italian journal of animal science

ASPA 23rd CONGRESS

Sorrento, June 11–14, 2019

Book of Abstracts

Guest Editors: Fulvia Bovera (Coordinator),
Marzia Albenzio, Mariangela Caroprese, Rosaria Marino,
Gianluca Neglia, Giovanni Piccolo, Angela Salzano.

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Italian Journal of Animal Science

The *Italian Journal of Animal Science* is an international peer-reviewed open access journal publishing original scientific papers, reviews and short communications.
The journal serves as essential reading for animal scientists, technicians and all those who research animal production.
The journal encourages submissions of international relevance on the following subjects:

- Animal derived food quality and safety
- Animal genetics and breeding
- Aquaculture, poultry, companion and wild game animals
- Livestock systems, management and environment
- Non-ruminant or ruminant nutrition and feeding
- Production physiology and functional biology of farmed, companions and wild game animals.
- Animal behavior
- Animal welfare
- In vitro studies that have an application to farmed livestock

Manuscripts must address topics based on research at molecular, cellular, organ, whole animal and production system levels. Manuscripts discussing milk or meat analysis and compositions must show a direct link to either livestock production system, product quality, animal feeding/nutrition, animal genetics or breeding. Manuscripts describing laboratory animal models will be considered where the study highlights a potential benefit to farmed livestock.

Submissions discussing epidemiology, parasitology, infective diseases, food-borne diseases do not fit with the aims and scope of the journal.

Meeting reviews, book reviews and conference supplements are also published, as well as news and guidelines from the Animal Science and Production Association (ASPA). We welcome submissions from ASPA members and non-members alike.

Article publishing charge

The standard article publishing charge (APC) for this journal is US$900 / €795 / £690. Depending on your location, these charges may be subject to local taxes.
For members of the Animal Science and Production Association, the publication fee is US$450 / €400 / £345. Depending on your location, these charges may be subject to local taxes. There are no submission charges for this journal.

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ASPA 23rd Congress

Sorrento, June 11–14, 2019

Guest Editors

Fulvia Bovera (Coordinator), Marzia Albenzio, Mariangela Caroprese,
Rosaria Marino, Gianluca Neglia, Giovanni Piccolo,
Angela Salzano

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23rd
Congress of the Animal Science and Production Association

23° Congresso dell'Associazione per la Scienza e le Produzioni Animali

Sorrento (Italy)
June 11th – 14th, 2019

Venue
Hilton Sorrento Palace
Congress Centre
Via S. Antonio 13, Sorrento

ASPA 23rd Congress

Sorrento, June 11-14, 2019

LEGEND
ES: *Environmental Sustainability*
LS: Livestock System
APHW: Animal Physiology, Health and Welfare
AP: Animal Products
ABG: Animal breeding and Genomics

Tuesday 11th June 2019

	Auditorium Ulisse	**Auditorium Tritone**	**Sala Stampa**	**Boardroom 6**
10.00 - 13.00		IJAS editorial board meeting		
11.00 - 15.00	**ARRIVAL - REGISTRATION**			
15.00-15.30		**INVITED LECTURE** *"Using metabolomics approaches in transition cow biology for basic and applied science"* **Helga Sauerwein**	**INVITED LECTURE** *"Leveraging on High-throughput Phenotyping Technologies to Optimize Livestock Genetic Improvement and Husbandry"* **Guilherme Rosa**	**INVITED LECTURE** *"Efficient aquafeed production and management in a circular future"* **Alessio Bonaldo**
15.30-16.45		**APHW** "Animal Physiology" (5)	**ABG** "Genetics of welfare, genetics of resilience" (5)	**LS** "Aquaculture" (5)
16.45-17.15	***Coffee Break***			
17.15-18.45		**LS** "GHG emissions in livestock" (5)	**APHW** "Tannins in animal nutrition" (6)	**ABG** "Poultry genetics" (5)
20.30	***CENA RUSTICA***			

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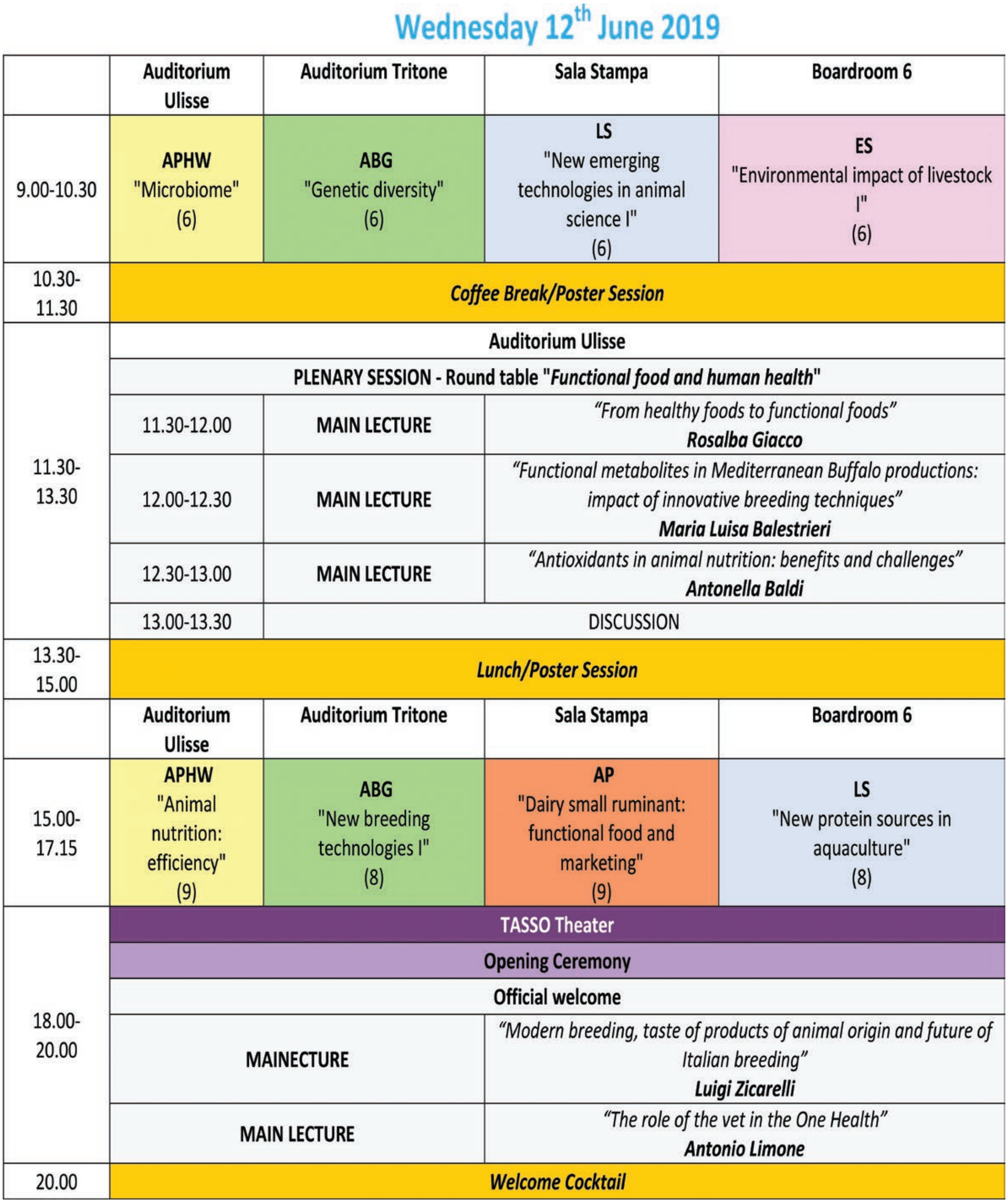

Wednesday 12th June 2019

	Auditorium Ulisse	Auditorium Tritone	Sala Stampa	Boardroom 6
9.00-10.30	**APHW** "Microbiome" (6)	**ABG** "Genetic diversity" (6)	**LS** "New emerging technologies in animal science I" (6)	**ES** "Environmental impact of livestock I" (6)
10.30-11.30	***Coffee Break/Poster Session***			
11.30-13.30	**Auditorium Ulisse**			
	PLENARY SESSION - Round table "*Functional food and human health*"			
	11.30-12.00	**MAIN LECTURE**	*"From healthy foods to functional foods"* ***Rosalba Giacco***	
	12.00-12.30	**MAIN LECTURE**	*"Functional metabolites in Mediterranean Buffalo productions: impact of innovative breeding techniques"* ***Maria Luisa Balestrieri***	
	12.30-13.00	**MAIN LECTURE**	*"Antioxidants in animal nutrition: benefits and challenges"* ***Antonella Baldi***	
	13.00-13.30	DISCUSSION		
13.30-15.00	***Lunch/Poster Session***			
	Auditorium Ulisse	**Auditorium Tritone**	**Sala Stampa**	**Boardroom 6**
15.00-17.15	**APHW** "Animal nutrition: efficiency" (9)	**ABG** "New breeding technologies I" (8)	**AP** "Dairy small ruminant: functional food and marketing" (9)	**LS** "New protein sources in aquaculture" (8)
18.00-20.00	**TASSO Theater**			
	Opening Ceremony			
	Official welcome			
	MAINECTURE		*"Modern breeding, taste of products of animal origin and future of Italian breeding"* ***Luigi Zicarelli***	
	MAIN LECTURE		*"The role of the vet in the One Health"* ***Antonio Limone***	
20.00	***Welcome Cocktail***			

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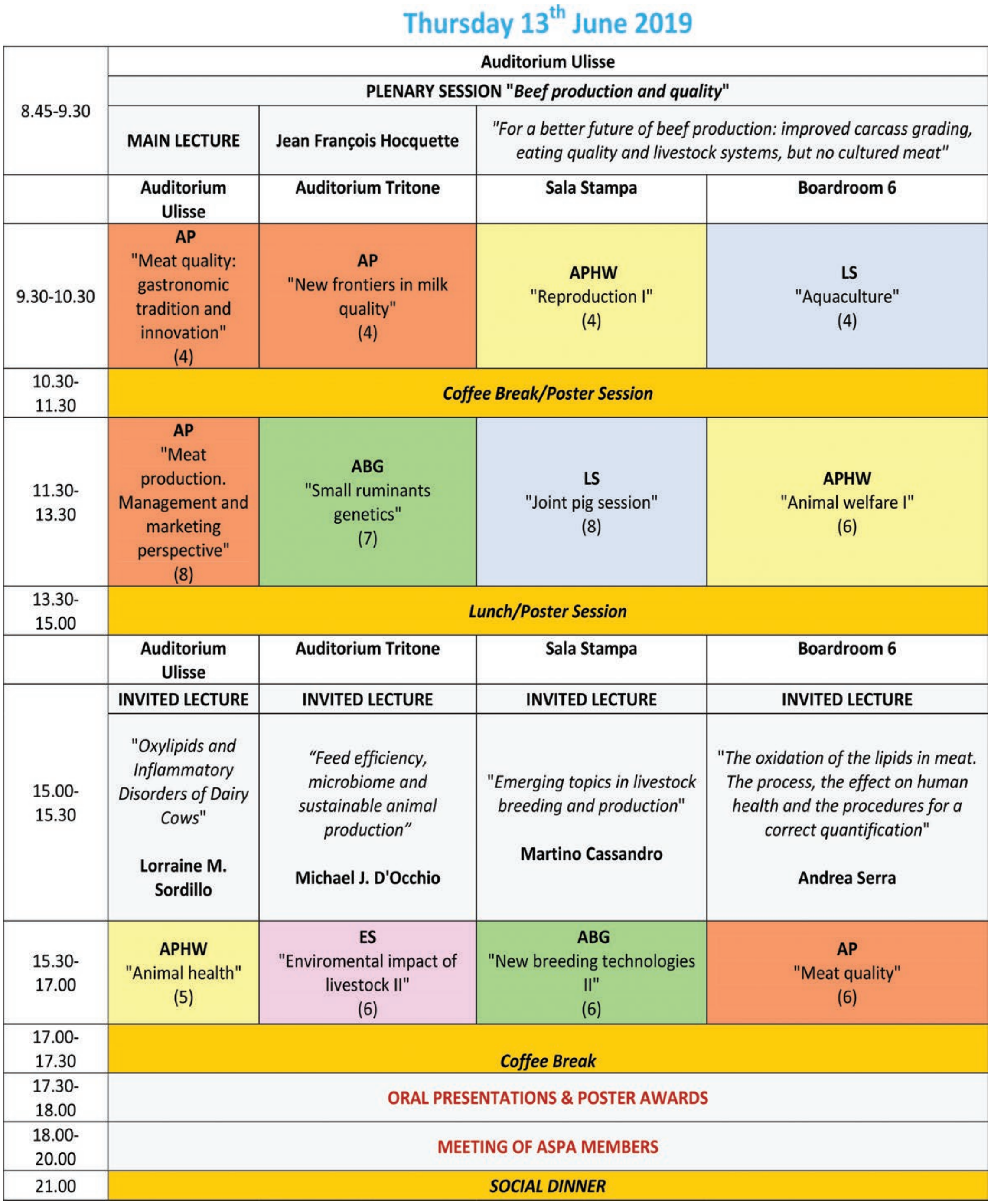

Thursday 13th June 2019

8.45-9.30	**Auditorium Ulisse**			
	PLENARY SESSION "*Beef production and quality*"			
	MAIN LECTURE	**Jean François Hocquette**	*"For a better future of beef production: improved carcass grading, eating quality and livestock systems, but no cultured meat"*	
	Auditorium Ulisse	**Auditorium Tritone**	**Sala Stampa**	**Boardroom 6**
9.30-10.30	**AP** "Meat quality: gastronomic tradition and innovation" (4)	**AP** "New frontiers in milk quality" (4)	**APHW** "Reproduction I" (4)	**LS** "Aquaculture" (4)
10.30-11.30	***Coffee Break/Poster Session***			
11.30-13.30	**AP** "Meat production. Management and marketing perspective" (8)	**ABG** "Small ruminants genetics" (7)	**LS** "Joint pig session" (8)	**APHW** "Animal welfare I" (6)
13.30-15.00	***Lunch/Poster Session***			
	Auditorium Ulisse	**Auditorium Tritone**	**Sala Stampa**	**Boardroom 6**
15.00-15.30	**INVITED LECTURE**	**INVITED LECTURE**	**INVITED LECTURE**	**INVITED LECTURE**
	"Oxylipids and Inflammatory Disorders of Dairy Cows" **Lorraine M. Sordillo**	*"Feed efficiency, microbiome and sustainable animal production"* **Michael J. D'Occhio**	*"Emerging topics in livestock breeding and production"* **Martino Cassandro**	*"The oxidation of the lipids in meat. The process, the effect on human health and the procedures for a correct quantification"* **Andrea Serra**
15.30-17.00	**APHW** "Animal health" (5)	**ES** "Enviromental impact of livestock II" (6)	**ABG** "New breeding technologies II" (6)	**AP** "Meat quality" (6)
17.00-17.30	***Coffee Break***			
17.30-18.00	**ORAL PRESENTATIONS & POSTER AWARDS**			
18.00-20.00	**MEETING OF ASPA MEMBERS**			
21.00	***SOCIAL DINNER***			

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Friday 14th June 2019

9.00-9.45	**Auditorium Ulisse**			
	Plenary Session "*Heat stress in cattle*"			
	MAIN LECTURE	**Geoffrey E. Dahl**	*"Late gestation heat stress in dairy cows: effects on dam and daughter "*	
	Auditorium Ulisse	**Auditorium Tritone**	**Sala Stampa**	**Boardroom 6**
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			"Nutritional modulation of canine and feline intestinal microbiota" **Giacomo Biagi**	*"Selection and conservation of Apis mellifera at the time of globalization"* **Giulio Pagnacco**
			APHW "Companion animals I" (2)	**ABG** "Genome editing" (2)
10.45-11.15	***Coffee Break***			
11.15-12.45	**INVITED LECTURE**	**INVITED LECTURE**	**LS** "Companion animals II" (5)	**LS** "New emerging technologies in animal science" (4)
	"Endocrine and physiological differences between bovine and buffalo females and their impact on assisted reproductive technologies" **Pietro S. Baruselli**	*"Animal Welfare Assessment: towards a simplification"* **Fabio Napolitano**		
	APHW "Reproduction II" (4)	**APHW** "Animal welfare II" (4)		

ASPA 23th Congress

Sorrento, June 11th–14th, 2019

Italian Journal of Animal Science
2019; volume XX

Agenda Invited Speakers

June, 11th

Auditorium Tritone, Animal Physiology, Health and Welfare – Animal physiology

Sala Stampa, Animal Breeding and Genomic – Genetics of welfare, genetics of resilience

Boardroom 6, Livestock Systems – Aquaculture

June, 12th

Auditorium Ulisse, PLENARY SESSION – Round Table "Functional food and human health"

Tasso Teather – Opening ceremony

June, 13th

Auditorium Ulisse, PLENARY SESSION – "Beef production and quality"

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Auditorium Ulisse, Animal Physiology, Health and Welfare – Animal health

Sala Stampa, Animal Breeding and Genomic – New breeding technologies II

Auditorium Tritone, Environmental Sustainability – Environmental impact of livestock II

Boardroom 6, Animal Products – Meat quality

June, 14th

Auditorium Ulisse, PLENARY SESSION – Animal Physiology, Health and Welfare, "Heat stress in cattle"

Sala Stampa, Animal Physiology, Health and Welfare – Companion animals I

Boardroom 6, Animal Breeding and Genomic – Genome editing

Auditorium Ulisse, Animal Physiology, Health and Welfare – Reproduction II

Auditorium Tritone, Animal Physiology, Health and Welfare – Animal welfare II

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Agenda for oral presentations

Tuesday, June 11th – Auditorium Tritone
Animal physiology, health and welfare – Animal physiology

Chairs: Mariangela Caroprese, Nicola Lacetera
15.30–15.45 Anna Benedet, Marco Franzoi, Mauro Penasa, Erika Pellattiero, Massimo De Marchi
Determination of blood metabolites in early lactation dairy cows using milk mid-infrared spectra O001
15.45–16.00 Erminio Trevisi, Matteo Mezzetti, Fiorenzo Piccioli Cappelli, Annarita Ferrari, Giorgia Lovotti, Andrea Minuti
Effect of the milk yield amount at the end of lactation on inflammation, oxidative stress, and metabolic changes during dry-off stage of dairy cow O002
16.00–16.15 Gabriela Avila Morales, Carlotta Catozzi, Valentina Zamarian, Cristina Lecchi, Fabrizio Ceciliani, Giulia Sala, Davide Pravettoni, Nicola Lacetera
Effect of heat stress on monocytes and lymphocytes in dairy cattle O003
16.15–16.30 Ludovica M.E. Mammi, Damiano Cavallini, Elisa Giaretta, Isa Fusaro, Attilio Mordenti, Andrea Formigoni
Study of inflammatory markers in dairy cows exposed to grain-rich diet O004
16.30–16.45 Alberto Stanislao Atzori, Antonello Ledda, Umberto Bernabucci, Loredana Basiricò, Mondina Francesca Lunesu, Antonello Cannas
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Tuesday, June 11th – Sala Stampa
Animal breeding and genomics – Genetics of welfare, genetics of resilience

Chairs: Alessio Cecchinato, Francesca Ciotola
15.30–15.45 Maurizio Marusi, Giulio Visentin, Claudio Cipolat-Gotet, Andrea Summer, Andrea Formigoni, Marco Nocetti, Martino Cassandro, Giorgio Burchiellaro, Giorgio Civati
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15.45–16.00 Alessio Cecchinato, Mehdi Momen, Gota Morota, Guilherme J.M. Rosa, Daniel Gianola, Sara Pegolo
Structural equation models for genome-wide association study (SEM-GWAS) of interrelationships among udder health traits in dairy cattle O007
16.00–16.15 Maria Martinez Castillero, Robert Tempelman, Gustavo de los Campos, Ana Ines Vazquez, Hugo Toledo Alvarado, Alessio Cecchinato
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16.15–16.30 Tania Bobbo, Ali Zidi, Mauro Penasa, Martino Cassandro
Cut-off values and genetic aspects of differential somatic cell count in dairy cows O009
16.30–16.45 Raffaella Finocchiaro, Jan-Thijs van Kaam, Guido Invernizzi, Giorgio Civati, Martino Cassandro, Giovanni Savoini
Setting up genetic evaluation for feed efficiency in Italian Holsteins O010

Tuesday, June 11th – Boardroom 6
Livestock systems – Aquaculture

Chairs: Pierpaolo Gatta, Alessandra Roncarati
15.30–15.45 Gloriana Cardinaletti, Donatella Volpatti, Chiara Bulfon, Paola Beraldo, Maria Messina, Emilio Tibaldi
Effects of nucleotide supplementation in diets high in soybean meal on growth, gut health and immune status of E. sea bass (D. labrax) O011

15.45–16.00 Roberto Cerri, Gloriana Cardinaletti, Fabrizio Capoccioni, Michela Contò, Giuliana Parisi, Natascia Biondi, Francesca Tulli, Emilio Tibaldi
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16.00–16.15 Genciana Terova, Simona Rimoldi, Elisabetta Gini, Federica Iannini, Marco Saroglia
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16.15–16.30 Lorenzo Morsiani, Luca Parma, Nicole Francesca Pelusio, Serena Busti, Matteo Zerlotin, Lorenzo Mariani, Pier Paolo Gatta, Alessio Bonaldo
Interaction between dietary composition and seasonal temperature changes in Gilthead sea bream (Sparus aurata, L. 1758): effects on growth, fat deposition, plasma biochemistry, enzymatic activity and gut bacterial community O014

16.30–16.45 Serena Busti, Luca Parma, Nicole Francesca Pelusio, Maurizio Mazzoni, Oliviero Mordenti, Francesco Dondi, Pierpaolo Gatta, Alessio Bonaldo
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Chairs: Giuseppe Luciano, Giovanni Savoini

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Tuesday, June 11th – Boardroom 6

Animal breeding and genomics – Poultry genetics

Chairs: Cesare Castellini, Martino Cassandro

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Chairs: Arianna Buccioni, Luciano Pinotti

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Wednesday, June 12th – Auditorium Tritone

Animal breeding and genomics – Genetic diversity

Chairs: Luca Fontanesi, Vincenzo Peretti

09.00–09.15 Sara Pegolo, Simone Savoia, Alberto Brugiapaglia, Liliana Di Stasio, Alfredo Pauciullo, Andrea Albera
Genome-wide association study and functional analysis of carcass and meat quality traits in double-muscled Piemontese cattle .O038

09.15–09.30 Roberta Ciampolini, Laurence Flori, Katayoun Moazami-Goudarzi, Salvatore Mastrangelo, Progetto BOVITA, Fabio Pilla, Martino Cassandro
Genomic Adaptation of Local Cattle Breeds in the Alpine Massif..........O039

09.30–09.45 Salvatore Mastrangelo, Elena Ciani, Slim Ben Jemaa, Gianluca Sottile, Baldassare Portolano, Fabio Pilla, BOVITA Consortium
The Italian cattle breeds in a worldwide context for a genome-wide diversity study..........O040

09.45–10.00 Cristina Sartori, Francesco Tiezzi, Nadia Guzzo, Roberto Mantovani
Genotype by environment interactions and response to selection for productive traits in a local cattle breed..........O041

10.00–10.15 Maria Chiara Fabbri, Christos Dadousis, Stefano Biffani, Riccardo Negrini, Riccardo Bozzi
Conservation status and rates of inbreeding of Italian autochthonous beef breeds..........O042

10.15–10.30 Elena Ciani, Salvatore Mastrangelo, Kaouadji Zoubeyda, Meghelli Imane, Andrea Criscione, Szilvia Kusza, Taki Karsli, Samir Bachir Souheil Gaouar, Rosaria Lucchini, Fabio Pilla, Bovita Consortium
Twenty shades of grey. Combined analysis of genome-wide SNP data in Steppe cattle and in Mediterranean grey cattle shed new light on the molecular bases of coat colour..........O043

Wednesday, June 12th – Sala Stampa

Livestock systems – New emerging technologies in animal science I

Chairs: Paolo Bani, Massimo De Marchi

09.00–09.15 Maddalena Zucali, Stefania Celozzi, Anna Sandrucci, Alberto Tamburini, Francesco Maria Tangorra, Lucio Zanini
How to measure milking efficiency in dairy cattle farms? O044

09.15–09.30 Monica Berlese, Stefano Bovolenta, Mirco Corazzin, Damiano Gianelle, Maurizio Ramanzin, Enrico Sturaro
Precision farming approach to assess grazing dairy cows adaptation to mountain pasture O045

09.30–09.45 Rosanna Marino, Francesca Petrera, Marisanna Speroni, Teresa Rutigliano, Andrea Galli, Fabio Abeni
Unravelling the relationship between milk quality and rumination by a PLF technology O046

09.45–10.00 Maura Sannino, Salvatore Faugno, Francesco Serrapica, Maria Luisa Varricchio, Fernando Grasso, Antonio Di Francia, Felicia Masucci
Automatic milking system in Mediterranean buffaloes: experimental results and considerations O047

10.00–10.15 Francesco Maria Tangorra, Veronica Redaelli, Fabio Luzi, Mauro Zaninelli
First evaluation of the infrared thermography as a tool for the detection of mechanical stress on cow udder teat due by milking O048

10.15–10.30 Marco Acciaro, Valeria Giovanetti, Corrado Dimauro, Giampaolo Epifani, Carla Manca, Salvatore Contini, Andrea Cabiddu, Mauro Decandia, Giovanni Molle
Milk production of Sarda suckler cows with different calving period O049

Wednesday, June 12th – Boardroom

Environmental Sustainability– Environmental impact of livestock I

Chairs: Fulvia Bovera, Claudio Cavani

09.00–09.15 Stefania Colombini, Sara Savoldelli, Daniela Lupi, Gianluca Galassi
Growth of Hermetia illucens on different food by-products: conversion efficiency and nutritive value. Part I O050

09.15–09.30 Luciana Bava, Costanza Jucker, Stefania Celozzi, Maddalena Zucali
Growth of Hermetia illucens on different food by-products: environmental sustainability. Part II O051

09.30–09.45 David Meo Zilio, Monica Guarino Amato
Preliminary results of a systematic review on chemical and nutritional characteristics of Musca domestica and Hermetia illucens as feed O052

09.45–10.00 Marco Meneguz, Antonio Franco, Andrea Dama, Christian Caimi, Vanda Malfatto, Patrizia Falabella, Laura Gasco
Impact of different protein levels and substrates fermentation on the development of the first stages of black soldier fly (Hermetia illucens) larvae O053

10.00–10.15 Marco Cullere, Yazavinder Singh, Giulia Tasoniero, Joris Michiels, Antonella Dalle Zotte
Black soldier fly (Hermetia illucens) as dietary source for laying quails: live performance and physical traits of eggs O054

10.15–10.30 Doriana E.A. Tedesco, Cecilia Conti, Jacopo Bacenetti
Environmental sustainability assessment: from fruit and vegetable waste to earthworm as feed sources O055

Wednesday, June 12th – Sala Stampa

Animal Products – Dairy small ruminant: functional food and marketing

Chairs: Marzia Albenzio, Antonio Pirisi

15.00–15.15 Adriana Campione, Bernardo Valenti, Luciano Morbidini, Marcella Avondo, Giuseppe Luciano, Martyna Wilk, Pawel Migdal, Barbara Król, Camilla Pomente, Mariano Pauselli
Dietary hazelnut skins: effects on milk quality of dairy ewes O056

15.15–15.30 Salvatore Claps, Maria Antonietta Di Napoli, Lucia Sepe, Giovanni Annicchiarico
Effect of destoned olive cake on the milk fatty acid profile O057

15.30–15.45 Nadia Musco, Serena Calabrò, Olga Lai, Maria Ferrara, Cristina Roncoroni, Federico Infascelli
Influence of breeding system on ewe milk quality O058

15.45–16.00 Annamaria Perna, Amalia Simonetti, Giulia Grassi, Emilio Gambacorta
Efficiency in sheep milk production: comparison between extensive and intensive management systems O059

16.00–16.15 Francesco Lai, Fabio Correddu, Antonello Ledda, Mondina Francesca Lunesu, Roberto Rubattu, Antonio Mazza, Antonio Fenu, Alberto Stanislao Atzori, Antonello Cannas
Effect of forage quality on milk production of Sarda dairy ewes O060

16.15–16.30 Annalaura Lopez, Mauro Vasconi, Vittorio Maria Moretti, Federica Bellagamba
Influence of different livestock production systems in caprine milk and cheese odd and branched chain fatty acids (OBCFA) composition O061

16.30–16.45 Matteo Daghio, Francesco Pini, Anna Espinoza-Tofalos, Luciana Giovannetti, Arianna Buccioni, Andrea Franzetti, Carlo Viti
Is pink discoloration defect in Pecorino cheese due to Thermus thermophilus presence? O062

16.45–17.00 Lucia Sepe, Rosanna Salvia, Francesco Paladino, Salvatore Claps, Giovanni Quaranta
The answer of the consumers to a cheese with natural vegetable rennet O063

17.00–17.15 Arzu Gökdai, Engin Sakarya, Barbara Contiero, Flaviana Gottardo
Socio-Economic Analysis and Current Problems of Saanen Goat Farms in Turkey: A case of Çanakkale Province O064

Wednesday, June 12th – Auditorium Tritone

Animal breeding and genomics – New breeding technologies I

Chairs: Niccolò Pietro Paolo Macciotta, Gianfranco Cosenza

15.00–15.15 Martina Zappaterra, Silvia Gioiosa, Giovanni Chillemi, Tiziana Castrignanò, Paolo Zambonelli, Roberta Davoli
Comparative analysis of Semimembranosus muscle transcriptome in Italian Large White pigs with high and low intramuscular fat content O065

15.15–15.30 Alberto Cesarani, Giustino Gaspa, Lorenzo Degano, Daniele Vicario, Nicolò Pietro Paolo Macciotta
Runs of homozygosity of Simmental bulls raised in five European countries O066

15.30–15.45 Enrico Mancin, Bolivar Samuel Sosa-Madrid, Agustin Blasco, Roberto Mantovani, Noelia Ibañez-Escriche
Efficiency of imputation for genomic selection in rabbits O067

15.45–16.00 Elisa Cotozzolo, Simona Mattioli, Alice Cartoni Mancinelli, Michele Schiavitto, Paola Principe, Alessandro Dal Bosco, Cesare Castellini
Assessment of a fertility and a maternal behavior index: effect of kindling order in three genetic lines of rabbits does O068

16.00–16.15 Mario Graziano Usai, Sara Casu, Bachisio Ziccheddu, Tiziana Sechi, Sabrina Miari, Patrizia Carta, Sotero Salaris, Antonello Carta
Using identity-by-descent probability to impute whole genome sequence variants in a nucleus flock O069

16.15–16.30 Sara Casu, Paolo Ajmone Marsan, Mario Barbato, Patrizia Carta, Marcello Del Corvo, Sotero Salaris, Tiziana Sechi, Mario Graziano Usai, Sabrina Miari, Antonello Carta
Biodivsel: Functional annotation of target regions based on whole genome resequencing of Sarda sheep O070

16.30–16.45 Barbara Auzino, Gianfranco Cosenza, Federica Salari, Mina Martini, Roberta Ciampolini
Gene structure analysis of donkey oxytocin-neurophysin I (OXT) gene and genetic variability detection O071

16.45–17.00 Samanta Mecocci, Stefano Capomaccio, Giovanni Chillemi, Andrea Giontella, Raffaele Cherchi, Maurizio Silvestrelli, Alessio Valentini, Katia Cappelli
Endurance exercise shifts mature mRNA towards introns: does stress enhance genome plasticity? O072

Wednesday, June 12th – Boardroom 6

Livestock systems – New protein sources in aquaculture

Chairs: Laura Gasco, Giovanni Piccolo

15.00–15.15 Federico Nassivera, Francesca Tulli, Emilio Tibaldi
Millennials' attitudes towards insects as feed for sustainable fish O073

15.15–15.30 Enrico Daniso, Maria Messina, Francesca Tulli
Molecular based identification of insect ingredients in animal feed O074

15.30–15.45 Ilaria Biasato, Elena Colombino, Giulia Chemello, Christian Caimi, Marco Meneguz, Sihem Dabbou, Maria Teresa Capucchio, Achille Schiavone, Francesco Gai, Laura Gasco
Dietary Tenebrio molitor larva meal inclusion in rainbow trout diets: preliminary results about animal and gut health implications O075

15.45–16.00 Christian Caimi, Ilaria Biasato, Giulia Chemello, Achille Schiavone, Marco Meneguz, Sihem Dabbou, Manuela Renna, Francesco Gai, Laura Gasco
High protein Tenebrio molitor larvae meal in rainbow trout diets O076

16.00–16.15 Simona Rimoldi, Laura Gasco, Chiara Ceccotti, Micaela Antonini, Genciana Terova
Black soldier fly (Hermetia illucens) larvae meal as a dietary protein source for rainbow trout (Oncorhynchus mykiss): effects on allochthonous and autochthonous gut microbial communities O077

16.15–16.30 Leonardo Bruni, Giulia Secci, Matteo Zarantoniello, Basilio Randazzo, Ike Olivotto, Fabio Mina, Francesca Tulli, Giuliana Parisi
Lipid metabolism and fillet quality of rainbow trout fed diets including Hermetia illucens full-fat larvae O078

16.30–16.45 Giulia Maricchiolo, Biagina Chiofalo, Letteria Caccamo, Giovanni Marco Cusimano, Lucrezia Genovese, Laura Gasco, Francesco Gai
Effects of dietary inclusion of defatted black soldier fly (Hermetia illucens) meal on gut health in gilthead seabream (Sparus aurata) O079

16.45–17.00 Giulia Secci, Leonardo Bruni, Monica Borgogno, Ikram Belghit, Erik-Jan Lock, Giuliana Parisi
Fillet qualitative characteristics of Atlantic salmon fed diets including Hermetia illucens larvae O080

Wednesday, June 12th – Auditorium Ulisse

Animal physiology, health and welfare – Animal nutrition: efficiency

Chairs: Federico Infascelli, Salvatore Claps

15.00–15.15 Vera Perricone, Alessandro Agazzi, Annamaria Costa, Aldo Calcante, Giovanni Savoini, Massimo Lazzari, Federica Cheli, Antonio Crotti, Diego De Nisi, Francesco Maria Tangorra
Sources of variation in TMR delivered to dairy cows: a field study O081

15.15–15.30 Fabio Omodei Zorini, Elena Mariani, Thi Xuan Nguyen, Guido Invernizzi
Evaluation of Feed efficiency in Italian Holstein Friesian heifers and lactating cows O082

15.30–15.45 Serena Calabrò, Ivan B. Koura, Raffaella Tudisco, Nadia Musco, Alessandro Vastolo, Monica I. Cutrignelli
Characteristics of forages collected in different areas of Benin O083

15.45–16.00 Gianni Matteo Crovetto, Giulia Gislon, Stefania Colombini, Andrea Rota Graziosi, Maria Chiaravalli, Luca Rapetti
Milk production and digestibility in cows fed diets with different forage basis O084

16.00–16.15 Massimo Todaro, Antonino Di Grigoli, Maria Luisa Scatassa, Cinzia Cardamone, Concetta Maria Messina, Francesca Mazza, Adriana Bonanno
Prickly pear pulp in the feeding of livestock ruminants: preliminary investigation O085

16.15–16.30 Luigi Gallo, Luca Carraro, Sudeb Saha
Body weight and measures of purebred Holstein and crossbred cows from Viking Red, Montbéliarde and Holstein sires O086

16.30–16.45 Sudeb Saha, Stefano Schiavon, Luigi Gallo
Production efficiency of purebred Holstein and crossbred cows from Viking Red, Montbéliarde and Holstein sires O087

16.45–17.00 Nicola Bettiga, Veronica Redaelli, Fabio Luzi, Giovanni Savoini
Quality assessment of corn silage by infrared thermography O088

17.00–17.15 Manuela Renna, Anne Ferlay, Carola Lussiana, Benoît Graulet, Ueli Wyss, Luca Maria Battaglini, Mauro Coppa
Factors affecting the fatty acid profile of permanent grasslands O089

Thursday, June 13th – Auditorium Ulisse

Animal Products – Meat quality: gastronomic tradition and innovation

Chiars: Gianluca Neglia, Rosaria Marino

09.30–09.45 Valeria Petrecca, Siria Tavaniello, Rossella Mucci, Daniela Prioriello, Giancarlo Salvatori, Ondřej Bučko, Giuseppe Maiorano
Quality and safety of meat from wild boar hunted in Molise region O090

09.45–10.00 Aristide Maggiolino, Rosaria Marino, Alessandra Tateo, Pasquale Centoducati, Jose Manuel Lorenzo, Pasquale De Palo
Effect of aging time on the volatile compounds profile of donkey meat O091

10.00–10.15 Hanane Smili, Samira Becila, Radjeb Ayad, Fatima Zohra Chakou, Marzia Albenzio, Mariangela Caroprese, Antonella Santillo, Agostino Sevi, Baaissa Babelhadj, Abdelkader Adamou, Abdelghani Boudjellal, Rosaria Marino
Dromedary meat proprieties: case of algerien sahraoui dromedary O092

10.15–10.30 Qianlin Ni, Matteo Bergamaschi, Franco Biasioli, Iuliia Khomenko, Giovanni Bittante
Effect of cooking method on the volatile organic compound profile of meat of different species identified by PTR-ToF-MS O093

Thursday, June 13th – Auditorium Tritone

Animal Products – New frontiers in milk quality

Chairs: Angela Gabriella d'Alessandro, Ada Bragheri

09.30–09.45 Angela Salzano, Jacopo Troisi, Anna Balestrieri, Giovanni Scala, Francesco Vinale, Giuseppe Campanile
Traceability of buffalo milk and mozzarella cheese through metabolomic profile O094

09.45–10.00 Nunzia D'Onofrio, Alessandra Monaco, Angelo Capasso, Rosario Casale, Gianluca Neglia, Giuseppe Campanile, Maria Luisa Balestrieri
Antioxidant and Anti-Inflammatory Activities of Buffalo Milk Bioactive Metabolites O095

10.00–10.15 Beatrice Tozzi, Domenico Gatta, Marco Mariotti, Lucia Casini, Sara Minieri
Study of nutraceutical substances (Rutin) in sheep's milk fed with buckwheat hay O096

10.15–10.30 Iolanda Altomonte, Elio Novembre, Giovanni Brajon, Fina Belli, Franco Corrias, Rosario Licitra, Federica Salari, Lucrezia Sarti, Mina Martini
Use of donkey milk in infant feeding O097

Thursday, June 13th – Sala Stampa
Animal physiology, health and welfare – Reproduction I

Chairs: Rossella Di Palo, Nicolaia Iaffaldano

09.30–09.45 Fabio Mosca, Ahmad Abdel Sayed, Maria Grazia Mangiagalli, Silvia Cerolini, Luisa Zaniboni
Quality of chicken semen cryopreserved with different N-methylacetamide concentrations combined with trehalose O098

09.45–10.00 Marek Bednarczyk, Maria Wiechetek, Ewa Lukaszewicz, Artur Kowalczyk
Influence of germ line chimerism on chicken reproductive traits O099

10.00–10.15 Michele Di Iorio, Giusy Rusco, Angelo Manchisi, Silvia Cerolini, Nicolaia Iaffaldano
How can the honey improve the post-thaw quality of turkey spermatozoa? O100

10.15–10.30 Giusy Rusco, Michele Di Iorio, Stefano Esposito, Pier Paolo Gibertoni, Bruno Testa, Silvia Cerolini, Alessandra Roncarati, Maria Silvia D'Andrea, Nicolaia Iaffaldano
Thawing rate effects on the cryosurvival of Mediterranean brown trout spermatozoa O101

Thursday, June 13th – Boardroom 6
Livestock systems – Aquaculture

Chairs: Giuliana Parisi, Francesca Tulli

09.30–09.45 Nicole Francesca Pelusio, Luca Parma, Serena Busti, Stefano Porcelli, Enric Gisbert, Maria Angeles Esteban-Abad, Federica D'Amico, Matteo Soverini, Oliviero Mordenti, Antonio Casalini, Pier Paolo Gatta, Alessio Bonaldo
Effects of rearing density on growth, welfare indicators and digestive conditions of Gilthead sea bream (Sparus aurata, L. 1758) fed different fish meal and fish oil dietary levels O102

09.45–10.00 Francesco Bordignon, Gerolamo Xiccato, Cristina Zomeño, Anton Pascual, Marco Birolo, Emanuele Rossetti, Angela Trocino
Pre-fattening of Manila clams in a Po Delta lagoon: effect of stocking density in suspended lanterns O103

10.00–10.15 Benedetto Sicuro, Beatriz Castelar
Integrated freshwater aquaculture: potentialities and limitations of freshwater bivalves utilization O104

10.15–10.30 Matteo Dell'Anno, Stefania Sotira, Monika Hejna, Valentina Caprarulo, Serena Reggi, Luciana Rossi
In vitro evaluation of macroalgae (Ascophyllum nodosum and Lithothamnium calcareum) and microalge (Schizochytrium spp.) for animal nutrition O105

Thursday, June 13th – Auditorium Ulisse
Animal Products – Meat production. Management and marketing perspective

Chairs: Giuseppe Maiorano, Pasquale Centoducati

11.30–11.45 Lorenzo Bussotti, Gabriele Ferrini, Federico Fava, Giulia Berni, Matilde Ciani, Christos Dadousis
In vitro cultured meat: an Italian consumer survey O106

11.45–12.00 Ruggero Menci, Mariano Pauselli, Adriana Campione, Camilla Pomente, Luciano Morbidini
Consumer test with salami from "Umbria" medium-heavy pork, fed traditional or experimental diet with hazelnut perisperm O107

12.00–12.15 Simona Mattioli, Alessandro Dal Bosco, Matics Zsolt, Gerencsér Zsolt, Marco Cullere, Szendrő Zsolt, Cesare Castellini, Antonella Dalle Zotte
Effect of dietary liquorice extract on the oxidative status of rabbit burgers O108

12.15–12.30 Simone Mancini, Roberta Nuvoloni, Francesca Pedonese, Simona Mattioli, Alessandro Dal Bosco, Gisella Paci
Effects of garlic powder and salt additions in rabbit meat burgers O109

12.30–12.45 Marisa Palazzo, Francesco Vizzarri, Angela Gabriella D'Alessandro, Giovanni Martemucci, Raffaella Rossi, Sabrina Ratti, Carlo Corino, Donato Casamassima
Dietary effect of dried bay (Laurus nobilis) meal on rabbit meat quality O110

Thursday, June 13th – Auditorium Tritone

Animal breeding and genomics – Small ruminants genetics

Chairs: Fabio Pilla, Elena Ciani

Thursday, June 13th – Boardroom 6

Animal physiology, health and welfare – Animal welfare I

Chairs: Fabio Napolitano, Silvana Mattiello

Thursday, June 13th – Sala Stampa

Joint pig session

Chairs: Giovanna Martelli, Emilio Gambacorta

Thursday, June 13th – Auditorium Ulisse

Animal physiology, health and welfare – Animal health

Chairs: Antonella Santillo, Giuseppe Pulina

Thursday, June 13th – Sala Stampa

Animal breeding and genomics – New breeding technologies II

Chairs: Emiliano Lasagna, Mauro Penasa

Thursday, June 13th – Auditorium Tritone
Environmental Sustainability – Environmental impact of livestock II

Chairs: Matteo Crovetto, Fabio Abeni

15.30–15.45 Enrico Sturaro, Marco Berton, Stefano Bovolenta, Maurizio Ramanzin, Chiara Spiegarelli, Anna Zuliani
Added value of dairy farming in mountain areas: an Ecosystem Services approach O146

15.45–16.00 Mondina Francesca Lunesu, Alberto Stanislao Atzori, Fabio Correddu, Francesco Lai, Roberto Rubattu, Gesumino Spanu, Antonello Cannas
Carbon footprint of dairy sheep farms located in different forage systems of Sardinia O147

16.00–16.15 Daniela Lovarelli, Giuliana D’Imporzano, Alberto Tamburini
Environmental sustainability of dairy farms producing milk for Grana Padano and Parmigiano Reggiano cheese production O148

16.15–16.30 Federico Froldi, Sara Corrado, Francesco Masoero, Maurizio Moschini
Parameters affecting the carbon footprint of milk at the farm gate O149

16.30–16.45 Giulia Gislon, Luciana Bava, Giorgio Borreani, Aldo Dal Prà, Maria Teresa Pacchioli, Ernesto Tabacco, Anna Sandrucci
Greenhouse gas emissions associated to cattle milk production in northern Italy O150

16.45–17.00 Severino Segato, Giorgia Riuzzi, Vittoria Bisutti, Lorenzo Serva, Flaviana Gottardo
Effect of the main dietary roughage source on the N-nutrient use efficiency (N-NUE) in dairy systems O151

Thursday, June 13th – Boardroom 6
Animal Products – Meat quality

Chairs: Marcello Mele, Girolamo Xiccato

15.30–15.45 Francesca Ciucci, Roberta Nuvoloni, Leonor Corrales Retana, Laura Casarosa, Barbara Turchi, Alice Cappucci, Domenico Cerri, Marcello Mele
Effect of rearing system and packaging method on lipid oxidation and shelf life of beef from Aubrac steers during storage O152

15.45–16.00 Miriam Iacurto, Michela Contò, Francesca Ciucci, Laura Casarosa
Cattle rearing at pasture or in feedlot farming: quality of meat O153

16.00–16.15 Silvia Parrini, Valentina Becciolini, Lapo Nannucci, Antonio Bonelli, Anna Acciaioli
Effect of different farming systems on meat quality of Massese lambs O154

16.15–16.30 Leonor Corrales Retana, Eleonora Bulleri, Alice Cappucci, Giuseppe Conte, Andrea Serra
Effect of Polyphenols supplementation on growth performance and meat shelf life of broiler chicken O155

16.30–16.45 Massimo Trabalza-Marinucci, Roberta Galarini, Raffaella Branciari, Patrizia Casagrande-Proietti, Laura Vieceli, Flavia Lucignani, Dino Miraglia, Maria Pia Franciosini, Gabriele Acuti
Effects of two different olive mill wastewaters on growth performance, quality of meat and Campylobacter loads in broiler chickens O156

16.45–17.00 Siria Tavaniello, Anna Slawinska, Micol Bertocchi, Rossella Mucci, Valeria Petrecca, Federico Sirri, Giuseppe Maiorano
In ovo delivery of GOS in fast-growing broiler chickens exposed to heat stress: implications for meat quality traits O157

Friday, June 14th – Auditorium Ulisse
Animal physiology, health and welfare – Climate

Chairs: Pasquale De Palo, Umberto Bernabucci

09.45–10.00 Luisa Magrin, Marta Brscic, Isabella Lora, Giulio Cozzi
Effect of different flooring systems on claw condition of finishing beef cattle during warm and cold seasons O158

10.00–10.15 Paolo Ajmone Marsan, Marcello Del Corvo, Emanuele Capra, Barbara Lazzari, Ludmilla Zavarez, Marco Milanesi, Yuri Tani Utsunomiya, Adam Taiti, Guillerme De Paula Noguiera, Josè Fernando Garcia
Methylomes of indicine and taurine cattle under heat challenge O159

10.15–10.30 Maria Usala, Maria Carabaño, Manuel Ramon, Cristina Meneses, Nicolò Macciotta, Clara Diaz
Individual thermal response of Avileña Negra-Ibérical calves at feedlot O160

10.30–10.45 Pasquale De Palo, Aristide Maggiolino, Umberto Bernabucci, Attilio Rossoni, Enrico Santus, Andrea Vitali
Heat stress in Italian Brown Swiss O161

Friday, June 14th – Sala Stampa
Animal physiology, health and welfare – Companion animals I

Chairs: Bruno Stefanon, Biagina Chiofalo

10.15–10.30 Monica I. Cutrignelli, Nadia Musco, Alessandro Vastolo, Marcia O.S. Gomes, Aulus C. Carciofi
Digestibility of extruded diets for cats: in vitro vs. in vivo O162

10.30–10.45 Elisa Scarsella, Misa Sandri, Michela Cintio, Sandy Sgorlon, Giuseppe Conte, Bruno Stefanon
DogBiome: the gut microbiome project for dog O163

Friday, June 14th – Boardroom 6
Animal breeding and genomics – Genome editing

Chairs: Bruno Ronchi, Antonino Nizza

10.15–10.30 Giulietta Minozzi, Gustavo Gandini, Barbara Lazzari, Elena Facchini, Cecilia Costa, Andrea Talenti, Giulio Pagnacco
Unveiling the Biodiversity of the Italian Honeybees by Next Generation Sequencing O164

10.30–10.45 Elena Ciani, Pamela Burger, Samantha Brooks, Faisal Almathen, the International Camel Consortium for Genetic Improvement and Conservation
The Illumina® Greater Good Initiative. A further step toward inclusion of camels in the "agrigenomic revolution" O165

Friday, June 14th – Auditorium Ulisse
Animal physiology, health and welfare – Reproduction II

Chairs: Bianca Gasparrini, Riccardo Negrini

11.45–12.00 Nageshvar Patel, Matteo Bergamaschi, Giovanni Bittante
Combined use of dairy sexed semen and double-muscled breed semen for beef crossbred calves production in dairy herds O166

12.00–12.15 Stefano Biffani, Roberta Cimmino, Giuseppe Campanile, Gianluca Neglia, Rossella Di Palo, Riccardo Negrini, Dario Rossi, Giacomo Bertolini
Developing a new selection index for the Italian Mediterranean Buffalo (Bubalus bubalis) O167

12.15–12.30 Angela Costa, Massimo De Marchi, Giuseppe Campanile, Riccardo Negrini, Gianluca Neglia
Effect of milk somatic cell level on lifetime milk related performances in Italian water buffaloes O168

12.30–12.45 Riccardo Negrini, Mario Barbato, Licia Colli, Yuri T. Utsunomiya, Jesus R.V. Herrera, Libertado Cruz, Pietro S. Baruselli, Maria E.J. Amaral, Marcela G. Drummond, José F. Garcia, John L. Williams, The International Water Buffalo Consortium, Paolo Ajmone-Marsan
Adaptive introgression of river-buffalo derived regions in the genome of Brazilian Carabao swamp buffalo O169

Friday, June 14th – Auditorium Tritone
Animal physiology, health and welfare – Animal welfare II

Chairs: Giuseppe de Rosa, Elisabetta Canali

11.45–12.00 Mirco Corazzin, Monica Berlese, Anna Zuliani, Edi Piasentier, Domenico Vecchio
Animal welfare and environmental sustainability of dairy buffalo farms in Northern Italy O170

12.00–12.15 Francesco Serrapica, Fabio Napolitano, Ada Braghieri, Giovanni Migliori, Felicia Masucci, Antonio Di Francia, Fernando Grasso, Giuseppe De Rosa
Risk factors associated with the expression of cross-sucking and intersucking in buffaloes O171

12.15–12.30 Monica Battini, Anna Agostini, Silvana Mattiello
How can we understand the emotional state of cattle for welfare evaluation purpose? O172

12.30–12.45 Alice Cartoni Mancinelli, Andrea Aliberti, Simona Mattioli, Elisa Cotozzolo, Monica Guarino Amato, Alessandro Dal Bosco, Cesare Castellini
Behaviour assessment of different poultry genotypes organically reared with computerized video recording system O173

Friday, June 14th – Sala Stampa
Livestock systems – Companion animals II

Chairs: Bruno Stefanon, Biagina Chiofalo

11.15–11.30 Bruno Stefanon, Michela Cintio, Elisa Scarsella, Lucilla Iacumin
Interplay between salivary biomarkers and gut microbiome in dogs O174

11.30–11.45 Natalia Russo, Elena Pagani, Achille Schiavone, Laura Gasco, Joana Nevy, Silvia Martínez, Fuensanta Hernandez, Josefa Madrid, Liviana Prola
In vivo and in vitro digestibility of extruded dog foods with Hermetia illucens O175

11.45–12.00 Alessia Candellone, Liviana Prola, Pier Giorgio Peiretti, Sonia Tassone, Erica Longato, Daniele Pattono, Natalia Russo, Giorgia Meineri
In vivo and in vitro digestibility, palatability and nutritive quality of extruded dog food based on mechanically separated chicken meat or meat by-product O176

12.00–12.15 Emiliano Lasagna, Simone Ceccobelli, Amparo Martinez Martinez, Hovirag Lancioni, Vincenzo Landi, Mario Cosenza, Anthony Gruppetta, Francesca Maria Sarti, Vincenzo Chiofalo, Carlos San José, Luis Monteagudo Ibanez, Luigi Liotta, George Attard
Genetic variability assessment in different Mediterranean canine breeds using microsatellite markers O177

12.15–12.30 Paola Crepaldi, Stefano Frattini, Alessio Negro, Matteo Cortellari, Andrea Talenti
Metigree: a genomic tool to enhance the well-being of Italian mongrel dogs O178

Friday, June 14th – Boardroom 6
Livestock systems – New emerging technologies in animal science

Chairs: Annamaria Perna, Antonio Di Francia

11.15–11.30 David J. Wilson, John Middleton, Pamela Adkins, Gregory M. Goodell
Conventional culture, MALDI-TOF and 16S rRNA compared for test agreement in diagnosis of bacteria in bovine milk samples O179

11.30–11.45 Carlotta Giromini, Raffaella Rebucci, Gabriella Tedeschi, Tamil Selvi Sundaram, Federica Cheli, Antonella Baldi
Angiotensin Converting Enzyme-1 inhibitory activity of milk proteins evaluated after in vitro digestion and peptidomic analysis O180

11.45–12.00 Corrado Dimauro, Salvatore Pier Giacomo Rassu, Rossella Cossu, Maria Gabriella Serra, Carla Manca, Andrea Cabiddu, Marco Acciaro, Giovanni Molle, Mauro Mameli, Mauro Decandia, Valeria Giovanetti
The ability of the partial least square regression to predict the number of bites in Sarda dairy sheep by using acceleration data O181

12.00–12.15 Nadia Piscopo, Andrea Amici, Maria Pia Pasolini, Giovanni Della Valle, Pierpaolo Coluccia, Orlando Paciello, Luigi Esposito
Evaluation of the foal Esperia ponies' mortality attributable to predation by roaming dogs or wolves O182

ASPA 23th Congress

Sorrento, June 11th–14th, 2019

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Agenda for poster presentations

ANIMAL BREEDING AND GENOMICS

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE

ANIMAL PRODUCTS

ENVIRONMENTAL SUSTAINABILITY

LIVESTOCK SYSTEMS

Foreword

The 23rd Congress of the Animal Science and Production Association (ASPA) is hosted in Sorrento, a wonderful location near Naples, where sea and mountain, deep valleys and citrus groves plateaux draw a unique, fantastic landscape. The Congress is organized by the Department of Veterinary Medicine and Animal Production of the University of Naples "Federico II" in cooperation with other Italian scientific Institutions, such as the Universities of Foggia, Bari "Aldo Moro" and Basilicata; the Council for Agricultural Research and Economics – CREA PCM (Rome) and the "Istituto Zooprofilattico Sperimentale del Mezzogiorno" (IZSM).

This edition of the ASPA Congress has received a total of 338 scientific contributions: 182 oral presentations and 156 posters have been selected. The Congress sessions with their relative number of papers are listed as follows: Animal breeding and genomics (90); Animal Physiology, Health and Welfare (101); Animal Products (68); Livestock Systems (55); Environmental Sustainability (24).

The scientific program is enriched by 18 main lectures, covering all the main topics.

Many thanks for the extraordinary job done to the President of the Organizing Committee, Prof. Gianluca Neglia, and to all components: Prof. Giuseppe Campanile, Prof. Pasquale Centoducati, Prof. Salvatore Claps, Prof. Angela Gabriella D'Alessandro, Prof. Giosuè De Simone, Prof. Antonio Di Francia, Prof. Emilio Gambacorta, Prof. Federico Infascelli, Dr. Antonio Limone, Prof. Vincenzo Peretti, Prof. Domenico Raimondo, Prof. Angela Salzano, Prof. Agostino Sevi, Prof. Alessandra Tateo. I also wish to thank all the members of the Scientific Committee, Prof. Marzia Albenzio (President), Prof. Giovanni Annicchiarico, Prof. Fulvia Bovera, Prof. Serena Calabrò, Prof. Mariangela Caroprese, Prof. Francesca Ciotola, Prof. Monica Isabella Cutrignelli, Prof. Pasquale De Palo, Prof. Giuseppe De Rosa, Prof. Luigi Esposito, Prof. Bianca Gasparrini, Prof. Emiliano Lasagna, Prof. Rosaria Marino, Prof. Felicia Masucci, Prof. Marcello Mele, Prof. Fabio Napolitano, Prof. Annamaria Perna, Prof. Giovanni Piccolo, Prof. Antonella Santillo, Prof. Raffaella Tudisco, and all who have contributed in different ways in the organization of the meeting. Finally, very special thanks are for Prof. Luigi Zicarelli, Honorary President of the Congress, and for Prof. Marcello Mele, Editor-in-Chief of the Italian Journal of Animal Science.

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IS001

Using metabolomics approaches in transition cow biology for basic and applied science

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The transition from late pregnancy to early lactation is a critical period in a dairy cow's life due to the rapidly increasing drain of nutrients from the maternal organism towards the foetus and into colostrum and milk. In order to cope with the challenges of parturition and lactation, comprehensive adaptive reactions comprising the endocrine and the immune system are required. The ability to cope with these challenges is highly variable. Both metabolic and infectious diseases summarised as 'production diseases', may occur during this phase thus impairing welfare, productive lifespan and economic outcomes.
The metabolome, the entity of all metabolites in an organism, represents the outcomes of the genome, transcriptome and proteome integrating also environmental effects. Metabolomics comprises the identification and determination of the set of metabolites in tissues, cells or body fluids under normal conditions as compared to altered states promoted by disease, dietary intervention, or other environmental factors and may also be used to compare physiological responses of different genotypes. Metabolomics technologies are increasingly applied in dairy science. We herein aim to provide a brief overview of the general methodological approaches that are available for assessing the metabolome. We will then review the physiological changes during the transition period and the related production diseases in dairy cows, focussing on metabolic diseases. The longitudinal changes of the metabolome during the transition period as affected by disease, feeding and dietary supplements will be summarised providing examples from the literature. Besides biological fluids such as blood and milk, we will put other metabolically relevant tissues in centre. The current and prospective view of the application of the recent achievements generated by metabolomics together with the bioinformatics evaluation of the data obtained for biomarker discovery, diagnosis and deep phenotyping will also be addressed. In addition, we will briefly introduce the new European Joint Degree Programme 'MANNA' in which metabolomics, amongst other OMIC techniques, are applied to animal nutrition and physiology studies.

Acknowledgements
This contribution would not have been possible without the Joint Mobility Programme PPP by the German Academic Exchange Service (DAAD) and the Italian Ministry of Education, Universities and Research (MIUR) whose funding is gratefully acknowledged.

ANIMAL BREEDING AND GENOMIC – GENETICS OF WELFARE, GENETICS OF RESILIENCE

IS002

Leveraging on high-throughput phenotyping technologies to optimise livestock genetic improvement and husbandry

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The advent of fully automated data recording technologies and high-throughput phenotyping (HTP) systems has opened up a myriad of opportunities to advance breeding programmes and livestock husbandry. Such technologies allow scoring large number of animals for novel phenotypes and indicator traits to boost genetic improvement, as well as real-time monitoring of animal behaviour and development for optimised management decisions. HTP tools include, for example, image analysis and computer vision, sensor technology for motion, sound and chemical composition, and spectroscopy. Applications span from health surveillance, precision nutrition, and control of meat and milk composition and quality. However, the application of HTP requires sophisticated statistical and computational approaches for efficient data management and appropriate data mining, as it involves large datasets with many covariates and complex relationships. In this talk, we will discuss some of the challenges and potentials of HTP in livestock. Some examples to be presented include the utilisation of automated feeders to record feed intake and to monitor feeding behaviour in broilers, milk-spectra information to predict dairy cattle feed intake, and image analysis and computer vision to monitor growth and body condition in pigs and cattle. HTP and big data will become an essential component of modern livestock operations in the context of precision animal agriculture, boosting animal welfare, environmental footprint and overall sustainability of animal production.

LIVESTOCK SYSTEMS – AQUACULTURE

IS003

Efficient aqua feed production and management in a circular future

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Aquaculture is the fastest-growing agricultural sector globally, accounting for almost 50% of the fish consumed in the world. If the projections for human population growth are achieved, an additional 40 million metric tons of aquatic food will be needed to maintain the current per capita consumption rate.

However, current protein production for aqua feed, both animal- and vegetal based is not easily scalable and has environmental impacts in terms of greenhouse gas emissions, land and water use. It is, therefore, of vital importance to find sustainable alternative protein sources that can be economically produced in quantities that meet growing aquaculture.

Terrestrial microorganisms, microalgae and insects are the most three promising sources of alternative proteins that can be produced through innovative and environmentally sustainable bioconversion processes using industrial waste streams.

Microorganisms such as bacteria, yeast, moulds and higher fungi and its products as single-cell protein (SCP) in aqua feeds is one of the best approaches to enhance aquaculture sustainability.

Microalgae are a promising future source for proteins, amino acids, fatty acids (omega-3, especially EPA and DHA), vitamins and bioactive compounds as high-quality ingredients in food and feed, but also represent a potential source of biofuel.

Many insect species have been studied as food ingredient over the last decades, but their cultivation through transformation of organic waste is a relatively new practice. However, despite the proposed benefits of using insects as food and feed ingredient, they remain marginal in the industry, mostly due to low availability and high prices in comparison to conventional protein ingredients.

At farm level, future methods for feeding management will need to be more advanced and smarter. Better precision and monitoring tools in feed delivery to farmed fish would improve the predictability and observability of feed consumption in the fish population, which in turn could enable reductions in production costs and environmental impacts while improving growth and feed efficiency. This could be solved by applying the principles of precision fish farming and the emerging technologies and automated system to shift feeding management from comprising largely experience-driven processes to become a more knowledge-driven procedure.

Round table: FUNCTIONAL FOOD AND HUMAN HEALTH

IS004

From healthy foods to functional foods

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A large body of evidence from epidemiological studies indicates that some foods and dietary patterns are beneficial for the prevention of non-communicable diseases as obesity, type 2 diabetes, cardiovascular diseases (CVD) and some types of cancer. This applies, in particular, to dietary fibre as well as to fruit, vegetables, nuts, wholegrain cereals and legumes. Moreover, mono- and n-6 polyunsaturated fatty acids, and in particular oleic and linoleic acid, n-3 long-chain polyunsaturated fatty acids, plant sterols, potassium, polyphenols, are associated to a lower risk of CVD (Assmann et al. 2014). The identification of the health benefits of some foods and nutrients have led the scientific community to search for food constituents that can influence specific body functions relevant for health thus characterising these foods as functional. Although the functional food market is constantly growing, to date no universally accepted definition for functional foods exists. According to the Concerted Action on Functional Food Science in Europe (FUFOSE) endorsed by the European Commission, a food can be regarded as 'functional' if it is 'satisfactorily demonstrated that it affects beneficially one or more target functions of the body, beyond adequate nutritional effects, in a way that is relevant to either an improved state of health and well-being and/or reduction of risk of disease'. A 'functional food' can be either a natural food (healthy food), or a food to which a component has been added, or a food from which a component has been removed by technological or biotechnological means. Functional foods, however, must remain foods and they must demonstrate their beneficial effects on health in clinical trials performed in humans utilising in amounts that can normally be expected to be consumed in the habitual diet: they are not pills or capsules, but part of a normal food pattern (Diplock et al. 1999). Consumers are informed about the health benefits of functional foods by health claims that are represented by any communication that states, suggests or implies by any means a relationship between a food or a food category and health; they include functional claims, reduction of disease risk claims, or claims referring to the growth and development of children. The European Food Safety Authority (EFSA) scientifically evaluates whether a food is able either to influence a biological function or to reduce the risk of disease. Examples of functional claims are 'sugar-free chewing gum helps reduce tooth demineralization' or 'plant sterols contribute to keep plasma cholesterol levels under control'. Reduction of disease risk claims are i.e. 'calcium reduces risk of osteoporotic fracture' (over 65 years) or 'dietary fibres contribute to decrease the risk of colon cancer'.

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A claim is the only guarantee for consumers of the health effects of a functional food and a necessary support to make the appropriate product's choice.

To authorise a claim, EFSA requires the following conditions:

1. the food /constituent is well characterised;
2. the claimed effect is well defined and physiologically relevant;
3. a cause-effect relationship exists between the food/constituent and the claimed effect for the target group and for the proposed conditions of use.

Benefits of functional foods have frequently been reproduced by providing isolated components of foods as supplements identified as nutraceuticals. It is mandatory that, before health claims for particular foods or nutraceuticals are authorised, clinical trials in humans are undertaken demonstrating their beneficial effects on human health.

An example of a new potential functional food is represented by a rusk made with a flour enriched in amylose (a type of starch with a slower digestibility) that improves postprandial glucose metabolism thus contributing to reduce the impact of glucose and insulin on the arterial wall thus, possibly, contributing to reduce the risk of cardiovascular diseases. In this respect, there might also be meat-derived products showing functionality in relation to the impact on risk factors for cardiovascular diseases. Along this line there are data showing that by manipulating the diet of rabbits it is possible to increase the meat levels of PUFA, CLA, EPA, DHA, vitamin E, selenium, molecules that play a key role in controlling relevant risk factors for CVD and other chronic diseases (Dalle Zotte and Szendro 2011). The implementation of this type of strategy applied to other animal species, combined with wild breeding could improve the quality of the meat and their derivatives in respect to potential health benefits.

Nowadays, functional foods represent a challenge for researchers and the food industry that needs to implement and/or develop innovative technologies able to improve the health properties of foods without compromising the organoleptic features that represent the basis for the everyday consumption. In this light, the use of appropriate intervention strategies able to increase the functionality of meat products would be of paramount importance, given the relevance of meat in the human diet in relation to its nutritive properties linked mainly to its important contribution of proteins of high nutritional value.

In conclusion, healthy foods and functional foods with demonstrated health effects which can be easily included in a real-life dietary pattern should represent the cornerstone of a feasible strategy to reduce the risk of non-communicable diseases in the general population.

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IS005

Functional metabolites in Mediterranean buffalo productions: impact of innovative breeding techniques

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Despite the considerable progress made in the epi-nutrients field, it is now clear the importance of advanced technologies aimed at improving the quality of the productions. The development of food production of 'nutritional and functional quality' cannot ignore animal welfare, food safety and environmental protection. In this context, the development of new breeding techniques capable of improving the functional metabolite profile of animal productions is the road ahead.

Mediterranean buffalo (Bubalus bubalis) milk has emerged to be particularly rich in bioactive components derived from carnitine, such as short-chain acylcarnitines (acetyl-carnitine, propionyl-carnitine, butyryl-carnitine), and betaine (glycine betaine, δ-valerobetaine). Among these, δ-valerobetaine (N,N,N,-trimethyl-5-aminovaleric acid), a water-soluble trimethylated compound, recently identified as constitutive betaine of ruminant milk and meat, is synthesised by rumen microbiota most likely during the anaerobic oxidative deamination of Nε-trimethyllysine and glycine (Stickland reaction). Of note, milk content of short-chain acyl-carnitines and δ-valerobetaine improve depending on the animal 'well-being' with a concomitant increase of the *in vitro* antioxidant and anti-inflammatory properties. These evidences have even greater relevance in light of the fact that milk δ-valerobetaine is active in modulating the epigenome via SIRT1 and SIRT6, suggesting a potential as a novel dietary compound with health-promoting properties.

Acknowledgements

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IS006

Antioxidants in animal nutrition: benefits and challenges

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Over the past decades, a number of studies investigated the importance of an adequate antioxidant status to sustain both animal health and production. Farm animals can experience oxidative stress (OS) during their life, such as in the periparturient period, at weaning, during transport or in condition of heat exposure. In dairy cow, particularly, the increased lipid mobilisation, associated with the onset of lactation, can increase the severity of OS and a consequent relationship with negative energy balance can be observed. Supplementation of dietary antioxidants or boosting endogenous antioxidant defences of the body have been proposed for maintaining the redox balance. Among dietary antioxidants, vitamin E and selenium are the most recognised, even though this category includes several classes of other compounds, such as vitamin C, carotenoids and polyphenols, all of them largely advised as feed additives or ingredients in farm animal nutrition. Recently, agri-food waste and by-products have been also proposed as possible sources of bioaccessible phenols and antioxidant compounds that can be used as functional ingredients in feed. Research made significant advances in the definition of adequate and standardised biomarkers of oxidative stress, making it possible to better define the subsequent effect of antioxidant supplementation, and address some regulatory discrepancies about these substances.

OPENING CEREMONY

IS007

Modern breeding, taste of products of animal origin and future of Italian breeding

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Modern breeding aims to reduce the cost of food of animal origin to encourage its consumption also for to the less well-off. The concept of quality did not take into account the sui generis taste but the longest possible shelf life, a primary requirement for large retailers. Even 'fresh' cheeses can have a commercial life until 30 days when the 'freshness' is a distant memory.

Paradoxically, the rules were dictated by the North Europeans who do not have to teach anything in the food sector but above all consider the economic factor. In fact, the areas that gave rise to 'typical' productions had to ask for exemptions to adapt production to the rules of those who 'copied' many products.

The cured meats were created using only salt thanks to the preservability of the meat of Mediterranean pigs. The Lucanian slaves introduced the 'lucaniche' to Rome, which in Brazil are called calabreza and, much earlier, the Etruscans (perhaps also the Spaniards), gave life to the ham.

Later, the Anglo-Danish genetics produced insipid meat not suitable for the production of typical cured meats for which, with the exception of PDO products, the use of nitrates is required. This genetic has considerably reduced family production that only involves the use of salt, a technology unsuitable for the preservation of this type of meat. Pork for direct consumption in Northern Europe is represented by increasingly lighter pigs, while in Italy, recently, by increasingly heavy pigs, and this reversal in 2018 has increased consumption by 4%.

The poor organoleptic quality of chicken meat did not prevent the increase in consumption for the price and the impact on GW caused by ruminants, which I totally disagree with. The beneficial effect on human health of chicken meat does not take into account the fact that its growth is faster than that of a neoplastic mass (50 times in 45 days). Also, the consumption of turkey meat is increased, animal that, if released in nature, would not be able to reproduce.

The classification of bovine carcases by the EU rewards subjects characterised by muscular hypertrophy similar to that obtained with anabolics. It does not matter if the meat of these subjects no longer has the flavour 'sui generis', so it can not be distinguished, if evaluated 'blindly', from that of the chicken or the pig. The carcases of a Rubia Gallega, a Chianina, a Nelore or a Podolico raised in the pasture are penalised. The carcase of an industrially reared 'Bleu Belga' beef provides highly appreciated tasteless meat. The omega-6 ratio: omega-3 of steak of industrial production can reach 30:1 while grazing animals have a ratio of 2:1. The cattle fed as 'ruminants' grow less but provide leaner meat that reduces LDL cholesterol levels 'bad' and three to five times richer in CLA and vitamin E. Buffalo meat, regardless of rationing, provides lean meat. Finnish women who have received higher levels of CLA in their diet have a 60% lower risk of breast cancer than those who have had lower levels of CLA. The pastures in Italy are scarce but it is not impossible, in the final phase, to use diets with more fodder to give the meat more favourable nutraceutical characteristics, exactly the opposite of what happens today.

Similar considerations apply to milk. Recently, thanks to an experimental plan devised by Infascelli, Lombardi and Mollica, it has been shown that cows' milk fed with a fodder/concentrate ratio: 70/30 favourably influences lipid metabolism in rats, the leptin/adiponectin ratio decreased the amount of inflammatory substances and those responsible for oxidative stress and the accumulation of lipids in the liver.

From these results, it can be said that the less productive breeds should be preferred, because they are more suitable for rationing with a forage/concentrate ratio of 70/30 capable of supplying nutraceutical milk capable of ending the demonisation of products of animal origin. Recently, a series of functional molecules useful for human health has been highlighted in the milk and meat of ruminants, whose presence can be influenced by breeding techniques (Campanile et al.).

The sheep and goat products are certainly the best from a nutraceutical point of view. However, their consumption has decreased gradually due to incomplete information on the media that have always avoided showing that the most long-lived Italian populations are those inland areas of Sardinia and Cilento which, together with the Caucasian ones, constantly consume products of sheep and goats.

Further reduction of costs, production of functional foods or exaltation of flavors? On the harmony between these needs will have to work the Italian zootechny of the future, not underestimating that the increase in production coincides with tasteless tastes. The consumption of increasingly 'tasteless' food is the premise to encourage the consumption of worms, crickets and 'Shmeat' meat, proposed, not by chance, by researchers belonging to peoples who have no taste and culinary traditions. The future problem of breeding is not the cost of products, but their taste and the presence of nutraceutical substances.

IS008

The role of the vet in the one health

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The Vet plays a pivotal role in the declination of the environment-health combination: a professional who is able to mediate between the environmental, food and health sectors. He identifies its strength in the overall view and, through the application of specific knowledge, he is able to analyse each single detail, thus defining the keys to reading and understanding both the phenomena and the risks.

The 'naturalistic sensitivity', which distinguishes either the training and the approach of the Vet, determines an innate sense of holistic protection towards environmental systems and their organisation: the vet becomes a driver in the paths of knowledge, study, operational strategy, useful for the single ENVIRONMENT system.

Thanks to the 'One Health' perspective, a multi- and inter- disciplinary profile is outlined, a collection of common intentions for the ecosystems protection and the safeguard of public health. From this point of view, by acquiring these skills and knowledge, the Vet becomes the main 'tool' in the resolution of the 'Environmental Crimes', in the conditions of illegality that blame the territory and burden as criminal offences on the environmental system. The skills of the Vet are to monitor, control and protect the welfare of the ecosystems, to safeguard biodiversity, to carry out and implement epidemiological-surveillance plans for zoonoses and environmental related diseases, to spread the culture of primary prevention medicine and public health, to apply an integrated and multidisciplinary approach for understanding the phenomena and to frame the actions in a systemic view, through a rational experimental approach of collective utility. Q4

During their activities, the judicial authorities draw from the technical-scientific support and the overall view of the Vet, who supports the elaboration of experimental evidences, towards the deterministic cause-effect interpretation: therefore, the Vet represents a fundamental figure in identifying correlation links and criminal responsibilities, according to the 'who pollutes, pays' principle. The principles of prevention, precaution, correction and integration are basilar for supporting the judicial authorities in order to avoid environmental crimes and crimes against public health and environment.

The Vet who works for the 'One Health' is not a mere utopian representation, but anticipates the needs of the system. In the last years the IZSM translated into practical experience this pioneering ideal, supplying its contribution to the Campania region, i.e. when the 'Terra dei Fuochi' phenomenon emerged, through experimental evaluations and investigations in different fields, from the health of the food to the environmental monitoring, the epidemiological observational study on susceptible population, up to a plan of primary prevention and oncological screening. The strong collaborations with ministerial and government institutions, public prosecutors and law enforcement agencies, provide scientific and operational tools to clarify the multiple and complex health-environmental phenomena and emergencies. In fact, the complexity does not hinder to read the phenomena, but put them through different levels of interactions and fields of investigation, requesting a dynamic and multidisciplinary vision in their analysis: this is the typical vision of the Vet in an 'One Health' perspective.

During the experimental activities that were focussed primarily on the link between food, environment, ecosystem welfare and human health, the IZSM defined a *modus operandi* based on in-depth and widespread knowledge of the territory, that identifies parameters of risk assessment and critical issues in order to define experimental and operational models. Thus, the 'Comparative Evaluation of Risk' and the 'Multivariate Analysis' become the basis of the experimental designs and planned analytical activities, aimed to assess the health status of several substrates (soil, air, water, vegetables, animals, food) and humans, and to identify critical issues and illegal activities.

The IZSM provides continuous support to the judicial authorities in the evaluation of crimes against the environment through several activities: those defined by the Ministerial Decree 'Traceability of the Buffalo Supply Chain' (Implementation Decree 9 September 2014); those regarding the Quality and Health Assessment System for Campania Products through the 'QRCODE Campania' project; the integrated monitoring plan on environmental, animal and plant matrices 'Campania Trasparente'; the human biomonitoring plan 'SPES – Exposure Study in Susceptible Population', an epidemiological observational study on the population resident in Campania, exposed to potential sources of contamination; the monitoring plan on emerging zoonoses 'HEV Hepatitis E Virus'; the training of law enforcement agencies for recognition of Hemp authorised crops, with reference to genetic and chemotypical database 'S'Attiva';
Q5 the cancer screening plans 'First Prevention' [1–4]. The implementation of research and development activities, the monitoring and certification of food products quality and health, allowed the IZSM to combine, in an integrated action, the needs and requirements of a territory considerably vexed from environmental emergencies. This encountered the will of the scientific community and of the authorities to reply, confirming or refuting the hypotheses on causes and correlations between environment, contamination, food and health. Everything was done raising the professional figure of the Vet to guide advancement and scientific vanguard, in support of the protection of the entire environmental system.

IS009

For a better future of beef production: improved carcase grading, eating quality and livestock systems, but no cultured meat

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So far, the beef market has been driven by carcase grading to satisfy all the actors of the supply chain from farmers to butchers and retailors. Therefore, classification systems have been solely designed to describe the carcase for the purchaser, with no real objective to satisfy consumers at the end of the whole chain. However, some grading schemes (such as the USDA classification) confer both a yield grade and a quality grade to each carcase. In addition, official and commercial quality schemes for retail beef have been developed in relationship with origin, tradition or eating quality, each with their own focus and methods of evaluating and describing carcases and/or beef. Consequently, the definition of quality varies for each destination market and mainly depends on characteristics of farming systems and/or animal/carcase. Despite all these initiatives, there is still a high level of variability in beef palatability and a low relationship between eating quality of beef and its price, which induces consumer dissatisfaction. Therefore, a great deal of research has been developed to set up tools and equations to predict beef eating quality based on physical measurements (mechanical methods or spectroscopy) or biological markers (genomic or phenotypic predictors). However, all these approaches are confronted with the complex determinism of quality parameters, especially tenderness. Concerning spectroscopy, the main encouraging results were obtained using Near Infra-red Spectroscopy (NIR) for the prediction of the composition and nutritional value of meats.

More recently, Australia has developed a unique and consumer-driven grading system called Meat Standards Australia (MSA). It is based on different beef eating quality scores (namely tenderness, flavour liking, juiciness, and overall liking) at the muscle level in interaction with the cooking method. The four eating quality scores are then combined into a unique global quality score, which is predicted from animal and carcase traits such as sex, age, breed, marbling, hot carcase weight and fatness, ageing time, etc. An MSA-like grading scheme could be used to predict beef eating quality in Europe and other countries, including in Asia and America. Hence, it could underpin the existing official labels or commercial brands by helping them to provide beef of satisfactory eating quality, which is a prerequisite to satisfy consumers. An MSA index has recently been proposed based on the global quality scores of each cut to recalculate an eating quality potential at the carcase level. It may be implemented to improve eating quality through genetics and management. Finally, because consumers from many countries are willing to pay more for a better quality product, such an eating quality guarantee system is likely to generate economic benefits to be shared along the beef supply chain from farmers to retailors.

Nowadays, the livestock and meat sectors are facing new and important challenges such as their environmental impact and role in global climate change; the need for increased availability of food due to the human population growth, the need to address societal needs (e.g. animal welfare and ethics) and the need to produce meat of high quality (health, safety, palatability). Therefore, at the consumer or citizen level, meat quality includes intrinsic qualities (characteristics of the product itself) and extrinsic qualities (animal health and welfare, environmental impacts, etc.), but there are trade-offs among and between intrinsic and extrinsic qualities. As for eating quality with the MSA grading scheme, a relevant combination of indicators related to intrinsic and extrinsic quality traits (including carbon footprint, animal welfare, grassland biodiversity, rural development, etc.) and economic efficiency (e.g. income of farmers, price for the consumer) will allow the prediction of the overall quality of beef for consumers, but also for any stakeholder of the supply chain.

The beef industry cannot respond to these changes in demand by business as usual. It must find solutions to many issues regarding not only eating quality (as already done by the MSA grading scheme), but also nutritional quality, animal welfare, health and more generally sustainability. It has to do so in the face of competition from emerging protein products including new plant-based products and cultured meat. Despite a high media interest, the latter is probably the worst alternative for different reasons: there are still numerous technological obstacles that have to be overcome to produce in vitro meat, its safety and nutritional value are not guaranteed, its advantages for the environment and its potential acceptance by consumers have not been demonstrated yet. Furthermore, the artificial nature of cultured meat goes against the growing demand for natural products in many countries, while conventional meat producers are moving towards concepts of agroecology to develop environmentally friendly and sustainable animal production systems.

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – ANIMAL HEALTH

IS010

Oxylipids and inflammatory disorders of dairy cows

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Dairy cattle are susceptible to increased incidence and severity of health disorders during the transition period. A common link contributing to the development of both metabolic and infectious diseases in transition cows is a dysfunctional inflammatory response. The role of inflammation is to eliminate the source of infection or tissue injury and then return tissues to normal function. An unlimited inflammatory response, however, can cause damage to host tissues and contribute significantly to the pathophysiology of economically important diseases such as mastitis. A delicate balance between pro-inflammatory and pro-resolving mechanisms is needed to ensure optimal pathogen clearance and the prompt return to immune homeostasis in transition cows. Therefore, inflammatory responses must be tightly regulated to avoid bystander damage to host tissues. Oxylipids are potent lipid mediators that can regulate all aspects of the inflammatory response. The oxylipid network is complex, highly interactive, and often cell-specific in orchestrating the onset or resolution of inflammatory responses. At present, there is ample evidence in dairy cattle to suggest that oxylipid biosynthesis is controlled at several levels including the amount and availability of polyunsaturated fatty acid substrates, the relative activity and substrate preference of oxidising enzymes, and the degree to which intermediate metabolites are catabolize to their end products. The biosynthetic profiles of oxylipids and the subsequent impact that these metabolites may have on the character of the inflammatory response is also likely depend on the timing of their subsequent metabolism through various oxidising pathways. This presentation will provide a brief overview of the bovine inflammatory response and the role that oxylipids play in contributing to the onset and resolution of inflammation especially as it pertains to mastitis. Factors associated with transition cows that can contribute to dysfunctional regulation of inflammation as a function of altered oxylipid biosynthesis will be described. Specifically, there is a dramatic increase in linoleic and arachidonic acids concentrations in both plasma peripheral blood leukocytes during the transition period of dairy cattle. The impact that linoleic-derived oxylipids play in regulating bovine mammary gland endothelial cell functions in response to challenge with mastitis-causing pathogens will be presented. We will also show how arachidonic acid-derived lipid mediators metabolised by the cytochrome P450 enzymatic pathways can contribute to bovine vascular dysfunction during coliform mastitis. Nonsteroidal anti-inflammatory drugs are often used to modify the cyclooxygenase pathways that control prostanoid biosynthesis. However, we will show that changing substrate availability and the degree of lipid mediator metabolism are alternative approached that can modify the expression of pro-inflammatory oxylipids. A greater understanding of the factors that can regulate the delicate balance between the initiation and resolution of inflammatory responses is needed in order to diminish the morbidity and mortality associated with health disorders of dairy cattle such as mastitis.

ANIMAL BREEDING AND GENOMIC – NEW BREEDING TECHNOLOGIES II

IS011

Emerging topics in livestock breeding and production

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The next future of livestock will be driven by two world needs, as the global population and global wealth both continue to increase so will the demand for livestock products, especially those which are highly nutritious and socially sustainable. However, high competition with other uses for land and water resources will also be intensified, hence more socially acceptable, efficient and sustainable livestock production will be needed. The application of emerging knowledge, integrated with new technologies, such as high-performance phenotyping, livestock farming precision, traits ontologies, genome editing, microbiome and deep learning, will play important roles in meeting these challenges. Aim of this review is to describe the emerging topics in livestock breeding and production sectors.

High-throughput phenotyping (e.g. near- and mid- infra-red spectrometry-MIR) of farm animals appears to be essential given the new societal and scientific challenges that will have to be faced to ensure sustainable animal production. It can be defined as a method of determining phenotypes that can be subject to rapid, repeatable and automated measurements that generate significant volumes of data. Another way to collect large amounts of data is achieved through many livestock farming precision technologies (LFP), using robot and sensors, for daily milk yield, meat and eggs recording, milk component monitoring (e.g. total fat, fatty acid profile, total protein, protein profile, acidity, coagulation properties, somatic cells count, differential somatic cells count), energy balance, enteric methane, pedometers, automatic temperature recording devices, milk conductivity indicators, automatic oestrus detection or calving monitors, and daily body weight measurements, already being utilised by producers. In this context of high-throughput phenotyping, thanks to LFP and sensors, obtaining phenotypic information that is accurate, reliable, repeatable and comparable between laboratories, countries or companies, is critical to gain a better understanding of the relationship between genes and phenotypes, and to develop LFP using robust and resilience animals. Up till now, it has been extremely difficult to combine various sources of different phenotypic data from databases of many different origins due to the variability in phenotyping techniques and the absence of any information on livestock breeding conditions. To remedy these problems, a common language with shared, unambiguous definitions of traits and their measurement methods is needed. A traits ontology is a formal, structured representation of a set of objects (in this case, animal traits or measurements), and of the relationships between these objects. In an ontology approach, the concepts are clearly defined and organised in a structured manner (often a hierarchical structure).

Genome editing (GE) is an emerging genetic way, that enables to modify genetic material in targeted ways. In the context of animal breeding one use of GE could be to fix a small number of undesirable alleles in individuals that have otherwise high breeding values. Such an approach could make GE very complimentary to genomic selection (GS). Indeed, individual animals could be first selected on the basis of GS and then have some of their unfavourable alleles 'fixed'. Livestock animals, mainly ruminants, have developed a symbiotic relationship with anaerobic microorganisms, being able to convert feed into food products useful for human consumption, such as meat, milk and eggs and for guaranteeing a good health status. Analysis of the animal gut microbiome, using next-generation sequencing studies, suggests that the diversity and composition of the microbial communities co-diversified with their hosts, being influenced by diet composition, host genetics, geographical location, and environmental factors. The know and use of microbiome will permits to improve how the symbiotic relationship between livestock animals and their microbes can affect the host productivity and welfare. The traits vary significantly with both genetics and environmental factors, and these variations can be predicted using learning algorithms that can learn from the current data to predict the animal's future performance. The term deep learning (DP) is often associated to artificial intelligence (AI), Big Data and analytics. The DP it is an approach to AI which is showing great promise when it comes to developing the autonomous, self-teaching systems which are revolutionising many industries and recently also the livestock sector and food industries. Essentially deep learning involves feeding a computer system a lot of data, which it can use to make decisions about other data. This data is fed through neural networks, as is the case in machine learning.

In conclusion, breeding and management of livestock animals are changing very fast due to the application of emerging knowledge, integrated with new technologies, such as high-performance phenotyping, livestock farming precision, robot, sensors, traits ontologies, genome editing, microbiome and deep learning. All these changing and emerging topics are requiring careful data quality validation, discussing on legal issues and to data accessibility in general. A clear understanding of these promising tools is strategic for their valorisation and to make this data useful, also off-farm and facilitating data definition, consolidation and integration layer. Novel concepts as traits ontology, gene editing and artificial intelligent need to be diffused and applied in the livestock sector. However, need to improve the cost-benefit relation and to solve the ethics aspects for these emerging technologies, which are currently some of the major obstacles for their large-scale adoption.

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ENVIRONMENTAL SUSTAINABILITY – ENVIRONMENTAL IMPACT OF LIVESTOCK II

IS012

Feed efficiency, microbiome and sustainable animal production

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The role of animal products (meat, dairy) in global food systems is projected to grow as a result of increasing demand that is being driven by world population growth, global demographics and dietary preferences. Food production requires significant natural resources and it has been estimated that one-third of the world's land surface and around 75% of freshwater are used for crop or animal production (IPBES 2019). There is mounting global pressure on the impact of animal production on the environment, which requires an urgent response.

The provision of feed accounts for 70–75% of the cost of animal production in both extensive and intensive systems. An improvement in the efficiency by which animals convert feed into energy benefits the environment (less natural resources including less land) and also improves profitability. Both resource requirements and profit are related to the ability to establish sustainable production systems. Feed efficiency has a genetic component as demonstrated in cattle (Li and Guan 2017; Lovendahl et al. 2018), goats (Dzakuma et al. 2004), pigs (McCormack et al. 2017), poultry (Leinonen and Kyriazakis 2016) and sheep (Paganoni et al. 2017). A genetic predisposition for good feed efficiency can be complemented by the provision of highly digestible and nutritious feed (Godinho et al. 2018). Hence, there is considerable scope to select and breed efficient animals that are provided with optimal diets, and with both animals and diet matched to production goals. In ruminants, good feed efficiency is associated with reduced methane production which is an additional environmental benefit (Lovendahl et al. 2018).

The new frontier in feed efficiency is research on the interrelationships between the gut microbiome and feed utilisation (Malmuthuge and Guan 2016). The microbiome refers to the collection of micro-organisms, including bacteria and bacteriophages, that inhabit the gut and intestine and which interact with the host in health and disease. Information is emerging on the microbiome in cattle (Meyer et al. 2015; Li and Guan 2017; Schären et al. 2018), goats (Mao et al. 2014), pigs (Holman et al. 2017) and poultry (Pan and Yu 2014; Waite and Taylor 2014). Relationships have been found between the microbiome profile and feed efficiency in cattle (Meyer et al. 2015; Li and Guan 2017) and pigs (McCormack et al. 2017; Yang et al. 2017). Diet was shown to influence the microbiome in pigs (Tilocca et al. 2017; Verschuren et al. 2018) and poultry (Pan and Yu 2014). These preliminary reports have shown how new knowledge on the microbiome can be utilised to transform feeding systems in animal production in order to optimise the efficiency of production, whilst reducing environmental impacts.

There is a clear opportunity for major advances in animal production by matching and integrating the genotype (feed efficiency) with the microbiome and feed. The result will be a reduced demand for natural resources and a move to greater sustainability in animal production.

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ANIMAL PRODUCTS – MEAT QUALITY

IS013

The oxidation of the lipids in meat. The process, the effect on human health and the procedures for a correct quantification

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Meat lipid oxidation (MLO) represents, with the exception of microbial alteration, the process which most negatively affects meat quality. Lipid oxidation is a non-spontaneous process (the molecular oxygen can not thermodynamically interact with a double bound of an unsaturated lipid), so needs some catalysing substances to take place. MLO starts from unsaturated lipid mainly contained in the phospholipids of membrane of cells and organelles. Fatty acid oxidation is no related to chain length, but to the number of double bounds and bis-allylic hydrogens. After the initiation, the process goes on to the hydroperoxides; this happens rapidly (is self-propagating and self-increasing) and intensively (many more than one hydroperoxide is produced per initiating event). Hydroperoxides are the primary oxidation products and can be produced also enzymatically in a no-radical way. Hydroperoxides are quite stable, however, a huge variety of reactions start from them, thus many secondary products such as cyclic peroxides, multi hydroperoxyl-derivative, aldehydes, ketones, alcohols can be formed. The cholesterol too is an unsaturated lipid which undergoes oxidation through analogous ways of the fatty acids, producing COPs (cholesterol oxidation products). Most of the secondary oxidation products, both of fatty acids and cholesterol are harmful for the human health.

Meat lipid oxidation is as a result of a very complex combination of pro-oxidant and anti-oxidant items. Former include the lipid composition, reducing metals content (iron specifically), myoglobin amount and type, pro-oxidant pool enzymes (both endogenously and exogenously produced); the latter comprise substances able to chelate metals or quench radicals, so contrasting the start or limit propagation of oxidation, respectively.

MLO arises throughout the whole process of meat production either before, during or after animal slaughtering. The species of animals is an important ‘before-slaughtering’ factor for MLO. Meat from ruminant shows a higher content of myoglobin (about 70–80% of total iron) than poultry or turkey meat (less than 30% and about 40% respectively). The haem iron is a pivotal item in the MLO. On the other hand, poultry and turkey meat show a more unsaturated fat (so, more prone to oxidation) than ruminant meat. The rearing system is another important before-slaughtering factor; for instance, grazing animals show more unsaturated lipids but, contemporaneously, higher content of vitamin and antioxidant substances than animals reared in feedlot; some lipid supplementation can modify fatty acids composition of meat. Moreover, animals’ movement can affect the kind of muscle fibres. They differ in dimension (red ones are smaller than the white ones) and in the metabolism (red, oxidative; white, glycolytic). Dimensions can affect the lipid composition (small fibre shows more phospholipids and cholesterol than bigger ones), while the kind of metabolism affect the enzymatic pool (oxidative or glycolytic).

Slaughtering represents a key step in MLO; wrong procedures stress the animals and induce them to the inflammatory cascade which involves some oxidised lipid (5-HPETE from arachidonic acid for instance). On the other hand, also a correct slaughtering procedure negatively affects MLO; after death, antioxidant enzymes activity is reduced, cell compartmentation is destroyed, and free metals are increased.

Obviously, a huge variety of actions such as meat conservation, cooking, manipulation, are able to affect MLO, by limiting or increasing the interaction of the lipids with oxygen, light and temperature. For instance, before consuming, meat from ruminants, is conserved under a modified atmosphere packaging containing 60% or more of oxygen. This preserve meat from discolouration, allowing to maintain myoglobin as oxymyoglobin (bright red), but, contemporaneously, oxymyoglobin is able to trigger MLO.

Analytical procedures for a correct quantification of MLO has to be carefully planned, as is it appropriate to check and quantify some proxies of all MLO steps: free fatty acids, conjugated dienes, primary and secondary oxidation products. The evaluation of secondary oxidation products of MLO is a key point. The most used test to do it is the TBARS test. This is a good proxy of oxidation of fatty acids with more than two double bounds, as MDA come from a breakdown of cyclic peroxides derived from these kinds of fatty acids. Nevertheless, the high unsaturated long chain fatty acids represent, often, a minor part of total fatty acids of meat; in this eventuality, the quantification of other substances respect to MDA is mandatory. Volatiles aldehydes quantification is useful, as they are able to give information on the oxidation of a specific fatty acid. Finally, the cholesterol oxidation has to be checked due to COPs harmfulness. Moreover, COPs can be considered as very good proxy of the whole MLO; the oxidation of B ring of cholesterol produces, in fact, only five or six compounds which do not undergo, like fatty acids, a molecule breakdown.

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE

IS014

Late gestation heat stress in dairy cows: effects on dam and daughter

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Heat stress has profound negative impacts on the productivity of dairy cows during lactation, but less is known regarding the effect of heat stress in dry cows. Recent studies from our laboratories indicate that heat stress of non-lactating, late gestation animals, i.e. dry cows, dramatically reduces milk yield in the next lactation and alters the developing foetus to induce epigenetic changes that may explain, in part, the less productive phenotype at maturity (Dahl et al. 2017; Skibiel et al. 2018). The design of our studies consists of cows being exposed to temperature humidity index (THI) above 72 from the time of dry off through parturition, in comparison to herdmates that have access to active cooling systems. The targeted length of the dry period is 45 days and all of the cows in our studies are second parity or greater, and other than heat stress, the management of the cows is identical. Heat stress cows then typically have rectal temperatures 0.4 to 0.5 °C higher than cooled cows, and a respiration rate of 70 to 80 breaths/min for the entire dry period. Thus, in the absence of access to fans and soakers, cows become profoundly heat stressed. The most apparent impact of heat stress in the dry period is lower milk yield in the subsequent lactation, usually in the range of 4–5 L/d for the entire lactation (Tao and Dahl 2013). Using a serial mammary biopsy approach, we determined that heat stress of dry cows limits mammary development late in gestation, likely as a result of reduced placental function which is characterised by altered endocrine output of the placenta. Heat stress also reduces dry matter intake, but other impacts on insulin and energetic metabolites are limited in the dry cow. Dry cows that experience heat stress have reduced immune status with lower leukocyte proliferation and antibody response to antigens (Tao and Dahl 2013). Thus, heat stress in late gestation depresses immune status, lowers intake and reduces mammary growth relative to cows that are actively cooled. This is consistent with results of a comparison of records of over 2500 cows on a commercial farm that had the dry period during the hottest (June–August) or coolest (December–February) months of the year. When dry in the summer, cows produced less milk, had higher incidence of mastitis and respiratory disease, and poorer reproductive outcomes relative to cows on the same farm that were dry in the coolest months (Thompson and Dahl 2012). We have estimated these negative effects on production to cost the US dairy industry over $800 million annually (Ferreira et al. 2016).

In addition to the effects on the dam, the developing foetus has altered physiological responses to adapt to elevated temperatures in utero. Calves from heat stressed dams are born at a lower bodyweight, have reduced passive transfer, and lower survival to puberty (Monteiro et al. 2016). In addition, in utero heat stress is expressed as a lower milk yield phenotype in the first lactation. This lower yield is not a result of growth related differences per se, as calves from heat stressed and cooled dams are of comparable bodyweight at first parturition, and no bodyweight differences are noted during the first lactation. Further studies have confirmed that the yield drag persists into the second and third lactation is transmitted to their offspring (Laporta et al. 2018). Most recently, we have determined that the pattern of DNA methylation is affected by in utero heat stress, and there is consistency of the affect across multiple tissues and ages (Skibiel et al. 2018). Collectively this evidence suggests that elevated temperatures in utero act via epigenetic mechanisms to confer altered, lower productivity phenotype on heifers gestated in dams under heat stress in the dry period.

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – COMPANION ANIMALS

IS015

Nutritional modulation of canine and feline intestinal microbiota

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The bacterial populations that inhabit the intestinal tract play a role of great importance in animal health, as they are involved in nutritional, functional and immunological processes (Suchodolski 2011). A healthy stable microbiota is able to reduce the risk that invading pathogens will cause disease. Intestinal microbes produce vitamins and take part in digestive processes, thereby providing nutritional support for the enterocytes. Moreover, the presence of the intestinal microbiota is essential for the development and stimulation of the immune system.

There is a clear evidence that the symbiotic relationship between intestinal microbiota and the host may be positively modulated through dietary strategies that include the utilisation of indigestible fermentable carbohydrates (prebiotics) and specific microorganisms (probiotics).

The intestinal microbiota of dogs and cats

Recently, thanks to the use of molecular identification methods, the intestinal microbiota of dogs (Hand et al. 2013) and cats (Minamoto et al. 2012) has been characterised. Based on these studies, like in other mammals, the phyla Firmicutes, Bacteroidetes, Proteobacteria, Fusobacteria and Actinobacteria account for over 99% of the bacterial species harboured in the intestine of dogs and cats. However, a huge variability exists among animals belonging to the same species.

Diets differing in their composition are likely to have an influence on the intestinal microbiota. For example, diets containing high levels of protein may lead to higher faecal concentrations of putrefactive compounds deriving from bacterial proteolytic reactions (Pinna et al. 2018). Feeding extruded commercial or raw diets may also result in differences in the gut microbiota of dogs (Sandri et al. 2016) and cats (Kerr et al. 2014).

Effects of prebiotics and probiotics on healthy animals

Prebiotics are non-digestible food ingredients (part of dietary fibre) that stimulate the growth and/or activity of beneficial bacteria in the hindgut. Prebiotics may reduce intestinal proteolysis and production of putrefactive compounds. Moreover, prebiotic substances result in increased production of short-chain fatty acids with a reduction of luminal pH (lowering the concentrations of pH-sensitive pathogens and improving mineral absorption; Pinna and Biagi 2014). Among prebiotics, inulin, beet pulp, and fructo-oligosaccharides are common ingredients added to commercial pet food. However, responses to dietary prebiotics are variable in pets, due to different baseline levels of colonic target-bacteria, dietary macronutrient intake, and antibiotic usage.

Probiotics have been defined as living organisms (bacteria and yeasts) that, upon ingestion, can improve the health of the host. Probiotics exert their effects through interactions with the immune system, modulating its functions. Furthermore, probiotics interact with the host microbiota through the production of antimicrobial factors (bacteriocins and organic acids) and competing for adhesion sites and nutrients (Wynn 2009). Interestingly, it has been seen that also fractions of yeast cell walls (mannan oligosaccharides and beta-glucans) possess the ability to stimulate immune function and increase resistance to intestinal pathogens (Broadway et al. 2015). However, probiotic action may vary with different dosages, bacterial strains, host age, health condition, and nutritional status. At present, two bacterial strains (*Enterococcus faecium* NCIMB 10415 and *Lactobacillus acidophilus* CECT 4529) are currently authorised in the European Union for dogs and cats whereas a strain of Bacillus subtilis (C-3102) has been authorised only in dogs.

Effects of prebiotics and probiotics in animals suffering from chronic kidney and liver disease

Prebiotics act as a source of energy for the intestinal microbiota, increasing bacterial protein anabolism and decreasing blood nitrogen concentrations. This mechanism, in combination with a dietary protein restriction, may provide benefit to animals suffering from hyperammonaemia or azotaemia (in case of liver failure and chronic kidney disease, respectively; Rochus et al. 2014). Even probiotics might be used in the treatment of chronic kidney disease, as some specific strains are supposed to lower systemic uraemic toxin loads as they use nitrogen as a growth source (Wynn 2009).

Effects of prebiotics and probiotics in animals suffering from gastrointestinal disorders

An appropriately balanced microbiota generates anti-inflammatory molecules such as short-chain fatty acids, polysaccharide A, and peptidoglycans (Tizard and Jones 2018). When dysbiosis occurs, intestinal homeostasis is altered and this may contribute to the development of inflammatory processes in the gut. It has been seen that dogs suffering from chronic enteropathy display lower bacterial diversity and distinct microbial communities when compared with healthy dogs (Minamoto et al. 2015). Due to beneficial effects on inflammatory processes, probiotics (including both bacteria and yeasts, e.g. *Saccharomyces boulardii*) may be used to treat acute and chronic enteropathies in dogs (D'Angelo et al. 2018).

Conclusions

Today, there is clear evidence that the gut microbiota of dogs and cats can be modulated by feeding specific dietary ingredients or supplements. Moreover, improved composition and metabolism of the intestinal microbiota may result in a more efficient immune system in healthy animals and may improve conditions of animals suffering from specific gastrointestinal, hepatic and renal diseases.

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ANIMAL BREEDING AND GENOMIC – GENOMIC EDITING

IS016

Selection and conservation of *Apis mellifera* at the time of globalization

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Despite beekeeping in Italy is widely considered one of the most advanced in Europe from the management techniques point of view, as regards the genetics and conservation aspects is far away from an optimal setting. Selection is based only on the maternal side and breeding of the virgin queens (VQ) is almost totally left to uncontrolled field mating. Italy is the home country of a particular variety of Apis mellifera, well known all over the world for a number of excellent characteristics, the Ligustica variety. Ligustica and Carnica (or Carniolan) bees belong to a same cluster spread toward Europe after the end of the last Glacial Era. Ligustica is well adapted to the mild climate of the Peninsula, while Carnica has its homeland on the North-Eastern side of the Alps and is widely bred on the Alpine Arch. In Sicily, there are still some examples of the original Sicula breed close to the Intermissa subspecies, present in all the South side of the Mediterranean sea. Before the introduction of the nomadism practice, the conservation of the three breeds present in Italy did not represent a problem since their geographic distribution had limited overlapping areas.

Due to its excellent numbers, the Ligustica strain was exported all over the world since the XIX century and well selected abroad. An important example is the Buckfast Abbey Bee, selected and improved in UK by Brother Adam during the XX century. Brother Adam included in this Buckfast honeybee the best traits of Ligustica, Carnica, Caucasic, Anatolian and several other geographic subspecies using empirical but well focussed techniques of gene introgression. In the meantime, in the former area of the Austro-Hungarian Empire, Carnica breed was selected by modern additive breeding techniques based on the control of mating in numerous isolated stations and, in more recent years, by BLUP genetic evaluation.

Now, in a rapidly globalising world, the diffusion of new breeds is fostered by superior performances in terms of honey production, resistance to pathogens and parasites, docility, reduced swarming attitude and other important management traits. In this context we are presently facing in Italy a progressive diffusion outside the Alpine area of Carnica breed. More remarkably, in an overall yearly market of about 700,000 queens, more than 100,000 are Buckfast that spread rapidly for their superior characteristics. It is therefore easily predictable that drones produced by these queens mating with VQ of the original geographic autochthonous breeds will determine a progressive genetic erosion of our national germplasm due to the lack in Italy of a proper culture of mating control. In honeybees, selection and genetic conservation of local breeds, in addition to the possible development of new biodiversity, would take enormous advantage from the use of isolated mating stations. In the Alps and in some districts of the Apennine chain is quite possible to find small secluded valleys or highlands where beekeeping is absent due to the lack of sufficient pasture. The access to these areas should also be limited to the beekeepers involved.

In the last years, thanks to the BEENOMIX project funded by Regione Lombardia, we have developed a model for the use of these isolated stations (IS) both for selection and for conservation. The general idea is that a breeder or a small group of associated breeders should develop a selection scheme in which a number (e.g. 100) of colonies are phenotyped, possibly genetically evaluated, and the best 8/10 selected. From the top best family 15 drone-producing queens (DPQ) are raised and placed in a ‘private’ IS to saturate the area with top selected drones. From the other best selected families (queen-producing queens, QPQ) a new cohort of 100 VQ is raised and taken to the station for breeding. The scheme can be implemented by any professional breeder, on any breed or strain, on any selection objective. By the use of 15 sister DPQs that represent a dummy father of future bees, a pedigree file can be registered providing an additive relationship matrix for BLUP indexing of the animal.

An IS is thought just to maximise the response to selection of that breeder and to provide improved animals both by maternal and paternal path. For the production of commercial queens, a different type of isolated station is necessary: a breeding area

(BA). A BA is thought to provide drones of a particular breed or strain, to breed a large number of commercial queens. Drones may come from many DPQ, eventually certificated for their breed origin, non-necessarily sisters, enough in number to guarantee the natural mating of even thousands VQ. The DPQ should be provided by several breeders selecting that same breed and the BA should be accessible to single beekeepers willing to breed their VQ to those specific drones. Commercial beekeepers should renew their queens buying these marked queens mated in a BA, to minimise the number of VQ available for free random mating. Nomadism and breeding of different breeds in a same area became therefore possible without risk of genetic pollution while conservation of any local breed is assured by selection in IS and diffusion thorough pure queens mated in BA.

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – REPRODUCTION

IS017

Endocrine and physiological differences between bovine and buffalo females and their impact on assisted reproductive technologies

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The combined use of assisted reproductive technologies (ART), such as, timed-artificial insemination (TAI), superstimulation (SOV), ovum pick-up (OPU), in vitro embryo production (IVEP) and timed-embryo transfer (TET) has a great potential to improve reproductive outcomes and disseminate selected genetics, improving milk and beef production in bovine an in buffalo herds. Nowadays, our expanding knowledge regarding endocrine and physiological differences between bovine and buffalo ovarian function during the oestrous cycle has given new approaches for the precise synchronisation of follicular development and ovulation to apply consistently ART. Unlike bovine, buffalo is a seasonal reproductive species and becomes sexually active in response to a decreasing day length (short-days) in late summer to early autumn. However, in recent decades several therapies have been proposed for manipulating ovarian follicle growth and ovulation, regardless of reproductive seasonality or cyclicity in buffalo and bovine. Ovarian follicular dynamics in buffalo are similar to those in bovine. The 2-wave cycle is the most common and the follicle deviation occurs 2–3 days after ovulation. In cattle and in buffalo the number of waves in a cycle is also associated with the luteal phase and with the oestrous cycle length. However, the number of follicles recruited per follicular wave is lower in buffalo than in cattle. Artificial insemination (AI) has proven to be a reliable technology for producers to improve genetic progress and control venereal diseases in their herds. However, the traditional AI programme is impaired by the low oestrous detection efficiency due to the poor manifestation of the symptoms of oestrus and to operational difficulties to detect oestrus in bovine and in buffalo. Furthermore, buffalo presents lower frequency of homosexual behaviour compared to cattle, affecting more negatively the oestrus detection efficiency. Recent synchronisation protocols are designed to control both luteal and follicular function and permit fixed-time AI with high pregnancy rates regardless the cyclicity in bovine and during the breeding (autumn-winter) and nonbreeding (spring-summer) seasons in buffalo. Superstimulation for *in vivo* embryo production and ovum-pick-up (OPU) for in vitro embryo production (IVEP) can rapidly enhance genetics through both the female and male superior lineage. Unlike bovine, the *in vivo*-derived (IVD) embryo production has been shown to be feasible in buffalo and low efficiency and limited commercial application has been documented in this species. However, a series of recent studies have demonstrated the potential of *in vitro* embryo production (IVP) in buffalo, similar to that observed in cattle. Additionally, synchronisation protocols provide opportunities to fixed-time embryo transfer in bovine and buffalo recipients, facilitating the process at the farm and increasing the efficiency of ET. These endocrinology and physiology differences between bovine and buffalo reveal a number of key points related to the manipulation of ovarian follicular growth which must be taken into account to improve the efficiency of assisted reproductive technologies in these species.

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – ANIMAL WELFARE

IS018

Animal welfare assessment: towards a simplification

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The aim of the present study is the identification of current trends in the field of on-farm animal welfare assessment. The Animal Needs Index was the first on-farm welfare monitoring scheme developed in Austria and Germany in the eighties with the objective to provide data on the level of animal welfare of the farms claiming organic certification, when a specific legislation on organic farming was still missing at European level. That scheme, albeit feasible and reliable within and between observers, mainly relied on design characteristics and was not scientifically validated neither at individual indicator nor at full scheme levels. This gap was tackled by the Welfare Quality project which increased the knowledge in the field, by providing a framework for the assessment of animal welfare for all farmed species, and produced a high amount of data for the validation of each indicator included in the scheme. However, this scheme proved to be time consuming (more than forty indicators to be recorded) and unable to discriminate among farms, as the majority is classified in the two intermediate welfare categories, and to provide a balanced summary of the general welfare conditions at farm level, as one indicator (absence of prolonged thirst, which is a resource-based measure) drives the overall farm classification. As a consequence, the Welfare Quality protocol has been seldom used in practice for certification purposes. A first effort to simplify the assessment was done in the project AWIN where in terms of welfare monitoring a two-stage approach increased the feasibility of the scheme (the first stage consists of a quick farm screening, which requires minimal animal handling; additionally, the number of indicators is roughly reduced by half as compared with the Welfare Quality scheme), whereas in terms of output no integration of individual indicators is needed and benchmarking is the main tool used to promote a continuous improvement of animal welfare. A further step towards simplification was taken in more recent studies conducted in Australia on extensively managed ewes where only ten scientifically validated animal-based measures were selected on the basis of extensive literature review and consultations with experts in the field of sheep welfare. This number was further reduced to six (body condition, fleece condition, skin lesions, tail length, dag score, lameness) due to lack of feasibility and/or reliability (rumen fill, hoof overgrowth, footwall integrity) and inappropriateness to extensive conditions (cleanliness). As also observed for the AWIN scheme, in this case study no indicator aggregation was applied, thus no overall farm classification could be achieved. With the aim to obtain a farm classification a simplified one-step aggregation system of single indicators has been recently proposed, which is based on expert opinion to establish the severity of a welfare issue for an individual animal to be then multiplied by the prevalence of that welfare issue in the farm. In the field of risk assessment, a simplified approach was used by the EFSA working group on sheep welfare which ranked seventeen welfare consequences across six different management systems through the elicitation of expert knowledge and associated the three to five most relevant to the related risk factors. However, in Italy an opposite trend can be observed as, following a holistic approach, the Italian National Animal Welfare Reference Centre issued the only currently applied welfare monitoring schemes at national level relying on a high number of indicators (more than eighty for dairy cattle) covering both biosecurity and welfare with Management, Housing and Animal-based measures as the three main aspects to be assessed. Additional aspects to be evaluated in order to get the welfare certification 'Classyfarm' include feeding, consumption of antimicrobial agents, lesions identified at the slaughter house, health and productive parameters. In conclusion, while at international level on-farm welfare monitoring is moving towards schemes with a limited number of indicators and simplified aggregation systems, in Italy an opposite approach is pursued where animal welfare per se is not the only focus and several other aspects, albeit relevant and related to animal welfare, are also covered with the aim to address a wide range of issues currently faced by the animal production sector.

ORAL PRESENTATIONS

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – ANIMAL PHYSIOLOGY

O001

Determination of blood metabolites in early lactation dairy cows using milk mid-infrared spectra

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During early lactation, dairy cows normally experience an unbalanced energy status that can lead to the occurrence of several metabolic disorders. Blood metabolic profile is a valid tool to monitor and identify metabolic diseases, but blood sampling and analysis is a time-consuming and expensive procedure, being also stressful for the animals. Mid-infrared (MIR) spectroscopy is routinely implemented for milk composition analysis of cow milk, being a cost-effective and non-destructive method. Thus, the aim of this study was to investigate the feasibility of predicting blood metabolites from milk MIR spectra. To achieve this goal, 20 herds rearing Holstein-Friesian, Brown Swiss or Simmental dairy cows, located in Trentino Alto Adige and Veneto regions, have been visited between December 2017 and June 2018. At each visit, blood and milk samples were collected within one hour from all lactating cows within 35 days in milk. Blood samples were analysed through reference procedures and milk MIR spectra were collected during milk analyses. Backward interval partial least squares (BiPLS) algorithm was applied to build prediction models for considered metabolic traits.

Results showed that BiPLS improved the predictive ability of the models for the studied traits compared with traditional PLS analysis. Blood β-hydroxybutyrate, urea, non-esterified fatty acids and cholesterol were the most predictable traits, with coefficients of determination in external validation of 0.71, 0.64, 0.55 and 0.45, respectively. On the other hand, prediction models for other analysed metabolites were not enough accurate for routine analysis or population studies. Results of the present study suggest the potential of milk MIR spectra to predict important blood metabolites, leading to the possibility to easily access to metabolic status information of early lactation cows.

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O002

Effect of the milk yield amount at the end of lactation on inflammation, oxidative stress, and metabolic changes during dry-off stage of dairy cow

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Deep changes in feeding behaviour, gastrointestinal tract, metabolism and immune parameters occurs in high-yielding cows at dry-off. Indeed, the release of cortisol, systemic inflammations and altered redox balance have been reported at milking interruption, and high milk yield (MY) aggravate such conditions.

Our study investigates the causes of metabolic changes occurring at dry-off and the contribution of MY in such alterations. A group of 13 Holstein dairy cows were dried off at 55 days from expected calving day. Concentrates were gradually reduced in the last week of lactation and eliminated at dry-off. Then cows were fed only hay for 10 days and a hay-based ration with soybean meal and corn silage thereafter. The cows were divided into two groups according to their MY in the last week of lactation, assuming a cut-off of 15 kg*d-1: low MY (6 cows) and high MY (7 cows). From –7 to 34 days from dry-off (DFD) feed intake and rumination time were measured. Blood samples were collected regularly to assess a wide hematochemical profile and to test white blood cell functions with a whole blood stimulation test with LPS (WBS). Data were submitted to ANOVA using the MIXED procedure (SAS Inst.), including MY at dry-off, time and their interaction as fixed effects in the model.

Increased fibre amounts of dry ration reduced DMI and increased rumination time ($p<.05$). Leukocytes migration into mammary gland to contribute in the involution phase decreased their abundance in blood at dry-off ($p<.05$). Moreover, the production of IL1 beta and IL 6 following the WBS increased after the dry off. Such activation of leukocytes at mammary site increased the abundance of nitrogen species in plasma ($p<.05$) and triggered a systemic inflammation in all the cows (higher positive and lower negative acute phase proteins). Inflammation impaired liver functions (higher gamma-glutamyl transferase, bilirubin and alkaline phosphatase; $p<.05$). After dry-off, the antioxidant systems were depleted from blood (lower thiol groups, tocopherol, β-carotene, ferric reducing antioxidant power and oxygen reactive antioxidant capacity). Cows with higher MY at dry-off showed the worst condition, likely for the deeper metabolic changes at milking interruption, and for greater amounts of mammary parenchyma to be reabsorbed. This study highlights the dry-off as a critical point for dairy cows' health and suggests for a relationship between dry-off and immune alteration that occurs at calving.

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O003

Effect of heat stress on monocytes and lymphocytes in dairy cattle

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Heat stress has a negative impact on dairy cows' health, milk production, reproductive performance and immune status. Cellular and molecular responses to high temperatures have been described in bovine polymorphonuclear cells and peripheral blood mononuclear cells (PBMCs). However, few studies on the effect of high temperatures on specific immune cell populations, like lymphocytes and especially monocytes, have been carried out. Therefore, here we sought to determine the effects *in-vitro* of high temperatures, simulating severe whole-body hyperthermia, on the immune function of monocytes and lymphocytes in dairy cattle. First, PBMCs from nine healthy animals were isolated from whole blood samples. Monocytes were further purified by positive selection with CD14+ antibody. After isolation, monocytes and lymphocytes were cultured overnight at 39 °C and 41 °C. The two temperatures were intended to mimic conditions of normothermia and hyperthermia, respectively. Apoptotic rate, reactive oxygen species (ROS) production, viability and expression of few cytokines were assessed. We found that the viability was higher in monocytes and lymphocytes incubated at 39 °C than 41 °C (p=.046 and p=.0039, respectively). In addition, we observed that apoptosis increased in monocytes and lymphocytes exposed to 41 °C, as compared to the control (p<.001). Conversely, no difference in ROS production was observed. In conclusion, the results obtained in this study suggest that the exposure to high temperatures affects viability and apoptosis, confirming that immune cells response is modified during hyperthermia. Further studies are needed to support these results and to ascertain whether the effects of high temperatures on immune cells are responsible for higher susceptibility of heat-stressed cows to infections.

O004

Study of inflammatory markers in dairy cows exposed to grain-rich diet

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Grain-rich diets are commonly fed to cows in modern dairy herds, exposing cows to the risk of development of Subclinical Acidosis (SARA). This research aims to explore the effects of SARA on inflammatory status of animals. For this purpose, 3 groups of 8 multiparous cows each, enrolled around 60 DIM, were housed in the experimental tie stall barn of the University and fed an acidogenic diet for 4 weeks. This diet had a forage: concentrate ratio of 25:75, while the pre-challenge diet had a F:C ratio of 45:55. During this period, individual dry matter and water intake and rumen pH were recorded continuously and blood was sampled 8 times: the first time before the beginning of the trial (day 0), 4 times in the 1st week to evaluate the acute response (day 1, 2, 3 and 7) and then at day 14, 21 and 28. On each sample, complete haematologic and biochemical profile were determined, together with IFN-γ, IL-1β and IL-6. Data were statistically analysed with a mixed model with time point, group and interactions as fixed effects and animal as random effect. Significant effects were then compared between time points with a t-student test.

As expected, rumen pH was reduced with the high concentrate diet: time with pH below 5.8 and 5.5 was significantly longer compared to the pre-challenge period (305 vs. 290 min/d < 5.8 and 53 vs. 34 min/d < 5.5). Blood analysis revealed an acute phase response of the organism following the acidogenic diet. In particular, IL-6 increased significantly after 7 days of challenge (+473 pg/mL, p<.05), as well as serum amyloid A and ceruloplasmin that, compared to T0, reached their maximum after 2 and 3 weeks respectively (+125.15 µg/mL and +0.62 µmol/L, p<.05). Accordingly, albumin decreased constantly after the intake of the experimental diet getting the minimum after 4 weeks of trial indicating a status of chronic inflammation (−1.69 g/L, p<.01). Reactive Oxygen Metabolites (ROMs) increased after 7 days of acidotic diet and remained high until the end of the trial, with a peak after 3 weeks (+3.53 mg H2O2/100 mL, p<.01). Ferric Reducing Ability of Plasma (FRAP) begins to decrease after 2 weeks of trial and reached the minimum after 21 days (−18.44 µmol/L, p<.01), confirming together with ROMs levels, the presence of oxidative stress in these animals. These results show that an excessive content of concentrates in the diet can trigger an inflammatory response that could have important consequences on the health of high producing or fresh animals.

O005

Starch exposure early in life to enhance metabolic programming of nutrient partitioning in lactating dairy sheep

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This work aimed at testing if sequences of glucogenic (from starch; S) and lipogenic (from high digestible fibre; F) diets, in foetal life, growing and mid-lactation, affect milk persistency of dairy sheep. Performance of 72 sheep from birth to first dry-off was described. A split-plot experimental design has been adopted. The 72 sheep were divided into 8 groups identified by a sequence of 3 letters: SSS, SSF, FSS, FSF, SFS, SFF, FFS and FFF. They indicated the diets received in foetal life (last 75 days), in growing and in mid-lactation (56 to 165 days in milk, DIM), respectively. Body weight, body condition score, feed intake and several blood parameters including plasma glucose and insulin concentrations were monitored. Glucose (GTT) and insulin (ITT) tolerance tests were also executed. Gene expression of adiponectin in subcutaneous fat from biopsies was determined in late lactation. At 55 DIM, fat and protein corrected milk (FPMY) was 1.59 ± 0.10 kg/d per ewe. Prenatal diet affected persistency: FPMY decay from 56 to 165 DIM was −0.728 vs. −0.992 kg/d in ewes exposed to S or F in foetal life, respectively (p=.04). FPMY persistency was greater (p<.05) in ewes fed SSF, exposed to starchy diets in foetal life and from weaning to first lambing (SS), and fed diet rich in high digestible fibre in mid-lactation (F). SSF and FFF showed the greatest and lowest FPMY after 56 DIM (1.483 ± 0.180 vs. 1.143 ± 0.198 kg/d, respectively; p<.05). FPMY was different (p<.05) among groups only when considering ewes with homogenous lactation stage (lambing the same decade). SS vs. FF diet sequence was associated to greatest levels of basal insulin during GTT in pregnancy and ITT in lactation ($p<.05$), to lower glucose uptakes in ITT ($p=.06$) and to greater values of the homeostasis model assessment index in lactation. Relative abundance of adiponectin gene expression was affected by prenatal and growing diets (2 times greater in FF vs. SS; $p<.05$) and by lactation diet (1.5 times greater in F vs. S; $p<.05$). These metabolic evidences are often associated with insulin resistance (IR). We concluded that exposure to glucogenic diets in foetal and growing phase (SS) might programme a default IR that could extend the homeorhetic IR of early lactation. After lactation peak, IR could affect milk persistency reducing nutrient uptake by body reserve and stimulating the nutrient utilisation by mammary gland.

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ANIMAL BREEDING AND GENOMICS – GENETICS OF WELFARE, GENETICS OF RESILIENCE

O006

Cheesemaking and sustainability index – Parmigiano Reggiano: a new breeding index for the Italian Holstein

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The Italian dairy industry is specialised in the manufacturing of high quality, long ripened cheese. The objective of the present study was to develop a new breeding index (ICS-PR) for Holstein cattle producing milk for long ripened cheeses such as Parmigiano Reggiano PDO; the final breeding goal is to maximise net profit per day of life. Net profit for commercially available AI bulls was calculated as the difference between total estimable revenue and the total estimable costs using EBV released with the official genetic evaluations. Costs included heifer rearing, cow feeding for maintenance and production, fertility, udder health, calving difficulty and milk delivery whereby revenue considered cream, cheese and whey production, as well as, culled cow value. Dry matter intake and cheese yield were estimated using available prediction formulae; economic values for both costs and revenue were retrieved from the literature. Stepwise multiple linear regression was employed to develop a prediction equation for net profit from official AI bulls EBV having rank >59 and reliability >50. The 10-year genetic gain achievable for official EBV with selection for ICS-PR was calculated from P and G matrixes constructed using (co)variances routinely used for BLUP genetic evaluation and it was compared to the 10-year genetic gain achievable with selection for PFT index. EBVs retained (relative emphasis) from the multiple linear regression were: kg of protein (30%) and fat (5%), stature (−2%), locomotion (3%), udder depth (2%), somatic cell score (14%), calving ease (9%), longevity (10%), mastitis resistance (5%), and fertility (20%). Moreover, results demonstrated that, although the gain in production traits with ICS-PR was 85–96% the potential gain achievable with PFT, the selection for ICS-PR would result in a much faster genetic gain for welfare characteristics, such as calving ease (22%), longevity (22%), udder health (18%), while the gain for fertility was three times larger. In 10 years' time, bovine production and productive life genetically will increase at a different rate depending on breeding decisions. However, in a scenario of selection for ICS-PR, cow lifetime production of Parmigiano Reggiano, calculated applying cheese yield prediction formulae used in the present study, is expected to be almost 100 kg higher compared to that achievable with selection for PFT.

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O007

Structural equation models for genome-wide association study (SEM-GWAS) of interrelationships among udder health traits in dairy cattle

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The quality and safety of milk products depend on animal health and welfare, and particularly on mammary gland health. It is well known that mastitis, either in its acute or sub-clinical form, greatly affects the nutritional and technological quality of milk. Apart from somatic cell count (SCC), milk lactose (LAC) content, pH and casein to protein ratio (CAS:PRT) are sensitive to inflammation of the mammary gland, and these variables could be incorporated together into an udder health indicator. Multi-trait model genome-wide association (MTM-GWAS) can be used to study associations between genomic regions and multiple traits, but it does not consider potential causal relationships among phenotypes. Alternatively, structural equation modelling (SEM) represents a powerful tool for modelling causal networks. Recently, SEM in combination with GWAS (SEM-GWAS) has been proposed to better understand the genetic basis and the relationships among a set of traits. SEM-GWAS can partition SNP effects into direct and indirect (i.e. mediated by an up-stream trait in the causal network) components. This study aimed to apply SEM-GWAS on a set of phenotypes related to udder health, i.e. milk yield (MY), somatic cell score (SCS), LAC, pH and CAS:PRT, in a cohort of 1158 Italian Brown Swiss cows. Animals were genotyped with the Illumina BovineSNP50 Bead Chip v.2. A causal phenotypic network was inferred in two stages: (1) a multi-trait model was employed to estimate covariance matrices of additive genetic effects and of residuals, and (2) the causal structure among phenotypes from the covariance matrix between traits, conditionally on additive genetic effects, was inferred by the Hill-Climbing algorithm. The residual (co)variance matrix was inferred using Bayesian Markov-chain Monte Carlo, with samples drawn from the posterior distribution. Results showed positive path coefficients for MY → LACT, LACT → CAS:PRT and SCS → pH, while negative values were obtained for LACT → SCS. Based on the identified phenotypic relationships structures, SEM-GWAS will be run, enabling to identify direct and indirect (i.e. mediated) SNP effects and providing a more complete picture of the genetic basis of these indicators of udder health.

Acknowledgements

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O008

Reaction norm analyses to infer genotype by environment interaction for fertility traits in Italian dairy cattle

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Different environments can contribute to differential expression of polygenic effects in dairy cows, which can lead to re-ranking of sires for economically important traits across environments. This genotype by environment interaction (G x E) may need to be taken into account for environment-specific breeding programmes. In this study, G x E was inferred using reaction norm models in Brown Swiss and Simmental breeds on the fertility traits calving interval (CI), interval from calving to first service, interval from first service to conception, days open, calving rate and non-return rate at day 56. Data was used from the Alto Adige province database, in the north of Italy. Herd-year specific daily milk energy production levels (dMEO, kg/day), were used to define the environmental gradient in terms of production management. Only animals with pedigree records and sires with at least 2 daughters across the environmental gradient were used. Analyses were carried out by software programmes AIREMLF90 and THRGIBBSF90. The animal model included parity number, year season and herd as non-genetic effects. Heterogeneous residual variances were assumed across the environmental gradient. For Brown Swiss breed and fertility trait CI, genetic parameter estimates showed a higher genetic variation between low (30–60 kg/day) and high levels of dMEO (90–110 kg/day) for genetic variance and heritability values, and small variation between low and medium (70–90 kg/day) levels of dMEO. Residual variances decreased across the environmental gradient from low to high levels of dMEO. Heritability estimates across the environmental gradient obtained ranged between 0.047 to 0.053 at low and medium levels of dMEO and increased at high levels of dMEO from 0.060 to 0.100 (standard deviation of 0.019). Genetic correlation across the environmental gradient ranged from 0.99 to 0.80 at low-medium levels of dMEO and between low-low, low-medium, medium-medium and medium-high levels of dMEO. However, genetic correlation decreased down to 0.60 between low and high levels of dMEO. Sire estimated breeding values re-ranked across the environmental gradient and correlations of those breeding values between environments ranged from 0.99 to 0.90. In conclusion, we observed G x E on fertility traits according to the energy level and this should be taken into account for breeding programmes.

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O009

Cut-off values and genetic aspects of differential somatic cell count in dairy cows

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The knowledge of the proportion of the different cell types in milk, rather than just the overall somatic cell count (SCC), could provide valuable information for a more precise definition of the health status of the udder. Recent developments in milk-testing technology have allowed cell differentiation in milk in a high throughput manner using flow cytometry. In the present study, we defined cut-off values of a novel indicator of udder health, i.e. the milk differential somatic cell count (DSCC), to identify subclinical mastitis. This novel trait represents the percentage of neutrophils plus lymphocytes in the total SCC. In addition, we investigated the genetic background of DSCC to gain knowledge on its potential inclusion in selection programmes aimed to improve mastitis resistance in dairy cattle. A total of 21,043 test-day records of 10,106 Italian Holstein cows from 406 herds were considered for ROC analysis, whereas a subset including 10,709 test-day records of 5142 cows from 299 herds was used for genetic analysis. The ROC procedure allowed the identification of the best DSCC thresholds (given by the highest combination of sensitivity and specificity) among all the possible cut-off values. Different thresholds were obtained for primiparous and multiparous cows, in early or mid-late lactation. In addition, our results revealed, for the first time, that DSCC is a heritable trait (h^2=0.08 ± 0.02), and heritability was lower in primiparous than in multiparous cows. Furthermore, bivariate analysis demonstrated that DSCC and traditional somatic cell score (SCS) are two different traits as their genetic correlation was less than one. This study provided DSCC cut-off values for a practical application of this novel trait to identify subclinical mastitis and findings on the genetic aspects of DSCC are a prelude to its potential use in breeding programmes aimed to select against mastitis occurrence.

Acknowledgements

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O010

Setting up genetic evaluation for feed efficiency in Italian Holsteins

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Feed cost is half of the total cost of dairy production. One possibility to increase the profitability of dairy production is to reduce feed cost by improving feed efficiency. Selection of animals with high feed efficiency can bring benefits in terms of productivity and environmental impact. Aim of this project is to set up a genetic evaluation for feed efficiency in Italian Holsteins. Currently, it is unfeasible to work on direct data collection for dry matter intake and therefore feed efficiency, however, it is possible to derive traits thanks to the availability of data from the national milk recording system. For this preliminary study feed efficiency (PFE_ECM) was estimated as a ratio between Energy Corrected Milk (ECM) and predicted dry matter intake (pDMI). The PFE_ECM, ECM and pDMI were on average equal to 1.27 ± 0.18 kg/d, 29.65 ± 7.70 kg/d and 23.30 ± 3.14 kg/d. All traits showed the same pattern through lactation, increasing in the first 3 months of lactation and then decreasing. A test-day repeatability model was applied. Fixed effects were the herd-test-date (HTD), and the interaction parity*stage of lactation. Random effects included the cow within and across lactation, the animal and the error. Parameter estimation has been undertaken using a data subset extracting randomly in the Italian Holstein population 300 herds, this procedure was repeated 5 times. Editing resulted in cows with lactation stage between 5 and 365 days in milk (DIM), with at least 2 test-day records and belonging to HTD classes with a minimum of 3 contemporary animals. Sires were required to have at least 5 daughters in 3 herds. Final data-set consisted of 632,840 repeated records from 39,574 cows and 1434 sires. Pedigree (76,268 animals) included individuals with records and their ancestors up to 6 generations back. Heritabilities of PFE_ECM, ECM and pDMI were equal to 0.103 ± 0.006, 0.096 ± 0.006 and 0.100 ± 0.006 respectively. Genetic parameters have been applied to the entire test-day data-set in order to estimate breeding values for PFE. In conclusion, several issues are discussed; for example, the lack of real DMI measurements (and the key assumption that body condition score is not affected by feed efficiency level), as

well as, the limitations of predicting the biological consequences of selection for feed efficiency. To overcome these limitations, an international effort is under discussion to bring together data on feed intake directly measured in dairy cows.

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LIVESTOCK SYSTEMS – AQUACULTURE

O011

Effects of nucleotide supplementation in diets high in soybean meal on growth, gut health and immune status of E. sea bass (*D. labrax*)

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Soybean meal (SBM), a widely used feed protein source, is known to impair gut health and functions in certain fish species when included at high levels in aquafeeds. Previous studies have shown that dietary addition of exogenous nucleotides (NT) could improve fish growth, gut integrity and immune response. In this context, a feeding trial was carried out in European sea bass to evaluate possible beneficial effects of a dietary NT supplementation to a diet rich in SBM. Three isoproteic (45.6% DM) and isolipidic (17% DM) diets were formulated: control (C), high in fish meal and oil and low in SBM (150 g/kg); diet rich in SBM (350 g/kg) and nucleotide-supplemented diet (N) obtained from the SBM one by adding 0.2 g/kg dry diet of a commercial source of nucleotides (Vannagen®). 108 fish (mean b.w. 204 ± 12.7 g) were divided into nine 300-L tanks included in a marine recirculating aquaculture system. They were randomly allotted in triplicate to the 3 diets offered to visual satiety over 105 days. At the end of the trial, the growth performance was recorded per group. Blood samples from 10 fish per diet were analysed for serum total protein (TP) level, lysozyme, antiprotease and peroxidase (PO) activity; six fish per diet were evaluated for respiratory burst (RB) and myeloperoxidase (MPO) activity of head kidney leucocytes. The digestive tract was processed for histology and analysed for intestinal brush border enzyme (BBE) gene expression and activity.
Growth response was unaffected by dietary treatments. Fish fed SBM and N diets showed higher TP relative to controls ($p<.05$). No diet effect was detected on lysozyme, PO and MPO activities whereas N diet induced the highest antiprotease activity ($p<.05$). The RB was depressed in fish fed the SBM diet compared to fish fed diets C and N ($p<.05$). In the posterior intestine, N diet resulted in improved BBE activities relative to the SBM one ($p<.05$). Irrespective of the intestinal tract, BBE expression was upregulated in fish fed N diet ($p<.05$) when compared to fish fed both C and SBM diets. Fish fed diets SBM and N showed a greater *villi* height compared to those given diet C ($p<.05$), while the thickness of intestinal epithelium was increased only in fish fed N diet. The present study demonstrated that supplementing SBM-rich diet with 0.2 g/kg NT does not affect growth performance of sea bass, leads to improved gut health and digestive-absorptive functions and just barely affects certain innate immune parameters.

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O012

Nutritive value of novel feed protein sources to rainbow trout (*O. mykiss*) and European sea bass (*D. labrax*)

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The dried biomass of certain microalgae and the meals obtained from arthropods like insects are deserving a growing interest as potential sources of protein in aquafeeds but very little information still exists on their actual nutritive value to fish. The present study investigated the nutrient composition and the *in vivo* nutrient digestibility in rainbow trout (*O. mykiss*) and sea bass (*D. labrax*), of a panel of three microalgae dry biomass (*Arthrospira platensis*, S, *Tetraselmis suecica*, Te, *Tisochrysis lutea*, Ti), two *Hermetia illucens* larval meals (HM1, 2) and of a meal obtained by processing whole Louisiana red swamp crayfish (RCM). The proximate, amino acid and fatty acid compositions of the test ingredients were determined with standard methods. Their nutrient and energy apparent digestibility coefficients (ADCs) were estimated with the indirect method and by difference relative to those of a reference diet. Six test diets were obtained by mixing each test ingredient and the reference diet at a 30:70 (w:w) ratio. All diets were added with acid insoluble ash (1500 mg/kg) and Yttrium oxide (100 mg/kg) as inert markers before being extruded into 3 mm pellets. The ADCs of macronutrients and energy of the reference and test diets were measured in juvenile rainbow trout (47 ± 1.5 g) and sea bass (37 ± 1.0 g) kept in tank-units each composed by three 65-L vessels stocked with 45 fish and fitted with a settling column for faecal collection. Each diet, offered to visual satiety in two daily meals, was evaluated over three independent 10-day faecal collection periods.

Despite obvious differences in crude protein content (CP) among test ingredients (range 36–53% as fed), they could be considered valuable sources of feed protein although, apart from RCM, they were limiting in sulphur amino acids relative to fish requirements. The macronutrient and energy ADCs were unaffected by the marker and resulted systematically lower in sea bass relative to those of trout ($p<.05$). Extremely high CP and energy ADCs were estimated for RCM (>98%, $p<.05$) and the lowest in case of Te (<82% and <66% for CP and energy respectively, $p<.05$) with the remaining ingredients having intermediate values.

The results of this study provide a first data set of the nutritive value of novel feed protein sources to two important fish species for the European aquaculture.

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O013

Monoglycerides of short- and medium-chain fatty acids: effects on growth performances and gut microbiota of gilthead sea bream (*Sparus aurata*)

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Fish gut microbiota is involved in the anaerobic fermentation of otherwise indigestible dietary components. The bacterial metabolism of these substrates yields bioactive metabolites, such as the short-chain fatty acids (SCFAs). Being organic acids, SCFAs decrease the pH in the gut and are therefore believed to prevent the overgrowth of pH-sensitive pathogenic bacteria. Because SCFAs are also antimicrobials, they may affect commensal microbiota.

Accordingly, the present study aimed to evaluate the potential beneficial effects of SCFA monoglycerides used as a feed additive on fish growth performances, and intestinal microbiota composition. For this purpose, a specific combination of short and medium chain 1-monoglycerides (SILOhealth108) was tested in juvenile gilthead sea bream (*Sparus aurata*) fed a plant-based diet. Six hundred fish of about 60 g mean initial weight were fed for 3 months with two different feed formulations. The control fish group received a plant-based diet without any form of butyrate supplementation, whereas the other group received the control feed supplemented with 0.5% of SILOhealth108. The Illumina MiSeq platform for high-throughput amplicon sequencing of 16S rRNA gene, and QIIME pipeline were used to analyse and characterise the whole microbiome associated with both, feeds and *S. aurata* intestine. The number of reads taxonomically classified according to the Greengenes database was 394,611. We identified 259 OTUs at 97% identity in sea bream faecal samples; 105 OTUs constituted the core gut microbiota, i.e. those OTUs found in at least 80% of the samples per dietary group and shared irrespective of the diet. The dominant phyla in both experimental groups were represented by Firmicutes, Proteobacteria and Actinobacteria.

In summary, our findings clearly indicated that SILOhealth108 positively modulated the fish intestinal microbiota. The specific composition of 1-monoglycerides of short and medium chain fatty acid contained in SILOhealth108 could have thus a great potential as feed additive in aquaculture.

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O014

Interaction between dietary composition and seasonal temperature changes in gilthead sea bream (*Sparus aurata*, L. 1758): effects on growth, fat deposition and plasma biochemistry

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The optimisation of feeding strategy in relation to the environmental condition needs further investigation in order to maximise performance, fish quality and fish health of Mediterranean farmed species. For this reason, the purpose of this study was to evaluate the effect of temperature and dietary protein and dietary energy ratio (DP/DE) changes on growth, feed efficiency, fat deposition in liver and intestine, somatometric indexes and plasma biochemistry of Gilthead sea bream (*Sparus aurata*) during seasonal changes. Two different extruded diets with high and low DP/DE ratio were tested in triplicated fish groups of 30 individuals (initial weight: 67.5 g) and raised at two different temperatures (17 and 23 °C) in the same recirculation system over 115 days. Fish were fed manually to visual satiation twice a day. After 58 days fish were exposed to a switch in temperature (fish kept at 17 °C were transferred to 23 °C and the fish kept at 23 °C were transferred to 17 °C) while continued to receive the same diet in each group. Specific growth rate (SGR), voluntary feed intake (VFI), feed conversion rate (FCR), viscerosomatic index (VSI), hepatosomatic index (HSI), condition factor (CF), carcase composition and plasma biochemistry were determined in the intermediate and in the final sampling. Fish exposed to low water temperature in the first period showed a reduced ($p<.05$) SGR, VFI and an increased FCR under both dietary regimes in comparison to the groups exposed to high temperature, while HSI was higher at low water temperature. No evident dietary effect on growth and fat deposition was detected, however, low DP/DE slightly improved performance of fish firstly exposed to low temperature. This study provided novel insight on the effects of DP/DE ratio on fat deposition and physiological parameters of gilthead sea bream fed in summer before entering winter and fed in winter before entering in spring.

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O015

Effects of different feeding frequencies on growth, feed utilisation, plasma biochemistry and digestive conditions of gilthead sea bream (*Sparus aurata*) fed with different fish meal/fish oil dietary levels

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The optimisation of feeding management is essential to improve the Key Performance Indicators (growth rates, mortality, quality traits and feed efficiency) of the Mediterranean aquaculture. This study was undertaken in order to assess the effects of different feeding frequencies on growth, feed digestibility, somatometric indexes, fish plasma biochemistry, gut histology and gut enzymes activity. Two isoproteic and isoenergetic extruded diets were formulated to contain high (HFM) and low (LFM) fishmeal (FM) and fish oil (FO) dietary level. Each diet was randomly assigned to triplicate fish groups of 60 individuals (initial weight: 88.3 g), each submitted to a different feeding frequency (1, 2 and 3 meals day-1) over 109 days. Specific growth rate (SGR), voluntary feed intake (VFI) and feed conversion rate (FCR) were determined. Furthermore, viscerosomatic index (VSI), hepatosomatic index (HSI), condition factor (CF) and fat index (FaI) were also calculated. Data were analysed by a two-way ANOVA followed by a Tukey's multiple comparison test. At the end of the trial, no significant differences ($p> .05$) due to feeding patterns were observed in terms of final body weight, SGR, VFI and FCR and no significant differences were found in VSI, HSI, CF and FaI values between different feeding frequencies. However, significant differences ($p<.05$) among diets were found. SGR and FaI values were slightly higher in HFM compared to LFM while FCR was higher in LFM. In conclusion, high FM/FO diet led to higher growth and feed utilisation in comparison to low FM/FO diet, and different feeding patterns did not seem to affect the overall growth performance and feed utilisation.

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – TANNINS IN ANIMAL NUTRITION

O016

Effect of vescalagin and gallic acid from chestnut tannins on microbial community profile: preliminary results

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The aim of the present study was to evaluate how the rumen community, the fatty (FA) and dimethylacetal (DMA) profile can be affected by vescalagin and gallic acid, the major components in chestnut tannins. Ewe rumen liquor (LR) was buffered with McDougall saliva and inoculated with four feeds (basal diet as negative control – diet C – formulated with tannin-free ingredients; basal diet supplemented with chestnut tannin extract as positive control – diet CHT – at the inclusion level of 1.6 g/100 g of dry matter (DM); basal diet supplemented with vescalagin – diet VES – at the inclusion level of 0.24 g/100 g of DM; basal diet supplemented with gallic acid – diet GAL – at the inclusion level of 0.032 g/100 g of DM) and fermented in batch for 24h. The formulation and the inclusion levels of the diets were the same as of a previous *in vivo* trial. At the end of the experiment, the rumen liquor was analysed to evaluate the potential effects of vescalagin and gallic acid on rumen microbiota, FA and DMA profile. Data for FAME and DMA determined by mass spectrometry and microbial relative abundance at genus level determined by high-throughput sequencing, were processed by GLM of SAS. The genera *Anaerovibrio*, *Bibersteinia*, *Escherichia/Shigella*, *Streptococcus* and the relative abundance of the genus *Pseudobutyrivibrio* increased with CHT compared to the other feeds while the genus *Arcobacter* decreased. Furthermore, a slight decrease in the relative abundance of the genus *Treponema* was observed in GAL fermenters compared to VES fermenters. Changes in microbial profile were not so substantial to induce variations in FA profile. In contrast, the presence of CHT extract, VES and GAL in the basal diet modified the concentration of several DMA. At 24 h DMA 12:0, DMA 13:0, DMA 14:0, DMA 15 iso, DMA 18:0 and DMA 18:1 trans11 were lower in C fermenters than in the others ($p \leq .05$). Results showed that VES and GAL did not exert singularly strong effects on microbiota. Hence, the antimicrobial efficiency of CHT may be due to the overall composition of the tannin extract.

O017

Effect of chestnut tannins and an extract from *Stevia Rebaudiana Bertoni* on *in vitro* rumen fermentation and microbiota

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Plant extracts can be used as safe dietary additives to improve nutritional efficiency and to reduce excretions of animals. The effects of dietary addition of chestnut tannins (T) and an extract from *Stevia Rebaudiana Bertoni* (S) on rumen fermentation parameters and microbiota were evaluated in three *in vitro* rumen fermentation experiments. The Experiment 1 and 3 batch culture systems (fermentation in 500 mL bottles for 24 or 48 h) were used while for Experiment 2 was used a continuous culture system (fermentation in 2 L bottles for 10 days). In Experiment 1 and 2 a control substrate (C) was added with 1.5% of T or S extracts, while Experiment 3 considered 7 doses of each extract added to C (additions of 0.2, 0.4, 0.6, 0.8, 1.0, 1.2 and 1.4 %) in separate runs performed for each extract.

The T addition lowered the *in vitro* rumen ammonia in all experiments: the reduction, in comparison to C, was 12 and 50% in the Experiment 1 and 2 ($p<.01$) and followed a linear trend in Experiment 3 (R^2=0.42; linear effect p=.05). Tannins effect on protozoa counts depended from the fermentation system, with a reduction in Experiment 1 (334 vs. 479 10^3 cell/mL, $p<.01$) and no effect in Experiment 2. In contrast, the S extract did not modify ammonia concentrations, but significantly lowered the protozoa counts in all three experiments. The reduction was 47 and 23 % in Experiment 1 and 2, respectively, and followed a curvilinear trend in Exp. 3 (R^2=0.63; quadratic effect $p<.05$).

Both the extracts did not affect the fermentation intensity measured as gas (Experiment 1 and 3) and VFA yield (Experiment 1 and 2). Changes in VFA profile induced by T were limited to reductions of iso-valerate (2.08 vs. 2.23 and 3.48 vs. 4.11 % total VFA, $p<.01$, in Experiment 1 and 2 respectively) and iso-butyrate (0.25 vs. 0.63 % total VFA, $p<.01$, Experiment 1). Tannins increased *Prevotella ruminicula* and *Selenomonas ruminantium* groups and decreased the *Ruminobacter amylophilus* ($p<.1$, $p<.05$ and $p<.05$, respectively). The S extract increased *Treponema saccarophylum* ($p<.05$).

In conclusion, both substances influence the rumen environment, having S reduced protozoa counts and T lowered rumen ammonia. The rumen effects of both extracts were appreciable at low dietary doses and negative impacts on fermentation were only limited to protein degradation reduction by T addition.

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O018

Hydrolysable tannins: inhibition of microbial abundance in the caecum thereby affecting skatole and indole production in entire male pigs

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Bioactive substance such as hydrolysable tannins (HT) are of great interest in livestock production due to their proposed positive impact on health. In this regard, the interaction between HT, gut microbiota and host needs to be considered. A bidirectional link between the biotransformation of HT into their metabolites by gut microbiota and the modulation of gut microbiota composition by HT and their degradation products exists, and it could affect gut health outcomes. In this study, 44 Swiss Large White entire male pigs (body weight 26.0 ± 4.95 kg, age 70 ± 10 days) were fed control (CT) and chestnut tannin extracts (Castanea sativa) enriched (3%) diets. Pigs were slaughtered at 170 days of age and the content of the caecum was collected within 30 min after exsanguination. The microbiota composition was investigated through the 16S rRNA gene sequences obtained by the next generation sequencing (Illumina MiSeq) and analysed by using the open source pipeline Quantitative Insights Into Microbial Ecology (QIIME), version 1.9.1. Analysis of similarity (ANOSIM) was used to evaluate whether gut microbiota and HT supplementation differed ($p<.05$) among diets, also considering the effect size of the test (R). Hydrolysable tannins reduced the bacteria abundance ($p=.001$) without affecting the within community bacterial biodiversity ($p=.167$). Both unweighted and weighted beta diversity analysis showed a clear clusterization ($p<.05$) of microbiota composition between the CT and the HT groups. Accordingly, substantial differences between the two dietary groups with respect to the abundance of several OTUs has been found, confirming the overall antibacterial effects of HT with no detrimental effects on the gut ecosystem. At the same time, HT supplementation could lead to lower apoptosis of intestinal epithelial cells due to the inhibition of the microbial growth, together with qualitative changes in the microbiota population between samples. This limits the availability of L-tryptophan from cell debris and consequently microbial mediated production of skatole production, the latter being a main component of boar taint in pork from entire males. Hydrolysable tannins are a promising alternative to conventional antibacterial additives with no detrimental effects on pig gut health and appealing properties for reducing synthesis of main components of boar taint.

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O019

In vitro evaluation of tannin-based additives for weaned piglets

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Postweaning diarrhoea (PWD) is one of the major concerns of swine livestock worldwide and it is responsible for important economic losses. PWD in piglets is commonly caused by enterotoxigenic *Escherichia coli* (ETEC) and the virulence of this pathotype is associated with the production of enterotoxins and adhesins, such as F18 and F4 fimbriae. Antibiotics are widely used for the control of PWD but alternatives are urgently needed because of the spread of antimicrobial resistance. Vegetable tannins possess various biological properties such as anti-microbial activity. Nevertheless, their effectiveness as an alternative to in-feed antibiotics showed great variability in the field. For these reasons, the main objective of this study was to evaluate the antibacterial activity of tannin-based additives, obtained from chestnut, quebracho, and a mix of them, against *E. coli* $F18^+$ and $F4^+$, the main *E. coli* serotypes responsible for PWD. The *E. coli* strains were characterised by PCR. Overnight-grown *E. coli* $F18^+$ and $F4^+$ were inoculated (10^7 CFU) into tubes containing 10 mL of Luria-Bertani medium (LB) supplemented with 0, 200, 400, 600, 800, and 1200 µg/mL of tannin extracted. Tubes were cultured aerobically at 37 °C and the bacterial growth-rate was determined spectrophotometrically by optical density at 600 nm (at 60 min intervals for 6 h and then after 24 h). The minimum bactericidal concentration (MBC) of tannins, ranged from 1 to 10 mg/mL, was evaluated through 96-well plate microdilution method on both *E. coli* strains at 24 h of incubation at 37 °C at 200 rpm. Results proved that tannins can exert a significant antimicrobial effect on the *E. coli* growth in a dose-dependent way in the first six hours of treatment ($p<.01$). The maximum percentage of inhibition, corresponding to a reduction of 68.01% of bacterial cells, was observed against F4+*E. coli* after 5 h of incubation with 1200 µg/mL of tannin extracted. At sub-MBC levels, no effects were observed after 24 h of tannins' exposure. The antimicrobial activities of both chestnut and quebracho tannins, analysed separately, resulted lower than the mix of them, suggesting a synergic effect. Tannins could be valuable feed additives for the control of *E. coli* infections in weaned piglets.

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O020

Effects of dietary supplementation of different tannin sources on productive performance and plasma metabolomics profile in broiler chickens

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Although condensed tannins are not absorbed by the animal and thereby completely excreted, the effects of the dietary administration of hydrolysable tannins on animal health and performance are not completely understood. This study aimed at evaluating the effect of the dietary supplementation of commercial tannin sources showing different composition on productive performance and plasma metabolomics profile in broiler chickens. A total of 2340, 1-day-old chicks was divided into 4 experimental groups (9 replications/group): CON, fed a commercial basal diet, and A, B and C groups receiving CON diet supplemented respectively with tannin source A (constant dosage 0.3%), B (0.13% in pre-starter and starter feed; 0.12% in grower and finisher feed) and C (0.13% in pre-starter and starter feed; 0.12% in grower and finisher feed) according to the manufacturer recommendations. Birds were weighed on a pen basis at housing (0 day), at each diet switch (10, 21, and 30 days), and at slaughtering (49 days). Feed intake was determined at the end of each feeding phase, while mortality was recorded on a daily basis. Daily weight gain (DWG), daily feed intake (DFI) and feed conversion ratio (FCR) were calculated accordingly. At processing, the incidence and severity of foot pad dermatitis (FPD) were assessed on all the birds using a 3 point-scale evaluation system while plasma was obtained from 9 birds/group (1 bird/replication) and subsequently analysed applying a nuclear magnetic resonance spectrometry approach. In the overall period of trial, the dietary administration of tannin significantly reduced DFI in groups A and C compared to CON (129 and 129 vs. 134 g/bird/d, respectively; $p<.001$) and negatively affected body weight in group C (3472 vs. 3599 g, respectively for C and CON; $p<.05$). FCR was not significantly influenced by the treatment except during the pre-starter phase where CON was significantly lower than A (1.507 vs. 1.562 respectively; $p<.05$). Similarly, FPD occurrence highlighted no significant change according to the dietary treatment. However, tannin administration significantly altered the plasma concentration of 9 metabolites (uridine, mannose, ascorbate, threonine, dimethyl sulphone, 2-oxoglutarate, pyruvate, acetate and propylene glycol) mainly involved in biological pathways related to energy and protein metabolism, adding important insights regarding the effects of these compounds on metabolic aspects of fast-growing broilers.

O021

Intramuscular fatty acids in lambs supplemented with different tannin extracts

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Thirty-five 2-month-old male lambs (initial BW 19.6 ± S.D. 2.06 kg) were individually penned and allocated at random to the 4 experimental diets (Control, Chestnut, Mimosa and Gambier) to evaluate the effect of the dietary inclusion of hydrolysable and condensed tannin-rich extracts on the fatty acid (FA) composition of lamb meat. The control group (CO) received a barley-based concentrate diet, while the other experimental groups received the same diet as the control lambs with the addition of 4% (as fed) of hydrolysable tannin extract from chestnut (CH) or two different condensed tannin extracts from mimosa (MI) and gambier (GA). During the experimental period (75 days), the lambs were fed *ad libitum* with their respective diets. Daily, individual intakes were measured according to the refusal left by each animal. At the end of the experimental period, all the lambs were slaughtered on the same day at a commercial abattoir. After 24-h-storage at 4 °C, carcases were halved and the entire *longissimus thoracis et lumborum* (LTL) muscle was removed from the right side, packed under vacuum and stored at –80 °C until fatty acids analysis.

Dry matter intake, final body weight and carcase weight were depressed by Chestnut extract as compared to the other treatments. Intramuscular fat, total saturated (SFA), monounsaturated (MUFA) and polyunsaturated fatty acids (PUFA) were not affected by tannins. While, the sum of odd and branched chain fatty acids (OBCFA) was lower in CH treatment than CO group. Rumenic and vaccenic (VA) acids were lower in Gambier than in Control meat. Tannin supplementation affected total *trans*- 18:1 ($p<.001$) and the VA / total trans- 18:1 ratio ($p<.05$). Iso FAs were higher ($p\leq.05$) in the Chestnut than in Mimosa or Gambier lambs. Our findings do not allow to describe specific mechanisms of action of the tannins on precise steps of the fatty acids metabolism but suggest a general action on the whole biohydrogenation process. Nevertheless, a multivariate approach to the data clearly discriminated the dietary treatments by means of the intramuscular fatty acid profile. The fatty acids with a high discriminant capacity arise from microbial metabolism and from the principal or alternative BH pathways. This confirms the impact that dietary tannins exert on the rumen BH and suggests that they can have a generalised influence on BH rather than effects on one or more specific steps of the process.

ANIMAL BREEDING AND GENOMICS – POULTRY GENETICS

O022

Genetic variability among Mexican, Italian, Narragansett and hybrid turkey populations using SNP and CNV markers

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The domestication of the turkey occurred in Mexico between 200 B.C. and 700 A.D. The Spanish conquerors introduced the species in Europe and since then, more than 500 years ago, the evolution of the Mexican turkey populations occurred independently from other European ones and from commercial hybrids. This study examines the genomic diversity of 6 Italian turkey breeds and the Mexican population using Copy Number Variants (CNV) and SNPs markers. A total of 31 Mexican turkeys, 116 individuals from 6 Italian breeds (Colle Euganei, Bronzato Comune Italiano, Parma e Piacenza, Brianzolo, Nero d'Italia and Ermellinato di Rovigo) 7 Narragansett and 38 commercial hybrids were genotyped using the Affymetrix 600K SNP turkey array. After stringent quality control, the HMM of PennCNV software was used to call CNVs using LogR Ratio and B allele frequencies. When overlapping the CNV were summarised into regions (CNVRs) at single breed/population level with BEDTools. Variability among populations was obtained by hierarchical clustering analysis using the pvclust R package and by principal component analysis (PCA). A total of 2987 CNV were identified covering 4.65% of the autosomes of the *Turkey_5.0/melGal5* assembly. The total number of CNVRs obtained across all the populations were 1659, 51% containing genes, 1190 gains, 412 losses and 57 complexes, covering a total of 2.55% of the genome with average CNVR length 13,612 bp. The average F_{IS} resulted -0.016 and average F_{ST} 0.249. Both SNPs markers and CNVs clustered the individuals according to breed origin and their geographical distribution. The Italian breeds result to be well distinct among them and from the other ones. Nevertheless, while the SNPs clustering can be interpreted as providing a spatial distribution of breeds associated with their actual geographical location and providing indication on past migration events and long-term directional selection, the clustering obtained for the CNVs indicates that grouping of individuals occurred more closely related to their adaptive evolution, as several research works developed in various species are suggesting. These results reflect the human action on domestication of wild turkey, its migration to Europe and the directional selection resulting in selected populations and in commercial hybrids in the last 40 years.

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O023

Analysis of run of homozygosity in turkey populations

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The aim of this study was the characterisation of run of homozygosity (ROH) in nine different turkey populations in order to identify the genomic regions with high ROH frequencies that may have occurred due to natural and artificial selection. The genotypes from the high-density Affymetrix 600K SNP turkey array were obtained for a total of: 116 individuals from 6 Italian breeds (Colle Euganei, Bronzato Comune Italiano, Parma e Piacenza, Brianzolo, Nero d'Italia and Ermellinato di Rovigo); 7 Narragansett turkeys; 38 turkey from a Commercial Hybrid; 31 Mexican turkeys. A total of 604,196 loci on autosomes were used to identify ROH that were defined setting a minimum of 1000 kb in size and 50 homozygous SNPs; additionally, a maximum gap between SNPs of 100 kb was predefined in order to assure that the SNP density did not affect the ROH. The ROH were obtained with the SVS 8.4 software of Golden Helix®. The proportion of the total genome length affected by ROH was calculated and represent an estimate of the genomic inbreeding F(ROH). The total number of ROH in the overall populations was 3782 with an average number per individual of 42. The breed with the largest average number of ROH (within breed) for individual was the Colli Euganei with 75 (min =11; max =157) while the Mexican (min =1; max =47) and the Hybrid (min =5; max =21) population showed an average number of ROH of 11 and 12 respectively. The Commercial Hybrid was the population with lower number and less variation among individuals of ROH comparable to the Mexican population. According to these results, the Mexican population appears to be under an outbreeding reproductive scheme: in fact, it is farmed as a free-range backyard population where animals are free to mate and migrate across family groups and villages. The two Italian populations with the largest average number of ROH are the Colli Euganei and the Brianzolo with 75 and 64 ROH respectively. The ROH varied in length from 1.4 Mb to 8.37 Mb. Gene Ontology (GO) and KEGG pathways terms for the genes contained in the ROH were identified. The average F_{IS} among all populations in this data set resulted 0.28. The average F(ROH) calculated on the autosome genome length of the Turkey_5.0 assembly resulted 0.031.

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O024

Genetic diversity assessment of Kwa-zulu natal native chickens using SSR markers

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Commercialization of breeding in domestic animals has gradually favoured the use of high productive exotic breeds and consequently led to lower population sizes of indigenous, low performing native breeds and South African local chicken populations are no exception to that. Indigenous chickens are recognised as an important component of the rural household livelihood by providing a source of income, and as gifts to strengthen social relationships at a cheaper cost. Characterisation of these important genetic resources can be the first step for their effective management and utilisation, which will facilitate their conservation. The aim of this study was to investigate genetic variation within and between four Kwa-Zulu Natal indigenous chicken populations using 19 microsatellites loci recommended by FAO 2004. Blood was collected from 199 animals of four different regions of Kwa-Zulu Natal: Jozini, Pietermaritzburg, Newcastle, and Port Shepstone. Pure breeds of some South African indigenous chicken breeds (Potchefstroom Koekoek, Ovambo and Venda) were included. One exotic breed (White Sussex) was also sampled to trace any cross breeding. The following parameters were analysed: genetic variation, genetic differentiation, genetic distance, genetic structure and admixture. A total of 161 alleles were observed with an average of 8.47 allele per locus across the 19 microsatellites loci in the eight studied populations. All studied markers were found to be polymorphic. The mean number of observed alleles ranged from 4.63 (Pietermaritzburg) to 5.32 (Port Shepstone). The highest observed heterozygosity (0.70) was detected in Jozini, whereas the lowest (0.61) in Pietermaritzburg. The inbreeding coefficient estimated ranged from −0.0382 in Newcastle to 0.0737 in Pietermaritzburg. The Reynolds weighted genetic distance revealed three distinct clusters; the first cluster included Port Shepstone, Newcastle, Pietermaritzburg and Jozini, the second had Ovambo and Venda, while the last was made of Koekoek and White Sussex. The structure analysis results ascertained that Kwa-Zulu Natal indigenous chickens have distinct gene pools with some level of genetic admixture. The analysed populations are characterised by a noticeable genetic variation; nevertheless, suitable conservation strategies must be planned out before their gene pool could be diluted by uncontrolled breeding with other exotic chickens.

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O025

Mating strategy based on DNA parentage information in Italian chicken breeds

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Bionda Piemontese is a slow growing chicken breed, mainly reared for egg and meat production. In this study, we assessed the effect on growth traits of a mating scheme base on genomic parentage information aimed to minimise progeny inbreeding. One hundred and twenty birds (63 males and 57 females) of Bionda Piemontese, were genotyped by a set of 14 microsatellite markers. For each subject, the genetic distances were calculated. Six family lines were identified and hens for each line were grouped in a single box; in every generation for each line the cock with highest genetic variability was identified and mated with the most distant female genetic line. Four hundred and forty individuals of three generations (G0, G1 and G2) were weighed every 15 days from hatch to 180 days of age. Gompertz linear model was used to describe the growth index over the three generations. Daily growth rate significantly increased ($p<.001$) over successive generations in males (G0 = 16 g/d; G1 = 20 g/d; G2 = 23 g/d) and in females (G0 = 12 g/d; G1 = 14 g/d; G2 = 18 g/d). Inflection point age significantly decreased in the last generation ($p<.001$) in males (G0 = 82 d; G2 = 64 d) and in females (G0 = 76 d; G2 = 61 d). Live weight in correspondence of the inflection point increase the last generations in males (G0 = 1035 g; G2 = 1067 g) and in females (G0 = 786 g; G2 = 795 g). Estimated weight at 180 days of age increased over the generations: +20% in cocks (G0 = 2212 g; G1 = 2453 d; G2 = 2657 g) and +17% in hens (G0 = 1713 g; G1 = 1752 g; G2 = 2000 g).

The results showed an improvement of growth performance as rate of growth and final body weight; this can be related to offspring heterozygosity increasing (G0 = 0.66%; G1 = 69%; G2 = 70%) and consequently to inbreeding depression reduction. The use of molecular parentage in mating schemes could be a reliable tool for the management of small size chicken populations and the improvement of their production.

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O026

Exploring polymorphisms in genes affecting energy metabolism and skeletal muscle in broiler chickens

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Chicken meat is one of the most popular sources of animal protein for human consumption worldwide. Through advances in genetic selection, farming practices and nutrition, the production of broiler chickens has become more efficient. Genetic selection has contributed significantly to the improvement in growth rate, biological efficiency, breast yield, longevity and leg health. Besides, any condition which impacts the quality of breast meat is of great importance to breeding companies and broiler producers. Carcases affected by breast muscle myopathies (BMM) can be downgraded or in some cases condemned, resulting in economic losses for poultry meat producers. In the present work, we investigated three genes, Peroxisome proliferator-activated receptor gamma coactivator 1-alpha (*PPARGC1A*), that plays a role in glucose and fatty acid metabolism, Growth differentiation factor-8 (*GDF8*) and E3 ubiquitin-protein ligase (*WWP1*), involved in skeletal muscle growth. For the experiment, 90 chickens of six different genetic lines were recruited. *Post mortem* breast fillets were scored for different degrees of white stripping and fatty acids were measured both in breasts and legs; DNA was extracted from breast meat. The III and VIII exons for *PPARGC1A*, I and III exons for *GDF8*, X exon for *WWP1* were amplified by polymerase chain reaction (PCR) by using primers designed in the flanking introns and then sequenced. For *PPARGC1A* four single nucleotide polymorphisms (SNPs), were identified, three in the flanking region of III exons, that did not influence splicing sites and one in VIII exon, that caused a missense mutation (C348W). Four SNPs, that caused synonymous mutation, were detected in *GDF8*. Fifteen SNPs were identified in the intron region of *WWP1*. Allelic, genotypic and haplotypic frequencies were calculated for all SNPs. The effect of the non-synonymous variant C384W on PPARGC1A protein function was evaluated by using the SIFT (**S**orting **I**ntolerant **F**rom **T**olerant) algorithm and the substitution of cysteine with tryptophan in the C-terminal region of protein was predicted not tolerated. Because proteins encoded by the three investigated genes are in the same metabolic network and have been shown to interact, the identified mutation could have an effect on skeletal muscle structure and growth.

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LIVESTOCK SYSTEMS – GHG EMISSIONS IN LIVESTOCK

O027

Gas and methane production from two rumen inoculums, used warm, refrigerated, chilled or freeze-dried

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The aim of this research was to evaluate the effect of two sources of rumen inoculum and the effects of its preservation method (warm, refrigerated, chilled, freeze-dried) on gas (GP) and methane production (CH_4) *in vitro* to identify a technique that makes the inoculum standardisable and constantly available, reducing costs and respecting animal welfare.
Both sources of inoculum were compared: rumen fluid collected at slaughtering house (RF) from four dry multiparous Holstein Friesian cows and fermentation fluid produced by a Rumen Continuous System (RCS). The sources of inoculum were preserved with four methods: warm at 39 °C (W), refrigerated at 4 °C (R), chilled at −20 °C (C) and freeze-dried (F). Both for RF and for RCS, W and R fluids were used as inoculum the same day of collection, while C and F were stored and used later in a separated incubation. Each inoculum (25 mL) was incubated with a Menke medium (50 mL) using 6 feeds (0.5 g, forages and concentrates) in 2 replicates. Data were analysed according to the following schema: 2 runs ×2 sources of inoculum ×4 methods of preservation ×6 feeds ×2 replicates. The Ankom Gas Production System was used to evaluate the GP and CH_4 up to 24 h of incubation.
Compared to the RF, RCS reduces GP (from 190 to 143 mL/g DM, $p<.01$), decreases CH_4 (from 23.1 to 12.8 mL, $p<.01$), but improved

the repeatability of the measurements reducing the coefficient of variation (CV, %) of measurements of GP (18 vs. 8%, respectively, $p<.01$) and CH_4 (19 to 16%, respectively, $p<.01$). The warm ruminal liquid (W) produces on average 189 mL of GP and 28.9 mL of CH_4 ($p<.01$). Compared to W, R increases GP (+5.9%) and CH_4 (+10.0%), and also slightly improved the repeatability of the measurements. Compared to W, C and F inoculums strongly reduced GP (−17.5 and −35.4%, respectively, $p<.01$) and of CH_4 (−63.0 and −99.7%, $p<.01$), but the repeatability of the measures was improved.

In conclusion, the use of a RCS to produce inoculum for *in vitro* study slightly reduced the fermentation of feeds but allowed to standardise the technique, improving the repeatability of measurements. Refrigeration improved the feed fermentation and the repeatability of measurements, proving to be useful for a short-term rumen fluid preservation. The procedures of chilling and freeze-drying of rumen fluid, adopted in this research, inhibited the microbial activity and methane production.

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O028

Fermentation pattern and volatile organic compounds to cluster poorly and well-preserved silages by a multivariate approach

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Q11

The aim was to cluster corn silage quality based on chemical composition, fermentative traits and volatile organic compounds (VOCs). Sixty corn samples were collected in 2018 and were characterised by chemical-biological parameter ($n = 11$), fermentative traits ($n = 16$), and VOCs ($n = 58$). Fermentative quality indexes (FQI), Flieg–Zimmer or Vanbelle scores were calculated. In particular, chemical and biological parameters were estimated using near-infra-red (NIR) spectroscopy. Fermentative traits (pH, volatile fatty acid, ethanol and ammonia nitrogen) and VOCs (alcohols, ketones, esters and aldehydes) were quantified by GC/FID apparatus. Lactic acid and propylene glycol by HPLC. The mean (±s.d.) values for FQI, Flieg-Zimmer and Vanbelle indexes were respectively 56.8 ± 16.1, 73.8 ± 13.6 and 79.7 ± 5.3. Then, a principal component analysis (PCA) was carried out and fourteen principal components (PCs) were extracted and biologically interpreted based on end-products related to homolactic, heterolactic, clostridia and aerobic fermentations. Further, other PCs appeared related to chemical reactions and other fermentations occurring during ensiling, with the latter still not linked to specific microorganism communities. Then, the Pearson's correlation coefficients were calculated between scores of each corn silage on the fourteen extracted and interpreted PCs and FQI, Flieg–Zimmer's, or Vanbelle's values. The FQI was positively related ($p<.05$) to homo- and hetero-lactic fermentations as well as oxidation of alcohols, whereas negative relationships ($p<.05$) were observed with PCs representing clostridia fermentations. The Flieg-Zimmer index showed a positive correlation ($p<.05$) with homolactic fermentation, whereas this relationship was not significant for Vanbelle index. Flieg-Zimmer or Vanbelle scores were negatively related to the two PCs representing heterolactic and *Lactobacillus buchneri* fermentations. Concluding, the FQI index appeared adequate to cluster poorly or well-preserved corn silages, whereas Flieg-Zimmer and Vanbelle indexes were unable to properly discriminate these silages for their fermentation quality. Furthermore, despite the multivariate approach used in current work permitted to identify the main chemical reactions and fermentation pathways occurring during ensiling, the biological interpretation of all PCs deserves of future investigation to be completely understood.

O029

Evaluation of an additive capable to improve ruminal fermentations through the use of an automated gas production system

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Aim of the present work was to test the effect of an additive, Bioflavex (Exquim S.A., Barcelona, Spain), a blend of natural flavonoid extracts comprising naringin (200 g/kg of product), extracted from bitter orange (Citrus aurantium) and grapefruit (Citrus paradisi; 400 g/kg of product) on the in vitro fermentation of a total mixed ration (TMR) for dairy cattle. Fermentation performances were evaluated through the gas production system, using the Gas Endeavour instrument which: (a) operate with 400 mL of rumen inoculum and the buffer solutions; (b) simulate the physiological rumen movements by discontinuous stirring; (c) operate using a substrate quantity between 2 and 8 g of dry sample or about 20 g of fresh forage, (d) measure and register continuously the amount of gas produced in 15 batches. Cumulative gas production is evaluated at a temperature of 39 °C along with stirring. All medium ingredients, as Van Soest buffer, macro and micromineral solutions, were prepared on the base of rumen fluid inoculum and sample substrate amount, and the whole mix proportioned to batches of 400 mL. The fermentation trials were made on dairy cows TMR typical of Parmigiano Reggiano cheese production area added with Bioflavex at the equivalent doses of: 50, 100, 200 and 400 g/cow/day. It was hypothesised that Bioflavex could exert some antimicrobial effects on ruminal bacteria. Five grams of pre-dried TMR control sample and 5 g of pre-dried TMR experimental samples were placed in three batches for each treatment. The fermentation trials lasted 24 h and were repeated twice. Volatile fatty acids (VFA) were determined on the fermentation fluids by HPLC. The average total amount of gas produced (mL/g DM), was similar between thesis, while the gas production rate was significantly modified by the presence of Bioflavex in the initial 8 h of fermentation. No effects were found in the following interval. In particular, Bioflavex increased the rate of gas production only at the dosages of 100, 200 and 400 g/cow/day compared to the control group (+20.4, +20.3 and +27.6% respectively; $p<.05$). Moreover, at the same doses, Bioflavex improved the synthesis of VFA in an average amount equal to +64.2% for the acetic acid, +37.2% for the propionic acid, +78.4% for butyric acid ($p<.05$). This effect was also confirmed by other researchers. Flavonoid supplementation by Bioflavex could be effective in improving rumen fermentation with increased VFA production.

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O030

Faeces as source of information on dairy cattle GHG emissions

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The methane emissions represent about 44% of the total livestock GHG emissions. The development of proxies able to predict CH_4 emission from individual cows to improve the mitigation strategies is a hot research point. Near infra-red spectra (NIRS) has been used to directly estimate methane emissions but never applied with this purpose on faecal samples and in combination with other phenotypic parameters. We aimed to verify the possibility to use NIRS of faeces (NIRSf) alone and in combination with other parameters available at a farm level to predict methane emission intensity (MI, g/kg milk) from individual lactating cows. The study was carried out on 34 Nordic Red dairy cows, fed three diets: a diet based on grass silage and concentrate (55:45 on DM basis), the same diet supplemented with rumen-protected conjugated linoleic acid or a concentrate containing a mixture of sunflower and fish oil (4% on DM basis). CH_4 emissions were measured in respiratory chambers during 4 days after 1 day of acclimatisation at 3, 7, 11, and 15 weeks of lactation, together with milk yield (MY, kg/d) and body weight (BW, kg). The total faecal collection was conducted in parallel to CH_4 measurements. A total of 115 faecal samples were used. Near infra-red reflectance spectra were recorded by a FT-NIR Analyser (MPA^{II}, Bruker Italia S.r.l., Milan, Italy). Calibration models were developed by partial least square method with test set validation. Mean observed value was 11.6 ± 2.4 g CH_4/kg milk; mean estimated value from NIRSf and from NIRSf-MY-BW were 11.9 ± 1.8 and 11.6 ± 2.1 g CH_4/kg milk, respectively. The accuracy of the calibration curves, expressed by the correlation coefficient R^2, was 0.73 and 0.79 and by RMSPE (Root Mean Square Prediction Error) was 1.24 and 1.10 g CH_4/kg milk, respectively, for NIRSf and NIRSf-MY-BW. Though methane can be predicted by models that incorporate dry matter intake together with other phenotypes, the availability of intake data represents a major limitation outside experimental conditions. Our results show that Near infra-red technique applied to faecal samples could allow fairly good prediction of methane emission intensity, although they need to be confirmed on larger datasets. In particular, when combined with other easily available phenotypic parameters, it can represent a valid option for large-scale measurements in commercial dairy farms, where individual DMI is usually not available, for genetic selection of environmentally more efficient dairy cows.

O031

Rumination patterns in dairy cows fed high concentrate ration

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The objective of this study was to investigate in high producing cows, the modifications in rumination patterns after changing diets and housing conditions. Twenty-four lactating Holstein cows divided into three consecutive blocks were exposed to two different periods: period one (PreChallenge-PC): standard diet (S, forage concentrate ratio 45:55), free-stall housing (FS); period two (Challenge-Ch): high concentrate diet (HC, forage concentrate ratio 25:75), tie-stall (TS). Both periods lasted for 4 weeks and the change was abrupt. Rumination time (RT) was measured continuously along all the time by collar, and health events were recorded. All data were statistically analysed in R (v3.5.1) with a factorial arrangement for period, block and health status using mixed model procedure, with day as repeated measure; k-means cluster analysis was performed to group cows by health status. The results obtained showed a dramatical drop in RT (−114.38 min in PC vs. Ch $p<.01$) after the abrupt change, and the major health problem occurred was the rumen hypomotility (RH). We defined RH as a drop in rumination below the physiological level, and we set the minimum at 200 min of RT per day. Number of days with RH increased in PC vs. Ch (+10.63%; $p<.01$). Using the RT and RH cases we divided the animals into clusters: cluster one (NoRH): high RT and no RH cases; cluster two (RH): low RT and RH cases. RH group resulted lower in RT for the whole trial (−155.72 min $p<.01$), but mostly in Ch period (−199.06 min $p<.01$). The RT S.D. resulted higher in RH (+14.90 min $p<.01$, the whole trial; +18.28 min $p<.05$ in Ch), and the drop in RT, after the abrupt change, resulted more than doubled in RH group (133.75 vs. 65.13 in RH and NoRH, respectively; $p<.01$). These data highlight that there are different responses in dairy cows fed high concentrate ration, being possible to divide the population into two groups with high and low susceptibility. Moreover, these innovative results set a new level of RT considered below the physiological minimum (≤200 min/d), acceptable to provide a rumen healthy condition.

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – MICROBIOME

O032

Effects of sugars on *in vitro* rumen microbial ecosystem

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Molasses are not properly characterised, even if considered as sugar sources. Moreover, no researches have been made to evaluate their impact on the rumen microbial ecosystem. In this study, four different molasses (2 cane and 2 beet, 77% D.M., 9.5% ash, 40% sucrose, as fed) were fermented using the traditional procedure for *in vitro* ruminal digestion. Each molasses was fermented in duplicate, by adding 1 g per flask containing 0.5 g of forage and 40 mL of buffer. Once anaerobiosis was reached, 10 mL of rumen fluid from lactating Holstein cows was inoculated. Time points analysed were 1 h, 8 h and 24 h. For each one of it, a blank sample containing the forage was also fermented. Flask's content was then poured into a falcon tube and placed at −80 °C prior DNA extraction. This procedure followed a specific protocol designed for rumen content. DNA concentration and quality were assessed using NanoDrop. V3–V4 region of the 16S rRNA gene was amplified, then sequencing was performed on Illumina MiSeq according to the manufacturer's instructions. Raw sequences were processed using PANDAseq, QIIME2 and DADA2. High-quality reads were clustered into high-resolution Operational Taxonomic Units (OTUs). Using taxonomic assignment, OTU tables were collapsed from phylum to genus level. Alpha diversity was computed using OTUs, Shannon index and Faith's diversity metrics. Beta diversity was estimated by computing weighted and unweighted UniFrac distances into Principal Coordinates Analysis (PCoA). Statistical analysis was performed using R studio 1.0.136. Significant differences in alpha or beta diversity were assessed by Wilcoxon test.

Results obtained with the Beta diversity indicates that molasses addition strongly impacted the microbial community during time. Highest differences were observed after 8 and 24 h, while high similarity was observed at 1 h among blank and molasses samples. Among known species, molasses positively impacted *Butyrivibrio spp.* (1% vs. 2.8% in blank and molasses, respectively, $p<.05$) as well as *M. elsdenii* (0.4% vs. 1.1% in blank and molasses, respectively, $p<.01$) and *S. bovis* (5% vs. 27% in blank and molasses, respectively, $p<.01$), while no differences among molasses and control were observed on *Prevotellae spp.* nor *Ruminococcaceae spp.* In conclusion, molasses addition severely impacted the rumen microbial ecosystem, and utilisation of such products could be useful to drive specific metabolic patterns while feeding the rumen with high energy sources.

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O033

Modelling *in vitro* dynamics of fermentation and methane production by cow rumen microbiota at different total mixed rations

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A mechanistic dynamic model has been developed to describe *in vitro* rumen fermentation. The model includes stoichiometry of fermentation, microbial metabolism, acid–base reactions and liquid–gas transfer. In particular, the rumen microbiota is represented by three functional microbial groups associated with the utilisation of hexoses, amino acids and hydrogen. The model is then represented by ordinary differential equations for 18 compartments. The model was able to capture the dynamics of fermentation of the database used for model calibration. However, to assess its robustness, it was identified as a crucial step to challenge the model predictive capabilities under different conditions. The objectives of this work were (i) to evaluate the model using independent data set and (ii) to assess whether the hydrolysis rate constants of diet polymers determined enzymatically could be integrated into the model as known parameters while keeping satisfactory predictions. Twenty-two total mixed rations (TMR) were collected by different dairy farms in 2018. All TMR were characterised for chemical parameters, along with NDF, crude protein and starch degradability rates. Six analytical replicates for each TMR were simultaneously incubated in diluted rumen fluid. At each time of incubation (i.e. 2, 5, 9, 24, 36 and 48 h) one replicate was used to determine the gas volume and composition, as well as rumen VFA, ammonia nitrogen and soluble sugar contents. In particular, two millilitres of gas was sampled from the headspace of these bottles and analysed by gas chromatography to determine gas composition (CH_4, O_2, H_2). The fermentation fluid (10 mL for each analysis) were used to determine ammonia nitrogen, VFA and soluble sugars. Model calibration was performed for VFA and CH_4 using a maximum likelihood estimator approach. The model represented satisfactorily VFA concentrations and CH_4 production with Lin's concordance correlation coefficient (CCC) of 0.91 in average. These results strengthen the capabilities of the model to predict rumen fermentation pattern and CH_4 production. They also show the potential of using prior values of hydrolysis rate constants obtained enzymatically into the model to prediction rumen fermentation pattern. Our perspectives include model extensions such as the incorporation of lipid metabolism and thermodynamic factors to enhance the model capabilities for the assessment of strategies for methane mitigation.

O034

Study of the *in vivo* periodic fluctuation of microbiome composition and enzymatic activity of rumen fluids used as inocula

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Aim of the present work was to evaluate the microbiome composition (MC) and the relative enzyme activities (EAs) to assess individual, daily and weekly fluctuations of rumen fluids (RFs) inocula degradative capacity. MC and cellulase (Ce), amylase (Am) and xylanase (Xy) activities were tested on the liquid fraction of RFs sampled before –T0–, and after 4 and 8 hours from the main meal –T4 and T8 respectively – in 3 days with one-week interval for a total of 3 weeks. RFs were collected from 4 Holstein cows (2 heifers, 2 adults) fed a hay-based diet. The liquid fraction of each sample was divided into 2 aliquots: one was centrifuged and filtered through 0.45 µm filter porosity for the EAs tests; the other was freeze-dried for the MC analysis. Bacteria were extracted using the Repeated Bead Beating Plus column method followed by PCR amplification of the V3–V4 hypervariable regions of the 16S rRNA gene. Libraries were sequenced on an Illumina MiSeq instrument with a paired-end 2 × 300 run. MC data were analysed by QIIME pipeline using Greengenes as reference database. The EAs were tested using the radial enzyme diffusion method using substrate concentrations of 0.5%, 0.5% and 0.1% for Ce, Am and Xy respectively, and expressed as area of the halos surface (mm^2). EAs statistical analyses were performed using the Linear mixed model. MC did not differ daily, but individual ($p \le 001$) and weekly ($p \le 001$) differences in bacterial composition (beta-diversity) were found. The individual differences were more marked among heifer and adult cows. Overall, individual, weekly and daily EAs variations were found ($p \le 001$) while not significant daily differences for Am were measured in analogy with MC findings. The lowest Xy was found in the 1st week (177.86 vs. 193.34 and 196.50 mm^2, $p \le 001$) while Ce showed a depression in the 2nd week (239.32 vs. 315.50 and 276.35 mm^2, $p \le 001$) and Am was depressed in the 3rd (110.75 vs. 136.36 and 153.35 mm^2, $p \le 001$) week. Regarding the daily variation, Xy generally increased starting from T0 to T4 and T8 (172.62, 192.44 and 202.64 mm^2 respectively) while Ce showed different results at T4 increasing in two cows and decreasing in the other two. Nevertheless, MC did not change over the day while weekly and individual variations appeared especially when heifer and adult cow were compared. Enzymatic activity of rumen fluids varied individually, weekly and daily, therefore degradative capacity could vary widely among inocula used for i*n vitro* procedures.

O035

Effect of physiological stages of dairy cows on rumen protozoal community composition revealed by RNA based amplicon sequencing

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Rumen protozoal communities produce a large amount of hydrogen as part of metabolism. Due to interspecies hydrogen transfer with their symbiotic partners (methanogens), protozoa play an important role in rumen methanogenesis. Therefore, in order to reduce ruminal methane emission, a thorough understanding of rumen protozoal community composition is very important. This study was aiming to monitor seven lactating Italian Simmental cows during different physiological stages, to observe possible modifications in their metabolically active rumen protozoal communities, based on 18S rRNA amplicon sequencing. The experiment was divided into 3 parts depending on different physiological stages of dairy cows: late lactation (248–332 DIM), dry period Q12 (8–51 days before calving), and postpartum (15–34 days after calving). Rumen fluid samples were collected using an oesophageal probe from each cow, maintained on a specific total mixed ration (TMR) at least for 10 days before collection. RNA extraction was done in duplicates and used as template for synthesis of cDNA, that was later amplified by PCR using specific primers for eukaryotic hypervariable V9 region of 18S rRNA gene, followed by sequencing using 300 bp paired-end Illumina Miseq platform. Furthermore, during each part of the experiment, the total number of protozoa in the rumen fluid samples was also counted manually using compound microscope. The physiological stages of dairy cows were found to have a clear effect on rumen protozoal community composition, with the most prominent effects observed on the relative abundance of following protozoal phyla (Ciliophora and Amoebozoa), and genera (Dasytricha, Eudiplodinium, Ostracodinium and Entamoeba). In conclusion, dry period resulted in higher protozoa number and an increase in abundance of ciliated protozoan populations. This study has potential future implications in microbial programming process, through controlled early life feeding managements to decrease hydrogen producing ciliated protozoans populations which play a key role in rumen methanogenesis, thereby improving health and production.

O036

Growth performance and gut microbiota in post-weaning piglets fed diets containing bakery/confectionary former food products as cereal substitute

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Former Foodstuffs (FFPs) are products that have lost their commercial value on the human consumption market, due to for example production errors. However, their nutritional value for animal feed purposes is not at all affected. Biscuits, bread, cereals, chocolate bars, pasta, savoury snacks and sweets leftovers from the food industry are all FFPs, high in energy content in the form of sugar, starch, oil or fat. For this reason, they can be considered an appealing alternative feed ingredient as valuable energy sources. This study aimed to investigate the effects of FFPs on growth performance and gut microbiota when they partially replace conventional cereal grains in post-weaning piglet's diets. After an adaptation period, 12 post-weaning piglets were housed for 16 days in individual pen and were fed two diets, including a standard wheat-barley-corn meal control diet and a diet containing 30% FFPs partially substituting for conventional cereals. The diets were isoenergetic and isonitrogenous and met NRC (2012) requirements. For the growth performance evaluation, individual feed intake and piglet's body weight (BW, kg) were recorded, while average daily gain (ADG kg/day), average daily feed intake (ADFI kg/day) and feed conversion ratio (FCR kg/kg) have been calculated. Stool samples have been collected for the next generation sequencing of the variable regions V3 and V4 of the 16S rRNA gene in order to characterise the faecal microbiota composition. At the end of the experiment, no differences in BW, ADG and ADFI have been observed between groups. Conversely, piglets on the FFP diet showed a better FCR ($p<.05$). The gut microbiota did not show differences in microbial taxa composition, while further investigations are necessary to clarify the effects of FFPs on gut bacterial abundance and biodiversity.

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O037

Administration of green tea and pomegranate extract in drinking water on broiler growth performance, total blood antioxidant activity and gut microbiota

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The plant-derived products as green tea extracts have been extensively used in poultry nutrition due to their antioxidant, anti-viral and anticoccidial properties. Furthermore, they have the capacity to lower cholesterol and reduce lipid peroxide level in plasma and meat, and improve the growth performance and meat quality of broiler. The pomegranate peel extract has also been showed pharmacological, anti-inflammatory, antioxidant and anticoccidial properties. The combination of green tea and pomegranate extract might be able to modulate intestinal microflora. The aim of this study was to evaluate the influence of the combination of natural extract of pomegranate and green tea extract on growth performances, blood antioxidant status and gut microbiota in broiler chicks. A completely randomised block design with two experimental treatments (CTR and plant extracts, PE) was carried out. Each treatment group included 240 male birds allotted in 12 pens (replicates). PE were supplemented through drinking water at the dosages of 0.2 mL/L at days 0–4, 10–11, 20–21 and 45–46. During the trial, birds weight and pen feed intake were measured. At slaughtering (day 50), blood samples and caeca content were collected from one broiler per pen. Moreover, carcase and breast percentage were also evaluated. Blood total antiradical activity was measured by KRL test. 16S amplicon sequencing for microbial community profiling was performed on caeca content. Feed conversion ratio was significantly affected by PE treatment from 21 to 50 days (p=.029). No other growth and slaughter parameters were affected. Total antioxidant activity on blood (p=.009) and red blood cells (p=.02) were higher in PE group. Microbial profiling analysis showed no difference in richness and diversity of caeca bacterial composition between groups. Microbiome analysis indicates a slight increase in treated group of beneficial bacteria such as *Rikenellaceae*, *Ruminococcaceae, Lactobacillaceae* that represent the most abundant families found in the gut environment and have been associated with the maintenance of gut health. The obtained results indicate that administration of pomegranate and green tea extracts through drinking water is able to ameliorate the total blood antioxidant activity and modulate gut intestinal health.

ANIMAL BREEDING AND GENOMICS – GENETIC DIVERSITY

O038

Genome-wide association study and functional analysis of carcase and meat quality traits in double-muscled Piemontese cattle

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Meat quality traits are important drivers of consumer acceptance and purchase intention. Recently, Piemontese meat was awarded the EU Protected Geographical Indication (PGI) status 'Vitelloni Piemontesi della coscia' which has increased the interest in improving the meat quality attributes for this breed. Therefore, aim of this study was to investigate the genomic regions and biological pathways controlling the variation in carcase and meat quality traits in double-muscled Piemontese cattle breed. We carried out genome-wide associations (GWAS) and pathway enrichment analyses for carcase traits (age at slaughtering [AS], carcase weight [CW], carcase daily gain [CDG], conformation score [EUS] and ribeye muscle area [REMA]) and meat quality traits (pH, Warner-Bratzler shear force [WBSF], purge loss [PL], cooking loss [CL] and colour parameters [lightness, L*; redness, a*; yellowness, b*; chroma, C*; hue, H*]) in a cohort of 1166 Piemontese young bulls. Animals were slaughtered at 539 days on average. Genotype data from the 'GeneSeek Genomic Profiler Bovine LD' array was used. A single marker regression model was applied for GWAS using the GenABEL R package and the GRAMMAR-GC approach. The functional enrichment analysis was conducted using the Cytoscape plugin ClueGo to identify significantly overrepresented pathways and ontologies (right-sided hypergeometric test with false discovery rate correction). In total, we identified 37 significant single nucleotide polymorphisms (SNPs), which were associated with 12 traits ($p < 5 \times 10^{-5}$). Notably, 14 SNPs associated with CW, CDG and AS were detected in the region at ~38.57–38.94 Mb on *Bos taurus* autosome (BTA) 6 and mapped within four genes, i.e. Leucine Aminopeptidase 3 (*LAP3*), Family with Sequence Similarity 184 Member B (*FAM184B*), Non-SMC Condensin I Complex Subunit G (*NCAPG*) and Ligand-Dependent Nuclear Receptor Corepressor-Like (*LCORL*). Significantly enriched GO terms and KEGG pathways were found for four meat quality parameters, i.e. a*, b*, C* and CL, and for two carcase traits, i.e. CW and EUS. In particular, pathways related to transmembrane transport (i.e. oxygen, calcium, ion and cation) were overrepresented for meat colour parameters. The information acquired might be useful for genomic selection programmes aimed at improving beef production and meat quality in the Piemontese breed.

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O039

Genomic adaptation of local cattle breeds in the alpine massif

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Understanding adaptive ability of livestock is a key factor in the context of global climate change and become a crucial focus in conservation and management for a sustainable farming in a changing environment. One way to better understand adaptation abilities is to identify genes underlying adaptation phenotypes. This goal can be achieved by genetically characterising livestock species and detecting footprints of selection in the animal genome. The recent availability of genome-wide SNP panels allows providing background information concerning genome structure in domestic animals, opening new perspectives to livestock genetics.

In order to investigate the genetic regions with a potential adaptive role, we investigate 24 cattle breeds, (high productive specialised breeds and several autochthonous populations) reared in six different neighbouring nations from East to West Alpine Massif. Several analyses have been performed to detect footprints of selection and genomic regions associated with climate variables. More precisely, using breed GPS coordinates, 4 climatic variables (annual mean temperature, annual precipitation, annual mean radiation, and annual mean moisture index) were extracted from the Climond database. In order to identify footprints of selection, a whole genome scan for adaptive differentiation were performed using Bovine 50K SNP chip genotyping data with the XtX model implemented in the BAYPASS software. In addition, across-population whole genome scans for association with the population-specific climatic variables were performed using the AUX model. Footprints of selection were detected on BTA6 and BTA18 pointing out several candidate genes (i.e. LCORL, PDGFRA, KDR and SPG7); moreover different genomic regions (on BTA 6, 10, 19 and 20) were associated with annual mean radiation. Ongoing analyses will specify candidate regions and genes involved in local adaptation in the Alpine massif.

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O040

The Italian cattle breeds in a worldwide context for a genome-wide diversity study

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The availability of genome-wide single nucleotide polymorphism (SNP) data has made it possible to provide a detailed assessment of the genetic relationship among different cattle populations across the world. These comprehensive studies, however, did not include several European populations, such as the Italian breeds. With a total of 3283 individuals, representing 205 different domesticated bovid breeds, we have now assembled the largest and most divergent dataset to place the Italian breeds into a global context and to perform a worldwide study on cattle using genome-wide SNPs. To understand these aspects, in this preliminary step, we carried out multidimensional scaling (MDS) and ADMIXTURE analysis, and inferred ancestry graphs using TREEMIX. Inbreeding based on runs of homozygosity ($F_{ROH>4Mb}$) was also estimated. Once again, the global cattle genetic diversity can be described as a triangle. Patterns of genetic differentiation, shared ancestry, admixture events, and the phylogenetic tree, all suggest the presence of gene flow, in particular among breeds that originate from the same geographical area. A comparison among European breeds pinpointed a close genetic relationship between the Balkan breeds (Turkish grey and Busa) and Sicilian and Podolian-derived breeds. The Northern Italian breeds are much closer to several European breeds, which indicates a contribution of European ancestry in the formation of these Italian breeds. The ROH coverage in the genome differs considerably within and among breeds and sub-species. The highest mean value of F_{ROH} (0.26) was observed in Bali (*B. javanicus*), most likely due to ascertainment bias of the SNP chip. Among African, American and Asian cattle, the highest values were observed for some Asian *B. t. indicus populations* (e.g. Hariana breed with F_{ROH} mean value of 0.10). Among the European breeds, the lowest levels of F_{ROH} (<0.02) was found in some French (e.g. Vosgienne, Limousin), Russian (Tagil, Kalmyk), and Northern Italian breeds (Piedmontese, Rossa Siciliana) whereas the highest values (>0.05) were observed in Spanish (Toro de Lidia), England (Beef Shorthorn) and Tuscany breeds (Pontremolese, Mucca Pisana). This finding reflects the fact that recent inbreeding impacted the genome of these cattle breeds. Multiple complementary methods will be applied to reveal the complex origin of Italian breeds and to understand the genetic relationships and admixture events among worldwide cattle.

O041

Genotype by environment interactions and response to selection for productive traits in a local cattle breed

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Genotype by Environment interactions (G × E) happen when individuals have diverse adaptations to local conditions. This study aimed to investigate G × E and response to selection (R) in dairy traits and somatic cells under the diverse environmental conditions in which the local Rendena cattle breed is reared. Target traits were milk, fat and protein yields (MY, FY, and PY; kg), fat and protein percentage (F% and P%), and somatic cell score (SCS), routinely collected over 12 years as test day data of nearly 10,000 cows daughters of ~600 sires. The G x E term was valued by Gibbs sampling analysis under the reaction norm model approach, i.e. in two-steps. First, a single trait test day repeatability animal model (M1) was carried out to obtain the solutions of the herd-test day (HTD) effect, used as environmental covariate in a second random regression sire model analysis (M2) to obtain the intercept G and the slope G x E across HTD levels. The gradient of the G and G x E variance across HTD on the total phenotypic variability (M2) was used as indication of R under different environmental conditions: (i) geographical area (plain/hill/mountain); (ii) type of housing (tie-stall/loose housing); (iii) feeding system (traditional/total mixed ration); and (iv) occurrence of summer pasture (yes/no). The sire EBVs under M1 resulted variable in different environmental conditions for some traits: e.g. an average correlation of 60% was found for the F% EBVs values of the same sires in different geographical areas, type of housing and feeding system. MY and P% showed the highest correlations in different environments (83% on average). The G x E component estimated via M2 explained from 6% (SCS) to 28% (P%) of the phenotypic variance. On average, a 1.04 times greater R was found under a target environmental condition as respect to the alternative(s) (e.g. loose housing vs. tie-stall). For productive traits, the higher R was found for total mixed ration feeding, loose housing and the absence of summer pasture. Opposite results were found for SCS. Greater R was found in plain for MY, FY, PY, P%. A rank correlation between 87% (SCS) to 96% (FY) was found between the bull EBVs estimated under the reaction norm model vs. a traditional approach (M2 vs. M1), and the ranking of the first 20 bulls resulted greatly changed under the two approaches. These results suggest the importance to include G × E when local breeds are selected.

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O042

Conservation status and rates of inbreeding of Italian autochthonous beef breeds

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Albeit the success of commercial beef cattle nowadays, local breeds are an important reservoir of traits that might be crucial to meet future challenges. Genetic analysis is a primary step to study populations in order to elaborate conservation measures and handle inbreeding. This study investigates the population structure of six local cattle beef breeds through pedigree data, namely: Calvana ($n = 2798$), Mucca Pisana ($n = 3339$), Pontremolese ($n = 328$), Sarda ($n = 97{,}163$), Sardo Bruna ($n = 74{,}981$) and Sardo Modicana ($n = 25{,}355$).
The current population size of the three Tuscan breeds is extremely low counting: 263 Calvana (37 males and 226 females), 346 Mucca Pisana (52 males and 294 females) and 52 Pontremolese (8 males and 44 females) extant animals. These figures classify Calvana and Pisana as endangered breeds, while Pontremolese as critical breed.
Genetic analysis was performed by the ENDOG 4.8 software using pedigree information. To overcome pedigree incompleteness, we estimated the effective population size (*Ne*) based on the equivalent generations calculated as $\sum(1/2)^n$, where n was the number of generations separating the individual from each of the known ancestors. Pontremolese and Sarda had the lowest *Ne* (14.62 and 16.64, respectively) while Sardo Modicana the highest value (39.79). The others showed similar *Ne*, around 19. The average inbreeding coefficient for the Tuscan breeds was 7.25, 5.10 and 3.64 % for Mucca Pisana, Calvana and Pontremolese, respectively. Sardinian breeds showed the smallest values ranging between 1.23% (in Sardo Bruna) to 1.90% (in Sarda). The average generation interval in years was rather high for all breeds, with the lowest value observed in Sardo Modicana (7.8 SD =9.75) and the

highest in Sardo Bruna (13.3 SD =16.7). Following *Ne*, the rates of inbreeding per generation ($\Delta F = 1/2Ne$) were ~2.6% for Calvana, Mucca Pisana and Sardo Bruna, 3% and 3.42% for the Sarda and Pontremolese, respectively, and ~1.26% for Sardo Modicana.

This preliminary data suggests the urgency of conservation strategies for the three Tuscan breeds. In the next steps, we will further integrate pedigree and genomic information to (i) explore the population structure and genetic diversity within and between breeds, (ii) identify signatures of selection and (iii) plan assortative matings to control the inbreeding.

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O043

Twenty shades of grey. Combined analysis of genome-wide SNP data in Steppe cattle and in Mediterranean grey cattle shed new light on the molecular bases of coat colour

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Coat colour is among the most distinctive phenotypes in cattle. To date, several genes underlying this complex trait have been identified. A number of breeds, mainly in Europe and Asia, share peculiar coat colour features such as the presence of (i) a diluted grey coat, (ii) lightly coloured inner sides of the legs and belly, (iii) aureoles around the eyes, a muzzle ring, black nasal mucosa, and dark tail tip, (iv) a fawn pigmentation of the calf at birth, turning over time to grey and (v) sexual dichromatism. The aim of this study was to investigate the molecular basis of the above phenotype by searching for selection signatures. An F_{ST}-outlier approach was adopted, as implemented in the software BayeScan, to a total of 22 pair-wise breed comparisons. In each breed comparison, any of the selected breeds displaying the 'diluted-grey phenotype' was contrasted with the same cattle breed showing a 'non diluted-grey phenotype' selected as external reference. In total, 3 breeds were adopted here as external references (Holstein, Angus and Charolais), for a sum of 66 pair-wise breed comparisons. Preliminary analysis of results highlighted the presence of several signals of differential selection, some of them harbouring genes related to pigmentation biology. In order to identify the most robust signals, only those observed in at least seven pair-wise breed comparisons for all the 3 external reference breeds, were further considered. Among them, an interesting signal was observed at chromosome 14 (intergenic SNP BTB-00557532). The window of ±250 kb up- and down-stream the SNP harbours nine genes (FAM110B, LOC101902490, UBXN2B, CYP7A1, TRNAG-CCC, LOC112449629, SDCBP, LOC112449508, NSMAF). Out of them, FAM110B (centrosome/spindle pole-associated protein) has been shown to interact with proteins [ADAM, BBS proteins, beta-catenin (Wnt signalling), DVL2 (substrate for HECW1), XRCC5 and XRCC6 (UV-induced DNA damage repair)] known to be involved in pigment biology. SDCBP has been shown to participate in melanin transport. UBXN2B contain a domain peculiar of proteins involved in ubiquitination, a process involved in tyrosinase (melanin precursor) degradation. Several inherited hypopigmentary diseases involve aberrant processing and/or trafficking of tyrosinase and its subsequent degradation. Interestingly, additional ubiquitination or de-ubiquitination signals were detected in this study (HECW1, RNF111, MINDY2) that may be related to the diluted-grey phenotype in cattle.

LIVESTOCK SYSTEMS – NEW EMERGING TECHNOLOGIES IN ANIMAL SCIENCE I

O044

How to measure milking efficiency in dairy cattle farms?

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Labour associated with milking remains a significant constraint on dairy farms; in fact, regardless of herd size, milking is the most time-consuming farm task.

The aim of this study was to compare different evaluation methods of milking timing in intensive dairy farms in order to identify the critical points of milking. The eight studied dairy farms were located in Lombardy (Italy) and were characterised by different milking routine and parlour size. Farms (average herd size 138 ± 61.8 milking cows) were visited twice a year (spring and summer). Three farms milked their herds three times a day, while the remaining five milked twice a day. The average milking parlour size was 9 ± 2 milking units/row. Electronic milk flow metres (Lactocorder®), video recordings and milking vacuum data loggers (Vadia®) were used to detect the time dedicated to each operation in the milking routine as well as to each milking phase. Data were analysed by ANOVA using the GLM procedure in SAS 9.4 (SAS Institute, Cary, NC, USA). The results showed an average milk production of 13.9 ± 2.53 kg/head per milking, without any effect of structural and management characteristics. Peak milk flow was on average 4.11 ± 0.30 kg/min and average milk flow was 2.62 ± 0.20 kg/min. Milking routine was not standardised in all farms: in particular, 25% of farms applied an irregular milking routine and 37.5% applied an incomplete routine. Moreover, the time dedicated to the udder preparation was found to be much longer in the farms adopting an incomplete (2.3 ± 0.14 min) and an irregular routine (5.07 ± 0.14 min) compared to those adopting a regular routine (0.56 ± 0.14 min). As milking parlour size increases, from 10 to 20 milking units/row, an increase in udder time stimulation, idle time between one cow and another, and quantity of milk by single operator was recorded. Idle time was higher as herd size increased (8.08 vs. 13.2 min, in small herds with <120 heads, compared to and large herds with >120 heads, respectively); this result could be due to difficulties in handling cow groups. The use of different evaluation methods, sensors and visual evaluations, is useful in order to analyse milking routine and identify hot spots; in particular, idle time resulted a critical point that need improvement to optimise labour efficiency.

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O045

Precision farming approach to assess grazing dairy cows adaptation to mountain pasture

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This study is part of SMartAlp project, which aims to propose a grazing animals management approach based on integrated application of sensors and advanced technologies. The aim of this contribution is to present the early results concerning the effects of dairy cows adaptation to grazing before transhumance to summer farms.

A group of 16 Italian Simmental dairy cows with previous grazing experience, homogeneous for production (20.3 ± 1.32 kg/d) and lactation stage (212 ± 9.5 d, days in milk), was followed for 10 days before and 10 after its transfer from the permanent farm to a summer farm (Malga Juribello, TN). The permanent farm was free stall and animals received a Total Mixed Ration; in summer farm pasture was integrated with 5 kg/d of compound feed. During the 10 days before the summer pasture, half of the cows spent progressively more time (1, 3, 5, 7, 9 h/d) in a pasture near the barn, while the others were moved to pasture without feeding adaptation. During the experimental period, milk yield and quality, feeding behaviour (measured with RumiWatchSystem, Itin and Hoch GmbH, Liestal, Switzerland) and body condition score were monitored. The variables were analysed with a mixed model for repeated measures that consider the group (adapted vs. non-adapted) and the sampling time as fixed and repeated factor, respectively.

It is commonly known that the absence of an adaptation from a permanent farm diet to a fresh herbage diet can be stressful for high-producing dairy cows, regarding health, behaviour and productive performances. The results of this study do not show significant differences (p>.05) between experimental groups (mean ± SEM) in terms of milk production (20.4 ± 0.70 kg/d), composition (fat, 4.26 ± 0.19%; protein, 3.51 ± 0.04%; lactose, 4.75 ± 0.05%), somatic cells score (2.66 ± 0.36 units), β-Hydroxybutyrate (0.051 ± 0.007 mmol/L). Conversely, milk urea was lower in adapted group (20.9 vs. 24.3 mg/dl; p<.05). Feeding behaviour was similar for both groups (grazing 394 ± 17 min/d; ruminating 316 ± 13 min/d; p>.05). Milk technological traits showed large variability, with a general improvement in cheese making traits after transhumance.

The results suggested that dual purpose breeds are able to adapt to pasture without particular negative effects with accurate management strategies, even without particular training before transhumance.

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O046

Unravelling the relationship between milk quality and rumination by a PLF technology

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Daily rumination time (RT) in cattle has been largely related to feeding behaviour and health status, whereas the relationship between RT and milk composition has not been quite examined. The aim of the study was to unravel the association between test-day (TD) milk records and RT for a period of 10 days before TD (RT_{10}) on a large field dataset.

Data from 558 Italian Friesian lactating cows, reared in two commercial farms and one experimental farm in Lombardy (Italy), were used. All animals were fitted with activity and rumination neck collars (SCR Heatime® HR System, Engineers Ltd., Netanya, Israel) and daily monitored over thirteen months. A dataset made up of official monthly TD milk yield and composition records as well as RT_{10}, was analysed by linear mixed models (lme4 package, R statistical software, CRAN Project).

For statistical analysis, the observations were stratified on the basis of the mean RT_{10} in: low RT ($LRT_{10} \leq 8$ h/d), medium RT (8 h < $MRT_{10} \leq 9$ h/d) and high RT ($HRT_{10} > 9$ h/d). Farms, parity (1, 2 and ≥3 calvings), lactation stages (<60, 61–180 and >181 DIM) and TD data were considered as fixed effects in the model, with cow as a random effect. The best statistical model for each milk trait was selected according to with the minimum Akaike's information criterion. Least squares means and 95% CI were calculated for milk parameters (more than 3400 observations/each). All the considered fixed effects were significant ($p<.001$) in explaining the variability of milk traits.

As expected, cows with HRT_{10} had greater daily milk yield (37.9 vs. 35.4 and 31.5 kg/d), protein (1.27 vs. 1.21 and 1.10 kg/d), lactose (1.85 vs. 1.72 and 1.52 kg/d), and casein yield (0.984 vs. 0.936 and 0.842 kg/d) compared to ones with MRT_{10} and LRT_{10}. Cows with LRT_{10} had higher fat to protein ratio (1.240 vs. 1.152), unsaturated fatty acid (31.5 vs. 29.8%), and monounsaturated fatty acid (26.03 vs. 24.92%) percentages, but lower fat yield (1.35 vs. 1.47 kg/d), and total saturated fatty acid percentage (65.5 vs. 66.5%) compared to animals with HRT_{10}.

Additionally, a classification analysis using SVM (support vector machine, supervised learning) showed higher validation accuracy (69.1%) when milk yield was included as predictor into the analysis, confirming its usefulness in the identification of the two extreme RT clusters.

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O047

Automatic milking system in Mediterranean buffaloes: experimental results and considerations

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The scarce implementation of automatic milking systems (AMS) in buffalo farming despite its primarily benefit, i.e. relief from the milking routine, is mainly due, beside to the initial investment, to concerns about adaptability of buffalo cows to robotic milking and a potential detrimental effect on milk quality. This study aimed to shed light on the operational and functional aspects of AMS in buffaloes and their relationships with milk yield and quality. The research was carried out over 3 years on a dairy buffalo farm equipped with both a traditional tandem parlour and four AMS, each serving about 50 cows. Efficiency of the AMS was evaluated by taking into consideration three issues: (1) yield and quality of milked milk; (2) system capacity, i.e. number of buffaloes that can be milked by AMS per year; (3) number of milking-failure visits, i.e. robot failures to attach milking cluster. The assessment of milk traits was performed by comparing (ANOVA) over the whole lactation milk yield and quality of buffaloes milked with the traditional tandem (TT, 45 cows) or the AMS (45 cows). The cows milked by the AMS had higher number of milkings/days and higher milk yield ($p<.05$) than those milked with TT system, whereas no differences were found for milk composition. The analysis of system capacity was carried out over two years by comparing (ANOVA) the performances of buffaloes milked in AMS serving 50 or 70 cows. Results highlighted that the number of buffaloes per robot may be increased up to 70 without impairing milking performance ($p<.05$), so making it possible to recoup the initial investment earlier. Factors causing milking-failure visits were studied over one year by installing, for each AMS, one camera on the robot arm applying teat cups and another one on the pre-selection gate. The video-recordings were downloaded and analysed remotely every week. The number/year of milking failure visits were 4053 ± 550 (mean ± SD), corresponding to about 10% of AMS working time. About 27% of the failure visits was due to the cow' behaviour, while 72% was related to malfunction of the milking robot (e.g. unexpected size, and/or orientation of one or more teats; obstructed view of the teats). It follows that the operational and functional aspects of AMS in buffaloes can be better adapted to the buffalo's characteristics. Overall results indicate that AMS may be a suitable alternative to conventional milking, offering new management prospects for buffalo farming.

O048

First evaluation of the infra-red thermography as a tool for the detection of mechanical stress on cow udder teat due by milking

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Faults in milking machine (e.g. incorrect vacuum, pulsation setting) or in milking management (e.g. overmilking, incorrect coupling between liner and teat) can mechanically stress the teats that can appear red or purple-coloured after the cluster removal. These colour changes can be associate with a change of blood circulation and fluid retention of the teat tissue. This effect, when occurs, can be at the basis of a teat skin surface temperature (SST) increase that could be detected, automatically and in a contactless way, by an infra-red thermography. In order to test this hypothesis, 40 thermographic images were collected from a herd of Holstein Friesian dairy cows. Thermographic images were acquired during a milking session, 30–60 seconds after the cluster removal. For each cow, a thermographic image of the lateral part of the udder was collected using a commercial infra-red camera positioned at udder level – at a distance of circa 0.5 m – in accordance with the layout of the milking system (a 2 × 12 herringbone parlour). For each thermographic image, and for each couple of udder teats visible in the image, three SSTs were calculated. They were the maximum temperatures measured in three rectangles (of 5 × 25 pixels) manually positioned at the base ($T_{max,B}$), at the centre ($T_{max,C}$) and at one centimetre over the tip of the teat ($T_{max,T}$). Data calculated, were statistically investigated and related to the colour changes of each teat classified by a researcher as pink, red or purple-coloured. Results obtained have shown that mean values of the maximum temperatures, measured at the base, centre and tip of the teat, increase significantly when the colour of the teat changes – after the milking – from pink to red or purple ($T_{max,B}$: 34.19 ± 0.14; 35.11 ± 0.15; 35.68 ± 0.20 – $T_{max,C}$: 35.12 ± 0.15; 36.19 ± 0.14; 36.53 ± 0.16 – $T_{max,T}$: 36.26 ± 0.13; 36.72 ± 0.14 37.31 ± 0.18 – means values and standard errors of teats classified as pink, red and purple). This finding suggests that a colour change of teats – after the cluster removal – might be classified through an infra-red camera, without the need of a visual inspection performed by a technician/milker. Thus, with this technology, it should be possible to develop an automatic system able to detect possible cases of teat stress. A reduction of these negative cases could allow, as a long term effect, a better management of the milking and therefore to increase the general health of a heard of dairy cows.

O049

Milk production ofSarda suckler cows with different calving period

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In the high hill and mountain range of Sardinia the suckler beef industry is traditionally based on the small framed Sarda cow grazing all year around. The milk yield of cows is one of principal factors influencing growth rate and weight at weaning of their calves. With the aim to enhance the knowledge on the performance of calves born during different seasons, the current study evaluated the effect of calving season on milk production of Sarda cows in a rangeland-based beef livestock system. Twenty multiparous Sarda beef cows (8 ± 4.5 years old), randomly selected from the larger herd of Foresta Burgos research farm owned by AGRIS, featured by two distinct calving periods (autumn, group A, 11 animals, average calving date 15/10/2016 ± 15 and winter, group W, 9 animals, average calving date 26/01/2017 ± 10) were used. Milk yield was measured by using the weigh-suckle-weigh technique once a day on five occasions for each group, with average days in milk of 32, 85, 116, 140 and 180 days. The total milk yield (TMY) throughout 180 days of lactation was estimated by fitting Legendre polynomials functions to the test-day records, because of its capacity to fit a great range of atypical lactation curves shapes. The test-day data were analysed by a mixed-effects model with calving period as fixed effect, live weight and age of cows as covariates and the animal as random effect using the lme procedure of R software. TMY data were analysed by a linear model (lm function of R) with calving season as fixed effect and live weight and age of cows as covariates. The group W showed higher average milk yield (5.86 ± 0.30 vs. 4.96 ± 0.27 kg/d, $p = 0.03$) and TMY (1090 ± 63 vs. 846 ± 57 kg, $p = 0.02$) than group A, likely because of pasture availability and quality. To conclude, the study suggests that winter calving outperforms autumn calving in terms of milk performance in cows grazing mountain range of Sardinia. These results, coupled with other basic data (calves growth rate, quantity and quality of grazed pasture, supplementation level), are of utmost importance for optimising economic returns from suckler beef industry.

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ENVIRONMENTAL SUSTAINABILITY – ENVIRONMENTAL IMPACT OF LIVESTOCK I

O050

Growth of *Hermetia illucens* on different food by-products: conversion efficiency and nutritive value. Part I

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Black soldier fly (BSF), *Hermetia illucens* (L.) has been studied for its capability to convert organic waste to high-quality protein, which can be used for animal nutrition. Nutrient composition of growing substrates influences larval growth and composition. Aim of the present study was to evaluate larval growth performances, chemical composition and nutritive value of BSF reared on 4 different substrates. The experimental diets were: commercial diet for laying hens (control), okara, brewer's spent grain mixed with trub and maize distiller's dried grains. Three growing trials were conducted in a climatic chamber at controlled temperature and relative humidity.
Substrates were characterised by a wide variability of chemical composition (% DM). Hen diet was characterised by the highest ash concentration (16.3) as compared to other treatments (on average 4.40). Okara was characterised by the highest value of EE (12.5), maize distillers were intermediate (8.96) and hen and brewer's grains were characterised by the lowest concentration of EE (overall means 6.83). Larval mortality was not affected by the rearing substrates. Larvae fed on hen diet and maize distillers had the highest final weights (2.29 and 1.97 g, respectively for ten larvae). Waste reduction index (WRI takes into account the substrate reduction and the time the larvae require to consume diet) ranged from a minimum of 3.01 in brewer's grain to a maximum of 4.90 in okara. Considering the efficiency conversion index, larvae grown on okara had a higher index (0.36), corresponding to a major attitude to convert the ingested diet.
The average chemical composition of the larvae was: ash 7.92; CP: 50.6; EE 28.4 with differences depending on the diet. Larvae grown on control had a higher ash content compared to other treatments; larvae grown on okara and maize distillers had a higher EE content (on average: 31.2) than larvae reared on brewer's grain and hen diet (on average: 25.6). The saturated fatty acid content (% total fatty acid) was lower for larvae grown on okara. The intestinal digestibility of the protein was higher for the larvae reared on maize distillers, intermediate for those raised on okara and brewer's grain and lower for those on control diet. Larvae of BSF can be a valuable feed for animal nutrition (piglets) and *in vivo* trials are needed to evaluate these alternative feed sources.

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O051

Growth of *Hermetia illucens* on different food by-products: environmental sustainability. Part II

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It is known that the *Hermetia illucens* (Diptera Stratiomyidae) larvae are able to convert food by-products into an interesting rich protein feed. The aim was to evaluate the environmental impact of production of larvae grown on three food by-products (okara, brewer's grain, maize distillers) and on hen diet throughout Life Cycle Assessment approach. The environmental impact of a simulated piglet diet containing insect meal as a partial substitution of protein feed was performed and compared with standard diet with 18% PG (on DM) and contain soybean meal. Larval growth performance was reported on companion abstract. A cradle to insect farm gate LCA was applied, and estimation of environmental impact of food by-products production was given. For maize distiller a value from Ecoinvent database was applied, for hen diet the impacts of each ingredients were evaluated. For okara and brewer's grain, that did not have an economic value and it was impossible to apply an economic impact allocation between product and by-product, the impact values were obtained through a substitution method based on value of soybean meal and protein content. The results showed that feed production activities resulted the main contribute to the environmental impact (until 99% of the impact depending on impact categories and substrates). The climate change for larvae produced (as kg of larvae) on hen diet resulted more impactful (5.76 kg CO_2 eq/kg larvae dry weight) than maize distiller (1.9 kg CO_2 eq/kg larvae), caused by the inclusion of soybean meal in the diet. When an impact was attributed to okara and brewer's grain production throughout a substitution method, the climate change of larvae production grown on these substrates was very high (8.7 and 7.8 kg CO_2 eq/kg larvae dry weight, respectively), so the best sustainable product resulted the larvae grown on maize distiller. The use of high impactful soybean meal, due to land use change, as substitution penalised these last substrates. The inclusion of larvae reared on maize distiller, that showed the best digestibility from *in vitro* test for monogastric, into a piglet diet revealed an environmental benefit with about 10% of reduction of climate change compared with standard diet. This study shows the necessity to deepen the methods to attribute an environmental weight to by-products to better evaluate the environmental advantage of producing insects as novel feed.

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O052

Preliminary results of a systematic review on chemical and nutritional characteristics of *Musca domestica* and *Hermetia illucens* as feed

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Insects have different feeding habits and can exploit several by-products; so, they can be reared under different conditions to optimise their nutritive value. Besides, due to the high variability and heterogeneity of available data, real nutritive properties are not yet well known or not fully exploited. *Hermetia illucens* (Hi) and *Musca domestica* (Md) were recently included in the EU regulation on animal protein (893/2017) and so far, they are used as feedstuff for fish and pet. Anyway, even though they both belong to Diptera order they seem different from a chemical and nutritional point of view. For crude protein content, for instance, the range observed in a review of the available literature is from 40.1 to 74.6% on a dry matter base for Md fed different diets and even to 76.23 on the base of the development phase (adult). The content varies between 21 and about 60% in Hi. While not so big differences come out for fat content, the composition in fatty acids was interestingly divergent. The most expressed fatty acid in Hi is lauric acid (C_{12}) while it is just 0.2% of total fatty acid in Md. The high content of C_{12} strongly impacts the saturated/unsaturated fatty acids ratio. On the other hand, Md shows a not negligible content (16.3%) of palmitoleic acid (C_{16}:1). Both these fatty acids are interesting from a nutritional point of view for humans and animals as well. Hi and MD are poor source of n-3 but this content might be improved by the use of high n-3 feed for insects rearing through the inclusion of fish residues or algae for example. Insects are, in general, supposed to have an amino acids profile similar to fish meal and better than soybean. Md seems higher in the most limiting amino acids (lysine, cysteine, methionine and tryptophan), particularly tryptophan. Tryptophan content in larvae, expressed as percentage of total amino acids, is two and a half times in Md when compared to Hi. Amino acids digestibility is expected high for both the insects. Regarding mineral content, Hi is much richer in Calcium and its Ca/P ratio is much more favourable. In conclusion, the use of different growing substrates and the choice of the insect and its biological phase, along with their combined use in feed formulation could lead to optimised feed as a function of the target animal species.

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O053

Impact of different protein levels and substrates fermentation on the development of the first stages of black soldier fly (*Hermetia illucens*) larvae

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Black soldier fly (BSF) is a dipteran species belonging to Stratiomyidae family. In the last ten years, this species became the object of many researches for the production of protein meal. Moreover, BSF can adapt to different substrates of low added value as organic waste.

Some researchers studied the effect of diet on BSF larvae development from 4-6 days old to adult stage and few data are available on the effects of rearing substrate pre-treatment on larvae development. Studies highlighted as diet can influence the growth performances and chemical composition of BSF larvae and adults, and how a pre-treated substrate can influence the availability of the nutrients of the diet. Instead, few data are available on the impact of the diet on the first days of development of BSF larvae and none on the impact of substrate pre-fermentation.

The aims of this research were to evaluate the impact of different levels of protein and of the pre-treatment of substrates, on the growth performances of the BSF larvae from day zero to day six. Eight dietary treatments were prepared formulating four isoenergetic diets having increasing levels of protein (CP) (13, 16, 19 and 22% of dry matter). Half of each diet (4 treatments) was fermented for 24 hours before its use. Five-hundred milligrams of BSF eggs from the same egg batch were weighed and placed on a net above plastic box containing one of the 8 diets, with 5 replicates per treatment. The plastic boxes were randomly allocated in a climatic chamber with controlled temperature (27.5 ± 1 °C) and humidity (70 ± 5%). The trial was stopped after 6 days. Larvae were then sieved and weighed. All data were subjected to a statistical analysis with SPSS software.

The preliminary results showed as the larval activity impacted on pH values from acidic (5.9–6.3 not fermented; 4.0–4.3 fermented) to basic pH (7.4 to 8.7). The increasing level of protein from 13% to 22% influenced the weights of the larvae from 2.4 ± 0.29 mg (13% CP) to 14.9 ± 0.84 mg (22% CP), moreover the fermentation of the diet influenced the grow performances. The not fermented CP content showed lower weights than fermented CP content, respectively from 2.4 ± 0.29 mg to 4.2 ± 0.30 mg in CP 13% and from 14.87 ± 2.07 mg to 18.32 ± 2.27 mg (CP 22%). The gap was higher in low CP diet compared to high CP content.

O054

Black soldier fly (*Hermetia illucens*) as dietary source for laying quails: live performance and physical traits of eggs

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Insects have been rapidly emerging as innovative feed ingredient to improve the sustainability of the poultry sector and the black soldier fly (BSF) is considered one of the most promising species in this sense. However, at present there is limited knowledge regarding the effect of dietary BSF on live performances and eggs quality of laying quails. Therefore, the present study tested the effect of two dietary inclusion levels with BSF defatted larvae meal on the live performances and physical egg quality of laying quails. A total of $n = 225$ laying quails were weighed and divided into 3 dietary groups, each consisting of five replicated cages (homogeneous for live weight). The control (C) group was fed on a diet with 0% BSF, and the diets of the H1 and H2 groups were formulated including 10% and 15% BSF, respectively. For 35 days, daily, all laid eggs were counted, checked for abnormalities and weighed. Feed intake was recorded weekly. At day 35, quails were weighed, and eggs were collected, marked and analysed. Recorded live performances consisted of initial and final weight of the laying quails, feed efficiency, egg production and egg weight, defected eggs and mortality. Eggs physical traits were egg weight, shape index, surface area, shell weight, thickness and incidence, edible portion, albumen pH, and yolk colour. Data were analysed by a one-way ANOVA with diet as fixed effect. The dietary inclusion of BSF larvae meal did not affect the productive performance of laying quails. Diversely, some physical egg traits were affected by the dietary treatments: quails of the H1 and H2 groups had the highest egg shape index ($p<.01$), shell weight ($p<.01$), thickness ($p<.05$) and thus incidence on the egg weight ($p<.01$). Therefore, the edible portion of the H1 and H2 eggs was lower than that of the C group ($p < 01$). Yolk colour was also affected by the dietary treatment: an increase in the dietary inclusion of BSF larvae meal progressively augmented the yolk colour intensity which was $C < H1 < H2$ ($p<.01$). Based on the present findings, the defatted BSF larvae meal showed to be a promising ingredient for laying quails, up to the 15% dietary inclusion level. The positive effect of the BSF larvae meal on the colour of the yolk deserves further investigation, as it could represent an effective manner to meet consumer demand, i.e. intensely coloured yolks, without using additional pigmenting agents.

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O055

Environmental sustainability assessment: from fruit and vegetable waste to earthworm as feed sources

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Europe's reliance on imported protein to feed livestock, particularly soybean, is inconsistent with sustainability objectives. An increasing dependence on soybean can lead to increasing environmental burden such as deforestation and greenhouse gas (GHG) emissions due to its transport over long distances, which

contributes to climate change. In particular, soybean produced in South America embeds a considerable impact in terms of greenhouse gas emissions related to land use change and indirect land use change. At the same time, food waste has already been recognised as an important global issue. On the environmental point of view, food waste has led to an unnecessary exploitation of natural resources (land, water and fossil energy) and to GHG production. Reduction and recycling of food waste is a key driver towards sustainable productive solutions. Among the possibilities to achieve this goal, this study explores the recycling of fruit and vegetable waste (FVW) discarded directly from juice and ready-to-eat food processing industries, as growth substrate for producing fresh earthworms then processed into dried meal. The dried meal is assumed adoptable for feed alternative purposes, which is the studied scenarios.
The aim of the present study is to assess the environmental impact of the bioconversion of FVW into earthworms dried meal as new feed source adopting the Life Cycle Assessment (LCA) method with an attributional approach and economic allocation. With LCA, other than the most widely known Climate Change expressed as kg CO_2 eq., several indicators of environmental impact are considered, among which particulate matter formation (PM), photochemical ozone formation (POF) and freshwater eutrophication (FE). By means of LCA, the environmental impact of the production of earthworm-dried meal was quantified. The results showed that climate change assessed for 1 kg of earthworm meal is 2.2 kg CO_2 eq. The feed substrate for earthworms consists of FVW that, therefore, is highly valorised respect to wasting. From this result, earthworms' meal can represent a future-generation feedstock with improved sustainability in order to reduce the dependence on imported soybean as protein source.

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ANIMAL PRODUCTS – DAIRY SMALL RUMINANT: FUNCTIONAL FOOD AND MARKETING

O056

Dietary hazelnut skins: effects on milk quality of dairy ewes

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The inclusion of agro-industrial by-products in the ruminant diet could be an alternative to conventional feeds and contribute to improve the quality of meat and milk. Hazelnut skin (HS), the perisperm of the hazelnut kernel, is a by-product of hazelnut manufacturing. Due to the high amount of phenolic compounds, the removal of HS is important to prevent the bitter flavour or the colour reaction in some food applications. Mono and polymeric forms of flavan-3-ols account up to 95% of total polyphenols. Dietary phenolic compounds can affect biohydrogenation (BH) promoting the accumulation of unsaturated fatty acids while reducing saturated fatty (SFA) in ruminant products. Also, healthy fatty acids such as oleic (OA), linoleic (LA) and linolenic acid (LNA) are predominant in the HS. This study evaluated the effect of dietary HS on milk yield and quality in dairy ewes. Nineteen Comisana ewes at 80 ± 10 days in milk were divided into control (C; $n = 9$) and hazelnut (HS; $n = 10$) group, balanced for live weight and milk yield. After the adaptation to the experimental diets, the two groups were kept indoor in sawdust bedded pens. For a 28-day experimental period, each animal received chopped alfalfa hay *ad libitum*. At each of the two-daily milking (7:30 and 17:30), ewes were individually offered 400 g of a conventional concentrate containing 370 g/kg of dried beet pulp (C) or 360 g/kg of HS. Milk yield was recorded, and samples were collected weekly. All data were analysed with a repeated measures ANOVA to account for the effects of diet, sampling time and their interaction. Milk yield was not affected by treatment (p= .525). In contrast, crude protein (p= .045) decreased, while fat tended to increase (p=.082) in the HS milk. Dietary HS affected milk fatty acids, with total SFA, OA and total MUFA (p=.001), greater in HS group. As for individual SFA, C6:0 ÷ C16:0 were lower in HS milk (p=.001). Regarding fatty acids involved in BH, 18:0, OA, t-11 18:1 (VA) and c-9 t-11 18:2 (RA) were greater in HS milk, while LA and LNA were not affected by dietary treatment. This suggests that the impairment of BH due to the polyphenols contained in the HS did not occurred in the way that could have been expected. However, dietary HS improved milk fat composition by reducing unhealthy SFA and favouring the accumulation of OA and healthy fatty acids originating from BH such as VA and RA, with no detrimental effect on milk yield and a limited reduction of the milk protein percentage.

O057

Effect of destoned olive cake on the milk fatty acid profile

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The feed industry is aiming to produce nutraceutical enriched feedstuff to answer to the consumer request for healthier animal products. More and more used source for animal feeding is the use of agro-industrial by-product still rich in primary and secondary metabolites. In the Mediterranean area, the olive oil extraction chain produces significant amounts of olive cakes and vegetation wastewater that in the past represented a cost both for disposal and for the environment. The olive cake was largely studied for its nutritive value, animal intake, degradability, animal feeding behaviour, effect on milk yield and meat quality. The object of the present study was preliminary investigating the effect of fresh destoned olive cake on the milk fatty acid profile.

The trial was carried out in Campania region, south Italy. Ten Italian Simmental cows were assigned to two groups (Control and Olive Cake), homogenous for weight (550 kg), lactation day (60–70 days) and milk yield (28 kg/d). Animals were fed the same diet (hay, silage, concentrate) for 2 weeks of adaptation plus 15 days as experimental period. In the OC group, 15% (DM) of concentrate feed was replaced with fresh destoned olive cake. Milk samples were collected and stored at −20 °C until analyses.

Fatty acids were separated using a Gas Chromatograph fitted with an automatic sampler and equipped with a FID detector. FA peaks were identified by comparison of retention times with those of known mixture of standard FA, then quantified as percentage of total FAME. Thrombogenic Index (TI) and Health Promoting Index (HPI) were calculated. Data were submitted to one-way variance analysis for diet variable (differences considered significant at $p \leq .05$).

The milk fatty acid profile was significantly affected by the feeding treatment. SFA showed a decrease from 65.35 to 57.24% FAME; PUFA increased from 5.03 to 6.32 while SCFA decreased from 6.75 to 4.27 ($p < .001$). Total CLA and Omega-3 increased respectively from 0.98 to 1.41 ($p < .001$) and from 0.81 to 1.10 ($p < .01$). LA and EPA significantly increased from 1.78 to 2.12 and from 0.23 to 0.47 ($p < .001$). HPI significantly increased from 0.46 to 0.68, while TI decreased from 2.72 to 2.07 ($p < .001$). The results showed that the nutritional value of Simmental cow milk was significantly affected by feeding with fresh destoned olive cake, opening interesting and practical perspective to the production chain by naturally enriching drink milk with by-products.

O058

Influence of breeding system on ewe milk quality

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Ewe breeding has relevant socio-economic importance in Central-Southern Italy. In recent decades, many factors led to reduction in farms number and consistency: lack of generational turnover in farm management, reduction of available pastures, productive levels conditioned by hygienic problems, increase of feeding costs, poor development of dairy products. Traditionally, sheep are bred in a semi-extensive system, even if, due to the influence of transalpine model, the current tendency is to establish a more intensive management. This is, where sheep breeds are potentially more productive, but different in quality of dairy products. Aim of this investigation was to evaluate the influence of breeding systems and feeding plans on ewe milk yield and quality. Forty ewes, homogenously selected in four farms (two extensive and two intensive ones), was monitored from lamb weaning up to five lactation months. Every 15 days milk yield was recorded and samples were collected to analyse quality parameters (e.g. chemical composition, cheese-making aptitude, fatty acids profile). Results were statistically influenced by breeds (Sarda and Comisana vs. Lacaune), as well as by management system (use of pasture) and feeding plan (high forage percentage). In particular, milk yield of Lacaune showed the highest values (mean 2520 g/d, $p < .05$). The highest ($p < .05$) percentages of fat (7.98%), protein (7.00%) and casein (5.54%) were found in one of the extensive farms, whereas urea content (40.6 mg/dl, $p < .05$) was the lowest ones. Regarding the cheese-making aptitude, no differences were detected for k_{20} and a_{30} and all the parameters fall within the normal range for this species, excepting r that resulted significantly higher (17.8, $p < .05$) in one the extensive farms, probably due to its lower milk production. Interesting differences also emerged among the management systems concerning milk fatty acid profile. In particular, all the fatty acids with beneficial effects on human health, were significantly ($p < .05$) more represented in fat milk from animals belong to one of the extensive farms (36.2, 30.9 and 5.23% UFA, MUFA and PUFA, respectively), whereas saturated fatty acids were the lowest (60.9%, $p < .05$). Moreover, milk CLA were also significantly higher (mean values: 1.57 vs. 0.38%, $p < .05$) in grazing ewes. These results confirm that feeding plans strongly influence nutritional and technological milk quality. Data concerning blood parameters evaluating animal welfare are still being processed.

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O059

Efficiency in sheep milk production: comparison between extensive and intensive management systems

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The aim of this study was to evaluate the effect of the breeding system (extensive vs. intensive) on performance of Gentile di Puglia sheep breed. The trial, started the last week of April, was carried out on 30 Gentile di Puglia sheep breed in lactation, raised under an extensive system. The animals were divided into 2 groups (15 subjects for each group): one group raised under an extensive system (E) and the second group raised under an intensive system (I). The subjects showed an initial average weight of 42.1 and 41.8 kg for E and I group, respectively. Controls started one week after giving birth and weekly. Group I sheep, after an adaptation period of about 20 days, were fed *ad libitum* with excellent clover hay with addition of barley to reach an average feed concentration of 0.7 units of forage (UF)/kg. On individual milk samples was determined the proximate composition: dry matter, protein (total $N \times 6.38$), ash, fat and lactose. For each subject live weight (LW; kg), $LW^{0.75}$ (kg), daily milk production (kg), composition and energy value of the milk were detected. Then the *Biological Efficiency* (BE) which indicates the productive potential of the animal – g product/kg LW, and the *Zootechnical Efficiency* (ZE) which provides elements on the energy cost for each unit produced – g product/kg $LW^{0.75}$ were calculated. The statistical analysis was performed by ANOVA. E group sheep showed a higher average milk production than I group (1131 vs. 789 g/day/sheep, respectively; $p<.001$); whereas, the fat and protein content and energy content was higher in the milk of the I group sheep (4.53 vs. 4.71% protein, 4.39 vs. 5.60% fat, 858 vs. 981 kcal/kg for E and I group, respectively). BE index was significantly higher in E group sheep than I group ones, both in terms of milk quantity (26.8 vs. 18.4 g/kg LW, respectively) and energy produced (23 vs. 18.1 kcal/kg LW, respectively; $p<.001$). Performance in relation to metabolic weight (ZE) were: milk 68.2 vs. 47.2 g/kg MW ($p<.001$) and energy 58.6 vs. 46.3 kcal/ $LW^{0.75}$ ($p<.001$) for E and I group, respectively. Every kg of metabolic weight of E group sheep produced 26.56% more energy compared to the sheep of I group. So, to get one kcal in milk are consumed 1.94 kcal in E group sheep and 2.19 kcal in I group ones. The results of this study showed that the efficiency of the extensive system was higher than that of the intensive system.

O060

Effect of forage quality on milk production of Sarda dairy ewes

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The production of high-quality forages represents not only a suitable strategy to increase animal productivity but also a way to reduce enteric emissions and feeding costs per kg of produced milk. Thus, it can be considered among the best practices for GHG mitigation. For this reason, this study evaluated the effect of two diets containing hay of different quality on milk production of lactating ewes.

Twenty-four Sarda dairy ewes in late-lactation (184 ± 15 DIM; mean ± standard deviation) were allocated to two experimental groups, homogeneous for milk production (0.83 ± 0.19 kg/d), body weight (44.79 ± 5.73 kg) and body condition score (BCS 2.84 ± 0.16). The groups were fed 2 diets differing for the 2 hays offered, characterised by high (H-NDF: 66.1% NDF; 7.6% CP, DM basis) and low (L-NDF: 54.3% NDF; 11.4% CP, DM basis) NDF content, respectively. The hay was offered once a day in the morning and was available *ad libitum*. Both groups received the same concentrate supplementation (soy meal, corn and flaked corn, 0.27, 0.20 and 0.17 kg/d DM basis, respectively). The ewes were kept indoor in individual boxes and fed individually for 5 weeks. Animals were milked at 7 am and 4 pm; two times a week individual milk yield was measured and sampled for chemical analyses. Data were analysed with the PROC MIXED of SAS including the dietary treatment, the sampling date and their interaction as fixed effects, and the animal as random effect.

The intake of hay was higher (+85%) in the L-NDF than H-NDF group (1.07 vs. 0.59 kg/d, respectively) ($p<.001$). This was due to the higher quality of the hay fed to this group of sheep. Milk production was significantly affected by diet × sampling interaction ($p<.01$). The L-NDF ewes exhibited a greater persistence of milk production than those of the H-NDF group, which had a rapid reduction of milk production already since the first part of the trial. On average, the L-group produced 35% more milk than the H-group (0.72 vs. 0.53 kg/d per head, respectively). Milk composition was not affected by the dietary treatment. Milk urea concentration was affected by the diet × sampling interaction and it was higher in the H-NDF than L-NDF groups (32.97 vs. 25.63 mg/dl, respectively) ($p<.001$). The results of this work emphasise the positive effects of high-quality forages on sheep milk production.

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O061

Influence of different livestock production systems in caprine milk and cheese odd and branched chain fatty acids (OBCFA) composition

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Odd and branched chain fatty acids (OBCFA) are produced by rumen biochemical mechanisms and by mammary lipogenic activity of ruminants. Although OBCFA represent a lesser component of milk, they are recognised as important bioactive components because of their positive role in gastrointestinal microbial ecology and their potential anti-cancer activity. In this study, we investigated the influence of the livestock production system on milk and cheese fatty acid composition of three different small goat farms: a conventional Saanen farm (CS) characterised by local ryegrass hay based diet, a conventional Alpine farm (CA) with dairy goats fed with alfalfa hay based diet and an organic Alpine farm (OA) characterised by grazing. Bulk raw milk samples were twice a month sampled from March to October and fresh cheese samples were monthly collected in the same period. Q13 Fat was extracted according to Folch (1957) and fatty acids methyl esters were prepared by base catalysed methanolysis of glycerides Q14 as described by Christie (1982) and analysed by gas-chromatography. Data were analysed by one way ANOVA and multivariate statistic (PCA). Results showed significant differences in the composition of fatty acids in goat milk and cheese. Particularly, OBCFA showed a satisfactory discriminant capacity for the livestock production system. Samples coming from the conventional Saanen farm (CS) resulted enriched in SFA and PUFA, whereas OA and CA milk and cheese were higher in MUFA and OBCFA. The highest OBCFA value (4.66%) was detected in milk of OA farm goats fed with the highest quantity of fibre, incoming from fresh grass and forage. On the contrary, Saanen goats, not grazing and fed with a higher quantity of concentrate, produced milk lacking in OBCFA (3.92%). The highest values of C13:0 and C15:0 resulted in OA milk (0.13% and 1.14%, respectively) at different level, if compared to CA and CS farm. C17:0 values resulted higher in OA (0.70%) and in CA (0.75%) than in CS (0.57%); we obtained similar results for C17:1, higher in OA (0.33%) and CA (0.32%) milk, than in CS (0.22%) milk. Cheese samples mirrored the same trend for OBCFA values observed in milk, with higher values in OA and CA than in CS. Considering the whole OBCFA, there were no statistically significant differences between OA and CA samples (4.95% and 4.82%, respectively) in fresh cheese. However, these samples showed a difference if compared to cheese samples coming from the conventional farm (4.14%).

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O062

Is pink discolouration defect in Pecorino cheese due to *Thermus thermophilus* presence?

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Pink discolouration defect can be observed in different varieties of ripened cheese and can lead to economic losses for the producers due to the impossibility to sell the defected cheese. Little knowledge is available on the causes of this defect; however, *Thermus thermophilus* has been recently identified as a possible responsible. Further understanding of the factors involved in the development of pink discolouration in cheese can give economic benefits.

The goal of this study was to characterise the microbial community in Pecorino cheese with pink discolouration defect.

DNA was extracted from cheese samples from two different lots (lot1, with not defected cheeses – 4 samples from 2 cheese units – CU; lot2 with both defected – 8 samples from 2 CU – and not defected cheeses – 4 samples from 2 CU). The V5-V6 hypervariable regions of the bacterial 16S rRNA gene were PCR amplified. Amplicons were sequenced by Illumina MiSeq. Bioinformatic elaborations were performed by DADA2. Forward and reverse reads were truncated at 200 and 160 bases, respectively. Primers' sequences were removed and low-quality reads (i.e. reads with expected errors higher than 0.5 and with Ns) were discarded. Error rates were estimated and used to infer the Amplicon Sequence Variants. The taxonomic classification was performed using the RDP database (confidence 80%).

The most abundant bacteria belonged to the genera that were found into the starter culture (i.e. *Lactobacillus* ranged from ∑2% to ∑10%, *Lactococcus* ranged from ∑2% to ∑7%, *Streptococcus* ranged from ∑77% to ∑91%). No sequences belonging to the genus *Thermus* were detected. A permutational multivariate analysis of variance (PERMANOVA) based on Hellinger transformed genus relative abundance data showed that the microbial

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communities in the cheeses from lot1 and lot2 were different (p=.001). A Kruskal–Wallis test was performed to identify the genera with a different relative abundance in lot1 and lot2. The relative abundance of the genus *Streptococcus* was higher in the cheeses from lot1, while the relative abundance of the genera *Escherichia/Shigella*, *Lactobacillus* and *Propionibacterium* was higher in the cheeses from lot2 (p<.05). The presence of *T. thermophilus* was not observed suggesting that this bacterium was not responsible for the pink discolouration defect. Further analyses are needed to verify the potential correlation between the detected populations and the pink discolouration of Pecorino cheese.

O063

The answer of the consumers to a cheese with natural vegetable rennet

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In the recent years, innovation transfer in the agricultural sector has been increasingly led by user-centred research systems, focussing on a new approach where innovation is the combined outcome of technological, social, economic and institutional change. Research has clearly shown that successful innovation requires consumers to be included in innovation processes. On these bases, the concept of social innovation comes out and promotes the active participation of the end users in every part of the innovation process, via built local connections and common learning culture.

Recently, the cheese market showed an increasing demand for animal rennet free cheeses, for ethical (vegetarian) or religious reasons. In order to satisfy the demand, the Research Centre for Animal production and Aquaculture in Bella Muro set up an innovative vegetable rennet, starting from local resources, the artichoke cultivar *Bianco di Pertosa*.

Soft cow milk cheeses were produced with this rennet from 2011 to 2013. The caciotta cheeses (28 days ripened) were offered for consumer tests during special events organised within the study (December 2011 and February 2012) and participating to large public events, such as ‘Vinitaly’, ‘Salone del Gusto’ and ‘Forum PA’ (2012 and 2013). After a short training on the cheese sensory evaluation, consumers received an acceptability inquiry form and an anonymous survey on their profile (gender, age, title/study and job) and cheese consume behaviour. The acceptability test was structured in a 1–10 point scale and free space for the comments. The results were elaborated and used, together with the comments, to identify possible corrective actions to improve the product. In particular, the cheesemaking process was adjusted in order to amend the characteristics less appreciated by the consumers who expressed acceptability score ≤6. The most significant comments such as ‘Complex taste but I did not appreciate very much the bitter note’ and ‘Too fresh’, contributed to lead to cheeses with lower bitter taste but creamier texture.

Over six consumer tests, the acceptability score increased progressively from 5.73 in 2011 to 7.91 in 2013 (38.0%), with a R^2=0.88. The percentage of forms with score <6 decreased from 41.9% to 8.8%.

The results confirmed the role played by the involvement of consumers in developing innovative products. The approach seemed to be very powerful also to reshape a food chain that is constrained by both healthy and environmental issues.

Acknowledgements

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O064

Socio-economic analysis and current problems of Saanen goat farms in Turkey: a case of Çanakkale province

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Goats still play an important socio-economic role in many rural areas in Turkey. The aim of this study is to evaluate the general structure and socio-economic aspects of the Saanen goat farms in Turkey. The fieldwork was carried out in the Çanakkale province, where is located in the west of Turkey and has a very intensive breeding and the largest number of Saanen goats (25.51%) in Turkey. Data has been collected by using a questionnaire from 92 farms, selected by using ‘Simple Random Sampling’. The farms are grouped according to number of animals as small-scale (25–75 heads), medium-scale (76–150 heads) and large-scale (151 and overheads). Variance Analysis (ANOVA) and χ^2 test were used to determine whether the difference between group averages was significant or not significant. As a result of the analysis, 91.3% of the farmers were male. Educational level of the farmers has been determined as primary (63%), secondary and high school (14%) and university (9%) respectively. It has been reported that 78% of the farms have another activity in order to make income besides goat breeding. Type of labour in the farms was mostly family labour (79%). Only 17% of the farms could afford a shepherd as a foreign labour in their farms and labour type difference between three scales of farms was significant ($F = 10.882$; $p<.05$). About 66% of the farms were a member of cooperatives and the most important reason to become a member was to decrease input cost (69.6%). Average milk production of farms has been determined as 35079.30 kg and average daily milk yield per animal was 1.95 kg. Most farmers (59.8%) have remarked that the low producer milk price was the most common problem seen in the dairy goat sector. It has been observed that even the farmers tend to raise Saanen goat breed to provide more milk yield, due to intense existence of small scale and scattered structure farms they face many problems such as difficulties in milk products marketing, some diseases which make important economic losses in the farms, insufficient subsidies and high input costs.

ANIMAL BREEDING AND GENOMICS – NEW BREEDING TECHNOLOGIES I

O065

Comparative analysis of *Semimembranosus* muscle transcriptome in Italian Large White pigs with high and low intramuscular fat content

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The implementation of high-throughput technology such as RNA-sequencing has become an effective tool in livestock sciences to disentangle gene expression networks involved in complex traits, such as meat quality. Pig meat quality depends on several organoleptic and nutritional characteristics of the cuts and is also influenced by the amount of intramuscular fat (IMF). Several studies showed that IMF content positively affects the flavour, tenderness and the technological characteristics of fresh and seasoned meat products. Therefore, the purpose of the present work was to analyse *Semimembranosus* muscle (SM) transcriptome of purebred pigs in order to find the genes whose expression is related to changes in IMF deposition. The used samples belonged to two groups of Italian Large White pigs, chosen among a population of 950 animals for their extreme and divergent IMF content. The groups were composed of six animals each (three gilts and three barrows) showing divergent contents of SM IMF (an average value of 0.67 ± 0.09% in low IMF group vs. the 6.81 ± 1.17% in high IMF samples). Paired-end RNA sequences were aligned to *Sus scrofa* genome assembly 11.1 and gene counts were used to perform the subsequent analyses using WGCNA and DeSeq2 packages. The lists of genes whose expression level in the studied samples was most significantly associated with IMF deposition were submitted to DAVID on-line tool for functional annotation analysis. Four WGCNA modules were significantly associated with IMF deposition, with the most significant one being the grey60 module (p=.003). The functional analysis of the genes contained in this module showed the strongest significant annotation cluster comprising genes belonging to primary cilium (p=.004), intraciliary transport (p=.005) and to cell projection (p=.02). DeSeq2 analysis identified 58 differentially expressed genes (DEG) between the two studied groups of samples with a Fold Change >1.5 and an adjusted $p<.05$. Other DEG were involved in the regulation of differentiating cells, in the control of RNA-processing, and in G-protein and ERK signalling pathways. Interestingly, most of the DEG found with DeSeq2 were also clustered in the most significant gene modules highlighted by WGCNA. The combined use of different statistical tools aimed at identifying both DEG and gene co-expression networks could provide the potential for the identification of genes acting as central players in fat-related traits.

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O066

Runs of homozygosity of Simmental bulls raised in five European countries

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Simmental is a worldwide spread cattle breed, even if it is raised with different purposes according to the country: e.g. in USA for meat production, while in Europe is recognised as a dual-purpose. Moreover, also in Europe, each national association establish the weight of each trait on the breeding programme. Such a difference in the breeding purpose, and therefore on the matings planning, can be reflected in genome differences among animals coming from different countries. In order to highlight these differences, runs of homozygosity (ROH) were investigated in Simmental bulls coming from Austria (AT, 351 animals), Switzerland (CH, 215), Czech Republic (CZ, 248), Germany (DE, 550) and Italy (IT, 2481). All bulls were genotyped with a customised chip starting from the medium density Illumina chip. Markers not matching the following parameters were discarded: mapped in BTA 3.1 on autosomal chromosomes, minor allele frequency >2%, call rate >97.5%, Hardy Weinberg equilibrium *p*-value>.01. After quality control, a total of 3845 bulls and 40,099 SNPs were used for further analyses. ROH were calculated through Zanardi pipeline and defined as 15 consecutive homozygotes, with no missing or heterozygotes, with a minimum length of 1 Mb. ROH inbreeding coefficient (F_{ROH}) was calculated as well. A total of 287,524 ROHs were identified. The average ROH/animal (overall of 77.8 ± 20.7) ranged from 52.2 (CZ) to 109.9 (CH). CZ and CH bulls showed also the lowest (119.2 Mb) and highest (324.9 Mb) genome length covered by ROH, respectively. The overall average F_{ROH} value was 7.5 ± 2.9%, with the lowest registered for CZ (4.6%) and the highest for CH (12.6%). Bulls raised in AT, DE and IT showed very close results for all considered ROH features. Analysing ROH in the European Simmental bull population some differences were highlighted, with CH showing features closer to selected breeds (e.g. Holstein and Brown Swiss) and CZ closer to small indigenous breeds.

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O067

Efficiency of imputation for genomic selection in rabbits

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Genomic selection could be applied in many sectors, including rabbit industry, using low SNP density arrays and an effective imputation strategy. The aim of this study was to unravel the best trade-off between imputation strategies and genotyping cost for litter size in a simulated rabbit population. Moreover, the accuracy of imputation on the reliability of estimated genomic breeding value (gEBV) was also examined. A rabbit population accounting 200 animals were obtained by simulation in AlphaSim software. Selection was carried out in two steps, i.e. (i) twenty generations were selected by BLUP followed by selection of further three generation by GBLUP; (ii) an additional last generation was simulated from random mating of all 200 individual to expand the population size to 1000 animal to measure the accuracy of imputation. Thirty-five replicates were examined and six different scenarios (S1–S6) were created considering the use of different SNP panels in the training population (i.e. the last two generations selected by GBLUP) and in the last generation of 1000 animals. In S1 and S2, all parents and grandparents were genotyped with high-density arrays (100K). In S3, females (dams and grand dams) were genotyped with medium density array (3K SNPs). In S4 and S5, dams and grand dams were genotyped with low-density arrays (300 SNPs) and for S6, only males were genotyped (dams and grand dams were not genotype). Individuals of the last generation were always genotyped with low density, and for S2 and S5, only half of individuals were genotyped. The accuracy of gEBVs obtained through BLUPF90 software implementing a single step approach was obtained for imputed genotypes considering only S1, S3 and S6 scenarios. Highest imputation accuracies were observed in S1 (0.93). Otherwise, S4 and S5 achieved the lowest accuracy (0.85 and 0.83, respectively). The best trade-off between cost of genotyping and accuracy was reached in S3, where a 0.91 accuracy was obtained in combination with a global cost near to 30,000 euros, i.e. about 6000 euro lower than in S1. The results of this study show that is possible to implement imputation as a strategy to reduce the genotyping costs having a good imputation accuracy. The accuracy of gEVB was greater in scenarios with greater imputation accuracy. However, this relationship could depend on the distribution of QTL and SNP across genome, so further studies are needed to address this relationship.

O068

Assessment of a fertility and a maternal behaviour index: effect of kindling order in three genetic lines of rabbits does

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The maintenance of an adequate level of fertility is of fundamental importance in rabbit breeding. In particular, animals with high fertility allows for improving functional welfare of the does and their economic sustainability. The selection for reproductive characters is difficult and the low genetic gain obtainable is due to the low inheritability of reproductive traits. Commercial production of rabbit meat is generally based on 2 or 3-way selected lines. The selection of the female lines generally includes characters such as the size and weight of the litter at birth and after 4 weeks also in relation to the female's maternal instinct. The

selection scheme applied by ANCI (Associazione Nazionale Coniglicoltori Italiani) Genetic Centre is based on well-established scheme of genetic improvement for 3 rabbit lines of the Genealogical Book: Bianca Italiana (BI), Macchiata Italiana (MI) and Argentata Italiana (AI). The aim of this work was the assessment of a Female Fertility (IFF) and Maternal Behaviour Index (IMB) of the ANCI genetic lines. For this purpose, 138,374 records were made. Only phenotypic variability of the different traits was evaluated. The fertility and litter size indices were combined into IFF (fertility per litter size) whereas IMB was the combination of litter weight at 19 days (milking ability) per weaned rabbits (mortality). IFF indicates the expected number of live-born per insemination, whereas IMB states the capacity of the does to have low mortality and very high milking ability. The effect of kindling order (0= nulliparous, 1 primiparous and 2 pluriparous) and genetic strain (BI, MI and AI) was evaluated for the different reproductive characters. The fertility rate was higher in nulliparous and multiparous does. Pluriparous also had higher litter size at birth and at weaning whereas primiparous had the lower fertility rate. Concerning the genetic strain, BI and MI had similar productive performances, whereas AI showed the worst. The IFF showed the lower value in primiparous and the best value for pluriparous and in BI strain. The same for the IMB, which was better in pluriparous and in BI. These indices can be used alone or included in the selection index applied to the centre's genetic lines. Further studies are needed to establish selection programmes aimed at obtaining the best genotypes in terms of reproductive traits.

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O069

Using identity-by-descent probability to impute whole genome sequence variants in a nucleus flock

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Genotype imputation is a process where a reference sample of a population, fully genotyped at a higher density panel, is used to statistically infer untyped genotypes in a target sample of the same population where only loci from a lower density panel are directly genotyped. Thus, genotype imputation allows exploiting an increasing number of loci for genome-wide association studies and genomic selection limiting genotyping costs.

Here, we propose a Bayesian procedure to infer the gametic phases in a population where only a small sample is fully genotyped for a dense marker panel. This procedure uses as prior information reference sample genotypes at loci to impute and the locus-wide identity-by-descent probability between all the gametes of the population estimated with a sparser markers panel.

The proposed procedure was applied to a nucleus flock of Sarda sheep (NF). NF was set up in 2000 and consists yearly of about 1000 ewes. Replacements (25%) are generated by mating ewes of NF with rams from the Sarda Herd Book in order to represent the genetic variability of the selected population. The gametes of 4000 ewes generated until 2015 are replicates of the base gametes carried by the ~1000 first generation dams and ~160 rams. All the ewes and rams were genotyped with the OvineSNP50 Beadchip (Illumina) and their gametic phases were reconstructed by linkage analysis. Identity-by-descent probabilities among base gametes and between base gametes and their replicates in NF ewes were estimated at each SNP position. A reference sample of 45 individuals (22 ewes and 23 rams) were Whole-Genome resequenced and used for imputation.

The proposed procedure was applied to four genomic regions on OAR 4, 6, 20 and 22 (total length of 65 Mb) associated with traits of economic interest. A total of 1,443,878 polymorphisms from Whole-Genome Resequencing were imputed trough identity-by-descent probabilities at 1190 loci from OvineSNP50 Beadchip.

Considering only the base gametes, the precision of imputation of polymorphisms from resequencing, measured as the variability of the probabilities of assigned alleles, was on average 0.95 ± 0.04 and the correlation between the observed Beadchip gametic phases and gametic phases inferred by imputation were 0.98 ± 0.02.

We concluded that the proposed imputation method is effective and robust mainly in population where identity-by-descent probabilities between gametes can be accurately estimated.

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O070

Biodivsel: functional annotation of target regions based on whole genome resequencing of Sarda sheep

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The Biodivsel project aims at detecting functional genetic variants valuable for selection by exploring genomic regions associated with traits of economic interest in Sarda dairy sheep. Analyses were carried out on the whole genome sequence of 24 Sarda individuals (target coverage of 12X/animal) and 21 Sarda rams (target coverage of 30X/animal) chosen to represent the genetic variability of the breed. Paired-end reads were mapped to the latest sheep reference sequence Oar_v4.0 (2015) using the BWA 0.7.15 aligner and variant call was performed with GATK. Three regions spanning ~10 Mb and surrounding locations significantly associated to milk fat content (FC), milk protein content (PC), and CLA and Vaccenic acid content ratio in milk fat were investigated on chromosomes 4 (7–18 Mb), 6 (80–90 Mb) and 22 (15–25 Mb), respectively. Additionally, a region on OAR20 (16–50 Mb) harbouring locations associated with nematodes resistance (FEC) and paratuberculosis (PTB) was also investigated. SnpEff v4.3t was used to annotate the genomic context of the identified variants. Candidate genes for each trait were inferred through KEGG enrichment analysis, after extending the background genes' panel with those genes significantly associated with each of the analysed trait according to literature. We identified 769 protein-coding genes and 1,543,664 variants (of which 1,231,096 were SNPs and 312,568 were INDELs), with a rate of one variant every 42 bases. Six genes on chromosome 4 were identified in pathways related to FC (KRIT1, GNG11, COL1A2, MIOS, RPA3, ICA1), four on chromosome 6 to PC (CSN1S1, CSN1S2, CSN2, CSN3), five on chromosome 22 to CLA (FGF8, NFKB2, SCD, WNT8B, LOC101120535), and 31 on chromosome 20 to FEC and/or PTB (DQA, IL17F, ILF17A, TNF, among others). Ninety-three variants with inferred 'high' impact (mainly frameshift variants) and 354 with 'moderate' impact (mainly missense variants) were identified in the target genes. Concerning the most evident candidate genes, nine missense variants were identified in the four caseins genes, 88 in DQA, two in IL17F and one in IL17A and TNF, respectively. Only low- and moderate-effect variants were found in the SCD gene.

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O071

Gene structure analysis of donkey oxytocin-neurophysin I (*OXT*) gene and genetic variability detection

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Oxytocin is a neurohypophysial peptide released into general circulation from posterior pituitary gland. It is involved in different physiological roles, including milk ejection from the mammary gland, stimulation of uterine smooth muscle contraction during labour and affects cognitive processes, tolerance, adaptation and complex sexual and maternal behaviour. The main objective of the present research was to determine the complete donkey oxytocin-neurophysin I encoding gene (*OXT*) sequences and to detect genetic diversity at this *locus*. Using the genomic DNA as template, we sequenced and compared the whole *OXT* gene of 5 Italian donkeys: 2 Amiatine, 2 Ragusana and 1 Grigio Siciliano donkeys. On average the gene extends over 946 bp, composed of 517 bp of exonic regions and 429 bp of intronic regions, with an A/T and G/C content of about 27% and 73%, respectively. Furthermore, the 5' flanking region (876 bp) was sequenced. The gene contains only three exons, ranging in size from 202 bp (exons 2) to 155 bp (exon 3), and two introns of 315 bp (intron 1) and 114 bp (intron 2). The first exon encodes a peptide leader (19 amino acids residues encoded by the nucleotides from 41 and 97), the nonapeptide hormone (from nucleotide 98 to 124), the tripeptide processing signal (GKR) (from nucleotides 125 and 133) and the first 9 of 94 residues of neurophysin I; the second exon encodes the central part of neurophysin I (67 aa), while the third exon encodes the COOH–terminal region of neurophysin I (18 aa). Among the different species of animals of species of livestock animals, the *OXT* donkey gene sequence showed the highest degree of similarity with the horse (98%) vs. ruminants (75%) and camelids (76%). The comparison of the sequences obtained allowed the identification of 5 SNPs: g.263A>G, g.334A>G, g.436T>C and g.304A>C in intron 1 and g.124G>A in promoter region. This last polymorphic site falls into a potential binding site for the transcription factor GATA-1 (www.generegulation.com). With this study, we provide the first contribution to the characterisation of the genomic sequence of the *OXT* gene and first examples of markers found at this *locus* in donkey. The detected polymorphisms represent a good opportunity to carry out studies focussed on the identification of significant association with the physiological processes controlled by this hormone as already performed in other species.

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O072

Endurance exercise shifts mature mRNA towards introns: does stress enhance genome plasticity?

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Physical exercise is recognised as stress and horse is probably the best animal model for investigating the related genomic response due to its natural aptitude for athletic performance and the homogeneity of its genetic background. The aim of this work is to dissect the modulation of Peripheral Blood Mononuclear Cells (PBMCs) transcription after exercise with a time course experiment that explores not only the expression of annotated genes but also the unexplored regions related to introns and intergenic portions. Illumina sequences from PBMCs of five 3 years old racehorses collected at rest (T0) and after a 2000-metre competition (T1) were produced. After mapping with Hisat2, reads counts evaluation was performed using the ReadCounter software that allowed us to differentiate reads falling within the exons and introns. Apart from DGE ascertainment between the two-time points ('exon': 648; 'intron': 1306), the complexity of transcription for alternative transcripts was identified using DEXseq. We noted a transcription shift from the coding to the non-coding regions. We further investigated the possible causes of this phenomenon focussing on repeats using a differential expression approach finding a strong general up-regulation of repetitive elements such as LINE (46.7%) and SINE (23.8%). Aware that repetitive elements modulation is associated to the 'exonization' – the recruitment of repeats that act with regulatory functions – we tried to explore if DGE belonged to some GO terms linked with the alternative splicing annotation. Many of them ('exon': 14; 'intron': 34) were associated to terms like 'RNA splicing', 'mRNA splicing via spliceosome' and 'RNA splicing regulation', suggesting that there might be an active regulation of this transcriptional shift. Our study could represent, thanks to the innovative approach, a model of transcriptomic investigation of stress on somatic cells.

LIVESTOCK SYSTEMS – NEW PROTEIN SOURCES IN AQUACULTURE

O073

Millennials' attitudes towards insects as feed for sustainable fish

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The decrease in the availability of fishmeal and fish oil prompted the detection of sustainable alternatives for aquaculture feeds. Insects, apparently part of the natural diet of fish, leave a small ecological footprint, have a limited need for arable land and seems to be a good substitute. Recently, studies of the replacement of fishmeal with insects in the diet of fish have emerged and the promising results have encouraged further research. Insects, in line with these researches, are considered a potential ingredient of feed and, due to their 'growing ability', product of an environmental-friendly farming process. Their production can be renewable and cost-effective since they can be produced also on waste. Despite the benefits, consumers in Western countries generally reject the practice of accepting insects as food and feed for animals. From an evolutionary point of view, when a new food product is introduced into a culture, it generally induces feelings of fear and refusal called 'neophobia'. The aim of this study was to investigate the attitude of a sample of Italian millennials towards insects, suitable candidates for providing sustainable proteins, as part of fish diets. Using data from a cross-sectional survey, this study proposed a theoretical model with the aim to analyse the relationships between latent constructs in consumer behaviour, investigating the applicability of the Theory of Planned Behaviour (TPB). A structured questionnaire, based on the TPB existing literature, was randomly administered to university students in the North East of Italy. A Structural Equation Model (SEM) was implemented to test the hypotheses and model fit. The results of this study allowed us to support the following model hypothesis: (a) millennials attitude towards insects as feed for fish had a mediation role in the relationship between subjective norms and buying intention of fish farmed using insect meals in their diets; (b) beliefs that eating fishes farmed using insect meals has positive effects on health and the environment, significantly affected attitudes and intention; (c) information about the environmental and nutritional benefits of introducing insect meals as feed for fishes had a positive effect on their acceptability. The value of this research lies in an in-depth exploration of direct and mediating effects of millennials' attitude towards insects, in order to efficiently promote acceptance and involvement among them.

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O074

Molecular-based identification of insect ingredients in animal feed

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Since January 1st 2018 seven different insects species have been allowed to be used in aquafeed (Regulation (EU) 2017/893) in the European Union. *Tenebrio molitor* and *Hermetia illucens* are the most attractive species that could suitably be used in aquafeed for their availability and market price. Taking into consideration that no official methods for insect detection in feed are available yet and the microscopy analysis have not a good accuracy, therefore, sensitive detection methods are urgently required to monitor compliance with the law since no threshold is in place. A nucleic acid-based approach like PCR is still considered straightforward method for target detection; for this reason, it was decided to design suitable primers against a conservative mitochondrial gene for the detection of the two insect species and consequently, the primers efficiency for the target and their cross-reactivity were evaluated using qPCR. DNA samples used in the qPCR were extracted from different untreated insects, insect meals and aquafeed with different percentages of target insect meal inclusion and were used to evaluate the accuracy and sensitivity of the analysis.

Results show that the designed pairs of primers do not cross-react with the no target samples. *Hermetia* DNA at 10ng/µl in purity gave a reproducible signal about Ct =23 and the *Tenebrio* DNA at 10 ng/µl gave a signal at Ct =20. The target species were detected in aquafeed with different insect inclusion and the qPCR analysis output gives different values of Ct due to the different level of inclusion, lower with high inclusion, higher with low inclusion level for both the target species.

The qPCR method has confirmed its accuracy and suitability for the detection of these ingredients in animal feeds within a range of 0.03 to 0.5 g/g ($r = 0.98$, $n = 10$). Moreover, these primers that revealed a good efficiency and affinity, are going to be used to implement a portable biosensor for field analysis based on a fluorescence detection technique as a faster and easier system of detection that could be useful at industrial level.

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O075

Dietary *Tenebrio molitor* larva meal inclusion in rainbow trout diets: preliminary results about animal and gut health implications

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The European Commission (Regulation No 2017/893) recently authorised the use of insect proteins in the aquaculture sector. Positive results in terms of animal (histological features) and gut (mucosal morphology and mucin composition) health have already been obtained in rainbow trout fed *Hermetia illucens*-based diets, but no such data in relation to *Tenebrio molitor* utilisation are currently available. The aim of this study was to investigate the effects of high protein *Tenebrio molitor* larva meal (TM) on histological traits of gut, liver, kidneys and spleen, and gut morphology of rainbow trout. A total of 252 fish were randomly divided into 4 experimental dietary treatments (12 tanks/diet with 3 replicates/tank): TM0 (the control diet without TM) and TM5, TM10 and TM20 (with 5, 10 and 20% of TM inclusion level, respectively, as a replacement of 25, 50 and 100% of fishmeal). All the diets were isonitrogenous and isoenergetic. At the end of the trial (154 days), 15 fish/diet (5 animals/tank) were killed by over anaesthesia. Samples of anterior and posterior gut, liver, kidneys and spleen were collected and fixed in 10% buffered formalin solution for anatomopathological investigations. Gut morphology was evaluated through morphometric measurements of villus height, while histopathological alterations of gut and organs were scored using a semiquantitative scoring system as follows: absent (score 0), mild (score 1), moderate (score 2) and severe (score 3). Data were analysed by IBM SPSS Statistics V20.0.0 software ($p<.05$). Gut morphology was not affected by dietary TM larva meal inclusion ($p>.05$), with the posterior gut showing higher villi than the anterior one in all the dietary treatments ($p<.05$). Dietary TM larva meal inclusion did not influence either development or severity of the observed histopathological alterations ($p>.05$). Gut showed mild, occasional mucosal and submucosal lymphoplasmacytic inflammation. Mild to moderate, multifocal lymphoplasmacytic inflammation, as well as mild to severe, multifocal to diffuse steatosis, were identified in liver. Kidneys showed mild to moderate, multifocal interstitial lymphoplasmacytic inflammation, while mild to moderate, focal to multifocal white pulp hyperplasia and haemosiderosis were observed in spleen. In conclusion, TM larva meal can be included in diets for rainbow trout without any negative effects on both the gut morphology and the health status of the fish.

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O076

High protein *Tenebrio molitor* larvae meal in rainbow trout diets

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In recent years, there has been a growing interest towards the use of insect protein meals in aquaculture feeds. The use of this alternative raw material has been successfully tested in different fish species, such as Atlantic salmon (*Salmo salar*), rainbow trout (*Oncorhynchus mykiss*), gilthead sea bream (*Sparus aurata*), and European sea bass (*Dicentrarchus labrax*). The aim of this study was to evaluate the substitution of fishmeal with a high protein *Tenebrio molitor* larvae meal (TM) in rainbow trout diets. At the beginning of the trial, 252 fish were randomly divided into 12 fibreglass tanks (3 replicates/diet). Four experimental diets were formulated to be isonitrogenous and isoenergetic: TM0 (without TM), and TM5, TM10 and TM20 (with 5, 10 and 20% of TM inclusion level, respectively) in substitution of 25, 50 and 100% of fishmeal. At the end of the trial (154 days), the following performance indexes were calculated: survival, individual weight gain, specific growth rate, feed conversion ratio and protein efficiency ratio. Fifteen fish per treatment were killed by over anaesthesia and individually weighed. Fish length was measured to calculate the Fulton's condition factor (K). The fish were dissected to determine the carcase yield (CY), hepatosomatic index (HSI), viscerosomatic index (VSI), and the coefficient of fatness (CF). An *in vivo* digestibility trial was also contemporarily performed to determine the apparent digestibility coefficients (ADC) of dry matter, crude protein and ether extract of the diets. One hundred and eighty trout (15 fish/tank/tank) were divided into 12 cylindroconical tanks (3 replicates/diet). The fish were fed twice a day with the same diets used in the growth trial. The faeces were collected daily from each tank for 4 consecutive weeks, using a Choubert' system. The faeces were freeze-dried and frozen (−20 °C) until analysed. All data were statistically analysed by IBM SPSS Statistics V20.0.0 software (p<.05). No differences between treatments were observed for the performance indexes. No differences were highlighted for K, CY, VSI, and CF, but HSI resulted higher in the fish fed TM20 compared to those fed TM0. About the digestibility, all the ADC values result higher than 93%. Differences were recorded for all the ADC parameters (p<.05). TM5 diet showed the highest values. The obtained results confirm the high potential of the inclusion of *Tenebrio molitor* larvae meal in rainbow trout feed.

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O077

Black soldier fly (*Hermetia illucens*) larvae meal as a dietary protein source for rainbow trout (*Oncorhynchus mykiss*): effects on allochthonous and autochthonous gut microbial communities

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Insects represent an economically sustainable high-value and safe protein-rich alternative to fishmeal in aquafeeds for farmed fish. Accordingly, the present study aimed to evaluate the effects of partial substitution of fishmeal with insect meal from *Hermetia illucens* in the diet of rainbow trout (*Oncorhynchus mykiss*), on fish gut microbiota composition.

For this purpose, three diets, with increasing levels of fishmeal replacement with insect meal (10%, 20% and 30%) and a control diet without insect meal were tested in a 12-weeks feeding trial. Fish growth and feed conversion ratio were evaluated. The Illumina MiSeq platform for high-throughput amplicon sequencing of 16S rRNA gene and QIIME pipeline were used to analyse and characterise the whole microbiome associated to fish intestinal mucosa (autochthonous or resident microflora) as well as to the digesta (allochthonous or transient microflora). We identified 74 and 450 OTUs at 97% identity in trout mucosa and faecal samples, respectively. *Tenericutes* and *Proteobacteria* constituted the most abundant bacterial phyla of gut mucosa. *Actinobacteria*, *Firmicutes* and *Proteobacteria* represented, instead, the dominant phyla of trout intestinal lumen.

Dietary inclusion of insect meal significantly reduced the amount of *Proteobacteria*, both in resident and transient microflora. Our findings clearly indicated that insect meal positively modifies trout gut microbiota, by increasing microbial diversity of intestinal lumen and mucosa. In particular, the profile of transient microbiota of fish fed with insect meal showed an increased number of beneficial lactic acid- and butyrate-producing bacteria in comparison to the control group. Furthermore, based on the present study and other published articles, we believe that the prebiotic effect of insect meal is principally due to the fermentable chitin.

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O078

Lipid metabolism and fillet quality of rainbow trout fed diets including *Hermetia illucens* full-fat larvae

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The use of insects as protein source in feed is a hot topic in the present aquaculture scenario. Aquaculture production escalation must be supported by an increase in feed production and the conventional dietary protein sources must be substituted by cheaper and more eco-friendly ingredients, such as insects. Although a substitution of up to 50% of the conventional protein sources with insect meals is possible without jeopardising *in vivo* performances of salmonids, the effect on fillet quality is still of concern.

The present study enquired the effects of three diets (Hi0, Hi25, Hi50) with increasing substitution levels (0, 25, 50%) of dietary fishmeal with *Hermetia illucens* full-fat larva meal on liver (L), pyloric caeca (C) and mid intestine (M) lipid metabolism and on fillet quality of rainbow trout (*Oncorhynchus mykiss*), after a feeding period of 98 days. The physical analyses on fillets included colour measurement by a Chroma Metre CR-200 (Chroma Metre CR-200, Tokyo, Japan), water holding capacity determination and pH instrumental measurement. The assessment of fillet fatty acid (FA) profile was also performed. The expression of genes related to lipid metabolism (*abcg5*, *cd36*, *elovl1*, *elovl2*, *fads2*, *fads6*, *hmgcr*, *ppar*α, *ppar*β, *ppar*γ, *ppar*δ, *srebp1*, *srepb2*) was investigated by two-step RT-qPCR.

Dietary treatments did not noteworthily affect physical characteristics. Since the insects were not defatted, saturated FAs (SFAs) were abundant in the diets including insect meal; as a consequence, fillet FA profile showed an increased SFA content comparing Hi50 and Hi0 ($p<.05$). Significant differences and clear trends were revealed between the treatments as concerns the genes related to the lipid metabolism.

To sum up, the inclusion of full-fat *H. illucens* meal in diets for rainbow trout did not alter the physical traits of fillet but affected fillet cardioprotective lipid profile as well as liver and intestine lipid metabolism. An understanding of lipid metabolism, cross-checked with the lipid status of the fillets, can give hints to modulate diet formulation, aiming to improve the nutritional and functional characteristics of the final product.

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O079

Effects of dietary inclusion of defatted black soldier fly (*Hermetia illucens*) meal on gut health in gilthead seabream (*Sparus aurata*)

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Fishmeal (FM) is the optimal source of animal proteins in commercial aquaculture feeds. However, it is becoming less available and not sustainable for environment, due to overfishing. Hence, research in this sector is oriented to find alternative sources of proteins. Insects proteins, specially, black soldier fly (*Hermetia illucens*-HI) meal has been investigated, recently, for its valuable nutritional properties. This study aims to evaluate, by histological approach, the effect of dietary inclusion of defatted HI meal on gut health of 360 gilthead seabreams. Fishes with an initial weight of 181.6 g (±13.5) were fed for 120 days with a control diet

(CTR: 100% fishmeal) and three experimental diets containing increasing levels of defatted HI meal in substitution of 25 (R25), 50 (R50) and 75% (R75) of fishmeal, leading to a HI inclusion of 9.2%, 18.4% and 27.6%, respectively. At the end of the feeding trial, growth performances such as specific growth rate (SGR), feed conversion rate (FCR) and protein efficiency rate (PER) were calculated and the gut of fifteen fishes (per treatment) were fixed in Bouin (24 h), dehydrated in ethanol and embedded in paraffin, to obtain 5 μm-thick sections stained with haematoxylin and eosin. In addition, on gut sections, the following morphometric indexes were measured: villi length; villi ramification; number of goblet cells; submucosa layer detachment; villi fusion; position of enterocytes nuclei; vacuolation of enterocytes.

Results on growth performances highlight that dietary inclusion of HI meal in seabream fed up to 18.4% of inclusion level did not influence any of the growth performance traits. On the other hand, HI inclusion level of 27.6% worsened significantly SGR (p=.003), FCR (p=.007) and PER (p=.007). Histological evaluation showed no significant differences between the CTR group and the R25 one. On the contrary, in some R50 subjects anatomo-functional changes of the gut were observed. More frequent and pronounced structural alterations were observed in R75 group, both in the anterior and posterior part of gut; in some cases, haemorrhagic spots were observed. Significant (p<.05) changes in the morphometric parameters of groups fed higher levels of HI (mostly R50 and R75) confirmed the results of anatomopathological exam. In conclusion, it is important to take into account not only the effects of HI meal on growth performances but also those on the histological analyses that highlight the onset and increasing of an irritant state of gut-associated with the increase of HI substitution level.

O080

Fillet qualitative characteristics of Atlantic salmon fed diets including *Hermetia illucens* larvae

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Aquaculture is expected to supply more than 65% of global fish production by 2030, thus increasing quantities of fish meal (FM) will be necessary as protein source in aquafeeds. However, the fish stocks (mainly anchovy and herrings) from which FM derives are nowadays overexploited. Hence, aquafeed production needs to be based on alternative protein sources. Among the other, insects are promising due to their fast growth, easy reproduction, few nutritional requirements and their ability to recycle organic waste. The present study tested the effects of the replacement of FM with partially defatted *Hermetia illucens* larva meal (HI) in the diet for seawater-phase Atlantic salmon (*Salmo salar* L.) on physico-chemical characteristics and consumer liking of fillets. Triplicate sea-cages of A. salmon were fed one of four isoproteic, isoenergetic and isolipidic diets for 16 weeks: a control diet (C, 0% of HI) and three diets with increasing substitution levels of FM with HI (33, 66 or 100%; IM33, IM66 and IM100, respectively). Then, fish were percussively slaughtered and filleted. Four salmon fillets from each sea-cage were allotted to physico-chemical analyses ($n = 6$) and consumers' test ($n = 6$). The physico-chemical analyses included colour evaluation (Chroma Metre Cr-200, Tokyo, Japan), water holding capacity (WHC), texture profile analysis (Zwick Roell® texturometer, Zwick GmbH & Co. KG, Ulm, Germany), and proximate composition determination. A blind product test was performed with 80 consumers who evaluated on a 9-points scale the liking for appearance, odour, flavour, texture, and the overall judgement; they evaluated the adequacy of colour, texture, appearance, aroma, flavour and salty intensity, juiciness and fibrousness. Colour, WHC, texture and proximate composition were not affected by diet. Only IM66 group contained higher amount (p<.05) of protein than IM100 (20.92 and 19.48g/100g of fillets, respectively), while C and IM33 assumed intermediate values. Consumers' liking was unaffected by the diet and received scores from 6 to 9. The main criticisms were related to the adequacy of colour and textural attributes. Indeed, IM100 group was considered the palest, the most fibrous and with the lowest aroma and flavour. Nevertheless, consumers highlighted their propensity to future consumption. To sum up, the inclusion of HI in diets for A. salmon determined physical modifications which were not instrumentally quantified, but which were perceived and accepted by consumers.

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – ANIMAL NUTRITION: EFFICIENCY

O081

Sources of variation in TMR delivered to dairy cows: a field study

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Total mixed ration (TMR) delivered to dairy cows can be subjected to daily variations in its chemical composition and particle size distribution (PSD). These variations could negatively affect nutrients provision and performance. Sources of variations in TMR depend on a number of factors: besides the variability of forages itself, some of the diet preparation procedures (DPP) could account. The aim of this study was to detect the variation sources in TMR with especial regard to DPP in field. The study was conducted using a horizontal self-propelled TMR mixer wagon (Gulliver 6014, Sgariboldi, Codogno, Italy) with desilage conveyor arm, cutting slasher, and mixer tank of 14 cubic metre capacity with paddle technology. Charging feeds times (CFT) and sequences (SEQ), feeds mixing times (FMT), and discharging times (DT) of TMR were considered in a dairy farm located in the north of Italy (Lombardy). The diet provided was based on corn, grass silages and hay. Samples of each TMR were collected at the beginning, middle and end of the feeding alley (FA) for chemical composition and PSD (Penn Particle Size Separator) of the diet. Feed samples were analysed for dry matter (DM), crude protein (CP), ether extracts (EE), neutral detergent fibre (NDF), acid detergent fibre (ADF) and ash content. Data were analysed by ANOVA of S.A.S. (v9.2) dividing CFT, FMT and DT in homogenous classes, while a total of four SEQ were considered. Significance was declared for $p<.05$. Obtained results showed as the chemical composition of the diet was significantly modified along the FA for DM, CP, EE and ash content, while no differences were found for NDF and ADF. In most of the cases accounted DPP such as SEQ, FMT and DT hugely influenced the chemical homogeneity of the diet in terms of DM, CP, EE, NDF, ADF and ash ($p< .01$), while no effects were detected for CFT on EE and NDF content. PSD of the diet was strongly modified in middle, lower and bottom sieves of Pen Particle Size Separator along the FA ($p< .01$), while the major effects of DPP were found on the lower and the bottom sieves ($p< .01$). The present study shows how each step of the TMR preparation can affect both the chemical content of nutrients and the PSD of the diet. In particular DM and PG diet content, together with lower and bottom sieves are the items most affected by all the DPP considered.

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O082

Evaluation of feed efficiency in Italian Holstein Friesian heifers and lactating cows

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At present, the improvement of livestock feed efficiency (FE) represents one of the main challenges for the future of the agri-food supply chain, aimed at increasing farm sustainability and profitability. The growing awareness of the relationships between FE and factors like dry matter intake, methane emission and thermoregulation, contributes to the interest in selecting for efficient animals. The aim of this work was to investigate the relationships between different indices of FE and performance parameters in a herd of Italian Holstein-Friesian dairy cattle. Trials were conducted at the Experimental farm of Animal Production Research and Teaching Centre of Lodi (CZDS). The first batch of 16 heifers was investigated for dry matter intake (DMI), body weight (BW), body condition score (BCS) and other growth parameters. Data obtained were used to calculate the residual feed intake (RFI) and feed conversion ratio (FCR) of each animal. After the ex-post division of the batch into two groups with the higher (H-RFI) and the lower (L-RFI) RFIs, data were analysed by a MIXED procedure of SAS. Significant differences were highlighted between DMI values of the two groups ($p=.01$) and also for the wither height gain character ($p<.05$). The second batch of 30 lactating cows was investigated for DMI, BW, BCS, milk yield and composition. Data obtained were used to calculate the RFI and milk to feed ratio (M:F) of each animal. The ex-post division of the batch into two groups with the higher and the lower RFIs highlighted a statistically significant difference between DMI values of the groups but not between milk production-related parameters. Statistical analysis also underlined a positive correlation (CORR procedure, SAS) between RFI and DMI values (PCC =0.80) but not between RFI and milk production-related parameters, as opposed to M:F values, characterised by negative relations with milk energy output (PCC= −0.69) and energy corrected milk (PCC= −0.63). Our results confirm the goodness of RFI as an index of FE, a character increasingly regarded as independent from growth and production level and more linked to the animal intrinsic metabolic processes.

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O083

Characteristics of forages collected in different areas of Benin

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The Republic of Benin is a little state of South-Western Africa, characterised by different environmental. Its economy is mainly based on agriculture and livestock; pasture is the most used practice. To improve ruminants' performances, the quality of forages represents one of the most important factors to keep under control. To choose the most appropriate botanical species to utilise in ruminant diets during forage shortage periods, 2 local forages (*Dactyloctenium aegyptium, Leptochloa caerulescens*) were collected in 3 peri-urban areas of southern Benin (Amandji, Gakpé, Zinvié). The areas differed for location, human and livestock activities, and soil characteristics, but are similar for climatic conditions. The samples were analysed for chemical composition and *in vitro* fermentation characteristics, including CH_4 production. *D. aegyptium* collected in Zinvié showed higher CP and energy level and lower NDF content even if ADL content was higher (12.5, 63.6 and 8.69% DM, respectively; $p<.05$). OM degradability ranked the areas as follows: Zinvié > Gakpé > Amandji, probably due to the high ash content (11.1% DM) in Zinvié. The latter area also showed the highest VFA production (98.8 mM/g; $p<.01$). Compared to Amandji, *L. caerulescens* showed higher CP and lower NDF content ($p<.05$) when collected in Zinvié and Gakpé, although, in both areas, ADL content was higher ($p<.05$). VFA production was higher ($p<.01$) when samples were taken in Amandji and Gakpé compared to Zinvié (82.3, 78.0 and 63.1 mM/g, respectively). Overall, CH_4 resulted higher (36.57 mL/g dOM) compared to previous data in forages produced in Mediterranean area of Southern Italy. On average, the nutritive value was quite poor. To meet animal requirements, nutritional strategies need (e.g. protein integration with local and cheaper sources). Many differences appear among the studied areas: both forages collected in Zinvié showed best nutritional characteristics (CP and NDF content, OM degradability), whereas in Amandji the least interesting. Probably, soil characteristics have contributed to these results: the presence of water has favoured Zinvié and the sandy and hydromorphic soil have negatively affected Amandji. Further studies are needed: increasing forage species, better characterising soil, using *in vivo* trials.

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O084

Milk production and digestibility in cows fed diets with different forage basis

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The environmental sustainability of milk production can be attained paying particular attention to the forages included in the diet and the related use of imported soybean meal on one side, and the soil carbon sink achievable by different forage crops and rotations on the other side.
Eight Italian Friesian lactating cows (2.4 parities, 127 DIM and 601 kg BW on average) were used in a repeated Latin Square design (4 dietary treatments × 4 periods × 2 squares) to evaluate the dairy efficiency and the digestibility of 4 diets formulated on different forage basis. The diets (% on DM) were: (1) CON (conventional): 49% maize silage and 17% Italian ryegrass hay; (2) HQ (high quality): 27% Lucerne silage, 19% Italian ryegrass silage; (3) WC (winter cereals): 20% wheat silage, 10% Lucerne silage, 10% Lucerne hay; (4) PR (Parmesan Reggiano): 25% Lucerne hay, 25% Italian ryegrass hay. Soybean meal, maize grain meal, and maize ear silage were used in different proportions to balance the protein and the starch content among the diets.
Diets were formulated to be similar for net energy and metabolisable protein content. ADFom content of the diets CON, HQ, WC and PR was respectively 21.8, 22.6, 23.8, 26.2% on DM.
Each period lasted 4 weeks, with 3 weeks of adaptation in the stall before the week of digestibility by total tract collection. Data were statistically analysed by proc mixed by SAS.
For CON, HQ, WC and PR respectively, DM intake was 19.8^{b}, 20.4^{b}, 20.3^{b} and 23.4^{a} kg/d ($p<.01$), milk yield 27.0, 27.3, 28.2, 29.3 kg/d with 4.38, 4.60, 4.71, 4.26% fat, and 3.58, 3.52, 3.56, 3.53% protein, and the dairy efficiency (kg milk/kg OM intake) 1.48^{ab}, 1.49^{a}, 1.51^{a} and 1.36^{b} ($p<.05$). Digestibility (%) of the diets CON, HQ, WC and PR was respectively: OM $=74.4^{a}$, 74.1^{a}, 70.5^{b}, 67.3^{b}; CP $=70.2^{a}$, 55.8^{b}, 67.9^{a}, 55.5^{b}; aNDFom $=48.8^{a}$, 45.0^{ab}, 39.4^{b}, 38.0^{b} ($p<.01$). PR diet enhanced feed intake but lowered digestibility and the dairy efficiency; forage protein was less digested than soybean meal protein.
In conclusion, good quality forages associated with maize to supply starch, allow to attain similar milk production and efficiencies as maize silage based-diet, with a lower purchase of soybean meal.

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O085

Prickly pear pulp in the feeding of livestock ruminants: preliminary investigation

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In Sicily, the current increasing cultivation of *Opuntia ficus-indica* (L.) Mill. corresponds to an availability of prickly pears pulp (PPP) that residues from fruits processing for juice extraction, comprised of peel, pulp and seeds. This investigation aims to evaluate the nutritional traits of PPP for ruminants feeding, and the effect of potassium metabisulfite (PMB) as a natural stabiliser to limit initial rapid PPP fermentation. The by-product, collected in August, included 28% of peel and pulp (PP) and 72% of seeds on dry matter (DM), with seeds higher in ether extract (EE) and NDF and comparable for DM and crude protein (CP) (DM 35.5, 35.9 and 35.6%; EE 6.5, 3.6 and 8.5% DM; CP 6.5, 6.0 and 6.7% DM; NDF 62.0, 41.5 and 73.3% DM, for PPP, PP and seeds). For PPP, the net energy for lactation was 1.1 Mcal/kg DM, total polyphenols were 2.71 mg gallic acid equivalent/g DM, the DPPH (IC_{50}) resulted 65.6 mg DM/mL, and the reducing power (EC_{50}) 109.4 mg DM/mL. The PPP showed a total microbial load of 1.7×10^5 CFU/g, mainly due to mesophilic cocci lactic acid bacteria (1.4×10^6 CFU/g), responsible of sugar fermentation, and hosted a certain presence of coliforms (1.2×10^5 CFU/g) derived from the environment, while *Escherichia coli*, *Listeria monocytogenes* and Salmonella spp. were not detected. Four heaps of 20 kg of fresh PPP, were placed on the ground, treated with different PMB doses (0, 50, 100 and 150 g/100 kg of PPP for T0, T1, T2 and T3), and covered by a breathable plastic sheet, simulating the storage technique in farms. The PMB doses were established to have a very low risk to reach the maximum amount recommended by WHO for human consumers (0.7 mg/kg of body weight). The initial pH of PPP (5.0) decreased more slowly in the treated heaps, reaching after 9 days values of 3.80, 4.05, 4.21 and 4.23 with T0, T1, T2 and T3. Similarly, the sugar content in the treated heaps decreased more slowly from day 1 (133.4 g/kg) to day 7 (8.8, 9.1, 9.5 and 9.7 g/kg with T0, T1, T2 and T3), suggesting a certain efficiency of higher PMB doses to slower the initial fermentation. Based on these first results, the PPP appears an interesting by-product to be used as a feeding source able to increase the environmental and economic sustainability of ruminants livestock. Proper researches need to set up methods to preserve the PPP safety during storage and evaluate its potential antioxidant properties that can be beneficial to livestock animals and consumers of their products.

O086

Body weight and measures of purebred Holstein and crossbred cows from Viking Red, Montbéliarde and Holstein sires

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Crossbreeding in dairy cattle has become of interest for commercial dairy farms to enhance fertility, health and longevity of cows, and most studies focussed on these traits, as well as on effects on milk yield. Conversely, effects on production efficiency have been scarcely investigated. Body weight and measures can be used as scaling factors for computing efficiency indicators, and for estimating energy requirements of cows. This study aimed to compare the purebred Holstein (Ho) with crossbred cows produced within a rotational crossbreeding scheme involving Viking Red (VR), Montbéliarde (Mo) and Ho sires on body traits and to develop prediction equations for estimating the body weight (BW) of cows. BW, body condition scores (BCS) and body measures were taken on 225 cows kept in a commercial dairy farm (number of sampled cows was 60 for purebred Ho and ranged between 15 and 35 within each breed combination). BW averaged 664 ± 88 kg and was similar for Ho and crossbreds, but Ho were ($p<.05$) taller, longer in body, had greater heart girth and lower BCS (3.03 vs. 3.34, $p<.05$) than crossbred cows. For developing prediction equations to estimate BW, records available were divided into a calibration (150 cows) and a validation (75 cows) data set. The former was used to run different prediction models of BW, using the regression procedure of SAS and including as independent variables BCS and body measures alone or combined with parity and days in milk class or breed effects or both; the latter was used for validating the reliability of models developed on an independent data set. Determination coefficients (R^2) of regressions for 4 models developed were constantly above 0.80. The model selected included breed effects (VR and Mo, with respect to Ho, which was included in the intercept), heart girth, body length and BCS. *R2* of the model was 0.83, with a root mean square error of 33.92 kg. Bias (the average difference between predicted and observed BW) and precision (the standard deviation of bias) was –5.7 and ±31.55 kg, respectively. The results obtained from this study provided a new insight into the phenotypic variation of crossbred cows obtained from a 3 breed rotational crossbreeding scheme. Besides, this prediction equation, as a simple and reliable method to estimate the BW of crossbred cows, can be used in further studies aimed to compare the performance of pure Ho and crossbred cows in term of production efficiency.

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O087

Production efficiency of purebred Holstein and crossbred cows from Viking Red, Montbéliarde and Holstein sires

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This study aimed to investigate the production efficiency of pure Holstein (Ho) compared to crossbred cows within a 3-way rotational crossbreeding scheme involving Viking Red in 1st (F1: VR × Ho), Montbéliarde in 2nd (F2: Mo × F1), and Ho sires in 3rd generation (F3: Ho × F2). Body condition scores (BCS) and body measures were assessed on 635 cows kept in 2 dairy herds. On the same cows, milk yield (MY) was recorded and milk samples were collected to assess the milk composition and individual cheese yield (CY). Body weight (BW) of cows was estimated through regression equations based on BCS and body measures. Net energy requirements were estimated using NRC equations based on predicted BW, MY and milk nutrients. Production indicators rely on daily yield (kg/d) of milk, fat, protein, casein, cheese and milk energy (MJ/d) taken as absolute values or scaled on BW, metabolic weight (MW), and total net energy requirements (NE). BCS averaged 3.24, predicted BW 678 kg, and MY 34.5 kg/d, with average 3.77% fat and 3.63% protein. Given an average %CY of 16.80%, cows provided around 5.75 kg/d of cheese. Crossbred cows had (p<.01) greater BCS, BW and total body fat compared to Ho. They yielded lower milk (−5%) with a greater protein content (+1.4%), therefore providing similar daily fat and protein yield. No difference between Ho and crossbreds was found for CY, but greater MY of Ho reflected in greater (5%) daily CY and energy produced in milk. Ho cows were superior to crossbreds also when comparisons were expressed in term of production efficiency indicators for daily yield of milk, milk components, milk energy and CY. However, the magnitude of difference between Ho and crossbreds varied according to the scaling unit considered. Superiority of Ho in comparison to crossbreds ranged between 5 to 9% when production indicators were expressed on BW or MW, with differences always significant (p<.05). Conversely, when production indicators were expressed on NE, differences between genetic lines (p<.05) was limited to 3% for daily CY and MY and to 2% for daily milk energy production, whereas daily yield of fat, protein and casein were not different between Ho and crossbreds.

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O088

Quality assessment of corn silage by infra-red thermography

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The qualitative assessment of feed intended for ruminants appears to be an increasingly interesting topic for the influence it may have on health of both animals themselves and humans as a result of the consumption of animal products, in particular of milk.

Corn silage may be the predominant element in the ration of both dairy and meat cattle; therefore, innovative and possibly rapid technologies are needed to assess its quality, in order to guarantee public health and improve the economic performance of animal's products.

In this study, an infra-red camera was used to identify areas subjected to aerobic deterioration in a corn trench silo, during the entire period of use, in a typical farm with 400 dairy cows in the north of Italy.

Over a period of 5 months, from April to September, the temperature of silage front was detected with an infra-red microbolometric camera, model AVIO G120 EX (320 × 240 pixel), early in the morning with no heating due to direct solar radiation on the trench front. The value of the emissivity was set at 0.97.

Chemical and nutritional analysis were also carried out on samples taken at representative points of the average silage condition and at points marked as warmer by the thermal imager. The statistical analysis (analysis of variance) was performed with the GLM procedure of the SAS programme.

In the warmer points, a higher pH value (p<.01) and a lower concentration (p<.05) of lactic acid were detected. More, the relative humidity in the last 3 months was higher in the points reported as anomalous from the infra-red camera (p<.01).

The composition of silage in dry matter, proteins, fats, starch and ash, did not undergo significant variations during the five months of use and was overall of excellent quality.

It should be noted that the thermally anomalous points have always been detected in peripheral areas of the trench front.

Therefore, during this test, the thermography has proved to be very useful in recognising, rapidly and in non-invasive way, areas of the silos subjected to aerobic deterioration.

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It should also be noted that, working remotely and without any contact with the material, this technique presents no risk of microbiological dissemination from the silages.

O089

Factors affecting the fatty acid profile of permanent grasslands

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Permanent grasslands are traditionally used as forage for ruminants in many European countries. The fatty acid profile of permanent grasslands can influence the nutritional quality of milk and meat products from ruminants. This study used available data ($n = 144$) collected in the years 2003–2016 by three research institutions from Italy, France and Switzerland to explore factors able to affect the fatty acid profile of semi-natural and biodiversified pastures used as grazing areas by commercial dairy herds. The dataset was built up to match a large altitude range (15–2500 m a.s.l.) and botanical diversity. The dataset included 14 variables comprising: herbage fatty acid profile [C16:0 (%), C18:0 (%), C18:1c9 (%), C18:2n6 (%), and C18:3n3 (%)], herbage proximate composition [dry matter (DM, g/kg), crude protein (CP, g/100 g DM), neutral detergent fibre (NDF, g/100 g DM), and acid detergent fibre (ADF, g/100 g DM)], herbage botanical composition [grasses (*Poaceae*, %), legumes (*Fabaceae*, %), and forbs (%)], herbage phenology (BBCH scale), and site altitude (m a.s.l.). Data were analysed by Principal Component (PC) Analysis using IBM SPSS Statistics v. 25 for Windows. The Kaiser-Meyer-Olkin measure of sampling adequacy and the Bartlett's test of sphericity were used to verify the adequacy of data analysis. The sum of the first three linear components accounted for 70.7% of the total explained variance. Considering factor loadings >0.6 (or <–0.6), the herbage C16:0, C18:0, C18:1*c*9 and C18:2n6 contents, the DM, NDF and ADF contents, and herbage phenological stage were positively related, whereas herbage C18:3n3 and CP contents were negatively related to the 1st PC (45.6% of explained variance). Legumes were positively related to the 2nd PC (13.1% of explained variance) and grasses and forbs were positively and negatively related to the 3rd PC (12.1% of explained variance), respectively. In conclusion, regardless the altitude, the variation in the fatty acid composition of semi-natural grasslands in on-farm conditions appeared to be much more related to herbage phenology (and consequently to the proximate composition and nutritive value), rather than to the botanical composition. The latter played anyway a role, even if less relevant than the herbage phenological stage.

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ANIMAL PRODUCTS– MEAT QUALITY: GASTRONOMIC TRADITION AND INNOVATION

O090

Quality and safety of meat from wild boar hunted in Molise region

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In recent decades, there has been a consistent increase in the population of wild boar in Italy and also in the Molise region, with a greater availability and consumption of their meat. The aim of the study was to evaluate pH, colour, total lipid, fatty acids composition and heavy metals content of wild boar meat of different estimated live weight (~50, 70 and 100 kg; $n = 25$, 24 and 18, respectively), sex and hunting area (A1: Bagnoli del Trigno-Poggio Sannita (IS); A2: Roccavivara-Civitacampomarano (CB)). The study was carried out on wild boars hunted between November 1st 2017 and January 31st 2018. At slaughter, *Longissimus dorsi* muscle was removed from carcases for the analyses. Meat quality data were analysed by GLM, live weight and sex were the main factors; heavy metals data were analysed considered also the hunting area factor. pH and colour were not affected by live weight and sex; however, meat from heavier boars (100 kg) was slightly darker than that of lighter ones (50 kg). Total lipid, total saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA) were not affected by weight. Differently, lighter boars showed a higher content of total polyunsaturated fatty acid (PUFA, $p<.05$), n-6 ($p<.05$) and n-3 (70 kg: $p<.01$; 100 kg: $p<.05$) PUFA, and PUFA/SFA ratio ($p<.01$) compared to heavier classes. Lighter boars had a better atherogenic index compared to boars from intermediate weight class (70 kg). Compared to females, males had a higher ($p<.05$) n-3 PUFA content and a lower ($p<.05$) n-6/n-3 ratio. Weight, sex and hunting area did not affect cadmium (0.001 mg/kg), lead (0.011 – 0.026 mg/kg), copper (0.696–1.151 mg/kg) and manganese (0.083 – 0.130 mg/kg) contents. Chromium was affected only by sex (males: 0.072 mg/kg; $p<.05$). Nickel content was higher ($p<.01$) in the heaviest boars (0.035 mg/kg) than the other weight classes (0.017 mg/kg). Wild boars hunted in the A2 area showed higher nickel concentrations ($p<.01$) than those hunted in the A1 area (0.026 vs. 0.018 mg/kg, respectively). Interactions ($p<.01$) were detected among all the three factors taken into consideration for nickel content. Sex did not affect nickel content. In conclusion, lighter boars showed a better meat quality from the nutritional point of view. In addition, the low content of heavy metals in the meat indicates a very low level of anthropogenic pollution of the areas under study, making wild boar meat safe from a health point of view.

O091

Effect of ageing time on the volatile compounds profile of donkey meat

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The aim of the work is to assess the ageing effect on the volatile compounds profile of vacuum packaged donkey meat stored at 4°C for 14 days. Ten Martina Franca breed male donkeys were included in the trial. They were slaughtered at the age of 12 months old. Samples of *Longissimus Lumborum* (LL) muscle were taken from the right half 24 h after slaughtering, sliced in 20 mm thick steaks, vacuum packed, stored at 4 °C and analysed at 1, 3, 6, 9 and 14 days after slaughtering. At each time, samples were grill cocked and Volatile Organic Compounds profile was performed by solid-phase microextraction (SPME) and gas chromatography-mass spectrometry (GC-MS). The VOC profile was analysed using a one-way ANOVA, where ageing time was set as independent variable. The values were given in terms of mean values and standard error of the means (SEM). When a significant effect ($p<.05$) was detected, means were compared using the Tukeýs test for repeated measures. Pentane and octane showed decreasing trend during ageing, with the lowest values at 9 and 14 ageing days compared to others ($p<.01$), with no differences showed in the total hydrocarbons ($p<.05$). Many researchers pointed out that these compounds made no significant contribution to meat flavour. On the contrary, aromatic hydrocarbons have an important contribution to meat flavour, and they showed a decreasing trend, with the lowest values observed from the 6th to the 14th ageing day ($p<.01$); furan 2-penthyl, in particular, showed lowest values from the 6th ageing days until the trial end ($p<.01$). Total aldehydes did not show to be affected by ageing time, although benzaldehyde was the only one that showed at 14 ageing days the highest values than other days ($p<.01$). Total alcohols, and in particular hexanol, showed lowest values from the 6th to 14th ageing days ($p<.01$). Total aldehydes did not show any differences during ageing time, although this family of volatile compounds was the most produced by donkey meat. Aldehydes have often also tallowy and meaty odours and hexanal, that is considered a good oxidation indicator, characterised by high aromatic potential, providing freshly cut-grass and green aroma notes, is the most present compound. The ageing of donkey meat under vacuum conditions for 14 days leads to small changes in the volatile compounds profile after grilling cooking. However, the volatile compounds that showed some variation included primarily those indicative of lipid oxidation.

O092

Dromedary meat proprieties: case of algerien sahraoui dromedary

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In the last years, due to climate changes, the choice of adequate management for food security and sustainable development becomes a necessity and, in this scenario, in Algeria, the dromedary can have an important role. The camel genus present in Algeria is the *Camelus Dromedarius* or Dromedary, its estimated number is about 379094 head in 2016. In the Southeastern region, Sahraoui dromedary population is widely present. The heat tolerance, high carcase yield under inexpensive management systems and the good nutritional properties of meat make dromedary an important food source in the arid areas. Particularly, dromedary meat is an interesting source of protein with high nutritional value and shows lower fat contents with the ratio of polyunsaturated fatty acid/saturated fatty acid close to the nutritional recommended value. However, the non-aboriginal consumers have a particular image on dromedary meat quality linked to less desirable palatability, which can result from a multitude of *ante-* and *post-mortem* factors. In order to better understand the potentialities of dromedary in terms of meat quality traits, the present study summarises some meat quality proprieties of Algerian Sahraoui dromedary, with a particular focus on tenderness.

The effects of ageing time (6, 8, 10, 12, 24, 48 and 72 h) were evaluated in *Longissimus lumborum* muscles from Six Sahraoui dromedaries (8–10 years old). pH, myofibrillar fragmentation index (MFI) and total collagen content were estimated. The measurements revealed a quick acidification of the muscle with a pH value of 6.15 at 6 h *post-mortem* while the pH at 24 h was 5.9.

MFI showed the highest value at 6 h *post portem* after decrease with a minimum value recorded at 8 h *post-mortem* and starting for 12 h increase. In contrast collagen content was lowest at 6 h then increases with highest value at 12 h and starting for 24 h post mortem decreases progressively. Beside this characterisation, a further investigation on particular proteolysis biomarkers of dromedary meat quality, especially in the early ageing times is suitable for a better valorisation of the product.

O093

Effect of cooking method on the volatile organic compound profile of meat of different species identified by PTR-ToF-MS

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The volatile organic compounds (VOCs) are important contributors of meat flavour. The aim of this preliminary experiment was to compare the VOCs profile of meat of different species cooked by different methods. Briefly, meat samples (rib eye for mammals and breast fillet for avians) from 100 animals of 5 species/categories (beef, veal calf, pork, chicken and turkey) were sampled during 10 sessions from 10 different retailers (5 supermarkets and 5 local butcheries). From each meat sample, 4 hamburgers were prepared and cooked. Two hamburgers, according to ASPA, were cooked in water bath at 70 °C for 40 min and two, according to AMSA, were grilled on a griddle heated at 150–160 °C for about 3 min per side, till an internal temperature of 70 °C was achieved. The subsamples were thawed overnight and analysed after 30 min at 25 °C. The raw meat subsamples, after analyses at 25 °C, were heated at 70 °C by PTR-ToF-MS, and analysed again. For detecting VOCs, Proton transfer reaction – time of flight – mass spectrometry (PTR-ToF-MS) was used. This technic revealed 383 peaks on each subsample, 129 of which, the most intense ones, were retained, and 56 were tentatively identified as relevant VOCs on the basis of their fragmentation patterns and available literature.

The VOCs profile of the 600 meat subsamples were analysed using a mixed model including the effect of session, species, meat sample (random), cooking method, interactions and error. Cooking increased VOCs amount in meat of all species/categories. VOCs released by raw meat increased after heating at 70 °C, but remained lower than those found in grilled and especially in boiled hamburgers. The increase was different for different species and VOCs. The large majority of peaks had significant difference between the different species. Most of the times, turkey had the greatest amount, beef and veal calf have an intermediate, pig and chicken the lowest. On the total of 129, 110 peaks showed a significant interaction between the meat type and the cooking method. Only 7 peaks showed differences between the retailer types (supermarkets vs. butcheries).

In conclusion: (a) PTR-ToF-MS showed to be a powerful instrument for analysing meat flavour, because of high throughput and precision; (b) meat flavour is characterised by a very large number of different VOCs; (c) the VOC profile of meat is largely affected by species, cooking method and their interaction and (d) new research opportunities are now available for characterising genetics, nutrition, and management of animals, and processing of meat.

ANIMAL PRODUCTS– NEW FRONTIERS IN MILK QUALITY

O094

Traceability of buffalo milk and mozzarella cheese through metabolomic profile

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The metabolome provides a comprehensive profile of compounds and has emerged as a reliable signature that differentiates the geographical origin and hence the authenticity of food products.

In Italy, ‘Mozzarella di Bufala Campana’ cheese, gained a very well-defined profile in the market, enhanced and protected by a Protected Designation of Origin (PDO) trademark. Since attempts are regularly made to place non-authentic PDO mozzarella in premium markets as a substitute for real PDO buffalo mozzarella, the aim of the present study was to combine advanced GC-MS and metabolite identification in a robust and repeatable technology platform for characterisation of the metabolome of buffalo milk and mozzarella. The study utilised 20 commercial buffalo dairies. Eleven dairies were located in a PDO area and nine dairies were located in non-PDO area in Italy. All dairies had a processing facility that produced mozzarella cheese exclusively from their own milk. Pooled samples of raw milk (100 mL) and mozzarella cheese (100 g) were obtained from each dairy. A total of 185 endogenous metabolites were consistently detected in both milk and mozzarella cheese. The PLS-DA score plots clearly differentiated PDO and non-PDO milk and mozzarella samples. Regarding milk samples, it was possible to divide metabolites into two class according to area: those with lower concentrations in the PDO (Galactopyranoside, hydroxybutyric acid, allose, citric acid) and those which have lower concentrations in non-PDO samples (Talopyranose, pantothenic acid, mannobiose, maltose, phosphate, mannofuranose, dodecanoic acid, lactose, palmitic acid, N-acetyl glutamic acid, N-acetyl glucosamine). The same was recorded in mozzarella samples with some metabolites (Talopyranose, 2,3-dihydroxypropyl icosanoate, Sorbose, 4-pnehyl glutamic acid. Oxalic acid, Galactose) higher in PDO samples while others (Tagatose, Lactic acid dimer, Ribitol, Dodecyl thioglycolate, N-acetyl glucosamine, Valine, Diethylene glycol) higher in non-PDO samples. At present, more studies and efforts should be done on the applicability of the use of a metabolomic approach for legal purposes.

O095

Antioxidant and anti-inflammatory activities of buffalo milk bioactive metabolites

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The interest on developing foods with health promoting and disease preventing properties is still increasing. Buffalo milk (*Bubalus bubalis*), the second most consumed milk worldwide, is a good source of substances with high nutritional value which are necessary for body maintenance and to fight against several disease conditions. The health promoting and functional ability of buffalo milk bioactive metabolites have been recently expanded. Indeed, recent results showed peculiar profile of betaines and short-chain acylcarnitines of buffalo milk, with regard to δ-valerobetaine (δVB), γ-butyrobetaine (γBB), acetylcarnitine, and propionylcarnitine.

Buffalo milk also shows high antioxidant and anti-inflammatory activities in counteracting the high-glucose (hGluc)-induced endothelial damage. These effects were amplified by δVB enrichment, resulting in decreased reactive oxygen species, lipid peroxidation, and cytokine release during hGluc treatment (p<.05). Of interest, δVB counteracted the hGluc activated inflammatory signal by modulating the expression levels of SIRT1, SIRT6 and NF-κB (p<.05). In conclusion, this study shows the first evidence of buffalo milk properties in reducing endothelial oxidative stress and inflammation, suggesting a potential role of δVB as a novel dietary compound with health-promoting properties.

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O096

Study of nutraceutical substances (Rutin) in sheep’s milk fed with buckwheat hay

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Different ruminants’ diet can change milk chemical and nutritive composition. Forages, such as buckwheat, could change the nutritive properties of milk, being a significant source of bioactive compounds, metabolites that are thought to have an important role of both human and animal diets. Recent research has demonstrated that flavonoids play a synergic antioxidant role against free radical damage. Flavonoids, such as rutin, are an important class of phytochemicals products found in most fruits, vegetables, herbs and forages.

Rutin is the dominant flavonol glycoside in common buckwheat and is well known that is a powerful antimicrobials and antioxidants with anti-inflammatory and immune system benefits. Two groups each with ten Massese sheep (body weight 60 ± 5 kg) at the same stage of lactation were studied. Blood samples of the two groups were collected before and at end of the trial in order to monitor the metabolic profile. Each animal was fed daily with 1 kg of buckwheat hay and 1 kg of concentrate. The rutin content of buckwheat hay was 2.26 g/kg.

The animals were monitored in order to ensure that the hay was completely ingested. Individual milk samples were collected at 0, 15, 20, 25, 30, 35, 40 days after the first buckwheat hay administration.

The rutin determination in buckwheat hay and milk was performed by a HPLC method. After 15 days of buckwheat hay administration, the lower rutin content in milk was 0.89 mg/kg. The presence of rutin in milk may constitute a non-hydrolysed aliquot or may indicate a difference between monogastrics and ruminants. This also could be in agreement with the literature which indicates that urinary and biliar excretion of flavonoids and their metabolites may be the main metabolic pathways for such compounds.

Is well known that flavonoids have biological activities and they could use for preparing fortified foods, these compounds could be added to milk and other food.

The study in progress will be useful to evaluate the rutin carry-over from buckwheat hay to milk and the possible nutraceutical applications of this milk to improve health benefits in humans.

O097

Use of donkey milk in infant feeding

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There are still few literature about the role of donkey milk (DM) in human nutrition and increasing knowledge is crucial in order to provide practical advice for DM consumption. The aim of this study was to monitor nutritional quality, hygiene and health risks, and the impact of DM in the feeding of children with cow's milk protein allergy (CMPA). DM was supplied by a farm located in central Italy, conforming with EU regulation 853/2004. Eighteen pasteurised milk samples (at 65 °C for 30 min) were taken monthly. Pasteurised DM showed a total average viable count of 4332.22 CFU/mL (±3046.78), a slightly alkaline pH (7.12 ± 0.17), a lactose percentage of 6.83 ± 0.34, a total protein percentage of 1.63 ± 0.19, while casein was 0.81%±0.11. Fat percentage (0.51 ± 0.52) was lower compared to ruminant milk and about 48% of the total milk fatty acids were unsaturated. In addition, DM contained 7.52 ± 2.49 g/100 g of fat of n3 linolenic acid. Eighty-one children with CMPA referred to the Allergy Unit of the Anna Meyer Children's Hospital were recruited. They underwent to an allergological work-up including an oral food challenge (OFC) with DM; during the OFC the palatability of the milk was also evaluated. In children ≥3 years of age, DM palatability was assessed with a specific Wong-Baker modified pain scale, while in children <3 years of age it was assessed through the physician's judgment. The results of the allergological work-up showed that DM did not caused allergic reactions in the 98.7% of patients, in addition, a good palatability of the milk was found. DM was included for six months in nutritional plans for 16 children with IgE-mediate CMPA (mean age of 20 ± 18.4 months at the beginning of the study) and six with Food Protein-Induced Enterocolitis Syndrome (mean age of 5.33 ± 1.75 months). The daily dose of DM varied from a maximum of 1000 mL to a minimum 200–250 mL according to the age of the children. Given the low fat of DM, the diet of the children was supplemented with extra virgin olive oil (EVO) according to the age (from a minimum of 1.5 g of EVO and 1.5 g of Medium Chain Triglycerides vegetable oil in each 100 mL of milk to a maximum of 8–10 mL of EVO added to the daily meals). All the children that underwent to the nutritional plans were monitored twice (at the beginning and at the end of the study) for the auxological parameters. The results showed that DM did not change the normal growth rate of allergic children.

Acknowledgements

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – REPRODUCTION I

O098

Quality of chicken semen cryopreserved with different N-methylacetamide concentrations combined with trehalose

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The study aimed to assess the effect of different N-methylacetamide (MA) concentrations combined with trehalose on the quality of post-thawing chicken semen. Forty adult Hubbard male chicken breeders were used. All chickens were housed in individual cages at 20 °C, exposed to a 14L:10D photoperiod and fed a commercial standard chicken diet. Males were divided into four different groups (10 males/group). All the ejaculates collected within one group were pooled together, then splitted into three aliquots, each one assigned to one treatment. Semen collection was repeated on three days, 36 semen samples (12 replicates/treatment) were collected into graduated tubes and volume and concentration were recorded. Semen aliquots were diluted to a concentration of 1.5×10^9 sperm/mL using a Lake pre-freezing modified extender added with 0.1M trehalose. The diluted semen was cooled at 4 °C for 30 min and transferred to the laboratory. MA was added into semen aliquots to obtain 6 (M-6), 3 (M-3) and 0% MA final concentration (M-0 treatment) and 1×10^9 sperm/mL working concentration. Semen was packaged in straws (0.25 mL), frozen for 10 min over a nitrogen bath at 3 cm of height and thawed in water bath at 38 °C for 30 s after 7 days. Sperm cell membrane integrity (SYBR-14/PI staining) and motility (SCA system) were assessed in both fresh and thawed semen. The recovery rates (%) of undamaged and motile cells after freezing/thawing were also calculated. Analysis of variance was performed considering time and MA concentration as sources of variation. Sperm quality parameters were significantly ($p<.001$) decreased by the freezing-thawing process and the overall mean values were consistent to those reported in scientific reports. Significant differences ($p<.05$) were found among the MA treatments: membrane integrity and sperm motility were improved in semen cryopreserved with 6% MA (15 ± 0.9% and 21 ± 1.3% respectively) compared to 3% (7 ± 0.9% and 15 ± 1.3%) and 0% (1 ± 0.9% and 14 ± 1.3%) MA. Several sperm kinetic parameters and the recovery rates of undamaged, motile and progressive motile sperm followed the same trend, presenting the highest value in M-6 treatment. The present results confirmed that the treatment with 6% MA is the most effective to prevent sperm membrane cryodamage and motion impairment.

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O099

Influence of germ line chimerism on chicken reproductive traits

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Germline chimaeras are one of the best tools for studying bird embryo development, as well as the transgenic bird's production or endangered species. The production of germline chimaeras involves the incorporation of exogenous primordial germ cells (PGCs) into the endogenous gonadal tissue of the recipient embryo. The experiment described here was designed to study the effect of germline chimerism on reproductive traits of chicken. Chimaeras were created from purified PGCs isolated from gonads of 5.5-day old donor Green-legged Partridgelike embryos and injected into blood vessels of 3-day old recipient White Leghorn embryos. Two hundred and nineteen chickens hatched from 327 injected embryos (incubation rate – 67%), and 217 of them (99%) were raised until maturity, and germline chimerism was identified by PCR method. In 34 germline chimaeras – CH group/19 cocks and hens 14/, identified by PCR method, and in 18 control birds – C group (not manipulated), raised and housed in the same environmental conditions, 41 reproductive traits were investigated. For statistical evaluation of the results, two-way analysis of variance (ANOVA) followed by Duncan's multiple-range test was applied. Chimerism significantly ($p\leq.01$) affected 9 of the studied traits. In most cases, they concerned the male traits. Sperm volume, sperm concentration, and percentage of live normal spermatozoa (0.338 mL; 365.8×10^6/mL; and 94.6%, respectively) were lower in chimaeras, compared to the control group (0.427 mL; 452.0×10^6/mL; and 97.6%). This impact affected also some kinematic parameters of motile spermatozoa, analysed using a computer-assisted sperm motility analysis (CASA) system. Especially, linearity coefficient (LIN), straightness coefficient (STR), and mean amplitude of lateral head displacement (ALH). In case of hens, only the per cent of late dead embryos was significantly higher (($p\leq.05$) in CH group (25.8), compared to C group (16.7). In addition, one hen and two sterile cocks were identified. Our study indicated that germline chimerism may affect some reproductive abilities of chicken chimaeras. Possible reasons for the observed reproductive traits depression were discussed in the light of PGCs competition in the recipient birds.

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O100

How can the honey improve the post-thaw quality of turkey spermatozoa?

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The most feasible biotechnologies for *ex situ* conservation of genetic resources in avian species is semen cryopreservation. However, research on turkey semen cryopreservation is not yet satisfactory to facilitate a turkey semen cryobank. Therefore, there is a clear need to standardise the freezing process to improve the post-thaw quality of turkey spermatozoa.

The most important factor that may affect the success of cryopreservation is the choice of the cryoprotectant (CPA). Recently, bee honey has been used as a non-permeable (NP) CPA with satisfactory results for sperm cryopreservation in some mammals. Thus, the goal of this study was to investigate the effect of different concentration of bee honey as NP-CPA on the cryosurvival of turkey semen.

Pool of semen (8–10 ejaculates/pool) were collected from toms (BUT) (Amadori group). Each pool was pre-extended (1:1; v/v) with Tselutin extender (TE) and cooled at 4 °C for 25 min. The semen was split into four aliquots and diluted (1:1; v/v) with four different freezing extenders composed of TE, containing 20% dimethylsulphoxide (DMSO) and 0, 1, 2 or 5% of honey. The diluted semen was packaged in 0.25 mL plastic straws and equilibrated at 4 °C for 20 min, then the straws were frozen by exposure at 10 cm above the liquid nitrogen level for 10 min. Lastly, the straws were dipped in liquid nitrogen at −196 °C. Semen samples were thawed at 50 °C for 10 s. Sperm motility (CASA system), DNA integrity (Acridine Orange), sperm viability and osmotic resistance (SYBR-PI) were assessed.

The freezing/thawing process caused a significant reduction of semen quality compared to fresh semen ($p<.05$). However, the post-thawing semen quality was affected differently in relation to the honey concentration used. Higher value ($p<.05$) of sperm viability (43.5 ± 1.3), total and progressive motility (35.6 ± 2.3, 3.9 ± 0.4 respectively) were found in semen frozen with the 1% of honey in respect to that with only DMSO and 5% of honey.

In conclusion, these data show that the use of 1% honey improves the cryosurvival of turkey spermatozoa. The benefits of the honey on sperm cryopreservation is due to the capacity to minimise the ice crystals formation inside the cytoplasm of sperm responsible for cryodamage. The development of an effective freezing protocol will lead to the take-off of a turkey sperm cryobank and preserve the genetic resources of autochthonous poultry breeds in Italy.

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O101

Thawing rate effects on the cryosurvival of Mediterranean brown trout spermatozoa

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The Mediterranean trout, *Salmo cettii*, is listed in Annexe II of the EU Habitats Directive and is included as 'critically endangered' in the Italian freshwater fish Red List. One of the most effective tools to avoid the extinction is the semen cryobank and, in this regard, the development of a successful sperm cryopreservation protocol is needed. Therefore, the purpose of the present study was to evaluate the effects of two different thawing rates on the *in vitro* and *in vivo* sperm quality of *S. cettii* populations of Molise.

Native trout were captured by electro-fishing. Semen was collected from 40 males to obtain five total pools, each diluted 1:3 (v:v) with a freezing extender composed of 0.3M glucose, 10% DMSO and 10% egg yolk. The semen was loaded in 0.25 mL straws and equilibrated at 5 °C for 10 min. The straws were frozen through the exposure at 5 cm above the liquid nitrogen level for 10 min, dipped and stored in the liquid nitrogen. Straws were thawed at two different thawing rates 30 °C for 10 s and 10 °C for 30 s, using a water bath. The sperm parameters evaluated were: motility and movement duration, viability (SYBR-14, PI) and DNA integrity (Acridine Orange). Fertilisation trials were performed using three groups of eggs ($N \simeq 100$) inseminated with: (a) fresh sperm (control group); (b) sperm thawed at 30 °C for 10 s; (c) sperm thawed at 10 °C for 30 s. The data obtained *in vitro* showed that the freezing process impaired the post-thaw sperm quality compared to the fresh semen ($p<.05$). In addition, we also found significantly higher motility and viability ($p<.05$) in sperm thawed at 10 °C for 30 s than the 30 °C for 10 s.

The fertilisation and hatching rates were significantly higher in fresh sperm (73.26 ± 5.17 and 68.89 ± 5.51) respect to the frozen semen. However, the best eyed and hatched rates were found using semen thawed at 10 °C for 30 s (58.6 ± 2.8% and 54.5 ± 2.8%) than sperm thawed at 30 °C for 10 s (32.9 ± 4.9% and 29.9 ± 4.4%) ($p<.05$).

In conclusion, the use of low thawing rate (10 °C for 30 s) improved the semen fertilisation ability of *S. cettii*. Our findings provide an important contribution for the creation of a sperm cryobank aiming at the restoration of *S. cettii* in Molise. Natural reproduction of native trout occurs on the spawning grounds at the main springs of Volturno and Biferno rivers ($T \simeq 10$ °C). Thus, the encouraging results at the thawing temperature of 10 °C, would facilitate the on-field artificial reproduction of wild breeders, using directly the spring water.

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LIVESTOCK SYSTEMS – AQUACULTURE

O102

Effects of rearing density on growth, welfare indicators and digestive conditions of gilthead sea bream (*Sparus aurata*, L. 1758) fed different fish meal and fish oil dietary levels

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In Mediterranean aquaculture, the improvement of feeding management represents a key factor to maximise productivity and improve sustainability. For this reason, the relation between feed composition and relevant farming parameters such as rearing density might be investigated and used to choose the best feed according the specific rearing conditions. This study explored the effect of high and low rearing density on growth, plasma biochemistry, skin mucus, gut enzymes activity and gut microbiome structure in Gilthead sea bream (*Sparus aurata*, L. 1758) fed low and high fishmeal (FM) and fish oil (FO) dietary level. Two isoenergetic diets with high and low FM/FO percentages, (30/15 and 10/3, respectively) were tested in triplicated fish groups until a final high (30 kg/m^3) and low (10 kg/m^3) biomass density. Fish (initial body weight: 96.15 g) were fed to satiation twice a day (10% overfeeding) over 97 days period. At the end of the trial, specific growth rate (SGR), voluntary feed intake (VFI), feed conversion rate (FCR), protein efficiency ratio (PER), gross protein efficiency (GPE), lipid efficiency ratio (LER), gross lipid efficiency (GLE) were estimated. Fish welfare was evaluated through plasma biochemistry and gut microbiome structure. Furthermore, skin mucus enzymatic activity was analysed. Two-way ANOVA was performed on data, followed by a Tukey's multiple comparison test. Differences among treatments were considered significant when *p*-value was <.05. According to the results, density seemed to negatively influence feed intake. Diet also had a significant effect on final weight, weight gain, growth rate and FCR, being higher in high FM/FO diet. Highest peroxidase and protease level were obtained in skin mucus from fish fed low FM/FO diet. Enzymatic activity of digestive enzymes and gut microbiota by Next-generation sequencing were also analysed. In conclusion, rearing density negatively affected feed intake but did not compromise fish health.

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O103

Pre-fattening of Manila clams in a Po Delta lagoon: effect of stocking density in suspended lanterns

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This study assessed the growth and mortality of Manila clams (*Tapes philippinarum*) (initial weight, 0.04 g) during a 3-month pre-fattening (April–June 2018) in suspended net lanterns at three stocking densities (low, 10,000 clams/m^2; medium, 20,000 clams/m^2; high, 30,000 clams/m^2; 21 lanterns/stocking density). Lanterns were located in two sites of Sacca degli Scardovari (Rovigo, Italy) characterised by different hydrodynamism. A total of 21 lanterns (7 per stocking density) were located at the northern site, which was at the most far and inner side of the lagoon with respect to the sea water entrance (average water temperature 23.4 ± 2.99 °C; dissolved oxygen: 6.18 ± 1.23 ppm). Other 42 lanterns (14 per stocking density) were located at the western site, at half of the lagoon (temperature 22.6 ± 3.20 °C; dissolved oxygen 7.12 ± 1.13 ppm). At the end of the pre-fattening (10 and 11 weeks in the northern and western sites, respectively), a subsample (about 50 g) was collected from every lantern to measure clam biometric traits. Data were analysed by PROC GLM (SAS), with stocking density as the main effect. In the northern site, after 10 weeks of pre-fattening, clams exhibited an average weight of 0.50 ± 0.23 g, a length of 13.45 ± 2.46 mm, and a width of 9.38 ± 1.48 mm. Clam growth varied as stocking density increased (p<.001): weight decreased from 0.75 g to 0.55 g to 0.47 g from low to medium to high stocking density; length from 15.6 mm to 13.9 mm and 13.2 mm, and width from 10.8 mm to

9.66 mm and 9.18 mm. Mortality was 1.8%, 0.9%, and 1.4% in lanterns kept at low, medium, and high stocking density, respectively (p>.10). In the western site, after 11 weeks of pre-fattening, clams exhibited an average weight of 0.64 ± 0.31 g, a length of 14.13 ± 2.46 mm, and a width of 10.25 ± 1.66 mm. Clam growth also varied as stocking density increased: weight decreased from 0.88 g to 0.62 g to 0.45 g; length from 16.1 mm to 14.3 mm and 12.7 mm, and width from 11.5 mm to 10.2 mm and 9.23 mm (p<.001). In this site, mortality increased from 1.0% to 4.3% and 8.9% as stocking density increased (p>.10). To conclude, under our conditions in Sacca degli Scardovari, clams in suspended lanterns successfully reached the minimum sowing size (0.3 g, length of 11 mm) in 3 months. Nevertheless, the increase of stocking density decreased clam growth and size in both sites.

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O104

Integrated freshwater aquaculture: potentialities and limitations of freshwater bivalves utilisation

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Aquaculture productions significantly differ from other sectors of land animal productions for the direct relation with aquatic ecosystems. Reduction of environmental impact of aquaculture is a central question and integrated marine aquaculture has presented good responses. Freshwater natural ecosystems are more vulnerable to aquaculture impact respect marine ones for their limited extension; therefore, the reduction of environmental impact is a mandatory question for the future of aquaculture in Italy and in Europe. Integrated marine aquaculture systems are based on co-production of two or more aquatic edible species, where fish are reared in combination with marine bivalves (such as blue mussels) and edible algae. Freshwater bivalves are optimal candidates for freshwater integrated aquaculture, but are investigated to little extent as they are not considered suitable for human consumption. The department of veterinary sciences of Torino university in 2017 has won an EU project financed (AQUAVAL) about freshwater integrated aquaculture and bioremediation. From May to November 2018, two consecutive experimental trials have been organised, in order to test the bivalve filtration efficiency in artificial conditions, using output water from a productive farm, in order to test the efficiency of bivalves in reducing bacteria concentration. Twelve aquaria have been equipped with 4 bivalve densities (60; 30; 15 and 7.5 kg/mc) and a control (without bivalves), in order to test the optimal density. In the second experimentation, the highest bivalve rearing density (60 kg/mc) was substituted with a lower density of 3.75 kg/mc. Despite some negative aspect have emerged during these experimentations, first results indicate a net bacteria reduction of almost 100% in 7.5 kg/mc density, one order of magnitude lower than the control. In the successive phases of the project, a bivalve meal will be tested as a potential ingredient of fish feeds and bivalve shell will be used as Calcium source in laying hens feeding. Finally, the possibility of bivalve utilisation as living sensors for water monitoring will be tested. This method, currently used in drinking water treatment plants, will be eventually transferred to aquaculture, in order to promote an innovative concept of hybrid integrated aquaculture.

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O105

In vitro evaluation of macroalgae (*Ascophyllum nodosum* and *Lithothamnium calcareum)* and microalgae (*Schizochytrium* spp.*)* for animal nutrition

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The growth of food demand and the scarcity of lands and water have led to interest in novel sustainable food and feed ingredients. Algal biomass as feedstock or as supplements in animal nutrition can positively improve the food security and environmental impact. Algae can be grown cheaply without arable land, they release oxygen into atmosphere and, if included in the diet of ruminants, are able to reduce methane emissions. Algae are characterised by high content of nutrients, minerals, complex carbohydrates, pigments and poly-unsaturated fatty acids. Although the nutritional profiles of algae vary considerably within species, different studies showed general antioxidant and antibacterial properties.
The aim of this study was to evaluate the antibacterial activity against O138 *E. coli*, a typical enteric pathogen of swine, and the antioxidant proprieties of *Ascophyllum nodosum, Lithothamnium calcareum* and *Schizochytrium* spp. Algal extracts were obtained starting from 5 g of dried algal flour samples, dissolved in 150 mL of acetone, were extracted in a Soxhlet apparatus for 6 h. After the evaporation of solvent under vacuum at 50 °C, the residue was dissolved with 20 mL of water. The overnight cultures of O138 *E. coli* in Luria-Bertani (LB) medium served as the inocula for

the experiments. Liquid culture-based growth inhibition assay with *E. coli* was performed with several concentrations of the algae extracts (in a ratio v/v ranging from 1:5 to 1:20). The growth rate of *E. coli* strains was monitored through the measurement of absorbance by spectrophotometer at optical density of 600 nm, from 0 hours to 6 hours of incubation at 37 °C. Algal extract with 10 mL LB liquid medium and 10 mL LB were used as negative controls, while 10 mL LB with *E. coli* was used as positive control. The antioxidant activity was tested by ABTS assay.

A. Nodosum showed a dose-response effect on the antioxidant properties and bacterial growth inhibition ability, particularly after 3h of incubation ($p<.001$). *Schizochytrium* spp. did not inhibit the bacterial growth even if it demonstrated dose-response antioxidant properties ($p<.001$). Our experimental conditions did not show any bioactivity related to *Lithothamnium calcareum* extract. In conclusion, algae could be considered as functional innovative raw material to improve the health status and performance of animal production.

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ANIMAL PRODUCTS– MEAT PRODUCTION, MANAGEMENT AND MARKETING PERSPECTIVE

O106

In vitro cultured meat: an Italian consumer survey

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Various studies have linked the meat industry with a negative impact on the environment, animal welfare and sustainability, while FAO has predicted a ~73% increase of meat consumption worldwide by 2050. *In vitro* cultured meat (IVCM; also known as lab-grown meat) has been proposed as an alternative solution. In brief, the technology is mimicking the process of regeneration of tissues after trauma and of embryonic myogenesis to obtain a differentiated cell capable of proliferating and differentiating into muscle fibres. As a result, meat is produced in lab without the need of slaughtering the animals.

Our objective was to investigate the attitude of the Italian consumers in terms of acceptance and perception of the in vitro cultured meat. We conducted an exploratory analysis on a web-based (via Google Forms) survey ($n = 736$). Six questions were considered: (1) have you ever heard about IVCM, (2) would you agree to its marketing, (3) would you taste it, (4) would you be willing to buy it at the same market price of conventional meat, (5) if a greater and positive impact on sustainability is shown, would you be willing to pay a higher price and (6) does its artificiality worry you.

Results showed that less than a quarter (22.3%) of the Italian consumers were aware of the IVCM. Despite this, 39.8% agreed with its marketing. Although 59.6% were worried about its artificiality, 69.3% were willing to taste it. Moreover, 41.1% were willing to purchase at a higher value compared to conventional meat if a greater and positive impact on sustainability compared to conventional meat could be proven. Given that the current population under 30 will be the main meat consumers in the future, we subset the samples <30 years old ($n = 354$) and re-analysed the data. Results were similar concerning the general knowledge of IVCM, but greater values observed for its marketing (50.9%) and willingness to pay at a higher price towards sustainability (45.5%). Although 49.4% were worried about its artificiality ~78% of the people <30 years old were willing to try it.

Our survey indicated that a large part of the Italian consumers is not yet familiar with a food product that is expected to be released in the market in the following years. Despite this, they appeared willing to try and pay for the IVCM, especially if it is related to higher sustainability. Further studies to shed more light into the Italian consumer's perceptions towards IVCM are recommended.

O107

Consumer test with salami from 'Umbria' medium-heavy pork, fed traditional or experimental diet with hazelnut perisperm

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The aim of this study is to assess the effect of hazelnut perisperm inclusion in finishing diet of pigs on the consumer's evaluation of salami. Twenty light-heavy pigs were divided into two groups and fed for 78 days a control diet (C) or an experimental diet (S) including 11% hazelnut perisperm (HP). After this period, the pigs were slaughtered and the meat was processed in two salami types, which differed only in the pigs' diet. On these salami, a Central Location Test (CLT) was carried out by about 100 untrained consumers, on three different steps: blind test (B), expectation test (E) and informed test (I), in this order. In B and I tests, consumers had to try slices of both salami types and evaluate Aroma (AR), Taste (TA), Colour (CO), Texture (TE) and Overall Liking (OL) on a 9-point hedonic scale; while in E test consumers had to evaluate only the OL, without tasting.

Statistical analysis of the data was carried out with a factorial model that included all the socio-demographic characteristics, feeding system (T) of the pigs, C or S, test conditions (A), B, E and I, and T x A interaction. To test the influence of information on sensory parameters (OL only), according to the type of salami offered to consumers, the χ^2 test was used.

People involved in CLT, coming from five different Italian regions, were mostly male (56%), middle-aged (between 45 and 64 years – 51%), graduated (36%), with elementary occupations (16%) or students (12%). The majority of participants in the test, eats cured meat more than once a week (44%), mainly at home (78%), and buys it sliced (49%) at the supermarket (53%).

Socio-demographic features of participant in the test had no significant influence on the results.

The results of the factorial analysis showed a significant interaction ($p<.05$) between test conditions and C salami testing: particularly, the mean of OL in B test was higher than the mean of OL in E test. The analysis of the effect of information on the acceptability, analysed only on OL, showed significant positive disconfirmation for the C salami ($p<.0001$), confirming that in B consumption was better than expected after information (E test). In conclusion, the use of HP in swine feeding did not penalise the taste of the salami and the information about its use and of its characteristics does not adversely affect the expected and the informed OL of the consumers involved.

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O108

Effect of dietary liquorice extract on the oxidative status of rabbit burgers

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The aim of this study was to quantify the effect of liquorice extract dietary administration on rabbit burgers oxidative status. For this aim, 60 rabbits fed a commercial diet (C) or C diet with 6 g/kg of liquorice extract (L) for 6 weeks. In the experimental group a liquorice extract (*Glycyrrhiza glabra* L.; Nutraceutica Srl, Monterenzio, BO, Italy) containing 18–22% of glycyrrhizin acid water solution was used. At weaning (5 weeks) rabbits were randomly selected, divided into two feeding groups and fed with the two dietary treatments. At 12 weeks of age, rabbits were transported to a slaughterhouse where after fasting and electro-stunning, were slaughtered by cutting the carotid arteries and jugular veins. Carcases were dissected according to the World Rabbit Science Association. Hind leg meat (*Biceps femoris*) of each group were trimmed (to reach 3 kg); the minced meat of each batch was further manually homogenised and shaped into 30 patties/treatment weighing 20 g each. The fatty acid profile, antioxidant content and lipid oxidation (TBARs) were determined and Atherogenicity and Thrombogenicity indexes calculated. A one-way ANOVA analysed the effect of dietary liquorice on the considered data. For the correlation analysis, Proc CORR was used (STATA-SAT®).

The burgers from L rabbits showed the highest content of α,γ-tocopherol, together with a reduction of Retinol and α-tocotrienol. According to these data and the higher antioxidant magnitude of α-tocopherol, the amount of peroxidative processes were lower in the experimental group (0.09 vs. 0.14 mg MDA/kg; $p<.05$). The same samples consequently showed the highest amount of polyunsaturated fatty acid (PUFA) and, significant differences were observed on the C20:5n-3 and C22:6n-3 contents (7.03 vs. 8.09 and 8.09 vs. 9.49 mg/100 g meat respectively for C and L group). Moreover, the L group had lower Atherogenicity (0.51 vs. 0.55; $p<.05$) and Thrombogenicity (0.98 vs. 1.07; $p<.05$) indexes, due to the higher n-3 PUFA concentration. Another interesting result was related to the correlation between TBARs and C22:6n-3 content; in the C group, the correlation index was −0.08 (n.s.), while in the experimental group it was equal to −0.44 ($p<.05$), reflecting a more linear trend between oxidation and polyunsaturated fatty acid content. Other studies are in progress in order to investigate the relationships between dietary supplementation and addition of liquorice extract directly in the burgers along the storage period.

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O109

Effects of garlic powder and salt additions in rabbit meat burgers

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The consumption of rabbit meat, commonly sold as whole carcase or at least as cut-up parts, is nowadays decreasing. Ready-to-eat products, like burgers, represent an important percentage of food production and they are sold as only meat or meat mixed with other ingredients, mainly spices and salt. In order to increase rabbit products relatability in the last year, few research studies were conducted on burgers manufacturing and on antioxidant compounds. One of the most used spices worldwide and very commonly used in rabbit culinary recipes is garlic. Salt, due to its antimicrobial properties could preserve meat products and also it could influence the activity of some enzymes responsible for flavour development. The objective of this research was to evaluate the influence of garlic powder and salt in rabbit burgers during a storage period of 7 days at 4 °C. Thus, meat quality, lipid oxidation and antioxidant capacity and fatty acids profile were determined on raw and cooked samples.

Four different formulations were produced: control (only meat), meat added with garlic powder (0.2%), meat added with salt (1%), and meat added with both garlic powder (0.2%) and salt (1%). Meat was mixed with the assigned ingredient and a total of 120 burgers of 100 g were formatted.

Burgers were statistically tested via two-way ANOVAs with formulation and storage time as main factors with Tukey's test as post hoc ($p<.05$). Garlic powder modified partially the colour (mainly b*) of the burgers and increased partially the antioxidant capacity (DPPH) of the raw samples during time ($p<.05$). Addition of salt modified the colour (mainly a*) and induced an increase in lipid peroxidation (TBARS) in raw burgers ($p<.001$). Moreover, further differences were highlighted in lipid peroxidation of cooked burgers in relation to the formulation and the storage time ($p<.001$). The burgers with both garlic powder and salt showed miscellaneous results. Mixing garlic powder and salt could be a potential practical application as the results partially showed that garlic could contrast the negative effects of the salt.

O110

Dietary effect of dried bay (*Laurus nobilis*) meal on rabbit meat quality

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Laurus nobilis (commonly called laurel or bay leaves) is known in the human field especially as food spice, rich in polar compounds such as phenols, flavonols and flavones having antioxidant and antimicrobial effects. The aim of this study was to evaluate the effect of dietary supplementation with dried bay leaves in growing rabbits on chemical composition and fatty acid profile of meat. The experiment lasted 56 days and was performed on 120 rabbits; at weaning (35 ± 2 days of age), the animals were divided into 4 groups (n. 30 per group) based on different dietary treatments. All rabbits were fed a basal diet. The experimental groups were: a negative control (CON) that received the basal diet without supplementation with dried bay leaves; a positive control group (CF) that received a 2.5% fat-enriched diet (pig fat in addition to the 2.6% of crude fat in basal diet); a supplemented group (SB) that received 1 g/kg feed of dried bay leaves meal; a combine-supplemented group (SBF) that received fat-enriched diet and dried bay leaves in the same doses used in CF and SB group. The dietary supplementation with dried bay leaves (SB and SBF) did not affect the gross (moisture, crude protein, ether extract, ash) composition, the pH value, the content in fat-soluble A and E vitamins and lipid oxidative stability of meat, whereas it reduced ($p<.05$) the cholesterol content in comparison with the positive control (CF). Concerning the fatty acid profile of meat, saturated fatty acids content decreased ($p<.001$) in the bay leaves groups SBF and SB compared to CON and CF groups; monounsaturated fatty acids were improved ($p<.001$) in SBF and SB groups compared to CF group, whereas polyunsaturated fatty acids were markedly higher ($p<.001$) in the experimental SB group compared to CON, CF and SBF groups. In conclusion, the dietary supplementation of dried bay leaves improved the lipid profile, particularly in the contents of cholesterol, and monounsaturated and polyunsaturated fatty acids, without affecting the chemical traits and oxidative markers of rabbit meat.

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O111

Evaluation of *Pectoralis minor* gaping defect in broiler chickens

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In the recent years, the occurrence of a new quality issue affecting the *P. minor* muscle such as separation of fibre bundles in different positions (gaping) has been signalled by a certain number of broiler abattoirs located in different Countries around the World. Thus, after defining the criteria to classify the affected muscles as Moderate (MOD) or Severe (SEV) cases, the incidence of gaping under commercial conditions was assessed on a total of 8.600 broiler chickens belonging from 43 flocks during a six-month period. In addition, 72 *P. minor* muscles (24/group) were selected to evaluate the impact of gaping defect on the main quality traits (pH, colour, drip and cooking losses, and shear force), proximate composition (moisture, protein, lipid, ash and collagen) and thermal properties. The total incidence of *P. minor* displaying gaping defect (16.8%, being 8.8 and 8.0% MOD and SEV affected cases, respectively) evidences that this defect represents an important quality issue as affected muscles are usually downgraded because of their impaired appearance. The analyses of the main quality traits and technological properties evidenced that the occurrence of gaping is associated with significantly higher ($p<.01$) lightness values coupled with remarkably lower ($p<.01$) ultimate pH and impaired water holding capacity of the meat. Indeed, a remarkable increase ($p<.05$) in cooking loss was found as the severity of the gaping defect increased. These quality traits overlap with those previously observed in pork *semimembranosus* muscles affected by destructuration and have strong similarities with the PSE-like condition formerly described in chicken meat. In addition, no significant differences among the groups were found concerning proximate composition and the enthalpy of thermal shrinkage associated to the intramuscular connective tissue thus suggesting that the occurrence of this defect is not associated to an alteration of the histological features of the muscle. In conclusion, the findings of the present study evidenced that nowadays gaping defect can represent an important quality issue which can result in economic losses for the broiler industry in view of the increasing consumer preference toward *P. minor* cut-up (commercially referred to terderloin or tender). This defect likely develops because of the biochemical processes taking place during the *post-mortem* conversion of muscle to meat and its severity might be related to the operations carried out during deboning.

O112

In ovo delivery of GOS in slow-growing broiler chickens exposed to heat stress: implications for meat quality traits

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Beside from commercial broilers, chickens are the extremely rich source of genetic diversity and could have different reaction to microbiome stimulation by *in ovo* delivery of prebiotics. The aim of the study was to evaluate carcase and meat quality traits in slow-growing chickens stimulated *in ovo* with trans-galactooligosaccharides (GOS) and exposed to heat stress. Fertilised eggs from Hubbard JA57 chickens were incubated. On the day 12 of egg incubation, 3000 eggs were divided into: prebiotic group (GOS) injected with a single dose of 3.5 mg GOS/egg, saline group (S) injected with physiological saline and control group (C) un-injected. After hatching, 900 male chicks were reared in floor pens: three groups (GOS, S and C, 6 pens/group, 25 birds/pen) reared in thermoneutral conditions (TN) and three groups (GOS, S and C, 6 pens/group, 25 birds/pen) reared under heat stress conditions (HS). Chronic heat stress (30 °C) was applied from day 36 to 50. At 50 days of age, 15 randomly chosen birds/treatment/temperature were slaughtered and pectoral muscle (PM) was removed for the analyses. Data were analysed by GLM procedure in a 3×2 factorial design: treatment and temperature were the main factors. Treatment had no influence on carcase weight. GOS reduced ($p<.05$) PM weight and its yield as compared to C group. As expected, temperature had a strong effect on carcase and PM weight, significantly lower in HS group. Muscle pH was higher ($p<.05$) in GOS group compared to C. Meat from GOS group showed a lower lightness and a higher redness compared to C and S groups. WHC, proximate composition, cholesterol and intramuscular collagen were not affected by treatment. Heat stress increased moisture ($p<.01$) and reduced protein ($p<.05$) content. Cholesterol and collagen contents were not affected by temperature. Neither fatty acid composition (total saturated, monounsaturated and polyunsaturated fatty acids) nor nutritional indices of meat were significantly affected by treatment and temperature. Significant interactions between factors were observed for carcase weight, protein and lipids contents, and for n6/n3 ratio. In conclusion, *in ovo* injection of the GOS prebiotic had no negative effect on physico-chemical and nutritional properties of meat produced by slow-growing broiler chickens reared under intensive conditions, but reduced weight and yield of breast muscle. Heat stress has a negative and marked effect on carcase traits and reduced the protein content.

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O113

^{1}H-NMR metabolite profiling of broiler breast meat affected by muscular abnormalities

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Nuclear Magnetic Resonance (NMR) spectroscopy represents a versatile technique that allows gathering, in a single spectrum, quantitative and qualitative information on the metabolic profile of a tissue. In the present study, ^{1}H-NMR was applied to quantify free amino acids, histidine compounds and metabolites involved in energy-generating pathways in *Pectoralis major* muscles affected by growth-related abnormalities (White Striping – WS, Wooden Breast – WB and Spaghetti Meat – SM). The study was conducted on a single flock of 48 days old broiler chickens (Ross 308, females, average live weight 2.8 kg) classified into the following groups: Normal (NORM), WS, WB and SM. The increased (p<.001) concentration of free amino acids observed in WS, WB Q15 and SM likely results from the degenerative processes taking place within these muscles as well as from a concurrent attempt of the tissue to overcome necrosis and repair the damaged fibres. This is corroborated by the significantly higher (p<.001) glutamine and glutamate content found in WS, WB and SM previously observed in *mdx* mice and dystrophic muscles and hypothesised to be related to the intense regenerative processes associated to these conditions. In addition, significantly higher (p<.001) glycine and branched amino acids (namely valine, leucine, isoleucine and proline) were found in WS, WB and SM thus suggesting an increased collagen synthesis and an intense extracellular matrix remodelling within these muscles. With regard to the histidine compounds, the lower (p<.001) levels of anserine and carnosine found in abnormal muscles confirms that oxidative stress can contribute to the aetiology of breast muscle abnormalities in broilers. Besides, WS, WB and SM exhibited significant variations in the metabolites involved in energy-generating pathways (lower lactate, creatine and inosine-5-monophosphate) thus supporting the assumption of metabolism perturbations associated to the development of these conditions. However, since no distinctive metabolites were found, similar causative mechanisms leading to WS, WB and SM can be hypothesised. In conclusion, the adoption of ^{1}H-NMR spectroscopy allowed to corroborate earlier findings (i.e. oxidative stress and perturbation of energy metabolism) and provide novel insight (i.e. higher content of free branched amino acids) into metabolic pathways involved in the development of growth-related abnormalities which are nowadays of great importance for the broiler industry.

ANIMAL BREEDING AND GENOMICS – SMALL RUMINANTS GENETICS

O114

VarGoats international initiative, a 1000 goat genomes project

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Goats were domesticated ~10,000 years ago from their bezoar wild ancestor *Capra aegagrus*. These founder animals spread out from domestication centres in Europe, Asia, and Africa during the next few thousand years, which resulted in many populations becoming locally adapted to diverse environmental constraints such as heat, altitude, disease resistance, etc. After a very long period of soft selection for production traits, the situation changed dramatically with the emergence of the breed concept about 200 years ago. Selection pressure was strongly increased, and gene flow between populations (or newly formed breeds) was seriously reduced, leading to the fragmentation of the initial gene pool. Currently, there are more than 1000 goat breeds worldwide, 18% of which are either threatened or already extinct.

Through the generation of domestic breeds sequence data and their analysis together with existing data of domestic and wild animals, VarGoats collaborative project aims at becoming the first step of a 1000 goat genomes project. VarGoats Consortium is coordinated by the Institut National de la Recherche Agronomique INRA (France), and includes an international team of partners (Laboratoire d'Ecologie Alpine, France; Parco Tecnologico Padano, Italy; Agricultural Research Service, USDA, USA; International Goat Genome consortium; African Goat Improvement Network; Roslin Institute and the Centre for Tropical Livestock Genetics and Health, UK). At present, a dataset containing 829 goat genomes has been assembled, including 815 sequences from 84 local and transboundary domestic populations, and 14 genomes from 7 wild goat species. Bioinformatic infrastructures for data storage and sharing are collecting the generated sequence data and making it available to VarGoats participants. Starting from the ADAPTMAP project experience, thematic working groups are in charge of performing data analyses to (i) explore genetic diversity, (ii) study domestication, (iii) detect adaptation and selective sweeps, and (iv) discover variants (SNPs, CNVs, structural variants, causal mutations) in various goat breeds, with the final goal to develop breeding solutions. Hybridisation between species is also under investigation, thanks to the availability of sequences from various *Capra* species. At the end of the project, data will be released in a public database.

O115

Copy Number Variations in Sicilian goat breeds inferred from GoatSNP50 BeadChip

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Copy number variations (CNVs) are structural variations of genome in the form of losses or gains of DNA fragments, ranging from kilobases to several megabases in size, that displayed variable copy numbers in comparison with a reference genome. There is increasing evidence that CNVs not only are important source of genetic variation but have significant influence on phenotypes such as coat colour and productive and reproductive traits. The aim of this research was to investigate the presence and distribution of CNVs, inferred from GoatSNP50 BeadChip, in four Sicilian goat breeds (Girgentana, Argentata, Messinese and Derivata di Siria). PennCNV software was used to identify CNVs in a total of 120 individuals using Log R Ratio and B allele frequency of 50,619 autosomal single nucleotide polymorphisms (SNPs) as input data. Seven individuals did not show CNVs, as well as chromosome 22. After filter applications on raw CNV calls, we identified 702 putative CNVs in 108 individuals, consisting of 202 deletions (LOSS) and 500 duplications (GAIN). Chromosome 10 showed the highest number of CNVs, i.e. 70 CNVs, while chromosomes 2, 20, 24 and 25 showed only one CNV each. The CNVs were merged into 160 CNV regions (CNVRs) spanning ~35 Mb of total length and corresponding to ~1.2% of the goat genome. We found 64 CNVRs with GAIN, 85 with LOSS, and 11 MIXED (with both deletions and duplications). Moreover, 85 CNVRs were unique, i.e. present in only one breed, and seven CNVRs have frequency >20%. Gene enrichment analysis was conducted within these seven CNVRs using Genome Data Viewer – NCBI Genome Browser with ARS1 assembly of *Capra hircus* genome. We found some important genes overlapping with these CNVRs, which are involved in local adaptations, such as coat colour and olfactory receptors, and immune response (e.g. ADAMTS20, EDNRA, OR7, T-cell receptor and GAL-9B). Even though this represents a preliminary study, our results on Sicilian breeds could be considered reliable since similar representation of CNVRs in immune-related genes, coat colour and OR genes was reported previously in livestock species, included goat.

O116

Coagulation ability of goat milk from 4 breeds predicted by Fourier-transform infra-red spectroscopy

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The aim of this study was to investigate the prediction reliability of traditional and modelled milk coagulation properties (MCP) applying Bayesian models on the Fourier-transform infra-red spectroscopy (FTIR) spectra of individual goat milk samples from 4 breeds. Milk samples from 770 individual goats reared in 22 farms were collected. Goats belonged to: Camosciata delle Alpi, Murciano-Granadina, Maltese and Sarda (CA, MG, Ma and Sr, respectively). The following traits were measured using a lactodynamograph (60 min): (I) traditional MCP (RCT, min; k_{20}, min; a_{30}, a_{45} and a_{60}, mm) were recorded; and (II) 240 individual measures of curd firmness (CF, mm; 1 every 15 s) were collected and used to estimate modelled MCP (RCT_{eq}, min; k_{CF} and k_{SR}, %/min; CF_p, mm; and t_{max}, min). For each milk sample, 2 spectra (absorbance) in the infra-red region (5011 to 925 cm^{-1}) were available and averaged before data analysis. A Bayesian approach was implemented to develop calibration models by using the BGLR (Bayesian Generalised Linear Regression) package in R software. Three different training-testing calibration procedures were adopted: (i) spectra of the entire data set were used, 80% for calibration and 20% for validation (randomly selected), repeated 10 times per each trait; (ii) spectra of each breed were separately used for calibration and validated on an external set of the remaining spectra; (iii) calibrations produced in (ii) were applied on each spectra set of each other individual breed. In all the prediction procedures, the observed and the predicted values of the testing subset were used to calculate the root mean square error of validation ($RMSE_{VAL}$). Using the entire dataset, the results obtained were comparable with those available in literature for bovine and ovine milk, exhibiting the highest performance of prediction for RCT and a_{60}, and for RCT$_{eq}$ and CF$_{p}$. Among breeds, MG (for RCT, RCT$_{eq}$, a_{30} and a_{45}) and CA (for k_{20}, a60, k_{CF} and t_{max}) calibrations showed the lowest RMSE$_{VAL}$ values when validated on the remaining spectra data set (calibration ii). The results obtained in the calibrations (iii) showed that the validation performances on each breed were not always accurate, with RMSE$_{VAL}$ higher than those obtained for the calibrations of the entire data set. Since the interest of the dairy goat industry on selection for milk with best MCP, a breed factor needs to be considered for the use of FTIR predictions at the population level.

O117

Identification of copy number variations in Sicilian sheep breeds

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Copy number variants (CNVs) are an important source of genetic variation complementary to single nucleotide polymorphisms (SNPs). CNVs are classes of polymorphic genomic regions including deletions, duplications and insertions of DNA fragments from at least 0.5 kb up to several Mb. Many CNVs represent neutral polymorphic variants, whereas several others cause or are associated with both Mendelian and complex traits. The aim of this study was to identify CNVs in five sheep breeds reared in Sicily (Valle del Belice, Comisana, Pinzirita, Barbaresca and Sarda) and performed a CNV-based population genetics analysis. Illumina OvineSNP50K BeadChip data and SVS Golden Helix software were used to identify CNVs and CNV regions (CNVRs) within the genome of 575 animals. We identified 12,026 CNVs, which can be merged into 1,014 CNVRs, spanning 169.58 Mb of total length and corresponding to 6.9% of the sheep genome. A total of 254 genes were found within the 101 CNVRs used for cluster analysis. Gene Ontology and KEGG pathway analyses showed that several of these genes are involved in olfactory transduction, neural development, fatty acid degradation, T-cell receptor signalling pathway.

Our results also indicated that segregating CNV express a certain degree of diversity across all breeds. The clustering based on matrices estimated from CNV counts has made evident the separation into more or less distinct groups, even if they are breeds belonging to the same geographical area, except the Sarda breed, and with low genetic differentiation. This is the first comprehensive study of the CNV population genetic properties in Sicilian sheep breeds. Additional studies are needed to evaluate the effects of these CNVs on phenotypic and production traits in Sicilian breeds and their usefulness in defining relationships among breeds.

O118

A genome-wide association study for somatic cell count in dairy sheep

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Mammary infection causes high somatic cell count (SCC) and results in important leading to economic loss in the dairy industry and therefore any method to reduce the incidence of mastitis would be welcomed. Genome-wide association studies (GWAS) have become a powerful tool to identify genomic regions associated with important traits in livestock. The aim of this study was to carry out a genome-wide association study (GWAS) to identify genetic loci associated with SCC, an indicator trait of mammary gland inflammation. A total of 481 Valle del Belice sheep were genotyped with OvineSNP50K Genotyping BeadChip and associated with SCC phenotypes. The phenotypic data included 5586 test-day for the trait considered. SCC were log10 transformed before the statistical analysis. After quality control performed using GenABEL package, 469 ewes and 37,228 SNPs were retained for the analysis. In order to handle multiple measures, the GWAS analysis was done using RepeatABEL package. Briefly, a mixed model including fixed (parity, litter size, lambing season, days in milk, herd by test-day interaction) and random effects (animal, permanent environmental across lactation) is fitted in a REML analysis. The variance component estimates were used to estimate the covariance structure of the data and used in a generalised least square fitting the fixed effects and the SNPs to be tested. Significant genome-wide and suggestive thresholds based on Bonferroni correction were of 1.34×10^{-6} and 2.68×10^{-5}, respectively. Fifteen SNPs located within or close to known genes were found to be associated with SCC. In particular two SNPs (rs401848358 and rs415331743) on OAR2 reached the genome-wide threshold and were found within genes PIP5K1B that are involved in the vitamin D pathway and ZNF618 that participate in numerous physiological processes, including cell proliferation, differentiation and apoptosis. Two SNPs (rs425714078 and rs430803312) on OAR14 and one SNP (rs398865179) on OAR22 were located within two known QTLs for SCC (id14000 and id14007) identified in an Awassi × Merino backcross population. Three SNPs on OAR3 (rs428821105, rs411731094 and rs409654401) and three SNP on OAR8 (rs411955601, rs422733508 and rs409088383) were found associated with SCC. A QTL associated with SCC was found in a previous study in Chios dairy sheep breed using the same genotyping array. Finally, four SNP located on chromosomes 5, 17, 19 and 22 reached the suggestive threshold for SCC.

O119

Regional heritability mapping approach to identify genomic regions associated with milk production traits in dairy sheep

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Regional heritability mapping (RHM) is a variance component approach that has been suggested to be more powerful than standard GWAS in capturing genetic variation, by combining the effects of both common and rare variants in a region. The aim of this study was to carry out an association analysis using RHM for milk production traits: milk yield (MY), fat yield and percentage (FY, F%), protein yield and percentage (PY, P%), in 481 Valle del Belice dairy sheep, genotyped with the OvineSNP50K Genotyping BeadChip. Each chromosome (OAR) was divided into regions of 100 adjacent SNPs, and the genetic variance attributable to each region estimated. Briefly, a mixed model including fixed (parity, litter size, lambing season, fortnights in milk) and random (additive genetic, permanent environmental within and across lactation, herd-test-day interaction and environmental) effects, was used. The additive genetic effects had two variance components: (i) a whole polygenic component modelled with a genomic relationship matrix (GRM) constructed with all SNPs in the genome; and (ii) a regional genomic component modelled with a GRM constructed with the SNPs within the region. The significance of the regional component was tested using a likelihood ratio test (LRT) against a null model with no variance associated with the tested region. The Bonferroni corrected thresholds were 13.48 and 9.20, for genome-wide ($p<.05$) and suggestive (one false positive per genome scan) significance levels, respectively. A region on OAR2 was found significant at the genome-wide level (LRT =16.25) for F%, with an adjacent region reaching the suggestive threshold. Three more regions (on OAR3 and 20) reached the suggestive threshold for F%. The same two regions on OAR3 reached the genome-wide threshold for P% (LRT =13.86 and 16.27). The genomic regions on OAR3 include several QTLs identified in Spanish Churra sheep and SNPs within known candidate genes for the considered traits. Genomic regions identified in the present study provide an opportunity to further develop genetic improvement programmes aiming at increasing milk production traits in the Valle del Belice dairy sheep.

O120

Influence of genotype on meat quality in kids

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The research meant to evaluate carcase characteristics and meat quality traits of kids from three genotypes bred in Southern Italy. The trial was carried out in a farm located in Basilicata on 15 male kids born as singletons of three genotypes: Garganica (G), Maltese (M) and Siriana (S) that were fed only milk, suckling from their dams until they were about 21 days old. Afterwards, the kids were weaned with a pelleted feed administered *ad libitum* and slaughtered at 60 days of age. Dams grazed for about ten hours during the day and received hay *ad libitum* and a commercial feed (500 g/head/day) at housing, in the evening. Genotype did not affect growth performances. As for slaughtering data, a significantly lower net cold dressing percentage was recorded for M kids (56.42%; SEM =3.723) in comparison with G (64.22%; $p<.01$) and S (62.51%, $p<.05$). In M kids the dissection of the right half carcase into meat cuts showed a significantly ($p<.05$) lower incidence of the loin (5.78% vs. 6.86 and 7.06%, respectively for S and G breeds; SEM =0.711) and leg (30.15%; SEM =0.388) as compared to S (29.69%; $p<.01$) and G (32.21%; $p<.05$). Separation of the leg and the loin into their tissue components did not show any significant difference between genotypes as for the proportion of lean, fat and bone. No significant differences aroused between breeds for pH and meat colour features of the *Longissimus dorsi* (Ld) and *Semimembranosus* (Sm) muscles. Shear force (WBS) of the *Ld* muscle of G kids was significantly lower ($p<.05$) compared to M and S breeds (5.15 vs. 7.95 and 7.27 kg/cm2, respectively; SEM =1.553) while no effect of breed was recorded on tenderness of the *Sm* muscle. The total collagen content assessed on the *Ld* muscle showed no significant difference between genotypes. Meat lipid oxidation, measured by 2-thiobarbituric acid reactive substances (TBARS), showed that *Ld* meat stored for 10 days at 4 °C had a significantly ($p<.01$) higher value of malondialdehyde content (mg MDA/kg meat) in the G breed compared to S and M (0.77 vs. 0.28 and 0.38, respectively; SEM =0.206).

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – ANIMAL WELFARE I

O121

Time-based feed restriction and group size in growing rabbits: effects on health status and growth performance

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The effects of the feeding programme (R, time-based feed restriction vs. L, *ad libitum*) and group size (from 6 to 32 rabbits/group) were evaluated on 368 crossbred rabbits from 31 to 73 days of age. In R group, the access time to feeders was reduced during the 1st week from 14 to 8 h/d, maintained at 8 h/d in the 2nd week, and then increased by 1 h/d during the 3rd and 4th week up to 24 h/d. Rabbits were housed in cages or pens with different dimensions and group sizes, i.e. conventional cages (8 cages of 0.33 m^2, 6 rabbits/cage), small pens (8 pens of 0.50 m^2, 8 rabbits/pen), medium pens (1.00 m^2, 16 rabbits/pen), and large pens (4 pens of 2.00 m^2, 32 rabbits/pen). Individual data of live weight (LW) and daily weight gain (DWG) were analysed by PROC MIXED (SAS), with the feeding programme, the housing system and their interaction as main effects and with the cage or pen as a random effect. Cage and pen data for feed intake (FI) and feed conversion (FC) were analysed by PROC GLM (SAS), with the feeding programme, the housing system, and their interaction as main effects. Mortality and morbidity rates, and health risk index (HRi) (mortality + morbidity) were analysed by the PROC GENMODE (SAS). L rabbits evidenced epizootic enteropathy (ERE) during the first two weeks on trial, whereas R rabbits fell ill only in the following two weeks during refeeding. In the whole trial, however, R rabbits reached a higher HRi as compared to L rabbits (16.3% vs. 11.9%; $p<.05$). During restriction, R rabbits exhibited lower FI (94 vs. 123 g/d), DWG (47.4 vs. 56.2 g/d), and FC (1.97 vs. 2.21) in comparison with L rabbits ($p<.001$) whereas during the refeeding period, R rabbits achieved higher DWG (52.2 vs. 47.4 g/d) and better FC (2.97 vs. 3.19) than L rabbits ($p<.001$). In the whole trial, R rabbits showed significantly lower FI (141 vs. 149 g/d), and FC (3.04 vs. 3.12), as well as lower DWG (46.5 vs. 47.7 g/d) and final LW (2695 vs. 2750 g) than L rabbits ($.001<p<.05$). Mortality rate tended to increase with increasing group size (3.4% in cages and pens with 6 and 8 rabbits vs.. 10.9% in pens with 16 and 32 rabbits; $p=.06$) without effects on growth performance. In conclusion, time-based feed restriction significantly improved feed efficiency but had some negative effects on health status and reduced final live weight of group-housed growing rabbits.

O122

Veal calves' abomasal lesions and rumen mucosa alterations investigated post-mortem: is there any way to differentiate 'good' from 'bad' farms?

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Several years of research on abomasal lesions and rumen mucosa alterations and on the predisposing and risk factors in intensively reared veal calves have not overcome the problem. Abomasal lesions, in particular, are the most frequent post-mortem findings (70–93% of animals). Since this problem is one of the major welfare concerns, this study aimed at assessing the prevalence of abomasal lesions and rumen mucosa alterations at a commercial slaughterhouse as strategic observation point and at differentiating 'good' from 'bad' farms.

The post-mortem assessments were carried out by a veterinarian who evaluated abomasa and rumens after their emptying, without interfering with the regular procedures and schedule of the slaughterhouse. Abomasa and rumens of the first 15–16 animals per batch were evaluated. The occurrence of lesions on torus pylorus and in the pyloric area of the abomasa was recorded as a binary. Lesions in the pyloric area were also classified according to 3 size classes: 1 = lesions with diameter <0.5 cm^2, 2 = 0.5–1 cm^2, and 3 = >1 cm^2. The presence of plaques (multiple patches with coalescing papillae covered by a sticky mass and hair), hyperkeratosis (hardened rumen papillae), and underdeveloped rumen mucosa (almost no papillae in atrium and ventral and dorsal rumen) were recorded as binary. Herd-level prevalence were calculated and submitted to descriptive statistics.

Results of this study regard a total of 653 abomasa and 653 rumens (15.9 ± 3.1 organs/batch (mean ± SD)) from 41 batches slaughtered in 13 days. The slaughter batch size was 60.6 ± 21.5 and carcase weight 162.7 kg ±23.4. Lesions on torus pylorus and in the pyloric area were present in 84.7%±15.1 and in 92.5%±10.0 of abomasa observed, respectively. Over 77% of abomasa presented at least a large lesion (size class 3). Plaques, hyperkeratosis, and underdeveloped mucosa involved 68.2%±21.6, 60.1%±22.6, and 11.1%±14.4 of rumens, respectively. The calculations of the quartiles for all the variables and of the correlations between variables (overall and for the batches in the worst quartile with the highest prevalence) allowed to differentiate farms, contextually at measure level. This differentiation of farms at slaughter could be used as an iceberg indicator to effectively address the on-farm welfare assessments and the investigation of risk factors in those with severe problems.

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O123

Relationships between farm productivity and housing space allowance. An analysis on a sample of Italian dairy farms

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The respect of animal welfare in productive contests is generally associated with ethical issues, then it is considered as a public good whose generates costs that cannot be completely covered through the market. Coherently, policies are mainly addressed to impose rules for livestock management, while giving financial incentives to farms to assure high levels of animal welfare. However, it was widely proved that chronic effects of stress from poor welfare can lead to adverse metabolic changes for animals, which in turn can have significant effects on their health and productivity.

The objective of this work was to evaluate housing space allowance, as a key animal welfare criterium, in terms of those benefits it provides to farms (e.g. increased milk yield and/or reduced total production costs). We focussed our attention on this topic, following literatures evidencing housing density among the major environmental stressors, that significantly affect freedom of movement, resting times and incidence of lameness of cows.

Using a sample of 121 dairy livestock, located in the plain of four North Italian regions, the analysis tried to explore the economic consequences of housing conditions which potentially influence the animal welfare.

Data were collected in 2015 from the Italian Farm Accountancy Data Network. A gross housing space allowance indicator (GHS) was calculated as total m^2 of stables over the number of livestock standard unit (LSU). GHS' distribution has a median of 7.3 m^2, a mean of 8.5 m^2 and a s.d. of 5.0 $m^{2.}$

Several regressions were performed to estimate the relationships between GHS and different proxies of farm performances, including other independent variables to take farm and herd characteristics into account. As dependent variables we used: (1) the milk yield per cow/year (2) the labour costs per LSU (3) a 'cost coverage ratio' (CCR), proxied by the ratio of the milk yield value per LSU over the fixed costs per LSU, measured by summing up labour cost and stable depreciation charge.

Main results indicated that GHS is positively correlated with productivity but evidencing a non-linearity around 14 m^2, where the relation becomes negative. Regards to costs, GHS shaped a convex curve, showing first decreasing and then increasing relation, with turning point around 15 m^2. Finally, a concave curve was shaped for the relationship with CCR, with turning point of 14 m^2, suggesting positive net effects of animal welfare on livestock economic performances.

O124

Transcriptome profiling of ruminants fed with a micronutrient-supplemented diet

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Iodine (I) is a micronutrient essential for the proper thyroid functionality in mammals. I requirement is dependent by the life stage and milk constitutes the main source of I intake for infant. For this reason, in dairy ruminants, the calibration of I is fundamental to keep the performance high. However, the effects of I supplementation on gene expression in dairy cow are not well documented. Thus, to improve knowledge in this field, we have assessed the effect of I supplementation dairy cattle using RNA-sequencing. In detail, we have randomly divided 22 dairy cows into two groups: the first group (CTR, 11 animals) received a customised diet with physiological level of I_2 (20 mg/day/animal of I2) while the second one (I, 11 animals) received the same diet with a I_2 supplementation (20 + 65 mg/day/animal of I2) for 8 weeks (56 days). At the end of supplementation, the whole blood from both groups is been collected and the transcriptome analysis is been performed. Filtering our data using a FDR <0.05, we identified 525 differentially expressed genes (DEGs) and in particular there were 274 and 248 genes downregulated and upregulated respectively in I group compared to the CTR one; they were sufficient to discriminate the two groups on a heat map scale but also on a hierarchical clustering analysis indicating the robustness of the experimental strategy. Next, we used our DEGs to evaluate the enrichment for any GO and thus identify a number of genes that share the same characteristic. Thus, we found that the most enriched pathway was associated with antigen processing and presentation o via MHC class I (GO:0002474), glycolytic process (GO:0006096), oxidation-reduction process (GO:0055114), regulation of cell proliferation (GO:0008285) indicating that iodine could positively influence the immune response and the productive performance of the animals. Moreover, our findings contribute to the growing body nutrigenomics research in ruminants.

O125

Post calving gene expression of whole blood leukocytes in Simmental and Holstein cows following pegbovigrastim treatment

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Pegbovigrastim is a commercial long-acting analogue of bovine granulocyte colony-stimulating factor (rbG-CSF) that promotes increase count and functionality of polymorphonuclear cells in dairy cows around parturition. We hypothesised that rbG-CSF administered to periparturient cows at approximately 7 d before parturition and at the calving day affects gene expression profiles involved in the leukocytes function. Blood was collected on d 3 after calving from treated group (PEG; 13 Holstein and 13 Simmental cows) that received rbG-CSF (pegbovigrastim; Imrestor; Elanco Animal Health), and control group (CTR; 13 Holstein and 13 Simmental cows). Blood from all cows was sampled from the jugular vein in PAXgene tubes (Preanalytix, Hombrechtikon, Switzerland) for RNA extraction and consequently to perform RT-qPCR analysis for mRNA expression. Normalised data were subjected to MIXED model of SAS (ver. 9.4) with treatment, breed, and their interaction as fixed effects. After calving, whole blood leukocytes of PEG cows showed higher expression of genes involved in recognition and immune modulation (*CD14*, *CD16*, *MYD88*, *TLR2*, and *TLR4*), cell adhesion (*ITGB2*, *ITGAL*, *TLN1*, *SELL*, *SELPLG*, and *CD44*), antimicrobial activity (*MMP9*, *LTF*, *LYZ*, and *LCN2*), and inflammatory cascade (*CASP1*, *TNFRSF1A*, *IL1B*, *IL1R*, *IL18*, *IRAK1*, *NRLP3*, and *S100A8*) compared to CTR group, suggesting an improvement of migration, adhesion, and antimicrobial ability, and an enhanced inflammatory response that in turn triggers immune cells activation. On the other hand, pegbovigrastim treatment led to a lower expression of *RPL13A*, *IL8*, *TLN2*, and *TNFA* compared to cows not treated. Overall, Simmental cows resulted in higher expression of *IDO1*, *RPL13A*, *CD44*, *CX3CR1*, *ITGB2*, and *TNFA* compared to Holstein cows, whereas expression of *CD16* and *TLR2* was lower expressed. Treatment with pegbovigrastim increased most of the genes involved in the processes investigated that suggests a complete activation of the immune machinery against the critical period of the post-partum. In addition, we shed light into some differences for the breed. The immune system of Simmental cows seems to be more responsive to the stimulation by pegbovigrastim, as revealed by the up-regulation of genes involved in immune system adaptation, suggesting differences in the post-calving adaptation of breeds.

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O126

Effects of the nutritional status before of slaughter on the transcriptome and differential expression analysis in rainbow trout (*Oncorhynchus mykiss*)

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The aim of the present research was to study the effects of the nutritional status of rainbow trout (*Oncorhynchus mykiss*) before the slaughter process on the post-mortem indices (rigor mortis, shell life, etc.), body quality characteristics and transcriptome expression analysis (RNAseq). Fish groups, each consisting of 40 specimens (695 g), were kept at 13.2 ± 0.1 °C and fed to apparent satiation with a commercial diet (Control), with a restricted feed intake (RR, 30% of control) and starved (R0) for three weeks before slaughter. Total RNA was purified using the Aurum total RNA fatty and fibrous tissue kit (Bio-Rad, USA) and mRNA was extracted from the muscle tissue ($n = 5$). The RNA integrity and concentration were measured with an Agilent Bioanalyzer 2100. Construction of RNA-seq libraries, barcoding and sequencing were performed using an Illumina Genome Analyser (HiSeq 2500) using 150 bp paired-end reads and 3 replicates for each biological sample. The Galaxy pipeline was used to perform the RNAseq workflow. RNA-seq reads were aligned to a previously published reference transcriptome and genome of *O. Mykiss*. ExpHTS-processed RNA-seq reads were quantified from each

sample using Salmon. EdgeR and DESeq2 software packages were used to conduct differential expression analyses. The Trinotate and Blast2GO suite pipelines were used to generate functional annotations for contigs. GO enrichment was visualised in REVIGO. In this study, several markers of nutrient deprivation were identified. Mapping reads to the reference genome identified new additional transcripts not annotated in the original reference genome. Statistical analysis of the RNAseq data and differential expression profiling led to the identification of distinct transcriptional clusters. In agreement with the proteolytic and lipolytic responses to starvation described in mammals, a large set of genes were overexpressed in the muscle of fasted trout. This study has demonstrated that muscle gene expression profiling after slaughter is modulated by the animal nutritional status.

JOINT PIG SESSION

O127

Genetic diversity and signatures of selection in Italian pig breeds and wild boars revealed by whole-genome resequencing

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Natural and artificial directional selection in cosmopolitan and autochthonous livestock and wild relative populations have shaped their genomes defining the level of variability and determining selective sweeps as final adaptation to different environmental conditions and production systems. In this study, we analysed the genetic variability and selection signatures in eight Italian pig breeds (three cosmopolitan breeds: Italian Large White, Italian Landrace and Italian Duroc; and five autochthonous breeds: Apulo-Calabrese, Casertana, Cinta Senese, Mora Romagnola and Nero Siciliano) and in Italian wild boars. For each breed or population, DNA pools were prepared using equimolar DNA from 30–35 animals. Whole genome resequencing (pool DNA-seq) was obtained using paired-end Illumina sequencing at 35–40X. About 800 M read pairs were mapped with BWA-MEM on the Sscrofa11.1 genome version and variants were detected with CRISP (a pool-seq variant caller) coupled to an ad-hoc bioinformatic pipeline. We detected about 20.5M of variants (15% not included in dbSNP yet), 1% of which affected gene products. Selection signature analyses were carried out by computing the Fixation Index (Fst) and the Pooled Heterozygosity (PH) statistics on 100 kbp genome windows. Putative selective sweeps regions were identified considering the 99.98th percentile of the related distributions (extreme lower end). Fst and PH analyses detected selection signatures in more than 40 genome regions on 10 out of 18 porcine autosomes (SSC). Several selective sweeps were already reported by other studies and included genes affecting body shape/size (e.g. *PLAG1* on SSC4, *LCORL* on SSC8 and *CASP10* on SSC15 and coat colour (e.g. *KIT* on SSC8). These results might provide a first global variability analysis in Italian *Sus scrofa* populations and have identified several potential causative mutations that could be associated with phenotypic traits that differentiate the studied pig breeds and wild boars.

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O128

Mora Romagnola pig breed production chain: *MC1R* and *NR6A1* polymorphisms can link the Herd Book to a DNA based system useful for the authentication of its pork products

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A sustainable strategy for the conservation of animal genetic resources is based on the commercial exploitation of their productions through the development of mono-breed labelled chains of meat and dairy products. These products are usually sold at a higher price compared to undifferentiated ones contributing to assure profitability to the farmers. As a drawback, the market added value attracts fraudsters. Therefore, the development of methods that can authenticate mono-breed products is a key issue for a successful construction of a mono-breed production chain. Mora Romagnola is one of the six Italian autochthonous pig breeds under a conservation programme managed by the National Pig Breeders Association (ANAS). This breed is mainly raised in the eastern side of the Emilia-Romagna region. In 2017, the Herd Book of this breed accounted a total of 339 registered sows and boars. Mora Romagnola pigs have a typical black–brown coat colour. In this study, we genotyped almost all Mora Romagnola registered sows and boars (290 pigs) for polymorphisms in two genes affecting morphological traits (the melanocortin 1 receptor gene or *MC1R*, for coat colour; and the nuclear receptor subfamily 6 group A member 1 gene or *NR6A1*, for vertebral number). The final aim was to set up a DNA based protocol that could link the registration of the Mora Romagnola pigs to its Herd Book and the possibility to authenticate Mora Romagnola pork products. At the *MC1R* gene, all genotyped Mora Romagnola pigs carried alleles *E*+ (wild type) or *e* (recessive red) apart from five animals that carried the dominant black *ED2* allele. At the *NR6A1* gene, only two pigs carried the wild type allele. All pigs carrying the *MC1R ED2* and the *NR6A1* wild type alleles were eliminated from the Herd Book because their genetic profiles were not compatible with the breed specific genotypes at these two loci. Based on these results and actions at the Herd Book level, we designed a DNA based system to authenticate pork products that could be assigned to Mora Romagnola only if the analysed meat has compatible genotypes at the *MC1R* and *NR6A1* genes. To our knowledge, this is the first example in a livestock breed for which it was possible to establish a direct relationship between the Herd Book registration of the animals and a DNA based method to defend its production chain from frauds.

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O129

Effectiveness of *Bacillus amyloliquefaciens* or *Bacillus subtilis* to counteract *Escherichia coli* F4ac infection in weaned pigs

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Probiosis is considered a valuable strategy to prevent the post-weaning diarrhoea (PWD) and to reduce the antibiotics' use in pigs. This study investigates the effect of Bacillus amyloliquefaciens DSM25840 (BAA) or Bacillus subtilis DSM25841 (BAS) supplementation on gut homeostasis of post-weaning pigs susceptible to enterotoxigenic *E. coli* F4ac (ETEC) and infected with this pathogen. Sixty-four post-weaning pigs (BW 7.7 kg ±0.6 kg) were randomly allocated to 4 groups and fed: control diet (CO); CO $+1.28 \times 10^6$ cfu/g of BAA; CO $+1.28 \times 10^6$ cfu/g of BAS; CO +1 g colistin/kg of feed (AB). At day (d) 7, pigs were challenged with 10^5 cfu/mL of ETEC. Pigs' health status and faecal score were daily recorded until d21. Pigs were weekly weighted. Blood was collected at d8 and d21 for metabolomics analysis. The intestinal functionality was investigated by quantification of cells apoptosis and mitosis and by microarray analysis of the jejunum mucosa at d21. CO had a higher number of pigs with severe impairment of health status and excluded (CO: 4) from the trial compared to the other groups (BAA:2; BAS:0; AB:0). CO had a more impaired faecal score than AB and BAS (at d14) ($p<.05$). AB improved BW at d7, d14 and d21, average daily weight gain (ADWG), average daily feed intake (ADFI) and gain to feed ratio (G:F) from d0 to d7 ($p<.05$). CO had a higher level of plasma arginine, lysine, ornithine, glycine, serine and threonine than other groups ($p<.05$) on d8 and a lower level of glycine and glutamine than BAS on d21. Gene sets related to adaptive immune response were enriched in jejunum of BAA, BAS and AB. CO had enriched gene set for nuclear structure and RNA processing indicating an increase of intestinal cells proliferation. The mRNA data were not supported by the immunohistochemistry results that showed a lower mucosal villus mitotic index in CO than in BAS and AB groups ($p<.01$) probably due to the different timing of activation of these parameters. In conclusion, dietary supplementation with *Bacillus subtilis* DSM25841 may inhibit the effects of ETEC F4ac infection in weaned piglets by modification of amino acid metabolism reducing the catabolic insult leading to muscle breakdown and by improving the intestinal health and the immune response, which provides a rationale for the reduced diarrhoea of the pigs.

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O130

Relationship between *in vivo* skin discolouration and carcase skin lesions in Italian heavy pigs

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Pre-slaughter handling of pigs involves several stressful procedures which can lead to *in vivo* skin discolouration and carcase skin lesions. The presence of these latter defects is not only an indication of poor animal welfare but also has great implications on economic returns due to a depreciation in carcase value. *In vivo* skin discolouration (dark red patches on some parts of the body) is extensive and hence easy to detect at the farm and during the pre-slaughter handling. Skin lesions (scratches or bite marks) on the other hand becomes apparent only after pigs have emerged from the scalding tank.

The aim of this study was to determine the relationship between *in vivo* skin discolouration and carcase skin lesions.

A total of 1680 Italian heavy pigs [Duroc x (Landrace x Large White)] from the same farm were examined during unloading and in the race to stunning by a trained observer. The pigs were transported for a short distance in 12 deliveries in a truck composed of a lorry and a trailer, each one equipped with three hydraulic decks containing 23–24 pigs/deck. Pigs exhibiting dark red patches on the ear, thigh and back were recorded. At 30 minutes post mortem, skin lesions were scored on the front, middle and hind quarters for each carcase using a four point scale (1 = none to 4 = severe) provided by the Danish Meat Research Institute. Skin lesions score for the whole carcase was determined using the highest score assigned to each quarter. For each deck, the frequencies of *in vivo* skin discolouration and carcase with mild to severe skin lesions (≥2) were calculated. Spearman rank correlations between these frequencies were determined.

Significantly high and positive correlation coefficients in the range between r= +0.35 and +0.61, with $p<.0001$ were observed, in particular, between red thigh with middle and hind quarters (r= +0.51 and +0.61, respectively).

These results suggest that skin discolouration *in vivo* could be used to estimate the occurrences of carcase skin lesion.

Acknowledgements

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O131

Cinta Senese protein requirements from 30 to 65 kg: effects of different dietary protein levels on slaughtering traits and protein deposition indexes

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Four diets, differing for their crude protein content (120, 140, 160 and 180 g/kg as fed basis, named CP12, CP14, CP16 and CP18, respectively), were tested on 24 Cinta Senese castrated males to assess the optimal protein requirements in the growing phase. The experimental diets, formulated according to the ideal protein composition, were added with 2% of bentonite as internal standard, to asses diet's digestibility. Animals were weighed weekly along the whole trial. Simultaneously, on eight more animals, nitrogen excretion in faeces and urines were detected using metabolic cages. After slaughtering, the right carcase side was dissected in: head, neck, shoulder, ribs, loin and ham. The main cuts were weighed and divided in: subcutaneous fat, intermuscular fat, lean and bone. Tissues were analysed for proximate composition, whereas bone was discarded. Results on *in vivo* performances showed a negative impact of increasing crude protein levels on feed intake, average daily gain and protein conversion indexes. Carcase composition and tissues percentage were not affected by dietary treatment but, a difference between low (CP12 + CP14) and high (CP16 + CP18) protein diets was observed for fat and lean gain, as well as for protein conversion in lean and in lean protein. The nitrogen balance corroborated these results, showing a significantly higher loss of N through urine as the dietary CP levels increased. The overall results indicated that the CP12 diet was adequate to fulfil Cinta Senese protein requirements during growing.

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O132

Effect of free-range grass grazing on two pigs' genetic types

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A total of 20 male pigs, 10 Cinta Senese (CS) and 10 Large White X Cinta Senese (CS×LW), were reared outdoor in a single pen of 7 ha for all the fattening period. Animals fed natural pasture (grass) and crop residuals (regrowth of oats) plus a feed supplementation consisted in 1.4 kg/head/d of commercial mixture. Animals started the trial with similar live weight and no differences between slaughter weight were found. The CS group showed highest thickness of backfat in all three measuring points (first and last thoracic vertebra and Glutaeus medius muscle) as well as highest dressing percentage. As regard commercial cuts, CS showed higher percentage of fat cuts, except for jowl (p>.68) and lower of lean cuts (total fat cuts, 35.06 vs. 27.46% and total lean cuts, 59.43 vs. 66.36% for CS and CS×LW respectively). The greater adiposity of CS was also confirmed by sample cut dissection (p<.05) with exception of intermuscular fat (p>.05). The chemical traits recorded not significant differences both in Longissimus lumborum (LL) and in Psoas major muscles. As regard physical analysis, cooking loss (21.24 vs. 24.24 % for CS and CS × LW respectively) and WB on cooked meat (100.3 vs. 136.6 N for CS and CS × LW respectively) resulted different in Longissimus lumborum muscle. The fatty acid profile of backfat showed differences for total lipids, MUFA and PUFA content. CS group recorded the highest value of C18:1, influencing thus the whole MUFA family (46.29 vs. 44.98% for CS and CS×LW respectively). PUFA, both n3 and n6, resulted different between groups, with C18:2 (p<.01) and C18:3 (p<.0001) fatty acids higher in CSxLW group. Among saturated fatty acids, only C12:0, C14:0 and C16:0 resulted higher in CS group (p<.05). Crossbreeding is thus a reliable solution to produce meat with desired qualitative characteristics also for Cinta Senese sector.

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O133

Use of FT- NIRS to estimate subcutaneous fatty acid groups in autochthonous European pig breeds

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The amount and proportions of fatty acids determine the degree of unsaturation of the backfat and represent a key factor in the technological quality of processed meat. Among the methods developed for a reliable determination of fatty acid content, near infra-red spectroscopy could provide a rapid and no-destructive characterisation. Nevertheless, the use of backfat of different origins (genotype, gender and live weights) can represent a challenge in the real application of spectroscopy techniques.
The aim of the present study is to evaluate the use of FT-NIRS for predicting the amount of total fat and fatty acid groups (MUFA; PUFA; PUFA 3, 4, 6; SFA) on pig grounded muscles. The research considered 152 fresh samples of backfat collected from 12 European local pig breeds.
For every sample, lipids were extracted from subcutaneous fat and fatty acid profile was determined by a gas chromatograph. Two aliquots of each sample were scanned using FT-NIRS Antaris II model (Thermo Fisher Scientific). Mathematical pre-treatments (MSC, smoothing, 1st and 2nd derivate) were applied and outliers' spectra were identified and removed. The entire set was randomly split into a calibration (80%) and a validation set (20%) in order to have an independent dataset. Partial least square regression on the average spectrum was applied and the chemometric results are evaluated in terms of coefficient of regression and root mean square errors in calibration (R^2-RMSE) and validation (Rp^{2}-RMSEP).
The best results in terms of accuracy (RMSE) and explained variability (R^2) were obtained for unsaturated fatty acid groups (MUFA, PUFA), their ratio (PUFA/SFA) and PUFA 6. These parameters achieved R^2 higher than 0.96 in calibration and higher than 0.94 in validation showing a high predictability capacity of FT-NIRS. PUFA3 and PUFA 4 appear more difficult to predict by NIRS; in fact, in their equations R^2 is between 0.89 and 0.76. SFA achieved a R^2 of 0.86 that is slightly lower than values reported in other studies probably because of the large variability of genotypes used.
Hence, FT-NIRS is a valid tool to rapidly estimate fatty acid groups in pigs' backfat, whereas single fatty acid content will require greater dataset before having reliable estimates.

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O134

Predicted glycemic index in bakery/confectionary former food products and in former food based pig diet

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This study evaluated the predicted glycemic index (pGI) in former food products (FFPs), and in two pig compound feeds formulated with or without the inclusion of FFPs. Six samples of FFPs and two pig compound feeds were used. Former food products were based on bakery and confectionary ex-food, while the pig compound feed was formulated by substituting 30% of cereals with

FFPs (FeedFFP30%). All samples were analysed using an in vitro Englyst assay that simulates gastric and small intestinal digestion, and which has been proposed to determine the hydrolysis index (HI) and predict the glycemic index (pGI) of cereal-based foods introduced into pig diets. Corn meal, heat processed wheat, and a conventional pig compound feed (FeedCTR) were included as control feed ingredients. In the assay, white bread was used as reference material. Compared to white bread (i.e. reference material) the lowest HI value was observed in maize meal (81.05). In the tested FFPs, the average HI value was 104.53, while the same variable in compound feed containing or not FFPs, was equal (81.17 and 83.74, respectively). Predicted GI value, ranged from 84 to 114, indicating a higher pGI in most of the tested FFPs compared to cereals. Considering the two experimental pig compound feeds any difference was observed between FeedCTR and FeedFFP30% for the same variables that reached for both, HI and pGI values slightly above 80. Accordingly, two main groups of samples can be distinguished in the study, namely: low HI samples with HI lower than 100, including corn meal, FeedCTR, FeedFFP30%, flacked wheat and FFP4; and high HI samples including all the other FFPs tested. The same classification can be adopted for pGI. Thus, combining HI and pGI results suggests that most of the FFPs tested in the present study were characterised by a high glycemic index potential, which seems to be linked to the starch/sugars HI. The inclusion of FFPs in a commercial compound feed, and its effect on HI and pGI merits further investigations.

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – ANIMAL HEALTH

O135

Multivariate factor analysis of milk fatty acid profile: effect of different somatic cells count on nutraceutical composition of single udder quarter

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Milk fat concurs to define the quality of dairy products. Its composition is related to the metabolic activity of the mammary gland (MG). Somatic cell count (SCC) is an indicator of MG health, being related to both MG cell turnover and cells from immune system, according to the presence or not of mammary infections. The levels of SCC of single quarters of MG are usually not related because the four quarters are anatomically and physiologically distinct from each other. Alterations in the health status of MG can lead to changes in SCC values and changes in the quality of milk, including milk fat content.

The aim of the present work was to evaluate the relationship between the level of SCC and the fatty acid (FA) composition of milk from single quarters.

The study involved 49 cows of Italian Holstein, with an average milk production of 23 ± 5.80 kg/day. Milk samples were collected two times (30 days between the two samplings) per cow. Milk sample from each individual quarter was collected into a separate tube. Milk FA profile was obtained by GC-FID, whereas SCC level was obtained by electronic fluorescence-based cell counting. Data of FA composition and SCC were analysed by a multivariate factorial analysis (MFA).

MFA extracted five latent factors able to explain about 95% of the original variance. The first was associated with medium-chain FA and C18:1c9 and was called '*de novo* synthesis' factor. The second latent factor was associated with C16:0, C16:1c9 and C18:0 and was defined as 'lipid source'. The third showed the largest loadings for C18:2n6 and C18:3n3 and was called 'diet FA' indicator. Rumenic acid and C18:1 trans isomers were associated with the fourth common factor, that has been interpreted as an index of 'biohydrogenation'. Finally, the last factor was called 'short chain FA' because it was linked with C4:0 and C6:0.

SCC significantly affects the '*de novo* synthesis', 'diet FA' and 'biohydrogenation' factors. Values of SCC higher than 10 x 10^3 cells/mL were negatively associated with scores of 'de novo synthesis' factor. This result suggested that FA synthesis decreased with increasing levels of SCC. The consequence is the reduction of de novo synthesis FA, with the increase of blood FA uptake. Contemporary, a significant increase of the 'biohydrogenation' factor in the quarters with more than 400 x10^3 cells/mL was observed. Milk fat from quarters with high levels of SCC contained more FA mainly from adipose tissue mobilisation or rumen metabolism.

O136

Intramammary infusion of casein hydrolysate for involution of a single mastitic quarter for the remainder of lactation in dairy cows

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Mastitis in a single quarter can cause high somatic cell count (SCC), clinical signs, or death in dairy cows. Casein hydrolysate

(CH) is an intramammary treatment reported to induce mammary involution. Objectives were to study cessation of milk production after CH infusion of one mastitic quarter for the rest of lactation, subsequent 3-quartered cow milk production and SCC. After the next calving, both total-cow and previously treated quarters' milk production, SCC and bacterial cure were studied. Criteria for enrolment: target quarter SCC > 10^6/mL, SCC ≤400,000/mL in the 3 non-mastitic quarters, target quarter producing >20% (front) or >25% (rear) of total-cow milk, cow producing ≥22.7 kg milk/day, 75–190 days until next due date, and mycoplasma-negative. Cows were blocked based on lactation number (1st or 2nd-plus) and culture status (at least one pathogen isolated or no growth), and 3 treatments were randomised in a 2:2:1 ratio: CH, non–hydrolyzed casein (NHC), and ceasing milking only (negative; N). Target mastitic quarters were milked once per day and treated for 3 days, then not milked until next calving. Total-cow (3 lactating quarters) milk and SCC were measured 7–14 days later. Post-calving, total-cow and previously treated quarters' SCC and production were measured once, 10–21 DIM. Bacterial cure defined: all 3 weekly cultures during the first 21 DIM negative for a pathogen isolated pre-treatment. Of 40 cows enrolled, 12 were culled and 28 calved again (14 CH, 9 NHC, 5 N). Pre-treatment measures were the same among treatment groups. Decreases in cow SCC (−966,000/mL) and cow production (−14%) for the rest of lactation after treatment of the target quarter were significant ($p <.02$, ANOVA), but not different among treatments. After calving, total-cow production (33.2 kg), previously treated quarter SCC (1.41×10^6/mL) and quarter production (25% of cow) were not different among treatments. The CH and N treated cows had significant decreases in target quarter SCC after next calving (-2.76×10^6/mL and -5.32×10^6/mL; $p < 0002$). Bacterial cure proportion (14/16, 88%) was not different among treatments, chi-square. Casein hydrolysate infusion was associated with involution of mastitic quarters, reduced SCC for the remainder of lactation, and all quarters returned to milk production with reduced SCC following calving.

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O137

Production, oxidation traits and health of dairy ewes fed diets supplemented with fungus myceliated grains

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Mushrooms contain many bioactive compounds beneficial for human health. Polysaccharides from mushrooms exhibited immunomodulatory, antibacterial, antiviral and antifungal properties, as well as antitumor activity. Some mushrooms showed antioxidant properties due to phenolic acids, flavonoids, polysaccharides, carotenoids, ascorbic acid and tocopherols contents. These properties indicate the potential use of mushrooms as performance-enhancing feed additives for livestock. The effects of mushrooms-based diets have largely been investigated in the poultry species, whereas few studies have involved the ruminants. This study was undertaken to evaluate the effects of fungus myceliated grains (FMG) in diets fed to dairy ewes on intestinal parasite control, milk production, milk fatty acid (FA) profile, and cheese oxidative stability. For 8 weeks, 21 Valle del Belice ewes were divided into 3 homogeneous groups fed hay *ad libitum* and 1.3 kg/day of a concentrate containing faba bean (50%), barley (30%) and sorghum (20%), this latter as myceliated or non-myceliated grains; accordingly, the concentrate included FMG at levels of 20% (FMG20), 10% (FMG10), or 0% (FMG0). To prepare FMG, sterile grains were incubated with mycelia of selected mushrooms at 25 °C for 8 weeks, dried at 60 °C for 24 h and stored at 4 °C until used. During the trial, the FMG20 ewes showed comparable dry matter and nutrients intake, a reduction in intestinal parasite infection, a tendency to increase milk yield, and a higher ($p<.05$) milk casein (4.8% vs. 4.3% in both FMG10 and FMG0 ewes). In cheeses manufactured 3 times in the last week, the FMG diets induced higher lightness and redness, and less intense yellow colour. Cheeses from FMG20 milk showed a lower secondary lipid oxidation, indicated by TBARs values, and a higher antioxidant capacity, detected by the TEAC assay, suggesting a major oxidative stability of cheese fat due to antioxidant compounds transferred by FMG. The FMG diets did not affect the content of health-promoting polyunsaturated FA in milk, except for n-3 eicosapentaenoic acid, found only in FMG milk. Based on these results, the antiparasitic and antioxidant effects of FMG showed to enhance the health of ewes, milk yield, and the health properties and oxidative stability of dairy products. These encouraging results require to be confirmed by further investigations into the use of mushrooms or FMG as natural feed additives for enhancing health and production of livestock ruminants.

O138

Use of selenium-enriched olive leaves in sheep diets: effects on performance, oxidative status and milk and plasma selenium levels

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Selenium (Se) is an essential trace element used as a dietary supplement for its wide range of beneficial effects that are primarily due to its important role in antioxidant defences. In addition, dairy ruminant diets supplemented with Se show an increased milk Se level with potential benefits for the consumers. Valorisation of agro-industrial by-products is an important environmental challenge under the perspective of a circular economy and sustainable agro-food chain. Sixteen Sarda lactating ewes (4th month of lactation) were randomly divided into two homogeneous groups and fed one of the following isoenergetic and isonitrogenous concentrates (350 g/ewe/day) for 56 days: (1) a pelleted feed (OL) containing ground dehydrated olive leaves (204.0 g kg^{-1}); (2) as in OL, except that the olive leaves were obtained from the pruning material (by-product) of trees treated with selenate (Se content in leaves: 7.83 mg kg^{-1}) to reduce drought stress and increase oil Se content (OL-Se). Lucerne hay was provided ad libitum. Body condition score (BCS) and milk yield were determined every two weeks. Total Se concentration and other parameters (lipid, total nitrogen, casein, urea, lactose, somatic cells count) were evaluated in raw milk obtained from the morning milk. At the same time intervals, blood samples were collected and tested for Se concentration. Plasma samples were evaluated for haemoglobin, haematocrit, total protein, triglycerides, glucose, cholesterol, LDH, GOT, GPT and indicators of the oxidative status such as superoxide dismutase (SOD), glutathione peroxidase, glutathione reductase, ferric reducing antioxidant power and antioxidant capacity. Milk yield increased in the OL-Se group (1.58 vs. 1.46 L, $p<.05$). Se supplementation did not influence BCS (2.68 vs. 2.66) and milk composition. Overall milk (90.8 vs. 30.3 ppb, $p<.001$) and blood (143.8 vs. 99.2 ppb, $p<.001$) Se concentration were markedly increased in the OL-Se group. Plasma parameters were not affected by treatment, except for SOD that increased in the OL-Se group (1.04 vs. 0.60 U/mg Hb, $p<.05$). In conclusion, under the adopted experimental conditions, dietary Se-enriched olive leaves improved milk yield and its nutraceutical properties. The oxidative status of the lactating ewe was not largely affected by the presence of organic Se in the feed. Treating olive leaves with Se can be a valid strategy to adopt in the perspective of a circular economy.

O139

Effects of by-products on oxidative status, metabolic profile and performance in dairy sheep

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The effects of diets containing different dried by-products on milk production traits, metabolic and oxidative status of dairy sheep have been studied. Thirty-six Sarda dairy sheep, were assigned to 4 experimental groups and fed diet containing: no by-product (CTR), exhausted berries from myrtle (EBM), tomato pomace (TM) and dry grape pomace (GP). The trial lasted 8 weeks. Blood samples were collected before starting and at 45 days of experimental feeding. Milk was measured weekly. Milk yield, milk composition and biochemical, and oxidative parameters were influenced by the by-product included in the diet. The GP group produced more milk, protein, and fat compared to CTR group, whereas the EBM group produce less compared CTR (1050 vs. 1220 g/d). The TM did not influence milk yield and its composition compared to CTR.

Values of plasma biochemical parameters were within the physiological range for the species in all groups, demonstrating the good health conditions of ewes throughout the experiment. A higher antioxidant capacity measured either by ferric reducing antioxidant power (FRAP) or the 2,2'-azino-bis(3-ethylbenzothiazoline)-6-sulphonic acid (ABTS) radical scavenging in blood of GP ewes in comparison with the CTR has been observed. An improvement in the oxidative status of milk from GP was evidenced by lower malondialdehyde (MDA) and protein carbonyls (PCs) contents. The EMB group showed higher ABTS in blood and lower PCs in milk when compared with CTR ewes. In the blood of TM group an increase in the MDA content, in comparison with the CTR group was observed. In conclusion, the GP at tested amounts showed to be the most appropriate by-product to improve milk yield and to preserve the health status of animals. Finally, the results suggested that dietary GP in the dose used may have a beneficial effect against oxidative stress.

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ANIMAL BREEDING AND GENOMICS – NEW BREEDING TECHNOLOGIES II

O140

Demonstrating the value of genetic improvement in the Australian dairy industry

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ImProving Herds was an Australian dairy industry project initiated in 2015 to accelerate genetic gain and increase usage of herd improvement tools by turning complex science into simple, data-driven decisions that deliver profits to farmers.
Twenty-seven Focus Farms provided data, had heifers genotyped, and took part in economic data collection. Ten years of data (2007–2016) was used to compare performance of herd contemporaries of differing genetic merit. Each cow's margin over feed and herd costs (MOFH) was calculated by summing income from milk, calf sales and final salvage value and subtracting cost of rearing, feed and maintenance; costs associated with mating and mastitis events.
On average, the top 25% of cows (ranked on Balanced Performance Index, BPI; Australia's dairy selection index) had a $300/cow/year greater MOFH than the bottom 25%. The top 25% of cows produced 88 kg/cow/year more milk solids and lasted, on average, eight months longer in the milking herd. Additional milk income easily compensated for higher feed costs of high BPI cows. Daughters of bulls who rank highly on the BPI performed better under Australian conditions, across dairying regions and feeding systems.
The relationship between pre-calving genomic estimated breeding values (GEBV) and first lactation production records corrected for fixed effects was evaluated in the Focus Farms. A strong relationship between GEBV and corresponding production records was found which was similar to published national mean reliabilities for equivalent traits. After accounting for genotyping costs, selecting the top 50% of heifer replacements based on GEBV is worth ~$40/head more than using parent average. GEBV can be confidently used to guide heifer selection decisions.
The project is creating change in industry; 86% of survey respondents said their thinking about genetics had changed after seeing project results at field days. Many Focus Farms have also become advocates of genetics, facilitating future farmer-to-farmer learning. The enthusiasm with which the project has been received, a suite of legacy resources and a commitment to continue extending results to industry, means Australia's dairy farms will reap the benefits of genetic improvement well into the future.

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O141

Revision of the aggregate fertility index for Italian Holstein-Friesian dairy cattle

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The aggregate fertility index was added to the national Holstein breeding objective in 2009, and is a linear combination of five selection criteria: angularity, milk yield (MY), interval calving-first service (DTFS), 56 days non-return rate (NR56), and calving interval. The objective of the present study was to include new traits, measured in both productive cows and heifers, to such aggregate index, by exploiting information from linear scoring and fertility-related events. Six subsets of ~15,000 animals calving since 1994 were extracted from the national database. Traits identified were: age at first insemination in heifers (AFI), interval first-last insemination (IFL), NR56, and conception rate (CR) in heifers and cows, and DTFS, MY, and body condition score (BCS) in cows. A multiple-trait animal model was employed to estimate (co)variances of these phenotypes, separately for cows and heifers. Fixed effects per trait were: herd-year of birth (AFI), month of birth (AFI), herd-year-season of birth (IFL, NR56, CR in heifers), month-year of insemination (IFL, NR56, CR in heifers), herd-year-season of calving (MY, DTFS, IFL, NR56, CR in cows), month of calving (DTFS, IFL, CR in cows) or insemination (NR56 in cows), age-year of calving (DTFS, IFL, NR56, CR in cows), herd-year-season of classification (BCS), age-stage of lactation at linear scoring (BCS), and year of calving (BCS). Animal additive genetic and residual were treated as random terms. The pedigree was traced back 5 generations. Estimates of G and R matrixes of all the six subsets were averaged to apply selection index methodology for deriving appropriate index weights, by fixing CR in cows and heifers as the only breeding goals. Heritability of cow fertility traits ranged from 0.013 (NR56) to 0.076 (DTFS), whereby estimates of heifer traits were between 0.012 (CR) and 0.071 (AFI). Relative emphasis placed in the selection criteria of the aggregate index for cows were: 0.55 (IFL), 0.16 (DTFS), 0.13 (NR56), 0.09 (MY), and 0.07 (BCS). Index weights for heifer traits were 0.47 (IFL), 0.43 (NR56), and 0.10 (AFI). Both indexes were scaled to have mean 100 and standard

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deviation 5. The aggregate fertility index published in the official genetic evaluation release is a combined index which includes both indexes by placing 90% emphasis on cow aggregate index and 10% emphasis on heifer aggregate index.

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O142

Variation of milk protein profile of cows from multi-breed herds

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The aim of present work was to study the variation of milk protein fractions (α_{s1}-CN, α_{s2}-CN, β-CN, κ-CN, β-LG, α-LA), expressed as g/L of milk, and as percentage of the total nitrogen (%N) of milk, in relation to the breed, parity, and days in milk (DIM) of the cow, and of *CSN2*, *CSN3*, *BLG* genotypes.
A total of 1500 dairy cows belonging to 6 breeds (3 specialised dairy breeds: Holstein-Friesian, Brown Swiss, Jersey; and 3 dual-purpose breeds: Simmental, Alpine Grey, Rendena) were sampled from 41 multi-breed herds (from 2 to 5 breeds per herd) located in Trentino-Alto Adige region (Northeast Italy). High-Performance Liquid Chromatography (HPLC) permitted the quantification of all the major protein fractions and the identification of β-CN, κ-CN and β-LG genetic variants. Protein fractions were analysed using a linear mixed model, which considered breed, parity, DIM, and *CSN2*, *CSN3*, *BLG* genotypes as fixed effects, herd as random effect and herd productivity as covariate.
Results showed that parity and DIM influenced all protein fractions, both expressed as g/L and %N. Also, the genotypes of *CSN2*, *CSN3* and *BLG* affected almost all the traits considered. Even after correcting for the effects of the protein genotypes, breed of cows showed large differences: Holstein-Friesian cows had the lowest milk protein content, but with the highest proportion of true protein, mainly because of a high proportion of β-CN and whey protein; Jersey cows had the highest milk protein, casein and particularly α_{s1}-CN, contents, but the lowest proportion of true protein and content of whey proteins; Brown Swiss and Simmental cows showed intermediate results; Alpine Grey cows were characterised by the highest proportion of true protein and whey protein (like Holsteins); lastly, Rendena cows showed the highest proportion of casein (like Jerseys), content of β-CN, as Holstein-Friesian, and proportion of α_{s2}-CN and κ-CN.
This study allowed to disentangle the effect of 6 dairy and dual-purpose breeds due to the major genes of *CSN2*, *CSN3* and *BLG* from those due to the other genes, in the same herds and taking into account the effect of herd productivity.

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O143

Genomic predictions of milk coagulation properties

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Among the characteristics that make milk optimal for cheese production, together with fat and protein, milk coagulation properties (MCP) must be included: rennet coagulation time (RCT), curd-firming time (k20) and curd firmness (a30). Moreover, an estimator of cheese yield, such as the individual laboratory cheese yield (ILCY), should be added. All these milk features are becoming influential in the breeding programmes oriented to cheese production, which is the main purpose for several livestock species or breed. The dataset was composed by 965 ewes; 769 of these were genotyped with Illumina Infinium Ovine SNP50 v1 BeadChip. A pedigree with more than six hundred animals was also available. All ewes had records for RCT and ILCY, while only 903 and 857 records were available for a30 and k20, respectively. In order to have a normal distribution of records, k20 was converted to its logarithm (logk20). Aim of this study was to estimate breeding values accuracy using two methods for variance component estimation: restricted maximum likelihood (REML) and Gibbs sampling (GIBBS). Using blupf90 family software, genomic breeding values (GEBV) were computed using a single-step approach. Accuracies (ACC) of GEBV were computed as the root square of reliability. The overall accuracy values for phenotyped ewes were 0.38 ± 0.16 and 0.35 ± 0.14 using REML and GIBBS, respectively. The highest ACC value was recorded for ILCY, with average values of 0.54 ± 0.04 (REML) and 0.49 ± 0.04 (GIBBS), followed by RCT that showed values of 0.44 ± 0.04 (REML) and 0.37 ± 0.04 (GIBBS), respectively. Strongly lower accuracies were observed for females without phenotypes: 0.11 ± 0.07 for a30 and 0.06 ± 0.04 for logk20. The most represented ram in the pedigree, with eleven daughters, showed an ACC average value of 0.62 ± 0.10. The quite promising results obtained, especially for ILCY and RCT, suggest the possibility to include these traits as breeding goal in dairy livestock.

O144

Genomic breeding values accuracies using phenotypes or genotypes

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The inclusion of genotype information in several cattle breeding programmes all around the world lead to the genomic selection (GS) era. One of the main advantages of GS over the traditional selection is the possibility of an early estimation of breeding values of candidates based on their genotypes. The optimisation of genotyping and phenotyping strategy represents a key point for GS programmes. Aim of this study was to evaluate the effect of using different phenotyping and genotyping strategies on sire breeding values (BV) accuracies. Using QMSim a dairy bovine population was simulated. Five replicates of ten recent populations were simulated starting from 200 males and 50,000 females. Best animals were selected and mated using a positive assortative mating design; sire and dam replacement were fixed at 40 and 30%, respectively. Ten identical chromosomes each with 1000 markers were generated for the last three generations. About 26,000 females per generation were available. The phenotypes of the last generation were masked in order to represent the candidates of a breeding programmes. Using blupf90 family programmes, breeding values were estimated for 160 sires of female belonging to the tenth generation. Some of those sires had daughters also in the ninth generation. BV accuracies were computed as root square of reliabilities. Masking phenotypic and genetic information of candidate females, the average BV accuracy of sires was 0.54 ± 0.48. The high standard deviation can be ascribed to the different number of daughters in the previous generations. Increases were observed when phenotypes or genotypes were added in the analyses: 0.92 ± 0.02 and 0.75 ± 0.15 for phenotypes and genotypes, respectively. The best scenario was found using both phenotypes and genotypes, with an average accuracy of 0.93 ± 0.01. Using phenotypes in lieu of genotypes lead to higher BV accuracies. However, the phenotypes registration implies longer generation intervals and increase of costs. The quite good accuracies highlighted using only genotypes and the low genotyping price at this point can suggest the use of this strategy instead of waiting for phenotypes collection also for the well-known advantage of having BV already available at candidate birth. As expected, the highest accuracies were found when all possible sources of information (phenotypes + genotypes) were included in the model.

O145

Signatures of selection in the genome of Italian Holstein cattle for cheese production

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Dairy cattle breeds have been exposed to intense artificial selection for milk production traits over the last fifty years. In Italy, where over 75% of milk is processed into cheese, selection has also focussed on cheese-making traits. The most widely spread PDO Italian cheeses in the world are the Parmigiano Reggiano and the Grana Padano, which cover 16% and 24% of the total Italian milk production. Intensive selection programmes that aim at increasing production yield can cause loss of genetic variability and increased genomic homozygosity. To this end, recent advances in genome mapping have the potential both to improve our understanding of selection and to be used in the breeding programme. The present study aims to detect potential selection signatures in the genome of Italian Holstein dairy cows bred for PDO cheese productions. High-density genotype information from SNP-chip was available for 400 cattle, reared in certified farms for Parmigiano Reggiano (PR =200) and Grana Padano (GP =200). The quality control (QC) of the data was performed by excluding both animals and SNPs with a call rate <90% and by removing both unassigned and on-sex-chromosome SNP. We then performed a genomic scan for runs of homozygosity (ROH) which were detected in PLINK 1.9 by using a sliding window approach. ROH were defined based on at least 1 Mb-long homozygous segments, and by allowing for a maximum of one missing and one heterozygous SNP.

All animals passed the QC and 311,501 SNP were used to estimate ROH. In total, 142,211 ROH were detected (355.5 segments per animal), with an average length of 1.59 Mb. The mean number of homozygous segments in the PR and GP cattle was similar (354 and 356, respectively). For 115 PR and 122 GP cows, at least one ROH longer than 16 Mb was found, probably indicating recent inbreeding. Six genomic regions with ROH shared among more than 70% of the 400 cattle were located on the following chromosomes: BTA 11, 14, 16, 18, 21 and 29. As a small amount of regions showed a different rate of homozygosity when comparing PR and GP cattle, further studies will investigate any potentially-different signs of selection among PDO dairy chains. Further research is ongoing to demonstrate the role of those homozygous segments on cheese-making traits.

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ENVIRONMENTAL SUSTAINABILITY – ENVIRONMENTAL IMPACT OF LIVESTOCK II

O146

Added value of dairy farming in mountain areas: an ecosystem services approach

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This study aims to analyse synergies and trade-offs between efficiency, environmental footprint and non-provisioning ecosystem services in dairy cattle farms in Eastern Alps (Veneto, South Tyrol, Friuli and Carinthia). Seventy-five dairy cattle farms conferring milk to 10 cooperative dairies were sampled. The following methodological approaches were applied: environmental footprint (Life Cycle Assessment, LCA); animal welfare; analysis of biodiversity in the grasslands managed by each farm. A LCA approach was used to calculate: Global Warming Potential (GWP), Eutrophication (EP), Cumulative Energy Demand (CED) and Land Occupation (LO). Production efficiency was calculated as gross energy conversion ratio, considering also the diverting of human edible resources. Animal-based indicators of welfare were assessed through direct observation of cattle or through data retrieval from farm records according to the EFSA protocol on animal welfare assessment on small-scale dairy farms. Correlations among the different indicators were tested to analyse synergies and trade-offs between efficiency, environmental footprint, animal welfare and grassland biodiversity. The average herd size was 42 ± 27 LU, the milk yield was 6299 ± 1923 FPCM/cow per year and the agricultural area was 27 ± 20 ha, mainly grassland, with a large variability of farm self-sufficiency (from 14 to 100% of diet dry matter). The impact categories calculated with LCA approach were in line with literature: GWP =1.32 ± 0.42 kg CO_2-eq; EP =7.96 ± 3.36 g PO_4-eq; CED =3.96 ± 2.14 MJ; LO =2.18 ± 1.33 m^2/y, 1 kg of FPCM as functional unit. The higher production, the lower impact categories, but the use of external inputs, especially human edible concentrates, affected this result. Farms with high self-sufficiency can maintain grasslands with a good balance between production and species richness. Results on animal welfare assessment were similar to the ones presented in previous studies: for example, the average percentage of dairy cows with a level of somatic cells higher than 400,000 was 9.4%±1.06 and the average percentage of lameness (mild and severe) was 8.6%±1.06. These results evidenced that the alpine dairy farming systems are able to produce high-quality products by using local resources. A multi-indicators approach is recommended to analyse the sustainability and the efficiency of these systems, considering the trade-offs between production and non-provisioning ecosystem services.

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O147

Carbon footprint of dairy sheep farms located in different forage systems of Sardinia

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The aim of this work was to estimate the carbon footprint (CF) of a sample of 15 Sardinia dairy sheep farms selected from 3 forage systems (FS) identified as: lowland (L), hill (H) and mountain (M). Farms were classified on the bases of their altimetry (<200, 200–500, >500 metres a.s.l.), flock size (low: <200 heads, medium: 200–500 heads, high: >500 heads) and type of soil (alluvial, calcareous, effusive and granitic land substrates).

Data were collected by interviewing the farmers for a complete life cycle inventory (LCI) of farm production processes from October 1st 2016 to September 30st 2017. The LCI included information on flock, animal diets, feed purchases, crops, farm stocks, and an energy use audit. Data were analysed with a modified Tier 2 of the IPCC (2006), by using coefficients of IPCC for estimation Q16 of animal and manure CH_4 and N_2O emissions and literature coefficients for purchased feeds and for energy. Reported values of CF were allocated 100% to milk yield. Average milk production level of the flock (PL) was equal to 217, 198 and 173 kg of fat and protein corrected milk (FPCM) head^{-1} year^{-1} for H, L and M forage systems, respectively. When expressed per animal, emissions resulted higher in H than in the M and L forage systems (627, 620 and 612 kg of CO_2equivalent (CO_2eq. head^{-1} year^{-1}) respectively. In contrast, emission intensity was on average higher in the M than in the H and L forage systems (3.72, 3.31, 3.15 kg CO_2eq kg FPCM^{-1}, respectively). Production level was the best predictor of CF in M ($CF = -0.016 \times PL + 6.4972$; R^2=0.73), H ($CF = -0.0139 \times PL + 6.3238$; R^2=0.79) and L ($CF = -0.0085 \times PL + 4.8104$; R^2=0.73) forage systems.

The contribution to the total emission was about 50% from enteric methane in each of the forage systems. In terms of percentage, L forage system manure management and energy use were the most important variables contributing to CF, whereas in H and M the most important variable was the amount of purchased feeds. In conclusion, in Sardinia sheep farms emission mitigation should take into account increases of: (a) flock production level, (b) the farm efficiency and (c) the amount of on-farm produced feeds. However, this preliminary study needs to be improved considering the carbon sequestration from soil, natural pasture and forests and ecosystem service provided by the forage systems.

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O148

Environmental sustainability of dairy farms producing milk for Grana Padano and Parmigiano Reggiano cheese production

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Dairy cattle farms are known to be part of very complex production systems that require a huge amount of inputs for animals' breeding (e.g. cultivated and purchased feed) and that release emissions in the environment (e.g. methane, ammonia, N and P).
Following the worldwide spreading interest in environmentally Q17 sustainable productions, in this study, 81 farms located in the province of Mantova (Lombardy Region) and producing milk for Grana Padano (GP) and Parmigiano Reggiano (PR) cheese production were analysed.
The aim was to compare data collected on farms between the year 2016 and 2017 and to identify the improvements or worsening of the main parameters affecting dairy farms and their environmental sustainability. In particular, by analysing separately farms producing milk to GP (47 farms) and to PR (34 farms), the following main parameters were calculated: Dairy Efficiency (kg FPCM/kg DM feed), dry matter intake (kg DM/d per cow), Income Over Feed Cost (€/d per cow), herd composition and number of dairy cattle (n.), livestock units (LU/ha), land area (ha), farm self-sufficiency (%) and nitrogen (N) and phosphorous (P) farm balances (calculated as difference between input and output at farm level). All calculated data derived from primary data collected on the farm through questionnaires.
In the 2 years evaluated, Dairy Efficiency improved by +2.3% in GP farms (1.29 in 2017), while reduced by −2.6% in PR farms (1.09 in 2017), and dry matter intake in dairy cow rations, herd dimension and land area lightly raised on both GP and PR farms. IOFC increased just for PR farms (12.2 €/d in 2017, +7.2% respect 2016), due to a sensible increased milk price in 2017 (+8.7%). This seems justified by a worse attention to management aspects in PR farms. On the opposite, the self-sufficiency decreased in GP farms (−3.8%), while increased in PR farms (+1.2%), particularly due to better cultivation performances and increase in hay price. The farm N and P balances decreased consistently in GP farms (−33% and −1% for N and P) and increased in PR farms (+14% and +16% for N and P), due to worse attention to management aspects.
Most other parameters showed no sensible variations between the 2 years, highlighting the difficulties to get improvements on the environmental and economic points of view, mostly for different farm management.

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O149

Parameters affecting the carbon footprint of milk at the farm gate

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The latest assessment report (2014) of the Intergovernmental Panel on Climate Change (IPCC) identified anthropogenic greenhouse gas (GHG) emissions at the highest levels ever achieved and estimates the emissions from the livestock sector at 7.1 gigatons of CO_2 eq per year, 14.5 % of the total global anthropogenic GHG emissions. To address the issue at the Italian milk sector, a survey on 61 dairy farms located in the Po Valley was carried out. The contribution of raw milk to GHG emission has been estimated with the Life Cycle Assessment (LCA) a methodology that underpins sustainable development in food production. The aim of the research was to perform the carbon footprint (CF) of milk production at the farm gate (cradle-to-farm-gate) without considering the transport, processing and distribution of milk. Primary data were collected for one year and the CF calculation was performed according to the IPCC guidelines. The functional unit used to report GHG emissions was 1 kg of carbon dioxide equivalents (CO_2-eq) per kg of FPCM at the farm gate. Meat was considered as a co-product. The environmental impacts were grouped into five main categories: purchased feeds, produced feeds, farm consumption, enteric fermentation, livestock manure. SimaPro© v8.0.5 and SPSS were used as modelling and statistical software, respectively. Dairy farms were classified according to the herd size and type and source of feeds being used in animal feeding. Expanding and restricting herds were not considered as this could bias the total CO_2 eq allocation either to meat or milk. Thus, 52 out the original 61 farms were considered in the LCA analysis. Results showed that of the 1.187 kg CO_2 eq per kg of Fat-Protein Corrected Milk (FPCM), 43.3 % were due to enteric fermentation, 29.1% to feed purchased, 14.6% to manure (9.5, 3.0, 2.1 % for CH_4, N_2O direct emission and N_2O indirect emission, respectively), 8.2% to feed production (2.7, 1.4, 4 % for CO_2 from fertilisers, CO_2 from diesel, N_2O from fertilisers, respectively), 4.5% other purchase (3.1, 0.08, 1.3 % for CO_2 from electricity, CO_2 from other energies and other CO_2, respectively).

The CO_2 eq per kg of FPCM was similar to the emissions estimates in European and North American farms (about 1.3 kg of CO_2 eq/kg of FPCM).

O150

Greenhouse gas emissions associated to cattle milk production in northern Italy

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Greenhouse gas (GHG) emissions associated with milk production are mainly generated by the rumen fermentations, the cultivation, processing and transport of feed, and the storage and use of manure. In the last years in northern Italy there has been a gradual change in the forage systems adopted in the dairy farms: for several reasons maize silage, that dominated for decades animal feed crops, has been partly or even completely replaced by other forages. To assess the effect of alternative forage systems on GHG emissions per kg of milk, 46 dairy farms in the Po Plain were selected on the basis of the representativeness of their forage systems. The six forage systems identified were: Conventional (based on maize silage); Winter Cereals (winter cereal silages), High Quality Forages (high moisture ear maize, Italian ryegrass and lucerne silages); Mixed (complex crop rotations), Dry Parmigiano Reggiano (hay), Fresh Parmigiano Reggiano (hay and fresh grass). Through direct interviews of the farmers, data about herd, rations, crops for cattle feeding, purchases, fuel and energy consumption, housing, storage and distribution of manure were collected. The GHG emissions per kg of fat and protein corrected milk (FPCM) were estimated through a Life Cycle Assessment approach. The average GHG emission was 1.36 (±0.31) kg CO_2 eq/kg FPCM. Contribution of CH_4 from enteric fermentation was ~45% of total GHG; N_2O from manure management was 20%; the remaining part was CO_2. In particular, CO_2 from land transformation varied between 8.15 and 22.2% mainly due to different use of purchased soybean meal in the diets. The lowest GHG emission per kg milk was obtained with the High-Quality Forage system: 1.14 (±0.12) kg CO_2 eq/kg FPCM. The system was associated to the highest dairy efficiency (1.51 kg milk/kg DM intake). The highest average GHG emission was estimated for the Fresh Parmigiano Reggiano system: 1.51 (±0.27) kg CO_2 eq/kg FPCM. With this system the highest CH_4 emission from rumen fermentation was obtained as a consequence of the high forage percentage in the rations. The High-Quality Forage system seems to allow a mitigation of GHG emission per kg FPCM in comparison both with the Conventional system based on maize silage (−14.3%) as well as with the Fresh Parmigiano Reggiano system (−25%) and with the average emission of all farms (−16%).

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O151

Effect of the main dietary roughage source on the N-nutrient use efficiency (N-NUE) in dairy systems

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Although dairy cows provide high nutritional quality products, they can contribute to GHG and N-compounds emissions. The raising of N-concentrate feed and N-fertilizer prices and the more stringent regulation aiming at reducing the nutrient loss led to increase N-nutrient efficiency in the dairy systems. Several indicators to assess the N-efficiency are proposed from boundary farm-gate model such as the N-NUE (N output over the N input), and the N surplus (N input – N output). The study took place in 2018 considering 14 specialised dairy farms located in a restricted area of the Veneto region and according to the main roughage dietary sources they were grouped in three experimental theses: high maize silage (HMS, 6 farms), high permanent meadow hay (HMH, 4 farms) and mixed crop-silage/hay (MSH, 4 farms). Since no crops and/or manure were sold, only milk and meat (calves and culling cows) were recorded ad N output.

The N-concentrate purchased feed and N-fertilisers were also established as N input and N atmospheric deposition was assessed as 20 kg·ha^{-1}·year^{-1}. The apparent N-NUE was equal to 0.34 ± 0.06, 0.30 ± 0.04 and 0.32 ± 0.03 for HMS, HMH and MSH, respectively. As regard to the surplus per surface unit (kg of N□ha^{-1}□year^{-1})/milk unit (kg of N□Mg^{-1} fat correct milk), it was equal to 286 ± 87/14.2 ± 2.7, 102 ± 54/15.4 ± 1.7 and 160 ± 41/14.1 ± 1.2 for HMS, HMH and MSH, respectively. The higher values of farm-gate N-NUE recorded in HMS- and MSH-farms seemed most correlated to a better dietary efficiency of ration based on maize silage (HMS) and/or other cereals silage (MSH) than ones based on permanent meadow hay (HMH). Moreover, HMS and MSH farms were characterised by the presence of high productive cows, thus lower amounts of N were necessary for maintenance and growing up the young stock replacements, even if this N supply also depends on calving rate. Additionally, the cultivation of a winter catch crop after maize lead to a reduction of the input of N-compound feed. Summarising, the sustainability seemed to be the highest in HMH-farm if related to the surplus per surface unit as a regional approach. However, the two systems based on cereal crops could be considered the more sustainable accounting the surplus per unit of production (Mg of FCM) even if it was detected a high variability within the experimental group.

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ANIMAL PRODUCTS – MEAT QUALITY

O152

Effect of rearing system and packaging method on lipid oxidation and shelf life of beef from Aubrac steers during storage

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The effect of different packaging method on lipid oxidation and spoilage were evaluated on beef samples from Aubrac steers maintained in different rearing systems: grazing vs. feedlot. At this aim a sample of *Longissimus dorsi* muscle of each animal was packed by adopting 3 types of packaging: SKIN (high vacuum) and 2 different modified atmospheres (MAP): MAP1 = 50% O_2 20% CO_2 30% N_2 and MAP2 = 65% O_2 24% CO_2 11% N_2. Beef samples were stored at refrigerated (4 °C) storage condition. During the storage period, part of beef samples was analysed after 7 days (T1) of storage and the remaining part at the end of the storage period (T2, 14 days). Lipid oxidation was determined through quantification of TBARS, cholesterol oxidation products (COPs) and the composition of volatile organic compounds by gas chromatography equipped with mass spectrometer (GCMS) coupled with solid phase micro-extraction (SPME). The microbial evolution was evaluated by total mesophilic bacterial count (TMBC), total Psychrophilic bacterial count (TPBC), quantitative determination of Enterobacteriaceae (EB), Pseudomonadaceae (PM), *Brochotrix thermosphacta* (BT) and lactic bacteria (LAB). The effect of rearing system was significant only for TBARS content: beef samples from confined steers (feedlot) contained more TBARS (0.80 mg/kg of beef) than beef samples from grazing steers (0.45 mg/kg of beef). Beef samples packed using MAP contained more volatile organic compounds, mainly composed by aldehydes and ketones. Part of aldehydes and ketones were probably present as secondary products of the oxidation of fatty acids. This data confirmed the significant higher content of TBARS and COPs in beef samples packed with MAP1 and MAP2 methods. The effect of packaging was also statistically significant for the microbial growth on beef samples. The values of TMBC and TPBC were significantly lower for beef samples packed with SKIN and they remained stable at 7 log ufc/g, whereas both MAP1 and MAP2 exceeded this limit. The values of BT and PM were significantly lower in the SKIN samples if compared to MAP1 and MAP2 samples. LAB content did not throughout the storage period, irrespective to the types of packaging (3.5 log ufc/g).

It is interesting to highlight that the skin packaging protected the samples from oxidation and microbiological spoilage regardless of the breeding system for the entire storage period.

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O153

Cattle rearing at pasture or in feedlot farming: quality of meat

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One of the top future challenges is the adoption of different management systems for beef cattle in view of achieving greening herds. The present work aims to investigate the potential of two cattle breeds, Aubrac (AU, 20 steers) and Maremmana (MM, 20 steers) and one crossbred from Piemontese*Maremmana (MM*PD, 16 steers) reared under pasture (28 steers) or under feedlot (28 steers). All animals were slaughtered to commercial maturity (581 kg weight and 21 months on average). Carcases were hanging at 14 or 21 days. After dissection, sample cuts were collected and frozen. The analyses were: pH_1 at thawing and pH_{48} after 48 h; WHC (%); drip and cooking loss (%); share force on cooking loss (kg/cm); MFI and colour (a*, b*, L, C, H). Analysis of variance was performed on the results of the analyses (Proc GLM, SAS 9.4) by including the following fixed factors: rearing system, breed, hanging and their interactions. Rearing system showed no influence on either analysed parameter. The breed had significant effects on weight and age at slaughtering: AU reached the highest weight at younger age (626.0 kg at 20.8 months), MM showed the lowest weight at older age (544.7 kg at 22.6 months). pH_1, pH_{48} and WHC were lower in AU and higher in MM (5.38, 5.44 and 8.56% vs. 5.57, 5.66 and 11.47%, respectively). The lowest share force was obtained by MM*PD compared to AU (2.00 vs. 2.28 kg/cm). MFI was lowest in AU compared to MM (61.97 vs. 120.29). As for meat colour, AU was the brightest with a more defined red colour, by achieving the highest values for L and H indices (L 39.36 vs. 35.52 on MM and H 47.71 vs. 40.08 on MM*PD, respectively), AU showed also the lowest a*index (13.50 vs. 16.32 in MM*PD). Share force differed between breeds after 21 days at hanging. Moreover, MM*PD showed a decline in share force during maturation. Also cooking losses differed between breeds at 21 days of hanging, while AU showed significantly different cooking losses between the two hanging times (22.60% at 14th day vs. 25.09% at 21st day). Our results did show significative differences in rearing systems, suggesting that it is possible to produce beef from grazing steers with optimal parameters for tenderness and colour, even when grazing upon rough agricultural lands like the Maremma. The present study showed that AU breed achieved the best results for both meat quality and growing performances. However, further studies are necessary to verify the adaptation of this breed to the Italian grazing lands.

O154

Effect of different farming systems on meat quality of Massese lambs

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Massese lambs are commonly fed exclusively on milk and slaughtered at around one month of age, in order to let milk available for cheese production. Nevertheless, if reared up to two months of age with supplementary fed, Massese lambs could be able to produce heavier carcases with greater muscle development representing an innovative product for this breed. Aim of this study was to investigate the meat quality of Massese lamb reared up to 65–70 days of age using different farming systems: stall (S) fed concentrate/hay, semi-free range (F) fed concentrate/hay and pasture, and free-range (P) fed only natural pasture. *Longissimus dorsi*, *Triceps brachii* and *Semimembranosus* muscles were analysed, and principal meat quality proprieties were studied. Data were subjected to ANOVA using the GLM procedure of SAS with feeding system, muscle type and sex as discrete effects and age as a continuous variable.

Most of the parameters are affected by rearing system: pH of S lambs is lower than P and F lambs (5.60 vs. 5.80 and 5.75, respectively; p=.001) confirming the fact that high energy diets protect animals against potentially glycogen depleting. Total lipid content is higher in S and F lambs. Texture profile analysis (TPA) shows that P lambs is less tender than other groups (hardness 22.6 vs. 13.7 and 8.5 for P, F and S, respectively; p=.001). Same result was obtained for the other texture parameters (cohesiveness, springiness, gumminess) where P lambs showed lower values (p=.001). No differences between sex are shown, whereas muscle type affected the meat quality characteristics. *Longissimus dorsi* shows less hardness and less colour intensity, while *Triceps brachii* shows smaller acidification and greater lipid content.

The results suggest that, at least in this range of age, the feeding system significantly affect meat characteristics and concentrate supplementation improves meat quality characteristics.

Acknowledgements

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O155

Effect of Polyphenols supplementation on growth performance and meat shelf life of broiler chicken

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The supplementation of broiler chicken diet with polyphenols may be a useful approach to affect both the growth performance of the animals and the shelf life of their meat. The improvement in growth performance represents an important item from economical point of view; the control against meat lipid oxidation is a pivotal goal in the product treating, as the peroxidation of both fatty acids and cholesterol can produce several detrimental substances with respect to the safety and the organoleptic properties of foods.

The experiment was aimed at studying the effect of polyphenol supplementation on growth performance of Ross-308 broilers and on the protection against lipid oxidation of meat. One-hundred and five animals were randomly allotted at 21 boxes (each of five animals), divided in three groups according to with diet. The control diet (*C*, no-supplementation) was compared with two diets supplemented with 2 g/kg of dray matter of following products; *OxilemTM* (*OX*), a low molecular weight polyphenol from cellulose and *OxiGemTM* (*OG*), obtained by *OxilemTM* plus *SanitresTM*, polyphenols from Pine tree. *OxilemTM* and *OxiGemTM* were obtained from *Green Innovation GmbH, Innsbruck AUSTRIA*. The experiment lasted 6 weeks.

OG was effective in improving the animal feed intake (0.215 vs. 0.202 g/day of *C* of last week), the daily live weight gain (90.47 vs. 80.98 g/days of *C* of last week); with respect to the meat proximate composition, *OG* affected the total protein content (23.69 vs. 22.28 g/100 g of meat from *C*). We relieved no effect on meat fatty acid composition. In order to monitor the oxidation status of fatty acids we performed TBARS test and volatile organic compounds (VOCs) by means of SPME; the content of cholesterol oxidation products (COPs) were used to estimate the oxidation status of cholesterol. *OG* and *OX* resulted effective in providing protection against meat lipid oxidation.

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O156

Effects of two different olive mill wastewaters on growth performance, quality of meat and *Campylobacter* loads in broiler chickens

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Food wastes are sources of different compounds that can be used as natural additives in the food and feed industry. The olive oil industry produces two main wastes: aqueous waste (olive mill wastewater, OMW) and solid waste (pomace or olive cake). These by-products are rich in phenols, which are antioxidant and antimicrobial compounds able to inhibit or delay the growth rate of several bacteria *in vitro*. In this experiment, the dietary effect of two different OMWs on growth performance, quality of meat and *Campylobacter* spp. load in broiler chickens was investigated. A commercial basal feed was supplemented with either OMW-1 or OMW-2 (tyrosol to verbascoside ratio 5:1 vs. 1:1 for OMW-1 and OMW-2, respectively), to reach a similar total polyphenol concentration in the feed (125 µg/g), and administered to chickens from the 22nd to the 42nd day of age, when the animals were slaughtered. A negative (CTR) and a positive control group (α tocopherol added at 245 mg/kg feed, VIT-E) were also included in the experimental design. Growth performance of broilers was monitored at day 22 (T0), 35 (T2) and 42 (T3). *Campylobacter* spp. shedding was evaluated at 22 and 42 days of age and neat quality assessed after slaughter. Both body weight (BW) gain and feed conversion ratio (FCR) of OMW-2 birds were similar to those recorded for the CTR and the VIT-E groups (average values: 146.1 g/d and 2.45 for BW gain and FCR, respectively). The OMW-1 group showed a lower ($p<.01$) BW gain (131.5 g/d) during the whole period of the trial in comparison to the other groups (143.2, 147.9 and 147.2 for OMW-2, CTR and VIT-E, respectively). However, the OMW-1 FCR tended to be lower (1.79) compared to those observed in the other groups (2.54, 2.41 and 2.40 for CTR, OMW-2 and VIT-E, respectively). Meat quality characteristics were not different among the four experimental groups. The *Campylobacter* isolates from faecal samples were identified by multiplex PCR as *C. coli* in all groups. At the end of the trial, OMW-2 group showed a lower ($p<.05$) *Campylobacter* load compared to the CTR and the VIT-E groups. These results highlight the potential use of OMW by-products against *Campylobacter* spp. in poultry. Further studies are needed to assess the palatability of OMW diets in broilers and to investigate possible synergistic effects among phenolic compounds.

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O157

In ovo delivery of GOS in fast-growing broiler chickens exposed to heat stress: implications for meat quality traits

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In ovo technology is proposed as a method for early stimulation of intestinal microbiota, which protects animals from heat-induced gut dysbiosis. A study was carried out to evaluate meat quality traits in fast-growing chickens stimulated *in ovo* with trans-galactooligosaccharides (GOS) and exposed to heat stress. On the day 12 of egg incubation, 3000 fertilised eggs (Ross 308) were divided into: prebiotic group (GOS) injected with a single dose of 3.5 mg GOS/egg, saline group (S) injected with physiological saline and control group (C) un-injected. After hatching, 900 male chicks were reared in floor pens: three groups (GOS, S and C, 6 pens/group, 25 birds/pen) were reared in thermoneutral conditions (TN) and three groups (GOS, S and C, 6 pens/group, 25 birds/pen) were reared under heat stress conditions (HS) induced on day 32 (30 °C) for 10 days. At 42 days of age, 15 randomly chosen birds/treatment/temperature were slaughtered and the pectoral muscle (PM) was removed for analyses. Data were analysed by GLM procedure in a 3 × 2 factorial design. Treatment had no effect on PM weight; however, PM from GOS group was slightly heavier (+9.7%) compared to C. pH and WHC were similar among groups. GOS and S birds had lighter (L*, $p<.01$) PM than C group; while, C group had a higher ($p<.05$) yellowness index (b*) compared to S group. Proximate composition, cholesterol and intramuscular collagen contents were not affected by treatment. Total content of saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA) was similar among treatment groups, while total polyunsaturated fatty acids (PUFA) content and n-6 PUFA were slightly lower in GOS group compared to S one. Heat stress had a detrimental effect on PM weight ($p<.01$) and increased meat pH ($p<.01$). PM from HS chickens was darker ($p<.05$) with a higher b* index ($p<.05$) than TN group. Compared to TN conditions, heat stress increased ($p<.01$) lipid content (+0.44%) but decreased ($p<.05$) total collagen amount; differently, it did not affect total content of SFA, MUFA and PUFA, but increased total n-3 PUFA. Significant interactions between factors were found for lipid, ash and fatty acid composition: GOS decreased ($p<.01$) SFA and increased ($p<.05$) MUFA contents in HS birds. In conclusion, *in ovo* injection of GOS prebiotic had no negative effect on physico-chemical and nutritional properties of meat from fast-growing broiler chickens. Heat stress reduced PM weight and slightly affect meat quality.

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – CLIMATE

O158

Effect of different flooring systems on claw condition of finishing beef cattle during warm and cold seasons

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The type of floor adopted on specialised fattening units for beef production is considered a relevant risk factor for animal health. In mainland Europe, the predominant flooring systems used on beef farms are the concrete fully-slatted floor (CS) and the deep litter (DL). Recently, covering concrete slats with rubber mats (RM) is the most common alternative solution. This study aims to investigate the effects of these three-floor types and of the finishing season on claw condition of beef cattle. A total of 2162 left hind feet belonging to 153 batches of cattle were collected after slaughter in commercial abattoirs. Dorsal wall (DW) and total hoof (TH) lengths, heel height (HH) and toe angle (TA) of lateral claws were measured. After trimming, a vet diagnosed specific claw disorders. Infectious (ILS) and non-infectious lesion (NILS) scores were calculated per each foot. Information regarding the type of floor adopted for each batch at the origin farm (45 batches housed on CS; 61 on DL; 18 on RM; 29 missing data) and the season during finishing (95 finished in winter; 58 in summer) were collected. A mixed model was used to test the effects of floor type and season on claw dimensions, lesion scores and the prevalence of specific disorders. Shorter DW, TH and HH with greater TA were recorded in CS bulls. Both bulls on DL and RM had longer DW and TH, but only RM bulls had lower HH and sharper claws. Season affected only claw HH, being greater in bulls finished in summer. Both mean claw lesion scores did not differ for floor type. Bulls finished in winter had worse average ILS than in summer, with a higher % of feet/batch having the worst scores and lower % of healthy feet. Prevalence of feet/batch affected by heel horn erosion was higher on DL and RM. Neither floor type nor season affected the % of feet/batch with interdigital dermatitis. Bulls finished in summer had worse average NILS than in winter, with a lower % of healthy feet/batch and higher % of feet grading the worst scores. The % of feet/batch showing sole haemorrhages and white line abscesses (WLA) was higher in summer. Prevalence of feet/batch grading the worst NILS and that of those with WLA were higher on CS and DL than RM. Sole ulcer prevalence was not affected by floor type or season. Results point out a seasonal variation of claw condition according to disorders' aetiology. Although rubber floors could prevent some non-infectious lesions, they could develop claw overgrowth or infectious disorders.

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O159

Methylomes of indicine and taurine cattle under heat challenge

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Heat stress has a detrimental impact on cattle health, welfare and productivity. Its impact on gene expression, metabolism and immune response has been investigated but little is known on the epigenetic mechanisms mediating the effect of heat at the cellular and organism level. To understand the role of epigenetic variation in cattle adaptation to heat stress, we investigated genome-wide DNA methylation in blood samples collected from 10 bulls, 5 Nellore (heat stress resilient) and 5 Angus (heat stress susceptible) exposed to the sun during the high temperature-high humidity season of the Brazilian Southeast region. Their methylomes were analysed by Reduced Representation Bisulphite Sequencing which provided genome-wide single-base resolution methylation profiles.

To date, an average of 14.8 million reads were produced per sample, with varying mapping efficiency (40.4–47.1%) and a narrow range of methylated Cs in CpG context between 66.0 and 68.2%. Significant methylation changes between stressful and recovery periods were observed in 822 genes. Among these, 352 were private to Angus, 367 were specific to Nellore, and 103 showed significant changes in methylation patterns in both breeds. We found that epigenetic signals related with response to heat challenge and recovery were mostly breed specific. Furthermore, among the 103 common genes, a relevant proportion showed opposite behaviour in the two breeds, having higher methylation in the stress period and lower in the recovery period in one breed and the opposite in the other. These genes were subjected to KEGG (Kyoto Encyclopaedia of Genes and Genomes) and Gene Ontology (GO) enrichment analyses, which confirmed a breed-specific response and showed that only a few cellular functions (a single one in GO enrichment analysis, phosphoric diester hydrolase activity) were shared between breeds. Interestingly, the differential methylation response to heat stress/recovery seemed more organised in Nellore, as compared to Angus, resulting in the identification of a higher number of enriched functions. In this breed, KEGG pathway analyses indicated that significant genes were mainly involved in stress responses and cellular defence. These preliminary results suggest that heat challenge does not promote significant changes in genome-wide
Q18 CpG methylation, but rather induce changes in methylation patterns in specific loci. These loci should be further scrutinised to assess their role in heat tolerance.

O160

Individual thermal response of Avileña Negra-Ibérical calves at feedlot

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The increasing demand for meat in developing countries, together with the rapidly increasing change of the world's climate conditions has made the thermal tolerance one of the most important adaptive aspects for cattle. The identification of thermally-tolerant breeds and also the selection of more resistant animals within the same breed is basic to maintain the productivity of the beef cattle production systems.

In this study, the aim was to investigate the genetic component of thermal response in male calves of Avileña-Negra Ibérica (ANI) as a first step for an eventual genetic evaluation to improve thermal tolerance in this breed. Three random regression models (RRM) with second and third order of Legendre polynomials for additive genetic and permanent environmental effects were fitted to estimate the variability in individual reaction to increasing heat loads of weight in feedlot. A total of 29,591 weight records belonging to 5876 animals collected between 2005 and 2017 were used. Pedigree file included 15,418 animals. The RRM were fitted for weight using average temperature as covariable through use of Blupf90 programmes. Range of average daily Ta at the location of the feedlot ranged from a minimum of −4 to a maximum of 30 °C.

EBV for changes in weight at different temperatures were estimated for all animals with results of Model 3 that had the best fit to the data. The results revealed a mean of variation of the EBV values of 215.63, 194.9 and 188.84 kilograms within cold, comfort and hot range of T class, respectively. The range of average variation between the minimum and maximum temperature class was equal to −129.96 and 85.67 kg for cold range; (112.35–82.57) and (103.79–85.05) for the comfort and hot range of temperature, respectively. Furthermore, among the temperature scale a range of the hereditability equal to 0.52 and 0.64 was recorded.

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O161

Heat stress in Italian Brown Swiss

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The aim of the present study is to estimate the upper critical thermo-hygrometric index (THI) thresholds beyond which production traits start changing in Italian Brown Swiss breed. The data set comprised 957,449 test day records, from 2008 to 2017, from 85,380 Brown Swiss lactating cows and 698 farms located in Italy. Investigated production traits were: milk yield, fat-corrected milk, fat and protein concentration (%), fat and protein yield, somatic cell score (SCS), urea (MUN), saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids. Environmental climate data were collected from 76 weather stations. Data-set was clustered according parity in 4 categories (primiparous, second diparous, third calving and equal or up to fourth calving). A linear model was used to estimate the THI on milk production traits, considering the THI of the same day of test day and of the five days before. After this, using a 2-phase linear regression procedure, the THI thresholds of the same day for milk traits in all parity categories considered were detected. An analysis of data showed that the daily THI at which milk yield start to decline ranged from 75 to 78 ($p<.001$) for all parity classes. Protein concentration started dropping at THI ranging from 68 to 74 ($p<.001$), while fat concentration started to decline at THI ranging from 74 to 79 ($p<.01$). The SCS started to increase at THI ranging from 64 to 71 ($p<.0001$) and MUN increased at THI ranging from 63 to 68 ($p<.001$). The SFA content started to decline at THI ranging from 65 to 72 ($p<.01$), instead MUFA and PUFA started to increase at THI ranging from 63 to 69 ($p<.01$). The negative effect of THI is recorded both on milk yield and on protein and fat yield and concentration. The study recorded also, over the calculated THI thresholds, a reduction of SFA milk content and an increasing of SCS, MUN, PUFA and MUFA. In particular, the negative effect of THI on milk yield and constituents showed that dairy cows at first parity are less sensitive than multiparous, showing often higher THI thresholds. Moreover, fat percentage showed lower sensibility to THI increasing, on the contrary SCS and urea showed to be particularly sensible to THI effects. Data obtained showed that Italian Brown Swiss have a higher thermal tolerance than Holstein, although the climatic conditions in Italy (but not only) represent a main concern also for this breed.

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – COMPANION ANIMALS I

O162

Digestibility of extruded diets for cats: *in vitro* vs. *in vivo*

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In animal, nutrition digestibility represents one the most important parameter to evaluate the nutritive characteristics of novel ingredients/diets, and this parameter is highly different among species and/or feedstuffs. In companion animals only, few digestibility data are available, due to the ethical concern related to society preconception on animal's trial. The *in vitro* digestibility trials can represent valid alternative to the *in vivo* ones because they are faster, cheaper and more observant of animal welfare. The aim of this study was to evaluate a new *in vitro* enzymatic method to predicting apparent digestibility (OMD_iv) of 18 extruded cat diets, comparing the data obtained with those achieved *in vivo* (OMD). The *in vivo* evaluation was carried out in Jaboticabal using six cats kept in metabolic cages, during the experimental period (10 days of adaptation +30 days of sampling). The same diets were incubated *in vitro* in Napoli, incubating 0.75 g of diet with pepsin (1%; pH2) for 2 h at 39 °C; then a pancreatin solution (10%; pH 6.8) was added and incubated for 4h at 39 °C. At the end, sulpho-salicylic acid (20%) was used to precipitate the residual protein. The undigested residue was filtered in glass crucibles, dried at 103 °C and ashed at 550 °C.
The mean chemical composition of the extruded diet was (% a.f.): dry matter 92.88 ± 1.05, ash 7.33 ± 0.79, crude protein 31.75 ± 3.64, ether extract 12.66 ± 2.13 and crude fibre 5.16 ± 2.26.
The determination coefficient ($r^2 = 0.7607$) suggest that the proposed *in vitro* method provided an accurate prediction of *in vivo* digestibility, even if on average OMD_iv values were lower than OMD ones. Correlating chemical composition parameters with OMD and OMD_iv values, the two methods showed similar trend. Only lipids content and structural carbohydrates significantly

affected OM digestibility: crude fibre was the main factor that influenced negatively ($p<.01$) digestibility (−0.697 and −0.598 for OMD and OMD_iv, respectively), while ether extract was positively correlated with digestibility (0.493; $p<.05$ and 0.589; $p<.01$, respectively).

These preliminary results suggest the goodness of the proposed method to predict OM digestibility, even if the digestibility of other nutrients have to be studied and a larger number of different pet-food have to be tested in order to better represent the wide variation of pet-food which exists on the market and, consequently, to achieve a better adjustment of the obtained equation.

O163

DogBiome: the gut microbiome project for dog

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Recently, a description of the microbial population of many niches of the organism, for example, the gastrointestinal tract, is feasible thanks to the use of advanced techniques such as High-throughput DNA sequencing that allows the simultaneous analysis of microbiota. Regarding the companion animals, a growing number of studies investigated faecal microbiome in healthy or affected subjects, although the methodologies used in the different laboratories do not allow a straight comparison among results. Despite this, they still have the interest in studying microbiome in depth, also to see how diet modifications can affect this latter.

In the present study, we report data collected from several in house researches carried out in healthy dogs, with the aim to describe the variability of microbial taxa in the faeces, providing a global picture of the composition of faecal microbiota. Overall, the database contains 334 samples from 132 dogs, which were collected during dietary intervention studies, where the diet composition and nutritional supply were under experimental control. According to the experimental design of the trials, for some dogs serial faecal samples were collected to assess the response of microbiota to the diet. The procedure of samples collection, storage, DNA extraction and sequencing, bioinformatic and statistical analysis followed a defined pipeline. The relative abundances (RA) of *Firmicutes*, *Bacteroidetes* and *Fusobacteria* represented more than 90% of the phyla, with minor percentages of *Proteobacteria* and *Actinobacteria*. At a genus taxonomic level, 72 genera were found with an RA >0.001%, although 20 of these genera accounted for more than 90% of the total RA. The genera with a threshold of RA >5% were *Clostridium*, *Blautia*, *Fusobacterium* and *Bacteroides*. Alpha and beta biodiversity were relevant complementary information to underpin faecal microbiome.

This DOGBIOME database represents a unique archive of dog faecal microbiota, which can form the basis for the identification of reference values, if any, of microbial community. The same data can also be used as a diagnostic tool to screen subject for gastrointestinal conditions. Nevertheless, the implementation of this archive will also help researchers to design diets and to investigate the effect of nutrients on gut functions.

Acknowledgement

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ANIMAL BREEDING AND GENOMICS – GENOMIC EDITING

O164

Unveiling the Biodiversity of the Italian honeybees by next generation sequencing

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Among the domestic animals, the honey bee (Apis mellifera) has an extremely important role both for the production of honey, wax, pollen and royal jelly, but above all for the pollination of arboreal and herbaceous species fundamental for human nutrition. Nevertheless, the genetic management of this species both at a selective and biodiversity conservation level has been proven to be difficult. One of the main reasons lies on the fact that the reproductive moment escapes possible human control as the virgin queen (VQ) is fertilised in flight by a variable number of drones present in the environment. Furthermore, free coupling opens the way to the genetic erosion of local varieties by different genetic subtypes. The Apis mellifera, in fact, presents over 30 subtypes described in detail in the ‘80s by Friedrich Ruttner on a morphometric basis and by Father Adam (Karl Kehrle) of Buckfast Abbey based on the production and behavioural traits. In this scenario, it becomes urgent to provide protection of the local varieties from the phenomena of genetic erosion.

To implement a correct genetic conservation plan it is necessary to describe the biodiversity present today in a robust manner.

The availability of the Honeybee genome allows using next-generation sequencing methods to define subtypes/subpopulations more precisely.
In this context, we analysed patterns of genetic variation of several populations of Honeybees sampled in Italy. In detail, whole genome sequencing has been performed on 125 bee samples by Illumina technology. Sequences have been mapped to the reference genome obtaining a mean coverage of 18.6X (minimum coverage 7.33X and maximum coverage 29.75X). In total, after quality check, 4,095,663 SNPs have been identified. The SNP dataset has been pruned based on linkage disequilibrium (LD) and principal component analysis (PCA) has been performed. The resulting pruned dataset contains 1,032,587 SNP. Results of the PCA analysis of the 125 honeybees allowed to identify and subgroup bees according to their subtype. The SNP collection can be mined to create a SNP panel for subtype testing.

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O165

The illumina® greater good initiative. A further step toward inclusion of camels in the ‘agrigenomic revolution’

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Camels represent a key livestock resource in several low-income countries where they can positively impact food security and resilience. Unlike the other livestock species, the genomic revolution is still in its early infancy for camels. Notwithstanding, a growing world-wide demand for innovation exists, as attested by (i) the recent establishment of the International Camel Consortium for Genetic Improvement and Conservation (ICC-GIC), now counting over than 80 members from various countries; (ii) the ongoing work to establish the organisation of the camel genome at the chromosome level; (iii) the publication of various dromedary transcriptomes. Here, we describe the project, supported by the Illumina Greater Good Initiative Grant, aiming at developing an Illumina Camel Genotyping BeadChip. Implementation of a selection of SNP loci in a SNP genotyping platform may allow rapid and cost-effective genotyping of hundreds of thousands markers in large numbers of animals, thus boosting downstream applications such as genome-wide association studies for production traits and genome-based selective breeding. As the risk of the ‘SNP ascertainment bias’ phenomenon has been shown to be one of the major drawbacks in SNP array design, the project will preliminary perform a whole-genome Q19 camel diversity study across the whole geographic range of camel distribution. Using the core collection of DNA samples available from the ICC-GIC members, a total amount of 20 terabases of Illumina NovaSeq sequencing data will be produced, partly using the 10X Genomics Linked-Read sequencing approach. This strategy will allow to obtain a deep insight on the genomic variation at a large geographic scale (over 400 animals will be whole-genome sequenced), provide better resolution of haplotypes, detect and characterise structural variants, improve the currently available reference genomes, deepen understanding of evolutionary processes (domestication, inter-specific hybridisation, dispersal and selection) that shaped the camel genomes, and possibly decipher the molecular basis of the peculiar physiological adaptation traits of camels. Also, in line with the mission of ICC-GIC, this initiative will allow to strengthen the relationships among the camel scientific community. Finally, the availability of a camel SNP genotyping platform may, in the mid-term, boosts national governments investments in country-based breeding programmes based on systematic phenotype and genealogical recording.

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – REPRODUCTION II

O166

Combined use of dairy sexed semen and double-muscled breed semen for beef crossbred calves production in dairy herds

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Increasing meat production from dairy farms could improve their economic efficiency, the ecological footprint of beef chain and the supply of Italian meat for internal market. These objectives were pursued by MeetBull project, consisting, for dairy cattle, in the insemination of heifers and best cows with dairy sexed semen and other cows with conventional beef semen. These objectives were pursued by MeetBull project, consisting, for dairy cattle, in the insemination of heifers and best cows with dairy sexed semen and other cows with conventional beef semen. A total of 140 dairy farms were involved and about 50,000 inseminations controlled. The slightly lower fertility of sexed semen was compensated by the greater fertility of beef semen, respect to conventional dairy semen. The aim of this study was to investigate the calves' performances, the quality of meat and the market value of MeetBull crossbred calves.

Data were collected from 1530 MeetBull crossbred calves: 245 calves fattened directly by dairy farmers (km0), 847 veal calves (Veal), and 438 calves fattened by commercial fatteners (Beef). Calves were mainly obtained from Holstein, Brown Swiss, Simmental and crossbred dams and from Belgian blue, Limousine, INRA95, Simmental and Piemontese sires. For productive performances, the km0, veal and beef calves showed an average age at slaughter of 637, 207, and 476 days, respectively; a slaughter-weight of 624, 308 and 665 kg; an average daily gain (ADG) of 0.98, 1.49 and 1.41 kg/d; a carcase yield of 54.4, 59.0 and 59.5%; and a carcase SEUROP of R, U and U-. For meat quality, the results for km0, veal and beef were: L* 40.6, 47.4, 40.4; haem iron 15.5, 6.0, and 16.2 mg/kg; cholesterol 41.1, 59.2, 43.7 mg/kg; cooking losses 33.6, 31.7, 35.1%; and shear force 22.2, 22.0, and 28.9 N/cm^2. Dam and sire breed significantly affected many traits. To characterise the nutritional value of meat obtained, also the detailed fatty acids, amino acids and minerals profiles were obtained. As a comparison, also imported purebred (Charolais) and national crossbreds (from suckler cows) stock calves were fattened with MeetBull calves. The calves from double-muscled sires and dairy cows showed characteristics similar to those of the beef stock calves.

In conclusion, the insemination of dairy cows with dairy sexed and double-muscled conventional semen demonstrated to be a viable strategy for improving the efficiency of dairy farms and the supply of Italian bovine meat of good quality and nutritional value.

Acknowledgements

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O167

Developing a new selection index for the Italian Mediterranean buffalo (*Bubalus bubalis*)

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The aim of this work was to develop a new selection index and to estimate the annual genetic response for milk yield (MY), milk components, composite feet and legs (FL) and mammary system (MS) in the Italian Mediterranean Buffalo (IMB) population under different selection indices and breeding objectives. Buffalo milk has excellent nutritional characteristics as a consequence of the high percentage of its constituents. These milk properties are desirable for the dairy industry because mainly used to make Campana buffalo mozzarella PDO cheese. Former breeding objective of the National Association of Buffalo Breeders (ANASB) was based only on the evaluation of kg of mozzarella production (PKM index). Hence, selection by PKM was mainly carried out according to the kg of fat and protein and consequently milk yield. To address the call of mozzarella cheese manufacturers and farmers community, who both asked for a more balanced breeding objective, annual genetic response for the aforementioned traits and correlated response with mozzarella cheese production were estimated using selection index theory. Genetic parameters among traits included in the selection index and traits included in the breeding objectives were estimated using data from 7199 buffalo cows and a pedigree including 19,574 buffalos. A multi-trait animal model was fitted using a Bayesian implementation via Gibbs sampling. Three alternative breeding objectives were formulated according to relative weight given to MY, cheese production, FL and MS: 5:45:30:20 (S1), 30:35:15:20 (S2) and 45:45:5:5 (S3). Traits included in the selection index were MY, fat and protein percentage, FL and MS. Cheese production had a negative genetic correlation with MY (−0.54), a null correlation with MS (−0.01), a positive correlation with FL (0.25), fat % (0.15) and protein % (0.28). S1 breeding objective had the largest and positive genetic response in terms of cheese production, fat and protein %. However, MY was strongly penalised, with a negative genetic response. S3 showed the best results with a positive genetic response for all traits included in the breeding objective ranging from 0.02 to 0.05 genetic standard deviation. Relative emphasis of selection criteria in breeding objective S3, namely MY, fat %, protein %, FL and ML were 21%, 15%, 20%, 24% and 20 %, respectively.

Acknowledgements

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O168

Effect of milk somatic cell level on lifetime milk related performances in Italian water buffaloes

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Dairy buffalo industry plays an important role in the Italian agricultural sector, especially for the high quality and market value of mozzarella cheese. Long living cows are supposed to provide more profit to the farmer, however, lactating animals keep memory of early productive life events and stressors, such as mastitis. The present study aimed to estimate the effect of somatic cell score (SCS) in first lactation on the lifetime cow performances. Data on 270-d milk, fat and protein yields, and on test-day somatic cell count were available for four lactations (first to fourth) of 5757 cows, for a total of 23.028 lactations. At least five test day records within each lactation were ensured to compute lactation mean SCS. Three classes of milk SCS in first lactation were defined using mean (2.67) and standard deviation (1.21): high (SCS > mean + SD), medium (SCS within mean ± SD) and low (SCS < mean – SD). Statistical analyses were performed separately by parity. Milk yield traits were analysed using a mixed linear model that included herd as random effect (minimum three cows/herd), and classes of SCS in first lactation, year of calving, season of calving and age at calving as fixed effects. Pearson correlations between lactation mean SCS treated as different trait in different parities ranged from 0.26 (first and fourth lactation) to 0.60 (second and third lactation). Approximately 47% of cows with high mean SCS in first lactation had high mean SCS also in second lactation. Cows that were in the high SCS class in first lactation produced significantly less milk in second lactation compared with cows that were in low or medium SCS class in first lactation. In subsequent lactations, milk yield did not differ significantly among SCS classes, even if a lower milk production was found in the high SCS class, compared to medium and low SCS classes. Overall, the lifetime milk yield loss was greater than 100 kg. The fat yield of cows that were in the high SCS class in first lactation was similar in the four lactations. Protein yield of second lactation cows was greater for medium SCS class, compared with high and low SCS classes. In conclusion, cows with high SCS in first lactation tended to produce less milk with high SCS for the entire life. Therefore, cow udder health has to be monitored starting from early career in order to reduce health-related costs and improve the farm profit.

O169

Adaptive introgression of river-buffalo derived regions in the genome of Brazilian Carabao swamp buffalo

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The domestication of water buffalo (*Bubalus bubalis*) happened ~5000–6000 years ago and has profoundly impacted human societies as the main livestock resource in tropical and subtropical regions. Currently, two sub-species have been identified, namely river and swamp buffalo, which show different morphology, behavioural traits, production purposes and geographical distributions. Previous genome-wide analyses performed on 31 river and swamp buffalo populations from 16 regions worldwide found evidence of composite river/swamp genomic makeup in Carabao, a swamp type water buffalo breed from Brazil. To better understand the nature and putative adaptive value of the river type-derived introgressed genomic regions, we applied local ancestry investigations on 18 Carabao individuals genotyped for 90k SNPs. For comparison, the same analysis was performed on a recently established crossbred river *x* swamp buffalo population from the Philippines. Local ancestry results showed larger haplotypes of river-derived ancestry in the Philippine population with respect to the Carabao breed, confirming the more recent occurrence of admixture in the Philippine population. Conversely, we identified shorter river type-derived haplotypes in Carabao, suggesting a more ancient event of introgression. We identified several admixed genomic regions, which appeared conserved among Carabao individuals, possibly due to their putative adaptive nature. Among them, a region located on chromosome 4 intercepted a QTL affecting somatic cell score in cattle. Further, a highly conserved region on chromosome 29 intercepted genes, which have been related to fertility in cattle. Our preliminary results suggest that adaptive introgression occurred in the Carabao population and provide better tools for future improvement of the species and a better understanding of the underlying demography.

ANIMAL PHYSIOLOGY, HEALTH AND WELFARE – ANIMAL WELFARE II

O170

Animal welfare and environmental sustainability of dairy buffalo farms in Northern Italy

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In Northern Italy, the buffalo sector began in the eighties, and today it encompasses 22 farms that are mainly devoted to the production of milk for cheese making. Despite the modest dimension, in recent years, the interest in this sector is growing as well as the number of animals registered in the herd-book. In addition, consumers are increasingly concerned about the conditions of farmed animals and about the environmental externalities related to livestock production. Aim of this study was to assess animal welfare and the environmental impact of dairy buffalo farms in Northern Italy. Five farms were considered. Animal welfare was assessed through the protocol (CReNBA) developed by Italian National Animal Welfare Reference Centre and National Reference Centre on Water Buffalo Farming and Productions Hygiene and Technologies (CReNBuf) that considers animal-based measures, farm management, personnel, facilities and equipment. The environmental impact of farms was assessed in a 'cradle to farm-gate' Life Cycle Assessment approach. Both on-farm and off-farm emissions were considered and calculated with Tier 1 and 2 procedure of IPCC. The impact categories were climate change, acidification and eutrophication. 1 kg of normalised buffalo milk (LBN) was used as functional unit. The assessment was performed either without allocation or using the physical allocation method which also accounted for meat production.

Considering the CreNBa protocol, the farms obtained an average total score (mean ± S.E.) of 64 ± 1.5%, and all the farms had a score higher than 60% that is the threshold for assuring an acceptable level of animal welfare. Considering the environmental impact and the no allocation method, the average greenhouse gas emission (GHG), acidification and eutrophication emissions were 10.94 ± 2.57 kg CO_2eq/kg LBN, 70.34 ± 6.53 g SO_2eq/kg LBN, and 15.46 ± 1.14 g PO_4^{3-}/kg LBN respectively. The enteric and the off-farm emissions represented 71% and 16% of the total GHG emissions, respectively. With the physical allocation method, that apportioned the emissions not only to milk, but also to meat, the calculated values were 5.30 ± 1.25 kg CO_2eq/kg LBN, 36.36 ± 6.65 g SO_2eq/kg LBN and 7.98 ± 1.32 g PO_4^{3-}/kg LBN for GHG, acidification and eutrophication respectively. The results of this study can contribute to communicate the positive externalities of the dairy buffalo husbandry system to the consumers.

O171

Risk factors associated with the expression of cross-sucking and intersucking in buffaloes

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The incidence and the potential risk factors for cross-sucking in calves and intersucking in lactating cows were assessed in 73 buffalo farms located in the area of PDO 'Mozzarella di Bufala Campana'. Two trained assessors collected the data through a questionnaire including resource and management measures, which were directly taken and asked to the farmer, respectively. The incidences of cross-sucking, intersucking and mortality were also asked to the farmer. The farm was used as experimental unit. For each discrete risk factor, an ANOVA was performed to assess the association with outcome variables (i.e. the incidence of cross-sucking and intersucking), whereas for continuous risk factors the association was tested using Pearson correlation coefficients. Cross-sucking and intersucking were observed in 91% and 45% of the farms, respectively; their mean incidences were 21.9 ± 22.3 and 1.2 ± 1.9 (mean ± SD), respectively. Cross-sucking was negatively correlated with the number of calves in the collective pen ($n = 34$, r= –0.47, p=.01). Furthermore, cross-sucking tended to be positively correlated with calf mortality from 24 h to weaning ($n = 34$, r=.27, p=.11). The incidence of intersucking was positively correlated with the number of buffalo cows on the farm ($n = 73$, r=.26, p=.03), the number of lactating buffalo in the pen ($n = 73$, r = 0.23, p=.04), the percentage of weaned calves cross-sucking ($n = 73$, r=.29, p=.01), the percentage of heifers and pregnant heifers cross-sucking ($n = 73$, r=.61, p=.001 and $n = 73$, r=.52, p=.001; respectively). As expected the percentage of buffalo cows with nose-ring was highly correlated with the incidence of intersucking ($n = 73$, r = 0.75, p=.001). Moreover, the expression of cross-sucking in the categories of weaned calves, heifers and pregnant heifers induced a higher incidence of intersucking ($F_{1,69}$=5.5, p=.02, $F_{1,70}$=28.8, p=.0001 and $F_{1,71}$=15.5, p=.0002; respectively). We conclude that appropriate management strategies should be implemented in order to reduce the incidence of cross-sucking and intersucking. In addition, the onset of cross-sucking in replacement stock, such as calves and heifers, may favour the development of intersucking in their later lives.

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O172

How can we understand the emotional state of cattle for welfare evaluation purpose?

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This study explored whether the visible eye white and ear posture can reliably contribute to interpret the level of arousal in dairy cows. The research was conducted in five Italian farms. Lactating cows were loose housed in cubicles, except in farm 3 (tie stalls), and they were fed a TMR, except in farm 5 (ventilated hay and fresh grass). In farm 5, cows also had daily access to pasture. More than 500 photos of cows' heads were taken during feeding, resting, grazing and an avoidance distance test at the feeding rack (ADF). From these photos, for each animal we classified eye white as 'high arousal' (EWhigh =1: eye white visible), and 'low arousal' (EWlow =0: no visible white, including half-closed eye); ear posture was classified as 'high arousal' (EPhigh =1: ear held upright or horizontally), and 'low arousal' (EPlow =0: ear held backwards or hung down). Eye white and ear posture were significantly correlated (Spearman rank correlation test: $\rho = 0.482$; p=.001). The context significantly affected the level of arousal (Chi Square test: p=.001): the highest percentage of EWhigh (95.5% of cows) and EPhigh (70.1%) were recorded during ADF test, as the most stressful situation, whereas the highest percentage of EWlow (77.3%) and EPlow (86.4%) were observed during grazing, when cows are usually relaxed. During feeding, the highest level of arousal was observed in farm 3 (EWlow =28.6%), where cows were housed in tie stalls, and 2 (EWlow =55.1%), that had the lowest ratio feeding places/cow (0.68), and the lowest level of arousal was recorded in farms 4 and 5 (EWlow =81.8% and 81.3%, respectively; Chi-Square test: p=.028). In farm 5, all cows with low arousal had half-closed eyes, which suggests that they were in a positive mood, probably due to the high number of feeding places/cow (1.72 feeding places/cow) and to the preference of cows for hay and grass, rather than for TMR distributed in all other farms. During testing, the level of arousal was significantly lower in farms 4 and 5 (Chi Square test: p=.001), that had the higher proportion of cubicles per cow (1.15/cow and 1.12/cow, respectively): EWlow was 85.4% in farm 4 and 100.0% in farm 5 and EPlow was 72.1% in farm 4 and 92.6% in farm 5. Results suggest that eye white and ear posture are indicators of the level of arousal in dairy cows, which may depend on some resource-based factors, and support their inclusion for welfare evaluation.

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O173

Behaviour assessment of different poultry genotypes organically reared with computerised video recording system

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The Regulation (EC) n. 889/2008 suggests that in organic production the breed choice should take into account the capacity to adaptation to local conditions. Unfortunately, the choice of genotypes is not compulsory and often the farmers base the choice only upon the productivity aspect.

The aim of the present study was to evaluate the behaviour of six different chicken genotypes organically reared using a computerised video recording system. The studied genotypes were: Ranger Classic (R1) Ranger Gold (R2), Rowan Ranger (R3), Campese (C), CYgen5 × JA87 (CY), M22 × JA87(M). The behaviour analysis was carried out through a Media Recorder for the video capture. The evaluation was done on the initial interest of bird and its activity from 5 to 10 m to the shelter. All the videos were elaborated using the Observer XT with a coding scheme that permits the identification of each behaviour. The main analysed behaviours were: resting, walking, roost, eating feed, eating grass, drinking and comfort. Data were statistically elaborated with a linear model and Bonferroni multiple *t*-test. The results showed a higher initial interest of the R3 and C genotypes, on the contrary, CY and R performed a low exploratory activity. Moreover, no subject, belonging to the M genotype, leave the shelter during the established time unit (5 minutes).

At 5 m from the shelter the genotypes did not show any difference on the walking behaviour. R3 had the lower frequency of rest and C the higher, CY exhibited intermediate frequencies. The roost behaviour showed a large variability between genotypes: R3 genotype exhibited the lower frequency; M and R1 were the genotypes that most expressed this behaviour while C, R2 and CY genotypes showed intermediate frequencies. The most frequent feed behaviour was observed in R2 genotype, while the R3 showed higher grass consumption.

At 10 m from the shelter the M and CY genotypes were observed occasionally due to their low explorative and kinetic activity. Concerning walking and resting behaviours there was no significant difference between the genotypes. R3 showed a low roosting behaviour followed by C. At this distance, R3 exhibited higher feed consumption while C higher grass consumption.

In conclusion, the behavioural analysis suggests that CY and M genotypes showed less adaptability to the organic rearing systems, on the contrary, C and R3 seem more suitable due to the active behaviours and grass intake at both the distances from the shelter.

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LIVESTOCK SYSTEMS – COMPANION ANIMALS II

O174

Interplay between salivary biomarkers and gut microbiome in dogs

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The study aimed at evaluating the role of a standardised polyphenol phytocomplex from grapes (GP) on the microbiome-gut-brain axis in dogs. For the trial, 24 dogs were recruited in a local shelter. The animals were housed in separated box and were adult, clinically healthy and of both sex and of mixed breed or different pure breeds. The dogs were assigned to 3 groups of 8 subject each and fed the same commercial kibble diets for 2 months before the beginning of the study. One group (CTR) received a placebo tablet without GP, the second group (D20) and the third group (D60) was supplemented with tablets to achieve 1 or 3 mg/kg live weight of GP, respectively. At day 0 (T0), after 14 (T14) and 28 (T28) days, faecal and saliva samples were collected from each dog. Furthermore, at T0 and T28 the hair was also collected. The DNA of faecal samples were extracted with commercial kit and sequenced in 300 bp pair ends for V3 and V4 regions of the 16S rRNA using an Illumina platform. Reads were annotated using the QIIME bioinformatic pipeline and operational taxonomic units used to build the taxonomic tree. Faecal samples were also analysed for short chain fatty acids (SCFA) and lactic acid (LA). The serotonin and cortisol concentrations of saliva and cortisol in hair were analysed with ELISA. The supplementation of GP to the canine diet caused a significant increase ($p<.05$) of serotonin and serotonin/cortisol ratio in the D60 group in comparison to CTR and D20 groups, suggesting a positive interplay between microbiota and the host. The endocrine variations were associated with some modifications of relative abundances of microbiota at a genus taxonomic level. Minor differences were observed for the faecal concentrations of SCFA and LA. Principal component analysis revealed that *Firmicutes* and *Bacteroidetes* phyla accounted for 93.6% of total variance (87.6% and 6.2%, respectively). However, not a clear clustering of the groups of dogs was observed. The results suggest that GP interferes with gut microbial communities, modifying their interaction with the host. If the observed variations of salivary serotonin are related to change in tryptophan metabolism in the gut deserves further evaluations.

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O175

In vivo and *in vitro* digestibility of extruded dog foods with *Hermetia illucens*

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Edible insects were used as high-quality and new-source of proteins. The larvae of black soldier fly (*Hermetia illucens*) is one of the most studied insect used as source of protein in feed. The aim of this study was to evaluate the effect on nutrient digestibility of the inclusion of larvae Hermetia illucens (HI) as source of protein, in extruded dog food.

Two extruded dog foods were formulated to meet FEDIAF dog nutrient requirements. In the test diet, the source of protein was represented by Hermetia larvae meal (HI) and the control diet was represented by a food with the same formula but where the source of protein was deer by-products meal. Substrates were analysed for dry matter (DM), organic matter (OM), ash, crude protein (CP), ether extract (EE), crude fibre (CF), Ca, P and amino acid (AA) contents and for *in vitro* and *in vivo* digestibility of DM, OM and CP. Test Protocol for *in vivo* trial used was total collection method and a Latin square scheme, to test same dogs. The chemical analyses were determined according to FEDIAF (2018) methods, and gave respectively following results HI 94% DM (94.17% MO, 5.82 Ash; 20.69% CP; 15.6% EE; 1.89% CF as DM) and control 92% DM (92.37% MO, 7.69 Ash; 16.97% CP; 17.4% EE; 2.06% CF as DM). Combined requirement for Cystine and Methionine was limiting AA.

The value of DM digestibility was higher for HI food. Preliminary results suggest that insects may be practical alternatives to traditional protein in pet foods. This higher digestibility could be explained with a lower collagen content in Hermetia meal respect to deer meal but this hypothesis needs to be confirmed with hydroxyproline analysis.

O176

In vivo and *in vitro* digestibility, palatability and nutritive quality of extruded dog food based on mechanically separated chicken meat or meat by-product

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Fresh meat, such as mechanically separated chicken meat (MSCM), is usually listed first as an ingredient on pet food labels when selected by industry as a protein component. This commercial strategy may influence consumer choice in the comparison among pet food products. Since most dry pet foods are produced by food extrusion, it is important to compare the nutritional value of a dog diet based on MSCM with one based on meat by-products (MBP), such as processed animal proteins, to determine whether the 'fresh meat' is the better choice. The objectives of this study were: (a) to evaluate the effect of inclusion of MSCM in dry dog food on fatty acid (FA) profile, *in vivo* and *in vitro* digestibility, and palatability as compared with dry dog food containing MBP; (b) to determine, whether or not, the inclusion of the one or the other ingredient changes the microbiology and the storage quality of the two food products; (c) to propose a new system (Daisy^{II} Incubator) to measure the *in vitro* digestibility of the two products. Despite their similar chemical composition, the MSCM product had lower palatability but better nutritional quality (with higher polyunsaturated FA content and lower saturated FA content) than the MBP product. The n-6/n-3 polyunsaturated FA ratio was 11.5 and 16.7 for the MSCM and the MBP diet, respectively. Microbiological risk assessment showed no microbiological hazards for either product. Q21 After 6 months storage, the total mesophilic bacterial count ranged between 1.77 log CFU/g and 2.09 log CFU/g feed, while polyamine values were found to be higher in the MSCM (0.37 g/kg) than in the MBP (0.27 g/kg). Our results indicate that MSCM or MBP may be advantageously included in pet food formulation as a source of animal protein. Moreover, the Daisy^{II} Incubator proved a valid instrument for the study of *in vitro* digestibility for dogs. Since it provided data simply, quickly, with less variability and cost than obtained with *in vivo* trials, it could represent the future for pet food digestibility studies.

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O177

Genetic variability assessment in different Mediterranean canine breeds using microsatellite markers

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The domestic dog (*Canis familiaris*) was the first species to be domesticated. Today, the 'Fédération Cynologique Internationale' distinguishes and recognises 360 canine breeds, classified in 10 ethnological groups. Group 5 'Spitz and primitive type' is subdivided into 7 sections. The Kelb tal Fenech (KTF) is allocated in section 6, while section 7 includes the Italian breed Cirneco dell'Etna (CIE) and different breeds (Valenciano, Ibicenco, Canario, Andaluz) of the Spanish Podenco (POD).

The KTF is Malta's national dog, esteemed for rabbit hunting. Some scholars suggest that it descended from the Ancient Egyptian Tesem. Two more canine breeds were well-regarded for rabbit hunting: the CIE, a Mediterranean breed from Sicily, and the POD originated in the Canary Islands.

All these breeds are morphologically quite similar, and it is tempting to assume that they are genetically related. Besides their common hunting utilisation, the genetic diversities and relationship within and among these dog breeds were investigated using STR markers. The genetic analysis was conducted using blood samples from each breed as source of DNA. Following the DNA extraction, multiplex PCRs were carried out using a panel of 17 ISAG suggested microsatellites. One breed (German Shepherd) was included as an outgroup. The following parameters were analysed: genetic variation, genetic distance, genetic structure and admixture.

An average of 6.30 alleles per locus were observed in the six studied breeds. The mean number of observed alleles ranged from 4.44 (KTF) to 7.88 (POD Valenciano). The highest observed heterozygosity (0.73) was detected in POD Andaluz, whereas the lowest (0.56) in KTF. The inbreeding coefficient estimated ranged from −0.0296 in POD Canario to 0.0670 in POD Valenciano. The Reynolds weighted genetic distance revealed three distinct clusters; the first cluster included the four POD populations, the second had CIE, while the last cluster consisted of KTF. The structure analysis results ascertained that some level of genetic admixture is reported especially in POD populations. KTF and CIE seem to have a clear genetic identity even if some genetic admixture with POD is highlighted for CIE.

The findings strongly suggest that the KTF and the POD Ibicenco seem to have a distinct genetic identity probably due to its geographic isolation. A genetic introgression of POD populations in CIE is appreciable. Moreover, a certain degree of genetic admixture in POD populations is detected.

O178

Metigree: a genomic tool to enhance the well-being of Italian mongrel dogs

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Mongrels and mixed-breed dogs are animals that do not belong to any of the officially recognised breeds. Figures vary, but one recent paper reported that 75% of the 1 billion dogs worldwide are free-ranging and breeding. Applying the same percentage to the current 7 million dogs in Italy would amount to 5 million mongrels, of which about 4 million have owners, 0.2 million are in pounds, and 0.8 million are strays.

Our research aimed at determining the ancestry of mongrel dogs to further promote, protect and safeguard their well-being. Saliva samples from 20 mongrels collected according to European rules were genotyped using the canine 230K SNP chip.

Using a combination of ranked supervised admixture and haplotype sharing, we compared our results with a database of 183 breeds and populations (including 24 Italian breeds and populations, wild dogs and wolves).

By comparing the percentages of genomic background associated with one or more breeds, we obtained 3 types of results when assigning membership breeds to each of our 20 dogs:

i. 'real mongrels': the first breed accounts for less 10% of their genomic background with no phenotype consistent for any breed of reference, with the exception of the size;
ii. 'mixed breeds': the first breed accounts for 11 to 74% of their genomic variance with phenotype resemblance ranging from mild to high;
iii. 'purebred dogs' without official pedigree: the first breed accounts for 75% or more of their genomic background.

We propose coining the word METIGREE, deriving from the Italian 'METIccio' (mongrel) and 'pediGREE', to reconstruct ancestral line for mongrels by using genomic analysis.

This scientific identification of mongrels would increase their chances of adoption and further our knowledge of their temperament and health issues, thus reducing medical costs while promoting the dogs' and their owners' happiness.

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LIVESTOCK SYSTEMS – NEW EMERGING TECHNOLOGIES IN ANIMAL SCIENCE

O179

Conventional culture, MALDI-TOF and 16S rRNA compared for test agreement in diagnosis of bacteria in bovine milk samples

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Comparison of culture, matrix-assisted laser desorption ionisation – time of flight (MALDI-TOF), and 16S rRNA genomic sequencing to identify mastitis pathogens was the objective. Milk (10 µl) from quarter samples submitted to The Dairy Authority (TDA) lab was streaked onto Columbia blood agar (CBA), MacConkey agar and Modified Hayflick medium. Colonies isolated within 48 hr were subcultured onto CBA, plates were paraffin sealed and shipped overnight to the University of Missouri (MU). Culture at TDA identified bacteria to the genus level except for *Staphylococcus aureus* and *Escherichia coli* speciation, and grouping of streptococcal-like organisms, per standard mastitis diagnostics. Mycoplasma speciation PCR was available if mycoplasma were isolated. At MU colonies were tested in duplicate using a MALDI-TOF mass spectrometer (Bruker Daltonics). Comparison with the Biotyper known bacteria database produced identification scores from 1.7 to 1.99 for genus-level identification and ≥2.0 for species level. 16S rRNA colony lysate PCR products were Sanger sequenced and compared to GenBank data using nucleotide-BLAST at MU. Microbiologists were blind to other test results. Positive test agreement (same microbe identified by different tests) analysis used McNemar's test; overall test agreement (whether negative for same organism by different tests also included) used Kappa test. Culture and MALDI-TOF tested 181 isolates; 16S rRNA tested 179 (2 were lost in storage). No *Streptococcus agalactiae* or *Mycoplasma* spp. were detected. Overall agreement between all 3 diagnostic methods was 94% (169/179); agreement between 16S rRNA and MALDI-TOF was 98% (176/179), both good by McNemar's test. Culture agreement with each of the other 2 methods was 95% (170/179 with 16S rRNA, 171/181 with MALDI-TOF). For individual pathogens, positive agreement ranged from 90% to 100%, nearly all 'good' by McNemar's. Overall agreement (negative agreement included) was 97% to 100% among all 3 methods, all 'very good' by Kappa. For 22 isolates defined by culture as *S. aureus*, *Enterobacter* spp., *Klebsiella* spp., *Pasteurella* spp., or *T. pyogenes*, agreement among all methods was 100%. These results suggest that for usage in milk quality and udder health monitoring, any of the 3 methods are valuable tools for the dairy industry.

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O180

Angiotensin Converting Enzyme-1 inhibitory activity of milk proteins evaluated after *in vitro* digestion and peptidomic analysis

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Milk proteins are relevant sources of bioactive peptides. Many hurdles still exist regarding the widespread utilisation of milk protein-derived bioactive peptides as they may be degraded during gastrointestinal digestion. A crucial issue in this field is the demonstration of a cause-effect relationship, from the ingested intact form to the bioactive form.
The aim of this study was to evaluate *in vitro* digestion, digestibility (IVD, using two different hydrolysis methods) and Angiotensin Converting Enzyme-1 inhibitory activity (ACE-1i) of milk and plant proteins (used as control). Based on ACE-1i activity, a peptidomic and proteomic profile analysis was performed on permeate and retentate samples.
In particular, milk and plant protein samples were *in vitro* digested, and the total digest was filtered using a 3 KDa membrane. A permeate fraction (<3 KDa) and retentate fraction (>3 KDa) were obtained. ACE-1i activity was measured as the ability of protein fractions (pre-digested, permeate and retentate) to decrease the hydrolysis of furanacroloyl-Phe-Glu-Glu (FAPGG) synthetic substrate for ACE enzyme. Furthermore, permeate was characterised by LC-nano ESI MS/MS using a shotgun-peptidomic approach, whereas retentate was further trypsin-digested prior the analysis with mass spectrometry using a shotgun-proteomic approach.
We found a positive correlation among the IVD methods tested ($p<.05$; $r = 0.85$). Milk proteins exhibited higher values of IVD

(>82.5%) with both methods used, compared with plant proteins. Milk proteins after *in vitro* digestion exhibited a significant increase in ACE-1i ($p<.05$) (> 23.91 ± 0.64%) compared with plant protein tested (10.40 ± 1.07%). Based on proteomic and peptidomic analysis performed, specific peptides associated with anti-hypertensive and ACE-1i effect have been identified in permeate and retentate fractions of milk proteins. Our results demonstrated that milk and plant proteins are highly digestible and, in particular, milk proteins may represent valuable sources of ACE-1i and anti-hypertensive peptides which may confer the ability to decrease blood pressure *in vivo*.

O181

The ability of the partial least square regression to predict the number of bites in Sarda dairy sheep by using acceleration data

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The number of bites taken by a grazing sheep is an important component of the equation that estimates the herbage intake. Recent development in technologies has promoted the use of tri-axial accelerometer devices to study the behaviour of grazing animals. Such instruments can produce a set of variables in the three dimensions that, combined each other, can accurately discriminate between grazing, ruminating and other activities. Number of bites can be also estimated with a moderate accuracy from the sum of accelerations recorded in X-axis with linear regression models. The aim of this study was to predict number of bites using all accelerometer variables with a multivariate approach, the partial least square regression (PLSR), that has become an established tool for modelling linear relations when a set of dependent variables has to be predicted from a set of independent variables highly correlated. For the scope, an experiment has been conducted under controlled conditions to test if the number of bites can be predicted from accelerometer data. Ten dairy Sarda sheep were fitted with a halter equipped with an accelerometer (BEHARUM device) and subjected to short term tests (6 min) using micro-swards of Italian ryegrass (*Lolium multiflorum* L.), alfalfa (*Medicago sativa* L.), oat (*Avena sativa* L.), chicory (*Cichorium intibus* L.) and a mixture (Italian ryegrass and alfalfa). Each animal was video recorded during the test to detect the number of bites. Accelerometer data sum, mean, variance and inverse of coefficient of variation calculated for the X-, Y- and Z-axes and the resultant were summarised with an epoch setting of one minute. A database inclusive of the acceleration variables and the number of bites detected was created. To verify if the acceleration variables could be used as predictors of the number of bites, the partial PLSR model was used. The precision and accuracy of PLSR predictions were evaluated implementing the Model Evaluation System, in which the predicted values were regressed against the observed ones, based on r^2, root-mean-square error of prediction (RMSEP) and Dent & Blackie test. The PLSR showed an overall good accuracy (Dent & Blackie test P = 1) and was proven precise for the estimation of number of bites (r^2=0.77, RMSEP =0.21).
To conclude, PLSR procedure can accurately estimate with high precision the number of bites of sheep equipped with the BEHARUM device and short term grazing Mediterranean forages.

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O182

Evaluation of the foal Esperia ponies' mortality attributable to predation by roaming dogs or wolves

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The Esperia's Pony is a horse population living in the Mountain *Aurunci* and *Ausoni* area. The survival of free-range livestock maintains a human presence in areas that seem destined to the abandonment and degradation thus, exposes ponies to the roaming dogs and wolf's predation. Aim of the study was to analyse the temporal distribution of foals' mortality related to complaints of wolf attacks presented by horse breeders. Number of mares, deliveries and foal deaths in a group of ponies in the *Appiolo* Mountain (300 h) were analysed over 15 years (2002–2016). Differences in foals mortality between years were evaluated. During the study period, predatory complains were classified as canids attacks because it was not possible to ascertain if they were due to roaming dogs or wolves. Results indicate that from 2002 to 2006 the number of mares increased, as well as the number of births (total average 113 ± 5.48/year). The foaling rate was highest in 2002 (97.3%) and progressively decreased. Foals mortality was low (<14%) from 2002 to 2009 and predation complains were absent in the same period. Canids attacks began in 2010 and foaling mortality became significant in 2011 (14.1% vs. 28.8% respectively in 2010 and 2011; $p<.01$). Foaling mortality continued to increase in the 2012 and 2013 (42.2 and 63.4%, respectively in 2012 and 2013; $p<.05$ in 2012 and $p<.01$ in 2013). The mortality significantly decreased in 2014 compared to 2013 rate (39.8% vs. 63.4% respectively in 2014 and 2013; $p<.05$) but remained still significantly higher ($p<.01$) compared to 2010. The highest rate of mortality was recorded in May (31%) and in June (28.2%), becoming residual in September (2.4%), October (0.9%) and November (0.9%). Data analysis suggest a significant increase of the attack complains, an influence of predation on the reproductive mares' outcomes, and a seasonal occurrence of mortality. Even if camera traps showed the presence of wolves in the study area (April–November), the consistency of the wolf population is still unknown, and information about the role of roaming dogs are lacking. Further studies are necessary to an integrated management plan in a protected area, including actions to conserve both endangered animals and the safeguard of sustainable activity.

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POSTERS

ANIMAL BREEDING AND GENOMICS

P001

Estimated breeding values of differential somatic cell count in Italian Holsteins

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Recently, we investigated the genetic aspects of milk differential somatic cell count (DSCC), a novel indicator of udder health that corresponds to the sum of the percentages of neutrophils and lymphocytes in the total somatic cell count (SCC). Information about the proportion of the different cell types in milk, in combination with the overall SCC, would be a valuable asset to better define the udder health status. Our findings served as a prelude for the potential inclusion of DSCC in breeding programmes aimed to enhance cow resistance to mastitis. In the present study, we reported estimated breeding values (EBV) for DSCC and Traditional Somatic Cell Score (SCS) in Italian Holsteins. Variance components used for breeding value estimation were obtained using a model that accounted for the fixed effects of herd-test-day, stage of lactation and parity, and for the random effects of additive genetic animal, permanent environment and residual. In general, we observed the genetic improvement of milk SCS and DSCC across the year of birth of the sires. Correlation between EBV for DSCC and SCS was calculated, and results suggest that a combination of both traits will help identify animals with the genetic potential for lower DSCC and SCC, i.e. bulls with greater genetic merit for mastitis resistance. In particular, 4 groups of bulls could be distinguished: 'healthy' (with EBV for both DSCC and SCS <0), 'susceptible' (with EBV for DSCC >0 and EBV for SCS <0), 'chronic' (with EBV for DSCC <0 and EBV for SCS >0) and 'mastitic' (with EBV for both DSCC and SCS >0). Mean EBV for DSCC and SCS by groups of bulls were analysed. Further research is needed to assess the proper weight DSCC should receive in a breeding programme aimed to enhance cow resistance to mastitis.

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P002

Preliminary results of survey on native poultry breeds in centre Italy

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The TUBAVI project (Valorisation of Italian Poultry Breeds) aims to recover and to preserve fifteen Italian poultry breeds through the maintenance of their genetic variability; also, the economic valorisation of these breeds is another goal of the project through the study of alternative rearing systems respectful of animal welfare and environmental sustainability.
Poultry breeds in Italy do not have genealogical books, therefore the first step of the project was to make a survey on the consistencies of some native breeds, followed by recording phenotypic (reproductive and morphological) and genotypic chicken data. For this aim, the diffusion of Ancona and Leghorn breeds have been scrutinised in three regions of central Italy (Lazio, Umbria and Marche). In these farms, a survey on the main structures, human resources and reared animals was carried out.

Of the fifteen farms analysed (seven in Lazio, two in Umbria and six in Marche), nine were represented by agricultural farms, while the others consisted in unprofessional farms. The technical skills are inadequate (only 1/5 made specific training courses) and the age of breeders was between 30 and 50 years. Structural characteristics of farms did not show significant differences between Regions. All the farms had poor rearing structures and were equipped with manual feeders and egg-laying nests. The main business of the farms was the selling of animals for poultry shows. Concerning the genotypes, Ancona represented 59% of the animals, and as expected, it was more widespread in its land of origin (Marche). In general, the average number of hens per farm was very low ($n = 15$), exceeding three cases with more than thirty subjects.
In conclusion, it could be affirmed that these two breeds in central Italy are not very diffused and the technical skills of farmers and rearing structures are very poor. The recent creation of a National Register and the possible diffusion of niche market for such breeds could be an incentive for the reorganisation of the Italian chicken breeds and for their conservation.

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P003

Application of genetic selection for ovine PrP resistance alleles in southern Italy

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Scrapie is a neurodegenerative disease affecting ovine and it is one of the several transmissible spongiform encephalopathies (TSEs). Scrapie is recognised as two forms – classical and atypical. Susceptibility or resistance of sheep to classical scrapie is under the control of the host prion protein (PrP) gene polymorphisms at codons 136, 154 and 171. This genetic resistance allowed the application of a breeding programme in Italy finalised to the increase of the frequency of the resistant ARR allele in sheep populations. The aim of this study was to identify the *PrP* gene polymorphisms in a total of 13,937 healthy sheep from Campania and Calabria regions, analysed from the beginning of 2016 to the first half of 2018. We identified 6 alleles and 19 genotypes based on codons 136, 154, and 171 of the *PrP* gene. The most frequent alleles were ARQ associated with higher susceptibility to scrapie (frequency 0.479), and ARR, responsible for higher resistance to scrapie (frequency: 0.428). In general, the ARR/ARQ genotype was predominant (0.360). The most resistant genotype to scrapie, ARR/ARR, was found with a frequency of 0.213. The most susceptible genotype, VRQ/VRQ, exhibited a frequency of 0.003. We also found the rare allele ARK, at a general frequency of 0.003. According to these results, 61.9% of all the genotyped sheep belong to risk groups R1 (21.3%) and R2 (40.6%). In the Calabria region, the presence of the R1, R2 and R3 groups exhibited a uniform distribution, while the risk groups R4 and R5 appeared mainly concentrated in the central area of the region. Our findings highlight the importance of the application of a sheep breeding programme aiming to increase the frequency of the resistant genotypes to reduce the risk of scrapie.

P004

Genetics of some maternal ability traits in the Italian beef cattle

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Genetic improvement in beef cattle has been focussed mainly on productive trait; however, reproductive and functional traits (body condition, locomotion, maternal ability, etc.) have gained interest in recent years. Calving ease and milk yield have been shown to be an important trait for calf survival and its subsequent growth, particularly in beef cattle in open pasture and loose system. Traits related to the udder and milk production can then influence calf growth and weaning weight, which has a direct effect on the farmer's income from livestock activities. This study was aimed to estimate genetic parameters for some maternal ability traits in Marchigiana (M), Chianina (C), Romagnola (R), Maremmana (MM) and Podolica (P) Italian beef cattle. Maternal ability was identified through linear type traits evaluated on cows as the udder volume (UV – evaluated as distance of the bottom part of the udder and the hocks), and teats size (TS). In addition, milk production (MP) obtained from interview to farmers was also analysed. Type traits were scored using a 1–5 scale system, i.e. from very small to abundant volume for UV and from very small to very big for TS. Also, MP, not directly measurable in beef cattle, was scored from 1–5 considering the information available on milking attitude of cows given by farmers. Animal evaluation was carried out on 1548 cows up to third parity by 36 classifiers in the period between 2017 and 2018. Data obtained from the ANABIC data-base included 780 M cows, 305 C, 119 R, 129 MM, and 215 P. Univariate procedure of SAS revealed a normal distribution for all three measured variables. A preliminary GLM of SAS allowed the identification of 3 main non-genetic factors affecting the scores. These factors were identified in the herd-year classifier within breed, the calving distance (i.e. interval from calving to type trait evaluation), and the age at calving by parity (in months). A final across-breed model was then constructed accounting all available data and these 3 factors as fixed in the model. Genetic parameters were estimated under a single trait animal model-REML accounting for 20,561 animals in the pedigree file. Heritability values obtained were intermediate for all traits analysed, ranging from 0.26 for MP to 0.38 for UV. Heritability of TS resulted closer to that of MP, reaching the value of 0.28. The three investigated traits revealed a significant genetic variance indicating the possibility to obtain breeding values for selection.

P005

Project preview on genetic characterisation of Italian local chicken populations using a high-density SNP's chip

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The poultry industry is characterised by a raised constant process of specialisation of genetic resources, commercial hybrid lines and breeding systems. This process brought undisputed improvements in productive performance, but also a progressive erosion of genetic variability and adaptability in local populations. According to a census carried out in 2001, about 61% of the 90 historically known Italian poultry breeds/populations must be considered extinct and only 9% are still widespread. In general, the size of indigenous breeds/populations has drastically reduced over the years with a consequent increase in inbreeding and worsening of performance.

The aim of the present project was based on genetic characterisation of 10 *Gallus gallus* local breeds/populations, through the evaluation of genetic variability, genetic structure and phylogeny of the studied breeds/populations using Single Nucleotide Polymorphisms (600 K SNPs array). The project was developed through three different steps. First, 24 blood samples were collected from each breed/population; then, genomic DNA was extracted from whole blood. Finally, after DNA quality control, the genetic analysis was done by a SNPs array. The results were useful to evaluate and propose conservation indexes based on the average kinship of the individual with the whole population, thus favouring virtuous methods in the mating management in order to minimise the inbreeding rate and encourage the maintenance of the maximum variability existing. The contribution of each breed/population to biodiversity and the extinction risk status was also analysed.

Acknowledgements

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P006

Feasibility of near infra-red spectroscopy to predict faecal composition in Italian Holstein Friesian cattle

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In recent years, feed efficiency has been studied to establish goals for productive and economic traits in dairy cattle. To increase animal production, one possibility is to improve feed efficiency, which means increase animal production or decrease use of resources. The amount of crude protein (CP), starch, ethereal extracts (EE), neutral detergent fibre (NDF) and acid detergent fibre (ADF) in faeces provides a real estimation of cow's diet digestibility, therefore a key point in order to reach feed efficiency goals. Selection for feed efficiency is hardly achievable because individual feed efficiency is not easily measurable on a large scale, due to high laboratory costs, time-consuming and high labour protocols. In the present study, a total of 100 faecal samples of Holstein Friesian cows were analysed in terms of CP, starch, EE, NDF and ADF using the reference laboratory methods. Data obtained by reference analyses have been used to evaluate the feasibility of near-infra-red spectroscopy (NIRS) as an alternative and cost-effective tool to predict the faecal composition of the same samples. Calibration equations were developed using WinISI software, and for each trait the goodness-of-fit statistics considered were coefficient of determination of calibration (R^2_C), coefficient of determination of cross-validation (R^2_{CV}), standard error of calibration (SE_C), standard error of cross-validation (SE_{CV}), and ratio performance deviation (RPD) calculated as SD/SE_{CV}. The lowest value of R^2_C was observed for starch (0.42) Q22 whereas the highest was observed for ADF (0.91). Values of SE_C ranged from 0.21 to 3.28 for EE and CP, respectively. Values of R^2_{cv} ranged from 0.29 for CP to 0.86 for ADF and the values of RPD were 1.19 and 2.68, respectively. The application of such NIR prediction models may allow to collect phenotypic data on large scale level, and therefore may be used for specific animal breeding programmes aiming to enhance feed efficiency. Further analyses are needed in order to enhance the accuracy of prediction models.

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P007

The new visual assessment scoring system in the Italian beef cattle breeds

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The National Association of Italian Beef Cattle Breeders (ANABIC) developed a new visual assessment scoring system that is significantly much simpler compared to the previous one. The work of assessors in the new method is faster; the low heritability traits were, in fact, no more evaluated or processed in minor detail. A great emphasis is given to the breed traits 'descriptors' such as: coat colour, pigmentation and horns; Body Condition Score (BCS) and maternal ability were also considered. This procedure is in agreement with the new selective targets that aim to obtain animals able to produce in harsh environments.
The study was focussed on three Italian beef cattle breeds with identical management and selective targets (Chianina: CH, Marchigiana: MC and Romagnola: RM) in order to compare the old and the new evaluation systems. Only the females that presented repeated evaluations carried out, by the same assessor, both with the old and the new system (93 CH, 1212 MC, 218 RM) were considered. The data were used to estimate correlation by Spearman test and all were significant ($p \leq .01$). The 'size' traits were in a range from 0.41 to 0.53 (CH), 0.32–0.46 (MC) and 0.21–0.44 (RM). The 'dimension' traits were in range from 0.46 to 0.52 (CH), 0.42–0.47 (MC) and 0.18–0.37 (RM). The 'muscularity' correlations ranged from 0.31 to 0.57 (CH), 0.31–0.45 (MC), and 0.16–0.43 (RM). The correlations between BCS, 'teats size', 'milk', 'volume', 'legs correctness' in the new evaluation and the related traits in the old one showed coefficients ranging between 0.22 and 0.38 (CH), 0.48 and 0.38 (MC), 0.19 and 0.34 (RM).
Interesting were the correlations between the total score in the two methods; the coefficients were high and equal to 0.70 (CH), 0.62 (MC) and 0.57 (RM). Because of the small number of observations, the estimated coefficients are quite low. These preliminary results, therefore, do not allow to obtain definitive conclusions; moreover, the comparison between the two evaluation systems is often carried out on animals which were in different physiological ages (in the first heifers and in the cows at first calving). Besides, the wide range of environments and management cause a wide variability in the animal's body condition. As a matter of fact, these results suggest that it will be possible to have a good conformity in the selection progress obtained through the two evaluation systems.

P008

Genotypic and allelic frequencies of *PRNP* gene polymorphisms in goat populations reared in central and South Italy

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Scrapie belongs to a group of diseases known as 'transmissible spongiform encephalopathies or prion diseases' that can occur in sheep and goat. They represent a family of inheritable, slowly progressive and invariably fatal neurodegenerative diseases affecting humans and animals.
The disease, which takes its name from an intense itch, leads the animals to produce even deep wounds rubbing against any surface. It affects adult animals between 2.5 and 4.5 years of age and, only rarely, it is found in animals less than 18 months of age. The incubation period generally ranges from 2 to 5 years. The clinical course of the natural disease ranges from 8 to 24 weeks in sheep and from 2 to 24 weeks in goat.
In sheep and goat, the different polymorphisms of the prion protein gene (*PRNP*) modulate and influence susceptibility or resistance to scrapie. The strong evidences on the resistance of some sheep *PRNP* allele induced the EU Authorities to implement breeding programme to eradicate scrapie from the ovine population. For goats, an EFSA opinion concluded that the K222, D146 and S146 alleles can confer genetic resistance against classical scrapie, although no EU legislative indication has been so far approved.
In order to elucidate the frequencies of these alleles in Italian goat population, 589 samples of blood were collected for the DNA extraction in 10 different goat breeds/populations (Camosciata delle Alpi, Saanen, Grigia Ciociara, Bianca Monticellana, Capestrina, Fulva del Lazio, Facciuta della Valnerina, Teramana, Garganica, and Grigia Molisana) and in crossbreeding, reared in Central and Southern Italy. The entire coding region of *PRNP*

gene was sequenced bi-directionally with an ABI PRISM® 3500 Genetic Analyzer. Sequences were aligned to *Capra hircus PRNP (*Accession number NM_001314247.1*)*. Polymorphisms, leading to an amino acid substitution, were observed in 15 codons. Two silent mutations were observed at codons P42P (ccg/cca) (220 goats) and S138S (agc/agt) (210 goats). Globally the amino acid substitutions were poorly represented, from 0.1% (T142) to 4.8% (Q211), except for the codons K222 (8.8%) and P240 (55%). Five animals were homozygotes for K222: 2 Camosciata delle Alpi, 1 Facciuta della Valnerina, 1 Garganica and 1 Fulva del Lazio.

These findings could be useful to implement a genetic selection plan in goat, like in the ovine species, to increase the frequency of the scrapie-resistant genotypes.

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P009

Sequence analysis and genetic variability of Mediterranean river buffalo prolactin gene (PRL)

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Prolactin (PRL) is one of the most versatile hormones of the pituitary gland in terms of biological actions. In particular, PRL is essential for mammary gland development, lactogenesis and milk protein gene expression. It has also considered as a potential quantitative trait *locus* affecting milk performance traits in dairy cattle. Aims of this study were to detect *PRL* gene polymorphisms and to investigate their effect in Mediterranean river buffalo milk yield. For this purpose, DNA was isolated from the blood collected from 757 unrelated lactating Mediterranean river buffaloes belonging to 14 farms located in Salerno, Caserta and Potenza provinces (Italy). Using primers designed on bubaline genome sequence we amplified and sequenced the entire *PRL* gene of 5 buffaloes randomly chosen. Seven-thousand one-hundred and fifty-one records for milk yield, measured monthly by the official recording system of the Italian Association of Buffalo Breeders on 1081 lactations of 544 buffaloes, were used. Association between *PRL* polymorphism and milk yield was investigated with a mixed linear model. The entire panel of Mediterranean river buffalo DNA samples were genotyped in outsourcing (KBiosciences, Herts, UK) for the exonic SNP. The buffalo *PRL* gene extends over 8.6 kb including 0.858 kb of exonic regions and 7.741 kb of intronic regions plus 361 bp of the 5′UTR. The main feature of the buffalo *PRL* gene is simple architecture. It contains 5 exons ranging in size from 82 (exon 1) to 306 bp (exon 5) and 4 introns from 629 bp (intron 4) to 2715 bp (intron 1). The sequence comparison of buffalo *PRL* gene showed 34 intronic polymorphisms (11 transversions, 22 transitions and 1 single deletions/insertions) and a transition C→T at the 108th nucleotide of the exon 5. The frequency of the *PRL* T108C SNP in the investigated population was 0.88 (genotype distribution: 589 T/T, 156 T/C, 12 C/C). The least-squares means for milk yield indicated a difference of 0.18 kg/day between the genotypes TT (n. subjects 432) and TC (105) or CC (7), whereas both TC and CC showed an average milk production of 8.32 kg/day. However, these differences were not significant. Therefore, this SNP appears to be selectively neutral in relation to milk yield.

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P010

Genetic variation of beta-lactoglobulin (*BLG*) gene and association studies with milk traits in Mediterranean river buffalo Q23

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Beta-lactoglobulin (β-lg) is the major whey protein in ruminants' milk and in these species has been suggested to play a role also in milk fatty acid metabolism in the newborn. In cattle, sheep and Q24 goat the coding gene (*BLG*) has been well investigated and alleles to this *locus* were associated with dairy performances. In contrast, data regarding *BLG* polymorphism and association studies with milk composition traits are scarce in buffalo and carried out only in Indian breeds. Therefore, the aim of the present research was to identify and analyse the variability of the *BLG locus* in the Italian Mediterranean river buffalo and to test the possible associations between polymorphisms and milk fat traits. For this purpose, individual fresh milk and blood samples were collected from 747 unrelated lactating Mediterranean river buffaloes reared in 14 farms in the province of Salerno, Caserta and Potenza (Italy). Three-hundred and six milk samples were used for FA analysis (fat content, fatty acid composition and fatty acid classes) whereas all blood samples were treated for genomic DNA isolation. We sequenced the entire *BLG* gene (over 4749 bp, including 789 bp in exonic regions, from exon 1 to exon 7) plus 229 bp of the 5′UTR and 241 bp of 3′UTR of 2 Mediterranean river buffaloes randomly chosen. The sequenced *BLG* exons had different lengths (from 42 bp for exons 6–179 bp for exon 7) whereas intron lengths ranged from 214 bp (intron 5) to 995 bp (intron 3). Alignments of the sequences among the 2 subjects showed 33 intronic polymorphisms (11 transversions, 11 transitions, 7 deletions/insertions and 4 inversions) and a transition C→T at the 67th nucleotide of the 5th exon. This mutation identifies a silent allele named *BLG* A1. Since the transition removes a *Hsp*92II endonuclease restriction site, the digestion of a PCR product of 353 bp, spanning the 5th exon and part of the 6th exon, allows a correct genotyping. The allele frequency of the *BLG* A1 allele, determined in 747 subjects was 0.70 (genotype distribution: 53 *BLG* A/A, 419 A1/A1, 275 A/A1). The present study suggests that no direct effect of the investigated polymorphism exists on these production traits. Although no association was found, the present work provides a starting point for additional association studies between *BLG* polymorphisms and other phenotypic traits in buffalo, such as milk yield.

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P011

Hong Kong feral cattle: a distinct genetic pool?

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Cattle are a fundamental resource for human livelihood, providing nutrients, traction, fertiliser, leather, fuel and wealth. There are more than 1000 cattle breeds worldwide, many of which are adapted to live and be productive in a variety of ecosystems.
Asia possesses a remarkable share of cattle diversity, hosting 241 breeds and almost 40% of the total world cattle population, with more than 100 million cattle and 53 local breeds. In Hong Kong, a feral cattle population accounts for 1200 individuals with a wide diversity of phenotypes (e.g. body shape with both taurine and indicine features, and pigmentation). The origin of this population is uncertain. Local government veterinary officers have hypothesised that this population is either a rare group of indigenous cattle, possibly related to Indonesian cattle, or that they are a heterogeneous group of mainly South China cattle origin that were released into the wild by farmers.
To shed light on the Honk Kong feral (HKF) cattle population origin and genetic diversity, we genotyped 21 HKF individuals with the Illumina BovineSNP50 v2 BeadChip. A subset of 3943 SNPs, that were heterozygous in two wild *Bos* species, Gaur (*Bos gaurus*) and Banteng (*Bos javanicus*), was selected to lessen ascertainment bias, and used in further analyses. For comparison, we also analysed 379 individuals from 18 cattle breeds, comprising ten European and two African taurine, two African sanga, and four Asian indicine breeds. Preliminary results suggest that, despite their heterogeneous phenotypes, HKF are genetically homogeneous, distinct from taurine, indicine and sanga cattle, and possibly related to wild Asian cattle species. Further studies are underway to explore the origin of this unique population.

P012

Genomic insights into the recent history of the Rossa Reggiana cattle breed

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The Rossa Reggiana is a taurine cattle breed native to the province of Reggio Emilia (Emilia-Romagna Region in Northern Italy), where it represented the most important breed until ∼1950s. Since, the substitution with more productive breeds led to a progressive population decline. From the late 1980s, conservation efforts were implemented on the remaining nucleus of ∼450 individuals, allowing the inversion of the demographic trend and preventing the extinction of the breed.
The present study aims to (i) evaluate the genetic variability of the Rossa Reggiana cattle in the national and international context and (ii) get insights into the recent history of the breed by identifying gene flow with commercial cattle breeds, which occurred during the last 100–150 years, according to historical records.

To this end, 32 individuals (four males and 28 females) have been sampled from 11 farms located throughout the Province of Reggio Emilia and genotyped at 777 k SNP using the high-density BovineHD Genotyping BeadChip.

For comparison, we analysed genotype data from 15 European taurine, four Asian indicine and two African *B. taurus*×*B. indicus* breeds. Genetic variability, effective population size, and population structure were investigated to estimate the genetic health of the population, evaluate breed relationships, and reconstruct past events of migrations and gene flow.

Preliminary results show that the genetic variability of the Rossa Reggiana breed (observed heterozygosity, Ho =0.34, expected heterozygosity, He =0.29) overlaps with the average values for European taurine cattle (Ho =0.32, He =0.31), and also highlights negligible inbreeding (F_{IS}= –0.017). In accordance with historical records, effective population size trend showed a progressive reduction over the last 50 generations. From the genomic point of view, population structure analysis showed that the ancestry composition of the Rossa Reggiana cattle shares common features with other breeds, as Piedmontese and Italian Limousine. Although marginal genomic exchanges were identified (mostly involving Holstein, Danish Red and Brown Swiss), Rossa Reggiana appears to retain its original genetic background.

P013

Whole genome resequencing analyses in gilthead seabream (*Sparus aurata*) identify a major locus determining a colouration defect

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Phenotypic abnormalities are major problems for the gilthead seabream (*Sparus aurata*) aquaculture sector. The most frequent observed defects span from skeletal malformations, lack of operculum and colouration abnormalities. These defects might be due to environmental and genetic factors, and their incidence might increase in inbred populations. Fry that does not fulfil specific standards are discarded causing major economic losses. The application of genomic approaches could be useful to characterise the genetic factors associated with these defects. The *Sparus aurata* genome has been recently sequenced and a preliminary assembled genome version for this species is publicly available. In this study, we used a whole-genome resequencing approach to identify a major locus affecting a colouration defect (i.e. a depigmentation) that was recently observed among cultivated fry of this aquaculture species. Equimolar DNA pools were constructed using DNA extracted from 30 normally coloured and 30 depigmented fries, sampled from the same commercial hatchery. Then whole-genome re-sequencing was carried out from the two DNA pools using Illumina paired-end technology at about 90×. Variant calling and allele frequencies were obtained using the CRISP software, which is specifically designed to handle pooled samples. Fst was calculated on sliding windows of different sizes (ranging from 100 kb to 2 Mbp). Only one sharp Fst peak, located on chromosome 6, was identified from this genome scan, suggesting the presence of a major colour locus in this region. This discovery will lead to the identification of the causative mutation and the development of breeding strategies to eradicate the defect in the analysed gilthead seabream population.

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P014

Comparative evaluation of genomic inbreeding parameters in Italian pig breeds

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The availability of commercial high throughput single nucleotide polymorphism (SNP) genotyping tools in all main livestock species, including the pig, has opened new opportunities to estimate the inbreeding level directly using whole genome information. Among the genomic approaches that have been proposed to this aim, the methods that use a genomic relationship matrix (GRM) and runs of homozygosity (ROH) have been evaluated in several livestock species. ROH, defined as continuous and uninterrupted chromosome portions showing homozygosity at all loci without any heterozygous genotype, can be detected using SNP chip data setting several parameters that might affect the outputs. In this study, we used several approaches to calculate genomic inbreeding coefficients (GRM, SNP heterozygosity and ROH) in a total of 2860 pigs belonging to seven Italian pig breeds (Italian Large White, Italian Landrace, Italian Duroc; Apulo-Calabrese, Casertana, Cinta Senese and Nero Siciliano) and compared these measures with their pedigree inbreeding coefficient. PLINK software version 1.9 was used to calculate SNP heterozygosity and ROH by setting nine different combinations of parameters (the minimum number of consecutive homozygous SNPs included in the ROH, the minimum length that constituted the ROH, the number of missing and heterozygous SNPs allowed in the ROH, the minimum density of SNPs in a genome window, the maximum gap between consecutive SNPs and the number of heterozygous SNPs allowed in the ROH). GCTA software was used to calculate GRM. ROH were grouped in different categories depending on their length. The categories that included the highest number of ROH were the 2–4 Mbp and 4–8 Mbp. Analysis of the genomic inbreeding coefficients indicated a high correlation between all ROH methods ($r > 0.90$). Among all genomic methods tested, in general, all ROH methods provided also the highest correlated coefficients with the pedigree inbreeding coefficients (ranging from 0.60 to 0.85). ROH identified setting no heterozygous SNPs in the ROH showed the highest correlated values with the pedigree inbreeding coefficients. Correlation between genomic and pedigree inbreeding measures was usually higher in the three cosmopolitan breeds (Italian Large White, Italian Landrace and Italian Duroc) than in the autochthonous breeds, mainly due to the lowest pedigree depth available for the animals of these latter breeds. Q25

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P015

Genome-wide association studies for teat number and related parameters in Italian heavy pigs

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Teat number is an important morphological trait that largely influences the mothering ability of the sows. The total number of teats (TNT) is derived by the individual side count (on the right and left lines, NTR and NTL, respectively). Other parameters related to this trait are the number of functional teats in the anterior and posterior parts of the body based on the umbilical line separation (NTA and NTP, respectively). Bilateral symmetry is defined as the absolute difference between the left and right teat numbers. In this study, we recorded all these parameters in 846 Italian Large White and 500 Italian Duroc pigs and wanted to identify loci affecting these morphological traits. All animals were genotyped with the Illumina PorcineSNP60 BeadChip array. Animals were also genotyped at the vertnin (*VRTN*) gene that has been already associated with the total number of thoracic vertebrae and TNT in several pig populations. Results indicated the presence of a highly significant locus on porcine chromosome 7 segregating in both breeds and affecting partial (NTR, NTL, NTA and NTP) and total number of teats. This region encompasses the *VRTN* gene. Conditional association analysis including as fixed factor the *VRTN* gene polymorphism in the statistical model did not report any significant results, suggesting that this gene might harbour the causative mutation for the identified QTL. Obtained results confirmed the role of the *VRTN* gene in affecting teat number parameters in heavy pigs, providing useful information that could be applied to improve reproduction performances in the analysed breeds.

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P016

Dispersion of *Apis mellifera* subspecies in the Emilia Romagna region (North of Italy) estimated using honey DNA analysis

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Different *Apis mellifera* subspecies and ecotypes are well adapted to a wide spectrum of ecosystems. *A. mellifera* subspecies have been grouped into four main evolutionary lineages characterised at the mitochondrial DNA (mtDNA) level: the West and North European M lineage (including: *A. m. mellifera*); the C branch of the East and Central Europe and North-Mediterranean region (including *A. m. ligustica*, which is also known as the Italian honey bee); the Oriental lineage (O) of the Middle East and central Asia; and the African lineage (A), which groups the African subspecies. Conservation of this biodiversity is threated by beekeeping activities that have mainly favoured the widespread dispersion of different subspecies and by the commercial distribution of hybrid queens. Thus, it is recognised that regional efforts to preserve local honey bee genetic resources are extremely important in maintaining *A. mellifera* biodiversity. To this aim, a few programmes have been established in several regions for

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the characterisation and conservation of locally adapted subspecies. In this study, we evaluated the dispersion of different *A. mellifera* subspecies in the Emilia Romagna region using a novel approach that took advantage from the possibility to define the entomological origin of the honey by analysing the environmental DNA that can be recovered from this food matrix. A total of 100 honey samples were collected from Emilia Romagna beekeepers who covered all nine provinces of this region. Extracted DNA was analysed using a PCR based fragment analysis method that we developed from an informative mtDNA region that can distinguish the A, M and C lineages. The C mitotype was observed in all analysed samples. The M and A mitotypes were identified in 17% and 10% of the analysed samples, respectively. A total of 83 samples showed only the C mitotype, seven samples showed both the C and M mitotypes whereas 10 other samples reported all three mitotype fragments (C + M+A). To our knowledge, this study provided for the first time a dispersion map of the different *A. mellifera* lineages in the Emilia Romagna region. These results could be useful to design a regional programme for the conservation of the autochthonous *A. m. ligustica* subspecies.

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P017

Selection signature analyses identify genomic regions highlighting the traditional role of the autochthonous Reggiana cattle breed in the Parmigiano Reggiano cheese production system

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Selection signature analyses based on single nucleotide polymorphism (SNP) chip data are carried out in cattle to identify loci under natural or artificial selection and identify peculiar genetic features that might be useful to describe breed-specific characteristics. Reggiana is an autochthonous cattle breed reared mainly in the province of Reggio Emilia located in Emilia Romagna region (North of Italy). This breed is characterised by a typical red coat colour. A conservation programme, linked to a new brand of Parmigiano-Reggiano cheese made only of Reggiana milk, started in the 1990s. This branded Parmigiano-Reggiano cheese reverted the decreasing trend of the Reggiana population observed in the 1960s–1980s reaching, at present, the number of about 2500 cows reared in about 180 different farms. In this study, we used SNP chip data to identify selection signatures in the Reggiana genome that could distinguish this autochthonous breed from other cosmopolitan breeds and that might be indirectly derived by the specialised use of Reggiana in the Parmigiano Reggiano cheese production system. A total of 3489 bulls of four cattle breeds (Reggiana, n. 168; Brown, n. 749; Simmental, n. 479; and Holstein, n. 2093) were genotyped with the Illumina BovineSNP50 v1 or v2 BeadChip arrays. Reggiana bulls included in this study were all sires born from 1975 to 2010 for which it was possible to obtain frozen semen in 2014. SNPs were used with their coordinates on the latest assembly of the bovine genome (ARS-UCD1.2). Three pairwise Fst and three pairwise Rsb analyses were performed comparing each time the Reggiana breed with one of the other cosmopolitan breeds. The two methods identified partially overlapping regions over the 99.5% percentile. In the three comparisons, a total of 17 regions and 24 genomic regions reached this threshold for the Fst and Rsb methods, respectively. Selection sweeps on chromosome 6 were identified with all methods, even if partially overlapping. Reggiana vs. Brown detected the highest number of regions (nine with Fst and nine with Rsb). Several genes affecting milk production and morphological traits included in these regions might explain genetic differences that could, at least in part, define Reggiana breed features needed to maximise milk production quality and cheese making properties.

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P018

Water buffalo (*Bubalus bubalis*) susceptibility to bovine tuberculosis is influenced by g.4002C > T polymorphism in interleukin-10 gene

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Interleukin-10 (IL-10) is a regulatory cytokine produced by different innate and adaptive immune cells during bovine tuberculosis (bTB) an infectious disease caused by *Mycobacterium bovis*. bTB is endemic in many countries where it is responsible for economic losses and is still considered a risk factor for humans. The interaction of *M. bovis* with its hosts is long-dated and therefore its genetic makeup may reasonably play a crucial role in the resistance to the pathogen. Thus, because polymorphisms in cytokines may have an important role in bTB, we decided to investigate the genetic variability in IL-10 of water buffalo related to resistance/susceptibility to bTB. To achieve the aim, we used 184 animals reared in different herds located in Campania region (south Italy) and grouped in cases (positive to hypersensitivity and microbiological test, 59 samples) and controls (negative to multiple hypersensitivity test, 125 samples). We amplified and sequenced a genomic DNA region of 300 nucleotides spanning the coding region and following alignment, we identified a point transversion in the position g.4002C>T, which is responsible for an amino acid substitution p.(Thr175Met) in the primary protein sequence. The analysis of all samples revealed that homozygous subjects TT are more represented in the cases group (22 out of 59; frequency: 0.37) compared to control groups (24 out of 125; frequency: 0.19); conversely, the genotype CC is more common in the control groups (52 out of 125; frequency: 0.42) compared to the cases group (14 out of 59; frequency: 0.27). The comparison of the CC vs. TT ratio between case and control subjects by Fisher's exact test, showed an odds ratio (OR) of 2.97; with the 95% confidence interval ranging between 1.231 and 6.667; p=.0088. This result strongly suggests a predisposition of TT subjects to bTB because the OR is in the range between 2.1 and 5.7. Moreover, as expected in case of association, genotypic frequencies at the *IL-10* locus were in equilibrium among controls and in disequilibrium among cases. In conclusion, our data demonstrated that susceptibility to bovine tuberculosis in water buffalo is influenced by the polymorphism g.4002C>T in *IL-10* gene probably consequent to a change in the protein secondary structure. This polymorphism might represent a valid tool for marker-assisted selection against bTB in water buffalo.

P019

Validation of a 14 STRs panel for parentage analysis in water buffalo

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The PCR-based STR profiling technique is widely used in forensic science, evolutionary and ecological studies, both for human and animal species. Microsatellites are ubiquitous, generally highly polymorphic, codominant and relatively easy to screen. On the other hand, genotyping errors can occur due to low DNA concentrations, preferential amplification of small alleles or stutter products caused by slippage during PCR amplification.

Water buffalo (*Bubalus bubalis*) is an economically important animal species bred in southern Italy, and breeding farms complying with high-quality management systems take advantage of parental diagnosis of newborns for genealogical certification in order to estimate the genetic variability.

In this study, we explored and validated a 14-plex PCR system for water buffalo genotyping.

Blood samples were collected from 5182 water buffaloes belonging to several farms in southern Italy. A 14-plex PCR system for parentage analysis in water buffalo was selected based on the PIC (Polymorphic Information Content) score as recommended by both the International Society for Animal Genetics (ISAG) and Food and Agricultural Organization (FAO). Two sets of primers were prepared, and each primer set was run in a multiplex touchdown PCR. The PCR products were resolved on an ABI Prism 3500 Genetic Analyzer. Microsatellite fragment analysis was performed by Gene MapperTM v. 3.7. Sensitivity was determined by analysing genetic profiles generated by different DNA concentrations, while species specificity was tested by analysing non-water buffalo samples (bovine, sheep, dog and pig DNA). Complete profiles were obtained from a minimum DNA amount of 187 pg with an analytical threshold of 50 relative fluorescence units. No amplification products were obtained from sheep, dog and pig DNA, while amplification peaks could be detected for half of the analysed loci from bovine DNA. Each locus included several alleles, ranging from 3 to 7. The observed and expected heterozygosity ranged from 0.62 (CSSM60) to 0.87 (CSSM42) and from 0.51 (CSSM38) to 0.76 (CSSM42), respectively. The allelic average size, standard deviation, intra-locus peak height ratios and allele frequencies were calculated for each allele at each locus.

Our results confirm the ability of our system to produce accurate and reliable genetic profiles and highlight the usefulness of our 14-plex PCR system for water buffalo parentage analysis.

P020

Investigation on muscle fatty acid composition in Italian Large White pigs and association study with *ACACA*, *ACLY*, *SCD* and *FASN* genes

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In recent years, a growing awareness of the association between diet and health has led nutritional quality to become a relevant factor in consumers' food choices, in particular, meat products with lower omega-6 to omega-3 ratio and enriched in omega-3 polyunsaturated fatty acids (PUFA) are preferred. However, consumers' dietary demands and technological requirements by the meat industry do not completely match: PUFA are more likely to incur in lipolytic and oxidative processes than saturated fatty acids (SFA), causing rancidity, abnormal flavours, fat softness and altered organoleptic properties of pork and dry-cured hams. The aim of this research is the identification of genetic associations between muscle fatty acid (FA) composition and candidate genes related to fat metabolism. DNA extraction was performed using *Semimembranosus* muscle samples of 430 Italian Large White pigs. FA composition was determined by capillary gas-chromatography and backfat thickness (BFT) was measured by calibre on the median line of each carcase. Single Nucleotide Polymorphisms (SNPs) in Fatty Acid Synthase (*FASN*), *Stearoyl-CoA Desaturase Delta-9* (*SCD*), *Acetyl-CoA Carboxylase Alpha* (*ACACA*), and *Fatty Acid Synthase* (*ACLY*) genes were utilised for this research. Statistical analyses were carried out using the MIXED procedure of SAS 9.4 considering sex, slaughtering day and genotype of the analysed SNPs as fixed effects; BFT was considered as covariate; sire effect was included as random factor. The *p* values were corrected using the adjusted-FDR values calculated by SAS MULTTEST procedure. Contrasts among the estimated least square means were calculated using the Tukey–Kramer test. Additive and dominant effects were obtained by *t*-test. Considering the adjusted *p* values, *FASN* and *SCD* SNPs resulted significantly associated with FA composition. *FASN* TT genotype showed an association with the content of C12 and C14 saturated FAs, in agreement with the biological role of this enzyme in the FA chain elongation. *SCD* TT genotype shows the highest monounsaturated FA content in agreement with the biological role of SCD enzyme in FA desaturation. The detected association between SNPs and parameters of fatty acid composition, once validated, could be useful to improve the knowledge of genes affecting pork FA composition.

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P021

Body weight estimation from linear measures in Cornigliese sheep breed

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In sheep meat breeds, it is important to accurately estimate the animal body weights, in order to slaughter them at the most convenient time. However, especially in extensive farms, it is really challenging to accurately measure this parameter, since scales are not usually on site. Thus, it is useful to estimate the body weight (BW) from equations obtained starting from body measures. The aim of this research is to propose equations to estimate the BW from body measurements in the Cornigliese sheep breed, a local breed raised in Emilia-Romagna region, used for meat purpose, with a limited number of heads and a late development. Body weight and linear body measures were submitted to correlation analysis and, subsequently, to multiple regression analysis. Two datasets were used: (1) 303 animals (178 females and 125 males, BW: 4.00–117.80 kg), on which height at withers (HW), chest circumference (ChC) and body length (BL) were detected; (2) 156 animals (109 females and 47 males, BW: 5.15–117.80 kg) on which, in addition to the previously mentioned measures, height at croup (HCr), chest width (ChW), chest depth (ChD) and croup width (CrW) were also considered. On each dataset, two models were applied, containing all variables (model 1 and 3, respectively for dataset 1 and 2) or a group of variables, selected by means of the stepwise procedure (model 2 and 4, respectively for dataset 1 and 2).

Body weight resulted highly correlated with all linear measures (from 0.852 for ChW to 0.950 for ChC; $p<.001$). Models 1 and 3 fitted the data better than models 2 and 4, both for all animals and for the subsamples of females and males. The proposed equations, for all animals, are as follow (body measures in cm):

Model 1: BW (kg) = 0.184 HW +0.980 ChC +0.236 BL −61.708 (SE: 8.21 kg; R^2: 0.910) for dataset 1;
Model 3: BW (kg) = 0.321 HW +0.744 ChC +0.319 BL − 0.206 HCr − 0.305 ChW +0.911 ChD +0.448 CrW − 71.454 (SE: 7.19 kg; R^2: 0.936) for dataset 2.
Model 4 gave best fits, showing lower AIC values, compared to model 2, both for all animals (778 vs. 1586) and for the subsamples of females (563 vs. 942) and males (140 vs. 622). We concluded that BW could be predicted by means of linear body measures in Cornigliese sheep breed. The best fits were obtained when the highest number of body measures was included in the model. Nevertheless, models 1 and 2 can be used more easily in practice in the extensive sheep breeding, since they require less parameters measured in the population.

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P022

Biodiversity in local population of Mediterranean trout in Molise

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Freshwater fishes are particularly interesting due to the high number of endemic species described in Italy. Among these, Mediterranean trout *(Salmo cettii, syn. Salmo macrostigma)* has considerable economic significance for fisheries management, aquaculture and conservation biology. Unfortunately, especially in Italian rivers, due to anthropogenic disturbance as pollution, water depletion for irrigation, dam constructions for power production, habitat alteration and over-fishing, the native Mediterranean trout populations are decreased during the past decades. For this reason, alien trout as the domesticated strains of *Salmo trutta* have been introduced in freshwater basins for the implementation of fishing activities without considering the presence of wild autochthonous populations of the interfertile *Salmo cettii*, leading to the introgression by alien genomes. The restocking with non-native domesticated strains produces the 'founder effect' due to low genetic variability of spawners. The described scenario is also common to Molise watersheds in which no data of genetic variability has ever been reported. Here we present preliminary results of gene variation study to assess trout population structures in Biferno and Volturno rivers. A total of 300 samples in 30 different areas (15 for each river) were collected. Adipose fin tissue fragments were cut, immediately transferred into a tube containing 100% ethanol and, once in the lab, stored at –20 °C until DNA isolation (by Qiagen blood and tissue kit). PCR-RFLP analysis on nuclear gene LDH- C1* using the Ldhxon3F/Ldhxon4R primer pair and *Bsl*I restriction enzyme was carried out.

Preliminary results, carried out on a representative subset of samples, showed a different introgression level considering the two rivers. In the Volturno river drainage, the degree of homozygous specimens for the native allele (*S. cettii*) was 86.9% vs. 13.1% of heterozygous fishes. The Biferno river basin, instead showed 50.0% of specimens with homozygous native alleles vs. 41.2% of heterozygous fishes and 8.8% of specimens with homozygous alien alleles (*S. trutta*). Further investigations are required using all the samples and analysing also mitochondrial 16S rDNA.

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P023

Identification of copy number variants in Braque Français type Pyrénées' dog using CanineHD array

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Copy number variants (CNVs) are an important source of genetic variation complementary to single nucleotide polymorphisms (SNPs). Only a few studies have been conducted in dogs on CNVs derived from high-density SNP array data, and many canine breeds still remain uncharacterised, e.g. the Braque Français, type Pyrénées breed (BRA). Therefore, in an effort to more comprehensively investigate the canine genome for CNVs, we used a high-density genome-wide array (170 K) to discover additional CNVs in BRA. A total of 48 BRA individuals (27 females, 21 males) were sampled. After excluding SNPs which were unmapped or mapped to sex chromosomes, a total of 167,183 markers were used. A total of 1047 CNVs were detected using PennCNV, with an average length of 107.123 kb and an average number of 40.3 CNVs per sample. The CNAM univariate segmentation of SVS identified 1638 CNVs with an average length of 110.41 kb and the average number of 63 CNVs per sample. By aggregating the overlapping CNVs from PennCNV, a total of 181 CNVRs were identified. The CNVs identified with SVS were aggregated into 280 CNVRs. Intersecting the two CNVR datasets, a total of 45 CNVRs, ranging from 3.5 kb to 458,716 kb in length were detected in 26 dog samples. Among the stringent CNVRs, 42 CNVRs were defined as pure deletions, and only 3 as loss/gain, meaning that both deletions and duplications were observed. Results overlap moderately in comparison with previous studies on CNVs in dogs, leading to the identification of 16 novel CNVRs. A total of 159 genes were annotated in the CNVRs. Some of these genes are associated with well-known phenotypes in dogs, such as: *SOX8* involved in sex determination, *SLC38A2* related to hypoxia adaptation, *MYOG* associated with the development of functional skeletal muscle. Moreover, the gene ontology enrichment analysis provided information on biological processes and cellular components related with muscle structure development and muscle cell differentiation. These are interesting results considering that BRA is a dog breed used for tracking, hunting, pointing and retrieving feathered game. These are hardy dogs, strong,

adequately muscled but without heaviness. We can hypothesise that selection for such hunting behaviour, for which particular anatomical features are required, could have shaped, at least in part, the genetic background of this breed and, consequently, the frequency/presence of the detected CNVRs in these genes.

P024

Genome-wide analysis identifies a new potential candidate marker associated with the coat colour sidedness in Cinisara cattle breed

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Coat colour is one of the most important phenotypic features in livestock breeds. Cinisara is a local cattle breed generally of uniform black colour which occasionally presents a particular phenotype, with animals typically displaying a white band along their spine, from the head to the tail, and on the ventral line (colour sidedness). Therefore, this breed provides an ideal model to study the genetic components underlying phenotypic variation in coat colour. In order to identify the potential causative markers affecting the phenotypic variability, we compared the two groups of Cinisara via a case-control genome-wide association study (GWAS) and a genome-wide F_{ST} analysis. A total of 63 animals, ten with sidedness phenotype and 53 with uniform black colour were genotyped with Illumina Bovine 50 K. After filtering for quality, the final number of markers retained for the analysis was 45,246. In the GWAS, at the $p<.05$ Bonferroni corrected, we identified a single strongly associated marker. An average inflation factor (λ) of 1.08 indicated that the GWAS was not inflated by population structure. To further support the association, a genome-wide F_{ST} case-control analysis was also performed. The marker with the highest F_{ST} value (F_{ST}=0.559) that differentiated between the groups overlapped with the significant SNP identify in the GWAS. Therefore, the comparison among GWAS and F_{ST} analysis revealed a single nucleotide polymorphism (SNP), *ARS-BFGL-NGS-55928*, at the position 21,048,672 bp on bovine chromosome (BTA) 20, significantly associated with the trait. Only one gene (*PLK2*) was annotated near the associated SNP in a window of ±200 kb. The protein encoded by this gene is a member of the polo-like kinase, the same family of several known coat-colour candidate genes. Based on the reported results, we draw the possible conclusion that the identified marker is potentially associated with the coat colour sidedness in Cinisara. Once again, the local breeds with their genetic variability represent an important resource and model to study the genetic basis affecting peculiar traits. Moreover, these results should be of value for future studies, and constitute a preliminary report to further genomic research on coat colour sidedness. Future studies would be particularly relevant to refine these results and to better understand the genetic basis for this phenotype.

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P025

Genome-wide characterisation of runs of homozygosity and estimation of genomic inbreeding in Sicilian goat breeds

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The availability of single nucleotide polymorphism (SNP) assays allows for the determination of autozygous segments based on runs of consecutive homozygous genotypes (ROH). ROH are widely used as predictors of whole-genome inbreeding levels in livestock species. In this study, we computed several ROH parameters to investigate different scenarios of contemporary goat breeding in five Sicilian breeds. Individuals of Argentata dell'Etna (ARG, $n = 48$), Derivata di Siria (DDS, $n = 32$), Girgentana (GIR, $n = 59$), Maltese (MAL, $n = 16$) and Messinese (MES, $n = 22$) were genotyped with the Illumina Goat SNP50 BeadChip. After filtering, the final number of animals and SNPs retained for analyses were 174 and 48,348, respectively. A total of 3687 ROH segments >2 Mb were detected. The ROH parameters revealed well-defined differences between breeds. The mean number of ROH per breed ranged from 3.02 (ARG) to 38.81 (MAL). The average length of ROH ranged from 4.98 Mb (ARG) to 9.61 Mb (DDS). MAL breed showed the highest value of inbreeding (F_{ROH}=0.125), followed by GIR (F_{ROH}=0.108), whereas ARG showed the lowest one (F_{ROH}=0.009). ARG also showed the highest number of samples for which no ROH were detected ($n = 7$). Each ROH segment was categorised based on its physical length in five categories, and the mean sum of ROH per breed was calculated. The results showed that, for all breeds, the majority of ROH segments were <8 Mb in length. DDS and MAL had a larger mean portion of their genome (98.97 Mb and 89.12 Mb, respectively) covered in longer ROH (>20 Mb). High ROH coverage within the short category

may indicate a relatively high contribution of more distant inbreeding in the breeds, whereas the higher coverage of long ROH suggests a larger effect of more recent inbreeding. Therefore, our results highlight that ancient and recent inbreeding have had an impact on the genome of the Sicilian goat breeds and suggest that several animals have experienced recent autozygosity events. In particular, ARG and MES appeared as the less inbred breeds, whereas MAL, GIR and DDS showed ROH patterns typically produced by recent inbreeding. The levels of ROH that we estimated here reflect the inbreeding history of the investigated goat breeds. Our results showed the need of implementing conservation programmes to control the rise of inbreeding, especially for breeds with a reduced population size, such as the GIR.

P026

GDF9 and *BMP15* genes sequence analyses in Mediterranean Italian River Buffalo

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Oocyte recovery rate is a main issue in genetic improvement since it allows to increase the number of embryos obtained from the same dam with artificial reproductive technologies thus facilitating genetic selection also through the maternal line.
The number of oocytes recovered is highly correlated with the amount of follicles recruited per follicle wave, an event regulated by various genes. In particular, *GDF9* and *BMP15* act synergistically through the regulation of several key granulosa cell enzymes that are essential for normal ovulation, fertilisation, and female reproduction. Single nucleotide polymorphisms (SNPs) in both genes have been related to an increased number of mature oocytes per cycle and to superovulation traits in different species.
The aim of this project is to look for SNPs that may be related to oocyte recovery rate from ovaries of Mediterranean Italian River buffaloes. To the purpose, we first analysed the genes *GDF9* and *BMP15*. In particular, the two exons and the intron of *GDF9* and the two exons of *BMP15* have been fully sequenced in the animals selected for this study.
GDF9 has been sequenced in 50 buffaloes: 47 females with different oocyte recovery rates, 2 hypofertile females and 1 healthy bull.
BMP15 has been sequenced in 30 buffaloes: 27 females with different oocyte recovery rates, 2 hypofertile females and 1 healthy bull.
All buffaloes were unrelated and reared in different farms of the Campania Region.
None of the genotyped animals showed SNPs in the analysed sequences of *GDF9* and *BMP15* suggesting that in Mediterranean Italian River Buffalo other genes may have a main role in oocyte recovery.

P027

Molecular traceability of food products obtained from a local goat population in Umbria region

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The safeguard of local genetic resources is important to avoid genetic erosion and to preserve their cultural and historical value. Among the different tools for breed traceability, DNA-based methods are widely recognised as the most powerful, because they can be applied at any stage of the production chain. So far, breed allocation based on molecular markers has been profitably applied for meat traceability in different animal species, including cattle and pigs, while in sheep and goat it is still difficult to implement for the high cost of genotyping relative to the economic value of the single animal, especially in minor breeds.
This work aims to develop simple screening procedures for the implementation of molecular traceability protocols along with the market and food chain for a typical genetic resource of the Umbria region: 'Capra della Valnerina'. This genetic type, also known by names that refer to the characteristic two white facial lists, is a local population reared primarily in Valnerina. For this study, blood samples were collected from 24 animals belonging to 9 different farms; 20 samples of two cosmopolitan breeds (Camosciata delle Alpi and Saanen) were included as controls. A panel of 16 SSR markers (selected from the list of recommended markers for genotyping analyses in goat breeds – FAO/ISAG) was used with the objective to select a minimum number of markers capable to reliably identify the local population from cosmopolitan ones. A Discriminant Analysis of Principal Component (DAPC) was carried out with the method implemented in the ADEGENET package within the statistical package R version 3.3.2, using 16, 10, and 7 microsatellite markers. DAPC was conducted without *a posteriori* group assignments by inferring the most likely number of genetic clusters (K) using the *find.clusters* function. The scatterplot of the first two components of the Discriminant Analysis showed that the local goat is clearly distinguishable from the cosmopolitan breeds using a minimum of 7 markers.
On the base of our results, actions directed to preserving genetic distinctiveness of 'Capra della Valnerina' can be undertaken

using the molecular markers identified in this work; simple, low-cost molecular tests for traceability can be easily implemented, by using a very small SSR panel.

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P028

Beta-casein A2 variant: is the frequency changing in Holstein cattle?

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Since the detection of bovine β-lactoglobulin genetic variants in 1957, genetic variation has been identified and characterised in all milk proteins. The effects of several variants as well as of casein cluster haplotypes on production traits have also been investigated, but their use in selection schemes was rarely exploited. In the late 1990s, after epidemiological evidence in New Zealand indicated that beta-casein CSN2*A1 milk consumption incremented mortality from ischaemic heart disease, a company, A2 Corporation, started marketing milk produced only by CSN2*A2A2 individuals. Bioactive peptide with opioid properties β-casomorphin-7 (BCM7), one of the several peptides released during milk protein digestion, was suspected to be the risk factor in this human disease. Furthermore, BCM7 has been suggested to be involved in sudden infant death syndrome, neurological disorders (e.g. autism and schizophrenia) and milk allergy. CSN2*A2 and CSN2*A3 differ from CSN2*A1, CSN2*B and CSN2*C for the substitution of a His with a Pro at position 67 in the mature protein. The presence of His67 determines the enzymatic cleavage, which releases BCM7 in the three variants. An European Food Safety Authority report (EFSA 2009) could not establish a cause-effect relationship BCM7 or related peptides and the aetiology of the abovementioned diseases. More recent publications pointed at possible intolerances and gastrointestinal effects of BCM7, associating CSN2*A1A1 milk consumption to delayed intestinal transit, looser stool consistency, and intestinal inflammation, and even if the number of studies is still limited, CSN2*A2A2 milk is now marketed in various countries. We analysed data from Illumina beadchip of 214210 (including directly genotyped and imputed SNPs) Holstein cattle bred in different countries born between 1952 and 2017 and found that the CSN2*A2 frequency remained substantially unchanged in the 1980s (44.75%) and in the 1990s (45.78%), but it increased substantially in the 2000s (50.46%) and it is still on the rise (53.94% in the 2010s and 54.21% in this decade). A similar trend was also observed in the 51871 Italian Holsteins analysed: 38.92% in the 1990s, 49.03% in the 2000s, 53.07% in the 2010s and 55.72% in individuals born since 2010. These data indicate an ongoing selection of bulls and cows carrying only the CSN2*A2 variant, both in Italy and worldwide.

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P029

Phenotypic traits correlation for hygienic behaviour in *Apis mellifera ligustica*

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Beekeeping is a farming activity with huge economic importance beneficial effects on rural development and ecological balance. Thanks to national beekeeping register, a total of over 50,000 beekeepers and over 1.1 millions of hives were recorded in 2017; it was estimated that the beekeeping sector is worth over 2 billion euros. However currently, more and more bees are dying, because of the extensive pesticides use and numerous diseases.

Honey bee (*Apis mellifera*) hygienic behaviour is performed by single bees in a colony and it is a specific mechanism of resistance against a number of important pathogens, including *Ascosphaera apis* (which induces chalkbrood disease), *Paenibacillus larvae* (which causes American Foulbrood) and the parasitic mite *Varroa destructor*. The hygienic activity usually involves uncapping behaviour of the diseased or damaged brood cells and removing behaviour that involves the removal of the pupae from the damaged cells.

The aim of the study was to characterise and pick out *Apis mellifera* colonies according to their hygienic behaviour first and then to their propolis and honey productions, and to their grooming behaviour. We evaluated a total of 50 colonies, among the most productive and strong ones on almost 900 colonies. Colony's strength was assessed by counting the number of frames with brood and frames covered by bees within the hives. The hygienic behaviour was carried out according to the liquid nitrogen-killed brood test (LNKB): a small portion of capped brood was frozen using liquid nitrogen and then returned to the colony. Initially,

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we selected frame to be tested, looking at frame with the best brood pattern. Twenty-four hours and forty-eight hours later the freeze-killed portion was checked to see how much dead brood the colony removed. The normal distribution of traits under investigation was verified by Shapiro–Wilk test. Pearson correlation was performed to explore the relationship among the different traits. All the statistical analyses were conducted in R environment. Overall hygienic behaviour highlighted suggestive trends with productive traits (i.e.: propolis yield). In particular, 18 colonies showed a relevant hygienic behaviour associated with a higher attitude in grooming behaviour. Further studies are needed to investigate the biological bases of our finding.

P030

Multivariate factor analysis of milk fatty acids profile for GWAS analysis in Comisana sheep breed

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Sheep milk is important for the economy of Mediterranean countries. A large part of its production is reserved for traditional cheese making, usually protected by the European Community regulations. Milk fat content balance between MUFA and PUFA is important for cheese flavour and, although it is well known that fat is significantly influenced by both genetic and environmental factors, there is no relevant literature reporting genetic or genomic parameters influencing it in sheep populations. Nowadays, genome-wide association (GWA) studies represent the gold standard method to investigate the genetic architecture of complex traits, such as milk fat composition. However, because of the nature of this multiple-trait, performing an effective association analysis is not an easy task. In fact, when correlated traits are analysed, because of GWAS stringent cut-off, the outcomes may lead to false negatives and broadly are difficult to interpret. Methods that generate uncorrelated traits represent useful and promising approaches to overcome this problem. In this study, a multivariate factor analysis (MFA) was performed to extract latent factors able to explain metabolic information of the original variables and usable in a GWA analysis. To achieve this, we first characterised milk samples from 200 Comisana ewes, reared in genetic Centre of Asciano for their fat profile by gas-chromatography. Subjects were genotyped with SNP array and a single-marker regression model for GWAS was performed. The extracted factors were representative of the following groups: *de novo* fatty acids, biohydrogenation, branched fatty acids and desaturation. Moreover, GWA analysis showed a general increase of significances compared with the analysis using the original variables. Nevertheless, this was not sufficient to reach the Bonferroni statistical threshold, except for a single SNP on Chr 6 in association with desaturation factor group. To the best of our knowledge, this was the first study performing a MFA on milk FA in Comisana. The preliminary results were interesting and in line with previously published studies on bovine.

P031

Association between transposable elements activity and genetic diversity in domesticated ungulates

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Transposable elements (TEs) are short DNA sequences that can move and replicate within the genome, modifying genes structure and expression, and generating variability. According to their implication in shaping the genomic structure, some authors recently proposed TEs activity as one of the drivers of genome evolution, being a modulator of differentiation and a carrier of speciation. Many studies showed that transposable elements have a substantial role in the evolution and speciation of mammals especially during events of adaptive radiation. In this context, the aim of this study is to investigate the potential role of transposable elements on shaping genomes during the domestication of cattle using the available genomes of *Bos taurus* and *Vicugna pacos* (Camelidae) as outgroup. To test this hypothesis, we used the Spearman correlation and a linear regression model based on the association between the number of active transposable families, the Density of Insertions (DI) and the Relative Rate of Speciation (RSS). DI summarises the TE activity level as a ratio between the number of TE insertions in a genome and its size, whereas RSS is defined as a conditional parameter used to compare a pair of taxonomical element at the same hierarchical level. According to our previous work, we found that the fast evolution and speciation of Bovidae respect to Camelidae correlated with the higher activity and accumulation of transposable elements in the former family that might have contributed to the evolution of novel phenotypes. Furthermore, here we show for the first time the comparison between the DI distribution of TEs in *Bos taurus* vs. *Bos taurus indicus* (zebù). In conclusion, this work shows that the activity of transposable elements contained in Bovidae genomes explained the variability and speciation of this taxon and provide helpful information about the activity of new TE families of *Bos taurus indicus*.

P032

Cabannina: genomic characterisation of a local Italian Breed

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Cabannina is a small dual-purpose cattle breed, reared almost exclusively in Liguria. Other local cattle breeds in the area might have experienced a genetic introgression with Cabannina. Moreover, a 1938 law stipulated the use of Bruna Italiana bulls to improve the productivity of local breed population. As a result, the Cabannina breed may have interbred with Bruna Italiana and/or other local breeds. This work aimed to disentangle the genomic background of Cabannina with respect to local/cosmopolitan breeds and to identify the effects of recent selection in this breed. Consequently, a subset of 170 individuals, belonging to 7 Italian cattle breeds and genotyped with BovineSNP50 BeadChip within the BOVITA project were used. As the first step, after an initial quality check that reduced the number of SNPs to 36,202, phylogenetic analysis was performed in order to determine the genetic background of Cabannina. Our results highlighted the uniqueness of the Cabannina breed, which is clearly distinguishable from the other breeds in MDS plots. Furthermore, Reynolds distances indicated the genetic proximity of Cabannina to Bruna Italiana. Haplotype analysis revealed that Cabannina shares the longest median with Bruna Italiana (5.13 Mb) and that 73% (of the cows) of these two breeds display haplotypes in common.

Subsequently, we investigated the signatures of recent selection on the genome of Cabannina breed respect to Bruna Italiana breed, using Integrated Haplotype Score and the Wright's Fixation Index (F_{ST}). The results from these two approaches enabled us to identify 3 different genomic regions, harbouring 3 genes (*SLC8A1*, *ARNT2*, *PRKCB*) that can be considered under selection in Cabannina but not in Bruna Italiana.

These genes are associated with QTLs related to milk production, fertility and morphological traits connected to grazing in mountainous and hilly areas.

In conclusion, this study allowed us to determine the genomic uniqueness of the Cabannina breed, despite its eventful history, which differentiates it from the other bovine populations studied.

Finally, the genomic regions we identified are linked to selection in response to Cabannina's adaptation to the local environment.

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P033

Metagenomic analysis of water bacteria communities in South Adriatic Sea (Kornati islands, Croatia)

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Isolation of environmental DNA (eDNA) is a new and promising method for analysing bacteria and aquatic species. In the present research, a variety of bacteria communities were analysed in the South Adriatic Sea (Kornati islands, Croatia) in three locations (Kurba, Klobucar, Silo Vela) and at two sea depths (5 and 20 m). The water was collected and filtered using a peristaltic pump and 0.22 µm, 50 mm diameter, cellulose filters (Millipore, USA). After filtration, filters were stored in liquid nitrogen for six days, and subsequently at –80 °C. eDNA was extracted from the filters using the PowerWater DNA Isolation kit (MoBio Laboratories Inc., Carlsbad, CA). DNA extracted from filters was sequenced by means of a Illumina Sequencer (HiSeq 2500), using a 16S rRNA profiling method. All trimmed and filtered data were processed using a custom script based on the QIIME software suite. All reads were classified to the lowest possible taxonomic rank using a reference dataset from the SILVA database. Biodiversity of the samples (alpha-diversity) was calculated with Chao1 and Shannon indexes. Similarities between samples (beta-diversity) were calculated by unweighted uniFrac. Overall, results showed the presence of a large variety of bacteria among four phyla. The most abundant phylum was the *Proteobacteria*, followed by *Actinobacteria*, *Bacteroidetes* and *Firmicutes*. Phyla *Proteobacteria* and *Bacteroidetes* were the most present at the Kurba location while phylum *Actinobacteria* was present at the Velo Silo. Phylum *Firmicutes* was more present at Klobucar. Regarding depths, phylum *Proteobacteria* showed the same abundance at the two depths while phylum *Firmicutes* showed a major presence at 5 m and phylum *Actinobacteria* at 20 m. The study identified the presence of some potentially pathogenic bacteria for marine animal organisms and also for humans, such as: *Staphylococcus sp.*, *Legionella sp.*, *Heliobacter sp.* and *Pseudomonas sp*. This study has clearly shown that diverse bacteria populations can be easily and rapidly detected by filtering the water and analysing by means of using NGS technologies.

P034

Utilization of progesterone-based treatments on anoestrus in Mediterranean buffalo

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The aim of this study was to evaluate the efficacy of a treatment based on progesterone for encountering the anoestrus in buffalo. The trial was carried out on 124 Italian Mediterranean buffaloes (91 primiparous and 33 pluriparous) bred in a commercial farm in Caserta province for three years. All buffaloes that were recognised in anoestrus after clinical and ultrasound examination, underwent a progesterone-based treatment. Briefly, it consists in an intravaginal progesterone-releasing device (PRID DELTA®, Ceva Animal Health, Agrate Brianza, MB, Italy) for 10 days and 750 U of PMSG (Folligon®, Intervet, Milan, Italy) and 500 mcg of PGF2ρ analogue (Cloprostenol, Estrumate®, MSD Animal Health, Segrate Milan, Italy) administered at removal. All buffaloes were maintained in open yards with bulls of proven fertility and underwent clinical examination 15 days apart. After pregnancy diagnosis or the subsequent calving, the intercalving period (ICP) and the treatment – conception interval (ITC) were calculated. Statistical analysis of the data was carried out by chi-square test and Student's *t*-test.

The pregnancy was ascertained in 60.0% (73/124) of buffaloes on the progesterone-induced heat or in the subsequent oestrus cycle, whereas the remaining animals resulted pregnant after the third cycle (p<.01). Pregnancy rate recorded in pluriparous buffaloes in the first two oestrus cycles after treatment was significantly (p<.01) higher than that recorded subsequently (87.8 vs. 12.2%, pregnant at the first and second oestrus cycle post-treatment vs. those pregnant subsequently, respectively). No significant differences were recorded in primiparous buffaloes (48.4 vs. 52.6%, pregnant at the first and second oestrus cycle post-treatment vs. those pregnant subsequently, respectively). A higher ITC was recorded in primiparous compared with pluriparous buffaloes (79.7 ± 77.0 vs. 24.5 ± 20.5 in primiparous and pluriparous buffaloes, respectively), and consequently a higher ICP (509.4 ± 86.9 vs. 486.5 ± 119.1 in primiparous and pluriparous buffaloes, respectively). No differences were observed regarding the month of treatment. In conclusion, the application of progesterone-based treatment, allows cycle resumption in almost all acyclic animals, guaranteeing 50% pregnancy rate, independently of the period of the year.

P035

Effects of feed supplementation with galactogogue and immunomodulator phytocomplexes on chromosome stability in Cilentana goat: preliminary results

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Nowadays, the demand from consumers of nutraceutical and organic products is constantly growing and the use of supplements in the diet of livestock is becoming a common practice. In particular, the administration of galactogogues (natural substances that stimulate milk production) and of phytocomplexed immunomodulators are thought to have a synergistic effect aimed to increase milk productions and animal health. This study aims to analyse the effects of two different feed supplements on lymphocytes chromosome stability as an index of immune system status. Fifteen Cilentana goats were divided into 3 groups (G1, G2 and G3 or control group). All goats were second parity females and received the same ration, except for the supplementation administered to G1 and G2 which received two different formulations of supplements. Supplement is composed of some officinal plants, omega3 unsaturated fatty acids and bioflavonoids (formulation subjected to copyright) with an action to improve milk production (increase in protein and unsaturated fatty acids) and immunomodulatory activity. G1 received 20 gr/die supplementation in two critical periods for the production cycle of the animal, from 14 February to 15 March and from 1 July to 1 August. G2 received from 15 March to 31 August a complementary feed (20 gr/die) enriched with vegetable oils and fats extracted from Meliaceae with immunomodulatory and antioxidant action. Two blood samples were taken at the beginning and at the end of experimentation. CA and aneuploidy test were performed, respectively, on 100 and 50 metaphases as previously reported by Ciotola et al. (2014). For each animal were performed plates for each animal were observed for aneuploidy and CA (chromatid and chromosome breaks) tests, respectively, under a fluorescence microscope, captured with a digital camera Nikon DS U1, transferred onto a PC and later processed by image analysis software. Aneuploidy was calculated as the percentage of cells with $2n \neq 60$. The analysis of the currently available data shows a different response in the G1 and G2 groups compared to the G3, which suggests a positive effect on lymphocytes chromosome stability.

P036

Genetics of somatic cells in autochthonous dual-purpose breeds

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Somatic cell count (SCC) in milk is considered an indirect measure of udder health being linked in most cases to mastitis. Mastitis has a large economic impact on dairy profitability and in many specialised dairy cattle, the log transformation of SCC (i.e. Somatic Cell Score; SCS) has been introduced in breeding programmes. This trait has gained interest even in autochthonous breeds. The aim of this study was to estimate heritability (h2) and genetic correlations (rg) between SCS and milk (M), fat (F), protein (P) yields in Alpine Grey (AG), Reggiana (REG) and Rendena (REN) local breeds. Test day for M, F, P and SCC (from 1997–2018) were obtained from national functional control carried out routinely by the Italian breeders association. After editing, test-day records for the first three lactations available for analyses in each breed were 406.918 for AG, 78.351 for REG, and 252.229 for REN, and the corresponding numbers of cows were 29.219, 6.149 and 15.716. A unique repeatability test day model was used. The linear model includes the fixed effects of herd test day (HTD), parity number, (separately from HTD for AG, combined with HTD in the other breeds), gestation, age at parity within lactation, month of parity within lactation. The shape of the lactation curve was described by 4th order Legendre polynomials for the fixed effects of parity number, age at parity and month of parity. Random permanent environment and additive genetic effects were also accounted in the model. The number of animals included in the pedigree files were 49.389 for AG, 8.506 for REG and 22.412 for REN, respectively. Bi-trait analyses by means of a Gibbs sampling algorithm were carried out. The values of estimated h2 ranged from 0.08 to 0.13 for SCS (high posterior densities – HPD: 95% 0.05–0.15), 0.16–0.22 for M (HPD: 95% 0.12–0.24), 0.09–0.16 for F (HPD: 95% 0.07–0.17), and 0.12–0.18 Q27 for P (HPD: 95% 0.08–0.20). Genetic correlations of SCS with productive traits resulted generally low. Particularly, the rg ranged from 0.07 to 0.14 considering SCS and M, from −0.11 to 0.07 between SCS and F and from 0.09 to 0.12 between SCS and P Q28 yields. Considering all analysed traits, the HPD 95% of rg accounted always zero, indicating genetic independency of SCS from M, F and P. On the basis of results obtained, SCC could be introduced as a breeding goal in the local Alpine Grey, Reggiana and Rendena breeds without negative effects on yields.

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P037

Influence of food type on the reproductive performance of *Octopus vulgaris* kept in captivity

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The octopus (*Octopus vulgaris* Cuvier, 1797) has been indicated as a new species for diversification of aquaculture due to the short life cycle, the fast growth and the high feeding conversion rate. During the reproduction, it is known that female nutrition can affect the quality of deposition since diet provides essential nutrients for embryonal and paralarval development.

Six adult octopus couples were divided into two groups (3 couples for each group) and fed *ad libitum* with two different diets: GF (Group Fish) octopus were fed with 90% of fish and 10% of crustacean, while GC (Group Crustacean) octopus were fed with 90% of crustacean and 10% of fish. Animals were kept in a recirculating aquaculture system (RAS) for 22 weeks and subjected to eco-physiological conditioning that included an increase in temperature (from 15 °C to 20 °C) and photoperiod (from 10 to 15 h of light) to obtain spontaneous spawning. Salinity was kept at 35 ± 2 ppt. During the trial, main reproductive performances were recorded and statistically analysed.

At the end of the trial, GC female showed a higher ($p \leq .05$) body weight increase compared to GF female; in particular, GC females had a body weight increase of 363%, while GF females displayed an increase of 270% compared to the initial weight of each group. Females fed with the highest percentage of crustacean (GC) produced more eggs (116.100 vs. 20.933) and paralarvae (115.367 vs. 19.763) ($p \leq .05$) due to the high number of the spawned

ovarian bunches. The length of bunches and the hatching percentage were not affected by the food administered. Likewise, paralarvae spawned by GC and GF octopus were not statistically different and had a high vitality that had been prolonged for different days in a fasting condition.
The results obtained showed that the growth and reproductive performances of octopus fed with natural diet depends on the species used as food, showing the great importance of crustacean in the diet of these cephalopods.

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P038

Genetic investigation of Rendena cattle biodiversity and evolutionary history

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Situated on the western side of the Province of Trento, the Rendena Valley was populated by Rhaetian and Celtic peoples from the bronze age, and later conquered by the Romans in the 1st century B.C. Settlements in this valley were relatively isolated for centuries, with a pastoral economy based on the rearing of the indigenous dark brown cattle. The modern Rendena cattle breed is the descendant of ancient Rendena Valley livestock and still plays an important commercial and cultural role in the local alpine agriculture. The project RENDENAGEN aims at reconstructing the genetic origin of this breed that, from preliminary analyses, appears rather unique. We will also attempt to connect the genetic diversity of the Rendena breed to the prehistoric history of the Rendena Valley inhabitants. We will collect genetic material from at least 200 Rendena dairy cows, representing all existing maternal lineages and fully sequence their mitochondrial DNA (mtDNA). A comparative analysis with public and proprietary mtDNA data from other breeds in Europe (e.g. Piedmontese, Angus) and Southwest Asia (e.g. Anatolian Black, Turkish Grey) will permit the reconstruction of the origin of the breed and past demographic events, as well as an assessment of its present diversity. Various stakeholders will benefit from this study, because the results may be used to estimate, protect and manage this agricultural and cultural heritage.

P039

Identification of a low-density SNP panel to evaluate hybridisation between feral and domestic sheep

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Increasing rates of hybridisation between many domestic and wild species have been documented in several livestock species as pig, goat and sheep. Hybridisation can be favoured by farming practices as free grazing and can lead to the loss of genetic variability in the wild species. The Mediterranean island of Sardinia hosts one of the last extant autochthonous European mouflon (*Ovis aries musimon*) populations. Conservation policies including reintroduction plans have been enforced to preserve Sardinian mouflon. However, although previous research described the overall genetic integrity of the Sardinian mouflon population, the occurrence of crossbreeding between wild and domestic sheep has been reported. Here, we identified a low-density SNP panel able to detect and quantify admixture between wild and domestic sheep hybrids. Medium-density SNP array genotyping data of 23 Sardinian mouflon and 23 Sarda sheep showing pure ancestry were used as a reference. After pruning for MAF, Hardy-Weinberg equilibrium, and call-rate, 33.481 SNPs were retained for further analyses. We applied a two-step selection algorithm. Principal component analysis was used as a pre-selection step and identified a set of ~1 k most significant genetic markers. Then, an iterative supervised-classification machine-learning approach (based on Random Forest) was implemented to generate two low-density panels of highly discriminant SNPs. The first panel of 131 SNPs was obtained after a single Random Forest run, while a second-panel accounting 51 SNPs was identified iterating the classification algorithm until no SNPs was further removed. To evaluate the efficacy of the two low-density panels to determine genetic admixture we tested (1) 28 mouflon × Sarda hybrids of known ancestry proportions, and (2) three synthetic mouflon × Sarda hybrids populations each counting 90 individuals, having 25, 50 and 75% sheep ancestry. We computed the ancestry proportions in the full-and low-density sets through supervised admixture. We obtained high correlation values for the 131 and

51 SNPs panels in both the natural and synthetic populations. The diagnostic SNP panels proposed in this work could be a useful tool to conservation practitioners to exclude hybrids from reintroduction plans, hence improving the implementation of biodiversity conservation strategies for mouflon populations.

P040

Molecular diversity of sheep breeds from the United Kingdom

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Sheep farming is the most important agricultural activity in the United Kingdom, with ~33 million sheep and 90 breeds, more than half of which are local breeds. The current changes in market demands and consequent plummeting of lamb meat price is causing a reduced interest of sheep breeders on the less profitable local breeds. Consequently, local breeds are experiencing a decrease in population size, loss of genetic diversity, and increase of inbreeding. With the aim to evaluate the conservation status of British local breeds, we investigated the genetic diversity and population structure of 14 local sheep breeds chosen among the most representative. Further, we included one commercial breed from UK and two from mainland Europe, all of them belonging to the Texel breed group. We analysed 515 individuals genotyped using the Ovine SNP50 BeadChip. We evaluated the molecular diversity through observed and expected heterozygosity, effective population size and linkage disequilibrium. Further, the population structure was investigated through Admixture, PCA and Neighbour-Net analyses. Our results highlighted medium- to high-levels of inbreeding and low effective population size for the majority of the local breeds analysed, such as for South Wales Welsh Mountain and Border Leicester breeds, and low values of observed heterozygosity. Conversely, few local breeds presented an increase of effective population size in the most recent generations (e.g. Soay), or limited inbreeding values, and moderately high *Ho* (e.g. Welsh Mountain). Such results are probably due to the increased awareness of authorities on biodiversity, and the consequent implementation of conservation policies for local breeds. Overall, despite a few positive exceptions, results underline that many local UK populations possess low levels of molecular diversity, along with unknown or partially incomplete history. High levels of inbreeding combined with decreasing effective population size, and low levels of heterozygosity may increase the degree of endangerment and the risk of extinction of many UK breeds, especially those having a reduced census size. In this context, our results underline the importance of monitoring genetic diversity across generations to design and implement educated conservation plans.

P041

Selection signatures in feral and domestic Sardinian sheep

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Since domestication, both artificial and natural selection have shaped the genomes of domestic species towards economically important traits and adaptation to the anthropogenic domain. Here, genome-wide single nucleotide polymorphism data of 27 Sardinian mouflon and ten Sarda sheep (a dairy breed) were investigated to identify selection signatures in co-distributed feral and domestic sheep in Sardinia using Cross Population Extended Haplotype Homozygosity (XP-EHH). A locus-specific empirical *p* value assessment approach was developed to define a confidence threshold for XP-EHH results and windows of selection were defined by including neighbouring positive results.
Despite differences in sample size in the two groups, of the 475 genomic selection signatures detected, 331 were exclusive to the Sarda, while 144 were exclusively found in the mouflon. The identified windows of selection harboured 261 and 18 annotated genes in sheep and mouflon, respectively. In sheep, genes of known agricultural value such as: milk and wool production, fat deposition and fertility were found. In contrast, genes known to be involved with body size traits that could be related to mating advantage, were identified in mouflon.
By investigating domestic and feral populations with genome-wide data and applying novel data filtering methods, we identified regions under strong selection and linked to the second wave of sheep domestication. The regions detected harbour physiologically relevant candidate genes in both feral and domestic sheep, providing a better understanding of the effects of domestication at the genome level.

P042

Canonical discriminant analysis to analyse goat breed diversity at alpha-s1-casein (*CSN1S1*) locus

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The goat AdaptMap project (http://www.goatadaptmap.org/) is an international effort developed to improve coordination among otherwise independent projects for genotyping, resequencing and phenotyping of goat breeds. Several studies detected signatures of selection across the different breeds, sub-geographical clusters, phenotypic and climatic groups. Considering contrasting phenotypes between breeds, goat caseins have raised considerable research interest because in relation to milk quality, composition and technological properties. The canonical discriminant analysis (CDA) was used to explain goat breeds diversity by using the αs1-casein (*CSN1S1*) locus alleles information. The known *CSN1S1* alleles are grouped into 4 classes on the basis of the αs1-CN milk content: strong, intermediate, weak, null. For the 144 genotyped (Illumina GoatSNP50 BeadChip) breeds included in the AdaptMap dataset, we mined literature to identify the ones with known *CSN1S1* alleles. A total of 1123 animals belonging to 17 breeds were grouped *a priori* according to the different reported *CSN1S1* alleles classes. The CDA analysis was performed around the regions containing the casein cluster by using 791 markers spanning 75–120 Mb on Chr 6. The canonical variables extracted, were able to discriminate animals according to the *CSN1S1* alleles classes. In particular, the CAN1 (60% of variability) discriminated the strong alleles, CAN2 (22% of variability) discriminated the weak alleles and CAN3 separated the animals carrying the null alleles. The most discriminant SNPs for the three CANs have been found to be located in the casein cluster. In particular two SNPs mapped in the *CSN1S1* chromosomal position and the breeds carrying the null alleles shown opposite allelic frequencies in relation to the other breeds as results of selective selection pressure. Mahalanobis distance, based on the group centroid position in the three-dimensional space, returns a trend of variation from the strong to null alleles classes. Moreover, the CDA analysis allowed to identify associations of *CSN1S1* alleles with other clustered casein genes. This result could help in the developing of a panel of SNPs useful for selection plans aimed to improve milk technological properties for cheese making or, conversely, to modify milk nutraceutical characteristics for goat's milk intolerance.

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P043

Re-sequencing of genes related to mastitis resistance in dairy cows

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Selection for mastitis resistance should be one of the first aims in dairy farms, since mastitis is a major cause of economic loss. Resistance to mastitis is a complex trait and expression profiles of mammary gland infected with different pathogens were conducted for a better understanding of the mechanism underlying this disease. Many different genes were found to be involved with mastitis, but only the identification of causative mutations could be useful for selection of resistant cows. Wide regions of six genes involved in immune response were re-sequenced to look for causative mutation of mastitis resistance: the pentraxin3 (*PTX3*), the chemokine C-X-C motif receptors (*CXCR1* and *CXCR2*), the toll-like receptor 4 (*TLR4*), the mannose-binding lectin 1 (*MBL1*), and serum amyloid A3 (*SAA3*) genes, respectively on BTA1, BTA2, BTA8, BTA28 and BTA29. DNA was extracted from semen of bulls in the positive (58 bulls) and negative (37 bulls) tails of the distributions of estimated breeding values for somatic cell score. Using a target re-sequencing approach by NGS technique on the MiSeq Illumina platform, we identified a total of 1535 polymorphisms (including SNPs and small indels). Excluding mutations, with a minor allele frequency lower than 0.05 only 384 polymorphisms remained. The original phenotypes were adjusted for population structure using the genomic relationship matrix calculated using this dataset and 4 individuals having an identity by state (IBS) > 0.95 were excluded from the following analysis. To test for associations, mutations with a correlation higher than 0.80 with any others were further excluded, together with polymorphisms deviating from Hardy-Weinberg equilibrium. Finally, 101 polymorphisms were tested for associations. A total of 7 SNPs resulted significantly associated with SCS ($p<=.05$): one on PTX3 (rs208223246, missense variant responsible for the amino acid exchange Glu347Lys), one on *CXCR1* (rs109694601, intron variant), one on *TLR4* (rs134052737, intergenic variant), one on *MBL1* (rs208247354 and rs208491630, respectively intron and upstream gene variant), and two on *SAA3* (rs137746604 and rs210417381, both upstream gene variants). These findings

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represent the first step toward the use of causative mutations in genetic selection for mastitis resistance in dairy cows.

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P044

Genetic diversity, productive and reproductive performance in Italian chicken breed Bianca di Saluzzo

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Bianca di Saluzzo (BS) is a chicken breed reared in Piedmont region and its substitution with commercial lines caused a reduction in size, associated with a progressive decline due to inbreeding. In this study, genetic diversity, productive and reproductive performance were examined. Birds were kept in standard environmental conditions. At hatching, 177 chicks were weighed and at six weeks of age were separated by sex and transferred to growing pens with free-access to water and were fed with a standard commercial starter diet *ad libitum* followed by a growing diet. All birds were genotyped by a set of 14 microsatellite markers chosen by their high polymorphism. Body weight (BW) was recorded every two weeks from hatching to the age of 27 weeks. Gompertz model was used to define the growth rate. The performance of laying hens was monitored over a period of 6 months. To evaluate fertility and hatchability, all eggs laid in 2 consecutive weeks were hatched for 12 weeks. Data analysis was carried out with the Excel software. Allelic richness was 3.6 and observed and expected heterozygosity were 0.68. Mean live weights for cocks and hens were 2779 ± 233g ($n = 79$) and 2089 ± 195 ($n = 98$). Growth rates in the linear phase were 26.95 ± 3.19 g/day and 16.63 ± 1.90 g/day for male and female birds. In egg collection period for hatch mean 179.5 eggs were taken. Fertility and hatchability were 87.2% and 92.3%. The overall mean chick survival rate to six months of age was 4.6%. The age at first lay ranged between 5 and 6 months, and egg number/month was 14.5, mean egg weight was 60 ± 3 g and laying percentage resulted in 51.6% (max: 76.2%). These results provide an important insight on the genetic and productive characteristic of this local poultry breed that can be used for managing new mating schemes aimed to preserve variability and increase productivity.

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P045

Genetic diversity, productive and reproductive performance in Italian chicken breed Bionda Piemontese

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Bionda Piemontese (BP) is a local slow-growing poultry breed that is mainly reared for meat; hens are also bred for egg production. In this study, the genetic diversity, productive and reproductive performance were examined. The birds were kept in standard environmental conditions at the Poultry Conservation Centre of the University of Turin (Italy). At hatching, 218 chicks were weighed, labelled with a wing metal tag. At six weeks of age, birds were separated by sex and transferred to growing pens with access to an external paddock (20 birds/pen). The birds always had free access to water and were fed with standard commercial starter diet *ad libitum* (0–6 weeks) followed by a growing diet. All birds were genotyped by a set of 14 microsatellite markers selected for their variability. The body weight (BW) was recorded individually every two weeks from hatching to the age of 27 weeks, for a total of 14 weightings. Gompertz model was used to define the growth rate. The performance of laying hens was monitored Q29 over a period of 6 months. To evaluate fertility and hatchability, all eggs laid in 2 consecutive weeks were hatched for 12 weeks. Data analyses were carried out with the Excel software.

The results showed a low genetic variability with an allelic richness of 3.3 but a good individual variability: the observed and expected heterozygosity were 0.675 ± 0.040 and 0.680 ± 0.015, respectively, Fis index was 0.016. The growth performance was good: the mean live weights for cocks was 2797 ± 253 g ($n = 102$) and for hens 2226 ± 249 g ($n = 116$). The growth rates in the linear phase resulted 26.47 ± 3.19 g/day and 18.42 ± 2.63 g/day for male and female. The fertility and hatchability were 86.5% and 94.2%, respectively showing a good reproductive performance. The mean chick survival rate to six months of age was 4.6%. In egg collection period for hatch mean 184.5 eggs were taken; in total 1107 eggs were collected (6 repetitions). The egg production was lower than commercial layer but consistent with local breed: the age at first lay ranged between 5 and 6 months and Iegg number/month was 14.5, egg weight mean 61 ± 3g and laying percentage 51.6% (max: 70.7%). In conclusion, the results highlight that BS is local slow-growing and dual-purpose breed with good productive performance.

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P046

Dynamic profile of active metabolic pathways in the subcutaneous fat tissue of Holstein cows during early lactation

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Early lactation is a challenging time for dairy cows, which have to simultaneously cope with milk production and body maintenance. The rapid increase in energy requirements is only partially met by feed and cows enter in a state of negative energy balance (NEB), frequently associated with metabolic diseases and reduced fertility. To counterbalance NEB cows mobilise body reserves (mainly fat tissue) with a complex interplay still far to be completely understood. RNA-Seq experiments provide a comprehensive understanding of the expression of tissue-specific genes as well as of targeted metabolic pathways. For a better understanding of how lipids are metabolised to respond to cows requirements in early lactation we performed RNA-Seq analysis of the subcutaneous fat tissue sampled from the tail of 7 healthy multiparous Holstein Friesian cows at three times point after calving: T0 = 2 days, T1 = 30 days and T2 = 90 days. Adipose tissue was collected and preserved in Qiagen AllPrep solution until extraction, performed using the Qiagen RNeasy Lipid Tissue Kit. RNA-Seq was performed on the Illumina Hiseq 3000 platform. Differential expression analysis was performed comparing the log-fold differences in gene counts at the three-time points and genes with a FDR-adjusted p value <.05 were considered significantly different and retained for gene functional analysis. An enrichment analysis was conducted to detect pathways significantly associated with the identified genes. We found 113, 324 and 17 genes differentially expressed in T0 vs. T1, T0 vs T2 and T1 vs. T2, respectively. These genes are involved in 142 metabolic pathways. The top 5 pathways for time comparisons include insulin secretion, oxytocin signalling, glycolysis/gluconeogenesis, pyruvate metabolism, insulin resistance, calcium signalling, gonadotropin-releasing hormone, mitogen-activated protein kinase and adipocytokine signalling, and the renin-angiotensin system. These pathways are associated with cellular processes, inflammatory response and energy production, which contribute to milk synthesis, foetal growth and homeostatic mechanisms. This knowledge could be potentially applied to provide better farming conditions reducing the negative impact on the health and economics of the herds.

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P047

Several hundred single nucleotide polymorphisms in candidate genes: association with six production, carcase, and meat quality traits in Italian Large White pigs

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Italian heavy pig selection programmes include production (average daily gain, ADG; feed gain ratio, FGR), carcase and meat quality traits (weight of LC: lean cuts; HW: ham weight; BFT: backfat thickness; HWLFS: ham weight loss at first salting; and VIF: visible intermuscular fat). In this study, we wanted to evaluate if a selection genotyping approach designed for BFT could identify markers associated with other traits. Italian Large White (ITLW) gilts were chosen according to their extreme estimated breeding values (EBV) for BFT (276 with the most negative and

279 with the most positive EBVs) within a population of about 12,000 sib-tested pigs. A total of 736 single nucleotide polymorphisms (SNPs) within candidate genes were used in this study. Of the genotyped markers, 280 SNPs in 211 genes that had minor allele frequency >5% and call rate >90 were used in association analyses with six traits (ADG, FGR, LC, HW, HWLFS and VIF). To deal with the multiple testing problem in a candidate gene approach, we applied the proportion of false positives (PFP) method, setting a threshold for significance of 0.10. Six SNPs were significant: four for HWLFS and two for LC. The significant SNPs for HWLFS were within genes encoding proteins involved in carbohydrate metabolism (phosphoglucomutase 3, *PGM3*; and glycogen phosphorylase L, *PYGL*), transcription regulation and development (mitogen-activated protein kinase 8, *MAPK8*), and cellular RNA processing and degradation (exosome component 1; *EXOSC1*). The two significant markers for LC were within the TBC1 (tre-2/USP6, BUB2, cdc16) domain family, member 1 (*TBC1D1*) gene encoding a protein implicated in regulating the trafficking of glucose transporter 4. These results reveal a possible genetic basis for the relations among BFT, HWLFS and LC, beyond the trivial consideration that fatter hams necessarily lose less weight and complement results obtained in genome-wide association studies.

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P048

Heritability and correlations among milk yield, Body Condition Score and type traits in the Italian Mediterranean Buffaloes (*Bubalus bubalis*)

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The objective of the present study was to estimate variance components between milk yield and type traits, including Body Condition Score (BCS) in the Italian Mediterranean Buffaloes. Type evaluation in the Italian Mediterranean Buffaloes began in 2000 including thirty traits, namely seven composites, twenty-two linear and BCS. Composite traits included final score (FS), structure (ST), feet and legs (FL), dairy characters (DC), dairy strength (DS), udder (UT) and yield potential (YP). Linear traits included twelve, two and eight traits respectively, for structure, feet and legs and udder. In 2009, the first official genetic evaluation for type traits was introduced but it did not include milk yield and BCS as correlated traits.

The data analysed included 6199 buffalo cows and a pedigree file with 17,773 animals. A multi-trait animal model was fitted using a Bayesian implementation via Gibbs sampling. The fixed part of model included the following effects: herd-year of evaluation-classifier, days in milk (30 days class), age nested within parity and number of lactations. Animal and residual effects were treated as random. Traits analysed included five composite traits (FS, ST, FL, YP, UT), BCS, nine linear scores (stature, body depth, body length, Foot angle, Fore Udder Attachment, Rear Udder Width, Udder Depth, Teat Placement and Teat Length) and milk yield at 270 days.

Heritability estimates for the type traits varied from 0.09 to 0.43, the largest was for stature. Genetic correlation between type traits and milk yield ranged from –0.21 (Udder Depth) to 0.50 (Rear Udder Width). BCS had a null genetic correlation with milk yield at 270 days (–0.03). This result suggests that the possible inclusion of BCS in an aggregate index will not affect genetic response for milk yield. Rear Udder Width was positively correlated with both Fore Udder Attachment (0.75) and Teat placement (0.48) but negatively correlated with Udder Depth (–0.31) and Teat Length (–0.12). Fore Udder attachment had the same pattern. Teat length and Teat Placement had a mild negative correlation (–0.16). Excluding FL, foot angle and Teat Length, whose heritability was around 0.10, all the other traits showed moderate to high heritabilities suggesting reasonable and accurate response to selection. A multi-trait approach will help to cope with the expected low accuracy of some traits.

P049

Evaluation of the methylation extent in horses during training with MCSeEd, a new method for detecting epigenetic changes

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Cytosine methylation plays an important role in regulating the response to various stresses. Traditionally, two main approaches have been applied to unveil this phenomenon: digestion with methylation-sensitive restriction enzymes coupled with electrophoresis (i.e. MSAP – Methylation-sensitive amplification polymorphism) and bisulphite conversion followed by NGS (i.e. BS-seq). Both of these approaches have constrains. BS-seq

Q30 requires a reference genome for the alignment of converted and unconverted sequences. MSAP, while not requiring a genome, provides only a small fraction of the overall methylation. Recently, to overcome this limitation, some strategies involving methylation-sensitive restriction enzymes combined with high-throughput sequencing were developed.

In order to quantitatively compare patterns of genome-wide methylation, here we propose Methylation Context Sensitive Enzyme ddRAD (MCSeEd), a new method to call both DNA methylation polymorphisms in symmetric and asymmetric contexts and SNPs between samples either in presence or absence of a reference genome. To do so, highly multiplexed libraries are generated with a simultaneous double restriction-ligation employing an enzyme sensitive to methylation and an enzyme insensitive to methylation and sequenced with Illumina technology. Enzymes can be chosen with a virtual digestion allowing to maximise the discovery potential in the regions of interest. The bioinformatic pipeline relies on recognised free algorithms and is optimised for parallel implementation. Beyond the differentially methylated positions and regions calling (DMP, DMR), MCSeEd is capable to return SNPs from the generated sequences allowing for association analysis of variation with methylation context.

To test its effectiveness, MCSeEd was applied to a time course experiment comparing leukocytes DNA of horses before and after a period of intense training to reveal its epigenetic signature.

Twenty-two years old horses from the same farm were followed for four months during the training sampling blood every 30 days. Five bulks of four randomly chosen individuals were created and digested with two enzyme combinations in four-time points. Bulks were processed with MCSeEd wet and *in silico* technique.

Our results, demonstrating the reliability of MCSeEd either in depict methylation state or variation, will be reported and discussed.

P050

Demography, inbreeding and breeders' management in Italian Weimaraner (*Canis familiaris*) population

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The aim of the present study is to investigate the demographic asset, the inbreeding levels and breeders' management in Italian Weimaraner (WMR) population in 46 years of data, entered in the ENCI-Italian Kennel Club Studbooks with the first records tracing back to 1970. Data were analysed using SAS ® and CFC software. The WMR, according to Federation Cynologique Internationale (FCI) standard n° 99 (26 May 2015) is a medium to large sized hunting dog with a functional working type. The breed is part of the 7th FCI group Pointing Dogs, Section 1.1 Continental pointing dogs, type 'Braque'. The most recognisable and distinguishing phenotypical trait is the unique 'silver, roe or mouse grey' coat colour. Two varieties are present: short-haired (SH) and long-haired (LH). In the observed period, a total of 9974 subjects were entered in Italian stud books (Registro Origini Italiano (ROI)+RSR (Registro supplementare riconosciuti), sex ratio 1:1, varieties are present with a dramatic difference: 99% SH–1% LH. A constant increase in puppies' entries has been recorded in the last ten years. It should be underlined that 23.7% of the population has foreigner origin, 76% is entered in ROI and only 0.3% of the population is constituted by RSR subjects (founders recognised as typical by official judges, 3 generation pedigree). A complete genealogy was recorded for 13,002 dogs, the reference population is sized on 11,579 dogs, 8364 subjects are inbred. A total of 1929 breeding males and 2385 breeding females have been calculated. The population average inbreeding coefficient (F) is 0.025, in inbred dogs mean F is 0.038, the maximum calculated F is 0.375. In the last 10 years, F levels have always been lower than 0.05. In the whole population, 27 full-sib, 121 half-sib and 75 parent-progeny combinations have been recorded. The top producing sire originated 216 puppies, the top producing dam originated 64 puppies. The maximum number of litters per sire recorded was 33. The maximum number of litters per bitch recorded was 9. The average dams age is 3.6 years, and the average sire age is 4 years. Some breeding activities (litter birth) under 1 year of age and over 7 years of age were recorded in dams and sires. Objective data have been supplied for WMR breed conservation and study and breeding strategies planning a constant attention towards breeder management and coefficient level could be suggested according to ENCI rules.

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ANIMAL PHYSIOLOGY, HEALTH AND WELFARE

P051

Stage of gestation and dietary carbohydrates change ruminal microbiota structure in sheep

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Nutrition during pregnancy significantly affects the metabolic homeorhesis of the mother and the metabolism of the foetus with permanent effects on offspring performance. Human studies highlighted a central role of diets and physiological stages, mediated by the gastrointestinal microbiome, in the regulation of the host metabolism for food energy harvesting and, in particular, to influence the hormonal regulation of glucose metabolism. The aim of this study was to study the effects of different dietary carbohydrates on the microbiota structure of dairy sheep in their last month of pregnancy. Two groups of 6 Sarda sheep (BW: 47.4 ± 5.4 kg and BCS: 2.7 ± 0.2) were fed a glucogenic (starch) or a lipogenic (fibre) diet (37.0% vs. 46.5% of NDF and 26.1 vs. 10.0 of starch and sugars on DM basis, respectively) from –75 to the day of parturition (DoP). From each group, a ruminal fluid sampling was performed with oesophageal probe at –35 and –5 DoP. Samples were immediately frozen until DNA extraction and analysis by quantitative real-time PCR and DGGE analysis. Hierarchical cluster analysis and linear mixed models were applied for qualitative and quantitative statistics, respectively. At –35 to DoP the ruminal microbiome was strongly influenced by the diet while microbial groups clustered accordingly with the diet at this phase. On the other hand, at –5 DoP the dietary effect on microbial groups was not detectable and the ruminal microbial structure of the starch and fibre ewes showed a very high inter-animal similarity.

The number of bacteria, methanogens, *Butyrivibrio fibrisolvens* and *Prevotella ruminicola* were not influenced by diets at –35 to and –5 DoP. *Protozoa*, both at –35 and –5 DoP tended to be higher in fibre ewes ($p<.01$). The diet had a significant effect on the number of *Fibrobacter succinogenes*, with higher values in fibre than in starch ewes ($p<.05$). Gestation stage influenced the Fungi population, which declined significantly from –35 to –5 DoP ($p<.05$). Observed microbial changes and the depression effect of the fungal growth rate could be due to the increase in ruminal passage rate with the advance of gestation. In conclusion, pregnancy stage has led to a change in the structure of the microbiome, which needs to be further investigated to better understand possible links with animal performances.

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P052

Evaluation of industrial by-products

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The use of industrial by-products increased constantly in the present years in order to reduce the economic and environmental impact of animal production system. Nevertheless, to use correctly residuals as feedstuffs, it is necessary to know their nutritional characteristics. The aim of the present study was to evaluate the chemical composition and *in vitro* fermentation characteristics of nine by-products obtained by different industrial processes: three by olive oil production (dry pomace DP1, DP2, DP3); four by fruit transformation (grapecake GC and orange fruits pulp OFP1, OFP2, OFP3) and two from sugar industry (glutamic beet pulp GBP and dried beet pulp BP). Each sample was analysed for chemical composition according to official methods. To consider the use of these by-products in swine nutrition, each sample was incubated at 39 °C for 96 h, into serum bottles under anaerobic condition with swine faecal inoculum. At the end of incubation, cumulative gas production (OMCV), organic matter digestibility (OMD), short chain fatty acids (SCFA) and NH_3 were determined. All data were statistically analysed. As expected, chemical composition of the tested by-products varied consistently in function of the productive process as well as the nature of raw material. Regarding protein, GPB showed the highest value (31.17% a.f.), while in the other by-products crude protein ranged from 5.57 to 13.14% a.f., in OFP1 and GC, respectively. Structural carbohydrates showed the highest levels in GC (NDF: 58.89; ADF: 55.97, ADL: 34.24% a.f.), while the lowest values were registered in OFP1, OFP2 and OFP3. The olive oil residuals showed lipid values exceeding 25.0% a.f. Both fermentation parameters and end-products were influenced by chemical composition. In particular, OMD was significantly ($p<.01$) correlated to dry matter and ether extract, while NDF, ADF and ADL content reduced significantly ($p<.05$) OM digestibility; OMCV was significantly ($p<.05$) related only to structural carbohydrates fraction. All chemical parameters influenced significantly ($p<.01$) SCFA production; on the contrary, no correlations were observed between NH_3 production and chemical parameters. The evaluation of chemical composition and *in vitro* fermentation parameters is an essential step in order to utilise by-products as feedstuffs in swine nutrition; the search for other compounds, as well as the direct *in vivo* confirmation, will represent a subsequent step.

P053

Responses to heat stress of three rabbit synthetic breeds: physical and sensory traits of the longissimus dorsi muscle

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Rabbits are very sensitive to high environmental temperature due to their limited number of sweat glands. This typically leads to decreased performance with possible effects on meat quality aspects. However, it has been shown that different rabbit genotypes might display different performances when farmed under heat stress. This research studied the effects of the rabbit breed and the farming temperature on physical and sensory quality of the longissimus dorsi (LD) muscle. A total of $n = 90$ LD muscles were obtained from three different synthetic rabbit breeds: Pannon Large (PL), Pannon White (PW) and Pannon Ka (PK). Each genotype was farmed at two different temperatures, 20 °C (normal) and 28 °C (heat-stress conditions). After slaughter, the LD samples were submitted to physical analysis (pH, $L^*\,a^*\,b^*$ colour, thawing and cooking losses, and Warner-Bratzler shear force – WBSF). A trained sensory panel ($n = 12$ members) received a list of descriptors (olfactory, gustative, textural) to score them on numerical and continuous scales from 0 to 10. For each sample, panellists were also asked to indicate if and which of a list of off-odours and off-flavours they could recognise. Data were analysed with a two-way ANOVA with breed and temperature as fixed effects. As for sensory data, a χ^2 test was performed on off-odours and off-flavours characterisation. The breed significantly affected water total losses as well as WBSF values of LD meat. The PW rabbits showed higher thawing loss compared to PL ones ($p<.05$). However, it was the PK breed that displayed the highest cooking loss ($p<.01$) total loss ($p<.05$), and meat WBSF values ($p<.01$). The farming temperature affected some physical meat traits: the LD pH increased when rabbits were outside their thermoneutral zone ($p<.01$) which determined lower thawing loss ($p<.05$), and b* value ($p<.05$). Overall, the breed did not affect the LD sensory traits, except for meat toughness, whose score was the highest ($p<.05$) in PK rabbits, thus supporting the results on the physical traits of the LD meat. The environmental temperature did not affect the LD sensory attributes, except the ‘pungent’ off-odour who increased in rabbit farmed under heat stress condition ($p<.\ 05$). It was concluded that the PK breed was the less acclimated to heat which negatively reflected on physical and sensory meat quality traits, and that the heat stress did not impair the LD meat quality traits.

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P054

Response of the bovine milk microbiome to different dry-cow treatment approaches

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Most lactating cows are routinely treated in each quarter with antibiotic at drying off. This is defined as the ‘blanket’ approach, and it is considered effective in preventing new infections and curing the existing infections. However, this farming practice can potentially interfere with the milk microbiota balance, as well as promote the selection for antimicrobial resistance. Therefore, the impact of this practice on the physiological milk microbiota deserves further investigation. The objective of our study was the evaluation of the possible effects that different treatments at drying-off can have on the milk microbiota of dairy cows. The study involved 5 cows from drying-off to 10 days after calving. The samples were collected at drying-off, 3 days (colostrum) and 10 days after calving, for a total of 60 samples. At drying-off, each animal received four different treatments: the left front quarter did not receive any treatment (control quarter), an internal teat sealant based on bismuth subnitrate was applied to the right front quarter, the right rear quarter received dry therapy with cefalonium and the left rear quarter was treated with another antibiotic (cloxacillin). For 16S rRNA-gene sequencing, bacterial DNA was extracted from 5 mL of milk samples, amplified using the primers for the V3-V4 hypervariable regions and sequenced in one MiSeq (Illumina) run with 2 × 250-base paired-end reads. There was a relatively high variability in the composition of milk microbiota between quarters already at drying-off. The variation in microbial composition among the different treatments was small. In particular, 10 days after calving there were no significant differences compared to the control quarter. In agreement with previous works, the bacterial phyla *Proteobacteria*, *Firmicutes*, *Bacteroidetes* and *Actinobacteria* were relatively abundant at all sampling times.

P055

Cow-calf morphometric relationships and birth dystocia in Piemontese breed

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Dystocia has been defined as a difficult birth resulting in prolonged calving or severe assisted extraction of the calf at birth, generally higher in primiparous than in multiparous cows. Several scales have been described to determine the degree of assistance provided during parturition, but for the practical purpose, a simplified scale can be used that only provides for the absence or presence of dystocia (easy or difficult calving). The reduction in the number of dystocic calving is an extremely interesting aspect for beef cattle breeders, particularly in Piemontese breed, and the identification of maternal-foetal morphometric relationships that can influence birth difficulties is a non-negligible aspect of the problem. To this aim, several measures relating the size of the cows (height at withers) and the development of the rump (rump length, width at hips, at thurls and at pins) were recorded on 162 Piemontese multiparous cows, to be related to calf measurements (weight, girth at head, at shoulders, at belly and at tail) at the time of delivery (all single calving); the delivery characteristics (easy =124 or difficult =38) so as the calf sex (male =86 or female =76) were also recorded and data analysed by GLM Univariate and Linear Discriminant Analysis to evidence differences among animal conformation related to dystocia and to identify discrimination thresholds for the choice of cows and heifers to be used in the farm. Only few cow measures are different in animal presenting easy or difficult calving (height at withers 127.29 vs. 125.25 cm, *p*=.024; and rump length 52.02 vs. 50.72 cm, *p*=.016), while almost all calf measures differ in calves with easy or difficult birth (weight 53.42 vs. 63.79 kg, *p*=.000; girth at head 75.49 vs. 85.05 cm, *p*=.000; at belly 79.00 vs. 87.95 cm, *p*=.000; and at tail 60.56 vs. 48.11 cm, *p*=.002). None of the recorded measure on calves is influenced by calf sex (*p*>.284). In the discriminant analysis applied to the cow and calf measures, the different calving was not easily identified. In fact, 82.7% of the original grouped cases were correctly classified: 94.4% of easy but only 44.7% of difficult calving. In detail, the discriminant function was mainly affected by all calf measurements while was less affected by cow dimensions (only width at thurls). Then, the use of the discriminant analysis applied to the cow and calf measurements does not seem to be effective in correctly predicting the calving characteristics in Piemontese cows.

P056

Influence of beef cattle management on blood biochemistry

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To investigate the effects of different beef production systems on some blood parameters considered indicators of welfare and health status, a first study (trial 1) was conducted on 29 Piemontese bulls (15 reared in pen in group in a fattening system, PP-FS; 14 reared in a tied stall in a cow-calf system PT-CS) to evaluate the effect of the housing and management system. A second study (trial 2) was conducted on 45 bulls reared through FS in pen in group (15 Blonde d'Aquitaine, BP; 15 Irish cross breed, CP; 15 Piemontese, PP) to evaluate the adaptability of the studied breed to the fattening period. In both the trials, the animals were housing on concrete and littered floor. Blood was analysed for reactive oxygen metabolites (d-ROMs), white blood cells (WBC), aspartate aminotransferase (AST) and creatinine (CREA) at the beginning (after the quarantine period; T0) and at the end of the fattening period (5 months; T1). Data were analysed by GLM ANOVA procedure. Trail 1 at T0 showed only CREA higher in PP-FS than PT-CS (2.7 and 2.1 mg dL^{-1}, respectively; *p*<.01). At T1, PT-CS showed the worst adaptation, in terms of higher d-ROMs than PP-FS (98.0 and 46.1 U CARR, respectively; *p*<.01). Always at T1, WBC showed normal values, but higher in PT-CS than PP-FS (7.8 and 6.0 10E03 cells μL^{-1}, respectively; *p*<.01). CREA was always higher in PP-FS than PT-CS (2.6 and 2.2 mg dL^{-1}, respectively; *p*<.05). No differences were found for AST. In the trial 2, at T0 BP showed lower d-ROMs values than CP and PP (39.1, 69.1 and 68.7 U CARR, respectively; *p*<.01) and the best adaptability with 81% d-ROMs reduction from T0 to T1. PP also showed a reduction of 33% in d-ROMs in the same period. Differently, CP remained unchanged and at T1 resulted the group with the worst adaptation, in terms of higher d-ROMs values than BP and PP (70.4, 7.4 and 46.1 U CARR, respectively; *p*<.01). BP showed the highest increase of WBC (+210%) and AST (+53%) between T0 and T1, with the final values being over the normal range (32.56 10E03 cells μL^{-1} and 131.00 U/L, respectively). CREA showed an increase between T0 and T1 for CP and BP, with final values being higher than BP (2.26, 2.60 and 1.22 mg dL^{-1}, respectively; *p*<.01). In conclusion, the rearing system with animals loose in group showed better blood indicators of welfare and health status than the animals reared in tied stall. The breed is an important factor affecting animal welfare that combined with a correct management can improve the animal health.

P057

Preliminary results of the efficacy of an alternative pain management approach for piglet castration

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Surgical castration of piglets without anaesthesia or analgesia is a painful and stressful procedure. Pharmacological treatments to reduce pain can present some limitations as shown by several studies performed on human neonates, due to possible adverse and discordant effects. Alternative non-pharmacological approaches seem to be effective and feasible. For instance, in human neonates, small amounts of sweet solutions were proven to be effective, presumably mediated by endogenous opioids.

The aim of this study was to evaluate the efficacy of an alternative pain management approach (oral glucose) on behavioural and physiological responses of piglets subjected to surgical castration.

Sixteen three-day-old male piglets were recruited from 4 different litters and allocated to 4 treatment groups: CG – surgical castration and oral administration of glucose 10%; CW – surgical castration and oral administration of water; MG – manipulation without castration and oral administration of glucose 10%; MW – manipulation without castration and oral administration of water. Surgical castration was carried out by a trained operator, in compliance with the Council Directive 2008/120/EC. Piglets of the same litter were isolated into a small nursery and filmed for 15 min before and 30 min after castration. Ninety-six pictures of the faces were collected pre- and post-procedure and scored with the Piglet Grimace Scale (PGS) by 3 treatment-blind observers. Heart rate before and during the procedure and latency at the first suckling were recorded. The inter-observer reliability was good with an overall interclass correlation coefficient value of 0.77. In the CW group, mean PGS score before castration was 1.89 ± 1.96 and increased to 3.11 ± 1.62, after castration. During the procedure, mean heart rate was lower in castrated piglets receiving glucose 10% (127 bpm) compared to ones receiving water (220 bpm). Piglets receiving water required a longer time (s) to suckle (CG =36.0; CW =121.0; MG =10.3; MW =28.3).

Although preliminary, these results suggest that an alternative non-pharmacological approach might represent an effective method to control pain induced by routine husbandry procedures in pigs. Further research providing oral glucose in combination with pharmacological treatment is required to identify the most feasible and effective pain relief treatment for on-farm surgical castration in pigs.

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P058

Upgrading the pipeline to analyse 16S rRNA-gene sequencing data

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In dairy cattle, microbiomes are known to affect milk production, well-being and health of the animals. Within the FARM-INN project, the rumen, milk and hindgut microbiomes of dairy cows will be analysed to better understand their contribution to the efficient production of high-quality milk for direct consumption and transformation into cheese and other dairy products. From a statistical and computational perspective, the available methods and software tools are continuously and rapidly evolving, making it fundamental to keep pace with the latest advancements. Aspects like parameters for quality filtering and normalisation, alpha- and beta-diversity metrics, software packages and reference databases (e.g. GreenGenes and Silva) are being thoroughly compared in this study. For instance, we showed that in a closed OTU picking approach a stricter (19) or looser (3) Phred threshold yielded similar results in terms of retained sequences (48.7% vs. 49.6%), although acting at different levels in the pipeline (reads filtering vs. alignment against the reference database). Which software package to use for which operation is also relevant: the most popular tools include Qiime (Quantitative Insights into Microbial Ecology), Mothur and Dada2 (a Bioconductor R package). The critical transition from Qiime 1.9 to Qiime 2 will also be addressed here and may bring about a major overhaul of existing pipelines. Finally, the most common algorithms for the prediction of microbial gene functions from microbiota composition, i.e. Picrust and Tax4Fun, will be benchmarked against each other: preliminary results showed a substantial overlap of retrieved orthologs and pathways (~92% common results). The objective of this work is to upgrade the current bioinformatics and biostatistics pipeline for the processing and analysis of 16S rRNA-gene sequencing data, by integrating the most recent advancements in terms of statistical analyses and bioinformatics processing. The upgraded pipeline will be published alongside full documentation and a set of user-friendly guidelines for reproducibility of results.

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P059

Investigation on the 'Club Foot' disorder in Arabian Pureblood horses reared in Italy

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Horses used in equestrian disciplines are subjected to intense exertion, making the athlete's physical integrity a vital point, in order to optimise performance as well as to limit the onset of traumatic disorders. Several parameters are involved in choosing a subject for sport activities, one of the most decisive being the absence of any disorder or malformation of the limbs. The aim of this research was to detect the incidence of the disorder commonly referred to as 'Club Foot' or 'Mismatched Foot' in Arabian Pureblood horses, attempting to understand its causes. In this breed, the pathology is widespread because, in their environment of origin, the rocky desert, a hard and almost goat's hoof is not disabling so selection against this disorder has never been done. The 'Club Foot' is defined as acquired or congenital flexural deformity of the distal interphalangeal joint, caused by a shortening of the musculotendinous unit of the deep digital flexor tendon. The hoof capsule is distorted and the palmar angle of the third phalanx increases to 60° or more, and the horse is forced to walk on his toes. Most commonly, this condition affects the forelimbs, one or both. In the congenital forms, the newborn is unable to extend the joint of the distal limb, it cannot place the foot on the ground and subsequently tries to walk on the toe. One hundred forty adult Arabian Pureblood horses (51 males and 90 females) belonging to 8 Italian different farms during the period 1982–2017 were considered. For each horse we observed the presence or absence of the disorder following these criteria: alignment of the front hooves, recognition of pathological signs through analysis of hoof axis, presence of dishing on the anterior hoof wall from the coronary band to the toe, hoof width, heel height, frog atrophy, presence of laminitis symptoms. Four grades of deformity were esteemed. Moreover, we considered the environmental condition of each farm: box stalls, paddocks, nutrition, orientation and other observations. The chi-square test was applied. Two grades of deformity were observed (I and II) with 20 females and 14 males (24.11% of the examined horses) displaying the disorder. No differences between males and females or between shod (59%) and unshod (41%) were observed. Environmental conditions do not influence the rate of pathology in the different farms, with a prevalence of the disorder ranging from 7.69% in one farm to 66.67% in two farms. Future works may include a larger number of animals.

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P060

Larvicidal activity of neem oil (*Azadirachta indica*) formulation against larva of *Aedes albopictus* mosquitoes to improve animal welfare

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Aside from their importance as vectors of disease agents of animals, mosquitoes are a cause of irritation, blood loss, and allergic reactions. They can also disrupt normal behaviour of livestock. For cattle, mosquito bites can result in decreased weight gains and milk production. *Aedes albopictus*, known as 'tiger mosquito', is an Asian insect, widespread in all continents and also recorded in Italy. The mosquito fight is directed especially against larvae. This is because the fight against adults is temporary, unsatisfied and polluting for the environment, while larval treatment is more localised in time and space, resulting less dangerous. The aim of our work is to develop new natural anti-mosquito formulations (the neem oil, p.a. azadirachtin). Among various tested substances the neem oil has shown good activity. The neem oil (AZ: 0.3%) was tested at different concentrations: 0.005 g; 0.01 g; 0.025 g; 0.05 g, 0.075 g on larvae at the L1–L2 stage and control. Four replicas were performed. The conditions were a 14-hour photoperiod and an average water temperature of 25 °C.
At the lowest doses (0.05 g and 0.01 g) the mean total death larvae compared to the control is not statistically significant. The doses 0.025 g, 0.05 g and 0.075 g are all equally effective, completely eliminating the larvae. Highly significant differences were found between the last 3 doses and the control. Analysing the results up to the eighth day of treatment, that is the day when the larvae in the control tests were either completely transformed into adults (48%) or dead (52%), differences in the timing of elimination of larvae between different doses were recorded. At the dose of 0.005 g at the eighth day, 48% were dead and another 1% died within 17 days. At the dose of 0.01g at the eighth day, 61% of the larvae were dead, 10% died within 56 days. At the dose of 0.025 g

at the eighth day, 85% of the larvae were eliminated and all died at the 26 day, at the dose of 0.05 g after 8 days, 94% were eliminated and all larvae died in 21 days, and at a dose of 0.75 g, all larvae died in 7 days. The results obtained are interesting if we consider the importance nowadays to develop new anti-mosquito drugs to supply the drug resistance of the used products.

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P061

Plasma isoprostane concentrations as biomarkers of oxidative stress during physiological transitions of dairy cows

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Oxidative stress contributes to the incidence and severity of dairy cattle diseases. Isoprostanes (IsoP) are molecules generated from interactions between free radicals and membrane phospholipids and serve as excellent indicators of free radical-mediated lipid damage. Previous studies showed that plasma IsoP concentrations increase in cows during times of oxidative stress such as around calving and during coliform mastitis. Although IsoPs are recognised as biomarkers of lipid damage, the biological functions of these molecules are not well defined. The objective of this study was to assess changes in different IsoP isoforms during major physiological transitions and to determine the impact of select IsoP on bovine endothelial cell responses to pro-oxidant challenge *in vitro*. Blood samples were collected from 7 healthy Holstein dairy cows beginning from 6 days before dry-off, at dry-off, 1 day after dry-off (d + 1), 2 days after dry-off (d + 2), 6 days after dry-off (d + 6), 12 days after dry-off (d + 12), when moved to close-up (cu) and again at 7 days post-calving (c + 7). Plasma was separated, flash frozen, and stored at −80 °C. Isoprostanes were extracted from plasma using solid phase extraction columns and quantified using LC/MS/MS. Primary bovine aortic endothelial cells were stimulated for 12 h with 10 nM of selected IsoP. The majority of IsoP isoforms studied showed no significant changes in concentration across the sampling period. However, concentrations of 8-iso PGA2 increase until calving but significant decrease at c + 7. Conversely, 15F2t-IsoP peaked at dry-off with a gradual decrease the days following dry-off until c + 7, where a significant increase was observed. Treatment of cultured bovine endothelial cells with 15F2t-IsoP significantly reduced endotoxin-induced apoptosis and reduced reactive oxygen species production following pro-oxidant challenge. Results of this study show that isoprostane production represents a stable biomarker of oxidative stress in periparturient dairy cattle. Specific isoforms of IsoP may have beneficial effects on vascular integrity during times of oxidative stress. Establishing threshold values for isoprostanes can facilitate the development of management strategies to control oxidative stress during times when dairy cattle are most susceptible to health disorders.

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P062

Semen freezability in Italian chicken breeds

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The purpose of this study was to assess semen sensitivity to cryopreservation in two Italian chicken breeds: *Mericanel della Brianza* (MB) and *Milanino* (MI). MB ($n = 27$) and MI ($n = 19$) cockerels were housed in outdoor pens and the semen was regularly collected twice a week. Soon after semen collection, the ejaculates were pooled into semen samples and diluted to 1.5×10^9/mL in pre-freezing Lake diluent, refrigerated at 5 °C and transferred to the DiMeVet laboratory (University of Milan) for further cryopreservation processing. In brief, semen samples were further diluted to 1×10^9/mL in pre-freezing Lake diluent added with DMA (6% final dilution) and trehalose (0.1 M), incubated for 1 min at 5 °C, loaded into 0.25-mL French straws and frozen in nitrogen vapours. Thawing was performed in a water bath at 38 °C for 30 sec. Ejaculate volume and concentration were recorded; sperm viability (SYBR14-PI staining) and motility (SCA System) were assessed in pooled semen samples before and after cryopreservation.

Good semen production was obtained in MI cockerels and 12 semen pools were processed for cryopreservation, whereas, poor semen production was obtained in MB cockerels and only 3 semen pools were processed. The quality of fresh semen was higher in MI compared with MB males: viability and motility were 91% and 84%, respectively in MI birds, and 77% and 51%, respectively in MB birds. As expected, cryopreservation caused a significant decrease in sperm quality parameters: viability and motility were reduced to 42% and 33%, respectively in MI males, and to 29% and 15%, respectively in MB males. The mean recovery rate of viable, motile and progressive motile sperm after cryopreservation was 46, 37 and 18%, respectively in MI birds, and 38, 33 and 51%, respectively in MB birds. These results highlighted the high variation in motility and progressive motility recorded among semen samples. The present data are considered preliminary results and further studies are required to confirm the different semen freezability found between and within the breed. The functional and biochemical characterisation of semen samples with extreme (highest vs. lowest) sensitivity to cryopreservation is of undeniable interest to identify biochemical markers and will be investigated.

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P063

Selective hunting plans as source of reproductive data in wild boar of the National Park of Sibillini Mountains

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The rapid increase of wild boar (*Sus scrofa*) in Italian Apennines and in many European areas is due to its generalist behaviour that has led to wide expansion causing damage to agriculture activities. Therefore, the Parks, in order to reduce density of the wild boar population, started monitoring and harvest plans, so since 1998, the Sibillini Mountains Park has implemented its wild boar management plan. Data, obtained from forms filled out by selective hunters, regarding 1821 wild boars hunted from 2015 to 2017, have been available. To evaluate the effect of year, season and age class (I and II) data on 557 wild females (their uteri were cut open to evaluate pregnancy rate and litter size) has been statistically analysed with JMP 10 software. The descriptive statistical analysis on hunted females showed: average age 26.11 ± 10.77 months, mean weight 63.11 ± 15.96 kg, mean foetus number 4.42 ± 1.25. The ANOVA considering year effect (2015, 2016, 2017) did not show significant differences for age, weight and foetus number during the three-year period. The effect of season showed significant differences ($p<.05$) for age and weight; wild females hunted in Winter were older than those hunted in Spring (27.07 vs. 23.77 months). Moreover, females hunted in Autumn reached a higher weight (68.12 kg) than the ones hunted in other seasons (Winter 62.81 kg, Spring 61.78 kg, Summer 61.68 kg). Age class II females showed heavier weight ($p<.05$) than those of class I (73.28 vs. 56.06 kg). In the three-year period, overall reproductive data on 550 wild females showed the following physiological status: 70.54% no pregnancy vs. 29.46% pregnancy. Four hundred and twenty-six out of 557 females, 22.76% were lactating vs. 77.23% no lactating. Sows hunted in Winter and Spring showed a higher ($p<.05$) pregnancy rate (62.35% and 33.33%, respectively) than those hunted in Summer and Autumn (0.62% and 3.70%, respectively). The pregnancy status was confirmed by the trend of hunted lactating females, showing a higher percentage of lactating sows in Spring (72.16%) and Summer (18.56%) than in Autumn and Winter (5.15% and 4.12%, respectively). The highest percentage of pregnant females was observed in age class II (57.41%). The analysis of data in the three-year period shows age, weight and litter size homogeneously distributed, while reproductive performance confirms the typical seasonal reproductive efficiency of wild boar with the main rutting period in Autumn-Winter seasons and lactations mainly in Spring.

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P064

In vitro digestibility protocol applied to BARF diets: pros and cons

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This study evaluated the *in vitro* dry matter and crude protein digestibility in natural pet food, also termed BARF diets. For this purpose, eight samples of dogs BARF diets and two commercial dog food, used as reference materials, were analysed and tested in the assay. The BARF diets were based on raw beef and poultry by-products, while the commercial pet food was one dry and one wet. All samples were analysed for dry matter (DM), crude protein (CP), ether extract (EE) and ash content. Furthermore, using an *in vitro* assay, simulating gastric and small intestinal digestion, Q31 both dry matter digestibility (IVD-DM) and crude protein digestibility (IVD-CP) have been measured. Briefly, after the pepsin (39 °C for 6 h) and pancreatin (39 °C for 18 h) incubation in the IVD-DM and IVD-CP test, the undigested residues were dried at 105 °C overnight. The IVD-DM was calculated from the difference between dry matter in the sample and the undigested residue. The IVD-CP was calculated from the difference between the nitrogen content in the original sample and the nitrogen content undigested residue measured by the Kjeldahl method. All BARF diets and wet pet food were characterised by high moisture content (DM: 380 $g{\cdot}kg^{-1}$), while in the case of dry pet food DM content was 920 $g{\cdot}kg^{-1}$. On average, BARF diets and commercial diets were characterised by the following values, on dry matter basis: CP, 368 $g{\cdot}kg^{-1}$; EE, 442 $g{\cdot}kg^{-1}$; ash, 52 $g{\cdot}kg^{-1}$. All BARF samples and reference materials were characterised by high digestibility values. Both IVD-DM and IVD-CP reached values higher than 80%. Of note, in the case of IVD-DM, a substantial variability within samples has been observed (SD: ±5.5). While in the case of IVD-CP value observed presented less variability (SD: ±1.4). In light of these results, it can be concluded that proposed IDV method has some potential in determining protein digestibility in BARF diets, while the assay seems to be limited for measuring DM digestibility, as indicated by the large SD recorded in the BARF diets. The reason for this is unclear and merit further investigations.

P065

Lactating ewes responded to a glucose tolerance test with higher glucose and insulin concentrations both in early and in mid-lactation compared to lactating goats

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In a recent study, we observed lower milk persistency and higher body fat accumulation in dairy ewes compared to dairy goats, especially during mid-lactation. Since these species might have a different regulation of glucose metabolism, in the same experiment we carried out glucose tolerance tests (GTT) to test this hypothesis.

After parturition, 30 Sarda ewes and 26 Saanen goats were fed a high-starch diet (HS: 20.4% starch; DM basis), whereas starting at 92 ± 11 days in milk (DIM) each species group was divided into two dietary subgroups, receiving HS or low starch diets (LS; 7.8% starch; DM basis). LS diet was obtained by replacing most of the corn meal and all of the barley meal of HS diet with soybean hulls, very rich in highly digestible fibre. At 50 and 148 DIM, GTT were performed in 18 ewes and 18 goats. One millilitre of a 50% glucose solution per kg of BW was injected into the jugular vein of each animal. Blood samples were collected before (−15 min (min)) and after (+5, +10, +15, +30, +45, +90 and +180 min) glucose injection. Data were analysed by the PROC MIXED procedure of SAS with repeated measurements.

The dietary starch level applied in mid-lactation did not affect any of the results. At 50 DIM, basal plasma glucose (*p*=.10) and insulin concentrations (*p*=.08) were numerically higher in ewes than in goats. After glucose infusion, glucose (206.3 vs. 177.8 mg/dL ±12.9, *p*=.048) and insulin (0.58 vs. 0.32 μg/L ± 0.10, *p*=.027) concentrations were significantly higher in ewes than in goats. At 148 DIM, basal plasma glucose (62.2 vs. 51.6 mg/dL ±3.62, *p*=.011) and insulin (0.34 vs. 0.13 μg/L ± 0.09, *p*=.036) concentration were significantly higher in ewes than in goats. After glucose infusion, glucose (*p*=.06) and insulin (*p*=.07) concentrations were numerically greatest in ewes. The area under the glucose concentration curve, fractional glucose turnover rate and half-time were not affected by species or diet, in both stages of lactation. Quantitative Insulin Sensitivity Check Index (QUICKI) and Revised QUICKI were higher in goats than in ewes in both stages of lactation, while the Homeostasis Model Assessment (HOMA) was highest in ewes. In conclusion, the highest plasma glucose, insulin and HOMA observed in ewes suggested the presence of an insulin resistance status, which was more marked in mid than early lactation and which can be the cause of greatest body fat deposition and lowest milk yield persistency observed in this species.

P066

Comparison among four different bacterial DNA extraction protocols for analysing milk metagenomics

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Bovine udder is colonised by a huge number of microorganisms that constitute the intramammary ecosystem, with a specific role in modulating not only the udder homeostasis and mastitis susceptibility but also the quality of the dairy products. Therefore, information on milk microbiota composition will facilitate the dairy industry in the production of safe and high-quality products. However, generating high-quality bacterial DNA could be critical.

In the present study, bacterial DNA from healthy milk samples was isolated by four different protocols to evaluate the effect of the extraction procedures on milk microbiota composition. For the characterisation of the milk microbiota by 16S deep sequencing, 500 mL of bulk tank milk samples were aseptically collected from three different farms and bacterial DNA was extracted by using an internal laboratory protocol and three commercial kits. Bacterial DNA was then amplified using the primers for the V3–V4 hypervariable regions and sequenced in one MiSeq (Illumina) run with 2 × 250-base paired-end reads. Data analysis was performed by using QIIME suite and SILVA 132 as a reference database for taxonomy.

The results showed that the four extraction kits performed very differently and showed a significant separation on both microbial richness (alpha-diversity) and composition (beta-diversity). In particular, the relative abundance of some genera (e.g.: *Lactobacillus*, *Acinetobacter* and *Microbacterium*) were consistently altered by the extraction method. Based on these data, then, particular attention must be kept in choosing the proper extraction method, for example, carefully evaluating eventual biases towards or against bacterial genera of interest. Moreover, we believe that, in order to define which kit best resembles the original bacterial community, an additional set of experiments with a mock community with known composition should be performed.

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P067

Effect of weeping teats on milk microbiome in goats

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In dairy goats, variability in teat conformation is often observed, such as the presence of the so-called ‘weeping teats’. This trait, observed in goat breeds selected for high milk production, is characterised by the presence of milk-secreting tissue in the wall of the teat where the milk can pass through skin pores out to the external epithelial surface and be released onto the skin surface, resulting in a ‘weeping teat’. The aim of the study is to characterise the milk microbiota composition of the abnormal trait ‘weeping teats’ in Italian Saanen and Alpine goats in order to understand the effect of this trait on the milk microbiome.

For the characterisation of the milk microbiota by 16S rRNA-gene sequencing, milk samples were aseptically collected from three different herds for a total of 46 weeping-teats and 32 normal teats. Bacterial DNA was extracted by using an internal laboratory protocol (Cremonesi et al. 2018), then amplified using the primers for the V3–V4 hypervariable regions and sequenced in one MiSeq (Illumina) run with 2 × 250-base paired-end reads. On average, 97,098 reads per sample were obtained: after quality filtering (Phred >19), 68.3% of the reads were retained for subsequent analysis. After removing OTUs with ≤10 counts in ≤2 samples, a total of 6675 OTUs were detected. The milk microbiota was dominated by the phyla *Firmicutes, Proteobacteria* and *Actinobacteria*, with little differences between weeping and normal teats (e.g. 46% vs. 42% *Firmicutes*, 26% vs. 29% *Proteobacteria*, 16.7% vs. 16.9% *Actinobacteria*, respectively). Alpha diversity metrics were very similar between weeping and normal teats, with no comparison showing a significant difference.

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P068

Gut microbiome response to dietary prevention regimes in pre-weaning piglets

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Post-weaning diarrhoea (PWD) represents the most important threat for the pig industry all over the world with a morbidity over 50% among weaned piglets during outbreaks of the disease. Given the public health concerns about the spread of multi-resistant bacteria due to the use of antibiotics in livestock, it is necessary to develop alternative strategies to restore microbial balance and control post-weaning diarrhoea in piglets. To date, the most promising alternative strategies are mainly based on the use of substances that act on bacteria indirectly by stimulating the immune system, or by improving gut health.

Therefore, the aim of our study was to evaluate the effect of an alternative treatment compared to antibiotics on supporting the health of the gut microbiota of pre-weaning piglets. Twenty-four litres were randomly divided into the following 3 treatment groups: (i) basal diet without any preventive treatments; (ii) basal diet supplemented with a mixture of garlic and oregano Q32 essential oil (500 g/ton of feed) and without any preventive treatments; (iii) basal diet not supplemented but using preventive antibiotics (cefquinome: 2 mg/kg BW for 5 days while castration and tattooing, tildipirosin: 4 mg/kg at weaning, amoxicillin: 20 mg/kg BW per os for 5 days at weaning) and antiparasitic (toltrazuril: 20 mg/kg BW at birth) treatments.

For metabarcoding analysis, rectal swab samples were individually collected at four time points (from birth to weaning) and the V3–V4 hypervariable regions of the bacterial 16S gene were sequenced in one MiSeq (Illumina) run.

Results revealed that the gut microbiota of pre-weaning piglets is dominated by the phyla *Firmicutes* (51%), *Bacteroidetes* (25%) and *Proteobacteria* (16%), which together make up for 92% of all microbes. The gut microbiota clearly changed over time: from the analysis of variance, all taxa showed significantly different relative abundances across time points, as well as alpha diversity indexes; based on the matrix of Bray-Curtis dissimilarities, samples clustered separately per time point. On the other hand, no clear differences among treatments were observed in terms of taxa abundances or alpha- and beta-diversity indexes. These preliminary results suggest that the tested treatments do not seem to exert major effects on general properties of pre-weaning piglets' gut microbiota, however, modifications of specific taxa may occur.

P069

Survey on mycotoxin content in feed and milk from sows

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The transmission of mycotoxins and their metabolites between animals and humans or between animals (mother-child) can occur through milk, but data for the pig are scarce. The study investigated if the intake of mycotoxins by the sow leads to a transfer of the same to piglets, through the suckled milk. Furthermore, the study evaluated the level of the main mycotoxins in farm feeds for sows.

Samples of the feed for lactating sows were obtained in 19 farms that (1) have an in-house feed mill; (2) administer the same batch of feed at least 5 days before the farrowing until the time of sampling (1st day of lactation); (3) preferably do not use chelating agents or warn of their presence. Per each farm, colostrum was obtained from 2 or 3 sows. The content of aflatoxin B1/B2/G1/G2 (AF), fumonisins (FU), deoxynivalenol (DON) and zearalenone (ZEA) of feeds was assessed by commercial ELISA kits, and confirmed by LC-MS at Laemmegroup lab.

Levels of mycotoxins present in the feeds were in general very low (10; 12; 17; 2 positive samples for AF, FU, DON and ZEA, respectively), always within the limits of the law or recommended, except for two samples (one for AF, one for DON). Based on values in the feed, colostrum samples from 13 farms were tested for at least one mycotoxin (AF; FU; DON). All colostra were negative for AF. The quantity of AF found in the feeds was apparently too low to detect their presence in the colostrum, in fact, in other studies, a transfer to pig milk was highlighted only at higher dosages. Despite FU was present in small quantities in the feed, a slight signal (under the standard quantification limit) was commonly observed in the colostrum (5/11). This indicates that attention should be paid in the control of feeds for sows for fumonisins content. DON, found averagely with values in feed less far from the suggested limit, was frequently present in colostrum (10/14). In the farm exceeding for law limits for DON in feed, the higher level in colostrum was seen (10.9 µg/kg). This value, considering the ratio of 1/5 between the dry matter of porcine colostrum and the reference feed, is about 1/20 of values, showing toxicity in the young pig. The absence of reference values for neonate pigs, their reduced immunocompetence, the risk of higher and longer ingestion of DON by sows, suggest improving the control of sow feed. More research on DON transfer and toxicity in piglet are desirable.

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P070

Variations in porcine milk oligosaccharide composition across breed and association with sow reproductive performance

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Oligosaccharides (OS) are naturally indigestible carbohydrates found in mammalian milk. They exert prebiotic and prophylactic effects, promoting growth and preventing disease, and are being explored as potential alternatives to antibiotic use in animal production. The composition of porcine colostrum OS (PCOS) may influence the health of the neonate giving rise to robust adult pigs, less susceptible to illness. The changes in structure and composition of PCOS across breed and parity order are unknown, as well as the connection with the litter performance. The aim of the present study was to characterise the PCOS composition from sows differing for breed and parity order and to correlate it with the sow mother traits.

Colostrum samples were gathered from 83 parturitions of sows belonging to three different breeds important in Italian heavy pig production: 44 Large White, 27 Landrace and 12 Duroc. Samples were taken between the birth of the first and the last piglet from sows not pharmacologically induced to farrowing, and between 1 and 8 parity order. The defatted colostrum samples were analysed by MALDI-ToF (19 compounds were detected).
The season of the farrowing and the reproductive performance data were recorded for each sow, including the number of alive piglets and the litter body weight at birth, at day 3 and at weaning, and corrected for the weight of the dead piglets. The litter weight gain (LWG) was then calculated. Four OS, including the bifidogenic sialyllactose, were detected in all the samples, one only in one sample, and one OS only in three samples. Among the 12 OS present in at least 50% of samples, in a 3-factor analysis of variance, the abundance values were affected by genotype (p<.05; 6 of 12), marginally affected by season (p<.10; 3 of 12) and never by parity order (1; 2; 3 or more). Abundance values of each OS component (16 of 20) were also standardised by Z-score scaling (within a component, μ=0 and SD =1), were then transformed by principal component analysis, and four similarity clusters were generated. Cluster membership, as an independent variable, was associated with LWG at 3 days (p=.063) and at weaning (p<.05), but not with piglet mortality within 3 days (in a linear model starting together with breed, season and number of pigs born). OS composition of colostrum may explain partially the variability within and between breed in the litter performance of sows. PCOS data base can be a further base for the development of new prebiotic products for the suckling and weaning pig.

P071

Effects of diet formulation on yield and nutritive value of *Hermetia illucens* prepupae intended for animal feeding

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The use of insect larvae as a feedstuff for aqua-feed represents a promising alternative, and it may be of particular interest obtaining insect-derived meals of desired characteristics starting from the rearing condition of the insects. The aim of this study was to evaluate how an experimental high protein diet (TMD3) (CP =13.8% DM; EE =4.0% DM; NDF =22.0% DM), including cereal co-products (55% wheat middlings; 30% alfalfa; 15% barley), can affect the survival rate, biomass increase and nutritive value of *Hermetia illucens* prepupae (HIP) intended for feed making. Three consecutive 21-days trials were performed in the same experimental conditions. Crude fat (EE), ash and crude protein (CP) contents of these HIPs were compared with those obtained, in the same growing conditions, by using a control diet (GHFD, 50% corn flour, 30% alfalfa; 20% wheat bran) (CP =10.6% DM; EE =4.2% DM; NDF =22.2 % DM) formulated by others. Both TMD3 and GHFD had similar gross energy content. Overall, the survival rate of larvae reared on TMD3 (67.5%) was lower than the one recorded for GHFD (83.8%) (p<.01). However, no significant difference was observed as far as the HIP fraction (71.2% vs. 63.7%, for TMD3 and GHFD) (p<.05). Neither total wet weighs (4.11 g vs. 4.21 g), nor total dry weights (1.35 g vs. 1.36 g) of HIPs reared on TMD3 and GHFD were found to be different (p<.05). The content of EE and CP were similar in HIP reared on both diets (32.50% DM and 50.63% DM for TMD3; 32.96% DM and 50.71% DM for GHFD). On the contrary, the ash content of HIPs obtained on TMD3 (10.9% DM) was significantly higher (p<.01) than the control (8.7% DM). Looking at the exhaust growing substrates, the pH of TMD3 (8.77) was significantly higher (p<.01) if compared to GHFD (7.25) suggesting that a more extensive deamination process took place in TMD3. Overall, the newly formulated, rich protein diet tested showed no improvements on the yield and compositional traits of *H. illucens* prepupae probably because the deamination of exceeding protein could have impaired their development. Further studies could help explain these preliminary results.

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P072

Iron supplementation of managed honey bee colonies as a sustainable strategy for preventing winter losses: a preliminary report

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Alimentary interventions can improve the resistance of managed colonies of *Apis mellifera* (HB) to maladies and parasites. The parasitic mite *Varroa destructor* (VM) can undermine the relationship between the effectiveness of social and individual immunity and the nutritional state of HB in several ways that can bring an HB colony to perish under winter conditions. In this preliminary work, the pre-wintering supplementation of divalent iron was studied because a literature survey suggests that it can

be helpful in reducing the winter loss of managed HB colonies. However, no data are available on the effect of iron supplementation in HB colonies on the parasitic pressure exerted by VM. Thus, the natural fall of VM was investigated as a possible explanation of the expected results. In September 2017, 16 healthy hives from two different apiaries located in the Lazio Region were selected and fed for four weeks with inverted sugar (IS) as a control (CON; 8 hives) or ferrous fumarate (FE; 8 hives) added to the IS to get a total dose of 1000 mg as Fe (II) per 5 kg of bees. During the supplementation phase (SUPP) and the following four weeks (POST), the fall of VMs from treated and control hives was bi-weekly monitored, counting separately mature (adult), damaged (as an indicator of grooming activity of the bees) and immature specimens (as an indicator of bees' hygienic behaviour against parasitised brood). In early April 2018, all the colonies were checked to assess the efficacy of the treatment. Treated and control hives showed no difference ($p<.05$) as far as the daily fall of mature (20.7 vs. 23.1 for FE and CON, respectively) and damaged VMs (0.8 vs. 0.7 for FE and CON, respectively) suggesting that the treatment gave no initial advantages in controlling
Q34 adult VMs. However, in the POST phase, supplemented hives showed a slight but significant ($p<.05$) higher daily fall of immature VMs in comparison to the control (1.2 vs. 0.5 for FE and CON, respectively) that can be regarded as a stimulatory effect of iron on the hygienic behaviour of worker HBs against parasitised/damaged brood. As far as the winter loss of HB colonies, different losses were observed between treated (12.5%) and controls (50.0%) (Odds ratio =7). These preliminary results suggest that Fe (II) supplementation may be a possible mitigation strategy of HB winter losses. Further studies are required to ascertain the linkage between treatment and reduction of parasitic pressure.

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P073

A novel by-product feed for lactating dairy sheep: effects on milk production traits and metabolic profile

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Cocoa husk (CH) is an important by-product derived from the processing of *Theobroma cacao L.* (Sterculiaceae), that represents an important source of income for a lot of tropical countries. CH is a good source of fibre (38–44%), protein (2.1–9.1%), carbohydrate (17.5–47%), lipid (0.6–4.7%) and mineral (K, Ca and Mn). Furthermore, CH contains a significant quantity of total phenolic compounds as epicatechin, catechin and procyanidins.

The aim of this study was to evaluate the effect of CH supply on milk production and composition, and on metabolic profile of dairy ewes. Twenty-four Sarda dairy ewes in middle lactation were allotted to 3 experimental groups. Each group was randomly assigned to one of three dietary treatments: a diet included a supplement of 100 g/d per head of soybean husks (CON), a diet included a supplement of 50 g/d per head of CH (CH50 diet), and a diet included a supplement of 100 g/d per head of CH (CH100 diet). Individual milk samples were collected weekly at morning and afternoon milking and the samples were analysed separately for fat, protein, casein, lactose, urea and for somatic cell count. Blood samples were collected at the beginning and at the end of the trial; the samples were analysed for principal biochemical parameters. All data were analysed as a completely randomised design with repeated measures using the PROC MIXED procedure of SAS version 9.2 (SAS Institute 2008). The inclusion of CH did not influence ($p>.05$) milk yield and principal milk components. Only the percentage of milk protein and casein was significantly affected ($p<.05$) by the interaction between treatment and sampling. In fact, at the end of the trial, the major percentage of protein milk was reached by animals fed with 100 g of CH per day (6.4%, 5.8% and 6.3% for CH100, CH50 and CON, respectively). During the experiment, the somatic count cells (SCC) increased significantly in CON ($p=.05$), whereas there were no differences in CH50 and CH100. As a regard to haematological parameters, most of them were not affected by treatments, except for lymphocytes (LYM) that were higher in CH50 and CH100 groups than CON ($p<.05$). The interaction between treatment and sampling was significant for some parameters, including basophil granulocytes, MHC, PLT and PCT.

In conclusion, the use of CH in ruminant nutrition is not having a negative effect on milk production and milk quality and it could be an alternative feed for animals.

P074

In vivo evaluation of tannin-based additives in weaned piglets

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Weaning is a critical phase that can lead young piglets more susceptible to enteric disorders and post-weaning diarrhoea. After the ban of antibiotics (EU Reg. 1831/2003) innovative alternatives to antibiotics for the control of enteric diseases are needed. Polyphenols, such as tannins from chestnut and quebracho, are characterised by antimicrobial and antiviral properties against post-weaning diarrhoea. The objective of this study was to evaluate the dietary effects of hydrolysable chestnut and quebracho tannins on growth performance, faecal bacterial count and faecal score of weaned piglets. A total of 108 piglets (Large White × Landrace), weaned at 30 ± 2 days, were randomly divided into four groups, two control (CTRL1, $n = 27$; CTRL2, $n = 27$) and two experimental (TAN3, $n = 27$; TAN4, $n = 27$). The groups were fed *ad libitum* the same diet that, for the TAN groups, was supplemented with 0.75% (as fed) of hydrolysable chestnut and quebracho tannins (Silvafeed Nutri P/ENC for Swine, Silvateam, Italy) from 0 to 42 days post weaning. Growth performances were evaluated and feed intake was measured weekly. A subset of animals ($n = 8$) for each group cohorts was individually scored for the consistency and colour of faeces, and faecal samples were collected on day 14, 28 and 42. A scale of four levels was used to score faecal consistency: 0 = normal (faeces firm and well formed), 1 = soft consistency (faeces soft and formed), 2 = mild diarrhoea (loose faeces, usually yellowish), 3 = severe diarrhoea (faeces watery and projectile). Faecal colour was evaluated using a three points scale: 0 = yellow, 1 = green, 2 = brown. Faecal coliforms, *Enterobacteriaceae* and *Escherichia coli* were enumerated using Violet Red Bile Glucose Agar (VRBGA), Violet Red Bile Agar (VRBA) and VRBA with MUG (4-methylumbelliferyl-ß-D-glucuronide) at the final concentration of 100 mg/L. Compared to CTRLs, the individual average body weight at 42 days of TAN groups was higher (17.60 ± 0.34 vs. 15.16 ± 0.34 kg; $p<.01$). Additionally, TANs showed higher average daily gain (from day 28 to day 42: 0.34 ± 0.02 vs. 0.25 ± 0.02 kg; $p<.05$), total feed intake and feed conversion ratio. The average faecal score of piglets of TAN3 and TAN4 was lower than CTRL1 and CTRL2 at 42 days ($p<.05$), even if no differences were observed from the microbiological evaluation. Concluding, tannins supplementation positively affected growth performance and could be an interesting alternative to antibiotics in livestock.

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P075

Fuel consumption for TMR preparation and distribution in different dairy farms

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The aim of the study was to evaluate the diesel fuel consumption (DFC) of a mixer wagon during feeds loading and mixing phases, and TMR distribution in dairy farms. In this work, the operating performance of a horizontal self-propelled TMR mixer wagon (Gulliver 6014, Sgariboldi, Codogno, Italy) with desilage conveyor arm, cutting slasher and mixer tank of 14 cubic metre capacity with paddle technology were analysed. The mixer wagon was equipped with a 100 kW turbo-diesel engine with a displacement of 3.6 L and it was characterised by hydrostatic transmission. Four different dairy farms located in the northern part of Italy (Lombardy) were involved in the study. All the farms had a corn and grass silage-based diet and a dietary content of hay from 2 to 16%. Fuel consumption was recorded as well as the duration of the different preparation and distribution phases, accounting for the amount of TMR produced in each farm and to the NDF content. Data were statistically analysed using simple linear regression.

The results showed a statistically significant correlation (R^2=0.90) between the amount of TMR produced and the consumption of diesel fuel. The relation between the NDF content of the TMR and the DFC had a coefficient of determination equal to 0.69, showing a higher consumption of fuel with the increase of the fibre contained in the ration. This is probably explainable with a greater absorption of power during loading due to a greater use of the cutting slasher to cut the fibrous parts. The relationship between increasing preparation and distribution times and fuel consumption showed, as expected, a high correlation (R^2=0.98). The hourly DFC of the three work phases analysed (loading, mixing and distribution) were similar over the four dairy farms involved in the study, with average values of 13.19 ± 0.33 kg/h, 13.75 ± 0.18 kg/h and 12.73 ± 0.47 kg/h, respectively. The average operating times measured were respectively of 25.06 ± 6.79 min for the loading phase (including the transfer time between the different feed storages in the dairy farm), 7.18 ± 1.39 min for mixing and 3.96 ± 0.9 min for the distribution, highlighting as the loading phase is the one requiring the most energy expenditure. This suggests the need – for the dairy farms – to optimise the internal logistics and the location of the feed storages.

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P076

Evolution of gastrointestinal microbiota structure in sheep: dietary and age effects

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The aim of this work was to study and describe the colonisation and evolution of microbiota from gastrointestinal tracts (GIT) in Sarda dairy sheep raised indoor from birth to 20 months of age (at middle of the first lactation). Since the day of 75 from conception, the sheep were divided into two groups and respectively exposed to two different dietary regimes. One group was fed a glucogenic diet with a high content of starch and sugars (starch) and the second was fed a lipogenic diet with a high content of high digestible fibre from soyhulls and beet pulp (fibre). Samples from different GIT of male lambs (rumen, ileum and rectum content) at birth and weaning were collected post-mortem. In addition, samples of rumen liquid and faeces were collected, *in vivo*, from female replacement lambs at birth, at weaning, at 6 months, gestation and first lactation phases. Samples were immediately frozen until DNA extraction, and microbial groups and structure were analysed by quantitative real-time PCR and DGGE. Metagenomic analyses of 16S-RNA were carried out on samples collected at birth and weaning, and are in progress for rest of the samples. Preliminary results showed that: – at birth (even before suckling) the GIT of lambs is characterised by a heterogeneous microbiota; – at weaning the microbiota is already structured, with developed mature microbial functions, that are dietary driven even maintaining the large individual differences observed at birth; – the adult-like composition of microbiota is reached around 1 year of age; – from prepuberal to first lactation the number of bacteria, protozoa and fungi increased significantly (p<.05). The number of fungi, *Fibrobacter succinogenes, Treponema saccharophilum* and *T. bryanthii* were higher in sheep Q35 exposed to fibre vs. starch diets (p<.05). The number of protozoa increased with age (p<.05) and tended to be higher in fibre fed ewes (p<.01). This work confirmed that diet was the first driver of the rumen microbial colonisation and its microbiota structure. However, during the last-trimester of pregnancy, the effect of diet on rumen microbiota was not significant, highlighting a possible host homeorhetic control on the microbiota structure. Further investigations should consider the effects of microbial structure and functionality on host performances.

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P077

Development and setting of a ventilated hood system to measure enteric methane emission from sheep

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Measuring methane (CH_4) is important to study the emission potential of ruminant feeds and diets. The ventilated hood head system, with respect to respiration chambers, allows to directly measure enteric CH_4 with easier animals' management. In this study, a ventilated hood system for CH_4 measurements in sheep was developed. The hood and the preliminary assessments to measure sheep CH_4 emission were described. The system, consisted of two parts: (i) a head hood (218 L volume), suspended on the front part of a metabolic cage for small ruminants, made by iron frame and transparent polycarbonate plates; in the front-bottom part a drawer allows to offer feed and water; the sheep, standing or laying in the cage, enters the head in the hood back, whereas the neck of the animal is tied with an impermeable airtight cloth; (ii) an air sampling system, consisting of air extraction and sampling from the hood; the air, drawn by a centrifugal fun and regulated by a manual valves system (3000 L/h), is first filtered, dried (silica gel) and sampled by a micropump (125 cc/min) in 10 L non-diffusing gas bags. Each bag-sample of air allows to accumulate samples from 1 h of emissions. Four Sarda dry ewes were allocated into two experimental groups fed *ad libitum* 2 different hays characterised by high and low NDF content (66.1 and 54.3% of NDF on DM basis for H-NDF and L-NDF, respectively). Both groups also received concentrate supply (0.63 kg/d of DM. Measurements lasted 12 days (1 sheep measured per day; 4 ewes ×3 replicates). The ewe was introduced in the hood at 23:00; the first 8 h were considered as adaptation period to the hood, whereas the other 16 h were used for the gas sampling. The daily ration of concentrate and hay was divided equally into three meals at 23:00 h, 7:00 h and 15:00 h. The CH_4 concentration of the sampled air was measured by a gas chromatographic method. Hay and total DM intakes resulted higher in the L-NDF than in the H-NDF (0.46 vs. 0.28 and 1.11 vs. 0.93 kg/d on DM basis; p<.05). Cumulative CH_4 daily emission was not different among groups (mean of 14.2 g/d) indicating that L-NDF animals emitted less CH_4 per kg of DM and NDF intake (15.22 vs. 12.55 g/kg of DMI, p<.05; and 53.49 vs. 41.86 g/kg of NDFI, p<.10). The hood allowed

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measuring the daily pattern and average values of methane emissions from sheep.

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P078

Selenium levels in sheep milk and blood from different flocks of Sardinia

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A survey was performed to evaluate the distribution of selenium (Se) concentration in milk and blood of sheep raised in different flocks of Sardinia. In 7 flocks located in the north and centre of Sardinia, serum and milk samples of adult ewes were collected and analysed for Se concentration during the year for every 6 weeks. Se concentration was measured in all dietary ingredients. Se intake from concentrate was calculated and Se from pasture has been estimated. Se concentration was determined by Inductively Coupled Plasma Mass Spectrometry (ICP-MS Perkin Elmer Nexion 300×). Se concentrations in milk and blood, and the average daily intake of Se, were analysed by GLM including in the model flock, sampling and their interaction. Correlation analysis was used to relate milk and blood Se and Se intake. The highest levels of Se concentration in milk were found during winter in correspondence of its supplementation in several farms, to avoid the white muscle dystrophy disease of suckling lambs. The levels of Se concentration in blood showed a peak in summer when diets of gestating ewes are characterised by the absence of green pasture. Weak relationship was found between Se in milk and Se in blood ($r = 0.21$; $p > .05$) and between estimated pasture Se intake and Se in milk. Positive relationship has been observed between Se in milk and Se intake from forage and concentrate ($r = 033$; $p < .05$). No relationship has been observed between Se in blood and Se intake. This is a preliminary study in Sardinia that investigates the relationship between diets and the presence of this important mineral in sheep milk and in blood.

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P079

Efficiency in sheep meat production: comparison between extensive and intensive management systems

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The aim of this study was to evaluate the effect of the suckler sheep breeding system (extensive vs. intensive) on performance of lambs. The trial, conducted in spring, was carried out on 30 Gentile di Puglia pregnant sheep with synchronised oestrus, raised under an extensive system. After delivery, the animals were divided into 2 groups (15 subjects for each group): one group raised under an extensive system (E) and the second group raised under an intensive system (I). Group I sheep, after an adaptation period of about 20 days, were fed *ad libitum* with excellent clover hay with an addition of barley to reach an average feed concentration of 0.7 units of forage (UF)/kg. In the initial phase, both the suckler sheep and the lambs showed a similar average weight: 42.1 and 41.8 kg for sheep, and 3.97 and 3.95 kg for lambs, E and I group, respectively. The lambs, born all from single birth, were fed with breast milk during the study period. On animals the following were determined: sheep live weight (LW; kg) at the beginning and end of the trial; lambs LW (kg) at birth and weekly; daily milk production (kg) using the double weighing technique, and milk composition (dry matter, protein, ash, fat and lactose); average daily gain (ADG; g/day) of lambs; *Biological Efficiency* (BE) which indicates the productive potential of the animal – g product/ kg LW; *Zootechnical Efficiency* (ZE) which provides elements on the energy cost for each unit produced – g product/ kg $LW^{0.75}$; feed conversion efficiency of lambs (g milk/g live weight gain). At 6 weeks of age, the lambs were slaughtered, hot carcase weight was recorded, and carcase yield was calculated. The statistical analysis was performed by ANOVA. The E group lambs showed a higher ADG (+34.4 g/day), BE (+2.7 g/kg LW), and ZE (+5.1 g/kg MW) than group I ones. ADG, BE and ZE were: 221.5 vs. 187.1 g/day, 29.0 vs. 26.3 g/kg LW and 47.7 and 42.6 g/kg MW for E and I group lambs, respectively ($p < .001$). E group lamb carcases showed a higher LW than those of I group (7.9 kg vs. 6.6 kg, respectively; $p < .001$), and thus a higher yield (66.0% vs. 61.6%; $p < .001$). This could be due to a biological limit to the growth rate, beyond which it is verified that the excess energy available is used for the fat synthesis. Thus, the results of this study showed that the sheep raised under an extensive system showed a higher production level which translates into an improvement in the *ante* and *post-mortem* performance of the lambs.

P080

Computerized management of cattle reared in an extensive system

Q36

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The innovations can be defined as such if they bring cost reductions and/or improvements in management and production efficiency. The problem that occurs in the extensive system, even more when grazing is spread over uneven surfaces or sprinkled with bushy or wooded areas, is the herd control. The aim of this study is the implementation of innovative information technology systems and digitisation of production processes, called ‘Boote System’, to reduce management costs, proportionally increasing the quality and sustainability of the production process and creating a territorial information network. The Boote System consists in the application to the animal of IoT hardware devices, allocated in the collars, which monitor in real time all bovine activities and some of its vital parameters, and by satellite control, they transmit the data (BIG DATA) to an IT management platform. This information within models specifically predisposed allows to define a series of behavioral-physiological aspects related to the single animal. The basic element detected instantly is the geolocation of the animal that provides information about the area covered per day and the relative transit speed. Moreover, the animal movement modalities allow to define elements related to pasture quality and hierarchical conditions in the herd, reproductive sphere and identification of the ‘delivery’ condition, and presence of predators. Results showed that the implementation of Boote System has reduced the need of personnel involved in breeding activities from ½ to ⅓ depending on the season of reference. In particular, the greatest reduction was observed in the phase in which the animals do not need milking or attention for the calves in the first phase of life. Furthermore, overall reproductive efficiency (number of calves per cow) has improved of about 10%; this is due both to the improvement of the conception phase (heat detection – coupling – conception) and to the control of the prepartum phase. The BIG DATA detected and collected by the IT platform through a food-blockchain allow the certification of both environmental sustainability of the process and total traceability of milk and/or meat.

P081

Study of a five sieves faecal particles separator to evaluate fibre and dry matter digestibility in dairy cattle

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The aim of the present study was to evaluate the undigestible neutral detergent fibre and dry matter (auNDFom and uDMom) in faecal particle fractions of lactating dairy cows. Faecal particles were stratified through a battery of five sieves with 4.6, 2.36, 1.18, 0.6 and 0.15 mm pore sizes. Four dairy farms located in the Parmigiano Reggiano cheesemaking area, feeding hay based total mixed ration (TMR), were involved. Five faecal samples were collected in each farm from 5 multiparous lactating Holstein cow of 60–90 days in milk (DIM). For each analysis, 250 g of faeces were weighted and put on the upper sieve, then washed with a water flow of 250 mL/s for 30 s. Washing was performed at each level of the sieves battery during sieves removal. Sieving was repeated three times for each sample. Residual material on each sieves was recovered, dried at 50 °C for 48 h and weighted. Triplicate of each sieve were pooled to carry out the *in vitro* digestion, for the determination of uDMom and auNDFom at 240 h of fermentation.

Statistical analysis was performed through the univariate procedure of the general linear model, using farm and sieves as fixed factors and cow as a random effect; *post-hoc* LSD test was performed.

Results showed that 2nd, 3rd and 4th sieves residues were similar for auNDFom proportion but differed from the 1st, 5th and escape residues – particles passing through the 5th sieve- (64.12, 67.55 and 64.18 %DM vs. 57.39, 55.77% and 16.90%, respectively; $p \leq .01$), while, considering the uDMom, the 3rd sieve residue showed the highest value ($p \leq .01$), being equal to 75.13 %DM. It appears that faecal particles having dimensions between 1.18 and 2.36 contained the lowest digestible dry matter residue, indicating the highest exploitation by the lactating cow, while this range is wider if auNDFom (%DM) is considered.

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P082

Assessment of livestock injuries in slaughterhouses and their economic impact in Argentina

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Meat slaughterhouses often receive animals with different muscle injuries which, after slaughter and during veterinary inspection, are confiscated. These muscular injuries are produced by blows and bruises suffered by animals, in the breeding establishment itself or during the sale of animals in livestock auctions. During transport, frequent injuries occur when loading and unloading animals on transfer trucks. They are influenced by the number of animals transported in each truck (density of load), the distance from the farms to the slaughterhouse. Other injuries occur in the slaughterhouse itself, during disembarkation and moving to rest pens and to the stun boxes. Acute muscle damage caused by blows has a strong negative impact. It affects the integrity and quality of commercial cuts. It obliges partial seizures of the injured area (of variable length) causing economic losses for the meat producer. The cattle that has suffered seizures during the Veterinary Inspection cannot be used for the international meat trade. The requirements established by the Animal Welfare standards of Argentine legislation require the application of Good Agricultural Practices (GAP) programmes to reduce the abuse and injuries that are frequently observed. From July 2016 to March 2018, a total of 2785 cattle were observed at a slaughterhouse located in Río Cuarto, Córdoba, Argentina. Acute lesions were observed in 52% (1448) of the animals. The average muscular confiscation was 550 grams per res, or 1 dollar per res, considering a mean value of 2 dollars per kilo, according to the meat market values.

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P083

Comparison of rumen fluid vs. faecal inoculum for the estimation of the uNDF fraction of feeds and faeces

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The accurate estimation of the undigestible NDF (uNDF) is important to assess forage quality and predict diet digestibility. *In vitro* uNDF is determined using ruminal inoculum, but also fresh faeces can be utilised as inoculum. The aim of this study was to compare rumen vs. faeces inocula to estimate uNDF concentration of 5 feed and 2 faeces samples.

The selected substrates were: barley meal, corn silage, alfalfa hay, lactating cow TMR, grass hay and 2 faecal samples collected from a dry and lactating cow. The inocula were prepared with (1) rumen liquor (R) collected from cannulated dry cows, (2) faeces collected from lactating cows fed a corn silage-based diet (F-CS) or (3) from lactating cows fed a hay-based diet (F-H). The concentration of uNDF was evaluated in 2 runs of incubation using the Daisy^{II} incubator and testing 2 incubation times (240 vs. 360 h). Each incubation jar was assigned to 1 source of inoculum. The inocula were renewed at 120 h and 240 h and 2 filter bags for each substrate and 2 blanks were removed at each time.

The results showed that uNDF content was affected by substrate, inoculum and incubation time ($p<.01$). The uNDF values were different among substrates ($p<.05$) except for barley meal and TMR. The faeces of dry cow had the highest uNDF value (408 g/kg DM), while the lower values were obtained for barley meal and TMR samples (74.9 g/kg DM and 83.4 g/kg DM, respectively). The values of uNDF (g/kg DM) obtained with the 3 inocula were significantly different (R: 212; F-CS: 241; F-H: 227; $p < .01$ for R vs. F-CS; $p=.02$ for R vs. F-H; $p=.02$ for F-CS vs. F-H), and the uNDF at 240 h of incubation (239 g/kg DM) was significantly higher than uNDF at 360 h (214 g/kg DM) ($p<.01$). uNDF decreased from 240 to 360 h more for F-CS (−15.6%) than for R (−9.00%) and F-H (−6.80%). Considering the results for each substrate, there were no uNDF differences at 240 h between R and F-H inocula. The inoculum F-CS at 240 h gave higher results ($p<.05$) for grass hay and corn silage, but the difference disappeared at 360 h. All linear regressions between R and the others inocula at 240 and 360 h resulted in high r^2 values (r^2=0.98 with R vs. F-CS at 240 h; r^2=0.996 with R vs. F-H at 240 h; r^2=0.98 with R vs. FC-CS at 360 h; r^2=0.994 with R vs. FC-H at 360 h). In conclusion, faecal inoculum seems to be a good alternative to rumen liquor for the estimation of uNDF on different samples. Further investigation is also required to estimate total tract digestibility.

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P084

Canonical discriminant analysis of body measures for the prediction of body weight in horses of different breed and morphology

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The estimation of horse body weight (BW) may represent an issue for the nutritionist when a scale is not available in stables. The adequacy of energy and nutrient content of the ration is calculated using BW as a variable. Modern horse breeds display huge morphological differences and alternative ways of BW prediction may display different extents of accuracy. This aspect still gathers the interest of the scientific community and prediction of BW by different means can lead to different outcomes. Also, body surface area (BSA) involved in different metabolic aspects for the sport horse covers an important role as to the nutritional management and body fluid distribution. Different scientific reports show complex calculations for BSA estimation based on morphometric measures, but the feasibility in field is low. In general, the more accurate BW estimation, the more reliable BSA. For this purpose, real BW (determined on digital scale) was comparatively examined with the estimated BW obtained by prediction equation (BWeq) and measuring tape (BWmt) for the horse. A total of 21 adult horses with a BCS of 4.5–5 (BCS 1–9 points scale) as pre-requisite for the enrolment in the trial, was weighed and assessed for the nutritional state. Different breeds were involved: French Trotter (2); Thoroughbred (4); Arabian (5); Saddle Italian (5); Saddle Belgian (3); Quarter horse (2). Two groups could be created as to meso-dolichomorph and dolichomorph breeds. Individual real BW was compared with estimated BWeq and BWmt by using a simple ANOVA model. Body measures used in the prediction equation for BW (wither's height, heart girth and fore-leg girth) were collected on each horse and used to develop a canonical discriminant analysis (CDA) to ascertain if those measures are able to discriminate the two groups.

Average BW, BWeq and BWmt displayed to statistically differ according to the morphology of animals ($p<.001$) but not as to methods of BW estimation ($p=.057$), in all cases being the prediction equation preferable to the measuring tape. In general, both estimation methods display to underestimate the real weight of the horse and this is more frequently observed in case of heavier horse breeds. CDA significantly separated the two groups and the best predicting measure appears to be the wither's height. In conclusion, other body measures than heart girth or fore-leg girth may be considered when the rear of the horse may contribute to the differences between real and estimated BW.

P085

Effects of cooling on postpartum dairy cows

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Different strategies can be adopted to alleviate the negative effect of heat stress on production performance of dairy cows. Our hypothesis was that cooling immediately after calving could improve the production performance of dairy cows by sustaining the homeorhetic shifts in metabolism for the onset of lactation, with an appropriate degree of inflammation. Forty-six multiparous lactating cows balanced for calving date, parity and mature-equivalent milk production during their previous lactation, were divided into two groups ($n = 23$); heat stressed (HS; parity =3 ± 0.12) and cooled (CL; parity =3 ± 0.25). Cows were housed in sand-bedded individual stalls equipped with misters and fans which were on from 1000 to 1800 h for CL group. Dry matter intake and milk yield were measured at calving until 7 weeks. Body Condition Score (BCS) and body weight (BW) were recorded weekly. Rectal temperature (RT) was measured daily at 14:00 h. Blood samples were collected weekly to measure the metabolic, oxidative and antioxidant status, the production of inflammatory cytokines and immunoglobulins. Mean daily maximum temperature, minimum relative humidity and maximum temperature-humidity index was 37.0 °C, 31.9% and 83.4 for HS and 27.3 °C, 44.9% and 75.7 for CL, respectively. HS cows showed higher RT (39.8 vs. 39.1 °C), lower feed intake (19.8 vs. 21.3 kg/d) and BCS than CL cows. Milk yield increased in CL cows in comparison with HS cows. The percentages of milk protein (3.25 vs. 3.06), lactose (4.73 vs. 4.58), and solids-not-fat (8.63 vs. 8.38) were higher in HS cows than CL cows. The metabolic status of CL cows displayed an increase in plasma concentrations of glucose, non-esterified fatty acids and β-hydroxybutyric acid in comparison to HS cows. A reduction of oxidative stress biomarkers concentration in plasma and a reduction of total antioxidant capacity was registered in CL cows. The immunological parameters, including plasma inflammatory cytokines (tumor necrosis factor-α, interleukin-1α and interleukin-2) and immunoglobulins, were higher in CL cows than in HS cows. Our data demonstrated that cooling lactating cows during early postpartum improved their productive performance, metabolic and oxidative status, and immune responses.

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P086

First semen cryobank of Italian rabbit breeds

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In Italy, there is an urgent need of conservation and safeguard programmes for Italian rabbit breeds in order to counteract biodiversity losses. Sperm cryopreservation is considered a secure method for the *ex situ* preservation of biodiversity facilitating the storage of their gametes in a gene bank. In this regard, thanks to the financed project Cun-Fu (MiPAAF 2016-2019) that aimed the safeguard and valorisation of Italian rabbit breeds, the first Italian semen cryobank has been created.
Semen cryopreservation activities within project have been facilitated thanks to the collaboration over the past several years between the University of Molise (Zoocolture laboratory) and Associazione Nazionale Coniglicoltori Italiani (ANCI-AIA). The University staff with the use of a mobile laboratory, were in charge for the semen cryopreservation. They moved in some Italian regions where events organised by ANCI i.e. local fairs and exhibitions of autochthonous rabbit occurred thanks to the participation of 'keeper's breeders'.
Males employed for the semen collection have been selected on sanitary status and specific morphological features. Semen samples were collected by an artificial vagina, the samples were then cooled at 5 °C for 90 min, then diluted to a ratio 1:1 (v:v) with an extender composed of Tris-citric acid-glucose (TCG) containing 16% DMSO and 0.1 M sucrose. The diluted semen was packaged into 0.25 mL plastic straws, equilibrated at 5 °C for 45 min. Lastly, semen was frozen by exposure at 5 cm above liquid nitrogen for 10 min, then was transferred into liquid nitrogen for storage at −196 °C. During the first year of the project, the semen from 114 rabbits of 26 breeds were collected and frozen, for a total of 431 doses. According to the Cun-Fu project goals, the number of doses for the Italian semen cryobank will be increased to preserve the autochthonous rabbit breeds.
In fact, this cryobank will play an important role i.e. to preserve resources with high genetic value to use in the medium or long term. In particular, these resources will be used: (1) to allow the backup of populations preserved *in vivo* in the event of genetic problems (inbreeding and genetic drift), to increase the effective population size; (2) to provide the possibility to reconstruct breeds in the event of extinction or drastic reduction of the population.

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P087

Characterisation of intestinal microbiome in overweight and non-overweight dogs of different breeds

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Obesity in dogs is as an emerging issue that affects canine health and well-being. Its development is attributed to several factors, including genetic predisposition and dietary management, and recent evidence suggests that the intestinal microbiota may be implied as well. Previous work has shown obesity to be associated with significant changes in the gut microbiota composition of humans and mice, but limited information is available about the role of the canine gut microflora.
The aim of this study was to investigate whether alterations in canine faecal microbiome may be influenced by breed and overweight condition. Eligible companion dogs were young adults, intact, healthy, fed mainly commercial extruded pet food, and had not to be receiving antibiotics, probiotics or immunosuppressant drugs in the previous six months. Labrador Retriever (LR) and Border Collies (BC) were chosen as reference breeds. Using 16S rRNA gene sequencing, the faeces of 15 lean (7 LR and 8 BC; BCS 4–5/9) and 14 overweight (8 LR and 6 BC; BCS >5/9) pet dogs were analysed. Moreover, for each dog, the daily intake of dietary protein, fat, carbohydrate, ash and fibre (g/d), and energy (kcal/d) were calculated according to an accurate feeding history collection.
Firmicutes and *Bacteroidetes* were the predominant phyla (51.5 ± 10.0% e 33.4 ± 8.5%, respectively), while the main classes resulted were *Bacteroidia* (33.3 ± 8.5%), *Clostridia* (29.9 ± 9.0%) and *Negativicutes* (13.7 ± 9.3%). The bioinformatic and statistical analysis of the microbiome composition revealed that no bacterial taxon differed significantly on the basis of body condition or breed ($p > 0.1$). No remarkable differences were recorded neither for the species richness in each sample (i.e. alpha diversity, p>.1) or for the divergence within the sample set (i.e. beta diversity, p>.05). However, there was a tendency for dogs to cluster together at a class level according to their lifestyle (living indoor vs.

outdoor). PERMANOVA test performed on single factors demonstrated the tendency of dietary protein to determine the recruited dogs' microbiota composition at operational taxonomic unit (OTU) level (p=.07) and order level (p=.1), while dietary ash might have played a role at family level (p=.09). Composition at OTU level appeared to be influenced also by daily energy intake (p=.08).

In conclusion, the faecal microbiome of dogs involved in this study showed no significant differences based on body condition or breed.

P088

Effect of crocin on post-thawed sperm quality in buffalo (*Bubalus bubalis*)

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Sperm cryopreservation offers great advantages for the application of reproductive technologies. However, cryopreservation determines considerable damages to motility apparatus, plasma membrane, acrosomal cap and DNA. Furthermore, buffalo sperm is more susceptible to hazards during freezing and thawing than cattle sperm. There is a strong evidence that antioxidants enhance sperm quality parameters maintaining the motility and the genetic integrity of sperm cells. Crocin is a water-soluble carotenoid and the chemical primarily responsible for the colour of saffron (*Crocus sativus*). Recent findings showed that crocin, under *in vitro* condition, improved sperm quality parameters preventing DNA fragmentation in bovine and red deer sperm. Therefore, the aim of this work was to evaluate the effect of crocin on motility, viability, membrane integrity and DNA fragmentation of buffalo frozen/thawed sperm. Frozen-thawed sperm from four different Mediterranean buffalo bulls (2 replicates each) were thawed at 37 °C for 40 s, separated by Percoll, and incubated in Talp medium with 0 (control), 0.5, 1 and 2 mM crocin for 2 h at 5% CO_2 in humidified air. After incubation, sperm motility was examined by phase contrast microscopy at 40× magnification on thermo-regulated stage at 37 °C while sperm viability and membrane integrity were assessed respectively by Trypan Blue/Giemsa and HOS test. DNA fragmentation was evaluated by TUNEL staining. Data were analysed by ANOVA. The incubation with 1 mM crocin increased (p<.05) sperm motility (65.0 ± 0.2, 67.5 ± 0.2, 73.8 ± 0.1 and 65.0 ± 0.5, respectively with 0, 0.5, 1 and 2 mM crocin) and viability (80.7 ± 1.8, 81.6 ± 2.2 86.8 ± 1.4 and 85.3 ± 1, respectively with 0, 0.5, 1 and 2 mM crocin) compared to the control. However, crocin did not affect sperm membrane integrity (56.3 ± 1.7, 56.4 ± 3.5, 60.1 ± 2.7 and 62.0 ± 2.1, respectively with 0, 0.5, 1 and 2 mM crocin). Interestingly, the percentage of sperm exhibiting DNA fragmentation significantly decreased at the highest concentration tested (11.0 ± 1.9, 12.6 ± 1.5, 14.5 ± 1.1 and 5.4 ± 0.9, respectively with 0, 0.5, 1 and 2 mM crocin; p<.05). In conclusion, these preliminary results indicated that crocin improved the quality of buffalo frozen-thawed sperm and the incubation with 2 mM crocin was also effective in reducing sperm DNA fragmentation. However, further investigations on fertilising ability are required to draw definite conclusions.

P089

Effects of diet supplementation with *Trichoderma spp.* on structure and ultrastructure of gastric tissues in buffaloes

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The effects of supplementing buffalo young bulls with a fibrolytic enzymes mixture derived from *Trichoderma harzianum* strain T22 on morphostructural and ultrastructural features of the pre-stomachs and abomasum were investigated. Sixteen intact Mediterranean Italian buffalo males (initial body weight 88.2 ± 7.7 kg) were divided into two homogeneous groups (Control and Treatment) and fed the same maize-silage based total mixed ration (TMR). The treatment consisted of spraying the TMR with the liquid fibrolytic enzymes mixture at the dose of 0.7 mL/kg of dry matter. Enzyme mixture was prepared weekly and it was applied to TMR, immediately after unloading, diluted in water (1:5 ratio vol/vol). The animals were slaughtered at 350 kg body weight and, for each of them, six samples from rumen, six from reticulum, three from omasum, and five from abomasum were collected. All samples were processed in order to perform scanning electron microscopy (SEM) and transmission electron microscopy (TEM) investigations. The results are a morphostructural description, so that no statistical analysis was performed. Image analysis allowed to highlight some general trends on morphofunctional changes determined by the treatment with *Trichoderma*. Treated animals showed apparent higher reticular mucosa crests, an apparent increment in number and size of the ruminal papillae, and of the abomasal laminae. Moreover, treatment induced more vascularisation of the superficial layers of the *lamina propria*, with a greater presence of dilated loops at the abomasal level, as well as an apparent greater number of glands in the different abomasal districts and a widening of the exchange surface between the mucosa and nearby structures. Enhancement of *lamina propria* vascularisation can improve absorption through the organ wall, as well as it can play an important role in the trophism of mucous membranes. Finally, it was highlighted in both pre-stomachs and abomasum districts a better development of the muscularis mucosae involved in the peristaltic movements. The results of this study showed that fibrolytic enzymes of *T. harzianum* strain T22 can promote a better growth of both pre-stomachs and abomasum of buffalo young bulls.

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P090

Ionomic profile of hair of growing and adult dogs fed different diets

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The aim of the study was to investigate element concentrations in the hair of growing and adult dogs. Overall, 39 dogs were recruited in a kennel and were divided into 4 groups: G1, 10 puppies of small size breeds; G2, 6 puppies of medium size breed; G3, 11 adult dogs of medium size breeds; G4, 12 adult dogs of toy breeds. The dogs of each group were fed 4 different complete diets, according to the respective nutrient requirements. Hairs were sampled with a scissor from the medial part of the left leg of each dog, taking care to cut as closer to the skin as possible. The same area was sampled with the same procedure after 60 days (D60). The D0 hairs were sampled on the 3 June and the D60 on the 2 August. Just after the collections, the samples were inserted in a paper envelope and stored until element analysis. A sample of 200 mg for each complete diet and hair was mineralised at 180 °C for 10 min in a microwave with a Teflon digestion tube with a solution composed by 1 mL of H_2O_2 at 30% (v/v) and 9 mL of HNO_3 at 65% (v/v). Liquid samples were filtered with PTFE filter with pores of 0.45 µm of diameter. Samples were analysed for the elemental contents using an inductively coupled plasma optical emission spectroscopy (ICP-OES) with detection limit <1 ppm. The adult dogs of the G3 group always showed in the hair the lowest concentrations of Al, Fe, Mg and Mn ($p<.05$) and Cu and Zn ($p<.01$), whilst the hairs of puppies fed G1 and G2 diets had a significant higher content of Al, Cu, Fe and Mn. Hair concentrations K, Li and Na were higher at D60 in comparison with D0 ($p<.01$). Principal component analysis on the data of D60 explained 86.2% for the first 3 components and clearly indicated separated clusters for Li, Na, K and for Mg, Ca, Sr, Ba. Correlations between concentrations in the hair of Al and Fe was highly significant ($p<.01$; $r=0.922$). The results did not support the use of elements in the hair to predict nutritional supply, but they seemed to more useful to assess health conditions and environmental exposure to toxic elements.

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P091

Effects of dietary supplementation with *Pinus taeda* hydrolized lignin on rumen microbiome in beef

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The aim of this study was to evaluate the *in vivo* effect of a commercially available herbal preparation (Oxyphenol®, I-Green, Padua, Italy) from *Pinus taeda* hydrolysed lignin (PTHL) on fattening beef rumen microbiota. Forty Limousine steers were randomly divided into two groups of twenty individuals each, reared in two separated pens. The two groups were fed with the same ratio, except for the supplementation with PTHL (35 g/day per head for the first 90 days of the trial and 70 g/day per head for the last 30 days) of one of the two groups (TREAT). The other group (CON) did not receive any supplement. Rumen fluid was collected, at each sampling time (day 0, before starting the trial, at 60 and 120 days after starting the inclusion of the supplement) from 10 animals for each group, randomly chosen, using an oesophageal tube under mild vacuum from the reticulum near the reticulo-omasal orifice. Samples of ruminal fluid (filtered through eight layers of gauze cloth) was collected in thermos flasks (previously filled with distilled water at 39 °C to avoid thermal shock to rumen fluid), insufflating in the headspace CO_2 to ensure anaerobic environment, and moved within 30 min to the laboratory where they have been processed and stored at −80 °C until metagenome sequencing. Analysis of rumen microbiota was carried out by 16S, 18S and ITS1 sequencing. After quality filtering and chimaera detection, reads from 16S and 18S sequencing were aligned against SILVA ribosomal RNA sequence database (https://www.arb-silva.de/) using MALT, while pre-processed ITS1 reads were aligned against UNITE fungal ITS sequence database (https://unite.ut.ee/) using BLAST. Taxonomic binning was performed with MEGAN Community Edition software v6.12. The data Q37 set was subjected to two-way ANOVA, where sampling time and feeding group were set as independent variables. Values were given as mean values and standard error of the means (SEM) of

the number of reads. When a significant effect ($p<.05$) was detected, means were compared using the Tukey's test for repeated measures. Yeasts concentration was not affected by the PTHL addition in the feed ($p>.05$). Differently, protozoa and methanogenic organisms showed a reduction in their number ($p<.001$). These observations need to be implemented with results about *in vitro* fermentation, gas production and apparent digestibility, in order to understand if there is a variation in animal performance and greenhouses gas production.

P092

Occurrence of gluten in canned and dry grain-free commercial pet food

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In recent decades, the anthropomorphism of companion animals has led to an increased inclusion into family units such that they are viewed as family members. The pet food manufacturers have responded to this culture shift with trendy diets that questionably align with animal health and physiology. They include the grain-free diet as one of the most well-known and advertise. Industries usually claim these products as high protein 'ancestral diets' considered healthier than their grain inclusive counterparts, mainly represented by gluten. However, confirmed reports of gluten-sensitive enteropathy (GSE) are rare in dogs and proved only in certain breeds (i.e. Irish setters, Border terriers). Moreover, establishing a direct cause and effect relationship between the intake of a dietary component and a disease is challenging, not least because pet food is a complex mixture of nutrients which may interact with each other, but also because of potential cross-contamination of proteins from origins not mentioned on the label (i.e. wheat or other cereals). Given the above, the aim of the present study was to ascertain the absence of gluten residues in pet food samples marked as 'grain-free' and 'gluten-free' diets, to assess the reliability of manufacturer labelling. Fifteen commercial diets labelled as grain- or gluten-free and 2 commercial diets containing wheat were sampled. An analytical procedure was developed and applied to determine specific markers of wheat gluten, through liquid chromatography interfaced with mass spectrometry with high power of resolution. The results are expressed as milligrams of wheat flour type '00' present in 1 g of feed. The quantification limit (LOQ) obtained in the wheat flour for ion m/z 894.5043, $z = 2$, is 4 mg of wheat flour per gram of feed, that is, weighing fewer grams of wheat flour, the quantifier ion is not detectable. Taking into account this calibration line and the LOQ obtained, the samples were prepared and analysed using the HPLC-HRMS method developed. Quantifier ion in 14 out of 15 samples from grain- or gluten-free diet was < LOQ, meaning that gluten residues from wheat or other cereals were not detectable, while in 1 out of 15 samples analysed, 10 mg of wheat flour/g feed were measured (in control samples of 50.3 and 53.4 mg of wheat flour/g feed, respectively).

P093

Establishment of inflammatory *in vitro* intestinal epithelial models for translational animal nutrition

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Many naturally available compounds as n3-polyunsaturated fatty acids (EPA: Eicosapentaenoic acidand DHA: Docosahexaenoic acid), conjugated linoleic acid, milk exosomes and plant extract from *Macleaya cordata* exhibits anti-inflammatory effects. From previous studies, these bioactive compounds demonstrated a multitude of beneficiary effects in both human and animal health and are considered as potential therapeutic agents with pharmaceutical properties. Due to their health benefits, new ways to incorporate them in human diet through poultry and livestock nutrition is extensively studied and therefore, it is first important to determine its anti-inflammatory effects in cell-based inflammatory models. The gastrointestinal tract (GI) is the first site where food is broken down and nutrients are absorbed and therefore the GI cell models are widely preferred for food/feed analysis. In this respect, it becomes of paramount importance to establish inflammatory cell line models of intestinal epithelia. Therefore, in the present study, we demonstrated the inflammatory response of IPEC-J2 cell lines of neonate porcine intestinal epithelium challenged against different stimuli as cell wall lipopolysaccharides (LPS) of Gram-negative bacteria as *Escherichia coli* and *Salmonella*, and chemical such as dextran sodium sulphate (DSS), analysed by MTT cell viability assay. The cells were treated with each stimulus in a dose-dependent manner (0.15–10% for DSS, 1.56–100 µg/mL for LPS) for 24 h and thereafter viability was measured and the concentration at 50% inhibition (IC50) was calculated using regression analysis. The IPEC-J2 cells exhibited an IC50 value of 2.89% for DSS challenge and 12.77 µg/mL for *Salmonella* LPS. The *E. coli* did not show any significant

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inflammatory response even with the challenge of highest dose as 100 µg/mL. These results suggest that the epithelial cells are specific for different biological challenge as bacterial LPS and the DSS chemical proves to be potent inflammatory agent even at small doses and can be effectively used to induce inflammatory response to study anti-inflammatory properties of food/feed additives.

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P094

NEFA, BHBa and γ-GT variation in the blood serum of weaned foals up to 18 months of age

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The correct energy balance estimation of the growing foal may represent an issue for the nutritionist, given the dynamic change in body composition and fat tissue depots. In fact, the maintenance of adequate body weight and body measures over time for optimal growth curve accomplishment and the fulfilment of nutrient requirements are fundamental to address adequate individual feeding plans of the future sport horse. The metabolic profile of selected metabolites could be helpful in interpreting growth homeostasis and some of these could be used as indicators of energy balance state and liver health. Against this background, the literature on circulating indicators of homeostasis perturbation during the growth of foals is poor. We hypothesised that metabolites indicating the energy balance like non-esterified fatty acids (NEFA), β-hydroxy-butyric acid (BHBa) and liver enzyme like γ-glutamyl-transferase (γ-GT) [interpreted in the light of circulating total bilirubin (TBIL) and aspartate aminotransferase (AST)] may be used to monitor the growing foals from weaning to 18 months of age. For this purpose, a total of 12 Anglo-Arab (AA) foals from the same stable were enrolled in this trial. All foals were serially weighed on a digital scale and sampled for total blood at weaning, at 12- and 18-months of age. Feeding and rearing conditions were adopted simultaneously equal for all the foals used. All data were processed by a repeated measure ANOVA. The foals involved appeared outwardly healthy and no signs of poor growth performance were pointed out as to body and size gain for the breed. However, the study of the metabolic profile gave rise to interesting patterns of parameters investigated. In fact, NEFA turned out to increase sharply in 12-months-old foals, when animals were sent to spring pasture, but BHBa displayed to increase when foals were aged 18 months and NEFA concomitantly decreased to reach similar levels to those found when foals were stabled during weaning. In addition, γ-GT and BHBa levels turned out to positively correlate ($p = 0.051$). While at 6- and 12-months, γ-GT dropped in the physiological reference range for the horse, at 18-months of age γ-GT levels exceeded the upper limit, when also BHBa tested to increase.

In conclusion, the liver enzyme increase could be associated with ketone bodies production for energy purposes when energy balance appears negative, following the drop of NEFA from body depots.

P095

Effect of fibre length, and amount on growth and behaviour of buffalo calves

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The objective of this study was to determine the effect of two different physical forms of forage, long and chopped, on the performance and feeding behaviour of young buffalo calves. Twenty-four Mediterranean buffalo calves were randomly assigned to 3 feeding treatments (2 males and 6 females/group). All the calves received daily 4 L of milk replacer (MR: 0.18 kg/L). From the 15th day of life, calves from Group LH received long hay (20 cm) and commercial pelleted starter *ad libitum*; calves from Group OS received only commercial pelleted starter *ad libitum* and calves from Group CH received chopped hay (3–4 cm) and commercial pelleted starter *ad libitum*. Food and water intake were recorded weakly while the calves were weighted twice a month. At weaning (91 days of age), 2 males of each group were slaughtered and the stomachs were removed and measured. Samples of feed were taken and analysed for chemical composition. The remaining calves were weighed again 85 days after weaning (173 days of age). Calves' behaviour was recorded in sessions of 1 min/animal, for 1 h/day (totally 3 min/day), 2 times per week during the last 2 weeks of pre-weaning. The total observation time per animal was of 12 min. Starter intake was higher in group OS ($p<.05$), whereas the hay intake was higher in group CH ($p<.05$) as compared with the other two groups. Body weight gain was not different between treatments at the weaning, whereas the weight of rumen, reticulum, omasum and abomasum was higher in group CH than in groups LH and OS ($p<.05$). Unexpectedly, 85 days after weaning the body weight of calves from Group CH was lower than the body weight of calves from Group OS ($p<.001$). Calves fed CH spent more time eating chopped hay ($p<.001$) and consequently ate less starter ($p<.05$) than the other two groups. Group CH spent and tended to spend less time performing non-nutritive oral behaviours than calves from Group LH ($p<.05$) and Group OS, respectively. Calves from Group OS spent more time standing than animals from Groups LH and CH ($p<.05$). In conclusion, the provision of chopped hay to young buffalo calves increased the stomach dimension and reduced non-nutritive oral behaviours as compared with offering long hay or only starter. On the other hand, calves from Group CH had a reduced growth as a possible consequence of the higher amount of time spent eating chopped hay than the starter. Conversely, the best growth performances were observed in Group OS where the calves only ate starter.

ANIMAL PRODUCTS

P096

Donkey cheese production using pure milk or in mixture with goat or cow milk: preliminary results

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Compared to conventional dairy species, the processing of donkey milk into cheese is very difficult due to its intrinsic chemical characteristics related to low contents in caseins and fat that determine poor clotting activity, and only a few researches have been made on cheese production. The aim of this study was to examine some cheesemaking parameters of cheese elaborated via appropriate technology using donkey milk alone or fortified with goat or cow milk. Three cheesemaking sessions on different days were performed in mini-vat heated by thermostatic water bath using fresh milk. In each trial, donkey milk and two mixtures (70/30, v/v) of donkey and goat milk, and donkey and cow milk were processed. After cooling, calcium chloride was added in milk and commercial starter cultures were inoculated. When the milk, maintained in a thermostatic water bath, reached the pH 6.3, liquid microbial rennet was added. The milk was then allowed to coagulate, and the coagulation behaviour was monitored by observation of milk clotting. After gel formation, the curd was cut, the whey was drained, and the obtained curds were placed into cheese moulds. The weight of cheeses was assessed, and the yields were calculated. Cheesemaking parameters were affected by the kind of milk, as pure donkey milk or in mixtures. Compared with the two mixtures, pure donkey milk exhibited longer time for gel formation (20 min) and for total in-vat working (170 min), and showed a visive weaker gel, highlighting a specific behaviour during cheesemaking. The cheese yield was 7.2% for donkey milk, 10.2% for the mixture of donkey and goat milk, and 11.4% for the mixture of donkey and cow milk. On the basis of visive evaluation, the cheese from pure donkey milk showed a more soluble texture and grainy structure compared to the cheeses from the milk mixtures. This preliminary study may be useful for further studies and potential industrial applications for the development of innovative kind of cheeses.

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P097

Effect of two sous-vide cooking methods on fatty acid composition and oxidative stability of longissimus thoracis muscle from pigs receiving a diet containing or not extruded linseed

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To verify the effects of two different sous-vide cooking conditions on lipid oxidation and fatty acid (FA) composition of longissimus thoracis (LT) muscle, 24 pigs, evenly divided into two groups of 12 subjects each, were used. One group received a barley-soya bean meal diet (C) and the second was given the same feed where 5% of extruded linseed partly replaced barley, to obtain a n-3 FA enriched diet (L). At slaughter, from each left half carcase, two samples of LT muscle were collected, packed under vacuum and stored at −18 °C until analysis. The samples were cooked in water bath according to two different methods: at high temperature (80 °C) and short-time i.e. samples left until the core temperature

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reached 70 °C (A); at low temperature (60 °C) and long-time (15 h) (B). After cooking, the samples were refrigerated (2 °C) for 24 h. Oxidative stability was measured by a dosage of the 2-thiobarbituric acid reactive substances (TBARS) content, expressed as milligrams of malondialdehyde (MDA)/kg of meat, and FA composition was determined by capillary gas chromatography. The content of each fatty acid is expressed as a percentage of the total FAs detected. The statistical analysis was performed by means of ANOVA, using the GLM procedure of SAS. Dietary treatment (C vs. L) and cooking condition (A vs. B) were used as independent variables. The different sous-vide cooking conditions affected neither lipid oxidation nor FAs percentage of LT muscle. Extruded linseed feeding brought about an increase of the percentage of total n–3 FA (2.67 vs. 0.98; $p<.01$) and also of polyunsaturated fatty acids (12.02 vs. 9.68; $p<.01$) in intramuscular fat but did not affect lipid oxidation. This enabled to obtain pork with a more favourable n–6/n–3 ratio (3.68 vs. 10.42 in L and C group, respectively; $p<.01$), according to the global health guidelines. Thus, an enriched linseed diet ameliorates the FA composition of pork. The effect of the two different sous-vide cooking methods on lipid composition and oxidative stability of pork does not differ, irrespective of dietary treatments.

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P098

Dietary tomato pomace: effects on pork quality

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Sixteen Nero Siciliano male pigs (initial body weight: 42.7 ± 2.31 [SD] kg) were assigned to two feeding treatments and fed for 88 days with: a commercial concentrate diet based on corn-barley (CON; $n = 8$), or a concentrate diet including 15% of dry tomato pomace (DTP) to partially replace corn (DTP; $n = 8$). Pigs were allocated in individual pens and fed *ad libitum* with the respectively experimental diet. At the end of the trial, animals were transferred to a commercial slaughterhouse and slaughtered by electrically stunned and exsanguinated. After 24 h at 4 °C, the longissimus dorsi muscle (7th–11th rib) was removed from each carcase. Then, muscle samples were divided into two aliquots. One aliquot was immediately vacuum-packaged and stored at −30 °C, until analyses. The remaining aliquot was divided into 3 slices (2 cm thick) and stored at 4 °C for 0 (3 h), 3 and 5 days to determinate colour and lipid stability.

Final body weight, carcase weight and carcase yield were not affected by the experimental treatment. Intramuscular fat, sum of saturated and monounsaturated fatty acids were greater in the meat of CON pigs as compared to DTP meat ($p<.05$). Palmitic (C16:0), stearic (C18:0) and oleic (C18:1 c9) acids were significantly lower in meat from DTP animals than CON pigs ($p<.05$). The sum of polyunsaturated fatty acids was not affected by dietary treatment. Retinol content was 4.7-fold greater ($p<.001$) in the muscle of DTP than CON pigs. While, α-, γ- and δ-tocopherol levels were not different ($p<.05$) between the two diets.

Meat lightness (L*) was lower ($p=.037$) in the meat of DTP, while yellowness index (b*) only tended to be lower ($p=.087$) as compared to CON group. The redness index (a*), chroma (C*) and hue angle (H*) were comparable between treatments. The lipid oxidation (TBARS) was not significantly different between the groups but tended to increase over time of storage ($p=.060$). In addition, a significant interaction between dietary treatment and time of storage (D × T) was recorded for the same parameter ($p=.018$). In the present study, the inclusion of tomato pomace in pig feeding allowed to obtain improvements on meat quality, in terms of intramuscular fat and vitamin A content, without compromising the animal performance and meat shelf-life.

P099

Effects of sodium nitrite replacement with natural antioxidants on aromatic profile and consumer acceptance of Cinta Senese dry-fermented sausages

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A mixture of natural antioxidants made of grape seed extract, olive pomace hydroxytyrosol and tocopherol (GSE) was tested as sodium nitrite (NIT) replacement for Cinta Senese dry-fermented sausages. The effects on aromatic profile and sensory traits were assessed by SPMS-GC coupled with GC-Olfactometry technique, whereas the consumer's acceptability was assessed through a consumer test on 120 potential consumers. The differences observed for the volatile profile were mainly related to the abundance of the single compounds. Considering the volatile compounds grouped for their aromatic notes, the olfactometric analysis showed a higher incidence of fruity-sweet and roasted-nutty notes in NIT samples than in GSE. However, the hedonic evaluation, performed by a blind test using a scale from 1 (extremely disliked) to 9 (extremely liked), did not show any statistical difference between the two products (6.60 for NIT and 6.58 for GSE). Moreover, the consumer's purchase intention was evaluated before and after the products' tasting. The purchase intention was confirmed for GSE products, while it decreased for NIT sausages. This suggests that consumers that have chosen the GSE product, were more interested in the health-related attributes, while the ones that have chosen the NIT product were more interested in the sensory-related attributes. Likely, the latter group was the most susceptible to change opinion after the sensory evaluation. In conclusion, the sodium nitrite replacement by natural antioxidants did not affect the overall aroma profile and the consumer's acceptability and might be an interesting strategy to enhance the healthiness of this type of Cinta Senese product.

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P100

Evaluation of the chemical-nutritional parameters and the aromatic profile of eggs obtained from laying hens fed with a dietary supplementation of extruded linseed

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Hen eggs represent a traditional food with an excellent nutritive value due to the presence of highly digestible proteins, vitamins, minerals and lipids, such as polyunsaturated fatty acids (PUFAs). Lipid composition of hen eggs is a subject of primary consumer concern, due to the relationship between specific dietary lipids and the development of coronary heart diseases (CHD). Nowadays, it is well known that ω-3 PUFAs provide important health benefits to humans as prevention and treatment of many chronic diseases. The most significant ω-3 PUFAs appear to be α-linolenic acid (ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) and for the mentioned reasons over the course of time a growing interest has been developed in the production of eggs rich in ω-3 fatty acids, by feeding laying hens with experimental feedstuffs containing these nutrients. For example, ALA is predominantly in seed oils such as flax (*Linum usitatissimum*). Linseed is unique among oilseeds because of their exceptionally high content of ALA ($\cong$50% of the total oilseed).

The aim of this work was to determine the effect of the hens diet integration with extruded flaxseed (7%) on productive parameters and on the quality of resultant Bio-ω-3 eggs.

At the end of the treatment, no significant difference was observed for eggs production while it was observed for the average egg weight (58.05 ± 1.94% for control eggs vs. 63.37 ± 2.14% for flax eggs). Regarding, instead, the chemical-nutritional parameters, significant differences were not observed for total lipids and in the TBARs-test while significant differences were observed in the acidic profile. Specifically, ω-3 PUFAs were higher in flax eggs ($p < .01$) while SFA were higher in control eggs ($p < .01$). Also, β-carotene was found higher in flax eggs (478.20 ± 15.19 μg/g vs. 324.80 ± 13.84 μg/g, $p < .001$). The aromatic profile was, also, analysed and significant differences were observed both for the alcohols and aldehydes compounds ($p < .05$). Finally, a significant difference in the colour was observed between the two types of eggs ($\Delta E_{ab}=1.77 \pm 0.23$, $p < .05$).

In conclusion, it is possible to assert that the integration of the laying hens diet gave positive results as it has not negatively affected the production parameters, has improved the ω-6/ω–3 ratio (5.9:1 for flax eggs vs. 62:1 for control eggs) and the β-carotene content, and has decreased the percentage of SFA guilty of cardiovascular pathologies.

P101

Effect of dried liquorice root supplementation on chemical-nutritional quality of dairy products obtained from goats

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Enriching the goat's diet with plant extracts, rich in bioactive compounds, allow to improve the chemical-nutritional properties of dairy products. Liquorice has anti-inflammatory, antimicrobial, antioxidative effects associated with the presence of compounds, as triterpene saponins, flavonoids and coumarins.

The aim of the present work was to evaluate the effect of dietary integration with 8% of dried liquorice root on chemical-nutritional quality of goat milk and cheeses. The study was conducted for 60 days on Saanen goats divided into two groups: a control group (CG) and an experimental group (LG+) whose diet was supplemented with liquorice. Milk samples were collected to determine chemical-nutritional composition and fatty acids profile (FA) at 30 and 60 days from the beginning of the study. At the end of the experiment, cheeses produced with CG and LG + bulk milk were analysed for chemical–physical parameters at 3 (T3) and 60 (T60) days of ripening.

A different FA profile and a significant increase in proteins ($p<.01$) and casein ($p<.01$) percentage but no variation in lipids, lactose, urea and in FA were observed in LG + milk samples respect to CG milk. Regarding cheeses, no variation in lipid and protein but a lower water content ($p<.05$) were found in LG + T3 and T60 cheeses, this reflected in a different texture. Indeed, the LG + cheeses were harder, more elastic and more gummy than the CG cheeses and these differences were observed both in fresh and aged cheeses. Moreover, the LG + cheeses showed a yellowish colour probably correlated to the presence in LG + milk of specific compounds of liquorice roots, such as tannins, carotenoids and ascorbic acid which have antioxidant activity and are also responsible for the yellow colour. A greater presence of antioxidant compounds deriving from liquorice roots can be also correlated with greater oxidative stability found in both LG + T3 ($p<.05$) and T30 ($p<.05$) cheeses. Different families of volatile compounds were detected in T30 cheeses obtained from the two groups. A significant reduction of octanoic acid ($p<.05$) and a significant increase in nonanal ($p<.01$) were found in LG + T3 cheeses, instead in LG + T60 cheeses, significant increases of 3-methyl, 1-butanol ($p<.01$) and acetoin ($p<.05$) have been found.

In conclusion, it is possible to assert that the integration with liquorice has modified chemical and technological properties of goat cheeses, improving the oxidative stability and inducing changes in texture and colour.

P102

Effects of polyphenols and algae supplement on rabbit meat quality

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Recently, natural extract has been studied as dietary supplement in animal feed for its antioxidant, anti-inflammatory, antiviral and antimicrobial properties. There is a growing interest on the use of natural supplements in rabbit nutrition in order to enhance meat quality, improving oxidative stability and nutritional parameters. Moreover, these nutritional strategies are able to support growth performances in rabbit antibiotic-free production, which is gaining increasing interest by producers and consumers. The aim of the study was to evaluate the effect of a dietary supplementation with natural extract mixture in growing male rabbit on *Semimembranosus* muscle quality parameters. One hundred and forty-four New Zealand White rabbits were housed in an antibiotic-free production system for 42 days and divided into 3 experimental groups (4 rabbits/cage) from weaning (35 days old) to slaughter. The first group fed a basal diet (C), the second (T1) and the third one (T2) received 0.3% and 0.6% of natural extract mixture containing polyphenols and brown seaweeds. At slaughter (average live weight 2.89 ± 0.33 kg) 10 carcases per groups were randomly selected, frozen and sent to lab for determination of meat quality and sensory parameters. Right thighs were selected and chemical parameters, pH, colour parameters, Vitamin E content, oxidative stability and sensory analyses were performed. Data on meat quality were analysed by one-way ANOVA and sensory parameters were analysed using three-way ANOVA. Physical and chemical parameters of *Semimembranosus* muscle did not differ ($p>.05$) among experimental groups. Vitamin E content was improved by dietary treatment (0.130 ± 0.001 mg/100 g C; 0.174 ± 0.001 mg/100 g T1; 0.164 ± 0.001 mg/100 g in T2; $p<.001$). However, oxidative stability was not affected by dietary treatments (0.136 ± 0.002 mg/kg C; 0.133 ± 0.001 mg/kg T1; 0.131 ± 0.002 mg/kg MDA in T2; $p>.05$). Sensory evaluation revealed that dietary supplementation with polyphenols and brown seaweed improved ($p<.05$) meat texture parameters and aroma. Overall, these results highlight that in rabbit antibiotic-free production, dietary supplementation with plant polyphenols and brown seaweeds, positively affects *Semimembranosus* muscle sensory parameters, enhancing Vitamin E content.

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P103

The production of typical cured meats from cattle of Cinisara breed

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The meat of Cinisara cattle could be used to obtain also processed products as commercial alternatives to fresh meat. The aim of this research was to produce bresaola and salami from adult cows (AC) and grazing (GB) or housed (HB) young bulls of Cinisara breed, evaluating their physico-chemical and sensory traits. In the last 3 months, all animals were fed with hay and concentrate; moreover, AC and GB were continuously grazing pasture until slaughtering. The carcases were dissected to separate *semimembranosus*, *semitendinosus* (ST) and *biceps brachii* muscles for bresaola and using the rest of meat for salami. The muscles, after removing fat and tendons, were salted for 14 days at 4 °C, dripped for 8 days at 4 °C, stuffed into natural casing and transferred to drying cells. For the salami, the meat was minced with 20% of lard from pigs of 'Nero dei Nebrodi' breed cut into cubes, and a mixture of salt and spices; each mixture was stuffed into natural casings of straight type (35 cm in length and 7 cm in diameter). During the phases of production, the salami and bresaola were stored in rooms with controlled temperature and relative humidity for 45 and 35 days, respectively. The fat content (% DM) was higher in AC for both bresaola (as muscles mean values: 9.68 vs. 4.78 in HB and 3.52 in GB; $p \leq .001$) and salami (41.46 vs. 36.96 in HB and 32.62 in GB; $p \leq .01$), due to the higher fat content of meat. The contents of SFA, MUFA and PUFA (% FA) in bresaola, as muscles mean values, showed differences among animals (45.24, 42.51 and 10.79 in AC; 41.73, 23.33 and 34.80 in GB; 45.20, 32.32 and 21.72 in HB; $p \leq .001$), as consequence of their different diet, age and especially, intramuscular fat; indeed, the increase of this latter, increasing the level of intracellular lipids, reduce the relative incidence of PUFA incorporated in phospholipid cell membranes. In salami, the differences were minor probably due to the lower lipid contribution of meat compared to the lard added. Sensory analysis of salami showed a better overall acceptability for AC. The bresaola from ST of grazing animals (AC, GB) showed higher shear force. In both products, the volatile organic compounds were higher in AC than in GB and HB. The multivariate statistical approach discriminated animal categories for both bresaola and salami. The results evidenced the possibility to obtain bresaola and salami from different animals and muscles, thus to improve the economic performance of autochthonous cattle.

P104

Evolution of instrumental and sensory properties of the traditional Caciocavallo of Castelfranco cheese in relation to cows' feeding

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Aim of the study was to examine during ripening the instrumental and sensory proprieties of Caciocavallo of Castelfranco cheese produced by using hay or silage as the sole forage source. Three batches of Caciocavallo were manufactured by using milk from two homogenous groups of cows fed isoenergetic and isonitrogenous rations containing 60% (on DM basis) of sorghum silage or hay. Analyses were carried out on curd after salting (time 0) and on cheese after 30, 60 and 90 days of ripening. Instrumental colour (CIELAB system) was determined by spectrophotometry. The compression test was performed by an Instron Universal testing machine. The sensory analysis was performed by using a quantitative–descriptive analysis (QDA) method. Two appearances (uniformity and colour), six odours/flavours (overall odour, overall flavour, milk, butter, grass, hay), three tastes (salty, sour, bitter, umami), and three textures (tenderness, elasticity, oiliness) attributes represented the consensus profile of sensory characteristics. Data were analysed by a mixed procedure to evaluate the fixed effects of diet and ripening times. Sensory profile data were subjected to a preliminary analysis of variance in order to verify the reliability of the panel. No serious defects (poor odour or taste, blowing, etc.) were highlighted in the cheeses. Moreover, no notable discrepancies were found between instrumental and sensory colour and texture. The intensity of yellow was remarkably higher in silage cheeses ($p < .05$), due to the preservation of plant pigments (mainly carotenoids) in ensiled sorghum. Cheese texture traits were not markedly influenced by the diet but changed during ripening with a significant reduction of hardness due to proteolysis ($p < .05$). As general trend, silage cheeses showed higher values of odour/flavour sensory attributes and were saltier and had more umami taste. The same attributes increased during ripening ($p < .05$). Overall, the study highlighted that forage preservation method may modify the organoleptic characteristics of Caciocavallo cheese. Some effects were due to the presence in the raw milk of compounds directly originated by diet (i.e. carotenes, ketones, VFA). However, several of these effects appeared different at different stages of maturation as a result of the interaction between molecules from milk and the formation of new compounds in cheese during ripening.

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P105

Quality parameters of freeze-dried donkey milk

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One of the most important nutritional characteristics of donkey milk is the similar chemical composition compared to human milk, which makes it the most appropriate mammalian milk in those cases in which cow's milk cannot be used. Donkey milk is not produced in large amount, and donkey lactation is concentrated in a few months per year. Use of thermal treatments to reduce microbial content and to prolong donkey milk shelf life received interest by the scientific community. The aim of the present study was to investigate the effects of freeze-drying treatment on donkey milk chemical composition, fatty acid profile and mineral content, in order to compare the nutritional properties of fresh and lyophilised donkey milk. Fresh donkey milk samples were collected in duplicate from 18 Ragusana breed pluriparous asses in mid-stage of lactation with an average body weight of 289 ± 24 kg. From the total 36 milk samples collected, 18 were subjected to freeze-drying process at CRUA (Consorzio di Ricerca Unico di Abruzzo), using a Beta 1–16 lyophilizer. To prepare the reconstituted donkey milk samples with a moisture content of 90% (wb), 0.9 g of freeze-dried donkey's milk was completely dissolved in 10 ml of distilled water. Data obtained were subjected to analysis of variance. Chemical composition determined in freeze-dried donkey milk powder showed lactose as the most represented nutrient found (73.2%), followed by proteins (16.1%), then fat (4.48%) and finally ash (3.20%): the Total Solid content was 96.9%. The moisture, protein, fat, lactose, and ash contents of the reconstituted milk were not significantly different compared to the values obtained in fresh donkey milk. Fatty acids content in fresh and in freeze-dried donkey milk did not show significant differences, showing both in fresh and in freeze-dried milk large amounts of PUFA, including essential fatty acids (n-6 and n-3). Fresh and lyophilised donkey milk showed a ratio UFA/SFA respectively of 0.89 in fresh and 0.90 in freeze-dried milk. Minerals content in both fresh and freeze-dried donkey milk showed the highest average concentration for Ca, followed by P, K, Cl, Na and Mg. Nutritional properties of freeze-dried donkey milk remained basically unchanged compared with fresh milk. Freeze-dried donkey milk is easy to transport, requires no special conditions for prolonged storage. Therefore, freeze-drying can be a useful tool to supply donkey milk on the market all-over the year.

P106

Nutritional characteristics of 'Kefir' fermented milk

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The fermented milk 'Kefir' is a drink of Caucasian origin, made by inoculating cow, goat, or sheep milk with kefir 'grains' which is a starter made up of proteins, lipids, bacteria and yeasts kept together by a liposoluble polysaccharide called 'Kefiran'. In the kefir, milk proteins are partially digested, therefore can be easily metabolised by the human organism, and are also characterised by a low allergenic potential. From a nutritional point of view, kefir is rich in essential amino acids, minerals and vitamins, it is also considered a probiotic drink since it contains a number of microorganisms useful for the proper functioning of the gut microbiota. Furthermore, kefir is a drink suitable for lactose-intolerant individuals since the 'grains' show a good β-galactosidase activity. The aim of the present work has been a proteomic analysis of kefir (derived from bovine milk) through two-dimensional electrophoresis (2DE). Kefir was centrifuged (13000 *g* for 20 min) and then filtered with 0.22μ filter in order to remove bacteria. The total protein present in the filtered kefir was concentrated by precipitation and subsequently treated with 2D-Clean-Up. The second dimension consisted of a 15% SDS-PAGE. The gel was analysed by the software PDquest for spot quantitation, determination of isoelectric point and molecular weight. The results obtained showed that most of the low molecular weight spots were proteins deriving from κ-β and α-caseins and some of them are bioactive peptides with important nutraceutical properties. Q38 In particular, in kefir are present κ-casein fragments containing the sequence of the Casoxin C (YIPIQYVLSR), a bioactive peptide able to bind to the μ opioid receptors, that regulates the emotional behaviour and the intestinal motility. Casoxin C is able to stimulate the release of histamine, leading to a rapid contraction of the intestinal smooth muscle and the release of prostaglandin E2. Finally, Casoxin C seems to stimulate the processes of phagocytosis. Another spot named KM has been identified as a peptide derived from the α_{s2}-casein that contains the sequence of Casocidin-1, a bioactive peptide with antimicrobial properties, able to inhibit the growth of Gram-negative and Gram-positive bacteria such as *Escherichia coli* and *Staphylococcus carnosus*. Data obtained permitted the identification of bioactive peptides in kefir which could be responsible for the nutraceutical effects that characterise this drink.

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Marchigiana breed and the PGI mark ‘Vitellone bianco dell’Appennino centrale’: analysis of the BovinMarche database

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Protected Geographical Indication (PGI) mark ‘Vitellone bianco dell’Appennino centrale’ is a tool to valorise autochthonous meat produced by Chianina, Marchigiana, and Romagnola beef cattle breeds, reared along the Apennine ridge of central Italy (Reg. EC 134/98). BovinMarche is a breeders’ association that, since the year 2000, plays an intermediation and marketing role between breeders and actors of the bovine meat supply chain; BovinMarche is also responsible for SEUROP carcase classification according to Reg. (EC) 1234/2007, 1249/2008, and (EU) 1308/2013. Moreover, on slaughtered beef cattle, BovinMarche collects in a database: bovine ID, birth date, age, sex, breed, carcase weight, carcase classification, mark (PGI or Bovinmarche), slaughterhouse, slaughtering date. Therefore 11,476 records, collected throughout the period 2011–2018, were analysed with JMP 10 software showing an average carcase weight of 423.99 ± 83.87 kg, and an average slaughter age of 21.73 ± 2.74 months. The descriptive statistical analysis showed that 82.67% of slaughtered animals were male and 17.33% female, mainly distributed in the age categories A (82.54%) and E (17.34%). According to carcase conformation, 70.15% of carcases were classified as U (very good muscle development), 21.13% R (good muscle development), 8.52% E (exceptional muscle development), 0.17% O (average muscle development), and 0.02% S (double muscled carcase type). Fat cover distribution highlighted that 54.31% of carcases were in class 2 (slight), and 45.38% in class 3 (average). Out of 11,476 slaughtered beef, 89.98% belonged to Marchigiana breed, 6.44% crossbreeds, and 3.58% other cattle breeds. Overall 89.48% and 10.52% of carcases were labelled with PGI and Bovinmarche mark, respectively. The U class was the most represented for both specification marks with a similar frequency (≈70%); remaining carcases were classified mainly in R class. The data analysis throughout 2011–2018 highlighted an increase of U class frequency from 2011 to 2014 (78.76%) and a progressive decrease with the lowest value in 2018 (61.79%). The frequency of the class R increased progressively from 2011 (12.43%) to 2018 (34.29%); moreover, class E showed a frequency steadily decrease in the eight-year period. These results suggest that, regardless of marks, beef cattle carcases monitored in the period 2011–2018 by BovinMarche are showing a reduction in the profiles’ convexity and in the round, back, shoulder muscles development.

P108

Poultry meat quality in antibiotic-free production has improved by natural extract supplement

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Farm sustainability is a key factor in animal production. In recent years, the consumers demand for products of animal origin coming from production chains certified for animal welfare has increased. Moreover, the institutions, have restricted antibiotic use in order to prevent the antibiotic resistance. For these reasons, antibiotic-free production chains are emerging. The search for innovative nutritional strategies able to support animal health and enhance product quality is required. Natural extracts containing polyphenols and seaweed are rich in bioactive compounds able to enhance animal health and product quality. The study was designed to investigate the effect of dietary supplementation with polyphenols and seaweed extract on meat quality parameters in Hubbard slow growth female chicken in antibiotic-free production. The animals were fed a control diet (C) and a diet integrated with 0.3% of polyphenols and seaweed extract (T1) and were slaughtered at 56 days of age at an average weight of 2.1 kg. Ten carcases per treatment were randomly selected for the determination of meat quality parameters. Chemical parameters, pH, colour parameters and oxidative stability were determined at 0, 3, 7 days of refrigerated storage on the right breast and thigh. Data on chemical parameters were analysed by one-way ANOVA and the other parameters were submitted to a repeated measure ANOVA. In breast, protein and ash content resulted higher in T1 group than in control ($p<.01$). The pH was affected ($p<.001$) by storage time in thigh. The redness values were unaffected ($p>.05$) by dietary treatments and storage time in both muscles. As expected, the lightness and yellowness values in thigh and breast were negatively affected by storage time ($p<.001$). The oxidative stability resulted higher in thigh of chicken fed polyphenols and seaweed extract ($p<.01$) than in control. As expected, storage time negatively affected ($p<.001$) oxidative stability in both muscles. Overall these results suggest that in chicken antibiotic-free production, dietary supplementation with polyphenols and seaweed extract, positively affects protein content in breast and protect muscle from oxidative processes, enhancing poultry meat quality parameters.

Acknowledgements

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P109

Effect of two sous-vide cooking methods on physicochemical characteristics of *Longissimus thoracis* muscle from pigs fed with or without extruded linseed

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Meat is recognised as a highly nutritive food. With cooking, the meat enhances its nutritional value and becomes more digestible. However, high cooking temperatures lead to several chemical modifications in meat. Therefore, there is an increasing interest in sous-vide cooking. The aim of this research was to study the influence of two sous-vide cooking methods on physicochemical characteristics of *Longissimus thoracis* (LT) muscle from 24 pigs fed with two different diets: control group (C) had a basal barley/soybean diet; in the linseed group (L), 5% of extruded linseed replaced the same amount of barley. At 24 h *post mortem*, LT muscles were sliced, vacuum sealed and stored at −18 °C until analysis. Samples were cooked in water bath at two different combinations of temperature and time: 'A' 80 °C of the bath water as long as the core temperature of the pork reached 70 °C; 'B' 60 °C for 15 h. After the cooking process, the samples were kept under refrigeration (2 °C) for 24 h. The day after, cooking loss, colour, pH, microbial growth and tenderness were determined. Microbial growth was analysed also in the raw meat. Statistical analysis was performed by means of ANOVA, using the GLM procedure of SAS. Dietary treatment (C vs. L) and cooking condition (A vs. B) were used as independent variables. Dietary treatments did not produce significant differences in pH, colour, cooking loss and tenderness. Colour parameters were affected by cooking method: a* values of the internal part of the sample and b* values of the external part were higher for the samples cooked at 60 °C (a* 4.74 vs. 3.97 for B and A, respectively, $p<.05$; b* 17.79 vs. 15.84, $p<.01$). The A cooking method led to higher ($p<.01$) shear force values (5.03 vs. 3.30 kg). The microbial load in the raw meat was significantly different ($p<.05$) between dietary treatments: C group showed higher total viable count (4.56 vs. 4.14 $\log_{10}$) and *Enterobacteriaceae* (2.65 vs. 1.94 $\log_{10}$) respect to L group. Low microbial growth was detected for both cooking methods. Total viable count was 0.50 $\log_{10}$ for A cooking method and 0.64 $\log_{10}$ for B, without significant differences among them. No *Enterobacteriaceae* growth was detected. Also, no differences were found for pH and cooking loss between A and B. Both cooking methods generated a meat safe from contamination. The B method: low temperature for long time also, generated tender meat.

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P110

Effect of different rearing system on sensory characteristics of meat in Massese Lamb

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Meat of lamb has specific flavour and taste, distinct from other popular red meats. The traditional Massese lamb is a suckling lamb and, in this study, they are reared up to two months of age and fed by supplement in order to increase meat production. The feeding system could affect the sensory properties of lamb meat: pasture and associated flavours may be unfamiliar to consumers of meat respect to meat produced from lamb fed with concentrate. Historically, Massese sheep farmers in mountainous environment used different farming systems based on changes in resources availability during the year and on their seasonality. In this research, Massese lambs were reared using two different farming systems: stall (S) including concentrate and hay as supplementary fed, and pasture (P) that includes only pasture as supplementary fed.

The sensory proprieties of roasted unsalted *Longissimus dorsi* samples were evaluated by a team of 11 trained panellists who did not receive any information regarding the lambs. For each sample, the panellist was asked to evaluate in a continuous scale from 1 to 10 the following parameters: tenderness, juiciness, flavour, taste and overall preference. Data were analysed by GLM of SAS using feeding system, panellist and trial day as discrete effects.

The farming system affected meat colour: meat of S lambs is brighter (51.4 vs. 32.1 for S and P groups respectively; $p<.001$) and has a less intense colour (33.6 vs. 45.4; $p<.001$). The taste does not show undesirable aroma except for a slightly metallic sense in both meat types.

The characteristic lamb flavour is not very intense while the characteristic lamb taste is greater in S group (33.2 vs. 27.4; $p<.05$). Effectively, the S lambs showed a greater amount of intramuscular fat that affected the aroma component of meat. The meat of both groups has similar tenderness, juiciness and acceptance level. The results of the present study underline that both groups obtained a high level of liking and acceptance.

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P111

Effects of dietary by-products in mealworm rearing

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Insects represent one of the answers to increasing demand for animal-based protein, related to both the growing world population and also to the increasing demand of protein from developing countries. The lower environmental impact of insects compared to conventional production animals (mostly linked to greenhouse gas production, use of water and use of arable land) and the higher reproductive capacity, higher nutritional quality, and higher feed conversion efficiency lead several researchers to study insects as food and feed.

Furthermore, insects can be reared on sustainable feeds, such as waste or by-products that do not meet the nutritional values needed by other farmed animals. By-products represent a valid source of energy with a marginal cost, and they could be used as feed ingredient in regard to their essential nutrients. Indeed, diet composition could affect insect's development rate and their body nutrient composition.

The objective of this research was to evaluate the influence on the chemical composition of mealworm larvae (*Tenebrio molitor*) of different diets based on food by-products. Five different diets were formulated: brewery spent grains, bread (leftover), cookies (near to the expiration date), 50% brewery spent grain and 50% cookies, and 50% bread and 50% cookies. Mealworms were harvested when the first pupa was observed. Larvae were analysed for the determination of the proximate composition, antioxidant status, fatty acids profile, amino acids profile, microbiological analyses (fasted and un-fasted).

Results of this study highlighted a strong effect of the diet on the final insect products. All the tested parameters were affected by the diet, in relation to the physico-chemical composition of the feeds. Also, microbiological analyses were affected by the diets and the fasting procedure.

Notably, the chemical composition of the larvae followed the chemical composition of the feed stuffs, highlighting that mealworm larvae express a wide rearing plasticity that lead to several possibilities of employment as food and feed.

P112

Effects of different blanching treatments on quality and microbiological profile of mealworms

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Nowadays, acceptability of edible insects is very low to Western consumers, mostly to European ones; nevertheless, in the last years several insect products have been sold online and in supermarkets with also an increasing interest of media.

Safety concern is one of the main factors that reduce the willingness of consumers to eat insects, related to allergic, parasitical, chemical and microbial hazards. Another important concern in consumers is the unwillingness to taste something unknown, this behaviour is also called food neophobia. Grinding is a possible way to include insect as small particles that could be easily mixed in a well-known product without altering its appeal. Anyhow, grinding could induce an enzymatic browning process that does not allow their use as ingredient.

In this study, the effects of different blanching treatments on the microbiological profile, pH and colour of mealworm (*Tenebrio molitor*) larvae were evaluated. The effect of 10 combinations of temperature (50, 60, 70, 80 and 90 °C) and time (2.5 and 5 min) was evaluated in comparison to fresh larvae (not blanched) and oven cooked larvae (10 min at 150 °C). Moreover, effect of 24 h starvation was also evaluated on the microbiological profile. Starvation affected only marginally the microflora, furthermore, in all samples *Escherichia coli*, *Bacillus cereus*, *Listeria monocytogenes* and *Salmonella* spp. were never detected. A blanching treatment at 60 °C for 5 min seems to be the lower time-temperature combination in order to achieve a significant decrease of microbial loads. Blanching treatments played a role also in pH and colour modifications: larvae blanched at least at 60 °C completely stopped browning effect, maybe in relation to an enzymatic inhibition. Among the blanching treatments tested, 60 °C for

5 min seems to be the more practical application in order to achieve the fixed goals. Lower temperature or time combinations showed to be unable to reduce microbial loads or to stop the browning effect, on the other hand, higher temperatures did not show an improvement in quality and microbiological parameters.

P113

Carcase characteristics of Massese lamb in different rearing systems

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Massese is a dairy sheep breed, autochthonous of the Tuscan-Emilian Apennines and widespread in this territory. Massese lambs are traditionally slaughtered at 30 days in order to preserve milk production for cheese making. However, their good size and growth suggest the possibility to extend slaughtering age to over 2 months of age, by gradually weaning and supplementing lambs with a balanced feed ration, in order to increase meat production. This research aimed at evaluating the convenience to extend the rearing of Massese lambs up to 70 days. We determined carcase yield, weight of commercial cuts and tissue composition of loin, shoulder and leg in 20 lambs reared in three systems: stall (S) fed with concentrate/hay, semi free-range (F) fed with concentrate/hay and pasture, and free-range (P) fed only with natural pasture. The lambs were kept constantly with their dams, thus suckling milk was always available. Data were analysed by Linear Model (R 3.4.1) including a covariate (age in days) and three categorical variables: sex, rearing system and type of calving (single or twin). Single-born lambs displayed higher liveweight (p=.03), carcase weight and yield compared to twin lambs, but also a higher amount of fat. The rearing system affected significantly liveweight, carcase weight and yield (p<.05): carcases were heavier in F and S (11 kg) than in P (7 kg), and yields were the highest in F (54%). The rearing system influenced also the quality of the carcase, in term of weight of the commercial cuts (expressed as proportion on carcase weight) and their tissue composition. Ribs and loins were heavier in S. Lambs reared in the free-range system had the lowest proportion of fat, while those reared in the semi free-range system displayed the highest proportion of muscles, an intermediate proportion of fat and the lowest proportion of bone tissue. Muscle/bone ratio resulted more favourable in S and F lambs (p=.02) so as the proportion of muscle in relation to the defatted carcase (p=.02). The study showed that Massese lambs reared up to 70 days of age are able to provide carcases which double in weight respect to lambs slaughtered at 1 month of age. However, this result is achievable in rearing systems that provide an adequate concentrate supplementation to forage resources.

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P114

Familiarity and preference of Lucanian consumers for animal-based products

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Within the Italian Taste project data from 192 Lucanian consumers, balanced for gender and three age classes (18–30; 31–45; 46–60 years), were gathered through questionnaires concerning demographic information, familiarity (on a 5-point scale, from 'I do not recognise it' to 'I regularly eat it') and liking (on a 9-point hedonic scale, from extremely disliked to extremely liked) for 48 animal products (18 dairy products and 30 items for meat, fish and eggs). Data on familiarity and liking were subjected to ANOVA using body mass index (BMI), gender, age, food neophobia (FNS) and orientations towards health and hedonic characteristics of foods (HTAS) as factors. The effect of familiarity on BMI was also studied. Respondents were 61% normal weight (NW), 26% overweight (OW) and 12% obese (O). Age significantly ($p < 0.01$) affected liking, with higher liking in the first age class compared with the third class for kebab (6.3 ± 0.4 vs. 3.7 ± 0.4), hamburger (7.3 ± 0.3 vs. 4.8 ± 0.3), salami (6.9 ± 0.3 vs. 6.7 ± 0.3), spicy salami (6.9 ± 0.3 vs. 5.4 ± 0.3), baked ham (7.7 ± 0.3 vs. 6.35 ± 0.3), sausage (8.2 ± 0.3 vs. 7.1 ± 0.3), chicken breast (8.1 ± 0.3 vs. 6.9 ± 0.27), cutlet (8.3 ± 0.3 vs. 7.1 ± 0.3) while this age class liked more (p<.05) lamb ribs (7.7 ± 0.4 vs. 6.3 ± 0.4) and cod (7.3 ± 0.4 vs. 6.1 ± 0.4). Women gave higher scores for skimmed milk, compared with males (6.3 ± 0.3 vs. 5.5 ± 0.3), while fresh and seasoned pecorino cheese, lamb ribs (p<.001) and spicy salami (p<.05) were preferred by males. Body mass index affected liking with higher scores in O consumers for whole milk (7.5 ± 0.5 vs. 6.1 ± 0.2, p<.05), hamburger (6.5 ± 0.5 vs. 5.6 ± 0.2, p<.05), rib beef (8.1 ± 0.4 vs. 7.0 ± 0.2, p<.05), chop (8.4 0.4vs 7.0 ± 0.2, p<.01), sushi (4.5 ± 0.7 vs. 2.9 ± 0.3, p<.05), ham (8.5 ± 0.4 vs. 7.3 ± 0.2, p<.01) and baked ham (7.7 ± 0.4 vs. 6.7 ± 0.2, p<.05) compared with NW. Regular consumers of lamb ribs (25.6 ± 3.9

vs. 19.3 ± 3.4, $p<.05$), chops (25.3 ± 4.6 vs. 15.7 ± 3.8, $p<.05$) and spicy salami (25.3 ± 4.1 vs. 19.6 ± 3.6, $p<.05$) showed higher BMI compared with non-consumers. We found a significant correlation between the HTAS subscale 'using food as reward' and liking for kebab (r = 0.22, p=.0033) and hamburger (r = 0.21, p=.0046). While 'food as pleasure' was negatively correlated with FNS values (r= −0.38, $p<.0001$). We conclude that liking of animal-based product in Lucanian consumers is affected by gender and age, while regular consumption of particular animal-based products may increase BMI.

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P115

Nutritional value of retail meat lipid fraction from Piemontese young bulls

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Compared to conventional animals, the meat of double-muscled subjects is characterised by lower levels of fat. The aim of this study was to investigate the nutritional value of the lipid fraction of Piemontese beef. Samples of *longissimus thoracis* muscle from 10 Piemontese double-muscled young bulls purchased at retail from local butcher shops were analysed. Proximate composition, cholesterol and fatty acid content were determined. In this study, the intramuscular fat and the protein content was 1.10% and 22.45%, respectively. Thus, one serving of meat (100 g) had 99.7 kcal of which only 9.9 kcal were from fat, that is only 9.9% of the total energy. Considering a recommended daily intake of 2000 kcal and 65 g of fat, 100 g of raw meat provided 1.69% of fat and 0.50% of fat energy. Piemontese beef showed Nutritional Quality Index values lower and higher than 1 for fat and protein, respectively, which are desirable from a nutritional point of view. The cholesterol content was 51 mg/100 g meat, which represents 17% of the maximum daily cholesterol intake recommended in adults (300 mg per day). The content of saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids was 469, 337 and 200 mg/100 g meat, respectively. Therefore, the PUFA/SFA ratio was above 0.4, which is the minimum value recommended. In particular, this is due to the low proportion of SFA. Aiming to a dietary pattern that achieves 5% to 6% of calories from SFA and considering a daily need of about 2000 kcal, no more than 100–120 kcal should come from SFA. Linoleic (LA) and α-Linolenic (ALA) fatty acids content was lower than the minimum recommended intake in human diet. In addition, the high level of n6 PUFA (188 mg/100 g meat) compared with the low level of n3 PUFA content (9.3 mg/100 g meat) led to an imbalanced n6/n3 ratio, which was higher than the recommended value of 4. In conclusion, Piemontese beef showed a desirable low fat and SFA content, but a very high n6/n3 ratio which is not desirable from a health point of view. Feeding strategies should be adopted to favour the deposition of n3 PUFA in beef in order to obtain a healthier product.

P116

Growth performances of rainbow trout (*Oncorhynchus mykiss*) juveniles fed on a diet containing processed fish as a partial substitute for fish meal

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The recovery of valuable nutrients from fishery discards and seafood processing is a promising alternative to the use of fish meal and fish oil, which are considered limited resources and non-sustainable feedstuffs. Studies focused on the replacement of fish meal with alternative sources, also consider the quality traits of by-products derived from wild and farmed aquatic organisms. In the past, these waste by-products were discarded without any attempt to recover or process into low market-value products, such as pet food and fertiliser. Recently, it has been ascertained that they still contain a fair amount of nutrients that could be better employed. By-products typically include muscle cuts (15–20% of the whole fish), skin and fins (1–3%), bones (9–15%), heads (9–12%), viscera (12–18%) and scales (5%). The use of discarded raw materials and processed by-products can reduce the pressure on fish stocks as well as diminish the waste and negative environmental impact associated with the processing of aquatic animals.

The present paper aims to evaluate the growth performances of rainbow trout juveniles fed on a diet including processed meal, obtained from seafood processing, as a partial replacement (50%) of fish meal. The processed meal was prepared after having extracted oil from discarded fish, which had been grounded and cooked at 70 °C, utilising both a hydraulic press and centrifugation.

The results were compared to those obtained by a control group, represented by conspecifics of the same initial mean body weight (25 ± 2 g), fed on a conventional diet (CD) composed of fish meal as the main protein source. The two feeds were isonitrogenous (45%) and isolipidic (21%). The fish were reared in duplicate tanks/group, with a recirculating aquaculture system. Growth performances and food conversion rate were evaluated after 90 days of feeding.

Good productive parameters were obtained in the two groups with similar performances. No significant differences were found in the final mean body weight, which ranged from 105 g and 110 g. The feed conversion ratio was around 1.2 and the survival rate was around 98% in both groups.

From an economic point of view, considering production and feed costs, the results suggest that there is an economic return in replacing 50% of the fish meal protein with processed seafood when this replacement is performed during the rainbow trout pre-growing phase.

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P117

Dietary selenium supplementation of Friesian cows modifies the aromatic profile of dairy products

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This study aimed to investigate the effect of dietary selenium (Se) supplementation of Friesian cows on the aromatic properties of dairy products. Thirty-two Friesian cows, balanced for parity, milk production and days in milk, were randomly assigned to 2 groups. The trial lasted 63 days in which the control group (CG) was fed with a conventional feeding strategy, while the experimental group (SeG) received daily selenomethionine (SeMet) supplementation. During the experimental period, the milk yield was monitored and samples of milk and related Caciocavallo cheese were collected and analysed in order to obtain information on chemical-nutritional composition. To evaluate the effect of ripening on the aromatic profile, the analysis was performed on cheese samples collected after 7 (T_7) and 120 (T_{120}) days after the cheese-making. The volatile compounds composition resulted positively affected by dietary Se intake, with a significant increase in concentration of carboxylic acids, esters and lactones ($p<.05$), reflecting the predominance of the lipolytic processes respect to the proteolytic events. The general increase of carboxylic acids in ripened cheese could be explained by the extent of starter cell autolysis, with the consequent release of peptidases and especially lipases that accelerate the lipolytic events. Among the most represented carboxylic acids must be indicated butanoic and hexanoic acids; such compounds are considered to be mainly involved in the determination of cheese flavour, giving origin to cheesy, rancid and sweaty odours. The increased production of such compounds could be probably explained by an increase of lipolysis of the triglycerides by microbial and endogenous milk enzymes, resulting in an augmented release of free fatty acids (FFAs). With regard to the esters, such compounds are characterised by a low odour threshold and are generally associated with the sweet, fruity, and floral notes of surface-ripened cheese flavour. Lactones, instead, are generally produced by a one-step transesterification reaction of hydroxylated FFAs which represent the main precursors. Such findings could contribute to the production of cheeses with interesting organoleptic properties, although further sensorial evaluations should be performed to deeply investigate these changes and confirm the consumer acceptability.

P118

Enzymatic injection of papain for improving meat tenderness

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One of the most important sensory properties of meat is tenderness. Traditional ageing of meat improves tenderness, however, is time consuming, expensive and its effectiveness varies between different commercial cuts and animals. The use of exogenous proteases from plants (papain from papaya, bromelain from pineapple, etc), bacteria and fungal sources are an alternative method to increase meat tenderness and decrease production costs.

Therefore, the aim of this research was to study the effect of enzymatic injection of papain on meat tenderness. As first step, the optimum of enzymatic concentration of papain, that represents a main limit of the application of exogenous enzyme, was identified. Subsequently, the effect of papain tenderisation was evaluated on 12 *Semitendinosus* muscles removed from six beef cattle carcasses 24 hours *post-mortem*. Each muscle was divided into 8 steaks (3 cm thick uniform-sized slices) and randomly assigned to two experimental treatments: control (C) without enzymes injection and enzymatic injection (EI) with papain solution and aged at 3 °C for 0, 24, 48 and 96 hours. At each ageing time, Warner–Bratzler shear force (WBSF), texture profile analysis (TPA), total collagen and changes in myofibrillar proteins were estimated. Results of mechanical properties highlight that the injection of papain solution improves meat tenderness accelerating ageing processing. On the contrary, ageing did not affect the WBSF and TPA profile only in EI samples. Lowest collagen content was found in meat treated with papain, while no significant ageing time effect was observed for all meat samples. Papain-treated samples showed an electrophoretic profile characterised by the lowest intensity and number of bands starting from t0 with a complete degradation of all myofibrillar proteins. Particularly, the total disappearance of all intact isoforms of TnT found in EI samples confirms more intense proteolysis in this treatment as also supported by the highest content of TnT-derived polypeptides (32–24 kDa).

Data suggest that the enzymatic treatment could be a promising technique to accelerate conventional ageing of meat determining energy and cost saving, and become very attractive especially for meat that is tougher and requires a long ageing time because of breed, age and type of cut.

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P119

Genomic meat traceability: from breeders to consumers

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In recent years, conventional traceability systems, based on paper documents, have been widely used to verify the origin of local Italian-bred beef. Nowadays, product authenticity regarding species, breed and animal can also be guaranteed by genomic methods.

Our study compared DNA analysis results regarding breed of origin with the label indications from beef samples taken from 4 big Italian retail shops.

The DNA of 24 beef samples were analysed, with the BovineSNP50 BeadChip v2 counting 54,609 SNP markers. The breed of origin was estimated using publicly available SNP data for pure breeds
Q39 as reference genotypes and using a probability clustering and assignment approach (Plink and ADMIXTURE software) to classify samples. An independent set of data (reference dataset) derived from 199 animal of known origin was used to assign each sample analysed to a breed. The reference dataset is derived from 10 most common breed in Italy. To evaluate assignment performance 100 bootstrapping replicas were performed with CLUMPP software, resulting in 97% similarity among the replicas.

The percentage of origin matched DNA results for 46%, i.e. 11 of the 24 samples analysed. Two types of inconsistencies were observed: 33% of the samples labelled ‘pure bred’ were identified as ‘crossbred’ or a different breed by DNA; while 21% labelled ‘crossbred’ were identified as ‘pure bred’ by DNA. In the first case, consumers and/or supply chain buyers pay more for products that are worthless. In the second case, beef is sold for less than its worth, with negative consequences for all along the production chains.

These results show how genomic traceability can protect consumers by guaranteeing authentic Italian beef at a fair value for their money. Lastly, everyone in the beef value chain, from producers to retailers, can benefit from a more detailed and scientifically correct evaluation of Italian beef.

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Physical characteristics of eggs from Hy-line Brown layers fed with *Hermetia illucens* larvae meal

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A total of 162, sixteen-week-old hens (average live weight 1.41 kg ±0.13) were divided into three groups: the control was fed a corn-soybean meal-based diet (SBM) covering the hens’ requirements according to the Hy-line Brown commercial line management guide (2016). In the diets of the treated groups (HI25 and

HI50), the soybean meal was partially replaced by an insect meal from *Hermetia illucens* (HI) larvae, in order to develop three isoprotein and isoenergetic diets. In the HI25 and HI50 groups, 25 and 50% of the diet protein were replaced by the HI protein, reaching an inclusion level of 7.3 and 14.6%, respectively. The hens were housed for 20 weeks in the same building in modified cages (800 cm^2/hen), under controlled environmental conditions. For each group, the hens were distributed into three cages (18 hens/cage), each of them was divided by two internal transects into three equal areas in order to obtain nine replicates of six hens/group. Feed and water were manually distributed. The dark:light cycle was 9:15 h. Six eggs per group and per week were weighed, and the weights and percentages of shell, yolk and albumen were recorded. The shell thickness was measured using an appropriate calliper. The yolk colour measurement was performed with a Konica Minolta colourimeter, according to the CIE system and expressed as lightness (L*), redness index (a*) and yellowness index (b*). The albumen pH was measured using a pH-meter. The eggs from the insect groups were heavier than those of the control (61.10 and 60.82 vs. 57.56 g, respectively for HI25, HI50 and SBM groups; $p<.05$) and the SBM group had a shell thicker than that of the HI50 group (541.9 vs.. 476.3 μm; $p<.05$). The yolk weight of the insect groups was higher ($p< .01$) than that of the control (14.30 and 14.33 vs. 12.93 g, respectively for HI25, HI50 and SBM groups). Nevertheless, the albumen and yolk did not show significantly different incidences on the whole egg weight. The yolk of the eggs did not differ for L* values among groups, but the a* index of the eggs obtained from the control was markedly higher ($p< .01$) than that of the insect groups (6.01 vs. 0.67 and 0.80, respectively for SBM, HI25 and HI50 groups). Furthermore, the b* index of the HI25 group was higher ($p<.05$) than that of HI50 (33.01 vs. 27.39). Both levels of HI larvae meal used in this trial modified the physical properties of the eggs inducing a heavier weight and a lower redness index of yolk and 25% of the diet protein content replaced by the HI protein seems to be more appropriate than 50% level of replacement as it did not modify the shell thickness.

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Healthy indices and oxidative stability of Marchigiana beef meat enriched with omega-3, vitamin E and treated with oregano and rosemary essential oils

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The use of the oilseed in the diet for ruminant is an interesting strategy to enrich the meat products with healthy fatty acids (FA), such as PUFA, omega-3 and CLA. However, it should be taken into account that PUFA are more susceptible of lipid oxidation due to the double bonds with negative impacts on organoleptic characteristics of the products. The aim of this study was to evaluate the effect on FA composition, lipid stability and antioxidant potential of meat from Marchigiana beef receiving dietary extruded linseed and vitamin E and treated with oregano and rosemary essential oils at (T0) and 9d after storage (T9) in modified atmosphere packaging (MAP, 66% O2, 25% CO2, 9% N2).

For the experiment 36 Marchigiana beef (421.7 ± 28.4 kg) divided in three homogeneous groups received during the finishing period until slaughter the following diets: group C (control diet), group L (linseed), group LE (linseed + Vitamin E). After an ageing period of 12d two sections of *Longissimus dorsi muscle* (2.5-cm thick steaks) from each carcase were cut. Half of the steaks from each LD muscle were dipped in the essential oil (EO) for 60 s (0.5 ml of rosemary +0.5 ml of oregano essential oil) and the rest were left untreated (UO). All samples underwent to *MAP and* stored at +4 °C until 9d and then sampled for subsequent analysis.

Meat from L and LE (T0 and T9) groups showed a significant and favourable impact on PUFA composition ($p<.05$), n-3 ($p<.01$) and CLA ($p<.05$).

At T9 TBARs values in meat from group L were higher than in LE and C (0.98 vs. 0.61 vs. 0.66; $p<.05$). At T9 EO affected TBARS significantly when compared to UO steaks ($p<.05$). Meat colour in samples from LE at T0 showed significantly higher values of the coordinate a* ($p<.05$) and Chroma (C*) ($p<.01$) compared to C and L; at T9 a* values were significantly higher in all samples ($p<.05$) treated with EO compared to UO. The antioxidant potential as expressed by FRAP was significantly higher in LE ($p<.01$) compared to L and C groups. Moreover, EO treatment significantly affected FRAP values ($p<.05$) after 9 days of storage. Our results evidenced the FA profile of meat treated with L and LE showed a higher value of healthy indices, moreover, the lipid oxidation is positively affected when diet is supplemented with vitamin E. The EO treatment contributes to enhance the antioxidant potential of meat enriched with EL after 9d of storage.

Acknowledgements

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P122

Feasibility study of NIRS technology to predict Merino wool quality parameters

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The wool market is gaining importance in recent years due to the rising interest of the textile industry to manufacture high-quality products, along with the valuable resource that wool represents by itself and the involvement it has on the biodiversity conservation. The determination of the quality parameters of wool fleece yield, fibre diameter, coefficient of variation, comfort index, brightness and luminosity are laborious, time-consuming and expensive, as they imply washing, weighing and the use of different instruments. Therefore, it would be quite useful to have an objective, reliable and fast analytical method to characterise wool according to its quality. In the present work, a feasibility study of near infra-red spectroscopy (NIRS) for the prediction of the parameters of importance in the assessment of the quality of Merino sheep wool has been carried out. A total of 127 greasy and 121 clean wool samples have been used. Wool samples were scanned in circular cups of 4 cm Ø, using a FOSS-NIRSystems 6500 NIR scanning monochromator. Spectral absorbance values were recorded in reflectance mode (400–2498 nm, every 2 nm). Spectral data were fitted to laboratory data. MPLS regression equations were developed applying several mathematical treatments in both data sets. The standard error of cross-validation (SECV) and the coefficient of determination of cross-validation (r^2_{CV}) were used to select the best models. The comparison of the results obtained in both groups showed promising prediction ability for fibre diameter, brightness, and luminosity both in greasy and clean wool sample sets (r^2_{CV} of 0.6 vs. 0.6, 0.8 vs. 0.7, 0.8 vs. 0.7, respectively), although errors for colour-related parameters were higher in the greasy one (SECV 1.2 vs. 1.2, 10.2 vs. 2.8, 4.8 vs. 1.5, respectively). However, better results were obtained for the prediction of fleece yield in greasy wool, as compared with the clean set (r^2_{CV} 0.6 vs. 0.3, and SECV 5.1 vs. 8.5, respectively). More limited results were obtained for the remaining laboratory measurements. Therefore, the possibility of getting quality parameters directly from the NIRS analysis of greasy wool samples will suppose an important saving in time and cost for farmers, wool traders and textile industries. Future works should be aimed at expanding the data sets, in search of greater variability and better distribution of the parameters of interest, to improve the precision and accuracy of the predictive models here initiated.

P123

Consumers' perception of insect meal as aquaculture feed ingredient

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The recently adopted Regulation No 2017/893 authorises the use of insect proteins originating from seven insect species [black soldier fly (*Hermetia illucens),* common housefly (*Musca domestica*), yellow mealworm (*Tenebrio molitor*), lesser mealworm (*Alphitobius diaperinus*), house cricket (*Acheta domesticus*), banded cricket (*Gryllodes sigillatus)* and field cricket (*Gryllus assimilis*)] in feed for aquaculture fish. To date, many researchers have successfully demonstrated that insect meals (IM) can be used as feed ingredient for a variety of fresh and seawater species. However, alongside legislation and scientific knowledge, consumers show increasing interest towards animal breeding, especially for welfare and feeding items. For this reason, we decided to perform a survey (using Google Forms®) among Italian consumers, in order to understand their perception and acceptance of insects as alternative protein source for aquaculture species. Six hundred and seventy-eight answers were collected. Both males and females (47.8 and 52.2%, respectively) aged from 18 to 75 years attended the questionnaire. Firstly, people were asked for general information (gender, age, country, income) and their food attitudes; only people usually consuming fish flesh could proceed with the subsequent questions (96.5%). For 53% of the interviewees, fish, especially seawater species, represent an important food in their weekly diet, and almost 49% asserted to prefer a meal with fish rather than one without. Nevertheless, half of the people said that they are indecisive when buying fish due to allergy, heavy metals, geographical origin, pollutants and sustainability of fishery supply chain. To the question 'How do you feel knowing that fish eat insects in nature?' the majority answered not to feel nervous (97%), and only 62 people (less than 10%) assured that they would not accept IM as an ingredient for aquafeed. Nineteen people (3%) were uncertain, basically because of their lack of knowledge, while the remaining participants affirmed that they would accept IM for feeding farmed fish. Finally, almost 78% believe that the use of IM is linked to the concept of 'sustainability' mainly because its production does not directly exploit wild fish, is considered 'natural', and because people consider IM production an environmentally friendly activity. In conclusion, from the present survey, it emerged that Italian consumers seem to be ready to accept insect meal as ingredient in aquafeed.

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P124

Is the European pig carcase classification grid still a valid method to select green hams for PDO circuit?

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The aim of the present work was to collect information at the slaughterhouse on the relationship between carcase composition (lean meat content assessed according to the EUROP classification grid) and the quality of the green hams. Quality was assessed based on indicators included in the product specifications for Parma ham: trimmed weight, thickness of the fat layer (measured vertically at the head of the femur -best end-), presence of fat at the 'coronet' (visually assessed), iodine number.

The research included 11 slaughter plants. In each plant, 10 slaughtering batches and about 20 green hams per batch were selected. All pigs assessed had a carcase weighing more than 110 kg, in agreement with the definition of heavy pig. Overall, 2352 green hams were evaluated. At least 25% of the selected green hams in each batch derived from a carcase classified as E in the EUROP grid (F-o-M classification). Out of them, 35.7% belonged to class E, 33.0% U, 23.3% R, 6.6% O, and 1.4% P.

In every slaughtering batch, 8 samples of subcutaneous fat (including both inner and outer fat layer) were collected, of which 4 from green hams belonging to the E class and 4 from green hams belonging to the other classes (U, R, O). Iodine number analysis is presently in progress, in order to assess if the green hams classified as E are more frequently non-compliant with Parma ham production rules compared to the green hams belonging to the other classes (U, R, O). It is worth remembering that, according to Parma ham production specifications, iodine number must be lower than 70.

As concerns the green hams classified as E, 73.7% of the hams weighed between 13 and 16 kg (which is the preferred weight class for Parma ham) whereas 11.5% had a weight below 13 kg and 14.8% weighed above 16 kg.

Based on these preliminary results, the majority of the raw hams classified as E are compliant to Parma ham production rules (at least 80% for fat thickness; 97.6% for fat at the 'coronet').

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P125

Chemical composition and fatty acid profile of milk from Mediterranean buffaloes fed with two different diets

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Milk provides essential nutrients and is an important source of dietary energy providing high-quality proteins and particularly fats. Mediterranean buffalo milk has a very high-fat content, twice that of cow's milk. Moreover, Mediterranean buffalo milk contains several beneficial compounds such as fatty acids, vitamins and other bioactive elements. Monitoring changes in buffalo milk composition is an important index in order to improve its supply chain and dairy products. The composition of buffalo milk reflects differences in farm management, feeding and environmental conditions. The aim of this study was to determine the effect of fresh forage on milk properties in lactating Mediterranean buffalo diet under intensive farming conditions. Mediterranean buffaloes were split into 2 groups fed with different diets: fresh feeding group (FRS) and dry feeding group (CTL). Milk samples were analysed for four months. Chemical (fat and protein content) and fatty acid compositions of each sample were determined in duplicate. Fat extraction for fatty acid composition was carried out and fatty acids were expressed as a percentage of total methylated fatty acids. Atherogenic index was also calculated. No significant differences in chemical composition between CTL and FRS milk were observed, even if fat content tended to be higher in CTL milk (average =8.17% vs. 7.99%). In the same way, no differences between FRS and CTL group regarding protein content (average =4.43%) were observed. Fatty acid composition of CTL and FRS milk did not differ, except C6:0 and C12:0, being lower in FRS. Higher contents of C18: 1n-9 cis, C18: 3n-3 in FRS milk were observed. As consequences, FRS milk presented higher levels of PUFA and MUFA (4.48% and 23.57%), lower percentages of SFA (72% vs. 76%) and a better atherogenic index (3.07 vs. 3.88). This study underlines that the use of fresh feeding in lactating Mediterranean buffalo diet is able to modify milk fatty acid composition. However, different diets did not have important effects on total fat and protein content. Fresh forage feeding may represent a low-cost technique to increase PUFA and CLAs content in milk. Studies about feeding composition are useful to increase bioactive compounds content, thus providing a tool for differentiation and improve dairy products.

P126

Development of a synergy model aimed to qualify and valorise the natural historic cheese of southern Italy in the Sicilian, Sardinia, Calabria, Basilicata and Campania regions

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The '*Canestrum casei*' project aims to focus the attention on those products at risk of extinction or which have objective difficulties in terms of their qualification and exploitation in the markets. Fifteen 'AGER' cheeses were selected, among the Natural History Cheeses in the South of Italy, by 9 partners. The 'AGER' Selection includes: Ragusano PDO; Provola dei Nebrodi PDO; Piacentinu Ennese PDO; Pecorino Siciliano PDO; Vastedda della Valle del Belìce PDO; Caciocavallo Palermitano; Maiorchino; Caprino Nicastrese; Fiore Sardo PDO; Casizolu del Montiferru; Pecorino di Filiano PDO; Canestrato di Moliterno PGI; Pecorino Carmasciano; Caciocavallo Podolico; Cacioricotta. Cheeses of the 'AGER' Selection will fill some aspects of scientific research, will be characterised by the nutritional profile in order to create 'talking' labels, and will be studied to develop a new marketing approach by IULM of Milan. The general objective of the '*Canestrum casei*' project is to develop a model of synergies to Qualify and Valorise the Natural Historic Cheeses of the South of Italy.
Specific objectives are:

1. Scientific characterisation of 'AGER' 'Canestrum casei' 'cheeses selection' by standardising information of greater interest for consumers in respect of regional production specificities (WP1);
2. Development of researches on the main production processes requiring scientific research to streamline and enhance processes and reduce defects or critical points (WP2);
3. Development of common strategies for communication, promotion and marketing of 'AGER' 'Cheeses Selection' (WP3);
4. Training of producers for the rationalisation of production processes and for a new approach to the sale of cheeses, and involvement of stakeholders in project activities (WP4);
5. Communication and dissemination of the results of the '*Canestrum casei*' project.

The '*Canestrum casei*' project aims to improve the market penetration of the 'AGER' Cheeses Selection by creating an economy in favour of small and medium-sized Italian companies producing these great cheeses with high nutritional value and safety properties, but with poor diffusion and presence in the commercial distribution. The development of joint bid and distribution strategies, along with appropriate promotion, packaging and marketing strategies at the point of sale and on the web, will contribute (expected results) to increase the commercial demand for these cheeses with a relative increase of production.

P127

Authentication of local Italian honey by support vector machines (SVM) based on NIR data

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Honey is a complex natural food product, which is highly appreciated for its both nutritional value and antioxidant properties, related to the presence of sugars and flavonoid and phenolic acids. Honey composition and characteristics are highly influenced by its botanical and geographical origin, therefore the possibility to find a reliable tool to assess these features would be extremely useful for the product traceability and fraud detection, but also for niche products exploitation. While the traditional way to assess honey's composition comprehend time-consuming and expensive analyses, the fast and non-destructive near infrared (NIR) spectroscopy technique has been proven to be a promising tool for the honey characterisation. Thus, the potential of NIR in association with a statistical machine learning method called support vector machine (SVM), were used to discriminate honey's geographical origin. Eighty-five ($n = 85$) samples were collected in 2017. They were classified according to their geographical origin into: Local ($n = 45$), collected from Veneto beekeepers; Italian ($n = 25$), from beekeepers of many Italian regions or acquired from great retail and labelled as produced in Italy; Foreign ($n = 15$), acquired from great retail or websites and produced outside from Italy. Samples were stored at 20 ± 1 °C in dark conditions. Before NIR analysis samples were mildly heated at 39 °C for 30 min in a cabinet, stirred for a better homogenisation and then scanned in triplicates in transflectance mode. A support vector machine (SVM) classifier with a 10-fold cross validation procedure was trained on the raw spectral data to perform a classification of the geographical origins. The best SVM was chosen as the one with the highest average Matthews correlation coefficient (MCC). The predictive performances of the SVM allowed a good classification of Local (MCC =0.66, accuracy =0.83) and Foreign (MCC =0.61, accuracy =0.89) honeys. The Italian class (MCC =0.42, accuracy =0.77) seemed to have in-between characteristics because of the heterogeneity of the samples coming from many regions each with specific flora and atmospheric conditions. Summarising, NIR coupled with SVM provide to be a sensing system for routine online control of honey origin even if it will need to consider a larger number of samples and the use of furthermore powerful spectroscopy tools such as the variable importance in projection (VIP).

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Comparative study of meat quality in thrush, woodcock and starling in Apulia

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The Apulian gastronomic tradition in the autumn-winter period includes several specialities based on the wintering avifauna hunting. Thrush and woodcock are among the most representative bird species hunted in December and January. The hunt of the starling, forbidden in the other Italian regions, has been authorised by derogation in the provinces of Bari and Brindisi, during the hunting seasons 2017/2018, in order to restrain the damage caused by these birds on olive crops in the area. The research meant to evaluate meat quality traits of thrush (T), woodcock (W) and starling (S) hunted in Apulia during the hunting season 2017/2018. Meat pH, colour, chemical and fatty acid composition were assessed on the *Pectoralis major* muscle in eight birds of each species. The pH value (6.38) was significantly $p< .01$) higher in meat from S as compared to W (5.77) and T (5.70). Meat from T was significantly ($p< .01$) darker (28.4 vs. 21.4–23.1, respectively for S and W) and had higher redness (15.2 vs. 3.7–4.7, respectively for S and W) and yellowness (7.8 vs. 3.6–3.9, respectively for W and S). T meat chemical composition showed the highest amount of fat (4.7 vs. 0.7 and 1.9%; resp. for S and W; $p< .01$) while the lowest one of protein (18.5 vs. 22.2 and 21.7%, respectively for W and S; $p< .01$). Fatty acid profile of meat from T showed the best nutritional properties as compared to the other two birds since a significantly ($p< .01$) higher concentration of MUFA was recorded (72.4 vs. 47.1 and 49.4%, respectively for S and W), in turn of a lower SFA content (14.5 vs. 36.1 and 32.2%, respectively for W and S). Meat from S showed the highest ($p< .01$) PUFA concentration (20.8%) in comparison with both T (13.5%) and W (14.3%). While no difference aroused between species regarding the $\omega3$ concentration, meat from S showed a significantly ($p< .01$) greater content of $\omega6$ fatty acids (15.7 vs. 8.9 and 9.0, respectively for W and T). As a consequence, also the $\omega6/\omega3$ ratio was higher ($p< .01$) in S meat (3.5) as compared to both W (1.8) and T (2.4). Meat from T was the healthiest since it showed the lowest ($p< .01$) IA (0.08 vs. 0.50 and 0.17, respectively for W and S) and IT (0.11 vs. 0.30 and 0.26, respectively for W and S) indices. This result may be due to the different eating habits of thrush which prefers berries and fruits instead of insects that, on the other hand, are the main feeding source for woodcock and starling.

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P129

Ham weight loss at first salting: correlations with in vivo performances, carcase, and ham quality traits in Italian Large White heavy pigs

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Ham weight loss at first salting (HWLFS), defined as the weight loss during the first week of curing, is a quality criterion included in the selection programme of Italian heavy pigs to assess the aptitude of the meat for salting and seasoning. This study investigated the relationships between HWLFS and seventeen phenotypic parameters related to growth and feed efficiency, carcase and fresh ham quality such as pH values, glycogen and lactate content, glycolytic potential and cathepsin B activity obtained on the semimembranosus muscle of 270 performance tested Italian Large White heavy pigs. The animals did not carry the *RYR1* recessive allele and the *PRKAG3* 200Q allele. A multivariate analysis was used to determine residual correlations between traits. HWLFS was affected by day and weight at slaughter whereas sex and slaughter age did not affect its variability. HWLFS was significantly correlated with several traits: back fat thickness (r= –0.61), lean cuts (r = 0.60), ham weight at trimming (r = 0.49) and ham weight after first salting phase (r = 0.45). The correlation between HWLFS and fresh ham quality traits ranged from 0.14 (cathepsin B activity) to –0.30 (pH_u). Results from this study indicated that higher ham fat content and pH_u, and lower glycolytic potential could reduce HWLFS of green hams transformed in PDO products.

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ENVIRONMENTAL SUSTAINABILITY

P130

Use of different models to estimate phosphorus excretion in heavy pig

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Tools to quantify flows of nutrients at animal level are very useful to evaluate the environmental impact of livestock production. For the swine industry, phosphorus (P) is one of the most dangerous nutrients that affect several environmental aspects. To estimate the P potential excretion in heavy pigs using mathematical models, 15 intensive farms sited in the Italian north-western plain were studied for a fattening period (7 months) to collect data on management, feeding systems, pigs weight gain, diet composition and feed consumption. The data collected were at first used to calculate the P excretion at animal level using the following mass balance (MB): $P_{excretion} = P_{intake} – P_{retention}$. The result was then used to evaluate the reliability of two empirical models to estimate P excretion. The first equation (EQ1) was: $P_{excretion} = P_{feed} \times FCR_{head} \times ADG_{head} \times D_{fattening} – (0.24 + 0.004\ LW_{final} – 0.007\ LW_{initial})$; where: P_{feed} = P ration mean content (g/kg); FCR = Feed Conversion Rate (kg as feed/kg); ADG = Average Daily Gain (kg); LW = Live Weight of head at the beginning or at the end of the fattening period (kg); D = fattening days (*n*). The second equation (EQ2) was: $P_{excretion}=–0.467 + 0.905\ P_{intake}$; where: P_{intake}=P intake per head per fattening period (g). The collected data were analysed altogether and then comparing farms grouped according to mitigation strategies (phase feeding or phytase addition) adopted (S0 = 3, not adopting strategies; S1 = 6, adopting only phase feeding; S2 = 6, adopting phase feeding and phytase addition) by GLM ANOVA procedure. Statistical differences for P excretion were found between MB and EQ1 (p<.01), but no differences were found between EQ2 and the other models due to the great variability among the data. Mean P excretion obtained with MB (1966 g) were 24% lower than EQ1 (2561 g) and 15% lower than EQ2 (2309 g). Comparing the feeding management, no differences in P excretion were pointed out between S0, S1 and S2 using both MB (1670, 2011 and 2020 g, respectively), EQ1 (2277, 2627 and 2590 g, respectively) and EQ2 (2067, 2403 and 2297 g, respectively). The overestimation obtained using EQ1 and EQ2 recommends the use of MB to estimate P excretion. Anyway, both MB and the two mathematical models tested in this study do not highlight the advantages that could derive from the adoption of environmental pollution mitigation strategies for pig farms due to the several variables that could affect P excretion in the different farms.

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Use of non-medicated feed with the addition of pomegranate seed extract in commercial rabbit fattening

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Following the EU tendency, a specific Spanish national strategy has been developed to minimise the use of medical products in production animals. We tested whether fattening commercial rabbits with non-medicated feed with the addition of 20% of pomegranate seed extract (POM, *n* = 18) affected productive traits and meat quality compared with a conventional industrial fattening strategy that uses medicated concentrate (CON, *n* 18). The study includes three replicates per treatment and the groups were equilibrated for initial live weight. The effect of replicate was tested before and was not significant. Finally, a one-way general linear model was used, with feeding strategy as the fixed factor (POM or CON). We assessed productive traits (average daily gain and conversion index) during the 30 days of fattening period, ultimate meat pH, meat colour in the CIELAB colour space, meat texture by *Warner Bratzler* and cooking losses. We also assessed sensory and fatty acid composition, but these results are not presented in this abstract. Productive and meat quality variables were similar between the two groups (*p*>.05). Only slight differences for total consumption were observed (higher in POM group), even the conversion index was similar. POM group presented slightly higher (*p*>.07) carcase yield (62%) compared with CON (60%). We conclude that we can substitute 20% of the industrial expensive concentrate with pomegranate seed extract (10 times cheaper), avoiding completely the use of medicated concentrate with similar o slightly better results than the conventional strategy of industrial fattening. POM strategy of rabbits fattening has another added value because the use of a by-product of local traditional production of pomegranate, which is rich in polyphenols.

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Organic livestock production: a bibliometric analysis

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The organic livestock sector in the European Union has been experiencing a fast growth and, lately, organic farming has become a trending topic. A bibliometric review has been performed to summarise the research developed on organic livestock farming. The bibliographic search was done on Web of Science during April 2018 selecting those papers that dealt with organic livestock. The analysis was carried out with the package 'bibliometrix' for R.

Since 1993, a total of 306 research outputs (254 journal articles and 52 proceeding articles) have been published focussing on organic livestock farming, with an annual growth rate of 1.70% and a clear increase since 2005, which seems to follow the increasing interest on that topic by consumers and markets. The 254 papers have been published in 109 journals, and only 61.5% of them are in journals of the 1st and 2nd quartile of the relevant subject area. Germany is the country with more papers published on organic livestock farming (50 documents), followed by Denmark (29), France (28) and the Netherlands (17). In Europe, the most important species reared organically, with the exception of poultry, are sheep (42%) – mainly in United Kingdom – and cattle (34%) – mainly in Germany and Austria, followed by pigs (9%) – mainly in Denmark – and goats (7%) – mainly in Greece. Authors' top 10 keywords were: organic farming (74 times), organic, animal welfare, animal health, cattle, livestock, farming, organic agriculture, organic livestock production and health (10 times each). Even if poultry is the most important species reared organically in Europe only the term 'cattle' appeared as an indicator of the species studied within those keywords. This could indicate that more research has been done in cattle because of the importance of this species in Germany. Moreover, the presence of the terms 'animal welfare' and 'animal health' within those keywords seems to indicate that the research on organic livestock production has been focussing on these two areas, which are the major concern for consumers on organic farming. This bibliometric analysis revealed that: (i) countries focalised their research on their main production, (ii) more research on organic livestock production in other species than cattle is needed, and (iii) more and more high-quality research on this topic is needed.

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Role of zinc and copper in the intensive swine production systems

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Animal manure represents a possible route of heavy metals and metalloids (HMMs) diffusion into the environment. HMMs can be released into groundwater, absorbed by crops and enter into food chain and could cause problems for animal and human health. Some HMMs are essential nutrients and they are widely used as additives. Nevertheless, after the digestive process, not absorbed elements are released in manure. The aim of this study was to estimate the input and the output of HMMs in swine production in order to establish strategy to implement the sustainability of livestock.

Samples of feed ($n=16$), faeces ($n=80$) and water ($n=4$) were collected from four typical swine farms located in northern Italy. The collection of samples (in airtight nylon bags) was carried out considering the representativeness of matrix according to AOAC procedure. Samples were analysed for the principal components (AOAC, 2005; 152/2009). Mineralised samples (Microwave Digestion System) and water samples were evaluated by Inductively Coupled Plasma Mass Spectrometry (Bruker Aurora

M90 ICP-MS), in triplicate, for the detection of Na, Mg, K, Ca, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Mo, Cd and Pb.

The principal nutrients' analysis presented proper composition linked with the swine nutritional requirements (NRC 2012). The undesirable elements (As, Cd, Pb, Co, Ni, Mo) did not exceed the thresholds levels (2002/32/EC) in the feed samples. Zinc (Zn) and copper (Cu) resulted widely applied in swine diets for their Q40 positive impact to help young animals to cope with pathogens. The evaluated Zn content of feed samples, probably related to its pharmacological use, was 821.74 ± 301.27 mg/kg (as fed) and 1737.88 ± 301.27 mg/kg (as fed) in the weaning and finishing phases, respectively. Pig diets also presented a high Cu concentration in the finishing (133.75 ± 11.55 mg/kg as fed) and in the weaning (160.09 ± 11.55 mg/kg as fed). The content of HMMs in faeces reflected their presence in the diet and swine manure represents a possible source of Zn and Cu pollution. In conclusion, Zn and Cu should be used as nutritional additives in a concentration lower than the maximum admitted level. Thus, alternative strategies are required to guarantee the health status of growing piglets, in line with the recent European Regulations (Reg. 1039/2018; EMA/394961/2017) aimed to reduce the total Cu level in complete feed and to ban the pharmacological use of ZnO.

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Environmental impact assessment of goat milk production

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Goat farms are spreading as an alternative to other milk production pathways. Given the wide interest in goat milk quality and composition and the different characteristics of goats breeding, their presence in intensive farming systems is increasing. Similarly, to food, environmental assessments play an important role also in goat milk production. However, very few studies have been performed on the environmental sustainability of goat farming systems and efficiency improvements are searched on several points of view, among which breeding solutions and milk production.

In this study, the aim is to evaluate the environmental impact of goat milk production, taking into account all the livestock breeding phases. The environmental impact was quantified by means of the Life Cycle Assessment method. An attributional approach was used with a 'from cradle to gate' system boundary. The selected Functional Unit of 1 kg of Fat and Protein Corrected Milk (FPCM) was used, and all inputs and outputs for milk production were referred to this unit. All data were gathered from questionnaires to farmers and using specific models for goat emissions (Tier 1 and Tier 2 by IPCC, 2006 and EEA, 2014) about enteric methane, manure storage and spreading on field, nitrate and phosphates to water. Also for feed intake and diets data were collected on farm and evaluated through specific models. Background data were taken from Ecoinvent database v3.0.

Twenty goat farms in Lombardy Region were visited to collect the inventory data on crops cultivation (area, yield, etc.) and goat livestock management (herd composition, milk production, effective length of the milking period, etc.), as well as on the composition of the diets. The ILCD characterisation method was used for the environmental impact assessment.

The environmental impact results showed a wide variability, which is broadly due to the differences among the studied farms in terms of herd dimension (from 26 to 450 goats), milk production (from 633 to 3817 g FPCM/head day), crops cultivation and livestock management. Enteric methane emission was estimated as 11.9 ± 1.78 kg/head year, following the equation suggested by INRA (2018), based on diet characteristics and dry matter intake. Environmental impact assessment can be a useful tool in order to improve farm management and to look for innovative mitigation strategies.

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LIFE TTGG: improving the supply chain efficiency of European hard or semi-hard PDO cheeses by development of an Environmental Decision Supporting System (EDSS) for Product Environmental Footprint (PEF) assessment

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In order to limit global warming, the reduction of GHG emissions in the coming decades will have to be very consistent and should cover all sectors: not only the energy sector, which is responsible for the largest quantities of direct emissions in global terms, but also the agricultural sector, which today is a direct source of about 10–12% of total emissions worldwide. Available projections indicate that an increase in food consumption, without action, will rise GHG from the agricultural sector.

The European dairy sector represents one of the principal players in the world in terms of both importation and exportation. In the European ranking Italy and France are big European cheese producers, Italy has the sixth highest production in terms of cheese in general. The Grana Padano PDO accounting for ~24% of total milk output in Italy, is one of the most important cheese in the country. The EU has founded the 'LIFE TTGG' project, which aims to reduce the carbon footprint of hard and semi-hard cheese production by 10% (111,000 tons CO_2 eq/y) in the following 2/3 years after the project considering: the 25% of Grana Padano production, the 5% of other IT hard cheese PDOs, the 10% of FR hard cheese PDOs, involved in PEF reduction. Solutions are needed to improve Italian and France cheeses supply chain efficiency and to analyse and reduce their Product Environmental Footprint (PEF). In this regard the project proposed underlines the importance of implementing environmental solutions to improve the Grana Padano PDO's supply chain efficiency, supporting the development of a tool (EDSS) for PEF analysis and reduction.

The project, coordinated by Politecnico di Milano, provides the development of a life-cycle analysis (LCA) on a representative sample of Grana Padano PDO (65 farms, 18 dairies and 18 packaging producers) and on a French hard or semi-hard PDO cheese (35 farms and 15 dairies); the implementation of a Life-Cycle Inventory (LCI) datasets for the production of 1 L of cows' milk; the production of two of main feeds used for cows; the production of 1 kg of Grana Padano PDO; the production of 1 kg of French hard or semi-hard PDO cheese, the production of three packaging solutions. The LIFE TTGG project will be a useful contribution to the implementation of European environmental policies and also represents an opportunity to test Product Environmental Footprint Category Rules (PEFCR).

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Mineral nutrient content of tomato plants in the aquaponics system as affected by the fish density

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Aquaponics is a technique combining fish rearing with the hydroponic cultivation of plants, especially vegetables, in a closed system based on a virtuous recycling of water and nutrients, according to the principles of the circular economy. In an aquaponic unit, wastewater from aquaculture, after being filtered mechanically in order to remove solid waste, passes through a bio-filter, where nitrifying bacteria convert ammonia into nitrate, then is used as a nutrient solution by the plants, and finally comes back to the fish tank, purified. Aquaponics is acknowledged as an innovative sustainable food production system with high potentiality for facing problems like population rise, climate change, soil degradation, water scarcity and food security, especially in developing countries. The main strength of this production system is that two different kinds of products are simultaneously obtained, allowing the virtuous use of resources and then the sustainability of the food chain.

The present study reports the results of the production of Nile tilapia (*Oreochromis aureus* Steindachner, 1864) and tomato (*Lycopersicon esculentum* L. Newton) in the classical aquaponic system (1 loop) with different fish density with emphasising the growth and mineral nutrient content of tomato plant. The experiment was conducted at the Faculty of Agriculture of Ankara University (Turkey), using in-door, small-scale aquaponic systems with coupled system.

Ninety-six tilapia (*O. aureus*) juveniles around 7 g of individual weight were stocked at different ratio: 25 kg/m^3 (Group I), 35 kg/m^3 (Group II) and 50 kg/m^3 (Group III) and fed with 45% raw protein feed at the level of 2% body weight for 126 days. Fish density affected the fish growth with better values in the highest fish density, however, the overall calculation of the condition factor did not show any significant difference due to fish density. Water quality parameters measured fluctuated in all experimental groups during the experiment. Total plant biomass was low due to the insufficient lighting of the in-door aquaponics system and the low level of water potassium. An increase in fish stocking rate caused significant increases in tomato plant leaves growth. The mineral nutrient content in the plant was altered by the fish stocking rate. The mineral nutrient content of the leaves of the plant was at the sufficiency rate with the exception of potassium when compared to tomato plants grown in soil.

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SheeptoShip LIFE – looking for an eco-sustainable sheep supply chain: preliminary results on the sustainability of dairy sheep diet

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SheepToShip LIFE project aimed at the mitigation of greenhouse gas emissions of dairy sheep value chain in Sardinia. Twenty farms located in contrasting pedo-climatic zones, were surveyed in year 2017 to gather primary data 'from cradle to farm gate' for Life Cycle Assessment. This work focuses on two case study farms whose data were analysed to compute indicators of sheep feeding related to the three dimensions of animal diet sustainability according to the Food and Agriculture Organisation: (i) *planet* - Carbon Footprint (CF), measured as kg of CO_2eq/kg of fat and protein corrected milk (FPCM), and use of local feed, as percentage of self-sufficiency; (ii) *people* - percentage of non-human edible feed; (iii) *profit* - ratio between total farm income and variable feed costs.

The farms, located in North Sardinia lowland, have an area of 72 and 52 ha, a flock size of 396 (375 ewes; 21 rams) and 255 adult head (248 ewes; 7 rams), and a stocking rates of 7.6 and 3.5 head/ha, in A and B respectively. FPCM yield per present ewe was 194 (A) and 154 (B) kg. Both farms used annual cereal-legume mixtures (20.0 and 36.3% of farm area), and legume monocultures (22.6 and 13.9%). Supplements consisted of hay, home-grown Q42 and purchased concentrates. Milk CF, based IPCC (2006) and 100% allocated to milk, was 3.1 and 3.3 kg CO_2eq/kg FPCM for A and B, respectively. These data are lower by 29% (A) and 23% (B) than the project baseline estimated for Sardinian sheep sector. Animal and manure emissions accounted for most of CF (63%, A; and 71%, B). Using protein mass allocation of milk, meat and wool, CF allocated to milk amounted to 2.7 (A) and 2.8 (B) kg CO_2eq/kg FPCM. The other sustainability indicators for A and B, were, respectively: (1) protein feed self-sufficiency equal to 62% and 75% of crude protein (CP); (2) non-human edible feed (including all forages and 35% of pelleted concentrates) equal to 75% and 76% of CP intake; (3) the ratio between total farm income and variable feed costs (local price-based) equal to 1.67 and 2.03.

These preliminary results suggest that dairy sheep farms from the same area but differing in forage systems and stocking rate converge towards high production sustainability according to the indicators chosen.

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Water use of livestock production system in Italy

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Water used in animal husbandry is the result of the sum of the volumes of water resources used to meet animals' demand and water to wash structures and equipment. In the livestock sector, the uses of water can also be influenced by environmental and management factors such as the air temperature and relative humidity, rearing technique (extensive or intensive), housing systems, production targets (e.g. meat or milk for cattle, meat or eggs for poultry) and feeding management. The content of water in the diet of animals, depending on the composition of the ration and the type of feedstuff, influences water demand, as the supply of feed with a relatively high humidity content reduces the amount of water required by the livestock. Based on official data on farm water consumption for the year 2016, in this work, we attempt to estimate the water requirements of the national livestock sector. The first estimates show that, in 2016, the volume of water used in animal husbandry was 318 million cubic metres. Cattle farming (5.7 millions of bovines) absorbed just under two thirds (66.0%) of the total volume used by Italian livestock. About 17.7% of the water volume was allocated to 8.4 millions of pigs and sows farming. Below a ten per cent threshold, main water consumption animal groups were buffaloes (overall 382 thousand that used 5.9% of the total water volume), 15.4 millions of poultry (4.8%) and 7.0 millions of sheep (3.9%). The case of goats, rabbits, and ostriches is quite marginal, with overall just over a million cubic metres of water used (about 0.3% of the total). Compared to 2013, the volumes of water used by animal farming increased by about 5.4%, although the total number of live animals decreased by 3.3%; this is due to the increase, in the three-year period analysed, of the species with a larger water footprint, in particular, dairy cows (+15.1%) and buffaloes (+5.7%). The territorial data analysis shows that farms in Northern Italy made use of slightly less than 70% of the total amount of water used, in parallel with the greater presence of livestock farming in these areas. In 2016, two regions, Lombardia and Veneto accounted altogether for 40.7% of water uses, respectively 28.0 and 12.7% of the total; these two regions are also characterised for the highest shares of cattle (18.3% and 36.9% of the total nationwide).

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Trend in land use and land cover change in Mediterranean rangelands: the case of Campo di Segni, Lepini mountains (Central Italy)

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Over the past five decades, European landscapes have undergone significant changes, largely due to natural and socio-economic dynamic processes. Marginalisation and abandonment of small and extensive agrosilvopastoral activities in mountain zones lead to a spontaneous reforestation of secondary rangelands and arable lands.

In this study we investigate landscape evolution in a mountainous area of central Italy (Campo di Segni, Lepini mountains, 3570 ha, elevation 600–1300 m a.s.l.), which has been shaped by centuries of animal and human presence and where an agrozootechnical activity is still present, albeit strongly resized (from 644.7 Animal Unit in 1977 to 499.3 in 2016).

In order to analyse landscape changes, we built a GIS database based on contemporary (2016) and past (1954) maps. The analysis spans a 62 years period. We used aerial photos and field surveys to map land-use/land-cover (LULC) classes, and a transition matrix to analyse gains and losses of each class over time. LULC classes were visually identified.

Our results show that, at lower altitudes, human settlements have increased (+99.2 ha), as new constructions have invaded croplands (–36.5 ha), chestnut groves (–37.9 ha) and, to a lesser extent, woodlands (–11.2 ha), rangelands (–19.5 ha) and other classes. A quote of arable lands (26.9 ha) has been replaced, for the most part, by rangelands (+11.4 ha) and chestnut groves (+14.1 ha). Due to the abandonment of cultivation, a quota of about 50 ha of chestnut groves has turned into forest. Regarding the general increase of wooded areas (+578.4 ha), we appreciated a different evolution of closed compared to open woodlands. Due to the transition to forest, the significant extensions of grazing land that existed in 1954 (1450.4 ha) have almost halved, decreasing to 731.4 ha only in 2016. The traditional structures (‘*cese*’, ‘*volubri*’, farm facilities) are still used for the existing pastoral activity.

Our analysis shows a dramatic change of the landscape in the study area, with the reduction of open and semi-open rangelands and a rapid and intense spontaneous reforestation. The causes are to be found in the reduction of the grazing load. Management decisions to support extensive agrosilvopastoral activities must be taken as soon as possible, to avoid complete abandonment and consequent loss of ecosystem services.

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Does the application of BEEF CARBON strategies reduce the carbon footprint? Results of the first year of activity

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A large number of nations have committed to keep the increase of the temperature of the preindustrial age lower than 1.5 °C and to cut at least 30% of the greenhouse gas (GHG) emissions in 1995 by 2030. Beef production is one of the main contributors of GHG emissions in agriculture. The main sources of emissions are methane from enteric fermentation and decomposition of manure organic matter, and nitrous oxide from manure management, and from fertiliser application. In addition, beef production causes indirect emissions of carbon dioxide from combustion of fossil fuels. There is a long list of strategies for reducing GHG emissions and increasing soil carbon sequestration. One hundred and seventy beef farmers have committed to reduce GHG emissions by 15% in France, Ireland, Italy and Spain within 10 years, by adopting one or more of available strategies. In Italy, carbon footprints of 20 intensive fattening farms located in Veneto and Piemonte were estimated before and after the application of a series of mitigation strategies, most of which aim to improve animal welfare, increase renewable energy, reduce synthetic fertilisers, increase animal performances. Life cycle assessment was estimated utilising Calcul des AUTOMATISE Performances Environnementales en Elevage de Ruminants-CAP2ER® (*Niveau* 2) developed at the French Institut de l’Elevage. The functional unit was 1 kg of live weight gain (LWG) and the environmental categories are: global warming, acidification, eutrophication, energy consumption, biodiversity and carbon storage. The activity was carried out in collaboration with ASPROCARNE and UNICARVE. At the beginning of the project, farms’ mean size was 66 ± 66 ha and 468 ± 334 places; the average LWG was 1.22 ± 0.21 and average live weight production was 213 ± 159 T/year.

The estimated carbon footprint was 8.61 ± 1.55 and 7.99 ± 1.48 kg CO_2eq/kg LWG before and after the application of the mitigation strategies, respectively, with a reduction of about 7%. The farms combining more than one mitigation strategies demonstrate to be able to achieve the goal of this project to reduce the carbon footprint by 15%.

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Influence of zeolite on ammonia concentrations, gains and mortality of animals in piglet housing

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Ammonia (NH_3) has a negative impact on the environment, performance and health of animals. In order to decrease its concentrations in piglet housing, we applied zeolite–clinoptilolide (0.5 kg per m^2 of floor area, three times, seven-day interval) into the slurry stored in the slurry pits of an experimental Q43 hall. A second hall was control without its application (both halls: 24 pens, 23 piglets per pen, forced ventilation, slatted floor, complete feed mixture). Ammonia concentration, temperature and humidity were measured every 60 minutes during 21 days using calibrated instruments placed in 3 locations (flaps, the middle of the hall and door). We found that the average weekly NH_3 concentrations ($mg.m^{-3}$) decreased with the fattening time (from 1st to 3rd week) in all locations of both halls. In the 1st and 2nd weeks, they were lower in the experimental hall (flaps: 3.78 ± 0.59, 3.21 ± 0.56; door: 5.35 ± 0.75, 4.40 ± 0.64) than in the control hall (flaps: 7.20 ± 0.96, 5.00 ± 0.88; door: 7.94 ± 1.63, 5.56 ± 1.40) with significance $p<.001$. In the 3rd week, the average weekly NH_3 concentrations were higher in the experimental hall (flaps: 3.17 ± 0.48, door: 3.44 ± 0.49) than in the control hall (flaps: 3.15 ± 0.62, door: 3.12 ± 0.78) with significance only for door location ($p<.05$). In the middle of the halls, the average weekly NH_3 concentrations were lower in the experimental hall (3.11 ± 0.56; 2.59 ± 0.30; 1.90 ± 0.31) than in the control hall (4.24 ± 0.54; 3.22 ± 0.64; 2.87 ± 0.39) in all weeks with significance $p<.001$. We also identified very high significant ($p<.001$) differences in NH_3 concentrations between individual weeks in all locations of both halls. Next, we detected that the average daily gains (kg) per piglet were the same in both halls and amounted 0.51 ± 0.05, 0.51 ± 0.07 in the experimental and control hall, respectively. The piglet mortality was not significantly different ($p=.25$) between halls and reached 2.8 % and 1.7 % in the experimental and control hall, respectively. Lower NH_3 concentrations were detected in the experimental hall (except 3rd week, location – flaps, door). This was caused by the application of zeolite which bound ammonia and did not allow its release to the housing area. Application of zeolite reduced NH_3 concentrations by 27% in the housing area, but its positive effect on gains and mortality of piglets was not demonstrated.

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LIVESTOCK SYSTEMS

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Managing future dairy farms producing more than 22,000 litres of milk per cow

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High managerial standards are fundamental in dairy farms to achieve high production levels. Annual increases of rolling herd averages will lead to very productive farms and challenging managerial choices. Nutritional and reproduction are the most important area to take into account in first instance. The most productive farm in the world already reached herd average of about 20,000 kg/cows per year. This work assumed that the top 100 Italian Holsteins for production level could represent the average herd of a future farm. Production records of 400 lactations of the top 100 pluriparous cows from the years 2014–2017 of the Italian farmer Association (AIA) database were analysed. According to previously published classifications, animals were divided into two groups characterised by Standard lactations (with length <390 DIM; $n = 197$) and Extended lactations (with length between 390 and 700 DIM; $n = 193$).

Standard lactations showed average milk yield of 20,065 ± 1284 kg/head of cumulative milk production and 678 ± 133 and 620 ± 61 kg/head of fat and protein in 341 ± 28 DIM from parturition to dry-off. Average daily yield was equal to 58.9 ± 3.0 kg/d of milk per cow with 3.38 ± 0.63% of fat and 3.09 ± 0.23 % of protein. Extended lactations showed average milk yield of 25,166 ± 3045 kg/head of cumulative milk production and 843 ± 188 and 800 ± 135 kg/head of fat and protein in 480 ± 79 DIM from parturition to dry-off. Daily milk yield was on average equal to 52.4 ± 3.0 kg/d of milk per cow with 3.35 ± 0.56% of fat and 3.18 ± 0.25 % of protein.

Cumulative production at 100 DIM was similar for both groups and close to 6750 ± 600 kg of milk per head.

Assuming that cows could reach 700 kg of BW, estimated DMI based on their average production level using NRC (2001) equations, will result in 33.0 and 30.9 kg/d per head for cows with Standard and Extended lactations, respectively. Assuming an average period of 45 dry days, the average day at conception resulted approximately close to 100 and 240 DIM for cows with Standard and Extended lactations, respectively. These results focus on the need to adequately manage nutrition in order to avoid metabolic disorders related to energy balance and also to specifically focus on reproduction. In particular, a specific target will be the early estimation of animal persistency (around 100 DIM) in order to manage the optimum insemination time for each individual cow and benefit from persistent lactations of high producing animals.

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Relationship between resting time in dry period, health status and inflammo-metabolic profile in early lactation of dairy cows

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In modern dairy farms, sensors to monitor physiological characteristics of cows have been introduced with the aim to facilitate the assessment of behaviour and health status. Resting is one of the most relevant activity of dairy cows, that spend about 50–60% of the day lying. It can be reduced by many factors, such as overcrowding, competition, inadequate environment and health status. Despite the resting time (RT) may be a useful indicator of the cow condition, at the moment it is poorly studied in transition period. This research aimed to investigate the relationship between RT and health status in periparturient dairy cow.

The RT of 34 Holstein multiparous cows was measured with AfiTag II pedometer (SAE Afikim, Israel) from –21 to 28 days from parturition (DFP). Blood samples were collected from jugular vein at –14, –3, 1, 3, 7 and 28 DFP for the analysis of the inflammo-metabolic profile. Body condition score (BCS) and body temperature were measured after each blood sampling and milk yield was recorded daily. Cows were retrospectively divided into three groups based on the first clinical disease diagnosed in the first month of lactation: control group (CTR, 13 cows without clinical disease); retained placenta (RP, 10 cows); metritis (MET, 11 cows). Data were evaluated by ANOVA using the MIXED procedure (SAS Inst.). Q44

The MET's RT showed a similar trend to CTR, while RP showed a greater RT compared to CTR in dry period (837 vs. 735 min/d; $p<.05$) and during the first week of lactation (741 vs. 659 min/d; $p<.01$). After that no differences among groups were observed and the mean resting time was 646 min/d. No differences were observed in milk production. Cows with a uterine disease (RP and MET) expressed a more severe inflammatory response after calving, with the increase of haptoglobin ($p<.01$ at 7 DFP) and the reduction of many biomarkers of the negative acute phase response (albumins, paraoxonase, cholesterol, zinc, vitamin A and E; $p<.05$). Moreover, RP showed higher body fat mobilisation (higher NEFA), lower efficiency of fat oxidation (higher ρ-hydroxybutyric acid) around calving and higher body temperature at 7 DFP ($p<.01$). The measurement of RT in dry period confirms its utility as prognostic index of some diseases such as RP, but studies are required to define the reference ranges of RT in healthy cows in different stages of lactation and in different environment conditions.

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Thermography analysis of silage: methodological aspects

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Analysis of face silage temperature during feed-out period could be a way to detect aerobic spoilage related to temperature variations. Besides the traditional use of visual inspection to detect critical zones, a global silo diagnostic may be an interesting tool to summarise structural and management problems.

Sixteen bunker silos filled with different silages (whole corn $n=8$; whole ear corn $n=5$; wheat $n=3$) were examined using thermography to identify descriptors able to quantify the risk for aerobic spoilage related to structural or management characteristics. Infra-red images were captured 1 hour after bunker unloading process during the early hours of winter days. Face silage images were normalised and the main descriptive features (FE) were extracted. Notable correlation between Maximum (Tmax) and Mean Temperatures was detected (Pearson coeff. 0.6718, $p=.004$), meanwhile no correlation was notable between

Minimum vs. Mean Temperatures and Tmax vs. Geometric Means. Vertical (Vgrad) and horizontal (Hgrad) temperature gradients were evaluated as well: a qualitative analysis, based on data plots, showed different spatial distribution of gradients where Vgrad displayed widest variations than Hgrad. Considering both descriptive FE and others from gradient matrices, a total number of 14 FE were explored by principal component analysis (PCA): the first two components already explained the 77.93% of total variance; moreover, 96.70% was reached by the first four principal components. PCA revealed a cluster formed by 3 bunkers; their visual technical inspection reported badly maintained and unusual building. A principal factor analysis (PFA) was carried out on the same dataset (16 observations and 14 FE): five principal factors (PF), explaining the 86%, were retained; the first two patterns were mostly populated (5 FE each) with greater scoring coefficients. In the first PF pattern, Standard Deviation and Tmax were linked to minimum gradients values; in the second pattern three over four FE related to Hgrad were grouped and combined with location measures (Mean and Mode).

We explored methods and achieved the preliminary result that descriptive FE are interesting and probably sufficient to investigate face silage temperature. Furthermore, using an unsupervised statistical approach, we identified 3 defective situations related to structural issues and independent of bunker content.

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Reduction of concentrated feed in dairy cows in the Parmigiano Reggiano area: effects on milk production and quality‡

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‡ A part of the results of the research was presented at 6th AITeL
Q45 Congress (Trento, September 20th 2018).

A project of technological research, funded by Emilia Romagna Region and aimed to a higher sustainability of dairy chain, involved an experimental trial testing the effect of a reduction of concentrate feed in Frisona cows producing milk for Parmigiano Reggiano Cheese making. Two groups of 15 cows each, with milk production at 9 days in milk (DIM) of 31.7 and 32.1 kg/d, were fed mixed meadow hay *ad libitum* and a growing amount of concentrate feed up to 15 kg (C group) or 12 kg (T group) at the peak of lactation (about 60 days). Milk production, milk fat and protein were recorded from each cow at 9, 47, 96, 131, 166, and 207 DIM; fatty acid composition of milk was analysed on 5 pooled samples/group at 47, 131, and 207 DIM. Milk collected from the two groups of cows at day 161 and 168 was used for micro cheese making. Milk production and milk fat were unaffected by diet, whereas milk protein was lower in T cows at 166 (3.18 vs. 3.39%; $p<.01$) and 207 (3.17 vs. 3.31%; $p<.05$) DIM. Fatty acid composition of T milk showed at 47 DIM significantly lower % of MUFA and PUFA, and higher % of SFA, resulting in higher SFA/UFA ratio (1.70 vs. 1.38; $p<.01$). Both n-3 and n-6 fatty acids were significantly lower in T milk at 47 DIM (n-3: 1.21 vs. 1.44%; n-6: 4.05 vs. 4.57%) and at 131 DIM (n-3: 1.18 vs. 1.37%: n-6: 3.89 vs. 4.83%). CLA levels were similar in the milk of the two groups. The milk collected for cheese making (60 l/group) had similar chemical and bacteriological composition, with the exception of a higher content of thermophilic Lactobacilli in T milk. Rennet coagulation characteristics showed lower curd firmness at 30' in T milk (a_{30}: 24.80 vs. 29.48 mm; $p<.05$), whereas the other cheese making traits were not significantly different between the two groups. Bacteriological traits of the ripened cheese (60 days) from the two groups were also similar.

The results of this field trial indicate that a reduction of 20% of concentrate feed in the diet of high producing cows did not affect milk amount, milk fat and cheese making traits, but caused a partial deterioration in milk protein, fatty acid composition and rennet coagulation. Further research might evaluate the whole lactation, reproductive performances, nutritional and health status.

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P146

Effects of a GH polymorphism on milk production traits in Modicana and Cinisara cows reared in different feeding systems

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GH gene encodes for growth hormone, a member of somatotropin/prolactin family of hormones, which plays an important role in milk production and metabolism. In bovine a single nucleotide polymorphism (SNP) in exon 5 (g.2141C>G) in GH gene changes the codon CTG to GTG and a Leucine to Valine (L127V) in the mature GH molecule. The aim of the study was to evaluate, in Modicana and Cinisara cows, the effects of genetic polymorphism at GH locus and its interaction with feeding system on milk traits. 155 individual blood samples were collected from 97 Modicana cows (32 in an extensive and 65 in a semi-extensive farm) and from 58 Cinisara cows (32 in 7 extensive and 26 in 6 semi-extensive farms). In the extensive, farms cows were fed exclusively with pasture; in the semi-extensive farms with pasture, hay and concentrate. Bovine GH (bGH) was analysed at the 5th exon according to Komisarek et al. (2011). In Modicana, monthly individual milk yield was recorded and individual milk samples were collected. In Cinisara, milk production was recorded once, the day of samples collection. Fat and protein were determined (Combi-foss 6000, Foss Electric, Hillerød, Denmark). In Modicana, milk yield, fat and protein were analysed by GLM procedure for repeated measures of SPSS (SPSS for Windows, SPSS Inc., Chicago, IL, USA). In Cinisara, milk yield, fat and protein were analysed by the univariate GLM procedure of SPSS. The analyses included main effects of GH, feeding system and the interaction GH × feeding system. Genotype frequencies were: LL, 0.61 and 0.34; LV, 0.37 and 0.57; VV, 0.02 and 0.09, respectively in Modicana and Cinisara. Although in both breeds milk yield was lower in VV cows by 2–3 kg/d (respectively in Modicana and Cinisara: LL 9.06 and 12.3, LV 9.24 and 11.4, VV 7.61 and 8.33), these differences did not reach the statistical significance, probably due to the low number of data. Also, milk gross composition was not affected by GH genotype (respectively in Modicana and Cinisara: fat, LL 4.08 and 3.32, LV 3.91 and 3.13, VV 4.04 and 3.62; protein, LL 3.67 and 3.51, LV 3.66 and 3.56, VV 3.64 and 3.71). Results are in line with the literature. The higher energy level of the diets offered in the semi-extensive farms did not interfere with GH genotype effect on milk traits: no significant interaction was found between genotype and feeding system. Nevertheless, the trend that emerged for milk yield needs to be confirmed in further investigations involving more animals.

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Effect of caspase-3 inhibitor Z-VAD-FMK on cryotolerance of in vitro bovine matured oocytes

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The aim of the work was to evaluate whether prevention of cryopreservation-induced apoptosis, by the caspase inhibitor Z-VAD-FMK (ZVAD) during vitrification, warming and post-warming-culture, would improve the cryotolerance of in vitro matured bovine oocytes. This was assessed by evaluating mitochondrial membrane potential, DNA fragmentation, as well as viability and developmental competence after *in vitro* fertilization. Abattoir-derived cumulus-oocyte complexes (COCs) were matured in vitro Q46 for 21 h at 39 °C with 5% CO_2. After maturation COCs were divided into three groups: a group of non-vitrified oocytes (F-CTR), and two groups of vitrified oocytes, respectively untreated (V-CTR) and treated with 20 μM ZVAD (V-ZVAD). All oocytes were placed for 1 h in fresh maturation medium (IVM) in presence (V-ZVAD) or absence of Z-VAD (F-CTR and V-CTR). Oocytes of group V-CTR and V-ZVAD were partially denuded and then vitrified by Cryotop. After warming, vitrified oocytes were transferred for 2 h in IVM with 0 and 20 μM ZVAD, to allow recovery. DNA fragmentation was assessed by TUNEL staining (46/group) and mitochondrial membrane potential by JC-1 staining (54/group). In addition, oocytes (110/group over 4 replicates,) were *in vitro* fertilized and cultured according to standard procedures. Vitrification decreased (p<.05) the mitochondrial membrane potential in absence of treatment (V-CTR; 1.0 ± 0.1 pixel/mm_2), while exposure to the caspase inhibitor was efficient to maintain levels (V-ZVAD; 1.2 ± 0.1) comparable to the F-CTR group (1.3 ± 0.1). Vitrification increased (p<.01) apoptosis, as indicated by higher percentages of oocytes showing DNA fragmentation in both vitrified groups compared to the F-CTR group (22.7 ± 7.2, 20.5 ± 7.2 vs. 4.4 ± 3.0, respectively). However, no significant differences were recorded between the vitrification groups. Furthermore, cryopreservation decreased cleavage (73.1 ± 6.1, 42.4 ± 1.7, 47.7 ± 9.9; p<.05, respectively for F-CTR, V-CTR and V-ZVAD groups) and blastocyst rates (40.1 ± 8.5, 6.7 ± 2.0, 6.2 ± 2.1; p<.01, respectively F-CTR, V-CTR and V-ZVAD groups), without differences between the vitrification groups. These results demonstrated that the treatment with Z-VAD before, during and after vitrification of bovine oocytes, is not effective in preventing cryopreservation-induced apoptosis and hence improving oocyte cryotolerance, despite its beneficial effect on the mitochondrial membrane potential.

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An integrated approach to reduce the use of antimicrobials in Italian heavy pigs, the experience of Emilia Romagna

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The reduction of the antibiotics (ABs) use in livestock is a key point to counteract the occurrence of antimicrobial-resistance. The quantification of ABs is essential to evaluate the effectiveness of the Regional/National strategies. This work is part of a PSR project aimed in quantify the ABs use on a sample of Emilia-Romagna pig farms and support the farmers in a process of ABs reduction. ABs data were collected from 30 farms during 2016-2017. Quantitative raw data were converted in Defined Daily Dose Animal for Italy (DDDAit).

The data showed that the DDDAit administered during 2016 were: 19.0 for sows, 22.6 for fatteners, 26.0 for suckling piglets and the highest use was registered in weaners (60.5%), with 103.9 DDDAit, among which 26.8% were Highest Priority Critically Important Antimicrobials (HPCIAs). The most used routes of administration were premix in weaners (53.2%) and fatteners (59.5%), oral powder in sows (60.2%) and injection in suckling piglets (79.1%).

The data concerning 2017 showed a reduction in overall ABs use (−22.1%) and of HPCIA (−47.3%). Colistin consumption was almost eliminated (−95.3%) also as consequence of DMS n. 117/2016. A sharp decline was observed in weaners (−53.5% overall ABs; −88.9% HPCIA), sows (−21.3% overall ABs; −62.9 HPCIA) and fatteners (−16.35% overall ABs, −45.8% HPCIA). In contrast, an increase was reported for suckling piglets (+2.5% overall ABs, +8.4% HPCIA). Regarding the route of administration, a decrease of premix use in favours of oral powder (43.1% vs. 46.2%) was registered in weaners, for the other rearing classes no variation in proportions were registered respect 2016; in fatteners the premix remains the most used (50.2%), the oral powder remains the most used in sows (73.9%) and the injection in suckling piglets (93.3%).

The present results highlighted the possibility to reduce the ABs use in the pig rearing system, by proving consistent data to the farmers and Vets, as well as supporting them in the decision making. Weaning and suckling pigs still represent a sensible target for ABs reduction. The collection of further data concerning 2018 is still in progress, as well, data on welfare and biosecurity will be collected to define possible correlation between management and ABs use.

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Effects of insect meal and microalgae as dietary ingredients on gut histology of Nile tilapia (*Oreochromis niloticus*)

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Although the value of fish-in fish-out ratio of Nile tilapia (*Oreochromis niloticus*) is below 1 (0.4) its high aquaculture worldwide production still requires a huge amount of fish meal. Fish meal can be partially substituted by insect meal without affecting growth performance of tilapia, but little information is available about the effects on gut histology although its important role in absorption of dietary nutrients and in immunological status. In this work, we tested the effects of partial replacement of fish meal with insect meal from *Sarcophaga carnaria* on histological characteristics of anterior intestine of tilapia. The dietary supplement use of lyophilised microalgae *Nannochloropsis gaditana* was also assessed. One hundred twenty Nile tilapia fingerlings (4.5 g ± 1.0) were randomly distributed into 12 tanks (20 L each) equipped with mechanical and biological filters. The isonitrogenous experimental diets were: fish meal based diet (FM); fish meal diet supplemented at 3% of DM with microalgae (FMA); insect meal based (70% DM) diet (IM); insect meal diet supplemented at 3% of DM with microalgae (IMA). Each diet was randomly assigned to triplicate tanks. Fish were fed 3 times a day for 5 days a week and 3 fish/tank were sampled after 80 days for histological study. The gut samples were fixed in Davidson’s fluid and embedded in Paraplast. Serial sections of 6 microns were stained with Alcian Blue pH 2.5 and PAS. Goblet cells (CG) were counted from six villi. Transmission electron microscopy (TEM) images were also obtained using standard procedures. The intestinal quotient was significantly different ($p<.05$) only between FMA and IMA groups (6.041 ± 0.267 vs. 4.721 ± 0.327, respectively). The main differences among the diets were found for the GC number/villus: 13 for FM and FMA, 21 for IM and 7 for IMA. The TEM imagines highlighted more chylomicrons in enterocytes of both microalgae supplemented diets (IMA and FMA) indicating a higher lipid absorption. An acid mucous film on the top of the villus was observed in the IMA group that could indicate chemical action related to the intake of chitinous materials, while along the walls of the villus there is evidence of neutral mucous secretions. In conclusions, the tested diets did not substantially alter tilapia gut histology, but the observed differences in mucous secretions among groups need further study.

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Post-weaning rumen fermentation of calves and its relationship with rumination time measured by Hr-Tag system in response to weaning age

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The present experiment aimed to investigate the effect of weaning age on rumen fermentation, dietary intake, growth and rumination behaviour around 3 weeks after weaning in Simmental calves. The relationship between rumination time (RT) measured by the Hr-Tag rumination-monitoring system (Hr-Tag; SCR Engineers Ltd, Netanya, Israel) and fermentation characteristics were also evaluated. Ten Simmental calves were randomly assigned at birth to be weaned at 45 (EW) or 60d (NW). EW calves were offered 6 L of whole milk/d from d 2 to 38d and 3 L/d from d 39 to 45, while NW were offered 6 L of whole milk/d from d 2 to 53d and 3 L/d from d 54 to 60. On d 21 after weaning, samples of rumen fluid were obtained via an oesophageal tube. These samples were used to determine pH, volatile fatty acids (VFAs), and lactate. The Hr-Tag system continuously recorded the RT by Hr-Tag loggers positioned on the left side of the neck. Normalised data were subjected to SAS ver. 9.4 (SAS Institute Inc., Cary, NC). NW calves had higher final body weight (BW), average daily gain (ADG), and overall starter intake compared to EW group. No differences were detected for total VFAs, acetate, propionate, and butyrate, whereas both L- and D-lactate concentrations were higher in NW compared to EW calves. Daily RT (average of 3 before rumen fluid collection) resulted positively correlated to pH (r = 0.98), and negatively correlated to concentration of total VFAs (r= –0.90), acetate (r=-0.85), and propionate (r= –0.90). Results herein provide evidence that anticipating the weaning time in Simmental calves fed whole milk did not impair rumen fermentations after weaning. Thus, calves weaned earlier resulted in a greater rumen fermentation efficiency considering the lower amount of starter intake compared to calves weaned 15 days later. Surprisingly, the use of Hr-Tags opened a certain interest for the prediction of rumen fermentation characteristics in calves after weaning, obtaining quick information on rumen health (i.e. avoiding a marked decrease of rumen pH) during this critical phase, from liquid to only solid feeds, when rumen of calves is not completely developed and efficient. The provision of these results on Hr-Tags in calves offer inputs for further research involving more subjects and also using this device in studies with different diets.

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Environmental management and pododermatitis in a breeding farm for hares: microbiological aspects

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The breeding of the hare in a cage for restocking seems to obtain better results when animals are introduced into the territory; however, the wildness of this animal makes it less adaptable to this kind of breeding and more prone to stress, resulting in immunocompromission and, consequently, to infectious diseases.
The aim of this work was to study the bacteria involved in podal diseases and the environmental conditions eventually predisposing to these pathologies.
The samplings were carried out in a medium size breeding farm of hares for repopulation, located in Piedmont region, in the province of Novara.
The breeding is composed of 70 cages for pairs divided into 5 rows, composed from 7 bicellular cages each, for a total of 140 breeders and from 70 cages for weaning and young hares arranged in 5 rows composed of 7 bicellular cages. A total of 53 swabs was carried out from 49 hares (four animals presented lesions at both posterior paws) using swabs with transport medium. The samples were streaked onto Sheep Blood agar plates (Microbiol, Italy) at the laboratory, and then, incubated for

18–24 hours at 37 °C in aerobic condition in order to obtain the growth of isolated colonies. The results obtained show, as already reported in the literature above all for the rabbit, the link between the presence of podalic lesions and *Staphylococcus aureus*. In fact, in our study 16/53 samples (30%) result positive for *S. aureus*; moreover, we have isolated *Pseudomonas aeruginosa* (2%), *Klebsiella spp* (2%) and *S. pseudintermedius* (2%) too. Our data agree with what reported for podalic infections in rabbit, in the breeding hare and in free-living species. Moreover, in our conditions, the presence of cages with a flooring with grids needs particular attention because it could be a predisposing factor to podal lesions.

In conclusion, this work allows us to confirm that *S. aureus* is the main aethiological agent of pododermatitis also in hares bred in a cage. It would be interesting in genotyping *S. aureus* strains to identify the origin (human or animal or environmental) and also to intervene for decreasing the onset. Finally, it would be important to evaluate that the actions started to counteract the microorganisms have been effective using monitoring activities.

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Fillet fatty acids and volatile organic compounds of Atlantic salmon fed diets including *Hermetia illucens* larvae

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Insects can convert biomass into high-quality protein that is used in feed formulation. When insect meal is included in aquafeeds, even if partly defatted, it carries some amounts of saturated fatty acids (SFAs), which negatively affect fish fillet FA profile. Moreover, rainbow trout fed with insects was perceived as different by trained panellists, compared to fish fed a control diet based on fishmeal. The current study aimed to evaluate the effects of the replacement of fishmeal with increasing levels of partially defatted meal of *Hermetia illucens* larvae in the diet for seawater-phase Atlantic salmon (*Salmo salar* L.) on fillet FA composition and volatile organic compounds (VOCs). Four isoproteic, isoenergetic and isolipidic diets were formulated: a control diet (C, 0% of insect meal) and three diets with increasing substitution levels of fishmeal with partially defatted *H. illucens* larva meal (33, 66 or 100%; IM33, IM66 and IM100, respectively). Additional fish oil was included in the diets with less fishmeal (IM33, IM66 and IM100) to ensure sufficient dietary long-chained highly unsaturated FAs. Fish were fed for 17 weeks, then they were percussively slaughtered and filleted. Twelve fillets per treatment were sampled and analysed to determine neutral and polar FAs and to determine VOCs using a proton transfer reaction time-of-flight mass spectrometer (PTR-ToF-MS).

Neutral lipids in fillets were differently affected by the different diets. SFA, monounsaturated FA (MUFA) and polyunsaturated FAn3 (PUFAn3) contents were higher in IM100 than in C group, while PUFAn6 content showed an opposite trend ($p<.01$). As regards polar lipids, only MUFA and PUFAn6 contents changed between the dietary treatments, peaking in C and dropping in IM100 fillets ($p<.05$ and $p<.01$ for MUFA and PUFAn6, respectively).

Overall, VOC profile was different between the different dietary groups. In detail, defatted *H. illucens* inclusion increased the total amount of fragments detected by PTR-ToF-MS; formaldehyde, propanale and eptadienale contents were higher in IM100 in comparison to the C group ($p<.01$). Contrariwise, group H100 resulted in a diminished acetic acid content ($p<.05$).

In summary, the inclusion of defatted *H. illucens* meal in diets for Atlantic salmon determined modifications of the FA and VOC profile, although no detrimental changes were of notice.

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P153

Assessment of aboveground biomass production in the Italian Apennines grazed rangelands using Sentinel 2 satellites and vegetation indices

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Rangelands are one of the most prevalent and widespread land cover vegetation types of mountainous landscape, and, from an agricultural perspective, they provide the cheapest feed source for ruminant livestock. For a proper planning of the carrying capacity, the estimate of the aboveground herbaceous biomass is necessary, as well as sward height, phenological stage, productivity level, chemical and species composition.

Remote sensing and modelling approaches allow for large scale monitoring, quantification and prediction at varying temporal and spatial resolutions, letting a precision farming approach also for extensive grazing. Using Sentinel 2 satellite imagery, the aims of this study was to examine the relationship between various Vegetation Indices (VI) derived from spectral reflectance and the aboveground fresh biomass of mountain Apennine pasture. The most performing VI was used to derive maps of herbage productivity.

The study was conducted during a two-year (2017–2018) growing seasons (June–October) in the southeast of the Lazio Region (Picinisco, Abruzzo, Lazio and Molise National Park, 1600–1900 m a.s.l.). After appropriate stratification, we built nine grazing exclusion fences (22 m × 12 m), including the Sentinel 2 acquisition grid (2 pixel of 10 m × 10 m). Inside each plot, we cut grass (5 m^2) at heading and regreening stages. The grass was immediately weighed (fresh weight, FW).

In a GIS environment, we calculated the following VIs based on red and NIR spectral bands: SR, NDVI, EVI2, SAVI, MSAVI and OSAVI; a correlation analysis was then conducted to study the relationship between them and fresh ground-collected biomass. In accordance with previous studies, data present an exponential behaviour, with the typical VIs saturation effect in the presence of high biomass values.

All the correlations present a good coefficient of determination (SR $r^2 = 0.575$, NDVI $r^2 = 0.667$, EVI2 $r^2 = 0.673$, SAVI r^2 = MSAVI $r^2 = 0.667$, OSAVI $r^2 = 0.641$). EVI2 presents the best correlation with fresh biomass and was used to derive map, based on Sentinel 2 acquisition data, of pasture productivity at 100 m^2 spatial and 5 days temporal resolution.

The integrated use of satellite spectroradiometer measurements and on-field sampling of biomass and species composition appears to have a great applicability for rangeland management, also with a view to adopt precision grazing techniques.

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Safety assessment: from fruit and vegetable waste to earthworm as feed sources

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The protein supply of farm animals is a central topic in Europe due to the reliance on imported protein, particularly soy products. On the other hand, livestock producers are in demand of sustainable, healthy and safe protein feed. Thus, it is necessary to develop sustainably feed innovations with a high level of safety. At the same time, food waste has already been recognised as an important global issue. From the environmental point of view, food waste has led to unnecessary exploitation of natural resources. Reduction of food waste is a key driver towards sustainable productive solutions and among the possibilities to achieve this goal, in this study is explored the recycling of fruit and vegetable waste (FVW) as growth substrate for producing fresh earthworms then processed into dried meal. The dried meal is assumed adoptable for feed alternative purposes. These topics are included in the philosophy of the circular economy. The safety and security assessment of the derived earthworm meal as future-generation feedstock is the aim of this study.

In order to consider safety aspects of earthworms grown on FVW as future-generation feedstock, this study evaluated the microbiological quality and chemical contaminants of FVW (i) used as growth substrate; fresh earthworms (ii) and earthworms' meal (iii) resulting from drying processes. The sampling and the analysis methods were carried out taking into account the Regulation (EC) No 152/2009 which laid down the methods of sampling and analysis for the official control of feed. The undesirable substances studied were: nitrites, the presence and the level of contamination of mycotoxins, pesticides, heavy metal and microbiological parameters.

Microbiological analyses revealed the absence of *Salmonella* spp. and *Listeria monocytogenes* in FVW, in fresh earthworms and in earthworms' meal. Microbial contamination was below the limit considered for animal feed. Analysis of persistent organic pollutants (POPs) and pesticides were also conducted showing the conformity of earthworms according to feed safety criteria used for food-producing animals. From this result, earthworms' meal can represent a safe future-generation feedstock with improved sustainability in order to reduce the dependence on imported soybean as protein source.

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Honey bee pollen inclusion in diets for meagre (*Argyrosomus regius*) juveniles: effects on growth performance and body indexes

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Previous researches have highlighted positive characteristics of honey bee pollen (HBP), such as its strong antioxidant activity through its constituent flavonoids and the positive effects on non-specific immune responses and growth performances of various animal species. Despite these favourable characteristics no researches have been carried out to evaluate the effects of the inclusion of pollen in diets for marine fish species. Thus, the aim of the trial was to evaluate the inclusion of increasing levels of commercial honey bee pollen in diets for meagre (Argyros*oumus regius*) and its effects on growth performance and body indexes. The experiment was conducted at the IMBRC of the Hellenic Centre for Marine Research (Crete, Greece). At the beginning of the trial, 360 fish were lightly anesthetised, individually weighed (3.35 ± 0.1 g) and randomly divided into 12 fibreglass tanks (3 replicates/diet) supplied by open-circulation borehole aerated sea water.
Four experimental diets were formulated to be isonitrogenous and isoenergetic: HBP0 (without HBP), HBP 1, HBP 2.5 and HBP 4 (with 1, 2.5 and 4% of HBP inclusion level, respectively). At the end of trial (89 days), the following performance indexes were calculated: individual weight gain (WG % IBW), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER). Twelve fish per treatment were killed by over anaesthesia and individually weighed. Fish length was measured to calculate the Fulton's condition factor (K). The fish were dissected to determine the hepatosomatic index (HSI), viscerosomatic index (VSI) and the gut length. All data were processed by ANOVA (SAS). The orthogonal contrast analysis was also performed to test the linear, quadratic and cubic effect among the means. All the growth parameters were significantly influenced by the diet. Feed intake increased as pollen inclusion in the diet increased ($p<.01$), while final weight ($p<.0001$), FCR ($p<.001$), SGR ($p<.01$), PER ($p<.001$) and WG %ABW ($p<.01$) where negatively linearly affected by pollen inclusion in the diet, reaching the worst values in the HBP4 diet. No effect of the diet was registered for VSI and HSI, while gut length and K showed higher and lower values respectively in HBP containing diets compared to control diet ($p=.04$). These preliminary results could be confirmed and find an explanation in the scheduled haemato-biochemical, histopathological and toxicological analyses.

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Characterisation of lactic acid bacteria (LAB) from Provolone del Monaco P.D.O. cheese: preliminary results

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Provolone del Monaco P.D.O. is a pasta filata cheese traditionally produced in Campania region (south of Italy) in the Lattari mountains areas. This type of cheese is produced from a mixture of 20% of milk originated from cattle native genetic type (TGA) Agerolese and from 80% of milk produced by other different breeds of cattle; in particular, it is manufactured with raw milk without the addition of starter cultures. Depending on the environmental and technological factors, all ripened cheese varieties harbour an autochthonous microbiota, which mainly diversifies cheeses throughout processing. The aim of the present work was to identify the lactic acid bacteria (LAB) playing a role in Provolone del Monaco cheesemaking and ripening using conventional microbiological methods followed by Pulse Field Gel Electrophoresis (PFGE) and 16S amplicon sequencing. A total of 371 presumptive LAB were isolated from samples (milk, curds, pasta filata and cheese) collected throughout the technological process and during the ripening. After PFGE analysis 105 different types were distinguished. *Lactobacillus* was the dominant genus in the samples; in particular the species *Lb. casei/paracasei* and *Lb. delbrueckii* were frequently detected. Moreover, *Lb. rhamnosus* was isolated from the beginning of the ripening until the end and *Lb. hilgardii*, *Lb. fermentum* and *Lb. helveticus* were isolated in milk and in aged cheese samples. Besides Lactobacillus genus, also *Streptococcus*, *Lactococcus* and *Enterococcus* genera were reported and among these genera *S. macedonicus*, *S. lutetiensis/infantarium*, *S. thermophiles*, *Lc. lactis*, *E. faecalis* and *E. faecium* were frequently identified. The presence of wild bacterial populations plays a critical role in the development of the unique characteristics of each cheese. The results of the present work show a high microbial diversity and a high species richness in the Provolone del Monaco cheese and thus it is important to preserve this type of P.D.O. cheese made from cow's milk.

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E-nose, e-tongue and e-eye for sensorial evaluation of anchovy fillets marinated in different flavoured olive oil mixture with anisakicidal activity

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Anisakis are responsible for an important fish-borne disease, anisakidosis. This human disease is related to the consumption of raw or undercooked fish, which are parasite hosts of this parasite. In order to reduce the incidence of human anisakidosis all seafood product destined to be consumed raw or almost raw is worldwide recommended the freezing treatments. In several fishery products, such as marinated anchovy, the freezing process, used to destroy *Anisakis* larvae, affects the sensory characteristics. For these reasons, fish marinated industries required new study on alternative process, in order to avoid the freezing. Natural compounds are more frequently used against these larvae and two different flavoured oil mixture commonly used in Italy and in Tunisia for industrial marinated anchovy showed interesting effectiveness against the parasites. Aim of this study was to evaluate the sensorial characteristics of marinated anchovy in different oil mixture with anisakicidal activity compared to traditionally marinated anchovy during storage. For the experiment 18 fresh anchovies (*Engraulis encrasicolus* Linneo, 1758), collected from local markets (Sicily, Italy), were processed within six hours harvesting. Each fish was eviscerated, decapitated and reduced in fillets. Three groups of marinated fillets were evaluated. Control group (CTR) prepared with unflavoured olive oil and two other groups with enriched olive oil in different mixtures against Anisakis larvae: the first one Parsley/Garlic/Lemon used in Italy (MIX) and the second one Cumin used in Tunisia for fish preparations (CUM). Samples were analysed at day 0, 7 and 14 of storage. An E-nose (FOX 4000, Alpha M.O.S.) equipped with 18 MOS sensors, a potentiometric E-tongue (αAstree, Alpha M.O.S.) and an E-eye (Iris Visual Analyser 400, Alpha M.O.S.) were used for asses the trial and to evaluate the difference between the three groups of marinated anchovy. Principal component analyses (PCA) was performed with the native instrumental software (AlphaSoft, v14.1). The three groups were similar overall in colour (DI= –10) and taste (DI= –2) and differed only for the volatile component (DI =62). During storage, the two MIX and CUM groups showed a similar trend to the CTR group, proving to be stable over time. Considering the low influence in sensory evaluation, the enriched oil mixtures can represent an effective method alternative to the freezing process required by European Regulation EC No 853/2004.

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