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## Poster 27

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## Poster 28

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## Session 08

### Molecular tests for milk quality in Romanian sheep and goats

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The casein and  $\beta$ -lactoglobulin polymorphisms are important and well known due to their effects on quantitative traits and technological properties of milk. The aim of this study was to analyze using PCR-RFLP and sequencing techniques the genotype distribution of  $\beta$ -lactoglobulin and  $\alpha$ -s1-casein in Karakul sheep and different goat breeds. DNA amplification was carried out by PCR and the amplicons were digested with restriction endonuclease *RsaI* and *MboII*. Restricted products were analyzed by electrophoresis in agarose gel stained with ethidium bromide. The determined genotypes were confirmed by sequencing. Both in sheep and goat the  $\beta$ -lactoglobulin A allele (AA) yields three bands of 66, 37, and 17 bp, the B allele (BB) gives two fragments of 103 and 17bp, and heterozygote (AB) have all four fragments. The genotypes detected were AB (57%) and AA (43%). In case of  $\alpha$ -s1-casein the different restriction enzyme patterns are: nonA homozygous yield two bands 306 and 66bp, A/A individuals yields three fragments 160, 146 and 66bp and heterozygous nonA/A all four fragments. The most frequent genotype obtained in our breeds was the non-A homozygous genotype. Our results show that the Romanian sheep and goats presents a high degree of variability, which opens interesting prospects for future selection programs, especially marker assisted selection between different genotypes of milk and cheese characteristics and also for preservation strategies.

## Session 08

### Three local cattle breeds from Tuscany (Italy): genetic diversity and similarity

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The aim of this research was to evaluate the genetic diversity of three local cattle breeds reared in Tuscany (Italy) by molecular markers. A total of 149 animals (63 Calvana, CAL; 23 Pontremolese, PON; and 43 Garfagnina, GAR) were genetically characterized by using 22 STR markers. Genetic similarities were calculated by performing all possible pair-wise comparisons between the individual multilocus genotypes. F-statistics, molecular coancestry and inbreeding coefficients and kinship distances were obtained using MolKin v.2.0. A high genetic differentiation between breeds was observed ( $F_{ST} = 0.172$  for CAL/PON, 0.166 for PON/GAR and 0.131 for CAL/GAR;  $P < 0.001$ ). Between-breeds genetic similarity was 0.250 for CAL/PON and PON/GAR and 0.252 for CAL/GAR. Within-breed genetic similarity was 0.378, 0.420 and 0.374 for CAL, PON and GAR respectively. The lower kinship distance ( $D_k$ ) was observed within the Pontremolese breed (0.500 vs 0.530 in CAL and 0.557 in GAR) while the mean coancestry value ( $f_{ij}$ ) and the inbreeding coefficient (F) were higher in PON than in the other breeds (0.278 vs 0.268 in CAL and 0.258 in GAR for  $f_{ij}$ ; 0.667 vs 0.596 in CAL and 0.516 in GAR for F). Molecular data corroborate the evidence, supported by demographic information, that all the three breeds, and particularly the more genetically distant Pontremolese breed, have suffered a severe erosion of the genetic variability.

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## Poster 30



**The Leccese local sheep breed from Apulia (Italy): a questionnaire survey**

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Leccese is a local sheep breed from Apulia, raised mainly for milk production. The population has suffered a dramatic bottleneck during the past three decades due to main changes in rural production systems. Today the breed counts about two thousand animals. In order to establish a successful conservation plan, an economically sustainable valorisation of the breed has to be considered. A questionnaire survey to examine structure and management of farms rearing Leccese sheep breed has been undertaken in the provinces of Lecce, Taranto and Brindisi. A total of 10 farms have been identified, only half of them being registered to the Genealogical Book. Fragmentation and reproductive isolation among farms, lack of labourers to be employed as herdsmen and massive presence of non pure-bred Leccese animals are the main weakness points. On the contrary, main strength points are the eco-sustainable pasture-based production system, higher resistance to local parasites of Leccese compared with non autochthonous milk breeds, widespread know-how in traditional and artisan cheese-making techniques and high quality of both dairy and lamb meat products. All these data suggest the need to adopt instruments, like traditional denominations and farmer consortia, as strategic tools to protect and promote the niche market penetration of both the local single-breed Leccese cheese and the light lamb meat.

**Characterization of MTNR1A gene polymorphism in Sarda breed sheep**

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The melatonin receptor 1a gene (MTNR1A) is polymorphic in many sheep breeds and appears to influence a number of seasonal reproductive responses. The objectives of the study were to obtain the sequence of the MTNR1A gene in Sarda breed sheep and to evidence if polymorphisms are present even in this breed. For the study 220 adult ewes, of an average age of  $3,2 \pm 1,5$  years, coming from several areas of Sardinia, were used. From each animal a blood sample was taken for DNA extraction to utilize for PCR. Amplification product was digested using two restriction enzymes, MnlI and RsaI, for polymorphism identification. Five samples of each genotype were sequenced to confirm the exact position of nucleotide substitution and to verify the presence of other correlated mutations. Furthermore, allele and genotype frequencies and Hardy-Weinberg equilibrium were calculated. Results point out the presence of two polymorphic sites in the Sarda sheep, in positions 605 (C→T) and 612 (G→A). For the mutation in position 605, C allele showed a frequency of 67% while T allele 33%; genotypic distribution resulted: CC 54%, CT 26% and TT 20%. For the mutation in position 612, G allele showed a frequency of 78% vs 22% of the A allele; genotypic distribution: resulted GG 68%, GA 21% and AA 11%. Population was in Hardy-Weinberg equilibrium. Data show the presence in Sarda breed sheep of the same polymorphic sites found in other breeds.

**Genomic selection against canine hip dysplasia in German shepherd dogs using QTL-associated SNPs**

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Canine hip dysplasia (CHD) is one of the most prevalent hereditary skeletal diseases in dogs with multifactorial origin and the involvement of major genes. Different linkage studies had been performed and revealed QTL for German shepherd dogs, Labrador retrievers and Portuguese water dogs. At the genome-wide level of significance, we identified nine QTL in German shepherd dogs and further ten chromosome-wide QTL. In these CHD-QTL, we developed about 180 single nucleotide polymorphisms (SNPs) in 65 different genes located within these CHD-QTLs. More than 100 SNPs were polymorphic in German shepherd dogs. This SNP markerset was completed with further publicly available SNPs in these QTL. We tested these SNPs for association with CHD in a case-control study design. Here, we included a group of 770 German shepherd dogs randomly sampled from the total German shepherd population (> 20,000) and with average coefficients of coancestry as low as possible among all registered German shepherd dogs. The study design was fully matched by sex and CHD-affection status. Only SNPs with significant odds ratios were retained in the final model and for these SNPs an increasing risk for CHD with an increasing number of cumulated CHD allelic effects was evident. Employing these SNPs for estimation of genomic breeding values, we could develop an efficient selection scheme for a rapid reduction of CHD in German shepherd dogs.

**The 'Bracco Italiano' Genetic and Genealogical Study (BIGGS) -**

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The Italian Bracco is one of the oldest pointing dog breed that has been used for hunting since the Renaissance period. Paintings of the 14th century show hunting scenes depicting dogs similar to the present day's Bracco. The breed has officially registered by the ENCI (the Italian Cynological Club) in 1949, when the definitive standard was established. The SABI (Società Amatori Bracco Italiano) is financing a research project aimed at investigating the demographic, genetic and genealogical structure of the breed. In the first phase of the project the complete electronic database of the breed was obtained. The total number of animals on record, born between 1970 and April 2007, was 20,499. The maximum number of traced generations was 14. Here, we show the main demographic parameters of this important breed, which highlights the trend of the inbreeding coefficient during this period. We wish to thank the President and the Board of Directors of the SABI for having actively promoted the present project among the Society's members.