

milk and detected an average yeast count of  $1.9 \times 10^5$  cfu/g and  $4.3 \times 10^4$  cfu/g, respectively. According to these authors a total yeast count of  $5.0 \times 10^5$  cfu/g still indicates good hygienic quality of those products.

Apart from the high counts of aerobic mesophilic bacteria, the most critical results were the numbers of enterobacteria and *E. coli* in several samples. The high contents of up to  $4.7 \times 10^6$  and  $1.7 \times 10^4$  cfu/ml, respectively, found already in the raw buffalo milk, strongly indicate a requirement for improvement of milking hygiene. A similar study by VIANNI *et al.* (9), conducted in Brazil, showed lower average counts of  $2.2 \times 10^5$  cfu enterobacteriaceae/ml raw buffalo milk. In another study (10), conducted in the UK,  $< 10$  *E. coli*/ml were found in five of eight samples of raw buffalo milk and  $< 10^2$  *E. coli*/ml in one of eight samples, in the remaining two samples no *E. coli* were detected. In raw buffalo milk mozzarella CORTESI *et al.* (11) reported up to  $1.8 \times 10^4$  enterobacteria/g. In accordance with our findings, they did not detect any *E. coli*. Neither did CONEDERA *et al.* (12), who examined 501 samples of buffalo milk mozzarella for *E. coli* O157.

Additionally, the high total aerobic bacterial counts in all tested samples of pasteurised milk, which exceeded the legal limits, showed that there is need to improve the process hygiene. For comparison, in a study conducted in the UK (10), the total viable count in unpasteurised buffalo milk was between  $10^3$ – $10^6$  cfu/ml.

Therefore, the producers were educated on improving processing, bottle cleaning and environmental hygiene in particular. Owing to the high counts of enterobacteriaceae and *E. coli* in yoghurt, curd cheese and mozzarella cheese, fundamental changes in process hygiene are also necessary for these products.

This exploratory analysis allows an initial assessment of the microbial quality of these economically valuable, niche-market products. Further samples will be taken to evaluate improvements of the process hygiene. In addition, it will thereby be possible to observe and record seasonal variations.

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## Genetic variability of milk rheological parameters in Italian Friesian dairy cows

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The aim of this study was to estimate the heritability of coagulation parameters ( $r$ , clotting time;  $k_{20}$ , curd firming time;  $a_{30}$  and  $a_{45}$ , curd firmness measured at 30 and 45 min after rennet addition, respectively), somatic cell count (SCC) and titratable acidity. Milk samples were taken from 238 Italian Friesian cows reared in a single herd located in the province of Pisa (northwestern Tuscany, Italy). All individual sample were taken from the morning milking. The heritability coefficient of the clotting time ( $r$ ) was  $0.26 (\pm 0.142)$ , while for the other technological parameters the coefficient was rather low ( $0.08 \pm 0.162$  for  $k_{20}$ ,  $0.09 \pm 0.195$  for  $a_{30}$  and  $0.12 \pm 0.204$  for  $a_{45}$ ). Also the heritability coefficient of the SCC was low ( $h^2 = 0.07 \pm 0.122$ ) but not different from values reported elsewhere, as for titratable acidity ( $h^2 = 0.03 \pm 0.201$ ) too.

### Genetische Variabilität von rheologischen Parametern der Milch bei italienischen Friesian-Kühen

Ziel dieser Studie war es, die Heritabilität der Koagulationsparameter (Gerinnungszeit  $r$ ; Bruchfestigungszeit  $k_{20}$ ; Bruchfestigkeit  $a_{30}$  bzw.  $a_{45}$  gemessen 30 und 45 min nach Zusetzen des Labs), der somatischen Zellzahl (SCC) und des

titrierbaren Säuregrads festzustellen. Milchproben wurden von 238 italienischen Friesian-Kühen aus einer einzelnen Herde in der Provinz Pisa, nordwestliche Toscana, Italien, genommen. Alle Einzelproben wurden den Morgengemelken entnommen. Der Heritabilitätskoeffizient der Gerinnungszeit  $r$  lag bei 0,26 ( $\pm 0,142$ ), während die Koeffizienten für die anderen technologischen Parameter recht niedrig waren ( $0,08 \pm 0,162$  für  $k_{20}$ ,  $0,09 \pm 0,195$  für  $a_{30}$  und  $0,12 \pm 0,204$  für  $a_{45}$ ). Auch der Heritabilitätskoeffizient des SCC war niedrig ( $h^2 = 0,07 \pm 0,122$ ) und unterschied sich nicht von den Werten anderer Autoren. Gleiches gilt für den titrierbaren Säuregrad ( $h^2 = 0,03 \pm 0,201$ ).

**50 Cheese milk** (rheological parameters, genetic variability)

**50 Käseemilch** (rheologische Parameter, genetische Variabilität)

## 1. Introduction

The limits imposed by milk quotas on quantity production as well as the European Community regulations concerning marketing and prices based on quality, stimulate producers to seek higher qualitative standards for milk destined for commercial purposes. Therefore, great importance must be given not only to percentages of fat and protein, but also to all other quality parameters, such as somatic cell count (13), and when transformation is involved, technological parameters (milk coagulation time, curd-firming time and curd firmness). It's well known that milk that coagulates and form a firm curd soon after addition of the clotting enzyme produces higher cheese yields than milk that coagulates slowly. Moreover, the somatic cell count, lactose, pH and titratable acidity of the milk, are all good indicators for diagnosing mastitis in lactating cows (1).

The choice of product direction (i.e., milk for fresh consumption) can frequently lead to neglecting milk quality in parameters which are fundamental to other product directions.

Furthermore, the study of the genetic variability fraction emphasizes the importance of environmental sources and their control. Also of fundamental importance is determining the genetic variability of the somatic cell count: the possibility of selection against somatic cells may lower the incidence of mastitis and lead to considerable economic benefits resulting from an increase in milk quality (18).

In this study we have attempted to evaluate the genetic variability of the technological parameters and of the SCC in milk from a single herd of Italian Friesian cattle raised in the Province of Pisa (Tuscany), with the aim of devising possible strategies for improvement.

## 2. Material and methods

The study was carried out on 238 Italian Friesian cows reared in the same herd in a medium-temperate climatic zone in the province of Pisa (northwestern Tuscany, Italy). All animals were fed the same diet: the cows were allowed to range freely, and were fed a unifeed diet; about 16 kg/head of the ration was distributed three times a day. The ration consisted of corn mash, polyphyte meadow silage, alfalfa hay first and second cut, straw, mixed feed (crude protein 21%), corn, barley, soybeans, cotton seeds, Na bicarbonate, zeolite and water.

The individual milk samples were collected from the morning milking from each cow and analyzed in triplicate. No preservatives were added.

The following procedures were carried out on every sample of fresh milk:

- SCC (Fossomatic 360), titratable acidity;
- rheological parameters were measured: rennet clot-

ting time ( $r$ =min), rate of curd firming ( $k_{20}$ =min), and curd firmness 30 and 45 min after rennet addition ( $a_{30}$  and  $a_{45}$  = mm) (Formagraph, Foss Electric).

For Somatic Cell Count a logarithmic transformation of data was effected to normalize the variance.

For the evaluation of the heritability the following mixed linear sire model was performed using JMP, ver. 5.0 for PC, of the SAS Institute (7):

$$Y_{ijk} = \mu + s_i + O_j + bX_{ijk} + \varepsilon_{ijk}$$

where  $Y_{ijk}$  = considered parameters;  $\mu$  = overall mean;  $s_i$  = random effect of the  $i^{\text{th}}$  sire ( $i=1, \dots, 6$ );  $O_j$  = fixed effect of the  $j^{\text{th}}$  parity ( $j = 1, \dots, 4$ );  $b$  = regression coefficient on the time elapsed since calving in days ( $X_{ijk}$ );  $\varepsilon_{ijk}$  = residual error.

All estimates of variance components for estimates of heritability were obtained with a REML procedure using a Newton-Raphson method (5, 16, 17). Because a sire model was used, estimates of ratios of sire to total variance were multiplied by 4 to yield heritability estimates.

Standard errors of heritabilities were approximated according to BECKER (3).

## 3. Results and conclusions

**Table 1: Means values of a milking**

Trait		Means	s.d.
CCS*; Somatic Cell Count	CCS/ml	12.75	1.444
pH; pH		6.70	0.164
r; Clotting time	min	27'33"	10'36"
$k_{20}$ ; Curd firming time	min	9'03"	7'21"
$a_{30}$ ; Curd firmness (1)	mm	11.55	10.692
$a_{45}$ ; Curd firmness (2)	mm	17.72	12.448
AT; Titratable acidity	"SH/50 ml	3.10	0.449

\*A logarithmic transformation of data was effected. (1) Curd firmness measured 30 min, (2) 45 min after rennet addition.

**Table 2: Estimates of variance components and heritabilities for coagulation parameters**

Trait	Additive genetic variance	Phenotypic variance	$h^2$	SE
Log CCS	6.07	86.71	0.07	0.122
pH	0.07	7.00	0.01	0.235
AT	0.06	2.00	0.03	0.201
$r$	19.74	75.92	0.26	0.242
$k_{20}$	2.00	25.00	0.08	0.205
$a_{30}$	9.77	108.55	0.09	0.261
$a_{45}$	14.97	124.75	0.12	0.208

As shown in Table 1, milk resulted of scarce reactivity. The heritability of clotting time " $r$ " with the value of 0.26 (Table 2) is in line with previously described values (6, 8, 9, 10, 15, 19) in cattle, while the heritability coefficients of curd firming time and curd firmness are rather low (0.08 for  $k_{20}$ , 0.09 for  $a_{30}$  and 0.12 for  $a_{45}$ ).

On the contrary other authors, in studies carried out on buffalo (14), and on Massese sheep (3, 4), report low heritability coefficients for all coagulation parameters.

Studies that have estimated the heritability coefficient of the SCC (11, 12, 13) report values ranging between 0.11 and 0.23, somewhat higher than those determined in this study ( $h^2=0.07$ ). At the sight of the heritability value it is not expected to obtain an important genetic response to the selection of SCC in dairy cows belonging to the Italian Friesian breed. So, for the time being, a reduction in SCC should particularly be based on improving hygiene and environmental conditions on farms.

The heritability values reported in this study are somewhat low for all parameters considered, the majority being close to zero, with the exception of clotting time, with a value of 0.26. The existence of a strong extra-genetic variability, probably due to the aims of the company (i.e., production of fresh milk) as well as to sample size, suggests and confirm the importance of further study in order to verify not only the accuracy of heritability values within the parameters themselves, but also to determine possible plans for environmental improvement.

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## Chemical studies of Pecorino Siciliano cheese throughout ripening

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Changes in the composition and level of proteolysis in the internal portions of Pecorino Siciliano cheese were studied. At the end of ripening, the average moisture, fat and protein concentrations were 33.36, 30.9 and 27.73%, respectively, for winter-made cheese and 35.63, 30.2 and 27.56%, respectively, for spring-made cheese. The average level of salt-in-moisture (g 100 g<sup>-1</sup>) was 11.55 and 6.16 in winter- and spring-made cheese, respectively. The average pH of both winter- and spring-made cheese was 5.36. Urea-polyacrylamide gel electrophoresis of the cheese samples indicated that  $\alpha_{s1}$ -casein was degraded in all samples leading to the formation of products with higher electrophoretic mobility.  $\beta$ -Casein was also degraded to equivalent levels in all cheeses at the end of ripening. Peptide profiles obtained by reversed phase high performance liquid chromatography indicated significant changes during ripening. The principal free amino acids (FAAs) found in all samples were Glu, Leu, Phe, Lys and Val.

### Chemische Untersuchungen von Pecorino Siciliano-Käse während der Reifung

Es wurden Veränderungen in Zusammensetzungen und Proteolyse-Level bei inneren Teilen von Pecorino-Siciliano untersucht. Am Ende der Reifung lagen durchschnittlicher Wassergehalt sowie Fett- und Proteinkonzentration bei 33,36 und 30,9 bzw. 27,73% für im Winter hergestellten Käse und bei 35,63 und 30,2 bzw. 27,5% für Frühlingskäse. Der durchschnittliche Salz-in-Wasser-Level (g 100 g<sup>-1</sup>) war 11,55 in Winter- bzw. 6,16 in Frühlingskäse, der durchschnittliche pH-Wert betrug bei beiden Käsen 5,36. Die Harnstoff-Polyacrylamid-Gelelektrophorese der Käseproben zeigte, dass  $\alpha_{s1}$ -Casein in allen Proben abgebaut wurde und zu Produkten mit höherer elektrophoretischer Mobilität führte. Auch  $\beta$ -Casein wurde bei allen Käse am Ende der Reifung gleichmäßig reduziert. Ebenso zeigten durch Umkehrphasen-Hochleistungs-Flüssigkeitschromatographie (RP-HPLC) erhaltene Peptidprofile während der Reifung signifikante Veränderungen. Die wesentlichsten freien Aminosäuren (FAAs) waren in allen Proben Glu, Leu, Phe, Lys und Val.