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## Genetic parameters for somatic cell score, differential somatic cell count, and milk electrical conductivity in dairy buffaloes

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### ABSTRACT

Understanding the genetic architecture of economically important traits in dairy buffaloes is essential for implementing genetic selection and breeding strategies aimed at improving the productivity and profitability of dairy buffalo farming. Our objective was to estimate genetic parameters of udder health indicators, that is, electrical conductivity (EC), total somatic cell score (SCS), and differential somatic cell count (DSCC), in Mediterranean buffalo populations. Data were collected from six dairy commercial herds located in the Basilicata Region (Italy) including 14,571 records from 1501 Mediterranean buffaloes. Covariance components and genetic parameters were estimated using a multi-trait animal model by a Bayesian inference approach. The animal model accounted for the systematic effects of herd, days in milk (DIM), parity order, year-season of calving, year-season of sampling, milk yield class, and temperature – humidity index (THI) as well as the random effects of additive genetic and permanent environmental effects. The heritability estimates (95% credibility interval) for EC, SCS and DSCC were 0.18 (0.12–0.23), 0.11 (0.05–0.17) and 0.085 (0.04–0.12), respectively. The genetic correlations were all positive and ranged from 0.045 (−0.26 to 0.32) (DSCC-EC) and 0.76 (0.55–0.92) (DSCC-SCS). Our findings highlighted that udder health indicators have additive genetic variation which is potentially exploitable for breeding purposes. However, the response to genetic selection will likely be slow, so selection should be practiced jointly with improvements in animal husbandry and environmental conditions.

### HIGHLIGHTS

- Heritability and genetic correlations for indirect indicators of mastitis were estimated in Mediterranean buffaloes.
- Genetic parameters for udder health indicators offer crucial insights to design breeding programs for milk yield and quality in dairy buffaloes.
- DSCC analysis holds promise as a novel tool for the early identification and monitoring of mastitis in dairy animals.

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## Introduction

Udder health indicators have not been traditionally considered as breeding objectives in dairy buffalo. Although Italian Mediterranean buffaloes appear less prone to udder infections compared to other dairy species (Fagiolo and Lai 2007), mastitis remains a widespread issue in the dairy industry (Ruegg 2017). Milk traits such as electrical conductivity (EC) and total somatic cell count (SCC) are widely recognised as

indirect indicators of mastitis (Heringstad et al. 2006; Matera et al. 2022). Recently, differential somatic cell count (DSCC), that is, the proportion of neutrophils and lymphocytes on the total SCC (Damm et al. 2017), has been suggested as a novel promising phenotype to prevent and manage mastitis. Collectively, these indicators provide a more comprehensive understanding of the health and physiological status of the mammary gland, which can inform more effective strategies for disease prevention, identification,

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management and genetic improvement within dairy herds. In dairy cattle, somatic cell score (SCS; log-transformation of SCC) and DS<sub>CC</sub> are heritable traits (Bobbo et al. 2019; Pegolo et al. 2021; Ablondi et al. 2023) and SCS in particular has been widely adopted as selection criteria in breeding programs for mastitis resistance (Jamrozik 2013; Wiggans and Carrillo 2022). In dairy buffaloes, although several studies have already estimated the genetic parameters for milk production and quality (Aspilcueta-Borquis et al. 2010; Cesaraní et al. 2021; Silva et al. 2023) udder health indicators have not been considered yet as selection criteria in breeding programs (Lázaro et al. 2024). So far, the knowledge about DS<sub>CC</sub> in other species other than cattle (e.g. buffalo, sheep, and goat) is limited, possibly due to the absence of a rigorous methodological validation in such species. Nevertheless, few studies have already been published including DS<sub>CC</sub> both in dairy buffaloes (Bobbo et al. 2023; 2024) and in dairy sheep (Tolone et al. 2023). Therefore, the aim of this study was to investigate genetic variation, heritability and genetic correlation of EC, SCS, and DS<sub>CC</sub> in Mediterranean buffalo populations in order to provide new knowledge for enhancing breeding programs focused on improving udder health and milk quality.

## Materials and methods

All data used in this study were obtained from pre-existing databases based on routine milk recording procedures and did not involve any invasive procedure, so approval of the animal welfare committee was not necessary. Monthly test-day records from August 2019 to April 2022 were provided by the Italian Breeders Association, collected from six dairy commercial herds located in the Basilicata Region (Italy). Cows were milked twice daily and individual samples were taken at consecutive am and pm milkings. All milk samples were analysed using CombiFoss 7 DC (Foss, Hillerød, Denmark) to determine milk composition (data not shown), EC (mS/cm), SCC (cells/mL), and DS<sub>CC</sub> (%). Outliers beyond four standard deviations were treated as missing values and removed. Additionally, records with SCC and DS<sub>CC</sub> equal to zero were also excluded. The SCC was log-transformed to SCS [ $SCS = \log_2 (SCC/100,000) + 3$ ] according to Ali and Shook (1980) to achieve a normal distribution, while DS<sub>CC</sub> did not require any transformation. The final dataset comprised 14,571 records from 1501 animals, including only animals with at least three test-day records within lactation and with less than

360 days in milk (DIM). The records contained information about herd, animals, sampling date, and daily milk production. In order to calculate Temperature–Humidity Index (THI) according to Bobbo et al. (2024) the daily average temperature at 2 m (T, °C) and relative humidity at 2 m (RH, %) were obtained from public stations from NASA Prediction of Worldwide Energy Resource – POWER (Sparks 2018) using the geographic coordinates (latitude and longitude) of the six herds. Descriptive statistics of EC, DS<sub>CC</sub>, SCS and THI have been reported in Table 1. Pedigree information was provided by the National Association of Italian Buffalo Breeders (ANASB), and included buffaloes with all their ancestors for a total of 4004 animals from 235 sires and 2631 dams. The covariance components and genetic parameters were estimated by Bayesian approach and Markov-chain Monte Carlo (MCMC) methods (Sorensen and Gianola 2002) in multi-trait analyses using GIBBSF90 and POSTGIBBSF90 programs (Misztal et al. 2022). For all traits (EC – mS/cm, SCS – unit, and DS<sub>CC</sub> – %) the animal model accounted the systematic effects of herd (six classes, A to F), DIM (10 classes of 30 days, with the last being an open class until 360 days), parity order (1, 2, 3, 4, 5 and ≥ 6), year-season of calving (11 classes), year-season of sampling (nine classes), milk production (four classes based on quartiles of average milk production by herd), and THI (four classes based on quartiles) selected previously by Bobbo et al. (2024). Additive genetic and permanent environmental effects were included as random effects. All traits were considered as continuous variables, and their values were assumed to be sampled from the multivariate normal distribution. Analysis consisted of a single chain of 250,000 cycles, a burn-in of 50,000 and a thinning interval of 100 cycles, totalling 2000 samples used to compute the marginal posterior distributions. The convergence was visually determined by plotting Gibbs samples, as well as using statistical tests of Heidelberger and Welch (1983) and using the coda package (Plummer et al. 2006) with the R program (R code Team R: A language and environment for statistical computing, 2014). For all analyses, convergence was successfully achieved.

**Table 1.** Descriptive statistics of electrical conductivity (EC), differential somatic cell count (DS<sub>CC</sub>), somatic cell score (SCS), and temperature – humidity index (THI).

	EC (mS/cm)	DS <sub>CC</sub> (%)	SCS, units	THI, units
N of records	14,413	14,571	14,569	14,571
Mean	592.75	54.45	3.92	57.03
Standard deviation	68.69	17.39	1.81	11.09
Minimum	327.70	5.00	-2.06	38.00
Maximum	942.10	97.00	10.73	77.00

**Table 2.** Estimates of variance components, heritability, and genetic correlations for electrical conductivity (EC), differential somatic cell count (DSCC) and somatic cell score (SCS)

	Heritability			Repeatability			
	$\hat{\sigma}_a^2$	$\hat{\sigma}_{pe}^2$	$\hat{\sigma}_e^2$	Mean	HPD95	Mean	HPD95
EC	635.3	453.4	2447.8	0.179	0.12–0.23	0.307	0.28–0.32
DSCC	20.19	43.26	172.95	0.085	0.04–0.12	0.268	0.24–0.28
SCS	0.243	0.488	1.45	0.111	0.05–0.17	0.335	0.31–0.35

<sup>1</sup> Estimates are expressed as mean of the marginal posterior density of the parameters; HPD95 = lower and upper bound of the 95% highest posterior density region,  $\hat{\sigma}_a^2$  = additive genetic variance,  $\hat{\sigma}_{pe}^2$  permanent environmental variance,  $\hat{\sigma}_e^2$  residual variance.

## Results and discussion

The estimates of the variance due to permanent environmental effects were 453.4, 43.26, and 0.488 for EC, DSCC and SCS, respectively (Table 2). Heritability estimates are presented in Table 2, but the comparison with the literature is challenging, as to the best of our knowledge there is no information about the genetic aspects of EC and DSCC in buffaloes. The heritability estimate for EC observed in our study was moderate (0.18), whereas in dairy cattle it is often variable. Norberg et al. (2004) found heritability estimates around 0.12 at the beginning of lactation in a multi-breed dairy herd, while Samaraweera et al. (2022) reported estimates ranging from 0.11 to 0.22 in dairy cows in temperate areas. In contrast, Norberg et al. (2004) found higher heritability estimates between 0.26 and 0.36 using a repeatability model in Holstein cows. These findings suggest potential genetic differences in EC across dairy breeds, species, and populations. Nonetheless, the moderate heritability observed in this research indicates that EC may be a viable selection criterion for improving dairy buffalo performance. The heritability estimate obtained for SCS (0.11) was medium-to-low but in accordance with the literature. Indeed, Aspilcueta-Borquis et al. (2010) reported for Murrah buffaloes SCS heritability of 0.255 (0.13–0.36) using a Bayesian approach, ranging from 0 to 0.43 across lactation. Our estimate for DSCC heritability in dairy buffaloes (0.085) was in line with that observed in dairy cattle, as reported by Bobbo et al. (2019) and Pegolo et al. (2021) in Holstein cows (0.08 and 0.11, respectively) and by Ablondi et al. (2023) in Simmental cows (0.08). The genetic and environmental correlations between all traits are presented in Table 3. The genetic correlation estimates were all positive, ranging from 0.045 (DSCC-EC) and 0.76 (DSCC-SCS); an intermediate value (0.33) was observed between EC and SCS. Considering the genetic correlation between DSCC and SCS in Italian Holsteins, (Bobbo et al. 2019) reported an estimate of 0.66, whereas Pegolo et al. (2021) a value of 0.60. Ablondi et al. (2023) found

**Table 3.** Genetic (upper triangle) and environmental (lower triangle) correlations<sup>1</sup> among electrical conductivity (EC), differential somatic cell count (DSCC) and somatic cell score (SCS).

	EC		DSCC		SCS	
	Mean	HPD95	Mean	HPD95	Mean	HPD95
EC	–	–	0.045	–0.26 to 0.32	0.330	0.05–0.59
DSCC	0.293	0.10–0.53	–	–	0.760	0.55–0.92
SCS	0.605	0.41–0.77	0.786	0.70–0.86	–	–

<sup>1</sup> Estimates are expressed as mean of the marginal posterior density of the parameters HPD95 = lower and upper bound of the 95% highest posterior density region.

instead a genetic correlation of 0.61 between DSCC and SCS in Simmental cows. Pegolo et al. (2021) also reported genetic correlations of 0.26 between DSCC and milk conductivity, and of 0.31 between SCS and milk conductivity. Interestingly, the genetic correlation between EC and SCS is always positive because both traits are influenced by genes related to immune response and mammary permeability. Generally, during mastitis there is an increase in SCC, but also an increase in ion leakage ( $\text{Na}^+$ ,  $\text{Cl}^-$ ), resulting in raised EC. The genetic correlation between EC and DSCC can be also negative (see confidence intervals reported in Table 3) because DSCC represents the proportion of the different cell types (neutrophils and lymphocytes) that compose SCC, not just the total count. Therefore, in acute mastitis, neutrophils dominate, increasing both SCC and DSCC. However, in chronic infections, macrophages may dominate, leading to high EC, high SCC, but low DSCC. This might explain why EC and SCS are positively genetically correlated, whereas EC and DSCC may show a negative or weak genetic correlation.

## Conclusions

In conclusion, our findings suggest that the genetic parameters for somatic cell score, differential somatic cell count, and electrical conductivity offer critical insights for enhancing breeding programs aimed at improving milk yield and quality in dairy buffaloes. Moreover, DSCC analysis emerges as a promising tool for the early identification and monitoring of mastitis in dairy animals. However, rigorous validation of this methodology across a variety of livestock species, beyond the well-studied cattle population is essential to ensure its wider applicability.

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This manuscript reflects only the authors' views and opinions; neither the European Union nor the European Commission can be considered responsible for them.

## Ethical approval

Animal Care and Use Committee approval was not obtained for this study because the data were obtained from an existing database. The authors did not have direct control over the care of the animals included in this study.

## Disclosure statement

No potential conflict of interest was reported by the authors.

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## Data availability statement

The results and analyses presented in this article are freely available upon request

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