

**COW AND HERD PREDICTORS OF UDDER HEALTH STATUS IN A RANDOM
SAMPLE OF ARGENTINIAN DAIRY HERDS**

**PREDICTORES A NIVEL DE VACA Y DE RODEO DEL ESTATUS DE SALUD MAMARIA EN UNA
MUESTRA ALEATORIA DE TAMBOS DE ARGENTINA**

Vissio.; Claudina^{1,2}; Richardet, Melina²; Dieser, Silvana Andrea²; Odierno, Liliana Mónica³;
Larriestra, Alejandro José¹

¹ Departamento de Patología Animal, Facultad de Agronomía y Veterinaria, Universidad Nacional de Río Cuarto, Córdoba, Argentina. Ruta 36 km 601, C.P. X5804ZAB, Río Cuarto, Córdoba, Argentina. ² Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina. ³ Departamento de Microbiología e Inmunología, Facultad de Ciencias Exactas, Físico-Química y Naturales, Universidad Nacional de Río Cuarto, Córdoba, Argentina

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Correspondencia e-mail: Claudina Vissio cvissio@ayv.unrc.edu.ar

RESUMEN

El objetivo del presente estudio fue evaluar la relación entre la salud mamaria, utilizando recuento celular somático de leche compuesta, y factores a nivel de vaca y rodeo en tambos de Córdoba, Argentina. La población de estudio fueron rodeos con 100 a 250 vacas en ordeño. En 48 tambos seleccionados aleatoriamente y visitados durante el año 2007, se inspeccionó la rutina de ordeño, se entrevistó al tambero y se colectaron en promedio 45 muestras compuestas de leche de vacas seleccionadas aleatoriamente, de estas se registró fecha y número de parto. El recuento celular somático fue determinado con equipo Somacount 300 y su valor transformado a logaritmo natural. Se ajustó un modelo de regresión lineal generalizado mixto con el rodeo como efecto aleatorio. La media de recuento celular somático en las 2.140 vacas seleccionadas fue 5,38 (Desvío estándar =1,56). Terapia al secado, despunte y edad del tambero, como factores de rodeo, días en lactancia y número de partos, como factores a nivel de vaca, fueron asociados al recuento celular somático. El efecto incremental para los factores a nivel de vaca fue mayor que para los factores de rodeo. La información provista podría contribuir a diseñar programas para mejorar la salud mamaria en Argentina.

Palabras clave: (recuento celular somático), (factores de riesgo), (modelo de regresión mixto).

SUMMARY

The objective of this observational study was to evaluate the relationship between cow- and herd-level factors and cow udder health status, measured by composite milk sample somatic cell count (CSCC) in dairy herds from Córdoba, Argentina. The study population was dairy farms of a size of 100 to 250 cows; 48 randomly selected dairy herds were visited during 2007. Each visit involved a milking routine inspection, an interview to the dairyman and the collection of 45 milk composite samples from cows selected by a random systematic procedure. Parity and days in milk were recorded for each cow sampled. The somatic cell count was determined with a Somacount 300, and then a natural logarithmic transformation of SCC values was used. A general linear mixed regression model looking at herd as a random effect was fitted. The mean of somatic cell count in 2,140 cows was 5.38 (SD=1.56). Dry cow therapy, milk strip and dairymen age as herd factors, lactation days and parity as cow factors, were associated with somatic cell count. The incremental effect on somatic cell count for cow attributes was greater than herd factors. This information will help to device herd programs to improve udder health in Argentina.

Keywords: composite somatic cell count, risk factors, mixed regression model.

INTRODUCTION

The dairy sector in Argentina is undergoing a process of deep transformations, showing an increase in the herd size and the adoption of herd management practices that are reshaping the

traditional pasture systems. This new context raises concerns about the udder health status. Bovine mastitis is the most costly disease for dairy herds worldwide. Recently, Vissio *et al.* (2015) reported average losses of 1 U\$\$/cow/day for dairy herds from Argentina.

Factors that influence the magnitude and variation of bovine mastitis in the herd can be defined at both herd and individual level, this implies a study properly designed and the fit of multilevel models. So far, no survey has yet reported in Argentina on mastitis prevalence considering herd and cow risk factors based on a random sample of medium and small size dairy herds. Such producer stratum represents around 60% of dairy farms in Argentina (SENASA, 2015). The primary objective of this research was to assess the relationship between cow- and herd-level factors and cow udder health status, measured by composite milk sample somatic cell count (CSCC) in dairy herds from Córdoba, Argentina. The secondary objective was to account the herd-level clustering effect for CSCC, relative to cow to cow variation.

MATERIALS AND METHODS

Study population

The target population was dairy farms located in the province of Córdoba (Argentina) with a herd size of 100 to 250 dairy cows (medium and small size). The study population was a sample of 48 dairies randomly selected from a roster provided by the producer's association; the sampling fraction was 14.2%. Overall, a total of 2,140 cows were sampled. From March to September of 2007, each farm was visited once and all milking cows were subjected to systematic random selection. The sample size was determined by assuming an expected subclinical mastitis prevalence of 20%, a precision of 10% and confidence level of 95%. The systematic sampling involved that the first cow was selected randomly followed by selection of cows at equal intervals. The selection interval was calculated dividing the total milking cows by the sample size.

Data collection

At the herd visits, the following steps were carried out: (1) Visual assessment of milking routine and parlour characteristics using a checklist, (2) Administration of an interview to the dairyman, and (3) Composite milk sample collection together with records on calving date and parity for all cows selected.

The questionnaire was designed by the first author and revised by four mastitis experts. Once the final version was available, the questionnaire was tested and fine-tuned on 5 dairy herds. All interviews were performed by the same person (first author). The questionnaire covered information on herd structure, level technology, parlor characteristics, mastitis control, milking routine, and socio-demographic dairymen characteristics.

Somatic cell count (SCC) was determined in each composite milk sample using Somacount 300 (Bentley, USA 1997).

Data analysis

Composite SCC (CSCC) was used as the response variable. To approximate the normal distribution, a natural logarithmic transformation of SCC values (lnCSCC) was used. The

predictors at herd level were gathered from checklist and questionnaire; parity (cow or heifer) and days in milk (DIM) were the variables at cow level evaluated.

Correlation and mean comparison analysis were used to assess associations between herd- and cow-factor, and lnCSCC. Only those factors with a p-value <0.20 were considered for further multivariate analysis. The relationship between lnCSCC and factors was determined using general linear mixed model (Snijders.; Bosker, 1999). The model was specified as $Y_{ij}=\alpha+\beta*Z_{ij}+v_j+\epsilon_{ij}$, where subscripts i and j denote the ith cow and the jth herd; Y_{ij} = lnCSCC from the ith cow (n=1928) for jth herd (n=48); α =regression intercept; β =coefficients associated with each covariate; Z =covariate vector for fixed effects; v_j =random effect reflecting clustering of cows within herds; and ϵ_{ij} =random effect to reflect residual variation for cow.

The proportion of variance attributed to herd was deduced using the Intra-class correlation coefficient (ICC) (Snijders.; Bosker, 1999). The value of ICC was deduced as, $ICC=\sigma_t^2/(\sigma_t^2+\sigma_r^2)$, where σ_t^2 =herd variance components, and σ_r^2 =residual variance components.

The proportion of variance explained by the predictors included in the final model was calculated as the ratio between the variance of the full model and null model (with no predictors) (Larriestra *et al.* 2005).

The analyses were performed using Proc Mixed (SAS Institute, Cary, North Carolina, USA).

RESULTS

An average of 45 (DS=3.1) cows within each herd were selected systematically at random. The mean of lnCSCC in 2,140 cows was 5.38 (SD=1.56). The lnCSCC average in cows before 90 DIM was 4.92 (SD=1.79) and in cows with 90 or more DIM was 5.63 (SD=1.41). The lnCSCC average for heifers and cows (>1 calving) were 4.81 (SD=1.66) and 5.58 (SD=1.51), respectively. The overall average of these herd means was 5.38 (range=3.88-6.49).

The explanatory variables offered to multivariate analysis after screening process were: cows in milk, milk level production, milk production records, artificial insemination, interval between milking machine test, treatment of clinical mastitis, microbiology analysis of milk sample from clinical mastitis, diagnosis of subclinical mastitis, drying-off management, dry cow therapy, udder washing before milking, teat pre-dipping, milk stripping, teat post-dipping, educational level and age of dairymen, use of gloves during milking.

The final model for CSCC retained dry cow therapy, milk strip and dairymen age as herd factors and DIM and parity as cow factors (Table 1).

The null model ICC was 0.13 ($\rho=\sigma_t^2/(\sigma_t^2+\sigma_r^2)=0.3293/(0.3293+2.1248)=0.13$), which indicates that 13% of the cow to cow variation in CSCC would be explained by the herd clustering. When predictors were added to the model ICC was 0.12 ($\rho=\sigma_t^2/(\sigma_t^2+\sigma_r^2)=0.2657/(0.2657+1.9461)$). Therefore the variance of the CSCC explained by the model was 10% ($1-(2.212/2.454)$).

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| Variable | Fixed effect | | | Least squares mean | | CSCC (cells/mL) |
|-------------------------------|------------------|------------|---------|--------------------|-----------|--------------------|
| | Estimate (SE) | 95% CI | p value | Estimate (SE) | 95% CI | Estimate |
| Intercept | 4.85 (0.23) | 4.39-5.30 | <0.01 | | | |
| Days in milk | | | | | | |
| >90 | 0.68 (0.07) | 0.55-0.82 | <0.01 | 5.60 (0.12) | 5.35-5.85 | 271375 |
| ≤90 | | | | 4.92 (0.13) | 4.66-5.18 | 136838 |
| Parity | | | | | | |
| ≥2 | 0.74 (0.08) | 0.59-0.90 | <0.01 | 5.63 (0.12) | 5.39-5.88 | 279667 |
| 1 | | | | 4.89 (0.13) | 4.62-5.16 | 132781 |
| Litres/cow/day | | | | | | |
| <15 | -0.53 (0.31) | -1.16-0.09 | 0.13 | 5.00 (0.27) | 4.46-5.54 | 148265 |
| 15-17 | -0.28 (0.18) | -0.63-0.08 | 0.09 | 5.25 (0.16) | 4.94-5.57 | 191062 |
| >17 | | | 0.12 | 5.53 (0.15) | 5.22-5.84 | 252598 |
| Dry cow therapy | | | | | | |
| No | 0.43 (0.21) | 0.00-0.85 | 0.05 | 5.47 (0.18) | 5.11-5.84 | 238579 |
| Yes | | | | 5.05 (0.14) | 4.77-5.33 | 155633 |
| Milk stripping | | | | | | |
| No | 0.38 (0.19) | 0.00-0.76 | 0.05 | 5.45 (0.17) | 5.10-5.80 | 232572 |
| Yes | | | | 5.07 (0.13) | 4.80-5.34 | 159653 |
| Age of dairymen (years) | | | | | | |
| <34 | -0.72 (0.26) | -0.20-1.24 | 0.01 | 4.97 (0.19) | 4.60-5.35 | 144705 |
| 34-50 | -0.58 (0.22) | -0.13-1.03 | 0.01 | 5.11 (0.13) | 4.85-5.38 | 166384 |
| >50 | | | | 5.69 (0.21) | 5.27-6.12 | 297198 |

Table 1. Final general linear mixed model for factors associated with composite somatic cells counts (CSCC) with herd as random effect

DISCUSSION

The SCC is probably the most widely test used to detect IMI, CSCC is a cost-effective option to reveal and describe the udder health status (Vissio *et al.* 2014). Using such sampling procedure and within the context of a probabilistic design, would be feasible to infer the udder health status variation among farmers of one of the most important Argentina's dairy sector. The herds studied showed big differences in their CSCC distribution, suggesting certain level of intra-herd clustering of SM, which resulted in only a minor fraction of variation in the CSCC (ICC=13%). The clustering effect at herd level for SM was assessed previously (De Vliegher *et al.* 2004; Nyman *et al.* 2009), which reported ICC values between 4% and 8%. The different study population may explain the disparities observed in the degree of

clustering. Nyman *et al.* (2009) and De Vliegher *et al.* (2004) included only freshened heifers in their studies, thus the ICC may only be the manifestation of underline effect of factors prior to calving. By contrast, in the present study, cows and heifers during whole lactation were recruited, so a more comprehensive set of management factors may have been involved, fact that may be responsible for the higher ICC found.

In Argentina, the udder health status is still being challenged by the high prevalence of *Staphylococcus aureus* (Dieser *et al.* 2014). That situation may reflect some variation in the adoption of practices well known for their efficacy and included in the so-called “five point plan” conducted at the National Institute for Research in Dairying in the 1960’s. In this context, Vissio *et al.* (2013) found farms in the region that differed markedly regarding the degree of adoption of a comprehensive udder health management program. Dry cow therapy and milk stripping, interventions included in the “five point plan”, were associated with CSCC in this study. The treatment of all udder quarters has been reported as effective to reduce the infection prevalence at dry off and the incidence of new infections during the dry period and after calving (Dufour *et al.* 2011). All herds that applied dry cow therapy, showed a significant reduction of CSCC at cow level (lowered in 83,000 cells/mL).

The milk strip, considered a secondary prevention strategy for early detection of clinical mastitis, resulted associated with CSCC (lowered 73,000 cells/mL) in this study. Dufour *et al.* (2011), conducting a standardized review of udder health management, did not found any association between this practice and SCC. Furthermore, fore-stripping has been found to be a risk factor for clinical mastitis (Schukken *et al.* 1991). It is certainly difficult to explain the discrepancy between studies. The contextual effect of the udder health management in each farm may change or mask the effect of individual practices in the population. In addition, contagious mastitis pathogen transmission could be favored for stripping fore-milk of cows with clinical mastitis (Schukken *et al.* 1991) or the association could be a result of detection bias (Peeler *et al.* 2000).

The postmilking disinfection is an intervention of well proven efficacy to reduce new infections. Dufour *et al.* (2011) found that post-dipping was consistently associated with low weighted average herd SCC. Hence, our result was not expected. In this study, around 40% (19/48) of the farmers applied post-dipping, however, many of them were not able to determine the degree of consistency and for how long this practice was used.

During milking, many decisions are implemented by farm workers and could be influenced by their level of instruction, motivation and according to working environment (Jansen *et al.* 2009). This study involved different aspects, such as dairymen age, level of instruction, specialized qualification and the time of the dairyman in the herd. However, only age of dairyman remained as associated with lnCSCC, showing lower CSCC in cows milked by younger dairyman. That association seems reasonable, however it may represent the effect of several underline factors associated with age, some of them not recorded in this study.

The variability from cow to cow was the largest source of variation, so that, known and unknown cow factors would be responsible for the higher CSCC within the herd. The cow factors most consistently associated with high CSCC, in accordance with the present findings, are cow parity and DIM (Reneau, 1986; Harmon, 1994). In general, older cows are more likely to be exposed to pathogens, including more infected quarters and more extensive tissue damage in long standing infections (Reneau, 1986). In the same way, the rate of infection

would increase prior to drying-off. In addition to that, at the end of lactation the cows produced less milk and a rise in SCC, this can be interpreted as a milk concentration (Reneau, 1986; Harmon, 1994).

The main research findings documented here applied only to small and medium dairy farms from Argentina, because the probabilistic design had that producer segment as target population. Further studies have to be done in order to explore the mastitis health status among large dairy herds.

CONCLUSION

Cow parity and days in milk, as cow factors, and milk strip, dry cow therapy and dairymen age, as herd factors, were all associated with cow CSCC. The CSCC variation rested mainly on differences from cow to cow, while the herd clustering accounted for 13% of the total variance.

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