



Acta Scientiarum. Animal Sciences

ISSN: 1807-8672

Editora da Universidade Estadual de Maringá - EDUEM

Scolari Botton, Fernanda; Moro Alessio, Dileta Regina; Busanello, Marcos; Corrêa Schneider, Catia Leticia; Hammes Stroeher, Fernanda; Pereira Haygert-Velho, Ione Maria

Relationship of total bacterial and somatic cell counts with
milk production and composition - multivariate analysis

Acta Scientiarum. Animal Sciences, vol. 41, e42568, 2019

Editora da Universidade Estadual de Maringá - EDUEM

DOI: <https://doi.org/10.4025/actascianimsci.v41i1.42568>

Available in: <https://www.redalyc.org/articulo.oa?id=303160553039>

- How to cite
- Complete issue
- More information about this article
- Journal's webpage in redalyc.org

EDUEM
redalyc.org

Scientific Information System Redalyc

Network of Scientific Journals from Latin America and the Caribbean, Spain and
Portugal

Project academic non-profit, developed under the open access initiative



Relationship of total bacterial and somatic cell counts with milk production and composition – multivariate analysis

Fernanda Scolari Botton¹, Dileta Regina Moro Alessio², Marcos Busanello³, Catia Letícia Corrêa Schneider¹, Fernanda Hammes Stroeher¹ and Ione Maria Pereira Haygert-Velho^{1*} 

¹Universidade Federal de Santa Maria, Campus de Palmeira das Missões, Avenida Independência, 3751, 98300-000, Palmeira das Missões, Rio Grande do Sul, Brazil. ²Universidade do Estado de Santa Catarina, Centro de Ciências Agroveterinárias, Lages, Santa Catarina, Brazil. ³Escola Superior de Agricultura “Luiz de Queiroz”, Universidade de São Paulo, Piracicaba, São Paulo, Brazil. *Author for correspondence. E-mail: ione.h.velho@ufsm.br

ABSTRACT. This study was realized to evaluate the monthly production, composition and quality of milk (total and defatted dry extract, lactose, fat and protein) and their relation to somatic cell count (SCC) and total bacterial count (TBC) using multivariate statistical analyses. The data are from a dairy farm for the period of two years (from January 2015 to December 2016). The SCC and TBC variables were transformed to somatic cell score (SCS) and \log_{10} (LogTBC). Factor analysis, discriminant analysis and cluster analysis were used. Through factor analysis, it was found two factors that together explained 69.5% of the total data variation. The first factor represented the inverse relationship between lactose versus fat and protein content, while the second factor represented the inverse relationship among monthly milk yield *versus* SCS and LogTBC. The discriminant analysis identified that lactose and protein contents and SCS were the variables that had the greatest participation in the separation of the groups formed by the cluster analysis. The groups differed mainly by the monthly production of milk, composition and SCS. Finally, there are important multivariate relations between the variables milk production, composition and quality.

Keywords: cluster analysis; factor analysis; *Bos taurus*; fat; lactose; protein.

Received on April 24, 2018.

Accepted on May 28, 2018.

Introduction

The milk composition enables to monitor the feeding of the dairy herd, aiming to correct failures that might compromise the production of lactose, fat and protein (Dias et al., 2017; Fagan, Jobim, Calixto Júnior, Silva, & Santos, 2010). In addition, it also enables the monitoring of total bacterial counts (TBC) and somatic cell counts (SCC), which are indicators that can be used to evaluate the hygienic management during milking and the health of the mammary gland of lactating cows (Busanello et al., 2017a; Paixão et al., 2017). Therefore, the hygiene practices adopted during milking, besides contributing with the maintenance of the microbiological quality of the milk, also collaborates in the health of the mammary gland. In this way, the discharge of the first three jets at the beginning of milking, which is directly associated with the SCC and TBC, helps in the prevention and identification of the occurrence of mastitis (Belage, Dufour, Bauman, Jones-Bitton, & Kelton, 2017; Eckstein et al., 2016).

Other factors directly related to the animals, such as number of lactation, stage of lactation, and body condition, affects milk yield and quality, especially when using cows with a high number of lactations and advanced lactation stage, as they tend to have a higher incidence of problems that affect the health of the mammary gland (Dias et al., 2017). In addition, the level of education of producers also influence the dairy production, as the more knowledge they have about their own business, the more quality and production practices are used (Eckstein et al., 2016). Considering the factors related to milk production and composition, which include the physiological status of lactating cows, feeding, hygiene conditions at the time of milking and the health of the mammary gland, the use of multivariate analysis is appropriate. The reason for that is because both factorial and cluster analysis allow to evaluate better the interactions between the various factors involved in milk production and composition, when compared to conventional (univariate) analysis that can measure the effect of only one or two factors singly (Macciotta, Cecchinato, Mele, & Bittante, 2012).

The cluster analysis is characterized as a set of procedures that aims to group and discriminate groups of variables, based on distances, which are mathematical measures of similarity. In the formation of groups, the closest observations are in the same group, while the more distant ones separate the group (Lebart, Morineau, & Piron, 2000). Therefore, the clustering analysis allows the better characterization and interpretation of the formed groups, and together with the factorial analysis, it allows the comparison within and between the production units that use the system (Tremblay et al., 2016). Therefore, this study aimed to evaluate the monthly production, composition and quality of milk considering the variables of the total dry extract (TDE) and defatted dry extract (DDE), lactose, fat, protein and its relation with SCC and TBC using techniques of multivariate statistical analysis of data from a smallholder dairy farm in a two-year period (from January 2015 to December 2016).

Material and methods

The database was provided by the owners of Olho d'Água dairy farm, located in the county of Jaboticaba, in the northwest of the state of Rio Grande do Sul, Brazil. The climate of the region is classified as Cfa type, sub-tropical, with hot summers and well distributed rains throughout the year, with average temperature of 22°C in the hottest period (January, February and March) (Peel, Finlayson, & McMahon, 2007). The temperature and rainfall data are showed in the Table 1.

The dairy farm had 21 hectares, of which 15.5 were intended for milk production and the remainder was occupied by improvements, facilities, vegetation areas and dams. The dairy herd was composed of 25 purebred lactating dairy cows of the Jersey and Holstein breeds, with different ages, number of lactations and lactation stages. The average production of the cows was 14 liters/cow/day, and both the milk production and the genetic composition of the herd varied during the studied months. The milk production varied accordingly to the management of the dairy farm, because there was no reproductive planning, so, in some months there was a higher concentration of lactating animals and, consequently, higher monthly milk production. Regarding the genetic group, there was a variation in the proportion of cows of each breed, and in all months the percentage of Jersey cows was higher.

The cows were reared in a semi-confined system, and the cows had free access to the feed building. The feed was provided in the trough, and it was composed of concentrated millet silage, which, sometimes, was replaced by haylage made from oat (*Avena strigosa*) and ryegrass (*Lolium multiflorum*). The cows had access to the pasture that was used intermittently, respecting the vegetative cycle of the pasture, and they had free access to potable water, in both the improvements and paddocks. In the summer, the pasture consisted of signal grass (*Brachiaria decumbens*), Áries grass (*Panicum maximum*), Tifton-85 (*Cynodon spp.*) and sorghum (*Sorghum bicolor*). In winter, it was used black oat (*Avena strigosa*), white oat (*Avena sativa*), dual-purpose wheat BRS Tarumã (*Triticum aestivum*) and annual ryegrass (*Lolium multiflorum*). Pasture fertilization was carried out with chemical fertilization (N-P-K), organic fertilization and urea. The concentrate (composed by corn, soybean meal, wheat bran, minerals and vitamins) was offered according to the milk production of each cow, where in the first 100 days of lactation, one kilogram of concentrate was provided for every three liters of milk produced. From 100 days to six months of production, each cow received one kilogram of concentrate for every four liters of milk produced, and from the 6th month of production, each cow received two kilograms of concentrate a day.

The drying-off of the cows was realized two months before calving, and the animals were placed on a paddock with perennial Tifton-85 pasture (*Cynodon spp.*), near the management center (milking room and feed building). Approximately 20 days before calving, the cows received a concentrate for the gradual adaptation to the feeding management, which was applied during lactation.

The milking parlor was made of wood, with concrete floor, and had a milking system of a four-joint fishbone type, piped until the cooling bulk tank. The milking was performed at 6 a.m. and 4 p.m., and the cows remained in the waiting room until the milking time. The order of milking was performed according to the sanity of the mammary gland, by first milking the healthy cows, then the cows that were once treated in their productive life, and finally the cows that were being treated, of which the milk was discarded. The milking procedure was initiated by cleaning the teats with pre-dipping (0.3% iodine) and drying with disposable paper towels. In all milking, the first milk jets were collected in a strip cup test to monitor possible clinical mastitis. Monthly, the California Mastitis Test (CMT) was performed to detect possible cases of subclinical mastitis. The teacup was maintained for a period of five to seven minutes, and in the end milking the post-dipping (iodine at 0.7-1.0%) was applied. After milking, the cows were taken to the feed building, where they were fed at the trough with corn silage or haylage of oat and ryegrass with concentrated. With that process, the cows stood for the necessary time to close the teat sphincter.

Table 1. Average temperature and total rainfall for Palmeira das Missões – RS, which is the city with the nearest meteorological station from the evaluated dairy farm, between the years of 2015 and 2016.

Month	Mean temperature (°C)	Total rainfall (mm)
January	22.5	92.8
February	22.3	147.8
March ¹	20.4	125.2
April ¹	20.6	134.6
May ¹	15.7	71.4
June ^{*1}	13.6	-
July ¹	13.7	100.8
August ¹	17.6	193.4
September ¹	15.9	48.4
October ¹	18.3	276.6
November	20.2	108.3
December	21.7	232.0

Source: Instituto Nacional de Meteorologia (INMET, 2017), with data referring to the meteorological station of Palmeira das Missões city, which is the nearest unit from the property; ¹The meteorological station had problems in some months with the equipment that measures precipitation, and the months indicated did not present data for the year 2015 (the presented value of the month refers to the year 2016); ^{*}in June 2016 the meteorological station presented a new problem, not allowing the precipitation measurement for that month.

The milking equipment was sanitized immediately after milking with water at a temperature of 40°C. Then, a solution of chlorinated alkaline detergent (pH greater than 11) was used, with a water inlet temperature of 70° to 75°C with a circulation time of 10 minutes. Afterwards, an acid detergent (pH less than 3) was used at a temperature between 30-35°C for 5 minutes.

The vaccines used in the herd were: Brucellosis (*Brucella sp.*) (Brucelina B19 vaccine from Vallee®), that was applied by a qualified veterinarian, only for females from three to eight months; Leptospirosis (Leptovac 6®) at a dose of 5 mL, in which the application took place every three months; Carbuncle (*Bacillus sp.*) (Sintoxan® Polyvalent from Merial), applying 2 mL subcutaneous per animal, once a year; Infectious Bovine Rhinotracheitis (IBR) and Bovine Viral Diarrhea (BVD), using the CattleMaster GOLD FP®5+L5 from Zoetis, at the dosage of 5 mL per cow, subcutaneously, with recommendation of application every six months. Finally, the vaccine for Foot-and-Mouth Disease (Bovitel, from Vallee®), applied 5 mL subcutaneously, according to the schedule of the Secretariat of Agriculture, Livestock and Irrigation of the State of Rio Grande do Sul. The worm treatment was carried out every four months on calves and heifers. In diagnosed cases of cows with clinical mastitis, the supportive therapy was based on antimicrobials and anti-inflammatories applied by intramuscular route for three days. This protocol was indicated by the veterinarian, and the cow's milk was discarded, respecting the grace period of each medicinal product that was applied. In milder cases, with the beginning of signs of inflammation such as redness, heat and edema, the therapy consisted only on anti-inflammatories and diuretics. In the drying process (60 days before delivery) an intramammary antibiotic was used as a preventive treatment.

The data about monthly production, composition and quality of milk were collected during the period from January 2015 to December 2016. The milk samples from the cooling bulk tank were collected by the professionals of the dairy company that bought the milk, with three collections per month. The milk samples were sent to the Laboratório de Serviços Leiteiros (SARLE) of the University of Passo Fundo (UPF). The laboratory was certified by the Brazilian Ministry of Agriculture, Livestock and Supply (MAPA) to determine the levels of TDE, DDE, lactose, fat, protein using the near infrared technique (NIRS, Bentley 2000, Bentley Instruments, USA), in accordance with the ISO 9622, in addition to determine SCC and TBC by flow cytometry (Somacount 300, Bentley Instruments, USA), in conformation with the ISO 13366-2. INMETRO IEC 17025: 2002 prescribed both methods, since the dairy company paid for milk quality, including bonus.

The descriptive statistics of the data set evaluated are presented in Table 2, as well as the transformations for the SCC and TBC, that adopted the following methodologies: (somatic cell score - SCE = $(\log_2(\text{SCC}/100) + 3)$ (Ali & Shook, 1980) and logarithm with base 10 (\log_{10}) (LogTBC), respectively. Subsequently, the data were evaluated using multivariate analysis techniques (factor analysis and clustering), using SAS® statistical software (Statistical Analysis System [SAS], 2000).

The factor analysis allows the evaluation of the relationship between the variables, while the cluster analysis aims to confirm these relationships and quantify the differences for each variable among the formed groups. The factor analysis was used to evaluate the relationship between the variables, addressing the reduction

of the original set of variables to a smaller number of factors, considering that the relation of the variables that compose it represents each factor. The factor analysis was performed using the PROC FACTOR, and Kaiser-Meyer-Olkin (KMO) statistic was used to verify the suitability of the model. The variables were selected to compose each of the factors by their communalities, which represent the proportion of the variation of the original variables that are explained by the factors. The number of factors was defined by the eigenvalue equal to one, and all eigenvalues greater than one were considered. The factor loadings were considered significant from 0.40 and the Promax rotation was used, which is an oblique rotation that considers the relation between the factors.

The clustering analysis was performed using PROC FASTCLUS to form groups of observations that presented similar characteristics within the group, and differences with the other groups. In order to achieve this, we used the Ward's hierarchical method based on the Euclidean distance to estimate the means of the groups. Then, the discriminant analysis was performed through PROC DISCRIM, in order to correctly classify the observations within each group, and the PROC STEPDISC, using the STEPWISE method aiming to identify the variables responsible for group differentiation. The canonical discriminant analysis (PROC CANDISC) was used to graphically demonstrate the distances within and between groups. For these analyzes, the assumptions of multivariate normality and homogeneity of the covariance matrix were tested and met. In this way, the observations were correctly distributed within their respective groups when verified by the Fischer linear discriminant function, using the algorithm of the k-nearest neighbor (KNN).

Finally, we performed the test of comparison of means for the groups formed by the cluster analysis, where the normality (Shapiro-Wilk test) and homogeneity of variances assumptions (Levene's test) were tested. Only the variables monthly milk production and lactose content did not meet these assumptions. The variables TDE, DDE, fat content, protein, SCS and LogTBC met the assumptions of normality and homogeneity of variances, and for them, the analysis of variance (ANOVA) was performed using PROC GLM. For the variables milk production and content of lactose, which did not meet the assumptions, PROC GLIMMIX was used, which considers the distribution of the response variable in the statistical model. The normal distribution was used for both variables. In addition, we transformed the seasons of the year into numerical variables in Likert scale, being: 1 = Summer - January, February and March; 2 = Autumn - April, May and June; 3 = Winter - July, August and September; and 4 = Spring - October, November and December. Thus, the variable season of the year was compared between the groups using the Kruskal-Wallis non-parametric test (PROC NPAR1WAY), because as a Likert scale variable (ordinal), it cannot be compared by parametric statistical analysis (Norman, 2010). The variable season of the year was not included in the multivariate analyses. When the results were significant, the means were compared by the Tukey test for PROC GLM, Tukey-Kramer test for PROC GLIMMIX, and Dwass, Steel, Critchlow-Fligner method (DSCF) for PROC NPAR1WAY. The statistical significance was considered at the 5% probability level for all analyses.

Results and discussion

In the factorial analysis of the monthly production, composition, SCC and TBC of the milk samples taken from the bulk tank in the evaluated smallholder dairy farm, 69.5% of the total variance was explained by the first two factors, with a KMO of 52.2 (Table 3). The first factor comprises the negative relation of the lactose content with the fat and protein content, which is demonstrated by the opposition of its high factorial loadings, as for example for the lactose level (-0.819) and fat level (0.956). Alessio et al. (2016) evaluated the milk composition of 73 herds of the Catarinense Cattle Breeders Association, using multivariate analysis, and they verified that milk production had a negative relation with milk solids, such as fat and protein. However, in our study, the milk production is represented indirectly by lactose content, because this component is the main determinant of milk production (Allen & Piantoni, 2014). Both results demonstrate the importance of the rural producers of the South of Brazil to carry out genetic improvement programs that allow the increase of the milk production as well as the production of total solids, since the greater total dry extract production maximizes the profitability of the industry, which, by its turn, can pay better by means of bonuses.

Table 2. Descriptive statistics on the production system of the Olho d'Água farm in the years 2015 and 2016.

Variables	Descriptive statistics				
	Minimum	Average	Median	Maximum	SD
Monthly milk production (L Month ⁻¹)	4.164	9.022	7.510	16.997	3.866
Total dry extract (%)	12.14	13.06	13.10	13.67	0.36
Defatted dry extract (%)	8.57	8.98	9.00	9.27	0.14
Lactose content (%)	4.18	4.45	4.46	4.57	0.10
Fat content (%)	3.46	4.08	4.11	4.54	0.26
Protein content (%)	3.16	3.50	3.48	3.87	0.16
SCC (cells mL ⁻¹)	114.330	249.790	208.000	834.000	154.880
SCS	3.19	4.13	4.06	6.06	0.72
TBC (CFU mL ⁻¹)	12.670	69.650	40.830	249.000	58.760
LogTBC	1.10	1.70	1.61	2.40	0.37

SCC = Somatic cell count; SCS = Somatic cell score; TBC = Total bacteria count transformed into log₁₀.

The second factor represents the positive relationship between SCS and LogTBC, which have a negative effect on monthly milk production, as well as on profitability when payment programs based on milk quality are adopted (Busanello et al., 2017a). Busanello, Rossi, Cassoli, Pantoja, and Machado (2017b) studied the prevalence and incidence of subclinical mastitis in 517 Brazilian herds. They found high values for these indicators (46.4% and 0.17 new cases/cow at risk/month, respectively). In addition, these values did not improve over their study period, highlighting the need to establish large-scale milk quality programs in Brazil. Therefore, the high commonalities for the variables in our study demonstrate the importance of these variables in the monitoring and evaluating the milk quality and herd's health.

According to Dillon, Hennessy, and Cullinan (2015), the SCS can influence milk production. In a study conducted in Ireland from 2008 to 2011, these authors found that approximately 30% of the herds had a mean SCS above of 300,000 cells mL⁻¹. In addition, when comparing herds with 400,000 cells mL⁻¹ with the ones with 300,000 cells mL⁻¹, a 2% reduction in productivity was verified. Although not statistically significant, it considerably reduced the annual profitability of milk producers. Afterwards, we applied the discriminant analysis. This analysis selected the variables that were determinant in the differentiation of the groups formed by the cluster analysis (Table 4). These variables were protein content, lactose content and SCS. All the determinant variables were considered, and the partial R² explains how much each variable represents in the differentiation of the groups. Thus, 66.3% of the distance between the groups is explained by the milk protein content, 41.5% by the lactose content and 34.1% by SCS.

Three groups of observations were formed from the cluster analysis (Table 5), and they were evidenced by the canonical discriminant analysis that graphically demonstrated the Euclidean distances used in the separation between and within the groups (Figure 1). Groups 1 and 2 differed for TDE, DDE, fat and protein content, whereas groups 2 and 3 differed for monthly milk production, lactose, fat and protein content, and SCC (Table 5). Groups 1 and 3 differed for milk production and milk composition, except for SCC. The variables for season and LogTBC did not differ for the groups formed. However, it is noteworthy that the group 1 was a reference to warmer months, whereas 2 and 3 referred to colder months.

Table 3. Factorial loadings, eigenvalue, percentage of variance and communality of each variable referring to the factorial analysis of the production system at the Olho d'Água farm for the years of 2015 and 2016.

Variables	Factors*		Communalities
	1	2	
Monthly milk production (L Month ⁻¹)	-0.137	-0.720	0.603
Lactose content (%)	-0.819	-0.119	0.749
Fat content (%)	0.956	-0.168	0.835
Protein content (%)	0.936	0.088	0.938
SCS	-0.058	0.882	0.301
LogTBC	-0.042	0.562	0.746
%Variance	48.6	20.9	

*Factors formed by factorial analysis; SCS = score of somatic cell count; TBC = total bacterial count transformed into log₁₀.

Table 4. Discriminant analysis of the determinant variables on monthly milk production in the production system at the Olho d'Água farm for the years 2015 and 2016.

Variables	Parcial R ²	F	P > F	Wilks Lambda	P < Lambda	ASCC	P > ASCC
TP (%)	0,663	20.73	<0.0001	0.336	< 0.0001	0.331	< 0.0001
TL (%)	0.415	7.10	=0.0047	0.196	< 0.0001	0.539	< 0.0001
SCS	0.341	4.92	=0.0190	0.129	< 0.0001	0.635	< 0.0001

ASCC: Average Squared Canonical Correlation; SCS= somatic cell score

Table 5. Groupings formed by the variables regarding to the production system at the Olho d'Água farm for the years 2015 and 2016.

Variables	Groups*			P
	1	2	3	
Season of the year ¹	1.00	3.00	3.00	=0.9135 ¹
Monthly milk production (L Month ⁻¹)	9.986 a	10.304 a	5.439 b	=0.0238
Total dry extract (%)	12.57 b	13.12 a	13.37 a	<0.0001
Defatted dry extract (%) (%)	8.80 b	9.03 a	9.01 a	=0.0013
Lactose content (%)	4.48 a	4.49 a	4.32 b	=0.0002
Fat content (%)	3.77 c	4.09 b	4.35 a	<0.0001
Protein content (%)	3.32 c	3.48 b	3.70 a	<0.0001
SCS	4.39 ab	3.73 b	4.79 a	=0.0033
LogTBC	1.51	1.69	1.88	=0.2709
Number of observations (Month)	5	13	6	

*Groups formed by cluster analysis; SCS = somatic cell score; LogTBC = total bacterial count transformed to log10; ¹Season of the year was measured on a Likert scale and compared by Kruskal-Wallis nonparametric method, and the values presented represent the median of the observations, being 1 - summer, 2 - autumn, 3 - winter and 4 - spring.

The increase in SCS and LogTBC presented a negative relation with milk production (factor 2, Table 3), and the increase in SCS caused a reduction in lactose content and milk production (group 3, Table 5). Ribas et al. (2014) found similar results, in which they verified a reduction in lactose contents with an increase in the bulk tank somatic cell count (BTSCC) when working with samples from bulk tanks in regions from the Paraná state. This demonstrated that the lactose content decreased from 4.63 to 4.14% with increasing the TSCC range from 0 to 17,000 cells mL⁻¹ to 2,263,000 to 4,525,000 cells mL⁻¹ in samples collected in 1,950,034 bulk tanks.

The SCC is used in several countries as a health indicator of the mammary gland and for genetic improvement of the herd (Haile-Mariam & Pryce, 2017). High concentrations of SCC in milk are an indicative of inflammation of the mammary gland of lactating cows. This occurrence causes variation in milk composition and quality due to changes in the total available solids and to the presence of pathogens that cause diseases in cows, which raises both prevalence and incidence of mastitis (Busanello et al., 2017b). Another factor that contributes to explain this indirect relationship of the lactose content with fat and protein content, which is not usually found, might be related to the feed that was given to the herd at the Olho d'Água farm in the years of 2015 and 2016. The lactose content is highly dependent of energy and related to high milk production, and due to the substitution of corn silage for oat and ryegrass haylages at the end of 2016, there might have been a reduction in the energy concentration of the diet, compromising the glucose availability for the synthesis of milk lactose. Lactose is formed from glucose (Lemosquet, Delamaire, Lapiere, Blum, & Peyraud, 2009), which is mainly generated from the propionate in the fermentation of non-fibrous carbohydrates at the rumen, that is metabolized into glucose in the liver. Therefore, the glucose is used in the synthesis of lactose, which is the main determinant of milk production (Allen & Piantoni, 2014; National Research Council [NRC], 2001). Considering the relationship between lactose content and milk production, this may indicate the indirect relationship of milk production with fat and protein contents. This relation is caused by the effect of dilution or increase in the concentration of fat and protein contents as a function of production, since the concentration of fat and milk protein decreases as the milk production increases (Alessio et al., 2016; Haile-Mariam & Pryce, 2017; Ribas et al., 2014).

In the group 3 (Table 5) the low milk production with high levels of fat and protein, with repercussion in TDE and DDE, can be explained by the higher SCS. In cows with elevated SCC, there is an increase in fat due to the reduction of milk production, so the fat is concentrated and has its content risen. The same occurs with the protein content (Table 3), which has a positive relationship with SCC and occurs due to tissue injuries caused by intramammary infection. This leads to a change in membrane permeability of the gland, increasing the influx of albumin and immunoglobulins into the gland, and, by its turn, raises the protein concentration in milk (Ribas et al., 2014). In this group, the relationship between milk production and lactose production was also evident, being that the lactose content is closely related to milk production (Haile-Mariam & Pryce, 2017).

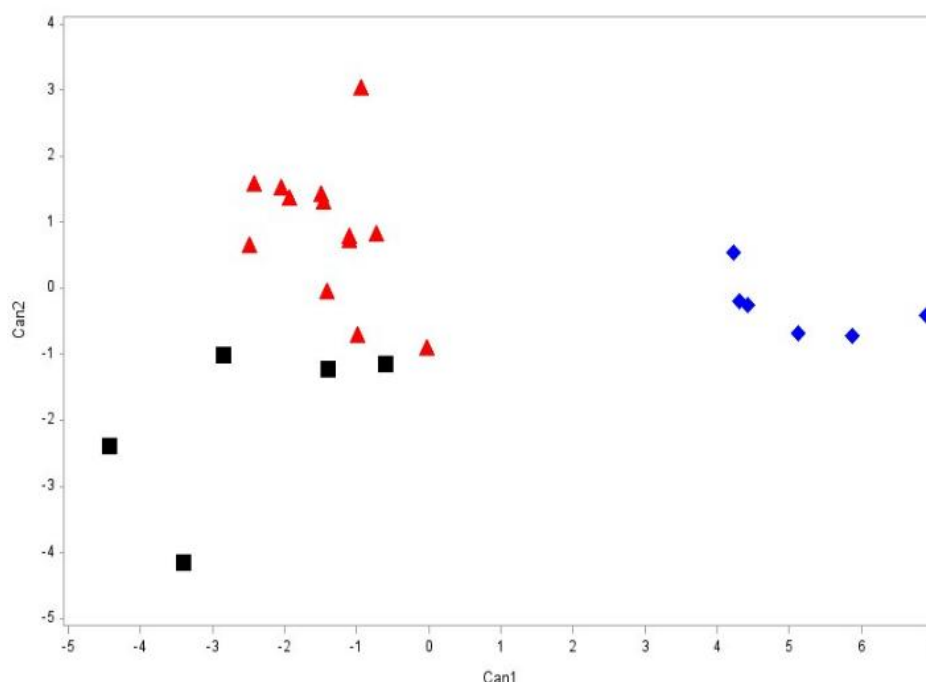


Figure 1. Canonical discriminant analysis showing the Euclidean distances between and within the groups formed by the variables for the observations (months) which represent the group 1 (■), group 2 (▲) and group 3 (◆) in relation to the milk production at the Olho d'Água farm for the years 2015 and 2016.

The low monthly milk production (group 3, Table 5) can also be explained by the reduction in the percentage of lactating cows in some months of the year, caused by the lack of reproductive planning. This affects the oscillation of the number of lactating cows, as well as the stage of lactation, being that a higher proportion of cows could be at the end of lactation. This explanation also justifies the increase in the content of milk solids, which might have been influenced by the proportion of lactating Jersey cows. It should be highlighted that these factors, such as the number of lactating cows, lactation stage and proportion of cows of a particular breed are commonly found in the routine of smallholder dairy farms in the Northwest Region of Rio Grande do Sul. In the milking management, another key procedure is the pre-dipping, which minimizes the presence of microorganisms in raw milk and the occurrence of pathogenic microorganisms in the teat canal, resulting in lower rates of TBC and SCC (Reche et al., 2015). Regarding to mastitis prevention, the use of post-dipping is essential, and 74% of the dairy farms from the Northeast Region of the State of Rio Grande do Sul use this milking management (Machado, Fischer, Stumpf, & Stivanin, 2017).

In Ireland and Netherlands, in order to achieve reduction of SCC and improvement of milk quality, the farms carried out training, extension services, discussion groups between farmers and dairy companies, and monitoring of individual cows and the quality of milk from the bulk tank. They improved the herd's health, which provides a better milk yield for the dairy industry. The dairy industry becomes more competitive by producing higher quality and for being responsible by the consumer's satisfaction of the demand (Dillon et al., 2015; Passetti et al., 2016). These recommendations have a fundamental importance for Brazil, concerning the improvement of the quality and productivity of milk.

Conclusion

In a multivariate way, two factors were found to be important to explain the relation between milk production, composition and quality. The first factor indicates the inverse relationship between lactose versus protein and fat, whereas the second indicates the inverse relationship between SCC and TBC versus total milk production of the herd. From these factors, three observations groups were formed, which are mainly differentiated by milk production and composition, in which lactose, protein and SCC were the determining factors for the separation of these groups. Finally, there is a strong multivariate relationship between the variables related to milk production, composition and quality.

Acknowledgements

The authors thank the owners of the Sítio Olho d'Água farm for assigning the data, which allowed the development of this project. This study has financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001.

References

- Alessio, D. R. M., Thaler Neto, A., Velho, J. P., Pereira, I. B., Miquelutti, D. J., Knob, D. A., & Silva, C. G. (2016). Multivariate analysis of lactose content in milk of Holstein and Jersey cows. *Semina: Ciências Agrárias*, 37(4), 2641-2652. doi: 10.5433/1679-0359.2016v37n4Supl1p2641.
- Ali, A. K. A., & Shook, G. E. (1980). An optimum transformation for somatic cell concentration in milk. *Journal of Dairy Science*, 63(3), 487-490. doi: 10.3168/jds.s0022-0302(80)82959-6.
- Allen, M. S., & Piantoni, P. (2014). Carbohydrate nutrition: Managing energy intake and partitioning through lactation. *Veterinary Clinics: Food Animal Practice*, 30(3), 577-597. doi: 10.1016/j.cvfa.2014.07.004.
- Belage, E., Dufour, S., Bauman, C., Jones-Bitton, A., & Kelton, D. F. (2017). The Canadian National Dairy Study 2015—Adoption of milking practices in Canadian dairy herds. *Journal of Dairy Science*, 100(5), 3839-3849. doi: 10.3168/jds.2016-12187.
- Busanello, M., Freitas, L. N., Winckler, J. P. P., Farias, H. P., Dias, C. T. S., Cassoli, L. D., & Machado, P. F. (2017a). Month-wise variation and prediction of bulk tank somatic cell count in Brazilian dairy herds and its impact on payment based on milk quality. *Irish Veterinary Journal*, 70(26), 1-13. doi:10.1186/s13620-017-0103-z.
- Busanello, M., Rossi, R. S., Cassoli, L. D., Pantoja, J. C. F., & Machado, P. F. (2017b). Estimation of prevalence and incidence of subclinical mastitis in a large population of Brazilian dairy herds. *Journal of Dairy Science*, 100(8), 6545-6553. doi: 10.3168/jds.2016-12042.
- Dias, M. B. d. C., Leão, K. M., Carmo, R. M., Silva, M. A. P., Nicolau, E. S., & Marques, T. C. (2017). Milk composition and blood metabolic profile from holstein cows at different calving orders and lactation stages. *Acta Scientiarum. Animal Sciences*, 39(3), 315-321. doi: 10.4025/actascianimsci.v39i3.34807.
- Dillon, E. J., Hennessy, T., & Cullinan, J. (2015). Measuring the economic impact of improved control of sub-clinical mastitis in Irish dairy herds. *The Journal of Agricultural Science*, 153(4), 666-675. doi: 10.1017/S0021859614001178.
- Eckstein, I. I., Pozza, M. S. S., Ramos, C. E. C., Zambom, M. A., Santos, G. T., Kazama, R., & Pozza, P. C. (2016). Typification of factors related to milk production and its impact on the sanitary quality of milk. *Scientia Agraria Paranaensis*, 15(1), 56-63. doi: 10.18188/1983-1471/sap.v15n1p56-63.
- Fagan, E. P., Jobim, C. C., Calixto Júnior, M., Silva, M. S., & Santos, G. T. (2010). Environmental and handling factors on the chemical composition of milk in dairy farms of Paraná State, Brazil. *Revista Brasileira de Zootecnia*, 32(3), 309-316. doi: 10.4025/actascianimsci.v32i3.8570.
- Haile-Mariam, M., & Pryce, J. E. (2017). Genetic parameters for lactose and its correlation with other milk production traits and fitness traits in pasture-based production systems. *Journal of Dairy Science*, 100(5), 3754-3766. doi: 10.3168/jds.2016-11952.
- Instituto Nacional de Meteorologia [INMET]. (2017). *Rede de Estações Climatológicas*.
- Lebart, L., Morineau, A., & Piron, M. (2000). *Statistique exploratoire multidimensionnelle*. Paris, France: Dunod.
- Lemosquet, S., Delamaire, E., Lapierre, H., Blum, J. W., & Peyraud, J. L. (2009). Effects of glucose, propionic acid, and nonessential amino acids on glucose metabolism and milk yield in Holstein dairy cows. *Journal of Dairy Science*, 92(7), 3244-3257. doi: 10.3168/jds.2008-1610.
- Macciotta, N. P. P., Cecchinato, A., Mele, M., & Bittante, G. (2012). Use of multivariate factor analysis to define new indicator variables for milk composition and coagulation properties in Brown Swiss cows. *Journal of Dairy Science*, 95(12), 7346-7354. doi: 10.3168/jds.2012-5546.
- Machado, S. C., Fischer, V., Stumpf, M. T., & Stivanin, S. C. B. (2017). Seasonal variation, method of determination of bovine milk stability, and its relation with physical, chemical, and sanitary

- characteristics of raw milk. *Revista Brasileira de Zootecnia*, 46(4), 340-347. doi: 10.1590/S1806-92902017000400010.
- National Research Council [NRC]. (2001). *Nutrient Requirements of Dairy Cattle* (7th ed.). Washington, DC: National Academy Press.
- Norman, G. (2010). Likert scales, levels of measurement and the “laws” of statistics. *Advances in health sciences education*, 15(5), 625-632. doi: 10.1007/s10459-010-9222-y.
- Paixão, M. G., Lopes, M. A., Costa, G. M., Souza, G. N., Abreu, L. R., & Pinto, S. M. (2017). Milk quality and financial management at different scales of production on dairy farms located in the south of Minas Gerais state, Brazil. *Revista Ceres*, 64(3), 213-221. doi: 10.1590/0034-737X201764030001.
- Peel, M. C., Finlayson, B. L., & McMahon, T. A. (2007). Updated world map of the Köppen-Geiger climate classification. *Hydrology and earth system sciences discussions*, 4(2), 439-473. doi: 10.5194/hess-11-1633-2007.
- Reche, N. L. M., Neto, A. T., Loredana, D., Felipus, N. C., Pereira, L. C., Cardozo, L. L., ... Picinin, L. C. A. (2015). Multiplicação microbiana no leite cru armazenado em tanques de expansão direta. *Ciência Rural*, 45(5), 828-834. doi: 10.1590/0103-8478cr20140542.
- Ribas, N. P., Junior, P. R., de Andrade, U. V. C., Valotto, A. A., de Jesus, C. P., & de Almeida, M. C. (2014). Escore de células somáticas e sua relação com os componentes do leite amostrados de tanque no estado do Paraná. *Archives of Veterinary Science*, 19(3), 14-23.
- Statistical Analysis System [SAS]. (2000). *SAS/STAT UserGguide, Version 8.1*. Cary, NC: SAS Institute Inc.
- Tremblay, M., Hess, J. P., Christenson, B. M., McIntyre, K. K., Smink, B., van der Kamp, A. J., ... Döpfer, D. (2016). Customized recommendations for production management clusters of North American automatic milking systems. *Journal of Dairy Science*, 99(7), 5671-5680. doi: 10.3168/jds.2015-10153.