

Blood Sugar and Protein Levels in the Milk of Dairy Cattle Based on Their Physiological State

Claudia Pânzaru¹, Marius Gheorghe Doliş¹, Răzvan Mihail Radu-Rusu¹,
Mădălina Alexandra Davidescu^{1*}

¹Iasi University of Life Sciences, Faculty of Food and Animal Sciences, Romania

Abstract

This study aimed to determine the correlations between blood glucose levels, milk protein levels, and the physiological status of cows in four categories within the Research and Development Unit for Cattle Farming in Dancu, Iaşi (pregnant cows aged between 4-6 years; cows aged 3-5 years, in the 3rd and 5th month of gestation, and were clinically healthy; cows aged 5-6 years, at 48 hours postpartum, and clinically healthy; cows aged 4-6 years, in the 3rd month of lactation, with subclinical mastitis). The results revealed blood glucose values ranging from 28.81 ± 7.15 mg/dl to 50.34 ± 10.31 mg/dl, which are within the normal range for the physiological state of the cows in these groups. The protein percentage ranged from 2.71% to 7.29%, with the lowest value observed in cows with subclinical mastitis and the highest in those in the colostrum period; these results indicate normal values for their specific stages. The findings demonstrate that the studied parameters align with the physiological state of the analysed cows. Overall, the results underscore the importance of metabolic monitoring and the enhancement of housing conditions for dairy cows in this unit.

Keywords: blood, cattle, glucose, milk, protein.

1. Introduction

Some data from the specialized literature indicate that the level of blood glucose accurately reflects the extent to which the animals' energy needs are met. Additionally, there are data suggesting that the level of proteins in milk can be used to assess the satisfaction of energy requirements. Blood glucose levels represent the indicator used to determine these requirements [1]. Furthermore, the determination of milk proteins provides information about the energy supply of the animals [2].

There is a wide range of environmental and housing factors that can influence the productivity and well-being of the animals, such as: temperature and climate conditions (cattle are

sensitive to temperature extremes; high temperatures can lead to heat stress, reducing feed intake and milk production. Adequate ventilation and cooling systems in housing facilities are crucial to mitigate heat stress); humidity (high humidity levels can exacerbate heat stress in cattle. Proper ventilation and drainage systems are essential to maintain optimal humidity levels within housing facilities); feed quality and availability (adequate nutrition is vital for milk production. Studies [3] often assess the quality and availability of feed, including factors such as protein content, fiber content, and access to fresh forage; water quality and availability (clean and readily available water is essential for maintaining hydration and milk production in cattle); housing design and density (the design and layout of housing facilities can significantly impact cattle comfort and productivity. Factors such as stall size, bedding material, and stocking density are often considered in studies on milk production); social environment (cattle are social animals, and

* Corresponding author: Mădălina Davidescu
Tel. 0332/407582, Email: mada.davidescu@gmail.com

their social interactions can influence stress levels and behavior, thereby affecting milk production. Studies [4] may assess the impact of group size, social hierarchy, and housing arrangements on milk yield); health and disease management: Disease outbreaks and health issues can have a significant impact on milk production; [5]; stress reduction measures (minimizing stressors in the environment, such as loud noises, overcrowding, or abrupt changes in routine, can help maintain optimal milk production levels; practices may explore the effects of stress reduction techniques, such as providing environmental enrichment or implementing low-stress handling methods); genetics and breeding: genetic factors play a crucial role in determining milk production potential in cattle); management practices (overall farm management practices, including milking routines, handling procedures, and staff training, can impact milk production efficiency and animal welfare).

Considering that the health of cows affects both the quantity and quality of the milk obtained, it was considered useful to conduct a study to observe aspects related to animal maintenance and how environmental conditions influence production. This can help take measures to optimize the factors contributing to these aspects.

2. Materials and methods

The analyzed material consisted of 43 cows from the Research and Development Unit for Cattle Farming in Dancu, Iași, Romania. They were divided into groups as follows: batch 1 consisted of 10 pregnant cows aged between 4 and 6 years; batch 2 comprised 15 cows aged 3-5 years, in the 3rd and 5th month of gestation, and were clinically healthy; batch 3 was composed of 8 cows aged 5-6 years, at 48 hours postpartum, and were clinically healthy; batch 4 consisted of 10 cows aged 4-6 years, in the 3rd month of lactation, with subclinical mastitis.

The working methods included: determining blood glucose levels, from plasma obtained from morning blood samples, collected from the jugular vein in EDTA vacuum tubes (Ethylenediaminetetraacetic acid). The method used was the ortho-toluidine method, and the readings were taken with a spectrophotometer in a 1 cm cuvette at $\eta=560$ nm. The calculation of the

glucose quantity was done using the formula $EP/ES \times 100 = \text{mg glucose}/100 \text{ ml blood}$ (EP represents the sample's absorbance, and ES represents the standard's absorbance). The protein quantity in milk was determined from individual samples using the Kjeldahl method, with the D. P. Lab Instruments Standard 268 machine.

The data were processed using Microsoft Excel and GraphPad Prism 8.4.2 (\bar{X} , $\pm s\bar{x}$, s, V%).

3. Results and discussion

In Table 1, the data regarding the blood glucose values for the four groups of cows are represented. The results indicated the highest average values in batch 1 (50.34 ± 10.31 mg/dl) and the lowest in batch 3 (28.81 ± 7.15 mg/dl). The absolute minimum value was 19.23 mg/dl, corresponding to the onset of lactation, the presence of an energy deficit during pregnancy, decreased liver function, or a negative hepatic balance (in cows that calved 48 hours before sample collection and were clinically healthy). The absolute maximum value was 62.35 mg/dl (lactating cows in the seventh month and pregnant in the fifth month, clinically healthy). As a result, the coefficient of variation exhibited significant fluctuations, ranging between 20.12% and 25.71%, indicating that this characteristic is highly heterogeneous within the studied population.

In Figure 1, comparisons were made between the values obtained in the four groups and the data from the literature [6]. It can be observed that the average values from the literature (65 mg/dl) are higher than the values obtained in all four groups (Batch 1=50.34 mg/dl; Batch 2=50.05 mg/dl; Batch 3=28.81 mg/dl; Batch 4=40.74 mg/dl). However, the maximum values are closer to those in the literature, except for batch 3, where the maximum value is approximately half of what is described by other authors [7]. In Romania, normal values are reported as follows: 47.06 ± 7.73 in early lactation (4 weeks after calving), 48.32 ± 9.90 in mid-lactation for pregnant animals in the 2nd and 3rd months, and 48.16 ± 8.76 post-weaning at 8 weeks postpartum.

It has been shown that the early lactation period is the most suitable for assessing the nutritional status of dairy cows because deficiencies are at their maximum and uncompensated during that interval [8]. In ruminants, vitamin B12 influences

the use of propionic acid in the hepatic gluconeogenesis process. Hyperglycemia occurs in cases of stress (catecholamine release), after

glucocorticoid treatment, which promotes gluconeogenesis, in magnesium deficiency, and likely in copper deficiency [9].

Table 1. The blood sugar levels, the average values and statistical indices for each batch (mg/dl)

Individual	Batch 1	Batch 2	Batch 3	Batch 4
1	45.81	30.20	20.78	52.15
2	60.55	41.56	26.15	47.42
3	62.35	60.22	19.23	41.10
4	61.90	52.00	32.84	63.25
5	54.00	54.54	26.00	39.72
6	50.12	49.17	37.50	51.17
7	33.15	37.42	30.44	54.30
8	42.38	69.11	28.60	43.88
9	37.82	29.16	-	66.55
10	55.35	55.74	-	-
11	-	46.27	-	-
12	-	69.00	-	-
13	56.2	56.23	-	-
14	-	62.73	-	-
15	-	37.44	-	-
X	50.34	50.05	28.81	40.74
$\pm s\bar{x}$	10.31	12.87	7.15	10.01
V%	20.48	25.71	24.81	20.12
Min.	33.15	29.16	19.23	36.88
Max.	62.35	69.11	39.11	66.55

X=the average values of blood sugar; $\pm s\bar{x}$ =standard error of mean; V%=the coefficient of variation; Min.=minimum value; Max.=maximum value.

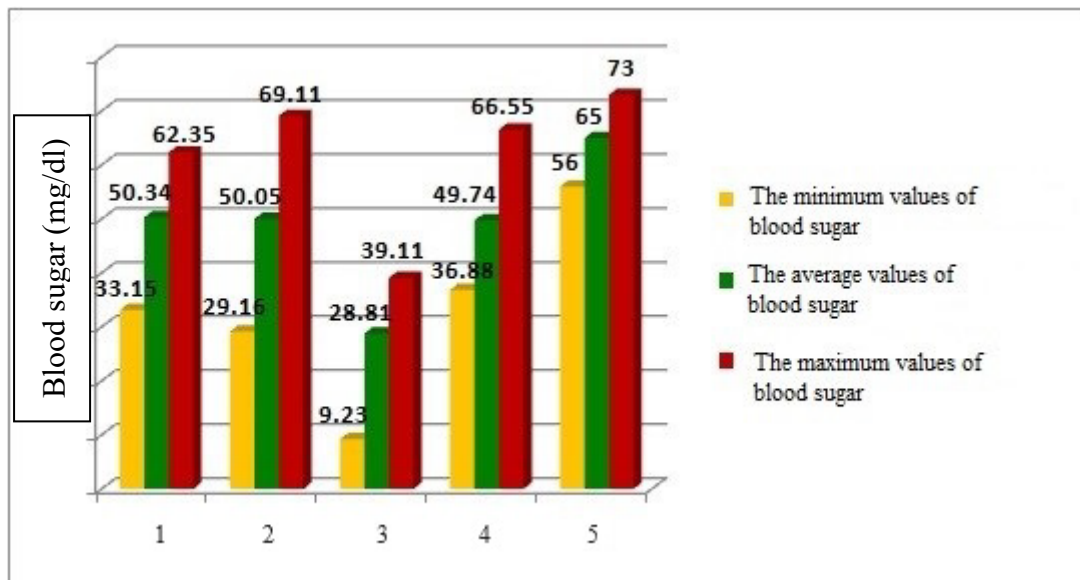


Figure 1. The comparison between the values of blood sugar and the literature

Figure 2 illustrates the comparison between the protein levels in milk obtained from the studied batches and data from the literature. It was observed that, in all cases, the protein levels exceeded the maximum values reported by other authors.

Notably, in batch 3, significant differences were noted due to the colostrum's specific characteristics, as this group was examined 48 hours after parturition (the maximum value is nearly double when compared to the normal protein values in milk: 8.4 g/dl).

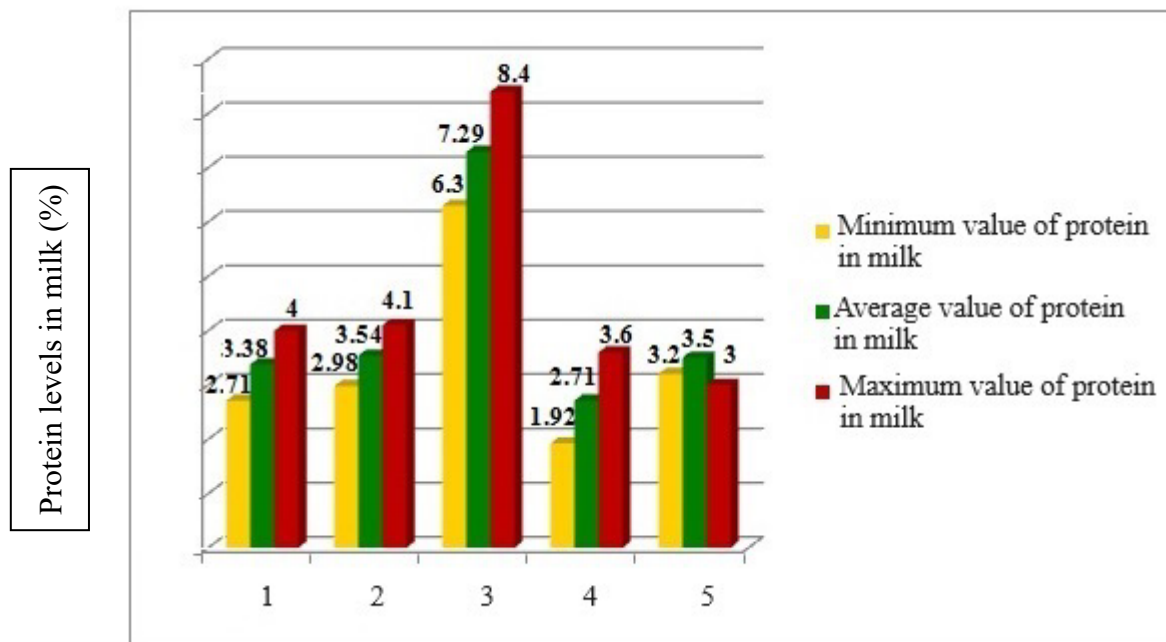


Figure 2. The levels of protein percent in milk (%)

In Table 2, the protein percentage is expressed, with average values ranging from 2.71% to 7.29%. The lowest value was recorded in batch 4 (cows aged 4-6 years, in the 3rd month of lactation, with subclinical mastitis). The statistically significant reduction observed in batch 4 (2.71 ± 0.57 g/dl) indicates the health status of the animals in this group, which consists of dairy cows with subclinical mastitis. Some authors have shown that dairy cows with mastitis experience a decrease in milk fat, casein, and lactose. The amount of casein in cows with mastitis was 2.25 g/dl, while in clinically healthy cows, it registered values of 2.79 g/dl, representing a decrease of 15-20% [9].

Other authors mention that milk's total casein content accounts for about 80% of all the proteins in milk [10].

For batch 3, the milk proteins had an average of 7.29 ± 0.76 g/dl, and the difference compared to batches 1, 2, and 4 was statistically significant. This batch consisted of clinically healthy cows, and the samples were collected 48 hours after calving. Colostrum is richer in total proteins, casein, fats, vitamins, and minerals (protein 16.78 g/dl, casein 3.45 g/dl), with values decreasing to 4.25 and 2.92 g/dl, respectively, at 3 days after calving [10].

Based on the primary data regarding blood glucose and milk proteins, the correlation coefficient (r), the standard error of the correlation

coefficient (Sr), and the significance of the correlation coefficient were calculated, and the results are presented in Table 3. Significant correlations ($p \leq 0.001$) were evident in batches 2 and 3: 0.873 ± 0.061 and 0.954 ± 0.031 , respectively. Correlations were non-significant for batches 1 and 4. Blood glucose levels allow us to assess the extent to which the body's energy needs are met. It was considered that milk protein levels accurately reflect how the energy requirements are met in dairy cows [1]. Milk proteins can also provide accurate information about the energy supply of the animals. This is obvious in the data obtained in batch 2, consisting of cows in the 5th month of lactation. However, it is not reflected in the data recorded in batch 1, consisting of cows in the 7th month of lactation. The increased demands related to the intensive development of the fetus in the latter part of gestation, involving plastic substances and proteins from the mother's body, as well as their levels in milk, could explain the absence of correlation in batch 1.

Regarding batch 3, a positive correlation can be observed between the low blood glucose level, which primarily reflects the energy deficit during gestation, and the physiologically high level of proteins in colostrum. In the case of batch 4, consisting of cows with subclinical mastitis, the pathological reduction in proteins, especially total caseins in milk, rendered the studied correlation non-significant

Table 2. The protein levels, the average values and statistical indices for each batch (mg/dl)

Individual	Batch 1	Batch 2	Batch 3	Batch 4
1	2.71	3.33	6.40	2.21
2	4.00	3.56	7.16	3.60
3	3.62	3.80	6.30	2.78
4	3.25	3.76	7.25	2.00
5	4.93	3.78	7.33	2.66
6	3.00	3.44	8.40	2.75
7	3.17	3.01	8.31	3.21
8	3.38	4.10	7.21	2.55
9	3.45	2.98	-	1.92
10	3.27	3.86	-	3.44
11	-	3.00	-	-
12	-	3.88	-	-
13	-	3.74	-	-
14	-	3.80	-	-
15	-	3.12	-	-
X	3.38	3.54	7.29	2.71
$\pm s\bar{x}$	0.39	0.36	0.76	0.57
V%	12.53	10.36	10.42	21.8
Min.	2.71	2.98	6.30	1.92
Max.	4.00	4.10	8.40	3.60

X=the average values of protein levels in milk (%); $\pm s\bar{x}$ =standard error of mean; V%=the coefficient of variation; Min.=minimum value; Max.=maximum value.

Based on the primary data regarding blood glucose and milk proteins, the correlation coefficient (r), the standard error of the correlation coefficient (Sr), and the significance of the correlation coefficient were calculated, and the results are presented in Table 3. Significant correlations ($p \leq 0.001$) were evident in batches 2 and 3: 0.873 ± 0.061 and 0.954 ± 0.031 , respectively. Correlations were non-significant for batches 1 and 4. Blood glucose levels allow us to assess the extent to which the body's energy needs are met. It was considered that milk protein levels accurately reflect how the energy requirements are met in dairy cows [1]. Milk proteins can also provide accurate information about the energy supply of the animals. This is obvious in the data obtained in batch 2, consisting of cows in the 5th

month of lactation. However, it is not reflected in the data recorded in batch 1, consisting of cows in the 7th month of lactation. The increased demands related to the intensive development of the fetus in the latter part of gestation, involving plastic substances and proteins from the mother's body, as well as their levels in milk, could explain the absence of correlation in batch 1.

Regarding batch 3, a positive correlation can be observed between the low blood glucose level, which primarily reflects the energy deficit during gestation, and the physiologically high level of proteins in colostrum. In the case of batch 4, consisting of cows with subclinical mastitis, the pathological reduction in proteins, especially total caseins in milk, rendered the studied correlation non-significant.

Table 3. The significance between the blood sugar and the protein level

Batch	$\pm r$	$\pm S_r$	Significance
1	0.409	0.364	$p \geq 0.05$
2	0.873	0.061	$p \leq 0.001$
3	0.954	0.031	$p \leq 0.001$
4	0.232	0.299	$p \geq 0.05$

r=correlation coefficient; S_r =standard error

4. Conclusions

Based on the results, the following conclusions can be drawn:

In the case of the studied batches the blood glucose levels varied within normal limits, except for batch 3, where statistically significant hypoglycemia was observed.

Milk protein levels remained within physiological limits for batches 1 and 2. The highest value registered in batch 3 was attributed to the colostrum's unique secretion characteristics. In batch 4, milk proteins experienced a statistically significant decrease ($p \leq 0.001$), which is specific to the conditions present in these animals.

Batch 3 exhibited positive correlations, although statistically non-significant, between blood glucose levels and milk protein levels, especially in colostrum secretion

References

1. Maciuc, V., Managementul creșterii bovinelor, Alfa Publ., Iași, 2006, pp. 56-59.
2. Ruginosu, E., Fiziologia și fiziopatologia parturii și post-parturii, PIM Publ., Iași, 2008, pp.71-73.
3. Belibasakis, N. G., Progia, E., Papaioannou, A., Comparison of maize and alfalfa silages on milk production, milk composition and blood components of dairy cows, *Vet.med.*, 1997, 42(8), 239-242
4. Bond, G.B., de Almeida, R., Ostrensky, A., Molento, C. F. M., Welfare assessment methods and critical points for dairy cattle, *Ciencia rural*, 2012, 42(7), 1286-1293,
5. Eslamizad, M., Dehghan-Banadaky, M., Rezayazdi, K., Moradi-Shahrbabak, M., *J. of Dairy Sci.*, 2010, 93(9), 4054-4061
6. Doreau, M., Boulot, S., Martinrosset, W., Effect of parity and physiological-state on intake, milk-production and blood parameters in lactating mares differing in body size, *Ani. Prod.*, part 1, 1992, 53, 111-118
7. Rhind, S. M., Bass, J., Doney, J. M., Pattern of milk-production of east Friesland and Scottish blackface ewes and associated blood metabolite and hormone profiles, *Ani. Prod.*, part 2, 1992, 54, 265-273
8. Mărgărint, I., Boișteanu, P. C., Halga, P., Bazele morfologice ale producției de lapte, *Vasiliana 98 Publ.*, Iași, 2001, pp. 102-106.
9. Megahed, A. A., Hiew, M. W. H., Townsend, J. R., Messick, J. B., Constable, P. D., Evaluation of an Electrochemical Point-of-Care Meter for Measuring Glucose Concentration in Blood from Periparturient Dairy Cattle, *Journal of Veterinary Internal Medicine*, 2015, 29(6), 1718-1727
10. Ivanova, R., Nikolov, V., Malinova, R., Study on hematological characteristics of native Rhodope Shorthorn cattle breed, *Scientific papers-series D-Animal science*, 2020, 63(2), 266-271.