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Article

Correlations between Selected Blood Markers of Hematological Profile and Milk Components in Dairy Cows

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Simple Summary: Clinical and laboratory diagnostics are especially important in dairy cows, whose metabolism is under constant pressure. Correlations between various milk and blood parameters may be very useful in veterinary diagnostics and in cattle practice. The aim of the study was to analyze the correlation between main hematological biomarkers and quantitative-qualitative parameters of cows' milk. Samples were collected three times at daily intervals from 11 clinically healthy late-lactation dairy cows, obtaining 33 sets of test results. In these dairy cows, significant positive correlations were recorded between milk production and HGB, HCT, MCV or MHC. Significant negative correlations were also found between SCC and HGB, SCC and HCT, SNF and MCV, SNF and MHC, and between MF and RBC. The efficiency of milk production in cows, as well as the composition of milk, is significantly dependent on almost all tested blood parameters of the hematological profile.

Abstract: Clinical and laboratory diagnostics are particularly important in monitoring the internal homeostasis of the body in high-yielding animals, especially in dairy cows, whose metabolism is under constant pressure to obtain high parameters of milk production and reproduction. Knowledge of laboratory test results and relationships based on significant correlations between various milk and blood parameters may be very useful in veterinary diagnostics and helpful in cattle practice in order to protect animal health and the quality of milk production. The aim of the study was to analyze the correlation between selected hematological biomarkers such as white blood corpuscles (WBC), red blood corpuscles (RBC), hemoglobin concentration (HGB), hematocrit (HCT), platelets (PLT), erythrocyte mean corpuscular volume (MCV), mean corpuscular hemoglobin content (MHC), mean corpuscular hemoglobin concentration (MCHC) and selected quantitative-qualitative parameters of cow's milk such as current milk production, somatic cells count (SCC), colony of bacteria forming units (CFU), milk fat (MF), milk protein (MP), milk lactose (ML), milk solids (S), solids non-fat in milk (SNF), daily milk production. Samples were collected three times at daily intervals from 11 clinically healthy late-lactation dairy cows obtaining 33 sets of test results. This repeated study of cows allowed for obtaining a homogeneous group in order to determine statistical correlations between selected blood and milk parameters. Many significant correlations were found between the examined parameters. In cows, significant positive correlations were recorded between milk production and HGB, HCT, MCV or MHC. Significant negative

correlations were also found between SCC and HGB, SCC and HCT, SNF and MCV, SNF and MHC, and between MF and RBC.

Keywords: dairy cows; blood; hematological profile; milk components; correlations

1. Introduction

Hematology and biochemistry tests are commonly performed in medical and veterinary diagnostics and are useful for examining the function of various important organs and body systems [31]. In dairy cows, the efficiency of these functions is particularly important for maintaining the required health of these animals, their fetus during pregnancy and obtaining high quality and quantity of milk during lactation [10,13,14,17,36]. Moreover, cows with high milk yields are significantly at risk of developing metabolic and deficiency diseases, as well as problems with the efficiency of the hematological system [12,16]. Different researchers indicate that both blood and milk tests are very useful in monitoring of health, as well as in the diagnosis of health problems in dairy cows [11,21,24]. The results of various blood tests analyzed in relation to the clinical condition of the cows, their milk yield, mammary gland quality and milk composition are particularly important in cattle practice from the point of view of animal welfare and the quality of dairy products [17,25,27,28]. In addition a milk sample can have great potential in detection of mastitis, assessing the composition of milk as well as in predicting blood metabolites and the metabolic status of cows [2,3,15,20,26]. High-production cows, in order to maintain the required parameters of fertility and achieve record milk volumes, needs the organs involved to be highly efficient metabolically, including blood as a tissue by enabling adequate intensive gas exchange and a supply of nutrients [6,7,33]. Furthermore, the results of other recent studies confirm that parameters related to metabolism, including blood oxygen and glucose, increase significantly in highly productive animals, and the quality of the blood cells that supports these processes is of significant importance for the health and level of milk yield of cows [29]. In conclusion, the authors pointed out that the reference values should be considered in the light of the stage of lactation and the level of milk production, as they may have an effect on the interpretation of changes in these parameters. It can be seen in various scientific studies, that the levels of the hematological parameters of cows are related to many factors, including the quantity and quality of milk parameters, and therefore requires analysis in different conditions and stages of lactation. Moreover, a basic health problem related to the hematological profile and the red blood cell system, such as anemia, regardless of its causal type, by definition, cannot be positively associated with high standards of animal welfare or productivity [1,32]. According to the studies of the cited authors, the hematological profile in some cases is a very important or the only certain way to diagnose health problems related to the functions of the blood system. In cattle practice, many dairy farms rarely use the hematological profile in herd monitoring, despite the real risk of various red blood cell disorders, which can be caused by many factors. For better monitoring of the dairy herd and production forecasting on a modern farm, it is also important to have basic knowledge whether or how and to what extent the individual blood parameters of the hematological profile are related to the milk parameters of cows. For this reason, among others, the aim of our work was to examine the value of interrelationships between selected blood markers in the hematological profile and milk parameters, using a model of tests repeated at daily intervals in late-lactating cows.

2. Materials and Methods

2.1. Animals and Study Design

The study protocol was approved by the 2nd Local Bioethics Committee in Wrocław (decision no. 24/2007). The material consisted of 11 randomly selected Holstein Friesian black and white cows aged 4-5 years, which showed similar BCS and were tested under similar conditions on the same dairy farm, in the autumn season, in the second half of the lactation period. The herd of cows was

under constant veterinary control, free from infectious diseases, and the average milk yield for the lactation period was about 8,500 kg. The cows in the clinical trials showed no signs of disease, but were in a herd with the usual risks for high-yielding animals, including the risk of developing metabolic disorders or mastitis. Each of the 11 selected cows was tested three times at daily intervals, resulting in 33 sets of blood and milk samples over three days. Milk samples were collected during morning milking around 5 am and blood samples were taken immediately after milking. The cows were kept under the same favorable environmental conditions in a hall for 90 milking cows with free access to beds, feed and fresh water. The cows were fed total mixed ration (TMR) served on the feed table. TMR was offered ad libitum twice a day in the morning and afternoon. TMR was developed in accordance with the INRA system using INRAtion 4.0 software, based on appropriate procedures for dietary components according to the 18th edition of official methods of analysis (AOAC) [5]. This feeding was supplemented with a commercial concentrate feed to cows in electronically controlled stations based on their current milk production. These animals had a high level of living comfort due to such equipment as special insulating mattresses on the positions, an automatic manure removal system, an automatic ventilation system and others. On the farm, the Alpro - Delaval milking system was used as a Parallel 2x4.

2.2. Blood Analysis

Blood samples were collected from the external jugular vein using Sarstedt, K3 EDTA 2 mL tubes. Hematological blood tests involved number of white blood corpuscles (WBC), red blood corpuscles (RBC), platelets (PLT), hemoglobin concentration (HGB), hematocrit (HCT), as well as mean corpuscle volume (MCV), mean corpuscular hemoglobin content (MHC) and mean corpuscular hemoglobin concentration (MCHC) in erythrocytes. These blood tests were performed in a specialized laboratory in the Department of Internal Medicine of Faculty of Veterinary Medicine in our University using the analyzer Animal Blood Counter abc vet – Horiba medical.

2.3. Milk Analysis

Milk samples were collected automatically from all milking phases of the cows. They were collected in a special tank used in the trial milking, and then placed in sterile polypropylene containers with a capacity of 50 mL (Prolab, Poland) for transport. These milk samples were analyzed using selected parameters of milk such as the somatic cell count (SCC), the number of colony forming units (CFU), milk fat (MF), milk protein (MP), milk lactose (ML), milk solids (S), and solids non-fat in milk (SNF) and current milk production received during the morning milking. Analyses of milk samples were performed in the specialized laboratory for milk in our University. The SCC tests were performed using an analyzer SOMACOUNT-150 Bentley (USA) by employing the fluoro-opto-electronic method. The determination of CFU determination was performed on the analyzer BACTOCOUNT-70 Bentley (USA) - which uses flow cytometry. Determination of the fractions of milk such as MF, MP, ML, S and SNF was done by the use the analyzer Milko-Scan 133 B Foss Electric (Denmark). The volume of milk yield during milking was recorded using the weight flow in the electronic system of Alpro - Delaval.

2.4. Statistical Analysis

Statistical analysis was carried out using the Statistica 10 StatSoft Inc. program. The samples were tested for normal distribution using the Shapiro-Wilk test. Linear regression was used to assess the strength and direction of relationships between variables of individual compared parameters and the Pearson Correlation Coefficient (PCC) (Person's r) was calculated. Each determined coefficient was also tested for statistical significance with the Student's t -test for $n-2$ degrees of freedom. The number of samples was 33 and the value of the correlation coefficient defining the threshold for a statistical significance level of 5% ($P < 0.05$) was $r = 0.344$. All the values of Pearson's negative or positive correlation coefficients between the variables, including those indicating their statistical significance, are presented in the appropriate Tables 1 and 2.

3. Results

The results of correlations between selected hematological profile markers and milk composition parameters in cows are shown in Tables 1 and 2. Significant statistical correlations are specifically marked.

WBC was significantly negatively correlated with MCHC ($r=-0.354$). RBC was significantly positively correlated to such parameters as: HGB ($r=0.532$), and HCT ($r=0.656$), but RBC was significantly negatively correlated with such parameters as: PLT ($r=-0.427$), MCV ($r=-0.494$), MHC ($r=-0.664$) and MCHC ($r=-0.598$). HGB was significantly positively correlated with HCT ($r=0.946$) and MCV ($r=0.411$), but HGB was significantly negatively correlated with PLT ($r=-0.526$). HCT was significantly negatively correlated with PLT ($r=-0.531$) and MCHC ($r=-0.580$). MCV was significantly positively correlated with MHC ($r=0.938$). MHC was significantly positively correlated with MCHC ($r=0.409$).

Table 1. Correlation results between the tested hematological markers in cows.

	WBC	RBC	HGB	HCT	PLT	MCV	MHC
WBC							
RBC	-0.217						
HGB	0.103	0.532*					
HCT	-0.027	0.656*	0.946				
PLT	0.133	-0.427*	-0.526*	-0.531*			
MCV	0.218	-0.494*	0.411*	0.318	-0.129		
MHC	0.320	-0.664*	0.268	0.089	-0.019	0.938*	
MCHC	-0.354*	-0.598*	-0.290	-0.580*	0.262	0.083	0.409*

* - Significant statistical correlation ($r=0.344$ indicates $p<0.05$), WBC white blood corpuscles, RBC – red blood corpuscles, HGB – hemoglobin concentration, HCT – hematocrit, PLT – platelets, MCV – mean cell volume, MHC – mean corpuscular hemoglobin content, MCHC -mean corpuscular hemoglobin concentration.

WBC was significantly negatively correlated with ML ($r= -0.356$). RBC was significantly negatively correlated with MF ($r=-0.346$). HGB was significantly positively correlated with milk production ($r=0.408$), but hemoglobin was significantly negatively correlated with SCC ($r=-0.371$). Similarly, hematocrit was positively correlated with milk production on the level ($r=0.355$) and negatively with somatic cells in milk ($r=-0.414$). In the case of the PLT blood parameter, this was significantly positively correlated with SCC in milk ($r=0.346$). MCV was significantly positively correlated with milk production ($r=0.455$), but it was significantly negatively correlated with MP ($r=-0.340$) and SNF ($r=-0.358$). MHC was significantly positively correlated with milk production ($r=0.385$), and MF ($r=0.341$) but was also significantly negatively correlated with SCC ($r=-0.361$).

Table 2. Results of correlation between selected blood markers of hematological profile and milk components in cows.

	WBC	RBC	HGB	HCT	PLT	MCV	MHC	MCHC
Production	0.033	-0.013	0.408	0.355	-0.219	0.455	0.385	-0.051
CFU	-0.083	-0.033	-0.054	-0.022	-0.181	0.066	0.056	-0.036
SCC	0.143	-0.266	-0.371	-0.414	0.346	-0.158	-0.361	0.282
MF	0.169	-0.346	-0.087	-0.165	0.265	0.303	0.341	0.268
MP	0.147	0.206	-0.076	-0.027	0.048	-0.340	-0.314	0.079
ML	-0.356	0.265	0.154	0.213	0.225	-0.129	-0.185	-0.229
S	0.187	-0.309	-0.092	-0.162	0.280	0.249	0.292	0.256
SNF	-0.036	0.305	0.007	0.077	-0.066	-0.358	-0.361	-0.175

* - Significant statistical correlation ($r = 0.344$ indicates $p<0.05$), WBC - white blood corpuscles, RBC - red blood corpuscles, PLT - platelets, HGB- hemoglobin concentration, HCT- hematocrit, MCV - mean red corpuscle volume, MHC- mean red corpuscular hemoglobin content, MCHC - mean corpuscular hemoglobin

concentration in erythrocytes. CFU – colony forming units, SCC – somatic cell count, MF – milk fat, MP – milk protein, ML – milk lactose, Solids – milk solids, SNF – milk solids non-fat.

Mean values and standard deviations of the tested blood and milk parameters in cows are presented in Table 3.

Table 3. The mean values and standard deviations (SD) of selected parameters of blood and milk in cows.

Parameters of blood and milk	Mean	SD	Unit
WBC	10.14	3.16	G/L
RBC	6.73	0.72	T/L
HGB	6.22	0.51	mmol/L
HCT	0.29	0.03	l/l
PLT	416.12	103.54	G/L
MCV	43.93	3.69	fL
MHC	0.93	0.08	fmol
MCHC	21.13	0.69	mmol/L
*Milk production	16.53	2.63	L
CFU	82.77	52.12	x 1000/mL
SCC	114.36	63.74	x 1000/mL
MF	4.30	1.81	%
MP	3.57	0.42	%
ML	4.64	0.22	%
Solids	13.14	1.56	%
SNF	8.82	0.48	%

WBC – white blood corpuscles, RBC – red blood corpuscles, PLT – platelets, HGB – hemoglobin concentration, HCT – hematocrit, MCV – mean red corpuscle volume, MHC – mean red corpuscular hemoglobin content, MCHC – mean corpuscular hemoglobin concentration in erythrocytes. *Values of milk parameters such as milk production, CFU – colony forming units, SCC – somatic cell count, MF – milk fat, MP – milk protein, ML – milk lactose, Solids – milk solids, SNF – milk solids non-fat as a necessary comparative basis for correlation assessment also for other blood parameters and profiles characterizing the functions of other organs or other body systems were published respectively [24–26].

4. Discussion

The bovine hematology and milk component tests used in our veterinary practice to monitor the health of cows and their production have proven to be useful and relatively inexpensive. The hematological profile and its interpretation are important not only in the diagnosis of disorders of the hematological system, but it is also helpful in diagnosing of many organ and systemic diseases [31]. Clinically healthy cows which were selected for our study were in late lactation and did not show any abnormalities or did not exceed the normal reference range the for the individual hematological parameters in blood tests as quoted by Winnicka [35]. Repeating the tests on the same, selected dairy cows within three days allowed to obtain homogeneous conditions and material for the precise assessment of the level and direction of changes measured by correlations between the tested parameters of blood and milk.

The negative significant correlation between WBC and ML reveals that the increase in the number of leukocytes was accompanied by a decrease in the amount of lactose in milk. An increase in the number of leukocytes in the blood accompanies various inflammatory processes in the clinical or subclinical course, as well as in the field of subclinical forms of mastitis in cows. In such cases, an increase in the number of somatic cells and a decrease in lactose concentration in milk are also observed [2]. Moreover, the cited authors suggest that changes in the lactose content in milk can be used as a diagnostic tool in the prevention of subclinical mastitis in cows.

The negative correlation between RBC and MF indicates that an increase in the number of erythrocytes is accompanied by a decrease in the amount of fat in milk. Milk fat depends significantly on the type of food fed and is synthesized from fatty acids produced during microbiological fermentation of crude cellulose fiber contained in the feed, and also partially from fatty acids and triglycerides contained in the blood. Their quantity and quality influences changes in RBC, in the composition of fatty acids in the erythrocyte cell membrane and their functions, resulting in a change in the release of ATP by these blood cells [30].

It is also interesting that there is a significant positive correlation between PLT and SCC. This correlation confirms the naturally occurring defense reaction of the body triggered by cytokine release and observed in connection with stress, inflammation initiated at any time, including subclinical and clinical mastitis with an adequate increase in the number of somatic cells in milk [31]. Moreover, this dependence even may be used for therapeutic administration of bovine platelet concentrates in the treatment of mastitis or metritis in cattle [9].

As the obtained results showed, such parameters as HGB, HCT, MCV MHC had a significant positive impact on milk production. Moreover, the negative significant correlation between HGB or HCT parameters and SCC indicates that the decrease in these blood parameters was accompanied by increases in the number of somatic cells in milk. These results are consistent with the summary of other studies showing differences in hematological values of HF cows with different levels of milk production [29]. In the studies of the cited authors, the observed values of hematological parameters such as HCT and HGB were lower in cows that produced less milk 18–20 kg of milk per cow compared to cows with a higher milk production, above 35 kilograms per cow per day, as well as those that did not exhibit mastitis in the California Mastitis Test (CMT).

The negative significant correlation between MCV and MP reveals that the increase in the mean volume of erythrocytes is accompanied by a decrease in the amount of protein in milk. The increase in the mean corpuscular volume MCV in healthy animals without typical absolute macrocytic anemia can result from a relative transient condition after drinking water or taking succulent feed, which usually effects dilution of the milk and a lower concentration of milk protein. This may be confirmed by the fact that the real increase in mean corpuscular hemoglobin (MCH) in erythrocytes was accompanied by an increase in fat concentration in milk, and ultimately proves that both MCV and MHC correlated negatively with the non-fat milk solids. The level of hydration and type of diet may have an influence on the range of hematological parameters in cattle considered normal [31], as well as on the composition of milk, including the profile of fatty acids in cow's milk [19]. Similarly, other common farm environmental factors affecting the herd, such as stress, including thermal stress, may also affect the hematological picture of blood in cattle [37] and the functions of the mammary gland and milk composition [34].

This confirms that the hematological system has a huge impact not only on maintaining appropriate vital functions, but also on obtaining an appropriate level of milk production in cows. Increased metabolism in high-yielding dairy cows, can be satisfied not only by providing the right amount and quality of nutrients [18], but also by the efficiency of all body systems, including blood tissue and gaseous exchange [8,29]. The volume of milk obtained from cows, shown in the results of our research, is clearly positively correlated with the quality of blood parameters in the hematological profile and the structure of the red blood cell system.

5. Conclusions

Simultaneous analysis of hematological indicators and milk components has proven to be a useful tool for monitoring welfare, health and production in dairy cows. Hematological tests combined with testing the cow milk were useful for our diagnostics of internal homeostasis and allowed for the demonstration of many important, significant relationships between the tested blood and milk parameters. The efficiency of milk production in cows, as well as the composition of milk, significantly depends on almost all tested blood parameters of the hematological profile. These studies increase the importance of using hematological tests to diagnose mammary gland diseases in dairy cows, which may contribute to improving milk yield and herd profitability.

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Data Availability Statement: Data is available upon request.

Conflicts of Interest: The authors declare no conflicts of interest.

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