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Full Length Research Paper

The analysis of variability of pH level and somatic cell count (SCC) in the colostrum and milk of złotnicka white sows

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The aim of the research was to analyze the pH level and somatic cell count (SCC) in sows' colostrum and milk depending on the reproductive cycle, litter size and consecutive pair of teats. The animal specimens were 30 sows of the Złotnicka White breed. Złotnicka pigs were subjected to the National Genetic Resources Conservation Programme. The experiment was made on the farm where pigs of native breeds were kept. The research material was collected in 24 h after parturition and on the 7th, 14th and 21st day of lactation. The somatic cell count (SCC) and pH were labelled in the colostrum and milk. The number and body weight of piglets' and their mortality were checked on the 1st, 7th, 14th, and 21st day of life. As a result of the research, the influence of the consecutive reproductive cycle and litter size on the somatic cell count and pH in sows' colostrum and milk was proved. The older and more numerous the litters were, the higher the somatic cell count was and it ranged between 2.48 and 3.05 × (10^6). On the other hand, the acidity (pH) of colostrum and milk decreased and assumed the values from 6.90 to 6.22. A positive correlation between the pH of colostrum and the weights of piglets aged one day was observed and a negative correlation between the pH of milk on the 21st day and the weights of piglets aged 7, 14 and 21 days. The research also proved dependence between piglets' body weight and the somatic cell count at the level of α ≤ 0.01 and α ≤ 0.05.

Key words: Sows, milk, somatic cell count (SCC), pH, native breeds.

INTRODUCTION

Pig production, like any other human activity, is burdened with potential risk and uncertainty. It is unique due to its cyclic nature. The necessity to include Złotnicka pigs in scientific analyses results from the fact that these breeds are characterised by their small population size, they are subject to the National Genetic Resources Conservation Programme (Górecki, 2003; Buczyński et al., 2008; Szulc et al., 2011) and differences between breeds affect the quality characteristics of sows' milk (Alston-Mills et al., 2000; Buczyński et al., 2001; McNamara and Pettigrew, 2002; Buczyński and Skrzypczak, 2003; Gourdine et al., 2006). The Mastitis-metritis-agalactia (MMA) syndrome causes high losses and dramatic changes; hence it is worthwhile to prevent this disease. Extensive prevention measures need to be introduced. Moreover, it is crucial to diagnose the disease promptly through different diagnostic methods, such as hematological and biological blood analyses, cytological analyses and physical analyses of milk (Rekiel, 1999; Skrzypek et al., 2007; Rekiel et al., 2008; Borne et al., 2011). Therefore, it is justified to start research on qualitative changes in the colostrum and milk of the sows kept as genetic reserve.

MATERIALS AND METHODS

The animals for analyses included 30 Złotnicka white sows. The

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experiments were conducted on the farm where pigs of native breeds were kept. The analyses were made between January 2010 and March 2011. All the sows subject to the experiment were kept in the same conditions that met welfare requirements. The sows were kept in single littering crates from about the 10th day before parturition to the 4th week of lactation. They were fed individually with standard mixture prepared according to Polish Nutrient Requirements for Pig (1993) twice a day with constant access to water. Moreover, the sows in the experiment were naturally mated according to the approved mating plan and their litters came from one boar. Farrowing sows were supervised by personnel. The sows were placed in individual confinement crates. The number of piglets on the 1st day refers to the total number of piglets in the litter. No stillborn piglets were recorded during the experiment.

Due to the specific character of mammary gland secretion in sows (from 40 to 60 s passage of milk in all teats at the same time), colostrum and milk were collected after earlier intramuscular injection of 2 to 4 ml of oxytocin. The amount of oxytocin injected depended on the day of lactation of a given sow rather than the weight of the animal. Samples of about 5 ml of colostrum and milk were manually collected from all active mammary glands to labelled test tubes with a preservative (MILKOSTAT). The teats were numbered in the order from the forelimbs to the tail. The material for analyses was collected 24 h after parturition and on the 7th, 14th and 21st day of lactation. Twenty-four hours after parturition, a total of 16 samples of colostrum were collected from each sow and the same amount of milk was collected on the 7th, 14th and 21st day of the experiment. A total of 1920 samples were collected for analyses (16 teats × 30 sows × 4 days).

During the experiment the following parameters were determined in all the sows:

1) Physical parameter: This includes the acidity (pH) of colostrum and milk from all active teats. Active acidity was determined with electrometry by measuring the activity of hydrogen ions with a pH-meter type pH 211 by Hanna Instruments and a specific electrode for milk measurements after a prior calibration of the apparatus in relation to three buffers with pH values of 4.01, 7.01 and 10.01.
2) Cytological parameter: The somatic cell count (SCC) of colostrum and milk – from all active mammary glands. Somatic cell counts were determined by means of a Somacount type 150 apparatus by Bentley. Due to the small volume of samples under analysis they were diluted at a 1:9 ratio with deionised water produced with a Millipore apparatus by Merck. Flow cytometry was applied to determine somatic cell counts. The samples under analysis were mixed with buffer and a fluorescent dye, which stains cell DNA. The sample of material under analysis injected into a flux of a carrier fluid encountered a laser beam, causing fluorescence of stained cells. A special photodetector detected shining cells and the results of the analysis of data conducted with a computer operation system were recorded on the disc.

The following parameters were monitored during the experiment: number of piglets aged one, seven, 14 and 21 days; body weight of piglets aged one, seven, 14 and 21 days; piglet mortality rates up to the age of 21 days. The source data was divided according to:

1) The reproductive cycle – consecutive litters of the sow (two, three, five, six to eight)
2) The litter size – the number of piglets in a litter (10, 11, 12, 14)
3) The teats (1R, 1L; 2R, 2L; 3R, 3L; 4R, 4L; 5R, 5L; 6R, 6L; 7R, 7L; 8R, 8L)

In order to estimate the significance of statistical differences caused by the abovementioned factors, a multivariate analysis of variance was conducted with Fisher's test (Ruszczyc, 1997) according to the following model:

$$Y_{ij=} \mu + M_i + W_j + S_k + e_{ijk}.$$

Where Y_{ij} is the expected value of a given trait, µ is the grand mean, M_i is fixed effect of i-th litter, reproductive cycle, where i = 1, 2, 3, 4 (2, 3, 5, 6 to 8), W_j is the fixed effect of j-th number of piglets in a litter, where j = 1, 2, 3, 4 (10, 11, 12, 14), S_k is the fixed effect of k-th pair of teats, where k = 1, 2, 3, 4, 5, 6, 7, 8 (1R, 1L; 2R, 2L; 3R, 3L; 4R, 4L; 5R, 5L; 6R, 6L; 7R, 7L; 8R, 8L) and e_{ijk} is the fixed effect of error.

Statistical analysis

The least significant difference (LSD) test for pairs of item means was applied to the groups formed as a result of division of the research specimens according to the factors included in the abovementioned model of analysis of variance. SAS package (2007) based on the ANOVA function was used for calculations according to the PROC GLM procedure with MENES LSD. Pearson's correlation method with SAS statistical package (2007) according to the PROC CORR Pearson procedure was applied to examine the dependence between the data concerning the qualitative traits of colostrum and milk and the weights of piglets. The correlation coefficient was estimated for the entire population under analysis. Due to the fact that the calculations were made by means of the analysis of variance, probit transformation described by Żuk (1998) and Lynch and Welsh (1998) was applied to the traits that are discrete random variables (the number of piglets aged one, seven, 14 and 21 days). It enables transformation from discrete random variables to continuous random variables. In order to obtain normal distribution of the somatic cell count, logarithmic transformation was applied according to the following formula (Ali and Shook, 1980):

$$Y = \ln (x + 10)$$

Where, x is the experimentally obtained somatic cell count.

RESULTS AND DISCUSSION

Although lactation is an essential component of mammalian reproduction, each species has evolved unique lactation strategies that optimise the growth and development of their young (Hartmann et al., 1997). Over many years, analyses conducted on sows' milk have shown that it is the most important component of diet for piglets, determining their rearing and thus having a significant effect on breeding performance (Buczyński et al., 2006; Farmer and Quesnel, 2009). The health condition of sows after parturition, the body weight of reared piglets and percentage of piglet mortality up to the age of 21 days is a measure of effectiveness of the prevention system against the agalactia syndrome or reduced milking performance. The results of experiments show that apart from other factors, piglets' weight gains depend on the amount and quality of consumed milk (Foisnet et al., 2010). The chemical composition of milk, its physical parameters (pH) and cytological parameters (SCC), along with main breeding performance traits (number of piglets born alive and weaned) enable a more comprehensive determination of the production value of sows (Lin et al., 2009; Quesnel, 2011). Figures 1 and 2 show the level of pH and SCC changes during lactation. For the entire population under analysis, the mean pH value increased on consecutive days of lactation and

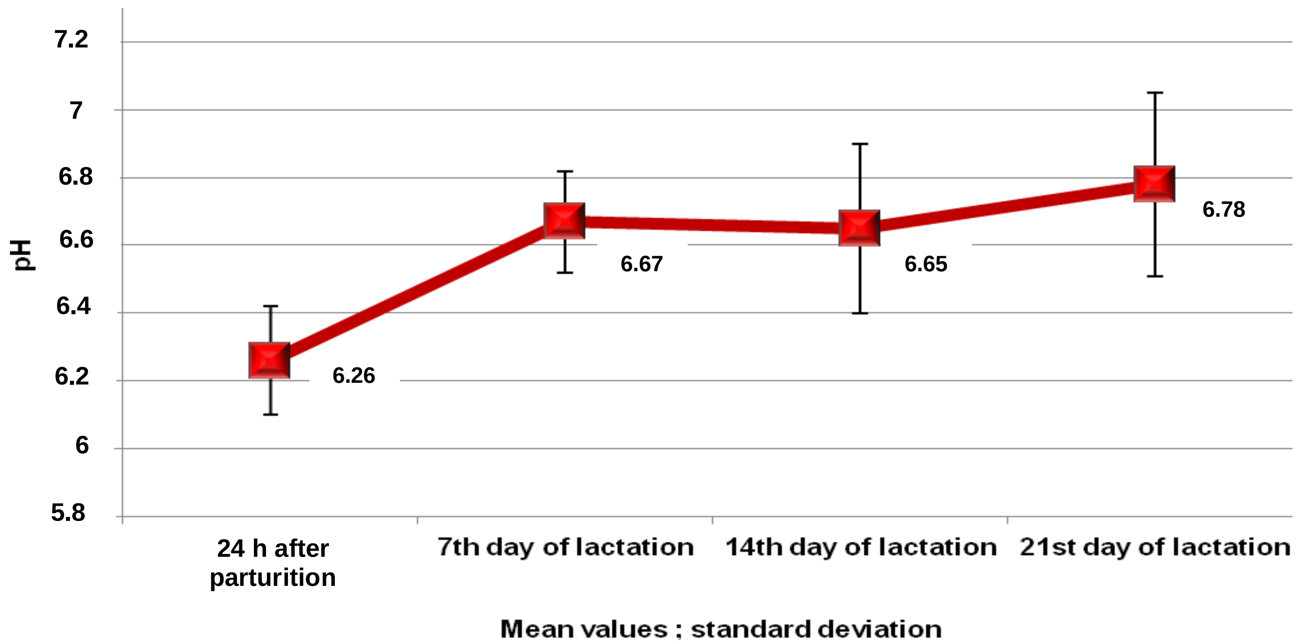

Figure 1. The pH level in sow's colostrums and milk.

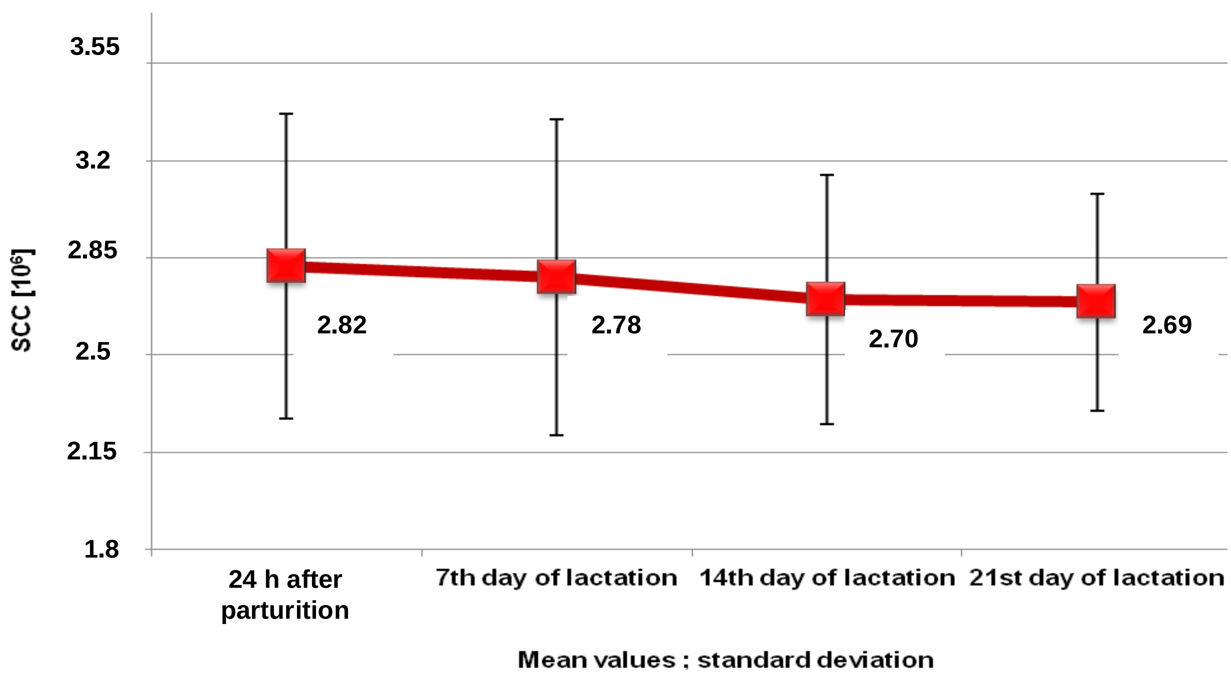

Figure 2. The SCC level in sow's colostrum ll count.

assumed the value of 6.26 ± 0.16 in 24 h after parturition, whereas on the 21st day it amounted to 6.78 ± 0.27. On the other hand, the SCC decreased each day. Furthermore, 24 h after parturition, this trait amounted to 2.82 × (10^6) ± 0.55 to reach the mean value of 2.69 × (10^6) ± 0.39 on the 21st day. On consecutive days of lactation, the pH of milk changes as a result of penetration of the serum from blood vessels to glandular follicles of active mammary glands (Coffey et al., 1982). Rekiel (2002) in her analyses reported acidity of less than 7.0, indicating good health condition of sows and their mammary glands.

Table 1. The qualitative traits of colostrum and milk vs the consecutive reproductive cycle.

Specification	Litter 2 (N = 6)	Litter 3 (N = 6)	Litter 5 (N = 9)	Litter 6, 8 (N = 9)
pH of colostrum in 24 hours after parturition	6.21 ± 0.11^{Bb}	6.29 ± 0.15^{a}	6.31 ± 0.13^{Aa}	6.24 ± 0.20
pH of milk on 7th day of lactation	6.64 ± 0.14^{Bb}	6.62 ± 0.15^{Bb}	6.65 ± 0.14^{Bb}	6.75 ± 0.15^{Aa}
pH of milk on 14th day of lactation	6.76 ± 0.15^{Aa}	6.72 ± 0.15^{Aab}	6.64 ± 0.30^{bc}	6.55 ± 0.26^{Bc}
pH of milk on 21st day of lactation	6.90 ± 0.15^{Aa}	6.70 ± 0.51^{Bb}	6.83 ± 0.13^{a}	6.71 ± 0.15^{Bb}
SCC of colostrum in 24 hours after parturition [10^6]	2.73 ± 0.45^{b}	2.69 ± 0.48^{Bb}	3.01 ± 0.64^{Aa}	2.76 ± 0.53^{b}
SCC of milk on 7th day of lactation [10^6]	2.70 ± 0.48	2.80 ± 0.51	2.76 ± 0.54	2.83 ± 0.68
SCC of milk on 14th day of lactation [10^6]	2.58 ± 0.35^{b}	2.76 ± 0.45^{a}	2.64 ± 0.39^{a}	2.79 ± 0.55^{a}
SCC of milk on 21st day of lactation [10^6]	2.55 ± 0.29^{Bb}	2.65 ± 0.31^{b}	2.67 ± 0.47^{b}	2.82 ± 0.38^{Aa}

Mean ± standard deviation (SD). Means labelled with different letters (A, B**)** are statistically different when $P \leq 0.01$; means labelled with different letters (a, b, c) are statistically different when $P \leq 0.05$.

Table 1 shows the dependence between the qualitative traits of colostrum and milk and the reproductive cycle of sows. The level of pH in colostrum after 24 h following the littering turned out to be statistically significant at $\alpha \leq 0.01$ and $\alpha \leq 0.05$ between litters 2 and 5. On the 7th day, the pH of milk from older sows (those which produced their 6th and 8th litters) increased consistently. The opposite situation was observed for the pH of milk on the 14th and 21st day of lactation. The milk obtained from older sows was characterised by lower acidity. These differences turned out to be statistically significant. During the experiment on the group of sows under analysis, the SCC exceeded the threshold values of over $2.0 \times (10^6)$. Despite that fact no clinical changes were observed. The recorded results indicate that the older the litter, the higher the SCC. This observation was also confirmed by the experiment conducted by Rekiel and Więcek (2002). In consecutive cycles, the percentage of sows with higher SCC increased, which may indicate a deterioration of the health condition of the mammary glands. In addition, Table 2 shows the qualitative traits of colostrum and milk depending on the litter size – the number of piglets. When analysing the pH and SCC of colostrum and milk in this study, the following dependence may be found: the more piglets in the litter, the poorer the quality of milk. In the initial period of mastitis, milk may be unchanged macroscopically, but it has a higher pH, amounting to as much as 7.0 (Rekiel, 2006; Rekiel et al., 2008). Such high pH was not recorded in this experiment, which may indicate that milk came from a mammary gland without inflammatory lesions. Csapo et al. (2001) and Boruta et al. (2009) in their analyses showed that the best parameters of colostrum and milk were found in the litters where the number of piglets was 10 to 12.

Table 3 shows the influence of consecutive pairs of teats on the qualitative traits of colostrum and milk. It was observed that in the posterior segments, pH and SCC assumed lower values than in the anterior segments. The data presented in the literature confirms varying susceptibility to inflammation of individual segments within a gland. Życzko et al. (1986) did not find dependence between the number of cells in milk and the position of the gland from which it came. Also the studies by Kempera and Gerjetsa (2009) did not confirm the influence of the position of the mammary gland on the content of bacteria in sows' milk and thus on piglet rearing. In some sows, as early as during gestation, teats in a well-developed gland but located low (especially in the posterior position) are at risk of contamination and damage, thus promoting infection. However, it was found that in young sows with a well-developed mammary gland, infection occurs most frequently in the anterior and central sections. This is still a debatable problem. Table 4 shows the phenotype correlation between piglet's average body weight and the pH and SCC of colostrum and milk on individual days of the measurement. In our analyses a positive correlation was found between the pH of colostrum and weight of piglets aged one day, whereas a negative correlation was noted between the pH on the 21st day and body weight on the 7th, 14th and 21st days. Moreover, a correlation between the weight of piglets on individual days of life and the SCC was proven at the level of $\alpha \leq 0.01$ and $\alpha \leq 0.05$. Babicz et al. (2011) proved that higher piglets' body weight was obtained when the SCC level was low. The positive relationship between the piglet body weight and milk consumption indicates that older, heavier piglets are able to remove more milk from the mammary glands of lactating sows (King et al, 1997).

To sum up, the results of the investigations proved significant dependence between the pH and somatic cell count (SCC) contained in sows' colostrum and milk and the traits under analysis. The investigations proved the influence of the sow's consecutive litter and its size on the SCC and pH of colostrum and milk. The older and more numerous the litters were, the higher the SCC was and the lower the acidity was. The investigations proved worse quality of colostrum and milk in the posterior segments of the mammary gland. The research also

Table 2. The qualitative traits of colostrum and milk vs. the size of a litter.

Specification	Number of piglets born in litter 10 (head); N=7	Number of piglets born in litter 11 (head); N=10	Number of piglets born in litter 12 (head); N=7	Number of piglets born in litter 14 (head); N=6
pH of colostrum in 24 h after parturition	6.36 ± 0.09^{Aa}	6.26 ± 0.11^{bc}	6.22 ± 0.22^{Bc}	6.31 ± 0.16^{ab}
pH of milk on 7th day of lactation	6.66 ± 0.09^{bc}	6.64 ± 0.15^{ab}	6.57 ± 0.15^{Aa}	6.74 ± 0.13^{Bc}
pH of milk on 14th day of lactation	6.74 ± 0.13^{a}	6.66 ± 0.20	6.60 ± 0.33^{b}	6.67 ± 0.23
pH of milk on 21st day of lactation	6.81 ± 0.12	6.79 ± 0.16	6.73 ± 0.43^{b}	6.89 ± 0.08^{a}
SCC of colostrum in 24 h after parturition [10^6]	2.89 ± 0.34^{bc}	2.65 ± 0.37^{Bc}	2.97 ± 0.66^{ab}	3.05 ± 0.80^{Aa}
SCC of milk on 7th day of lactation [10^6]	2.72 ± 0.41	2.65 ± 0.46	3.03 ± 0.73	2.65 ± 0.36
SCC of milk on 14th day of lactation [10^6]	2.62 ± 0.47	2.60 ± 0.41	2.90 ± 0.47	2.63 ± 0.42
SCC of milk on 21st day of lactation [10^6]	2.48 ± 0.13	2.62 ± 0.34	2.89 ± 0.46	2.63 ± 0.34

Mean ± standard deviation (SD). Means labelled with different letters (A, B) are statistically different when P ≤ 0.01; means labelled with different letters (a, b, c) are statistically different when P ≤ 0.05.

Table 3. The qualitative traits of colostrum and milk vs. the pair of teats.

Specification	1st pair of teat (1R, 1L)	2nd pair of teat (2R, 2L)	3rd pair of teat (3R, 3L)	4th pair of teat (4R, 4L)	5th pair of teat (5R, 5L)	6th pair of teat (6R, 6L)	7th pair of teat (7R, 7L)	8th pair of teat (8R, 8L)
pH of colostrum in 24 h after parturition	6.26 ± 0.10^{Bb}	6.32 ± 0.14^{Bb}	6.25 ± 0.14^{Bb}	6.24 ± 0.12^{Bb}	6.28 ± 0.18^{Bb}	6.26 ± 0.12^{Bb}	6.21 ± 0.19^{Bb}	6.56 ± 0.76^{Aa}
pH of milk on 7th day of lactation	6.62 ± 0.09^{Bb}	6.63 ± 0.11^{Bb}	6.70 ± 0.18^{Bb}	6.66 ± 0.09^{Bb}	6.73 ± 0.16^{Bb}	6.73 ± 0.14^{Bb}	6.62 ± 0.19^{Bb}	6.94 ± 0.24^{Aa}
pH of milk on 14th day of lactation	6.70±0.20	6.69±0.15	6.65±0.19	6.70±0.19	6.58±0.40	6.65±0.22	6.60±0.31	6.59±0.02
pH of milk on 21st day of lactation	6.76±0.12	6.80±0.17	6.80±0.15	6.76±0.11	6.86±0.16	6.64 ± 0.63^{b}	6.84±0.15	6.89 ± 0.10^{a}
SCC of colostrum in 24 h after parturition [10^6]	2.75 ± 0.44^{Bbc}	2.60 ± 0.45^{Bc}	2.63 ± 0.32^{Bbc}	2.72 ± 0.62^{Bbc}	3.09 ± 0.76^{b}	2.93 ± 0.42^{Bbc}	2.98 ± 0.55^{Bbc}	3.68 ± 1.28^{Aa}
SCC of milk on 7th day of lactation [10^6]	2.48 ± 0.21^{Cd}	2.49 ± 0.23^{Cd}	2.68 ± 0.59^{BCcd}	2.73 ± 0.54^{BCcd}	3.19 ± 0.72^{Bb}	3.01 ± 0.56^{BCbc}	2.82 ± 0.47^{BCbcd}	3.80 ± 1.27^{Aa}
SCC of milk on 14th day of lactation [10^6]	2.71 ± 0.49^{Bbcd}	2.54 ± 0.40^{Bcd}	2.49 ± 0.26^{Bd}	2.51 ± 0.25^{Bcd}	2.96 ± 0.56^{Bb}	2.87 ± 0.42^{Bbc}	2.77 ± 0.44^{Bbcd}	3.42 ± 0.95^{Aa}
SCC of milk on 21st day of lactation [10^6]	2.48 ± 0.15^{Bd}	2.54 ± 0.18^{Bcd}	2.59 ± 0.23^{bcd}	2.58 ± 0.23^{bcd}	2.97 ± 0.63^{Aa}	2.84 ± 0.33^{ab}	2.86 ± 0.52^{ab}	2.77 ± 0.14^{abc}

Mean ± SD – standard deviation. Means labelled with different letters (A, B) are statistically different when P ≤ 0.01; means labelled with different letters (a, b, c) are statistically different when P ≤ 0.05.

Table 4. The phenotype correlation between the average body weight of piglets and the pH and SCC of colostrum and milk on individual days of the measurement.

Specification	Body weight (aged 1 day (kg))	Body weight (aged 7 days (kg))	Body weight (aged 14 days (kg))	Body weight (aged 21 days (kg))
pH of colostrum in 24 hours after parturition	0.16*	0.04	0.08	0.06
pH of milk on 7th day of lactation	-0.15	-0.12	-0.08	-0.08
pH of milk on 14th day of lactation	-0.04	-0.05	-0.03	-0.07
pH of milk on 21st day of lactation	0.02	-0.17*	-0.21**	-0.20**
SCC of colostrum in 24 hours after parturition [10^6]	-0.12	-0.12	-0.19*	-0.16
SCC of milk on 7th day of lactation [10^6]	-0.28**	-0.21**	-0.16	-0.15
SCC of milk on 14th day of lactation [10^6]	-0.23**	-0.23**	-0.22**	-0.15
SCC of milk on 21st day of lactation [10^6]	-0.30**	-0.24**	-0.26**	-0.20*

*Significance level P ≤ 0.05; ** significance level P ≤ 0.01.

proved dependence between piglets' weights and the somatic cell count at the level of $\alpha \leq 0.01$ and $\alpha \leq 0.05$.

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