

## STUDY ON HEMATOLOGICAL CHARACTERISTICS OF NATIVE RHODOPE SHORTHORN CATTLE BREED

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### Abstract

*We studied some hematological parameters in 36 cows from the autochthonous brachycephal Bulgarian breed - Rhodope short cattle. The blood for the study was taken twice - in spring and autumn, from cows bred in three different regions. We proved that the region and the season have a significant effect on hematological parameters. In the different regions, blood sugar ranges from 3.288 to 4.714 mmol/L, total protein from 50.67 to 58.48 g / L ( $P < 0.001$ ), cholesterol from 2.616 to 2.815 mmol/L, triglycerides from 0.258 to 0.595 mmol/L ( $P < 0.001$ ), creatinine from 217.8 to 254.1  $\mu\text{mol/L}$ , Ca from 1.521 to 1.748 mmol/L, P from 3.073 to 3.218 mmol/L ( $P < 0.05$ ) and Mg from 0.649 to 1.497 mmol/L ( $P < 0.001$ ). Glucose, cholesterol ( $P < 0.001$ ), triglycerides ( $P < 0.05$ ) and minerals Ca, Mg ( $P < 0.001$ ) ( $P < 0.01$ ) levels are higher in the fall compared to the spring while the levels of total protein ( $P < 0.001$ ) and creatinine ( $P < 0.05$ ) are lower in the fall compared to the spring.*

**Key words:** Rhodope short cattle; hematological parameters; season; regions.

### INTRODUCTION

The blood biochemical parameters, which are kept within certain reference values but at the same time are extremely sensitive to conditions leading to change in their functions (Ivanova et al., 2009), are used by the clinical practice as primary indicators of possible occurrence and development of pathological processes. By understanding the specificity and dynamics of the physicochemical and morphological blood changes within reference values and in case of pathology, there are ample opportunities to assess the condition of the individual.

According to Ježek et al., (2006, 2013), the blood serum biochemical analysis is a useful method for ascertaining the metabolic and health status of animals, however, when blood parameters are used, it is necessary detailed studies as they are influenced by the breed, age, physiological condition, and the lactation stage of the cow (Doornenbal, 1988).

In a study of glucose, total protein, hemoglobin, cholesterol, Ca and P levels in the blood of cows from the Bulgarian Rhodope Cattle breed during the autumn-winter period, Ivanova et al., (2009) found that the season had a significant influence only the phosphorus levels ( $P < 0.05$ ) which are lower during the winter when compared to the autumn. Apart from the hemoglobin, all other

parameters examined in the Bulgarian Rhodope Cattle breed were higher when compared to that of the Bulgarian Black and White Cattle breed. The autumn-winter dynamics of the Bulgarian Rhodope Cattle parameters was less expressed than that of the Bulgarian Black and White Cattle.

Nikolov et al. (2009) report age differences in the cholesterol levels of cows from the Bulgarian Rhodope Cattle breed- its content is higher in younger animals. During the examination high ASAT/ALAT ratio, high hemoglobin and creatinine and low cholesterol levels, which are specific for the breed, were reported.

### MATERIALS AND METHODS

The levels of major hematological parameters- glucose, total protein, triglycerides, cholesterol, creatinine, magnesium, calcium and phosphorus were examined during two critical periods- before and after the winter season. For this purpose, 36 blood samples were taken from *v. jugularis*. The samples were taken twice - in spring and autumn, from cows with an equalized physiological status kept in three different regions, there were six blood samples per region. The hematological parameters were reported through a semi-automated analyser

with tests of "BIOMED" firm. The data were statistically processed via analysis of variance with the help of a specialized software (SPSS 21, IBM).

## RESULTS AND DISCUSSIONS

The cows from the Rhodope Shorthorn Cattle breed are reared under harsh natural and climatic conditions and have a relatively poor diet. Taking the above mentioned in the account and searching for an explanation of the clearly exhibited significant seasonal differences in the reproductive ability, we examined some hematological parameters in cows before and

after the period of mass calving- in autumn and spring- in different regions of the main areal. The blood glucose level is a major interior index reflecting the balance of the carbohydrate-metabolism in the organism. In two of the regions, Smolyan and Haskovo, the cows from the Rhodope Shorthorn Cattle breed we examined exhibited values which were higher than the upper reference values (Table 1). The values were higher than that reported from Nikolov et al. (2012) with reference to the same breed in the same regions (2.40 nmol/l). The latter, however, were obtained during the winter but this study was carried out in autumn and spring.

Table 1. Serum blood sugar, total protein, cholesterol, triglycerides and creatinine content in the cows from different regions

Traits	Regions	N	LSM	± SE	SD	Min.	Max.
Blood sugar, mmol/L	Kurdzhali	12	3.288	0.181	0.937	1.940	4.920
	Smolyan	12	4.526	0.181	1.953	2.430	7.150
	Haskovo	12	4.714	0.181	2.039	2.110	7.460
Total protein, g/L	Kurdzhali	12	50.67	0.846	6.475	41.10	58.67
	Smolyan	12	55.28	0.846	3.598	49.70	62.65
	Haskovo	12	58.48	0.846	11.56	40.30	73.59
Cholesterol, mmol/L	Kurdzhali	12	2.616	0.139	0.870	1.390	4.100
	Smolyan	12	2.815	0.139	0.860	1.820	4.260
	Haskovo	12	2.640	0.139	0.549	1.910	3.650
Triglycerides, mmol/L	Kurdzhali	12	0.258	0.036	0.114	0.120	0.470
	Smolyan	12	0.595	0.036	0.227	0.190	0.920
	Haskovo	12	0.436	0.036	0.132	0.240	0.680
Creatinine, µmol/L	Kurdzhali	12	229.8	9.090	28.624	173.0	285.0
	Smolyan	12	254.1	9.090	45.231	170.1	325.0
	Haskovo	12	217.8	9.090	26.292	175.7	268.0

Note: LSM – average; SE – standard error; SD – standard deviation

Table 2 indicates that in spring, the average blood sugar levels are within reference values and are close to those typical for the winter period (Nikolov et al., 2012). Of all three seasons examined, apparent differences in the carbohydrate metabolism can be observed only during the autumn. The seasonal changes can be clearly seen in Table 3.

Actually, the autumn is an unfavourable period for the cattle in the researched areal. The air

temperature is high, the pastures are scarce with hay mostly, and the cattle is not supplementary fed by the farmers.

Shrikhande et al., (2008) have not found considerable seasonal variations in the blood glucose levels of lactating cows- they were respectively 2.46 and 2.67 nmol/L during the winter and spring period.

Table 2. Serum blood sugar, total protein, cholesterol, triglycerides and creatinine content in cows in the spring and the autumn

Traits	Season	N	LSM	± SE	SD	Min.	Max.
Blood sugar, mmol/L	Spring	18	2.712	0.148	0.453	1.940	3.600
	Autumn	18	5.640	0.148	1.353	3.390	7.460
Total protein, g/L	Spring	18	61.16	0.691	6.319	54.03	73.59
	Autumn	18	48.47	0.691	4.306	40.30	54.50
Cholesterol, mmol/L	Spring	18	2.108	0.114	0.299	1.390	2.510
	Autumn	18	3.272	0.114	0.610	2.340	4.260
Triglycerides, mmol/L	Spring	18	0.374	0.030	0.188	0.120	0.700
	Autumn	18	0.485	0.030	0.227	0.170	0.920
Creatinine, µmol/L	Spring	18	245.8	7.422	39.41	173.0	325.0
	Autumn	18	222.0	7.422	30.55	170.1	309.8

Note: N - Number of animals from each farm; LSM – average; SE – standard error; SD – standard deviation;

Table 3. Influence of the Season and the region on some hematological parameters of cows from the Rhodope Shorthorn Cattle breed

Traits	F- criteria and confidents level		
	Region	Season	Region of the season
Blood sugar, mmol/L	18.25***	195.40***	10.53***
Total protein, g/L	21.56***	168.72***	21.29***
Cholesterol, mmol/L	0.61	52.41***	1.28
Triglycerides, mmol/L	21.79***	7.08*	10.09***
Creatinine, µmol.l <sup>-1</sup>	4.15*	5.13*	2.16
P, mmol/L	4.03*	9.44**	14.94***
Ca, mmol/L	2.06	968.78***	0.96
Mg, mmol/L	42.57***	178.77***	42.77***

Note: \*\*\*P<0.001; \*\*P<0.01; \*P<0.05; F-criteria and Sig – ration of the mean sums of squares of the regression equation and the residual and significance level

Figure 1 shows that in spring, the blood sugar levels of the animals from the three regions examined are practically the same. In autumn, however, the parameters of the cows from Kurdzhali region are more than 1/3 lower than those of the animals in the other two regions; the lowest levels are exhibited by the animals from Haskovo region.

Similar regional differences (Table 3) are also observed with reference to the serum triglycerides content (Figure 1).

As a whole, in the autumn their content is 29.6 % higher than that in spring (Table 2). Nevertheless, it is clear from Figure 1 that the above mentioned parameters refer two of the researched regions - Kurdzhali and Haskovo. The blood triglycerides content of the cows reared in Haskovo region is 35.1% higher in spring than in the autumn.

With reference to the cholesterol, the season is a source of variation ( $P < 0.001$ ) while the region does not affect its values (Table 3).

According to Quiroz-Rocha et al. (2009), the total serum cholesterol concentration may be used as an index for energy balance of cows. The cholesterol is of vital importance for the proper function and the structure of the cell membranes. It is a compound of the steroid hormones, including estrogen, progesterone, testosterone and hormones related to the supra-renal function. Furthermore, the cholesterol acts as one of the organism natural barriers, protecting the cells from dehydration and infections. (Kaslow, 1997). In the light of the results we obtained, the latter is really interesting. Although they are within the established physiological references, during the less favourable autumn season, the blood cholesterol levels of the researched cows are 55.2% higher than in spring. The total protein levels are lower than the reference values for the cattle, being close to the minimum in spring. The feeding is proved to be the major factor influencing the blood protein levels. They are also affected by the condition of the liver, kidneys,

gastrointestinal tract, the stress, water loss and so on. During the less favourable autumn season, the total protein content in blood is ( $P < 0.001$ ) 20.4% lower, the decline being most considerable in Haskovo region (Figure 2)-30.5%, and least noticeable in Smolyan region-9.6%.

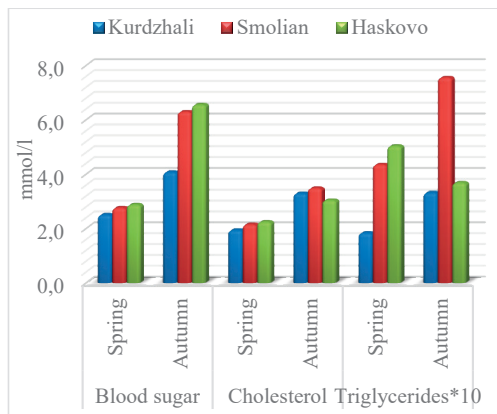


Figure 1. Seasonal dynamics of the blood sugar, cholesterol and triglycerides content in blood of cows from the Rhodope Shorthorn Cattle breed, reared in the different regions

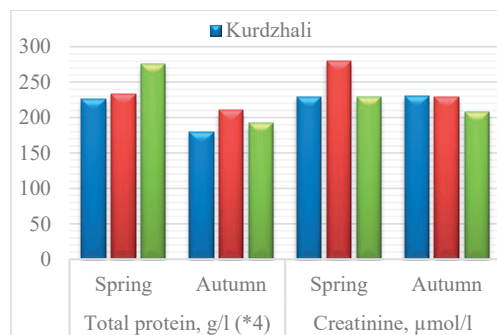


Figure 2. Seasonal dynamics of total protein and creatinine blood levels of cows from the Rhodope Shorthorn Cattle breed, reared in the different regions

Creatinine forms in muscles and usually has constant levels. In the case of the animals examined by us, the blood creatinine content during the two seasons is within close references but is significantly above the physiological norm (165 μmol/l). Even higher values were ascertained by Nikolov et al. (2012) upon examining the population during the winter period. Values higher than the upper reference levels were also reported with reference to the

Bulgarian Rhodope Cattle, originating from the Rhodope Shorthorn Cattle (Nikolov et al., 2009).

Taking into account that the creatinine is formed in the muscles and that during intensive muscle activity the macroergic phosphate residue of the creatine phosphate is transferred to ADP forming ATP, and the creatine phosphate turns into creatinine, the above mentioned authors assume that the maintaining of high creatinine levels can probably be attributed to the energetic nature of the two breeds and the noticeable physical activity during grazing. Increased creatinine content after intensive muscle activity is also observed by Sato et al. (2001).

The Rhodope Shorthorn Cattle is extremely active and dynamic animal. Therefore, we can presume that the ascertained creatinine levels are probably close to the physiological ones which are typical of the breed. The region influences ( $P < 0.5$ ) the creatinine blood content of Rhodope Shorthorn Cattle, however, within the season, the regional differences are insignificant (Table 3).

Considerable (Table 4) and significant (Table 3) are the seasonal differences in the mineral blood content of the Rhodope Shorthorn Cattle breed. Such seasonal differences in the content of mineral substances in the blood of the cows are also observed by Ivanova et al. (2009).

All examined blood mineral values are higher in autumn than in spring. This can probably be accounted to the fact that in autumn, the concentration of mineral in the hay is higher.

The difference in phosphorus levels is minimal, while the calcium levels are two times higher in the autumn. In spring, the magnesium is within the lower physiological references and in autumn, it is 2.7 times higher. Although the minerals levels after the winter are considerably lower than those during the autumn, they are still within the physiological reference which indicates well functioning homeostatic mechanisms. According to Martens and Schweigel (2001), there are significant calcium and phosphorus reserves in the bones, but it is difficult for the organism to compensate the low magnesium content due to the lack of hormonal mechanisms of homeostasis.

Its content in the blood is mainly influenced by its intake with the food. At the same time the Mg-ATP complex is vital for all biosynthesis

processes in the body (glycolysis, energy dependent membrane transport, formation of cyclic AMP and transfer of genetic code) (Djokovic et al., 2014).

Our study ascertained significant regional differences regarding magnesium content in the blood of Rhodope Shorthorn Cattle breed (Table 5).

Table 4. Content of minerals in the blood of cows in spring and autumn

Traits	Season	N	LSM	± SE	SD	Min.	Max.
P, mmol/L	Spring	18	1.523	0.047	0.174	1.280	1.820
	Autumn	18	1.726	0.047	0.357	1.090	2.280
Ca, mmol/L	Spring	18	2.241	0.041	0.239	1.910	2.770
	Autumn	18	4.061	0.041	0.089	3.890	4.250
Mg, mmol/L	Spring	18	0.628	0.056	0.067	0.500	0.740
	Autumn	18	1.695	0.056	0.820	0.560	2.870

Note: N - Number of animals form each farm; LSM – average; SE – standard error; SD – standard deviation

Table 5. Content of minerals in the blood of cows reared in different regions

Traits	Reginos	LSM	± SE	SD	Min.	Max.
P, mmol/L	Kurdzhali	1.604	0.057	0.266	1.280	2.050
	Smolyan	1.748	0.057	0.307	1.370	2.280
	Haskovo	1.521	0.057	0.289	1.090	1.880
Ca, mmol/L	Kurdzhali	3.218	0.051	0.918	2.130	4.120
	Smolyan	3.073	0.051	0.940	1.990	4.030
	Haskovo	3.163	0.051	1.036	1.910	4.250
Mg, mmol/L	Kurdzhali	1.339	0.069	0.786	0.620	2.870
	Smolyan	0.649	0.069	0.073	0.540	0.760
	Haskovo	1.497	0.069	0.954	0.500	2.810

Note: LSM – average; SE – standard error; SD – standard deviation

Magnesium activates more than 300 enzymes. When its levels are too low, the organism defense systems decrease and the possibility of infections increases. Furthermore, the growth is slowing down and productivity and sexual activity diminish (Davoudi and Fazel, 2012).

In Smolyan region, its levels are two times lower when compared to the Mg levels in Eastern Rhodopes regions. They remain low in both examined periods (Figure 3).

Cattle have physiologically lower levels of inorganic phosphorus when compared to the other domestic animals due to the fact that the latter consume feeds which are richer in Ca. The phosphorus levels of the animals we studied are the highest in Smolyan region

where the lowest levels of calcium and magnesium are reported (Table 5). The lowest

phosphorus level is reported in Haskovo region where the level of magnesium is the highest. With the exception of Mg no significant differences are observed between regions in different seasons with reference to all minerals examined (Figure 3). In magnesium, there was a general upward trend in its content in the blood during the autumn with the exception of Smolyan region where its levels were almost the same as those during the spring. The Mg content in the other two regions - Kurdzhali and Haskovo, during the autumn is 3 and 4 times higher.

The Ca content in all regions during the autumn is 75-85% higher than it is during the spring. Taking the quality of the food consumed during the two seasons into account, this is considered logical.

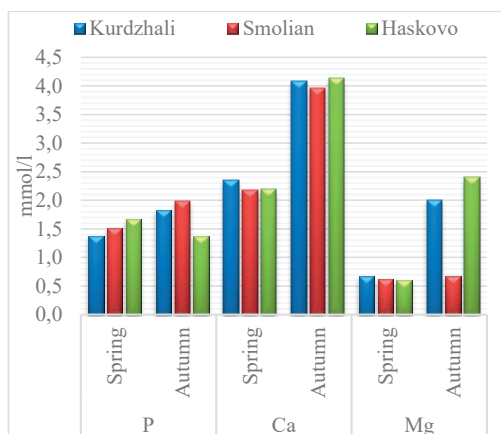


Figure 3. Seasonal dynamics of Ca, P and Mg content in the blood of cows from the Bulgarian Shorthorn Cattle breed reared in the different regions.

## CONCLUSIONS

It can be concluded that the glucose, total protein, cholesterol, triglycerides, creatinine, Ca, P and Mg content in the Rhodope Shorthorn Cattle blood is seasonally affected.

There have also been regional differences ascertained regarding all parameters except the cholesterol and Ca.

The levels of total protein are lower and those of creatinine- higher than the physiological norms in all studied seasons and regions.

The blood sugar content is considerably above the reference values, the cholesterol and triglycerides levels are higher, and those of creatinine and total protein are lower in autumn. In spring, the levels of minerals in the blood are lower than in the autumn; all of them are under the physiological norms.

As a whole, the mineral content is lower in Western Rhodopes, and more significant changes in the carbohydrate and protein metabolism are indicated in Haskovo region during the autumn.

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