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SERUM CHEMISTRY PARAMETERS OF FREE-RANGING MOOSE (Alces alces Linnaeus, 1758) OF DIFFERENT SEX AND AGE GROUPS

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Abstract

Moose (Alces alces, Linnaeus 1758) is a perspective species for game farming, characterized by a high growth rate, as well as a wide potential for economic use for meat and milk production. In Russia and the world, there are few studies of the biochemical parameters of moose blood. (S.A. Becker et al., 2010; A.W. Franzmann et al., 1978; M.A. Keech et al., 1998; V. Reshetnyak et al., 2021; M.K. Rostal et al., 2012). The reference range of biochemical blood parameters is necessary for assessing the physical condition of animals, nutrition quality, habitat, stressors, the impact of infectious and invasive diseases (A.A. Cunningham et al., 1998; P. Daszak et al., 1999; W.F. Frick et al., 2010). The article presents for the first time the biochemical parameters of the blood of moose from different sex and age groups. The goal of the work is to determine the biochemical parameters of blood in moose of different sex and age living in the Kirov region. Whole blood was taken from animals (n = 90) bagged during the hunting seasons of 2006-2020. Samples were collected from October to December in the experimental hunting ground of Zhitkov Russian Game Management and Fur Farming Research Institute (Kirov region). All animals were wild and moved freely, feeding on local vegetation. Whole blood was studied from the following sex and age groups: 20 young females aged 6-7 months; 20 adult females aged 2.5-7 years; 20 young males aged 6-7 months; 30 adult males aged 2.5-7 years. All animals were considered clinically healthy. Blood samples were taken by cutting the jugular vein immediately after the animal was shot. Blood serum studies were performed on a Biochem SA semi-automatic biochemical analyzer (High Technology Inc., USA). The analysis included the determination of the activity of aspartate aminotransferase (AsAT), alanine aminotransferase (AlAT), alkaline phosphatase, lactate dehydrogenase (LDH), alpha-amylase, total protein, albumin, total bilirubin, direct bilirubin, creatinine, urea, cholesterol, glucose. In the blood serum of adult moose, high activity of AsAT was noted (in females and males, respectively, 253.8±52.38 and 250.9±47.52 U/l), alphaamylase $(29.2\pm7.20 \text{ and } 32 \text{ } 0\pm8.77 \text{ U/l})$, creatinine content $(183.9\pm18.59 \text{ and } 182.1\pm23.66 \text{ mmol/l})$, urea (6.2 ± 0.82 mmol/l and 5.9 ± 0.87 mmol/l). In young individuals, AsAT (161.2 ± 28.30 and 160.0±30.92 U/l), alpha-amylase (24.6±4.91 and 23.2±5.46 U/l), the content creatinine (152.8±20.32 and 149.1 ± 23.78 mmol/l), urea (2.4 ± 0.63 and 2.6 ± 0.98 mmol/l) were significantly lower. In young animals, on the contrary, high activity of AlAT (60.8 ± 6.42 and 58.7 ± 6.74 U/l), alkaline phosphatase (230.4±40.79 and 222.2±31.14 U/l) /l), LDH (805.2±185.57 and 822.9±237.13 U/l), glucose content $(6.3\pm1.01 \text{ and } 6.3\pm1.03 \text{ mmol/l})$. Adult animals were characterized by lower levels of AlAT (43.6 \pm 7.35 and 41.9±6.33 U/l), alkaline phosphatase (69.3±12.62 U/l and 69.9±11,31 U/l), LDH (614.1±98.11 U/l and 598.2 \pm 129.37 U/l), glucose (5.3 \pm 1.02 and 5.3 \pm 1.14 mmol/ l). Statistically significant (p < 0.05) differences in the content of total protein in blood serum between adult females $(57.4\pm7.48 \text{ g/l})$ and males (68.1 ± 4.93) were established. In young females, an average correlation was found between the

content of total and direct bilirubin (r = 0.57, p = 0.01), in adult females — between the activity of AsAT and alkaline phosphatase (r = 0.50, p = 0.02) and between AsAT and LDH activity (r = 0.66, p = 0.00). A statistically significant (p < 0.05) effect of age on the content of AsAT, AlAT, alkaline phosphatase, LDH, alpha-amylase, direct bilirubin, creatinine, urea and glucose, as well as the sex of animals on the content of total protein was shown. The dependence of the AsAT, AlAT, alkaline phosphatase, LDH, alpha-amylase, total protein, direct bilirubin, creatinine, urea and glucose levels on body weight was revealed. The obtained biochemical parameters of moose blood have a similar trend in most parameters with the results obtained on other artiodactyls in the wild. Differences are due to species of animals, living conditions, nutrition, age, sex, as well as the method used in blood sampling.

Keywords: moose, females, males, age groups, biochemical blood parameters, blood serum

Farm hunting has emerged in the last decade as a separate area, which involves keeping animals in semi-free conditions and in an artificial habitat, which requires hunting and veterinary measures. In this regard, elk (*Alces alces* Linnaeus, 1758) is a promising species characterized by a high growth rate and wide possibilities for economic use for the production of dietary meat and medicinal milk [1].

The biochemical factor influences the adaptability of the animal body to environmental conditions. Changes in enzymatic reactions make it possible to successfully cope with unfavorable factors or their fluctuations. The biochemical features identified in various organisms became understandable and accessible to study only against the backdrop of modern advances in biochemistry. It is biochemical adaptation that allows species living in different natural conditions to maintain external similarities [2].

Basic values of biochemical blood parameters are necessary to assess the physical condition (function of internal organs, metabolism) and health of animals both at the population and individual levels, the quality of nutrition and habitat, the impact of anthropogenic and environmental stressors, the potential impact of existing and emerging diseases infectious and invasive nature, as well as to determine appropriate strategies to combat them [3-7]. In addition, biochemical blood parameters make it possible to assess the metabolic patterns of elk of different ages depending on environmental conditions [8].

Even in a healthy animal in relatively good body condition, nutrient deficiencies can create physiological imbalances that affect population performance [9, 10]. It has been proven that nutrient limitation and deficiency for moose, especially in winter, leads to decreased reproductive function and survival, especially among young animals [11-14].

When developing a reference range, it is important to consider the presence of diseases in the animals being studied, which may affect the results of the study, especially in individuals with infectious or invasive diseases, iron deficiency anemia, some types of cancer, and other systemic diseases [15-17]. Various conditions of the body will also affect the biochemical parameters of the blood. For example, in emaciated ruminants, there is a decrease in albumin, lactate dehydrogenase (LDH), gamma-glutamine transferase (GGT) and creatinine phosphokinase (CPK), an increase in creatinine and urea (due to muscle catabolism), as well as a change in the amount of beta-hydrobutyric acid. Obesity causes increased glucose and triglyceride levels [18].

Although sampling from animals in the wild is difficult and sometimes virtually impossible, basic blood chemistry ranges have been established for some wild species [19].

Studies of biochemical parameters in moose in Russia and the world are rare [20-25]. The values of chemical indicators were derived from a small number of samples with no age-sex differences, obtained by different methods from hunted, caught, immobilized individuals or from animals kept in captivity, and therefore cannot be fully representative, but nevertheless represent scientific value, and similar studies should be continued.

In this work, for the first time, biochemical blood parameters of moose from different sex and age groups were established.

The purpose of the work is to study biochemical blood parameters in moose of different sexes and ages living in the Kirov region.

Materials and methods. Whole blood was taken from animals (n = 90) hunted during the 2006-2020 hunting seasons. Sampling was carried out from October to December in the scientific experimental hunting farm of the Professor Zhitkov All-Russian Research Institute of Hunting and Fur Farming (Kirov Province), located in the northern part of the moose habitat (northeast of the European part of Russia; 58°33'04"N, 50°43'42"E). The total area of the farm is 66,250 hectares. The climate is continental with moderately cold winters and warm summers. All animals were wild and moved freely, feeding on local vegetation.

Whole blood was studied in 20 young females aged 6-7 months; 20 adult females aged 2.5-7 years; 20 young males aged 6-7 months; 30 adult males aged 2.5-7 years. All animals were considered clinically healthy (at the time of sampling there were no signs of disease; the health of the animals was assessed by a veterinarian who is part of the hunting team). Body weight was 127.0-185.0 kg (158.6 \pm 23.97 kg) in young females, 363.0-432.0 kg (391.8 \pm 29.04 kg) in adult females, 178.0 -201.5 kg (191.2 \pm 10.58 kg) in young males, and 280.0-420.5 (340.9 \pm 53.36 kg) in adult males.

Blood for the study was taken from the jugular vein (*venae jugularis*), which was cut immediately after the animal was shot; no medications were used. The death of an animal from a gunshot wound occurred at lightning speed, or in most cases the agonal period did not exceed several minutes (this corresponded to the lightning-fast rate of dying, in which the agonal period is no more than 15-30 minutes). Blood was collected into 4 ml UNIVAC vacuum tubes (Eiliton, Russia) with a coagulation activator. Before sending to the laboratory (16-24 hours), the blood was stored in a refrigerator at 4 $^{\circ}$ C. Hemolyzed samples were discarded to avoid analytical errors.

In the laboratory, the blood was centrifuged at 1500 rpm for 20 minutes (Liston C 2204 centrifuge, Liston, Russia). Blood serum studies were carried out immediately after delivery (a semi-automatic biochemical analyzer Biochem SA, High Technology, Inc., USA) with a set of reagents (Eco-Service, Russia) to determine the activity of aspartate aminotransferase (AsAT), alanine aminotransferase (AlAT), alkaline phosphatase, lactate dehydrogenase (LDH), alpha-amylase, total protein content, albumin, total bilirubin, direct bilirubin, creatinine, urea, cholesterol, glucose.

Statistical analysis was performed using Microsoft Excel 2019 and Statgraphics (19-X64) software using standard methods [26]. To describe the samples, means (*M*), standard deviations (\pm SD), medians (*Me*), 25% and 75% percentiles were determined. When comparing indicators between groups, the nonparametric Wilcoxon-Mann-Whitney test (U) was used. Relationships between characteristics were assessed using Spearman rank correlation. To assess the influence of three factors (age, sex, body weight) on biochemical blood parameters, single- and multivariate analysis of variance (ANOVA, MANOVA) was used. The influence of the factor was considered statistically significant at p < 0.05.

Results. The results of biochemical studies of blood serum of moose of various sex and age groups are given in the Table.

In young females, an average correlation was established between the content of total and direct bilirubin (r = 0.57, p = 0.01), in adult females between the activity of AsAT and alkaline phosphatase (r = 0.50, p = 0.02) and between AsAT and LDH activity (r = 0.66, p = 0.00).

Parameter	Youngsters up to 1 year old, \bigcirc	Adults, \bigcirc	Youngsters up to 1 year olf,, 3	Adults, δ
	(n = 20)	(n - 20)	(n = 20)	(n - 30)
Aspartate aminotransferase, U/l:	117 5 210 1	157 1 241 6	120 2 207 5	162 3 242 5
M+SD	161 2+28 30	253 8+52 38	120.2-207.3 160.0+30.92	250 9+47 5
Me	155.6 ^a	244.1ª	154.7 ^a	243.3ª
25 %-75 %	146.9-178.9	219.6-296.0	131.6-193.5	208.4-299.4
Alanine aminotransferase, U/l:				
Min-max	47.2-72.0	29.4-55.4	48.0-71.2	30.4-53.2
M±SD Ma	60.8±6.42	$43.6\pm /.35$	58./±6./4	41.9±6.33
25 %-75 %	56 4-63 9	39 1-49 7	54 8-64 7	38 3-46 9
Alkaline phosphatase, U/l:	0011 0019	0,111,010	0110 0117	0010 1019
Min-max	165.2-296.0	49.0-89.1	169.2-270.8	46.6-88.3
M±SD	230.4 ± 40.79	69.3±12.62	222.2±31.14	69.9±11.31
Me	232.0a	69.0 ^a	219.1 ^a	70.4 ^a
25 %-75 % Lactate dehydrogenase U/I:	19/./-269.2	57.5-79.3	200.9-253.8	61.4-79.2
Min-max	502 9-1118 6	417 0-803 0	504 9-1222 0	380 5-864 1
M±SD	805.2±185.57	614.9±98.11	822.9±237.13	598.2±129.37
Ме	847.5 ^a	622.9 ^a	778.2 ^a	594.4 ^a
25 %-75 %	700.4-926.2	558.3-672.1	659.3-1062.7	515.3-699.6
Alpha amylase, U/l:	160.050	17 1 41 4	15.2.246	10.0.47.6
Min-max M+SD	16.9-35.9	17.1-41.6	15.3-34.6	18.9-47.6
M±SD Ma	24.0±4.91 24.30	29.2±7.20 27.3c	23.2 ± 5.40 23.4a	32.0 ± 8.77 32.2a
25 %-75 %	21 3-27 0	27.3-	19 4-25 9	31 3-35 1
Total protein, g/l:	21.5 27.0	21.7 33.2	19.1 25.9	51.5 55.1
Min-max	46.5-83.0	45.4-70.1	45.5-86.1	59.4-79.4
M±SD	61.6±9.91	57.4±7.48	66.4±13.53	68.1±4.93
Me	62.0	57.8d	70.5	68.7d
25 %-75 %	53.75-67.0	52.4-62.0	53.2-77.8	63.4-71.4
Albumen, g/l:	33.0.40.1	30 0 53 8	32 2 53 3	33 4 53 2
M+SD	40 9+4 48	43 7+5 62	42.9+6.94	42 3+5 65
Me	41.8	44.9	42.9	41.5
25 %-75 %	38.2-43.7	40.2-46.5	36.0-50.0	38.8-46.6
Total bilirubin, mmol/l:				
Min-max	5.6-10.9	6.7-10.7	5.6-10.1	6.5-11.5
M±SD	7.9±1.54	8.6 ± 1.02	8.4±1.11	9.1±1.43
Me 25 %_75 %	8.0 6 3-9 1	8.5 81_93	8.4 7.6-9.3	9.2 8 3-10 1
Direct bilirubin, mmol/l:	0.5-7.1	0.1-9.5	7.0-7.5	0.5-10.1
Min-max	1.45-3.7	1.6-3.7	0.9-3.8	1.3-4.0
M±SD	2.6 ± 0.60	2.6 ± 0.55	2.4 ± 0.90	2.7±0.79
Me	2.6	2.7	2.4	2.8
25 %-75 %	2.3-3.1	2.2-2.9	1.8-2.8	2.1-3.3
Creatinine, mmol/l:	117 7 190 7	156 4 225 7	100 2 190 4	126 2 222 2
MHI-HAX M+SD	152 8+20 32	183 9+18 59	149 1+23 78	182 1+23 66
M±SD Me	152.0 <u>+</u> 20.52	185.7°	142.95	181.2 ^c
25 %-75 %	139.8-169.5	169.6-195.3	133.3-173.0	167.5-200.1
Urea, mmol/l:				
Min-max	1.3-3.7	4.8-7.4	1.0-4.0	4.2-7.3
M±SD	2.4 ± 0.63	6.2 ± 0.82	2.6 ± 0.98	5.9 ± 0.87
ме 25 %_75 %	2.20	0.4° 5.5.7.0	2.20	0.0° 5.3_6.7
Cholesterol mmol/l	2.0-2.0	5.5-7.0	1.7-3.0	5.5-0.7
Min-max	0.5-0.8	0.2-0.9	0.3-0.7	0.2-0.8
<i>M</i> ±SD	0.6±0.09	0.5±0.18	0.5±0.10	0.5±0.14
Me	0.6	0.5	0.5	0.5
25 %-75 %	0.5-0.7	0.4-0.7	0.4-0.6	0.4-0.6
Glucose, mmol/l:	1276	2672	4077	22.70
M+SD	4.2-7.0 6.2+1.01	3.0-/.2 5.3+1.02	4.9-7.7	5.5-7.9 5.3+1.14
Me Me	6.2±1.01	5.5 <u>+</u> 1.02	5.9 ^b	5.5±1.14 5 3b
25 %-75 %	5.7-7.2	4.5-6.0	5.5-7.5	4.8-6.0
a, b, c Dfferences between young an	imals and adults are sta	tistically significant	nt, at $p = 0.000$, $p =$	0.003, p = 0.03,
respectively; d differences between fe	emales and males are stat	tistically significan	t at $p = 0.000$.	,

Blood biochemical parameters in moose (*Alces alces* Linnaeus, 1758) of different sex and age groups (Kirov Province, 2006-2020)

It should be noted that biochemical blood parameters can be influenced by

various factors. The conducted single- and multifactorial analysis (ANOVA, MANOVA) made it possible to establish the influence of physiological factors (age, gender, weight). According to the results of MANOVA testing, a significant effect of age on the activity of AsAT (p = 0.00), AlAT (p = 0.00), alkaline phosphatase (p = 0.00), LDH (p = 0.00), alpha amylase (p = 0.00), direct bilirubin (p = 0.01), creatinine (p = 0.00), urea (p = 0.00) and glucose (p = 0.00). A statistically significant effect of gender on the amount of total protein was proven (p = 0.00).

The individuals we studied from different sex and age groups differed significantly in body weight. Using ANOVA, the dependence of the following biochemical parameters on body weight was established: activity of AsAT (p = 0.00), AlAT (p = 0.00), alkaline phosphatase (p = 0.00), LDH (p = 0.00), alpha-amylase (p = 0.00), total protein (p = 0.00), direct bilirubin (p = 0.01), creatinine (p = 0.00), urea (p = 0.00) and glucose (p = 0.00).

Due to the few publications of information on the biochemical parameters of the blood of moose and the lack of sex and age distinctions in them, we considered it possible to compare our data with those obtained for other species of the suborder *Ruminantia*. When compared with the results of foreign and domestic researchers, some differences were identified, but the trend for adult animals and young animals under 1 year of age was similar in most indicators.

AsAT and AlAT play an important role in amino acid metabolism, and their greatest activity is observed in skeletal muscle, myocardium and liver. Our studies established significant age-related differences in the content of AsAT, AlAT, alkaline phosphatase, and LDH.

M.K. Rostal et al. [25] who studied blood biochemical parameters in moose in Norway, described a trend in blood biochemistry similar to our results, but in their studies, the animals were not differentiated by sex. Thus, AsAT activity in young animals and adults averaged 130.0 U/l, which is lower than our values by 16.2 and 46.7%, respectively. The AIAT content in young animals and adults was also lower by 44.5% (33.0 U/l) and 36.3% (27.0 U/l). From the work of S.A. Becker et al. [21] who studied blood parameters in adult female Shirasi elk (*A. a. Shirasi* Nelson, 1914) in the northwestern state of Wyoming (USA), it is known that the AsAT content in adult females was lower than the values in our study by 57.5% (103.7 U/l). A.L. Miller et al. [27] studied the biochemical composition of the blood serum of free-ranging adult Norwegian wild reindeer (*Rangifer tarandus* Linnaeus, 1758) in southwestern Norway. Their results are also presented without taking into account the sex of the animals. The concentrations of AsAT and AIAT were lower than in our study by 60.7 (96.0 U/L) and 23.1% (34.0 U/L), respectively.

For the differential diagnosis of various pathologies, the de Ritis coefficient is of great importance. Having calculated the ratio of the activity of AsAT and AlAT enzymes, we found that it increased unidirectionally with the age of the animals. In young animals under 1 year of age, this indicator was at the upper limit of the norm for domestic cows (*Bos taurus taurus* Linnaeus, 1758) [28] (2.5 for females, 2.7 for males). In adult animals, it was 5.5 and 6.0, respectively, which can be explained by an increase in the metabolic load on skeletal muscles and myocardium when pursuing animals during hunting.

A.E. Weber et al. [29] found that in the blood serum of moose, the activity of the transamination enzymes AsAT and AlAT varies with the seasons. It was maximum in spring, minimum in autumn. The spring rise is due to increased intake and utilization of ammonia, as well as high activity of protein metabolism. In autumn, the intensity of transamination decreases significantly which is apparently due to the attenuation of synthetic processes in the body. In addition, according to M. Koseoglu et al. [30], hemolysis causes a significant increase in AsaT levels. Obviously, the parameters must be interpreted taking this factor into account. The total activity of alkaline phosphatase in the circulating blood of healthy animals consists of the activity of liver and bone isoenzymes, which is greatest in growing animals [31]. Alkaline phosphatase is involved in the formation of the skeleton during ontogenetic development. Our data are consistent with these statements. The values in young and adult moose from Norway exceeded our values of alkaline phosphatase activity by 20.0% [25], and in adult females of Shiras elk [21] and Norwegian reindeer [27] by more than 70.0%. According to E.V. Gromyko [32], in cows the physiological norm for alkaline phosphatase levels is 55.0-80.0 U/l.

It should be noted that the development of the skeleton and musculoskeletal system is of utmost importance in the formation of the respiratory function of the blood in onto- and phylogenesis [33]. In newborn mammals, the entire red bone marrow is active, and it is in it that the hematopoies occurs, while in adult animals a certain part of it is replaced by yellow adipose bone marrow [34].

According to researchers studying the physiology of moose in the Pechora-Ilychsky Nature Reserve (Komi Republic), newborn moose calves grow very quickly. A comparison of the growth rate of elk calves and young cattle shows that the ratio of body weight gain to body weight in the 1st month of life in moose calves is 50.8 [35], in calves 31.3 [36]. According to A.E. Knorre [37], the relative increase in body weight of elk calves is 2 times higher than that of calves. That is, doubling body weight occurs faster in those species whose milk contains a higher concentration of protein and ash elements (calcium, phosphorus), necessary for the formation of the skeleton and muscles. Among ungulates, these are elk and reindeer [38]. Relative growth rate peaks at 3 months of age. In the fall, when switching to twig food, the growth of moose calves slows down sharply. The growth rate during the first half of life in elk calves reaches 1500%, and in cattle 400-407.0% [39]. High growth rates and active metabolism, the formation of physiological maturity of organs and systems of elk calves in the summer determine the success of their survival in winter, when nutrition is limited to twig-poor protein food, and serve as evidence of the adaptive plasticity of the elk body. In addition, the rapid development of the musculoskeletal system is important for more efficient respiratory function of the blood.

Lactate dehydrogenase is an enzyme that catalyzes the reversible conversion of lactate to pyruvate during glycolysis. High LDH activity is inherent in many tissues, primarily the liver, skeletal muscles, myocardium, as well as lung tissue, kidneys, pancreas and stomach. A.W. Franzmann et al. [22] showed that LDH content is significantly higher in young animals. The same trend was observed in our studies, as well as in other works [21, 25]. In addition, significant changes in LDH content were recorded in bovine blood during storage. Thus, this indicator in the blood serum of cattle increases after 24 hours of storage in the refrigerator [25]. Studies of human blood have shown a similar increase in the amount of LDH in refrigerated sera stored for 7 days [40]. In these studies, other biochemical blood parameters did not change significantly.

Alpha-amylase is a hydrolytic enzyme that breaks down complex carbohydrates into maltose and glucose. In our studies, alpha-amylase activity increased statistically significantly (p < 0.05) with the age of the animals. In adult moose, it turned out to be higher by 11.1% in females and 27.3% in males than in young animals, which indicates a more intense carbohydrate metabolism in adults compared to young animals. In the studies of M.K. Rostal et al. [25] such a trend was not identified; the concentration of alpha-amylase in young animals and adults did not have statistically significant differences with the indicators in adult moose from the Kirov region. The activity of alpha-amylase in Norwegian reindeer exceeded our values by 33.8% (45.0 U/l).

The correspondence of protein nutrition to the biological needs of the body

is assessed by the concentration of total protein and its fractions in the blood serum. The significantly higher content of total protein in the blood of adult males (p < 0.05) that we established was apparently associated with stimulation of its synthesis by the male steroid hormone testosterone. In addition, a high concentration of albumin in the blood indicates the activation of the processes of creating energy and plastic reserves of the body in the summer and autumn periods [41]. Differences in diet and sampling season may also have been reflected in the total protein values in our experiments (32.0% lower) compared to those reported by S.A. Becker et al. [21]. In the autumn and winter periods, the concentration of total protein in the blood serum decreases, which indicates a lower supply of nitrogen to the body when feeding on twig food than in summer [29]. In addition, the causes of hypoproteinonemia may be protein starvation, poor absorption of proteins from food, as well as the mobilization of proteins as energy sources [42]. In Norway moose [25] and reindeer [27], the concentration of total protein in young animals and adults was practically no different from our results. According to E.V. Gromyko [32], the physiological norm for total protein content in cows was 70.0-80.0 g/l, which is probably due to the inclusion of additional protein in the animals' diet through feed additives.

The pigment bilirubin is the end product of the breakdown of hemoglobin. The total bilirubin indicator includes the total content of direct and indirect bilirubin. Direct bilirubin is indirect bilirubin processed by the liver, which is subsequently excreted from the body of animals with bile. In our studies, the content of total and direct bilirubin did not have statistically significant differences and differed by more than 60.0% from the concentration of total bilirubin in the works of other researchers [25]. These differences may indicate the extent of hemoglobin breakdown processes in the animal body.

Creatinine is an end product of metabolism that diffuses into the bloodstream and is then freely filtered by the glomeruli of the kidneys. We noted changes in the amount of creatinine depending on the age of the animals. A decrease in creatinine levels was found in young animals because they had less muscle mass compared to older animals [43]. Creatinine concentrations in our studies were comparable to values obtained in moose [25] and reindeer [27] from Norway.

Urea is the main end product of protein metabolism, synthesized by the liver from amino acids in the Krebs cycle with the participation of enzyme systems. In young animals, due to increased protein synthesis, the amount of urea is slightly reduced compared to the norm for adults [44]. Differences in urea concentrations between adult animals and young animals may also be explained by differences in diet, since calves and adult animals eat different plants. Therefore, reducing protein intake may lead to lower urea levels [25]. Nutritional deficiencies and fasting are important factors affecting blood urea levels, particularly as reported by A.L. Miller et al. [27], low rates were observed in semi-domesticated reindeer under poor nutritional conditions.

Comparison with the results of other studies showed that the concentration of urea in the blood serum of adult moose from the Kirov region is 40% higher than that of moose from Norway [25], and 26.8% higher than that of reindeer [27]. In cows, the physiological norm for urea content has been established to be 3.0-5.6 mmol/l [32]. According to A.E. Weber et al. [29], a high urea content in the blood of moose indicates the intensity of protein metabolism; in contrast to domestic cows, in the fall the amount of urea in moose does not decrease, which is associated with increased rumen-hepatic circulation of nitrogen.

In general, it is believed that an increase in the amount of protein in the blood and a decrease in urea indicate an improvement in nitrogen metabolism. The deterioration of nitrogen metabolism is accompanied by an increase in urea content and a decrease in the concentration of total protein [29].

Cholesterol is an amphipathic lipid synthesized by all cells of the body, but the bulk of production occurs in liver cells and is excreted in bile. In all age and sex groups of moose we studied, we did not establish statistically significant differences in the intensity of lipid metabolism. M.K. Rostal et al. [25] came to the same conclusions, but in their studies, as in the work of A.L. Miller et al. [27], the concentration of cholesterol in the blood averaged 1.3 and 1.5 mmol/l, respectively.

Glucose is the leading diagnostic indicator of the state of carbohydrate metabolism. The concentration of glucose in the blood is a derivative of the activity of the processes of glycogenesis, glycogenolysis, gluconeogenesis and glycolysis. It should be noted that the glucose content in the blood serum of elk in our work was 2.5 times higher than that of domestic cows [32]. According to A.E. Weber et al. [29], the peculiarity of carbohydrate-energy metabolism in moose compared to domestic ruminants is the high concentration of glucose in the blood and the fact that this indicator varies from month to month. So, in May-June it is 4.6 mmol/l, in July it reaches 5.3 mmol/l, and by October, due to the lower intensity of metabolic processes, it becomes a third lower, 3.4 mmol/l. The high blood glucose levels in moose are probably due to the increased role of the glycolytic energy supply system of these animals. In winter, this is associated with a high energy requirement against the background of slow aerobic oxidation, and in summer, with biochemical adaptations of muscles to rapid transitions from rest to movement. As a result, carbohydrate metabolism in moose is characterized by a high intensity of gluconeogenesis not only due to nutritional glucogenic compounds, but also due to hormonal regulation.

It has also been established that the heat source during thermogenesis activated by thyroid hormones is the terminal energy-rich ATP bond. The cleavage of this bond is catalyzed by the Na^+K^+ -ATPase system of membranes of calorigenic tissues - skeletal muscles, liver and kidneys. For example, after the administration of thyroid hormones, the activity of the ATPase system increases and, along with this, heat production in the body increases [2].

It is known that the concentration of glucose in the blood increases in response to sympathetic stimulation. The sympathetic nervous system constricts blood vessels and increases blood pressure, thereby diverting blood from organs whose functions in a stressful situation are not necessary for the survival of the body, and, on the contrary, increasing blood flow to the vital skeletal muscles necessary during stress. Also, the increased serum glucose values in our work and in adult female and male Norwegian wild reindeer [27] may be due to the hyperglycemic effects of catecholamines and glucocorticoids released when pursuing animals during a hunt. Other researchers have come to similar conclusions [45-47]. In addition, the high glucose content of moose from the Kirov region and reindeer from Norway, compared with domestic ruminants, facilitates survival in harsh winter conditions [19, 27, 48].

According to A.Yu. Kovtunenko [49], when domestic cows were exposed to negative temperatures of 20 °C, significant changes in biochemical blood parameters were noted. In particular, the glucose content increased by 36.2%, bilirubin by 76.7%, AIAT activity by 79.3%, AsAT by 221.0% compared to the control.

The harvest method of wild animals determines changes in serum biochemical parameters and should be taken into account when assessing or comparing serum composition between animals from different groups or between different studies [19]. In our case, the death of the animal from a gunshot wound occurred at lightning speed or the agonal period did not exceed several minutes. A number of authors [50-53] report that in this case, pulmonary and cerebral edema is absent or slightly expressed. Congestion of the capillary bed of the internal organs and hemorrhages in the tissue without reactive changes were noted. There are completely no signs of disseminated intravascular coagulation and manifestations of respiratory distress syndrome. Signs of shock changes in hemodynamics are not recorded. The specialized closing arteries of the lungs and brain are spasmed. E.K. Perry et al. [54] indicate that with an increase in the rate of death, a significant decrease in the activity of glutamate decarboxylase, phosphofructokinase, and pH is observed. At the same time, the content of phenylalanine, lysine, leucine and tryptophan increases in brain tissue.

It should be noted that medical workers and expert tanologists mainly use clinical medicine data, which are adapted to forensic practice [55-57]. "Normal" values of laboratory parameters are determined in the process of clinical trials based on the results of measurements of the test analyte in a large population of healthy individuals or other biological objects, selected and grouped by age, sex, biological and other indicators. The obtained data lead to an average value, taking into account statistically possible standard deviations and obtaining a range of values in which the reference values are located. The reference interval gives an idea of the lower and upper limits of the norm of the indicator. We also recommend following this framework to evaluate your results.

Differences in serum biochemical parameters between physically and chemically captured animals have been documented in several species of wild ungulates, including red deer (*Cervus elaphus* Linnaeus, 1758) [58] and white-tailed deer (*Odocoileus virginianus* Zimmermann, 1780) [59]. According to M.K. Rostal et al. [25], when moose were immobilized using etorphine from a helicopter, the average AsAT concentration in the blood serum was 130.0 U/L in young and adult animals, and AIAT was 27.0 and 33.0 U/L, respectively. In the work of S.A. Becker et al. [21] adult female Shiras elk were immobilized using the drugs thiafentanil or carfentanil. At the same time, AsAT activity in their blood averaged 103.7 U/l. A.L. Miller et al. [27] who also used helicopter immobilization with a combination of medetomidine and ketamine to collect blood from adult female and male Norwegian reindeer, obtained an average AsAT value of 104.0 U/L.

I. Marco et al. [58] and M.J. Fettman et al. [16] reported significant increases in serum AlAT, total protein, albumin, sodium, and chloride in physically captured red deer compared with values in chemically immobilized animals. J.M. Arnemo et al. [45] also indicate that AsAT activity and glucose levels increase under stress and exercise, caused, as in our case, by hunting pursuit. A.L. Miller et al. [27] found that stress-induced reindeer had increased values of AAT, glucose, alkaline phosphatase, and urea.

Although we provide data on the serum biochemistry of moose living in the Kirov region, there are some limiting factors that should be taken into account when conducting comparative studies. These are primarily differences in blood collection methods, biochemical analyzers, laboratory diagnostic methods and animal habitats. Obviously, parameter ranges must be interpreted in light of the factors given.

Many issues have not received sufficient discussion in the presented article due to their poor study. Research on moose metabolism will be continued and expanded, and will also be compared with data on other ungulates. We believe that the biochemical parameters of blood can be analyzed in connection with environmental and anthropogenic factors (environment, diet, nutrition, lifestyle, physical activity, etc.).

Thus, it is now important to establish baseline values for significant blood biochemical parameters in wild animals, particularly moose. This will provide veterinarians and game biologists with reference data that can be used to assess population status, as markers of various pathologies and potential reproductive efficiency, and ultimately to assess habitat quality. In the blood serum of adult moose living in the Kirov region, we found high activity of aspartate aminotransferase (in females and males 253.8±52.38 and 250.9±47.52 U/l, respectively), alpha-amylase $(29.2\pm7.20 \text{ and } 32.0\pm8.77 \text{ U/l})$, high levels of creatinine $(183.9\pm18.59 \text{ and }$ 182.1±23.66 mmol/l), urea (6.2±0.82 mmol/l and 5.9±0.87 mmol/l). In young individuals, indicators for AsAT (161.2 ± 28.30 and 160.0 ± 30.92 U/l), alpha-amylase $(24.6\pm4.91 \text{ and } 23.2\pm5.46 \text{ U/l})$, creatinine $(152.8\pm20.32 \text{ and } 149.1\pm23.78 \text{ mmol/l})$, urea $(2.4\pm0.63 \text{ and } 2.6\pm0.98 \text{ mmol/l})$ levels were significantly lower. In young animals, on the contrary, high activity of AIAT (60.8±6.42 and 58.7±6.74 U/l), alkaline phosphatase (230.4±40.79 and 222.2±31.14 U/l), lactate dehydrogenase (805.2±185.57 and 822.9 ± 237.13 U/l), high glucose content (6.3±1.01 and 6.3±1.03 mmol/l) were noted. Adult animals were characterized by a decrease in the activity of alanine aminotransferase (43.6 ± 7.35 and 41.9 ± 6.33 U/l), alkaline phosphatase (69.3 ± 12.62 and 69.9 ± 11.31 U/l). 1), LDH (614.1 ± 98.11 U/l and 598.2 ± 129.37 U/l), glucose content $(5.3\pm1.02 \text{ and } 5.3\pm1.14 \text{ mmol/l})$. Statistically significant differences in the content of total protein in blood serum were found between adult females $(57.4\pm7.48 \text{ g/l})$ and males (68.1 ± 4.93). A significant effect of the age of animals on the content of AsAT, AlAT, alkaline phosphatase, LDH, alpha-amylase, direct bilirubin, creatinine, urea and glucose was shown. A significant influence of the sex of animals on the content of total protein has been established. The dependence of the activity of AsAT, AlAT, alkaline phosphatase, LDH, alpha-amylase, total protein content, direct bilirubin, creatinine, urea and glucose on body weight was revealed. The biochemical values presented in the work have a similar trend for most indicators with the results obtained on other artiodactyls in the wild. The differences are due to the type of animal, living conditions, nutrition, age, gender, as well as the method used to collect blood. Further comparative studies, inclusion of other age categories, and studies of blood chemistry in more species will expand our knowledge of the biology of wild animals. The results obtained can be useful in monitoring the environment, assessing the influence of anthropogenic factors on the parameters of the blood system and the condition of the body as a whole.

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