Seasonal changes of some hematological parameters of *Ovis aries* L. reared in ecologically extreme conditions (Karnabchul steppe, Uzbekistan)

Baxritdin Bazarov[1*], *Feruza* Normuhammedova[1], *Oysora* Tojikulova[1], *Mars* Safin[1], *Mirzoxid* Kuziev[1], and *Khayratdin* Seytkamalov[2]

[1]Samarkand State University named after Sharof Rashidov, Samarkand, 140104, Uzbekistan
[2]Nukus branch of the Tashkent University of Information Technologies named after Muhammad al-Khorazmi, Nukus, 230100, Uzbekistan

Abstract. The work describes the results of research aimed at determining the effect of the change of seasons on some blood parameters of the organism of the Korakul breed of *Ovis aries* L., which are kept in extreme conditions in the desert region of Uzbekistan. Morphological indicators and leukogram of sheep's blood were determined in the experiments according to the seasons. The amount of proteins in the sheep's blood was determined, and the seasons in which proteins were kept at a normal level were determined. It has been shown that in extreme conditions, immune parameters in the blood of sheep change depending on the seasons. As a result, the variation of blood parameters in the organism of Korakul breed of *Ovis aries* L. depends on the climatic parameters of the external environment.

1 Introduction

In the world, climate change as a global problem has a negative impact on various sectors of the economy. Climate change has become a serious threat to the stability of human food security. The situation is increasing the relevance and necessity of studies related to climate change criteria, seasonal change, temperature and rainfall [1-2].

Living organisms are a self-regulating system in response to the influence of various biotic and abiotic factors in relation to the natural environment. Climate variability has specific effects on various physiological and biochemical indicators of animals [3-6], plants [7] and microorganisms [8].

Domestic sheep (*Ovis aries* L.) is one of the livestock that is of particular importance in ensuring the stability of food security for humans [9]. Among sheep, the Korakul breed differs from other breeds in that it can be used for meat, milk, and skin. At the same time, this breed is known for its ability to grow in extremely difficult living conditions. The Korakul sheep breed supplies the best Korakul skins in the world. In the fur industry and

[*] Corresponding author: normuhammedovafer@mail.ru

sale, known as the Korakul trade, the demand for Korakul is very high in all countries, that is why the Korakul breed has spread so widely and is bred in more than fifty countries in Asia, Africa, Europe and the Americas. The average number of Korakul sheep breeds and hybrids of their type in the world is more than 30 million heads, and the production of Korakul skins is 9-10 million pieces [10]. Therefore, in order to effectively care for sheep in extreme conditions and increase their productivity, there is an increasing need to carry out research aimed at evaluating the effects of extreme climate variability on the physiological state and biochemical indicators of sheep.

The blood system returns a perfect response reaction to any negative impact of the external environment in the animal organism. The fact that blood has such a feature is characterized by the fact that it is a functional system that unites many systems of the body [11]. Therefore, changes in the blood system have constantly attracted the attention of researchers. Any negative effects are interconnected with the hematopoietic cell and immune systems and are manifested in clinical immunological and hematological processes. An objective assessment of the complications of adverse factors on the condition of the blood of animals serves as a basis for taking into account the dynamics of the body's adaptive mechanisms [12]. Analyzing hematological deviations caused by adverse factors into the characteristics of phenotypic adaptations and the characteristics of participation of secondary functional systems leads to important results [13].

In scientific sources, the lack of information on the effect of climate changes and the change of seasons on the variability of blood parameters of sheep indicates that in-depth research has not been conducted in this regard.

Based on the data analyzed above, in our study we aimed to determine the effect of seasonal changes on blood parameters in the Korakul breed of *Ovis aries* L.

2 Materials and methods

2.1 Study area

A desert region with an extreme climate and nutrient base was chosen as the research area. Location of the study area are presented in Figure 1.

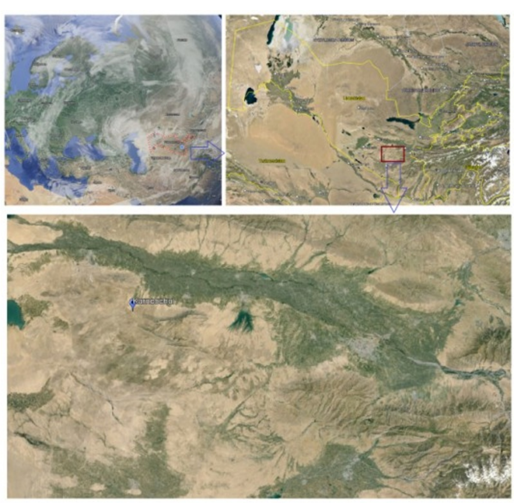

Fig. 1. Map of the study area (Location: Karnobchul; Coordinate: 39°51'32.2"N, 65°36'10.6"E; Altitude (m asl.): 500-700).

2.2 Animal material and experiential procedures

In the experiments, the Korakul breed of O. aries was selected as a research object. 30 sheep not less than one year old were selected for the experiment. Data obtained at the beginning of the experiment were used as a control option for all indicators. Feeding of sheep was carried out by providing additional rations in the winter season along with other sheep in the herd, and by grazing in other seasons. Blood samples were taken in January for the winter season, April for the spring season, July for the summer season, and October for the fall season.

2.3 Determination of the morphological composition of the blood

The method of Khayitov (2023) was used to determine morphological indicators of blood [14]. Analysis of the amount of erythrocytes, leukocytes, and hemoglobin in the blood were performed on the Automated hematology analyzer BC-5000 (Mindray). In the experiments, the blood taken from the research subjects was initially placed in vacuum tubes containing 1 mL of EDTA (Ethylenediaminetetra-acetic acid) solution. The test tube was left for 2-3 minutes. The precipitate from the test tube was taken into the device tube and placed in the analyzer. The obtained results were analyzed and average indicators were determined.

2.4 Determination metabolism of proteins in the blood

The method of Duda et al. (2020) measured the amount of proteins in the blood [15]. Analysis to determine the amount of albumins and globulins (a, b, and g) in the blood were performed on the Automated clinical chemistry analyzer XL-200 (ErbaMannheium). In the experiments, the blood obtained from the research subjects was placed in a test tube of 2 mL and centrifuged at 1,500 g for 10 minutes. Next, 1 mL of the resulting supernatant was poured into the cuvette of the analyzer and placed in the analyzer. The obtained results were analyzed and average indicators were determined.

2.5 Determination of indicators of immunity in the blood

The method of Khayitov (2024) was used to determine the indicators of immunity in the blood [16]. Analysis to determine the amount of T-lymphocytes, T-helpers, T-suppressors, T-active fraction, IgA, IgM, and IgG in the blood were performed on the Fully chemiluminescence immunoassay analyzer MAGLUMI-600 device. In the experiments, the blood obtained from the research subjects was placed in a test tube of 2 mL and centrifuged at 1,500 g for 10 minutes. Next, 1 mL of the resulting supernatant was poured into the cuvette of the analyzer and placed in the analyzer. The obtained results were analyzed and average indicators were determined.

2.6 Statistical analysis

Statistical processing and drawing of the results were performed using Microsoft Excel 2013 (USA) computer program. The results of the experiment were statistically summarized by evaluating the arithmetic averages of 5 repeated experiments at the level of statistical reliability of $p \leq 0.05$. In the mathematical-statistical analysis, the mean values and deviations of the indicators, as well as the calculation of the probability, were carried out according to the method of [17].

3 Results and Discussion

3.1 Morphological composition of the blood of *O. aries* (Korakul)

In our research, experiments aimed at evaluating the effect of the change of seasons on the morphological parameters of the sheep's blood, which were the object of the research, were conducted. According to the results of the research, the morphological parameters of the blood of rabbits can change depending on the seasons (Table 1). Table 2 shows that the amount of erythrocytes in the blood of Karakol sheep was 8.0±0.5 million/mm^3 in winter, and 11.2±0.3 million/mm^3, 12.6±0.4 million/mm^3 and 10.0±0.4 million/mm^3 in spring, summer and autumn, respectively. found to be mm^3. Taking into account that the amount of erythrocytes in sheep is 7.5-12.5 million/mm^3 is a normal physiological state, the decrease (in winter) and increase (in summer) of air temperature causes the amount of erythrocytes in the blood to decrease or increase to the minimum and maximum limits. The amount of leukocytes in the blood of sheep was found to be from 6.8±0.2 thousand/mm^3 to 10.9±0.6 thousand/mm^3. The minimum number of leukocytes was recorded in the summer season, and the maximum number was recorded in the winter season, indicating that the increase in temperature leads to a decrease in the number of leukocytes. It was observed that the amount of hemoglobin in the blood of sheep changes according to the amount of erythrocytes, the highest amount of hemoglobin is in the summer season, and the minimum amount of hemoglobin is in the winter season. The above-mentioned trend was observed in changes in platelet count (Table 1).

Table 1. Morphological composition of the blood of *Ovis aries* L. reared in ecologically extreme conditions, Karnobchul, Uzbekistan (n=30).

Indicators	Control (Mohammed et al., 2014; Nafisat et al., 2021; Nedeva et al., 2019)	Seasons			
		Winter	Spring	Summer	Autumn
Erythrocytes, million/mm^3	7.5-12.5	8.0±0.5	11.2±0.3	12.6±0.4	10.0±0.4
Leukocytes, thousand/mm^3	6.0-11.0	10.9±0.6	8.0±0.4	6.8±0.2	9.4±0.3
Platelets, thousand/mm^3	300.0-400.0	298.6±10.9	330.2±13.2	347.3±11.6*	320.1±13.2
Hemoglobin, g/l	90-140	90.8±2.4	120.4±3.7	124.9±4.5	118.8±3.6

Note. *P<0.05.

The results of the experiments aimed at evaluating the effect of the change of seasons on the morphological indicators of the blood of O. aries showed that with a decrease in temperature, the amount of erythrocytes in the blood and, accordingly, hemoglobin decreases, and the amount of leukocytes in the blood increases. As a result of research conducted on different breeds of sheep, it was noted that the amount of erythrocytes in sheep decreases by 17-25%, and the amount of leukocytes increases by 12-20% due to the influence of climate change, urbanization, diseases and various environmental factors on the variability of blood morphological indicators [13]. This information confirms that the information received by us is reasonable.

During our research, we analyzed leukograms of blood samples taken from sheep in different seasons. As a result of the experiments, it was noted that the amount of basophils in the blood was 0.30±0.01% in winter, while in summer their amount decreased by 2 times with the increase in temperature. It was found that the percentage of eosinophils in the blood decreased from spring to winter. Also, the percentage of neutrophils in sheep's blood

had different values depending on the change of seasons. In particular, in the winter, the amount of neutrophils with segmental and rod nuclei in the blood was 4.0±0.1% and 36.3±1.2, respectively, while in the summer, their amount decreased to 2.4±0.1% and 28.2±1.5%, respectively. From the data of Table 3, it can be seen that the amount of lymphocytes in the blood of sheep has a maximum value in spring and a minimum value in autumn, and the amount of monocytes in blood has a maximum value in winter and a minimum value in spring (Table 2).

Table 2. The influence of the seasons on the leukogram of the blood of *Ovis aries* L. reared in ecologically extreme conditions, Karnobchul, Uzbekistan (n=30).

Indicators	**Control (Mohammed et al., 2014; Nafisat et al., 2021; Nedeva et al., 2019)**	**Seasons**			
		Winter	**Spring**	**Summer**	**Autumn**
Basophils, %	0.2-0.8	0.30±0.01	0.22±0.02	0.15±0.01	0.18±0.02
Eosinophils, %	2.0-8.0	2.5±0.2	4.4±0.2	3.5±0.3	3.3±0.2
Neutrophils with rod nuclei,%	0.4-4.0	4.0±0.1	2.6±0.2	2.4±0.1	3.1±0.2
Neutrophils with segmented nuclei, %	27.0-41.0	36.3±1.2	27.7±1.2	28.2±1.5	28.4±1.0
Lymphocytes, %	50.0-62.0	60.6±1.9	62.2±2.2	61.2±1.6	53.1±1.8
Monocytes, %	1.4-5.8	4.5±0.4	3.6±0.7	3.8±0.5	4.1±0.8

Note. *P<0.05.

Leukocytes indicate the state of the body's natural resistance factors [18]. Adaptation processes in the body of sheep undergoing a period of adaptation to new conditions or climate were manifested, first of all, by an increase in the number of leukocytes. Because white blood cells, i.e. leukocytes, are responsible for the implementation of protective reflexes in the body [11].

3.2 Proteins in the blood of O. aries (Korakul)

During our research, the protein content of blood samples taken from sheep in different seasons was studied. It was noted that the highest value of total proteins in the blood is in the summer season, and the lowest value is in the winter season. The amount of albumin in the blood was equal to 53.4±2.5% in winter, and it was found to be 27.6±3.6%, 31.6±1.7% and 44.2±1.5% in spring, summer and autumn, respectively (Figure 2).

Table 3. The influence of the seasons on proteins of the blood of *Ovis aries* L. reared in ecologically extreme conditions, Karnobchul, Uzbekistan (n=30).

Indicators	**Control (Mohammed et al., 2014; Nafisat et al., 2021; Nedeva et al., 2019)**	**Seasons**			
		Winter	**Spring**	**Summer**	**Autumn**
Total proteins, g/l	6.6-9.0	6.1±0.6	8.5±1.3	9.1±2.1	7.0±2.0
Albumins, %	30.0-60.0	53.4±2.5	27.6±3.6	31.6±1.7	44.2±1.5
Globulins, %	40.0-70.0	46.6±1.0	72.4±2.1	68.4±2.1	55.8±1.4
α-globulin	8.0-12.0	11.3±0.5	8.4±0.7	8.8±0.5	11.2±0.8
β-globulin	6.0-16.0	12.1±1.0	13.8±2.8	15.4±1.9	7.6±1.0
γ-globulin	20.0-60.0	23.3±2.4	50.2±4.3	44.2±3.1	37.0±1.4

Note. *P<0.05.

When the amount of globulins (a, b and g) in the blood of rabbits was analyzed according to the seasons, it was found that the amount of globulins in sheep's blood also changed proportionally (Table 3). The obtained data showed that with the increase in temperature, the amount of albumins in the blood of rabbits decreases and the amount of globulins increases.

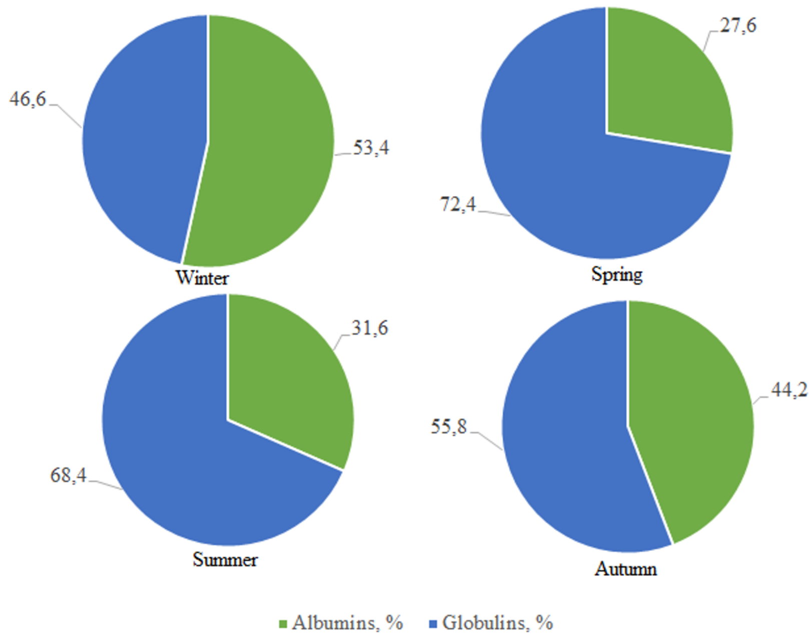

Fig. 2. The influence of the seasons on proteins of the blood of *Ovis aries* L. reared in ecologically extreme conditions, Karnobchul, Uzbekistan.

The main function of albumins is to ensure oncotic pressure in the blood due to its hydrophilicity, small size, and high concentration. Also, it mainly acts as a plastic, building material in the body [16].

Globulins combine with neutral salts in blood serum and are divided into two fractions - euglobulins and pseudoglobulins. The fraction of euglobulins mainly consists of γ-globulins, and the fraction of pseudoglobulins consists of α-, β- and γ-globulins. β-globulins cover a number of functionally important proteins. In particular, it is the transferrin protein that is responsible for the transport of iron. α- and β-globulin fractions are composed of lipoproteins, metal-bound proteins, and the γ-globulin fraction is found in a large amount of antibodies in blood serum. If the proteins of this fraction decrease, the formation of immune cells of the body decreases and the functional activity decreases [10].

3.3 Indicators of immunity in the blood of *O. aries* (Karakul)

The immune system of the body is the most dynamic system. The activity of various links of immunity undergoes constant changes under the influence of biological, physical and chemical factors of the external environment. Therefore, in our research, indicators of cellular and humoral immunity in the blood of sheep were studied depending on the change of seasons.

Indicators of cellular immunity in the blood of animals are determined by the ratio of T-lymphocytes, T-helpers, T-suppressors, T-active percentage and T-helpers and T-

suppressors in the blood. Table 4 shows that the amount of T-lymphocytes in winter (55.7±0.6%) is the lowest compared to other seasons. All parameters were observed to increase in direct proportion to the increase in temperature (Table 4).

Table 4. The influence of the seasons on the immune system cell indicators in the blood of *Ovis aries* L. reared in ecologically extreme conditions, Karnobchul, Uzbekistan (n=30).

Indicators	Control (Mohammed et al., 2014; Nafisat et al., 2021; Nedeva et al., 2019)	Seasons			
		Winter	Spring	Summer	Autumn
T-lymphocytes, %	55.0-65.0	55.7±0.6	57.2±0.8	63.0±0.4	62.8±0.5
T-helpers (Th), %	45.0-55.0	45.8±0.3	47.8±0.2	52.0±0.5	51.6±0.7
T-suppressors (Ts), %	8.0-12.0	8.7±0.5	9.3±0.4	11.0±0.4	11.2±0.4
T-active, %	45.0-55.0	43.7±0.7	45.0±0.9	52.5±0.2	53.2±0.3
Th/Ts	4.0-6.0	5.26	5.17	4.77	4.60

Note. *P<0.05.

Molecules of two types are involved in the process of differentiating foreign antigens that appear in human and animal bodies under the influence of unfavorable factors. One of these is immunoglobulins, and the other is antigen receptors of T-cells. Immunoglobulins, or antibodies, are glycoproteins found in the serum and tissue fluids of all mammals. Immunoglobulin molecules are produced in large quantities by plasma cells.

In our study, the amount of IgA, IgM and IgG was determined as indicators of humoral immunity in blood. According to the data of Figure 4, the amount of IgA in sheep increased in the sequence of winter→ spring→ autumn→ summer. Seasonal analysis of IgM level showed that it decreases in autumn→ summer→ aspring→ winter. The change of seasons did not have a significant effect on the amount of IgG in the blood of sheep (Figure 3).

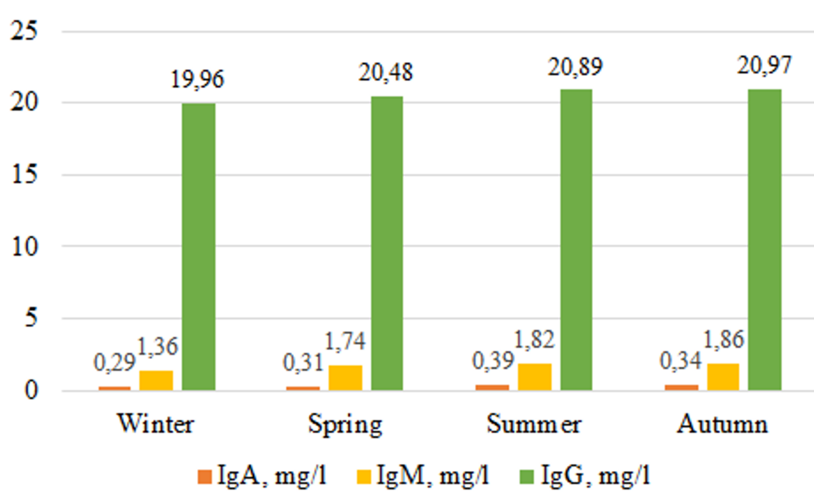

Fig. 3. The influence of seasons on the indicators of humoral immunity in the blood of *Ovis aries* L. reared in ecologically extreme conditions, Karnobchul, Uzbekistan (n=30).

IgA makes up 10-15 percent of the total amount of immunoglobulins in blood serum. These molecules are often found in the secretions of the mucous membranes, such as saliva, tear and nasal fluid, skin and lung secretions, and protect its mucous membrane from the influence of microorganisms from the external environment. IgM is the first to appear as a primary immunological response. IgM causes agglutination and cell lysis very easily. They are antigen-detecting receptors in the membrane of V-lymphocytes [19-20].

IgG is found in more tissue fluids and is important in neutralizing bacterial toxins and binding to microorganisms. In addition, IgG, forming complex compounds with bacteria, activates the complement system and causes leukocytes to chemotaxis.

4 Conclusion

It was noted that changes in air temperature and amount of rain and the change of seasons affect the blood parameters in the Korakul breed of *Ovis aries* L. An increase in temperature leads to an increase in the amount of erythrocytes, hemoglobin, a decrease in leukocytes and its components, a decrease in the amount of albumins, an increase in the amount of globulins, an increase in the number of T-lymphocytes, T-helpers, T-suppressors, the percentage of T-active and T-helpers, and an increase in the amount of IgA., causes a decrease in the amount of IgM and IgG.

Acknowledgement

The authors would like to thank the Vivary (biophysiological and biochemical research) laboratory of Samarkand State University named after Sharof Rashidov for providing appropriate equipment and reagents for conducting research experiments.

References

1. A. Mohammed, M. Campbel, F.G. Youssef, Serum Copper and Haematological Values of Sheep of Different Physiological Stages in the Dry and Wet Seasons of Central Trinidad. Veterinary Medicine International, 1- 7 (2014) http://dx.doi.org/10.1155/2014/972074
2. B. Bazarov, Z. Rajamuradov, M. Safin, A. Rajabov, D. Khayitov, M. Kuziev, S. Aminjonov, M. Ismayilova, J. Kudratov, M. Khujabekov, D. Khaydarov, The productivity, chemical composition and nutritional value of pastures dominated by Artemisia diffusa and Cousinia resinosa in arid lands of southwestern Uzbekistan, Biodiversitas, **24**, 3916-3923 (2023) https://doi.org/10.13057/biodiv/d240730
3. F. Khalimov, M. Rakhimov, U. Usanov, R. Khamzaev, E. Abdullaev, Composition and Structure of the Entomofauna of Ferula (Ferula kuhistanica) in Different Sections of the Zarafshan Ridge. Journal of the Entomological Research Society, **25**, **2**, 275–286 (2023) https://doi.org/10.51963/jers.2023.8
4. J. Kudratov, A. Pazilov, Z. Maxammadiyev, R. Urazova, B. Otakulov, B. Bazarov, K. Keldiyarov, Z. Soatova, X. Urinova, Diversity and ecology of molluscs (Gastropods) in mountain streams, Nurota mountain range, Uzbekistan, Biodiversitas, **24**, **4**, 2402-2408 (2023) https://doi.org/10.13057/biodiv/d240455
5. J. Kudratov, A. Pazilov, D. Zokirova, B. Otakulov, J. Jalilov, X. Urinova, F. Khalimov, Z. Izzatullaev, Complexes of terrestrial molluscs of various biotopes in the Gissar Range, Uzbekistan. Acta Biologica Sibirica, **10**, 349–359 (2024) https://doi.org/10.5281/zenodo.11160044
6. L.K. Alimova, M.S. Umurzakova, D.F. Zokirova, R.A. Khamzaev, G. Normuradova, B. Otakulov, A. Pazilov, J.A. Kudratov, R.S. Urazova, F.Z. Khalimo, Diversity and features of the fauna of herpetobiont beetles (Carabidae, Tenebrio-nidae, Elateridae, Scarabaeidae) of the Lower Zeravshan, Uzbekistan. Biosystems Diversity, **32**, **1**, 73–82 (2024) https://doi.org/10.15421/012407

7. S.K. Urokov, M. Usmanova, D. Xadjayev, Z. Jurayeva, A. Khujanov, M. Raimkulova, Effects of Salinity and Drought of Germination Parameters of Seeds of Triticosecale. Journal of Ecological Engineering, **25**, **7**, 178-186 (2024) https://doi.org/10.12911/22998993/188333
8. T.M. Al-Thuwaini, The relationship of hematological parameters with adaptation and reproduction in sheep; A review study. Iraqi Journal of Veterinary Sciences, **35**, **3**, 575-580 (2021) https://doi.org/10.33899/ijvs.2020.127253.1490
9. M. Muhammad, J.E. Stokes, L. Manning, Positive Aspects of Welfare in Sheep: Current Debates and Future Opportunities. Animals , **12**, 3265 (2022) https://doi.org/10.3390/ani12233265
10. S.A.A. Musavi, A.M. Khadimiyan, A.M. Azimi, Morphological Characterization of Karakul Sheep in North Part of Afghanistan. Voice of the Publisher, **8**, 16-25 (2022) https://doi.org/10.4236/vp.2022.81003
11. A. Nafisat, B. Boyi, S.T. Mbap, T. Elizabeth, Y. Ibrahim, A.M. Ja'afar, A. Shuaibu, Effect of breed, sex, age and season on the haematological parameters of sheep in Bauchi state, Nigeria. Nigerian J. Anim. Sci, **23**, **2**, 28-37 (2021)
12. I. Nedeva, T. Slavov, I. Varlyakov, V. Rade, D. Panayotov, Haematological and blood biochemical parameters in Lacaune dairy sheep. Bulgarian Journal of Agricultural Science, **25**, **1**, 91–95 (2019)
13. S.D. Rathwa, A.A. Vasava, M.M. Pathan, S.P. Madhira, Y.G. Patel, A.M. Pande, Effect of season on physiological, biochemical, hormonal, and oxidative stress parameters of indigenous sheep, Veterinary World, **10**, **6**, 650-654 (2017) https://doi.org/10.14202/vetworld.2017.650-654
14. D.G. Khayitov, Biological Sensitization of Blood Immune Cells of Rabbits to External Environmental Stress Factors (Temperature And Seasons). Bulletin of Pure and Applied Sciences-Zoology, **42A**, **1**, 163-169 (2023) https://doi.org/10.48165/bpas.2023.42A.1.15
15. Y.V. Duda, M.P. Prus, R.S. Shevchik, L.V. Koreyba, R.V. Mylostyvyi, V.V. Samoiliuk, Seasonal influence on biochemical blood parameters in males of Californian rabbit breed. Ukrainian Journal of Ecology, **10**, **4**, 262-268 (2020) https://doi.org/10.15421/2020_197
16. D. Khayitov, Z. Rajamuradov, B. Bazarov, M. Kuziev, M. Ismayilova, N. Allanazarova, S. Aminjonov, O. Tojikulova, N. Rakhmatova, D. Namazova, M. Normurodova, M. Khujabekov, D. Khaydarov, N. Rajamuradova, Diversity of blood parameters of Oryctolagus cuniculus var. domestica L. reared in different ecological regions of Zarafshan Oasis, Uzbekistan. Biodiversitas, **25**,1465-1471 (2024) https://doi.org/10.13057/biodiv/d250414
17. G.F. Lakin, Biometrics (Higher school, Moscow, 1990)
18. D. Adamchuk, M. Kuziev, E. Gurman, B. Niyazmetov, Influence of papaverine and commercial dietary supplements on blood glucose and body weight in obese dogs. Georgian Med News, **336**, 28-31 (2023)
19. K. Yunusov, K. Boymurodov, A. Egamkulov, G. Dilmurodov, F. Djalilov, Distribution of hydrobionts in aquatic ecosystems in different parts of the akdaryo river. E3S Web of Conferences, **539**, 01012 (2024)
20. M. Tibbo, T. Jibril, M. Woldemeskel, F. Dawo, K. Aragaw, J.E.O. Rege, Factors affecting haematological profiles in three Ethiopian indigenous goat breeds. International Journal of Applied Veterinary Medicine, **2**, **4**, 297 –309 (2024)