

PHYSIOLOGICAL STATE OF ANIMAL HEALTH IN THE CONDITIONS OF TECHNOGENIC AGROECOSYSTEMS

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ABSTRACT

The physiological state of the animal's body can be characterized based on hematological indicators since blood plays a special role in the body. Moving nutrients and active biological substances, the blood plays the regulator role of all vital functions of the body. Around each cell, it creates the internal environment of the body, the stability of the conductor, which is necessary for the life of all organs and tissues. It is widely known that the state of a living organism depends on the protective properties and adaptability of its body. Biochemical analysis of the blood composition can characterize these properties. The obtained data were compared with the reference indicators of metabolism in the body of cows. The determining results of total protein and protein fractions content in the blood serum indicate that the individuals of the experimental groups as a result of the influence of adverse environmental factors on the animal body revealed a small decrease in the concentration of total protein and manifest significant changes in the proteinogram, while the protein composition of the blood serum undergoes significant changes when compared with the normal spectrum of proteins. It was found that the total concentration of sugar in the blood of cows is significantly lower than the reference values indicating a direct relationship with the concentration of total protein and the nature of the serum proteinogram. Changes in the most important metabolic concentrations of carbohydrates in the blood of cows are narrowly directed and confirm the data obtained on liver dysfunction, but the rate of increase in their level is different. For the level of pyruvic acid, the main metabolites of glycolysis and glyconeogenesis are significantly higher than the reference values.

Disciplinary: Biotechnology, Bioengineering, Zoological Engineering; Agricultural Science, Biology, Veterinary Medicine, Biochemistry.

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1 INTRODUCTION

In modern conditions, it is more true and relevant than ever that the main factor of the biosphere is chemical: "Having a geochemical approach to the study of geological processes, we cover all the surrounding nature in a single atomic aspect."

Identification of areas with excessive or insufficient content of trace elements in the soil makes it possible to control the level of their content for the production of agricultural products of high quality and the prevention of endemic diseases of animals and humans (Titov, et al., 1994). To do this, it is necessary to study the immunobiochemical state and the nature of exchange processes in agricultural animals at a narrow territorial level, consider the environmental features of polluted regions and find ways to increase the adaptive capabilities of the body. Our scientific article involves these issues.

2 ANIMALS IN THE TECHNOGENIC BIOGEOCHEMICAL PROVINCE, FOREST-STEPPE ZONE OF THE SOUTHERN URALS

The current state of animal husbandry in agricultural sectors of all forms of ownership, research of blood, nutrition, animal organs, suggests that one of the main reasons for the low efficiency of this sector of farms (low productivity of breeding stock and male producers, the birth rate of low-power animals, low quality and low quality of animals) is the constant instability in other environmentally disadvantaged areas (Taranov and Sabirov, 1987). But, despite the huge number of studies on the study of microbiogenic components in the plant-soil-animal system, a lot of questions about the mechanism of origin of the processes of imbalance in metabolism in microelementosis from appearance to clinical manifestation, until now, little has been studied. At the same time, it should be noted that during their work, veterinarians began to encounter diseases that are very difficult to diagnose and there are no effective ways to treat new previously unknown diseases (Sysoev, 1981).

Research conducted in different regions of Russia has revealed a General trend: the General condition of animals in different regions, which are characterized by different stress conditions, differ in some specific parameters.

This indicates the need to study the relationship between a living organism and the environment at the regional level, taking into account the ecological properties of these territories. Also, special attention should be paid to the study of the effect of excess or lack of trace elements on changes in the biogeochemical food chains and intermediate metabolism, the adaptation of the animal body to the geochemical conditions of the surrounding environment, the appearance of biogeochemical endemics (Taranenko, 1982)

3 MATERIALS AND METHODS OF RESEARCH

To perform the task, groups of cows were formed on the principle of analogs: age from 6 to 7 years, the living weight of which - from 500 to 520 kg, the term of insularity - from 3.0 to 3.5 months (I group). A similar group of animals was formed in a region characterized by a similar biogeochemical situation (Group II).

To calculate the concentration of total protein in the blood serum of an animal, blood was taken from the jugular vein in a special tube containing a coagulating substance see Appendix. Before taking blood for research, the animal is kept on a starvation diet for 8 hours. Blood is taken before the

use of medicinal drugs that may affect the research results. The qualitative set of plasma proteins is very diverse (Fatkullov, 2014). The total protein was divided into separate fractions using the electrophoresis method, which is based on the difference between protein suspensions based on different mass values and a specific charge of a single protein. In electrophoretic separation, depending on the carrier, the number of protein fractions of the total protein varies.

Albumins are synthesized in the liver and are simple proteins containing up to 6 amino acid residues. They are highly soluble in water. The normalized value is 56.5-66.8.

Serum albumin accounts for approximately 60% of total protein. Albumins play an important role in transporting many biological materials, especially hormones. They can bind to CS, bilirubin. A significant portion of calcium in the blood is also associated with albumin. Albumins can combine with various drugs (Titov, *et al.*, 1994).

Plasma globulins are many different proteins. During electrophoresis, they go for albumin. The connection with lipids provides a complex of globulins with a soluble state and transport to various tissues.

3.1 CONDUCTING ELECTROPHORESIS

Dry membranes are carefully placed on the surface of the buffer for electrophoresis, avoiding their rapid immersion, and allowed to stand until completely wetted (Gaal *et al.*, 1982). Wet membranes should be wetted between sheets of thick filter paper to prevent them from drying out. Before use, samples need to carry out a phase of Preparata (ready). To do this, place the membrane in the electrophoresis chamber and activate the current in the selected mode for 10 minutes. The preparatory stage can be replaced by prolonged soaking of the membrane in a buffer solution (several hours) (Osterman, 1981).

We use a conductor to apply the analyzed images of blood serum at a distance of 2-3 cm from the cathode edge of the membrane (Telitchenko and Ostroumov, 1990). We place the membrane in the electrophoretic chamber and connect the current. After disconnecting the current, the membrane is carefully transferred to the dye solution for 3-5 minutes, then twice for 3 minutes in a 5-7% acetic acid solution (before bleaching the background). The electrophoregram is processed using a scanner and a computer program.

Biochemical and morphological studies of blood were carried out according to generally accepted methods based on the interdepartmental laboratory of the South Ural state university.

4 HEMATOLOGICAL PARAMETERS OF BLOOD OF ANIMALS

Age stages and periods of individual development of an animal, as well as conditions not peculiar to a healthy body, are accompanied by certain mechanisms of biochemical reactions that underlie the adaptive abilities of the body to respond to changes in the environment. The following biochemical adaptive factors directly affect the course of these reactions: 1) immunobiological 2) neuroendocrine 3) genetically determined.

Biochemical parameters to judge the condition of the body systems that makes it possible in turn to judge the intensity and nature of the disease (pathogenesis), for the study of adaptive abilities of animals when contaminated ecosystems with toxic elements, identify the characteristics of specific

reactions of metabolism studies have been conducted biochemical indices of mineral status, lipid, protein, and carbohydrate metabolism in cows. Laboratory tests were selected based on clinical and biochemical criteria for evaluating the pathological conditions of the animal body (Sova and Kusaykin, 2006).

The obtained data were compared with reference values of metabolism in the body of cattle. The results of the research content in the blood serum of cows total protein and protein fractions suggests that individuals in the experimental groups as a consequence of effects on the body, adverse environmental factors environment has a small reduction of the concentration of total protein and the significant changes in proteinogram, with all this protein in serum of cows technogenic zones is undergoing tremendous changes when compared with the conventional range protein (Fatkullin, R. R., 2010). Conclusions concerning the Genesis of the content of protein fractions are put forward based on the results of the statistical processing of available data.

Based on the data from Table 1, it can be seen that the concentration of total protein in group I individuals is 82.24 ± 0.31 grams/l, which is 4.3% lower than normal ($P > 0.05$) (Figure 1). In individuals of the second group, the decrease in the level of total protein is more noticeable and is 79.32 ± 0.41 gram/l, this indicates, first of all, the inhibition of protein biosynthesis processes in the body due to the violation of protein-synthetic liver function.

These proteinograms indicate that the decrease in the level of total protein concentration is due to the low-molecular fraction of albumins, copper-containing protein-ceruloplasmin, and protective proteins: β_2 -globulins and γ -globulins, which entails the redistribution of protein fractions in the blood serum of experimental animals.

Table 1: total protein and serum fractions in the blood of cows (g / l-10; n=10).

Indicator		Total protein	serum proteins						β -Lp	
			Alb	Palb	Tf	Cp	Globulins			
							β_2 -micro	α_2 -macro- + γ_1 -		γ_2 -
Reference	$\bar{X} \pm Sx$	85.92 ± 0.21	40.32 ± 0.24	7.52 ± 0.04	8.09 ± 0.01	2.42 ± 0.04	4.82 ± 0.03	7.11 ± 0.02	9.92 ± 0.02	6.06 ± 0.07
I	$\bar{X} \pm Sx$	82.24 ± 0.31	32.85 ± 0.18	9.48 ± 0.08	10.46 ± 0.01	1.54 ± 0.03	4.31 ± 0.02	6.42 ± 0.04	8.65 ± 0.06	8.0 ± 0.12
	%	-4.30	-18.53	+26.05	+29.29	-36.36	-10.59	-10.71	-12.81	+32.01
II	$\bar{X} \pm Sx$	79.32 ± 0.41	31.24 ± 0.08	9.34 ± 0.05	10.84 ± 0.02	1.52 ± 0.09	4.10 ± 0.02	6.47 ± 0.07	8.01 ± 0.12	7.28 ± 0.12
	%	-7.70	-22.52	+24.20	+33.99	-37.19	-14.94	-9.01	-19.25	+20.13

At the same time, the concentration of serum albumin, which has a unique ability to deliver physiological metabolites and control the level of their content in the body due to their low molecular weight, is reduced, if compared with standard indicators, in individuals of group I by 18.53 and group II by 22.52%, which in the first place, evidence of the oppression of detoxification function, albumin and the formation of individual metabolites in the free biologically active state, also the decrease concentrations of albumins, entails the inhibition processes to replace outdated new proteins in the cell structures of the body.

This situation in the proteinogram is worsened by a 1.26-fold increase in the I –st group of subjects and 1.24-in the 2-nd group of postalbumins that can inhibit enzymatic catalysis in accelerated normal or abnormal proliferative processes and have the immunosuppressive ability.

Immunosuppressive activity is also shown by α_2 -macroglobulin and haptoglobin of the $\alpha_2 + \gamma_1$ -globulin fraction zone. But the total content of protein fractions of this zone was reduced in individuals of both groups by 9.01 and 10.71% when compared with the reference indicators. We

have come to the General opinion that the low content of α_2 -globulin fraction is due to the ability of α_2 -macroglobulin to attract and move the Nickel in the form of nickelplating. In the human body, this protein is contained in an amount of 43% of the total content, but the physiological purpose of this metalloprotein is not exactly known (Tonegawa, 1985).

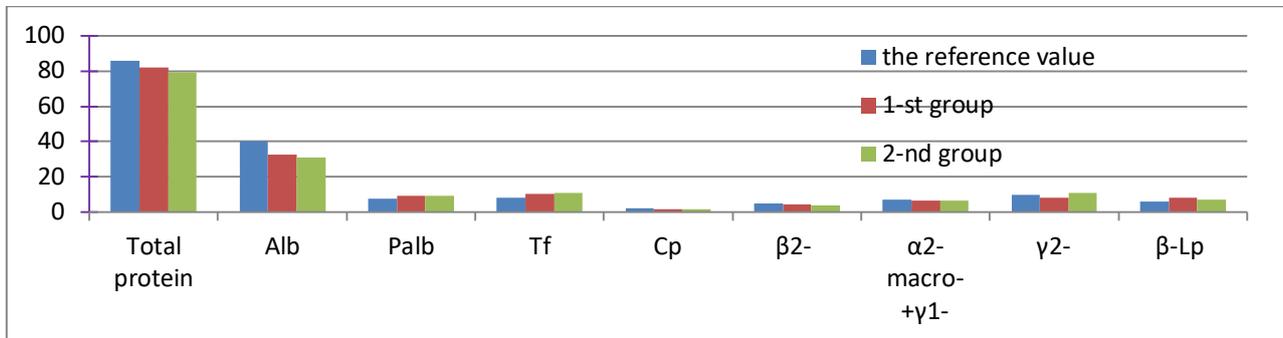


Figure 1: Total protein and serum fractions in the blood of cows (g / l-10; n=10)

The concentration of the iron-protein fraction (TF), on the contrary, increases, and its content in individuals of the I-st group was 10.46 ± 0.01 ($P < 0.001$), and the II-nd - 10.84 ± 0.02 grams/l ($P < 0.001$), this is higher than the reference values, by at least 31.64%. We believe that this increase is due to the joint effect of transferrins in hematopoiesis, with the peculiarities of their structural structure. It is proved that one of the significant components of the prosthetic group of transferrins is considered sialic acids, they are a kind of obstacle to infectious agents even before the formation of specific antibodies since iron-rich transferrin suppresses the development of bacteria and viruses. We also know that sialic acids are represented as acylated derivatives of neuraminic acids - the labile component of glycoproteins (Golitsyn, 1999). According to the results of our studies, the concentration of neuramic acid in the blood serum of experimental animals was 41.95 ± 2.31 ($P < 0.01$) and 45.31 ± 1.95 mmol/l ($P < 0.001$), respectively, in the 1st and 2nd groups of subjects. In our opinion, the decrease in the level of neuraminic acid detected by us, on average, by 20.79% is associated with the accelerated synthesis of its derivatives. Also, the concentration of transferrins in the blood serum can play the role of an indicator of pathological processes developing in the body. Some authors believe that increasing the content of transferrins has a positive effect on the condition of animals (Kamyshnikov, 2000).

The content of another transport protein α_2 -globulin fraction-copper-containing oxidase (ceruloplasmin) tends to decrease both in the I-th group of subjects and in the II-th group and is 1.54 ± 0.03 ($P < 0.001$) and 1.52 ± 0.09 grams/l ($P < 0.01$), respectively in the 1st and 2nd groups (Table1). Despite a large number of biological properties of ceruloplasmin, the decrease in its content, in our opinion, is primarily due to the inhibition of the antioxidant dynamics of copper oxidase, due to which auto-oxidation of lipids is activated and toxic radicals of superoxidation – a product of defective aerobic metabolism-accumulate in the animals of the technogenic zone (Tutelian, 1995).

Our hypothesis about a decrease in the anti-oxidant function of the blood plasma of the subjects is confirmed by an increased content of b-lipoproteins. Thus, their levels in both animal groups ($P < 0.001$) are higher than the reference values by 20.13 and 32.01% (Table 1).

Changes in the protein picture in the body of the subjects had a great impact on the level of

b2-globulins, which are protective blood proteins, the content of these proteins was reduced in the first group of subjects to 4.31 ± 0.02 , and in the second group of animals 4.10 ± 0.02 grams/l ($P < 0.001$). This decrease in the level of b2-globulins is most likely due to the toxic effects of cadmium because the increased content of cadmium in the kidneys and the harmful changes in the proximal renal tubules inhibit the reabsorption of b2-globulins.

The obtained data from the study of the protein spectrum of the blood of test animals indicate that in response to the prolonged influence of a qualitatively new environmental factor (toxic elements), an imbalance of homeostasis occurs, which through higher regulatory mechanisms of the brain (hypothalamic-pituitary system) activates systems that are responsible for adapting to changed conditions (Podvoysky and Fatkullin, 2013) The hypothalamus through releasing hormones implements the connection of the nervous system with the endocrine system (nerve impulses are transformed into humoral ones). However, the functional system that is specifically responsible for adaptability, the growth of cellular structures that limit the level of functioning of organs and cells responsible for the body's adaptability. Simultaneously elevated levels of immunosuppressors, which leads to a reduced immune function generally affect the processes responsible for the division of immune cells, and in our study, this is reflected in the decrease in the content of gamma 2-globulins in the body of the subjects of both groups to 8.65 ± 0.06 8.01 ± 0.12 g/l ($P < 0.01$), respectively, in the groups, with the average statutory rate of 9.92 ± 0.056 g/L. It is worth taking into account that the synthesis of gamma-2-globulins is a specific function of immunocompetent cells and can characterize the overall state of the cellular immune system (Zaytsev and Biochemistry, 2004).

In studying the nature of specific protein exchange pathways, the enzymes aspartate-aminotransferase (ASAT) and alanine-aminotransferase (Alat), which belong to the group of indicator enzymes or organ-specific, are of crucial importance. We found that fluctuations in the activity of serum aminotransferases in both groups of subjects have a relatively uniform character. In particular, the activity index of aspartate-aminotransferase with a high degree of confidence was increased in the individuals of group I by 86.03% and amounted to 4650.69 ± 45.26 nkat/l ($P < 0.001$), and in group II of the subjects higher than the comparison value by 52.61% (3815.16 ± 29.72 nkat/l, $P < 0.001$). It was found that ASAT, similarly to Alat, has two molecular forms – cytosolic and mitochondrial, but about 1/3 of the total activity of ASAT is concentrated in the mitochondria, and the predominant part of Alat is in the cytosol (Mironova and Sirota, 1977). According to the results of research, it became known that the critical concentration of trace elements in environmental objects mobilizes a more increased increase in the activity of the cytoplasmic enzyme (Stoyanovsky and Stupnitsky, 1985). Thus, the activity of Alat in group I subjects increased by 2.27 times, in group II-by 1.73, and the activity of ASAT-by 86.03 and 52.61%, respectively, in each group of subjects, which indicates a violation of the function and structure of hepatocytes, which is preceded, first of all, by the release and entry into the blood of the cytosolic enzyme – Alat (Tumasheva, *et al.*, 1973).

Our assumption about the presence of pathological changes in the liver of the subjects is proved by the values of the de Ritis coefficient (2.49 units; $P < 0.01$ and 2.68 units; $P < 0.001$, in both groups, respectively). An increase in this indicator indicates that pathological processes occurred in the liver, which contributed to an increase in the permeability of liver cell membranes (Urazaev *et al.*, 1990)

Due to the liver, there is an imbalance in amino acid and protein metabolism, and since the liver is the biosynthesis of all albumins, 78-90% alpha - and gamma-globulins, then the alterations of transaminases, everything seems to be directly dependent not only on the nerve-humoral regulatory

effect through inhibitors and activators, but also from inhibition of protein synthesis in the diseased liver. At the same time, an increase in the activity of aminotransferases in the blood of subjects may be an adaptive response of the body, one of the protective measures in the process of adaptation to stress-related environmental influences, which it seems to us, can be attributed to the influence of harmful environmental factors (Fatkullin *et al.*, 2016).

5 CONCLUSION

Based on the results of biochemical studies of the blood serum of subjects according to the list of values of protein metabolism in the body and their comparison with reference indicators, we can conclude that the complex influence of environmental pollution of anthropogenic nature leads to the phenomenon of self-adaptability of animals, during which latent pathological processes are briefly balanced. In the conditions of the biopathogenic zone, which is characterized by a poor content of zinc and cobalt in the diet of cows and at the same time an abundant content of Nickel, lead, manganese, copper, iron, and cadmium, there is an imbalance of metabolic processes in female cattle, which adversely affects their productivity. Prolonged exposure of animals to harmful environmental factors reduces the concentration of albumins, beta-and gamma-two-globulins in the blood, which indicates the inhibition of the humoral factor of the natural protective function of cows in the first period of pregnancy and therefore reduces the provision of the necessary plastic materials for actively developing embryos (Golitsyn, *et al.*, 1999)

Thus, inhibition of biosynthesis of proteins that perform the function of immunobiological resistance in the body of cows contributes to the involvement of amino acids in indirect transamination and deamination, which is confirmed by the nature of changes in the activity of transaminases – the most important enzymes that ensure the normal course of the glyconeogenesis process. the inclusion of amino acid metabolism products in this process, and contributes to the suppression of the activity of alkaline phosphatase, thereby exacerbating energy metabolism. As a result, there is a serious restructuring of the immune regulatory and neurohumoral mechanisms of the adaptive process of the body, accompanied by increased energy consumption.

Given the fact that animals in contaminated ecosystems with toxic elements require huge amounts of energy to adapt to the environment, it was necessary to assess the status of carbohydrate metabolism, because of carbohydrates, namely, glucose play a role as a main source of energy for cellular structures of the body. For this purpose, we determined the content of the main metabolites of carbohydrate metabolism: pyruvic acid, lactic acid, and glucose (Table 1).

According to the results of the study, it was found that the total concentration of sugar in the blood of cows of the first and second groups is significantly lower than the reference values by 13.85-15.82%. This level of glucose in the blood of cows (2.98 ± 0.09 to 3.05 ± 0.18 mmol/l), in our opinion, has a direct relationship with the concentration of total protein and the nature of the serum proteinogram (Table 1). Apparently, glucose under such conditions begins to arrive, mainly due to the reaction of glyconeogenesis from amino acids included in the metabolic processes, and directly glyconeogenesis is provided by the influence of the ordering participation in the metabolic processes of the parasympathetic nervous system, which prevents the emergence of a pronounced hypoglycemic state (Vikhlyantsev, *et al.*, 2006).

Changes in the most important metabolic concentrations of carbohydrates in the blood of cows

are narrowly directed and confirm the data we have obtained on liver dysfunction, but the rate of increase in their level is different (Georgievsky *et al.*, 1979). Thus, if the level of pyruvic acid, the main metabolite of glycolysis and glyconeogenesis, in both groups individuals are significantly higher than the reference values, on average, by 21.0%, then the level of the lactate, the final product of glycolysis, increases more significantly (1.98 ± 0.03 mmol/l and 2.22 ± 0.06 mmol/l, in both groups, respectively, $p < 0.001$) and exceeds the comparison values by 62.17 and 82.04%, respectively.

6 AVAILABILITY OF DATA AND MATERIAL

Information can be made available by contacting the corresponding author.

7 CONFIRMATION

The work was carried out in accordance with the State program for the development of competitiveness of the Russian Federation for the South Ural State Agrarian University.

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