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REFERENCE VALUES OF BLOOD BIOCHEMICAL PARAMETERS FOR THE ZHABE TYPE KAZAKH HORSE IN THE SOUTHERN REGION OF THE REPUBLIC OF KAZAKHSTAN

Abstract

The reference values for biochemical parameters in blood serum of the zhabe type Kazakh horse are presented in this article. To obtain reference biochemical parameters, whole blood samples of 50 clinically healthy adult zhabe-type horses of both sexes and raised on the free pasture of the "Kalka" farm in Zhambyl region, Shu district, Baluan Sholak, were examined on an automatic biochemical analyzer A15 (Biosystems, Spain). Biochemical results of zhabe type horses were close to the established normal indicators. The levels of enzymes of the hepatobiliary system (aspartate aminotransferase, alkaline phosphatase), triglyceride content in horses of the zhabe type were 3-5 times higher than normal. Lower concentrations of albumin and zinc were found (22.22 ± 3.67 g/l and 4.86 ± 1.49 mmol/l, respectively), as well as creatinine concentrations within the lower normal values (77.84 ± 14.19 mmol/L). Therefore, reference intervals of biochemical blood parameters (albumins, total protein, urea, uric acid, cholesterol, creatinine, bilirubin, triglycerides, glucose, calcium, phosphorus, magnesium, iron, zinc, alkaline phosphatase, alanine aminotransferase and aspartate aminotransferase) were determined for the zhabe type horse in the conditions of the Southern region of the Republic of Kazakhstan, typical for the normal course of metabolic processes in the body.

Key words: *biochemical parameters, blood, serum, zhabe type horses, reference values, intervals, mineral metabolism.*

Introduction

Meat and milk horse breeding in the republic is represented mainly by zhabe (more than 34.0% of the total number of horses in Kazakhstan), which are bred in all regions of the country. The priority of herd horse breeding in Kazakhstan is determined by the presence of large natural pastures in areas remote from large settlements, where there is an unlimited potential for obtaining high quality, horsemeat and koumiss in conditions of low-cost production. Zhabe type Kazakh breed horses were formed on the territory of Kazakhstan by natural selection under the influence of a harsh climate for many years. Zhabe differ from most Kazakh horses in high live weight and relatively large measurements (Fig. 1).



Figure 1 - Karak stallion of the zhabe type Kazakh horse breed

In recent years, in the horse breeding of the Republic of Kazakhstan, the name “zhabe” has been adapted for this type [1-3]. The Kazakh horse is polytypic: in the eastern region it is close to the Mongolian horse, in the southern and southwestern regions to the Adaev horse, in Central Kazakhstan to the steppe Kazakh horse (Fig. 2) [4]. Zhabe type Kazakh horses have a big head with massive jowls, a developed powerful dental system and chewing muscles, which allows horses to chew the coarse vegetation that they feed on. The neck of horses is fleshy and short, stallions have a large fat layer, where a kind of nutrients “reserve” accumulates. Zhabe horses have a long and deep torso, which is associated with a voluminous digestive tract adapted to the metabolizing of fiber feed. Legs bony, strong, with short but thick fetlocks; these fetlocks protect the horse legs from injury during the winter period. Covering hair (coat), mane and tail are well developed. Under favorable pasture conditions, Zhabe horses accumulate large amounts of fat underneath the skin and inside the abdomen (on the internal organs), these reserves are gradually spent to compensate for the lack of forage in winter or drying of pasture vegetation during the summer. The genetic potential of zhabe mares for live weight reaches approximately 485 kg, and stallions - 520 kg, which indicates the possibility of their further improvement on this economically useful trait [3].

Their adaptive characteristics in relation to the conditions of the breeding area deserve high attention. Stallions of this type have proven themselves as excellent improvers of local herd horses of a productive direction in a wide variety of natural conditions from the desert to northern latitudes. While the realities of the modern world where market relations dominate and only the highest quality products are in demand, it is necessary to take measures to improve livestock products and become competitive in the domestic and foreign markets. To do this, it is require to conduct breeding work on a scientific basis using advanced technologies for care, maintenance, feeding and taking into account the breed, productive characteristics and environmental and climatic conditions of animal breeding. [4].



a, b – Kazakh zhabe horse



c – Kazakh zhabe horse (Kyzylachshy)



d – Kazakh zhabe horse (Seletinskiy breed type)

Figure 2 - Zhabe type Kazakh horse [4].

Thus, previous research on zhabe horses has focused on conservation of breed, characterization of genetic diversity, and study of meat productivity along with morphological and biochemical parameters [5-6]. Studies of blood in herd horse breeding, in scientific and practical terms, are given due attention, since by studying its composition one can most deeply understand the features of the interior of animals and the biological essence of the ongoing processes. The conducted research allowed to identify the main characteristics for the zhabe type Kazakh horses [7-11]. The study of reference intervals for existing zhabe type Kazakh horses will help improve the management and maintenance of their populations. Therefore, the purpose of this study was to determine the reference biochemical parameters of blood for the zhabe type horse in the conditions of the Southern region of the Republic of Kazakhstan, typical for the normal course of metabolic processes in the body.

Materials and methods

During the study, zhabe type horses were kept in the “Kalka” peasant farm of the Zhambyl region, Shu area, Baluan Sholak rural district (Republic of Kazakhstan). The study included 50 clinically healthy adult horses (40 mares and 10 stallions). The age of the horses ranged from 3 to 8 years. Blood samples was collected from the jugular vein in the morning for one day in August 2021. Sampling was carried out in accordance with the requirements of "On approval of the Rules for sampling moved (transported) objects and biological material", Order of the Minister of Agriculture of the Republic of Kazakhstan dated April 30, 2015, № 7-1/393.

Determination of the biochemical parameters in horse blood serum was carried out on an automatic analyzer A15 (Biosystems, Spain) using reagent kits from Biosystems (Spain). The concentration of albumin, total protein, urea, uric acid, creatinine, bilirubin, cholesterol, triglycerides, glucose, minerals (calcium, phosphorus, magnesium, iron and zinc), activity of ALP (alkaline phosphatase) and transamination enzymes ALT (alanine aminotransferase) / AST (aspartate aminotransferase) were determined in horse blood serum.

Reference ranges of biochemical parameters with depending on the nature of the data distribution during processing of the study results, were determined by parametric or non-parametric methods of calculation using the Microsoft Excel 2013 application program [12].

The normality of data distribution was assessed using Shapiro-Wilk test; parameters with non-normal distribution were normalized by the Box-Cox transformation [12-13]. The blood profile was compared with reported control values for horses of foreign breeds [14-17] as an international standard as there were no previous reports of blood biochemical analyzes for zhabe type horses with a registered data.

Results and Discussion

In this study we established biochemical reference values for Zhabe horses and evaluated them in the context of known reference values for other breeds horses in general.

This is the probably more full set of reference interval data reviewed for the Zhabe horse under these conditions, although other studies have assessed the biochemical characteristics of horse blood using study animals from distinct populations from around the world.

The experiments result demonstrated that the reference intervals of biochemical parameters in blood of zhabe type horses were corresponded to the normal course of the processes of all types of metabolism in the body of the animal. The data obtained will allow for effective biological correction of the functional state of organs.

The biochemical parameters determined in this study are shown in Table 1. No differences in blood biochemical data between the two sex groups (mares and stallions) were found.

Table 1 - Reference values of biochemical parameters of zhabe type horses

Parameters	Zhabe type Kazakh horse (n=50)		Normal limits
1	2		3
Albumin (g/l)	22,22±3,67	(14,89-29,56)	25-38
Alkaline Phosphatase (U/L)	622,63±224,25	(174,14-1071,12)	70-227
Alanine aminotransferase (U/L)	12,3±4,18	(3,95-20,66)	2.7-21
Aspartate aminotransferase (U/L)	337,8±50,68	(236,44-439,16)	116-287
Bilirubin total (µmol/L)	10,08±4,49	(1,10-19,06)	5.4-51
Calcium (mmol/L)	2,7±0,23	(2,25-3,16)	2.6-3.3
Cholesterol (mmol/L)	2,59±0,26	(2,07-3,1)	1.8-3.7
Creatinine (µmol/L)	77,84±14,19	(49,46-106,22)	77-175
Glucose (mmol/L)	4,96±1,13	(2,69-7,23)	3.5-6.3
Iron (µmol/L)	19,84±5,73	(8,37-31,3)	13-23
Magnesium (mmol/L)	1,02±0,13	(0,76-1,27)	0.7-1.1
Phosphorus (mmol/L)	1,65±0,21	(1,23-2,07)	0.7-1.7
Total protein (g/L)	77,27±7,44	(62,38-92,16)	57-79
Triglycerides (mmol/L)	1,16±0,21	(0,75-1,57)	0.1-0.4
Urea (mmol/L)	4,84±0,75	(3,34-6,34)	3.7-8.8
Uric acid (µmol/L)	73,82±17,39	(39,04-108,61)	59-95
Zinc (µmol/L)	4,86±1,49	(1,87-7,84)	6.3-12 [15]

Data are expressed as mean ±2 standard deviation (2SD) or intervals. Mean ranges from – 2SD to +2SD are indicated in parentheses, alkaline phosphatase, total bilirubin, aspartate aminotransferase, magnesium and zinc ranges are calculated from normalized data using the Box-Cox transformation [12]. These limits of the norm are indicated according to reference materials [14-17].

The reference ranges of these parameters in zhabe type horses were almost within the normal range (Table 1). The concentrations of urea and uric acid of the zhabe type horses (4.84 ± 0.75 mmol/L and 73.82 ± 17.39 μ mol/L, respectively) were within physiological norm, with the exception of albumin (22.22 ± 3.67 g/L) (Table 1). A decrease in albumin shows a lack of protein at the initial stage, and at first it is compensated by an increase in the amount of globulins [10-11]. However, in our studies, the level of total protein is within the normal range, and small differences in indicators can be explained by different conditions of animal management and feeding, including the physiological characteristics of the breed [18-19].

The activity of aspartate aminotransferase (AST) was slightly higher in zhabe horses than the physiological norm. An increase in the activity of alkaline phosphatase (ALP) in the entire group of animals by 3-5 times was established. To find out the reason for the higher activity of alkaline phosphatase, it is necessary to determine the activity of gamma-glutamyl transpeptidase to exclude the pathology of bone tissue and liver. However, the reason for the somewhat higher levels of AST and ALP in zhabe horses is unclear, perhaps due to the high fat concentration in the feed.

Serum creatinine in zhabe horses was in the lower range, and serum levels may reflect muscle mass and daily exercise in small pastures.

Lipid metabolism is controlled in the blood by the concentrations of total lipids, total cholesterol, low and high density cholesterol, triglycerides. Table 1 shows ranges of cholesterol and triglyceride reference values. The cholesterol concentration was 2.59 ± 0.26 mmol/L, which corresponded to the normal limits. The levels of triglycerides (TG) in zhabe horses amounted to 1.16 ± 0.21 mmol/L, which exceeds the established norms by 2.9 times. A higher TG suggests a negative energy balance. However, most of the animals were of moderate body weight and were not overweight or malnourished.

To determine the balance of energy rations, it is necessary to assess the level of glucose, with a lack of which the animal's body seeks to compensate for the energy deficit by fat consumption. In this study, glucose levels corresponded to the normal limits (4.96 ± 1.13 mmol/L).

The usefulness of the mineral nutrition of horses depends on the serum concentration of calcium, phosphorus, magnesium, iron and zinc (Table 1). The concentration of mineral elements is in the range of normal values. The exception was zinc with a content of 4.86 ± 1.49 mmol/L, which is 0.7 times lower than normal.

Thus, comparing the obtained results with the published reference intervals for biochemical parameters of horse blood, no significant contradictions were revealed. It is difficult to determine whether the differences point to a specific characteristic of the zhabe horse because the biochemical parameters in blood of horses depends on various factors, including sex, age, season, breed, and area. [20-23]. These results confirm that it is necessary to create intra-assay databases of reference ranges, and the obtained values can be influenced by a number of factors - breed, housing and feeding conditions, climatic features. Thus, the acquisition of data will allow for a retrospective analysis of large data, obtained from intact animals and kept in equal conditions using the same analytical methods. This discussion also highlights the importance of using current and recent reference intervals for clinical assessment of veterinary cases.

Conclusions

The serum blood samples of zhabe type Kazakh horses breed, the reference intervals of biochemical parameters characteristic of the normal course of metabolic processes in the body were studied in the Southern region of the Republic of Kazakhstan. Differences in blood biochemical data between the two sex groups (mares and stallions) were not found. The reference ranges of these parameters in zhabe type horses were almost within the normal range. The concentrations of urea and uric acid in zhabe type horses (4.84 ± 0.75 mmol/L and 73.82 ± 17.39 μ mol/L, respectively) were within physiological norm, with the exception of albumin (22.22 ± 3.67 g/L). The activity of aspartate aminotransferase (AST) was slightly higher than the physiological norm in zhabe type horses. It was also established that an increase in the activity of alkaline phosphatase (ALP) in the entire group of animals by 3-5 times. The levels of triglycerides (TG) in zhabe type horses amounted to 1.16 ± 0.21 mmol/L, which exceeds the established normal ranges by 2.9 times. The concentration of mineral

elements is within the range of normal values. The exception was zinc with concentration of $4,86 \pm 1,49 \mu\text{mol/L}$, which is 0.7 times lower than normal range.

The analysis of reference intervals using the classical approach in comparison with the indicated data in the literature revealed the comparability of their values, which can be considered additional confirmation of the satisfaction of our results.

The calculated reference intervals of blood biochemical parameters characterizing the normal course of protein, lipid-carbohydrate, mineral metabolism in the body of zhabe type horses will allow preventing alimentary disorders and correcting the functional state of the body.

The obtained results enabled the establishment of biochemical reference values in the blood of Zhabe horses that will in turn support clinical diagnosis and further research into horse physiology.

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ҚАЗАҚСТАН РЕСПУБЛИКАСЫНЫҢ ОҢТҮСТІК ОБЛЫСЫНДАҒЫ ЖАБЕ ТИПІНДЕГІ ҚАЗАҚ ЖЫЛҚЫСЫНЫҢ ҚАН БИОХИМИЯЛЫҚ ПАРАМЕТРЛЕРІНІҢ РЕФЕНЦИЯЛЫҚ МӘНДЕРІ

Аңдатпа

Мақалада қазақтың жабы типі жылқы тұқымының биохимиялық көрсеткіштерінің референстік мәндерін зерттеу деректері көрсетілген. Жамбыл облысы, Шу ауданы, Балуан Шолақ ауылдық округінде орналасқан "Қалқа" шаруа қожалығында еркін жайылымда өскен, дені сау толық жетілген 50 бас жабы типі жылқы тұқымының биелері мен айғырларынан, эталонды биохимиялық көрсеткіштерді алу үшін А15 (Biosystems, Испания) автоматты биохимиялық анализаторында қан үлгілері зерттелді. Жабы типті жылқыларының биохимиялық нәтижелері белгіленген қалыпты көрсеткіштерге жақын болды. Гепатобилиарлық жүйе ферменттерінің деңгейі (аспартатаминотрансфераза, сілтілі фосфатаза), триглицеридтердің мөлшері жабы типті жылқыларда, қалыпыты жағдайдан 3-5 есе жоғары болды. Альбумин мен мырыштың неғұрлым төмен концентрациясы анықталды (тиісінше $22,22 \pm 3,67$ г/л және $4,86 \pm 1,49$ мкмоль/л), сондай-ақ креатинин концентрациясы қалыпты жағдайдың төменгі мәнінде ($77,84 \pm 14,19$ мкмоль/л) анықталды. Осылайша, Қазақстан Республикасының Оңтүстік өңірі жағдайында жабы типті жылқылардың организмдегі зат алмасу процестерінің қалыпты жүруіне тән, қанның биохимиялық референтік аралық көрсеткіштері (альбуминдер, жалпы ақуыз, мочевино, несеп қышқылы, креатинин, билирубин, холестерин, триглицеридтер, глюкоза, кальций, фосфор, магний, темір, мырыш, сілтілі фосфатаза, аланинаминотрансфераза және аспартатаминотрансфераза) анықталды.

Кілт сөздер: биохимиялық көрсеткіштер, қан, сарысу, Жабе типті жылқылар, анықтамалық мәндер, интервалдар, минералды зат алмасу.

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РЕФЕРЕНТНЫЕ ЗНАЧЕНИЯ БИОХИМИЧЕСКИХ ПОКАЗАТЕЛЕЙ КРОВИ ДЛЯ КАЗАХСКОЙ ЛОШАДИ ТИПА ЖАБЕ В ЮЖНОМ РЕГИОНЕ РЕСПУБЛИКИ КАЗАХСТАН

Аннотация

В статье представлены данные исследований референсных значений биохимических показателей Казахской породы лошадей тип жабе. Для получения эталонных биохимических показателей были исследованы на автоматическом биохимическом анализаторе А15 (Biosystems, Испания) образцы цельной крови 50 клинически здоровых взрослых лошадей тип Жабе обоих полов, выращиваемых на вольном выпасе КХ «Қалқа» Жамбылской области, Чуйского района, с.о. Балуан Шолақ. Биохимические результаты у лошадей Жабе были

близки к установленным нормальным показателям. Уровни ферментов гепатобилиарной системы (аспартатаминотрансфераза, щелочная фосфатаза), содержания триглицеридов у лошадей тип жабе были выше нормы 3-5 раз. Были установлены более низкие концентрации альбумина и цинка ($22,22 \pm 3,67$ г/л и $4,86 \pm 1,49$ мкмоль/л, соответственно), а также концентрации креатинина в пределах нижних значений нормы ($77,84 \pm 14,19$ мкмоль/л). Таким образом, были определены референсные интервалы биохимических показателей крови (альбуминов, общего белка, мочевины, мочевой кислоты, холестерина, креатинина, билирубина, триглицеридов, глюкозы, кальция, фосфора, магния, железа, цинка, щелочной фосфатазы, аланинаминотрансферазы и аспартатаминотрансферазы) для лошади тип жабе в условиях Южного региона Республики Казахстан, характерных для нормального течения обменных процессов в организме.

Ключевые слова: биохимические показатели, кровь, сыворотка, лошади тип Жабе, референсные значения, интервалы, минеральный обмен.