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Biochemical Blood Profile of the Kazakh White-Headed Breed Depending on Age

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Abstract

The reference boundaries of biochemical parameters of blood in males of the Kazakh white-headed breed have been determined, depending on the age and biological development of the body. The object of the study was to repair bulls during the rearing period and service bulls during production use. The research material was blood taken from bulls at 8, 11, 15, 18, and 21 months of age, from bulls at the age of 3, 5, and 8 years. It was established that in statistical sampling of the breed the reference boundaries of biochemical parameters of blood fluctuated in the following intervals: total protein 63.32-86.24 g/l, albumin 23.41-43.41 g/l, urea 2.25-5.16 mmol/l, AAT 1.40-4.20 mmol/h·l, ALT 0.95-3.30 mmol/h·l, total lipids 0.50-4.37 g/l, triglycerides 0.11-0.89 mmol/l, cholesterol 1.24-5.01 mmol/l, HDL cholesterol 0.94-2.53 mmol/l, LDL cholesterol 0.59-2.20 mmol/l. These boundaries for blood parameters can be used as normative indicators in animal health monitoring, conducting therapeutic measures, and developing breeding work models.

Disciplinary: Animal and Veterinary Science, BioScience.

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1 Introduction

In recent years, to meet the needs of the population and the food industry in beef, agricultural enterprises have been raising beef cattle, which has the high adaptive ability, is unpretentious to errors in feeding and maintenance, and has high growth energy. In the zones of "extreme animal husbandry" of Russia and Kazakhstan, cattle of the Kazakh White-headed breed

are bred [1]. Many breeding units have been created for the reproduction of their livestock, which has significantly increased the number of breeding animals.

To improve the efficiency and organization of breeding work, to assess the clinical status of animals, it is necessary to know the reference boundaries of blood parameters depending on age, gender, productivity, a physiological state in the context of a particular breed [2]. At the same time, some biochemical parameters of blood are species-specific. Their collection and systematization will create a data bank that can be used in breeding work.

Currently, there are a limited number of reference books that provide normative data on blood composition. At the same time, they are generalized for the type of animals (large or small cattle, horses, pigs, rabbits, poultry, etc.) [3] and do not consider their productivity direction, breed, age, genotype, etc.; they are given for the average weight of an adult animal [4]. This actualizes research in the field of age physiology of breeding cattle animals since the correct character of the variability of each blood parameter in the corresponding period of physiological development allows to assess the level of body health, the state of metabolic processes, etc.

Blood is one of the most important tissues in an animal's body. Its composition is a "variable" regulated by homeostatic mechanisms [5]. Through the blood in the animal body, almost all processes of vital activity are supported since they are based on metabolism. It is the blood that supports interstitial metabolism, as it delivers metabolic substrates to the cells of the body and removes products of oxidative decomposition [6].

When assessing the variability of blood components, it is necessary to consider that the body's ability to maintain the constancy of the internal environment decreases with age [5]. This determines the appearance of large fluctuations in the key parameters of blood in individuals of the same species in the same conditions of existence. At the same time, the regulation of homeostatic equilibrium uses not only the principle of feedback but also positive relations.

In addition to age, the level of blood indicators in cattle is influenced by the direction of breed productivity (dairy, meat). Animals of meat breeds are significantly superior to their counterparts of dairy breeds. This is due to differences in live weight, intensity and direction of metabolic processes, energy costs for the formation of productivity, etc. [3, 7- 9]. The metabolic rate of growing animals is higher than that of adults. This determines what the key parameters of the blood are, for example, nitrogenous compounds (total protein, albumin, urea, creatinine) predictors of productive qualities [10]. The breed of animals and the genetic characteristics of individuals affect the direction of metabolic flows in the body when exposed to endogenous and exogenous factors [11]. The physiological state of animals is associated with the biochemical composition of blood. Therefore, it is used to assess animal health [12], physiological and nutritional status [13]. For example, cows after calving experience a period of insulin resistance, which creates the basis for a negative energy balance and the development of ketosis [13], and indicators such as β -hydroxybutyrate, glucose, and calcium in the blood serum can serve as indicators of the risk of developing diseases of the reproductive organs [14-15]. According to [16],

variations in serum calcium, magnesium, urea, AAT, and cholesterol are associated with placenta retention, vaginal prolapse, and abortion in cows. In the body of bulls, such a blood indicator as insulin-like factor 3 is a predictor of producer fertility [17].

Thus, the biochemical composition of animal blood depends on many factors, which prompts researchers to specify their variability not only in the context of the animal species but also a specific breed. Because the sources of variations that cause physiological differences between the parameters of the biochemistry of the bull blood serum have not been studied in detail [18], the purpose of our work is to determine the reference boundaries of the biochemical parameters of blood in the Kazakh White-headed breed (3), reflecting the biological variability of the organism depending on age.

2 Materials and Methods

The work was carried out in the Republic of Kazakhstan (Kostanay region, Mendykarinsky district, Budenovka village) in the conditions of a livestock farm of the breeding company LLP Agro-industrial firm Borovskoye in 2017-2020. The choice of the farm was determined by its specialization in the cultivation of breeding young cattle of (\mathcal{C}) Kazakh White-headed breed for commercial and own purposes. This made it possible to research both repair bulls and servicing bulls. The farm uses rounded calved animals, which are massively held in February-March.

Data on repair bulls were collected in 2017-2018. The experimental group was formed after weaning young animals from mothers at 8 months of age on the principle of approximate analogs in the autumn months (September-October 2017). 20 bull calves were selected into it, whose clinical status was regularly monitored. The live weight of the bull calves at weaning exceeded the standard of the 1st class of the breed and amounted to 210-217 kg. Steers were raised separately from heifers, using loose maintenance. In winter, they were kept indoors on deep litter with access to walking grounds; in summer, pasture maintenance was used. When drawing up the feeding ration, they were guided by the norms of VIZ [19] and used feed of their own production.

For biochemical studies in bull calves at 8, 11, 15, 18, and 21 months of age, blood was taken from the subcaudal vein by the vacuum method before feeding into sterile test tubes without anticoagulants. In a thermal container, it was delivered to the laboratory of the SUrSAU (city of Troitsk, Chelyabinsk region) on the first day after sampling. The blood serum was separated by centrifugation at a speed of 3000 rpm and then used for analysis. A total of 80 blood samples were collected during the research period.

Data on servicing bulls were collected during 2016-2020. The experimental groups were formed in accordance with the principle of balanced groups. Bulls by live weight corresponded to the Elite class. In total, 3 groups of 10 heads each were formed, considering the age of the animals: the first group included 3-year-old bulls, the second – 5-year-old, and the third - 8-year-old. Servicing bulls were used for free mating and during this period were kept together with cows. The annual load on each bull was 25 heads. The feeding ration of the bulls corresponded to the norms of the VIZh [19].

Blood was taken from the bulls as well as from the repair bulls, and then delivered to the university laboratory.

Biochemical studies were performed using an automatic biochemical analyzer Super Z (manufacturer Raito Life and Analytical Sciences Co., Ltd, China) using a set of reagents from the manufacturer. It included the determination of total protein, albumins, urea, the activity of ALT (alanine aminotransferase) and AST (aspartate aminotransferase), total lipids, triglycerides, cholesterol, high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C).

Statistical data processing was performed in the MS Excel program using the "Analysis Package" application. At the same time, the average value of the parameter (X) at a certain age and its standard error (Sx), the variation of the trait in the interval "minimum – maximum", the standard deviation (SD), showing the dispersion of variables of the relative average value, were calculated. In addition, the combined mean value and its error, the standard deviation for the breed as a whole, were determined, including all variables for a specific indicator in the statistical matrix.

3 Results and Discussion

The analysis of age-related variability of key parameters of protein and lipid metabolism in the blood of male individuals of the Kazakh White-headed breed showed that the value of the standard deviation of the average value of a particular parameter increases with age. It reaches its maximum value in a statistical sample of 8-year-old bulls. Consequently, as the animals grow older, the spread of values determined by analogues in the age group becomes wider. This confirms the assumption [5] that as the body ages, deviations in regulatory mechanisms accumulate in it, which is reflected in the change in the level of the regulated variable.

The key parameter of protein metabolism is the total protein, the level of which in the blood characterizes the body's protein supply. It is a combination of all protein molecules circulating in the blood and is one of the most strictly regulated constants [20]. Its concentration steadily increased in the body of growing animals. At the same time, individuals in each age group had significant differences in the amount of total protein in the blood, since the value of the standard deviation varied from 1.24 to 3.69 (Table 1). The average value of the parameter for the breed was 74.46 \pm 0.59 g/l and the variables considered in its calculation varied in the range of 63.32-86.24 g/l.

According to [21-23], the change in total protein in the blood of individuals is determined by the quantitative variability of enzymatic parameters aimed at maintaining its level in the blood, that is, in an individual organism, the level of total protein in the blood is a "reference constant", and not a value that varies in the range of reference boundaries.

Table 1 . Age-related variability of key parameters of protein metabolism (n=20)											
Age	Total protein,		Albumins,		Urea,		AST,		ALT,		
	g/l		g/l		mmol/l		mmol/h·l		mmol/h·l		
	X±Sx	SD	X±Sx	SD	X±Sx	SD	X±Sx	SD	X±Sx	SD	
Repair bulls											
8 months	68.95±0.55	1.57	26.69±0.45	141	4.86±0.09	0.27	1.78±0.09	0.29	1.14±0.07	0.21	
	(67.32-72.79)		(23.41-27.14)		(4.38-5.16)		(1.40-2.20)		(0.95-1.40)		
11	69.21±0.48	1.54	27.88±0.51	1.60	4.41±0.12	0.39	1.98±0.10	0.30	1.37±0.10	0.33	
months	(68.94-72.97)		(25.98-30.25)	1.00	(3.91-4.94)		(1.55-2.40)		(1.05 - 1.90)		
15	72.88±0.49	1.44	29.60±0.45	1.42	3.01±0.13	0.39	2.76±0.12	0.37	2.28 ± 0.07	0.21	
months	(70.39-74.24)		(27.41-31.12)	1.42	(2.45-3.61)	0.39	(2.10-3.05)		(2.10-2.65)		
18	73.31±0.53	1.61	31.29±0.54	1.83	3.61±0.14	0.40	2.58 ± 0.09	0.27	1.98 ± 0.10	0.30	
months	(69.39-74.34)	1.01	(28.18-33.08)	1.05	(3.21-4.58)	0.40	(2.20-2.90)		(1.60-2.40)		
21	76.31±0.69	2.17	32.29±0.52	1.65	3.37±0.14	0.40	2.74 ± 0.09	0.27	2.08 ± 0.09	0.29	
months	(73.07-79.34)	2.17	(29.64-34.18)	(3.01-4.4	(3.01-4.48)		(2.40-3.20)		(1.60-2.50)		
Servicing bulls											
3 years	79.97±1.36	2.45	35.48±1.06	3.51	3.06±0.14	0.44	3.80±0.10	0.31	2.29±0.12	0.38	
	(72.39-84.24)		(30.94-40.25)		(2.46-3.88)		(3.40-4.20)		(1.90-2.90)		
5 years	81.57±0.90	2.85	39.40±1.17	3.71	3.01±0.19	0.63	3.76±0.17	0.54	2.68±0.16	0.49	
	(78.39-86.24)		(34.79-43.41)		(2.25-4.00)		(2.75-4.10)		(2.13-3.30)		
8 years	73.51±1.17	3.69	28.26±1.30	4.11	4.03±0.21	0.67	3.17±0.18	0.56	2.36±0.17	0.54	
	(68.39-77.34)		(25.25-32.80)		(3.03-5.08)		(2.40-4.20)		(1.50-3.20)		
By breed	74.46±0.59	5.28	31.36±0.97	4.83	3.67±0.09	0.83	2.82±0.09	0.77	2.03±0.09	0.59	
	(63.32-86.24)		(23.41-43.41)		(2.25-5.16)		(1.40-4.20)		(0.95-3.30)		

Table 1: Age-related variability of key parameters of protein metabolism (n=20)

The main component of the total blood protein is albumin, which is synthesized only in liver cells [22-23]. It is the main protein providing suspension and colloidal properties of blood, due to its relatively small molecular weight, a high percentage of total blood protein, and hydrophilicity [24]. For this reason, it also belongs to the "reference" indicators of blood [21]. The concentration of albumins is maximal in the blood of growing animals and during the period of the active working capacity of the body (Table 1). Nevertheless, in the body of repair bulls, the indicator is characterized by a standard deviation of variables from the average value of 1.41-1.65, and in adults 3.51-4.11. It is logical to assume that albumin, as a biochemical metabolite [Rosly L] under conditions of active growth of the organism, has a stricter enzyme-dependent dependence on the most important "metabolic highways" than in the body of adult animals.

The albumin concentration in the average statistical sample of the breed was 31.36 ± 0.97 g/l, it had fluctuation limits from 23.41-43.41 g/l, SD = 4.83 (Table 1).

Urea is an indicator of protein catabolism, reflecting the intensity of their use in the synthesis of their own protein molecules [23, 25]. The level of urea in the blood of bull calves and servicing bulls decreased with age, reflecting the active use of protein substrates in the metabolic pathways of the body, with the exception of 8-year-old animals. At the same time, a similar dynamic of the values of the standard deviation was revealed. The average urea value calculated from the statistical aggregate of the breed was equal to 3.67 ± 0.09 mmol/l with fluctuations in the min-max range from 2.25 to 5.16 mmol/l SD=0.83.

From the dynamic indicators of protein metabolism, we present data on the age variability of aminotransferases (alanine aminotransferase (ALT), aspartate aminotransferase (AST)). The metabolic role of ALT reflects the readiness of the body to use the amino acid alanine through pyruvate in glucose synthesis (gluconeogenesis), and AAT - the intensity of formation of

oxaloacetic acid from aspartic acid, followed by its use in the cycle of tricarboxylic acids (a marker of the state of mitochondria) [21-23, 25-26]. The activity of the AST enzyme in the blood of animals increased with age, reaching a maximum value at the age of 5. A similar dynamic was typical for the ALT. The exception was the age of 8 years, at which a decrease in the concentration of enzymes in the blood was noted. If we assume that the metabolism of almost all amino acids is initiated by aminotransferases [27], then the rate of their use in the biochemical reactions of males of the Kazakh White-headed breed increased with age, providing the intensity of protein metabolism. On average, the activity of AST for the breed was 2.82 ± 0.09 mmol/h·l, varying in the range from 1.40 to 4.20 mmol/h·l (SD 0.77), and ALT - 2.03 ± 0.09 mmol/h·l, varying from 0.95 to 3.30 mmol/h·l (SD 0.59). At the same time, we would like to emphasize that although the activity of enzymes changes its value in the age scale, but in an individual organism, it remains a specific value corresponding, first, to the concentration of total protein. In general, the direction of the change in the activity of aminotransferases was combined with the age dynamics of the total protein.

Age	Total lipids, g/l		Triglycerides, mmol/l		Cholesterol, mmol/l		HDL cholesterol, mmol/l		LDL cholesterol, mmol/l		
-	X±Sx	SD	X±Sx	SD	X±Sx	SD	X±Sx	SD	X±Sx	SD	
Repair bulls											
8 months	2.84±0.17 (2.07-3.54)	0.55	0.36±0.02 (0.26-0.45)	0.07	4.29±0.10 (3.79-5.01)	0.39	1.36±0.02 (1.25-1.42)	0.06	0.97±0.03 (0.85-1.09)	0.09	
11 months	3.37±0.24 (2.13-4.07)	0.74	0.45±0.02 (0.38-0.53)	0.06	4.11±0.10 (3.81-4.55)	0.31	1.41±0.01 (1.36-1.49)	0.05	1.05±0.02 (0.99-1.14)	0.06	
15 months	3.61±0.18 (2.97-4.27)	0.58	0.55±0.03 (0.47-0.75)	0.11	3.53±0.11 (2.85-4.65)	0.36	1.65±0.02 (1.56-1.73)	0.08	1.09±0.03 (0.97-1.21)	0.09	
18 months	3.79±0.19 (2.79-4.27)	0.60	0.61±0.04 (0.53-0.88)	0.13	3.08±0.10 (2.72-3.51)	0.32	1.67±0.03 (1.54-1.79)	0.11	1.11±0.06 (0.86-1.29)	0.19	
21 months	3.81±0.20 (2.69-4.37)	0.66	0.63±0.04 (0.55-0.89)	0.12	2.99±0.12 (2.50-3.51)	0.39	1.66±0.03 (1.54-1.77)	0.10	1.13±0.05 (0.93-1.30)	0.16	
Servicing bulls											
3 years	1.96±0.13 (1.12-2.77)	0.43	0.43±0.06 (0.15-0.69)	0.20	2.86±0.15 (2.17-3.45)	0.49	1.98±0.07 (1.75-2.31)	0.22	1.14±0.13 (0.59-1.69)	0.35	
5 years	2.09±0.12 (1.05-2.27)	0.40	0.48±0.06 (0.27-0.81)	0.26	2.91±0.17 (2.25-3.53)	0.55	2.13±0.11 (1.69-2.53)	0.35	1.17±0.09 (0.68-1.82)	0.35	
8 years	1.08±0.11 (0.50-1.78)	0.41	0.26±0.05 (0.11-0.53)	0.27	2.00±0.18 (1.24-2.76)	0.56	1.28±0.12 (0.94-1.99)	0.39	1.67±0.12 (1.09-2.20)	0.37	
By breed	2.82±0.12 (0.50-4.37)	1.08	0.47±0.02 (0.11-0.89)	0.18	3.21±0.09 (1.24-5.01)	0.81	1.64±0.04 (0.94-2.53)	0.33	1.16±0.03 (0.59-2.20)	0.31	

 Table 2: Age-related variability of key parameters of lipid metabolism (n=20)

Lipids perform several important biological functions in the animal body: they participate in the exchange of energy and the transport of substances, the formation of the structure of cell membranes, and the regulation of their physical properties, determining their functional activity [28]. The key parameters of the blood lipidogram are total lipids, the level of which reflects the direction of the exchange of its components [29]. The number of total lipids increased in the blood of repair bulls from 2.84 ± 0.17 (8 months) to 3.81 ± 0.17 (21 months) g/l. At the same time, the indicator was characterized by a fluctuation with SD from 0.55 to 0.74, reflecting the differences of individuals in the value of the parameter from the average value. In the blood of servicing bulls, the

concentration of total lipids was significantly lower, ranging from 1.08 ± 0.11 to 2.09 ± 0.12 g/l (SD = 0.40-0.43). Consequently, the age determined the demand for lipids in the energy metabolism of animals. If we base on the fact that changes in the level of lipids in the blood reflect the lipid metabolism in the cells of the body [30], then in the body of growing repair bulls, lipid metabolism has a higher intensity.

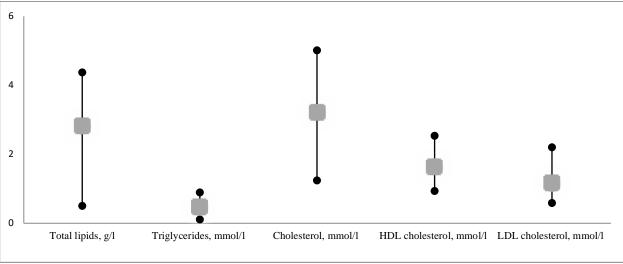


Figure 1: Reference boundaries of blood lipids in males of the Kazakh white-headed breed.

On average, the concentration of total lipids in the blood of the breed was 2.82 ± 0.12 g/l, but the level of the indicator varied from 0.50 to 4.37 g/l (SD= 1.08) (Figure. 1).

Triglycerides perform an important energy function from blood lipid parameters, the metabolic properties of which are determined by the ability, firstly, to be extracted from fat depots, and secondly, to hydrolyze to free fatty acids with their subsequent use for plastic or energy purposes [31]. At the same time, in the body of cattle, triglycerides are not used in the synthesis of fat in fat depots, but mainly propionate, acetate, or glucose [32]. Repair bulls outperformed servicing bulls in the concentration of triglycerides in the blood (Table 2). At the same time, the value of the standard deviation in the corresponding age sample was less than in adult individuals, reflecting the correspondence of their level in the body of bulls to the activity of lipid catabolism at the appropriate age. In the statistical sample of servicing bulls, significant differences between individuals in the concentration of triglycerides were revealed, determining the individual needs of the body in energy substrates.

The average value in the breed sample was 0.47±0.02 mmol/l, the parameter varied from 0.11 to 0.89 mmol/l, and the standard deviation was 0.18 (Figure 1).

One of the "reference" indicators of lipid metabolism is cholesterol [20-21]. In the animal body, homeostatic mechanisms are aimed at maintaining its level in the blood in an amount sufficient to ensure the processes of vital activity [33]. The concentration of cholesterol in the blood of males of the Kazakh White-headed breed decreased with age and reached a minimum value in the blood of 8-year-old bulls (Table 2). This was accompanied by an increase in the spread of individual values from the average value: SD in repair bulls 0.32-0.39, in servicing bulls - 0.49-

0.56. The average value of cholesterol in the blood of animals in the statistical sample of the breed was 3.21±0.09 mmol/l, varying from 1.24 to 5.01 mmol/l (SD=0.81) (Figure 1).

In the body of animals in conditions of physiological state and regulated homeostasis, the source of cholesterol is endogenous synthesis and its intake from the extracellular environment. In animals, cholesterol levels are interrelated with the process of formation and maintenance of fertility, as it is a substrate for the synthesis of sex hormones. The direction of cholesterol metabolism can be characterized by the age dynamics of HDL cholesterol and LDL cholesterol, which are transport molecules of cholesterol in the blood [34-35]. Thus, the level of HDL cholesterol and LDL cholesterol increased with age, except for 8 years of age. Nevertheless, the amount of HDL cholesterol increased more significantly than LDL cholesterol (Table 2). At the same time, the SD value of servicing bulls was higher than that of repair bulls. Their number on average for the breed was 1.64 ± 0.04 (HDL cholesterol) and 1.16 ± 0.03 (LDL cholesterol) mmol/l. The ratio of lipoproteins testified that cholesterol in the body of repair bulls and servicing bulls was used mainly in the synthesis of compounds of a steroid nature, including sex hormones.

Summing up our research, it can be noted that the optimal functioning of the body of the Kazakh White-headed breed males is associated with maintaining several key parameters of protein and lipid metabolism in the blood within certain limits. In general, by breed and depending on age in the context of the sex of animals, homeostatic mechanisms determine the variation of average biochemical parameters, some of which are "reference" and some "dynamic".

4 Conclusion

The results of calculating the average value of the parameter and its standard error, the range of variation of values in the range min-max, the standard deviation allowed to characterize the biological variability of key parameters of protein and lipid metabolism in the blood of males of the Kazakh White-headed breed depending on age and in general on the statistical sample of the breed. When analyzing the data, it was revealed that the standard deviation of the average value of the determined parameters increases with age, reflecting shifts in the regulation of homeostasis during changes in the physiological state of animals.

The concentration of biochemical parameters in the statistical sample of the breed was characterized by the following values: total protein 74.46 \pm 0.59 g/l (63.32-86.24 g/l, SD =5.28), albumins 31.36 \pm 0.97 g/l (23.41-43.41 g/l, SD =4.83), urea 3.67 \pm 0.09 mmol/l (2.25-5.16 mmol/l, SD=0.83), AST 2.82 \pm 0.09 mmol/h·l (1.40-4.20 mmol/h·l, SD=0.77), ALT 2.03 \pm 0.09 mmol/h·l (0.95-3.30 mmol/h·l, SD=0.59), total lipids 2.82 \pm 0.12 g/l (0.50-4.37 g/l, SD=1.08), triglycerides 0.47 \pm 0.02 mmol/l (0.11-0.89 mmol/l, SD=0.18), cholesterol 3.21 \pm 0.09 mmol/l (1.24-5.01 mmol/l, SD=0.81), HDL cholesterol 1.64 \pm 0.04 mmol/L (0.94-2.53 mmol/L, SD=0.33), LDL cholesterol 1.16 \pm 0.03 mmol/L (0.59-2.20 mmol/l, SD=0.31).

The obtained values can be used as normative for the quantitative assessment of the variability of biochemical parameters in the population of purebred males of the Kazakh White-

headed breed when monitoring the health of animals or carrying out therapeutic measures. In addition, they can serve as a reference database for the development of breeding models.

5 Availability of Data and Material

Data can be made available by contacting the corresponding author.

6 Ethical Statement

The study was approved by the Bioethics Committee of the South Ural State Agrarian University, Russia.

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