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INFLUENCE OF NATURAL SELECTION ON THE REPRODUCTIVE FUNCTION OF BULLS

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Aim. To investigate the influence of natural selection of bulls during tethered and non-tethered keeping on their reproductive function. **Methods**. Observation of analogue groups in different conditions of keeping, yet with the identical level of feeding and using, ethological methods of the behavior of animals in a herd and in a "mini-herd", zootechnical, genetic-mathematical, biometric and modelling methods. **Results.** It was established that dominant animals in the herd oppress the sexual ability of their subordinates, causing the decrease in their libido, erection, quality of sperm (as far as to the impotence). **Conclusions**. The influence of natural selection on the sexual potency of breeding bulls during their non-tethered keeping and some ethological specificities during the tethered keeping were established along with the reactions of an organism depending on the type of body composition and build. The application of the revealed regularities facilitates the work regarding the selection of bulls while evaluating the quality of the sperm production and ensuring the increase in the number of their offspring in further generations.

Keywords: a bull, keeping, reproduction, sperm.

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INTRODUCTION

The biological possibilities of forming and normal functioning of the reproductive system of bulls is not paid sufficient attention in many breeding enterprises due to the fact that the implementation of the genetic potential of breeding bulls in this direction depends on many factors, related to natural selection and specific conditions of keeping and using animals [1]. The production conditions are often unfavorable, which first of all has negative impact on sperm indices, which spontaneously get worse even in clinically healthy bulls [2]. It is very hard to improve using previously known methods [3]. There is a need for reliable and safe ways of influencing the reproductive system of bulls for accelerated enhancing of their sperm production and ensuring the stability of the industrial process of improving breeder's stocks, which completely depends on the satisfactory condition of the reproductive sphere of breeding bulls [4, 5].

However, there are insufficient data in the scientific literature on the influence of natural selection and elimination in the conditions of a herd or non-tethered group keeping on the reproductive function of livestock. Little has been published on the connection between zoological society adaptation, reaction of domination and subordination, and the reproductive function of animals within a herd, which is an artificial population. There are scarce studies of ethological-physiological mechanisms of the formation and keeping the herd hierarchy and its role in the processes of reproducing the offspring, as well as in natural regulation of the number of productivity of the herd due to intrasexual and epigamic selection. It has not been studied in what way the type of the nervous system influences the rank of animals within the herd.

There has practically been no studies of the ways of restricting the negative impact of natural selection using different ways of keeping the breeding bulls during the period of rearing them, learning to

get sperm and using it in the network of artificial insemination.

Taking the abovementioned into consideration, the aim of this work is to provide scientific substantiation and elaboration of the ways of keeping, using, and stimulating the reproductive function of bulls.

The analysis of the mentioned problems is of great scientific and economic relevance, it allows developing much more efficient, ethologically and ecologically grounded technology of keeping, using, and stimulating the reproductive system of bulls in the breeding farms [3].

MATERIALS AND METHODS

Multi-year researches and approbation of the results obtained were conducted in the breeding farms and agricultural enterprises of Ukraine (Crimea, Volyn, Drohobych, Sumy, Dnipropetrovsk breeding companies, Institute of Animal Breeding of the Central Districts NAAS of Ukraine), Russian Federation (Kaluga experimental station, All-Russia Animal Breeding Institute), and Tajikistan (Agro-industrial company "Khovaling", Tajikistan Research Institute for Animal Breeding) using the livestock of over 1,400 breeding bulls and 10,000 bull-calves on feed and 20,000 cows and heifers of Red Steppe, Ukrainian White-and-Black, Simmental, Schwyz, Kostroma, Kazakh Whitehead breeds aged from 12 months till 3 years old. The main methods of investigation were the observation of analogue groups in different conditions of keeping, yet with the identical level of feeding and using, ethological methods of the behavior of animals in a herd and in a "mini-herd", zootechnical, genetic-mathematical, biometric and modelling methods.

RESULTS OF INVESTIGATIONS

The impact of natural selection factors in herds was identical to that of modelling selection [6]. The ethological studies of synchronous intrasexual and epigamic selection in the herd demonstrated that in conditions of unbound mating with cows only two dominant bulls out of six mainly took part in natural reproduction. During the period of investigations the leader bull of the herd mated with 23 cows, and its closest rival – with 12. At the same time four subordinate bulls mated one cow each, *i.e.* they almost did not participate in reproduction due to the rank-related stress (Table 1).

In almost 100 herds of Khovaling company, dominant bulls had negative impact on the libido of subordinate males, which resulted in the impotence of over 60 % of the latter, which developed and persisted on the basis of non-specific adaptation reactions and stress reaction.

Due to this impact of sexual selection, the cows of the herd also had different adaptation level, i.e. the ability to survive and reproduce – 26–28 % of them did not have any calves. Here only 43.1 % of the most adapted animals had calves at foot and mated again within natural terms, better for future offspring (Table 2).

The difference in the reproduction for dominant and subordinate cows in the herd was 32 %. The live weight of heifers, calved by dominant mothers, was 400 kg, *i.e.* it almost did not change compared to the

Table 1. Mating of dominant and subordinate bulls with cows and heifers

Ethological belonging	Hierarchy rank	Number of cows and heifers, the bull mated with	Including in %
Leader bull	1	23	58.99
Dominant bull	2	12	30.77
Subordinate	3	1	2.56
Subordinate	4	1	2.56
Subordinate	5	1	2.56
Subordinate	6	1	2.56
Total	6	39	100.00

Table 2. Live weight and calving of dominant and subordinate cows and their heifers in herds, $\overline{X} \pm \overline{x}$

Ethological belonging of cows	Active factors of natural selection	Number of animals	Live weight, kg		Calving, %	
			Mothers	Daughters	Mothers	Daughters
Reproducing animals	Stabilizing, leading and sexual	423	346 ± 2.66	–	71 ± 2.5	–
Including dominant	Stabilizing and sexual	178	399 ± 3.06	400	85.9 ± 2.4	81.3
subordinates	Leading and sexual	245	301 ± 8.3	331	53.8 ± 4.9	68.2
Difference	–	–	97.6	69	32.1	13.1

mother's weight, but the conception rate decreased by 4.6 %. At the same time, the live weight of heifers, calved by subordinate cows, increased by 30 kg, and the conception rate – by 4.4 %, which may be explained by the influence of natural selection, which "pulls up" the subordinates of the herd to the average norm. The direction of natural selection (or its vector) was the same as for targeted artificial selection, which should be used while selecting cattle in the "cow-calf" system [7, 8].

Therefore, in practice it is reasonable to introduce the activity of natural selection factors, improving the conditions of feeding, keeping, and breeding animals, as the decrease in productivity and fertility as well as impaired reproductive function of herd subordinates, caused by the natural regulation of the herd number, are the consequence of negative influence of the selection on the efficiency of the industry as a whole, which is also confirmed in other works [9, 10].

Sexual selection is active at the level of the herd which is an artificial population. It is influenced by such probable causes as the number of dominant bulls in the herd as well as the number of cows in mating mood per time unit, and the duration of mating with cows and heifers in mating mood. At the same time there are two factors in the herd, related to the decrease in its number. First of all, it is the access of animals to all the herd resources, which ensures the reproduction of dominant animals at the expense of the subordinates. In addition, there is a factor of availability which is manifested when all the bulls are busy mating with cows, while probable animals in mating mood are removed from reproduction, though they could have their offspring [11].

Here the number of cows, which have had their mating and the ones, removed from mating, conditions the elimination, which acts on the basis of adaptation reactions and stress reaction. This leads to the impotence of subordinate bull-calves and to the absence of calves from subordinate cows due to the impact of zoo-social ranking stress [12].

More plausible observations of the influence of natural selection required not mere determination of the bulls, left out of natural reproduction, but also defining the direction or vector of the selection and proving selective elimination of subordinates and selective survival and participation of dominant herd members in the reproduction. Here the direction of sexual selection is manifested in circadian behavior of animals, registered by genetic memory and occurring during the relations of animals within the herd from generation to generation without any teaching.

Dominant bulls had first access to feeds, water, salt, and better places of rest. They ate more concentrated feeds and spent considerably less time to play compared with subordinate breeding bulls in the herd. These bulls demonstrated higher ability of extrapolating [1].

Dominant bulls moved less, they mostly lay down or stood, feeling more comfortable than subordinate breeding bulls, and they demonstrated their sexual activity only towards cows and heifers in mating mood. Contrary to dominant bulls, subordinate ones had sexual arousal only towards each other. This fact was related to absence of their stable libido towards cows and heifers in mating mood [13].

Most subordinate bulls had natural impotence, conditioned by ranking stress, due to which they could not mate even with a tethered cow in mating mood [14].

Thus, the study of daily (circadian) behavior of bulls demonstrates objective evidence of the fact that indeed it manifests the vector of sexual selection, directed at predominant survival and reproduction of dominant animals in herds.

Primitive emotions of defensive type, which were shown by animals in competitive conditions of the herd, related to the vector of sexual selection, had considerable influence on somatic reconstruction of the organism, which was manifested in their ethological and physiological indices and sperm production [2].

It was established that higher adaptive possibilities were noted for dominant animals with strong, balanced, flexible type of higher nervous activity (HNA) compared to subordinate animals with similar type of HNA, but with significant differences in the indices of the number of differentiations and the number of positive responses as well as transformation of negative reaction into positive one ("–" into "+"). This fact is related to the balance of nervous processes which testifies to higher ability of dominants to study and analyze external factors.

Live weight and sperm of dominant Schwyz bull-calves was considerably better compared to subordinate animals of the herd during the whole period of breeding animals (Table 3).

There was considerable difference in the live weight. Sperm indices of dominant and subordinate bull-calves were also considerably ($P < 0.01$) different in terms of activity (motility) of sperm cells which may be ex-

plained by the impact of zoo-social stress on subordinate animals. Therefore, new data, proving that sperm also reflects adaptive processes in the animal organism and may serve as objective characteristics of its state, have been obtained.

The behavior of live animals in nature regulates reproductive processes, which are the basis of species evolution via isolation, which restricts their mating and migration to other populations (herds).

It was experimentally established that sexual reactions of bull-calves towards each other in the process of hierarchy formation after they have been transferred from tethered to non-tethered keeping or after "new" animals have been introduced to the previously formed herd may be divided into three periods.

During the first period (a week), especially during the first hour after the transfer, bull-calves demonstrated energetic sexual reactions towards each other. Here both pulse and breathing rates of animals increased twice or more. Blood tests of 20 Schwyz bull-calves, which had been tethered, made a day after they were transferred to non-tethered keeping, demonstrated that these animals were stressed. During the first day the live weight of bull-calves decreased by 20–30 kg. Sperm activity dropped sharply ($P < 0.05$). The functional activity of sexual glands of bull-calves was inhibited. The breathing coefficient also dropped sharply – from 1.0 to 0.7.

During the second period (20 days), there was some stabilization of homosexual reactions of bull-calves in the herd, which was accompanied with further inhibition of sexual and defensive refluxes. The sperm production of bull-calves was improving with statistical reliability. After 10 days of non-tethered keeping, blood plasma indices of bull-calves decreased almost to the initial level, but were still higher compared to the period before the transfer. At this time the sexual function of bull-calves was not subject to such overpressure.

The third period (30–40 days) was remarkable for clear determination of hierarchy among the animals of the herd. There was first phenomenon of relative extinction of sexual arousal (libido) of bull-calves to the animals of their herd. However, as soon as "new" bull-calves (one or several) were transferred to the herd, sexual reactions of previously transferred ones towards "new" animals were manifested again. During this period they regained live weight, lost before, with average daily gain of almost 900 g, and sperm production improved.

Thus, new data have been obtained, proving that the basis of regularities of the formation and maintenance of hierarchy in the herd of bull-calves lies in complicated complexes of behavior and physiological reactions. It was first determined for leader bulls that there was such a specific quality of hierarchical rank as its being determined by HNA properties, which is relevant for the practice of selecting and evaluating animals.

The place of the leader of the herd was always taken by the most aggressive bull-calves of strong, balanced, flexible type of HNA, whose sperm quality almost did not decrease, which testified to absence of any stress. The distribution of non-leading bull-calves of different hierarchical ranks and types of HNA in the herd was random. Bull-calves with weak type of HNA did not participate in active fight for the hierarchy rank and took subordinate position in the herd. The selection of dominant bulls may be conducted in specialized premises, evaluating them by productive qualities

Table 3. Live weight and sperm quality of 18-month-old bull-calves $\overline{X} \pm \overline{x}$

Index	Dominant, $n = 5$	Subordinates, $n = 6$	± Dominant to subordinates	Statistical reliability of td difference at $P < 0.01$
Live weight (kg) at the age of (months)				
9	236 ± 10.6	221 ± 10.5	+ 15	1.0
12	302 ± 13.2	286 ± 13.3	+ 16	0.8
15	363 ± 15.6	330 ± 13.2	+ 33	1.4
18	439 ± 19.8	389 ± 12.4	+ 50	2.0
Sperm quality:				
volume, ml	2.9 ± 0.2	2.2 ± 0.9	+ 0.7	0.76
activity, points	5	4	+ 1	1.0
concentration, billion/ml	0.672 ± 0.17	0.487 ± 0.61	+ 0.185	0.3
overall quality of sperm cells, billion	1.95 ± 0.03	1.07 ± 0.11	+ 0.88	7.8

of offspring for further use in selection. The new data, obtained by us, and our conclusions were confirmed by other scientists as well [8, 11, 15].

To study the possibilities of limiting the negative impact of intrasexual selection on the growth, development, physiological state and sperm production of bull-calves during the period of sexual maturation, the experiment was conducted in which one group of bull-calves, aged from 6 to 18 months (12 animals) were kept tethered, and the other (12 animals) – non-tethered, *i.e.* not limiting natural selection factors.

During the experiment the live weight of bull-calves in both groups exceeded the requirements for elite-record class animals. At the age of 18 months the difference in live weight between bull-calves of experimental groups was 30 kg, registered for bull-calves which were kept tethered (480 vs 450 kg respectively) ($P < 0.1$).

The use of feeds to keep animals tethered was 2.7 thousand feed units, while 3.0 thousand feed units were spent for the non-tethered group, which is 12.0 % more. Almost all the body measurements and composition indexes for 18-month-old non-tethered bull-calves were higher than those for tethered animals. The results of animal slaughter (three bull-calves from each group) demonstrated that the tethered animals had only 2.5 % less weight of muscles (with fat) and tendons and 1.5 % less weight of spleen compared to non-tethered bull-calves. At the same time they exceeded their non-tethered analogues in the mass of bones by 4.0 %, in the mass of heart – by 7.0 %, lungs – by 21.0 %, stomach – by 6.0 %.

The development of internal organs, conditioned by different influence of natural selection factors, was evidently manifested in the growth gain of the main tissues which ensures higher viability of non-tethered bull-calves [15].

The study of gas exchange in bull-calves during sexual arousal and ejaculation demonstrated (Table 4) that its restoration to the initial level in tethered bull-calves ended a day after, while it took non-tethered bull-calves 3–4 hours.

It is known that sperm cells, naturally kept in the tail of epididymis, are in conditions of decreased oxygen pressure, where oxygen content in freshly obtained sperm without any air contact is 200 %, and after dilution with synthetic medium – 325 % (relative). Thus, oxygen in sperm is spent 6 h later, which leads to the decrease in redox-potential and oxidation-reduction index. It is possible to restore oxygen content in such sperm only using artificial oxygenation. On the contrary, in hydrogen culture medium the oxidation-reduction regime in sperm decreases rapidly [16].

The analysis of the gas exchange process and oxidation-reduction index in the sperm of bull-calves allowed determining for the first time the phenomenon of increased oxygenation of the organism (including sexual sphere) of males during sexual arousal and ejaculation, targeted at ensuring optimal starting conditions for mating with females.

Contrast conditions of keeping experimental bull-calves also influenced their mobility. The calculation of steps demonstrated that the number of stepping motions for bull-calves of the experimental group per day was 16 % higher than that of the control.

The study of the orientation and conditioned reflexes demonstrated that the reaction of animals to external factors depended on previous conditions of their keeping. Inadequate reaction of bull-calves to external factors was the main reason of their active defensive, and for some – even aggressive, behavior, conditioned by

Table 4. Gas-energy exchange in bull-calves before and after sperm sampling (% to average daily level)

Group	Released CO_2, l		Consumed CO_2, l		Heat production, kcal	
	Age at the moment of sampling, months					
	12	18	12	18	12	18
Experimental non-tethered group:						
before sampling	208	140	171	140	182	141
after sampling	191	133	171	125	178	126
Control tethered group:						
before sampling	240	212	208	183	215	184
after sampling	210	231	193	207	196	207

parabiotic balanced or inhibiting phases of the activity of brain cortex. Objective evaluation of typological specificities of the nervous system and composition type of Schwyz bull-calves allowed determining that 63.9 % of animals with strong types of HNA belonged to broad-bodied and 36.1 % – to shallow-bodied composition types. Thus, the culling of rearing bull-calves of undesired composition types prior to evaluating potential productive features of offspring will allow leaving only animals of the desired type of the nervous system for breeding.

To determine the connection between typological properties of the nervous system of animals and the change in the number of eosinophils, the impact of adrenocorticotropic hormone (ACTH) on the system of hypophysis-suprarenal complex and sperm production of experimental bull-calves with different types of the nervous system was studied (Table 5).

The changes in the morphological picture of blood depended on functional status of the system of hypophysis-suprarenal cortex of bull-calves, grown in different conditions of keeping and at the impact of intrasexual selection. The morphological state of blood in bull-calves of different types of HNA also depended on the functional state of adrenohypophysis system. The comparison of the stress reaction to the introduction of ACTH in bull-calves of weak and strong types of HNA demonstrated that it depended on typological properties of the nervous system, which correlated positively with the rate of exhausting suprarenal cortex.

The stress, artificially caused by ACTH introduction (Table 6), resulted in the decrease in sperm indices of all the bull-calves regardless of the type of their nervous system. There was especially notable (2-fold) decrease in the index of activity (motility) of the sperm of bull-calves after the introduction of ACTH.

Thus, the index of sperm cell activity in freshly obtained (native) sperm may indeed be used to evaluate stress resistance and stress sensitivity of bulls.

Due to the impact of intrasexual selection, sperm indices and number of eosinophils in peripheral blood of bull-calves of the experimental group actually reflected their hierarchical interaction in the herd. Therefore, compared to the leader of the herd, dominant bull-calves of the experimental group ejaculated 45.6 % fewer and subordinates – 82.3 % fewer sperm cells. The reliable difference between dominant and subordinate bull-calves, favorable to the former, was noted in terms of ejaculate volume and sperm activity.

Table 5. Content of eosinophils and non-mature forms of neutrophils before and after ACTH introduction (%, average data)

Type of nervous system	Group	Eosinophils			Neutrophils		
		Before ACTH introduction	After ACTH introduction, h		Before introduction of ACTH	After ACTH introduction, h	
			4	9		4	9
Weak	Control	10.5	6.0	12.5	6.2	12.2	8.7
	Experimental	3.0	1.0	4.5	2.5	8.2	9.5
Strong	Control	5.8	5.0	2.8	6.5	11.5	14.3
	Experimental	2.7	3.2	2.7	9.2	16.2	20.2

Table 6. Sperm indices of bull-calves before and after ACTH introduction

Index	Before introduction	After introduction	Statistical reliability of td difference at $P < 0.01$
Number of bulls	5	5	–
Number of ejaculates	7	12	–
Sperm indices:			
volume, ml	2.10 ± 0.18	1.77 ± 0.25	1.28
activity, points	6.00 ± 0.01	3.00 ± 0.50	6.00
concentration, billion/ml	0.61 ± 0.12	0.70 ± 0.11	0.69
total number of sperm cells, billion	1.28	1.24	–

There were 25.4 % more flawed ejaculates among subordinate bull-calves compared to the leader bull, and 20.4 % more compared to the dominant bull-calves of the herd. At the same time the bull-calves of the control group with tethered keeping had 0.6 ml bigger volume of ejaculate compared to the leader bull, and 1.7 ml bigger – compared to dominant and subordinate bull-calves. In terms of the total number of sperm cells in all the ejaculates, the bull-calves of the control group exceeded dominant ones by 34.2 % and subordinate bull-calves of the experimental group – by 79.7 %. However, the leader bull of the experimental group exceeded bull-calves of the control one in terms of this index by 19.8 %. All the presented differences are statistically reliable.

Sperm indices of experimental bull-calves for the whole period of keeping (Table 7) were also different for the control and experimental groups.

New data, obtained by us, demonstrate that domination of some bull-calves of the experimental group was conditioned by the impact of intrasexual selection and had objective influence on growth, development, sexual activity, and sperm production of all the bull-calves of the herd.

Thus, the fertility of animals, closely connected to the hierarchical rank, is the main link of multi-stage natural regulation of intrasexual selection, which takes place via complicated complexes of behavior, physiological adaptation reactions and stress, which condition regular system of adaptations to environment with normal level of sexual system functioning for each animal of the herd.

CONCLUSIONS.

Due to the impact of sexual selection in conditions of natural reproduction via group tethered and non-tethered keeping (with feeding in circular corridors), the main regularity is the decrease in libido (up to impotence), erections, prostate-testicular interaction and sperm production in subordinate bulls as a result of pain, fear, and aggressiveness of animals. These states may be eliminated using different ways of keeping and stimulating, which ensure rational use of breeding bulls in conditions of modern enterprises.

The ability of animals to reproduce depends on their place in herd hierarchy. Higher calving indices of dominant cows compared to subordinate cows of the herd (85.9 vs 53.8 %) ensure higher representation of their offspring in the next generation.

The main regularity of forming and maintaining the relations between bull-calves (hierarchy) in the herd is the occurrence of complicated complexes of behavior and adaptation reactions and reaction of stress as well as phenomenon of relative extinction of selective component of libido to animals of the same group, which was restored in the formed herd towards “new” animals.

It was established that there was an important quality of the hierarchical rank of bull-calves of the herd – being determined by the type of the nervous system – which was relevant for selection, evaluation, and use in selection. Dominant bull-calves of strong balanced flexible type of higher nervous activity with higher ability of extrapolation became leaders of herds.

There were most evident contrast differences in the ways of restricting the impact of intrasexual selection at the age of 18 months, when bull-calves of the experimental group (non-tethered keeping) ceded to their control analogues (tethered keeping) in the live weight by 50 kg ($P < 0.1$) and in the total number of sperm cells in the ejaculates – the bull-calves of the control group exceeded dominant bull-calves of the experimental group by 34.2 and subordinate ones – 79.7 %.

The regularity of inadequate reaction of bull-calves, kept tethered or in conditions of restricting intrasexual selection, to external factors was first proven, which became the main reason of their active defensive and

Table 7. Sperm indices of experimental bull-calves, aged 12 to 18 months $\overline{X} \pm \overline{x}$

Index	Group		Difference
	Control	Experimental	
Number of ejaculates	224	160	64
Volume, ml	1.97 ± 0.06	1.37 ± 0.06	0.60
Activity, points	0.7	0.7	–
Concentration, billion/ml	0.735 ± 0.03	0.747 ± 0.04	0.012
Total number of sperm cells, billion	1.446 ± 0.09	1.021 ± 0.01	0.425

aggressive behavior, caused by balanced and inhibiting phases of the activity of brain cortex. The restoration of reactions to external factors to the norm was observed in bull-calves only after their transfer to the group non-tethered keeping and the formation of hierarchy.

Objective evaluation of HNA type and composition type of Schwyz bull-calves allowed first determining that 63.9 % of animals with strong types of HNA belonged to broad-bodied and 36.1 % – to shallow-bodied composition types. It facilitates the selection of the desired type for the purpose of evaluation and use in selection considerably.

Вплив природного відбору на відтворювальну функцію бугаїв

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Мета. Дослідити вплив природного відбору бугаїв при безприв›язному і прив'язному утриманні на їхню відтворювальну функцію. **Методи.** Спостереження за групами-аналогами при різних умовах утримання, але при однаковому рівні годівлі і використанні, етологічні методи поведінки тварин в стаді та «міні-стаді», зоотехнічні, генетико-математичні, біометричні та методи моделювання. **Результати.** Доведено, що домінуючі тварини в стаді пригнічують статеву здатність підлеглих, у яких погіршується лібідо, ерекція, якість сперми (аж до імпотенції). **Висновки.** Встановлено вплив природного відбору на статеву потенцію бугаїв-плідників при їхньому безприв'язному утриманні і етологічні особливості – при прив'язному, а також реакції організму залежно від типу конституції і тілобудови. Використання виявлених закономірностей спрощує роботу в селекції бугаїв при оцінці якості спермопродукції і забезпеченні підвищення кількості їхніх нащадків у наступних поколіннях.

Ключові слова: бугай, утримання, відтворення, сперма.

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