Dependence between behavioural acts and sperm parameters of boars of modern and local breeds of Ukraine



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Abstract This study aimed to investigate the relationship between behavioural activities and sperm parameters in modern and local breeds of Ukrainian boars. Visual observations were conducted on 30 boars, aged 12 and 24 months, with five boars from each of the following breeds: Large White, Landrace, Ukrainian Meat, Pietrain, intrabreed type of Duroc breed of Ukrainian selection "Steppovyi", and the terminal line "Maxter". Behaviours such as rest, movement, feed, and water intake during 24 hours were recorded. Semen samples were manually collected from each boar and evaluated for quantitative and qualitative indicators of sperm quality and fertilizing capacity according to the "Instructions for Artificial Insemination of Pigs", which included parameters such as ejaculate volume, sperm concentration in the ejaculate, percentage of correctly motile spermatozoa, survival of spermatozoa, and fertilizing ability of boars. The study found that certain behavioural activities significantly influenced the qualitative and quantitative indicators of sperm parameters in boars of different breeds. Specifically, time spent on rest and movement, as well as the index of movement activity (at 12 months of age), significantly (*P* < 0.05) influenced ejaculate volume, sperm concentration, and the percentage of correctly motile spermatozoa. Moreover, the effect of these behavioural acts on ejaculate volume had a curvilinear character. On the other hand, the survival of spermatozoa and fertilizing ability of boars were mainly determined by their time spent on feed and water intake (at 24 months of age), and the relationship detected in this case was asymptotic.

Keywords: age, artificial insemination, behavioural patterns, boar, breed, ejaculate traits

1. Introduction

The preservation of local breeds of pigs, such as the Ukrainian Meat intrabreed type of Duroc breed of Ukrainian selection Steppovyi, is crucial for the national culture of pig farming in Ukraine, both in the context of the ongoing war and in the post-war reconstruction of the country (Kramarenko et al 2018; Lugovoy et al 2018; Voitenko et al 2019). These breeds are well-adapted to the Ukrainian ecosystem and have great potential for increasing the genetic variability of Ukrainian pig herds. They are also characterized by excellent adaptability, high reproductive and fattening meat characteristics, and suitable technological parameters for the culinary processing of pork (Lykhach et al 2020).

However, due to the introduction of specialized imported breeds and the ongoing hostilities that have resulted in the aggressor country's shelling of many pig farms since February 24, 2022, the local breeds have suffered a substantial decrease (Jagtap et al 2022; Mamonova 2022). Therefore, researchers and pork producers in the country are

developing measures to improve the situation and preserve the population of local pig breeds in Ukraine.

Boars that produce high-quality ejaculates in the shortest duration possible are particularly valuable. This aspect can be the primary factor in inbreeding development and the improvement of swine utilization (Kondracki et al 2021). Boars can be evaluated based on quantitative and qualitative parameters of sperm, which are essential criteria for high-quality artificial insemination and have an economic effect on pork production (Kawęcka et al 2008; Poliana et al 2019). The accurate determination of the volume, concentration and percentage of correctly motile spermatozoa is essential for artificial insemination, and sperm motility is an important parameter of boar sperm (Kunowska-Slósarz and Makowska 2011; Oberlender et al 2012).

Researchers have noted that sperm production in boars depends on different factors, such as breed, breed combination, seasons, age, frequency of use of the boar, and behavioural patterns (Banaszewska and Kondracki 2012; Frydrychová et al 2011; Knecht et al 2017; Miclea et al 2016;

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Okere et al 2005; Smital 2010; Stančić et al 2003; Wolf and Smital 2009). Therefore, a comprehensive study of the characteristics of boar behaviour is an equally important component in improving the efficiency of the pig farming industry. Understanding and predicting the functional manifestation of productive traits through studying behavioural acts can increase these traits in industrial pork production technology conditions. However, limited search results are available on the dependence of behavioural acts and boar sperm indicators. In connection with this, the experiment aims to study the dependence of behavioural acts and sperm parameters of boars of modern and local breeds of Ukraine.

2. Materials and Methods

2.1. Experimental design

Experimental studies were conducted at breeding farms in Ukraine, including PJ-SC "Plemzavod "Stepnoy" in the Zaporizhzhya region, LLC "Tavriyski svini" in the Kherson region, and APC "Agrofirma "Mig-Service-Agro" in the Mykolayiv region, from 2020 to 2022. The boars were kept in individual pens with an area of 7 m2 on a concrete floor with thermal and moisture insulation. The technological conditions of feeding and keeping the boars at the farms were identical, and electronically controlled mechanical ventilation was used in the pig houses. The boars of different breeds selected for the experiments were clinically healthy. They were individually fed with a complete pelleted feed, "Eber", that had a nutritional value of 2.8-3.0 kg per head/day and consisted of 202.630 g/kg crude protein content and 12.406 MJ/kg exchange energy. The composition of 1 kg of granulated feed "Eber" produced by "PC "Alternative" LLC contained the following ingredients (%): corn (20.000), wheat (18.355), wheat bran (25.000), soybean cake (22.645), sunflower meal (10.000), and AminoMix Eber (4.000) according to the Technical Conditions of the State Standard of Ukraine 4508:2005. The boars were fed twice daily at 8:00 a.m. and 4:00 p.m. and had constant access to drinking water from the nipples of watering devices. The microclimate variables during the keeping of the boars corresponded to the Departmental Norms for Technological Design - Agro-Industrial Complex - 02.05 Pig-breeding enterprises (complexes, farms, small farms) at an air temperature of 17 °C (15-19 °C), relative air humidity of 40-75%, and air movement speed of 0.3-1.0 m/s.

2.2. Behaviour

During the experiment, Full HD 1080p video recorders were used (with a maximum resolution of 1920×1080, 30 frames/s) with AVI recording format to measure the timing of behavioural acts of the boars of different breeds. A total of 30 heads of behavioural acts (rest, movement, feed and water intake during 24 hours) were studied through visual observations, at the ages of 12 and 24 months, with five heads each of the following breeds: Large White, Landrace, Ukrainian Meat, Pietrain, intrabreed type of Duroc breed of

Ukrainian selection Steppovyi and the terminal line Maxter. The assessment of the behavioural acts of each breed of boars was measured in absolute terms as the number of minutes spent on the implementation of a particular behavioural indicator during the day. The index of movement activity was calculated by integrating the time calculation for movement acts of behaviour: IMA = $\Delta T/T$, where IMA is the index of movement activity, ΔT is the time of the act of behaviour, and T is the total observation time.

2.3. Performance data

Semen sampling from boars was manually conducted on a phantom. The quantitative and qualitative parameters of the sperm and fertilizing capacity were then assessed based on the Instructions for Artificial Insemination of Pigs. After filtration of the gelatinous fraction, the ejaculate volumes were measured using electronic scales to determine the ejaculate weight. Sperm concentration in the ejaculates was determined using a photometric method with a spectrophotometer (V-120). The percentage of correctly motile spermatozoa was identified through microscopic examination. A drop of fresh semen was placed on a MICRO med slide (XS-5520) heated to 37 °C, and the percentage of correctly motile spermatozoa was determined in the overall number of spermatozoa in the field of vision at a 200-fold zoom. The survival of spermatozoa was determined in a thermostat at 38°C after three hours. The fertilizing ability of boars was determined by counting the number of sows that were inseminated and qualitatively fertilized (%).

2.4. Statistical analysis

The data were analyzed using Statistica 12.0 (StatSoft Inc., 2014, www.statsoft.com). The results are presented as mean \pm standard deviation (X \pm SD). The study used the following significance levels: P < 0.05, 0.01, and 0.001. The coefficient of determination (R²) was applied to determine the dependence of parameters based on the generally accepted formula.

3. Results and Discussion

Table 1 shows the duration of behavioural acts of boars of different breeds at various ages. Video observations revealed that Maxter terminal line boars, selected for improved fattening and meat traits, exhibited greater movement at 12 months of age (479.5 min) (F = 2.73; df1 = 6; df2 = 30; P = 0.043) and at 24 months of age (439.2 min) (F = 2.63; df1 = 6; df2 = 30; P = 0.049), compared to other boar breeds. Signoret et al (1975) also noted that boars increase the period of rest during their lifespan (from 12 to 24 months), move less, take a sitting position, and lie more on their stomachs.

Table 2 shows the ejaculate traits obtained from boars of different breeds. The data indicate that the ejaculate traits changed depending on the boar's breed. Boars of the Ukrainian Meat breed produced ejaculates with the highest utility, with the largest volume (295.3 mL), allowing for the

preparation of the most insemination doses. Sperm concentration in the ejaculate was dominated by the intrabreed type of Duroc breed of Ukrainian Steppovyi (305.4 million/mL), indicating a breed peculiarity. This genetically perpetuated trait of the breed was confirmed by the results of this study and numerous others (Smital et al 2004; Wysokińska and Kondracki 2014). Boars of the Pietrain (97.1%) and Maxter (97.0%) breeds had the highest percentage of high motile spermatozoa compared to other

breeds studied. Landrace boars exhibited the longest survival time of spermatozoa (70 hours), maintaining their high fertilizing ability at 81.2%, compared to other boar breeds. Similar results were obtained by Kawęcka et al (2008), Kondracki et al (2013), Kunowska-Slósarz and Makowska (2011), Savić and Petrović (2015), Smital (2010), Szostak and Sarzyńska (2011), Wolf and Smital (2009), and Zapryanova and Malinova (2019).

Table 1 The duration of behavioural acts of boars of different breeds depending on age, min, $X \pm SD$

Behavioural act	Age	Breed or line, (n = 5)						. F	Dualua
	(months)	LW	UM	L	Р	DUSS	М	– F	<i>P</i> -value
Rest	12	940.3	930.2	908.6	904.3	984.9	881.3	3.45	0.017
		±24.61	±16.80	±17.72	±18.70	±21.83	±14.65		
	24	1049.8	1052.6	1031.0	1044.0	1068.5	963.4	0.72	0.612
		±42.30	±37.44	±58.22	±46.00	±39.55	±34.81		
Movement	12	418.9	434.4	449.2	459.6	406.1	479.5	2.73	0.043
		±16.22	±14.65	±18.90	±14.38	±20.81	±11.20		
	24	354.3	362.2	385.6	378.2	339.8	439.2	2.63	0.049
		±22.32	±25.10	±21.88	±24.70	±19.20	±13.46		
Feed and water	12	82.0	75.4	82.2	76.1	49.0	79.2	2.62	0.050
		±5.60	±9.40	±6.80	±8.60	±8.20	±7.4		
intake	24	35.9	25.2	23.4	17.8	31.7	37.4	2.62	0.050
	24	±4.50	±4.78	±5.19	±4.21	±4.82	±4.96		
Index of	12	0.29	0.30	0.31	0.32	0.28	0.33	-	-
movement activity	24	0.25	0.25	0.27	0.26	0.24	0.30	-	-

N= number; F= dispersion relation; p= level of significant; LW=Large White; UM= Ukrainian Meat; L= Landrace; P= Pietrain; DUSS= intrabreed type of Duroc breed of Ukrainian selection "Steppovyi"; M= terminal line "Maxter".

Table 2 Ejaculate traits obtained from boars of different breeds ($X \pm SD$).

Breed or line	Res	earched	Ejaculates volume	Sperm concentration	Correctly motile	Survival of spermatozoa	Fertilizing	
	boars	ejaculates	(mL)	(million/mL)	spermatozoa (%)	(hours)	ability (%)	
LW	3	80	270.2	288.8	96.0	64	79.4	
	3		±2.20	±3.30	±0.52	±0.74		
UM 4	4	80	295.3	290.4	95.7	68	80.5	
	4	80	±3.60	±3.00	±0.56	±1.20		
L 3	2	40	280.6	285.2	96.7	70	81.2	
	3	40	±2.60	±2.64	±0.40	±1.00		
P 3	40	250.8	272.3	97.1	50	72.4		
	3	40	±3.10	±2.82	±0.44	±1.20	12.4	
DUSS 2	40	224.2	305.4	95.4	66	80.2		
	2	40	±2.20	±3.20	±0.30	±1.34	60.2	
М	2	40	221.6	280.8	97.0	65	78.0	
	2		±2.80	±3.10	±0.36	±1.18		
F			116.94	13.41	2.64	39.84	-	
<i>P</i> -value			< 0.001	< 0.001	0.049	< 0.001	-	

F= dispersion relation; p= level of significant; LW=Large White; UM= Ukrainian Meat; L= Landrace; P= Pietrain; DUSS= intrabreed type of Duroc breed of Ukrainian selection "Steppovyi"; M= terminal line "Maxter".

The timing observations of boars of modern and local breeds of Ukraine allowed the establishment of a significant impact of behavioural acts on quantitative and qualitative indicators of sperm. A dependence between the duration of rest (at 12 months) and the ejaculate volume of boars of different breeds were established (Figure 1). Maxter boars had the smallest rest duration (881.3 min.), and their ejaculate volume was also the smallest (221.6 mL). With

increased resting time, the ejaculate volume increased, reaching its maximum in Landrace and Ukrainian Meat breeds (280.6 mL and 295.3 mL, respectively). However, further duration of the resting period negatively affected the ejaculate traits of boars, as seen in the example of the intrabreed type of Duroc breed of Ukrainian selection Steppovyi animals, where the volume of the ejaculate was low (224.2 mL) at the maximum rest period (984.9 min).

Generally, the dependence between the duration of rest (at the age of 12 months) and the ejaculate volume of boars of different breeds can be described by a second-degree polynomial with a determination level of $R^2 = 0.856$ (P < 0.05).

A similar pattern was observed regarding the relationship between the duration of movement (at 12 months old) and ejaculate volume in boars of different breeds (Figure 2). The shortest movement duration (at 12 months of age) was noted in the boars of the intrabreed type of Duroc breed of Ukrainian selection Steppovyi, which was accompanied by low ejaculate volume (224.2 mL). The breeds of Ukrainian Meat and Landrace spent an average of 434.4-449.2 min in movement, but at the same time, they were characterized by the highest ejaculate volume (280.6-295.3 mL). High ejaculate volume was also noted in Landrace boars of different selections in research conducted by Nacu et al (2011), Savić and Petrović (2015), and Zapryanova and Malinova (2019). An increase in the duration of movement by animals at this age led to a decrease in the ejaculate traits of boars, as can be seen in the example of animals of the Maxter breed, for which, at the maximum duration of movement (479.5 min), the ejaculate volume was very low (221.6 mL). In general, the dependence between the time of movement (at the age of 12 months) and the ejaculate volume of boars of different breeds is also described by a second-degree polynomial with a determination level of $R^2 = 0.874$ (P < 0.05).

As expected, a curvilinear dependence was observed between the index of movement activity (at the age of 12 months) and ejaculate volume in boars of different breeds (Figure 3). The boars of the intrabreed type of Duroc breed of Ukrainian selection Steppovyi and Maxter breeds, at the highest (0.28) and (0.33) of the index of movement activity, respectively, showed low mean ejaculate volumes (224.2 mL and 221.6 mL, respectively), which is consistent with research by Savić and Petrović (2015), who found the lowest ejaculate volume in Duroc boars of 102.55 mL. On the contrary, boars with an average manifestation of the index of movement activity for the group - 0.29-0.31 (Large White, Landrace, Ukrainian Meat) showed the maximum expression of ejaculate volume in the range of 270-295 mL. In general, the dependence between the index of movement activity of boars of different breeds (at the age of 12 months) and ejaculate volume is adequately described by a second-degree polynomial with a determination level of $R^2 = 0.948$ (P < 0.05).

Significant positive dependence between sperm concentration in ejaculates and duration of rest was observed in boars of different breeds at the age of 12 months (r = 0.880; n = 6; P < 0.05) (Figure 4). The researched breeds, Maxter, Pietrain, and Landrace, had a short resting period (881.3-980.6 min) at this age and the lowest sperm concentration in ejaculates (272.3-285.2 million/mL). Meanwhile, the Duroc breed of Ukrainian Steppovyi had the longest resting period on average (984.9 min) and the highest sperm concentration in ejaculates of 305.4 million/mL. The sperm concentration in ejaculates increases by almost 8 million/mL on average, increasing boars' resting duration by 30 minutes.

A negative correlation between sperm concentration in ejaculates and duration of movement was established for the studied breeds of animals at the age of 12 months (r=0.825; n=6; P<0.05) (Figure 5). The Pietrain and Maxter breeds spent the most time on movement (459.6 min and 479.5 min, respectively) and had the lowest sperm concentration in ejaculates (272.3 million/mL and 280.2 million/mL, respectively). In contrast, the Duroc breed of Ukrainian selection Steppovyi spent an average of 406.1 minutes on movement, and their sperm concentration in ejaculates was the highest (305.5 million/mL). The sperm concentration in ejaculates decreases by almost 10 million/mL on average, increasing boars' movement duration by 30 minutes.

The sperm concentration in the ejaculates of boars from different breeds displays a curvilinear dependence on the feed and water intake duration (at 24 months), as depicted in Figure 6. Thus, when the duration of feed and water intake is increased from 17.8 minutes (in Pietrain boars) to 31.7 minutes (in the intrabreed type of Duroc breed of Ukrainian selection Steppovyi), the sperm concentration in the ejaculates rises from 272.3 to 305.5 million/mL. However, further increases in the feed and water intake duration, with the maximum values found in Large White and Maxter breeds (35.9 and 37.4 min, respectively), resulting in a decrease in the average sperm concentration in the ejaculates to 288.8 million/mL and 280.8 million/mL, respectively. The dependence between the feed and water intake duration from different breeds (at the age of 24 months) and the sperm concentration in the ejaculates is approximated by a second-degree polynomial with a determination level of R² = 0.779 (P < 0.05).

A linear inverse trend in the sperm concentration in the ejaculates of boars from different breeds was observed with an increase in the index of movement activity (at 12 months old), as shown in Figure 7. Boars of the intrabreed type of Duroc breed of Ukrainian selection Steppovyi are characterized by the lowest level of the index of movement activity (0.28) and, accordingly, a high sperm concentration in the ejaculates (305.4 million/ml). Conversely, in animals of the Pietrain and Maxter breeds, the opposite trend was noted, with the lowest sperm concentration in the ejaculates observed in animals with the maximum manifestation of the index of movement activity (0.32 and 0.33, respectively, ranging from 272.3 to 280.8 million/mL). Thus, a negative correlation was found between the index of movement activity and sperm concentration in the ejaculates of boars from different breeds (r=- 859; n=6; P < 0.05). On average, a 0.05-unit increase in the index of movement activity results in a decrease in the sperm concentration in the ejaculates by nearly 25 million/mL.

A linear reverse trend was observed in the percentage of correctly motile spermatozoa in boars of different breeds, with an increase in the duration of rest (at 12 months old) (Figure 8). Boars of the Maxter, Pietrain, and Landrace breeds had the shortest resting period (averaging 881.3-908.6 minutes), characterized by the highest percentage of

correctly motile spermatozoa (96.7-97.1%). Conversely, animals with the lowest percentage of correctly motile spermatozoa (intrabreed type of Duroc breed of Ukrainian selection Steppovyi) spent a lot of time resting (984.9 minutes). Thus, a negative correlation was found between

the percentage of correctly motile spermatozoa and the duration of rest for the studied breeds of animals (r= -0.901; n=6; P < 0.05). On average, with an increase in the rest duration for the boars by 30 minutes, the percentage of correctly motile spermatozoa decreased by 0.5%.

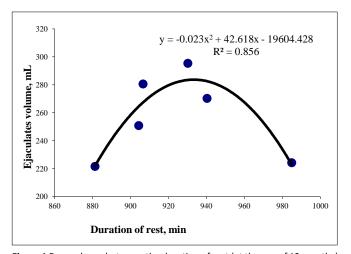


Figure 1 Dependence between the duration of rest (at the age of 12 months) and the ejaculates volume of boars of different breeds.

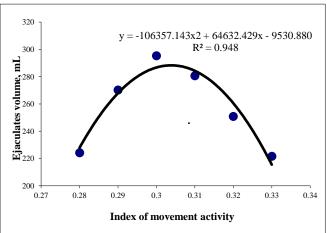


Figure 3 Dependence between the index of movement activity (at the age of 12 months) and the ejaculates volume of boars of different breeds.

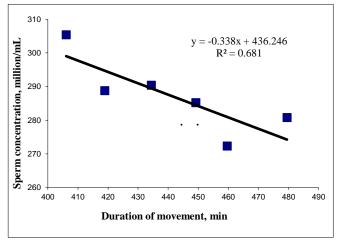


Figure 5 Dependence between the duration of movement (at the age of 12 months) and sperm concentration of boars of different breeds.

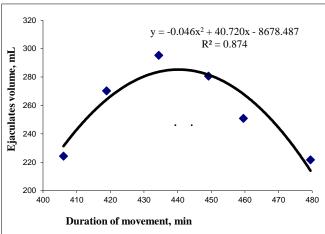


Figure 2 Dependence between the duration of movement (at the age of 12 months) and the ejaculates volume of boars of different breeds.

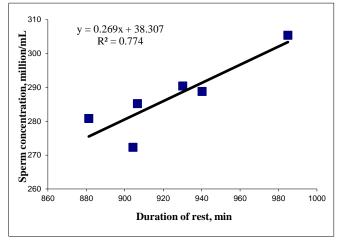


Figure 4 Dependence between the duration of rest (at the age of 12 months) and sperm concentration of boars of different breeds.

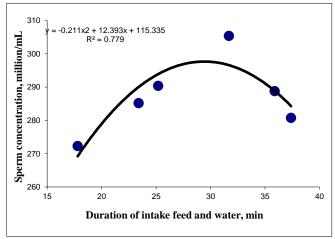


Figure 6 Dependence between the duration of intake feed and water (at the age of 12 months) and sperm concentration of boars of different breeds.

On the other hand, the percentage of correctly motile spermatozoa was positively associated with the duration of movement in boars of different breeds (at 12 months old) (Figure 9). Boars that spent less time moving (intrabreed type of Duroc breed of Ukrainian selection Steppovyi, Large White, Ukrainian Meat) had the lowest percentage of correctly motile spermatozoa (95.4-96.0%). Conversely, animals of the Pietrain and Maxter breeds, characterized by the highest percentage of correctly motile spermatozoa (97.0-97.1%), spent more time moving (averaging 459.6-479.5 minutes). Thus, a significant positive correlation was found between the percentage of correctly motile spermatozoa and the movement duration for the studied breeds of animals (r=0.903; r=6; P<0.05). On average, with an increase in the movement duration of

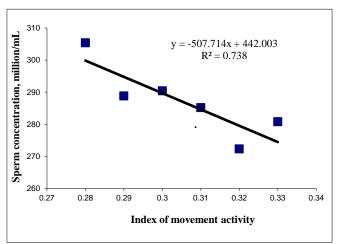


Figure 7 Dependence between the index of movement activity (at the age of 12 months) and sperm concentration of boars of different breeds.

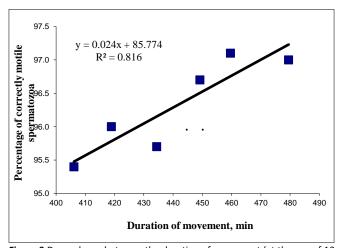


Figure 9 Dependence between the duration of movement (at the age of 12 months) and percentage of correctly motile spermatozoa of boars of different breeds.

The correctly motile spermatozoa (%) is significantly affected by the index of movement activity of boars of different breeds (at 12 months of age) (see Figure 11). Boars with the lowest index of movement activity (intrabreed type of Duroc breed of Ukrainian selection Steppovyi, Large White, Ukrainian Meat) were characterized by the lowest

boars by 30 minutes, the percentage of correctly motile spermatozoa increased by 0.7%.

The percentage of correctly motile spermatozoa is also associated with the time spent on boar movement (at 24 months of age), but this relationship is non-linear (see Figure 10). An increase in the percentage of correctly motile spermatozoa from 95% to 97% in boars is observed with an increase in movement time from 340 to 380 minutes. However, further increases in movement time do not improve the quality of sperm parameters. In general, the relationship between the duration of movement in boars of different breeds and the percentage of correctly motile spermatozoa can be adequately described by a second-degree polynomial with a determination level of $R^2 = 0.813$ (P < 0.05).

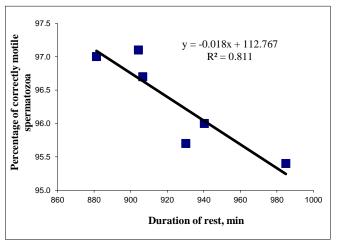


Figure 8 Dependence between the duration of rest (at the age of 12 months) and percentage of correctly motile spermatozoa of boars of different breeds.

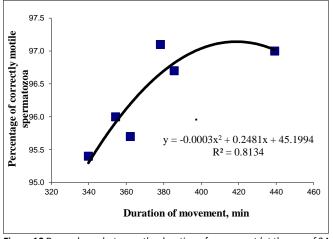


Figure 10 Dependence between the duration of movement (at the age of 24 months) and percentage of correctly motile spermatozoa of boars of different breeds.

percentage of correctly motile spermatozoa (95.4-96.0%). In contrast, animals of the Pietrain and Maxter breeds were characterized by high values of the index of movement activity (0.32-0.33) and the highest percentage of correctly motile spermatozoa (97.0-97.1%). Thus, a significant positive correlation was found between the percentage of correctly

motile spermatozoa and the index of movement activity for the studied breeds of animals (r=0.848; n=6; P < 0.05). On average, the percentage of correctly motile spermatozoa increased by 1.8%, with an increase of 0.05 units in the index of movement activity.

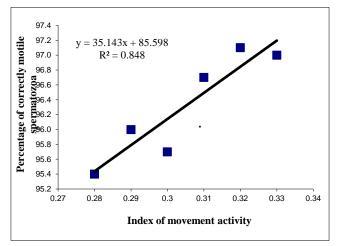


Figure 11 Dependence between the index of movement activity (at the age of 12 months) and percentage of correctly motile spermatozoa of boars of different breeds.

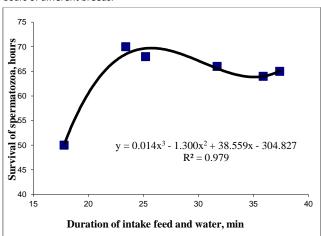


Figure 12 Dependence between the duration of intake feed and water (at the age of 24 months) and survival of spermatozoa of boars of different breeds.

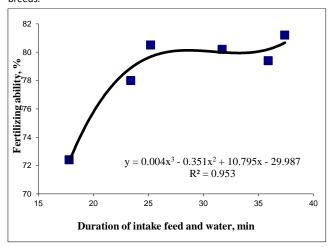


Figure 13 Dependence between the duration of intake feed and water (at the age of 24 months) and fertilizing ability of boars of different breeds.

A non-linear relationship was observed between the survival of spermatozoa and the duration of feed and water intake by boars at the age of 24 months, as shown in Figure 12. A similar relationship was noted between the fertilizing ability of sperm and the duration of feed and water intake by boars at the same age, as depicted in Figure 13. When boars' feed and water intake increased from an average of 17 to 25 minutes, the survival of spermatozoa increased from 50 to 70 hours, while their sperm's fertilizing ability rose from 72 to 80%. However, further increases in the duration of feed and water intake did not have an impact on either the survival of sperm or their fertilizing ability. Overall, the dependence between the feed and water intake duration by boars of different breeds and the survival of spermatozoa and their fertilizing ability could be approximated by a third-degree polynomial with a determination level of $R^2 = 0.953-0.979$ in both cases (P < 0.05).

4. Conclusions

The qualitative and quantitative parameters of boar sperm in modern and local breeds of Ukraine were significantly influenced by behavioural acts. The index of movement activity was found to be significantly dependent on the ejaculate volume, sperm concentration in the ejaculate, and percentage of correctly motile spermatozoa. These factors should be considered in breeding and commercial farms to improve the artificial insemination of pigs.

Ethical considerations

The requirements for animal treatment in the experiment fully complied. The conditions for feeding, watering, housing, care, prevention and treatment were organized following European legislation on the protection of animals and their comfort, including Council Directive 2008/120/EU - On the establishment of minimum standards for the protection of pigs of December 18, 2008, European Parliament and of the Council 2010/63/EU - On the protection of animals used for scientific purposes of September 22, 2010, and the Order of the Ministry of Economy of Ukraine - On approval of the requirements for the welfare of farm animals during their housing of February 18, 2021. The experimental protocol on Bioethics of the National University of Life and Environmental Sciences of Ukraine was approved by the local Commission.

Conflict of interest

The authors declare that they have no conflict of interest.

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