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CORRELATION OF FREQUENCY OF SPERMATOZOA MORPHOLOGICAL ALTERATIONS WITH SPERM CONCENTRATION IN EJACULATES OF POLISH LANDRACE BOARS

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The experiments were performed on 448 ejaculates obtained from 41 Polish Landrace boars. Ejaculates collected from each boar at one-month intervals for approximately 10 months were analysed. Sperm morphometric measurements were taken from each boar and assessment of semen morphology was done on the basis of examination under a microscope of preparations made from fresh ejaculates. The ejaculates were classified based on the criterion of sperm concentration and divided into three groups. An attempt was made in the present study to assess the correlation of ejaculate parameters, morphological sperm alteration incidence and morphometric sperm parameters with the sperm concentration in ejaculates of Polish Landrace boars. It should be stated that morphometric traits of spermatozoa are related to sperm concentration. The spermatozoa in concentrated ejaculates had smaller heads than the spermatozoa in the ejaculates with lower sperm concentrations. This can mean that the high fertility of males that produce highly concentrated semen does not only result from a high sperm concentration, but also from the fact that the spermatozoa in such ejaculates have smaller heads. The highest frequency of morphologically well-formed spermatozoa was identified in ejaculates with the sperm concentration ranging from 400 to 500 thousand/mm^3.

Key words: boar, ejaculate, sperm concentration

INTRODUCTION

Sperm generation varies in males of different species. It is conditioned both by genetic and environmental factors which affect the quality and quantity of generated semen (Holt *et al.,* 1997). The biological function of any spermatozoon consists in reaching and penetrating the egg cell. Only morphologically well-formed spermatozoa are capable of this. Studies conducted to date have proved that a large number of morphologically altered spermatozoa is a strong indicator of reduced fertility in men (Kruger *et al.,* 1993), stallions (Jasko *et al.,* 1990), bulls (Sekoni & Gustafsson, 1987) and goats (Chandler *et al.,* 1988). Many researchers have also analysed the influence of morphometric sperm parameters on male fertility. Some of their studies have revealed variability in morphometric sperm parameters in relation to sperm concentration in ejaculates of dogs (Rijsselaere *et al.,* 2004), stallions (Davis *et al.,* 1993) and boars (Banaszewska *et al.,* 2009, Kondracki *et al.,* 2011). There are also studies available that indicate differences in the dimensions and shapes of spermatozoa within one species, but in different breeds of boars (Kondracki *et al.,* 2005, Saravia *et al.,* 2007) Ejaculates of boars of different breeds also differ in their volume, sperm concentration, total number of spermatozoa in the ejaculates and, consequently, the number of insemination doses. The Polish Landrace is one of the most popular pig breeds in Poland. The boars of this breed produce good-quality ejaculates with considerable volumes and high sperm concentration, as well as with a high percentage of progressively motile spermatozoa. (Kondracki, 2003). An attempt was made in the present study to assess the correlation of ejaculate parameters, morphological sperm alteration incidence and morphometric sperm parameters with the sperm concentration in ejaculates of Polish Landrace boars.

MATERIALS AND METHODS

The study covered 448 ejaculates collected from 41 Polish Landrace boars used at an insemination centre. The ejaculates were collected manually (King & Macpherson, 1973). The analysed ejaculates were collected from each boar at one-month intervals for approximately 10 months.

The ejaculates were classified into the following three groups according to the criterion of sperm concentration in an ejaculate:

- group I - ejaculates with sperm concentration of less than 400 thousand/mm^3 (138 ejaculates),
- group II - ejaculates with sperm concentration of 400 - 500 thousand/mm^3 (158 ejaculates),
- group III - ejaculates with sperm concentration of more than 500 thousand/mm^3 (152 ejaculates).

Directly after sampling, the ejaculates were analysed for the following physical parameters: ejaculate volume, sperm concentration and the percentage of progressively motile spermatozoa per ejaculate. Ejaculate volumes were measured after isolating the gelatinous fraction. Sperm concentration in the ejaculates was determined with a photometric method using a Cassou spectrophotometer. Sperm motility was assessed under a microscope. Based on the results, the following calculations were made: the total number of progressively motile spermatozoa and the number of insemination doses to be obtained from one ejaculate. The total number of spermatozoa in the ejaculates and the number of insemination doses obtained from one ejaculate were calculated using WINSUL software.

Sperm morphometric measurements were taken from each boar and assessment of semen morphology was done on the basis of examination under a microscope of preparations made from fresh ejaculates. The method of slide preparation had been described in the previous work by Kondracki *et al*. (2006). The preparations were examined under the light microscope Nikon E-50i using 100x immersion lens. Morphological structure of 500 spermatozoa was evaluated in each slide, indicating the number of spermatozoa with appropriate morphology and morphologically changed ones which were further divided into forms with major and minor changes according to the Blom classification (1981). Additionally, for each slide, morphometric measurements were taken of 15 randomly selected spermatozoa characterised by normal morphology and well visible under the microscope. The measurements were done by means of a computer image analysis software (Screen Measurement v. 4.1, Laboratory Imaging S.r.o. LIM Czech Republic, Praha) following methodology prepared by Kondracki *et al*. (2005). A total of 1 125 measurements were taken. They included: sperm head length, sperm head width, sperm head perimeter, sperm head area, sperm tail length, total sperm length. The following parameters of sperm morphology were calculated on the basis of the results of the morphometric measurements.

Statistical analysis

Study results were statistically processed using the analysis of variance according to the following mathematical model:

$$Y_{ij} = \mu + a_i + e_{ij}$$

where:

Y_{ij} – trait value,
μ – population mean,
a_i – sperm concentration effect,
e_{ij} – error.

Differences between means were tested using the Tukey's test at $P\leq0.05$ and $P\leq0.01$.

RESULTS

Table 1 shows data for the physical parameters of the ejaculates in relation to sperm concentration in the ejaculates. The data indicate that ejaculate

Table 1. Physical traits of ejaculate depending on the sperm concentration in boar ejaculate

Item	Sperm concentration (x $10^3/mm^3$)		
	Group I < 400	Group II 400-500	Group III >500
Number of ejaculates	138	158	152
Sperm concentration (x $10^3/mm^3$)	345.88±51.32 ^{A}	457.82±31.95 ^{B}	571.58±46.89 ^{C}
Volume of ejaculate (ml)	283.85±88.02 ^{A}	267.72±87.23 ^{B}	240.66±91.07 ^{C}
Total number of progressively motile spermatozoa	76.81±4.68 ^{a}	77.59±4.29 ^{a}	77.37±5.24 ^{a}
Total number of spermatozoa in the ejaculates (mld)	75.71±23.11 ^{A}	94.82±32.75 ^{C}	105.53±39.06 ^{C}
The number of insemination doses	26.10±7.47 ^{A}	32.68±10.86 ^{B}	34.32±13.30 ^{B}

Different superscripts mean significant differences among means within particular rows; lower-case letters: $p \leq 0.05$. upper-case letters: $p \leq 0.01$

Table 2. Frequency of occurrence of spermatozoa morphologically changed depending on the sperm concentration in boar ejaculate

Item	Sperm concentration (x $10^3/mm^3$)		
	Group I < 400	Group II 400-500	Group III >500
Number of ejaculates	138	158	152
Sperm concentration (x $10^3/mm^3$)	345.88±51.32 ^{A}	457.82±31.95 ^{B}	571.58±46.89 ^{C}
Normal spermatozoa (%)	95.72±5.58 ^{ab}	96.21±4.04 ^{a}	94.71±8.76 ^{b}
Sperm with major abnormalities (%)	0.62±0.82 ^{a}	0.76±2.03 ^{a}	0.51±0.77 ^{a}
Sperm with minor abnormalities (%)	3.66±5.43 ^{abAB}	3.03±3.21 ^{aA}	4.84±8.68 ^{bB}

Different superscripts mean significant differences among means within particular rows; lower-case letters: $p \leq 0.05$. upper-case letters: $p \leq 0.01$

volume diminished as the sperm concentration in the ejaculates increased. The highest volumes were identified in the ejaculates with sperm concentrations below 400 thousand/mm^3. The volume of these ejaculates was over 16 mL higher than the volume of the ejaculates with the sperm concentration within 400 to 500 thousand/mm^3 and 27 mL higher than the volume of the ejaculates with the sperm concentration higher than 500 thousand/mm^3 (P≤0.01). The highest total number of spermatozoa in the ejaculates was identified in Group III in which the sperm concentration was higher than 500 thousand/mm^3. The mean number of spermatozoa in these ejaculates exceeded 105 billion and was approximately 10.7 billion spermatozoa higher than in Group II, and almost 30 billion spermatozoa higher than in the Group I (P≤0.01). The ejaculates with the sperm concentration higher than 500 thousand/mm^3 (Group III) also provided the most insemination doses, more than the Group I ejaculates by over 8 doses (P≤0.01).

Table 3. Morphometric traits of spermatozoa depending on the sperm concentration in boar ejaculate

Item	Sperm concentration (x 10^3/mm^3)		
	Group I < 400	Group II 400-500	Group III >500
Number of ejaculates	138	158	152
Sperm concentration (x10^3/mm^3)	345.88±51.32 ^{A}	457.82±31.95 ^{B}	571.58±46.89 ^{C}
Sperm head length (μm)	9.21±0.35 ^{a}	9.14±0.35 ^{ab}	9.12±0.35 ^{b}
Sperm head width (μm)	4.80±0.29 ^{aA}	4.73±0.28 ^{bAB}	4.70±0.27 ^{bB}
Sperm head perimeter (μm)	23.64±0.87 ^{aA}	23.30±0.96 ^{bB}	23.47±0.92 ^{abAB}
Sperm head area (μm^2)	40.86±2.58 ^{aA}	40.13±2.58 ^{bB}	40.38±2.39 ^{abAB}
Sperm tail length (μm)	45.29±1.38 ^{a}	45.30±1.46 ^{a}	45.18±1.58 ^{a}
Total sperm length (μm)	54.51±1.53 ^{a}	54.44±1.67 ^{a}	54.30±1.76 ^{a}

Different superscripts mean significant differences among means within particular rows; lower-case letters: p≤0.05. upper-case letters: p≤0.01

Data showing the correlation of morphological sperm alteration incidence with the sperm concentration in the ejaculates are presented in Table 2. The data reveal that the semen in Group II had the best morphology. More than 96.2% of spermatozoa in this semen had the correct morphological structure – 1.5% more than in the ejaculates classified in Group III (P≤0.05). The percentage of spermatozoa with morphological alterations was chiefly conditioned by the occurrence of spermatozoa with secondary defects. The highest frequency of secondary changes in spermatozoa (almost 5%) was observed in the ejaculates with the highest sperm concentration (Group III). In Group II, which had the ejaculate sperm concentration of 400–500 thousand/mm^3, in turn, the percentage of spermatozoa with secondary changes was 1.81% lower than in the Group III ejaculates which had the highest sperm concentration (P≤0.01).

Table 4. Indices sperm morphology depending on the sperm concentration in boar ejaculate

Item	Sperm concentration (x 10^3/mm^3)		
	Group I < 400	Group II 400-500	Group III >500
Number of ejaculates	138	158	152
Sperm concentration (x10^3/mm^3)	345.88 ±51.32 ^{aA}	457.82 ±31.95 ^{bB}	571.58 ±46.89 ^{cC}
Width to length ratio of sperm head x100	52.08±3.09 ^{a}	51.83±3.16 ^{a}	51.51±3.12 ^{a}
Ratio of head length to total sperm length x100	16.91±0.56 ^{a}	16.79±0.49 ^{b}	16.81±0.54 ^{ab}
Ratio of head length to sperm tail length x100	20.35±0.82 ^{a}	20.19±0.70 ^{a}	20.21±0.79 ^{a}
Ratio of tail length to total sperm length x100	77.10±18.55 ^{a}	81.19±11.15 ^{b}	78.21±17.07 ^{ab}
Ratio of sperm head perimeter to total sperm length x100	47.20 ±11.82 ^{aA}	44.12±7.35 ^{bB}	46.37±10.93 ^{aA}
Ratio of sperm had area to total sperm length x100	72.48±10.27 ^{a}	72.90±6.94 ^{a}	72.19±9.15 ^{a}
Ratio of sperm head length and width to total sperm length x100	78.01±12.76 ^{a}	78.43±8.39 ^{a}	76.25±11.05 ^{a}

Different superscripts mean significant differences among means within particular rows; lower-case letters: p≤0.05. upper-case letters: p≤0.01

Table 3 shows the dimensions of spermatozoa in relation to the sperm concentration in the ejaculates. The data reveal certain interrelations between sperm dimensions and the sperm concentration in the ejaculates. In the ejaculates with the lowest sperm concentration of below 400 thousand/mm^3 (Group I), the spermatozoa had larger heads than the spermatozoa in the ejaculates with higher sperm concentrations. In the ejaculates with the lowest sperm concentration (Group I) sperm heads were longer and broader than in the ejaculates with higher sperm concentrations ($P \leq 0.05$). Spermatozoa in Group I also had larger head perimeters and areas than the spermatozoa in the ejaculates with higher sperm concentrations ($P \leq 0.01$).

A relationship was also identified between the dimensional proportions of the spermatozoa and the sperm concentration in the ejaculates (Tab. 4). The spermatozoa in the ejaculates with lower concentrations (Group I) had a higher ratio of head length to total sperm length and, at the same time, a lower ratio of tail length to total sperm length than the spermatozoa in Group II and III ejaculates ($P \leq 0.05$) Additionally, the spermatozoa in Group II ejaculates, with the sperm concentration of 400–500 thousand/mm^3, had a slightly different shape, as evidenced in the lower ratio of head perimeter to total sperm length as compared with the spermatozoa in Group I and III ejaculates ($P \leq 0.01$).

DISCUSSION

The results of the present study have revealed a correlation of physical ejaculate parameters and morphological sperm parameters with the sperm concentration in the ejaculates. Differences were found in the quantitative ejaculate traits and morphological sperm parameters between ejaculates with different sperm concentrations. Ejaculates with the highest sperm concentration can be used to produce more insemination doses. At the same time, the ejaculates with the highest sperm concentration were observed for a greater number of spermatozoa with morphological anomalies which mainly resulted from a greater incidence of spermatozoa with secondary alterations. A positive correlation was identified between ejaculate sperm concentration and the percentage of morphologically well-formed spermatozoa and the percentage of pronuclei generated during extracorporeal insemination in humans (Morgentaler *et al.,* 1996; Azis *et al.,* 1998). The majority of researchers agree in the view that assessments of the fertilization potential of ejaculates should be supplemented with analyses of spermatozoon morphology (Kruger *et al.,* 1986; Lim *et al.,* 1998), as sperm morphology is closely associated with male fertility (Xu *et al.,* 1998; Alm *et al.,* 2006). It has also been found that the numerical strength of sow litters is correlated with the frequency of certain morphological changes in spermatozoa, including loose and malformed heads and acrosome defects (Sutkevičienė & Žilinskas, 2004; Frangeî *et al.,* 2005). Research done by Rijsselaere *et al*. (2004) suggests that the incidence of spermatozoa with morphological alterations is lower in ejaculates which have a lower sperm concentration, whereas a rise in ejaculate sperm concentration is accompanied with an increase in the number of morphologically anomalous spermatozoa. This may be associated with a higher sperm concentration in the semen outlet ducts where spermatozoa are stored and undergo necessary transformations for the process of fertilization. The present study revealed the highest percentage of spermatozoa with morphological alterations in ejaculates that had the highest sperm concentrations, which confirms the hypothesis of Rijsselaere *et al*. (2004). Abnormaly shaped sperm heads, mid-piece alterations and involuted tails undoubtedly reduce the insemination capacity of spermatozoa. The shape of the sperm head plays an essential role during the fusion of the spermatozoon with the zona pellucida of the oocyte (Szczygieł & Kurpisz, 1999). The use of deformed spermatozoa with head defects lowers the quality of embryos (De Jarnette *et al.,* 1992). Sperm head deformations may result from chromatin condensation disorders. This leads to a lowered fertilization capacity of spermatozoa.

The results of the present study reveal a relationship between the morphometric parameters of spermatozoa and the sperm concentration in the ejaculates. The spermatozoa in the less concentrated ejaculates had larger heads than the spermatozoa in ejaculates with higher sperm concentrations. This is confirmed by the results of studies of Pietrain (Banaszewska *et al.,* 2009) and Duroc boars (Kondracki *et al.,* 2011) and bulls (Kondracki *et al.,* 2012) which have demonstrated that spermatozoa have larger heads in ejaculates in which the sperm concentration is lower. On the other hand, analyses of dog semen have shown that ejaculates with lower sperm concentration contained spermatozoa that had shorter and narrower heads with smaller areas and perimeters in comparison with the spermatozoa in ejaculates with higher sperm concentrations (Rijsselaere *et al.,* 2004). An effect of sperm concentration on morphometric parameters of spermatozoa has also been observed in stallions (Davis *et al.,* 1993). Stallions that provided highly concentrated ejaculates were found to produce spermatozoa with smaller and shorter heads than those of the spermatozoa in the less concentrated ejaculates. Analyses of pbz boars have shown that the sperms in ejaculates containing numerous spermatozoa had smaller head dimensions as compared with the spermatozoa in ejaculates with a low sperm count (Wysokińska *et al.,* 2009). Other studies demonstrated that Duroc boar semen contained spermatozoa whose heads were larger and more elliptical as compared with the spermatozoa of other boar breeds (Saravia *et al.,* 2007). The identification of differences in sperm head dimensions makes it possible to more precisely evaluate sire spermiograms (Severa *et al.,* 2010). Additionally, on the basis of alterations of sperm heads it is possible to recognise fertile animals and those with limited fertility (Gravance *et al.,* 1996). Attempts at defining the fertility of males in relation to morphometric traits of spermatozoa have been made in the case of different animal species, e.g. horses (Casey *et al.,* 1997), pigs (Hirai *et al.,* 2001, Peña *et al.,* 2005), bulls (Ostermeier *et al.,* 2001), deer (Esteso *et al.,* 2006) or dogs (Núñez-Martínez *et al.,* 2007). Morphometric analyses of stallion and goat spermatozoa have revealed that the spermatozoa of males with reduced fertility have larger heads than the spermatozoa of fertile animals (Casey *et al.,* 1997; De Paz *et al.,* 2011). The fact is confirmed in studies of boars which show highly fertile boars to have spermatozoa with smaller and shorter heads than the boars which were found to be less efficient in fertilization (Hirai *et al.,* 2001). These data show sperm head dimensions to be associated with fertility of mammalian males – the smaller the sperm heads, the higher is the male's fertility. The present work demonstrated spermatozoa contained in highly concentrated ejaculates to have smaller heads. In the context of observations made by Hirai *et al*. (2001) this may signify that the higher fertility of boars that produce highly concentrated ejaculates does not only stem from the high sperm concentration but also from the fact that they generate spermatozoa with smaller head dimensions. The shape and size of the sperm head can also be associated with sperm motility. The results obtained by Gil *et al*. (2009) suggest that the spermatozoa with deformed heads were less motile, which could eventually lead to reduced fertilization capacity. Sperm motility is also affected by the length of the sperm mid-piece and tail. The spermatozoa contained in semen with reduced motility were found to have shorter tails than those contained in semen with a high percentage of progressively motile spermatozoa (Noorafshan and Karbalay-Doust, 2010). The present authors identified the longest tails in spermatozoa classified in the group with the lowest sperm concentration in the ejaculates. There is a likelihood that spermatozoa with longer tails may be more competitive in comparison with other spermatozoa and capable of reaching the egg cell faster.

In conclusion, it should be stated that morphometric traits of spermatozoa are related with sperm concentration. The spermatozoa in the more concentrated ejaculates had smaller heads than the spermatozoa in the ejaculates with lower sperm concentrations. This can mean that the high fertility of males that produce highly concentrated semen does not only result from a high sperm concentration but also from the fact that the spermatozoa in such ejaculates have smaller heads. A correlation was also observed between the indices of the morphological structure of spermatozoa and ejaculate sperm concentration. The highest frequency of morphologically well-formed spermatozoa was identified in ejaculates with the sperm concentration ranging from 400 to 500 thousand/mm^3.

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REFERENCES

1. *Alm K, Peltoniemi O, Koskinen E, Andersson M*, 2006, Porcine field fertility with two different insemination doses and the effect of sperm morphology, *Reprod Dom Anim,* 41, 210-13.
2. *Azis N, Buchan I, Taylor C,* 1996, The sperm deformity index: a reliable predictor of the outcome of oocyte fertilization in vitro, *Fertil Steril*, 66, 1000-8.
3. *Banaszewska D, Kondracki S, Wysokińska A,* 2009, Effect of sperm concentration on ejaculate for morphometric traits of spermatozoas of the pietrain breed boars, *J Cent Eur Agr,* 10, 383-96.
4. *Blom E,* 1981, The morphological estimation of the spermatozoa defects of bull II. The proposal of new classification of spermatozoa defects (in Polish), *Med Wet***,** 37, 4, 239-42.
5. *Casey PJ, Gravance CG, Davis RO, Chabot DD, Liu IKM,* 1997, Morphometric differences in sperm head dimensions of fertile and subfertile stallions. *Theriogenology*, 47, 575-82.
6. *Chandler JE, Painter CL, Adkinson RW, Memon MA, Hoyt PG,* 1988, Semen quality characteristics of dairy goats, *J Dairy Sci,* 71, 1638-46.
7. *Davis RO, Gravance CG, Casey PJ,* 1993, Automated morphometric analysis of stallion spermatozoa, *Am J Vet Res,* 54, 11, 1808-11.
8. *De Jarnette JM, Saake RG, Barne J, Volger CJ* , 1992, Accessory sperm: Their importance to fertility and embryo quality, and attempts to alter their numbers in artificially inseminated cattle, *J Anim Sci*, 70, 484-91.
9. *De Paz P, Mata-Campuzano M, Tizado EJ, Álvarez M, Álvarez-Rodríguez M, Herraez P et al.,* 2011, The relationship between ram sperm head morphometry and fertility depends on the procedures of acquisition and analysis used, *Theriogenology* 76, 1313-25.
10. *Esteso MC, Soler AJ, Fernández-Santos MR, Quintero-Moreno AA, Garde JJ,* 2006, Functional significance of the sperm head morphometric size and shape for determining freezability in Iberian red deer (*Cervus elaphus hispanicus*) epididymal sperm samples, *J Androl*, 27, 662-70.
11. *Frangeî R, Gider T, Kosec M*, 2005, Frequency of boar ejaculate collection and its influence on semen quality pregnancy rate and litter size, *Acta Vet Brno,* 74, 265-73.
12. *Gil MC, García-Herreros M, Barón FJ, Aparicio IM, Santos AJ, García-Marín LJ* , 2009, Morphometry of porcine spermatozoa and its functional significance in relation with the motility parameters in fresh semen, *Theriogenology*, 15, 71, 2, 254-63.
13. *Gravance CG, Liu IKM, Davis RO, Hughs JP, Casey PJ,* 1996, Quantification of normal stallion sperm-head morphometry, *J Reprod Fertil***,** 108, 41-6.
14. *Hirai M, Boersma A, Hoeflich A, Wolf E, Föll J, Aumüller R et al.,* 2001, Objectively measured sperm motility and sperm head morphometry in boars (Sus scrofa): Relation to fertility and seminal plasma growth factors, *J Androl*, 22, 104-10.
15. *Holt C, Holt WV, Moore HD, Reed HC, Curnock RM*, 1997, Objectively measured boar sperm motility parameters correlate with the outcomes of on-farm inseminations: results of two fertility trials, *J Androl,* 18, 312-23.
16. *Jasko DJ, Lein DH, Foote RH,* 1990, Determination of the relationship between sperm morphologic classifications and fertility in stallions: 66 cases (1987-1988), *J Am Vet Med Assoc*, 197, 389-94.
17. *King GJ, Macpherson J,* 1973, A comparison of two methods for boar semen collection, *J Anim Sci*, 36, 563-5.
18. *Kondracki S***,** 2003, Breed differences in semen characteristics of boars used in artifical insemination in Poland. *Pig News Inform,* 24, 4, 119-22.
19. *Kondracki S, Banaszewska D, Mielnicka C,* 2005, The effect of age on the morphometric sperm traits of domestic pig (*Sus scrofa domestica*), *Cell Mol Biol Lett,* 10, 1, 3-13.
20. *Kondracki S, Banaszewska D, Wysokińska A, Sadowska A*, 2006, Ejaculate traits and spermatozoa morphology as related to spermatozoa concentration in ejaculates of

Polish Large White boars, *Anim Sci Pap Rep,* 24 Supplement 3, 111-9.

21. *Kondracki S, Wysokińska A, Iwanina M, Banaszewska D, Sitarz D,* 2011, Effect of sperm concentration in an ejaculate on morphometric traits of spermatozoa of Duroc boars, *Pol J Vet Sci*, 14, 1, 35-40.
22. *Kondracki S, Banaszewska D, Wysokińska A, Iwanina M,* 2012, The effect of sperm concentration in the ejaculate on morphological traits of bull spermatozoa, *Folia Biol-Krakow,* 60, 1-2, 85-91.
23. *Kruger TF, Menkveld R, Stander FSH,*1986, Sperm morphometric features as a prognostic factor in in vitro fertilization, *Fertil Steril*, 46, 1118-23.
24. *Kruger TF, DuToit TC, Franken DR, Acosta AA, Oehninger SC, Menkveld R, Lombard C,* 1993, A new computerized method of reading sperm morphology (strict criteria) is as efficient as technician reading, *Fertil Steril,* 59, 202-9.
25. *Lim CC, Lewis SEM, Kennedy M,* 1998, Human sperm morphology and in vitro fertilization: sperm tail defects are prognostic for fertilization failure, *Andrology,* 30, 43-7.
26. *Morgentaler A, Fung MJ, Harris DH,* 1996, Sperm morphology and in vitro fertilization outcome: a direct comparison of World Health Organization and strict criteria methodologies, *Fertil Steril*, 64, 1177-82.
27. *Noorafshan A, Karbaly-Doust S,* 2010, A simple method for unbiased estimating of ejaculated sperm tail length in subjects with normal and abnormal sperm motility, *Micron*, 41, 96-9.
28. *Núñez-Martínez I, Moran JM, Peña FJ,* 2007, Sperm indexes obtained using computer-assisted morphometry provide a forecast of the freezability of canine sperm, *J Androl*, 30, 182-9.
29. *Ostermeier GCh, Sargeant GA, Yandell BS, Evenson DP, Parrish JJ*, 2001, Relationship of bull fertility to sperm nuclear shape, *J Androl*, 22, 4, 595-603.
30. *Peña FJ, Saravia F, García-Herreros M, Núñez-martínez I, Tapia JA, Johannisson A et al.,* 2005, Identification of sperm morphometric subpopulations in two different portions of the boar ejaculate and its relation to postthaw quality, *J Androl,* 26, 716-23.
31. *Rijsselaere T, Soom A, Hoflack G, Meas D, Kruif A*, 2004, Automated sperm morphometry and morphology analysis of canine semen by the Hamilton-Thorne analyser, *Theriogenology*, 62, 1292-306.
32. *Saravia F, Núñez-Martínez I, Morán JM, Soler C, Muriel A, Rodríguez-Martínez H et al.,* 2007, Differences in boar sperm head shape and dimensions recorded by computer-assisted sperm morphometry are not related to chromatin integrity, *Theriogenology*, 68, 2, 196-203.
33. *Sekoni VO, Gustafsson BK,* 1987, Seasonal variations in the incidence of sperm morphological abnormalities in dairy bulls regularly used for artificial insemination, *Br Vet J*, 143**,** 312-17.
34. *Severa L, Máchal L, Švábová L, Mamica O*, 2010, Evaluation of shape variability of stallion sperm heads by means of image analysis and Fourier descriptors, *Anim Reprod Sci*, 119**,** 50-5.
35. *Sutkevičienė N, Žilinskas H,* 2004, Sperm morphology and fertility in artificial insemination boars, *Vet Zoot*, 26**,** 48, 11-3.
36. *Szczygieł M, Kurpisz M,* 1999, Teratozoospermia and its effect on male fertility potential, *Andrology*, 31**,** 63-75.
37. *Wysokińska A, Kondracki S, Banaszewska D,* 2009, Morphometrical characteristics of spermatozoa in Polish Landrace boars with regard to the number of spermatozoa in an ejaculate, *Reprod Biol*, 9**,** 3, 271-82.
38. *Xu X, Pommier S, Arbov T, Hutchings B, Sotto W, Foxcroft G,* 1998, In vitro maturation and fertilization techniques for assessment of semen quality and boar fertility, *J Anim Sci*, 76, 3079-89.

KORELACIJA FREKVENCE MORFOLOŠKIH ALTERACIJA SPERMATOZOIDA I NJIHOVE KONCENTRACIJE U EJAKULATIMA POLJSKIH LANDRAS NERASTOVA

KONDRACKI S, BANASZEWSKA D, BAJENA M, KOMOROWSKA K i KOWALEWSKI D

SADRŽAJ

Ova ispitivanja su izvedena na 448 ejakulata dobijenih od 41 nerasta rase poljski landras. Ejakulati su uzimani za analizu u jednomesečnim intervalima u toku perioda 10 meseci. Morfometrijska merenja su vršena u svežim ejakulatima upotrebom mikroskopa. Ejakulati su bili klasifikovani u tri grupe na osnovu koncentracije spermatozoida. U ovoj studiji je napravljen pokušaj da se kod nerastova poljskog landrasa utvrdi korelacija između parametara kvaliteta ejakulata, učestalosti morfoloških alteracija spermatozoida i njihovih morfofometrijskih vrednosti sa koncentracijom spermatozoida. Potrebno je naglasiti da su vrednosti morfometrijskih parametara povezane sa koncentracijom. Spermatozoidi iz koncentrovanijih ejakulata su imali manje glave nego oni iz retkih. Ovo bi moglo da znači da je visoka plodnost nerastova koli daju koncentrovane ejaculate, ne samo rezultat visoke koncenracije, već i činjenice da spermatozoidi imaju manje glave. Najveća zastupljenost morfološki pravilno formiranih spermatozoida je utvrđena u ejakulatima sa koncentracijom od 400 do 500 $x10^3/mm^3$.