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An Assessment of the Breeding Maturity of Insemination Boars Based on Ejaculate Quality Changes

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We examined the sexual development and insemination capability of boars representing five breeds used at Polish sow insemination stations. The speed of attaining full breeding maturity by the boars and their insemination usefulness was assessed based on the results of experiments analysing age-related changes in the physical characteristics of the 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. The total number of spermatozoa in the ejaculates and the number of insemination doses obtained from one ejaculate were calculated using SYSTEM SUL v. 6.1 software. The sexual development of boars continues during insemination use, leading to improved ejaculatory performance. The physical parameters of the ejaculate change along with the age of the boar. The Duroc and Hampshire boars represent a pattern of sexual development that is different from that of the PLW, PL and Pietrain males, with an evident earlier termination of growth. The American boars (Duroc and Hampshire) acquire top ejaculatory performance approximately 6 months earlier.

Key words: Boar, ejaculate, age, breed.

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The reproduction performance of pigs partly depends on the fertility of the boar. Fertility is influenced by many external environmental and internal genetic factors (KUNAVONGKRIT *et al*. 2005). Genetic improvements aiming to raise the fattening and slaughter parameters cause young animals to grow faster. Increasingly younger animals are selected for reproduction. At the same time, as a result of strict selection and breeding progress, the carcasses tend to have a higher meat and lower fat content, which may impinge on reproductiveness (YOUNG & KING 1986). Meat-content-oriented selection can delay the puberty of pigs (JOHANSSON & KENNEDY 1983; GAUGHAN *et al*. 1995), inhibit testicular development and, consequently, deteriorate semen parameters and lower sex hormone concentration in the blood serum (ANDERSSON *et al*. 1999). The age is an inhibitive factor for the reproductive efficiency of boars. It may be regarded as a factor in the development of sexual functions during growth (FUENTES *et al*. 1987) – still before reaching sexual maturity. Otherwise, it may be considered as a differentiating factor of breeding usefulness of sexually mature boars (KENNEDY & WILKINS 1984; COLENBRANDER & KEMP 1990; KUNAVONGKRIT *et al*. 2005; HUANG *et al*. 2010). Young boars attain sexual maturity at the age of around 5-6 months, some already at 4 months. However, insemination boars normally start being used for breeding only at the age of 7-8 months. Such animals have generally reached sexual maturity, although they develop their full breeding potential at a much more advanced age. On this account, the ejaculates sampled in the first months of insemination use of boars may be highly variable as concerns their quantitative and qualitative characteristics. Sexual development usually corresponds with an improvement in boar ejaculate quality (GREGOR & HARDGE 1995; CLARK *et al*. 2003; BERTANI *et al*. 2002). Sex hormone levels gradually change. This concerns testosterone as well (ELLISON *et al*. 2002), which is crucial for the continuation of spermatogenesis. Spermatogenesis takes place throughout life until age-related fertility loss. During that time, as the male grows older, changes occur in the functioning of gonads (KIDD *et al*. 2001; ESKENAZI *et al*. 2003; HASSAN *et al*. 2003; KÜHNERT & NIESCHLAG 2004). Such changes have also been observed in men (Ng *et al*. 2004). Older men were found to have lower ejaculate volumes and lower sperm counts per ejaculate in comparison with younger men. It was observed that spermatogenesis efficiency in men decreases with age, which can result from falling numbers of Leydig cells that condition the process (Neaves *et al*. 1984). Smaller ejaculate volume may be caused by an incorrectly functioning prostate gland. Additionally, epididymal duct dysfunction can result in lower sperm motility (Neaves *et al*. 1984) which is also associated with prostate secretions (fructose, phosphatase). A crucial role in the control of testicular development and functionality is also played by estrogens (Kula *et al*. 2001; Oliveira *et al*. 2001). Estradiol prevents the apoptosis of gametes. If gonadotropins are absent or the gonads are dysfunctional, it also induces spermatogenesis, (Petikäinen *et al*. 2000; Ebling *et al*. 2000). The potential for estrogen synthesis is much greater in boar testes than in the testes of males of other species. Estrogens are also an important factor in the maturation of Sertoli cells (At-Taras *et al*. 2006) that affect spermatogenesis and testis weight. Hence, the dynamically rising weight of testes during pubescence as well as the number of reproductive and somatic cells within the parenchyma of testes may be determined by oestrogen levels. The testes of sexually mature males contain a relatively constant number of Sertoli cells (Berndtson *et al*. 1987). The number of correctly functioning Sertoli cells during embryonic development and in the postnatal period is crucial for the future reproductive capacity and fertility of a male (Franca *et al*. 2000; Sharpe *et al*. 2000). Both Sertoli cells and Leydig cells are vitally important for testis differentiation and development during embryogenesis (Franca *et al*. 2000; Cool & Capel 2009; Avelar *et al*. 2010). Two culminant periods of Sertoli cell multiplication have been observed in pigs – the first one directly after birth and the second one at the age of around 4 months (Franca *et al*. 2000). The number of Sertoli cells in the sinuous seminiferous tubules in the testes of males representing various species determines the number of spermatozoa in the ejaculate and, consequently, the reproductive efficiency of a male (Franca *et al*. 2000; Leal & Franca 2009).

One of the most important reasons for differing insemination capacity of breeding males are genetic differences, primarily the breed (Wysokińska *et al*. 2006). Pig breeds differ in the time of pubescence onset. Therefore, age-related ejaculate parameter changes may have different profiles in different breeds (Bertani *et al*. 2002; Kondracki 2003; Kondracki *et al*. 2005). This is suggested by the results of studies that have shown considerable differences of physical parameters of ejaculates collected from young boars (Kennedy & Wilkins 1984; Rathje *et al*. 1995; Bertani *et al*. 2002). This justifies the purpose of undertaking research to identify the nature of ejaculate parameter changes connected with sexual development in young boars of different breeds.

Material and Methods

The sexual development and insemination capability of boars representing five breeds used at Polish sow insemination stations at the Mazovian Centre for Animal Breeding and Reproduction in Łowicz were assessed. The analysed breeds were: Polish Large White (PLW), Polish Landrace (PL), Hampshire, Duroc and Pietrain. The speed of attaining full breeding maturity by the boars and their insemination usefulness was assessed based on the results of experiments analysing age-related changes in the physical characteristics of the ejaculates. Young boars of approximately 7-8 months of age – still before being used for breeding – were selected for analyses. The assessment of the physical parameters involved ejaculates sampled from 80 boars – 16 boars per each breed. All the boars selected for analyses were in good shape, without apparent developmental defects and with a correct sexual urge. The ejaculates were sampled manually (King & Macpherson 1973), once or twice a week at about 6 a.m. The assessment concerned all the ejaculates collected from the selected boars, starting from the first day of breeding use until the depletion of their semen. Each boar provided at least 100 ejaculates for analysis. Altogether 15668 ejaculates were assessed.

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 SYSTEM SUL v. 6.1 software.

The obtained numerical data were classified according to the age of the boars.
The first subgroup contained data on the ejaculates of boars under 9 months of age. The second subgroup included data on the ejaculates of nine- to

ten-month-old boars, while each successive subgroup involved data from an assessment of the ejaculates of boars that were incrementally older by 2 months.

The obtained material was statistically analysed using the ANOVA method. The significance of the differences between the groups was assessed with the Tukey test.

Results

Table 1 contains data that enable an assessment of the age-related ejaculate volume changes for boars representing the particular breeds. The average ejaculate volume of the youngest boars under one year of age generally did not exceed 210 ml. At this age, the largest ejaculate volumes were those

Table 1

Volume of ejaculate depending on the age and breed of boar (ml)

Age of boar (months)	Breed fo boar				
	PLW	PL	Hampshire	Duroc	Pietrain
	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$
<9	**151.03** ± 49.15	**168.37** ± 73.66	**227.76** ± 76.59	**140.06** ± 64.80	**180.34** ± 65.79
9-10	**194.94** ± 77.81	**191.88** ± 69.88	**220.02** ± 72.02	**131.77** ± 60.41	**180.36** ± 89.72
11-12	**208.32** ± 82.03	**210.75** ± 69.79	**222.99** ± 68.06	**140.79** ± 51.41	**210.48** ± 105.37
13-14	**225.75** ± 82.97	**229.19** ± 84.73	**230.17** ± 65.87	**148.59** ± 45.97	**229.68** ± 107.91
15-16	**248.30** ± 105.84	**231.39** ± 84.56	**234.12** ± 62.41	**159.69** ± 47.57	**244.51** ± 97.90
17-18	**255.14** ± 113.71	**246.89** ± 88.78	**250.10** ± 70.71	**165.89** ± 46.24	**240.29** ± 86.08
19-20	**253.41** ± 104.97	**256.23** ± 81.55	**260.01** ± 93.05	**169.71** ± 47.54	**243.29** ± 82.20
21-22	**256.48** ± 113.90	**262.32** ± 89.97	**275.75** ± 92.87	**174.55** ± 42.22	**264.11** ± 101.97
23-24	**269.51** ± 102.56	**276.04** ± 89.88	**271.50** ± 77.30	**171.83** ± 43.46	**272.47** ± 111.76
25-26	**273.41** ± 105.98	**277.61** ± 97.40	**280.19** ± 81.96	**167.23** ± 45.53	**283.54** ± 104.27
27-28	**291.66** ± 121.31	**303.05** ± 104.95	**263.16** ± 68.79	**171.71** ± 49.86	**283.68** ± 109.38
29-30	**282.61** ± 118.12	**284.18** ± 87.70	**250.90** ± 79.20	**174.51** ± 56.06	**294.19** ± 120.47
31-32	**288.82** ± 133.52	**284.63** ± 89.20	**289.68** ± 93.76	**172.13** ± 50.88	**277.29** ± 105.65
33-34	**260.82** ± 114.76	**302.91** ± 100.12	**291.65** ± 100.30	**177.47** ± 43.24	**284.56** ± 137.96
35-36	**237.76** ± 90.34	**302.40** ± 88.79	**311.27** ± 96.32	**176.96** ± 43.85	**273.96** ± 133.26
37-38	**260.61** ± 58.46	**298.45** ± 83.97	**298.04** ± 78.68	**169.48** ± 44.19	**287.66** ± 115.96
39-40	**257.22** ± 78.03	**269.60** ± 81.92	**271.38** ± 93.53	**153.29** ± 45.57	**268.24** ± 88.71
41-42	**238.33** ± 61.53	**240.19** ± 72.23	**266.47** ± 91.07	**168.00** ± 60.58	**252.38** ± 78.17
43-44	**228.00** ± 49.91	**276.28** ± 86.16	**280.89** ± 85.94	**159.85** ± 55.97	**225.53** ± 62.46
45-46	**227.25** ± 38.83	**313.33** ± 87.48	**314.17** ± 88.67	**175.19** ± 47.42	**256.10** ± 100.04
47-48	**249.71** ± 42.87	**300.67** ± 52.15	**332.42** ± 89.44	**170.00** ± 39.80	**284.83** ± 119.60
49-50	**317.60** ± 50.93	**285.53** ± 68.01	**285.95** ± 70.61	**174.13** ± 39.47	**300.36** ± 117.44
51-52	–	**265.45** ± 62.67	**267.06** ± 64.78	**160.00** ± 33.13	**295.93** ± 110.19
53-54	–	**288.00** ± 33.60	**312.22** ± 58.69	**140.57** ± 27.00	**272.50** ± 111.30
55-56	–	–	**395.00** ± 51.55	**138.50** ± 24.55	**266.74** ± 105.33
57-58	–	–	**413.33** ± 52.92	**160.56** ± 85.23	**150.00** ± 33.51
59-60	–	–	**365.56** ± 35.39	**120.00** ± 25.00	–
61-62	–	–	**288.33** ± 32.76	**170.00** ± 55.68	–
63-64	–	–	**344.38** ± 45.31	**168.33** ± 37.13	–
65-66	–	–	**449.09** ± 76.61	**120.00** ± 18.71	–
67-68	–	–	**468.67** ± 52.63	–	–
69-70	–	–	**432.00** ± 63.80	–	–
Total	**247.03** ± 104.05	**257.03** ± 93.45	**268.79** ± 88.06	**162.75** ± 50.52	**257.97** ± 109.58

$LSD_{0.05} = 4.805$; $LSD_{0.01} = 6.194$

PLW – Polish Large White; PL– Polish Landrace

$\bar{x}$ – arithmetic mean; Sd – standard deviations; LSD – least significant difference.

of the young Hampshire boars. The volume of the ejaculates of boars of this breed, collected before the age of one year, exceeded 220 ml and was significantly higher as compared with the other breeds ($P \leq 0.01$). As the boars grew older, they produced increasingly larger ejaculate volumes. However, the rate and scale of those changes differed depending on the breed. The volume of the semen fraction in the ejaculates of the young PLW, PL and Pietrain boars rose from the start of insemination use until the age of about 27-28 months, reaching the level of approximately 280-300 ml. The ejaculate volume increase in those breeds was very significant during that time and amounted to 100-140 ml, which means that the ejaculate volume rose by 58-93 % during 1.5 years of insemination use. Particularly high dynamics of ejaculate volume increase was observed at the beginning of exploitation, at the age of 8-16 months. During that time, the ejaculate volume of the PLW boars rose by over 97 ml, i.e. by approximately 12.2 ml per month on average. Simultaneously, the ejaculate volume of the PL and Pietrain boars increased by approximately 63-64 ml, i.e. by ca. 8 ml monthly. At the age of 17-28 months, the rate of ejaculate volume increase in the boars of those breeds was already considerably slower. The ejaculate volume rose then by 3.6 ml per month in the PLW boars and by 5.6 ml and 4.3 ml per month in the PL and Pietrain breeders, respectively.

Ejaculate volume changes in the Duroc and Hampshire breeders had a slightly different course in comparison with the PLW, PL and Pietrain males. The ejaculate volume of the Duroc and Hampshire boars also rose along with the age of the breeder but the dynamics of the change was very poor. Until the age of 27-28 months the volume of the ejaculates collected from the Hampshire boars grew by approximately 35 ml, while that of the Duroc boars by 32 ml. Assuming the volume of the ejaculates collected in the first month of insemination use to be 100, this shows an increase of about 15 % and 23 %, respectively, i.e. an increase that was a number of times lower than in the case of the PLW, PL and Pietrain boars. The differences in the dynamics of ejaculate volume changes of the particular boar breeds are shown in Figure 1. A period of intensive ejaculate volume increase is followed by a period of stabilisation in the volume of ejaculations at a level close to the maximum for a given breed. In the case of the analysed boars, it took place at the age of 27-38 months.

Ejaculate sperm concentration gradually fell as the boars grew older (Fig. 2). A fall in sperm concentration was more prominent in the PLW, PL and Pietrain ejaculates. Sperm concentration in the ejaculates of the PLW boars was generally higher as compared with the PL, Hampshire and Pietrain ejaculates. In spite of a fairly regular downward trend, until the age of 36 months sperm concentration in the ejaculates of the PLW boars exceeded 500K/mm^3. Falling ejaculate sperm concentration was observed in the Pietrain boars until the age of 30 months. At more than 30 months of age, sperm concentration in the ejaculates of this breed did not decrease. A completely different trend in sperm concentration changes was observed in the Hampshire and Duroc boars. The ejaculate sperm con-

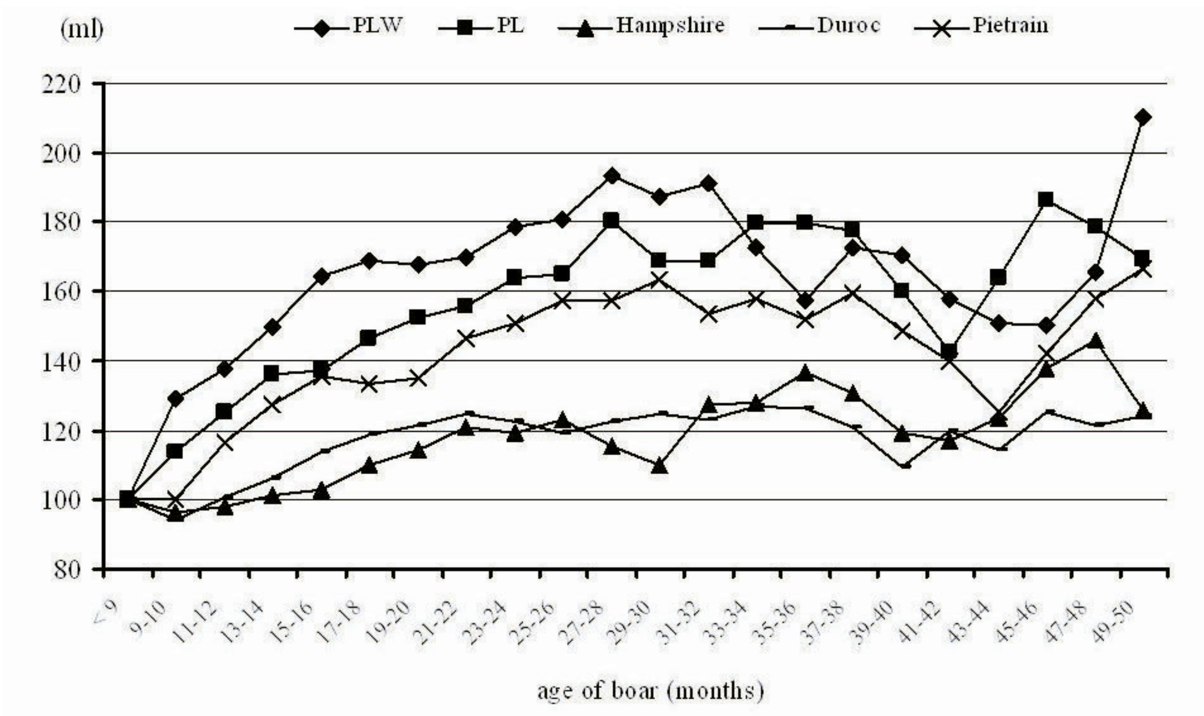

Fig. 1. The dynamics of ejaculate volume changes in the particular boar breeds depending on age (age below 9 months = 100).

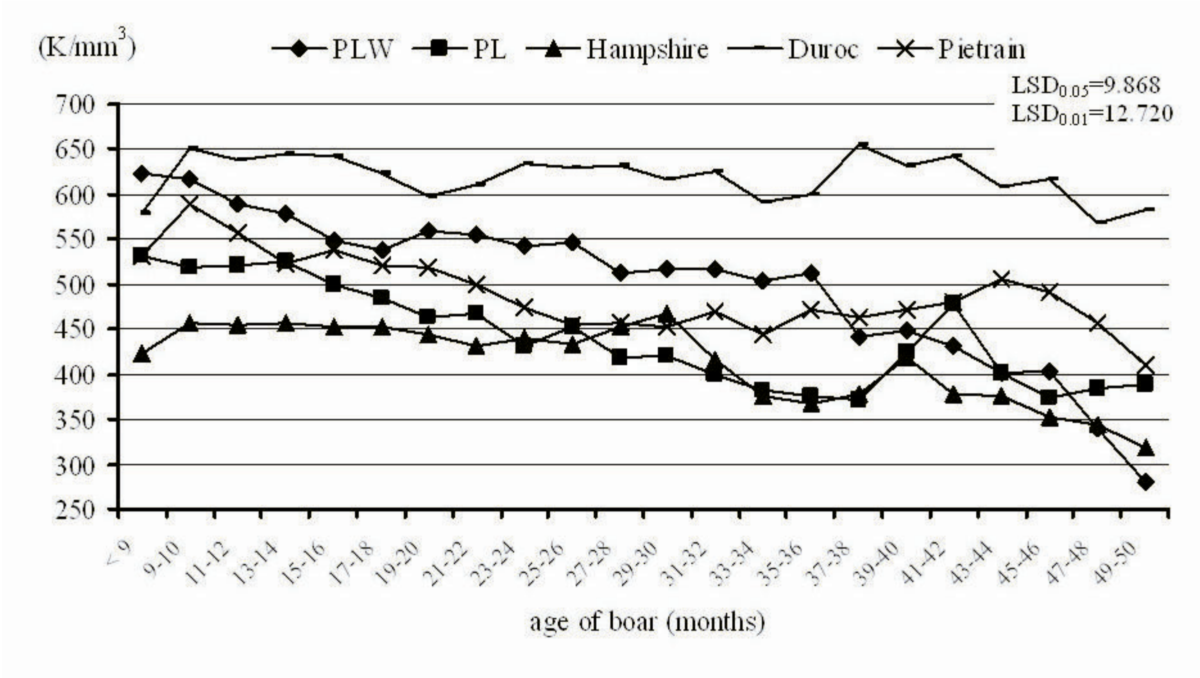

Fig. 2. The changes of sperm concentration in ejaculate in the particular boar breeds depending on age.

centration of the Hampshire boars did not significantly diminish until the age of approximately 29-30 months. A fall in sperm concentration in the ejaculates of these breeders was observed only at the age of more than 30 months. Sperm concentration in the ejaculates of the Duroc boars did not decrease with the age of the breeder and remained at a relatively steady level of about 567-654K/mm^3. A slight fall in sperm concentration in the ejaculates of these breeders was observed in boars more than 54 months. The ejaculate sperm concentration of the Duroc boars was higher than that of the other breeds, regardless of breeder age.

The percentage of spermatozoa with correct motility in the ejaculate slightly grew along with the age of the boars (Fig. 3). The scale of sperm motility changes in the ejaculate was nevertheless insignificant and, generally, did not exceed a few per cent. The most regular sperm motility growth was observed in the Hampshire ejaculates. The per-

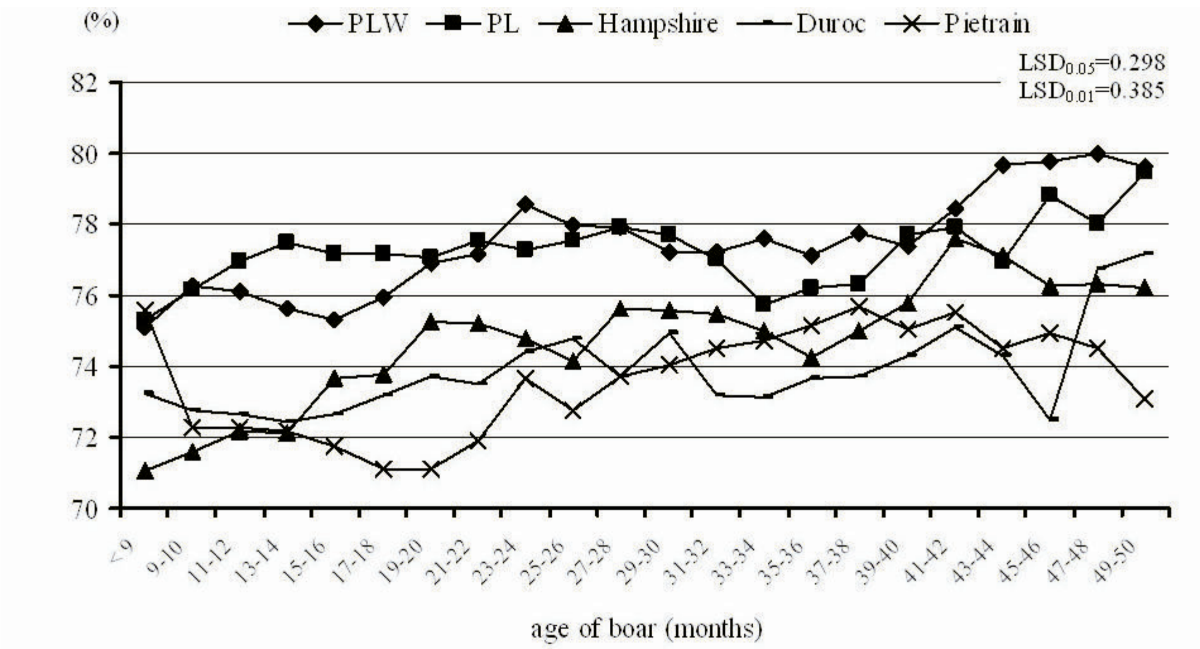

Fig. 3. The changes of total number of progressively motile spermatozoa in ejaculate in the particular boar breeds depending on age.

centage of progressively motile spermatozoa in the ejaculates of the boars of this breed steadily rose from around 71% at under 9 months of age to over 77.5 % at the age of 41-42 months. The growth in sperm motility in the Hampshire boars was then significantly more dynamic than in the other breeders. The age-related rise in sperm motility in PLW, PL and Duroc boars was also relatively regular, although less dynamic than in Hampshire boars. It is worth noting that the level of sperm motility in the ejaculates of the PLW boars was higher than in the case of the other animals, regardless of age. At above 20 months of age, the mean percentage of spermatozoa with progressive motility in the ejaculates of this breed exceeded 77 %.

Table 2 contains data illustrating the age-related changes in the total number of spermatozoa with

Table 2

Total number of spermatozoa in the ejaculates depending on the age and breed of boar (mld)

Age of boar (months)	Breed fo boar				
	PLW	PL	Hampshire	Duroc	Pietrain
	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$	$\bar{x} \pm Sd$
9	**67.93** ± 26.41	**64.02** ± 29.95	**66.52** ± 22.77	**56.83** ± 25.28	**67.31** ± 16.46
9-10	**86.28** ± 30.27	**72.34** ± 24.72	**70.77** ± 27.01	**58.16** ± 21.32	**69.03** ± 23.17
11-12	**88.39** ± 31.41	**80.98** ± 28.25	**71.03** ± 24.31	**60.22** ± 18.69	**75.77** ± 24.09
13-14	**92.17** ± 26.43	**89.48** ± 33.20	**73.26** ± 24.51	**66.57** ± 19.77	**77.50** ± 26.66
15-16	**96.12** ± 35.60	**87.68** ± 29.85	**77.27** ± 27.97	**72.50** ± 24.46	**85.90** ± 28.54
17-18	**95.59** ± 34.14	**88.83** ± 32.70	**81.44** ± 30.97	**74.61** ± 28.42	**82.77** ± 25.42
19-20	**101.08** ± 35.25	**89.66** ± 30.56	**83.08** ± 31.46	**72.77** ± 24.38	**85.07** ± 28.23
21-22	**103.49** ± 36.29	**93.59** ± 30.67	**86.43** ± 36.02	**76.58** ± 26.71	**89.72** ± 27.37
23-24	**108.27** ± 29.04	**92.00** ± 31.68	**84.79** ± 30.19	**81.22** ± 32.63	**87.72** ± 28.81
25-26	**107.92** ± 28.84	**95.84** ± 29.70	**86.90** ± 35.69	**78.69** ± 29.61	**90.38** ± 30.97
27-28	**107.77** ± 29.79	**97.81** ± 31.85	**87.33** ± 32.51	**79.82** ± 34.59	**90.35** ± 26.47
29-30	**108.19** ± 37.88	**93.38** ± 26.79	**85.71** ± 38.33	**78.84** ± 34.87	**92.53** ± 28.78
31-32	**106.92** ± 32.52	**89.65** ± 29.87	**86.91** ± 34.90	**75.82** ± 27.19	**91.88** ± 28.19
33-34	**99.54** ± 34.49	**87.22** ± 29.66	**82.01** ± 35.40	**74.72** ± 27.53	**86.00** ± 30.14
35-36	**90.35** ± 26.06	**84.82** ± 26.35	**83.98** ± 35.01	**76.54** ± 28.38	**87.57** ± 30.23
37-38	**92.68** ± 24.91	**82.33** ± 29.55	**83.34** ± 34.25	**80.25** ± 25.41	**93.29** ± 28.09
39-40	**90.11** ± 27.78	**84.24** ± 30.08	**85.08** ± 35.13	**70.76** ± 23.74	**92.52** ± 29.90
41-42	**84.09** ± 25.33	**82.46** ± 30.16	**76.43** ± 28.37	**79.50** ± 30.27	**90.48** ± 24.52
43-44	**75.38** ± 21.08	**76.98** ± 23.27	**79.19** ± 22.97	**71.42** ± 33.00	**85.49** ± 26.33
45-46	**72.09** ± 20.37	**83.03** ± 20.37	**86.96** ± 32.18	**76.78** ± 25.94	**94.73** ± 36.91
47-48	**66.65** ± 16.18	**86.41** ± 33.96	**87.78** ± 38.59	**73.35** ± 28.86	**94.57** ± 41.87
49-50	**69.20** ± 16.98	**81.73** ± 32.71	**69.05** ± 20.78	**76.89** ± 25.50	**83.55** ± 27.41
51-52	–	**108.02** ± 25.46	**73.36** ± 25.42	**67.30** ± 25.50	**76.20** ± 21.33
53-54	–	**72.01** ± 26.89	**94.87** ± 12.02	**55.79** ± 19.71	**68.10** ± 19.86
55-56	–	–	**77.51** ± 18.66	**49.39** ± 10.64	**80.08** ± 22.14
57-58	–	–	**95.28** ± 23.90	**48.78** ± 13.04	**56.55** ± 24.04
59-60	–	–	**96.11** ± 14.95	**45.04** ± 17.62	–
61-62	–	–	**54.97** ± 10.83	**52.11** ± 10.89	–
63-64	–	–	**58.74** ± 11.73	**53.44** ± 17.78	–
65-66	–	–	**78.32** ± 29.02	**35.96** ± 9.14	–
67-68	–	–	**61.75** ± 12.56	–	–
69-70	–	–	**52.38** ± 14.64	–	–
Total	**95.31** ± 32.97	**87.12** ± 30.82	**80.97** ± 31.97	**72.06** ± 28.06	**85.69** ± 28.88

$LSD_{0.05} = 1.618$; $LSD_{0.01} = 2.086$

Symbols as in Table 1

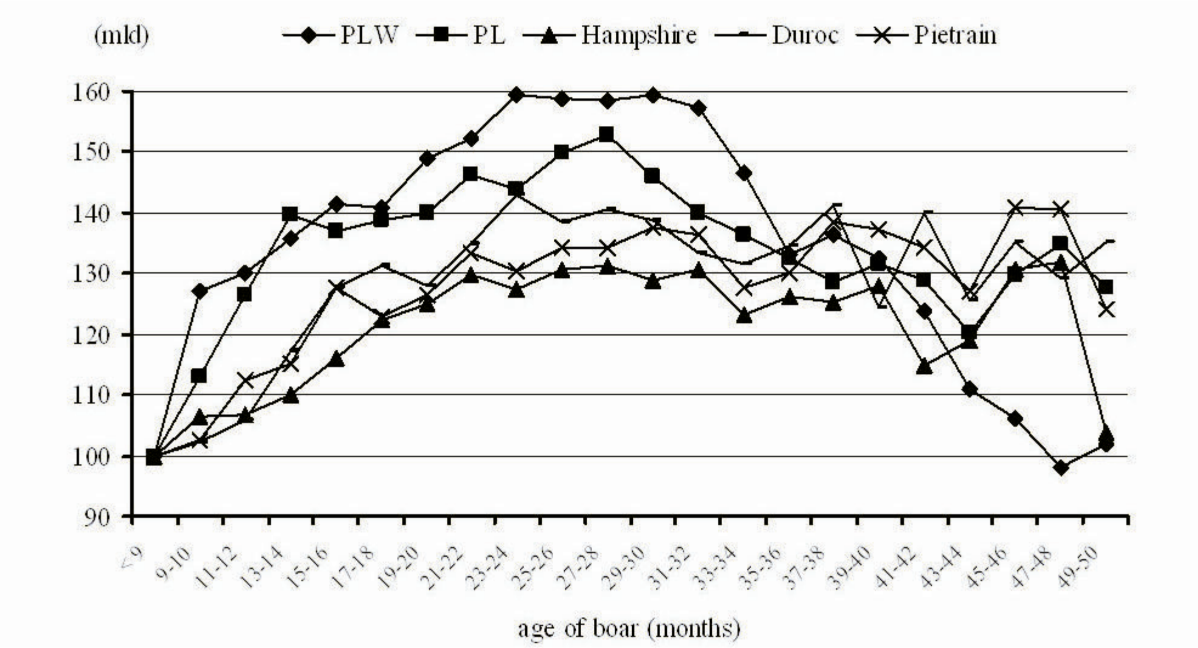

Fig. 4. The dynamics of total number of spermatozoa in the ejaculates changes in the particular boar breeds depending on age (age below 9 months = 100).

progressive motility in the ejaculates of the particular breeds. The total number of progressively motile spermatozoa rose as the boars grew older. Sperm content in the ejaculates systematically increased from the start of breeding use until the age of around 27-28 months, when the total number of spermatozoa reached a level close to the maximum. The Hampshire and Duroc boars attained maximal sperm production levels slightly earlier than the other boars – at the age of about 21-24 months. Until that age, the number of spermatozoa in the ejaculates of these boars increased by 20-24 bn. Moreover, the sperm count in the ejaculates of the Hampshire and Duroc breeders was lower than that of the other breeds, regardless of the age of the male. The scale of the sperm count changes in the ejaculates of the Hampshire and Duroc boars aged above 21-24 months was already insignificant and remained at a relatively stable level until the age of 37-40 months. The sperm count in the ejaculates of the Pietrain boars rose until the age of 30 months, in the ejaculates of the PL boars until 28 months, and in those of the PLW boars until the age of 23-24 months. At 23-32 months of age, the number of spermatozoa in the ejaculates of the PLW boars was approximately 105-110bn, i.e. significantly more than in the ejaculates of the other breeds ($P \leq 0.01$). A fall in the sperm count in the ejaculates of the PLW breeders was observed only at the age of more than 34 months. The highest dynamics of ejaculate sperm count increase was observed during the first months of insemination use (Fig. 4). However, certain interracial differences were observed in the dynamics. At the age of 8-10 months, the highest rate of sperm count increase was observed in the PLW boars. At that time, the mean sperm count in the ejaculates of this breed rose by over 18bn, i.e. by over 9bn per month. A similar tendency for the ejaculate sperm count to rise was observed in the PL boars whose sperm count in the ejaculates collected until the age of 13-14 months increased by over 4bn monthly. A relatively intensive sperm count increase until the age of about 15-16 months was also observed in the ejaculates of the Duroc and Pietrain boars. However, in comparison with the PLW and PL males, sperm count growth in the ejaculates of these breeders was much smaller during that period, at 2.0-2.3bn per month.

With age, the number of insemination doses obtained from one ejaculate rose at a rate similar to the growth of ejaculate volume and sperm count (Fig. 5). The highest number of insemination doses was obtained from the ejaculates collected at the age of around 25-30 months. A particularly dynamic increase in the number of insemination doses obtained from one ejaculate was identified for the PLW boars. Until the age of 27-28 months the number of insemination doses obtained from the ejaculates of this breed rose up to over 32 portions from approximately 17 portions at under 9 months of age, i.e. almost twice. The ejaculates of the PLW boars collected at over 2 years of age produced over 30 semen portions, i.e. over 10 portions more than the ejaculates of the two-year-old Duroc boars ($P \leq 0.01$), over 6 portions more than the Hampshire boars ($P \leq 0.01$), and almost 4 portions more than the ejaculates of the Pietrain boars

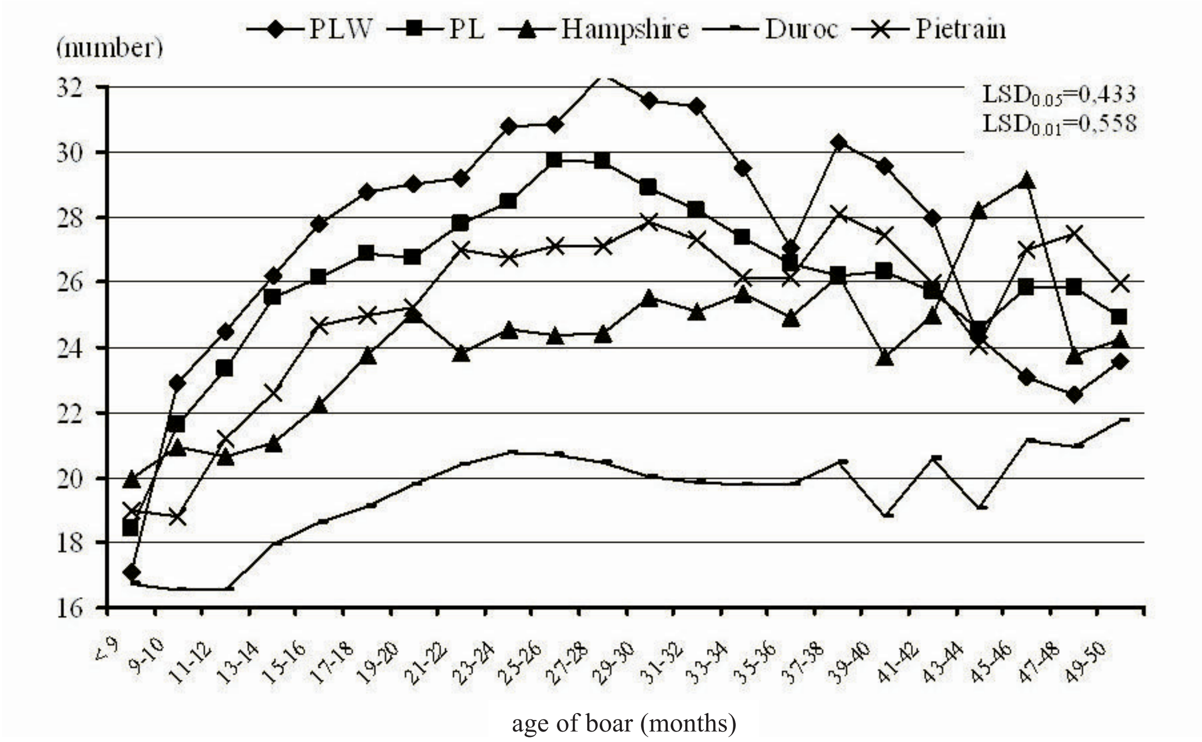

Fig. 5. The number of insemination doses in the particular boar breeds depending on age.

($P \leq 0.01$) collected at the same age. At over 30 months of age, the number of insemination doses sampled from the ejaculates of the PLW boars gradually decreased. However, even the oldest animals of this breed did not produce fewer than 22 portions per ejaculate. The fewest insemination doses were obtained from the ejaculates of the Duroc boars. The number of insemination doses obtained from the ejaculates of this breed rose until the age of 23-24 months, though at a much slower rate than that of the other breeds. The number of semen portions prepared from the ejaculates of the Duroc boars collected at the age of more than 2 years did not appreciably change and equalled 19-21 doses.

Discussion

The sexual development of pig males is not over at 8-9 months of age, when boars start to be used for insemination, but proceeds until a much more advanced age. This is indicated by the results of the present work which show that the ejaculate volume and sperm count of most of the boars increased until the age of approximately 27-28 months. Based on abundant and reliable experimental material, the data show that the sexual development of insemination boars proceeds until the age of more than 2 years. Until now, this pattern has not been supported with sufficient experimental data. Most studies in this area have focused on a very early period in the sexual development of boars (CAMERON 1985; HARAYAMA & KATO 1992; RATHJE *et al.* 1995; BERTANI *et al.* 2002). Research on the breeding performance of boars at a more advanced period in their insemination use has seldomly been undertaken. Most studies did not present unequivocal results showing an improvement of boar reproductive characteristics at the age of more than 1.5 years (CLARK *et al.* 2003). The results of some studies even indicate that the parameter variability of insemination boar ejaculates significantly decreased already at the age of 12-16 months (CARTEE *et al.* 1986; GREGOR & HARDGE 1995; FLOWERS 2008). However, there are studies available showing that the ejaculate volume and sperm count can increase until a much more advanced age. Some authors have reported that boar ejaculate volume grows until the age of around 29-30 months (KENNEDY & WILKINS 1984; JANKEVICIUTE & ZILIKSKAS 2002). Others have observed rising boar ejaculate volumes and sperm counts until approximately 25 months of age (STANČIĆ *et al.* 2003), or even 30 months (SURIYASOMBOON *et al.* 2004). In turn, FALKENBERG (1992) suggests that boars attain optimal ejaculatory efficiency only after fully growing up, i.e. at the age of about 2.5-3 years. SMITAL's (2009) research revealed that boar sperm production rose until the third year of life. The culmination was observed by the author at the age of 3.5 years. Then, a fall in the number of spermatozoa per ejaculate followed. CLARK *et al.* (2003) reported a dynamic rise in the ejaculate sperm counts of boars aged from 8-10 months to 14 months, followed by a stabilisation of the number of spermatozoa per ejaculate. WOLF and SMITAL (2009a) found that ejaculate volumes increased until the age of 2 years. The most dynamic ejaculate volume rise was observed until 18 months of age. The maximum ejaculatory performance measured with the total sperm count per ejaculate was attained by the boars at the age of 21 months. Afterwards, the sperm count remained at a relatively steady level.

The results presented in the present work unequivocally prove that the sexual development of boars continues during the breeding use, until the age of more than 2 years. This obviously affects the quality of the ejaculates and the efficiency of using boars for insemination. During this time, the development of the seminiferous epithelium in boar testes that determines the sperm count and the development of the secretory functions of the accessory sexual glands whose secretions affect the ejaculate volume proceed. This is evidenced in the continued increase of ejaculate volumes and sperm counts demonstrated in the present paper. The further development of sexual glands in sexually mature and active breeding boars is confirmed by testicular morphology analysis. It was shown that boar testes increase in size until the age of 20 months (CLARK *et al.* 2003). However, the largest growth of the testicular and epididymal mass was observed in young boars aged from 2 to 6 months. During that time, the mass of these organs increased more than threefold (KUMARESAN *et al.* 2008). According to RATHJE *et al.* (1995) the physical parameters of the ejaculates of boars with higher daily weight gains and larger testes are definitely better.

By analysing the present material it is possible to identify the age limits at which significant quantitative changes of the boar ejaculate parameters occur. The age of about 12 months is of particular significance. The ejaculates of all the breeds at this age had significantly lower volumes and contained fewer spermatozoa than the ejaculates of older breeders. They produced fewer insemination doses. At the age of 12-30 months, a gradual increase in ejaculate volume and total ejaculate sperm count was observed. The most dynamic changes of ejaculate parameters took place in the first months of breeding use, i.e at the age of about 8-16 months. Similar observations are presented in a study by FLOWERS (2008). A dynamic ejaculate sperm count increase was observed until the age of about 13 months. Afterwards, the sperm count changes were modest and remained at a fairly stable level.

Completely different changes relating to ejaculate sperm concentration were observed in the present work. Sperm concentration did not increase in parallel to ejaculate volume and sperm count. On the contrary, it gradually decreased. Published data often indicate an entirely different direction of sperm concentration changes. In general, they demonstrate rising ejaculate sperm concentration along with the age of the boars (HOVORKA & SLECHTA 1985; STANČIĆ *et al.* 2003). Sperm concentration increase in the ejaculates of eight- to eighteen-month-old boars was also observed by CAMERON (1985). Similar observations were also made by other researchers who noted rising boar ejaculate sperm concentration until the age of about 30 months (KENNEDY & WILKINS 1984). On the other hand, ČEŘOVSKÝ *et al.* (2005) identified lower sperm concentration and total sperm count in older boars aged over 2.5 years as opposed to those several months younger. In turn, JANKEVICIUTE's and ZILIKSKAS' (2002) study shows that ejaculate sperm concentration grows until the age of 18-24 months. Afterwards, at the age of 24-30 and above 30 months, a decrease follows. There are also data available that confirm an age-related decrease in boar sperm concentration. WOLF and SMITAL's (2009b) show a gradual fall in sperm concentration from up to approximately 12 months of age until the age of about 3 years, following a short period of sperm concentration increase in young animals. CARTEE *et al.* (1986) did not observe, however, any significant differences between ejaculate sperm concentration in boars aged 9 and 15 months. Some data indicate that ejaculate sperm concentration remains at a steady level in boars aged from 16.5 to 21 months (KOZINK *et al.* 2004).

Independently from the abovementioned general tendencies, significant differences were found between the particular breeds in the levels of the physical parameters of the ejaculates and in the evolution of the age related changes in those parameters. This points at differences in the sexual development of the boars representing the analysed breeds. The collected data confirm that Hampshire and Duroc boars attain full breeding efficiency earlier than PLW, PL and Pietrain breeders. The former already provide the most voluminous and sperm-rich ejaculates at the age of 21-24 months, which suggests that boars of this breed are quicker to develop both the seminiferous epithelium of the testicular seminal tubules and the secretory capacity of the accessory sexual glands. The development and improvement of the sexual functions of PLW, PL and Pietrain boars is somewhat slower, lasting until the age of approximately 28-30 months. Until that time, a very large increase in ejaculate sperm count and ejaculate volume was observed in those breeds. The Duroc boars displayed specific ejaculate characteristics. The ejaculate volume of this breed was considerably lower as compared with the other breeds. In turn, the sperm concentration in the ejaculates of the Duroc boars was always higher than in the other breeds, regardless of the age of the breeder. Considering that the scale of the age-related ejaculate parameter changes in this breed was insignificant, it can be assumed that the functioning of the accessory sexual glands and the development of the seminal tubules in the testes of Duroc boars is already at an advanced stage at the age of breeding use initiation. The opinions on the performance of boars of this breed are ambiguous. There are publications available that document a gradual increase in the ejaculate volume of Duroc boars aged from 9 to 33 months (SURIYASOMBOON *et al*. 2004). Moreover, HUANG *et al*. (2010) observed that the ejaculatory performance climax is attained by this breed at approximately 3 years of age. After reaching the climax, the ejaculate volume of Duroc boars decreases at a slower rate than the other semen parameters (HUANG *et al*. 2010). This means that spermatogenesis efficiency is more susceptible to the effect of ageing as compared with the functionality of the accessory sexual glands that determines the ejaculate volume. Regardless of the progressive increase in certain semen parameters, analyses of the physical characteristics of Duroc boar ejaculates generally reveal relatively low ejaculate volumes and low ejaculate sperm counts, with a relatively high sperm concentration (KONDRACKI 2003). Some of the experiments conducted on very young Duroc boars led to the conclusion that lower ejaculate volume and a low sperm count result from a later maturation of this breed. However, low levels of the basic physical ejaculate parameters were also characteristic of older Duroc boars (BORG *et al*. 1993; PARK & YI 2002; KONDRACKI *et al*. 2004; KONDRACKI *et al*. 2011), which does not confirm the hypothesis of later maturation as the reason for lower semen parameters of young boars of this breed. The data presented in this paper even suggest the possibility that Duroc boars accomplish their sexual development earlier than the other breeds. This is evidenced in lower dynamics of sperm count increase as compared with the other breeds and, especially, ower dynamics of the age-related ejaculate volume increase and an earlier attainment of the threshold of uppermost ejaculatory performance. The data support the view that the sexual development of Duroc boars ends earlier than in the other breeds. It may well be that the hypothesis of an earlier termination of the sexual development is also well-founded in the case of Hampshire boars. The results of the present work also show that the ejaculatory performance climax for boars of this breed comes at an earlier age than for PLW, PL and Pietrain breeders.

Apart from the age-related improvement in the quantitative parameters, certain studies report a parallel sperm motility increase in fresh ejaculates (ŠERNIENĖ *et al*. 2002; STANČIĆ *et al*. 2003; WOLF & SMITAL 2009b). This not only implies positive transformations during spermatogenesis, but also advantageous changes in the composition of the semen plasma, a natural medium for spermatozoa. The most frequent reasons for lowered sperm motility are spermatogenetic disorders, epididymal epithelium dysfunctions, as well as an impairment of the functioning of the accessory sexual glands (PINART *et al*. 1999). It was concluded that sperm motility in young boars aged up to 18 months is relatively low. It rises along with age, culminating at 18-24 months, and declines in older animals (JANKEVICIUTE & ZILIKSKAS 2002). Some researchers also indicate interracial differences in sperm motility. The semen of Lithuanian White boars collected at the age of 10-18 months contained a smaller percentage of progressively motile spermatozoa in comparison with Pietrain boars of the same age (ŠERNIENĖ *et al*. 2002).

The age of the boar was found to affect the number of insemination doses obtained from one ejaculate (RUTTEN *et al*. 2000). Our results show that certain breeds produce significantly fewer semen doses regardless of the age of the breeder. These primarily included the majority of the Duroc boars that provided very few insemination doses. Collected at an age of above 17 months, the ejaculates of this breed provided roughly 19-20 semen portions, this number being close to the maximum. At the same age, the PLW boars produced 29-32 insemination doses per ejaculate, i.e. 10-12 portions smore than the Duroc boars.

The correlation between ejaculate parameters and age, as well as ejaculate parameter changes occurring during sexual development were identified in humans (CHEN *et al*. 2003; MUKHOPADHYAY *et al*. 2010), boars (FLOWERS 2001; BANASZEWSKA *et al*. 2011), bulls (AL-MAKHZOOMI *et al*. 2008) and even chimpanzees (MARSON *et al*. 1991). Experiments conducted by HUANG *et al*. (2010) also confirm the hypothesis that boars do not manifest the highest ejaculatory performance after having attained breeding maturity. According to the literature, the best semen quality in the temperate climate zone is that of twenty four- to twenty nine-month-old boars (KENNEDY & WILKINS 1984), while the best age in the tropical zone is 33 months. It is possible to identify age-related thresholds of optimal ejaculatory performance not only for animals. Men's ejaculatory efficiency has also been found to increase until the age of about 40-45 years, with a subsequent decline of these parameters (HASSAN *et al*. 2003; PASQUALOTTO *et al*. 2005).

In summary, we conclude that the sexual development of boars continues during insemination use, leading to improved ejaculatory performance. The physical parameters of the ejaculate change along with the age of the boar. However, both the direction and dynamics of the changes vary depending on the breed and between particular ejaculate parameters. The Duroc and Hampshire boars represent a pattern of sexual development that is different from that of the PLW, PL and Pietrain males, with an evident earlier termination of growth. The American boars (Duroc and Hampshire) acquire top ejaculatory performance approximately 6 months earlier. The scale and dynamics of the age-related changes of the physical ejaculate parameters in the Duroc and Hampshire insemination boars are much lower as compared to the European breeds. The most advantageous course of sexual development was observed in the PLW boars. Both the ejaculate volume and the number and motility of spermatozoa in this breed dynamically rose with age. Thus, regardless of the age of the boar, it was possible to prepare more insemination doses per ejaculate in contrast to the other boar breeds.

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