

Influence of Collection Frequency on Boar Ejaculate Quality Parameters

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Abstract

The aim of study was to examine the effect of the boar ejaculate collection frequency on the parameters of its quality. The research covered a three-year period, and four most common boar breeds in our area. A total of 1705 ejaculates were taken from Yorkshire boars, 1693 ejaculates from Landrace boars, 1106 ejaculates from Pietrain boars and 669 ejaculates from Duroc boars. Within each breed, groups were formed according to the frequency of ejaculate taking (once, twice, three, four, five, or six times a month). Evaluation of ejaculate quality was performed in a reprocentre laboratory, according to their quality standards. Quality parameters that have been determined were the volume of ejaculate, sperm concentration, progressive sperm motility, live sperm count, the number of doses per ejaculate and the overall assessment of ejaculate. Results of this study show that the volume and progressive motility increase with the increase of frequency in ejaculate taking, while the sperm concentration drops, except for the Pietrain boars. The best results were observed in ejaculates collected 5-6 times during the month. The highest mean volume of ejaculate was found in the Landrace boars (292.22 ml), in ejaculate taking frequency of five times per month. The highest average progressive sperm motility was 92% in the Yorkshire boars, on collection frequency of six times a month, while the highest concentration of spermatozoa was found in the Duroc boars in collection frequency of one taking per month (447.32 mil/ml). The results of this study indicate that the frequency of ejaculate taking affects the boar semen quality parameters.

Key words: ejaculate, frequency of collection, sperm quality

Introduction

Frequency of ejaculation, i.e. reproductive exploitation of boar, is one of the main factors affecting the quantity and quality of ejaculate (Pruneda et al., 2005; Smital, 2009). Sperm taking (every 24 or 48 hours) can often lead to a maximum amount of sperm per time unit, but its fertility may be reduced. On the other hand, if the ejaculates are taken irregularly and with longer intervals, a significant number of the sperms would be excreted through the urine, which results in incomplete exploitation of the boar reproductive potential (Singleton & Flowers, 2001). The interval between the two ejaculate takings should be long enough to renew the sperm reserve in epididymis, but not too long that the production of the sperms exceeds the capacity of the reserves, and the sperm is lost through urine excretion (Briz et al., 1995, 1996; Singleton & Belstra, 1997).

Audet et al. (2009) state that the rhythmically high frequency of semen collection does not have a significant effect on the quality of ejaculates, while other authors claim that semen quality is worse at more frequent collection, and that poor semen quality is a consequence of the rapid passage of sperms through the epididymis, due to which sperms do not have sufficient time to complete their maturation (Bonet et al., 1991; Strzezek et al., 1995; Pruneda et al., 2005). Most studies, however, indicate that too high frequency of ejaculate collection leads to a decrease in sperm count per ejaculate, resulting in a decrease in the number of insemination doses produced (Bonet et al., 1991; Singleton & Flowers, 2001; Pruneda et al., 2005). Strzezek et al. (1996) found that too frequent reproductive exploitation of boars leads to a decrease in the percentage of progressive motility, concentration and total sperm volume, with an increase in the proportion of abnormal spermatozoa and spermatozoa with damaged membranes. Wolf and Smital (2009a) analyzed the effect of collection frequency on ejaculate quality parameters in Czech Large White and Czech landrace boars, and found that sperm concentration gradually decreased, while ejaculate volume increased, with shortening of the interval between two consecutive ejaculate collections. Miclea et al. (2007) found that the average volume of ejaculates increased by 7% with decreasing frequency of taking (from every three to four, and from every four to five days), while at the same time an increase of average sperm concentration by 11% was observed.

According to these authors, the frequency of ejaculate collection did not significantly affect the percentage of progressive motility of sperms. These authors state that daily ejaculate collection results fast in reduced values for most quality parameters and recommend ejaculate collection once a week, whereas more frequent collection is acceptable only when it is highly needed and should not take long.

Assuming all the above mentioned, the aim of this study was to examine the effect of the boar ejaculate collection frequency on the parameters of its quality in conditions of commercial reproductive centre.

Materials and Methods

The research covered a three-year period, and four most common boar breeds in our area. A total of 1705 ejaculates were taken from the Yorkshire boars, 1693 ejaculates from the Landrace boars, 1106 ejaculates from the Pietrain boars and 669 ejaculates from the Duroc boars. Within each breed, groups were formed according to the frequency of ejaculate collection (once, twice, three, four, five, or six times a month, Table 1.).

Tab. 1. Number of ejaculates collected in different boar breeds and frequency of collection per month

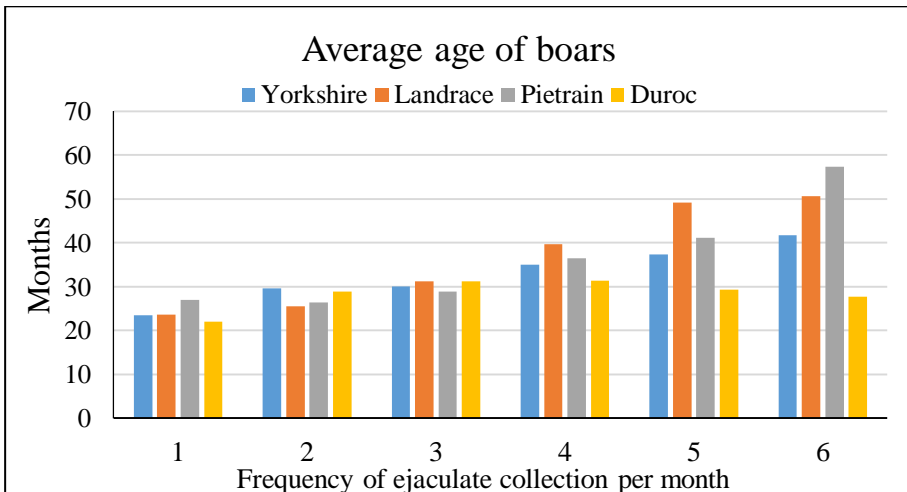
Breed of boar	Frequency of ejaculate collection per month						Total number of ejaculates
	Once	Twice	Three times	Four times	Five times	Six times	
Yorkshire	33	146	460	674	355	37	1705
Landrace	77	418	621	460	99	18	1693
Pietrain	32	308	401	282	65	18	1106
Duroc	19	48	130	242	170	60	669

Evaluation of ejaculate quality was performed in a reproductive centre laboratory, by an expert evaluator and according to the reproductive centre quality standards. Quality parameters that have been determined were: the volume of ejaculate (ml), the percentage of progressively motile spermatozoa (%), and sperm concentration (in 10^6 /ml). In addition to these three basic parameters of ejaculate quality, the number of ejaculates (N), age (months), the percentage of living sperm (%), the number of doses per ejaculate and the overall assessment of ejaculate were monitored.

The volume of ejaculate was measured in a graduated glass. The concentration of spermatozoa was determined spectrophotometrically. The number of produced insemination doses per ejaculate was calculated on the basis of the concentration of spermatozoa and the required dilution rate to obtain the desired number of fertility-capable spermatozoa in the insemination dose, and the overall assessment of ejaculate according to expert evaluator's experience. A number of living or dead spermatozoa was determined by an HOS test, described by Pérez-Llano et al. (2001). The SPSS-22 software packages were used for statistical analysis.

Results and Discussion

A question that is often asked in the boar reproductive exploitation is what the most optimal frequency of ejaculate collection is to obtain the maximum number of insemination doses without compromising the quality of the semen (Kanokwan, 2011; Jotanović & Savić, 2017, Savić et al., 2017). On the one hand, too frequent ejaculate collection leads to a decrease in its quality, while not collecting ejaculate often enough is economically unjustified because of the costs of boar keeping and feeding, as well as the absence of ejaculation results in the resorption of spermatozoa in the epididymis (Briz et al. 1995; 1996, Singleton & Belstra, 1997; Singleton & Flowers, 2001, Frangež et al., 2005). Singelton (2001) states that the optimum frequency of ejaculate collection is about 4.7 times a month on average, or slightly more than once a week. Stančić and Veselinović (2002) state that the usual norm for boar in reproductive centres is one collection in three days, i.e. about 10 times per month. Wolf and Smital (2009b) consider that a time interval of 7 to 10 days between two ejaculates is the best because the values of all ejaculate quality parameters are within the optimum range. Uzelac and Vasiljević (2011) state that the frequency of ejaculate collection in adult boars should be at intervals of 4 to 5 days, while in young boars, the intervals of 6 to 7 days are recommended. The average age of different boar breeds included in this study is presented in Graph 1.

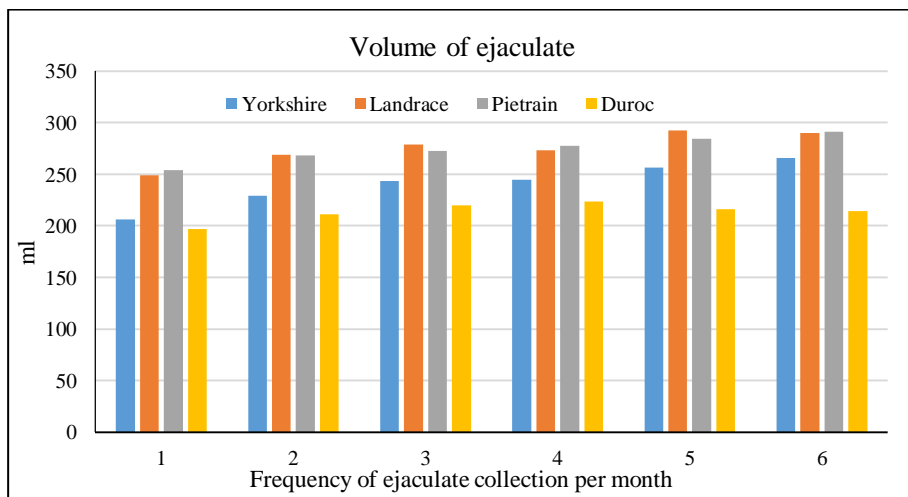


Graph 1. Average age of different boar breeds included in the study according to the ejaculate collection frequency (source: Sarajlić, 2018)

The age of the boars has increased along with the increase in the frequency of ejaculate collection (Graph 1), which is the expected and usual trend of increasing the reproductive exploitation rate of the boars in reproductive centres with the increase of their age (Uzelac & Vasiljević, 2013). This trend is found in all breeds except Duroc, in which the highest age was found in boars whose ejaculates were collected three and four times a month.

The volume of ejaculates is determined immediately after collection, in a graduated vessel, or approximately, based on the weight of the ejaculate. Data from the literature indicate that the range of variations for ejaculate volume is 50 to 1000 ml, with an average value of about 200 to 250 ml (Ardelean, 2002; Kyriazakis & Whittemore, 2006). The dominant factor affecting ejaculate volume is boar's age, since younger boars, at the beginning of exploitation, give ejaculates with less volume than older boars (Kanokwan, 2011, Banaszewska & Kondracki, 2012). Other factors that may influence ejaculate volume include breed or hybrid of boar, season, diet, housing, reproductive exploitation intensity, and others (Pruneda et al., 2005; Kunavongkrit et al., 2005; Smital, 2009; Wolf, 2009, Savić et al., 2017)

The average volume of ejaculates in different boar breeds, according to the collection frequency is presented in Graph 2.



Graph 2. Volume of ejaculates in different boar breeds according to the ejaculate collection frequency (source: Sarajlić, 2018)

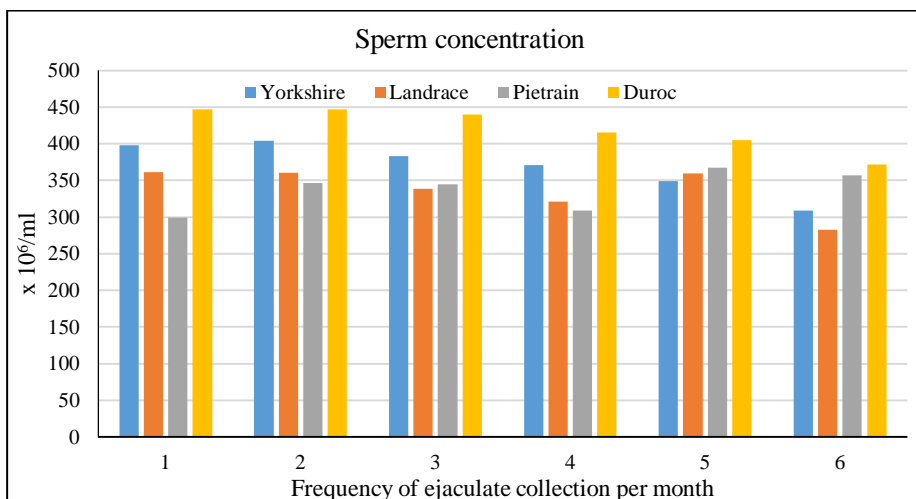
The data presented indicate a general trend of an increase in the average ejaculate volume with an increase in the frequency of ejaculate collection, with variations in different breeds being noticeable.

The Landrace and Pietrain boars gave ejaculates of the highest volume, regardless of the frequency of administration, as opposed to those of Duroc, whose ejaculates had the lowest volume in each frequency of administration. Compared to the results of other authors (Miclea et al., 2007; Frunza et al., 2008, Uzelac & Vasiljevic, 2013), the results of this study are slightly lower, but are within the scope of physiological variations. When interpreting the results obtained in this study, it is important to note that ejaculate volume is greatly influenced by a boar's age, as it is common for the intensity of reproductive exploitation of boar to increase with age, which is confirmed by the results of this study.

Sperm concentration, together with ejaculate volume, is a key factor in determining the total number of sperm in the ejaculate, possible degree of ejaculate dilution and the number of insemination doses that can be produced (Savić et al., 2013). Since it is negatively correlated with ejaculate volume, it is expected that an increase in the frequency of ejaculate collection will have a negative effect on sperm concentration (Rothschild & Ruvinsky, 2011), leading to its decrease, due to the faster emptying of spermatozoa reserve in the epididymis. It is important to keep in mind that reducing the sperm concentration with increasing frequency of ejaculate collection does not necessarily mean a decrease in the total fertility capacity of ejaculates, as increasing the frequency of ejaculate collection leads to an increase in ejaculate volume, and thus often an increase in the total number of sperm in the ejaculate (Stančić et al., 2003; Knecht et al., 2014; Jotanović & Savić, 2017). Cergolj and Samardžija (2006) found sperm 25 to 300 x10⁶/ml concentration limits, which is lower than the results of this study. According to Wolf and Smital (2009b), these limits are significantly larger, and range from 50 to 900 x10⁶/ml, which is in accordance with the results of this study.

Comparing the results of this study with those of Frunza et al. (2008), who found an average sperm concentration of 262 x10⁶/ml and a limit value of 100-600 x10⁶/ml, it is noticeable that the results of this study have a higher average value for this parameter, and are in range within the stated range of limit values. Our results for all breeds included, especially for Landrace, are lower than the values obtained by Wolf and Smital (2009b) for boars of Czech Landrace. According to these authors' study, the average sperm concentration for the Pietrain boars 453 x10⁶/ml, which is higher than our results for all breeds included.

The average sperm concentration in ejaculates in different boar breeds according to the collection frequency is presented in Graph 3.



Graph 3. Sperm concentration in different boar breeds according to the ejaculate collection frequency (source: Sarajlić, 2018)

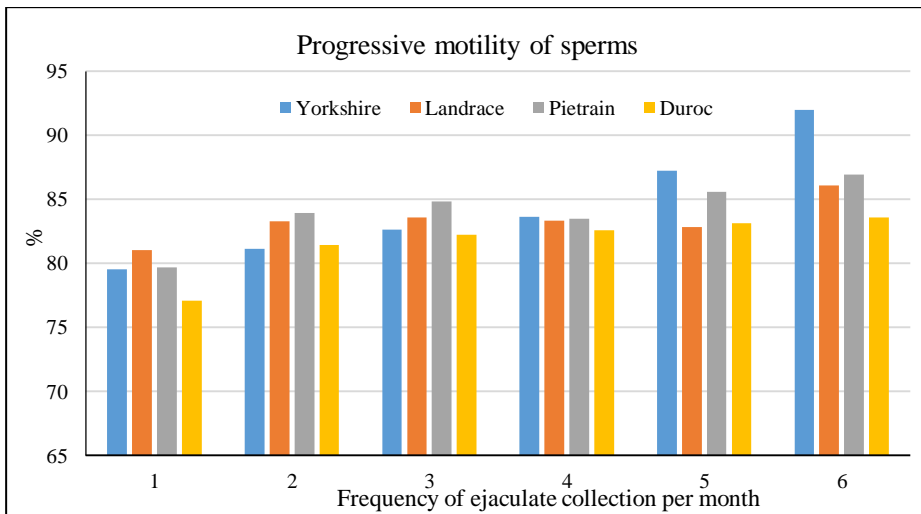
The data presented (Graph 3) indicate a general trend of a decrease in the average sperm concentration with increasing frequency of ejaculate collection in boars of all breeds included, with variations with respect to breed characteristics, which is similar to the results of other authors (Borg et al., 1993; Savić et al., 2015). The results presented for sperm concentration in this study confirm the literature claiming that it is negatively correlated with ejaculate volume, which is particularly noticeable in the Duroc boars (Savić et al., 2013, 2015, Savić, 2014).

The progressive motility of sperm is considered to be practically the most important feature of sperm, which allows it to pass through the reproductive organs of sows and reach fallopian tubes to perform fertilization (Feitsma, 2009). The percentage of progressive sperm motility determined by the visual method is one of the limiting factors for evaluating ejaculates and taking them into further procedure (Kunowska-Słószarz & Makowska, 2011; Savić et al., 2017; Jotanović & Savić, 2017), because ejaculates with insufficient progressive sperm motility (generally below 70%) are rejected. The exception to this rule includes ejaculates of particularly genetically valuable boars, such as breeds that are classified as genetic resources or boars of particular importance in terms of selection. Miclea et al. (2007) found progressive sperm motility of 72.85% at a frequency of ejaculate collection once a week, which was lower than all values found in this study. Umesiobi (2010) found that progressive sperm motility was 87.1% in ejaculates collected

every four days, which is at the upper limit of the range of values found in this study.

Stančić et al. (2003) examined the influence of season, breed, and age of boar on ejaculate quality parameters and found progressive sperm motility ranging from 75% to 83%, which is consistent with the results of this study. Looking at the results of this study, which range from very low to higher ejaculation rates, and the studies of Miclea et al. (2007), Umesiobi (2010) and Savić et al. (2016) in which the frequencies go from high (daily collection) to lower (collection once a week), it can be said that, in order to preserve the percentage of progressive sperm motility, the most optimal frequency of ejaculate collection should be from three times in two weeks to once a week (four to six times a month).

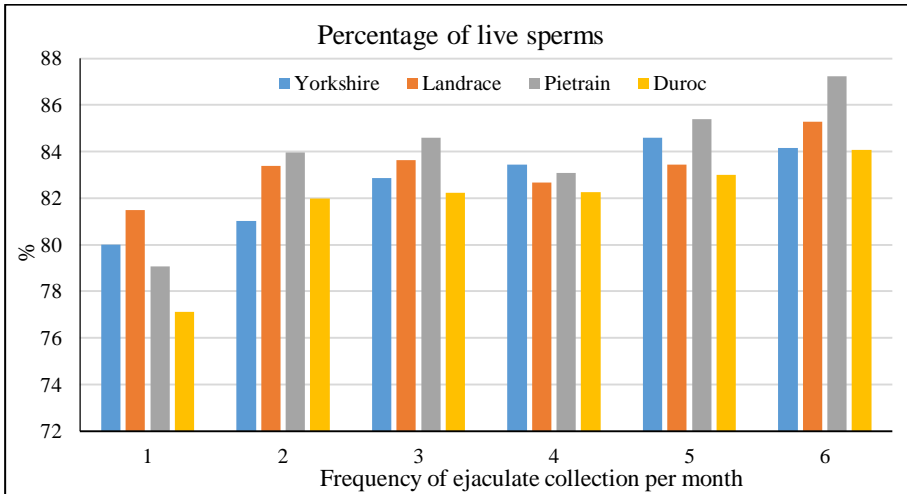
The average progressive motility of sperms in ejaculates in different boar breeds according to the collection frequency is presented in Graph 4.



Graph 4. Progressive motility of sperms in different boar breeds according to the ejaculate collection frequency (source: Sarajlić, 2018)

According to the literature, frequency of ejaculate collection has a generally positive effect on the percentage of progressive sperm motility, as frequent emptying of the sperm reserve in the epididymis enables their faster flow and shorter retention, which generally has a beneficial effect on the sperm vitality. The results of this study confirm the stated, because in all breeds examined a positive relationship was found between the frequency of ejaculate collection and the percentage of progressive sperm motility (Graph 4).

The average percentage of live sperms in ejaculates in different boar breeds according to the collection frequency is presented in Graph 5.



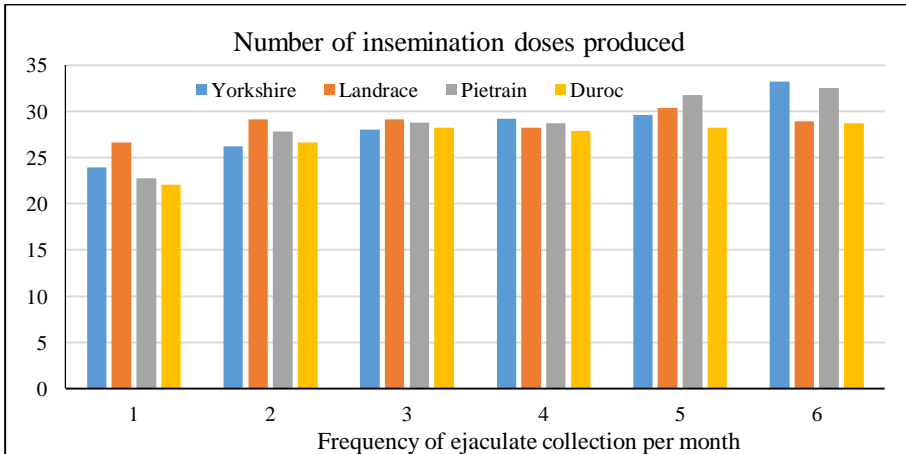
Graph 5. Percentage of live sperms in ejaculates of different boar breeds according to the ejaculate collection frequency (source: Sarajlić, 2018)

The results of this study generally indicate a slight increase in the percentage of live sperms with an increase in the ejaculate collection in all breeds included (Graph 5), which can be attributed to the effect of faster discharge of spermatozoa from reserves, i.e. their faster flow and shorter retention in the epididymis. It is important to emphasize that the percentage of live sperm in most of the boar groups was maintained over 80%, which indicates high fertilization potential of ejaculates and the insemination doses produced. Given the characteristics of spermatogenesis in boars and the time required for maturation of young spermatozoa, it is expected that a further increase in ejaculate collection, especially if lasting for a long period, would result in an increase in the total number of young and immature spermatozoa, and an increase in the proportion of sperm with different deformities of the head and tail, which would adversely affect the overall fertilization capacity of the ejaculate and insemination doses produced.

The number of insemination doses that can be produced from one ejaculate is determined by the total number of sperm in the ejaculate and the required number of sperm in the insemination dose (Stančić et al., 2003; Jotanović & Savić, 2017; Savić et al., 2017). Although each reproductive centre has its own quality standards for insemination doses, it is a common practice that a 100 ml insemination dose, intended for intracervical insemination, should contain at least

three billion spermatozoa, which corresponds to a sperm concentration of $30 \times 10^6/\text{ml}$ (Stančić & Veselinović, 2002).

The average number of insemination doses produced from ejaculates of different boar breeds according to the collection frequency is presented in Graph 6.



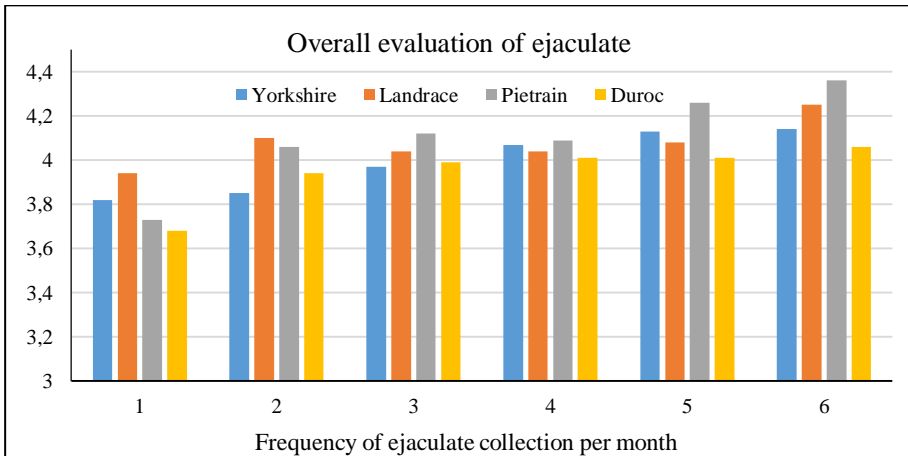
Graph 6. Number of insemination doses produced from ejaculates of different boar breeds according to the ejaculate collection frequency (source: Sarajlić, 2018)

The results of this study indicate a general increase in the number of insemination doses produced with an increase in the frequency of ejaculate uptake, which is certainly a consequence of an increase in ejaculate volume and total sperm count in the ejaculate. In addition, when interpreting the results of this study in terms of the number of insemination doses produced, it should be taken into account that this parameter also varied depending on the demand for doses of a particular breed and boar, which is why the reproductive centre needed to dilute their ejaculates to a greater or lesser extent, to produce more or less insemination doses, in accordance with market demands.

The average overall evaluation of ejaculates in different boar breeds according to the collection frequency is presented in Graph 7.

On the basis of the values of all the above quality parameters, a final ejaculate evaluation is made, which is essential for the reproductive centres business, the selection of boars, and the planning of their reproductive exploitation. Since each centre has its own quality standards for the insemination doses produced, this rating is formed by an expert involved in ejaculate collection and dilution, as well as the production of insemination doses. In addition to the quality of ejaculates, boar's libido and other parameters are taken into account when final ejaculate evaluation is made, which forms the

basis for planning their use for insemination of sows (Savić et al., 2013, Savić et al., 2017).



Graph 7. Average overall evaluation of ejaculates in different boar breeds according to the ejaculate collection frequency (source: Sarajlić, 2018)

The results of this study indicate a slight increase in the average ejaculate score with an increase in the frequency of ejaculate uptake, most likely as a result of an increase in the volume and total number of spermatozoa in the ejaculate, which allows producing more insemination doses of satisfactory quality.

Conclusion

The results of this study indicate the need to observe the quality of ejaculate from several aspects, in order to determine the optimal model of reproductive exploitation for each particular boar, i.e. the frequency of taking the ejaculate by which the maximum number of insemination doses of the desired quality is obtained, and thus an appropriate economic effect. To do so, it is necessary to take into account the breed or hybrid of boar, its age, the way of keeping, the nutrition, the season, the libido, and other factors that can influence the quality of ejaculate, and to plan the reproductive exploitation of the boar so that, at the annual level and during its production life, it gives the maximum number of insemination doses of satisfactory quality. By applying this approach to the reproductive exploitation of boar, it is possible to maximize the exploitation of the reproductive potential of genetically high-quality boars, while reducing the costs of their upbringing, accommodation and keeping, which would

mean achieving additional economic profit for reproductive centres and pig farms.

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Утицај учесталости узимања на параметре квалитета ејакулата нераста

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Сажетак

Циљ истраживања био је да се испита утицај учесталости узимања ејакулата нераста на параметре његовог квалитета. Истраживање је обухватило период од три године и четири најчешће расе свиња на нашем подручју. Укупно је узето 1705 ејакулата од нерастова расе јоркшир, 1693 ејакулата од нерастова расе ландрас, 1106 ејакулата од пиетрен нерастова и 669 ејакулата од нерастова расе дурок. У оквиру сваке расе, формиране су групе према учесталости узимања ејакулата (једном, два, три, четири, пет или шест пута мјесечно). Оцјена квалитета ејакулата извршена је у лабораторији репроцентра, у складу са њиховим стандардима квалитета. Параметри квалитета који су утврђени били су запремина ејакулата, концентрација сперматозоида, прогресивна покретљивост сперматозоида, број живих сперматозоида, број доза по ејакулату и укупна оцјена ејакулата. Резултати овог истраживања показују да се запремина ејакулата и прогресивна покретљивост сперматозоида повећавају са повећањем учесталости узимања ејакулата, док концентрација сперматозоида опада, осим код нерастова пиетрена. Најбољи резултати су установљени код ејакулата узиманих 5-6 пута мјесечно. Највећа просјечна запремина ејакулата установљена је код нерастова ландраса (292,22 ml), уз учесталост узимања ејакулата од пет пута мјесечно. Највећа просјечна прогресивна покретљивост сперматозоида била је 92% код нерастова јоркшира, при учесталости узимања од шест пута мјесечно, док је највећа концентрација сперматозоида установљена код нерастова дурика при учесталости узимања једном месечно (447,32 mil./ml). Резултати овог истраживања указују на то да учесталост узимања утиче на параметре квалитета ејакулата нераста.

Кључне ријечи: ејакулат, учесталост узимања, квалитет сперме.

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