

REGULAR ARTICLE

SOWS FERTILITY AFTER INTRACERVICAL OR POSTCERVICAL ARTIFICIAL INSEMINATION (AI) IN WORM AND COLD SEASON

Ivan Stančić*, Blagoje Stančić, Saša Dragin, Ivan Radović, Aleksandar Božić

Adress: Ivan Stančić, University of Novi Sad, Faculty of Agriculture, Trg D. Obradovića 8, 21000 Novi Sad, Serbia, Phone: +381 21 485-34-96.

*Corresponding author: <u>dr.ivan.stancic@gmail.com</u>

ABSTRACT

Reduced fertility of boars and sows during the warmer period of the year significantly reduces the reproductive efficiency of pigs in intensive production. The aim of this study was to determine whether the application of intrauterine (postcervical) artificial insemination (AI), with twice reduced dose volume (50 mL) and the number of spermatozoa per dose $(2x10^9)$, compared to the classical intracervical insemination (dose volume 50 mL with $4x10^9$ spermatozoa), can increase the fertility of sows inseminated in the warm season of the year. After classical intracervical insemination, farrowing rate was significantly (P<0.01) lower in the warmer (80% and 76.7%), compared to the cold period (66.7% and 53.3%), using both spermatozoa number per dose $(4x10^9 \text{ or } 2x10^9)$. Using new intrauterine (postcervical) AI technology, the farrowing rate was not significantly different (P>0.05), neither depending on the season, or depending on the number of spermatozoa per dose (78% and 75% in warm, or 86% and 83% in cold season of the year). The results show that the use of intrauterine insemination, with doses twice reduced in volume and sperm number, can significantly reduce the negative impact of the warm season on sows fertility.

Key words: Intracervical, postcervical, AI, fertility, season, sow

INTRODUCTION

Significantly lower parameters of sows fertility within the warmer part of the year, is permanently demonstrated in the most intensive pig farm units, all over the world, during the past 40 years. Reduced sows fertility is manifested in the increased occurrence of anestrous, prolonged weaning-to-estrus interval, irregular estrus cycles, reduced farrowing rate and lower litter size (Almond, 1992; Stančić, 2005, Xue et al., 1994; Stančić, 1994; Bassett et al., 2001; Prunier and Quesnel, 2000; Stančić et al., 2010). The season influences the variation of the native sperm quality parameters (ejaculate volume, concentration and total sperm in the ejaculate, and progressive motility). It is known that the parameters of sperm quality are much worse during the warmer periods of the year. The practical consequence is to obtain a smaller number of insemination doses per ejaculate, with reduced ejaculate fertilization capacity, on the one side, and reduced sows fertility, on the other side (Setchell, 1998; Corcuera et al., 2002; Stančić et al., 2003; Okere, 2003). Fertility decreasing during the warm season, is associated with negative effects of high ambiental temperature and extended daily photoperiod on reproductive function in pigs (Gordon, 1997).

In the classical technology of artificial insemination diluted liquid semen doses, volume 100 mL, with 3 to 4x10⁹ progressively motile sperm is used (Almin et al., 2006; Stančić et al., 2009). In that case, it can be obtained average 21 insemination doses per one ejaculate (Singleton, 2001). Insemination dose is usually preserved 1 to 2 days at 17°C, before using for insemination (Johnson et al., 2000). However, due to lower values of semen fertilization capacity parameters, during the warmer season, significant lower doses number can be formed per ejaculate, compared to the cold season. The formation of double insemination doses number per ejaculate, with reduced dose volume (50 mL) and sperm number (2x10⁹), could be the solution of this problem. On this way, a sufficient number of insemination doses per one ejaculate, can be obtained during the warm season. Such reduced dose is possible to use with intrauterine insemination technology, while the achieved level of sows fertility is similar to that obtained using classical intracervical insemination (doses of 100 mL volume, with 4x10⁹ spermatozoa) (Vansickle, 2002; Roseboom et al., 2004; Mesalira et al., 2005, Serret et al., 2005; Stančić et al., 2006; Stančić et al., 2007; Stančić et al., 2008; Stančić et al., 2010).

The aim of this study was to determine whether intrauterine insemination with doses of twice reduced volume and sperm number increase the sow fertility during the warm period of the year.

MATERIALS AND METHODS

Classical, intracervical insemination and intrauterine (postcervical) insemination was performed with dose volume of 50 mL, contained $4x10^9$ or $2x10^9$ progressively motile sperm. Insemination was performed during the warm (May-September) and cold (October-April) season. The total of 480 sows, second to the fifth farrowing parity, (60 per each group) was inseminated by each insemination procedure and dose sperm number (60x4x2 = 480). Insemination was performed in the estrus detected at day 5 after weaning. Lactation lasted 28 days. The first insemination was carried out 12h, and second 36h after standing estrus detection. For conventional insemination were used sterile disposable catheters (Foamtip safe blue®), and for intrauterine insemination were used sterile disposable catheters Foamtip "Verona"® (Minitübe, Germany). Semen were diluted with BTS1, for short-term storage of liquid diluted boar semen (Minitübe, Germany). Value for farrowing rate and litter size were recorded. For the statistical analysis, "Statistica 10" software was used.

RESULTS

Using classical (intracervical) insemination, by both $4x10^9$ or $2x10^9$ progressively motile sperm per dose, achieved farrowing rate was statistically significant (P<0.01) lower in the warm season (66.7% and 53.3%) compared to the cold season (80% and 76.7%). However, farrowing rate, after postcervical (intrauterine) insemination was not significantly (P>0.05) varied depending on the season (78.3% and 75% in the warm or 86.7% and 83.3% in the cold season). The farrowing rate, after intrauterine insemination, were statistically significant (P<0.01) higher, compared to intracervical insemination, both in the warm and cold season (Table 1).

Tabla 1	Effort of in	acamination	mothad an	d gnorm nu	mhar i da	ca on for	rowing rate
I able 1	Ellect of II	iscillilation	i incinou ai	ia sperm na	moer ruo	se on iai	rowing rate

	Season of year				
Method of insemination	War	m	Cold		
	4x10 ⁹	2x10 ⁹	4x10 ⁹	2x10 ⁹	
Classic	$66.7\%^{\mathrm{B}}$	53.3% ^B	80.0% ^A	76.7% ^A	
Classic	(40/60)	(30/60)	(48/60)	(44/60)	
Intrauterine	78.3% ^A	75.0% ^A	86.7% ^A	83.3% ^A	
mirauternie	(47/60)	(45/60)	(52/60)	(50/60)	

^{AB} Values with different superscript are statistically significant (P<0.01).

In parenthesis: (No. farrowed/No. inseminated).

Sperm number in a dose had no effect on farrowing rate, within the same season. However, the intracervical insemination with reduced dose sperm number (from 4 to $2x10^9$), in the warm season, statistically significant (P<0.01) decrease farrowing rate (53.3%), compared to the cold season (76.7%), which is not the case with intrauterine insemination method (Table 1).

Table 2 Average litter size at farrowing

Method of	Litter size (n)	Season of year				
insemination		Wa	ırm	Cold		
msemmation		4x10 ⁹	2x10 ⁹	4x10 ⁹	2x10 ⁹	
	Live	9.45 ^A	9.53 ^A	10.04^{B}	10.64^{B}	
Classic	Dead	0.55	0.59	0.46	0.54	
	Total	10.00	10.12	10.50	11.18	
	Live	10.10^{B}	10.35^{B}	10.48^{B}	10.58^{B}	
Intrauterine	Dead	0.50	0.60	0.46	0.48	
	Total	10.60	10.95	10.94	11.06	

AB Values with different superscript are statistically significant (P<0.01)

The average number of live born piglets per litter, after intracervical insemination, was statistically significant (P<0.01) higher in the cold (10.04 and 10.64) compared to the warm season of the year (9.45 and 9.53). After intrauterine insemination, the average number of live born piglets did not differ depending on the season (10.10, 10.35, 10.48 and 10.58), but these

values were significantly (P<0.01) higher than those obtained after intracervical insemination (Table 2).

DISCUSSION

Our results clearly show that the intrauterine insemination, with double reduced dose volume (50 ml) and sperm number ($2x10^9$), result in statistically significant (P<0.01) higher farrowing rate, in the warm (75%) and in the cold season (83%), compared to the intracevical insemination (53% warm and 77% in cold season). The average number of live born piglets per litter was significantly (P<0.01) higher after intrauterine insemination with reduced dose sperm number, compared to the intracervical insemination, only in the warm season (10.35 vs. 9.53).

Using intrauterine (postcervical) insemination with different doses volume (100, 85, 50, 30 and 20 mL) and different sperm number per dose (4, 3, 1.5 and 1x10⁹), result with 78 and 96% farrowing rate and 9 to 12 live born piglets per litter (Vansickle, 2002; Roseboom et al., 2004; Mesalira et al., 2005, Serret et al., 2005; Stančić et al., 2006; Stančić et al., 2007; Stančić et al., 2008; Stančić et al., 2010). By the sperm deposition in the cranial parts of the female reproductive tract (the body of the uterus, uterine horns, uterotubal junction or fallopian tubes), the volume of insemination dose and sperm number per dose can be radically reduced, with the same or higher fertility of inseminated sows, compared with the classical intracervical insemination (Mezalira et al., 2005; Stančić et al., 2007). Numerous studies show that the optimal value of the sows fertility has been achieved when insemination is performed approximately 24 hours before ovulation, with doses contained 2x10⁹ spermatozoa. Increasing the sperm number per dose does not affect sows fertility, while reducing the number of sperm under the 2x10⁹ leads to a decrease in sows fertility parameters (Knox, 2004; Stančić et al., 2007; Stančić et al., 2010).

Although numerous studies (Liao et al., 1996; Kunavongkrit et al., 2005; Ciereszko et al., 2000; Jankevičiute and Žilinskas, 2002; Chukwuemeka et al., 2005) consistently showed significantly lower values of the boar sperm fertilizing capacity parameters during warm period, bath the precise physiological mechanism of this phenomenon is not fully understood. However, most researches shows that this is a consequence of the elevated ambient temperature (Suriyasomboon et al., 2004) and extended daily photoperiod (Sancho et al., 2004), during the warm periods of the year, on the process of spermatogenesis and testosterone synthesis. In addition, some researches suggest that this phenomenon could be

due to genetic heritage of the domestic breeds from their wild relatives. Namely, it is known that wild boars are highly seasonal sexually active, and that they have the best quality of semen during the breeding season, which lasts from late fall to early winter (Kozdrowski and Dubiel, 2004; Macchi *et al.*, 2010).

Due to significantly lower values of sperm fertilizing capacity parameters, during the warm season, it can be prepared significant lower classic insemination doses number per one ejaculate (100 mL volume with $4x10^9$ sperm), compared to the cold period of the year. The formation of twice more doses number from the same ejaculate, requires twice reduction of the sperm number in a dose, and double degree of ejaculate dilution proportion. However, using a twice smaller dose volume (50 mL) and sperm cells number (2x10⁹), it is not necessary to double the degree of ejaculate dilution. Adding large amounts of artificial extender in native semen, leads to a reduction in sperm progressive motility and agglutination (Stančić et al., 2003). This is due to reduction in amount of native protein and natural antioxidants, and other natural ingredients of seminal plasma, which are essential for the normal integrity and function of sperm cell membrane (Kommisurd et al., 2002; Boe-Hansen et al., 2005). In addition, the sperm plasma has a significant impact on the process of sperm transport in the female reproductive tract (Stančić et al., 2012) and is a significant factor in the regulation time of ovulation (Weitze et al., 1994). On the other hand, it was found that the semen of a large number of boar does not tolerate the increasing degree of dilution. Namely, the results of numerous studies indicate that semen in only 20 to 30% of boars retained $\geq 65\%$ progressive motility during 72h of storage, on +17°C, in dilution rate 1:4 (Weitze, 1990; Stančić et al., 2003).

Practical contribution to the results of our research consists in the fact that twice a smaller dose volume and sperm count can be used in the application of postcervical (intrauterine) insemination technology, without significant decrease in sow fertility. Thus it is possible to get the same or similar number of insemination doses per ejaculate during warm and cold seasons and, consequently, to significantly reduce the negative impact of warm season on reduced the number and quality of insemination doses.

CONCLUSION

Based on obtained results, it can be concluded:

- 1. Farrowing rate was significantly lower in the warm, against the cold season of the year, using both classical (intracervical) or intrauterine (postcervical) insemination of sows.
- 2. Using postcervical insemination, with twice reduced dose volume and sperm number, it is possible to significantly increase the sows farrowing rate in the warm season, compared with classical (intracervical) insemination.
- 3. By using intrauterine insemination, it is possible to increase the reproductive efficiency of boars and sows reproductive performance, during the warmer periods of the year.

Acknowledgments: This work is a part of the project supported by Ministry of Education, Science and Technological Development of the Republic of Serbia, TR-31081, 2011.-2014.

REFERENCES

ALMIN, K. - PELTONIEMI, O.A.T. - KOSKINEN, E. - ANDERSSON, M. 2006. Porcine field fertility with two different insemination doses and the effect of sperm morphology. In *Reproduction of Domestic Animals*, vol. 41, 2006, p. 210-213.

ALMOND, W.G. 1992. Seasonal infertility in female pigs. In *Healthy Hogs*, vol. 1, 1992, p.1-5.

BASSETT, J.M. - BRAY, C.J. - SHARPE, C.E. 2001. Reproductive seasonality in domestic sows kept outdoors without boars. In *Reproduction*, vol. 121, 2001, p. 613–629.

BOE-HANSEN, B., G. - ERSBØLL, K.A. - GREVE, T. - CHRISTENSEN, P. 2005. Increasing storage time of extended boar semen reduces sperm DNA integrity. In *Theriogenology*, vol. 63, 2005, p. 2006-2019.

CHUKWUEMEKA, O. - AVIS, J. - EZEKWE, M. 2005. Seasonal and genotype variations in libido, semen production and quality in artificial insemination boars. In *Journal of Animal Veterinary Advances*, vol. 4, 2005, p. 885-888.

CIERESZKO, A. - OTTOBRE, S.J. - GLOGOWSKI, J. 2000. Effect of season and breed on sperm acrosin activity and semen quality of boars. In *Animal Reproduction Sciences*, vol. 64, 2000, p. 89-96.

CORCUERA, D.B. - HERNANDEZ,G.L.R. - DE ALBA, C. - MARTIN RILLO, S. 2002. Relationship of environmental temperature and boar facilities with semen quality. In *Livestock Production Sciences*, vol. 74, 2002, p. 55-62.

GORDON, I. 1997. In: Controlled Reproduction in Pigs (Vol. 3). CAB Int., Oxon, UK.1997.

JANKEVIČIUTE, N. - ŽILINSKAS, H. 2002. Influence of some factors on semen quality of different breeds of boars. In *Veterinaria ir Zootechnika*, (abstr.). vol. 19, 2002, p. 41,

JOHNSON, L.A. - WEITZE, K.F. - FISER, P. - MAXWELL, W.M.C. 2000. Storage of boar semen. In *Animal Reproduction Sciences*, vol. 62, 2000, p. 143-172.

KNOX, V.R. 2004. Practicalities and pitfalls of semen evaluation. In *Advences in Pork Production*, vol. 15, 2004, p. 315-322.

KOZDROWSKI, R. - DUBIEL, A. 2004. The effect of season on the properties of wild boar (*Sus scrofa* L.) semen. In *Animal Reproduction Sciences*, vol. 80, 2004, p. 281–289.

KUNAVONGKRIT, A. - SURIYASOMBOON, A. - LUNDEHEIM, N. - HEARD T.W. - EINARSSON, S. 2005. Management and sperm production of boars under differing environmental conditions. In *Theriogenology*, vol. 63, 2005, p. 657-667.

KOMMISRUD, E. - PAULENZ, H. - SEHESTED, E. - GREVLE, S.I. 2002. Influence of boar and semen parameters on motility and acrosome integrity in liquid boar semen stored for five days. In *Acta Veterinaria Scandinavica*, vol. 43,2002, p. 49-55.

LIAO, C.W. - SHEN, T.F. - CHYR, S.C. 1996. Monthly changes in the semen characteristics of Duroc boars. In *Journal Taiwan Livestook Research*, vol. 29, 1996, p. 137-144.

MACCHI, E. - STARVAGGI CUCUZZA, A. - P. BADINO, P. - ODORE, R.F. - RE, F. - BEVILACQUA, L. - MALFATTI, A. 2010. Seasonality of reproduction in wild boar (Sus scrofa) assessed by fecal and plasmatic steroids. In *Theriogenology*, vol. 73, 2010, p. 1230–1237

MEZALIRA, A. - DALLANORA, D. - BERNARDI, L.M. - WENTZ, I. - BORTOLOZZO, F.P. 2005. Influence of sperm cell dose and post-insemination backflow on reproductive performance of intrauterine inseminated sows. In *Reproduction of Domestic Animals*, vol. 40, 2005, p. 1-5.

OKERE, C. 2003. Seasonal infertility in modern domestic pigs: What's News? In *Tch Service*, vol. 6, 2003, p. 1-6.

PRUNIER, A. - QUESNEL, H. 2000. Influence of the nutritional status on ovarian development in female pigs. In *Animal Reproduction Sciences*, vol. 32, 2000, p.185–197.

ROSEBOOM, J.K. - REICKS, L.D. - WILSON, E.M. 2004. The reproductive performance and factors affecting on-farm application of low-dose intrauterine deposition of semen in sows. In *Journal of Animal Sciences*, vol. 82, 2004, p. 2164-2168.

SERRET, C.G. - ALVARENGA, M.V.F. - CÓRIA, A.L.P. - DIAS, C.P. - CORCINI, C.D. - CORRÊA, M.N. - DESCHAMPS, J.C. - BIANCHI, I. - LUCIA, T. jr. 2005. Intrauterine artificial insemination of swine with different sperm concentrations, parities, and methods for prediction of ovulation. In *Animal Reproduction*, vol. 2, 2005, p. 250-256.

SANCHO, S. - PINART, E. - BRIZ, M. - GARCIA-GIL, N. - BADIA, E. - BASSOLS, J. - KADAR, E. - PRUNEDA, A. - BUSSALLEU, E. - YESTE, M. - COLL, M.G. - BONET, S. 2004. Semen quality of postpubertal boars during increasing and decreasing natural photoperiods. In *Theriogenology*, vol. 62, 2004, p. 1271-1282.

SETCHELL, P.B. 1998. The Parkers Lecture: Heat and the testis. In *Journal of Reproduction Fertility*., vol. 114, 1998, p. 179-194.

SINGELTON, W.L. 2001. State of the art in artificial insemination in the Uniteted States. In *Theriogenelogy*, vol. 56, 2001, p. 1305-1310.

STANČIĆ, B. - BOŽIĆ, A. - RADOVIĆ, I. - GRAFENAU, P., sen. - PIVKO, J. - CHRENEK, P. - STANČIĆ, I. 2007. Sows artificial insemination with reduced spermatozoa number in dose (a rewiev). In *Contemporary Agriculture*, vol. 56, 2007, p. 1-11.

STANČIĆ, B. - GAGRČIN, M. - RADOVIĆ, I. - STANČIĆ, I. 2008. Sows fertility after transcervical intrauterine insemination (the summarize of ours results). In *Lucrari stiintifice Zootehnie si Biotehnologii (Romania)*, vol. 41, 2008, p. 624-628.

STANČIĆ, B. - GAGRČIN, M. - RADOVIĆ, I. 2003. Effect of season, breed and age of boar on sperm quolity. 2. Extended sperm. In *Biotechnology in Animal Husbandry*, vol. 19, 2003, p. 25-29.

STANČIĆ, B. - RADOVIĆ, I. - BOŽIĆ, A. - GAGRČIN, M. - ANDERSON, R. 2009. Sow fertility after conventional AI with insemination doses of various volumes and spermatozoa number. In *Contemporary Agriculture*, vol. 58, 2009, p. 62-66.

STANČIĆ, B. - RADOVIĆ, I. - STANČIĆ, I. - DRAGIN, S. - BOŽIĆ, A. - GVOZDIĆ, D. 2010. Fertility of sows after intracervical or intrauterine insemination with different spermatozoa number in reduced volumen doses. In *Acta veterinaria (Beograde)*, vol. 60, 2010, p. 257-262.

STANČIĆ, B. - RADOVIĆ, I. - STANČIĆ, I. - GAGRČIN, M. - BOŽIĆ, A. - KRAGIĆ, S. - GRAFENAU, P. jr. - CHRENEK, P. - PIVKO, J. 2006. Fertilitet krmača posle intracervikalne

i intrauterine inseminacije dozama redukovanog volumena. In *Biotechnology in Animal Husbandry*, vol. 22, 2006, special issue, pp. 273-281.

STANČIĆ, B. 1994: Factors that influence on some parameters of sows reproductive performance. In *Veterinary glasnik (Belgrade)*, vol. 48, 1994, p. 345-472.

STANČIĆ, B. 2005. In Pig Reproduction (monography). University of Novi Sad, Faculty of Agriculture, 2005.

STANČIĆ, I. - DRAGIN, S. - STANKOVIĆ, B. - JOTANOVIĆ, S. 2102. Effect of protein contents in seminal plasma on sperm motility in diluted boar semen. *Proc. 1st International Symposium on Animal Science, November 8 – 10, 2012., Belgrade, Serbia. Pp.149-154.*

SURIYASOMBOON, A. - LUNDEHEIM, N. - KUNAVONGKRIT, A. - EINARSSON, S. 2004. Effect of temperature and humidity on sperm production in duroc boars under different housing systems in Thailand. *In Livestoock of Production Science*, vol. 89, 2004, p. 19-31.

VANSICKLE, J. 2002. New Insemination Tool Still Unproven. In *National Hog Farmer*, vol. 7, 2002, p. 1-3.

WEITZE, F.K. 1990. The use of long-term extender in pig AI: A view of the international situation. In *Pig News and Information*, vol. 11, 1990, p. 23-26.

WEITZE, K.F. - WAGNER-REITCHEL, H. - WABERSKI, D. - RICHTER, L. - KRIETER, J. 1994. The onset of oestrus after weaning, oestrus duration and ovulation as major factors in AI timing in sows. In *Reproduction of Domestic Animals*, 29, 1994, p. 433-439.

XUE, J.L. - DIAL, G.D. - MARSH, W.E. - DAVIES, P.R. 1994. Multiple manifestations of season on reproductive performance of commercial swine. In *JAVMA*., vol. 204, 1994, p. 1486–1489.