



*Original Contribution*

## DEEP INTRAUTERINE AND TRANSCERVICAL INSEMINATION OF SOWS AND GILTS

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### ABSTRACT

The use of low count spermatozoa in low semen volume to artificially inseminate sows and gilts is a technique that should introduce a more efficient use of semen doses from genetically superior boars. The objective of this experiment was to determine whether reproductive performance of sows and gilts would be influenced by inseminating  $0.15 \times 10^9$  sperm cells per dose with a deep intrauterine horn catheter, inseminating  $1.5 \times 10^9$  sperm cells per dose with an intrauterine body catheter, or inseminating  $3.0 \times 10^9$  sperm cells per dose with a traditional cervical catheter. This work demonstrated that farrowing rate was not significantly reduced when sows and gilts were artificially inseminated by transcervical insemination ( $1.5 \times 10^9$  sperm cells per dose) or inseminated by deep intrauterine horn method ( $0.15 \times 10^9$  sperm cells per dose) compared with a traditional cervical insemination ( $3.0 \times 10^9$  sperm cells per dose). However, total number of piglets born per litter was significantly reduced when sows were inseminated by deep intrauterine horn method with  $0.15 \times 10^9$  sperm cells per dose compared with sows inseminated by cervical catheter ( $3.0 \times 10^9$  sperm cells per dose).

**Key words:** sows, gilts, deep intrauterine insemination, transcervical insemination, farrowing rate.

### INTRODUCTION

Artificial insemination (AI) technology is widely used in the pig industry. During the early years when artificial insemination was being studied, it was recommended that each sow be inseminated with a large volume of semen (50 to 200 ml) and a high number of sperm cells per dose (5 to 10 billion) to ensure high fertility and fecundity (1-5). A survey of pig farmers found that on average a sow receives 2.2 insemination doses per service with each insemination dose containing 2.5 to  $4 \times 10^9$  sperm cells in a 70 to 100 ml volume (6). These numbers suggest sows mated by AI receive 5.5 to  $8.8 \times 10^9$  sperm cells per service. Pig farmers could make a considerable economic savings on the purchase of semen if the number of sperm

cells per dose and doses per mating could be reduced without affecting reproductive performance of the sows and gilts (7). Currently, the two methods used to reduce the volume of semen inseminated and numbers of sperm cells per dose are intrauterine body insemination (7-9) and non-surgical deep intrauterine horn insemination (10, 11). The results of previous studies indicate that a 3 to 20 reduction in the total number of spermatozoa did not decrease fertility when semen was deposited into the uterine body or into the upper first third of one uterine horn.

The objective of this study was to confirm the effect of low-dose application of deep intrauterine horn (DUHI), transcervical-uterine body (TCUB), and cervical insemination (CI) of sows and gilts on reproductive performance in a pig farm.

### MATERIAL AND METHOD

#### *Animals*

This study used Danube White females (132 sows and 47 gilts) that were housed on one

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pig farm. The gilts at time of first service were 237 to 266 days of age and weighed approximately 110 kg. The body weight of the sows ranged from 250 to 280 kg. The lactation length was 25 days. Sows were weaned into individual gestation stalls. The stalls were 0.61 m wide and 2.13 m long. Depending on sow's body conditions, the sows were daily fed 1.80 to 2,30 kg of balanced diet for gestating sows. Gilts were housed in pens containing 6 to 8 individual animals. The amount of space per animal ranged from 0.80 to 1.00 square metres. The gilts were fed approximately 2.6 kg per animal per day. All sows and gilts were provided *ad libitum* access to water. All animals were observed daily for aspects concerning their health, welfare and body conditions.

### **Treatment**

Semen was collected no more than twice per week from Duroc boars (DanBred<sup>INT</sup>) housed at the same farm by using the gloved hand method. Ejaculates were individually assessed for volume, number of sperm cells, and percent of motile spermatozoa. The spermatozoa and minimum standards of an acceptable ejaculate were: volume - > 80 ml; colour - milky; odour - normal; number of sperm per ml - >  $100 \times 10^6$ ; motility - > 70 %; agglutination - < 30 %; and pH - 7.4 to 7.8. Volume (without gel fraction) was determined by using a graduated glass vessel with precision of 0.01 ml. Number of sperm cells was determined by using a Thoma counting chamber. Percentage of motile spermatozoa (0 to 100 %) was subjectively evaluated with a light microscope at a magnification of 100X. Spermatozoa were diluted in BTS extender (Magapor, Ejea de los Caballeros, Spain) and packaged in 100 ml plastic bottles and cooled within 20 to 30 minutes after dilution. The dilution rate of pooled semen was 1:3 - 1:5 (v/v). The number of sperm cells per dose was checked after dilution. Diluted sperm was stored in a cooler unit at 17° C until used within 24 to 48 hours after collection.

### **Oestrus detection and artificial insemination**

Oestrus detection of weaned sows was performed twice per day (09:30 to 10:30 and 15:00 to 16:00) starting on day 3 after weaning. Duration of oestrus was not recorded. Eight to 10 weaned sows were placed in a pen and checked for oestrus using a mature boar. Gilts were exposed twice per day to the boar to facilitate detection of

oestrus. Animals that showed a standing reflex were used for the experiments and were randomly allotted to each treatment within weaning-to-oestrus-interval (WEI). Stored semen was evaluated for motility prior to insemination and >65% was motility required for use. Sows and gilts were randomly assigned to one of the following three treatment groups: Treatment 1 (n = 48) - DUHI of  $0.15 \times 10^9$  spermatozoa in 5 ml with FIRFLEX<sup>®</sup> catheter (Magapor, Ejea de los Caballeros, Spain), Treatment 2 (n = 32 sows, n = 19 gilts) - TCUB insemination of  $1.5 \times 10^9$  spermatozoa in 50 ml with Magaplus<sup>™</sup> catheter (Magapor, Ejea de los Caballeros, Spain), and Treatment 3 (n = 52 sows; n = 28 gilts) - CI of  $3.0 \times 10^9$  spermatozoa in 100 ml with Foam type catheter (Magapor, Ejea de los Caballeros, Spain). Deep uterine horn inseminations were performed on each sow in gestation crates without sedation. The DUHI procedure was accomplished by first inserting a Foam type catheter through the vagina into the cervix, secondly inserting the flexible FIRFLEX<sup>®</sup> catheter (1.8 m in length, 4 mm in diameter) through the Foam type catheter, and thirdly passing the FIRFLEX<sup>®</sup> catheter carefully through the cervical canal and moving it forward in one uterine horn until its total length had been inserted. Transcervical inseminations were performed by carefully pushing the Magaplus<sup>™</sup> catheter (70 cm in length, 4.3 mm in diameter) through the rings of the cervix into the body of the uterus. Cervical inseminations were performed in the traditional manner by inserting a Foam type catheter through the vagina into the cervix. Animals were inseminated twice at 8 h and 24 h after onset of standing reflex. Pregnancy was diagnosed at 28 to 30 days after insemination by ultrasonography (PREG-TONE<sup>®</sup>, Renco Corporation, USA). All pregnant animals were allowed to carry litters to term. The experimental period was from October 2003 to June 2004. Data were recorded for weaning-to-oestrus interval, farrowing rate, total number of piglets born per litter, number of piglets born alive per litter, and birth weight of piglets.

### **Statistical analysis**

Prior to data analysis sows were categorized into two weaning-to-oestrus interval (WEI) of  $\leq 6$  days or  $> 6$  days. A one-way MANOVA with fixed effects statistical analysis was used. The post hoc comparisons were done by LSD test.

The additive model was:

$$y_{ijl} = \mu + \alpha_i + \beta_j + \varepsilon_{ijl},$$

where  $y_{ijl}$  was  $l^{\text{th}}$  observation of investigated trait,  $\alpha_i$  was the effect of the method of artificial insemination (number of spermatozoa per dose),  $\beta_j$  was the effect of the interval between weaning and insemination, and  $\varepsilon_{ijl}$  was the error. In the gilts a one-way ANOVA with fixed effects statistical analysis was used. The additive model was:  $y_{ij} = \mu + \alpha_i + \varepsilon_{ij}$ , where  $y_{ij}$  was the  $j^{\text{th}}$  observation of investigated trait,  $\alpha_i$  was the effect of the method of artificial insemination (number of spermatozoa per dose) and  $\varepsilon_{ij}$  was the error.

All calculations were made with the statistical package, StatSoft® (STATISTICA, Tulsa, OK).

## RESULTS

**Table 1** indicates the reproductive performance of sows artificially inseminated

by DUHI ( $0.15 \times 10^9$  sperm per dose), TCUB ( $1.5 \times 10^9$  sperm per dose) and CI ( $3.0 \times 10^9$  sperm per dose) procedures. The method of insemination did not affect farrowing rate, average number of piglets born alive per litter, fecundity index, or average total birth weight of litter. The average number of total piglets born per litter was greater ( $p < 0.01$ ) for sows inseminated by the CI method compared with sows inseminated by the DUHI method (10.28 vs 8.88 piglets). The average number of total piglets born from sows inseminated by the TCUB method (9.04 piglets) was not significantly different from sows inseminated by DUHI or CI methods. Average birth weight of piglets was lower ( $p < 0.01$ ) for the sows inseminated by CI method compared with sows inseminated by DUHI method (1.43 vs 1.54 kg). Average birth weight of piglets born from sows inseminated by TCUB method was lower ( $p < 0.01$ ) compared with piglets born from sows inseminated by DUHI method.

**Table 1.** Effect of deep intrauterine insemination, transcervical insemination or cervical insemination method on reproductive performance – numbers of sperm per dose (Mean  $\pm$  SE)

Traits	Deep intrauterine insemination $0.150 \times 10^9$ sperm	Transcervical insemination $1.5 \times 10^9$ sperm	Cervical insemination $3.0 \times 10^9$ sperm
Number of sows	48	32	52
Farrowing rate, % <sup>a</sup>	88.99	82.07	90.48
Average total number piglets born per litter	8.88 $\pm$ 0.41 <sup>A</sup>	9.04 $\pm$ 0.73	10.28 $\pm$ 0.39 <sup>A</sup>
Average number of piglets born alive per litter	8.69 $\pm$ 0.42	8.69 $\pm$ 0.69	9.81 $\pm$ 0.37
Fecundity index <sup>b</sup>	773	713	888
Average total birth weight of litter, kg	13.21 $\pm$ 0.57	14.16 $\pm$ 0.96	13.90 $\pm$ 0.58
Average birth weight per piglet, kg	1.54 $\pm$ 0.04 <sup>A</sup>	1.63 $\pm$ 0.05 <sup>B</sup>	1.43 $\pm$ 0.03 <sup>AB</sup>

<sup>a</sup> Farrowing rate is the number of sows that farrowed divided by the number of sows bred

<sup>b</sup> Fecundity index is farrowing rate times average number of piglets born alive per litter times 100

<sup>AB</sup> Same superscripts within row indicate differences between means ( $p < 0.01$ )

**Table 2** indicates the reproductive data according to the weaning-to-oestrus interval within the insemination methods. The method of insemination did not significantly affect farrowing rate, average total number of piglets born per litter, average number of piglets born alive per litter, fecundity index or average birth weight of the litter. Although not significant, sows cycling within 6 days after weaning and inseminated by the DUHI ( $0.15 \times 10^9$  sperm cells) or TCUB ( $1.5 \times 10^9$  sperm cells) method had a greater farrowing rate (DUHI, 96.2 %; TCUB, 86.4%) when compared with sows cycling greater than 6 days after weaning (DUHI, 81.8 %; TCUB,

77.8 %). When sows were inseminated by the CI method ( $3.0 \times 10^9$  sperm cells), there was only a 1.6 percentage point difference in farrowing rate between sows cycling within 6 days and those sows cycling greater than 6 days after weaning. Average birth weight of piglets was less ( $p < 0.01$ ) for sows with a WEI greater than 6 days and inseminated by the CI method (1.37 kg) compared with sows inseminated by DUHI (WEI  $\leq$  6 days, 1.58 kg) or TCUB (WEI  $\leq$  6, 1.61 kg; WEI  $>$  6 days, 1.67 kg). Average birth weight of piglets was less ( $p < 0.01$ ) for sows with a WEI less than 6 days and inseminated by the CI method (1.48 kg) compared with sows

inseminated by TCUB method (WEI > 6 days, 1.67 kg).

**Table 3** indicates the reproductive performance of gilts artificially inseminated by TCUB ( $1.5 \times 10^9$  sperm cells) and CI ( $3.0 \times 10^9$  sperm cells) method. Although not significant, gilts inseminated by CI method had a greater average total number of piglets

born per litter, average number of piglets born alive per litter, and fecundity index compared with gilts inseminated by the TCUB method. Average birth weight per piglet was reduced ( $p < 0.01$ ) for gilts inseminated by the CI method compared with gilts inseminated by the TCUB method (1.28 vs 1.58 kg).

**Table 3.** Reproductive performance of gilts artificially inseminated by transcervical insemination or cervical insemination (Mean  $\pm$  SE)

Parameters	Transcervical insemination ( $1.5 \times 10^9$ sperm per dose)	Cervical insemination ( $3.0 \times 10^9$ sperm per dose)
Number of gilts	19	28
Farrowing rate, % <sup>a</sup>	89.47	85.71
Average total number of piglets born	9.07 $\pm$ 0.76	10.3 $\pm$ 0.57
Average number of piglets born alive	8.69 $\pm$ 0.66	10.0 $\pm$ 0.55
Fecundity index <sup>b</sup>	777	857
Average birth weight per litter, kg	13.5 $\pm$ 0.88	12.7 $\pm$ 0.66
Average birth weight per piglet, kg	1.58 $\pm$ 0.06 <sup>A</sup>	1.28 $\pm$ 0.03 <sup>A</sup>

<sup>a</sup> Farrowing rate is the number of sows that farrowed divided by the number of sows bred

<sup>b</sup> Fecundity index is farrowing rate times average number of piglets born alive per litter times 100

<sup>A</sup> Same superscript within row indicate differences between means ( $p < 0.01$ )

## DISCUSSION

A common goal for most pig production enterprises is to improve efficiency. For artificial insemination, one of the most compelling ways to achieve efficiency is to decrease the number of spermatozoa in each dose of semen without compromising fertility. Currently, most insemination doses contain between 2 to 4 billion viable spermatozoa. A large number of spermatozoa need to be inseminated because the site of normal semen deposition for artificial mating is the cervix. After insemination, a large number of spermatozoa are trapped in the crypts and folds of cervix and do not have an opportunity to fertilize ova. The two methods by which spermatozoa are lost in the uterus are backflow of the semen and polymorphonuclear leukocyte phagocytosis (12,13). The clearance of spermatozoa from the uterus starts soon after insemination (14). Several different types of catheters have been designed to physically by-pass the cervical crypts and folds of the uterus during insemination. This research project evaluated whether depositing a low number of spermatozoa in the uterine body or deep into the uterine horn would influence reproductive performance.

The average number of total piglets born per litter for sows inseminated by CI with  $3.0 \times 10^9$  sperm cells per dose was 1.40 piglets per litter greater ( $p < 0.01$ ) compared with sows inseminated by DIHU with  $0.15 \times 10^9$  sperm cells per dose. A similar study (11)

found a small increase (0.27 piglets) in total number of piglets born per litter for sows inseminated by CI with  $3 \times 10^9$  sperm cells per dose compared with sows inseminated by DUHI with  $0.15 \times 10^9$  sperm cells per dose.

Although a significant difference was not found in total number of piglets born per litter when comparing CI versus TCUB insemination methods, sows and gilts inseminated with  $3 \times 10^9$  sperm cells per dose produced 1.2 more piglets per litter than sows and gilts inseminated with  $1.5 \times 10^9$  sperm cells per dose. The standard error of the mean for sows inseminated by TCUB was about twice the value for the CI treatment. Some authors (15) found a significant decrease in total pigs born (1.6 piglets) and total pigs born alive (1.5 piglets) when sows were inseminated by TCUB with  $1.0 \times 10^9$  sperm cells per dose compared with sows inseminated by CI with  $4 \times 10^9$  sperm cells per dose. The results from these studies indicate that litter size cannot be maintained when a sub-optimal number of sperm cells per dose are deposited into the uterine body.

The results from the present study confirm the findings from others authors that farrowing rate is not affected when sows are inseminated by CI with  $3 \times 10^9$  sperm per dose compared with sows inseminated by DUHI with  $0.15 \times 10^9$  sperm per dose (11). However, as the number of sperm cells per dose decreases below  $2.5 \times 10^7$  per insemination by DUHI method a decrease in pregnancy rate and farrowing rate occurs as

compared with sows inseminated by CI and  $3 \times 10^9$  sperm cells per dose (11). Although not different, farrowing rate was 8.4 percentage points lower for sows inseminated by TCUB with  $1.5 \times 10^9$  sperm per dose compared with sows inseminated by CI with  $3.0 \times 10^9$  sperm per dose. Rozeboom et al. (2004) did not find a significant difference in farrowing rate when sows were inseminated by TCUB ( $1.0 \times 10^9$  sperm per dose) or CI ( $4.0 \times 10^9$  sperm per dose).

In the present study the WEI category did not significantly influence any of the reproductive traits recorded. Some authors (16) reported that sows with weaning-to-conception intervals of 2 to 4 days had the highest litter sizes, while litter size decreased progressively for sows with weaning-to-conception intervals of 5 to 7 days. Sows with weaning-to-conception intervals of 7 to 10 days had the smallest litters. The small number of sows per treatment in the current study may explain why the WEI did not influence the number of piglets born.

The significant difference in average birth weight per piglet between treatments is related to the number of piglets born per litter. More total piglets were born for sows and gilts inseminated with  $3.0 \times 10^9$  sperm per dose compared with sows inseminated with

$1.5$  or  $0.15 \times 10^9$  sperm cells per dose. A negative relationship between litter size and weight of piglets at birth does exist (17).

## CONCLUSIONS

Farrowing rate is not significantly reduced when sows and gilts are artificially inseminated by transcervical insemination ( $1.5 \times 10^9$  sperm cells per dose) or inseminated by deep intrauterine horn method ( $0.15 \times 10^9$  sperm cells per dose) compared with a traditional cervical insemination ( $3.0 \times 10^9$  sperm cells per dose).

Total number of piglets born per litter is significantly reduced when sows are inseminated by DUHI with  $0.15 \times 10^9$  sperm cells per dose compared with sows inseminated by CI ( $3.0 \times 10^9$  sperm cells per dose) on a pig farm.

## ABBREVIATIONS

- AI -artificial insemination
- DUHI - deep intrauterine horn insemination
- TCUB - transcervical-uterine body insemination
- CI - cervical insemination
- WEI - weaning-to-estrus interval

**Table 2.** Effect of sperm number per dose and weaning-to-oestrus interval (WEI) on reproductive performance of sows artificially inseminated by deep intrauterine insemination, transcervical insemination or cervical insemination (Mean  $\pm$  SE)

Parameters	Deep intrauterine insemination ( $0.15 \times 10^9$ sperm per dose)		Transcervical insemination ( $1.5 \times 10^9$ sperm per dose)		Cervical insemination ( $3.0 \times 10^9$ sperm per dose)	
	WEI		WEI		WEI	
	$\leq 6$ days	$> 6$ days	$\leq 6$ days	$> 6$ days	$\leq 6$ days	$> 6$ days
Number of sows	26	22	22	10	29	23
Farrowing rate, % <sup>a</sup>	96.20	81.80	86.40	77.80	89.71	91.30
Average total number of piglets born per litter	8.83 $\pm$ 0.58	8.94 $\pm$ 0.59	9.00 $\pm$ 0.80	9.14 $\pm$ 1.58	10.15 $\pm$ 0.58	10.43 $\pm$ 0.51
Average number of piglets born alive per litter	8.75 $\pm$ 0.57	8.61 $\pm$ 0.58	8.53 $\pm$ 0.77	9.14 $\pm$ 1.58	9.65 $\pm$ 0.55	10.00 $\pm$ 0.48
Fecundity index <sup>b</sup>	841	704	737	711	865	913
Average total birth weight of litter, kg	13.55 $\pm$ 0.84	12.74 $\pm$ 0.73	14.03 $\pm$ 1.10	14.54 $\pm$ 2.09	14.20 $\pm$ 0.88	13.52 $\pm$ 0.72
Average birth weight per piglet, kg	1.58 $\pm$ 0.05 <sup>A</sup>	1.49 $\pm$ 0.06	1.61 $\pm$ 0.06 <sup>B</sup>	1.67 $\pm$ 0.08 <sup>CD</sup>	1.48 $\pm$ 0.05 <sup>C</sup>	1.37 $\pm$ 0.04 <sup>ABD</sup>

<sup>a</sup> Farrowing rate is the number of sows that farrowed divided by the number of sows bred

<sup>b</sup> Fecundity index is farrowing rate times average number of piglets born alive per litter times 100

<sup>ABCD</sup> Same superscripts within row indicate differences between means ( $p < 0.01$ )

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