Effect on fertility of sperm motility in pooled Duroc semen

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Semen doses with pooled semen from three Duroc boars, all with good sperm motility, had the same fertilizing capacity as doses with pooled semen from three boars with poor sperm motility and from three boars with good sperm motility.

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Abstract

Sperm with reduced motility has reduced fertilizing capacity. If low-motile Duroc sperm is mixed with high-motile sperm, the fertilizing capacity of that dose is equal to semen doses exclusively containing high-motile Duroc sperm. With doses that only contained low-motile sperm, the number of total born piglets per litter dropped by minimum 0.5.

Duroc boars were selected on the basis of the percentage of motile sperm assessed with SpermVision CASA system version 3.0, and divided into three groups:

Group 1: Semen with high motility
Group 2: Semen with reduced motility
Group 3: Equal share of semen from groups 1 and 2

4,528 LY sows from three Danish sow farms were randomly allocated to the three groups and inseminated with semen from the selected boars.

Farrowing rates were identical in all three groups, but the number of total born piglets differed significantly between the groups (P=0.0001). The number of total born piglets was identical in groups 1 and 3, whereas in group 2 it was minimum 0.5 lower than in groups 1 and 3.

Danish commercial semen consists of semen from up to ten different Duroc boars. The outcome of this trial demonstrates that this procedure guarantees a good fertility of the semen dose in relation to sperm motility.

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Background

In Danish pig production, finishers are most commonly produced with semen doses that consist of semen from several different boars. Pooling semen is considered a guarantee that fertility is not adversely affected by one boar with reduced fertility. However, the exact effect of pooled semen is unknown.

The effect of pooling semen of good quality with defect sperm often depends on the defect. Defects can be classified as:

1. Defects that result in non-fertile sperm that does not affect other sperm
2. Defects that result in sperm fertilizing an egg, but the egg dies
3. Defects that result in non-fertile sperm that also affects other sperm negatively.

Curled tails is one example of a defect that does not affect other sperm. Sperm with curled tail is intact, but does not swim efficiently and will therefore never fertilize an egg.

However, sperm with genetic defects is capable of fertilizing an egg, but the egg will die. The consequence is likely a drop in litter size because of the actual competition between functional and defect sperm.

The process from sperm is damaged and finally dies appears to be gradual. The sperm is likely to start by losing its ability to fertilize an egg; then motility drops and finally it becomes completely immotile. Today, sperm motility is used as a parameter of whether the sperm has a sufficient shelf-life – if it looks alive after storage. When cells in the body die, they release substances that are harmful to other cells [1]. This is a familiar process for cells in the body, and this process may be assumed to apply also when sperm dies.

Pig Research Centre tested the Computer Assisted Sperm Analysis (CASA), which is an instrument for assessing sperm motility [2]. In an investigation with Duroc semen, CASA revealed large differences in sperm motility between different boars [3]. Consequently, differences in sperm fertility are expected. Results also revealed a significant correlation between sperm motility parameters assessed with CASA on Large White semen and sperm fertility [4]. It is not known whether sperm with reduced motility affects the quality of the sperm when semen is mixed.

The aim of this present investigation was to analyse whether fertility of mixed semen is adversely affected when a dose contains a mix of high-motile sperm and low-motile sperm.

Materials and methods

Semen from Duroc boars from one boar station was analysed to select boars for each group (see Table 1). Boars with high-motile sperm were allocated to group 1; boars with low-motile sperm were allocated to group 2; and semen mixed of the two constituted group 3. Sperm from all three groups was used on three Danish sow farms.

Table 1. Trial design (the table shows the number of boars represented in each group).

Group	1	2	3
Sperm motility	High	Low	Equal mix of 1 and 2
Number of boars represented in the doses	≥ 3	≥ 3	≥ 6

Selection of boars for the trial

Semen from one ejaculate from each Duroc boar was analysed three days after collection. A semen dose was randomly collected from each boar and stored at 17 °C until analysis. Analyses included assessment of sperm motility with CASA.

Analysis of sperm motility

On day 3 after collection, sperm motility was assessed. 10 ml semen was transferred to a 10 ml polycarbonate tube (NUNC, Denmark). The tubes were turned five times to mix the semen before being placed in a water bath for 50 minutes to reactivate the sperm. Upon reactivation, the semen was mixed by turning the tube ten times. From this tube, 2.4 µl semen was collected and placed in a 20 µl deep Leja 4 chamber (Leja, the Netherlands). The sperm were analysed in the part of the chamber with the least damaging effect on sperm motility. This position is the area closest to the sample inlet [5]. The analyses were performed with SpermVision CASA version 3.0, and included assessment of sperm motility in 15 different areas. As variables, the analysis included the percentage of motile sperm (MOT) and variations in the swimming speed of motile sperm (R-VCL), as these variables have proven

a correlation with sperm fertility [3]. All analyses were made as two separate consecutive measurements.

Production of semen doses

The semen doses were produced in accordance with “Guidelines for AI stations – semen preservation and health control” [5] and only semen from approveed boars was used. Semen that did not meet normal quality standards was discarded. Semen doses were produced weekly (Sundays) from minimum six different boars representing minimum three boars with good sperm motility and minimum three with reduced sperm motility (see Table 1). Group 3 contained an equal amount of semen from groups 1 and 2. Each dose of approx. 80 ml contained approx. 2.2 billion sperm. One dose from each group was used in the quality control. The dose was stored at 17°C until analysis. Semen doses used on commercial farms were shipped to the farms according to normal AI station routines.

Quality control of semen used in the trial

From each group and on each day of production, one semen dose was selected for quality control and analysed to record the number of sperm per dose and sperm motility. Semen was weighed, and for recording of sperm concentration 1.00 ml semen was diluted with 10.00 ml Reagent S100 (Chemometec, Allerød, Denmark). A sample of the diluted semen was collected with NucleoCassette SP1 (Chemometic) and analysed in NucleoCounter SP100 (Chemometic). All analyses were made as two separate consecutive measurements.

On-farm insemination

The trial included two phases in order to prevent losses in case of very large differences between the groups. Phase 1 ran on two sow farms in the period June 2008 – February 2009, and phase 2 on three sow farms in the period July 2009 – August 2011. All participating farms are briefly described in Appendix 1.

The trial comprised 4,528 LY sows randomly allocated to three groups. Only sows inseminated for the first time on day 4 or 5 post-weaning were included in the trial as the semen in this trial was produced 3 days before the first insemination and had to be used within 3-4 days after production. The sows were inseminated with semen from the same group within the same round of heat. Returners and gilts were not included.

Procedure for insemination

Oestrus detection started from day 4 post-weaning by stimulating the sows according to the five point plan. Sows in standing heat on day 4 post-weaning were inseminated the first time on day 5 post-weaning. A sow in standing heat was inseminated twice with a 24-hour-interval. For each sow, one of the following was recorded: farrowing, returner, culled empty.

Statistical analyses

Selection of boars

Boars were selected on the basis of sperm motility and ranked according to the percentage of motile sperm (MOT). Boars among the 25% highest in MOT were allocated to group 1, and boars with the lowest MOT were allocated to group 2.

Sperm motility and sperm concentration in the doses used on the five farms were analysed descriptively.

Differences in litter size between group 1, 2 and 3 were subjected to an analysis of variance. The model included group, phase, sow and parity as explaining variables and farm as random effect. Differences in farrowing rates between groups 1, 2 and 3 were analysed with logistic regression. The model included group, phase, sow and parity as explaining variables and farm as random effect. The models are shown in Table 2.

Results and discussion

Fertility results are shown in Table 2.

Table 2. Fertility results.

Group	1	2	3
	High-motile	Low-motile	Sperm mixed from groups 1 and 2
Sows inseminated	1,245	1,251	2,032
Average parity	3.92	3.86	3.80
Litters	1,154	1,144	1,896
Farrowing rate, ± SEM, %	92.7 ± 0.75	91.3 ± 0.75	93.1 ± 0.60
Litter size, total born, ±	17.4^{a} ± 0.10	16.8^{b} ± 0.10	17.3^{a} ± 0.08

There were no significant differences in farrowing rate between the groups. There was a significant difference in the number of total born piglets between the groups (P=0.0001). In groups 1 and 3, the number of total born piglets was identical. However, in group 2 the number of total born piglets was minimum 0.5 lower per litter than in groups 1 and 3.

The number of sperm and average motility in the semen doses are shown in Table 3.

Table 3. Number of sperm per dose and motility.

Group	1	2	3
	High-motile	Low-motile	Sperm mixed from groups 1 and 2
Concentration of sperm, ± SEM, billion sperm per dose	2.06^{a} ± 0.02	1.99^{b} ± 0.02	2.05^{a} ± 0.02
Percentage of motile sperm (MOT), ± SEM, %	72.61^{a} ± 1.00	49.26^{c} ± 1.00	62.74^{b} ± 0.99

The concentration of sperm differed significantly between the groups ($P<0.020$) with group 2 containing fewer sperm per dose than groups 1 and 3. The reason for this difference was not clarified, but a difference of approx. 0.06-0.07 billion sperm will not result in the reduction in total born piglets seen in group 2. Previous trial results demonstrated that it requires a significantly greater reduction in concentration to detect a reduction in the number of total born piglets in a litter. A reduction from 2.0 billion sperm to 1.0 billion sperm per dose reduced litter size by 0.2 piglets [6].

The percentage of motile sperm differed significantly between the groups ($P<0.0001$): the percentage of motile sperm was highest in group 1 and lowest in group 2, with group 3 in between.

The percentage of motile sperm is correlated with fertility. However, this effect was neutralized when semen with high motility was mixed with low-motility semen. Semen consists of seminal plasma and sperm. Seminal plasma contains proteins believed to affect sperm positively, but the effect varies between boars [7]. Different boars may have different concentrations of these proteins. It is therefore possible that sperm and plasma from a boar with high concentrations of these proteins may improve the fertilizing capacity of sperm from other boars. This may explain the results in this trial, but it cannot be determined for certain. It is also possible that the morphology of the sperm affected the result.

Conclusion

When sows are inseminated with high-motility Duroc semen with equal amounts of low-motility Duroc semen, the farrowing rate and the number of total born piglets are the same as if the semen dose only contained semen with high motility. If the semen dose exclusively contained semen with low motility, the number total born piglets dropped by 0.6.

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Appendix 1

Farm data

Farm	1 (109588)	2 (55572)	3 (13279)
Sows/year	1,100	1,120	580
Service facility	Crates with boar alley	Groups, 1 feeding stall per sow; tethered first 4 wks + boar/feed alley	Crates
Batch size	60	60	39
Weekly batch	1	1	2
Use of boar	Two boars	Boar	No boar
Feed	Liquid feed mixed on-farm	Liquid feed mixed on-farm	Liquid feed mixed on-farm
Gestation facility	Group-housed ESF	Group-housed, one feeding stall per sow	Group-housed with T pens