Effect of Extender, Cooling Method and Incubation Time on Storage of Extended Boar Semen at 5°C

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Summary

A study was carried out to determine if cooling method, incubation time and extender affected the success of storing boar semen at 5°C. Extended boar semen can be stored at 5°C with acceptable values of sperm progressive motility, total motility and viability over time. This success depends upon incubating the sperm at 17°C for at least 24 hours prior to 5°C storage.

Extended boar semen can be stored at 5°C with acceptable results.

Introduction

Extended boar semen is usually stored at a temperature of 17-18°C, but this temperature is difficult to maintain, especially during transport. Fluctuations in temperature during shipping or storage that affect the quality of the extended semen can often go unnoticed. Storing extended boar semen at 5°C would allow for the transport of semen using readily available cooling units.

A study to determine the effects of cooling method and incubation time on boar semen extended in three different extenders and stored at 5°C was carried out at PSC Elstow Research Farm Inc. The objectives of this project were to determine the effect of 1. three commercial extenders, and 2. stepwise cooling combined with different final incubation times at 17°C, on the ability to store extended boar semen at 5°C.

Experimental Procedures

Eighteen fresh ejaculates from 7 boars were split and 8 insemination doses (2 billion sperm, 70 mL each) were extended in each of three commercial extenders (Ext A, B and C) at 35°C. Within extender, each dose was subjected to one of eight cooling rate by incubation time treatment combinations:

Treatment	Cooling Method	Incubation	Final Storage Temperature
1	Stepwise 35 - 17°C	None	17°C
2	Stepwise 35 - 17°C	None	5°C
3	Stepwise 35 - 17°C	4 Hr @ 17°C	5°C
4	Stepwise 35 - 17°C	24 Hr @ 17°C	5°C
5	Direct 35 - 17°C	None	17°C
6	Direct 35 - 17°C	None	5°C
7	Direct 35 - 17°C	4 Hr @ 17°C	5°C
8	Direct 35 - 17°C	24 Hr @ 17°C	5°C

Stepwise cooling consisted of placing the 35°C extended semen into consecutive water baths at 32, 29, 25, 22, 19 and 17°C, changing the extended semen from one bath to the next every 30 min. For direct cooling, the semen doses were placed into a 17°C storage cabinet. Total motility (percentage of sperm moving), progressive motility (percentage of sperm moving in a forward fashion) and viability (percentage of living sperm), were measured on the extended semen samples for six consecutive days.

Results and Discussion

Extender has an effect on sperm motility. Progressive motilities were greater in extenders A and B than in C on day 1, but from day 2 on, progressive motilities were greater in extender A, intermediate in B and lower in C (Figure 1).

Progressive motilities in the 8 cooling rate/ incubation time treatments were typically the greatest in sperm stored at 17°C (Figure 2; Treatments 1 and 5) followed by sperm incubated at 17°C for 24 hr prior to storage at 5°C (Treatments 4 and 8). Cooling method (direct or stepwise) did not affect these results.

Total motility was lower in extender A and B than in C for days 1 through 5 and lower in extender A than B and C on day 6.

Total motility on day 1 was greater when sperm were stored at 17°C or incubated for 24 h at 17°C prior to storage at 5°C (Figure 3; Treatments 1, 5, 4 and 8), than when sperm were stored at 5°C or incubated for 4 h at 17°C prior to storage at 5°C (Treatments 2, 6, 3 and 7).

By day 2, total motilities were greatest for sperm stored at 17°C (Treatments 1 and 5), followed by sperm incubated for 24 hr at 17°C prior to storage at 5°C (Treatments 4 and 8), which in turn were followed by sperm incubated for 4 h at 17°C prior to storage at 5°C (Treatments 3 and 7). Sperm stored at 5°C (Treatments 2 and 6) had the lowest values for total motility and this pattern continued on through to day 6.

Extender A, regardless of cooling rate/ incubation time combination, maintained both progressive and total motility values at the greatest levels over time. When stored at 5°C without prior incubation at 17°C, total motility values were generally greater for sperm that had been cooled stepwise compared to those cooled directly. While total motility values were superior when sperm were stored at 17°C, acceptable total motility was possible when sperm were stored at 5°C but depended on a 24h incubation period at 17°C.

Viability results differed among the three extenders. In Extender A, viability on day 1 was greater for all cooling methods compared to sperm directly cooled and stored at 5°C (Treatment 6). By day 2 there were essentially no differences.

Viability in Extenders B and C was variable, with sperm stepwise or directly cooled and stored at 5°C without incubation (Treatments 2 and 6) usually yielding values among the lowest. Cooling method (direct or stepwise) did not affect viability in any of the three extenders.

Implications

These results indicate that extended boar semen can be stored at 5°C with acceptable values of sperm progressive motility, total motility and viability over time. Achieving these acceptable values at a storage temperature of 5°C depends upon incubating the sperm at 17°C for at least 24 hours prior to 5°C storage. Extender choice influences this success, as does incubation time, but only total motility was affected by a direct or stepwise cooling method. It is important to note that these are laboratory results and an insemination trial is required to confirm these laboratory findings prior to implementation in the field.

Acknowledgements

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Figure 1: Progressive Sperm Motility

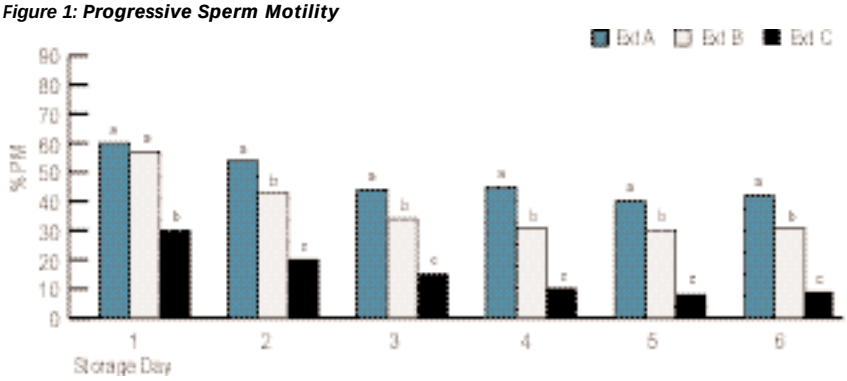

Figure 2: Progressive Sperm Motility

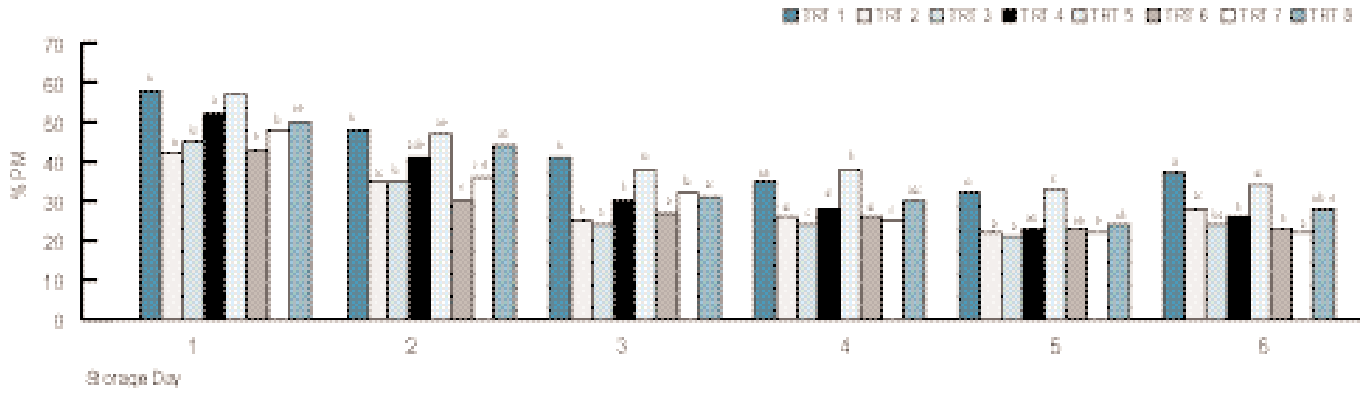

Figure 3: Total Sperm Motilty