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## FLUCTUATING-TEMPERATURE STORAGE OF BOAR SEMEN: SPERM-IA

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### Introduction

Plasma membrane of boar spermatozoa is highly sensitive to any temperature changing (Maxwell and Johnson 1997). Most extenders are formulated to storage boar semen at 15°C. Inappropriate changes in temperature during transport and storage can induce functional and biochemical damage to the sperm. The aim of this research was to design an extender to preserve semen quality at fluctuating temperatures and 5°C.

### Material and Methods

Previously, every component of the extender (sugar, buffers, antioxidants, antibiotics and cryoprotectors) was properly selected by several trials. Five ejaculates were fractionated and then diluted into Sperm-IA and stored at two different temperature conditions: fluctuating temperature from 5°C to 20°C (T1) and 5°C (T2) for 3 and 8 days. To evaluate sperm quality, next parameters were measured: total motility and progressive motility (ISAS®; Proiser; Spain), hypoosmotic swelling test (HOST) and thermal resistance (ORT), mitochondria status (Rhodamine) and viability and acrosome integrity (FITC-PNA-CFDA-PI). *In vitro* fertilization capacity was evaluated after 3 and 6 days for T1. Three extenders were included in this study as controls.

### Results

The use of SPERM-IA for semen storage at fluctuating temperatures showed higher response to HOST (0.0393) as well as better mitochondria status (<0.0383) at 3 days comparing to controls. There were differences in total motility (0.0142), progressive motility (0.0006) and HOST (0.0307) in favour of our experimental extender after 8 days in T1. Higher percentages of alive and intact acrosome status were found in Sperm-IA (0.0001). Regarding fertilization, upper penetration rates were shown by novel extender (Figure 1).

Total motility, progressive motility and HOST percentages were significantly higher for Sperm-IA after 8 days (Table 1).

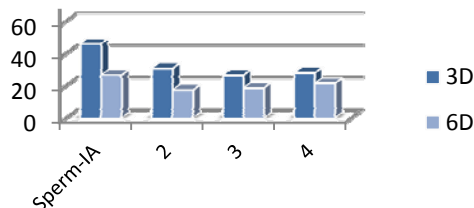


Figure 1. *In vitro* penetration capacity after 3 and 6 days in fluctuating temperatures.

TM	PM	HOST
93,60 ± 2,89 a	51,60 ± 9,23 a	37,20 ± 4,84 a
78,80 ± 2,83 ab	38,80 ± 4,78 ab	22,60 ± 5,79 ab
52,00 ± 10,81 b	20,00 ± 7,50 b	15,00 ± 3,57 b
70,80 ± 6,32 ab	33,40 ± 10,01 ab	14,00 ± 2,19 b
<b>0.0039</b>	<b>0.0174</b>	<b>0.0055</b>

Table 1. Semen quality after 8 days conservation. At 5°C in terms of total motility (TM), progressive motility (PM), test hypoosmotic (HOST) and resistance osmotic (ORT).

### Conclusions

Sperm-IA, novel extender of Magapor, protects semen from fluctuations in temperatures during storage and maintains quality at constant low temperatures. Previously, other extenders have been designed to maintain semen quality at constant temperatures, at 5°C (Correa et al. 2006) and at 10°C (Le Thi et al. 2011) but not fluctuating or at fluctuating one but not 5°C (Wabersky et al. 2010).

### References

1. Correa MN et al.: 2006, Anim Reprod 3:41-43.
2. Le Thi X et al.: 2011, Unit for Repr. Med. Hannover
3. Maxwell WM and Johnson LA: 1997, Mol Reprod Dev 46(3): 408-418