

Introduction



Responsible artificial insemination (AI) organizations strive to ensure that semen sold has the potential to achieve acceptable levels of fertility when used in herds of fertile cows and heifers. Accordingly, straw handling by the inseminator should be limited, in order to reduce the damages to the semen quality.

Materials and Methods



This study tested different conditions of handling before AI:

Time condition

straws are kept out of the liquid nitrogen during

- 0 sec
- 5 sec
- 10 sec
- 15 sec

Frequency condition

straws are kept out of the liquid nitrogen 0-3-6-9 times during 5 sec

Liquid nitrogen level

Level 1



Level 2



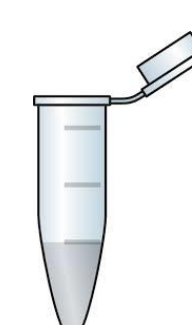
straws are kept in vapour nitrogen (instead of liquid) during 24-48h before previous described handling conditions



Straws provided from 5 Belgian Blue bulls

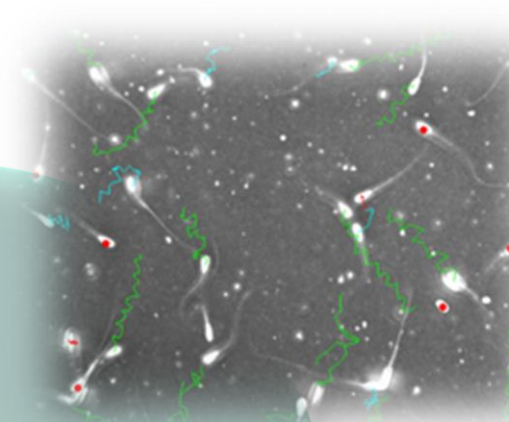
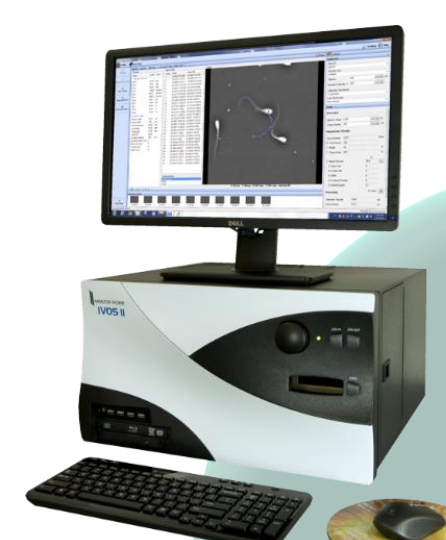


Thawing, 30 sec at 37°C

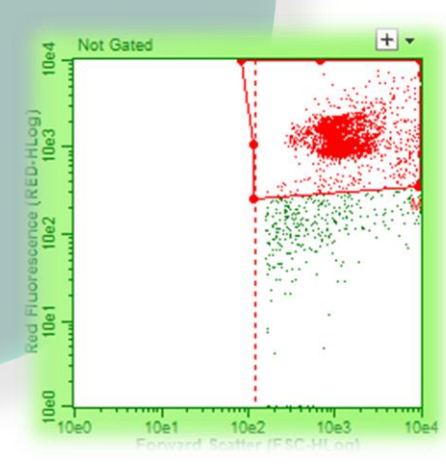


Data were analysed by analysis of variance (SAS software)

Microscope (CASA system – IMV tech.)



Flow cytometry (easycyte – Millipore R)



Viability
Acrosomal integrity
Mitopotential

Results and Discussion

Semen quality was not significantly affected by straw handling (Time and Frequency conditions) under liquid nitrogen storage.



Parameters	Time effect			Frequency effect		
	5 secs	10 secs	15 secs	3 x 5 secs	6 x 5 secs	9 x 5 secs
% motility	No	No	No	No	No	No
% progressive	No	No	No	No	No	No
% viability	No	No	No	No	No	No
% alive with stables phospholipids	No	No	No	No	No	No
% spz with intact acrosome	No	No	No	No	No	No
% spz alive with intact acrosome	No	No	No	No	No	No
% spz with polarized mitochondria	No	No	No	No	No	No



Level 2

After 24h in vapour nitrogen storage condition, the acrosomal integrity and the mitopotential level significantly decreased even without additional straw handling.

Keeping the straws during 48h in vapour nitrogen with additional handling (the straws were kept out of the tank 9 times during 5sec) decreased the semen quality for all the measured parameters.

Parameters	Liquid nitrogen level effect		Liquid nitrogen level + manipulation effect
	Low level 24h	Low level 48h	Low level 48h 9 x 5 secs
% motility	No	No	Yes
% progressive	No	No	Yes
% viability	No	Yes	Yes
% alive with stables phospholipids	No	Yes	Yes
% spz with intact acrosome	Yes	Yes	Yes
% spz alive with intact acrosome	No	No	Yes
% spz with polarized mitochondria	Yes	Yes	Yes

The results of this study suggest that caution should be taken before AI by the users, especially if the straws are maintained under atmospheric vapour of nitrogen instead of liquid nitrogen immersion, to preserve the fertility potential of the bulls.

Conclusions