

Development of a semen freezing extender in cheetahs

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EnvA

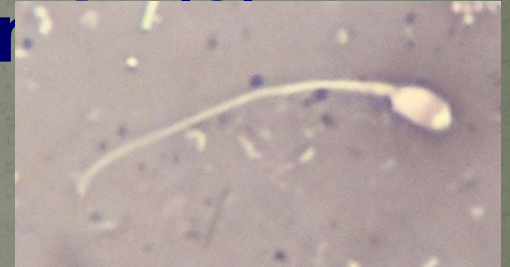
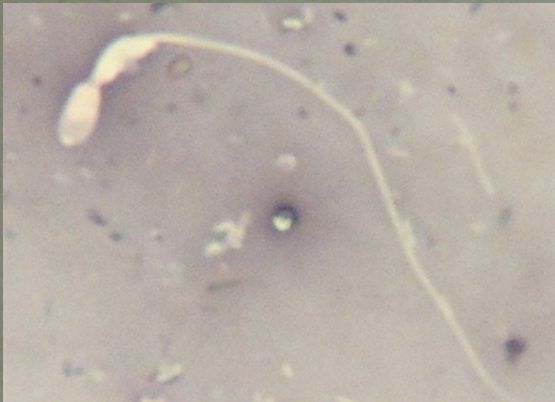
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INTRODUCTION

- Damage caused to cells spermatics by cold shock is seen as an irreversible loss of motility after cryopreservation and thawing and includes lesions in the membranes and changes in metabolic function (1).



- Semen freezing is difficult in felids and especially in Cheetahs because of the high incidence of teratospermia



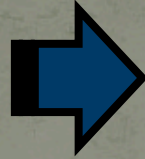
- Particularly if the semen is frozen to be used for artificial insemination.



- The aim of this research was to develop a semen freezing protocol for Cheetahs in one of the wildlife reserves in South Africa.



MATERIALS AND METHODS



anesthetic protocol:

0.04 mg/kg medetomidine plus

2.5 mg/kg ketamine and after collection

cheetahs 0.5mL IM and 0.5 mL IV of atipemazole
()

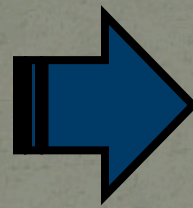
80 stimuli (30, 30 and
20) from 2 to 3V.

Semen
collected by
electroejaculation



During semen collection

**Diluted 1:1 with Extender TRIS/
citric acid/glucose/without egg
yolk**



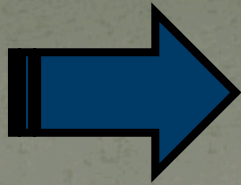
Evaluations of samples:

Spermatic progressive
motility, progressive velocity,
ejaculate concentration,
integrity of membranes,
spermatic abnormalities

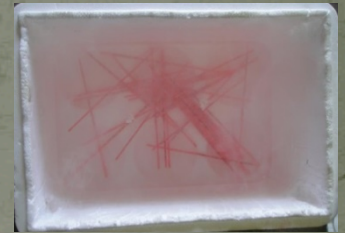
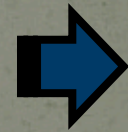
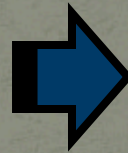
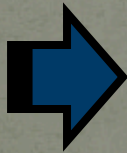
pH



Centrifuged
at 300g/10
min



The pellets were diluted
in Tris / Citric Acid /
glucose / 20% egg yolk /
1% Orvus ES Paste / 4%
glycerol, pH 8,0.

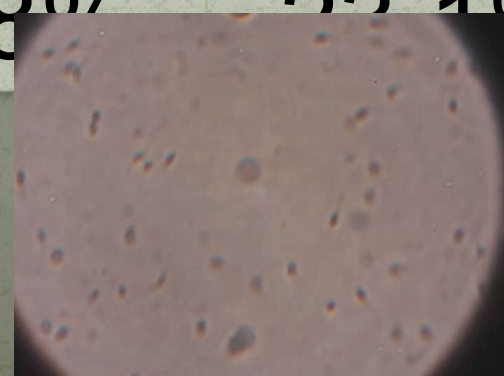
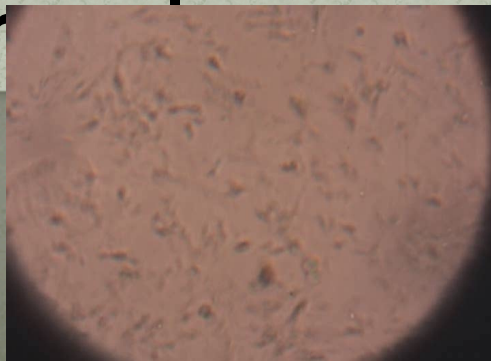


They were packed in 0.25 mL straws,
equilibrated at 4°C/3h and frozen in
nitrogen vapor for 10 min; thus stored at
-196°C. The straws were thawed at 37°C

RESULTS

Median values of semen characteristics were:

Evaluation	Fresh	Thawed
Motility	78%	50%
Sperm velocity	4.0	2.9
Normal morphology	28.8%	22.1%



RESULTS

The most frequent spermatic alterations were:

Morphology	Fresh	Thawed
Acrosome defects	19.6%	27.6%
Intermediary piece	19%	25.5%
Tail defects	25%	24.5%
Head defects	2%	4%

We observed an important decrease in the spermatozoa membrane integrity after freezing (23% versus 11%).

CONCLUSION



The main difficulty in freezing semen of cheetahs had been that all male had less than 30% of normal spermatozoa and therefore were more sensitive to cryopreservation.

In conclusion, despite strong teratospermia detected, it is possible to freeze semen from Cheetah.

REFERENCE

(1) PUKAZHENTHI, B.S., PELICAN, K., WILDT, D.E., HOWARD, J.G. Sensitivity of domestic cat (*Felis catus*) sperm from normospermic versus teratospermic donors to cold-induced acrossomal damage. **Biol. Reprod.**, v. 61, p.135-41, 1999.

(2) PUKAZHENTHI, B.S., WILDT, D.E., HOWARD, J.G. The phenomenon and significance of teratospermia in felids. **J. Reprod. Fert. Suppl.**, v.57, p.423-33, 2001.

(3) MARTINS et al. Paper in preparation.

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