

PROGRESS IN ALPACA RESEARCH AT THE UNIVERSITY OF SYDNEY SPRING 2006

Alpaca research at Sydney University has been progressing at a great rate. Our first progress report to RIRDC has been received with praise.

We're pleased to announce that Dr. Jeff Downing has joined the team. Dr. Downing holds a Diploma of Agriculture from Charles Stuart University, BSc and PhD from Macquarie University. He is currently a lecturer in the Faculty of Veterinary Science, University of Sydney. His research interests have concentrated on various aspects of animal reproductive and stress physiology. The interaction between nutrition, ovarian follicle development and ovulation rate in ewes has been central to much of his research interests as has been the manipulation of ovulation rate using steroid immunization and hormone treatments. He has acquired extensive expert skills in reproductive endocrinology; nutrition-reproduction interactions; oocyte maturation and advanced surgical skills becoming proficient in performing ovarian, utero-ovarian and adrenal autotransplants in sheep. Procedures such as embryo collection and transfer and intrauterine AI have been routine operations. He has published 52 refereed papers and 56 conference papers.

The research team comprises Professors Gareth Evans and Chis Maxwell, Drs. Jeff Downing and Katherine Morton, and two honours students (starting in 2006), Ms. Zamira Gibson, and Ms. Sarah Wilson. Unfortunately, Jorge Reyna (PhD student) resigned from the University in December.

Our progress to date includes

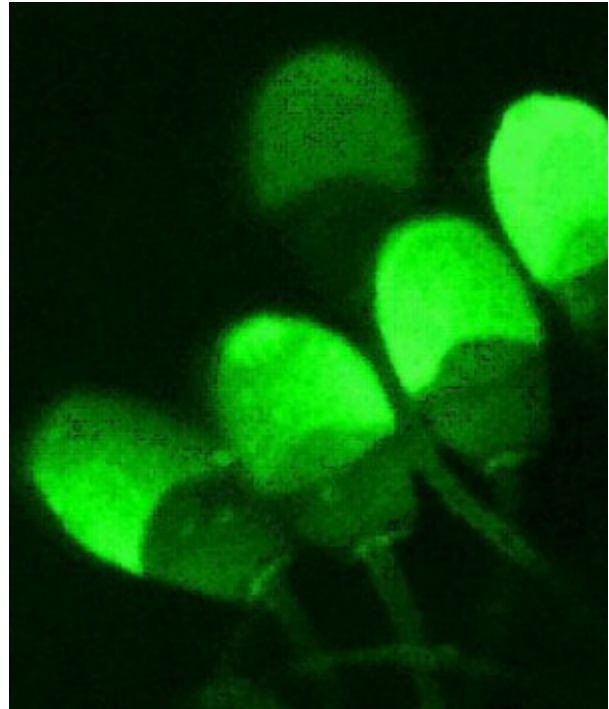


Figure 4. FITC-PNA stained alpaca sperm with intact acrosomes (stained green; upper region of the sperm head).

Previous studies in many species have used older style stains (such as Giemsa) to examine acrosome integrity. However, these are being replaced with fluorescent stains. The project is also developing a fluorescent CTC-stain to examine the capacitation status of sperm (capacitation is a process the sperm must undergo before they undergo the acrosome reaction and fertilise an oocyte). The development of these stains will represent the first use of a fluorescent stain with alpaca sperm, and the first assessment of the capacitation status of camelid sperm.

The computer assisted sperm analyser (CASA) machine has been adapted for alpaca sperm. The CASA measures

establishing a herd of males at Camden. The males are being trained for semen collection and the preliminary results are encouraging. During the on-farm collection and the training period, three sequential ejaculates were obtained from males (n=10) to examine the variability between males and ejaculates. Ejaculate volume (Figure 1), sperm concentration (Figure 2) and sperm motility (Figure 3) were highly variable between males and between ejaculates. Interestingly, some males did not produce any semen despite mating with the mannequin indicating that further improvements to the collection technique were required.

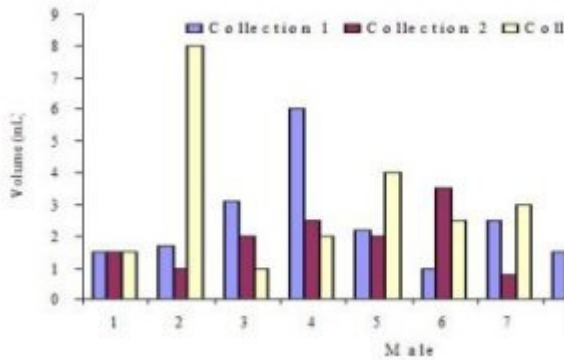
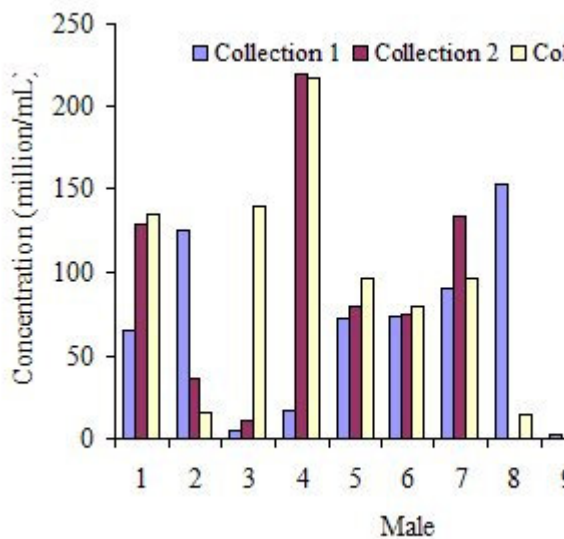


Figure 1. Semen volume in three consecutive ejaculates during repeated semen collection from alpaca males (n=10).



sperm motility (movement) and patterns of sperm movement, permitting objective and precise determination of alpaca sperm motility patterns.

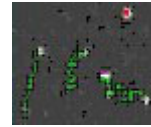


Figure 5. Motility of sperm as described by the CASA. The green lines are the CASA tracing the movements of the sperm. The red dot is an immotile sperm.

Normally sperm motility is assessed subjectively (visual assessment by trained personnel) but because of the variable sperm concentrations and viscosity of the seminal plasma subjective assessment are often unreliable.

Progress with epididymal sperm has been limited owing to the uncertain supply of testes. Camel and alpaca testes were utilised to establish the collection, chilling and shipping protocols. We have developed “kits” for the collection of testes and anyone interested in helping us by donating testes is strongly encouraged to contact us by phone 02 9351 3463 or email kmorton@vetsci.usyd.edu.au.

Sperm were then diluted in three different extenders to determine the appropriate medium for chilling and cryopreservation. Motility of epididymal sperm at room temperature (immediately after harvesting) and after chilling (at 4°C), diluted in lactose, citrate or Salamon’s buffer is presented in Figure 6.

Figure 2. Sperm concentration in three consecutive ejaculates during repeated semen collection from alpaca males.

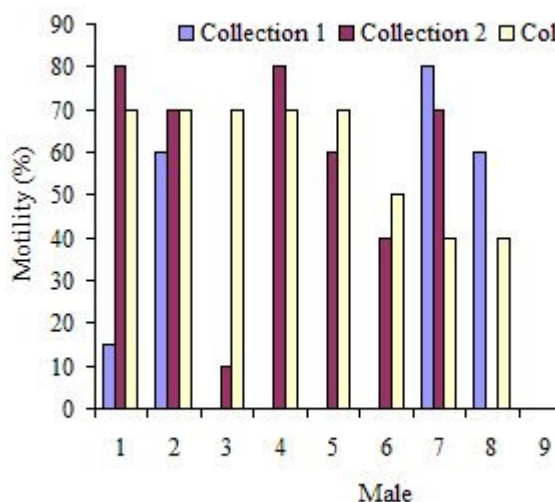


Figure 3. Sperm motility of three consecutive ejaculates during repeated semen collection from alpaca males.

A high variation in the parameters of alpaca ejaculates has been reported previously, and the project is continuing to establish the basic parameters for alpaca ejaculates. The following results are based on 55 ejaculates. Results are presented as mean±s.e.m. (range)

Mating length (mins)	18.14±1.27	range	4-43 mins
Ejaculate volume (mL)	1.867±0.2	range	0.0-8.0 mL
Motility (%)	44.9±4.43	range	0.0-80 %
Concentration (x 10 ⁶ mL ⁻¹)	71.2±9.6	range	0.0-220

Considerable progress has been made in the development of laboratory and in vitro tests for alpaca sperm. Basic laboratory handling procedures, staining protocols and in vitro tests are paramount to all experimental work. We have commenced research to

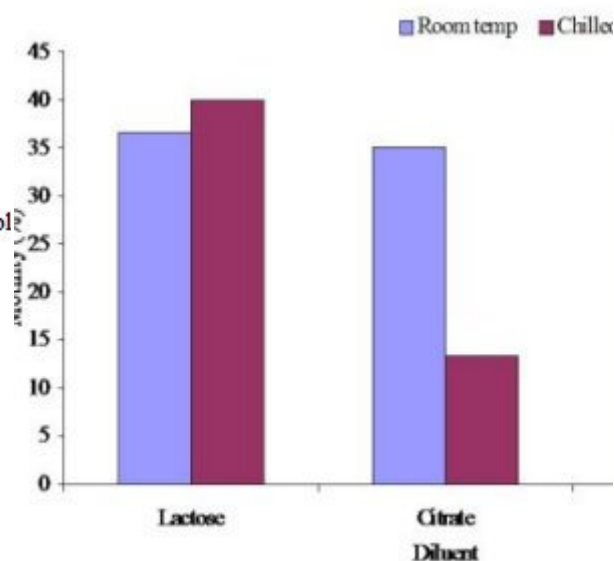


Figure 6. Motility of epididymal sperm at room temperature and after dilution in lactose, citrate and Salomon's buffer.

Presently there are in excess of 120 pellets of frozen epididymal alpaca sperm, which will be used to establish post-thaw in vitro fertility tests such as a zona-binding assay (the binding of frozen-thawed alpaca sperm to cattle oocytes) and to investigate the longevity of frozen-thawed sperm.

Future plans include investigating the effects of some enzymes on semen viscosity and sperm motility, and testing different diluents and temperatures for the liquid-storage of alpaca sperm. If anyone is interested in more information about our research, or would like to participate in research please do not hesitate to contact me.

Dr. Katherine M. Morton
 Post-doctoral Research Fellow
 Centre for Advanced Technology in
 Animal Genetics and Reproduction
 (ReproGen)
 Faculty of Veterinary Science, The
 University of Sydney, 2006.

Phone: 02 9351 3463

determine the appropriate handling medium for alpaca sperm, modified protocols for percoll centrifugation (a process by which live sperm are separated from dead sperm), and fluorescent staining for determining alpaca sperm viability and acrosome integrity (the acrosome is a membrane encasing the head of the sperm that has an integral role in fertilisation).

Fax: 02 9351 3957

Mobile: 0412 187 824

Email: kmorton@vetsci.usyd.edu.au

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