

The Australian alpaca industry is blessed with serious money backing research into new opportunities for their industry. The **Rural Industries Research and Development Corporation** has produced three new fact sheets that sum up the work done that we are reproducing in this issue.

The first fact sheet looks at the quantitative genetic analysis of micron blow out in alpacas, crucial knowledge for farms breeding alpacas for their fibre. The second is about a new breakthrough in the long road to artificial insemination that if it becomes commercial would give alpaca breeders a better chance of accessing the top genetics in the world. Thirdly there is more research on using therapeutic antibodies from alpacas which could be a new market opportunity.

MATTER OF FACTS



FACT SHEET



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Quantitative genetic analysis of micron blowout in alpacas

The issue

Micron blowout is one of the major factors affecting long-term financial return on alpaca fibre. Fibre diameter is one of the most important fibre quality traits in apparel textiles because it is strongly related to softness and comfort of the garment. Some alpacas maintain fine fibre throughout life, while others suffer from significant coarsening of fibre as they age; this trait is known as micron blowout. Micron blowout results in reduced productivity, through reduced yield of high quality fibre over the life of an animal.



What is micron blowout?

For alpacas, as is the case with many fibre-producing species, the finest fibre is of the greatest value. It has been well established in many fibre-producing species that fibre diameter increases with increasing animal age; a phenomenon known as micron blowout. In addition, micron blowout can also be caused by over-nutrition, or by a combination of both factors.

Although all fibre producing species have the potential to suffer from micron blowout, the fault is particularly problematic for alpacas, because alpacas are kept as fibre producers for longer than sheep. McGregor and Butler (2004) calculated that the average micron blowout for alpacas over seven years is 7.5µm. This variation can lower the value of fibre from an older animal to less than 20% of the value of fibre from a young animal. And, because alpacas are retained as breeding and production animals for much longer than seven years, this can mean that the majority of the animals' productive life is at the lesser economic value. Micron blowout affects males and females, Suri and Huacaya of every colour; it is thus a problem for all alpaca producers.

As a result of micron blowout, animals produce less valuable fleeces over time, reducing the life-time value of fibre from the individual. Maintenance of low fibre diameter is one aspect of fibre biology that is of importance for the alpaca industry to assist in increasing yield and productivity. However, it is not an easy task to measure micron blowout, because of the many factors that affect micron diameter (for example, genetics, age of dam at parturition, nutrition status, health). Therefore, to accurately identify animals that either have little or lots of micron blowout, longitudinal data on a group of animals kept under the same conditions must be used. With accompanying extensive phenotype and pedigree records, the underlying genetic potential of the animals can be revealed.

Alpacas and micron blowout

In order to carry out research on the inheritance of micron blowout in alpacas, researchers worked with Grupo Inca (Peru), who own and manage a large alpaca herd at their farm, Pacamarca S.A. Pacamarca has kept extensive phenotype records on their animals, and has developed and successfully implemented in-house selection measures to generate estimated breeding values on their animals. This data was used to carry out a complex quantitative genetics analysis, to determine whether micron blowout in alpaca is heritable.



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The project results showed that age, sex, colour, birth year, measurement year and measurement month all significantly affect micron blowout. White and fawn animals were less prone to micron blowout than coloured animals, while males showed a higher propensity to blowout than females. This may not have been due to any effect of sex; rather, it was probably due to the effect of different management between the groups.

Micron blowout was found to have moderately low heritability in alpacas. The heritability of micron blowout from year one to year two is the highest, but very low in the subsequent age categories. This implies that selection against micron blowout may be successful in reducing the extent of the problem, however more data is required to verify micron blowout at later ages.

Industry Opportunities

The results of this research confirm that micron blowout has a moderate genetic component in alpacas. Therefore, it may be possible for breeders to select animals that do not exhibit the trait (or do not exhibit it as strongly), thereby improving the yield of high quality fibre over the lifetime of an animal. However, progress on this research will not be fast, due to the time taken to get successive fibre diameter measurements to allow the detection of micron blowout.

The recommendation arising from this research is that breeders should directly select for reduced fibre diameter and include "change in year one to year two fibre diameter" as a component in their selection process, to take into account genetic predisposition for micron blowout.



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Advancing Artificial Insemination in Camelids, Particularly the Alpaca



The issue

Cryopreservation of sperm (freezing for later use) is an important technique in many species, as it permits long-term storage for artificial insemination (AI). Despite over a decade of research focus, cryopreservation of camelid sperm still cannot be satisfactorily achieved. This means that the alpaca industry cannot take advantage of the many benefits of AI, such as decreased animal shipping costs, increased revenue from stud fees, and most importantly, rapid genetic gains leading to improvement in traits such as fleece quality and quantity.

Background

Previous research has identified that viscosity of alpaca seminal plasma is the principal reason that semen cannot satisfactorily undergo cryopreservation. Typically, a semen extender that contains cryoprotectants is added before freezing, and it is believed the plasma thickness prevents these cryoprotectants from interacting with, and protecting, the sperm cell. Thus, an essential step to successful cryopreservation is reducing the viscous component of the seminal plasma without impairing sperm function.

The Project

This project aimed to:

- Identify the cause and source of the viscosity of alpaca seminal plasma
- Investigate ways to reduce the viscosity without impairing sperm function
- Determine the effect of viscosity reduction on the success of sperm cryopreservation.

Results

In an important breakthrough, a protein Mucin 5B was revealed as the cause of the high viscosity of alpaca seminal plasma. The source of this protein was identified as the bulbourethral gland.

A literature review, and investigations into the use of numerous enzymes, indicated the protease papain as the most suitable enzyme to degrade Mucin 5B. A reliable and cost-effective protocol was developed for the use of papain to reduce alpaca seminal plasma viscosity.

Through adapting and validating sperm analysis methods used in other species, it was confirmed that this protocol does not affect the viability, acrosome integrity, or DNA integrity of the sperm.



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Recommendations

To maximise the success of camelid sperm cryopreservation:

- Semen should be diluted 1:1 in a suitable semen extender, then treated with 0.1mg/ml papain (final concentration) for 20 minutes at 37°C to completely eliminate viscosity. The effect of papain should then be halted by adding 10µM E-64 (final concentration) at 37°C for 5 minutes
- A final seminal plasma concentration of 10% should be used to maximise sperm function. This can be achieved by diluting the semen prior to freezing, or by removing all seminal plasma and returning 10% post-thawing.

Following these recommendations leads to a significant improvement in post-thaw sperm motility rates, however, unfortunately they still do not reach commercially viable levels.

Where To From Here

To benefit fully from the findings of this project, further research is essential. This should focus on:

- Developing an industry standard semen extender for alpaca sperm cryopreservation
- Investigating the effect of sperm storage (pellets or straws) on sperm function
- Determining optimal freeze/thaw rates
- Improving knowledge of alpaca reproductive physiology and optimal AI protocols.

Implications

These findings provide a platform on which to develop successful cryopreservation and a subsequent AI protocol that can be used commercially within the alpaca industry. This would place the Australian alpaca industry at the forefront of the development of AI in camelids bringing, among previously noted benefits, the reputation of being a world leader in this area.



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- RIRDC publication 12/O16: "Advancing Artificial Insemination in Camelids. Particularly the Alpaca" is available from the RIRDC website www.rirdc.gov.au or by phoning 1300 634 313.

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Therapeutic Antibodies from Alpaca: A New Market Opportunity

The issue

This proof of concept study has explored the use of alpaca for the production of medical grade therapeutic antibody products. Alpaca are members of the camelid family. Camelids produce a unique class of immunoglobulin molecules in their blood stream¹. These immunoglobulins can be harvested and refined for production of specialised therapeutic medical products.

The unique properties include reduced allergenic potential, greater heat stability and greater capacity for inactivating certain enzymes^{2,5}. Australia is an ideal location for sourcing alpaca blood because it is recognised as one of the lowest disease risk countries in the world for producing medical products derived from animals³. There are over 130,000 alpaca currently in Australia. This early stage research has successfully demonstrated that alpaca can be used to produce therapeutic antibodies. These findings could lead to a new high-value niche market for producing alpaca derived medical products in Australia.



What are therapeutic antibodies?

Therapeutic antibodies are molecules found in the bloodstream that can bind to, and inactivate, a wide range of chemical substances. There is a billion dollar global demand for therapeutic antibodies⁴. A diverse range of medical products with well-defined markets already exists. Amongst these include various anti-toxin serums for snakes, spiders, bacterial infections (tetanus, rabies, botulism, anthrax), emerging anti-cancer therapies, and a host of other applications⁴. Currently these products are produced primarily in horses, sheep, rabbits and a small range of other animal species. Camelid antibodies have tremendous potential to improve the quality of products available in this existing product marketplace⁵.

How are therapeutic antibodies made?

The production of antibodies requires stimulating the immune system with the product that antibodies are desired for. This process takes time for the animal's immune system to respond with high levels of a specific antibody. Blood is then collected from the animal and processed to concentrate the specific antibody fraction. The product is then tested for its potency and a range of other physiochemical parameters. Typically, these products are final-packaged in sterile glass vials for human or veterinary medical use.

Alpaca derived anti-toxin antibodies:

This proof of concept project explored the potential for alpaca to make therapeutic antibodies against a range of different snake venom toxins. Snake venoms were chosen because, for an antibody to be effective, it must neutralise the actions of the venom. This approach provides a very robust demonstration of all the steps required to produce the neutralising antibody. (continues overleaf)



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Snake venom is a complex pharmacological substance with many different components. This project has also examined the antibody response of alpaca to different venom components. Alpaca respond to venom immunisation with rapid development of neutralising antibodies. Extremely low doses of venom were used, which did not harm the animals. Blood tests were performed regularly to monitor the animals' general health and these showed no abnormalities. All animal procedures were approved by the Victorian Department of Primary Industries Wildlife and Small Institutions Animal Ethics Committee (Approval numbers 16.11 and 06.16). The alpaca serum was successfully processed with modification to existing protocols used for other species. A very pure and concentrated antibody product was produced from the alpaca serum. When tested for venom neutralising capacity the experimental products were effective in neutralising venom.

Industry opportunities:

This proof-of-concept study has demonstrated that alpaca can be used successfully to make therapeutic camelid-type antibody products. For such an industry to develop in Australia, a commercial partner would be required who would develop the antibody products and support them through the relevant government testing and approvals processes. Australia has potential for such an industry given it has an abundant number of alpaca, at an affordable price with low disease risk³.



Manual restraint of alpaca



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