Lowering Sperm Dose Rates in Frozen Semen for Bovine Artificial Breeding

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By

Colin John Pitt

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Abstract

The New Zealand Dairy industry relies on artificial breeding to produce high genetic merit replacement stock. Proven bull semen is extended and preserved as either ambient temperature product or as frozen doses. High fertility is essential to maintain synchronicity of lactation with the spring surge of grass growth.

To improve efficiency in the utilization of the very best bulls producers try to lower sperm dose rates without compromising fertility. Livestock Improvement Corporation's (LIC) Long Last Liquid™ (LLL) is most commonly used during the peak season and is their most important product. However, shortfalls and specifically targeted matings are met with frozen semen. Lowering sperm dose rates in both liquid and frozen semen maximises the usage of elite sires increasing genetic gain, lowering overheads and garnering premium prices for the semen producer.

A product for improving frozen semen technology was developed whereby a discrete quantity of pretreated semen was placed alongside a beneficial post-thawing redilution medium in a standard semen straw. This emulated a larger-scale process developed to freeze semen in times of low demand and redilution into the LLL form for use as an ambient temperature product. This rediluted product has been proved to enhance semen survival and to allow fertility to be maintained at sperm dose rates lower than the widely accepted standard for frozen semen.

The physical packaging of the semen into the industry standard single dose straw in a configuration that separated incompatible components was novel. This configuration was essential to prevent damage to the sperm during the freezing process from low solubility components in the post-thaw redilution medium. Separation between the semen and diluent within the straw was achieved by the introduction of a gas partition or air bubble between the two liquids.

A large-scale field trial showed that the new product could produce savings of up to 25% of the sperm needed for a semen dose to achieve equivalent fertility. Furthermore, the production cost per dose was lowered in comparison to the standard semen processing system used at LIC.

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Glossary of terms used in this thesis

AB. Artificial Breeding.

AI. Artificial Insemination.

BST. Bovine Somatotrophin hormone used to extend lactation in dairy animals.

BW. Breeding Worth. A measure of the genetic potential of an animal.

Caprogen. The liquid semen diluent used by LIC for its LLL products.**CASA.** Computer Aided Semen Assessment. Using a computer to analyse various morphometric and motility parameters of sperm to provide a picture of the viability of an ejaculate.

CASA Computer Aided Semen Assessment. Systems using computerized image analysis to quantify sperm motility and morphology parameters.

Cold-shock. A term describing the changes that occur when sperm are exposed to low temperature.

Cryogenic. Extremely low temperature.

Cryoprotectant. A substance that protects cells from freeze damage.

NZ. New Zealand.

CIDR-B. Controlled Intravaginal Drug Releasing device. Device to assist in oestrus synchrony in ruminants by emulating reproductive hormone release cycle.

Cryovet. A rival company to IMV that produces straws for frozen semen.

DataMate. Hand-held data logger used by LIC inseminating technicians to record information relevant to matings.

Flow cytometry. A system that analyses individual cell parameters using specific wavelengths of light to induce fluorescence of dyed cellular components.

Fonterra. New Zealand's largest dairy cooperative. Processes and markets over 90% of New Zealand's dairy production.

FPD. Farm Production Division of the NZ Dairy Board. A division of the NZ Dairy Board that was the forerunner of LIC and became part of LIC when it was incorporated.

IMV. Instrumentes du Medecine Veterinaire. A French company that developed the frozen semen technology used through most of the developed world.

Kaptan. Plastic used for cryogenic packaging.

KiwCross™. A breed of dairy cattle developed in New Zealand from Holstein-Friesian and Jersey ancestry.

KNEX. The code name given to the project and the frozen semen product described in this thesis.

LIC. Livestock Improvement Corporation. A large dairy breeding and milk testing company based in Hamilton, New Zealand.

LLL. Long Last Liquid™. Livestock Improvement Corporation's flagship ambient temperature semen product.

LN_2. Liquid Nitrogen. Used as cryogenic refrigerant.

Minitub. Minitub GmbH is the German company supplying PVC straws for LIC's LLL product.

mwco. Molecular Weight Cut-off. Term used to describe the cut-off point above which a dialysis membrane excludes molecules in solution.

NRR. Non-Return Rate. A measure of fertility derived from the ratio of females returning to service, that is, failing to conceive to a service.

NZDB. New Zealand Dairy Board. Former New Zealand government body established to oversee dairy production and marketing.

OSH. Occupational Heath and Safety act that governs workplace safety.

PD. Pregnancy diagnosis. Most commonly performed by rectal palpation and/or ultrasound imagery.

PG. Prostoglandin. Hormone used in oestrus synchrony programs.

Pistolette. Inseminating device to contain the straw for deposition of semen into the female.

PVA. Poly Vinyl Acetate. Water soluble glue that can be powdered in its anhydrous state.

PVC. Poly Vinyl Chloride. Plastic used for many applications including straws for artificial breeding.

RDF. Rediluted Frozen Semen. An artificial breeding product used in NZ consisting of straws or bags of concentrated frozen semen that was thawed and rediluted in the semen production laboratory into a volume of Caprogen ready for use as liquid semen.

ROS. Reactive Oxygen Species. A term to describe the various oxidizing compounds that are formed during metabolism or in cellular breakdown.

SCC. Somatic Cell Count. An indicator of the level of infection in a milk sample. Used as a test for mastitis in lactating dairy animals.

Sempak. An artificial breeding product consisting of a straw of concentrated frozen semen that was thawed and rediluted in the field ready for use as liquid semen.

SPD. Sperm per inseminate Dose

SPS. Sire Proving Scheme. Program to breed and prove the genetic worth of sires.

TMR. Total Mixed Rations. Feed for intensively farmed cattle in feedlot systems.

Tris. Trishydroxymethylamine. An organic buffer used extensively as a component in frozen semen diluent.

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1 Introduction

The New Zealand Dairy industry is one of the most cost efficient producers of milk and processed dairy products in the world. It relies on pasture-based farming systems to produce cheap feed for animals bred to efficiently convert grass to milk products [2], [3; 4]. The seasonal surge in grass growth that occurs in spring is the most cost-efficient feedstock for the lactating cows. Cows have to be at the optimum point in their lactation to fully utilise the available feed. Therefore, parturition and hence, conception must occur to a strict timetable. High fertility is essential if the herd is to lactate at the right point in the calendar [5], [6], [7].

New Zealand farmers formed cooperatives such as Livestock Improvement Corporation (LIC) for provision of farm services such as animal evaluation, product testing and breeding of suitable dairy animals[8]. Semen research at LIC is mainly focussed on lowering sperm dose rates and extending the life of its liquid and frozen semen products. This maximises the usage of elite bulls proven to enhance production traits in their daughters. An over-riding consideration is that fertility must be maintained.

LIC's operation is based in two important products: Long Last Liquid^{TM} semen (LLL), which is used during peak demand and deep frozen semen to supplement LLL or for use during off peak season.

Most frozen semen systems are constrained by high sperm dose rates compared to LLL, but LIC has previously developed a frozen semen system where semen frozen at high concentrations is rediluted into the LLL form for distribution. This product was named Rediluted Deep Freeze (RDF). RDF sperm dose rates are more favourable than other frozen systems. The major limitation of this product is its perishability after thawing and redilution. Its positive attribute of lowering sperm dose rates lead to this project.

There is a basic need therefore, for a single dose product capable of overcoming some of the deficiencies in LIC's existing products.

In this thesis, the product development is described along the principles of classical chemical product development [9].

That is:

- Identification of industry needs.
- Generation of ideas.
- Screening between ideas and selecting the most promising idea.
- Product manufacture.

The work is based on research located at LIC's semen research facility at Newstead, Hamilton, New Zealand and this thesis is limited to cattle artificial breeding. Furthermore, possible solutions are constrained by current practices, especially within the infrastructure developed for storing and distributing frozen semen.