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Recommendations for Purchasing Frozen Canine Semen

Introduction

Incorporation of frozen semen into your breeding program is complex and can be expensive. There are 3 fundamental factors that determine the success of artificial insemination with frozen semen:

1. Breeding management and ovulation timing,
2. Artificial Insemination,
3. Frozen semen quality and quantity

We can control and optimise points one and two, but not point three. Where we put the semen, and when we put the semen in, are things we can control to give you the best chance of a pregnancy. When the semen has been frozen elsewhere it is out of our control and unfortunately the quality and sometimes the quantity of frozen semen allocated per AI dose that we receive is sub optimal and we cannot compensate for these deficiencies.

Poorly frozen semen is the most common cause for failure to achieve pregnancy. Published data in a peer-reviewed reproduction journal shows the results from an analysis of over 1000 bitches inseminated at MVS GlenBred over 7 years. It was found that the most significant factor affecting pregnancy rate was semen quality.

Therefore, to try and maximise your chance of purchasing and importing doses of frozen canine semen containing an adequate number of sperm and ultimately of high quality we have put together some guidelines:

Semen Freezing – the facts!

Canine semen has been frozen commercially in New Zealand for about 40 years. Once frozen, and so long as the semen is kept in well maintained liquid nitrogen tanks designed for long term storage, it will last forever without any significant decline in quality. We have produced a number of litters of puppies from semen frozen by Dr Marion Wilson (the founder of GlenBred) 30 years ago.

Factors that affect the quality and quantity of frozen semen you are considering purchasing:

Freezing Method:

There are several methods employed around the world to freeze canine semen. There is also enormous variation between centres and individuals that freeze canine semen in regards to their skills, experience, techniques and knowledge. It is therefore very important that whoever is collecting the ejaculate from your chosen dog for freezing is experienced and reputable. Be cautious of those who freeze canine semen using proprietary or ‘secret’ extenders and methods, or those that claim superior results are achieved with semen

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they freeze in pellets compared to straws. There is no objective evidence to this claim, and in many species it has been scientifically proven that there is no difference in post thaw motility and fertility between pellets or straws. Furthermore, as with other species, there are also many steps involved in freezing canine semen as well as a variety of different extenders that can be used, but there is neither one single method nor one extender that is significantly superior to others.

Regardless of what extender and what method is used, the main determinants of good post thaw motility, and ultimately fertility, is the skill of the person collecting and freezing the dog, the individual dog and his inherent fertility. There are many factors that affect a dog's fertility. Some of the more important factors include his age at the time of semen freezing, health status (especially in regard to his reproductive organs such as the prostate, testicles, epididymis), how often he is used at stud and importantly his reproductive history (especially whether he is a proven sire - ideally with both natural mating and with his frozen semen).

Semen Assessment:
After an ejaculate is collected, and at each step in the freezing process, including assessment after thawing, a sample of semen should be removed and assessed under the microscope for a number of basic seminal parameters including motility, concentration, morphology, and cytology. A number of operators are also using a "CASA" system which is a computer assisted assessment method of assessing semen, which should reduce the subjectivity of assessing semen. These systems provide print outs of a number of seminal parameters. These findings should be provided in a **semen report** which you can request prior to committing to the purchase of the frozen semen.

Unfortunately, what is reported is not always representative of what we see. It is not uncommon to thaw frozen semen with significantly poorer seminal characteristics than what was reported by the person who froze the semen. This is extremely disappointing and often heart breaking for the owner of the bitch who has put in considerable time and significant expense to import the semen all the way to New Zealand. However, at least having a report gives the bitch owner and purchaser a record of what they believe was purchased, in case compensation, replacement or reimbursement of the frozen semen needs to be discussed. Be sceptical when someone reports a post thaw progressive motility of greater than 80%. This would be an unusual finding, with most frozen-thawed canine semen having an average 50-55% progressively motile sperm after thawing.

Thaw Media
Thaw media is a simple extender, usually containing the base chemicals of the freezing extender being used for freezing but without egg yolk or glycerol. For some freezing techniques, especially those that freeze in pellets, it is just saline that the frozen semen is thawed in. It is important to understand, that in most situations thaw media is not essential for thawing semen and won't make sub-standard semen great semen, nor does it change the fertility of the semen; there are no magical "accelerants". However, you should check with the providers of the frozen semen if using a thaw media is essential.

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Thaw media can be advantageous in regard to the volume of the insemination dose. Larger volumes can facilitate sperm transport in the uterus so if a thaw media is provided, we will always endeavor to import it and use it. It is also important to note that not all thaw media can automatically enter New Zealand, so we need to investigate its eligibility prior to importation.

What is in an insemination dose?

It is generally accepted internationally by those who freeze and use canine semen that the ***minimum*** number of frozen-thawed sperm per insemination dose is *100 million progressively motile sperm.* Unfortunately, unlike many other species, there is little objective scientific research in this area of canine reproduction as large-scale fertility trials are not possible. Determination of the optimal number of frozen-thawed canine sperm to use for AI has therefore been determined from analysis of retrospective breeding databases.

Furthermore, there is enormous variation in fertility and 'freezability' between individual dogs as the selection pressure for fertility is low. This variation is not only related to inherent individual factors but also breed, age, health and how often a dog has been used at stud.

What we recommend:

- If possible, use two AI doses per heat/breeding (i.e. 200 million motile frozen-thawed sperm rather than 100 million motile frozen-thawed sperm):
  In our experience, and that of others, particularly in Europe, this results in significantly higher pregnancy rates and litter sizes. Unfortunately, this is often not economically feasible as a stud fee arrangement is often sold as a 'dose' and most canine AI doses are frozen as 100 million sperm. Although litters have been produced with only 20 – 50 million frozen sperm, this does not happen "commonly" and is inadvisable when purchasing frozen semen.

- Purchase an ejaculate rather than a dose of semen if possible:
  One ejaculate can provide between 5 and 10 AI doses. Limiting ourselves to a single AI dose of '100 million sperm' when the costs involved in importing frozen semen from overseas, monitoring a bitch for ovulation timing and ultimately carrying out an intrauterine insemination are significant, is not ideal. Therefore, if you are able to negotiate the purchase of an 'ejaculate' rather than an AI dose this will give you much greater control over the number of sperm inseminated per bitch. Stud fees can then easily be determined after AI or after pups are born and should not be linked to a minimum sperm number per AI dose.

Intrauterine Insemination: TCI vs Surgical AI

Too often the discussion of using frozen semen is entirely focused on the intrauterine insemination technique. You will be bombarded with stories about the pros and cons of TCI versus surgical AI. It is important to understand that both these techniques provide the same

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result: sperm are deposited into the same place - that being the uterine horns. Regardless of where the semen is initially deposited in the uterine horns, the sperm will travel up and down each uterine horn and eventually form a reservoir at the opening of each of the oviducts (=site of fertilization). This has been demonstrated by contrast radiographs and is true not only for bitches but ALL female species (cows, mares, ewes, does, humans and sows are all well documented).

Much more emphasis should be focused on frozen semen quality, sperm numbers, male fertility, bitch selection for AI with frozen semen and timing of the insemination, as this is where the variation in success lies (or cause for failure).

Putting it simply, one method (surgery) is invasive and requires a general anaesthetic and therefore carries the risks of mortality, morbidity and is an in-hospital ½ day procedure. For this reason, the procedure cannot be carried out twice, so two inseminations in one heat is not possible.

The other method (TCI) is carried out using an endoscope and is non-invasive, usually requires no sedation and is a fast (5 minutes), visual out-patient procedure, with the owner present always. This means an insemination can be carried out twice or any number of times (depending on semen availability) in the same heat if required. This technique is not offered by many practitioners in NZ due to the costs involved in setting up the endoscopic equipment and the experience required to carry out this procedure accurately.

What we recommend:

- If you as the bitch owner are paying to have the male dog collected and frozen, and also paying a stud fee then you should purchase the WHOLE ejaculate not just one or two doses.
- If you are paying a stud fee then you should request that at least 200 million motile sperm are provided in that dose.
- If the dog owner provides you with less than the international minimum, we recommend that you pay a stud fee only when the bitch is pregnant or if the bitch isn't pregnant, have an agreement in place in that a second dose of semen is provided at no extra cost to you.
- Discuss semen quality with us *prior* to committing to the purchase of frozen semen and before importing semen. Quality is more important than the technique used to freeze the semen. Specifically ask the owner of the dog for a **semen report**.
- Investigate the fertility history of the dog – has he produced puppies? Has his frozen semen produced puppies?
- Import or purchase as much semen as possible – importing can be costly so import more semen than is required so that you have extra doses readily available in New Zealand.

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What is in it for us?

We want what you want – a healthy litter of puppies! We believe that having invested the time and expense, you are entitled to receive sufficient, good quality semen to impregnate your bitch and give her the best possible chance of success.