What Every Mare Owner Needs To Know About Using Frozen Stallion Semen.

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The goal of using frozen stallion semen is of course to get your mare pregnant. The reasons may be varied as to why frozen versus fresh, versus fresh, extended, cooled and transported semen. But, in general, the mare can be kept at home or at least closer to home and inseminated, and the stallion can be from an entirely different part of the country, if not another country entirely. In some cases, the stallion may actually not be alive. To get her pregnant she must have her egg or ovum fertilized, and two events must coincide for fertilization to take place:

1. the mare must ovulate a viable egg (oocyte) during a window of opportunity when there is a viable population of stallion sperm available for fertilization in her reproductive tract; sperm viability can be anywhere from 1 to 48 hours, sometimes up to 14 days, but not reliably.
2. there must be sufficient numbers of viable sperm in the reproductive tract of the mare during the window of opportunity when a viable egg is present and available for fertilization; egg viability is anywhere from 1-6 hours post-ovulation, sometimes up to 12 hours, but not reliably.

Seems simple and straightforward enough. Then why are there so many problems with fertility and conception when using frozen semen? The longevity (i.e., livability or viability) of frozen and thawed stallion semen once deposited into the mare's uterus is quite a bit shorter than when raw, or fresh, extended, cooled and transported semen is used for AI. This makes timing of the insemination in concert with the mare's ovulation much more challenging and critical to success. The 'window of opportunity' (i.e., period of post-thaw viability) for some stallion's frozen / thawed semen may only be from 1 to 6 hours. This problem, while a fact of life when using frozen stallion semen, then also becomes significant to the mare owner as well. He or she must make certain that their mare is examined frequently enough to allow an appropriately timed insemination. Since breeding doses are limited to one or three per cycle, or maybe only one per season, the timing of the AI more often than not can become even more critical to successful fertilization and conception than the stallion's post-thaw semen quality.

There is a great deal of individual variation as to how well a stallion's spermatozoa will survive the challenges of collection, processing for cryopreservation (freezing), storage, thawing, and insemination. Some stallions have excellent conception rates (CR) with artificial insemination (AI) on the farm, or even with fresh, extended, cooled and transported semen. But the same ones may not have respectable CR when their semen is inseminated following freezing and thawing. The following table represents fertility or reproductive efficiency data on 100 stallions selected for good to excellent semen quality when evaluated following freezing and thawing. The

numbers represent the resulting efficiency of getting their respective mares pregnant. The first cycle conception rate (1st cycle CR) is a very useful parameter to know when deciding to select a particular stallion's frozen semen. This means that 50% of the time, an insemination with frozen semen on the mare's first estrous cycle of the season for that year of breeding resulted in conception. The cycles per pregnancy is also a useful measure, and should correlate well with the 1st cycle CR. It makes sense that if the 1st cycle CR was around 50%, then the number of cycles a particular mare may need before becoming pregnant would be close to 2. The seasonal pregnancy rate (PR) is less useful. It means that over the entire breeding season the stallion may have been able to induce pregnancy in 70% of the mares that were inseminated with his frozen semen. This is good information, but it does not tell the mare owner how many breeding doses / inseminations with the frozen semen, or how many estrous cycles each mare had to go through before becoming pregnant with the frozen semen. It is therefore less useful as a parameter of reproductive efficiency.

Reproductive efficiency parameters for frozen stallion semen

Parameter	Selected Stallions
Number of stallions	100
Number of mares bred	641
1st cycle CR (%)	51.3
Seasonal PR (%)	71.9
Cycles per pregnancy	2.08

From Loomis, P.R. The equine frozen semen industry. Anim Repro Sci 68:191, 2001

So ask the stallion owner or breeding manager or the freezing center personnel some or all of the following questions before you select a stallion for frozen semen use in your breeding program.

Q. How many mares was this stallion bred to last season?.....and in the season(s) before that?
A. A useful number to determine, as it may give you an idea of his success rate, unless there were no breedings last year at all. A stallion that bred a good number of mares last year (e.g., > 25) and in the year before that (again > 25) would be an indication that he has 'repeat business', and that the mare owners using him were comfortable with their results. A stallion with no data could be suspect, unless this is his first year in use as a breeding sire with frozen semen available. Again, remember his success rate with fresh on the farm breeding, and fresh, extended, cooled, and transported semen has no bearing on his potential success when using frozen semen.

Q. How many mare-cycles were inseminated last year?
A. If you can get the number of mares booked or inseminated and then the number of mare-cycles inseminated, you now have an idea as to how many cycles each mare was inseminated, an indication of efficiency. But remember, not all mares inseminated 'out there' received similar care with respect to timing of insemination.

Some mares probably received excellent care and others very inappropriate care. Some mares may have been really good breeding candidates, some not. So the information obtained here, while helpful for having some bearing on stallion reproductive efficiency, is biased to a large extent by mare-side factors (e.g., mare age, mare readiness to breed, semen handling and thawing proficiency, timing of insemination, etc.).

Q. How many mares conceived or became pregnant last season?
A. If this is known, then a comparison to the number of mares bred or inseminated in the same season gives you information on the stallion's seasonal pregnancy rate. Conception is defined as a diagnosis of pregnancy between days 14 and 45 after ovulation. Pregnancy is defined as a diagnosis of pregnant (or continued pregnancy) after day 45. The difference is that many mares may be pregnant at day 14-25 after ovulation, but lose the pregnancy before day 45. This is considered an early embryonic loss or early embryonic death (EED). Why the embryo may not have survived is still open to conjecture, and most of the time the mare is at fault. But some stallions are also known to have an inherently high EED rate.

Q. How many insemination doses were required for each mare that became pregnant?
Or
Q. What were the number of mare cycles per pregnancy for this stallion last year?
A. As the table above indicates, a target number here would be around 2; less than this would be excellent. Greater than this may indicate a stallion semen problem; but it may also indicate that some (or many) mares were not managed appropriately, and inseminated at an inopportune time.

Q. What is known about this stallion's 1st cycle CR?
A. This is sometimes a difficult number to obtain, since the data is not collected as often as it needs to be. It is the best information that you could hope to get. If greater than 50%, this should tell you that under optimal breeding management, your mare has a high likelihood of getting pregnant. For all Thoroughbred stallions in the US, breeding by live cover or natural service, the national average 1st cycle CR is not much better than 60%. The 1st cycle CR for Quarter Horse (or Paint, Arabian, Morgan, Standardbred, etc., no favoritism intended) stallions when managed for fresh, on the farm, AI is around 65%. Very few stallions will have a 1st cycle CR >60% with frozen semen. Most will be in the 40-50% range.

Q. What do you expect that his post-thaw motility should be when his semen is appropriately handled?
A. For optimal breeding success, most freezing centers or labs prefer not to send out frozen stallion semen for use in the field with ≤ 30% post-thaw motility. Good stallions should have between 50-60% post-thaw motility. Understand that motility of the sperm population is only one part of the sperm's function required for fertilization or conception. Some stallions have excellent post-thaw motility but fertilize very few of the mares to which they are bred. This is because their sperm cell membranes have

been damaged or altered by the freeze-thaw process, and even though still alive, and still moving, they are unable to attach to the viable egg (oocyte) and complete the events required for fertilization. The breeding industry to date does not yet have a good laboratory method to assess sperm cell membrane functional capacity after freezing and thawing.

Q. What is a calculated insemination/breeding dose for this stallion?
A. Most laboratories or freezing centers will attempt to include at least 400 million progressively, motile sperm in each breeding or mare insemination dose. Some may include as much as 900 million progressively, motile sperm. The breeding or mare insemination dose is NOT the volume (e.g., 0.5 or 5 mL), nor is it the number of straws required for each insemination (e.g. 1 to 8 straws). It is the number of living, potentially functional sperm present when inseminated into the mare at the optimal time for breeding. Stallion fertility or reproductive efficiency is a numbers game!

Q. How many straws are required for each insemination/breeding dose?
A. Important to know for your veterinarian or breeding technician to be able to administer the correct number of sperm for each insemination, but has little or nothing to do with stallion reproductive efficiency.

Q. What am I purchasing?,… a breeding contract with a live foal guarante?… or an insemination dose, or two?
A. A breeding contract will be for your mare to get pregnant, no matter how many times it may take, or in how many breeding seasons. Sometimes the contract may specify a set number of estrous cycle attempts (e.g., 3 or 4) before a mare change or even a stallion change is required / allowed. It may or may not include a live foal guarantee, which means the foal must be born alive, and suckle (nurse), and in some contracts live for the first 5 days after foaling, before the contract is considered executed in full. Such contracts might be considered by some mare owners as good assurance that they will indeed get the foal they intend to produce, however, they can also work against the mare owner. You have a contract to breed to the specified stallion, and he may have less than optimal reproductive efficiency with regard to use of his frozen semen. You may be stuck with a dud! Or the contract may specify that in the event the mare does not settle to the stallion of choice, arrangements can or will be made to provide semen from a ‘back up’ stallion. Not the one you chose to begin with! An alternative that is growing in popularity is to purchase only one or two breeding or insemination doses. If the frozen semen doesn’t work the way it is supposed to, you are not bound by a contract to continue trying to get your mare pregnant. You are free to seek another stallion, and his frozen semen’s assistance. You are generally out less of an economic investment as well.

Mare owners are the consuming public for frozen stallion semen and as a group should demand a quality product. At least as close to quality as is possible with the inherent constraints discussed above kept well in mind. They hold the key to improving frozen semen success rates. They will be the ones that will need to have their mares examined more closely, more frequently, often by a reproductive

specialist to ensure the best result with the frozen semen they have acquired. They must sometimes invest more time and money on their side of the transaction to get their mare pregnant with frozen semen than the stallion owners will invest in procuring frozen semen from their horse.

Of course, all of these concerns may go out the window, if the only stallion you want to breed your mare with is only available to you by frozen semen. But at least you need to understand the risks and the problems. The chances of success are usually never better than 50%, and sometimes a whole lot less. So you can expect to breed your mare on more than one estrous cycle to get her pregnant with frozen semen; you should at least expect 2 cycles. You may have to breed her four or more cycles in the same season to get her pregnant, and yet, she may still not conceive. Breeding with frozen semen is partly a science, partly a technique, partly an art, and partly chance! Never should you expect it to go like 'clock-work', and never should you expect it to be 100% successful.

What can you do to optimize your chances of success? That will be the topic of my next article.