



HANDLING OF FROZEN STALLION SEMEN

Puget Sound Equine Reproduction Center

These instructions are specific only to 5.0 ml straws prepared at PSERC. It is critical to precisely follow the instructions provided by the facility where the semen originates.

Each insemination dose consists of one billion progressively motile spermatozoa in a 4.0 ml volume packaged in 5.0 ml hard plastic straws sealed with spheres of steel or plastic. The name of the stallion, farm name, registration number, and freezing extender are written on the outside of the straw. The straw should not be lifted above the neck of the liquid nitrogen container for more than a couple seconds until it is time to thaw it. **SUCCESS DEPENDS UPON FOLLOWING THE INSTRUCTIONS CAREFULLY!**

THAWING

Thaw the semen in 37 degree water for 60 seconds. Preferably use a transparent container which is at least seven inches high and of approximately one quart capacity so that the entire straw can be submerged in water. A large glass beaker or plastic thermos works well. Fill the container with water at 37 degrees and transfer the straw from the liquid nitrogen container to the warm water as fast as possible and start the timer. The straw should be thawed for exactly 60 seconds in an upright position and should be held with a long forceps. You may want to watch the air bubble in the center of the straw rise to the top. This will indicate the semen is thawed.

WARNING

ALWAYS USE PROTECTIVE GLASSES WHEN WORKING WITH LIQUID NITROGEN. STRAWS MAY EXPLODE AT THAWING IF THERE IS A CRACK IN THE PLASTIC. DIRECT THE TOP END OF THE STRAW AWAY FROM YOU AND OTHER PEOPLE WHEN THAWING.

INSEMINATION

Wipe the straw dry and cut off the ball in one end with heavy-duty scissors. Invert the open end into a sterile 10cc red top test tube maintained at 37 degrees C. Cut off the top end of the straw allowing semen to flow into the test tube. Then attach an artificial insemination breeding pipette to the syringe and aspirate the semen. The entire balance of semen will be in the pipette so handle it very carefully.

Introduce the pipette into the uterus as usual and deposit the semen. Be careful so that the semen is not lost from the pipette before reaching the site of insemination - the volume is small! You may add up to 10cc's of pre-warmed (37 degree C) extender (E-Z Mixin BF) to the semen sample at the time of insemination to provide a larger volume.