# **Thawing Instructions for Frozen Semen**

This is strictly a standard thawing protocol

Most of the time, the person responsible for freezing the semen, will include the specific thawing instructions because every stallion freezes and thaws at a relative different rate. I be very cautious to follow the specific recommended thawing directions for the stallion whose semen you are about to thaw. These instructions are "basic" in that if all eles fails and you have no directions, these are the industry standard for the past 7-10 years.

## Step #1 – **Preparing the Water Bath**

- 1. Warm up a small Styrofoam cooler with hot water so that the sides take and hold temperature.
- 2. Set your water temperature using a digital temperature to 37° C coming out of your tap.
- 3. After the water temperature stabilizes.
- 4. Begin to fill the Styrofoam cooler until it is about half full.

# Step #2 – $N_2$ Care and Selecting the Semen Straws

- 1. Roll your N<sub>2</sub> holding tank close to the cooler
- 2. Make sure you know the straws per dose so that you do not thaw more straws than needed.
- 3. Quickly identify the straws desired for thawing.
- 4. Drop back the canister into the nitrogen tank so that the other straws do not spoil.
- 5. Drop each straw into the warm water as quickly as possible and let them warm up for 3 to 5 minutes.

## Step #3 - Handling the Straws of Semen Post Thaw

There are two methods of handling a straw of semen

### Method #1

- 1. Pull the completely thawed straw of semen out of the Styrofoam cooler.
- 2. Thoroughly dry off the straws with a paper towel
- 3. Make sure that no water has contact with the semen.
- 4. Using a straw cutter
  - a. Clip off the opposite end that has the cotton plug
  - b. Clip off one end if plastic or steel balls are used to plug the straw.
- 5. Drop in disposable insemination gun and inseminate under normal protocol.

### Method #2

- 1. Pull the completely thawed straw of semen out of the Styrofoam cooler.
- 2. Thoroughly dry off the straws with a paper towel
- 3. Make sure that no water has contact with the semen.
- 4. Using a straw cutter
  - a. Clip off the opposite end that has the cotton plug
  - b. Clip off one end if plastic or steel balls are used to plug the straw.
- 1. Remove the top/plug from a red top blood tube
- 2. Placing the straw into the tube, slowly begin to cut the top of the straw so that the vacuum is lost in the straw.
- 3. The semen will easily drain into the blood tube, you may need to tap the top of the straw if a bubble persists.
- 4. After the entire dose has been deposited into the blood tube, using an insemination pipette draw out all of the semen into the pipette
- 5. Make sure you draw the semen at least half way up into the pipette, so that a vacuum dose does not draw the semen into the vagina prior to entering the uterine body.
- 6. Simply inseminate to a standard protocol.

This procedure saves from having to reload the insemination gun for each straw.