FROZEN SEMEN THAWING DIRECTIONS

IMPORTANT! PLEASE READ BEFORE THAWING STRAWS!

EQUIPMENT NEEDED:

- Modena Extender or SGI Ready Made Extenders
- Distilled water (can be purchased from select grocery or department stores)
- Graduated plastic insemination bottles (capacity of 80 ml minimum)
- Sterile glass or plastic container for mixing extender (1-2 Liter capacity)
- Sterile glass or plastic beaker for pouring extender into bottles; OR ready-made extenders purchased from SGI
- Water bath OR sink to thaw straw(s)
- Timer with minutes and seconds
- Tongs or large tweezers to remove straws from tank
- Paper towels
- Scissors
- Celsius or Fahrenheit thermometer

FROZEN SEMEN EXTENDER—MODENA: The SGI freezing process is performed using Modena Extender due to its past and present success in our freezing process. Please have Modena Extender or SGI Ready Made’s on hand for the thawing process. Modena Extender and SGI Ready Made can be purchased from SGI.

READY MADE EXTENDERS Upon arrival, place Ready Made Extenders in a freezer (-20 degrees Celsius) future use. Extenders expire 6 months after production day and the bottles contain the day of expiration. To thaw, set the Ready Made Extenders out at room temperature overnight.

MODENA EXTENDER Prepare SGI Modena Extender as directed and keep at 35°C. Pour 80 mL of extender into graduated plastic insemination bottles, which can be purchase from SGI. The extender is now ready for use or may be stored in a freezer for 6 months.

EXTENDER TEMPERATURE: Extender for thawing needs to be at 20°C/68°F. Warming or cooling the extender may be necessary to achieve this temperature. This may be done by placing bottles
of extender in a refrigerator or sink of cold water for a few minutes if it is too warm, or placing in a sink of warm water if the extender is too cold.

**WATER BATH TEMPERATURE:** The water used to thaw the straws must be at 50°C/122°F. If using an electrical water bath, be sure the temperature is set to 50°C. Always check the temperature of the water bath with a thermometer before thawing. If using a sink, fill with hot tap water and adjust the temperature as necessary by either adding boiling water or ice water.

**THAWING THE STRAW:** Place the liquid nitrogen tank close to the water bath being used to thaw the straws. This will reduce the amount of time the straws will be exposed to ambient temperature during the transfer. Quickly remove one straw at a time from the nitrogen tank with a pair of tongs and place in the water bath. Pulling a canister up into the neck of the nitrogen tank to remove straws will be fine, but remember to lower the canister back into the nitrogen one you have removed a straw. Allow the straw to thaw for exactly 45 seconds.

After 45 seconds, remove the straw from the water bath and dry off the straw well with a paper towel. Any excess water that may come in contact with the semen will kill the sperm cells. Cut the straw below the colored or silver bead and place the open end into the extender bottle; allow the air bubbles in the straw to gather at the closed end of the straw. Once the air bubbles have all gathered cut the closed end as well.

Once all the semen has flowed out of the straw, take out the straw, put the cap back on the bottle and gently mix. You are now ready to inseminate your animal.

**ANALYZING (OPTIONAL):** You may analyze the quality of the semen in the straw that was just thawed. To do this, remove a small sample of the extended semen and place in a small sample tube, small sealed container or Ziploc baggie. Make sure to warm this sample for 30 minutes prior to analyzing. To warm, microwave a gel pack until it is warm to the touch of the inside of your wrist and place the sample on top of the gel pack in a small closed environment. A Styrofoam container or cooler will work.

While the sample is warming, insemination needs to be taking place. Frozen semen viability decreases over time and should not be used passed 1 hour of thawing.

When analyzing the sample, make sure to use both regular slides and caffeine slides. This helps to accurately estimate the cells potential as it would be if located in the reproductive tract. When analyzing frozen semen, samples with 40% or more live cells is normal.

**TRANSPORTATION TO BREEDING AREA:** Place the extended semen in an insulated container with warm gel packs. Remember to use the semen as soon as possible and to not use it past 1 hour of thawing for maximum conception rates.
**SHORTENED INSTRUCTIONS**

1) Warm the bottles of extender to 20°C (68°F)
2) Heat the water bath or sink of water to 50°C (122°F)
3) Put the straws of semen into the water bath/sink for 45 seconds
4) After 45 seconds remove the straws of semen from the water bath/sink. Make sure to dry the straws well; any excess water that gets into the semen will kill the sperm cells
5) Cut the straw below the colored/silver bead and place the open end into the extender bottle; allow the air bubbles in the straw to gather at the closed end of the straw; once the air bubbles have all gathered cut the closed end as well
6) Gently shake the bottle with the semen and extender in it; your semen is now ready for insemination

**INSTRUCTIONAL VIDEO:** [https://www.youtube.com/watch?v=9_wPfl2RZr8&feature=youtu.be](https://www.youtube.com/watch?v=9_wPfl2RZr8&feature=youtu.be)

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<th>Single Insemination</th>
<th>Double Insemination</th>
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<td><strong>Gilts</strong></td>
<td>29-32 hours</td>
<td>1&lt;sup&gt;st&lt;/sup&gt; 24-29 hours</td>
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<td><strong>Sows</strong></td>
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<td>1&lt;sup&gt;st&lt;/sup&gt; 29-32 hours</td>
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The above times are hours after the sow or gilt first exhibits standing estrus (wait the above number of hours after the sow or gilt will stand for a boar to mount before insemination). Double insemination is recommended.

**DRY SHIPPER PROCEDURE:**

1. Once you receive the dry shipper, transfer the frozen straws into a liquid nitrogen tank or use as soon as possible. The dry shipper only holds a charge for 7 days.
2. To send the dry shipper back to SGI, please call the office and let us know you are ready to have SGI issue a call tag through UPS.
3. Set the empty dry shipper in the same location it was delivered to. UPS will print a label and pick up the tank to ship back to SGI.
4. Your rental of the dry shipper is for a maximum of 10 days. After 10 days, you will be charged $50 each week until the tank is returned.
5. The dry shipper is property of Swine Genetics International.