# Nidacon News

The news letter from your ART supplier • No 1 • 2023



# Sperm CryoProtec

The new bottle for Sperm CryoProtec was launched about a year ago and we hope that you are satisfied with the new clear bottle with a screw cork instead of the former darker bottle with a stopper.

The stopper was replaced with a screw cork in order to make it easier to use, the small stopper was a bit difficult to handle and therefore easy to contaminate. The screw cork ensures a safer handling in the lab.

The composition of SpermCryoProtec has not been changed. It is still using glycerol as cryoprotectant, the proportion reduced as far as possible to minimize toxicity to sperm, while still providing maximum cryoprotection. SpermCryoProtec maintains a high concentration of glucose as an osmotic agent,

however, in order to reduce intracellular water and reduce the risk of damage due to ice-crystal formation.

### Advantages of freezing sperm with Sperm Cryo Protec

- Maximum procedure flexibility
- Maximum survival rate
- Long shelf life, even after opening
- No antibiotics

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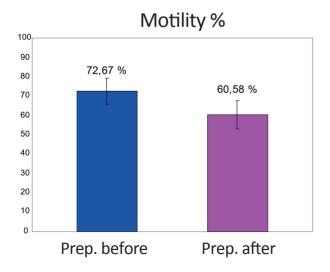
Naturally, obtaining the best survival results when freezing and thawing sperm requires a suitable cryoprotective medium. The medium is however not all that is needed. A suitable protocol for freezing and thawing can give not only better results but also more consistent and controllable results.

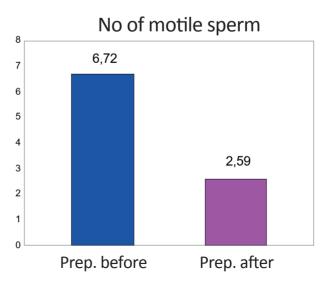
In this newsletter we will go through all the steps of the methods in order to help you get the best results possible when freezing sperm.

While it is possible to freeze unprocessed semen, studies performed at Nidacon have shown that using gradient prepared sperm for freezing provides a higher recovery rate and a higher percentage of motile sperm compared to freezing ejaculates followed by post-thaw gradient preparation.

Using a gradient before freezing removes seminal plasma as well as ROS and their sources, ensuring optimal recovery of sperm on thawing. While this entail a bit more work for the lab before freezing, the results are well worth the effort

### Preparation before and after freezing





## How to freeze gradient prepared sperm

Reagents and Equipment needed SpermCryoProtec (Nidacon) Steile pipettes and tubes Plastic cryostraws CryoFloater (Nidacon) Steel tray Container with liquid nitrogen Forceps

 Add 1 part of SCP to 3 parts of sample. Add slowly – one drop at a time – and mixing after adding each drop.

#### Tips

- You can find a dilution table on Nidacon website, www.nidacon.com
- Spermatozoa should be diluted slowly to allow gradual osmotic adjustment to take place between the intra- and extracellular compartments, thus, preventing or minimizing the osmotic shock phenomenon from occurring. SCP has an osmolality around 3000 before dilution and the medium around 290, the final diluted solution will have an osmolality around 1000.
- Use a pipette or a syringe to fill the straws with the sperm suspension. Seal the straws

3. Place the straws in the refrigerator for 10-60 minutes for equilibration.

#### Tips

- Use a steel tray for the equilibration in the fridge, that way the straws will remain cold when removed from the refrigerator. The tray can also be pre-cooled and used when preparing the straws.
- The equilibration time is set to provide some flexibility in the labwork. The results
- will improve with incubation in the fridge compared to room temperature, 10 minutes is however enough for a good survival rate, only a slight increase for the full 60 minutes.
- 4. Place the cryofloater on the liquid nitrogen, then place the straws using a forceps on the floater.

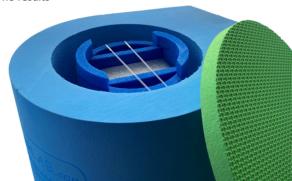
#### Tips

- Do not touch the straws with your hands after the equilibration, they will quickly warm up. Use forceps.
- The CryoFloater provides a stable raft with a constant and correct height above the nitrogen surface, guaranteeing optimal freezing temperature and the best possible result.

5. Leave on Cryofloater for 10-30 minutes.

#### Tips

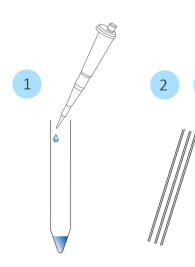
- Lid can be used or not, it will not affect the survival rate.
- Once again, a flexible time range and for this range no difference in result between 10 and 30 minutes. Just make sure that the amount of liquid nitrogen is enough if using the 30 minutes.

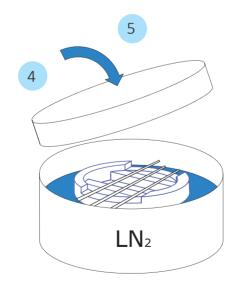


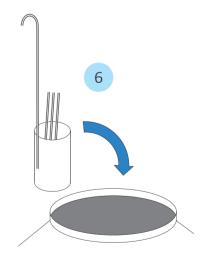
**6.** Tip the floater so that the straws are submerged in the liquid nitrogen. Straws are ready for storage.

#### Tips

- Transfer to storage device, avoid any warming of the straws.
- Be kind to the CryoFloater after the freezing procedure, it is a bit fragile when cold.







### Thawing gradient prepared sperm

#### Reagents and equipment needed:

PureSpermWash (Nidacon) Sterile pipettes and tubes Scissors Water bath Forceps Centrifuge

1. Remove the straws from the liquid nitrogen tank. Check individual straws for cracks etc.

- If you suspect a crack etc, the straw can be thawed in your hand.
- 2. Place the straws in 37°C water for 30s.

- Thawing the straws with frozen sperm with water at 37°C is recommended. This temperature will give a better survival rate compared to room temperature.
- Instead of using a water bath filled with warm water, a 50 ml tube containing water can be preheated in the incubator to 37°C. Easy way to ensure having the correct temperature.
- 3. Dry the surface of the straw.

4. Cut one end of the straw with scissors. Hold the straw over a tube containing 5 ml PureSpermWash and cut the other end of the straw. Any sperm suspension remaining in the straw can be expelled using a pipette.

#### Tip

- If the sample is going to be used for intrauterine insemination (IUI) the washing step is no necessary. If used for IVF/ICSI washing should be performed.
- 5. Centrifuge for 5 minutes at 500xg. Do not use the brake.

- The proper RPM for your centrifuge can be calculated using a formula on Nidacon website; https://nidacon. com/support/#rpm
- 6. Aspirate PureSpermWash supernatant, leaving as much liquid as required for desired concentration or remove all and replace with fertilisation medium.
- 7. The sample is now ready for use.

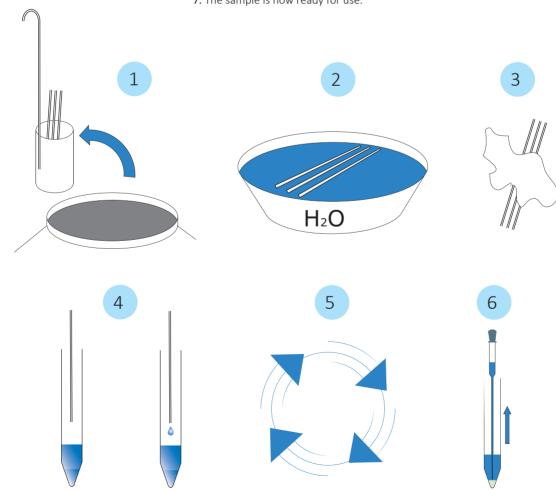
#### Storage and Stability

When stored between 2° and 30°C. Sperm CryoProtec and PureSpermWash has a shelflife of 12 months. Avoid tempera tures above or below these values. The expiry date is shown on both bottles and cartons.

Take care to open and close bottles under aseptic conditions. After opening, store between 2° and 8°C when not in use. Shelflife on the product label applies when the product is stored and handled according to manufacturer's recommendations.

Nidacon uses no added antibiotics, unstable additives, or preservatives in the manufacture of Sperm CryoProtec and PureSpermWash.







## Basic Semen Workshops

We conducted multiple basic semen workshops throughout the year, primarily at our lab in Gothenburg.

However, our latest session took place in Buenos Aires. It's always fascinating to connect

with embryologists from diverse corners of the globe.

Rest assured, we will be organizing more workshops after the summer season!



Nidacon will remain open throughout
the summer, ready to assist you with all your orders,
requests, and any questions you may have.

We wish you a delightful summer
and look forward to welcoming
you at ESHRE!

## Coming up

■ ESHRE 39th Annual Meeting
Nidacon booth no C3-125
Copenhagen, Denmark 25-28 June 2023



■ ASRM 2023 The Past. The Present, and The Pipeline New Orleans, Louisiana , US. October 14-18



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