Nidacon News No 2 • 2012

Frequently asked questions

Some recent questions and answers that might be useful to you.

Are antibiotics included in PureSperm®100?

– No, PureSperm®100 does not contain antibiotics, for the following reasons:

1. The gradient will remove most, if not all, of the bacterial contamination present in the ejaculate, provided that the retrieval of the sperm pellet is carried out according to the instructions given in the package insert.

2. Antibiotics commonly used in cell culture media are toxic to sperm.

What device should I use with the VitriBlast®kit?

Any device can be used. In the manual we have used the cryoloop but it works with all other devices on the market.

Is there any human serum albumin in PureSperm®40 and PureSperm®80?

– Research in our laboratory showed that the yield of motile sperm recovered from the gradient was approximately the same, regardless of whether or not the gradient layers contained human serum albumin. Therefore, we have not included any protein in PureSperm®40 and PureSperm®80. The colloid in these products eliminates aggregation of sperm and reduces their sticking to the centrifuge tube. However, human serum albumin must be added to the "wash" solution, as provided in PureSperm®Wash.

If you have other questions, do contact us!

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Upcoming events

AAEP – Equine conference
2-5th of December 2012,
Los Angeles, United States

21-23rd of March 2013,
Berlin, Germany
Controversies in cryopreservation of stem cells, reproductive cells, tissue, organs & cryo surgery.

SSRM
– Swedish society meeting
12-13th of April 2013,
Stockholm, Sweden

ESHRE –7-10th of July 2013,
London, United Kingdom

Christmas holiday – Nidacon will be closed from the 24th of December until 2nd of January

We believe in the strength of networking and the exchange of knowledge.

Therefore, we are available on Facebook! We publish the latest news, discussions, pictures from conferences, invitations to workshops, campaigns and much more.

You can find a link on our website www.nidacon.com

facebook

Please become our friend on Facebook and you will also keep updated on our latest happenings!

Who to contact

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Nidacon News

The news letter from your ART supplier • No 2 • 2012

Contents

The device that will make your sperm preparation easier

This novel device consists of an insert within a centrifuge tube. The outer chamber of the ProInsert™ is designed for easy layering of PureSperm gradient layers and for adding the semen sample on the gradient prior to centrifugation.

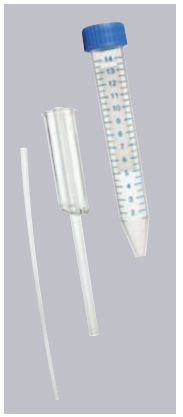
> you will achieve a very clean preparation, and avoid the risk of recontamination.

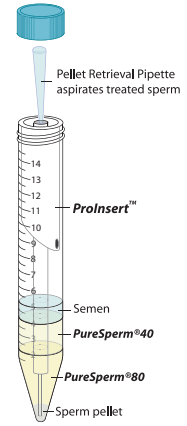
During preparation of sperm from a semen sample using a density gradient separation, the ProInsert™ facilitates both the gradient preparation and the pellet retrieval.

The central channel is designed for the safe and easy removal of the sperm pellet after centrifugation. You will achieve a very clean preparation, and avoid the risk of recontamination.

You will also save time; studies have shown that it takes half the time to prepare a sample using the ProInsert™ compared to the standard method for density gradient preparations.

The kit consists of 5 sterile tubes including the insert, tubes for the washing of the pellet and 5 specially designed pipettes for retrieval of the pellet.





"Elimination of bacteria from human semen during sperm preparation using density gradient centrifugation with a novel tube insert"
J. Fourie, N. Loskutoff & C. Huyser
Andrologia Vol 44, pp 513-517, 2012

Use of a novel washing method combining multiple density gradients and trypsin for removing human immunodeficiency virus-1 and hepatitis C virus from semen
Loskutoff, Huyser, Singh,Walker, Thornhill, Morris, Webber, Path
Fertility and Sterility Vol.84 No 4, October 2005

This device has been tested in several IVF clinics and is used routinely at the clinic in Pretoria, South Africa; results from there have been published by Dr. Carin Huyser and her group.

All components are gamma sterilized and mouse embryo tested.
If you are interested, please contact Nidacon and we will be happy to supply you with a sample.

Ordering information:
5 Sterile one patient kits in each package.
The product cat. no. is PI15-5

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Evaluation of the effects of different in vitro incubation conditions on sperm DNA integrity

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Abstract

Introduction:

Prolonged in vitro incubation of spermatozoa has been shown to have adverse effects on sperm motility, vitality as well as on DNA integrity. Knowledge regarding how shorter incubation periods prior to the IVF/ICSI procedure affect semen quality is however limited.

The aim of the present study was to examine if sperm DNA integrity was affected during incubation in three different conditions for 2 hours after sperm preparation prior to the IVF/ICSI procedure.

Materials and Methods:

Density gradient centrifuged samples from two hundred men undergoing infertility work-up were included in the study.

Following gradient centrifugation one reference sample was frozen immediately.

Thereafter samples were divided into three aliquots and incubated for two hours in either

1) room temperature (23-24°C);

2) in a 37°C humidified incubator with 6%CO_2 and 5%O_2 or

3) in a 37°C humidified incubator with atmospheric air. The Sperm Chromatin Structure Assay (SCSA) was used to assess the extent of sperm DNA damage. Sperm DNA fragmentation was expressed as DNA fragmentation index (DFI).

Mona Bungum, laboratory manager.

Results:

A statistically significant increase in DFI was seen in density gradient prepared samples incubated for 2 hours at 37°C, 6%CO_2 and 5%O_2 compared to the reference sample taken immediately after preparation.

This was the case also for samples incubated at 37°C in atmospheric air. Moreover, statistically significant lower DFI levels were seen in the group incubated at room temperature compared to those incubated at 37°C, 6%CO_2 and 5%O_2 or at 37°C in atmospheric air.

Conclusions:

In order to prevent against further sperm DNA damage after density gradient preparation prior to the IVF/ICSI procedure, spermatozoa should be stored at room temperature.

This poster was presented at ESHRE in Rome 2011 by the Skåne University Hospital, Reproductive Medicine Centre, Malmö, Sweden.

Research at the clinic is an important part of their work and, today they have extensive research in both already established areas and new ones.

Research, development and the clinical operation of the centre in Malmö are closely connected.

VitriBlast™ and ThermoBlast™

Now even longer shelf life!

After trials carried out at Fertility Centre ART-clinic at Carlanderska Hospital, Gothenburg we are now able to prolong the shelf life to 12 months instead of 9 months.

This will help us ensure that products are always on the shelf for you and, also that you can use the same batch for a longer period.

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Two useful products from Nidacon

Sperm CryoProtec™II

This is a cryo medium designed especially for human sperm. The concentration of the cryoprotectant glycerol has been reduced as far as possible, and significantly minimises toxicity to sperm, while still providing cryoprotection. Moreover, a high glucose concentration functions as an osmotic agent, to reduce intracellular water. In turn ,this reduces ice-crystal formation and membrane damage.

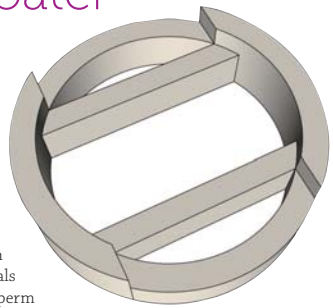
As well as optimising the formulation of Sperm CryoProtec™II, the methodology for using this product has been carefully adjusted to give optimal sperm survival during the cryopreservation.

The methododology recommended by Nidacon is easy and will give a high proportion of motile sperm after thawing.

You can find more information about the methodology on our website www.nidacon.com

CryoFloater™

We have now also the two different CryoFloaters™ to help improve cryo results even further. The CryoFloater™ is a floating device made of Polyethylene. Its intended use is to float on liquid nitrogen carrying straws or vials of semen/prepared sperm for freezing. It provides a constant distance between the sample and the nitrogen surface, to standardize the freezing rate.



Studies carried out at Nidacon showed that different heights of the cryofloater did give different survival rates of the sperm.These studies have lead to optimisation of the CryoFloater™ height to give the best result, and still be stable enough and easy to put the straws and vials on.

Test of CryoFloater™ height

Motility after thawing, percentage and grade of motility. Test of 2 different heights over the liquid nitrogen surface.

Spermdonor	1.5 cm over LN_2	3 cm over LN_2
	% motility	% motility
1	65 (3)	10 (2)
2	60 (2-3)	25 (2)
3	55 (2-3)	10 (1-2)
4	55 (2-3)	25 (no forward motility)
Average	58.75	17.5

Comparing controlled rate cryopreservation with Nidacon's manual method with a CryoFloater™ The Academic University Hospital in Uppsala, Sweden has been in combination with SpermCryoProtec™II for several years using controlled rate cryopreservation equipment for freezing.

They have now completed a study where they compared the controlled-rate cryopreservation (CR) with the simplified manual method from Nidacon, using CBS straws for both methods.The conclusion of the study showed :

1) no significant difference between CR and manual cryopreservation of sperm

2) the manual method is cheaper and easier to perform

News from the animal world

Nidacon has ongoing interesting collaborations with Schockemöhle Pferdehaltung, Germany, Cecolfes IVF clinic, Colombia and IVI Barcelona, Spain regarding sex determination of equine embryos.

In horses, pre-implantation genetic diagnosis (PGD) could be a very valuable technology for the equine breeding industry, as it would allow elimination of devastating genetic diseases and selection of genetic traits in the offspring produced.

> In horses, pre-implantation genetic diagnosis (PGD) could be a very valuable technology...

Unfortunately, so far, very few biopsied equine embryos have resulted in pregnancies and the genetic results were only determined after the embryos had been transferred. During the studies, the timing was evaluated for carrying out the biopsy and sex determination.

The results were very promising, showing that embryos of a certain size could be biopsied without compromising pregnancy rates. Further studies will follow for improving the technique and making it possible to include all sizes of embryos.