

Nidacon News

The news letter from your ART supplier • No 2 • 2016

Optimize your sperm preparation

PureSperm®90 has joined the PureSperm family and is now available.

This product makes the line of ready-to-use products more complete and it is now possible to perform different density

gradient preparations very easily. All of the ready-to-use PureSperm products have also been updated and their formulations optimized.

The raw material, the silane coated silica is still our own unique material. It is produced by Nidacon and only used for the PureSperm products.

All of the ready-to-use PureSperm products have also been updated and their formulations optimized.



Contents

Optimize your sperm preparation	1
The importance of storing the sperm	2
The 2016 Ig Nobel Prize Winner	2
Frequently asked questions	3
Processing of semen by density gradient centrifugation selects spermatozoa with longer telomeres for assisted reproduction techniques	3
Tips & tricks	4
New distributors	4
Upcoming events	4
Who to contact	4

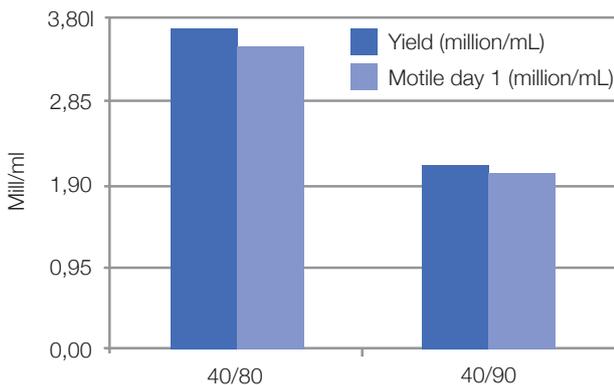
But is it necessary to have different density gradient preparations? No, it's not necessary, if 40/80 is used or 40/90 for all your patients, you will have very good preparations, regardless.

It is, however, possible to use 40/80 for inseminations, donors, samples where you need a high yield and use 40/90 for samples where a higher percentage of motile sperm is desired. As shown in the two graphs the difference is not large but sometimes it's the small things that make a difference.

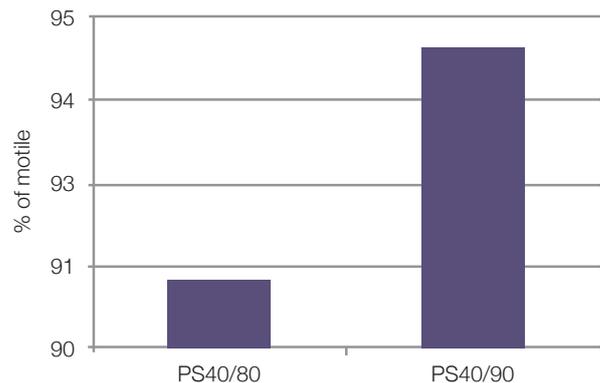


Product Specialist
Ms. Ann-Sofie Forsberg
Direct +46 31 703 06 42
ann-sofie@nidacon.com

Comparison of yield



Comparison of % of motile sperm



► The importance of storing the sperm sample in Room Temperature and avoiding temperature fluctuations

It has proven to be of great importance for the outcome that the sperm sample is maintained in a temperature as constant as possible and that the temperature should be around room temperature. We here intend to further clarify with references to publications on the subject.

1. Activation of enzymes

The ejaculate contains enzymes which start working directly after ejaculation. This results in an increase in osmolality. If the sperm sample is stored in an incubator (37°C), the enzymes work more efficiently and the increase of the osmolality will occur much faster. The difference in osmolality between the ejaculate and the gradient or swim-up medium will be higher than if the sample had been stored at room temperature. As is well known, sperm do not like shocks (cold-shock, dilution shock, osmolality shock and so forth)

Ref: Osmotic shock induces structural damage on equine spermatozoa plasmalemma and mitochondria. L. Gonzalez-Fernandez et al, Theriogenology 78 (2012) 415-422

Similar studies have been performed on a number of different species. A PhD student at Nidacon will soon publish results with human sperm where this is confirmed. The results so far also show negative effects on motility and DNA-integrity.

2. Temperature fluctuation

In the reference below, temperature fluctuation and motility is discussed. The conclusion is that the slightest jump up or down in temperature has a negative effect on motility, and causes hyperactivity.

Ref: Behavioral mechanism of human sperm in thermotaxis: a role for hyperactivation, Sergii Boryshpolets et al, Hum Rep, 2015

The findings of this study show that the fraction of hyperactivated spermatozoa is tightly dependent on the temperature and, especially, on temperature changes. This is because hyperactivation events could be generated as a result of temperature fluctuations. With the extreme temperature sensitivity of human spermatozoa and their response to even subtle temperature changes (Bahat et al., 2012), it is of utmost importance



to maintain a well-controlled constant temperature in studies of sperm motility.

3. The WHO-manual

The WHO-manual recommends storage of sperm samples in constant temperature. However, it does define the temperature to be between 22-37°C, but still constant in that interval. "Avoid changes in temperature".

With above in mind, the optimal storage temperature is room temperature, from ejaculation until the washed sperm are introduced to the oocyte.



Animal Product Specialist
Ms. Anna Niläng Laessker
Direct +46-31-703 06 30
anna@nidacon.com

► The 2016 Ig Nobel Prize Winner

The Ig Nobel Prizes honour achievements that make people LAUGH, and then THINK. The prizes are intended to celebrate the unusual, honour the imaginative – and spur people's interest in science, medicine, and technology

REPRODUCTION PRIZE [EGYPT] –

The late Ahmed Shafik, for studying the effects of wearing polyester, cotton, or wool trousers on the sex life of rats, and for conducting similar tests with human males.

Effects of different types of textiles on male sexual activity

Ahmed Shafik European Urology, vol 24 no 3, 1993

Abstract: The effect of different types of textile underpants on sexual activity was studied in 50 men. All the subjects were potent and sexually active. They were divided into 5 equal groups: 4 tests and 1 control. Each of the 4 test groups were

dressed in one type of textile underpants made of 100% polyester, 50/50 polyester/cotton mix, 100% cotton, or 100% wool.

Sexual behaviour was assessed before and after 6 and 12 months of wearing the pants, and 6 months after their removal. Behavioural response was rated as potent if the subject's penis became erect, entered the vagina, and ejaculated. The rate of potent intromission (I) to mounts (M) (I/M ratio) was determined. The electrostatic potentials (EP) generated on the penis and scrotum was measured by an electrostatic kilovolt-meter. The I/M ratio at 6 and 12 months of wearing the polyester and polyester/cotton mix

pants was significantly reduced compared to the pre-test levels and the controls (p < .001).

The reduction was more manifest in the pure polyester than in the polyester/cotton mix group, and at the 12-month than at the 6-month examination.

The polyester-containing pants generated EP, which may induce electrostatic fields in the intrapenile structures and could explain the diminished sexual activity. The cotton and wool textiles did not generate EP. Thus, polyester underpants could have an injurious effect on human sexual activity.

Conclusion: If you are thinking about buying underpants for your husband or anyone else for Christmas, make sure that it's of the right material.



▶ Frequently asked questions

How do I calibrate my centrifuge to make sure that I use the correct g-force?

To achieve the optimal result using a density gradient, it is critical, to make sure that your centrifuge uses the correct g-force. You can either use the equation and do the calculation or use the RCF nomograph

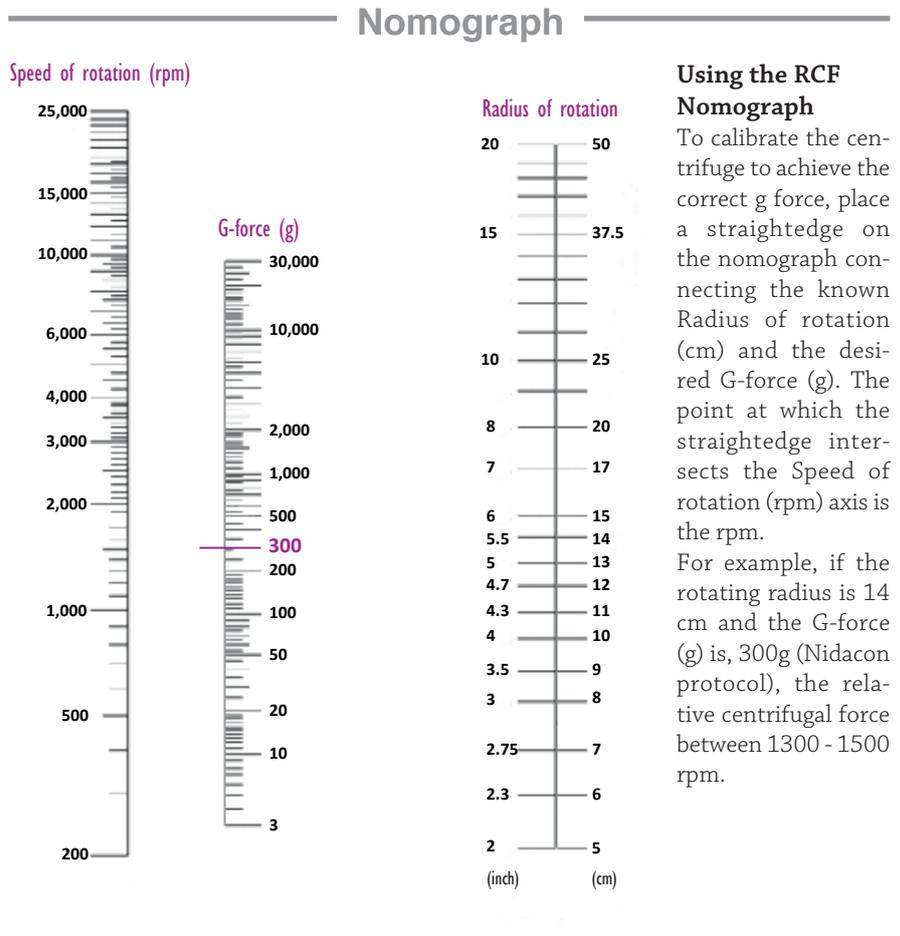
$$Rpm = v[(g/(1.118 \times r))] \times 10^3$$

- r = rotational radius, the distance (mm) from the center of the rotor to the bottom of a centrifuge tube in the bucket when raised to horizontal position

For example; to achieve 300 x g when radius = 165 mm the centrifuge speed must be:

$$Rpm = v[(300/(1.118 \times 165))] \times 10^3 = 1275$$

- By using the RCF Nomograph



Using the RCF Nomograph

To calibrate the centrifuge to achieve the correct g force, place a straightedge on the nomograph connecting the known Radius of rotation (cm) and the desired G-force (g). The point at which the straightedge intersects the Speed of rotation (rpm) axis is the rpm.

For example, if the rotating radius is 14 cm and the G-force (g) is, 300g (Nidacon protocol), the relative centrifugal force between 1300 - 1500 rpm.

▶ Processing of semen by density gradient centrifugation selects spermatozoa with longer telomeres for assisted reproduction techniques *Qingling Yang et al,*



Abstract

The ends of eukaryotic chromosomes contain specialized chromatin structures called telomeres, the length of which plays a key role in early human embryonic development.

Although the effect of sperm preparation techniques on major sperm characteristics, such as concentration, motility and morphology have been previously documented, the possible status of telomere length and its relation with sperm preparation techniques is not well-known for humans. The aim of this study was to investigate the role of density

gradient centrifugation in the selection of spermatozoa with longer telomeres for use in assisted reproduction techniques in 105 samples before and after sperm processing.

The possible status of telomere length and its relation with sperm preparation techniques is not well-known for humans.

After density gradient centrifugation, the average telomere length of the sperm

was significantly longer (6.51 ± 2.54 versus 5.16 ± 2.29 , $P < 0.01$), the average motile sperm rate was significantly higher (77.9 ± 11.8 versus 44.6 ± 11.2 , $P < 0.01$), but average DNA fragmentation rate was significantly lower (11.1 ± 5.9 versus 25.9 ± 12.9 , $P < 0.01$) compared with raw semen. Additionally, telomere length was positively correlated with semen sperm count ($rs = 0.58$; $P < 0.01$).

In conclusion, density gradient centrifugation is a useful technique for selection of sperm with longer telomeres.

► Tips & Tricks

We hope you have had some use of the tips & tricks that we have started to send to you.

Our hope is to provide you with the small details that can make a big difference but are difficult to find in literature and not often talked about at conferences.



If you have any questions regarding small details, how to make the most of the procedure or product, please let us know, most likely, many others have asked themselves the same thing.

If you are not receiving our monthly tips&tricks, send us an e-mail.



KAM, Latin America
Mr. Mauricio Lucena
Direct +46 31 703 06 39
mauricio@nidacon.com

► Upcoming events

- Repronion workshop Sperm DNA damage and strategies for its reduction, Copenhagen, Denmark November 28th.



- Swedish Society for Reproduction Annual meeting Uppsala, Sweden March 31th-April 1st.



- ESHRE <https://www.eshre.eu/Annual-Meeting/Geneva-2017>.



► New distributors

Polycompany International group S.A is a new Nidacon distributor in Chile.

We would like to welcome Alejandro Mucientes and colleagues at Polycompany International group S.A as our new distributor in Chile.



In Vitro Life is a new Nidacon distributor in Peru.

We would like to welcome Marco Arturo Escobar Aguilar and colleagues at In Vitro Life as our new distributor in Peru.



► Who to contact



Product Specialist
Ms. Ann-Sofie
Forsberg
ann-sofie@nidacon.com
Tel: +46-31-703 06 42



Logistics
Mr. Dennis
Johansson
dennis@nidacon.com
Tel: +46-31-703 06 37

