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.

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.

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

Comparison of yield

<table>
<thead>
<tr>
<th></th>
<th>Yield (million/mL)</th>
<th>Motile day 1 (million/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>40/80</td>
<td>3.80</td>
<td>2.85</td>
</tr>
<tr>
<td>40/90</td>
<td>2.85</td>
<td>1.90</td>
</tr>
</tbody>
</table>

Comparison of % of motile sperm

<table>
<thead>
<tr>
<th></th>
<th>% of motile</th>
</tr>
</thead>
<tbody>
<tr>
<td>PS40/80</td>
<td>90</td>
</tr>
<tr>
<td>PS40/90</td>
<td>94</td>
</tr>
</tbody>
</table>
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

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.

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.
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.

\[ \text{Rpm} = \frac{v}{g/(1.118 \times r)} \times 10^3 \]

- \( r \) = 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:

\[ \text{Rpm} = \frac{v}{(300/(1.118 \times 165))} \times 10^3 = 1275 \]

- By using the RCF Nomograph

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.

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.

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New distributors

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

We would like to welcome Alejandro Muniches 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.

Upcoming events

- Reprounion 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.


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