An innovative alternative to PVP for ICSI

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Introduction
To enable sperm to be drawn into a pipette for intracytoplasmic sperm injection (ICSI), their motility needs to be reduced, for example, by placing them in a viscous medium. Previously, the only products commercially available for slowing sperm motility contained polyvinylpyrrolidone (PVP).

However, some PVP is injected into the oocyte along with the sperm, which has been reported to cause problems. For example, Dustner et al. (1995) reported that exposure to PVP prior to ICSI damages the sperm plasma membrane, allowing thiol-reducing agents to gain access to the sperm nucleus, and furthermore, the PVP may prove interferes with sperm nucleus decondensation.

Feichtinger et al. (1995) observed a much lower incidence of chromosomal abnormalities in newborn babies where a modified ICSI technique (which did not require the use of PVP) had been used, compared to some other groups using PVP. Other researchers, for example Hinika et al., (1998), Meng Yin Tai et al., (2000), also agree that the use of PVP should be avoided and have developed a modified ICSI technique to capture sperm without PVP. For some years, a physiological alternative to PVP has been needed for reducing sperm motility to facilitate their capture. Moreover, the product should possess specific characteristics, as follows:

(i) Be viscous, to slow sperm motility sufficiently to aspirate them into an ICSI pipette;
(ii) Be sufficiently fluid, to facilitate ease of aspirating and dispensing the sperm and a little of the liquid into, and from, the pipette;
(iii) Be able to prevent sperm sticking to either the plastic culture dish or the glass ICSI pipette;
(iv) Have no deleterious effects on post-ICSI zygote development.

Objectives
This study was designed to evaluate SpermCatch™ (NidaCon International, Gothenburg), a viscous liquid containing only substances which occur naturally in the human reproductive tract. Being wholly physiological, SpermCatch™ may prove to be a substitute for PVP to reduce sperm motility.

Materials and Methods
Ejaculates from healthy volunteers, whose fertility status was unknown, were prepared on PureSperm® density gradients by centrifugation at 300 g. After washing the resulting sperm pellet in PureSperm® Wash by centrifugation at 500 g, and resuspending in a gamete holding medium, spermAssist™, aliquots of the sperm preparations were added to 10 µL drops of SpermCatch™ under oil (NadOil™). All materials were obtained from NidaCon International AB, Gothenburg.

In a separate study, oocytes were injected with sperm exposed to either SpermCatch™ or a commercially available PVP-containing product (ICSI-100; VitroLife, Gothenburg, Sweden). Post-ICSI rates of cleavage and development were compared between the two groups.

Results
Sperm motility was modified immediately in SpermCatch™, with sperm appearing to swim in “slow motion.” Progression forward motility was decreased without any noticeable change in the lateral movement of the sperm head. Motility was reduced compared to sperm in gamete holding medium alone. Although the sperm did not cease moving completely, it was possible for experienced workers to immobilise and catch them individually and draw them into an injection pipette. The SpermCatch™ medium was sufficiently viscous to allow fine control during both sperm aspiration into, and expulsion from, the injection pipette. Sperm did not stick to the plastic culture dish or to the glass pipette.

Computerised sperm motility analysis was performed on sperm exposed to either SpermCatch™ or the proprietary PVP product. Sperm had mean velocities of 49 and 44.5 µm/sec respectively (figure 1), compared to a mean curvilinear velocity of 88.25 µm/sec for sperm diluted in sperm maintenance medium, SpermAssist™ (n = 4). The rates of zygote cleavage and development after injection of the oocyte with sperm exposed to either SpermCatch™ or the PVP product are shown in Table 1. The rates were not different between the two treatment groups.

Conclusions
The new product, SpermCatch™, was capable of slowing sperm motility sufficiently for them to be caught, was easy to handle in the ICSI pipette and prevented sperm from sticking to the culture dish or to the pipette. Although sperm motility was somewhat faster in SpermCatch™ than in the PVP product, there was no difference between the two products in terms of zygote development after ICSI. Therefore, SpermCatch™ represents a safer, physiological alternative to PVP for modulating sperm motility prior to aspirating a single sperm into an ICSI pipette.

Table 1. Proportion of oocytes cleaving and zygotes developing after intracytoplasmic sperm injection with sperm exposed to SpermCatch™ or PVP product.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No. of oocytes injected</th>
<th>No. with two pronuclei (fertilisation)</th>
<th>Cleavage</th>
</tr>
</thead>
<tbody>
<tr>
<td>SpermAssist™</td>
<td>121</td>
<td>90 (75%)</td>
<td>88 (98%)</td>
</tr>
<tr>
<td>PVP product</td>
<td>109</td>
<td>77 (71%)</td>
<td>76 (99%)</td>
</tr>
</tbody>
</table>

References


Figure 1. Motility of sperm exposed to gamete maintenance medium. SpermCatch™ and Polyvinylpyrrolidone (PVP).

Motility velocity (µm/sec)

Gammetes
SpermAssist™
Polyvinylpyrrolidone