The Relationship between the Sperm Deformity Index (SDI), Apoptosis and Sperm Penetration Capacity

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Objective: The sperm deformity index was previously reported to be a powerful predictor of in vitro fertilization outcome. The inclusion of apoptotic sperm during in vitro fertilization was suggested as one of the reasons for suboptimal success rates. The binding with annexin-V microbeads during magnetic cell separation (MACS) can effectively eliminate apoptotic spermatozoa. This study aimed to evaluate the relationship between apoptosis in sperm, SDI, and sperm fertilizing ability utilizing the zona-free hamster oocyte penetration assay (SPA) as a suitable model. Design: Prospective-controlled study. Materials and Methods: Semen specimens collected from 16 healthy donors were prepared by density gradient centrifugation (DGC) followed by MACS using annexin V-conjugated microbeads. A non-separated aliquot of DGC preparation of each sample served as control. Apoptosis was evaluated in spermatozoa using flow cytometry. The oocyte penetration capacity of the annexin-V negative (non-apoptotic) and annexin-V positive (apoptotic) fractions was assessed using SPA. Results of SPA were evaluated as the percentage of oocytes penetrated by sperm. Sperm morphology was assessed using Tygerberg’s strict criteria and the SDI score was calculated by dividing the total number of deformities observed by the number of sperm randomly selected and evaluated, irrespective of their morphological normality.

Results: The mean percentage of normal sperm morphology and the mean SDI scores in the non-separated sperm aliquot and the annexin-negative and the annexin-positive fractions are given in the table. There was a statistically significant difference in the mean SDI scores. Significantly high SDI sores were seen in the apoptotic fraction compared to the non-apoptotic fraction and to the non-separated control sperm aliquot ($P < 0.0001$, and $P < 0.03$, respectively). The mean percentage of normal sperm morphology was similar in the 2 separated fractions and in the control sperm aliquot. The mean SPA for the 2 sperm fractions and the control were statistically different. Annexin-negative spermatozoa had significantly higher SPA compared to annexin-positive fraction and the non-separated control ($P < 0.0001$, and $P < 0.0001$, respectively). In the pooled data there was a significantly negative correlation between the SDI scores and SPA outcome ($r = -$
There was no significant correlation between normal sperm morphology and SPA results ($P = 0.28$).

**Table:** The mean (SD) of SDI scores, percentage normal morphology and SPA results in annexin-negative (non-apoptotic) and positive (apoptotic) sperm fraction and non-separated sperm control. Means were tested utilizing analysis of variance.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Annexin-negative spermatozoa</th>
<th>Annexin-positive spermatozoa</th>
<th>Control</th>
<th>F Statistics ($P$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean SDI Score</td>
<td>1.32 (± 0.1)</td>
<td>1.62 (± 0.13)</td>
<td>1.4 (± 0.1)</td>
<td>29.62 ($P &lt; 0.0001$)</td>
</tr>
<tr>
<td>Mean normal morphology (%)</td>
<td>19 (± 7)</td>
<td>16 (± 6)</td>
<td>19 (± 8)</td>
<td>1.46 ($P = 0.24$)</td>
</tr>
<tr>
<td>SPA (%)</td>
<td>44 (± 12)</td>
<td>20 (± 5)</td>
<td>33 (± 7)</td>
<td>28.57 ($P = 0.0013$)</td>
</tr>
</tbody>
</table>

Conclusion: The study data demonstrate SDI scores to be a powerful surrogate measure of sperm integrity and function. Andrology laboratories may find the assessment of SDI superior to routine assessment of normal sperm morphology in predicting the fertilizing ability of semen samples in routine clinical work.

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