Changing old trends in IVF laboratory cleaning and disinfection

**Aims**

1) To evaluate efficacy of 6% hydrogen peroxide, 70% ethanol and Oosafe® using differential bacterial counts

2) To evaluate safety of the three products on mouse embryonic development

**Method**

**Differential bacterial counts**

Samples were obtained using Difco™ Hycheck™ non-selective agar slides. Replicates were taken from various locations in a student embryology laboratory.

**Mouse embryo testing**

Ethical approval was obtained from Monash Medical Centre Animal Ethics Committee. At the two-cell stage, embryos were exposed to:

1) Direct disinfectant contact

2) Disinfectant residue

3) Media equilibrated in MINC™ incubators recently cleaned by wiping with disinfectant or water (control). Blastocyst formation at 72 h was the experiment end point.

**Sperm toxicity testing**

Semen samples with 90% motility were analysed after a 2h incubation in disinfectant cleaned chambers.

**Results**

<table>
<thead>
<tr>
<th>Treatment group</th>
<th>Average colony growth per Difco™ side (24 h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>13.0 ± 1.0(^a)</td>
</tr>
<tr>
<td>Dry wipe</td>
<td>0.3 ± 0.3(^b)</td>
</tr>
<tr>
<td>6% hydrogen peroxide</td>
<td>0.0(^b)</td>
</tr>
<tr>
<td>Oosafe®</td>
<td>0.0(^b)</td>
</tr>
<tr>
<td>70% ethanol</td>
<td>0.3 ± 0.3(^b)</td>
</tr>
</tbody>
</table>

\(^a\)Different superscripts within the same column indicate significance difference (P<0.05).

**Mouse embryo testing**

- Direct disinfectant contact and residue inhibited embryo growth at the 2-cell stage in all cases.
- 92% of embryos incubated in the Oosafe® cleaned MINC™ reached blastocyst stage, similar to control levels giving 67% blastocyst development (Fig. 1A,C).
- 70% ethanol and 6% hydrogen peroxide cleaning inhibited growth at the 2-cell stage (Fig. 1B,D).

**Significance**

Embryos are extremely sensitive to disinfectant residue and fumes. There is no completely safe option and all IVF laboratories are encouraged to re-evaluate their cleaning protocols.

This study indicates that Oosafe® poses the least risk to embryo development and sperm viability. Ethanol and hydrogen peroxide both had a detrimental effect on embryos and sperm, but could still be used for general laboratory cleaning. A combination of disinfectants might be ideal for effective cleaning protocols.

Great care must be taken so that embryo growth and subsequent pregnancy rates are not affected by essential cleaning protocols.