Changing old trends in IVF laboratory cleaning and disinfection

BACKGROUND

Air quality is crucial for the success of IVF.

70% ethanol is the most commonly used IVF laboratory disinfectant. This is a concern, as alcohol-based disinfectants release VOCs which are directly toxic to embryos.

Hydrogen peroxide is a potential alternative cleaning product, which decomposes to water and oxygen.

Oosafe® is a commercially available product that contains quaternary ammonium compounds and no VOCs.

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 2 h incubation in disinfectant cleaned chambers.

Results

Differential bacterial counts

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

*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).

Sperm toxicity testing (% motility)

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.

Figure 1 72 h incubation in disinfected MINC™. Control (A), 6% hydrogen peroxide (B), Oosafe® (C) and 70% ethanol (D) treatment. Scale bar: 50 μm.