Methods for the Evaluation of Hand Disinfectants

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Medical University of Vienna

Evaluation of Hand Disinfectants (1)
Parameter: Reduction of bioburden

- In vitro
  - MIC of disinfectant vs. selected strains
  - MBC of disinfectant vs. selected strains
  - Quantitative suspension tests for (bacteri-, fungi-, viru-)cidal properties
  - Kill-time studies suspension tests

Evaluation of Hand Disinfectants (2)

- In vivo
  - Controlled laboratory tests simulating practical conditions on hands of volunteers
  - Field trials

Evaluation of Hand Disinfectants (3)
Parameter: Reduction of infections

- Clinical trial
  - Comparative trial with the aim
    ⇒ Novum > Reference
  - Equivalence study

SAMPLE SIZE NECESSARY FOR SIGNIFICANT DIFFERENCE OF PROPORTIONS
(Example for comparative trial)

<table>
<thead>
<tr>
<th>Present Ratio of Hand-Transmitted Nosocomial Infections be</th>
<th>Desired Reduction of Infection Ratio be</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 %</td>
<td>50 %</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>New Ratio of Hand-Transmitted Nosocomial Infections intended be</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 %</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Level of significance (one-sided)</th>
<th>Power of statistical test</th>
</tr>
</thead>
<tbody>
<tr>
<td>α = 5 %</td>
<td>1 - β = 90%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Sample Size (Number patients per experimental arm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2500</td>
</tr>
</tbody>
</table>

Laboratory Test Methods

Hygienic Hand Disinfection:
(Hygienic Hand *Wash* and Hygienic Hand *Rub*)

- In vitro
- In vivo
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Bactericidal Properties of Hand Disinfectants
Proposed Suspension test prEN 12054

In vitro
Test organisms
- Staphylococcus aureus
- Pseudomonas aeruginosa
- Escherichia coli K 12
- Enterococcus hirae
Temperature 20°C
Contact time
- hygienic 1 min (optional 0.5 min)
- surgical 5 min (optional 1.2,3,4 min)
Requirement
- reduction: rub 5.0 lg,
  wash 3.0 lg

Hygienic Handwash with antiseptic soap – EN 1499. Simulating practical Conditions

In vivo
Volunteers 12-15
Test organism Escherichia coli K 12
Recovery Fingertip rub before and after treatment
Application
- product 30 or 60 s, according to manufacturer
- reference 60 s handwash with unmedicated soap
Requirement Product significantly (p = 0.01 unidirectional)
  more efficacious than soap
Discrimination Means ≥ 0.5 lg different (Power: 0.90)

Hygienic Handrub – EN 1500
Simulating practical Conditions

In vivo
Volunteers 12-15
Test organism Escherichia coli K 12
Recovery Fingertip rub before and after treatment
Application
- product 30 or 60 s, according to manufacturer
- reference 2 x 30s (=60 s) handrub with 2x3 ml
  60% (vol) 2-propanol
Requirement Product not significantly (p = 0.1, unidir.)
  less efficacious than 2-propanol 60%, 1min
Discrimination Means ≥ 0.6 lg different (Power: 0.95)

Hygienic Hand Disinfection – ANOVA: non-standardized
Results (lg RFPi): 5 agents, 5 repetitions, 2 laboratories,
with 15 volunteers in each

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>DF</th>
<th>Mean Squares</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Agents (5)</td>
<td>4</td>
<td>102,4</td>
<td>327,5</td>
<td>&lt;0,0001</td>
</tr>
<tr>
<td>Volunteers (15)</td>
<td>14</td>
<td>2,6</td>
<td>8,1</td>
<td>&lt;0,0001</td>
</tr>
<tr>
<td>Laboratories (2)</td>
<td>1</td>
<td>5,8</td>
<td>18,6</td>
<td>&lt;0,0001</td>
</tr>
<tr>
<td>PxV</td>
<td>42</td>
<td>0,7</td>
<td>2,2</td>
<td>&lt;0,0001</td>
</tr>
<tr>
<td>VxL</td>
<td>3</td>
<td>3,4</td>
<td>10,8</td>
<td>&lt;0,0001</td>
</tr>
<tr>
<td>PxL</td>
<td>14</td>
<td>3,1</td>
<td>10,0</td>
<td>&lt;0,0001</td>
</tr>
<tr>
<td>PxVxL</td>
<td>42</td>
<td>0,7</td>
<td>2,4</td>
<td>&lt;0,0001</td>
</tr>
<tr>
<td>Error</td>
<td>470</td>
<td>0,3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

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Hygienic Hand Disinfection – ANOVA:
standardized results (lg RFPi-lg RFRi):
4 [products–reference], 5 repetitions , 2 laboratories,
with 15 volunteers in each

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>DF</th>
<th>Mean SQ</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Products-Reference</td>
<td>3</td>
<td>90,2</td>
<td>117,5</td>
<td>&lt;0,00001</td>
</tr>
<tr>
<td>Volunteers (15)</td>
<td>14</td>
<td>2,7</td>
<td>3,5</td>
<td>&lt;0,00001</td>
</tr>
<tr>
<td>Laboratories (2)</td>
<td>1</td>
<td>3,5</td>
<td>4,6</td>
<td>n.s.</td>
</tr>
<tr>
<td>PxV</td>
<td>42</td>
<td>0,9</td>
<td>1,1</td>
<td>n.s.</td>
</tr>
<tr>
<td>VxL</td>
<td>3</td>
<td>1,6</td>
<td>2,1</td>
<td>n.s.</td>
</tr>
<tr>
<td>PxL</td>
<td>14</td>
<td>1,7</td>
<td>2,1</td>
<td>&lt;0,01</td>
</tr>
<tr>
<td>PxVxL</td>
<td>42</td>
<td>0,9</td>
<td>1,2</td>
<td>n.s.</td>
</tr>
<tr>
<td>Error</td>
<td>466</td>
<td>0,8</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Health-Care Antiseptic Drug Products-FDA (1)
In vitro
- Antibacterial spectrum of
  - active ingredient
  - carrier
- MICs with 20 microbial species, 50 strains of each
  (50% fresh clinical strains)
  - 9 gramnegative spp.
  -10 grampositive spp.
- Candida (incl. C. albicans)
  (= approx. 1000 strains)
- Development of resistance study
- Time-kill study (suspension test) with selected strains for
  0, 3, 6, 9, 12, 15, 20, 30 min

Health-Care Antiseptic Drug Products - FDA (2)
Antiseptic handwash and Health-Care Personnel handwash
(originally ASTM E 1174)

In vivo
- Volunteers (N):
  approx. total of 108
- Test preparation:
  approx. 54
- Positive control:
  approx. 54
- Contaminant bacterium: Serratia marcescens , (E. coli)
- Contamination and application: 10 times on an experimental day
  (Test and control in parallel)
- Samplings:
  - after 1st contamination (baseline)
  - after 1st, 3rd, 7th, 10th wash (rub)
- Required reduction within 5min:
  - after 1st handwash: 2 lg
  - after 10th handwash: 3 lg

Standard Test Method for Determining the Bacteria Eliminating Effectiveness of Hygienic Handwash and Handrub. ASTM E 2276
In vivo
(Similar to “Virus-eliminating“ Test acc. to ASTM E-1838)

Finger Pads including thumbs of at least 2 volunteers
Test bacteria: S. marcescens, E. coli, S. aureus, S.epiderm.
Requirement: not defined, but in comparison to a
negative and positive control

Virucidal Hand Disinfection Tests
Suspension Tests

<table>
<thead>
<tr>
<th>Suspension Tests</th>
<th>In vivo Tests</th>
</tr>
</thead>
<tbody>
<tr>
<td>prEN 14476</td>
<td>Finger pad, Whole hand</td>
</tr>
<tr>
<td>DVV</td>
<td>ASTM E-1838 (acc. to Sattar)</td>
</tr>
<tr>
<td>Adeno (human 4)</td>
<td>Polio 1</td>
</tr>
<tr>
<td>Rota (human) Wa</td>
<td>Adeno</td>
</tr>
<tr>
<td>Rhino (human) 37</td>
<td>Adeno</td>
</tr>
<tr>
<td>Hepatitis A HM-175</td>
<td>Polio 1</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Virucidal Hand Disinfection Test ASTM E-1838</th>
</tr>
</thead>
<tbody>
<tr>
<td>Preparation for the test</td>
</tr>
<tr>
<td>Marking of test areas</td>
</tr>
<tr>
<td>Test area with wet Virus suspension</td>
</tr>
<tr>
<td>Test area with dried Virus suspension</td>
</tr>
<tr>
<td>20-30 minutes</td>
</tr>
</tbody>
</table>

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Laboratory Test Methods

Surgical Hand Disinfection:
(Surgical Hand Wash and Surgical Hand Rub)

In vitro
In vivo

Laboratory Test Methods

Surgical Hand Disinfection:

In vitro

Same Test Method as for
Hygienic Hand Disinfection

Laboratory Test Methods

Surgical Hand Disinfection:

In vivo

Surgical Handrub/wash – prEN 12791
Simulating practical conditions

In vivo
Volunteers (N): 18-20
Test organism: Resident skin flora
Recovery: Fingertip rub before and after treatment
Application of
- Product: acc. to manufacturer, max. 5 min handrub/wash
- Reference: 3 min handrub with 60%/vol 1-propanol (mx3 ml)
Immediate effect sample: immediately after end of treatment (one hand)
3-hours effect sample: 3 hrs after end of treatment (other [gloved] hand)
Requirement: Product not significantly (imm: p=0.1; 3-hrs: 2p =0.01)
less efficacious than reference
Discrimination: imm. Effect: Means > 0.5 lg different (Power: 0.95)
Sustained effect: Optional claim: At 3 hrs, product significantly (p=0.01, unidirectional) more efficacious than reference
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Surgical Hand Disinfection according to EN 12791
4 disinfectants, 5 laboratories, both hands

<table>
<thead>
<tr>
<th>Hands</th>
<th>Agents</th>
<th>Conc.</th>
<th>Mean (lgRFi)</th>
<th>Median (lgRFi)</th>
<th>deviation (lgRFi)</th>
</tr>
</thead>
<tbody>
<tr>
<td>l</td>
<td>n-propanol</td>
<td>60%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>r</td>
<td>n-propanol</td>
<td>60%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>l</td>
<td>ethanol</td>
<td>85%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>r</td>
<td>ethanol</td>
<td>85%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>l</td>
<td>iso-propanol</td>
<td>70%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>r</td>
<td>iso-propanol</td>
<td>70%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>l</td>
<td>CHG</td>
<td>4%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>r</td>
<td>CHG</td>
<td>4%</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
</tbody>
</table>

Surgical Hand Disinfection - ANOVA: non-standardized results (lgRFi):
4 agents, 2 hands, 5 laboratories, 20 volunteers in each

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>DF</th>
<th>F</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Agents (4)</td>
<td>3</td>
<td>53.97</td>
<td>&lt; 0.001 *</td>
</tr>
<tr>
<td>2 Laboratories (5)</td>
<td>4</td>
<td>5.22</td>
<td>&lt; 0.001 *</td>
</tr>
<tr>
<td>3 Hands (2)</td>
<td>1</td>
<td>4.93</td>
<td>0.029</td>
</tr>
<tr>
<td>1 x 2</td>
<td>12</td>
<td>1.67</td>
<td>0.073</td>
</tr>
<tr>
<td>1 x 3</td>
<td>4</td>
<td>1.37</td>
<td>0.249</td>
</tr>
<tr>
<td>2 x 3</td>
<td>3</td>
<td>0.66</td>
<td>0.579</td>
</tr>
<tr>
<td>1 x 2 x 3</td>
<td>12</td>
<td>1.55</td>
<td>0.107</td>
</tr>
</tbody>
</table>

Surgical Hand Disinfection - ANOVA: standardized results (lgRFPi - lgRFri):
3 [products –reference], 5 laboratories, 2 hands, 20 volunteers in each

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>DF</th>
<th>F</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 [Products-Reference] (3)</td>
<td>2</td>
<td>42.16</td>
<td>&lt; 0.001 *</td>
</tr>
<tr>
<td>2 Laboratories (5)</td>
<td>4</td>
<td>1.67</td>
<td>0.162</td>
</tr>
<tr>
<td>3 Hands (2)</td>
<td>1</td>
<td>0.70</td>
<td>0.404</td>
</tr>
<tr>
<td>1 x 2</td>
<td>8</td>
<td>1.66</td>
<td>0.109</td>
</tr>
<tr>
<td>1 x 3</td>
<td>4</td>
<td>3.41</td>
<td>0.012</td>
</tr>
<tr>
<td>2 x 3</td>
<td>2</td>
<td>0.63</td>
<td>0.533</td>
</tr>
<tr>
<td>1 x 2 x 3</td>
<td>8</td>
<td>0.40</td>
<td>0.918</td>
</tr>
</tbody>
</table>

EFFICACY OF SURGICAL HAND DISINFECTION (5 min)

Surgical Hand Disinfection: FDA (orig. ASTM E II5)
Volunteers (N): approx: 100 (150)
per arm:
- Test: approx. 50
- Positive control: approx. 50
- ( Placebo ) ( 50 )

Testbacteria: normal resident handflora
Application of Product:
acc. to manufacturer’s instruction or without any: apply product 2 x 5 min, then rinse hands for 1 min
„Baseline“:
- rinse hands for 30 s, wash hands for 30s,
- rinse hands for 3s
Positive control: FDA-approved antiseptic; all parameters as product; concurrent testing
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Surgical Hand Disinfection: FDA in-vivo model
Schedule for disinfecting and sampling and required lg bacterial reduction

<table>
<thead>
<tr>
<th>Sampling Times (hrs)</th>
<th>Day of test period</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>1/60</td>
<td>x</td>
</tr>
<tr>
<td>3</td>
<td>O</td>
</tr>
<tr>
<td>6</td>
<td>&lt;bl</td>
</tr>
</tbody>
</table>

X: Desinfection: Day 1 (1/60), day 2, 3, 4 (1/60, 3, 6), day 5 (1/60)
O: Sampling: Day 1, 2 and 5: after 1/60, 3 and 6 hrs with gloves
bl: Baseline

Further Information is available!

Other 2005 Teleclasses
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- April 21 - Creutzfeldt-Jakob Disease: Recommendations for Disinfection and Sterilization with Dr. William Rutala
- April 28 - Overcoming the Resistance of Biofilms with Dr. Peter Gilbert
  Sponsored by Virox Technologies Inc. www.virox.com
- May 19 - Antiseptic Practice & Procedure with Susan Crow
  Sponsored by 3M Canada www.3m.ca
- May 26 - Canadian Response to West Nile Virus with Dr. Paul Sockett
- June 7 - Measuring the Cost of Hospital Infection with Dr. Barry Cookson

Questions? Contact Paul Webber paul@webbertraining.com

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