Eradication of *Pseudomonas aeruginosa* Biofilm in Endotracheal Tubes Based on Lock Therapy: Results From an In Vitro Study

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Ventilator-Associated Pneumonia (VAP) accounts for 22% of hospital-acquired infections\textsuperscript{1, 2}.

Up to 27% of intubated patients develop VAP (46% in critical care patients with >48h of MV)\textsuperscript{3}.

Overall VAP-ICU rate in HGUGM 2016: 7 episodes/1,000 days of MV.

Clinical impact
- Attributable mortality rate: 13\%\textsuperscript{4}.
- Extra days of MV: 7-11\textsuperscript{1}.
- Extra days of hospitalization: 11-13\textsuperscript{1}.
- Associated costs: 11,000€ (Spain)-40,000$ (USA)\textsuperscript{5, 6}.

\textsuperscript{1} Kalil et al. Clin Infect Dis 2016.
\textsuperscript{3} Hortal J et al. Crit Care 2009.
\textsuperscript{4} Ralsen WG et al. Lancet Infect Dis 2013.
\textsuperscript{6} Kollef MH et al. Infect Control Hosp Epidemiol 2012.
BACKGROUND
Bacterial biofilm plays an important role in the pathogenesis of VAP\textsuperscript{1,2,3,4}.

Advanced ETT biofilm stage is related to VAP (not days of MV)\textsuperscript{5,6,7,8}.
**PREVENTIVE MEASURES in HGUGM**

- Continuous training
- Hand hygiene
- Subglottic aspiration (100 mm Hg) \(^1,2,3\)
- Continuous cuff pressure monitoring (25 cm H\(_2\)O)
- Position (30-45°)
- *S. aureus* decolonization
- Selective digestive decontamination (SDD)

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Catheter-related bloodstream infections

SDD subglottic space
To apply an in vitro bench-top model to assess the efficacy of SDD solution applied in the subglottic space for eradication of *P. aeruginosa* biofilm in ETTs.

**HYPOTHESIS**

The application of SDD solution as “lock therapy” in the subglottic space can reduce *P. aeruginosa* biofilm in ETTs.
MATERIAL AND METHODS

72-h mature *P. aeruginosa* biofilm

2-hour application of SDD or saline (+C)

Cuffed ETT (TapeGuard Oral Tracheal Tube Evac Murphy Eye, Mallinckrodt™)

3 ml $10^8$ cfu/ml

composition of SDD solution
nystatin 2.6 mU, tobramycin 15.6 mg/ml, colimycin 13 mg/ml

Single-dose 5-day

ALT

SLT

Sonication of ETT segment

No. live/dead cells by CLSM

Visualization of ETT segments by CLSM and SEM

*SDD solution composition*

Cuffing ETT (TapeGuard Oral Tracheal Tube Evac Murphy Eye, Mallinckrodt™)

*ATCC 15442*

Biofilm production was previously assessed using the crystal violet assay

All experiments were performed in triplicates.
## Sonication of ETT Segment

### RESULTS

<table>
<thead>
<tr>
<th>Variable</th>
<th>Lock Therapy</th>
<th>5-day</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Single-dose</td>
<td>5-day</td>
</tr>
<tr>
<td></td>
<td>ALT</td>
<td>SLT</td>
</tr>
<tr>
<td>Median (IQR) cfu counts/ml</td>
<td>3.12x10⁵ (9.7x10⁴-0)</td>
<td>8.16x10⁷ (7x10⁷-0)</td>
</tr>
<tr>
<td>Median (IQR) % live cells</td>
<td>53.2 (50.9-57.2)</td>
<td>91.5 (87.3-93.9)</td>
</tr>
</tbody>
</table>
RESULTS

VISUALIZATION OF BIOMASS

VISUALIZATION OF SESSILE CELL STRUCTURE

ALT

SLT

Single-dose

5-day

Single-dose

5-day
CONCLUSIONS

- Preliminary results: ALT with SDD solution significantly reduced 72-h biofilm of *P. aeruginosa* based on culture and microscopy

- Efficacy of ALT against biofilm of other microorganisms (prevention and therapy)

- Future studies: evaluate the efficacy of SDD solution as a prophylactic measure for VAP in patients under mechanical ventilation

- Our bench-top model = practical for simulation of colonization of ETT (diagnosis and therapy)
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