Correlation between biofilm removal and bacterial killing in a model mimicking disinfection of contaminated endoscopes

Wafi Siala¹,², Martine Weickmans¹, Guy Heynen¹, Françoise Van Bambeke², Thomas Vanzieleghem¹

¹ OneLIFE SA, Louvain-la-Neuve (Belgium), ² Louvain Drug Research Institute, UCL, Brussels, Belgium.

Introduction

Growth of biofilms inside endoscope channels can result in failure of the endoscope reprocessing. Contaminated endoscopes can be the cause of device-related nosocomial outbreaks. Therefore, it is essential to clean and disinfect them effectively between patients to avoid this risk. The objective of this study was to examine the impact of biofilm removal in the cleaning phase on the levels of bacterial eradication achieved by high-level disinfection in biofilm models that mimic the accumulation of bacteria within endoscopes.

Materials and Methods

Biofilms were grown in a model mimicking an endoscope environment: the Buildup Biofilm model (BBF) as described by Da Costa and colleagues in 2016. Briefly, biofilm was developed in MBEC 96-well plate (Innovotech, Canada). Bacteria were suspended in Artificial Test Soil (ATS US patent 6447990) to achieve 10⁸ CFU/ml. Biofilm was formed at room temperature in ATS, with rocking action, on plastic pegs over eight days. Four rounds of high level disinfection (HLD) using 2.6% glutaraldehyde were included. In this study, one reference strain and one clinical isolate of two clinically relevant species: *P. aeruginosa* and *K. pneumoniae* were used.

Biofilms were then treated with four commercially available detergents intended for endoscope cleaning (recommended dosage, 60 min, 40°C, absence of friction). Optionally, after the treatment with cleaners, biofilms were exposed to peracetic acid (900 ppm, 3 min, 40°C). Control biofilms were exposed to water at 40°C for 1 hour.

Remaining biofilm biomass was assessed using crystal violet assay as previously described (1) and data were expressed as the percentage reduction in biomass compared to control. Bacterial viability was quantified by CFU counting and converted to log₁₀ CFU/cm² as previously described (2).

<table>
<thead>
<tr>
<th>Biofilm biomass removal (%)</th>
<th>Viable counts (log₁₀ CFU/cm²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cleaner - / PAA -</td>
<td>Cleaner + / PAA -</td>
</tr>
<tr>
<td>Cleaner + / PAA -</td>
<td>Cleaner + / PAA +</td>
</tr>
</tbody>
</table>

**Figure 1** – (a) Biofilm biomass removal (%) and (b) viable counts (log₁₀ CFU/cm²) observed in control BBFs (Cleaner - / PAA -) and in BBF that were exposed to cleaners only (Cleaner + / PAA -) or to cleaners followed by PAA treatment (Cleaner + / PAA +).

**Figure 2** – Correlation plots for bacterial counts and remaining BBF biomass observed in biofilms exposed to cleaners followed by PAA treatment.

Conclusions

Strong correlation between biofilm removal and CFU reduction observed in the BBF models suggest that treatment with a potent biofilm-disruptive cleaner before disinfection is key to achieve successful decontamination of biofilm-colonized endoscopes.