Electronic Supporting Information (ESI)

Rapid antibacterial activity of anodized aluminum-based materials impregnated with quaternary ammonium compounds for high-touch surfaces to limit transmission of pathogenic bacteria

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Fig. S1: Manufacturing of A3S materials:
(1) Anodization step: electrolytic process performed in an acid bath allows the formation of nanoporous anodic aluminium oxide, with controlled depth and diameter, on the surface of aluminum sample.
(2) Impregnation step: antibacterial solution (AgNO₃ and/or QACs) is incorporated into the nanopores, followed by a sealing process as final step.

Table S1: Characteristics of bacterial strains used in the present study

<table>
<thead>
<tr>
<th>Name</th>
<th># Origin</th>
<th>Gram</th>
<th>Morphology</th>
<th>Cellular dimension (average)</th>
<th>E-test (resistance)</th>
<th>Gene</th>
<th>Culture Medium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staphylococcus aureus</td>
<td>ATCC # 29213</td>
<td>Gram-</td>
<td>Cocci (grape-like clusters)</td>
<td>0.5-1.5 µm diameter</td>
<td>-</td>
<td>-</td>
<td>Mueller Hinton Agar/Broth</td>
</tr>
<tr>
<td>Clostridioides difficile</td>
<td>Epidemic strains R20291</td>
<td>Gram-</td>
<td>Bacilli</td>
<td>1.0 µm wide by 3.0-4.0 µm long</td>
<td>-</td>
<td>-</td>
<td>Tryptone Yeast extract (TY) Agar/Broth</td>
</tr>
<tr>
<td>Enterococcus faecium</td>
<td>Clinical from CHUS #422</td>
<td>Gram-</td>
<td>Cocci</td>
<td>0.6-2.5 µm diameter</td>
<td>Vancomycin: &gt; 256 µg/ml</td>
<td>VanA</td>
<td>Brain Heart Infusion (BHI) Agar/Broth</td>
</tr>
<tr>
<td>Enterococcus faecalis</td>
<td>Clinical from CHUS #55</td>
<td>Gram-</td>
<td>Cocci</td>
<td>0.6-2.5 µm diameter</td>
<td>Vancomycin: 8 µg/ml</td>
<td>VanB</td>
<td>BHI Agar/Broth</td>
</tr>
<tr>
<td>Klebsiella pneumoniae</td>
<td>Clinical from CHUS #455814871</td>
<td>Gram-</td>
<td>Capsulated bacilli</td>
<td>0.3-0.8 µm wide by 1.0-3.0 µm long</td>
<td>-</td>
<td>-</td>
<td>Mueller Hinton Agar/Broth</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>ATCC # 29532</td>
<td>Gram-</td>
<td>Bacilli</td>
<td>1.0 µm wide by 1.0-2.0 µm long</td>
<td>-</td>
<td>-</td>
<td>Luria-Bertani (LB) Agar/Broth</td>
</tr>
<tr>
<td>Salmonella typhimurium</td>
<td>SL1344</td>
<td>Gram-</td>
<td>Bacilli</td>
<td>0.7-1.5 µm wide by 2.2-5.0 µm long</td>
<td>Streptomycin: 100 µg/ml</td>
<td>-</td>
<td>LB Agar/Broth + 100 µg/ml Streptomycin</td>
</tr>
</tbody>
</table>
Fig. S2: Standard curve representing the absorbance at a wavelength (λ) of 215 nm in function of the concentration of QACs contained in the stock solution used for material impregnation. The coefficient of determination $R^2 = 0.9948$, of the standard curve $y = 0.0117x + 0.012$, demonstrates the good reliability of this linear regression model. Results are means ± SEM (n=5; N=15).

Fig. S3: Sporicidal effect of impregnation solutions (QACs and AgNO$_3$+QACs) on *C. difficile* spores. From an initial inoculum of $\approx 1.6 \times 10^5$ spores/mL, the number of surviving spores was determined following a contact of 1, 4 and 24 h with impregnation solutions (QACs and AgNO$_3$+QACs) and sporicidal control (1/16 (v/v): Virox$^\text{TM}$5/water). Results are means ± SEM (n=4; N=16). Significant effect compared to sporicidal control: *p<0.001; NA: no spore counted.
Fig. S4: Swab liquid-inoculation assay evaluating the antibacterial activity of different materials on S. aureus, C. difficile and streptomycin-resistant S. typhimurium. The reference of 99.9% antibacterial activity is indicated by the dotted red line. Results are means ± SEM (n=2; N=8). Statistical effect compared to copper: * p< 0.001; NA: no bacteria counted.
**Fig. S5:** Humid-transfer inoculation assay from contaminated gel evaluating the antibacterial activity of materials. The graph shows total counts of bacteria that survived (A) on the gel or (B) after transfer from the gel to the different materials. The reference of 99.9% antibacterial activity is indicated by the dotted red line. Results are means ± SEM (n=2; N=8). Statistical significance when compared to copper: *p<0.001; NA: no bacteria counted.
Fig. S6: Dry-transfer inoculation assay evaluating the antibacterial activity of materials after contact with a contaminated filter under low humidity conditions. Counts in log_{10} CFU/surface indicate the number of bacteria that survived on (A) the contaminated filters and (B) the materials, after transfer from the filter. Results are means ± SEM (n=2; N=8). Statistical effect compared to AA: *p<0.001 and to copper: #p<0.001.
Fig. S7: Swab liquid-inoculation assay evaluating the sporicidal activity of the materials on *C. difficile* spores following a contact of 1 and 24 h. Results are means ± SEM (n=3; N=12). No statistical effect was observed.