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Isolation and Characterization of Clostridioides difficile spores from contaminated “single-use” surgical gowns.

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Introduction

Clostridium difficile is a Gram-positive, spore-forming anaerobe that comprise either toxigenic or non-toxigenic strains. Toxigenic C. difficile usually possess Toxin A (TcdA) and Toxin B (TcdB); although some strains can be variant [1]. C. difficile can exist in vegetative bacterial form or as metabolically dormant, highly disinfectant resistant spores.

Sporos can attach to clinical surfaces for months via structures such as the exosporium and are implicated in organism transmission [2]. C. difficile infection (CDI) is the leading cause of antibiotic-and healthcare-associated diarrhoea globally [3]. One reason for high incidence rates is due to the adherence of spores to surgical gowns which can ‘trap’ the spores and transfer them to stainless steel surfaces and hospital floor vinyl [4].

Aim of Study

To determine whether C. difficile can be isolated from “used” hospital gowns. Any presumptive C. difficile will also be identified.

<table>
<thead>
<tr>
<th>Suspected C. difficile number</th>
<th>NO2 growth (CCFA)</th>
<th>Growth on CCFA</th>
<th>Odour</th>
<th>Chartreuse under UV</th>
<th>Gram Stain</th>
<th>Produces spores</th>
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Table 1: Phenotypic identification of the suspected C. difficile colonies extracted from contaminated gowns.

Figure 3: Life cycle of C. difficile in the gut. (A) toxigenic C. difficile may be present in healthy human microbiota. (B) dysbiosis occurs (e.g. due to antibiotics) which allows for colonisation of C. difficile. (C) C. difficile toxins lead to inflammation of the distal gut and thus C. difficile mediated disease [5]

Results

• 23 colonies were isolated from the gowns. After phenotypic analysis (Table 1) only 8 isolates showed characteristic C. difficile growth. CDIFF QUIK CHEK COMPLETE® (TECHLAB) analysis confirmed these results (Figure ).
• After PCR analysis via 16S-23rRNA inter-spacer region [6] and toxin A and toxin B PCR [7], this was reduced to 5 final samples which are presumed to be C. difficile.
• These samples are currently undergoing final confirmatory testing at the National Anaerobic Reference unit, Cardiff, UK.

Conclusions and Future Work

• It was concluded that the surgical gowns from Rideout Hospital, USA, were contaminated with C. difficile.
• Future work will include conformational testing of all presumptive C. difficile strains at the National Anaerobic reference Unit, Cardiff, UK.
• Implications include gowns acting as fomites and the need to dispose of gowns immediately after use to prevent spore transfer
• Biocide testing will establish if current infection control measures and disinfectants (sporicides) are working.
• This work aims to limit the prevalence of spores and spread of infection in hospitals.

References