

O140

Oral Session

Enterococcal resistance

MATURE BIOFILMS OF ENTEROCOCCUS FAECALIS AND ENTEROCOCCUS FAECIUM ARE HIGHLY RESISTANT TO ANTIBIOTICS

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Objectives:

E. faecalis and *E. faecium* are among the leading pathogens isolated from nosocomial infections and are increasingly recognized as important pathogens in infections of medical devices such as prosthetic joints. Bacterial biofilm formation is central in the pathogenesis of infections related to foreign material, and *E. faecalis* and *E. faecium* can form biofilm. There is compelling evidence that bacteria within biofilms are more tolerant to antibiotics than are planktonic cells. Treatment of *E. faecalis* and *E. faecium* infections are complicated both by the antimicrobial resistance of the bacterium and by the ability of the bacterium to form biofilm. In this study we explore the antibiotic sensitivity of bacteria in young and mature biofilms

Methods:

E. faecalis and *E. faecium* isolates were from infections of prosthetic hip and knee joints. Biofilm formation was measured in a microtiter plate-based assay. The young and mature biofilm was analyzed in scanning electron microscopy (SEM). Minimal biofilm eradication concentration (MBEC) for vancomycin, linezolid, ciprofloxacin, daptomycin, tigecycline, and rifampicin alone and in combinations was determined for biofilms of different ages. Enterococcal biofilms of different ages on beads of bone cement were treated with the same antibiotics for 24 hours followed by plating to quantify surviving bacteria.

Results:

The mature biofilms showed higher OD than young biofilms in the microtiter-plate assay. The surfaces of the beads were visualized by SEM, and an amorphous biofilm was seen embedding the bacteria. In 24 hours-old biofilm, *E. faecalis* and *E. faecium* isolates displayed MBECs for the various antibiotics in the 8mg/l - 512 mg/l range. When combined with 4 mg/l rifampicin, MBEC dropped by two to four dilutions. In biofilms formed on cement beads during 24 hours significant bacterial killing was detected (> 2 log decrease in cfu) after 24 hrs exposure to antibiotics.

All *E. faecalis* and *E. faecium* isolates displayed MBECs between 128-(>1024) for the different antibiotics in 120 hours-old biofilm. When combined with 4 mg/l rifampicin, MBEC dropped 1 dilution or not at all. In biofilms grown on cement beads for 120 hours no significant bacterial killing was detected (0.2- 1.2 log decrease in cfu) after 24 hrs exposure to antibiotics at physiological concentrations. When combining ciprofloxacin, rifampicin, tigecycline, and daptomycin in an attempt to eradicate *E. faecalis* or *E. faecium* biofilms on cement beads grown for 120 hours, a 1.2-1.8 log reduction in cfu was detected.

Conclusions:

This study indicates that the age of biofilms profoundly affects the antibiotic susceptibility of enterococci in biofilms. Here the mature biofilms of *E. faecalis* and *E. faecium* were highly resistant to all antibiotics used. The duration of a device related infection might be an important factor to take in consideration for prediction of treatment-outcome in the clinical setting.