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P0161 Performance of a new EUCAST disk diffusion method using fastidious anaerobe agar (FAA) for clinical Bacteroides fragilis group isolates

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Background: Resistance in anaerobic bacteria is increasing. In the *Bacteroides* genus of bacteria this increase is most problematic, especially considering the limited number of antibiotics available for treatment of infections with anaerobic bacteria. The emergence of resistance to even the broadest-spectrum antibiotics makes the development of a reliable and easy to use susceptibility testing method imperative. We have tested the Fastidious Anaerobe Agar (FAA) and found it to be a suitable medium for disk diffusion of rapidly growing anaerobic bacteria. The aim of this study was to further investigate this medium for clinical isolates of the *Bacteroides fragilis* group.

Materials/methods: Disk diffusion testing was performed on 175 clinical isolates of the *Bacteroides fragilis* group (*Bacteroides fragilis* = 116 isolates, *Bacteroides thetaiotaomicron* = 25 isolates) with a McFarland 1.0 (0.9-1.1) inoculum suspension on in-house produced FAA plates (LabM) supplemented with 5% defibrinated horse blood. Zone diameters and quality of growth were assessed after 16-20 hours of anaerobic incubation at 37°C. The obvious zone (the zone of clearest inhibition of bacterial growth) was measured for clindamycin (10 µg), metronidazole (5 µg), meropenem (10 µg) and piperacillin-tazobactam (30-6 µg). MIC determination with agar dilution on brucella blood agar was used as reference.

Results: All 175 isolates showed confluent growth after 16-20 hours of incubation. Zone diameters were easily readable for most antibiotics, except sometimes for beta-lactam antibiotics (especially for *Bacteroides thetaiotaomicron).* Reading the most obvious zone edge correlated well with the reference MICs for metronidazole (Figure 1A). For piperacillin-tazobactam and meropenem (Figure 1 B) there was an area of poorer separation below the fully susceptible population. Reading of clindamycin zones after 16-20 hours of incubation resulted in false susceptible results, but all resistant isolates were detected after prolonged incubation (48 hours).

Conclusions: Disk diffusion on FAA is a feasible method for susceptibility testing of the *Bacteroides fragilis* group with sufficient distinction between resistant and susceptible isolates for metronidazole, meropenem and piperacillin-tazobactam already after 16-20 h whereas clindamycin, at least for some species, seemed to require longer incubation for all resistance to express itself.

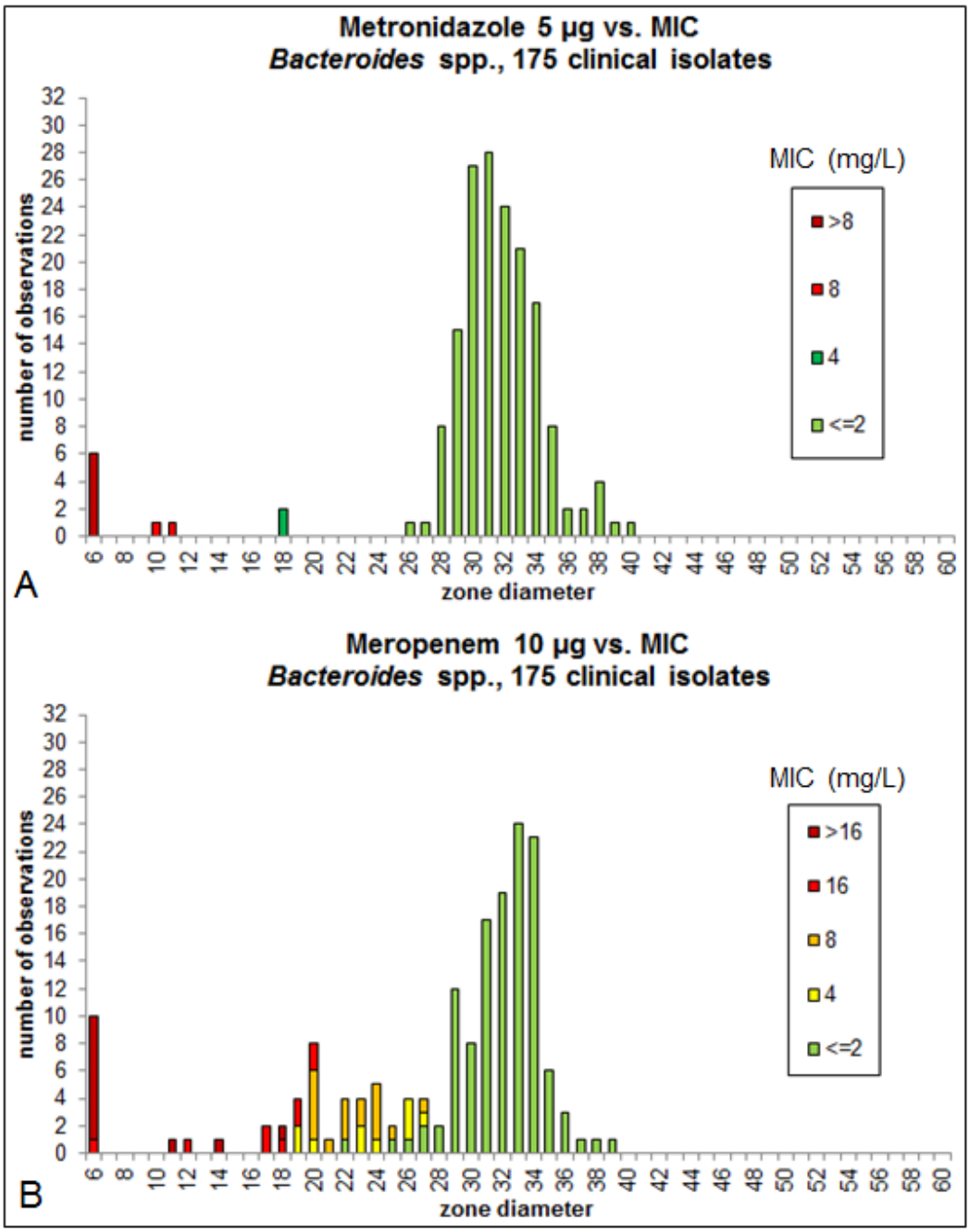

Figure 1. Inhibition zone diameter distributions for *Bacteroides fragilis* group isolates (n=175) with corresponding MIC values as coloured bars for A) metronidazole 5 µg and B) meropenem 10 µg.