OP 9

Impact of routine laboratory culture media on *in-vitro* biofilm formation of *Pseudomonas aeruginosa, Staphylococcus aureus* and *Enterococcus faecalis*

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Objectives: This study was aimed to determine the efficacy of four routine laboratory culture media on biofilm formation of *Pseudomonas aeruginosa, Staphylococcus aureus* and *Enterococcus feacalis*.

Methods: A sterile flat bottom 96 well plate was inoculated using 0.5 McFarland equivalent standard cell suspension of *P. aeruginosa, S. aureus* and *E. feacalis* and the growth rate of planktonic cells was quantified by measuring the optical density (OD492) at two hour intervals. Influence of culture medium on adhesion of bacteria as an initial step of biofilm formation in the presence of four culture media (Nutrient broth (NB), Brain Heart Infusion (BHI) broth, Luria-Bertani (LB) broth and RPMI 1640) was quantified using MTT (3-[4, 5- dimethylthiazole-2-yl]-2, 5- diphenyltetrazolium bromide) assay after 90 minutes adhesion. Biofilms of *P. aeruginosa, S. aureus, E. feacalis* and their 1:1 mixed biofilms were developed and the growth was quantified using MTT metabolic activity at 24 hour time intervals. Scanning electron microscopy (SEM) was performed to assess the ultrastructure.

Results: On comparing the relative growth of the bacteria in different culture media, the maximum growth of all three planktonic cultures was achieved using BHI broth. All mono species and mixed species cultures exhibited their maximum adhesion in the presence of RPMI 1640. All biofilm exhibited the maximum growth in BHI broth. SEM imaging had shown the enhanced growth of ultrastructure of the biofilm with the presence of BHI broth.

Conclusions: The maximum planktonic and biofilm growth was achieved with BHI broth. However, bacterial adhesion was enhanced in the presence of RPMI 1640.