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Anti-biofilm effect of colistin and ciprofloxacin alone and in combination with various QSIs on mono and polymicrobial biofilms of P.aeruginosa

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Background: *Pseudomonas aeruginosa is* one of the major pathogen related with the biofilm infections and significant problem in hospital infections. Recent studies have indicated that many clinically relevant biofilms are polymicrobial Quorum sensing is an alternative strategy in fighthing biofilm infections and some natural quorum sensing inhibitors (QSI) were found successful to interfere with the infections.

Material/methods: Polymicrobial biofilms were reproducibly grown, consisting of *S. aureus, E.faecium, P. aeruginosa and C. albicans* in a 96-well microtiter plate. We determined the antibiofilm effects of ciprofloksasin and colistin alone or in combinations with some natural QSIs on sessile cells of susceptible *P.aeruginosa* and carbapenem resistant *P.aeruginosa* isolates respectively in both mono and multispecies biofilm model in order to gain more insight into the role of biofilm composition on efficacy of the agents. Ciprofloksasin and colistin combinations of QSIs including cinnamaldehyde, resveratrol, L-canavanin, 4-nitropyridine N-oxide, p-benzoquinon, farnesol, epigallocatechin gallate, catechin hydrate, curcumin, baicain hydrate and esculin hydrate and combinations with cyclic di-GMP inhibitors such as sulfatiyazol and azathioprine were tested with MBEC assay and MBIC, MBEC and log reduction of the antimicrobials were determined.

Results: Ciprofloxasin significantly decreased the biofilm cells of *P.aeruginosa in* mono biofilms in the concentration range of $0.0625$to 4 µg/ml. All of the combinations with ciprofloxasin and QSIs except sulfatiyazol and epigallocatechin killed completely the biofilm cells in particular concentrations tested. Sessile cells of the susceptible *P.*aeruginosa isolates in polymicrobial biyofilm were found fewer susceptible to the combinations of ciprofloxasin with all QSIs except for esculin hydrate and cinnamaldehyde compared to the cells in monomicrobial biofilms. In monospecies biofilm, colistin concentrations in range of 0.0625 to 8 µg/ml didn't show any significant decreases in the biofilm cells of *P.aeruginosa* however, combinations of curcumin, 4-nitropyridine N-oxit, p-benzokinon, azatiyoprine and farnesol with colistin significantly decreased the biofilm cell counts. The study also showed that

P.aeruginosa biofilm cells in polymicrobial biofilm were more susceptible by comparison with those in monomicrobial biofilm.

Conclusions: Except for sulfatiyazol and epigallocatechin, all QSIs increased the effect of ciprofloxasin as an antibiofilm agent. The drug susceptible isolate of *P.*aeruginosa in polymicrobial biyofilm were found fewer susceptible wheras, carbapenem resistant isolates of P.aeruginosa were more susceptible to the combinations of ciprofloxasin with the QSIs when compared to the cells in monomicrobial biofilms.