Title: Study of the effect of Allicin (an active component of garlic) Sub-MIC Concentrations on Biofilm Formation by Proteus mirabilis

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Abstract: **Background & Objective:** *P. mirabilis* is one of the common agents involved in urinary tract infections, especially in patients undergoing urinary catheterization. *P. mirabilis* is capable of living as biofilm and/or planktonic forms. Biofilm plays a significant role in *P. mirabilis* infections. Cells in the biofilm show higher degree of resistance to antimicrobial therapy and host immune responses compared with planktonic cells. So inhibition of biofilm formation could help the body's immune system to combat the bacteria and improve the clinical outcomes for antimicrobial therapy. This study was aimed to investigate the inhibitory effect of allicin on biofilm formation by *P. mirabilis*.

**Material & Methods:** This study was carried out using a *P. mirabilis* ATCC12453. Allicin was purified using semi preparative HPLC procedure. MIC of allicin was determined by microdilution method using serial dilutions of aqueous allicin solution (4-512 µg/ml) in Mueller-Hinton broth. Biofilm inhibition was assayed using Microtiter plate method in the presence of sub-MIC concentrations (4µg-32µg) of allicin. The plates were incubated for 18 hours at 37 ºC. Bacterial biofilms were stained with 0.2% safranin. Dye was solubilized using alcohol-Aceton as solvent and the optical density (OD) was measured at 492 nm wavelength. The extent of biofilm formation was determined (OD of sample well/OD of control well*100). Each assay was performed in triplicate and repeated two times.

**Results:** The allicin MIC was 64 µg/ml for *P. mirabilis* ATCC12453. The results indicated that allicin at concentrations of 16 and 32µg/ml significantly diminished biofilm formation (P<0.01). This concentration did not have significant influence on bacterial growth rate.

**Conclusion:** The results showed that allicin can inhibit the biofilm formation by *P. mirabilis*.