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Determination of antimicrobial effect of protamine by transmission electron microscope and SDS PAGE on *Pseudomonas aeruginosa* isolates from diabetic foot infection

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Diabetic foot infection is one of the major complications of diabetes causing high morbidity and mortality and often leading to lower limb amputations. 285 pus samples from diabetic patients having foot infections were collected from different hospitals of Karachi and Capital Health hospital, Halifax, Canada. Clinical history of each patient was recorded. Bacterial isolates were cultured on appropriate media; identification was done by morphology, cultural and biochemical tests. Effect of protamine against multi drug resistant strains of *P. aeruginosa* was checked by Minimum inhibitory concentration in 96 well micro titer plates. These bacteria were grown in bactericidal concentration of protamine to isolate mutants. Effect of Protamine on protein expression was checked by SDS- PAGE and ultra structural morphological changes due to its effects were observed by Transmission Electron microscopy. Results indicate prevalence of infection as 92 % in diabetic patients with foot ulcers. Major bacterial isolates were *Staphylococcus aureus* (23%), *P. aeruginosa* (20%), *Klebsiella spp* (13%), *Proteus mirabilis, E.coli* (12%) *and Serratia marscenses* (10%). These isolates were highly resistant to different antibiotics. MIC value of protamine was 500µg/ml against P. aeruginosa. SDS-PAGE analysis revealed that Protamine can suppress various virulence proteins expression and Electron micrographs indicated condensation of cytoplasm and accumulation of protamine in cytoplasm without damage to cell membrane. *P. aeruginosa* and *S. aureus* were major isolates expressing multi drug resistance. Transmission electron microscopy results revealed disruption of cellular activities due to condensation of cytoplasm.

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