Abstract

Biofilm formation in *Pseudomonas aeruginosa* (*P. aeruginosa*) is controlled by about 1% of its chromosomal genes; from which four genes were selected for prospective work. The aim of this study was to determine the biofilm formation in *P. aeruginosa* clinical isolates and to evaluate the role of the selected genes in biofilm formation. A total of fifty isolates were recovered from different clinical samples isolated from Egypt by isolation on cetrimide agar media. These isolates were then identified by gram stain, culture characters and biochemical reactions. All the fifty isolates were identified as *P. aeruginosa*. The antimicrobial resistance pattern of the planktonic cells of all isolates was then determined. The antibiogram of isolates showed that amikacin was the most potent antibiotic against all isolates followed by ceftazidime and imipenem. Biofilm quantification (matrix, live and dead cells) of isolates was done by the microtiter plate method using crystal violet (CV) assay. According to the optical density (OD) readings, isolates were classified into the following categories: strong, moderate or weak biofilm producers. Result of CV assay showed that 42 isolates (84%) were strong biofilm producers; 4 isolates (8%) were moderate biofilm producers and the last 4 isolates (8%) were weak biofilm producers. Quantification of the metabolic activity of cells within biofilms (live cells only) was done by the microtiter plate method using XTT reduction assay. Then data analysis and comparison between results of both CV assay and XTT reduction assay was done. Data analysis revealed that there was a significant difference between the results of both assays. Finally, studying the effect of four selected genes (*RhlI, PilA, PilT, PelB*) on biofilm formation in 13 isolates (9 strong biofilm producer isolates and 4 weak biofilm producer isolates) was done using PCR technique. Screening of these genes revealed their presence in all screened isolates including both strong and weak biofilm producer isolates. These final results made us concluded the importance of these genes in biofilm formation and concluded the presence of other factors which may contribute in determining the degree of biofilm formation in *P. aeruginosa*.

***Key words: P. aeruginosa*; Biofilm; Crystal violet; XTT; Quorum sensing; Twitching motility; Exopolysaccharide; Biofilm genes**