

The Effect of Some Enzymes on the Formation of Biofilm from Bacterial *Pseudomonas aeruginosa* Isolated from Pathological Samples

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ABSTRACT

This study of effect of the lipase enzyme on the production of biofilm from the *Pseudomonas aeruginosa* bacteria. This bacterium was isolated from various pathological samples where 30 samples were obtained, included samples isolated from urine, burns, and wounds were the most samples that gave a large number of isolates wound samples 15 samples With an isolation rate of 50%, burns followed by 10 isolates, with an isolation rate of 33.3%, 5 isolates from urine samples of with an isolation rate of 16.6%. The effect of the lipase enzyme on the production of biofilm using it There are three concentrations of the enzyme 40, 80 and 160%. The results showed

that there were significant differences in the effect of the enzyme lipase on the production of biofilm. The concentration of 160% was better than the rest of the effect in its effect.

Keywords: biofilm, lipase, *P.aeruginosa*

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INTRODUCTION

Pseudomonas aeruginosa is an environmental bacterium and the opportunistic pathogen, which performs a dominant function as the causative agent of acute and chronic, generally biofilm-associated, infections. (1,2) Bacterial biofilms are quite recalcitrant to antibiotic treatment plans due to more than one tolerance mechanisms. The involvement of *Pseudomonas aeruginosa* in a huge range of biofilm-related infections regularly leads to remedy failures. Indeed, few contemporary antimicrobial molecules are nonetheless splendid illiberal sessile cells. (3,4) The biofilm surfaces that may be live or inanimate by means of material comprehensively and generically blinded "unrestricted adhesion Microbial blindness.(5,6) Complex organic polymers excrete the microbial cells psychologically outward The material forms the basis on which blindness is based on the formation of an ecosystem Functionally effective and independent. It is also known as An Blindness The endogenous organism that results from adhesion, grounding, surface secretion and secretion of substances.(7,8) The process of formation and progression of a mature biofilm is multi-stage and Wii The number of variants depends on the type of SMU nature Surface to blind microbe, environmental factors and gene expression.(9,10) Within the biological membrane is increasing The ability of cells to efficiently exchange antibiotic-resistant plasmids Through the pairing process and assist in that spatial proximity is more than It occurs between cells and is stuck For the genomics responsible for creating the biofilm(11,12)

MATERIAL AND METHODS

Collection of bacterial samples

30 scientific samples had been gathered from Ramadi Teaching Hospital which had been from a quantity of sources along with burns, wounds, UTI infections, These samples had been amassed through sterile cotton swabs. While urinary tract infections two (UTI) samples were gathered through way of a sterile container.

Bacterial isolation and identification

Bacterial isolates have been subjected to a variety of cultural and biochemical tests for identification of these isolates(13)

Methods for investigating the viability of isolated bacteria upon formation Biofilm

Loopful transplantation from the bacteria under study in ml of medium Glass tubes and then incubated at C°37 for 82 hours After the lap time has expired, the medium was removed from the tubes and grafted several times broth soy Trypticase containing 0% glucose sugar in PBS buffer ((pH = 7.3) Phosphate buffer Saline) He previously attended and left To dry completely at room temperature and then stained with Crystal% 1.0 violet. The excess dye was drained, drenched with distilled water and left(14)

The effect of enzymes on the formation of biofilm

The culture medium containing the active isolation was added by 200 µl with five replicates, and the non-containing culture medium was added to the isolation as a control incubated the dishes at 37 ° C for 24 hours after washing with distilled water, then the enzymes were added to three concentrations 40,80,160 µg / ml with five replications for each Concentrate the incubation dishes for an hour after which were proven by adding 200 µl for

15 minutes. The methanol poured the solution and washed with distilled water. Then I added a tinted crystal

violet. Then I washed it with distilled water. Then I added the acetic acid and read it with the Elisa device⁽¹⁵⁾

RESULT AND DISCUSSION

Table 1: Percentages of *Pseudomonas aeruginosa* isolated from pathological samples

Source of sample	Number	percentage
Burn	10	33.3%
Urine	5	16.6%
Wound	15	50 %
Total	30	100%

Table (1) shows the number of isolates and their percentages isolated from pathological samples, where the number of isolates isolated from wounds was 15 out of a total of 30 isolates with a rate of 50%, ten isolates from

burns with a percentage of 33.3% and five isolates from yields with a percentage of 16.6 %

Table 2: Initial screening to know efficient isolation with biofilm production

No. isolates	Biofilm formation degree			
	Strong	Moderate	Weak	No formation
Isolate No. 1	+			
Isolate No. 2			+	
Isolate No. 3			+	
Isolate No. 4		+	+	
Isolate No. 5	+			
Isolate No. 6	+			
Isolate No. 7				+
Isolate No. 8			+	
Isolate No. 9			+	
Isolate No. 10	+		+	

Table (2) shows the initial screening of the most efficient isolation in the production of biofilm. Isolation No. 1, 5, 10 gave the highest production of the biofilm measured by tube method, while the isolates 2,3,9,8 were poor

production for the biofilm and the number 4 was produced in the middle of the isolation 7 did not give any biofilm production.

Table 3: The effect of the lipase enzyme on the production of biofilm from *P.aeruginosa*

Concentration	production of biofilm ug / ml				
	F(1)	F(2)	F(3)	F(4)	F(5)
40 % of lipase	0.77	0.78	0.72	0.73	0.73
80 % of lipase	0.53	0.52	0.53	0.56	0.55
160 % of lipase	0.44	0.43	0.45	0.46	0.43
Control	1.8	1.5	1.9	1.2	1.7

Table (3) shows the effect of the lipase enzyme on the production of biofilm from bacterial *pseudomonas aeruginosa*. The results showed significant differences for the production of biofilm using the lipase enzyme with control, where three different concentrations of this

enzyme were used. 40,80,160% gave the concentration 160% of the enzyme the best results compared With the control of the concentrations and with control, there were no significant differences for the enzyme concentrations together.

Table 4: ANOVA table of effect of the lipase enzyme on the production of biofilm from *P.aeruginosa*

ANOVA					
production of biofilm					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	5.343	3	1.781	377.719	.000
Within Groups	.075	16	.005		
Total	5.418	19			

Table (4) shows the statistical analysis of the effect of an enzyme on the production of biofilm from bacterial *pseudomonas aeruginosa*. The results of the statistical analysis showed that there were significant differences for all treatments with control

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