

Experimental Study the role of LasA Protease of *Pseudomonas aeruginosa* in the Treatment of Bacterial Keratitis Caused by *Staphylococcus aureus*

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Summary:

Background: LasA protease play a major role in the colonization of the bacteria to the cornea during bacterial keratitis by preventing other bacteria from colonization to the cornea, for example in the mixed infection with *S. aureus* the enzyme eradicate the bacteria by their lysis it and finally eliminate the competitive for *P. aeruginosa* bacteria.

Objective: To study the role of LasA protease of *Pseudomonas aeruginosa* in the treatment of experimental keratitis caused by *S. aureus*.

Patients and methods: One hundred - twenty clinical samples (corneal scraping) were collected from patients suspected with bacterial keratitis presenting to Ibn Al-Haitham Teaching Hospital from May 2013 until November 2013. The bacterial isolate of *P. aeruginosa* that harbored LasA gene and capable of production of enzyme was analyzed by Real - time PCR. LasA protease enzyme was extracted by cold centrifugation and purified by gel - filtration chromatography.

Results: The results of the experimental treatment of bacterial keratitis (in vivo) of infected rabbits eyes caused by *S. aureus* has showed that the efficacy of LasA protease was effective as Lysostaphin drug in eradicating the *S. aureus* from the infected corneas. While Vancomycin drug gave very little potency in the eradicating *S. aureus* from corneas in comparison with potency of LasA protease and Lysostaphin during this time but showed good potency with very late period (approximately after 3 days) after application of treatment.

Conclusion: The current study revealed that the LasA protease efficacy in the experimental treatment of bacterial keratitis of rabbit's eye corneas was good and similar to that of Lysostaphin in eradicating *S. aureus* from corneas of these animals and was higher than that of Vancomycin.

Key words: LasA protease, treatment of bacterial keratitis, *S. aureus*.

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Introduction:

Pseudomonas aeruginosa produces several proteases and toxins, the best characterized being exotoxin, elastase (LasB), LasA protease, alkaline protease and protease IV. Proteases enzymes are important predisposing factors that's mediates the establishment of bacterial infection and enhanced damage to the corneas (1,2,3).

LasA protease (also called staphylolysin) is a 20 kDa and elastolytic and staphylolytic endopeptidase that are secreted by *P. aeruginosa*. LasA protease play a major role in the colonization of the bacteria during bacterial keratitis by preventing other bacteria from colonization to the cornea, for example in the mixed infection with *S. aureus* the enzyme eradicate the bacteria by lysis it and finally eliminate the competitive bacteria for *P. aeruginosa* (4). LasA protease can

cause lysis of a wide range of). *S. aureus* strains, and it has also been shown to inhibit the growth of *S. aureus* cells in vitro (5,6). This favor LasA protease as another useful agent in enzyme - based treatment of *S. aureus* infections. LasA protease is not a major virulence factor in experimental *P. aeruginosa* keratitis (7,8,9).

Patients and methods:

Specimens collection: One - hundred and twenty patients with suspected microbial keratitis (corneal ulcer) presenting to Ibn Al-Haytham Teaching Eye Hospital. from May 2013 to November 2013, were included in this study. Corneal scraping was done by an ophthalmologist using a sterile bent-tipped needle.

Isolation : The corneal scraping was inoculated onto 4 types of cultural media: which were included: Blood agar inoculated (aerobically and an aerobically) for detecting the type of haemolysis, MacConkey agar to differentiate organisms according to their ability to ferment lactose, Chocolates agar

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