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Plasmid Mediated Quinolone Resistance in Clinical Isolates of *Klebsiella Pneumonia*

A Thesis Submitted to the Council of collage of Science /Al Muthanna University as Partial Fulfillment of the Requirements for the Degree of Master of Science in Biology

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Abstract

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Klebsiella pneumoniae is a gram-negative bacteria , is a significant member of the *Enterobacteriaceae*family .Is a facultative anaerobe can survive in environments with varying oxygen levels, making. This bacterium is commonly found on the skin, in the mouth, and in the intestines as part of the normal flora.. It is responsible for causing pneumonia, which is characterized by severe inflammation in the lungs. Quinolones are among the most commonly prescribed antimicrobials because of their broad-spectrum activity. Plasmid-mediated quinolones resistance (PMQR) mechanisms play a significant role in quinolone resistance. The main purpose of this study was to investigate the dissemination of PMQR genes among clinical isolates of *K. pneumoniae* recovered from samawwa hospitals. During the study period, a total of 215 from both sexes and from different sources (urine, sputum and pharyngeal swabs). These samples were collected from Al-Hussein Hospital, women and Children Hospital in city of Samawah, from September to December, 2022. The *Klebsiella pneumonia* were identified by traditional biochemical tests, and confirmed by VITEK-2 system.

The results of isolation have been shown 85 isolates of *Klebsiella pneumoniae* (39%) were isolated and diagnosed using Specialized media (Hichrom *Klebsiella* selective agar media, and Extend Spectrum β_Lactam ESBL media), Also, the ability of the isolates to resist antibiotics (disc diffusion method)was studied. It was found that these isolates were resistant to the following antibiotics (Amoxicillin, Amoxicillin_culvanic acid, Cefotaxime, Ceftriaxone, Trimethoprim-sulfamethoxazole, Ceftazidime, and Tetracycline) as percentage(100%,22%,62%,100%,,25%,88.8%,18%) respectively. Quinolone Antibiotics (Ciprofloxacin, Giatifiloxacin , Nitrofurazole ,Ofloxacin , Levofloxacin

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,Norfloxacin and Nalidixic acid)showed resistance As percentage (90% ,48%,38%,35%,45%,93%,and100%) respectively. Also, these isolates demonstrated their sensitivity towards the two antibiotics (Meropenem and Imipenem) as percentages (70% and 78%) respectively.

The isolates were tested for the ability to produce biofilm (by Micro titter plate method) as one of the most important factors of their virulence. It was found that the 45(66%) were biofilm-producing at a moderate rate majority from urine and sputum samples, and the 22(34%) was not productive for this factor.

And by using molecular diagnostics, the genes of resistance to quinolone antibiotics (*qnr a, qnr b* and *qnr s*) were investigated. Our study showed the presence of (*qnr s* and *qnr b*) in different proportions in isolates with no presence of (*qnr a*) in any isolates in this study.

In order to compare the isolated genes' bases with those of other global isolates, the sequencing analysis of those bases was examined. One isolate (designated T1 isolate) had no nucleic acid variation when *QnrB* sequences were aligned with the most similar *Klebsiella pneumoniae* reference sequences 100%identifies (GenBank acc. no. CP095426.1). Additionally, none of the ten isolates (U1- U10) that are designated to amplify this locus had any nucleic acid variants,according to the alignment of *QnrS* sequences with the most comparable reference sequences of *Klebsiella pneumoniae* identifies 100%(GenBank acc. no. CP124706.1).