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Molecular detection of *rec A* gene from some pathogenic bacteria and related with antibiotic resistance

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By

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Abstract

The emergence of multidrug-resistant bacterial pathogens is considered to be a public health risk. Emergence of antimicrobial resistance in studied isolates (*Proteus mirabilis*, *Vibrio cholera*, *Pseudomonas aeruginosa*, *Escherichia coli* and *Salmonella typhi*) was related to the acquisition of extra genetic elements. In this work, the molecular characterization of studied bacterial *RecombinaseA* gene was compared to isolates from the GenBank . In the present study, a total of 67 clinical specimens were collected from urine, burns, urine and blood for *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Escherichia coli* and *Salmonella typhi* respectively from patients who had been admitted to Al Hussein Educational Hospital in addition to Women and Children Educational Hospital in Al- Samawa city during the period from June 2020 to November 2020. Ten DNA samples were collected for *Vibrio cholera* from stool samples obtained from patients infected with cholera disease. Then the specimens were cultured in selective media and identified by using bacteriological , biochemical tests ,API20E and VITEC2 compact system.

Antimicrobial susceptibility was carried out against 7 different antibiotics for studied isolates (*Proteus mirabilis*, *Pseudomonas aeruginosa*, *Escherichia coli* and *Salmonella typhi*). Bacterial isolates showed high resistance in different levels toward all tested antibiotics according to (CLSI,2019). The highest rate of resistance was detected in *P. mirabilis* against Sulphamethazol-trimethoprim as it reached 10/15(66.6%) while *P. aeruginosa* against Ceftazidime as it reached 14/17(82.3%). The antimicrobial resistant tests showed a high resistant of *E.coli* isolates to the Ampicillin and Trimethoprim as it reached 20/20(100%) and the study proved that the antimicrobial resistance rate of *S.typhi* high resistance rate for Ampicillin was 15/15 (100 %); for Amoxicillin-clavulanate was 15/15 (100 %) ; for Cefotaxime was 15/15 (100%),and Ciprofloxacin was 15/15 (100%).

Abstract

Genomic DNA was extracted from the bacterial culture then tested for *recA* gene using PCR technique. Molecular results of the present study showed that all bacterial isolates contained *recA* gene with PCR technique. In this study, two comparisons were made to detect the sequence homology of *recA* gene among studied samples, the first comparison was against reference sequence, while the second comparison was used against the closest reference and representative international isolates. Sequencing results of *recA* gene showed different levels of sequence similarity between studied samples and reference gene. The similarity with reference gene of *recA* gene in *P. mirabilis* showed only one sample (A2) give 100% Similarity to *recA* gene, while the other samples (A1,A3,A4,A5 and A6) give 99%. The similarity with *recA* gene in *V. cholera* showed all samples (B2,B3,B4,B5 and B6) give 100% Similarity to *recA* gene except one sample (B1) give 92% in comparison with reference gene. Four samples (C2,C3,C4 and C6) give 100% identity to *recA* gene, while the samples (C1 and C5) give 99% Similarity to *recA* gene for *P. aeruginosa*. For *E. coli* the similarity with *recA* gene showed one sample (D3) give 100% identity to *recA* gene, while the second one (D2) give 92%. Also, the three samples (D1,D5 and D6) give 99% identity to *recA* gene whereas (D4) gives 98% identity. The similarity with *recA* gene in *Salmonella typhi* showed all samples (E1,E2,E3,E4,E5 and E6) give 100% Similarity to *recA* gene.

Phylogenetic tree analysis showed that each bacteria (except *salmonella typhi*) grouping into different clusters with different clades that were branched from the tree root with different bootstrap value indicating that these strains were genetically diverse in comparison with reference *recA* gene.