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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.