

MOLECULAR DETECTION OF OPPORTUNISTIC BACTERIA *PANTOEA SPP.* FROM DIFFERENT HUMAN SAMPLES

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Abstract

The aim of this study was molecular detection of opportunistic bacteria *Pantoea spp.* From different human samples. From June 2021 to June 2022, 141 clinical samples were gathered from clinics throughout the Babylon province. Swabs were taken from various clinical samples, including 28 samples from the pharynx, ears (26), stool (22), wounds (18), pus (15), cough (17) and burns (16). These samples were cultured on primary medium, then biochemical tests were done for identification of bacteria. The DNA of bacteria were extracted by using kit purchased from (Promega, Madison). *Pantoea* species were identified using gene 16sRNA primers. By bacteriological methods, out of 140 samples, 6 isolates were suspected as *Pantoea spp.* On MacConkey agar, smooth, convex, punctate, umbilicated fermenting lactose, shining colonies were produced, after Gram staining revealed Gram negative rods. The molecular way demonstrates that the isolates are from *Pantoea* species. During this investigation, the PCR method has a 100% sensitivity rate for isolating isolates.

In conclusion, *Pantoea spp.* was isolated from different human sources in significant percentage, which may be causative agent for different diseases.

Keywords: Sport Psychology, Exercise, Molecular, *Pantoea spp.*, Human

Introduction

Various hosts, including plants, animals, insects, as well as humans, are often found in close proximity to *Pantoea* strains [1,2]. Although several *Pantoea* species are well-known plant pathogens [3-5], they have also been identified from clinical samples. *Pantoea agglomerans* has been identified from the blood of children suffering from bacteremia, septicemia, peritonitis, osteomyelitis, septic arthritis, pneumonia, as well as septic arthropathy [6]. Cases of *P. agglomerans* in humans are often the consequence of hospital-acquired infections or contamination of a wound by plant matter [7]. Isolates of other species, including as *Pantoea eucalypti*, *Pantoea ananatis*, *Pantoea dispersa*, as well as *Pantoea septica*, have also been found in many clinical sources, such as blood, wounds, CSF, faeces, skin, cysts, abscesses, fractures, as well as urethra & trachea [2].

Additionally, *pantoea* has been linked to several epidemics that have killed newborns [8,9]. The pathogenic potential of some *Pantoea* species for humans is still being challenged [10], despite data showing that many clinical strains are not *Pantoea* at all. Many strains that were formerly thought to be members of the *Pantoea*

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genus have been reassigned to other genera as a consequence of taxonomic and nomenclatural changes [11]. Moreover, it is difficult to place *Pantoea* strains into a species group using solely their metabolic profile, which has led to the misidentification of some *Pantoea* isolates [12].

The enterobacteriaceae family, which does not stain positively on the Gram stain, is known to include the bacteria *Pantoea spp.* [13]. The primary issue is that, although not being a mandatory infection, *Pantoea spp.* bacteria are linked to opportunistic illnesses [14].

The cellulase enzyme, for instance, is useful for studying the cellulose polymer that makes up the walls of plant cells, which both defends the plant against invasion and makes the bacterium more dangerous. The genes responsible for the synthesis of exopolysaccharide (EPS) in *Pantoea spp.* [15] have been identified, and EPS plays an important role in the adherence and pathogenicity of *Pantoea spp.* The regulatory genes *rcaA* and *rcaB* regulate the synthesis of the exopolysaccharide known as capsule polysaccharide (EPS) [16].

Producing more pilius is facilitated by the *hpaA* gene, which in turn improves host pathogenicity [17] by making bacteria more adept at sticking to host tissue and interfering with cellular host function.

The aim of this study was molecular detection of opportunistic bacteria *Pantoea spp.* From different human samples.

Materials and Methods

From June 2021 to June 2022, 141 clinical samples were gathered from clinics throughout the Babylon province. Swabs were taken from various clinical samples, including 28 samples from the pharynx, ears (26), stool (22), wounds (18), pus (15), cough (17) and burns (16). These samples were cultured on primary medium, then biochemical tests were done for identification of bacteria [18].

The DNA of bacteria were extracted by using kit purchased from (Promega, Madison). *Pantoea* species were identified using gene 16sRNA primers. The gene sequence was:

F CCTGGACAAAGACTGACGCT, R CGCTTCTCTTTGTATGCGCC

The reaction was carried out in 34 cycles, each lasting 1 minute at 95°C, 45

seconds at 53°C, and 2 minutes at 72°C after a 5-minute initial denaturation. Five l of the PCR product were run on 1% (w/v) agarose gel after PCR amplification, stained with ethidium bromide, and examined under a UV transilluminator.

Results and Discussions

By bacteriological methods, out of 140 samples, 6 isolates were suspected as *Pantoea spp.* On MacConkey agar, smooth, convex, punctate, umbilicated fermenting lactose, shining colonies were produced, after Gram staining revealed Gram negative rods.

By using API 20E assays, the *Pantoea spp.* were biochemically described. All six isolates fit the description of the *Pantoea* genus. They lacked urease, did not decarboxylate lysine or ornithine, and did not create H₂S from thiosulfate. To the best of the API 20E system's knowledge, no species-level identifications have been made with any of the isolates. However, only *Pantoea* species are included in the current database; these findings have been verified in the past by [19].

In this work, the isolates of *Pantoea spp.* were identified by using of gene 16sRNA. The findings in Fig. 1 demonstrate that the gene 16sRNA, which is the diagnostic gene responsible for this bacteria, is present in all isolates and has a molecular weight of (523 bp). This demonstrates that the isolates are from *Pantoea* species. During this investigation, the PCR method has a 100% sensitivity rate for isolating isolates (Figure 1).

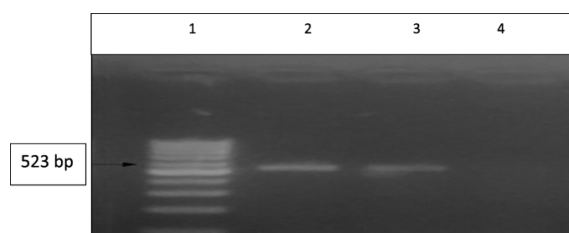


Figure 1: The 16sRNA gene investigation of the *Pantoea spp.* is shown by agarose gel electrophoresis of the PCR experiment. Positive *Pantoea* species are isolated using the ladder 100bp, the gene was have 523bp.

The 16S rRNA gene was used to screen 18 isolates for *Pantoea spp.*, and 9 of the isolates were found to be members of this bacterial family, as reported in the study [20]. A study of 21 isolates of the genus *Pantoea* found that 95.6% had the 16S rRNA gene, making them potentially dangerous to humans and other animals [21].

Conclusion

Pantoea spp. was isolated from different human sources in significant percentage, which may be causative agent for different diseases.

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