

Providencia Rettgeri Common Causes of UTI

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ABSTRACT

Wasit city, Iraq provided a total of twenty-four different samples of urine for the researchers to examine. The samples were first cultured onto Ma-Cconkey agar and Blood agar, and placed into brain heart infusion broth. Microscopic inspection, cultural characteristics, biochemical assays, and the Api20 E system were all utilized to independently verify the isolates' identities. Only two out of twenty-four human samples (8.3%) included *Providencia* species, with *P. rettgeri* being the most prevalent of those species. Using the disc diffusion technique, tests of antimicrobial susceptibility were carried out against 10 different antibiotics. All of the *Providencia* isolates had evidence of multidrug resistance (MDR), and the level of absolute resistance to cefoxitin, methicillin, vancomycin, doxycycline, clarithromycin, and trimethoprim/sulfamethoxazole was 100%. They exhibited a high degree of sensitivity to ofloxacin, chloramphenicol, cefixime, and teimethoprim, with a percentage of 100%.

KEYWORDS: *Providencia*, *rettgeri*, UTI

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1. INTRODUCTION

The genus *Providencia* constitutes flora that is naturally found in the human digestive system. The species *Phaeobacter alcalifaciens*, *Phaeobacter rettgeri*, *Phaeobacter stuartii*, *Phaeobacter rustigianii*, and *Phaeobacter heimbachae* are the ones that are most often seen. It is frequently discovered in places such as the soil, the water, and the sewage. (O'Hara et al., 2000). Urine, throat, perineum, axillae, feces, blood, and wound specimens from humans have all been analyzed and found to include human isolates of the *Providencia* species. In the past, it had a reputation for being a rare disease since very little was known about the potential for it to induce a nosocomial infection. It is now well acknowledged as a dreaded opportunistic pathogen that is capable of triggering a wide range of nosocomial illnesses.. (O'Hara et al., 2000 ; Hee et al., 2015)

UTIs, gastroenteritis, and septicemia are typical illnesses caused by *Providencia*. Multiple additional illnesses, such as burns, pneumonia, newborn sepsis, community- and hospital-acquired neuroinfection, etc., are also reporting an increase in this phenomenon. (Hee et al., 2015 ; Sharma et al., 2017 ;

Gupta and Kachru .2017 ; Siddharth et al ., 2017 ; Maiti et al ., 2013) Isolation from the therapeutic environment is strongly linked to the prevalence of long-term urinary catheter usage in patients who are critically ill, those with diabetes, and those with other immunocompromised illnesses. Recent studies have shown an increase in *Providencia*-related illnesses and have called for much more research into the topic. (O'Hara et al ., 2000 ; Hee et al ., 2015 ; Sharma et al ., 2017 ; Gupta and Kachru .2017 ; Siddharth et al ., 2017 ; Maiti et al ., 2013 ; Shiwani et al ., 2021)

Because clinical strains are inherently resistant to several routinely used antibiotics, such as ampicillin, first-generation cephalosporins, polymyxins, and tigecycline (O'Hara et al., 2000), treatment of these infections is a challenging endeavor. In addition, during the past several years, an ever-increasing incidence of antibiotic resistance has been documented. As a result, this is a significant newly emerging pathogen. Their antibiogram is undergoing rapid change, and it has also been reported that extended-spectrum beta-lactamase (ESBL) and carbapenemase-producing *Providencia* strains are becoming more prevalent. (Abdallah

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and Balshi .2018 ; Mataseje et al ., 2014 ; Tatsuya et al .,2014). A significant problem in clinical practice is the growing isolation of such strains from the samples of patients. In lately, there have also been sporadic reports of epidemics coming from other regions of the world. (Saeam et al .,2018) .

MATERIAL AND METHODS

Sample Collection

Twenty-four urinalysis samples were collected. samples have been obtained from individuals at a hospital in Wasit. Human participants were employed to gather the samples. After the samples were taken, they were streaked over the nutritional medium and incubated at 37 degrees Celsius for a whole day in Ma-Cconkey and blood agar

Identification of the Isolates

After using an API 20E suspension medium to create the bacterial suspension from pure isolated colonies, the turbidity was brought down to 0.5 McFarland tubes. (1-1.5x10⁸ CFU. ml-1). The bacteria suspension was transferred to the twenty microtubes using a sterile Pasteur pipette. The microtubes were inoculated according to the manufacturer's instructions, and they were incubated for 24 hours at 37 degrees Celsius. After this, the isolates were identified by using the numerical coding of the API 20E system for confirmatory identification at the species level..

Antibiotic Sensitivity Test (Qualitative Disk Method)

Antimicrobial susceptibility testing was determined using the disk diffusion protocol. Bacterial suspension was adjusted to McFarland tube No. 0.5 that contained 1-1.5 10⁸ CFU/ml streaking on Mueller-Hinton (MH) agar (Oxoid, UK). Then the antimicrobial disks were applied firmly to the surface of the inoculated agar plates and incubated at 37°C for 18–24 hours, and the inhibition zone diameter results were measured and recorded (CLSI 2019; EUCAST 2020). Ten antibiotic disks (Bioanalyse, Turkey) were used, including cefoxitin, methicillin, vancomycin, doxycycline, clarithromycin, trimethoprim/sulphamethoxazole, ofloxacin, chloramphenicol , cefixime, and teimethoprim. Multidrug resistance (MDR) and the multiple antibiotic resistance (MAR) index were determined according to Krumperman et al. (1983; Magiorakos et al. (2012).

RESULTS AND DISCUSSION

1. Isolation and Characterization of Providencia spp.

Twenty four urine samples had been collected from in wasit cities patients hospitals. as showing in table (1). On the basis of their cultural and microscopic characteristics, two local isolates were characterized. Species and genera have been determined using biochemical assays and the API 20 E confirming test.

Table 1. Distribution of Providencia rettgeri in urine samples, including sample types, numbers, and percentages

| Types of sample | Number of samples | Number of isolates | Percentage% |
|-----------------|-------------------|--------------------|-------------|
| Human | 24 | 2 | 8.33 % |

Cultural Characteristics

The isolates displayed various colonies on selective and differential culture media that have grown in 24 hours at 37°C. The colonies of the three isolates showed a pale-yellow due to lactose non-ferment on MacConkey agar; on nutrient agar, these colonies appeared milky-white round and slightly convex. One isolate was β hemolysis on BA . These results are comparable with many results (Jasim, 2015 ; Procop et al., 2017).

Microscopic Characteristics.

Under a light microscope examination, the isolates appeared as single, rod gram-negative bacilli, after 24 hours post-incubation at 37°C.

Biochemical tests

The results of biochemical tests using the API-20E system 2 (Table 2), that showed two isolates of Providencia rettgeri the percentage of isolates were identified on 99.4% . (Somvanshi *et al.*, 2006; Khunthongpan *et al.*, 2013).

Table 2. Api 20E technique of Providencia rettgeri

| No. | Active ingredients | Symbol test | Results |
|-----|---------------------------------------|------------------|---------|
| | Ortho NitroPhenyl-Bd-Galactopyranside | ONPG | - |
| | L-arginine | ADH | - |
| | L-Lysine | LDC | - |
| | L-Ornithin | ODC | - |
| | Trisodium citrate | CIT | + |
| | Sodium thiosulfate | H ₂ S | - |
| | Urea | URE | + |
| | L-tryptophane | TDA | + |
| | L-tryptophane (indole production) | IND | - |
| | Sodium pyruvate | VP | - |
| | Gelatin (bovine origin) | GEL | - |

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| | | | |
|--|------------------------|-----|---|
| | D-Glucose | GLU | + |
| | D-Mannitol | MAN | - |
| | Inositol | INO | + |
| | D-Sorbitol | SOR | - |
| | L-Rhamnose | RHA | - |
| | D-Saccharose (sucrose) | SAC | - |
| | D-Melibiose | MEL | - |
| | Amygdaline | AMY | + |
| | L-Arabinose | ARA | - |

Antimicrobial susceptibility

The *Providencia rettgeri* antimicrobial susceptibility test isolates from humans shows that resistant was 100% to cefoxitin, methicillin, vancomycin, doxycycline, clarithromycin and Trimethoprim/ Sulphamethoxazole . they

were highly susceptible (100%) to Ofloxacin ,chloramphenicol, cefixime, and teimethoprim. the prevalence of MDR was 100%, the MARI was 0.5 as shown in Table (3) .

Table 3. Susceptibility as antimicrobial versus isolates of *P. spp.* in human

| No. | Antibiotic | S % | R % |
|-------------|---------------------------------------|------------|-----|
| 1 | Ofloxacin 5 µg | 100 | 0 |
| 2 | Chloramphenicol 30 µg | 100 | 0 |
| 3 | Trimethoprim/ Sulphamethoxazole 25 µg | 0 | 100 |
| 4 | Methicillin 10 µg | 0 | 100 |
| 5 | Vancomycin 30 µg | 0 | 100 |
| 6 | Cefoxitin 30 µg | 0 | 100 |
| 7 | Cefixime 5 µg | 100 | 0 |
| 8 | Trimethoprim 5 µg | 100 | 0 |
| 9 | Doxycycline 30 µg | 0 | 100 |
| 10 | Clarithromycin 15 µg | 0 | 100 |
| MARI | | 0.5 | |

Enterobacteriaceae cause severe infections and also members of this family have become progressively resistant to a wide range of antimicrobial drugs used against them (Denton, 2007). The isolates showed MDR to antimicrobial agents and the MAR index was more than 0.2 indicates that isolates are at risk, according to (Krumperman, 1983; Magiorakos et al., 2012). The results were following other studies, which showed *Providencia spp* were MDR (Cao et al., 2017; Al-Gburi 2020). *P. rettgeri* are reported resistant to many antimicrobials such as polymyxins, first-generation cephalosporins, ampicillin, and tigecycline (Magiorakos et al., 2012). Moreover, in last year's *P. rettgeri* has become significantly important due to the emergence of carbapenemase-producing strains (Mataseje et al., 2014; Shin et al., 2018). In the present study, all of *Providencia rettgeri* isolated from human, showed resistance to Trimethoprim-Sulfamethoxazole, Methicillin, Vancomycin, Cefoxitin, and Clarithromycin, and all isolates shows sensitive to Ofloxacin , Trimethoprim, Cefixime and Chloramphenicol, these results are close to many studies with some differences, the agreement of the results with Al-Janabi, (2012) and Sharma et al., (2017) who found *P. rettgeri* isolates were resistant 100% to Tetracycline, Clarithromycin and Amoxicillin-Clavulanate.

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