

Clinical Correlation, Isolation, Speciation, and Antibiotic Susceptibility Pattern of Tribe Proteeae from Various Clinical Samples

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Abstract

Introduction: Organisms belonging to Tribe Proteeae are usually present as commensals in the environment but can cause urinary tract and wound infections. There is a need for regular studies on sensitivity pattern of various bacteria causing infections to decide a suitable empirical therapy and there are not many studies on Tribe Proteeae.

Aim: To clinically correlate, isolate, speciate and determine the antibiotic susceptibility pattern of Tribe Proteeae from various clinical samples.

Materials and Method: All samples were cultured on standard culture media as per routine protocol. Isolates were identified by standard biochemical reactions. Antibiotic sensitivity was done by Kirby Bauer's disk diffusion method as per CLSI guidelines. ESBL production was detected by double disk diffusion method using ceftazidime (30µg) disk and ceftazidime/clavulanic acid (30/10µg) disk. Clinical data was collected from the record section of the hospital.

Results: Sixty samples yielded bacteria belonging to Tribe Proteeae. Most of the patients belonged to male gender 46 (76.7%) and >50 years 38 (63.3%) of age. *P. mirabilis* was the predominant isolate 42 (70%). Maximum rate of isolation of *Proteus spp* was from diabetic foot ulcer 21 (35%). The predisposing conditions were diabetes mellitus, tracheostomy cases and only one case of carcinoma tongue. Most of our isolates were sensitive to various drugs tested. There were only three (5%) ESBL producers. Most of the isolates of *P.mirabilis* were sensitive to antibiotics than *P.vulgaris* isolates.

Conclusion: Maximum rate of isolation of tribe Proteeae was from diabetic foot ulcer cases. *P.mirabilis* was the predominant isolate and was more sensitive to the antibiotics tested than *P.vulgaris*.

Key words: Beta lactamases, Diabetic foot, *Proteus mirabilis*, *Proteus vulgaris*

Introduction

The three genera, *Proteus*, *Providencia* and *Morganella* belong to Tribe Proteeae These bacteria are

usually found in water, sewage and soil as they are the normal flora of the intestine. ⁽¹⁾ They cause a variety of community-acquired and nosocomial diseases, which include bacteremia, wound infections and urinary tract infection. ⁽¹⁻⁴⁾

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Among genus *Providencia*, *P. rettgeri* is most frequently isolated from urine of the catheterized patients in hospitals and nursing homes.⁽⁴⁾ The genus *Morganella* causes opportunistic infections in humans. ^(1,4,5)

The antibiotic resistance due to the inadequate and inappropriate use of antibiotics is a public health

problem, hence the need for regular studies on sensitivity pattern of various bacteria causing infections to decide a suitable empirical therapy.⁽⁶⁾ The present study aims to clinically correlate, isolate, speciate and determine antibiotic susceptibility pattern of Tribe Proteeae from various clinical samples.

Materials and Method

It was a prospective time bound study conducted from October 1st 2017 to March 31st 2018 in Microbiology department of a tertiary care hospital in South India. All samples received at microbiology laboratory which were positive for Tribe Proteeae and the gram's stain picture indicates active infection from October 1st 2017 to 31st March 2018 were included in the study. Samples which did not yield Tribe Proteeae, which were received on dates other than the period of study and which yielded scanty mixed growth were excluded. Clinical data was taken from the records section of the hospital. Gram staining was done on all samples. All samples were cultured on Mac Conkey's agar, Chocolate agar and Blood agar. Semi quantitative standard loop method were used for urine samples and they were cultured on CLED, Mac Conkey agar and on blood agar.⁽⁷⁾ Isolates were identified by standard biochemical reactions.⁽⁷⁾

Antibiotic sensitivity was done by Kirby Bauer's disk diffusion method according to CLSI guidelines.⁽⁸⁾ ESBL production was detected by double disk diffusion method wherein Ceftazidime (30µg) disk and Ceftazidime/clavulanic acid (30/10µg) disk was used. If the difference between the two zones is > 5 mm the test was considered as positive.⁽⁸⁾

Statistical Analysis

Statistical analysis was performed by using SPSS version 16.0. Also between two variables statistically significant correlation will be done using Chi Square test. P value <0.05 was considered as significant.

Results

Sixty samples yielded bacteria belonging to Tribe Proteeae. Of these there were 46 (76.7%) male and 14 (23.3%) female; 38 (63.3%) were >50 years of age, 15 (25%) were 30-50 years of age and 7 (11.7%) belonged to <30 years of age. Of the 60 samples, there were 21 (35%) cases of diabetic foot ulcer, 18 (30%) cases of wound infection, 8 (13.3%) cases of endotracheal tube tip, 5 (8.3%) cases of urinary tract infection, 4 (6.6%)

cases of skin and mucous membrane infection, 2 (3.3%) cases of sepsis were blood culture became positive, one (1.6%) case of endophthalmitis and one case (1.6%) of cholangitis where bile was received (Figure 1). Of the different skin and mucous membrane infection we had one case of carcinoma tongue with an ulcer over the tongue, 2 cases of perineal and perianal abscess, discharging sinus over axilla, one case of eczema and cellulitis. Diabetic foot ulcer patients were treated with magnamycin (cefaparazone sodium) and clindamycin. Tracheostomy cases were put on cefaperazone sulbactam, Cholangitis was treated with meropenem, eczema, cellulitis with augmentin (amoxycylav) and clindamycin, discharging sinus over the axilla was treated with linozelid and chloramphenicol, perianal abscess was treated with amoxiclav, metronidazole, clindamycin. Almost all patients had underlying co morbidities like diabetes mellitus or carcinoma.

Of the different isolates, there were 42 (70%) *Proteus mirabilis*, 15 (25%) *P.vulgaris*, 1(1.7%), *P.penneri*, 2 (3.3%) *Providencia* spp. Most of the isolates were sensitive to all the drugs tested. There were only three isolates which were ESBL producers (one isolate was *P. mirabilis* from urine sample, one isolate of *P. vulgaris* from diabetic foot ulcer and the third isolate was *P. penneri* from diabetic foot ulcer). Comparatively *P.vulgaris* showed more resistance to drugs then *P. mirabilis* (Table 1). *P. penneri* and *Providencia* spp showed resistance to most of the drugs studied but the number of isolates were very less to conclusively say so. We did not find any statistically relevant correlation between different clinical conditions and the antibiotic resistance.

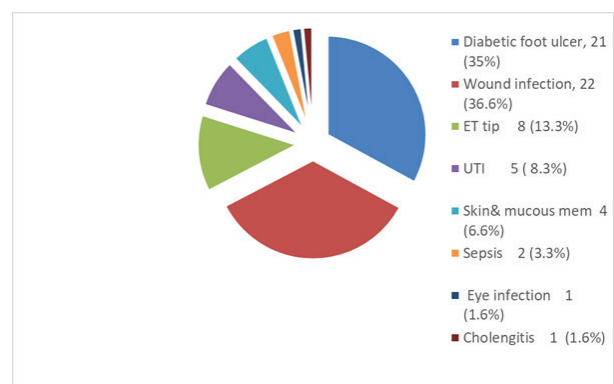


Figure 1: Clinical Correlation

Discussion

In a study conducted in Rajasthan, India, of 1876 clinical samples 5.4% (101/1876)

yielded *Proteus* species. Maximum isolation was from pus 80.2% (81/101) followed by urine 8.9% (9/101) and also maximum proteus species was isolated from inpatients than out patients and 91% (92/101) were ESBL producers. *P. mirabilis* was the predominant isolate.⁽²⁾

In a study conducted in Chandigarh, India, 60 consecutive non-repeated strains of *P. mirabilis* were evaluated for production of ESBLs, AmpC b-lactamases, and carbapenemases. Of these, 36 (60%) isolates were found to be ESBL producers, and 7 (12%) were positive for production of AmpC b-lactamases.⁽³⁾

In a study conducted in Ghana, Africa, of the 200 *Proteus* species obtained from patients suspected of bacterial infection, wound isolates were the highest (64.5 %) followed by ear swab. *P. mirabilis* was the commonest species (61.5 %), followed by *P. vulgaris* (30.5 %), and *P. penneri* (8 %). There was no significant difference between the out-patient and in-patient cases, just as there were no preferences for gender or age of the patients. All the species were resistant to chloramphenicol, ampicillin and co-trimoxazole. About 72.9 % of the isolates produced β -lactamase and 88.5 % were resistant to more than 2 antibiotics. *P. penneri* was the most resistant among the recovered species.⁽⁹⁾

In a study conducted in Mumbai, India, three *Proteus* species were recovered from 56 (1.12%) of the 4995 clinical samples collected, 38 of these samples (67.85 %) were taken from male patients and 18 (32.14 %) from females. Wound samples contributed the highest percentage of *Proteus* (67.85%) followed by urine. ESBL production was highest in *Proteus penneri* 58.33% followed by *Proteus vulgaris* 33.33% and *Proteus mirabilis* 8.33%..⁽¹⁰⁾

In a study conducted in Tamil Nadu, India, out of 3972 clinical samples 121 (3.04%) *Proteus* strains were isolated. *Proteus mirabilis* was more commonly isolated than *Proteus vulgaris*. Pus (44.6%) and urine (50.4%) were predominant samples. There were more numbers of male (63.6%) patients than female (36.4%). Isolates of *Proteus* spp were highly sensitive to imipenem, piperacillin-tazobactam, ofloxacin and highly resistant to ampicillin, amoxycillin-clavulanic acid and 24.8% were ESBL producers. ⁽¹¹⁾ In the present study also most of the patients were male and *P. mirabilis* was the predominant isolate but ESBL production was only 5% (3/60).

In another study, 100 isolates of Tribe Proteaceae were collected over a period of 6 months from clinical specimens obtained from patients suspected of bacterial infection. Wound infections were the highest (52%) followed by urine (47%). *Proteus mirabilis* was the predominant species (61.5 %), followed by *Proteus vulgaris* (30.5%). More than 80% were resistant to ampicillin and cefazolin, 34% to quinolones, 32% to aminoglycosides and 48% to Co-trimoxazole. About 49 % of the isolates produced ESBL and 16 % were multidrug resistant. *Providencia stuartii* was the most resistant among the recovered species. Maximum sensitivity was noted with carbapenems and maximum resistance with ampicillin and first and second generation cephalosporins. ⁽¹²⁾

In a study conducted at Republic of Cameroon, of the 1136 culture positive clinical specimens, 164 (14.4%) isolates were identified as members of Proteaceae of which 110 (67.1%) were from urine, 37 (22.6%) from wounds and burns, 10 (6.1%) from blood and 7 (4.3%) from CSF. Speciation of the Proteaceae isolates showed that 111 (67.7%) were *Proteus mirabilis*, 21(12.8%) *Proteus vulgaris*, 11 (6.7%) *Providencia alcalifaciens*, 6 (3.6%) *Providencia stuartii*, 4 (2.4%) *Morganella morganii* and 5 (3.0%) *Proteus penneri* and *Providencia rettgeri*. Most Proteaceae isolates were susceptible to imipenem, ceftazidime, chloramphenicol, gentamycin, nalidixic acid, ofloxacin and amikacin.⁽¹³⁾

In the present study as in studies conducted in past, most of the Tribe Proteaceae were isolated from diabetic foot ulcers, but we did not find much antibiotic resistance. But as in the past studies, *P. vulgaris* was more resistant to antibiotics than *P. mirabilis*.

Conclusions

Most of the patients belonged to male gender and >50 years of age. As in previous studies, *P. mirabilis* was the predominant isolate. Maximum rate of isolation of *Proteus* spp was from diabetic foot ulcer. The predisposing conditions were diabetes mellitus, tracheostomy cases and only one case of carcinoma tongue. Most of our isolates were sensitive to various drugs tested. There were only three ESBL producers. *P. mirabilis* was more sensitive to antibiotics than *P. vulgaris*.

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