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Research Article

Identification of *Proteus* Species in Different Clinical Samples and their Antibiotic Susceptibility Pattern at a Tertiary Care Hospital

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Abstract

The present study aimed to determine the prevalence of *Proteus* species in various clinical samples and to evaluate their antibiotic susceptibility patterns. A total of 2400 clinical specimens were analyzed over a six-month study period. All samples underwent direct microscopy and culture for the isolation and identification of *Proteus* species. Out of the total samples, 67 isolates of *Proteus* species were identified, of which *Proteus vulgaris* accounted for 38 cases (56.72%) and *Proteus mirabilis* for 29 cases (43.28%). The majority of isolates were obtained from pus and urine samples. Antibiotic susceptibility testing by the Kirby–Bauer disc diffusion method revealed high sensitivity to piperacillin-tazobactam (89.55%), amikacin (65.67%), and ceftriaxone (61.19%). In urine isolates, higher sensitivities were observed with fosfomycin (82.76%) and nitrofurantoin (65.67%). High resistance rates were recorded for colistin (100%), levofloxacin (44.77%), and imipenem (41.79%). Given the rising antimicrobial resistance among *Proteus* species, their increasing prevalence may pose a significant public health concern.

Keywords: *Proteus vulgaris*; Pus Samples; Antibiotics Sensitivity Testing

Introduction

The tribe Proteeae, which also comprises *Morganella* and *Providentia*, includes the genus *Proteus*, which is a member of the Enterobacteriaceae family. This group of Gram-negative, pleomorphic, motile, aerobic, and facultative anaerobic bacilli includes three unidentified genomospecies and five named species: *P. mirabilis, P. vulgaris, P. myxofaciens, P. hauseri,* and *P. penneri* [1]. Among the microorganisms that are frequently linked to both community-acquired illnesses and hospital infections are *Proteus* species [2]. This pathogen can infect many body anatomical areas due to its varied mode of transmission. Soil, tainted water, food, equipment, intravenous solutions, and the hands of patients and medical staff are some of the incriminating sources of transmission [3,4]. *Proteus* infection prevalence rates in MGM have been shown to range from 9.8% to 14.6% [5,6]. *Proteus* species are frequently challenging to remove from the host, particularly in patients with complex wounds, ulcers, catheterization, or urinary tract functioning problems [7]. *Proteus* infections are the third most common cause of infections linked to medical care, with a reported incidence of 9.8% to 14.6% worldwide [8]. *Proteus* species are responsible for about 5% of hospital-acquired UTI cases and 10–15% of severe UTI cases, primarily those related to catheterization [9]. Additionally, 10% of wound infections are caused by *Proteus* species, which can significantly increase morbidity and mortality [10]. Prolonged hospital stays, crowded wards, and careless antibiotic usage are likely the causes of the high prevalence of *Proteus* infections.[11] Since penicillin and cephalosporins are the most often used antibiotics for empirical treatment, *Proteus's* sensitivity pattern to these drugs is influenced by both intrinsic and extrinsic resistance mechanisms, such as the production of β lactamases [12]. *Proteus* resistance to different antibiotic groups has been rising, necessitating ongoing monitoring for improved therapy response [13]. The creation of β-lactamases is the main mechanism for resistance to β-lactam antibiotics, and drug resistance has been observed more frequently for this genus. The most prevalent β-lactamases are AmpC β-lactamases and extended spectrum β-lactamases (ESBLs) [14].

Materials and Methods

In the microbiology department of Era Lucknow Medical College and Hospital, a cross-sectional analysis was done from the month of October 2024 to March 2025. Chi-square test was used for data analysis. All samples have been collected and processed as per standard Microbiology protocols. The Sample size is calculated based on prevalence among the clinical samples using the formula $n=za^2pq/L^2$. The total samples are 2400 in size.

Inclusion criteria

All samples received in bacteriology lab (pus, urine, sputum, ear swab, blood, tissue, wound swabs, Ascitic Fluid) from patients of all age groups attending various OPD'S and IPD'S of ELMCH were included for the study.

Exclusion criteria

Stool and CSF samples were not included. Isolates other than *Proteus* species were excluded from the study due to inadequate amount of sample.

Direct examination

The laboratory received all of the samples shortly after they were all collected. On pristine sterile glass slides, the smears were created from each sample. They were dried, fixed with heat, stained using the Gram's Method, and examined under a microscope in immersion oil. All biochemical tests, the Indole, Methyl Red Urease test, the Citrate utilization test, Triple sugar iron test and sugar fermentation test were performed in order to identify *Proteus* species.

The clinical laboratory standard institute (CLSI) guidelines were used to interpret the result of an investigation into the antibiotic sensitivity pattern of *Proteus* species isolates to Colistin, Imipenem, Piperacillin-Tazobactam, Gentamycin, Tobramycin, Amikacin, Ceftriaxone, Ciprofloxacin, Levofloxacin by Kirby-Bauer method on Muller Hinton Agar (MHA).

Statistical analysis

Simple ratio and percentage statistic were used for the statistical study. To create the tables, Microsoft office 2007 was used.

Result

In the present study, 2400 clinical samples collected over a six-month period yielded 67 (2.79%) isolates of *Proteus* species. Out of 67 isolates 39 isolates were from OPD and 28 are from IPD. Maximum isolates were obtained from urine sample i.e., 48.28% and 37.31% from pus; other samples contributed less in number of isolates. Out of 67 isolates two species of *Proteus* present i.e., 40 *P. mirabilis and 27 P. vulgaris* (Table 1).

Table 2 presents the antibiotic susceptibility pattern of 67 isolates tested against various antibiotics. The highest sensitivity was observed with piperacillin–tazobactam (89.55%), followed by fosfomycin (82.76%) and amikacin (65.67%), indicating these drugs may be more effective treatment options. Complete resistance (100%) was noted for colistin, while high resistance

S. NO.	Sample	*P. vulgaris*	*P. mirabilis*	Total
1.	Urine	7	22	29
2.	Pus	14	11	25
3.	Tissue C/S	0	1	1
4.	Ascitic fluid	0	1	1
5.	Blood	2	3	5
6.	Sputum	2	1	3
7.	Endotrachial fluid	2	0	2
8.	Tip C/S	0	1	1
Grand total		27	40	67

Table 1: Species wise distribution of *Proteus* species in different clinical samples.

S. NO.	Antibiotics (n = 67)	Sensivity		Intermediate		Resistance	
		No.	%	NO.	%	NO.	%
1.	Amikacin	44	665.67%	11	16.42%	12	17.91%
2.	Amoxicillin-clavulanic	35	52.24%	11	16.42%	21	31.34%
3.	Ceftriaxone	41	61.19%	8	11.94%	18	26.87%
4.	Imipenem	33	49.25%	6	8.95%	28	41.79%
5.	Colistin	0		0		67	100%
6.	Levofloxacin	20	29.85%	17	25.37%	30	44.77%
7.	Ciprofloxacin	23	34.33%	19	28.36%	25	37.31%
8.	Cefoperazone-sulbactam	37	55.22%	8	11.94%	22	32.84%
9.	Piperacillin-tazobactam	60	89.55%	2	2.99%	5	7.46%
10.	Gentamicin	36	53.73%	14	20.90%	17	25.37
11.	Netilmicin	34	50.75%	10	14.93%	23	34.33%
12.	Tobramycin	34	50.75%	9	13.43%	24	35.82%
13.	Cefepime*	40	59.70%	4	5.97%	23	34.33%
14.	Fosfomycin*	24	82.76%	0		5	17.86%
15.	Norfloxacin*	6	20.69%	1	3.45%	22	75.86%
16.	Nitrofurantoin*	19	65.51%	2	6.89%	8	27.58%

Table 2: Antibiotic susceptibility pattern of *Proteus* species.

rates were also seen with norfloxacin (75.86%) and levofloxacin (44.77%). Imipenem showed moderate sensitivity (49.25%) with a relatively high resistance rate (41.79%).

Overall, the data indicate varying levels of effectiveness among the tested antibiotics, highlighting the importance of performing culture and sensitivity testing before selecting appropriate antimicrobial therapy.

S. No.	Antibiotic	Abbr.	Sensitivity (%)
1.	Piperacillin-Tazobactam	PTT	89.555
2.	Fosfomycin*(urine samples)	FO	82.76%
3.	Amikacin	AK	65.67%
4.	Ceftriaxone	CTR	61.19%
5.	Nitrofurantoin*(urine sample)	Nit	65.51
6.	Cefepime	CPM	59.705
7.	Cefoperazone-sulbactam	CFS	55,22%
8.	Gentamicin	GEN	53.73%
9.	Amoxicillin-clavulanic acid	AMC	52.24%
10.	Netilmicin	NET	50.75%
11.	Ciprofloxacin	CIP	49.25%
12.	Levofloxacin	LE	29.85%
13.	Norfloxacin*(urine sample)	NX	20.69%

Table 3: Antibiotic sensitivity pattern of *Proteus* species.

Table 3 shows the percentage sensitivity of different antibiotics against the tested isolates. Piperacillin–Tazobactam (89.55%) demonstrated the highest sensitivity, followed by Fosfomycin (82.76%) and Amikacin (65.67%), indicating strong effectiveness.

Among cephalosporins, Ceftriaxone (61.19%) and Cefepime (59.70%) showed moderate sensitivity. Aminoglycosides such as Gentamicin (53.73%) and Netilmicin (50.75%) also exhibited moderate activity. Fluoroquinolones like Ciprofloxacin (49.25%), Levofloxacin (29.85%), and Norfloxacin (20.69%) showed comparatively lower sensitivity rates, suggesting increased resistance. Overall, the data highlight that beta-lactam/beta-lactamase inhibitor combinations and selected urinary antibiotics appear more effective than fluoroquinolones in this study.

S. NO.	Antibiotic	Abbr.	Resistance (%)
1.	Colistin	CL	100%
2.	Norfloxacin*(urine samples)	NX	75.86%
3.	Levofloxacin	LE	44.777%
4.	Imipenem	IPM	41.77%
5.	Ciprofloxacin	CIP	37.82%
6.	Tobramycin	TOB	35.82%
7.	Cefepime	CPM	34.33%
8.	Netilmicin	NET	34.33%
9.	Cefoperazone-sulbactam	CFS	32.34%
10.	Amoxicillin-calvulanic acid	AMC	31.34%
11.	Nitrofurantoin*(urine sample)	NIT	27.58%
12.	Ceftriaxone	CTR	26.87%
13.	Gentamicin	GEN	25.37%
14.	Amikacin	AK	17.91%
15.	Fosfomycin*(urine samples)	FO	17.24%
16.	Piperacillin-Tazobactam	PIT	7.46%

Table 4: Antibiotic Resistance Pattern of *Proteus* species.

Table 4 presents the percentage resistance of different antibiotics among the tested isolates. Colistin shows the highest resistance rate (100%), indicating complete resistance in all samples. Norfloxacin (75.86%) and Levofloxacin (44.77%) also demonstrate high resistance, suggesting reduced effectiveness of fluoroquinolones.

Moderate resistance was observed with Imipenem (41.77%), Ciprofloxacin (37.82%), Tobramycin (35.82%), Cefepime (34.33%), and Netilmicin (34.33%). Beta-lactam combinations such as Cefoperazone–sulbactam (32.34%) and Amoxicillin–clavulanic acid (31.34%) also showed notable resistance levels.

Lower resistance rates were seen with Amikacin (17.91%), Fosfomycin (17.24%), and particularly Piperacillin–Tazobactam (7.46%), indicating these antibiotics may remain more effective treatment options. Overall, the data highlight significant resistance patterns and emphasize the need for culture-guided antibiotic therapy.

Discussion

In this study, 67 *Proteus* isolates were obtained from a total of 2400 clinical samples, accounting for an isolation rate of 2.79%. This finding highlights the relatively low but significant contribution of *Proteus* species as clinical pathogens, consistent with studies such as Bavitha C., *et al*. (2020) [15], who reported an isolation rate of 1.12%, and other reports showing low prevalence around 2% in clinical settings. Among the isolates, *Proteus vulgaris* (56.72%) was more prevalent than *Proteus mirabilis* (43.28%), which contrasts with most previous studies, including those by Suhartono., *et al*. (2022) [16], where *P. mirabilis* was reported as the predominant species.

The higher isolation of *P. vulgaris* in this study may reflect geographical variation, hospital-specific flora, or sampling differences. Urine (43.28%) and Pus (37.31%) samples were the most common sources, aligning with findings from previous reports where urinary tract infections and wound infections were the primary sites of *Proteus* related infections. *Proteus* infections were more common in males (61.19%) than females (38.80%), a pattern also noted by C. Bavitha., *et al*. (2020) [15]. and Suhartono., *et al*. (2022) [16].

The majority of infections occurred in elderly patients (≥61 years, 31.34%), followed by adults aged 31-50 years, highlighting increased vulnerability among older populations possibly due to comorbidities such as diabetes, urinary instrumentation, or impaired immunity. The study also indicated a significant proportion of multidrug-resistant (MDR) *Proteus* isolates, consistent with the growing global concern regarding MDR strains, especially those producing ESBL (Extended Spectrum Beta-Lactamase), AmpC β-lactamase, and carbapenemases. Prior research by Mandal., *et al*. (2015) [17] and Serry FM., *et al*. (2018) [18] have documented similar findings, reinforcing the urgent need for antimicrobial stewardship programs.

The highest number of *Proteus isolates* were obtained from urine (43.28%) and pus (37.31%) samples. This finding correlates well with the well-established role of *Proteus* species, particularly *P. mirabilis,* as important uropathogens. The data is in agreement with previous studies such as those by Treska Dh. Kamil., *et al*. (2019) [19], who found that the majority of *Proteus* isolates were recovered from urine samples, followed by wound swabs. Similarly, Bavitha., *et al*. (2020) [15], found a significant number of isolates from diabetic foot ulcers and wound infections. The increasing prevalence of antibiotic-resistant bacteria poses a major threat to public health globally, and *Proteus* species are no exception. The present study found piperacillin-Tazobactam (89.55%) to be the most effective antibiotic against *Proteus* isolates, followed by fosfomycin (82.76%) and amikacin (65.67%). This is consistent with the findings of Priya., *et al*. (2022) [20] and Mona Shaaban., *et al*. (2022) [21] where *Proteus* isolates showed high sensitivity to these antibiotics. These antibiotics continue to remain reliable treatment options for *Proteus* infections, although their effectiveness may vary based on regional prescribing habits and resistance patterns ceftriaxone (61.19%), norfloxacin (75.86%) and cefepime (59.70%) also demonstrated good efficacy, although declining trends in susceptibility suggest increasing resistance. A moderate resistance pattern was observed with gentamicin, netilmicin, and tobramycin, with sensitivity rates ranging from 50.75% to 53.73%.

The study's most alarming finding was the 100% resistance to colistin, a last-resort antibiotic often used against multidrug-resistant (MDR) gram-negative infections. This trend has been previously reported by Serry FM., *et al*. (2018) [18], who also noted complete resistance to colistin and tetracycline among *P. mirabilis* isolates. The presence of colistin-resistant *Proteus* is particularly concerning given the limited therapeutic options available for such infections. Additionally, resistance to imipenem (41.79%), norfloxacin (78.86%) indicates the emergence of carbapenem-resistant *Proteus* (CRP), likely mediated by the production of carbapenemases and alterations in porin channels. Mona Shaaban., *et al*. (2022) [21] confirmed carbapenemase production in 12.1% of *Proteus* isolates and identified co-expression of ESBL, AmpC, and carbapenemase genes such as blaVIM-1, blaSHV, and blaAmpC. These findings underline the serious threat posed by resistance gene dissemination via plasmids and other mobile genetic elements.

Conclusion

In the present study a total of 67 (2.79%) *Proteus species* were isolated from 2400 clinical samples among them *Proteus vulgaris* was the most prevalent (56.72%) followed by *Proteus mirabilis* (43.28%) the majority of isolates were recovered from and urine and pus samples with a higher infections rate observed in males

(61.19%) compared to females (38.80%) the highest incidence was noted among patients aged 61 years and above.

Antibiotic susceptibility testing revealed that piperacillin tazobactam (89.55%) fosfomycin (82.76%) amikacin (65.67%) nitrofurantoin (65.67%) and ceftriaxone (61.19%) were the most effective antibiotic against *Proteus* isolates conversely a high level of resistance was observed against colistin (100%) norfloxacin (75.86%) levofloxacin (44.77%) and imipenem (37.31%) the growing concern of multidrug resistance among *proteus* species.

This study emphasizes the need for continuous surveillance of antimicrobial susceptibility patterns prudent use antibiotic and strict infections control practices to manage infections caused by *Proteus species* effectively the findings also stress the importance of early identification and appropriate antimicrobial therapy to prevent the spread of resistant strains and to ensure better clinical outcomes.

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