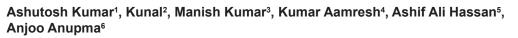
## ORIGINAL ARTICLE

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# Microbiological analysis of urinary calculi: A study from a tertiary care hospital of Eastern India



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# ABSTRACT

Background: Among various renal pathology, urolithiasis among patients of urinary tract infections (UTIs) forms a major group. The pathophysiology of such patients varies from the group of urolithiasis patients without infection. Aims and Objectives: The current study was planned and conducted to find out the bacteriological profile of urinary calculi among patients of UTI. Materials and Methods: A descriptive study was conducted by the Department of Microbiology and Surgery of JLNMC, Bhagalpur, Bihar. A total of 100 patients were enrolled in the study, having urinary calculi who either visited the outpatient department or who were admitted at the hospital for planned procedure for removal of stone between January 2021 and December 2021. Results: Male predominance was noted with a mean age of presentation at 44.3 years. 67% of the patients had some or the other symptoms of UTI, most common symptom being excruciating groin pain. Recurrent episodes of UTI were noted among female patients. Bacterial growth on culture was obtained from the urine samples of 46 patients. Escherichia coli were the most common isolated organism. Most common type of stone was a mixed type of stone formed by calcium oxalate and calcium phosphate followed by struvite stones and uric acid stones. Conclusion: Various factors of clinical significance among patients of urolithiasis and UTI are gender, age, and clinical symptoms. Gram-negative bacilli are the most common bacteria found in UTIs in patients with urolithiasis.



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**Key words:** Urolithiasis; Urinary tract infection; Gram negative bacteria; Coliform bacilli; Bacterial sensitivity tests

# **INTRODUCTION**

Urinary calculi or urolithiasis has been recognized as the most common renal morbidity globally as well as in India.<sup>1</sup> This condition dates back to 7000 years old during Egyptian mummy. Although urinary calculi are not fatal, it causes excruciating pain to the patient and requires immediate medical care.<sup>2</sup> The underlying mechanism is still not clear. It affects individuals of any age, and the incidence is reported to be one in ten individuals.<sup>3</sup> Numerous factors have been postulated to pose a risk to develop urinary calculi. To name a few, slow metabolism, dietary factors, race, gender, obstructive uropathy, and infections of urinary tract. Among these, the most frequently documented association is that of urolithiasis and urinary tract infections (UTIs). UTI plays an important role in precipitation of stone.<sup>4</sup> The reported incidence of this association varies from 3% to as high as 34% in various literatures.<sup>5</sup> Among other, the most frequent organisms encountered in the nuclei of urinary stones are Staphylococcus and *Escherichia coli*. These organisms hydrolyze urea that precipitates carbonate, bicarbonate, and ammonium ions, hence rendering urine alkaline. Stones are known to precipitate in alkaline urine in the

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form of calcium phosphate or as magnesium phosphate.<sup>6</sup> Non-urease-producing organism facilitates crystallization and formation of stone.<sup>7</sup> As bacteria are inter-wined in the spaces of stones, they evade antimicrobial action. Furthermore, antibiotic susceptibility of the microbes keeps on modifying continuously. This increase patient mortality and morbidity and also exaggerates expenses on patient care.<sup>8</sup> As far as knowledge of the author goes, there is no study documented on microbacteriological profile of ureteric stones and association with UTI from this part of the country. To fill this dearth, the current study was planned and conducted to find out the bacteriological profile of urinary calculi among patients of UTI presenting to the outpatient department or admitted in the wards of surgery.

#### Aims and objectives

The current study aimed to study patients with urinary calculi. Objectives of the study were to study the clinical presentations of these patients, perform culture and biochemical analysis of the stones derived from these patients.

#### **MATERIALS AND METHODS**

A cross-sectional descriptive study was conducted by the Department of Microbiology and Surgery of JLNMC, Bhagalpur, Bihar. A total of 100 patients were enrolled in the study, having urinary calculi who either visited the Out Patient Department or who were admitted at the hospital for planned procedure for removal of stone between January 2021 and December 2021. Duly signed informed consent forms were taken from the patients before confirming participation. Patients unfit for surgery and those who did not want biochemical analysis of their renal stone were excluded from this study. Prior ethical clearance was obtained from the Institutional Ethics Committee. A pre-designed pre-tested schedule was used to collect information from the patients that included demographic details and information on associated factors or related comorbidity.

The midstream urine specimen was collected from each participant before they underwent stone removal procedure. Post-operative stone collection was done. Colony-forming units per milliliter (CFU/mL) were identified and further processed. Species were identified. Collected stone samples were washed thoroughly using sterile water. Stone matrices were extracted from the "nidus" (nucleus) portion by scraping. The remaining part of the stone was grinded to fine powder. The powder was subjected to culture and chemical analysis. Culture was done to isolate organism and chemical analysis yielded the composition such as carbonate, calcium, magnesium, phosphate, oxalate, and uric acid. Parts from the stone samples were also inoculated in brain–heart infusion broth, incubated at 35°C for 6–8 h, inoculated onto blood, MacConkey, CLED agar (HiMedia Laboratories), and incubated at 35°C for 24 and 48 h, respectively. Bacteria isolated thus were identified by standard biochemical tests.

#### **Statistics**

Data were entered and analyzed using the Statistical Package for the Social Sciences version 24.0. Data have been described using mean with standard deviation or number and percentage. Result has been depicted in the form of text, tables, or figures.

#### **RESULTS**

#### **Demographic details**

A total of 100 patients with urinary calculi were enrolled for the study. There was a slight male predominance noted in the study population with a male-to-female ratio of 1.17:1. The mean age of male patients was 41.2 years with a standard deviation (SD) of 12.7 years, while that for the female population was older with a mean age of 46.3 years and a SD of 9.3 years (Table 1).

#### **Clinical presentation**

Nephrolithiasis was most common followed by ureterolithiasis and then urinary bladder stone (12.5%). Among comorbidities reported by the patients, hypertension (82%) was most common followed by diabetes mellitus (34%).

Symptoms of UTI were seen among 67 patients. Most common presenting complaint was excruciating groin pain that was reported by all patients. This was followed by burning micturition and increased frequency of urine. Gross hematuria was reported by seven patients, while constitutional symptom such as nausea, vomiting, and fever were complained by 27, 19, and 9 patients, respectively. History of recurrent episodes of UTI was noted for 21% of the patients, predominantly in female patients (Table 2).

#### **Culture report**

Out of 100 samples, bacterial growth on culture was obtained from the urine samples of 46 patients. Whole stone culture of 24 patients showed bacterial growth, while nidus culture was positive for 22. Bacteriological profile of patients as shown in various culture materials has been shown in Table 3.

#### **Biochemical analysis of stone**

The biochemical analysis revealed that the most common type of stone was a mixed type of stone formed by calcium

Table 1: Demographic characteristics of studypopulation			
Demographic characteristics	n		
Gender			
Male	54		
Female	46		
Age (years)			
18–30	2		
31–40	29		
41–50	36		
51–60	21		
>60	12		
Mean BMI (kg/m²)			
Males	29.6		
Females	32.1		
BMI: Body mass index			

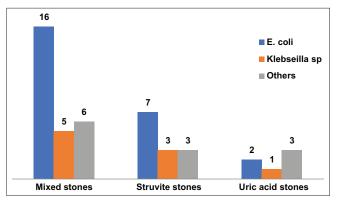
#### Table 2: Clinical characteristics of the study population **Clinical characteristics** n Symptoms 67 Groin pain Burning micturition 33 26 Increased frequency Gross hematuria 7 27 Nausea Vomiting 19 Fever 9 Previous episodes of UTI 7/54 Males 14/46 Females Co-morbidity Hypertension 82 **Diabetes mellitus** 34

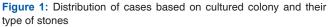
UTI: Urinary tract infection

oxalate and calcium phosphate followed by struvite stones and uric acid stones. Similarly, the majority of the culture positive urine samples were obtained from the patients with mixed stones (Figure 1 and Table 4).

# DISCUSSION

The current study was conducted to through light on the bacterial profile isolated from the urine and stone of patients diagnosed with urinary calculi. We further tried to determine the antimicrobial sensitivity pattern of the isolated culture. The burden of urinary calculi has shown many fold increase in the past few decades. The study enrolled 100 patients, majority of whom were male participants. Although UTI is known to be common among female population,<sup>2,9</sup> as the study envisages UTI in patients with diagnosed urinary stones, any of the previous researchers have documented increased high prevalence and male predominance among patients.<sup>1,10</sup> Contrary to our findings, female predominance has been reported by Koirala et al.<sup>11</sup> A new trend with more female involvement





has been noted over near past that may be attributed to change in lifestyle.<sup>12,13</sup>

Out of 100 samples, bacterial growth on culture was obtained from the urine samples of 46 patients. Worldwide, varied prevalence both higher and lower has been reported.<sup>10-13</sup> This variation is because of different antibiotic prescription patterns in various countries, and varying susceptibility toward antibiotics in different population. The most common isolated organisms were E. coli, Klebsiella sp., followed by others such Proteus sp., Enterococcus sp., Staphylococcus sp., and Pseudomonas aeruginosa. Many previous researchers have reported unanimously that the most common organism isolated from culture of the urine samples obtained from the patients with urinary calculi is E.coli.1,5,8,14 Whole stone culture of 24 patients showed bacterial growth, while nidus culture was positive for 22 samples. The bacteriological profile obtained was similar to that noted on urine culture. E. coli was most common isolated pathogen from mixed type of stones as well as from struvite stones, whereas uric acid stones have varied type of colony-forming organism. The finding of our study is contrary to the fact that E. coli is not urease-producing organism and hence is least likely to precipitate urinary stones. Still, many have documented E. coli to be very common pathogen isolated from stone culture. There are authors who have similar findings.<sup>2,8,15-17</sup>

Stone samples obtained from all the patients underwent biochemical analysis. The most common type of stone was a mixed type of stone formed by calcium oxalate and calcium phosphate, followed by struvite stones and uric acid stones. Calcium oxalate is known to be the most common compound to get precipitated as urinary stones. This pattern of biochemical composition of stone has been shown to be associated with dietary habits such as more consumption of animal protein along with high calcium and oxalate intake in the diet, particularly rice. Some have

#### Table 3: Pattern of bacteria isolated from urine and stone matrices culture

Bacteria	Sample used to inoculate culture			
	Urine (46), n (%)	Whole stone (24), n (%)	Nidus (22), n (%)	
Escherichia coli	25 (54.3)	17 (70.8)	12 (54.5)	
Enterococcus sp.	4 (8.7)	1 (4.2)	2 (9.1)	
Klebseilla sp.	9 (19.6)	2 (8.3)	4 (18.2)	
Proteus mirabilis	3 (6.5)	1 (4.2)	2 (9.1)	
Staphylococcus aureus	3 (6.5)	-	1 (4.5)	
Citrobacter koseri	1 (2.2)	-	-	
Pseudomonas aeruginosa	1 (2.2)	2 (8.3)	1 (4.5)	

#### Table 4: Distribution of stones based on their biochemical analysis and isolated organism

Type of stones that yielded	Isolated organism		
positive growth on culture (46)	Escherichia coli	Klebseilla sp.	Others
Mixed stones (27), n (%)	16 (59.3)	5 (18.5)	6 (22.2)
Struvite stones (13), n (%)	7 (53.8)	3 (23.1)	3 (23.1)
Uric acid stones (6), n (%)	2 (33.3)	1 (16.7)	3 (50.0)

documented relationship with high content of groundnuts, tomato, and spinach in the diet.<sup>17-20</sup>

#### Limitations of the study

The study enrolled a small number of patients at this level. Antibiotic sensitivity of the cultured organism can not done as a part of the current study.

#### CONCLUSION

Hence, we conclude that urinary stone disease is very common to encounter in our population with a male dominance, though females are more predisposed to get UTIs due to their anatomical structure. *E. coli* stands to be the most common organism isolated both from the urine samples and from the stone culture followed by *Klebsiella* sp. Biochemically, mixed stone of CaOx and CaPO4 is very common.

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#### Authors Contribution:

**AK-** Definition of intellectual content, Literature survey, Prepared first draft of manuscript, implementation of study protocol, data collection, data analysis, **K-** Concept, design, clinical protocol, manuscript preparation, editing, and manuscript revision; **MK-** Manuscript preparation and submission of article, Coordination and Manuscript revision. **KA-** Design of study, statistical Analysis and Interpretation; **AAH-** Review Manuscript; **AA-** Literature survey and preparation of Figures.

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