

## Research Article

# A study on positive stone culture and its association with rate of sepsis after urological procedures

M. C. Songra, Mahendra Damor, Rohit Kumar Namdev\*,  
Naveen Kumar Patbamniya, Pragye Nawalakhe, Romil Jain

Department of General Surgery, Gandhi Medical College & Associated Hamidia Hospital, Bhopal, M.P., India

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**\*Correspondence:**

Dr. Rohit Kumar Namdev,

E-mail: dr.rknamdev@gmail.com

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## ABSTRACT

**Background:** This prospective study is carried out to determine if there is a correlation between urine and/or stone cultures with postoperative sepsis in patients treated for renal and ureteral calculi. And to determine the correlation between different sites of urine sampling, including stones, and also ascertained which is more predictive of urosepsis. Correlation of urosepsis with prognosis and final outcome of patients of urolithiasis.

**Methods:** This prospective study was carried out in Department of surgery, Gandhi medical collage Bhopal and associated Hamidia hospital Bhopal between July 2013 to October 2014. All patients are divided in 4 groups according to their diagnosis.

**Results:** The incidence of renal stone was more in male (N 67) (67%) as compared with female (N 33) (33%) i.e., in the ratio of 2:1. Out of 100 patients 45 cases showed infection in preoperative urine culture, while rest 55 were sterile. Similarly 65 patients were positive for stone culture and rest 35 were sterile. 30 patients had both stone and urine culture positive. Infected urine was more common in females (66.67%). Most common bacteria isolated in urine culture were E. coli (24.44%), Klebsiella (17.77%). While correlating the result of concurrent bacteriological analysis of stones and urine culture, it is evident that same organisms from urine culture and stone culture were isolated in 8 (17.77% of positive urine culture) cases but different organisms in 26 (57.77% positive urine culture) cases. Post operatively 37% (N 37) patient show signs of SIRS out of which 17 patients had both preoperative urine and post-operative stone core culture positive. 14 Patient had only stone core culture positive, 5 pt had only urine culture positivity, and in one patient both culture were negative. Out of 37 patients who had sepsis 31 patients were positive for stone culture while 23 patients were positive for urine culture. The result was significant on chi square test (Observed P value is 0.036).

**Conclusions:** The results of this study suggest that in patients undergoing surgery for urolithiasis, stone cultures are better predictors of urosepsis than bladder urine C and S. Positive stone culture may guide clinicians regarding selection of antibiotics, especially in cases of severe urosepsis.

**Keywords:** Urolithiasis, Urosepsis, Stone culture, Prediction of urosepsis, Urine and stone culture

## INTRODUCTION

Urinary Tract Infections (UTIs) and their complications represent one of the most common causes of medical

consultation with high cost to medical services and high morbidity and mortality. The association between infection of the urinary tract and urinary calculi is well known and has been documented for many years.

Incidence of urinary tract infection in stone patients varies from 7% to 60% reported in previous studies. Infection favours the formation of urinary calculi. The predominant bacteria found in the nuclei of urinary calculi are *Staphylococcus* and *Escherichia coli*. Urea splitting organisms like *Proteus*, *Pseudomonas* & *Klebsiella* are under the urine alkaline and hence are known to promote stone formation in both clinical and experimental studies. Recurrence of urinary tract infection and stone is commonest and major health problem and it is necessary to eradicate infection. For study of aetiology and treatment of patients with urinary calculi it is necessary to perform urine and stone culture. Also it is necessary to find out the chemical composition of stone and antimicrobial sensitivity of urine and stone culture isolates. Most urea splitting bacteria lead to stone formation as hydrolysis of urea increases the carbonate, bicarbonate and ammonium ionic concentration and urinary pH. These are the required conditions for the formation of magnesium ammonium phosphate calculi which are generally staghorn. Other bacteria like *E. coli* commonly observed in urinary infection are not urea splitting.<sup>1</sup>

The combination of both clinical scenarios urinary tract infection and urinary stone is common and can trigger a Systemic Inflammatory Response Syndrome (SIRS) before, during or after medical treatment (i.e. antibiotics) and/or surgical manipulation of infected urinary stones. It is also not uncommon for patients with a sterile preoperative urine culture to develop a postoperative infection after stone removal or fragmentation. It is believed that SIRS is due to the release of bacteria & their endotoxins from infected urinary stones, developing endotoxemia, bacteremia and urosepsis. If not controlled, Multiple Organ Failure syndrome (MOF) and death of the patient may occur urosepsis due to manipulation during urological procedure can be catastrophic despite prophylactic antibiotic coverage, and negative mid-stream urine Culture and Sensitivity testing (C&S).<sup>2</sup> Stone may be infected with a different organism than that infecting the bladder and urine. Bacteria that reside within urinary calculi may differ significantly from the bacteria that are present in voided urine, thereby potentially evading the initial antibiotic coverage this is particularly important in women because the predominant bacteria in urine may be something other than what exists in the stone. The increased incidence of infection stone in females may be due to increased incidence of recurrent urinary tract infection in them which is due to close proximity of urethra to anus, short urethra and sexual activity additionally serves to increase chance of bacterial contamination of female urethra.

Perioperative urine cultures yield a high number of false-negative and false-positive results and do not accurately identify infective organisms. Low Penetration of antibiotics in the stone prevents complete eradication of urinary tract infection by conventional antibiotic therapy and thus the development of resistant organisms with

intermittent shedding in urine. Thus a vicious cycle starts, infection bringing about stone formation and stone formation causing infection.

In this prospective study we shall determine the correlation between different sites of urine sampling, including stones, and also ascertained which is more predictive of urosepsis.

## METHODS

Present study was carried out in department of surgery, Gandhi medical collage & associated Hamidia hospital Bhopal between July 2013 to October 2014.

### *Patient selection*

#### *Inclusion criteria*

1. Both male and female between age of 14 to 70 years.
2. Patients with urolithiasis who undergone stone extraction surgery.

#### *Exclusion criteria*

1. Patients < 14 years and > 70 years.
2. Patients with urogenital malignancy.
3. Patients with others causes of sepsis.
4. Severely immunocompromised patients.

In this study we analysed various culture specimens, namely bladder urine and crushed stones, and to identify that most predictive of urosepsis. We also identify other preoperatively determinable factors that might contribute to systemic infection.

Urine culture will be taken 1-2 weeks prior to scheduled surgery and 1 day or less for unscheduled surgery. All patients with negative preoperative urine culture or were treated with antibiotics for 1-7 days prior to surgery for a positive preoperative urine culture is included in study. Stone fragments will be obtain during procedure and send for culture and antibiotic sensitivity. pelvic urine sample will be collected when possible and will be send for culture & sensitivity and analysis.

The primary end point is sepsis, Systemic Inflammatory Response Syndrome (SIRS).

### *SIRS is defined as*

- Temperature <36°C or >38°C
- Heart rate >90/min
- WBC count <4000 or >12000
- Respiratory rate >20/min.

Patients who show two or more signs of SIRS are further investigated by urine c/s, blood c/s. Patient who had documented evidence of post-operative culture positivity or suspected microbiological cause of infections are considered as sepsis.

Pt who developed sepsis is treated with antibiotic vasopressor such as dopamine, phenylephrine, nor adrenaline to sustain blood pressure.

### Urine culture

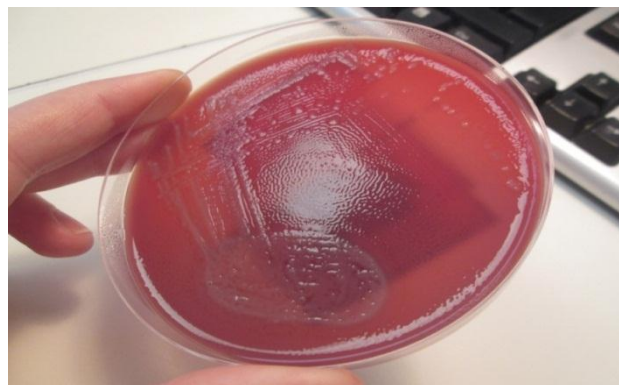
Before giving antibiotic treatment pre-operative mid-stream sample of urine was collected in sterile container after a rigorous cleaning of the external genital organs. Samples were inoculated using calibrated (4mm dia.) loop providing fixed quantity on blood agar Mac Conkeys agar. Cultures were incubated at 37°C for 24 hours. Also microscopy and Gram stain of urine samples were performed.<sup>3-10</sup> If no growth observed after 24 hours of incubation samples were considered sterile. If the colony forming units less than hundred was observed the unit was considered as significant bacteria. And if the colony forming units more than hundred was observed then, it was considered as presence of urinary tract infection.<sup>11-13</sup>

The growth from culture plate was examined and colony characters were seen. The colony was picked up and inoculated into peptone water, which was used to study the morphology, motility, biochemical test and antibiotic sensitivity. The identification of bacterial isolate was done by using standard biochemical test. Antibiotic sensitivity was done by using disc diffusion (Kirby and Baur) method.<sup>12</sup> The sensitivity of organisms to antibiotics will be studied. (Ampicillin, chloramphenicol, tetracyclin, fluoroquinolones, cephalosporin group of antibiotics, imipenem, meropenem aminoglycosides, macrolide group of antibiotics, linezolid etc.

### Stone culture

Urinary calculus was collected after the operation in a sterile container. Culture of calculus by giving 4-5 washes and finally by crushing the calculus in sterile mortar and pestle with 5ml in sterile saline. The crushed calculi core was cultured in 5 ml thioglycolate broth which was incubated at 37°C for 18-24 hours, and then subcultures were made on blood agar and Mac Conkey's agar plate for isolation of etiological agents. Cultures were incubated at 37°C for 24 hours. The growth from culture plates were examined for number of colonies from wash and crush fluids of calculus.<sup>3-6,8,14,15</sup>

The colony was picked up and inoculated into peptone water for study of morphology, motility, biochemical reactions. The identification of bacterial isolates were done by conventional methods. Also the antibiotic sensitivity of bacterial isolates was done by using disc diffusion (Kirby Baur) method.<sup>12</sup>



**Figure 1: Proteus growth on culture media.**



**Figure 2: X-ray KUB showing Staghorn renal calculi.**

## RESULTS

Present study was carried out in department of surgery, Gandhi medical collage Bhopal & associated Hamidia hospital Bhopal between July 2013 to October 2014. Total 100 cases of urolithiasis were enrolled in this study out of which 67 were male & 33 were female. All patients are divided in 4 groups according to their diagnosis. Table 1 showing distribution of patients according to their diagnosis.

**Table 1: Distribution of patients with urolithiasis.**

Type of urolithiasis	No. of patients
Renal stone	31
Pelvic stone (Staghorn)	33
Ureteric stone	15
Vesical stone	21
<b>Total</b>	<b>100</b>

The incidence of renal stone was more in male (N 67) (67%) as compared with female (N 33) (33%) i.e., in the ratio of 2.03:1.

Out of 100 patients observed 45 cases showed infection in preoperative urine culture, while rest 55 urine culture were sterile. Similarly 65 patients were positive for stone culture & rest 35 were sterile. 30 patients had both stone & urine culture positive.

**Table 2: Result of urine & stone culture.**

Specimen	Positive	Negative	The P value is 0.004474 result is significant at P <0.05
Preoperative urine culture	45	55	
Post-operative stone core culture	65	35	
<b>Total</b>	<b>100</b>	<b>100</b>	

Infected urine was more common in females (66.67% in female vs. 34.32% in male approx. ratio 2:1), however infected stone rates are almost similar in male & female (65.67% in male vs. 63.60% in female). Table 3 showing urine & stone culture positivity in male & females.

**Table 3: Urine & stone culture positivity (Male vs. Female).**

Category	Total No. of patients	Infected urine	% of infected urine	Infected stone	% of infected stone
Male	67	23	34.32%	44	65.67%
Female	33	22	66.67%	21	63.63%

Most common bacteria isolated in urine culture were E. coli (24.44%), Klebsiella (17.77%), coagulase negative staphylococcus (17.77%), pseudomonas (13.33%) & streptococcus (8.88%) etc. Table 4 showing various organism isolated from preoperative urine culture.

**Table 4: Preoperative urine culture results.**

Organism	No. of patients (N) (%)
E. coli	11 (24.44%)
Klebsiella	08 (17.77%)
Coagulase negative staph	08 (17.77%)
Pseudomonas	06 (13.33%)
Streptococcus	04 (8.88%)
Proteus	03 (6.66%)
Coagulase positive staph	03 (6.66%)
Citobacter	02 (4.44%)
<b>Total</b>	<b>45</b>

Most of the bacteria are resistant to all antibiotics (33.33%), 31.11% of these showed sensitive to carbapenem group of antibiotics, 26.67% to third generation cephalosporin, 20% to fluoroquinolons & 17.77% to aminoglycosides. Some bacteria were sensitive to more than one antibiotics. Table 5 showing pattern of antibiotic sensitivity in urine culture.

Stone culture was done in all 100 cases, out of which 65% of stone were culture positive. In 30 patients previous urine culture was positive and in remaining 35 patients previous urine culture was negative (only stone culture positive). Most common bacteria identified was

pseudomonas followed by Klebsiella, Proteus and E. coli. Table 6 showing various organism in stone culture.

**Table 5: Preoperative urine culture sensitivity.**

Antibiotics	% of organism sensitive (N)
Carbapenems	14 (31.11%)
3 <sup>rd</sup> generation cephalosporins	12 (26.67%)
Fluoroquinolons	09 (20.00%)
Aminoglycosides	08 (17.77%)
Resistance to all	15 (33.33%)

Note - Some organism were sensitive for more than one antibiotic

**Table 6: Post-operative stone culture.**

Organism	No. of patients (65) (%)
Pseudomonas	32 (49.23%)
Klebsiella	25 (38.46%)
Proteus	21 (32.30%)
E. coli	12 (18.46%)
Coagulase negative staphylococcus	02 (3.0%)
Citobacter	03 (4.6%)

Note - Some patients had more than one organism in stone culture

Most of these organisms were resistant to all antibiotic, 33% show sensitivity to carbapenem group of antibiotic, 25 to 3<sup>rd</sup> generation cephalosporins 20% to fluoroquinolons and 15% to aminoglycosides. Table 7 showing antibiotic sensitivity pattern in stone core culture.

**Table 7: Postoperative stone culture sensitivity.**

Antibiotics	No. & % of organism sensitive
Resistance to all	26 (40.00%)
Carbapenems	27 (41.53%)
3 <sup>rd</sup> generation cephalosporins	16 (24.61%)
Fluoroquinolons	13 (20.00%)
Aminoglycosides	10 (15.38%)

Note - Some organism were sensitive for more than one antibiotic

While correlating the result of concurrent bacteriological analysis of stones and urine culture, it is evident that same organisms from urine culture and stone culture were isolated in 8 (17.77% of positive urine culture) cases but different organisms in 26 (57.77% positive urine culture) cases. In 11 patients (24.44% of positive urine culture) who had infected urine, no organism was grown. Thus it



appears that voided urine does not always reflect the bacteriology of urinary tract stones.

**Table 8: Bacteriological analysis of stones and urine culture.**

No. of urine culture positive patients (N 45)	Stone culture results	Percentage
08	Same organisms in both culture	(17.77%)
26	Different organism in both culture	(57.77%)
11	Urine culture positive stone culture negative	(24.44%)

The comparison of micro-organisms isolated from crushed stones and pre-operative urine, showed that *E. coli* was the predominant micro-organism isolated from preoperative urine and *Pseudomonas* from crushed stone core culture.

Post operatively 37% (N 37) patient show signs of SIRS & evidence of UTI infection (post-operative urine culture positive). Out of which 17 patients had both preoperative urine and post-operative stone core culture positive. 14 Patient had only stone core culture positive, 5 pt. had only urine culture positivity, and in one patient both culture were negative. When we compare only 1 variable, 31 patients out of 37 patients of sepsis had positive stone culture when compared to 22 patients who had positive urine culture.

**Table 9: Comparison between different group & post-operative sepsis (SIRS).**

Group	Stone & urine culture positivity	Total No. of patients	Sepsis absent	Sepsis present (%)
I	Both urine & stone culture positive	30	13	17 (45.94%)
II	Only stone culture positive	35	21	14 (37.83%)
III	Only urine culture positive	15	10	05 (13.51%)
IV	Both culture negative	35	34	01 (02.7%)

**Table 10: Comparison between different groups.**

Groups compared	P value (Chi square test)	Result
I & IV	P <0.05	Significant
II & IV	P <0.05	Significant
III & IV	P <0.05	Significant

For comparison of various groups chi square test was used. When we compared group I (both culture positive) & group II (only stone culture positive) & group III (only urine culture positive) with group IV. We observed that chances of sepsis are more in group I & II & III when compared with group IV. P value in all three cases was <0.05 which is significant (Table 10).

From these results it is concluded that chances of sepsis is very high in a group where both cultures are positive, followed by group in which only stone culture was positive.

Isolated urine culture has less predictive value for sepsis than stone culture. Lowest chance of sepsis is observed in a group where both cultures are negative.

**Table 11: Comparison between urine & stone culture & occurrence of sepsis.**

Name of specimen	Total No. of patients	Sepsis present	Sepsis absent	P value Chi-square test
Stone culture positive	65	31	34	The P value is 0.001478 This result is significant at P <0.05.
Stone culture negative	35	06	31	
Urine culture positive	45	22	23	The P value is 0.025922 This result is significant at P <0.05.
Urine culture negative	55	15	40	

All patients who had sepsis were managed conservatively by appropriate antibiotic according to urine & stone culture. No patients developed septic shock. There was no death.

**Table 12: Culture in patients of sepsis (Total 37).**

Category	Sepsis present	Sepsis absent	
Stone culture positive	31	06	The P value is 0.03625. This result is significant at P <0.05.
Urine culture positive	23	14	

Note - 17 patients of sepsis has both urine & stone culture positive

This result is significant on chi square test. (Observed P value is 0.036).

It can conclude from above results that chances of sepsis are more in patients who had positive stone culture reports.

**Table 13: Overall comparison between type of stone, urine & stone culture & incidence of sepsis.**

Type of stone	No. of patents	Urine culture positive	Stone culture positive	No. of patient with sepsis (sirs)
Renal stone	31	12	21	11
Pelvic stone	33	12	24	12
Ureteric stone	15	05	06	05
Vesical stone	21	16	14	09
<b>Total</b>	<b>100</b>	<b>45</b>	<b>65</b>	<b>37</b>

## DISCUSSION

It has been standard practice to test urine for infection at least a week prior to urosurgery by Mid-stream urine culture & sensitivity. Patients with infection were treated with appropriate antibiotics for 7 days and then urine C&S was repeated. Antibiotics prophylaxis has been done in accordance with European Association of Urology (modified from Infectious Diseases Society of America, and European Society of Clinical Microbiology and Infectious Diseases) guidelines.<sup>17</sup> Despite this careful preoperative preparation patients still have systemic and sometimes catastrophic infection. Urosepsis and shock have been found to occur in direct proportion to the duration of the procedure, urine bacterial load, severity of obstruction by stone and infection in the stone.<sup>18</sup> O'Keefe et al. retrospectively reviewed a series of 700 patients undergoing upper tract manipulation.<sup>19</sup> Severe septicemia developed in 9 patients and 66% died. At the same center Rao et al. observed minor forms of septicemia in 37% of 27 patients undergoing PCNL.<sup>20</sup>

A stark comparison to this is series of Charton et al. of 216 patients who underwent PCNL with no prophylactic antibiotics and no major septic complications, although 35% had infected urine preoperatively.<sup>21</sup>

Our study was conducted on 100 patients of urolithiasis which include identification of causative micro-organisms from preoperative urine & crushed stone core culture. The higher incidence of renal stones in males in comparison to females may be due to increased serum testosterone level which favours increased endogenous oxalate production by liver which in turn predisposes to oxalate stone formation. Moreover, increased urinary citrate concentration in females may help in protection against calcium urolithiasis.<sup>22</sup>

In our study we found that infected urine is more common in females as compared to male (approx. ratio 2:1). However infected stone rates are almost similar (65.67% in male vs. 63.60% in female). The increased incidence of infected urine in females may be due to increased incidence of recurrent urinary tract infection in them which is due to close proximity of urethra to anus,

short urethra and sexual activity additionally serves to increase chance of bacterial contamination of female urethra. The pregnancy causes anatomical and hormonal changes that favour development of urinary tract infection. A change in genitourinary tract mucosa due to menopause may play a role in colonization of the introitus by coliforms, a major background factor for recurrent bladder infection in females.<sup>23</sup>

In our study urolithiasis was mainly observed in kidney (64%) and urinary bladder (21%) as compared to stones lodged in ureter (15%). Baron EJ, Peterson IR et al.<sup>23</sup> found that 82.97% kidney stones were found to be sterile on culture whereas in urinary bladder, infection stones were more frequent (48.57%). This could be that kidney acts as a first barrier filter for crystals thereby damaging tubular epithelium which acts as a nidus for sterile stone formation.

Recurrent urinary tract infection pre disposes to infected renal stone formation in urinary bladder due to proximity of bladder to urethra.<sup>24</sup> In contrast in our study we found 70.31% infection rate in kidney stone, whereas 66.67% in bladder stone.

Urine cultures were found positive in 45% of patients. This figure is lower than that noted by Bratell S. & Colleagues who reported 60% of patients with positive urine cultures.<sup>24</sup> It correlates well with figure reported by Bratell S. & Colleagues. The figure is more than the figure reported by Lewi H. (48%)<sup>24</sup> & Hugosson J & colleagues (45.67%).<sup>26</sup> In present study *E. coli* (24.44%) is predominant isolated organism from urine culture. It correlates well with Jennis F & co-workers, they found *E. coli* (25.7%).

In present study stone culture were found positive in 65.00% out of total 100 stones examined. Dewan B & co-workers found 47% positive stone cultures.<sup>27</sup> Those using the Nemoy and Stamey method for culturing stones have described the incidence of infected stones as 5.6% to 77.3%.<sup>16</sup> In present study *Pseudomonas aeruginosa* (49.23%) was most frequently isolated from stone culture. Gault M. H. & co-workers found more frequent isolations of *Pseudomonas*, from the stone culture.<sup>28</sup>

In a similar study of 328 patients Jairam R. Eswara & Ahmad Sharif et al. found that 3% (11/328) developed postoperative sepsis. 73% (8/11) had positive stone cultures, while none had a positive preoperative urine culture. 8% (8/96) with positive stone cultures and 1% (3/232) with negative stone cultures developed sepsis ( $P = 0.003$ ). The stone culture grew the same pathogen as the urine culture obtained on readmission in 64% (7/11) of the patients, while 9% (1/11) of preoperative urine cultures correlated with the readmission pathogen ( $P = 0.02$ ). The pathogen causing infection had a significantly higher correlation with the organism grown on stone culture than the preoperative urine culture. The patients who developed sepsis did so despite preoperative

antibiotics, and the pathogen grown on the preoperative urine culture was different from that seen post operatively. These results suggest that stone culture is more informative than preoperative urine culture for determining treatment of postoperative sepsis.<sup>29</sup>

## CONCLUSION

The results of this study suggest that in patients undergoing surgery for urolithiasis, stone cultures are better predictors of urosepsis than bladder urine C&S. Many times urine C&S may be negative or stones may be infected with different organism, in such cases positive stone culture may guide clinicians regarding selection of antibiotics, especially in cases of severe life threatening urosepsis.

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