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Prevalence of Asymptomatic bacteriuria among pregnant women in a Community Center Hospital in Shillong city, Meghalaya

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Abstract
Background: *Asymptomatic bacteriuria (ASB), a common problem in pregnancy with prevalence of 4% to 7%. Untreated asymptomatic bacteriuria (ASB) adversely effects on maternal and fetal outcome specially in a resource poor country due to lack of standard guideline that recommends screening of asymptomatic bacteriuria (ASB) among pregnant females for screening bacteriuria by Semi-quantitative culture of urine which is relatively expensive, time consuming and laborious. This study was aimed to determine the prevalence of asymptomatic bacteriuria with aim to device a single or combined rapid screening method as an acceptable alternative to urine culture. for early detection and targeted antimicrobial therapy based on anitbiogram among pregnant women attending antenatal clinic.*

Methods: *Early Morning Mid-Stream Clean Catch Urine samples were collected from 210 pregnant females, aged between 18-45 years attending antenatal clinic, for a period of 08 months (September 2020 and April 2021). Screening tests such as pus cell count, nitrite test and leukocyte esterase test of uncentrifuged urine were done using rapid dipstick kits. Identification of organisms and antibiotic sensitivity tests were performed as per standard methods.*

Results*: Out of the 210 pregnant women, 12 (5.71%) had significant bacteriuria. Among the positive cultures 62% of them were between 21-30 yrs. High percentage of asymptomatic bacteriuria was seen in 2nd trimester (51.4%) and in Primi gravida (41%). E. coli (67%) was the most common organism. The results of Direct Pus cell count showed low sensitivity of 42% but good specificity of approximately 90%. When the samples were screened by combination of Leukocyte esterase and Nitrite dip strip test, and analyzed value showed sensitivity of 83.3%, specificity of 98% with negative predictive value of 99%. Asymptomatic bacteriuria (ASB) were correctly identified in almost 100% of the cases, as single dipstick with both the parameter.*
Conclusion: *Asymptomatic bacteriuria in pregnancy can be identified by simple and combined rapid screening methods and urine culture along with antimicrobial susceptibility(AST) for early treatment to prevent complications. Strict adherence and compliance with antibiogram is of utmost importance in combating global antimicrobial resistance.*
Keywords: *Asymptomatic bacteriuria, urinary tract infection, UTI, pregnancy, pyelonephritis, drug resistance, screening for bacteriuria.*
Conflict of interest: None. Disclaimer: Nil.

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I. Introduction

Asymptomatic bacteriuria (ASB) is the presence of 1 or more species of bacteria growing in the urine at specified quantitative counts ($\geq 10^5$ colony-forming units [CFU]/mL or $\geq 10^8$ CFU/L), irrespective of the presence of pyuria, in the absence of signs or symptoms attributable to urinary tract infection (UTI). Pregnant women are at an increased risk of urinary tract infection due to their anatomic structure (short urethra) and physiological changes during pregnancy making a favourable environment for bacterial proliferation. The source of bacterial contamination is usually from the gastrointestinal tract, periurethreal or vaginal flora. During pregnancy secretions of progesterone causes smooth muscle relaxation, dilatation of the ureters and renal pelvis

which along with compression from the enlarging dextrorotated uterus leads to relative stasis of the urine (due to reduced peristalsis of the ureters) making pregnant ladies prone for asymptomatic bacteriuria (ASB). Glycosuria of pregnancy and transient/physiological decline in the immunity, also encourages the growth of both commensal and non-commensal microorganisms (1,2). Pregnancy enhances the progression from asymptomatic to symptomatic bacteriuria which could lead to pyelonephritis and adverse obstetric outcomes such as prematurity, low birth weight and higher foetal mortality rates (4). The adverse effects of undiagnosed asymptomatic bacteriuria on mother and child have made researchers to suggest routine culture screening for all pregnant women attending antenatal clinic in order to prevent mother and child from any form of complication that may arise due to infection.

Asymptomatic bacteriuria is one of the major risk factors for the development of UTIs during pregnancy which accounts for about 70% of the cases. Globally, Studies have confirmed the prevalence rate of asymptomatic bacteriuria (ASB) in pregnancy to be 2% - 11% with majority of investigators reporting it to be between 4% to 7% (2,3). But it is reported at different level in different parts of the world.

Burden of this asymptomatic bacteriuria among pregnant women in India also varies with the location of study. In India there is no guideline that recommends screening of asymptomatic bacteriuria (ASB) for the pregnant women regardless of the symptoms and sign. If untreated at the time of asymptomatic bacteriuria (ASB), it causes about 40% of cystitis (infection of bladder) and 30% pyelonephritis (infection of kidney) (4). It also affects the infants by causing premature delivery, low birth weight, intrauterine growth retardation, preterm labour, intrauterine foetal death and in general it increases prenatal mortality and morbidity (5). It is estimated that 30–50% of pregnant women without on-time treatment of asymptomatic bacteriuria (ASB) in pregnancy will progress to symptomatic urinary tract infection. Therefore, appropriate screening and treatment of asymptomatic bacteriuria (ASB) will reduce the infection rate.

However, in many hospitals in developing countries including India, routine urine culture test is not carried out for antenatal patients probably due to cost implication and time factor for culture result (usually 48 hours period) instead many clinicians opt for the strip urinalysis method for assessing urinary tract infection in pregnant women. The true picture of such urine specimen cannot be fully assessed as the strip cannot quantify the extent of infection in such a patient as well as provide antimicrobial sensitivity which is usually seen in the case of culture test. In many health centres in developing countries, the attention of clinicians and health care providers is usually on the presence of glucose and protein in urine specimens with less attention on possible asymptomatic infection. Against this background, this work was aimed to determine the prevalence of asymptomatic bacteriuria among pregnant women attending antenatal clinic in a community health centre in Shillong City, Meghalaya.

II. Aims And Objectives

AIM

This study aimed to determine the prevalence of asymptomatic bacteriuria in pregnant women attending our community centre.

Objectives

1. Early detection of the asymptomatic bacteriuria in pregnant ladies by using conventional cultural method.

2. To detect the prevalent organism and prepare an antibiogram based on culture in our study population.

III. Materials And Methods

Study Population: Pregnant women attending antenatal clinic at the centre were assessed for asymptomatic bacteriuria between September 2020 and April 2021

METHODOLOGY: -

1. Conventional Methods

- Macroscopic examination of the urine

- Urine microscopy (Pus cells)

- Urine dip stick examination (Nitrate and Leucoesterase) using Siemens Multistix

- Conventional bacteriological culture and antibiotic susceptibility test (AST)

STUDY DESIGN

Cross sectional prospective study at our community center located in the heart of Shillong city in Meghalaya state from September 2020 to April 2021.

STUDY SETTING

Early Morning Mid-Stream Clean Catch Urine after verbal and written consent in a prescribed format as per the inclusion criteria of the study

LOCATION

Community Center Hospital located in the heart of Shillong city in Meghalaya, a state in North East India.

SAMPLE SIZE: -

Sample size was calculated based on the assumptions of minimum 80% power and 5% significance level (significant at 95% confidence level). In the reference study (Asymptomatic Bacteriuria among Pregnant Women Attending Antenatal Care at Hiwot Fana Specialized University Hospital, Harar, Eastern Ethiopia: Magnitude, Associated Factors, and Antimicrobial Susceptibility Pattern), the overall prevalence of asymptomatic bacteriuria was **19.9%**. With the above mentioned assumptions and at 95% confidence level and a margin of error (confidence interval) of + 5%, the sample calculations will be **245** subjects.

Sample size calculated using the formula: - $N= Z2 * P(1-P)/\sigma 2$

where P = Prevalence; σ = Precision (Here Z = 1.96; P = 0.199; 1-P = 0.801; σ = 0.05)

Statistical analysis : -

Categorical variables was presented in number and percentage (%) and continuous variables will be presented as mean ± SD.

Statistical tests was applied as follows: -

1. Qualitative variables was compared using Chi-Square test /Fisher's exact test.

2. Sensitivity/specificity, PPV, NPV and accuracy was calculated.

3. Any other test, if applicable, at the time of analysis was considered.

Data was analysed using statistical package for social sciences (SPSS) 24.0 (SPSS Inc., Chicago, IL).

Study Protocol

The study was conducted over a period of 08 months in the Department of Lab Sciences (Microbiology) and Antenatal clinic, Department of Obstetrics and Gyneacology, at a Secondary care hospital, Shillong. All clinically relevant aseptically collected samples (Early Morning Mid-Stream Clean Catch Urine) received in the Microbiology lab were screened and processed for culture and sensitivity. A total of 210 samples aged between 18- 45 years were included in the study. Study participants were selected by a convenient sampling technique.

Ethical Consideration

The study protocol was reviewed and ethical clearance was obtained before starting the data collection process from the Research Ethics Review Committee (RERC) of the Hospital. The study participants were

informed of their right to refuse or decline participation in the study at any time and that refusing to participate in the study will not affect them. Informed voluntary, written, and signed consent was obtained from all respondents before the study. The confidentiality of participant's information was assured by excluding names and other identifiers.

Inclusion Criteria

1. All asymptomatic pregnant women

2. Participants giving verbal and written informed consent

Exclusion criteria

1. Pregnant women with a history of UTI symptoms (dysuria, frequency and urgency, etc).

2. Pregnancy induced diabetes mellitus/ hypertension.

3. History of antibiotic therapy taken in the previous two weeks, pyrexia of unknown origin, known congenital anomalies of the urinary tract, were excluded from this study.

Processing of samples

Sample collection and storage: - The study participants were explained the importance of the quality of the sample and explained in their own language about sample collection and were asked to collect 5-10 mL of early morning midstream urine by the "clean catch" method. Mid-stream clean catch urine samples collected in sterile, wide mouthed containers covered with tight-fitting lids, transported to the laboratory for analysis within 1 hrs optimally accepted (to maintain the neutrophil integrity with routine microscopy) and in case of delay in urine processing, the samples were stored at 4C. Sample from all eligible candidates were collected in sterile urine container and were processed for Urine Microscopic examination for Pus cells and Screening tests, followed by culture and antibiotic susceptibility.

Screening procedures

1. Pus cell count of the uncentrifuged urine

Pyuria is the hallmark of inflammation and also correlates with urinary tract infection. This study included the pus cell counting as a screening test and was calculated as standard method and formula. After proper mixing, 0.05 ml urine sample was transferred to the middle of the microscopic slide and cover slip was applied (22 X 22 mm) in dimensions, avoiding the trapped bubbles. The film with an excess of fluid along the edges of the cover slip with approximately 0.1 mm of depth and considering the area of HPF of 0.15 mm^2 and the volume of the urine observed in the area of 0.15 mm^3. Under these conditions the findings of one leucocyte per seven high power field corresponds with 10^4 leucocytes per ml of urine which indicates significant pyuria.

2. Leucocyte esterase test

Evidence of a host response to infection is the presence of Polymorphonuclear leucocytes in the urine. Because inflammatory cells produce Leukocyte esterase, a simple and rapid method that measures this enzyme has been developed.

3. Nitrite reductase test (Dipstick test)

It is a screening procedure that looks for the presence of urinary nitrite, an indicator of UTI. Nitrite reducing enzymes that are produced by the most common urinary tract pathogens reduce nitrate to nitrite.

The strip dipped in the well-mixed uncentrifuged urine for no longer than a second, after withdrawing the strip, excess urine along the rim of vessel was wiped. After 1 minute the colour change in the strip was compared with the colour scales provided with the kit. Pink colour produced was considered as nitrate positive and a violet colour was considered as leucocyte esterase positive. Any colour change appearing only along the edges of the test patches or developing after more than 2 minutes were considered insignificant. The manufacturer's instructions were followed.

Culture and Identification

All aseptically collected samples prior to the screening tests were subjected to culture (to avoid contamination) and antibiotic susceptibility testing. The samples were processed under aseptic condition by using standard microbiological procedures. The aseptically collected and well-mixed urine sample was inoculated on to Cysteine-Lactose-Electrolyte Deficient agar (CLED Agar), Blood Agar, and MacConkey Agar by streaking method using a standard calibrated wire loop having capacity of containing 0.001ml of urine. The inoculated plates were incubated aerobically at 37°C for 18–24 hours. Primary isolation of bacteria was made based on their colony characteristics observed on the agar plate, then the confirmed colony was counted from CLED media and multiplied by 1000 to determine the number of bacteria per ml (CFU) of the original urine specimen. A specimen was considered positive for ASB if a single organism was growing at a concentration of $\geq 10^5$ colony forming units/ml. Colony Gram stain were done to identify the organism, Lactose-fermenting yellow, moist colonies on CLED agar resembles like *E. coli* while *K. pneumoniae* produced striking mucoid colonies on the agar plate. Further Identification of bacterial organism to species level was carried out by taking an isolated colony, further inoculated onto different biochemical media for identification. Gram-negative bacteria were identified by conducting a series of Biochemical tests such as Catalase, Oxidase, Triple Sugar Iron Agar (TSI), MethyRed (MR), VogesProskauer (VP), Indole, Urease, Citrate, Triphenyltetrazolium chloride (TTC) media. Whereas Gram positive bacteria were identified using catalase and coagulase and smear Gram stain. A single isolated bacterium was inoculated onto nutrient agar slant and stored in a refrigerator after 24 hours of incubation for the maintenance of isolated bacteria.

Antimicrobial susceptibility

Antimicrobial susceptibility testing was performed using Kirby-Bauer Disc Diffusion Method as described by the Clinical Laboratory Standards Institute (CLSI). Pure isolated colony was transferred to a tube containing 5ml sterile normal saline (0.85% NaCl) and mixed gently until it formed a homogeneous suspension. The turbidity of the suspension was adjusted to the optical density equivalent to 0.5 McFarland. A sterile cotton swab was then dipped into the suspension and the excess was removed by gentle rotation of the swab against the surface of the tube. The swab was distributed evenly over the entire surface of Mueller-Hinton agar.

The inoculated plates were left at room temperature to dry for 3 to 5 minutes. The antimicrobial discs (Hi Media, Mumbai) representative of the Penicillin group (Amoxicillin 10µg, Amoxy-clavulanic acid 30µg), Cephalosporin (Cephalexin 30µg, Cefuroxime 30µg, Cefotaxime 30µg, Ceftriaxone 30µg), Aminoglycosides (Amikacin 30µg), Lincosamide (Clindamycin 2µg), Sulphonamide (Cotrimoxazole 25µg) Fluoroquinolones (Ciprofloxacin 5µg), Carbapenems (Imipenem 10µg) Nitrofurantoin (300µg), were placed on the inoculated plates and incubated at 37°C for 18– 24 hours.

The results of antibiotic sensitivity test of isolates performed on Mueller Hinton agar plates inhibition zone diameter was measured and interpreted as per the Clinical and Laboratory Standards Institute (CLSI) guidelines. For quality control of Disc Diffusion tests control strains of *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 25923 were used. Ten percent of the samples were taken to another Civil Hospital Microbiological diagnostic laboratory for quality control purposes. The results of prevalence of asymptomatic bacteriuria, isolated organisms, their antibiograms and their distribution among pregnant females are expressed as percentages. Microsoft excel was used for the interpretation of these results. For comparisons of screening tests - sensitivity, specificity, positive predictive value and negative predictive values were calculated using SPSS-24 version software (SPSS Inc., Chicago, IL).

IV. Results

Out of the total 210 pregnant women included who were screened in this study, 12 (5.71%) cases were identified by culture to have significant bacteriuria. Among the positive cultures 62% of them were between 21-30 yrs. High percentage of asymptomatic bacteriuria was seen in 2nd trimester (51.4%) and in Primi gravida (41%) (Table 1).

Table 1: Age and gestational characteristics of pregnant women screened for asymptomatic bacteriuria.

Characteristics	Number of cases (n=210)	Number of Significant bacteriuria (%) (n= 12)
Age in years		
< 20	30	2
21-30	130 **(62%)**	7

31-40 >40	38 12	2 1
Parity Primi gravida Multi gravida	86 **(41%)** 124	8 4
Gestational age 1st Trimester 2nd Trimester 3rd Trimester	85 108 **(51.4%)** 17	2 9 1

This study included evaluation of three screening tests i.e. Direct Pus cell count, combined Leukocyte esterase test and Nitrite test of uncentrifuged urine. The results of Direct Pus cell count showed low sensitivity of 42% but good specificity of approximately 90%. When the samples were screened by combination of Leukocyte esterase and Nitrite dip strip test, and analyzed value showed sensitivity of 83.3%, specificity of 98% with negative predictive value of 99%. Asymptomatic bacteriuria (ASB) were correctly identified in almost 100% of the cases, as single dipstick with both the parameter. (Table 2)

Table 2: Distribution of statistical values for various screening tests

Test	True positive	True negative	False positive	False negative
Direct wet mount of pus cells(uncentrifuged urine)	5	178	20	7
Combined Lecoesterase and Nitrite Dip Stick Test	10	194	4	2

Culture results showed the Gram negative organisms as commonest (92%) causative agents than gram positive organisms (8%) (Fig. 1). Among the Gram negative organisms, *E. coli* (67%) was the predominant organism isolated followed by *K. pneumoniae* (25%) and CoNS (Staph. saprophyticus) (8%). (Table 3 and Fig. 2)

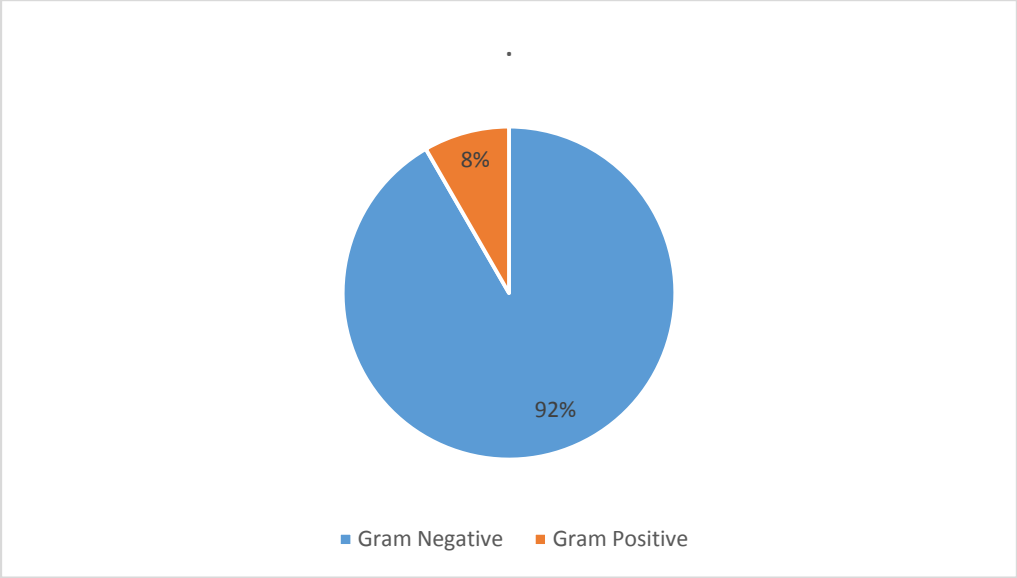

Figure 1: Percentage of Gram positive & Gram negative organisms causing asymptomatic bacteriuria

Table 3: Organisms causing asymptomatic bacteriuria expressed in percentage

Organism	No. of organism (n=12)	Percentage (100%)
E. coli	08	66.6%
K. pneumoniae	03	25%
CoNS (Staph. saprophyticus)	01	8.33%
Total	12	100%

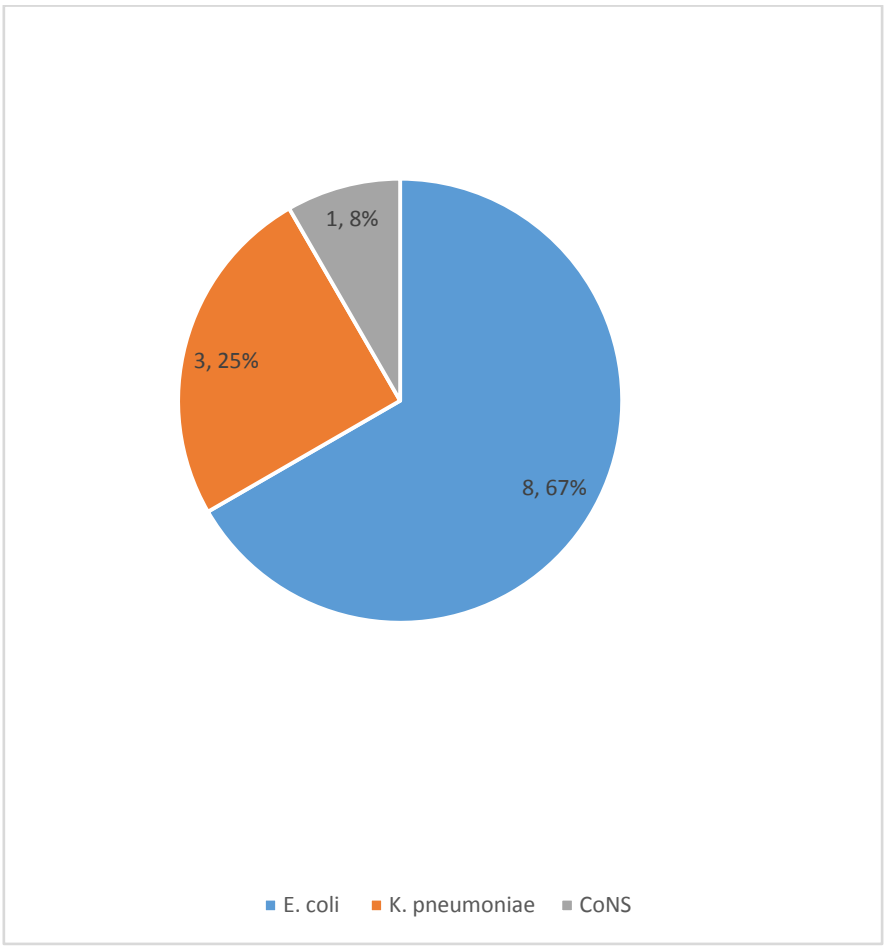

Figure 2: Relative percentage of organisms causing significant bacteriuria (n=12).

Antibiogram

The overall rate of sensitivity of the Gram negative organisms to Amoxicillin, Amoxy-clavulanic acid, Ceftriaxone, Cefuroxime, Cephalexin, Cefotaxime, Nitrofurantoin, Ciprofloxacin, Imipenem, Clindamycin, Amikacin, Cotrimoxazole were 72.7%, 100%, 100%, 54.5%, 36%, 100%, 81.8%, 18.1%, 100%, Not Done (ND), 36.3%, 81.8%, respectively (Table 4). Most of the Gram negative isolates showed good sensitivity to frequently used antibiotics with least sensitivity to Ciprofloxacin (18%), Amikacin (36%), Cephalexin (36%). *E. coli* showed the highest sensitivity to Augmentin, Ceftriaxone, Cefotaxime and Imipenem (100%), followed by Cotrimoxazole and Nitrofurantoin (91%), Amoxicillin (75%), Cephalexin (50%), Amikacin (38%), Cefuroxime (63%), and Ciprofloxacin showed the least sensitivity (16%). *Klebsiella. pneumonia*, the second most frequent organism which was grown on culture, showed almost similar sensitivity pattern as *E. coli*.

Among Gram positive isolate, identified as CoNS, (*Staph. saprophyticus*) (Catalase + / Coagulase –) and Novobiocin resistant (using Novobiocin disc (30µg), with an overall sensitivity of almost 100% sensitivity to Amoxy-clavulanic acid, Ceftriaxone, Cefuroxime, Cephalexin, Cefotaxime, Nitrofurantoin, Imipenem, Clindamycin, Amikacin, Cotrimoxazole. In this study it was observed that the isolate had comparable sensitivity to 1st and 2nd generation Cephalosporin. Isolate was found to be resistant to Amoxicillin and Ciprofloxacin in this study (Table 4).

Table 4: Pattern of antibiotic sensitivity of bacterial isolates.

Antibiotics	*E. coli* (n= 08)	*K. pneumoniae* (n=03)	CoNS (n=01)	Total sensitive isolates	Total % sensitive (n=12)
Amoxicillin	6 (75%)	2 (67%)	R	8	67%
Amoxicillin clavulanic acid	8 (100%)	3 (100%)	1 (100%)	12	100%
Ceftriaxone	08 (100%)	3 (100%)	1 (100%)	12	100%
Cefuroxime	5 (63%)	1 (33.33%)	1 (100%)	7	58%
Cephalexin	4 (50%)	R	1 (100%)	5	42%
Cefotaxime	8 (100%)	3 (100%)	1 (100%)	12	100%
Nitrofurantoin	7 (91%)	2 (66.66%)	1 (100%)	10	83%
Ciprofloxacin	2 (16%)	R	R	2	17%
Imipenam	8 (100%)	3 (100%)	1 (100%)	12	100%
Clindamycin	ND	ND	1 (100%)	1	8.33%
Amikacin	3 (38%)	1 (33.33%)	1 (100%)	5	42%
Cotrimoxazole	7 (91%)	2 (67%)	1 (100%)	10	83%

V. Discussion

Asymptomatic bacteriuria of pregnancy needs special attention, due to lack of symptoms & its adverse consequences in pregnancy. A cost evaluation study reported that screening for pyelonephritis is appropriate when the prevalence of asymptomatic bacteriuria (ASB) is greater than 2% (11,12).

The present study was conducted to know the prevalence of asymptomatic bacteriuria (ASB) using various screening methods with culture as a gold standard further preparing an antibiogram based on the sensitivity pattern of the isolates for early initiation of the targeted therapy to prevent the maternal and fetal complication.

In this study, the prevalence was observed to be higher in women belonging to 21-30 years of age group (61.90%) (7), in Primi gravida (41%) and during second trimester (51.42%) which was similar to that seen in the study of reported by Turpin et al., (7). These findings were similar to those reported in other studies done by Mukherjee K et al., where 61.9% of cases belonged to 21-30 years of age, 52.38% were Primi gravida and 42.86% of cases were in second trimester respectively (21). However, a study done in Ghana showed higher prevalence of asymptomatic bacteriuria (36.80%) in 30-34 years age group, while a study done in Hassan showed 61.77% of cases in third trimester of pregnancy.

In this study, the overall prevalence of asymptomatic bacteriuria (ASB) was 5.7%, which was almost similar to a study in Iran (6.1%) (4). Studies at Pakistan have showed a prevalence of 4.8% (5), while Jayalaxmi et al., in India showed a prevalence of 7.4% (6). But studies done in Nepal and Srilanka showed higher prevalence rates of 26%, 16.9% respectively (5,6). On the contrary, our finding was relatively high when compared with study in the antenatal care at Mbale Hospital (3.75%), Eastern Uganda and Egypt (13,14). The prevalence of asymptomatic bacteriuria in pregnant women in the world which is known to vary and the difference in findings might be attributed to differences in sample size, geographical variation, and socioeconomic condition, awareness, and predisposing factors.

This study encountered three (03) isolates with a Gram negative organisms predominance (92%), which agrees with the study report by Mukherjee K et al., (21) and from Adama and Ethiopia (72.6%) (5). This finding is supported by the fact that Gram-negative bacteria have a unique structure which assists in attachment to the uroepithelium and prevents pathogens from being flushed away by urine, allowing proliferation and tissue invasion resulting in invasive infection and pyelonephritis during pregnancy. Gram positive organism were isolated only one in number (8%). Among the Gram negative organism, *E. coli*, the most dominant isolate in this study (67%) similar in other studies done by Chandel et al., (5,6,7,8), was followed by *K. pneumoniae* (25%), Coagulase-negative staphylococci (8%) similar to Enayat et al., (5,6,7,9) reported that up to 16.8% of the causative organisms were Coagulase negative Staphylococcus (CoNS), whereas one study done in Nigeria showed *S. aureus* as the most common pathogen (72%) and E. coli being the least common (2%), (23).

In this study, *E.coli*, the most common isolate was sensitive to Cotrimoxazole, Nitrofurantoin, Amoxy-clavulanic acid, Ceftriaxone, Cefotaxime, and Imipenem. The Gram negative bacilli in this study showed less sensitivity to commonly used antibiotics ie. Amoxicillin, Cephalexin, Amikacin, Cefuroxime and Ciprofloxacin showed the least sensitivity. This is similar to what has been found in other studies (23).

Similarly, in this study the gram positive isolated as Coagulase - negative Staphylococcus (CoNS) *namely S. saprophyticus* showed sensitivity to almost all tested antibiotics (Amoxy- clavulanic acid, Ceftriaxone, Cefuroxime, Cefuroxime, Cefotaxime, Clindamycin, Amikacin, Cotrimoxazole and Imipenem. Isolates showed resistance to Amoxicillin, and Ciprofloxacin (23,24). The resistance to the antibiotics could be due overuse or misuse of these antibiotics. In this study we observed that the isolate had comparable sensitivity to 1st and 2nd generation Cephalosporin which being less frequently prescribed in this modern era, giving an antibiotic holiday may have been contributed in the sensitivity to these older antibiotics, a tool for combating global antimicrobial resistance.

Prescription of antibiotics without laboratory guidance and over the counter sales of antibiotics without prescription are a probable factor for increased bacterial resistance to antimicrobial agents (24, 25), for eg. Ampicillin and other frequently used antimicrobials are freely available in local pharmacies, and people could purchase without prescription and use them with less adherence in developing countries including India. Having said that there is an urgent need to make the antimicrobials as prescribed drugs and to implement antimicrobial stewardship program ensuring the rational usage of antimicrobials. Present study has aimed to prepare an antibiogram based on the antimicrobial sensitivity pattern further combating global antimicrobial resistance.

An ideal screening test should be simple, rapid and accurate and must identify all positive cases. Thus, a sensitive test with a high negative predictive value is desirable. In the present study, three screening tests; Pus cell count uncentrifuged urine, Combined Nitrite test and Leukocyte esterase tests were evaluated.

The pus cell count of uncentriuse urine is a very accurate method, however it showed low sensitivity of 42% in our study. The low sensitivity of pyuria observed probably due to delay in the processing leading to loos of integrity of the pus cells (6).

In the present study, Combination of two screening test values were analyzed, as single dipstick with both the parameters Combined Leukocyte esterase and Nitrite tests demonstrated sensitivity of 83.3% high specificity of 98% and Positive Predictive Value (PPV) 71.42% and negative predictive value 99%. When both nitrite and leukocyte esterase tests were positive, positive cases of asymptomatic bacteriuria were correctly identified in almost 100% of the cases. A study done by Mokube et al., (22) showed similar results compared to our study. Using combination of these two tests, all patients with infections even caused by gram-positive bacteria which would have been missed out using nitrite test alone, have been correctly diagnosed using the combined Leucoesterase and Nitrite test. Combined Leukocyte esterase and Nitrite tests may provide an acceptable alternative for screening all asymptomatic pregnant women with urine culture.

VI. Conclusion

Our study was conducted to find out the prevalence of asymptomatic bacteriuria in pregnant women, to identify the common pathogens and to prepare an antibiogram based on their susceptibility pattern. Asymptomatic bacteriuria (ASB) is diagnosed in a considerable number of pregnant women and is associated with serious complications which exacerbate maternal and neonatal morbidity. This highlights asymptomatic bacteriuria (ASB) in pregnancy as a significant problem for public health. Of the total isolated bacteria Gram negative organisms were predominant (92%) and only 8 % were the Gram positive isolate. The most frequently identified isolate was *E. coli* (67%) followed by *K. pneumoniae* (25%), Coagulase-negative staphylococci (CoNS) (8%). The Gram negative isolate were sensitive to almost all tested antibiotics i.e. Amoxy-clavulanic acid, Ceftriaxone, Cefuroxime, Cefotaxime, Cotrimoxazole and Imipenem and the same may be advised in the treatment of asymptomatic bacteriuria (ASB). The gram positive isolate showed resistance to Amoxicillin, and Ciprofloxacin and hence cannot be advised for asymptomatic bacteriuria (ASB) treatment. *E.coli*, the most common isolate was sensitive to Amoxicillin clavulanic acid Ceftriaxone, Cefotaxime, Nitrofurantoin Cotrimoxazole and Imipenem which may be used as the targeted therapy for treating asymptomatic bacteriuria (ASB). The gram negative isolates showed resistance to Ciprofloxacin and Amikacin and hence cannot be advised for asymptomatic bacteriuria (ASB) treatment in the study population. Microbiological examination remains gold standard for the isolation and culture identification of the bacteria causing asymptomatic bacteriuria (ASB) and should be performed with selection of the appropriate antibiotic based on the antimicrobial susceptibility pattern of isolated bacteria There is particular need for guidelines defining the basic principles to be followed in antibiotic treatment of asymptomatic bacteriuria (ASB) in pregnant women. We hold the opinion that it is also necessary to routinely screen for asymptomatic bacteriuria (ASB) by both rapid screening test followed by culture and susceptibility in all pregnant women at first diagnosis of pregnancy. Thus targeted antimicrobial therapy and strict adherence to antibiotic policy is of utmost importance in combating global antimicrobial resistance.

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