



PREVALENCE AND ANTIBIOTICS RESISTANCE OF BACTERIA ISOLATED FROM URINARY TRACT INFECTION OF PREGNANT WOMEN ATTENDING ISUOCHI GENERAL HOSPITAL ABIA STATE

Unegbu Nnachetam Valentine¹, Okoronkwo Christopher^{*2}, Ike Christian Chukwuemeka³, Ogwuegbu Happiness⁴ and Okakpu Lilian Obianuju⁵

^{1,3}Department of Microbiology, University of Agriculture and Environmental Sciences Umuagwo Imo State, Nigeria.

²Department of Microbiology, Abia State University Uturu, Abia State, Nigeria.

^{4,5}Department of Microbiology, Spiritan University Nneochi University Abuja, Nigeria.

Received date: 22 August 2023

Revised date: 12 September 2023

Accepted date: 02 October 2023

*Corresponding Author: Okoronkwo Christopher

Department of Microbiology, University of Agriculture and Environmental Sciences Umuagwo Imo State, Nigeria.

ABSTRACT

Background and objective: Urinary tract infection (UTI) is the most common medical complications of pregnancy. They are associated with multiplication of organisms in the urinary tract. This study is designed to investigate the prevalence and antibacterial resistance of bacteria isolated from urinary tract infection of pregnant women attending Isuochi general hospital. **Methods:** A cross-sectional study was conducted at Isuochi general hospital from September 2022 to December 2022. A semi-structured interviewer-administered questionnaire was used to collect the socio-demographic characteristics of patients. Urine specimens were collected from 80 pregnant women. Subsequent identification was done based on morphology and biochemical tests. Susceptibility pattern of the isolates were done using the disk diffusion method. **Result:** The prevalence of urinary tract infection was 54(68%). *E. coli* was the most prevalent pathogen 25(46.3%), followed by *Staphylococcus aureus* 14(26%), followed by *Klebsiella spp* 9(16.7%) and then *Pseudomonas aeruginosa* 6(11.1%) and there was statistical significance ($p=0.04$) among the organisms isolated. *S. aureus* was sensitive to levofloxacin, gentamicin, erythromycin, nitrofurantoin and streptomycin which made it the highest sensitive organism to these antibiotics, followed by *E. coli* which was sensitive to nitrofurantoin, augmentin, and ciprofloxacin. *Klebsiella spp* was sensitive to ciprofloxacin while *Pseudomonas aeruginosa* was sensitive to ciprofloxacin. **Conclusions:** The prevalence rate of UTI in pregnant women in this study was very high with *E.coli* being the most prevalent pathogen. Majority of the isolated organisms were resistant to the antibiotics used in this study. It is therefore recommended that pregnant should start attending antenatal clinic from the first trimester and that routine antibiotic susceptibility test should be performed on them to detect and eliminate drug resistant bacteria.

KEYWORDS: Urinary, Tract, Antibiotics, Prevalence, Infection, Resistance, Bacteria.

INTRODUCTON

Urinary tract infections (UTIs) are associated with multiplication of organisms in the urinary tract. Urinary tract infection (UTI) is a common bacterial infection during pregnancy and a significant cause of perinatal and maternal morbidity and mortality (Onoh *et al.*, 2013). It may be symptomatic, in form of urethritis, cystitis, pyelonephritis; or it may remain asymptomatic. Urinary Tract Infection is more common in women than in men due to their short urethra, promoting ascending infection of the bladder (cystitis) and occasionally the kidney. (Gupta *et al.*, 2011) When it affects the lower urinary

tracts, it is known as cystitis and when it affects the upper urinary tracts, it is known as pyelonephritis (Belete, 2020). Acute cystitis refers to infection of the bladder (lower urinary tract); it can occur alone or in conjunction with pyelonephritis (infection of the kidney-the upper urinary tract). (Gupta *et al.*, 2011) There has been an increasing resistance by the bacterial agent to the commonly available antibiotics (Onoh *et al.*, 2013). The prevalence of UTI is increased by several risk factors. Poor socio-economic status is reported to be a major risk factor with indigent patients having a five-fold increased risk (Joanna *et al.*, 2015). Other risk factors include age,

high parity, poor perinatal hygiene, history of recurrent UTI, diabetes mellitus, neurogenic bladder retention and anatomic bladder retention, anatomic or functional urinary tract abnormality and increased frequency of sexual activity (Joanna *et al.*, 2015). More than 90% of urinary tract infections are caused by bacteria species that are part of the normal body flora; and consequently, can readily contaminate the genital area and invade the urinary tract. *Staphylococcus aureus* has been reported to colonize the vagina in 4%-22% of pregnant women (Karina *et al.*, 2012). *Escherichia coli* are responsible for between 72% and 55% of cystitis cases in younger women, and more than 50% in women over 50 years (Derese, *et al.*, 2016). Of 500 asymptomatic pregnant women screened, 433 clinical specimens showed significant bacteriuria, representing an incidence of 86.6% (Al-Naqshbandi, *et al.*, 2019). Of this number, 38 (7.4%) were mixed bacteria colonies, while 395(91%) were of single bacteria colonies. *Staphylococcus aureus* (29.8%), *Escherichia coli* (29.1%) and *Klebsiella pneumoniae* (21.5%) were most frequently isolated pathogens (Bradley *et al.*, 2019). On the average, the pathogens were sensitive to Ciprofloxacin (99.7%), Cefazidime (81.6%), Cotrimoxazole (79.4%), Augmentin (71.4%), Nalidixic Acid (61.7%), Nitrofuradantoin (61%), Gentamicin (56.9%) and Ampicillin (25.4%) (Al-Naqshbandi, *et al.*, 2019). The positive culture rate seen in previous studies carried out in Nigeria are: 46.5% in Ebonyi, Eastern Nigeria; 35.5% in Ilorin (North Central Nigeria); 31.6% in Kano (Northern Nigeria); 32.7% in Benin (Southern Nigeria) (Onoh *et al.*, 2013). There have been reported cases of resistance to antibiotics by the UTI-causing organisms (Onoh *et al.*, 2013). Following frequent use of broad-spectrum antibiotics, the prevalence of these resistant bacteria is mainly due to widespread use of antibiotics in people and animal feeds. This study is designed to investigate the Prevalence and Antibiotic resistance of Bacteria Isolated from Urinary Tract Infection of pregnant women in Isuochi General Hospital, Abia State.

MATERIALS AND METHODS

Study Area

This study was carried out at the antenatal clinic of Isuochi General Hospital Abia State Nigeria

Study Design

This was a cross – sectional prospective study conducted from September 2022 to December 2022.

Sample size and technique

This involved eighty (80) pregnant women with UTI who attended the antenatal clinic at Isuochi General Hospital. A total of 16 pregnant women without UTI were monitored as control. A semi-structured interviewer-administered questionnaire was administered to all consenting participants in order to obtain information on their socio-demography.

Inclusion Criteria

1. Pregnant women attending Isuochi General Hospital within the study period.
2. Pregnant women between the ages 15year to 55 years.

Exclusion Criteria

1. Patient that discontents to be part of the study.
2. Pregnant women less than fourteen years and above 55 years.
3. Non-pregnant women.

Ethical clearance

Ethical permission was obtained from the hospital authorities of Isuochi General Hospital and the consent of patients were obtained and patient's confidentiality was ensured.

Specimen collection

A total of 80 clean catch midstream urine specimens were collected in sterile screw capped universal containers from consenting pregnant women attending the antenatal clinic. The women were advised to clean the genital area with lukewarm water before collecting the specimens. The sample bottle were labeled accordingly and transported to the microbiology laboratory of Spiritan University in an iced pack for processing within two hours of collection, to ensure maximum isolation of the micro-organisms.

Urinalysis

10mls of the midstream urine were transferred into a transparent test tube to check the appearance of urine. Then combi 9 medi test strip was deeped into the urine and was compared to the color chart to know the degree of changes in constituent of urine. The changes in pH, protein, glucose, ketone, blood, urobilinogen, bilirubin, nitrite, and Ascorbic acid were recorded. (CLSI, 1014).

Urine culture

A loop full (0.01 ml) of well mixed un-centrifuged urine was streaked on the surface of a dried Cysteine Lactose Electrolyte Deficient (CLED) medium and nutrient agar. These plates were incubated at 37°C for 24hours aerobically. The colony count was then determined at the end of incubation period by spreading a loopful of the culture on agar plate to get individual cells far apart enough from each other and expressed as the colony forming unit (cfu). Bacteria growth and isolated colonies were sub-cultured on a freshly prepared CLED agar incubated at 35°C for 24hours. The purified cultures were stored on nutrients agar slants and identified based on their morphology, gram staining and biochemical reaction (CLSI, 1014).

Antibiotic sensitivity test

Antimicrobial susceptibility testing was performed for bacterial isolates using disk diffusion method on Muller-Hinton agar. Briefly 3-5 colonies of the test organisms were emulsified in 5ml of nutrient broth squarely and

mixed gently. The suspension was incubated at 37° C. The antimicrobial agent impregnated disks were placed using sterile forceps on the agar surface and the plates were incubated at 37°c for 24 hours and the zone of inhibition was determined. The antimicrobial agents on the disks and their concentrations are as follows: gentamicin (GEN, 10mcg), ciprofloxacin (CPX, 10mcg), erythromycin (E, 30mcg), levofloxacin (LEV, 20mcg), streptomycin (STR, 30mcg), augmentin (AUG, 30mcg), septrin (SXT, 30mcg) and nitrofurantoin (NIT, 300mcg). The zones of inhibition were measured to the nearest millimeter using a transparent meter ruler (CLSI, 1014).

Statistical analysis

Data were analysed using Analysis of Variance (ANOVA). Discrete variables expressed as percentages

and proportions were compared using Chi-square test. Statistically significant differences were considered at value of $p \leq 0.05$.

RESULTS

Table 1 shows the socio-demographic characteristics of the study participants. The prevalence rate of UTI in this study is 54(68%). the highest prevalence of UTI of (44.4%) was seen in the age range of 26-35 years while the least was in age range of 46-55(3.7%) and this was not statistically significant ($p=0.06$). there was no statistical significance among marital status ($p=0.14$). Gestational age, education and occupation had statistical significance of ($p=0.01$, 0.001 and 0.03) respectively.

Table 1: Socio-demographic characteristics of study participants.

Variable	No. of patients (%)	percentage positive %	P-value
Age (years)			
15-25	17(21.25)	12(22.2)	0.06
26-35	36(45.00)	24(44.4)	
36-45	21(26.25)	16(29.6)	
46-55	6(7.5)	2(3.7)	
Total	80(100)	54(68)	
Marital status			
Single	19(23.75)	17(31.4)	0.14
Married	41(51.25)	27(50.0)	
Separated	11(13.75)	3(5.6)	
Widow	9(11.25)	7(13.0)	
Total	80(100)	54(68)	
Gestational age			
1 st trimester	14(17.5)	37(68.5)	0.01
2 nd trimester	39 (48.75)	2(3.7)	
3 rd trimester	27 (33.75)	15(27.7)	
Total	80(100)	54(68)	
Education			
Primary	40(50.00)	35(64.8)	0.001
Secondary	27(33.75)	12(22.2)	
Tertiary	13(16.25)	7(12.9)	
Total	80(100)	54(68)	
Occupation			
House wife	31(38.75)	24(44.4)	0.03
Employed	20(25.00)	11(20.3)	
Self-employed	29(36.25)	19(35.1)	
Total	80(100)	54(68)	

Table 2 shows the result obtained from the sample using dipstick urinalysis. The age range of 26-35 had ascorbic acid present in 2 patients, glucose in 1 patient, ketone in 2 patients, protein in 1 patients, nitrates in 15 patients

and their PH ranges from 5.0 in 10 patients, 6.0 in 16 patients, 7.0 in 4 patients and 8.0 in 5 patients. Other age range result is found in table 2. There was no significance $p=0.003$ among the parameters.

Table 2: Dipstick urinalysis results.

Age	No. tested	ASC	GLU	BIL	KET	BLD	PRO	URO	NIT	PH				p.value
										5.0	6.0	7.0	8.0	
15-25	17	0(0)	0(0)	0(0)	2(11.8)	0(0)	0(0)	0(0)	1(5.9)	2(11.8)	15(88.2)	0(0)	0(0)	0.003
26-35	36	2(5.6)	1(2.8)	0(0)	2(5.6)	0(0)	1(2.8)	0(0)	15(41.7)	10(27.8)	19(52.8)	7(19.4)	0(0)	
36-45	21	1(4.8)	6(28.6)	0(0)	0(0)	0(0)	3(14.3)	0(0)	8(38.1)	4(19.0)	8(38.1)	4(19.0)	5(23.8)	
46-55	6	0(0)	0(0)	0(0)	1(16.7)	0(0)	0(0)	0(0)	2(33.3)	3(50.0)	1(16.7)	2(33.3)	0(0)	

ASC=Ascorbic acid, GLU=Glucose, BIL=Bilirubin, KET=Ketone, BLD=Blood, PRO=Protein, URO=Urobilinogen, NIT=Nitrite

Table 3 shows the result of the percentage occurrence of bacteria isolates. *Escherichia coli* was the most prevalent

organisms, 25(46.3%), followed by *Staphylococcus aureus*, 14(26.0%), *Klebsiella* spp with 9(16.7%) and *Pseudomonas aeruginosa* as the least prevalent organisms, 6(11.1%). There was statistical significance of $p=0.04$ among the isolated organism.

Table 3: Percentage occurrence of isolated organisms from urine samples of patients.

Organisms	No. of occurrence	Percentage occurrence (%)	P- Value
<i>Escherichia coli</i>	25	46.3	0.04
<i>Staphylococcus aureus</i>	14	26.0	
<i>Klebsiella</i> spp	9	16.7	
<i>Pseudomonas aeruginosa</i>	6	11.1	
Total	54	100	

Table 4 shows the antibiotics susceptibility profile of isolates. *Staphylococcus aureus* was sensitive to levofloxacin (16), gentamicin (13), erythromycin (12), nitrofurantoin (11), septrin (13), and streptomycin (16) which made it the highest sensitive organism to these antibiotics, followed by *Escherichia coli* which was

sensitive to nitrofurantoin (22), augmentin (17), and septrin (17). *Klebsiella* spp was sensitive to nitrofurantoin (8), while *Pseudomonas aeruginosa* was sensitive to nitrofurantoin (3). *Pseudomonas aeruginosa* was resistant to erythromycin (3), levofloxacin (3) and ciprofloxacin (2).

Table 4: Antibiotics susceptibility profile of bacteria isolates.

Bacteria Isolates		S ₁ (%)	CN (%)	AU(%)	NFN(%)	ERY(%)	SXT (%)	CPX (%)	LEV.(%)
E coli (n=26)	S	17(65.4)	12(46.2)	17(65.4)	22(84.6)	0(0)	8(30.8)	4(15.4)	0(0)
	R	9(36.0)	14(56.2)	9(34.6)	4(15.4)	26(100)	18(69.2)	22(84.6)	
S. aureus 16(94.1)	S	3(17.6)	13(76.5)	0(0)	11(64.7)	12(70.6)	0(0)	13(76.5)	
	R	14(82.4)	4(23.5)	17(100)	6(35.3)	5(29.4)	17(100.0)	4(23.5)	1(5.58)
K. spp (n=9)	S	5(55.6)	2(22.2)	1(11.1)	8(88.8)	0(0)	3(33.3)	4(44.4)	0(0)
	R	4(44.4)	7(77.7)	8(88.8)	1(11.1)	9(100.0)	6(66.6)	5(55.5)	
P. aeruginosa (n=3)	S	2(66.7)	1(33.3)	0(0)	3(100)	0(0)	0(0)	1(33.3)	0(0)
	R	1(33.3)	2(66.7)	3(100.0)	0(0)	3(100.0)	3(100.0)	2(66.6)	3(100.0)
Total (n=55)									

CPX=Ciprofloxacin; CN=Gentamycin; AU=Augmentin; NFN=Nitrofurantoin; ERY=Erythromycin; SXT=Septrin; S=Streptomycin; LEV=Levofloxacin; S=Sensitive; R=Resistance, n= Number of isolate tested, no (%) = Number and the percentage of isolate susceptible/resistant to each antibiotic.

DISCUSSION

This study showed that the prevalence of UTI is 68%, which is high. The prevalent rate in this study was high compared to UTI prevalence recorded in previous studies showing pregnant women from other parts of the state recording 46.5%, in Ebonyi, Eastern Nigeria (Onoh *et al.*, 2013), and 61% in Akure, Southwest Nigeria

(Simon-Oke *et al.*, 2019). The different prevalence recorded could be attributed to UTI perception, mode of screening and other risk factors, such as age, type of toilet used, and socio-economic status of the pregnant women. However, the high prevalence of UTI in this study might be due to the lack of proper personal and environmental hygiene, population susceptibility, low socio-economic status and sexual intercourse among pregnant women (Vasudevan, 2014).

Urinary tract infections were more prevalent among women of the age category of 26-35 years. The finding differs from the research of (Ifeyanyi *et al.*, 2021) and (Kawser *et al.* 2011) which shows that UTI was more prevalent among women of 36-40 years. The high

incidence of UTI among women of ages 26-35 years maybe due to a high rate of untreated UTI history, diabetes mellitus, high blood pressure, and lack of personal hygiene which are the known risk factors of UTI prevalence. A higher incidence of UTI was found among married women. This study recorded high urinary tract infection in the gestation period of second trimester because most pregnant women start their antenatal in their second trimester. The increase in prevalence could be due to a higher number of pregnancies. Pregnant women primary educated had a higher incidence of UTI and this might be as a result of illiteracy and lack of knowledge. This study recorded high urinary tract infection among housewives which might be as a result of ignorance.

The urinalysis test result recorded a high positive result in the age category of 26-35 where 15 patients had nitrates present in their urine sample, ketone in two patients sample, ascorbic acid 1 patient's sample and glucose in 1 sample.

E. coli was the most frequently isolated common UTI pathogens. The high prevalence of *E. coli* could be that it is the most common commensal organism. Previous researchers have shown that the most prevalently isolated bacteria were *E. coli*, *S. aureus*, and *K. pneumoniae* (Azami, *et al*, 2019) and (Nwachukwu *et al*, 2018). There was statistical significance of $p=0.04$ among the isolated organisms.

The antibiotic susceptibility pattern showed that the pathogens were highly sensitive to most of the antibiotics except ciprofloxacin, erythromycin and levofloxacin which recorded high resistance. This contradicts the report of (Muhammed, 2014) who recorded high sensitivity of pathogens to ciprofloxacin.

CONCLUSION

The prevalence of UTI in this study was very high. The physiological changes of pregnancy predispose women to UTI. Urinary tract infections were more prevalent among women of the age category of 26-35 years. *E.coli* was the most frequently isolated common UTI pathogens followed by *S. aureus*, *klebsiella. spp*, and *P. aeruginosa*. The pathogens were highly sensitive to most of the antibiotics except ciprofloxacin, erythromycin, and levofloxacin which recorded high resistance.

Recommendation

It is therefore recommended that pregnant women should start attending antenatal clinic from the first trimester and that routine antibiotic susceptibility test should be performed on them to detect and eliminate drug resistant bacteria. Also, personal and environmental hygiene should be taught during antenatal visits.

REFERENCES

1. Al-Naqshbandi, A.A., Chawsheen, M.A and Abdulqader, H.H "Prevalence and antimicrobial susceptibility of bacterial pathogens isolated from urine specimens received in rizgary hospital-Erbil," *Journal of Infection and Public Health*, 2019; 12(3): 330–336.
2. Azami, M., Jaafariz, Z., Masoumi, M., Shohani, M., Badfar, G., Mahmudi, L., & Abbasalizadeh, S., The Etiology and prevalence of urinary tract infection and asymptomatic bacteriuria in pregnant women in Iran: A systemic review and Meta-Analysis. *BMC Urology*, 2019; 19: 43-45.
3. Belete, M.A. "Bacterial profile and esbl screening of urinary tract infection among asymptomatic and symptomatic pregnant women attending antenatal care of north western region," *Infection and Drug Resistance*, 2020; 13.
4. Bradley I.G., Stewardson, A.J., Abbott, I.J and Anton, Y.P. "Nitrofurantoin and fosfomycin for resistant urinary tract infections: old drugs for emerging problems," *Australian Prescriber*, 2019; 42(1): 14–19.
5. Clinical Laboratory Standards Institute (CLSI), "Performance standards for antimicrobial susceptibility testing," Twentyfourth informational supplement. CLSI document M100-S24, 2014; 34: 50–57, Clinical and Laboratory Standards Institute, Wayne, PA, USA.
6. Derese, B, Kedir, H, Teklemariam, Z, Weldegebreal, F and Balakrishnan, S. "Bacterial profile of urinary tract infection and antimicrobial susceptibility pattern among pregnant women attending at Antenatal Clinic in Dil Chora Referral Hospital, Dire Dawa, Eastern Ethiopia," *@erapeutics and Clinical Risk Management*, 2016; 12: 251–260.
7. Gupta, K., Hooton, T.M., & Naber, K.G., International clinical practice guidelines for the treatment of acute uncomplicated cystitis and pyelonephritis in women: A 2010 update of infectious Disease society of America and the European Society for microbiology and Infectious Diseases. *Clinical Infectious Diseases*, 2011; 52: 103-106.
8. Ifeanyi, A.E., Ruth, A.A., Felix, C.O., Chidimma, R.C., Ifunaya, M.O., Christie, O.O., Marian, N.U., & Emmanuel, A.E., Prevalence of urinary tract infections and associated risk factors among pregnant women in Enugu metropolis, Nigeria. *Journal of biosciences and medicines*, 2021; 9: 156-171.
9. Joanna, M., Jolanta, M., & Monika, W., Urinary tract infections in pregnancy: old and new unresolved diagnostic and therapeutic problems. *Archives of medical Science*, 2015; 11(1): 67-77.
10. Karina, A., Amanda, B., Sussan, W., Adam, J.R., & Lisa, S., Predictors of Staphylococcus aureus rectovaginal colonization in pregnant women and risk for maternal and neonatal infections. *Journal of*

- the paediatric infectious diseases society*, 2012; 1(1): 7-15.
11. Kawser, P., Afroza, M., Arzumath, A.B., & Monowara, B., Prevalence of urinary tract infection during pregnancy. *Journal of Dhaka National Medical Collection of Hospital*, 2011; 7(2): 8-12.
 12. Muhammed, M., Urinary tract infection amongst pregnant at tending a medical centre in Kaduna, Nigeria. *African Journal of Clinical and Experimental Microbiology*, 2014; 6: 7-11.
 13. Nwachukwu, E., Onyebuchi, O., & Micheal, O., Prevalence of urinary tract infection in pregnant women in Onitsha, Nigeria. *Journal of Bacteriology and Mycology*, 2018; 6(1): 284-285.
 14. Onoh, R., Umeora, O., Egwuatu, V., Ezeonu, P., & Onoh, T., Antibiotic Sensitivity Pattern of uropathogens from pregnant women with urinary tract infections in Abakaliki, *Nigeria Infection Drug Resistance*, 2013; 6(2): 225-233.
 15. Simon-Oke, I.A., & Odeyemi, O.J., Incidence of urinary tract infections and antimicrobial susceptibility patten among pregnant women in Akure, Nigeria. *Scientific Africana*, 2019; 6(26): 1-5.
 16. Vasudevan, R., Urinary tract infection: an overview of the infection and the associated risk factors. *Journal of microbiology and experimentation*, 2014; 1(2): 42-55.