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## SPECTRUM OF UROPATHOGENS AND ITS ANTIBIOTIC SUSCEPTIBILITY IN PREGNANT WOMEN WITH SYMPTOMATIC URINARY TRACT INFECTION IN A NIGERIAN TEACHING HOSPITAL. RUNNING HEADLINE: URINARY TRACT INFECTION IN PREGNANCY

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### ABSTRACT

**Background:** Urinary tract infections (UTI) are the most common bacterial infections in pregnancy and associated with maternal and perinatal morbidity and mortality.

**Objectives:** To determine the current uropathogens and their antibiotic susceptibility pattern and to compare the pregnancy outcome among clinical UTI and non clinical UTI cohorts.

**Patients and methods:** This was a prospective matched cohort study carried out between 1st January, 2012 and 30th June, 2012 at the department of Obstetrics and Gynaecology of the University of Ilorin Teaching Hospital, Ilorin, Nigeria. The study population was made up of 200 pregnant women with clinical signs and symptoms of urinary tract infections and 200 pregnant women without clinical signs and symptoms of urinary tract infection as control matched with maternal age group, parity and gestational age.

**Results:** Of 3442 obstetric patients seen 200 had clinically diagnosed UTI in pregnancy giving a rate of 5.8%. Age bracket 21- 30years and multipara had highest frequency of significant bacteriuria. Low social status and third trimester of pregnancy were identified risk factors for UTI in pregnancy. Frequency of maternal anaemia ( $p=0.02$ ) and hypertension ( $p=0.03$ ) were significantly higher among subjects than control.

The common bacterial uropathogen isolated were *Escherichia coli* (46.7%), *Staphylococcus aureus* (17.9%), *Proteus spp.*(13.3%) and *Klebsiella spp.* (11.1%). The antibiotics with highest coverage included

Co-amoxyclave (81%), Gentamicin (68.8%) and Cefuroxime (54.4%).

**Conclusion:** Maternal anaemia and hypertension were significantly higher among subjects than control. Gram negative isolates were predominant and *E. coli* was the most common isolated bacteria. Co-amoxyclave had highest coverage against the

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bacteria. Therefore, co-amoxycylave is recommended for empirical use for urinary tract infection in pregnancy in this locality.

**Keywords:** Uropathogens, urinary tract infection, pregnancy, antibiotic sensitivity.

## INTRODUCTION

Urinary Tract Infections (UTIs) are among the most common health problems during pregnancy and most common cause of admission in obstetrical wards in USA.<sup>1</sup> it occurs in 17 -20% of pregnancies hence constitutes a public health problem.<sup>2</sup>

The physiological and anatomical changes during pregnancy predispose pregnant women to developing UTIs.<sup>3-5</sup> UTI in pregnancy may be asymptomatic or symptomatic. The prevalence of asymptomatic bacteriuria in pregnancy varies between 2-10% and 30-50% of patients with asymptomatic bacteriuria will develop symptomatic infection later during pregnancy.<sup>2</sup> UTI is associated with increase maternal and perinatal morbidity namely spontaneous rupture of membrane, preterm labour and delivery, septicaemia, preterm baby and neonatal infection.<sup>2</sup>

In diagnosis of UTI, various tests have been used but urine culture remains the most reliable tool [6]. *Escherichia coli* is the most common bacterial isolate of UTI in pregnancy in many centres.<sup>4,6-8</sup>

The predominant and specific bacterial agents as well as their antibiotic sensitivity pattern vary according to time in any geographical and regional location.<sup>9-11</sup> Majority of the treatments begin or are done completely empirically, therefore, the knowledge of the organisms, their epidemiological characteristics and antimicrobial susceptibility are important to curb increasing resistance to antibiotics worldwide.

Against this background, this study is aimed at determining the current uropathogens and their antibiotic susceptibility pattern and to compare the pregnancy outcome among clinical UTI and non clinical UTI cohorts at the University of Ilorin Teaching Hospital, Ilorin, Nigeria.

## MATERIALS AND METHODS

*Study design and setting:* This prospective matched cohort study was conducted at the University of Ilorin Teaching Hospital, Ilorin, Kwara State, Nigeria.

*Study population:* A total of 200 consented pregnant women with symptoms and signs of urinary tract infections who presented at the antenatal clinic and emergency unit of the department of Obstetrics and Gynaecology, University of Ilorin Teaching Hospital, between January 1<sup>st</sup>, 2012 and June 30<sup>th</sup>, 2012, were recruited for the study. And another two hundred consented healthy pregnant women without signs and symptoms of UTI were selected as the control group matched with the subject using maternal age group, parity and gestational age through simple random sampling technique. The control group was recruited from the antenatal clinic of UITH, Ilorin.

Inclusion criteria were consenting pregnant women with signs and symptoms of UTI irrespective of the parity and gestational age who have not used antibiotics within one week prior to presentation, while the exclusion criteria included antibiotic usage by the patient within one week of presentation, presence of known

medical conditions in pregnancy and refusal to participate in the study.

Recruited pregnant women (both subjects and control) were followed up in the clinic till delivery. Those admitted and managed were also followed up at the antenatal clinic after discharge. Subjects were re-evaluated by urine microscopy, culture and sensitivity test two weeks after treatment with sensitive antibiotic. Women who delivered during the ongoing UTI had their babies evaluated for sepsis by taking swab around the navel and eye swabs for microscopy, culture and sensitivity immediately after birth. All infants were followed up for seven days after birth.

### ***Collection of Samples and Microbiological Analysis***

Ten to twenty millilitre of clean-catch midstream urine sample was collected from each patient into sterile universal bottles with screw cap tops. The specimens were delivered to the Medical Microbiology and Parasitology laboratory of the University of Ilorin Teaching Hospital, for processing within 30 minutes to 1 hour of collection. When a delay in delivery of more than 2 hours was anticipated, urine specimens were refrigerated at 4-6°C and thereafter subjected to microscopic examination, Gram staining culture and sensitivity. Pus cell equal to or greater than 10 cells per micro litre of urine was considered significant to indicate infection.<sup>12</sup>

A calibrated sterile platinum wire loop was used for the plating and it has a 1.0mm diameter designed to deliver 0.001ml of urine. A loopful of well mixed urine sample was inoculated on to plates of Cystine Lactose Electrolyte Deficient (CLED) media and Blood agar. After 24 hours of aerobic incubation of all plates at 35-37°C the

plates were examined macroscopically for bacterial growth and under the Microscope for gram reaction. Colony forming unit was determined by semi quantitative method. The bacterial colonies were counted on each plate and multiplied by 100 to give an estimate of the number of bacteria present per millilitre of urine. A significant bacteriuria was considered as any count equal to or in excess of  $10^5$  cfu/ml.<sup>13,14</sup>

The antibiotic discs that were used include Nitrofurantoin (200µg), Co-amoxyclave (10µg), Amoxicillin (10µg), Erythromycin (5µg), Gentamycin (10µg), Ciprofloxacin (5µg), Cefuroxime (30µg), Ceftriaxone (30µg), Cotrimoxazole (1.25µg+23.75µg), and Ampicillin (10µg) [15].

Zones of inhibition around each antibiotic disc were measured using a calibrated ruler and interpreted according to National Committee for Clinical Laboratory Standards (NCCLS, 1990). Zones of Inhibition of 18mm were considered sensitive, 13-17mm intermediate and <13mm resistant.<sup>9,16</sup>

### ***Sample Size Determination***

Sample size was determined using the formula  $n = z^2 pq / d^2$  [17]. Where n represents desired sample size, z is Standard normal deviation, usually set at 1.96 which corresponds to the 95% confidence interval. Letter p stands for the proportion in the target population estimated to have a particular characteristics, which is 6.2%.<sup>4</sup> in this case i.e. 0.062. and q is 1-p= 0.938. The calculated sample is 97.2 with provision for attrition (10% of the sample size). Therefore, the minimum estimated sample for the study was 100 subjects. However, 200 subjects were recruited together with 200 controls, who were matched for maternal age range, gestational age and parity.

*Ethical considerations:* Ethical approval was obtained for this study from the ethical and research committee of the University of Ilorin Teaching Hospital, Ilorin, Nigeria.

*Data analysis:* The data obtained was analyzed using the Statistical Package for the Social Sciences (SPSS), version 20. The data was presented in frequency tables and histograms. Test of significance was based on 95% confidence interval and P-value < 0.05 was considered significant.

## RESULTS

A total of two hundred clinically diagnosed cases of UTI in pregnancy were recorded between January 1<sup>st</sup>, 2012 and June 30<sup>th</sup>, 2012, out of three thousand four hundred and forty two obstetric patients seen at the department of Obstetrics and Gynaecology of UITH, Ilorin, giving a rate of 5.8%. Of the 200 subjects, 112(56%) had clinical cystitis and 88(44%) had pyelonephritis. There were 90(45%) laboratory confirmed cases of UTI in pregnancy with significant bacterial growth.

Table 1 shows the age distribution of patients with urinary tract infection in pregnancy among subjects. Over half of the pregnant women with significant bacterial growth from urine was in the age group 21-30 years, while those less than or equals to 20 years had the least frequency (4.4%). This difference was statistically significant (p=0.01).

Positive urine culture results was most common among Multiparous women 36(40.0%) followed by Nulliparous women with 30(33.3%) cases. No case of positive urine culture among grandmultiparae. The difference was statistically significant (p= 0.02). Detail is in figure 1. The highest frequency 52(57.8%) of positive urine results were in the social class IV.

Social classes I, II, III and V had 25(27.8%), 8(8.9%) and 3(3.3%) respectively. The difference was statistically significant (p=0.01). Positive urine culture result was most common 65(72.2%) among women in third trimester and least common 10(11.1%) in women in first trimester of pregnancy. However the difference was not statistically significant (p= 0.39). It is depicted in figure 2.

Table 2 shows the frequency of maternal and foetal outcome among subjects and controls. There was higher frequency of anaemia, hypertension, preterm birth, premature rupture of membrane,abruption placenta, low birth weight and perinatal death among the subjects compared to the control. But only the difference in anemia and hypertension variables among the two groups was statistically significant. There was a case of miscarriage among the control and none among subjects.

*Escherichia coli* was the most common isolated uropathogen and it accounted for 46.7% of the laboratory confirmed cases. Other organisms isolated in order of frequency were *Staphylococcus aureus* (17.9%), *Proteus species* (13.3%) *Klebsiella* (11.1%) *Pseudomonas*(4.4%) and *Candida species* (2.2%). This is depicted in figure 3.

Co-amoxyclave had the widest coverage, was effective against 81% of all the organisms isolated. Other antimicrobial sensitivity pattern in order of frequency were Gentamycin (68.8%), Cefuroxime (54.4%), Ciprofloxacin (49.8%), Ceftriaxone (28.8%), Nitrofurantoin(25.5%), Erythromycin(15.5%), Amoxicillin(5.6%) and Cotrimoxazole (4.4%). Detail in figure 4.

Repeated microscopy, culture and sensitivity carried out two weeks after treatment showed only 12 cases of candida albicans and the rest

had no significant bacterial growth. Also, eight patients delivered during the course of UTI and their babies had no significant bacterial growth from the nasal and eye swab microscopy, culture and sensitivity.

## DISCUSSION

Urinary tract infections are among the most common problems in obstetrics and constitute a public health challenge. It comprises of cystitis and pyelonephritis, which could be symptomatic or asymptomatic.

Low socio-economic status was one of the identified risk factors for urinary tract infection in pregnancy in this study. This finding is similar to Amiri et al's<sup>18</sup> and Schnarr et al's<sup>19</sup> findings.

As pregnancy advances susceptibility to UTI increases because of urethral dilatation, increased bladder volume, and decreased bladder tone which leads to urinary stasis.<sup>1,20</sup> Also risk of developing glycosuria increases, with advancing gestational age which encourages bacteria growth in the urine.<sup>20</sup> In this series, third trimester of pregnancy had the highest frequency of UTI, which is comparable to Tugrul et al's finding.<sup>21</sup> Other factors such as sexual activity, washing genitals pre-coitus, post-coitus and washing genitals from back to front are implicated in other studies<sup>1,18</sup> but were not investigated in this study.

Gram negative isolates was the predominant isolates and *E. coli* was the most common (46.7%) aetiological organism isolated in this series. This is in keeping with findings from Khartoum North Hospital, Sudan (42.4%)<sup>1</sup>, Bugando Medical centre, Mwanza, Tanzania (47.2%)<sup>22</sup>, Jhalawar, India (63%)<sup>23</sup>, Ethiopia (47.5%)<sup>20</sup> and McMaster University, Hamilton, Canada.<sup>19</sup> The other common

bacteria isolated included *Staphylococcus aureus*, *Proteus sp.*, and *Klebsiella*. Organisms from the anorectal flora are implicated in several reports to be responsible for most UTIs thereby lending credence to the theory that the lower gastrointestinal tract is the reservoir of most organisms causing UTI.<sup>4</sup> However, this is contrary to the findings of Ajayi et al's study on asymptomatic bacteriuria in pregnancy in the same study centre, where *Staphylococcus aureus* was the most common organism (72%) and *E. Coli* accounted for (4%) of cases.<sup>24</sup>

There are many drugs available to treat UTIs, but it's difficult to define the optimal antibiotic regimen. The ideal drug is expected to be active against the majority of pathogens likely to be involved, able to maintain adequate serum and tissue levels throughout the treatment, not associated with development of antimicrobial resistance, inexpensive and safe for fetus.<sup>2</sup> A major problem in the management of UTI is the emergence of drug resistant organisms. Though some resistance developed de novo as part of the evolutionary process of adaptation, majority were due to wrong choice of antibiotics or inappropriate use of the antibiotics.<sup>4</sup> Problems of antibiotic resistance are even more difficult to deal with in obstetric practice because of the limited choice of safe antibiotics in pregnancy.<sup>4</sup> This study identified Co-amoxyclave (81%) as having the highest and widest spectrum of activity against organisms responsible for UTI. Other drugs with good spectrums are Gentamycin (68.8%), Cefuroxime (54.4%) and ciprofloxacin (49.8%). Nitrofurantoin (25.5%), Ceftriaxone (28.8%), Amoxycillin (5.6%), E r y t h r o m y c i n ( 1 5 . 5 % ) a n d Cotrimoxazole (4.4%) have a limited range of effectiveness. Ampicillin didn't show any degree of effectiveness to all uropathogens

isolated in this study, probably because of its indiscriminate use among the general populace. This pattern of drug sensitivity is in contrast to the findings of Ezechi et al, where Nitrofurantoin was identified as having the highest and widest spectrum of activity against organisms responsible for UTI [4]. It is also different from the pattern of sensitivity from the urine of general populace in Ilorin described by Akanbi II et al where fluoroquinolones and third generation cephalosporins had the highest effectiveness against uropathogens [9]. However, this present study reaffirms the findings of earlier research in this centre by Abdul et al where Gentamicin was identified as being highly effective against organisms isolated from pregnant patients' urine, but its use should be restricted to situations where the benefits outweigh the risk of teratogenicity.<sup>3</sup>

Maternal anaemia and hypertension were the only complications that were significantly higher among the subjects than the control. In Mazor-Dray et al's series, maternal UTI was independently associated with pre-term delivery, pre-eclampsia, intrauterine growth restriction and caesarean delivery. But not associated with increased rates of perinatal mortality compared women without UTI.<sup>25</sup> There was no significant difference observed in preterm birth in this study, while intrauterine growth restriction and caesarean delivery were not investigated.

In conclusion, low social status and advanced gestational age are the identifiable risk factors for urinary tract infection in pregnancy. Maternal anaemia and hypertension are significantly higher among pregnant women with UTI. Gram negative isolates are predominant and *E. coli* is the most common causative organism isolated and Co-amoxycylave

has the widest coverage against the uropathogens. It is therefore recommended that Co-amoxycylave should be used empirically for urinary tract infection in pregnancy in this locality while awaiting urine culture and sensitivity result.

**Table 1:** Age Distribution of Patients with UTI in Pregnancy

Age group (years)	Subjects N(%)	Culture		Cystitis N(%)	Pyelonephritis N(%)
		Positive N(%)	Negative N(%)		
=20	9(4.5)	4(4.4)	5(4.5)	5(4.5)	4(4.5)
21-25	39(19.5)	23(25.6)	16(14.6)	29(25.9)	10(11.4)
26-30	82(19.0)	28(31.1)	54(49.1)	41(36.6)	41(46.6)
31-35	38(19.0)	15(16.6)	23(20.9)	21(18.8)	17(19.3)
36-40	24(12.0)	14(15.6)	10(9.1)	8(7.1)	16(18.2)
>40	8(4.0)	6(6.7)	2(1.8)	8(7.1)	0(0)
<b>Total</b>	<b>200(100)</b>	<b>90(45)</b>	<b>110(55)</b>	<b>112(56)</b>	<b>88(44)</b>

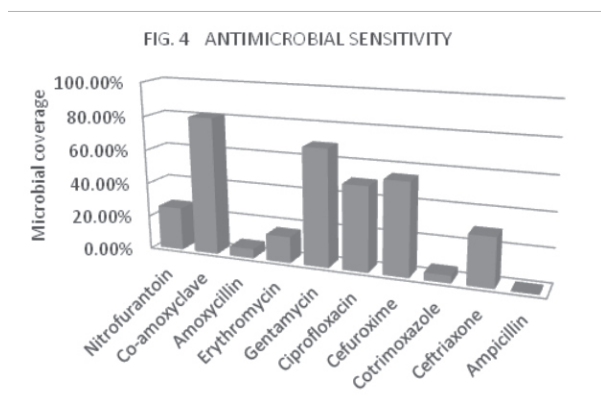
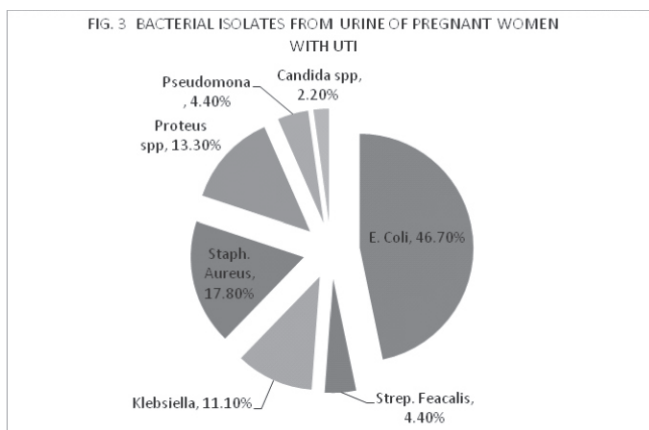
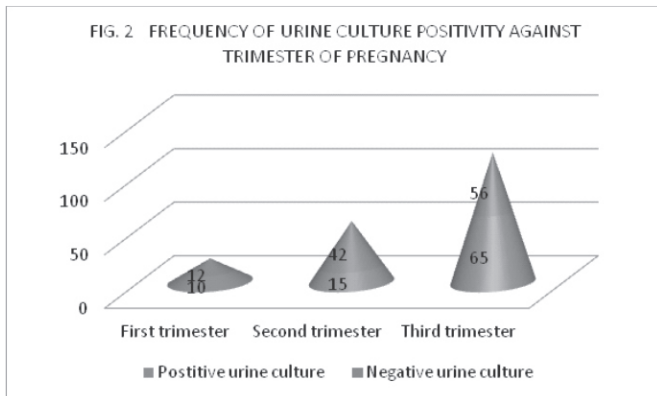
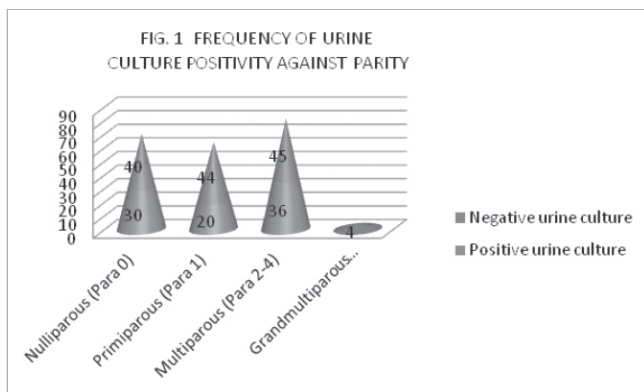
P=0.01

**Table 2:** Maternal And Foetal Outcome Among Subjects And Controls

Outcome measures	Subjects N(%)	Control N(%)	P value	Odds ratio 95% confidence interval
Anaemia	54(27)	24(12)	0.02	1.8(1.5-2.3)
Hypertension	19(9.5)	8(4)	0.03	2.4(5.6-8.3)
Renal failure	0	0	Not applicable	Not applicable
Miscarriage	0	1(0.5)	Not applicable	Not applicable
Preterm birth	8(4)	7(3.5)	0.42	16.2(-0.1-1.4)
Premature rupture of membrane	6(3)	5(2.5)	0.30	13.1(-4.9-7.9)
Abruption placenta	2(1)	1(0.5)	0.20	1.3(-3.1-4.5)
Low birth weight	12(6)	4(2)	0.92	9.3(-0.3-4.6)
Perinatal death	10(5)	4(2)	0.21	19.3(-0.8-1.8)

**Table 3:** Uropathogens and Their Antibiotic Sensitivity Pattern Among Subjects

Microbial isolates N(%)	E.Coli 42(46.7)	Streptococcus 4(4.4)	Staphylococcus 16(17.9)	Klebsiella 10(11.1)	Proteus 12(13.3)	Pseudomonas 4(4.4)	Total (%)
<b>Antibiotic sensitivity</b>							
Nitrofurantoin	11(12.2)	0	0	8(8.9)	4(4.4)	0	25.5
Co-amoxycylave	35(38.9)	4(4.4)	16(17.8)	4(4.4)	10(11.1)	4(4.4)	81
Amoxicillin	5(5.6)	0	0	0	0	0	5.6
Erythromycin	2(2.2)	0	11(12.2)	0	1(1.1)	0	15.5
Gentamicin	38(42.2)	0	12(13.3)	2(2.2)	10(11.1)	0	68.8
Ciprofloxacin	22(24.4)	4(4.4)	4(4.4)	4(4.4)	11(12.2)	0	49.8
Cefuroxime	18(20.0)	4(4.4)	11(12.2)	6(6.7)	6(6.7)	4(4.4)	54.4
Ceftriaxone	22(24.4)	0	0	2(2.2)	2(2.2)	0	28.8
Cotrimoxazole	4(4.4)	0	0	0	0	0	4.4
Ampicillin	0	0	0	0	0	0	0



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