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Urinary Pathogens amongst Parturients at the University of Port Harcourt Teaching Hospital, South-South, Nigeria

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Authors' contributions

This work was carried out in collaboration among authors. Author IFK designed the study, wrote the protocol and the first draft of the manuscript. Authors JOA and CIA did the statistical analysis and literature searches and analysis. All authors read and approved the final manuscript.

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ABSTRACT

Aim: To identify the microorganisms in urine before and after delivery. Also, to identify the incidence, risk factors and antibiotic susceptibility pattern in postpartum bacteriuria.
Methods: A quasi-interventional study of 50 women who had term vaginal deliveries at the University of Port Harcourt Teaching Hospital (UPTH) Nigeria, between September 1, 2016 and December 31, 2016. Clean catch midstream urine samples collected in sterile containers from parturients before and after delivery were processed. All significant isolates were subjected to antimicrobial susceptibility testing. Socio-demographic data and data regarding labour and risk factors were collected on a pre-designed proforma and entered on a spread sheet. Statistical analysis was done using SPSS version 22.0.
Results: Majority 49 (98%) of the parturients were booked (women that received antenatal care). Eight (16%) of the parturients had bacteriuria pre and post-delivery. Four of the parturients who were negative for bacteriuria before delivery had postpartum bacteria, giving an incidence of 8% for

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postpartum bacteriuria. The commonest micro-organism isolated post-delivery was *Escherichia coli* (50%). The number of vaginal examinations, vacuum delivery, episiotomy, perineal tear and bladder catheterization did not significantly increase the risk of postpartum bacteriuria.
Conclusion: *Escherichia coli* is the most common urinary pathogens and sensitive to Nalidixic acid. Routine screening of women for bacteriuria after delivery and an understanding of the antimicrobial sensitivity will help in reducing associated morbidities.

Keywords: Asymptomatic bacteriuria; postpartum; antimicrobial; Port Harcourt; Nigeria.

1. INTRODUCTION

Urinary tract infection (UTI) is a common clinical problem which affects the urethra, urinary bladder, ureters and kidneys. UTI affects all age groups and can be asymptomatic or symptomatic [1]. Previous studies have shown that women are more at risk than men. The incidence of asymptomatic bacteriuria during pregnancy varies from 2% to 7% [1]. The clinical significance of bacteriuria in causing upper and lower urinary tract infection during pregnancy has been well established, with about 25% of bacteriuric women developing acute symptomatic infection subsequently during the pregnancy [1]. Undetected and untreated asymptomatic bacteriuria can lead to pyelonephritis later in pregnancy or during puerperium [2]. Bacteriuria is defined as the presence of bacteria in urine. Asymptomatic urinary tract infection is when there is significant bacteriuria without obvious clinical manifestation. Significant bacteriuria is defined as a urine sample containing more than 10^5 colonies/ml of bacteria (10^8/L) in pure urine culture using a standard calibrated bacteriological loop. The anatomical proximity of the lower urinary tract, lower gastrointestinal tract and the lower genital tract exposes the urinary tract to bacteria present in the environment. The physiological changes imposed on the urinary tract by pregnancy as well as pressure on the ureters by the gravid uterus and the muscle relaxant effect of progesterone predisposes women with asymptomatic bacteriuria to urinary tract infection [1,3]. Iatrogenic procedures during labour and delivery may additionally predispose the parturient to bacteriuria [4,5].

Urine is a good medium for bacterial growth so it is not surprising that many bacteria can grow in the urinary tract and do so frequently [4,6]. The spectrum of bacteria causing complicated UTI is much broader than of those causing uncomplicated UTI. However, the most commonly encountered microorganisms globally are Gram negative bacteria including *Escherichia coli, Citrobacter* spp., *Enterobacter aerogenes, Pseudomonas aeruginosa,* and *Proteus vulgaris,* whereas *Klebsiella* spp, *Staphylococcus aureus,* and *Salmonella* spp are rarely found [7,8,9]. Increasing multidrug resistance in bacterial uropathogens is an important and emerging public health problem. The Infectious Disease Society of America (IDSA) identified some microorganisms for new effective therapies. Those microorganisms were called “ESKAPE pathogens” which include *Enterococcus faecium, S. aureus, Klebsiella* spp, *Acinetobacter* spp, *Pseudomonas* spp, and *Enterobacter* spp. Increasing drug resistance in UTI needs regular monitoring of the antibiotic susceptibility of uropathogens in a particular area [10]. Since most UTIs are treated empirically, the criteria for the selection of antimicrobial agents should be determined on the basis of the most likely pathogen and its expected resistance pattern in a geographic area. The use of urine culture as a screening measure may prove to be cost effective if bacteriuria is a common finding in the puerperium and it is clinically significant. Most studies have sought to identify pathogens in the urine of pregnant women; however, the aim of this study was to identify the microorganisms found at onset of labour and 24 hours after delivery and also to highlight the risk factors and antibiotic sensitivity pattern.

2. MATERIALS AND METHODS

2.1 Study Site

This study was carried out at the Obstetric unit of the University of Port Harcourt Teaching Hospital (UPTH) from September 1, 2016 to December 31, 2016. The UPTH is an 882-bed hospital located at Alakahia in Obio-Akpor local government area of Rivers State, South-South Nigeria. An average of 2,800 deliveries are conducted annually. It has the highest delivery rate among the health facilities in Rivers state. The unit has a total of 40 beds in the postnatal ward, 40 beds in the unbooked ward, 13 beds in the first stage room, and 8 beds in the private/semi-private rooms. There are five units, each unit has five consultant obstetricians, five

specialist senior registrars and five registrars with many experienced midwives.

2.2 Methods

The sample size was calculated using the Kish formula. All consecutive parturient who presented to the labour ward, who satisfied the eligibility criteria and consented to the study were recruited until the sample size was obtained. Parturients who withheld consent, those with prelabour rupture of fetal membranes, human immuno-deficiency virus infection, diabetes mellitus, haemoglobinopathies, contraindications to vaginal delivery, medical history of recurrent UTI not related to pregnancy, known renal pathology, on antibiotic therapy up to a week before and during labour, on immune-suppressive therapy and those who had an emergency caesarean section due to intra-partum complications were all excluded from the study.

On admission into the labour ward, a well-labeled, wide mouthed, sterile screw capped universal container was given to each participant to collect 5mls of mid-stream urine. The clean catch mid-stream urine is the preferred type of specimen because of the reduced incidence of cellular and microbial contamination. The parturient was required to first wash her hands with soap and running water and offered examination gloves, then while standing with legs astride, the labia was parted and the urethral area cleansed with a downward stroke. The first portion of urine was voided. A clean catch mid-stream urine was then collected into the sterile container. Any excess urine was voided. Care was taken to avoid touching the rim of the container. Trained midwives supervised the sample collection. The specimen was promptly transported to the Department of Medical Microbiology and Parasitology, University of Port Harcourt Teaching Hospital, for immediate processing. The sample was processed within two hours of collection. Macroscopic examination was done to assess the appearance of the urine sample. Thereafter, the colour-coded portion of the dip stick was immersed into the urine sample for 5 seconds. The dipstick was removed and left for 20 seconds for the reactions to occur and then compared against the chromatic scale within 2 minutes. Microscopy was done by placing one drop of un-centrifuged urine on a clean grease pre-labeled frosted slide and covered with a cover slip. It was examined under a microscope for white blood cells, red blood cells, yeast cells and various types of casts. This procedure was repeated using sediment obtained from the 5 mls of centrifuged urine. The urine was then cultured using a calibrated sterile loop (that can hold 1/500 ml i.e. 0.002 ml of urine) to collect a loop full of urine and inoculated into the pre-labeled sterile culture media; Cystine lactose electrolyte deficient agar and blood agar and incubated aerobically at 35ºC to 37ºC overnight. All isolates that are considered significant using the interpretation criteria were identified using standard biochemical methods. The number of isolated colonies (colony forming units) on the medium was counted using a counting chamber and then multiplied by a factor of 500 to estimate significant bacteriuria. A count more than 10^5 per ml of urine was taken as significant bacteriuria; less than 10^4 per ml was taken as not significant while counts between 10^4–10^5 per ml was considered doubtful and the urine samples was re-examined. Antimicrobial sensitivity was done by Disc diffusion test. A disc of blotting paper impregnated with a known volume and appropriate concentration of an antimicrobial agent was placed on a sensitivity test agar (Mueller Hinton) inoculated with the test organism. The preparation of the inoculums was done using a broth (peptone water). A swab stick was used to spread the broth containing the organisms evenly. The medium was incubated overnight and the zone of inhibition measured around each disc. The growth of the organism was inhibited up to a distance from the disc where the concentration of the drug is roughly equal to its minimum inhibitory concentration, the zone of inhibition. The zone of inhibition was compared with that of a control isolate for each antibiotic.

Isolates were classified as either sensitive or resistant based on the definition of the Clinical and Laboratory Standard Institute. An isolate was considered multi-drug resistant if it was resistant to at least three of the antibiotics.

Each patient was assigned a special number which was used to collate results on a spread sheet. A second mid-stream urine sample was collected 24 hours after delivery by the same method described above and subjected to the same tests. Patients with positive urine culture were treated with appropriate antibiotics and a test of cure repeated after two weeks.

2.3 Statistical Analysis

Statistical package SPSS 22 was used for data analysis. The results are presented as mean,

standard deviation, and percentages. Paired t test was used to compare differences in means pre- and post-delivery while independent t test was used to compare the difference in mean between women with postpartum bacteriuria and those without it. Chi-square test and Fisher's exact test were used to compare the differences in proportions. The comparison of the differences in bacteriuria across the pair (pre- and post-delivery) was done using Mcnemar chi square test. Statistical significance was set at $P < 0.05$.

3. RESULTS

Sixty women met the inclusion criteria and were recruited into the study. Three parturients were subsequently withdrawn due to emergency caesarean sections carried out for obstetric indications, two had received antibiotics in the course of labour and 5 parturients had insufficient samples. Therefore, only 50 of the women's samples were suitable for analysis. The mean age of respondents was 30.82±5.04 years, median parity was 2 and the mean gestational age was 38.32±1.00 weeks. Majority (72%) had tertiary level of education, 54% of the participants were Multiparas. About half (46%) of the women were unemployed. This is shown in Table 1. Table 2 shows the relationship between socio-demographic characteristics of the parturients and postpartum bacteriuria. There was a significant association between employment status and postpartum bacteriuria (p value = 0.021). None of the obstetric characteristics was significantly associated with postpartum bacteriuria (p>0.05). Eight (16%) of the 50 parturients had bacteriuria both pre and post-delivery. A total of 12 women had bacteriuria in the postpartum period. Four of the paturients whose samples were negative on admission (pre-delivery) acquired bacteriuria during labour, giving an incidence of postpartum bacteriuria of 8%. However, the difference in occurrence of bacteriuria across the pair (pre- and post-delivery) was not statistically significant (p value =0.074). *Escherichia coli* (*E. coli*) *and Staphylococcus aureus* (*S. aureus*) were the most common (37.5%) organisms isolated in the pre-delivery samples while *E. coli* alone was the commonest in the post-delivery samples. Combining all the isolates in both pre- and post-delivery, *E. coli* was the commonly identified organism with a frequency of 45% while the least common isolates were *Klebsiella spp.* (10.0%) and *Pseudomonas spp.* (10.0%). There was no statistically significant difference in the distribution of isolates before and after delivery (p value =1.000). This is shown in Table 3. Bacterial uropathogens isolated from the urine samples of the parturients revealed high levels of single and multiple antimicrobial resistances against commonly prescribed drugs considered safe in pregnancy. *E coli* which was the predominant organism isolated was resistant to Cefuroxime and Augmentin but sensitive to all other agents, whereas, *S. aureus* which was the second most common organism was resistant to Augmentin, Cefuroxime and Ceftriaxone. Nalidixic acid was the only antimicrobial sensitive to all the identified isolate in the study as shown in Table 4. Test of cure done two weeks later in patients with positive urine culture, who were treated with appropriate antibiotics, showed no growth. Vacuum delivery, episiotomy, perineal tear and bladder catheterization did not significantly increase the risk of acquiring postpartum bacteriuria. The mean number of vaginal examinations was higher among those with postpartum bacteriuria but this difference was not statistically significant. This is shown in Table 5.

Table 1. Socio-demographic characteristics

Variables	Frequency	%
Age		
15– 24 years	5	10.0
25 – 34 years	34	68.0
35 – 44 years	11	22.0
Parity		
Para 0	10	20.0
Para 1	13	26.0
Para2 and above	27	54.0
Educational level		
Primary	1	2.0
Secondary	13	26.0
Tertiary	36	72.0
Employment status		
Unemployed	23	46.0
Employed	27	54.0
Booking status		
Unbooked	1	2.0
Booked	49	98.0

4. DISCUSSION

Bacterial infection of the urinary tract is one of the common reasons for seeking medical attention [11]. Effective management of patients with bacteriuria depends on the identification of the type of organism(s) that caused the disease and the selection of an effective antibiotic agent [12]. The incidence of postpartum bacteriuria was 8%. This is similar to that of previous studies

[4,13,14]. In contrast, the retrospective cohort study done by Gundersen, et al. showed that 4.6% of women with intended caesarean delivery and 3.5% of women with intended vaginal delivery were treated for postpartum urinary tract infection [15].

Although, mid-stream catch and not supra-pubic aspiration was employed in this study, the urine sample collection was done to minimize contamination. The screening of women for bacteriuria before and after delivery in this study showed a slightly higher rate of bacteriuria in

Table 2. Socio-demographic/Obstetric characteristics and postpartum bacteriuria

Variables	Postpartum bacteriuria				
	Yes n (%)	No n (%)	Total n (%)	Statistical test	P value
Age					
15– 24 years	1 (20.0)	4 (80.0)	5 (100.0)	1.941**	0.513
25 – 34 years	2 (5.9)	32 (94.1)	34 (100.0)		
35 – 44 years	1 (9.1)	10 (90.0)	11 (100.0)		
Parity					
Para 0	1 (10.0)	9 (90.0)	10 (100.0)	0.529**	1.000
Para 1	1 (7.7)	12 (92.3)	13 (100.0)		
Para 2 and above	2 (7.4)	25 (92.6)	27 (100.0)		
Educational level					
Primary	0 (0.0)	1 (100.0)	1 (100.0)	2.033**	0.597
Secondary	0 (0.0)	13 (100.0)	13 (100.0)		
Tertiary	4 (11.1)	32 (88.9)	36 (100.0)		
Employment status					
Employed	0 (0.0)	30 (100.0)	30 (100.0)	**	0.021*
Unemployed	4 (20.0)	16 (80.0)	20 (100.0)		
Booking status					
Booked	4 (8.2)	45 (91.8)	49 (100.0)	**	1.000
Unbooked	0 (0.0)	1 (100.0)	1 (100.0)		

**Statistically significant; **Fishers exact test*

Table 3. Distribution of isolates pre and post-delivery

Isolate	Pre-delivery	Post-delivery	Total
Escherichia coli	3 (37.5)	6 (50.0)	9 (45.0)
Staph aureus	3 (37.5)	4 (33.4)	7 (35.0)
Pseudomonas spp.	1 (12.5)	1 (8.3)	2 (10.0)
Klebsiella spp.	1 (12.5)	1 (8.3)	2 (10.0)
Total	**8 (100.0)**	**12 (100.0)**	**20 (100.0)**

Fisher's exact= 0.977; p value = 1.000

Table 4. Antimicrobial susceptibility pattern of identified isolates

Isolates	Antimicrobial sensitivity					
	CXM	NIT	NA	AUG	CRO	CIP*
Escherichia coli (n=9)	0 (0.0%)	9 (100.0%)	9 (100.0%)	0 (0.0%)	4 (44.4%)	9 (100.0%)
Staph aureus (n=7)	0 (0.0%)	7 (100.0%)	1 (14.3%)	0 (0.0%)	0 (0.0%)	7 (100.0%)
Klebsiella spp. (n=2)	0 (0.0%)	0 (0.0%)	2 (100.0%)	0 (0.0%)	0 (0.0%)	2 (100.0%)
Pseudomonas spp. (n=2)	0 (0.0%)	0 (0.0%)	2 (100.0%)	0 (0.0%)	0 (0.0%)	2 (100.0%)
Total (n=20)	0 (0.0%)	16 (80.0%)	14 (70.0%)	0 (0.0%)	4 (20.0%)	20 (100.0%)

**Antimicrobial used as control ; n=number of isolates*

CXM-Cefuroxime; NIT-Nitrofurantoin; NA-Nalidixic acid; AUG-Augmentin; CRO- Ceftriaxone; CIP-Ciprofloxacin

Table 5. Intrapartum risk factors and postpartum bacteriuria

Variables	Postpartum bacteriuria				
	Yes n (%)	No n (%)	Totaln (%)	Statistical test	P value
Vacuum delivery					
Yes	0 (0.0)	3 (100.0)	3 (100.0)	**	1.000
No	4 (8.5)	43 (91.5)	47 (100.0)		
Episiotomy					
Yes	2 (13.2)	13 (86.7)	15 (100.0)	**	0.574
No	2 (5.7)	33 (94.3)	35 (100.0)		
Perineal tear					
Yes	1 (5.6)	17 (94.4)	18 (100.0)	**	1.000
No	3 (9.4)	29 (90.6)	32 (100.0)		
Bladder catherization					
Done	3 (8.8)	31 (91.2)	34 (100.0)	**	1.000
Not done	1 (6.2)	15 (93.8)	16 (100.0)		

***Fishers exact test*

comparison to other studies [13,15]. This therefore highlights the need for routine screening for bacteriuria in postpartum period.

The major aetiological agent causing bacteriuria is *E. coli,* this is in keeping with other studies [16,17]. This could be explained by the fact that *E. coli* can persist in the urinary tract as reservoir for recurrent infections [18]. This result is consistent with reports from other studies [1,2,19,20,21] but differs from a report that showed *S. aureus* as the most predominant organism in bacteriuria [22]. Some researchers found *Enterococcus fecalis*, *S. aureus* and *Klebsiella spp* to be the second most predominant isolates [4,23]. These show the worldwide variations in the pattern of urinary tract infection.

The findings of *E. coli*, *S. aureus* and *Klebsiella spp* all being sensitive to nalidixic acid, and all resistant to cefuroxime in the present study is consistent with other studies [16,24]. Whereas *S. aureus* and *Klebsiella spp* were both resistant to ceftriaxone, a third generation cephalosporin. Resistance to the third generation cephalosporins may infer that many of the organisms are extended-spectrum betalactamases (ESBLs) producers [25,26]. Consequently, Nitrofurantoin has been found to be an effective alternative in such cases of ESBLs [27]. The other possible explanation for this resistance could be that the third generation cephalosporins have been abused overtime. Hence, the organisms have developed resistant mechanisms [28].

Concerning the risk factors for postpartum bacteriuria, some studies found that urethral catheterization significantly increased the rate of bacteriuria in vaginal deliveries [29,30]. However, this was not the case in this study.

This may be because not all the patients were catheterized and the few patients catheterized were done aseptically. This could explain the absence of significant association between catheterization and bacteriuria in present study.

Operative delivery, number of vaginal examinations and epidural anesthesia did not increase the risk of postpartum bacteriuria. This is in contrast with previous reports which showed that the number of vaginal examinations performed, duration of epidural analgesia as well as vacuum delivery correlated significantly with the risk of postpartum bacteriuria [4]. The strength of this study lies in it being a prospective study, identifying the rate of bacteriuria before and after delivery, the causative organisms and the antimicrobial profile in our environment. However, it had some limitations because it is a hospital-based study with a small sample size, which may not be reflective of the general population.

5. CONCLUSION

Variation in pathogens comparative in child birth was obvious in the face of multiplicity of risk factors. This is more so in the index study. The incidence of postpartum bacteria is high (1:12 women). *E. coli* is the most prevalent causative organism as seen in this research. *S. aureus* was the second most prevalent. There was no significant difference in the pattern of isolates before and after delivery. Nalidixic acid plays a major role in the treatment of postpartum

bacteriuria. The organisms isolated were all resistant to the third generation cephalosporins. This study did not identify any risk factor with significant correlation to the risk of acquiring postpartum bacteriuria.

CONSENT

As per international standard, patient's written consent has been collected and preserved by the author(s).

ETHICAL APPROVAL

All authors hereby declare that the study was approved by the ethical review board of the hospital and was carried out in accordance with the ethical standards laid down in the 1964 Helsinki declaration.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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