

Original Research Article

Antimicrobial profile and Asymptomatic Urinary Tract Infections among Pregnant Women
Attending Antenatal Clinic in Bolgatanga Regional Hospital, Ghana

ABSTRACT

Background: Urinary tract infection (UTI) in pregnancy is associated with significant morbidity for both the mother and the baby. Proper investigation and prompt treatment are needed to prevent serious life-threatening condition and morbidity associated with UTI in pregnant women. **Aim:** This study was designed to detect common uropathogens and their antibiotic susceptibility pattern among asymptomatic pregnant women attending antenatal care in the Bolgatanga Regional Hospital. **Methodology:** Mid-stream urine samples were collected from 200 individuals and inoculated onto cysteine lactose electrolyte deficient (CLED) agar media. Colony counts yielding bacterial growth of $\geq 10^5$ CFU /ml was regarded as significant bacteriuria. Pure isolates of bacterial pathogen were characterized by colony morphology, Gram-stain and standard biochemical procedures. Kirby Bauer disc diffusion method was used for antimicrobial susceptibility testing of all identified isolates. **Results:** The overall prevalence of bacteria-associated asymptomatic UTI was 17.5%. *Escherichia coli* (42.9%) was the most isolated organism followed by *Staphylococcus aureus* (34.3%), *Klebsiella pneumoniae* (11.4%), *Staphylococcus saprophyticoccus* (5.7%) and *Proteus mirabilis* 2 (5.7%). Yeast cells and *Schistosoma haematobium* were also recorded in 2% of the women. Isolates showed significant sensitivity to commercially prepared antibiotic discs. However, higher level of resistance was recorded with tetracycline, nitrofurantoin and nalidixic Acid. **Conclusion:** Early screening for UTI should be done for all pregnant women and those found to be infected need to be treated with appropriate antimicrobial agents to avoid complications.

26 **Key words**

27 Urinary tract infection, Bacteriuria, Pregnant women, Antimicrobial resistance

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31 **1 INTRODUCTION**

32 Urinary tract infection (UTI) is an infection caused by the presence and growth of
33 microorganisms anywhere in the urinary tract. The proximity of the vagina to the anus more
34 frequently predisposes females to UTI's due to easy contamination of the urinary tract with fecal
35 flora [1]. More so, the presence of short urethra, absence of prostatic secretion, and pregnancies
36 are few factors deemed to foster rapid progression of UTI's in females [2].

37 Pregnancy is a special condition presented with marked changes in the anatomical and
38 physiological make-up of the urinary tract. During pregnancy, there is a profound alteration in
39 hormonal levels and physiological and anatomical changes in the female urinary tract. Changes
40 during pregnancy are usually marked by glucosuria, gradual dilatation of renal pelvis and ureters
41 in the eighth week, increased levels of estrogen and progesterone, and also the displacement of
42 the bladder superiorly and anteriorly [3, 4]. The combination of anatomical, hormonal and
43 physiological changes during pregnancy contributes to significant changes in the urinary tract,
44 which has a profound impact on the acquisition of bacteriuria during pregnancy [5].

45 UTI infections usually present as asymptomatic or symptomatic. Nonetheless, both
46 asymptomatic or symptomatic could potentially cause considerable morbidity or mortality if not
47 detected early and treated. In symptomatic pregnant women, urinary tract infection may manifest
48 as preterm delivery, low birth weight, pre-eclampsia toxemia, pyelonephritis, anaemia, chronic
49 renal failure and foetal mortality [6]. Diagnosis and treatment of UTI's should ideally be prompt

50 and accurate especially in this era where there are increasing reports of antimicrobial resistance
51 worldwide. Unfortunately, in many developing countries management of UTIs is usually
52 empirical, and exhaustive screening in pregnancy is not considered as an essential part of
53 antenatal care. This study is designed to detect common uropathogens and their antibiotic
54 susceptibility pattern in pregnant women.

55 **2 METHOD**

56 **2.1 Study population and sample collection**

57 This study included urine samples from 200 pregnant women attending antenatal clinic at the
58 Bolgatanga Regional Hospital in the Upper East region of Ghana. The age of the participants
59 ranged from 15 to 46 years. Information on parity and gestational age were extracted from the
60 antenatal records of pregnant women. Other information was obtained using a well-structured
61 questionnaire. Clean-catch midstream urine specimens were collected. The participants were
62 instructed on how to collect the urine sample by using leak-proof, wide-mouth sterile plastic
63 containers. The urine specimens were then delivered to the laboratory and processed within one
64 hour. Pregnant women who were on antibiotic treatment two weeks prior to their initial visit, those
65 who exhibited clinical signs and symptoms of UTI and those at 38 weeks of gestation or more were
66 excluded from the study.

67 **2.2 Bacteria culture and identification**

68 Using sterile calibrated wire inoculating loop, approximately 0.01ml urine samples were
69 inoculated onto cysteine lactose electrolyte deficient (CLED) agar plate. Cultures were incubated
70 under an aerobic condition at 37°C for 24 hrs. Colonies were counted to check the presence of
71 significant bacteriuria. Colony count yielding bacterial growth of 10^5 CFU/ml of urine was
72 regarded as significant bacteriuria [7]. All positive cultures with significant bacteriuria were

73 then identified by their colony characteristics, gram-staining reaction and standard biochemical
74 protocol [8].

75 **2.3 Urine Chemistry, Microscopic and Macroscopic Examination**

76 A volume of about 6ml of well-mixed urine sample was centrifuged at 3000 rpm for 10 minutes
77 and the supernatant was subsequently dispensed into a clean sterile tube. The supernatant was
78 tested against combi 10 urine reagent strips (URIT Medical electronics, China). The sediment
79 obtained after centrifugation was mixed and a drop was placed on a microscopic slide for
80 examination using a compound light microscope for the presence of parasites, pus cells,
81 epithelial cells, cast and crystals.

82 **2.4 Antimicrobial susceptibility testing**

83 Kirby Bauer disc diffusion method was used for antimicrobial susceptibility testing of all
84 identified isolates following the Clinical and Laboratory Standards Institute protocol [9].
85 Commercially available antibiotic discs including Cefoperazone (CPZ), Ciprofloxacin (CIP),
86 Ceftriaxone (CTR), Piperacillin (PIT), Cefotaxime (CTX), Tetracycline (TET), Levofloxacin
87 (LEV), Nitrofurantoin (NIT), Gentamicin (GEN), Norfloxacin (NX), Amikacin (AMK) and
88 Nalidixic Acid (NA) were used for the antimicrobial susceptibility testing. *Escherichia coli*
89 ATCC 25299 was used as a quality control strain. The diameter of the zone of inhibition was
90 measured as the index for isolate susceptibility.

91 **2.5 Ethical Consideration**

92 All experiments have been examined and approved by the appropriate ethics committee and have
93 therefore been performed in accordance with the ethical standards laid down in the 1964
94 Declaration of Helsinki

95 **2.6 Data analysis**

96 Socio-demographic, clinical and laboratory data were entered and analyzed using IBM SPSS
97 version 20. Chi-square analysis was used to compare all categorical data. Fisher's exact analysis
98 was used for cells with an expected count less than 5. An alpha level of 0.05 was used for all
99 statistical tests.

100 **3 RESULTS**

101 A total of 200 pregnant women were included in the study. The age of the study participants
102 ranged from 19 years to 45 years. Notably, most of the participants (57.5%) had only primary
103 education. Anaemia was more prevalent 31(15.5%) among the study participants. Details on the
104 baseline characteristics of the study participants are provided in Table 1.

105 **3.1 Urine Chemistry, Microscopic and Macroscopic Examination**

106 The appearances of the urine samples examined during this study were as follows; clear
107 171(85.5), cloudy 7 (3.5%), hazy 22 (11%) amber 14(7%) and 186(93%) straw in colour.
108 Majority of the participant with urine pH less than 7 were 169 (80.5%) and those ≥ 7 were 59
109 (29.5%). The study showed that 169 (84.5%) of the subjects' urine had specific gravity < 1.025 .
110 Further, the urine of 7(3.5%) participants was positive for haematuria. There were 19 (9.5%)
111 positive cases of leucocytes esterase and 3 (1.5%) leucocyte esterase trace. In addition, positives
112 for protein were 2 (1%), 9 (4.5%) for trace and 189(94.5%) were negative for proteins.
113 Moreover, glucosuria was found in 5 (2.5%) of the participants whereas 3 (1.5%) of the
114 participants were positives for ketones. Microscopic examination showed 4 (2%) were infected
115 with *Schistosoma haematobium* and varying proportions of epithelial cells, crystals and cast
116 (Table 2). Notably, of the "cast and crystal" identified, the proportion of calcium oxalate was
117 relatively high.

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122 **Table 1 Baseline characteristics of study participants in the Bolgatanga Regional Hospital**

VARIABLE	NUMBER(n=200)	PERCENTAGE (%)
<i>Parity</i>		
Primigrividae	59	29.5
Pausigravidae	26	13
Multigravidae	115	57.5
<i>Marital Status</i>		
Single	4	2.5
Married	196	97.5
<i>Religion</i>		
Muslim	25	12.5
Christian	175	87.5
<i>UTI In</i>		
<i>Previous Pregnancy</i>		
Yes	37	18.5
No	163	81.5
<i>Place of Treatment</i>		
Hospital	21	56.8
Others	16	43.2
<i>Awareness of UTI</i>		
Yes	84	42
No	116	58
<i>Knowledge of Mode of Infection</i>		
Sexual Contact	40	20
Uncleanness	10	5
No Idea	150	75
<i>Education</i>		
Primary	115	57.5
Secondary	9	4.5
Tertiary	49	24.5
Vocational Skills	5	2.5
Illiterate	21	10.5
<i>Complications in Previous Pregnancy</i>		
Anaemia	31	15.5
Eclampsia	1	0.5
Hypertension	6	3
Preterm Delivery	2	1

BMI(Kg/M2)

Lean (<20)	23	11.5
Normal (20-24.9)	107	53.5
Overweight (25-29.9)	43	21.5
Obese (≥ 30)	27	13.5

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124 **3.2 Prevalence of bacteria-associated urinary tract infection**

125 The results showed that of the 200 samples cultured, 165 (82.5%) had insignificant bacterial
 126 growth while 35 (17.5%) showed significant bacterial growth. Of the total recovered isolates were
 127 15 (42.9%) *Escherichia coli*, 4 (11.4%) *Klebsiella pneumoniae*, 12 (34.3%) *Staphylococcus*
 128 *aureus*, 2(5.7%) *Staphylococcus saprophyticus* and 2 (5.7%) *Proteus mirabilis*. This study reports
 129 an overall prevalence of bacterial urinary tract infection of 17.5% (35/200) among pregnant
 130 women attending a clinic in Bolgatanga regional hospital.

131 **Table 2. Microscopic examination of urine samples recorded**

URINE DEPOSITES	COUNT (%)
Pus Cells <10	189(94.5)
Pus Cells ≥ 10	11(5.5)
Epithelial Cells <10	150(75)
Epithelial Cells ≥ 10	50(25)
Crystals & Casts	
Calcium Oxalate	10(5)
Calcium Phosphate	1(0.5)
Hyaline Cast	1(0.5)
Triple Phosphate	1(0.5)
Parasites	
Yeast cells	4(2)
<i>Schistosoma haematobium</i>	4(2)

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133 **3.3 The distribution of uropathogens among pregnant women**

134 There was evidence of an association between *E. coli* and *S. aureus* infection and parity ($p < 0.05$).

135 Overall, there was evidence of an association between uropathogen infection and parity among
136 pregnant women. Table 3 depicts the distribution of uropathogens among pregnant women
137 stratified into age, parity, and gestational age.

UNDER PEER REVIEW

Table 3. The distribution of uropathogens among pregnant women stratified into age group, parity and gestational age in the Bolgatanga Regional Hospital

ISOLATE	Age			P-value	Parity			P-value	Gestational age			P-value
	(≤19) N=19	(20-30) N=122	(>30) N=59		Primigravidae N=59	Paucigravidae N=26	Multigravidae N=115		1 ST Trimester N=35	2 ND Trimester N=0	3 RD Trimester N=0	
<i>E. Coli</i>	3(15.8)	9(7.4)	3(10.0)	0.304	5(8.5)	7(26.9)	3(2.6)	<0.05	15(42.9)	0(0.0)	0(0.0)	-
<i>Klebsiella pneumoniae</i>	0(0.0)	4(3.3)	0(0.0)	0.535	2(3.4)	1(3.8)	1(0.9)	0.240	4(11.4)	0(0.0)	0(0.0)	-
<i>Proteus mirabilis</i>	0(0.0)	2(1.6)	0(0.0)	1.000	0(0.0)	1(3.8)	1(0.9)	0.130	2(5.7)	0(0.0)	0(0.0)	-
<i>S. aureus</i>	2(10.5)	5(4.1)	5(8.5)	0.259	3(5.1)	9(34.6)	0(0.0)	<0.05	12(34.3)	0(0.0)	0(0.0)	-
<i>S. saproprophyticus</i>	0(0.0)	2(1.6)	0(0.0)	1.000	0(0.0)	1(3.8)	1(0.9)	0.330	2(5.7)	0(0.0)	0(0.0)	-
TOTAL INFECTION	5(26.3)	22(18.0)	8(13.6)	0.435	10(16.9)	19(73.1)	6(5.2)	<0.05	35(100)	0(0.0)	0(0.0)	-

Data is presented in count (percentage)

3.4 Antimicrobial susceptibility testing

Evaluation of the susceptibility pattern of the organisms to the antibiotics showed Amikacin was the most effective, recording 94.3% susceptibility. Ceftriaxone and ciprofloxacin were also active against the organisms, recording 91.4% and 85.7% susceptibility respectively. On the other hand, tetracycline and nalidixic Acid were the least effective antimicrobial agents recording 20% susceptibility each. Details on the antimicrobial susceptibility pattern are provided in Figure 1.

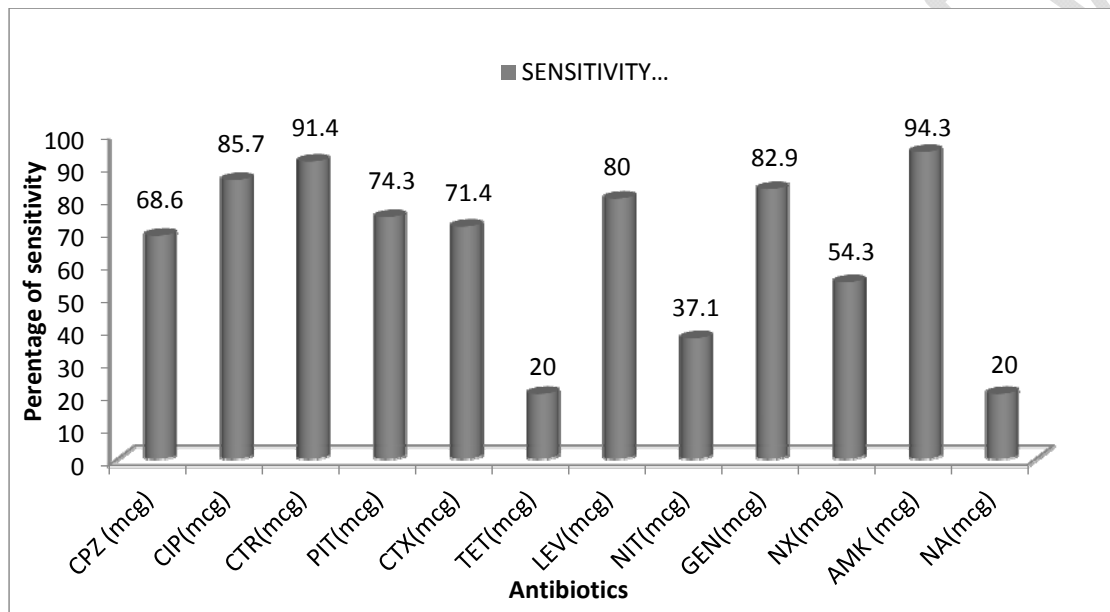


Figure 1. Susceptibility of the uropathogens to the antimicrobial agents used.

Cefoperazone (CPZ), Ciprofloxacin (CIP), Ceftriaxone (CTR), Piperacillin (PIT), Cefotaxime (CTX), Tetracycline (TET), Levofloxacin (LEV), Nitrofurantoin (NIT), Gentamicin (GEN), Norfloxacin (NX), Amikacin (AMK) and Nalidixic Acid (NA)

4 DISCUSSION

It is of great importance to detect and effectively treat urinary tract infections early especially, among the risk group of pregnant women. The signs and symptoms of UTI's in asymptomatic patients are not always obvious hence the need for routine screening to aid in early detection. The

study sought to investigate the incidence of asymptomatic uropathogens infections among pregnant women aged between 19-35 years using microscopic and microbiological methods.

The results of this present study showed that 17.7% of pregnant women attending a clinic at Bolgatanga Regional hospital were infected with bacteria-associated UTI. The prevalence of UTI recorded by this present study discords with earlier findings obtained previously in Ghana. This is significantly higher than the findings obtained previously in Ghana [10, 11], [12], and Ethiopia [13] but comparable to that conducted in Northern Tanzania [14]. The discrepancies in prevalence could reflect the difference in sociocultural practice, the economic status between the study areas, sexual contact, personal hygiene [15], the period the studies were conducted and the variations in diagnostic protocols employed.

Findings from this study indicate that *E. coli* and *Staphylococcus aureus* were the most prevalent bacteria implicated in urinary tract infection among the pregnant women and this harmonizes with these earlier studies in Ghana [16] and other African countries [17, 18]. In this study, the second dominated isolate was coagulase-negative *Staphylococci* 12(34.3%), which is slightly higher than other studies [10, 12] but comparable to reports elsewhere [5, 19]. Urogenital infection with *Schistosoma haematobium* is associated with severe morbidity and mortality and it is characterized by granulomas formation in uterus, fallopian tube, and ovaries [20]. This usually occurs when *Schistosoma haematobium* successfully penetrates the urinary tract into the genital region. This can result in infertility. The study reported *Schistosoma haematobium* infections among four participants, highlighting the need to look at the source of drinking water of the catchment areas surrounding Bolgatanga.

Age, parity, and gestation are purported to be a risk factor for the acquisition of UTI among pregnant women [21, 22]. In agreement with this posit, Chi-square analysis revealed a significant association ($p < 0.05$) between parity and urinary tract infection. However, no evidence of association was found between gestational age and UTI and this is in contrast to the findings by Obeng-Forson et al. [16]. The incidence of bacteria-associated urinary tract infection was highest among pregnant women age < 20 years followed by 20-30 years and > 30 years. This finding discords with earlier studies where UTI was found to be more prevalent among women age > 30 years [4, 16, 23]. Further, all UTI cases were recorded among pregnant women in their 1st trimester and none among pregnant women in their 2nd and 3rd trimesters. This disagrees with a study in Ghana Obeng-Forson et al. [16] where UTI was most prevalent among women in their 2nd and 3rd trimesters.

Substantially high sensitivity rates were recorded for the following antibiotics in descending order, AMK 94.3%, CTR 91.4%, CIP 85.7%, GEN 82.9%, LEV 80% and PIT 74.3%. The activeness of AMK against the isolates could be attributed to the fact that AMK is reserved for the treatment of serious infections and are not available over the counter. This may result in low drug-pressure and consequently the development of low-level resistance against AMK. A similar finding was reported against bacteria isolates in a different study carried out in Ghana [24]. On the other hand, low susceptibility rates were observed for NX, NIT, TET and NA. The findings of this study are similar for some antibiotics reported in other studies [5, 10, 25].

5 CONCLUSION

The present study estimated the overall prevalence of urinary tract infection among pregnant women attending a clinic at Bolgatanga regional hospital at 17.5 %. Of the isolated, *Escherichia coli* and *Staphylococcus aureus* were the most prevalent implicated in the UTI infection. The study

also found evidence of association between parity and UTI infection. Though the study reported low-level schistosomiasis infection, there is the need to assess the water source in these areas. Amikacin seems to be the most effective antibiotic against all the recovered bacteria isolates.

6 CONFLICT OF INTEREST STATEMENT

The authors declare that there is no conflict of interest regarding the publication of this paper.

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