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Prevalence of *Schistosomia haematobium* and *Staphylococcus aureus* among Pregnant Women in Akure

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Authors' contributions

This work was carried out in collaboration between all authors. Author TAO designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors FCA and ASF managed the analyses of the study. Author OAO managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Schistosomiasis is one of the most important parasitic disease of man. *Staphylococcus aureus* has also been observed as one of the causal agents of Urinary Tracts Infectionsand can be regarded as threat. This study focuses on the prevalence of *Schistosoma haematobium* infection and Staphylococcal infection and the relationship between these infections among pregnant women in semi – urban communities of Akure, Nigeria. A survey of Schistosomiasis and Staphylococcal infection were carried out among pregnant women between ages of 15-44 years that were selected randomly at Mother and Child Hospital, Akure, Ondo State in February 2016. Questionnaires were administered to obtain demographic data. One hundred urine samples were examined, comprising 30 from first trimester and 35 both for second and third trimester. The overall prevalence of *S. heamatobium* was 4% while *S. aureus* was 82%, the overall prevalence was much in age group 20-24 years for *S. aureus* and 40-44 years for *S. heamatobium* and first trimester had the highest prevalence for both cases. The highest prevalence was associated with water and toilet facility.

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There was association between *S. aureus, S. heamatobium* and toilet and water facilities. Microscopic examination of the urine sample also revealed yeast cells, RBC and crystals, which have health implications. This study revealed a relative declinein the prevalence of Schistosomiasis in the study area while Staphyloccocal infection had a substantial prevalence.

Keywords: Schistosomiasis; Schistosoma haematobium; Staphylococcus aureus; pregnant women and prevalence.

1. INTRODUCTION

Urinary tract infections (UTIs) are mostly common infection diagnosed in both hospitalized and outpatients and has been reported to be one of the most common bacterial infections among women with high morbidity and mortality rate and accompanied with increase in the cost of hospital care [1]. International Classification of Diseases in 2010 (ICD-10) recognized urinary tract infections as infections that affects organs responsible for urine secretion and elimination [2]. UTIs is observed to exceed 0.5 in a year among women that are sexually active [1]. In developing countries, UTIs are worse and empiric antibiotic treatment are mostly administered before laboratory diagnostic test results are available for therapeutic guide [2]. Currently, there is an increasing problem of resistance to antibiotics especially in developing countries, prompting the need for a good knowledge of antibiotics resistance patterns of these pathogens to ensure efficient treatment and eradication [1].

An UTI during pregnancy is considered complicated. During gestation, untreated UTIs can lead to several pregnancy complications, such as low birth weight infants, premature delivery, and occasionally, stillbirth. Prompt and efficacious treatment of symptomatic UTIs is warranted in pregnant women. Nevertheless, there is still some controversy regarding the screening and treatment of asymptomatic forms during gestation [3]. Complicated UTIs may occur in men, children and pregnant women, but it is especially common in the elderly as well as in immuno-compromised/suppressive patients like pregnant women and in individual with neurologic disorders [4].

Schistosomiasis is a parasitic disease caused by blood flukes (Trematodes) of the genus *Schistosoma*. About 700 million people are at risk of Schistosomiasis infection in 76 countries [5]. The disease is endemic among people whosetheir anthropological activities around rivers such as farm work, domestic chores and recreational activities expose them to infested water [6]. Schistosomiasis is the third most devastating tropical disease in the world, out of which 85% of infected individuals live in Africa [6].

There are about fifteen species of Schistosomes that are prevalent in different areas of the world and produce different symptoms. Some of the important species are *S. mansoni, S. haematobium, S. mekongi, S. japonicum* and *S. intercalatum.* Among all these species, *S. mansoni*is widely distributed in Africa, the Eastern – Mediterranean, the Caribbean and South America and can only infect humans and rodents [6]. Complications caused by this parasite has been documented and the most common complication is periportal fibrosis, also termed Symmers clay pipe-stem fibrosis. This leads to portal hypertension and gastrointestinal hemorrhage. Liver failure is uncommon, except in persons with concomitant chronic hepatitis or cirrhosis. *S. haematobium* causes lesions in the female lower genital tract (i.e. cervix, vulva, and vagina). Female genital Schistosomiasis (FGS) has been identified as a major social and medical problem that may facilitate the spread of some sexually transmitted diseases such as HIV and human papillomavirus (HPV) [7].

Infected patients require diagnosis, treatment and education about how to avoid re-infecting themselves and others. Adequate healthcare facilities need to be available, water systems must be treated to control snail populations, and sanitation must be improved to prevent the spread of the disease. After more than 20 years of Schistosomiasis control programmes, chemotherapy has been shown to be a very important tool. Several therapeutic measures have been developed against Schistosomiasis, such as metrifonate, oxamniquineand praziquantel (Biltncide), which are safe and effective [8]. Praziquantel is the most effective against all forms of Schistosomiasis with few side effects. This drug is given in either two or three doses over the course of a single day [8]. Oxamniquine is used mostly in Africa and South

America to treat intestinal Schistosomiasis [8]. Staphylococcal UTI is one of the most difficult UTI infections that can be controlled due to the antibiotic resistant strains of this species and if this medical condition is associated with Schistosomiasis, the threat that it will pose on the concerned pregnant woman cannot be underestimated. Some studieshas revealed the association of *S. haematobium* infection and Sexually Transmitted Infections caused by *Neisseria gonorrhea, Chlamydia trachomatis, Mycoplasma genitalium* and *Trichomonas vaginalis* among pregnant women in Edo [8]. However, few studies has been done on the prevalence of *S. haematobium* and the frequency of occurrence of *S. aureus* among pregnant women. This study therefore tends to reveal this prevalence of *S. haematobium* and *S. aureus* among pregnant women in Akure, Nigeria.

2. MATERIALS AND METHODS

2.1 Study Area

The study was carried out in Mother and Child Hospital, which is the leading hospital especially for women receiving anti-natal and post-natal care within Akure metropolis and Ondo State as a whole. The samples collected were transported on ice packed to Microbiology laboratory, FUTA for analysis.

2.2 Study Population

One hundred pregnant women from Mother and Child Hospital were randomly selected for the study. The convenience sampling method was used in which only the willing and available pregnant women at the time of the study were recruited into the study. The women examined cut across a trimester of pregnancy with a total of thirty women in the first trimester, thirty-five women for both the second and third trimesters were examined. The bottles containing the urine samples were labeled properly.

2.3 Ethical Clearance

Ethical clearance was sought for and granted by Ethics Committee of Mother and Child Hospital, Akure, Ondo State. Permission was also obtained from administrators and nurses of the Hospital before the study commenced. Informed consent was obtained from all the participants.

2.4 Administration of Questionnaires

Structured questionnaires were administered to the pregnant women from as samples were collected. Demographic data obtained were location, occupation name and age of the women, sources of water for drinking and household activities, housing sanitation, types of toilets, history of bloody urine and terminal haematuria and outdoor activities of the women such as swimming.

2.5 Collection of Urine Samples

After demographic information (e.g. age, trimester and anti - biotic use for the past week) were obtained from their folders in the antenatal clinic, the pregnant women were adequately educated on how to take mid-stream urine sample of their first urine into sterile capped, dry, wide-necked, leak-proof, and labeled sample tubes which were given to each of them. A volume of 10 ml mid-stream urine samples were collected from the anti-natal patients. All the urine containers are well labeled properly and were transported on ice to Microbiology laboratory, FUTA.

2.6 Laboratory Analysis of Samples

2.6.1 Physical examination of urine

The physical characteristics of the urine samples such as color were recorded immediately after collection from the pregnant women.

2.6.2 Urine chemistry

Each urine sample was divided into two sub-samples. One subsample was used for rapid assessment of microhaematuria and proteinuria using Medi-Test strip. Each reagent strip was dipped into urine sample and matched with colour chart on test strip container for detecting the presence and values of these biochemicals. The urine samples were subjected to a semi-quantitative test using a urinary dipstick combi-9 from ACON laboratories, Inc, USA was used for urinalysis tests to characterize the presence and levels of chemical entities such as proteins, nitrite, blood, glucose, Ketones etc. This was done by dipping a test strip into each urine sample and comparing the observed color changes on the strip to a reference color chart provided on the package of the test strip [9].

2.6.3 Urine microscopy

Ten ml of the second sub-sample urine was centrifuged at 2000 rpm in a 15 ml conical centrifuge tube for five minutes. The clear portion of the urine was discarded and the supernatant was thorough mixed with a glass rod and placed on a clean slide covered with a cover slip. It was examined under a light microscope using both low and high power objective lens (10X and 40X) with the condenser iris closed sufficiently to give good contrast for identification of pus cells, Schistosoma ova, pus, epithelial cells, crystals, and other unusual components [9].

2.6.4 Isolation, identification and counting of bacteria isolates

A loopful of urine sample was taken using a standard loop calibrator and inoculated on a Mannitol Salt Agar (MSA) using streaking methodand was incubated aerobically at 37^{o}C for 24hours in an IPF400 Precision incubator (Memmert, Germany). The different bacteria colonies were identified on the basis of their morphological and biochemical characteristics as described by Cheesbrough [9].

2.7 Antimicrobial Susceptibility Test (AST)

The antibiogram of the isolates to selected conventional antibiotics was determined by the disc diffusion method [10]. Using antibiotic-impregnated paper discs (Medicare Nig. Ltd.) containing the following antibiotics: Pefloxacin, gentamicin, Ampliclox, erythromycin, Zinnacef, Amoxacillin, Rocephin, Ciprofloxacin, streptomycin and SeptrinSterile Petri dishes were seeded aseptically with 1 ml each of 18 h old pure cultures of the test organisms each while about 15 ml of sterilized Muller-Hinton agar was poured aseptically on the seeded plates. The culture was first standardized using spectrophotometer and plate count methods at 2.0 ×104 cfu/ml. McFarland standard at 540 nm (0.050 spectrophotic reading) was used. The plate were swirled carefully for even distribution and allowed to gel. With the aid of sterile forceps the antibiotics discs were placed firmly on solidified plates and incubated for 24 h at 37℃. After incubation, zones of inhibition were measured in millimeter (mm). The experiment was carried out in triplicate [10].

2.8 Statistical Analysis

Statistical Package for Social science was used for all statistical analyses. One -way Analysis of Variance (ANOVA) was used to determine the significance of the differences in UTI prevalence among the trimesters, source of water, toilet facilities and age groups. P ≤0.05 was considered statistically significant in all analysis.

3. RESULTS

3.1 Urine Macroscopic Examination

According to the macroscopic examination done on the urine samples, different colours of urine were observed, the number and percentage of people infected with both infections are shown in Table 1. Pale yellow was the most normal colour.

Table 1. Macroscopic examination of urine samples

S/N	Colour	Number of urine samples
1	Pale-straw yellow	23
2	Dark yellow	18
3	Amber	30
4	Pink-reddish	29
	Total	**100**

3.2 Chemical Component and Microscopic Examination of Urine Samples

Among the 100 urine samples that were analyzed, 40% tested positive for protein,14% had glucose, 30% of pregnant women had nitrate in urine. This can be observed in Table 2. For microscopic examination, 27% had yeast cells, 4% eggs of Schistosoma, 5% Red Blood Cell, 18% epithelial cells and 34% hadpus cells that signifythe invasion of UTI organisms (Table 3).

3.3 Socio-demographical Data and Obstetric Characteristics

3.3.1 Age- related prevalence of schistosomiasis and staphylococcal infection

Age 20-24 years recorded highest prevalence (94.4%) of *S. aureus* compared with other age ranges. For urinary Schistosomiasis, age group 40-44 recorded the highest prevalence (33.3%) than those of other age groups with 2 infected persons out of 3 persons in this age group of *S. haematobium* eggs in urine. On the other hand, individuals in age group 25-39 years were found not infected with Schistosomiasis. The data is significant for both infections on the age group, $p< 0.05$. This is revealed on Table 4.

Table 2. Chemical component of urine samples

Chemical entity	Number of positive test (%)	Number of negative (%)
Protein	40	60
Ketone	2	98
Bilirubin	1	99
Nitrate	30	70
Blood	15	85
Glucose	14	86

Table 3. Microscopic examination of urine samples

Deposit	Number of positive test (%)	Number of negative test (%)
Yeast cells	27	73
S. haematobium	4	96
Red Blood Cell	5	95
Pus cell	34	66
Epithelial cells	18	82

3.3.2 Trimester-based prevalence

The highest prevalence for *S. aureus* was among first trimester of pregnancy with 93.3% prevalence while Schistosomiasis also had the highest prevalence of 10% among the first trimester. This shows 100% relationship among the infections in the trimester. Although the infection rate of Schistosomiasiswas statistical insignificant ($p>0.05$) while the trimester was significant for bacterial infection ($p<0.05$), (Table 5).

3.4 Prevalence Rate among People who have Undergone Treatment on *S. haematobium*

Two forms of treatments were considered in this study; treatment from the hospital and traditional treatment. Among patients (10) who had visited hospital to receive treatment for Schistosomiasis, it was observed that many of them were uninfected (9; 90.0%) and percentage of uninfected was also higher (2; 66.7%) among patients that undergone treatment locally. This can be observed in Table 6.

3.5 Antimicrobial Sensitivity Test

The antimicrobial sensitivity pattern for all the *S. aureus* isolated was shown in Figs. 1-2. Most of the drugs were sensitive to the microorganisms isolated except for Ampiclox (12.2%) and Zinnacef (9%). Pefloxacin had the highest percentage of sensitivity on the *S. aureus* isolated (Fig. 3).

Table 4. Age interval of infected women

Age group	Number tested	*S. aureus*, (%)	*S. haematobium*, (%)
15-19	7	6 (85.6)	1(14.3)
20-24	18	17 (94.4)	2(11.1)
25-29	34	24 (70.6)	-
30-34	24	20 (83.3)	-
35-39	14	13 (92.9)	-
40-44	3	2 (66.7)	1 (33.3)
Total	100	82 (82)	4 (4)

Table 5. Infection by pregnancy trimester

Trimester	No examined	No infected with *S. aureus*, (%)	No infected with *S. haematobium*, (%)
First	30	28 (93.3)	3 (10)
Second	35	31 (88.6)	1 (2.86)
Third	35	23 (65.7)	
Total	100	82 (82.0)	4 (4.0)

Table 6. Prevalence rate for people who have undergone treatment on *S. haematobium* (n=15)

Treatment	Total number	Number infected, (%)	Number of uninfected, (%)
Hospital	10	1 (10.0)	9 (90.0)
Local	3	1 (33.3)	2 (66.7)
No action	2	2 (100)	0 (0.00)
Total	15	4 (26.7)	11 (0.11)

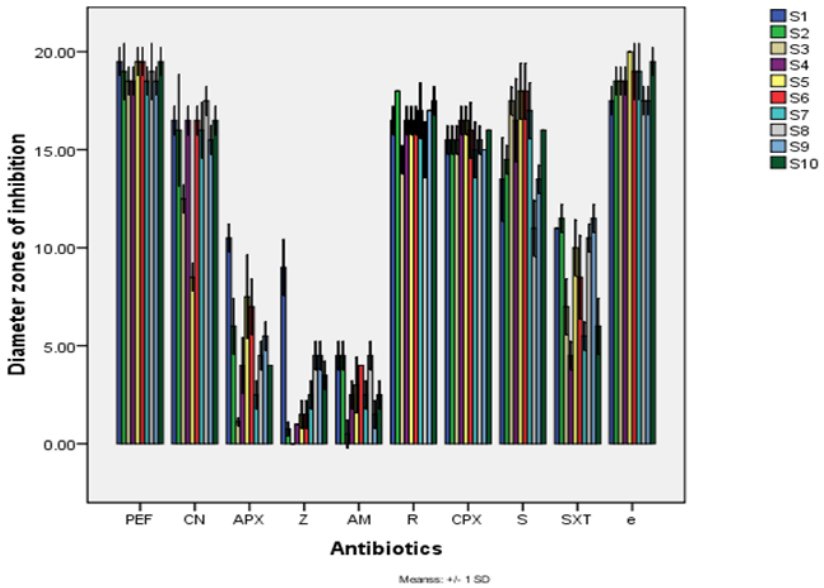

Fig. 1. Antibiotic sensitivity pattern of *S. aureus* isolated from urine sample

Keys: P- Pefloxacin, Cn- Gentamicin, Apx- Ampliclox E- Erythromycin, Z- Zinnacef Sxt-Septrin, Cpx- Ciprofloxacin, Am- Amoxacillin, Z-Zinnacef, R-Rocephin

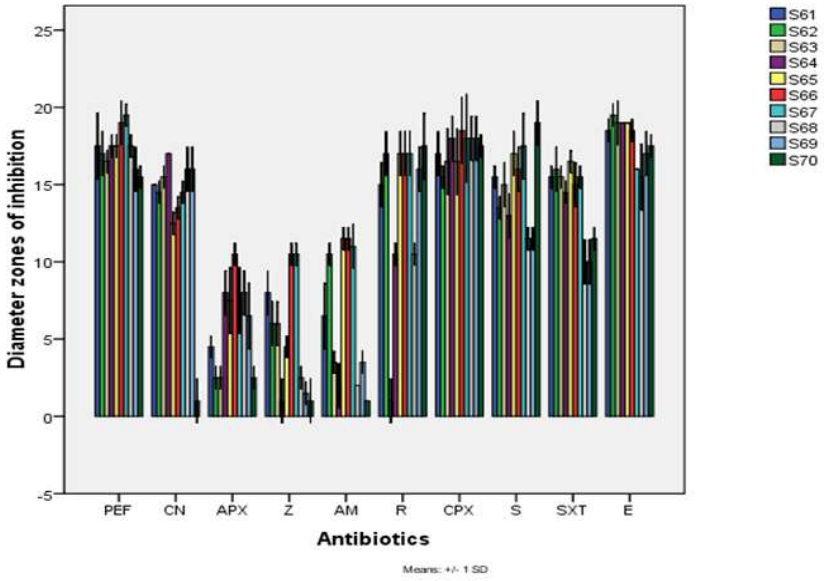

Fig. 2. Antibiotic sensitivity pattern of *S. aureus* isolated from urine samples

Keys: P- Pefloxacin, Cn- Gentamicin, Apx- Ampliclox E- Erythromycin, Z- Zinnacef Sxt-Septrin, Cpx- Ciprofloxacin, Am- Amoxacillin, Z-Zinnacef, R-Rocephin

4. DISCUSSION

The main findings of this study was that the prevalence of *S. aureus* among pregnant women was 82% while *S. haematobium* was 4%. The result of the urinalysis showed 40% of the samples contained proteins while 30 pregnant women had nitrate in their urine. The presence of protein in the urine could be due to pathological disorders. Proteinuria in some of the samples could be anon-specific biomarker for UTI [11]. Nitrites were always detected in urine samples for which Gram-negative bacteria were present therefore could cause the indication. Gram-

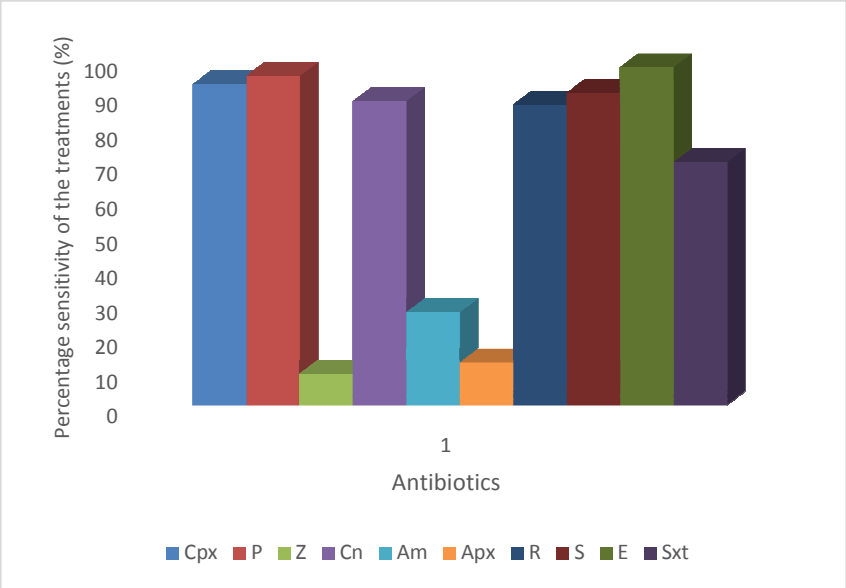

Fig. 3. Percentage resistance of *S. aureus* isolated from urine to antibiotics

Keys: P- Pefloxacin, Cn- Gentamicin, Apx- Ampliclox E- Erythromycin Z- Zinnacef Sxt-Septrin, Cpx- Ciprofloxacin, Am- Amoxacillin, Z-Zinnacef, R-Rocephin

negative organism such as *E. coli*, *Kl. pneumonia and P. mirabilis* have been observed to be able to reduce nitrate to nitrite in urine, hence could account for the nitrate present [12]. This agrees with Cheesbrough [10] who had linked bacteria invasion to the presence of protein and nitrite in urine.

Microscopic examination of urine samples showed that34% of the samples had pus cells, while 27% of the samples had yeast cells. The high percentage of pus cells in the urine samples is an indication of the presence of urinary tract infection. In addition, the presence of high percentage of glucose in this study might be responsible for the high number of yeast cells in the urine. Sugar is known to be a good culture medium for the growth of fungi. 18% epithelial cell present may not all be in case of infection, normally in few people epithelial cells can be seen. Normally few RBCs can be seen in urine of normal individuals but large numbers may be as result of clinical issues, the 5% RBC is associated with the 4% urinary Schistosomiasis, in other cases, RBC may be because of other complications. In this study, eighty-two (82) urine samples gave significant growth representing 82% prevalence. This study is similar discoveries reported in previous studies, for example the report is comparable to similar study by Mbata [13] who recorded 77.9% Staphylococcal infection among prison inmates and also in the dry season and much of the snails are not around.

Schistosomiasis is usually common during rainy season Herbert [14] and most infected patients might have used medication to conquer it since in the response to the questionnaires shows some people had blood case in their urine but have undergone treatments. In addition, since Akure is an urban area and not village with relatively high percentage of infrastructure and education, Schistosomiasis is not common and it is not one of the pandemic cities for the parasite.

The relationship between infection of parasitic UTI and asymptomatic UTI is 100% explaining that for each individual having asymptomatic UTI also have the parasitic UTI. In this survey the highest prevalence is among people that use pit latrine, it is noticed that most pit- latrines are not clean and the intermediate host-snail may be present in the pit environment. Also *S. aureus* is high among these people because there is no portable water available to these people because the most response in the questionnaire suggests they are using well water and those that uses river frequently has 100% prevalence. Water facility and toilet facility could be the major factor contributing the infections.

The type of treatment gotten is an important factor in the blood cases of the infected patients.

From the response from the questionnaire, it was discovered that 15 patients have blood history in their urine. It may be due to some other medical conditions necessary not Schistosomiasis but it was discovered only 1% of people that has undergone hospital treatment that has definitely used antibiotics or other anti-helminth drugs has Schistosomiasis, 33.3% of people who has undergone traditional treatment has Schistosomiasis while 100% of people that took no action has the infection. It is concluded the hospital treatment has reduced the probability of infection cases. For the parasitic infection, two people has light infection of between 2-6 eggs contributing to 2% of the population while 2% also has heavy infection of 11-15 eggs. There was resistance towards some of the most common antibiotics in use, the highest sensitivity is with Erythromycin, followed by pefloxacin**.** This issimilar to the study of Obiazi et al. [15] who recorded Erythromycin showing the highest susceptibility.The observation that ampicloxdid not have good inhibitory activities on the isolates of Gram-positive bacterial species isolated in this study agrees with the findings of Olukitibi and Adebolu [16] who reported that *S. aureus* showed resistance to treatment of antibiotics [16].

5. CONCLUSION AND RECOMMENDATION

5.1 Conclusion

The study revealed that UTI caused by *S. aureus* is one of the major health problem among pregnant women in Akure. This complication can lead to increase in mortality and morbidity rate. There is also a relationship between the parasitic infections caused by *S. haematobium* and *S. aureus* which suggests the invasion of the parasite is accompanied with UTI. The age of women, trimester and other socio-economic status can as well contribute to the rate of staphylococcal UTI in women and *Schistosoma* infection during pregnancy, therefore, there is a need for frequent and consistent evaluation of the prevalence in order to reduce the devastation effects of urinary tract infections in pregnancy on both maternal and foetal health.

5.2 Recommendation

Routine and regular mass screening for asymptomatic UTI and parasitic infection should be carried out because untreated cases may lead to pylonephrititis, renal scaring and kidney failure. Good personal hygiene should be kept and the toilet and water facilities that contributed to the overall prevalence should be neat and portable respectively.

CONSENT

As per international standard or university standard, patient's written consent has been collected and preserved by the authors.

ETHICAL APPROVAL

As per international standard or university standard, written approval of Ethics committee has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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