# Bacteriological Profile of Urinary Tract Infections in Pregnant Women at the Atakora Departmental Hospital, Benin

**Aikou Arielle, Aikou Nadine and Aikou Nicolas\***

*Department of Human Biology, National University of Sciences, Engineering and Mathematics, Laboratory of Clinical Biochemistry and Medical Microbiology, Benin*

**\*Corresponding Author:** AIKOU Nicolas, Department of Human Biology, National University of Sciences, Engineering and Mathematics, Laboratory of Clinical Biochemistry and Medical Microbiology, Benin.

## Summary

*Introduction:* Urinary tract infections (UTIs) are the second most common infectious disease in humans after respiratory diseases [1]. In pregnant women, any UTI is by definition at risk of complication [1]. Urinary tract infection (UTI) can have harmful consequences for the mother and the fetus. It can manifest itself in three forms: gravidic urinary tract colonization, acute gravidic cystitis and acute gravidic pyelonephritis (AP). The urine cytobacteriological examination (UCE) is the only way to confirm the diagnosis and to guide the treatment. It is also by far the most frequently requested examination from a microbiology laboratory [2].

*Objectives:* The aim of our study is to establish the bacteriological profile of germs involved in urinary tract infections in pregnant women at the Departmental Hospital Center of Atacora, Northwestern BENIN from March to October 2021.

*Methods:* This is a prospective, descriptive, cross-sectional study with an analytical focus that included 192 ECBU samples collected from pregnant women in prenatal consultation at the Departmental Hospital Center of ATACORA. On these samples we performed the following examinations: ECBU followed by antibiogram.

*Results:* The study revealed that out of 192 ECBU 19 were positive, i.e. a frequency of 9.9%. This result was confirmed by [3] and [4] who found 10% and 9.9% respectively. The most frequent germ was *Escherichia coli (68.4%)* followed by *Staphylococcus aureus (21.1%)* and finally *Klebsiella pneumoniae (10.5%).* However, we found a resistance to (90.1%) ampicillin and a sensitivity to (100%) ceftriaxone in Gram-negative bacteria, in Gram-positive bacteria we noted an effective activity of third generation cephalosporins, fluoroquinolones.

*Conclusion:* The results of this study allow us to highlight the problem of treatment of urinary tract infections in pregnant women. In the absence of bacterial identification and antibiogram, the problem of the choice of antibiotics arises. One possibility is to choose the less expensive aminopenicillins, knowing that very few of the isolated strains are sensitive to amoxicillin and that the combination of amoxicillin and clavulanic acid is only active on a small proportion of the isolated strains. The choice of other more active antibiotics is limited, either by the numerous contraindications (quinolones), or by their high cost, especially the third generation cephalosporins

*Keywords:* Urinary tract infection, Bacteria; Asymptomatic bacteriuria; Acute gravid cystitis; Acute gravid pyelonephritis

## Introduction

UTI is the bacterial colonization of the urine and/or urinary tract from the kidneys to the urethral meatus [5]. UTI is a relatively common condition during pregnancy; 5-10%, a 2-2.5 fold increase in risk [6]. It is benign in the majority of cases, generally limited

to the lower urinary tract. It most often associates bacteriuria with leukocyturia; however, the latter may be absent in asymptomatic infections. According to KASS, quoted by KASS: [7] we speak of urinary tract infection whenever bacteriuria is greater than $10^5$/ml of urine and leukocyturia greater than $10^4$/ml of urine. UTI can occur at any age during pregnancy. It ranges from asymptomatic bacteriuria to sepsis through the intermediate stages which are: cystitis and pyelonephritis. Given its high frequency during pregnancy, several researchers have conducted studies on the issue. A study conducted at CSRéf CII in 2005 and 2006 by (*Coulibaly D.*,2007) revealed that one pregnancy out of 12 is associated with a urinary tract infection, a frequency of 9.9%. The aim of our work is to study the bacteriological profile of germs causing urinary tract infections in pregnant women at the Departmental Hospital Center of Atacora (Natitingou). It will therefore be a question of:

- To determine the prevalence of urinary tract infections among pregnant women at the Atacora departmental hospital
- Determine the nature of the germs responsible for these urinary tract infections.
- To establish the bacteriological profile of these germs
- Evaluate the sensitivity of the identified germs to the tested antibiotics.

## Patients and methods

This is a cross-sectional, descriptive, analytical study of the bacteriological profile of urinary tract infections in pregnant women during the period from July 10 to September 10, 2021. The study was open to pregnant women who came for prenatal consultation and in whom the diagnosis of pregnancy was confirmed. On the basis of the questioning and information contained in the health record, pregnant women under antibiotic treatment or having completed such treatment less than three months ago were excluded from our study. Our study included 192 pregnant women. For each pregnant woman, various parameters were collected from the records and prenatal consultation diaries. Data were collected using an individual survey form filled out during the prenatal consultation with the help of health care providers. The consent of the pregnant women was requested and obtained; the ethics committee also gave its favorable opinion. The pregnant women were included in the study on the basis of free consent.

## Method

The urine is collected in a sterile tube and transported to the laboratory as quickly as possible. The main objective of this step is to collect the bladder urine in a sterile way, avoiding its contamination during miction by commensal flora, which colonize the urethra and the perineal region. In order to perform a qualitative and quantitative bacteriological analysis of the urine.

The macroscopic examination of the sample allows to assess the appearance of the urine: color, turbidity and odor.

The microscopic examination of the sample is done in two parts. The first part is the quantitative examination of the total urine: A drop of urine is placed in a Malassez cell. A count of leukocytes and red blood cells/mL is performed. Then, in the fresh state, this step consists in searching for the presence of germs, their mobility and even parasites; the quantification of red blood cells and leukocytes. This result is given in red blood cells/leukocytes/minute. It is carried out by:

- The deposit of a few drops of the urine pellet on the central area of the slide using a sterile pipette.
- Cover with a slide, avoiding air bubbles.
- Observe quickly in low light without oil at the x40 objective.

### *Gram staining*

This examination is carried out before and after a culture during a 24h incubation period at 37°C. It allows to highlight the dyeing properties of the bacteria.

### Operating mode

- Put some gentian violet for 1min;
- Rinse and put Lugol's for 40s
- After rinsing put alcohol for 30s;
- Put the fushin for 1min;
- Rinse, dry, and observe

The staining time depends on the dilution of the dye.

Choosing the culture medium: the choice of the culture medium depends on the Gram observation.

### CLED agar

Medium on which all urinary germs grow.

### Chapman agar

Selective medium for Cocci Gram (+) but more selective for Staphylococcus.

### EMB agar

Selective medium for Gram (-) bacilli.

### Bacteriological examination
### Bacterial count

The counting of the colonies that will eventually grow is essential to declare an infection. The technique is performed as follows:

- Homogenize the urine tube.
- Take 0.1ml of the urine pellet, place and streak on the entire surface of an agar plate.
- Incubate at 37°C for 18 to 24 hours in a bacteriological oven.
- Counting is done by counting the number of colonies on the surface of the culture medium.

### Reading

The orientation of the diagnosis is based on the appearance of the colonies which appear on the culture medium and which can be, depending on the germ: pigmented, transparent, mucous, round, bulging, flattened. Some colonies may have characteristic colors and aspects depending on the medium used.

### Cytobacteriological criteria of an infection

Normal urine is sterile, the result of the urine count is one of the most reliable criteria for the diagnosis of infection. During the macroscopic observation of colonies and depending on the case we can have: a- Negative culture - After 24 hours of culture in the absence of colonies, render: a negative culture result . b-Positive mono-microbial culture, N $\geq 10^3$ germs/ml.

Proceed to the identification of the germ then the realization of its antibiogram. c-Multiple types of colonies: poly-microbial contaminations.

### Biochemical identification
### Classic Gallery

In our study we used the classical gallery for the search of biochemical characters, in order to identify the bacteria responsible for

the urinary infection. An isolated colony or a few strictly identical colonies are collected with a loop previously flamed and cooled, then emulsified in a sterile tube containing physiological water, from this inoculum, we seed the different identification media chosen according to the nature of the culture after bacterial counting. These media are incubated for 18 to 24 hours at 37°C. The reading allows to identify the genus and the species of the isolated germ.

### The Kliger Hajna medium

The Kliger Hajna medium is a solid medium, a rapid identification medium for enterobacteria. It allows to highlight the fermentation of glucose, lactose, sucrose and the production of hydrogen sulfide.

### Urea Indole medium

Urea Indole medium is a yellow-orange liquid medium, which allows the detection of indole and urease. Bacteria possessing urease transform urea into ammonium carbonate resulting in alkanization which causes a purplish red coloration of the medium in the presence of phenol red. Some bacteria degrade tryptophan with a tryptophanase by forming indole, this reaction is confirmed after addition of Kovacs reagent.

### Mannitol Mobility Medium

Mannitol mobility medium is a solid medium, which allows the study of mannitol fermentation as well as the mobility of the strain, the use of mannitol as a carbon source and pH indicator.

### Simmons citrate medium

Simmons citrate medium is a solid medium, which allows to highlight the use of citrate as the only source of carbon and energy.

### ONPG test

The ONPG test (Ortho-Nitro-Phenyl-Galactopyranoside) is essential for the study of lactose degradation in enterobacteria.

### Catalase test

This test is used for the identification of Gram-positive bacteria. Catalase is an enzyme with the property to decompose hydrogen peroxide ($2HO_2$) with the release of oxygen.

### Antibiogram

It is performed according to the classical method of diffusion of the antibiotic on agar from discs according to the standards of the Clinical and Labolatory Standard Institute (CLSI). It allows guide the choice of treatment and determine the sensitivity and resistance profile.

- Antibiotics tested

In our study six(6) antibiotics were tested:

- *The β- lactams:* Amoxicillin (AMX), amoxicillin + clavulanic acid (AMC), Ampicilin (AMP)
- *Fluoroquinolones:* Ciprofloxacin (CIP)
- *Cephalosporins:* Ceftriaxone (CRO)
- *Macrolides:* Erythromycin (ERY)

### Preparation of the inoculum

Using a sterile swab, an isolated colony is removed from the culture medium and placed in a tube containing 2.5 ml of water physiological.

The suspension is homogenized and must have an opacity of 0.5 Mac Farland used as a reference to adjust the turbidity of the bacteria in suspension (by addition of colonies if the turbidity is low and by addition of physiological water if the turbidity is high).

### Plating of plates

The medium used to perform an antibiogram is the Mueller-Hinton medium. Using a sterile swab dipped in the inoculum and after having discharged it by pressing it against the wall of the tube, the inoculation is done by tight streaks on the whole surface of the plate three times, turning the plate of 60° each time; without forgetting to pass it on the periphery of the agar, making a circular movement.

### Antibiotic disc arrangement

Antibiotic discs are applied with a pair of sterile forceps, leaving a distance of 25-30 mm between discs, while gently pressing each disc to ensure uniform contact with the medium. The plates are then incubated at 37°C for 18 to 24 hours.
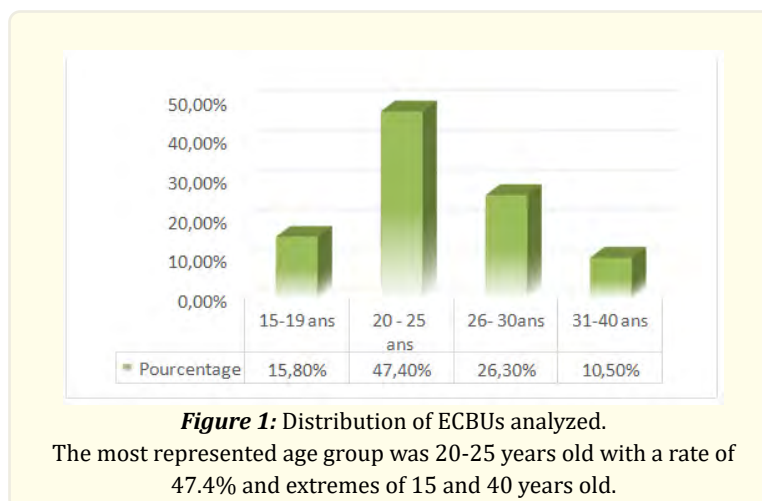
### Reading and interpretation of results

After incubation, the inhibition diameters are measured with a caliper and the strains are classified according to their sensitivities according to the critical values of (CLSI).
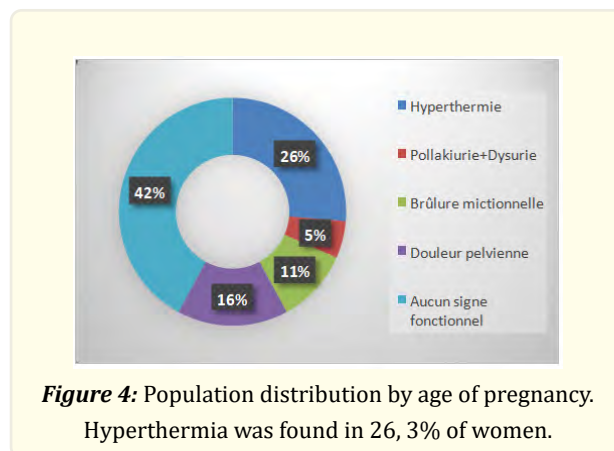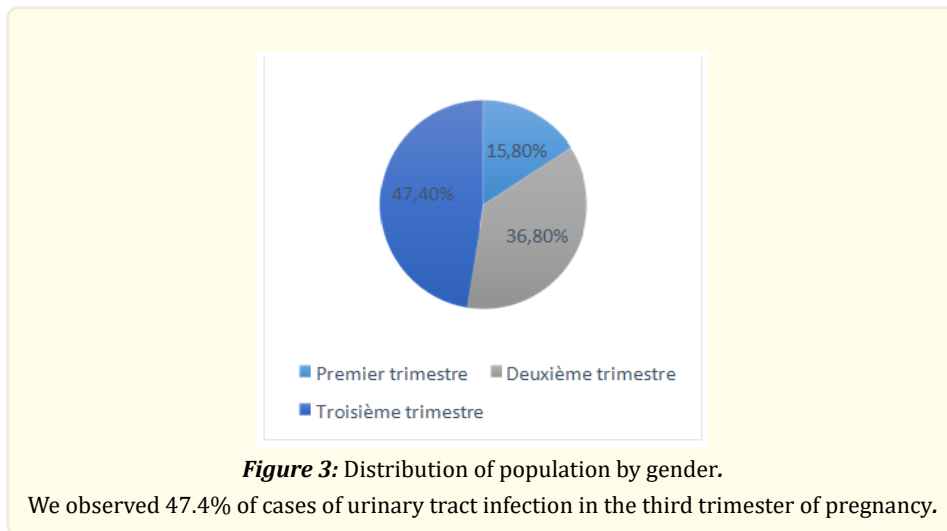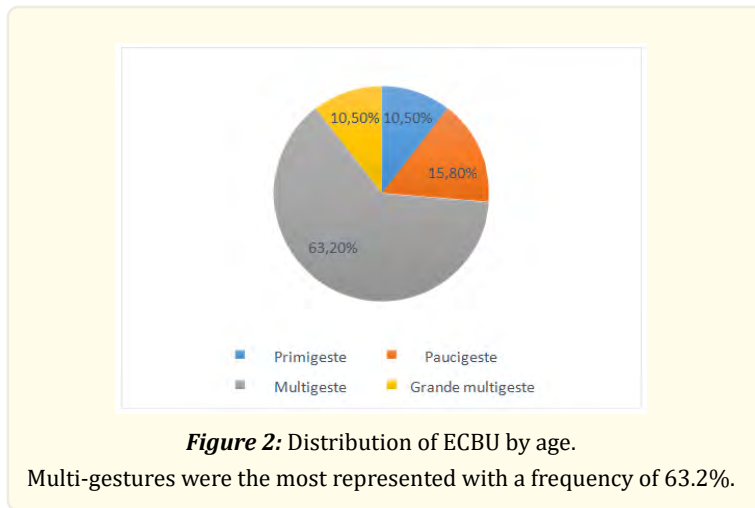
- In cases where the diameters obtained are greater than the critical diameters, the bacteria are declared sensitive (S).
- In cases where the diameters obtained are less than the critical diameters, the strain is declared resistance (R).
- In cases where the diameters obtained are equal to the critical diameters, the bacteria is declared intermediate (I).

## Result

### our results are shown on the following graphs

The urine cytobacteriological examination is the main examination performed in the bacteriology laboratory of the Centre Hospitalier Departemental de l'atacora. Out of 192 urine cytobacteriological examinations (UCE) performed during the study period, 19 UCE were positive. We therefore recorded 19 cases of urinary tract infection out of a total of 192 pregnant women, i.e. a frequency of 9.9%.
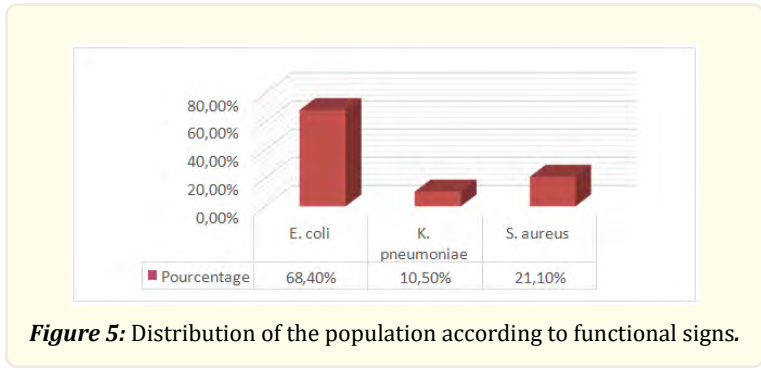


***Figure 1:*** Distribution of ECBUs analyzed.
The most represented age group was 20-25 years old with a rate of 47.4% and extremes of 15 and 40 years old.

***Figure 2:*** Distribution of ECBU by age.

Multi-gestures were the most represented with a frequency of 63.2%.



***Figure 3:*** Distribution of population by gender*.*

We observed 47.4% of cases of urinary tract infection in the third trimester of pregnancy*.*



***Figure 4:*** Population distribution by age of pregnancy.

Hyperthermia was found in 26, 3% of women.

*Figure 5:* Distribution of the population according to functional signs*.*



*Figure 6:* Distribution of ECBUs according to the germs isolated.
Escherichia coli is resistant to ampicillin in 92.3% of cases but sensitive to ceftriaxone in 100% of cases.
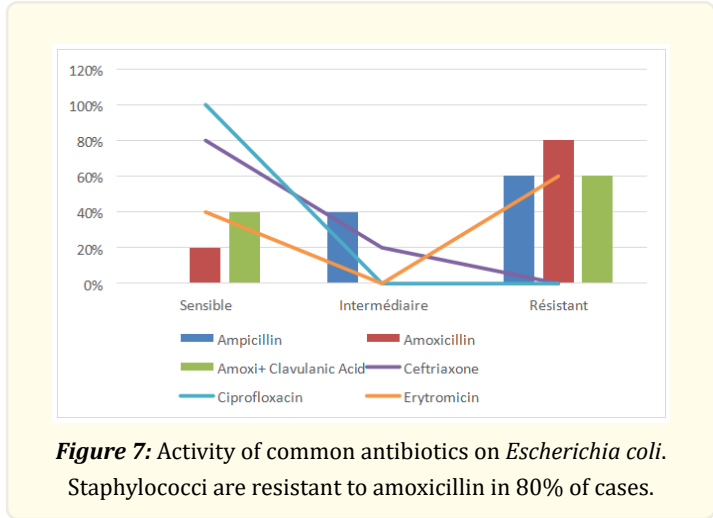


*Figure 7:* Activity of common antibiotics on *Escherichia coli*.
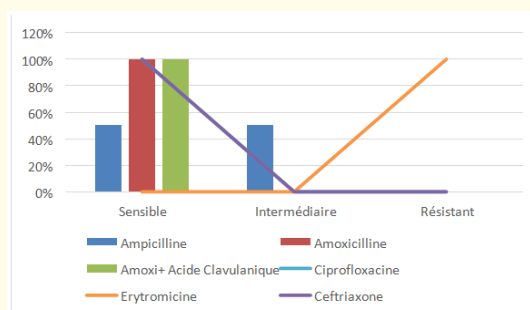Staphylococci are resistant to amoxicillin in 80% of cases.

***Figure 8:*** Activity of common antibiotics on *Staphylococcus aureus*. *Klebsiella pneumoniae* is susceptible to β-lactams and third generation cephalosporins(100%).

## Discussion

Urinary tract infections are infections affecting the urinary system. The germs responsible are generally bacteria. There are three types of urinary tract infections

### Presence of bacteria in the urine in the absence of symptoms of infection:

It is estimated that 2-15% of pregnant women suffer from this. If left untreated, this type of infection can become complicated and can affect the kidneys.

### Bladder infection

This infection affects 1 to 4% of pregnant women.

### Kidney infection

This complication occurs in 1-2% of pregnant women. In 80-90% of cases, it occurs during the second or third trimester of pregnancy.

### Epidemiological aspect

Our study took place from July 10 to September 10, 2021 at the Departmental Hospital of Atacora. We recorded 19 cases of positive ECBU in pregnancy out of a total of 192 ECBU. This represents a prevalence of 9.9%. This result is confirmed by *Brassme T*; *Querlin D*; *Bisserte J*. and *Coulibaly D*. [8] who found 10% and 9.9% respectively. It is slightly higher than the results obtained by Togo A. and Diassana HK [9], who noted 8.8% and 8.54%.

### Socio-demographic characteristics

The age group of 20 to 25 years was the most represented in our study, i.e. 47.4% with extremes of 15 and 40 years. This finding had been made by *Schultz R*, [10] and *Diassana* [11], who found respectively 40%, 38.2% and 36%. This high rate could be explained by our very young population, being at the beginning of their sexual life. Indeed, in this age group, sexual intercourse is generally very frequent, which would favor the spread of germs to the upper urinary tract. The multigestas were the most represented in our study with 63.2%, this rate is higher than that of [12] with a rate of 47.9% among multigestas, this could be explained by the predominance of professional housewives in our study with a frequency of 57.9%.

*Frequency of urinary tract infection according to the term of pregnancy*

In our study, the frequency of UTI was higher in the third trimester with a rate of 47.4%. This rate is similar to that of *Coulibaly D* and *Togo A* who found a high frequency in the third trimester with respective rates of 48% and 45.9%. Gram-negative bacteria were found to be responsible for urinary tract infections in 78.9% of cases. The bacterial species most frequently isolated in our study were, in decreasing order: E. coli (68.4%); Staphylococcus aureus (21.1%); Klebsiella pneumoniae (10.5%).

Escherichia coli was the most common germ found with a rate of 68.4%. This distribution of germs isolated during our study is in line with the results found in several international studies, which allows us to generalize our results.

A study conducted at the military hospital Mohammed V of Rabat showed a clear predominance of enterobacteria which represented 82.4% of isolates. At the head of the line, we find E. coli with a frequency of 57.2% followed by Klebsiella spp (16.6%). Grampositive cocci accounted for 12.1% of isolates [13].

In the laboratories of the city of Nouakchott, gram-negative bacilli represented 94% of the bacterial strains isolated, of which 92.2% are enterobacteria (E. coli 64.4%, Klebsiella 42.1%), 6% are gram-positive cocci.

The Bio 24 medical laboratory in Dakar (Senegal) records that Enterobacteriaceae represent the majority of isolated germs (88.20%), followed by gram positive cocci (8.13%) and non-fermentative bacteria (3.66%). The detailed study of isolated enterobacteria showed that they are mainly Escherichia coli with a rate of 77.84%, followed by Klebsiella (13.90%), Enterobacter (2.34%), Proteus (2.55%).

*Overall antibiotic resistance profile of isolated enterobacteria*
*β-lactam resistance profile*

The strains of enterobacteria isolated from urine are resistant to ampicilin (AMP)92,3%, amoxicillin (AMX)76.9% and amoxicillin + clavulanic acid (AMC) with a rate of 53.8%. The frequency of amoxicillin was found to be higher than that of *Honderlick et al* [14] who reported 47.8% of resistance. The rate for the combination of amoxicillin + clavulanic acid is close to that found by *Alemu et al, 2012* [15] 40.7%, however, it is higher than that found by *Honderlick et al* [16], *Akortha; Filgona* [17] reporting 16.6% and 21.3% respectively.

*Fluoroquinolone resistance profile*

Ciprofloxacin (CIP) resistance (7.7%) is noted, this frequency is close to that of *Honderlick et al* [16] who indicate 12%, but it is higher than that of *Alemu et al* [15] with the percentage of 3.7% and it is lower than that of *Taiwo; Aderounmu,; Akortha; Filgona,* [17] reporting a percentage of 56.4% and 36.4% respectively. The main mechanism of resistance to quinolones and fluoroquinolones involves an accumulation of mutations in the genes encoding the DNA gyrase and topoisomerase IV enzymes. The development of resistance to several families of antibiotics may seriously compromise the use of β-lactams to treat infections due to Gram-negative bacteria. *Activity of usual antibiotics on Staphylococci.*

Staphylococcus aureus showed a good sensitivity to Ciprofloxacin (100%) and to Ceftriaxone (80%). On the other hand, there was a low sensitivity to aminopenicillins (20%).

## Conclusion

Urinary tract infections are a major public health problem since they are responsible for a significant morbidity and mortality. Knowledge of the bacteriological profile of these infections is essential for efficient management. Our study was carried out on 192 UTIs, 19 of which were positive, from pregnant women attending antenatal clinics during the period from March to October 2021. Urinary tract infection is a frequent condition in pregnant women (9.9%). It is very often asymptomatic (42.1%). The epidemiological characteristics are that UTI is the prerogative of young pregnant women aged 20 to 25 years (47.4%). It is also most often found

during the third trimester (47.4%) and especially in multigestational women (63.2%). Screening for UTI should therefore be systematic in all pregnant women from the first prenatal visit, particularly in younger women. From a biological point of view, cloudy urine is most often infected. The germs responsible are mostly enterobacteria (78.9%) and they present a good sensitivity to quinolones and third generation cephalosporins. Gram positive cocci are also isolated and are highly sensitive to quinolones. The results of this study highlight the problem of the treatment of urinary tract infections in pregnant women. In the absence of bacterial identification and antibiotic susceptibility testing, the problem of choice of antibiotics arises. The choice of other more active antibiotics is limited, either by the numerous contraindications or by their high cost. An effective fight against these infections requires a global prevention strategy which implies a close collaboration between gynecologists, epidemiologists, clinicians, bacteriologists, hygienists and the health care team.

## Recommendations

We propose, in the light of the results obtained, the implementation of some measures for a better control of urinary tract infections, the protection of the fetus and that of the mother:

- Systematically perform an ECBU during infectious syndromes and respect the conditions for taking urine samples.
- To fight against the over-the-counter sale of antibiotics by pharmacies and to raise awareness about the danger of self-medication.
- Avoid probabilistic prescriptions of antibiotics.
- Avoid the frequent use of the same class of antibiotics in our health structures and make practitioners aware of the need for rational prescription of antibiotics, preferably guided by a correctly performed and interpreted antibiogram.
- Avoid prescribing amoxicillin and/or amoxicillin + clavulanic acid in the treatment of UTIs (very limited efficacy)
- To set up a therapeutic strategy adapted to the local epidemiology for the treatment of urinary tract infections.

## Acknowledgements

## Authors' Contributions

All the authors contributed positively to the realization of this study. Each of us played his or her part until the manuscript was written. After the first manuscript, they all worked for; the completion of the final version. They remain even willing in case of possible corrections to work for a completion of the publication of our original research article.

## Conflict of Interest

There are no conflicts of interest of any kind. We all worked in a cordial and collegial manner.

## References

1. THOULON JM, PUECH F and BOOG G. "Bacterial infections and pregnancy". obstetrics. 3rd ed Ellipses Paris (1995): 985.
2. LEGRAIN M., et al. Abstract of nephrology: Urinary tract infection. 2nd ed Masson. Paris (1985): 399.
3. GELLE P., et al. Pyelitis and urinary tract infections in pregnancy and childbirth. Their monitoring at by means of the isotonic nephrogram. J. Gyn. Obst. Biol. Reprod 1 (1972): 676-690.

4.  AGHAYAN M, THOUSIN H and LAMBOTTE R. Therapeutic strategies for gravidic bacteriuria. Rev méd. Liège: September 45 (1990): 433-439.

5.  PROVIDED A and LESSOURD-PONNIER F. Urinary tract infections during pregnancy. Enc Méd. Chir (Gynaecology/obstetrics). Paris 5 (1996): 10.

6.  LANSAC JC, BESNIER JM and COTTY F. Bacterial, viral and parasitic infectious disease during pregnancy. Lansac JC, Berger C and Magnin G. Obstetrics for the practitioner. 3rd ed Masson, Paris (1997): 125-164.

7.  ANDDRIOLE V T and PATTERSON TF. Urinary tract infection in pregnancy: Epidemiology-natural history and management. Med. Clin. North. Am. March 75.2 (1991): 338-359.

8.  COLAU JC. "Pyelonephritis in pregnancy". Rev of Prat 43.9 (1993): 1091-5.

9.  AMINON AK. Contribution to the study of the association between urinary tract infection and pregnancy 84 cases. Dissertation of medicine. Cocody Abidjan (RCI) (1989): 52.

10.  CISSE CT, NIANG MM, NAJLAA O, MORCEAU JC.

11.  TOGO A. Study of the association between urinary tract infection and pregnancy in the gynecologicalobstetric service of the Point "G" National Hospital. A propos de 61 cas). Thesis Med. Bko (1993): 20.

12.  KODIO A. Study of urinary tract infections at the laboratory of the National Hospital of Point "G" (in About 2000 bacteriological examinations). Pharmaceutical teas. Bamako(1988): 27

13.  DIASSANA HK. "Urinary tract infection and pregnancy at the Renée Cissé maternity hospital in Hamdallaye". Thése méd. Bamako (2002): 151.

14.  COULIBALY D. "Urinary tract infections and pregnancy in the reference health center of the district of Bamako". Thesis med. Bamako (2007): 81.

15.  Aminata T. Urinary tract infection and pregnancy in the reference health center of the commune II of Bamako, Thesis Med (2012).

16.  SCHULTZ R., et al. Genitourinary infections in pregnant women and low birth weight. birth weight.

17.  CISSE CT, NIANG MM, NAJLAA O, MORCEAU JC.

18.  BAUDET JH. The "usual" forms of urinary tract infection in pregnant women. Ann. Urol 18 (1984): 345-346.

19.  Nezha RACHIDI. Epidemiology and antibiotic resistance of bacteria isolated from urinary tract infections at the Mohammed V military training hospital in rabat. Faculty of Medicine and Pharmacy of Rabat, Mohammed V University (2014).

**Volume 2 Issue 2 February 2022**