
ORIGINAL PAPER

PREVALENCE OF URINARY TRACT INFECTIONS ON PREGNANT WOMEN

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SUMMARY

Introduction: Urinary tract infections in pregnant women complicate the course of pregnancy. It is necessary to know not only the pathogen type but also to prescribe the safest antibiotics for adequate treatment of urinary tract infections.

Materials and methods: The material for the study was urine. The study was conducted by a quantitative method. The identification of microorganisms was realized by MALDI-TOF using mass-spectrometer Microflex (Bruker). Determination of the sensitivity of the isolated strains to antimicrobial agents was performed by disk methods in accordance with the recommendations of CLSI (2012). Statistical processing was performed using WhoNet 6.3 program.

Results: The results of the research showed that the positive samples with a titer 10^6 - 10^8 CFU/ml were obtained in 87 pregnant women. *E. coli* were isolated in 55.17% cases. The most active antibacterial agents against *E. coli* were the drugs of fluoroquinolones group. All *E. coli* strains were susceptible to fosfomycin, trimethoprim, amikacin, isepamicin, netilmicin. High activity had nitrofurantoin (93.8%), ampicillin-sulbactam (97.3%), gentamicin and sisomicin (95.2%). 53.3% isolated *E. coli* strains were sensitive to ampicillin.

Conclusion: The prevalence of urinary tract infections in pregnant women was 22.5%. *E. coli* was the most common causative agent of urinary tract infections in pregnant women. High antibacterial activity was registered in cephalosporins, fosfomycin, and nitrofurantoin.

Key words: urinary tract infections, pregnant women, MALDI-TOF, antibacterial drugs

RÉSUMÉ

Prévalence des infections urinaires sur les femmes enceintes

Introduction: Les infections urinaires chez les femmes enceintes compliquent le déroulement de la grossesse. Il est nécessaire de connaître non seulement le pathogène mais aussi de prescrire les antibiotiques les plus sûrs pour le traitement adéquat des infections des voies urinaires.

Matériels et méthodes: Le matériel pour l'étude était l'urine. L'étude a été effectuée par une méthode quantitative. L'identification des micro-organismes a été réalisée par MALDI-TOF, utilisant le spectromètre de masse Microflex (Bruker). La détermination de la sensibilité des souches isolées des agents antimicrobiens a été effectuée par des procédés de disque conformément aux recommandations du CLSI (2012). L'étude statistique a été réalisée en utilisant le programme WhoNet 6.3.

Résultats: Les résultats de la recherche ont montré que les échantillons positifs avec un titre 10^6 - 10^8 UFC / ml ont été obtenus chez 87 femmes enceintes. *E. coli* ont été isolés dans 55,17 % des cas. Les agents antibactériens les plus actifs contre *E. coli* étaient les médicaments du groupe des fluoroquinolones. Toutes les souches de *E. coli* étaient sensibles à la fosfomycine, triméthoprime, amikacine, isépamicine, nétilmicine. Aussi les agents suivants avaient une forte activité: la nitrofurantoïne (93,8 %), l'ampicilline sulbactam (97,3 %), la gentamicine et la sisomicine (95,2 %). 53,3 % des souches isolées de *E. coli* étaient sensibles à l'ampicilline.

Conclusion: La prévalence des infections des voies urinaires chez les femmes enceintes était de 22,5 %. Le *E. coli* est l'agent responsable le plus courant des infections des voies urinaires chez les femmes enceintes. Une forte activité antibactérienne a été enregistrée en céphalosporines, fosfomycine, nitrofurantoïne.

Mots-clés: infections des voies urinaires, femmes enceintes, MALDI-TOF, médicaments antibactériens

INTRODUCTION

Urinary tract infections (UTIs) often complicate the course of pregnancy, and the rate may reach 8% [3, 20]. The risk of UTIs in pregnant women is higher than in non-pregnant women; and it is associated with physiological changes during pregnancy. The stretching of ureter develops in 90% of women from the 6th week of pregnancy, and may persist until the birth. The increased size of the bladder, with its tone reducing is the cause of urinary retention and development of vesicoureteral reflux. Urinary stasis and disturbance of physiological anti-reflux mechanism are the favorable conditions for bacterial growth and ascending infection. Additional contributing factors may include specific biochemical changes in urine: a higher content of glucose, amino acids, hormones, degradation products that increase urine pH [5, 6].

As in non-pregnant women, UTIs in pregnant women are classified as asymptomatic bacteriuria (ASB), when infection is limited by the bacterial growth in urine, or symptomatic infections, as acute cystitis, acute pyelonephritis, when bacteria invade the urinary tract tissue, causing an inflammatory reaction [13].

The spectrum of micro-organisms that cause UTIs in pregnant women does not differ from the pathogens causing UTIs in non-pregnant women. 80-90% of all infections are caused by *Escherichia coli*, with the remaining 10-20% caused by Gram-negative flora, such as *Klebsiella pneumoniae*, *Proteus mirabilis*. Gram-positive bacterial infections, such as the streptococci and staphylococci are rare [4, 10,16,19].

In Kazakhstan, as well as throughout the world, there is a high level of resistance relating to the main pathogen of outpatient UTIs in pregnant women. This is the resistance of *Escherichia coli* to semi-synthetic penicillins (amoxicillin/clavulanate), cephalosporins of 1-2 generations (cefazolin, cefuroxime, cephalexin, cefaclor) [7]. Thus UTIs in pregnancy should require the special attention in terms of diagnosis and treatment.

The gold standard of UTIs diagnosis is a culture method. It has advantages over microscopy and other rapid diagnostic techniques. Firstly, it has a high sensitivity ($\geq 10^2$ CFU/ml), allowing the infection caused by one type of microorganism to be distinguished from several types of other microorganisms. Second, this method allows determination of the sensitivity of the pathogen; this is essential for the rational use of antimicrobials. Third, the method provides the doctor with epidemiological data about the structure of the pathogen and the possible mechanisms of antibiotic resistance [1].

It is necessary to execute the sampling of material correctly for the analysis. The collecting of urine should be carried out in the morning after a preliminary cleaning of genitals using the plain water with mild soap. It is not recommended to use antiseptics, as this can lead to an artificial reduction of CFU. A sample of midstream urine should be examined not later than for 2 hours after collection.

The choice of antibiotic for the treatment of UTIs in pregnant women is decided not only by the activity of the drug in relation to major pathogens, but also the require-

ments for the safety of the antibiotic. At present the beta-lactam antibiotics: ampicillin, cephalosporins are commonly used; these may be assigned in pregnancy with a high degree of security [2, 12, 14].

AIMS OF THE STUDY

The purpose of the study is the definition of sensitivity to antibiotics of microbial strains isolated from pregnant women with UTIs.

MATERIAL AND METHODS

The material for the study was an average portion of the morning free flow of urine obtained after the cleaning of external genitals. The study was conducted by a quantitative method. The material was applied to a blood agar with the calibration loop (10 microlitres), the isolated microorganism included in the study were clinically significant bacteriuria. If more than three kinds of microorganisms grew on plates, the result was regarded as contamination, and it was recommended to repeat material sampling. The identification of microorganisms was undertaken using MALDI-TOF, using mass-spectrometer Microflex (Bruker Daltonics, Germany). By using MALDI-TOF we made double (duplicated) applied cultures, grown on blood agar, on 96-cells steel panel (Bruker Daltonics, Germany) followed by air-drying. Then, we added 2 microliters of matrix: saturated solution of α -cyano-4-hydroxycinnamic acid in 50% acetonitrile and 2.5% trifluoroacetic acid (Bruker Daltonics, Germany) and re-dried at room temperature. Mass-spectra were calibrated using ribosomal protein *Escherichia coli* (bacteria standard). The protein spectra were analyzed using MALDI Biotyper (version 3, Bruker Daltonics, Germany). The results were expressed as the estimated coefficient (score) in the range from 0 to 3. At the values of score $> 1,7$ the result corresponds to a high degree of reliability to identify the genus, and at the values of score $> 2,0$ – to a reliable identification to species. At the same time, the value of score $> 1,7$ is assessed as the minimum value of the evaluation factor required for species identification [18]. At the values of score $< 1,7$ authentication result was considered as invalid and the study must be repeated.

Determination of the sensitivity of the isolated strains to antimicrobial agents was performed by disk methods in accordance with the recommendations of the Institute of Clinical and Laboratory Standards (CLSI 2012) [12]. Statistical processing was performed by calculating of the 95% confidence interval using WhoNet 6.3 program (World Health Organisation).

RESULTS AND DISCUSSION

845 samples of urine, obtained from pregnant women, were investigated. 387 samples gave no growth on the plates and the results were considered as negative. The growth of more than three microorganisms was obtained in 74 cases, and the result of research was regarded as

contamination and these were not processed further for bacteriological examination. Positive samples with a titer 10^6 - 10^8 CFU/ml were obtained in 87 pregnant women.

The study of etiological structure of the obtained strains showed that the most common pathogens were bacteria of Enterobacteriaceae group, and *Escherichia coli* strains dominated (55.17%), less frequently the following microorganisms were isolated: coagulase-negative staphylococci, enterococci, streptococci, fungi of *Candida* genus (fig. 1). Our data on the etiological structure of microorganisms at UTIs were confirmed by the results of numerous studies in obstetric practice [9, 11, 15].

Treatment of UTIs in women during pregnancy is a serious and unsolved problem in obstetric practice and nephrology. This is due to a limited selection of antimicrobial agents, which is because of possible transplacental transition and adverse effects on the fetus, the development of resistance of pathogens to the previously used drugs, and often latent or recurrent infections during pregnancy [8, 11, 17]. In this regard, we determined the sensitivity of the most frequent causative agent of UTIs *Escherichia coli* to antimicrobial drugs.

The most active antibacterial agents against *Escherichia coli* were fluoroquinolones (ciprofloxacin, levofloxacin, norfloxacin, ofloxacin) – 93% sensitive strains (95% CI 1.8-20.2). All *Escherichia coli* strains were sensitive to fosfomycin – the first-line therapy drug at UTIs in pregnant women today (95% CI 0.0-10.4). Also 93.3% (95% CI 0.0-10.4) *Escherichia coli* strains were sensitive to trimethoprim. Nitrofurantoin had activity 93.8% (95% CI 0.0-10.4). This drug for a long time been used in obstetric and nephrology practice in the 80s of the 20th century and the present time.

We obtained the following data when determining the sensitivity to penicillins and cephalosporins: 53.3% (95% CI 28.8-65.4) isolated strains were sensitive to ampicillin, 97,3% *Escherichia coli* strains were sensitive to ampicillin-sulbactam. All the strains were sensitive to ceftazidime, cefepime, meropenem and imipenem, aztreonam (95% CI 0.0-10.4).

The results were as follows when determining the sensitivity to aminoglycosides: all the tested *Escherichia coli* strains were susceptible to amikacin, isepamicin and netilmicin (95% CI 0.2-19.6). 95.2% of strains were susceptible to gentamicin and sisomicin (95% CI 0.8-17.5). Thus,

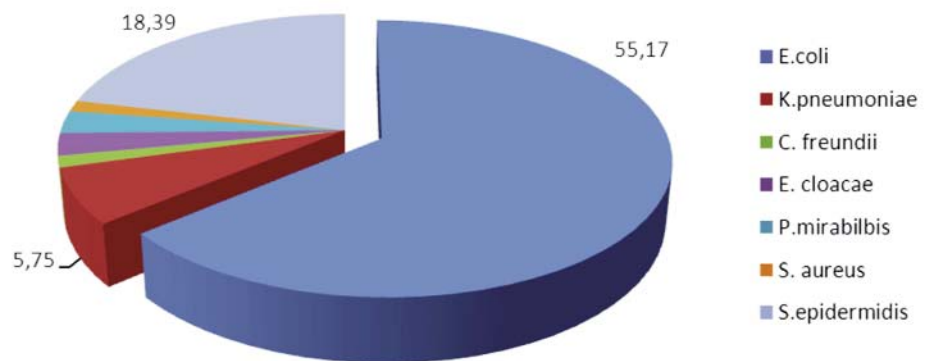


Figure 1 - Etiological structure of UTIs in pregnant women

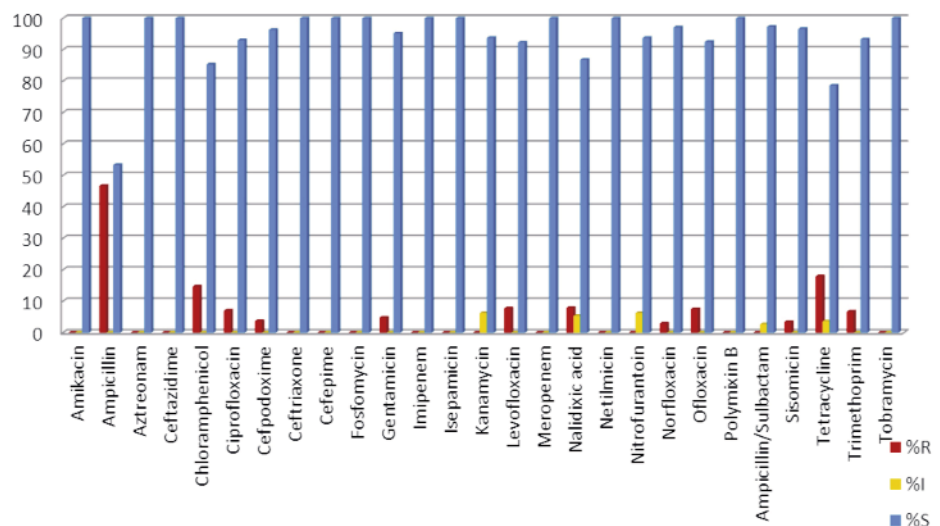


Figure 2 - Sensitivity of Escherichia coli to antimicrobial drugs

the prevalence of urinary tract infections in pregnant women was 22.5%.

CONCLUSION

Escherichia coli is the most common causative agent of urinary tract infections in pregnant women, as evidenced from the samples taken from a range of pregnant women. High antibacterial activity was registered in cephalosporins, fosfomycin, nitrofurantoin. Based on our findings, these antibiotics may be prescribed to pregnant women in order to address infections (this is in keeping with the 2014 FDA criteria).

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