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ORIGINAL RESEARCH

The Prevalence of Asymptomatic Bacteriuria Among Jordanian Pregnant Women

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Objective: Asymptomatic bacteriuria (ASB) is common in pregnant women and can lead to serious complications. During pregnancy, routine screening for bacteriuria is often not standard in many developing countries, leading to underdiagnosis. In Jordan, there is limited data on ASB in pregnant women. This study aimed to determine the prevalence, causative organisms, antibiotic sensitivity, and associations with maternal conditions and diseases.

Methods: This cross-sectional, prospective study conducted at Jordan University Hospital assessed the prevalence of ASB in pregnant women. Pregnant women aged ≥18 years at any trimester, without symptoms of urinary tract infection, and who had not utilized antibiotics for two weeks, were included. Midstream urine samples were collected, analyzed, cultured, and tested for antibiotic susceptibility in the hospital's Microbiology laboratory.

Results: Out of 319 women, 23.5% (n=76) showed positive urine culture. The chi-square and logistic regression analysis revealed that gestational age and grand multipara were associated with ASB. Neither diabetes nor hypertension (true or gestational) was associated with ASB. Fifty-one of the bacteriuria cases (68%) were positive for pure culture of *Escherichia coli*, and 4% were mixed with Gram-positive organisms. ASB due to Gram-negative organisms was significantly associated with older age group ($p = 0.025$). Furthermore, the antibiotic sensitivities of the isolated *E. coli* showed high percentages (≥90%) to nitrofurantoin, aminoglycosides, and some types of cephalosporins, but low (56%) to amoxicillin-clavulanic acid.

Conclusion: The prevalence of ASB among Jordanian women was higher than in developed countries and some low-to-middle-income countries, and ASB was significantly associated with gestational age and parity. These findings emphasized the importance of routine screening for ASB during pregnancy, particularly in the third trimester and in grand multiparous women. Although no significant association was found with maternal age or common comorbidities such as diabetes and hypertension, further extensive studies are needed.

Keywords: asymptomatic bacteriuria, gestational age, Jordan, parity, pregnancy

Introduction

Bacteriuria is colonization of one or more bacterial organisms in the urine and is identified in a significant number (≥10^5 colony-forming units [CFU]/mL). Bacteriuria can be symptomatic and indicative of a urinary tract infection, or it can be asymptomatic bacteriuria (ASB).[1–3] ASB is very common and can affect people of all age groups and both sexes, but its incidence increases with age and during pregnancy.[2–4]

During pregnancy, several anatomical and physiological changes in the urinary tract occur that facilitate bacteriuria.[5,6] Of these changes, the ureters and renal pelvis dilate, the urinary bladder is displaced, and progesterone induces smooth muscle relaxation, all of which lead to a decrease in ureteral peristalsis, an increase in bladder capacity, and urinary stasis. Additionally, changes in urine pH and osmolality, as well as pregnancy-induced glycosuria and aminoaciduria, may facilitate bacterial growth.[6]

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If ASB is left untreated, the adverse maternal outcomes, including symptomatic cystitis, pyelonephritis, preterm labor, and delivery, increase.[7–12] ASB is also linked to adverse fetal outcomes such as prematurity, low birth weight, and increased perinatal mortality.[10–12] However, when ASB is treated with appropriate antibiotics, the incidence of pyelonephritis and complications such as preeclampsia, preterm labor, low birth weight, and premature rupture of membranes is significantly reduced.[7,8,13] Although the Clinical Consensus document issued in 2023 by the American College of Obstetricians and Gynecologists[14] established a protocol and recommended that clinicians screen for ASB with a urine culture at least once during an early antenatal visit, this remains a non-standard operating procedure in many countries, particularly developing countries. As a result, there is limited understanding and diagnosis of this condition.

Furthermore, it has been documented that there is a substantial difference in the prevalence of ASB during pregnancy among different populations. For instance, the range of ASB during pregnancy in African countries ranged from 2 to 55%,[15–17] whereas in certain Asian countries, the percentages could range from 4.5 to 26.5%,[18–22] compared to 2 to 10% in industrialized nations.[13]

In Jordan, there were more than 181,991 births during 2022, with a birth rate of 20.681 births per 1000 people, according to the Department of Statistics.[23] Despite ASB's impact on maternal and fetal health and the high birth rate, there is limited data on the prevalence and management of ASB among pregnant women in Jordan. Addressing this gap can provide insights for improving antenatal care protocols in the region. Therefore, this study was conducted at Jordan University Hospital, a tertiary center and the largest teaching hospital in Jordan, to determine the prevalence of ASB during pregnancy, identify the causative organisms, and evaluate the association between common medical conditions and the relationship with pregnancy trimesters.

Method

Study Design

This study was a combination of a cross-sectional and prospective design, incorporating the patient's history with laboratory analysis of urine and culture. Subjects were asked to participate during their routine antenatal care visit at the obstetrics and gynecology outpatient clinics of Jordan University Hospital during February and March 2024.

Study Approval

A study protocol was submitted and approved by the University of Jordan Hospital's ethics committee, and each participant provided her informed consent to participate in the study. The study was conducted in accordance with the Declaration of Helsinki and the guidelines of Jordan University Hospital.

Study Population

Participants included pregnant women who met the following inclusion and exclusion criteria.

The inclusion criteria were pregnant women aged 18 years or older attending antenatal care clinics at Jordan University Hospital, pregnant women at any trimester, and those with no signs or symptoms of urinary tract infection (UTI). Exclusion criteria included women who used antibiotics within the past two weeks, the presence of any signs or symptoms of a UTI, and patients who declined to provide consent.

The sample size was calculated based on the assumption that the population has a probability between 25% and 28% of having ASB, with 95% confidence and a 5% margin of error. A total of 310 pregnant women were required to establish the sample size; however, 319 were enrolled.

Data Collection

Clinical and obstetric history were obtained and recorded in both the hospital system and a standardized form, ensuring participant privacy. Gestational age was calculated using either an ultrasound scan or based on the first day of the last menstrual period.

Urine Sample Collection and Analysis

Participants were educated on how to properly collect a clean-catch midstream urine sample. Approximately 15–20 mL of urine was collected from each participant, using the proper collection technique in a sterile container. The urine samples were sent to the Microbiology Department at the Jordan University Hospital for analysis, culture, and antibiotic sensitivity testing. Briefly, urine samples were cultured using a calibrated loop on blood and MacConkey agar plates. To check and ensure the efficacy and accuracy of laboratory results, reference standards from the Clinical & Laboratory Standards Institute were implemented. A positive culture is defined by having $\geq 10^5$ colony-forming units (CFU)/mL of a single isolate, except for group B streptococcus (10^4 CFU/mL). If two organisms were isolated and CFU for at least one of the species had CFU of $\geq 10^5$/mL and the second had CFU of $\geq 10^4$, both organisms were identified, and sensitivity testing was performed. Bacterial identification system and antibiotic sensitivity testing were performed using VITEK-2 Compact (bioMérieux, Marcy-l'Etoile, France). The results were collected by the research team and recorded for further analysis. Regarding the choice of antibiotics, a list of approved antibiotics during pregnancy is part of the protocol at the University of Jordan Hospital; however, the selection for treatment is left to the clinician and the pregnant woman's health, gestational age, and infant's safety.

Statistical Analysis

Data was processed using IBM SPSS Statistics 25. Continuous variables were expressed as mean ± standard deviation, and categorical variables were expressed using frequencies and percentages. Differences in the continuous variables were assessed using the *t*-test, and the frequencies were analyzed using the Chi-square test. A logistic regression analysis was performed to find the association between variables and bacteriuria to estimate the odds ratio and 95% confidence interval (%95 CI). A p-value < 0.05 was considered statistically significant.

Results

Frequency of ASB and Associated Variables Among Pregnant Jordanian Women

A total of 319 pregnant women were included in the study, with a mean age of 32.2 years (±5.5) and a gestational age of 20.4 weeks (±10.1). Two hundred and seventy-two (85.3%) were medically free of any disease condition, whereas 1.9% were diabetic, 5.6% had gestational diabetes, 1.6% were hypertensive, 0.3% had gestational hypertension, 4.7% had hypothyroidism, and 0.3% had hyperthyroidism.

The frequency of ASB was 23.5% (n = 75) in the studied population (Table 1). There was no difference in age or age groups between women with negative and positive bacteriuria. However, bacteriuria was associated with gestational age (p = 0.004). In addition, bacteriuria was associated with parity (p = 0.019). Neither diabetes nor hypertension (as total or gestational) was associated with bacteriuria.

The logistic regression analysis was performed on three variables: parity, gestational age, and maternal age. The results revealed that gestational age and parity were associated with bacteriuria (Table 2). The odds ratios for women in

Table 1 Characteristics of the Study Population, with the Differences Between the Two Groups: Negative for Bacteriuria and Positive for Bacteriuria

Parameter	Negative for Bacteriuria n = 244 (76.5%)	Positive for Bacteriuria N = 75 (23.5%)	P value**
Age (years) (mean ± SD)	32.2 ± 5.5	32.1 ± 5.3	n.s.
≤ 25 years	27 (77.1%)	8 (22.9%)	n.s.
26-35 years	146 (78.1%)	41 (21.9%)	
≥ 36 years	71 (73.2%)	26 (26.8%)	

(*Continued*)

Table 1 (Continued).

Parameter	**Negative for Bacteriuria n = 244 (76.5%)**	**Positive for Bacteriuria N = 75 (23.5%)**	**P value****
Gestational age (weeks) (mean ± SD)	19.1 ± 10.0	22.8 ± 9.9	0.005
1st Trimester	74 (80.4%)	18 (19.6%)	0.004
2nd Trimester	108 (82.4%)	23 (17.6%)	
3rd Trimester	62 (64.6%)	34 (35.4%)	
Medical Conditions			
Medically-Free	212 (77.9%)	60 (22.1%)	n.s.
Diabetes mellitus (DM)	3 (50%)	3 (50%)	
Gestational DM	12 (66.7%)	6 (33.3%)	
Hypertension	3 (60.0%)	2 (40.0%)	
Gestational hypertension	1 (100%)	0 (0%)	
Gestational hypertension and DM	1 (100%)	0 (0%)	
Hyperthyroidism	0 (0%)	1 (100%)	
Hypothyroidism	12 (80%)	3 (20%)	
Parity			
Primigravida	31 (67.4%)	15 (32.6%)	0.019
Primipara	58 (78.4%)	16 (21.6%)	
Multipara	143 (80.8%)	34 (19.2%)	
Grand multipara*	12 (54.5%)	10 (45.5%)	

Notes: *Women who had given birth to five or more live infants. **The p-value for means was calculated based on the *t*-test, whereas for 2×2 frequencies, the chi-square analysis was used, and for 2x3, the Pearson Chi-square was used.
Abbreviations: DM, diabetes mellitus; n.s., non-significant; SD, standard deviation.

the first and second trimesters to have bacteriuria were significantly lower than those of women in the third trimester. Furthermore, the odds ratio for multipara, but not for primigravida, to have bacteriuria is significantly less than that of grand multipara women (women who had given 5 or more live births) (Table 2). Although the odds ratio for primipara was comparable to that of multipara women in relation to grand multipara, the p-value did not reach a significant value.

Table 2 The Logistics Regression Analysis of Parity, Gestational Age, and Maternal Age to Have Bacteriuria Among Pregnant Jordanian Women with Asymptomatic Bacteriuria

Parameter	**Odds Ratio**	**95% CI**	**P value**
Parity			
Primigravida	0.698	0.230–2.118	0.525
Primipara	0.383	0.133–1.100	0.075
Multipara	0.336	0.130–0.873	0.025
Grand multipara*	Reference		

(*Continued*)

Table 2 (Continued).

Parameter	Odds Ratio	95% CI	P value
Gestational age			
1st Trimester	0.472	0.239–0.932	0.031
2nd Trimester	0.406	0.217–0.762	0.005
3rd Trimester	Reference		
Maternal Age (years)			
≤ 25 years	0.484	0.258–1.902	0.484
26-35 years	0.718	0.382–1.315	0.283
≥ 36 years	Reference		

Notes: *Women who had given birth to five or more live infants.

Isolated Bacterial Organisms in Jordanian Pregnant Women with ASB

Fifty-one of the bacteriuria cases (68%) were positive for pure culture of *Escherichia coli*, and 4% were mixed with Gram-positive organisms (2 cases with *Staphylococcus spp*. and 1 case with *Enterococcus spp*). Other Gram-negative isolated bacteria were *Klebsiella pneumonia* (2.7%) (Table 3). As for Gram-positive bacteria, the most commonly isolated type was *Staphylococcus spp*. (22.7%); however, almost half of these cultures were mixed with Enterococcus spp., and only one case (1.3%) was positive for methicillin-resistant *Staphylococcus aureus* (MRSA). Other organisms, such as pure *Enterococcus spp*. (2.7%) and *Streptococcus agalactiae* (2.7%) were also isolated.

Table 3 Isolated Organisms from Pregnant Jordanian Women with Asymptomatic Bacteriuria

Isolated Organism	Frequency	Percent
Escherichia coli (E.coli)	51	68.0
Klebsiella pneumoniae	2	2.7
Enterococcus faecalis	1	1.3
Enterococcus faecium	1	1.3
Staphylococcus aureus (MRSA)*	1	1.3
Staphylococcus haemolyticus	1	1.3
Staphylococcus spp. (coagulase negative)	2	2.7
Staphylococcus spp.	3	4.0
Streptococcus agalactiae	2	2.7
Mixed culture		
E. coli+ Staphylococcus spp.	2	2.7
E. coli + Enterococcus spp.	1	1.3
Staphylococcus spp. + Enterococcus spp.	8	10.7
Total	75	100

Abbreviation: *MRSA, methicillin-resistant *Staphylococcus aureus*.

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Table 4 Association of Gram-Type Bacteriuria with Age Group Among Jordanian Pregnant Women with Asymptomatic Bacteriuria

Age Group	Gram-Positive	Gram-Negative	P value*
≤ 25 years	5 (62.5%)	3 (37.5%)	0.025
26-35 years	7 (17.1%)	34 (82.9%)	
≥ 36 years	7 (26.9%)	19 (73.1%)	

Notes: *The Pearson Chi-square was used.

Table 5 Antibiotic Sensitivity of the Isolated *E. coli* Versus All Types of Bacteria from Jordanian Pregnant Women with Asymptomatic Bacteriuria to Different Classes of Antibiotics

Antibiotic	*E. coli*	Overall
	% Sensitivity	
Amikacin	60.0	69.2
Amoxicillin-Clavulanic	55.6	69.2
Ceftazidime	66.7	75.0
Ceftriaxone	93.9	95.7
Cefuroxime	89.6	89.9
Ciprofloxacin	87.5	84.5
Gentamicin	100.0	98.4
Nitrofurantoin	91.8	94.4

ASB due to Gram-negative organisms was significantly associated with the age group ($p = 0.025$) (Table 4). It was apparent that bacteriuria in pregnant women under 26 years of age was associated with low percentages of Gram-negative bacteria, whereas Gram-negative bacteriuria was isolated more frequently in pregnant women above 26 years of age.

Furthermore, the antibiotic sensitivities of the isolated *E. coli* to different classes of antibiotics were similar to those of all types of bacteria, with high percentages of sensitivity to nitrofurantoin, aminoglycosides, and some types of cephalosporins (Table 5). On the other hand, the percentage sensitivity to amoxicillin-clavulanic acid was low.

Discussion

The prevalence of ASB in Jordanian pregnant women was 23.5%, and was significantly higher in the third trimester group (35.7%), and in women with grand multipara (45.5%), followed by primigravida (32.6%). *E. coli* was the most frequently isolated bacterium, accounting for 72%, and more than 90% of these isolates were sensitive to nitrofurantoin and ceftriaxone, which are considered safe types of antibiotics to take during pregnancy.

The ASB prevalence in the present study is two to five times higher than studies from developed countries such as the Netherlands 2.3 to 4.7%,[24] United Kingdom (8.04%),[4] United States (9.5%),[25] and Australia (4.9%).[26] Furthermore, the prevalence of ASB was approximately twice from other low to middle income countries such as Ethiopia (13.8%)[27] and 16.9%,[28] Kenya (16.3%),[29] Egypt (10%),[30] India (13.2%),[20] Pakistan (17.1%),[31] Bangladesh (4.5%)[19] and (16.5%),[18] but similar to a study from south India (24.5%),[32] Pakistan (26.5%)[21] and much less than from studies from Nigeria (45%)[16] and (55%).[17] Although several studies included factors such as environmental, social habits, socioeconomic status, personal hygiene standards, and education levels that could be attributed to ASB prevalence differences between different populations, the present study emphasizes the importance of the percentage of women in the third trimester and the percentage of women with grand multipara, which are additional contributing factors in these differences.

In the present study, the percentage of women in their third trimester was 30.1%. Among these women, the percentage of ASB was 35.4%, compared with 19.6% and 17.6% for first- and second-trimester women, respectively. On the contrary, the study from Bangladesh[19] involved all women who were less than 20 weeks pregnant, and the study from India involved women in their 15–16 weeks of gestation.[20] Similar to the present findings, other studies have shown a higher prevalence of ASB in the third trimester, but the percentage of women in the third trimester in these studies was low.[29,33,34] In a multicenter study in Pakistan, 48.1% of the studied population was in the third trimester of pregnancy, and the prevalence of ASB was 26.5%.[21] These figures indicate that the prevalence of ASB is higher in women in the third trimester, and it is a contributing factor to the prevalence differences between the studied populations. The latter finding is consistent with the anatomical and physiological changes in the urinary tract system during pregnancy that prevail during the third trimester, such as urinary stasis, which increases the risk of bacteriuria more than in the first and second trimesters.[5,6] Therefore, the higher bacteriuria incidence in the third trimester emphasizes the importance of routine screening and timely treatment to prevent complications such as preterm labor and pyelonephritis.

In the present study, 22 women were grand multiparas, and 10 (45.5%) of them had ASB, with 60% of these cases occurring in the third trimester. Therefore, grand multipara is significantly associated with ASB, especially in the third trimester. Similar findings were reported by others,[35,36] where they found that grand multiparous women had the highest percentage of ASB. In a murine model, it was demonstrated that multiparity was associated with a significantly increased risk of chronic high-titer *E. coli* cystitis and ascending pyelonephritis, suggesting that childbearing causes permanent changes in the urinary tract, which increases susceptibility to bacteriuria.[37]

Similar to most studies from different populations, the most causative ASB in pregnant Jordanian women was *E. coli* (68% pure and 4% mixed growth), followed by Staphylococcus spp. (22.7%, almost half of these were mixed growth).[17,27,28,30–34] Although the number of pregnant women with ASB in the ≤ 25 years old group was small, an interesting observation was that Gram-positive bacteria were statistically more common causes of bacteriuria in this age group than in older pregnant women. In Ordindo et al[29] study, the majority of the population (60%) tested was below 24 years of age, and 56.8% of the women's cases with ASB were caused by Gram-positive bacteria. Furthermore, the isolated *E. coli* from the ASB were mainly sensitive to nitrofurantoin, aminoglycosides, and some types of cephalosporins and showed considerable resistance to other types of cephalosporins (eg ceftazidime) and amoxicillin with clavulanic acid. These results are equivocal to the previous studies performed in Jordan on acne and nasal swab isolated organisms[38,39] and raise a major concern regarding the emergence of resistant bacteria to the safe choice of antibiotics in pregnancy. The emergence of resistant strains is partly due to the inappropriate prescription of antibiotics and the increased use of antibiotics during the COVID-19 pandemic.[40]

Our study found no significant association between ASB and common maternal comorbidities, including diabetes mellitus, gestational diabetes mellitus, hypertension, or hypothyroidism. These findings align with other studies that reported no significant difference in the prevalence of ASB between mothers with and without diabetes or gestational diabetes.[19,24,28] Conversely, existing literature presents conflicting evidence regarding the relationship between bacteriuria and hypertension. For instance, a study by Marlina et al[41] reported a significant association between hypertension and bacteriuria, suggesting a potential link that warrants further investigation. The latter study highlights the need for additional large-scale, multi-center studies to better understand the role of maternal comorbidities in the development of ASB during pregnancy.

This study has limitations. Although the sample size is considerable, it may not be sufficient to detect minor yet clinically significant associations, particularly regarding past medical conditions. However, a larger and more diverse cohort would enhance the generalizability of the findings. Other potential confounding variables that could impact urine culture results, such as body weight, dietary practices, socioeconomic status, or a history of recurrent urinary tract infections, were not considered in the study design. Future research should incorporate these factors to gain a more comprehensive understanding of risk factors. Due to the study's focus on a single demographic, the findings may not be representative of other regions or healthcare environments. Multicenter studies with a broader demographic representation are needed to confirm these results and improve their generalizability.

Conclusion

In conclusion, this study showed that the prevalence of ASB among Jordanian women was higher than in developed countries and some low-to-middle-income countries, and ASB was significantly associated with gestational age and parity. These findings emphasized the importance of routine screening for ASB during pregnancy, particularly in the third trimester and in grand multiparous women. Although no significant association was found with maternal age or common comorbidities such as diabetes and hypertension, further extensive studies are needed. The findings may support the use of universal screening; however, larger, multicenter studies are needed to confirm the results and identify additional risk factors that may lead to improved maternal and fetal outcomes.

Data Sharing Statement

The datasets used and/or analysed during the current study are available from the corresponding author upon request.

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Disclosure

The authors declare that they have no conflicts of interest.

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