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Factors affect the prevalence of vaginal infection and susceptibility pattern of bacterial isolates among pregnant women in Zliten city

Abstract

Vaginitis is the infection in the vagina, bacterial vaginosis is the most common infection among women, especially pregnant women. It occurs due to the disturbance of the microbiome in the vagina *Lactobacilli spp.*, which affects the growth of any microorganisms pathogenic.

Objectives: The study determined prevalence of bacterial infections in vagina, and their relationship to age, education levels and discharge abnormal. Identification and isolation bacterial infection to assess the incidence of their antimicrobial pattern in pregnant women, attending private clinics in Zletin city, Libya.

Material and methods: A descriptive study was conducted on 115 pregnant women during period from March 2016 to December 2016. Pregnant women were screened for high vaginal swabs to isolate and identify pathogenic bacteria. By microscopic examination (wet mount, gram stain). Culture by (blood agar, chocolate agar and MacConkey agar), macroscopic examination (bacteria hemolysis on blood agar, ferment and nonferment on MacConkey agar and catalase test), identification and antibiotics sensitivity patterns by BD Phoenix™ system.

Statistical analysis was performed using SPSS *P*. value < 0.05 was considered statistically significant.

Results: (77.39%) of pregnant women detected with frequency high vaginal swabs were bacterial infection, the highest infection was found in the age group (20-25) years old, the higher percentage of infection was between the (secondary education level) which found the percentage of this level was (47.2%) from the total, the highest infection rate was recorded with consistent thick white discharge was represented by 44 samples (49.4%) of the total samples. There are several different bacterial species were isolated, of which the most prevalent is Enterococcus faeca*lis* 16 (17.8%), followed by *Klebsiella pneumonia* 14 (15.55%), *Escherichia coli 12* (13.33%), and *Staphylococcus aureus* 10 (11.11%). The antibiotic Levofloxacin was the most sensitive agent against Gram-negative bacteria, and Linezolid was the most sensitive against gram-positive bacteria, additional Cefepime, Daptomycin, and Vancomycin were the most sensitive against gram-positive bacteria *streptococcus spp*.

Keywords: bacteria isolated, vaginitis, antibiotic susceptibility, Zliten

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Introduction

The human body contains as much bacteria as commensalism normal micro flora. These bacteria live outside and inside, including the vagina. The microbial can affect human health through nutrition, immunity and pathogen resistance.[1–3] The vagina is an important part of the female genital system that contains the normal flora in the lower reproductive tract. The complex bacterial community found in the human vagina is referred to as the vaginal microbiome, which plays an important role in a woman's reproduction and the health of the newborn. But when imbalances occur in these microorganisms, it may result in many negative conditions such as bacterial vaginosis, increased exposure to the pelvis inflammatory disease increased transmission of pathogens of Sexually Transmitted Diseases such as *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and human immunodeficiency virus.[3–6] The female reproductive system is considered one of the important organs because it is related to pregnancy and childbearing. It includes a variety of organs and epithelial surfaces. The external genitalia include mons pubis, labia majora, labia minora, bartholin's glands and clitoris. The area that contains these organs is called the vulva. The internal reproductive organs form a pathway (reproductive system), and this pathway consists of the vagina, uterus, and fallopian tubes, in addition to the ovaries first described syndrome *Haemophilus vaginalis*. It was concludere that vaginal infection is sexually transmitted diseases and the causative agent is *Haemophilu vaginalis*. Until it was renamed *Gardnerella vaginalis*, which was named after the scientist who discovered it.[7] In contrast Bacterial Vaginosis was identified a polymicrobial condition brought on by a complex alteration of the normal vaginal microbiota, which increases the number of anaerobic bacteria (Spiegel, 1991). One of the most common causes of vaginitis in women is a condition characterized by some common symptoms including odour vaginal discharge (often smells fishy) and abnormal discharge usually (white or grey) in colour (Ahmed et al., 2014).[8] The increased vaginal PH due to the replacement of *lactobacilli spp.,* which is the normal flora of the vagina by bacteria anaerobic species.[9] These microorganisms include *Gardnerella vaginalis, Mycoplasm hominis, Mobiluncus spp, Bacteroides spp*. and *Prevotella spp*.[10–13] The vaginal infection microbiome can be caused by *Gardnerella vaginalis* virulence factors such as adherence to epithelial cells, biofilm production,[14] cytolysin

activity,[15] and sialidase activity.[16] There are bacterial pathogens associated with vaginal infection and they are *Escherichia coli, Staphylococcus aureus, Streptococcus pyogenes*, *Enterococcus faecalis, Clostridium perfringens, Klebsiella spp*, *Streptococcus spp*.[8,17,18] Bacterial vaginosis occurs in up to 20% of pregnant women (Ahmed et al., 2014). Vaginal bacterial infection is one of the risk factors that cause pregnancy complications of premature rupture of membranes, spontaneous abortion, chorioamnionitis and other complications.[11,19] Several factors can be associated with an increased rate of vaginal colonization by bacterial infections and can disturb the balance of normal flora vaginae such as irregular sugar, antibiotics inhibiting normal flora, poor personal hygiene, the use of irritating detergents, use of high oestrogen and oral contraceptives.[8,17,20]

Materials and methods

Culture and identification procedures

This study was conducted at the department of Medical Laboratories and Microbiology Unit in Zliten Teaching Hospital, during the period from March 2016 to December 2016. The study population included 115 pregnant women with mild to moderate symptoms, their ages ranged from about less than of 20 years to over 40 years. It was completed by obtaining consent from all patients after clarifying the purpose of this study, before collecting samples. Aquestionnaire was created to collect much information as possible for the cases from which samples are taken in a special form designed for this study. It contains some parameters such as age, history of the disease, educational level and symptoms of the disease such as the presence of secretions, the color and consistency of the secretions and others (the questionnaire form is attached) see appendix D.

Collection of vaginal swabs

Samples collection by using two sterile vaginal swabs for each patient using a vaginal speculum one of the swabs to be used for direct examination, and the other one for culture media.

Diagnosis bacterial isolates and antibiotic sensitivity

Pure isolates of the bacterial pathogen were preliminarily characterized by some tests. Identification of bacteria to genus and/or species level was carried out by using BD Phoenix™ Automated Microbiology System which is intended for the *in-vitro* using rapid identification (ID) and antimicrobial susceptibility testing (AST) of clinically significant bacteria. The BD Phoenix™ system provides rapid results for most aerobic and facultative anaerobic gram-positive bacteria as well as most aerobic and facultative anaerobic gram-negative bacteria of human origin.

Results

This study was conducted on different infections of vaginal bacteria in the city of Zliten. The samples were selected from pregnant Libyan women, most of whom suffer from symptoms of vaginal symptoms such as excessive secretionsor vaginal itching, foul odour and lower abdominal pain. The present study aimed to detect the frequency of High Vagina Swab and susceptibility patterns of bacterial isolates among pregnant women who attended Zliten Clinics during the study period, out of the 115 pregnant women registered in these clinics agreed to participate in this study.

The age of pregnant women which have bacterial infection ranged less than 20 years to more than 40 years old, where the highest infection was found in the age group (20-25) years old, with a percentage of (30.3%) of the total infected, followed by the age group (31-35) years old, where the infection rate was (23.6%) of the total cases of infection, followed by the age group (26-30) years old, at a rate of (18.0%), the age group (36 and over) years old found a rate of (14.6%), and the age group of (20>) years old was the least affected among the registered age groups with a percentage of (13.5%). *p*-value = 0.639 (Table 1).

Table 1 The distribution of bacterial infection status regarding age for the study population

Age in years	Bacterial infection status				p-value
	Absent		Present		
	Count	%	Count	%	
< 20	2	7.7	12	13.5	0.639
20-25	10	38.5	27	30.3	
26-30	7	26.9	16	17.9	
31-35	4	15.4	21	23.6	
36 or more	3	11.5	13	14.6	
Total	26	22.6	89	77.4	

The educational levels are divided into three levels, the first level (Primary education), the second level (Secondary education) and the third level (university education and above). The secondary educational level for pregnant women which have bacterial infection were higher than other levels, the percentage which found of this level was (47.2%) from the total for infection, followed by the first level the infection per cent was (33.7%) from the total of infection. The third level represented (19.1%) from the total infection. *P* = 0.909 (Table 2).

Table 2 The distribution of bacterial infection status regarding educational level for the study population

Educational Level	Bacterial infection status				p-value
	Absent		Present		
	Count	%	Count	%	
Primary education	9	34.6	30	33.7	0.909
Secondary education	13	50	42	47.2	
University or above	4	15.4	17	19.1	
Total	26	22.6	89	77.4	

Vaginal secretions are considered normal unless they are accompanied by secretions of abnormal colors (white, yellow and green) accompanied by unpleasant odours, they indicate the presence of health problems. In our study were secretions of different colors were detected, where the white color majority of infections 37 samples (41.6%) of the total bacterial infection , followed by color yellow secretions in 28 samples (31.4%) of the total bacterial infection, *P* = 0.835 (Table 3).

Table 3 The distribution of bacterial vaginosis status regarding discharge colour for the study population

Discharge Colour	Bacterial infection status				p-value
	Absent		Present		
	Count	%	Count	%	
Yellow	7	26.9	28	31.5	0.835
White	13	50	37	41.6	
Green or Gray	6	23.1	24	26.9	
Total	26	22.6	89	77.4	

Abnormal vaginal discharge is one of the diagnostic signs that indicate the presence of infection, usually accompanied by pain or itching in the vagina. These are symptoms of a problem. The highest infection rate was recorded with consistent thick discharge was

represented by 44 samples (49.4%) of the total samples, followed by consistency frothy, which were represented by 28 samples (31.5%) of the total samples, and consistency watery which were represented by 17 samples (19.1%) of the total number of samples p-value = 0.256 (Table 4).s

Table 4 The distribution of bacterial infection status regarding discharge consistency for the study population

Consistency of Discharge	BV status				p-value
	Absent		Present		
	Count	%	Count	%	
Watery	7	26.9	17	19.1	0.256
Frothy	4	15.3	28	31.4	
Thick	15	57.7	44	49.4	
Total	26	100	89	100	

Prevalence rates of various types of bacterial infections

More than 20 different bacterial species were isolated, and the most dominant causative agent of high vaginal swabs were percent gram positive bacteria (51.1%) and gram negative bacteria (48.9%). All bacteria isolated were single growth except one case was mixed. The high percent bacteria were *Enterococcus Faecalis* 16(17.8%), followed by *Klebsiella pneumonia* 14(15.55%), *Escherichia coli* 12(13.33%), *Staphylococcus aureus* 10(11.11%), and *Staphylococcus haemolyticus* 8 (8.9%), this study have Showed that the Gram-negative isolates were most sensitive to Levofloxacin 43(97.72), followed by Cefepim 38(86.36), Ceftazidime, Amikacin 37(84.1%), Ciprofloxacin 36(81.81),Meropenem, Piperacillin-Tazobactam 34(77.72%), Gentamicin, Azetronam, Imipenem, Ertapenem 33(75%) respectively, Nitrofurantoin, Ceftriaxone 28(63.63) the Gram-negative isolates showed low response to Cefuroxime 20(45.45), Cephalothin 17(38.63), Amoxicillin-Clavulanate 15(34.1), Ampicillin 4(9.1%). In addition the current study have showed also that bacteria (*Streptococcus spp*) isolates are highly sensitivity to Cefepime, Daptomycin, Vancomycin 5(100%), Levofloxacin, Clindamycin, Meropenem 4(80%), Amoxicillin, Penicillin G 3(60%). This study resistance to Erythromycin, Cefotaxime, Chloramphenicol 2(40%), and Cefuroxime 0(00%).

Discussion

In this study highlights the prevalence of bacterial vaginal infections among pregnant women in Zliten, Libya, with significant findings regarding the distribution of infections across age groups, educational levels, and bacterial species.

Our findings indicate that the highest infection rate was observed among women aged 20–25 years (30.3%), which aligns with studies by Ibrahim et al.[11] and Afolabi et al.,[21] who identified increased risk in this reproductive age group due to heightened sexual activity and elevated hormonal levels. Conversely, the lowest infection rate (13.5%) was recorded in women aged 36 years and above, likely due to reduced sexual activity and declining levels of glycogen, estrogen, and the maintenance of normal vaginal pH, as reported by Bhakta et al.[22] in their study

Our study also found that secondary education level accounted for the highest proportion (47.2%) of infections, a finding consistent with Ibrahim et al.[11] Shayo et al.[23] reported a similar trend, with the highest infection rates observed among women with primary and secondary education levels, suggesting that education may influence hygiene practices and healthcare awareness.

A variety of bacterial species were identified, with gram-positive bacteria comprising 51.1% of isolates and gram-negative bacteria accounting for 48.9%. This is consistent with findings from Dahash[24] in Libya. The most frequently isolated bacteria in this study, **Enterococcus** spp., align with findings by Atia.[25] Similarly, **Escherichia coli** was isolated in 13% of cases, closely matching studies by Sáez-López et al.[26] in Spain and Mostafa et al.[27] in Menoufia, Egypt.

However, some discrepancies were noted when comparing the prevalence of bacterial species with other studies. For example, Mohammed et al.[28] in Iraq reported higher isolation rates of **E. coli** (21.6%) and **Staphylococcus aureus** (23.8%) than observed in this study. Additionally, **Pseudomonas aeruginosa**, reported in Iraq by Mohammed et al.[28] and Dahash,[24] was not isolated in our study. These variations may be attributed to differences in geographic, demographic, and methodological factors.

The prevalence of **Klebsiella pneumoniae** in our study aligns with findings by Razzak et al.[29] and Ali[30] but diverges from Dahash,[24] who reported **Klebsiella** spp. as less prevalent. Similarly, **Streptococcus agalactiae** (Group B Streptococcus) was identified in our study, consistent with findings by Sahar and Al-Saliem[31] and Dashtizade et al.,[32] but in disagreement with Khan et al.[33]

The antibiotic susceptibility testing revealed that **Streptococcus agalactiae** isolates were highly sensitive to vancomycin, linezolid, and cefepime, findings that align with studies by Sadaka et al.,[34] Atia,[25] and Shabayek et al.[35] However, resistance to erythromycin was observed, which is consistent with previous research and underscores the need for careful antibiotic selection.

Conclusion

Our study found a several different bacterial species were isolated, of which the most prevalent is *Enterococcus faecalis*, *Klebsiella pneumonia*, *Escherichia coli*, and *Staphylococcus aureus* respectively, Vancomycin, Linezolid and Cefepime were the most effective antibiotic for bacterial vaginosis as a very prevalent among pregnant women in the study area, where routine culture of vaginal samples must be performed on women that attending to gynecology clinics. Therefore, monitoring the action of antibacterial medications is of concern mainly among pregnant women as the choice of treatment is limited by their side effects, and a proper use of antibiotics should be encouraged.

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Conflicts of interest

No potential conflict of interest was reported by the author(s).

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