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ORIGINAL ARTICLE

# Bacterial vaginosis and Mycoplasma infections in reproductive-age women: Clarifying the association with risk factors

**S. Eroglu**[a,*], **N. Asgin**[b]

[a] *Semra Eroglu, Samsun University, Department of Gynecology and Obstetrics, Samsun, Turkey*
[b] *Nergis Asgin, Karabuk University Faculty of Medicine, Department of Microbiology, Turkey*

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**KEYWORDS**
Bacterial vaginosis;
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Prevalence;
*Ureaplasma urealyticum*

**Abstract**
*Objectives:* This study seeks to examine the association between predisposing risk factors and the prevalence of bacterial vaginosis (BV) as well as *Mycoplasma hominis* (MH) and *Ureaplasma urealyticum* (UU) infections in reproductive age women and investigate its relationship with infertility.
*Methods:* This cross-sectional, prospective study was carried out using sexually active females who presented at the Gynaecology Clinic with complaints of vaginal discharge. Two cervical smear samples were taken from the endocervical canal using sterile cotton swabs for each patient. The patients were questioned to obtain their demographic data and potential risk factors for lower genital tract infections, and their responses were recorded.
*Results:* Of 348 patients, BV was detected in 46.3%, UU in 26.7%, MH in 3.7% and UU and MH co-infection in 13.2%. The prevalence of BV concomitant with UU and/or MH was significantly high ($p = .001$). The most prominent risk factors for BV were UU and MH infection (AOR = 6.79, 95% confidence interval (CI): [2.63–17.56]), vaginal douche use (AOR = 6.80, 95% CI: [03.60–12.83]), abortion history (AOR = 2.82, 95% CI: [1.55–5.12]) and high body mass indexes (BMI) (AOR = .81, 95% CI: [.74–.89]). The prevalence of BV, UU and MH was significantly higher in infertile patients than fertile patients ($p = .002$).
*Conclusions:* Bacterial vaginosis, MH, and UU co-infection were common in patients with vaginal discharge, and it was detected considerably higher in infertile patients than in fertile patients.


* Corresponding author.
*E-mail address:* drsemraeroglu@gmail.com (S. Eroglu).

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**PALABRAS CLAVE**
Vaginosis bacteriana;
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Prevalencia;
*Ureaplasma urealyticum*

**Vaginosis bacteriana e infecciones por micoplasma en mujeres en edad reproductiva: aclarando la asociación con factores de riesgo**

**Resumen**
*Objetivos:* Este estudio busca examinar la asociación entre los factores de riesgo predisponentes y la prevalencia de la vaginosis bacteriana (VB), así como las infecciones por *Mycoplasma hominis* (MH) y *Ureaplasma urealyticum* (UU) en mujeres en edad reproductiva e investigar su relación con la infertilidad.
*Métodos:* Estudio transversal y prospectivo que se llevó a cabo con mujeres sexualmente activas que acudieron a la Clínica de Ginecología con quejas de flujo vaginal. Con hisopos de algodón estériles, se tomaron dos muestras de frotis cervical del canal endocervical. Se interrogó a las pacientes para obtener sus datos demográficos y se registraron los posibles factores de riesgo de infecciones del tracto genital inferior y sus respuestas.
*Resultados:* Entre 348 pacientes, se detectó VB en el 46,3%, UU en el 26,7%, HM en el 3,7% y coinfección por UU y HM en el 13,2%. La prevalencia de VB concomitante con UU y/o MH fue significativamente alta ($P = 0{,}001$). Los factores de riesgo más destacados para la VB fueron la infección por UU y MH (AOR = 6,79, intervalo de confianza (IC) del 95%: [2,63-17,56]), uso de duchas vaginales (AOR = 6,80, IC del 95%: [03,60-12,83]), antecedentes de aborto (AOR = 2,82, IC del 95%: [1,55−5,12]) e índices de masa corporal (IMC) altos (AOR = 0,81, IC del 95%: [0,74-0,89]). La prevalencia de BV, UU y MH fue significativamente mayor en pacientes infértiles que en pacientes fértiles ($P = 0{,}002$).
*Conclusiones:* Se encontró que la coinfección por BV, MH y UU era común en pacientes con flujo vaginal, y también este aumento fue significativamente mayor en pacientes infértiles que en pacientes fértiles.


## Introduction

The vaginal flora is a balanced system containing lactobacilli and numerous other bacteria. The lactobacilli found in this system have a protective function with acidic pH. As a result of physical and chemical irritation, a decrease in the density of lactobacilli occurs, the vaginal pH increases, anaerobic microorganisms proliferate, and bacterial vaginosis (BV) develops. Although BV is usually asymptomatic, it is the most common cause of vaginal discharge in reproductive age.[1] BV has been reported at a rate of 17−30.7% in Turkey, 29% in the US, 50% in patients of African-American ethnicity, and 15−20% in pregnant women.[2,3] A relationship has also been established between cervicitis and post-hysterectomy vaginal-cuff cellulitis, postabortal infection, endometrial bacterial colonization, and plasma cell endometritis.[4] These have been linked with the incidence of missed abortions in early pregnancy, preterm premature membrane rupture, endometritis, preterm labor, and postpartum fever.[5] Other risk factors that increase the incidence of BV include being of the African race,[6] smoking,[7] having sexual multiple partners, female-female sexual intercourse,[8] vaginal douching,[9] obesity,[6] and estrogen deficiency.[7] However, these issues have received little attention in Turkey.[10]

The *Mycoplasma hominis* (MH) and *Ureaplasma urealyticum* (UU) are small intracellular bacteria. They are often isolated in vaginal smears and urine samples of healthy, sexually active males and females. They have been reported to be associated with urethritis, cervicitis, cystitis, pelvic inflammatory disease, and endometritis.[5] They can also lead to preterm births, spontaneous abortions, tubal factor infertility, and an increase in HIV transfer.[8−12]

It has been estimated that there is co-infection between BV, MH, and UU, causing chronic pelvic infections in women of reproductive age.[13] In this context, the present study determined the prevalence of BV, MH, and UU; their co-infection rates, and associated risk factors in sexually active HIV-negative women of reproductive age.

## Methods

### Study design, sampling technique, and sample size

Approval for this prospective, cross-sectional study was granted by the Non-interventional Ethics Committee of Karabuk University (decision no: 7/10, date: 04.07.2018). Informed consent was obtained from all the study participants.

The patients included were females aged 18−45 years who presented at the Karabuk University Training and Research Hospital Gynaecology Polyclinic between July and December 2018 with complaints of vaginal discharge. They were all sexually active and HIV-negative. Patients were excluded from the study if they were pregnant, had any genetic disease, anatomic disorder, endocrine disease, had used antibiotics in the previous month, or did not wish to participate in the study.

### Questionnaire survey

A record was made for each patient regarding their age, gravidity, parity, number of abortions, and body mass index (BMI). The patients were questioned about their level of education, history of preterm birth, infertility, and type of delivery at birth. BMI was calculated as weight in kilograms divided by height in meters squared. According to the European Society of Human Reproduction and Embryology definition, infertility was defined as no pregnancy despite 12 months or more of sexual intercourse without contraception. Preterm birth was accepted as birth at 20–37 gestational weeks. Also, questions related to potential BV risk factors in the literature were included.[14,15] The questionnaire added smoking, vaginal douche habits, and contraception methods (e.g., withdrawal method, condom, oral contraceptive, intrauterine device (IUD), injectables, and tubal ligation). The patients were asked about vaginal discharge, dysuria, itching, dyspareunia, and lower abdominal pain. All their responses were recorded.

### Sample collection and microbiological analysis

Two cervical smear samples were taken from the endocervical canal using sterile cotton swabs. The samples were placed in Stuart Transport medium (COPAN, Brescia-Italy) and transferred to the Microbiology Laboratory for processing.

Gram staining was applied to the first swab to investigate the presence of BV, and the stained smear was examined under a microscope at $\times$100 magnification. The preparations were evaluated according to the Nugent scoring system.[16] Cervical smears scored as $\geq$7 points were considered as positive.

The second swab was used for MH and UU cultures. The samples were processed using the colorimetric Autobio Mycoplasma IES Kit (Autobio Diagnostics Co. Ltd., Zhengzhou, China) that utilizes biochemical tests. The test kit contain 30-well strips for two different concentrations of 11 antibiotics. Also, it can detect the MH and UU simultaneously. The test procedure was performed according to the manufacturer's instructions.

### Data analysis

The study was an institution-based, cross-sectional study using a systematic sampling technique. The sample size was determined based on the single population proportion formula with a seroprevalence value ($p$ = 42%) taken from a previous study from Turkey with a desired absolute precision of 5% and 95% confidence intervals (CI).[10] The final study population size was 348.

### Statistical analysis

The data were analyzed statistically using the SPPS 20 software (IBM Corp. Released 2011. IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY, US). Descriptive statistics were stated as mean $\pm$ standard deviation values and number ($n$) and percentage (%). Variance homogeneity from the assumptions of parametric tests was assessed with the Levene test. Conformity of the data to normal distribution was evaluated with the Shapiro–Wilk test. To evaluate differences between 2 groups, the Student's $t$-test was performed on data showing normal distribution and the Mann–Whitney $U$ test on data not showing normal distribution. For the comparison of 3 or more groups, One-way Variance analysis and the Tukey test or the Kruskal–Wallis test with Bonferroni correction was used as appropriate. Categorical data were analyzed using Fisher's Exact test and the Chi-square test. Binary logistic regression analysis was used to determine correlations between independent and dependent variables. Values of $p < 0.05$ and $p < 0.01$ were considered statistically significant.

## Results

Out of the 348 patients, BV positivity was found in 46.3%, UU in 26.7%, MH in 3.7% and UU and MH co-infection in 13.2%. Among patients, 20.9% ($n$ = 73) of women were infertile and 79.1% ($n$ = 275) were fertile group. The mean age of the patients was 36.68 $\pm$ 8.66 years (range, 18–45 years), and the most common age range was 26–35 years (40.5%). The most common complaint on presentation was vaginal discharge (62.1%, 62.73%), followed by vaginal itching (23.9%), pelvic pain (10.15%), and dysuria (1.1%). Almost all the patients were married, the vast majority (77.9%) were housewives, and the most common education level was high school (37.9%). In the fertile group, previous births were reported as normal vaginal delivery in 59.2% and as cesarean section in 28.4%; however, 12.4% of the participants had not given birth. According to the Nugent score, the patients were separated into BV positive or negative groups. BV positivity was significantly increased at a younger age and increased the number of abortions ($p$ = 0.03, $p$ = 0.04). The demographic characteristics of the participants are shown in Table 1.

Almost all (97.4%) of the patients had one sexual partner. BV prevalence was significantly low in patients who were university graduates ($p$ = 0.016) (Table 2). Vaginal douche was used by 65%, and BV positivity was extremely high in such patients ($p$ = 0.001). The most common contraception method was IUD (15.5%). BV prevalence was significantly low in patients who used the withdrawal method, condom, or tube ligation ($p$ = 0.043). BV prevalence was considerably high in infertile patients ($p$ = 0.002) (Table 2).

BV positivity with the co-infection of UU and MH was significantly high ($p$ = 0.001). Women in UU and MH positive groups were significantly younger and had higher BMI than those negative for UU and MH ($p$ = 0.001, $p$ = 0.001). The UU and MH positivity was remarkably low at an older age ($\geq$46 years) ($p$ = 0.022) and significantly high in those with a history of preterm birth ($p$ = 0.003), those with vaginal douching habits ($p$ = 0.001), patients who smoked ($p$ = 0.037) and infertile ($p$ = 0.002) (Tables 3 and 4).

The multivariate logistic regression analysis showed that the most significant risk factors for BV were the presence of Ureaplasma and Mycoplasma (AOR = 6.79, 95% CI: [2.63–17.56]), vaginal douching (AOR = 6.80, 95% CI: [03.60–12.83]), a history of abortion (AOR = 2.82, 95% CI:

**Table 1** Relationships between demographic characteristics and bacterial vaginosis.

| Characteristic | BV positive (*n*, %) *N* = 161 (46.3%) | BV negative (*n*, %) *N* = 187 (53.7%) | *p* value |
|---|---|---|---|
| Age (years) | 35.59 ± 7.92 | 37.61 ± 9.16 | 0.03 |
| Gravida | 1.99 ± 1.44 | 2.01 ± 1.36 | 0.94 |
| Parity | 1.7 ± 1.11 | 1.83 ± 1.14 | 0.28 |
| Past abortion/s | 0.3 ± 0.75 | 0.16 ± 0.51 | 0.04 |
| Body mass index (kg/m$^2$) BMI | 26.99 ± 3.12 | 26.44 ± 4.02 | 0.94 |

Data expressed as Mean ± SD and absolute count with percentage in parenthesis. The Student's *t*-test was used to compare the differences between groups.

**Table 2** Relationships between risk factors and occurrence of Bacterial Vaginosis.

| Characteristic | Total (*n*, %) *N* = 348 | BV positive (*n*, %) *N* = 161 (46.3)[a] | BV negative (*n*, %) *N* = 187 (53.7)[a] | *p* value[b] |
|---|---|---|---|---|
| *Age category (years)* | | | | 0.199 |
| 15–25 | 29 | 14 (48.3) | 15 (51.7) | |
| 26–35 | 141 | 69 (48.9) | 72 (51.1) | |
| 36–45 | 126 | 70 (55.6) | 56 (44.4) | |
| ≥46 | 52 | 34 (65.4) | 18 (34.6) | |
| *Education status* | | | | 0.016 |
| Illiterate | 5 | 2 (40) | 3 (60) | |
| Primary | 45 | 26 (57.8) | 19 (42.2) | |
| Secondary | 113 | 60 (53.1) | 53 (46.9) | |
| High school | 132 | 58 (43.9) | 74 (56.1) | |
| University | 53 | 15 (28.3)[a] | 38 (71.1)[b] | |
| *Occupation* | | | | 0.137 |
| Housewife | 271 | 131 (48.3) | 140 (51.7) | |
| Teacher | 15 | 5 (33.3) | 10 (66.7) | |
| Manual/clerical employee | 32 | 14 (43.8) | 18 (56.3) | |
| Healthcare worker | 29 | 10 (43.5) | 19 (56.5) | |
| *Premature birth history* | | | | 0.983 |
| No | 332 | 154 (46.6) | 178 (53.4) | |
| Yes | 15 | 7 (46.7) | 8 (53.3) | |
| *Vaginal douche* | | | | 0.001 |
| No | 120 | 27 (22.5) | 93 (77.5) | |
| Yes | 228 | 134 (58.8) | 94 (41.2) | |
| *Method of contraception* | | | | 0.043 |
| None | 187 | 95 (50.8) | 92 (49.2) | |
| Withdrawal | 40 | 17 (42.3)[a] | 23 (57.5)[b] | |
| Condom | 30 | 10 (33.3)[a] | 20 (66.7)[b] | |
| IUD | 56 | 27 (48.8) | 29 (51.2) | |
| Tube ligation | 25 | 10 (40)[a] | 15 (60)[b] | |
| OCP | 10 | 5 (50) | 5 (50) | |
| *Smoking* | | | | 0.037 |
| No | 294 | 129 (43.9) | 165 (56.1) | |
| Yes | 54 | 32 (59.3) | 22 (40.7) | |
| *Infertility* | | | | 0.004 |
| No | 275 | 116 (42.2) | 159 (57.8) | |
| Yes | 73 | 45 (61.6) | 28 (38.4) | |

BV: bacterial vaginosis, IUD: intrauterine device, OCP: oral contraceptive pill.
[a,b]Statistically significant difference when compared with BV positive and BV negative group.
[a] All values are expressed as *N*(%).
[b] Fisher's Exact Test and the Chi-square test were used.

**Table 3** Relationships between risk factors and the presence of Ureaplasma urealyticum (UU) and Mycoplasma hominis (MH).

| Characteristic | Total *N* (%) *N* = 348 | No reproduction (*n*, %) *N* = 196 (56.3)[a] | UU(+) (*n*, %) *N* = 93 (26.7)[a] | MH(+) (*n*, %) *N* = 13 (3.7)[a] | UU and MH(+) (*n*, %) *N* = 46 (13.2)[a] | *p* value[b] |
|---|---|---|---|---|---|---|
| *Age (years)* | | | | | | 0.022 |
| 15–25 | 29 | 14 (48.3) | 9 (31) | 0 | 6 (20.7) | |
| 26–35 | 141 | 77 (54.6) | 39 (27.7) | 5 (3.5) | 20 (14.2) | |
| 36–45 | 126 | 65 (51.6) | 36 (28.6) | 7 (5.6) | 18 (14.3) | |
| ≥46 | 52 | 40 (76.9)[a] | 9 (17.3)[b] | 1 (1.9)[ab] | 2 (3.8)[b] | |
| *Education status* | | | | | | 0.642 |
| Illiterate | 5 | 3 (80) | 0 | 2 (0) | 0 | |
| Primary | 45 | 25 (55.5) | 12 (26.6) | 2 (4.4) | 6 (13.3) | |
| Secondary | 113 | 68 (54.0) | 32 (23.31) | 2 (1.8) | 12 (10.6) | |
| High school | 132 | 74 (56.06) | 29 (21.9) | 7 (5.3) | 22 (16.6) | |
| University | 53 | 25 (52.8) | 22 (26.4) | 1 (3.8) | 6 (17) | |
| *Occupation* | | | | | | 0.137 |
| Housewife | 271 | 157 (57.9) | 75 (27.7) | 7 (2.6) | 32 (11.8) | |
| Teacher | 15 | 8 (53.3) | 4 (66.7) | 2 (13.3) | 1 (6.7) | |
| Manual/clerical employee | 32 | 14 (43.8) | 18 (56.3) | 3 (9.4) | 7 (21.9) | |
| Healthworker | 29 | 19 (56.5) | 6 (43.5) | 1 (3.44) | 5 (17.24) | |
| *Premature birth history* | | | | | | 0.003 |
| No | 332 | 193 (58.1) | 87 (26.2) | 13 (3.9) | 39 (11.7) | |
| Yes | 15 | 3 (20) | 6 (40) | 0 (0) | 6 (40) | |
| *Vaginal douche* | | | | | | 0.001 |
| No | 120 | 90 (75) | 21 (17.5) | 2 (1.7) | 7 (5.8) | |
| Yes | 228 | 106 (46.5) | 72 (31.6) | 11 (4.8) | 39 (17.1) | |
| *Methods of contraception* | | | | | | 0.06 |
| None | 187 | 114 (61) | 46 (24.6) | 6 (3.2) | 21 (11.2) | |
| Withdrawal method | 40 | 21 (52.5) | 7 (17.5) | 4 (10) | 8 (20) | |
| Condom | 30 | 19 (63.3) | 9 (30) | 0 | 2 (6.7) | |
| IUD | 56 | 20 (37) | 23 (42.6) | 1 (1.9) | 10 (18.5) | |
| Tube ligation | 25 | 7 (63.6) | 3 (27.3) | 0 | 1 (9.1) | |
| OCP | 10 | 3 (30) | 3 (30) | 1 (10) | 3 (30) | |
| *Smoking* | | | | | | 0.037 |
| No | 294 | 168 (57.1) | 79 (26.9) | 11 (3.7) | 36 (12.2) | |
| Yes | 54 | 28 (51.9) | 14 (25.9) | 2 (3.7) | 10 (18.5) | |
| *Infertility* | | | | | | 0.002 |
| No | 275 | 167 (60.7) | 71 (25.8) | 10 (3.6) | 27 (9.8) | |
| Yes | 73 | 29 (39.7) | 22 (30.1) | 3 (4.1) | 19 (26) | |

[a,b,ab] Statistically significant difference when compared between groups.
[a] All values are expressed as *N* (%).
[b] Fisher's Exact Test and Chi-square test were used.

**Table 4** Multivariate analysis of risk factors for bacterial vaginosis.

| Variables | Adjusted odds ratio (95%CI) | *p* value |
|---|---|---|
| MH | 2.98 (0.721–12.334) | 0.031 |
| MH and UU | 6.79(2.63–17.56) | 0.000 |
| Abortion history | 2.82 (1.55–5.12) | 0.001 |
| BMI (kg/m$^2$) | 0.81 (0.74–0.89) | 0.001 |
| Infertility | 2.04 (0.99–4.22) | 0.052 |
| Vaginal douche | 6.80 (3.60–12.83) | 0.001 |

[1.55–5.12]) and higher BMI (AOR = 0.81 95% CI: [0.74–0.89] (Table 4).

## Discussion

Bacterial vaginosis is the most common cause of vaginal discharge in women of reproductive age. It is evaluated as a sexually enhanced disease, and it lays the ground for the transmission of other pathogens.[17] In our region, BV prevalence has been reported as 46.3%. The BV prevalence varies among regions and personal hygiene conditions. Rates have

been reported as 28.1–29.2% in the US,[6] 20% in Europe,[18] 31% in Africa, 51% in Nigeria[1] and 12–48% in Turkey.[3,10,19]

It was found that BV positive patients were younger than the BV negative patients. These results are similar to previous studies.[1,20–22] Some studies indicated no relation with age,[23] and another BV is common in older age.[24] These differences can be attributed to social structure, sexual preferences, and the age of starting sexual intercourse. It is thought that a higher educational level and high economic status enable more access to healthcare services. Being well-informed about vaginal hygiene rules and early diagnosis and treatment reduces the prevalence of BV. BV positivity was significantly low in patients who were university graduates. Similar results have been reported[1,20,24] and low education and socioeconomic status were increased risk of BV.[22,25]

It was found that the prevalence of BV is higher in obese women. Although the relationship has not been fully clarified, this may be attributed to the disruption of the vaginal flora in obese patients with hormonal and metabolic problems and poor dietary habits, blood in the vagina due to menstrual irregularities, and vaginal douching practices.[6] Also, we found that BV was 6.08-fold higher in women who regularly vaginal douched. Similarly, other studies reported that vaginal douching increases the risk of BV positivity by 1.21–2.06 fold.[22] Also, Klebanoff indicated that when vaginal douching was stopped, BV recurrence was reduced.[26] Interestingly, Kasaro reported that vaginal douching is not a risk factor if done with water but constitutes a risk factor when chemical agents such as soap are used.[15] In contrast, Jespers found no relationship.[27]

We found a low BV prevalence for those women who used the withdrawal contraception method, condoms, or tubal ligation. It has been reported that condom use protects against BV and STDs.[7,22] As tubal ligation is a method usually preferred at an older age, this could explain the low rate of BV observed in the older age bracker. Since semen pH is higher than the vaginal acidity, it may increase the prevalence of BV by disrupting the acidity of the vaginal environment. Therefore, we believe that the prevalence of BV is less common in the withdrawal method.One study has reported that the use of condoms reduces BV recurrence.[24] We did not find a relationship between BV and IUD. While Mardh reported no relationship,[7] Tibaldi found higher BV prevalence in patients using an IUD.[24] By changing the cervicovaginal flora, an IUD can prepare the ground for BV. In this study, UU was 26.7%, UU and MH combined 13.2%, and MH at 3.7%. These results are consistent with earlier reports in the literature. For instance, Robinson reported MH at the rate of 20–50% and UU at 40–80%.[28] UU was reported at 28.4% and MH at 0.8% in Portugal[8] and UU 12.7%, MH 3.4% and UU and MH 0.4% in Turkey.[29] These variations in what has been reported may be due to cultural differences and whether or not the regional culture and religious beliefs are monogamous. The UU and MH positivity were significantly low in the $\geq$46 years group. This can be explained by several reasons, such as increased awareness of protective methods together with age and an increase in monogamy. In the literature, these pathogens are common at a younger age.[24] Silva reported that UU and MH occur most in the 25–29 years, and the second peak was found in the 35–44 years age group.[8]

We found a significant relationship between BV positivity and MH and UU. Similarly, Goje reported that the combination of BV and Mycoplasma occurs at a high rate of 58.3%.[30] For us, however, we found a relationship between vaginal douching and MH and UU positivity, which may have been due to the development of BV consequent to vaginal flora disruption.

We have found a high BV, UU, and MH prevalence in infertile women. The increasing severity of infertility is a significant public health problem. A prevalence of approximately 20%-35% affects a vast population. Although ovulation disorders, endocrine disorders, and tubal-peritoneal diseases are important causes of infertility, there has been a recent focus on microorganisms infecting the reproductive system. The MH and UU, which are within the Mollicutes family, may cause urinary infection, chorioamnionitis, pelvic inflammatory disease, and associated infertility.[5] The relationship between infertility and BV has not been clarified yet, and many factors are responsible. As a result of adhesions in the uterus and tubes with chronic infection formed in the lower genital system, the development of the zygote is more challenging, leading to ectopic pregnancies and premature membrane rupture. It has been reported that BV spreads to the upper genital tract due to vaginal flora infection and is closely related to adverse reproductive outcomes. BV microbiota and related inflammation and cytokine increase are responsible for decreased fertility. Another factor is that bacteria break down cervical mucus with sialidase and mucinases, facilitating the transmission of infection in the upper genital tract. In addition, BV contributes to infertility by facilitating the transmission of STDs. The prevalence of tubal factor infertility and BV in IVF patients suggests that there may be similar pathogenesis. While many studies found these pathogens are associated with tubal infertility, Salah found high levels in unexplained infertility and PCOS group.[9,11] The differences between these studies can be attributed to differences in populations and the number of patients who were anovulatory and had tubal infertility. Lastly, regulation of vaginal microbiota with the use of probiotic vaginal tablets can reduce the frequency of BV by suppressing inflammation.[31,32] In patients whose infertility has underlying infectious causes, complications can be overcome with early treatment, and fertility can be recovered within a short period.

According to the regression analysis, MH, MH, and UU, abortion history, high BMI, and vaginal douching habits were risk factors for BV. To the best of our knowledge, this is the first study in Turkey to have shown the high prevalence of BV with MH and UU co-infection, especially in infertile patients. In the light of these results and considering the severe complications that BV, MH, and UU may cause, it is necessary to screen suspected cases for all of them. Vaginal infections should not be ignored in reproductive-age women with fertility expectations. The pregnancy can occur through appropriate antibiotic treatment. Paying attention to this fact may help avoid unnecessary medical expenses in the infertility investigation.

There are some limitations to this study. First, the population in this region is predominantly Muslim. As most of the patients were from rural communities, almost all did not wish to answer questions related to

a single sexual partner, the number of previous sexual partners, female–female sexual intercourse, and sexual behaviors. Therefore, these parameters did not include in the study.

## Conclusion

We observed that BV, MH, and UU co-infection was common in patients with vaginal discharge, significantly higher in infertile patients. Preventive measures should be taken to eliminate these risk factors that lead to the increase of mentioned pathogens, and individual awareness should be raised with training programs. As a result, these steps can help overcome the challenges of infertility and maternal complications.

## Ethical disclosures

**Protection of human and animal subjects.** The authors declare that the procedures followed were in accordance with the regulations of the relevant clinical research ethics committee and with those of the Code of Ethics of the World Medical Association (Declaration of Helsinki).

**Confidentiality of data.** The authors declare that they have followed the protocols of their work center on the publication of patient data.

**Right to privacy and informed consent.** The authors have obtained the written informed consent of the patients or subjects mentioned in the article. The corresponding author is in possession of this document.

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None.

## Conflict of interest

The authors declare that they have no conflict of interest.

## References

1. Wariso KT, Igunma JA, Oboro IL, Olonipili FA, Robinson N. Prevalence of bacterial vaginosis among patients with vulvovaginitis in a tertiary hospital in Port Harcourt, Rivers state, Nigeria. Asian J Med Health. 2017;7:1–7.
2. Allsworth JE, Peipert JF. Prevalence of bacterial vaginosis: 2001–2004 national health and nutrition examination survey data. Obst Gynecol. 2007;109:114–20.
3. Karaer A, Boylu M, Avsar AF. Vaginitis in Turkish women: symptoms, epidemiologic–microbiologic association. Eur J Obst Gynecol Reprod Biol. 2005;121:211–5.
4. Ugwumadu AH. Bacterial vaginosis in pregnancy. Curr Opin Obstet Gynecol. 2002;14:115–8.
5. Lis R, Rowhani-Rahbar A, Manhart LE. Mycoplasma genitalium infection and female reproductive tract disease: a meta-analysis. Clin Infect Dis. 2015;61:418–26.
6. Brookheart RT, Lewis WG, Peipert JF, Lewis AL, Allsworth JE. Association between obesity and bacterial vaginosis as assessed by Nugent score. Am J Obst Gynecol. 2019;220:476–81.
7. Mårdh PA. Bacterial vaginosis: a threat to reproductive health? Historical perspectives, current knowledge, controversies and research demands. Eur J Contracept Reprod Health Care. 2000;5:208–19.
8. Silva J, Cerqueira F, Teixeira AL, Bicho MC, Campainha R, Amorim J, et al. Genital mycoplasmas and ureaplasmas in cervicovaginal self-collected samples of reproductive-age women: prevalence and risk factors. Int J STD AIDS. 2018;29:999–1006.
9. Durugbo II, Nyengidiki TK, Bassey G, Wariso KT. Bacterial vaginosis among women with tubal factor infertility in Nigeria. Int J Gynecol Obst. 2015;131:133–6.
10. Rahman S. Some frequent genital tract infections and related factors in women who admitted to elazig Sara Hatun Obstetric and Gynecology Hospital with the complaint of vaginal discharge; 2008.
11. Salah RM, Allam AM, Magdy AM, Mohamed AS. Bacterial vaginosis and infertility: cause or association? Eur J Obst Gynecol Reprod Biol. 2013;167:59–63.
12. Cao CJ, Wang YF, Fang DM, Hu Y. Relation between mycoplasma infection and recurrent spontaneous abortion. Eur Rev Med Pharmacol Sci. 2018;22:2207–11.
13. Lokken EM, Balkus JE, Kiarie J, Hughes JP, Jaoko W, Totten PA, et al. Association of recent bacterial vaginosis with acquisition of *Mycoplasma genitalium*. Am J Epidemiol. 2017;186:194–201.
14. Ryckman KK, Simhan HN, Krohn MA, Williams SM. Predicting risk of bacterial vaginosis: the role of race, smoking and corticotropin-releasing hormone-related genes. Mol Hum Reprod. 2009;15:131–7.
15. Kasaro MP, Husnik MJ, Chi BH, Reid C, Magure T, Makanani B, et al. Impact of targeted counseling on reported vaginal hygiene practices and bacterial vaginosis: the HIV Prevention Trials Network 035 study. Int J STD AIDS. 2017;28:467–75.
16. Nugent RP, Krohn MA, Hillier SL. Reliability of diagnosing bacterial vaginosis is improved by a standardized method of gram stain interpretation. J Clin Microbiol. 1991;29:297–301.
17. Hay P, Ugwumadu A. Detecting and treating common sexually transmitted diseases. Best Pract Res Clin Obst Gynaecol. 2009;23:647–60.
18. Kenyon C, Colebunders R, Crucitti T. The global epidemiology of bacterial vaginosis: a systematic review. Am J Obst Gynecol. 2013;209:505–23.
19. Ortaylı N, Amca M, Say L, Özgür S, Nalbant H. Frequency of bacterial vaginosis and related factors. Türkiye Klinikleri J Gynecol Obst. 2000;10:57–62.
20. Diadhiou M, Ba Diallo A, Barry MS, Alavo SC, Mall I, Gassama O, et al. Prevalence and risk factors of lower reproductive tract infections in symptomatic women in Dakar, Senegal. Infect Dis: Res Treat. 2019;12, 1178633719851825.
21. Modak T, Arora P, Agnes C, Ray R, Goswami S, Ghosh P, et al. Diagnosis of bacterial vaginosis in cases of abnormal vaginal discharge: comparison of clinical and microbiological criteria. J Infect Dev Ctries. 2011;5:353–60.
22. Singh A, Kanti V, Dayal S, Shukla SK, Mishra N. Prevalence and risk factors of bacterial vaginosis among women of reproductive age attending rural tertiary care institute of Western Uttar Pradesh. J Evol Med Dent Sci. 2016;5:2695–702.
23. Holzman C, Leventhal JM, Qiu H, Jones NM, Wang J, BV Study Group. Factors linked to bacterial vaginosis in nonpregnant women. Am J Public Health. 2001;91:1664–70.
24. Tibaldi C, Cappello N, Latino MA, Masuelli G, Marini S, Benedetto C. Vaginal and endocervical microorganisms in symptomatic and asymptomatic non-pregnant females: risk factors and rates of occurrence. Clin Microbiol Infect. 2009;15:670–9.
25. Ashraf Ganjoei T. Risk factors for bacterial vaginosis in women attending a hospital in Kerman, Islamic Republic of Iran. EMHJ. 2005;11:410–5.

26. Klebanoff MA, Nansel TR, Brotman RM, Zhang J, Yu KF, Schwebke JR, et al. Personal hygienic behaviors and bacterial vaginosis. Sex Transm Dis. 2010;37:94.
27. Jespers V, Crucitti T, Menten J, Verhelst R, Mwaura M, Mandaliya K, et al. Prevalence and correlates of bacterial vaginosis in different sub-populations of women in sub-Saharan Africa: a cross-sectional study. PLOS ONE. 2014;9:e109670.
28. Taylor-Robinson D, Jensen JS. Genital mycoplasmas. Atlas Sex Transm Dis AIDS. 2010:64–75.
29. Ozturk S, Yildiz S, Dursun P, Ilce BY, Kaymaz O. Mycoplasma hominis profile in women: culture, kit, molecular diagnosis, antimicrobial resistance, and treatment. Microb Pathogen. 2019;135:103635.
30. Goje O, Munoz JL, Nolte FS, Soper DE. Prevalence and clinical significance of *Mycoplasma genitalium* in gynecologic patients. J AIDS Clin Res. 2017;8:2.
31. Ravel J, Moreno I, Simón C. Bacterial vaginosis and its association with infertility, endometritis, and pelvic inflammatory disease. Am J Obst Gynecol. 2021;224:251–7.
32. Ding C, Yu Y, Zhou Q. Bacterial vaginosis: effects on reproduction and its therapeutics. J Gynecol Obst Hum Reprod. 2021;50:102174.