

Prevalence of Bacterial Vaginosis and Impact of Genital Hygiene Practices in Non-Pregnant Women in Zanjan, Iran

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Abstract

Objectives: Bacterial vaginosis is one of the most common causes of reproductive tract infection (RTI), its prevalence is influenced by many factors. The aim of this study is to determine the prevalence of bacterial vaginosis and impact of sexual and genital hygiene practices and socio-demographic characteristics in non pregnant women of Zanjan province in Iran.

Methods: 500 non-pregnant, married women were randomly selected for this study. Following gynecological examination and vaginal sample collection by physicians, bacterial vaginosis was confirmed by Nugent criteria, trichomoniasis by direct microscopy and candidiasis by direct microscopic observation and evaluation of presenting clinical signs of vulvovaginitis.

Results: The prevalence of RTI was 27.6%. Out of which 16.2% was devoted to bacterial vaginosis (BV), 6.6% to trichomoniasis and 4.8% to Vulvovaginal candidiasis (VVC). In contrast to coital hygiene, there was a significant correlation between menstrual and individual vaginal hygiene and BV ($P<0.01$ and $P<0.001$) respectively. There was a significant correlation between BV and education ($P<0.025$), number of pregnancy ($P<0.05$) and

method of contraception ($P<0.005$). No significant correlation was observed between age, age of marriage and abortion.

Conclusion: The data obtained suggests that the prevalence rate of BV is relatively high and could be affected by hygiene behaviors and certain socio-demographic characteristics, which indicate the need for comprehensive, scheduled programs of healthcare educations, aimed at reducing BV prevalence.

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Introduction

Data from epidemiological surveys show that within countries and between countries in the same region, the prevalence and incidence of genital tract infections may vary widely even in similar population groups.¹ In developing countries, patients with genital tract infections often bypass formal healthcare services and seek care in the informal sector.² Diverse studies performed, with the objective of establishing the frequency of the most common infectious agents for vaginitis, have shown varying results. The prevalence found for BV has varied, ranging between 8% and 75%; VVC has presented 2.2 - 30% and trichomoniasis between 0% and 34 %.³ BV is the most common vaginal infection of reproductive age women and is the most frequently cited cause of vaginal discharge and malodor.⁴ It is characterized by an increased vaginal pH and the replacement of vaginal lactobacilli (particularly those that produce hydrogen peroxide) with *Gardnerella vaginalis* and anaerobic Gram-negative rods.⁵ Women with BV may be at increased risks of sexually transmitted disease (STD), HIV, and a number of other adverse reproductive outcomes.⁶ VVC is an extremely common infection in women of childbearing age of all strata of society. Since it has now been excluded from the ranks of

sexually transmitted diseases and is also not a notifiable disease, not much information regarding its incidence and epidemiology is available.⁷ VVC is the second most common cause of vaginitis in the United States and the most common cause in Europe.⁸ Trichomoniasis is known to be responsible for an estimated 180 million new infections per year, making it the most prevalent non-viral sexually transmitted pathogen worldwide.⁹ Infection of the female genital tract can result in vaginitis, cervicitis, and urethritis, and trichomoniasis has been associated with adverse pregnancy outcomes.¹⁰ Behavioral factors such as vaginal douching or menstrual hygiene practices have been suggested as important factors that might influence vaginal flora composition, but little data is available from Iranian populations.¹¹ Therefore, the present study was carried out to determine the prevalence of BV, VVC and Trichomoniasis and association of BV with socio-demographic characteristics and sexual and hygiene-related variables in women attending primary health care centers in Zanjan, Iran.

Methods

Prior to the initiation of the experiment, study protocols were reviewed and approved by the ethical committee from the research

office of Zanzan University of Medical Science. Declaration of Helsinki was considered in this study. Women were enrolled after giving informed consent according to local ethics committee guidelines, and clinical research was conducted in accordance with guidelines for human experimentation issued by the authors' institutions.

To calculate the sample size based on a projected RTI prevalence of 30% (precision 5%, confidence interval 95%) and adjusting for the design effect, approximately 500 married women were enrolled. Out of 10 primary healthcare centers in Zanzan, 5 were randomly selected and in each selected center, 100 non pregnant married women, between the ages of 15 - 45 (mean age 36 years), were randomly recruited. Primary healthcare centers are evenly situated in different parts of the Zanzan city and are managed under direct supervision of the Zanzan University of Medical Science and most of the people prefer to consult at these centers for charge free health and medical services.

A standardized questionnaire elicited sexual and hygiene behaviors and socio-demographic characteristics. The purpose of the study was explained to all eligible women and with their permission; interviews were conducted in complete privacy. Under supervision of in - charge midwife of present study, and an attendant physician at each primary healthcare center, conducted the required gynaecological examinations and sampling. Swab samples were collected for Gram stain, wet mount preparation and determination of pH, by a pH indicator paper (pH indicator strips; Merck Laboratories, Darmstadt, Germany).

For diagnosis of bacterial vaginosis, vaginal smear slides were heat fixed, Gram-stained and examined under oil immersion objective (1000x magnification) and graded as per standardized, quantitative, morphological classification method developed by Nugent *et al.*¹² The method involved assigning a score between 0 and 10 based on the quantitative assessment of the Gram-stain for three different bacterial morphotypes: (i) large Gram-positive rods (indicative of *Lactobacillus* spp), (ii) small Gram-negative or variable rods (indicative of *Gardnerella*, *Bacteroides* and other anaerobic bacteria), and (iii) curved, Gram-variable rods (indicative of *Mobiluncus* spp). Scores between 0 and 3 represented 'normal vaginal flora', between 4 and 6 'intermediate vaginal flora', and scores between 7 and 10 were considered diagnostic for 'BV'. In this study, microbiological definition of BV was a score of 7–10 by Nugent's method. Quality control of the readings was checked by rereading 10% of the slides by a second experienced microbiologist for Nugent's score. The diagnosis of VVC was suggested in women who presented symptoms and/or signs of vaginitis, eg.

vaginal discharge (sometimes and often minor; can be thin or thick like cottage cheese when it occurs with no particular odor), itch or discomfort, external dysuria and vulvovaginal erythema. Spores, hyphae, or yeast buds identified on wet mount confirm the diagnosis. The addition of potassium hydroxide to the wet mount slide will remove debris that may obscure the hyphae, and assists in making an accurate diagnosis.¹³ In present study, VVC was diagnosed on the basis of clinical symptoms of vaginitis and direct microscopic examination. Trichomoniasis was diagnosed by visualisation of motile trichomonads in the wet mount smear, immediately after sampling. All Laboratory assessments were performed by microbiologists.

With the help of SPSS (Statistical Package for social Sciences, Version 11) software package, the data was analyzed, using X² tests, to test the significance of associations between categorical variables.

Results

500 married non pregnant women, aged between 15 and 45 (mean age 36 years), were enrolled in the study. The overall prevalence of women with one RTI was 27.6%. The prevalence of BV prevalence, as determined by Nugent's score, was 16.2. Using wet preparation microscopy, 6.6% of the women were positive for *T vaginalis* and 4.8% for *Candida* spp. Principal component analysis was performed separately on the hygiene-related variables and socio-demographic characteristics, (Tables 1, 2, and 3). Hygiene-related variables were divided in to three categories of menstrual, individual vaginal and coital hygiene. Statistical analysis showed a significant correlation between BV menstrual status and individual vaginal hygiene ($P < 0.01$ and $P < 0.001$ respectively). In contrast, no significant correlation was observed between BV and coital hygiene.

Socio-demographic characteristics include age, age of marriage, educational background, method of contraception, number of pregnancy and history of abortion. Literacy was divided in to 5 groups; illiterate, primary, secondary, higher secondary and college graduate, in which 34.2% of the affected and 90% of non affected individuals were illiterate and college graduate respectively. Oral contraception, condom, intra uterine contraceptive device (IUD), sexual interruption and tubectomy were the most popular methods of contraception employed by the participants, in which 36.1% of affected and 88.1% of non affected individual reported IUD and condom use respectively. Those who had 3-5 pregnancy, showed 32.2% BV and 76.1% of non affected individual had up to 2 pregnancies.

Table 1: Frequency distribution of menstrual hygiene

| Menstrual hygiene | BV | | Yes | | | | | | No | | | | | | X ² |
|---|-------------|------|--------|------|----------|------|-------|------|--------|------|----------|------|---|---|----------------|
| | Performance | | always | | sometime | | never | | always | | sometime | | never | | |
| | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | |
| 1- Bathing during menstruation period | 100 | 78.1 | 102 | 68.9 | 160 | 71.4 | 28 | 21.9 | 46 | 31.1 | 64 | 28.6 | X ² =3.08 non significant df=2 | | |
| 2- Genital douching during menstruation period | 268 | 74.4 | 64 | 68.8 | 30 | 63.8 | 92 | 25.6 | 29 | 31.2 | 17 | 36.2 | X ² =3.04 non significant df=2 | | |
| 3- Using sanitary pad | 142 | 76.8 | 170 | 83.3 | 50 | 45 | 43 | 23.3 | 34 | 16.6 | 61 | 55 | X ² =55.2 Significant df=2 p<0.001 | | |
| 3- Pad change after each urination | 76 | 83.5 | 221 | 71.1 | 65 | 66.3 | 15 | 16.5 | 90 | 28.9 | 33 | 33.7 | X ² =7.68 Significant df=2 p<0.025 | | |
| 4- Sexual abstinence during menstruation period | 349 | 73.6 | 10 | 43.5 | 3 | 100 | 125 | 26.4 | 13 | 56.5 | 0 | 0 | X ² =6.87 Significant df=1 p<0.01 | | |

There was a significant correlation between BV and education ($P<0.025$), method of contraception ($P<0.005$) and number of pregnancy ($P<0.05$). On the other hand, no significant correlation was found between BV and age, age of marriage, and history of abortion.

Discussion

The main objective of this study was to determine the prevalence of BV among women attending primary healthcare centers in the Zanzan province, Iran and to relate this to sexual and vaginal hygiene behaviors and sociodemographic characteristics. Despite peoples' concerns, accurate epidemiologic data on RTI are scarce and existing information yields a widely varying RTI prevalence that ranges from 20% to 70%.¹⁴ In comparison with other similar studies, using the same diagnostic method, the results from this study were considered to be slightly higher but also, in some cases, lower prevalence. In a study conducted in the rural area of Shandong province in China, the prevalence of BV, trichomoniasis and candidiasis were 6.6, 2.9 and 3.9% respectively.¹⁵ In another study performed in Hamedan province, Iran, the prevalence of candidiasis, trichomoniasis, and BV was 17.2, 18.1, and 28.5%, respectively.¹⁶ Among women referred to hospital in Vientiane, the capital of Laos, the prevalence of BV, trichomoniasis and candidiasis were 24.5, 3.7 and 39.5% respectively.¹⁷ In another

study conducted in the rural area of Northeast Brazil, 20% of women had BV, 4.1% trichomoniasis and 12.5% candidiasis.¹⁸ With attention to the above findings, it can be concluded that RTI has a varying degree of prevalence rate among people of different communities which might be due to certain factors such as hygiene behaviors and sociodemographic characteristics. Therefore, it is important to try to establish a correlation between BV and factors affecting its prevalence. In this study, menstrual, personal and coital hygiene were the hygiene-related variables. Consistent with other studies, there was a significant correlation between individual and menstrual hygiene and prevalence of BV, whereas other findings showed no significant correlation in this regard.^{11,19,20,21,22} In the current study, there was a significant correlation between methods of contraception and BV. Data showed that among different methods of contraception employed in the current study, IUD was most commonly used but the use of condoms was relatively low. However, as compare to condom, BV was diagnosed significantly more frequent in women with IUD, which is in line with other studies.^{21,23} The results also showed a significant correlation between BV and educational status, which in comparison to other similar studies, it was evident that the lack of education has been found to be significantly associated with BV among women in Zanzan, whereas certain studies contradict this finding.^{15,24,23} Similar to other studies, there was no association observed between

Table 2: Frequency distribution of individual ginal hygiene

| Individual vaginal hygiene | Performance | | Yes | | | No | | | X ² | | | | |
|--|-------------|------|-----|------|-----|------|-----|------|----------------|--------|----------|-------|---|
| | No. | % | No. | % | No. | % | No. | % | | | | | |
| | | | | | | | | | | always | sometime | never | always |
| 1- Regular underwear changing | 180 | 74.7 | 70 | 72.2 | 112 | 69.1 | 61 | 25.3 | 27 | 27.8 | 50 | 30.9 | X ² = 1.47 non significant df=2 |
| 2- Underwear washing with hot water | 176 | 76.2 | 87 | 77 | 99 | 63.5 | 55 | 23.8 | 26 | 23 | 57 | 36.5 | X ² = 9.12 significant df=2 p<0.025 |
| 3- sunlight drying of underwear | 300 | 72.5 | 33 | 79.6 | 19 | 59.4 | 114 | 27.5 | 11 | 20.4 | 13 | 40.6 | X ² =4.18 non significant df=2 |
| 4- use of loss underwear | 291 | 72.2 | 65 | 82.3 | 6 | 33.3 | 112 | 27.8 | 14 | 17.7 | 12 | 66.7 | X ² =17.65 Significant df=2 p<0.001 |
| 5- Ironing underwear before use | 25 | 69.4 | 37 | 74 | 300 | 72.5 | 11 | 30.6 | 13 | 26 | 114 | 37.5 | X ² =0.2 non significant df=2 |
| 6- driness of underwear during day | 204 | 77.9 | 119 | 68 | 39 | 61.9 | 58 | 22.1 | 56 | 32 | 24 | 38.1 | X ² =9.03 Significant df=2 p<0.025 |
| 7- front to back douching and cleaning of genital area | 270 | 77.1 | 40 | 59.7 | 52 | 62.7 | 80 | 22.9 | 27 | 40.3 | 31 | 37.3 | X ² =13.27 Significant df=2 p<0.005 |
| 8- drying of genital area after douching | 97 | 76.4 | 39 | 79.6 | 226 | 69.8 | 30 | 23.6 | 10 | 20.4 | 98 | 30.2 | X ² = 3.38 non significant df=2 |
| 9- use of privet bathroom | 230 | 74.9 | 22 | 73.3 | 110 | 67.5 | 77 | 25.7 | 8 | 26.7 | 53 | 32.5 | X ² =2.86 non significant df=2 |
| 10- use of personal bathing equipment | 202 | 78 | 32 | 72.7 | 128 | 65 | 57 | 22 | 12 | 27.3 | 69 | 35 | X ² =9.44 Significant df=2 p<0.01 |
| 11 - sanitation of bathroom before use | 272 | 79.5 | 68 | 58.6 | 22 | 52.4 | 70 | 20.5 | 48 | 41.4 | 20 | 47.6 | X ² =28.14 Significant df=2 p<0.001 |
| 12 - bathing in standing position | 98 | 81.7 | 60 | 69.8 | 204 | 69.4 | 22 | 18.3 | 26 | 30.2 | 90 | 30.6 | X ² =6.8 Significant df=2 p<0.05 |
| 13 -sun drying of bathing equipment | 253 | 77.6 | 49 | 60.5 | 60 | 64.5 | 73 | 22.4 | 32 | 39.5 | 33 | 35.5 | X ² =13.01 Significant df=2 p<0.005 |
| 14 - shaving of hair in genital area | 325 | 73.2 | 10 | 52.6 | - | - | 129 | 26.8 | 9 | 47.4 | - | - | X ² = 3.85 non significant df=1 |

Table 3: Frequency distribution of coital hygiene

| Coital hygiene | BV | | | | | | Yes | | | | | | No | | | | | | X ² |
|---|-------------|------|--------|------|----------|------|-------|------|--------|------|----------|------|--|--|--|--|--|--|----------------|
| | Performance | | always | | sometime | | never | | always | | sometime | | never | | | | | | |
| | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | | | | | | | |
| 1- Genital douching before intercourse | 215 | 75.2 | 86 | 65.6 | 61 | 73.5 | 71 | 24.8 | 45 | 34.4 | 22 | 26.5 | X ² =4.1 non significant df=2 | | | | | | |
| 2-Urination after intercourse | 218 | 75.2 | 90 | 65.7 | 54 | 74 | 72 | 24.8 | 47 | 34.3 | 19 | 26 | X ² =4.26 non significant df=2 | | | | | | |
| 3-Frequency of intercourse (less than 7 times a week) | 164 | 77.4 | 148 | 71.8 | 50 | 61 | 48 | 22.6 | 58 | 28.2 | 32 | 39 | X ² =7.99 Significant df=2 p<0.025 | | | | | | |

the prevalence of BV and age, as almost equal prevalence was seen in women between 15 to 45 years.⁵ However, other studies showed a significant correlation between BV and different age groups.^{18,23,24} The causes for the age distribution patterns of BV are difficult to disentangle, as probably various behavioral, physiological, and immunological variables interact. In contrast to other findings, no significant correlation was observed between BV and number of abortion in this study.¹⁵ Except in medically indicated and pathogenic cases, the induction of abortion/curatage, particularly in unexpected pregnancies, is an illegal act in Iran, and it is mostly performed by local midwives or non authorized persons, and as a result not only the data is not officially recorded but also it may not be reported by the participant during the interview. Therefore, no significant correlation can be deduced, partly, due to incorrect information in this regard. No significant correlation was found between BV and the age at marriage. Intact hymen is a traditional sign of virginity in Iran, particularly in the Muslim community, and it should be maintained, some how until marriage. That is why the age at marriage is considered as the first intercourse in present study population. There are certain limitations in this study. Firstly, since the risk factors were self reported, it is possible that there was under reporting and misclassification of risk behaviors. Secondly, the survey involved the collection of temporally distant and sensitive sexual and hygiene behaviors, therefore, there is a possibility of measurement error that may lead to residual confounding associations.

Conclusion

In conclusion, this study could provide important epidemiologic data on BV for future risk behaviors and population based

studies. The data suggests that the prevalence rate of BV was relatively high and it was affected by hygiene behaviors and certain sociodemographic characteristics, which indicate the need for comprehensive, scheduled programs of healthcare educations, aimed at reducing BV prevalence as well as to guide the planning and resource allocation of decision makers for future interventions. Furthermore, since there is strong evidence in the literature that BV is associated with STIs including HIV, further studies are needed to understand the potential role of screening and treatment of BV in STI/HIV prevention programs.

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