Prevalence of Bacterial Vaginosis among Sexually Active Females in a Tertiary Institution in Benin Metropolis, Nigeria

*Moses-Otutu IM, Ngemegwai CC

ABSTRACT

Bacterial vaginosis is the most prevalent cause of abnormal vaginal discharge in women between the ages of 15 and 44 years. Bacterial vaginosis, if untreated can progress to pelvic inflammatory disease, preterm labour and ultimately infertility. The aim of this study was to determine the risk factors and their association with the prevalence of bacterial vaginosis among sexually active females in a tertiary institution in Benin metropolis. This study recruited 113 randomly selected sexually active females in a tertiary institution in Benin metropolis. The age range was 16 years to 37 years. After informed consent, sterile swab sticks were used to collect high vaginal swab and immediately sent for laboratory analysis. Nugent scoring method was applied for bacterial vaginosis. Results obtained were statistically analyzed. A total prevalence of 47.8% bacterial vaginosis was obtained among sexually active females in a tertiary institution in Benin metropolis. The highest prevalence of bacterial vaginosis was among age 21-25years (35.2%), 16-20years (29.6%) while 31-35 years had the least prevalence (3.7%). Bacterial vaginosis varied significantly with tribes, Bini recorded the highest prevalence (38.9%), Esan (22.2%), others (14.8%), Etsako (13.0%) and Yoruba's recording the least prevalence (3.7%). Marital status and frequency of sex did not significantly influence the prevalence of bacterial vaginosis (P=1.000; P=0.065) among sexually active females in a tertiary institution in Benin metropolis. Candida albicans, a fungus was also found (45.1%) among the study population. Public enlightenment on Bacterial vaginosis, its symptoms and effects, cessation of douching and smoking and limiting the number of sex partners is advised.

Keywords: Bacterial vaginosis, Microscopy, Nugent Scoring, Asymptomatic, Candida albicans

INTRODUCTION

Bacterial vaginosis (BV) is described as a shift in the balance of the vaginal microflora characterized by an increase in the vaginal pH, a reduction in lactobacilli (hydrogen peroxide producing species) and an increase in the number and type of facultative anaerobic bacteria. 1, 2 This shift in vaginal microflora causes bacterial overgrowth resulting in the common symptoms of vaginal discharge and vaginal odour as experienced by individuals.³ The first bacterium identified to represent BV infection was G. vaginalis, and is still the known primary pathogen associated with BV diagnosis. BV has been linked with an increased risk of acquiring sexually transmitted infections (STIs) such as: human immunodeficiency virus (HIV), gonorrhea, chlamydia and herpes simplex virus. 4 Moreso,

Department of Medical Laboratory Science, School of Basic Medical Sciences, College of Medical Sciences, University of Benin, Benin City, Edo State, Nigeria.

*Corresponding author: ifueko.moses-otutu@uniben.edu Date manuscript was received: 4/8/2021 Date manuscript was accepted: 2/11/2021 BV infection can lead to pelvic inflammatory disease, preterm birth, post-hysterectomy and postpartum vaginal infections. BV is also one of the genital infections common among women of reproductive age, but its major etiology and if it is sexually transmitted are unknown.

The known risk factors that increases the risk of developing BV include: sexual activity, mostly unprotected sexual intercourse, an increased number of sexual partners, women who have sex with women, Africans or African American descent, douching and having an intrauterine device insitu.7 BV has also been associated with sexual behavior related characteristics such as young age at coitarche, life time number of sex partners, a recent history of multiple sex partners and a recent history of a new sex partner. These inconsistencies make it difficult to define what genuinely represents high-risk sexual behaviour to the acquisition of BV. Moreover, some risk factors identified may be proxy variables to the true risk of exposure; that is a new sex partner might be predictive to frequency of sexual intercourse. However, whether BV pathogenesis does actually involve sexual transmission of pathogenic micro-organisms from men to women is still unknown.9 Also identified to confer an increased risk of BV acquisition is heterosexuality among women, non-coital sexual behaviours, including receptive oral sex, receptive anal sex, and non-penetrative digito-genital contact. 10 Higher prevalence rates of G. vaginalis have been found in sexually abused girls as compared to nonabused girls in most studies.8 Aside G. vaginalis carriage, BV in children is rare. 11 According to studies, once beyond the menarche, BV also occurs among sexually inexperienced adolescents and virginal women but at lower rates on average as compared to sexually active reproductive aged-women.12 Up to one third of nonsexually active adolescents harboured G. vaginalis, which was significantly less than in sexually active adolescents (60%). Several studies support that BV incidence is increased by sexual activity but clearly also contradict exclusive heterosexual transmission. As such. preventive strategies of BV involves targeting the risk factors or behaviours of BV.

BV can occur in any age group and globally is most prevalent cause of abnormal vaginal discharge in women of childbearing age (15 years to 44 years). Although the prevalence of BV differs widely from country to country within the same region and even within similar population groups, it has been estimated to be in the range of 8% to 75%. 15 Clinical signs and symptoms of BV are: an increased grey-white vaginal discharge with consistency similar to milk and with a strong odour of fish. The vaginal discharge has a PH > 4.5 and is usually more noticeable after sexual intercourse and menstruation. The more severely affected individuals experience an offensive fishy-smelling discharge that frequently recurs around the time of menstruation.9 BV is associated with an increased risk from several pathological states including post-surgery infections which arise

after hysterectomy, post abortal pelvic inflammatory disease and plasma cell endometritis. Facultative anaerobes and anaerobic bacteria (such as G. vaginalis, Mycoplasma hominis, Ureaplasma urealiticum and Mobiluncus) are also found besides Lactobacillus in the vaginal microbiota of healthy women.

In addition to the physical risks, qualitative studies have shown that women who suffer from BV, particularly recurrent BV, experience a decrease in their quality of life (QOL). The disruptions in QOL includes: embarrassment about any perceived odour, decreased self-esteem, interruption of intimate relationships, social isolation and decrease in work productivity. This epidemiological study can help in understanding the contribution of sexual activity to the development of BV, recurrence of BV after initial treatment and is also important at reducing the burden of the disease in infected females.

MATERIALS AND METHODS Study Area and Population

This study was carried out among sexually active females in a tertiary institution in Benin metropolis, the capital of Edo State. Edo State is in the South-South geo-political zone of Nigeria and lies between latitudes 6.1°N and 7.3°N and longitude 5.0°E and 6.5°E. It has a total land area of 19,281.93Km². The State is bounded by Delta State to the South, Kogi State to the North, Ondo State to the West and the River Niger along the Eastern border. Benin City is located at 6.3°N latitude and 5.6°E longitude.

The sample population consisted of sexually active females in a tertiary institution in Benin metropolis that was randomly selected.

Study Design

A total of 113 sexually active females in a tertiary institution in Benin metropolis were randomly selected for this study. This study referred to sexually active females as females who are already exposed to sexual activities irrespective of the age, time and duration of exposure; not necessarily those that practice commercial sex. The participants gave their verbal consent after a thorough explanation of the rationale for the study and vagina swabs were then collected by trained medical personnel from each participant. After the samples were collected, they were sent to the laboratory for microbiological assessment.

Ethical approval

Permission was sought and obtained from the Ministry of Health, Edo State for ethical approval to carry out this research in Benin City metropolis. In addition, each of the participants was given a written consent form to fill to indicate their willingness to participate in the study.

Sample Collection

Two vaginal swabs were collected aseptically by trained medical personnel. A sterile speculum was used to dilate the cervix, with the speculum in situ, the tip of a rayon-tipped applicator swab stick was passed through the speculum to the posterior fornix of the vagina, then the swab was rotated for 10-15 seconds in the posterior fornix ensuring to swab any discharge present. Each swab stick was then placed inside the transport tube (one tube containing normal saline while the other tube was dry) and the tubes were closed tightly. The two vaginal swabs were transferred without delay to the microbiology laboratory for analysis.

Sample processing Vagina Wet Mount

The swab stick in normal saline was observed by wet mount microscopy to view the presence of Clue cells, pus cells and *Trichomonas vaginalis*. For yeast, germ tube test was carried out and *Candida albicans* was identified by its ability to produce germ tubes when incubated in serum at 37°C for 2 hours.

Microscopic Examination

For the diagnosis of bacterial vaginosis, the dry swab stick was used to make a smear which was heat-fixed, Gramstained and examined under oil immersion objective. Each slide was examined for yeast cells, clue cells, pus cells and normal flora. The Gram stained slide was then scored and graded as per the standardized quantitative morphological classification method developed by Nugent and colleagues.¹⁸ This assign a score between 0 and 10 based on the following various bacterial morphotypes: large Gram-positive rods (Lactobacillus morphotypes), small Gram-variable rods (G. vaginalis morphotypes), small Gramnegative rods (Bacteroides spp. morphotypes), curved Gram-variable rods (Mobiluncus spp. morphotypes), and Grampositive cocci. Each morphotype was quantitated from 1 to 4+ about the number of morphotypes per oil immersion field as represented in the table below and the score of each morphocyte added together to get the Nugent score.

Nugent Scoring Table

Score	Lactobacillus morphotype per field	Garderella morphotype per field	Curved bacteria (Mobiluncus) per field
0	>30	0	0
1	5-30	<1	1-5
2	1-4	1-4	>5
3	<1	5-30	
4	0	>30	

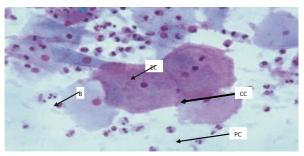


Figure 1: Bacterial vaginosis seen on a light microscope after Gram staining using oil immersion objectives (×100 objective)

KEY: B- Bacteria, EC- Epithelial Cell, CC- Clue Cell, PC- Pus Cell

Statistical Analysis

The categorical variables obtained from laboratory investigations in this study were tabulated, encoded and statistically analyzed using Statistical Package for Social Sciences (SPSS version 16) program. Test of significance was done using Chi-square and the levels of significance were accepted at p<0.05.

RESULTS

Prevalence of Bacterial Vaginosis among Sexually Active Females in a Tertiary Institution in Benin Metropolis, Nigeria

Based on Nugent scoring method, a total prevalence of 47.8% BV was obtained among sexually active females in a tertiary institution in Benin metropolis, Nigeria (Table 1). Age of participants did not significantly influence the prevalence of BV, as the prevalence of BV was higher among age groups 21-25 years (35.2%), 16-20years age group (29.6%), age group 26-30 years (24.1%), >35years (7.4%) while the least prevalence of 3.7% was recorded among 31-35years age group (Table 2).

Tribe significantly influenced the prevalence of BV among sexually active females in a tertiary institution in Benin metropolis, Nigeria (P=0.028). As prevalence of bacterial vaginosis was higher among the Binis (38.9%), Esan tribe (22.2%), other tribes (14.8%), Etsako (13.0%), Akoko-Edo (7.4%) with Yoruba tribe recording the least prevalence (3.7%) (Table 2).

Table 1: Prevalence of Bacterial Vaginosis among Sexually Active Females in a Tertiary Institution in Benin Metropolis, Nigeria.

Bacterial Vaginosis	Frequency	Percent
Positive	54	47.8
Negative	59	52.2
Total	113	100.0

Table 2: Sociodermographic Parameters and Frequency of Bacterial Vaginosis among Sexually Active Females in a Tertiary Institution in Benin Metropolis, Nigeria.

Variable	No examined (%)	No. positive (%)	X ²	P- value
Age (years)				
16-20	28(24.8)	16(29.6)	3.416	0.491
21-25	48(42.5)	19(35.2)		
26-30	24(21.2)	13(24.1)		
31- 35	06(5.3)	02(3.7)		
>35	07(6.2)	04(7.4)		
Tribe				
Yoruba	04(3.5)	02(3.7)	12.516	0.028
Bini	38(33.6)	21(38.9)		
Esan	17(15.0)	12(22.2)		
Etsako	11(9.7)	07(13.0)		
Akoko-Edo	12(10.6)	04(7.4)		
Other	31(27.4)	08(14.8)		
Total	113(100)	54(100)		

Effect of some Associated Risk Factors on the Prevalence of Bacterial Vaginosis among Sexually Active Females in a Tertiary Institution in Benin Metropolis, Nigeria

Marital status as a risk factor was not significantly associated with the prevalence of Bacterial vaginosis among sexually active females in a tertiary institution in Benin metropolis, Nigeria (OR=1.077, 95% CI = 0.338, 3.431, P=1.000). Bacterial vaginosis was however higher among single participants (89.0%) when compared to their married counterpart (11.0%) (Table 3).

Frequency of sex as a risk factor did not significantly influence the prevalence of bacterial vaginosis among sexually active females in a tertiary institution in Benin metropolis, Nigeria (P=0.065). However, females having sexual intercourse few times a month had a higher percentage prevalence of bacterial vaginosis (48.1%) followed by those having sexual intercourse few times a week (37.0%), those rarely having sexual intercourse recording 11.1% while the least prevalence was recorded among females having daily frequent sexual intercourse (3.7%) (Figure 1).

Table 3: Marital Status as a Risk Factor for the Prevalence of Bacterial Vaginosis among Sexually Active Females in a Tertiary Institution in Benin Metropolis, Nigeria.

Marital status	No examined (%)	No. positive (%)	OR	95% CI	P-value
Single	100(88.5)	48(89)	1.077	0.338 - 3.431	1.000
Married	13(15.5)	06(11)			
Total	113(100)	54(100)			

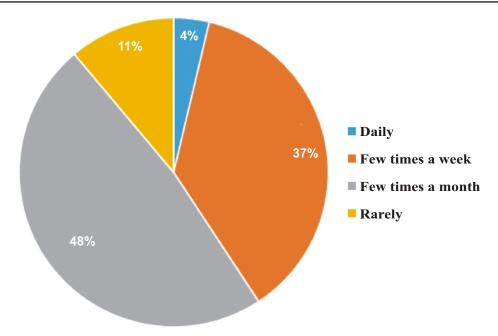


Figure 2: Prevalence of BV with Frequency of Sex among Sexually Active Females in a Tertiary Institution in Benin Metropolis, Nigeria.

DISCUSSION

The prevalence of BV has been shown from previous studies to differ widely from country to country within the same region and even within similar population groups. This study reported a total prevalence of 47.8% BV among sexually active females in a tertiary

institution in Benin metropolis, Nigeria. This value though relatively high lies within the reported prevalence range of 8% to 75% of BV from previous studies obtained globally ¹⁵ Similar studies on BV among sexually active women in USA and Cameroon recorded a prevalence of 29.2% ²¹ and 38% ^{19,20}

respectively. The differences in prevalence may be due to geographical distribution, race, ethnicity and method of analysis.

This study recorded the highest prevalence of BV among age's 21-25years (35.2%), age's 16-20years (29.6%) and age's 26-30 years (24.1%) while 31-35 years age group had the least prevalence (3.7%). Age had no statistical significant relationship with prevalence of BV among our study population (p=0.491). A study in Cameroon also recorded the highest prevalence of BV among age group 20-25 years (48.1%) with a statistically significant relationship between age and prevalence of BV among sexually active women. 20 Also observed from another study is an association among 17-21-year-old females with oral sex and non-penetrative digitogenital contact and the occurrence of BV.²⁷The reasons behind our results may be because more of the risk factors for acquisition of BV such as young age at coitarche, douching, non coital sex behavior, receptive anal and oral sex and non-penetrative digito genital contact 10, 22 occurs mostly at ages 16-25 years due to teenage and youthful exuberance. Moreso, once beyond the menarche, BV also occurs among sexually inexperienced adolescents and virginal women but at lower rates on average as compared to sexually active reproductive aged-women.¹²

BV prevalence varied significantly with the tribes: Bini's had the highest prevalence (38.9%), Esan (22.2%), other tribes (14.8%), Etsako (13.0%), Akoko-Edo (7.4%), with the least prevalence among the Yoruba's (3.7%). Race and ethnicity are known risk factors for acquiring bacterial vaginosis among sexually active black women.²³ BV is known to affects blacks differently and predisposes them to the acquisition of sexually transmitted diseases, human immunodeficiency virus, preterm labour, late miscarriage and other gynecologic infections.^{21,23}

This study also recorded no statistically significant relationship between marital status and prevalence of BV (p=1.000), though single women had higher prevalence (89%) when compared to their married counterparts. Single sexually active women

are more prone to BV because while the married females tend to stick to their spouse as the only sex partner, the single women practice more of the risk factors for acquiring BV such as: having multiple sexual partners²⁴. having a new sex partner as well as douching which can upset the balance of bacteria in the vagina and predispose them to bacterial vaginosis.²⁶

Frequency of sex as a risk factor did not significantly influence the prevalence of BV among sexually active females in our study (P=0.065). The highest prevalence of BV was seen among females who engage in sexual intercourse a few times a month (48.1%), females who had sexual intercourse a few times a week (37.0%), those who rarely had sexual intercourse recording 11.1% while those having sexual intercourse daily recording the least prevalence (3.7%). However, there was a statistical significant relationship between the prevalence of BV and the sexuality of respondents in another study.²¹ Though frequency of sex is a critical factor in the acquisition of BV, differences in our studies may be because higher prevalence rates of G. vaginalis have been found in sexually abused girls as compared to nonabused girls in most studies.8 BV also occurs among sexually inexperienced adolescents and virginal women but at lower rates on average as compared to sexually active reproductive aged-women.¹² Several studies support that BV incidence is increased by penetrative sexual activity, non penetrative digito-genital contact, oral sex but clearly also contradict exclusive heterosexual transmission. Moreso, unprotected receptive anal sex before vaginal intercourse also predisposes females to BV prevalence. 27,28

T.vaginalis was not isolated in the wet mount during the course of this study. A previous study among Nigerian students recorded 25% prevalence of BV from cultural study. 9.27 T.vaginalis is among the most prevalent cause of sexually transmitted infections globally, this makes its examination mostly done as part of the recommendation in the investigation of vaginal discharge. The wet mount microscopy is the most cost effective

diagnostic test for BV but lack of sensitivity contributes to the under diagnosis. This is because delay in transport and evaporation of moisture from the specimen reduces motility and consequently diagnostic sensitivity which might be the case in our study.

Candida albicans, a fungus was isolated among the study participants. The lactobacillus normally present in the female vagina creates an environment that does not encourage yeast overgrowth but disturbances in its delicate balance can lead to the development of yeast infection. Moreso, Candida albicans though not a sexually transmitted infection, can spread through oral-genital contact, during sexual intercourse and is also associated with history of recent masturbation with saliva by the participant as well as the sexual partner. ^{29,30,8}

CONCLUSION

This study recorded a high prevalence of BV among sexually active females in a Tertiary Institution in Benin Metropolis, Nigeria. The risk factors studied had no significant relationship with the acquisition of BV among the study population.

RECOMMENDATIONS

Public enlightenment on the existence of BV, its symptoms and effect, instructing women to stop douching, cessation of smoking and limiting the number of sex partners is advised.

ACKNOWLEDGEMENTS

The contribution of Mr Nosakhare Lawrence Idemudia to the laboratory analysis is acknowledged.

CONFLICTS OF INTEREST

The authors declare that there are no conflicting interests.

REFERENCES

- 1. Eschenbach DA, Thwin SS, Patton DL. Influence of the normal menstrual cycle on vaginal tissue, discharge and microflora, Clinical Infectious Diseases 2000;30:901-7.
- 2. Lakshmi K, Aishwarya CS, Menezes GA. Review on infectious vaginitis. Research Journal of Pharmaceutical,

- Biological and Chemical Sciences 2013;4:679-82.
- 3. Amabebe E, Anumba DO. The vaginal microenvironment: the physiologic role of Lactobacilli. Front Med (Lausanne) 2018;5:181-3.
- 4. Jones A. Bacterial Vaginosis: A review, Recurrence and Disparities. The Journal for Nurse Practitioners 2019:15:420-3.
- 5. Machado A, Cerca N. Influence of biofilm formation by Gardnerella vaginalis and other anaerobes on bacterial vaginosis. Journal of Infectious Diseases 2015;212:1856-61.
- 6. Allsworth JE, Peipert JF. Prevalence of bacterial vaginosis: 2001-2004 national health and nutrition examination survey data. Obstetrics and Gynecology 2007;109:114-20.
- 7. Coleman J, Gaydos C. Molecular diagnosis of bacterial vaginosis: an update. Journal of Clinical Microbiology 2018;56:342-8.
- 8. Verstraelen H, Verhelst R, Vaneechoutte M, Temmerman M. The epidemiology of bacterial vaginosis with sexual behaviour. BMC Infectious Diseases 2010:10:81-3.
- 9. Hay P, Ugwumadu A. Detecting and treating common sexually transmitted diseases. Best Practice Research Clinical Obstetrics Gynaecology 2009;23:647-60.
- 10. Tchamouroff SE, Panja SK. The association between receptive cunnilingus and bacterial vaginosis. Sexually Transmitted Infections 2000;76:144-5.
- 11. Papanikolaou EG, Tsanadis G, Dalkalitsis N, Lolis D. Recurrent bacterial vaginosis in a virgin adolescent: a new method of treatment. Infection 2002;30:403-4.
- 12. Vaca M, Guadalupe I, Erazo S, Tinizaray K, Chico ME, Cooper PJ. *et al*. High prevalence of bacterial vaginosis in adolescent girls in a tropical area of Ecuador. BJOG 2010;117:225-8.

- 13. Bump RC, Sachs LA, Buesching WJ. Sexually transmissible infectious agents in sexually active and virginal asymptomatic adolescent girls. Pediatrics 2006;77:488-94.
- 14. Donders GGG, Vereecken A, Bosmans E, Dekeersmaecker A, Salembier G, Spitz B. Definition of a type of abnormal vaginal flora that is distinct from bacterial vaginosis: aerobic vaginitis, BJOG: An International Journal of Obstetrics and Gynaecology 2002; 109:34-43.
- 15. Murta EFC, Silva AO, Silva EAC, Adad SJ. Frequency of infectious agents for vaginitis in non- and hysterectomized women. Archives of gynaecology and obstetrics 2005;273:152-6.
- 16. Numanoviæ F, Hukiæ M, Gegiæ M, Nukiæ M, Delibegoviæ Z, Pašiæ S. *et al.* Bacterial Vaginosis Presence in Sexually Active Women in Tuzla Canton Area. Bosnian Journal of Basic Medical Sciences 2008;8:322-30.
- 17. Bradshaw C, Sobel J. Current treatment of bacterial vaginosis. Limitations and need for innovation. Journal of Infectious Diseases 2016;214:S14-S20.
- 18. Nugent RP, Krohn MA, Hillier SL. Reliability of diagnosing bacterial vaginosis is improved by a standardized method of gram stain interpretation. Journal of Clinical Microbiology 1991;29:297-301.
- 19. Yudin MH, Money DM. Screening and management of bacterial vaginosis in pregnancy. Journal of obstetrics and gynaecology 2008;30:7028.
- 20. Achondou AE, Fumoloh FF, Aseneck AC, Awah AR, Utokoro AM. Prevalence of Bacterial Vaginosis Among Sexually Active Women Attending the CDC Central Clinic Tiko, South West Region, Cameroon. African journal of infectious diseases 2016;10:96-101.
- 21. Koumans EH, Sternberg M, Bruce C, McQuillan G, Kendrick J, Sutton M. *et al*. The prevalence of bacterial vaginosis in the United States, 2001-2004; associations with symptoms,

- sexual behaviours and reproductive health external icon. Sexually Transmitted Diseases 2007;34:864-9.
- 22. Kalra G, Subramanyak A, Pinto C. Sexuality: Desire, activity and intimacy in the elderly. Indian Journal of Psychiatry 2011;53:300-6
- 23. Brumley, J. "Testing a Model of Bacterial Vaginosis among Black Women," Doctor of Philosophy Dissertation, 2012;pp.44-5
- 24. Schwebke JR, Rivers C, Lee J. Prevalence of Gardnerella vaginalis in male sexual partners of women with and without bacterial vaginosis. Sexually Transmitted Diseases 2009;36:92-4.
- 25. Ranjit E, Raghubanshi BR, Maskey S, Parajuli P. Prevalence of Bacterial vaginosis and Its Association with Risk Factors among Non-pregnant Women: A hospital Based Study. International Journal of Microbiology 2018;9:10-23.
- 26. Tabrizi SN, Fairley CK, Bradshaw CS, Garland SM. Prevalence of Gardnerella vaginalis and Atopobium vaginae in virginal women. Sexually Transmitted Diseases 2006;33:663-5.
- 27. Fether KA, Fairley CK, Hocking JS, Gurrin LC, Bradshaw CS. Sexual risk factors and bacterial vaginosis: a systematic review and meta-analysis. Clinical Infectious Diseases 2008;47:1426-35.
- 28. Sharma AK, Ranjan R, Mehta G. Prevalence and determinants of reproductive tract infections among women. Journal of Communicable Diseases 2004;36:93-9.
- 29. Reed BD, Zazove P, Pierson CL, Gorenflo DW, Horrocks J. Candida transmission and sexual behaviours as risks for a repeat episode of Candida vulvovaginitis. Journal of Women's Health (Larchmt) 2003;12:979-89.
- 30. Rylander E, Berglund AL, Krassny C, Petrini B. Vulvovaginal candida in a young sexually active population: prevalence and association with orogenital sex and frequent pain at intercourse. Sex Transmitted Infection 2004;80:54-7.