Incidence of Bacterial Vagniosis and Aerobic Vaginitis before and after Pessary Insertion in Indonesia

Eighty Mardiyan Kurniawati¹, Faria Toma¹, Harry Parathon¹, Gatut Hardianto¹, Azami Azinar Denas¹, Kartuti Deborah², Firas Farisi Alkaff³

¹Departement of Obstetrics and Gynaecology, ²Departement of Microbiology, ³Department of Pharmacology, Faculty of Medicine, Universitas Airlangga, Dr. Soetomo General Hospital, Surabaya, Indonesia

ABSTRACT

Background: Pelvic organ prolaps (POP) is the most common gynecological condition in women and its incidence increases with age. Among all treatments option available, the most commonly used was pessary treatment. However, the use of pessary has several side effects, and the most common one is leucorrhoea due to Bacterial Vaginosis (BV) or Aerobic Vaginitis (AV). This study aims to evaluate the incidence of BV and AV before and after pessary insertion in postmenopausal women with POP in Indonesia.

Method: This study was an observational analytical study conducted at outpatient clinic Department of Obstetric and Gynecology Dr. Soetomo General Hospital on August-October 2016. Subject of this study was postmenopausal women with POP who were about to be treated with pessary. Vaginal secretion samples were taken before pessary insertion and 4 weeks after insertion. Gram staining, microbiology culture, and pH evaluation was done for BV and AV evaluation.

Result: There were a total of 20 patients included in this study. The incidence of BV before and 4 weeks after pessary insertion was significantly higher (0 vs 16, p < 0.05), while the incidence of AV before and 4 weeks after pessary insertion was not significantly higher (1 vs 5, p = 0.29). There was a significant increase in the pH evaluation before and after pessary insertion (6.2 \pm 0.62 vs 6.4 \pm 0.53, p= 0.008). The most common microorganism found in vaginal swab was *Escherichia coli*.

Conclusion: BV incidence was significantly increased after pessary insertion in postmenopausal women with POP.

Keywords: Aerobic Vaginitis, Bacterial Vaginosis, Postmenopause, Pelvic organ prolapse, Pessary

Introduction

Pelvic organ prolaps (POP) is the most common gynecological condition in women and its incidence increases with age. It is defined as the descent of uterus and vaginal walls through vaginal canal ¹. Not only causing discomfort, POP is also having a negative impact on quality of life.

Corresponding Author:

Eighty Mardiyan Kurniawati
Departement of Obstetrics and Gynaecology,
Faculty of Medicine, Universitas Airlangga,
Dr. Soetomo General Hospital,
Surabaya, East Java, Indonesia
Phone: +6287851971399

Email: eightymardiyankurniawati@gmail.com

Treatment for POP is offered when the patients develop symptoms attributable to the prolapse that they find bothersome. Until now there are 2 available option for treating POP, which are conservative treatment and surgical treatment. However, surgical treatment is generally reserved when conservative treatment has failed 2. One of the conservative treatments for POP is the use of pessary. From many types of pessaries, ring pessary have been widely used as conservative therapy for women with pelvic organ prolapse because of its easiness for insertion and removal. Compare to surgical therapy, patients with pessary reported to experience similar improvement in micturition, bowel movements, and quality of life 3. However, the use of pessary has several side effects, and the most common one is leucorrhoea. A study in postmenopausal women

using pessaries reported a significant increase in vaginal discharge, leukocytes, and parabasal cells which illustrate the presence of vaginal inflammation ⁴. The inflammation is caused by either Bacterial Vaginosis (BV) or Aerobic Vaginitis (AV) due to the changes in the vaginal ecosystem ^{4,5}.

BV is a clinical condition which is characterized by changes in vaginal ecology that normally contains morphotype Lactobacilli spp flora into a mixture of flora, specifically a combination of anaerobic bacteria and Gardnerella vaginalis ⁶. AV, on the other hand, is a condition where Lactobacillus flora is disrupted, followed by signs of inflammation and predominance of aerobic bacteria composed of commensal or pathogenic enteric bacteria ^{6,7}.

The composition of Lactobacillus in the vagina of each individual is different, and it depends on geographic location, race and ethnicity *. In Indonesia, analytic studies examining the effect of the pessary insertion in pelvic organ prolapse patients on the incidence of BV and AV is scarce. Given the geography, race and ethnicity of Indonesian women that are different from other countries, this study aims to analyze the difference of BV and AV incidence before and after pessary insertion in postmenopausal women with POP.

Material and Method

This study was an observational study conducted from August to October 2016 at Department of Gynaecology Dr. Soetomo General Hospital Outpatient Clinic, Surabaya, Indonesia. Subject of this study was postmenopausal women with POP who were about to be treated conservatively with pessary. POP were diagnosed according to the Pelvic Organ Prolapse Quantification System (POP-Q) 9. Patients with intrauterine device, underwent hormonal replacement therapy, and had an uncontrolled diabetes mellitus were excluded from this study. Drop-out criteria for this study was patients whose pessary was expulsed before four weeks of insertion and those who consumed antibiotics during the first four weeks of pessary insertion.

Vaginal secretion samples were taken before pessary insertion and 4 weeks after insertion. Gram staining, microbiology culture, and pH evaluation was done for BV and AV evaluation. BV was evaluated using Nugent criteria scoring system ¹⁰. AV was evaluated using

microscopic diagnosis criteria 7. Microbiology culture was done in blood agar plate for aerobic organism and MacConkey agar plate for gram-negative aerob organism. Culture was done at the Clinical Microbiology Laboratory Dr. Soetomo General Hospital.

Acquired data was analysed using SPSS version 18.0. Data distribution was analysed using Shappiro-Wilk test. Incidence of BV and AV before and after pessary insertion was analysed using McNemar test. pH evaluation before and after pessary insertion was analysed using paired T-test. The p-value of < 0.05 was considered as statistically significant.

This study follows the principles of the Declaration of Helsinki. This study had received ethical clearance from Dr. Soetomo General Hospital before the study began. All subjects gave their informed consent prior to their inclusion in the study. Information for informed consent was given before subjects signed the informed consent. Details that might disclose the identity of the subjects under study were omitted.

Results

There were 26 patients with POP that met the criteria to be involved in this study. During the study period, 6 patients were dropped out from the study because of pessary expulsion and antibiotic use. The average age of the study subject was 63.8 ± 8.2 years. The menopause duration was 14.9 ± 8.5 years. Most of the study subjects were in the POP stage 3. There were 2 patients who had an intercourse during the first 4 week of pessary insertion (table 1).

Table 1: Characteristics of the Study Subject

Characteristics	N= 20 n (%)
Age (mean \pm SD)	63.8 ± 8.2
Menopause duration (mean ± SD)	14.9 ± 8.5
BMI (mean ± SD)	25.3 ± 2.39
Education attainment	
Elementary school	9 (45)
Middle school	7 (35)
High school	4 (20)
Degree of Prolapse	
Stage 2	3 (15)
Stage 3	17 (85)

Conted ...

Smoking	
Yes	0
No	20 (100)
Vaginal Douching	
Yes	0
No	20 (100)
Intercourse	
Yes	2 (10)
No	18 (90)

Before pessary insertion, no patient had BV and only 1 patient were positive for AV. Four weeks after pessary insertion, 16 patients were positive for BV and 5 patients were positive for AV (table 2). From 16 vaginal swab samples of BV patients, none of the samples had *Lactobacillus* and *Mobiluncus*. Six samples had *Bacteroides/Gardnerella* as many as 5-30/visual field, and 10 samples had more than 30/visual field. No clue cell was found in all samples. The pH value

of patients' vaginal swab specimens before and after pessary insertion was significantly different $(6.2 \pm 0.62 \text{ vs } 6.4 \pm 0.53, \text{ p}=0.008)$. Microbiological culture of vaginal swab specimens showed that the most common microorganism before and after pessary insertion was *Escherichia coli*, and the second most common was *Staphylococcus coagulase negative* (table 3).

Table 2: Incidence of Bacterial Vaginosis and Aerobic Vaginitis before and 4 weeks after pessary insertion

	Pre-pessary insertion n (%)	Post-pessary insertion n (%)	p-value
Bacterial Vaginosis	0	16 (40)	<0.05**
Aerobic Vaginitis	1 (2,5)	5 (12,5)	0.219"

*p < 0.05 was considered statistically significant #McNemar test was used

Table 3: Distribution of microbiological culture from vaginal swab specimens before and 4 weeks after pessary insertion

Isolates	Pre-pessary insertion N= 20 n (%)	Post-pessary insertion N = 20 n (%)
Gemella haemolysan	1 (1,9)	1 (1,9)
Streptococcus α haemolyticus	2 (3,8)	1 (1,9)
Corynebacterium spp.	2 (3,8)	1 (1,9)
Escherichia coli	8 (15,3)	12 (23)
Proteus mirabilis	1 (1,9)	0
Staphylococcus haemolyticus	2 (3,8)	1 (1,9)
Staphylococcus aureus	1 (1,9)	2 (3,8)
Streptococcus viridans	1 (1,9)	1 (1,9)
Streptococcus mutans	1 (1,9)	1 (1,9)
Streptococcus non haemolyticus	1 (1,9)	0
Pseudomonas spp.	1 (1,9)	0
Morganella morgani	0	1 (1,9)
Streptococcus uberis	0	1 (1,9)
Streptococcus β haemolyticus	0	1 (1,9)
Staphylococcus coagulase negative	3 (5,7)	4 (7,69)
Streptococcus agalactiae	1 (1,9)	0

Discussion

Characteristics of Study Subjects: Subjects of this study were postmenopausal women ranging from 48 to 80 years. Previous study from South Korea found that the prevalence of POP increases with age and reach it peaks on the age of 75 years old ¹¹. Majority of the patients had elementary education, followed by secondary education.

Educational level is known to correlate with individual hygiene and sanitation. A study involving 500 married non-pregnant women revealed a significant association between the incidence of Bacterial Vaginosis and level of education (p <0.025) ¹². Another study involving 85 sex workers also revealed that the level of education was significantly related to the incidence of sexually transmitted diseases ¹³.

Behaviors such as sexual intercourse, smoking and douching also have an effect on the incidence of BV. In this study, all patients did not smoke or douche. There were 2 patients that had sexual intercourse during the study, with a frequency of 1 to 2 times in four weeks. A prospective study involving 1248 women who did not initially suffer BV reported that the incidence of BV was associated with smoking, douching, and not using contraception, with risk factors for non-white races including decreased *Lactobacillus*-produced H₂O₂, having two or more sexual partners in in the past four months and have had sex more than 3 times a week ¹⁴.

Incidence of BV before and after pessary insertion: BV is a clinical condition characterized by changes in vaginal ecology, which in normal conditions contains morphotype flora Lactobacilli spp into a mixture of flora combined of anaerobic bacteria and Gardnerella vaginalis. It is defined as an abnormal vaginal ecosystem characterised by a decreased concentration of Lactobacillus and an increased concentration of anaerobic bacteria such as Bacteroides spp., Mobiluncus spp., and Gardnerella vaginalis. There are four bacteria related to BV, including Gardnerella vaginalis, Gram negative anaerobic rod bacteria, Mycoplasma hominis, and Mobiluncus spp.

Incidence of BV before and after pessary insertion in this study was statistivally significant (p< 0.05). A case control study at Mount Sinai Hospital involving 44 women with POP using pessary with a control of 176 women found that pessary insertion significantly increased the incidence of BV (OR 4.37), where the incidence mostly occured in the first 6 months after the insertion ⁵.

BV is evaluated based on Nugent criteria and/or the presence of clue cells ¹⁰. In this study, the diagnosis of BV is based on a decrease in the number of *Lactobacillus* and an increase in *Bacterioides/Gardnerella like*. In microscopic examination of vaginal swab specimens from patients with positive BV, *Lactobacillus*

morphotype was found in large quantities. The absence of Lactobacillus in vaginal swabs in this study is most likely because subjects were postmenopausal women, hence the decrease in estrogen resulted in a decrease in the amount of Lactobacillus. This is in accordance with severeal studies which state that in postmenopausal women, the decrease in estrogen results in a reduced in glycogen metabolism in epithelial cells, which causes a decrease in the amount of Lactobacillus and an increase in vaginal pH ^{6,15}. However, no clue cells were found in all positive BV specimens. We argue that the absence of clue cell formation in this study is likely due to fewer amounts of Gardnerella vaginalis compared to other anaerobic bacteria ¹⁶.

In this study, it was found that the average of vaginal pH before pessary insertion was 6.2 ± 0.62 and significantly increased to 6.4 ± 0.53 in 4 weeks after the insertion. An increase in vaginal pH will result in a shift in vaginal flora towards the mixed flora that causes BV and AV ¹⁷. In normal condition, the average pH of postmenopausal women is > 5.3 ¹⁸. According to one of the Amsel's criteria for the diagnosis of BV, vaginal pH had to be > 4.5 ¹⁹. Therefore, Amsel's criteria could not be used to diagnose BV in postmenopausal women.

Incidence of AV before and after pessary insertion: AV is an abnormal condition vaginal ecosystem with reduced concentration of Lactobacillus and increased concentration of aerobes such as Escherichia coli, Staphylococcus aureus, group B Streptococcus and Enterococcus. AV is diagnosed based on an increased leukocyte count of ≥10, decreased Lactobacillus, and the presence of parabasal cells and aerobic bacteria in vaginal swabs ⁷. In this study, AV is defined as an AV score of ≥3.

Incidence of AV post-pessary insertion was higher compare to pre-pessary insertion. However, the difference was not statistically significant. The result of this study is in accordance with a study involving 100 postmenopausal women, which showed an increase in leukocytes and parabasal cells at 2 weeks, 3 months and 6 months of pessary insertion. Pathological organisms tend to increase after pessary insertion although the increase is not significant.

The most common bacterial isolate in vaginal fluid culture was *Escherichia coli*. This finding is consistent with a study by Donders *et al.* which states that the most common microorganism found in AV is Escherichia coli
7. A study by Hitti et al, reported that Escherichia coli was the only microorganism found in the vaginal fluid culture of women with vaginal infections ¹⁴.

Conclusion

Incidence of BV in Indonesia increases significantly after pessary insertion in postmenopausal women with POP, while AV incidence did not increase significantly.

Conflict of Interest: The authors declare that there is no conflict of interest regarding the publication of this article.

Source of Funding: This study was privately funded by the authors.

REFERENCES

- Aytan H, Ertunc D, Tok EC, Yasa O, Nazik H. Prevalence of pelvic organ prolapse and related factors in a general female population. Turkish journal of obstetrics and gynecology. 2014;11(3):176-80.
- Barber MD. Pelvic organ prolapse. BMJ. 2016;354:i3853.
- Atnip SD. Pessary use and management for pelvic organ prolapse. Obstetrics and gynecology clinics of North America. 2009;36(3):541-63.
- Collins S, Beigi R, Mellen C, O'Sullivan D, Tulikangas P. The effect of pessaries on the vaginal microenvironment. American journal of obstetrics and gynecology. 2015;212(1):60 e1-6.
- Alnaif B, Drutz HP. Bacterial vaginosis increases in pessary users. International urogynecology journal and pelvic floor dysfunction. 2000;11(4):219-22; discussion 22-3.
- Nikolaitchouk N, Andersch B, Falsen E, Strombeck L, Mattsby-Baltzer I. The lower genital tract microbiota in relation to cytokine-, SLPIand endotoxin levels: application of checkerboard DNA-DNA hybridization (CDH). APMIS: acta pathologica, microbiologica, et immunologica Scandinavica. 2008;116(4):263-77.
- Donders G, Bellen G, Rezeberga D. Aerobic vaginitis in pregnancy. BJOG: an international journal of obstetrics and gynaecology. 2011;118(10):1163-70.
- Rampersaud R, Randis TM, Ratner AJ. Microbiota of the upper and lower genital tract. Seminars in fetal & neonatal medicine. 2012;17(1):51-7.

- Kuncharapu I, Majeroni BA, Johnson DW. Pelvic organ prolapse. American family physician. 2010;81(9):1111-7.
- Muthusamy S, Elangovan S. Comparison of Amsel's Criteria, Nugent Score and Culture for the Diagnosis of Bacterial Vaginosis. National Journal of Laboratory Medicine. 2016;5(1):37-40.
- Yuk J-S, Lee JH, Hur J-Y, Shin J-H. The prevalence and treatment pattern of clinically diagnosed pelvic organ prolapse: a Korean National Health Insurance Database-based cross-sectional study 2009–2015. Scientific Reports. 2018;8(1):1334.
- Bahram A, Hamid B, Zohre T. Prevalence of bacterial vaginosis and impact of genital hygiene practices in non-pregnant women in zanjan, iran. Oman medical journal. 2009;24(4):288-93.
- Solomon MM, Smith MJ, del Rio C. Low educational level: a risk factor for sexually transmitted infections among commercial sex workers in Quito, Ecuador. International journal of STD & AIDS. 2008;19(4):264-7.
- Hitti J, Hillier SL, Agnew KJ, Krohn MA, Reisner DP, Eschenbach DA. Vaginal indicators of amniotic fluid infection in preterm labor. Obstetrics and gynecology. 2001;97(2):211-9.
- Farage MA, Miller KW, Sobel JD. Dynamics of the Vaginal Ecosystem—Hormonal Influences. Infectious Diseases: Research and Treatment. 2010;3:IDRT.S3903.
- Scott TG, Smyth CJ, Keane CT. In vitro adhesiveness and biotype of Gardnerella vaginalis strains in relation to the occurrence of clue cells in vaginal discharges. Genitourinary medicine. 1987;63(1):47-53.
- Zhou X, Bent SJ, Schneider MG, Davis CC, Islam MR, Forney LJ. Characterization of vaginal microbial communities in adult healthy women using cultivation-independent methods. Microbiology. 2004;150(Pt 8):2565-73.
- Panda S, Das A, Singh AS, Pala S. Vaginal pH: A marker for menopause. Journal of mid-life health. 2014;5(1):34-7.
- Udayalaxmi, Bhat G, Kotigadd S, Shenoy S. Comparison of the Methods of Diagnosis of Bacterial Vaginosis. JCDR. 2011;5(3):498-501.