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Correlation vaginal microbiota and risk factor on pelvic organ prolapse

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Abstract

Background: Pelvic organ prolapse (POP) is a common pelvic floor disorder that markedly affects women's quality of life. Recent evidence suggests that vaginal microbiota imbalance may play a role in pelvic floor dysfunction; however, its association with POP risk factors remains poorly understood.

Aim: This study aimed to evaluate the relationship between vaginal microbiota composition and clinical risk factors in women with pelvic organ prolapse.

Method: A cross-sectional analytical study was conducted involving women diagnosed with POP and a comparison group without POP. Vaginal swab samples were analyzed to determine microbial composition at the phylum and genus levels.

Result: Age above 60 years and menopausal duration of five years or more were significantly associated with increased POP risk. Bacillota was the predominant phylum in both groups, with higher proportions in the non-POP group. Women with POP showed greater microbial diversity and reduced protective bacterial profiles, indicating vaginal dysbiosis.

Conclusion: Advanced age and prolonged menopause are key risk factors for POP and are associated with alterations in vaginal microbiota. These findings suggest that aging-related microbial shifts may contribute to POP development.

Key words: Pelvic organ prolapse, vaginal dysbiosis, menopause, bacterial diversity, risk factors

Introduction

Pelvic organ prolapse (POP) is a condition characterized by the descent of the anterior, posterior, and/or apical vaginal compartments, resulting in the protrusion of one or more pelvic organs such as the

bladder, uterus, post-hysterectomy vaginal vault, or rectum into the vaginal canal. This condition arises from the loss of structural support of the pelvic organs and significantly affects women's quality of

life. The pathophysiology of POP involves injury and degeneration of the muscles, nerves, and connective tissues that support the pelvic floor and its contents.¹ Pelvic organ prolapse represents a major burden on women's health services worldwide, with reported prevalence reaching up to 40%.²

The incidence of POP increases with advancing age, and it is more commonly observed in women over 40 years old, with prevalence rates reported as high as 50%. Globally, POP prevalence ranges from 2.9% to 41.1% in low-income countries.³ In Indonesia, although national epidemiological data are limited, a study reported a POP prevalence of 26.4%, indicating that the condition constitutes a substantial public health concern.⁴ Several risk factors have been identified for the development of pelvic organ prolapse, including advanced age, parity, vaginal delivery, and increased body mass index (BMI), all of which contribute to pelvic floor tissue damage. A family history of POP has also been recognized as a significant risk factor, suggesting a possible genetic predisposition.⁵

Although POP is not a life-threatening condition, its symptoms can profoundly impair physical functioning, emotional well-being, and overall quality of life.⁶ Clinically, POP may cause a sensation of vaginal fullness or a visible or palpable bulge at the vaginal introitus. Women with POP frequently experience stress urinary incontinence, particularly when urethral support is compromised, as well as voiding dysfunction due to urethral kinking. These urinary disturbances can manifest as increased urinary frequency, hesitancy, weak urinary stream, and incomplete bladder emptying. Such conditions promote urinary stasis, which creates a favorable environment for bacterial growth.⁷

Decreased pelvic floor muscle strength and vaginal wall laxity may facilitate microbial invasion into the vaginal canal, thereby increasing susceptibility to pathological conditions such as bacterial vaginosis

(BV) and pelvic inflammatory disease (PID).⁸ Previous studies in Indonesia have reported a high prevalence of bacterial vaginosis among women with pelvic organ prolapse, reaching 76.7%. Age and menopausal status were found to be significantly associated with the occurrence of BV in this population. Pelvic organ prolapse has also been identified as a risk factor for pelvic inflammatory disease, in which vaginal microbiota and pathogenic organisms play critical roles in disease development and in related infertility issues.⁹

The vaginal microbiome is a complex and dynamic microecosystem that undergoes continuous fluctuations throughout the menstrual cycle and across a woman's lifespan. The vaginal mucosa consists of stratified non-keratinized squamous epithelium covered by cervicovaginal secretions.¹⁰ Due to limited direct blood supply, oxygen, glucose, and other nutrients diffuse from the underlying submucosal tissues, creating a relatively anaerobic environment. Within this niche, diverse microbial communities coexist in symbiosis with the host.¹¹ Lactobacillus species dominate healthy vaginal microbiota by producing antimicrobial substances such as lactic acid, hydrogen peroxide (H₂O₂), and bacteriocins, thereby maintaining an acidic environment and providing defense against pathogenic organisms. The composition of vaginal microbiota is therefore essential for maintaining vaginal health and preventing disease.¹²

Material and methods

Study Design

This study employed an analytical observational design using a cross-sectional approach. The design was selected to assess the correlation between vaginal microbiota composition and associated risk factors in women diagnosed with pelvic organ prolapse at a single point in time. The study was conducted at several teaching hospitals affiliated with the Department of Obstetrics and Gynecology, Faculty of Medicine, Hasanuddin University, including Dr.

Wahidin Sudirohusodo General Hospital and other affiliated teaching hospitals. Data collection was carried out from January to July 2025. The study population consisted of all women diagnosed with pelvic organ prolapse who attended the Department of Obstetrics and Gynecology at the teaching hospitals of the Faculty of Medicine, Hasanuddin University, Makassar, during the study period.

Sample Size and Sampling Technique

The study sample included menopausal women diagnosed with pelvic organ prolapse who met the inclusion and exclusion criteria. Participants were recruited using a consecutive sampling technique, whereby all eligible patients presenting at the study sites during the data collection period were enrolled until the required sample size was achieved. The minimum sample size was determined using the Harry King Nomogram method. This method estimates sample size by drawing a straight line from the total population size through the desired margin of error to determine the corresponding sample proportion.

Eligibility Criteria

Inclusion Criteria: Participants were included if they met the following criteria: women aged over 40 years, clinically diagnosed with pelvic organ prolapse, and willing to participate in the study as evidenced by signing a written informed consent form. **Exclusion Criteria:** Participants were excluded if they had used antibiotics within one week prior to sampling, were undergoing hormone replacement therapy, were menstruating or pregnant at the time of data collection, had documented mental disorders, or had consumed probiotic supplements within the preceding two months.

Data Collection Procedures

Eligible participants who agreed to take part in the study were consecutively enrolled. Prior to data col-

lection, participants received a detailed explanation regarding the study objectives, procedures, potential risks, and benefits, after which written informed consent was obtained. Demographic data and clinical characteristics were collected using a structured research form. Anamnesis and physical examination were performed, including assessment of vital signs and documentation of established risk factors for pelvic organ prolapse. Vaginal sample collection was conducted after providing adequate explanation to reduce participant anxiety. Participants were positioned in the lithotomy position, and aseptic procedures were strictly followed, including hand hygiene and the use of personal protective equipment. Using a sterile speculum, a sterile swab was gently inserted 1-2 cm into the endocervical canal, rotated carefully, and withdrawn slowly. Endocervical swab specimens were obtained under sterile conditions.

Study Instruments and Materials

Materials used in this study included informed consent forms, structured research questionnaires, sterile gloves, masks, sterile swabs, specimen labels, and documentation equipment. Collected data were entered and tabulated using Microsoft Excel before statistical analysis. Univariate analysis was conducted to describe baseline characteristics of the study population, presented as frequency distributions and percentages in tabular form. Bivariate analysis was performed to assess the association between vaginal microbiota composition and identified risk factors using the chi-square test for categorical variables. Statistical significance was set at a p-value < 0.05 with a 95% confidence level. All analyses were conducted using SPSS version 27.

Ethical Approval

This study was conducted in accordance with ethical principles for medical research involving human subjects. Written informed consent was

obtained from all participants prior to enrollment. Ethical approval was granted by the Biomedical Research Ethics Committee of the Faculty of Medicine, Hasanuddin University, and the Health Research Ethics Committee (KEPK) of RSPTN UH-RSWS, with approval number: 372/UN4.6.4.5.31/PP36/2025.

Results

A comparative analysis of 60 Pelvic Organ Prolapse (POP) patients and 60 non-POP controls revealed significant demographic and clinical differences

(Table 1). POP patients were significantly older ($p < 0.001$) and had a longer duration of menopause ($p < 0.001$). All POP patients (100%) engaged in heavy lifting, a significantly higher proportion than controls ($p = 0.006$). Vaginal deliveries were more prevalent in the POP group (91.7% vs. 76.7% in controls; $p = 0.031$). Notably, 100% of non-POP controls used genital cleansers, while 21.7% of POP patients did not ($p < 0.001$). BMI and parity did not differ significantly between groups (Table 1).

Table 2. presents the distribution and relative

Table 1. Characteristics of research subjects.

CHARACTERISTICS	POP (N = 60) n (%)	NON-POP (N = 60) n (%)	P-VALUE
Age			
≤ 60 years	21 (35.0)	50 (83.3)	< 0.001
> 60 years	39 (65.0)	10 (16.7)	
BMI			
Underweight	1 (1.7)	3 (5.0)	0.360
Normal	23 (38.3)	15 (25.0)	
Overweight	11 (18.3)	14 (23.3)	
Obesity	25 (41.7)	28 (46.7)	
Parity			
Primipara	1 (1.7)	3 (5.3)	0.285
Multipara	33 (55.0)	36 (63.2)	
Grand multipara	26 (43.3)	18 (31.6)	
Duration of menopause			
No	4 (6.7)	21 (35.0)	< 0.001
< 5 years	5 (8.3)	15 (25.0)	
≥ 5 years	51 (85.0)	24 (40.0)	
Work			
Activity < 10 kg	0 (0.0)	8 (13.3)	0.006
Activity ≥ 10 kg	60 (100.0)	52 (86.7)	
POP complaint duration			
< 5 years	39 (65.0)	0 (0.0)	-
≥ 5 years	21 (35.0)	0 (0.0)	
Labor			
Vaginal	55 (91.7)	46 (76.7)	0.031
Caesarean section	0 (0.0)	5 (8.3)	
Vaginal and caesarean section	5 (8.3)	9 (15.0)	
Use of genital cleansers			
Yes	47 (78.3)	60 (100.0)	< 0.001
No	13 (21.7)	0 (0.0)	

Chi-square test, significant ($p < 0.05$)

Table 2. Percentage of bacterial genera in POP.

GENUS OF GERMS	N	%	PHYLUM	INFORMATION
Gram positive				
Lactobacillus	9	15.0	Bacillota	Commensal
Atopobium	9	15.0	Actinomycetota	Pathogen
Eggerthella	2	3.3	Gordonibacter	Pathogen
Gordonibacter	2	3.3	Gordonibacter	Pathogen
Olsenella	2	3.3	Firmicutes	Pathogen
Streptomyces	2	3.3	Actinomycetota	Pathogen
Adlercreutzia	2	3.3	Actinomycetota	Pathogen
Coriobacteriales	2	3.3	Actinomycetota	Pathogen
Amylolactobacillus	2	3.3	Bacillota	Pathogen
Pediococcus	2	3.3	Bacillota	Pathogen
Companilactobacillus	2	3.3	Bacillota	Pathogen
Lentilactobacillus	2	3.3	Bacillota	Pathogen
Schleiferilactobacillus	1	1.7	Bacillota	Pathogen
Corynebacterium	1	1.7	Actinomycetota	Pathogen
Bifidobacterium	1	1.7	Actinomycetota	Pathogen
Micrococcus	1	1.7	Actinomycetota	Pathogen
Gram negative				
Prevotella	12	20.0	Bacteroidota	Pathogen
Gardnerella	11	18.3	Actinomycetota	Pathogen
Escherichia	8	13.3	Pseudomonads	Pathogen
Bacteroides	5	8.3	Bacteroidota	Pathogen
Shigella	5	8.3	Pseudomonads	Pathogen
Hoylesella	4	6.7	Bacteroidota	Pathogen
Pseudomonas	3	5.0	Pseudomonads	Pathogen
Phocaeicola	3	5.0	Bacteroidota	Pathogen
Salmonella	2	3.3	Pseudomonads	Pathogen
Stenotrophomonas	2	3.3	Pseudomonads	Pathogen
Chryseobacterium	2	3.3	Bacteroidota	Pathogen
Phyllobacterium	1	1.7	Pseudomonads	Pathogen
Segatella	1	1.7	Bacteroidota	Pathogen
Xylanibacterium	1	1.7	Bacteroidota	Pathogen
Flavobacterium	1	1.7	Bacteroidota	Pathogen
Gammaproteobacteria	1	1.7	Pseudomonads	Pathogen
Rhizobium	1	1.7	Pseudomonads	Pathogen
Arcobacte	1	1.7	Proteobacteria	Pathogen

abundance of bacterial genera identified in vaginal microbiota among patients with pelvic organ prolapse (POP). The data are categorized based on Gram staining characteristics (Gram-positive and Gram-negative bacteria), bacterial genus, number of isolates (n), percentage (%), phylum classification, and biological role (commensal or pathogen). A shift in vaginal microbiota composition in POP patients, characterized by reduced dominance of protective commensal *Lactobacillus* and increased presence of diverse pathogenic bacterial genera, particularly Gram-negative anaerobic and facultative anaerobic bacteria. This microbial imbalance (dysbiosis) may contribute to inflammatory processes and tissue vulnerability, potentially playing a role in the pathophysiology of pelvic organ prolapse.

Table 3 present the association between age, duration of menopause, and vaginal bacterial profiles among women with pelvic organ prolapse (POP). The presence and absence of *Lactobacillus*, *Gardnerella vaginalis*, *Prevotella*, *Atopobium*, and *Fannyhessea vaginae* were analyzed in relation to age categories and menopausal duration. Regarding age, *Lactobacillus* was found in 14.3% of women aged ≤ 60 years and 15.4% of those aged >60 years, with no statistically

significant association observed ($p = 1.000$). Similarly, no significant differences were found between age groups for the presence of *Gardnerella vaginalis* ($p = 0.493$), *Prevotella* ($p = 0.737$), *Atopobium* ($p = 0.142$), or *Fannyhessea vaginae* ($p = 0.142$). These findings indicate that age was not significantly associated with the distribution of these bacterial genera in women with POP. In terms of menopausal duration, no significant associations were identified between the duration of menopause and the presence of *Lactobacillus* ($p = 0.663$), *Gardnerella vaginalis* ($p = 0.382$), *Prevotella* ($p = 0.967$), *Atopobium* ($p = 0.618$), or *Fannyhessea vaginae* ($p = 0.393$). Both women who had not yet reached menopause and those with menopausal durations of less than 5 years or 5 years and above showed comparable bacterial distribution patterns.

Table 4. show the non-POP, Gram-positive taxa were primarily derived from the phyla Bacillota, Fusobacteriota, and Campylobacterota. *Lactobacillus* dominated the Gram-positive community, identified in 83.3% of subjects, followed by *Streptococcus* (43.3%) and members of Fusobacteriota (16.7%). Gram-negative organisms in non-POP women originated from the phyla Bacillota and Legionella, with

Table 3. Association Between Age, Duration of Menopause, and Vaginal Bacterial Profiles in Women With Pelvic Organ Prolapse.

RISK FACTOR	LACTOBACILLUS			GARDNERELLA VAGINALIS			PREVOTELLA			ATOPOBIUM			FANNYHESSEA VAGINAE		
	FOUND (N= 9)	NOT FOUND (N = 51)	P-VALUE	FOUND (N= 11)	NOT FOUND (N = 49)	P-VALUE	FOUND (N= 12)	NOT FOUND (N = 48)	P-VALUE	FOUND (N= 21)	NOT FOUND (N = 39)	P-VALUE	FOUND (N= 21)	NOT FOUND (N = 39)	P-VALUE
	N (%)	N (%)		N (%)	N (%)		N (%)	N (%)		N (%)	N (%)		N (%)	N (%)	
Age															
≤ 60 years	3 (14,3)	18 (85,7)	1,000	5 (45,5)	16 (32,7)	0,493	5 (23,8)	16 (76,2)	0,737	1 (4,8)	8 (20,5)	0,142	1 (4,8)	8 (20,5)	0,142
> 60 years	6 (15,4)	33 (84,6)		6 (54,5)	33 (67,3)		7 (17,9)	32 (82,1)		20 (95,2)	31 (79,5)		20 (95,2)	31 (79,5)	
Duration of menopause															
Not yet	0 (0)	4 (100)	0,663	1 (25,0)	3 (6,1)	0,382	1 (25)	3 (75)	0,967	0 (0,0)	4 (7,3)	0,618	0 (0,0)	4 (100)	0,393
≥ 5 years	1 (20)	4 (80)		2 (40,0)	43 (87,8)		10 (19,8)	41 (80,4)		5 (100,0)	46 (83,6)		9 (17,6)	42 (82,4)	
< 5 years	8 (15,7)	43 (84,3)		8 (15,7)	3 (6,1)		1 (20)	4 (80)		0 (0,0)	5 (9,1)		0 (0)	5 (100)	

Table 4. Percentage of bacterial genera in non-POP.

GENUS OF GERMS	N	%	PHYLUM	INFORMATION
Gram positive				
Lactobacillus	50	83.3	Bacillota	Commensal
Streptococcus	26	43.3	Bacillota	Opportunistic pathogens (Bacterialvaginosis,)
Fusobacteriota	10	16.7	Fusobacteriota	Opportunistic pathogens (Bacterial vaginosis)
Campylobacter	9	15	Campylobacterota	Pathogen (Diarrhea)
Gram negative				
Dialister	21	35	Bacillota	Pathogen (Bacteremia)
Legionella	15	25	Legionella	Pathogen (Pneumonia)

Dialister (35%) and Legionella (25%) as the predominant genera.

Table 5. present the associations between demographic, obstetric, and clinical factors and the presence of vaginal bacterial genera (*Prevotella*, *Gardnerella*, *Escherichia*, *Shigella*, and *Hoylesella*) among women with pelvic organ prolapse. Overall, no statistically significant associations were identified between age, body mass index, parity, duration of menopause, mode of delivery, duration of pelvic organ prolapse, history of pessary use, or use of genital cleansing products and the detection of any bacterial genus (all p-values > 0.05). The distribution of bacterial presence was relatively comparable across all examined categories. Although certain trends were observed such as a higher proportion of *Hoylesella* among women aged ≤60 years and primiparous participants, and predominant bacterial detection among women with ≥5 years of menopause or vaginal delivery these differences did not reach statistical significance.

Discussion

This study suggest that advanced age and duration of menopause longer than 5 years are important risk factors for POP. These results are consistent with the studies by Nizomy et al, 2013, which reported

a significant and moderately strong correlation between menopausal status, increasing age, and POP occurrence.¹³ POP has been shown to occur more frequently in women aged over 60 years. A retrospective study by Pipara et al, 2015 involving patients who underwent urogynecological surgery between January 2011 and December 2014 demonstrated that most cases occurred in women older than 60 years.¹⁴ Furthermore, data from the Global Burden of Disease (GBD) 2019 study across 195 countries indicated that women aged over 50 years represent the greatest global burden of POP. POP is highly prevalent among women over 40 years, elderly women, and postmenopausal women, with an estimated prevalence of 41%-50%.¹⁵

The prevalence of POP increases with age, peaking among women aged 60-69 years. Age has also been shown to influence vaginal microbiota composition. Other studies reported that women younger than 60 years exhibited higher abundances of *Murdochella*, *Megasphaera*, *Peptoniphilus*, and *Ezakiella*, whereas women older than 60 years showed greater abundance of *Escherichia* *Shigella* and *Hungatella*, particularly in critically ill patients.¹⁶ Similarly, elderly women (>60 years) demonstrate the greatest differences in uterine microbiota compared with other age groups. In contrast, variations in vaginal microbiota appear earlier, with the most pronounced

Table 5. The relationship between POP risk factors and the genus of Gram-negative bacteria.

Factor	Prevotella			Gardnerella			Escherichia			Shigella			Hoylesella			p-value
	Found	Not Found	p-value	Found	Not Found	p-value	Found	Not Found	p-value	Found	Not Found	p-value	Found	Not Found		
	n (%)	n (%)		n (%)	n (%)		n (%)	n (%)		n (%)	n (%)		n (%)	n (%)	n (%)	
Age	5 (41.7)	16 (33.3)	0,737	5 (45.5)	16 (32.7)	0,421	3 (37.5)	18 (34.6)	1,000	2 (40.0)	19 (34.5)	1,000	3 (75.0)	18 (32.1)	0,119	
> 60 years	7 (58.3)	32 (66.7)		6 (54.5)	33 (67.3)		5 (62.5)	34 (65.4)		3 (60.0)	36 (65.5)		1 (25.0)	38 (67.9)		
BMI																
Underweight /Normal	4 (33.3)	20 (41.7)	0,746	4 (36.4)	20 (40.8)	0,785	4 (50.0)	20 (38.5)	0,702	3 (60.0)	21 (38.2)	0,380	1 (25.0)	23 (41.1)	0,643	
Overweight/Obesity	8 (66.7)	28 (58.3)		7 (63.6)	29 (59.2)		4 (50.0)	32 (61.5)		2 (40.0)	34 (61.8)		3 (75.0)	33 (58.9)		
Parity																
Primipara	6 (50.0)	28 (58.3)	0,602	5 (45.5)	29 (59.2)	0,406	4 (50.0)	30 (57.7)	0,717	2 (40.0)	32 (58.2)	0,644	4 (100.0)	30 (53.6)	0,126	
/Multipara	6 (50.0)	20 (41.7)		6 (54.5)	20 (40.8)		4 (50.0)	22 (42.3)		3 (60.0)	23 (41.8)		0 (0.0)	26 (46.4)		
Grande multipara																
Duration of menopause	1 (8.3)	3 (6.3)	0,967	1 (9.1)	3 (6.1)	0,382	0 (0.0)	4 (7.7)	0,443	0 (0.0)	4 (7.3)	0,618	0 (0.0)	4 (7.1)	0,414	
Not yet	1 (8.3)	4 (8.3)		2 (18.2)	3 (6.1)		0 (0.0)	5 (9.6)		0 (0.0)	5 (9.1)		1 (25.0)	4 (7.1)		
< 5 years	10 (83.4)	41 (85.4)		8 (72.7)	43 (87.8)		8 (100.0)	43 (82.7)		5 (100.0)	46 (83.6)		3 (75.0)	48 (85.7)		
≥ 5 years																
Delivery	11 (91.7)	44 (91.7)	1,000	11 (100)	44 (89.8)	0,573	8 (100.0)	47 (90.4)	1,000	5 (100.0)	50 (90.9)	1,000	4 (100.0)	51 (91.1)	1,000	
Vaginal and cesarean section	1 (8.3)	4 (8.3)		0 (0.0)	5 (10.2)		0 (0.0)	5 (9.6)		0 (0.0)	5 (9.1)		0 (0.0)	5 (8.9)		
Duration of POP	7 (58.3)	32 (66.7)	0,737	7 (63.6)	32 (65.3)	1,000	5 (62.5)	34 (65.4)	1,000	3 (60.0)	36 (65.5)	1,000	2 (50.0)	37 (66.1)	0,606	
< 5 years	5 (41.7)	16 (33.3)		4 (36.4)	17 (34.7)		3 (37.5)	18 (34.6)		2 (40.0)	19 (34.5)		2 (50.0)	19 (33.9)		
History of Pessary	2 (16.7)	9 (18.8)	1,000	2 (18.2)	9 (18.4)	1,000	0 (0.0)	11 (21.2)	0,330	0 (0.0)	11 (20.0)	0,330	0 (0.0)	11 (19.6)	1,000	
Yes	10 (83.3)	39 (81.3)		9 (81.8)	40 (81.6)		8 (100.0)	41 (78.8)		5 (100.0)	44 (80.0)		4 (100.0)	45 (80.4)		
No																
Use of genital cleansers	8 (66.7)	39 (81.3)	0,271	9 (81.8)	38 (77.6)	1,000	6 (75.0)	41 (78.8)	1,000	3 (75.0)	44 (78.6)	1,000	3 (75.0)	44 (78.6)	1,000	
Yes	4 (33.3)	9 (18.8)		2 (18.2)	11 (22.4)		2 (25.0)	11 (21.2)		1 (25.0)	12 (21.4)		1 (25.0)	12 (21.4)		

Uji chi-square, bermakna (p<0.05)

differences observed in women aged 41-60 years. Significant alterations in both uterine and vaginal microbiota were observed in women aged ≥ 40 years, consistent with marked changes in alpha and beta diversity, indicating substantial microbial disruption during these age ranges.¹⁷

In the present study, most POP patients had experienced menopause for ≥ 5 years, whereas the majority of healthy controls were premenopausal or had menopausal duration of less than 5 years. This finding is in agreement with studies reported a significant association between POP and menopausal duration exceeding five years.¹⁸ This relationship may be explained by estrogen deficiency associated with prolonged menopause and advanced age, which contributes to pelvic floor dysfunction due to the presence of estrogen receptors in pelvic floor support structures. Further noted that longer menopausal duration and extended hypoestrogenic periods predispose older women to prolapse.¹⁹

Pelvic organs and their muscular and connective tissue components are estrogen-responsive, and aging reduces the collagen I/III ratio. Increased expression of type III collagen and matrix metalloproteinase-9 (MMP-9) has been implicated in the pathogenesis of POP.²⁰ The vaginal microbial profile among POP patients in this study consisted predominantly of *Prevotella* (20.0%), *Gardnerella* (18.3%), *Lactobacillus* (15.0%), *Atopobium* (15.0%), *Escherichia* (13.3%), *Shigella* (8.3%), *Bacillota* (6.7%), *Hoylella* (6.7%), *Pseudomonas* (5.0%), and *Phocaeicola* (5.0%). In contrast, non-POP women exhibited a vaginal microbiota dominated by *Lactobacillus* (83.3%), followed by *Streptococcus* (43.3%), *Dialister* (35.0%), *Legionella* (25.0%), *Fusobacteriota* (16.7%), and *Campylobacter* (15.0%). Both groups shared the presence of the phylum *Bacillota*; however, its prevalence was markedly higher among non-POP women. Nearly all non-POP participants harbored *Bacillota*, which is consistent with previous evidence

that this phylum dominates the vaginal environment in healthy women.²¹

These findings demonstrate a markedly lower prevalence of *Lactobacillus* in POP patients compared with non-POP women. The presence of *Lactobacillus* is a hallmark of vaginal eubiosis and is associated with a healthy vaginal environment.²² *Lactobacillus* maintains vaginal acidity through lactic acid production, thereby inhibiting the growth of potential pathogens such as *Neisseria gonorrhoeae*, *Gardnerella vaginalis*, and *Chlamydia trachomatis*. Additionally, *Lactobacillus* plays a critical role in preserving vaginal mucosal integrity and immune defense by supporting innate immune cells and limiting pathogen invasion.²³

The reduction of *Lactobacillus* and the presence of diverse pathogenic genera in POP patients indicate vaginal dysbiosis. Predominant pathogens identified included *Atopobium*, *Prevotella*, *Gardnerella*, *Escherichia*, *Bacteroides*, and *Shigella*. This pattern is characteristic of vaginal dysbiosis, which is defined by decreased *Lactobacillus* abundance and increased anaerobic bacteria.²⁴ Similar findings were reported who observed significantly higher relative abundance of *Bacteroidota* and *Fusobacteriota* in women with pelvic floor dysfunction.²⁵ The observed microbial profile in POP patients corresponds to community state type IV (CST-IV), characterized by low *Lactobacillus* dominance and increased anaerobic diversity, a pattern previously reported as the most common vaginal community type in women with POP.²⁶ Notably, *Prevotella* and *Gardnerella* were detected in 20.0% and 18.3% of POP patients, respectively. Shifts toward CST-IV, marked by increased anaerobic diversity and dominance of *Prevotella* and *Gardnerella*, are commonly observed during menopause.²⁷

The presence of anaerobic bacteria such as *Prevotella* and *Atopobium* is further influenced by sexual activity, particularly unprotected intercourse, which has been shown to significantly alter vaginal

microbiota composition.²⁸ These findings indicate that POP patients in this study experienced vaginal dysbiosis, likely associated with bacterial vaginosis. This observation aligns with previous Indonesian data reporting bacterial vaginosis in 76.7% of POP patients, particularly among women aged over 60 years and those who were postmenopausal.²⁹ The underlying mechanism linking vaginal dysbiosis and POP may involve inflammation driven by estrogen deficiency, reduced Lactobacillus-mediated protection, increased oxidative stress, and upregulation of MMP expression, leading to degradation of pelvic floor connective tissue and progression of prolapse.³⁰

Conclusion

In conclusion, age >60 years and a menopausal duration of ≥5 years were associated with an increased risk of pelvic organ prolapse (POP). Both POP and non-POP women harbored the phylum Bacillota, although its relative abundance was higher in the non-POP group, while women with POP showed greater vaginal microbial diversity and a predominance of non-Bacillota phyla, indicating vaginal dysbiosis. Women aged >60 years were more frequently colonized by Atopobium and Fannyhessea vaginae, whereas Prevotella predominated among women with a menopausal duration of ≥5 years. Although these associations were not statistically significant, the findings suggest an age and menopause related shift toward vaginal anaerobic dysbiosis in women with pelvic organ prolapse. These findings suggest that advanced age and prolonged menopause contribute to reduced protective Lactobacillus levels, increased pathogenic colonization, and vaginal dysbiosis, which may play an important role in the development of pelvic organ prolapse.

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Disclosure

The authors report no conflicts of interest in this work.

Authors Contribution

WM drafted the manuscript. ECJ and DL contributed to data interpretation and manuscript revision. ECJ, DL provided critical input on clinical methodology and supervision ECJ, NA, SNA and FH contributed to laboratory analysis, data validation, and visualization. All authors reviewed and approved the final version of the manuscript and agreed to be accountable for all aspects of the work

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