



## Genital Mycoplasmas and Genital Disorders Among Women of Childbearing Age in Franceville, South-Est Gabon

Kelly Hornelia Mbombe Moghoa<sup>\*,1,2</sup>, Richard Onanga<sup>1</sup>, Romeo Wenceslas Lendamba<sup>1,3</sup>, Michelle Bignoumba<sup>1,2</sup>, Jean Ulrich Muandze-Nzambe<sup>1</sup>, Yann Mouanga-Ndzime<sup>1</sup>, Saidou Mahmoudou<sup>3</sup>, Romuald Be Mba<sup>3</sup>, Klara Pecmann<sup>3</sup>, Amahani Gafou<sup>1</sup>, Roland Fabrice Kassa Kassa<sup>1</sup>, Cyrille Bisseye<sup>4</sup>

### Abstract

**Objective:** This study aimed to evaluate the prevalence of *Ureaplasma* spp. and *Mycoplasma hominis* and their association with genital disorders (bacterial vaginosis, aerobic vaginitis, and vulvovaginal candidiasis).

**Methods:** The IST2 kit was used to identify genital mycoplasmas. The Nugent score and microscopic observation of neutrophils and bacteria were employed to diagnose bacterial vaginosis and aerobic vaginitis. Statistical analysis was performed using the chi-square or Fisher's exact test.

**Results:** The prevalence of *M. hominis* and *Ureaplasma* spp. was estimated at 2% and 41%, respectively, with a *Ureaplasma* spp./*M. hominis* coinfection rate of 32%. The prevalence of bacterial vaginosis, vulvovaginal candidiasis, and aerobic vaginitis was found to be 68%, 35%, and 54%, respectively. *Group B Streptococcus* was not associated with *M. hominis* ( $P=0.006$ ,  $OR=0.41$ ,  $95\% CI=0.22-0.77$ ) or *Ureaplasma* spp./*M. hominis* ( $P=0.002$ ,  $OR=0.30$ ,  $95\% CI=0.14-0.63$ ).

**Conclusion:** This study reports a high prevalence of genital mycoplasmas. Although vulvovaginal candidiasis, aerobic vaginitis, and bacterial vaginosis were frequently encountered, their association with genital mycoplasmas was not statistically significant.

**Keywords:** BV; AV; VVC; *M. hominis*; *Ureaplasma* spp.; Gabon

### Background

Genital disorders could be seen as results of a disturbance of the vaginal ecology with bacterial vaginosis (BV) being found in more than 30% of women of childbearing age worldwide. BV is characterized by an imbalance in the vaginal bacterial flora, involving the reduction of lactobacilli and proliferation of bacteria such as *Gardnerella vaginalis* (*G. vaginalis*), *Mobililincus* spp., and resident anaerobic vaginal bacteria [1][2]. Genital mycoplasmas, specifically *Mycoplasma hominis* (*M. hominis*) and *Ureaplasma* spp., are opportunistic bacteria that are also part of the commensal vaginal flora, with the potential to increase during an imbalance in the vaginal ecosystem. These bacteria are commonly found in the genital tract of both symptomatic and asymptomatic women [3]. Several studies have suggested their association with other microorganisms in lower genital disorders, including bacterial vaginosis [4, 5]. Those genital mycoplasmas can be diagnosed in conjunction with bacterial vaginosis (BV), aerobic vaginitis (AV), and vulvovaginal candidiasis (VVC).

A previous study by Kouegnigan Rerambiah (2015) found a 68.5% rate of mycoplasma infections among Gabonese women suspected of having

### Affiliation:

<sup>1</sup>Medical Research and Analysis Unit (URAM): Bacteriology Laboratory, Interdisciplinary Center for Medical Research of Franceville (CIRMF), BP 769, Franceville, Gabon.

<sup>2</sup>Doctoral School of Fundamental and Applied Sciences, University of Central Africa Regional Doctoral School in Tropical Infectiology (ECODRAC), University of Science and Technology of Masuku (USTM), BP 876, Franceville, Gabon.

<sup>3</sup>Centre de Recherches Médicales de Lambaréné (CERMEL), Department of Clinical Operations, BP 242, Lambaréné, Gabon.

<sup>4</sup>Department of Biology, Laboratory of Molecular and Cellular Biology (Lab Head) University of Science and Technology of Masuku (USTM) BP 943, Franceville, Gabon.

### \*Corresponding author:

Kelly Hornelia Mbombe Moghoa, Medical Research and Analysis Unit (URAM): Bacteriology Laboratory, Interdisciplinary Center for Medical Research of Franceville (CIRMF), BP 769, Franceville, Gabon.

**Email:** moghoakelly@yahoo.com

**Citation:** Kelly Hornelia Mbombe Moghoa, Richard Onanga, Romeo Wenceslas Lendamba, Michelle Bignoumba, Jean Ulrich Muandze-Nzambe, Yann Mouanga-Ndzime, Saidou Mahmoudou, Romuald Be Mba, Klara Pecmann, Amahani Gafou, Roland Fabrice Kassa Kassa, Cyrille Bisseye. Genital Mycoplasmas and Genital Disorders Among Women of Childbearing Age in Franceville, South-Est Gabon. Archives of Microbiology and Immunology. 7 (2023): 468-474

**Received:** October 24, 2023

**Accepted:** October 31, 2023

**Published:** December 1, 2023

urogenital disorders [6]. A Gabonese study published in 2022 indicated a polymicrobial aetiology of vaginitis in southeastern Gabon that was linked to several associations and the most frequently identified vaginal infections were BV, AV, and VVC [7]. Also, a high prevalence of BV with genital mycoplasmas has been found in this area [7], [8]. Molecular analyses have been applied in multiple studies to better understand vaginal imbalances [4], [9]. Meanwhile, the Nugent score is the gold standard in low-resource country clinical laboratories for diagnosing bacterial vaginosis. This technique is sensitive and specific for evaluating BV, the most common vaginal disorder [2]. Thus, the present study aimed to evaluate the prevalence of *Ureaplasma* spp., *M. hominis*, and other genital disorders such as AV, BV, and VVC and assess the association of genital mycoplasma with these disorders.

## Materials and Methods

### Study population criteria and sample collection

Between January 2019 and August 2020, 350 sexually active women, referred to the laboratory for a diagnostic genital swab and residing in Franceville and its surrounding areas, were encouraged to participate in the study. Franceville is located in the administrative centre of the Haut-Ogooué province in southeastern Gabon, the second most populous province in Gabon. Eligible participants should not have had sexual intercourse during the two to three days preceding sampling, should not have performed private hygiene on the day of selection, and should not have been under antibiotics or genital ovules medication. Social and demographic data were collected from the participants using a semi-structured pretested questionnaire.

### Ethical considerations

Participants were individually recruited after signing informed consent forms, and confidentiality was ensured through assigned participant numbers. The research license for this study was obtained from the Scientific Commission on Research Authorizations of the National Centre of Scientific and Technological Research (CENAREST) (permit 7 no. AR0033/17/MESRSFC/CENAREST/CG/CST/CSAR, dated 4 July 2017). This study was conducted following the Declaration of Helsinki.

### Sampling

Each outpatient was examined for inflammation or ulceration and to characterize vaginal discharge (colour, consistency, pH, and odour). Vaginal samples were then collected by trained and experienced nurses according to standard operating procedures. In non-pregnant women, three swabs were used: one vaginal swab for diagnosing BV, another from the endocervix for *M. hominis* and *Ureaplasma* spp. research, and the remaining one from the posterior

vaginal fornix for inflammation of the vaginal mucosa, VVC, and AV. The swab for *M. hominis* and *Ureaplasma* spp. search was taken from the posterior vaginal fornix in pregnant women. To prevent contamination, all specimens were processed within an hour of sampling.

### *M. hominis* and *Ureaplasma* spp. detection, quantification, and identification

The identification and *quantification* of *Ureaplasma* spp. and *Mycoplasma hominis* were determined using a commercial *Mycoplasma* IST2 kit (bioMérieux, Marcy-l'Etoile, France) as instructed by the manufacturer.

### Diagnosis of vaginal dysbiosis

**Diagnosis of BV:** Following the sampling step, all vaginal swabs underwent a microscopic examination of Gram-stained smears to establish a Nugent score after assessing the presence of three bacterial morphotypes (*Lactobacillus* spp., *Gardnerella* spp., *Mobiluncus* spp.), and clue cells. A Nugent score higher or equal to 7 was considered positive for BV [2].

**Diagnosis of AV and associated germs:** Diagnosis of aerobic vaginitis (AV) was performed based on microscopy using the refined Schröder's classification done in 2005 by Donders et al. [10]. The other swabs taken from the posterior vaginal fornix were used for isolating, based on colony morphology, the other germs by immediately cultivating them on several media. Only growth above 1+ (growth over more than half of the incubation plate) was included in the analysis. BCP dextrose agar medium (BIOKAR Diagnostics, France) was used to culture fast-growing Gram-negative bacilli, while CNA + 5% Sheep Blood agar medium (bioMérieux, France) was used for Gram-positive bacteria whereas the specific isolation of *Neisseria* spp. was made using Chocolate + PolyViteX VCAT3 agar medium. Rapid and accurate biochemical identification of all isolated germs was performed using the Vitek 2 automated system (bioMérieux, France).

**VVC:** The evaluation of vulvovaginal candidiasis was made using Sabouraud Chloramphenicol (SAB-CHL) agar medium (bioMérieux, France) for presumptive *Candida* spp. isolation. The isolated colonies were then identified using the Vitek 2 automated system (bioMérieux, France).

**Trichomoniasis research:** *Trichomonas vaginalis* (TV) was identified microscopically (under an optical microscope's immersion oil objective) based on its morphological characteristics on Giemsa-stained smears of vaginal swabs [11].

### Statistical Considerations

Data were analyzed using R version 4.2.2 software. Prevalences and proportions were used to summarize and present the data. The association between independent and

dependent variables was tested using chi-square or Fisher's exact test, as appropriate, and the odds ratio (OR) and its corresponding 95% CI were reported. Results were considered significant for  $p < 0.05$ . Furthermore, logistic regression analysis was also performed to correlate the presence of other lower genital disorders with genital mycoplasmas.

## Results

Three hundred fifty (350) women living in Franceville and surrounding areas were enrolled in this study. The study population age was ranging from 13 to 55 years, with an average age of 32.2 years.

### Prevalence of genital disorders:

The results of this study show a prevalence of *M. hominis* and *Ureaplasma* spp. of 74.86%. In our study population, *Ureaplasma* spp. was diagnosed at a rate of 42%, followed by *Ureaplasma* spp./ *M. hominis* co-infection (31%), and *M. hominis* alone (2%). The prevalence of other lower genital disorders was as follows: 68% for BV, 35% for VVC, 54% for AV, and 0.3% for *Trichomonas vaginalis* and *Neisseria gonorrhoea* infections.

### Distribution of *M. hominis* and *Ureaplasma* spp. among women:

Seventy-five per cent (262/350) of women tested positive for genital mycoplasmas at different rates: *M. hominis* (2%), *Ureaplasma* spp. (41%), and the co-infection of

*Ureaplasma* spp./ *M. hominis* (32%). Women whose ages ranged between 21 and 40 years old were not only the most numerous (79%) but also the most affected by genital mycoplasmas with a prevalence of 77.9% for *Ureaplasma* spp., 89.7% for *M. hominis*, and 79.4% for *Ureaplasma* spp./ *M. hominis*. Approximately 66.9% of the women did not use contraceptives and 92.3% experienced symptoms. Most of them (92%) were non-pregnant, 42.0% of patients were in an unmarried cohabiting relationship and 51.5% reported having a job. Although the prevalence of genital mycoplasmas was relatively high in this study, it was found that statistically, genital mycoplasmas were associated with age, contraceptive use, and symptoms ( $P < 0.05$ ) (Table 1).

### Association of *M. hominis* and *Ureaplasma* spp. with VVC, BV, and AV:

BV was observed to be the most prevalent genital disorder associated with *M. hominis* and *Ureaplasma* spp. in this study i.e. 25%. The rates of association of BV with *Ureaplasma* spp., *M. hominis*, and the coinfection *Ureaplasma* spp./*M. hominis* were respectively 23%, 57% and 31%. This was followed by mixed infection BV/AV (21%) with 25%, 29%, and 31% for *Ureaplasma* spp., *M. hominis*, and *Ureaplasma* spp./*M. hominis*, respectively. Univariable logistic regression analyses were performed to show an association between *M. hominis* and *Ureaplasma* spp., BV, VVC, and AV (Table 2). BV ( $P=0.005$ ,  $OR=0.12$ ,  $95\% CI=0.02-0.43$ ), AV ( $P=0.041$ ,

**Table 1:** Overview of Socio-demographic characteristics of outpatients according to genital mycoplasmas

Variable	N	<i>Ureaplasma</i> spp.			<i>Mycoplasma hominis</i>			<i>Ureaplasma</i> spp./ <i>Mycoplasma hominis</i>			
		Negative	Positive	p-value	Negative	Positive	p-value	Negative	Positive	p-value	
All	350	203	147		343	7		242	108		
Age Group	<= 20	22 (6.4%)	11 (7.6%)	0.725	22 (6.5%)	0 (0%)	0.782	12 (5.0%)	10 (9.3%)	0.192	
	21-40	274 (79.2%)	161 (80.1%)		268 (79.1%)	6 (85.7%)		189 (79.1%)	85 (79.4%)		
	> 40	50 (14.5%)	29 (14.4%)		21 (14.5%)	49 (14.5%)		1 (14.3%)	38 (15.9%)		12 (11.2%)
Contraception	With	116 (33.1%)	64 (31.5%)	0.522	113 (32.9%)	3 (42.9%)	0.884	74 (30.6%)	42 (38.9%)	0.161	
	without	234 (66.9%)	139 (68.5%)		95 (64.6%)	230 (67.1%)		4 (57.1%)	168 (69.4%)		66 (61.1%)
Gestation	No	322 (92.0%)	190 (93.6%)	0.274	316 (92.1%)	6 (85.7%)	1	219 (90.5%)	103 (95.4%)	0.18	
	Yes	28 (8.0%)	13 (6.4%)		15 (10.2%)	27 (7.9%)		1 (14.3%)	23 (9.5%)		5 (4.6%)
Marital status	Concubine	147 (42.0%)	92 (45.3%)	0.252	142 (41.4%)	5 (71.4%)	0.276	97 (40.1%)	50 (46.3%)	0.191	
	Married	83 (23.7%)	48 (23.6%)		35 (23.8%)	82 (23.9%)		1 (14.3%)	64 (26.4%)		19 (17.6%)
	single	120 (34.3%)	63 (31.0%)		57 (38.8%)	119 (34.7%)		1 (14.3%)	81 (33.5%)		39 (36.1%)
Occupations	Student	30 (9.9%)	22 (12.2%)	0.25	28 (9.5%)	2 (28.6%)	0.079	23 (10.7%)	7 (8.0%)	0.691	
	With	156 (51.5%)	89 (49.2%)		67 (54.9%)	155 (52.4%)		1 (14.3%)	108 (50.2%)		48 (54.5%)
	without	117 (38.6%)	70 (38.7%)		47 (38.5%)	113 (38.2%)		4 (57.1%)	84 (39.1%)		33 (37.5%)
Symptoms	With	323 (92.3%)	183 (90.1%)	0.119	316 (92.1%)	7 (100.0%)	0.954	223 (92.1%)	100 (92.6%)	1	
	without	27 (7.7%)	20 (9.9%)		7 (4.8%)	27 (7.9%)			19 (7.9%)		8 (7.4%)

OR=0.19, 95% CI=0.03–0.79), BV/VVC (P=0.018, OR=0.14, 95% CI=0.02–0.61), BV/AV (P=0.021, OR=0.17, 95% CI=0.03–0.63), and BV/VVC/AV (P=0.003, OR=0.1, 95% CI=0.01–0.37) showed a reverse odds of having *Ureaplasma* spp./*M. hominis* in women (Table 2).

### Association of *M. hominis* and *Ureaplasma* spp. with different germs:

The proportions of women with Group B *Streptococcus* showed reverse odds of having *M. hominis* (P=0.006, OR=0.41, 95% CI=0.22–0.77) and *Ureaplasma* spp./*M. hominis* (P=0.002, OR=0.30, 95% CI=0.14–0.63) (Table 3).

### Discussion

Vaginal disorders are considered the most common gynaecological problem in women of reproductive age and the most common cause of gynaecological medical care. The prevalence of *M. hominis* and *Ureaplasma* spp. (74.86%) found in this study matches that previously reported in Gabon, Cameroon, and China in pregnant women [6][12][13]. In our study population, *Ureaplasma* spp. was the most frequently diagnosed microorganism (42%), followed by *M. hominis*/*Ureaplasma* spp. co-infection (31%) and *M. hominis* (2%). Similar findings have been observed in Libreville, the capital of Gabon [6] and China [14]. In a healthy vagina, *Ureaplasma*

**Table 2:** Association of genital mycoplasmas to other lower infection VVC, BV, and AV

Variable	Ureaplasma spp				Mycoplasma hominis				Ureaplasma spp_Mycoplasma hominis			
	Positive	OR	95% CI	p.value	Positive	OR	95% CI	p.value	Positive	OR	95% CI	p.value
No Infection	14 (9.5%)	1			1 (14%)	1			2 (1.9%)	1		
BV	34 (23%)	1.43	0.61, 3.35	0.41	4 (57%)	0.73	0.04, 5.21	0.784	33 (31%)	0.12	0.02, 0.43	<b>0.005</b>
VVC	10 (6.8%)	0.56	0.15, 1.92	0.362	0	NA	NA	NA	2 (1.9%)	0.52	0.06, 4.70	0.533
AV	12 (8.2%)	2.1	0.78, 5.79	0.144	0	NA	NA	NA	11 (10%)	0.19	0.03, 0.79	<b>0.041</b>
BV & VVC	13 (8.8%)	1.15	0.41, 3.25	0.792	0	NA	NA	NA	10 (9.3%)	0.14	0.02, 0.61	<b>0.018</b>
BV & AV	37 (25%)	0.96	0.40, 2.27	0.923	2 (29%)	1.3	0.06, 14.1	0.831	23 (21%)	0.17	0.03, 0.63	<b>0.021</b>
VVC & AV	14 (9.5%)	0.93	0.33, 2.65	0.896	0	NA	NA	NA	6 (5.6%)	0.27	0.04, 1.31	0.132
BV, VVC & AV	13 (8.8%)	2.51	0.96, 6.73	0.062	0	NA	NA	NA	21 (19%)	0.1	0.01, 0.37	<b>0.003</b>

**Table 3:** Distribution of pathogens according to genital mycoplasmas (N = 396)

	Ureaplasma spp				Mycoplasma hominis				Ureaplasma spp_/Mycoplasma hominis			
	Positive (n= 288)	OR	95% CI	p.value	Positive (n= 131)	OR	95% CI	p.value	Positive (n= 122)	OR	95% CI	p.value
<b>Aerobic Vaginitis</b>												
No Infection	188 (65%)	1			84 (64%)	1			76 (62%)	1		
<i>Enterococcus faecalis</i>	7 (2.4%)	1.97	0.56, 6.42	0.264	2 (1.5%)	2.25	0.57, 14.9	0.305	2 (1.6%)	1.95	0.49, 12.9	0.398
<i>Enterococcus spp</i>	5 (1.7%)	2.19	0.53, 8.55	0.253	3 (2.3%)	0.98	0.25, 4.74	0.974	3 (2.5%)	0.83	0.21, 4.04	0.799
<i>Escherichia coli</i>	11 (3.8%)	0.25	0.01, 1.30	0.182	1 (0.8%)	5.42	1.03, 100	0.109	1 (0.8%)	4.65	0.88, 85.8	0.145
<i>Group B Streptococcus</i>	39 (14%)	0.77	0.35, 1.55	0.477	26 (20%)	0.41	0.22, 0.77	<b>0.006</b>	26 (21%)	0.36	0.19, 0.68	<b>0.002</b>
<i>Klebsiella pneumonia ssp</i>	2 (0.7%)	2.69	0.32, 22.9	0.328	2 (1.5%)	0.53	0.06, 4.49	0.529	1 (0.8%)	1.34	0.17, 27.4	0.802
<i>Kocuria kristinae</i>	11 (3.8%)	0.99	0.27, 3.01	0.989	2 (1.5%)	3.1	0.83, 20.1	0.143	2 (1.6%)	2.66	0.71, 17.3	0.205
<i>Neisseria gonorrhoeae</i>	1 (0.3%)	2.81	0.11, 72.0	0.469	0	NA	NA	NA	0	NA	NA	NA
<i>Sphingomonas paucimobilis</i>	5 (1.7%)	1.04	0.15, 4.96	0.966	2 (1.5%)	1.26	0.26, 8.92	0.789	2 (1.6%)	1.1	0.23, 7.80	0.913
<i>Staphylococcus aureus</i>	18 (6.2%)	0.76	0.24, 2.00	0.601	9 (6.9%)	0.69	0.29, 1.74	0.421	9 (7.4%)	0.61	0.25, 1.52	0.27
<i>Trichomonas vaginalis</i>	1 (0.3%)	5.54	0.52, 120	0.166	0	NA	NA	NA	0	NA	NA	NA
<b>Vulvovaginal Candidiasis</b>												
No Infection	186 (65%)	1			89 (68%)	1			81 (66%)	1		
<i>Candida albicans</i>	67 (23%)	0.92	0.51, 1.61	0.774	26 (20%)	1.46	0.85, 2.58	0.179	25 (20%)	1.4	0.80, 2.49	0.247
<i>Candida spp</i>	35 (12%)	1.14	0.56, 2.23	0.712	16 (12%)	1.21	0.63, 2.40	0.582	16 (13%)	1.06	0.55, 2.12	0.861

**Citation:** Kelly Hornelia Mbombe Moghoa, Richard Onanga, Romeo Wenceslas Lendamba, Michelle Bignoumba, Jean Ulrich Muandze-Nzambe, Yann Mouanga-Ndzime, Saidou Mahmoudou, Romuald Be Mba, Klara Pecmann, Amahani Gafou, Roland Fabrice Kassa Kassa, Cyrille Bisseye. Genital Mycoplasmas and Genital Disorders Among Women of Childbearing Age in Franceville, South-Est Gabon. Archives of Microbiology and Immunology. 7 (2023): 468-475.

spp. can be found at rates ranging from 30 to 50% compared to *M. hominis* mostly present at 10% [15], thus probably explaining the higher prevalence of *Ureaplasma* spp. [16].

The most prevalent form of vaginal disorders in our study was BV (68%), with similar results reported in Yemen and Ethiopia [17][18], but with a lower rate in the Rummyantseva study [5]. Gabonese women can use vaginal ovules without medical recommendations and this could lead to BV relapses. Additionally, the high rates of BV observed in this study may be indicative of therapeutic failures, potentially resulting from a lack of patient adherence to treatment or the presence of bacteria that are naturally resistant to metronidazole (e.g., *Atopobium vaginae*) and capable of producing biofilms (e.g., *Gardnerella vaginalis* and *Atopobium vaginae*) [19], [20]. While VVC and AV had reasonably high prevalence rates (respectively 35% and 54%), similar results were found in previous results conducted in Senegal, Gabon and Bosnia [21][22] [23]. Contrasting with our study, Yemen reported a prevalence of 6.6% for VVC [17] while Salinas observed that AV was the most prevalent form of vaginal infection in Ecuador, followed by BV and VVC [9]. In contrast, *Trichomonas vaginalis* and *Neisseria gonorrhoea* infections were the least frequent vaginal infections, as reported by Maha et al. (2019) and Angelica et al. (2017) [17][24].

The age range of the women in this study varied from 13 to 55 years, with an average age of 32.2 years. In our study, women belonging to the age group between 21 and 40 years old were the most affected by *M. hominis* and *Ureaplasma* spp. (79.2%) with a prevalence of 77.9% for *Ureaplasma* spp., 85.7% for *M. hominis*, and 79.4% for *Ureaplasma* spp./*M. hominis* co-infection. Similar results have been obtained in other studies on sex workers [25] and pregnant women [13], while lower results were observed in Iranian studies [26]. Among the 67% of women not using condoms, 65% had *Ureaplasma* spp., 57% had *M. hominis*, and 61% had both associated. This indicates that the less we have protected sex, the higher is the risk of contracting genital mycoplasmas. In 2005, Chinese sex workers were shown to reduce the risk of urogenital diseases, particularly mycoplasma infections, by using condoms [25].

Most women in this study (92%) were experiencing vaginal discomfort as was reported in a study performed in Korea [27]. The prevalence of vaginal discomfort in the presence of genital mycoplasmas was 95% for *Ureaplasma* spp., 100% for *M. hominis*, and 93% for *Ureaplasma* spp./*M. hominis*, which is not in line with results found in Poland [28]. Marovt et al. found that women co-infected with *U. urealyticum* and *U. parvum* had significantly higher odds of experiencing symptoms than women with *U. urealyticum* alone [29]. Plummer showed that only *M. hominis* was associated with symptoms/signs, which were manifestations

of BV. Significantly, *M. hominis* was not associated with symptoms/signs in women without BV [32]. Regardless of socio-demographic characteristics, the prevalence of genital mycoplasma was relatively high, and statistically, no significant difference was observed. This suggests that these factors do not influence the acquisition of genital mycoplasmas. Based on our data, the prevalence of all mycoplasmas was fairly higher in BV than in AV. This leads us to conclude that mycoplasmas may thrive in symbiotic relationships with BV-associated bacteria, mainly *Gardnerella vaginalis* and *Mobiluncus* spp. This result corroborates the study by Donders GG [5] in which the proportions of women with BV, AV, BV/VVC, BV/AV, and BV/VVC/AV that showed a reverse odd of *Ureaplasma* spp./*M. hominis* in women. Even though the study of Dandan et al., highlighted that *M. hominis* was more abundant when associated with TV instead of BV [31], several studies have shown that *M. hominis* is highly associated with BV [4][5]. It should be noted that the diagnosis of BV and genital mycoplasma is usually associated with an imbalance in vaginal flora. In our study, AV, VVC and the imbalance leading to BV may favour the proliferation of *Ureaplasma* spp./*M. hominis*. According to Rummyantseva et al., mycoplasmas require more than just a favourable pH in the environment to thrive; they may survive better in symbiotic relationships with anaerobic BV-associated bacteria, such as *Gardnerella vaginalis*, *Atopobium vaginae*, *Mobiluncus* spp., and other anaerobes [5]. According to Robinson, *M. hominis* loads significantly increase in cases of bacterial vaginosis (BV), and to a lesser extent, so do *Ureaplasma* species [32].

In genital complaints, *Group B Streptococcus*, *Escherichia coli*, *Staphylococcus aureus*, and *Trichomonas vaginalis* are frequently reported [33]. In this study, the association between *M. hominis*, and *Ureaplasma* spp./*M. hominis*, and *Group B Streptococcus* showed a relatively high prevalence of 20% and 21%. The proportions of women with *Group B Streptococcus* showed reverse odds of having *M. hominis*, *Ureaplasma* spp./*M. hominis* in women. *Streptococcus agalactiae* in symptomatic women with microscopic evidence of inflammation should be considered a causative agent of vaginitis. *Group B Streptococcus* or *Streptococcus agalactiae* can cause severe damage to the host, resulting in devastating clinical outcomes in pregnant women and newborns [34]. The association of *Group B Streptococcus* and two opportunistic mycoplasmas could be very dangerous because many studies have shown that *Mycoplasma hominis* and *Ureaplasma* spp. could lead to severe diseases and complications during or outside pregnancy [35][36]. It is essential to further investigate this association. *Escherichia coli* and *Enterococcus faecalis* mainly cause AV, while other causes haven't been found in this research, although Donders states that the cause of AV is *Escherichia coli*, *Enterococci*, *Staphylococcus* spp., and *Group B streptococcus* [37]. However, there are some

significant limitations in the present study since molecular methods were not applied and we did not look for sexually transmitted infections such as *Mycoplasma genitalium*, *Chlamydia trachomatis*, and *Neisseria gonorrhoea* by PCR to increase diagnostic sensitivity. In addition, the fact that they are not directly related to other lower infections may help to understand their implications for lower genital disorders.

## Conclusion

Although there is no statistically significant link between genital mycoplasmas (specifically *Ureaplasma spp.* and *M. hominis*) and genital disorders such as AV, BV, and VVC, their prevalence in this study is reasonably high in sexually active women living in Franceville and surrounding areas. Moreover, Group B *Streptococcus* carriage does not influence the occurrence of *M. hominis*, *Ureaplasma spp./M. hominis*. The association of genital mycoplasma with Group B *Streptococcus* should be further studied, especially in pregnant women where these microorganisms have a particular impact.

## Disclosure of Interests

All authors declared no conflict of interest.

## Ethical Considerations and Consent to Participate

The study protocol was reviewed and approved by the Ethics Committee of CIRMF. Female participants were recruited for the study after they or their legal representatives read and signed the informed consent form. To ensure confidentiality, participants were assigned lab numbers corresponding to their survey sheets and samples. Once the diagnosis was made, the results of all participants were transmitted with strict anonymity.

## Acknowledgements

This work is part of the first author's doctoral thesis. We want to thank URAM (Unité de Recherche et d'Analyses Médicales) of CIRMF (Centre Interdisciplinaire de Recherches Médicales de Franceville) for its involvement and financial support, and our staff in the Genomic Platform for its technical assistance and availability.

## Author Contributions

All authors contributed significantly to the work reported, whether in the conception, study design, execution, data acquisition, analysis, and interpretation. Kelly Hornelia MBOMBE MOGHOA, Richard ONANGA, Wenceslas LENDAMBA, Michelle BIGNOUMBA, Jean Ulrich MUANDZE-NZAMBE: conception, study design, execution, data acquisition, analysis, and interpretation. Klara PECMANN, Yann MOUANGA NDZIME, Romeo Wenceslas LENDAMBA, Cyrille BISSEYE: review

and translation; Romeo Wenceslas LENDAMBA, Kelly Hornelia MBOMBE MOGHOA Amahani GAFOU: analysis; Saidou MAHMOUDOU, Romuald BE MBA: statistical analysis; Roland Fabrice KASSA KASSA: Medical referral of patients.

## Funding Statement

The CIRMF funded this study.

## Consent for Publication

Not applicable.

## Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author upon reasonable request.

## List of abbreviations and acronyms

URAM: Unité de Recherches en Analyses Médicales.

CIRMF : Centre International de Recherches Médicales de Franceville.

USTM : Université des Sciences et Techniques de Masuku.

ECODRAC : Ecole Doctorale Régionale d'Afrique Centrale en Infectiologie Tropicale

USTM : Université des Sciences et Techniques de Masuku.

CERMEL : Centre de Recherches Médicales de Lambaréné.

VVC: vulvovaginal candidiasis.

BV bacterial vaginosis.

AV: Aerobic vaginitis.

*M. hominis*: *Mycoplasma hominis*.

*G. vaginalis*: *Gardnerella vaginalis*.

SAB-CH: Sabouraud chloramphenicol

MGG: May-Grünwald Giemsa

BCP: Bromocresol purple

VCA3: Chocolat PolyViteX VCAT3

CNA: Columbia NaladixicAcid

PVX: Chocolate agar + PolyViteX™

PTB: Preterm birth

## References

1. Sobel JD. "Bacterial vaginosis," Pathophysiology (2000): 349–356.

2. Nugent RP, Krohn MA, and Hillier SL. "Reliability of diagnosing bacterial vaginosis is improved by a standardized method of gram stain interpretation," *J. Clin. Microbiol* 29 (1991): 297–301.
3. Judlin P. "Genital mycoplasmas," *Gynecol. Obstet. Fertil* 31 (2003): 954–959.
4. Cox C, Watt AP, McKenna JP, and Coyle PV, "Mycoplasma hominis and Gardnerella vaginalis display a significant synergistic relationship in bacterial vaginosis," *Eur. J. Clin. Microbiol. Infect Dis* (2016).
5. Rummyantseva T, Khayrullina G, Guschin A, and Donders G. "Prevalence of Ureaplasma spp. and Mycoplasma hominis in healthy women and patients with flora alterations Tatiana," *Diagnostic Microbiol. Infect. Dis* (2018): 227–231.
6. Rerambiah LK, Ndong J, and Medzegue S. "Genital Mycoplasma infections and their resistance phenotypes in an African setting," (2015).
7. Bignoumba M, et al. "An overview of vaginal infections' etiologies in south-eastern Gabon," *Int. J. of Women's Heal* (2022).
8. Lendamba RW, et al. "Bacterial Vaginosis: Prevalence in Sexually Active Women Living in the City of Franceville (Gabon) and its Surroundings," 1–8.
9. Salinas AM, Osorio VG, Herrera DP, Vivanco JS, Trueba AF, and Machado A. "Vaginal microbiota evaluation and prevalence of key pathogens in Ecuadorian women: an epidemiologic analysis," *Sci. Rep* 0123456789 (2020): 1–18.
10. Donders GGG, Vereecken A, Bosmans E, Dekeersmaecker A, Salembier G, and Spitz B. "Aerobic vaginitis: Abnormal vaginal flora entity that is distinct from bacterial vaginosis B" 1279 (2005): 118–129.
11. Mason PR, Heather Super, and Fripp PJ. "Comparison of four techniques for the routine diagnosis of Trichomonas vaginalis infection," *J. Clin. Pathol* 29 (1976): 154–157.
12. Nkwabong E and Dingom MAN. "Acute Pelvic Inflammatory Disease in Cameroon : A Cross-Sectional Descriptive Study," 19 (2015): 87–91.
13. Lee MY, Kim MH, Lee WI, Kang SY, and La Jeon Y. "Prevalence and antibiotic susceptibility of mycoplasma hominis and ureaplasma urealyticum in pregnant women," *Yonsei Med* 57 (2016): 1271–1275.
14. Sha BE et al. "Female genital-tract HIV load correlates inversely with Lactobacillus species but positively with bacterial vaginosis and Mycoplasma hominis," *J. Infect. Dis* 191 (2005): 25–32.
15. Leli C, et al. "Prevalence of cervical colonization by Ureaplasma parvum, Ureaplasma urealyticum, Mycoplasma hominis, and Mycoplasma genitalium in childbearing age women by a commercially available multiplex real-time PCR: An Italian observational multicentre study" *J Microbiol Immunol Infect* (2017): 1–6.
16. Kim Y, Kim J, Lee KA. "Prevalence of sexually transmitted infections among healthy Korean women: Implications of multiplex PCR pathogen detection on antibiotic therapy" *J Infect Chemother* 20 (2014): 74–76.
17. Abdul-Aziz M, et al. "Bacterial vaginosis, vulvovaginal candidiasis, and trichomonal vaginitis among reproductive-aged women seeking primary healthcare in Sana 'a city, Yemen" 3 (2019): 1–10.
18. Mulu W, Yimer M, Zenebe Y, Abera B. "Common causes of vaginal infections and antibiotic susceptibility of aerobic bacterial isolates in women of reproductive age attending at Felegehiwot referral Hospital, Ethiopia: a cross-sectional study" (2015): 1–9.
19. De Backer E, et al. "Antibiotic susceptibility of Atopobium vaginae," *BMC Infect. Dis* 6 (2006): 1–6.
20. Livengood CH. "Bacterial vaginosis: an overview for 2009" *Rev. Obstet. Gynecol* 2 (2009): 28–37.
21. "Candidoses vulvo-vaginales au laboratoire de Parasitologie-Mycologie du Centre Hospitalier Universitaire de Fann, Dakar (Sénégal)" 5 (2017): 21–27.
22. Bignoumba M, et al., "Vulvovaginal Candidiasis among Symptomatic Women of Childbearing Age attended at a Medical Analysis Laboratory in Franceville, Gabon ScienceDirect Vulvovaginal candidiasis among symptomatic women of childbearing age attended at a Medical Analysis Laborat," *J. Mycol. Med* (2019).
23. Candidiasis V. "Clinical Characteristics of Aerobic Vaginitis and Its" 67 (2013): 428–430.
24. Miranda AE, Silveira MF, Gabriela A, Tenório T, Cristina I, De Lannoy L. "Original article Prevalence of Chlamydia trachomatis and Neisseria gonorrhoea and associated factors among women living with Human Immunodeficiency Virus in Brazil: a multicenter study," *Brazilian J. Infect. Dis* 21 (2017): 402–407.
25. Pingmin W, Yuepu P and Jiwen Z. "Prevalence survey on condom use and infection of urogenital mycoplasmas in female sex workers in China," *Contraception* 72 (2005): 217–220.
26. Seifoleslami M, Safari A and Khameneie MK. "Prevalence of Ureaplasma urealyticum and Mycoplasma hominis in high vaginal swab samples of infertile females," *Iran. Red Crescent Med. J* 17 (2015): 0–4.

27. Jang YS, Min JW, Kim YS. "Positive culture rate and antimicrobial susceptibilities of *Mycoplasma hominis* and *Ureaplasma urealyticum*" *Obstet. Gynecol. Sci* 62 (2019): 127–133.
28. Zdrodowska-Stefanow B, Kłosowska WM, Ostaszewska-Puchalska I, Bułhak-Kozioł V, Kotowicz B. "Ureaplasma urealyticum and *Mycoplasma hominis* infection in women with urogenital diseases.," *Adv. Med. Sci* 51 (2006): 250–253
29. Marovt M, Ke D, Kotar T, Kmet N and Miljkovi J. "Ureaplasma parvum and *Ureaplasma urealyticum* detected with the same frequency among women with and without symptoms of urogenital tract infection" (2015).
30. Plummer EL, et al. "Are *Mycoplasma hominis*, *Ureaplasma urealyticum* and *Ureaplasma parvum* Associated with Specific Genital Symptoms and Clinical Signs in Non-pregnant Women?," *Clin Infect Dis* 73 (2021): 659–668.
31. Yuan D, Chen WJ, Qin, Shen D, Qiao Y and Kong B. "Associations between bacterial vaginosis, candida vaginitis, trichomonas vaginalis, and vaginal pathogenic community in Chinese women," *Am J Transl Res* 13 (2021): 7148–7155.
32. Taylor-Robinson D. "Mollicutes in vaginal microbiology: *Mycoplasma hominis*, *Ureaplasma urealyticum*, *Ureaplasma parvum*, and *Mycoplasma genitalium*," *Res. Microbiol* 168 (2017): 875–881.
33. Donders GGG, Vereecken A and Bosmans E. "Definition of a type of abnormal vaginal flora that is distinct from bacterial vaginosis: aerobic vaginitis" 109 (2002): 34–43.
34. "Version of Record <https://www.sciencedirect.com/science/article/pii/S2468718919301102>" 0–21.
35. Donders GGG, Ruban K, Bellen G and Petricevic L. "Review article *Mycoplasma / Ureaplasma* infection in pregnancy: to screen or not to screen" 45 (2017): 505–515.
36. "Mycoplasmas in pregnancy" no. Table 1 (2010): 164–174.
37. Donders GGG. "Definition and classification of abnormal vaginal flora," *Best Pract. Res. Clin. Obstet. Gynaecol* 21 (2007): 355–373.