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Sexually transmitted infections and bacterial vaginosis in women of child-bearing age in Antananarivo, Madagascar: prevalence and risk factors from a cross-sectional study

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Abstract

Background Recent data on sexually transmitted infections (STIs) and bacterial vaginosis (BV) in the general population of Madagascar is scarce. Our objective is to estimate the *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (NG), *Trichomonas vaginalis* (TV), *Mycoplasma genitalium* (MG), and BV prevalence among women and to determine associated risk factors.

Methods We recruited women aged 18–45 years consulting at a hospital in Antananarivo in a cross-sectional study. We collected socio-demographic and behavioral data. Vaginal swabs were collected for each individual. STIs were detected by qPCR and BV using the Nugent score. We explored factors associated with having an STI using logistic regressions.

Results Of the 501 recruited women, 242 (48%) were married. Only one woman in three ever used condoms. The prevalence of CT was 11.8% (95% Confidence Interval: 9.0, 14.6), NG: 4.2% (2.4, 5.9), TV: 14.8% (11.7, 17.9), MG: 8.2% (5.8, 10.6), and BV: 39.6% (35.3, 43.9). Factors associated with an STI were having sex in exchange for favors (adjusted Odds Ratio (aOR): 4.25, 95%CI: 1.27, 16.6), BV (aOR: 2.14, 95%CI: 1.34, 3.47) or intermediate vaginal microbiota (aOR: 2.10, 95%CI: 1.19, 3.72), and being in a non-marital relationship compared to married (aOR: 1.86, 95%CI: 1.16, 2.98).

Conclusions To address the high prevalence of STIs, prevention programs should target women who are more likely to engage in sex in exchange for favors and empower them with negotiation skills on condom use. Given the elevated prevalence of dysbiosis and its association with STIs, it is crucial to prioritize research efforts toward a comprehensive understanding of this relationship.

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Keywords Sexually transmitted infections, Bacterial vaginosis, Epidemiology, Madagascar, Women

Background

Women in Africa are strongly affected by STIs, with more than 34 million prevalent cases of *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (NG), and *Trichomonas vaginalis* (TV) [1]. At the global level, women in Africa bear the highest prevalence of gonorrhea and trichomoniasis [1]. Only few studies have estimated the prevalence of *Mycoplasma genitalium* (MG) among women in Africa which was estimated to 3.20% in Tanzania [2] and 2.70% in Madagascar [3].

These STIs may result in urethritis, cervicitis, and pelvic inflammatory disease (PID). PID, in particular, can evolve into infertility, ectopic pregnancy, and chronic pelvic pain [4]. Infection during pregnancy is related to adverse pregnancy outcomes such as miscarriage or preterm delivery and in the newborn, low birth weight and neonatal infections such as pneumonia and conjunctivitis [5–7].

BV is a dysbiosis of the vaginal microbiota characterized by a replacement of *Lactobacilli* spp. by anaerobic bacteria such as *Gardnerella vaginalis*, *Fannyhessea vaginae*, and *Mycoplasma hominis* [8]. Global prevalence of BV ranges from 23 to 29% across regions and reaches 25% in sub-Saharan Africa [9]. Similarly to STIs, BV can lead to adverse health outcomes. During pregnancy, BV is associated with an increased risk of preterm birth, fetal death, and perinatal infections [8].

Multiple factors influence the risk of STI acquisition. In African countries, inconsistent condom use, multiple sexual partners, new relationships, lack of knowledge about STIs, and alcohol consumption have been reported to be associated with STIs [10–13].

Specifically in Madagascar, epidemiological data on STIs in the general population remains scarce. Although some studies on syphilis and HPV in the country have recently been published [14–16], to the best of our knowledge, the latest study on CT, NG, TV, MG, and BV was published in 2005 on a study conducted in 2002 [3], more than two decades ago. This study also explored risk factors and found that women and young age (15–24 years) were associated with STIs; however, it did not investigate social, behavioral, or biological factors.

This paper thus addresses the current lack of epidemiological data and understanding of factors associated with STIs in Madagascar. We provide recent estimates on the prevalence of CT, NG, TV, MG, and BV in women of child-bearing age who attended a hospital in Antananarivo. Additionally, we investigated the factors associated with the presence of at least one of the four tested STIs.

Methods

Study design

We conducted a cross-sectional study between March and November 2022 at the outpatient and family planning departments of the Gyneco-Obstetrics Befelatanana University Hospital (CHUGOB) in Antananarivo, the capital city of Madagascar. We chose this University teaching hospital since it serves as the primary referral center for gynecology and obstetrics. This study was a feasibility study for a larger trial on the evaluation of a rapid diagnostic test, the Genital InFlammation Test (GIFT), which aims to detect genital inflammation in women.

Study procedures

Our inclusion criteria were women aged 18 to 45 years, excluding pregnant women. Women with metrorrhagia or bleeding during examination or swabbing were also excluded because the presence of blood on the swab could interfere with pathogen detection. Menstruating women or women who had their last day of menstruation in the past three days were invited to return at a later date to the hospital for enrolment. Lastly, we excluded women who had taken antibiotics in the past 30 days as they would have cleared the pathogen but fragments of pathogens' DNA could still be detectable and would yield false positive cases.

Participants answered a questionnaire in Malagasy that was administered in a dedicated consultation room by either a nurse or a midwife, both of whom had clinical research experience. The questionnaire included questions on socio-demographic data, sexual history, and risk behavior information. Following the interview, a gynecologist or midwife inquired about symptoms, conducted a genital examination, and documented clinical signs.

Clinicians followed established procedures and prescribed treatment for suspected STIs. If the clinician did not initially suspect an STI but the laboratory (Experimental Bacteriology Unit (UBEX), Institut Pasteur in Madagascar (IPM)) later identified it on samples, participants were notified by phone to collect treatment at the hospital for free. Testing and enrollment did not involve any charges or incentives for the participants.

The gynecologist and the midwives collected three vaginal specimens with a speculum in place. One cotton and two Copan flocked® swabs (Copan, Brescia, Italy) were used to collect vaginal fluid from the lateral vaginal wall. The two flocked swabs were introduced simultaneously to detect CT, NG, TV, MG, and the fungal infection by *Candida albicans* (CA). The national STI/AIDS program offered HIV testing with capillary blood [17].

To test for BV, a cotton swab was used to prepare immediately after collection a vaginal smear for Nugent scoring. The vaginal smear was heat-fixated for 1 min on a heating plate at 40 °C and stored at room temperature until transport to UBEX at IPM.

Copan flocked swabs were kept at 2–8 °C until transport. The transport of swabs to UBEX was done on the same day using a cool box and icepacks, where they were stored at -20 °C until testing.

Laboratory methods

After thawing one flocked swab per participant, the vaginal secretions were eluted for at least 15 s in 1.2 ml of diluted phosphate-buffered saline (dPBS) (pH 7.4–1:9, PBS: saline). We extracted the DNA from an aliquot of 200 µL using the QUICK-DNA HMW magbead kit (Zymo Research, USA) according to the manufacturer's instructions. We used Presto CT/NG (Goffin Molecular Technologies, Beesd, The Netherlands) for the detection of CT and NG, according to the manufacturer's procedure. We used previously published methods for the detection of TV, MG, and CA [18–20]. We utilized the CFX platform (Bio-Rad, California, USA) for all amplifications.

We Gram-stained the fixated vaginal smear and examined it using the Nugent score. BV was defined by a Nugent score of above or equal to 7, an intermediate microbiota by a score between 4 and 6, and optimal vaginal microbiota by a score below 4.

Statistical methods

We determined the sample size using the prevalence of CT, NG, TV, MG, and BV from former studies conducted in Madagascar [3, 21]. For each pathogen, we employed a precision of 0.05 for prevalence rates equal to or exceeding 10% and a precision of 0.025 for rates below 10%, using two-sided 95% confidence intervals. The largest required sample size, 470, was retained and it was increased to 515 in order to accommodate potential attrition or sample loss.

We determined the prevalence of the infections and their Wald 95% confidence intervals (CI). We defined the outcome as having an STI, i.e., a positive test for either CT, NG, TV, or MG. We described the socio-demographic characteristics, behaviors, and medical features of participants. We compared those infected by an STI and those with no STI using Chi-2 or, in the case the application conditions were not met, the Fisher test. We created three age groups using standard categories for ease of comparison with other studies: 18–24, 25–34, and 35-45 [3, 12, 22]. We conducted univariable analyses to explore the associations between socio-demographic and behavioral characteristics of the participant and the outcome. Finally, we performed multivariable logistic regression to examine associations with STI infection, adjusting for age. We included the variables with $p \leq 0.2$ in the univariable analyses to build the final model using a backward approach. For all the logistic regressions conducted, our measures of effect were odd ratios (OR) in univariable analysis and adjusted odd ratios (aOR) for multivariable analysis. The statistical significance of variables in the final model was assessed at the 0.05 level. We used R (version 4.1.1) to perform the analyses.

This study followed the STROBE statement.

Ethical consideration

The ethical committee (Comité d'Ethique de la Recherche Biomédicale, CERBM) in Madagascar approved the study protocol (N° 124-MSANP/SG/AMM/CERBM). All participants gave their informed consent prior to joining the study.

Results

Between March and November 2022, 529 eligible women attended the CHUGOB. Among them, 14 did not consent to participate in the study, five bled during vaginal swabbing, three were positive for pregnancy after testing, three were lost to follow-up between the interview and sampling, and three withdrew consent, resulting in 501 included women (Fig. 1). Two-thirds of the women were recruited at the outpatient department (339/501, 68%), and a third were recruited at the family planning service (162/501, 32%). The main reasons for attending the outpatient department were abnormal vaginal discharge (220/339, 64.9%), lower abdominal pain (89/339, 26.3%), infertility (57/339, 16.8%), gynecological check-up (39/339, 11.5%), and abnormal vaginal bleeding (32/339, 9.4%) (Supplementary Fig. 1). We compared the most common reasons for attendance among women with STIs or BV and women without these conditions and found that 17% women with no STI/BV came for a gynecological check-up versus 6.8% of women with STI/BV ($p = 0.003$) (Supplementary Table 1). Additionally, we compared the prevalence of each STI, CA, and BV in the outpatient and family planning department and found no evidence of a difference, except for CA (19.2% at the outpatient department versus 8.6% at the family planning, $p = 0.004$) (Supplementary Fig. 2).

Study participants characteristics

The median age was 27 years (interquartile range (IQR): 23–32). More than half of the participants went at least to high school (132/501, 26%) or had higher education (189/501, 38%) (Table 1). A third of the participants (160/501, 32%) were employed by the public service or companies, whereas 151/501 (30%) owned their own businesses. The majority of them were either married (242/501, 48%) or in a non-marital relationship (191/501,

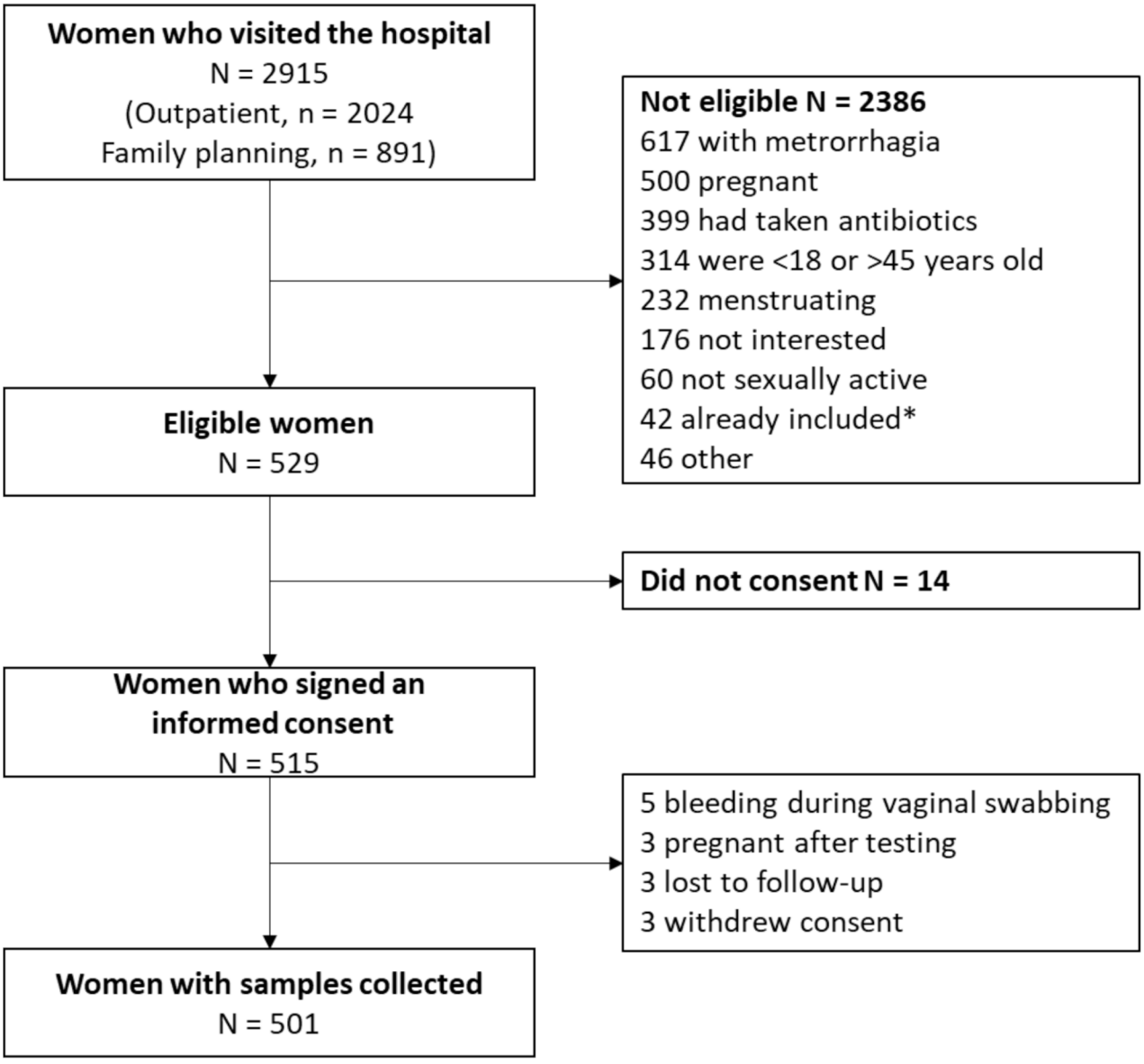


Fig. 1 Number and selection of women attending CHUGOB, Antananrivo, 2022
* These women already participated in the study and were no longer eligible to participate again. Per study design, women could be included only once

38%). In contrast, 68/501 (14%) were single or divorced. Most never used condoms (355/501, 71%). Although the majority of the women declared that they had a permanent partner (471/501, 94%) and lived with them for more than a year (444/501, 89%), concomitant partners were not uncommon with 61/444 (14%) of the latter stating they had other partners in the past 12 months. Similarly, 127/444 (28%) indicated that their permanent partner had other partners in the past 12 months. Circumcision among permanent partners was extremely common (460/471, 98%). Previous STIs were self-reported by 46/501 (9%) of the participants. Of note, 74/501 (15%) of the women declared having ever experienced forced sexual intercourse.

Prevalence of CT, NG, TV, MG, BV, and vaginal CA

The prevalence of CT was 11.8% (59/501, 95% CI: 9.0, 14.6), for NG 4.2% (21/501, 95%CI: 2.4, 5.9), for TV 14.8% (74/501, 95%CI: 11.7, 17.9), and for MG 8.2% (41/501, 95%CI: 5.8, 10.6). For BV, 11 vaginal smears contained too little material; consequently, the Nugent score could not be interpreted. Of the available samples, the prevalence was 39.6% (194/490, 95%CI: 35.3, 43.9). The prevalence of vaginal CA was 15.8% (79/501, 95%CI: 12.6, 19.0). Due to a shortage of HIV tests, only the first 281 women were

Table 1 Characteristics of women attending CHUGOB included in the study, stratified by infection status, Antananarivo, 2022

Characteristics	No STIs* (*N* = 349)		At least one STI* (*N* = 152)		Overall (*N* = 501)		*p*-value†
Socio-demographic							
Age							0.004
18–24	115	(33%)	70	(46%)	185	(37%)	
25–34	159	(46%)	65	(43%)	224	(45%)	
35–45	75	(21%)	17	(11%)	92	(18%)	
Education level							0.953
No schooling or primary school	43	(12%)	19	(13%)	62	(12%)	
Middle school	80	(23%)	38	(25%)	118	(24%)	
High school	92	(26%)	40	(26%)	132	(26%)	
Higher education	134	(38%)	53	(35%)	189	(38%)	
Marital status							< 0.001
Single / Divorced	44	(13%)	24	(16%)	68	(14%)	
In a relationship	116	(33%)	75	(49%)	191	(38%)	
Married	189	(54%)	53	(35%)	242	(48%)	
Activity							0.775
Owner of their own business	105	(30%)	46	(30%)	151	(30%)	
Civil servant / Private sector	116	(33%)	44	(29%)	160	(32%)	
Jobless / Housewife / On leave	79	(23%)	39	(26%)	118	(24%)	
Student	49	(14%)	23	(15%)	72	(14%)	
Behavior / Exposure to STI/HIV							
Personal hygiene							0.392
Wash vulva	244	(70%)	100	(66%)	344	(69%)	
Wash vulva and vagina	104	(30%)	52	(34%)	156	(31%)	
Prefer not to say‡	1	(< 1%)	0	(0%)	1	(< 1%)	
Has a permanent partner							0.025
No	17	(4.9%)	7	(4.6%)	24	(4.8%)	
Yes, not living with partner	69	(20%)	46	(30%)	115	(23%)	
Yes, living with partner	261	(75%)	95	(63%)	356	(71%)	
Prefer not to say‡	2	(< 1%)	4	(2.6%)	6	(1.2%)	
Other partner(s) in the past 12 months							0.009
No	302	(87%)	113	(74%)	415	(83%)	
Yes, one other partner	34	(9.7%)	24	(16%)	58	(12%)	
Yes, two or more other partners	11	(3.2%)	11	(7.2%)	22	(4.4%)	
Prefer not to say‡	2	(< 1%)	4	(2.6%)	6	(1.2%)	
Ever had sex in exchange for favors							0.004
No	345	(99%)	143	(94%)	489	(97%)	
Yes	4	(1.1%)	9	(5.9%)	13	(2.6%)	
Frequency of condom use							0.868
Always	5	(1.4%)	2	(1.3%)	7	(1.4%)	
From times to times	94	(27%)	45	(30%)	139	(28%)	
Never	250	(72%)	105	(69%)	355	(71%)	
Permanent partner had other partner(s) in past 12 months							0.028
No	137	(39%)	39	(26%)	176	(35%)	
Yes	88	(25%)	51	(34%)	139	(28%)	
Prefer not to say‡	0	(0%)	1	(< 1%)	1	(< 1%)	
Not concerned§	19	(5.4%)	11	(7.2%)	30	(6.0%)	
Do not know	105	(30%)	50	(33%)	155	(31%)	
Current partner(s) reported STI symptom							0.136
No	253	(72%)	98	(64%)	351	(70%)	
Yes	92	(30%)	50	(33%)	142	(28%)	
Do not know†	4	(1.5%)	4	(2.6%)	8	(1.6%)	
Biological testing							

Table 1 (continued)

Characteristics	No STIs* (*N* = 349)		At least one STI* (*N* = 152)		Overall (*N* = 501)		*p*-value†
Candida albicans	66	(18.9%)	13	(8.6%)	79	(15.8%)	0.009
Bacterial vaginosis (BV)¶	120/341	(35.2%)	74/149	(49.7%)	194/490	(39.6%)	
Intermediate microbiota	59/341	(17.3%)	36/149	(24.2%)	95/490	(19.4%)	0.003

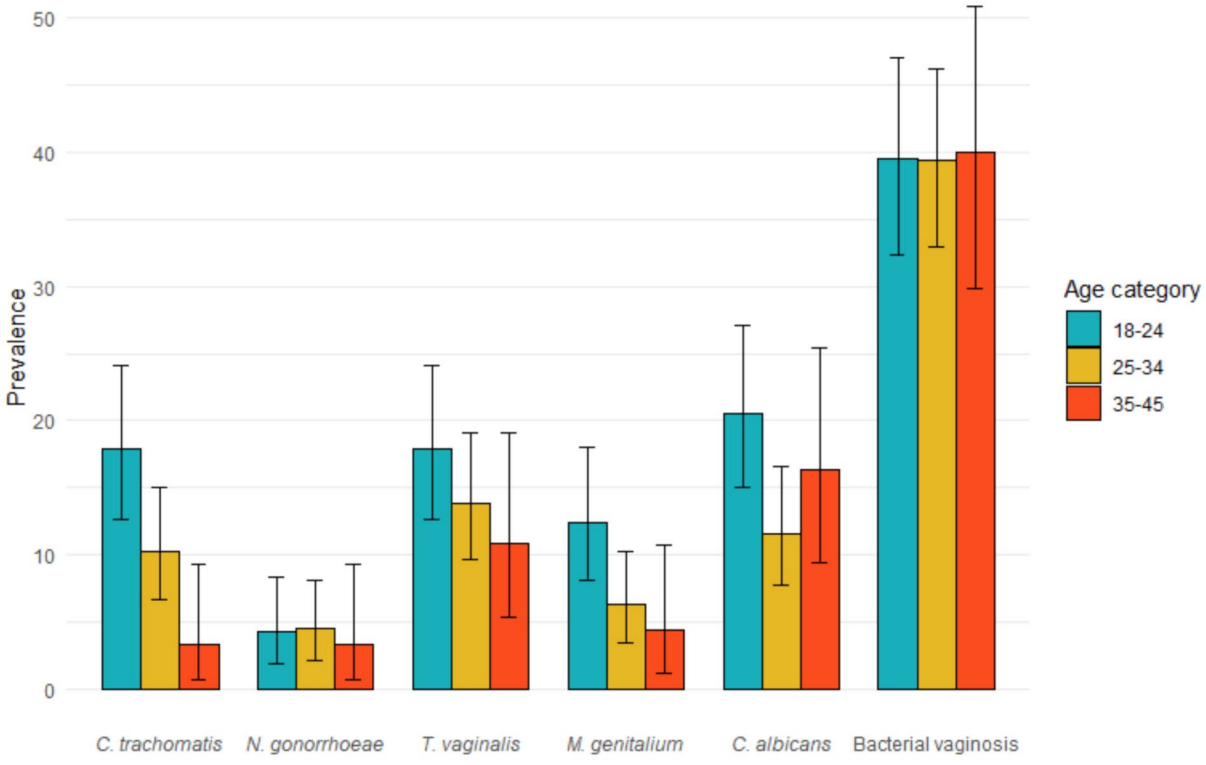
* STI refers to the detection of either CT, NG, TV, or MG

† Using Chi² or Fisher test

‡ Excluded from the analyses with the Chi² or Fisher tests

§ No permanent partner or did not want to say if they had one

¶ 11 women could not be tested for BV



	CT	NG	TV	MG	CA	BV
18-24 (n=185)	17.8 (12.6, 24.1)	4.3 (1.9, 8.3)	17.8 (12.6, 24.1)	12.4 (8.0, 18.1)	20.5 (15.0, 27.1)	39.6 (32.4, 47.1)*
25-34 (n=224)	10.3 (6.6, 15.0)	4.5 (2.2, 8.1)	13.8 (9.6, 19.1)	6.3 (3.5, 10.3)	11.6 (7.7, 16.5)	39.4 (32.9, 46.3)†
35-45 (n=92)	3.3 (0.7, 9.2)	3.3 (0.7, 9.2)	10.9 (5.3, 19.1)	4.3 (1.2, 10.8)	16.3 (9.4, 25.5)	40.0 (29.8, 50.9)‡

Fig. 2 Prevalence of CT, NG, TV, MG, CA, and BV by age groups, with 95% confidence intervals, among women attending CHUGOB, Antananarivo, 2022
* Denominator is 182 for BV in 18–24 age group
† Denominator is 218 for BV in 25–34 age group
‡ Denominator is 90 for BV in 35–45 age group

tested, and among them, two were HIV seropositive, resulting in an HIV prevalence of 0.7% (95%CI: 0.1, 2.5).

We identified that 152/501 (30.3%) women had at least one infection by CT, NG, TV, or MG. Thirty women (35/501, 7.0%) had a dual infection, with the combinations of CT-NG and CT-TV being the most common co-infections (8/30 each).

When stratifying by age category, we observed that as age increased, CT prevalence decreased ($p = 0.004$). A similar but not significant trend was observed for TV and MG (Fig. 2).

Table 2 Factors associated with at least one of CT, NG, TV, or MG among 463 women attending CHUGOB in Antananarivo, 2022

Characteristic	*n*	Univariable analyses			Multivariable analysis		
		OR	95% CI	*p*-value	aOR	95% CI	*p*-value
Age category				0.005			0.100
18–24	172	2.71	1.46, 5.27	0.002	2.05	1.06, 4.15	0.039
25–34	204	1.95	1.06, 3.79	0.038	1.69	0.89, 3.34	0.12
35–45	87	-	-		-	-	
Marital status				0.001			0.034
Married	226	-	-		-	-	
In a relationship	177	2.25	1.46, 3.51	< 0.001	1.86	1.16, 2.98	0.010
Single or divorced	60	1.67	0.88, 3.11	0.110	1.34	0.68, 2.58	0.4
Permanent partner				0.063			
Yes, living with partner	337	-	-				
Yes, not living with partner	103	1.74	1.09, 2.77	0.019			
No permanent partner	23	0.97	0.34, 2.41	> 0.9			
Participant had other partners*				0.035			
No	392	-	-				
Yes, one	50	1.80	0.97, 3.29	0.058			
Yes, two or more	21	2.45	1.00, 5.98	0.047			
Partner had other partners*				0.052			
No	167	-	-				
Yes	131	1.97	1.18, 3.29	0.009			
Do not know	142	1.68	1.02, 2.81	0.044			
Not concerned	23	1.24	0.42, 3.23	0.7			
Ever had sex in exchange for favors							
No	451	-	-		-	-	
Yes	12	5.05	1.56, 19.2	0.007	4.25	1.27, 16.6	0.019
Vaginal microbiota				< 0.001			0.003
Optimal	194	-	-		-	-	
Intermediate	90	2.30	1.32, 3.54	0.003	2.10	1.19, 3.72	0.010
Bacterial vaginosis	179	2.21	1.39, 3.54	< 0.001	2.14	1.34, 3.47	0.002

* in the past 12 months

Among women with an STI, 92/152 (60.5%) had at least one symptom that corresponded to the entry points of the WHO syndromic management algorithms for the investigated STIs: vaginal discharge syndrome (which includes: abnormal vaginal discharge and/or irritation and/or pruritus) and lower abdominal pain [23]. Of the infected women, 60/152 (39.5%) were asymptomatic. Among the women with abdominal pain, 25% were infected by TV.

Risk factors associated with STIs

We conducted the analyses on complete data ($n = 463$), excluding participants with missing BV results (11/501, 2.2%) and those who preferred not to answer or did not know the response to specific questions (27/501, 5.4%). We compared age, education, marital status, activity, and STI status between those excluded and included in the risk factors analysis and found no differences in the socio-demographic characteristics (Supplementary Table 2).

In the multivariable analysis, adjusting for age, we found that women in a relationship but not married had 86% increased odds of having an STI compared to married women (aOR: 1.86, 95%CI: 1.16, 2.98, $p = 0.01$) (Table 2). We found that the odds of having an STI were 2.14 higher in those with BV compared to those with an optimal microbiota (95%CI: 1.34, 3.47, $p = 0.002$); likewise, women with intermediate microbiota had twice the odds of having an STI (aOR: 2.10, 95%CI: 1.19, 3.72, $p = 0.010$). Lastly, women who had sex in exchange for favors (e.g., money, goods, or jobs) had 4-fold increased odds of having an STI (aOR: 4.25, 95%CI: 1.27, 16.6, $p = 0.019$). Following this result, we then compared the socio-demographic characteristics of women who had sex in exchange for favors with those who did not. We found that women who had sex in exchange for favors had a lower education level than the other women ($p = 0.008$). Analysis of factors associated with each STI separately showed that CT was associated with younger age, NG with sex in exchange for favors and TV with sex in exchange for favors and perturbation of the vaginal microbiota (Supplementary Tables 3–5). No factor was found to be associated with MG.

Discussion

We identified a high prevalence of STIs in women of child-bearing age attending CHUGOB in Antananarivo. One third of the women were infected with at least one STI.

We estimated the prevalence of CT and NG to be 11.8% (95%CI: 9.0, 14.6) and 4.2% (2.4, 5.9), respectively. Our findings are similar to those determined in rural Madagascar (for which we calculated the 95%CI): 9.3% (95%CI: 6.4, 13.0) and 5.4% (3.2, 8.4), respectively [3]. In our study, the prevalence of MG exceeded that reported in this study (8.2% [5.8, 10.6] compared to 2.7% [1.2, 5.1]). Conversely, the prevalence of TV was lower in our investigation (14.8% [11.7, 17.9]) compared to the cited study (23.4% [19.0, 28.3]). However, these estimates must be compared critically, since the two studies were conducted 20 years apart, and one in the capital city and the other in rural areas. BV prevalence in our study (39.6% [35.3, 43.9]) was consistent with findings among women in rural Madagascar (33.0% [28.0, 38.4]) [3] and very similar to estimates found in the rest of Africa [24].

In a meta-analysis that estimated STI prevalence among women in different African regions, our figures were closer to the estimates found among young women in East Africa and at high risk, i.e., recruited from recreational facilities or engaged in transactional sex [24]. The prevalence of CT in our study was similar to the estimate for this high-risk group: 10.3% (7.4, 14.1). Likewise, we found a TV prevalence of 14.8% (11.7, 17.9), comparable to the 12.7% (7.5, 20.6) estimate among women at risk.

The prevalence of MG in our study was 8.2% (5.8, 10.6), which was higher than the estimate found in Tanzania (3.2% [2.8, 4.2]) [2], but comparable to those in pregnant women in Sub-Saharan Africa (13.5% [4.0, 27.2]) [25]. However, it was lower than estimates from adolescents and young women at risk (< 25 years) in South Africa (22.1% [19.3, 25.2]) [26] but more than six times higher than in countries with high development index (1.3% [1.0 to 1.8]) [27].

Our results raise concerns about the broad circulation of STIs among Malagasy women of child-bearing age. The average level of education in the participants of our study was higher than the level representative samples of women from Antananarivo [28], which may lead them to be better informed of their health issues or STI-associated risk behaviors and consult more often. Thus, we may have not captured women with intermediate to high risk of STI, and consequently the true prevalence of STI may be even more substantial than the one we observed.

6% of the women were co-infected by two STIs or more, a result that is much lower in our study in Madagascar than other studies conducted in Tanzania (19%) [29] and South Africa (14%) [30]. Consequences of a dual infection remain unclear. The simultaneous presence of STIs can complicate diagnosis and treatment, and the potential effects on long-term health outcomes have yet to be studied.

Among factors related to having an STI, we found that being in a relationship was associated with an increased odds of having an STI, using being married as the reference. This association can be explained by a higher number of concurrent partners among unmarried women in couple: our data shows that 5.7% of the married women had other partners in the past 12 months, compared with 20.3% of unmarried women in a relationship. Relationships that overlap in time are not uncommon among unmarried young individuals in Madagascar [31] and have been shown to be associated with an increased risk of STIs compared to single relationships [32].

Although sex in exchange for favors concerned only less than 3% of the participants, the magnitude of this association was the highest among all explored risk factors. The proportion of women who declared having ever exchanged sex for favors in our study was comparable to another study conducted in Antananarivo among the 15-24-year (5.9%).[31] The proportion of women who exchanged sex for favors in our study is consistent with demographic health surveys (DHS) data in Africa summarized in a meta-analysis where 4.34% (3.16, 5.51) of the women were concerned [33]. Analyses of the features of those women in our data showed an association with low education, a factor also stressed in the meta-analysis [33]. Women with low education may have difficulty finding a job or have jobs with low pay, and they may therefore turn to selling sex as a complementary income.

Lastly, we found that the women with BV or intermediate microbiota had twice the odds of having an STI compared to women with an optimal microbiota, an association extensively documented in previous studies [34, 35] However, it remains unclear whether BV increases susceptibility to STIs or if STIs predispose individuals to BV. Several cohort studies have investigated the incidence of STIs in women with BV, reporting a higher risk of CT, NG, and TV infections [36]. From a biological standpoint, this association may be attributed to BV-associated reductions in lactobacilli, which normally produce antimicrobial compounds, as well as the activity of sialidase and mucinase enzymes produced by BV-associated bacteria that compromise immune and physical barriers [37]. Conversely, other studies have reported an elevated risk of BV among women with STIs [38]. Further research is needed to elucidate the underlying immunological and mechanistic pathways by which these conditions influence each other.

Our findings provide additional context on the demographic distribution of STIs. Women aged 18–24 years were more likely to have a CT infection than those aged 35–45. This asymmetric burden among people below

25 years has also been documented in national surveillance data in the United States [39] and in a large study in China [22]. This increased risk among young women can be explained by behavioral factors such as a higher number of sexual partners, but also biological factors such as cervical ectopy [40].

We also observed that condoms were rarely used, a finding shared by other studies conducted in Madagascar [41, 42]. In the study conducted by Rahamefy et al., low condom use was justified by steady relationships; [42] however, our study did not find a difference in condom use between married and unmarried women. According to the last DHS conducted in Madagascar, condom uptake with a partner that was neither the spouse nor the person they were living with increases with education: it ranges from 1.2% among those who have never been to school to 12.6% among those who went to university [28]. When comparing with other African countries, a study pointed out that, out of 18 countries, Madagascar has the lowest condom use among men aged 15–29 years at last intercourse and the lowest percentage of men who know about condoms [41]. Historically, the Malagasy society was influenced by Christian values and condom use was not favorably regarded. This attitude may have endured over time, shaping cultural perceptions and impacting sexual health practices [43].

Paradoxically, despite high proportions of STIs, the prevalence of HIV in our study was low (0.7%) and consistent with the estimate found in a study conducted between 2010 and 2015 in Madagascar (0.4%) [15]. Low HIV prevalence could be explained by the isolated island status of Madagascar and the fact that most men are circumcised. While evidence that circumcision is protective against HIV has been established, a recent review found mixed evidence against the acquisition and transmission of CT or NG and no evidence against MG [44].

One of our important results is that nearly 40% of the women with an STI did not present with symptoms of the WHO syndromic management guidelines. This result indicates that, outside the context of our study, a substantial fraction of the women with an STI would remain untreated, as they showed no symptoms, and they may develop complications and sustain the transmission of STIs. We also noted that among women with lower abdominal pain, one in four were infected by TV. Other studies have also reported an association between TV and lower abdominal pain [45, 46]. However, the WHO guidelines recommend treatment only for only for CT, NG, bacteria associated with BV, and MG in case of lower abdominal pain [23]. The importance of lower abdominal pain in TV infection warrants further investigation.

Our study had several limitations. Firstly, we conducted this study in the capital city, and the results may not be representative of the women in Madagascar. In our study population, 88% of the participants went to middle school or above, whereas at the national level, only 31% of girls do [28]. However, the DHS shows that knowledge on HIV prevention and condom uptake increases with the level of education [28]. Therefore, women included in our study may be less at risk for STIs, and our estimates, which were already high, may be even higher in the Malagasy female population.

Secondly, our questionnaire contained sensitive questions which have likely caused social desirability bias. This bias may have affected the associations in our analysis.

Despite these limitations, this study has several strengths. It is the first study on CT, NG, TV, and MG among women who are not sex workers, in the past twenty years [3]. Additionally, we had no missing data for determining the prevalence rates except for BV, for which the inability to analyze the samples was due to random events. We found no difference in characteristics between the 95% of participants with complete data and the 5% with incomplete data. Thus, we believe our results are strongly reliable. Finally, the laboratory team was rigorously trained, and emphasis was made on the quality of the procedures using the most recent recommendation for the detection of the pathogens.

In conclusion, the high prevalence of STIs in this urban, educated female population showed a need for intervention and prevention, such as the promotion of and negotiation skills on condom use for both men and women who have occasional partners, especially those who exchange sex for favors. Young people should be targeted in priority due to their higher likelihood of being infected and their misconceptions about condoms [47].

Further studies on the prevalence of STIs and HIV in other cities and rural areas should be considered, especially in Southern regions where polygamy and extramarital relationships are far more common. Similarly, STI diffusion could follow a different pattern in coastal cities where sex tourism is common [48]. A comprehensive picture of the epidemiology of STIs, including HIV, at the country level is needed to serve as a guide for future public health prevention and intervention programs.

Abbreviations

aOR	Adjusted Odds Ratio
BV	Bacterial vaginosis
CA	*Candida albicans*
CT	*Chlamydia trachomatis*
CHUGOB	Gyneco-Obstetrics Befelatanana University Hospital
DHS	Demographic and Health Surveys
HIV	Human Immunodeficiency Virus
IPM	Institut Pasteur de Madagascar
MG	*Mycoplasma genitalium*
NG	*Neisseria gonorrhoeae*
STIs	Sexually Transmitted infections
TV	*Trichomonas vaginalis*
UBEX	Experimental Bacteriology Unit
WHO	World Health Organization

Supplementary Information

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Supplementary Material 1

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Author contributions
CF designed the study, analyzed the data, and wrote the manuscript.ANH designed the study and supervised the coordination.SBR supervised the coordination.TFR coordinated the study.HLA coordinated the study.RTM supervised data quality.DTR developed the electronic case report forms and ensured data quality.LFR coordinated and carried out the biological analysis. TTR carried out the biological analysis.LR carried out the biological analysis. BTH contributed to the design, the analysis, the results, and the writing of the manuscript.RVR contributed to the design, the implementation, the analysis, the results and the writing of the manuscript, and supervised the field study. TC contributed to the design, the implementation, the analysis, the results and the writing of the manuscript, and supervised the biological analyses.

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Data availability
The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate
The ethical committee (Comité d'Ethique de la Recherche Biomédicale, CERBM) in Madagascar approved the study protocol (N° 124-MSANP/SG/AMM/CERBM). All participants gave their informed consent prior to joining the study.

Consent for publication
Not applicable.

Disclosure
This work was, in part, presented at the STI & HIV 2023 world congress.

Competing interests
The authors declare no competing interests.

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