

# Clinical Characteristics Associated with *Mycoplasma genitalium* among Female Sex Workers in Nairobi, Kenya

Ayodele Gomih-Alakija,<sup>a</sup> Jie Ting,<sup>a</sup> Nelly Mugo,<sup>b</sup> Jessie Kwatampora,<sup>b</sup> Damon Getman,<sup>c</sup> Michael Chitwa,<sup>b</sup> Suha Patel,<sup>d</sup> Mugdha Gokhale,<sup>a</sup> Joshua Kimani,<sup>b</sup> Frieda S. Behets,<sup>a</sup> Jennifer S. Smith<sup>a,e</sup>

Department of Epidemiology, Gillings School of Global Public Health, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, USA<sup>a</sup>; Kenyatta National Hospital/University of Nairobi, Nairobi, Kenya<sup>b</sup>; Hologic Gen-Probe, San Diego, California, USA<sup>c</sup>; Department of Obstetrics, Gynecology, and Reproductive Sciences, University of California, San Francisco, California, USA<sup>d</sup>; Lineberger Comprehensive Cancer Center, Chapel Hill, North Carolina, USA<sup>e</sup>

The prevalence of *Mycoplasma genitalium* is high in vulnerable populations of women in low-resource settings. However, the epidemiology of infection in these populations is not well established. To determine the prevalence of *Mycoplasma genitalium* and its association with cervical cytology and other correlates, we recruited 350 female sex workers (FSW) who were 18 to 50 years old in Nairobi, Kenya, for a cross-sectional study. A questionnaire was administered at baseline to obtain information on sociodemographics and sexual behaviors. Women underwent a pelvic exam, during which a physician collected cervical-exfoliation samples for conventional cytology and sexually transmitted infection (STI) testing. Samples were tested for *M. genitalium* and other STI organisms (*Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Trichomonas vaginalis*) and the E6/E7 mRNA of human papillomavirus (HPV) by Aptima nucleic amplification assays. The prevalence of *M. genitalium* was 12.9%. FSW who engaged in sexual intercourse during menses were less likely to have *M. genitalium* infection than those who did not (odds ratio [OR], 0.3; 95% confidence interval [95% CI], 0.1, 0.9). *M. genitalium* was also less prevalent among FSW who had worked in prostitution for >5 years (6.2%) than among those who had worked for <3 years (17.6%) (OR, 0.3; 95% CI, 0.1, 0.8). FSW who reported more frequent condom use were more likely to be infected with *M. genitalium* than those who reported less frequent use (OR, 3.8; 95% CI, 1.2, 11.6). These correlates differ from those found in *M. genitalium* studies conducted with FSW from West Africa and China. Further longitudinal analyses assessing associations with persistent *M. genitalium* infection are needed.

*Mycoplasma genitalium* is an emergent bacterium that is transmitted via sexual activity and has been associated with complications in the female genital tract, including endometritis, tubal-factor infertility, pelvic inflammatory disease (PID), cervicitis, and ectopic pregnancy (1–9). *M. genitalium* may also increase the risk of HIV transmission and associated shedding (10–13).

Vulnerable populations of women in low-resource settings have a higher-than-average susceptibility to sexually transmitted infections (STIs) and subsequent complications of the reproductive tract, including those caused by *M. genitalium*, *Chlamydia trachomatis*, and *Neisseria gonorrhoeae* (14). In the case of female sex workers (FSWs), STI risk is increased due to a greater exposure to unsafe sexual behaviors, such as unprotected sexual intercourse and high numbers of clients/sexual partners (15).

The prevalence and associated risk factors for *M. genitalium* have been investigated both in the general population and among populations at high risk for STIs in sub-Saharan Africa, Asia, the United Kingdom, and the United States (4, 16–23). Across many of these studies, a young age, little education, single marital status, and high numbers of sex partners remained consistently associated with infection (4, 9, 16, 18, 21, 23, 24). Findings are less consistent among the few studies that examined risk factors among FSWs. In the literature, studies have shown similar associations of *M. genitalium* infection with basic demographic factors, such as education and marital status. However, there is less agreement for sexual-behavioral and clinical correlates, such as condom use, duration of sex work, and history of STIs (16, 18, 23). Chinese sex workers who did not present STI symptoms in the year prior to the study were more likely to have infection with *M. genitalium* than those who did (23). In contrast, a study conducted in sub-Saharan Africa found that sex workers who presented clin-

ical symptoms were more likely to have *M. genitalium* infection than those who were asymptomatic (18).

In studies examining concurrent *M. genitalium* infection with other STIs in the general population, *M. genitalium* was found to be associated with urethritis, cervicitis, and PID (25–28). However, more-recent research indicates the role of *M. genitalium* in nonchlamydial nongonococcal urethritis (NCNGU), supporting its independent role as an etiological factor for NCNGU and other urogenital complications (29, 30). While the role of human papillomavirus (HPV) in the etiology of cervical cancer is well established, evidence demonstrates a positive relationship between *M. genitalium* and HPV infection as well as abnormal cervical cytology (8, 31–34). *M. genitalium* was associated with a higher risk of HPV among Japanese men with urethritis, with both organisms present in the urethral tract (33). Among a cohort of FSWs, *M. genitalium* was positively associated with the prevalence of high-risk HPV (hrHPV) (32). An *in vitro* study of human cells infected with four urogenital mycoplasmas found that *M. genitalium* infection altered the gene expression of cervical epithelial cells, supporting its potential role in cervical dysplasia and cervical cancer progression (35). Infection with *M. genitalium* has also been pos-

Received 11 April 2014 Returned for modification 14 May 2014

Accepted 16 July 2014

Published ahead of print 6 August 2014

Editor: E. Munson

Address correspondence to Jennifer S. Smith, [jennifers@email.unc.edu](mailto:jennifers@email.unc.edu).

Copyright © 2014, American Society for Microbiology. All Rights Reserved.

doi:10.1128/JCM.00850-14

TABLE 1 Prevalence of *Mycoplasma genitalium* and other sexually transmitted infections among female sex workers in Kenya

Parameter	Value for subjects infected with <sup>a</sup> :					
	<i>Mycoplasma genitalium</i>	<i>Chlamydia trachomatis</i>	<i>Neisseria gonorrhoeae</i>	<i>Trichomonas vaginalis</i>	HIV-1	hrHPV
Overall % of subjects infected	12.9	3.7	2.3	7.2	24.0	29.5
Median age in yr (range)	26 (19–46)	23 (19–31)	26 (21–35)	30 (22–44)	32 (21–48)	32 (20–43)
% in age group (yr):						
18–24 (n = 89)	18.0	11.4	3.4	9.0	7.9	32.6
25–29 (n = 108)	13.9	1.9	2.8	3.7	18.5	29.6
30–34 (n = 72)	8.3	1.4	1.4	4.2	36.6	30.6
≥35 (n = 80)	10.0	0.0	1.3	12.5	38.5	25.0
P trend	0.07	<0.001	0.34	0.40	<0.001	0.33
% with indicated duration of prostitution (yr)						
<3 (n = 74)	17.6	10.8	4.1	5.4	18.9	27.0
3–5 (n = 145)	16.6	2.8	1.4	9.7	21.7	31.7
>5 (n = 130)	6.2	0.8	2.3	5.4	29.5	28.5
P trend	<0.01	<0.001	0.64	0.78	0.07	0.94

<sup>a</sup> HIV-1, human immunodeficiency virus type 1; hrHPV, high-risk human papillomavirus.

itively associated with cervical inflammation, a condition associated with high-risk cervical neoplasia (8, 9, 36, 37).

There are widespread implications of understanding the characteristics of *M. genitalium* infection in populations susceptible to STIs and related complications. Therefore, the objective of this study was to determine the prevalence of *M. genitalium* infection, identify correlates for infection, and explore the relationship between *M. genitalium* and cervical cytology in a cohort of FSWs in Kenya.

## MATERIALS AND METHODS

**Study population.** Ethical approval was granted by the Institutional Review Boards (IRB) at Kenyatta National Hospital (Nairobi, Kenya) and the University of North Carolina (Chapel Hill, NC, USA). From August 2009 to March 2011, FSWs attending the Korogocho clinic in Nairobi, Kenya, were invited to participate in this study to determine the prevalence of correlates for *M. genitalium* infection. The clinic provided counseling and medical care, including screening and treatment of cervical cancer as well as of STIs for FSWs in the Korogocho slum area. Detailed information on the study population and sample collection has been previously described (38). Briefly, a total of 350 FSWs aged 18 to 48 years provided written informed consent and were subsequently enrolled from among approximately 425 FSWs who were invited to participate in the study. Each participating woman underwent a pelvic examination, where the physician collected cervical specimens for cytology, histology, and STI testing, including for *M. genitalium* and HPV infections.

***M. genitalium* and STI testing.** Encoded, deidentified cervical samples were transported to Hologic Gen-Probe in San Diego, CA, for *M. genitalium* DNA testing using the Aptima research-use-only assay (Hologic Gen-Probe), which uses target capture, transcription-based amplification (TMA), and hybridization protection to capture, amplify, and detect *M. genitalium* 16S rRNA (39). The specimens were also tested (i) for *Chlamydia trachomatis* and *Neisseria gonorrhoeae* by the Aptima Combo 2 assay, (ii) for *Trichomonas vaginalis* by the Aptima TV assay, and (iii) for HPV by the Aptima HPV assay, which qualitatively detects E6/E7 mRNA of 14 hrHPV types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68) (Hologic Gen-Probe, Inc., San Diego, CA). All assays utilized target capture, TMA, and hybridization principles similar to those used for *M. genitalium* detection.

Serum was tested for HIV antibodies by enzyme-linked immunosorbent assay (ELISA) (Detect HIV-1; Biochem ImmunoSystems, Inc., Montreal, Canada), with positive results confirmed by a second ELISA. Peripheral blood CD4 cells were also enumerated. The HIV ELISA and CD4 assays were conducted at the University of Nairobi. All assays were performed according to the manufacturer's instructions, with the technicians blind to cervical cytology, laboratory, and other study results.

**Cervical cytology.** The Pap smears were evaluated at the University of Nairobi and classified according to the 2001 Bethesda System (40). All smears were independently read by two cytopathologists blind to *M. genitalium* and all other study results. In cases of discordant diagnoses between the two cytology readings, the slides were reassessed a third time, with the final diagnosis being the consensus of the opinions of the reviewing cytopathologists. Study participants were notified of their cytology results within 2 weeks after their screening visit. Women with low-grade squamous intraepithelial lesions (LSIL) were instructed to undergo a repeat cytology 4 months later. Women with high-grade squamous intraepithelial lesions (HSIL) were immediately referred for a colposcopy and biopsy. In cases of histological cervical intraepithelial neoplasia 2 (CIN-2) or more-severe neoplasia (at least CIN-2), the women received standard care and treatment at Kenyatta National Hospital.

**Statistical analysis.** Of the 350 FSWs recruited, 1 woman was missing baseline questionnaire data and was excluded from analyses, resulting in a final sample size of 349. Distributions of *M. genitalium* and other STIs were assessed by univariate analyses. Odds ratios (ORs) and 95% confidence intervals (CI) were calculated by unconditional logistic regression to examine crude associations between baseline characteristics and *M. genitalium*. The Wald test was utilized to determine the inclusion of variables for the multivariable logistic-regression model. Any correlate associated with *M. genitalium* at a *P* of 0.15 were selected for the multivariable model. Additionally, *M. genitalium* was reviewed as a correlate of cervical cytology. Binary and categorical variables with a sample size of less than 10 in a stratum (*N. gonorrhoeae*, CD4 count) were eliminated from the regression analyses when looking within strata of *M. genitalium* infection. Analyses were performed using SAS version 9.2.

## RESULTS

The median age of study participants was 28 years overall (range, 18 to 48 years). The median age at sexual debut was 16 years (range, 10 to 25 years), and the average duration of prostitution

**TABLE 2** Sociodemographic, sexual-behavioral, and biological factors associated with *Mycoplasma genitalium* among 349 female sex workers in Kenya

Parameter	Overall no. of subjects (n = 349)	% infected with <i>Mgen</i> <sup>a</sup> (n = 45)	Crude OR (95% CI) <sup>b</sup>
<b>Sociodemographic characteristics</b>			
Age (yr)			
18–24	89	18.0	1.0 <sup>f</sup>
25–29	108	13.9	0.7 (0.3, 1.6)
30–34	72	8.3	0.4 (0.2, 1.1)
≥35	80	10.0	0.5 (0.2, 1.3)
Marital status <sup>c</sup>			
Single (never married)	154	11.7	1.0
Married/cohabiting	3	0.0	
Widowed/divorced/separated	191	14.4	1.2 (0.6, 2.3) <sup>d</sup>
Education <sup>c</sup>			
Primary or less	265	13.6	1.0
Secondary or more	83	10.8	0.8 (0.4, 1.7)
Income/mo <sup>c,e</sup>			
≤4,000 Ksh	201	10.5	1.0 <sup>f</sup>
>4,000 Ksh	147	16.3	1.7 (0.9, 3.1)
Smoking status			
Never	217	12.4	1.0
Past	46	17.4	0.9 (0.4, 2.0)
Yes, still	86	11.6	1.5 (0.6, 3.5)
Alcohol use <sup>c</sup>			
No	57	10.5	1.0
Yes	291	13.4	1.3 (0.5, 3.3)
<b>Sexually transmitted infection characteristics</b>			
HIV-1 serology <sup>c</sup>			
Negative	291	12.0	1.0
Positive	83	17.2	1.2 (0.6, 2.4)
HPV RNA			
Negative	246	11.4	1.0
Positive	103	16.5	1.5 (0.8, 3.0)
<i>Chlamydia trachomatis</i> <sup>c</sup>			
Negative	335	12.8	1.0
Positive	13	7.7	0.6 (0.1, 4.5)
<i>Trichomonas vaginalis</i>			
Negative	324	12.7	1.0
Positive	25	16.0	1.3 (0.4, 4.0)
<b>Sexual-behavioral characteristics</b>			
Avg charge for sex <sup>c,e</sup>			
≤200 Ksh	199	10.6	1.0
>200 Ksh	149	16.1	1.6 (0.9, 3.1)
Duration of prostitution (yr)			
<3	74	17.6	1.0 <sup>f</sup>
3–5	145	16.6	0.9 (0.4, 2.0)
>5	130	6.2	0.3 (0.1, 0.8) <sup>g</sup>
Avg no. of clients/wk			
≤8	120	14.2	1.0
9–14	127	8.7	0.6 (0.3, 1.3)
≥15	102	16.7	1.2 (0.6, 2.5)

**TABLE 2** (Continued)

Parameter	Overall no. of subjects (n = 349)	% infected with <i>Mgen</i> <sup>a</sup> (n = 45)	Crude OR (95% CI) <sup>b</sup>
<b>Avg frequency of condom use<sup>c</sup></b>			
Half the time or less	92	4.4	1.0 <sup>f,g</sup>
Most of the time	168	16.7	4.4 (1.5, 13.0)
Always	88	14.8	3.8 (1.2, 12.2)
<b>Has a regular partner(s)</b>			
No	100	13.0	1.0
Yes	249	12.9	1.0 (0.5, 2.0)
<b>Age at menarche (yr)</b>			
≤13	100	8.0	1.0 <sup>f</sup>
14–15	147	14.3	1.9 (0.8, 4.5)
≥16	102	15.7	2.1 (0.9, 5.3)
<b>Age at sexual debut (yr)</b>			
≤14	136	13.2	1.0
15–18	121	13.2	1.0 (0.5, 2.1)
≥19	92	12.0	0.9 (0.4, 2.0)
<b>Age at parity (yr)</b>			
≤17	150	12.0	1.0
18–20	138	14.5	1.2 (0.6, 2.5)
≥21	61	11.5	1.0 (0.4, 2.4)
<b>Depo/Norplant use</b>			
No	239	11.7	1.0
Yes	110	15.5	1.4 (0.7, 2.6)
<b>Douche after sex<sup>c</sup></b>			
No	85	10.6	1.0
Yes	177	14.1	1.4 (0.6, 3.1)
More than once per day	20	30.0	2.8 (0.9, 9.0)
Once per day or less	157	12.1	0.9 (0.4, 2.0)
<b>Sex while in menses</b>			
No	271	15.5	1.0 <sup>f,g</sup>
Yes	78	3.9	0.2 (0.1, 0.7)
<b>Anal sex<sup>c</sup></b>			
No	268	13.4	1.0 <sup>f</sup>
Yes	80	11.3	0.2 (0.03, 1.7)
<b>Lubricant use during sex</b>			
No	222	11.7	1.0 <sup>f</sup>
Yes	127	15.0	0.8 (0.4, 1.4)

<sup>a</sup> *Mgen*, *Mycoplasma genitalium*.

<sup>b</sup> OR, odds ratio; 95% CI, 95% confidence intervals.

<sup>c</sup> Numbers do not add up to the total sample size due to missing data.

<sup>d</sup> The odds ratio represents a comparison between the categories “married/cohabiting” and “widowed/divorced/separated” combined (collapsed due to sparse data) and the referent “single/never married.”

<sup>e</sup> 4,000 Kenyan shillings (Ksh) is equivalent to \$50 U.S.

<sup>f</sup> A *P* of ≤0.15 was selected for the multivariable logistic-regression model.

<sup>g</sup> *P* < 0.05.

was 5.5 years (Table 1). Most of the women had never been married or were widowed, divorced, or separated and had completed a primary school education (Table 2). Among the 349 FSW with baseline sample data, the prevalences of *M. genitalium*, *C. trachomatis*, *N. gonorrhoeae*, and *T. vaginalis* infection were 12.9%, 3.7%, 2.3%, and 7.2%, respectively.

**Trends of sexually transmitted infections by age.** The median age for women infected with *M. genitalium* was 26 years (range, 19 to 46 years) (Table 1). The prevalence of both *M. genitalium* and *C. trachomatis* infection was higher among 18- to 24-year-olds than

among older women ( $P$  trends, 0.07 and  $<0.001$ , respectively). However, age and HPV infection were positively correlated in that women aged 35 years and older had a higher prevalence of infection than younger FSWs ( $P$  trend,  $<0.001$ ). Women who had worked in prostitution for less than 3 years had a high prevalence of *M. genitalium* infection (17.6%), but infection decreased with more years in prostitution (6.2%) ( $P$  trend,  $<0.01$ ).

**Independent correlates of *Mycoplasma genitalium*.** The duration of prostitution was negatively associated with *M. genitalium*; women who had worked in prostitution for  $>5$  years were less likely to be infected than women who had practiced it for  $<3$  years (OR, 0.3; 95% CI, 0.1 to 0.8) (Table 2). Women who self-reported using condoms most of the time or all the time were more likely to have *M. genitalium* infection than those who self-reported using condoms less frequently (OR, 4.4; 95% CI, 1.5 to 13.0; OR, 3.8; 95% CI, 1.2 to 12.2, respectively).

Participants who reported having sexual intercourse during menses were less likely to have *M. genitalium* infection than those who did not (OR, 0.2; 95% CI, 0.1 to 0.7). *M. genitalium* did not appear to be associated with other STIs (*N. gonorrhoeae* tests were not conducted due to insufficient sample size) or participant sociodemographic characteristics (Table 2).

**Multivariable analysis.** Based on the univariable analyses, variables selected for the multivariable model included age, duration of prostitution, average frequency of condom use, age at menarche, sexual intercourse during menses, anal sex, income, and use of a lubricant during sex (Table 3). Sex during menses remained inversely associated with *M. genitalium* infection; women who engaged in intercourse were less likely to have infection than those who did not (OR, 0.2; 95% CI, 0.1 to 0.8). FSW who reported frequent condom use were more likely to be infected with *M. genitalium* than women who reported using a condom half the time or less (OR, 3.2; 95% CI, 1.0 to 9.8). *M. genitalium* was less common among women who worked in prostitution for  $>5$  years than in women who worked for  $<5$  years (OR, 0.4; 95% CI, 0.1 to 1.0).

***M. genitalium* and cervical cytology.** The prevalence of *M. genitalium* was highest in women with severely abnormal cytology results (high-grade squamous intraepithelial lesion [HSIL] and squamous cell carcinoma [SCC]) and atypical cytology results (atypical cells of undetermined significance [ASCUS] and atypical glandular cells of undetermined significance [AGUS]), at 20% and 21.4%, respectively. However, there was no evidence of an association between *M. genitalium* infection and abnormal cytology (OR, 1.1; 95% CI, 0.5 to 2.3), compared to the incidence of *M. genitalium* infection in women with normal cytologies (Table 4).

## DISCUSSION

The prevalence of *M. genitalium* in this population of FSWs was 12.9%, which is higher than the prevalences of *C. trachomatis*, *N. gonorrhoeae*, and *T. vaginalis*. Compared to prevalences in other studies in non-Westernized regions, the prevalence of *M. genitalium* infection was lower than in a population of FSWs in Kenya, Ghana, and Benin (26%) but comparable to that of a high-risk cohort in Japan (12.6%) (16, 18, 19). The variation in *M. genitalium* prevalences across studies may be due to differences in study methods, such as the means of sampling of the source population, user variation in specimen collection, and the detection assays utilized. The age-specific prevalence of *M. genitalium* in this study was comparable to that of other studies among female sex work-

TABLE 3 Multivariable analysis<sup>b</sup> of correlates for *Mycoplasma genitalium*

Parameter	OR (95% CI) <sup>a</sup>
Age (yr)	
18–24	1.0
25–29	0.8 (0.3, 1.8)
30–34	0.5 (0.2, 1.4)
$\geq 35$	0.6 (0.2, 1.7)
Duration of prostitution (yr)	
$<3$	1.0
3–5	1.0 (0.4, 2.3)
$>5$	0.4 (0.1, 1.0)
Income/mo <sup>c</sup>	
$\leq 4,000$ Ksh	1.0
$>4,000$ Ksh	1.6 (0.8, 3.1)
Anal sex	
No	1.0
Yes	0.3 (0.04, 2.7)
Sex during menses	
No	1.0
Yes	0.2 (0.1, 0.8) <sup>d</sup>
Lubricant use during sex	
No	1.0
Yes	0.6 (0.3, 1.2)
Age at menarche (yr)	
$\leq 13$	1.0
14–15	1.7 (0.7, 4.3)
$\geq 15$	1.9 (0.7, 4.8)
Avg frequency of condom use	
Half of the time or less	1.0
Most of the time	3.2 (1.0, 9.8)
Always	2.5 (0.8, 8.7)

<sup>a</sup> OR, odds ratio; 95% CI, 95% confidence intervals.

<sup>b</sup> Multivariable logistic-regression analyses, controlling for all other variables in table.

<sup>c</sup> 4,000 Ksh is equivalent to \$50 U.S.

<sup>d</sup>  $P < 0.05$ .

ers, in whom the prevalence of *M. genitalium* generally decreased as age increased (18, 23).

Women who reported using condoms with clients more than 50% of the time were more likely to be infected with *M. genitalium* than women who reported less frequent condom use. These findings are inconsistent with previous studies showing lower rates of HIV/STI transmission among individuals with more frequent condom use and may reflect potential overreporting of condom use by study participants (41, 42). Comparatively, among female sex workers in Ghana and Benin, women who reported using condoms with all clients had a lower prevalence of *M. genitalium* than women who reported not using a condom with all clients (24% versus 33%,  $P = 0.02$ ) (18). However, similar findings of a positive association between *M. genitalium* prevalence and condom use were observed in a study conducted among young men and women attending an STI clinic (31). Social desirability bias may be the cause of overreported condom use in the study; however, it



**TABLE 4** *Mycoplasma genitalium* as a risk factor for abnormal cervical cytology

Cytology <sup>d</sup>	Overall no.	% infected with <i>Mgen</i> <sup>c</sup>	OR (95% CI) <sup>b</sup>	OR (95% CI) in HPV-positive women <sup>b</sup>
Normal	282	16.2	1.0	1.0
ASCUS/AGUS	14	21.4		
LSIL	38	7.9		
HSIL/SCC	15	20.0	1.1 (0.5, 2.3) <sup>d</sup>	1.1 (0.4, 3.2) <sup>d</sup>

<sup>a</sup> Results of a Pap smear test are indicated as follows: ASCUS, atypical cells of undetermined significance; AGUS, atypical glandular cells of undetermined significance; LSIL, low-grade squamous intraepithelial lesion; HSIL, high-grade squamous intraepithelial lesion; SCC, squamous cell carcinoma (40).

<sup>b</sup> OR, odds ratio; 95% CI, 95% confidence interval.

<sup>c</sup> *Mgen*, *Mycoplasma genitalium*.

<sup>d</sup> The odds ratio represents a comparison between the categories "ASCUS/AGUS," "LSIL," and "HSIL/SCC" combined (collapsed due to sparse data) and the referent, "Normal."

merits further investigation of other mechanisms that may produce contradictory outcomes (43).

We also found that women who engaged in sexual intercourse during menses were less likely to have *M. genitalium* infection (OR, 0.2; 95% CI, 0.1 to 0.7), in contrast to previous research which found that engaging in sexual intercourse during menses increased the risk of STI, including chlamydia, genital herpes, *Trichomonas* infection, and syphilis (44). Previous research has shown a relationship between phases of the menstrual cycle and *M. genitalium* infection; women in the proliferative phase (days 6 to 14) were more likely to have infection, but no women in the menstrual phase (days 1 to 5) in the previous study were infected with *M. genitalium* (9). This may suggest that a biological mechanism connected to the menstrual hormone cycle may have an impact on becoming infected and/or on clearance of infection with *M. genitalium*.

This study is the first to our knowledge to examine a wide spectrum of potential correlates, including HPV infection and abnormal cervical lesions, in a population of female sex workers. The prevalence of *M. genitalium* infection was higher among women with severely abnormal (HSIL/SCC) and mildly abnormal (ASCUS/AGUS) cytologies, though there was no statistical evidence of an association. However, *M. genitalium* may still have a contributory role in the development of cervical neoplasia, via cervicitis and/or HPV infection, as described in other studies (32). While the natural history of *M. genitalium* infection is not yet well understood, it has been positively associated with cervical inflammation, a condition associated with a higher risk of cervical neoplasia (9, 25, 33, 36, 37). This relationship warrants further investigation into the prevalence and persistence of *M. genitalium* infection and cervical outcomes.

This study has several strengths. Data for *M. genitalium* were obtained using the Aptima *Mgen* laboratory assay, which employs TMA-based methods (39). This diagnostic assay has been shown to have a higher clinical sensitivity and specificity for *M. genitalium* detection than culture, which can be highly susceptible to error due to improper specimen handling (45), and to have a sensitivity and specificity for detection of *M. genitalium* similar to those of PCR assays for *M. genitalium* detection (46). The inclusion of HPV and abnormal cervical cytology in the analyses with *M. genitalium* was another unique aspect of this study.

One limitation of our findings is the smaller sample size of the population. As a result, the 95% confidence intervals of OR estimates from analyses were relatively imprecise (confidence limit ratio, >0.25) for several variables (47). Based on previous literature, we expected to observe associations between *M. genitalium* and factors such as age, education, marital status, and sexual-behavioral factors, including number of sexual partners/clients, frequent douching, and hormone contraceptive use (16, 18, 22, 31). However, a larger study sample would likely improve our power to detect correlations with *M. genitalium* and improve the precision of the present findings. Our results were also limited by the cross-sectional study design, as baseline data were assessed from the collection of biological, sociodemographic, and sexual behavioral data. Thus, we lacked the ability to establish temporality, and in the case of coinfections, we were unable to determine whether a participant was infected with *M. genitalium* prior to another pathogen and, consequently, whether the presence of one infection impacted the presence of another infection. Additionally, we collected data on sexual behaviors via self-reporting, which is subject to recall and social desirability bias (43, 48). This may have been the case with condom use frequency and other reported sexual behaviors.

Overall, the results suggest that greater frequency of condom use and duration of prostitution are associated with *M. genitalium* infection. To further elucidate the correlates for *M. genitalium*, longitudinal analyses might produce more-robust results, facilitating a better understanding of *M. genitalium* infection in relation to sexual behavior and other STIs. Targeted public health interventions could then be directed to treating and preventing *M. genitalium* and concomitant reproductive health complications in populations with higher-risk women and low-resource settings.

## ACKNOWLEDGMENTS

We acknowledge the help and support of Craig Hill. We also thank the participants for their contribution to the study.

This study was supported by Hologic Gen-Probe and the UNC Center for AIDS Research (CFAR grant P30-AI50410). Suha Patel was a Howard Hughes Medical Research Fellow from 2009 to 2010. Damon Getman is an employee of Hologic Gen-Probe. Jennifer S. Smith has received unrestricted educational grants, consultancy, and research grants from Hologic Gen-Probe over the past 5 years. The remaining authors have no conflict of interest to declare.

## REFERENCES

- Haggerty CL, Totten PA, Astete SG, Ness RB. 2006. *Mycoplasma genitalium* among women with nongonococcal, nonchlamydial pelvic inflammatory disease. *Infect. Dis. Obstet. Gynecol.* 21:65–69. <http://dx.doi.org/10.1155/IDOG/2006/30184>.
- Jurstrand M, Jensen JS, Magnuson A, Kamwendo F, Fredlund H. 2007. A serological study of the role of *Mycoplasma genitalium* in pelvic inflammatory disease and ectopic pregnancy. *Sex. Transm. Infect.* 83:319. <http://dx.doi.org/10.1136/sti.2007.024752>.
- Moi H, Reinton N, Moghaddam A. 2009. *Mycoplasma genitalium* in women with lower genital tract inflammation. *Sex. Transm. Infect.* 85:10. <http://dx.doi.org/10.1136/sti.2008.032748>.
- Oakeshott P, Hay P, Taylor-Robinson D, Hay S, Dohn B, Kerry S, Jensen JS. 2004. Prevalence of *Mycoplasma genitalium* in early pregnancy and relationship between its presence and pregnancy outcome. *BJOG* 111: 1464–1467. <http://dx.doi.org/10.1111/j.1471-0528.2004.00276.x>.
- Schlicht MJ, Lovrich SD, Sartin JS, Karpinsky P, Callister SM, Agger WA. 2004. High prevalence of genital mycoplasmas among sexually active young adults with urethritis or cervicitis symptoms in La Crosse, Wisconsin. *J. Clin. Microbiol.* 42:4636. <http://dx.doi.org/10.1128/JCM.42.10.4636-4640.2004>.

6. Uuskula A, Kohl PK. 2002. Genital mycoplasmas, including *Mycoplasma genitalium*, as sexually transmitted agents. *Int. J. STD AIDS* 13:79–85. <http://dx.doi.org/10.1258/0956462021924695>.
7. Taylor-Robinson D. 2002. *Mycoplasma genitalium*—an up-date. *Int. J. STD AIDS* 13:145–151. <http://dx.doi.org/10.1258/0956462021924776>.
8. Reference deleted.
9. Manhart LE, Critchlow CW, Holmes KK, Dutro SM, Eschenbach DA, Stevens CE, Totten PA. 2003. Mucopurulent cervicitis and *Mycoplasma genitalium*. *J. Infect. Dis.* 187:650. <http://dx.doi.org/10.1086/367992>.
10. Perez G, Skurnick JH, Denny TN, Stephens R, Kennedy CA, Regivick N, Nahmias A, Lee FK, Lo SC, Wang RYH. 1998. Herpes simplex type II and *Mycoplasma genitalium* as risk factors for heterosexual HIV transmission: report from the heterosexual HIV transmission study. *Int. J. Infect. Dis.* 3:5–11. [http://dx.doi.org/10.1016/S1201-9712\(98\)90088-1](http://dx.doi.org/10.1016/S1201-9712(98)90088-1).
11. Manhart LE, Mostad SB, Baeten JM, Astete SG, Mandaliya K, Totten PA. 2008. High *Mycoplasma genitalium* organism burden is associated with shedding of HIV-1 DNA from the cervix. *J. Infect. Dis.* 197:733. <http://dx.doi.org/10.1086/526501>.
12. Mavedzenge SN, Van Der Pol B, Weiss HA, Kwok C, Mambo F, Chipato T, Van der Straten A, Salata R, Morrison C. 2012. The association between *Mycoplasma genitalium* and HIV-1 acquisition in African women. *AIDS* 26:617–624. <http://dx.doi.org/10.1097/QAD.0b013e32834ff690>.
13. Vandepitte J, Weiss HA, Kyakuwa N, Nakubulwa S, Muller E, Buvé A, Van der Stuyft P, Hayes R, Grosskurth H. 2013. Natural history of *Mycoplasma genitalium* infection in a cohort of female sex workers in Kampala, Uganda. *Sex. Transm. Dis.* 40:422. <http://dx.doi.org/10.1097/OLQ.0b013e31828bfccf>.
14. Mayaud P, Mabey D. 2004. Approaches to the control of sexually transmitted infections in developing countries: old problems and modern challenges. *Sex. Transm. Infect.* 80:174–182. <http://dx.doi.org/10.1136/sti.2002.004101>.
15. Moses S, Muia E, Bradley JE, Nagelkerke NJD, Ngugi EN, Njeru EK, Eldridge G, Olenja J, Wotton K, Plummer FA. 1994. Sexual behaviour in Kenya: implications for sexually transmitted disease transmission and control. *Soc. Sci. Med.* 39:1649–1656. [http://dx.doi.org/10.1016/0277-9536\(94\)90079-5](http://dx.doi.org/10.1016/0277-9536(94)90079-5).
16. Cohen CR, Nosek M, Meier A, Astete SG, Iverson-Cabral S, Mugo NR, Totten PA. 2007. *Mycoplasma genitalium* infection and persistence in a cohort of female sex workers in Nairobi, Kenya. *Sex. Transm. Dis.* 34:274. <http://dx.doi.org/10.1097/01.olq.0000237860.61298.54>.
17. Morency P, Dubois M, Gresenguet G, Frost E, Masse B, Deslandes S, Somse P, Samory A, Mberyo-Yaah F, Pepin J. 2001. Aetiology of urethral discharge in Bangui, Central African Republic. *Sex. Transm. Infect.* 77:125–129. <http://dx.doi.org/10.1136/sti.77.2.125>.
18. Pepin J, Labbe A, Khonde N, Deslandes S, Alary M, Dzokoto A, Asamoah-Adu C, Meda H, Frost E. 2005. *Mycoplasma genitalium*: an organism commonly associated with cervicitis among West African sex workers. *Sex. Transm. Infect.* 81:67. <http://dx.doi.org/10.1136/sti.2003.009100>.
19. Tsunoe H, Tanaka M, Nakayama H, Sano M, Nakamura G, Shin T, Kanayama A, Kobayashi I, Mochida O, Kumazawa J. 2000. High prevalence of *Chlamydia trachomatis*, *Neisseria gonorrhoeae* and *Mycoplasma genitalium* in female commercial sex workers in Japan. *Int. J. STD AIDS* 11:790–794. <http://dx.doi.org/10.1258/0956462001915291>.
20. Uno M, Deguchi T, Komeda H, Hayasaki M, Iida M, Nagatani M, Kawada Y. 1997. *Mycoplasma genitalium* in the cervixes of Japanese women. *Sex. Transm. Dis.* 24:284. <http://dx.doi.org/10.1097/00007435-199705000-00009>.
21. Hancock EB, Manhart LE, Nelson SJ, Kerani R, Wroblewski JKH, Totten PA. 2010. Comprehensive assessment of sociodemographic and behavioral risk factors for *Mycoplasma genitalium* infection in women. *Sex. Transm. Dis.* 37:777–783. <http://dx.doi.org/10.1097/OLQ.0b013e3181e8087e>.
22. Vandepitte J, Muller E, Bukonya J, Nakubulwa S, Kyakuwa N, Buvé A, Weiss H, Hayes R, Grosskurth H. 2012. Prevalence and correlates of *Mycoplasma genitalium* infection among female sex workers in Kampala, Uganda. *J. Infect. Dis.* 205:289–296. <http://dx.doi.org/10.1093/infdis/jir733>.
23. Xiang Z, Yin Y-P, Shi M-Q, Jiang N, Han Y, Wang H-C, Zheng B-J, Liang G-J, Chen X-S. 2012. Risk factors for *Mycoplasma genitalium* infection among female sex workers: a cross-sectional study in two cities in southwest China. *BMC Public Health* 12:414. <http://dx.doi.org/10.1186/1471-2458-12-414>.
24. Andersen B, Sokolowski I, Østergaard L, Kjølhøth Møller J, Olesen F, Jensen JS. 2007. *Mycoplasma genitalium*: prevalence and behavioural risk factors in the general population. *Sex. Transm. Infect.* 83:237. <http://dx.doi.org/10.1136/sti.2006.022970>.
25. Falk L, Fredlund H, Jensen J. 2005. Signs and symptoms of urethritis and cervicitis among women with or without *Mycoplasma genitalium* or *Chlamydia trachomatis* infection. *Sex. Transm. Infect.* 81:73. <http://dx.doi.org/10.1136/sti.2004.010439>.
26. Gaydos C, Maldeis NE, Hardick A, Hardick J, Quinn TC. 2009. *Mycoplasma genitalium* as a contributor to the multiple etiologies of cervicitis in women attending sexually transmitted disease clinics. *Sex. Transm. Dis.* 36:598. <http://dx.doi.org/10.1097/OLQ.0b013e3181b01948>.
27. Huppert JS, Mortensen JE, Reed JL, Kahn JA, Rich KD, Hobbs MM. 2008. *Mycoplasma genitalium* detected by transcription-mediated amplification is associated with *Chlamydia trachomatis* in adolescent women. *Sex. Transm. Dis.* 35:250. <http://dx.doi.org/10.1097/OLQ.0b013e31815abac6>.
28. Simms I, Eastick K, Mallinson H, Thomas K, Gokhale R, Hay P, Herring A, Rogers P. 2003. Associations between *Mycoplasma genitalium*, *Chlamydia trachomatis* and pelvic inflammatory disease. *J. Clin. Pathol.* 56:616–618. <http://dx.doi.org/10.1136/jcp.56.8.616>.
29. Totten PA, Schwartz MA, Sjöström KE, Kenny GE, Handsfield HH, Weiss JB, Whittington WLH. 2001. Association of *Mycoplasma genitalium* with nongonococcal urethritis in heterosexual men. *J. Infect. Dis.* 183:269–276. <http://dx.doi.org/10.1086/317942>.
30. Jensen JS. 2004. *Mycoplasma genitalium*: the aetiological agent of urethritis and other sexually transmitted diseases. *J. Eur. Acad. Dermatol. Venereol.* 18:1–11. <http://dx.doi.org/10.1111/j.1468-3083.2004.00923.x>.
31. Manhart LE, Holmes KK, Hughes JP, Houston LS, Totten PA. 2007. *Mycoplasma genitalium* among young adults in the United States: an emerging sexually transmitted infection. *Am. J. Public Health* 97:1118. <http://dx.doi.org/10.2105/AJPH.2005.074062>.
32. Yin Y-P, Li H-M, Xiang Z, Liang G-J, Shi M-Q, Zhou Y-J, Han Y, Wang G-Q, Wang B, Chen X-S. 2013. Association of sexually transmitted infections with high-risk human papillomavirus types: a survey with 802 female sex workers in China. *Sex. Transm. Dis.* 40:493–495. <http://dx.doi.org/10.1097/OLQ.0b013e31828b32b8>.
33. Shigehara K, Kawaguchi S, Sasagawa T, Furubayashi K, Shimamura M, Maeda Y, Konaka H, Mizokami A, Koh E, Namiki M. 2011. Prevalence of genital *Mycoplasma*, *Ureaplasma*, *Gardnerella*, and human papillomavirus in Japanese men with urethritis, and risk factors for detection of urethral human papillomavirus infection. *J. Infect. Chemother.* 17:487–492. <http://dx.doi.org/10.1007/s10156-010-0203-0>.
34. Walboomers JM, Jacobs MV, Manos MM, Bosch FX, Kummer JA, Shah KV, Snijders PJ, Peto J, Meijer CJ, Munoz N. 1999. Human papillomavirus is a necessary cause of invasive cervical cancer worldwide. *J. Pathol.* 189:12–19. [http://dx.doi.org/10.1002/\(SICI\)1096-9896\(199909\)189:1<12::AID-PATH431>3.0.CO;2-F](http://dx.doi.org/10.1002/(SICI)1096-9896(199909)189:1<12::AID-PATH431>3.0.CO;2-F).
35. Zhang S, Wear DJ, Lo SC. 2000. Mycoplasmal infections alter gene expression in cultured human prostatic and cervical epithelial cells. *FEMS Immunol. Med. Microbiol.* 27:43–50. <http://dx.doi.org/10.1111/j.1574-695X.2000.tb01410.x>.
36. Castle PE, Giuliano AR. 2003. Genital tract infections, cervical inflammation, and antioxidant nutrients, assessing their roles as human papillomavirus cofactors. *JNCI Monogr.* 2003:29–34. <http://dx.doi.org/10.1093/oxfordjournals.jncimonographs.a003478>.
37. Castle PE, Hillier SL, Rabe LK, Hildesheim A, Herrero R, Bratti MC, Sherman ME, Burk RD, Rodriguez AC, Alfaro M. 2001. An association of cervical inflammation with high-grade cervical neoplasia in women infected with oncogenic human papillomavirus (HPV). *Cancer Epidemiol. Biomarkers Prev.* 10:1021.
38. Ting J, Mugo N, Kwatampora J, Hill C, Chitwa M, Patel S, Gakure H, Kimani J, Schoenbach VJ, Poole C. 2013. High-risk human papillomavirus messenger RNA testing in physician-and self-collected specimens for cervical lesion detection in high-risk women, Kenya. *Sex. Transm. Dis.* 40:584–589. <http://dx.doi.org/10.1097/OLQ.0b013e31828e5a91>.
39. Gen-Probe Inc. 2008. Aptima HPV assay package insert. Gen-Probe, Inc, San Diego, CA.
40. Solomon D, Davey D, Kurman R, Moriarty A, O'Connor D, Prey M, Raab S, Sherman M, Wilbur D, Wright T, Jr. 2002. The 2001 Bethesda System. *JAMA* 287:2114–2119. <http://dx.doi.org/10.1001/jama.287.16.2114>.
41. Holmes KK, Levine R, Weaver M. 2004. Effectiveness of condoms in

- preventing sexually transmitted infections. *Bull. World Health Organ.* 82: 454–461.
42. Zenilman JM, Weisman CS, Rompalo AM, Elish N, Upchurch DM, Hook EW, III, Celentano D. 1995. Condom use to prevent incident STDs: the validity of self-reported condom use. *Sex. Transm. Dis.* 22: 15–21. <http://dx.doi.org/10.1097/00007435-199501000-00003>.
  43. Howard GS, Dailey PR. 1979. Response-shift bias: a source of contamination of self-report measures. *J. Appl. Psychol.* 64:144. <http://dx.doi.org/10.1037/0021-9010.64.2.144>.
  44. Tanfer K, Aral SO. 1996. Sexual intercourse during menstruation and self-reported sexually transmitted disease history among women. *Sex. Transm. Dis.* 23:395–401. <http://dx.doi.org/10.1097/00007435-199609000-00009>.
  45. Hardick J, Giles J, Hardick A, Hsieh YH, Quinn T, Gaydos C. 2006. Performance of the Gen-Probe transmission-mediated amplification re- search assay compared to that of a multitarget real-time PCR for *Mycoplasma genitalium* detection. *J. Clin. Microbiol.* 44:1236–1240. <http://dx.doi.org/10.1128/JCM.44.4.1236-1240.2006>.
  46. Wroblewski JKH, Manhart LE, Dickey KA, Hudspeth MK, Totten PA. 2006. Comparison of transcription-mediated amplification and PCR assay results for various genital specimen types for detection of *Mycoplasma genitalium*. *J. Clin. Microbiol.* 44:3306–3312. <http://dx.doi.org/10.1128/JCM.00553-06>.
  47. Poole C. 2001. Low P-values or narrow confidence intervals: which are more durable? *Epidemiology* 12:291–294. <http://dx.doi.org/10.1097/00001648-200105000-00005>.
  48. Catania JA, Gibson DR, Chitwood DD, Coates TJ. 1990. Methodological problems in AIDS behavioral research: influences on measurement error and participation bias in studies of sexual behavior. *Psychol. Bull.* 108:339. <http://dx.doi.org/10.1037/0033-2909.108.3.339>.