

Risk Factors for Prevalent and Incident *Trichomonas vaginalis* Among Women Attending Three Sexually Transmitted Disease Clinics

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Goal: *Trichomonas vaginalis* is the most common nonviral sexually transmitted infection in the United States and may be associated with adverse birth outcomes and may also increase susceptibility to or transmissibility of human immunodeficiency virus. The purpose of this analysis is to describe the epidemiology of *T. vaginalis* in Sexually Transmitted Disease clinics and characterize the risk factors associated with prevalent and incident *T. vaginalis* within the same population.

Methods: We analyzed data from visits occurring during February 1999–December 2001 from 3 sexually transmitted disease clinics in Newark, NJ; Long Beach, CA; and Denver, CO. Data were analyzed from 1462 women aged 15 to 39 years who were tested by culture at their initial visit for *T. vaginalis*, and for 1269 women with at least 1 follow-up visit. Risk factors for prevalent infections at baseline and incident infections among treated or previously uninfected women were assessed.

Results: At baseline, 13.0% of the women had a prevalent infection; risk factors included the following: older age (≥ 20 years), black race, having less than 12 years of education, and having a concurrent chlamydial infection. At follow-up, 4.6% of women had an incident infection; risk factors included the following: older age (35–39 years), black race, having a concurrent chlamydial infection, having had multiple sexual partners in the 3 months before incident infection, and having had *T. vaginalis* at the visit before their incident infection.

Conclusions: *T. vaginalis* incidence is high in women. Risk factors for prevalent and incident infection are similar. *T. vaginalis* was associated with older age in women, unlike other sexually transmitted infections.

TRICHOMONAS VAGINALIS IS ONE of the most common sexually transmitted infections (STI) in the world and thought to be the most common nonviral STI in the United States. There are

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an estimated 7 million new cases of *T. vaginalis* occurring annually in women in the United States, with an estimated prevalence of 3.1%.^{1,2}

Trichomonas infections range from an asymptomatic carrier state to acute inflammatory disease, with up to 50% of infected women being asymptomatic.^{3,4} Asymptomatic *T. vaginalis* infection in women has been associated with adverse birth outcomes, such as premature rupture of the membranes, preterm delivery, and low birth weight, and may also increase susceptibility or transmissibility of human immunodeficiency virus (HIV).^{5,6}

Several studies have identified risk factors and markers for having a *T. vaginalis* infection in women.^{3,7–15} Most of these studies have included a cross-section of a population and measured the prevalence (the number of infections that exist in a population at a given point in time). Very few studies have measured the incidence (the number of new infections). The factors associated with having a prevalent infection may be different from those associated with acquiring an incident infection. For example, women with prevalent infections may be more likely to be asymptomatic because some women with symptomatic infections seek treatment and are cured. Increased risk for a prevalent trichomonas infection in women has been associated with black race, having multiple sexual partners, greater lifetime number of sexual partners, greater years of sexual activity, concomitant sexually transmitted diseases, and both younger and older age.^{3,7–13} Increased risk for an incident trichomonas infection in women has been associated with black race, having multiple sexual partners, and older age.^{14,15} There have been no published studies that evaluated prevalence and incidence of trichomonas infection in the same population of women.

It is still unclear as to whether age is a risk factor for either incident or prevalent trichomonas infection. Both younger age and older age have been found to be associated with prevalent trichomonas infections,^{8–12} whereas older age has been found to be associated with incident trichomonas infection with the highest incidence among women aged 40 to 44 years.¹⁵

The authors thank Lin H. Tian for assistance in developing the application that was useful in analyzing the data. The findings and conclusions in this report are those of the authors and do not necessarily represent the views of the Centers for Disease Control and Prevention.

Supported, in part, by appointment of D.J.H. to the Research Participation Program for the Centers for Disease Control and Prevention, administered by the Oak Ridge Institute for Science and Education.

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Received for publication August 15, 2007, and accepted December 4, 2007.

We analyzed data from an HIV prevention trial that included women seen in sexually transmitted disease (STD) clinics who were tested for *T. vaginalis*, treated, and then followed up for incident trichomonas infections. We identified risk factors for prevalent and incident *T. vaginalis* and compared those risk factors. In addition, we assessed the relationship between age and trichomonas infections.

Materials and Methods

We analyzed data from 1462 women screened for *T. vaginalis* upon enrollment in RESPECT-2, a multicenter, randomized, controlled trial comparing the efficacy of counseling and testing for HIV using 2 different testing methods. The trial took place in 3 STD clinics in Denver, CO; Long Beach, CA; and Newark, NJ. Primary analyses and detailed methodology are described elsewhere.^{16,17} Briefly, eligible clients were those who came to the clinics for a full diagnostic examination for STIs, were HIV negative at enrollment, reported having vaginal or anal sex in the preceding 3 months, and were aged 15 to 39 years. Participants were randomized at enrollment to receive either a rapid HIV test with 2 counseling sessions in 1 visit or a standard HIV test and counseling in 2 visits.^{16,17} Half the participants in each HIV testing group were further randomly assigned to receive an additional booster counseling session 6 months later.^{16,17}

At the baseline visit, women were tested for gonorrhea, chlamydia, *T. vaginalis*, HIV, and syphilis. We did not analyze HIV and syphilis test results for this article because the women were not tested for these infections at each follow-up visit. Tests for gonorrhea and chlamydia were performed by Long Beach and Newark STD clinic laboratories using ligase chain reaction (LCx Uriprobe; Abbott Diagnostics Division, Abbott Park, IL); and the Denver STD clinic laboratory used polymerase chain reaction (Cobas Amplicor CT PCR and Cobas Amplicor GC PCR; Roche Diagnostic Systems, Inc., Branchburg, NJ) initially, but 18 months after the study began changed to strand displacement amplification (BDProbeTec ET CT/GC; BD Diagnostic Systems, Sparks, MD). *Trichomonas vaginalis* culture was done using vaginal swabs by InPouch TV test (BioMed Diagnostics Inc., San Jose, Cal.) or modified Diamond's medium as the culture medium. At follow-up visits, vaginal swabs were self-collected (Denver and Long Beach) or clinician-collected (Newark), depending on local clinic policy. Cultures were maintained for at least 5 days, with specimens examined for growth on Days 1, 2, and 5. Audio computer-assisted self-interview technology was used to collect sexual behavioral history and self-reported symptoms of vaginitis (discharge, pruritis, rash, or redness) at enrollment and at each study follow-up visit. Multiple sexual partners were defined as having ≥ 2 sexual partners in the previous 3 months.

Each woman was scheduled for 4 follow-up visits in 13-week intervals, scheduled 3, 6, 9, and 12 months from the date of enrollment in the study. At each follow-up visit, women were screened for gonorrhea, chlamydia, and *T. vaginalis*. Per clinic protocols, patients with positive test results were treated with Centers for Disease Control recommended treatment regimens. Assessment and treatment of partners was conducted according to local clinic policy but its completeness could not be determined. No active partner notification occurred, but infected women were told to have their partners treated. In Denver, patients were given medication and treatment instructions for their partners. Women who were due for a return follow-up visit were screened for STDs and interviewed if they visited the clinic any time from 1 week before their scheduled visit to 12 weeks after their scheduled visit. Women could return anytime during a 13-week interval if symp-

tomatic, and if they were positive for infection at any of those visits, they were counted as having a positive infection during that interval. A trichomonas infection was defined as having a positive culture result.

Analysis

For this analysis we compared the risk factors for incident trichomonas infections during follow-up to the risk factors for prevalent infections at baseline. All analyses were done using SAS version 9.1 (SAS Institute, Cary, NC).

Baseline trichomonas prevalence was estimated by dividing the number of positive *T. vaginalis* test results by the total number of positive and negative *T. vaginalis* test results. Potential risk factors and markers analyzed included demographic (enrollment site, age, race/ethnicity, years of education, employment status), clinical characteristics (previous STIs, current STIs, and symptoms of vaginitis), and behavioral factors (new sexual partners and number of sexual partners). Potential predictors of prevalent trichomonas infections were identified by univariate odds ratios and 95% confidence intervals. Logistic regression modeling was used to determine factors associated with prevalent trichomonas infections. We assessed interaction between age and behavioral risk factors.

An incident trichomonas infection was estimated by dividing the number of positive *T. vaginalis* test results (positive results had to either be preceded by a negative result or detected more than 14 days after documented treatment with metronidazole) by the total number of positive and negative *T. vaginalis* test results. Women who had a baseline test for trichomonas infection and returned for at least 1 follow-up visit were included in the incident analysis. Potential predictors of incident trichomonas infections were identified by univariate odds ratios and 95% confidence intervals.

Unconditional logistic regression modeling using generalized estimating equations (GEE), which accounted for within-participant correlations of repeated measures, was used to determine factors associated with incident trichomonas infections.¹⁸ We used an independence working correlation matrix, in addition to unstructured, exchangeable, and autoregressive correlation structures. Only the results of the unstructured correlation matrix are shown because all correlation structures had similar final model estimates. This correlation matrix was the least restrictive of the 4 correlation structures; it did not assume all correlations to be equal (assumption with exchangeable), any correlation to be zero (assumption with independence), or that the correlation depends on interval of time between responses (an assumption with autoregressive), as all intervals were 13-weeks apart in our analysis.

Results

Among the 1462 women tested for *T. vaginalis* at baseline, there were 190 with prevalent *T. vaginalis*, for an overall prevalence of 13% (Table 1). Baseline *C. trachomatis* prevalence was 12% and baseline *N. gonorrhoeae* prevalence was 6%. *T. vaginalis* prevalence was highest among women from Newark (19%), women aged 35 to 39 years (18%), black women (20%), women with 12 or less years of education (15%), and women who tested positive for chlamydia (22%) or gonorrhea (23%) at baseline. The strongest predictors for a woman having a prevalent *T. vaginalis* infection at baseline were age 35 to 39 years compared to 15 to 19 years (odds ratio 2.8, 95% CI 1.6–4.9) and black race compared to white race (odds ratio 3.9, 95% CI 2.4–6.5).

Using logistic regression modeling, we found that factors significantly associated with prevalent trichomonas infection included older age (compared with women aged 15–19 years), black race

TABLE 1. *Trichomonas vaginalis* Baseline (Prevalent) and Follow-Up (Incident) Infection Among Women Aged 15–39 Years Participating in Respect-II, 1999–2001

Characteristic	Baseline					Follow-Up				
	N	No. Positive	Prevalence (%)	OR	(95% C.I.)	n*	No. Positive	Incidence per 3-mo Interval (%)	OR	(95% C.I.)
All	1462	190	13.0			3955	180	4.6		
Site of enrollment										
Newark, NJ	498	95	19.1	1.0	—	1043	86	8.2	1.0	—
Denver, CO	594	64	10.8	0.5	(0.4–0.7)	1797	69	3.8	0.4	(0.3–0.6)
Long Beach, CA	370	31	8.4	0.4	(0.3–0.6)	1115	25	2.2	0.3	(0.2–0.4)
Age group (yr)										
15–19	359	26	7.2	1.0	—	918	31	3.4	1.0	—
20–24	461	64	13.9	2.1	(1.3–3.3)	1236	47	3.8	1.1	(0.7–1.8)
25–29	278	37	13.3	2.0	(1.2–3.3)	748	26	3.5	1.0	(0.6–1.8)
30–34	195	33	16.9	2.6	(1.5–4.5)	584	40	6.8	2.1	(1.3–3.4)
35–39	169	30	17.8	2.8	(1.6–4.9)	466	36	7.7	2.4	(1.5–3.9)
Race/ethnicity										
White	307	18	5.9	1.0	—	909	14	1.5	1.0	—
Hispanic	260	17	6.5	1.1	(0.6–2.2)	736	14	1.9	1.2	(0.6–2.6)
Other†	170	13	7.6	1.3	(0.6–2.8)	504	10	2.0	1.3	(0.6–2.9)
Black	725	142	19.6	3.9	(2.4–6.5)	1803	142	7.9	5.5	(3.1–9.5)
Education level										
>12 yr	494	45	9.1	1.0	—	1407	31	2.2	1.0	—
High school diploma or GED	555	78	14.1	1.6	(1.1–2.4)	1441	85	5.9	2.8	(1.8–4.2)
<12 yr	413	67	16.2	1.9	(1.3–2.9)	1104	64	5.8	2.7	(1.8–4.2)
Current employment status										
Student	470	47	10.0	1.0	—	1301	44	3.4	1.0	—
Working	569	76	13.4	1.4	(0.9–2.0)	1586	65	4.1	1.2	(0.8–1.8)
Unemployed	423	67	15.8	1.7	(1.1–2.5)	1065	71	6.7	2.0	(1.4–3.0)
No. sex partners in the previous interval‡										
0	N/A	N/A	N/A			614	26	4.2	1.0	—
1	747	85	11.4	1.0	—	2418	91	3.8	0.9	(0.6–1.4)
2	387	53	13.7	1.2	(0.9–1.8)	600	39	6.5	1.6	(0.9–2.6)
≥3	320	51	15.9	1.5	(1.0–2.2)	303	21	6.9	1.7	(0.9–3.0)
New sex partner in the previous interval										
No	454	54	12.6	1.0	—	2383	80	3.4	1.0	—
Yes	744	105	14.1	0.9	(0.6–1.2)	1552	97	6.3	0.5	(0.4–0.7)
STI in the previous interval:										
Chlamydia										
No	1346	174	12.9	1.0	—	3393	151	4.5	1.0	—
Yes	102	15	14.7	1.2	(0.7–2.1)	229	12	5.2	1.2	(0.7–2.2)
<i>T. vaginalis</i>										
No	1402	183	13.1	1.0	—	3262	119	3.6	1.0	—
Yes	50	6	12.0	0.9	(0.4–2.2)	244	29	11.9	3.6	(2.3–5.5)
Gonorrhea										
No	1401	178	12.7	1.0	—	3527	150	4.3	1.0	—
Yes	43	9	20.9	1.8	(0.9–3.9)	96	11	11.5	2.9	(1.5–5.6)
Had self-reported symptoms of vaginitis§										
No	710	80	11.3	1.0	—	2915	107	3.7	1.0	—
Yes	752	110	14.6	1.4	(1.0–1.8)	664	46	6.9	2.0	(1.4–2.8)
STI diagnosed at visit										
Chlamydia										
No	1268	150	11.8	1.0	—	3685	161	4.4	1.0	—
Yes	181	39	21.5	2.1	1.4–3.0	150	15	10.0	2.4	(1.4–4.2)
Gonorrhea										
No	1355	166	12.3	1.0	—	3754	162	4.3	1.0	—
Yes	97	22	22.7	2.1	(1.3–3.5)	78	13	16.7	4.4	(2.4–8.2)

*Total number of tests in all 4 follow-up intervals.

†Other indicates Native American, Asian, Pacific Islander, Biracial.

‡Total number of tests in all 4 follow-up intervals.

§Symptoms of vaginitis include discharge, pruritis, and rash or redness.

TABLE 2. Logistic Regression and GEE Models for Risk Factors of Prevalent and Incident Trichomonas Infection, Women Aged 15–39 Years Participating in Respect-II, 1999–2001

Characteristic	Baseline (Prevalent Infection) Logistic Regression Model		Follow-Up (Incident Infection) GEE Model	
	Adjusted OR	(95% CI)	Adjusted OR	(95% CI)
Age (15–19)	1.00	—	1.00	—
Age (20–24)	2.70	(1.48–4.94)	1.31	(0.62–2.41)
Age (25–29)	3.06	(1.59–5.91)	1.22	(0.62–2.41)
Age (30–34)	3.49	(1.78–6.84)	1.95	(0.98–3.89)
Age (35–39)	3.73	(1.86–7.46)	2.26	(1.14–4.48)
All sites vs. Newark	1.06	(0.69–1.63)	1.03	(0.66–1.61)
Black women vs. all other race	3.03	(1.90–4.83)	3.31	(2.04–5.37)
<12 yr of education vs. 12 yr or more	1.46	(0.99–2.16)	1.44	(0.95–2.20)
Unemployed	1.05	(0.71–1.54)	1.06	(0.69–1.64)
Chlamydia*	2.37	(1.44–3.90)	2.37	(1.08–5.20)
Gonorrhea*	1.81	(0.96–3.40)	1.62	(0.55–4.75)
Symptoms of vaginitis [†]	1.26	(0.88–1.80)	1.36	(0.87–2.11)
Gonorrhea in previous interval [‡]	2.14	(0.93–4.94)	1.18	(0.49–2.87)
≥2 sex partners in past 3 mo	1.37	(0.93–2.02)	1.71	(1.13–2.60)
New sex partner in the past 3 mo	1.18	(0.79–1.77)	1.17	(0.78–1.74)
Trichomonas infection in the previous interval [‡]	0.44	(0.15–1.30)	3.12	(1.93–5.03)

*Concurrent infection.

[†]Symptoms of vaginitis include discharge, pruritis, and rash or redness.

[‡]At baseline, refers to interview and at follow-up, refers to laboratory diagnosis.

(compared with all other race/ethnicity), and having a concurrent chlamydial infection (Table 2).

During the 3955 follow-up visits by women, there were 180 incident trichomonas infections; an overall incidence of 4.6% per 3-month interval over the 1-year follow-up period (estimated annual incidence of 18%) (Table 1). Similar to our findings for prevalence, incidence was highest among women from Newark (8%), older women 35 to 39 years (8%), black women (8%), women with 12 or less years of education (6%), and women who tested positive for chlamydia (10%) or gonorrhea (17%) in that interval. Risk of incident infection was higher in women having had trichomonas (12%) or gonorrhea (12%) diagnosed during their previous clinic visit. Women who tested positive for trichomonas infection during their previous visit had 3.6 times greater risk of an incident trichomonas infection than women who tested negative for trichomonas infection during the previous visit (95% CI 2.3–5.5). The strongest predictor of a woman having an incident trichomonas infection was black race compared to white race (odds ratio 5.1, 95% CI 3.0–8.8).

Using GEE modeling, and controlling for clinical and behavioral risk variables, independent risk factors and markers for incident trichomonas infection included the following: older age (compared with women aged 15–19 years), black race (compared with all other race/ethnicity), concurrent chlamydial infection, multiple (≥2) sexual partners in the 3 months before incident infection, and having a diagnosed and treated trichomonas infection in the interval before incident infection (Table 2).

Comparison of the multivariate models for risk factors associated with prevalent (logistic regression) and incident infections (GEE) showed some interesting similarities and differences. Factors associated with both prevalent and incident trichomonas infection included the following: older age, black race, and concurrent chlamydial infection. Having multiple (≥2) sexual partners and having had a trichomonas infection in the previous interval were each risk factors for incident trichomonas infection.

Discussion

This study sought to assess and compare risk factors for prevalent trichomonas infections with risk factors for incident trichomonas infections within the same population of women. Few studies have evaluated the incidence of trichomonas infection in women and no other study has compared prevalence and incidence of trichomonas infection within the same population. Both prevalence and incidence of trichomonas infection were high within this population of women; 13% of women had a prevalent infection, and 4.6% of women acquired an incident infection in a 3-month time period (approximately 18.4% for a 1-year incidence). To be included in this study, these women had to come into the STD clinic for an exam, and therefore, we would expect a high prevalence of *T. vaginalis* as well as other STIs—after all, they are coming in because they think they have an STI. During follow-up, this population would be expected to have a higher risk than the general population for acquiring an STI or HIV. However we are following up all women regardless of symptoms and not just testing the women who came back for another clinic visit, and the number of incident infections was still high.

When we compared incidence to prevalence we found that older age was a risk factor for both incident and prevalent trichomonas infection. Women aged 35 to 39 years were more likely to acquire an incident trichomonas infection than woman aged 15 to 19 years; women aged 20 to 39 years were more likely to have a prevalent trichomonas infection than women aged 15 to 19 years. The higher incidence of trichomonas infection in women aged 35 to 39 years in this study is consistent with a previous incidence study that found the overall age-specific incidence of *T. vaginalis* was highest in women aged 30 to 49 years.¹⁵ Four other studies showed the prevalence of *T. vaginalis* to increase monotonically with age among women.^{8–10,19} This association with older age is in contrast to the association of age and other curable STIs. The highest age-specific rates for chlamydia and gonorrhea are among women under 25 years.²⁰ The reason the association of *T. vaginalis* with older age in women exists remains to be determined. Possible

hypotheses include biologic changes in older women that could increase their risk for acquiring a *T. vaginalis* infection or for having persistent infection after standard treatment with metronidazole, or a higher prevalence in older men because of persistence or inadequate response to treatment,²¹ in turn leading to higher prevalence and incidence in older women.

As in previous studies, black race was found to be a significant predictor of both prevalent and incident trichomonas infection. Cotch et al. found the most significant predictor of *T. vaginalis* to be black race, with black women 3.5 times as likely to be colonized as non-black women after adjusting for age, marital status, income, education level, and several indices of sexual behavior.¹³ Although black race was found to be significant in our study, even after adjusting for other risk factors in the model, there is no biologic or physiological explanation for this increase in risk and black race is more likely a surrogate marker for some unknown underlying risk factor. Other factors that were not taken into account, including personal health practices such as douching, and socioeconomic factors such as sexual networks leading to higher prevalence partners, may explain the association of black race and *T. vaginalis*.

Having a concurrent chlamydial infection was a predictor of both prevalent and incident *T. vaginalis* after controlling for other risk factors. A previous study showed that having a chlamydial infection increased the risk of concurrent trichomonas infection in women.¹⁹ Another study found that having a history of other STIs and having a concomitant STI increased the risk of having *T. vaginalis*.¹⁴ Women who are diagnosed with chlamydial infections may be at a high risk for *T. vaginalis* infection as well because of higher risk behaviors in themselves, their partners, or their sexual networks.

In this study, one of the most significant predictors of an incident *T. vaginalis* infection was having had *T. vaginalis* at the previous visit. Although all infections were treated with highly effective single-dose therapy, some of the infections may have been related to treatment failure and others may have been because of lack of partner treatment. In addition, the majority of the new infections were asymptomatic. Without scheduled rescreening those repeat infections would have been missed, and the benefits of rescreening should be investigated further.

The results of this study apply to women in STD clinics. Women who attend STD clinics may be very different from the general population. Because the study wasn't specifically designed to study risk factors associated with trichomonas infection, it did not gather information for all possible confounders to the associations of risk factors and trichomonas infection. Additional information regarding, income, forms of birth control being used, and whether the women douched may have strengthened the study results. Also, there may be misclassification of incident infections because of the low sensitivity of culture, in that some baseline infections may have been missed and then counted as incident infections when detected at follow-up. Misclassification may have also occurred because of a drug resistant infection, which can occur in approximately 5% to 10% of the population,^{22,23} resulting in persistent baseline infections detected at a later visit.

Most of the incident infections were asymptomatic. Risk factors for prevalent and incident infection in women were similar: older age, black race, and concurrent chlamydial infection. In addition, having a trichomonas infection in the previous interval and having multiple (≥ 2) sex partners were risk factors for incident infection. Unlike other STIs, such as gonorrhea and chlamydia, we found that *T. vaginalis* was more often found in older compared with younger women. Understanding why this difference exists is an

important next step. Further investigation should assess the benefits of routinely screening women in STD clinics for *T. vaginalis*.

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