

Identification and Evaluation of *Trichomonas vaginalis* in Wesley Guild Hospital Ilesha, Nigeria

Olaniran Olarinde^{1*}, Osevwe Ayodele.J², Balogun Abiodun.I², Afolabi Olusegun.T³
Hassan-Olajokun Rachel.E¹, Oyetoke Olufemi.O², Awoyeni Ayobami.A¹

1. Department of Medical Microbiology and Parasitology, Obafemi Awolowo University Ile- Ife, Nigeria.
2. Department of Medical Microbiology and Parasitology, Obafemi Awolowo University Teaching Hospital complex, Ile- Ife, Nigeria.
3. Department of Community Health, Faculty of Clinical Sciences College of Health sciences, Obafemi Awolowo University Ile- Ife, Nigeria.

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The Identification and evaluation of *Trichomonas vaginalis* among women was performed over a period of three months in Ilesha. The aim of this study was to determine the prevalence of *Trichomonas vaginalis* in women in relation to different samples collected and level of education. 310 samples from high vaginal, endocervical and urethral regions of female patients were analyzed using Gram staining and direct microscopy. 25 samples (8.1%) were positive for *Trichomonas vaginalis*. Out of the positive samples, 52%, 48% and 0% were collected from high vaginal, endocervical, and urethral regions, respectively. Age group 16-25 years had the highest prevalence: 15 (60%), followed by 26-35 years: 8 (32%), and the least represented age group correspond to 6-15 years: 2 (8%). Less educated women had the highest prevalence of *Trichomonas vaginalis* infection with 15 (60%) cases while 10 (40%) cases were present in educated women. The prevalence of Trichomoniasis in the present study was high, but there was no significant difference in overall distribution of the parasite in high vaginal or endocervical swabs. Therefore, either of the samples can be used for investigating *Trichomonas vaginalis* infection in patients presenting the symptoms.

Keywords: Prevalence, *Trichomonas vaginalis*, high vagina swab, endocervical swab, urethral swab, Nigeria

T*richomonas vaginalis* is an anaerobic flagellate protozoan parasite that causes Trichomoniasis. It is sexually transmitted and exists in trophozoite form only (1). It is oval in shape with five flagella; four of which extend together

immediately outside the cell, while the fifth flagellum wraps backwards along the surface of the organism and serves as the axostyle which may be used for attachment to surface (1). The genus *Trichomonas* has three species which occur in

*Correspondence: Department of Medical Microbiology and Parasitology, Obafemi Awolowo University, Ile-Ife.
E-mail: olarinde.olaniran@yahoo.com

humans; *Trichomonas tenax*, *Trichomonas hominis* and *Trichomonas vaginalis*. *Trichomonas tenax* and *Trichomonas hominis* are nonpathogenic trichomonads while *Trichomonas vaginalis* is pathogenic. *Trichomonas tenax* is found in the tartar of the teeth in the mouth, while *Trichomonas hominis* inhabit the intestine or the caecum of man (2, 3). *Trichomonas vaginalis* is an established cause of sexually transmitted diseases (STDs) as 170 million people are affected worldwide; more than gonorrhea, syphilis and chlamydia combined (1, 4-6). This parasite is the cause of the most parasitic sexually transmitted infections in the world and estimated at 3 million infections in the United States annually (7). The sexually transmitted parasite has become increasingly wide spread over the past several decades in association with relaxation of social taboos, the availability of oral contraceptives; inadequate education of adolescence about the responsibilities associated with sexual intercourse, and a decrease in funding for tracking contact of diagnosed cases (8). *Trichomonas vaginalis* though generally believed to be a female agent can also be detected in men, it is the sole organism detected in the urethral region and seems to affect women more severely than men because the urethra in women is short, about 1.5 inches, compared to 8 inches in males (9) producing symptoms that range from annoying to life threatening (10). The female genital tract plays major role in infection. In women; ovary, fallopian tube and uterus cells can be destroyed by *Trichomonas* infection (11). Greenish-yellow frothy vaginal secretion and itching are the commonest manifestations in women. Infertility, premature rupture of membranes, preterm labour and abortions have also been associated with the parasite (12, 13). The diagnosis is usually obtained by microscopic examination of endocervical swab, urine, high vaginal swab, discharge or prostatic secretion in a drop of fresh physiological saline (14). *Trichomonas*

vaginalis has also been reported to cause pneumonia, bronchitis and oral lesions and is cytopathic to vaginal cells (15, 16). It also induces epithelial monolayer disruption, creating a micro-environment conducive to HIV- 1 replication (17), a phenomenon which could encourage the spread of HIV infection (18-20). Other parasites which are sexually transmitted include: Pubic louse, *Entamoeba histolytica* and *Giardia lamblia*. Also, vaginitis may be due to infection including *Candida species* and *Gardnerella vaginalis* as well as anaerobic bacteria (21). The main objective of this work was to estimate the prevalence of *Trichomonas vaginalis* infection among female patients and investigate its relationship with age and educational background in Ilesha.

Materials and methods

Place of study

The study was conducted over a three months period at Wesley Guild hospital, Ilesha, Osun state, Nigeria. Wesley Guild hospital is an arm of Obafemi Awolowo teaching hospital complex, Ile Ife, Osun state, Nigeria.

Study design and data collection

This was a cross-sectional study design. All females consecutively referred for swab preparation upon gynaecological complaints at the gynaecology clinic of Wesley Guild hospital were serially recruited for the study.

A total of 310 swabs comprising 150 high vaginal, 150 endocervical swabs and 10 urethral discharges were collected after verbal consent was obtained from the patients. Their bio-data were also abstracted into a pro-forma sheet. All experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 declaration of Hel-

Table 1. Overall prevalence of *Trichomonas vaginalis* in high vaginal, endocervical and urethral swab samples collected from patients

Type of Swab	Positive N (%)	Negative N (%)	Total N (%)
High vaginal	13 (4.2)	137 (44.2)	150 (48.4)
Endocervical	12 (3.9)	138 (44.5)	150 (48.4)
Urethral	0 (0.0)	10 (3.2)	10 (3.2)
Total	25 (8.1)	285 (91.9)	310 (100.0)

sinki.

Specimen collection

High vaginal swab: The soft tip of the sterile swab stick was carefully inserted into the lower third of the vagina or about 2 inches past the introitus (the entrance into the vagina) and rotated for 10-30 s, making sure the swab touches the walls of the vagina so that it absorbs the vagina fluid or moisture. The swab was withdrawn without touching the skin and immediately placed into the swab tube. Care was taken that the soft tip was not touched and the swab was placed into the tube in a manner to avoid contamination. The cap was tightened (14).

Endocervical swab: the cervix was reached with the aid of a sterile speculum. Excess mucus from the cervix and surrounding mucosa was removed. Then the swab stick was inserted into the endocervical canal, gently rotated clock wisely for 10-30 s to ensure adequate sampling. It was carefully withdrawn to avoid contact with the vaginal mucosa and placed back into the swab stick tube and recapped tightly. The speculum was washed and sterilized after each use (21).

Urethral swab: the patient should not have urinated for at least one h prior to specimen collection. Area around the urethral opening was cleansed using a swab moistened with sterile physiological saline. Gently the urethra was massaged from above downward. Using a sterile swab stick, the discharged mucus was collected (21).

Samples preparation

About 0.5 ml sterile normal physiological sal-

ine was introduced into each swab container and agitated thoroughly in order to ensure that the vaginal, endocervical or the urethral contents get into the saline. Then, one or two drops from the content were put on a clean grease free slide, covered with a clean grease free cover slip and observed under x10 and x40 objective lens for motile flagellate observation with the condenser and iris being sufficiently closed to give a good contrast.

Giemsa staining

The smear was prepared by emulsifying the swab sample into a drop of sterile normal physiological saline on a clean grease free slide. This was allowed to air dry and was fixed with methanol for about 1 min. Then, the fixed smear was flooded with 10% Giemsa stain for 45 min and washed off with clean water; the back of the slide was wiped clean with dry cotton wool. Afterwards, the slide was placed on a warmer dish to allow drying of the stained smear. Slides were viewed using oil immersion objective lens (x100).

Results

310 samples of high vaginal, endocervical and urethral swabs were obtained from female patients with median age of 27 years with 62% of them being educated (at least secondary education).

A total of 25 (8.1%) samples were positive for *Trichomonas vaginalis* while 285 (91.9%) were negative. Out of the 25 positive samples, 13 (52%) were obtained through high vaginal swab, 12 (48%) A total of 25 (8.1%) samples were positive for

Table 2. Distribution of *Trichomonas vaginalis* in high vaginal, endocervical and urethral swab samples according to age groups

	Age range (years)					Total N (%)
	6-15 N (%)	16-25 N (%)	26-35 N (%)	36-45 N (%)	46-45 N (%)	
High vaginal	1 (0.32)	8 (2.58)	4 (1.29)	0 (0.00)	0 (0.00)	13 (4.2)
Endocervical	1 (0.32)	7 (2.36)	4 (1.29)	0 (0.00)	0 (0.00)	12 (3.9)
Urethral	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Total	2 (0.65)	15 (4.48)	8 (2.60)	0 (0.00)	0 (0.00)	25 (8.1)

Trichomonas vaginalis while 285 (91.9%) were negative. Out of the 25 positive samples, 13 (52%) were obtained through high vaginal swab, 12 (48%) from endocervical swab, while urethral swabs showed no infection (Table 1).

The investigation of *Trichomonas vaginalis* in high vaginal, endocervical, and urethral swabs according to age showed that the 16-25 years age group had the highest occurrence of *Trichomonas vaginalis* with 15 (60%) cases, followed by 26-35 years group with 8 (32%) cases, and 6-15 years group with 2 (8%) cases. The age groups 36-45 years and 46-55 years showed no infection (Table 2).

Table 3 represents the relationship between educational status and prevalence of *T. vaginalis* with 5.2% of educated women being infected compared to 12.7% of uneducated women. This difference was statistically significant ($P= 0.02$).

The prevalence of *Trichomonas vaginalis* obtained from different anatomical regions in patients with different educational status is presented in Table 4. It was revealed that the uneducated patients had the highest infection rate:

Table 3. Prevalence of *Trichomonas vaginalis* according to the educational status of patients

T. vaginalis infection status	Educational status		
	Educated N= 192	Not educated N= 118	Statistic
Positive	10 (5.2)	15 (12.7)	$\chi^2=5.55$ $P= 0.02$
Negative	182 (94.8)	103 (87.3)	

15 (60%), comprising 8 (53.3%) in high vaginal swabs, and 7 (46.7%) in endocervical swabs.

Discussion

Trichomonas vaginalis is one of the major causes of vaginitis, cervicitis and urethritis in humans (9). In this study 310 samples of high vaginal, endocervical, and urethral swabs were collected. *Trichomonas vaginalis* was detected in high vaginal swab and endocervical swab of the subjects while the urethral swab gave a negative result.

It was observed that the age group 16-25 years had the highest occurrence of *Trichomonas vaginalis*, in high vaginal swab 8.0 (32%) and endocervical swab 7.0 (28%), probably because they are a very sexually active group who may not have been referred for performing a test due to the lack of symptoms of infection and also due to the fact that this type of infection is the most common curable STD in sexually active young women (23, 24). The same rate of prevalence of *Trichomonas vaginalis* in

Table 4. Prevalence of *Trichomonas vaginalis* in high vaginal, endocervical and urethral swab samples according to the educational status of patients

Type of Swab	Uneducated N (%)	Educated N (%)	Total N (%)
High vaginal	8 (2.58)	5 (1.61)	13 (4.19)
Endocervical	7 (2.26)	5 (1.61)	12 (3.87)
Urethral	0 (0.00)	0 (0.00)	0 (0.00)
Total	15 (4.84)	10 (3.23)	25 (8.1)

ages 26-35 years: 4.0 (16%) in both high vaginal and endocervical swabs, can be compared with the Hillier's report suggesting that active sexual life of the patients may cause the high rate of Trichomoniasis (24). Patients within 36-45 years and 46-55 years showed no *Trichomonas vaginalis* occurrence for both vaginal and endocervical swabs. This may be associated to reduction in sexual activity of people in this age group and change in vaginal pH to semi acidic which is not conducive for the parasite growth as reported by other studies (18, 25).

Absence of the *Trichomonas vaginalis* in urethral swab in this study may be attributed to the regular flushing of urethral lining by urine and lack of exposure to other routes of infection as also documented by other studies (23, 26, 27).

The effect of educational status on *Trichomonas vaginalis* infection is apparent in this study. This may be attributed to lack of good personal hygiene as documented by other studies which have shown a high correlation between education and hygiene. Jombo and Opajobi documented that *T. vaginalis* infection may be more predominant in populations where sex education is minimal or non-existent (28).

In conclusion, *Trichomonas vaginalis* is fairly common in this population; commoner amongst those newly initiated into sexual activities (16-25 years) and in the uneducated persons. Therefore, awareness creation is vital if gains are to be made in reducing prevalence of *T. vaginalis* infection. Either high vaginal or endocervical swab samples of patients can be collected to demonstrate *Trichomonas vaginalis* infection, because there was no significant difference in the overall distribution of the parasite in the two type of swab samples collected.

Conflict of interest

The authors declared no conflict of interest.

References

1. Schwebke J R, Lawing L F. Improved detection by DNA amplification of *Trichomonas vaginalis* in males. *J Clin Microbiol*. 2002;40:3681-3.
2. Brooks G F, Jawetz E, Melnick J L, et al. *Jawetz, Melnick & Adelberg's medical microbiology*. New York; London: McGraw-Hill Medical ; McGraw-Hill [distributor]; 2013.
3. Arora D R, Arora B. *Trichomonas Vaginalis in Medical Parasitology*. Second Edition ed. New Delhi, Bangalore, India: CBS publishers and distributors; 2007.
4. Shelton J D. Reproductive tract infections and abortion among adolescent girls in rural Nigeria. *Lancet*. 1995;345:870.
5. Crowell A L, Sanders-Lewis K A, Secor W E. In vitro metronidazole and tinidazole activities against metronidazole-resistant strains of *Trichomonas vaginalis*. *Antimicrob Agents Chemother*. 2003;47:1407-9.
6. Quan M. Vaginitis: meeting the clinical challenge. *Clin Cornerstone*. 2000;3:36-47.
7. Secor W E, Meites E, Starr M C, et al. Neglected parasitic infections in the United States: trichomoniasis. *Am J Trop Med Hyg*. 2014 May;90:800-4.
8. Cohen M S. Sexually transmitted diseases enhance HIV transmission: no longer a hypothesis. *Lancet*. 1998;351 Suppl 3:5-7.
9. Nester E W, Roberts C E, Nester M T, et al. *Microbiology : a human perspective*. Dubuque, Iowa: WCB/McGraw-Hill; 1998.
10. Bauer H M, Ting Y, Greer C E, et al. Genital human papillomavirus infection in female university students as determined by a PCR-based method. *JAMA*. 1991;265:472-7.
11. Hook E W, 3rd, Marra C M. Acquired syphilis in adults. *N Engl J Med*. 1992;326:1060-9.
12. Meri T, Jokiranta T S, Suhonen L, et al. Resistance of *Trichomonas vaginalis* to

metronidazole: report of the first three cases from Finland and optimization of in vitro susceptibility testing under various oxygen concentrations. *J Clin Microbiol.* 2000;38:763-7.

13. Soper D. Trichomoniasis: under control or undercontrolled? *Am J Obstet Gynecol.* 2004;190:281-90.

14. Alderete J P, Castella P C. Method and device for trichomonas detection. Google Patents; 2010.

15. Sorvillo F, Smith L, Kerndt P, et al. Trichomonas vaginalis, HIV, and African-Americans. *Emerg Infect Dis.* 2001;7:927-32.

16. Wang C C, McClelland R S, Reilly M, et al. The effect of treatment of vaginal infections on shedding of human immunodeficiency virus type 1. *J Infect Dis.* 2001;183:1017-22.

17. Guenthner P C, Secor W E, Dezzutti C S. Trichomonas vaginalis-induced epithelial monolayer disruption and human immunodeficiency virus type 1 (HIV-1) replication: implications for the sexual transmission of HIV-1. *Infect Immun.* 2005;73:4155-60.

18. Bennett V R, Brown L K, Myles M F. Textbook for midwives. Fourth Edition ed. Edinburgh; New York: Churchill Livingstone; 2000.

19. McClelland R S, Wang C C, Mandaliya K, et al. Treatment of cervicitis is associated with decreased cervical shedding of HIV-1. *AIDS.* 2001;15:105-10.

20. Rendon-Maldonado J, Espinosa-Cantellano M, Soler C, et al. Trichomonas vaginalis: in vitro attachment and internalization of HIV-1 and HIV-1-infected lymphocytes. *J Eukaryot Microbiol.* 2003;50:43-8.

21. Cheesbrough M. District laboratory practice in tropical countries. Cambridge: Cambridge University Press; 2000.

22. Zhang Z-F, Begg C B. Is *Trichomonas vaginalis* a cause of cervical neoplasia? Results from a combined analysis of 24 studies. *International Journal of Epidemiology.* 1994;23:682-90.

23. Center for disease control and prevention. Sexually transmitted disease treatment guidelines. MMWR 55: 11. 2006.

24. Klebanoff M A, Carey J C, Hauth J C, et al. Failure of metronidazole to prevent preterm delivery among pregnant women with asymptomatic *Trichomonas vaginalis* infection. *N Engl J Med.* 2001;345:487-93.

25. Krieger J N, Alderete J F. *Trichomonas vaginalis* and Trichomoniasis in K. Holmes P. Markh, P Sparling et al (Eds). Third edition. ed. McGraw-Hill. New York, 1999.

26. Govender L, Hoosen A A, Moodley J, et al. Bacterial vaginosis and associated infections in pregnancy. *Int J Gynaecol Obstet.* 1996;55:23-8.

27. Kirby H. The structure of the common institutional Trichomoniasis of man. *J Paras.* 2000;31:163-75.

28. Jombo G, Opajobi S. Genital Trichomoniasis Among Women At A University Teaching Hospital: Findings From A Study On 8,443 Clinical Urogenital Samples In Jos, North Central Nigeria. *The Internet Journal of Parasitic Diseases.* 2006;2:Number 1.