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Trichomonas vaginalis infection in a low-risk women attended in Obstetrics and Gynaecology Clinic, Universiti Kebangsaan Malaysia Medical Centre



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ABSTRACT

Objective: To investigate the presence of trichomoniasis among women attending the Obstetrics and Gynaecology Clinic, Universiti Kebangsaan Malaysia Medical Centre.

Methods: A total of 139 high vaginal swabs were taken from the subjects and sent to the laboratory in Amies gel transport media. The specimens were examined for the presence of *Trichomonas vaginalis* using wet mount, Giemsa staining and cultured in Diamond's medium. Sociodemographic characteristics and gynaecological complaints were obtained in private using structured questionnaire applied by one investigator.

Results: The median age was 32 years, with an interquartile interval of 9.96. Most of the subjects were Malays (76.9%) and the remaining were Chinese (15.1%), Indians (2.2%) and other ethnic groups (5.8%). One hundred and thirty eight (99.3%) of the women were married and 98.6% had less than 6 children. More than half (75.5%) of the women's last child birth was less than 6 years ago. Forty seven percent of them were involved in supporting administrative work and 64.7% of the women gave a history of previous or current vaginal discharge.

Conclusions: The present study reported zero incidence rate of trichomoniasis. The low incidence rate was postulated due to all women who participated in this study were categorized into a low-risk group.

1. Introduction

Trichomonas vaginalis (*T. vaginalis*), which is an obligate single-celled protozoan parasite, is the most common curable

sexually transmitted disease (STD) and despite high prevalence rates ranging from 5% to 75% has received little attention globally [1]. It is predominantly spread via unprotected intercourse with an infected partner but might also be spread through the fingers after masturbation [2]. There are several factors that associated with high incidence of infection such as poor personal hygiene, multiple sex partners, low socioeconomic status and underdevelopment [3]. Globally, *T. vaginalis* infection affects approximately 57–180 million people with the majority living in developing countries [2,4].

This infection is strongly related with the existence of other STD such as chlamydia, gonorrhoea and HIV. Previous history of STD is an important sign of trichomoniasis among adolescents and better STD treatment can decrease the incidence of HIV infection [5,6]. *T. vaginalis* infection usually involves the urogenital tract, affecting both female and male of the reproductive age. The common sites of infection are vagina

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The study protocol was performed according to the Helsinki declaration and approved by Ethical Committee of Universiti Kebangsaan Malaysia Medical Centre. Informed consent was sought and obtained from individuals prior to participation in this study.

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and urethra in female and male, respectively. However, most of them may appear asymptomatic especially in male and they become the source of infection [7]. Symptoms typically occur after a period of incubation of 4–28 days. In women, the usual presenting complaint is vaginal discharge (yellowish to greenish in color), which often has a foul smell and is frothy. There may be complaints of itching sensation in the vulva, vagina and medial aspects of the thighs. They may also have urethritis, dyspareunia, cystitis, cervicitis or infection of the Skene's ducts and Bartholin's gland [8].

Trichomoniasis can cause a serious discomfort to women such as adverse pregnancy outcomes, manifested by preterm rupture of membranes, low-birth-weight infants, infertility and cervical cancer. It also increases the transmission of HIV [9]. The Centers for Disease Control and Prevention guidelines recognize that typical diagnosis is made by microscopic examination of wet mount preparations and the measurement of vaginal pH, which has a sensitivity of approximately 60% [10]. When there is a high index of suspicion for trichomoniasis unconfirmed by microscopic examination, culture of vaginal specimens should be performed as this method improves the sensitivity to 95% [11].

In Malaysia, there is a minimal research done to survey the prevalence of *T. vaginalis* infection in a low-risk group or in general population. Most of the researches focused on sub-populations which are known to be at high-risk for contracting and spreading STD such as sex workers and drug abusers. Hence, this study was undertaken to investigate the presence of *T. vaginalis* among women, which were categorized in a low-risk group.

2. Materials and methods

2.1. Study design and study subjects

The study was a cross-sectional report performed from April to May 2012 in the Obstetrics and Gynaecology Clinic, Universiti Kebangsaan Malaysia Medical Centre, Bandar Tun Razak, Kuala Lumpur, Malaysia. A total of 139 reproductive-age women (age 19–49 years) were enrolled randomly when they came for follow up at Obstetrics and Gynaecology Clinic, Universiti Kebangsaan Malaysia Medical Centre. Subjects who were virgin, underage and HIV positive were excluded from this study. Subject information was obtained from participants by use of structured questionnaire. They were interviewed in a confidential setting regarding their sociodemographic characteristics, reproductive history, presenting complaints, history of any vaginal discharge and risk behavior. Each subject voluntarily completed the questionnaire before specimen collection and clinical examination.

2.2. Sampling and sample processing

High vaginal swabs were taken by a well-trained medical officer by scraping at the wall of posterior fornix and immediately placing the swab into the Amies gel transport media. Specimens were labeled with reference numbers and tests were conducted anonymously. The specimens were transported in Amies gel agar which has the ability to maintain viability of *T. vaginalis* up to 24 h and transported to culture laboratory on the same day of sample collection for laboratory assessment [12].

Laboratory trainings were given to all staffs prior to the commencement of the study. The specimens were investigated for the presence of *T. vaginalis* using three different methods which were wet mount, Giemsa staining and cultivation in Diamond's medium. The first swab was used to produce a wet mount after mixing with normal saline onto a microscope slide covered with a cover slip for direct microscopic examination. Motile and dead *T. vaginalis* were observed microscopically at a magnification of 400× and 20 fields. In Giemsa staining, the smears were air-dried and fixed immediately with methyl alcohol prior to staining in order to preserve the morphology of the *T. vaginalis*. The smeared glass slides were reviewed by an experienced laboratory technologist for quality control. A third swab specimen was immediately placed in 5 mL of Diamond's medium which was pre-warmed to room temperature and kept in screw-capped tubes prior to use. Cultures were incubated at 37 °C for 7 days and examined using the wet mount method every day in order to detect *T. vaginalis* motile trophozoites.

2.3. Statistical analysis

The data were analyzed using the SPSS software program for Windows version 20 (Chicago, IL, USA). For descriptive analysis, rate (percentage) was used to describe the prevalence of *T. vaginalis*. *P*-values less than 0.05 were considered significant.

2.4. Ethical approvals

The study protocol (Reference No. UKM 1.5.3.5/244/FF-165-2011) was reviewed and approved by the Ethical Committee of Universiti Kebangsaan Malaysia Medical Centre. Informed consent was sought and obtained from individuals prior to participation in this study.

3. Results

Most of the subjects were Malays (76.9%), Chinese (15.1%), other ethnic groups (5.8%) and finally Indians (2.2%). Thirty one percent of them aged between 26 and 31 years, followed by those aged between 32 and 37 years (29.5%). Most subjects involved in this study were young, with a median age of 32 years (interquartile interval of 9.96).

One hundred and thirty eight (99.3%) subjects were married with a stable sexual partner. Almost all of them (98.6%) have less than 6 children in the family. More than half (75.5%) of the women's last child birth was less than 6 years ago. Forty seven percent of them were involved in supporting administrative work, whereas the other half of the subjects were almost equally categorized into professional work (27.3%) and housewife (25.9%). Among the subjects, 90 (64.7%) of them complained of previous or current vaginal discharge. Forty four subjects with vaginal discharge (48.9%) also complained of vaginal itchiness.

Examination of 139 high vaginal swabs using wet mount and Giemsa staining technique were found negative for *T. vaginalis*, whereby culture of 139 high vaginal swabs on Diamond's medium also revealed no growth.

4. Discussion

Trichomoniasis is a well-known sexually transmitted infection in the world and is considerably prevalent in a high-risk

groups. According to the analysis of National Health and Nutrition Survey (2001–2004), the prevalence of *T. vaginalis* increased with age and greater number of lifetime sex partner. Furthermore, non-Hispanic black ethnicity also posed a higher prevalence of trichomoniasis compared to other racial groups [13]. A national survey which was carried out by the Division of Disease Control, Ministry of Health Malaysia revealed that 0.5% of women and 1% of prostitutes attending antenatal clinic were found positive with *T. vaginalis* [14]. Ramachandran and Ngeow reported the incidence rate of 9.5% among 370 prostitutes and it was ranked fifth most common STD in Malaysia [15]. Another research involving 130 female drug abusers at a rehabilitation center reported 19.2% incidence rate [16]. On the other hand, a recent study conducted at the STD clinic and National Population and Family Development Board clinic reported incidence rate of 0% and 0.36%, respectively [17]. Likewise, the present study reported of zero percent incidence rate which is in agreement with Okonko *et al.* who also reported zero cases of trichomoniasis among patients attending a genitourinary medicine clinic [18]. The low incidence rate and decrease of *T. vaginalis* infection in the current study during the past 20 years may be associated with several factors. Firstly, all women who participated in this study were categorized into a low-risk group, in which most of them were married and had one stable sexual partner. They were either housewives or had secure occupation. Another possibility might be due to the women using vaginal washing and antiseptics after coitus with their partners or the existence of health education programs about STD arranged by the maternal care office.

Despite having symptoms which are suggestive of the presence of trichomoniasis, none of the subjects were found positive with *T. vaginalis*. Thus, vaginal discharge and itchiness reported may be due to infection with candidiasis, bacterial vaginosis or increased vaginal secretion due to pregnancy. Chen *et al.* also revealed that the reported symptoms of vaginal discharge or clinical signs are poor predictors for *T. vaginalis* infection [13]. A study performed by Madhivanan *et al.* disclosed no significant difference in the prevalence among women who complained of abnormal vaginal discharge and those who did not complain of abnormal vaginal discharge with trichomoniasis [19]. Interestingly, a positive result of *T. vaginalis* infection was observed in an incidental finding on a Papanicolaou smear. However, no swab was taken from this subject as there was a breach in the inclusion criteria. The subject was older than the intended reproductive age criteria. This clearly showed that trichomoniasis may occur regardless of age and the possible spread of STD from the sexual partner.

In this study, three methods were used to detect the presence of *T. vaginalis* among the subjects. Wet mount preparation for microscopy is currently the method of choice for diagnosis of trichomoniasis because it is simple, rapid and cheap [20]. However, this method has been reported less sensitive for the diagnosis of the disease, especially in male patients [21]. Giemsa staining make diagnosis more efficient as the samples are concentrated and morphology of the parasite is preserved [22]. Culture of vaginal specimen is the gold standard for the diagnosis of this infection. The sensitivity of this method was reported to be at 95% [22]. Although molecular methods such as PCR are currently being designed, a combination of both wet mount examination and *in-vitro* culture has been recommended as being more sensitive and specific in establishing diagnosis either one alone [23].

In conclusion, the present study reported a zero incidence rate of *T. vaginalis* infection as the prevalence is low in Malaysia. The low incidence rate was expected as all women who participated in this study were categorized into a low-risk group. The symptom of vaginal discharge with or without vulva itchiness does not necessarily suggestive of trichomoniasis. Therefore, a large scale and multi-centred studies should be conducted in order to determine the prevalence of *T. vaginalis* infection in a low-risk group as well as the association of gynecological symptoms with trichomoniasis. More studies are needed to develop and disseminate cheap, sensitive point-of-care diagnostic tests, allowing a greater understanding of the epidemiology of *T. vaginalis* and targeted screening of asymptomatic individuals.

Conflict of interest statement

We declare that we have no conflict of interest.

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