

The Investigation of the Association Between the Frequency of *Trichomonas Vaginalis* and Using Intrauterine Contraceptive Device

Rahim İçi Araç Kullanımı ve Trichomonas Vaginalis Sıklığı Arasındaki İlişkinin Araştırılması

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Objectives: We investigated the association between the frequency of *Trichomonas vaginalis* and using intra-uterine contraceptive device (IUD).

Patients and Methods: A total of 1058 vaginal swab samples were collected. The study population included 402 symptomatic and 656 asymptomatic women, between 18-45 years of age. Specimens were taken with a speculum and sterile cotton swabs from the posterior fornix of the vagina. Wet mount preparations were examined first, and then inoculated into Trypticase-yeast extract-maltose medium.

Results: *T. vaginalis* was determined in 35 (3.31%) samples out of 1058 specimens using both diagnostic methods (direct examination and/or culture). While a total of 402 women were symptomatic, 656 women were asymptomatic. Among 190 IUD users, the frequency of *T. vaginalis* was determined as 6.84%, and among 868 non-users, this ratio was determined as 2.53%. Moreover, while among 100 symptomatic women with IUD, 11 women were positive for *T. vaginalis* (11%), of the 302 symptomatic women without IUD, 17 were positive (5.63%) for *T. vaginalis*. Significant correlation was determined between the use of IUD and the presence of *T. vaginalis* ($p<0.05$).

Conclusion: These results showed that IUD use may increase the growth of *T. vaginalis* in the vaginal mucosa. To reduce the incidence of this pathogen in IUD users, they should be followed up closely.

Key words: *Trichomonas vaginalis*; intrauterine contraceptive device; vaginal infection; symptomatic infection; asymptomatic infection.

Amaç: Bu çalışmada rahim içi araç (RİA) kullanımı ve *Trichomonas vaginalis* sıklığı arasındaki ilişki araştırıldı.

Hastalar ve Yöntemler: Toplam 1058 vajinal sürüntü örneği toplandı. Çalışmaya dahil edilen kadınların 402'si semptomatik, 656'sı asemptomatik olup yaşları 18-45 arasındaydı. Steril eküvyon ile vajina arka forniksinden alınan örnekler, Trypticase-yeast extract-maltose besiyeri içine alınarak direkt bakı ve kültür yöntemi ile incelendi.

Bulgular: Direkt bakı ve kültür yöntemi ile incelenen 1058 örneğin 35'inde (%3.31) *T. vaginalis* saptandı. Kadınların 402'si semptomatik infeksiyonlu iken, 656'sı da asemptomatikti. RİA kullananlar arasında *T. vaginalis* sıklığı %6.84 olarak bulunurken, RİA kullanmayan kadınlar arasında bu oran %2.53 olarak saptandı. Bunun yanında RİA kullanan 100 semptomatik kadın arasında *T. vaginalis* sıklığı %11 olarak bulunurken, RİA kullanmayan 302 semptomatik kadında *T. vaginalis* varlığı %5.63 olarak bulunmuştur. RİA kullanımı ile *T. vaginalis* varlığı arasında anlamlı bir korelasyon olduğu saptanmıştır ($p<0.05$).

Sonuç: Bu sonuçlar RİA kullanımının vajinal mukozada *T. vaginalis*'in üremesini artırdığını göstermektedir. Bu patojenin RİA kullanıcıları arasında ortaya çıkmasını azaltmak için RİA kullanıcıları yakından takip edilmelidir.

Anahtar sözcükler: *Trichomonas vaginalis*; rahim içi araç; vajinal infeksiyon; semptomatik infeksiyon; asemptomatik infeksiyon.

Trichomonas vaginalis (*T. vaginalis*) is a pathogenic protozoon flagellate, affecting 180 million people worldwide. Trichomoniasis is an infection of urogenital tract, infection in women can cause vaginitis, cervicitis, and urethritis. *T. vaginalis* infection is the most prevalent non-viral sexually transmitted disease in the world. Symptomatic women with trichomoniasis usually complain of vaginal discharge, vulvovaginal soreness, and/or irritation.

Dysuria and dyspareunia are also common.^[1,2] *T. vaginalis* can be asymptomatic in 10 to 50% of women.^[3,4] Signs of infection include vaginal discharge (42%), odor (50%), and edema or erythema (22 to 37%). The discharge is classically described as frothy, but it is actually frothy in only about 10% of the patients. Other complaints may include dysuria and lower abdominal pain. Therefore, if these women are not screened, the diagnosis will be missed.^[5] Males are most frequently considered to be asymptomatic carriers which represent an important vector and reservoir.^[5,6] *T. vaginalis* infection is among the most common sexually transmitted infections worldwide.^[7]

There are some important risks factors for spreading of these kind of infections. One of the reported risk factors is the intrauterine contraceptive devices (IUD). Using IUD is one of the most prevalent and effective methods of contraception worldwide. It has been inserted by millions of people yearly. Intrauterine contraceptive devices are highly effective, long-term methods of contraception, however, IUD use is limited due to concerns about the risk for complications linked to its use, particularly pelvic inflammatory disease and subsequent complications.^[1] One of the most common problem among IUD users is genital infections.^[8] In several researches, it was suspected that the presence of an IUD in the uterus may increase the infection risk because of host susceptibility. The projection of part of the device through the cervical canal is thought to allow easy access of vaginal bacteria to the upper genital tract.^[9,10]

Recently, both bacterial and fungal infections have been reported with increasing frequency

in association with IUD use.^[11] But the association between *T. vaginalis* and IUD have not been extensively researched by scientists. Because of the lack of statistical information in this area, in this study, we aimed to investigate the association between trichomoniasis and IUD use.

PATIENTS AND METHODS

This study was carried out as a prospective study from June 2005 - September 2007. A total of 1058 women were randomly selected among gynecologic patients with various complaints who presented to the Gynecology Polyclinic of the Mustafa Kemal University Research Hospital.

This study consisted of 402 symptomatic and 656 asymptomatic women. Symptomatic patients had one or more of the following symptoms: irritating discharge, dyspareunia, dysuria, and lower abdominal discomfort. A total of 190 women were using an IUD.

Informed consent was obtained from each participant woman. The women included the study ranged in age from 18 to 45 years. The duration of IUD use ranged from 1 to 5 years. The protocol was approved by the Human Research Ethics Committee of Mustafa Kemal University.

Vaginal samples were obtained with a vaginal speculum with sterile cotton swabs from the posterior fornix of vagina. All samples were examined by two methods: wet mount preparations and cultivation method.

Wet mount preparations

Vaginal swab samples were obtained after the insertion of the speculum. Each swab sample obtained from the patients was placed in 1 ml of sterile phosphate-buffered saline (PBS), pH 7.2. Fresh preparations were made in sterile PBS. All vaginal samples were examined carefully. Each sample was examined under the light microscope for the presence of *T. vaginalis* by using two objectives (X10, X40). The slides were examined at X10, looking for motile trichomonas, and then at X40 to confirm motility, flagellar movement and morphologic features of the organisms. Negative wet mounts were examined for at least ten minutes.

Table 1. The presenting complaints of the 190 IUD users

| Complaints | Number | (%) |
|--------------------------------|--------|------|
| Discharge | 100 | 52.6 |
| Backache | 94 | 49.5 |
| Pruritus | 89 | 46.8 |
| Vaginal itching | 72 | 37.9 |
| Lower abdominal pain | 64 | 33.7 |
| Dysuria | 51 | 26.8 |
| Menorrhagia | 40 | 21.1 |
| Dysmenorrhea | 23 | 12.1 |
| Dyspareunia | 7 | 3.7 |
| Both pruritus, vaginal itching | 60 | 32 |

Cultivation method

Vaginal samples were taken in 2 ml of trypticase-yeast extract-maltose (TYM) transport medium and these samples were examined at the Parasitology Laboratory of Mustafa Kemal University, Medical Faculty. Wet mount preparation was made of each swab sample after collection. Both microscopically *Trichomonas* negative and positive swab samples were cultured in TYM medium for culture of *T. vaginalis*. The organisms were cultured in vitro at 36 °C in TYM medium, without agar, pH 6.0, supplemented with 10% heat inactivated bovine serum, penicillin (1000 UI ml⁻¹) and streptomycin sulphate (1 mg ml⁻¹). Isolates were subcultured every 48 h in TYM medium.

The cultures were incubated at 36 °C and then wet mount preparations from them were examined at 24 hourly intervals for seven days before they were discarded as negative.

Statistical analysis

The statistical analyses were performed using the Statistical Package for Social Sciences software (SPSS for Windows, version 11.5, Chicago,

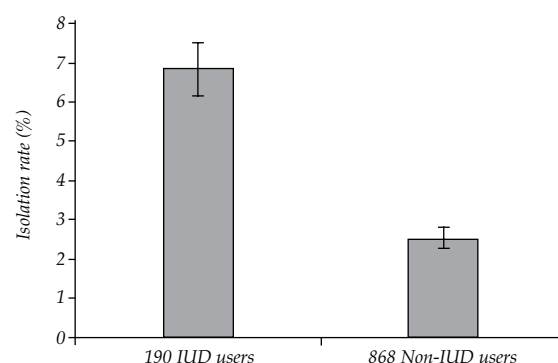


Fig. 1. The isolation rate of *Trichomonas vaginalis* among IUD users and non-users.

USA). Statistical analysis was performed using the chi-square test. $P < 0.05$ was taken as the level of statistical significance.

RESULTS

A total of 1058 women were screened. While a total of 190 women were IUD users, 868 women were non-users. The presenting complaints of the IUD users included vaginal discharge (52.6%), backache (49.5%), pruritus (46.8%), vaginal itching (37.9%), lower abdominal pain (33.7%), dysuria (26.8%), menorrhagia (21.1%), dysmenorrhea (12.1%) and dyspareunia (3.7%). Moreover, a total of 32 (16.8%) women had both pruritus and vaginal itching (Table 1).

The isolation rate of *T. vaginalis* was given in Figure 1. While a total of 402 women were symptomatic, the 656 women asymptomatic. One hundred and ninety of the 1058 women were IUD users. Among IUD users, the frequency of *T. vaginalis* was determined as 6.84% (13/190), and among the non-users, this ratio was determined as 2.53% (22/868). There was statistically significant correlation between the IUD users and the positivity of *T. vaginalis*. ($p < 0.001$, Figure 1).

Table 2. Comparison of sensitivity of wet mount against culture techniques for the examination of vaginal swab samples

| Method | Positive number for <i>T. vaginalis</i> (%) | Negative number for <i>T. vaginalis</i> (%) | Total (%) |
|-----------|---|---|------------|
| Wet mount | 15 (1.42) | 1043 (98.58) | 1058 (100) |
| Culture | 35 (3.31) | 1023 (96.69) | 1058 (100) |

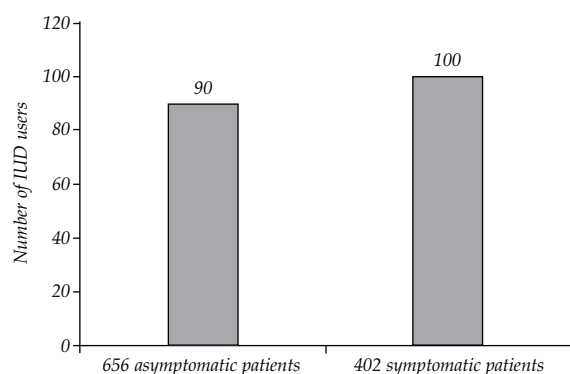


Fig. 2. The IUD use rate among the symptomatic and asymptomatic patients.

A total of 1058 vaginal samples taken from randomly selected women and screened, 35 (3.31%) were positive for trichomoniasis as detected by culture techniques. The direct microscopic examination of wet mount preparation detected only 15 (1.42%) positive cases (Table 2). Twenty of the negative samples and the 15 positive samples under the wet mount examination were found positive by culture method. The cultivated samples were incubated at 37 °C and microscopic examination was made after 24, 48 and 72 hours. For all cultures, positivity was determined maximally by the 72nd hour of incubation.

While *T. vaginalis* was detected in 28 (6.97%, 28/402) patients among the 402 symptomatic patients, seven (1.1%, 7/656) patients were found to be positive in the 656 asymptomatic patients. Results showed that there was a significant difference between the isolation rates of *T. vaginalis* from symptomatic and asymptomatic patients ($p < 0.001$). Of the 1058 women, 190 (17.96%, 190/1058) were IUD users, and the others (868 women; 82.04%) were non-IUD users. The IUD use rate among symptomatic and asymptomatic patients was given in Figure 2.

The women were separated into four groups according to the presence and absence of IUD and *T. vaginalis* (Table 3). Among 190 IUD users, trichomoniasis was detected as positive in 13 women (Group IV), Among this group, trichomoniasis prevalence was found as 6.84%. Twenty-two of 868 non-users (Group II) also had trichomoniasis giving a prevalence of 2.53%. There was a statistically significant difference between two

Table 3. The distribution of women regarding the presence of IUD and *T. vaginalis*

| Groups | Diagnosis | Number | (%) |
|-----------|---|---------|-------|
| Group I | IUD absent, <i>T. vaginalis</i> negative | 846/868 | 97.46 |
| Group II | IUD absent, <i>T. vaginalis</i> positive | 22/868 | 2.53 |
| Group III | IUD present, <i>T. vaginalis</i> negative | 177/190 | 93.16 |
| Group IV | IUD present, <i>T. vaginalis</i> positive | 13/190 | 6.84 |

groups (Groups II and IV) (Table 3).

Moreover, while among the symptomatic women with IUD, the frequency of *T. vaginalis* was 11% (11/100), of the 302 symptomatic women without IUD, 17 were positive (5.63%, 17/302) for *T. vaginalis*. A statistically significant difference was found between these two groups ($p < 0.01$). Similarly, among asymptomatic women with IUD, while *T. vaginalis* positivity rate was detected as 2.22% (2/90), this rate was determined as 0.88% (5/566) among asymptomatic women not wearing an IUD. There was a significant correlation between these two groups and the presence of *T. vaginalis* ($p < 0.05$), (Table 4).

DISCUSSION

Intrauterine contraceptive device is a commonly used birth spacing method which is inserted into the uterus to prevent pregnancy. In several studies, it was reported that using these devices are important for ascertaining their side effects, the risk of genital tract infection.^[12,13]

Table 4. The positivity rates of *T. vaginalis* among symptomatic and asymptomatic women

| Diagnosis | Number | (%) |
|---------------------------------|--------|------|
| Symptomatic women, IUD present | 11/100 | 11 |
| Symptomatic women, IUD absent | 17/302 | 5.63 |
| Asymptomatic women, IUD present | 2/90 | 2.22 |
| Asymptomatic women, IUD absent | 5/566 | 0.88 |

Using IUD has been demonstrated to be a significant risk for microbial colonization and infections. The IUD is an effective contraceptive method, but its use has been associated with increased incidence of various vaginal infections. Recently *T. vaginalis* infections have been reported with increasing frequency in association with IUD use.^[13-16]

T. vaginalis is a sexually transmitted disease of worldwide importance. Trichomoniasis has been associated with various morbidities such as vaginitis, cervicitis, urethritis and pelvic inflammatory disease.^[17] In this study, we employed wet mount smear examination and culture method for the detection of *T. vaginalis* in vaginal samples. The method of wet mount smear examination has some limitations. In this method, only motile organisms can be detected.^[18]

Culture method to detect of *T. vaginalis* is the most sensitive method, especially in asymptomatic cases.^[19] For this reason, we preferred to use the culture method in addition to the wet mount smear examination for the detection of the association between the presence of *T. vaginalis* and IUD.

In the present study, when the microscopic and cultivation method were compared for the diagnosis of *T. vaginalis*, our results showed that better performance was obtained by using culture method ($p < 0.05$) (Table 2).

The association between *T. vaginalis* and IUD use was mentioned in limited previously published studies.^[20] It has been demonstrated in a few studies that the infection prevalence of *T. vaginalis* is higher in IUD users compared with non-IUD users.

Determination of *T. vaginalis* in various risk groups in the society plays a significant role in taking the necessary precautions against this disease.^[13,20]

In this study, we investigated the incidence of *T. vaginalis* infection among the women using IUD. While the frequency of *T. vaginalis* was 6.84% (13/190) in IUD users, this rate was 2.53% (22/868) among the non-IUD users. We found a highly significant difference in prevalence of

T. vaginalis between women using an IUD and non-users ($p < 0.05$). We think that these findings may be due to IUD, and susceptible to infections.

It is known that infection has been one of the main concerns in IUD use. The use of IUD results in quantitative changes in vaginal microbial flora. In this study, significant increase of the trichomonal growing was determined. Similarly, Agarwal et al.^[13] also found significant increase in the frequency of *T. vaginalis*.

Studies on the prevalence of *T. vaginalis* infection among IUD users are limited in our country. In a study conducted by Demirezen^[21] in 2001, while 46 (4.32%) of 1065 non-IUD users were seropositive for *T. vaginalis*, 10 (22.22%) of 45 women were positive for *T. vaginalis* in IUD users. The difference was found to be significant ($p < 0.05$).

Similarly, Nasir et al.^[20] investigated the association between the use of an IUD and the presence of *T. vaginalis* in vaginal smears. Among 45 IUD users, while the presence of *T. vaginalis* was determined in 15 women (33.33%), among non-users, this rate was 5.56% (42/755). A significant correlation between the use of IUD and the presence of *T. vaginalis* was also found ($p < 0.05$). In our study, *T. vaginalis* was found 6.84% in the IUD users. Our results are quite low in terms of the seroprevalence of *T. vaginalis* compared to that reported by Demirezen^[21] and Nasir et al.^[20]

In another study carried out in 2006, Demirezen et al.^[14] investigated whether there was an association between bacterial vaginosis and IUD use. They found a significant correlation between the use of IUD and the presence of bacterial vaginosis ($p < 0.05$) alike the relation between the presence of *T. vaginalis* and IUD.

It is reported in the literature that the chance of microbial colonization increases with the use of an IUD. It is found that there appears to be a direct relationship between IUD use and the development of microbial infection.^[13,14] In our study, we found a significant increase in the

colonization of *T. vaginalis* among IUD users which is consistent with the literature.

In several studies, some authors suggest that microbial colonization increases with the duration of IUD use. A direct relationship was shown between the duration of IUD use and the development of microbial infection.^[22,23] On the contrary, Kalaichelvan et al.^[16] found no significant association between the duration of IUD use and the microbial colonization in vaginal smears in their study. Similarly, we found no significant association between the duration of IUD use and *T. vaginalis* incidence in vaginal swabs.

In conclusion, we determined that the rate of infection by *T. vaginalis* was prevalent among IUD users. The use of IUD poses risk for *T. vaginalis* infection. In addition, it is recommended that IUD users who are susceptible to *T. vaginalis* infections should be identified by both wet mount smear examination and culture method for detection of *T. vaginalis* in vaginal samples. Therefore, patients using the IUD should be screened for *T. vaginalis* after starting using IUD to prevent this infection. Hence, women who use an IUD require a regular gynecologic follow-up, clinical examination, counseling and further investigation if required.

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