Original Article

Investigation of the frequency and relationship between trichomonas infection in the preterm delivery (a case-control study in Amir Al-Momenin Hospital, Semnan)

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Abstract

Introduction and Purpose: Preterm delivery is a common complication during pregnancy periods and imposes a high cost on the healthcare system due to the care needs of premature babies. Sexually transmitted infections are one of the effective factors in the occurrence of preterm delivery and the diagnosis and treatment of these infections are effective in reducing complications and preventing preterm delivery. In this study, the role of *Trichomonas vaginalis* (*T. vaginalis* [TV]) infection in preterm delivery has been evaluated. **Methods:** In a prospective case–control study, women with preterm birth were assigned to the case group, and women with full-term delivery on the same day were also assigned randomly to the control group. After receiving the history and physical examination, a sample was taken from the cervix for TV culture. The data were included in the SPSS version 23 software. A significance level of less than 0.05 was considered. **Findings:** The overall prevalence of this infection was 10%. The prevalence of chlamydial infection was 2% among mothers with full-term delivery and 16.4% among mothers with premature birth, and there was a significant difference between the two groups ($P = 0.021$). The logistic regression analysis to determine the effect of *Trichomonas* infection on premature birth showed that there was the probability of the occurrence of premature delivery increases in mothers with trichomoniasis infection with lower age, higher body mass index, the presence of underlying disease, lower educational level, housewives, lower parity and gravity and having a history of fetus abortion more than 13 times with its occurrence probability occurs in mothers without *Trichomonas* infection ($P = 0.046$, Exp (β) =13.266). **Conclusion:** According to the present results, TV screening for pregnant women, especially in high-risk groups, is emphasized to reduce the incidence of preterm delivery and related complications, especially neonatal complications.

Keywords: Pregnancy complications, preterm delivery, *T. vaginalis*

Introduction

The preterm delivery or birth before the end of the 37th week of pregnancy has a global prevalence of 10.6%, which varies from 8.7% to 13.4% in different regions.[1] Preterm delivery can be considered a global problem despite the fact that more than 60% of premature birth occurs in Africa and South Asia.[2] The complications of preterm delivery are the main cause of mortality for children under 5 years of age, and despite the advancement of knowledge about the risk factors and mechanisms related to preterm delivery, the rate of preterm delivery and its burden are increasing in most developed countries.[2-4] In one of the studies, it

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Received: 25-08-2023 **Revised:** 13-11-2023
Accepted: 05-12-2023 **Published:** 22-04-2024

Access this article online
Quick Response Code:
Website: http://journals.lww.com/JFMPC
DOI: 10.4103/jfmpc.jfmpc_1411_23

How to cite this article: Saffarieh E, Tarahomi S, Pazoki S, Khaleghian A. Investigation of the frequency and relationship between trichomonas infection in the preterm delivery (a case-control study in Amir Al-Momenin Hospital, Semnan). J Family Med Prim Care 2024;13:1362-70.

has been reported that the cost of healthcare services for children born under preterm delivery up to the age of 10 years is equal to 2.17 to 6.55 times of a child born under full-term delivery.[5] The cause of infant mortality in preterm delivery is 10% of cases, and preterm delivery is the second most common cause of death in children under 5 years after pneumonia.[6,7] Also, almost 75% of perinatal deaths are due to preterm delivery.[8,9] Other complications caused by preterm delivery include respiratory distress syndrome and chronic lung disease,[10] cerebral palsy, neurological disorders and sensory-motor disability, heart defects, increased risk of cardiovascular diseases, diabetes during puberty periods, necrotizing enterocolitis, visual impairment and hearing, patent ductus, asphyxia, retinopathy, bronchopulmonary dysplasia, bleeding and intracranial infection, sudden death syndrome, long-term disability in growth and development processes,[11] as well as cognitive, behavioral and social problems in adulthood and weak academic progress.[12] One of the unknown dimensions of preterm delivery is physiology and molecular biology.[13] A combination role of genetic and environmental factors in premature birth is probable. Some of these factors include non-white skin race, short intervals between pregnancies, physical or psychological stress of the mother and excessive weight loss, some diseases, such as bacterial vaginosis, intrauterine infections, non-feminine infections, abdominal surgery in the mother, endocrine disorders of the mother, multiple births, shortness of cervix length (less than 3 cm), smoking use, uterine abnormalities and placenta previa, uterine abnormalities, placental abruption, excessive enlargement of the uterus, for example in multiple births, increased amniotic fluid, diabetes, gingivitis (severe gum infection), lupus, severe asthma, hepatitis, intestinal diseases, kidney infection, lung infection, abdominal trauma, abdominal surgery during pregnancy periods, age less than 18 year or more than 35 years, height shortening, previous history of preterm delivery, vaginal bleeding in more than one trimester, genetic factors, hard and long-term work play a role in the onset of labor and preterm delivery.[8,14-16] The most important infection affecting fetus abortion and preterm delivery is urinary–genital tract infection whose contribution among the infections affecting the incidence of preterm delivery reaches 40%. The role of microorganisms such as *Mycoplasma urealyticum*, *Mycoplasma hominis*, *Neisseria*, and *Chlamydia trachomatis* have been emphasized in many studies and some studies have also implied *T. vaginalis* (TV).[17-21] TV infection is the most common non-viral sexually transmitted infection (STI) worldwide. Its prevalence rate is varied among different ethnicities and races. There is conflicting evidence of a relationship between trichomoniasis and perinatal adverse outcomes. In one of the studies, it has been reported that the risk of premature birth increases by about 30% in people with TV infection.[22] This organism is highly resistant to the drugs and 5-nitroimidazole remains as the only treatment option.[23] According to the overall prevalence of preterm delivery in Iran, which has reported to equal to 2.9%[24] and the high cost which is spent on caring for premature babies and the impact of infectious agents, and the importance of preventing preterm delivery due to its costly consequences; in this study, we will evaluate the role of infectious agents, specifically TV infection in preterm delivery. It is clear that the results obtained in this study will be effective in reducing maternal and neonatal complications and adopting necessary and timely measures to prevent preterm delivery.

Materials and Methods

In a prospective case–control study, pregnant mothers referred to the Amir al-Momenin Hospital in Semnan were selected using a convenient sampling approach. Mothers with premature birth were assigned to the case group and mothers with full-term delivery on the same day were assigned to the control group. The case group consisted of women who were referred with preterm delivery pain before the 37th week of pregnancy according to the definition of full-term labor and did not have the exclusion criteria. The control group included healthy women who met the inclusion criteria and whose gestational age was more than 37 weeks. Mothers were randomly included in the study. The inclusion criteria included maternal age from 15 to 45 years and body mass index (BMI) from 20 to 35 kg/m^2. Mothers with multiple pregnancies, intrauterine death, preeclampsia, severe bleeding, bacterial vaginosis and uterine anomaly were excluded from the study. Data collection was performed using a checklist including demographic information and clinical information. The presence of chlamydial infection was determined through specialized diagnostic tests (polymerase chain reaction [PCR]). Before inclusion in the study, by explaining the objectives of the study to the subjects, informed consent was received from them. First, information related to age, occupation, education, weight, height, previous marriage, and history of pregnancy is performed. Then, the mother's underlying diseases including diabetes, high blood pressure, autoimmune disease, and neurological and mental diseases were recorded. Other information, including bleeding, smoking, and history of previous pregnancy with an emphasis on premature birth was examined and recorded. Vaginal examination, dilatation, effusion, amniotic fluid drainage, bleeding, vaginitis and cervicitis were recorded. Then, the patient was examined using a speculum and in case of the presence of cervicitis, the findings were recorded. In case of the presence of bleeding, membrane rupture or bacterial vaginosis, the intended case was excluded from the study. In the case of the presence of erythema, congestion, edema, and mucopurulent secretions from the endocervix and cervicitis were recorded. A sample was taken from the cervix to examine the presence of *T. vaginalis* and sent to the laboratory for culture and PCR. Before inclusion in the study, by explaining the objectives of the study to the subjects, informed consent was included in the study. First, questions related to age, occupation, education, weight, height, previous marriage, and history of pregnancy were taken. The mother's underlying diseases including diabetes, high blood pressure, autoimmune disease and neurological and mental diseases were recorded. Other information, including bleeding, hydraminos, uterine anomaly, fetal anomaly, multiple births and smoking, as well as previous pregnancy history with an emphasis on premature birth were examined and recorded. The sampling was under sterile conditions. The method of

sampling and performing PCR was as follows: After placing a sterile speculum, a Dacron swab (Delta lab, Spain) after four times rotation in the endocervix, in the specific medium of TV (sucrose phosphate buffer, 0.2 M or 2SP at pH = 7.2 with 10% fetal bovine serum and antibiotics (50 μg/mL streptomycin, 100 μg/mL vancomycin, 10 μg/mL gentamicin) (Sigma, Germany) were added. All samples were transferred to the Biochemistry Research Laboratory of the Medical School of Semnan City by following the principles of cold chain. The samples were kept at −75°C until DNA extraction. The DNA extraction was performed with the Accuprep genomic DNA kit (Bioneer Company, Korea), and in this way, about 20 μL of protease K and 200 μL of binding buffer were added to the sample. After mixing, it was incubated at 60°C for 20 min. Then, about 100 μL of isopropanol was added. This mixture was centrifuged at 8000 rpm for 1 min. Washing buffers 1 and 2 (500 μL) were added to the microtubes and after centrifugation, 200 μL of elution buffer was added and finally, it was placed at 8000 rpm for 1 min. The extracted DNA was kept at −20°C until performing PCR. The primers used in the PCR test to detect the 18 s rRNA gene of TV had the following sequence which as follow:

Forward primer: 5'-TAATGGCAGAATCTTTGGAC-3'

Reverse primer: 5'-GAACTTTAACCGAAGGACTTC-3'

The first round of PCR was carried out in the following method: From 10X PCR buffer (Fermentas, Germany) with the amount of 5 μL, (50 μm) $MgCl_2$ (Fermentas) with an amount of 0.75 μL, (10 mμ) dNTP with the amount of 1 μL, primers with a concentration of 10 pmol (Bioneer, Korea) with an amount of 1.75 μL, HOT Taq DNA polymerase (5 U/mL) with an amount of 0.3 μL and DNA (300 ng/mL) with an amount of 7 μL and deionized water, were used. In the Master Mix, the second round is the same as the first round, with this difference that the amount of DNA was added to the materials with the amount of 4 μL. The implementation program for the first round was this way after placing the microtubes in a thermocycler (Eppendorf, Germany) with a pre-denaturation program of 95 degrees Celsius for 3 min, 94°C for 30 s, annealing at 58°C for 40 s and synthesis at 67°C for 40 s was set in 35 cycles. The nested PCR for the second round was in this way that the amplification at 95°C for 30 min and respectively denaturation at 94°C for 30 s, annealing at 56°C for 40 s and 67°C for 40 s for 35 cycles and 72°C for 5 min for final synthesis was planned and regulated. The final PCR product was electrophoresed in 2% agarose gel and was placed with ethidium bromide in the gel documentation device (Vilber journal, France) and then a band of about 453 bp was observed.

Data analysis

The comparison of the basic and numerical dependent variables in the two groups was applied after confirming the assumptions of normality and equality of variances using the *t*-test, otherwise, the non-parametric Mann–Whitney test was used. The qualitative variables are reported as values and percentages. If necessary and possible, the qualitative variables were compared in two groups using Fisher's exact test. The level of confidence in all tests was considered 95% and in the normality verification test (Kolmogorov–Smirnov) equal to 99%. Also, logistic regression was used to investigate the effect of chlamydial infection on the incidence of preterm delivery. The used software was SPSS version 23 software.

Ethical considerations

This study was performed after the approval of the proposal in the Research Council of Abnormal Uterine Bleeding Research Center of Amir al-Momenin Hospital in Semnan and the University Research Council, and finally, receiving the code of ethics from the University Ethics Committee with the code number: IR.SEMUMS.REC.2020.163. It is worth mentioning that during completing the checklists, the principles of confidentiality and trustworthiness were observed.

Findings

In this study, the frequency determination of trichomoniasis infection was examined in two groups of case and control (mothers with preterm and full-term delivery) to investigate fetal and maternal affecting factors and their effect on preterm delivery. The average age in the case group was equal to 29.83 ± 5.7 years and the control group was equal to 27.59 ± 5.6 years. The average BMI in the case group was equal to 25.24 ± 3.9 and the control group was equal to 25.77 ± 3.7 kg/m^2. There was no significant difference between the two groups in terms of average age and body mass index (P = 0.083 and P = 0.592, respectively). The studied mothers in the case group (preterm delivery) and the control group (full-term delivery) had no significant difference in terms of occupation, education and underlying diseases [Table 1].

The studied mothers in the case (preterm labour) and control (full-term delivery) groups did not have fetal anomalies in any case, and only one of the mothers had a history of stillbirth, which was not significant (P = 0.555). None of the studied patients had a uterine anomaly, multiple births, intrauterine infection, uterine bleeding, cervical infection or hydraminos. The presence of cervical dilatation had no significant difference in the two groups (P = 0.379). Gravity, parity, live birth and history of abortion did not significantly differ between the two groups [Table 2].

The main objective was to determine the frequency of trichomoniasis infection in two groups of mothers with full-term and preterm delivery and the relationship of this infection with preterm delivery. The frequency of trichomoniasis infection and the comparison of its frequency between two cohorts of mothers with and without preterm delivery are shown in Table 3. The frequency of trichomoniasis infection had a significant

Table 1: Frequency comparison of demographic and background characteristics in studied mothers

Background feature	Preterm (*n*=49)		Full-term (*n*=61)		Total (*n*=110)		*P*
	Number	Percentage	Number	Percentage	Number	Percentage	
Education level							0.379
Illiterate	17	38.6	27	61.4	44	100	
Less than diploma	14	42.4	19	57.6	33	100	
Diploma and above	18	54.5	15	45.5	33	100	
Occupation							0.136
Housekeeper	43	42/1	59	57/9	102	100	
Employee	6	0.75	2	0.25	8	100	
Underlying disease							
Without disease	38	43.7	49	56.3	87	100	0.487
Diabetes	4	0.05	4	0.50	8	100	0.550
Diabetes + blood pressure	0	0	1	100	1	100	0.850
Diabetes + hypothyroidism	4	0.80	1	0/20	5	100	
Hepatitis	0	0	1	100	1	100	
Blood pressure	0	0	2	100	2	100	
Hypothyroid	3	0.05	3	0.50	6	100	

Table 2: Frequency comparison of pregnancy records characteristics in studied mothers

Background feature	Preterm (*n*=49)		Full-term (*n*=61)		Total (*n*=110)		*P*
	Number	Percentage	Number	Percentage	Number	Percentage	
Gravity							
1	16	33.3	32	66.7	48	100	0.069
2	20	62.5	12	37.5	32	100	
3	6	37.5	10	62.5	16	100	
4 and more	7	50.0	7	50.0	14	100	
Parity							
0	22	37.9	36	62.1	58	100	0.273
1	17	58.6	12	41.4	29	100	
2	7	50.0	7	50.0	14	100	
3 and more	3	33.3	6	66.7	9	100	
Live birth							
0	22	37.9	36	62.1	58	100	0.113
1	20	62.5	12	37.5	32	100	
2	4	36.3	7	63.7	11	100	
3 and more	3	33.3	6	66.7	9	100	
Fetus abortion							
0	38	43.1	50	56.9	88	100	0.707
1	7	43.8	9	56.2	16	100	
2	3	60.0	2	40.0	5	100	
3	1	100	0	0	1	100	

Table 3: Frequency comparison of *Trichomonas* infection in the studied mothers

Background feature	Full-term (*n*=49)		Preterm (*n*=61)		Total (*n*=110)		*P*
	Number	Percentage	Number	Percentage	Number	Percentage	
Trichomonas infection							
No	48	0.98	53	9.86	101	8.91	0.035
Yes	1	0/2	8	1/13	9	2/8	
Total	49	100	61	100	110	100	

difference between the two groups (P = 0.035). The overall prevalence of this infection was equal to 8.2%. The prevalence of trichomoniasis infection among mothers with full-term delivery was 2% and among mothers with preterm delivery was equal to 13.1%.

The results of logistic regression analysis [Table 4] showed that the probability of preterm delivery in mothers with trichomoniasis infection increases significantly compared to mothers without chlamydia infection (P = 0.035, t = 2.130).

To more accurately determine the effect of *Trichomonas* infection on preterm delivery and the effect of other maternal factors, multivariate logistic regression analysis was performed [Table 5]. The results of the regression analysis showed that the probability of incidence of preterm delivery occurs in mothers with *Trichomonas* infection with lower age, higher BMI, presence of underlying disease, higher education, working women and parity, lower gravidity and live birth and having a history of abortion more than 2.5 times of its probability of incidence in mothers without *Trichomonas* infection ($P = 0.009$). In the initial model, the relationship between *Trichomonas* infection with preterm delivery and working people was significant, and the same relationship was also significant in the final model.

Discussion

In this study, the number of 110 mothers in the case group (preterm delivery, 61 people) and the control group (full-term delivery, 49 people) were examined in terms of the effect of chlamydial infection on the incidence of preterm delivery. The main objective of this study was to determine the frequency of trichomoniasis infection in two groups of mothers with full-term delivery and preterm delivery and the relationship of this infection with preterm delivery which there was a significant difference between the two groups ($P = 0.035$). The overall prevalence of this infection was equal to 8.2%. The prevalence of trichomoniasis infection among mothers with full-term delivery was 0.2% and among mothers with preterm delivery was equal to 13.1%. The results of logistic regression analysis showed that the probability of the incidence of preterm delivery in mothers with trichomoniasis infection was more than seven times the probability of its incidence in mothers without trichomoniasis infection ($P = 0.035$, odds ratio [OR] = 7.245). The results of the regression analysis showed that the The results of regression analysis showed that the incidence of premature birth in mothers with trichomoniasis infection was related to age, BMI, underlying disease, higher education, being employed, having a history of abortion more than 2 times, and live birth.

In the initial model, there was a significant relationship between the *Trichomonas* infection with preterm delivery and working people and the same relationship was also significant in the final model. Preterm delivery is one of the most important complications of pregnancy periods, which is the result of premature birth and occurs under the influence of various causes. The most important causes of preterm delivery include stress, infection, placental abruption, placenta previa, medication use, history of abortion, inadequate prenatal care, smoking use, maternal age less than 18 or more than 40 years, improper nutrition, low BMI, fetal abnormal, fetal growth restriction, oligohydramnios, polyhydramnios, vaginal bleeding, premature rupture of membranes (PPROM) and environmental factors.[25] Shoja *et al.* (2011) in their study, have considered multiple births, premature rupture of the amniotic sac, preeclampsia, baby breech

Table 4: Regression analysis estimation of the probability of incidence of preterm delivery in the presence of trichomonas infection

	β	Standard error	Beta standard coefficient	*P*	*t*	95%CI for (β)	
						Lower limit	Upper limit
Incidence of preterm delivery	0.020	0.039	0.201	0.035	2.130	0.008	0.214
Constant	0.111	0.052	1	0.559	0.527	0.056	0.097

Table 5: Regression analysis estimation of the effect of background factors and pregnancy on the incidence of preterm delivery in the presence of *Trichomonas* infection

	β	Standard error	Beta standard coefficient	*P*	*t*	95%CI for (β)	
						Lower limit	Upper limit
Complete model							
Incidence of preterm delivery	0.154	0.058	0.278	2.666	0.009	0.039	0.265
Age	-0.006	0.005	-0.115	-1.069	288.0	-0.016	0.005
Education	0.035	0.034	0.104	1.017	311.0	-0.033	0.102
Occupation	0.229	0.107	0.217	2.143	0.035	0.017	0.441
History of fetus abortion	0.081	0.120	0.172	0.672	0.503	0.320	0.156
Gravity	0.055	0.126	0.209	0.437	0.663	-0.195	0.305
Parity	-0.035	0.168	-0.122	-0.208	0.836	-0.368	0.298
Live birth	-0.006	0.205	-0.022	-0.032	0.975	-0.414	0.401
Underlying disease	-0.046	0.068	-0.068	-0.675	0.501	-0.182	0.089
Body mass index	0.030	0.054	0.055	0.566	0/572	-0.076	0.137
Constant	-0.215	0.217		-0.992	0.324	-0.647	0.216
Final model							
Incidence of preterm delivery	0.133	0./052	0.240	2.548	0.012	0.029	0.236
Occupation	0.224	0.099	0.212	2.249	0.027	0.027	0.421
Constant	-0.231	0.118		-1.956	0.053	0.465	0.003

position, the use of modern pregnancy methods, and pregnancy infection as the main causes of preterm delivery.[26] Sohrabi *et al.* (2006) in their study, have introduced maternal age, multi-parity, history of placenta previa, placental abruption and urinary tract infection as effective factors in preterm delivery.[14] One of the most important causes of preterm delivery is related to urinary–genital infections and almost 40% of spontaneous preterm delivery are caused by infection. The causative organisms in these infections include *Neisseria gonorrhoeae*, TV *Chlamydia trachomatis*, *Ureaplasma urealyticum* and other infections such as hepatitis, AIDS and syphilis. Maternal systemic febrile infections such as pyelonephritis are also associated with preterm delivery.[27] Several studies imply the role of TV infection in the incidence of preterm delivery.[28-36] Nourian *et al.* (2010) have reported the prevalence rate of TV infection equal to 3.3%, which has been related to the incidence of preterm delivery ($P = 0.009$). Higher parity and urbanization were also associated with a significant increase in the risk of vaginal trichomoniasis ($P < 0.05$). There was no observed significant statistical relationship between TV infection with education level, method of delivery, household income rate and frequency of birth low weight cases in this study.[29] In Akbarian *et al.* (2004) study, the prevalence of TV was equal to 2.9%, and there was no observed significant relationship between infections with the mother's age, gestational age, number of pregnancies, number of fetus abortions, and clinical symptoms, and only 1 case (0.3%) of women infected with TV had preterm delivery and low birth weight babies.[37] Mohammadi *et al.* (2014) also reported the prevalence rate of TV equal to 2.4% with confirmation of the diagnosis by culture method.[38] In our study, the overall prevalence of trichomoniasis infection was equal to 8.2%, which is different from other studies. In the study by Bagchesarai *et al.* (2019), the prevalence rate of trichomoniasis was equal to 6.4%,[39] in the study by Nourian *et al.* (2010) it was equal to 3.3%,[29] in the study by Dolatsara *et al.* (2016) in Ardabil, the overall prevalence of trichomoniasis was equal to 3%.,[33] in the study by Mann *et al.* (2009) its prevalence rate was equal to 3%,[34] in the study by Hamouda *et al.* (2022) it was equal to 12% with culture confirmation[35] and in the study by Mohamed *et al.* (2018) its prevalence with culture confirmation was equal to 11.7%[36].According to this important fact, the diagnostic method in our study was PCR, which was different from the other methods and more accurate. The prevalence of trichomoniasis in different populations has been reported to range from less than 5% in the general population to more than 60% in high-risk groups,[40] and its global prevalence is estimated at 8.1% in women and 1% in men.[41] Also, its prevalence has been reported at about 20% in South Africa,[42] 4.1% in Brazil in the general population and 5.9% in pregnant women,[43] 0.9% in Arak (Iran),[44] 8.08% in Southeast Asia.,[45] 24.2% in India's rural population and 15.7% in urban population[46] and 0.2% in Slovenia in men and 1.6% in women. In our study, the overall prevalence of this infection was equal to 8.2%, which is consistent with its global prevalence. The prevalence of trichomoniasis infection among mothers with full-term delivery was 0.2% and among mothers with preterm delivery was equal to 13.1%. The probability of preterm delivery in mothers with trichomoniasis infection was more than seven times its probability of incidence in mothers without Trichomoniasis infection ($P = 0.035$, OR = 7.245). The results of the regression analysis showed that the probability of incidence of preterm delivery in mothers with trichomoniasis infection was related and it was much higher in working women than housewives. Akbarian *et al.* (2004), in their study, have reported that the prevalence rate of TV was equal to 2.9%, and there was no observed significant relationship with the age of the pregnant mother, gestational age, number of pregnancies, number of abortions, and preterm delivery.[37] Although the findings of our study in most cases are in line with the findings of the study of Akbarian *et al.*, some findings are different.[37] Our study, like the findings of Akbarian *et al.*, did not show a significant relationship between trichomoniasis infection and the age of the pregnant mother, gestational age, number of pregnancies, and number of abortions, but there was a significant relationship with preterm delivery. This difference should be found in the study method. In this regard, Baghchesaraie *et al.* (2009) in their study, have reported that the prevalence rate of trichomoniasis infection was equal to 6.4%, which there was no observed significant relationship with age, education level, contraceptive method, age of marriage, place of residence, employment status, and number of births.[39] Our study did not show a relationship between *Trichomonas* infection and the level of education, number of births, abortions and body mass index, but the average age of these women was slightly lower than the women that had no trichomoniasis infection. At a younger age, sexual activity and vaginal acidity changes provide favorable conditions for the growth of the organism. In terms of education level, the prevalence of *Trichomonas* infection is relatively higher in lower education levels and this is due to non-compliance with health recommendations to prevent infection in people with lower education levels. The findings of our study indicated a significant relationship between *Trichomonas* infection and preterm delivery. The probability of the incidence of preterm delivery in mothers with trichomoniasis infection was more than 7 times the probability of its incidence in mothers without trichomoniasis infection ($P = 0.035$, OR = 7.245). The probability of the incidence of preterm delivery in mothers with *Trichomonas* infection with lower age, higher BMI, presence of underlying disease, higher education, working women and parity, lower gravity and live birth, and having a history of abortion was greater, the effect of being employed was significant and the probability of the incidence of preterm delivery in working women with trichomoniasis infection was increased up to 15.8 times ($P = 0.029$, $\text{Exp}(\beta) = 15.801$). Mohamed *et al.* (2018) also reported the high prevalence of preterm delivery and adverse outcomes after birth in trichomoniasis patients, which has a significant relationship with age, place of residence (rural or urban), education level, and socioeconomic status.[36] In this regard, the findings of Hamouda *et al.*'s study (2022) indicated a significant relationship between preterm delivery and trichomoniasis infection, especially in cases where there was no adherence to treatment.[35] Maternal infection plays a role in the pathogenesis of preterm delivery through the intrauterine

inflammatory response. Trichomoniasis, chlamydia, gonorrhea, and syphilis are among the most common STIs worldwide. In parallel with the upward trend in preterm delivery, the US Centers for Disease Control and Prevention (CDC) surveillance reports showed that the prevalence of STIs has increased nationally in the general population from 2013 to 2018.[47,48] Trichomoniasis is a curable sexually transmitted disease that affects more than 25 million pregnant women worldwide every year.[49] The studies indicate a relationship between adverse birth outcomes (ABO), preterm delivery (PTD), and PROM with trichomoniasis.[50] Studies indicate increased IgE levels and increased production of interleukin (IL)-4 and IL-10 in the presence of trichomoniasis infection.[51] Trichomoniasis caused by TV is one of the common causes of vaginitis in women, and one of the aggressive factors of this organism is a cysteine protease that destroys the extracellular part and facilitates its connection to vaginal epithelial cells. The proteases secreted by the organism stimulate a TH2 response,[52] and thus trichomoniasis is associated with type 2 immunity. However, it is not clear which allergens and organism components are sensed by the host immune system and how host immune responses, particularly TH2 and IgE responses, are induced in response to genital system infection.[53] The physiological mechanism associated with trichomoniasis and adverse birth outcomes is not well understood. One theory is that PTD and PROM in pregnant women with trichomoniasis are related to the inflammatory response of the maternal innate immune system after infection, which includes increased cervical interleukin-8 (IL-8) levels and vaginal defensive levels.[54,55] These cytokines are markers of neutrophil activation associated with PTD and PPROM.[56] The cervical IL-8 induces cervical maturation and dilatation and supports its potential role in ABOs.[57,58] In pregnant women infected with TV, it spreads beyond the genital tract. In these women, an increase in the level of serum C-reactive protein has been reported.[54] It is assumed that endometriosis inflammatory associated with trichomoniasis interferes with placental circulation,[50] but further studies are needed to prove this issue.

Limitations and strengths

The main strength of this study is that it was conducted in a population-based group and the PCR method was used as a very accurate method in the diagnosis of TV. Then, it was conducted in a low-risk population, which increases generalizability to the general population. Some limitations should be noted which like most prospective cohorts, follow-up loss, may have resulted in the selection of a healthier population. There is this probability that the prevalence of trichomoniasis during the pregnancy period is higher in those who are not followed up. Finally, we did not examine high-risk behaviors, spousal contamination, or previous STD treatment among women with trichomoniasis during the pregnancy period. As with any observational study, confounding factors are considered as an issue. One of these confounding factors is the receipt of antibiotic treatment during pregnancy, but because our study was focused mainly on subclinical trichomoniasis infection, we assume that the large majority of women were not treated with antibiotics and this confounding effect is negligible.

Conclusion

In our study, the overall prevalence of trichomoniasis was equal to 8.2%, and in mothers with full-term delivery was 0.2% and in mothers with preterm delivery was equal to 13.1%. The probability of the incidence of preterm delivery in mothers with trichomoniasis infection was more than seven times the probability of its incidence in mothers without trichomoniasis infection. In our study, being employed was one of the influential factors in increasing the chance of the incidence of preterm delivery in mothers with trichomoniasis infection. The pregnant mother's age, gestational age, number of pregnancies, and number of abortions had no effect on preterm delivery in the presence of trichomoniasis infection.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

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