



# Frequency and Associated Risk Factors of *Entamoeba gingivalis* and *Trichomonas tenax* among Patients with Periodontitis in Western Iran

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## ABSTRACT

**Background:** The present investigation aims to estimate the frequency of *Entamoeba gingivalis* and *Trichomonas tenax* in periodontitis patients referring to Khorramabad Dental Faculty, Lorestan Province, Iran.

**Methods:** The present cross-sectional survey was carried out on 76 periodontitis patients (based on the Periodontology Association protocols) referring to Khorramabad Dental Faculty during August 2017 to May 2018. The samples (saliva and dental plaque) after collection were smeared on a glass slide, and in next step using Giemsa stain were stained; finally by means of a light microscope were tested. A prepared questionnaire considering a number of data such as age, gender, use of toothbrush, etc. were provided by each patient.

**Results:** Among the 76 patients with periodontitis, these parasites were found in 24 (31.6%) patients including 13 (17.1%) patients for *E. gingivalis*, 11 (14.5%) patients for *T. tenax*.

The obtained results indicates that although there was no significant relationship between age, gender, education, residence, smoking and the prevalence of *E. gingivalis* and *T. tenax* among the periodontitis patients; but a significant correlation was observed with mouthwash and teeth brushing ( $p < 0.05$ ).

**Conclusion:** The findings of current survey revealed that the remarkable prevalence of *E. gingivalis* and *T. tenax* in patients with periodontitis in Lorestan Province, Iran; indicating that *E. gingivalis* and *T. tenax* may possibly related to the progress of periodontitis. Nevertheless, consideration to oral health ideals, mainly brushing and use of mouthwashes may protect people from expose with these parasites.

**Key words:** Oral cavity parasites, Periodontal diseases, Mouthwash, Teeth brushing, Iran

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## INTRODUCTION

Periodontitis is considered as one of the most common oral diseases which is described by gingival hemorrhage, damage in connective tissue ligament, progress of periodontal pocket and alveolar bone loss [1,2]. Periodontitis is a multifactorial disease, in spite of numerous studies regarding the etiology of periodontitis,

there is still no known specific cause of the disease; however some studies showed to the role of several etiologic factors such as host responses, environmental factors, and microbial infections [2,3].

*Entamoeba gingivalis* is a protozoan that lives in the upper gastrointestinal areas such as the oral cavity, around the teeth, the teeth, and the margin of the gums. The prevalence of infection with this parasite is increased, in the conditions such as non-compliance with oral and dental hygiene and gum problems [4]. *Trichomonas tenax* is a protozoan parasite resides in the oral cavity which observed in lack of oral hygiene

with the estimated prevalence of 4%-53% [5]. The main transmission routes of these parasites are through saliva, kissing, as well as the use of common dishes for eating and drinking, etc., [6].

Although in the past, researchers and dentists believed that these protozoans were not directly related to oral cavity diseases, but new studies have shown that cannot be ignored the pathogenic potential of these microorganisms [7,8]. As, for example, *T. tenax* may be aspirated and lead to transonic or pulmonary transient infections. Studies have also shown that *E. gingivalis* can cause periodontal diseases, osteomyelitis, and even infections caused by intrauterine contraceptive device (IUD) [9-11].

Considering the importance of periodontitis and the necessity for additional investigations on its causes, the current survey aims to assess the frequency of *E. gingivalis* and *T. tenax* among patients with periodontitis referring to Khorramabad Dental faculty, Lorestan Province, Iran.

## METHODS

### Ethics

This survey was permitted by Ethics Committee of Lorestan University of Medical Sciences, Khorramabad, Iran.

### Participants

The present cross-sectional descriptive survey was carried out on 76 periodontitis patients (based on the Periodontology Association protocols) referring to Khorramabad Dental faculty, Lorestan Province, Iran during August 2017 to May 2018. In this study, patients who have taken systemic antibiotics in the last three month and also immunocompromised patients were excluded.

### Questionnaire

A prepared questionnaire with some information including age, sex, place of residence, smoking, toothbrush, mouthwash, etc., was completed for each patient.

### Collection of samples and microscopic assessment

At first, informed consent was obtained from the each patient. Then, two specimens were collected using sterile swabs from saliva and dental plaques. The samples (saliva and dental plaque) after collection were smeared on a glass slide, and in next step using Giemsa stain were stained; finally by means of a light microscope were tested.

## Statistical analysis

By SPSS software 23.0 obtained data were analyzed. To examine the univariate correlation among variables and prevalence Chi-square test was employed. Then, the multifactorial logistic regression models were employed to find the correlation between prevalence and the risk factors.  $P < 0.05$  was termed statistically significant.

## RESULTS

### Participants

The average age of the patients was  $39.2 \pm 10.4$  years; aging from 13 to 65 year. The majority of patients were female (54,71.1%) and the rest were male (22,28.9%). Totally, 63 patients (82.9%) lived in urban areas and 13 (17.1%) patients living in rural areas. By education, 53 (69.7%) patients were under diploma; but 23 (30.3%) were diploma and above. In term of smoking, 71 (93.4%) of patients had no smoking. Among the patients, 10 (13.2%) patients were used the mouthwash daily and 66 (86.8%) patients had brushing daily (Table 1).

**Table 1: Demographic data and the frequency of *E. gingivalis* and *T. tenax* among the patients with periodontitis from Lorestan Province, Iran**

Variables	No. (%)	Positivity for parasite	P value
<b>Gender</b>			
Male	54 (71.1)	17 (31.5)	0.52
Female	22 (28.9)	7 (31.8)	
<b>Age groups</b>			
<20 years	2 (2.6)	1 (50)	0.18
20-40 years	47 (61.8)	18 (38.3)	
≥ 30 years	27 (35.5)	5 (18.5)	
<b>Residential place</b>			
Rural	13 (17.1)	7 (53.8)	0.06
Urban	63 (82.9)	17 (27.1)	
<b>Education</b>			
<Diploma	53 (69.7)	20 (37.7)	0.06
≥ Diploma	23 (30.3)	4 (17.4)	
<b>Smoking</b>			
No	71 (93.4)	21 (29.6)	0.17
Yes	5 (6.6)	3 (60)	
<b>Mouthwash</b>			
No	66 (86.8)	24 (36.4)	0.01*
Yes	10 (13.2)	0 (0.0)	
<b>Brushing</b>			
No	10 (13.2)	7 (70)	0.009*
Yes	66 (86.8)	17 (25.8)	

\* $P < 0.05$ , difference is statistically significant

### Prevalence and risk factors of *E. gingivalis* and *T. tenax*

Among the 76 patients with periodontitis, totally 24 (31.6%) patients were found positive for oral cavity parasites including 13 (17.1%) patients for *E. gingivalis*, 11 (14.5%) patients for *T. tenax*.

As shown in Table 1, although the prevalence of *E. gingivalis* and *T. tenax* was higher in male patients (17,31.5%); but no significant difference was observed. In the case of age, although in the age group of 20-40 years, the highest rate of the prevalence was observed; but no significant correlation among the age groups and the prevalence of *E. gingivalis* and *T. tenax* was found.

Regarding the place of residence, the prevalence of oral cavity parasites was 53.8 and 27.1% among the patients of living in the rural and urban areas, respectively; indicating that there was no significant correlation between the prevalence of oral cavity parasites and the place of residence.

Considering the education level of participants, the higher prevalence rate of oral cavity parasites was observed among patients with education of less than diploma (20,37.7%). However, the statistical analysis exhibited no significant correlation among education and the prevalence of *E. gingivalis* and *T. tenax* was found.

Obtained results exhibited in term of smoking, the frequency of these parasites among those who smoke and do not smoke was 29.6 and 60%, respectively; indicated that no remarkable differences was found in the frequency of *E. gingivalis* and *T. tenax* and smoking.

Among the patients without using the mouthwash, 24 (36.4%) patients were found positive for *E. gingivalis* and *T. tenax*; while none of the people using mouthwashes were infected with *E. gingivalis* and *T. tenax*. The data analysis showed a considerable difference (p=0.01) in the prevalence of *E. gingivalis* and *T. tenax* and the use of mouthwash among patients.

From patients, who did not toothbrush, 7 (70%) of patients have *E. gingivalis* and *T. tenax*; whereas among the rest patients with tooth brushing, these parasites were observed in 17 (25.8%) of patients; demonstrating a significant difference (p=0.009) in the prevalence of *E. gingivalis* and *T. tenax* among these two groups of patients.

After perform the multifactorial logistic regression models, the results exhibited that, the mouth washing and teeth brushing were reflected significantly correlated to the prevalence of *E. gingivalis* and *T. tenax* (Table 2).

**Table 2: The evaluation of the potential factors related with the frequency of *E. gingivalis* and *T. tenax* among the patients with periodontitis in Lorestan Province, Iran**

Variables	OR (95% CI)	P value
<b>Age groups</b>		
<20 years	-	
20-40 years	0.5 (0.13-2.1)	0.34
≥ 30 years	-	
<b>Residential place</b>		

Urban	-	0.66
Rural	0.72 (0.16-3.1)	
<b>Education</b>		
<Diploma	2.46 (0.64-9.3)	0.18
≥ Diploma	-	
<b>Smoking</b>		
No	0.86 (0.04-11.6)	0.92
Yes	-	
<b>Mouthwash</b>		
No	4.8 (1.1-7.8)	0.042*
Yes	-	
<b>Brushing</b>		
No	6.3 (1.1-9.6)	0.038*
Yes	-	

\*P<0.05 was statistically significant.

## DISCUSSION

Periodontitis is an infectious and inflammatory disease that by engaging the periodontal tissue, causes alveolar bone disappears. Moreover, loss of teeth is one of the most prevalent problems of this disease and occurs when treatment is not taken [1,2]. Given the high rate of infectious agents, especially parasitic factors involved in oral diseases, as well as high prevalence of *E. gingivalis* and *T. tenax* in people with oral infections, it seems necessary to collect epidemiologic data, to determine the frequency of these two protozoa and investigate the probable risk factors. In the recent study we designed to determine the prevalence and also related risk factors of *E. gingivalis* and *T. tenax* in the periodontitis patients in Lorestan Province, Iran.

Among the 76 patients with periodontitis, totally 24 (31.6%) patients were found positive for oral cavity parasites including 13 (17.1%) patients for *E. gingivalis*, 11 (14.5%) patients for *T. tenax*.

Many investigations have revealed the frequency of *E. gingivalis* and *T. tenax* in many parts of the world, including: Pakistan (57% *E. gingivalis* and 3.3% *T. tenax*), Poland (14% *E. gingivalis* and 87% *T. tenax*), Nigeria (11.3% *E. gingivalis* and 4.9% *T. tenax*), and Tehran (41.7% *E. gingivalis* and 9.2% *T. tenax*) [12-15].

Regarding the prevalence of *E. gingivalis* and *T. tenax* in patients with periodontitis, a number of investigations have performed; for example, Ebrahim et al. in Iraq, the frequency of *E. gingivalis* and *T. tenax* in people with periodontitis was reported 70% and 30%, respectively [16]. Yazar et al. have demonstrated that among 107 patients with periodontitis in Egypt, the prevalence of *E. gingivalis* and *T. tenax* was 35.5% and 2.8%, respectively [17].

Ghabanchi et al. have reported that the rate frequency of *E. gingivalis* and *T. tenax* among the 50 patients with periodontal disease in Shiraz, southern Iran was 12% and 6%, respectively [18]. Maraghi et al. demonstrated

the frequency of *E. gingivalis* and *T. tenax* in patients with periodontitis in Khozestan Province, Iran was reported 0.5% and 0% for just in *E. gingivalis* and *T. tenax* infection, respectively [19]. This difference between the findings of our study and previous investigation may be due to some factors such as studied sample sizes, diagnostic techniques employed, and cultural and religious habits of studies areas [15,20].

Here we found that although there was no significant association between age, gender, education, residence, smoking and the prevalence of *E. gingivalis* and *T. tenax* among the patients with periodontitis; but there was significant correlation among the positivity to these parasites and mouth washing and teeth brushing; representing that considering the oral and dental health through brushing and mouth washing can limit the infection in people. Consistent with our findings, Gharavi et al. showed that there was no significant correlation among the age and prevalence of *E. gingivalis* and *T. tenax* [15]. Moreover, Ghaffari et al. showed that although no correlation was observed between age, gender, place of residence, smoking and the prevalence of *E. gingivalis* and *T. tenax* in periodontitis patients; but, a considerable correlation was observed between tooth brushing and flossing and the frequency of *E. gingivalis* and *T. tenax* in patients with periodontitis [21]. However, it has been previously proven that the prevalence of *E. gingivalis* and *T. tenax* is depended on a wide range of factors such as age, gender, socioeconomic conditions, cultural habits, etc., [20].

### CONCLUSION

The findings of this study indicate that the remarkable prevalence of *E. gingivalis* and *T. tenax* in patients with periodontitis referring to Khorramabad Dental Faculty, Lorestan Province, Iran; indicating that *E. gingivalis* and *T. tenax* could contribute to the development of periodontitis. However, attention to oral and dental health standards, especially brushing and use of mouthwashes can prevent people from infection with these parasites.

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