OPEN ACCESS

Orapuh Journal

REVIEW ARTICLE

Prevalences and trends of human oral protozoan parasites

Adamu, V. E. [1, 2], Amaechi, A. A.[3], Ajaero, C. M. U.[3], Eneojo, N. I. F. [1], Nwoke, B. E. B. [3], & Ukaga, C. N. [3]

[1]*Allied Health & Biological Sciences Department, Legacy University, Banjul, The Gambia.*
[2] *School of Global Health & Bioethics, Euclid University (Pôle Universitaire Euclide)*
[3]*Animal & Environmental Biology Department, Imo State University, Owerri, Nigeria.*

ARTICLE INFO

Received: 18 September 2020
Accepted: 24 October 2020
Published: 20 December 2020

Keywords:
Oral protozoan parasites,
Entamoeba gingivalis,
Trichomonas tenax

Peer-Review: Externally peer-reviewed

Correspondence to:
Lead-Author: Dr. V. E. Adamu
veadamu@gmail.com

To cite:
Adamu, V. E., Amaechi, A. A., Ajaero, C. M. U., Eneojo, N. I. F., Nwoke, B. E. B., & Ukaga, C. N. (2020). Prevalences and trends of human oral protozoan parasites. *Orapuh Journal*, *1*(2), e711.

ISSN: 2644-3740

ABSTRACT

Protozoan parasites are microscopic, unicellular organisms, of a group of phyla of the kingdom Protista, that can be free-living or parasitic. These organisms can live, and reproduce their kind, in human tissues. This accounts, in part, for why just a single protozoan can survive and infect humans so profoundly. A lot of people suffer severe morbidity connected to parasites. Parasitic protozoan parasite infections are associated with a lack of sanitation and access to safe and potable water. These infections make it impossible for poor populations to enjoy the full potentials of a healthy body and vitality and this hampers their productivity and, consequently, their social life and economic progress. Protozoan parasites that adapt to and live in the oral environment are called, 'oral protozoan parasites'. The appreciation of the fact that human oral protozoan parasites exist has long been documented by the empirical works of several researchers and scholars. Many of these works also indicated that *Entamoeba gingivalis* and *Trichomonas tenax*, which are parasitic oral protozoan parasites, are found only in the oral cavity. It is well known that the presence of these oral protozoan parasites may be established both in persons with pathological alterations in the oral cavity and those with no such symptoms. Human oral protozoan parasites cause gingival itch, palatal sore, halitosis, fatigue, fever, headaches, and periodontal tissue damage. Researchers have reported that the presence and impact of oral protozoan parasites may vary with age, gender, oral hygiene measures, general and oral health status, immune status, and alcohol and tobacco usage. Oral protozoan parasites constitute a growing concern to health authorities and researchers because they can either compromise oral integrity and health or complicate clinical intervention goals. The intentional control of oral parasites remains the only valid clinical option to improve the quality of life for sufferers and, by extension, all humans.

INTRODUCTION

Protozoan parasites are microscopic, unicellular organisms, of a group of phyla of the kingdom Protista, that can be free-living or parasitic. These organisms can live in, and reproduce their kind in human tissues. This accounts, in part, for why just a single protozoan can survive and infect humans so profoundly (Roberts & Janovy, 2010; Centers for Disease Control & Prevention [CDC], 2020).

Protozoan parasites of medical importance can be classified into four groups (this classification is based on their mode of movement):

1. Amoeba, e.g., *Entamoeba histolitica*;
2. Flagellates, e.g., *Giardia lamblia;*
3. Sporozoans, e.g., *Toxoplasma gondii*, and *Plasmodium species* and
4. Ciliates, e.g., *Balantidium coli*

(CDC, 2020).

Parasitic infections are associated with a lack of sanitation and access to safe and potable water. These infections make it impossible for poor populations to enjoy the full potentials of a healthy body and vitality and this hampers their productivity and, consequently, their social life and economic progress (World Health Organization [WHO], 2005; Mehraj et al., 2008).

Approximately 300 million people suffer severe morbidity associated with these parasites, at least 50% of which are school-age children affected by massive infections (Keiser & Utzinger, 2008).

According to the CDC, 2020), parasitic infections cause a tremendous burden of disease in both the tropics and subtropics as well as in more temperate climates. Of all parasitic diseases, malaria causes the most deaths globally. Malaria kills approximately 660,000 people each year, most of them young children in sub-Saharan Africa. CDC further stated that the Neglected Tropical Diseases (NTDs), which have suffered from a lack of attention by the public health community, include parasitic diseases such as lymphatic filariasis, onchocerciasis, and Guinea worm disease. The NTDs affect more than 1 billion people—about one-seventh of the world's population—largely in rural areas of low-income countries. These diseases exert a huge toll on endemic populations, including lost ability to attend school or work, retardation of growth in children, impairment of cognitive skills and development in young children, and the serious economic burden placed on entire countries. However, parasitic infections also affect persons living in developed countries.

Protozoan parasites are also present in the oral cavity. The category of protozoan parasites that live in the oral cavity is called oral protozoan parasites. Oral protozoan parasites are present in all populations and they dwell only in the oral cavity (Roberts & Janovy, 2010). According to Jackson and Rawdin (1996), up to 50% of persons with a healthy oral cavity may harbor oral protozoan parasites.

HUMAN ORAL PROTOZOAN PARASITES

Protozoan parasites that adapt to and can live in the oral environment are called, 'oral protozoan parasites'.

The appreciation of the fact that human oral protozoan parasites exist has long been documented by the empirical works of several researchers and scholars (Sonne & Gradwohl, 1980; Borwn & Neva, 1983; Dao et al., 1983; Gharavi, 2004; Ozumba et al., 2004; and Onyido et al., 2011). These studies have also revealed the existence of two parasitic oral protozoan parasites, namely, *Entamoeba gingivalis* and *Trichomonas tenax*.

Entamoeba gingivalis

E. gingivalis belongs to the Entamoebaidae family and the sub-order, Tubulinae (Albert et al., 1988; Gharavi, 2004). This parasite is found only in trophozoite form, which varies from 5-35 µm (Sonne & Gradwohl, 1980; Borwn & Neva, 1983; Dao et al., 1983; Gharavi, 2004).

E. gingivalis phagocytes bacteria and some other organisms, red blood cells and leukocytes. Like *E. histolytica*, the karyosome of *E. gingivalis* is located in its center and it is big, like that of an *E. coli*. This organism is structurally similar to *E. histolytica* and diagnosing it requires enough attention so that this protozoan parasite could be differentiated from *E. histolytica* released from lung abscesses. This amoeba has a large number of pseudopodia and may be found on gingival margins, in interdental spaces, carious lesions, paranasal sinuses, the alveolar pyorrhoea, tonsillary crypts, and in bronchial mucus. It has also been found in the contents of lung abscesses (Wantland et al., 1958; Sonne & Gradwohl, 1980; Lyons et al., 1983; Beaver et al., 1984; Dao, 1985; Markell et al., 1986; Derda et al, 2011). If the protozoan is found in bronchial secretion, it is necessary to differentiate between *E. gingivalis* and *E. histolytica*, which is based on the ability of *E. gingivalis* to phagocyte leucocytes (Jian et al., 2008). In some cases, it has been isolated from tonsillary crypts and tonsil tissue sections (Beaver et al., 1984; Markell et al., 1986; Gharavi, 2004). Clark and Diamonds (1992) opined that the vagina and uterus are regarded as suitable growth media for the organism.

Transmission of *E. gingivalis* to humans is usually during oral contact or through the common usage of dishes and crockery (Derda et al, 2011). Close contact, contaminated food, dishes, and oral droplets have been reported as ways of transmitting the protozoan parasites (Wantland & Laver, 1970; Beaver et al., 1984). The role of this protozoan in the etiology of periodontal disease has been established by some studies (Gottlieb & Miller, 1971; Sonne et al, 1980; Lyons et al., 1983; Linke et al., 1989).

This amoeba causes gingival itch, palatal sore, halitosis, fatigue, fever, headaches, and periodontal tissue damage

(Gharavi et al, 2006). It is well known that the presence of these protozoan parasites may be established both in persons with pathological alterations in the oral cavity and those with no such symptoms (Feki & Molet, 1990).

To date, the pathogenicity of *E. gingivalis* has not been demonstrated. The impact of the infection on the course of inflammatory processes in the oral cavity may be supported by the fact that *E. gingivalis* occurs more frequently amongst people with alterations of the mucous membrane of the oral cavity, inflammation of the palatal tonsils and paranasal sinuses, as well as amongst those with bad oral cavity hygiene and the sick with a lowered body immunity (Liu et al., 2001).

Trichomonas tenax

Trichomonas tenax, on the other hand, is a small trichomonad that usually occurs in the oral cavity of 5-10% of humans. This protozoan is of the Trichomonadidae family members (Albert et al., 1988; Gharavi et al, 2006). *T. tenax* is only found in the trophozoite form and has a size of 5µm to12 µm (Beaver et al, 1984).

Although considered a non-pathogenic anaerobic commensal, the harborage of this protozoan is most common among individuals with poor oral hygiene or dental diseases (Onyido et al., 2011). However, the organism could enter the respiratory tract by aspiration from the oropharynx and can result in bronchopulmonary trichomoniasis (Mallat et al., 2004; Mahmoud & Rahman, 2004; Chinche et al., 2005).

T. tenax is usually transmitted by kissing or common use of eating or drinking utensils and are usually resistant to changes in temperature and could live for several hours in drinking water (Hersh, 1985; Talaro & Talaro, 2002; Brooks et al., 2007; Roberts & Janovy, 2010).

PREVALENCES AND TRENDS OF HUMAN ORAL PROTOZOAN PARASITES

Frequency

The study of Ullah et al. (2006) indicated that the overall prevalence of human oral protozoan parasites in their study population was 40% and *E. gingivalis*, 57%. The reports of Wantland and Wantland (1960) indicated a prevalence of 39%, 23%, and 17.7% for *Entamoeba gingivalis, Trichomonas tenax,* and the mixed infection respectively in the population they studied. The works of Onyido et al. (2011) showed a 31.67% prevalence of *Entamoeba gingivalis* and 35% of *Trichomonas tenax*. Gharavi et al. (2006) reported a prevalence of 41.7% for *E. gingivalis*, 9.2% prevalence for *T. Tenax, and* 3.3% for the mixed infection respectively. Ozumba et al. (2004) inferred that the prevalence of *Entamoeba gingivalis* stood at 11.3% and that of *Trichomonas tenax*, 4.9% among the patients they studied.

Type

The work of Ozumba et al. (2004) declared that *E. gingivalis* was more common than *T. tenax* in their study population. Ibrahim and Abbas (2012) also reported a higher occurrence of *Entamoeba gingivalis* than *Trichomonas tenax*.

Gender

Gharavi et al. (2006) asserted that since people of the female gender respect oral care more than people of the male gender, the *E. gingivalis* infection is less prevalent among them. In contrast, El Hayawan and Bayoumy (1992) reported that the prevalence of *E. gingivalis* was generally more common among the female patients they surveyed than the male patients; similarly, in a study by Cavalcanti et al. (2011) both parasites (*E. gingivalis* and *T. tenax*) were more common in the female than the male participants. The studies of Gharavi, et al. and Cavalcanti, et al could not establish any relationship between gender and infection with human oral protozoan parasites.

Age

Several researchers have reported that the presence of oral protozoan parasites increases with age (Vrablic et al., 1991; Jawad, 2011). Ibrahim and Abbas (2012) reported that *Entamoeba gingivalis* was found at a higher rate in the age group, '61-70 years', and found *Trichomonas tenax* was enhanced in the age group, '31-40 years'. However, this assertion is at variance with several other studies, including the one by Gharavi, et al. (2006), which inferred that the infection was more prevalent in the age group of '21-30 years old'. The study by Vrablic et al. (1992) and the one by Onyido et al. (2011) elicited results that were similar to those of Gharavi, et al. in their own respective different study populations. There is a long-recognized assertion that human oral protozoan parasites are rarely found in children less than 20 years old (Cambon et al., 1979; Gharavi et al., 2006; Onyido et al., 2011;). Chunge et al., (1998) and Ibrahim and Abbas (2012) reported a

positive association between age and the prevalence of oral protozoan parasites.

Chewing stick use

Uller et al. (2006) indicated a 50% prevalence of human oral protozoan parasites for chewing stick users among the population they studied.

PCV/Hb levels

Packed Cell Volume (PCV) is the proportion of blood volume occupied by red blood cells (Purves et al., 2004) while haemoglobin (Hb) is the iron-containing oxygen-transport metalloprotein in the red blood cells of humans (Anthea et al., 1993). Decreased PCV/Hb levels could indicate life-threatening diseases such as leukemia (Udel.org, 2015; National Cancer Institute [NCI], 2015; DoctorsLounge, 2015). It can also be related to other conditions, such as malnutrition, water intoxication, anemia, and bleeding (Medlineplus, 2015). All these conditions suppress the body's ability to defend itself against infection, including that of parasites.

HIV/AIDS

Lucht et al. (1998) described the presence of oral protozoan parasites amongst the HIV-positive patients they studied. Cembranelli et al. (2013) also found oral parasites in HIV-positive patients. Both studies found only *E. gingivalis* in the HIV-positive patients they studied.

Salivary pH

Cavalcanti et al. (2011) inferred that the salivary pH of his study participants ranged from 6.0 to 8.0, but the peak incidence of commensals in salivary samples occurred between pH 6.0 and 6.5. These findings are also similar to the findings of some other studies (Souza, 1982; Zdero et al., 1999). The study of Ponce de León et al. (2001) found no relationship between salivary pH and the presence of human oral protozoan parasites.

Soft tissues

Oral mucosa ulceration and sub-madibular lymphadenopathy, more often than not, denote a drop in body immunity, following an infection.

Feki and Molet (1990), Cielecka et al. (2000), Chomicz et al. (2002a), and Chomicz et al. (2002b) demonstrated the pathogenicity of oral protozoan parasites in patients with lowered body immunity.

Cavalcanti, et al. (2011) suggested that *Entamoeba gingivalis* was were more common in the early stages of periodontitis. Wantland et al. (1958) inferred that both *E. gingivalis* and *T. tenax* were prevalent in individuals exhibiting pyorrhea and periodontitis in the population they studied. The prevalence of human oral protozoan parasites, especially, *E. gingivalis* is positively correlated with advanced periodontal disease. While it is known that *E. gingivalis* and *T. tenax* are commensal protozoan parasites commonly found in the human oral cavity, it is most probable that they are opportunists especially in the lesions of gingivitis and periodontal pockets (Talaro & Talaro, 2002). Ghabanchi et al. (2010) reported that parasitic infections were relatively common among patients with periodontal diseases.

Hard tissue

Dental caries or tooth cavities give preferentiality to the buildup of food debris and the development of dental biofilm (plaque), which constitutes a superb starting point for the growth of fungi and bacteria, as well as oral protozoan parasites. Roberts and Janovy (2010) inferred that the prevalence of human oral protozoan parasites was higher in participants with dental caries than participants with sound dentition. According to this study, among other cites of interest, oral protozoan parasites, especially *T. tenax* were most abundant in tooth cavities.

Oral hygiene

Some research works have implicated a poor state of oral hygiene, rated by the presence and magnitude of oral deposits, to the enhanced prevalence of oral protozoan parasites (Wantland & Laver, 1970; Dao et al., 1983; Chunge, 1998; Ozumba et al., 2004).

Body Immunity

CD4+ cell count, WBC count, and WBC differentials (neutrophils, lymphocytes, basophils, monocytes, and eosinophils) count measure the body's immune status. Feki and Molet (1990); Cielecka et al. (2000); Chomicz et al. (2002a); Chomicz et al. (2002b) have demonstrated the pathogenicity of human oral protozoan parasites in patients with immune-suppression, genetic diseases, and lowered body immunity. AIDS.org (2015) asserted that WBC, neutrophil, lymphocytes, and monocytes count lower than the normal range portend low immunity, which in turn favours enhanced prevalence of microorganisms, including parasites. AIDS.org further

inferred that eosinophils are involved with allergies and reactions to parasites and that a high count may indicate the presence of parasites. The site also inferred that basophils are not well understood, but that they are involved in long-term allergic reactions such as asthma or skin allergies.

Alcohol & tobacco usage

Ullah et al. (2006) found the highest oral protozoan parasite infestation in smokers (55%) as compared to non-smokers (40%). According to Preber et al. (1955), smoking is an important risk factor for both adult-onset periodontitis and early-onset periodontitis. Smoking may modulate the subgingival microbiota and increase the prevalence of certain pathogens. In the mouth of participants who used tobacco in the form of snuff chewing, biological changes occur. In the view of Uller et al., the pH of the mouth of snuff addicts is so deteriorating that the protozoan parasites cannot survive there. The studies by Ibrahim and Abbas (2012) and Cavalcanti et al. (2011) demonstrated a statistically significant relationship between tobacco usage and infection with human oral protozoan parasites.

Cavalcanti et al. (2011) did not find a relationship between the presence of *Entamoeba gingivalis* or *Trichomonas tenax* and alcoholism.

CONCLUSIONS

Oral protozoan parasites constitute a growing concern to health authorities and researchers because they can either compromise oral integrity and health or complicate clinical intervention goals. Understanding the dynamics of this interesting concern may help clinicians plan better oral health management protocols.

The intentional control of oral parasites remains the only valid clinical option to improve the quality of life for sufferers and, by extension, all humans.

Acknowledgment: The authors appreciate every researcher and scholar whose work has helped to shape this review article. The colour you have brought to this paper is enormous.

Ethics Approval: Nil needed.

Conflict of Interest: None declared.

OrCID iDs: Nil identified.

REFERENCES

AIDS.org (2015). *The complete blood count (CBC)*. http://www.aids.org/topics/complete-blood-count

Albert, D. M., Christy N. P., Conn, R. B., & Cruse J. M. (1988). *Medical Dictionary*. W.B. Saunders Co.

Anthea, M., Hopkins, J., McLaughlin, C. W., Johnson, S., Warner, M. Q., LaHart, D., & Wright, J. D. (1993). *Human Biology & Health*. Prentice Hall.

Beaver, P. C. Jung, R.C., & Cupp, E. W. (1984). *Clinical Parasitology,* Lea & Febige;

Borwn, W. B., & Neva, F. A. (1983). *Basic Clinical Parasitology*. Appleton- Century- Crofts.

Brooks, G. F., Carroll, K. C., Butel, J. S., Morse, S. A., & Jawetz, M. (2007). *Adelberg's Medical Microbiology*. The McGraw-Hill Companies, Inc.

Cambon, M., Petavy, A. F., Guitot, J., Glandier, I., Deguillaume, J., & Coulet, M. A. (1979). Study of the frequency of protozoa and yeasts isolated from the paradontium of 509 subjects. *Pathol Biol*; *27*, 603-606

Cavalcanti, R. L., Moura, C., Alcântara, W., Lopes, J., & Albuquerque, F. (2011). Incidence of *Entamoeba gingivalis* & *Trichomonas tenax* in samples of dental biofilm & saliva from patients with periodontal disease.*Rev Gaúcha Odontol*., *59*(1), 35-40

Cembranelli, S. B. S., Souto, F. O., Ferreira-Paim, K., Richinho, T. T., Nunes, P. L., Nascentes, G. A. N., Ferreira, T. B., Correia, D., Lages-Silva, E. (2013). First evidence of genetic intraspecific variability & occurrence of *Entamoeba gingivalis* in HIV(+)/AIDS. *PLoS ONE*. *8*(12), 1

Centers for Disease Control & Prevention [CDC] (2020). *About parasites*. http://www.cdc.gov/parasites/about.html

Chinche, L., Donati, S., Corno, G., Benoit, S., Granier, I., & Chouraki, M. (2005). *Trichomonas tenax* in pulmonary & pleural diseases. *Presse Med*. *34*, 1371-72

Chomicz, L., Piekarczyk, J., Fiedor, P., Starosciak, B. J., Szubinska, D., & Wojtowicz, A. (2002a). Screening evaluation of oral cavity microorganisms in dialyzed & kidney allograft recipients under chronic immunosuppression. *Transplant Proc*. *34*, 675-676.

Chomicz, L., Piekarczyk, J., Starościak, B., Fiedor, P., Piekarczyk, B., Szubińska, D., Zawadzki, P. J. & Walski, M. (2002b). Comparative studies on the occurrence of protozoans, bacteria & fungi in the oral cavity of patients with systemic disorders. *Acta Parasitol*. *47*, 147-153.

Cielecka, D., Chomicz, L., Piekarczyk, J., Walski, M., Zawadzki, P., Bednarczyk, A., & Szubinska, D. (2000). Oral cavity condition & the occurrence of parasitic protozoans in patients with genetic diseases. *Acta Parasitol*. *45*, 107-112.

Chunge, R.N., Manyi, F., & Amwuayi, P. (1998). Oral protozoa in a Kenyan population. *East African Med Jour*. *63*(3), 203-207

Clark, C. A. & Diamonds, L. S. (1992). Colonization of the uterus by the oral protozoan *Entamoeba gingivalis*. *American Journal of Tropical Medicine & Hygiene*. *46*, 158-60.

Dao, A. H., Robinson D. P. & Wong S. W. (1983). Frequency of *Entamoeba gingivalis* in human gingival scrapings. *American Journal of Clinical Pathology*. *80*, 380-83.

Derda, M., Hadaś, E., Antczak, E., & Wojt, W. J. (2011). Incidence of *Entamoeba gingivalis* in the oral cavity of students. *Journal of Stomatology*. *64*(10), 784-795.

Doa, A. H. (1985). *Entamoeba gingivalis* in sputum smears. *Acta Cytol*. *29*, 632-33.

DoctorsLounge (2015). *Packed Cell Volume (PCV)*. http://www.doctorslounge.com/hematology/labs/hematocrit.htm

El Hayawan, I. A. & Bayoumi, M. M. (1992). The prevalence of *Entamoeba gingivalis* & *Trichomonas tenax* in periodontal disease. *J Egypt Soc Parasitol*. *22*, 101-5.

Feki, A., Molet, B. (1990). Importance of *Trichomonas tenax* & *Entamoeba gingivalis* protozoa in the human oral cavity. *Rev Odontostomatol*. *19*(1), 37-45

Ghabanchi, J., Zhibaei, M., Afkar, M. D., & Sarbazie, A. H. (2010). Prevalence of oral *Entamoeba gingivalis* & *Trichomonas tenax* in patients with periodontitis & healthy population in Shiraz, South Iran. *Ind Jour Dent Res*. *21*(1), 89-91

Gharavi, M. J. (2004). *The Textbook of Clinical Protozoology*. Teimoorzadeh Publishers.

Gharavi, M. J., Hekmat, S., Ebrahimi, A., & Jahani, M. R. (2006). Buccal Cavity Protozoa in Patients Referred to the Faculty of Dentistry in Tehran, Iran. *Iranian Journal of Parasitology*. *1*(1), 43-46.

Gottlieb, D. S., & Miller, L. H. (1971). *Entamoeba gingivalis* in periodontal disease. *J Periodont*. *42*, 412-15.

Hersh, S. M. (1985). Pulmonary Trichomoniasis and *Trichomonas tenax*. *Journal of Medical Microbiology*. *20*, 1-10.

Ibrahim, S., & Abbas, R. (2012). Evaluation of *Entamoeba gingivalis* and *Trichomonas tenax* in patients with periodontitis and gingivitis and its correlation with some risk factors. *Basic Science. 24*(3), 158-162

Jawad, S. Q. (2011). Frequency of *Entamoeba gingivalis* & *Trichomonas tenax* among patients with dental prosthesis-fixed or removable. *J of College of Basic Education*. *68*, 97-100.

Jian, B., Kolansky, A. S., Baloach, Z .W., & Gupta, P. K. (2008). *Entamoeba gingivalis* pulmonary abscess–diagnosed by fine needle aspiration. *Cytology Journal*. *30*, 12.

Keiser, J., & Utzinger, J. (2008). Efficacy of current drugs against soil-transmitted helminth infections: systematic review and meta- analysis. J*AMA*. *299*, 1937–1948

Linke, H. A., Gannon, I. T. & Obin, J. N. (1989). Clinical Survey of *Entamoeba gingivalis* by multiple sampling in patients with advanced periodontal disease. *Inter J Parasitol*. *19*, 803-8.

Liu, G. Y., Chen, J. F., Wen, W. R., Chen, W. L., Lin, L. Q. & Hong, H. (2001). Experimental study on the pathogenesis of *Entamoeba gingivalis*. *Zhonghua Yi Xue Za Zhi*. *19*, 229-232.

Lucht, E., Evengard, B., Skott, J., Pehrson, P., & Nord, C. E. (1998). *Entamoeba gingivalis* in human immunodeficiency virus type1-infected patients with periodontal disease. Clin Infect Dis. *27*, 471-473.

Lyons, T., Scholten, T. N., Palmer, J. C. & Stanfield, E. (1983). Oral Amoebiasis: The Role of *Entamoeba gingivalis* in periodontal Disease. *Quintessence International*. *12*, 1245-48.

Mahmoud, M. S. & Rahman, G. A. (2004). Pulmonary trichomoniasis improved diagnosis by using polymerase chain reaction targeting *Trichomonas tenax* 18S r RNA gene in sputum specimens. *J Egypt Soc Parasitol*. *34*, 197-211

Mallat, H., Podglajen, I., Lavarde, V., Mainardi, J. L., Frappier, J., & Cornet, M. (2004). Molecular characterization of *Trichomonas tenax* causing pulmonary infection. *J Clin Microbiol*. *42*, 3886-7.

Markell, E. K., Voge, M., & John, D. T. (1986). *Medical Parasitology*. W.B. Saunders Co.

Medlineplus (2015). *Haematocrit*. http://www.nlm.nih.gov/medlineplus/ency/article/003646.htm

Mehraj, V., Hatcher, J., Akhtar, S., Rafique, G., Beg, M. A. (2008). Prevalence and factors associated with intestinal parasitic infection among children in an urban slum of Karachi. PLoS ONE. *3*, 1–6

National Cancer Institute [NCI] (2015). *Adult Acute Myeloid Leukemia Treatment (PDQ®)*. http://www.cancer.gov/cancertopics/pdq/treatment/adultAML/Patient/page1#_3

Onyido, A. E., Amadi, E. S., Olofin, I., Onwumma, A. A., Okoh, I. C., & Chikwendu, C. I. (2011). Prevalence of *Entamoeba gingivalis* & *Trichomonas tenax* among dental patients attending Federal School of Dental Technology & Therapy clinic, Enugu, Nigeria. *Nature & Science*. *9*(9)

Ozumba, U. C., Ozumba, N., & Ndiokwelu, E. M. (2004). Oral protozoa in a Nigerian population. *African Journal of Clinical & Experimental Microbiology*. *5*(1), 15-19.

Ponce de León, P., Zdero, M., Vasconi, M.D., Nocito-Mendonza, I., Lucca, A., & Perez, B. (2001). Relation between buccal protozoa and pH and salivary IgA in patients with dental prothesis. *Rev Inst Med Trop S Paulo. 43*(4), 241-2.

Preber, H., Linder L., & Bergstorm, J. (1955). Periodontal healing and perio pathogenic microflora in smokers and non-smokers. *J clinical Periodontol. 22, 946-952*

Purves, W. K., Sadava, D., Orians, G. H., & Heller, H. C. (2004*). Life: The science of biology* (7th ed.). Sinauer Associates.

Roberts, L. S., & Janovy, Jr., J. (2010). *Gerald D. Schmidt & Larry S. Roberts' Foundations of Parasitology* (8th ed.). The Mcgraw-Hill Companies, Inc.

Sonne, N. A. C., & Gradwohl, J. L. (1980). *Clinical Laboratory Methods & Diagnosis.* Mosby Co.

Souza, E. M. (1982). Estudo de Entamoeba gingivalis em boca de crianças. *Rev Microbiol*. *13*(2), 173-9.

Talaro, K. P. & Talaro, A. (2002). *Foundation in Microbiology*. The McGraw-Hill Companies Inc.

Ullah, Z., Khan, M., Jan, A. H. &Ali, I (2006). Mouth protozoa in North West Frontier Province of Pakistan. *Pakistan Oral & Dental Journal*. *27*(2), 245-8

Udel.edu (2015). *Diagnosis of myelogenous leukemia.* http://udel.edu/~kschieff/leukemiadiagnosis.html#References

Vrablic, J., Tomova, S., Catar, G., & Randova, L. (1991). Morphology & diagnosis of *Entamoeba gingivalis* & *Trichomonas tenax* & their occurrence in children & adolescent. *Bratisl Lek Listy*. *92*, 241- 6

Wantland, W. W., & Laver D. (1970). Correlation of some oral hygiene variables with age, sex & incidence of oral protozoa. *Journal of Dental Research*. *49*, 293-7

Wantland, W. W., Wantland, E. M., Remo, J. W., & Winquist, D. L. (1958). Studies on human mouth protozoa. *Journal of Dental Research*. *37*, 949-50.

Wantland, W. W., & Wantland, E. M. (1960). Incidence, ecology, & reproduction of oral protozoa. *J Dent Res. 39*, 863.

World Health Organization [WHO] (2005). Deworming for health and development. *Report of the third global meeting of the partners for parasite control*. World Health Organization, Geneva

Zdero, M., Ponce de Leon, P., Vasconi, M. D., Nocito-Mendonza, I., & Lucca, A. (1999). *Entamoeba gingivalis* & *Trichomonas tenax*: hallazgo en poblaciones humanas con y sin patología bucal. *Acta Bioquím Clín Lat-Amer*. *30*, 359-65.