

ORIGINAL RESEARCH

Medicine Science 2019;8(1):77-80

Epidemiology of Blastocystis spp. in primary school students at a central village of Ordu province**Ulku Karaman¹, Zeynep Koloren², Emine Ayaz², Umit Gur²**¹*Ordu University, Faculty of Medicine, Department of Parasitology, Ordu, Turkey*²*Ordu University, Faculty of Arts and Sciences, Department of Molecular Biology and Genetics, Ordu, Turkey*

Received 02 July 2018; Accepted 07 August 2018

Available online 18.10.2018 with doi:10.5455/medscience.2018.07.8912

Copyright © 2019 by authors and Medicine Science Publishing Inc.

Abstract

Blastocystis spp. is one of the most common intestinal parasites in humans and its pathogenicity is still controversial. The epidemiology of this parasite varies depending on the socio-cultural, socio-economic and geographic circumstances. In this study, Blastocystis spp. was investigated in stool samples of a Primary School students' in a Central Village in Ordu province who did not have any intestinal complaints. Before starting work, we were granted permission from National Education Directorate and the School Board. Blastocystis spp. was investigated in Primary School Students' stool samples by direct microscopy and Trichrome staining. The stool samples which showed any form of Blastocystis spp. were evaluated as "microscopically positive" regardless of the number per microscope field. Eighty and 85 of the 165 students participating in working were male and female, respectively. As a result of the microscopic investigation, 45 (27.3%) students were identified as positive. Eighteen of the positive students (40%) were male and 27 (60%) were female. Direct microscopic inspection, sedimentation and permanent staining are routine methods used in diagnosis of Blastocystis spp. This study also evaluated trichrome staining for each stool samples. In our research, this parasite was identified in 27.3% of asymptomatic Primary School students' stool samples. This suggests the parasite is relatively common in the region. Blastocystis spp. infects via the fecal-oral route. As a result, we presented recommendations for the enhancing personal hygiene, social education and the provision of water treatment for the students to protect themselves from infection with Blastocystis spp.

Keywords: Blastocystis spp., Epidemiology, Ordu, Primary school students**Introduction**

Blastocystis spp. which is defined in 1988 by Zierdt can be caused complication in gastrointestinal tract in many animal and human [1]. There are different opinions about pathogenicity, genetic diversity, host spectrum, and treatment options, although the knowledge of parasite morphology for long time [2]. There are many zoonotic genotypes in humans [3]. There are studies in the literature supporting the pathogen of Blastocystis [4-6], whereas, some authors have reported that Blastocystis spp. is not responsible the clinical symptoms of blastocystosis [7,8]. Tan [3] asserts that some populations may be more susceptible to infection with Blastocystis spp. and risk factors of this parasite are immunodeficiency, journey to the tropics, animal low hygiene, contact with contaminated water and food. It is reported that in the rare cases, detection of Blastocystis spp. is only amoeboid form can be shown severe diarrhea and while Blastocystis spp. can lead to

protracted and recurrent diarrhea in immunocompromised persons, especially AIDS patients [9]. In addition, Blastocystis infections cause symptoms such as diarrhea, abdominal pain, discomfort, bloating, gas, cramps, vomiting, dehydration, anorexia, insomnia, and nausea, lack of appetite, weight loss, fatigue, itching and tenesmus [10].

Blastocystis spp. has taken in the first row within the gastrointestinal protozoa in epidemiologic study, as well [6,11,12]. The economic developments, increasing human migration, environmental and demographic changes have been reported to be effective in the distribution of parasite. Subtropical climatic zone which is also including Turkey and evolves the proliferation of parasitic disease pathogens provides condition of optimal temperate climate [13].

Nowadays, the prevalence of parasite is different rates in developing countries and developed countries. The prevalence of Blastocystis spp. is the most common gastrointestinal protozoan with 1.5-10% in developed countries, while they are at the rate of 30-50% in developing countries. This situation have been connected to low hygiene, contacting with animals and contaminated water and food

*Corresponding Author: Ulku Karaman, Ordu University, Faculty of Medicine, Department of Parasitology, Ordu, Turkey
E-mail: ulkukaraman44@hotmail.com

[2,14]. It has been reported that the prevalence of this parasite was 1% in Japan [15], 40.9% in Brazil [16] and 33.3% in Egypt [17] while, the rate of this parasite ranged from 0.48-44.4% in Turkey [13,18].

Recent studies suggest that it is impossible to explain the relationship between *Blastocystis* and irritable bowel syndrome when intestinal microbiota is not considered. Researchers have suggested that the environment may become a condition that supports or does not support the multiplication of *Blastocystis* due to the changing intestinal flora in chronic bowel diseases [19].

Meta-genomic analysis in a retrospective study *Blastocystis* was detected in 20% of patients with ulcerative colitis (UC), but not in Crohn's disease [20].

According to our best reached knowledge, many researches were reported regarding as intestinal pathogens in different sites of Turkey. However, this research is the first for the detection on epidemiology of the intestinal parasites in children in Ordu province. In this study, *Blastocystis* spp. was investigated in stool samples of Primary School students who did not have any intestinal complaints at a Central Village in Ordu province of Black Sea Region in Turkey.

Material and Methods

Before starting work, we were granted permission from National Education Directorate and the School Board. The research was conducted in Department of Medical Parasitology laboratory at School of Medicine in Ordu University by examining 5-14 age group of 165 children's stool samples collected from Kokenli Elementary School in Ordu province between the periods of February, 2015-March, 2015.

The cover-sealed containers for stools were used for each specimen in research. The class teachers described how to take stool samples for students and it should not be at a rate of no more than 1/3 of the stools container. Fresh stool samples were examined without waiting after reached to the laboratory. The stools were macroscopically evaluated for the color, consistency, whether the presence of blood and mucus and parasitic. The stools slides were examined by direct microscopy and trichrome staining. All slides were screened by the use of direct microscopy under X10 and X40 magnifications and stained slides have been assessed by scanning 100 magnification.

Results

In the study, 80 and 85 out of 165 students participating in this research were male and female, respectively. As a result of the microscopic investigation, 45 (27.3%) students were identified as positive. Eighteen of the positive students were found to be male and 27 were female. *Blastocystis* spp. found over 5 in each field was evaluated as positive. Sixty percent of women and 40% of men has been found to be infecting with parasites. The parasites were found to be in the same samples by Trichrome stain.

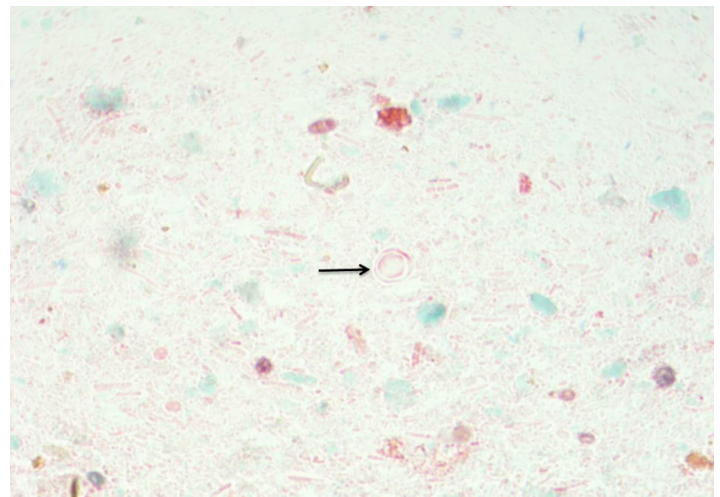


Figure 1. *Blastocystis* vacuolar form by Trichrome staining (100X magnification)

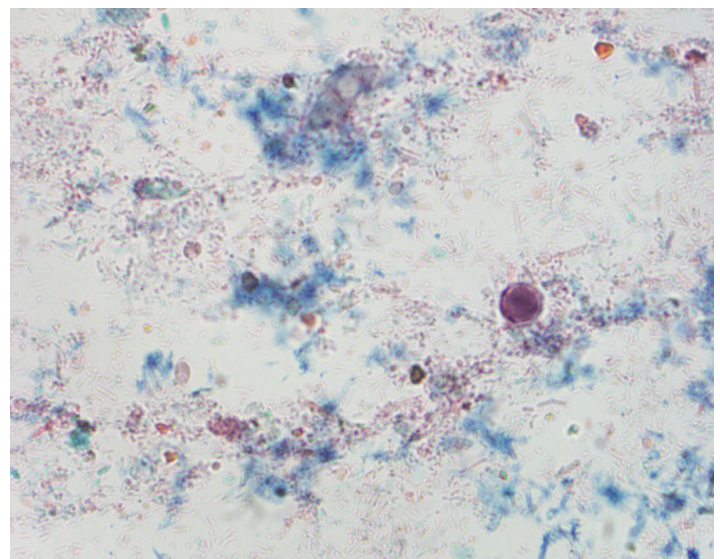


Figure 2. *Blastocystis* vacuolar form by Trichrome staining (100X magnification)

Discussion

Blastocystis spp. is one of the most common intestinal protozoa in humans. In nature there are many genotypes and human is the host for many zoonotic genotypes [3,21]. The pleomorphic structure of parasite and the lack of standardized diagnostic methods lead to misinterpretation in data about *Blastocystis* spp. The complexity about the pathogenesis of this parasite have originated from the presence of pathogenic and nonpathogenic genotypes [22]. Epidemiological, in vivo and in vitro studies related with parasite reinforce its the pathogen data [23].

The methods used in the routine diagnosis of *Blastocystis* spp. have been reported as directly screen, condensation and permanent staining. When directly screen comparing with condensation and Trichrome staining, these methods are reported to be more sensitive than directly screen [6].

Korkmaz et al [12] reported that the difficulty can be experienced in direct microscopic examination of blastocystosis because of different sizes and different morphological types of them and confusable with yeast and leukocytes by inexperienced persons. Even though, the authors found that the presence of the parasite

with trichrome staining higher than native-lugols, this method is also time consuming, laborious, and requires experience. In this study, both directly screen and trichrome staining method were used together.

Mumcuoğlu [24] reported that Blastocystis is found to be positive in 16 patients (29.1%) with irritable bowel syndrome were treated with direct microscopy, 18 (32.7%) with trichrome staining and culture method. Control group 2 without gastrointestinal system complaints; Blastocystis spp. was detected in 1 person (2.0%) by direct microscopy and Trichrome staining and in 3 persons (6%) by culture method. In control group 1, no other pathogens were found in all six patients who were detected 5 and up Blastocystis spp. on direct microscope. In addition to this situation, because of the absence of post-treatment complaints, reported that Blastocystis spp. Is a pathogen that needs to be evaluated by complaints and microscopy adherence?

Korkmaz et al [12] reported the occurrence of Blastocystis spp. in a total of 350 stool samples belonging to patients with diarrhea (n=157) and non-diarrhea (n=193) by native-lugol examination, trichrome staining and direct fluorescent antibody (DFA) methods. Researchers; Blastocystis spp. were found in culture of a total 66 (19%) fecal specimens which were 26 of diarrhea patients (16.6%) and 40 of non-diarrhea patients (21%). Blastocystis spp. was found to be positive in samples with 12% (42/350) Native-lugol, 17% (58/350) Trichrome staining and 19% (66/350) DFA method.

In our study, Native-lugol and Trichrome staining were also used for diagnostic purposes of Blastocystis spp.

It has been reported that parasites may be opportunistic pathogens in children and those with immunosuppression and should be monitored more carefully [25]. In people with Blastocystis infection, clinical findings are considered to be due to many factors such as Blastocystis genotype, microbiota, host immune response, Blastocystis concentration and other accompanying infection [19]. Blastocystis infection has been associated with numerous gastrointestinal and some dermatological symptoms (abdominal pain, diarrhea, constipation, weakness, vomiting, headache, dermatological manifestations, joint pain and psychiatric findings) in studies that have identified Blastocystis as pathogenic [26].

In this study, according to anamnesis, it was determined that the parasitized children have digestive system disorders. However, they have reported that they did not go to the doctor with the reasons for their complaints. This can be explained by the fact that the treatment of children should be taken into consideration.

Mumcuoğlu [24], Blastocystis spp. has found to be with 6 (7.5%), 8 (10.0%) and 15 (18.8%) of patients in the control group 1 with acute gastroenteritis complaints by direct microscopy, Trichrome staining and culture method, respectively. In addition, Blastocystis spp. has found to be with 1 (2.0%) and 3 (6%) of patients in the control group 2 with non-acute gastroenteritis complaints by direct microscopy, Trichrome staining and culture method, respectively. Blastocystis spp. has been identified in all of 6 patients in the control group 1 by direct microscopy in each field with the number of its 5 and up. But, no other pathogens were found in direct microscopy. Blastocystis spp. should be assessed by taking into consideration with microscope data and patient's complaints.

In the resource information reached with parasite epidemiology, the prevalence of this parasite was found to be with 1.5-10% in developed countries, while it was between 30-50% in the developing countries [25]. In America (USA?) and Brazil, the prevalence of Blastocystis was found to be with 18% and 26.5%, respectively [26,27], while, Blastocystis infection rate in Vietnamese pregnant women is reported as 20.4% [28].

In our country, the prevalence of Blastocystis spp. had been reported at ranging from 0.48-44.4% [13,18,29,30]. The prevalence of Blastocystis spp. was found to be with 0.48% in Kütahya [30], 2.1% in Istanbul [31], 22% in Ankara [32], 4.38% in Izmir [11], 7% in Eskişehir [33], 7.64% in Manisa [34], 23.2% in Kocaeli [35], 23.57% in Bursa [36]. According to Korkmaz et al. [12], Blastocystis spp. isolation have been reported in a total of 19% of the stool sample culture with 16.6% of patients with diarrhea and 21% of non-with diarrhea. Twelve percent of the stool samples are positive by native-lugols, 17% and 19% of them were positive by trichrome staining and DFA method, respectively. In this study, parasite was detected in children who not to go to the doctor because of any complaints.

Conclusion

This study represented that this parasite is relatively common in this region. Enhancing personal hygiene and public education and the provision of water treatment are necessary to protect themselves from Blastocystis infection.

Acknowledgements

We thank 2nd grade students in Faculty of Medicine for sample collection.

Competing interests

The authors declare that they have no competing interest

Financial Disclosure

The financial support for this study was provided by the investigators themselves.

Ethical approval

Before the study, permissions were obtained from local ethical committee.

Ulku Karaman ORCID: 0000-0001-7027-1613

Zeynep Koloren ORCID: 0000-0003-3982-5459

Emine Ayaz ORCID: 0000-0001-9708-2716

Umit Gur ORCID: 0000-0002-1511-1147

References

1. Tan TC, Suresh KG. Predominance of amoeboid forms of Blastocystis hominis in isolates from symptomatic patients. Parasitol Res. 2006; 98:189-193.
2. Roberts T, Stark D, Harkness J, et al. Update on the pathogenic potential and treatment options for Blastocystis sp., Gut Pathog. 2014;6-17.
3. Tan KS. New insights on classification, identification, and clinical relevance of Blastocystis spp. Clin Microbiol Rev. 2008;21:639-65.
4. Leder KM, Hellard E, Sinclair MI, et al. No correlation between clinical symptoms and Blastocystis hominis in immunocompetent individuals. J Gastroenterol Hepatol. 2005;20:1390-4.
5. Andiran N, Acikgoz ZC, Turkay S, et al. Blastocystis hominis, an emerging and imitating cause of acute abdomen in children. J Pediatr Surg. 2006;41:1489-91.
6. Ozcikir O, Gureser S, Erguven S, et al. Characteristics of Blastocystis hominis infection in a Turkish university hospital. Turkey Parazitolo J. 2007;31:277-82.
7. Muller RA, Minshev BH. Blastocystis hominis: an organism in search of a disease. Rev Inf Dis. 1988;10:930-8.

8. Senay H, MacPherson D. Blastocystis hominis: epidemiology and natural history. *J Inf Dis.* 1990;162:987-90.
9. Windsor JJ, Whiteside TM, Chalmers RM, et al. Blastocystis hominis: a common yet neglected human parasite. *Brit J Biomed Sci.* 2001;58:129-30.
10. Godekmerdan A. Prevalence of Blastocystis hominis in patients presenting dispensary and its relationship with gastro-intestinal complaints. Erciyes University Institute of Health Sciences. 1995;4:5-13.
11. Inceboz T, Uslu S, Over L, et al. The epidemiology research of Blastocystis hominis in the Dokuz Eylul University Medical Faculty Hospital between 2005 and 2009. *Turkey Parazitol J.* 2011;35:72-6.
12. Kormaz GA, Doğruman ALF, Mumcuoğlu I. Investigation of stool specimens of Blastocystis sp. by microscopic, culture and molecular methods. *Mikrobiol Bul.* 2015 49:85-97.
13. Akdemir C, Helvacı R. Evaluation of parasitology laboratory results of a group of people older than 15 years of age in Kutahya. *Turkey Parazitol J.* 2007;31:37-40.
14. Alfellani MA, Stensvold CR, Vidal-Lapiedra A, et al. Variable geographic distribution of Blastocystis subtypes and its potential implications. *Acta Trop.* 2013;126:11-8.
15. Hirata T, Nakamura H, Kinjo N, et al. Prevalence of Blastocystis hominis and Strongyloides stercoralis infection in Okinawa, Japan. *Parasitol Res.* 2007;101:1717-9.
16. Aguiar J I, Goncalves AQ, Sodre FC, et al. Intestinal protozoa and helminths among Terena indians in the State of Mato Grosso do Sul: high prevalence of Blastocystis hominis. *Rev Soc Bras Med Trop.* 2007;40: 631-4.
17. Rayan HZ, Ismail OA, El Gayar EK. Prevalence and clinical features of Dientamoeba fragilis infections in patients suspected to have intestinal parasitic infection. *J Egypt Soc Parasitol.* 2007;37:599-608.
18. Atambay M, Karaman U, Aycan O, et al. The frequency of intestinal parasites and head lice among students of the Aksemsettin Primary School for deaf students. *Turkey Parazitol J.* 2007; 31:62-5.
19. Stensvold CR, Clark CG. Current status of Blastocystis: A personal view. *Parasitology International* 2016;65:763-71.
20. Andersen LO, Bonde I, Nielsen HB, et al. A retrospective metagenomics approach to studying Blastocystis. *FEMS Microbiol Ecol.* 2015;91.
21. Noël C, Dufernez F, Gerbod D, et al. Molecular phylogenies of Blastocystis isolates from different hosts: implications for genetic diversity, identification of species, and zoonosis. *J Clin Microbiol.* 2005;43:348-355.
22. Clark CG. Extensive genetic diversity in Blastocystis hominis. *Mol Biochem Parasitol.* 1997;87:79-83.
23. Giacometti A, Cirioni O, Fiorentini A, et al. Irritable bowel syndrome in patients with Blastocystis hominis infection. *Eur J Clin Microbiol Infect Dis.* 1999;18:436-9.
24. Mumcuoğlu I, Coskun FA, Aksu N, et al. Role of Dientamoeba fragilis and Blastocystis spp. in irritable bowel syndrome. *Turkey Parazitol J.* 2013;37:73-7.
25. Stenzel DJ, Boreham PF. Blastocystis hominis revisited. *Clin Microbiol Rev.* 1996;9:563-84.
26. Nascimento SA, Moitinho Mda L. Blastocystis hominis and other intestinal parasites in a community of Pitanga City, Paraná State, Brazil. *Rev Inst Med Trop S Paulo* 2005;47:213-7.
27. Eassa SM, Ali HS, El Masry SA, et al. Blastocystis hominis among immunocompromised and immunocompetent children in Alexandria, Egypt. *Ann Clin Laboratory Res.* 2016;4.
28. Hameed DM, Hassanin OM, Zuel-Fakkar NM. Association of Blastocystis hominis genetic subtypes with urticaria. *Parasitol Res.* 2011;108:553-60.
29. Amin OM. The epidemiology of Blastocystis hominis in the United States. *Res J Parasitol.* 2006;1:1-10.
30. Cheng HS, Haung ZF, Lan WH, et al. Epidemiology of Blastocystis hominis and other intestinal parasites in a Vietnamese female immigrant population in Southern Taiwan, Kaohsiung. *J Med Sci.* 2006;22:166-70.
31. Celik T, Daldal N, Karaman U, et al. Incidence of intestinal parasites among Primary School Children in Malatya. *Turkey Parazitol J.* 2006;30:35-8.
32. Malatyali E, Ozcelik S. Isolation of Blastocystis sp. from human hosts and in vitro determination of different morphological forms. *Turkey Parazitol J.* 2011;35:19-22.
33. Koksali F, Baslanti I, Samasti M. A retrospective evaluation of the prevalence of intestinal parasites in İstanbul, Turkey. *Turkey Parazitol J.* 2010;34:166-71.
34. Gulmez D, Saribas Z, Akyon Y, et al. The results of Hacettepe University Faculty of Medicine Parasitology Laboratory in 2003-2012: Evaluation of 10 Years. *Turkey Parazitol J.* 2013;37:97-101.
35. Dogan N, Demirustu C, Aybey A. The Prevalence of intestinal parasites according to the distribution of the patients' gender and parasite species for five years at the Osmangazi University Medical Faculty. *Turkey Parazitol J.* 2008;32:120-5.
36. Duzyol D, Kilimcioglu AA, Cengiz Ozyurt B, et al. Incidence of intestinal parasites detected in the Department of Parasitology in Celal Bayar University Hospital between 2006 and 2010. *Turkey Parazitol J.* 2012;36:147-51.
37. Tamer GS, Caliskan S, Willke A. Distribution of intestinal parasites among patients who presented at the Parasitology Laboratory of the Kocaeli University School of Medicine Hospital. *Turkey Parazitol J.* 2008;32:126-9.
38. Alver O, Oral B, Tore O. The distribution of intestinal parasites detected in the Uludag University Medical School Hospital between 2005 and 2008. *Turkey Parazitol J.* 2011;35:194-8.