

An Outbreak of Gastroenteritis Associated with Intestinal Parasites

Sema TUNCAY¹, Songül DELİBAŞ¹, Tonay İNCEBOZ¹, Leyla ÖVER¹,
Ahmet Murat ORAL², Çiler AKISÜ¹, Ümit AKSOY¹

¹Dokuz Eylül University, School of Medicine, Department of Parasitology,
²Provincial Health Directorate, Contagious Diseases Department, İzmir, Türkiye

SUMMARY: Intestinal protozoa are one of the leading causes of waterborne outbreaks. Stool samples of 196 residents from a village of İzmir, using the public water supply, were collected during an outbreak of gastroenteritis. Patients were asked to fill out a questionnaire reporting on gender, age, gastrointestinal symptoms, whether or not there was a toilet in the house, their hygiene practices, and similar symptoms in the household members. Of the patients who had gastrointestinal symptoms (74.5%), diarrhea was observed in 69.5% whereas bloody and mucoid stools were observed in 20.4 %. The stool samples were examined for intestinal parasites by wet mount and trichrome stain and were also cultured in the Robinson medium. Pathogenic parasites were detected in 11 samples (5.6%) as follows: *Giardia intestinalis* in 7, *Hymenolepis nana* in 1 and *Blastocystis hominis* in 4. *Entamoeba histolytica/dispar* was not detected by direct wet mount in any of 8 patients who had *E. histolytica/E. dispar* in culture whereas it was detected in the trichrome stained slides of 3 patients. Amoeba prevalence in the 15-44 age-group was significantly high when compared with the 0-14 age group. The prevalence of pathogenic parasites was high among the people who had a toilet outdoors. Drinking water was thought to be a principal source of this outbreak.

Key Words: Intestinal parasites, outbreak

İntestinal Parazitlerin Eşlik Ettiği Bir Gastroenterit Salgını

ÖZET: İntestinal protozoanlar, su kaynaklı salgınlara önemli nedenlerinden birisidir. İzmir'in bir köyünde çıkan gastroenterit salgını sırasında, bu bölgede oturan ve şebeke suyu kullanan 196 kişiden gaita örneği toplandı. Hastaların cinsiyet, yaş, gastrointestinal semptomlar, tuvaletin evin içinde olup olmadığı, hijyen alışkanlıkları ve diğer aile fertlerinde benzer semptomlarla ilgili bir anket formu doldurması istendi. Gastrointestinal bulguları olan hastalarda (%74,5), ishal %69,5 oranında, kanlı ve mukuslu dışkı ise %20,4 oranında gözlemlendi. Dışkı örnekleri direkt bakı ve trichrom boyası ile boyanarak incelendi ve Robinson besiyerine ekim yapıldı. Örneklerin 11'inde (%5,6) patojen parazitler saptandı: 7 *Giardia intestinalis*, 1 *Hymenolepis nana*, ve 4 *Blastocystis hominis*. *Entamoeba histolytica/dispar* 8 hastanın kültüründe saptandı. Bu hastaların üçünde, trikrom boyada *E. histolytica/E.dispar* görülmesine karşın kültür öncesinde yapılan direkt bakıda *E. histolytica/E.dispar* saptanamadı. Yaş grupları göz önüne alındığında, amip prevalansı 15-44 yaş grubunda, 0-14 yaş grubuna göre anlamlı olarak yüksekti. Patojen parazitlerin prevalansı, tuvaleti evin dışında olanlarda yüksekti. Salgının temel kaynağının içme suyu olduğu düşünüldü.

Anahtar Sözcükler: Bağırsak parazitleri, salgın

INTRODUCTION

Water is a major conduit for protozoan parasites, and contaminated water is an important source of human infection either by direct consumption or by the use of contaminated water in food processing or preparation. The use of human

feces for fertilizer is also an important source of infection. Waterborne outbreaks of protozoan parasites are far more common than outbreaks due to helminths because of the smaller sizes of their transmissible stages (3, 9, 10).

At the beginning of October 2005 an outbreak of gastroenteritis had come out in a village of İzmir. The village, named Karacağaç, is in Buca district and is about 27 kilometers from İzmir (Figure 1). The population of the village is approximately 1500 and the villagers are mostly involved in agriculture. All residents in the village were using the same public water supply and water is pumped from 200 meters underground.

Makale türü/Article type: **Araştırma / Original Research**

Geliş tarihi/Submission date: 17 Aralık/17 December 2007

Düzeltilme tarihi/Revision date: 19 Mart/19 March 2008

Kabul tarihi/Accepted date: 23 Mart/23 March 2008

Yazışma /Corresponding Author: Sema Tuncay

Tel: (+90) (232) 412 45 47 Fax: -

E-mail: smtuncay@gmail.com

11. Uluslararası Parazitoloji Kongresinde (ICOPA XI) sunulmuştur.

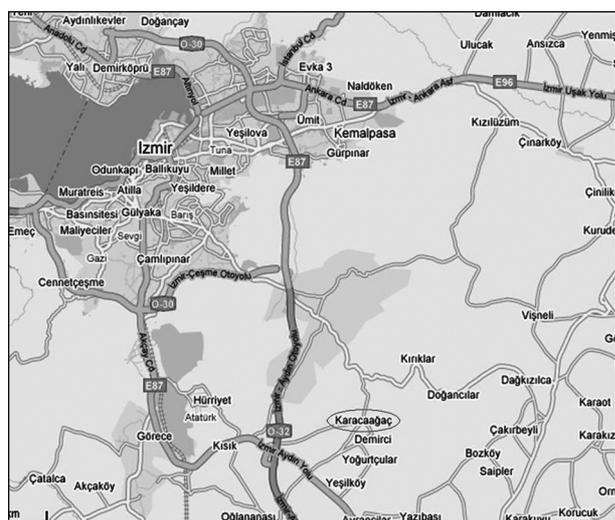


Figure 1. Map of Izmir (Karacağağaç village)

As the city is in the Mediterranean climate zone, it is dry and hot in summer while it is mild and rainy in winter. Seventy seven percent of the rain falls during October through May (12).

A midwife in the village informed the local health authorities about the occurrence of several cases of diarrhea. The local authorities conducted an investigation in the school and found out that 35 primary school children had diarrhea and vomiting. The same symptoms were also seen among adults. Stool samples from the school children were sent to the Public Health Laboratory for direct microscopy and amoeba trophozoites and cysts were seen in many samples. The lack of any common activity among cases and the homogenous spread of cases all over the village led the hypothesis of a waterborne outbreak.

MATERIAL AND METHODS

Six days after the notification of the first case, an investigation was started to determine the magnitude of the outbreak, the source of infection and to prevent further transmission. Since amoeba trophozoites and cysts were seen in many samples in the first examination, an outbreak of intestinal protozoans was suspected. A detailed stool examination was planned for people with gastrointestinal symptoms such as diarrhoea, abdominal pain or nausea. They were invited to present at the local clinic and were asked to fill a questionnaire. Stool samples were collected by the investigation team with representatives from the Parasitology Department of Dokuz Eylul University and the local health authorities.

Environmental investigation: Environmental health officials also collected water samples from the water distribution system for bacteriological and chemical analyses.

Epidemiological investigation: 196 residents took part in the investigation. One stool sample from each of 196 residents were collected. They were asked to fill a questionnaire

reporting on gender, age, symptoms (diarrhea, abdominal pain, flatulence, vomiting, nausea, fever, and blood or mucus in stool), whether or not there was a plumbed toilet in the house, their hygiene practices like hand washing, whether other household members had similar symptoms.

The data was analysed by SPSS 11.0.

Parasitological investigation: The stool samples were first examined by direct wet mount with saline and 2% iodine solution. Permanent smears of specimens fixed in Schaudinn fixative were stained with trichrome and examined by light microscopy for protozoa. In the meantime the samples were cultured in the Robinson medium and examined 24 and 48 hours later. The smears prepared from the cultures were also stained with trichrome. The microbiological examination of the stool samples for bacterial and viral enteric pathogens could not be done.

RESULTS

Environmental investigation: Analysis of the water samples revealed that there was no residual chlorine in the water at the time of the outbreak, although the head of the village had reported that the water was chlorinated. In addition, high levels of fecal indicator bacteria (primarily coliform counts) indicated faecal contamination in the public water supply.

Epidemiological investigation: Eighty seven (44%) of the residents were men and 109 (56%) were women.

Fifty seven percent of the residents were in 0-14 age group (Figure 2).

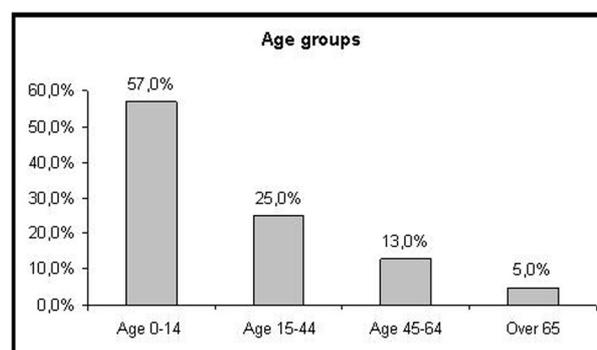


Figure 2. Distribution of age groups

One hundred forty one villagers completed the questionnaire (Response rate, 72%). Ninety six percent of the people were using public water supply. There was no toilet indoors in 8.7% of the houses (Figure 3).

Gastrointestinal symptoms like diarrhea, abdominal pain, flatulence, vomiting, nausea, and fever were reported by 105 (74.5%) of the cases (Figure 4). Seventy three (69.5%) of these villagers had diarrhea and the duration of the diarrhea was less than a week in 82% of the cases. Twenty two (20.4%) patients with diarrhea had bloody and mucoid stool.

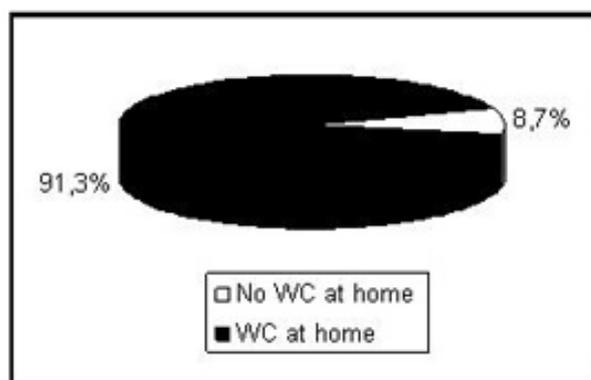


Figure 3. Condition of the toilets

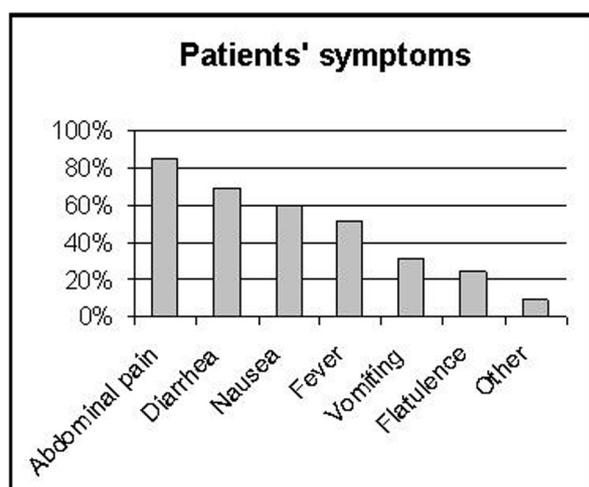


Figure 4. Symptoms of the patients in the outbreak

Fifty seven percent of the people stated that their family members had similar complaints.

Parasitological investigation: Pathogenic (5.6%) and non-pathogenic (29.1%) parasites were detected with various methods. Intestinal parasites were detected in 68 (34.7%) of the stool samples.

Pathogenic parasites were detected in 11 samples (5.6%), in which one sample had two different parasites (*G. intestinalis* and *B. hominis*). Of these parasites; *G. intestinalis* in 7; *H. nana* in 1; *B. hominis* (5 or more parasites in one field by 40x) in 4 samples.

E. histolytica /*E. dispar* was also detected in 8 patients. But it was not detected by direct wet mount in any of 8 patients who had *E. histolytica* /*E. dispar* in culture whereas it was detected in the trichrome stained slides of 3 patients. The prevalence of pathogenic parasites was high among the people who had a toilet outdoors compared to the ones who had a toilet indoors ($p < 0.05$).

Nonpathogenic parasites were detected in 57 (29.1%) patients. Of these parasites; *B. hominis* (fewer than 5 parasites in one field by 40x) in 42; *E. coli* in 8; *E. nana* in 7; *E. hartmanni* in 6; *I. bütschlii* in 4; *T. hominis* in 4 samples.

Two or more nonpathogenic parasites were detected in 10 patients.

When age groups were considered, amoeba were most prevalent (27.9%) in 15-44 age group whereas the prevalence was the lowest (8.1%) in the 0-14 age group. The amoeba prevalence in 15-44 age group was significantly high when compared with the prevalence in 0-14 age group ($p < 0.05$).

DISCUSSION

The combination of increased surveillance, improved detection methods, and testing requirements should result in a marked improvement in the ability to detect, investigate, and control food and waterborne parasitic protozoal agents (4).

In this outbreak that had come out in October 2005, pathogenic parasites were detected in 5.6% and nonpathogenic parasites in 29.1% of 196 villagers. Ingestion of food and water contaminated with *E. histolytica* /*E. dispar* and *G. intestinalis* cysts from human faeces and direct faecal oral contact are the commonest means of infection. Transmission of *E. histolytica* /*E. dispar* and *G. intestinalis* cysts by untreated drinking water is common in developing countries where much of the water supply for drinking is untreated and fecally contaminated (3, 7, 10).

Ninety six percent of the people in the village were using public water supply and 57% of the people stated that their family members had similar complaints. These data indicate that contaminated drinking water was the most likely cause of this outbreak, either through inadequate municipal water treatment or contamination of water in the distribution system. Analyses for coliform bacteria can help to document water contamination and provide information about the source of contamination (2). Microbiological investigation of the water in the village indicated that the most probable cause of contamination was the introduction of fecal bacteria during heavy rainfalls in September, 2005. Weather conditions may have been an important contributing factor in outbreaks (5). The average annual rainfall in Izmir is 700 mm, with the highest rainfall usually in December, January and February (12). An unusually heavy rainfall during a short period in September may have contributed to this outbreak by overloading the old sewage system and causing cross-contamination between sewage and drinking-water pipes.

Non chlorinated ground water systems may be also susceptible to waterborne outbreaks and constitute a risk to rural populations that could be reduced by chlorination of water (5). Chlorination will effectively eliminate faecal indicator bacteria from the water, but chlorine-resistant pathogens such as protozoan parasites can still be present (8). There was no residual chlorine in the municipal water during this outbreak.

Drinking water in the village could not be tested for *E. histolytica* /*E. dispar* and *G. intestinalis* because of lack of

appropriate equipment and other resources. The methods necessary for recovery of pathogenic protozoa are expensive because they are highly specific, labor intensive, and time consuming. Besides waterborne transmission can be established without evidence of the pathogen in the water system. At a workshop in the United States in December, 1998, it was recommended that emphasis should be given to the collection of water quality information that will help assess sources of fecal contamination rather than focus on the detection of waterborne pathogens during outbreak investigations (2).

An important issue is the delay in the recognition of an outbreak. Improved surveillance and reporting activities can help to detect outbreaks and reduce the time required to initiate an investigation (2). Enhancing surveillance activities and increasing laboratory support for testing clinical and water samples are recommended by the Centers for Disease Control (CDC) in United States to improve waterborne outbreak investigations (6).

At the beginning of 2005, a new and completely revised communicable disease notification system was launched nationwide in Turkey. Recent changes in disease epidemiology and developments in diagnostic capability meant that the notifiable diseases and the surveillance methods in the Turkish communicable disease notification system needed to be revised (1).

The new list of mandatorily notifiable diseases consists of 51 diseases, divided into four groups. *E. histolytica* and *G. intestinalis* are in Group D. Group D involves the notification of an infectious agent. This is an important innovation that involves the direct participation of laboratories in the notification system. The aim is to get data on the source of communicable diseases that remain a public health problem, and to study the epidemiology of these diseases when necessary. Only laboratories using acceptable diagnostic techniques will be able to notify cases. Group D data are notified to the provincial health directorates who implement actions (1).

In this new system case definition of *E. histolytica* is made either by identification of trophozoites containing erythrocytes in trichrome stained slides or by ELISA which can differentiate *E. histolytica* from the nonpathogenic *E. dispar*. *E. histolytica* should be notified to the Provincial Health Directorate in 24 hours (11).

Giardiasis was not a notifiable disease before 2005 and was included in the list of notifiable diseases in this new system. Case definition of *G. intestinalis* is made either by examination of fresh stool, duodenal aspiration or biopsy material for cysts and trophozoites, or by detecting *G. intestinalis* antigens by ELISA or DFA (11).

In conclusion leaking sewage pipes combined with insufficient water treatment was the likely cause of this outbreak. It should remind us that towns with decaying water treatment facilities

and worn out water distribution systems are at risk for waterborne disease outbreaks, especially with organisms that are resistant to chlorine disinfection. Developing efficient and rigorous disinfection methodologies, for example the use of ultraviolet radiation and ozone in addition to chlorination can help in controlling chlorine-resistant protozoans. Proper disposal of human and animal wastes to prevent contamination of foods and drinking water sources is also an excellent and basic strategy for preventing many parasitic infections that are transmitted by the fecal–oral route. Ensuring quality standards in the drinking water and sewage systems is very important to prevent further outbreaks.

REFERENCES

1. **Bayazit Y, Buyurgan V, Tumay S**, 2005. New communicable disease notification system launched in Turkey. *Euro Surveill*, 10: E050421.5.
2. **Craun GF, Frost FJ, Calderon RL, Hilborn ED, Fox KR, Reasoner DJ, Poole CL, Rexing DJ, Hubbs SA, Dufour AP**, 2001. Improving waterborne disease outbreak investigations. *Int J Environ Health Res*, 11: 229–243.
3. **Haque R, Huston CD, Hughes M, Houpt E, Petri WA, Jr.**, 2003. Current concepts: Amebiasis. *N Engl J Med*, 348: 1565–1573.
4. **Jaykus LA**. 1997. Epidemiology and Detection as Options for Control of Viral and Parasitic Foodborne Disease. *Emerg Infect Dis*, 4: 529–539.
5. **Kuusi M, Klemets P, Miettinen I, Laaksonen I, Sarkkinen H, Hänninen ML, Rautelin H, Kela E, Nuorti JP**, 2004. An outbreak of gastroenteritis from a non-chlorinated community water supply. *J Epidemiol Community Health*, 58: 273–277.
6. **Liang JL, Dziuban EJ, Craun GF, Hill V, Moore MR, Gelting RJ, Calderon RL, Beach MJ, Roy SL**, 2006. Surveillance for waterborne disease and outbreaks associated with drinking water and water not intended for drinking--United States, 2003–2004. *MMWR Surveill Summ*. 55(12): 31–65.
7. **Markell EK, Voge M, John DT**, 1992. *Medical Parasitology*. 7th. ed. Philadelphia, WB Saunders Co. p.22–95.
8. **Nygård K, Schimmer B, Søbstad Ø, Walde A, Tveit I, Langeland N, Hausken T, Aavitsland P**, 2006. A large community outbreak of waterborne giardiasis- delayed detection in a non-endemic urban area. *BMC Public Health*, 6: 141.
9. **Slifko TR, Smith HV, Rose JB**. 2000. Emerging parasite zoonoses associated with water and food. *Int J Parasitol*, 30: 1379–1393.
10. **Stanley SL Jr**, 2003. Amoebiasis. *Lancet*, 361: 1025–1034.
11. **T.C. Sağlık Bakanlığı**, 2004. Bulaşıcı Hastalıkların İhbarı ve Bildirim Sistemi, Standart Tanı, Sürveyans ve Laboratuvar Rehberi. [Ministry of Health, Guidebook for Notification System, Standart Diagnosis, Surveillance and Laboratory of Communicable Diseases] p.6–8, p.124–137.
12. **www.izmirturizm.gov.tr**. accessed: 2006, Feb 07