Distribution of Intestinal Parasites Detected in Ankara Training and Research Hospital between 2017 and 2020

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INTRODUCTION

Intestinal parasitic infections continue to maintain their importance as a common public health problem, especially in countries with low socioeconomic status. Low level of education, inadequate sanitation of drinking water and food, and lack of attention to personal hygiene are the main factors that increase the prevalence of intestinal parasitic infections [1].

Although intestinal parasites are common, the often non-acute course of the parasitic infections may cause clinicians to skip the diagnosis [2]. However, intestinal parasitic infections are of great importance, especially in children, as they can cause malnutrition, anemia, growth and developmental delay, deterioration in cognitive skills and decrease in school success [3,4]. At the same time, intestinal parasites can cause serious and widespread infections that can be life-threatening in immunocompromised patients [5].

The most common method for the diagnosis of gastrointestinal parasites is direct microscopic examination (native-lugol). The application of concentration methods in the examination of intestinal parasites increases the chance of diagnosis, especially in stool samples with low parasite loads [7].
In this study, we aimed to determine the prevalence of protozoa and helminths in the samples sent to Ankara Training and Research Hospital Medical Microbiology Laboratory for the purpose of examination of intestinal parasites between 2017-2020 and to evaluate the distribution of these parasites according to symptoms, age, clinics and nationality.

MATERIALS AND METHODS

Ethics Committee Approval
Ethics committee approval dated 06.07.2021 and numbered E-21-644 was obtained from Ankara Training and Research Hospital Clinical Research Ethics Committee.

Selection of Patient Groups
In the study, patients who applied to our hospital with gastrointestinal complaints and asked for intestinal parasitic examination via cellophane tape method and stool concentration method by the clinicians during the three-year period between 2017 and 2020 were evaluated retrospectively. Data on demographic and clinical parameters of patients were obtained from the laboratory information management system.

Parasitological Examinations
Fresh stool samples taken into a commercial fecal concentration tube with fixative solution (Parasep® Fecal Parasite Concentrators, Apacor, USA) were delivered to the laboratory and the sediment obtained after centrifugation was examined by native-lugol microscopy to detect intestinal parasites in stool samples [8,9]. Entamoeba spp., Dientamoeba fragilis etc. suspected specimens were stained with the Wheatley trichrome staining method [10]. Cryptosporidium spp. suspicious samples were stained with the Modified Kinyoun acid-fast staining method [11]. Cellophane tape method was used to detect Enterobius vermicularis eggs [12].

Statistical Analysis
Statistical analysis was performed using SPSS 23 (IBM Inc, New York, USA). Chi-square test was used to compare the gender, age group distributions and Turkish citizenship status between positive and screened cases, and p<0.05 was considered statistically significant. Descriptive statistics were given as percentage and frequency.

RESULTS

Results of Cellophane Tape Method
A total of 2348 patients were evaluated for E. vermicularis eggs by the cellophane tape method. Of these patients, 1169 (49.8%) were males and 1179 (50.2%) were females, with a mean age of 11.99 (0-81) and 13.05 (1-79), respectively. E. vermicularis eggs were found in 169 (7.2%) of 2348 samples and Taenia spp. eggs were found in four samples (0.2%). Among patients with E. vermicularis, 88 (52.1%) were males, with a mean age of 9.53 (1-51) and 81 (47.9%) were females, with a mean age of 11.91 (3-73). The distribution of these patients by age, gender and nationality is given in Table 1. When the distribution of patients with E. vermicularis was examined in terms of symptoms/clinical diagnosis, abdominal pain was observed in 44 (26%), parasitic infection was suspected in 35 (20%), and gastroenteritis was observed in 16 (9%) patients.

Results of Stool Examination
Stool samples of 4211 patients were evaluated with native-lugol microscopic examination after concentration method. Of these patients, 2127 (50.5%) were males, with a mean age of 11.56 (0-90), 2084 (49.5%) were females, with a mean age of 12.61 (0-85). One or more intestinal parasites were detected in 765 (18.2%) of 4211 samples. Among the patients in whom intestinal parasites were detected, 394 (51.5%) were males, with an average age of 13.1 (0-76), and 371 (48.5%) were females, with an average age of 13.8 (1-78). The distribution of these patients by age, gender and nationality is given in Table 2, and their distribution in terms of symptoms/clinical diagnosis is given in Figure 1.

Blastocystis sp. was the most common intestinal parasite and detected in 611 (14.5%) of the stool samples. Wheatley trichrome stain was applied to the suspicious samples that were examined by the concentration method and D. fragilis was detected in 119 (2.8%), Entamoeba histolytica/dispar in
Table 1. Distribution of samples examined by cellophane tape method according to age, gender and nationality.

|                      | E. vermicularis positivity (n/%) | Total number of samples examined (n/%) | p value |
|----------------------|----------------------------------|---------------------------------------|---------|
| Gender:              |                                  |                                       |         |
| Female               | 81 (48%)                         | 1179 (50.2%)                          | 0.538   |
| Male                 | 88 (52%)                         | 1169 (49.8%)                          |         |
| Age:                 |                                  |                                       |         |
| <6                   | 32 (18.9%)                       | 636 (27.1%)                           |         |
| 6-18                 | 124 (73.4%)                      | 1360 (57.9%)                          | 0.000*  |
| 19-39                | 9 (5.3%)                         | 195 (8.3%)                            |         |
| ≥40                  | 4 (2.4%)                         | 157 (6.7%)                            |         |
| Nationality:         |                                  |                                       |         |
| Turkish citizen      | 160 (94.7%)                      | 2223 (94.7%)                          | 0.679   |
| Other                | 9 (5.3%)                         | 125 (5.3%)                            |         |
| TOTAL                | 169 (%7.2)                       | 2348                                  |         |

*p<0.05

Table 2. Distribution of stool samples examined for intestinal parasites by age, gender and nationality.

|                      | Number of samples with intestinal parasites (n/%) | Total number of samples examined (n/%) | p value |
|----------------------|--------------------------------------------------|---------------------------------------|---------|
| Gender:              |                                                  |                                       |         |
| Female               | 371 (48.5%)                                      | 2084 (49.5%)                          | 0.244   |
| Male                 | 394 (51.5%)                                      | 2127 (50.5%)                          |         |
| Age:                 |                                                  |                                       |         |
| <6                   | 140 (18.3%)                                      | 1337 (31.7%)                          | 0.012*  |
| 6-18                 | 534 (69.8%)                                      | 2396 (56.8%)                          |         |
| 19-39                | 48 (6.3%)                                        | 235 (5.8%)                            |         |
| ≥40                  | 43 (5.6%)                                        | 243 (5.7%)                            |         |
| Nationality:         |                                                  |                                       |         |
| Turkish citizen      | 626 (16.3%)                                      | 3831 (91%)                            | 0.240   |
| Other                | 139 (36.6%)                                      | 380 (9%)                              |         |
| TOTAL                | 765 (%18.2)                                      | 4211                                  |         |

*p<0.05

15 (0.3%) of the samples. With Modified Kinyoun acid-fast staining method, Cryptosporidium spp. was detected in one patient. The distribution of intestinal parasites detected by the concentration method is given in Figure 2.

DISCUSSION

Intestinal parasitic infections are important public health problem, especially in underdeveloped and developing countries with low socioeconomic status. Although many preventive strategies have been implemented to control these infections, the methods used to determine the prevalence of intestinal parasites are insufficient. It is important to use advanced techniques in order to accurately determine the prevalence of intestinal parasites and to develop effective control strategies [13].

In our study one or more intestinal parasites were found in 18.2% of the stool samples examined by the concentration method. Native-lugol direct microscopic examination is the most commonly used method for the detection of intestinal parasites, as it is a fast and easily applicable method. However, the sensitivity of this method is low, and the chance of diagnosis decreases, especially in samples that are not delivered to the laboratory immediately [14]. Taking the stool sample into the fixative and delivering it to the laboratory and applying the concentration method to the stool increase the chance of diagnosis of intestinal parasites [15].
The prevalence of intestinal parasites in our country varies according to the methods used in the studies and the region where the study was conducted. In the study conducted by Cengiz et al. in Van, in which 11-year retrospective data were analyzed, one or more intestinal parasites were detected in 34.1% of 69633 patients. The most common parasites were Blastocystis sp., G. intestinalis, Entamoeba coli and Ascaris lumbricoides [16]. In the study conducted by Öncel, intestinal parasites were observed in...
31.6% of 7353 stool samples examined in Şanlıurfa. The most frequently detected parasites were Blastocystis sp., E. coli and G. intestinalis [17]. In the study of Yula et al. in Mardin, intestinal parasites were observed in 27.6% of 1620 stool samples examined and G. intestinalis and T. taeniae were most commonly detected parasites. This finding was attributed to the high consumption of raw meat in the region [18]. In the study conducted by Kırkkoyun Uysal et al. in Istanbul and examining 25-year data, intestinal parasites were found in 5% of the stool samples of 111889 cases. The most common parasites were G. intestinalis and E. vermicularis [19]. In the study conducted by Gülmez et al. in Ankara, 10-year data were analyzed and intestinal parasites were found in 4.2% of 85707 stool samples [20]. It is noteworthy that the prevalence of intestinal parasitic infections is quite high in regions with low socioeconomic status in our country. In our study, when cellophane tape and stool concentration methods were evaluated together, intestinal parasite positivity was found in 14.2% of 6559 patients examined over a three-year period. The higher intestinal parasite detection rates than in similar regions, is thought to be related to the low sociocultural level of patients.

In our study, the most frequently detected protozoa were Blastocystis sp. and D. fragilis, the most frequently detected helminths were E. vermicularis and T. taeniae. Blastocystis sp. and D. fragilis are intestinal protozoans whose pathogenicities are still controversial, despite their increasing incidences in recent years. Due to the difficulties in diagnosis by routine microscopic examinations, the true prevalence of D. fragilis is not well known. In our study, D. fragilis prevalence was found 2.8% by microscopic examination. In the recent studies conducted in Turkey, the prevalence of D. fragilis was found 11.9% and 12.04%, respectively, by molecular methods [21-23].

In our study, intestinal parasite positivity was two times higher among refugee/immigrant population than in Turkish citizens. Indeed, intestinal parasitic infections are reported more prevalent in refugees worldwide. In a study conducted in Denmark, the prevalence of G. intestinalis and Blastocystis sp. was found high in asymptomatic refugee population [24]. In another study in Thailand, pregnant women from the refugee camp were found two times more likely to be infected with soil-transmitted helminth infections [25]. In the current study soil-transmitted helminths (STH) were found in any of the patients, probably due to the non-endemic living areas for STH. In a study conducted in Canada, it is reported that refugees were at greater risk of parasitic infections with a high prevalence of intestinal parasites, like as our study indicate [26].

Although parasitic infections are mostly asymptomatic, it has been reported that these infections may trigger conditions such as diarrhea, malabsorption, dyspepsia, irritable bowel syndrome or anemia [27]. In a study conducted in Turkey, it has been reported that Blastocystis sp. and D. fragilis might play a role in chronic urticaria and indicated that parasitic infections should not be neglected in patients with cutaneous manifestations [28]. But on the contrary, in another study in Iran, the prevalence of various parasites between case and control groups was not found significant [29]. In our study, it was observed that in addition to gastrointestinal complaints such as abdominal pain and gastroenteritis, non-gastrointestinal complaints such as malnutrition, growth retardation, dermatitis and urticaria were quite common in patients with intestinal parasites. It should be kept in mind that intestinal parasites may also be a factor in patients presenting with these complaints, as in asymptomatic patients.

**Limitations of the study**

Due to nature of the retrospective study, cellophane tape method and stool concentration methods could not be applied to all patients concurrently. Since the modified acid-fast staining method was not used in routine parasitological examinations, the prevalence of sporozoan parasites such as Cryptosporidium spp., Cyclospora cayetanensis and Cystoisospora belli could not be determined in our study. However, in order to determine the prevalence of these protozoa, it is important to apply the modified acid-fast method, especially in watery stool samples. Another limitation of the study was single-day examination of samples. For an ideal parasitological examination, at least three samples taken periodically should be examined.
CONCLUSION

In Turkey, intestinal parasitic infections are still an important public health issue. It has a great importance to determine the prevalence of parasitic infections to develop optimal prevention and treatment strategies. More studies with advanced diagnostic tests are needed to accurately determine the prevalence of intestinal parasites and to understand their pathogenic roles.

Author contribution

Study conception and design: FK and MK; data collection: MK; analysis and interpretation of results: FK and MK; draft manuscript preparation FK. All authors reviewed the results and approved the final version of the manuscript.

Ethical approval

The study was approved by the Non Interventional Ethics Board/Committee (Decision number: E-21-644, 06/07/2021).

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Conflict of interest

The authors declare that there is no conflict of interest.

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