Original Article

Frequency of Intestinal Parasitic Infections among Individuals Referred to the Medical Center Laboratories in Nahavand City, Hamadan Province, Western Iran

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Abstract

Background: Intestinal parasitic infections (IPIs) cause serious public health problem in the world, especially those located in tropical and subtropical areas. This study was conducted with the aim of obtaining frequency of intestinal parasites in referred people to the Nahavand city laboratories, Hamadan province, western Iran.
Materials and Methods: A comparative cross-sectional study was conducted among checkup individuals and patients referred to laboratories of Nahavand County. A total of 371 stool samples (150 from checkup individuals and 221 from patients) were selected by using systematic random sampling during summer 2014. The stool specimens were examined macroscopically, and microscopically by using direct slide smear (saline wet mount and lugol staining), formaldehyde - diethyl ether concentration, trichrome staining and modified Ziehl-Neelsen staining techniques. The results were analyzed using SPSS version 16 and Chi-square test.
Results: Ninety two patients (24.8%) were infected with single or multiple intestinal parasites. The overall prevalence of IPIs in checkup individuals and patients was 21.3% and 27.1%, respectively. The frequency of the observed intestinal parasites was: *Blastocystis spp.* 72 (19.4%), *Entamoeba coli* 7 (1/9%), *Endolimax nana* 7 (1/9%), *Giardia lamblia* 5 (1/3%), *Cryptosporidium* spp. 3 (0.8%), *Entamoeba hartmanni* 3 (0.8%), *Entamoeba histolitica/E. dispar* 1 (0.3%), *Trichomonas hominies* 1 (0.3%), *Chilomastix mesnili* 1 (0.3%), *Iodamoeba butschlii* 1 (0.3%) and *Enterobius vermicularis* egg l (0.3%).
Conclusion: The proportion of observed protozoan parasites 91 (24.5%) is higher than helminthes infection 1 (0.3%). The worm infections in Nahavand city was dramatically decreased over the past decades, induced increases in public health at the community level. *Blastocystis* spp. was the predominant intestinal parasite in people referred to the Nahavand city laboratories. Proportion of pathogenic IPIs among patients 4.07% (9 of 221) was higher in compare to the checkup individuals in which only one out of 150 (0.66%) *Giardia lamblia* was observed.

Keywords: Frequency, Intestinal parasitic infections, Nahavand, Iran

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Introduction

Intestinal parasite infections (IPIs) are still main health problems, especially in tropical and subtropical areas[1]. IPIs have many effects on quality of life around the world, especially in developing countries. The high prevalence of IPIs is closely correlated with poor sanitation, lack of access to safe water, malnutrition, improper hygiene and impoverished health services[2,3]. IPIs are most common in school age children, which are particularly associated with poor growth in children, anemia, growth retardation, reduced physical forces, and impaired learning ability[4-6].

Entamoeba histolitica (*E. histolitica*) and *Giardia lamblia* are commonly associated with travelers' diarrhea, especially among children and infected patients are either asymptomatic or suffering from mild or severe diseases. Moreover, *E. histolitica* can cause amebic liver abscess (and rarely to the lung and the brain) with about 40,000 to 100,000 deaths annually[7-9].

Cryptosporidium is known as opportunistic protozoan parasite, cause of diarrheal disease particularly in young children and immunocompromised patients with worldwide distribution. The prevalence of *Cryptosporidium* in patients with gastro-enteritis was 1-4% in Africa, Asia, Australia, South and 3-20% in Central America[10].

On the other hand according to WHO report, more than 2 billion people in the world suffering from helminthes infection especially soil-transmitted helminthiasis including *Ascaris lumbricoides* (1 billion people), *Trichuris trichiura* (795 million) and *Hookworms* (740 million)[11].

Several prevalence studies on IPIs in the past decades from different regions of Iran[12-16], and also in Hamadan province have reported[17,18]. However, no study regarding the prevalence of IPIs in Namaland County was found. Therefore, this study was conducted to determine the frequency of IPIs among individuals referred to the Nahavand's medical centers laboratories.

Methods

Population study

A total of 371 stool specimens consist of 221 patients referred for various medical reasons to the medical laboratories for stool exam and 150 individuals who referred to the health emergency medical center laboratories for checkup or given healthy card during summer 2014 by simple random selection were obtained. Demographical data including age, gender and location were collected from each subject. The mean age of checkup individuals was 30.62±16.71 years and 30.63±22.87 years for the patients referred to the laboratories.

Stool examination

Macroscopic examination

Fresh collected fecal samples were transferred daily to the parasitology research laboratory of Ayatoallah Alimoradian hospital in Nahavand County for parasitological analysis. At first all specimen were mainly examined macroscopically to determine the presence of adult worms, or segment, blood, mucosa, etc.

Microscopic examination

All the stool specimens were examined microscopically by using direct slide smear (saline wet mount and lugol staining) and formaldehyde - diethyl ether concentration. All the samples stained with a modified Ziehl-Neelsen technique for detection of *Cryptosporidium* spp. and also trichrome staining was used for accurate determination of the amoeba parasites[19]. All the slides were observed using 10X, 40X and the stained slides with 100X objectives.

Statistical analysis

Data were analyzed using SPSS software version 16.0 (Chicago, USA). Pearson-chi square tests as well as Fisher's exact test were used to test the association between IPIs and study variables and the t-test were used to compare the mean age. Statistical values were considered statistically significant when $P<0.05$.

Results

Out of the 371 samples, 150 were referred individual for medical checkup (group 1) and 221 were patients referred to laboratories (group 2). The overall

Table 1: Frequency of the IPIs among individuals referred for checkup and the patient referred to the medical laboratories in Nahavand County.

Parasites	Patients (N: 221) N (%)	Checkup (N: 150) N (%)	P value	Total (N: 371) N (%)
Blastocystis spp.	45 (20.4)	27 (18)	0.595	72 (19.4)
Entamoeba coli	4 (1.8)	3 (2)	1.00	7 (1.9)
Giardia lamblis	4 (1.8)	1 (0.7)	0.652	5 (1.3)
Endolimax nana	4 (1.8)	3 (2)	1.00	7 (1.9)
Cryptosporidium spp.	3 (1.4)	0 (0)	0.276	3 (0.8)
Entamoeba hartmanni	2 (0.9)	1 (0.7)	1.00	3 (0.8)
E. histolitica /E. dispar	1 (0.5)	0 (0)	1.00	1 (0.3)
Iodamoeba butschlii	1 (0.5)	0 (0)	1.00	1 (0.3)
Chilomastix mesnili	1 (0.5)	0 (0)	1.00	1 (0.3)
Trichomonas hominis	1 (0.5)	0 (0)	1.00	1 (0.3)
Enterobius vermicularis	1 (0.5)	0 (0)	1.00	1 (0.3)
Total	60 (27.1)	32 (21.3)	0.203	92 (24.8)

proportion rate of IPIs obtained was 24.8% (92 out of 371). The frequency of IPIs was 21.3 % (32 of 150) in checkup individuals and 27.1% (60 of 221) in the patients (Table 1).

The frequency of the microscopically observed parasites was as follows: *Blastocystis spp.* 72 (19.4%), *Entamoeba coli* 7 (1/9%), *Endolimax nana* 7 (1/9%), *Giardia lamblia* 5 (1/3%), *Cryptosporidium* spp. 3 (0.8%), *E. histolitica/E. dispar* 1 (0.3%), *Entamoeba hartmanni* 3 (0.8%), *Trichomonas homanis* 1 (0.3%),

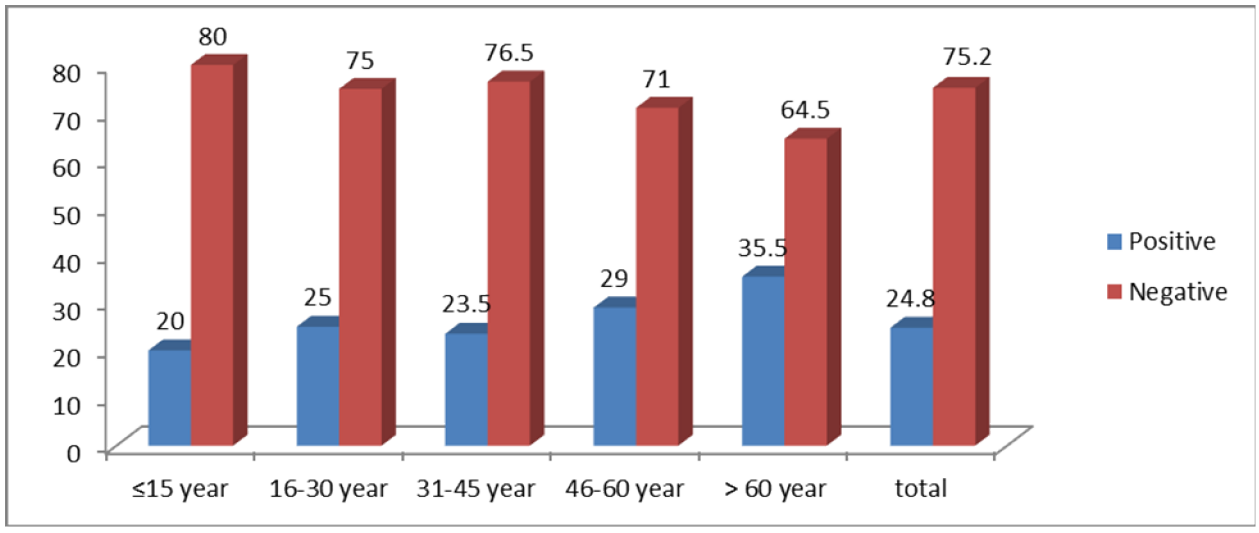

Figure 1. Frequency of the IPIs among checkup group and referred patient to the medical laboratories in Nahavand County, 2014

Table 2: Frequency of the IPIs by sex, location and age among checkup group in Nahavand County, 2014.

Variable	Positive N (%)	Negative N (%)	Total N (%)	P-value
Gender				**0.046**
Male	25 (27.2)	67 (72.8)	92 (100)	
Female	7 (12.1)	51 (87.9)	58 (100)	
Age (Year)				**0.08**
≤15	3 (9.7)	28 (90.3)	31 (100)	
16-30	13 (27.7)	34 (72.3)	47 (100)	
31-45	8 (18.2)	36 (81.8)	44 (100)	
46-60	5 (21.7)	18 (78.3)	23 (100)	
>60	3 (60)	2 (40)	5 (100)	
Location				**0.163**
Rural	20 (26.7)	55 (73.3)	75 (100)	
Urban	12 (16)	63 (84)	75 (100)	

Chilomastix mesnili 1 (0.3%), *Iodamoeba butschlii* 1 (0.3%), and just 1 (0.3%) *Enterobius vermicularis eggs* (Table1).

In stool samples of the patients 20.4% (45 of 221) *Blastocystis spp*. 1.4% (3 of 221) *Cryptosporidium* spp., 1.8% *Giardia lamblia,* 0.5% (1 of 221) *E. histolitica/E. dispar,* 0.5 % *Iodamoeba butschlii,* 0.5% *Chilomastix mesnili,* 0.5% *Trichomonas homanis* and 0.5% *Enterobius vermicularis* egg, were detected. In the checkup individuals no pathogenic parasite except one out of 150 isolates (0.66%) *Giardia lamblia* was seen (Table1). Figure 1 shows frequency of the IPIs among 371 studied individuals by sex. 20% of the positive isolates were 15 or less and 35.5 % were 60 or more years old. Frequency of the IPIs by sex, location and age among checkup group illustrate in table 2.

In this group, twenty five (27.2%) of male and 7 (12.1%) of females were infected with the intestinal parasites. 16 % of the positive persons were lived in urban and 26.7% in rural regions. Frequency of the IPIs by sex, location and age among patient illustrate in table 3.

In this group, twenty six (26.8%) of male and 34 (27.4%) of females were infected with the intestinal parasites. 30% and 24.8% among positive patients lived in urban and rural area, respectively.

Eighty two (22.1%) out of 371 studied individuals were infected by single parasites and in 10 (2.7%) isolates co-infections with two parasites were seen. *Blastocystis* spp was the predominant protozoan parasite observed in all of the co-infected isolates.

Discussion

To the best of knowledge this is the first study on intestinal parasite infections among patients referred for various medical reasons to the medical laboratories for stool exam and also in checkup individuals in Nahavand County. Totally about a quarter of studied individuals 24.8% (92 of 371) were infected with pathogenic or nonpathogenic intestinal parasites. However, there was significantly different ratio among pathogenic parasites in the two studied groups. 4.7% of the isolates in patient were positive with pathogenic parasites which is higher than 0.66% in checkups group. The most prevalent pathogenic parasites were: *Giardia lamblia, and Cryptosporidium. However,* in the checkup group only a single case of *Giardia lamblia* cyst was seen. Frequency of IPIs in patients referred to laboratories (27.1%) was higher than checkups group (21.3%). However, there was not statistically significant correlation between IPIs in two studied groups (P>0.05). The most prevalent pathogenic parasite was *Giardia lamblia* with 0.7% and 1.8% in group 1 and 2 respectively (Table 1).

Table 3: Frequency of the IPIs by sex, location and age among referred patient to the medical laboratories in Nahavand County, 2014.

Variable	Positive N (%)	Negative N (%)	Total N (%)	P value
Gender				**1.0**
Male	26 (26.8)	71 (73.2)	97 (100)	
Female	34 (27.4)	90 (72.6)	124 (100)	
Age (Year)				**0.754**
≤15	18 (24.3)	56 (75.7)	74 (100)	
16-30	10 (22.2)	35 (77.8)	45 (100)	
31-45	11 (29.7)	26 (70.3)	37 (100)	
46-60	13 (33.3)	26 (66.7)	39 (100)	
>60	8 (30.8)	18 (69.2)	26 (100)	
Location				**0.475**
Rural	30 (24.8)	91 (75.2)	121(100)	
Urban	30 (30)	70 (70)	100 (100)	

A significant association between the IPIs and sex in the checkup individuals was identified (Table 2). Frequency of *Cryptosporidium* spp. in children < 12 years was also significantly higher than adults (P<0.05). It was identified in 1.5% of the rural regions people, while was not observed in urban regions individuals. However, there was no significant difference among frequency of intestinal parasites in urban area compare to the rural in two groups of study. It can be due to close contact between urban and rural residents and the most of the urban people are also farmer and ranchers.

Protozoan parasites (24.8%) were predominant intestinal parasite in this study and *Blastocystis* spp. (19.4%) was the predominant protozoan parasites among them. *Giardia lamblia* was the most common intestinal pathogenic protozoan with the frequency of 5 (1.3%). *Cryptosporidium spp* 3 (0.8%) and *E. histolitica/E. dispar* were observed only in 1 (0.3%) of the stool samples of the patients. Only a single *Enterobius vermicularis* infection was detected in the stool sample of a referred patient. It may be more if cellophane tape especially in children was used.

This study and some reported studies in other regions[6,16] represent a dramatic decline of helminthic infections in Iran and other countries[20,21] in the past decades[14]. It may be due to increasing public and individual health and access to the safe food and water sources. Frequency of parasitic protozoan is also declining, but *Blastocystis* in the past decades was increased[18, 22]. Frequency of the obtained parasites in July (26.1) with no significance value was slightly higher than August (25.2%) and September (22.9%), P≤0.832.

Direct transmission, replicate, simple life cycle, resistance of protozoan cysts and healthy transporters, can cause higher rate of intestinal protozoa parasites infection[23].

Nahavand County is one of the leading agricultural and animal husbandry area of Hamadan province. Contact with domestic animals and soil pollutant with feces of animals cause increasing of IPIs, but the great reduction of parasitic helminthes particularly, soil-transmitted helminthes, may be due to the indiscriminate use of fertilizers and pesticides in this area. However, abundance of protozoan parasites can be due to direct transmission from human to human or contaminated food and water.

Co-infection of *Blastocystis spp.* with *E. hartmanni, E.histolitica/E.dispar, Endolimax nana, Cryptosporidium and Chilomastix mesnili is* notable, and is similar to Hamadan finding[18].

The most common protozoan was *Blastocystis spp*., which is different to some reported studies[18, 24]. Since

some investigators accept as true that this parasite is pathogen, we should be more tentative to deal with this parasite[25].

The obtained frequency of IPIs in this study in comparison with the results of some studies conducted in recent decades in other provinces of Iran, such as Mazandaran[26] and Zahedan[12] are approximately equal. However our result is different from some other parts of Iran for example, 48.8% in Bandar Abbas[24], 16.4% in Khorramabad[13], 7.6% in Kashan[27] and 35.1% in Hamadan[18]. Frequency of *Blastocystis* spp in Nahavand County found in this study (19.4%) is higher than Hamadan city (14%) which reported by Jafari et al. in 2014[18].

Additional studies with larger groups, particularly in infected patients and other population will be improved our knowledge about epidemiology of intestinal parasites in this area.

Conclusion

IPIs are highly prevalent in urban and rural region in Nahavand County. Age is an important predicator of *Cryptosporidium* spp. in 12< years old children. Also this study shows that the frequency of helminthic infections has been decreased significantly during recent years, but, intestinal protozoan parasites are still prevalent.

There was no significant statistically relationship between IPIs and age (Except for *cryptosporidium)*, gender and location (urban & rural) among patients referred to the medical laboratories. However in checkups group there was significant relationship between frequency of IPIs and gender.

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