



Intestinal Parasites Associated with Opportunistic Coccidial Infections Among Immunocompromised Individuals in Central Iran: A Cross Sectional Study

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Abstract

Background: Protozoa and helminthic parasites are the most common opportunistic parasites infections associated with the gastrointestinal tract in immunocompromised patients.

Objectives: There have been very few studies addressing this issue in central Iran and our purpose was to determine the frequency of the intestinal parasitic infections (IPIs) in different groups of immunocompromised patients admitted to the referral hospitals in Isfahan, Iran.

Methods: A cross-sectional study was performed on 204 immunocompromised patients (HIV/AIDS, lymphoma, leukemia, renal transplant and other transplants) between 2015 - 2016. Stool samples were analyzed for intestinal parasites using direct-smear, formol-ether concentration method and modified Ziehl-Neelsen staining techniques.

Results: The total rate of any parasites was 43.1% (88/204) in the patients. The prevalence of parasites was 32.7% (17/52), 39.6% (19/48), 46.2% (18/39), 56.0% (28/50), and 40.0% (6/15) in HIV/AIDS, lymphoma, leukemia, renal transplant recipients, and the other transplant recipients, respectively. *Blastocystis hominis* (30.4%), *Cryptosporidium* spp. (3.9%), *Entamoeba coli* (6.3%), *Giardia lamblia* cyst (5.4%), *Endolimax nana* (2%), ova of *Fasciola* spp. (0.5%) and *Dicrocoelium dendriticum* (0.9%) were the overall parasites that were found in this study. The most common parasites which were related to diarrhea were *Blastocystis hominis* and *Cryptosporidium* spp. The parasitic infection was significantly higher in urban patients and females ($P < 0.05$). Nevertheless, no significant relationship was observed between the prevalence of parasitic infections and age, occupation and level of education.

Conclusions: Our findings highlighted that IPIs are a common health problem among immunocompromised patients, in central Iran. Therefore these patients should be screened routinely for intestinal parasites and treated promptly.

Keywords: Intestinal Parasites, Immunocompromised Patients, Opportunistic Parasitic Infection

1. Background

According to a report of the World Health Organization (WHO) about 3.5 billion people are affected as a result of intestinal parasitic infections (IPIs) worldwide (1). Due to progressive decline in immunity status, patients with congenital/acquired immunodeficiency disorders, persons who suffer from malignancy and are undergoing chemother-

apy, organ transplant recipients and patients with human immunodeficiency virus (HIV), are highly susceptible to a variety of common and opportunistic intestinal infections.

Among the protozoa and helminthic parasites that affect the gastrointestinal system, the *Cryptosporidium* spp., *Isospora belli*, *Entamoeba histolytica*, *Giardia lamblia*, *Blastocystis hominis* and from the helminths *Strongyloides sterco-*

ralis, *Ascaris lumbricoides*, *Trichuris trichiura* and *hookworm* are the most important intestinal parasites that are the principal cause of intestinal sickness in people. These parasites cause chronic, and serious diseases that can cause iron deficiency and anemia, weight loss, tiredness and impaired mental function, especially in immunocompromised patients, helping to trap them into the health deprivation in which the disease flourishes with intense morbidity and mortality (2-4). Meanwhile, such infections can also trigger a host of other complications such as severe allergic reaction, digestive bleeding, bowel obstruction and peritonitis (2-4). Coccidial infection is a parasitic disease of the intestinal tract of human and animals caused by the Coccidian protozoa. Diarrhea, which may become bloody in severe cases, is the primary symptom. Most cases infected with coccidia are asymptomatic, but young or immunocompromised patients may suffer severe symptoms and death (1). Epidemiological reports from several regions of the world have shown a high prevalence of IPIs among immunocompromised patients (5-8). However, in Iran few studies have been conducted on the distribution and prevalence of coccidian and the other opportunistic parasites among immunocompromised individuals (9).

2. Objectives

The aim of this study was to measure of the prevalence of IPIs in patients with immunodeficiency and to identify related sociodemographic and environmental factors as well as behavioral customs in Isfahan province, Iran.

3. Methods

3.1. Ethical Consideration

The ethics board of the Isfahan University of Medical Sciences permitted the study procedures (ethical approval number: IR.MUI.REC.1396.3.305). Written informed consent was obtained from each study participant.

3.2. The Study Design and Population

A cross-sectional study was conducted on different groups of immunocompromised patients in order to determine the frequency of intestinal parasites and their relation with diarrhea. Among the three groups of immunocompromised patients, 204 fecal samples containing 52 (25.4%) HIV/AIDS individuals, 87 (42.6%) patients suffering from blood malignancy and 65 (31.8%) organ transplant recipients, were collected and stored in 2.5% potassium dichromate at 4°C. The inclusion criteria were: (1) All patients with AIDS, blood malignancies and transplanted individuals who are identified as a person with immunodeficiency according to their hospital records. (2) They should

be residents of the studied provinces. (3) In the last month they should not have used anti-parasitic drugs. The exclusion criteria were: (1) Patients who were unable to cooperate with the study for any reason. (2) Patients whose inquiries were incomplete. (3) Patients who died during the survey. (4) Patients who used anti-parasitic drugs during the last month.

3.3. Stool Examination

First, to gather socio-demographic characteristics, a comprehensive questionnaire was completed and a single fecal sample was collect in a pre-labeled, leak free, plastic specimen container from all accepting participants selected for the study. Stool samples were submitted to the parasitology laboratory of Isfahan University of Medical Sciences (Isfahan, Iran) to find intestinal parasites. Each sample was examined for intestinal parasitic microorganisms by both simple and concentration techniques. All samples were examined by microscopic observation of direct smears applying normal saline, Lugol's iodine staining and formalin ether concentration techniques (10). A permanent slide was prepared for each sample, stained with the modified Ziehl-Neelsen acid-fast technique for detection of oocysts of the opportunistic intestinal coccidian.

3.4. Statistical Analysis

Demographic records such as age, gender, job, marital status, educational level, and place of residence were collected by a questionnaire. Statistical analysis was performed by using SPSS software version 16 and chi-square and the Fisher exact test were used where appropriate to compare groups. A statistically significant P value less than 0.05 was considered.

4. Results

4.1. Characteristics of Study Participants

Important socio-demographic characteristics are given in Table 1. The total rate of any parasites was 43.1% (88/204) in the patients as well as the prevalence of protozoan parasites was 32.7% (17/52) in HIV/AIDS patients, 39.6% (19/48) in *lymphoma* patients, 46.2% (18/39) in leukemia patients, 56.0% (28/50) in renal transplant recipients and 40.0% (6/15) in the other transplant recipients (Table 1).

4.2. Prevalence of the Parasites Among the Study Participants

As shown in Table 2 the overall prevalence of the parasites found in this study included *Blastocystis hominis* (30.4%), *Cryptosporidium* spp. (3.9%), *Entamoeba coli* (6.3%), *Giardia* cyst (5.4%), *Endolimax nana* (2%), ova of *Fasciola* spp. (0.5%) and *Dicrocoelium dendriticum* (0.9%). *Blastocystis hominis* was significantly more abundant in patients than the other parasites (P = 0.003).

Table 1. Important Socio-Demographic Characteristics Among Immunocompromised Patients^a

Variable	HIV/AIDS, N = 52	Lymphoma, N = 48	Leukemia, N = 39	Renal Transplant, N = 50	Other Transplant, N = 15	Total, N = 204
Sex						
Female	24 (46.2)	23 (47.9)	12 (30.8)	26 (52.0)	4 (26.7)	89 (43.6)
Male	28 (53.8)	25 (52.1)	27 (69.2)	24 (48.0)	11 (73.3)	115 (56.4)
Age group						
≤ 45	15 (28.8)	22 (45.8)	6 (15.4)	22 (44.0)	7 (46.7)	72 (35.3)
46 - 59	27 (51.9)	13 (27.1)	21 (53.8)	20 (40.0)	7 (46.7)	88 (43.1)
≥ 60	10 (19.2)	13 (27.1)	12 (30.8)	8 (16.0)	1 (6.7)	44 (21.6)
Education						
Illiterate	15 (28.8)	7 (14.6)	6 (15.4)	2 (4.0)	1 (6.7)	31 (15.2)
Elementary school	19 (36.5)	15 (31.2)	18 (46.2)	13 (26.0)	1 (6.7)	66 (32.4)
Secondary school or higher	18 (34.6)	26 (54.1)	15 (38.5)	35 (70.0)	13 (86.6)	107 (52.4)
Residence						
Urban	42 (80.8)	39 (81.2)	38 (97.4)	49 (98.0)	14 (93.3)	182 (89.2)
Rural	10 (19.2)	9 (18.8)	1 (2.6)	1 (2.0)	1 (6.7)	22 (10.8)
Marital status						
Single	14 (26.9)	11 (22.9)	3 (7.7)	12 (24.0)	1 (6.7)	41 (20.1)
Married	28 (53.8)	33 (68.8)	32 (82.1)	36 (72.0)	13 (86.7)	142 (69.6)
Separate	10 (19.2)	4 (8.3)	4 (10.3)	2 (4.0)	1 (6.7)	21 (10.3)
Occupation						
Employee	9 (17.3)	4 (8.3)	5 (12.8)	6 (12.0)	6 (40.0)	30 (14.7)
Retired	3 (5.8)	3 (6.2)	3 (7.7)	2 (4.0)	2 (13.3)	13 (6.4)
Self-employed	17 (32.7)	15 (31.2)	20 (51.3)	15 (30.0)	3 (20.0)	70 (34.3)
Housekeeper	15 (28.8)	21 (43.8)	10 (25.6)	23 (46.0)	3 (20.0)	72 (35.3)
Unemployed	8 (15.4)	5 (10.4)	1 (2.6)	4 (8.0)	1 (6.7)	19 (9.3)
Infection						
Parasited	17 (32.7)	19 (39.6)	18 (46.2)	28 (56.0)	6 (40.0)	88 (43.1)
Non-parasited	35 (67.3)	29 (60.4)	21 (53.8)	22 (44.0)	9 (60.0)	116 (56.8)

^aValues are expressed as No. (%).**Table 2.** The Overall Prevalence of the Parasites Found in This Study^a

Parasite	HIV/AIDS, N = 52	Lymphoma, N = 48	Leukemia, N = 39	Renal Transplant, N = 50	Other Transplant, N = 15	Total, N = 204
<i>Blastocystis hominis</i>	16 (30.7)	14 (29.2)	14 (35.9)	14 (28.0)	4 (26.7)	62 (30.4)
<i>Entamoeba coli</i>	2 (3.8)	2 (4.2)	2 (5.1)	7 (14.0)	0 (0.0)	13 (6.3)
<i>Giardia lamblia</i>	0 (0.0)	2 (4.2)	1 (2.6)	8 (16.0)	0 (0.0)	11 (5.4)
<i>Cryptosporidium spp.</i>	0 (0.0)	1 (2.1)	2 (5.1)	3 (6.0)	2 (13.3)	8 (3.9)
<i>Endolimax nana</i>	1 (1.9)	2 (4.2)	1 (2.6)	0 (0.0)	0 (0.0)	4 (2.0)
<i>Dicrocoelium dendriticum</i>	1 (1.9)	0 (0.0)	0 (0.0)	1 (2.0)	0 (0.0)	2 (0.9)
<i>Fasciola spp.</i>	0 (0.0)	0 (0.0)	1 (2.6)	0 (0.0)	0 (0.0)	1 (0.5)

^aValues are expressed as No. (%).

4.3. Factors Associated with Parasite Infection

The percentages of parasites according to sex, age, and educational levels are summarized in Table 3. According to Table 3, the parasitic infection in females (53.9%) was more than males (34.8%) ($P = 0.006$). In addition, interestingly, the infection level was significantly higher in urban patients (46.2%) than the rural (18.2%) ($P = 0.01$). Nevertheless, no significant relationship was observed between the

prevalence of parasitic infections and age, occupation and level of education.

4.4. Clinical Characteristics

Among the samples, 24 (11.7%) were categorized as diarrheic stools and 180 (88.2%) were not. The most common parasites which were related to diarrhea were *Blastocystis hominis* and *Cryptosporidium spp.* with the prevalences

Table 3. Sociodemographic Characteristics of Immunocompromised Patients, According to the Presence or Absence of Parasites, Isfahan, Iran, 2015/2016^a

Variable	Parasited, N = 88	Non-Parasited, N = 116	Total, N = 204	P Value
Sex				0.006
Female	48 (53.9)	41 (46.1)	89 (43.6)	
Male	40 (34.8)	75 (65.2)	115 (56.4)	
Age group				0.80
≤ 45	29 (40.3)	43 (59.7)	72 (35.2)	
46 - 59	40 (45.5)	48 (54.5)	88 (43.1)	
≥ 60	19 (43.2)	25 (56.8)	44 (21.5)	
Education				0.22
Illiterate	14 (45.2)	17 (54.8)	31 (15.1)	
Elementary school	33 (50.0)	33 (50.0)	66 (32.3)	
Secondary school or higher	41 (38.3)	66 (61.6)	107 (52.4)	
Residence				0.01
Urban	84 (46.2)	98 (53.8)	182 (89.2)	
Rural	4 (18.2)	18 (81.8)	22 (10.7)	
Type of stool				0.048
Normal	74 (41.1)	106 (58.9)	180 (88.2)	
Diarrhea	14 (58.3)	10 (41.7)	24 (11.7)	

^aValues are expressed as No. (%).

of 30.4% (13 of 24 cases) and 20.8% (5 of 24 cases) respectively, as well as the highest rate of diarrhea was in organ transplant recipients, (46.7%) and the lowest in HIV/AIDS patients (13.5%). The uppermost frequency of parasitic infections was in renal transplant recipients (56%) and the lowest in HIV/AIDS patients (32.7%).

5. Discussion

According to our findings the overall frequency of the parasitic microorganisms was 43.1% in the immunocompromised patients. Many studies have been conducted on the prevalence of gastrointestinal parasites in Iran (4, 11-13), but conforming to our knowledge in the central Iran region, few studies have been done on IPIs in immunocompromised patients. As reported by an early study (1997) that was performed in Isfahan, 51.9% of the studied children were infected by one or several intestinal parasites and the most prevalent intestinal parasites was *Giardia lamblia* (29.8%), *Entamoeba coli* (17.1%) and *Enterobius vermicularis* (16.3%), respectively. Those study results emphasized that there is not a significant difference between the prevalence of parasitic infections in rural and urban parts of Isfahan province (14) which is not in agreement with our findings, as in our study the infection level was significantly higher in urban patients (46.2%) than the rural (18.2%) ($P = 0.01$). In the study of Azami et al. in Isfahan area, out of 150 renal transplant recipients, 33.3% (50), were infected with one or more type of intestinal parasites. In this study, *Entamoeba coli* (10.6%) was the first most prevalent parasite detected in patients followed by *Endolimax nana* (8.7%)

and *Giardia lamblia* (7.4%). This study stressed the importance of testing for intestinal parasites among renal transplant recipients (15). In another study, from a population of 100 children of 3 to 6-year-old kindergarteners in Isfahan, 19 children were found to be infected with parasites as *Enterobius vermicularis* (8.0%), *Giardia lamblia* (4.0%) and *Hymenolepis nana* (2.0%) were most frequent parasites (16), while in our study, the presence of ova of helminths was lower (*Fasciola* spp. and *Dicrocoelium dendriticum* 0.5% and 0.9%, respectively). In a study carried out by Emami et al. on kidney transplant recipients, a total of 150 kidney transplant recipients were studied and the overall prevalence of IPIs was 33.3%. In addition, the most prevalent intestinal parasite was *Entamoeba coli*, which was seen in 9.3% of the study patients (17). In the study of Mohaghegh et al. in the Isfahan region, out of 330 dialysis patients, (3.0%) were infected with *Cryptosporidium*. In this study, there was a significant difference between age and infection (18).

IPIs are a considerable public health problem across the world with a rich variety of prevalence among disadvantaged communities. The rate of infection in Ethiopia, India, Egypt, Cameroon, Malaysia, Indonesia, China, Iran, Australia, and Turkey was 35.8%, 60.5%, 94%, 59.5%, 37.9%, 57.8%, 4.3%, 33.3%, 50%, and 76%, respectively (15, 19-27). Our study shows that the infection level was significantly higher in urban patients (46.2%) than the rural (18.2%) ($P = 0.01$). This unusual and confusing outcome can be due to a number of challenges and barriers to good hygiene among low income marginalized people existing in megacities like Isfahan. However, our result is not in agreement with

reports elsewhere (11, 13, 15). We analyzed our findings by gender, thus, the parasitic infection in females (53.9%) was more than males (34.8%) with a statistically significant difference ($P = 0.006$) that is in contrast with the results of previous reports (4, 17, 28, 29). In this study, patients were classified for age, educational levels and occupation (Tables 1 and 3). However, for none of the above mentioned variables, there was no statistically significant difference with parasitic infection, while different studies reported an opposite association between educational levels and IPI rates (30, 31).

The limitation of our study should be acknowledged. Firstly, we performed a descriptive sectional study. Secondly, due to financial problems and lack of patient compliance, only one stool sample was collected from each patient. Finally, due to cultural and social issues, many patients, especially AIDS-related individuals, did not want to collaborate with the study.

5.1. Conclusions

Iran is a developing country, located in the Middle East and IPIs are still a considerable public health problem in several areas including Isfahan. In our study, the prevalence of IPIs among immunocompromised patients was moderately high and dominated by *Blastocystis hominis* and *Cryptosporidium* spp. and was more frequent in renal transplant recipients. In conclusion, the importance of IPIs in immunocompromised patients must not be neglected and we recommend additional, larger, well-designed studies to determine the prevalence of these microorganisms to be performed.

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Footnotes

Authors' Contribution: Shahrokh Izadi designed the study, collected both the field and lab data, and was involved in the analysis and manuscript preparation. The original research was the requirement for his PhD in Medical parasitology. Seyed-Hossein Hejazi, Zahra Ghayour-Najafabadi and Maryam Yavari were primary thesis advisors. Dhammika Leshan Wannigama, Mehdi Azami and Mohammad-Ali Mohaghegh reanalyzed the data and were

involved in the preparation of the manuscript. All authors read and approved the final manuscript.

Conflict of Interests: It is not declared by the authors.

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