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Intestinal parasitic infections in patients attending a tertiary care centre

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Introduction: Intestinal parasitic infections are prevalent worldwide with a range of 11- 90% in developing countries like India. The clinical presentations vary from self-limiting diarrhea in immunocompetent patients to profuse diarrhea in immmunocompromised individuals. Objectives: To determine the prevalence of intestinal parasites in patients presenting with diarrhea. Material and Methods: Stool specimens from patients with diarrhea were received in the department of Microbiology, G.I.P.M.E.R during 6 months duration from September 2017 to March 2018. All specimens were examined grossly for the color, consistency and presence of any visible adult worms or their segments. Microscopic examination was done by direct saline/iodine wet mount. The specimens from immunocompromised patients were subjected to Formol-ether concentration technique and Modified Ziehl Neelson's stain. Results: Total 57 stool specimens were received from 34 female and 23 male diarrheic patients. Majority of cases were in the 31-40 years age group. Intestinal parasites were detected in 10 specimens, out of whom 7 were Protozoa and 3 were Helminths. Over all, Cryptosporidium oocysts were the most common. Among the 10 positive cases, 7 were immunocompromised with 4 cases of AIDS/HIV, 2 of Ulcerative colitis and one case with Myasthenia gravis and Diabetes mellitus. Conclusion: The prevalence of parasitic infections indicates the socioeconomic status of the society. Recently many studies across India have reported protozoa to be common than the other intestinal parasites. The preponderance of coccidians in the present study highlights the importance of aggressive screening of these parasites, especially in immunocompromised group of patients.

Keywords: Parasitic, Cryptosporidium oocysts, Cystoisospora belli

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Introduction

Intestinal parasitic infections are prevalent worldwide and are one of the major health challenges faced by developing countries due to large population size, poverty, malnutrition, lack of availability of proper sanitation facilities and poor personal hygiene conditions [1,2]. Earlier, these infections were considered to be restricted to the developing countries, but now with the increase in the immigration, cases have been reported from the developed world too.

Worldwide, around 450 million people are infected by the intestinal parasites with a range of 11- 90% being reported in developing countries like India. [3, 4]. Intestinal parasites causing infections belong to the Protozoa and Helminth classes of parasites. Protozoa are unicellular structures transmitted by food and water contaminated with their cysts or oocysts. Most common Protozoa causing intestinal infections are Entamoeba histolytica/dispar, Giardia lamblia, Balantidium coli, Cryptosporodium species, Cyclospora cayetanensis, Cystoisospora belli and Microsporidia species. As per a WHO report, Entamoeba histolytica/ dispar was the third most common protozoa causing intestinal infections [5].

helminths include Ascaris *lumbricoides* (roundworm), Ancylostoma duodenale Trichuris trichiura, (Hookworms), Enterobius vermicuralis, Hymenolepis nana and Strongyloides stercoralis. Ascaris lumbricoides and Hymenolepis nana are the most common helminths affecting approximately1 billion people [5]. In recent years, parasitic infections in immunocompromised patients are being increasingly reported. The various immunocompromised conditions associated with the intestinal parasitic infections are HIV/AIDS, Human T-Cell Lymphotropic Virus-(HTLV-1), corticosteroid treatment, malignancy, organ transplantation, malnutrition, hypogammaglobuli-naemia, collagen/ vascular diseases and metabolic disorders.

Coccidian parasites were infrequently reported before the era of AIDS/HIV emergence.

Among the coccidians, Cryptosporidiosis was classified as one of the AIDS defining illnesses [6]. The infection dose of these opportunistic parasites is as low as 10 oocysts [7].

Other than the coccidians, various other parasites reported to cause intestinal infections in immunocom-promised groups are *S. stercoralis*,

G.lamblia/ intestinalis, E.histolytica, Blastocystis species, A. lumbricoides, Hookworms, H.nana etc [8].

The clinical presentation of enteric parasitic infections vary from short term, self-limiting diarrhea in immunocompetent patients to profuse, life threatening diarrhea in immunocompromised individuals.

Enteric parasitic infections continue as a major health problem in endemic countries like India. Within our country, prevalence and distribution of these parasites varies with the different geographical region. This study was conducted to determine the prevalence and pattern of distribution of the parasites in diarrheic patients attending tertiary care centre.

Material and Methods

Type of study: This was a cross sectional study carried at the department of Microbiology, GIPMER, Delhi.

Duration of study: 6 months from September 2017 to March 2018.

Sampling method: convenient sampling method was used where in all samples received in the above mentioned duration were included in the study.

Inclusion criteria: Patients with diarrhea, with or without fever, vomiting, weight loss, loss of appetite and any associated abdominal symptoms were included in the study.

Exclusion criteria: Patients receiving antiprotozoal or anti- helminthic drugs in previous 3 months period were excluded from the study.

Ethical permission: Permission of the Ethical committee of the G.I.P.M.E.R, Delhi was obtained.

Collection and processing of specimen: Stool sample from the patients were collected in clean, wide-mouthed container. Processing of the samples was done in the department of Microbiology, GIPMER, Delhi.

The samples were processed within 2 hours. The samples where delay in transport was assumed were preserved in 10% buffered formalin.

Macroscopic examination: Stool samples were examined macroscopically for color, consistency, and presence of blood, mucus, adult worms or their segments.

Microscopic examination: Microscopic examination was done for presence of motile trophozites, cyst, oocyst, pus cells and red blood cells.

The methods employed were Direct wet mount (saline/iodine) with and without Concentration technique of Formol ether sedimentation method and special staining like modified Ziehl Neelson stain for oocyst of coccidian parasites.

Results

A total 57 stool specimens were examined from 57 cases during the 6 months duration. Majority of cases were in the age group of 31-40 years (29.82%) followed by 41-50 years (24.56%). In the study, 34 (59.64%) were female and 23 (40.35%) were males with a male-female ratio of 0.67:1.

The prevalence of parasitic infections in patients with diarrhea was 17.54%. Out of these 10 cases, 7 different species of intestinal parasites were identified (Figure 1-6). The Protozoa (70%) were more common than the helminths (30%).

Overall, *Cryptosporidium species* were the most common observed parasites followed by *Isospora species*. (Table 1) Among the 10 positive cases, 7 had associated immunocompromised conditions.

Majority of immunocompromised patients had HIV/AIDS (57.14%) followed by ulcerative colitis (28.57%) (Table 2).

Table-1: Frequency of the parasites isolated.

Type of parasite detected	Frequency (n=10)		
Cryptosporidium species	3		
Isospora belli	2		
Giardia lamblia	1		
Entamoeba histolytica/dispar	1		
Hymenolepis nana	1		
Ascaris lumbricoides	1		
Strongyloides stercoralis	1		

Table-2: Distribution of parasites in Immunocompromised conditions

Parasites isolated	Immunocompromised conditions		
	AIDS/HIV	Ulcerative colitis	Myasthenia gravis
	(N=4)	(N=2)	(N=1)
Isospora belli	2		
Cryptosporidium sp	1	2	
Giardia lamblia	1		
Strongyloides			1
stercoralis			



Figure-1: Cysts of Entamoeba histolytica in saline wet mount (10 X magnification)

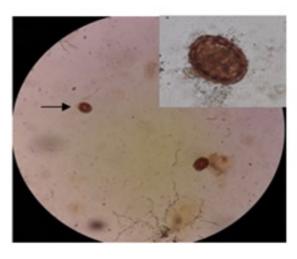


Figure-2: Eggs of Ascaris lumbricoides in saline wet mount (10 X magnification)>

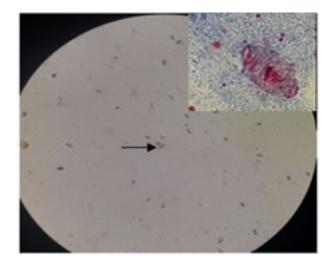


Figure-3: Oocyst of Isospora belli in saline wet mount (10 X magnification) and Kinyon's stain 100 X magnification (inset)

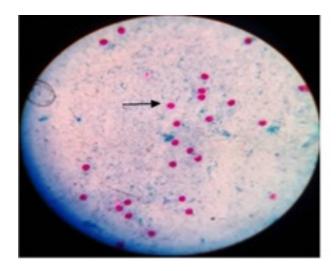


Figure-4: Oocysts of Cryptosporidium parvum in Modified Kinyon's stain (100 X magnification)

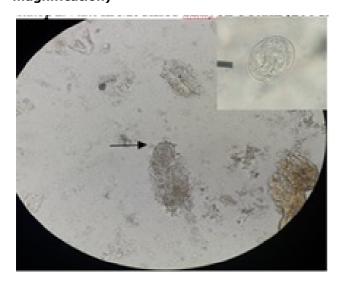


Figure-5: Eggs of Hymenolepis nana in saline wet mount (40 X Magnification)

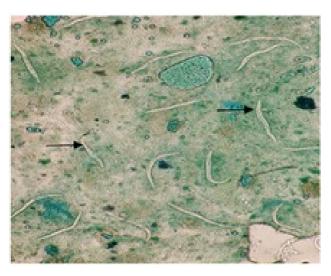


Figure-6: Rhabditiform larvae of Strongyloides stercoralis (Methylene blue stain 100 X magnification)

Discussion

Hot and humid climate of the tropical and subtropical areas is conducive for the growth and development of parasites and our country is no exception. In the present study, most of the cases were adults with age group of 31-40 years being the most common. Similar results are reported by Manuchitra K et al and Kamki Y et al from Imphal whereas a study by Shobha M and Mathuria YP noted maximum patients to be children in the age group of 0-10 years and 5-14 years respectively [1, 5, 9, 10]. In the present study females were more commonly affected than males. This is in contrast to many studies from across the country that report enteric parasitic infections more common in males [3, 10]. A few studies have reported findings similar to the present study [5, 11].

The prevalence of enteric parasitic infections in the present study was 17.54%. This rate is similar to the studies by Shobha M and Kumar S but less than those reported from other parts of the country [1, 3, 5, 12]. Wide variations from 11% to 97% in prevalence of enteric parasitic infections have been reported from different regions of India [1].

The present study was conducted in a super specialty hospital encompassing various medical and surgical departments. Patients with the chief complaints of diarrhea with no underlying comorbidity mainly attend the general hospitals. Thus, the load of such patients in our hospital is less and so is the prevalence of enteric parasitic infections. In the present study, maximum cases presenting with diarrhea were from the gastrosurgery and gastroenterology departments.

Majority of these cases were diagnosed with certain gastrointestinal conditions which have diarrhea as one of the associated complaints. The distribution of parasites detected showed that infection with the protozoan (70%) was more common as compared to the helminths (30%). Since the era of our earliest recorded history, helminthic infections were more prevalent in the tropical and sub-tropical developing countries [13].

However, recent reports from across the country and globe, have reported the higher prevalence of protozoa than the helminthes [1,5,11,14]. Protozoa are the unicellular parasites that require human body for their multiplication and existence as human to human transmission occurs through the feco-oral route.

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Many reports elucidate *E. histolytica* and *G. lamblia* as the common protozoa [1, 2, 15]. In the present study, among the protozoa as well as when total number of parasites were considered, *Cryptosporidium species* were the most common 5 (50%) followed by *Isospora belli* (20%).

The higher percentage of these coccidians can be explained by the fact that the present study group included both immunocompetent and immunocompromised cases with the higher number of immunocom-promised individuals (70%). Cryptosporidium species were isolated from HIV/AIDS and ulcerative colitis cases whereas Isospora belli from HIV/AIDS. A study by Kaur R et al also reported a high isolation rate of Cryptosporidium species as compared to the other protozoa. [16].

With the advancement in diagnostic and therapeutic medical science come the detrimental effects of introducing various immunocompromised conditions. Over the last two decades. Cryptosporidiosis is being increasingly reported in developed and developing countries. Though, Cryptosporidiosis is one of the opportunistic pathogen in the immunocompromised groups, its infection in the immunocompetent individuals is not uncommon [17]. Various factors contribute towards the ubiquity and pathogenicity of Cryptosporidiosis like zoonotic nature, lack of host specificity, very low infectious dose (< 10 oocysts), oocysts highly resistant to the common disinfectants (chlorine) and discharged in large numbers from severely infected individuals and ability to cause autoinfection [7].

Helminthic infections affect 2 billion of the world's population with India contributing to nearly 25% of this burden [18]. Though, India is an endemic region for these parasites, only three cases (30%) were observed infected with helminths in the present study, one case each by *A. lumbricoides, H. nana* and *S. stercoralis.* This may be due to the fact that the predominantly affected strata in the present study were adults and helminthic infections are more commonly found in children.

Around 85% of studies from different regions of India report that soil-transmitted helminthic infections are common in young children. Also, Delhi state falls in the zone of less than 20% affected region by the helminthes [18]. Furthermore, WHO recommends the Kato-Katz technique as the most reliable, accurate and effective diagnostic tool for the helminths.

The present study had the limitation of not using this technique. These could be the reasons for the lower prevalence of helminthic infection in the present study. Similar low rate of infection by helminths is reported by Kaur R et al from Delhi [16].

The helminthic infections are transmitted through contaminated soil, either through ingestion of infective eggs or penetration of the skin by larvae. Preventive strategy for soil transmitted helminthic infections recommended by WHO include deworming with a single dose of albendazole or mebendazole to the high risk population and improved hygiene and sanitation facilities. The health program under NRHM that provides deworming biannually, has contributed further towards the low prevalence of helminthic infections.

The number of immunocompromised patients attending the hospitals is increasing day by day. In the present study, out of 10 cases infected with the enteric parasites, 7 cases had underlying immunocompromised conditions of which HIV/AIDS was the most common followed by ulcerative colitis. Many other studies have also reported AIDS as the most common immunocompromised condition whereas some other studies state malignancy as the commonest condition [4, 9, 19].

The most common parasite detected in AIDS was Isospora belli followed by Cryptosporidium species and *G.lamblia*. This finding is consistent with the study by Kumar SS et al whereas Vanathy K et al surprisingly did not find a single case of Cryptosporidiasis in this group of patients [19, 20]. Though the prevalence of coccidians reported in AIDS patients is high, is still assumed to be under reported owing to the asymptomatic and irregular shedding of oocysts, prophylactic treatment with Trimethoprim/ sulpho-methoxazole in these patients that provide protection against coccidians like *Isospora* and lack of availability of accurate and standard diagnostic modalities like PCR. [20].

The diagnosis of the coccidians is mainly based on the conventional methods of microscopy in many resource poor settings. The results by these methods are prone to misdiagnosis as it requires good expertise. In present study, It was found that 2 cases of ulcerative colitis and one case of myasthenia gravis affected by *Cryptosporidium species* and *S. stercoralis* respectively. Very less data is available on Cryptosporidiasis in Ulcerative colitis from India and across the world. [21, 22].

A study from Mexico found protozoa like *Blastocystis hominis, Endolimax nana* and *E.histolytica* as the parasites in ulcerative colitis patients (23) whereas a case report has published the occurrence of *Giardia* in ulcerative colitis [24]. In the present study, the case affected with *Strongy-loides* larvae had myasthenia gravis along with diabetes mellitus. Few case reports have reported this parasite in these autoimmune conditions. [25, 26, 27].

The most commonly used anti-protozoal drugs are nitroimidazoles like metronidazole or tinidazole, nitazoxanide, trimithoprim- sulfamethoxazole, paramomicin etc while albendazole, mebendazole, pyrantel pamoate etc are the commonly used anti-helminthic drugs. The cases in the present study were mostly treated with metronidazole for protozoal infections and albendazole for helminthic infections.

Conclusion

Among the pathogenic enteric parasites, the protozoans like coccidians predominate. Though the diagnostic technology has evolved with time, it largely depends on conventional methods in resource constraint settings. An accurate diagnosis is of utmost importance as treatment varies as per the infecting parasite. Furthermore, conditions like Cryptosporidiosis should be diagnosed early as they are responsible for relapse as well as exacerbation of symptoms especially in immunocompromised conditions.

What this study adds to existing knowledge?

Majority of positive cases detected in the present study were affected with some immunocompromised conditions. Thus, in the present study highlights that the occurrence of intestinal parasites is increasing in this group of patients. Many studies have demonstrated the occurrence of these intestinal parasites in HIV/AIDS patients.

The present study reports the existence of these pathogens in various other immunosuppressed conditions like ulcerative colitis, myasthenia gravis etc. The clinical presentation of these intestinal disorders is with non-specific symptoms. Hence, the diagnosis mainly depends on the high clinical suspicion and reliable laboratory techniques requiring expertise.

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