



Original Research Article

Prevalence of parasitic infection and comparison of different types of concentration techniques

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ABSTRACT

Background: The intestinal parasitic infection is the major problem in the developing countries. The prevalence depends on not only the geographical location but also various socioeconomic factors such as climate, hygiene and age.

Material and Methods: A cross sectional study was conducted at Santosh Medical College, Ghaziabad during Jan. 2019 to June 2019. Total 200 stool samples were collected and each was examined by direct wet mount (iodine & saline mount) and concentration techniques such as simple salt flotation and formal ether concentration.

Results: In the present study the prevalence of the intestinal parasitic infection was found 34%. The most common parasite was found to be *Entamoeba histolytica*, with the prevalence rate of 57.35%, followed by *Giardia lamblia* 39.71%. A male predominance was noted 37.70% over the female 28.20%. The highest prevalence of parasitic infection was found in the age group between 21-30 years of age 42.85% followed by in age group 31-40.

Conclusion: In this study it was concluded that the formal ether sedimentation technique shown a high sensitivity for the parasite detection in comparison to the normal saline wet mount and salt flotation technique.

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1. Introduction

Parasitic infections are a major public health problem globally; especially in the developing countries.¹ The parasitic infection, prevalence varies with the level of sanitation and is generally higher in the tropics and sub-tropics in comparison to temperate climates.^{2,3}

It is estimated that 3.5 billion peoples are affected worldwide, and that 450 million are ill as a result of these parasitic infections, the majority being suffered are children. Recent studies suggest that even moderate intensity of infection may have adverse effects on growth, iron deficiency anemia and cognitive function, practically for children of school age.^{4,5} Intestinal parasitic infections causes diarrhea, dysentery, weight loss, malnutrition, anemia,

abdominal pain, and other gastrointestinal ailments. Chronic parasitic infections also impairs physical development and cognitive functions of growing children.^{6,7} The prevalence of intestinal parasitic infections not only varies in different parts of the world but also in different regions of same country.⁸

The direct wet mount and the iodine mount are the conventional methods which are used for the detection of intestinal parasites from stools samples. These methods have less sensitivity in the detection of parasites in the stool specimens (eggs, cysts, trophozoites and larvae).

The parasites detection in the stool samples is increased by the use of concentration procedures such as formal-ether concentration, formalin-ethyl acetate concentration, formalin-acetone sedimentation and saturated salt flotation, zinc sulfate flotation techniques. Concentration techniques increase the detection of the helminthic eggs, larvae and the

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protozoan cysts in stool samples. In view of the increasing parasitic infection burden in developing countries, required a sensitive diagnostic test, those are simple, easy to perform, cost effective to be used.

2. Materials and Methods

A cross sectional study was conducted in department of Microbiology in a tertiary level care hospital in Ghaziabad during the period from Jan. 2019 to June 2019. 200 stool samples were collected from symptomatic and asymptomatic cases who reported to the microbiology laboratory. Stool samples were collected in sterile container and all individuals were requested to provide a morning fecal sample, avoiding after antiparasitic drugs and avoiding contamination with urine. Those stool samples, which were contaminated with urine of patient were not accepted to include in study. Formed and unformed stools samples were examined immediately after receiving in the laboratory.

Stool samples were examined by the following methods:

2.1. Macroscopic examination

Macroscopically stool were examined to see the color, consistency and the nature of the stool.

2.2. Microscopic examination

2.2.1. Direct wet mount

A drop of saline and Lugol's iodine are placed on left and right halves of the slide respectively, a small amount of feces (~2mg) is mixed with a stick to form smooth suspension and examined under low power objective at 10X and 40X.

2.2.2. Formal-ether sedimentation techniques:

$\frac{1}{2}$ teaspoon of fresh stool into 10 ml of 5% or 10% formalin in a 15 ml shell vial, let stand for about 30 minutes for adequate fixation. Filter all the material through two layer of gauze into a 15 ml centrifuge tube. Add normal saline or 5% or 10% formalin to within 1, 2 inch of the top and centrifuge for 10 minutes at 500Xg and wash until the supernatant fluid is lighter tan or clear. Add ~3 ml of ethyl acetate solution in tube and stopper to be applied to the tube, shake it vigorously at least for 30 seconds, and then centrifuge the tubes for 10 minutes at 500Xg. Four layers formed the sediment at the bottom of tube to be used for microscopic examination. A plug of fecal debris on top of the formalin layer; and a layer of ether substitute at the top. After proper decanting, a drop or two drop of fluid remaining on the side of the tube drains down to the sediment. Mix the fluid with the sediment and prepare a wet mount for examined.

2.2.3. Saturated salt flotation technique

In a 20 ml conical glass test tube 1 gm of stool sample was emulsified in 3-4 ml of saturated salt solution. The solution

was stirred properly and more salt solution was added until the container was nearly full upto the top. Then a glass slide were placed on the top of the tube. The slide should remain in contact with the fluid in tube. Then slide was allowed to stand for 30 minutes in contact with the fluid. After that remove the slide and place a cover slip over it and examined under the microscope for the presence of ova, cyst to detect the oarasite.

3. Result

200 stool samples were examined, out of which 68 (34%) samples were found positive for intestinal parasites. Overall prevalence of male and female was 37.70% and 28.20%. Males were more infected compare than females. The prevalence rate in all age group is higher under 21-30 years age 27 (42.85%).

Among the parasites *Entamoeba histolytica* was the found to be the most common parasite 57.35%, followed by *Giardia lamblia* (39.71%), *Hymenolepis nana* (1.47%) and *Entamoeba coli* (1.47%) respectively as shown in Table 1. In the study, it was found, some cases of double intestinal infection, the prevalence rate of mixed infection *Entamoeba histolytica* and *Giardia* 8 (11.76%).

Formal-ether sedimentation technique is more sensitive 60.29% in comparison of saturated salt flotation technique 48.52% and direct wet mount 38.23% was less sensitive.

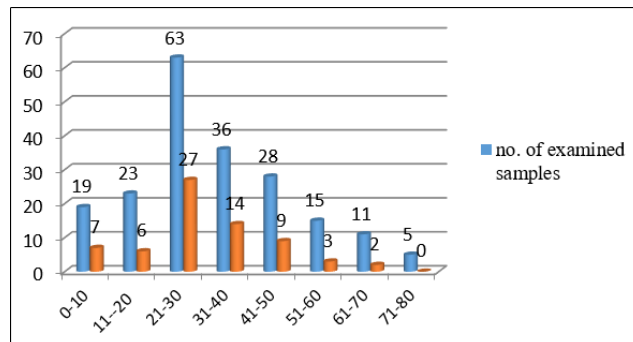


Fig. 1: Age-wise prevalence of parasitic infection

4. Discussion

In India, different studies have shown different prevalence rates of the parasitic infection. In present study, 200 samples were included to find out the parasitic infection and comparison the concentration technique. In this study, overall prevalence rate is 34% [Table 2], which was higher than previous study in India 15.19% to 29.2% and in Turkey 13.8% and 22.4%.^{5,9,10} In contrast. It was notably high in studies 46.50% - 71.2%.^{11,12} This high prevalence of intestinal parasitic infection may be due to the lack of awareness about personal cleanliness, hygiene and illiteracy

Table 1: Prevalence of parasitic infection

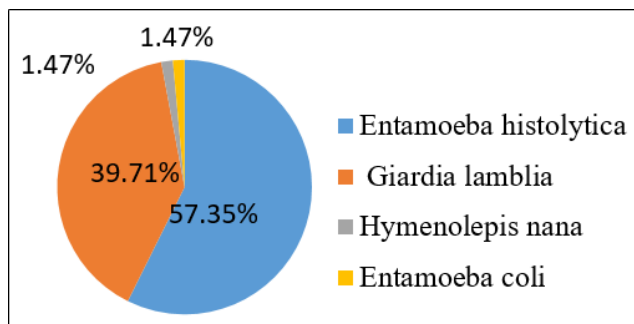
S. No.	Name of Parasite	No. of infected patients (n=200)	Prevalence (%)
1	Pathogenic Parasite Entamoeba histolytica	39	57.35
2	Giardia lamblia	27	39.71
3	Hymenolepis nana	1	1.47
4	Non Pathogenic Parasite Entamoeba coli	1	1.47
Total		68	100

Table 2: Sex wise prevalence of parasitic infection

S. No.	Sex	No. of examined patient	No. of infected patient	% of infected patient
1	Male	122	46	37.70
2	Female	78	22	28.20
Total		200	68	34

Table 3: Sensitivity of different methods of parasitic examination

Techniques	No. of positive for parasite (N= 200)	Percentage (%)
Direct wet mount	26	38.23
Saturated salt flotation	33	48.52
Formalin- ether sedimentation	41	60.29

**Fig. 2:** Parasite wise distribution of intestinal parasites

among rural population. This wide variation among studies could be attributed to the time and period of study, the age of the study population, variation in diet, habits and occupations, different sampling techniques and research methodologies, geographical differences and the inclusion of non- pathogenic intestinal parasites in the analysis. The parasitic infection rates were based on the examination of a single stool specimen per individual.

In the present study, Entamoeba histolytica 57.35% was found to be the most prevalent parasite followed by Giardia lamblia 39.71% [Table 1 and Figure 2]. Where as the lower prevalence were found by the studies done by Marothi Y et al.,¹³ and Bisht D et al.¹⁴ Entamoeba histolytica was reported to be responsible for approximately 50 million cases of invasive amoebiasis and upwards of 100,000 death/year (WHO 1997).

The prevalence rate was found higher in males 37.70% as compared to the females 28.20% (Table 2). The finding was comparable to the study of Parmeshwarappa KD et al.¹⁵ In their study, they found the prevalence rate is higher in males 33.39% as compared to females 21.29%. While other studies showed increased infection among the females, (Hamad and Ramzy 2012) in Erbil¹⁶ and Raza 2009 in Sulaimani.

The patients were from the age group between 0 to 80 years. The highest prevalence of parasitic infection was in the age group of 21-30 years as compared to study of Ahmed et al.¹⁷ The high incidence of intestinal parasitic infection in males in the age group of 21-30 years is probably due to outdoor eating and drinking habits, which increases to the transfer there from, to the mouth (Figure 1).

The routine stool wet mount examination to find out the parasitic burden lacking the sensitivity. The stool concentration technique should be perform routinely to find out the correct burden of parasites. These procedures allow the increased recovery of egg and protozoal cysts, the trophozoites are destroyed during concentration procedure. In the present study it was shown that, 60.29% of the cases were detected by the formal-ether method, followed by simple salt flotation concentration technique 48.52% and then direct wet mount techniques which showed the lowest detection rate 38.23%, These results were compared with the other study done by Parameshwarappa KD et al.,¹⁵ which showed that the formal-ether sedimentation concentration technique is more sensitive 56.88% as compared simple salt flotation technique 42% and less sensitive was direct wet mount technique 38%.

The result of our study disagreed with the study of (Hearsh et al., 2015 and Mahmood et al., 2014) in their study flotation concentration technique (49.3%, 60%) is more sensitive than sedimentation technique (43.3%, 58.7%) and direct wet mount (22.6%, 39.3%). All these methods are rapid, cost effective and easy to perform in rural settings with minimum basic infrastructure requirements. This study shown that there was a significant increase in the number of parasites detection by application of the concentration methods. If we include two or more methods along with different concentration techniques with different principles into the routine diagnostic tests to find out the correct prevalence of parasites in community. The direct wet mount showed the lowest sensitivity from the concentration methods, but the advantage of this method is to provide a quick diagnosis of a heavily infected specimen, to check organism motility and to diagnose parasites that may be lost in concentration techniques (Robyn, 1997).

5. Conclusion

This can be concluded from the study that the prevalence of parasitic infection was higher in males compared to females because of outdoor eating & drinking and unhygienic condition. The formal ether technique is more sensitive rather than other techniques.

6. Source of Funding

Nil.

7. Conflicts of Interest

None.

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