Orginal Article

A Study on Different Laboratory Methods for Diagnosis of Intestinal Protozoal Infections

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Abstract

This cross sectional study was done from January 2009 to June 2010 in Microbiology department of Dhaka Medical College, Dhaka on different laboratory methods for diagnosis of intestinal protozoal infections and their distribution among selected study population. Of the 375 stool samples evaluated, 103 (27.5%) samples were positive for the intestinal protozoa. The prevalence of G. intestinalis was highest (25.78%) followed by E. histolytica/dispar (8.98%). Other protozoa found were, E. coli (1.56%), B. hominis (1.95%), C. mesnilli (1.17%) and C. parvum (0.78%). Iron-hematoxyline stain showed highest sensitivity for detection of protozoa.

Key Words: Intestinal protozoa, Iron-haematoxyline stain (Permanent stain).

Introduction:

Intestinal parasitic infections are globally endemic and have been described as constituting the greatest single worldwide cause of illness and disease.[1] The World Health Organization (WHO) estimates that 3.5 billion people worldwide are infested with some type of intestinal parasite, and as many as 450 million of them are sick as a result.[2] Giardia intestinalis, causing giardiasis, is the most common protozoan parasite worldwide.[3] Direct stool smear technique is quick to prepare and inexpensive when compared with other methods but it can miss protozoa (cysts, trophozoites, oocysts) if concentration is too low or if too much debris or fat is present.[4] There is need for increase probability of finding the parasite in the fecal samples to allow for accurate diagnosis, hence there is need to practice other methods.[5] It has been proved that Iron-haematoxylin staining offers many advantages over direct stool smear technique for detecting intestinal protozoa. If performed correctly, this method is sensitive, simple, economical and easy to carry out.[5,6]

Material and Methods:

This cross sectional study included 375 person of all age group of the patients attending at outpatient department of Dhaka Medical College, people of two villages Konakhola and Malancha in Keraniganj Upazila, Dhaka, children from an orphanage in Dhaka city and among people of two urban slum Korail and Kamrangirchar in Dhaka city. The sample was selected by simple random sampling. After labeling, a plastic container was supplied to each person to collect the stool in the next morning. The container of stool samples was collected during visit in the next morning and was transported to the microbiology laboratory as early as possible. The collected stool was immediately examined macroscopically and microscopically followed by Iron-haematoxyline staining, formol petrol concentration technique and iodine wet mount. The results of the study were recorded systematically. Data analysis was done by Microsoft Excel version 2007.

Results:

A total of 375 stool samples from healthy people of different age and sex were included in this study. Among these, 103 (27.5%) samples were positive for the intestinal protozoa. The prevalence of G. intestinalis was highest (25.78%) followed by E. histolytica/dispar (8.98%). Other protozoa found were, E. coli (1.56%), B. hominis (1.95%), C. mesnilli

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Table I: Protozoal species identified among the stool positive cases (n = 103).

	Total
Entamoeba histolytica/dispar	23 (08.98)
Giardia intestinalis	66 (25.78)
Entamoeba coli	04 (01.56)
Blastocystis hominis	05 (01.95)
Chilomastix mesnili	03 (01.17)
Cryptosporodium parvum	02 (00.78)

Iron-haematoxyline staining, formol petrol concentration technique and iodine wet mount were superior to saline preparation in detection of intestinal protozoa.

Table II: Detection of protozoa in different procedures.

Protozoa	Routine microscopy		Formol petrol	Iron Haematoxyline
	Saline	Iodine	method	Staining
G. intestinalis	34	60	63	64
E. histolytica/dispar	15	21	21	22
E. coli	02	03	03	03
B. hominis	01	04	04	03
C. mesnili	02	01	02	00
C. parvum	00	00	00	02
Total	54	89	93	94

Discussion:

Giardia intestinalis had the highest prevalence of 25.78%. This could be due to the poor hygienic conditions and inadequate sanitation in the selected area promoting fecal-oral transmission of cysts. This prevalence found in this study was higher than Azam et al. where the prevalence was 16.4%. This might be due to the fact that Azam and other researcher carried out the study among 6-10 years age group whereas all age group were included in this study. In addition, Azam et al. followed a single procedure (wet film microscopy) in the study whereas three procedures (wet film microscopy, formol petrol concentration and staining) followed in this study.[7]

In this study, 23 (08.98%) were infected by Entamoeba histolytica/dispar. Similar observation was found by Hauque et al. in Dhaka where the prevalence was 5% and Azam et al. in Gazipur where the prevalence was 3.6%. The prevalence seems to be high in this study. This might be due to unhygienic condition of the study area and samples were not collected from any selected age group.[7,8]

In this study, 54 protozoa were detected by direct microscopy of saline wet mount, 89 by iodine wet mount, 93 by formol petrol concentration method and 94 by Iron-haematoxyline staining. Iron-haematoxyline staining showed the highest sensitivity (96.77%) to detect protozoa in stool. These findings correlate with Gardner et al. (1980) where he found that permanent stained smear (58.5%) was much more effective method for detecting protozoa in stool than the direct wet mount (4.8%).[9]

Conclusion:

It was observed that different procedures used in this study detected more intestinal parasitic infection than direct smear method which is in practice in most of the laboratories of Bangladesh. Protozoa was best identified by Iron-haematoxyline staining followed by formol petrol concentration method. So concentration technique should be practiced for diagnosis of intestinal protozoal infection.

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