Original article:

Comparison of stool concentration methods for detection of prevalence of enteroparasitic infection in rural tertiary care teaching hospital of Maharashtra

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Abstract:

Background: Intestinal parasitic infection is one of the major health problems in many developing countries. The prevalence of intestinal parasitic infections not only varies in different parts of the world but also in different regions of same country.

Methods: A total of 856 patients attending out-patients department and admitted in wards, who presented with complaints of diarrhoea, vomiting, abdominal pain and weight loss were included in the study. Stool samples collected from these patients were screened for the presence of intestinal parasites as per standard parasitological protocols. Comparison of formol ether sedimentation and zinc sulphate centrifugal floatation technique for detection of enteroparasites was done.

Results: Intestinal parasites were detected in 145 stool samples by direct saline and iodine mount. Ascaris lumbricoides followed by Ancylostoma duodenale and Taenia species were the common intestinal parasites. Entamoeba histolytica was the most common intestinal protozoa. Coccidian parasites like Cryptosporidium parvum and Isospora belli were detected in the HIV infected patients only. The total prevalence of intestinal parasites by formol ether sedimentation technique is 26.75% while by zinc sulphate centrifugal floatation technique prevalence is 17.64%.

Conclusion: Intestinal parasitic infections are worldwide in distribution. Their prevalence in rural areas is high due to poor sanitation and lack of personal hygiene. Early and prompt diagnosis of intestinal parasitic infections is important as in addition to morbidity and mortality, they contribute to malnutrition, growth retardation and diminished work capacity.

Keywords: Ascaris lumbricoides, coccidian parasites, Entamoeba histolytica, stool concentration techniques.

Introduction:

Intestinal parasitic infection is one of the major health problems in many developing countries.1) Gastrointestinal tract is the primary involvement site of parasites during their lifecycle.2) Amoebiasis, ascariasis, hookworm infection and trichuriasis are among the top ten most common infections in the world.3) Ascariasis can result in often-fatal intestinal obstruction; hookworm infection can cause iron deficiency anaemia; trichuriasis is associated with chronic dysentery and rectal prolapse. Amoebiasis can result in dysentery and extraintestinal complications. Giardiasis is associated with acute diarrhoea, steatorrhea and lactose intolerance.3) The prevalence of intestinal parasitic infections not only varies in different parts of the world but also in different regions of same country.4) Geographical conditions, poor nutritional and low socioeconomic status contribute to making rural area a favourable condition for parasitic infections. Recognizing features of parasitic infection is very essential for prompt diagnosis and treatment. It also helps to avoid complications and unnecessary surgical intervention. The present study was conducted with an aim to determine the prevalence of intestinal parasitic infection in a rural tertiary care teaching hospital and to compare the stool concentration techniques for detection and identification of enteroparasite.
Materials and methods:
The present study was conducted in Department of Microbiology, Swami Ramanand Teerth Medical College and Rural Hospital, Ambajogai, Maharashtra. A total of 856 patients attending out-patients department and admitted in wards, who presented with diarrhoea, vomiting, abdominal pain and weight loss were included in the study. A total of 3 freshly voided stool samples were obtained from these subjects in sterile, screw capped, disposable plastic containers. The samples were immediately transported to Department of Microbiology for further parasitological study. All stool samples were macroscopically observed for consistency, colour, presence of mucus and blood, adult intestinal helminths and segment of tapeworm. Microscopic examination of stool samples was done using direct preparation (saline and iodine wet mount) and after formol ether concentration technique. The wet mounts were observed under low power (10X) and high power (40X) for detection of trophozoites and cysts of intestinal protozoa and ova of helminths. Modified Ziehl Neelsen staining was done on smears prepared from fresh stool samples after methanol fixation. The smears were screened under low power (10X), high power (40X) and oil immersion (100X) objectives of light microscope for detecting oocysts of coccidian parasites like Cryptosporidium, Cyclospora and Isospora. For concentration techniques, the stool samples were added to 10% buffered formalin. The specimen in formalin were subdivided into two parts. One part was processed by zinc sulphate centrifugal floatation technique and the other part was processed by formol ether sedimentation.

Results:
In the present study, out of 856 patients, 502 (58.64%) were males and 354 (41.36%) were females. Majority of patients were from age group of 16-25 years (n=206 (24.07%)) followed by 26-35 years age group (n=197(23.01%)). Intestinal parasites were detected in 145 stool samples by direct saline and iodine wet mount preparation. Species wise distribution of intestinal parasites is shown in Figure 1. Ascaris lumbricoides followed by Ancylostoma duodenale and Taenia species were the common intestinal parasites. Entamoeba histolytica was the most common intestinal protozoa. Coccidian parasites like Cryptosporidium parvum and Isospora belli were detected only in HIV infected patients. Comparison of prevalence of intestinal parasites by formol ether sedimentation and zinc sulphate centrifugal floatation technique is shown in Table 1. In our study, the total prevalence of intestinal parasites by formol ether sedimentation technique is 26.75% while by zinc sulphate centrifugal floatation technique prevalence is 17.64%.

Discussion:
Intestinal parasitic infections rank among the most significant causes of morbidity and mortality in the world. In our study A. lumbricoides followed by A. duodenale and Taenia species were the most common intestinal parasites. Several studies have demonstrated a high prevalence of intestinal helminthic infections in underprivileged community. Intestinal helminthic infections are common in poor and socioeconomically deprived communities in the tropics and subtropics; where poverty, overcrowding, poor environmental sanitation and low level of education are more apparent. It is estimated that 25% of the world population are infected by A. lumbricoides and this causes up to a million cases of disease annually. E. histolytica and G. lamblia were the intestinal protozoa detected in the stool of the patients. E. histolytica is an inhabitant of the intestinal lumen. Amoebiasis affects 10% of the world population, resulting in 100,000 deaths per year. It is the 3rd
most common cause of parasitic diseases. Most of the infected patients are asymptomatic, but about 10% may present with clinical symptoms such as frank dysentery and a liver, lung, or brain abscess.\(^{(2)}\) The trophozoites containing ingested erythrocytes were identified as *E. histolytica*, but the cysts of *E. histolytica* and *E. dispar* are morphologically similar and therefore can’t be differentiated microscopically.\(^{(7)}\) The cysts were presumed to be that of *E. histolytica*, as *E. dispar* is non pathogenic and non invasive. *Cryptosporidium parvum* and *Isospora belli* were coccidian parasites found to be associated with diarrhoea in HIV infected patients. Predominance of coccidian parasites in HIV infected patients was also noted in the study of Deorukhkar et al.\(^{(7)}\) and Basak et al.\(^{(8)}\) Coccidian parasites usually cause a self-limiting illness in immunocompetent individuals but as the immune status of the patients falls, they are known to cause life-threatening profuse watery diarrhoea.\(^{(9,10)}\)

In our study, we compared formol ether sedimentation and zinc sulphate centrifugal floatation technique for detection of intestinal parasites. Stool concentration methods are usually performed when direct smear fails to reveal any parasites. The concentration of stool allows detection of small number of parasites in the stool specimen. These procedures allow the increased recovery of eggs and protozoal cysts. The trophozoites are destroyed during concentration procedure. In this investigation, additional 6 intestinal parasites were detected by zinc sulphate centrifugal floatation technique. Zinc sulphate floatation and saturated salt floatation are most frequently used. Many helminthic eggs and protozoal cysts can be recovered and demonstrated by floatation methods but the high specific gravity of the fluid may cause distortion in the morphology of eggs and cysts of enteroparasites. The basic disadvantage of the floatation technique is that unfertilized eggs of *Ascaris lumbricoides*, eggs of *Taenia saginata* and *Taenia solium*, all trematode eggs and larvae of *Strongyloides stercoralis* do not float because these are heavier than the suspending liquid.\(^{(5)}\)

In the present study additional 84 intestinal parasites were detected by formol ether sedimentation technique. The total prevalence of intestinal parasites by formol ether sedimentation technique is 26.75% while by zinc sulphate centrifugal floatation technique prevalence is 17.64%. Sedimentation techniques are easy to perform and technical error is also less. They are more sensitive than floatation methods and morphology of helminthic eggs and protozoal cysts is preserved.\(^{(5)}\)

**Conclusion:**

Intestinal parasitic infections are worldwide in distribution. Their prevalence in rural areas is high due to poor sanitation and lack of personal hygiene. Early and prompt diagnosis of intestinal parasitic infections is important as in addition to morbidity and mortality, they contribute to malnutrition, growth retardation and diminished work capacity. Formol ether sedimentation technique is better technique for detection of cysts, eggs and larvae.
Table 1. Comparison of prevalence of intestinal parasites by formol ether sedimentation and zinc sulphate centrifugal floatation technique.

<table>
<thead>
<tr>
<th>Sr. no</th>
<th>Intestinal parasite</th>
<th>Formol ether sedimentation (n)</th>
<th>(%)</th>
<th>Zinc sulphate centrifugal floatation technique (n)</th>
<th>(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ascaris lumbricoides</td>
<td>56</td>
<td>6.54</td>
<td>48</td>
<td>5.61</td>
</tr>
<tr>
<td>2</td>
<td>Ancylostoma duodenale</td>
<td>38</td>
<td>4.44</td>
<td>37</td>
<td>4.32</td>
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<tr>
<td>3</td>
<td>Taenia species</td>
<td>33</td>
<td>3.86</td>
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<td>0</td>
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<tr>
<td>4</td>
<td>Hymenolepis nana</td>
<td>26</td>
<td>3.04</td>
<td>27</td>
<td>3.15</td>
</tr>
<tr>
<td>5</td>
<td>Trichuris trichiura</td>
<td>8</td>
<td>0.70</td>
<td>7</td>
<td>0.82</td>
</tr>
<tr>
<td>6</td>
<td>Strongyloides stercoralis</td>
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<td>0.47</td>
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<td>0</td>
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<tr>
<td>7</td>
<td>Enterobius vermicularis</td>
<td>3</td>
<td>0.35</td>
<td>3</td>
<td>0.35</td>
</tr>
<tr>
<td>8</td>
<td>Entamoeba histolytica</td>
<td>17</td>
<td>1.99</td>
<td>15</td>
<td>1.75</td>
</tr>
<tr>
<td>9</td>
<td>Giardia lamblia</td>
<td>8</td>
<td>0.93</td>
<td>7</td>
<td>0.82</td>
</tr>
<tr>
<td>10</td>
<td>Entamoeba coli</td>
<td>5</td>
<td>0.58</td>
<td>7</td>
<td>0.82</td>
</tr>
<tr>
<td>11</td>
<td>Cryptosporidium parvum</td>
<td>24</td>
<td>2.80</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>12</td>
<td>Isospora belli</td>
<td>5</td>
<td>0.58</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>229</td>
<td>26.75</td>
<td>151</td>
<td>17.64</td>
</tr>
</tbody>
</table>

References:


