**Prevalence of Intestinal Protozoa in Primary Schools in Zawia City, Libya**

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**Abstract:** A total number of 605 stool samples were collected among eight primary school in Zawia city, during the period of November 2011 to April 2012 and analyzed for intestinal protozoa. Stool examination was done by direct and formalin ether technique. Total 64 (10.6%) cases were found with intestinal protozoa in their stools. The most common protozoa was *Entamoeba coli( E. coli*) 22 (3.6%), *Entamoeba histolytica (E. histolytic)* found in 19 (3.1%) cases and *Gardia lamblia (G. lamblia)* in 11 (1.8%) cases. Double infection was seen in only three samples. The low prevalence of intestinal protozoa among the study groups reflects the outstanding health and hygienic care in primary schools visited.

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**1- Introduction**

Intestinal parasitic infection is endemic throughout the world. They affect an estimated 3.5 billion persons and cause clinical morbidity in approximately 450 million (**WHO, 2000**). Human intestinal parasites are found worldwide both in developing and industrialized countries. Pathogenic intestinal parasites produce disease by infecting the small and \or large intestine. Some intestinal parasites have their major impact on children, while others infect all age groups but have their most profound effect on adults (**Zakai, 2004**). Parasites are one of the important casual agents of gastrointestinal disorders such as diarrhea, loss of weight, abdominal pain, nausea, vomiting, lack of appetite, abdominal distention and also sometime mentally related disorders. Several factors such as poor hygienic environment, poor socioeconomic status, climate and overcrowding of people are the major causes of increasing incidence of parasitic infections (**Niyyati** ***et al****,* **2009**). The distribution of intestinal parasitic infection depends on many factors. These include temperature, humidity, intermediate host, socioeconomic status of society, customary nutrition of people, and immigration (**Yilmaz *et al****,* **1999**). **Mahfouz *et al*** (**1997**) studied the ecological determinants of intestinal parasitic infections among preschool children in an urban squatter settlement of Egypt in 658 preschool children below 5 years of age. Overall, the prevalence rates of infectionswith the intestinal Helminthes and Protozoa were 47.3% and 31.5%, respectively. **Yilmaz *et al*** (**1999**) studied the distribution of intestinal parasites in two societies with different socio-economic status in Van, Turkey the results revealed that one or more intestinal parasites in 149 (55.6%) out of 268 immigrants, and 55 (27.3%) out of 205 university residents were detected. The finding of their study suggests that socio-economic status is an important factor on the distribution of intestinal parasites. Another study done by **(Menezes *et al*, 2008**) showed that out of 472 Brazilian children aged 3- 6 years, 24.6% had some types of intestinal parasites and 6.6% was have more one type of parasites. Among protozoa, *E. coli* (14.0%) and *G. duodenalis* (9.5%) were the most prevalent. A total of 205 cases from patients referred to a Hospital in Tehran, tested for the presence of intestinal parasites using direct examination, formalin- ether concentration and staining with Ziehl-Neelsen. The results showed that the positive rate was (29.75%). The rates was as follows *: B. hominis* 20.9%, *G. lamblia* 5.36%, *I. butschelii* 0.48%, *Dientamoeba fragilis* 0.48%, *Trichomonas hominis* 0.48%, *E. nana* 0.97%, *E. vermicularis* 0.48% and *Taenia* 0.48%. (**Niyyati e*t al,*****2009**). In Libya there are some studies which studied intestinal parasites prevalence among human. The first report on the prevalence of intestinal parasites in Libya was conducted by **Dar *et al*** (**1979**) on prevalence of parasitic infection in primary school children in Benghazi. The study showed that the prevalence of intestinal parasites was 27.6% among school children in Benghazi, Libya. The detected parasites were *G.* *lamblia* (11.4%)*, Hymenolepis. nana (*0.3%), *A. lumbricoides* (2,5%), *T. trichiura* (1.3%), *E. vermicularis*(0.8%), *S. stercoralis* (0.2%), *Hookworm* (0.2%), *I. butschelii* (0.3%). Another study by **EL-Boulaqi *et al*** (**1980**) for the prevalence of intestinal parasites in primary school children in Benghazi city, Libya, the results showed a very high affection with the intestinal protozoa 75.6% and intestinal helminthes 68.8%, and the most common of parasite are *A. lumbricoides*, *E. coli*, *E. histolytica* respectively. **EL-Buni and Khan 1998** conducted a study on intestinal protozoan infections in Benghazi. The overall prevalence rate was (12.88%). The most common parasite was (6.24%) *G. lamblia*, (3.94%) *E. histolytica*, (2.62%) *E. coli* and (0.07%) *T. hominis.* **Sadaga** (**2006**) studied the prevalence of intestinal parasites among primary school children in Derna district, Libya. The result showed that (31%) of the children were infected with intestinal parasites. The detected parasites were *G. lamblia* (12.7%), *Blastocysti. hominis*(6.7%), *E. histolytic* (6.6%), *E. coli* (3.2%), *E. vermicularis* (0.6%), *A. lumbricoides* (0.1%), *Entamoeba Hartman* (1.0%) and *Hymenolepis. nana* (0.1%). **Alkilani *et al*** (**2008**) studied the incidence of intestinal parasitosis in Nalout popularity, western Libya. The results showed an overall rate of 29.6% of these patients, 7.59% had double or triple parasitic infections. Children were more infected than adults. The infection rates were were *E. histolytica* (21%), *E. vermicularis* (7.5%), *G. lamblia* (2.5%), *E. coli* (0.38%), and *H. nana* (0.38%). A sudanese immigrant employee had *S. stercoralis* larva in their stool culture. The objective of this study is to determine the prevalence of intestinal protozoa infection among primary school children in Zawia city- Libya by using Direct Smear and Formalin Ether Concentration Technique and comparing the prevalence rate between the urban children and rural children.

**2-Materials And Methods**

Study group: This study was conducted in Eight primary school were selected by systematic random sampling using the master list of the schools In Zawia city- Libya, between the first week of November 2011 and the last week of April 2012.

A total of 605 random fresh stool samples were collected from primary school children aged between 6 and 14 years. Permissions to visit schools and obtain samples from students were obtained from the ministry of education.

Collection stool samples: Each student was given small package containing a covering latter, screw capped universal plastic container, questionnaires and was asked to provide, on the next day, a single fecal sample. At the second visit, the containers were collected. One or two additional visits were paid to get the undelivered the container and or the questionnaire. Then transported to the laboratory. A code was given to each questionnaire. This cod was also written on the samples.

Laboratory investigation: The Laboratory investigation included direct wet smears in normal saline, Lugol’s iodine and formalin-ether concentration methods.

Statistical analysis: The Statistical Package for Social Science (SPSS) version which is software for biostatic analysis was used to achieve valid and reliable results in this study.

**3- Results**

A total of 605 stool samples were examined. The prevalence of intestinal protozoa infection differed with the diagnostic procedure used. The results of the stool sample analysis revealed that, 45 (7.4%) of primary school children samples were positive by Direct method, while by sedimentation method the positive rate were 64 (10.6%). These results were statistically significant (p <0.05). Table (1).

Table 1. Comparison between two methods in the diagnosis of intestinal protozoa

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| The result | | Sedimentation  method | | Total |
| Negative | Positive |
| Direct method | Negative | 541(89.4%) | 19 (3.1%) | 560 (92.6% |
| Positive | 0 (0.0%) | 45 (7.4%) | 45 (7.4%) |
| Total | | 541(89.4%) | 64 (10.6%) | 605 (100%) |

The positive rate of intestinal protozoan infections was 63(10.4%). The most common parasites detected in subject sample were *E. coli* (3.6%), followed by *E. histolytica* (3.1%), *G. lamblia* (1.8%), *Endolimax nana* (1.1%), *B. hominis* (0.1%), *E. vermicularis* (0.1%) and mixed infections *E. coli& G. lamblia* (0.1%), *E. nana& B. hominis* (0.1%), *E. nana& Iodamoeba butschelii* (0.1%). Of the 64 positive samples, 61(10.0%) samples yielded single parasite, where as 3 (0.4%) samples had mixed parasites (Table 2).

**The Prevalence of intestinal protozoa according to sex: Table (3):** In males, out of 240 stool samples which were collected from the eight primary school of the study a total of 27(4.5%%) Were found to be positive for intestinal protozoa infection. In females, out of 365 stool samples which were collected from the eight primary school of the study a total of 37 (6.1%%)Were found to be positive for intestinal parasites infection. These results showed no statistical significance difference between intestinal protozoa infection and sex (p>0.05).

Table 2. Protozoa detected in primary schoolchildren

|  |  |
| --- | --- |
| Parasites | No. infected |
| Single infection | 61(10.0%) |
| *Entamoeb coli* | 22 (03.6%) |
| *Entamoeb histolytica* | 19 (03.1%) |
| *G. lamblia* | 11(01.8%) |
| *Endolimax nana* | 07(01.2%) |
| *Blastocystis hominisx* | 01 (00.2%) |
| Mixed infections | 03 (0.04%) |
| *E. coli& G. lamblia* | 01 (00.2%) |
| *E. nana& B. hominis* | 01 (00.2%) |
| *E. nana& Iodamoeba butschelii* | 01(00.2%) |
| Total | 64 (10.6%) |

Table 3: Association between prevalence of intestinal protozoa and sex.

|  |  |  |  |
| --- | --- | --- | --- |
| The result | Sex | | Total |
| Male | Female |
| Negative | 213 (88.75%) | 328 (89.86%) | 541 |
| Positive | 27 (11.25%) | 37 (10.14%) | 64 |
| Total | 240 (39.7%) | 365 (60.3%) | 605 |

Table 4: Prevalence of Intestinal protozoa according to sources of drinking water.

|  |  |  |  |
| --- | --- | --- | --- |
| Source of Water | The Result | | Total |
| Negative | Positive |
| Artesian wells | 332 (90.71%) | 34 (09.29%) | 366 (100%) |
| Artesian wells &  filtered water | 02 (50%) | 02 (50%) | 04 (100%) |
| Artesian wells &  water of pipes | 10 (76.92%) | 03 (23.08%) | 13(100%) |
| Filtered water | 03 (100%) | 0 (0.0%) | 03 ( 100%) |
| Water of pipes | 189 (88.32%) | 25 (21.68%) | 214(100%) |
| Water of pipes & filtered water | 05 (100%) | 00 (0.0%) | 05 (100%) |
| Total | 541 (89.4%) | 64 (10.6%) | 605(100.0%) |

**The Prevalence of intestinal protozoa according to sources of drinking water: Table (4)** The highest infection was observed in students which drinking water from wells (5.6%). In contrast, students which drank from filtered water are not infected. The results showed that there was a significant relation between the Prevalence of intestinal protozoa and the sources of drinking water (p <0.05).

**The Prevalence of intestinal protozoa according to sewage system: Table (5):** In a Private sewage, Out of 605 stool samples which were collected from the eight primary schools from of the study, the positive rate was 37(9,59%). In a public sewage, Out of 605 stool samples which were collected from the eight primary schools from of the study, the positive rate was 27(12,33%).

These results showed no statistical significance difference between intestinal protozoa infection and sewage system (p>0.05)

Table 5: Shows the relationship between Prevalence of intestinal protozoa and sewage system.

|  |  |  |  |
| --- | --- | --- | --- |
| The result | Sewage system | | Total |
| Private sewage | Public sewage |  |
| Negative | 349(90,41%) | 192(87,76%) | 541(89.4%) |
| Positive | 37(9,59%) | 27(12,33%) | 64(10.6%) |
| Total | 386(100%) | 219(100%) | 605(100.0%) |

The Prevalence of intestinal protozoa according to location of school (Urban /Rural): In the urban area, a total of 256 stool samples which were collected from the three primary schools from of the study, the positive rate were 31(12,11%). In the rural area, a total of 349 stool samples which were collected from the five primary schools from of the study, the positive rate were 33(9,46%) These results (Table 6) showed statistical significance between intestinal parasites infection and location of school [Urban /Rural] (p <0.05).

Table 6: Prevalence of Intestinal protozoa according to location of school (urban /rural).

|  |  |  |  |
| --- | --- | --- | --- |
| The Result | Location of school | | Total |
| Urban | Rural |
| Negative | 225(87.89%) | 316(90.54%) | 541(89.4%) |
| positive | 31(12,11%) | 33(9,46%) | 64(10.6%) |
| Total | 256(100%) | 349(100%) | 605(100.0%) |

**4- Discussion**

This study provides the first estimate of parasitic prevalence among primary schoolchildren in Zawia District. The formalin ether concentration sedimentation technique was used in the present study because of its higher diagnostic sensitivity of intestinal parasites than the direct smear technique. The prevalence of intestinal parasites infections estimated in this study was 10.6%. This reflects the hygienic standard in Zawia District in general. Furthermore, the low prevalence of intestinal parasites reflects the outstanding health and hygienic care in primary schools visited.

This finding is consistent with a study done by **EL–Buni and Khan** (**1998**) where a prevalence of 12.88% intestinal protozoan found in Benghazi. Another study conducted by **Ben Musa** (**2007**) among school-children reported a prevalence of 14.6%. This result was lower than another school-based study conducted in Derna by **Sadaga** (**2007**) who reported a prevalence of 31%. This is in agreement with several studies carried out in different parts of Libya, high prevalence of protozoan infection may due to their simple life cycle and way of transmission.

In this study, the detected protozoa were *G. lamblia* (1.98%), *E. histolytica* (3.1%), *E. coli* (3.8%), *H. nana* (1.4%), *B. hominis* (0.3%), *I. butschelii* (0.1%) and *E. vermicularis* (0.1%). *E. coli* detected as the most common protozoa with a rate of 3.8%, different rates were reported in Benghazi, 11,4% (Dar et al 1979), 19.7% (**EL-Boulaqi et al 1980**), and 6.24% (**EL –Buni and Khan, 1998**). The second most frequently identified intestinal Protozoa a among our study population were *E. histolytica* with a rate of (3.1). Different data for this parasite were reported in this country, o.3% in Benghazi (**Dar et al, 1979**), 15,7% in Benghazi (**EL-Boulaqi et al, 1980**), 6.6% in Derna district (**Sadaga, 2007**) and 32.8% in Zawia district as well as 20.45% in Sohag Governorare, Egypt (**EL-Masry *et al,* 2007**), 11.7% in Sana, a, Yemen (**Azazy and Raja 2003**), 18.5% in Ecuador (**Mamie-Eleanor et al., 2003**), 11% in Delhi, India (**kaur *et al*, 2002**).

Our study showed a high rate (10.1%) of single infection and low rate (0.5%) of mixed infection. This agrees with results by (**Dar et al, 1979 and Sadaga, 2007**).

The present study found that there is no statistical significance (p>0.05) between age groups and rate of infection, This finding agrees with results by **Dar et al, 1979 and Sadaga, 2007** as well as **Chandrashekhar et al, 2005**. On the contrary, **Gelaw et al. 2013** where prevalence of intestinal parasites was high in age group of 10–12 years compared to other age groups. This might be due to children in this age group usually move around over a wider territory, increasing the possibility of acquiring infections from contaminated environment.

In this study the positive rate of intestinal protozoa in male was higher than in female. The percentage was (10.14%) & (11.25%) in female and male respectively, but it is not statistically significant (p>0.05). On the contrary, Okyaya et al.,2004 found the positive rate was (50.1%) in females and (49.1%) in males.

We found that the highest rate of infection in samples who usually drinking wells water, compared to those who used Tap water or Filtered water. The infection can occur by drinking water that is contaminated with sewage due to close running of artesian water and sewage lines. These findings appear to be in accordance with the findings of **Mukhopadhyay *et al*, 2007**, which reported that the main source of the infection relates to poor quality drinking water and poor personal hygiene. But not consistent with results by **Sadaga, 2007**, who found the high rate in cases water of pipes.

Our results also showed that there is no statistical significance (p>0.05) between intestinal parasites infections and sewage system.

On the other hand, the present study found that the higher prevalence of protozoa infections in rural school than urban school could be due to low socio-economic status, poor hygienic habits and lack of sanitation prevailing in the rural school.

**5- Conclusion**

This study showed that the prevalence of intestinal parasites among primary school children in Zawia city- Libya is lower than others cities. The prevalence of protozoa infection was higher than that of helminthes. In the present study, the children in rural schools had a significantly higher prevalence of intestinal parasites than urban schools. The study data suggest that drinking wells water might be the most important routes of intestinal parasites transmission in our children. This study has shown that Formalin-ether concentration method has a higher detection rate compared with direct smear method. Although Direct smear is simpler to perform, Formalin-ether concentration method is far more accurate in providing correct laboratory diagnosis but with additional laboratory requirements such as additional reagents necessitating additional time and effort. Thus, Formalin-ether concentration method should be the technique used in screening stool samples of children.

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