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ENTEROPARASITIC HELMINTHES INFECTIONS AMONG SCHOOL CHILDREN AND SOME UNIVERSITY STUDENTS IN PESHAWAR

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Abstract: The present survey was carried out to determine the prevalence of intestinal parasites in school going children and some University students of Peshawar. During the study, 200 fecal specimens were examined from December 2010 to July 2011 at Different schools and some University students of Peshawar city. Stool analysis was performed by direct microscopic examination. Out of 200 subjects examined, 23 (11.5%) were found harboring Helminths parasites. *Hymenolepis nana* was more prevalent (8%) in whole survey as compared to other helminths *e.g., Ascaris lumbricoides* (1.5%), *Enterobius vermicularis* (1.5%) and *Trichuris trichiura* (0.5%). Statistically no significant difference (χ^2=6.88, df=3, α=0.05) was found in the infection rate in different age groups. Among Protozoa the commonest and non-pathogenic protozoan *Entamoeba coli* were more prevalent (66%), than *Entamoeba histolytica* (32%) and *Giardia lamblia* (17.5%). Statistically there was no association (χ^2=6.528, df=3, α=0.05) of protozoan parasitic infection with age. The frequency of helminth parasite infections is lower in Peshawar city may be due to improved socio-economic status, health education, and comparatively good sanitation system.

Keywords: Intestinal Parasites, school children, Harboring Helminths,

Introduction

In many developing countries intestinal parasites remain a major health problem. The helminthes infection significantly impairs childhood nutrition, intestinal obstructions and adversely affects cognitive functions (Nishiura *et al*., 2002). The parasites which are transmitted from soil include *Ascaris lumbricoides*, *Trichuris trichiura*, hookworms *(N. americanus*, *A. duodenale*) and *Enterobius vermicularis*. From large scale control program and economic development the infections of intestinal parasites are reduced in number (Steinmann *et al.,* 2010). Millions of people are plagued by a number of so-called neglected tropical diseases caused by soil transmitted helminthes (STH, *i.e., Ascaris lumbricoides*, Hookworms, *Trichuris trichiura* and intestinal protozoa (Gualdieri *et al.,* 2011). The infections of intestinal parasites are linked to the climatic condition and it is a major health problem (Tellez *et al*., 1997). In human the intensity of helminthes parasites are related to different ecological and behavioral aspects (Taylor *et al*., 2003). Among the helminthes parasites, *Ascaris*, hookworms, *Trichuris trichiura* and *E.histolytica* are the most common one (Alum *et al*., 2010). About 3.5 billion people are affected and majority of infections are in children (Jumbo *et al.,* 2010). About 209 million people are infected with *Enterobius*

vermicularis (pin worm) and 0.8 to 1.2 billion people are infected from *Ascaris lumbricoides* in the world (Fung *et al.,* 2009). Current estimates show that most of the infected people live in developing countries and at least more than quarter of the world's population is chronically infected with intestinal parasites which are major cause of morbidity and mortality globally (Alemu *et al.,* 2011). In addition to other pathological effects Helminthes are also responsible for stunted, underweight and anemic children at school going ages in Ethiopia (Degerege and Erko, 2013).

This project aimed to determine and compare the prevalence and intensity of STH infections in school children in Peshawar city and some Peshawar University students studying at department of Zoology.

Materials and Methods

Prevalence of intestinal parasites was observed among school going children and some Peshawar university students in Peshawar. A total of 200 stool specimens were collected from different schools and department of Zoology, University of Peshawar. Collection was made from both boys and girls (comprised 92 boys and 108 girls) of different age groups from 2 to 22years.

On first visit to each institution, the objectives of the research were discussed with the headmaster, principal and teachers. After the permission of the head of institute the small plastic container having reference number and 2-5ml of 10% formalin solution were distributed among the willing students. A questionnaire was handed over to each student after writing name, age and sex. On next day the student was asked to bring the container with stool specimen of morning defecation along with a questionnaire which was filled by their parents/ class teacher or the student him/herself. The stool specimens were brought to the Parasitology Lab at Department of Zoology, University of Peshawar to identify parasites microscopically.

Analysis of Stool

Stool analysis is common laboratory test. Each stool specimen was examined microscopically using 4x (40x), 10x (100x), and 40x (400x) microscope objectives. The following methods and technique were used.

Direct Method

In this method the stool was thoroughly mixed with 10% formalin preservative, 20µL of suspension was taken from plastic container and placed on a glass slide. Cover slip was placed on glass slide in such a way that no air bubble formation occurred. This slide was then examined under low (10x10) and high power (40x10) objectives for identification of parasitic eggs and other developmental stages by using authentic printed keys.

Sedimentation Method (Telemann's Method)

In this method 1gm of stool was suspended in 5ml of 5% acetic acid by shaking gently. The suspension was settled for 1 minute and then an equal amount of ether was added. This mixture was then centrifuged for 2 minute at 1200 RPM in Sigma's centrifuge machine. The sediment containing eggs, cysts and other stages were settled in lower layer of centrifuge tube, the acid and dirt layer in middle of centrifuge tube while ether was layered on top. The dirt, acid and ether layers

were discarded all at once so that only sediment was left. This sediment was diluted in 1 to 2mL of water. Approximately 20µL of this suspension was taken and placed on glass slide which was covered by using a glass cover slip by avoiding bubble formation. This glass slide was observed under low and high power objectives of the compound microscope respectively. The sediment contained eggs and oocysts of parasites in positive stool samples.

Willis's Brine Floatation Method

This method has been recommended for diagnosis of nematodes especially the Hook worm and whip worm's eggs. In this method about 1.0 gm of faeces was emulsified in 20mL of Brine (solution that has exactly 15.8% NaCl by weight) in a flask or glass cylinder of 2.5 cm in diameter. A clean glass slide was placed at the top of the touching container or cylinder meniscus, and this was allowed to remain for 1 hour. The slide was removed and inverted. Cover slip was then placed and examined for eggs under low and high power objectives.

Results and Discussion

During this study a total of 200 stool specimens were collected from December 2010 to July 2011. During every month collection was made from both girls and boys of different schools (Government and private) and in the month of April from female students studying in the final year of M.Sc zoology. Of 200 stool specimens, 108 were obtained from female and 92 were from boys which have been divided into four groups on the basis of age as shown in table 1. The 4 age groups were made with varying age range based on their educational level; therefore, variation in each group can clearly be seen.

Of 200 subjects examined, only 23(11.5%) were found harboring helminthes (Table 2). The highest infection rate 14.63(n=123) was observed among children with age range of 6-12 years, and lowest parasitic infection 14.28(n=35) was observed in children with age range of 13-18 years. Interestingly none of the stool specimen obtained from the children with age range of 2-5 years and university students with age range of 18-22 years was found positive for any of the helminthes. Statistically there was no significant difference (χ^2=6.88, df =3, α=0.05) in association of helminthes parasites with age. Comparing this study with another survey conducted in rural areas of Peshawar showed significantly higher helminthes infection (Ullah *et al.,* 2009), where *Ascaris lumbricoides* (45.5%) followed by *Hymenolepis nana* (8%), *Enterobius vermicularis* (4%), and *Trichuris trichiura* (3.5%) in children of age 5-14years were reported.

Table 1 Distribution of Subjects by Age

Age group	Age range (Years)	Male	Female	Total	Percentage
I (Pre-school)	2-5	5	3	8	4%
II (Primary- school)	6-12	58	65	123	58%
III (Secondary- school)	13-17	29	6	35	17%
IV(University -students)	18-22	Nil	34	34	20.5%
Total		92	108	200	100.00%

Our survey showed significantly lower enteroparasitism than that found by Brouwer *et al.* (2007) in rural areas of Vietnam, where *Trichuris trichiura* (76%) and *Ascaris lumbricoides* (71%) were most prevalent parasites. The children having these parasites were suffering from iron deficiency anemia. However much lower prevalence was reported by Saksirisampant *et al.* (2006), where *E. vermicularis* (0.19%), *T. trichiura* and hookworm (0.19% each) were detected in urban area of Thailand. Some international investigators also shown interest in investigating intestinal parasitism in children of Northern Areas in Pakistan *e.g.*, Nishiura *et al.* (2002) reported close association of Socio-cultural and behavioral factors, age relation and infection intensity for *Ascaris lumbricoides* among children in rural communities.

Of 200 Subject studied 23 individuals were found harboring helminthes parasites. Of 23 helminthes parasites positive cases 8% were infected with Cestodes *i.e.*, *Hymenolepis nana* and 3.5% were infected with Nematodes i.e. *Enterobius vermicularis, Ascaris lumbricoides* and *Trichuris trichiura*.

Table 3 shows month wise distribution of various helminthes among 200 fecal specimens collected. During the month of February *Enterobius vermicularis* was more prevalent nematode reported in 9.09% (n=11) of the cases, in December the prevalence rate was 6.67% (n=15), while in June the lowest prevalence of 3.12% (n=32) of enterobiasis was recorded. *H.nana* was the mostly reported helminth, recorded in every month except in month of April. For *H.nana* maximum prevalence was reported during February. In February the highest prevalence rate of 27.2% (n=11) of *Hymenolepis nana* was recorded and in December (2010) the infection rate was 13.3% (n=15), the lowest rate 11.76% (n=17) was recorded in the month of March. Ascariasis was recorded in March and June only. In June the rate of infection was 6.25% (n=32) and in March the rate of infection was 5.889% (n=17). *Trichuris trichiura* infection was only reported in 5.88% (n=17) of the subject in March, 2011.

Similar results were also reported previously by Steinmann *et al.* (2010) among school going children in Osh oblast, Kyrgyzstan. One of the most prevalent helminthes reported in their study was *E. vermicularis* (19.3%) followed by *Hymenolepis nana* (4.4%) in Kyrgyz school age children.

Table 2 Rate of helminthes parasites and age distribution pattern among children and university students (n= 200) [(n= %)]

Age Interval	Infected		Uninfected	
Year	**Number**	**%age**	**Number**	**%age**
2-5 (n=8)	Nil	Nil	8	100%
6-12(n=123)	18	14.63%	105	85.36%
13-18(n=35)	5	14.28%	30	85.71%
18-22(n=34)	Nil	Nil	34	100%
2-22(n=200)	23	11.5%	177	88.5%

Table 3 Month wise distribution of helminthes parasites

Months / **Parasite Identified**	**Dec (n=15)**	**Jan (n=16)**	**Feb (n=11)**	**Mar (n=17)**	**Apr (n=40)**	**May (n=20)**	**Jun (n=32)**	**July (n=49)**
E.vermicularis	1 6.67	Nil	1 9.09	Nil	Nil	Nil	1 3.12	Nil
H.nana	2 13.3	2 12.5	3 27.2	2 11.76	Nil	1 5	2 6.25	4 8.16
A.lumbricoides	Nil	Nil	Nil	1 5.88	Nil	Nil	2 6.25	Nil
T.trichiura	Nil	Nil	Nil	1 5.88	Nil	Nil	Nil	Nil

Conclusion

The percentage of parasitic infection n=200(11.5%) is less in Peshawar city. The pattern revealed that *Hymenolepis nana* was in high prevalence rate (8%) during the whole survey period, while *Ascaris lumbricoides* and *Enterobius vermicularis* both having infection rate of (1.5%). Thus it can be concluded that the intestinal helminthes are less common among children of Peshawar city and it may be due to public health education, clean water supply, sanitation facilities, hygienic environment and personal hygiene as compared to villages and rural area of Peshawar.

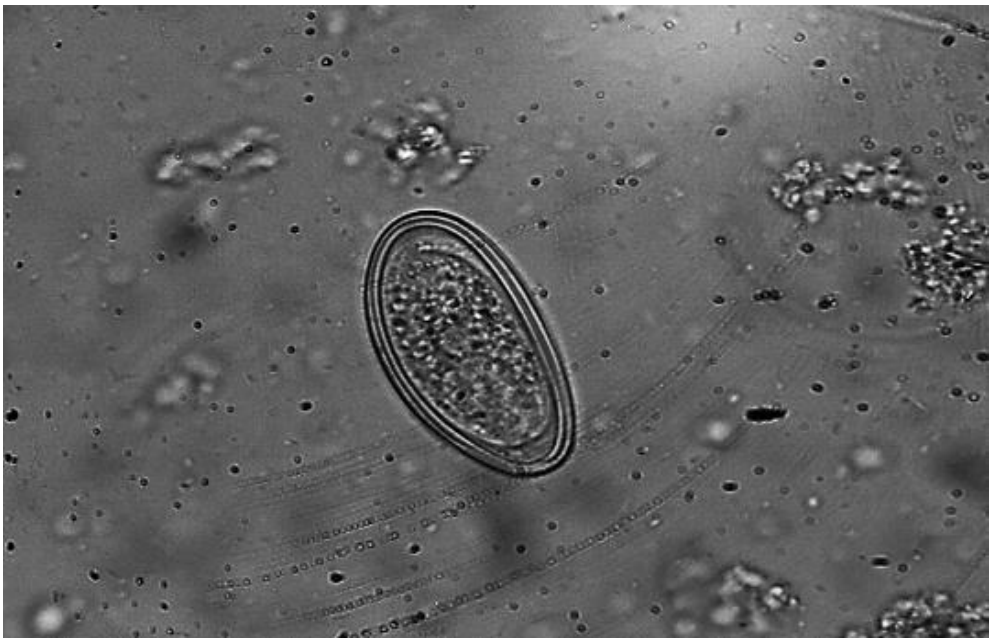

Fig.1. An ovum of *Enterobius vermicularis* (Magnification x 400) as observed in our study

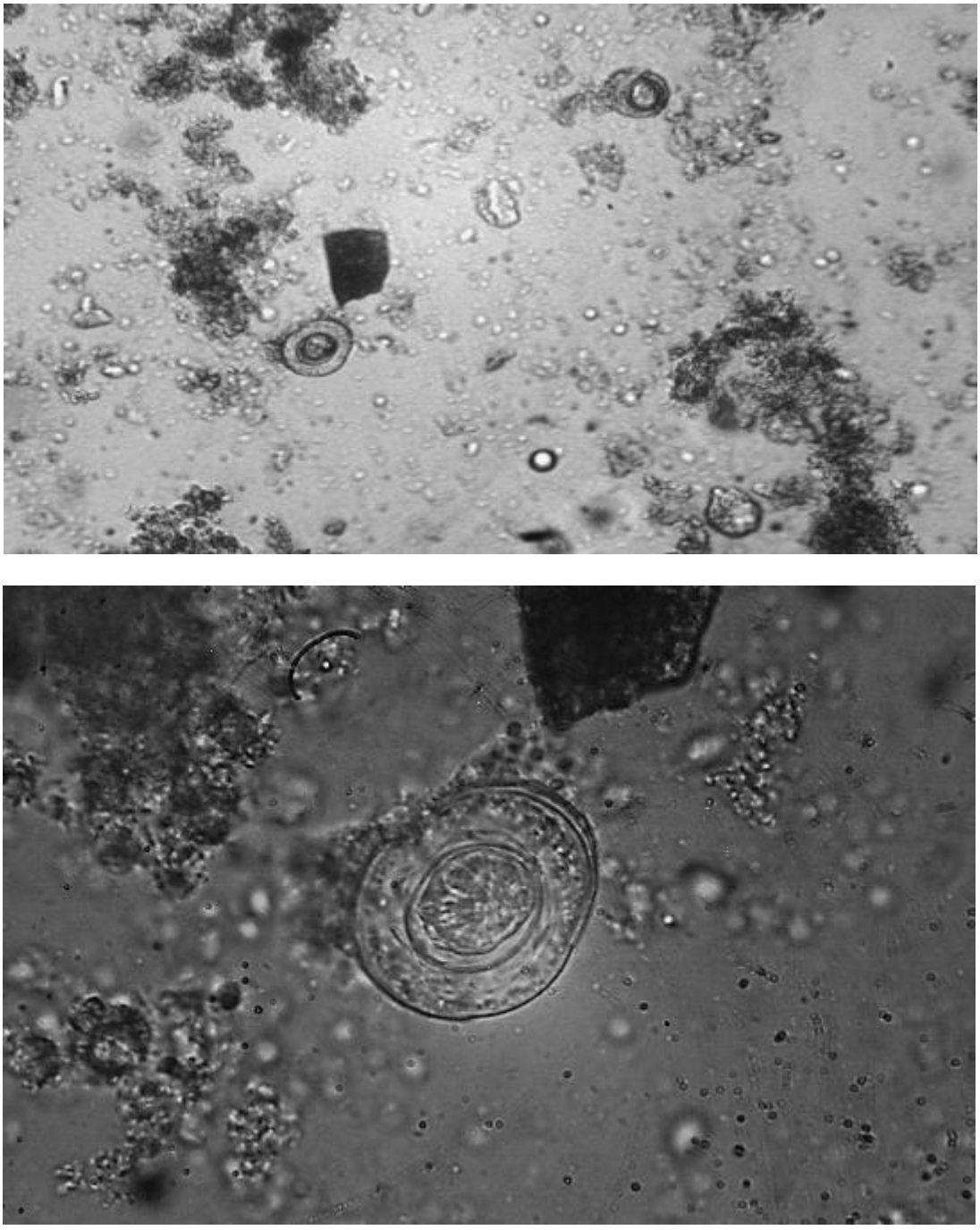

Fig.2. *Hymenolepis nana* fertilized ovum showing an Oncosphere (above Magnification x 100 and below x400) as indicated by an arrow

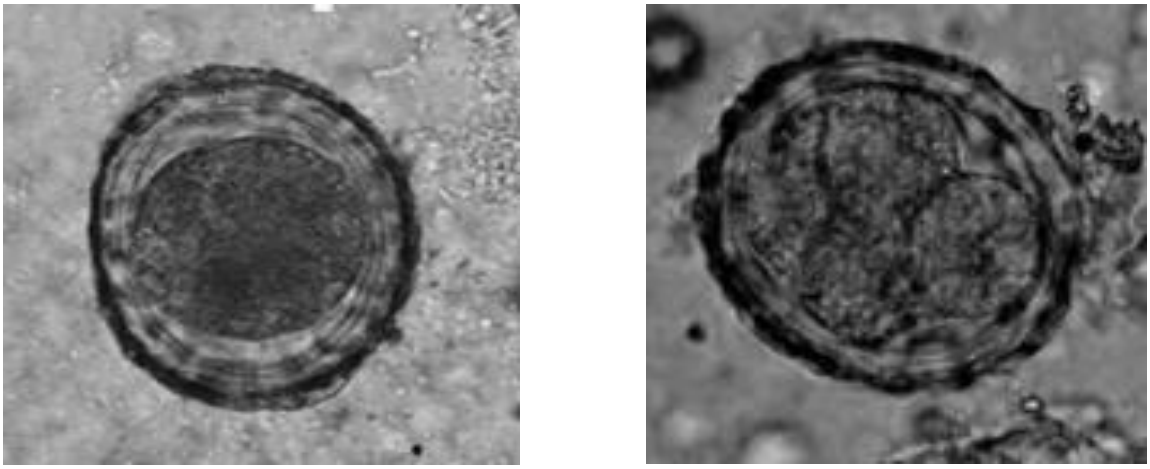

Fig.3. Ascaris lumbricoides fertilized ova with strong coarse mamilation (Magnification x 400)

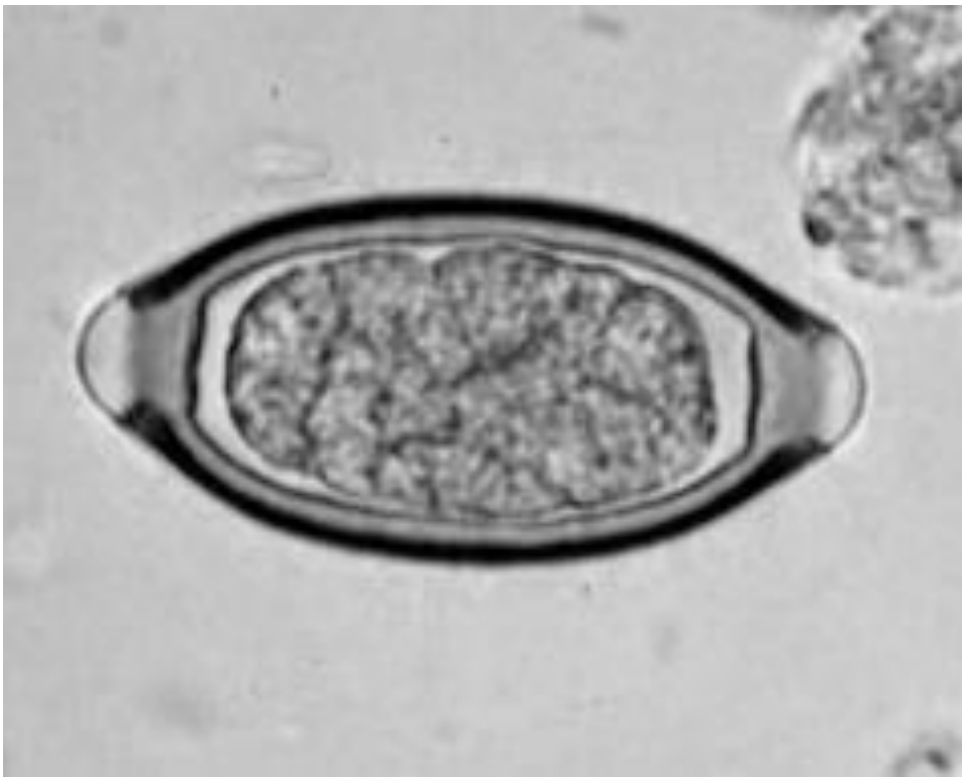

Fig.4. *Trichuris trichiura* egg showing mucoid plugs at both ends (Magnification x 400)

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