Epidemiology of Intestinal Polyparasitism among Schoolchildren In Sohag, Egypt

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Abstract

Introduction: Intestinal parasitic infections **(IPI)** are still public health problems in many communities, particularly among children in developing countries.This cross-sectional study aimed to investigate the current prevalence and risk factors associated with intestinal polyparasitism (the concurrent infection with multiple intestinal parasite species) among schoolchildren in Sohag governorate, Egypt.

Methodology / Principal findings: Fecal samples were collected **from 200** schoolchildren **(51.5% boys and 48.5% girls),** preserved and examined by using formalin-ethyl acetate concentration technique and modified Kinyoun acid fast stain. Demographic information was collected by using a standardized questionnaire. Overall, **63.5%** of the children were found to be positive for at least one parasite species. Of these, **23.5%** were polyparasitized. The overall prevalence of *Cryptosporidium*, *Giardia duodenalis*, *Entamebahistolytica/ dispar*, *Blastocystis*infections were **34%, 14.5%** and **13%,** respectively. Univariate and multivariable logistic regression models showed that none of the age, gender, residence or family size were risk factors for polyparasitism.

Conclusions/Significance: Intestinal polyparasitism is prevalent among children in Sohag, Egypt. Hence, effective and sustainable control measures, including school-based periodic chemotherapy, providing adequate health education focused on good personal hygiene practices and proper sanitation, as well as safe drinking water supply should be implemented to reduce the prevalence and consequences of these infections in this population.

Keywords: intestinal parasitic infections; polyparasitism; schoolchildren.

Introduction

IPIs are among the most prevalent of human infections worldwide, causing significant morbidity and mortality. More than **3.5** billion people are affected, the majorities being children and have been termed as cancers of the developing nations **[1,2].**

Over **600** million school-aged children live in areas where these parasites are intensively transmitted, and are in need of treatment and preventive interventions **[3,4].** The lack of access to safe water, sanitation, and hygiene are the key factors for the high intensity of intestinal parasites that, in children, frequently have the clinical expression of malabsorption syndrome and gastrointestinal morbidity **[5].**

The feco-oral route is the most significant in the transmission of IPIs to humans. It can occur anthroponotically, zoonotically, food-borne or waterborne. When the water/soil is contaminated, the resilient infective forms **(eggs, cysts or spores)** of the pathogenic organisms can be transported to vegetables, fruit, hands, tools, handles doors, currency, etc. and be easily swallowed accidentally by humans. They can be transmitted also by flies **[6].**

Negative effects of helminth infections include diminished physical fitness and growth retardation, and delayed intellectual development and cognition **[7].**

In particular, intestinal protozoans, such as *Cryptosporidium* spp., *G. intestinalis* and *E. histolytica* are major causes of diarrhea in children **[8,9].**

Recent studies support the pathogenic nature of *Blastocystis*. It is now accepted that the classic clinical features of infection with this parasite include gastrointestinal symptoms such as nausea, anorexia, flatulence, and acute or chronic diarrhea **[9].**

There is a general acceptance that severe IPIs are likely to result in failure to thrive and poor educational performance. Moreover, recent studies highlighted the impact of polyparasitism on the child's immunity and showed that polyparasitism is associated with higher mortality rates and may increase the susceptibility to other infections relative to infection with a single parasite **[2].**

Subjects and methods

Ethics Statement

This cross sectional study was conducted from January **2015** to December **2015** after being authorized by the scientific ethics committee of our institute.

Participation of schoolchildren was voluntary after explaining the aim of the study. Individual written informed consents were obtained from enrolled children or their guardians before data and sample collection with brief explanation of the procedure and the purpose of the study.

Study Area

Sohag is one of the governorates of upper Egypt. The capital, Sohag city, is located **467** km south Cairo. It is composed of **11** districts stretched from North to South along the Nile.

Selection criteria

200 school-aged children between **6 and 12** years, who attend primary schools in our governorate, were randomly recruited from four primary schools to be included in the study namely, Yousof Al-Kashif**,** Nag' Fadel**,** Arab Al Atawla&Saqolta Al-Qadema Primary Schools.

Questionnaire survey

A standardized questionnaire was completed by children or their parents, who had given informed consent, in order to obtain a demographic description including the age, gender, residence, presence of symptoms and number of family members.

Parasitological methods

Stool specimens were collected in clean, dry, wide-mouth containers with tight-fitting lids. The acceptable amount of stool required for O/P examination ***is 2 to 5*** g, often referred to as the size of a walnut.

The specimen container was labeled with the patient's name and identification number and the date and time of sample collection.

Specimens were immediately preserved after passage due to the lag time till they reach the laboratory. Preservatives used in this study were formalin **10% in 100** cases and sodium acetate-acetic acid-formalin **(SAF)** in the other **100**.

Microscopic examination was done after performing the formalin ethyl acetate concentration technique followed by staining with the kinyoun's acid fast stain for intestinal coccidia **[3].**

Statistical analysis

Results were gathered, organized and tabulated in an Excel **2010** spreadsheet. Data were analyzed using **SPSS** program for Windows version **(22.0).** Quantitative data were expressed as means ± standard deviation. Qualitative data was expressed as frequencies and percentages. Chi-Square test **(χ^2)** and

Fisher's Exact test were used when appropriate for comparison between qualitative variables. used in this study. Univariate and Multivariate logistic regression analysis were performed for identification of certain risk factors for polyparasitism. A **5%** level was chosen as a level of significance in all statistical tests.

Family size was categorized into two groups **(<5 and ≥ 5 members),** and age of participants was categorized into two groups that were **<10 years and ≥ 10** years according to previous studies.

RESULTS

POPULATION PROFILE

200 schoolchildren aged between **6 and 12** years **(Age Mean ± SD = 8.9±1.9)** had participated in this study.

Table 1. Demographics of the studied children (n=200).

Demographic characteristics	n (%)
Age (Mean ± SD = 8.9±1.9)	
<10 years	**119 (59.5)**
≥10 years	**81 (40.5)**
Gender	
Girl	**97 (48.5)**
Boy	**103 (51.5)**
Residence	
Urban	**100 (50)**
Rural	**100 (50)**
Family size (Mean ± SD = 3.5 ±1.13)	
< 5 members	**42 (21)**
≥ 5 members	**158 (79)**

Overall, **127 (63.5%)** of the children were found to be positive for at least one intestinal parasite species **(Table 2).**

Table 2. Infection prevalence among the studied children (n=200).

	n	%
No. of parasitized children	**(127 /200)**	**63.5%**
No. of non-parasitized children	**(73/ 200)**	**36.5%**

Monoparasitized children constituted **80 (40%)** of children while **(23.5%)** were polyparasitized. This is demonstrated in table **3.**

Table 3. Types of parasitism (n=200).

Parasitism	n	(%)
Monoparasitism	80	40%
Polyparasitism*	47	23.5%

In the current study, **(23.5%)** of the studied children were polyparasitized. Out of them**, (17.5%)** had pure protozoal co-infections and **(6%)** were parasitized by protozoa and helminths simultaneously.

Table 4. Frequencies of protozoan and helminthic parasitisms (n=200).

	n (%)
Monoparasitism	**80(40)**
Helminths	**8 (4)**
Protozoa	**72 (36)**
Polyparasitism	**47 (23.5)**
Helminths+ Protozoa	**12 (6)**
Protozoa + Protozoa	**35 (17.5)**
No parasitism (n= 73)	**73 (36.5)**

The prevalence of infections with pathogenic protozoa is shown in table **5**. *Cryptosporidium* was the most common pathogenic protozoan with a prevalence of **68(34%)** followed by ***G. intestinalis*(14.5%), *E. histolytica*/ *dispar*(13%),** *Blastocystis* **(10.5%)** and ***C. cayetanensis* (6.5%)** based on microscopy.

Table 5.Parasite frequencies and percentages in descending manner.

	n (%)
Protozoa	
Pathogenic	
Cryptosporidium	**68 (34)**
G. intestinalis	**29 (14.5)**
E. histolytica/ dispar	**26 (13)**
Blastocystis	**21 (10.5)**
C. caytanensis	**13 (6.5)**
Non-pathogenic	
E. coli	**7 (3.5)**
I. butscilli	**5 (2.5)**
C. mesnilli	**3 (1.5)**
E. hartmanni	**3 (1.5)**
Helminths	
H. nana	**10 (5)**
A. lumbricoides	**4 (2)**
E. vermicularis	**4 (2)**
A. duodenale	**3 (1.5)**

Results showed that children exhibited various forms of poly-parasitism. **Two-**species infection was the most common **(24.4%)** followed by Three-species infection (**7.9%),** four-species infection **(3.9%)** and lastly one child had five-species infection **(0.8%).**

Table 6. Frequencies of different forms of polyparasitism (mixed protozoal&helminth infections).

	n	%
Double infection	**31**	**24.4**
Triple infection	**10**	**7.9**
Quadruple infection	**5**	**3.9**
Pentaple infection	**1**	**0.8**

Polyparasitism with two protozoan parasites were found **in (17.3%)** of children. The most common dual infection was with *E. histolytica/ dispar*and*Blastocystis* with a prevalence of **(3.1%).** In addition**, (2.4%)** of children exhibited triple infections with*E. histolytica/ dispar, G. intestinalis*and*Blastocystis*. Other cases of polyparasitism are shown in table **7.**

Table 7.Protozoal co-infections among parasitized children (n = 127).

	n (%)
Double infection	
E. histolytica/ dispar, Blastocystis	**4 (3.1)**
Giardia, Blastocystis	**3 (2.4)**
E. histolytica/ dispar, Giardia	**5 (3.9)**
Giardia, C. mesnilli	**1 (0.8)**
Giardia, E. coli	**5 (3.9)**
Blastocystis, I. butscilli	**2 (1.6)**
E. histolytica/ dispar, I. butscilli	**2 (1.6)**
Total	**22(17.3)**
Triple infection	
E. histolytica/ dispar , Giardia, Blastocystis	**3 (2.4)**

Results revealed that the type of polyparasitzed children were more frequently symptomatized than monoparasitized children. A statistical significance was found between diarrhea and the type of parasitism. P-value was **0.012** as shown in table **8.** Other symptoms showed no statistical significance.

Table 8. Relation between symptoms and type of parasitism amongst parasitized children (n=127).

	Monoparasitism (N=80)	Polyparasitism (N=47)	P-value
Diarrhea			
Yes	**12 (42.9%)**	**16 (57.1%)**	**0.012***
No	**68 (68.7%)**	**31 (31.3%)**	
Pain			
Yes	**6 (42.9%)**	**8 (57.1%)**	**0.098**
No	**74 (65.5%)**	**39 (34.5%)**	
Dysentery			
Yes	**0 (0.0%)**	**2 (100%)**	**0.135**
No	**80 (64%)**	**45 (36 %)**	
Perianal itching			**0.894**
Yes	**2 (66.7%)**	**1 (33.3%)**	
No	**78 (62.9%)**	**46 (37.1%)**	

P- value was calculated by Chi square test and Fisher's Exact Test
***Statistically significant**

RISK FACTORS FOR POLYPARASITISM

Rates of polyparasitism were relatively lower than monoparasitism in both age groups. No statistical significance was denoted. P value was **0.425.**

Boys and girls from both urban anr rural areas were more commonly monoparasitized. No statistical significance was found.

Also family size did not significantly affect the type of parasitism. P value was **0.295.**

Table 9. Type of parasitism and demographic features among infected children (127).

	Monoparasitism (N=80)	Polyparasitism (N=47)	P-value
Age			
< 10 years	**50 (65.8%)**	**26 (34.2%)**	**0.425**
≥ 10 years	**30 (58.8%)**	**21 (41.2%)**	
Sex			**0.425**
Boys	**45 (66.2%)**	**32 (33.8%)**	
Girls	**35 (59.3%)**	**24 (40.7%)**	
Residence			**0.919**
Urban	**35 (62.5%)**	**21 (37.5%)**	
Rural	**45 (63.4%)**	**26 (36.6%)**	
Family size			**0.295**
< 5 members	**14 (73.7%)**	**5 (26.3%)**	
≥ 5 members	**66 (61.1%)**	**42 (38.9%)**	

P- value was calculated by Chi square test
***Statistically significant**

Univariate and multivariable logistic regression models were performed to identify risk factors that were expected to be significantly associated with intestinal polyparasitism. According to our data, none of the age, gender, residence or family size were risk factors for polyparasitism. See tables **10 & 11**.

Table 10.Univariate logistic regression analysis of factors associated with polyparasitism.

	OR (CI 95%)	P - value
Age	**0.7 (0.4 – 1.5)**	**0.426**
Sex	**0.7 (0.3 – 1.5)**	**0.425**
Residence	**0.9 (0.5– 1.9)**	**0.919**
Family size	**1.8 (0.6 – 5.3)**	**0.3**

OR, Odds ratio.CI, Confidence interval.
Significant association (unadjusted P,0.05).

Table 11. Multivariate logistic regression analysis of factors associated with polyparasitism.

	Adjusted OR (CI 95%)	P - value
Age	**0.8 (0.4 – 1.7)**	**0.579**
Sex	**0.8 (0.4 – 1.6)**	**0.456**
Residence	**1.03 (0.5– 2.2)**	**0.936**
Family size	**1.7 (0. 5 – 5.1)**	**0.374**

OR, Odds ratio.CI, Confidence interval.
Significant association (P,0.05).

Discussion

IPIs are still public health problems in many communities, particularly among children in developing countries. **IPI** are associated with high morbidity particularly among schoolchildren **[2].** This study attempted to assess the prevalence of intestinal polyparasitism, and its associated risk factors.

The findings of the present cross-sectional survey conducted in four randomly chosen governmental Primary schools in urban and rural Sohag governorate, Egypt showed that **(63.5%)** of the participating children harboured at least one parsite species.

The almost non-changing situation of IPIs in Sohag is evident from the agreement of the present finding with that ofHamed*et al*., **2013** who reported the infection of **(63.31%)** of rural children aged less than **12** in rural Sohag **[11].**

In contrast, a lower prevalence rate of **(38.5%)** has been previously reported for IPIs among primary schoolchildren in Sohag **[12].** Direct smear examination and **FECT** were used for fecal examination, scotch tape for ***E. vermicularis*and** urine sample examination for *S. hematobium* but they did not perform any special staining procedures to detect *Cryptosporidium* spp.

However, Osman *et al*.**, (2016)** recorded an overall prevalence of **(85%)** among schoolchildren in Tripoli, Lebanon. This was explained by performing fecal examination by both microscopy and molecular tools in their study **[10].**

In this study,a predominance of protozoal compared to helminthic infections **(53.5% vs. 4%)** among schoolchildren was found. Such a prevalence is high relied on the collection of a single stool sample per child, instead of the ideal three consecutive samples.

Monoparasitism was the most predominant type of infection, being prevalent among **(40%)** of schoolchildren, while only **(23.5%)** were harboring double and triple infections. Our results are consistent with the results of across-sectional survey which was carried out in earlier **2011** by Matthys*et al*. in western Tajikistan where **(40.9%)** of all children had a single species infection, whereas **17.3%** had a dual species infection and **4.9%** harboured at least three intestinal pathogenic parasite species concurrently **[8].**

Co-infections were common, affecting **(32.5%)** of schoolchildren in the Plateau Central and Centre-Ouest regions of Burkina Faso as reported by Erismann*et al*., **(2017).** This indicates that these environments are extremely contaminated **[13].**

Cryptosporidium **(34%)** was strikingly found to be the highest prevalent parasite in this study, The major risk factor for *cryptosporidium* infection is drinking water that is contaminated with oocysts. Moreover, oocysts can survive in chlorine used for water treatment **[14].**

The most frequent protozoan detected in Central Maluku regency, Indonesia

schoolchildren was *Cryptosporidium* sp**. (24.7%) [15].** The authors reported using the modified Ziehl–Neelsen staining technique.

Today, *Blastocystis* spp. is considered an under-reported parasite, with a worldwide distribution and a prevalence far exceeding that of other intestinal parasites in the human population. Indeed, its prevalence can reach **100%** in developing countries and has been reported at between **1.5% and 20%** in industrialized countries **[10].**

Univariate and multivariate logistic regression models revealed that none of the age, gender, residence or family size were risk factors for polyparasitism. This is consistent with **(Al-Delaimy*et al*., 2014) [2].**

In the current study, **(23.5%)** of study population were polyparasitized. Out of them, **(17.5%)** had pure protozoalpolyparasitism and **(6%)** were parasitized by protozoa and helminths simultaneously.

22 (17.3%) of the parasitized children had two species infections. The most common dual infection was with *E. histolytica/ dispar*and*Blastocystis* spp. with a prevalence of **(3.1%).** In addition, **(2.4%)** of children exhibited triple infections with *E. histolytica/ dispar*, *G. intestinalis*and*Blastocystis*.

Jafari*et al***. (2014)** stated that interestingly *E. nana* and *C. mesnili* were seen more in humans with loose feces, but we should note that some co-infections between these protozoans has been observed and consequently we don't know which one/ones were responsible for loose feces **[16].**

Interestingly, **(5.4%)** of the polyparasitized children were infected by five different parasite species (mainly the three **STH** species, ***G. duodenalis*** and ***E. histolytica/ dispar*) [2].**

STUDY LIMITATIONS

First, the findings presented here cannot be generalized for all Sohag governorate schoolchildren. The random selection of schools with a sample size not large enough to be representative of the targeted community.

Second, this study was obliged to rely on a single fecal sample per child instead of the ideal three consecutive samples because of limitation of resources. Thus, the prevalence rate of parasitic infections is likely to be underestimated due to the temporal variation in egg excretion and cyst passage over hours and days.

A limitation of the present study was the use of infection prevalence but not infection intensity with STHs. Also, *S. mansoni* was never detected.

The shortcomings of this study perhaps made the rate of *E. vermicularis* infection clearly low among the most susceptible group, that is, schoolchildren.

Many villages with no road access and therefore were not covered by the current study. Thus, further investigations are necessary to more precisely evaluate the current status of the research problem.

Despite these limitations, our findings highlight a number of important issues. First, this study could be considered as a basis for conducting further studies that account for the determination of infection intensity, which could reflect the transmission dynamics of such parasites and their associated morbidity.

Conclusion

The findings of the present study revealed that IPIs are very common among schoolchildren in Sohag, Egypt. Intestinal polyparasitism is a significant problem among schoolchildren in Sohag, Egypt. Cryptosporidiosis was the most common infection detected. Hence, there is an urgent need to implement an innovative and integrated control program to reduce the prevalence and intensity of these infections significantly and to save these children from their negative impacts.

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