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TITLE: HELMINTHIC INFECTION A PUBLIC HEALTH PROBLEMS IN CHILDREN: A CROSS-SECTIONAL STUDY IN A TERTIARY CARE INSTITUTION OF CENTRAL INDIA.

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Abstract:
Background: About two billion people in the world's population are affected by the soil-transmitted helminth infection, with children being the most affected with Soil Transmitted Helminth infection.[1] India alone contributes nearly 25% to the total global cases, with 220.6 million children in need of preventive chemotherapy. [2] In view of the above context, the present study was conducted with the aim and objective of to the prevalence of soil-transmitted helminth (STH) infection and its risk factors. Material and Methods: Study Design: A Cross-sectional Observational Study Study Setting: Department of Microbiology, AIIMS, Raipur, from January 1 to June 30, 2023 (6 months). Sample Size: 384 ($n=Z^2$ 1-ά/2 $P(1-P)/d^{2)}$ P=50%, CI=95%, d=5% Absolute Precision. Results: STH prevalence was 13.3%, more in males (68.8%), and significantly high (62.5%). $P < 0.05$) in school-going children between 6 and 12 years of age. The predominant STH was *Ancylostoma duodenale* (56.2%); STH infection was much less (12.5%) in those practising handwashing with soap. Fifty percent of children had an STH infection even after receiving deworming within the past six months. More egg counts—216 eggs per gramme of faeces—were found in 29 cases by the KK method. *Entamoeba histolytica* (56.5%) was predominant among non-STH infections. Conclusion: Contrary to the assumption of 50% prevalence, the actual prevalence was very low, only about 13.3%. School-going children aged 6–12 years were more affected, and handwashing with soap was the key factor in preventing STH infection. The proportion of participants having toilet facilities and using footwear regularly had no role in STH prevention.

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Keywords: Soil-transmitted helminth (STH), Children, Kato-Katz (KK) method

MAIN TEXT:

Introduction: Our understanding of the prevalence of soil-transmitted helminth infections (STH) is necessary to plan control strategies and focus on highly endemic regions for preventive chemotherapy and improved sanitation facilities. India is known to be endemic to soil-transmitted helminth infections. The soil-transmitted helminths (STHs) are so-called as they require appropriate soil to develop into the infective form and transmit primarily through contaminated soil. [3] More than 1.5 billion people (24% of the world's population) are infected with STH. They are widely distributed in tropical and subtropical areas and affect the poorest and most deprived communities. STH are transmitted by eggs present in human faeces that contaminate soil in areas of poor sanitation. Over 267 million preschool-age children and over 568 million school-age children live in such areas and are in need of treatment and preventive intervention. [4] The species infecting humans are *Ascaris lumbricoides*, *Trichuris trichiura, Necator americanus*, and *Ancylostoma duodenale.* [4] STH infection impairs the nutritional status of the host in multiple ways, including increased malabsorption of nutrients and loss of iron, protein, and blood as they feed on host tissues, leading to chronic intestinal blood loss and anaemia. Some STH also causes loss of appetite and, therefore, a reduction in nutritional intake and physical fitness. In particular, *T. trichiura* can cause diarrhoea and dysentery. The global target is to eliminate morbidity due to soil-transmitted helminthiases in children by 2020. [4] In India, the reported prevalence of STH ranges from 4.8% to 66%, with varying prevalence rates for individual parasites. [5-8] In recent years, a number of steps have been initiated in India to decrease the morbidity of the STH through a mass drug administration programme involving school-going children and building sanitary latrines in urban as well as rural areas. Studies for STH prevalence are reported from different parts of India, but none of the studies were reported from Tribal State, Chhattisgarh. As the urban slums lack all types of basic amenities and the residents are often deprived of medical facilities due to a lack of awareness, the present study was conceived to determine the prevalence and pattern of common STH among children, including school-going children, in slum areas of Raipur and to assess risk factors associated with STH infection in them.

MATERIALS AND METHODS:

A cross-sectional observational study was conducted among children (3–15 years old) in two slums of District Raipur, Chhattisgarh, from January 1 to June 30, 2023. Study setting: Department of Microbiology, AIIMS,Raipur,Chhattisgarh. 384 samples (n = Z2 1-/2 P(1-P)/d2), with P = 50%, CI = 95%, and d = 5%. Absolute Precision. Out of 384, 24 were not responding. So a total of 360 subjects were enrolled in the analysis. Stool samples were collected from each child after getting written informed consent from their parents or guardians. As it was difficult to get population details in the slum area, we adopted purposive sampling for the study population selection by house-to-house visit; half of the calculated samples were from each slum area. Demographic information such as age, sex, toilet facility, parent's education, occupation, footwear habits, deworming medication intake, and personal hygiene were collected in a pretested, semi-open questionnaire. Training was given to Mitanin/ASHA for door-to-door Collection of stool samples where the subject is available.

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The samples were transferred to the microbiology laboratory in a cold box within 6 hours of collection. Statistics Collected data and examination findings were entered into an Excel sheet and analysed by SPSS Software version 26. The chi-square test was used for the level of significance set at $P < 0.05$. Laboratory procedures Direct microscopy was done with a saline mount for the presence of eggs, larvae, or any other finding (iodine mount if cyst) under ×10 and confirmed under ×40 magnification. The Kato-Katz (KK) technique was used to quantify the number of eggs per gramme of faeces. Kato-Katz technique A glass slide labelled with the sample number was taken, and a plastic template with a hole at the centre was placed on it. A small amount of faeces was put on a newspaper, and with the help of a nylon screen on top, the faeces were sieved using a spatula. Some of the sieved faeces were scraped up and put in the template to fill the hole (approximately holding 41.7 mg), avoiding air bubbles. Then, the template was lifted off carefully to leave the faeces on the slide. One piece of cellophane strip of the size of a cover slip was cut and immersed in glycerol malachite green (1 ml of 3% aqueous malachite green added to 100 ml glycerol and 100 ml water) and soaked for 24 h before use. The soaked cellophane was placed over a faecal sample. Then, the slide was inverted, and the faecal matter was firmly pressed over the cellophane to evenly spread the faeces between the slide and cellophane. Then, the slide was removed carefully without lifting off the cellophane. Excess faecal matter was cleaned, then the slide was allowed to dry and placed under a microscope with cellophane upwards and examined in a systematic manner, and the number of eggs of each species was reported. Finally, the number of eggs was multiplied by the appropriate number (24 for the 41.7 mg template) to give the number of eggs per gramme. [9-11]

RESULTS:

Among the studied children (n = 360), 95% were going to school, 70% were in the primary school, 80% of the houses had toilets, 70% of the children were using soap for hand washing, and the same proportion were regularly using footwear during a toilet visit. The overall positivity rate for parasites was found to be 117 (32.5%), STH was positive in 13.3% (48/360), and parasites other than STH were found in 19.16% (69/360). Among the 48 STH positives, 33 (68.8%) were males and 15 (31.3%) were females, and *A. duodenale* was found in 27 (56.2%) cases, followed by A. lumbricoides 6 (12.5%), and the rest of the children had mixed infections [Table 1]. School-going children (6–12 years) were affected more (62.5%) in comparison to smaller children (3-6 years), and the difference was found to be statistically significant [Table 2]. The history of receiving deworming medication in the last 6 months was found in 24 (50%) of STH-positive and 144 (46.2%) of STH-negative study participants, but the association between STH-negative participants and deworming treatment was not statistically significant. All the participants used to eat from roadside vendors, and all of them were of low socioeconomic status. Unfortunately, The participants having toilets at home and regularly using footwear had a higher proportion of STH infection (62.5% among regular footware users in comparison to nonusers, 37.5%) [Table 2]. STH infection was positive in six (12.5%) participants using soap for hand washing after a visit to the toilet. Out of 48 STH positive, saline mount detected egg counts of 1–2, 2–4, and 4–9/cover slip area in 28, 14, and

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6 cases, respectively. In contrast, the KK method detected 1–2, 2–4, and 4–9 eggs per cover slip area, or 48, 96, and 216 eggs per gramme of faeces in 6, 13, and 29 cases, respectively [Table 3]. *Entamoeba histolytica* (39/69–56.5%), *Giardia lamblia* (27/69–39%), and *Hymenolepis nana* (3/69–4.3%) were found among the participants having infections other than STH.

DISCUSSION:

The present study observed that the prevalence of STH was found to be 13.3%, which is closely related to the findings by Kattula *et al.*, where the prevalence was 5.9%–12.1%. [8] However, several other studies have shown higher prevalence rates ranging from 30%–70%. [12-16] Ganguli *et al.* had reported a still higher prevalence of 75.6% (95% CI: 71.2–79.5) in the state of Uttar Pradesh. [17] A recent review reported a prevalence of more than 50% in 19 different states of India, including Tamil Nadu, Andhra Pradesh, Bihar, Assam, and West Bengal. STH positivity was higher in males (33,68.8%) and school-going children (6–12 years) were affected more (62.5%) as compared to Avhad, Hiware, and Greenland *et al.*, where they reported 33.9% and 42.5% in the same age group, respectively. [18,19] The most common helminth found was *A. duodenale* (56.25%), followed by *A. lumbricoides* (20.83%). A mixed infection of *A. duodenale plus A. lumbricoides* was seen in 22.91% of children. Many studies reported *A. lumbricoides* as the most prevalent parasite, and nearly 90% have more than one parasite species in the same sample population. [13,20,21] However, Odinaka *et al*. reported a higher prevalence of *A. duodenale (94.2%)*. [15] The T. *trichiura* infection was not detected at all in the present study. The prevalence of *T. trichura* was 4.6% in a study from North India. [17] More than 50% of the prevalence of *T. trichiura* was reported in Assam and Andhra Pradesh. Jalaripet in Andhra Pradesh was a unique location from which a high prevalence of all three parasites—*A. lumbricoides (*91.12%), *T. trichiura* (71.5%), and hookworm (50.2%)—was reported. [20] Open defecation practises, poor handwashing and footwear use, a low literacy rate, and poor socioeconomic status were significant predictors of the prevalence of STH infections in most of the above studies. In the present study, the awareness regarding personal hygiene and sanitation habits was satisfactory among the study children, which was reflected by the higher percentage practising hand washing with soap and using toilets and footware while defecation. However, the proportion of STH infections was higher among the children who had toilet facilities at home and used their footwear regularly. Kalipan *et al*. had also reported that poor usage of footwear does not significantly increase the risk of STH infection. [14] There was a statistically significant association between hand washing with soap and the infection rate. Although deworming medication was provided regularly in government schools, in our study, half of the children had not received the medication in the past 6 months, and there was no significant difference in STH positivity between those who received deworming medication. Regarding STH detection, a high egg intensity of 4–9/cover slip area or 216 eggs/g of faeces could be detected by the KK method in 29 cases, whereas a saline mount could detect only 6 cases. The majority of saline mounts

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detected low egg intensity (1–2 eggs), where there is a chance of missing detection of STH. The efficacy of the KK method was also reported by many investigators. [12,17,19]

CONCLUSION:

The prevalence in the slums of Raipur city is much lower in comparison to other reported studies as well as recent reviews. The common age group affected was school-going (6–12 years), and hand washing habits with soap were the key factors detected to prevent STH infection in the community. KK is a simple and rapid method that can be used routinely to detect egg burden and monitor therapy. The limitation of this study was a limited sample size, as it was difficult to convince for consent as well as specimen collection and transportation within the stipulated time. However, a larger sample study in the future can be planned to strengthen the similar findings. Deworming interventions should be carefully monitored and documented for better coverage and compliance.

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Table-01 Distribution of helminths among positive children (*n*=48)

Name of helminths	***n* (%)**
A. duodenale	27 (56.25)
A. lumbricoides	10 (20.83)
A. duodenale + A. lumbricoides	11 (22.91)
Total	48 (100)

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Table-02 Demographic distribution with positivity of soil-transmitted helminth infection

Demographic profile	**STH positive *n* (%), (*n*=48)**	**STH negative *n* (%) (*n*=312)**	**Total *n* (%) (*n*=360)**	**χ2, df, *P***
Sex				
Male	33 (68.8)	207 (66.3)	240 (33.3)	0.10, 1, 0.74
Female	15 (31.3)	105 (33.7)	140 (66.7)	
School going age				
Toddlers (3)	6 (12.5)	12 (3.8)	18 (5.0)	8.28, 3, 0.04
Preschool (3-6)	6 (12.5)	66 (21.2)	72 (20.0)	
School going (6-12)	30 (62.5)	183 (58.7)	213 (59.2)	
Teen (12-15)	6 (12.5)	51 (16.3)	57 (15.8)	
Education status				
Preschool	15 (31.3)	69 (22.1)	84 (23.3)	5.81, 2, 0.05
Primary	27 (56.3)	225 (72.1)	252 (70.0)	
Higher secondary	6 (12.5)	18 (5.8)	24 (6.7)	
Availability of toilet at home				
Available	33 (68.8)	255 (81.7)	288 (80.0)	4.38, 1, 0.03
Not available	15 (31.3)	57 (18.3)	72 (20.0)	
Regular footwear use				
Yes	30 (62.5)	222 (71.2)	252 (70.0)	1.48, 1, 0.22
No	18 (37.5)	90 (28.8)	108 (30.0)	
Handwashing practice after defecation				
With soap	6 (12.5)	225 (72.1)	231 (64.1)	7.47, 2, 0.02
Without soap	27 (56.3)	39 (12.5)	66 (18.3)	
With other material	15 (31.3)	48 (15.4)	63 (17.5)	
Deworming (within last 6 months)				
Done	24 (50.0)	144 (46.2)	168 (46.7)	0.24, 1, 0.61
Not Done	24 (50.0)	168 (53.8)	192 (53.3)	
Total	48 (13.3)	312 (86.7)	360 (100)	

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Table-03 Distribution of worm burden in saline mount and per gram of faeces by Kato-Katz technique (*n*=48)

Number of eggs/cover slip area	Saline mount (*n*)	Kato-Katz count/gram of feces (multiplying factor=24) (*n*)
1-2	28	2×24=48 (6)
2-4	14	4×24=96 (13)
4-9	6	9×24=216 (29)