Original Research Article

A study of common parasites associated with peripheral blood eosinophilia in pediatric cases attending a tertiary care institute

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Abstract

Background: Eosinophils are a striking feature of many parasitic diseases. Helminthic infections are the most common parasitic diseases that produce eosinophilia in tropical countries. But the significance of association of high absolute eosinophilia with non-helminthic parasitic diseases is varied. **Aim and Objective:** The present study was aimed to identify common parasites associated with peripheral blood eosinophilia in pediatric cases, attending a tertiary care center in Lucknow, Uttar Pradesh with objective of determining the prevalence of various parasitic infections among children with peripheral blood eosinophilia. **Materials and Method:** In the present cross-sectional observational study children ≤14 years of age with absolute eosinophil count ≥440/µL, in routine blood counts were enrolled. Three consecutive stool samples were collected from children and dispatched to the microbiology laboratory. Faecal samples were inspected with naked eyes followed by microscopic examination using normal saline and lugol's iodine mounts on grease free slides. The findings observed in stool samples were recorded on the proforma. Peripheral blood smears were also prepared from the blood samples of participating children to detect microfilaria larva. **Results:** Out of 384 participants, 326 belonged in the range of mild eosinophilia, 50 in moderate eosinophilia and 8 in severe eosinophilia range. The prevalence of parasitic infection was 7% (27 out of 384 children). Out of 326 children of mild eosinophilia, 24 (7.4%) children had been detected with parasites in their faecal samples. Among the moderate eosinophilic children prevalence was 6%. In severe eosinophilic category none of them showed parasites in their stool samples. The association between mild, moderate and severe eosinophilia and parasitic findings was statistically non-significant (p=0.95).Out of total 27 children showing parasitic infection 2 children showed mixed type of infection while remaining 25 children harboured only one type of parasite in their stool samples. The most common parasite observed was *Giardia lamblia* (8 in single type and one case in mixed type). Among 223 male patients, 13 were positive cases (5.8%) and among 161 female patients, 14 were positive cases (8.7%). In the age group of 0-5 years of age 5.5% children harboured parasites. Among 6-10 years of age 9.3% children were found with parasites in their stool specimen. **Conclusion:** Thus we conclude that the prevalence of parasitic infection in children with eosinophilia was 7%. Commonly encountered parasites in the present study was cysts/ trophozoites of *Giardia lamblia* (2.3%), cysts/trophozoites of *Entamoeba histolytica* (2.1%), ova of *Hymenolepis nana* (1.8%), cysts of *Blastocystis hominis* (1.3%), and trophozoites of *Trichomonas hominis* (0.3%). The parasitic infection was seen commonly among females of 6-10 years of age group.

Key Words: Eosinophilia, parasitic infections, pediatric cases.

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Eosinophilia is a condition in which eosinophil count in peripheral blood exceeds 440-500 cells/microliter. Most common cause of eosinophilia is allergic reactions or parasitic infections. Helminthic infections are the most common parasitic diseases that produce eosinophilia. Nematode infections account for the majority of patients with eosinophilia in tropical countries, especially in areas where filariasis, ascariasis and hookworm infection are endemic[1]. The pattern and degree of eosinophilia in parasitic infections is determined by the development,

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migration, maturation, burden and distribution of the parasite within the host as well as by the host's immune response. Parasites tend to provoke marked eosinophilia when they or their products interact with immune effect or cells in tissues, chiefly during migration (e.g. trichinosis, ascariasis, gnathostomiasis, and filarial parasites). Provocation of eosinophils in blood is usually absent when there is a mechanical hurdle between the parasite and the host (e.g. adult tapeworms that are solely intraluminal or hydatid cysts that are enclosed in a cystic structure)[2,3]. It appears well proven that the presence of absolute eosinophilia is attributable, in a high percentage of cases, to the presence of helminthic infection [4-8].But the significance of association of high absolute eosinophilia with non-helminthic parasitic diseases is varied. Non-helminthic parasites such as protozoans,flagellates,Apicomplexans,coccidian parasites and ciliates pose increased risk to patients in terms of morbidity such as diarrhoea, malabsorption and abdominal discomfort and high mortality in untreated cases.

MATERIALS AND METHODS

The present cross-sectional observational study was done with the aim to screen the parasitic infections among paediatric cases with peripheral blood eosinophilia. The study was conducted in the Department of Microbiology of IIMSandR, Lucknow,over a period of six months from January 2017 to June 2017.The study was approved by the Institutional Review Committee (IRC) and the Ethical Review Committee (ERC). Following inclusion and exclusion criteria was used to select the study subjects.

Inclusion Criteria

- Children ≤14 years of age, attending the department of Pediatrics IIMSandR, Lucknow with absolute eosinophil count ≥440/µL, in routine blood counts.

Exclusion Criteria

- Children who suffered from known causes of eosinophilia such as allergies, asthma, atopic dermatitis, malignancies or immunodeficiency in the past or at the time of study were excluded.
- Children who had received antihelminthic drugs within 3 months of beginning of study were excluded.
- Accompanying guardian who were not willing to give consent for their children's participation in the study.

The sample size was calculated by using the formula $n = z^2 PQ/d^2$ with P=50 and d=5%. Thus by using the above mentioned inclusion and exclusion criteria total 384 children wereselected. A predesigned questionnaire was used to obtain detailed related clinical history and socio-demographic details such as age, gender, place of residence etc.To detect and identify intestinal parasites, three consecutive stool samples were collected from each study participant and dispatched to the microbiology laboratory of Integral Institute of Medical Sciencesand Research, Lucknow, where it was further processed. Faecal samples were inspected with naked eyes. The faeces were formed, semi formed, watery or loose. The colour was found to be yellow, green, brown or black. Faecal samples were also examined for any accompanying blood, pus, mucus, intestinal worms or proglottids etc. which could have indicated severe dysentery and the presence of the helminths. The stool sample was examined microscopically using normal saline and lugol's iodine mounts on grease free slides. The findings observed in stool sample were recorded on the proforma. Peripheral blood smears were also prepared from the blood samples of participating children to detect microfilaria larva.The collected data was entered in Microsoft excel. The categorical data was analyzed by calculating proportion.The Chi Square test was used to see the association among categorical variables. However, Fisher exact test was used in case of small frequency in different cells. $P<0.05$ was taken as significant.

RESULTS

Table 1: Prevalence of parasitic infection in eosinophilic children

AEC/µL (≥440/µL)	Total no.of cases	No. of positive cases	p value
Mild (440-1500/µL)	326	24 (7.4%)	
Moderate (1500-5000/µL)	050	03 (6.0%)	0.95
Severe (>5000/µL)	008	00 (0.0%)	
Total	**384**	**27 (7.0%)**	

It was seen that out of 384 participants, 326 belonged in the range of mild eosinophilia, 50 in moderate eosinophilia and 8 in severe eosinophilia range. It was observed that the prevalence of parasitic infection was 7% (27 out of 384 children). Out of 326 children of mild eosinophilia 24 (7.4%) children had been detected with parasites in their faecal samples. Among the moderate eosinophilic childrenprevalence was 6%. In severe eosinophilic category none of them showed parasites in their stool samples. The association between mild, moderate and severe eosinophilia and parasitic findings was statistically non-significant (p=0.95).

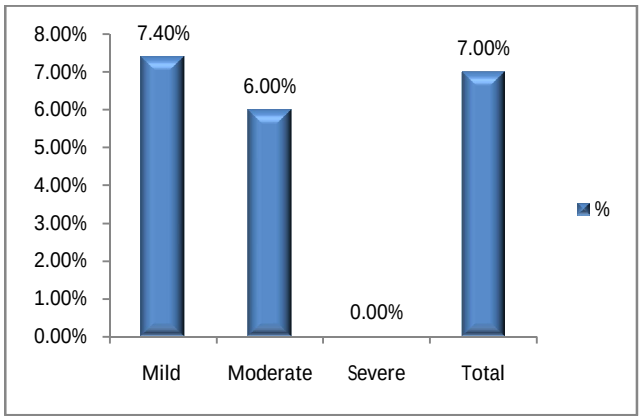

Figure 1: Prevalence of parasitic infection

Table 2: Parasites detected in children

Parasites	Single (one type of parasite)	Mixed infection	Total
Giardia lamblia	08	01	09(2.3%)
Entamoeba histolytica	06	02	08(2.1%)
Hymenolepis nana	07	00	07(1.8%)
Blastocystis hominis	03	02	05(1.3%)
Trichomonas hominis	01	00	01(0.3%)
Total	**25**	**05**	**30(7.8%)**

Out of total 27 children showing parasitic infection, 2 children showed mixed type of infection while remaining 25 children harboured only one type ofparasite in their stool samples.The most common parasite observed was *Giardia lamblia*(8 in single type and one case in mixed type).Thus total 9 children (2.3%) were found positive for Giardia *lamblia* in their stool specimen.In 6 children *Entamoeba histolytica* was observed as single infection while in 2 cases it was mixed infection. So, total 8 children (2.1%) had *Entamoeba histolytica* in their stool specimen.

Table 3: Distribution of parasites in mild, moderate and severe eosinophilic patients

Parasites	Mild	Moderate	Severe	Total
Giardia lamblia	6	2	0	8
Entamoeba histolytica	6	0	0	6
Hymenolepis nana	6	1	0	7
Blastocystis hominis	3	0	0	3
Trichomonas hominis	1	0	0	1
E. histolytica and *B. hominis*	1	0	0	1
E. histolytica, *G. lamblia* and *B. hominis*	1	0	0	1
Total	**24**	**03**	**00**	**27**

It was observed that majority of the parasites observed were in mild eosinophilia patients followed by moderate eosinophilia.

Table 4: Distribution of patients according to gender

	Total no. of cases	Positive cases	p value
Males	223	13 (5.8%)	0.38
Females	161	14 (8.7%)	
Total	**384**	**27 (7.0%)**	

It was seen that out of 384 children, 223 were males and 161 were females. Among 223 male patients, 13 were positive cases (5.8%) and among 161 female patients, 14 were positive cases (8.7%). Total Male: Female ratio was 1.4: 1 while, Positive Males: Positive Females were 0.92: 1. The association between male and female patient samples for parasites were statistically non-significant (p=0.38)

Table 5: Distribution of patients according to age groups

Age Group	No. of children in the study (n)	Positive cases	p value
0-5 years	145	08 (5.5%)	0.51
6-10 years	108	10 (9.3%)	
11-14 years	131	09 (6.9%)	
Total	**384**	**27 (7.0%)**	

Out of 145 children in the age groupof 0-5 years of age, 8 (5.5%) children harboured parasites. Among 6-10 years of age, 108 children were reported for eosinophilia. Out of these, 10 (9.3%) children were found with parasites in their stool specimen. Third group belonged to 131 children of 11-14 years of age. Out of these, 9 were found to be positive for parasites. The association between age groups for parasites were statistically non-significant (p=0.51). Peripheral blood smears were also prepared for detecting microfilariae larva from the blood samples of participating children but none of the samples showed the presence of microfilariae.

DISCUSSION

In the present study, out of total 384 patients, 27 were found to harbor parasites in their fecal samples showing prevalence rate of 7.03% in 6 months of study period. In a similar study done by Thakur and Rai[9], it was documented that worm infestation by stool or sera examination was seen in 37 (32%) children. Khadka et. al.[10], reported the prevalence of 15%in their study. Out of total 384 cases of eosinophilia, the most commonly encountered parasite was cysts/ trophozoites of *Giardia lamblia*. Other parasitic findings were cysts of *Entamoeba histolytica*, cysts of *Blastocystis hominis*, trophozoites of *Trichomonas hominis*, and ova of *Hymenolepis nana*. No other protozoa or helminthes were detected in the participants from this locality. Peripheral blood smears were prepared for detecting microfilariae larva from the blood samples of participating children but none of the samples showed the presence of microfilariae. In contrast Javier Pardo *et al*[4] in their study observed that

filariae (n =63, 29.6%) were the most frequently isolated parasite, followed by *schistosomes* (n=37, 17.4%), hookworms (n=36, 16.8%), and *Trichuris spp*. (n=18, 8.4%). This study is in contrast to our study as in our study it was found that protozoa were commonly detected in the participants as compared to helminthes. In a study done by Khanna et. al.[1], for the presence of eosinophilia, cases whose samples were positive for *Plasmodium vivax*, *Plasmodium ovale*, *Taenia solium*, *Echinococcus spp*., *Toxoplasma gondii*, *Leishmania donovani*, *Pneumocystis carinii*, *Trichomonasspp.* and *Entamoeba coli* had normal relative eosinophil count. However, *Trichuris trichiura*, *Ascaris lumbricoides*, *Strongyloides stercoralis*, filarial worm and hookworm accounted for eosinophilia in 88.9%, 87.5%, 63.6%, 53.8% and 35.5% of the cases respectively. In another similar study done by Khadka et. al.[10], it was seen that among 15 positive cases, 5 types of parasites were detected. The most common intestinal parasite was *Giardia lamblia* which was reported as 33.3% (5/15 cases).This finding is similar to present study where *Giardia lamblia* was most commonly encountered in our participants too. Occurrence of other intestinal parasites was 26.6% *Entamoebahistolytica* (4/15 cases), 26.6% *Ascaris lumbricoides* (4/15 cases), 6.6% *Trichuris trichura* (1/15 cases) and 6.6% Hook worm (1/15 cases) respectively. In our study,out of 384 children, 27 cases were detected with parasitic infections that had eosinophilia. In these 27 cases, 25 were detected with one type of parasites and 2 were detected with mixed infections. In a similar study conducted by Javier Pardo *et al*[4], among 161 eosinophilic cases 116 (54.5%) had 1 parasite, 30 (14.1%) had 2, and 15 (7.0%) had >3 types of parasites. *Giardia lamblia* was seen in 8 cases and in 1 case as mixed infection (total 9 cases), involving 2.3% of total population. 6 cases of *Entamoebahisolytica* were detected in this study and 2 were found with mixed infections (total 8 cases), invading 2.1% of participating population. In 7 cases *Hymenolepis nana* were detected with 1.8% of involvement in the population. 3 cases of*Blastocystis hominis* were detected while 2 cases appeared with mixed infections (total 5 cases) thus,infecting 1.3% of population. Also, 1 female child presented with the infection of *Trichomonas hominis* in this study showing 0.2% of involvement in population. Thus a total of 30 parasites were detected in this study. It could be seen that protozoal infections were more prevalent in this locality. Only one cestode is detected and no nematode was found during this study. Less number of parasite detection could be due to more awareness of people about healthcare these days. In rural areas most of the children wear footwear now, being barefooted otherwise could have been a risk factor for few of nematode infections. The absence of hookworm in participants of this study can be related to above mentioned cause. Blood samples were also taken to detect microfilariae in participants but none of the filarial case wasdetected. This could be attributed to less exposure of children to filarial worms in this locality. The area must not be prone to filarial infections. Age group could also be a factor for absence of microfilariae from the participants. It was seen that out of 384 children, 223 were males and 161 were females. Among 223 male patients 13 were positive cases (5.8%) and among 161 female patients 14 were positive cases (8.7%). Total Male: Female ratio was 1.4: 1 while, Positive Males: Positive Females were 0.92: 1. The association between male and female patient samples for parasites were statistically non-significant (p=0.38) In a similar study done by Khadka et. al.[10]prevalence of parasitic infection was 15% (15/100) in which 14% (7/50) were boys and 16% (8/50) were girls. Prevalence of parasitic infection was higher in girls than in boys but the difference was statistically also not-significant. This finding was similar to the present study. In contrast Thakur and Rai[9] in 2016, documented in their study that participating male: female ratio was 1.8:1 (M:73, F:41). It was seen that the prevalence of parasitic infection was maximum in 6-10 years of age (9.3%) followed by 11-14years of age (6.9%) and 0-5 years of age (5.5%). Similar findings were also observed by Khadka et. al.,[10]and Thakur and Rai[9] in their study.

CONCLUSION

Thus we conclude that the prevalence of parasitic infection in children with eosinophilia was 7%.Commonly encountered parasites in the present study was cysts/ trophozoites of *Giardia lamblia* (2.3%), cysts/trophozoites of *Entamoeba histolytica* (2.1%), ova of *Hymenolepis nana* (1.8%), cysts of *Blastocystis hominis* (1.3%), and trophozoites of *Trichomonas hominis* (0.3%).The parasitic infection was seen commonly among females of 6-10 years of age group.

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