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RESEARCH ARTICLE

IRON DEFICIENCY AND ANTHROPOMETRY IN CHILDREN INFECTED WITH *ENTAMOEBA HISTOLYTICA*

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ABSTRACT

A study examining the association of *Entamoeba histolytica* infection with anemia was conducted. A total of 44 children aged two months to twelve years-old were studied. Data were collected using structured questionnaires, anthropometric measurements and laboratory analysis of blood and stool samples. Analysis of the results showed that 65.90% of children were infected with *E.histolytica*, as well as significant differences were found among children with and without E.histolytic in Body Mass Index (P=0.02994), ferritin (P=0.00000649) and serum iron (P=0.00000311). *E.histolytica* infection was statistically correlated with iron deficiency anemia in this study population results showed that 13.79 % of infected children were considered to have iron deficiency anemia 65.51 % of infected children were at risk of iron deficiency anemia while no child of the non infected group was considered to have or were at risk of iron deficiency anemia.

INTRODUCTION

Entamoeba histolytica is an intestinal protozoan parasite; it is the third leading parasitic cause of death in humans after malaria and schistosomiasis. Globally, it is responsible for 40000–100000 deaths per year (Sebastiaan *et al*., 2007). It is distributed worldwide and causes an especially serious health threat in tropical and subtropical developing regions (Ohmishi et al., 2004) as well as it is a problem in the developed country (Haque *et al*., 2006).In Iraq literatures showed a different incidence level of *E.histolytica* infection ranges from 3.66% to 48% (Atia, 2009; Al-Warid, 2012). Iron deficiency can be defined as that moment when body iron stores become depleted and a restricted supply of iron to various tissues becomes apparent. Iron deficiency is one of the most common nutritional deficiencies worldwide (Raza *et al*., 2011) and it is mainly caused by low intake of bioavailable iron from diet, rapid growth, and iron losses due to intestinal malabsorption, or parasitic infections (De Vizia *et al*., 1992; Olivares *et al*.,2004) on other hand data on prevalence and intensity of infection with intestinal parasites, which are considered one possible cause of anaemia, are available only from small, unrepresentative sample surveys (Curtale *et al*.,1998). More data iron deficiency -related patterns of *E. histolytica* infection are needed in our country especially in children, so this study aims to evaluate serum iron, serum ferritin and hemoglobin among children with *E.histolytica* infection.

MATERIALS AND METHODS

Subjects

This study was conducted from May to July 2012 at AL-Kadimya Hospital for Pediatrics in Baghdad, Iraq. Forty four children were included in this study: 25(56.81%) males, and 19(43.19%) female, their ages range from 2 months-12 years. A qestionnaire about some clinical features were constructed and data on age, gender was gathered. Data collection was done in cooperation with children parents and physicians.

Anthropometry

All children were checked for anthropometric measurements as follows: the children were weighed without shoes using a portable digital electronic balance with a capacity of 0-150 ± 0.1 kg (IndiaMart, India) and the standing height was measured for children who could stand using a stadiometer, 205 ± 0.1 cm ((Holtain Ltd, UK), while recumbent length was measured for children who couldn't stand using a roll meter (KOH, Switzerland).To reduce intra-individual error, weight and height were measured twice and the mean value was used for the analysis

Blood Collection

Venous blood samples were collected from children and transferred to the laboratory for biochemical tests. Serum was separated by blood centrifugation at 3000 rpm for 5 minutes. Samples were stored in -20°C.

Serum ferritin

Serum ferritin concentrations were analyzed using VIDAS Ferritin kit (bioMérieux SA).

Serum Iron

Serum iron concentration was determined by a colorimetric assay using Iron-NP Colorimetric Test (Egyptian Co. for Biotechnology).

Hemoglobin concentration

Hemoglobin concentration was determined by the cyanmethemoglobin method, using a spectrophotometer (WHO, 2003).

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Fecal Examination

Fecal samples were collected from each child using a clean screw cap container, labeled with the number and date of collection. The samples were concentrated by formalin-ether method. A drop was taken from each deposit by pasture pipette and was smeared on a glass slide and then was examined by light microscope (Atia, 2009).

Statistical Analyses

Statistical analyses were performed with T-Tests and Chi square. The data were analyzed using SPSS statistical software (10^{th} version). Statistical significance was set at $p<0.05$.

RESULTS AND DISCUSSION

The results showed that the total infectivity rate of *E. histolytica* in this study was 65.90% (Figure 1). The number of positive samples was 29 out of 44 children; this percentage was higher than the other studies done in Iraq (Sa'el, 2009; AL-Warid, 2011)**.** The differences from one study to another may due to different factors such as: environmental, nutritional, socio-economic, geographical conditions, demographic and health-related behavior, number of patients samples in screening study and diagnostic method used (Atia, 2009; Norhayati *et al*., 2003; Ettehad, *et al*, 2010).

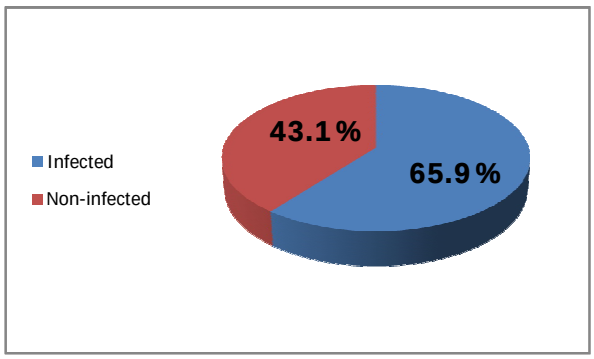

Figure 1 Percentage of infected and non infected children with *Entamoeba histolytica*

Results also showed that there were significant differences between infected children with *E.histolytica* and non infected children in Body Mass Index(BMI), higher BMI were noticed in non infected children 28.34 ± 12.09 kg/m^2 with range of 9.37-44.37, while the infected group showed low BMI 20.59 ± 12.57 kg/m^2 with range of 9.68-44.44 (Figure 2), the resultswas in agreement with the result of (Garba & Mbofung, 2010), who showed that mean body mass index was a remarkable difference between infected and non-infected children as well as they noticed that children infected with parasites were thinner than the non-infected ones, other also showed that the higher rates of intestinal parasitic infections frequently found in malnourished children with lower body mass index (Le *et al*., 2007). Some intestinal parasites produce adverse effects on weight gain wich may lead to inadequate food intake which in turn may cause poor appetite, metabolic and clinical disturbances (Garba & Mbofung, 2010).

Ferritin levels were also significantly varied in the current study (Figure 3); it was higher in non infected children 133.97 ± 55.03 ng/mL while the infected children showed lower levels 24.20 ± 21.16 ng/mL, these results were agreed with the results of (Le *et al*., 2007) who showed that the ferritin was in its low level in children infected with some intestinal parasites such as *Ascaris* and *Trichuris*, while our results were disagreed with the results of (Silva *et al*.,2009) who showed that ferritin serum concentrations were higher in infected than non infected children with *Giardia lamblia*, the differences between our result and the results of (Silva *et al*.,2009) may due to the following: age of children, the type parasites that infect the children and the sample size.

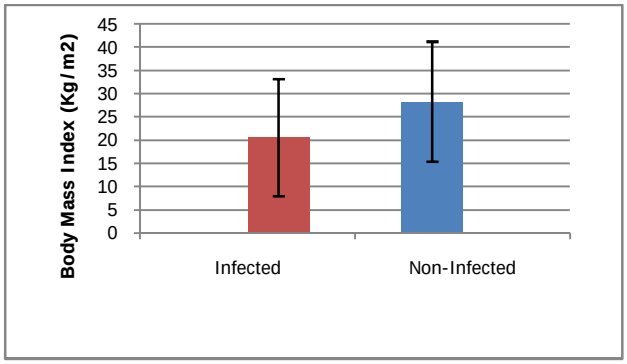

Figure 2 Body Mass Index of infected and non infected children with *Entamoeba histolytica*

Our results also showed that 13.79 % of infected children were considered to have iron deficiency anemia (serum ferritin < 12 ng/mL), as well as 65.51 % of infected children were at risk of iron deficiency anemia, (serum ferritin between 12-24 ng/mL) according to (WHO, 2001) while no child of the non infected group were considered to have or were at risk of iron deficiency anemia. On the other hand 20.68% of infected children and 100% of non infected children were not considered to have iron deficiency anemia or were at risk of iron deficiency anemia (Table 1), statistical analysis showed there were significant relation ($p < 0.05$) between infection with *E.histolytica* and the risk of iron deficiency anemia in children, this results agreed with the results of (Hesham *et al*., 2004) who reported that *E.histolyitica* is known to be predictors for iron deficiency anemia.

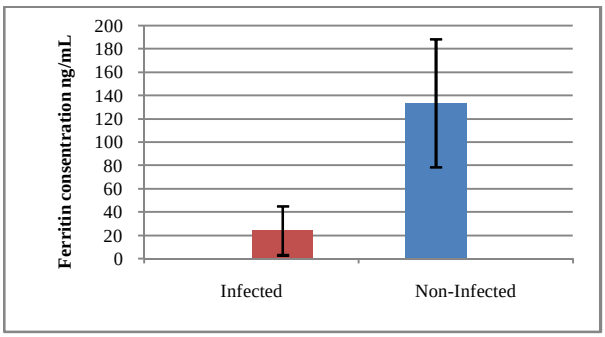

Figure 3 Ferritine concentration of infected and non infected children with *Entamoeba histolytica*

Results showed that there were significant differences ($p < 0.05$) in serum iron levels among infected children and non infected children, serum iron were in their high level in non infected children while it was in their low level in non infected children, these level were 80.83 ± 36.11 µg/dL and 29.13 ± 15.55 µg/dL respectively (Figure 4), this result was agreed with the result of (Olivares *et al*., 2004) who showed significant differences for serum iron between children with and without some intestinal parasites. *E. histolytica* requires a high concentration of iron to survive. This parasitic protozoan is able to obtain iron from the host proteins (Lopez-Soto, 2009) so heavy infection with *E. hostolytica* may cause decreasing in iron level in the host.

The current results showed that there were no significant differences between infected and non infected group in their

Table 1 Correlation between ferritin and infection with *E.histolytica*

Categories	Total Number of Cases	Number of Infected Children (%)	Number of Non-infected Children (%)
Children with iron deficiency anemia (serum ferritin < 12 ng/mL),	4	4(13.79)	0(0)
Children with iron deficiency anemia (serum ferritin 12-24 ng/mL),	19	19(65.51)	0(0)
Children without iron deficiency anemia (serum ferritin > 24 ng/mL),	21	6(20.68)	15(100)
	44	29	15

X^2, 24.926; degree of freedom (df), 2; P value, 0.00000387

hemoglobin concentration which were 11.20 ± 1.18 g/dL and 11.72 ± 1.73 g/dL respectively (Figure 5) as well as our results showed that 41.37% of anemic children (hemoglobin < 11g/dL) (WHO, 1968) were infected with *E.histolytica* while 40% of anemic subjects were non infected with this parasite, statistical analysis using Chi square showed that there was no significant relation ($p<0.05$) between infection with *E.histolytica* and the occurrence of anemia among children according to their hemoglobin concentration.

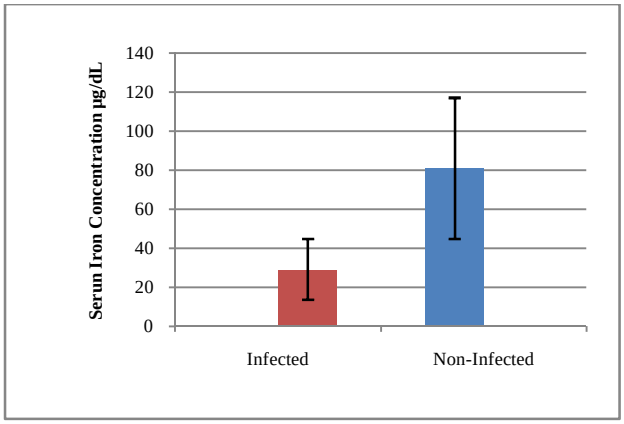

Figure 4 Serum iron concentration of infected and non infected children with *Entamoeba histolytica*

These result were disagreed with the result of (Le *et al*., 2007) who showed that children infected with some intestinal parasites showed a lower hemoglobin concentration and a higher prevalence of anemia, while (Al-Qadhi & Al-Warid, 2012) showed that there no association between hemoglobin concentration and infection with *E.vermicularis*.

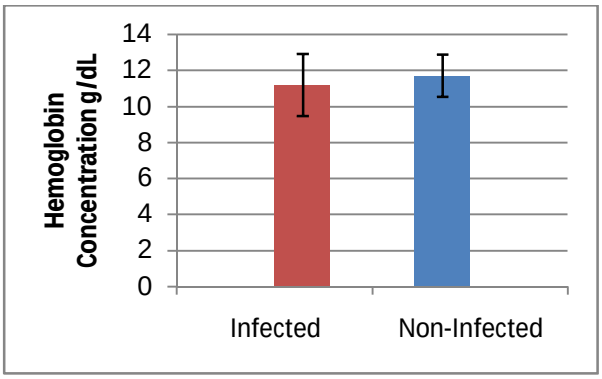

Figure 5 Hemoglobin concentration of infected and non infected children with *Entamoeba histolytica*

Our findings indicated that there were significant relation between *E.histolytica* infection and the prevalence of iron deficiency anemia depending on ferritin and serum iron, these findings were agreed with other findings (Breymann, 2001) which suggested that serum ferritin level still the best standard to evaluate iron deficiency, as well as we can recommend future studies to detect the prevalence of anemia in patients infected with other parasites.

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