

Association of *Giardia Duodenalis* Infection with Serum Vitamin A Status in Children: A Comparative Case Control Study

Mohammed H. Dakhil AL-Ghazzi^{1*}, Khaldoun M. Al-Rishawi², Ali Moaid Raheem³

¹Department of Medical Laboratory Technologies, College of Health and Medical Technologies, Imam Jaafar Al-Sadiq University/ AL-Muthanna, Iraq

^{2,3}Directorate General of Education in Al- Muthanna, Ministry of Education, Iraq

ABSTRACT

Background: *Giardia duodenalis* remain as common intestinal protozoan parasitic infection all over the world. Therefore, current study was purposed to explore the association of serum vitamin A concentrations with infection of *Giardia duodenalis* in children.

Methods: This comparative case control study was done on 50 children aged 5 to 10 years, and was conducted on 25 *G. duodenalis* infected children and 25 apparently healthy children as control in Al-Muthanna and Al-Diwaniyah (Iraq). The stool samples were assessed microscopically for incidence of *G. duodenalis* cysts and trophozoites. Blood samples were collected and serum was separated to measure vitamin A concentrations.

Results: The serum vitamin A level in *Giardia*-infected group was less (20.48 ± 9.91 µg/dL) compared to control (31.48 ± 6.19 µg/dL) group. The variation between groups was notably significant ($p < 0.001$). The median vitamin A concentration was (16 µg/dL) in infected children and (32 µg/dL) in controls. Serum vitamin A concentrations ranged from (11 to 45µg/dL) in infected group and from (20 to 44µg/dL) in control group. Sixteen infected children (64.0%) had vitamin A concentrations of (≤ 20 µg/dL), compared with only two children (8.0%) in the control group. The difference in frequency of low vitamin A concentrations between two groups was remarkable significant ($p < 0.001$).

Conclusion: Children infected with *G. duodenalis* had significantly lower serum vitamin A level than children in control group. The high proportion of infected children with low vitamin A concentrations supports a significant association between giardiasis and impaired vitamin A status. This relationship may be related to intestinal dysfunction, malabsorption, reduced dietary intake, or infection-associated alterations in vitamin A metabolism.

KEYWORDS: *Giardia duodenalis*, vitamin A, malabsorption, blood samples.

INTRODUCTION

In general *Giardia duodenalis* is an intestinal flagellated protozoan parasite of great importance in public health. It is described by other names as well, such as *Giardia lamblia* and *Giardia intestinalis*. It is found in humans and many species of mammals and is one of the most common intestinal parasites of the world [1].

Giardiasis is very common in developing countries; over 200 million cases are diagnosed worldwide each year. It has been a part of the World Health Organization's Neglected Diseases Initiative since 2004 [2].

It mainly infects the upper small intestine, where the trophozoite stage exists in the intestinal epithelium and grows extracellularly. The infective stage is the environmentally resistant cyst which is used to transmit disease from one host to another. Transmission to humans is primarily fecal-oral route via ingestion of cysts in contaminated water, food or by direct person-to-person transmission [3-5]

Children are especially vulnerable to giardiasis as are people living in areas with poor access to clean drinking water, sanitation and proper hygiene. Infection symptoms can range from being mild to severe. While some infected people do not get symptoms, others experience acute or chronic gastrointestinal illness with diarrhea, abdominal pain, abdominal cramps, nausea, vomiting, bloating, excessive intestinal gas and loss of appetite [6,7].

Persistent or chronic giardiasis may have important nutritional consequences. When the small intestine is inflamed, absorptive and digestive processes may be disrupted and malabsorption and malnutrition may be caused. In the past, giardiasis has been linked to stunted growth, malabsorption and interference with the absorption of various nutrients such as fat soluble vitamins (8-10).

Association of *Giardia Duodenalis* Infection with Serum Vitamin a Status in Children: A Comparative Case Control Study

Vitamin A is a vital and fat soluble micronutrient and is important for vision, immunity, epithelial integrity, cell differentiation, growth and development. It is especially important in children that there is an adequate vitamin A status because deficiency can compromise immune functions and predispose children to infection [11-13]. A serum retinol/serum or plasma retinol level below 0.70 $\mu\text{mol/L}$ is indicative of protein-bound retinol deficiency while a level below 0.35 $\mu\text{mol/L}$ is considered severe deficiency as per the World Health Organization [14].

The relationship between the occurrence of giardiasis and vitamin A status is biologically plausible as known. Vitamin A is fat soluble and will be absorbed only if there is sufficient digestion in the gastrointestinal tract and absorption capability in the intestine. Thus, damage or disturbances of function in the small intestine can reduce the absorption of vitamin A [15]. The present study was designed to determine the correlation between *Giardia duodenalis* infection and serum Vitamin A concentration in children (5-10 years) of Al-Muthanna and Al-Diwaniyah Governorates, Iraq.

METHODS

Patients and study design

This study was designed as a case-control comparative investigation study with the participation of 50 children attended private medical laboratories in the governorates of Al-Muthanna and Diwaniyah in southern Iraq during a six-month period from the beginning of February to the end of July 2026. Before starting the laboratory procedures, the consent of each participant's parents was confirmed. The study included both males and females aged between (5-10) years; those of other ages and those who declined to participate were excluded.

The study was categories into two groups:

- *Giardia*-infected group: 25 children with confirmed *Giardia duodenalis* infection.
- Control group: 25 apparently healthy children from the same geographical region.

The control group allowed comparison of serum vitamin A concentrations between children with giardiasis and apparently healthy children. The data were processed descriptively using SPSS software, tabulated and categorized, and differences were identified using appropriate statistical tests, including Welch's test for independent samples and Fisher's exact test, with $p < 0.001$ considered statistically different.

Stool Sample Collection

Children's stool samples were taken in clean, sterile, properly labeled containers. The samples were collected and immediately transported to the laboratory for examination and processed to maintain the morphometric features of protozoan parasites.

Microscopic Examination

Direct microscopic examination was carried out with wet mounts. A few drops of fresh stool were placed on a clean glass slide and covered with a slide cover. Further preparations were done with Lugol's iodine solution for better visualization of internal structures of protozoan cysts. The prepared slides were examined under a light microscope. *Giardia duodenalis* trophozoites and Cysts were identified according to its characteristic morphological features, as shown in figure (1). Children in the infected group were considered positive when *G. duodenalis* cysts and/or trophozoites were detected during microscopic examination.

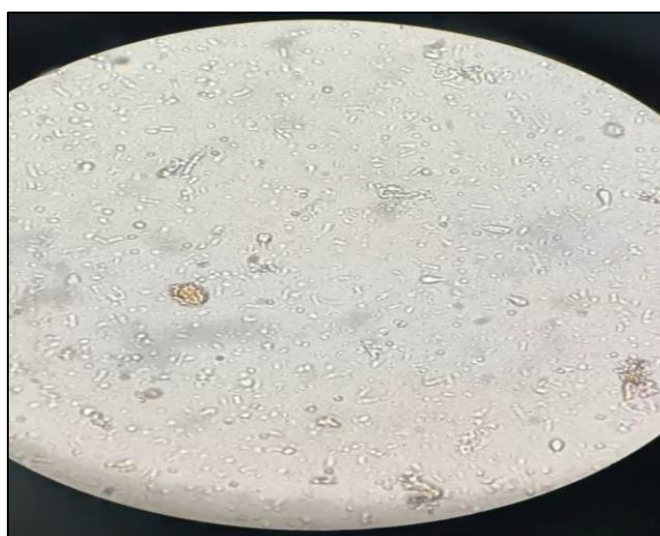


Figure (1): showed *Giardia lamblia* cyst under 40x.

Association of *Giardia Duodenalis* Infection with Serum Vitamin A Status in Children: A Comparative Case Control Study

Blood Sample Collection and Serum Preparation

5 mL of venous blood was collected from each participant using sterile serum-separation tubes. Following collection, the blood samples were allowed to clot under appropriate conditions. The samples were subsequently centrifuged at $1000 \times g$ for approximately 20 minutes to separate the serum. The separated serum was transferred into clean aliquots and stored at -20°C until vitamin A analysis.

Determination of Serum Vitamin A

Serum vitamin A concentrations were determined using an enzyme-linked immunosorbent assay (ELISA) according to the kit manufacturer's instructions.

Before analysis, serum samples and reagents were allowed to equilibrate to room temperature.

- Serum samples were diluted using phosphate-buffered saline according to the manufacturer's recommended procedure.
- Calibration standards were prepared according to the kit protocol.
- 50 μL of each standard and diluted serum sample was added to the appropriate wells of a microtiter plate. Detection reagent was subsequently added to each well.
- The plate was incubated at 37°C for the recommended incubation period by the manufacturer.
- Wells were washed several times after incubation to get rid of any unbound materials.
- Then each well was filled with (TMB) substrate solution and the plate was incubated in the dark to develop color.
- The reaction of the enzymes was stopped with the stop solution provided.
- Absorbance was determined by microplate reader at wavelength recommended by the manufacturer.
- The calibration curve determined from the standards was used to calculate the amounts of vitamin A present.
- All vitamin A values used in present study are given in $\mu\text{g/dL}$.

Interpretation of Vitamin A Concentrations

The WHO recommends that a serum retinol level below $0.70\mu\text{mol/L}$ is an indicator of subclinical vitamin A deficiency in the population and below $0.35\mu\text{mol/L}$ is severe deficiency.

RESULTS

All 25 stool samples were positive for *Giardia duodenalis* cysts and/or trophozoites when examined under a microscope from the infected group.

These children experienced gastrointestinal and systemic complaints such as diarrhea, steatorrhea or fatty diarrhea, abdominal pain, cramps, nausea, abdominal bloating, excess intestinal gas, loss of appetite, headache and fever.

The clinical symptoms are typical of symptomatic giardiasis and can lead to decreased appetite, poor nutritional absorption and impaired nutrition.

A marked difference in serum vitamin A concentrations was observed between *Giardia*-infected children and the control group. As explained in Table (1) mean serum vitamin A concentration among *Giardia*-infected children was $20.48 \pm 9.91 \mu\text{g/dL}$. On the contrary, control children had a mean serum vitamin A concentration of $31.48 \pm 6.19 \mu\text{g/dL}$. The median vitamin A concentration was $16 \mu\text{g/dL}$ in infected group and $32 \mu\text{g/dL}$ in control children. The vitamin A concentration ranged from 11 to $45 \mu\text{g/dL}$ among infected children and from 20 to $44 \mu\text{g/dL}$ among controls.

Table 1: Vitamin A serum concentration levels between the two groups

Parameter	Groups	
	Giardia-infected (n=25)	Control (n=25)
Mean ($\mu\text{g/dL}$)	20.48	31.48
Standard deviation	9.91	6.19
Median ($\mu\text{g/dL}$)	16	32
Minimum ($\mu\text{g/dL}$)	11	20
Maximum ($\mu\text{g/dL}$)	45	44
Range ($\mu\text{g/dL}$)	34	24

The difference in mean serum vitamin A concentration between the two groups was $11.00 \mu\text{g/dL}$. Statistical analysis demonstrated that serum vitamin A concentrations were significantly lower in *Giardia*-infected children than in the control group ($p < 0.001$).

Association of *Giardia Duodenalis* Infection with Serum Vitamin A Status in Children: A Comparative Case Control Study

The frequency of low vitamin A concentrations was substantially higher among *Giardia*-infected children. Sixteen of the 25 infected children (64.0%) had serum vitamin A concentrations ≤ 20 $\mu\text{g/dL}$. In contrast, only two of the 25 control children (8.0%) had concentrations ≤ 20 $\mu\text{g/dL}$.

Table 2: Frequency of low serum vitamin A concentrations

Study group	Vitamin A ≤ 20 $\mu\text{g/dL}$	Vitamin A > 20 $\mu\text{g/dL}$	Total
Giardia-infected children	16 (64.0%)	9 (36.0%)	25
Control children	2 (8.0%)	23 (92.0%)	25

The difference in the frequency of low vitamin A concentrations between two studied groups was considerable ($p < 0.001$). These findings indicate that children infected with *G. duodenalis* were considerably more likely to have low serum level of vitamin A compared to control children.

DISCUSSION

The current study designed to evaluate associated of *Giardia duodenalis* infection with serum vitamin A level in children (5-10 years) from Al-Muthanna and Al-Diwaniyah Governorates, Iraq. The main result showed that children with *G. duodenalis* infection had significantly lower serum vitamin A concentrations than apparently healthy children from the control group. In addition, 64.0% of children with infection had vitamin A levels ≤ 20 $\mu\text{g/dL}$ while 8.0% of control group had levels in this range. This significant difference is indicative of a high level of association between *Giardia* infection and low vitamin A levels in the population studied.

Undoubtedly, chronic infection can cause disruption in intestinal activity and malabsorption, as it is the main common site for the absorption of nutrients and for infection with the intestinal giardia [16,17].

Previous studies have shown that impaired absorption of vitamin A can be seen in children with giardiasis and improvement after successful eradication of giardiasis [18-20]. Other studies indicated that *Giardia* infection could impact vitamin A status through intestinal absorption and hepatic vitamin A metabolism. This study included school children, with evidence that giardiasis was linked to changes in vitamin A liver stores and that treatment of the parasite resulted in improvement of the vitamin A-related indicators [21].

The finding of the present results is also in agreement with the general knowledge that chronic intestinal parasites infection may lead to nutritional defects [22]. Malabsorption syndrome, growth disturbances, and malnutrition have been linked to *Giardia* infection, especially when it occurs repeatedly and over extended periods, particularly in children [23]. There are several scientific explanations for the low levels of this vitamin in affected children. Giardiasis can disrupt nutrient absorption in the intestines, and because it is a fat-soluble vitamin, any impairment in fat digestion or absorption can reduce its absorption. In addition, chronic diarrhea induces nutrient loss and reduces its absorption. Affected children may also experience loss of appetite, which can lead to a decrease in their consumption of foods rich in this vitamin [24].

Moreover, dietary vitamin A intake, overall nutritional status, socioeconomic factors, repeated gastrointestinal infections, other intestinal parasites and inflammatory status and differences in access to health and nutrition might account for the differences observed between groups [25,26]. The results also have potential public health implications, and assessment of nutritional status may be especially significant in children with intestinal parasitic infections in areas where such infections are endemic. Finally, proper nutrition coupled with timely diagnosis and treatment of giardiasis may contribute in health improving of affected children.

CONCLUSION

This study confirmed a considerable relationship between *Giardia duodenalis* infection with level of serum vitamin A in children aged 5- to 10-year-old in Al-Muthanna and Al-Diwaniyah Governorates in Iraq. The results indicate that children with giardiasis are likely to be vitamin A deficient. The possible mechanism involves intestinal dysfunction, malabsorption, diarrhoea, decreased food intake and change in vitamin A metabolism due to the infection.

REFERENCES

- 1) Adam, R. D. (2021). *Giardia duodenalis*: biology and pathogenesis. Clinical microbiology reviews, 34(4), e00024-19.
- 2) Savioli, L., Smith, H., & Thompson, A. (2006). *Giardia* and *Cryptosporidium* join the neglected diseases initiative. Trends in Parasitology, 22(5), 203–208.
- 3) Faubert, G. (2000). Immune response to *Giardia duodenalis*. Clinical microbiology reviews, 13(1), 35-54.

Association of *Giardia Duodenalis* Infection with Serum Vitamin a Status in Children: A Comparative Case Control Study

- 4) Silva, R. K., Pacheco, F. T., Martins, A. S., Menezes, J. F., Costa-Ribeiro, H., Ribeiro, T. C., et al. (2016). Performance of microscopy and ELISA for diagnosing *Giardia duodenalis* infection in different pediatric groups. *Parasitology International*, 65(6), 635–640.
- 5) Cernikova, L., Faso, C., & Hehl, A. B. (2018). Five facts about *Giardia lamblia*. *PLoS Pathogens*, 14(9), e1007250.
- 6) Ali QA, Ghareeb OA. Drinking Water Quality and Its Impact on Public Health: Review. *Academia Repository*, Sep. 11, 2023; 4(9): 48-64.
- 7) Bhise, M. P., Padhi, A., Chaudhary, A., Rajyaguru, B., Chauhan, K., Meena, P., & Agrawal, A. (2025). Giardiasis in pediatric populations: A case series highlighting nutritional and growth impacts. *Journal of Family Medicine and Primary Care*, 14(11), 4883.
- 8) Belkessa, S., Ait-Salem, E., Laatamna, A., Houali, K., Sönksen, U. W., Hakem, A., ... & Stensvold, C. R. (2021). Prevalence and clinical manifestations of *Giardia intestinalis* and other intestinal parasites in children and adults in Algeria. *The American journal of tropical medicine and hygiene*, 104(3), 910.
- 9) Barasa, E., Indusa, W., & Were, T. (2025). *Giardia duodenalis* in human health: A comprehensive review of its epidemiology, transmission, pathophysiology, diagnostics, and control strategies. *Afro-Egyptian Journal of Infectious and Endemic Diseases*, 15(2), 109-126.
- 10) Trelis, M., Taroncher-Ferrer, S., Gozalbo, M., Ortiz, V., Soriano, J. M., Osuna, A., & Merino-Torres, J. F. (2019). *Giardia intestinalis* and fructose malabsorption: a frequent association. *Nutrients*, 11(12), 2973.
- 11) Awasthi, S., & Awasthi, A. (2020). Role of vitamin A in child health and nutrition. *Clinical Epidemiology and Global Health*, 8(4), 1039–1042.
- 12) Kundu, S., Rai, B., & Shukla, A. (2021). Prevalence and determinants of Vitamin A deficiency among children in India: Findings from a national cross-sectional survey. *Clinical Epidemiology and Global Health*, 11, 100768
- 13) Rao, S., Li, T., Li, X., Hou, L., & Sun, W. (2025). Vitamin A and its related diseases. *Food Science & Nutrition*, 13(7), e70630.
- 14) Bastos Maia, S., Costa Caminha, M. D. F., Lins da Silva, S., Rolland Souza, A. S., Carvalho dos Santos, C., & Batista Filho, M. (2018). The prevalence of vitamin A deficiency and associated factors in pregnant women receiving prenatal care at a reference maternity hospital in Northeastern Brazil. *Nutrients*, 10(9), 1271.
- 15) Quihui-Cota, Astiazarán-García, Valencia, Morales-Figueroa, Lopez-Mata, & Ortiz, V. (2008). Impact of *Giardia intestinalis* on vitamin A status in schoolchildren from northwest Mexico. *International journal for vitamin and nutrition research*, 78(2), 51-56.
- 16) Fauziah, N., Aviani, J. K., Agrianfanny, Y. N., & Fatimah, S. N. (2022). Intestinal parasitic infection and nutritional status in children under five years old: a systematic review. *Tropical medicine and infectious disease*, 7(11), 371.
- 17) Solaymani-Mohammadi, S. (2022). Mucosal defense against giardia at the intestinal epithelial cell interface. *Frontiers in immunology*, 13, 817468.
- 18) Roshidi, N., Hassan, N. H. M., Hadi, A. A., & Arifin, N. (2021). Current state of infection and prevalence of giardiasis in Malaysia: a review of 20 years of research. *PeerJ*, 9, e12483.
- 19) Lima, A. A., Soares, A. M., Lima, N. L., Mota, R. M., Maciel, B. L., Kvalsund, M. P., ... & Guerrant, R. L. (2010). Effects of vitamin A supplementation on intestinal barrier function, growth, total parasitic, and specific *Giardia* spp infections in Brazilian children: a prospective randomized, double-blind, placebo-controlled trial. *Journal of pediatric gastroenterology and nutrition*, 50(3), 309-315.
- 20) De Medeiros, P. H. Q., Pinto, D. V., De Almeida, J. Z., Rêgo, J. M., Rodrigues, F. A., Lima, A. Â. M., ... & Oriá, R. B. (2018). Modulation of intestinal immune and barrier functions by vitamin A: implications for current understanding of malnutrition and enteric infections in children. *Nutrients*, 10(9), 1128.
- 21) Astiazaran-Garcia, H., Lopez-Teros, V., Valencia, M. E., Vazquez-Ortiz, F., Sotelo-Cruz, N., & Quihui-Cota, L. (2011). *Giardia lamblia* infection and its implications for vitamin A liver stores in school children. *Annals of Nutrition and Metabolism*, 57(3-4), 228-233.
- 22) Ghareeb OA, Ali QA, Abed QJ. Iodine: a micronutrient essential for human development and survival. *Disease and Health Research developments*. BP International. 2024 Dec 18:18-31.
- 23) Prabakaran, M., Weible, L. J., Champlain, J. D., Jiang, R. Y., Biondi, K., Weil, A. A., ... & Ojo, K. K. (2023). The gut-wrenching effects of cryptosporidiosis and giardiasis in children. *Microorganisms*, 11(9), 2323.
- 24) Rogawski, E. T., Bartelt, L. A., Platts-Mills, J. A., Seidman, J. C., Samie, A., Havt, A., ... & MAL-ED Network Investigators. (2017). Determinants and impact of *Giardia* infection in the first 2 years of life in the MAL-ED birth cohort. *Journal of the Pediatric Infectious Diseases Society*, 6(2), 153-160.

Association of *Giardia Duodenalis* Infection with Serum Vitamin a Status in Children: A Comparative Case Control Study

- 25) Fauziah, N., Aviani, J. K., Agrianfanny, Y. N., & Fatimah, S. N. (2022). Intestinal parasitic infection and nutritional status in children under five years old: a systematic review. *Tropical medicine and infectious disease*, 7(11), 371.
- 26) Kleinert, M., Górski, M., Kwiatkowski, A., Lepich, T., & Polaniak, R. (2026). Gastrointestinal disorders due to parasites—a narrative review of the literature. *Pomeranian Journal of Life Sciences*, 72(2).