

Research Article

Assessment of Interleukin38 and Some Parameters in Patients With Giardiasis

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How to cite this article:

Lahmod M A, Hussein M H, Assessment of Interleukin38 and Some Parameters in Patients With Giardiasis. *Epidem Int.* 2026;11(1):13-18.

Date of Submission: 2026-02-16

Date of Acceptance: 2026-03-19

A B S T R A C T

Background: Giardiasis, caused by the parasite protozoan *Giardia lamblia*, is regarded as one of the most prevalent parasitic disorders globally, with thousands of individuals affected. The sickness is beginning yearly. This condition is related with around 280 million cases of diarrhea each year.

Objective: This study is to assess the levels of interleukin-38 (IL-38) and various hematological parameters in patients infected with *Giardia lamblia*.

Methods: The present investigation will span from September 2024 to March 2025. (45) Patients originally suspected of having *G. lamblia* were sent to the laboratory for further examination, where the parasite was confirmed, and their blood was subsequently collected. Moreover, blood samples from 45 healthy people were obtained to serve as a control group. The present investigation was conducted from September 2024 to March 2025.

Results: This study revealed that interleukin-38 levels were considerably increased in individuals with parasitic infections ($p < 0.05$). The examination of haematological characteristics in individuals afflicted with parasites revealed a significant reduction in red blood cell count and mean corpuscular haemoglobin concentration (MCHC). The levels of HB, PCV, and MCH were considerably greater in patients who were infected with *Giardia lamblia* than in controls.

Conclusion: *Giardia lamblia* infection is a significant public health concern associated with notable immunological and hematological changes. The findings indicate a substantial increase in interleukin-38 (IL-38) levels in infected patients.

Keywords: *Giardia*, Giardiasis, Interleukin 38, Diarrhea

Introduction

Giardia lamblia is a parasite that is prevalent in humans and other mammals. Giardiasis is transmitted via the fecal-oral route, either directly or indirectly via the consumption of infectious cysts in water or food that is polluted with feces.^{1,2} The life cycle of *Giardia lamblia* is additionally composed of

two separate stages based on its shape. These two stages represent the growth phase of the trophozoite phase, as well as the infective stage of the cyst.³ The symptoms of giardiasis are comprised of symptoms on the intestinal wall, such as diarrhea, abdominal pain, nausea, and a loss of weight that is not significant, as well as extraintestinal symptoms, such as fever, lymphadenopathy, urticaria,

maculopapular rash, polyarthritis, and pulmonary issues.⁴ In developing countries, the majority of the population (around 33%) suffers from this disease. In developed countries, the typical cause of giardiasis is travelers, who account for 2-5% of the population.⁵ The host's response to *G. lamblia* is complex, including a protective Th17 response, the recruitment of mast cells, and increased levels of IgA, nitrates, and mucus.^{6,7} The response to *G. lamblia* that is adaptive includes both immunological mechanisms that are cellular as well as humoral. CD4+ T cells, especially Th17 and Th1 cells, are integral to this response. Th17 cells facilitate the recruitment of neutrophils, they also induce the production of anti-inflammatory peptides in the intestinal wall through the production of IL-17. Th1 cells facilitate the production of IFN- γ and activate macrophages.⁸ Giardiasis is determined by analyzing the excrement of individuals infected with the disease. The infection was confirmed if parasites or cysts of *Giardia* were observed during the microscopic analysis of stool samples.⁹

Materials and Methods

Blood Samples Collection

After the disease was suspected of having the symptoms of *G. lamblia*, 45 blood samples were collected for analysis by microscopy. Microscopic analysis showed that they were parasitized by the parasite. 45 blood samples were obtained from individuals with typical blood. The samples came from males and females of different age groups (aged 20-45, children aged 4 months-10 years).¹⁰

Examination Stool Samples

Fecal samples were assessed by eye for color and consistency. They were then evaluated for the presence of *Giardia* trophozoites and cysts, as well as for the detection of other parasites in a optical microscope (Olympus CH series, Japan and Primo Star ZEISS microscope, Germany). A blind solution of salt (normal salt) was created by dissolving 8.5 g of pure sodium chloride (NaCl) in 1 liter of distilled water, the mixture was then observed using an optical microscope to determine the presence of trophozoites and cysts in *Giardia*.¹⁰

Human Interleukin 38 ELISA kit Assay Principle

The kit is composed of an antibody that is linked to an enzyme. The plate is previously covered with the antibody against IL-38 that is human. The antibody from the sample that is specific for IL-38 is incorporated, this antibody interacts with the antibody that is present in the wells. Next, a biotinylated antibody that recognizes the IL-38 in the sample is incorporated. This antibody interacts with the IL-38 in the instance. Next, Streptavidin-HRP is added and attaches to the antibody that is biotinylated against

IL-38. After incubation, the unbound streptavidin-HRP is excised in a washing procedure. The substrate solution is then added, and the color of the substrate is proportional to the volume of human IL-38. The response is inhibited by adding a solution with a high pH, the absorbance is then measured at 450nm

Determination Of blood parameters

Blood samples in EDTA containers were thoroughly mixed in a blender, then 0.2 ml of blood was taken from each sample using a detector (Sysmex, XP 300 Japan) that is specifically designed to count the number of red blood cells. We obtained 45 blood samples from patients at primary health centers that had no previous history of *Giardia*, these patients appeared to have a healthy physical profile. Data on the age, gender, and residence of each child at the current location as well as the results of stool analyses, the presence of *Giardia*, and associated parasites were recorded in a unique manner.¹¹

Hematology analyzer

Hematology analyzers are devices that count the amount of cells in the blood, classify white blood cells, and assess the hemoglobin content of the cells. Two sorts of detecting technologies are prevalent: electrical and optical. Resistance and radio frequency conductivity are both instances of electrical principles, whereas laser scattering and spectrophotometry are both examples of optical principles.

Principles of blood cell counting

- **Resistance principle:** Blood cells are unable to carry out blood flow. If a suspension of blood cells is traversed via a tiny aperture with a continuous flow on both sides, the resistance of the region will grow owing to the poor conductivity of the cells compared to the electrolyte solution. This causes a voltage to vary and resulting in a pulsed voltage through. To gather cell count data, the signal must be amplified, filtered, and shaped before being transferred to the system for measurement. The cell's volume is also approximated based on the pulse's intensity, this creates a graphical depiction of the distribution of cell volumes.
- **The fundamentals of flow cytometry and the light-scattering characteristics of blood cells:** When the sheath fluid flows across the zone of detection, it is irradiated by the laser, this causes the cells to separate, the fluid is then dynamically aggregated. The light that is dispersed by the cells is identified by the photodetector, this creates pulses that are proportionate to the size of the cells that are impacted. The count of cells is the number of pulses.

Principles of white blood cell classification

Leukocytes are divided by their velocity into three classes: low, medium, and high. When differing quantities of white blood cells flow through a short area, the ensuing pulse will have a variable magnitude depending on the idea of resistance. The instrument recognises leukocytes by the size of their waves. After introducing a chemical that hemolyzes the red blood cells to the sample, the device calculates the number of electrical waves that are more than 35fl and are recognised by the leukocyte detector as the leukocyte count. The volume of the pulse is governed by the quantity of leukocytes in the blood: small cell clusters (35-90 fl), medium cell clusters (91-160 fl) and big cell clusters (161-450 fl).

Its qualities include rapid performance and user-friendliness, these traits are capable of considerably boosting efficiency. The device may be applied to identify the degree to which leukocytes are routinely examined in standard exams. Samples that are positive should be analysed with a microscope. For persons who are considered to have blood disorders, blood films and microscopic investigations should be undertaken even if the instrument seems to be normal.

Five-point categorisation principle: From the three-point classification of the resistance technique to the exploitation of several technologies (laser, radio frequency and chemical staining) to view single cells in detail, and completely evaluate the experimental data to produce the five-point classification of white blood cells. This approach is capable of classifying a huge number of cells with a high degree of specificity, it's now the most prevalent way of blood cell classification.

Principles of hemoglobin measurement

In triple or quintuple blood analyzers, hemoglobin is measured via spectrophotometry. When coupled with a chemical that produces hemolysis, red blood cells are destroyed and release their hemoglobin. It reacts with certain components of the hemolytic material to generate a stable hemoglobin derivative that has a specified hue in a specific range of light wavelengths (530-550 nm). Since the change in absorption is directly connected to the concentration of hemoglobin in the blood, the concentration of hemoglobin may be calculated.

Results

The study included 90 blood participants, of whom 45 were diagnosed with giardia lamblia. The prevalence of Giardia was found to be 50% with a higher incidence in children. The data suggest that the factors such as study the effect of some factor like age, sex, months, socioeconomic status, hygiene practices and exposure to contaminated water on infection rate of parasite.

Immunological parameter

The current study's findings revealed an increase in the level of interleukin-38. It was 49.39 higher than those of the controls group 39.59 as shown in (Table 1).

Hematological parameters

The results of study showed a significant decrease in RBCs, MCHC and there was a significant increase in the HB, PCV, MCV in patients infected with giardia lamblia in comparison to control group as shown in (table 2).

Table 1. Distribution of Giardiasis according to gender

Gender	No. of examined sample	No. of Positive sample	Infection rate (%)
Male	27	33	35.48 %
Female	18	17	26.56%
Total	45	50	31.847%

Table 2. The comparison of the concentrations of interleukin (IL-38) between infected patients and healthy individuals

Groups	IL-38 (Pg/ml)		P. value
	No.	Mean \pm S.E.	
Control	45	39.59 \pm 2.54 A	0.08 Sig.
Patients	45	49.39 \pm 4.11 B	
LSD		3.77	

Multiple pairwise comparisons./ least significant difference (LSD) post hoc test
SD; Standard Deviation of mean, sig: significant. P. value

Table 3. The comparison of blood properties between patients infected with Giardia lamblia and controls

parameter	Patients N = 45	Control healthy N= 45	P.value
RBCs (X106/mm3)	4.8065	4.9255	0.71
PCV (%)	40.5351	35.75	0.03
Hb g/dl	13.42	12.89	0.091
MCV (mm3)	84.55	79.81	0.011
MCHC (g/dl of RBCs)	32.965	36.045	0.047

Discussion

Table (1) demonstrated that adult and infant blood samples with a Giardia infection parasite lamblia, which numbered 45 samples, and 45 samples of healthy people missing the parasite, were subjected to a study of cytokines (IL-37) and a blood parameters study. The outcomes showed a higher rate of infection in males than females (35%), (26%) respectively. Blood samples from children and adults infected with the parasite Giardia lamblia (45 samples were included) as well as 45 samples from healthy persons without the parasite were examined for cytokines (interleukin 38) and other blood parameters. The outcomes of this investigation demonstrated a rise in interleukin-38 levels, as is displayed in Table 2. Giardia lamblia induces the production of various chemokines and pro-inflammatory cytokines, the latter of which is constituted of 38 distinct cytokines obtained from human epithelial and stromal cells of different types. The release of IL-38 is of essential relevance in lowering the production of inflammatory cytokines induced by the infection of Giardia. Interleukin 38 is a cytokine that combats inflammation, it reduces the production of other cytokines such Th1 and Th17, these are implicated in inflammation.¹² Interleukin (IL)-38 is a newly found anti-inflammatory cytokine that is part of the IL-1 β family and has a major influence on numerous autoimmune disorders. The IL-38 is generated by numerous distinct cells of the immune system, including B cells. Since IL-38 is formed from a B cell, the absence of this molecule is connected with increased inflammation in the general circulation, metabolic and cardiovascular illnesses.¹³ The normal expression of IL-38 occurs in the human heart, thymus and other organs, but not in the tonsil cells. Additionally, IL-38 binds to receptors via the nuclear factor κ B (NF- κ B), the activated protein 1 (AP-1) and the c-Jun N-terminal kinase (JNK) pathways, this induces the production of inflammatory cytokines.¹⁴ These statistics show that IL-38 may be connected with autoimmune disorders. Additionally, IL-38 is also capable of influencing the process of autoimmune illnesses by modulating the ratio of anti-inflammatory to pro-inflammatory chemicals.

The IL-38 is generated by epithelial cells, monocytes, macrophages and immunological cells. The precursor of IL-38 is cleaved at the N-terminus, this leaves the active form of the molecule reliant on the receptor and other downstream components. The reactions that mates make are crucial in human physiology and pathology, and the cytokine IL-38 is regarded helpful in sick situations.¹⁵

The findings indicated that the content of IL-38 in the serum of patients infected with G. lamblia was substantially lower than that of controls. This may be caused by a decreased cellular immune response, which leads to a reduction in the synthesis of cytokines by the immune cells engaged in host defense, these cells have a production of harmful cytokines. The findings revealed that the concentration of interleukin (IL-38) was larger in patients than in controls (Table 2). The rise in IL-38 was considerable (P<0.05). The conclusions of this analysis accord with previous studies (16,17). Key cellular products include cytokines, which are critical to the course and outcome of infection, including varieties that are pro-inflammatory and anti-inflammatory, the latter of which causes varied reactions to immunogens throughout various phases of infection. The method in which cytokines react to the same stimuli is dependant on the disease's kind, aetiology, and persistence. The infection of Giardia intestinalis causes the breakdown of the brush border of the intestinal cells, which leads in a shortening of the microvilli and a modification of the epithelial barrier. The cause of acute diarrhea in giardiasis is attributed to increased apoptosis in the enterocytes, impaired barrier function in the intestinal wall, activated lymphocytes, CD8+ cells that shortened the brush border of the microvilli (with or without villous atrophy), disaccharidases that are lacking, intestinal malabsorption, anion over-secretion, and increased intestinal travel rate.¹⁸

Table (3) demonstrates the average values of RBCs, PCV, Hb, MCV, and MCHC in individuals infected with Giardia lamblia in comparison to controls. The findings revealed that the amount of RBCs and MCHC was dramatically reduced, while the quantity of PCV, MCV, and Hb was raised in patients infected with Giardia lamblia. The reduction in

various blood parameters relating to the digestive system is ascribed to the colonization of the system by intestinal parasites, this is especially true of the duodenum and jejunum. These iron absorption sites contribute to the malabsorption of carbohydrates, lipids, vitamins (such as D and B12), folic acid, iron, and zinc. Iron malabsorption produces iron shortage, which is followed by iron entering into hemoglobin, which leads in anemia.¹⁹ It also creates proteins that examine the tissues and cells of the host and devour red blood cells.²⁰ Additionally, parasitic infection can cause the intestinal wall to become necrotic, this causes the intestine to have a lack of important substances and to suffer from tissue damage; parasites are closely associated with the development of anemia because they cause the intestine to lack essential substances and have a lack of tissue.

Conclusion

Based on the results of this study we concluded that *Giardia lamblia* infection is a significant public health problem and associated with significant immunological and hematological changes long term complications like vitamins and minerals deficiencies and need treatment. Contamination of municipal water supplies should be considered as potential risk factor, Housing improvement with appropriate sewage system and educational measures to improve personal hygiene are an important factors to reduce prevalence of infection.

Author Contributions

All authors had seen and approved the submission of the manuscript with full responsibility, and this research had not been published or under consideration by any other journal.

Sources of Funding: None

Conflict of Interest: None

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