Medical Science Journal for Advance Research

Vol. 4 , No. 2 , July 2023
e-ISSN: 2774-4892

Effect Of *Ascaris Lumbricoides* On Some Hematological Parameters And The Concentration Levels Of Interleukin-10 And Interleukin-22

Eman Hasani AL-Salami[1], Nadia Habeeb Sarhan[2], Dina A. A. Al-Roubaey[3]

[1]*Department of Microbiology, College of Medicine, Kufa University, Iraq*
[2]*Department of Basic Science, Faculty of nursing, University of Kufa, Iraq*
[3]*Department of Basic Science, Faculty of nursing, University of Kufa, Iraq*
E-mail: emanh.alsalami@uokufa.edu.iq
Email: nadiah.alshaibani@uokufa.edu.iq
Email: dinaa.alroubaey@uokufa.edu.iq

ABSTRACT

Background: *The parasite Ascaris lumbricoides modifies the host's immunological response. This study aims to determine the interleukin-10 (IL-10) and interleukin-22 (IL-22) serum levels in specific patients and their associations with the severity of infection. The present study also requires to study a few blood markers in Ascaris lumbricoides-infected patients.Patients were referred to the laboratory for testing from Al Sader General Hospital in AL-Najaf Province, Al Furat General Hospital, AL-Zahra maternity and pediatrics, and AL-Hakeem Hospital. Samples Collected 32 samples were taken from patients and 20 healthy individuals who attended clinics at the AL-Sadder Teaching Hospital and AL-Hakeem Hospital in the province of AL-Najaf between July 2021 and June 2022. Male and female patients between the ages of 10 and 50 had their stool samples taken and put in clean, wide-mouth specimen bottles. The same patients' veins were also punctured to get five milliliters of blood, four milliliters of which were then deposited in specimen tubes. After 30 minutes at room temperature, stool samples were examined using the Formol-ether concentration and Kato-Katz thick smear procedures. According to the findings, ascariasis patients had significantly higher serum levels of interlukin-10 and interlukin-22 than the healthy group (P0.05). The findings showed that the iron level concentration in ascariasis patients decreased significantly (P0.05), and that the Hb, PCV, and RBC count also decreased significantly (P0.05).*

Keywords : Ascariasis , Interleukin 10&22 , Hematological Parameters

Article Information

Received: June 13, 2023; Revised: June 26, 2023; Online: July 31, 2023

INTRODUCTION

Human pathogen Ascaris lumbricoides is a big roundworm with a widespread range. In tropical nations around the world, gastrointestinal parasite infection has been highlighted as a severe public health hazard (1,2,3). An estimated 1.5 billion infections are brought on by A. lumbricoides alone. These infections are linked to significant morbidity(4). According to research, children are more likely than adults to contract soil-transmitted helminth (STH) parasites in environments with low hygiene and sanitation. (6) Through oral ingestion of infectious eggs, A. lumbricoides infects the mammalian host. (7,8) Ascariasis is prevalent in regions where feces are dumped carelessly into open latrines. (9,10).

As a result, food and water contamination are very common (11). Malaria and hookworm infections are two parasitic illnesses that are frequently associated with anemia (12). Ascariasis is more prevalent in places with poor hygiene and is associated to malnutrition, iron deficiency anemia, and problems with cognition and growth. (13)

https://doi.org/10.46966/msjar.v4i2.131

Published by: READ INSTITUTE

One of the most frequent reasons for a patient's referral to a gastroenterologist is iron deficiency anemia (IDA). Reduced nutritional intake, reduced absorption, and blood loss are common causes of IDA, albeit low absorption is typically the least frequently mentioned among these factors. In countries with abundant resources, malabsorption is a very uncommon cause of iron deficiency, and only some of those few cases can be linked to parasite infections. (1). Ascaris infection is one such instance where an infection is being demonstrated to affect the nutritional status of the host. In locations with plentiful resources, malnutrition raises concerns about physiological conditions that hinder nutritional absorption or, in rare instances, opportunistic infections that feed off the nutrients the patient consumes. Although there are 819 million instances of Ascaris worldwide, it is uncommon in resource-rich nations and more frequently associated with areas of poverty and hunger. Ascaris is also particularly surprising if one does not go to endemic places (12,13). IL-22 is an indicator of the cytokine family IL-10, which also produces IL-19, IL-20, IL-24, and IL-26 (15). Regulatory T cells and even TH 1 cells have been linked to immune control and inflammation (14). A heterodimeric receptor complex consisting of the IL-22R1 and IL-10R2 subunits is how IL-22 interacts. (16). IL-22R1 is only present on the surfaces of non-hematopoietic cells, such as epithelial cells, hepatocytes, and keratinocytes, whereas IL-10R2 is present on nearly all cell types (17). IL-22 can particularly target innate cell populations in organs such the gastrointestinal tract, liver, skin, kidneys, and lungs because to the restricted expression of IL-22R1 on non-hematopoietic cells (18). In light of the fact that tumor necrosis factor alpha (TNF-) is an early mediator involved in the production of IL-6, chemokines, and CRP as well as the recruitment of leucocytes during inflammatory reactions, we conducted this study to illustrate the immunological milieu of A. lumbricoides (19,20), and is a member of cytokines that stimulate the acute phase reaction; its main function is the regulation of immune cells; it can also cause apoptotic cell death to cause inflammation (21); and Th2 cytokines such as IL-10 and IL-22; to determine whether there is any relationship between these cytokines and various well-known clinical symptoms of ascariasis, such as cramps in the abdomen, diarrhea, and atopy. It was also noted how these cytokines' concentrations and the age, illness severity, and age all relate to one another

The aim of the study :

1- The aim of the study is to determine the interleukin-10 (IL-10) and interleukin-22 (IL-22) serum levels in some patients and how they relate to the severity of diseases.

2-Study some blood parameters of patients infected with the Ascaris lumbricoides

MATERIALS AND METHODS

Samples Collected 32 samples were taken from patients and 20 healthy individuals who attended clinics at Al Furat General Hospital in the AL-Najaf Province, Al Sader General Hospital, AL-Zahra maternity and pediatrics, and AL-Hakeem Hospital between July 2021 and June 2022. Male and female patients between the ages of 10 and 50 had their stool samples taken and put in clean, wide-mouth specimen bottles. Five milliliters of blood were also drawn from the same patients via vein puncture, four milliliters were placed in specimen tubes, and 30 minutes were spent at room temperature. The serum was then separated from the samples using a centrifuge (Backman/counter, Germany) at 3000 rpm for 5 minutes, and the serum was then collected in additional sterile tubes. Each serum sample was then split into two parts to be placed in a

deep freezer at -20Co until it was time to determine the levels of iron and ferritin. To evaluate the haematological parameters Hb, PCV, and RBCS, the final one milliliter of blood was collected into a tube containing anticoagulated EDTA (Abott/Jordan.

2-Iron Serum (Colorimetric Test) To determine serum iron levels using RANDOX reagents, code HB012, (RANDOX Kit, U.K.), the Cypress Diagnostics Biochemistry Analyzer using the colorimetric test method.

3-Parasitological stool examination

On alternate days, three stool samples preserved in 10% formalin were used to investigate helminth eggs using the spontaneous sedimentation method (Hofmann). The Kato-Katz method was applied to determine the number of eggs per gram of feces (OPG).

4- The examination of IL-10,22 in supernatant fluids

Using a sandwich ELISA carried out in accordance with the manufacturer's instructions, the levels of IL-10 and IL-22 were assessed. 19.5 pg/mL was the assay's detection limit, and data below this threshold were given the detection limit value.

STATISTICAL ANALYSIS

The information was entered into the Microsoft Excel program. The prevalence of intestinal parasite infections and risk variables were examined and analyzed using the Chi-square (2) test in order to identify correlations between the two in both infected and uninfected children. The threshold for statistical significance was established at 0.05

RESULT

The Relationship Between Ascariasis Patients' IL-10 and the Control Group

Figure 1 shows a comparison of ascariasis patients and a healthy group, with a significant increase (P0.05) in serum interlukin-10 (IL-10) concentration in ascariasis patients (355.23 0.221 vs. 64.54.21 Ug/dl in the healthy group.

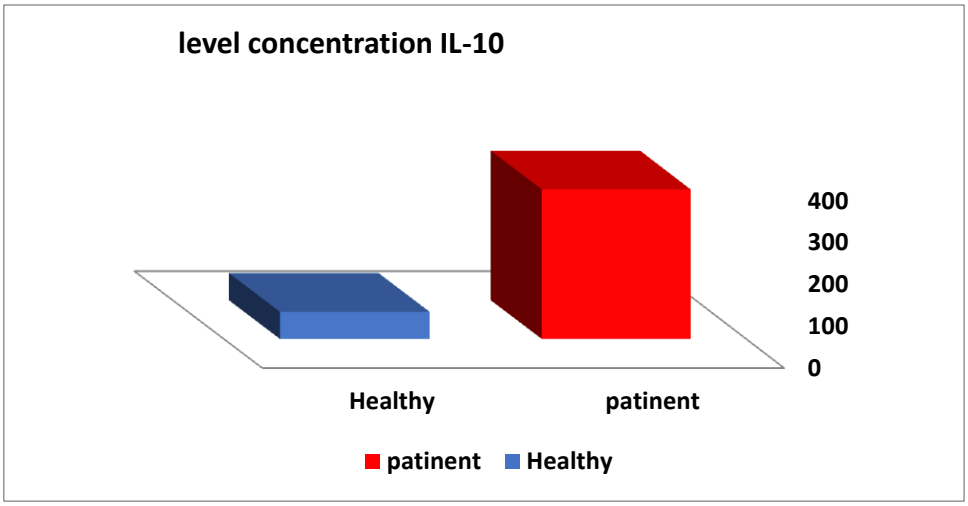

Fig.1. comparison between level concentration of IL-10 of ascariasis patients and Healthy group

Interlukin-22 (IL-22) levels in Ascariasis patients and healthy controls

Figure 2 shows a comparison of ascariasis patients and a healthy group, with a significant increase (P0.05) in serum interlukin-22 concentration in ascariasis patients (562.564 2.791 vs. 100.88 0.923 in the healthy group.(

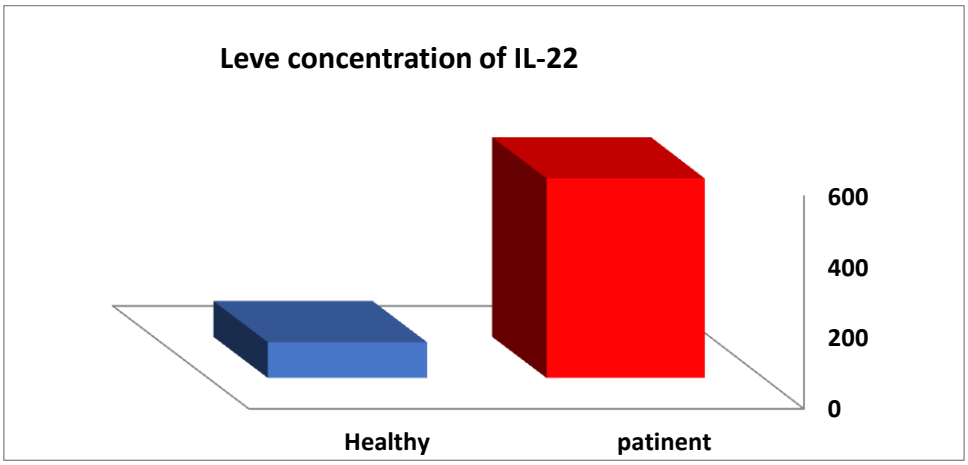

Fig.1. comparison between level concentration of IL-22 of ascariasis patients and Healthy group

comparison of the iron levels in Ascariasis patients and the healthy group

Ascariasis patients had significantly lower levels of iron than the healthy group (22.793 0.322 Ug/dL versus 266.321 4.113 Ug/dL, P0.05) in the comparison between the two groups of patients.

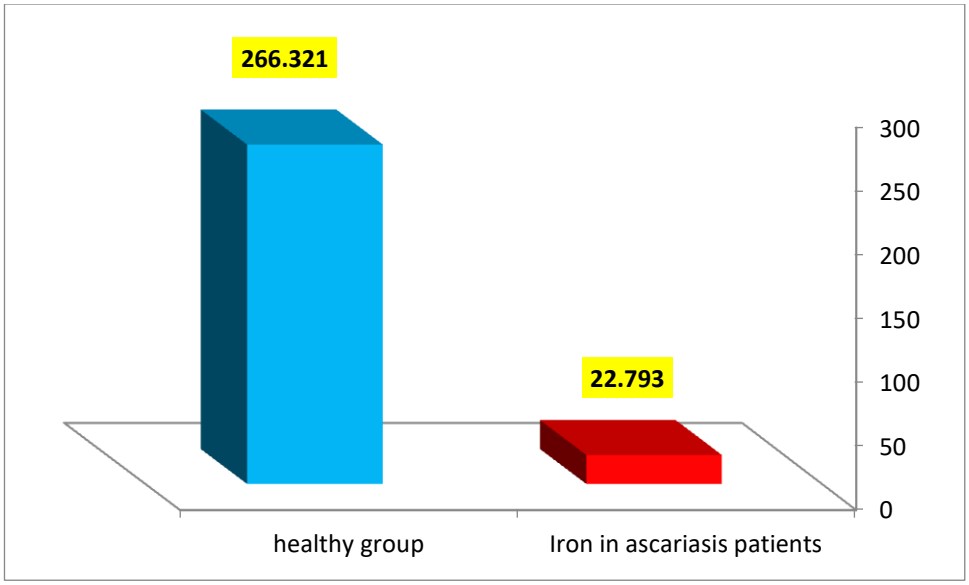

Figure (3) comparison Levels of Iron in Ascariasis Patients and the Control Group

Hematological parameters

When compared to individuals in the healthy group, ascariasis patients' Hb, PCV, and RBC counts significantly (P 0.05) decreased, with ascariasis patients' values being 9. 548 0.581 gm/dL, 13.548 0.160 gm/dL, 29.871 (%), and 3.755 106 respectively, compared to participants in the healthy group .

Table1: Comparing laboratory test results between ascariasis patients and the healthy group

	Patients	Controls	*P*-value
Gender(male/female)	32	20	0.730
WBC ($\times10^9$/L)	6.49 (4.78, 8.86)	6.19 (5.71, 7.48)	0.947
RBC ($\times10^{12}$/L)	3.67 ± 0.90	4.63 ± 0.39	0.000
HGB (g/L)	102.44 ± 22.95	145.48 ± 8.51	0.000
MCV (fL)	86.98 (73.32, 91.88)	91.04 (88.51, 91.22)	0.000
MCH (pg)	27.36 (21.96, 31.40)	29.81 (29.02, 30.72)	0.000
MCHC (g/L)	335.05 (322.50, 322.15)	322.70 (329.00, 325.80)	0.000
PLT ($\times10^9$/L)	265.30 (201.62, 310.20)	222.30 (208.70, 270.90)	0.002
NEU ($\times10^9$/L)	3.74 (2.69, 5.43)	3.48 (2.83, 4.23)	0.327
LYM ($\times10^9$/L)	1.26 (1.07, 1.97)	2.25 (1.96, 2.50)	0.000
EOS ($\times10^9$/L)	0.45 (0.17, 0.62)	0.12 (0.08, 0.30)	0.000

DISCUSSION

According to the current study, patients with A. lumbricoides infections had significantly lower RBC counts, Hb concentrations, and PCV concentrations than patients in the control group. This outcome might have been brought on by the A. lumbricoides worm hemolyzing RBCs, which might have led to a reduction in the amount of RBCs. A drop in the concentration of Hb might have additionally been brought about by the hemolysis of RBCs. (22). Anemia and parasite infestation are related by a pathogenophysiologic process. (23,24,25) By lowering iron absorption from the gut, directly sucking blood , and interfering with iron metabolism both directly and indirectly, the helminthic infection leads to anemia shortage. (26,27,28,29) ascaris affect anemia by destroying the intestine's mucosal structure, which affects how well micronutrients like iron are absorbed. These methods all impact the nutritional state of the hosts , which consequently changes their immune system. (30,31). Ascaris infection is one such instance where an infection has been shown to affect the nutritional state of the host. However, especially in middle-aged and older persons, IDA is typically thought to be brought on by blood loss until otherwise demonstrated. Gynecologic issues are another typical IDA etiology in females. A physiological condition that interferes with nutritional absorption or, in rare instances, an

opportunistic infection that feeds off the nutrients the patient consumes can be causes of malnutrition in resource-rich countries. Even though there are 819 million cases of Ascaris worldwide, it is a disease that is unusual in resource-rich nations and more frequently occurs in locations where there is malnutrition and low socioeconomic position. It is also particularly unexpected when there hasn't been travel to areas where it is endemic (32,33).

A. lumbricoides infection in humans has been associated with eosinophilia, fever, dyspnea, and cough, sometimes known as Loeffler's syndrome, during the larval migratory phase through the lung and abdominal pain during the intestinal phase. Ascariasis is distinguished by eosinophilia, mast cell hyperplasia, and elevated levels of immunoglobin E (IgE) in the blood. (34,35) Our findings are consistent with IL-10 and IL-22, with the family having a significant role in helminth infection immunity. IL-10 and IL-22 both stimulate eosinophils. function as a link between the parasite and a neighboring co-activated cell on the parasite's surface. The inhibition of IL-10 upstream in the immunological disease produced by A. lumbricoides infection during the lung migration phase has a substantial influence on IL-22 levels in the screened patients(36). In our area, A. lumbricoides infection is associated with significantly elevated levels of IL-10 and IL-22. (37,38).

CONCLUSION

In many parts of the world , where poor sanitation makes infection and reinfection more likely , parasitic helminth infestation is a widespread condition affecting billions of people, In individuals with ascariasis, intestinal parasite infection had a deleterious impact on hematological profiles and was a reliable indicator of anemia . In patients infected with the Ascaris parasite, intestinal parasitic infection resulted in a considerable reduction in the levels of HGB, HCT, MCV, MCH, and MCHC. Deworming can be an effective method of getting rid of these worms in ascariasis patients in order to minimize anemia.

REFERENCES

1. Oliveira F.M.S., et al. Comorbidity associated to ascaris suum infection during pulmonary fibrosis exacerbates chronic lung and liver inflammation and dysfunction but not affect the parasite cycle in mice. *PLoS Neglected Trop. Dis.* 2019;13(11):1–30. doi: 10.1371/journal.pntd.0007896. [

2. Degarege, A., Animut, A., Medhin, G., Legesse, M. & Erko, B. The association between multiple intestinal helminth infections and blood group, anaemia and nutritional status in human populations from Dore Bafeno, southern Ethiopia. *J. Helminthol.* 88, 152–159 (2014).

3. Abdullah, D. A., Mahmood, M. A., AL hatemi, M. D. (2011). The levels of cytokines IL-4, IL-10, IL12, P40, IFN-ɣ during acute Toxoplasmosis. *Journal of the Faculty of Medicine Baghdad*, 53(4): 1-5.

4. Varyani F, Fleming JO, Maizels RM. Helminths in the gastrointestinal tract as modulators of immunity and pathology. Am J Physiol - Gastrointest Liver Physiol [Internet]. 2017;312(6):G537-49.

5. Alelign, T., Degarege, A. & Erko, B. Soil-transmitted helminth infections and associated risk factors among Schoolchildren in Durbete Town, Northwestern Ethiopia. *J. Parasitol. Res.* 2015, 641602 (2015).

6. Srivastava, A., Mahmood, S. E., Srivastava, P. M., Shrotriya, V. P. & Kumar, B. Nutritional status of school-age children: a scenario of urban slums in India. *Arch. Public Health* 70, 8 (2012).

7. Zheng, P.-P., Wang, B.-Y., Wang, F., Ao, R. & Wang, Y. Esophageal space-occupying lesion caused by *Ascaris lumbricoides*. *World J. Gastroenterol.* 18, 1552–1554 (2012).

8. Shang, Y. *et al.* Stunting and soil-transmitted-helminth infections among school-age pupils in rural areas of southern China. *Parasites Vectors* 3, 97 (2010).

9. Damen JG, Luka J, Biwan EI, Lugos M. Prevalence of intestinal parasites among pupils in rural North Eastern Nigeria. Niger Med J. 2011;52:4–6.

10. Kabatereine, N. B. *et al.* Short communication: Soil-transmitted helminthiasis in Uganda: epidemiology and cost of control. *Trop Med Int Health* 10, 1187–1189 (2005).

11. Tarafder, M. R. *et al.* Estimating the sensitivity and specificity of Kato-Katz stool examination technique for detection of hookworms, *Ascaris lumbricoides* and T*richuris trichiura* infections in humans in the absence of a 'gold standard'. *Int. J. Parasitol.* 40, 399–404 (2010).

12. Eraky, M. A., Rashed, S. M., Nasr, M.E.-S., El-Hamshary, A. M. S. & Salah El-Ghannam, A. Parasitic contamination of commonly consumed fresh leafy vegetables in Benha, Egypt. *J. Parasitol. Res.* 2014, 613960 (2014).

13. Lamberton, P. H. L. & Jourdan, P. M. Human ascariasis: diagnostics update. *Curr. Trop. Med. Rep.* 2, 189–200 (2015).

14. Al-Hatemy, M. D. B., Mohsin, M. I., Al-Roubaey, D. A. A. (2022). The correlation between Interleukin-6 and D-dimer, Serum ferritin, CRP in COVID-19 patients in Al-Najaf province. *Kufa Journal for Nursing Sciences*, 12 (1): 163-173.

15. Hansen, E. P., Fromm, B., Andersen, S. D., Marcilla, A., Andersen, K. L., Borup, A., et al. (2019). Exploration of extracellular vesicles from ascaris suum provides evidence of parasite-host cross talk. *J. Extracell. Vesicles* 8, 1578116–1578116. doi: 10.1080/20013078.2019.1578116

16. Hepworth, M. R., Daniłowicz-Luebert, E., Rausch, S., Metz, M., Klotz, C., Maurer, M., et al. (2012). Mast cells orchestrate type 2 immunity to helminths through regulation of tissue-derived cytokines. *Proc. Natl. Acad. Sci. U.S.A.* 109, 6644–6649. doi: 10.1073/pnas.1112268109

17. Reina Ortiz M, Schreiber F, Benitez S, et al. Effects of chronic ascariasis and trichuriasis on cytokine production and gene expression in human blood: a cross-sectional study. PLoS Negl Trop Dis. 2011;5(6): e1157.

18. Allen JE, Maizels RM. Th1-Th2: reliable paradigm or dangerous dogma? Immunol Today. 1997;18:387–392.

19. Layedh, N. H., Al-Rubbaey, Y. A. , Fahad, A. H., Al-Roubaey, D. A. A. (2021). Salivary IL-6 and TNF- α in patients with periodontitis. *Annals of Tropical Medicine & Public Healt*, 317-325. DOI: http://doi.org/10.36295/ASRO.2021.24546.

20. AL-Roubaey, D. A. A. (2017). The correlation between oral warfarin intake and two proinflammatory cytokines (IL-6 and TNF-α) and their effects on atherosclerosis in deep venous thrombosis disease. Al-Kufa University Journal for Biology, Print ISSN: 2073-8854 & Online ISSN: 2311-6544.

21. AL-Roubaey, D. A. A. (2013). The correlation between anti-neutrophil cytoplasmic antibody and two cytokines (IL-18, TNF-α) in rapidly progressive glomerulonephritis disease. *AL-Taqani*, 26(3): 73-82.

22.Sarhan, N.H.,Waheed, Z.A., Zayed, S.S., Rasool, M.B., Rasool, B.R.(2022).Studing of some biochemical parameters for thalassemia patients in AL-Najaf province.International Journal of Health Science,6(S4), 1623-1629.

23. Jourdan, P. M., Lamberton, P. H. L., Fenwick, A., and Addiss, D. G. (2018). Soil-transmitted helminth infections. *Lancet* 391, 252–265.

24. Khalid, K. E., Nascimento, M. S. L., Sacramento, L. A., Costa, D. L., Lima-Júnior, D. S., Carregaro, V., et al. (2017). T1/ST2

deficient mice display protection against leishmania infantum experimental infection. *Acta Trop.* 172, 1–6. doi: 10.1016/j.actatropica.2017.04.011

25.AL-Salami, E.H., AL-Asaedi, M.R.M., Sarhan, N.H., Yasir, S.J.(2021). Interleukin-8 Concentration Correlation with the Size of Hydatid Cyst in Patients with Hydatidosis and Hepatitis C mixed infection(Case Control Study). Indian Journal of Forensic Medicine & Toxicology.V(15).Issue(2).p.1823-1828.

26. Karagiannis-Voules DA, Odermatt P, Biedermann P, et al. Geostatistical modelling of soil-transmitted helminth infection in Cambodia: do socioeconomic factors improve predictions? Acta Trop. 2015;141: 204–212.

27. Copper PJ, Chico ME, Sandoval C, et al. Human infection with Ascaris lumbricoides is associated with a polarized cytokine response. J Infect Dis. 2000;182(4):1207–1213.

28. Mathison, B. A., and Pritt, B. S. (2018). A systematic overview of zoonotic helminth infections in North America. *Lab. Med.* 49, e61–e93. doi: 10.1093/labmed/lmy029

29. AL-Salami, E.H.,Albu-Saabar, S.R., Sarhan, N.H .(2022).Study of Some haematological and immunological parameters in patients infected with Giardia Lambliain Najaf Governorate.International Journal of Health Science,6(S4), 6020-6029.

30. Reynolds LA, Finlay BB, Maizels RM. Cohabitation in the Intestine: Interactions among Helminth Parasites, Bacterial Microbiota, and Host Immunity. J Immunol [Internet]. 2015;195(9):4059-66.

31. Wright VJ, Ame SM, Haji HS, et al. Early exposure of infants to GI nematodes induces Th2 dominant immune responses which are unaffected by periodic anthelminthic treatment. PLoS Negl Trop Dis. 2009; 3(5):e433.

32. Goddey NOP, Osagie ID, Maliki A. Serum cytokines profiles in Nigerian children with Ascaris lumbricoides infection. Asian Pac J Trop Med. 2010;3:288–291.

33. Souza V, Medeiros D, Sales I, et al. Ascaris lumbricoides infection in urban schoolchildren: specific IgE and IL-10 production. Allergol Immunopathol (Madr). 2014;42(3):206–211.

34. Gill GV, Beeching NJ. Tropical Medicine. 4th ed. Oxford: Blackwell Publishing Company; 2007:352. 27. Kouro T, Takatsu K. IL-5 and eosinophil-mediated inflammation: from discovery to therapy. Int Immunol. 2009;21(12):1303–1309.

35. Al-Qenaei, A. (2008): The role of iron in rheumatoid arthritis. Ph. Thesis Department of Pharmacy and pharmacology, University of Bath, Syria.pp:160.

36.Rehman, H. (2008): Hemolytic Anemia following Mycoplasma infection, The Internet Journal of Hematology. 4(1):132-139.

37.Harhay, MO.; Horton, J. andOlliaro, PL.(2010). "Epidemiology and control of human gastrointestinal parasites in children". Expert Review of Anti-infective Therapy 8 (2): 219–34.doi:10.1586/eri.09.119. PMC 2851163. PMID 20109051.

38.Murray, Patrick R.; Rosenthal, Ken S.; Pfaller, Michael A. Medical Microbiology, Fifth Edition. United States: Elsevier Mosby, 2005.

