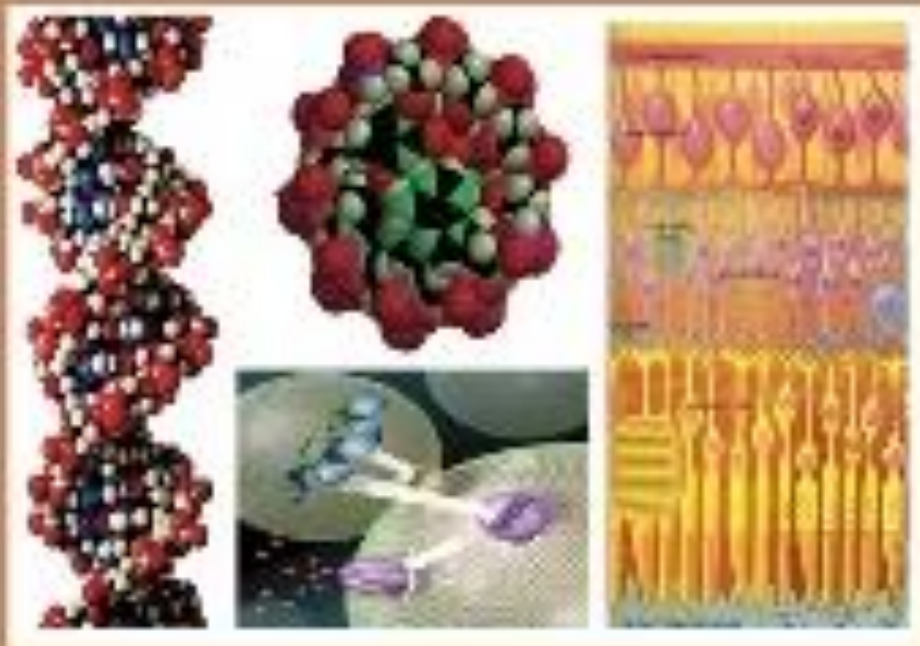




C

EGYPTIAN ACADEMIC JOURNAL OF
BIOLOGICAL SCIENCES

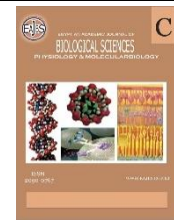
PHYSIOLOGY & MOLECULAR BIOLOGY



ISSN
2090-0767

WWW.EAJBS.ORG.EG

Vol. 15 No. 2 (2023)



**Quantitative Assessment of Serum Specific IgE in the Diagnosis of
*Echinococcus granulosus***

Fatima S. Kadhim¹ and Rasha A. Noori²

Department of Laboratory Investigation, Faculty of Science, University of Kufa, Iraq.

*E-mail: fatimasafa1305@gmail.com ; rashaa.altufaili@uokofa.edu.iq

ARTICLE INFO

Article History

Received:24/9/2023

Accepted:8/11/2023

Available:13/11/2023

Keywords:

Cystic
echinococcosis,
*Echinococcus
granulosus*, IgE,
Immunology.

ABSTRACT

A total of 50 serum samples were collected from patients with *Echinococcus granulosus* and 38 serum samples from healthy humans were taken from the Al-Furat AL-Awsat Hospital, Maternity and Children Teaching Hospital and Al-Diwaniyah Teaching Hospital in Al-Diwaniyah and from Specialized Hospital for Gastroenterology and Hepatology in AL-Najaf for the period from November 2022 to April 2023. The test was performed on 5 ml of venous blood, which was collected from each sample for an Enzyme-linked immunosorbent assay (ELISA) test to detect Echinococcosis IgE. The Distribution of Hydatid Cysts according to the site of the infection was the high rate of infection in the liver compared with other organs. The concentration level of IgE was increased in patients compared with the control, with a significant difference at ($P \leq 0.05$) while the level of IgE was decreased after the surgery compared with the before the surgery in patients with a significant difference at ($P \leq 0.05$). The levels of IgE concentrations in patients according to the location of the cyst were the highest level in the liver.

INTRODUCTION

Hydatid disease, a zoonotic parasitic infection, has a global impact on livestock, wildlife, and human populations (Deplazes *et al.*, 2017). Disease in both animal and human intermediate hosts is attributed to becoming infected with the larval stage of *Echinococcus*, including multiple species (Wilson *et al.*, 2019). Accidental getting of intermediate host status by humans can occur through fecal-oral transmission, which is facilitated by close proximity to definitive hosts, including domestic and wild canids. An additional mode of infection involves the consumption of embryonated eggs of the parasites, which can be present in food, water, soil, and fomites, as documented by Tamarozzi *et al.* (2020). Definitive hosts acquire infection through the ingestion of viable hydatid cysts present in the internal organs of intermediate hosts, which encompasses a wide range of mammalian species (Thompson, 2017; Mahdi Fakhar, *et al.*, 2021). The primary organs involved in this process are the liver, lungs, spleen, and heart, with occasional involvement of other organs. Due to the involvement of the liver and lungs of intermediary hosts in the life cycle of its larvae, they do not present significant issues during their early stages. Once they reach maturity, however, these organisms induce jaundice when they inhabit the liver. Hepatic damage is associated with the development of digestive disorders, whereas the presence of cysts in other organ systems is known to contribute to the occurrence of disorders specific to those organs (Toparlak *et al.*, 2000; Çinar *et al.*, 2018).

According to Eckert and Deplazes (2004), Budke et al. (2006), and Ahmadi and Bodi (2011), the incubation period of hydatid disease is characterised by a lengthy duration, requiring considerable time for detection by clinicians. The clinical manifestations of this disease are contingent upon factors such as the size, location, and proximity to adjacent organs. The onset of symptoms can vary significantly, with an incubation period spanning several months to years. In cases of hepatic hydatid cysts, patients may experience abdominal pain, hepatomegaly, cholestasis, biliary cirrhosis, and ascites. Conversely, pulmonary hydatid cysts can give rise to chronic cough, dyspnea, expectoration, hemoptysis, lung abscesses, and pleuritis. According to the studies conducted by Romig (2003), Jenkins (2005), and Tiaoying *et al.* (2005), hydatid cysts continue to pose a significant public health risk in regions endemic to this condition, including Mediterranean countries, The regions encompassing North and eastern Africa, Western and Central Asian China, South American, and Australia are of interest in this context. The occurrence of *Echinococcus granulosus* infection in humans in regions where it is prevalent exhibits a range of 1 to 10 new cases per 100,000 individuals annually (Spies, *et al.*, 2008). Various serological tests have been developed for the purpose of diagnosing (HD). While none of these methods can be considered the ultimate solution, they do offer additional data for the identification of cases and monitoring of patients post-treatment (Zhang *et al.*, 2003). The enzyme-linked immunosorbent assay (ELISA) has been employed for the immunodiagnosis of hydatid disease, as demonstrated by Farag *et al.* (1975) and Afferni *et al.* (1984).

MATERIALS AND METHODS

Sampling of Cases of *Echinococcus granulosus* Patients:

This study was conducted from November 2022 to April 2023 at the Al-Ameen Center for Research and Advanced

Biotechnology / Imam Ali Holy Shrine. A total of 50 serum samples were collected from patients with *Echinococcus granulosus* and 38 serum samples from healthy humans, including females 74 % compared with 26% for males were taken from the Al-Furat AL-Awsat Hospital, Maternity and Children Teaching Hospital and Al-Diwaniyah Teaching Hospital in Al-Diwaniyah and from Specialized Hospital for Gastroenterology and Hepatology in AL-Najaf.

Test Principle of Human IgE ELISA Kit:

The intended use of this IgE ELISA kit is limited to laboratory research purposes and it is not suitable for diagnostic or therapeutic procedures. The colour transformation from blue to yellow is induced by the addition of the Stop Solution, followed by the quantification of colour intensity at a wavelength of 450 nm utilising a spectrophotometer. The IgE ELISA Kit incorporates a series of calibration standards to facilitate the quantification of IgE concentration in the given sample. The calibration standards are assayed concurrently with the samples, the operator is able to generate a standard curve that establishes a relationship between Optical Density and IgE concentration. The quantification of immunoglobulin E (IgE) concentration in the samples is subsequently evaluated by comparing the optical density (O.D.) of the samples to the standard curve.

RESULTS

From Table (1) it can be noted that the concentration level of IgE was increased in patients compared with the control (53.630±6.179) in control and (78.896±6.845) in patients and with a significant difference at ($P \leq 0.05$). From Table (2) it can be noted that the levels of IgE concentrations in patients according to the location of the cyst were (99.445±8.55), (62.291±14.547), (43.714±12.387) and (53.718±8.632) in Liver, lung, kidney and other organs respectively, where the highest level was in the liver.

Table 1: levels of IgE antibody in Controls and patients.

Parameters	Groups		P-value
	Control N (38) Mean±SE	Patients N (50) Mean±SE	
IgE	53.630±6.179	78.896±6.845	0.015*

* significant difference at ($P \leq 0.05$). Independent samples t-tests for variables.

Table 2: levels of IgG, IgE, and IgM antibodies in patients concentrations in patients according to the location of the cyst.

Parameters	Groups				P-value
	Liver N (26) Mean±SE	Lung N (14) Mean±SE	kidney N (5) Mean±SE	Others N (5) Mean±SE	
IgE	99.445±8.55 ^a	62.291±14.547 ^{ab}	43.714±12.387 ^b	53.718±8.632 ^b	0.012*

* significant difference at ($P \leq 0.05$).

DISCUSSION

The study of Al-Husseini, (2014) recorded infection rates of females and males (77.7%, 22.2%). The studies by Al-Mukhtar and Qasim, (2017) confirmed that the rate of infection of females amounted to (8%), while males were less by a lesser rate of (2.86%) of the total number of examined samples amounted to 480 cases, just 24 cases were positive. Also, the study (Baraak, 2014) indicated, that females are more likely to have the hydatid cyst than males, as the percentage of females is 60%, whereas the percentage of infection in males is 40% for the groups of patients chosen from the country.

The incidence of CE exhibited a statistically significant disparity between females and males, aligning with the outcomes reported in previous research studies (Khan *et al.*, 2020). Based on the study conducted by AL-Masoudi *et al.* (2021), it was observed that out of the total 53 patients diagnosed with CE, 37 individuals (56.9%) were identified as female, while the remaining 16 individuals (43%) were identified as male.

(Ismail *et al.*, 2023) record that the present study encompassed a cohort of individuals diagnosed with hydatid disease, who were admitted to the "Al-Sadr" General Hospital during the period spanning from January 1, 2020, to December 31, 2020. The total number of patients included in this study

amounted to 58. Based on the research findings, it has been determined that rural areas exhibit the highest prevalence of this particular ailment, with the peasantry emerging as the demographic group most significantly impacted among females. The rationale behind this phenomenon can be attributed to the inherent characteristics of their agricultural and livestock-related activities. The results of the study also indicated that the prevalence of infection is higher among females compared to males.

The reason for the higher incidence in youth than other ages may be due to the activity of this group in farming and social work being closer contact to pathogens, maybe breed of pets and eating food from public restaurants furthermore silence of cyst development and long incubation period of hydatidosis by Al-Husseini, (2014) and Baraak, (2014) may be due to differences in categories distribution.

Hydatid disease is an exceptional parasitic problem that exhibits the ability to proliferate and thrive in various anatomical locations within the human body. This condition presents a diverse range of clinical presentations, which are contingent upon the developmental stage of the parasite, the presence of associated complications, and the specific tissues affected (Jenkins, 2005). The most commonly involved organ is the liver (59-75%) and others include lung (27%),

kidney (3%), bone (1 - 4%), and brain (1 - 2%) (Yuksel *et al.*, 2007). The results showed that the liver was the more affected organ in comparison to other organs this result agreed with previous study on human hydatidosis by Baraak, (2014) in Iraq which recorded rates of 61.8%, 55.6%, 76.67% and 50% respectively; as well it matches the study of Brundu *et al.*, (2014) in Italy and Zhang *et al.*, (2015) in China which recorded 83.6% and 95.08% respectively.

(Omidinia *et al.*, 2020) showed that in Alborz Province the most infected organ was the liver 21 (80.8%) followed by lungs 4 (15.4%) and one case of pelvic cyst 1 (3.8%).

(Al-marsomy, 2022) reported that the study demonstrates that infection can involve multiple organs. Specifically, the liver was found to be infected in 60 patients, while the lung, kidney, and spleen were affected in only 2 patients.

(Li *et al.*, 2020) reported that *Echinococcus granulosus* cysts induce a strong antibody response in most patients, In the chronic phases of CE, the level of IgE is frequently elevated. (Sterla *et al.*, 1999; Zhang *et al.*, 2008) found that the anti-hydatid antibody responses in clinically and radiologically diagnosed patients were found positive in 55 of 62 (IgE). (Naik *et al.*, 2015) showed that the diagnostic sensitivity in the clinically and radiologically suggestive cases (n = 62) for IgE was 89 %. (Tao *et al.*, 2022) recorded that the plasma level of total IgE was significantly increased in CE patients compared with healthy donors. However, Force *et al.* (1992) reported that ELISA IgG was the most sensitive test (91 %) the ELISA IgE (Force *et al.*, 1992).

The elevated concentration of IgE antibodies plays a significant role in the manifestation of allergic reactions, which is considered one of the clinical indicators observed in affected individuals. This phenomenon is involved in the modulation of eosinophil proliferation and the regulation of histamine and mast cell concentrations within the bloodstream. Moreover, a notable elevation in the concentration of immunoglobulin IgE stands as a significant

hallmark of parasitic worm infection (Wilson *et al.*, 2019).

Conclusion

1. The prevalence rate of Echinococcosis is significantly higher among the 21-40 years age group.
2. The prevalence of hydatidosis was higher in females than males.
3. The hydatid cysts were mostly observed in the liver rather than other organs.

REFERENCES

- Afferni, C., C. Pini, P. Misiti-Dorello, L. Bernardini, M. Conchedda, and G. Vicari. 1984. Detection of specific IgE antibodies in sera from patients with hydatidosis. *Clinical and Experimental Immunology*, 55:587-592.
- Ahmadi, N.A. and Bodi, F. (2011). Clinical Presentation, Localization and Morphology of Hepato-Pulmonary Hydatid Cysts in Patients Operated in Tehran. *World Applied Sciences Journal*, 12(9): 1544-1548.
- Al-Husseini, N.A.M. (2014). Identify Genetic Variation for Parasitic Strains of *Echinococcus granulosus* in Human and Some Intermediate Host by Using The Polymerase Chain Reaction (PCR) and to Determination The Most Common Strain in The Human at Al-Qadisyiah Province. M.Sc. Thesis, College of Education/Al-Qadisyiah University.
- Al-marsomy, W. A. (2022). Epidemiology of Hydatid Disease in Iraq : A Study of Hydatidosis Patients in Epidemiology of Hydatid Disease in Iraq: A Study of Hydatidosis Patients in Baghdad Province. March.
- AL-Masoudi, H. K., Al-Hamadani, K. C., & Khiarull, I. A. (2021). Interleukin 17 Cytokine Profiles in Patients with Cystic Echinococcosis in Babylon Province, Iraq. *Archives of Razi Institute*, 76(5), 1089–1096. <https://doi.org/10.22092/ari.2021.355855.1730>.

- Al-Mukhtar, A. and I. J. Qasim (2017). Serological Survey of Hydatid Disease in Asymptomatic Peoples in Mosul City, Iraq. *Rafidain Journal of Science*, 26(1): 1-8.
- Baraak, M. (2014): Molecular study on cystic echinococcosis in some Iraqi patients, PhD thesis, University of Baghdad. University of Baghdad, Iraq.
- Brundu, D.; Piseddu, T.; Stegel, G.; Masu, G.; Ledda, S. and Masala, G. (2014). Retrospective Study of Human Cystic Echinococcosis in Italy Based on the Analysis of Hospital Discharge Records Between 2001 and 2012. *Acta Tropica*, 140: 91-96.
- Budke, C.M.; Deplazes, P. and Torgerson, P.R. (2006). Global Socioeconomic Impact of Cystic Echinococcosis. *Emerging Infectious Diseases*, 12: 296-303.
- Deplazes, P., Rinaldi, L., Alvarez Rojas, C. A., Torgerson, P. R., Harandi, M. F., Romig, T., Antolova, D., Schurer, J. M., Lahmar, S., Cringoli, G., Magambo, J., Thompson, R. C. A., & Jenkins, E. J. (2017). Global Distribution of Alveolar and Cystic Echinococcosis. In *Advances in Parasitology*, (Vol. 95). Elsevier Ltd. <https://doi.org/10.1016/bs.apar.2016.11.001>.
- Eckert, J., & Deplazes, P. (2004). Biological, epidemiological, and clinical aspects of echinococcosis, a zoonosis of increasing concern. *Clinical microbiology reviews*, 17(1), 107–135. <https://doi.org/10.1128/CMR.17.1.107-135.2004>.
- Farag, H., D. Bout, and A. Capron. 1975. Specific immunodiagnosis of human hydatidosis by the enzyme-linked immunosorbent assay (ELISA). *Biomedicine*, 23:276-278.
- Force, L., Torres, J. M., Carrillo, A., & Buscà, J. (1992). Evaluation of eight serological tests in the diagnosis of human echinococcosis and follow-up. *Clinical infectious diseases*, 15(3), 473-480.
- Ismail, M. A., Mohammed, S. J., & Hassan, N. H. (2023, April). The epidemiology of hydatid cysts liver diagnosis in Najaf governorate. In *AIP Conference Proceedings*, Vol. 2776 (1). AIP Publishing.
- Jenkins, D.J. (2005). Hydatid Control in Australia: Where It Began, What We Have Achieved and Where to From Here. *International Journal of Parasitology*, 35: 733-740.
- Khan, A., Ahmed, H., Simsek, S., Afzal, M. S., & Cao, J. (2020). Spread of cystic echinococcosis in Pakistan due to stray dogs and livestock slaughtering habits: research priorities and public health importance. *Frontiers in Public Health*, 7, 412.
- Li, Z. D., Mo, X. J., Yan, S., Wang, D., Xu, B., Guo, J., Zhang, T., Hu, W., Feng, Y., Zhou, X. N., & Feng, Z. (2020). Multiplex cytokine and antibody profile in cystic echinococcosis patients during a three-year follow-up in reference to the cyst stages. *Parasites and Vectors*, 13(1), 1–10. <https://doi.org/10.1186/s13071-020-4003-9>.
- Mahdi Fakhar, Masoud Keighobadi , Hajar Ziaei Hezarjaribi , Mahbobeh Montazeri , Elham S. Banimostafavi , Shahram Sayyadi , Mohammad M. Ghaffari Hamadani , Ali Sharifpour , Rabeeh Tabaripour , Samira Asadi , Masoud Soosaraei, A. A. K. (2021). Two decades of echinococcosis/hydatidosis research: Bibliometric analysis based on the web of science core collection databases, (2000–2019).
- Naik, M. I., Tenguria, R. K., & Haq, E. (2015). Detection of specific IgG, IgM, IgE and IgG subclass antibodies for serological diagnosis of human cystic echinococcosis. *Helminthologia (Poland)*, 52(2), 85–88. <https://doi.org/10.1515/helmin-2015-0016>.

- Omidinia, N., Zibaei, M., Hosseini, H., Pourrostami, K., Vafae Eslahi, A., & Badri, M. (2020). Human hydatidosis in Alborz Province: a 5-year retrospective epidemiological analysis of hospitalized cases (2014-2019). *Annals of Parasitology*, 66(4), 587-592. <https://doi.org/10.17420/ap6604.302>.
- Romig, T. (2003). Epidemiology of Echinococcosis. *Langenbeck's Archives of Surgery*, 388: 209-217.
- Spies C, Weisskopf M, Ohnsorge JA (2008) Intraspinal echinococcosis within the lumbar spine of an 18-year-old male patient. *z orthop unfall journal*, 146:463-467.
- Sterla, S., H. Sato, and A. Nieto. 1999. Echinococcus granulosus human infection stimulates low avidity anticarbohydrate IgG2 and high avidity antipeptide IgG4 antibodies. *Parasite Immunology*, 21:27-34.
- Tamarozzi, F., Deplazes, P., Casulli, A., 2020. Reinventing the wheel of Echinococcus granulosus sensu lato transmission to humans. *Trends Parasitology*, 36 (5), 427-434. <https://doi.org/10.1016/j.pt.2020.02.004>.
- Tao, J., Du, X., Liu, K., Wang, C., Lv, Y., Wang, M., Yang, Z., Yang, J., Li, S., Wu, C., Li, M., & Zhao, W. (2022). Clinical characteristics and antibodies against Echinococcus granulosus recombinant antigen P29 in patients with cystic echinococcosis in China. *BMC Infectious Diseases*, 22(1), 1-14. <https://doi.org/10.1186/s12879-022-07597-8>.
- Tiaoying, L.; Jiamin, Q.; Wen, Y.; Craig, P.S.; Xingwang, C. and Schantz, O.M. (2005). Echinococcosis in Tibetan Populations, Western Sichuan Province, China. *Emerging Infectious Diseases*, 11: 1866-1873.
- Toparlak M, Tüzer E. 2000. Veteriner Helminoloji. İstanbul, Turkey: İstanbul University; (in Turkish).
- Wilson, C. S., Brookes, V. J., Barnes, T. S., Woodgate, R. G., Peters, A., & Jenkins, D. J. (2019). Revisiting cyst burden and risk factors for hepatic hydatid disease (Echinococcus granulosus sensu stricto) in Australian beef cattle. *Preventive Veterinary Medicine*, 172(April), 104791. <https://doi.org/10.1016/j.prevetmed.2019.104791>.
- Yuksel, M., Demirpolat, G., Sever, A., Bakaris, S., Bulbuloglu, E., & Elmas, N. (2007). Hydatid disease involving some rare locations in the body: a pictorial essay. *Korean Journal of Radiology*, 8(6), 531-540.
- Zhang W., Li J., McManus D.P.(2003). Concepts in immunology and diagnosis of hydatid disease. *Clinical Microbiology Reviews*, 16, 18-36.
- Zhang, T.; Zhao, W.; Yang, D.; Piao, D.; Huang, Sh.; Mi, Y.; Zhao, X.; Cao, J.; Shen, Y.; Zhang, W. and Liu, A. (2015). Human Cystic Echinococcosis in Heilongjiang Province, China: A Retrospective Study. *BMC Gastroenterology*, 15(29): 1-5.
- Zhang, W., Ross, A. G., & McManus, D. P. (2008). Mechanisms of immunity in hydatid disease: implications for vaccine development. *The Journal of Immunology*, 181(10), 6679-6685.