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# Seroepidemiological Survey of Human Hydatidosis in Patients Referred to the Health Service Centers of Hamadan in 2019

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#### Abstract

**Background and aims:** According to evidence, a hydatid cyst is one of the most important health issues in most parts of the world. This study aimed to investigate the frequency of human hydatidosis in patients referred to health service centers in Hamadan province in 2019 using the enzyme-linked immunosorbent assay (ELISA)method.

**Methods:** In this cross-sectional study, blood samples were collected from 300 patients referred to medical diagnostic laboratories in Hamadan and evaluated for the presence of immunoglobulin G antibodies against hydatid cysts by ELISA. The data were analyzed using SPSS software.

**Results:** Out of 300 samples, 16 (5.3%) cases were positive, including 10 (5.6%) cases of the residents of urban areas and 6 (4.9 %) cases from those residing in rural areas. Moreover, of all the positive cases, 8 (50%) cases were males and 8 (50%) cases were females. The highest percentage of infection (45.5%) was observed in the age group 20-30 years.

**Conclusion:** The findings indicated that human hydatidosis is a public health problem in this province. The implementation of control and prevention programs, as well as increasing the knowledge of people, can help in controlling and reducing infection in humans and livestock.

Keywords: Hydatid cyst, ELISA, Seroepidemiology, Hamadan

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### Introduction

Hydatidosis is one of the most important zoonotic diseases caused by Echinococcus granulosus.<sup>1,2</sup> This infection is common between humans and animals in most parts of the world, especially in countries with vast sheep rising. It annually leads to plenty of health and economic losses to the community and family economy.<sup>3</sup> The causative agent of this infection is E. granulosus, the most important definitive hosts of which are dogs and other Canidae, and its intermediate host is herbivores, especially sheep.<sup>4</sup> Humans are accidental intermediate hosts and biological dead-ends for this parasite. Humans take infections by drinking water, eating raw vegetables, and having direct contact with dogs and contaminated soil. The larval stage of this parasite is one of the most pathogenic stages of a parasite for humans.<sup>5</sup> Hydatidosis is usually caused by the growth of metastatic cysts in the intermediate host, including the human. Accordingly, in humans, the severity of symptoms and complications depends on the severity of the infection and the location of the larvae formation (cysts) in different organs. In general, cysts are often located in the liver and lungs, but other organs such as muscles, spleen, soft tissues, bone marrow, heart, and brain can also be considered as the larval location (hydatid

cyst).<sup>6-8</sup> This disease has been reported in all provinces of Iran, including Khorasan province with the highest rate of infection in humans (4.5/100000) and Hormozgan province with the lowest rate (0.1/100000). For the whole country, the average rate of surgery cases due to hydatid cysts in an under-estimated evaluation was reported as 1.2 in 100 000.<sup>9</sup>

In recent years, about 10 sub-species or genotypes have been defined for *E. granulosus*. Hydatid cyst diagnosis is difficult because of the lack of specific clinical signs and symptoms, and it requires imaging techniques or serologic methods to diagnose the disease. <sup>10</sup> However, some workers suggested that serological tests are preferred to imaging techniques because of the easy use, cost-effectiveness, and lack of a need to complex equipment.

The use of immunological methods is important because the rapid and accurate diagnosis of the disease is necessary for controlling and treating the disease and preventing its recurrences. Among these methods, immune-electrophoresis, skin, indirect hemagglutination, and immunofluorescence tests have shown less sensitivity and specificity in identifying the anti-parasitic antibodies of *E. granulosus* compared to enzyme-linked immunosorbent assay (ELISA) and Western blot. Thus, they are not

suitable for seroepidemiological studies.<sup>11</sup> The antigen Bisa-specific protein of a hydatid cyst with a molecular weight of 120-160 kDa can also be used to diagnose human hydatidosis by the ELISA serology method.<sup>11-13</sup>

Therefore, this study aimed to investigate the frequency of human hydatidosis in the target population using the ELISA method in Hamadan in 2019. On the other hand, it is an important issue in health policy-making for controlling hydatidosis in the community and adopting preventive policies to control the transfer from reservoirs to humans.

# Materials and Methods Study Area

Hamadan, the capital of Hamadan province in the west of Iran, is located on the northern slope of Alvand Mountain. This city is located at an altitude of 1741 m. The province lies between 59° and 33′ to 49° and 35′ north latitude and 34° and 47′ to 34° and 49′ east of the Greenwich Meridian.

The maximum and minimum absolute temperatures in this province are 36.8°C and -29.6°C, respectively, and the average temperature is 9.6°C. The hottest months of the year, with a maximum temperature of 35°C, are July and August, while the coldest month of the year, with an average of -25.5°C, is February. Hamadan is considered as one of the coldest cities in Iran. According to the latest official Census of the country, the population of Hamadan province is 1758 268 (Figure 1).

# Study Population, Sampling, and Sample Size

In this cross-sectional study, 8 city health centers were determined by the cluster random sampling method, and 300 blood samples (40 from each health center because 20 samples were not eligible for the test) were randomly collected from the people who referred to Hamadan health centers to obtain different services in 2019. According to the results of some reports on the epidemiology of hydatidosis in Iran and because of some

financial limitations, the total studied proper samples reached 300 individuals.

Accordingly, blood samples were collected after obtaining informed consent from all participants and completing a questionnaire. A 3 mL blood sample was taken from 20 men and 20 women referred to each health center. Thereafter, their blood samples were poured into the tubes and then centrifuged at 2500 rpm to separate the sera. The sera were kept at -70°C until use.

Anti-hydatid cyst antibody (Immunoglobulin G) was detected by the ELISA method using the kit (Pishtaz Teb, Tehran, Iran) according to the manufacturer's instructions.

The absorption values above 0.27 (cut off=0.27) were considered positive. Borderline cases were also considered positive due to their proximity to the cut-off range. The obtained data were analyzed using SPSS software (version 14) and compared by the chi-square test.

#### Results

Of a total of 300 studied people, 150 (50%) and 150 (50%) subjects were men and women, respectively. In terms of education, 108 (36%), 129 (43%), 46 (15.3%), and 17 (5.7%) cases were illiterate, junior high school students, high school students, and university educated, respectively. In addition, 169 (56.3%), 87 (29%), and 43 (14.3%) individuals washed their vegetables with tap water, disinfectants, and salt water, respectively. The highest age group in this study was 60-70 years old with 48 (16%) persons, while the lowest age group was over 80 years old with 14 persons. Out of the total population, 19 (3%), 124 (41.3%), 51 (17%), and 106 (38.7%) cases were unemployed, housewives, employees, and self-employed, respectively. According to the results, 16 subjects (5.3%) were positive for hydatid cyst antibodies. The majority of positive cases (56.25%) were in the age range of 20-40 years old (Figure 2). Out of 16 infected persons, 8 cases were women and 8 cases were men (Table 1); of

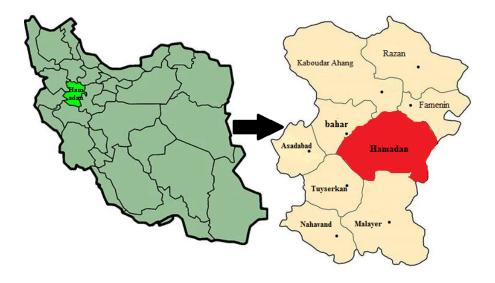


Figure 1. Map of Hamadan Province and Hamadan City. Source: Ghobadi et al<sup>15</sup>

Table 1. Percentage and Frequency of Infection With Hydatid Cysts According to the Studied Variables

Studied Variable		Total	<b>ELISA Test Results (Positive)</b>	ELISA Test Results (Negative)	P Value
Residence place	City	178	10 (5.6%)	168 (94.4%)	0.791
	Village	122	6 (4.9%)	166 (95.1%)	
Gender	Man	150	8 (5.3%)	142 (94.7%)	0.999
	Female	150	8 (5.3%)	142 (94.7%)	
Contact history with dog	Yes	22	1 (4.5%)	21 (95.5%)	0.864
	No	278	15 (5.4%)	263 (94.6%)	
Education	Illiterate	108	4 (35.7%)	104 (96.3%)	0.088
	Junior high school	129	5 (3.9%)	124 (96.1%)	
	Diploma	46	6 (13%)	40 (87%)	
	University	17	1 (5.9%)	16 (94.1%)	
How to wash vegetables	Water	169	14 (8.3%)	155 (91.7%)	0.035*
	Disinfectants	87	1 (1.1%)	86 (98.9%)	
	Salt water	43	1 (2.3%)	42 (97.7%)	
Job	Unemployed	19	2 (10.5%)	17 (89.5%)	0.480
	Housewife	19	4 (3.2%)	120 (96.8%)	
	Employee	51	3 (5.9%)	48 (94.1%)	
	Self-employed	106	7 (6.6%)	99 (93.4%)	

Note. ELISA: Enzyme-linked immunosorbent assay.

whom 4 subjects (25%) were housewives, and among men, 2 (12.5%), 3 (18.75%), and 7 (43.75%) cases were unemployed, employed, and self-employed, respectively. A total of 22 subjects had a contact history with dogs (7.33%), of whom 1 person (4.5%) was recognized to be positive (Table 1). Additionally, 6 (4.9%) out of 122 (100%) rural inhabitants and 10 (5.6%) out of 178 (100%) urban inhabitants were positive. All patients had a history of consuming raw vegetables, among them, those who washed vegetables with tap water were 14 persons (8.3%); only one person used to wash them with disinfection (1.1%), and only one person washed vegetables with salt water (2.3%). Further, the highest frequency in people

aged 60-70 years old was 48 (16%), and the mean age of the patients in this study was 45.1 years old.

#### Discussion

The findings of this study indicated that hydatidosis is a major problem in this region of Iran. Recent research in some provinces of Iran demonstrated the seroprevalence of hydatidosis in the whole population from 1.7% in Khuzestan province in the southwest of Iran to 4.7% in East Azerbaijan<sup>4</sup> and 2.2% in the Golestan Province in the northeast of Iran. Currently, hydatidosis is one of the most important parasitic diseases worldwide. This disease is observed in all five continents with a considerable

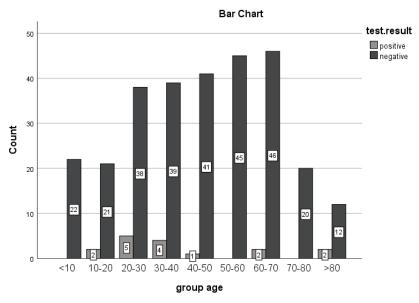


Figure 2. The Results According to Different Age Groups

distribution.<sup>14</sup> The health and economic loss caused by this disease cannot be ignored in most parts of the world, especially in the Middle East, Southern and Central Europe, China, and Africa. Rafiei et al reported a 13.8% prevalence rate of hydatid cysts in Khuzestan province, in the southwest of Iran. 16,17 In another study, Zhang et al reported 92.3% and 89.9% sensitivity and specificity of this test using crude hydatid cyst fluid, respectively.<sup>18</sup> Likewise, Pappas et al19 determined the sensitivity and specificity of this test as 96% and 98% using crude hydatid cyst fluid, respectively. Jalusian et al also performed a Dot-ELISA method with the crude hydatid cyst fluid antigen and determined the sensitivity and specificity as 100% and 97%, respectively. According to the findings of Khan et al, most cases of infection were in the age group of 20-70 years age, and most cases of infection in foreign reports were in the age group of 20-29 years old.<sup>20</sup>

Considering that this disease can become a chronic infection, it is usually diagnosed after passing several years after being infected.21 Some studies reported that the infection rate was higher in women than in men. Our findings showed that the education level is an effective factor in the infection rate of hydatid cysts because the prevalence rate of infection in people with low levels of education is more than in those with high levels of education; in addition, it seems that the literacy level is directly related to the awareness of people about the transmission route of this disease.<sup>22</sup> In the study by Wilson et al,23 the illiteracy factor was significantly different in the group of patients with hydatid cysts and the control group. Accordingly, the illiteracy rate in the patient group was 13.3% compared to the control group (1.9%). The consumption of improper washed raw vegetables and the type of drinking water are among the most important routes of infection to hydatidosis in humans; thus, there was a significant relationship between seropositivity and the type of raw vegetable washing.

In their study, Dalimi et al reported that the prevalence of hydatidosis by immunofluorescence was 9.5% and 3.3% in the Diwandareh region of Kurdistan and in the suburb of Sanandaj, respectively, and about 70% of the positive cases were women.24 In a seroepidemiologic study of human hydatidosis in the Shahriar region by Sedaghatghohar et al, the infection rate in two rural areas around Shahriar was 5.9% and 3.2%, respectively (80.7% women and 19.3% men).25 In a seroepidemiologic study of hydatidosis in Zanjan by Haniloo et al, the prevalence rate of hydatidosis was reported as 3% in the Islamabad area by the ELISA method, including 62% women, and there was a significant relationship between gender and hydatidosis. Likewise, Sadjjadi et al reported that the prevalence of positive serum cases of hydatid cysts by the ELISA method was 7.2%, of which 50.4% were women and 49.6% were men.26 In a seroepidemiologic study of hydatid cysts in patients referred to some medical centers in Golestan Province, Baharsefat et al<sup>27</sup> reported a prevalence rate of 2.34% and 2.2% by immunofluorescence and ELISA methods, respectively. In this study, although the number of positive serum cases was higher in women than in men, no significant differences were reported in terms of gender. Based on the results obtained from the ELISA test, the percentage of positive serum cases of human hydatidosis in Hamadan was determined 16/300 (5.3%). Although the percentage of positive serum cases of hydatid cyst disease in men and women was 8 (5.3%) and 8 (5.3%), respectively, no significant relationship was observed regarding gender (P=0.60). There are some limitations in the seroepidemiologic studies, including the present study. The studied population, including those who refer to health centers, could not be the real representative of the whole population, and this method only indicates the history of exposure to the infective agent rather than a positive case of the disease.

#### Conclusion

The prevalence rate of hydatid cyst infection in Hamadan was found to be almost equal to the other parts of the country. The health and economic importance of hydatidosis in humans and animals can lead to the imposition of significant costs to society, thus the implementation of infection control and prevention programs, as well as increasing the level of awareness of people can help in controlling and reducing infection with this common parasite in humans and livestock in this region and the country. According to the obtained data, some limitations were not fully addressed in some aspects, which are suggested to be examined in future studies.

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#### **Author Contributions**

MF designed the study, collected data, administrated the project, and wrote the original draft. MM designed the study and did formal analysis and investigation, and MB prepared methodology, supervision, validation, visualization, writing, reviewing, and editing. In addition, all authors read and approved the final manuscript.

#### **Conflict of Interest Disclosures**

The authors declare no conflict of interests.

#### **Ethical Approval**

Ethical approval for the study was obtained from the Ethics Committee of Chancellor for Research and Technology, Hamadan University of Medical Sciences according to confirmation of informed consent form of assigned it an exclusive code of IR.UMSHA.REC.1399.436.

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