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Seroprevalence and Risk Factors of Cystic Echinococcosis among Children in Lorestan Province, Western Iran

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ABSTRACT

Hydatidosis is as a zoonotic disease can be transmitted to human by consumption of the parasite's eggs through vegetables, water, food and also via having contact with dog. Children because of contact with the animals and lack of hygiene standards in the consumption of food, water, fruits and vegetables are far more at risk of hydatidosis. This study was aimed to evaluate seroprevalence and risk factors Cystic echinococcosis (CE, hydatid cyst) in children (2-15 year old) referred to health centers of Lorestan province, Iran. This cross-sectional study was performed from August 2016 to March 2017 on 316 children (aged 2-15 years), referring to health centers of Lorestan province, Iran. All the serum samples were tested using the commercially ELISA kit of NovaLisa® Echinococcus IgG (Novatec, Germany) according to the manufacturer's instructions. Statistical analysis was carried out using SPSS 17.0 software. Multifactorial logistic regression models were applied to determine association between CE seropositivity and the potential risk factors. Of the 316 children, 9 (2.84%) tested seropositive for anti-hydatid IgG antibody. In multifactorial logistic regression models, some risk factors such as living in rural regions (4.3; 90% CI: 1.9-9.6) and contact with dogs (3.8; 90% CI: 1.7-8.9) were significantly related to seropositivity to anti-hydatid IgG antibodies. To conclude, we found that CE is prevalent among in children referred to health centers of Lorestan province, Iran. The obtained findings may be a warning for health centers to pay special attention to CE among children and also design screening programmers for prevention of CE.

Key words: Hydatid cyst, ELISA, *Echinococcus granulosus*, Iran

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INTRODUCTION

Cystic echinococcosis (CE, hydatid cyst) as one of the most important zoonotic parasitic infections around the world caused by larval stage of dog tapeworm, *Echinococcus granulosus* [1]. In this disease, the final host is the dog, in which adult worm attached to the intestinal mucosa and exerted eggs into the environment through the feces [2]. Human and domestic livestock are normally infected through ingesting food contaminated with *E. granulosus* eggs. After ingestion, the eggs hatched and larvae penetrate the intestine wall and broadly dispersed in the various organs such as liver, lungs, muscle or other organs, where the hydatid cysts develop [3]. Although human cystic echinococcosisis asymptomatic, however death can happen due to hydatid cyst rupture, which may result in anaphylacic shock and/or cause secondary infections [4-6].

Primary diagnosis of *Cystic* echinococcosis is complicated because of being asymptomatic in early steps whereas using physical imaging; mainly ultrasound (US) assessment is beneficial for both early and late stages diagnosis [7]. The early diagnosis of CE is based on available immunodiagnostic techniques with specific immunodominant antigens such as Ag B and US imaging [8]. Methods for detecting specific antibodies can provide opportunities for early treatment of the disease [8,9]. Nowadays, immunodiagnostic approaches such as ELISA and IFA have been applied for overall screening of people in endemic area by evaluating anti-Echinococcus IgG antibody [9,10].

In Iran as one of the main endemic regions the prevalence of *cystic* echinococcosis according to a random-effects meta-analysis was 5.0% (95% CI: 4.0%, 6.0%). Moreover, the prevalence of rate *cystic* echinococcos is in stray dogs as definitive host is 5%-94% in various

regions of Iran [11]. Children because of contact with the animals and lack of hygiene standards in the consumption of food, water, fruits and vegetables are far more at risk of CE; therefore it is crucial that to study the prevalence anti-Echinococcus antibodies in the children in this age range [1]. To the best of our knowledge and according to a survey of the literature, little is known about the seroprevalence of *cystic* echinococcosis in children in Iran. Therefore, this cross-sectional study aims to determine the prevalence of anti-Echinococcus IgG antibody and the associated risk factors among the children (2-15 year old) referred to health centers of Lorestan province, Iran.

MATERIAL AND METHODS

Ethics

This study was approved by Ethics Committee of Lorestan University of Medical Sciences, Khorramabad, Iran. Parents/guardians provided informed consent on behalf of all child participants.

Questionnaire

Before collection of blood samples, the applied questionnaire was based on demographic data including age, gender, and education. Moreover, possible risk factors, such as animal contacts (dog), unwashed vegetables/fruit consumption, and residence were also evaluated.

Study design and sample collection

In this cross-sectional study, totally 316 children (aged 2-15 years), referring to health centers of Lorestan province, Iran were studied during August 2016 to March 2017. This province is located between the valleys of Zagros Mountains in the west of Iran, bordering the provinces of Markazi, Hamedan, Kermanshah, Khuzestan, Ilam, and Isfahan. Lorestan covers an area of 28,294 km^2 with a population of approximately 2 million people. 5 mL of blood was obtained from each of the patient by means of venipuncture, under sterile conditions. The samples were centrifuged at 1000 rpm and the sera were stored at -20°C until serological examination [12,13].

Enzyme-linked immunosorbent (ELISA) test

To determine the anti-Echinococcus IgG antibody, all the serum samples were tested using the commercially available ELISA kit of NovaLisa® Echinococcus IgG (Novatec, Germany). Analyses were carried out according to the manufacturer's instructions. Based on the ELISA kit, positive and negative results for anti-Echinococcus IgG antibody were defined as index values of ≥ 11 and ≤ 9 NovaTec units (NTU), respectively. Also, values between 9 NTU and 11 NTU were considered as inconclusive results.

Statistical analysis

Statistical analysis was carried out using SPSS 17.0 software (SPSS Inc., Chicago, IL, USA). Descriptive statistics were reported in terms of per cent (for categorical) and mean (SD) (for continuous) variables. We used Chi-square test to examine the univariate association between independent variables and outcome. Next, all the variables in univariate analysis that had a $p<0.25$ were arrived into multivariate analysis (logistic regression). Multifactorial logistic regression models were applied to determine association between CE seropositivity and the potential risk factors. $P<0.05$ was considered to be statistically significant.

RESULTS

Participants

In this cross-sectional investigation, totally 316 children referred to the health centers of Lorestan province, Iran were studied to determine the prevalence of anti-Echinococcus IgG antibody and the associated risk factors among them. The mean age of the children was 7.9 ± 2.5 years; whereas the majority of children were boys (176, 55.7%). Among the participants, 240 children (75.9%) lived in urban areas, but the 76 (24.1%) lived in rural parts. Out of 316 children, 85 children (26.9%) were in contact with dogs; while remaining (73.1%) did not have any contact with dogs. Between participants, 32 children (10.1%) consumed raw or unwashed vegetables and fruits; whereas 284 (89.9%) did not consumed raw or unwashed vegetables and fruits (Table 1).

Table 1: Demographic characteristics and seroprevalence of anti-hydatid antibodies among children (aged 2-15 years), referred to health centers of Lorestan province, Iran

Variables	No. (%)	IgG positive	P value
Gender			
Male	176 (55.7)	6 (3.4)	0.84
Female	140 (44.3)	3 (2.1)	-
Age groups			
<7 years	105 (33.2)	2 (1.9)	-
≥ 7 years	211 (66.8)	7 (3.3)	0.087
Residential place			
Urban	240 (75.9)	4 (1.6)	-
Rural	76 (20.1)	5 (6.6)	0.0031*
Being in contact with dogs			
No	231 (73.1)	4 (1.7)	-
Yes	85 (26.9)	5 (5.9)	0.0024*
Unwashed vegetables/fruit consumption			
No	284 (89.9)	8 (2.8)	-
Yes	32 (10.1)	1 (3.15)	0.65

*P<0.05, difference is statistically significant

Seroprevalence of anti-hydatid antibodies

Out of the 316 children, 9 (2.84%) tested seropositive for anti-hydatid IgG antibody. The obtained results showed that seroprevalence of anti-hydatid IgG antibodies were higher between boy children; so that six (3.4%) boy were positive for IgG anti-T. canis antibodies compared

to 3 (2.1%) girls (p=0.84).

From 240 children, who living in urban areas, 4 (1.66%) were found seropositive for anti-hydatid antibodies; while out of 76 children that lived in rural regions, 5 (6.6%) children tested seropositive for anti-hydatid antibodies. These results showed that there was a significant difference in the prevalence of anti-hydatid antibodies among the children, who living in urban and those living in rural areas (p=0.0031).

Among the 85 children that have contacted with dog, 5 (5.9%) tested seropositive for anti-hydatid antibodies, but from 231 children that did not have any contact with dog, only 4 (1.7%) children were found positive for anti-hydatid antibodies; indicated that there was significant difference in the prevalence of anti-hydatid antibodies among the children who contacting with dog and those children who no contacting with dog (p=0.0024).

Among the tested children, who consumption raw or unwashed vegetables and fruits, just 1 (3.15%) were seropositive for anti-hydatid antibodies; while from 284 children who did not eat raw or unwashed vegetables and fruits, 8 (2.8%) children were found seropositive for anti-hydatid antibodies; indicating that there was no significant association in the prevalence of anti-hydatid antibodies among the children who consumption raw or unwashed vegetables and fruits compared to children who did not eat raw or unwashed vegetables and fruits (p=0.65). Furthermore, statistical analysis also demonstrated that seroprevalence of anti-hydatid IgG antibodies was not different with age among the children between 2 to 15 year (Table 1).

In multifactorial logistic regression models, some risk factors such as living in rural regions (4.3; 90% CI: 1.9-9.6) and contact with dogs (3.8; 90% CI: 1.7-8.9) were significantly related to seropositivity to anti-hydatid IgG antibodies (Table 2).

Table 2: Logistic regression analysis of the potential factors associated with anti-hydatid antibodies among children (aged 2-15 years), referred to health centers of Lorestan province, Iran

Variables	OR (95% CI)	P value
Residential place		
Urban	1	-
Rural	4.3 (1.9-9.6)	0.012*
Being in contact with dogs		
No	1	-
Yes	3.8 (1.7-8.9)	0.000*

*P<0.05 was statistically significant

DISCUSSION

Hydatidosis is as a zoonotic disease can be transmitted to human by consumption of the parasite's eggs through vegetables, water, food and also via having contact with dog [1]. The CE is broadly distributed generally in regions where sheep are raised as a main industry [1]. In general, clinical manifestations of CE expand consequently this space-occupying lesion producing pressure on nearby organs [2]. Moreover, laceration of the cyst and linkage of the cyst contents (protoscoleces) can cause anaphylactic shock and secondary CE infections. Nowadays, surgery consider the selection treatment for most patients who suffering from hydatidosis [3,6]. For the reason that the current therapies are not completely useful and no safe and effective vaccines are existing, it is important to make efforts for diminishing transmission of hydatidosis to decrease the serious manifestations of disease. Early diagnosis of CE in human is complicated because of being asymptomatic in early phases whereas some physical imaging such as ultrasound (US) tests can be used in the all stages of disease [6]. To date, serological tests such as ELISA, IHA, Western Blot, etc., due to reliable sensitivity and accuracy broadly have been used to diagnosis of CE as well as in the research studies around the world [10].

According to the previous investigations, the overall seroprevalence of human hydatidosis in was 5%; while the prevalence of hydatidosis in dogs has been reported between 5%-94% in various regions of Iran [11]. Children in the first and second life decades due to contact with the definitive host (dogs) and lack of hygiene standards in the consumption of food, water, fruits and vegetables are far more at risk of CE; therefore it is crucial that to study the prevalence anti-Echinococcus antibodies in the children in this age range [1]. The present investigation was designed to evaluate the prevalence of anti-Echinococcus IgG antibody and the associated risk factors among the children (2-15 year old) referred to health centers of Lorestan province, Iran. Out of the 316 children, 9 (2.84%) tested seropositive for anti-hydatid IgG antibody. Previously a number of studies have been done on seroprevalence of hydatidosis in Iran: In the study conducted by Yousofi et al. [14] the total prevalence anti-hydatid antibodies in the in Chaharmahal and Bakhtiari province, Iran was 4.8%. Rafiei et al. [15] have reported the seroprevalence anti-hydatid antibodies 1.8% in nomads Khozestan province, southwest of Iran. In the other study, Aflaki et al. [16] the seroprevalence of hydatidosis infection in Ilam province, Iran was only 1.2%. Based on the study conducted by Asghari et al. [17] the seroprevalence of anti-hydatid antibodies in Markazi province, Iran was also 3.46%.

In the present study, in line with the study conducted by Asghari et al. [17] and Aflaki et al. [16] we found a significant difference in the prevalence of anti-hidatid antibodies among the people living in urban and those living in rural areas; however, it was in contrast to the findings of Yang et al. [18]. These differences might be related to work-related activities associated to contact with dogs and having lower hygienic life style.

The obtained results showed that contacting with dog is a risk factor for hydatidosis seropositivity. Similarly,

Yang et al. [18] and Aflaki et al. [16] demonstrated that contacting with dog as a risk factor for hydatidosis. In contrast, Rafiei et al. [14] and Asghari et al. [17] have reported no significant association between prevalence of anti-hydatid antibodies and having contact with dogs. Our results demonstrated no significant difference between age, gender, and consumption of raw or unwashed vegetables and fruits on the one hand and hydatidosis seropositivity on the other. Likewise, Rafiei et al. [15], and Aydın et al. [19] have reported no significant difference in term of age, sex, and consumption of unwashed vegetables and fruits. These disparities in the seroprevalence of anti-hydatid antibodies among the children could be related to sociocultural behaviors, geographical and environmental parameters, and transmission routes in the evaluated population [1,20]. Moreover, it should be mentioned that due to some limitations such as low sample size and not using of the more accurate techniques.

CONCLUSION

Based on the obtained findings, we found that hydatidosis is prevalent among in children (2–15 year old) referred to health centers of Lorestan province, Iran with the overall seroprevalence rate of 2.84%. Although there are some limitations such as the low sample size and not using of the more accurate techniques in this study, however the obtained findings may be a warning for health centers to pay special attention to hydatidosis among children (2-15 year old) and also design screening programmers for prevention of this disease.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests.

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