Seroepidemiological Survey of Human Hydatidosis in Western Part of Iran

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Summary
A total number of 4138 sera from apparently healthy volunteers living in 8 different provinces of the western part of Iran were collected and tested by a standard ELISA. Of those, 3908 (94.45%) were negative and 230 (5.55%) were positive. The results are monitored according to ethnic groups, sex, age, occupation, training, province and season.

***Key words*:** hydatidosis , human , antigen , ELISA , *Echinococcus granulosus*

Introduction

Hydatidosis is an important parasitic disease for herbivorous and man caused by the larval stage of *Echinococcus* species. Distribution of the disease is related to its intermediate and definitive hosts (Lukshenko 1971).

E.granulosus lives in the small intestine of carnivores, as infective hosts. Herbivorous and man acquire the larval stage through ingestion of infective eggs shed via the faeces of infected dogs. Human infection may occurs by direct contact with dogs or from contaminated environment. When ruminants are slaughtered their disposed viscera may be eaten by definitive hosts. The adult worm is then developed in their intestines (Lukshenko 1971, Mc connell 1979). Hydatid disease in human is potentially dangerous, organ type and cyst sizes are very important in the final pathogenicity of parasite (Matossian 1977).

Numerous studies based on the detection of humoral response of the host against the parasite have been carried out on development immunodiagnostic test(s) for hydatid disease in man (Laplante 1991, Liu *et al* 1992a, b). Iran is one of the endemic areas in the Middle East. Her provinces, suitable for husbandry, are located around the Zagros mountain including the affected areas (Eslami 1997, Noorja 1988).

In this study the human humoral response against *Echinococcus* antigen, to show a clear feature of the prevalence of the disease in above mentioned areas, by ELISA assay was detected.

Materials and Methods

Samples. 4138 sera from apparently healthy volunteers who living in 8 different provinces in the western part of Iran were randomly collected by cluster population method. The collected sera were frozen at -20C until use.

Solutions and buffers. All the buffers and solutions were prepared in the laboratory according to Deplazes and Felix instruction (1991) and kept in the refrigerator (4C) until use.

Antigen. 96 well microplates (Nunc immunoplates) were coated with hydatid fluid (sheep origin) as antigen. The substrate and conjugate were obtained from Dr.P.Deplazes, University of Zurich, Switzerland.

ELIZA assay. The ELISA assay was carried out according to the method described by Deplazes (1991). Antigen solution was diluted (1:200) in coating buffer. 100ml of diluted antigen was pipetted into each well of 96-well microplate and incubated overnight at 4C. Then the plates were washed with washing buffer. The plates were blocked with second with second buffer. Serum sample dilutions were made 1:200 in blocking buffer was added and incubated for 90 min at 37C.100ml/well detection antibody (conjugate) was added and incubated for 90 min at 37C. The plates were washed in washing buffer, and substrate (100ml/well) was added and incubated for 5-15 min at 37C. Known positive and negative controls were included in all test plates. The optical density (OD) for each test was calculated immediately such as average of negative calibration sera (n1, n2 and n3) multiplied by factor of two.

Statistical Analysis. All data were analyzed statistically by x^2 test.

Results

The results according to sex, age, ethnic groups and occupation and according to province, locality, season and training are showed in tables 1 and 2, respectively. The analysis of data, showing the significant differences between hydatidosis and sex ($P<0.0001$), province ($P<0.005$), locality ($P<0.005$) and season ($P<0.025$). The disease was not affected by age, ethnic groups, occupation and training.

Discussion

According to the results of this study (Tables 1,2) the prevalence of hydatid disease in the western part of Iran is 5.6%. Previous studies, which have been done based IFA

method, had showed fewer ratios (Arbabi & Masoud 1992). However, specificity and sensitivity of ELISA test could be one of its reasons, as well as high infection rates of carnivores and wild animals (20-45%) in these areas (Eslami 1997, Noorja 1988).

Table 1. *Frequency and relative frequency of ELISA according to variables*

Variables \ Results of ELISA		Negative		Positive		Total	
		No.	%	No.	%	No.	%
Sex	Male	95.3		4.7		100	
	Female	93.8		6.2		100	
	Total	3908	94.4	230	5.6	4138	100
Age	>20	785	94.4	47	5.6	832	100
	21-40	1840	94.3	111	5.7	1951	100
	41-60	901	94.9	84	5.1	949	100
	>60	382	95	24	5	402	100
	Total	3908	94.4	230	5.6	4138	100
Ethnical Groups	Turk	1990	94.8	110	5.2	2100	100
	Kord	1301	93.9	85	6.1	1386	100
	Lour	501	94.2	31	5.8	232	100
	Other	116	96.7	4	3.3	120	100
	Total	3908	94.4	230	5.6	4138	100
Occupation	Staff	496	95	26	5	522	100
	Worker	377	94	24	6	401	100
	Farmer	323	94.7	18	5.3	341	100
	Housekeeper	1778	93.6	121	6.4	1899	100
	Hunter	25	92.6	2	7.4	27	100
	Carpet-weaver	33	94.3	2	5.7	35	100
	Other	876	95.9	37	4.1	913	100
	Total	3908	94.4	230	5.6	4138	100

It is clear from the results reported that prevalence of hydatid disease was affected by sex, in female this was significantly higher ($P<0.0001$) than male. Because of, it is likely, women work in the farm and are more exposed to the animals than men. It is confirmed by previous study (Zarif-fard & Masoud 1998). Similar results were obtained in rural population of Ardabi, Eastern Azarbijan and kermanshah specially in summer and autumn. Direct contact with dogs, handling farm animals and face less public health could be important reasons for high prevalence of hydatidosis in these areas.

This study confirms and extends previous report showing that prevalence of hydatid disease is influenced by sex and locality (Zarif-fard & Masoud 1998). According to our observations, prevalence of the disease is not affected by age, ethnic groups, occupation and training. Differences between our results and the other reports could be to follow on more populations and areas.

Table 2. *Frequency and relative frequency of ELISA according to variables*

Variables \ Results of ELISA		Negative		Positive		Total	
		No.	**%**	**No.**	**%**	**No.**	**%**
Province	Ardebil	720	91.7	38	8.3		
	E.Azabaijan	648	91.4	64	8.6	100	
	W.Azarbaijan	360	97	11	3	748	100
	Eilam	775	94.7	43	5.3	371	100
	Kordestan	399	95.4	19	4.6	818	100
	Hamedan	352	94.9	19	5.1	418	100
	Lourestan	495	95	26	5	371	100
	Total	3908	94.4	230	5.6	521	100
						4138	100
Locality	Town	2383	95.4	114	4.6	2497	100
	Village	1499	93	112	7	1611	100
	Total	3882	94.5	226	5.5	4108	100
Season	Spring	853	94.5	50	5.5	903	100
	Summer	837	92.4	69	7.6	906	100
	Automn	1573	95.3	78	4.7	1651	100
	Winter	645	95.1	33	4.9	678	100
	Total	3908	94.4	230	5.6	1508	100
Education	Illiterate	1410	93.5	98	6.5	1508	100
	Primary.S	1276	94.4	76	5.6	1352	100
	Middle.S	999	95.4	48	4.6	1047	100
	Collage	223	96.5	8	3.5	231	100
	Total	3908	94.4	230	5.6	4138	100

References

Arbabi, A., Masoud, J.(1992). Seroepidemiological study in Hamadan province by IFA. M.S. thesis, School of Public Health, Medical Sience, Tehran University (In persian).

Deplazes, P., Gottstein, B.(1991). A monoclonal antibody against *Echinococcus* antigen. *Journal of Parasitology* 103: 41-49.

Eslami, A.(1997). Cestoda. In: *Veterinary Helmintology*, Vol 2. (In persian).

Laplante, J.(1991). Serological mass screening of alveolar Echinococcosis in france-comte (Abs.english). *Archive de la hydatidosis* 30:825-829.

LIU, D., Liyhtowlers, M.W. and Rickard, M.D.(1992a). Evaluation of a monoclonal antibody-based competition ELISA for the diagnosis of human hydatidosis. *Journal of Parasitoligy* 104:357-361.

LIU, D., Rickard, M.D. and Lightowlers, M.W.(1992b). Further characterization of monoclonal anthibody to *Echinococcus granulosus* antigen 5 and Antigen B. *International Journal of Parasitology* 22(3):391-394.

Lukshenko, N.P.(1971). Problems of epidemiology and prophylax of alveococcosis (multilocular. Echinococcosis) A general review-with particular reference to the U.S.S.R. *International Journal of Parasitology* 1:125-134.

McConnell, J.D., Green, R.(1979). The control of hydatid disease in Tasmania. *Australian Veterinary Journal* 55:140-145.

Matossian, R.M., Rickard, M.D. and Smyth, J.D.(1977). Hydatidosis: a global problem of increasing importance. *Bulletin of the world Healt Organization* 155(4):507.

Noorja, N.(1988). Hydatidosis-Echinococcosis. Ph.D. Thesis. School of Public Health. Medical Science. Tehran University. (In persian).

Zarif-fard, M.R., Massoud, J.(1998). Study of *Echinococcus granulosus* and *Echinococcus multilocularis* infections in conidiae in Ardabile province of Iran. *Archives of Razi Institute* 48-49:47-52.