Some Epidemiological and Serological Studies on Hydatidosis in Najran Region

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Abstract: This study was designed to record the prevalence rate of hydatidosis among farm animals and human in Najran area. Experiment I recorded the rate of hydatidosis in camels, sheep and goats. A total of 48139 animals (camels = 4531, sheep= 29916, goats= 13692) were employed in this experiment. The animals were examined at post- mortem for the presence of hydatid cysts. The results showed that the rate of prevalence differs (p<0.001) among animal species with higher rates of infection in sheep (6.8%), camels (5.4%) and goats (2.2%). The overall rate of prevalence was 5.3%. Experiment II is carried to determine the prevalence of hydatidosis in human. At total of 1142 human serum samples were tested with ELISA and IHA (ELISA =276, IHA=866) for the presence of hydatid antibodies. Fifty two serum samples (ELIZA = 17, IHA= 35) were positive for hydatid antibodies (4.5%) with no significant difference (p >0.05) between ELISA and IHA results. However, out of 57 negative samples evaluated with IHA, 4 samples were positive when tested with ELISA and out of 35 IHA positive samples 4 samples were negative when tested with ELISA. Consequently, the sensitivity, specificity and accuracy of IHA were 88.5 %, 92.9% and 91.3%, respectively. In conclusion hydatidosis prevails in Najran area at an overall rate of 5.3% in farm animals and 4.5% in human. Additionally, ELIZA and/or IHA can be used to screen hydaitdosis in human.
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1. Introduction

Hydatidosis, the known as cystic echinococcosis (CE), is caused by the larval stages of the tapeworm *Echinococcus granulosus*. This disease is one of the most important parasitic infections of livestock because it spreads worldwide and it is a serious zoonotic disease ***(Craig et al., 2007; Cringoli et al., 2007)***. CE accounts for more than 95 % of the estimated 2 – 3 million human global cases affected by Echinococcus parasites ***(Budke, 2006)***.

Hydatidosis spreads on all the continents with high prevalence rates in Eurasia, north and east Africa, Australia, and South America ***(Eckert et al., 2001).*** *Echinococcus granulosus* is found in the northern and southern hemispheres, with heaviest burdens reported in Latin America, Africa, coastal Mediterranean regions, states of the former USSR, the northern Middle East, and Southeast Asia. In a review by ***Ammann and Eckert, (1996)*** the hydatidosis infects up to 220 in 100,000 population.

The life cycle of *Echinococcus granulosus* involves domestic and wild carnivores as definitive hosts, which are infected by the ingestion of offal containing the larval forms (hydatid cysts) with viable protoscoleces producing adult stage in the intestine. Dogs are the main source of infection, although in some areas jackals, hyenas, foxes, and wolves could also play a role as definitive hosts. A wide range of domestic, wild mammals and humans act as intermediate hosts for this parasite wherein the larval stages develop after ingesting the eggs of *Echinococcus granulosus* ***(Seimenis, 2003).*** Accidental rupture of hydatid cyst during trauma can provoke severe anaphylactic reactions in human ***(Boyano et al., 1994).*** About 70% of human CE presents as chronic cystic lesions of the liver, nevertheless cystic lesions may be found in other organs including the brain ***(Wen and Yang, 1997).***

Immunodiagnostic techniques such as indirect hemagglutination (IHA) and enzyme-linked immunosorbent assay (ELISA) are used for the diagnosis of hydatidosis in human. These serological tests provide an extremely useful diagnostic methods for the disease ***(Rebhandl et al., 1999).***

In Najran area hydatidosis is not investigated and its prevalence is not known. Thus, this study was designed to record the prevalence rate of hydatidosis among farm animals and human in Najran area. Furthermore the efficacy of IHA versus ELISA was also evaluated

2. Material and Methods

Study area

This study was carried in Najran, south of Saudi Arabia which lies between 17° 30' 20" North, 44° 11' 3" East. average temperature in Najran ranges from

14.6 to 30.9 °C and the average annual rainfall is 83 mm.

The animals

The animals used in this study came from different location of Najran to the abattoirs between August 2011 – August 2012. They were different species (camels, sheep and goats) after slaughtering the animals were examined for the presence of hydatid cyst in the livers and lungs. Any animal found infected with cyst was recorded. Furthermore, some cysts were examined under microscope to determine whether they are fertile or not and the fertility is assessed by the presence of protoscoleces. Most of the positive cases were examined to determine their age.

The human samples

Total of 1142 human serum sample were collected from patient admitted to different hospitals in Najran. The samples were kept frozen at -30°C until used.

Serological Survey

The sera were screened with an indirect haemagglutination assay (IHA) (Echinococcosis, Fumouze Laboratoires, France) and with ELISA (Hydatid Cyst (Echinococcus) 96 wells ELISA – Gentaur, France)

Experimental Procedures:

Experimental I was carried out to assess the prevalence of hydatidosis in sheep, goats and camels. A total of 48139 animals (camels = 4531, sheep= 29916, goats= 13692) were employed in this experiment. The animals were examined post-mortem for the presence of hydatid cysts. The animals having hydatid cyst in the liver or lung were recorded. Some hydatid cyst were photographed and hydatid fluid was aspirated from some cysts and transferred to test tube and centrifuged at 5000 r.p.m for 10 minutes and the sediment was examined directly under microscope (×10).

Experimental II was done to determine the prevalence rate of hydatidosis in citizens in Najran area. A total of 1142 human serum samples were collected and tested with ELISA (276 samples) and IHA (866 samples) for the presence of antibodies of hydatidosis (*Rebhandl et al., 1999)*. Some of negative (n= 57) and all the positive (n=35) samples of IHA were retested with ElISA as described by *(Timmreck, 1994).* The re-evaluation of the IHA test was done as follow:

Table 1.

	IHA TEST		Total
Results	positive	Negative	
Positive cases	True positive (T+) a	False positive (F+) b	a+b
Negative cases	false Negative (F-) c	True Negative (T-) d	c+d
Total	a+c	b+d	a+b+c+d

Accordingly the sensitivity, specificity and accuracy of IHA test was calculated as follow:

a. Sensitivity %= a / a + c ×100

b. Specificity %= d / b + d ×100

c. Accuracy %= a + d / a + b + c +d ×100

Statistical analysis:

The data were analyzed with Chi-$\times^{2.}$ Probabilities of $P<0.05$ were considered significant.

3. Result

Experiment I

The results of this experiment showed that the prevalence rate of hydatidosis in animals is 5.3%. As shown in **fig. (1)** the prevalence rate was significantly higher ($P<0.001$) in sheep than in camels and goats (n=2041, n=245, n= 309). The percentage of infection in sheep, camels and goats was 6.8%, 5.4% and 2.2% respectively. The infection of sheep was so severe that a single animal has many cysts in the lung and the liver **(Fig.2).** Most of the cyst obtained from sheep when examined microscopically were fertile **(Fig.3).**

Experiment II

As shown table (1) the overall of prevalence rate in human was 4.5%. When the sera were tested with ELISA the prevalence rate was high (6%) as compared with the prevalence rate measured with IHA (4%) with no significant difference (p >0.05). Furthermore when the positive (n=35) and negative (n=57) cases found in IHA results were retested with ELISA, 4 negative cases were found among the positive IHA and 4 positive case were found among the negative IHA cases. Therefore, sensitivity, specificity and accuracy of IHA were evaluated as 88.5 %, 92.9% and 91.3% respectively.

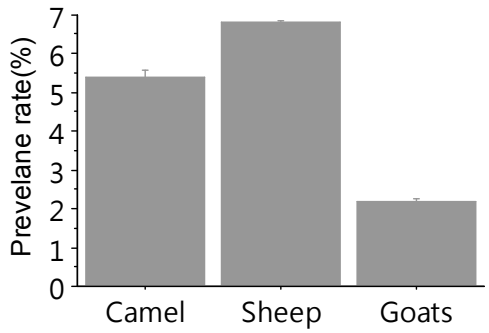
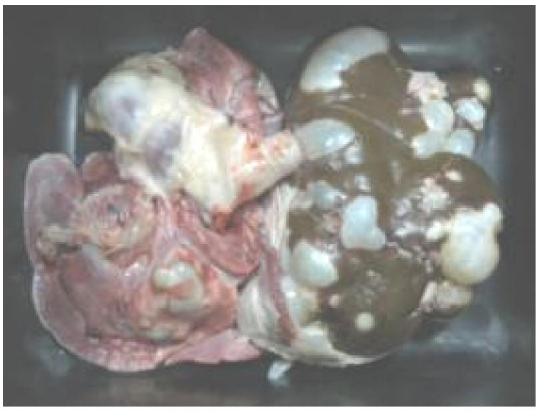


Fig. (1). The rate of prevalence of hydatidosis among farm animals in Najran area. [a,b,c] $p<0.00$



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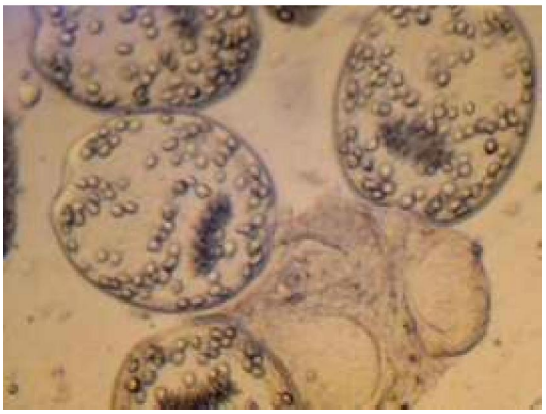

Fig. (3): Micrograph of a fertile hydatid cyst with a the clear hydatid sand

Table 2. The rates of hydatidosis in human as tested by ELISA and IHA.

Test	No. of samples	positive	Percentage
ELISA	276	17	6 %
IHA	866	35	4 %
TOTAL	1142	52	4.5 %

4. Discussion

The results of the present study indicated that hydatidosis prevails in Najran area both in human and animals. Furthermore, screening of the disease can be done serologically with ELISA and/or IHA.

Hydatid disease (*Echinococcus granulosus*) is endemic in the Middle East as well as in other parts of the world, including India, Africa, South America, New Zealand, Australia, Turkey and Southern Europe *(Goel et al., 1995).* In Mekka district, Saudi Arabia, the rate of prevalence of hydatidosis was recorded as 16% in camels (*Haroun et al., 2008).* In the present study the prevalence rate of hydatidosis in camels was 5.4%. The discrepancy between the two results is probably due the difference in animals age and that the camels slaughtered at Mekka abattoirs are brought from area outside of Mekka, either imported or from other areas of Saudia Arabia. Also the prevalence of infection in camels in Albaha was recorded as 32.85% *(Ibrahim, 2010)* which is extremely high compared to our findings. In Ibrahim study the incidence was high among old ages while in young ages was low. In the present study most of the camels slaughtered in Najran abattoirs were young. Usually the hydatid cysts increase gradually in size and number with age. Also Ibrahim reported a prevalence rate in sheep of 12.6% which is as twice as what we found (6.8%). In the goats he recorded a rate of 6.5% which is thrice our recorded rate (2.2%). The differences between the two studies could be attributed to environmental factors, management and the social behaviors. Where in Najran the camel meat consumers prefer the meat of camels calves.

Many cases of hydatidosis were diagnosed in pregnant women in the southwestern region of Saudi Arabia particularly in Abha *(Abu-Eshy and Elamin Ali, 1999).* Also *Hossain et al., 1985* found a rate of 57 % human Hydatidosis in Riyadh, Saudi by IHA. In the present study the prevalence rate among patients examined with IHA was 4% which is very low. However, when sera were examined with ELIZA the prevalence rate increased (6%). Probably this difference is due to the different kits used, localities and or the differences in standards of living which changed drastically between the eighties and the twenties. The improvement of the standards of livings and the increment of the awareness of zoonotic disease threat may contributed to this reduced rate of prevalence in Najran.

Many workers (*Jastaniah et al. ,1997; Abu-Eshy, 1998; Abu-Eshy and Elamin Ali, 1999; Alam, 1999; Adewunmi and Basilingappa, 2004; Hijazi; Al-Ansari, 2007)* also reported that hydatid disease is common in Asir, the southwestern region of Saudi Arabia.

Some of IHA results found in this study were tested versus ELISA to evaluate the sensitivity, specificity and accuracy of IHA. Some sample that gave false negative with IHA, gave positive results when tested with ELISA. This is probably due to infections with sterile and/ or calcified cysts. While the false positive results may have been caused by cross reaction with another parasites. The efficacy of IHA obtained in this result is similar to that obtained by *(Rajaii, 2005; Eris et al., 2009).*

In Najran hospitals the physicians used to diagnose hydatidosis with X rays, ultrasound, CT scan or MRI. The same methodology is done elsewhere *(Nunnari et al., 2012)*. The results of these techniques are usually limited because many cases of hydatidosis could be diagnosed as tumor growth or abscesses or vice versa.

In conclusion, hydatidosis prevails in Najran area at an overall rate of 5.3% in farm animals and 4.5% in human. Additionally, ELIZA and/or IHA can be used to screen and confirm the hydaitdosis in human.

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