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# Biochemical Profiles of Hydatid Cyst Fluids of Echinococcus Granulosus of Human and Animal Origin (Sheep, Goat, Cattle and Camel)

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**Abstract:** A comparative study on the biochemical parameters in hydatid cyst fluids of sheep, goat, cattle, camel and human cystic forms of Echinococcus granulosus have been made in Mazandaran. Hundred and twelve samples of hydatid fluids were collected from the liver cysts of different hosts (sheep, cattle, goat, camel and human) in slaughterhouses of sari and Ghaemshahr and Immam hospital respectively. All cyst fluids were centrifuged at 4500 rpm at 4°C for 45 min and the supernatants were analyzed for various biochemical parameters. Quantitative variations in the levels of Sodium, Glucose, Urea, Alanin Aminotransferase (AST), were found in the cystic fluids of different host origins although these differences were statistically insignificant. However, differences in the concentration of Potassium, Calcium, Triglycerides, Cholesterol, Uric acid, Creatinin, Albumin, Gamma glutamyl transferase, Aspartat Aminotransferase (AST) and Creatinine Phosphokinase (CPK), in different hydatid cyst fluids were statistically significant. Differences in the biochemical composition of different hydatid cyst fluids suggest existence of more than one strain of Echinococcus granulosus in human and other domestic animal intermediate hosts in Mazandaran.

**Key words:** Human, hydatid cyst, animals, biochemical compounds

## INTRODUCTION

Hydatidosis or *Ecchinococcosis* is the most important disease, common between human and cattle, which has worldwide spread. In our country, *Echinococcosis* has been found in more attention because of its significance in medicine and veterinary sciences. In the undeveloped countries, the people where live in villages are more vulnerable to *Echinococous* infection because of their direct contact with domestic and wild animals particularly with dog and dog family. In Iran, the contaminated vegetables and fruits among the citizen women and ashayer mostly spread this infection. Hydatidosis is a broad infection in animal and human which cause by *Echinococous granulosus* larva[1-3]. Hydatidosis is one of endemic diseases in this country and several domestic animals including sheep, goat, camel and cattle are the intermediate hosts of this infection[4]. Human infection cases are usually reported from different regions of Iran. For this reason, a significant percentage of surgery belongs to therapy of this infection in many hospitals Iran[5-7]. Different species and sub-species of *Echinococous* strain have been reported from much area where infection is endemic. Most of these reports give valuable information about the structure and natural specification of parasite. A collection of different sub-species exists in these areas, which indicated by the reports of researchers in recent years[8,9]. This variation definitely effects on the epidemiology, pathology, prophylaxis and finally control of this disease[9,10]. In addition, there has been evidence that some strain of the parasite is more infectious for man than others[11]. Therefore, study on this parasite assigns a significant position in research particularly, when there are several intermediate hosts and different transferable ways of infection to human in these regions[12]. Biochemical studies on the hydatid fluid's components has important role to determine the sub-species taxonomy of the parasite-causing cyst in Iran[13]. In 1991, Thompson and Lymbery suggested that the study on different species of parasite has significance epidemiology of hydatid cyst disease in a region[14].

Due to the lack of wide biochemical studies on the hydatid cyst in Iran[13,15], the present study proposed to determine the biochemical components of hydatid cyst fluid in different intermediate hosts including sheep, goat, camel, cattle and human in Sari and Ghaemshahr year 2004.

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## MATERIALS AND METHODS

Similar to other studies and in a descriptive manner, hundred and twelve samples of hydatid cyst fluid only isolated from liver organ were biochemically analyzed.

These sterile samples were collected from 16 sheep, 12 goats, 64 cattle and 10 camels from slaughterhouses in Sari, Ghaemshahr and 10 human samples were collected from Imam Hospital in Sari. Attempt was made to select only the active and fertilized cysts and the calcific or infected cases were removed from study. All cyst fluids were centrifuged at 4500 rpm at 4°C for 45 min and the supernatants were analyzed for various biochemical parameters. Supernatant solutions were used as fresh and free zed samples. The flame photometry method was utilized to measure the amount of $Na^+$ and $K^+$. Creatinin, $Ca^{2+}$ and albumin were measured by the colorimetric technique. The measurement of Glucose, cholesterol, Triglycerides (TG), urea and uric acid was made by the enzymatic methods. Three enzymes, *aspartate aminotransferase* (AST, GOT) (working base on converting of L-asp to OAA), *alanine transferase* (ALT, GPT) (working base on converting L-ala to pyr) and *lactate dehydrogenase* (LDH) (working base on converting pyr to lac or reverse) are measured by the quantity techniques IFCC and DGKC, respectively. All the kits were prepared from the two man chemistry and "zist chemistry companies in Iran. The statistic method of uni-variance test was employed to calculate and analyze the obtain database on their p values.

## RESULTS

The obtained data from biochemical analysis of hydatid fluid components is shown in the Table 1.

The amount of $Na^+$, TG, LDH and γ-glutamyl transferas (GGT) from sheep, $Ca^{2+}$ and ALT from goat, cholesterol and albumin from camel and finally Glucose, urea and creatinin from human cysts were found in higher quantity than other cases. However, AST from goat, Creatin Phosphokinase (CPK) from sheep and uric acid from human cyst showed elevated high levels.

## DISCUSSION

In the present study, biochemical components of hydatid cyst fluids isolated from different intermediate hosts (human, sheep, cattle, goat and camel) were analyzed and compared. This study could probably be helpful to determine sub-species of *Echinococous granulosus* parasite in the north regions of Iran. Of course, this should be followed with further bio-molecular evaluations in the future. The organic and in-organic compounds play a main role in physiology, metabolism and immunity functions of hydatid cyst[16-19]. The GGT enzyme is a trans-membrane protein, which transfers amino acids and short peptides from outside cell to inside via membrane[20]. In 1984, McManus and McPherson[21] reported that there are quantity differences in metabolism of hydatid cyst and its biochemical components because of different intermediate hosts. In 1991, Thompson[22] expressed that the growth of either a species or sub-species and its adoption in new media could cause metabolic activity changes in parasite. This study showed that the existence amounts of glucose, urea and ALT in different cysts fluid have no significant values but for $Ca^{2+}$ and $K^+$ showed significant values (Table 1). Sultan sheriff[17] and I.A. Shaafie *et al.*[23] reported no significant values for in-organic compounds, in contrast of this study. The amount of $Na^+$ has no significant value in different cysts indicated by these three reports. The amount of albumin showed no significant changes in different intermediate hosts but TG showed elevated high level in cyst fluid from sheep compare to others. I.A. Shaafie *et al.*[23] reported elevated high levels of proteins and TG in cyst fluid isolated from sheep compare to other intermediates[23] but on the other hand, Sultan Sheriff *et al.*[17] found no significant changes in cysts fluid isolated from human and sheep for protein and TG[24]. Also, I.A. Shaafie[23] reported elevated high level of uric acid in hydatid cyst fluid from human[23]. The present study found that the activity of GGT enzyme in cyst fluid from sheep is higher than the other intermediates and also, showed significant changes for this enzyme activity in cyst fluid of all studied

Table 1: Comparison of biochemical components of hydatid cyst in various infected intermediate host (±SD)
mmol $l^{-1}$

| Ca2+ | Na+ | K+ | Uric Acid | Triglyceride | Cholesterol | Glucose | Urea | Host |
|---|---|---|---|---|---|---|---|---|
| 10.25±3.9 | 133.3±9.6 | 7.52±1.1 | 0.26±0.15 | 52.87±32.15 | 16±10.66 | 54.75±31.4 | 36.52±5.46 | sheep |
| 16.66±3.72 | 125.16±6.01 | 6.66±3.72 | 0.20±0.002 | 6.83±6.36 | 11.33±5.35 | 32.5±9.89 | 36.12±4.4 | Goat |
| 9.00±.6.41 | 126.2±3.89 | 7.58±0.64 | 0.57±0.13 | 41.56±5.63 | 33.12±8.68 | 54.45±6.92 | 38.08±6.56 | Camel |
| 10.34±4.39 | 126.85±17.18 | 5.83±1.6 | 0.41±0.33 | 37.34±44.08 | 16.46±6.97 | 47.59±26.94 | 39.87±10.04 | Cattle |
| 15.72±3.72 | 121.9±7.33 | 5.28±.69 | 0.87±0.12 | 16.03±1.48 | 27.86±7.28 | 64.76±8.68 | 51.14±6.99 | human |
| | Creatinin (μmol $^{1-1}$) | Albumin (g $L^{-1}$) | GGT(IU $L^{-1}$) | AST(IU $L^{-1}$) | ALT(IU $L^{-1}$) | CPK(IU $L^{-1}$) | LDH(IU $L^{-1}$) | Host |
| | 0.38±0.19 | 0.73±0.31 | 55.87±3.90 | 11.37±7.60 | 11.37±10.67 | 72.25±57.90 | 58.12±35.44 | sheep |
| | 0.30±0.15 | 0.30±0.15 | 50±2.60 | 35.50±29.75 | 35.50±29.75 | 24.66±10.36 | 45.66±26.7 | Goat |
| | 1.09±0.34 | 0.80±0.04 | 44.06±3.91 | 3.84±0.61 | 3.84±0.61 | 12.42±1.73 | 13.16±3.21 | Camel |
| | 0.36±0.13 | 0.71±0.27 | 46.25±0.55 | 11.81±9.16 | 11.81±9.16 | 27.12±21.41 | 39.59±25.21 | Cattle |
| | 1.13±0.41 | 0.70±0.11 | 42.42±3.47 | 5.7±2.41 | 12.5±4.51 | 19.52±3.19 | 17.08±3.31 | human |

$p<0.05$, $p<0.001$

intermediates. The lower activity of this enzyme in isolated cysts fluid from other intermediates compare to sheep's apparently, is due to the existence of low level of albumin. However, it is difficult to determine the nature and source of this enzyme in hydatid cyst fluid. Anyway, a relationship would apparently be between the level activity of this enzyme which produced by cyst generative layer membrane and growth and developing of hydatid cyst. Also, in this research, some biochemical components of isolated cyst from human showed similarity to those isolated from sheep and camel. This could be concluded that one or more species of *Echinococous granulosus* exist in common between these hosts in these regions of Iran. Dalimi *et al.*[25] showed that there was a similarity between some biochemical components of isolated cysts from human, sheep and camel but although, there was a profound differences among some other components isolated from these hosts.

They employed the PCR technique for their research and they concluded that at least, two strains of *Echinococous granulosus* apparently exist in Iran, which they can make human, infected. In china, Bowles and McManus[26,27] isolated a sheep's species of *Echinococous granulosus* that also, cause infection in human, cattle, camel and pig. In 1990, Lymbery[28] and Bowles and McManus[29] reported to found an isolated form of *Echinococous granulosus* common between human, sheep, cattle and pig. In Africa and Middle East including Iran, camels usually get infected. Although, in McManus *et al.*[29] also, Eckert *et al.*[30], Wachira *et al.*[31] suggested no common form of *Echinococous granulosus* between human and camel but McManus and Rishi[32] reported an isolated camel's form, common between this host and other domestic animal hosts[32]. Regarding, the data in this research and existence of some biochemical components differences between human and other animal intermediate hosts. Apparently, there must be more than one strain of *Echinococous granulosus* in Mazandaran province. Therefore, further biochemical, pathologic, parasitological and particularly, bio-molecular studies must be complete to determine these strains in Mazandaran province.

## REFERENCES


1. Craig, P.S., M.T. Rogan and J.C. Allan Detection, 1996. Screening and Community epidemiology of Taeniid cestode Zoonoses: Cystic *Echinococcosis,* Alveolar *Echinococcosis* and neurocysticercosis. Adv. Parasitol, 38: 169-250.
2. Craig, P.S., D. Liu, C.N.L. Macpherson and S. Dazhong *et al*., 1992. A large focus of alveolar *Echinococcosis* in central China. Lancet., 340: 826-831.
3. Marie-France Biava, M.C.U.P.H. and M.D. Anne Dao, 2001. Laboratory Diagnosis of Cystic Hydatid Disease. World J. Surg., 25: 10-14.
4. Dalimi, A. and I. Mobedi, 1997. Helminth parasits of Carnivores in northern Iran. Ann. Trop. Med. Parasitol., 86: 395-397.
5. Moghaddar, N., A. Oryan and M.R. Hanife Paur, 1992. Helminthes recoverd from the liver and lung of camel with special references to their incidence and pathogenesis in Shiraz, Islamic Republic of Iran. Indian j. Anim. Sci. , 62: 1018-1023.
6. Oryan, A., N. Moghaddar and S.N.S. Gaur, 1994. Metacestodes of Sheep with special reference to their epidemiological status, pathogenesis and economic implication in Fars province, Iran. Vet. Parasitol., 51: 231-240.
7. McManus, D.P. and J.D. Smyth, 1986. Hydatid disease (hydatidosis) change concept in epidemiology and speciation. Parasitol. Today, 2: 163-168.
8. Bowles, J. D. Blair and D.P. McManus, 1992. Genetic Species within the genus *Echinococous* identified by mitochondrial DNA sequencing. Mol. Biochem. Parasitol., 54: 156-174.
9. Bowles, J. and D.P. McManus, 1993. Rapid discrimination of *Echinococous* species and strains using a PCR-based RELP method. Mol. Biochem. Parasitol., 57: 231-239.
10. Bowles, J. D. Blair and D.P. McManus, 1994. Molecular genetic characterization of the Cervid strain (northern form) of *Echinococous granulosus*. Parasitol., 109: 215-221.
11. Thompson, R.C.A., 1995. Biology and Systematics of *Echinococous*. In Thompson, R.C.A. and Lymbery, A.J. (Eds) *Echinococous* and hydatid disease. Wallingford, CAB Intl., pp: 33-49.
12. Eslamirad, Z., A. Deylami Asl and A. Moubedi, 2000. comparison of protein and lipid levels in hepatic Hydatid Cyst Fluid isolated from sheep, cattle and goat. Journal of Ghazvin Medical Science University, 11: 22-26.
13. Thompson, R.C.A., A.J. Lymbery and C.C. Constantine, 1994. Variation in *Echinococous* towards a taxonomic revision of the genus. Advances in Parasitology, 35: 145-176.
14. Vatankhah, A. and S. Rohani, 2003. Quantity Evaluation of Enzymes in Sterile and Fertilized Hydatid Cysts of *Echinococous granulosus* in sheep, Journal of Health Faculty and Institute of Hygienic Research, 3: 49-55.
15. Smyth, J.D., 1977. Strain differences in *Echinococous granulosus* with special reference to the status of equine hydatidosis in the United Kingdom. Transactions of the Royal Society of Tropical Medicine and Hygiene, 71: 93-100.

16. Frayha, G.J. and R. Haddad, 1980. Comparative chemical composition of protocolices and hydatid cyst fluids of *Echinococous granulosus*. Intl. J. Parasitolo., 10: 359-364.
17. Sultan Sheriff, D., F.K. Dar and S.A. Kidwai, 1984. Metabolic elements in hydatid fluids. J. Helmintolo., 58: 335-336.
18. Richards, K.S., E. Iderton and H.J. Yardley, 1987. Lipids in the laminated layers of liver, lungs and daughter hydatid cysts of equine *Echinococous granulosus* (Cestoda).,? Comparative Biochem. and Physiolo., 86B: 209-212.
19. Chowdhury, N. and R. Singh, 1993. Distribution of some elements in hydatid cysts of *Echinococous granulosus* from buffalo (Bubalus bubalus). J. Helmintholo., 67: 112-114.
20. Rambabu, K. I.A. Shaafie, A. Ansari S.A. Basha and M.M. Ziu, 1991. Studies on γ-glutamyl transpeptidase in primary idiopathic hypothyroidism patients. Biochem. Med. and Met. Biolo., 46: 140-144.
21. McManus, D.P. and A.K. Rishi, 1987. Genetic heterogenecity within *Echinococous granulosus* isolates from different hosts and geographical areas characterized with DNA markers. J. Helminth Zoonoses, 24: 29-36.
22. Thompson, R.C.A., 1991. *Echinococous* and Giardia: Variation on a theme. Intl. J. Parasitology. 77: 75-82.
23. Shaafie, L.A., A.H. Khan and K. Rambabu, 1999. Biochemical profiles of hydatid cyst fluids of *Echinococous granulosus* of human and animal origin in Libya. J. Helmintology., 73: 255-258.
24. Sultan Sheriff, D., M. EI-Fakhri and S.A. Kidwai, 1989. Lipids in hydatid fluids collected from lungs and liver of sheep and man. J. Helminthol., 63: 266-268.
25. Dalimi, A., N. Ahmadi and M. Sadeghizadeh, 2000. Molecular Evaluation of Hydatid Cyts Isolated from Human, Sheep and camel hosts by PCR Technique, Medical Sci. J. Tarbieat Modares, 2: 53-56.
26. Bowles, J. and D.P. McManus, 1993. Rapid discrimination of *Echinococous* soecies and strains using a PCR-based RELP method., Molecular Biochemistry and Parasitology, 57: 231-239.
27. Bowles, J. and D.P. McManus, 1993b. Molecular variation in *Echinococous*, Acta Tropica, 53: 291-305.
28. Lymbery, S.M., R.C.A. Thompson and R.P. Hobbs, 1990. Genetic diversity and genetic differentiation in *Echinococous granulosus* from domestic and sylvatic hosts on the mainland of Australia., Parasitology, 101: 283-289.
29. McManus, D.P., A.J.G. Simpson and A.K. Rishi, 1987. Characterization of the hydatid disease organism *Echinococous granulosus* from Kenya using cloned DNA markers. J. Helminth Zoonoses, 24: 29-36.
30. Eckart, J., R.C.A. Thompson, S.A. Michael, L.M. Kumaratilake and H.M. EI-Sawah, 1989. *Echinococous granulosus* of camel origin: development in dogs and parasite Morpholo. Parasitology Research, 75: 536-542.
31. Wachira, T.M., J. Bowles, E. Zeyhle and D.P. Mcmanus, 1993. Molecular examination of the sympatry and distribution of sheep and camel strains of *Echinococous granulosus* in Kenya, American J. Trop. Med. and Hygiene, 48: 473-479.
32. Mcmanus, D.P. and A.K. Rishi, 1989. Genetic heterogenecity within *Echinococous granulosus* isolates from different hosts and geographical areas characterized with DNA probe., Parasitology, 99: 17-29.