

Biochemical Characterization of *Cysticercus Tenuicollis* in Iranian Fat-tailed Sheep

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Abstract: The present study was designed to evaluate the biochemical profiles of *C. tenuicollis* in sheep. 36 cysts were collected from Iranian fat-tailed sheep slaughtered at the main abattoir of Shiraz (Southwest Province, Iran). Cysticerci were attached to the omentum, mesenteries and liver and transferred to the laboratory. All cyst fluids were centrifuged separately and the supernatants were analyzed for various biochemical parameters. Various biochemical parameters (glucose, total protein, urea nitrogen, uric acid, triglycerides, cholesterol, creatinine, calcium, sodium and potassium) were measured using conventional laboratory methods. The concentrations of glucose, triglyceride, cholesterol, total protein, urea nitrogen, creatinine, uric acid, Na⁺, K⁺, Ca²⁺ and phosphorous in cyst fluid were 6.265± 0.792 mmol/L, 0.777± 0.120 mmol/L, 0.167 ± 0.031 mmol/L, 3.18± 0.03 g/L, 15.407 ± 1.160 mmol/L, 71.957 ± 14.055 µmol/L and 33.147 ± 8.926 µmol/L, 115.71 ± 6.40 mmol/L, 9.357 ± 0.984 mmol/L, 2.307 ± 0.495 mmol/L and 0.867 ± 0.194 mmol/L respectively. The activities of AST and ALT in cyst fluid were 152.39 ± 48.94 and 14.29 ± 3.63 U/L respectively.

Key words: Biochemical parameters, *Cysticercus tenuicollis*, Iranian fat-tailed sheep

INTRODUCTION

Livestock may act as the intermediate hosts for the tapeworms of humans and other animals. Cestodes of the family *Taeniidae* which infect the dog (definitive host) are transmitted to a range of intermediate host species where they cause echinococcosis, cysticercosis or coenurosis (Flisser *et al.*, 1982; Eckert *et al.*, 1984; Thompson and Lymbery, 1995). The larval tapeworms (metacestodes) develop as fluid-filled cysts, each at a typical site in the body. They act as space-occupying lesions and cause condemnation at meat inspection (Radoszits *et al.*, 2007). The cysticercus of the canine taeniid tape worm *Taenia hydatigena* migrates through the liver tissue and encysts on the peritoneal membranes of cattle, sheep, swine, and certain wild ungulates. Massive invasions, such as when entire tapeworm segments are ingested, result in acute traumatic hepatitis, and even small numbers of migrating *T. hydatigena* larvae are capable of precipitating black disease in the presence of *C. novyi* (Bowman *et al.*, 2003).

Iran is considered as one of the endemic areas of *T. hydatigena* in dogs and wild carnivores as final hosts, and livestock and wild herbivores as intermediate hosts. Infection rate with *Cysticercus tenuicollis* in Iran was 12.87% in sheep (Radfar *et al.*, 2005). Morphological, immunological, physiological and some biochemical variation have been described in some Taeniid metacestodes, including *T. taeniaeformis*, *T. crassiceps*, *Hymenolepis diminuta* and *Echinococcus spp* (Hustead *et al.*, 1977; Mills *et al.*, 1983; Rosen *et al.*, 1994; Radfar *et al.*, 2004; Vinaud *et al.*, 2007). There has been no work on the biochemical aspects of *C. tenuicollis* in Iran. The present study was designed to evaluate the biochemical profile of *C. tenuicollis* in Iranian fat-tailed sheep.

MATERIALS AND METHODS

36 Cysticerci of *T. hydatigena* were collected from the omentum, mesenteries and livers of infected Iranian fat-tailed sheep slaughtered at the main abattoir of Shiraz, Southwest of Iran. The cysts were transferred to the parasitology laboratory and their fluids were aspirated aseptically, centrifuged at 15000 rpm at 4 °C for 30 min, and the supernatants analyzed for various biochemical parameters including glucose, total protein, urea nitrogen,

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uric acid, triglycerides, cholesterol, creatinin, sodium, potassium, calcium, phosphorus, AST and ALT. The biochemical parameters and used methods were as follows: glucose by the glucose oxidase method; total protein by the Biuret method; urea nitrogen (UN) by diacetyl monoxim method, uric acid by the phosphotungstic acid (PTA) method; triglycerides by the enzymatic procedure of McGowan *et al.* (1983); cholesterol by a modified Abell-Kendall/Levey-Brodie (A-K) method, creatinine by Jaffe method, phosphorus by ammonium molybdate method and aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities by the colorimetric method of Reitman and Frankel (Burtis *et al.*, 2006). The concentration of sodium and potassium were measured using flame photometry (FLM2, Bach-Simpson Ltd, Ontario, Canada). Samples were analyzed for calcium by atomic absorption spectroscopy (Shimadzo AA-670, Shimadzu Corporation, Kyoto, Japan).

RESULTS AND DISCUSSION

The biochemical parameters of *Cysticercus tenuicollis* in Iranian fat-tailed sheep are presented in Table 1.

Qualifying information on the nutritional state of the host at the time of necropsy is necessary for data analysis of the chemical profiles of parasitic worms. In animal parasites, the environment of the infectious stages is the host. A more important route of macromolecules absorption in immature stage is endocytosis, though supporting data come only from studies on *Taenia crassiceps*. The carbohydrate content has variations based on dry mass, and total glycogen content may vary in the range of 6-48 % of the dry mass. Larval cestodes generally show a more constant glycogen content than the corresponding adults; this may reflect the more stable intermediate host environment, usually the coelomic cavity or tissues. Our results showed that the concentration of glucose in the cyst fluid of *Cysticercus tenuicollis* was 6.265 ± 0.792 mmol/L. Investigations showed that tapeworm and metacestode parenchymal tissues contain numerous gap junctions to uptake glucose (Lumsden and Hildreth, 1983; Conn, 1993; Willms *et al.*, 2003). Glucose is absorbed against a steep concentration gradient by Na⁺ dependent glucose transport system in mammals and many other organisms. This transport system is found in the external cyst wall of *T. solium* neurocysticerci and also in the apical membrane of the tegument of adult worm.

The concentrations of triglyceride and cholesterol in the cyst fluid of *Cysticercus tenuicollis* were 0.777 ± 0.120 and 0.167 ± 0.031 mmol/L, respectively.

Sultan Sheriff *et al.* (1989) measured the levels of lipids in hydatid cyst fluid and showed the presence of phospholipids, cholesterol and glycerides from which the triacylglycerol and diacylglycerol are major lipids. Mills *et al.* (1981) reported the highest proportions at the earlier stages. The distribution was as follows: neutral lipid 27-45%; glycolipid 5-11%; and phospholipids 50-61%. The major neutral lipid was cholesterol and minor neutral lipids were sterol esters, triglycerides, diglycerides and monoglycerides. It has been showed that lipid accounts for 21% of the dry weight of the parasite at week 3, drops to 7% by week 7 and remains at this percentage throughout the rest of the larval growth of *Taenia taeniaeformis*. It is probable that these lipids play a vital role in the establishment and development of the parasite (Mills *et al.*, 1983).

The concentration of total protein in the cyst fluid of *Cysticercus tenuicollis* was 3.18 ± 0.03 g/L. Protein content depends on the age, degree of maturation and previous metabolic history of the worm. Some studies have been carried out on larval *E. granulosus* (Agosin and Repetto, 1967) and larval *T. crassiceps* (Naquira *et al.*, 1977). Immunoprecipitation (Shepherd and McManus, 1987) and immunoblot analysis (Shapiro *et al.*, 1992) have confirmed the presence of several host proteins, including serum albumin and immunoglobulins in hydatid cyst fluid. Host protein may enter the hydatid cyst by diffusing through fissures in the cyst membranes, by endocytosis or by specific filter or transport mechanisms. The ionic nature of proteins may also play an important role in their own absorption (Smyth and McManus, 1989). Radioiodinated proteins were taken up in vitro by larvae of both *T. taeniaeformis* and *T. crassiceps* and were shown to retain their physicochemical and antigenic characteristics. Rates of uptake were similar in the 2 species and were not related to the molecular weight of the proteins. Taeniid metacestodes are capable of absorbing a variety of proteins, and these macromolecules can retain their structural and functional integrity following transport. This absorptive capacity accounts for the presence of host serum components within bladder fluids (Hustead and Williams, 1977). The concentrations of urea nitrogen, creatinine and uric acid in the cyst fluid of *Cysticercus tenuicollis* were 15.407 ± 1.160 mmol/L, 71.957 ± 14.055 μ mol/L and 33.147 ± 8.926 μ mol/L respectively. The concentration of minerals such as Na⁺, K⁺, Ca²⁺ and phosphorous in the cyst fluid of *Cysticercus tenuicollis* were 115.71 ± 6.40 , 9.357 ± 0.984 , 2.307 ± 0.495 and 0.867 ± 0.194 mmol/L respectively.

The activities of AST and ALT in the cyst fluid of *Cysticercus tenuicollis* were 152.39 ± 48.94 and 14.29 ± 3.63 U/L respectively. Aspartate transaminase (AST) is a cytoplasmic and mitochondrial enzyme that catalyses the transamination of L-aspartate to oxaloacetate and glutamate, and AST activity is found in almost all cells. Alanine Transaminase (ALT) is a cytoplasmic enzyme that catalyzes the reversible transamination of L-alanine and 2-oxoglutarate to pyruvate and glutamate (Burtis *et al.*, 2006). Sanchez and Sanchez (1971) and Radfar and Iranyar (2004) carried out a very comprehensive comparative study on hydatid fluid from cyst of human, sheep, cattle and camel origin. Many of the components were different qualitatively or quantitatively, depending on the cyst location and host origin, possibly reflecting strain characteristics.

Table 1: Biochemical parameters of *Cysticercus tenuicollis* in Iranian fat-tailed sheep (n=28).

Parameter	Min	Max	Mean	S.E
Glucose mm. L ⁻¹	0.222	16.206	6.265	0.792
Urea nitrogen mm.L ⁻¹	4.984	32.752	15.407	1.160
Creatinin µm.L ⁻¹	0.0	309.40	71.957	14.055
Uric acid µm.L ⁻¹	0.0	142.826	33.147	8.926
Triglyceride mm. L ⁻¹	0.0	2.386	0.777	0.120
Cholesterol mm. L ⁻¹	0.0	0.775	0.167	0.031
Total protein g.L ⁻¹	1.0	7.0	3.18	0.03
AST U. L ⁻¹	2.0	972	152.39	48.94
ALT U. L ⁻¹	0.0	79	14.29	3.63
Na mm.L ⁻¹	6.0	165	115.71	6.40
K mm.L ⁻¹	0.6	18.5	9.357	0.984
Ca mm. L ⁻¹	0.473	15.189	2.307	0.495
Phosphorus mm.L ⁻¹	0.032	3.392	0.867	0.194

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