



Diagnostic Study on accidental *Hymenolepis diminuta* Infection in Laboratory Rats in Iraq

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Abstract

Rats are the most common laboratory animals used in

biological testing and research. They are seldom examined for native ecto- and endoparasites before their usage in the experiments. Parasitic infestation is one common problem in the laboratory animals, which can change the interpretation of final results. In this study, high accidental infection with *Hymenolepis diminuta* (*H. diminuta*) was reported in experimental *Sprague-Dawley* male rats. The diagnosis was based upon the viewing of numerous tapeworms attached to the intestinal wall, and the presence of *H. diminuta* characteristic eggs in the intestinal content. The authors suggest to pay special attention for this zoonotic tapeworm in contact animal attendants. In addition, extended survey and investigations need to be done for all units of laboratory animals, to assure the healthy environmental conditions and the well-being needed to the animals used in experimental trials. An educational control programs are also need to implement in the animal houses.

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Introduction

Understanding the better knowledge of physiological and pathological processes in human and animal can be achieved by using the living laboratory animals in experimental biomedical studies (Tanideh *et al.*, 2010). Laboratory rats are the most commonly used species as animal models of diseases in medical, biomedical and veterinary research. The well-being of the experimental rats throughout their life is one of the important factors that give correct value of the research models. Therefore, inadequate and unhealthy environmental conditions would subside the

animals' well-being of the laboratory animals and lead to affect negatively on the experimental results (Casebolt *et al.*, 1988; Bicalho *et al.*, 2007). There are about (150-200) zoonotic infectious agents that may be transferred from laboratory animals to human, including leptospirosis, salmonellosis, tuberculosis, lymphocytic choriomeningitis, hemorrhagic fever and rat bite fever. Moreover, various parasitic worms such as *H. diminuta*, *Hymenolepis nana*, *Schistosoma*, and *Trichinella* are incriminated (Huq *et al.*, 1985; Gilioli *et al.*, 2000).

H. diminuta is a rat tapeworm that occurs throughout the world. Arthropods, specifically beetles, Tenebrionidae family, is the necessarily intermediate host in the completion the life cycle of *H. diminuta*. Rodents are the principal definitive hosts of *H. diminuta*. Even though, in rare situations, it can infect humans, when by incidental ingestion of infected arthropods, cysticercoids move and invade the small intestine (King, 1995). *H. diminuta* infection in the rat can be associated with slow growth and pot-bellied syndrome (Owen, 1992). Variation in the prevalence of *Hymenolepis* in brown rats has been reported from UK (Webster and Macdonald 1995), Iran (Sadjjadi and Massoud, 1999), India (Somvanshi, 1997) and Jamaica (Waugh *et al.*, 2006). *H. diminuta* was reported in wild rodents from several studies in Iraq. It is reported in Basra (Al-Hadithi *et al.*, 1985; Al-Zihiry, 2002), Baghdad (Mahmoud, 1974; Jawdat and Mahmoud, 1980; Al-Barwari *et al.*, 1987; Al-Zahidy, 2001), Erbil (Hussein, 1986; Molan *et al.*, 1988), Mosul (Salih, 1975) and in Hilla city (Al-Morshidy, 2001)

Regular evaluation of the laboratory animals is necessary to control the healthy condition and to investigate parasites, viruses, bacteria, and fungi, moreover to recognize the genetic disorders. In Iraq, information on the incidence of *H. diminuta* in rat is not available. This study aims to report the unintentional incidence of *H. diminuta* in the rats reared at the unit of laboratory animals / Faculty of Veterinary Medicine / Kufa University and to describe its macroscopic and microscopic pathological changes.

History of cases

Four adult Sprague-Dawley male rats weighing 180 to 250 g were used as experimental animals to study the effects of one herb plant. They were obtained from the unit of Laboratory Animal / Faculty of Veterinary Medicine (FVM), Kufa University (KU), Iraq. The rats were grouped randomly into two groups, and maintained under standard laboratory conditions. Free access to the water ad libitum and standard dry pellet diet were allowed. The experimental and control rats were orally gavaged with herb extract and distilled water respectively. At the end of the experiment, the rats were subjected to necropsy investigation following a humanely euthanasia using chloroform. Visible gross lesions were recorded. After necropsy examination, tissue pieces from different visceral organs (liver, spleen, kidney, pancreas, and lymph nodes) and intestine positive for *H. diminuta* fixed in situ were preserved in 10 percent formalin for histopathological evaluation. Standard histopathological and Haematoxylin and Eosin (H&E) technique were performed according to method described by Luna *et al* 1968 and Culling *et al.*, 1985. All the procedures described were reviewed and approved by the Kufa

University Research and Animal Ethical Committee (KU.FVM.AEC number 0706-2013).

Results

In the present investigation 3/4 (75%) rats were found to be infected with tapeworms. The infected rats revealed 5- 8 adult worm that were attached on the intestinal wall. The intestinal contents of the infected animals were examined under the microscope to identify the eggs. It revealed numerous tapeworm eggs, which were appeared as globular oval shaped. The embryophore were appeared with thick rudimentary poles (Figure 1.), however the space between membranes appeared faintly granular.

Grossly, whitish-yellow structures were seen through the intestinal wall before cutting the intestine wall. Attached worms length 15-45 cm were seen in different location of the intestine. The rats revealed mild to moderate enteritis with grossly visible congestion, which were also observed in the other visceral organs. The intestinal lumen of the infected rats was dilated due to presence of multiple tapeworms (Figure 2 A, B, C).

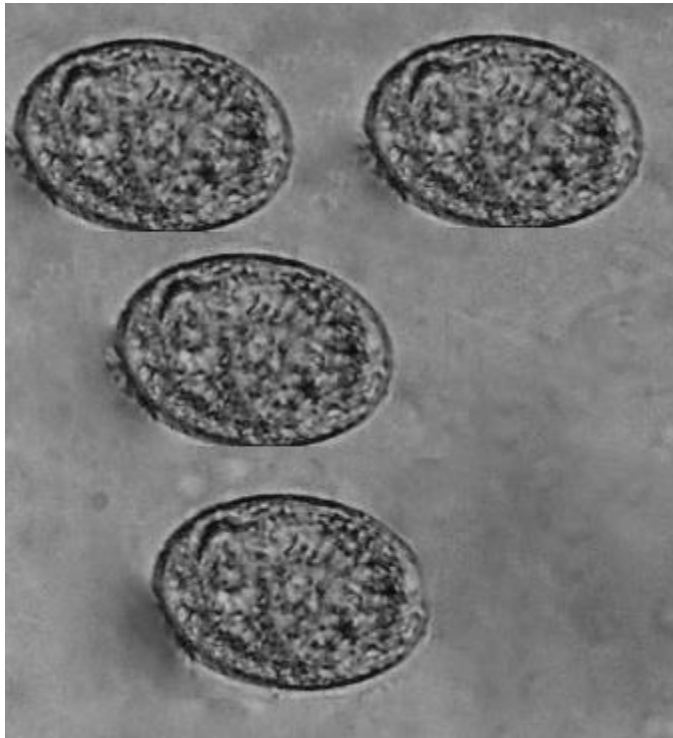


Figure 1. Shows the globular oval shaped eggs of *H. diminuta* isolated from the intestinal contents. (X100)

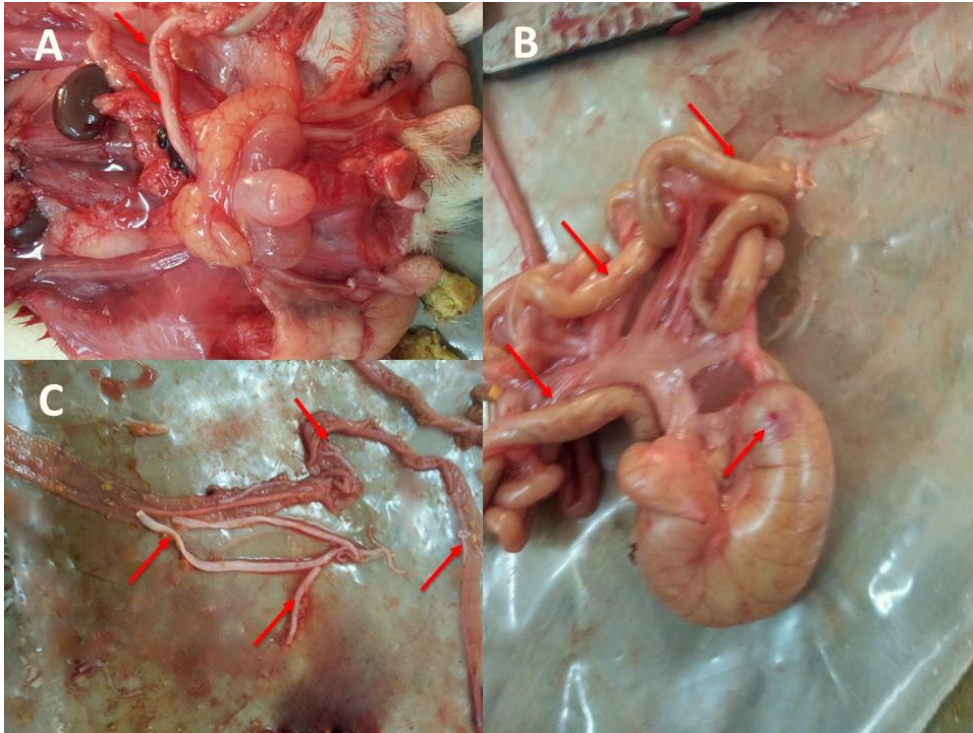


Figure 2. A & B Adult tapeworm (*Hymenolepis diminuta*) visible through the serosa of intestine of rat, C, Adult tapeworm (*Hymenolepis diminuta*) attached on the intestinal wall.

Microscopical examination of the infected small intestine revealed indistinguishable tapeworm segments of *H. diminuta*. The segments showed serrated borders and uterus, eggs, testes and other visceral organs in its centre. Some *H. diminuta* eggs and scolex were seen attached with the mucosa.

The intestinal sections also showed degeneration and desquamations of lining mucosal epithelial cells and excessive mucin secretion were mixed with the luminal debris. Atrophy in the intestinal villi also was seen. Eosinophilic cellular infiltration of the intestinal mucosa were frequently seen. Moreover, mononuclear cellular infiltration were also observed in the red pulp in spleen, kidneys and lung.

Dilatation of sinusoids and presence of destructive alterations in the hepatic parenchyma, were appeared in the infected rats in addition to, infiltration of mononuclear cell and necrotic cells with pyknotic nuclei. These necrotic cells were grouped in foci.

Discussion

Rodents are the definitive host for *H. diminuta*, which has a worldwide distribution. *H. diminuta* infects the small intestine of rats and mice. Human infection is rare and may occur by accidental ingestion of infected arthropods harbouring cysticercoids, infective larvae of the parasite (King, 1995; Schantz, 1996). In present study, the

75 percent of *H. diminuta* incidence, is in accordance with reports of other workers (Sadjjadi and Massoud, 1999; Somvanshi, 1997; Webster and Macdonald, 1995). Previous study mentioned that wild rats are the important source of *H. diminuta* in laboratory animals (Goswami *et al.*, 2011). They found that *H. diminuta* is commonly appeared in areas, where large amount of favorite foods for wild rats (dry feed products) are stored. In this study, the dry food was stored in the room near to the animal's unit. Moreover, there were many wild rodents that commonly entered through the holes in the wall. Therefore, the possible source of *H. diminuta* infection, was the visiting of the infected wild rats looking for food inside the unit of laboratory animals. Researchers are believed that the high infection rate is indicative of poor hygiene in the animal's unit. The prevalence *H. diminuta* and *H. nana* was detected in brown rats as 7 and 0 percent in Belgium (Cotteleer *et al.*, 1982) and 11 and 22 percent in UK (Webster and Macdonald, 1995). However, relatively low prevalence rate (3.8 %) of *H. diminuta* was reported in Jamaica in two species of wild rats (Waugh *et al.*, 2006), whereas, in India, high incidence rates were reported (39.6% and 27%) in laboratory and Wild rats, respectively (Somvanshi, 1997). High incidence of *H. nana* 31.3% and *H. diminuta* 12.5%, was also recorded in wild rodents from Khuzestan, South-West Iran (Sadjjadi and Massoud, 1999). In the present accidental investigation of high infection of *H. diminuta* in laboratory rats, the attention needs to be directed towards the possibility of zoonotic nature in contact animal attendants. *H. diminuta* are reported in human beings and causes diarrhea and abdominal pain in heavy infections while in rodents this infection can be associated with slow growth and pot-bellied syndrome.

In this study, multiple numbers of tapeworms were attached on the walls of the intestine. The confirmed diagnosis was based on the examination of the intestinal content and presence of *H. diminuta* characteristics eggs. Previous workers (Arai, 1980; Soulsby, 1969) reported that intestinal contents samples revealed 70 micron diameter, spherical eggs, with a striated outer membrane and a thin inner membrane and containing six central hooklets but no polar filaments (rudimentary) and differentiated from *H. nana* eggs, which have a similar appearance but are smaller and have two evident polar thickenings, from each of which arise four to eight polar filaments.

The present histopathological findings are in agreement with other previous report (Somvanshi, 1997). Excessive mucin secretion and desquamation of epithelial cells may be due to irritation caused by serrated border of segments of tapeworm. Enteritis in rats associated with *H. diminuta* was also observed in previous study (Sriram *et al.*, 1980). Looking for the high incidence of *H. diminuta* and its zoonotic nature for man, extended survey and investigations need to be done for all units of laboratory animals. In addition, educational control programs are also need to implement in the animal houses.

In conclusion, the high incidence of *H. diminuta* was found accidentally in the rats reared at the unit of laboratory animals. Gross and conventional intestinal contents examination were better methods in diagnosis of *H. diminuta* infection. *H. diminuta*

infection in laboratory rats needs special attention as zoonotic concern in contact persons.

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