

ORIGINAL ARTICLE

Hydatidosis: The Time Required for Implantation of Protoscolices in the Peritoneal Cavity of Rats

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ABSTRACT

Background: Hydatid disease is a parasitic infection caused by the larval stage of *Echinococcus granulosus* (*E. granulosus*), whose life cycle involves livestock and canids as intermediate and definitive hosts, respectively. Surgery is a common treatment for hydatid cysts, but cyst rupture and spillage of the contents may cause secondary implantation of protoscolices. The objective of this study was to determine the time required for implanting protoscolices in various organs.

Materials and Methods: In this experimental study, thirty-six rats were divided into six groups (n=6 in each group). The rats in five groups were intraperitoneally injected with 2 ml of hydatid fluid containing 100 protoscolices, but the rats in the sixth group (as the control group) were injected with 2 ml of normal saline.

The rats in groups I, II, III, IV, and V were sacrificed and opened after 1, 24, 48, 72, and 168 hours, respectively. All the internal organs were evaluated carefully and three samples from the liver, omentum, and intestine were prepared for optical microscopy.

Results: There were no protoscolice implantations in the tissues of the first, second, and third groups of rats. However, protoscolices were implanted in tissue samples of the fourth and fifth groups that were sacrificed 72 and 168 hours after injection.

Conclusion: The protoscolices of hydatid cyst require at least 72 hours for implantation after intraperitoneal spillage.

Key Words: Echinococcosis, Intraperitoneal injection, Rats

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INTRODUCTION

Hydatid disease is a parasitic infection caused by the larval stage of *Echinococcus granulosus* (*E. granulosus*) [1]. The parasite's domestic life cycle involves livestock and canids as the primary intermediate and definitive hosts, respectively. Canids harboring adult *E. granulosus* worms excrete eggs into the environment, where intermediate hosts become infected through ingestion of the eggs. Humans can also act as aberrant intermediate hosts if they ingest the infective parasite ova either through contaminated food or directly from an infected canid. After entering the body, cystic larva are gradually formed in different organs of the intermediate host [2].

Because of the slow progression of the

disease, it might be asymptomatic initially or with very little clinical manifestations [3]. In numerous parts of the world, including Iran, surgical approach is a common treatment of hydatid cysts. However, all surgical techniques are invasive and pose risk of recurrence. Rupture of cyst and spillage of the contents may cause anaphylactic shock and secondary cyst formation [2].

Hydatid disease is endemic to Iran, where several species of the intermediate host are commonly infected with *E. granulosus* [4]. Human hydatid disease is regularly reported from medical centers in different parts of Iran and its incidence is estimated at 1.18-3 per 100,000 cases [5, 6].

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Despite the developments in surgical treatments for hydatid disease, we still face critical challenges, and determining the time required for implanting protoscolices in other organs after perforation of hydatid cyst is of utmost importance. Effective administration of prophylactic treatments also depends on such findings.

Accordingly, the objective of this study was to determine the time required for implanting protoscolices in the abdominal cavity of rats.

MATERIALS AND METHODS

In this experimental study, hydatid fluid was aspirated from human hydatid cyst under sterile conditions and was left to settle for 30 min. The sedimented protoscolices were used for the injections. In so doing, they were diluted with normal saline to reach a concentration of 50 protoscolices/ml. The viability of the protoscolices was confirmed by flame cell activity and 0.1% eosin staining test under an optical microscope. Thirty-six Sprague Dawley rats were divided into six groups (n=6 each group). The rats in groups I, II, III, IV, and V were intraperitoneally injected with 100 protoscolices of fresh human hydatid fluid in 2 ml of normal saline. The rats in the sixth group (as the control group) were injected with two ml of normal saline. All the rats were kept under the same conditions regarding temperature and feeding. The rats in groups I, II, III, IV, and V were sacrificed and opened after 1, 24, 48, 72, and 168 hours, respectively.

In addition, one of the control mice was sacrificed at each mentioned time point. All the internal organs were observed carefully and three samples were taken from the liver, omentum, and intestine. The samples were separately sent to the pathology laboratory in formalin containers and were prepared for optical microscopy.

This study was approved by the Ethics Committee of Arak University of Medical Sciences, Arak, Iran (No.91-140-10).

RESULTS

In this study, thirty-six rats were experi-

mented in six groups. The pathological studies revealed that there were no implantations of protoscolices in tissue samples of the first, second, and third groups that were sacrificed 1, 24, and 48 hours after the injection. Nonetheless, tissue samples of the fourth and fifth groups that were sacrificed 72 and 168 hours after the injection were implanted with protoscolices. All the tissue samples of the control group were negative in this regard (Table1).

DISCUSSION

Hydatidosis is a chronic, long-term infection of domestic livestock and humans [7]. The slow-growing cysts may persist for several years after the initial infection with protoscolices. The results of our study demonstrated that the protoscolices of hydatid cyst need at least 72 hours for implantation after intraperitoneal injection in rats.

Mousavi and Tappeh reported that experimentally infected mice with protoscolices presented with hydatid cysts from 0.3 to 2 mm after 20 weeks [8]. According to a study by Rogan [7], parasitic cysts were first observed two weeks after intraperitoneal injection in mice, and parasitic cysts with distinctive laminated layers were first observed three-month post-infection. However, based on our findings, implantation could occur as soon as 72 hours after injection.

CONCLUSION

The protoscolices of hydatid cyst require at least 72 hours for implantation after intraperitoneal spillage.

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CONFLICTS OF INTEREST

None declared.

Table 1. The results of pathological study in rats injected with protoscolices of hydatid cyst

Groups	Number of mice	Positive implantation sign	Negative implantation sign
I	6	0	6
II	6	0	6
III	6	0	6
IV	6	2	4
V	6	3	3
VI	6	0	6

I: The group of mice that were sacrificed after 1 h, II: group of mice that were sacrificed after 24 h, III: group of mice that were sacrificed after 48 h, IV: group of mice that were sacrificed after 72 h, V: group of mice that were sacrificed after 168 h, and VI: control group

REFERENCES

1. Mandal S, Mandal MD. Human cystic echinococcosis: epidemiologic, zoonotic, clinical, diagnostic and therapeutic aspects. *Asian Pacific journal of tropical medicine*. 2012;5(4):253-60.
2. Eckert J, Deplazes P. Biological, epidemiological, and clinical aspects of echinococcosis, a zoonosis of increasing concern. *Clinical microbiology reviews*. 2004;17(1):107-35.
3. Holzheimer R, Mannick J. *Surgical Treatment: Evidence-Based and Problem-Oriented*. Munich: Zuckschwerdt; 2001.
4. Sadjjadi SM. Present situation of echinococcosis in the Middle East and Arabic North Africa. *Parasitology international*. 2006;55:S197-S202.
5. Rokni M. The present status of human helminthic diseases in Iran. *Annals of tropical medicine and parasitology*. 2008;102(4):283-95.
6. Rokni M. Echinococcosis/hydatidosis in Iran. *Iranian Journal of Parasitology*. 2009;4(2):1-16.
7. Rogan M. T-cell activity associated with secondary infections and implanted cysts of *Echinococcus granulosus* in BALB/c mice. *Parasite immunology*. 1998;20(11):527-33.
8. Mousavi J, Tappeh KH. Production of Experimental Hydatid Cyst in the Eye, Peritoneum and Liver of BALB/C Mice. *Turkiye parazitolojii dergisi/Turkiye Parazitoloji Dernegi= Acta parasitologica Turcica /Turkish Society for Parasitology*. 2009;34(1):21-3.