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ORIGINAL ARTICLE

Comparing the effectiveness of albendazole and combination of albendazole and praziquantel in experimental hydatidosis

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ABSTRACT

Background: Due to danger of spillage of cyst materials during surgery, chemoprophylaxis is routinely recommended for cyctic echinococcosis. However, for prophylactic purposes the time for starting and the duration of administration of prophylactic drug regimen is not yet clear. The aim of the present study was to evaluate the chemoprophylactic efficacy of one month administration of albendazole or a combination of albendazole plus praziquantel against protoscoleces stage of Echinococcus granulosus when administered before inoculation with the protoscoleces.

Patients and methods: Two days before infestation, 30 mice were assigned in three equal groups of control, albendazole and albendazole plus praziquantel. Control group received dimethylsulfoxide (DMSO). The drugs were suspended in DMSO and administered by gavage for one month (5 consecutive days and two days off every week) in doses of 50mg/kg/day albendazole or 50mg/kg/day albendazole plus 600mg/kg/day praziquantel. The mice were then infected by IP injection of 2000 protoscoleces. Then, three months after the last dose of drug, mice were sacrificed and the number of infected animals, the number, wet weight and cyst sizes were recorded.

Results: The results showed that albendazole chemoprophylaxis reduced the number, weight and cyst sizes by 63.78%, 79.39% and 60.98%, respectively. The corresponding results for the combination of albendazole plus praziquantel were 91.70%, 90% and 80.3%, respectively. All the differences were statistically significant.

Conclusion: Combination of albendazole and praziquantel is more effective than albendazole alone in controlling the development of secondary hydatidosis, when administered for a period of 4 weeks.

Keywords: *Hydatidosis, Hydatid cyst, Echinococcus granulosus, Albendazole, Praziquantel*.
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INTRODUCTION

Cystic echinococcosis (CE) caused by Echinococcus granulosus, is a zoonotic helminth infestation (1) and is the most prevalent zoonotic helminthic infections worldwide, especially in sheep-raising areas. Surgery is still the main form of treatment. However, the danger of spillage of the microscopic cyst materials predisposes the patients to recurrence in other and possibly more dangerous sites of the body (2,3). Albendazole, a benzimidazole compound (4), and praziquantel, an isoquinolone derivative, have been shown effective against the hydatid cyst in the intermediate host (5), however, the employment of a non-invasive method of treatment using ultrasound or computed

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tomography guided percutaneous drainage shows also promising results (6). Nevertheless, the risk of rupture and spillage of cyst contents is still high.

The combination of albendazole and praziquantel was shown to be more effective not only for chemoprophylaxis but also for CE treatment (7-9). However, the current prolonged recommended duration of drug therapy, which may last up to 6 months, predisposes to toxicity and makes the search for a more effective therapy with a shorter duration of chemotherapy necessary (10). Furthermore, investigations on the effectiveness of agents employed at different doses, for varying durations of therapy and on different animal models, reported varying degree of improvement and cure rates. In addition, controversies regarding the response to chemoprophylaxis in respect to the starting time and the duration of drug administration still exist. The aim of this study was, therefore, to compare the effectiveness of albendazole with a combination of albendazole and praziquantel when given before protoscolex inoculation.

PATIENTS and METHODS

E. granulosus protoscoleces were aseptically aspirated from liver of sheep infected with hydatid cysts at municipal abattoir in Ahwaz, Iran. The protoscolex viability was tested by methylene blue exclusion test, flame cell activity and microscopic examination and was found between 90-95%.

Mouse infestation and in vivo treatment: The animals were housed in stainless steel cases in groups of 10, maintained under an environmental conditions of 25-28°C and had free access to water and hard palette food. The cages were cleaned daily.

Thirty female Balb/c mice were randomly assigned in three equal groups. Group 1 (placebo): received only dimethylsulfoxide (DMSO (0.2 ml)). Group 2 received albendazole 50mg/kg/day and group 3 received a combination of albendazole (50mg/kg/day) and praziquantel (600mg/kg/day). The drugs were suspended in 0.2 ml of DMSO and administered by gavage in a single daily dose for one month (5 consecutive days and two days break every week). Two days after drug administration, the mice were infested by IP injection of 2000 protoscoleces in 0.5 ml of PBS solution.

The mice were killed by cervical dislocation under ether anesthesia three months after the last dose of the drugs, dissected and examined by direct macroscopic observation for larval growth. The number, wet weight and the sizes of the developed hydatid cysts were determined in placebo and treatment groups. The size of the cysts were measured by a simple ruler with 1mm accuracy, the wet weight was measured in mg by an electronic balance with 0.1 mg accuracy.

The number of animals that developed cysts, as well as the number and the wet weight and the size of cysts were compared by Dunnet's t-test. The efficacy rate of treatment (based on the number of infected animals, number of cysts, and their weight and sizes) were calculated by use of the following formula: measured parameters in placebo group minus the measured parameters in testing groups divided by the measured parameters in placebo group.

RESULTS

The number of infected animals that developed cysts in the placebo group was 90%, however, this figure was reduced by 55.5% and 22.2% in the albendazole plus praziquantel and the albendazole group, respectively (table 1). On the other hand, the mean number of developed cysts was 4.5±0.9 in placebo group compared with 1.6±0.4 and 0.4±0.3 in albendazole and albendazole plus praziquantel regimen, respectively ($p<0.001$) (table 1).

While the efficacy rates of reduction in the wet weight and size of the cysts in the albendazole-treated group were 79.4% and 60.9%, the corresponding values for the combination treatment

were 90.8% and 80.4%, respectively (p<0.01, and p<0.001, respectively).

Table 1. The number of mice developed hydatid cyst when infected with protoscoleces, the mean (±SEM) number of cysts, wet weight (mg) and size (mm) three months following the therapy in each group

	Placebo (n=10)	**Albendazole (n=10)**	**Albendazole+ Praziquantel (n=10)**
Number[¶]	9	7	4
% of efficacy[#]		22.2	55.5
No. of cysts	4.5±0.9[*]	1.6±0.4	0.4±0.3[***]
% of efficacy		63.8	91.7
Wet weight (mg)	58.2±7.9[*]	12.0±0.6	5.7±0.2[**]
% of efficacy		79.4	90.8
Size (mm)	4.99±0.43[*]	1.95±0.07	0.98±0.05[***]
% of efficacy		60.9	80.4

[¶] Number of mice developed cysts among all infected mice

[#] When compared with the placebo group, [*] p<0.001 when compared with other groups, [**] p<0.01 when compared with the albendazole group, [***] p<0.001 when compared with the albendazole group

DISCUSSION

At present there is discrepancy regarding the drug of choice and duration of treatment prior to surgery of human hydatidosis. In this study, we showed that a 5-day per week treatment with combination of praziquantel plus albendazole for 4 weeks when given 24 hours prior to protoscoleces infestation could significantly reduce the number of developed infection, as well as the number, wet weight and size of hydatid cysts. However, reduction in the number of the cysts is less than what reported by Casado et al., in which the same oral dose of these drugs were administered 48 hours after the inoculation of protoscoleces and continued for one month, with the associated reduction in the number and weight of cysts by 97.7 and 97.15%, respectively (9). These discrepancies in results may be in part explained by differences in the host characteristics, parasite strain, absorption profile of the drugs and their sources, as well as experimental conditions.

In our study, the reduction in number and wet weight of developed cysts following albendazole alone was 63.8% and 79.4%, respectively, when compared with the placebo group. The associated figures were 43.7% and 56.9% in Perez-Serrano study, however, they had administered the drug for 3 months at the same dose and frequency as ours, but the drugs were commenced 3 days post-inoculation (11). This fact suggests that time-point of administration has an important role in the efficacy of albendazole on the free protoscoleces when administered before inoculation.

Albendazole is the most commonly used drug in the medical treatment of echinococcosis. Time course of pre- and pos-operative treatment is still not clear. Our results indicate that a short-term (one month) course of prophylaxis with albendazole could effectively reduce cyst size and weight in animal model. Although albendazole showed good cure rate in human cases, it is routinely prescribed for a period of up to 6 months with adverse side effects (12). On the other hand, the combination of praziquantel and albendazole was significantly more effective than albendazole alone.

The reduction of therapy course to 1 month had not significantly reduced the effectiveness. Our results are in agreement with those reported by Taylor and Morris, and Casado et al (9,13). In our experiment treatment was started two days before inoculation of protoscoleces while these studies started treatment few days after protoscolex injection. In our opinion, administration of drugs before inoculation is advantageous for two reasons: firstly, in clinical practice drugs are administered at least one week before surgery and treatment is continued for several months. Secondly, since the spilled protoscoleces can undergo various stages of differentiation of the parasite material after its release from the surgically excised cyst, administration before the formation of the protective laminated layer (14) can be a more accurate predictor of efficacy of protoscolicidal agent at that particular time-point. It is recognized

that extrapolation of data from animal studies to man should be very guarded, especially with regards to the doses of the employed drugs. Prudently, further similar studies specifically designed to evaluate the efficacy of different doses of these agents need to be tested if these data to be related clinically.

In conclusion, various time-points of drug administration, doses and durations of treatment with albendazole and/or praziquantel has been studied both in human and animals. The results of this study clearly demonstrated that combined chemoprophylaxis with albendazole and praziquantel, administered for a relatively short time course of 4 weeks in an experimental animal model, is more effective than albendazole alone and it could be considered as an option for further investigations in clinical practice.

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REFERENCES

1. Zhang W, Li J, McManus DP. Concepts in immunology and diagnosis of hydatid disease. Clin Microbiol Rev 2003;16:18-36.

2. Jimenez-Mejias ME, Alarcon-Cruz JC, Marquez-Rivas. Orbital hydatid cyst: treatment and prevention of recurrences with albendazole plus praziquantel. J Infect 2000;6:105-7.

3. Pawlowski ZS, Eckert J, Vuitton DA. Echinococcosis in humans: clinical aspects, diagnosis and treatment. In: Eckert J, Gemmell MA, Meslin FX, Pawlowski ZS, editors. WHO/OIE manual on echinococcosis in humans and animals: a public health problem of global concern. Office International des Epizooties and World Health Organization, Paris, 2001; p:20-71.

4. Morris DL, Chinnery JB, Hardcastle JD. Can albendazole reduce the risk of implantation of spilled protoscoleces. An animal study. Trans R Soc Trop Med Hyg 1986;80:481-84.

5. Urrea-París MA, Casado N, Moreno MJ, Rodriguez-Caabeiro F. Chemoprophylactic praziquantel treatment in experimental hydatidosis. Parasitol Res 2001;87(6):510-2.

6. Raymond A, Smego JR, Sebanego P. Treatment options for hepatic cystic echinococcosis. Int J Infect Dis 2005;9:69-76.

7. Cobo F, Yarnoz C, Sesma B, Fraile P, Aizcorbe M, Trujillo R, et al. Albendazole plus praziquantel versus albendazole alone as a pre-operative treatment in intra-abdominal hydatisosis caused by Echinococcus granulosus. Trop Med Int Health 1998;3(6):462-6.

8. Mohamed AE, Yasawy MI, Karawi MA. Combined albendazole and praziquantel versus albendazole alone in treatment of hydatid disease. Hepatogastroenterol 1998;45:1690-94.

9. Casado N, Urrea-París MA, Moreno MJ, Rodríguez-Caabeiro F. Combined praziquantel and albendazole chemoprophylaxis in experimental hydatidosis. Parasitol Res 2001;87(9):787-9.

10. Moreno MJ, Casado N, Urrea-París MA, Rodríguez-Caabeiro F. Could ivermectin have a synergic effect with albendazole in hydatidosis therapy?. Parasitol Res 2002;88(6):563-7.

11. Perez-Serrano J, Denegri G, Casado N, Redruguez F. In vivo effect of oral albendazole-sulfoxide on development of secondary Echinococcosis in mice. Inter J Parasitol 1997;27:1341-45.

12. Kern P. Echinococcus granulosus infection: clinical presentation, medical treatment and outcome. Langenbecks. Arch Surg 2003;388:413-20.

13. Taylor DH, Morris DL. Combination chemotherapy is more effective in postspillage prophylaxis for hydatid disease than either albendazole or praziquantel alone. Br J Sug 1989;76:954-59.

14. Urrea-París MA, Moreno MJ, Casado N, Rodríguez-Caabeiro F. Relationship between the efficacy of praziquantel treatment and the cystic differentiation in vivo of Echinococcus granulosus metacestode. Parasitol Res 2002;88(1):26-31.