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Preventive effect of artemether in rabbits infected with *Schistosoma japonicum* cercariae[1]

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KEY WORDS *Schistosoma japonicum*; Schistosomiasis/prevention & control; drug administration schedule; artemether

AIM: To study the effect of artemether (Art) for prevention of schistosomal infection. **METHODS**: Rabbits with single infection or reinfection with *Schistosoma japonicum* cercariae were treated intramuscularly (im) or intragastrically (ig) with Art 5 – 20 mg·kg^{-1} on d 7 – 15 after the first infection, followed by various regimens. **RESULTS**: When rabbits were injected im Art 7.5 mg·kg^{-1} (ie, one half of the effective dose given ig on d 7) followed by once every week for twice, the ♀ worm reduction rate was only 42 %. In infected rabbits treated ig with Art 10 – 20 mg·kg^{-1} given in the same administration schedule, the ♀ worm reduction rates were >91 %. When Art 15 mg·kg^{-1} was given to rabbits on d 7 – 14 and the following dose of the drug was given at intervals of 7 – 14 d, the ♀ worm reduction rates were >94 %. In rabbits reinfected with cercariae, the ♀ reduction rate of Art given ig once a week for 3 times since d 8 after the first infection was 96 % which was similar to that given once a week twice since d 14 after the first infection. **CONCLUSION**: Art should be given ig on d 7 – 15 after infection, followed by repeated dosing once every 7 – 15 d for a total of 3 doses. Art given ig daily for 2 consecutive days or given at 1-wk intervals since 7 – 15 d after infection also showed preventive effect.

When dogs or rabbits with single infection or reinfection with *Schistosoma japonicum* cercariae were treated ig with artemether (Art) at a single dose on d 7 after the first infection, followed by once every 1 or 2 wk for 3 – 4 times, a promising preventive effect was seen which included apparent reduction of mean numbers of total and ♀ worm, negative response in some parameters related to acute schistosomiasis and protection of the liver from damage induced by schistosome eggs[1]. Field trials indicated that Art exhibited a promising preventive effect on reducing infection rate and intensity of infection as well as controlling acute schistosomiasis[2-7]. Thus, the Art can be used as a preventive measure against schistosomal infection in endemic areas. In this paper we reported the effect of Art injected im, and comparison of several regimens of Art used for prevention of schistosomiasis in rabbits.

MATERIALS AND METHODS

Parasites *Schistosoma japonicum* cercariae (Anhui isolate), released from artificially infected *Oncomelania hupensis*, was provided by the Department of Vector Biology and Control of our Institute.

Drug Art was the product of Kunming Pharmaceutical Corporation (lot № 880701). For intragastric gavage Art was suspended in 1 % tragacanth at the concentrations of 10 – 20 g·L^{-1}, while the Art dissolved in corn oil at the concentrations of 5 – 7.5 g·L^{-1} was used for im injection.

Rabbits New Zealand strain rabbits (♀ and ♂) weighing 2.2 – 2.5 kg were provided by our Institute (Certificate № 02-32-1). Rabbits infected with 198 – 202 schistosome cercariae via shaved abdominal skin or infected with 48 – 52 schistosome cercariae once every other day for 5 times were treated ig with Art 10, 15 or 20 mg·kg^{-1}·d^{-1} on d 7 – 15 for 2 d or once every 1 – 2 wk for 2 – 3 times. In another experiment, the im dosage was one half of the effective dose used in intragastric gavage. All rabbits were killed 4 wk after the last medication and the preventive effect was evaluated by numbers of total and ♀ worm.

RESULTS

Preventive effect of im Art In rabbits treated im with Art 5 mg·kg^{-1} on d 7 (group 2), no

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apparent effect was seen. With 7.5 mg·kg^{-1} (group 3), the total worm, but not the ♀ worm were less than those of the control. Rabbits treated ig with Art 10 or 15 mg·kg^{-1} (group 4 and group 5) resulted in total and ♀ worm reduction rates > 73 % *vs* the control. When above-mentioned dosages of Art were injected im once every wk for 2 times (group 6 and group 7) followed the first medication on d 7 after infection, the total and ♀ worm were less than those of the control but higher than those of the groups treated ig with Art 10 or 15 mg·kg^{-1} (group 8 and group 9) (Tab 1).

Comparison of regimens When Art 10 or 20 mg·kg^{-1} was given ig to rabbits once on d 7 and d 8 (group 11 and group 12) or d 15 and d 16 (group 15 and group 16) for 2 d, the total and ♀ worms of each group fluctuated a little ($P > 0.05$). Apart from group 12 the total and ♀ worm reduction rates were over of 90 %. In rabbits treated ig with Art 10 or 20 mg·kg^{-1} on d 7 after infection, followed by repeated dosing once every wk for 2 times (group 13 and group 14), the total and ♀ worms were less than those in groups treated with Art on d 7 and d 8 or d 15 and d 16. In group 13 and group 14, 1 or 2 out of each 3 rabbits were free from the ♀ worm (Tab 2).

In rabbits treated ig with Art 15 or 20 mg·kg^{-1} on d 15 after infection, followed by repeated dosing every 2 wk for once (group 18 and group 19) or twice (group 20 and group 21), no apparent differences in total and ♀ worm between these groups were seen (Tab 2).

Medications at different intervals When Art 15 mg·kg^{-1} was given ig to rabbits on d 7 after infection, followed by once every 7, 10, or 14 d for twice (group 23, 24, 25), the numbers of total and ♀ worms of group 24 and group 25 were similar to those of group 23. In rabbits treated ig with Art 15 mg·kg^{-1} on d 14, followed by once every 1 or 2 wk for twice (group 26 and 27), the numbers of total and ♀ worms of group 26 tended to be less than those of group 23 ($P > 0.05$). The numbers of total and ♀ worms of group 27 were significantly higher than those of group 26, but no differences ($P > 0.05$) were seen when those were compared with group 23 (Tab 2).

Regimens used in reinfection When rabbits infected with 48 – 52 cercariae once every other day for 5 times were initially treated ig with Art 15 mg·kg^{-1} on d 8 after the first infection, followed by once a week for 2 times (group 29), the numbers of total and ♀ worms (worm reduction rates > 95 %) were less than those of the control. In rabbits treated ig with Art at the same dose on d 13 and d 14 (group 31), or on d 14 and d 21 (group 32), the mean numbers of total and ♀ worms were similar to those of the group 29. Whereas the rabbits initially treated ig with Art 15 mg·kg^{-1} on d 8 or d 14 after the first infection, followed by a second dosing at 2-wk intervals (groups 30 and 33), the numbers of total and ♀ worm were higher than those of the group 29, although the total and ♀ worm reduction rates were >87 % as compared with those of the control (Tab 2).

Tab 1. Preventive effects of artemether given im or ig on rabbits infected with *Schistosoma japonicum* cercariae. $x \pm s$. $^aP > 0.05$, $^bP < 0.05$, $^cP < 0.01$ *vs* the control.

Group	Route	Dose/ mg·kg^{-1}	Time of medication	Rabbits	Total worm	Worm reduction rate/ %	Female worm	Female worm reduction rate/ %
1	Control	0	–	6	131 ± 10	–	57 ± 9	–
2	im	5	d 7	6	123 ± 12^a	6	58 ± 7^a	0
3	im	7.5	d 7	6	98 ± 27^b	25	44 ± 14^a	23
4	ig	10	d 7	6	32 ± 15^c	76	15 ± 7^c	74
5	ig	15	d 7	6	34 ± 11^c	74	15 ± 6^c	74
6	im	5	d 7, 14, 21	6	78 ± 10^c	41	37 ± 4^c	35
7	im	7.5	d 7, 14, 21	6	70 ± 11^c	47	33 ± 6^c	42
8	ig	10	d 7, 14, 21	5	7.4 ± 6.1^c	95	3.4 ± 2.6^c	94
9	ig	15	d 7, 14, 21	5	2.2 ± 1.5^c	98	0.8 ± 0.4^c	99

Tab 2. Rabbits infected with *Schistosoma japonicum* cercariae and treated ig with artemether at different regimens after infection. $\bar{x} \pm s$. [a] $P > 0.05$ *vs* corresponding 20 mg·kg^{-1} group. [c] $P < 0.01$ *vs* corresponding group 13 (10 mg·kg^{-1}) and group 14 (20 mg·kg^{-1}). [d] $P > 0.05$ *vs* group 23. [g] $P > 0.05$, [h] $P < 0.05$, [i] $P < 0.01$ *vs* group 29.

Group	Time of medication	Rabbits	Dose/ mg·kg^{-1}	Total worm	Worm reduction rate/ %	Female worm	Famale worm reduction rate/ %
10	Control	5	0	146 ± 10	–	72 ± 5	–
11	d 7,8	4	10	11 ± 5[ac]	92	5.3 ± 2.0[ac]	93
12	d 7,8	3	20	16 ± 4[c]	89	8.0 ± 2.0[c]	89
13	d 7,14,21	3	10	2.3 ± 2.1[a]	98	0.7 ± 1.2[a]	99
14	d 7,14,21	3	20	2.0 ± 2.0	99	1.0 ± 1.0	99
15	d 15,16	4	10	14 ± 6[ac]	90	6.3 ± 2.6[ac]	91
16	d 15,16	4	20	14 ± 6[c]	90	6.3 ± 2.6[c]	91
17	Control	5	0	157 ± 7	–	58 ± 8	–
18	d 15,29	4	15	16 ± 13[a]	90	7.5 ± 6.0[a]	87
19	d 15,29	6	20	12 ± 7	92	5.5 ± 3.5	91
20	d 15,29,43	4	15	17 ± 9[a]	89	8.3 ± 4.8[a]	86
21	d 15,29,43	4	20	11 ± 7	93	5.0 ± 3.7	91
22	Control	7	0	120 ± 16	–	60 ± 8	–
23	d 7,14,21	5	15	4 ± 3	97	1.8 ± 1.6	97
24	d 7,17,27	5	15	2.2 ± 1.8[d]	98	1.0 ± 1.0[d]	98
25	d 7,21,35	4	15	6.8 ± 5.5[d]	94	3.3 ± 2.6[d]	94
26	d 14,21,28	5	15	0.6 ± 0.9[d]	99	0.2 ± 0.4[d]	99
27	d 14,28,42	5	15	4.8 ± 3.9[d]	96	2.2 ± 1.6[d]	96
28	Control	4	0	156 ± 18	–	78 ± 9	–
29	d 8,15,22	4	15	8 ± 4	95	3.5 ± 1.9	96
30	d 8,22	5	15	20 ± 7[i]	87	9.4 ± 4.1[h]	88
31	d 13,14	4	15	11 ± 6[g]	93	5.5 ± 2.6[g]	93
32	d 14,21	6	15	5 ± 3[g]	97	2.3 ± 1.5[g]	97
33	d 14,28	4	15	21 ± 5[i]	86	10 ± 3[h]	87

DISCUSSION

In experimental treatment of mice infected with *Plasmodium berghei*, the effect of im Art (tea seed oil preparation) 15 mg·kg^{-1} was better than that of ig Art (suspension in tragacanth gum) 60 mg·kg^{-1} [8]. Therefore, it needs to determine and compare the preventive effects of Art given by these two routes. Although the ig LD_{50} of Art suspension and im LD_{50} of Art oil preparation in mice were 977 mg·kg^{-1} [9] and 263 mg·kg^{-1} [8], respectively, we selected only one half of the ig effective dose used for im medication. Unexpectedly, only a less preventive effect was seen. This might be ascribed to the low concentration of the drug in the circulation blood. Because after absorption of Art from the local injected site the drug was rapidly diluted by the blood in the circulation. Thus, the drug concentrations distributed in the liver and the mesenteric veins, where schistosomules located, were rather low.

After analysis of the results obtained from the several Art regimens used for prevention in rabbits, the key points can be summarized as follows, ie 1) since Art given at 10, 15, or 20 mg·kg^{-1} exhibits similar preventive effects, the adjustment of dose used for field trials seems to be unnecessary; 2) the first dosing of Art can be given on d 7 – 15 after the infection with cercariae; 3) after the first dosing of Art, the following dosing can be given at 7, 10, or 15 d intervals; 4) Art given daily for consecutive 2 d also shows preventive effect but less than that of the drug given at 1-wk intervals; 5) in reinfection with cercariae Art can be given once a week since d 7 after the first infection, or given 2 doses at 1-wk intervals since d 14 after the first infection.

According to the above-mentioned and previous experiments[1] as well as the field trials[2-7], the following suggestions would be helpful for using oral Art in prevention of schistosomiasis in endemic areas, ie Art 6 mg · kg^{-1}, a recommending effective dose[3-7], is initially given to the individuals on 7 – 15 d after contact with the infested water, following by repeated dosing once every 15 d during the whole transmission season. In the individuals, who work in the infested water or contact with the infested water in short time period, will be initially treated with Art on 7 – 15 d after contact with the infested water, and the same dose is given again every 15 d during continuing contact with the infested water. After the individuals withdraw from the pilot site one more dosing of Art is given 7 – 15 d later.

REFERENCES

1 Xiao SH, You JQ, Yang YQ, Wang CZ. Experimental studies on early treatment of schistosoma infection with artemether. Southeast Asian J Trop Med Public Health 1995; 26: 306 – 18

2 Xiao SH, Shi ZG, Zhou SJ, Wang CZ, Zhang ZG, Chu B, *et al*. Field studies on the preventive effect of oral artemether against schistosomal infection. Chin Med J 1996; 109: 272 – 5

3 Xiao SH, Wang JL, Wang CZ, Yang Z, Chu B, Yang H, *et al*. Protection of the residents from schistosome infection using oral artemether in mountainous area. Chin J Parasitol Parasitic Dis 1996; 14: 11 – 4.

4 Tian ZY, Xiao SH, Xiao JW, Zhou YC, Liu DS, Zheng J, *et al*. Observation on reducing schistosomal infection rate in an islet with embankment type endemic area after prophylaxis with oral artemether given to the population throughout the whole transmission season Chin J Parasitol Parasitic Dis 1997; in press.

5 Wang JL, Xiao SH, Yang Z, Wang MK, Yang H, Liu YH, *et al*. Effect of oral artemether in controlling schistosomiasis in Yunnan mountainous endemic area Chin J Parasitol Parasitic Dis 1997; 15: 138 – 42.

6 Xu MS, Xiao SH, Song Q, Tao CG, Xia CG, Wang H, *et al*. Observation on the effect of artemether on controlling schistosomiasis japonica in an endemic area of marshlands Chin J Parasitol Parasitic Dis 1997; 15: 212 – 5.

7 Song Y, Xiao SH, Wu W, Zhang SJ, Xie HQ, Xu XP, *et al*. The preventive effect of artemether in protection of people from schistosome infection during fighting against flood in Poyang lake. Chin J Parasitol Parasitic Dis 1997; 15: 133 – 7.

8 Gu HM, Liu MZ, Lu BF, Xu JY, Chen LJ, Wang MY, *et al*. Antimalarial effect and toxicity of artemether in animals Acta Pharmacol Sin 1981; 2: 138 – 44

9 Le WJ, You JQ, Yang YQ, Mei JY, Guo HF, Yang HZ, *et al*. Studies on the efficacy of artemether in experimental schistosomiasis. Acta Pharm Sin 1982; 17 : 187 – 93

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关键词 日本血吸虫；血吸虫病预防和对照；给药计划表；蒿甲醚

目的：观察蒿甲醚(Art) im 及其适宜给药方案预防血吸虫感染的效果。**方法**：兔感染日本血吸虫尾蚴后 d 7 – 15 im 或 ig Art 观察几种给药方案的预防作用。**结果**：感染兔每周 im Art 7.5 mg·kg^{-1} 1 次，共 3 次，减虫率仅 42 %。兔感染后 d 7 – 15 ig Art 10 – 20 mg·kg^{-1}，然后每 7 – 14 d 给药 1 次，共 2 次，减虫率均在 91 %以上；或 ig 1 剂 Art，继而每间隔 7 – 14 d ig 1 次，共 3 次，减虫率 > 94 %；重复感染兔自首次感染后 d 8 每 wk ig Art 1 次，共 3 次，或自首次感染后 d 14，每 wk ig Art 1 次，共 2 次的减虫率为 95 % – 97 %。**结论**：本文所述几种给药方案对预防兔感染血吸虫尾蚴均有效果。