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Albendazole–soybean oil emulsion for the treatment of human cystic echinococcosis: Evaluation of bioavailability and bioequivalence

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Abstract

A single 12.5 mg/kg dose of albendazole (Abz) in tablet form (AbzT) followed 2 weeks later by an equivalent dose of Abz emulsified in 30% soybean oil (AbzE) was administered orally 2 h after the first morning meal to 7 male adult patients with cystic echinococcosis caused by *Echinococcus granulosus*. Serum samples were taken 1, 3, 5, 7, 8, 9, 11, 14, 18, 24, 36, and 48 h post medication from each patient to measure the serum concentrations of albendazole sulfoxide (AbzSOX), the principal bioactive metabolite of Abz. AbzSOX concentrations were measured by reverse phase HPLC. The data were subjected to pharmacokinetic analysis to compare the relative bioavailability and bioequivalence of AbzT and AbzE. The results demonstrated that the mean peak concentrations (C_{max}) for AbzT and AbzE were 1.06 ± 0.38 mg/l and 1.71 ± 0.47 mg/l, respectively; the area under the concentration–time curves (AUC) were 13.24 ± 4.93mg·h/l and 21.01 ± 7.54 mg·h/l, respectively. The relative bioavailability of AbzE was $F_{Flu} = 1.59$. Two one-sided tests procedure and $(1 - 2\alpha)$ 90% confidence interval methods were used to evaluate the bioequivalence of AbzE and AbzT. The results demonstrated that the bioavailability of AbzE was greater than AbzT.

Keywords: Albendazole; Albendazole sulfoxide; Cystic echinococcosis; Pharmacokinetics; Bioavailability; Bioequivalence

1. Introduction

Hydatid disease caused by *Echinococcus granulosus* is a major zoonotic infection that is detrimental to both human health and animal husbandry in West China. The disease is highly endemic in the provinces and autonomous regions

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of Xinjiang, Qinghai, Gansu, Ningxia, Nei Mongolia, Tibet, and Sichuan where it is estimated that approximately 50 million individuals are at risk for acquiring the infection (Jiao W., personal communication). It is estimated that at the present time there are 4–500 000 cases with echinococcosis in China.

Benzimidazole anthelminthics have been used for the treatment of hydatid disease since the 1970s when mebendazole (Mbz) was discovered (Ammann et al., 1979). Since then WHO-sponsored studies demonstrated the superiority of albendazole (Abz) relative to Mbz (Davis et al., 1986, 1989). Afterwards, further studies indicated that both Abz and Mbz evidenced certain efficaciousness, and Abz was better than Mbz (Horton, 1989; De Rosa and Teggi, 1990; Todorov et al., 1992; Nahmias et al., 1994). Abz is now considered the chemotherapeutic treatment of choice for human echinococcosis. Based on almost two decades of experience, the cure rate for human cystic echinococcosis with Abz is estimated to range between 20 and 40% (Nahmias et al., 1994; Horton, 1997). In order to improve its therapeutic efficacy for use in China, our laboratory has reformulated Abz by emulsifying the drug with different combinations of vegetable oils. Our recent experience with Abz in a 30% soybean oil emulsion (AbzE) demonstrated its superiority over Abz with respect to its bioavailability in experimental echinococcosis in mice (Xiao et al., 2002). Mice infected with *E. granulosus* and treated with AbzE also experienced improved therapeutic responses relative to infected mice treated with Abz (Xiao et al., 2002). Based on the apparent success of treating murine echinococcosis with AbzE we undertook an initial evaluation of this agent in humans with *E. granulosus* infection.

2. Materials and methods

2.1. Albendazole formulations

Abz in substance was purchased from the Hanjiang Pharmaceutical Corporation, China (Lot 9502010). AbzE was prepared by mixing Abz powder in the presence of distilled water, 30% soybean oil (v/v), emulsifiers as described previously (Xiao et al., 2002). Albendazole tablets (AbzT) each containing 200 mg of Abz were the product of Yixing Pharmaceutical Factory, Jiangsu Province (lot 960605).

2.2. Patients enrollment

Seven male patients from Xinjiang Autonomous Region with clinically diagnosed cystic echinococcosis were selected for the study. Written informed consent was obtained based on a study protocol approved by the Institute Review Board (IRB) of the Institute of Parasitic Diseases of the Chinese Academy of Preventive Medicine (IPD-CAPM). The IPD-CAPM IRB is registered with the Office of Protection from Research Risks (OPHR) of the US Department of Health and Human Services. The patients ranged in age from 24–38 years (mean 29.3 ± 5.5 years); their body masses ranged between 55–81 kg (mean 66.4 ± 9 kg). The subjects had no other pre-existing morbid conditions and they had not taken any other medicines within 2 weeks prior to enrolling in the study. All subjects were determined to have normal liver and renal function.

2.3. Oral treatment

Either AbzE or AbzT was administered at a dose corresponding to 12.5 mg/kg of the Abz component (maximal dose of 1 g). For the initial dose, AbzT was administered to each patient approximately 2 h after the first morning meal. Two weeks following the initial AbzT dose, AbzE was subsequently administered to each patient, again approximately 2 h after the first morning meal. Blood samples were taken before and 1, 3, 5, 7, 8, 9, 11, 14, 18, 24, 36, and 48 h post each medication. The serum from each sample was separated and kept frozen at -20 °C. All patients were hospitalized during their Abz treatments and for blood-sampling; they were provided low fat meals and there was no significant variation between study and control groups in terms of diet.

2.4. Serum Abz sulfoxide measurements

Serum concentrations of the Abz bioactive metabolite, albendazole sulfoxide (AbzSOX), were measured in 1 ml of serum as described previously (Wang et al., 1991). Briefly, serum samples were subjected to reverse phase HPLC with a CLC-ODS Column (150×6.0 mm^2) containing ODS 5 μm or a preparatory column (30 mm in length) containing ODS 37–50 μm. The drug Mbz was used as an internal standard. Mbz was synthesized and donated by the Shanghai Institute of Pharmaceutical Industry. AbzSOX was synthesized and purified as described previously (Huang and Hu, 1995). The sensitivity of the assay, i.e. minimal concentration of AbzSOX that it can be measured in the plasma is 5 μg/l.

2.5. Pharmacokinetic modeling and evaluation

The bioequivalence of AbzE was evaluated by either the two one-sided tests procedure or the $(1-2\alpha)$ 90% confidence interval method, as described previously (Shuirmann, 1987; Huang and Han, 1993). The Students t-test was used to analyze the statistical difference in matched-pair tests. The parameters evaluated included the area under drug–time curve (AUC) calculated by the trapezoidal method (Zeng et al., 1982), or its logarithmic conversion (ln AUC).

3. Results

The mean serum AbzSOX concentrations at different time intervals measured from each of the 7 patients treated consecutively with AbzT followed by AbzE were shown in Table 1. Observed curves of AbzSOX serum drug concentrations over time fitted by computer conformed a two-compartment model with r values of 0.999 and 0.998 for both AbzE and AbzT, respectively (Fig. 1). The mean AUCs of AbzT and AbzE were estimated by the trapezoidal method (Zeng et al., 1982) to be 13.24 ± 4.93 mg·h/l and 21.01 ± 7.52 mg·h/l, respectively (Table 2). The relative bioavailability of AbzE was $F_{\text{Flu}} = 1.59$. The two one-sided tests calculated demonstrated that the two preparations of Abz did not exhibit biological equivalence. Similar results were observed when calculations were made substituting ln AUC (AbzE, 3.00 ± 0.32; AbzT, 2.53 ± 0.34). Furthermore, calculation by the $(1-2\alpha)$ 90% confidence interval method (Shuirmann, 1987; Huang and Han, 1993), the AUC value of AbzE was 21.01 (Table 2), which was beyond the 90% confidence intervals for AbzT (12.23 and 14.26), indicating biological unequivalence between AbzE and AbzT. Absence of biological equivalence was also confirmed in calculations using ln AUC, i.e. the

Table 1
Blood AbzSOX concentration (mg/l) at different intervals in 7 cases with cystic echinococcosis treated orally with AbzT or AbzE at a single dose of 12.5 mg/kg

Time after administration (h)	AbzT	AbzE
1	0.52 ± 0.23	0.76 ± 0.24
3	1.06 ± 0.38	1.71 ± 0.47
5	0.91 ± 0.36	1.57 ± 0.53
7	0.72 ± 0.30	1.29 ± 0.42
8	0.61 ± 0.28	1.09 ± 0.62
9	0.61 ± 0.31	0.98 ± 0.42
11	0.45 ± 0.16	0.79 ± 0.38
14	0.30 ± 0.15	0.47 ± 0.18
18	0.21 ± 0.11	0.39 ± 0.28
24	0.14 ± 0.10	0.18 ± 0.14
36	0.09 ± 0.05	0.10 ± 0.06
48	0.07 ± 0.04	0.07 ± 0.05

$x \pm$ SD. The two preparations were administered consecutively at 2-week interval.

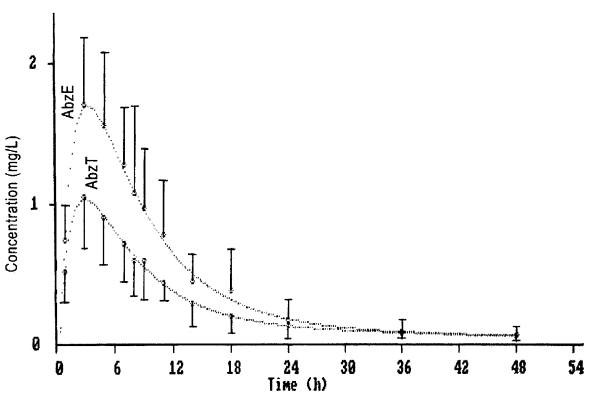

Fig. 1. The fitting curve (observed: ○, calculated: …) in AbzSOX pharmacokinetics of AbzT and AbzE.

Table 2
AbzSOX AUC (mg·h/l) in cystic echinococcosis cases treated orally with AbzT and AbzE by trapezium approximate calculation and its ratio

Case no.	AbzT	AbzE	AbzE/AbzT
1	7.86	19.14	2.43
2	22.82	36.26	1.59
3	10.46	23.77	2.27
4	16.07	20.90	1.30
5	13.27	15.42	1.16
6	11.29	13.51	1.20
7	10.92	18.07	1.63
X	13.24 ± 4.93	21.01 ± 7.52	$X_{\mathrm{AbzE}/\mathrm{AbzT}} = 1.59$

ln AUC value of AbzE (3.00) was dropped outside the 90% confidence intervals of ln AUC value of AbzT (2.22, 2.85). Comparative analysis on the maximum blood AbzSOX concentrations showed that the mean $C_{\max}$ was 1.06 ± 0.39 mg/l in AbzT group and 1.75 ± 0.47 mg/l in the AbzE group with an AbzE/AbzT ratio of 1.65. Statistical analysis revealed $t = 5.10$ and $t > t_{0.005}(4.32)$ ($P < 0.005$).

4. Discussion

AbzSOX is the active echinococcocidal metabolite of Abz following oral administration of the Abz parent compound in humans and animals (Xiao et al., 1994; Prez-Serrano et al., 1997). By measuring the serum concentrations of AbzSOX following oral administration of either AbzT or AbzE followed by pharmacokinetic modeling, the relative bioavailability and bioequivalence of the two compounds was determined. Both Abz absorption and serum levels of AbzSOX were increased following AbzE administration relative to AbzT administration. Previously, it was demonstrated that if the relative bioavailability (F_{Flu}) of two preparations of a drug derived from the AUC fell in the range 0.75–1.25, then bioequivalence of the two preparations could be established (Zhao, 1993). In our experiment, the relative bioavailability of AbzE $F_{\mathrm{Flu}} = 1.59$ surpassed this range, demonstrating the absence of bioequivalence of the two preparations. Striking individual differences in the AUC levels were seen after oral administration of AbzT and AbzE, particularly in AbzE group. The standard deviation of AbzE group was higher than that of AbzT group, indicating the higher individual differences in oral absorption of AbzE. Even though the difference of mean AUC levels between AbzT and AbzE groups is significant ($P < 0.05$). The two one-sided test for examining the equivalence threshold (with or without logarithmic conversion) reproducibly demonstrated statistically-significant differences in bioequivalence. This observation was supported by a matched-pair analysis on the two groups, in which comparison of AUC and $C_{\max}$ also showed significant difference. It was shown previously that there are no significant pharmacokinetic differences with respect to Abz metabolism between patients with hydatid disease and healthy volunteers. (Pan et al., 1992). Due to its significant improvement of bioavailability, AbzE will be recommended for evaluation in the clinical treatment of echinococcosis (Chai et al., 2001).

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