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Pharmacokinetics of tablet huperzine A in six volunteers[1]

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AIM: To study pharmacokinetics of tablet huperzine A (Hup-A) in Chinese volunteers to help establishing its drug administration schedule. **METHODS**: For 6 volunteers after a single oral dose of 0.99 mg, drug concentrations in plasma were assayed by reverse phase high pressure liquid chromatography (HPLC) at 0.5, 0.75, 1.0, 1.25, 1.5, 2, 4, 6, 8, and 10 h. The pharmacokinetic parameters were calculated with a 3P87 program by computer. **RESULTS**: The time course of plasma concentrations conformed to a one-compartment open model with a first order absorption. The pharmacokinetic parameters were as follows: $T_{\frac{1}{2}k_a} = 12.6$ min, $T_{\frac{1}{2}k_e} = 288.5$ min, $T_{max} = 79.6$ min, $C_{max} = 8.4$ μg L^{-1}, AUC = 4.1 mg L^{-1} min. **CONCLUSION**: Hup-A was absorbed rapidly, distributed widely in the body, and eliminated at a moderate rate.

KEY WORDS huperzine A; cholinesterase inhibitors; high pressure liquid chromatography; pharmacokinetics; phase I clinical trials

Huperzine A (Hup-A), a new alkaloid first isolated from Chinese herb *Huperzia serrata* (Thunb) Trev[1], exhibited a selective inhibition on acetylcholinesterase (AChE)[2]. It potentiated the skeletal muscle contraction and increased muscle tones[3], and enhanced rodent learning and memory[4]. Clinically, Hup-A improved muscle weakness of myasthenia gravis[5] and memory in patients with impaired memory or Alzheimer's disease[6]. The plasma level of Hup-A following iv or ig [^{3}H]Hup-A 13.9 MBq kg^{-1} in rats declined in two phases, the distribution phase and the elimination phase, with half-lives of 6.6, 149 min (iv) and 10, 203 min (ig) respectively[7]. This paper was to study the pharmacokinetics of Hup-A in healthy volunteers to help establishing its drug administration schedule in clinic.

MATERIALS AND METHODS

Drug According to Chinese National Standard tablet Hup-A (batch № 940112) was prepared by the Institute of Materia Medica, Zhejiang Academy of Medical Sciences. The purity of Hup-A was 99.5 %. Each tablet contains Hup-A 0.09 mg. (±)-Dinor Hup-A as internal standard was synthesized and presented kindly by Dr HE Xu-Chang, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, and 3 mg L^{-1} was used for experiment.

Subjects Six Chinese volunteers (M 3, F 3), aged 27±6 a and weighing 58±7 kg were all healthy, not in pregnant or menstruation. Each volunteer was told about the aim and process of the study. Agreements were obtained from them before study. Each subject was given a single oral dose of 0.99 mg Hup-A tablet at 8 am after an overnight fasting. Breakfast was served at 10 am. Blood (5 mL) was collected from an indwelling catheter in antecubital vein before and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 2, 4, 6, 8, and 10 h after *po*. Plasma (2 mL) was taken for HPLC. Pharmacokinetic parameters were obtained by first calculating the parameters from each person and then taking average of the 6 parameters, using a 3P87 program provided by Chinese Mathematic-Pharmacological Society on the computer.

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HPLC Shimadzu LC-6A liquid chromatography was connected to SPD-6A uv spectrophotometric detector (Shimadzu) and Rheodyn 7125 sampler, recorded on C-R3A integrator (Shimadzu). The column was a Spherisorb C18 (150 mm · 5 mm inner diameter; 5 μm particle size). The mobile phase was methanol: water (45:55, vol/vol), 1.0 mL min^{-1} at 30 C column oven. The column effluent was monitored at 313 nm.

Plasma sample Add (±)-dinor Hup-A 100 μL to plasma 2 mL, add Na_2CO_3-$NaHCO_3$ buffer 1 mL (using NaOH 1 mol L^{-1} to adjust pH to 11.9). Then add chloroform 7.5 mL, shake 2 min, and centrifuge at 1000 × *g* for 10 min. The organic phase was blown to dryness by N_2 at 40 C. Dissolve the residue with HPLC mobile phase 50 μL, and 20 μL was applied to HPLC. Hup-A peak and (±)-dinor Hup-A peak were separated clearly. The retention times (Rt) of (±)-dinor Hup-A and Hup-A were 3.5 and 8.3 min, respectively (Fig 1).

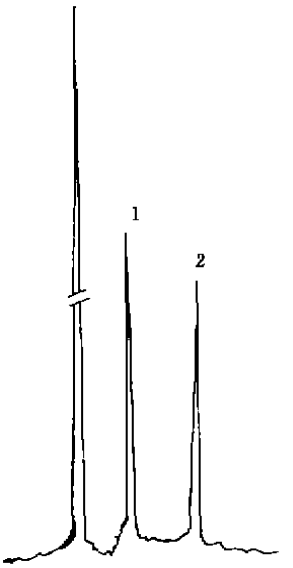

Fig 1. Chromatogram of blank plasma spiked with internal standard (peak 1, retention time 3.5 min) and Hup-A (peak 2, retention time 8.3 min).

Standard curve To the plasma containing (±)-dinor Hup-A add Hup-A 2.20, 4.43, 7.08, 8.85, and 17.70 μg L^{-1}, according to the ratio of Hup-A peak area to (±)-dinor Hup-A peak area in HPLC, a linear equation $Y = 0.0188X - 0.0069$ was obtained ($r = 0.9988$). The minimal detect limit of plasma Hup-A was 1.60 μg L^{-1}. The recovery of Hup-A was 95.7 ± 5.5 % ($n = 9$) and coefficient of variation was 6.4 %. According to measurements of 3 standard plasma Hup-A concentrations, intraday and interday variances were 5.5 %—7.4 % ($n = 9$) and 6.0 %—8.9 % ($n = 9$), respectively.

RESULTS

The plasma concentrations of Hup-A after oral administration of 0.99 mg within 10 h were fitted well to a one-compartment open model with a first-order absorption (Fig 2).

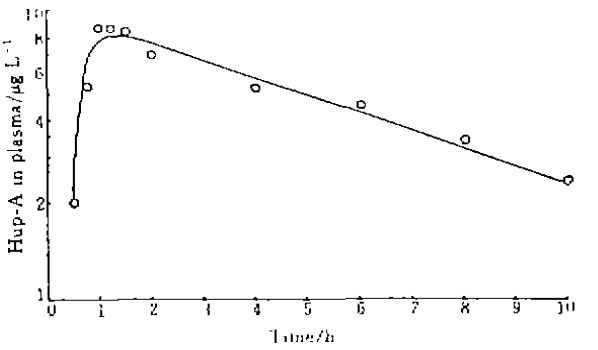

Fig 2. Mean plasma concentration-time curve after *po* tablet Hup-A 0.99 mg in 6 adults.

Hup-A was absorbed quickly after *po* with $T_{\frac{1}{2}k_a} = 12.6$ min and time peak for plasma averaged 79.6 min. It indicated that Hup-A was released and absorbed quite well *in vivo*. Plasma mean peak concentration after *po* was 8.4 μg L^{-1}, V_d/F was 0.108 L kg^{-1}, indicating that Hup-A was widely distributed *in vivo*. Mean elimination half life $T_{\frac{1}{2}k_e}$ was 288.5 min, suggesting that Hup-A have a mild elimination rate (Tab 1).

DISCUSSION

Hup-A showed some advantages, compared with the first generation of ChE inhibitors such as physostigmine (Phy) and tetrahydroaminoacridine (THA), LD_{50} value in mice for Hup-A ip was 1.8 mg kg^{-1} and

Tab 1. Pharmacokinetic parameters of Hup-A after *po* tablet 0.99 mg in 6 healthy volunteers. $\bar{x}\pm s$.

Parameter		$\bar{x}\pm s$
K_a	min^{-1}	0.061±0.017
K_e	min^{-1}	0.0025±0.0006
$T_{\frac{1}{2}K_a}$	min	13±5
$T_{\frac{1}{2}K_e}$	min	288±63
T_{max}	min	80±9
C_{max}	$\mu g\ L^{-1}$	8.4±0.9
T_{lag}	min	25.4±1.8
V_d/F	$L\ kg^{-1}$	0.108±0.008
AUC	$mg\ L^{-1}\ min$	4.1±1.2

that for Phy was 0.6 mg kg^{-1}[3]. Hup-A at optimal doses has a long term inhibition of AChE in rat brain(up to 360 min) and only 60 min for Phy[9]. The results of this paper showed that in human being $T_{\frac{1}{2}K_e}$ of Hup-A was 288.5 min. However, for Phy the $T_{\frac{1}{2}K_e}$ was 20 min[8]. Hup-A was absorbed rapidly, distributed widely in the body and eliminated at a middle rate[7]. Therefore it is better to take tablet Hup-A orally 2—3 times a day.

As a new ChE inhibitor, Hup-A shows some interesting cholinomimetic properties and its effects satisfy more closely established criteria for therapeutic use than effects of previously tested compounds. Hup-A is a new promising ChE inhibitor.

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REFERENCES

1 Liu JS, Zhu YL, Yu CM, Zhou YZ, Han YY, Wu FW, *et al*. The structures of huperzine A and B, two new alkaloids exhibiting marked anticholinesterase activity. Can J Chem 1986; **64**: 837—9.

2 Wang YE, Yue DX, Tang XC. Anti-cholinesterase activity of huperzine A. Acta Pharmacol Sin 1986; **7**: 110—3.

3 Yan XF, Lu WH, Lou WJ, Tang XC. Effects of huperzine A and B on skeletal muscle and elecrroencephalogram. Acta Pharmacol Sin 1987; **8**: 117—23.

4 Xiong ZQ, Tang XC. Effect of huperzine A, a novel acetylcholinesterase inhibitor, on radial maze performance in rats. Pharmacol Biochem Behav 1995; **51**: 415—9.

5 Cheng YS, Lu CZ, Ying ZL, Ni WY, Zhang CL, Sang GW. 128 Cases of myasthenia gravis treated with huperzine A. New Drugs Clin Remedies 1986; **5**: 197—9.

6 Zhang SL. Therapeutic effects of huperzine A on the aged with memory impairment. New Drugs Clin Remedies 1986; **5**: 260—2.

7 Wang YE, Feng J, Lu WH, Tang XC. Pharmacokinetics of huperzine A in rats and mice. Acta Pharmacol Sin 1988; **9**: 193—6.

8 Giacobini E. The second generation of cholinesterase inhibition; pharmacological aspects. In: Becker R, Giacobini E, editors. Cholinergic basis for Alzheimer therapy. Boston; Birkhauser, 1991, 247—262.

9 Tang XC, De Sarno P, Sugaya K, Giacobini E. Effect of huperzine A, a new cholinesterase inhibitor, on the central cholinergic system of the rat. J Neurosci Res 1989; **24**: 276—85.

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石杉碱甲片在六名志愿者体内的药物动力学

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目的：了解石杉碱甲片在人体内的药物动力学过程，为设计临床用药方案提供依据. **方法**：用反相高效液相色谱法测定六名健康志愿者口服片剂0.99 mg后的血药浓度，按3P87程序计算动力学参数. **结果**：石杉碱甲片在体内的药时过程符合一级吸收的一室开放模型. 主要动力学参数：$T_{\frac{1}{2}K_a}$ 12.6 min，$T_{\frac{1}{2}K_e}$ 288.5 min，T_{max} 79.6 min，C_{max} 8.4 $\mu g\ L^{-1}$，AUC 4.1 $mg\ L^{-1}\ min$. **结论**：石杉碱甲吸收迅速，属于中等速率消除类药物.

关键词 石杉碱甲；胆碱酯酶抑制剂；高压液相色谱法；药物动力学；I期临床试验

药代动力学