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Huperzine-A in capsules and tablets for treating patients with Alzheimer disease[1]

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KEY WORDS huperzine-A; cholinesterase inhibitors; Alzheimer disease; double-blind method; randomized controlled trials; memory; cognition disorders; free radicals

ABSTRACT

AIM: To compare the efficacy and safety between huperzine-A (Hup) in capsules and tablets for treating patients with Alzheimer disease (AD). **METHODS**: Using multicenter, prospective, double-blind, double-mimic, parallel, positive controlled and randomized methods, 60 patients meeting with the NINCDS-ARDRA criteria of AD were divided into 2 equal groups. Patients in the capsule group received 4 capsules of Hup (each contains 50 μg) and 4 tablets of placebo (lactose and starch inside); while the tablet group received 4 tablets of Hup (each contains 50 μg) and 4 capsules of placebo, *po*, twice a day for 60 d. All the patients were evaluated with a lot of related ranting scales, and physiological and laboratory examination. **RESULTS**: There were significant differences ($P < 0.01$) on all the psychological evaluations between 'before' and 'after' the 60-d trial of 2 groups, but there was no significant difference between 2 groups by group t test ($P > 0.05$). The changes of oxygen free radicals in 2 groups showed marked improvement. No severe side effect besides moderate to mild nausea was found in both groups. **CONCLUSION**: There is equal efficacy and safety between Hup in capsule and tablet for treating patients with AD, and Hup can reduce the pathological changes of the oxygen free radicals in the plasma and erythrocytes of patients with AD.

INTRODUCTION

The loss of cholinergic neurons of the brain observed in AD is considered an important element of dementia[1], therefore, a series of cholinesterase inhibitors such as physostigmine, galanthamine[2], E2020[3], and tacrine[4], *etc* were studied to supplement the cholinergic function for its symptomatic treatment[3]. Hup, a new *Lycopodium* alkaloid first isolated from Chinese herb *Huperzia serrata* (Thunb) Trev by Chinese[5], is a potential, centrally active and reversible cholinesterase inhibitor with better therapeutic index[6-8], its clinical efficacy and safety were proved by our previous trials[9,10]. This study was to compare the efficacy and safety of Hup in capsules and tablets by more advanced rating scales and, observation of its action on the oxygen free radicals in plasma and erythrocytes.

MATERIALS AND METHODS

Patients Patients ($n = 60$) who met with AD criteria of NINCDS-ADRDA[11] and DSM-Ⅲ R were selected for this study. Their entrance criteria were age ranged from 50 - 80 a; memory quotient (MQ) $\leqslant 70$; mini-mental state examination (MMS) score $\leqslant 23$; Hachinski ishemic scale < 4; depression, severe physical or psychotic disorders, and non-AD dementia were ruled out. Their procurators agreed with the patients to participate in this study, and completed the

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procedure of informed consent.

Methods Patients were abstained from any CNS stimulants, steroids, and nootropies for 2 wk. They were randomly divided into 2 equal groups, patients in Hup capsules group received 4 capsules of Hup (Haboyin, each contains 50 μg, manufactured by Joysun Aggregation/Henan Zhulin Pharmaceutical Company Ltd, Health Reg, No 970312 ZJ) and 4 tablets of placebo (lactose and starch inside); while the Hup tablets group received 4 tablets of Hup (each contains 50 μg, Health Reg, No 961001 HN) and 4 capsules of placebo, *po*, twice a day for 60 d. All the capsules and tablets, either Hup or placebo, were provided by Ningbo Lihua Pharmaceutical Company Ltd. The clinicians and the patients were double blind.

Assessment Blood pressure, heart rate, hemoglobin, white blood cells, blood urea nitrogen, creatinine, alkaline phosphatase, alanine transaminase, P-LPO (plasma lipoperoxide), E-LPO (erythrocyte lipoperoxide), P-SOD (plasma superoxide dismutase), E-SOD (erythrocyte superoxide dismutase), MMS, HDS-R (Hasegawa dementia scale revised), IADL (instrumental activity of daily living), GBS-SDS (Gottfries-Bräne-Steen scale for dementia syndromes), and TESS (treatment emergent symptom scale) were measured monthly; urine routine, ECG, EEG, and WMS (Wechsler memory scale) were measured at the beginning and end of the trial; CGI (clinical global impression) and CCGI (caregiver clinical global impression) were evaluated at the end of trial.

Data analysis The statistical analysis of the results were performed by new drug statistical treatment (NDST) software provided by professor SUN Rui-Yuan, paired t test was used for all the measured data before and after the trial; group t test was used for comparison between 2 groups. The clinical efficacy identified by MQ, MMS, CGI, and CCGI were compared between 2 groups with Ridit analysis or χ^2 method.

Duration of trial From 1997-04-01 to 1998-03-30.

RESULTS

The blind was declassified on 1998-03-30 in Ningbo Thirty patients were in capsule group, and 30 patients in tablet group. The pretreatment data between the 2 groups showed no significant difference (Tab 1).

Tab 1. Background data between the 2 groups of AD. n = 30. All data showed no statistical significance between capsule and tablet groups.

	Tablet	Capsule
Male	13	10
Female	17	20
Age/a	72 ± 10	71 ± 6
Range	52 – 80	55 – 79
Occupation		
Cadre	5	6
Technician	4	2
Peasant	4	8
Worker, miscellaneous	17	14
Culture		
College	0	3
Senior high	5	4
Junior high	4	5
Elementary	21	18
Marriage		
Married	18	19
Unmarried	1	0
Divorced	1	0
Single	10	11
Course/a	2.7 ± 0.8	2.4 ± 1.1
< 2	4	10
2 – 4	22	16
≥ 4	4	4
MQ baseline	43 ± 12	40 ± 13
MMS baseline	15 ± 4	13 ± 5
Identified cerebral atrophy by CT or MR	17 (57 %)	15 (50 %)

MQ: memory quotient; MMS: mini-mental state examination.

Psychological assessment MQ, MMS, and HDS-R were increased by 8 ± 10, 5 ± 4, and 4 ± 4 in capsule group, and 6 ± 10, 4 ± 4, and 4 ± 4 in tablet group respectively, IADL and GBS-SDS were decreased by 2.2 ± 2.4 and 11 ± 12 in the former, 2.6 ± 2.7, and 15 ± 15 in the latter respectively. Both of 2 groups showed significant differences on MQ, MMS, HDS-R, IADL and GBS-SDS between 'before' and 'after' the 60-d Hup treatment ($P < 0.01$), but there was no significant difference on them between 2 groups ($P > 0.05$, Tab 2).

The intraclass correlation (ICC) ICC on MMS, HDS-R, IADL, and GBS-SDS from 4 evaluators were 0.92, 0.93, 0.93, and 0.89 sequentially ($P > 0.05$).

Oxygen free radical changes Both of 2

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Tab 2. Comparison of all the psychological rating score between 2 groups of AD. $n = 30$.
$^cP < 0.01$ *vs* before treatment. $^dP > 0.05$ *vs* tablet.

Item	Tablet	Capsule
MQ baseline	43 ± 12^d	40 ± 13
60-d trial	49 ± 16^{cd}	48 ± 17^c
MMS baseline	15 ± 4^d	13 ± 5
30-d trial	16 ± 5^{cd}	14 ± 7^c
60-d trial	19 ± 6^{cd}	18 ± 8^c
HDS-R baseline	13 ± 5^d	11 ± 5
30-d trial	15 ± 6^{cd}	13 ± 6^c
68-d trial	17 ± 7^{cd}	16 ± 8^c
IADL baseline	22 ± 6^d	21 ± 5
30-d trial	21 ± 6^{cd}	20 ± 5^c
60-d trial	19 ± 6^{cd}	19 ± 6^c
GBS-SDS baseline	51 ± 21^d	52 ± 23
30-d trial	44 ± 22^{cd}	47 ± 22^c
60-d trial	36 ± 24^{cd}	40 ± 25^c

MQ: memory quotient; MMS: mini-mental state examination; HDS-R: Hasegawa dementia scale revised; IADL: instrumental activity of daily living; GBS-SDS: Gottfries-Bräne-Steen scale for dementia syndromes

groups displayed significant differences on P-LPO, E-LPO, P-SOD, and E-SOD between 'before' and 'after' the 30-d or 60-d Hup treatment ($P < 0.01$) (Tab 3). Comparison of these free radical changes between 2 groups showed no significant differences ($P > 0.05$) (Tab 3).

Tab 3. Comparison of P-SOD, E-SOD, P-LPO, and E-LPO between 2 groups of AD. $n = 30$.
$^cP < 0.01$ *vs* before treatment. $^dP > 0.05$ *vs* tablet.

Item		Tablet	Capsule	RV
P-SOD/	baseline	23 ± 9^d	25 ± 10	35 ± 10
$kU \cdot L^{-1}$	30-d trial	25 ± 10^{cd}	27 ± 10^c	
	60-d trial	27 ± 10^{cd}	30 ± 10^c	
E-SOD/	baseline	1644 ± 187^d	1693 ± 204	1913 ± 189
$U \cdot g^{-1}$(Hb)	30-d trial	1681 ± 190^{cd}	1733 ± 209^c	
	60-d trial	1719 ± 195^{cd}	1797 ± 221^c	
P-LPO/	baseline	13.9 ± 2.2^d	14.4 ± 2.4	11.9 ± 1.8
$\mu mol \cdot L^{-1}$	30-d trial	13.5 ± 2.3^{cd}	13.9 ± 2.3^c	
	60-d trial	12.9 ± 2.3^{cd}	13.2 ± 2.4^c	
E-LPO/	baseline	39 ± 4^d	40 ± 5	34 ± 4
$nmol \cdot g^{-1}$	30-d trial	38 ± 4^{cd}	39 ± 5^c	
(Hb)	60-d trial	37 ± 4^{cd}	38 ± 5^c	

RV: reference value of the health aged; P-SOD: plasma superoxide dismutase; E-SOD: erythrocyte lipoperoxide; P-LPO: plasma lipoperoxide; E-LPO: erythrocyte lipoperoxide.

Clinical efficacy evaluation The improvement rates of both groups ranged from 43 % to 70 % (Tab 4).

Tab 4. Comparison of clinical efficacy between 2 groups of AD. $n = 30$.

Verdict methods	Tablet	Capsule	u (Ridit)	χ^2
MQ improvement				
≥20 % (marked improvement)	9	9		
11 % – 19 % (improvement)	6	7	0.16	
≤10 % (less effective)	15	14		
MMS improvement				
≥3 points (response)	15	21		
<3 points (non-response)	15	9		2.5
CGI (marked improvement)	0	1		
(improvement)	16	12	0.34	
(less effective)	12	17		
(worse)	2	0		
CCGI (improvement)	18	16		
(less effective)	11	12	1.61	
(worse)	1	2		
The range of improved rate	50 % – 60 %	43 % – 70 %		

MQ: memory quotient; MMS: mini-mental state examination; CGI: clinical global impression; CCGI: caregiver clinical global impression.

Comparison of the clinical efficacy between 2 groups with Ridit analysis or χ^2 method displayed no significant difference ($P > 0.05$). The psychological evaluating odds of 2 groups were qualified with equivalent test (Tab 5).

Tab 5. Comparison of psychological evaluating odds between 2 groups of AD by equivalence examination (EE). $n = 30$. All EE were qualification.

Item	Tablet	Capsule	Max permissible odds
MQ	6 ± 10	8 ± 10	2.53
MMS	4 ± 4	5 ± 4	0.90
HDS-R	4 ± 4	4 ± 4	0.92
IADL	-2.6 ± 2.7	-2.2 ± 2.4	1.05
GBS-SDS	-15 ± 15	-11 ± 12	5.50

MQ: memory quotient; MMS: mini-mental state examination; HDS-R: Hasegawa dementia scale revised; IADL: instrumental activity of daily living; GBS-SDS: Gottfries-Bräne-Steen scale for dementia syndromes.

Side effects The incidence of mild to moderate nausea was 23.3 % – 33.3 % at the 30-d Hup

treatment, 13.3 % – 20 % at the 60-d Hup treatment; the incidence of mild to moderate insomnia was 23.3 % – 26.6 % and 13.3 % – 20.0 %, but there was no significant difference between 2 groups ($P > 0.05$). No any laboratory and physiological change was found in the 60-d Hup treatment (Tab 6, 7).

Tab 6. Comparison of TESS score between 2 groups of AD. $n = 30$. $^cP < 0.01$ *vs* before treatment. $^dP > 0.05$ *vs* tablet.

TESS	Tablet	Capsule
Baseline	1 ± 4^d	0.6 ± 2.3
30-d trial	2 ± 3^{cd}	1.7 ± 2.7^c
60-d trial	1.4 ± 2.4^{cd}	1.1 ± 2.5^c

TESS: treatment emergent symptom scale.

Tab 7. Comparison of side effect between 2 groups of AD. $n = 30$. All data showed no statistical significance between the capsule and tablet group.

Symptom	Tablet						Capsule					
	30-d trial			60-d trial			30-d trial			60-d trial		
	Ⅰ	Ⅱ	Ⅲ	Ⅰ	Ⅱ	Ⅲ	Ⅰ	Ⅱ	Ⅲ	Ⅰ	Ⅱ	Ⅲ
Nausea and vomiting	5	0	2	4	0	0	6	3	1	6	0	0
Insomnia	5	3	0	5	1	0	6	1	0	3	1	0
Fainting	4	0	0	1	0	0	2	1	0	2	0	0
Anorexia	3	0	1	2	0	0	3	0	1	2	0	0
Exciting	3	0	0	2	0	0	2	0	0	1	0	0
Visual blurring	2	0	0	2	0	0	1	0	0	1	0	0
Miscellaneous	22	1	0	14	0	0	19	2	0	13	1	0

Ⅰ: slight or mild; Ⅱ: moderate; Ⅲ: severe.

DISCUSSION

This study was designed as a double-mimic trial in order to avoid the visual influences of the clinicians and patients. According to high ICC in evaluators, multicenter, parallel, double-blind, positive controlled, randomized method and many advanced psychological rating scales for referring, and comparable background data between 2 groups, as same as our previous studies[9,10], the results of this study are also reliable.

There was no significant difference on all the evaluations between 2 groups ($P > 0.05$), at the same time, they were also qualified with equivalent test[12], so we consider that the efficacy of Hup in capsule and in tablet is equal. In other words, Hup is sure a promising drug for symptomatic treatment of AD.

From the viewpoint of free radical theory, the balance between oxidation and antioxidation in the aging or atrophy illness are serious imbalances[13], whereas AD includes these two involving factors, whether the imbalance would exist in the patients with AD? Whether Hup could reduce these changes? it had been not reported at home and abroad, therefore we tried to inspect these changes in the patients with AD. In this study, although we found increment of P-SOD and E-SOD, and decrement of P-LPO and E-LPO in patients with AD during the Hup treatment, there was a distance from the reference value of healthy aged yet ($P < 0.01$). In other words, Hup could only correct these pathological changes partially, it suggests that symptomatic treatment in patients with AD must be undertaken for long.

It is considered generally that any cholinesterase inhibitor without higher affinity for the central nervous system might inhibit general acetylcholinesterase activity, resulting in significant peripheral side effects[14]. In this study, owing to the strict design and wide observation along with the skilled clinicians participated in previous similar trial, the incidence of peripheral cholinergic side effects of Hup revealed was up to 33 %. Although they belonged to almost mild to moderate degree in the early stage of trial, and most of them diminished or disappeared at the end of trial, the fact must be still paid attention. We think that the grading administration of Hup at the beginning for reducing the peripheral cholinergic side effects is essential. No any lethal side effect was found in the whole duration, and all of the physiological and laboratory examination showed no change, it illustrates that Hup is a relatively safe drug for long-term treatment of patients with AD.

By reason that the samples of this study are not enough large, and there is no long-term observation and no direct clinical comparison with other cholinesterase inhibitors, the further studies will be undertaken to complete methods for confirming the efficacy and safety of Hup.

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石杉碱甲胶囊及片剂治疗阿尔采末病[1]

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关键词 石杉碱甲; 胆碱酯酶抑制剂; 阿尔采末病; 双盲法; 随机对照实验; 记忆; 认知障碍; 自由基

目的: 比较石杉碱甲胶囊和片剂对阿尔采末病的疗效和安全性. **方法**: 采用双盲双模拟法将入选的60例阿尔采末病人随机等分两组, 胶囊组每次口服石杉碱甲胶囊4粒和安慰剂片4片; 片剂组每次口服石杉碱甲片(哈伯因)4片和安慰剂胶囊4粒, 两组均每日两次, 60日为一疗程. 所有病人都测量WMS, MMS, HDS – R, IADL, GBS痴呆综合征量表和各种检查以及氧自由基. **结果**: 两组所有心理量表值疗后均有显著改善($P < 0.01$), 而两组间无明显差异($P > 0.05$), 且等效检验合格; 两组的自由基病理变化疗后也明显减轻, 但也无组间差异($P > 0.05$); 两组未发生严重不良反应. **结论**: 石杉碱甲胶囊和片剂治疗阿尔采末病时疗效和安全性相等.

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