Research Article

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# The efficacy of butylphthalide soft capsules combined with donepezil tablet in patients with alzheimer disease and its effect on Serum A beta, GSH-Px and SOD

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**Objective:** To investigate the efficacy of Butylphthalide Soft Capsules combined with donepezil tablet in patients with Alzheimer disease (AD) and its effect on serum amyloid beta (Aβ), glutathione peroxidase (GSH-Px), superoxide dismutase (SOD) and inflammatory cytokines.

**Methods:** A total of 104 patients with AD enrolled in our hospital from January 2015 to January 2017 were selected as the subjects. According to the random number table, they were divided into observation group and control group, 52 cases in each group. Patients in both groups were given conventional treatment. On such basis, the control group were given oral medication of donepezil each day, besides which, the observation group were treated with Butylphthalide Soft Capsules. The clinical efficacy and adverse reactions in the 2 groups were observed and the levels of serum Aβ, GSH-Px, SOD and inflammatory cytokines (IL-1β, IL-6 and TNF-α), as well as the changes in the cognitive function and the activity of daily living were compared between the two groups before and after treatment.

**Results:** Before treatment, there was no statistically significant difference between the 2 groups in the levels of serum Aβ, GSH-Px and SOD (P>0.05), while after treatment, the Aβ level in the 2 groups was significantly lower than that before treatment (P<0.05), obviously lower in the observation group than in the control group (P<0.05), and the GSH-Px and SOD levels in the 2 groups were significantly higher than those before treatment (P<0.05), obviously higher in the observation group than in the control group (P<0.05). Before treatment, there was no significant difference in the levels of IL-1β, IL-6 and TNF-αbetween the 2 groups (P>0.05), while after treatment, the levels of IL-1β, IL-6 and TNF-αin the 2 groups were significantly lower than those before treatment (P<0.05), obviously lower in the observation group than in the control group (P<0.05). Before treatment, there was no significant difference in the scores of the mini-mental state examination (MMSE) and ADL between the 2 groups (P>0.05), while after treatment, the scores of MMSE and ADL in the 2 groups were significantly higher than those before treatment (P<0.05), obviously higher in the observation group than in the control group (P<0.05). The total effective rate of the observation group was 94.23%, which was significantly higher than that of the control group (76.92%), and the difference was statistically significant (P<0.05). During treatment the incidence of adverse reactions in the control group was 9.62%, higher than that in the observation group, that was 7.69%, but the difference was not statistically significant.

**Conclusion:** The treatment of Butylphthalide Soft Capsules with donepezil tablet leads to few side effects and can improve the levels of serum Aβ, GSH-Px and SOD, reduce inflammatory cytokines and enhance cognitive function as well as the activities of daily living in patients with AD, thus worthy of clinical application.

**Keywords: butylphthalide soft capsules ▪ donepezil tablet ▪ alzheimer disease ▪ GSH-Px ▪ superoxide dismutase ▪ inflammatory cytokines**

## Introduction

Alzheimer disease (AD) is a degenerative disease of the central nervous system in the early or middle stage in the elderly, mainly characterized by behavioral dysfunction as well as progressive cognitive impairment and manifested as language fluency dysfunction, visual spatial dysfunction, memory dysfunction, personality changes and behavior changes, which can significantly

affect the activities of daily living and reduce the quality of life in patients [1]. In recent years, with the increasingly rising longevity and the severity of aging population in China, the incidence of AD is increasing year by year. The pathogenesis of AD is complex and there is no special therapeutic method in clinic in this regard. Therefore, strengthening the research on AD and seeking for more effective drugs and treatment methods are of great medical and social significance, becoming a hot topic in the field of medicine [2,3]. The disease is currently given conservative treatment in which butylphthalide is commonly used [4]. Related studies have confirmed that [5] butylphthalide is a drug for cerebral microcirculation reconstruction and that in the early stage it can significantly improve the dementia severity in AD patients with mild or moderate cognitive impairment, enhance their cognitive level and improve their memory function yet with great side effects. Donepezil is a cholinesterase inhibitor [6]. In recent years, the treatment of AD with combined drugs has attracted more and more attention. At present, there are few reports about the efficacy of Butylphthalide Soft Capsules combined with donepezil in the treatment of AD in serology. In this study Butylphthalide Soft Capsules plus donepezil is adopted to treat AD, aiming to seek suitable methods for treatment of the disease, the details now reported as follows.

## Data and methods

### General data

A total of 104 patients with AD enrolled in our hospital from January 2015 to January 2017 were selected as the subjects.

*Inclusion criteria*

i. patients met the diagnostic criteria for AD proposed in "the Chinese Classification and Diagnosis of Mental Diseases;

ii. patients were 50 years old or older;

iii. patients signed informed consent and were approved by the hospital ethics committee.

*Exclusion criteria*

i. patients had vascular or Louis dementia;

ii. patients had the dementia induced by brain injury or other factors;

iii. patients had serious heart, liver or kidney disease;

iv. patients had a history of cerebrovascular disease or brain surgery:

v. patients took other drugs for AD treatment in recent 4 week:

vi. patients had serious drug allergy history or history of alcohol abuse. Among the selected 104 patients, there were 60 males and 44 females, aged 52~80 with an average age of (70.6 ± 2.7) years and a disease course of 3~10 years, (7.5 ± 1.3) years on average; the dementia severity was determined with clinical dementia rating (CDR) and it turned out that there were 30 mild cases, 60 moderate cases and 14 severe cases. According to the random number table, the subjects were divided into the observation group and the control group, 52 cases in each group, and there was no significant difference in the general data between the 2 groups (P>0.05), as shown in **TABLE 1**.

### Methods

All patients experienced 2 weeks washout period and were given routine treatments including neurotrophic treatment and lipid lowering treatment. Patients in the control group took an oral of donepezil (donepezil film-coated tablets, specifications: 5 mg/tablet, 5 mg Eisai (manufactured by Pharmacy Co. Ltd., China), once a day; Besides the treatment in the control group, patients in the observation group were additionally treated with oral administration of 0.2 g Butylphthalide Soft Capsules (specifications: 0.1 g/capsule, manufactured by CSPC Butylphthalide Pharmaceutical Co., Ltd.), 3 times a day. Patients in the 2 groups were treated for 9 months.

*Observation index*

i. The clinical efficacy was evaluated according to the rate of MMSE score improvement [MMSE score improvement rate = (the score before treatment-the score after treatment)/the score before treatment]. The improvement rates of over 20%, 10%~20% and below 10% were respectively defined as significantly effective, effective and ineffective

treatment, and the total effective rate is a sum of significantly effective rate and effective rate.

ii. The cognitive function was evaluated before and after treatment by the MMSE scale, including the evaluations of computing power, directional power, application and verbal skill, memory and visual space, the full score was 30 points; the higher the score, the better the cognitive function of patients.

iii. The activity of daily living was assessed with Activity of Daily Living Scale (ADL) respectively before and after treatment, including the assessments on physical self-maintenance such as dressing, going upstairs and downstairs, bathing and grooming as well as instrumental activities of daily living such as doing the laundry, taking transportation and going shopping, the full score was 64 points, the higher the score, the better the patient's ADL.

*Enzyme linked immunosorbent assay (ELISA)*

The levels of serum amyloid β (Aβ), glutathione peroxidase (GSH-Px) and superoxide dismutase (SOD) were measured before and after treatment respectively; the elbow vein blood (5 mL) was extracted from patients on an empty stomach, centrifuged at the speed of 2000r/min, and then the upper serum was taken for detection of Aβ, GSH-Px and SOD with Aβ kit and enzyme-linked immunosorbent assay, with the whole process operated strictly according to the kit instructions. The changes in the levels of serum inflammatory cytokines such as IL-1β, IL-6 and TNF-αwere assessed with ELISA method and were compared in the two groups before and after treatment.

*Statistical analysis*

The data analysis was carried out by SPSS 21 statistics software. The measurement data were expressed as "$\overline{x} \pm s$" and assessed by t test. The count data were represented by the number of case or rate, and rank sum test or $x^2$ test were used in comparing between groups, P<0.05 suggested that there was statistically significant difference.

## Results

### Comparison of clinical efficacy between the two groups

The total effective rate of the observation group was 94.23%, which was significantly higher than that of the control group (76.92%), and the difference was statistically significant (P<0.05), as shown in **TABLE 2**.

### Comparison of the levels of serum Aβ, GSH-Px and SOD between the 2 groups

Before treatment, there was no statistically significant difference between the 2 groups in the levels of serum Aβ, GSH-Px and SOD(P>0.05), while after treatment, the Aβlevel in the 2 groups was significantly lower than that before treatment (P<0.05), obviously lower in the observation group than in the control group(P<0.05), and the GSH-Px and SOD levels in the 2 groups were significantly higher than those before treatment (P<0.05), obviously higher in the observation group than in the control group(P<0.05), as shown in **TABLE 3**.

**Table 1. Comparison of the basic clinical data of two groups of patients**

| Groups | n | Gender (Male/Femal) | Age (Year) | Course of disease (Year) | Mild cases | Moderate cases | Severe cases |
|---|---|---|---|---|---|---|---|
| Control group | 52 | 24/28 | 69.1 ± 2.6 | 7.7 ± 1.2 | 16 | 24 | 6 |
| Observation group | 52 | 25/27 | 71.8 ± 3.2 | 7.6 ± 1.4 | 14 | 26 | 8 |
| F | - | 1.352 | 1.242 | 1.285 | 1.062 | 1.159 | 1.256 |
| p | - | > 0. 05 | > 0. 05 | > 0. 05 | > 0. 05 | > 0. 05 | > 0. 05 |

**Table 2 Comparison of clinical efficacy between the two groups [n (%)]**

| Groups | n | Significantly effective | Effective | Ineffective | Effective rate |
|---|---|---|---|---|---|
| Observation group | 52 | 33(63.46) | 16(30.77) | 3(5.77) | 49(94.23) |
| Control group | 52 | 22(42.31) | 18(34.62) | 12(23.08) | 40(76.92) |
| $X^2$ | - | - | - | - | 5.045 |
| P | - | - | - | - | 0.028 |

### Comparison of serum levels of inflammatory cytokines between the two groups

Before treatment, there was no significant difference in the levels of IL-1β, IL-6 and TNF-αbetween the 2 groups (P>0.05), while after treatment, the levels of IL-1β, IL-6 and TNF-αin the 2 groups were significantly lower than those before treatment (P<0.05), obviously lower in the observation group than in the control group(P<0.05), as shown in **TABLE 4**.

### Comparison of MMSE and ADL scores between the two groups

Before treatment, there was no significant difference in the scores of MMSE and ADL between the 2 groups (P>0.05), while after treatment, the scores of MMSE and ADL in the 2 groups were significantly higher than those before treatment (P<0.05), obviously higher in the observation group than in the control group(P<0.05), as shown in **TABLE 5**.

### Comparison of adverse reactions between the two groups

During treatment the incidence rate of adverse reactions in the control group was 9.62%, higher than that in the observation group, that was 7.69%, but the difference was not statistically significant, as shown in **TABLE 6**.

## Summary

Before treatment, there was no statistically significant difference between the 2 groups in the levels of serum Aβ, GSH-Px and SOD (P>0.05), while after treatment, the Aβ level in the 2 groups was significantly lower than that before treatment (P<0.05), obviously lower in the observation group than in the

**Table 3 Comparison of the levels of serum Aβ, GSH-Px and SOD between the 2 groups ( $\bar{x} \pm s$ )**

| Groups | n | Aβ(ng/L) | | GSH-Px(U/mL) | | SOD(U/mL) | |
|---|---|---|---|---|---|---|---|
| | | Before treatment | After treatment | Before treatment | After treatment | Before treatment | After treatment |
| Observation group | 52 | 87.54 ± 9.05 | 67.36 ± 7.09 | 82.51 ± 8.06 | 132.17 ± 9.65 | 60.78 ± 3.17 | 85.33 ± 6.47 |
| Control group | 52 | 87.61 ± 9.14 | 74.47 ± 7.12 | 82.48 ± 8.11 | 107.24 ± 9.34 | 60.73 ± 3.06 | 69.26±5.28 |
| t | - | 0.448 | 4.093 | 0.159 | 7.892 | 0.209 | 9.003 |
| P | - | 0.152 | 0.035 | 0.195 | 0.017 | 0.674 | 0.012 |

**Table 4 Comparison of serum levels of inflammatory cytokines between the two groups( $\bar{x} \pm s$ , pg/ml)**

| Groups | n | IL-1β | | IL-6 | | TNF-α | |
|---|---|---|---|---|---|---|---|
| | | Before treatment | After treatment | Before treatment | After treatment | Before treatment | After treatment |
| Observation group | 52 | 35.78 ± 4.16 | 26.45 ± 3.09 | 6.76 ± 1.03 | 4.53 ± 0.87 | 148.94 ± 12.57 | 106.15 ± 9.44 |
| Control group | 52 | 35.59 ± 4.22 | 30.32 ± 3.15 | 6.71 ± 1.04 | 5.76 ± 0.68 | 148.87 ± 11.16 | 127.49 ± 8.06 |
| t | - | 0.902 | 5.672 | 0.165 | 5.053 | 0.542 | 8.776 |
| P | - | 0.133 | 0.031 | 0.134 | 0.019 | 0.601 | 0.016 |

**Table 5 Comparison of MMSE and ADL scores between the two groups ( $\bar{x} \pm s$ , points)**

| groups | n | MMSE | | ADL | |
|---|---|---|---|---|---|
| | | Before treatment | After treatment | Before treatment | After treatment |
| Observation group | 52 | 15.35 ± 2.86 | 21.57 ± 2.04 | 37.94 ± 3.05 | 46.56 ± 3.32 |
| Control group | 52 | 15.27 ± 2.95 | 17.16 ± 2.48 | 37.87 ± 3.12 | 43.47 ± 3.57 |
| t | - | 0.782 | 5.676 | 0.924 | 4.985 |
| P | - | >0.05 | <0.05 | >0.05 | <0.05 |

**Table 6. Comparison of adverse reactions between the two groups [n (%)]**

| groups | n | Increased transaminase | Fatigue and lassitude | Gastrointestinal reactions | Skin rashes | Total rate of adverse reactions |
|---|---|---|---|---|---|---|
| Observation group | 52 | 2(3.85) | 1(1.92) | 1(1.92) | 0(0) | 4(7.69) |
| Control group | 52 | 2(3.85) | 1(1.92) | 1(1.92) | 1(1.92) | 5(9.62) |
| $X^2$ | - | - | - | - | - | 1.204 |
| P | - | - | - | - | - | 0.056 |

control group(P<0.05), and the GSH-Px and SOD levels in the 2 groups were significantly higher than those before treatment (P<0.05), obviously higher in the observation group than in the control group (P<0.05); before treatment, there was no significant difference in the levels of IL-1β, IL-6 and TNF-αbetween the 2 groups (P>0.05), while after treatment, the levels of IL-1β, IL-6 and TNF-αin the 2 groups were significantly lower than those before treatment (P<0.05), obviously lower in the observation group than in the control group(P<0.05); before treatment, there was no significant difference in the scores of MMSE and ADL between the 2 groups (P>0.05), while after treatment, the scores of MMSE and ADL in the 2 groups were significantly higher than those before treatment (P<0.05), obviously higher in the observation group than in the control group (P<0.05); the total effective rate of the observation group was 94.23%, significantly higher than that of the control group (76.92%), and the difference was statistically significant (P<0.05); and during treatment the incidence of adverse reactions in the control group was 9.62%, higher than that in the observation group, that was 7.69%, but the difference was not statistically significant. These results suggest that the treatment of Butylphthalide Soft Capsules with donepezil tablet leads to few side effects, can improve the levels of serum Aβ, GSH-Px and SOD, reduce inflammatory cytokines and enhance cognitive function as well as the activities of daily living in patients with AD and is thus worthy of clinical application.

## Discussion

AD is a common type of dementia in clinic and has such main pathological changes as senile plaque, reduced nerve cells and neurofibrillary tangles. Data show that [7,8] cerebrovascular disease is closely associated with AD, which can accelerate the process of neurodegeneration. Cerebral ischemia can lead to cerebral vascular dysfunction followed by reduced neovascularization, thereby causing disorders of blood flow regulation and decreasing brain perfusion. The occurrence of AD is related to abnormal blood brain barrier and microvascular degeneration. Aβ is the core component of senile plaques with its generation and degradation in equilibrium in the brain, and the imbalance, if there is, can lead to abnormal Aβ deposition, accelerate microvascular degeneration and seriously damage the blood-brain barrier with its ability to scavenge Aβlowered, and aggravates cerebral vascular injury as well as the condition of AD by reactions [9]. In addition, the abnormal deposition of Aβ can accelerate inflammatory reactions in the brain, activate a large number of inflammation factors and result in neuronal apoptosis, synaptic loss as well as nerve fiber tangles, ultimately triggering AD. Oxidative stress plays an important role in the development of AD. Patients with AD have a high level of free radical, which can promote the accelerated oxidation of membrane lipid so as to damage the cell membrane and cause the apoptosis of neurons. GSH-Px can facilitate the decomposition of glutathione (GSH), reduce the toxic peroxides and transform them into non-toxic hydroxyl compounds so as to alleviate the damage of free radicals to cell membrane [10]. SOD can reduce the injury of membrane structure by removing and inhibiting free radicals [11].

Donepezil can improve the cholinergic nerve function in AD patients and effectively delay the progression of the disease [12]. Ding Bentai, as a new anti-cerebral ischemia drug, can effectively treat the brain dysfunction [13]. By acting on the microcirculation, it enables to effectively protect the mitochondrial function, improve cerebrovascular prostacyclin levels, reduce the calcium level in the brain, enhance the antioxidant effects and inhibit free radicals as well as platelet aggregation so as to reduce the abnormal deposition of Aβ, protect the blood-brain barrier, accelerate the neovascularization, enhance brain energy metabolism, increase cerebral blood flow, inhibit neuronal inflammation and increase synapse. The results of this study show that the MMSE and ADL scores in the observation group were better than those in the control group, suggesting that Butylphthalide Soft Capsules combined with donepezil tablet can effectively improve the cognitive function and daily living function of AD patients. It is also found that the levels of GSH-Px, SOD and Aβ in the observation group were better than those in the control group, suggesting that Butylphthalide Soft Capsules combined with donepezil tablet can improve the clinical efficacy by improving the serum levels of GSH-Px, SOD and Aβ.

In recent years, the inflammatory mechanism of AD has been paid more and more attention, and some people suggest that chronic inflammatory reaction may be another important pathological feature. In the inflammatory cytokine network, the balance between pro inflammatory cytokines and anti-inflammatory cytokines is the key to moderate immune response in the body, and IL-1β, IL-6 as well as TNF-αare considered as typical proinflammatory cytokines. Research reveals that [14] the release of proinflammatory mediators can promote apoptosis of hippocampal neurons, inhibit long-term potentiation (LTP) and then lead to cognition disorders, and that such pro-inflammatory mediators as IL-1β, IL-6 and TNF-α affect the central nervous function either directly or indirectly, in whcih IL-1β and TNF-α can enter the brain through the blood-brain barrier in ventricle and then induce neuroinflammation. It is revealed that the stimulation of TNF-αcan lead to decrease of PPARγ; Furthermore, it has been proved that [15] PPARγ ligand has anti-inflammatory effects and that the activation of PPARγ can reduce the inflammation response with lower expression of inflammatory factors. It is also shown that the expression of PPARγ is decreased with the increase of IL-1β and TNF-α, suggesting that inflammatory factors may down-regulate the expression of PPARγ. This study found that compared with the control group, after treatment the serum levels of IL-1β, IL-6, TNF-αwere significantly decreased in the observation group with the difference being statistically significant. These results indicate that patients with AD may suffer from inflammatory reactions of varying degree, and the treatment of Butylphthalide Soft Capsules combined with donepezil tablets, by activating PPARγ, can inhibit the expression of inflammatory cytokines and reduce neuroinflammation so as to delay the progress of AD course and improve the patients' cognition.

AD need long-time oral medication, during which some patients may suffer adverse drug reactions such as the increase of transaminase, fatigue, gastrointestinal reaction and skin rashes, the results of this study show that the incidence rate of the above adverse reactions in the observation group is not significantly different from that in the control group, suggesting that the treatment with Butylphthalide Soft Capsules and donepezil tablets dose not lead to more adverse drug reactions. the limitation of sample size and treatment course, there also exists some deficiencies of the study and needs further modification and improvement in the future.

To sum up, the treatment of Butylphthalide Soft Capsules with donepezil tablet leads to few side effects and can improve the levels of serum Aβ, GSH-Px and SOD, reduce inflammatory cytokines and enhance cognitive function as well as the activities of daily living in patients with AD, thus worthy of clinical application.

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