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STUDY PROTOCOL

Effectiveness and Safety of Shenxiong Huanglian Detoxification Granule Combined with Donepezil for the Treatment of Alzheimer's Disease: Study Protocol for a Multicenter, Pragmatic, Randomized Controlled Clinical Trial

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Background: Alzheimer's disease is a degenerative condition that causes patients to experience progressive memory decline and a significant decline in overall cognitive ability at any given moment. The increase in the elderly population has resulted in a notable surge in the prevalence of Alzheimer's disease, as has the global impact of the disease. Significant clinical efficacy of traditional Chinese medicine in combination with Western medicine for the treatment of Alzheimer's disease has been demonstrated in previous studies. The main purpose of this trial is to assess the effectiveness and safety of Shenxiong Huanglian Detoxification Granule combined with donepezil in individuals diagnosed with mild-to-moderate Alzheimer's disease.

Methods: This is a multicenter, pragmatic, randomized controlled trial. A total of 386 eligible individuals with mild to moderate Alzheimer's disease will receive random assignment and equal access to the test or control group. The effectiveness and safety of Shenxiong Huanglian Detoxification Granule in combination with donepezil will be observed. The primary outcome is the alteration in scores acquired from the Alzheimer's Disease Assessment Scale-Cognitive Subscale. Secondary outcomes include the assessments of the Traditional Chinese Medicine Syndrome score scale, Mini-Mental State Examination, Clinical Dementia Rating, and Activity of Daily Living scale. We will also analyze blood biomarkers of Alzheimer's disease, inflammatory indicators, oxidative stress indicators, and hemorheology indicators. In addition, safety assessments will be conducted at baseline, after 12 weeks, and after 24 weeks of treatment.

Discussion: These findings will offer reliable clinical evidence regarding the effectiveness and safety of Shenxiong Huanglian Detoxification Granule in combination with donepezil for treating patients with mild-to-moderate Alzheimer's disease. Additionally, this study will support the integration of traditional Chinese and Western medicine into mainstream treatment for Alzheimer's disease, promoting a multitarget strategy.

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Introduction

Alzheimer's disease (AD), is a long-term degenerative condition of the brain that primarily impacts the elderly population. This condition is marked by the progressive deterioration of brain function, which involves the loss of memory, alterations in behavior, and a decrease in cognitive capabilities.[1] Statistics from the World Health Organization show that there are presently more than 55 million instances of dementia globally, with nearly 10 million new cases emerging annually. Among these, AD comprises 60–70% of the cases.[2] Given the rising occurrence of AD and the subsequent disease burden, it is crucial to promptly achieve AD prevention, delay disease progression, and symptom enhancement via effective treatment.

According to the dominant perspective, the accumulation of neurofibrillary tangles caused by excessive phosphorylation of tau within neurons, along with the extracellular deposition of amyloid β (Aβ), is considered the primary factor in the development of AD. This process is accompanied by various intricate pathological alterations, including synaptic dysfunction, neuroinflammatory reactions, and loss of neurons.[3] A variety of drugs, such as memantine, galantamine, and donepezil, have been approved for the treatment of different pathological conditions. These drugs can merely delay the progression of AD and have no direct impact on the fundamental pathophysiology of the ailment.[4] Furthermore, single-target treatments often do not improve cognitive function.[5] The distinct benefits of various targets and pathways in traditional Chinese medicine (TCM) can compensate for the lack of currently available medications. Studies have demonstrated the notable advantage of combining Chinese herbal medicine with conventional drugs for treating AD, as opposed to using only conventional treatments. This combination has shown a stronger impact on symptoms as time progresses.[6]

According to TCM theory, the interaction of kidney deficiency, blood stasis, and toxins is the key to the pathogenesis of AD.[7] Shenxiong Huanglian Detoxification Granule (SHDG) was created by Professor Li Hao, a Chinese scholar of Qihuang, based on this theory and the therapeutic principle of tonifying deficiency and damage, activating blood circulation and detoxifying. The traditional herbal formula is composed of Renshen (*Panax ginseng*) 9g, Chuanxiong (*Ligusticum wallichii*) 6g, Huanglian (*Coptis chinensis*) 5g, Zhizi (*Gardenia jasminoides Ellis*) 5g, and Bingpian (*Borneolum Syntheticum*) 0.15g. The dosage of the above five herbs is within the dosage range specified in the Chinese Pharmacopoeia.[8] Renshen in the formula can greatly invigorate vitality, dispel evil Qi, improve intelligence and strengthen detoxification, Huanglian and Zhizi can clear away heat and dampness, purge fire and detoxify, Bingpian can awaken the mind, clear away heat and disperse toxins, Chuanxiong can activate blood circulation and remove blood stasis, promote Qi and open depression, and play the function of supplementing Qi, detoxifying and removing blood stasis. Earlier research has shown that the extracts of Renshen, Chuanxiong, and Huanglian in SHDG can greatly improve spatial learning and memory retention, alleviate cognitive dysfunction, and activate autophagy to facilitate Aβ clearance.[9] These findings provide a useful scientific basis for the combined use of SHDG and donepezil. Currently, a multicenter pragmatic randomized controlled trial (RCT) is underway to validate the safety and effectiveness of SHDG in combination with donepezil for treating mild to moderate AD individuals.

Methods

Study Design

The trial is a multicenter, pragmatic RCT in which the superiority test is used. The protocol is meticulously formulated in line with the guidelines provided by the SPIRIT-TCM Extension 2018, as demonstrated in Supplementary Table 1.[10] A total of 386 individuals with mild-to-moderate AD will be recruited for this trial. They will be randomly divided into either the test group or the control group at a 1:1 ratio. The patients in the test group will be administered a combination of SHDG and donepezil, while the control group will solely receive donepezil therapy. Figure 1 shows the research flowchart. The Medical Ethics Committee of the Third Affiliated Hospital of Guangzhou University of Chinese Medicine

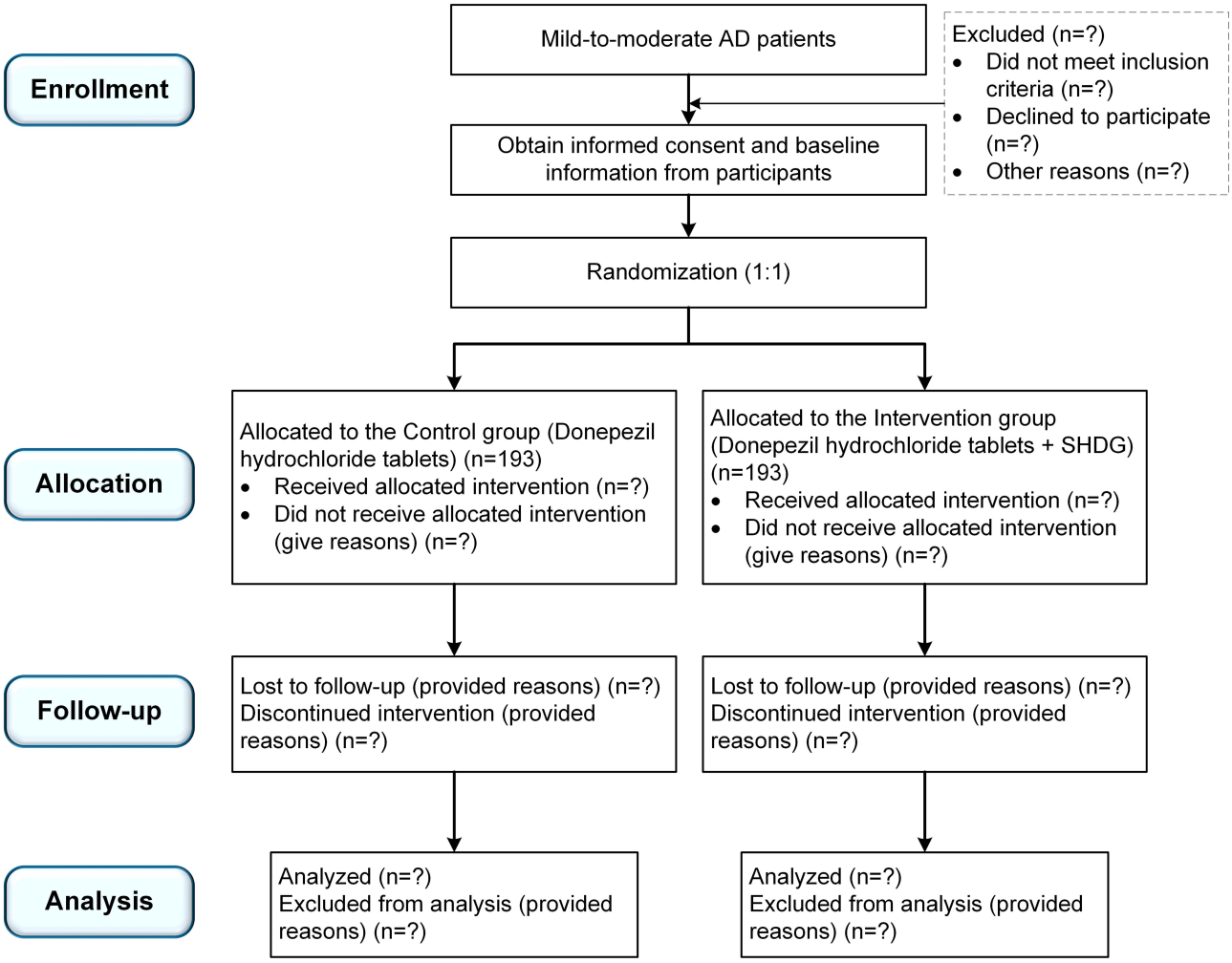

Figure 1 Flowchart illustrating the clinical trial of SHDG in combination with donepezil.

has approved the trial (Approval Number: PJ-KY-20230523-002), which was subsequently registered in the Chinese Clinical Trial Registry on 25 June 2023 (ChiCTR2300072768).

Participant Recruitment

Participants will be recruited at eight research centers in China. A list of the specific research centers is provided in Supplementary Table 2. The Shanghai Geriatric Institute of Chinese Medicine is a scientific research institution with the main task of carrying out research on TCM for geriatrics. The other seven research centers are all Class-A tertiary hospitals with rich experience in RCT. The medical ethics committee will review the recruitment subject advertisements, which will be posted in the hospital. The recruitment time will continue until December 31, 2025.

To guarantee the well-being of participants, as well as to encourage participant retention and successful follow-up, the recruitment of participants in this trial is carried out by clinicians who have undergone extensive training and obtained certificates in good clinical practice (GCP). These research physicians will thoroughly explain the trial's objectives, procedures, potential advantages, and risks to individuals who express interest in participating. Informed consent will be obtained from these patients or their authorized surrogates who have fully understood the trial. As shown in Supplementary Material 3, informed consent will require potential participants or authorized surrogates to grant permission to the research physicians for the reasonable collection of the participants' medical data and biospecimens. This collection will be done in accordance with the protocol for assessing the effectiveness and safety of the intervention

drugs. If they agree, potential participants will undergo further physical examination to determine whether they are eligible to participate in this study.

Inclusion Criteria

The following criteria must be met by participants:

(1) Individuals who satisfy the diagnostic criteria of the National Institute on Aging-Alzheimer's Association for probable AD dementia (2011);[11]

(2) Individuals who satisfy the diagnostic criteria for TCM patterns of "deficiency, blood stasis, and toxicity", as outlined in the Guiding Principles of Clinical Research on New Drugs of Traditional Chinese Medicine (2002);[12]

(3) Individuals diagnosed with mild to moderate AD have Mini-Mental State Examination (MMSE) scores ranging from 10 to 26 points, while their Clinical Dementia Rating (CDR) scores range between 1 and 2 points;

(4) Participants aged 55 to 85 years;

(5) Patients whose disease duration is not less than 6 months;

(6) Patients are capable of completing neuropsychological tests and cooperating with cranial MRI (resting for more than 20 minutes); if they are unable to cooperate with cranial MRI, at least complete cranial CT examination is performed;

(7) Patients who have reliable caregivers;

(8) Patients who voluntarily participate in the trial must provide informed consent, and in cases where a patient with dementia is unable to do so, a legal representative will provide consent on their behalf.

Exclusion Criteria

Individuals who meet the specified exclusion criteria will not be included as participants:

(1) Individuals experiencing dementia resulting from alternative factors such as vascular dementia, frontotemporal dementia, Lewy body dementia, brain tumors, hydrocephalus, or alcoholism;

(2) Patients with severe circulatory, respiratory, digestive, or other diseases;

(3) Participants with severe liver and kidney dysfunction;

(4) Participants who had started taking any medication that affects cognitive function before the baseline assessment;

(5) Patients with an allergic constitution or with a known allergy to the pharmaceutical ingredients used in this trial.

Interventions

All participants who are eligible for participation will be assigned randomly to either the test or control group and will be required to receive treatment with donepezil hydrochloride tablets. Donepezil hydrochloride tablets are manufactured by Zein Biotechnology Co., Ltd. The tablets should be taken at bedtime, once every night, with a dosage of 5 mg per intake. And the dosage will be adjusted to 10 mg per night after 4 weeks of maintenance. The total course of treatment with donepezil hydrochloride tablets is 24 weeks. Furthermore, the participants in the test group will need to undergo the SHDG (5.94 g/sachet) intervention twice a day with one sachet at a time for 24 weeks. Table 1 displays the detailed composition information of SHDG. Beijing Chunfeng Chinese Medicine Co., Ltd., will prepare SHDG by decocting, separating solid and liquid, drying under reduced pressure and other methods, and package it in small bags, each weighing 5.94g. In addition, they are also responsible for drug identification, quality control, and safety assessment.

Throughout the trial, the investigators will check and record the usage of the intervention drugs on a weekly basis. Patient adherence to medication is tracked by examining the quantity of medication left at follow-up visits. The actual dosage taken is determined by calculating the difference between the total number of doses prescribed and the number of doses still available. This approach facilitates an accurate evaluation of how well patients are following their prescribed treatment plan. Compliance = (actual dosage/should be dosage) × 100%. Deviation from the study protocol is considered significant if the adherence is less than 80% or exceeds 120%. Regular telephone follow-up and science popularization on social media may be effective strategies for improving participants' adherence to interventions.

In addition, it is recommended that participants avoid using any additional medications to treat AD to guarantee the evaluation of drug effectiveness and safety. When participants have other concurrent diseases requiring routine treatment,

Table 1 Dosages and Constituents of SHDG

Chinese Name	Latin Name	Origin	Pharmacological Effects	Weight (%)
Renshen	*Panax ginseng*	The dried root or rhizome of Panax ginseng C.A. Mey	Regulation of the central nervous system; bidirectional effects on learning and memory function	31.63
Chuanxiong	*Ligusticum wallichii*	The dried rhizome of Ligusticum chuanxiong Hort	Excitation of medullary respiratory, vasomotor, and spinal reflex centers	17.57
Huanglian	*Coptis chinensis*	The dried rhizome of Coptis chinensis Franch	Antioxidant; anti-inflammatory; Excitatory cerebral cortex	9.84
Zhizi	*Gardenia jasminoides Ellis*	The dried and mature fruit of Gardenia jasminoides Ellis	Sedation	14.59
Bingpian	*Borneolum Syntheticum*	$C_{10}H_{18}O$	Crossing the blood-brain barrier	26.36

Abbreviation: SHDG, Shenxiong Huanglian Detoxification Granule.

the investigators must record the details of concomitant treatment, including the name, dosage, administration frequency, and timing in the electronic case report forms (eCRFs), for summary analysis.

Criteria for Discontinuing Allocated Interventions

Participants can voluntarily withdraw from the trial at any time for any reason. In the case of any adverse event (AE) that poses a threat to life or significantly impacts daily routine, the trial for individuals with AEs will be discontinued. Additionally, if the participant experiences severe and persistent anaphylaxis, initiates unauthorized treatments, or significantly deviates from the prescribed regimen, this will also lead to the withdrawal of the affected participant.

Provisions for Post-Trial Care

If the participants have any damage related to the trial, the research center will pay the medical expenses incurred and provide appropriate financial compensation through laws and regulations.

Outcomes

Primary Outcome

The Alzheimer's Disease Assessment Scale-Cognitive Subscale (ADAS-cog) is a commonly utilized tool for assessing the effectiveness of clinical trials of AD drugs.[13] It evaluates various aspects including memory, language proficiency, operational skills, and attention. Education and language skills have little impact. The degree of cognitive impairment in AD patients can be effectively gauged by examining their performance on the tasks included in the scale. The ADAS-cog has a score ranging from 0 to 70. A higher score indicates more serious cognitive impairment. In this trial, the ADAS-cog assessment will be performed at the beginning, after 12 weeks, and after 24 weeks of treatment.

Secondary Outcomes

As the secondary outcomes, the TCM Syndrome score, MMSE score, CDR score, and activity of daily living (ADL) score will be assessed at the beginning, after 12 weeks, and after 24 weeks of treatment.

The TCM Syndrome score scale was created according to the Guiding Principles of Clinical Research on New Drugs of Traditional Chinese Medicine (2002), specifically targeting TCM patterns characterized by "deficiency, blood-stasis, and toxicity".[12] This scale provides a reference for evaluating the severity of clinical symptoms and objectively evaluating the efficacy of TCM syndromes. The research physicians will rate the patients in accordance with symptoms and signs such as memory dysfunction, lassitude, low back and knee soreness, dizziness and tinnitus, and dim complexion. The efficacy index will be subsequently calculated according to the ratio of the score difference before and after the intervention to the pretreatment score.

The MMSE is a cognitive function assessment tool that exhibits a considerable degree of sensitivity and specificity when diagnosing AD. It also helps determine the best threshold and educational adjustments. The primary components

assessed in the MMSE are orientation, immediate recall, delayed recall, cognitive functions, spatial abilities, problem-solving abilities, and language skills.[14] The highest possible score on the scale is 30 points. A lower score is associated with more severe cognitive impairment.

The CDR is a tool used to identify and assess the progression of AD dementia, making it appropriate for monitoring the cognitive condition of patients over an extended period. The assessment items of the CDR encompass six areas: recall, alignment, decision-making and critical thinking, social engagement, household and interest, and self-care.[15] These six aspects will be evaluated according to the standard for total score assessment, and the severity of dementia will be graded.

The ADL scale is an evaluation tool for the daily activities of AD patients with high stability and internal consistency. The assessment of ADL consists of two sections: one focuses on physical tasks performed in daily life, while the other assesses activities that require more complex skills.[16] A lower comprehensive score indicates more serious cognitive impairment.

Other Measurements

Aβ42, total tau, phosphorylated tau181, and neurofilament light chain are blood biomarkers with good diagnostic performance for AD and can accurately distinguish AD patients from participants with complete cognitive function or other neurodegenerative diseases.[17] The diagnostic performance of these biomarkers will be further confirmed by detecting their levels at the beginning, after 12 weeks, and after 24 weeks of treatment.

The harmful loop caused by the interplay of oxidative stress and inflammation can worsen nerve injury as AD progresses.[18] During the baseline, week 12, and week 24 of treatment, we will examine the presence of antioxidant substances (superoxide dismutase and glutathione peroxidase), the product of free radical reactions (malondialdehyde), and inflammation indicators (tumor necrosis factor-α, interleukin-1β, and interleukin-6). This analysis aims to determine whether the combined use of SHDG and donepezil can interfere with oxidative stress and inflammation during the progression of AD. Compared to individuals without cognitive impairments, patients with AD exhibit reduced cerebral blood flow, and this reduction in cerebral blood flow is linked to an increased risk of developing AD.[19] The detection of hemorheology indicators (whole blood viscosity, hematocrit, plasma viscosity, whole blood reduced viscosity at low shear, whole blood reduced viscosity at high shear, fibrinogen, erythrocyte aggregation index, and erythrocyte deformability index) at the beginning, week 12, and week 24 of treatment will reflect the alterations in hemorheology before and after intervention.

Safety Assessments

For any kind of intervention, safety evaluation and efficacy evaluation are equally important. Especially for TCM compound preparations with complex components, safety evaluation is the basis for improving the recognition of TCM. In this study, the participants' vital signs, hepatic and renal function, routine blood test results, routine urine test results, routine stool test results, electrocardiogram results, and other indicators will be dynamically monitored based on the intervention and follow-up duration. Physical examination is also one of the main aspects of safety evaluation. In the case of AEs, the nature, severity, consequences, or frequency of adverse reactions should be judged in time, and corresponding measures should be taken and recorded in detail.

Participant Timeline

In Figure 2, the enrollment, interventions, and assessments of the participants are shown.

Sample Size

The primary purpose of this trial is to determine whether the effectiveness of SHDG in combination with donepezil is superior to that of donepezil alone for treating patients with mild to moderate AD. A superiority test will be conducted. The formula for sample size calculation is as follows:[20]

	STUDY PERIOD					
	Enrolment	Allocation	Post-allocation			Follow-up
TIMEPOINT:	Day -7-0	Day 0	0-12 weeks	Week 12 ± 7days	Week 24 ± 7days	Week 48 ± 7days
ENROLMENT:						
Eligibility screen	X					
Informed consent	X					
Demographics collection	X					
Collection of medical records	X					
Allocation		X				
INTERVENTIONS:						
SHDG + Donepezil hydrochloride tablets			←	—	→	
Donepezil hydrochloride tablets			←	—	→	
ASSESSMENTS:						
Vital signs	X			X	X	X
Physical examination	X			X	X	X
Cranial MRI or CT	X					
Electrocardiogram	X			X	X	
Alzheimer's Disease Assessment Scale-Cognitive Subscale	X			X	X	
Traditional Chinese Medicine Syndrome score scale	X			X	X	
Mini-Mental State Examination	X			X	X	
Clinical Dementia Rating	X			X	X	
Activity of Daily Living scale	X			X	X	
Blood, urine, and stool routine	X			X	X	
Hepatic and renal function	X			X	X	
AD blood biomarkers	X			X	X	
Inflammation indicators	X			X	X	
Oxidative stress indicators	X			X	X	
Hemorheology indicators	X			X	X	
Adverse events				X	X	X
Record of concomitant medication	X			X	X	X

Figure 2 SPRIT figure of participants' enrollment, interventions, and assessments.

$$n_C = \frac{\left(Z_{1-\alpha} + Z_{1-\beta}\right)^2 \sigma^2 \left(1 + \frac{1}{K}\right)}{\left(\mu_T - \mu_C - \delta_U\right)^2}$$

In the given equation, n_C represents the estimated number of patients in the control group, $\mu_T - \mu_C$ denotes the disparity between the average values of the test group and the control group, σ signifies the combined standard deviation, and δ_U represents the boundary value of superiority. Furthermore, α represents the likelihood of committing a Type I error, while β represents the likelihood of committing a Type II error. K represents the ratio between the test group and the control group. The primary outcome in this trial is determined by the average change in the ADAS-cog score. Typically, in a clinical setting, a 4-point alteration in the ADAS-cog is employed to assess the efficacy of the medication, denoted as $\delta_U = 4$.[21] In conjunction with data from prior research, $\mu_T - \mu_C = 2$ and $\sigma = 6.59$.[22] When the allocation ratio

of this trial was 1:1 (K = 1), the significance level was 0.025 (one-sided test), and the power was 80% (β = 0.2), the estimated minimum total sample size was 340. Considering the 10% drop-out rate and the actual clinical situation, a sample size of 386 is required to ensure the reliability of the trial.

Randomization, Allocation Concealment, and Blinding

The Department of Statistics at Guangzhou University of Traditional Chinese Medicine completed the random allocation scheme. By using the central stratified block randomization method, participants were equally divided into test and control groups. The block length and the number of centers were determined by SAS software, and a random number table was generated. The block length is temporarily confidential and will be revealed after the study is completed. Opaque envelopes will contain random numbers. After the envelope subpackaging is completed by the personnel who do not participate in the clinical trial, the participants will randomly select the envelope.

According to the random numbers in the envelopes extracted by the participants, the pharmacists will confirm the group of participants and give the participants the drugs of the corresponding group. Only statisticians who created the random number table knew the complete random allocation scheme. The study implementers are not informed of the random allocation scheme, so they have no impact on the group to which the participants are assigned.

However, since this study is a pragmatic RCT without a placebo, the study implementers and participants can know the participants' group after the participants obtain the drug. To minimize the potential impact of personal bias on the results, the study implementers will not participate in the outcome assessments. Moreover, the study outcome assessors and statistical data analysts are blinded, and they are unaware of the participants' allocation of groups and intervention measures. The study implementers and participants are not allowed to provide information related to the intervention to blinded assessors.

Data Collection and Management

Collection of Data

All the data will be gathered by the investigators at each research center through case report forms (CRFs). To guarantee the quality of the gathered information, the personnel responsible for the data collection, who possess GCP certification, will undergo comprehensive training regarding the protocol. This training ensures the precision, completeness, and punctuality of the CRF data. To address the insufficiency of the data and enhance the integrity of the data, investigators will enhance the follow-up process, proactively resolve participant issues in this trial, and promptly notify participants of visit schedules to enhance participant retention.

Data Management

The data managers established the database and constructed the eCRFs according to the requirements of the protocol. After receiving the CRF, the data entry clerk who has received relevant training on the database will conduct a pre-entry inspection of the CRF. Once the CRF data are input into the database, the data managers verify the accuracy, coherence, presence of missing values, and adherence to the normal value range of the data. If any errors are found in the data, the investigators will be notified in a timely manner for correction. After completing the data quality assurance program, the database will be locked to prevent unauthorized access. Only the principal investigator (PI) has permission to read, modify, and export the data. After the database is locked, any data modification records will be retained. In addition, all the documents related to quality control, such as the CRF, the original informed consent signed by the participants, and the detailed records of drug distribution will be properly kept.

Confidentiality

To maintain the privacy of participants' medical information, only the investigators, the department of drug regulation, and the ethics committee have permission to view participants' medical records. The personal information of any of the participants in any public report on the results of this trial will not be disclosed.

Collection of Biological Specimens

The blood, urine, and fecal samples of participants will be gathered and examined following the study protocol. These include analyzing specific molecules in the blood and measuring their levels to monitor disease progression and evaluate

treatment efficacy. The identification, transfer, preservation, and other relevant records of all biological samples and sample storage files will be established. After the data are reported, all samples will be destroyed.

Statistical Analysis

The study data will undergo statistical analysis using SAS software. Moreover, two-sided tests will be used and $p<0.05$ is considered significant. For analysis, we will choose the full analysis set (FAS), the per-protocol set (PPS), and the safety set (SS). The FAS aims to adhere as closely as possible to the intention-to-treat (ITT) principle by including all randomized subjects to minimize bias and ensure fairness. The exclusion criteria include violations of important enrollment criteria, never receiving treatment with study drugs, and no observed data after randomization. The PPS is a subset of the FAS that includes patients who have completed all drug treatment and evaluation contents and no obvious protocol deviation. The SS is an analysis set consisting of all randomized subjects who have taken study drugs at least once and have safety index records.

In this trial, the FAS will be used to analyze baseline characteristics, including demographic data. The FAS and PPS will be simultaneously used for efficacy evaluation. Increasing the credibility of the results can be achieved if the conclusions from FAS and PPS analysis align. The SS will be used for the safety evaluation of the study. When the primary efficacy indicators are missing, the last observation is carried forward according to the ITT. The missing values of secondary efficacy indicators and safety indicators will not be carried forward and will be analyzed based on the actual data. The processing of outliers is performed by the PI and the statistical data analysts in the data blind review stage.

Descriptive analysis will be conducted on the baseline data, with qualitative indicators presented as percentages, constituent ratios, or frequency tables. The mean ± standard deviation, interquartile range, minimum, and maximum values will be utilized to express the quantitative data. Dichotomous data will be analyzed using logistic regression. A mixed effects model will be utilized to analyze both the primary and secondary outcome measures. In the safety analysis, the adverse reactions and AEs that occur during the study will be summarized. AEs will be characterized based on the frequency and incidence of AEs, and comparisons of the incidence of AEs between groups will be conducted using either the chi-square test or Fisher's exact test.

Oversight and Monitoring

Quality and Safety Assurance

The responsibility for monitoring the quality and safety of this study will lie with the Medical Ethics Committee of the Third Affiliated Hospital of Guangzhou University of Chinese Medicine, which is independent of the investigators and the sponsor. In their role as the committee responsible for monitoring the quality and safety of this study, they will assess the study protocol to guarantee the well-being of the participants and the effectiveness of any intervention methods employed in the trial. If necessary, they will propose suggestions for protocol amendments. The PI will amend the protocol according to the opinions and communicate protocol amendments to the ethics committees, the competent department, and the investigators. If the amendments affect participants, they will be informed. Informed consent forms must be signed by all participants before enrollment.

Moreover, the research quality and safety monitoring committee will send coordinators to review each center through the electronic system every 6 months and ensure at least one on-site visit every year. At each visit, the study protocol, ethics approval, informed consent, CRFs, and other data will be checked, and the study progress will also be evaluated.

Data Monitoring

A committee responsible for the data monitoring will be established by the Medical Ethics Committee of the Third Affiliated Hospital of Guangzhou University of Chinese Medicine. The data monitoring committee is a third party independent of the investigators and sponsors and has no conflicts of interest with the research team. To guarantee the integrity of the research data, the data monitoring committee will perform a biannual review of the data.

Adverse Event Reporting and Harms

AEs encompass any negative medical occurrences that occur following the administration of medications to a participant. These events may present as symptoms, disease, or abnormal laboratory test results, but they are not necessarily directly caused by the treatment. In this trial, we will closely monitor and prevent AEs. During week 12, week 24 of treatment, and the follow-up period, participants must honestly report their medication situation and undergo necessary safety assessments. In the case of AE, the study implementers should preliminarily assess the grade of the AE and its correlation with the study medications according to the condition, handle the adverse effects symptomatically, and closely observe the outcome of the AE until the symptoms and signs disappear and laboratory indicators return to normal. Moreover, the study implementers should also record AEs in the CRF in detail, including symptoms, signs, laboratory results, start time, end time, severity, correlation, outcome, concomitant medication, and treatment measures. In the event of a serious AE, investigators must promptly take appropriate actions to ensure the participants' safety. They must also notify the PI within 24 hours of the incident, who will then report it to the research quality and safety monitoring committee for documentation.

Dissemination Plan

The findings of this trial will be shared through publications in globally recognized scholarly journals.

Discussion

The meta-analysis revealed that TCM when used alongside conventional Western medicine, demonstrated notable benefits in enhancing cognitive function and daily activities among AD patients. Moreover, no safety concerns were reported in the monitoring data.[23] The use of TCM as an adjuvant treatment to conventional anti-dementia Western medicine is a viable and safe treatment option for AD.

The SHDG was created by our team based on 20 years of clinical experience in the treatment of AD, aiming at the key pathogenesis of "deficiency, blood stasis, and toxicity". Our previous research confirmed that the components derived from Renshen, Chuanxiong, and Huanglian in SHDG can reduce Aβ accumulation and enhance autophagy by modulating the mTOR pathway, thereby providing neuroprotective effects in elderly rats with cognitive impairment.[9] When used in isolation, donepezil, a cholinesterase inhibitor, has a favorable therapeutic impact on mild to moderate AD, but its efficacy is limited. It is advocated to combine this treatment with other drugs.[24] Currently, there is no direct clinical proof regarding the effectiveness and safety of SHDG in combination with donepezil for treating mild to moderate AD. To further assess the effectiveness and safety of this combined treatment, we designed a multicenter, pragmatic RCT.

This study will be carried out at eight research centers in six regions of China, which can expand the applicability of the results to a wider population. Pragmatic RCTs are often deemed more relevant to clinical practice than traditional explanatory RCTs due to their focus on conditions that mirror real-world scenarios. By incorporating actual settings and diverse patient populations, these pragmatic RCTs provide data that is more applicable and relevant for making medical decisions. Furthermore, the trial design incorporates multiple time points to assess the potential influence of the duration of AD treatment on the therapeutic effect.

This study inevitably has several limitations. As a pragmatic RCT, it is not blind to the study implementers or participants, which will affect its internal authenticity. However, through randomization and allocation concealment, the selection bias of the study can be minimized. By blinding outcome assessors and statisticians, we can also try to overcome the reporting bias caused by knowing the randomization.

Trial Status

The subject recruitment of this study started in October 2023, and the first subject was enrolled on October 26, 2023. Currently, the recruitment of participants is still ongoing, and the study will continue through December 31, 2025. The current study protocol version is 2.0, September 14, 2023.

Permission Statement for MMSE

The trial has obtained the necessary license for the use of MMSE® from Par Inc. An unauthorized version of the Chinese MMSE was used by the study team without permission; however, this has now been rectified with PAR. The MMSE is a copyrighted instrument and may not be used or reproduced in whole or in part, in any form or language, or by any means without written permission of PAR (www.parinc.com).

Abbreviations

AD, Alzheimer's disease; Aβ, amyloid β; TCM, traditional Chinese medicine; SHDG, Shenxiong Huanglian Detoxification Granule; RCT, randomized controlled trial; GCP, good clinical practice; MMSE, Mini-Mental State Examination; CDR, Clinical Dementia Rating; eCRFs, electronic case report forms; AE, adverse event; ADAS-cog, Alzheimer's Disease Assessment Scale-Cognitive Subscale; ADL, activity of daily living; CRFs, case report forms; PI, principal investigator; FAS, full analysis set; PPS, per-protocol set; SS, safety set; ITT, intention-to-treat.

Ethics Approval and Consent to Participate

The Medical Ethics Committee of the Third Affiliated Hospital of Guangzhou University of Chinese Medicine has granted approval for this trial (approval number: PJ-KY-20230523-002). The trial will comply with the Declaration of Helsinki. All participants will be required to provide signed informed consent before participating.

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Disclosure

The authors declare that they have no competing interests.

References

1. Penke B, Szucs M, Bogár F. New pathways identify novel drug targets for the prevention and treatment of Alzheimer's disease. *Int J Mol Sci*. 2023;24(6):23. doi:10.3390/ijms24065383
2. World Health Organization. Fact sheets of dementia. 2023. Available from: https://www.who.int/news-room/fact-sheets/detail/dementia. Accessed July 1, 2024.
3. Tiwari S, Atluri V, Kaushik A, Yndart A, Nair M. Alzheimer's disease: pathogenesis, diagnostics, and therapeutics. *Int J Nanomed*. 2019;14:5541–5554. doi:10.2147/IJN.S200490
4. Liu YS, Zhao HF, Li Q, Cui HW, Huang GD. Research progress on the etiology and pathogenesis of Alzheimer's disease from the perspective of chronic stress. *Aging and Disease*. 2023;14(4):1292–1310. doi:10.14336/AD.2022.1211
5. Zhang BY, Zhao J, Wang Z, Guo PF, Liu AL, Du GH. Identification of multi-target anti-AD chemical constituents from traditional Chinese medicine formulae by integrating virtual screening and *in vitro* validation. *Front Pharmacol*. 2021;12:19. doi:10.3389/fphar.2021.709607
6. Shi J, Ni JNA, Lu T, et al. Adding Chinese herbal medicine to conventional therapy brings cognitive benefits to patients with Alzheimer's disease: a retrospective analysis. *BMC Complement Altern Med*. 2017;17:7. doi:10.1186/s12906-017-2040-5
7. Lin ZH, Gu J, Xiu J, Mi TY, Dong J, Tiwari JK. Traditional Chinese medicine for senile dementia. *Evid-Based Complement Altern Med*. 2012;2012:1–13. doi:10.1155/2012/692621
8. Chinese PC. *Chinese Pharmacopoeia*. Vol. 1. Beijing, China: China Medical Science Press; 2020.
9. Yang Y, Wang ZY, Cao Y, et al. Yizhiqingxin formula alleviates cognitive deficits and enhances autophagy *via* mTOR signaling pathway modulation in early onset Alzheimer's disease mice. *Front Pharmacol*. 2019;10:1041. doi:10.3389/fphar.2019.01041
10. Dai L, Cheng CW, Tian R, et al. Standard protocol items for clinical trials with traditional Chinese medicine 2018: recommendations, explanation and elaboration (SPIRIT-TCM Extension 2018). *Chin J Integr Med*. 2019;25(1):71–79. doi:10.1007/s11655-018-2999-x
11. McKhann GM, Knopman DS, Chertkow H, et al. The diagnosis of dementia due to Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. *Alzheimer's Dement*. 2011;7(3):263–269. doi:10.1016/j.jalz.2011.03.005
12. Zheng XY. *Guiding Principle of Clinical Research on New Drugs of Traditional Chinese Medicine(Trial)*. Beijing, China: China Medical Science Press; 2002.

13. Rosen WG, Mohs RC, Davis KL. A new rating-scale for Alzheimer's-disease. *Am J Psychiat*. 1984;141(11):1356–1364. doi:10.1176/ajp.141.11.1356
14. Folstein MF, Folstein SE, McHugh PR. Mini-mental state - practical method for grading cognitive state of patients for clinician. *J Psychiatr Res*. 1975;12(3):189–198. doi:10.1016/0022-3956(75)90026-6
15. Morris JC. The clinical dementia rating (CDR) - current version and scoring rules. *Neurology*. 1993;43(11):2412–2414. doi:10.1212/WNL.43.11.2412-A
16. Lawton MP, Brody EM. Assessment of older people: self-maintaining and instrumental activities of daily living. *The Gerontologist*. 1969;9(3):179–186. doi:10.1093/geront/9.3_Part_1.179
17. Cai HM, Pang YN, Fu XF, Ren ZY, Jia LF. Plasma biomarkers predict Alzheimer's disease before clinical onset in Chinese cohorts. *Nature Communications*. 2023;14(1):6747. doi:10.1038/s41467-023-42596-6
18. Shi JZ, Zheng XM, Zhou YF, et al. Cornuside is a potential agent against Alzheimer's disease via orchestration of reactive astrocytes. *Nutrients*. 2022;14(15):3179. doi:10.3390/nu14153179
19. Weijs RWJ, Oudegeest-Sander MH, Vloet JIA, Hopman MTE, Claassen JAHR, Thijssen DHJ. A decade of aging in healthy older adults: longitudinal findings on cerebrovascular and cognitive health. *Geroscience*. 2023;45(4):2629–2641. doi:10.1007/s11357-023-00790-w
20. Chow SC, Shao J, Wang H. *Sample Size Calculations in Clinical Research*. Boca Raton: Taylor & Francis; 2003.
21. Molnar FJ, Man-Son-Hing M, Fergusson D. Systematic review of measures of clinical significance employed in randomized controlled trials of drugs for dementia. *J Am Geriatr Soc*. 2009;57(3):536–546. doi:10.1111/j.1532-5415.2008.02122.x
22. Yang Y, Liu JP, Fang JY, et al. Effect and safety of Huannao Yicong formula in patients with mild-to-moderate Alzheimer's disease: a randomized, double-blinded, donepezil-controlled trial. *Chin J Integr Med*. 2019;25(8):574–581. doi:10.1007/s11655-018-3054-7
23. Kim MG, Ooi SL, Kim GW, Pak SC, Koo BS. Effectiveness and safety of pattern identification-based herbal medicine for Alzheimer's disease: a systematic review and meta-analysis. *Journal of Integrative and Complementary Medicine*. 2023;29(10):605–620. doi:10.1089/jicm.2022.0806
24. Moss DE. Improving anti-neurodegenerative benefits of Acetylcholinesterase inhibitors in Alzheimer's disease: are irreversible inhibitors the future? *Int J Mol Sci*. 2020;21(10):3438. doi:10.3390/ijms21103438