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Original Research

A 6-week, phase IIb, randomized, double-blind, placebo-controlled trial of Anyu Peibo capsules for the treatment of major depressive disorder in adults

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Abstract

Background: Almost one-third of patients with major depressive disorder (MDD) do not respond to conventional antidepressants, and new treatments for MDD are urgently needed.

Objectives: This phase IIb clinical trial was designed to evaluate the efficacy and safety of Anyu Peibo capsules in the treatment of adults with MDD.

Design: A multicenter, randomized, double-blind, placebo-controlled, parallel-group, fixed-dose study.

Methods: A total of 172 patients with MDD from nine study centers were randomized (1:1) to receive placebo ($n = 86$) or oral Anyu Peibo capsules (0.8 g) twice per day ($n = 86$) for 6 weeks. The primary endpoint was the change in the Montgomery Åsberg Depression Rating Scale (MADRS) total score from baseline to week 6, analyzed using an analysis of covariance (ANCOVA) approach with the baseline MADRS score, center effect and center by group interaction as the covariates. Other efficacy endpoints and variables included clinical response and remission rates according to the MADRS and the 17-item Hamilton Depression Rating Scale (HAMD-17) scores, the change in the HAMD-17, Clinical Global Impression – Severity scale and Clinical Global Impression – Improvement scale scores and the reduction in the Hamilton Anxiety Scale from baseline to week 6.

Results: The mean baseline MADRS total scores were 29.20 and 29.72 in the Anyu Peibo ($n = 82$) and placebo groups ($n = 81$), respectively. The least squares mean change in the MADRS score from baseline to week 6 was 16.59 points in the Anyu Peibo group and 14.51 points in the placebo group. Although there were greater reductions in the MADRS score from baseline to week 6 in the Anyu Peibo capsule group compared to the placebo group, the difference did not reach statistical significance (least-squares mean difference, 2.07 points; 95% confidence interval, −0.27 to 4.41; $p = 0.0819$). The results of sensitivity analyses by ANCOVA with the last observation carried forward method for missing data indicated that the administration of Anyu Peibo capsules may lead to a significant reduction in depressive symptoms compared to the placebo (least-squares mean difference: 3.29 points; 95% confidence interval: 0.64–5.93; $p = 0.0152$). Furthermore, Anyu Peibo capsules showed significant benefits over placebo when the change in the HAMD-17 score from baseline to week 6 was evaluated as the secondary analysis ($t = 2.01$; 95% confidence interval, 0.03–4.23; $p = 0.0464$).

Conclusion: Anyu Peibo capsules may have an effective and safe antidepressant effect, which warrants further research.

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Introduction

Major depressive disorder (MDD) is one of the most common mental health disorders and can lead to substantial disability and high economic costs.[1,2] A WHO report predicted that MDD will become the leading cause of disability in the world by 2030.[3] Currently, there are several classes of antidepressant drugs, including selective serotonin reuptake inhibitors (SSRIs), selective serotonin and norepinephrine reuptake inhibitors (SNRIs), monoamine oxidase inhibitors, tricyclic antidepressants and mirtazapine. However, despite the availability of antidepressants in multiple classes, almost one-third of MDD patients do not respond to conventional antidepressant medications.[4,5] Thus, there is an urgent need to develop novel treatments for MDD.

The raw material in Anyu Peibo capsules is Piper laetispicum C. DC., a herb traditionally used in China for invigorating blood circulation and reducing detumescence, as well as for its analgesic properties.[6–8] A previous study showed that Piper laetispicum had antidepressant-like effects in an animal model of depression.[9–11] For example, Piper laetispicum could improve depression-like behavior by regulating inflammatory cytokines and apoptosis cytokines in chronic unpredictable mild stress (CUMS) model animals.[12,13]

The active ingredients in Anyu Peibo capsules are mainly amide alkaloids. The alkaloids include *N*-isobutyl deca-trans-2-trans-4-diene amide, 1-[(2*E*,4*E*)-2,4-decadiene acyl] pyrrolidine, 3′,4′-methylenedioxy cinnamic acid isobutyl amide, 5′-methoxyl-3′,4′-methylenedioxy cinnamic acid-isobutyl amide, piperamide C5:1(2*E*), 5′-methoxyl-3′,4′-methylenedioxy cinnamic acid-pyrrolidine, 4,5-dihydro piperlonguminine, and piperamide. Some alkaloids have also been shown to have antidepressant effects, such as piperamide and 4,5-dihydro piperlonguminine.[14,15] The detailed extraction and preparation process have also been reported and patented in the European Patent Office [International publication number: WO 2006/058487 (08.06.2006 Gazette 2006/23)].

Moreover, preclinical pharmacological data showed that Anyu Peibo capsules could inhibit the reuptake of 5HT and NE *in vitro* and *vivo* (unpublished data). Importantly, Anyu Peibo capsules were well tolerated in healthy controls during phase I clinical trials (the data presented are unpublished and were sourced from CTR20131456 at www.chinadrugtrials.org.cn). The purpose of phase I studies is to assess the preliminary tolerability and safety of investigational drugs in human subjects, aiming to establish a safe dosage range for administering Anyu Peibo capsules in phase II clinical trials for MDD treatment. Furthermore, Anyu Peibo capsules lead to a significant improvement in depressive symptoms compared to the placebo and exhibited good safety in patients with MDD during the phase IIa study (ClinicalTrials.gov Identifier: NCT02380066) (unpublished data). Herein, we performed a phase IIb trial to further evaluate the efficacy and safety of Anyu Peibo capsules in patients with MDD.

Material and methods

Ethical approval

The study was conducted at nine research sites in China and approved by ethics committees at each site. The leading institution for this study was Shanghai Mental Health Center. The research protocol was approved by the Ethics Committee of Shanghai Mental Health Center (Approval number: 2017-12). Written informed consent was obtained from all participants prior to enrollment in the study. The study was conducted following the principles of Good Clinical Practice and in compliance with the Declaration of Helsinki guidelines. The study was registered in the Clinical Trials registry (NCT03183505).

Study design

This was a Phase IIb multicenter, randomized, double-blind, placebo-controlled, parallel-group, fixed-dose study of Anyu Peibo capsules, which was conducted in nine centers in China, including Shanghai Mental Health Center, Beijing

HuiLongGuan Hospital, Beijing Anding Hospital, Xi'an Mental Health Center, Wuhan Mental Health Center, Sichuan West China Hospital, Brain Hospital of Jilin Province, The Second Xiangya Hospital of Central South University and Jiangxi Mental Hospital.

The results of the phase I clinical trial showed that the maximum tolerable dose of Anyu Peibo capsules was 1.6 g/day for both single and repeated administration in healthy Chinese subjects. Moreover, the results of the phase IIa clinical trial showed that Anyu Peibo capsules (0.8 g twice daily) showed a significant benefit over placebo. Thus, Anyu Peibo capsules (0.8 g twice daily) were used for this study.

The study comprised a 1-week screening and washout period (a minimum washout period of 3 days) if the patient was previously taking antidepressants within the 2 weeks before screening, followed by a 6-week double-blind treatment period and a 1-week double-blind down-taper period. Following completion of baseline assessments, patients were randomized in a 1:1 ratio to the Anyu Peibo capsule or placebo groups using the random number generator in the PROC PLAN procedure of Statistical Analysis Software (SAS) version 9.4 software by the method of block randomization (block size of 4), stratified by the Clinical Global Impression – Severity (CGI-S) scale. The random number generator used in this study was provided by the Department of Health Statistics of Shanghai Second Military Medical University, which was not involved in subject recruitment or data collection. The sequentially numbered, opaque, sealed envelope technique was used for allocation concealment. The size, shape, color, taste and packaging of the Anyu Peibo capsules (0.2 g per capsule) and placebo (0.2 g per capsule) were identical to ensure double-blind conditions. Suzhou YiHua Biomedical Technology Co., Ltd (Suzhou, China). manufactured and supplied the Anyu Peibo capsules and the placebo for use in the clinical trial.

Since this was a pilot study, a sample size calculation was not performed. We referred to the measures for the administration of drug registration in China to determine the appropriate sample size for a phase II trial. These measures state that each group in a phase II trial should have more than 100 subjects. The methodological basis for both phase IIa and phase IIb trials is essentially the same. The inclusion criteria for patients, as well as the primary and secondary outcomes, were identical. The primary outcome measure was 'The change in the total score on the Montgomery Åsberg Depression Rating Scale (MADRS) score from baseline to week 6 [Time Frame: 6 weeks]'. The treatment duration and statistical analysis methods were also consistent. Based on the results of the phase IIa trial and considering the guidelines outlined in the 'Measures for the Administration of Drug Registration' in China, we determined the sample size for the phase IIb trial. Given the previous phase IIa trial that involved 24 patients in both the 1.6 g Anyu Peibo capsule and placebo groups, we planned to include 86 patients in the Anyu Peibo capsule and placebo groups at a 1:1 ratio for this phase IIb trial.

Patient selection

Adult outpatients or inpatients between the ages of 18 and 65 years (inclusive) were eligible if they were diagnosed with MDD according to the Diagnostic and Statistical Manual of Mental Disorders, 5th Edition.[16] The inclusion criteria were as follows: (1) adults with a primary diagnosis of MDD based on the criteria of the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5), with single or recurrent episodes; (2) patients with a MADRS total score ⩾24 points at both the screening and baseline visits; (3) patients with a 17-item Hamilton Depression Rating Scale (HAMD-17) total score ⩾18 points and a score ⩾2 points on the first item (depressed mood) at both the screening and baseline visits; (4) patients with a CGI-S scale total score ⩾4 points at both the screening and baseline visits; and (5) patients who understood and consented to take part in this clinical trial. The subjects signed an informed consent form.

The exclusion criteria mainly included the following: (1) subjects who had a suicide attempt within the last year, a currently significant risk of suicide, or a score ⩾3 points on Item 3 (suicide assessment) of the HAMD-17; (2) subjects with a current psychiatric diagnosis other than depression; (3) subjects with unstable cardiovascular, hepatic, renal, blood, endocrine, or other medical diseases; (4) subjects with any neurological disease (such as Parkinson's disease, cerebrovascular accident and epilepsy) or cerebral injury (traumatic or disease-related injury); (5) subjects with clinically significant abnormal laboratory values; (6) subjects who used at least two different antidepressants with the recommended dose and an adequate duration of treatment (maximum dosage for at least 4 weeks

according to the label) and still had no response; (7) subjects who had received regular antidepressant treatment in the 2 weeks prior to screening; (8) subjects who received electroconvulsive therapy (ECT), transcranial magnetic stimulation, or other physical therapy within 3 months; (9) subjects who had a history of substance abuse (including alcohol, drug or other psychoactive substance abuse) within 1 year before screening; and (10) women who were pregnant, breastfeeding, or serum human chorionic gonadotropin (HCG) (+) on screening or were planning to become pregnant. A more detailed list of the inclusion/exclusion criteria can be found at http://clinicaltrials.gov (NCT03183505).

Outcome measures

The HAMD-17 scale, widely utilized as a clinician-administered assessment for depression, consists of 17 items.[17] It primarily focuses on evaluating cognitive and vegetative symptoms associated with depression and incorporates fewer items pertaining to social, motor, anxiety, and mood factors. Higher scores on the HAMD-17 scale indicate a greater severity of depression. Similarly, the MADRS is a widely used scale specifically designed to assess the severity of depressive symptoms.[18] It consists of 10 items that evaluate symptoms not covered in the HAMD-17, such as the inability to experience emotions and difficulties with concentration. With scores ranging from 0 to 60 points, higher MADRS scores indicate a greater severity of depression. The MADRS has been found to have superior internal reliability and sensitivity to change when compared to the HAMD-17.[19]

For measuring the severity of anxiety, the Hamilton Anxiety Scale (HAMA) scale is commonly used.[20,21] It consists of 14 items that evaluate different aspects of anxiety. Scores on the HAMA scale range from 0 to 56 points, with higher scores indicating greater levels of anxiety. In addition to these specific rating scales, the CGI-S and Clinical Global Impression – Improvement (CGI-I) scales are widely used assessment tools in clinical drug trials for various mental disorders.[22] The CGI-S scale evaluates the overall severity of an individual's condition, while the CGI-I scale assesses improvements or changes over time. The CGI-S scale is rated on a scale from 1 (healthy, not at all ill) to 7 (among the most extremely ill patients), while the CGI-I scale is rated from 1 (very much improved) to 7 (very much worse). These scales have gained popularity due to their conciseness and ease of administration, and the Chinese version is widely used in clinical trials for MDD.[23–26]

Efficacy assessments were conducted at weeks 1, 2, 4, and 6 by independent interviewers who were blinded to treatment allocation. The primary efficacy outcome was the change in the total score on the MADRS from baseline to the end of 6 weeks. The secondary efficacy outcomes included the following: (1) the clinical response rate according to the MADRS score; (2) the clinical remission rate according to the MADRS score; (3) the clinical response rate according to the HAMD-17 score; (4) the clinical remission rate according to the HAMD-17 score; (5) the change in the HAMD-17 score from baseline to week 6; (6) the changes in the CGI-S and CGI-I scale scores from baseline to week 6; and (7) the change in the HAMA total score and the reduction in the HAMA total score from baseline to week 6. A clinical response was defined as a reduction $\geqslant$50% in the MADRS or HAMD-17 total scores (improvement) from baseline to week 6. Remission was defined as a MADRS score $\leqslant$ 10 points or a HAMD-17 score $\leqslant$ 7 points. Safety measures included adverse events (AEs), laboratory parameters, vital signs and electrocardiogram (ECG) parameters.

Statistical methods

Three analysis populations were defined: the full analysis set (FAS; all randomized subjects who received at least one dose of the study medication and had at least one postbaseline efficacy assessment) for the primary analysis, the per-protocol set (PPS; subjects in good compliance with the study protocol), and the safety set (SS; all randomized subjects who took at least one dose of the study medication). The FAS and PPS were applicable for efficacy analysis. The change in the MADRS total score from baseline to week 6 in the FAS was the primary efficacy outcome in this study, which was evaluated by the analyzed using an analysis of covariance (ANCOVA) model, including treatment, center, baseline MADRS score (covariate), and the interaction between treatment and center as the covariates. Moreover, to further assess the robustness of the primary analysis results, the mixed-effects model for repeated measures (MMRM) analysis and ANCOVA with the last observation carried forward (LOCF) method or missing values were

both used as sensitivity analyses. MMRM analysis was carried out with treatment group and visit as fixed factors. Center effect and an unstructured variance covariance matrix were used for the random effect, and the baseline MADRS total score was included as a covariate. In the secondary analysis, the response and remission rates were analyzed using a Cochran–Mantel–Haenszel test stratified by pooled center. Moreover, the mean changes in the HAMD-17 and HAMA scores and the reduction in the HAMA score from baseline to week six and other secondary efficacy indices were compared between the two groups by t tests or rank-sum tests. Safety analyses were conducted in the safety population, defined as all patients who took at least one dose of the study medication. The incidence and severity of Aes and withdrawal due to Aes were recorded throughout the study. The chi-squared test or Fisher's exact test was used to compare the difference in the frequency of Aes between the experimental and control groups. A rank-sum test was conducted to analyze the independent ordered multiple category data when cases were divided into different degrees of Aes. Secondary outcomes were exploratory, and thus, the statistical analysis plan did not include a correction for multiplicity for secondary outcomes. Therefore, the secondary results should not be used to infer treatment effects. Analyses were performed using SAS, version 9.4 (SAS Institute, Inc. Cary, NC, USA). A two-sided p value <0.05 was considered significant.

Results

Patient population

This study was conducted at nine centers in China. Of the 233 patients screened, 172 eligible patients with MDD were randomized to receive Anyu Peibo capsules or placebo (see Figure 1). Because of obvious quality problems at the centers (the original records were incomplete), eight subjects (four in the Anyu Peibo capsule group and four in the placebo group) were excluded from this study. Moreover, one patient did not receive the experimental drug and therefore was not included in the FAS and safety analysis. Thus, 163 patients received treatment and were included in the safety analysis set. A total of 19 patients prematurely discontinued the study. Because the study drug compliance of one of the 19 patients in the placebo group was more than 75%, this patient was still included in the PPS dataset. Finally, the FAS and PPS populations comprised 163 patients and 145 patients, respectively. There were no significant differences between the Anyu Peibo capsule and placebo groups with respect to baseline demographics or disease characteristics (Table 1).

Extent of exposure and treatment compliance

In the FAS, the mean drug exposure per patient was 62.2 g and 58.2 g in the Anyu Peibo capsule and placebo groups, respectively. There was no difference in drug exposure between the two groups ($p=0.1166$). Moreover, no difference in compliance rates was found between the Anyu Peibo capsule group (91.5%; 75/82) and the placebo group (85.2%; 69/81) ($p=0.2323$). Compliance rates ranged from 80 to 120% and were defined as [the total actual number of capsules taken $\times$ 100]/the total planned number of capsules taken).

Efficacy

The mean baseline MADRS total scores for the Anyu Peibo group ($n=82$) and placebo group ($n=81$) were 29.20 and 29.72 points, respectively. For the primary efficacy endpoint, the least squares mean (standard error) change in the MADRS score from baseline to week 6 in the FAS was 16.59 ± 0.89 points in the Anyu Peibo capsule group and 14.51 ± 0.96 points in the placebo group (least-squares mean difference, 2.07 points; 95% confidence interval, −0.27 to 4.41; $p=0.0819$) (Figure 2 and Table 2).

It is likely that Anyu Peibo capsules were superior to placebo based on the results of sensitivity analyses by the MMRM in the FAS (least-squares mean difference, 2.4 points; 95% confidence interval, −0.08 to 4.88; $p=0.0574$). Moreover, sensitivity analyses for missing data using the LOCF method showed that the least-squares mean (standard error) change in the MADRS score from baseline to week 6 was -16.04 ± 1.03 points in the Anyu Peibo capsule group and -12.75 ± 1.06 points in the placebo group (least-squares mean difference, 3.29; 95% confidence interval, 0.64–5.93; $p=0.0152$). The results of the above two sensitivity analyses suggest that Anyu Peibo capsules may be more effective than placebo. Secondary efficacy endpoints, including the response rate according to the MADRS score, the remission rate according to the MADRS score, the response rate according to the HAMD-17 score, the remission rate according

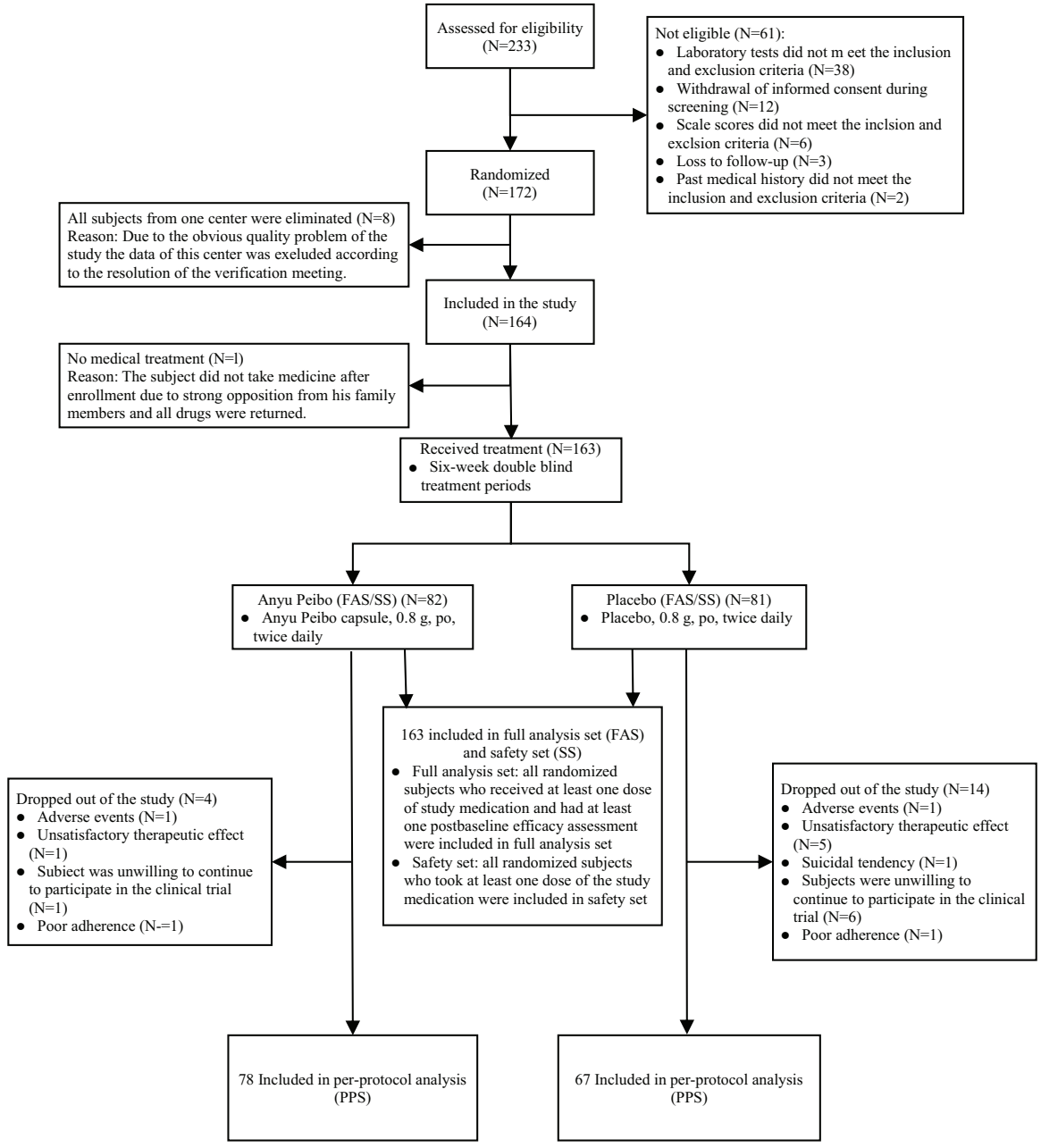

Figure 1. Consolidated standards of reporting trials (CONSORT) diagram.

to the HAMD-17 score, and the change in the HAMD-17, CGI-S, CGI-I and HAMA scores from baseline to week 6, were all numerically higher for the Anyu Peibo capsule group than for the placebo group (Table 2). However, Anyu Peibo capsules did not significantly improve the other secondary outcomes except for the change in the HAMD-17 score from baseline to week 6 and the reduction in the HAMA total score (Table 2). Significant improvements over placebo were observed for Anyu Peibo capsules when the change in the HAMD-17 score from baseline to week 6 was evaluated using the mixed model for repeated measures as the secondary analysis ($t=2.01$; 95% confidence interval, 0.03–4.23; $p=0.0464$). Moreover, Anyu Peibo capsules led to significantly greater improvements in the reduction (%) in the HAMA total score compared to placebo (51.90 ± 24.09 *versus* 42.09 ± 34.63, $p=0.0462$). However, no inference can be made because of the lack of adjustment for multiplicity in the secondary outcome analyses.

Table 1. Demographic and clinical characteristics of the participants between Anyu Peibo and placebo groups.

Characteristic	Anyu Peibo	Placebo	*p*
Age (mean ± SD)	37.49 ± 13.48	35.90 ± 14.27	0.4665[a]
Gender			
Male (%)	25 (30.49)	25 (30.86)	1.0000[b]
Female (%)	57 (69.51)	56 (69.14)	
Race/ethnicity			
Han (%)	80 (97.56)	80 (98.77)	1.0000[b]
Minority (%)	2 (2.44)	1 (1.23)	
Disease state			
First episode (%)	42 (51.22)	50 (61.73)	0.2073[b]
Recurrence (%)	40 (48.78)	31 (38.27)	
MADRS scores (mean ± SD)	29.20 ± 4.31	29.72 ± 4.35	0.4435[a]
HAMA scores (mean ± SD)	19.18 ± 6.59	18.37 ± 5.76	0.4035[a]
CGI-S scores (mean ± SD)			
4	52 ± 63.41	54 ± 66.67	0.8588[b]
5	22 ± 26.83	18 ± 22.22	
6	8 ± 9.76	9 ± 11.11	
HAMD-17 scores (mean ± SD)	22.67 ± 3.77	22.49 ± 3.66	0.7618[a]

[a]Independent samples *t* test.
[b]Fisher's exact test.
CGI, Clinical Global Impression; HAMA, Hamilton Anxiety Scale; HAMD-17, 17-item Hamilton Rating Scale for Depression; MADRS, Montgomery–Åsberg Depression Rating Scale; SD, standard deviation.

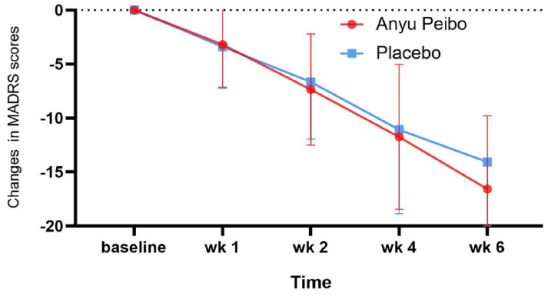

Figure 2. Change from baseline in MADRS score.
MADRS, Montgomery Åsberg Depression Rating Scale.

Adverse events

A total of 163 subjects were included in the safety analysis set, including 82 in the Anyu Peibo capsule group and 81 in the placebo group. There was no significant difference in the overall incidence of Aes between Anyu Peibo capsule and placebo groups (52.44% *versus* 48.15%, $p = 0.6396$). Common AEs (incidence of 5% or more) in patients treated with Anyu Peibo capsules mainly included upper respiratory tract infection (14.63%), nausea (7.32%), constipation (6.10%) and diarrhea (6.10%). Patients receiving placebo generally had a lower incidence of Aes than Anyu Peibo-treated patients. No AE with an incidence rate of 5% or more was observed in the placebo group. Some AE incidence rates in the Anyu Peibo capsule group were two or more times those in the placebo group, including upper respiratory tract infection, nausea, constipation, diarrhea, positive urinary leucocyte tests, insomnia, epigastric pain, decreased T wave amplitude in ECG and increased blood bilirubin levels. The

Table 2. Primary and secondary efficacy endpoints assessed after 6 weeks treatment.

End point	Anyu Peibo	Placebo	Difference (95% CI)	*p*
Primary endpoint				
The least squares mean change in the MADRS score from baseline to week 6 (SE)	16.59 (0.89)	14.51 (0.96)	2.07 (–0.27, 4.41)	0.0819[a]
Secondary endpoint				
MADRS response rate (%)	65.38	57.97	7.41 (–8.31, 23.13)	0.79[b]
MADRS remission rate (%)	42.31	34.78	7.53 (–8.18, 23.23)	0.3319[b]
HAMD-17 response rate	58.75	50.67	8.08 (–7.55, 23.72)	0.3197[b]
HAMD-17 remission rate	35.00	26.67	8.33 (–6.14, 22.80)	0.2138[b]
Change of HAMD-17 from baseline	12.00 ± 6.12	9.87 ± 7.09	2.13 (0.03, 4.23)	0.0464[c]
Change of CGI-S from baseline	1.95 ± 1.06	1.74 ± 1.26	–	0.2990[d]
Change of CGI-I from baseline	1.86 ± 0.82	2.00 ± 1.14	–	0.3627[d]
Change of HAMA from baseline	9.60 ± 5.19	8.06 ± 6.66	1.54 (–0.39, 3.48)_	0.1169[c]
Reduction in HAMA total score compared with baseline (%)	51.90 ± 24.09	42.09 ± 34.63	9.81(0.17, 19.44)	0.0462[c]

[a]The mixed-effects model for repeated measures analysis.
[b]Cochran-Mantel-Haenszel test.
[c]Independent sample *t*-test.
[d]Mann–Whitney test. Data are presented as mean ± standard deviation unless otherwise indicated.
CGI-I, Clinical global impression – improvement Scale; CGI-S, Clinical Global Impression – Severity Scale; CI, confidence interval; HAMA, Hamilton Anxiety Scale; HAMD-17, 17-item Hamilton Rating Scale for Depression; MADRS, Montgomery–Åsberg Depression Rating Scale; SE, standard error.

incidence rates of upper respiratory tract infection (14.63% *versus* 2.47%, $p = 0.0095$) and nausea (7.32% *versus* 0%) in the Anyu Peibo capsule group were higher than that in the placebo group (Table 3). Upper respiratory tract infection and nausea are both commonly reported AEs in clinical trials of antidepressant drugs. In this study, the majority of these AEs were mild. Only one case of upper respiratory tract infection and one patient with nausea experienced moderate AEs. There were no serious AEs related to respiratory tract infection or nausea. Moreover, none of the patients withdrew from the study specifically due to upper respiratory tract infection or nausea.

There were no deaths during this trial. Serious Aes (SAEs) were reported by one patient in the Anyu Peibo capsule group (1.22%) and one patient in the placebo group (1.23%). The SAEs were soft tissue injury and suicide attempt, respectively. One patient receiving placebo and one patient receiving Anyu Peibo capsules had Aes that led to study discontinuation. The Aes that led to discontinuation were insomnia (one Anyu Peibo-treated patient), nausea and dysgeusia (one patient receiving placebo). There were no significant differences in Aes, SAEs or Aes that led to study discontinuation between the two groups ($p = 0.6396$, $p = 1.0000$, and $p = 1.0000$, respectively).

Discussion

Anyu Peibo capsules are an innovative natural medicine product. Anyu Peibo is extracted from Piper laetispicum C. DC. (Piperaceae), which is a climbing glabrous plant that grows in southern China and is used for invigorating circulation, reducing detumescence and stasis, and its analgesic effects.[7] Piper laetispicum has been

Table 3. Analysis of the incidence of AEs > 2%.

Item	Anyu Peibo			Placebo			*p* Value
	Aes reported	Cases	Incidence (%)	Aes reported	Cases	Incidence (%)	
Upper respiratory tract infection*	12	12	14.63	2	2	2.47	0.0095[a]
Nausea*	7	6	7.32	0	0	0	0.0284[a]
Constipation*	7	5	6.1	1	1	1.23	0.2101[a]
Diarrhea*	5	5	6.1	2	2	2.47	0.4429[a]
Elevated lipid	4	4	4.88	2	2	2.47	0.6816[a]
Headache	6	4	4.88	3	3	3.7	1.0000[a]
Positive urinary leucocyte*	4	3	3.66	1	1	1.23	0.6204[a]
Insomnia*	4	3	3.66	1	1	1.23	0.6204[a]
Mouth dryness	2	2	2.44	2	2	2.47	1.0000[a]
Abdominal discomfort	2	2	2.44	1	1	1.23	1.0000[a]
Epigastric pain*	2	2	2.44	0	0	0	0.4969[a]
Decreased T wave amplitude in ECG*	2	2	2.44	0	0	0	0.4969[a]
Increased blood Bilirubin*	2	2	2.44	0	0	0	0.4969[a]
Drowsiness	2	2	2.44	3	3	3.7	0.6816[a]
Dizziness	3	2	2.44	2	2	2.47	1.0000[a]
Flatulence	1	1	1.22	2	2	2.47	0.6204[a]
Profuse sweating	1	1	1.22	2	2	2.47	0.6204[a]
Rash	1	1	1.22	2	2	2.47	0.6204[a]

[a]χ^2 test.
*The incidence rate of AEs in Anyu Peibo-treated patients was two times or more than two times higher than that in patients receiving placebo, or 0% in the placebo group and ⩾2% in the Anyu Peibo group.
AE, adverse event.

demonstrated to have an antidepressant effect as an inhibitor of 5-HT and NE reuptake through the tail suspension test and forced swimming test in mice. These findings have also been patented by the European Patent Office [International publication number: WO 2006/058487 (08.06.2006 Gazette 2006/23)]. Moreover, multiple studies have provided evidence of the notable antidepressant-like effect of the extract derived from Piper laetispicum C. DC., attributing this effect to its active component, laetispicine.[27] In a previous study, the other active ingredient, the-*N*-isobuty-3′,4′-methylenedioxy-5′-methoxy-cinnamamide, exhibited a moderate level of antidepressant activity when tested in behavioral despair experiments.[15] Moreover, Piper laetispicum extract could alleviate depressive-like behavior in CUMS mice by decreasing inflammatory cytokines and increasing BDNF.[12] Furthermore, in a previous study, it was demonstrated that

G11-5 [3-(3,4-methylenedioxy-5-trifluoromethyl phenyl)-2*E*-propenoic acid isobutyl amide] was synthesized based on the chemical structure of an active compound found in Piper laetispicum C. DC., exhibited a significant antidepressant effect through the TSPO-mediated mitophagy pathway.[28] Therefore, these studies clearly demonstrate that Piper laetispicum C. Dc. and its extract may have antidepressant effects.

This study was the first large, randomized, double-blind, placebo-controlled trial to evaluate the safety and efficacy of Anyu Peibo capsules for patients with MDD. Patients receiving fixed-dose Anyu Peibo capsules (0.8 g twice daily) and placebo both experienced significant improvement in depressive symptoms following 6 weeks of treatment. Depressive symptoms demonstrated numerical improvement in patients who received Anyu Peibo capsules compared to those who received placebo. However, based on the study's primary outcome measure, no significant difference was observed. Nonetheless, there was a noticeable trend toward clinical benefit in patients receiving Anyu Peibo capsules, especially as supported by sensitivity analysis. Additionally, Anyu Peibo capsules exhibited superior efficacy compared to the placebo in two secondary outcomes. Significant benefits were observed for Anyu Peibo capsules over placebo in terms of the change in HAMD-17 scores from baseline to week 6 and the reduction in the HAMA total score after 6 weeks of treatment.

Compared with previous trials, the magnitude of improvement for patients receiving Anyu Peibo capsules was similar to that observed in the positive efficacy study of novel antidepressants (vortioxetine, vilazodone) in recent years (the changes in the MADRS score from baseline to week 6 were 14.4–24.2 in the experimental group and 10.8–17.1 in the placebo group).[29,30] Moreover, based on the MADRS and HAMD-17 scores, the response rates of the Anyu Peibo capsule and placebo groups were 65.38% and 58.75%, respectively, and the remission rates were 42.31% and 35.00%, respectively. These rates were comparable to those reported in a previous study including SSRIs, SNRIs and the novel antidepressants vortioxetine and vilazodone (the response rate was 53–74%, and the remission rate was 31–46%).[29–31] However, the improvements in the placebo group (least-squares mean difference in change, 14.51) were higher than those seen in another study (least-squares mean difference in change: −11.0,[32] −10.3,[33] respectively), suggesting a slightly stronger placebo effect.

Thus, higher placebo-related improvements may likely degrade our ability to detect a significant difference between Anyu Peibo capsules and placebo for the primary outcome. On the other hand, there was a statistically significant difference in the expulsion rate between the two groups (4.88% in the Anyu Peibo capsule group *versus* 18.29% in the placebo group) in our trial, which indicated that expulsion rate of the placebo group was much higher than that of the Anyu Peibo capsule group. The main reasons for expulsion in the placebo group were poor efficacy and unwillingness to continue trial participation. Therefore, applying the LOCF method to deal with the missing values for primary outcome analysis could better reflect the actual efficacy of Anyu Peibo capsules.

Treatment with Anyu Peibo capsules was safe and well tolerated in this trial. During this study, the most common AEs (incidence ⩾5%) were upper respiratory tract infection, nausea, constipation and diarrhea. Serious AEs were rare, and most AEs were mild in severity. Moreover, the overall incidence of AEs was similar in both the Anyu Peibo capsule and placebo groups. No significant differences in the incidence of total AEs or severe AEs between groups were reported. Furthermore, rates of AEs leading to suspension and expulsion did not significantly differ between the Anyu Peibo capsule and placebo groups. There were no deaths or clinically significant safety signals in participants who received Anyu Peibo capsules. The incidence rate of treatment-emergent adverse events (TEAEs) observed in this study was 52.44%, which was lower than the findings of some meta-analyses.[31] An average incidence rate of 76.4% was reported for antidepressant arms in a large meta-analysis of placebo-controlled depression trials.[34] Specifically, for the medication escitalopram, the incidence of TEAEs ranged from 73.3% to 73.6%, while that of other SSRIs such as citalopram, fluoxetine, paroxetine, and sertraline was 78.2%.[31] Additionally, SNRIs (venlafaxine and duloxetine) had a TEAE incidence rate of 77.4%.[31] In comparison, the AE incidence rate of 48.15% for the placebo group in this trial was lower than the 63.0% average reported for the placebo arms in a meta-analysis.[34]

In terms of discontinuation rates due to TEAEs, the Anyu Peibo capsule group exhibited favorable results. The rate of discontinuations due to AEs in the Anyu Peibo capsule group (1.22%) was lower than the average rate of 7% reported for antidepressant arms in another comprehensive meta-analysis of placebo-controlled depression trials.[35] In contrast, the 1.23% rate of discontinuation due to AEs for the placebo group in our trial was lower than the 4% average reported for the placebo arms in that same meta-analysis. Additionally, when comparing Anyu Peibo capsules to other commonly prescribed antidepressants, the discontinuation rate for venlafaxine (37.5–225 mg/day) was 12%, while for desvenlafaxine, the discontinuation rates were 4.1%[31] at 50 mg/day and 8.5% at 100 mg/day. For vortioxetine, the discontinuation rates were 5%, 6%, 8%, and 8% for doses of 5, 10, 15, and 20 mg/day, respectively.[36] Therefore, the safety and tolerability profile of Anyu Peibo capsules appears to be superior to that of other commonly prescribed antidepressants, as indicated by these findings.

Above all, these results indicate that Anyu Peibo capsules may be safe and have the potential to reduce depressive symptoms within 6 weeks for patients with MDD. While the primary outcome of this study was negative, it is worth considering the significant trend in the primary outcome *p* value, the positive findings in the sensitivity analysis, and the presence of a large placebo effect. The discrepancy between the primary LOCF analysis and the observed case data raises questions about the robustness of the primary analysis. Therefore, further validation of the effectiveness and safety of Anyu Peibo capsules through subsequent studies is necessary.

In light of these results and the advantages of Anyu Peibo capsules as a natural product, we are currently conducting a phase III clinical trial (the Clinical Trials registry: NCT04210973).[37] The phase IIb study results provide essential research evidence for initiating phase III clinical trials. Through the phase IIb trial, we aim to gain a better understanding of the drug's adverse reactions, optimize the trial design, determine appropriate endpoints and sample sizes, and improve the success rate of the phase III trial. Furthermore, this study is the first to report on the efficacy and safety of Anyu Peibo capsules for MDD patients, providing preliminary evidence of its potential effects in improving depression symptoms.

Our study has a number of limitations. First, it had a short-term follow-up. The duration of Anyu Peibo treatment was 6 weeks. There was a potential of continuous improvement in the Anyu Peibo capsule group at the endpoint; thus, the 6-week treatment duration may not be long enough to witness the effectiveness of Anyu Peibo capsules. Second, the sample size was small, although the post hoc analysis showed that the power value was more than 0.95. Further research with larger samples and longer-term follow-up is needed to better ascertain the efficacy of this intervention among patients with MDD. We increased the treatment duration to 8 weeks in phase III trials of Anyu Peibo capsules. Third, the exclusion of patients with comorbidities is not reflective of clinical practice. Fourth, the current trial did not include an active antidepressant group, which limits comparison of the results with those of existing antidepressants. Additionally, it is important to note that this study had a relatively high placebo effect, which prevented the identification of a statistically significant efficacy advantage of Anyu Peibo capsules over placebo.

Conclusion

Anyu Peibo capsules exhibited a trend toward greater benefits in relieving depressive symptoms compared to placebo, although the difference did not reach statistical significance. Moreover, this trial provided evidence of the safety profile of Anyu Peibo capsules. These results suggest that Anyu Peibo capsules, which originate from natural products, may have an effective and safe antidepressant effect. Further research is warranted to validate these preliminary findings and enhance the design of future studies, including sample size calculation for full-scale randomized controlled trials.

Declarations

Ethics approval and consent to participate

The study was conducted at nine research sites in China in compliance with ICH-Good Clinical Practice guidelines. This study was approved by ethics committees in each site (Shanghai Mental Health Center Ethics Committee, Beijing HuiLongGuan Hospital Ethics Committee, Beijing Anding Hospital, Capital Medical University Ethics Committee, XI'AN Mental Health Center Ethics Committee, Wuhan Mental Health Center Ethics Committee, West China

Hospital Ethics Committee, Brain Hospital of Jilin Province Ethics Committee, The Second Xiangya Hospital of Central South University Ethics Committee, Jiangxi Mental Hospital Ethics Committee). Ethics approval number: IIB 2017-12. Written informed consent was obtained from all participants prior to enrollment in the study. The study was registered in the Clinical Trials registry (NCT03183505).

Consent for publication

All authors give their consent for publication.

Author contributions

Shen He: Formal analysis; Methodology; Validation; Writing – original draft.

Yimin Yu: Methodology; Writing – review & editing.

Jingjing Huang: Methodology; Writing – review & editing.

Jiahui Yin: Formal analysis; Writing – review & editing.

Yajuan Niu: Investigation; Writing – review & editing.

Yazhou Lu: Investigation; Writing – review & editing.

Bin Wu: Investigation; Writing – review & editing.

Maosheng Fang: Investigation; Writing – review & editing.

Xue Wang: Investigation; Writing – review & editing.

Zhiping Tao: Investigation; Writing – review & editing.

Lehua Li: Investigation; Writing – review & editing.

Kan Li: Investigation; Writing – review & editing.

Yan Li: Methodology; Writing – review & editing.

Xiujuan Ding: Formal analysis; Writing – review & editing.

Yifeng Shen: Conceptualization; Project administration; Resources; Supervision; Writing – review & editing.

Huafang Li: Conceptualization; Project administration; Resources; Supervision; Writing – review & editing.

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Competing interests

All authors declare no direct conflicts with this work. Xiujuan Ding is an employee of Suzhou YiHua Biomedical Technology Co., Ltd.

Availability of data and materials

The data that support the findings of this study are available from the corresponding author on reasonable request.

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References

1. Global Health Estimates. *Depression and other common mental disorders*. Geneva: World Health Organization, 2017. Licence: CC BY-NC-SA 3.0 IGO.
2. Benziger CP, Roth GA and Moran AE. The global burden of disease study and the preventable burden of NCD. *Glob Heart* 2016; 11: 393–397.
3. UN Department of Economic and Social Affairs-Issue: Mental Health and Development, https://www.un.org/development/desa/disabilities/issues/mental-health-and-development.html (2021).
4. Jackson WC, Papakostas GI, Rafeyan R, *et al.* Recognizing inadequate response in patients with major depressive disorder. *J Clin Psychiatry* 2020; 81: OT19037BR2.
5. Lundberg J, Cars T, Lööv SÅ, *et al.* Association of treatment-resistant depression with patient outcomes and health care resource utilization in a population-wide study. *JAMA Psychiatry* 2023; 80: 167–175.
6. Martins J and Brijesh S. Phytochemistry and pharmacology of anti-depressant medicinal

plants: a review. *Biomed Pharmacother* 2018; 104: 343–365.

7. Wang MT, Wang JH, Zhao KK, *et al*. Complete plastome sequence of Piper laetispicum (Piperaceae): an endemic plant species in South China. *Mitochondrial DNA B Resour* 2018; 3: 1035–1036.
8. Lu D, Wang Z, Gao S, *et al*. A rapid and sensitive LC-MS/MS method for determination of the active component K6 in serum of patients with depression. *J Pharm Biomed Anal* 2022; 213: 114691.
9. Yao CY, Wang J, Dong D, *et al*. Laetispicine, an amide alkaloid from Piper laetispicum, presents antidepressant and antinociceptive effects in mice. *Phytomedicine* 2009; 16: 823–829.
10. Xie H, Yan M, Jin D, *et al*. Studies on antidepressant and antinociceptive effects of ethyl acetate extract from Piper laetispicum and structure-activity relationship of its amide alkaloids. *Fitoterapia* 2011; 82: 1086–1092.
11. Pan S, Xie J, Qian F, *et al*. Antidepressant amides from Piper laetispicum C. DC. *Yao Xue Xue Bao* 2005; 40: 355–357.
12. Xie H, Jin D, Kang Y, *et al*. The effect of Piper laetispicum extract (EAE-P) during chronic unpredictable mild stress based on interrelationship of inflammatory cytokines, apoptosis cytokines and neurotrophin in the hippocampus. *BMC Complement Altern Med* 2015; 15: 240.
13. Li Q, Qu FL, Gao Y, *et al*. Piper sarmentosum Roxb. produces antidepressant-like effects in rodents, associated with activation of the CREB-BDNF-ERK signaling pathway and reversal of HPA axis hyperactivity. *J Ethnopharmacol* 2017; 199: 9–19.
14. Perviz S, Khan H and Pervaiz A. Plant alkaloids as an emerging therapeutic alternative for the treatment of depression. *Front Pharmacol* 2016; 7: 28.
15. Han M, Ma X, Jin Y, *et al*. Synthesis and structure-activity relationship of novel cinnamamide derivatives as antidepressant agents. *Bioorg Med Chem Lett* 2014; 24: 5284–5287.
16. American Psychiatric Association, DSM-5 Task Force. *Diagnostic and statistical manual of mental disorders: DSM-5™*. 5th ed. American Psychiatric Publishing, Inc, United States of America, 2013.
17. Zheng YP, Zhao JP, Phillips M, *et al*. Validity and reliability of the Chinese Hamilton depression rating scale. *Br J Psychiatry* 1988; 152: 660–664.
18. Zhong B, Wang Y, Chen H, *et al*. Reliability, validity and sensitivity of Montgomery-Asberg depression rating scale for patients with current major depression disorder. *Chin J Behav Med Brain Sci* 2011; 1: 85–87.
19. Ma S, Yang J, Yang B, *et al*. The patient health questionnaire-9 vs. the Hamilton rating scale for depression in assessing major depressive disorder. *Front Psychiatry* 2021; 12: 747139.
20. Maier W, Buller R, Philipp M, *et al*. The Hamilton anxiety scale: reliability, validity and sensitivity to change in anxiety and depressive disorders. *J Affect Disord* 1988; 14: 61–68.
21. Zhang MY. *Psychiatry assessment scale manual*. Changsha: Hunan Science & Technology Press, 1998, pp. 39–42.
22. Guy W. ECDEU assessment manual for psychopharmacology. US Department of Health, Education, and Welfare, Public Health Service, Alcohol, Drug Abuse, and Mental Health Administration, National Institute of Mental Health, Psychopharmacology Research Branch, Division of Extramural Research Programs, Rockville, Md., 1976.
23. Zhang T, Huang Y, Jin Y, *et al*. Treatment for major depressive disorder by repetitive transcranial magnetic stimulation in different parameters: a randomized double-blinded controlled trial. *Front Psychiatry* 2021; 12: 623765.
24. Yu YM, Gao KR, Yu H, *et al*. Efficacy and safety of agomelatine vs paroxetine hydrochloride in Chinese Han patients with major depressive disorder: a multicentre, double-blind, noninferiority, randomized controlled trial. *J Clin Psychopharmacol* 2018; 38: 226–233.
25. Su L, Jia Y, Liang S, *et al*. Multicenter randomized controlled trial of bifrontal, bitemporal, and right unilateral electroconvulsive therapy in major depressive disorder. *Psychiatry Clin Neurosci* 2019; 73: 636–641.
26. Su L, Zhang Y, Jia Y, *et al*. Predictors of electroconvulsive therapy outcome in major depressive disorder. *Int J Neuropsychopharmacol* 2023; 26: 53–60.
27. Xie H, Liu J, Yu M, *et al*. Structure-activity relationship of 39 analogs of laetispicine with antidepressant properties. *Am J Chin Med* 2013; 41: 1377–1392.
28. Wei Q, Zhou W, Zheng J, *et al*. Antidepressant effects of 3-(3,4-methylenedioxy-5-trifluoromethyl phenyl)-2E-propenoic acid isobutyl amide involve TSPO-mediated

mitophagy signalling pathway. *Basic Clin Pharmacol Toxicol* 2020; 127: 380–388.

29. Jacobsen PL, Mahableshwarkar AR, Serenko M, *et al*. A randomized, double-blind, placebo-controlled study of the efficacy and safety of vortioxetine 10 mg and 20 mg in adults with major depressive disorder. *J Clin Psychiatry* 2015; 76: 575–582.
30. Mathews M, Gommoll C, Chen D, *et al*. Efficacy and safety of vilazodone 20 and 40 mg in major depressive disorder: a randomized, double-blind, placebo-controlled trial. *Int Clin Psychopharmacol* 2015; 30: 67–74.
31. Mi W, Di X, Wang Y, *et al*. A phase 3, multicenter, double-blind, randomized, placebo-controlled clinical trial to verify the efficacy and safety of ansofaxine (LY03005) for major depressive disorder. *Transl Psychiatry* 2023; 13: 163.
32. Croft HA, Pomara N, Gommoll C, *et al*. Efficacy and safety of vilazodone in major depressive disorder: a randomized, double-blind, placebo-controlled trial. *J Clin Psychiatry* 2014; 75: e1291–e1298.
33. Gunduz-Bruce H, Silber C, Kaul I, *et al*. Trial of SAGE-217 in patients with major depressive disorder. *N Engl J Med* 2019; 381: 903–911.
34. Cheung CP, Thiyagarajah MT, Abraha HY, *et al*. The association between placebo arm inclusion and adverse event rates in antidepressant randomized controlled trials: an examination of the Nocebo effect. *J Affect Disord* 2021; 280: 140–147.
35. Schalkwijk S, Undurraga J, Tondo L, *et al*. Declining efficacy in controlled trials of antidepressants: effects of placebo dropout. *Int J Neuropsychopharmacol* 2014; 17: 1343–1352.
36. TRINTELLIX (vortioxetine) tablets, for oral use Initial U.S. Approval: 2013.
37. Huang J, Yu Y, Jiang Y, *et al*. The efficacy and safety of Anyu Peibo Capsule in the treatment of patients with major depressive disorder in China: study protocol for a randomized placebo-controlled trial. *Trials* 2021; 22: 585.