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EDITED BY
Xin Xu,
Sichuan University, China

REVIEWED BY
Delong Zhang,
South China Normal University, China
Sheng Li,
Guangzhou University of Chinese Medicine, China

*CORRESPONDENCE
Zhuo-Xin Yang
001188@gzucm.edu.cn
Min- Pi
pm0305@gzucm.edu.cn

†These authors have contributed equally to this work

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Fecal microbiota as a predictor of acupuncture responses in patients with postpartum depressive disorder

Yu-Mei Zhou[1†], Jin-Jun Yuan[1†], Yu-Qin Xu[1], Yan-Hua Gou[1], Yannas Y. X. Zhu[1], Chen Chen[2], Xing-Xian Huang[1], Xiao-Ming Ma[1], Min- Pi[1*] and Zhuo-Xin Yang[1*]

[1]Department of Acupuncture, The Fourth Clinical Medical College of Guangzhou University of Chinese Medicine, Shenzhen, Guangdong, China, [2]Department of Acupuncture and Tuina, Shenzhen Maternal and Child Health Care Hospital, Shenzhen, China

Background: There are several clinical and molecular predictors of responses to antidepressant therapy. However, these markers are either too subjective or complex for clinical use. The gut microbiota could provide an easily accessible set of biomarkers to predict therapeutic efficacy, but its value in predicting therapy responses to acupuncture in patients with depression is unknown. Here we analyzed the predictive value of the gut microbiota in patients with postpartum depressive disorder (PPD) treated with acupuncture.

Methods: Seventy-nine PPD patients were enrolled: 55 were treated with acupuncture and 24 did not received any treatment. The 17-item Hamilton depression rating scale (HAMD-17) was used to assess patients at baseline and after eight weeks. Patients receiving acupuncture treatment were divided into an acupuncture-responsive group or non-responsive group according to HAMD-17 scores changes. Baseline fecal samples were obtained from the patients receiving acupuncture and were analyzed by high-throughput 16S ribosomal RNA sequencing to characterize the gut microbiome.

Results: 47.27% patients responded to acupuncture treatment and 12.5% patients with no treatment recovered after 8-week follow-up. There was no significant difference in α-diversity between responders and non-responders. The β-diversity of non-responders was significantly higher than responders. *Paraprevotella* and *Desulfovibrio* spp. were significantly enriched in acupuncture responders, and these organisms had an area under the curve of 0.76 and 0.66 for predicting responder patients, respectively.

Conclusions: *Paraprevotella* and *Desulfovibrio*are may be useful predictive biomarkers to predict PPD patients likely to respond to acupuncture. Larger studies and validation in independent cohorts are now needed to validate our findings.

KEYWORDS

fecal microbiota, postpartum depressive disorder, acupuncture, the gut-brain axis, predictive biomarker, *Paraprevotella*, *Desulfovibrio*

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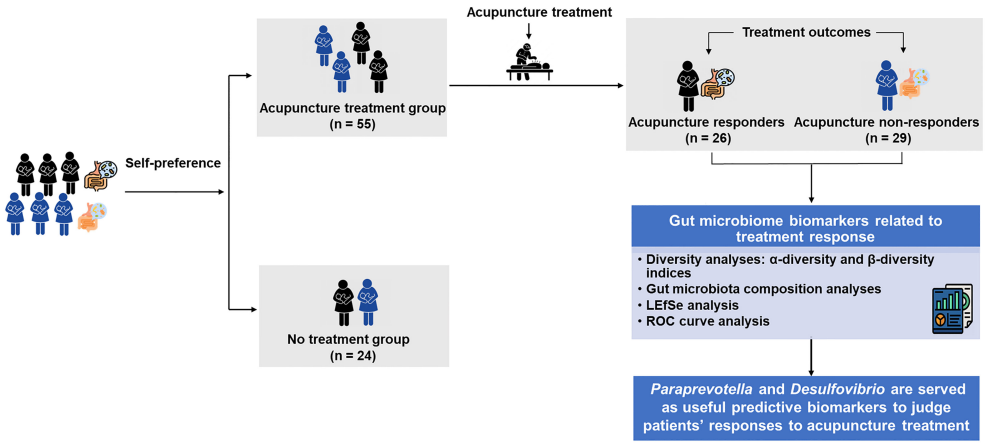

GRAPHICAL ABSTRACT

1 Introduction

Postpartum depressive disorder (PPD) is a common, disabling, but treatable psychiatric condition (Howard et al., 2014). However, without prompt diagnosis and treatment, maternal suicide and infanticide may be extreme outcomes of PPD (Grigoriadis et al., 2017; Netsi et al., 2018). With a global prevalence of 17.22% (Wang et al., 2021), PPD is a significant maternal and family health burden worldwide. While PPD is most commonly treated with antidepressants and psychological therapies, the efficacy of these approaches varies due to high clinical and functional heterogeneity (Consortium, P.D.A.T.C.a.T.P, 2015; Santos et al., 2017). Indeed, antidepressants have been reported to be only ~42% effective (Brown et al., 2021), and psychotherapy only benefits about a third of PPD patients (Huang et al., 2020). Furthermore, antidepressants have side effects (Carvalho et al., 2016), and any adverse events to the baby during lactation must also be considered (Davanzo et al., 2011). Psychotherapy cannot generally be widely used due to its high cost over long periods of time. An increasing number of PPD patients are seeking safe and effective complementary treatments with few side effects.

Acupuncture is safe and effective in pregnant women (Ormsby et al., 2016; Li et al., 2018; Tong et al., 2019; Li et al., 2020). In a study of 31 meta-analyses and 59 randomized controlled trials, acupuncture was shown to be superior to awaiting treatment, control acupuncture (invasive or non-invasive sham control), and antidepressants in terms of reducing the severity of depression (Hamilton, 1960; Li et al., 2020). Another relatively recent meta-analysis highlighted that acupuncture can significantly reduce Hamilton depression rating (HAMD) scores in PPD patients (Li et al., 2019). However, just like other treatments, the efficacy of acupuncture varies between individuals. PPD therapy urgently requires specific biomarkers to predict therapeutic responses to antidepressant treatments, including acupuncture, so that the correct patients can be prescribed the best treatments at the right time.

Several demographic and clinical therapeutic response predictors to traditional antidepressants in PPD have been reported in robust clinical trials including being white/non-Hispanic (Yonkers et al., 2008), having a major depressive episode within four weeks of delivery (Sharp et al., 2010; Hantsoo et al., 2014), concomitant anxiety symptoms (Cohen et al., 2001), an absence of concomitant psychiatric illness (Yonkers et al., 2008), early response to treatment (Cohen et al., 2001), and improvement within one week after initiation of antidepressants (Appleby et al., 1997). Predictors of non-response included a lifetime history of substance use disorder (Yonkers et al., 2008), concomitant anxiety symptoms (Nonacs et al., 2005), and Hispanic or Black ethnicity (Yonkers et al., 2008). However, the generalizability of these predictors is limited by significant methodological variability including a wide range of studied postpartum periods (2–24 months), comorbid diseases (lifetime alcohol abuse, alcohol dependence, drug abuse, drug dependence, or anxiety disorder) (Nonacs et al., 2005; Suri et al., 2005; Misri et al., 2016), and different severities of depression (minor depression or major depression) (Appleby et al., 1997). In recent years, it has been found that the consistency of quantitative electroencephalographic, the default pattern network with different discrete topological structures in the left and right hemispheres and the variance of the global signal are related to the terminal clinical results of antidepressant treatment of MDD (Hunter et al., 2010; Hou et al., 2016; Zhu et al., 2018), however, the acquisition of the above indicators is undoubtedly complicated, and there are few research results on the efficacy prediction of PPD. Furthermore, predictors focus on clinicodemographic factors and there have been few studies on biomarkers– such as genetic and inflammatory markers (Sharma et al., 2020) – to advance the goal of developing objective and clinically acceptable biomarkers that predict treatment outcomes and guide individualized therapy.

There is now mounting evidence supporting a role for the intestinal microbiota in mental health disorders (Rieder et al., 2017; Dubois et al., 2019; Nikolova et al., 2021; McGuinness et al., 2022). This biochemical signaling pathway, also known as the gut-brain axis, is thought to influence cognitive function and mood via neural, metabolic, hormonal, and immune-mediated mechanisms (Foster

and McVey Neufeld, 2013). Previous studies (Chung et al., 2019; Zhou et al., 2020; Nikolova et al., 2021) have found differences in the diversity and composition of gut microbial communities between PPD patients and healthy controls. Changes in the intestinal microflora can affect the efficacy of treatment for some diseases (Ma et al., 2019), and intestinal microflora has recently been shown to be a non-invasive diagnostic biomarker for colorectal adenoma and cancer (Liang et al., 2020). In a systematic review, probiotic therapy showed modest benefits in alleviating depressive symptoms in patients with major depressive disorder over four to nine weeks (Alli et al., 2022). Furthermore, *Lactobacillus rhamnosus* HN001 administered as a probiotic significantly reduced maternal depression and anxiety scores (Slykerman et al., 2017). It is also found that 919 syrup can relieve PPD by regulating the structure and metabolism of intestinal microorganisms and affecting the function of GABA/glutamic acid system in hippocampus (Tian et al., 2021). Additionally, it can be also used to predict responses to cancer immunotherapy in metastatic melanoma patients (Limeta et al., 2020), and dynamic changes in the intestinal microbiota can provide an early prediction of immunotherapy outcomes in patients with hepatocellular carcinoma (Zheng et al., 2019). Recently, some studies have also shown that responses to antipsychotic drugs are related to gut microbiota composition (Schwarz et al., 2018). It is found that the changes of intestinal microbial composition and metabolic function may be related to the response of antidepressants, which provides a potential predictor for the prediction of the curative effect of MDD and can even be used to distinguish MDD from generalized anxiety disorder (Dong et al., 2021; Dong et al., 2022). Therefore, characterizing the nature and impact of the intestinal microbiota on PPD therapy and its value as a biomarker of therapeutic responses would be highly clinically valuable. Acupuncture, as a common complementary alternative therapy, can reduce depressive-like behaviors in chronic unpredictable mild stress (CUMS) rats by regulating intestinal microbes and neurotransmitters (Li et al., 2021). Jiang found that acupuncture can effectively treat all stages of stroke and regulate intestinal flora, thus improving depressive symptoms (Jiang et al., 2023). Therefore, the intestinal microflora may act as clinically relevant biomarkers of therapeutic responses in individuals with mental health diseases, including in those receiving acupuncture.

Here, we first aimed to assess the efficacy of acupuncture in PPD patients. A secondary aim was to identify any differences in the intestinal microbiota in responders and non-responders to acupuncture, with the objective to identify microbiome-based predictors of acupuncture response.

2 Materials and methods

2.1 Study design

This was a prospective cohort study approved by the Ethics Committee of Shenzhen Hospital of Traditional Chinese Medicine [K2020-027-01]. The study was registered with the Chinese Clinical Trial Registry (http://www.chictr.org.cn/index.aspx; ChiCTR2100041687). Patients with PPD were recruited from the Shenzhen Traditional Chinese Medicine Hospital and Shenzhen Maternity & Child Healthcare Hospital (Shenzhen, China). All procedures used in this study conformed to the ethical standards of national and institutional human experimental committees and the Declaration of Helsinki. All subjects supplied written informed consent (Graphic Abstract).

2.2 Participant recruitment

Patients were initially screened for PPD using the Edinburgh Postnatal Depression Scale (EPDS) and then further evaluated using the 17-item Hamilton depression rating scale (HAMD-17) by physicians. All patients were assigned into acupuncture treatment group or no treatment group according to their own preference.

2.3 Diagnostic criteria

PPD was diagnosed by the evaluating physician according to the Fifth Edition of the Diagnosis and Statistics of Mental Illness (*DSM-V*) (Battle, 2013; First, 2013). Patients needed to meet five or more of the following symptoms, including at least the first or second symptoms, and the symptoms should have lasted for at least two weeks: (1) low mood and depressive emotion; (2) lack of interest in or loss of enjoyment in activities; (3) significant weight gain or loss; (4) poor sleep, insomnia, or lethargy; (5) psychomotor excitement or retardation; (6) a feeling of fatigue or weakness; (7) a sense that life is worthless, self-accusation, or self-guilt; (8) decline in cognition or difficulty concentrating; and (9) recurrent thoughts of death.

2.4 Inclusion and exclusion criteria

The inclusion criteria were: (1) patients between 20 and 49 years of age; (2) a diagnosis of PPD made by a psychiatrist; (3) illness appearing within a year of delivery; (4) HAMD-17 scores between 7 and 24; and (5) providing informed consent, voluntarily participating in the study, and able to complete the assessment instrument.

Exclusion criteria were: (1) severe psychiatric disorders such as bipolar affective disorder and schizophrenia; (2) mental disorder due to brain diseases or for other reasons, and unable to understand the contents of the questionnaire and cannot be effectively evaluated; (3) pregnancy; (4) patients with a HAMD suicide score >2 points; (5) anyone attempting suicide in the past year; and (6) anyone taking antibiotics or probiotics in the past month.

2.5 Interventions

Patients in the acupuncture group were treated with acupuncture therapy by an acupuncturist with a doctor's license and at least three years of clinical experience. Before patient enrollment, all acupuncturists participated in standardized operating procedure training, including locating the acupoints and needle manipulation.

The acupoints selected in this study including Baihui (DU20), Yintang (EX-HN3), Zhongwan (RN12), Qihai (RN6), Guanyuan (RN4), Neiguan (PC6), Shenmen (HT7), Hegu (LI4), Sanyinjiao (SP6) and Taichong (LR3). The location of acupoints has been shown in Table 1. When participants were supine, the skin around acupoints were routinely sterilized with 75% alcohol cotton swab, then disposable sterile needles (Product type: HuanQiu, Suzhou, China; 0.3 mm × 40 mm/0.3 mm × 75 mm; C-160630) were inserted into each acupoint to achieve the *deqi* sensation (a sensation of soreness, numbness, swelling, or radioactivity indicating the effectiveness of acupuncture). Paired alligator clips from the electroacupuncture (EA) apparatus (Hwato brand, Suzhou Medical Appliance Factory) were attached transversely to the needle holders at Baihui (DU20) and Yintang (EX-HN3), Zhongwan (RN12) and Qihai (RN6). The EA stimulation lasted for 30 minutes with a continuous wave of 2Hz and a current intensity of 0.1 to 1 mA depending on the participants comfort level. Acupuncture treatment consisted of 16 sessions, each for 30 minutes, and were administered over 8 weeks.

Patients in the no treatment group didn't receive any therapy.

2.6 Clinical outcomes

The clinical outcome was the response rate. The HAMD is a commonly used scale for clinical evaluation of depressive state (Hamilton, 1960). Depressive symptoms of PPD patients were assessed by HAMD-17 scale (17 items, scored from 0 to 52, higher scores representing more severe the depressive symptoms).

TABLE 1 Localization of acupoints selected in this trial.

Acupoint	Localization
Baihui (DU20)	7 cun above the middle of the posterior hairline, on the middle of the top of the head
Yintang (EX-HN3)	On the forehead, between the brows
Zhongwan (RN12)	On the anterior median line, 4 cun superior to the umbilicus
Qihai (RN6)	On the anterior median line, 1.5 cun caudal to the umbilicus
Guanyuan (RN4)	On the anterior median line, 3 cun caudal to the umbilicus
Neiguan (PC6)	2 cun proximal to the processus styloideus radii, between the tendons of the palmaris longus and the flexor carpi radialis
Shenmen (HT7)	In the wrist, the ulnar end of the transverse striation of the carpal palmar, and the radial depression of the flexor tendon of the ulnar carpal
Hegu (LI4)	On the highest point at m. interosseus dorsalis
Sanyinjiao (SP6)	3 cun proximal to the medial malleolus
Taichong (LR3)	Between metatarsal I and II, just distal to the caput

Patients were defined as responders if the HAMD-17 score reduced by ≥50% or the HAMD-17 score was <7 after treatment. Patients were defined as non-responders if the reduction in HAMD-17 score was <50% (Keller, 2003).

2.7 Fecal samples collection

Fecal samples of PPD participants in acupuncture treatment group were collected once at baseline and placed in sterile plastic cups, then frozen at −80°C immediately after defecation. The details of fecal sample collection are described elsewhere (Zhou et al., 2019).

2.8 DNA extraction and 16S ribosomal RNA gene sequencing

DNA was extracted using the MOBIO PowerSoil® DNA Separation Kit according to the manufacturer's instructions, and stored at −80° in Tris-EDTA buffer solution before microbial MiSeq sequencing. The V4 region of 16S rRNA gene was amplified by PCR with primers 515F (5'-GTGYCAGCMGCCGCGGTAA-3') and 806R (5'-GGACTACNVGGGTWTCTAAT-3'), along with barcode sequences, as previously described (Zhou et al., 2020). PCR mixtures contained 1 μl of each forward and reverse primer (10μM), 1 μl of template DNA, 4 μl of dNTPs (2.5mM), 5 μl of 10× EasyPfu Buffer, 1 μl of Easy Pfu DNA Polymerase (2.5 U/μl), and 1 μl of double-distilled water in a 50-μl reaction volume. Thermal cycling consisted of an initial denaturation step at 95° for 5min, followed by 30 cycles of denaturation at 94° for 30 s, annealing at 60° for 30 s, and extension at 72° for 40 s, with a final extension step at 72° for 4min. Amplicons were run for each sample on an agarose gel. Expected band size for 515f-806r was ~300–350 bp. Amplicons were quantified with Quant-iT PicoGreen dsDNA Assay Kit (P11496; Thermo Fisher Scientific, Waltham, MA, USA) according to manufacturer's instructions. The amplicon library for high-throughput sequencing on the Illumina MiSeq V3 reagent PE150 (300 cycles) platform was combined to an equal amount and subsequently quantified using KAPA Library Quantification Kit (KK4824; Illumina, Inc., San Diego, CA, USA) according to manufacturer's protocols.

2.9 High-throughput sequencing of 16S ribosomal RNA gene and microbial analysis

High-throughput sequencing analysis was performed using Quantitative Insights into Microbial Ecology (QIIME) 2.0 according to the manufacturer's instructions. Raw Illumina read data were deposited into tags, reads belonging to each sample were separated with barcodes, and low-quality reads were removed. The processed tags were clustered into amplicon sequence variants (ASVs) using the commonly used 97% similarity threshold. ASVs were assigned to taxa by matching to the SILVA database. A phylogenetic tree of representative sequences was constructed. α-

diversity indices such as evenness, observed species, Shannon, and Faith-PD indices were calculated by Wilcoxon rank sum test. For β-diversity indices, firstly, Wilcoxon rank sum test was used to analyze the inter-group and intra-group differences. The former indicated differences in microbial composition between samples within the same group; the later indicate the differences in microbial composition of pairwise samples from different groups. Secondly, the Bray-Curtis dissimilarity and unweighted unifrac calculated by principal coordinate analyses were used for β-diversity indices. To further identify specific bacteria as biomarkers at the genus level, linear discriminant analysis effect size (LEfSe) was applied through the Huttenhower Lab Galaxy Server (Segata et al., 2011) after taxa summaries were reformatted. LEfSe settings were as previously described (Zhou et al., 2020), and systemic forms with a linear discriminant analysis (LDA) cutoff of 2.0 and a $P < 0.05$ in the built-in rank sum test were considered statistically significant. Finally, biomarker data (specific bacteria) calculated by LEfSe were further analyzed by receiver operator characteristic (ROC) curve analysis, and area under the curve (AUC) was used to assess the ROC effect. The cut-off value associated with optimal sensitivity and specificity was used to distinguish acupuncture responders and non-responders.

2.10 Analysis of clinical data

The demographic and clinical outcomes were analyzed using the SPSS 22.0 software (IBM Statistics, Armonk, NY, USA). Normally distributed data were analyzed using Student's *t*-test, while non-parametric data were analyzed using the Mann-Whitney U-test with data expressed as medians with interquartile ranges (IQR). Categorical data were compared using the chi-squared test. A *P*-value < 0.05 was considered statistically significant.

3 Results

Among 179 patients screened, 88 were enrolled at baseline between March 25 and November 22, 2021. According to patient preference, 60 received acupuncture treatment (acupuncture group) and 28 received no treatment (control group). During the study, nine (10.23%) patients dropped out: five (8.33%) received acupuncture and four (14.29%) had not received acupuncture. Seventy-nine patients completed the eight-week follow-up and assessments (Figure 1).

3.1 Effectiveness of acupuncture therapy on PPD

3.1.1 Clinical characteristics of acupuncture therapy and control patients

The baseline demographic and clinical characteristics are shown in Table 2. There were no significant differences in age, body mass index (BMI), number of days postpartum, number of parturitions, duration of disease, delivery mode, family history, and EPDS or HAMD-17 scores between those receiving acupuncture and those not receiving acupuncture at baseline.

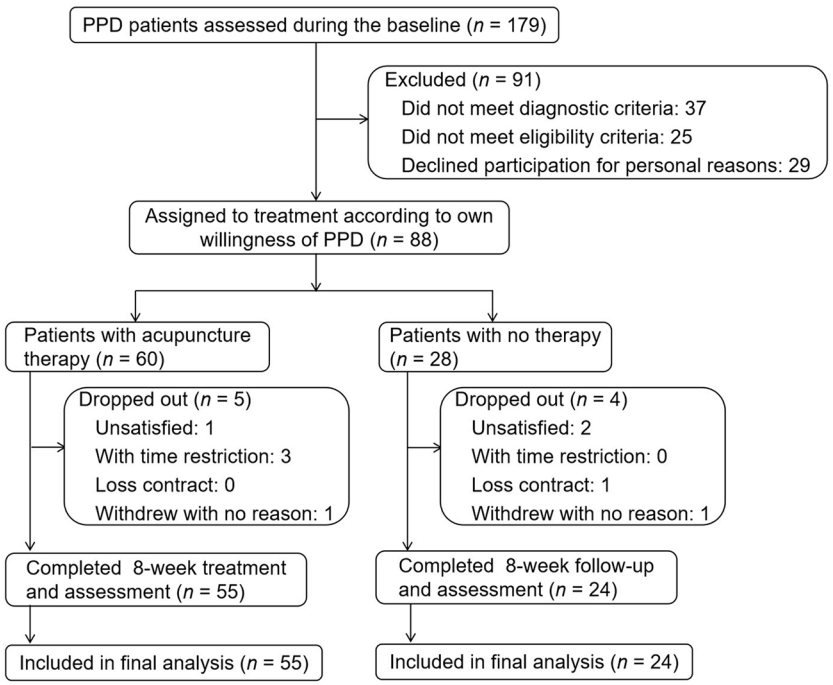

FIGURE 1
Trial flow diagram.

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TABLE 2 Clinical characteristics of patients in the acupuncture therapy and the control groups.

Characteristic	Acupuncture therapy group (N = 55)	Control group (N = 24)	*P*-value
Age (years, mean ± SD)	35.35 ± 4.01	32.21 ± 3.16	0.001
BMI ($kg.m^{-2}$, mean ± SD)	22.56 ± 3.31	21.58 ± 3.71	0.249
Number of days postpartum (days, mean ± SD)	142.24 ± 101.67	125.83 ± 87.60	0.185
Number of parturition(n)	1.53 ± 0.60	1.33 ± 0.57	0.470
Duration of disease (months)	3.78 ± 2.84	3.27 ± 2.52	0.449
Delivery mode (A/B/C)*	32/22/1	18/6/0	0.326
Family history (y/n)	4/51	1/23	0.602
EPDS	15.67 ± 4.57	15.71 ± 5.14	0.976
HAMDS	14.00 ± 3.51	13.33 ± 4.78	0.544

*A, Natural childbirth; B, Cesarean section; C, Women miscarried but pregnant for more than seven months.

3.1.2 Comparison of response rates between groups

47.27% responded to acupuncture treatment in the acupuncture group, and 12.5% patients not receiving treatment recovered after 8-week follow up. This difference was significant ($P = 0.003$) (Table 3).

3.1.3 Comparison of HAMD reduction rate between groups

Compared with the HAMD reduction rate between two groups, the results showed reduction rate in acupuncture group was superior than that in control group. This difference was significant (P <0.001) (Table 4).

3.1.4 HAMD changes between patients receiving acupuncture and controls

Compared with baseline, HAMD scores in the control group did not significantly decrease ($P = 0.113$). However, the HAMD score decreased significantly in patients receiving acupuncture treatment (P <0.001) (Table 5).

TABLE 3 Comparison of response rates between acupuncture group and control group.

	Response N (%)	No response N (%)	Effect size	*P*-value
Acupuncture group (N = 55)	26(47.27%)	29(52.73%)	8.696	0.003
Control group (N = 24)	3(12.5%)	21(87.5%)		

TABLE 4 Comparison of HAMD reduction rate between acupuncture group and control group.

	HAMD reduction rate (%, median (Q1, Q3)*)	Effect size	*P*-value
Acupuncture (N = 55)	44.44 (25.00, 64.71)	1126.50	<0.001
Control group (N = 24)	9.19 (-7.55, 20.00)		

*Q1, upper quartile; Q3, lower quartile.

3.2 Characteristics of the gut microbiota between responders and non-responders before treatment

3.2.1 Baseline clinical characteristics between responders and non-responders

There were no significant differences in age, BMI, number of days postpartum, number of parturitions, length of disease, delivery mode, family history, and EPDS or HAMD-17 scores between responders and non-responders (Table 6).

3.2.2 Sequencing characteristics

A total of 55 samples from all recruited subjects were sequenced on an Illumina MiSeq sequencer. For downstream analysis, 2259092 qualified reads from 2373462 raw reads were filtered.

3.2.3 Gut microbial diversity changes in acupuncture responders and non-responders

We next used different diversity indices (evenness, Faith PD, observed species, Shannon diversity) to assess gut microbial α-diversity. There were no significant differences in diversity between acupuncture responders and non-responders ($P = 0.7856$, $P = 0.4276$, $P = 0.6679$, and $P = 0.7208$, respectively). However, the gut microbial diversity, as estimated by evenness, Faith PD, observed species, and Shannon diversity, tended to be higher in responders than non-responders (Figure 2).

To better understand differences in overall community composition between the samples, we calculated Bray-Curtis distances and unweighted UniFrac distances, which were both higher in non-responders than responders ($P = 0.0065$ and $P = 5.5e^{-05}$) and between groups ($P = 0.019$ and $P = 0.0005$) (Figures 3A, C), To further demonstrate differences in species diversity between samples, we applied the Principal Coordination Analysis and non-metric multidimensional scaling (Figures 3B, D). The gut microbial composition was similar between groups, with a

TABLE 5 Comparison of HAMD score before and after treatment in two groups.

Groups	Before treatment	After treatment	Effect size	*P*-value
Acupuncture (N = 55)	14.00± 3.51	7.76 ± 3.83	9.699	<0.001
Control group (N = 24)	13.33 ± 4.80	12.38 ± 4.10	1.647	0.113

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TABLE 6 Clinical characteristics of acupuncture responders and non-responders.

Characteristic	Responders (N = 26)	Non-responders (N = 29)	*P*-value
Age (year, mean ± SD)	34.85 ± 3.96	35.79 ± 4.07	0.386
BMI (kg.m^{-2}, mean ± SD)	22.87 ± 3.81	22.27 ± 2.83	0.510
Number of days postpartum (day, mean ± SD)	129.19 ± 91.29	153.93 ± 110.44	0.373
Number of parturitions (n)	1.54 ± 0.58	1.52 ± 0.63	0.898
Duration of disease (months)	3.19 ± 2. 12	4.31 ± 3.31	0.138
Delivery mode (A/B/C)*	13/12/1	12/17/0	0.418
Family history (y/n)	1/25	3/26	0.354
EPDS	15.65 ± 3.73	15.69 ± 5.28	0.977
HAMDS	14.42 ± 3.22	13.62 ± 3.77	0.403

*A, Natural childbirth; B, Cesarean section; C, The Women miscarried who are but were pregnant for more than seven months.

tendency to being more centralized in the responder group than in the non-responder group, although this was not statistically significant (P = 0.959 and P = 0.911).

3.2.4 Comparison of gut microbiota composition in acupuncture responders and non-responders

At the phylum level, *Firmicutes, Actinobacteria, Bacteroidetes*, and *Proteobacteria* were the most abundant organisms in the gut microbiota (Figure 4A). The genera of *Faecalibacterium, Blautia, Ruminococcaceae, Roseburia, Gemmiger, Megamonas* and *Bifidobacterium* were dominant in the two groups. The 5 genera *Faecalibacterium, Ruminococcaceae, Roseburia, Megamonas* and *Bifidobacterium* had higher abundance in the PPD group (9.99, 6.45, 6.09, 4.04, and 3.96%, respectively) as compared to those in the control group (9.42, 4.08, 6.06, 3.87, and 3.28%, respectively). The 2 genera *Blautia* and *Gemmiger* had lower abundance in the PPD group (9.51 and 4.16%, Gemmiger) as compared to those in the control group (12.37 and 5.41%, respectively). However, the results didn't reach significance (all *p*-values were more than 0.05) (Figure 4B).

3.2.5 Specific genera associated with acupuncture treatment responses

LEfSe analysis (p <0.05, LDA > 2) was used to identify specific bacteria associated with acupuncture treatment responses. *g_Desulfovibrio*, *g_Paraprevotella*, and *Paraprevotella_xylaniphila* were enriched in the responder group (Figure 5).

Having identified these three genera (biomarkers), we performed ROC curve analysis to evaluate their predictive accuracy. The area under the curve (AUC) was 0.76 and 0.66 for *g_Paraprevotella* and *g_Desulfovibrio*, respectively. The AUC of combining genera *g_Paraprevotella* and *g_Desulfovibrio* was 0.65 (Figure 6).

4 Discussion

This trial showed that acupuncture alleviated depressive symptoms in patients with PPD over an 8-week treatment period, and 47.27% patients significantly responded to acupuncture treatment. Additionally, based on gut microbiota profiling, we successfully predicted responses to acupuncture and improvements in clinical symptoms in PPD patients after treatment. *Desulfovibrio* and *Paraprevotella* were identified as specific predictive genera.

So far, there have been few reports on the predictors of therapeutic effect of PPD. Pinna speculated the neurosteroid biosynthesis and endogenous cannabinoid system might be able to predict antidepressive treatment, but lack of rigorous experimental studies to confirm this idea (Pinna, 2023). Additionally, there are also some limited explanations, such as the influence of running on cortisol, which can also affect the therapeutic effect of PPD in the later stage (Gobinath et al., 2018). These prediction methods are often incomplete and indirect. Therefore, gut microbiota as a predictor is seem to be more objective.

In the present study, our results further confirm the close relationship between the gut microbiota and mental health disorders. It has long been known that the enterotype distribution varies according to depression status, with *Bacteroides enteritidis* type 2 more prevalent in depressed patients than healthy controls (Valles-Colomer et al., 2019). It has also been shown that intestinal

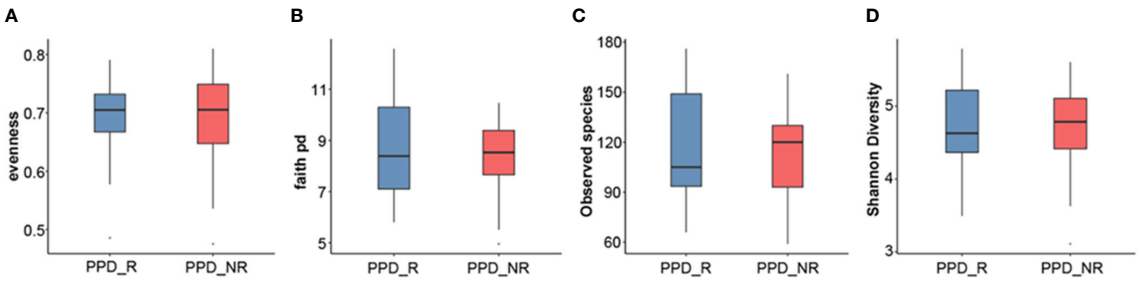

FIGURE 2
Microbial α-diversity analyses. Evenness index **(A)**, Faith PD **(B)**, observed species **(C)**, Shannon diversity **(D)**. PPD_R, PPD patients who are responsive to acupuncture treatment; PPD_NR, PPD patients who are not responsive to acupuncture treatment.

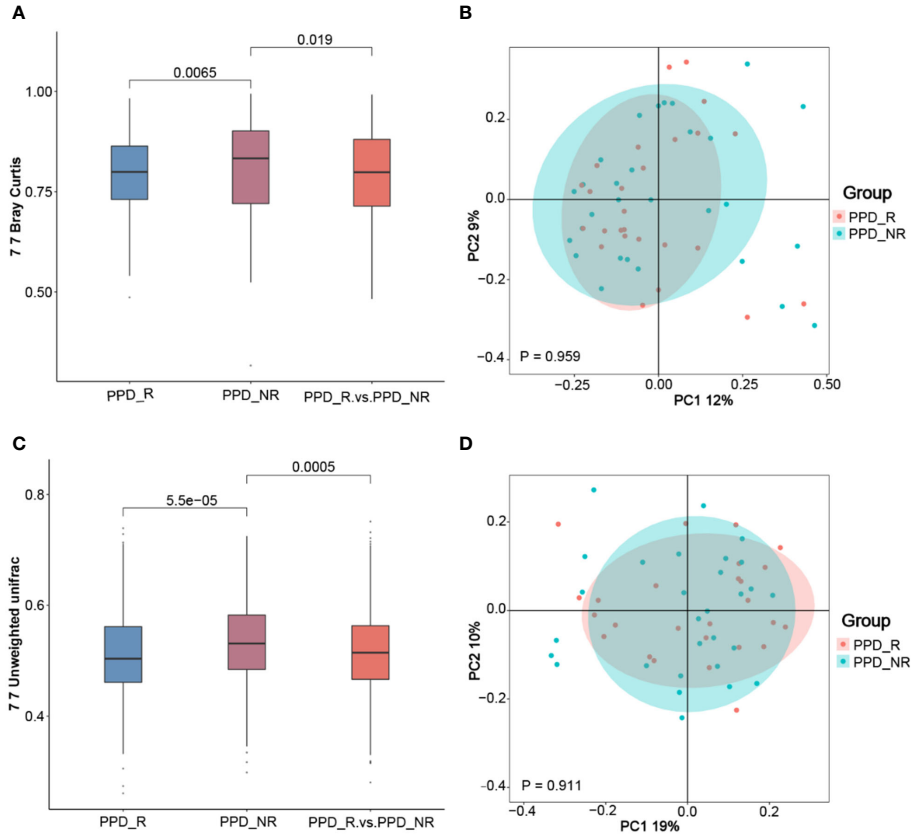

FIGURE 3
Microbial β-diversity analyses. The Wilcoxon rank-sum test analysis **(A)** and principal coordinates analysis plots **(B)** of the fecal microbiomes based on the Bray-Curtis distance; Wilcoxon rank-sum test analysis **(C)** and principal coordinates analysis plots **(D)** based on the Unweighted-UniFrac distance metric. PPD_R, PPD patients who are responsive to acupuncture treatment; PPD_NR, PPD patients who are not responsive to acupuncture treatment.

microbiota disorder is a characteristic of major depressive disorder (MDD) patients (Zheng et al., 2016; Zheng et al., 2020). Duan et al. studied treatment responses to escitalopram in a CUMS mouse depression model, comparing changes in metabolic function before and after treatment, and found that treatment responses were related to microbial composition, providing new insights into the mechanisms underlying variable antidepressant efficacy (Duan et al., 2021). Therefore, the flora structure is closely related to the intrinsic pathobiology of depression, suggesting that intestinal microbial biomarkers may be good predictors of antidepressant treatment responses.

Most studies employ a multifaceted approach to characterizing the gut microbiota, usually including measures of both α- and β-diversity. α-diversity is commonly used as a surrogate of

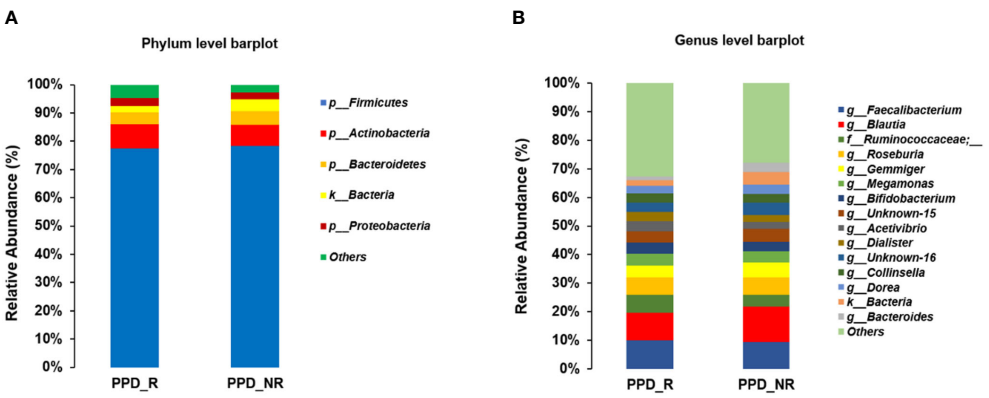

FIGURE 4
Microbiome composition differences at the phylum **(A)** and genus **(B)** levels between the two groups. PPD_R, PPD patients who are responsive to acupuncture treatment; PPD_NR, PPD patients who are not responsive to acupuncture treatment.

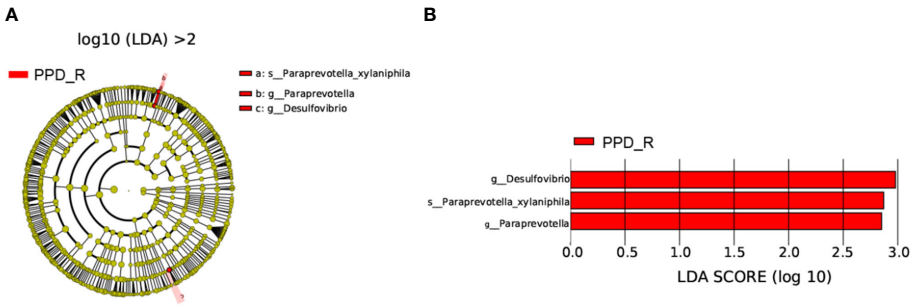

FIGURE 5
Differences in bacterial taxa between acupuncture responders and non-responders. Cladogram showing the most differentially abundant taxa identified by LEfSe. Red indicates clades enriched in the responder group **(A)**. Comparisons of gut microbiota between acupuncture responders and non-responders **(B)**. Only genera meeting a linear discriminant analysis score threshold >2 are shown. PPD_R, PPD patients who are responsive to acupuncture treatment; PPD_NR, PPD patients who are not responsive to acupuncture treatment.

community stability and function, which are thought to be beneficial to the host (Shade, 2017). Jiang et al. found that the intestinal microflora α-diversity was higher in antidepressant drug non-responders than responders in MDD patients compared with healthy controls (Jiang et al., 2015). In addition, the α-diversity of the gut microbiota was not significantly different in MDD patients with different treatment responses (Dong et al., 2022). In our study, we found that there were no significant differences in diversity between PPD patients who did and did not response to acupuncture, nor were there differences in the abundance and uniformity of the gut microbiota between the two groups. This mirrors the inconsistent results of previous studies, and the specific reasons underlying these differences need further study.

β-diversity reflects relationships between samples by analyzing the species composition and abundance (Anderson et al., 2011). Our β-diversity analysis showed that responders were significantly separated from non-responders, and the responder group had a more similar species composition. Kelly et al. and Zheng et al. both reported significant differences in β-diversity between individuals with depression and healthy controls (Kelly et al., 2016; Zheng et al., 2016). In the CUMS-induced depression study in mice, the β-diversity was also different between non-responders and responders (Duan et al., 2021), as was the β-diversity in patients who did and did not benefit from anti-programmed cell death protein 1 (PD-1) immunotherapy (Mao et al., 2021). The latter study found that the intestinal microflora affected the spectrum of immunotherapy-related adverse events, with high species diversity and relative abundance perhaps protective against immunotherapy-related adverse events (Mao et al., 2021).

We further analyzed and identified specific genera associated with acupuncture treatment responses. At the genus level, *Desulfovibrio* and *Paraperevotella* were enriched in responders, consistent with previous studies reporting a high abundance of *Paraprevotella* and *Desulfovibrio* at the genus level in patients with

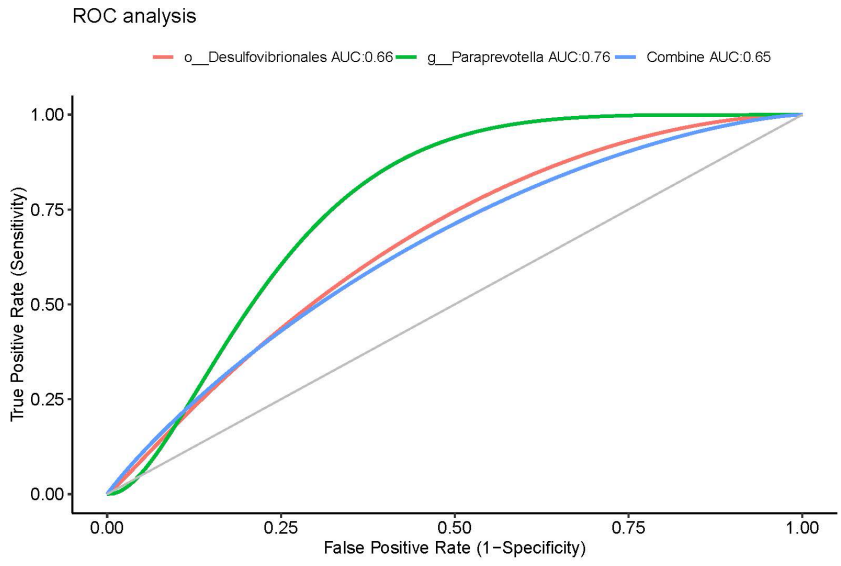

FIGURE 6
ROC curves using fecal microbiota to distinguish responders from non-responders. AUC, area under the curve; ROC, receiver operating characteristics.

depression (Naseribafrouei et al., 2014; Chen et al., 2018a; Chen et al., 2018b). *Desulfovibrio* are present in the oral and intestinal tract of approximately 50% of people, where they release hydrogen sulfide as a product of sulfate reduction (Devereux et al., 1990). Hydrogen sulfide is involved in the natural prevention of many digestive tract diseases (Pires et al., 2006), and there is a well-established link between desulfurization bacteria and individual intestinal diseases (Verstreken et al., 2012). For example, Scanlan et al. found significantly more desulfurization bacteria in the feces of colon cancer patients than healthy people (Scanlan et al., 2009), and similarly Rowan et al. found a significantly higher relative abundance of desulfurization bacteria in the intestinal tracts of patients with ulcerative colitis than those of healthy controls (Rowan et al., 2010). Additionally, we discovered that *Paraprevotella* was enriched in the gut microbiota of patients responding well to acupuncture. *Paraprevotella* belongs to the *Prevotellaceae* family, and another family member *Prevotella* is associated with a healthy plant-based diet and probiotic use (Ley, 2016). *Prevotella* can also act as an opportunistic pathogen associated with periodontal and dental inflammation, intestinal inflammation, rheumatoid arthritis, and bacterial vaginitis (Arweiler and Netuschil, 2016; Randis and Ratner, 2019; Bertelsen et al., 2021; Jia et al., 2021).

Desulfovibrio and *Paraperevotella* have different potential pathogenic mechanisms. For example, *Desulfovibrio* organisms co-cultured with human oral epidermoid carcinoma (KB) cells increased interleukin (IL)-6 production, implicating them in immune responses (Bisson-Boutelliez et al., 2010). Colonization of the intestine with *Prevotella* leads to metabolic changes in the microbiota that reduce IL-18 production (Iljazovic et al., 2021), thus aggravating intestinal inflammation and possibly leading to systemic autoimmunity. Furthermore, *Prevotella* can damage intestinal mucosal barrier function by producing sulfatase, which induces and degrades mucus, thus helping itself and other harmful bacteria to access intestinal epithelial cells to generate local inflammation (Wright et al., 2000). In addition, these two genera as Gram-negative bacteria might help to explain the role of microbiota in the development/maintenance of depression. Gram-negative bacteria contain lipopolysaccharides in the outer cell membrane leaflet (Al Bander et al., 2020), and lipopolysaccharides interacts with macrophages and stimulates immune responses through pro-inflammatory cytokine release. Supporting this, increased levels of proinflammatory cytokines including IL-1β and IL-6 and decreased levels of anti-inflammatory cytokines including IL-4 and IL-10 have been detected in people living with depression (Berk et al., 2013; Wong et al., 2016).

An increasing number of studies show that the occurrence and development of depression are closely related to inflammation and immunity (Simmons and Broderick, 2005; Maes et al., 2012; Kelly et al., 2015), Inflammatory cytokines and kynurenine pathway have been found as potential therapeutic targets for PPD, because the increase of plasma IL-6 and IL-8 and the decrease of serotonin, IL-2 and quinolinic acid are related to the severity of depressive symptoms, which increases the risk of PPD (Achtyes et al., 2020). These results indicate that the increased level of some inflammatory biomarkers in PPD patients means that the disease is related to the impaired adaptability of the immune system (Bränn et al., 2020).

Therefore, the high expression of these genera in PPD patients may correspond to increased levels of inflammatory biomarkers, and several studies have shown a strong association between persistent inflammatory responses and antidepressant therapy resistance (Carvalho et al., 2013). Electroacupuncture can downregulate inflammatory factors such as IL-6 in the hippocampus of depressed rats, suggesting that electroacupuncture may relieve depression through immune regulation (Guo et al., 2014; Yue et al., 2018). Indeed, α7nAChR is activated by acetylcholine released from cholinergic nerve endings and is a key target for inhibiting pro-inflammatory cytokines release by macrophages (Stakenborg et al., 2017). Acupuncture can reduce inflammatory cytokine production through the vagus nerve by activating α7nAChR (Yang et al., 2021). Acupuncture can also regulate the interaction between the gut microbiota and the brain-gut axis, inhibit proinflammatory cytokine production, alter the number and proportion of the gut microbiota, restore its stability, improve intestinal barrier function, and further adjust body function (Jang et al., 2020; Wang et al., 2020). In acupuncture treatment of PPD, the unique regulation mechanism of immune intestinal flora also played an important role. Finally, we found that acupuncture might inhibit inflammation and improve depression via two pathways: (1) inhibiting the release of inflammatory cytokines by activating the vagus nerve; and (2) regulating the brain-gut axis through the intestinal microflora, some predecessors put forward the same argument previously. (Yang et al., 2022).Therefore, accumulation of *Desulfovibrio* and *Paraperevotella* in the intestinal tracts of responsive patients could mediate the immune response induced by acupuncture to better regulate and alleviate depressive symptoms. The conclusion of our research results accords with the above conclusion, which can be understood as that acupuncture has played a better and more sensitive role in the flora of the responders.

5 Limitations

The study has several limitations. The sample size of present study was relatively small, and further studies in larger sample sizes are needed to confirm the findings with more advanced analyses methods, such as machine learning methods.

6 Conclusion

In conclusion, *Paraprevotella* and *Desulfovibrio* predicted early responses to antidepressants in patients with PPD receiving acupuncture. These results may help clinicians optimize their management of individual PPD patients in the future. Baseline enrichment and metabolism of *Paraprevotella* and *Desulfovibrio* intestinal microbiota in PPD patients were related to treatment outcomes. These findings pave the way for a new approach to personalize and maximize the efficacy of acupuncture treatment in PPD patients and provide potential new and accurate biomarkers for managing PPD patients.

Data availability statement

The original contributions presented in the study are publicly available. This data can be found here: https://www.ncbi.nlm.nih.gov/, with accession number PRJNA976190.

Ethics statement

The studies involving humans were approved by Ethics Committee of Shenzhen Traditional Chinese Medicine Hospital. The studies were conducted in accordance with the local legislation and institutional requirements. The participants provided their written informed consent to participate in this study.

Author contributions

Y-MZ and J-JY contributed equally. M-P and Z-XY are the corresponding authors. Y-MZ conceived and planned the experiments. Y-MZ and J-JY wrote the manuscript. Y-QX, X-MM, YYXZ, Y-HG, CC and X-XH executed the experiments. M-P and Z-XY contributed to revise the final manuscript. All authors contributed to the article and approved the submitted version.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

The reviewer SL declared a shared parent affiliation with the authors Y-MZ, J-JY, Y-QX, Y-HG, YYXZ, X-XH, X-MM, M-P, Z-XY to the handling editor at the time of the review.

Publisher's note

References

Achtyes, E., Keaton, S. A., Smart, L., Burmeister, A. R., Heilman, P. L., Krzyzanowski, S., et al. (2020). Inflammation and kynurenine pathway dysregulation in post-partum women with severe and suicidal depression. *Brain Behav. Immun.* 83, 239–247. doi: 10.1016/j.bbi.2019.10.017

Al Bander, Z., Nitert, M. D., Mousa, A., and Naderpoor, N. (2020). The gut microbiota and inflammation: an overview. *Int. J. Environ. Res. Public Health* 17 (20), 7618. doi: 10.3390/ijerph17207618

Alli, S., Gorbovskaya, I., Liu, J., Kolla, N., Brown, L., and Müller, D. (2022). The gut microbiome in depression and potential benefit of prebiotics, probiotics and synbiotics: A systematic review of clinical trials and observational studies. *Int. J. Mol. Sci.* 23 (9), 4494. doi: 10.3390/ijms23094494

Anderson, M. J., Crist, T. O., Chase, J. M., Vellend, M., Inouye, B. D., Freestone, A. L., et al. (2011). Navigating the multiple meanings of β diversity: a roadmap for the practicing ecologist. *Ecol. Lett.* 14 (1), 19–28. doi: 10.1111/j.1461-0248.2010.01552.x

Appleby, L., Warner, R., Whitton, A., and Faragher, B. (1997). A controlled study of fluoxetine and cognitive-behavioural counselling in the treatment of postnatal depression. *Bmj* 314 (7085), 932–936. doi: 10.1136/bmj.314.7085.932

Arweiler, N. B., and Netuschil, L. (2016). The oral microbiota. *Adv. Exp. Med. Biol.* 902, 45–60. doi: 10.1007/978-3-319-31248-4_4

Battle, D. (2013). Diagnostic and statistical manual of mental disorders (DSM). *CoDAS* 25 (2), 191–192. doi: 10.1590/s2317-17822013000200017

Berk, M., Williams, L. J., Jacka, F. N., O'Neil, A., Pasco, J. A., Moylan, S., et al. (2013). So depression is an inflammatory disease, but where does the inflammation come from? *BMC Med.* 11, 200. doi: 10.1186/1741-7015-11-200

Bertelsen, A., Elborn, J. S., and Schock, B. C. (2021). Microbial interaction: Prevotella spp. reduce P. aeruginosa induced inflammation in cystic fibrosis bronchial epithelial cells. *J. Cyst Fibros* 20 (4), 682–691. doi: 10.1016/j.jcf.2021.04.012

Bisson-Boutelliez, C., Massin, F., Dumas, D., Miller, N., and Lozniewski, A. (2010). Desulfovibrio spp. survive within KB cells and modulate inflammatory responses. *Mol. Oral. Microbiol.* 25 (3), 226–235. doi: 10.1111/j.2041-1014.2009.00550.x

Bränn, E., Fransson, E., White, R. A., Papadopoulos, F. C., Edvinsson, Å., Kamali-Moghaddam, M., et al. (2020). Inflammatory markers in women with postpartum depressive symptoms. *J. Neurosci. Res.* 98 (7), 1309–1321. doi: 10.1002/jnr.24312

Brown, J. V. E., Wilson, C. A., Ayre, K., Robertson, L., South, E., Molyneaux, E., et al. (2021). Antidepressant treatment for postnatal depression. *Cochrane Database Syst. Rev.* 2 (2), Cd013560. doi: 10.1002/14651858.CD013560.pub2

Carvalho, A. F., Sharma, M. S., Brunoni, A. R., Vieta, E., and Fava, G. A. (2016). The safety, tolerability and risks associated with the use of newer generation antidepressant drugs: A critical review of the literature. *Psychother. Psychosom* 85 (5), 270–288. doi: 10.1159/000447034

Carvalho, L., Torre, J., Papadopoulos, A., Poon, L., Juruena, M., Markopoulou, K., et al. (2013). Lack of clinical therapeutic benefit of antidepressants is associated overall activation of the inflammatory system. *J. Affect. Disord.* 148 (1), 136–140. doi: 10.1016/j.jad.2012.10.036

Chen, Z., Li, J., Gui, S., Zhou, C., Chen, J., Yang, C., et al. (2018b). Comparative metaproteomics analysis shows altered fecal microbiota signatures in patients with major depressive disorder. *Neuroreport* 29 (5), 417–425. doi: 10.1097/wnr.0000000000000985

Chen, J. J., Zheng, P., Liu, Y. Y., Zhong, X. G., Wang, H. Y., Guo, Y. J., et al. (2018a). Sex differences in gut microbiota in patients with major depressive disorder. *Neuropsychiatr. Dis. Treat* 14, 647–655. doi: 10.2147/ndt.S159322

Chung, Y. E., Chen, H. C., Chou, H. L., Chen, I. M., Lee, M. S., Chuang, L. C., et al. (2019). Exploration of microbiota targets for major depressive disorder and mood related traits. *J. Psychiatr. Res.* 111, 74–82. doi: 10.1016/j.jpsychires.2019.01.016

Cohen, L. S., Viguera, A. C., Bouffard, S. M., Nonacs, R. M., Morabito, C., Collins, M. H., et al. (2001). Venlafaxine in the treatment of postpartum depression. *J. Clin. Psychiatry* 62 (8), 592–596. doi: 10.4088/jcp.v62n0803

Consortium, P.D.A.T.C.a.T.P (2015). Heterogeneity of postpartum depression: a latent class analysis. *Lancet Psychiatry* 2 (1), 59–67. doi: 10.1016/s2215-0366(14)00055-8

Davanzo, R., Copertino, M., De Cunto, A., Minen, F., and Amaddeo, A. (2011). Antidepressant drugs and breastfeeding: a review of the literature. *Breastfeed Med.* 6 (2), 89–98. doi: 10.1089/bfm.2010.0019

Devereux, R., He, S. H., Doyle, C. L., Orkland, S., Stahl, D. A., LeGall, J., et al. (1990). Diversity and origin of Desulfovibrio species: phylogenetic definition of a family. *J. Bacteriol* 172 (7), 3609–3619. doi: 10.1128/jb.172.7.3609-3619.1990

Dong, Z., Shen, X., Hao, Y., Li, J., Li, H., Xu, H., et al. (2021). Gut microbiome: A potential indicator for differential diagnosis of major depressive disorder and general anxiety disorder. *Front. Psychiatry* 12. doi: 10.3389/fpsyt.2021.651536

Dong, Z., Shen, X., Hao, Y., Li, J., Xu, H., Yin, L., et al. (2022). Gut microbiome: A potential indicator for predicting treatment outcomes in major depressive disorder. *Front. Neurosci.* 16. doi: 10.3389/fnins.2022.813075

Duan, J., Huang, Y., Tan, X., Chai, T., Wu, J., Zhang, H., et al. (2021). Characterization of gut microbiome in mice model of depression with divergent response to escitalopram treatment. *Transl. Psychiatry* 11 (1), 303. doi: 10.1038/s41398-021-01428-1

Dubois, T., Reynaert, C., Jacques, D., Lepiece, B., and Zdanowicz, N. (2019). Role of gut microbiota in the interaction between immunity and psychiatry: a literature review. *Psychiatr. Danub* 31 (Suppl 3), 381–385. Available at: https://pubmed.ncbi.nlm.nih.gov/31488756/

First, M. B. (2013). Diagnostic and statistical manual of mental disorders, 5th edition, and clinical utility. *J. Nerv Ment. Dis.* 201 (9), 727–729. doi: 10.1097/NMD.0b013e3182a2168a

Foster, J. A., and McVey Neufeld, K. A. (2013). Gut-brain axis: how the microbiome influences anxiety and depression. *Trends Neurosci.* 36 (5), 305–312. doi: 10.1016/j.tins.2013.01.005

Gobinath, A. R., Richardson, R. J., Chow, C., Workman, J. L., Lieblich, S. E., Barr, A. M., et al. (2018). Voluntary running influences the efficacy of fluoxetine in a model of postpartum depression. *Neuropharmacology* 128, 106–118. doi: 10.1016/j.neuropharm.2017.09.017

Grigoriadis, S., Wilton, A. S., Kurdyak, P. A., Rhodes, A. E., VonderPorten, E. H., Levitt, A., et al. (2017). Perinatal suicide in Ontario, Canada: a 15-year population-based study. *Cmaj* 189 (34), E1085–e1092. doi: 10.1503/cmaj.170088

Guo, T., Guo, Z., Yang, X., Sun, L., Wang, S., Yingge, A., et al. (2014). The alterations of IL-1Beta, IL-6, and TGF-beta levels in hippocampal CA3 region of chronic restraint stress rats after electroacupuncture (EA) pretreatment. *Evid Based Complement Alternat Med.* 2014, 369158. doi: 10.1155/2014/369158

Hamilton, M. (1960). A rating scale for depression. *J. Neurol. Neurosurg. Psychiatry* 23 (1), 56–62. doi: 10.1136/jnnp.23.1.56

Hantsoo, L., Ward-O'Brien, D., Czarkowski, K. A., Gueorguieva, R., Price, L. H., and Epperson, C. N. (2014). A randomized, placebo-controlled, double-blind trial of sertraline for postpartum depression. *Psychopharmacol. (Berl)* 231 (5), 939–948. doi: 10.1007/s00213-013-3316-1

Hou, Z., Wang, Z., Jiang, W., Yin, Y., Yue, Y., Zhang, Y., et al. (2016). Divergent topological architecture of the default mode network as a pretreatment predictor of early antidepressant response in major depressive disorder. *Sci. Rep.* 6, 39243. doi: 10.1038/srep39243

Howard, L. M., Molyneaux, E., Dennis, C. L., Rochat, T., Stein, A., and Milgrom, J. (2014). Non-psychotic mental disorders in the perinatal period. *Lancet* 384 (9956), 1775–1788. doi: 10.1016/s0140-6736(14)61276-9

Huang, R., Yang, D., Lei, B., Yan, C., Tian, Y., Huang, X., et al. (2020). The short- and long-term effectiveness of mother-infant psychotherapy on postpartum depression: A systematic review and meta-analysis. *J. Affect. Disord.* 260, 670–679. doi: 10.1016/j.jad.2019.09.056

Hunter, A. M., Muthén, B. O., Cook, I. A., and Leuchter, A. F. (2010). Antidepressant response trajectories and quantitative electroencephalography (QEEG) biomarkers in major depressive disorder. *J. Psychiatr. Res.* 44 (2), 90–98. doi: 10.1016/j.jpsychires.2009.06.006

Iljazovic, A., Roy, U., Gálvez, E. J. C., Lesker, T. R., Zhao, B., Gronow, A., et al. (2021). Perturbation of the gut microbiome by Prevotella spp. enhances host susceptibility to mucosal inflammation. *Mucosal Immunol.* 14 (1), 113–124. doi: 10.1038/s41385-020-0296-4

Jang, J. H., Yeom, M. J., Ahn, S., Oh, J. Y., Ji, S., Kim, T. H., et al. (2020). Acupuncture inhibits neuroinflammation and gut microbial dysbiosis in a mouse model of Parkinson's disease. *Brain Behav. Immun.* 89, 641–655. doi: 10.1016/j.bbi.2020.08.015

Jia, Y. J., Liao, Y., He, Y. Q., Zheng, M. Q., Tong, X. T., Xue, W. Q., et al. (2021). Association between oral microbiota and cigarette smoking in the chinese population. *Front. Cell Infect. Microbiol.* 11. doi: 10.3389/fcimb.2021.658203

Jiang, H., Deng, S., Zhang, J., Chen, J., Li, B., Zhu, W., et al. (2023). Acupuncture treatment for post-stroke depression: Intestinal microbiota and its role. *Front. Neurosci.* 17. doi: 10.3389/fnins.2023.1146946

Jiang, H., Ling, Z., Zhang, Y., Mao, H., Ma, Z., Yin, Y., et al. (2015). Altered fecal microbiota composition in patients with major depressive disorder. *Brain behavior Immun.* 48, 186–194. doi: 10.1016/j.bbi.2015.03.016

Keller, M. (2003). Past, present, and future directions for defining optimal treatment outcome in depression: remission and beyond. *JAMA* 289 (23), 3152–3160. doi: 10.1001/jama.289.23.3152

Kelly, J. R., Borre, Y., C, O. B., Patterson, E., El Aidy, S., Deane, J., et al. (2016). Transferring the blues: Depression-associated gut microbiota induces neurobehavioural changes in the rat. *J. Psychiatr. Res.* 82, 109–118. doi: 10.1016/j.jpsychires.2016.07.019

Kelly, J. R., Kennedy, P. J., Cryan, J. F., Dinan, T. G., Clarke, G., and Hyland, N. P. (2015). Breaking down the barriers: the gut microbiome, intestinal permeability and stress-related psychiatric disorders. *Front. Cell Neurosci.* 9. doi: 10.3389/fncel.2015.00392

Ley, R. E. (2016). Gut microbiota in 2015: Prevotella in the gut: choose carefully. *Nat. Rev. Gastroenterol. Hepatol.* 13 (2), 69–70. doi: 10.1038/nrgastro.2016.4

Li, M., Niu, J., Yan, P., Yao, L., He, W., Wang, M., et al. (2020). The effectiveness and safety of acupuncture for depression: An overview of meta-analyses. *Complement Ther. Med.* 50, 102202. doi: 10.1016/j.ctim.2019.102202

Li, P., Huang, W., Yan, Y. N., Cheng, W., Liu, S., Huang, Y., et al. (2021). Acupuncture can play an antidepressant role by regulating the intestinal microbes and neurotransmitters in a rat model of depression. *Med. Sci. Monit* 27, e929027. doi: 10.12659/msm.929027

Li, S., Zhong, W., Peng, W., and Jiang, G. (2018). Effectiveness of acupuncture in postpartum depression: a systematic review and meta-analysis. *Acupunct Med.* 36 (5), 295–301. doi: 10.1136/acupmed-2017-011530

Li, W., Yin, P., Lao, L., and Xu, S. (2019). Effectiveness of acupuncture used for the management of postpartum depression: A systematic review and meta-analysis. *BioMed. Res. Int.* 2019, 6597503. doi: 10.1155/2019/6597503

Liang, J. Q., Li, T., Nakatsu, G., Chen, Y. X., Yau, T. O., Chu, E., et al. (2020). A novel faecal Lachnoclostridium marker for the non-invasive diagnosis of colorectal adenoma and cancer. *Gut* 69 (7), 1248–1257. doi: 10.1136/gutjnl-2019-318532

Limeta, A., Ji, B., Levin, M., Gatto, F., and Nielsen, J. (2020). Meta-analysis of the gut microbiota in predicting response to cancer immunotherapy in metastatic melanoma. *JCI Insight* 5 (23), e140940. doi: 10.1172/jci.insight.140940

Ma, Q., Li, Y., Li, P., Wang, M., Wang, J., Tang, Z., et al. (2019). Research progress in the relationship between type 2 diabetes mellitus and intestinal flora. *Biomedicine pharmacotherapy = Biomedecine pharmacotherapie* 117, 109138. doi: 10.1016/j.biopha.2019.109138

Maes, M., Kubera, M., Leunis, J. C., and Berk, M. (2012). Increased IgA and IgM responses against gut commensals in chronic depression: further evidence for increased bacterial translocation or leaky gut. *J. Affect. Disord.* 141 (1), 55–62. doi: 10.1016/j.jad.2012.02.023

Mao, J., Wang, D., Long, J., Yang, X., Lin, J., Song, Y., et al. (2021). Gut microbiome is associated with the clinical response to anti-PD-1 based immunotherapy in hepatobiliary cancers. *J. immunotherapy Cancer* 9 (12), e003334. doi: 10.1136/jitc-2021-003334

McGuinness, A. J., Davis, J. A., Dawson, S. L., Loughman, A., Collier, F., O'Hely, M., et al. (2022). A systematic review of gut microbiota composition in observational studies of major depressive disorder, bipolar disorder and schizophrenia. *Mol. Psychiatry* 27 (4), 1920–1935. doi: 10.1038/s41380-022-01456-3

Misri, S., Swift, E., Abizadeh, J., and Shankar, R. (2016). Overcoming functional impairment in postpartum depressed or anxious women: a pilot trial of desvenlafaxine with flexible dosing. *Ther. Adv. Psychopharmacol.* 6 (4), 269–276. doi: 10.1177/2045125316656297

Naseribafrouei, A., Hestad, K., Avershina, E., Sekelja, M., Linløkken, A., Wilson, R., et al. (2014). Correlation between the human fecal microbiota and depression. *Neurogastroenterol Motil.* 26 (8), 1155–1162. doi: 10.1111/nmo.12378

Netsi, E., Pearson, R. M., Murray, L., Cooper, P., Craske, M. G., and Stein, A. (2018). Association of persistent and severe postnatal depression with child outcomes. *JAMA Psychiatry* 75 (3), 247–253. doi: 10.1001/jamapsychiatry.2017.4363

Nikolova, V. L., Hall, M. R. B., Hall, L. J., Cleare, A. J., Stone, J. M., and Young, A. H. (2021). Perturbations in gut microbiota composition in psychiatric disorders: A review and meta-analysis. *JAMA Psychiatry* 78 (12), 1343–1354. doi: 10.1001/jamapsychiatry.2021.2573

Nonacs, R. M., Soares, C. N., Viguera, A. C., Pearson, K., Poitras, J. R., and Cohen, L. S. (2005). Bupropion SR for the treatment of postpartum depression: a pilot study. *Int. J. Neuropsychopharmacol.* 8 (3), 445–449. doi: 10.1017/s1461145705005079

Ormsby, S. M., Smith, C. A., Dahlen, H. G., Hay, P. J., and Lind, J. M. (2016). Evaluation of an antenatal acupuncture intervention as an adjunct therapy for antenatal depression (AcuAnteDep): study protocol for a pragmatic randomised controlled trial. *Trials* 17, 93. doi: 10.1186/s13063-016-1204-9

Pinna, G. (2023). Biomarkers and treatments for mood disorders encompassing the neurosteroid and endocannabinoid systems. *J. Neuroendocrinol* 35 (2), e13226. doi: 10.1111/jne.13226

Pires, R. H., Venceslau, S. S., Morais, F., Teixeira, M., Xavier, A. V., and Pereira, I. A. (2006). Characterization of the Desulfovibrio desulfuricans ATCC 27774 DsrMKJOP

complex–a membrane-bound redox complex involved in the sulfate respiratory pathway. *Biochemistry* 45 (1), 249–262. doi: 10.1021/bi0515265

Randis, T. M., and Ratner, A. J. (2019). Gardnerella and prevotella: co-conspirators in the pathogenesis of bacterial vaginosis. *J. Infect. Dis.* 220 (7), 1085–1088. doi: 10.1093/infdis/jiy705

Rieder, R., Wisniewski, P. J., Alderman, B. L., and Campbell, S. C. (2017). Microbes and mental health: A review. *Brain Behav. Immun.* 66, 9–17. doi: 10.1016/j.bbi.2017.01.016

Rowan, F., Docherty, N. G., Murphy, M., Murphy, B., Calvin Coffey, J., and O'Connell, P. R. (2010). Desulfovibrio bacterial species are increased in ulcerative colitis. *Dis. Colon Rectum* 53 (11), 1530–1536. doi: 10.1007/DCR.0b013e3181f1e620

Santos, H.Jr., Tan, X., and Salomon, R. (2017). Heterogeneity in perinatal depression: how far have we come? A systematic review. *Arch. Womens Ment. Health* 20 (1), 11–23. doi: 10.1007/s00737-016-0691-8

Scanlan, P. D., Shanahan, F., and Marchesi, J. R. (2009). Culture-independent analysis of desulfovibrios in the human distal colon of healthy, colorectal cancer and polypectomized individuals. *FEMS Microbiol. Ecol.* 69 (2), 213–221. doi: 10.1111/j.1574-6941.2009.00709.x

Schwarz, E., Maukonen, J., Hyytiäinen, T., Kieseppä, T., Orešič, M., Sabunciyan, S., et al. (2018). Analysis of microbiota in first episode psychosis identifies preliminary associations with symptom severity and treatment response. *Schizophr. Res.* 192, 398–403. doi: 10.1016/j.schres.2017.04.017

Segata, N., Izard, J., Waldron, L., Gevers, D., Miropolsky, L., Garrett, W. S., et al. (2011). Metagenomic biomarker discovery and explanation. *Genome Biol.* 12 (6), R60. doi: 10.1186/gb-2011-12-6-r60

Shade, A. (2017). Diversity is the question, not the answer. *Isme J.* 11 (1), 1–6. doi: 10.1038/ismej.2016.118

Sharma, V., Khan, M., Baczynski, C., and Boate, I. (2020). Predictors of response to antidepressants in women with postpartum depression: a systematic review. *Arch. Womens Ment. Health* 23 (5), 613–623. doi: 10.1007/s00737-020-01044-w

Sharp, D. J., Chew-Graham, C., Tylee, A., Lewis, G., Howard, L., Anderson, I., et al. (2010). A pragmatic randomised controlled trial to compare antidepressants with a community-based psychosocial intervention for the treatment of women with postnatal depression: the RESPOND trial. *Health Technol. Assess.* 14 (43), 1–153. doi: 10.3310/hta14430

Simmons, D. A., and Broderick, P. A. (2005). Cytokines, stressors, and clinical depression: augmented adaptation responses underlie depression pathogenesis. *Prog. Neuropsychopharmacol. Biol. Psychiatry* 29 (5), 793–807. doi: 10.1016/j.pnpbp.2005.03.009

Slykerman, R., Hood, F., Wickens, K., Thompson, J., Barthow, C., Murphy, R., et al. (2017). Effect of lactobacillus rhamnosus HN001 in pregnancy on postpartum symptoms of depression and anxiety: A randomised double-blind placebo-controlled trial. *EBioMedicine* 24, 159–165. doi: 10.1016/j.ebiom.2017.09.013

Stakenborg, N., Gomez-Pinilla, P. J., and Boeckxstaens, G. E. (2017). Postoperative ileus: pathophysiology, current therapeutic approaches. *Handb. Exp. Pharmacol.* 239, 39–57. doi: 10.1007/164_2016_108

Suri, R., Burt, V. K., and Altshuler, L. L. (2005). Nefazodone for the treatment of postpartum depression. *Arch. Womens Ment. Health* 8 (1), 55–56. doi: 10.1007/s00737-005-0071-2

Tian, X. Y., Xing, J. W., Zheng, Q. Q., and Gao, P. F. (2021). 919 syrup alleviates postpartum depression by modulating the structure and metabolism of gut microbes and affecting the function of the hippocampal GABA/glutamate system. *Front. Cell Infect. Microbiol.* 11. doi: 10.3389/fcimb.2021.694443

Tong, P., Dong, L. P., Yang, Y., Shi, Y. H., Sun, T., and Bo, P. (2019). Traditional Chinese acupuncture and postpartum depression: A systematic review and meta-analysis. *J. Chin. Med. Assoc.* 82 (9), 719–726. doi: 10.1097/jcma.0000000000000140

Valles-Colomer, M., Falony, G., Darzi, Y., Tigchelaar, E. F., Wang, J., Tito, R. Y., et al. (2019). The neuroactive potential of the human gut microbiota in quality of life and depression. *Nat. Microbiol.* 4 (4), 623–632. doi: 10.1038/s41564-018-0337-x

Verstreken, I., Laleman, W., Wauters, G., and Verhaegen, J. (2012). Desulfovibrio desulfuricans bacteremia in an immunocompromised host with a liver graft and ulcerative colitis. *J. Clin. Microbiol.* 50 (1), 199–201. doi: 10.1128/jcm.00987-11

Wang, L., An, J., Song, S., Mei, M., Li, W., Ding, F., et al. (2020). Electroacupuncture preserves intestinal barrier integrity through modulating the gut microbiota in DSS-induced chronic colitis. *Life Sci.* 261, 118473. doi: 10.1016/j.lfs.2020.118473

Wang, Z., Liu, J., Shuai, H., Cai, Z., Fu, X., Liu, Y., et al. (2021). Mapping global prevalence of depression among postpartum women. *Transl. Psychiatry* 11 (1), 543. doi: 10.1038/s41398-021-01663-6

Wong, M. L., Inserra, A., Lewis, M. D., Mastronardi, C. A., Leong, L., Choo, J., et al. (2016). Inflammasome signaling affects anxiety- and depressive-like behavior and gut microbiome composition. *Mol. Psychiatry* 21 (6), 797–805. doi: 10.1038/mp.2016.46

Wright, D. P., Rosendale, D. I., and Robertson, A. M. (2000). Prevotella enzymes involved in mucin oligosaccharide degradation and evidence for a small operon of genes expressed during growth on mucin. *FEMS Microbiol. Lett.* 190 (1), 73–79. doi: 10.1111/j.1574-6968.2000.tb09265.x

Yang, N. N., Lin, L. L., Li, Y. J., Li, H. P., Cao, Y., Tan, C. X., et al. (2022). Potential mechanisms and clinical effectiveness of acupuncture in depression. *Curr. Neuropharmacol* 20 (4), 738–750. doi: 10.2174/1570159x19666210609162809

Yang, N. N., Yang, J. W., Ye, Y., Huang, J., Wang, L., Wang, Y., et al. (2021). Electroacupuncture ameliorates intestinal inflammation by activating α7nAChR-mediated JAK2/STAT3 signaling pathway in postoperative ileus. *Theranostics* 11 (9), 4078–4089. doi: 10.7150/thno.52574

Yonkers, K. A., Lin, H., Howell, H. B., Heath, A. C., and Cohen, L. S. (2008). Pharmacologic treatment of postpartum women with new-onset major depressive disorder: a randomized controlled trial with paroxetine. *J. Clin. Psychiatry* 69 (4), 659–665. doi: 10.4088/jcp.v69n0420

Yue, N., Li, B., Yang, L., Han, Q. Q., Huang, H. J., Wang, Y. L., et al. (2018). Electro-acupuncture alleviates chronic unpredictable stress-induced depressive- and anxiety-like behavior and hippocampal neuroinflammation in rat model of depression. *Front. Mol. Neurosci.* 11. doi: 10.3389/fnmol.2018.00149

Zheng, P., Yang, J., Li, Y., Wu, J., Liang, W., Yin, B., et al. (2020). Gut microbial signatures can discriminate unipolar from bipolar depression. *Adv. Sci. (Weinh)* 7 (7), 1902862. doi: 10.1002/advs.201902862

Zheng, P., Zeng, B., Zhou, C., Liu, M., Fang, Z., Xu, X., et al. (2016). Gut microbiome remodeling induces depressive-like behaviors through a pathway mediated by the host's metabolism. *Mol. Psychiatry* 21 (6), 786–796. doi: 10.1038/mp.2016.44

Zheng, Y., Wang, T., Tu, X., Huang, Y., Zhang, H., Tan, D., et al. (2019). Gut microbiome affects the response to anti-PD-1 immunotherapy in patients with hepatocellular carcinoma. *J. Immunother. Cancer* 7 (1), 193. doi: 10.1186/s40425-019-0650-9

Zhou, Y., Chen, C., Yu, H., and Yang, Z. (2020). Fecal microbiota changes in patients with postpartum depressive disorder. *Front. Cell Infect. Microbiol.* 10. doi: 10.3389/fcimb.2020.567268

Zhou, Y., Yu, H., Guo, Y., Chen, C., Huang, X., Gou, Y., et al. (2019). Efficacy of acupuncture versus sham acupuncture for postpartum depression disorder: Study protocol for a randomized controlled trial. *Eur. J. Integr. Med.* 31, 100982. doi: 10.1016/j.eujim.2019.100982

Zhu, J., Cai, H., Yuan, Y., Yue, Y., Jiang, D., Chen, C., et al. (2018). Variance of the global signal as a pretreatment predictor of antidepressant treatment response in drug-naïve major depressive disorder. *Brain Imaging Behav.* 12 (6), 1768–1774. doi: 10.1007/s11682-018-9845-9

10.3389/fcimb.2023.1228940

Glossary

anti-PD-1	anti-programmed cell death protein 1
AUC	area under the curve
CUMS	chronic unpredictable mild stress
DSM-V	Fifth edition of the Diagnostic and Statistical Manual of Mental Disorders
EPDS	Edinburgh Postnatal Depression Scale
HAMD	Hamilton Depression Rating
HAMD-17	17-item Hamilton Depression Rating
IL	interleukin
LDA	linear discriminant analysis
LEFSe	linear discriminant analysis effect size
MDD	major depressive disorder
PPD	postpartum depressive disorder
PPD_R	postpartum depressive disorder patients who are responsive to acupuncture treatment
PPD_NR	postpartum depressive disorder patients who are not responsive to acupuncture treatment
ROC	receiver operating characteristic curve
rRNA	16S ribosomal RNA