General Psychiatry

Neural connectivity biotypes: predictors of clinical outcomes and improvement patterns of iTBS treatment in adolescents and young adults with depression

Weicheng Li,[1,2,3,4] Yanan Yin,[1,3,4] Zerui You,[1,2,3,4] Min Zhang,[1,3,4] Chengyu Wang,[1,3,4] Xiaofeng Lan,[1,3,4] Siming Mai,[1,3,4] Fan Zhang,[1,2,3,4] Zhibo Hu,[1,3,4] Guanxi Liu,[1,2,3,4] Xiaoyu Chen,[1,3,4] Haiyan Liu,[1,3,4] Zhanjie Luo,[1,3,4] Yexian Zeng,[1,3,4] Yiying Chen,[1,3,4] Yifang Chen,[1,3,4] Robin Shao,[5] Hanna Lu,[6] Roger S McIntyre,[7,8,9,10,11,12] Yanling Zhou ,[1,3,4] Yuping Ning[1,2,3,4]

To cite: Li W, Yin Y, You Z, *et al*. Neural connectivity biotypes: predictors of clinical outcomes and improvement patterns of iTBS treatment in adolescents and young adults with depression. *General Psychiatry* 2025;**38**:e101749. doi:10.1136/gpsych-2024-101749

► Additional supplemental material is published online only. To view, please visit the journal online (https://doi.org/10.1136/gpsych-2024-101749).

WL, YY and ZY contributed equally.

WL, YY and ZY are joint first authors.

Received 19 June 2024
Accepted 25 March 2025

For numbered affiliations see end of article.

Correspondence to
Dr Yanling Zhou;
zhouylivy@aliyun.com

Dr Yuping Ning;
ningjeny@126.com

ABSTRACT

Background The heterogeneity of depression limits the treatment outcomes of intermittent theta burst stimulation (iTBS) and hinders the identification of predictive factors. This study investigated functional network connectivity and predictors of iTBS treatment outcomes in adolescents and young adults with depression.

Aim This study aimed to identify default mode network (DMN)-based connectivity patterns associated with varying iTBS treatment outcomes in depression.

Methods Data from a randomised controlled trial of iTBS in depression (n=82) were analysed using a data-driven approach to classify homogeneous subgroups based on the DMN. Connectivity subgroups were compared on depressive symptoms and cognitive function at pretreatment and post-treatment. Furthermore, the predictive significance of baseline inflammatory cytokines on post-treatment outcomes was evaluated.

Results Two distinct subgroups were identified. Subgroup 1 exhibited high heterogeneity and greater centrality in the posterior cingulate cortex and retrosplenial cortex, while subgroup 2 showed more homogeneous connectivity patterns and greater centrality in the temporoparietal junction and posterior inferior parietal lobule. No main effect for subgroup, treatment or subgroup×treatment interaction was revealed in the improvement of depressive symptoms. A significant subgroup×treatment interaction related to symbol coding improvement was detected (F=5.22, p=0.026). Within subgroup 1, the active group showed significantly greater improvement in symbol coding compared with the sham group (t=2.30, p=0.028), while baseline levels of interleukin-6 and C-reactive protein emerged as significant indicators for predicting improvements in symbolic coding (R^2=0.35, RMSE (root-mean-square error)=5.72, p=0.013). Subgroup 2 showed no significant findings in terms of cognitive improvement or inflammatory cytokines predictions.

Conclusions Data-driven network analyses offer valuable insights into iTBS treatment outcomes in depression, providing clues for predicting cognitive improvements from an inflammatory perspective.

Trial registration number ChiCTR2100042346.

WHAT IS ALREADY KNOWN ON THIS TOPIC

⇒ Intermittent theta burst stimulation (iTBS) efficiently regulates cortical activity within a mere 3-min time frame, offering a time-effective and economical option while preserving effectiveness and minimising negative reactions. However, the mechanism of antidepressive effect and distinctiveness of iTBS remain indeterminate.

WHAT THIS STUDY ADDS

⇒ The response to iTBS therapy in adolescents and young adults with depression may exhibit variations in network patterns based on the default mode network. Pretreatment levels of inflammatory markers can serve as potential predictors for cognitive improvements within specific biotypes.

HOW THIS STUDY MIGHT AFFECT RESEARCH, PRACTICE OR POLICY

⇒ The study results hold the potential to furnish valuable insights regarding data-driven network analyses into iTBS treatment outcomes in depression, providing clues for predicting cognitive improvements from an inflammatory perspective.

INTRODUCTION

Episodes of major depressive disorder (MDD) often initiate during adolescence and early adulthood (age 18–25) and may persist as lifetime prevalent depression, contributing to compromised academic

progress and social functioning, along with an elevated susceptibility to suicide.[1 2] Repetitive transcranial magnetic stimulation (rTMS) is acclaimed for its safety, efficacy, minimal side effects, and non-invasiveness in treating depression.[3] Intermittent theta burst stimulation (iTBS), a newer variant of rTMS, efficiently regulates cortical activity in approximately 3 min, making it a more time-efficient and cost-effective option with significant clinical potential. While the antidepressant effects of iTBS have been extensively studied, the underlying mechanisms remain poorly understood, and a subset of patients does not respond to treatment. One potential reason for this is the neurobiological heterogeneity of depression, particularly in adolescent cases. Therefore, it is crucial to investigate the neurobiological heterogeneity of depression to clarify the mechanisms behind the antidepressant effects of iTBS and identify factors that influence treatment outcomes. This research aimed to optimise therapeutic results and enhance the precision of treatment delivery.

iTBS typically targets the dorsolateral prefrontal cortex, a critical brain region involved in mood regulation, where its activation is often associated with improved treatment outcomes.[4] It is currently believed that iTBS induces alterations in the functional connectivity between the dorsolateral prefrontal cortex and other key brain regions, such as the default mode network (DMN), which plays a pivotal role in its antidepressant effects. The DMN, comprising the medial prefrontal cortex, posterior cingulate cortex and areas of the posterior parietal cortex, is associated with spontaneous cognition, mind-wandering, self-reflection and envisioning future scenarios.[5] Current research indicates that adolescent depression is linked to DMN network abnormalities.[6] For example, studies have shown increased DMN connectivity in adolescents with MDD compared with healthy controls.[7] Conversely, other research has found decreased anterior DMN connectivity in adolescent depression.[8] Moreover, abnormalities in the DMN of adolescents with depression have been positively correlated with the symptom severity,[7] and, crucially, the DMN is closely associated with the efficacy of antidepressant treatments. For instance, reduced DMN connectivity has been observed in patients who do not respond to antidepressant medication.[9] Considering the pivotal role of the DMN in the pathogenesis and treatment of depression, our study aimed to investigate the performance and effectiveness of DMN neural network connectivity in the impact of iTBS. We employed a within-person-specific approach to the DMN activity map at baseline to cluster individuals with similar patterns of connectivity using a data-driven algorithm. This approach categorised individuals into subgroups based on the similarities and distinctions in their individual networks, thereby revealing patterns of heterogeneity (ie, increased variations in individual-specific connectivity) and homogeneity (ie, reduced variations in connectivity), aiding in the investigation of varying therapeutic responses to iTBS across distinct DMN patterns.

In addition to studying the heterogeneity of depression, substantial investigation has focused on the association between biological markers and therapeutic responses in depression. Emerging evidence suggests that adolescents with depression exhibit abnormal levels of plasma inflammatory cytokines, which are associated with the effectiveness of antidepressant treatments.[10] For example, after antidepressant therapy, pro-inflammatory cytokine levels in adolescents with depression decrease, with non-responders showing significantly higher levels of pro-inflammatory cytokines compared with responders.[11] Additionally, reductions in depressive symptoms and decreases in plasma interleukin-6 (IL-6) and C-reactive protein (CRP) levels have been observed following cognitive behavioural therapy or exercise interventions in adolescents with depression.[12 13] Based on these considerations, inflammatory cytokines were incorporated into this study.

Critically, beyond identifying specific DMN-based network patterns that could offer further elucidation on the treatment outcomes of iTBS in depression with differential neural activities, there is a pressing need to delve deeper into the biological markers influencing iTBS response. The present study examined DMN-based network patterns relating to iTBS treatment in a sample of 82 adolescents and young adults with depression. This secondary analysis of the randomised controlled trial sought to assess the veracity of the following hypotheses: (1) the data-driven neural connectivity network would delineate subgroups of adolescents and young adults with depression exhibiting varying treatment outcomes in a 2-week period of iTBS treatment; and (2) these connectivity subgroups are expected to manifest diverse susceptibilities in treatment outcomes of iTBS with regard to inflammatory cytokines. These questions were examined using functional network analyses at a critical time (adolescents and young adults) for the high incidence of disease to uncover variations in the treatment outcomes of iTBS while delving into the relationship between treatment outcomes and the potential predictors (inflammatory cytokines).

METHODS

Participants

A total of 82 drug-free patients from a randomised, double-blinded and sham-controlled trial of iTBS (China Clinical Trials Registry, ID: ChiCTR2100042346) with neuroimaging data collected prior to treatment were included. The study flow diagram is shown in figures 1 and 2. The written informed consent was obtained from all participants and their parents or legal guardians. The study adhered to the Consolidated Standards of Reporting Trials guidelines. The original study design encompassed the random allocation of eligible participants into one of three groups (twice-daily iTBS group, once-daily iTBS group or sham group). However, for the purposes of this paper, the two active iTBS stimulation cosets are treated as

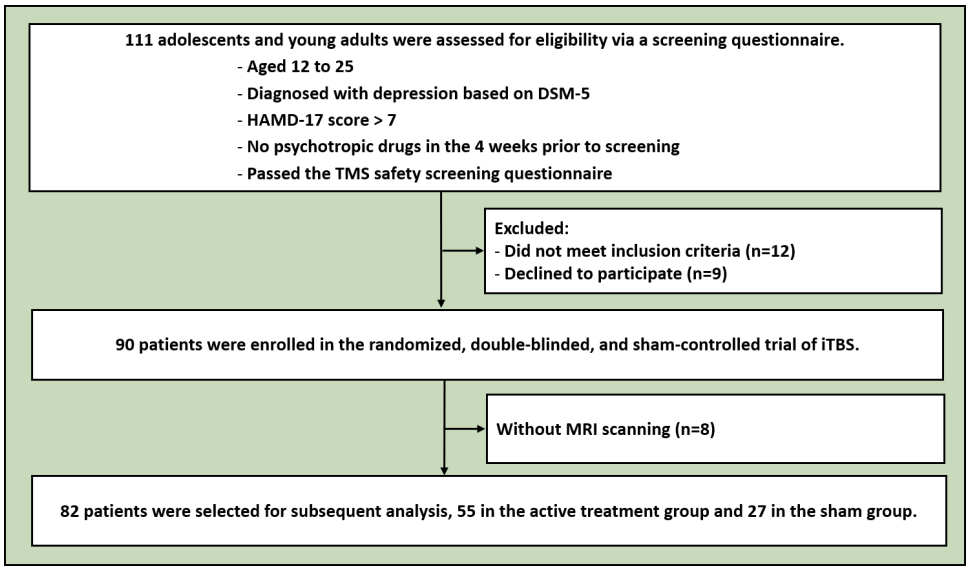

Figure 1 Flowchart of the study. DSM-5, Diagnostic and Statistical Manual of Mental Disorders-5; HAMD-17, Hamilton Depression Scale, 17-item; iTBS, intermittent theta burst stimulation; MRI, magnetic resonance imaging; TMS, transcranial magnetic stimulation.

one group (iTBS stimulation group), compared with the sham group. The ratio of participants in the iTBS stimulation group to those in the sham group was nearly 2:1. The inclusion criteria were as follows: (1) age 12–25 years; (2) diagnosed with MDD or bipolar depression according to the Structured Clinical Interview for the Diagnostic and Statistical Manual of Mental Disorders-5; (3) Hamilton Depression Rating Scale, 17-item (HAMD-17) score>7; (4) no use of psychotropic drugs within the 4 weeks prior to screening; (5) ability to adhere to the study schedule and cooperate during assessments; (6) passing the transcranial magnetic stimulation safety screening questionnaire. Exclusion criteria were: (1) alcohol or substance dependence; (2) presence of serious or unstable medical conditions, including neurological, endocrine, rheumatic disorders, brain disease, traumatic brain injury or surgery and infectious diseases; (3) previous receipt of rTMS or electroconvulsive therapy; (4) high risk of suicide; (5) contraindications for magnetic resonance imaging (MRI) scan and iTBS treatment, such as cardiac pacemaker, nerve stimulator, artificial metal heart valve, intracranial aneurysm clip, cochlear implant and other types of metal implants (except for oral brace); (6) history of epilepsy; and (7) pregnancy. More in-depth accounts of the iTBS treatment can be found in the reference materials.[14]

Patient and public involvement

The research included in this manuscript was conducted and designed without patient or public involvement in study design, outcomes or interpretation of results.

Clinical measures

The HAMD-17, which is the most common and classic scale to evaluate depression severity, was assessed at baseline and 2 weeks of iTBS treatment by well-trained psychiatrists. The rate of reduction in HAMD score was calculated using the formula: [(total pretreatment score − total post-treatment score) / total pretreatment score] × 100%. Cognitive evaluation encompassed assessments of symbol coding, working memory and visual memory, administered through the MATRICS consensus cognitive battery (MCCB). The cognitive scores were independently normalised to a T-score, with adjustments made for confounding variables, including sex, age and education. The average T-score was set at 50, with a standard deviation (SD) of 10. A higher T-score indicates superior cognitive functioning. To determine the influence of the treatment on cognitive performance, the calculation involved the subtraction of values obtained pretreatment and post-treatment.

Functional MRI acquisition and processing

MRI acquisition and preprocessing are detailed in the online supplemental materials.

Data-driven analysis: subgrouping group iterative multiple model estimation

Subgrouping group iterative multiple model estimation (S-GIMME)[15] was implemented for analysing extracted temporal MRI data in the Gimme R package (V.0.7–15). S-GIMME is a unified structural equation modelling method that identifies subgroups of individuals with similar functional connectivity profiles by considering the contemporaneous and first-order lagged connections among predetermined regions of interest (ROIs).[16] Differences in the presence and orientation of these connections signify individual consistency and diversity, which can be leveraged to estimate subgroups. This study used the default parameter of S-GIMME, a selection that was validated through numerical simulation investigation.[15 17] Commencing with a null model, common connections (group-level) are identified as those present in at least

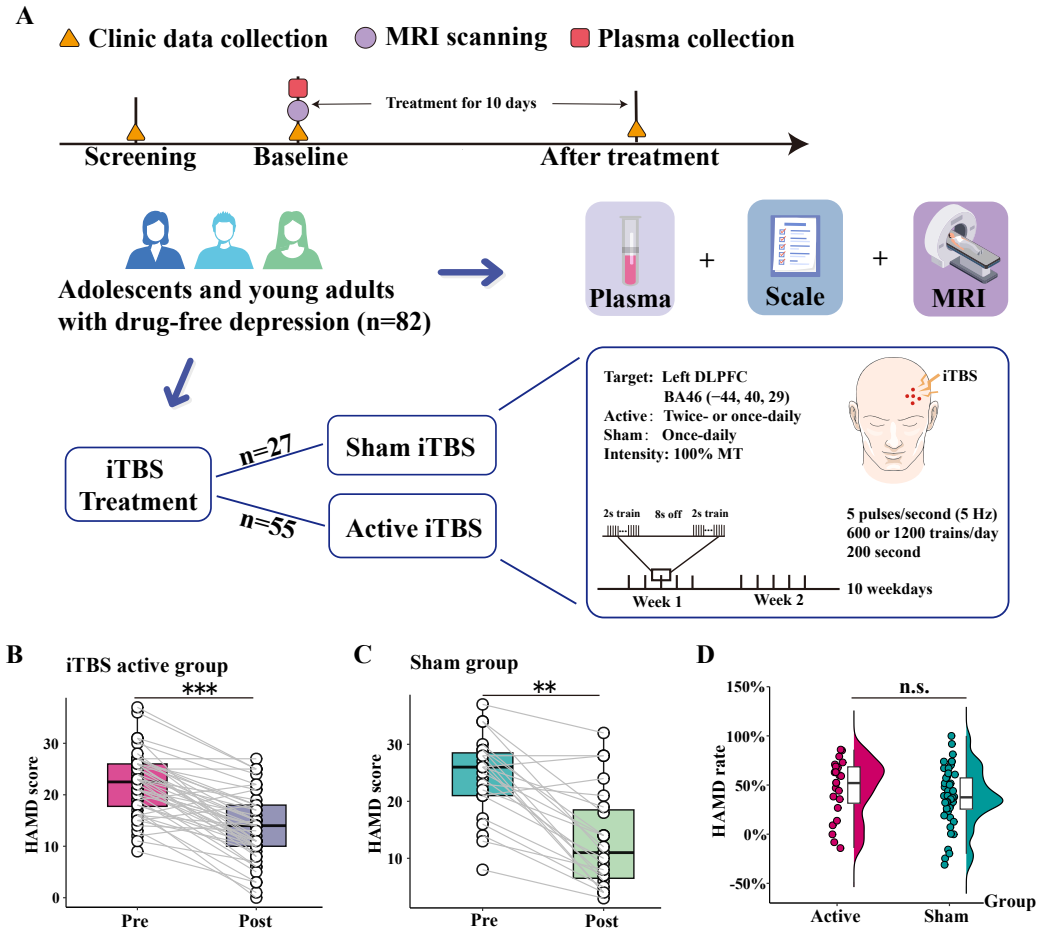

Figure 2 Schematic workflow and clinical characterisation among treatment and subgroups. (A) Schematic diagram of workflow and iTBS intervention. At baseline, adolescents and young adults with drug-free depression (n=82) were assessed with clinical scales and resting state functional connectivity; their plasma was collected after screening for clinical information. Patients were then randomly allocated to either active or sham groups to receive one or two treatment sessions per day for a total of 10 consecutive weekdays. Each session comprised 600 pulses of 5 Hz iTBS. After 2 weeks of iTBS treatment, patients were measured with clinical scales again. (B–D) HAMD scores pretreatment and post-treatment in the iTBS stimulation group (B) and in the sham group (C), and the difference in the HAMD reduction rate between the two groups (D). MRI, magnetic resonance imaging; BA, Brodmann area; DLPFC, dorsolateral prefrontal cortex; HAMD, Hamilton Depression Scale; iTBS, intermittent theta burst stimulation; MRI, magnetic resonance imaging; MT, motor threshold; n.s.,not significant. ***p < 0.001; **p < 0.01; *p < 0.05.

75% of the sampled population and have a significant impact on improving model fit. Following that, the Walktrap[15] algorithm was used to categorise individuals into data-derived subgroups without relying on any preconceived assumptions. Lastly, subgroup-specific connections were integrated for each individual in the subgroup if they contributed significantly to improving the model fit for a minimum of 50% of members, and individual-level connections were estimated for each person until the networks fit optimally. During iteration, the process of finding, adding and pruning each connected path was repeated until no new paths improved the model-fit indices in the group-level, subgroup-level or individual-level model estimation.[16]

When using S-GIMME to identify directed connectivity pathways and subgroups, it is advisable to incorporate a selection of 5–15 ROIs (https://gimme.web.unc.edu/63-2/gimme-basics/). We adopted a hypothesis-driven approach for ROI selection, concentrating on regions within the DMN that are closely associated with depression. Based on previous research, the ROIs in our study were selected from 11 regions within the DMN.[18] The locations of these 11 ROIs are detailed in online supplemental table 1 and figure 3. The extracted time series from these ROIs were then entered into the S-GIMME analysis described below.

In this study, the robustness of the model was primarily assessed: confirmatory fit index (CFI) ≥0.95; non-normed fit index ≥0.95; standardised root mean square residual (SRMR) ≤0.05; root mean square error of approximation ≤0.05.[17] Considering the similarity and robust correlation displayed between the contemporaneous and lagged paths, we primarily computed and depicted the visual representation of the contemporaneous pathway

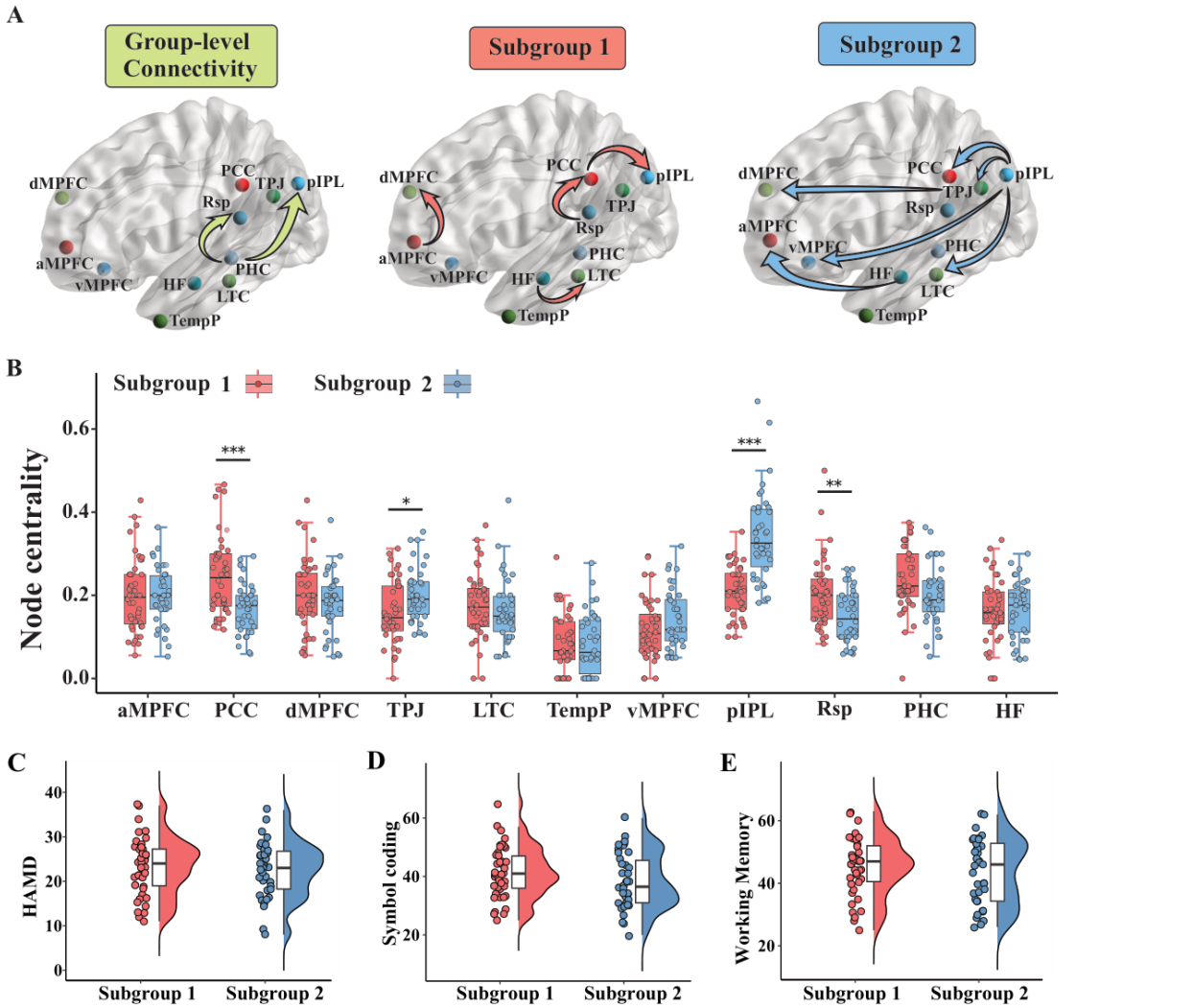

Figure 3 S-GIMME identified group- and subgroup-level effective connectivity. (A) S-GIMME group-level effective connectivity paths. S-GIMME identified the connectivity pathway for subgroup 1 and subgroup 2. Green arrows represent connections common to most subjects. The red arrows indicate a pathway that is significantly stronger in subgroup 1, while the blue arrows represent a pathway that is significantly stronger in subgroup 2. The diagram omits autoregressive and lagged pathways. (B) Node centrality within each ROI plotted separately for each subgroup. (C) Baseline HAMD scores between subgroups. (D–E) Baseline expression of cognitive function between S-GIMME subgroups. The Bonferroni method adjusted the p value. ***p<0.001; **p<0.01; *p<0.05. aMPFC, anterior medial prefrontal cortex; dMPFC, dorsal medial prefrontal cortex; HAMD, Hamilton Depression Scale; HF, hippocampal formation; LTC, lateral temporal cortex; PCC, posterior cingulate cortex; PHC, parahippocampal cortex; pIPL, posterior inferior parietal lobule; ROI, region of interest; Rsp, retrosplenial cortex; S-GIMME, subgrouping group iterative multiple model estimation; TempP, temporal pole; TPJ, temporal parietal junction; vMPFC, ventral medial prefrontal cortex.

to enhance simplicity and clarity.[19] Node centrality was calculated by determining the proportion of contemporaneous connections linked to corresponding nodes (ie, ROIs) relative to the total number of contemporaneous connections.[20] To address multiple comparisons, models incorporating various node centrality measures underwent Bonferroni correction. Another network index, such as graph level degree centrality, was created using the R package 'igraph' (V.2.0.3) (https://github.com/igraph). Following the identification of subgroups identified by S-GIMME, we conducted a comparative analysis of clinical and therapeutic measures across these subgroups. Comprehensive connectivity maps are showcased in figure 3A.

Cytokine assays

We measured two critical inflammatory cytokines, IL-6 and CRP, at baseline to explore the predictive role of inflammatory cytokines in iTBS intervention. As part of the study enrolment process, participants' blood samples were collected. Blood specimens were collected using tubes coated with ethylenediaminetetraacetic acid (EDTA) and subjected to centrifugation at a speed of 3000 revolutions per minute for 10 min at 4 °C. Subsequently, the plasma was separated and stored at a temperature of −80 °C in preparation for subsequent analysis. IL-6 was detected by the human high sensitivity T cell magnetic bead panel (Millipore, Billerica, Massachusetts, USA, HSTCMAG-28SK), and CRP was detected by human

neurodegenerative disease magnetic bead panel 2 (Millipore, HNDG2MAG-36K) via the Luminex Magpix-based assay (Luminex Corporation), as per the manufacturer's guidelines. The resulting data underwent cubic curve fitting and background correction using the Millipore Analyst V.5.1 software (EMD Millipore, Billerica, Massachusetts, USA) for accurate evaluation. Additionally, to achieve normality for statistical analysis, a natural logarithm transformation was applied to all peripheral cytokine values. The coefficients of variation for intra- and inter-assay were below 10% and 15%, respectively.

Statistical analysis

Continuous and categorical demographic and clinical variables were analysed using independent t-tests (or the Mann-Whitney U test) and χ^2 tests, respectively. Pearson product-moment correlation was performed to examine the associations between and within subgroups of symptom improvement and inflammatory cytokines. The general linear model was used to analyse the effects of the subgroup, treatment group (Active (iTBS active simulation) or Sham (iTBS sham simulation)), and their interaction on clinical features to investigate treatment outcome differences. In subsequent analyses, we assessed the models by including baseline expression levels and the primary covariates as sensitivity analyses. To control for potential confounding effects, sensitivity analyses incorporated the following covariates into statistical models: sex, age, diagnosis (MDD or bipolar depression), duration (months), medication (fluoxetine equivalent after 2 weeks), family history (presence/absence) and age of onset (years). Furthermore, in line with previous literature and hypotheses, we evaluated the effects of baseline levels of two key inflammatory cytokines (IL-6 and CRP) on treatment response. We explored the ability of two key inflammatory cytokines to predict the outcome of iTBS using the general linear model. The root-mean-square error (RMSE) and the coefficient of determination (R^2) were used to evaluate the performance of the models in predicting the model. Unless stated otherwise, all analyses and models were conducted using R (V.4.3.2) and SPSS (IBM SPSS Statistics 25). The significance level was set at $p<0.05$.

RESULTS

Clinical characterisation among iTBS treatment

The study flow diagram is shown in figure 2. Table 1 and figure 2B–D display the demographic and clinical information for participants. Both the active group and the sham group demonstrated similar demographic and clinical characteristics and cognitive functions pre-iTBS and post-iTBS treatment. The sham group also exhibited a favourable antidepressant effect, with no significant difference in the HAMD reduction rate compared with the stimulation group (t=1.19, p=0.240). Besides, there were no significant differences in diagnosis (MDD or bipolar depression, p=0.136), medication (fluoxetine equivalent, p=0.231) and use of mood stabilisers (χ^2=1.77, p=0.183) between the two groups.

Data-driven neural network subgroups and connectivity patterns

The final model fit indices demonstrated an outstanding fit across all subjects, with CFI=0.95and SRMR=0.02. S-GIMME identified two distinct subgroups characterised by both shared

Table 1 Clinical characterisation among iTBS treatment groups of patients with depression

Characteristics	Total (n=82)	Active group (n=55)	Sham group (n=27)	Statistics t/χ^2/z	P value
Sex, male, n (%)*	16 (19.51)	8 (14.55)	8 (29.63)	2.62	0.105
Age (years), median (IQR)†	15.00 (13.00–16.00)	15.00 (14.00–17.00)	14.00 (13.00–15.50)	1.85	0.064
Education (years), median (IQR)†	9.00 (7.25–10.00)	9.00 (8.00–11.00)	8.00 (7.00–10.00)	1.62	0.106
Duration (months), median (IQR)†	18.00 (9.25–24.75)	24.00 (12.00–30.00)	15.00 (6.00–24.00)	1.53	0.127
Smoking, smoker, n (%)*	4 (4.88)	3 (5.45)	1 (3.70)	0.12	0.729
Diagnosis, bipolar, n (%)*	14 (17.07)	7 (12.73)	7 (25.93)	2.23	0.136
HAMD, mean (SD)‡	23.02 (6.25)	22.40 (5.98)	24.30 (6.71)	1.25	0.219
Symbol coding, mean (SD)‡	39.88 (9.18)	40.62 (8.32)	38.31 (10.80)	0.96	0.341
Working memory, mean (SD)‡	44.62 (9.75)	45.98 (9.39)	41.73 (10.07)	1.81	0.076
Medication (fluoxetine equivalent), mean (SD)‡§	24.09 (66.85)	16.36 (47.30)	39.82 (94.29)	1.22	0.231
Mood stabilisers, yes, n (%)*§	6 (7.32)	0 (0.00)	6 (22.22)	1.77	0.183

Continuous variables are expressed as mean (SD) or median (IQR).
*Analysed by the χ^2 test.
†Analysed by Mann-Whitney U test.
‡Analysed by independent two-sample t-test.
§All participants were drug-free at enrolment. During the treatment, psychiatrists prescribed medications based on individual patient needs.
HAMD, Hamilton Depression Scale; IQR, interquartile range; iTBS, intermittent theta burst stimulation; SD, standard deviation.

and distinctive patterns of effective network connectivity. First, across all subjects, S-GIMME identified two contemporaneous directed connectivity pathways (figure 3A). These connectivity paths consisted of directed paths from the parahippocampal cortex to the posterior inferior parietal lobule and to the retrosplenial cortex. While these pathways are consistent among subgroups, variations in strength and direction within the DMN were observed across the two identified networks. Ultimate connectivity profiles revealed that subgroup 1 (n=44, figure 3A) had four subgroup-level connections, while subgroup 2 (n=38) exhibited a set of six distinct subgroup-level connections. The number of group-level paths is less than those found within the two distinct subgroups, indicating that the partitioning into subgroups revealed enhanced within-subgroup homogeneity compared with within-sample homogeneity. Subgroup 1 network was characterised by greater heterogeneity (ie, less graph level degree centrality; Subgroup 1: mean (SD), 0.16 (0.05); Subgroup 2: mean (SD), 0.19 (0.07); t=−2.08, p=0.041) compared with subgroup 2 network. Moreover, individuals in subgroup 1 showed significantly greater centrality in the posterior cingulate cortex (t=4.59, *p*FDR<0.001) and retrosplenial cortex (t=3.39, *p*FDR=0.004, figure 3B). In contrast, individuals in subgroup 2 showed significantly greater centrality in the temporal parietal junction (t=2.85, *p*FDR=0.015) and posterior inferior parietal lobule (t=6.57, *p*FDR<0.001; see more information in online supplemental table 2).

Depressive symptoms and cognitive function of two S-GIMME subgroups

The demographic and clinical characteristics of the patients within the two subgroups enrolled are displayed in online supplemental table 3 and did not differ significantly. The HAMD scores and baseline cognitive function of the two S-GIMME subgroups did not differ at baseline (figure 3C–E). No main effect for subgroup, treatment or subgroup×treatment interaction was revealed in the improvement of depressive symptoms (all p>0.05, figure 4A,D). A significant subgroup×active group interaction related to symbol coding improvement (F=5.22, p=0.026; figure 4B,E, online supplemental table 4) was detected, but no such interaction was observed in working memory (figure 4C,F). This interaction was adjusted for potentially confounding variables, including sex, age, diagnosis, duration, medication, family history, age of onset and baseline symbol coding function. Specifically, within subgroup 1, the active group showed significantly greater improvement in symbol coding compared with the sham group (t=2.30, p=0.028; figure 4E). However, there is no such difference within subgroup 2. Additionally, at both pretreatment and post-treatment, the symbolic coding scores of patients receiving active stimulation were higher than those receiving sham stimulation in subgroup 1 (active group vs sham group, pretreatment: t=2.20, p=0.038; post-treatment: t=3.30, p=0.003; figure 4B).

Predictive role of inflammatory cytokines in active stimulation treatment outcomes

The main objective was to explore potential distinctions in baseline levels of two pivotal inflammatory cytokines (IL-6 and CRP), along with the correlation of symptoms and their impact on treatment response among the groups. No significant distinctions were observed between the active group and the sham group, as well as between subgroup 1 and subgroup 2 (online supplemental table 5). No significant correlations between baseline depressive symptoms and cognitive function and inflammatory cytokines were found between the DMN-based network subgroups or the iTBS group (online supplemental tables 6 and 7). Inside subgroup 1, the symbol coding change for those receiving active stimulation was positively correlated with IL-6 (r=0.45, p=0.030, figure 4G and online supplemental table 8), and the evidence maintained its statistical significance and became even stronger after controlling for covariates (β=0.45, r=0.57, p=0.035). However, no significant correlation was found in the sham group of subgroup 1, nor in either of the groups within subgroup 2 (figure 4H). In the active group, compared with subgroup 2, baseline levels of IL-6 and CRP in subgroup 1 were predictive of improvement in symbol coding (R^2=0.35, RMSE=5.72, p=0.013; figure 4I and online supplemental table 9).

DISCUSSION

Main findings

The current study aimed to investigate whether connectivity subgroups among depression patients are associated with the outcomes of iTBS treatment and to identify predictors within these connectivity subgroups. Data-driven analyses identified subgroup 1, characterised by high heterogeneity and low graph-level degree centrality, and subgroup 2 with similar patterns of connectivity in key neural networks (DMN). We observed that among depression patients receiving active stimulation, subgroup 1 showed greater improvement in symbolic coding scores compared with subgroup 2. Furthermore, in subgroup 1, the pretreatment levels of IL-6 and CRP can predict the outcomes of active stimulation in improving symbolic coding ability. Both subgroups showed similar improvement in depressive symptoms following active stimulation. These findings imply that using data-driven network analyses could offer new insights into clinical outcomes of iTBS treatment and provide clues for predicting cognitive improvements from an inflammatory perspective.

Implications

The DMN has garnered significant attention in the field of clinical neuroscience of MDD. It is recognised that the DMN is not a singular entity but rather consists of various anatomical-functional systems, each fulfilling distinct functions.[18] Our study found that S-GIMME identified two concurrent connectivity pathways based on the similarity of connectivity in DMN among all patients, but there were four and six pathways in subgroup 1 and subgroup 2, respectively. This result suggests that partitioning into subgroups resulted in greater homogeneity within the subgroups compared with the overall sample, as evidenced by fewer group-level pathways compared with those found within the two distinct subgroups. These results also indicate significant

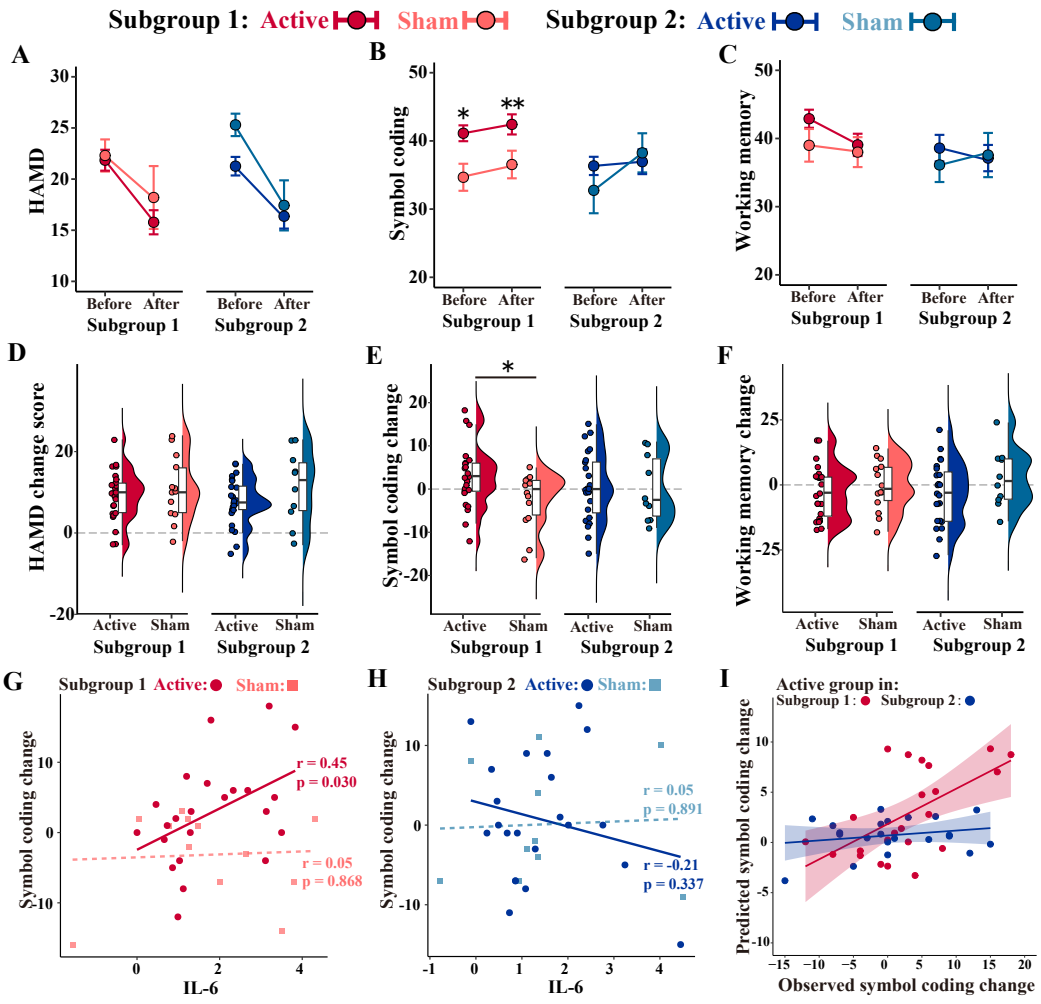

Figure 4 Depressive symptoms and cognitive function among different subgroups pretreatment and post-treatment, as well as correlation and predictive analysis of symbol coding changes with baseline inflammatory cytokine levels. (A–C) The figures represent the pretreatment and post-treatment values of HAMD scores (A) and cognitive function (B–C). (D–F) The change in HAMD scores and cognitive function in each subgroup post-treatment. The distributions of warm and cool colours correspond to subgroup 1 and subgroup 2, respectively. 'Active' and 'Sham' represent patients who received active and sham iTBS stimulation, respectively. (G–I) Correlation between baseline inflammatory factor IL-6 and symbol coding change in subgroup 1 (G) and subgroup 2 (H) with different iTBS treatment. (I) Inflammatory cytokines (IL-6 and CRP) were used to predict changes in symbol coding following iTBS treatment in subgroup 1 (red) and subgroup 2 (blue) with active iTBS treatment. CRP, C-reactive protein; HAMD, Hamilton Depression Scale; IL-6, interleukin-6; iTBS, intermittent theta burst stimulation. **p<0.01; *p<0.05.

heterogeneity in patterns of connectivity in DMN among patients with depression. Similar to previous research findings, Liang and his colleagues used a data-driven approach to analyse the connectivity of the DMN in 690 patients with MDD, identifying two subgroups of MDD.[21]

In a recent meta-analysis of sham-controlled randomised controlled studies conducted in patients with MDD, it was shown that rTMS demonstrates modest effect size improvements in processing speed, but no changes were observed in other cognitive dimensions.[22] Currently, scholars hold the view that rTMS can improve specific cognitive dimensions in some patients with MDD.[23] Our study findings support this view—subgroup 1 showed significant improvement in symbolic coding scores (the primary component of processing speed) after active stimulation, while subgroup 2 showed no significant improvement. These results are consistent with the notion that there may be greater neural flexibility in the biological features of cognitive improvement.[24] The DMN consists of three heterogeneous subsystems: the core DMN subsystem, the dorsal medial prefrontal cortex subsystem and the medial temporal lobe subsystem.[18] In our study, subgroup 1 exhibited greater centrality in the posterior cingulate cortex (PCC) and retrosplenial cortex than subgroup 2. The PCC, a central hub in the core DMN subsystem, connects the three subsystems.[18] Its higher centrality in subgroup 1 suggests increased DMN flexibility. In contrast, subgroup 2 displayed more homogeneous connectivity patterns, with greater centrality in the temporoparietal junction and posterior inferior parietal lobule, regions linked to the dorsal medial prefrontal cortex and medial temporal lobe subsystems, respectively.[25] These areas are crucial for theory of mind (to infer the representational

mental state of another individual, such as a belief or intention) and autobiographical memory. Previous research has shown that during rumination,[25] the left medial temporal lobe subsystems exhibit increased functional connectivity with other brain regions,[26] which may lead to more severe impairments in cognitive flexibility.[27] These results support the idea that greater neural flexibility in cognitive-related biological features may contribute to cognitive improvement. It is also important to note that we did not observe differences in depressive symptom improvement between the subgroups post-treatment. Since our iTBS intervention lasted only 2 weeks, extending the treatment duration may reveal further antidepressant efficacy.

We found no differences in depression-related risk factors (IL-6 levels and CRP levels) between subgroups. However, the significant correlation between pretreatment IL-6 levels and symbolic coding improvement in subgroup 1 persisted even after controlling for confounding variables. These findings suggest that IL-6 is an independent predictor of symbolic coding improvement in subgroup 1. Previous studies have used iTBS to treat treatment-resistant depression patients but found that baseline levels of several inflammatory cytokines cannot predict treatment outcomes.[28] We speculate that the neural heterogeneity of depression may be a potential cause of the negative results. Patients with depression exhibit loss of the endothelial cell tight junction protein Cldn-5, leading to increased blood–brain barrier permeability.[29] Peripheral inflammatory cytokines may penetrate the blood–brain barrier due to this increased permeability, inducing the generation of neuroinflammation. We hypothesise that the DMN connectivity patterns in subgroup 1 regulate the relationship between IL-6 levels and CRP levels and cognitive improvement. Furthermore, our predictive model suggests that IL-6 levels and CRP levels can predict cognitive function after iTBS intervention in subgroup 1, indicating that IL-6 levels and CRP levels are potential predictors of cognitive improvement.

Limitations

Several limitations should be noted. First, the sample size in this study was relatively small. However, S-GIMME simulation work indicates robust model fits and a reduction in model overfitting, even with small sample sizes as low as 25.[15] Future research should include more participants to ascertain the replicability and validity of the subgroups observed in our study. Second, combining subjects with MDD and bipolar depression in this study may introduce variability due to disease heterogeneity. Previous research has shown that both MDD and bipolar depression exhibit decreased whole-brain functional connectivity and similar alterations in the DMN.[30] Additionally, future studies could benefit from integrating multimodal MRI techniques to more comprehensively explore the neural subgroups of depression.

CONCLUSION

Our study suggests that the outcome of iTBS treatment in adolescents and young adults with depression may be influenced by DMN-based network patterns. Pretreatment levels of IL-6 and CRP serve as potential predictors for cognitive improvements within specific biotypes. These findings imply that using data-driven network analyses could offer new insights into clinical outcomes of iTBS treatment and provide clues for predicting cognitive improvements from an inflammatory perspective.

Author affiliations
[1]Department of Child and Adolescent Psychiatry, The Affiliated Brain Hospital, Guangzhou Medical University, Guangzhou, Guangdong, China
[2]The First School of Clinical Medicine, Southern Medical University, Guangzhou, Guangdong, China
[3]Guangdong Engineering Technology Research Center for Translational Medicine of Mental Disorders, Guangzhou, Guangdong, China
[4]Key Laboratory of Neurogenetics and Channelopathies of Guangdong Province and the Ministry of Education of China, Guangzhou Medical University, Guangzhou, Guangdong, China
[5]State Key Laboratory of Brain and Cognitive Sciences, Department of Psychology, The University of Hong Kong, Hong Kong, China
[6]Department of Psychiatry, The Chinese University of Hong Kong, Shatin, Hong Kong, China
[7]Canadian Rapid Treatment Center of Excellence, Mississauga, ON, Canada
[8]Mood Disorders Psychopharmacology Unit, Poul Hansen Depression Centre, University Health Network, Toronto, ON, Canada
[9]Department of Psychiatry, University of Toronto, Toronto, ON, Canada
[10]Institute of Medical Science, University of Toronto, Toronto, ON, Canada
[11]Brain and Cognition Discovery Foundation, Toronto, ON, Canada
[12]Department of Pharmacology, University of Toronto, Toronto, ON, Canada

Acknowledgements The authors express their gratitude to the patients who participated in the trial and acknowledge the valuable contributions of all investigators. The authors would like to extend their appreciation to the members of the Clinical Research Ethics Committees at the Affiliated Brain Hospital of Guangzhou Medical University for their assistance and diligent oversight throughout the study.

Contributors WL, YY, ZY: conceptualisation, methodology, software, visualisation, data curation, writing—original draft, writing—review and editing. MZ, CW, XL: methodology, resources, investigation, writing—review and editing. MS, FZ, ZH, GL, XC, HL, ZL, YZ, YiyingC, YifangC: methodology, resources, investigation. RS, HL: writing—review and editing, investigation. RSM: methodology, data curation. YZ: conceptualisation, funding acquisition, project administration, methodology, writing—review and editing. YN: conceptualisation, funding acquisition, investigation, supervision, project administration. Author YN is responsible for the overall content (as guarantor).

Funding This work was supported by the Guangzhou Municipal Key Discipline in Medicine (2021-2023), the Guangzhou High-level Clinical Key Specialty, the Guangzhou Research-oriented Hospital, the Innovative Clinical Technique of Guangzhou (2024-2026), the Guangdong Basic and Applied Basic Research Foundation (grant number 2022A1515011567, 2020A1515110565), the Guangzhou Science, Technology Planning Project (grant number 202201010714, 202103000032), the National Natural Science Foundation of China (grant number 82471546), the Guangdong College Students Innovation and Entrepreneurship Training Project (grant number S202310570038), the Guangzhou Health Science and Technology Project (grant number 20231A010038), the Guangzhou Traditional Chinese Medicine and Integrated Traditional Chinese and Western Medicine Technology Project (grant number: 20232A010013), and the Science and Technology Plan Project of Guangzhou (2023A03J0842). The funding source had no role in the study design, analysis or interpretation of data or in the preparation of the report or decision to publish.

Competing interests None declared.

Patient consent for publication Not applicable.

Ethics approval This is a secondary analysis, using data from the randomised controlled trial of iTBS in depression (n=82) which has undergone peer review and has been published in a reputable scientific journal. The trial took place at the Affiliated Brain Hospital following approval from the research ethics boards (ChiCTR2100042346), and written informed consent was obtained from all participants as well as their parents or legal guardians.

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement Data are available upon reasonable request.

ORCID iD

Yanling Zhou http://orcid.org/0000-0002-5727-2782

REFERENCES

1 Waszczuk MA, Zavos HMS, Gregory AM, *et al*. The phenotypic and genetic structure of depression and anxiety disorder symptoms in childhood, adolescence, and young adulthood. *JAMA Psychiatry* 2014;71:905–16.
2 Lan X, Wang C, Zhang F, *et al*. Efficacy of repeated intravenous esketamine in adolescents with anxious versus non-anxious depression. *Gen Psychiatr* 2023;36:e101007.
3 McClintock SM, Reti IM, Carpenter LL, *et al*. Consensus recommendations for the clinical application of repetitive transcranial magnetic stimulation (rTMS) in the treatment of depression. *J Clin Psychiatry* 2018;79:16cs10905.
4 Deckersbach T, Peters AT, Shea C, *et al*. Memory performance predicts response to psychotherapy for depression in bipolar disorder: a pilot randomized controlled trial with exploratory functional magnetic resonance imaging. *J Affect Disord* 2018;229:342–50.
5 Liu R, Qi H, Guan L, *et al*. Functional connectivity of the default mode network subsystems in patients with major depressive episodes with mixed features. *Gen Psychiatr* 2022;35:e100929.
6 Chahal R, Gotlib IH, Guyer AE. Research review: brain network connectivity and the heterogeneity of depression in adolescence - a precision mental health perspective. *J Child Psychol Psychiatry* 2020;61:1282–98.
7 Ho TC, Connolly CG, Henje Blom E, *et al*. Emotion-dependent functional connectivity of the default mode network in adolescent depression. *Biol Psychiatry* 2015;78:635–46.
8 Cao J, Ai M, Chen X, *et al*. Altered resting-state functional network connectivity is associated with suicide attempt in young depressed patients. *Psychiatry Res* 2020;285:112713.
9 Kim SM, Park SY, Kim YI, *et al*. Affective network and default mode network in depressive adolescents with disruptive behaviors. *Neuropsychiatr Dis Treat* 2016;12:49–56.
10 Toenders YJ, Laskaris L, Davey CG, *et al*. Inflammation and depression in young people: a systematic review and proposed inflammatory pathways. *Mol Psychiatry* 2022;27:315–27.
11 Amitai M, Taler M, Carmel M, *et al*. The relationship between plasma cytokine levels and response to selective serotonin reuptake inhibitor treatment in children and adolescents with depression and/or anxiety disorders. *J Child Adolesc Psychopharmacol* 2016;26:727–32.
12 Gazal M, Souza LD, Fucolo BA, *et al*. The impact of cognitive behavioral therapy on IL-6 levels in unmedicated women experiencing the first episode of depression: a pilot study. *Psychiatry Res* 2013;209:742–5.
13 Pace TWW, Negi LT, Dodson-Lavelle B, *et al*. Engagement with cognitively-based compassion training is associated with reduced salivary C-reactive protein from before to after training in foster care program adolescents. *Psychoneuroendocrinology* 2013;38:294–9.
14 Zhang M, Li W, Ye Y, *et al*. Efficacy and safety of intermittent theta burst stimulation on adolescents and young adults with major depressive disorder: a randomized, double blinded, controlled trial. *J Affect Disord* 2024;350:214–21.
15 Gates KM, Lane ST, Varangis E, *et al*. Unsupervised classification during time-series model building. *Multivariate Behav Res* 2017;52:129–48.
16 Beltz AM, Gates KM. Network mapping with GIMME. *Multivariate Behav Res* 2017;52:789–804.
17 Gates KM, Molenaar PCM. Group search algorithm recovers effective connectivity maps for individuals in homogeneous and heterogeneous samples. *Neuroimage* 2012;63:310–9.
18 Andrews-Hanna JR, Reidler JS, Sepulcre J, *et al*. Functional-anatomic fractionation of the brain's default network. *Neuron* 2010;65:550–62.
19 Stout DM, Harlé KM, Norman SB, *et al*. Resting-state connectivity subtype of comorbid PTSD and alcohol use disorder moderates improvement from integrated prolonged exposure therapy in Veterans. *Psychol Med* 2023;53:332–41.
20 Hardi FA, Goetschius LG, McLoyd V, *et al*. Adolescent functional network connectivity prospectively predicts adult anxiety symptoms related to perceived COVID-19 economic adversity. *J Child Psychol Psychiatry* 2023;64:918–29.
21 Liang S, Deng W, Li X, *et al*. Biotypes of major depressive disorder: neuroimaging evidence from resting-state default mode network patterns. *Neuroimage Clin* 2020;28:102514.
22 Martin DM, McClintock SM, Forster JJ, *et al*. Cognitive enhancing effects of rTMS administered to the prefrontal cortex in patients with depression: a systematic review and meta-analysis of individual task effects. *Depress Anxiety* 2017;34:1029–39.
23 Iimori T, Nakajima S, Miyazaki T, *et al*. Effectiveness of the prefrontal repetitive transcranial magnetic stimulation on cognitive profiles in depression, schizophrenia, and Alzheimer's disease: a systematic review. *Prog Neuropsychopharmacol Biol Psychiatry* 2019;88:31–40.
24 Qiao L, Xu M, Luo X, *et al*. Flexible adjustment of the effective connectivity between the fronto-parietal and visual regions supports cognitive flexibility. *Neuroimage* 2020;220:117158.
25 Andrews-Hanna JR, Reidler JS, Huang C, *et al*. Evidence for the default network's role in spontaneous cognition. *J Neurophysiol* 2010;104:322–35.
26 Chen X, Chen N-X, Shen Y-Q, *et al*. The subsystem mechanism of default mode network underlying rumination: a reproducible neuroimaging study. *Neuroimage* 2020;221:117185.
27 Altan-Atalay A, Kaya-Kızılöz B, İlkmen YS, *et al*. Impact of abstract vs. concrete processing on state rumination: an exploration of the role of cognitive flexibility. *J Behav Ther Exp Psychiatry* 2022;74:101691.
28 Chou P-H, Lu M-K, Tsai C-H, *et al*. Antidepressant efficacy and immune effects of bilateral theta burst stimulation monotherapy in major depression: a randomized, double-blind, sham-controlled study. *Brain Behav Immun* 2020;88:144–50.
29 Menard C, Pfau ML, Hodes GE, *et al*. Social stress induces neurovascular pathology promoting depression. *Nat Neurosci* 2017;20:1752–60.
30 Siegel-Ramsay JE, Bertocci MA, Wu B, *et al*. Distinguishing between depression in bipolar disorder and unipolar depression using magnetic resonance imaging: a systematic review. *Bipolar Disord* 2022;24:474–98.

Weicheng Li is currently pursuing a PhD in Psychiatry and Mental Health at the Affiliated Brain Hospital of Guangzhou Medical University in China. His research focuses on the pathogenesis of depression and the mechanisms of action of antidepressant treatments. He has published six SCI papers as the first author or co-first author in journals such as Depression and Anxiety, Journal of Affective Disorders, and Journal of Psychiatric Research.