Plasma Kynurenine Level is Associated With Left DLPFC Activity and Can Predict Treatment Response in Major Depressive Disorder

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Abstract

To establish treatment response biomarkers that reflect the pathophysiology of depression, it is important to use an integrated set of features that are promising as biomarkers. This study aimed to determine the relationship between blood metabolites related to treatment response to escitalopram and regional brain activity at rest and to find the characteristics of depression that respond to treatment. Blood metabolite levels and resting brain activity were measured in patients with depression (N = 65) before and after 6 weeks treatment with escitalopram and healthy controls (N = 36). Thirty-two patients (49.2%) showed clinical response (>50% reduction in Hamilton Rating Scale for Depression score) and were classified as Responders, and the remaining 33 patients were classified as Nonresponders. Pretreatment plasma kynurenine level and fractional amplitude of low-frequency fluctuations (fALFF) of the left dorsolateral prefrontal cortex (DLPFC) were lower in Responders, and their elevations after treatment were correlated with improvement in symptoms. Moreover, fALFF of the left DLPFC was significantly correlated with plasma kynurenine level in pretreatment patients with depression and healthy controls. Decreased kynurenine level and resting-state regional brain activity of the left DLPFC may be involved in the pathophysiology of depression in response to escitalopram treatment.

Introduction

Antidepressants are recommended as the first-line treatment for moderate to severe depression[1]. However, response and remission rates to antidepressant therapy remain disappointingly low. The largest pragmatic trial of depression to date, the Sequenced Treatment Alternatives to Relieve Depression (STAR*D) trial, reported that only 48.6% of patients respond to initial antidepressant treatment and 36.8% of patients achieve remission[2]. To make appropriate treatment choices and prevent unnecessary drug administration, it is necessary to develop biomarkers that can objectively assess the characteristics of patients with depression who respond to specific antidepressants and help predict treatment response.

Several promising biomarkers for treatment response have been reported in the fields of genetic variation, gene expression profiling, proteomics, metabolomics, neuroendocrinology, electrophysiology, and neuroimaging, but none have been established yet[3]. To establish a treatment response biomarker that reflects the pathophysiology, it is important to clarify the relationship between each biomarker in the treatment response and use them in an integrated manner.

Metabolites are the final products of interactions between gene expression, protein function, and cellular environment[4]. Thus, metabolomics approach holds great promise for the identification of pathways involved in antidepressant response and pathophysiology of depression[5]. For example, with regard to the treatment response to sertraline, studies reported that the metabolic pathways of phenylalanine, tryptophan (TRP), purine, and tocopherol are associated[6] and that a decrease in kynurenine (KYN)/melatonin (MEL) ratio and 3-hydroxykynurenine (3-HK)/MEL ratio are associated[7]. Studies also showed that treatment response to escitalopram is associated with a low glycine level[8] and that a high

serotonin level and a low KYN/TRP ratio are related[9]. Recently, we also reported that low KYN and kynurenic acid (KYNA) levels were associated with treatment response to escitalopram[10].

Conversely, in neuroimaging studies, functional magnetic resonance imaging (fMRI) has the advantage of high spatial resolution and noninvasive assessment of brain function, and several regional brain activities involved in treatment response have been reported. For example, in a study using task-based fMRI, the group that achieved remission after treatment with escitalopram and sertraline had the same pretreatment dorsolateral prefrontal cortex (DLPFC) activation during response inhibition as control participants[11]. Studies reported that low activation of the anterior cingulate gyrus, DLPFC, thalamus, and caudate nucleus in response to negative word stimuli is associated with a favorable treatment response to escitalopram[12] and that low activation of the amygdala in response to negative facial expressions is associated with a favorable treatment response to paroxetine[13]. In a study using resting-state fMRI, a decrease in resting-state functional connectivity in the cognitive control network was associated with non-remission[14] and that the strength of functional connectivity between the anterior insula and left DLPFC was associated with early favorable treatment response[15]. We also reported that fractional amplitude of low-frequency fluctuations (fALFF) in the right thalamus is overactive in treatment-resistant depression and that its activity is negatively correlated with treatment response in untreated depression[16].

Thus, blood metabolomics and fMRI are both promising in the development of biomarkers for predicting response to treatment of depression, but each has its advantages and disadvantages. Although samples of blood metabolomics are easy to obtain, it cannot directly evaluate brain function, which is the locus of pathology. Conversely, fMRI has the advantage of noninvasively assessing the function of each region of the whole brain but does not provide information on metabolic mechanisms. It is possible to obtain more insight into the pathogenesis of depression by measuring and analyzing both in the same subject in an integrated manner. However, few studies have examined how both biomarkers relate to each other in antidepressant treatment response or changes before and after treatment, and it is unclear how blood metabolites are related to activity in regional brain regions and contribute to treatment response.

In this study, we investigated the interrelationship between blood metabolites and resting-state regional brain activity, which affect escitalopram treatment response, by measuring blood metabolite levels and performing resting-state fMRI before and after escitalopram treatment in patients with depression.

Materials And Methods

Participants

A total of 65 patients with major depressive disorder (MDD) in the acute phase of the disease were recruited from the Hiroshima University and local clinics according to the following inclusion criteria: (a) age between 25 and 75 years; (b) outpatient status; (c) presentation of moderate or more severe depressive symptoms, as determined by a score on the 17-item Hamilton Rating Scale for Depression

(HRSD) of 14 or more[17]; and (d) diagnosis of non-psychotic MDD and current depressive episode, as determined by an experienced psychiatrist according to the Diagnostic and Statistical Manual of Mental Disorders, Fourth edition (DSM-IV) and verified through Mini-International Psychiatric Structural Interview (MINI)[18,19] conducted by trained valuators. The exclusion criteria for patients were as follows: (a) diagnosis of neurological disease, current or previous psychotic disorder, current high risk of suicide, current or previous substance abuse, and serious somatic disease as determined by the MINI conducted by trained valuators; (b) left-handedness, which was defined as a score < 0 on the Edinburgh handedness test[20]; and (c) current pregnancy or nursing. They had not started using antidepressants for their current depressive episode or had been taking escitalopram for less than 14 days.

Healthy control subjects (HC) were recruited from the local community, and their ages and sex were matched with the MDD patient group. They were interviewed with the MINI, and none showed a history of psychiatric disorders according to DSM-IV criteria.

Patients with MDD provided blood samples and underwent MRI during the acute phase of the disease (within 2-week administration of treatment [T1]) and after approximately 6 weeks of treatment with escitalopram (T2). HRSD was administrated to measure each patient's severity of depression at each session. HC participants provided blood samples and underwent MRI once.

The study was conducted in compliance with relevant guidelines and regulations and the latest version of the World Medical Association's Declaration of Helsinki. The current study protocol was approved by the Ethics Committee of Hiroshima University and the Institutional Ethics Committee of the University of Tokushima Graduate School. Before the administration of any experimental procedure, written informed consent was obtained from all participants.

Quantitative metabolome analysis

Blood sampling was performed during their visit to the Hiroshima University and local clinics. Samples were sent from Hiroshima University to Tokushima University, where each 50 μL sample was mixed with 450 μL of methanol containing internal standards (10 μM) and vortexed. Chloroform (500 μL) and Milli-Q water (200 μL) were added, mixed thoroughly, and centrifuged (2300 × g, 4°C, 5 min). Moreover, 375μL of the aqueous layer was filtered through a 5-kDa cutoff filter (EMD Millipore, Billerica, MA, USA) to remove macromolecules. The filtrate was lyophilized and dissolved in 50 μL of Milli-Q water containing the reference compound before MS analysis.

Metabolome measurements were conducted at Tokushima University. Plasma metabolite profiling and mixture of 110 standard metabolites (50 μM each, HMT, Tsuruoka, Japan) were analyzed using capillary electrophoresis electrospray ionization time-of-flight mass spectrometry (CE-ESI-TOFMS) system (Agilent 7100 CE − 6230 TOFMS, Agilent Technologies, Palo Alto, CA, USA) in cation and anion modes, with a mass range of 50–1000. Metabolites in the samples were analyzed using fused silica capillary (50 μm i.d. × 80 cm length) filled with electrolyte buffer solution (HMT, Tsuruoka, Japan) with the applied voltage or cation and anion modes set at 27 kV and 30 kV, respectively.

Peak information, such as mass-to-charge ratio (m/z), migration time (MT), and peak area, was extracted from the peaks detected by CE-TOFMS using MassHunter integration software (Agilent Technologies). The peaks were annotated to the metabolites inferred from the standard metabolites based on their MT and m/z values. The tolerance range of peak annotation was ± 0.2 min for MT and ± 10 ppm for m/z. The peak areas were normalized to the internal standard.

MRI data acquisition, preprocessing, and analysis

Functional brain images were collected using four different 3T MRI scanners. All scans were acquired with echo planar imaging sequences. Details on MRI acquisition are summarized in Supplementary Table S1. During resting-state fMRI, subjects were instructed to look at a central fixation point, lie still, stay awake, and not think of anything specific.

The first eight images were discarded to ensure steady-state fMRI signals during acclimation of the subjects. Then, images were preprocessed using CONN version 20.b[21]. After slice timing correction, realignment, normalization, smoothing (6-mm FWHM Gaussian filter), the images were co-registered to the structural data using a linear transformation, and normalized to MNI space using a nonlinear transformation. The Artifact Detection Tools (ART) in CONN identified outlier images if the head motion in x, y, or z direction was greater than 1 mm or if the global mean intensity in the image was greater than three standard deviations from the mean image intensity for all images. Individual T1-weighted images were segmented into gray matter, white matter, and cerebrospinal fluid (CSF) and generated three masks. Linear regression was performed to remove the confounding effects of ART-based scrubbing parameters containing invalid scans and BOLD signals from the white matter and CSF, which were used for aCompCor, head motion confounding defined by six rigid body motion parameters and six first-order time derivatives. Then, band-pass filtering (0.008–0.09 Hz) and linear detrending were applied to the resulting residual BOLD time series. Subsequently, using the CONN toolbox option, voxelwise fALFF was calculated within each voxel. fALFF describes the level of spontaneous activity of each individual element in the resting state from all aspects of energy and is used to evaluate the regional spontaneous activity level of the brain[22]. It has been suggested that fALFF analysis is the most sensitive measure for detecting the effects of metabolic states on resting state brain activity[23,24].

Statistical analyses

Demographic and clinical data were compared using the chi-square test for categorical variables and two-sample t-test for quantitative variables. Statistical analyses were conducted using R version 4.0.3 software. Significance was set at a two-tailed P-value < 0.05.

For each metabolite, the Mann–Whitney U test was used to compare Responders and Nonresponders, and the Kruskal–Wallis and Dunn–Bonferroni post hoc tests were used to compare Responders, Nonresponders, and HC. A significance test of Spearman's rank correlation coefficient was used to examine relationships between escitalopram treatment response and change of each metabolite level in patients with MDD. Changes in KYN levels before and after treatment with escitalopram were compared

using the Wilcoxon signed-rank test. These statistical analyses were performed using IBM SPSS Statistics version 26.0 (IBM Corp., Armonk, NY, USA). Significance was set at a two-tailed P-value < 0.05.

A two-sample t-test was performed on the fALFF values for each voxel using Statistical Parametric Mapping (SPM12) software (Welcome Department of Cognitive Neurology, London, UK). The significance level was set at familywise error (FWE)-corrected P-value < 0.05 for cluster level inference. Correlation analysis of left DLPFC activity to KYN and treatment response was performed using semipartial correlation with each MRI scanner as a control variable. One-way analysis of covariance with each MRI scanner as a covariate was used for comparison among the three groups in the activity of the left DLPFC. These statistical analyses were performed using R version 4.0.3 software. Significance was set at a two-tailed P-value < 0.05.

Results

Demographics

Demographic information, including sex, age, and HRSD17 score; body mass index (BMI); verbal IQ; and escitalopram dose are shown in Table 1. Verbal IQ was assessed by the Japanese version of the Adult Reading Test (JART)[25]. Responders and Nonresponders did not differ significantly in sex (16/16 [50.0% male] vs. 15/18 [45.5% male], P = 0.71, chi-square test), age (38.5 ± 9.8 vs. 41.9 ± 11.6, P = 0.21, two-sample t-test), and average baseline HRSD17 score (19.1 ± 4.5 vs. 20.8 ± 5.0, P = 0.15, two-sample t-test). Follow-up HRSD was recorded for all 65 patients, and there was a significant difference in the HRSD scores recorded at baseline and follow-up (P < 0.05, paired-sample t-test). Moreover, 32 patients (49.2%) showed clinical response (>50% reduction in HRSD score) and were classified as Responders; the remaining 33 patients (50.7%) were classified as Nonresponders. Furthermore, 25 patients (38.5%) achieved remission (follow-up HRSD score < 8).

Comparison between Responders and Nonresponders at baseline (T1)

To detect metabolites useful for escitalopram therapy, we performed the metabolomic analysis of plasma metabolites at 2 time points, baseline (T1) and post-treatment (T2), in 65 patients with depression. We also performed metabolomic analysis of metabolites in plasma at one time point for 36 healthy control subjects. We identified 34 metabolites in those metabolomic analyses. The Mann–Whitney U test was used to compare the blood levels of Responders and Nonresponders at baseline (T1) for each of the 34 metabolites. Among them, KYN levels were significantly lower in Responders than in Nonresponders (Table 2).

To identify the brain regions related to treatment response, we compared Responders and Nonresponders at baseline (T1) for each voxel's fALFF value. As a result, Responders had significantly lower fALFF values than Nonresponders in the left DLPFC (FWE-corrected P = 0.022 for cluster level inference, 65 voxels) (Fig. 1).

Comparison of KYN level and left DLPFC activity at baseline (T1) in the three groups

KYN levels were significantly lower in Responders than in both HC and Nonresponders, although no difference was observed between Nonresponders and HC (Fig. 2a). However, this trend was not observed in other metabolites (Supplementary Table S2).

The 65 voxels of the left DLPFC determined to be significant at the cluster level after comparing Responders and Nonresponders were used as the region of interest (ROI). The activity of each left DLPFC was assessed by the mean fALFF value of the left DLPFC ROI. The lower mean fALFF value of the left DLPFC ROI was significantly lower in Responders than in Nonresponders and HC (Fig. 2b).

Relationship between plasma KYN level and left DLPFC activity at baseline (T1)

Then, we examined the relationship between baseline (T1) plasma KYN level and left DLPFC activity for each individual with MDD and HC. The results showed that there was a significant positive correlation between the mean fALFF value of the left DLPFC ROI and plasma KYN level in both patients with MDD and HC (Fig. 3).

Relationship between treatment response and treatment changes in plasma KYN level and left DLPFC activity

To investigate the relationship between escitalopram treatment response and treatment changes in KYN level in patients with MDD, Spearman's rank correlation test was performed. Treatment response was defined as % reduction {[(score at T1 − score at T2) / score at T1] *100} of HRSD. The change in KYN level was calculated as delta values (concentration level at T2 − concentration level at T1). Accordingly, our results showed a significant positive correlation ($\rho = 0.26$, $P = 0.035$) between the change in KYN level and treatment response (Fig. 4a). See Supplementary Table S3 for the results of correlation analysis between treatment changes in each metabolite and treatment response.

Likewise, a semipartial correlation analysis was performed to examine the relationship between escitalopram treatment response and changes in left DLPFC activity in MDD. The change in the left DLPFC activity was calculated as delta values (mean fALFF value of the left DLPFC ROI at T2 − mean fALFF value of the left DLPFC ROI at T1). Accordingly, our results showed a significant positive correlation between the change in the left DLPFC activity and treatment response ($r = 0.31$, $P = 0.013$) (Fig. 4b).

Next, we examined whether there was any relationship between changes over the course of treatment, but there was no significant correlation and no direct relationship between their changes over the course of treatment (Fig. 4c).

Comparison of baseline (T1) and post-treatment (T2) plasma KYN levels and left DLPFC activity in Responders

KYN level was significantly increased in Responders after escitalopram treatment (Fig. 5a). However, the mean fALFF value of the left DLPFC ROI did not significantly increase (Fig. 5b). By contrast, Nonresponders showed no significant change in KYN level or in the mean fALFF value of left DLPFC ROI.

Discussion

This study revealed that depression that responds well to escitalopram is characterized by lower plasma KYN level and resting-state regional brain activity in the left DLPFC. Both were significantly lower in the Responders than in both Nonresponders and HC. The degree of increase in both left DLPFC activity and plasma KYN level before and after treatment reflected a favorable treatment response, suggesting an association with a condition that improves with escitalopram. Furthermore, there was a significant correlation between individual differences in pretreatment regional brain activity of the left DLPFC and plasma KYN level in both MDD and HC, suggesting a relationship between them. However, there was no significant correlation between the degree of increase in KYN level before and after treatment and degree of increase in regional brain activity of the left DLPFC, indicating that the increase in KYN caused by escitalopram treatment does not immediately lead to an increase in regional brain activity of the left DLPFC.

KYN is a metabolite that crosses the blood–brain barrier[26,27], and in the central nervous system, approximately 60–80% of KYN is supplied from the periphery[28–30], and plasma KYN level and cerebrospinal fluid KYN level have been reported to be correlated to some extent[31,32]. Pretreatment plasma KYN/TRP and plasma KYN levels have been reported in previous studies to be negatively correlated with treatment response to antidepressants[9,32,33], and the present results are consistent with these findings. Kocki et al. (2012) reported that treatment with SSRIs increased KYN levels[34], and in this study, plasma KYN levels increased significantly before and after treatment. Moreover, the degree of increase in KYN level before and after treatment correlated positively with treatment response. Taken together, the decrease in plasma KYN level reflects part of the pathology of depression in response to escitalopram treatment, suggesting that escitalopram contributes to treatment response by correcting pathologies related to KYN pathway.

The left DLPFC plays an important role in cognitive functions, such as working memory and attention[35–38], and depression is known to reduce brain activity in the left DLPFC during rest and task execution[39–45]. This study revealed that patients with depression with low fALFF value of left DLPFC responded better to treatment with escitalopram.

As for the effects of SSRIs on the DLPFC, it has been reported that glucose metabolism in the left DLPFC increases in depression that responds well to 6 weeks of treatment with paroxetine[46] and that activation of the right DLPFC during performance of cognitive tasks is increased after 8 weeks of treatment with escitalopram, sertraline, and paroxetine[47]. Conversely, Ichikawa et al. (2020) reported that the resting-state functional connectivity of the left DLPFC and posterior cingulate cortex was still not in the same

direction of correlation as in healthy subjects after 6–8 weeks of antidepressant treatment but was in the same direction of correlation as in healthy subjects during the recovery period[48], and Okada et al. (2009) found that the activation of the left DLPFC during verbal fluency task remained lower in patients with depression in remission than in healthy subjects[44]. Furthermore, Takami et al. (2007) reported that left DLPFC activity did not differ from that of healthy subjects during recovery[49]. It is conceivable that the activity of the left DLPFC has not fully improved in terms of its function and activity at 6–8 weeks of antidepressant treatment and that it may recover later than the improvement in abnormal KYN metabolism, which may disrupt the correlation between the treatment change in KYN level and treatment change in fALFF value of the left DLPFC.

Although KYN itself does not directly excite or inhibit neural activity, the downstream metabolites of KYN, quinolinic acid and 3-HK, are known to have N-methyl-D-aspartate (NMDA) receptor agonist effects, while KYNA is known to have NMDA receptor antagonist effects[50]. The balance of both affects neural activity via NMDA receptors[51], and therefore, it may be related to brain functions, such as attention and working memory, in which NMDA receptors play an important role[52–55]. In the DLPFC, a brain region important for working memory and attention, NMDA receptors are known to be involved in attention and working memory by activating pyramidal cells to fire synapses persistently[56,57]. Therefore, it is presumed that the neural activity of DLPFC is indirectly influenced by the downstream metabolites of KYN on NMDA receptors. In the present study, we found a positive correlation between plasma KYN level and fALFF of the left DLPFC in healthy subjects and patients with depression. This suggests that blood KYN is supplied to the brain and has an indirect effect on regional brain activity in the left DLPFC, which may also be related to treatment response to escitalopram.

Our study has several limitations that should be mentioned. First, the sample size of the recruited participants was relatively small. Larger-scale studies are needed for reproduction and validation. Second, all participants are in the same ethnic group. To verify whether they are replicated across regions and species, studies that recruit participants from different regions are needed. Third, we did not distinguish between placebo responses; to evaluate the escitalopram treatment response, it is necessary to exclude patients who may respond to placebo in advance. Fourth, this study did not consider the potential effects such as diet, exercise, renal function, and smoking status, which may affect the concentrations of metabolites of the KYN pathway[58–60]. Fifth, since the purpose of this study was not to identify new metabolomic biomarkers, sample size was small for that purpose and multiple testing correction was not performed. Therefore, no conclusions can be drawn about metabolites other than KYN. Finally, the level of metabolites downstream of KYN pathway has not been measured. Downstream metabolites of KYN pathway, such as KYNA and quinolinic acid, have antagonist and agonist effects on NMDA receptors, as described above. Although there is a problem that these metabolites do not cross the blood–brain barrier, it seems necessary to add these measurements to the study to promote understanding of the pathophysiology.

Despite the abovementioned limitations, this is the first study to use metabolomics and fMRI to measure blood metabolites and regional brain activity related to the therapeutic effects of escitalopram in the same group of subjects and examines the relationship between the two. This study suggests that decreased KYN levels and decreased resting-state regional brain activity of the left DLPFC may be involved in the pathophysiology of depression in response to escitalopram and may serve as biomarkers for predicting treatment response. Furthermore, there was a correlation between individual differences in plasma KYN level and individual differences in regional brain activity of the left DLPFC, suggesting an association between the two, but the increase in KYN level was not correlated with the increase in the left DLPFC activity before and after treatment. Previous studies have suggested that the brain activity of the DLPFC is mediated by downstream metabolites of KYN, which has a neural effect on NMDA receptors, and that recovery of DLPFC activity and function is delayed compared to the improvement of depressive symptoms. Further studies are needed to clarify these complex relationships.

Declarations

Data availability

The original data for this study will not be made publicly available due to the involvement of patient data. It can be obtained upon request from the Department of Psychiatry and Neurosciences, Hiroshima University, Japan (OY, oy@hiroshima-u.ac.jp). The Institutional Review Board imposing these restrictions on our data is the Ethics Committee for Epidemiology of Hiroshima University (contact: Shoji Karatsu kasumi-kenkyu@office.hiroshima-u.ac.jp).

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Author contributions

T.K. conducted the data analysis and drafted the manuscript. T.K. G.O., E.I., Y.M., M.T., S.Y., A.Y., and Y.O. were involved in the experimental design and data collection. K.M., S.N., A.T., and T.O. measured the metabolites analysis. T.K., G.O., and Y.O. discussed the interpretation of the data. All authors discussed the results and commented on the final manuscript. All authors read and approved the final manuscript.

Competing interests

The authors declare no competing interests.

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Tables

Table 1. Demographics of study participants

Variable	HC (N = 36, 19 men)		MDD (N = 65, 31 men)				P-value*	
			Responders (N = 32, 16 men)		Nonresponders (N = 33, 15 men)		HC vs. MDD	Responders vs. Nonresponders
	Mean	SD	Mean	SD	Mean	SD		
Age (years)	41.3	11.0	38.5	9.8	41.9	11.6	0.62	0.21
BMI	22.4	3.5	22.2	3.6	23.5	5.1	0.59	0.25
Verbal IQ	111.4	8.4	109.2	10.4	112.1	10.9	0.71	0.27
Dose of escitalopram (mg)	-	-	12.5	4.3	13.3	4.7	-	0.34
HRSD17 (T1)	-	-	19.1	4.5	20.8	5.0	-	0.15
HRSD17 (T2)	-	-	5.4	3.5	15.2	4.3	-	0.00

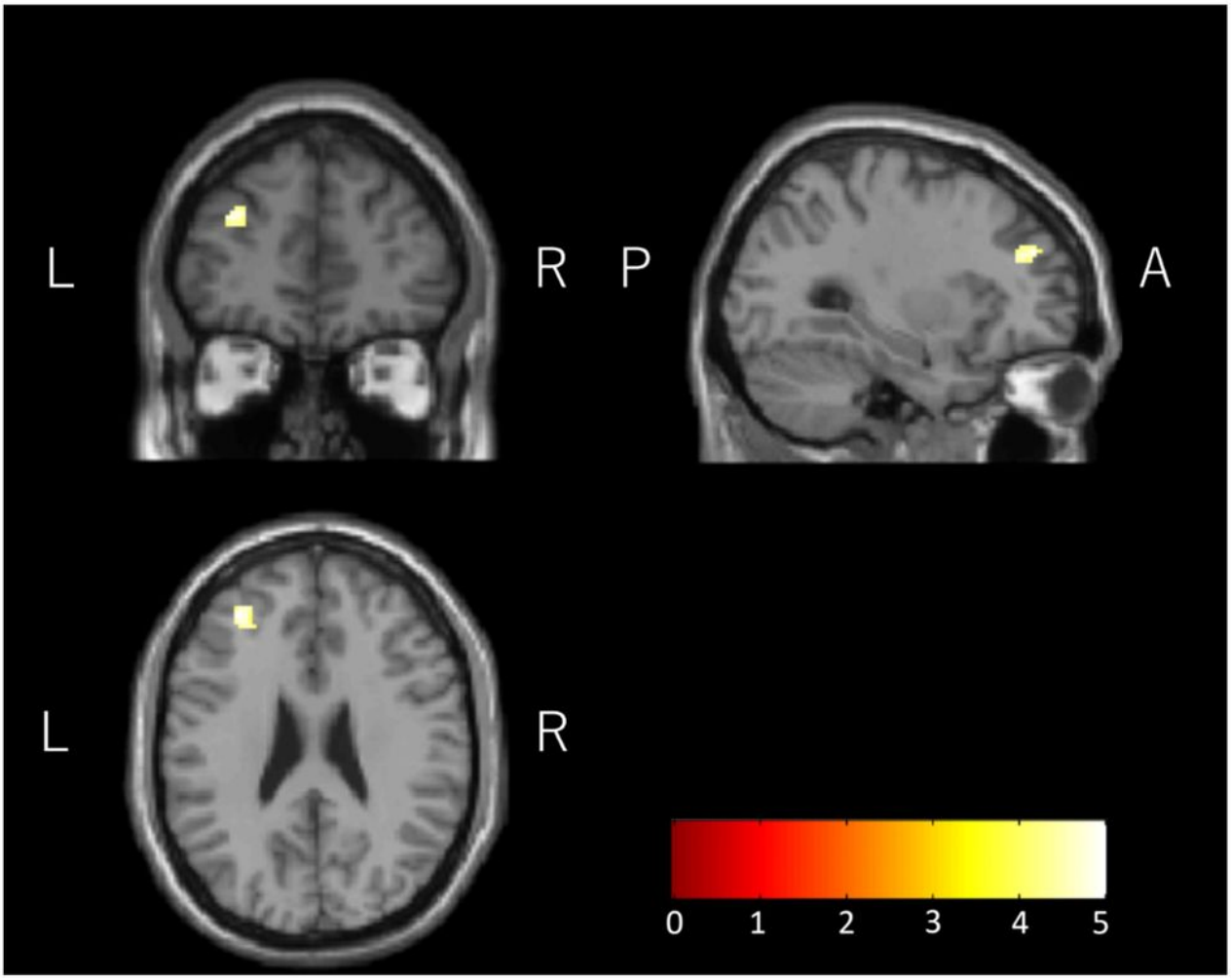
BMI, body mass index; Verbal IQ, The Japanese version of the Adult Reading Test (JART) was used.; HRSD17, Hamilton Rating Scale for Depression 17-item; SD, standard deviation; P-value*, two-sample t-test

Table 2. Comparison of Responders and Nonresponders in 34 metabolites.

Compound name	Responder (T1) N = 32		Nonresponders (T1) N = 33		P-value
	Mean	SD	Mean	SD	
Glycine	237.9	55.75	240.1	72.38	0.42
Alanine	420.2	115.7	425.9	105.6	0.84
γ-aminobutyric acid (GABA)	2.400	1.846	2.786	2.359	0.65
N,N-Dimethylglycine	3.318	1.376	3.537	1.260	0.44
Choline	18.43	4.958	17.17	3.974	0.19
Serine	124.0	25.51	126.0	28.28	0.98
Uracil	246.4	86.35	210.5	62.36	0.079
Creatinine	63.78	13.61	65.23	15.72	0.54
Proline	196.3	64.75	192.1	53.79	0.96
Valine	232.5	51.69	245.5	61.03	0.57
Betaine	52.16	16.22	54.93	16.05	0.97
Threonine	129.8	28.25	126.1	36.10	0.36
Cysteine	1.998	0.846	2.068	0.679	0.44
Thymine	67.42	97.72	45.88	20.01	0.40
Hydroxyproline	11.53	6.468	11.10	4.566	0.88
Creatine	36.82	17.08	39.98	17.21	0.40
Isoleucine	64.30	18.89	65.52	24.00	0.98
Leucine	114.3	36.10	118.3	35.57	0.68
Asparagine	44.54	6.538	45.03	11.66	0.71
Ornithine	55.81	16.61	60.05	17.82	0.35
Aspartic acid	5.846	2.571	5.597	2.026	0.88
Hypoxanthine	3.220	2.742	2.472	1.896	0.20
Anthranilic acid	1.084	0.402	1.049	0.288	0.80
Glutamine	620.8	97.20	641.0	91.13	0.47
Lysine	191.5	45.18	207.3	46.46	0.18
Glutamate	78.36	55.31	88.16	57.38	0.50
Methionine	20.22	4.461	19.52	7.298	0.17

Histidine	78.13	11.02	80.84	12.27	0.51
Phenylalanine	53.27	10.60	54.58	12.31	0.67
Arginine	69.41	21.20	70.04	21.87	0.96
Citrulline	29.69	6.538	31.47	7.840	0.45
Tyrosine	54.82	13.39	60.21	18.07	0.25
Tryptophan	53.46	12.06	55.01	10.89	0.57
Kynurenine	1.407	0.290	1.668	0.446	0.024

Figures



BA	Side	$P_{FWE-corr}$	k_E	Z	x	y	z
46	L	0.022	65	3.99	-32	42	26

Figure 1

Region where Responders (T1) were significantly lower than Nonresponders (T1) in cluster-level inference. Voxel level threshold were P (uncorrected) < 0.001, and cluster size threshold were P (FWE corrected) < 0.05. T-map of the activation greater or equal to 65 voxels was reported at the threshold P = 0.001, uncorrected. BA, Brodmann area; L, left; PFWE-corr, familywise error corrected P-value at cluster level; Z, Z value of the peak activation within the cluster. Coordinates for the peak voxel are listed as MNI

coordinates. A color scale represents t-value from 0 to 5. The figure was created using SPM12 (Welcome Department of Cognitive Neurology, London, UK) and Microsoft PowerPoint 2016 (Microsoft, Redmond, WA).

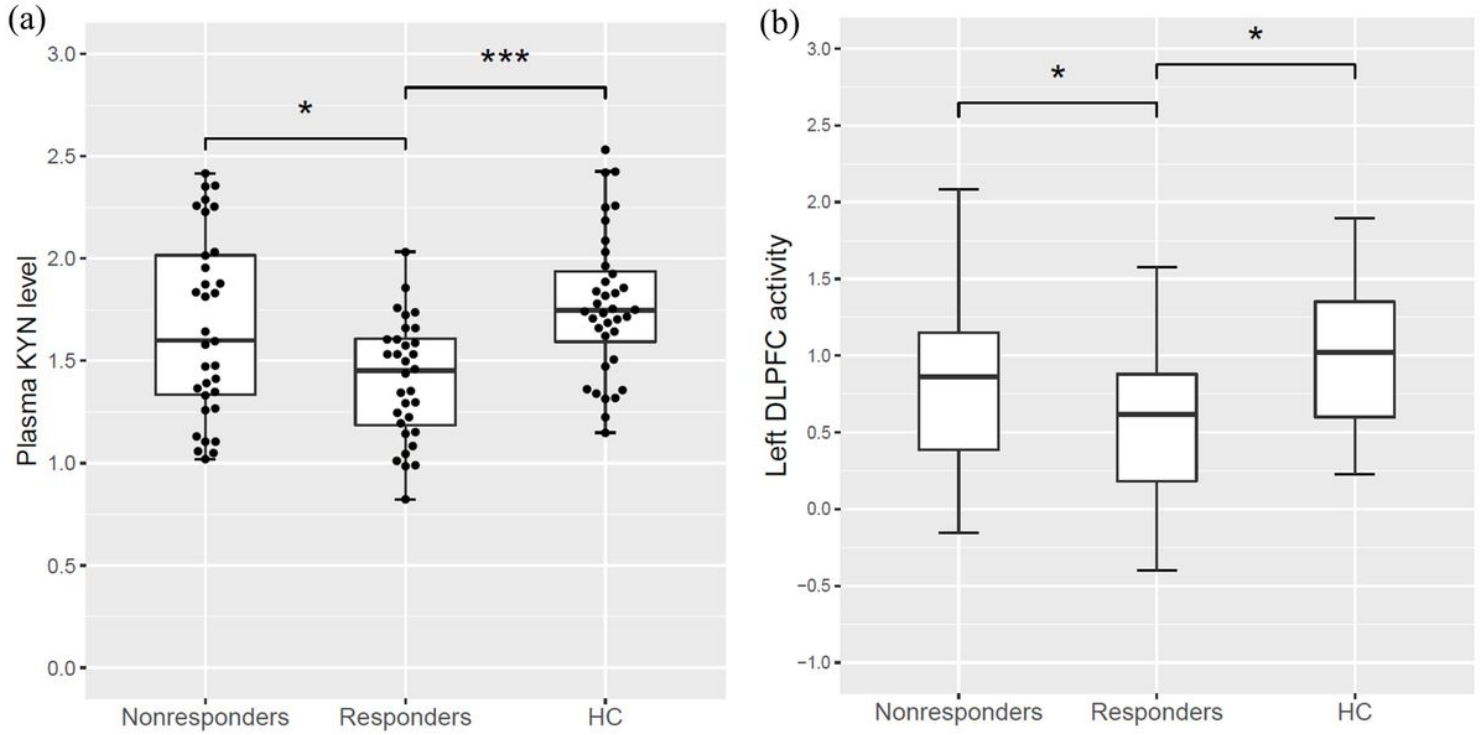


Figure 2

(a) Boxplots and beeswarm plots of plasma KYN levels in each group. Responders had significantly lower KYN than Nonresponders (P = 0.031) and HC (P = 0.00024). (b) Box plots of the mean fALFF of the left DLPFC ROI for each group. Responders had significantly lower left DLPFC activity than Nonresponders (P = 0.028) and HC (P = 0.015). *P < 0.05, ***P < 0.001, Dunn–Bonferroni post hoc test. The figures were created using the ggplot2 package for the R platform and Microsoft PowerPoint 2016 (Microsoft, Redmond, WA).

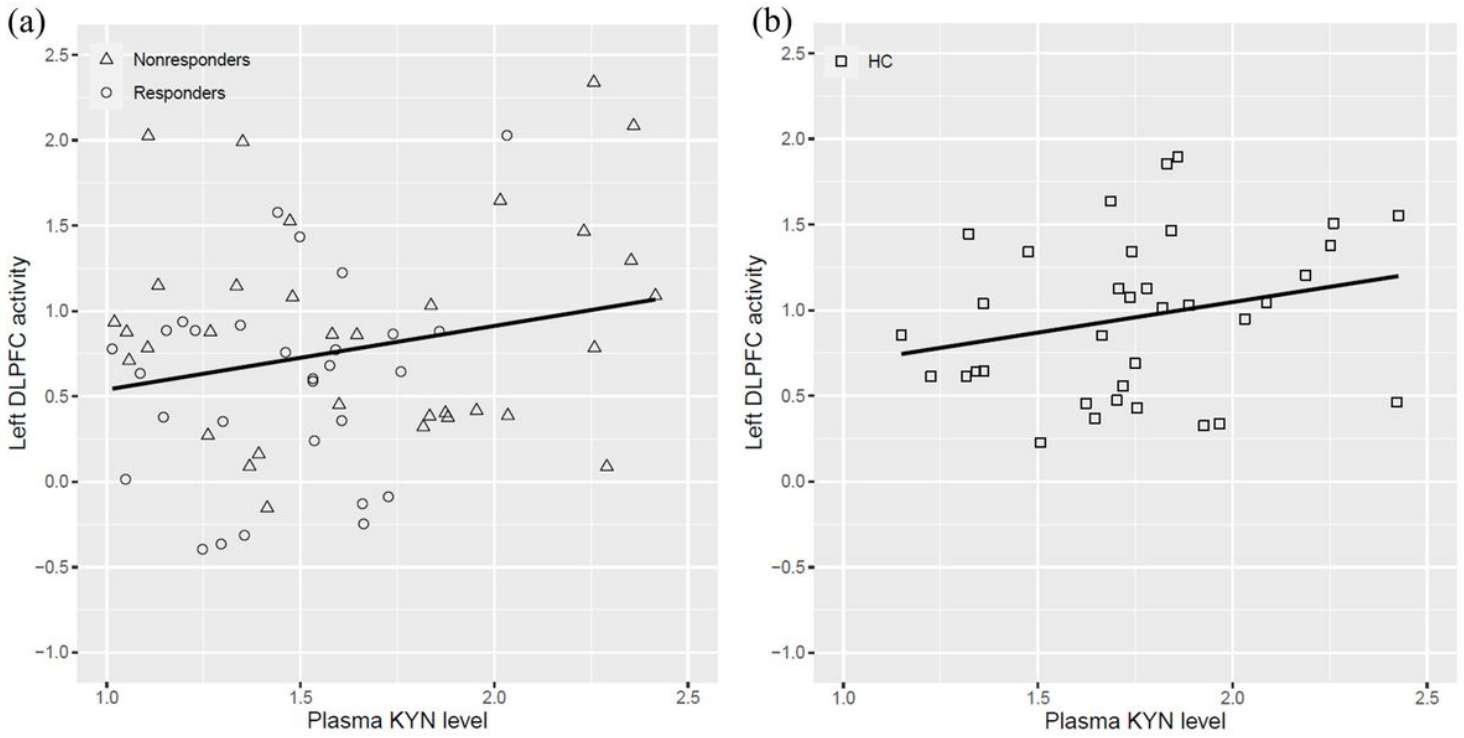


Figure 3

Mean fALFF value of left DLPFC ROI was correlated with plasma KYN level in MDD ((a) r = 0.28, P = 0.023) and HC ((b) r = 0.47, P = 0.005), respectively. Scatter plots illustrate these correlations. A semipartial correlation analysis was performed with each MRI scanner as a control variable. The figures were created using the ggplot2 package for the R platform and Microsoft PowerPoint 2016 (Microsoft, Redmond, WA).

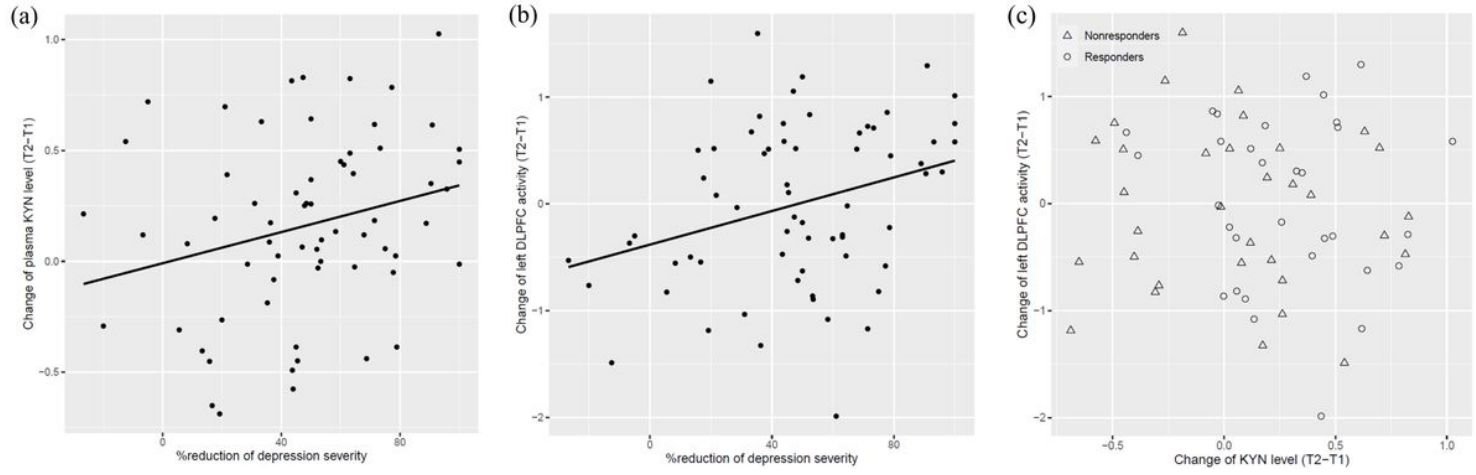


Figure 4

(a) Change in plasma KYN level (T2 − T1) showed a significant positive correlation with the %reduction of depression severity (r = 0.26, P = 0.035) using Spearman's rank correlation. (b) Change in the left DLPFC activity (T2 − T1) showed a significant positive correlation with the %reduction of depression severity (r = 0.31, P = 0.013) using semipartial correlation analysis with each MRI scanner used as a control variable.

(c) No significant correlation was observed between change in the left DLPFC activity (T2 − T1) and change in plasma KYN level (T2 − T1). Scatter plots illustrate these correlations. The figures were created using the ggplot2 package for the R platform and Microsoft PowerPoint 2016 (Microsoft, Redmond, WA).

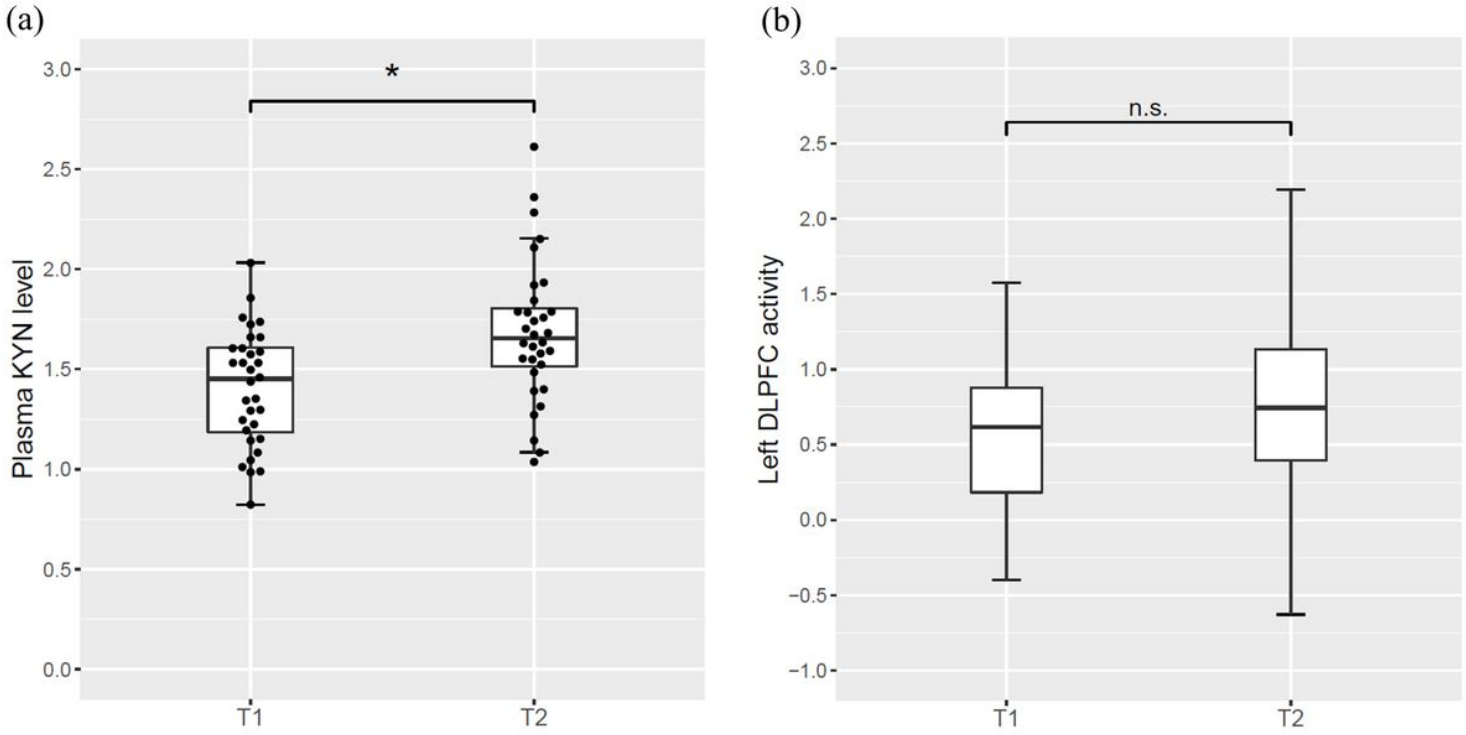


Figure 5

Comparison of baseline (T1) and post-treatment (T2) comparison in Responders. (a) Plasma KYN level was significantly increased (P = 0.00011). (b) Left DLPFC activity was not significantly increased (P = 0.15). *P < 0.05, Wilcoxon signed-rank test. n.s., not significant. The figures were created using the ggplot2 package for the R platform and Microsoft PowerPoint 2016 (Microsoft, Redmond, WA).

Supplementary Files

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