Psychological Medicine

cambridge.org/psm

Association of the plasma complement system with brain volume deficits in bipolar and major depressive disorders

Hua Yu[1,2,*], Peiyan Ni[3,*], Yang Tian[3], Liansheng Zhao[3], Mingli Li[3], Xiaojing Li[1,2], Wei Wei[1,2], Jinxue Wei[3], Xiangdong Du[4], Qiang Wang[3], Wanjun Guo[1,2], Wei Deng[1,2], Xiaohong Ma[3], Jeremy Coid[3] and Tao Li[1,2]

[1]Department of Neurobiology, Affiliated Mental Health Center & Hangzhou Seventh People's Hospital, Hangzhou, Zhejiang, China; [2]NHC and CAMS Key Laboratory of Medical Neurobiology, MOE Frontier Science Center for Brain Science and Brain-machine Integration, School of Brain Science and Brain Medicine, Zhejiang University, Hangzhou, Zhejiang, China; [3]The Psychiatric Laboratory and Mental Health Center, West China Hospital, Sichuan University, Chengdu, Sichuan, P R China and [4]Suzhou Psychiatry Hospital, Affiliated Guangji Hospital of Soochow University, Suzhou, 215137, Jiangsu, China

Original Article

*These authors contributed equally to this work.

Cite this article: Yu H *et al* (2023). Association of the plasma complement system with brain volume deficits in bipolar and major depressive disorders. *Psychological Medicine* **53**, 6102–6112. https://doi.org/10.1017/S0033291722003282

Received: 4 October 2021
Revised: 14 September 2022
Accepted: 30 September 2022
First published online: 26 October 2022

Key words:
Bipolar disorder; brain structure; complement component; inflammation; major depressive disorder

Author for correspondence:
Tao Li,
E-mail: litaozjusc@zju.edu.cn

Abstract

Background. Inflammation plays a crucial role in the pathogenesis of major depressive disorder (MDD) and bipolar disorder (BD). This study aimed to examine whether the dysregulation of complement components contributes to brain structural defects in patients with mood disorders.

Methods. A total of 52 BD patients, 35 MDD patients, and 53 controls were recruited. The human complement immunology assay was used to measure the levels of complement factors. Whole brain-based analysis was performed to investigate differences in gray matter volume (GMV) and cortical thickness (CT) among the BD, MDD, and control groups, and relationships were explored between neuroanatomical differences and levels of complement components.

Results. GMV in the medial orbital frontal cortex (mOFC) and middle cingulum was lower in both patient groups than in controls, while the CT of the left precentral gyrus and left superior frontal gyrus were affected differently in the two disorders. Concentrations of C1q, C4, factor B, factor H, and properdin were higher in both patient groups than in controls, while concentrations of C3, C4 and factor H were significantly higher in BD than in MDD. Concentrations of C1q, factor H, and properdin showed a significant negative correlation with GMV in the mOFC at the voxel-wise level.

Conclusions. BD and MDD are associated with shared and different alterations in levels of complement factors and structural impairment in the brain. Structural defects in mOFC may be associated with elevated levels of certain complement factors, providing insight into the shared neuro-inflammatory pathogenesis of mood disorders.

Introduction

Major depressive disorder (MDD) and bipolar disorder (BD) are severe psychiatric diseases with overlapping symptomatology (Niu et al., 2017). Despite the high global lifetime prevalence of MDD (13.2%) (Hasin, Goodwin, Stinson, & Grant, 2005) and bipolar spectrum disorder (2.4%) (Merikangas et al., 2007), the underlying pathophysiology is poorly understood (Niu et al., 2017). BD is characterized mainly by recurrent mood exacerbations of opposite polarity, ranging from major depressive to manic episodes (Gitlin, 2006), with depression the most frequent presentation (MacDonald et al., 2019). Current diagnostic systems, which are limited by the lack of reliable biomarkers, split BD and MDD into two independent diagnostic entities based on symptoms (Parker, 2014). However, genetic studies have established that BD and MDD share a similar polygenic component (Schulze et al., 2014). A large survey in the USA detected MDDs spanning the spectrum from MDD as least severe, BD-II as more severe and BD-I as most severe (Moreno et al., 2012). These results suggest that dimensional approaches may contribute to describing mood disorders more accurately (Moreno et al., 2012).

Activated peripheral inflammation plays a key role in the pathophysiology of BD and MDD (Bai et al., 2020; Solmi et al., 2021), although the severity of such inflammation in each disorder is unclear (Bulut, Yorguner, & Çarkaxhiu Bulut, 2021). Many patients with BD remain misdiagnosed with MDD for many years, leading to inadequate treatment, high healthcare costs, and poor prognosis involving, for example, mania and increased suicidality (Han, De Berardis, Fornaro, & Kim, 2019). Therefore, analyzing levels of inflammatory factors in BD and MDD may help differentiate the two disorders and improve treatment (Felger, 2017; Rosenblat et al., 2016).

Structural abnormalities in the brain have recently been identified as promising endophenotypes for affective disorders, so their analysis may improve the treatment and even prevention of these conditions (Opel et al., 2020). Since changes in gray matter volume (GMV) and cortical thickness (CT) can reflect degeneration, loss of neuroplasticity, or neurotoxicity (Lan et al., 2014), volume-based gray matter measures such as voxel-based morphometry (VBM) and CT-based measures such as surface-based morphometry (SBM) are widely used for structural analysis of the brain (Kong et al., 2015). For instance, previous work found that GMV in the medial orbital frontal cortex (mOFC) was reduced to a similar extent in BD and MDD patients (Wise et al., 2017). In a VBM-based study, both MDD and BD patients showed smaller GMV in the left anterior cingulate cortex (ACC) and right hippocampus than healthy controls (HCs), while GMV in the left superior frontal gyrus (SFG) and left ACC was lower in MDD than in BD patients (Chen et al., 2018b). In another study, BD patients showed significantly smaller CT in the frontal lobe than controls or patients with MDD, while both patient groups showed significantly smaller CT in the left inferior temporal cortex than controls (Niu et al., 2017). In contrast, Fung et al. (Fung et al. 2015) found no CT differences between MDD or BD patients. These findings suggest that the two mood disorders feature common and unique alterations in GMV and CT, for which the underlying neuropathology needs to be elucidated.

Clinical and experimental studies suggest that structural changes in the brains of patients with mood disorders are due to chronic low-grade inflammation (Leonard, 2018). The inflammatory response involves cytokine cascades, pathways of cellular immunity, increased levels of C-reactive proteins, and altered levels of complement components (Berk et al., 2011). BD patients are known to present more severe dysregulation of interleukin (IL)-6, tumor necrosis factor-α, and C-reactive protein than MDD patients (Bai et al., 2020; Chang et al., 2017), but whether plasma levels of complement factors differ between the two types of patients is unclear. The complement cascade, a key component of the inflammatory system, is a first-line defense against pathogens and waste materials. It also serves as a bridge between innate and acquired immune responses (Woo, Pouget, Zai, & Kennedy, 2020). There are three major pathways of complement activation: classical, mannan-binding lectin, and alternative (Pillai et al., 2019). All pathways converge on the generation of C3 convertase, which cleaves complement C3 into C3a and C3b (Pillai et al., 2019). Upon antigen-antibody binding, C1q is the primary initiator of the classical pathway, and it contributes to antigen recognition in adaptive and innate immunity (Reid, 2018). The complement components C1q, iC3b, C3, and C4 are involved in classical pathways (Kopczynska et al., 2019). C3b, generated by the classical or lectin pathways, activates or reinforces the alternative pathway, which involves the proteins properdin, factor B, factor H, and iC3b (Kopczynska et al., 2019). MDD patients show increased serum levels of C1q, C3, C4, and factor H (Shin et al., 2019; Yao & Li, 2020). The corresponding serum levels in BD patients are unclear: for example, different studies have reported increased or decreased C3 levels (Reginia et al., 2018; Yang et al., 2018). A comprehensive investigation of the complement cascades in BD and MDD is needed.

Changes in complement activity could underpin aberrant neurodevelopment or neuropathology by triggering excessive synaptic pruning, neurogenesis, and cell death (Gallego, Blanco, Morell, Lencz, & Malhotra, 2021). Increased *C4* gene expression was found in the dorsolateral prefrontal cortex and in the parietal cortex of schizophrenic patients (Rey et al., 2020), which implicates excessive complement activity in the development of schizophrenia and may help explain the reduced numbers of synapses in the brains of patients (Sekar et al., 2016). An analysis of 33 003 brains of healthy individuals and genetic data in the UK Biobank linked higher expression of the isoform *C4A* expression was significantly associated with reduced surface area for the transverse temporal sulcus (O'Connell et al., 2021). Animal studies have consistently shown that activation of C1q, C4, and C3 contribute to synapse loss in Alzheimer's disease, leading to gray matter loss and brain atrophy (Wu et al., 2019). However, few studies have explored the association between complement components and brain structure in mood disorders. Levels of *C3* mRNA were significantly higher in post-mortem prefrontal cortex of people who committed suicide than in the corresponding brain region of HCs (Crider et al., 2018). In addition, complement components have been found to enhance pro-inflammatory signaling, leading to increased IL-1β production (Zhang et al., 2007). These findings justify further exploration of the relationship between complement cascades and brain structure in mood disorders.

The present study builds upon the abovementioned evidence of shared and unique alterations in inflammatory biomarkers and brain structures in BD and MDD to comprehensively investigate complement cascade factors of all three complement pathways, as well as to explore correlations of their levels with changes in brain structure. We hypothesized that both disorders would be associated with increased levels of complement factors, with BD showing greater increases. We also hypothesized that the disorders would show shared and unique alterations in GMV and CT. Finally, we hypothesized that the disorders would share associations between complement factors and structural alterations in the brain, which may help clarify how the two mood disorders arise through shared pathways (Lanz et al., 2019).

Methods

Participants

Patients with BD and MDD were recruited from the inpatient and outpatient departments of West China Hospital of Sichuan University. BD or MDD was diagnosed using the structured clinical interview for DSM-IV Axis-I disorders (Patient Version) (First, Spitzer, Gibbon, & Williams, 2002). All subjects were between 16 and 55 years old and were right-handed Han Chinese who were physically and neurologically healthy. Their demographic characteristics were recorded, including disease duration, treatment history, illness stage, age, sex, education level, and body mass index (BMI). Manic and depressive symptoms were assessed using the Young Mania Rating Scale (Young, Biggs, Ziegler, & Meyer, 1978) and the Hamilton Rating Scale for Depression (Hamilton, 1960).

Patients with concurrent neurological illness, intellectual disability, cardiovascular disease, schizophrenia, anxiety disorder, alcohol or drug abuse, and history of loss of consciousness were excluded from the study. None of the patients had received electroconvulsive therapy before participation.

For comparison, demographically matched HCs were recruited from the local community through advertising. HCs were included if they (1) were 16–55 years old; (2) did not meet the diagnostic criteria of the structured clinical interview for DSM-IV (Non-patient Version) (First et al., 2002); (3) had no current or past significant medical or physical disease, such as diabetes, thyroid diseases, hypercholesteremia, liver disease,

https://doi.org/10.1017/S0033291722003282 Published online by Cambridge University Press

epilepsy, stroke, or systemic lupus erythematosus; and (4) had no history of psychiatric illness among first-degree relatives.

In order to exclude individuals with any intellectual disability, we assessed the intelligence quotient (IQ), verbal IQ, and performance IQ of all participants using the seven-subtest short form (information, arithmetic, digital symbol, digital span test, block design, picture completion, and similarities) on the revised Wechsler Adult Intelligence Scale in Chinese (Wechsler, 1981). The IQ scores of all participants were over 70.

All participants provided written informed consent after the study procedures were explained. The study was approved by the Institutional Review Board of West China Hospital of Sichuan University. Additional details about inclusion and exclusion criteria are provided in the Supplementary Materials.

Measurement of complement component levels

Blood samples without fasting were collected by venipuncture between 4:00 p.m. and 4:30 p.m. using ethylenediaminetetraacetic acid as an anti-coagulant. Peripheral blood mononuclear cells were removed by refrigerated centrifugation at 1000 ***g*** for 10 min, and the separated plasma was immediately divided into 0.5-mL aliquots and stored at −80 °C. Plasma factors were evaluated using MILLIPLEX® MAP kits (Merck KGaA, Darmstadt, Germany). The human complement panel 2-immunology multiplex assay (HCMP2MAG-19 K, Merck Millipore, Billerica, MA, USA) was used to measure the levels of C1q, C3, C3b/iC3b, C4, factor B, properdin and factor H, according to the manufacturer's instructions. Data were analyzed using a FLEXMAP 3D® instrument (Luminex, Merck Millipore) operated with xPONENT® software (version 4.0, Luminex). Standard curves were generated using standard samples in the multiplex assay kit. Median fluorescent intensity data were analyzed using a weighted 5-parameter logistic method to calculate the factor concentrations.

The manufacturer-specified sensitivities of the assays of complement components were as follows: C1q, 0.048 ng/mL; C3, 0.120 ng/mL; C3b or iC3b, 3.639 ng/mL; C4, 0.191 ng/mL; factor B, 0.024 ng/mL; factor H, 0.135 ng/mL; and properdin, 0.0032 ng/mL. The intra-assay coefficient of variation was 10% for the panel.

Image acquisition

Magnetic resonance imaging (MRI) was performed on a 3.0-T MR scanner (Achieva; Philips, Amsterdam, The Netherlands) using an eight-channel phased-array head coil. High-resolution T1 images were acquired using a 3D magnetization-prepared rapid gradient-echo sequence with the following parameters: repetition time, 8.37 ms; echo time, 3.88 ms; flip angle, 7°; in-plane matrix resolution, 256 × 256; field of view, 24 × 24 cm^2; and number of slices, 188. During MRI, none of the participants showed more than 2 mm displacement in the *x*, *y*, or *z* direction or more than 2° angular motion.

VBM and SBM

VBM and SBM analyses were conducted using the Computational Anatomy Toolbox 12 (CAT12, version 12.5, Jena University Hospital, Jena, Germany, http://dbm.neuro.uni-jena.de/cat/), which is an extension of Statistical Parametric Mapping software (University College London, London, UK, http://www.fil.ion.ucl.ac.uk/spm/software/spm12/), according to the developers' instructions (http://dbm.neuro.uni-jena.de/cat12/CAT12-Manual.pdf).

T1 MRI scans of all subjects were registered to the Montreal Neurological Institute space. Whole-brain structural data were then segmented into white matter, gray matter, and cerebrospinal fluid, and bias was corrected in order to remove intensity non-uniformities. The images of segmented gray matter were preserved to allow assessment of volume changes based on spatial registration (Hong et al., 2020). The total intracranial volume (TIV) of each subject was calculated and used as a covariate in subsequent statistical analyses.

Normalized gray matter images were smoothed using a Gaussian filter with a full-width half-maximum (FWHM) of 8 mm. The CT of the left and right hemispheres was automatically estimated by CAT12 using the projection-based thickness method, and the corresponding images were smoothed with a 15-mm FWHM Gaussian kernel (Zhang et al., 2017b).

Statistical analysis

Statistical analyses were carried out using SPSS 24.0 (IBM, Chicago, IL, USA). Differences in demographic and clinical data were assessed for significance using one-way analysis of variance for continuous variables and Fisher's chi-squared test for categorical variables. Data normality was evaluated by visual inspection and the Kolmogorov–Smirnov test. The concentrations of all complement components were $\log_{10}$-transformed prior to analysis in order to normalize their distributions. Complement factors were compared using analysis of covariance (ANCOVA) while controlling for age, sex, education level, and BMI. Clinical parameters between the two patient groups were compared using the two-samples *t* test. To explore correlations of complement components with clinical parameters, partial correlation analysis was conducted after controlling for demographic parameters. To explore effects of drugs on complement factors, we performed simple comparisons between drug-treated or untreated groups (online Supplementary Methods). Differences associated with $p < 0.05$ were considered statistically significant.

Image analysis

Voxel-wise GMV and vertex-wise CT differences among the three groups were investigated using ANCOVA, with age, sex, education level and BMI treated as covariates. For GMV analysis, all voxels with GMV probability value < 0.1 (absolute threshold; range, 0–1) were excluded in order to avoid possible edge effects around the margin between different tissue types. While controlling for age, sex, education level, and BMI, statistical maps were created to identify potential linear correlations between plasma levels of complement components and whole-brain GMV or CT in BD patients, MDD patients and HCs considered separately as three groups or together as one large group (Tseng et al., 2021).

We also conducted linear modeling to test whether observed relationships differed among the groups. Group and complement factor level were entered as regressors of interest, and contrasts for group by complement factor level interaction (Group × complement factor > 0 and Group × complement factor < 0) were generated, while age, sex, education and BMI were entered as regressors of non-interest (Tseng et al., 2021). TIV was also controlled as a covariate in GMV analysis. Significance was defined as $p < 0.05$ after correcting for family-wise error rate at the cluster level with conservative voxel-wise $p < 0.0005$.

https://doi.org/10.1017/S0033291722003282 Published online by Cambridge University Press

Table 1. Demographic and clinical characteristics of BD, MDD and HC

	BD M (s.d.) (n = 52)	MDD M (s.d.) (n = 35)	HC M (s.d.) (n = 53)	ANOVA			
				F/χ^2	df	p, 2-tail	Post hoc test
Age (Years)	28.92 (11.45)	30.26 (10.26)	27.47 (8.98)	0.793	2, 139	0.46	–
Education (years)	13.12 (3.23)	12.49 (4.15)	15.15 (3.60)	6.86	2, 139	**0.001****	BD, MDD < HC
BMI	23.10 (3.86)	21.10 (2.95)	21.33 (3.23)	4.78	2, 139	**0.010***	BD > MDD, HC
Gender (male /female)	21 (M)/ 31 (F)	15 (M)/ 20 (F)	22 (M)/ 31 (F)	0.053	2, 139	0.97	–
Smoking (yes/no)	8 (yes)/38(no)	7 (yes)/28 (no)	5 (yes)/44 (no)	1.73	2, 129	0.422	–
Cognitive function							
VIQ	104.92 (21.97)	98.32 (31.16)	112.48 (13.45)	1.48	2, 139	0.23	–
PIQ	96.35 (22.29)	94.23 (30.32)	108.35 (13.05)	1.90	2, 139	0.15	–
WAIS IQ	101.63 (21.94)	96.74 (30.87)	111.65 (13.34)	1.71	2, 139	0.19	–
Complement component							
Log_{10} C1q	2.75 (0.42)	2.59 (0.29)	2.30 (0.17)	22.47	2, 137	**0.000*****	BD, MDD > HC
Log_{10} C3	2.24 (0.35)	2.00 (0.24)	1.97 (0.23)	10.30	2, 138	**0.000*****	BD > MDD, HC
Log_{10} C4	2.99 (0.18)	2.89 (0.12)	2.78 (0.10)	24.72	2, 138	**0.000*****	BD > MDD > HC
Log_{10} Factor B	2.96 (0.31)	2.87 (0.25)	2.67 (0.14)	16.20	2, 133	**0.000*****	BD, MDD > HC
Log_{10} Factor H	3.12 (0.33)	2.97 (0.19)	2.79 (0.12)	21.59	2, 138	**0.000*****	BD > MDD > HC
Log_{10} Properdin	2.10 (0.32)	2.02 (0.22)	1.79 (0.15)	18.52	2, 136	**0.000*****	BD, MDD > HC
Clinical parameters							
HAMD total score	10.73 (7.55)	21.83 (5.26)	–	−7.55	85	**0.000*****	BD < MDD
YMRS total score	10.07 (11.31)	–	–	–	–	–	–
GAF	54.11 (13.63)	53.10 (10.05)	–	0.18	73	0.73	–
Age first episode (years)	23.85 (10.00)	27.21 (11.20)	–	−1.41	78	0.16	–
Duration of illness (months)	64.60 (59.11)	39.20 (54.97)	–	2.02	85	**0.046***	BD > MDD
Depressive episodes (n)	2.24 (1.46)	1.77 (1.07)	–	1.50	70	0.14	–
Manic/hypomanic episodes (n)	1.98 (1.62)	–	–	–	–	–	–
Mood stabilizers (n of users)	30/52 (57.69%)	0/35	–	30.82	1	**0.000*****	BD > MDD
Antipsychotics (n of users)	31/52 (59.62%)	3/35 (8.57%)	–	22.89	1	**0.000*****	BD > MDD
Antidepressants (n of users)	24/52 (46.15%)	18/35 (51.43%)	–	0.23	1	0.67	–

1.BD, bipolar disorder; MDD, major depressive disorder; HC, healthy controls; WAIS, Wechsler Adult Intelligence Scale Vocabulary Score (37 items); IQ, intelligence quotient; VIQ, verbal intelligence quotient; PIQ, performance intelligence quotient; TMT-A, Trail Making Test Parts A; TMT-B, Trail Making Test Parts B; DSST, digit symbol substitution test; C1q, Complement component 1q; C3, Complement component 3; C4, Complement component 4; Factor B, Complement factor B; Factor H, Complement factor B; HAMD, Hamilton depression rating scale (17 items); YMRS, Young Mania Rating Scale (11 items); GAF, Global Assessment of Functioning; n, number; s.d., standard variance; M, male; F, female. For the comparison of complement factors, age, gender, education years and BMI were co-variated out.
2.*p < 0.05, **p < 0.01, ***p < 0.001. Bold represents statistically significant results (p < 0.05).

To further analyze the relationship between GMV or CT and other variables, GMV and CT were calculated for various brain regions defined according to the anatomical automatic labeling atlas (AAL) or the Desikan–Killiany (DK40) cortical atlas. These regions were matched to brain areas that differed significantly between patients and controls or between the two types of patients, and the regions were separately matched to neuroanatomical alterations that correlated significantly with levels of complement components in whole-brain analysis. The values were then subjected to partial correlation analysis using SPSS 24.0, with the significance level defined as $p < 0.05$. During this partial correlation analysis outside SPM, we did not correct for multiple comparisons because the brain areas had survived multiple-comparison corrections in whole-brain analysis.

Results

Demographic and clinical characteristics of patients and HCs

A total of 140 subjects, including 52 BD patients, 35 MDD patients, and 53 age- and sex-matched controls were enrolled in the study (Table 1). Across the three groups, there were no significant differences in sex or smoking status. All three groups had similar estimated average IQ scores. Between BD and MDD patients, there were no significant differences in age at onset, number of depressive episodes, general functioning, or anti-depressant usage. Both patient groups had a lower education level than HCs, while BD patients had a higher BMI than MDD patients and HCs. The BD group had longer illness duration than the MDD group. Although BD patients had lower Hamilton depression total scores

https://doi.org/10.1017/S0033291722003282 Published online by Cambridge University Press

Table 2. Significant difference of GMV and CT between BD, MDD, and HC groups

Model	Cluster size (voxels/vertexs)	*F*/*t* value	Corrected *p*	MNI coordinates			BA	Anatomical label
				X	*Y*	*Z*		
Gray matter volume								
Three group comparison	1234	18.52	**0.000*** **	−8/5/2	−26/−18/−12	39/48/56	BA31/BA6/BA24	Middle cingulum
	1582	13.91	**0.000*** **	−12/−2/−3	53/33/42	−6/−15/−12	BA10/BA11	Frontal medial orbital cortex
	598	12.56	**0.016***	30/6/20	−30/−35/−26	56/72/65	BA4/BA6	Right precentral gyrus
Post-hoc: BD < HC	2040	5.84	**0.000*** **	−8/5/−15	−26/−18/−38	38/48/38	BA31	Middle cingulum
	2202	4.80	**0.000*** **	−2/−12/−9	33/53/47	−15/−8/−12	BA10/BA11	Frontal medial orbital cortex
Post-hoc: MDD < HC	1306	4.98	**0.002****	30/6/20	−30/−35/−24	56/72/66	BA4/BA6	Right precentral gyrus
	656	4.66	**0.027***	−6/8	−24/−17	42/51	BA31	Middle cingulum
	1190	4.59	**0.002****	−14/−14/−6	50/41/44	−8/−5/−11	BA10/BA11	Frontal medial orbital cortex
Cortical thickness								
Three group comparison	104	12.83	**0.014***	−36/−28	−16/−17	67/70	BA6	Left precentral gyrus
	85	10.47	**0.032***	−8	33	40	BA8	Left superior frontal gyrus
Post-hoc: BD < HC	115	4.29	**0.018***	−8	32	41	BA8	Left superior frontal gyrus
Post-hoc: MDD < HC	421	4.93	**0.000*** **	−36/−28/−21	−16/−17/−5	67/70/62	BA6	Left precentral gyrus

1. BD, bipolar disorder; MDD, major depressive disorder; HC, healthy controls; BA, Brodmann area; MNI, Montreal Neurological Institute.
2. Age, sex and education years were co-variated out in GMV and CT analysis. The intracranial volume was additionally controlled in GMV analysis. Voxel-wise threshold was set at $p < 0.0005$. Significance level was set at $p < 0.05$, cluster wise family wise error corrected. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$. Bold represents statistically significant results.

than the MDD group, they were treated more often with mood stabilizers and/or antipsychotics.

Levels of complement components

Levels of C1q were under the limit of detection in 3 subjects (2 BD patients, 1 HC). Levels of C3, C4 and factor H were under the limit of detection in 2 subjects (1 BD patient, 1 HC). Levels of factor B were under the limit of detection in 6 subjects (5 BD patients, 1 HC). Levels of properdin were under the limit of detection in 4 subjects (3 BD patients, 1 HC). C3b was not detected in more than half of subjects, so it was excluded from the analysis.

The $\log_{10}$-transformed concentrations of all complement components were higher in the BD group than in HCs. The same trend was also observed in the MDD group, except for C3. Levels of C3, C4, and factor H were higher in patients with BD than in those with MDD, after controlling for age, sex, education level, and BMI (Table 1). No other significant correlations were found between levels of complement components and clinical parameters in either patient group (online Supplementary Table S1). Within each group of patients, levels of complement components did not differ significantly between patients who received certain drug treatments or not (online Supplementary Table S3a–S3d).

Altered GMV and CT in BD and MDD patients

Voxel-based comparison of GMV in the mOFC, middle cingulum, and right precentral gyrus using age, sex, education years, and intracranial volume as covariates indicated significant differences among the three groups (Table 2 and Fig. 1a). Post-hoc analysis identified brain volume deficits in the mOFC and middle cingulum common to both patient groups (Table 2 and Figs 1b–c), and MDD patients additionally showed lower GMV in the left precentral gyrus than HCs (Table 2 and Fig. 1c).

Vertex-based comparisons of CT in the left precentral gyrus and left SFG revealed significant differences among the three groups after controlling for age, sex, and education level (Table 2 and Fig. 1d). CT of the left precentral gyrus was significantly smaller in the MDD group than in HCs, while CT in the left SFG was significantly smaller in the BD group than in HCs (Table 2 and Fig. 1d). However, no significant differences were observed in GMV or CT between BD and MDD patients after

https://doi.org/10.1017/S0033291722003282 Published online by Cambridge University Press

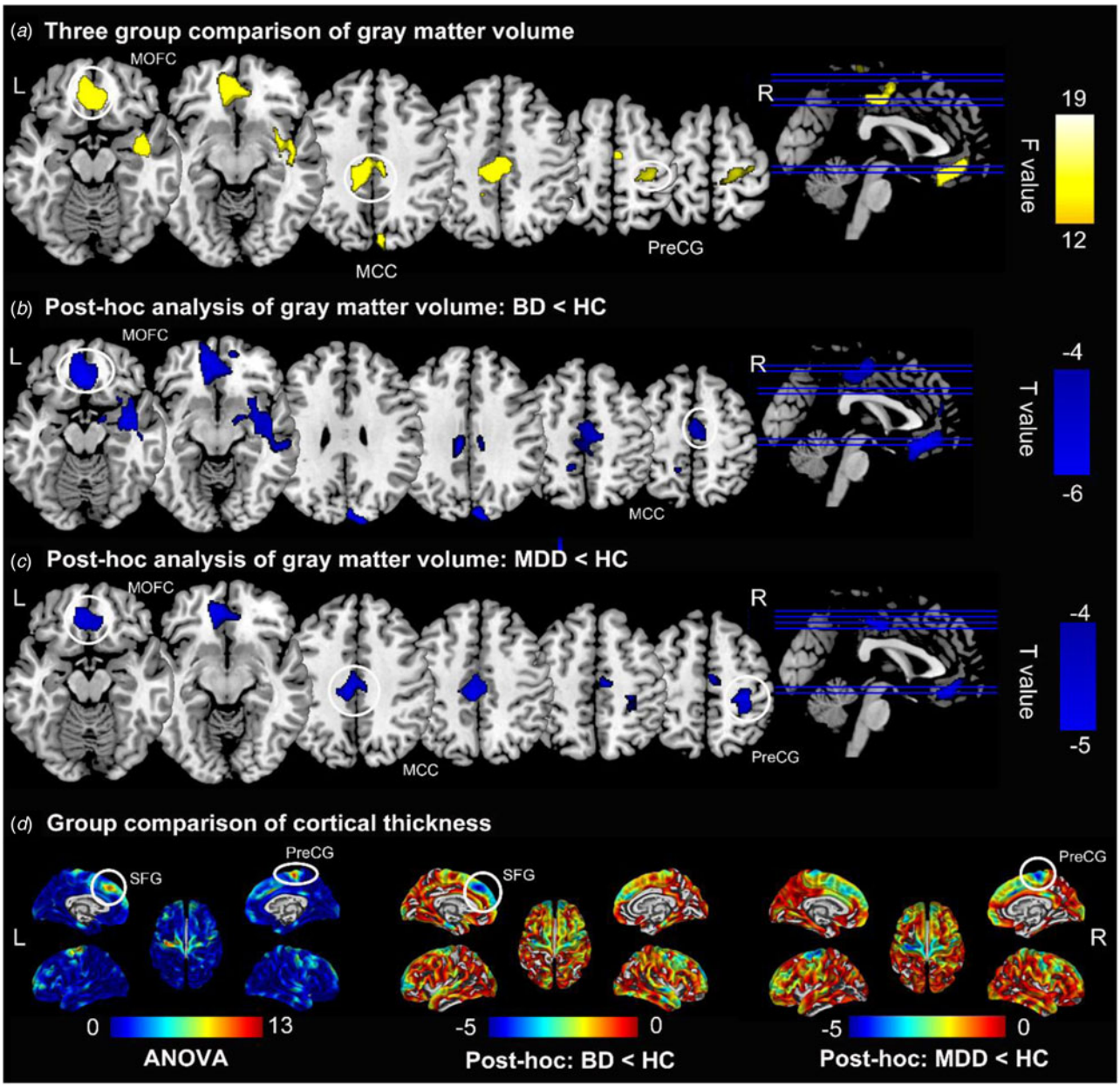

Fig. 1. GMV and CT differences between participants with BD and MDD, compared to HCs. (a). Three group comparisons: The GMV of the middle cingulum, mOFC, right superior temporal gyrus, right pre/post central gyrus and right occipital gyrus differed significantly among the three groups (color in yellow); (b). Compared with controls, BD patients showed decreased GMV in middle cingulum, right superior temporal gyrus, medial frontal orbital cortex and right superior occipital cortex (color in blue); (c). Compared with controls, the MDD patients showed decreased GMV in right pre/postcentral gyrus, middle cingulum and mOFC (color in blue); (d). The figure in the left showed three group comparison results of the CT, including bilateral precentral gyrus and left SFG (color in red); the figure in the middle showed that CT in the left SFG was decreased in bipolar patients group compared with controls (cold color); the figure in the right showed that compared with controls, MDD patients showed decreased CT in bilateral precentral gyrus (cold color). (All the results were co-variated out for age, gender and education years, and the TIV was additionally co-variated out for GMV analysis, voxel threshold significance was set at $p < 0.0005$, cluster level was set at $p < 0.05$, family wise error corrected). BD, bipolar disorder; MDD, major depressive disorder; HC, healthy control.

controlling for individual variations in intracranial volume (for GMV), age, sex, and education level.

Correlation of plasma levels of complement components with whole-brain GMV or CT

Whole-brain correlation analysis across all subjects after controlling for age, sex, education level, BMI, and TIV revealed a negative association between GMV in the mOFC and $\log_{10}$-transformed levels of C1q, factor H, and properdin (Table 3 and Fig. 2). However, whole-brain correlation analysis did not reveal significant correlations of CT with plasma levels of complement components, whether among all subjects or between different groups. Nevertheless, CT of several brain areas correlated significantly with several complement factors in SPSS (online Supplementary Table S2).

https://doi.org/10.1017/S0033291722003282 Published online by Cambridge University Press

Table 3. Whole brain-based negative correlation analysis of GMV with complement component factors

Samples	Complement component	Lateral	Cluster	*p* cluster	BA	Anatomical label	*t*	MNI peak coordinate		
								X	*Y*	*Z*
All subjects	Log_{10} C1q	Bilateral	1343	**0.001****	11	Medial orbital frontal cortex	4.46/4.27/3.93	−2/5/−8	32/41/43	−21/−18/−15
	Log_{10} FH	Bilateral	903	**0.008****	11	Medial orbital frontal cortex	4.32/4.18/3.59	0/5/−9	33/42/54	−23/−18/−14
	Log_{10} Properdin	Bilateral	1456	**0.001****	10/11	Medial orbital frontal cortex	4.20/4.00/3.93	0/0/−6	35/42/48	−21/−17/−9

1. Age, sex, education years and body mass index were controlled. BA, Brodmann area; MNI, Montreal Neurological Institute.
2. Voxel-wise threshold was set at $p < 0.0005$. Significance level was set at $p < 0.05$, cluster wise family wise error corrected. $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$. Bold represents statistically significant results. Bold represents statistically significant results.

Discussion

Consistent with previous findings, we found that complement component levels were significantly higher in both patient groups than in HCs, suggesting that both BD and MDD are associated with increased levels of inflammatory biomarkers (Reginia et al., 2018; Wei et al., 2018). The log_{10}-transformed levels of C3, C4, and factor H were significantly higher in BD than MDD patients, even after controlling for age, sex, years of education, and BMI. These results suggest that patients with BD might involve more severe inflammatory response than MDD. Group comparison of brain structures identified structural alterations that were shared and different between BD and MDD patients. Compared to HCs, both patient groups showed reduced GMV in the middle cingulum and mOFC. However, MDD patients showed lower GMV in the right precentral gyrus and smaller CT in the left precentral gyrus than HCs, while the BD group showed smaller CT in the left SFG than HCs. Finally, we found that plasma levels of C1q, factor H, and properdin were negatively associated with GMV in the bilateral mOFC across all subjects, which further supports the idea that BD and MDD share elements of their etiology (Lanz et al., 2019). To the best of our knowledge, this is the first HC-controlled study examining relationships between brain structure and complement components in mood disorders.

Our study associated BD and MDD with increased levels of complement components involved in the classical pathway (C1q and C4) and alternative pathway (factor B, factor H and properdin). C1q initiates the classical pathway upon antigen-antibody binding (Akcan, Karabulut, İsmail Küçükali, Çakır, & Tüzün, 2018). Patients with chronic BD show significant elevation of C1q mRNA expression in peripheral blood (Akcan et al., 2018). C3, C4 and factor B, whose levels in BD are much better studied than levels of C1q, are elevated in the peripheral blood of patients with manic or psychotic BD (Fontana et al., 1980; Wadee et al., 2002). Patients with chronic BD show elevated levels of mRNAs encoding C4 and factor B in peripheral blood mononuclear cells (Akcan et al., 2018). Our observation of increased C1q and factor H in MDD patients is consistent with previous work (Shin et al., 2019; Yao & Li, 2020). Other studies have shown that depressed subjects have significantly higher plasma levels of C4 than normal controls (Kronfol & House, 1989; Maes et al., 1997). Consistent with our observation that properdin, a key regulator of the alternative complement pathway (Chen, Cortes, & Ferreira, 2018a), is increased in patients with BD or MDD, previous studies have reported dysregulation of the alternative pathway in neurological and neuropsychiatric disorders such as schizophrenia and epilepsy (Kopczynska et al., 2018; Zhang, Lv, Fan, Tang, & Yi, 2017a). Taken together, our results are consistent with the idea that inflammatory markers are associated with mood disorders such as BD and MDD, without being specific to a particular disorder (Swardfager, Rosenblat, Benlamri, & McIntyre, 2016).

We found that plasma levels of C3 were significantly higher in BD patients, but not MDD patients, than in HCs. Our results are consistent with other studies in which no difference in C3 was found between MDD and controls (Berk, Wadee, Kuschke, & O'Neill-Kerr, 1997). In fact, we found that BD patients had higher levels of C3, C4, and factor H than MDD patients. While we are unaware of previous studies directly comparing levels of these complement components between the two patient groups, our results are consistent with a previous report that BD patients have higher levels of pro-inflammatory cytokines than MDD

https://doi.org/10.1017/S0033291722003282 Published online by Cambridge University Press

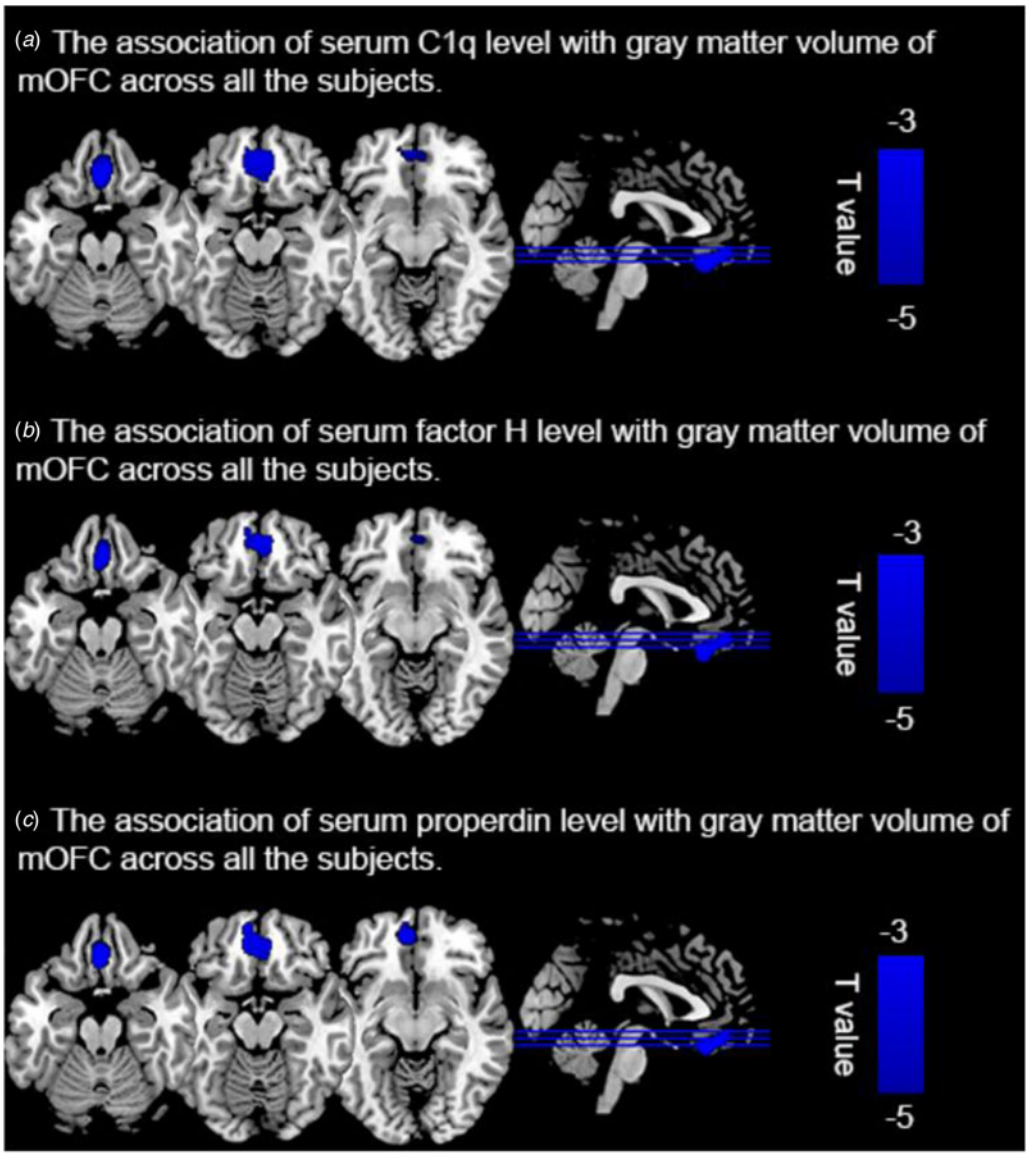

Fig. 2. (a) Figure shows significant negative correlation between log_{10}-transformed C1q and volume of medial orbital prefrontal cortex (cold color) across all the subjects. (b) Figure shows significant negative correlation of the log_{10}-transformed factor H and GMV in medial orbital prefrontal cortex (cold color) across all the subjects. (c). Figure shows significant negative correlation of the log_{10}-transformed properdin and GMV in medial orbital prefrontal cortex (cold color) across all the subjects. For the correlation analysis, age, gender, education years, TIV and body mass index were set as the covariates. Significance was set at a threshold with a peak-level uncorrected $p < 0.0005$, with a family-wise error rate (FWE)-corrected cluster level of $p < 0.05$). MOFC: medial orbital frontal cortex.

patients (Bai et al., 2020). Nevertheless, our results should be verified in future comparisons of complement components between the two disorders. If verified, our findings suggest that BD involves more severe inflammatory response than MDD.

Our findings of shared structural alterations in the brains of BD and MDD patients are consistent with the results of a recent meta-analysis, where the volume of the ventromedial prefrontal cortex was found to be lower in both patient groups, suggesting a volume deficit common to the two disorders (Wise et al., 2017). A previous study also reported that middle cingulum volume was significantly smaller in MDD patients who did not respond to treatment than in HCs or in patients who did respond to treatment (Batail et al., 2020). Although no such reduction has been reported for BD patients, our results suggest that altered middle cingulate gyrus volume may underlie brain pathology in both disorders.

We identified structural impairments that may be specific to BD or MDD. Our MDD patients had significantly smaller GMV in the precentral and postcentral gyri, as well as smaller CT in the left precentral gyrus than HCs. As both the precentral and postcentral gyri are key parts of the sensorimotor network (Chenji et al., 2016), our results confirm the potential role of the precentral region in MDD pathogenesis. Our BD patients had significantly smaller CT in the left SFG than HCs did. The SFG is the gyral-based representative of the dorsolateral prefrontal cortex (Niu et al., 2017) and is involved in self-awareness, complex cognitive behavior, executive function and emotional regulation (Yang et al., 2019). Our results are consistent with a previous whole-brain meta-analysis showing widespread cortical thinning in the SFG of BD patients, suggesting that structural defects in the SFG may reflect the main emotional regulatory and processing symptoms of BD (Hanford, Nazarov, Hall, & Sassi, 2016).

https://doi.org/10.1017/S0033291722003282 Published online by Cambridge University Press

In both patient groups, we found that structural defects of mOFC were associated with elevated levels of C1q, factor H, and properdin, which may provide insight into the shared neuro-inflammatory pathogenesis of mood disorders (Lanz et al., 2019). The mOFC is part of the medial prefrontal cortex (mPFC), which is a stress-sensitive brain area (McEwen & Morrison, 2013) that is involved in the regulation of emotions, cognition, and innate immunological responses to stress (Belleau, Treadway, & Pizzagalli, 2019; Savitz, Price, & Drevets, 2014). Human and rodent studies have shown that in mood disorders, repeated environmental stress can activate innate immune receptors, such as Toll-like receptors, through exogenous or endogenous ligands, thereby triggering inflammation and attenuating neuronal response, while inducing dendritic atrophy and activating microglia in the mPFC (Nie et al., 2018). More severe mPFC thinning in mood disorders has been associated with higher levels of peripheral pro-inflammatory cytokines, greater expression of inflammation-related genes, and reduced expression of neuroprotective genes (Belleau et al., 2019). Given that C1q initiates the classical complement pathway, while factor H and properdin are key regulators of the alternative pathway (Chen et al., 2018a; Noris & Remuzzi, 2013), the present study suggests that both classical and alternative complement pathways contribute to mOFC structural defects in BD and MDD.

Limitations

Our study has some limitations, including its cross-sectional design. The single assessment of peripheral blood samples could not provide evidence of causality, leaving open the question of whether the observed alterations in complement levels and brain structure are a cause or consequence of BD and MDD. The second limitation is the unbalanced sample size in our study, which may decrease statistical power. Third, we did not correct for the seven complement components in the partial correlation analysis that we conducted outside SPM.

Fourth, the association between elevated complement components and mOFC structural changes needs to be carefully interpreted. In the present study, patients were excluded if they had severe physical illnesses such as systemic lupus erythematosus, and blood samples were collected from all subjects at the same time to avoid potential confounding due to variation in inflammation state. However, other factors affecting complement levels were not taken into account, such as concurrent immunological conditions (infections, atopy, autoimmune disorders) or sleep level (Nilsson & Ekdahl, 2012). Future studies should address potential confounding from pre-existing inflammatory conditions.

Fifth, our BD patients did not show severe symptoms, despite showing increased levels of inflammatory markers. This may limit the generalizability of our findings to different BD subpopulations.

Sixth, all our BD patients were receiving medication at the time of the study. Traditional mood stabilizers such as lithium and valproate can substantially reduce neuroinflammation (Muneer, 2016), which may have affected our results. However, we believe that medication was not a strong confounder because we did not observe significant differences in complement factors between patients treated with drugs or not. In addition, BD patients showed high levels of inflammatory markers despite receiving treatment. Nevertheless, the potential influence of medication history should be rigorously assessed in future studies of complement components in mood disorders.

Finally, the patients in our study with BD had disease considerably longer than patients with depression. Nevertheless, we did not find any significant correlation between illness duration and levels of complement components. Future studies may wish to enroll only first-episode patients in order to exclude potential influence from illness duration on inflammatory response.

Conclusion and outlook

To the best of our knowledge, our study is the first to analyze the association between brain structures and complement cascades in BD patients, MDD patients, and HCs. Our results identify alterations in complement proteins and brain structures that are shared and unique between BD and MDD. Our findings underscore the need for combining dimensional and categorical approaches when describing mood disorders. Our work also reveals aspects of neuro-inflammatory pathogenesis that may be shared among mood disorders, which may help develop novel therapeutic strategies (Felger, 2017).

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S0033291722003282

Acknowledgments. The authors would like to thank all their coworkers at the Affiliated Mental Health Centre & Hangzhou Seventh People's Hospital, West China Hospital, the State Key Laboratory of Biotherapy in West China Hospital and Suzhou Psychiatry Hospital for their contributions to this research study.

Author contributions. H.Y., P.N., J.C., and T.L. developed the study, had full access to all data, and take responsibility for data integrity and accuracy. H.Y., P.N., Q.W., and W.G. drafted the manuscript. W.W., J.W., X.D., W.D., and X.M. collected the data, and Y.T., L.Z., M.L., and X.L. performed all data analyses. All authors agree to be accountable for all aspects of the work, ensuring that questions related to the accuracy or integrity of the data and results are appropriately investigated and resolved. All authors critically revised and approved the final version of the manuscript.

Financial support. This work was supported by the National Natural Science Foundation of China Key Project (81630030 and 81920108018 to T.L.), National Natural Science Foundation of China (81871054 and 81501159 to P.N., 82101598 to H.Y.), Special Foundation for Brain Research from Science and Technology Program of Guangdong (2018B030334001), 2021 Project for Hangzhou Medical Disciplines of Excellence & Key Project for Hangzhou Medical Disciplines, 1.3.5 Project for Disciplines of Excellence at West China Hospital of Sichuan University (ZY2016103, ZY2016203, and ZYGD20004 to T.L.), and Introductory Project of the Suzhou Clinical Expert Team (SZYJTD201715 to X.D. and T.L.).

Conflict of interest. The authors declare no conflicts of interest.

Ethical standards. The study was approved by the Ethics Committee of West China Hospital, Sichuan University, and complied with the principles of the Declaration of Helsinki. All participants received a complete description of the study and provided written informed consent. A copy of the written consent is available for review by the Editor of this journal.

Consent for publication. Not applicable.

Availability of data and materials. All data used in the current study are available from the corresponding author on reasonable request.

References

Akcan, U., Karabulut, S., İsmail Küçükali, C., Çakır, S., & Tüzün, E. (2018). Bipolar disorder patients display reduced serum complement levels and

elevated peripheral blood complement expression levels. *Acta Neuropsychiatrica*, *30*(2), 70–78. doi: 10.1017/neu.2017.10

Bai, Y. M., Chen, M. H., Hsu, J. W., Huang, K. L., Tu, P. C., Chang, W. C., … Tsai, S. J. (2020). A comparison study of metabolic profiles, immunity, and brain gray matter volumes between patients with bipolar disorder and depressive disorder. *Journal of Neuroinflammation*, *17*(1), 42. doi: 10.1186/s12974-020-1724-9

Batail, J. M., Coloigner, J., Soulas, M., Robert, G., Barillot, C., & Drapier, D. (2020). Structural abnormalities associated with poor outcome of a major depressive episode: The role of thalamus. *Psychiatry Research: Neuroimaging*, *305*, 111158. doi: 10.1016/j.pscychresns.2020.111158

Belleau, E. L., Treadway, M. T., & Pizzagalli, D. A. (2019). The impact of stress and major depressive disorder on hippocampal and medial prefrontal cortex morphology. *Biological Psychiatry*, *85*(6), 443–453. doi: 10.1016/j.biopsych.2018.09.031

Berk, M., Kapczinski, F., Andreazza, A. C., Dean, O. M., Giorlando, F., Maes, M., … Malhi, G. S. (2011). Pathways underlying neuroprogression in bipolar disorder: Focus on inflammation, oxidative stress and neurotrophic factors. *Neuroscience & Biobehavioral Reviews*, *35*(3), 804–817. doi: 10.1016/j.neubiorev.2010.10.001

Berk, M., Wadee, A. A., Kuschke, R. H., & O'Neill-Kerr, A. (1997). Acute phase proteins in major depression. *Journal of Psychosomatic Research*, *43*(5), 529–534. doi: 10.1016/s0022-3999(97)00139-6

Bulut, N. S., Yorguner, N., & Çarkaxhiu Bulut, G. (2021). The severity of inflammation in major neuropsychiatric disorders: Comparison of neutrophil-lymphocyte and platelet-lymphocyte ratios between schizophrenia, bipolar mania, bipolar depression, major depressive disorder, and obsessive compulsive disorder. *Nordic Journal of Psychiatry*, *75*(8), 624–632. doi: 10.1080/08039488.2021.1919201

Chang, H. H., Wang, T. Y., Lee, I. H., Lee, S. Y., Chen, K. C., Huang, S. Y., … Chen, P. S. (2017). C-reactive protein: A differential biomarker for major depressive disorder and bipolar II disorder. *The World Journal of Biological Psychiatry*, *18*(1), 63–70. doi: 10.3109/15622975.2016.1155746

Chen, J. Y., Cortes, C., & Ferreira, V. P. (2018a). Properdin: A multifaceted molecule involved in inflammation and diseases. *Molecular Immunology*, *102*, 58–72. doi: 10.1016/j.molimm.2018.05.018

Chen, L., Wang, Y., Niu, C., Zhong, S., Hu, H., Chen, P., … Huang, R. (2018b). Common and distinct abnormal frontal-limbic system structural and functional patterns in patients with major depression and bipolar disorder. *NeuroImage: Clinical*, *20*, 42–50. doi: 10.1016/j.nicl.2018.07.002

Chenji, S., Jha, S., Lee, D., Brown, M., Seres, P., Mah, D., & Kalra, S. (2016). Investigating default mode and sensorimotor network connectivity in amyotrophic lateral sclerosis. *PLOS ONE*, *11*(6), e0157443–e0157443. doi: 10.1371/journal.pone.0157443

Crider, A., Feng, T., Pandya, C. D., Davis, T., Nair, A., Ahmed, A. O., … Pillai, A. (2018). Complement component 3a receptor deficiency attenuates chronic stress-induced monocyte infiltration and depressive-like behavior. *Brain. Behavior, and Immunity*, *70*, 246–256. doi: 10.1016/j.bbi.2018.03.004

Felger, J. C. (2017). The role of dopamine in inflammation-associated depression: Mechanisms and therapeutic implications. *Current Topics in Behavioral Neurosciences*, *31*, 199–219. doi: 10.1007/7854_2016_13

First, M., Spitzer, R. L., Gibbon, M. L., & Williams, J. (2002). *Structured clinical interview for DSM-IV-TR axis I disorders, research version. Biometrics research department*. New York: New York State Psychiatric Institute.

Fontana, A., Storck, U., Angst, J., Dubs, R., Abegg, A., & Grob, P. J. (1980). An immunological basis of schizophrenia and affective disorders? *Neuropsychobiology*, *6*(5), 284–289. doi: 10.1159/000117771

Fung, G., Deng, Y., Zhao, Q., Li, Z., Qu, M., Li, K., … Chan, R. C. (2015). Distinguishing bipolar and major depressive disorders by brain structural morphometry: A pilot study. *BMC Psychiatry*, *15*, 298. doi: 10.1186/s12888-015-0685-5

Gallego, J. A., Blanco, E. A., Morell, C., Lencz, T., & Malhotra, A. K. (2021). Complement component C4 levels in the cerebrospinal fluid and plasma of patients with schizophrenia. *Neuropsychopharmacology*, *46*(6), 1140–1144. doi: 10.1038/s41386-020-00867-6

Gitlin, M. (2006). Treatment-resistant bipolar disorder. *Molecular Psychiatry*, *11*(3), 227–240. doi: 10.1038/sj.mp.4001793

Hamilton, M. (1960). A rating scale for depression. *Journal of Neurology, Neurosurgery and Psychiatry*, *23*(1), 56–62. doi: 10.1136/jnnp.23.1.56

Han, K.-M., De Berardis, D., Fornaro, M., & Kim, Y.-K. (2019). Differentiating between bipolar and unipolar depression in functional and structural MRI studies. *Progress in Neuro-Psychopharmacology & Biological Psychiatry*, *91*, 20–27. doi: 10.1016/j.pnpbp.2018.03.022

Hanford, L. C., Nazarov, A., Hall, G. B., & Sassi, R. B. (2016). Cortical thickness in bipolar disorder: A systematic review. *Bipolar Disorders*, *18*(1), 4–18. doi: 10.1111/bdi.12362

Hasin, D. S., Goodwin, R. D., Stinson, F. S., & Grant, B. F. (2005). Epidemiology of major depressive disorder: Results from the national epidemiologic survey on alcoholism and related conditions. *Archives of General Psychiatry*, *62*(10), 1097–1106. doi: 10.1001/archpsyc.62.10.1097

Hong, W., Zhao, Z., Wang, D., Li, M., Tang, C., Li, Z., … Chan, C. C. H. (2020). Altered gray matter volumes in post-stroke depressive patients after subcortical stroke. *NeuroImage: Clinical*, *26*, 102224. doi: 10.1016/j.nicl.2020.102224

Kong, L., Herold, C. J., Zöllner, F., Salat, D. H., Lässer, M. M., Schmid, L. A., … Schröder, J. (2015). Comparison of grey matter volume and thickness for analysing cortical changes in chronic schizophrenia: A matter of surface area, grey/white matter intensity contrast, and curvature. *Psychiatry Research: Neuroimaging*, *231*(2), 176–183. doi: 10.1016/j.pscychresns.2014.12.004

Kopczynska, M., Zelek, W., Touchard, S., Gaughran, F., Di Forti, M., Mondelli, V., … Morgan, B. P. (2019). Complement system biomarkers in first episode psychosis. *Schizophrenia Research*, *204*, 16–22. doi: 10.1016/j.schres.2017.12.012

Kopczynska, M., Zelek, W. M., Vespa, S., Touchard, S., Wardle, M., Loveless, S., … Morgan, B. P. (2018). Complement system biomarkers in epilepsy. *Seizure - European Journal of Epilepsy*, *60*, 1–7. doi: 10.1016/j.seizure.2018.05.016

Kronfol, Z., & House, J. D. (1989). Lymphocyte mitogenesis, immunoglobulin and complement levels in depressed patients and normal controls. *Acta Psychiatrica Scandinavica*, *80*(2), 142–147. doi: 10.1111/j.1600-0447.1989.tb01316.x

Lan, M. J., Chhetry, B. T., Oquendo, M. A., Sublette, M. E., Sullivan, G., Mann, J. J., & Parsey, R. V. (2014). Cortical thickness differences between bipolar depression and major depressive disorder. *Bipolar Disorders*, *16*(4), 378–388. doi: 10.1111/bdi.12175

Lanz, T. A., Reinhart, V., Sheehan, M. J., Rizzo, S. J. S., Bove, S. E., James, L. C., … Kleiman, R. J. (2019). Postmortem transcriptional profiling reveals widespread increase in inflammation in schizophrenia: A comparison of prefrontal cortex, striatum, and hippocampus among matched tetrads of controls with subjects diagnosed with schizophrenia, bipolar or major depressive disorder. *Translational Psychiatry*, *9*(1), 151. doi: 10.1038/s41398-019-0492-8

Leonard, B. E. (2018). Inflammation and depression: A causal or coincidental link to the pathophysiology? *Acta Neuropsychiatrica*, *30*(1), 1–16. doi: 10.1017/neu.2016.69

MacDonald, K., Krishnan, A., Cervenka, E., Hu, G., Guadagno, E., & Trakadis, Y. (2019). Biomarkers for major depressive and bipolar disorders using metabolomics: A systematic review. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *180*(2), 122–137. doi: 10.1002/ajmg.b.32680

Maes, M., Delange, J., Ranjan, R., Meltzer, H. Y., Desnyder, R., Cooremans, W., & Scharpé, S. (1997). Acute phase proteins in schizophrenia, mania and major depression: Modulation by psychotropic drugs. *Psychiatry Research*, *66*(1), 1–11. doi: 10.1016/s0165-1781(96)02915-0

McEwen, B. S., & Morrison, J. H. (2013). The brain on stress: Vulnerability and plasticity of the prefrontal cortex over the life course. *Neuron*, *79*(1), 16–29. doi: 10.1016/j.neuron.2013.06.028

Merikangas, K. R., Akiskal, H. S., Angst, J., Greenberg, P. E., Hirschfeld, R. M. A., Petukhova, M., & Kessler, R. C. (2007). Lifetime and 12-month prevalence of bipolar spectrum disorder in the national comorbidity survey replication. *Archives of General Psychiatry*, *64*(5), 543–552. doi: 10.1001/archpsyc.64.5.543

Moreno, C., Hasin, D. S., Arango, C., Oquendo, M. A., Vieta, E., Liu, S., … Blanco, C. (2012). Depression in bipolar disorder versus major depressive disorder: Results from the national epidemiologic survey on alcohol and related conditions. *Bipolar Disorders*, *14*(3), 271–282. doi: 10.1111/j.1399-5618.2012.01009.x

Muneer, A. (2016). Bipolar disorder: Role of inflammation and the development of disease biomarkers. *Psychiatry Investigation*, *13*(1), 18–33. doi: 10.4306/pi.2016.13.1.18

Nie, X., Kitaoka, S., Tanaka, K., Segi-Nishida, E., Imoto, Y., Ogawa, A., … Furuyashiki, T. (2018). The innate immune receptors TLR2/4 mediate repeated social defeat stress-induced social avoidance through prefrontal microglial activation. *Neuron*, *99*(3), 464–479, e467. doi: 10.1016/j.neuron.2018.06.035

Nilsson, B., & Ekdahl, K. N. (2012). Complement diagnostics: Concepts, indications, and practical guidelines. *Clinical & Developmental Immunology*, *2012*, 962702–962702. doi: 10.1155/2012/962702

Niu, M., Wang, Y., Jia, Y., Wang, J., Zhong, S., Lin, J., … Huang, R. (2017). Common and specific abnormalities in cortical thickness in patients with major depressive and bipolar disorders. *EBioMedicine*, *16*, 162–171. doi: 10.1016/j.ebiom.2017.01.010

Noris, M., & Remuzzi, G. (2013). Overview of complement activation and regulation. *Seminars in Nephrology*, *33*(6), 479–492. doi: 10.1016/j.semnephrol.2013.08.001

O'Connell, K. S., Sønderby, I. E., Frei, O., van der Meer, D., Athanasiu, L., Smeland, O. B., … Djurovic, S. (2021). Association between complement component 4A expression, cognitive performance and brain imaging measures in UK Biobank. *Psychological Medicine*, *3*, 1–11. doi: 10.1017/s0033291721000179.

Opel, N., Goltermann, J., Hermesdorf, M., Berger, K., Baune, B. T., & Dannlowski, U. (2020). Cross-disorder analysis of brain structural abnormalities in six major psychiatric disorders: A secondary analysis of mega- and meta-analytical findings from the ENIGMA consortium. *Biological Psychiatry*, *88*(9), 678–686. doi: 10.1016/j.biopsych.2020.04.027

Parker, G. (2014). The DSM-5 classification of mood disorders: Some fallacies and fault lines. *Acta Psychiatrica Scandinavica*, *129*(6), 404–409. doi: 10.1111/acps.12253

Pillai, A., Bruno, D., Nierenberg, J., Pandya, C., Feng, T., Reichert, C., … Pomara, N. (2019). Complement component 3 levels in the cerebrospinal fluid of cognitively intact elderly individuals with major depressive disorder. *Biomarkers in Neuropsychiatry*, *1*, 100007. doi: 10.1016/j.bionps.2019.100007

Reginia, A., Kucharska-Mazur, J., Jabłoński, M., Budkowska, M., Dołęgowska, B., Sagan, L., … Samochowiec, J. (2018). Assessment of complement cascade components in patients with bipolar disorder. *Frontiers in Psychiatry*, *9*, 614–614. doi: 10.3389/fpsyt.2018.00614

Reid, K. B. M. (2018). Complement component C1q: Historical perspective of a functionally versatile, and structurally unusual, serum protein. *Frontiers in Immunology*, *9*, 764. doi: 10.3389/fimmu.2018.00764

Rey, R., Suaud-Chagny, M. F., Bohec, A. L., Dorey, J. M., d'Amato, T., Tamouza, R., & Leboyer, M. (2020). Overexpression of complement component C4 in the dorsolateral prefrontal cortex, parietal cortex, superior temporal gyrus and associative striatum of patients with schizophrenia. *Brain. Behavior, and Immunity*, *90*, 216–225. doi: 10.1016/j.bbi.2020.08.019

Rosenblat, J. D., Kakar, R., Berk, M., Kessing, L. V., Vinberg, M., Baune, B. T., … McIntyre, R. S. (2016). Anti-inflammatory agents in the treatment of bipolar depression: A systematic review and meta-analysis. *Bipolar Disorders*, *18*(2), 89–101. doi: 10.1111/bdi.12373

Savitz, J. B., Price, J. L., & Drevets, W. C. (2014). Neuropathological and neuromorphometric abnormalities in bipolar disorder: View from the medial prefrontal cortical network. *Neuroscience & Biobehavioral Reviews*, *42*, 132–147. doi: 10.1016/j.neubiorev.2014.02.008

Schulze, T. G., Akula, N., Breuer, R., Steele, J., Nalls, M. A., Singleton, A. B., … McMahon, F. J. (2014). Molecular genetic overlap in bipolar disorder, schizophrenia, and major depressive disorder. *The World Journal of Biological Psychiatry*, *15*(3), 200–208. doi: 10.3109/15622975.2012.662282

Sekar, A., Bialas, A. R., de Rivera, H., Davis, A., Hammond, T. R., Kamitaki, N., … McCarroll, S. A. (2016). Schizophrenia risk from complex variation of complement component 4. *Nature*, *530*(7589), 177–183. doi: 10.1038/nature16549

Shin, C., Ham, B. J., Ko, Y. H., Pae, C. U., Park, M. H., Steffens, D. C., … Han, C. (2019). Increased plasma complement factor H is associated with geriatric depression. *International Psychogeriatrics*, *31*(1), 101–108. doi: 10.1017/s1041610218000558

Solmi, M., Suresh Sharma, M., Osimo, E. F., Fornaro, M., Bortolato, B., Croatto, G., … Carvalho, A. F. (2021). Peripheral levels of C-reactive protein, tumor necrosis factor-α, interleukin-6, and interleukin-1β across the mood spectrum in bipolar disorder: A meta-analysis of mean differences and variability. *Brain, Behavior, and Immunity*, *97*, 193–203. doi: 10.1016/j.bbi.2021.07.014

Swardfager, W., Rosenblat, J. D., Benlamri, M., & McIntyre, R. S. (2016). Mapping inflammation onto mood: Inflammatory mediators of anhedonia. *Neuroscience & Biobehavioral Reviews*, *64*, 148–166. doi: 10.1016/j.neubiorev.2016.02.017

Tseng, H. H., Chang, H. H., Wei, S. Y., Lu, T. H., Hsieh, Y. T., Yang, Y. K., & Chen, P. S. (2021). Peripheral inflammation is associated with dysfunctional corticostriatal circuitry and executive dysfunction in bipolar disorder patients. *Brain, Behavior, and Immunity*, *91*, 695–702. doi: 10.1016/j.bbi.2020.09.010

Wadee, A. A., Kuschke, R. H., Wood, L. A., Berk, M., Ichim, L., & Maes, M. (2002). Serological observations in patients suffering from acute manic episodes. *Human Psychopharmacology: Clinical and Experimental*, *17*(4), 175–179. doi: 10.1002/hup.390

Wechsler, D. (1981). *WAIS-R Manual: Wechsler adult intelligence scale-revised*. London: Psychological Corporation.

Wei, J., Liu, Y., Zhao, L., Yang, X., Ni, P., Wang, Y., … Ma, X. (2018). Plasma complement component 4 increases in patients with major depressive disorder. *Neuropsychiatric Disease and Treatment*, *14*, 37–41. doi: 10.2147/ndt.S151238

Wise, T., Radua, J., Via, E., Cardoner, N., Abe, O., Adams, T. M., … Arnone, D. (2017). Common and distinct patterns of grey-matter volume alteration in major depression and bipolar disorder: Evidence from voxel-based meta-analysis. *Molecular Psychiatry*, *22*(10), 1455–1463. doi: 10.1038/mp.2016.72

Woo, J. J., Pouget, J. G., Zai, C. C., & Kennedy, J. L. (2020). The complement system in schizophrenia: Where are we now and what's next? *Molecular Psychiatry*, *25*(1), 114–130. doi: 10.1038/s41380-019-0479-0

Wu, T., Dejanovic, B., Gandham, V. D., Gogineni, A., Edmonds, R., Schauer, S., … Hanson, J. E. (2019). Complement C3 is activated in human AD brain and is required for neurodegeneration in mouse models of amyloidosis and tauopathy. *Cell Reports*, *28*(8), 2111–2123, e2116. doi: 10.1016/j.celrep.2019.07.060

Yang, X., Tao, H., Xiao, L., Li, C., Tang, Y., & Liu, Y. (2018). Increased Serum C3 and decreased UA in patients of bipolar disorder in Chinese Han population. *Frontiers in Psychiatry*, *9*, 381. doi: 10.3389/fpsyt.2018.00381

Yang, Y., Liu, S., Jiang, X., Yu, H., Ding, S., Lu, Y., … Lv, L. (2019). Common and specific functional activity features in schizophrenia, major depressive disorder, and bipolar disorder. *Frontiers in Psychiatry*, *10*, 52. doi: 10.3389/fpsyt.2019.00052

Yao, Q., & Li, Y. (2020). Increased serum levels of complement C1q in major depressive disorder. *Journal of Psychosomatic Research*, *133*, 110105. doi: 10.1016/j.jpsychores.2020.110105

Young, R. C., Biggs, J. T., Ziegler, V. E., & Meyer, D. A. (1978). A rating scale for mania: Reliability, validity and sensitivity. *The British Journal of Psychiatry*, *133*(5), 429–435. doi: 10.1192/bjp.133.5.429

Zhang, C., Lv, Q., Fan, W., Tang, W., & Yi, Z. (2017a). Influence of CFH gene on symptom severity of schizophrenia. *Neuropsychiatric Disease and Treatment*, *13*, 697–706. doi: 10.2147/ndt.S132108

Zhang, J., Wu, Y.-L., Su, J., Yao, Q., Wang, M., Li, G.-F., … Du, X. (2017b). Assessment of gray and white matter structural alterations in migraineurs without aura. *The Journal of Headache and Pain*, *18*(1), 74. doi: 10.1186/s10194-017-0783-5

Zhang, X., Kimura, Y., Fang, C., Zhou, L., Sfyroera, G., Lambris, J. D., … Song, W. C. (2007). Regulation of toll-like receptor-mediated inflammatory response by complement in vivo. *Blood*, *110*(1), 228–236. doi: 10.1182/blood-2006-12-063636