significant contiguous voxels in the above analysis. The same VOIs were applied to $\lambda_1 - \lambda_3$ images, and $\lambda_1 - \lambda_3$ values were extracted. Axial (λ_1) and radial diffusivity ($[\lambda_2 + \lambda_3]/2$) were compared.

2.5. Statistical analysis

Group differences in demographic characteristics between patients and healthy controls were examined by unpaired t-test and Pearson χ^2 test. To examine the group differences of FA values and axial/radial diffusivity in VOIs shown in the voxel-based analysis, we performed ANCOVA with age and gender as covariates. We included age and gender as covariates because they reportedly affect white matter integrity (Inano et al., 2011). Paired t-tests were performed to examine the changes in the mRS, MMSE, SDS, and HAM-D scores and the FA values of patients and controls during the 6-month period after their initial examinations. We computed Pearson's correlations to examine the relationship between FA values and depressive symptoms at the first assessment and at the 6-month follow-up assessment. Pearson's correlations were also used to examine the relationship between the change in depression scale scores and the ratio of the FA values (FA values at second vs. initial examination) in patients. To examine whether the ratio of the FA values was related to the change in depression scale scores (SDS and HAM-D scores at second minus initial examination), we performed a multiple regression analysis with the change in depression scale scores as the dependent variable and the ratio of the FA values as the independent variable, after adjustment for age and gender.

All statistical tests were 2-tailed and reported at $\alpha < 0.05$. Bonferroni correction was applied to avoid type I errors due to the multiplicity of statistical analyses. Statistical analysis of the data was performed using SPSS for Windows 19.0 (IBM Japan Inc., Tokyo, Japan).

3. Results

3.1. Demographic and clinical data

Table 1 summarizes the demographic and clinical characteristics of the participants. Patients differed significantly from healthy control subjects in MMSE, SDS and HAM-D scores. The MMSE score was lower, and SDS and HAM-D scores were higher, among the patients. Table 1 also shows the mRS score and the location and volume of the infarctions among the patients. The main locations of the infarctions were the basal ganglia (44.8%), the subcortical white matter in the frontal lobe (20.7%), and the thalamus (13.8%).

3.2. Between-group comparisons of FA values

In the voxel-based analysis of FA values, the patient and healthy control groups differed in white matter FA values in the left and right anterior limbs of the internal capsule [left anterior limb of internal capsule: $(x, y, z) = (-24, 16, 16)$, cluster voxel size $= 189$, $T = 6.41$; right anterior limb of internal capsule: $(x, y, z) = (16, 6, 10)$, cluster voxel size $= 756$, $T = 6.86$] (Fig. 1a). Table 2 shows the quantification of the differences in FA value and radial/axial diffusivity in these affected regions. These regions revealed decreased axial diffusivity but no change in radial diffusivity. When we added the MMSE, SDS, and HAM-D scores as covariates in the ANCOVA, the results did not change.

3.3. Change in FA values of patients over 6 months

There were no significant differences in demographic data between participants who were followed and those who were lost to follow-up, except the age of the healthy control groups (Table 3). Table 4 shows the changes in psychometric scores and FA values and axial/radial diffusivity over 6 months in the followed-up patients and controls. Healthy controls showed no significant change of FA values in the anterior limb of the internal capsule 6 months after the initial examination (Table 4). Patients showed significantly increased FA values in the anterior limb of the internal capsule 6 months after the infarction, although their FA
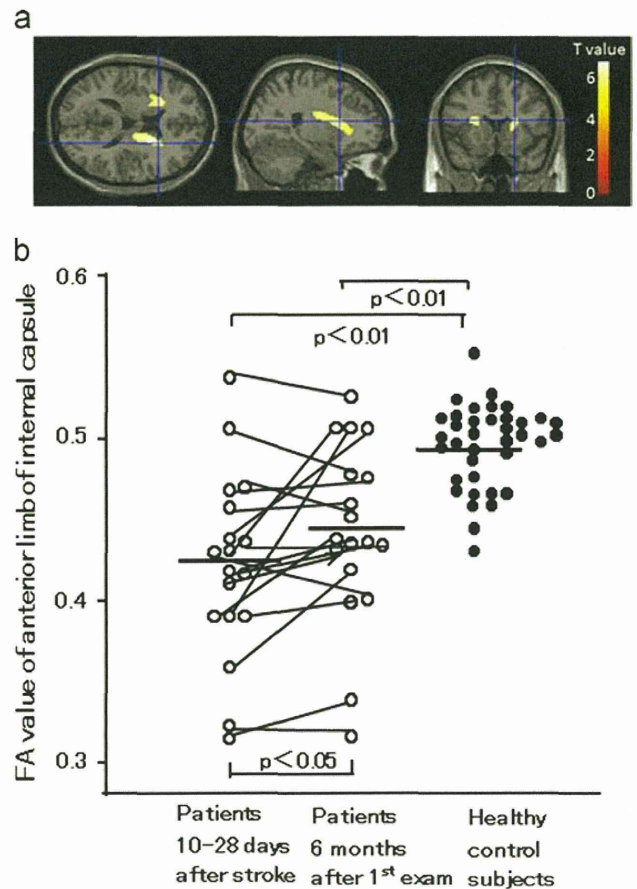

Fig. 1. White matter Fractional Anisotropy (FA) differences in voxel-based comparisons between stroke patients ($n=29$) and control subjects ($n=37$) (Fig. 1a), and scatter plots of FA values in the region of FA reduction among stroke patients ($n=18$) at 10–28 days after stroke/after 6-month follow-up and of control subjects ($n=37$) (Fig. 1b). (a) Images are presented in radiological orientation. Statistical parametric mapping projections were superimposed on a representative magnetic resonance image ($x=24, y=16, z=16$). Patients showed reduced FA in the right and left anterior limbs of the internal capsule. Statistical inferences were made with a voxel-level statistical threshold ($p < 0.05$) after family-wise error correction for multiple comparisons with a minimum cluster size of 50 voxels. (b) Significant increase in FA of the patients was observed at 6-month follow-up ($p < 0.05$), although the FA values of the patients were still lower than those of healthy subjects at both the initial and follow-up examinations ($p < 0.01$).

values were still lower relative to those of healthy control subjects at this time point (Table 4, Fig. 1b). There were no significant changes in MMSE, SDS and HAM-D scores 6 months after the initial examination in the groups of patients and healthy controls (Table 4).

There were no significant relationships between FA values and depressive symptoms at the first assessment and at the assessment performed 6 months later, but we found a significant negative correlation between the increased ratio of the FA values and the change in the depression scores on the SDS and HAM-D at 6-month follow-up ($r = -0.59$, $p = 0.01$ for SDS; $r = -0.56$, $p = 0.02$ for HAM-D) (Fig. 2). With Spearman's correlational analysis, the results were not changed ($r = -0.56$, $p = 0.02$ for the SDS; $r = -0.73$, $p = 0.001$ for the HAM-D). When we considered the influence of the volume of infarcts and lesion location [cortex ($n=2$), basal ganglia ($n=8$), thalamus ($n=1$), and subcortical white matter ($n=7$)] as a covariate in correlational analysis, the correlations were not changed ($r = -0.59$, $p = 0.02$ for SDS; $r = -0.57$, $p = 0.02$ for HAM-D). When we considered the influence of the degree of handicap by including the change in mRS scores as a covariate in the correlation analysis, which showed a significant decrease during follow-up (mRS score $= 2.1 \pm 0.8$ at first examination, 1.6 ± 0.6 at 6-month follow-up, $t = 4.12$, $p < 0.01$, paired

Table 2
Differences in values of FA and axial/radial diffusivity in VOIs between patients and healthy control subjects.

FA and axial/radial diffusivity	Patients (n=29)	Healthy controls (n=37)	Analysis of covariance [a]	
			F (1, 62)	P
Left anterior limb of internal capsule				
FA	0.44 ± 0.05	0.49 ± 0.05	16.7	< 0.001**
Axial diffusivity (× 10^{-3})	4.16 ± 0.36	4.37 ± 0.35	5.46	0.02 *
Radial diffusivity (× 10^{-3})	3.94 ± 0.33	4.03 ± 0.33	1.38	0.25
Right anterior limb of internal capsule				
FA	0.45 ± 0.05	0.51 ± 0.05	21.8	< 0.001**
Axial diffusivity (× 10^{-3})	4.15 ± 0.36	4.36 ± 0.36	5.19	0.03 *
Radial diffusivity (× 10^{-3})	3.91 ± 0.33	4.01 ± 0.33	1.38	0.24
Average of right and left anterior limbs of internal capsule				
FA	0.44 ± 0.05	0.50 ± 0.05	20.8	< 0.001**
Axial diffusivity (× 10^{-3})	4.15 ± 0.36	4.36 ± 0.35	5.35	0.02 *
Radial diffusivity (× 10^{-3})	3.92 ± 0.33	4.02 ± 0.33	1.38	0.24

Data are mean ± S.D.
[a] Age and gender are entered as covariates.
* $p < 0.05$.
** $p < 0.01$.

Table 3
Demographic characteristics of patients and healthy control subjects who could be followed up and who were lost to follow-up.

	Follow-up	Lost to follow-up	t or χ^2	P
Patients	n=18	n=11		
Age (years)	69.2 ± 8.0	67.6 ± 9.0	0.58	0.57
Female sex (n, %)	5 (26.3)	1 (9.1)	1.45	0.24
mRS score	2.1 ± 0.8	2.4 ± 0.9	1.00	0.33
MMSE score	27.6 ± 3.5	27.5 ± 2.8	0.47	0.64
SDS score	26.9 ± 5.6	27.1 ± 7.3	0.42	0.68
HAM-D score	3.1 ± 2.7	2.3 ± 3.0	0.46	0.65
Controls	n=19	n=18		
Age (years)	69.1 ± 8.0	65.8 ± 8.1	5.22	< 0.01
Female sex (n, %)	10 (55.5)	10 (55.5)	0.03	0.86
MMSE score	29.2 ± 1.2	29.2 ± 0.8	0.10	0.92
SDS score	22.1 ± 1.8	26.2 ± 1.8	1.64	0.11
HAM-D score	0.4 ± 0.6	1.8 ± 2.1	1.61	0.12

t-test), the correlational results were unchanged ($r= -0.61$, $p=0.01$ for the SDS; $r= -0.58$, $p=0.02$ for the HAM-D).

With multiple regression analysis evaluating whether the increased ratio of FA values was related to the change in the SDS and HAM-D depression scores at 6-month follow-up, the ratio of FA values was negatively related to both the changes in the SDS scores (beta= -0.44, $p=0.03$) and the HAM-D scores (beta= -0.46, $p=0.04$).

4. Discussion

Our findings showed that stroke patients had lower FA in the bilateral anterior limbs of the internal capsule relative to healthy control subjects. Six months after onset, a significant increase in FA was noted, and it was associated with a reduction in depression scale scores. The association of the increase in FA and the reduction in depression scale scores remained significant when we considered the influence of the volume of infarcts and lesion location and the severity of neurological symptoms as a covariate in the partial correlation analysis.

The reduced FA level of patients was associated with decreased axial diffusivity. Axonal damage leads to a marked decrease in axial diffusivity, while demyelination leads to an increase in radial diffusivity (Song et al., 2005). Therefore, our finding was not a result of demyelination but rather of gross reduction in axonal number and/or size, possibly reflecting Wallerian degeneration secondary to neuronal loss due to stroke (Thomalla et al., 2004). From an anatomical perspective, the anterior limb of the internal capsule represents the intercept point in the course of the frontal-subcortical circuits (Axer and Keyserlingk, 2000), and it has extensive connectivity with the cortical and subcortical areas. Its reduced FA may reflect the conjunctive focus of degeneration due to stroke in spatially different sites of cortical and subcortical areas.

The frontal-striatal-thalamic-cortical circuits play an important role in behavioral regulation (Duran et al., 2009), and microstructural change in the anterior limb of the internal capsule was shown to be related to the severity of depressive symptoms in adults with Major Depressive Disorder (MDD) using MRI (Zou et al., 2008). Degeneration in this region may relate to a loss of white matter integrity of these neural circuits (Budde et al., 2007), and this abnormality might trigger the onset of negative mood change. Our finding of a significant negative correlation between the increased ratio of FA values and the change in the depression scale scores 6 months after the stroke might reflect the association between axonal damage of the internal capsule and the negative mood change in stroke patients, and it indicated that recovery from microstructural abnormalities decreased the vulnerability to post-stroke depression, as predicted by elevated depression scale scores.

Our study has some limitations. First, the sample size was not large enough to elucidate the moderate size of difference between groups, and further study with increased numbers of subjects is necessary to draw any definitive conclusions. Second, the stroke patients had predominantly suffered a modest ischemic insult. The extent to which our findings are related to the severity of the ischemic insult was uncertain. Third, patients with significant comprehension deficits were excluded because clinical verbal interviews could not be conducted. Finally, we were not able to control precisely for important variables such as social support, pre-morbid personality, and concurrent and past medication histories. Further analysis, inclusive of consideration of these points, is needed to confirm our present findings.

In conclusion, the present study suggests that FA reduction in the bilateral anterior limbs of the internal capsule is evident in stroke patients. This regional axonal damage should be related to abnormality of neuroanatomical pathways in frontal-subcortical circuits, recent studies of which have highlighted the specific relevance in the pathophysiology of depression due to stroke (Terroni et al., 2011; Paradiso et al., 2013), and it may increase

Table 4

Change in psychometric scores and FA values and axial/radial diffusivity over 6 months in patients ($n=18$) and controls ($n=19$).

	10–28 Days after stroke	6 Months after first exam	Paired t-test	p
Patients				
Age (years)	69.2 ± 8.0	–	–	–
Female sex (n, %)	5 (26.3)	–	–	–
mRS score	2.1 ± 0.8	1.6 ± 0.6	$t_{17}=4.12$	< 0.01**
MMSE score	27.6 ± 3.5	29.0 ± 2.1	$t_{17}=2.08$	0.05
SDS score	26.9 ± 5.6	29.2 ± 11.5	$t_{17}=0.93$	0.37
HAM-D score	3.1 ± 2.7	2.8 ± 4.9	$t_{17}=0.21$	0.84
Anterior limb of internal capsule				
FA	0.42 ± 0.06	0.44 ± 0.06	$t_{17}=2.16$	0.05 *
Axial diffusivity ($\times 10^{-3}$)	4.13 ± 0.37	4.29 ± 0.45	$t_{17}=1.32$	0.21
Radial diffusivity ($\times 10^{-3}$)	3.91 ± 0.34	4.01 ± 0.37	$t_{17}=1.38$	0.37
Controls				
Age (years)	69.1 ± 8.0			
Female sex (n, %)	10 (55.5)			
MMSE score	29.2 ± 1.2	29.7 ± 0.5	$t_{18}=0.95$	0.36
SDS score	22.1 ± 1.8	22.8 ± 2.6	$t_{18}=1.80$	0.10
HAM-D score	0.4 ± 0.6	0.3 ± 0.6	$t_{18}=1.00$	0.33
Anterior limb of internal capsule				
FA	0.49 ± 0.03	0.48 ± 0.03	$t_{18}=0.76$	0.46
Axial diffusivity ($\times 10^{-3}$)	4.18 ± 0.66	4.30 ± 0.55	$t_{18}=0.92$	0.37
Radial diffusivity ($\times 10^{-3}$)	3.86 ± 0.60	4.00 ± 0.46	$t_{18}=0.75$	0.46

mRS=Modified Rankin Scale. MMSE=Mini-Mental State Examination. SDS=Zung Self-Rating Depression Scale. HAM-D=Hamilton Rating Scale for Depression.

Data are mean ± S.D.
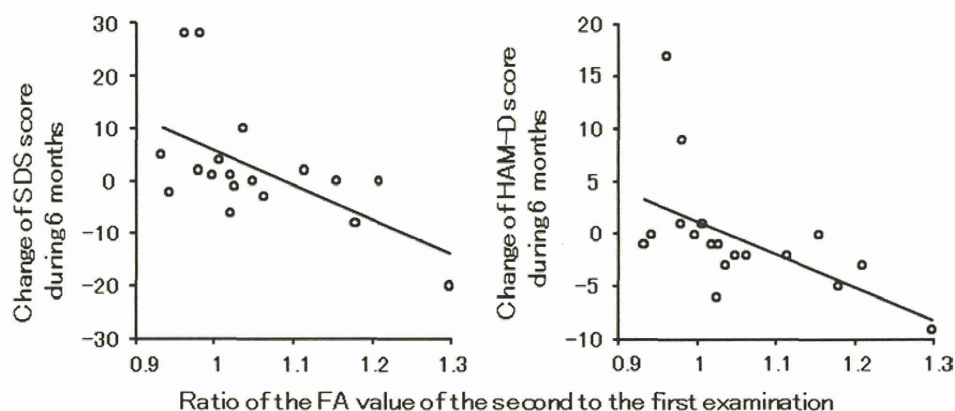
* $p < 0.05$.

** $p < 0.01$.

Fig. 2. Scatter plots showing the relationship between the ratio of the FA values of the second to the first examination and the change of depression scale scores among patients ($n=18$) Significant correlations were observed between the ratio of the FA values of the second examination to the first examination, and the changes in depression scale scores ($r=-0.59$, $p=0.01$ for the SDS; $r=-0.88$, $p=0.0001$ for the HAM-D).

the risk of post-stroke depression in conjunction with psychosocial factors following stroke. Further, a significant increase of FA in this region was noted 6 months after the stroke, and an association with a reduction in depression scale scores was revealed. Our findings suggested that the promotion of recovery from the axonal damage of stroke patients might prevent the onset of depressive symptoms. Further investigations are needed to clarify whether and how axonal damage of the internal capsule in stroke patients affects the clinical presentation, treatment response and outcome of depression or other psychiatric conditions in stroke survivors.

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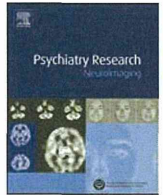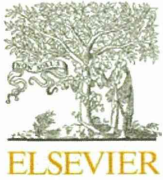
Increased binding of 5-HT$_{1A}$ receptors in a dissociative amnesic patient after the recovery process

CrossMark

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ARTICLE INFO

ABSTRACT

Dissociative amnesia is characterized by an inability to retrieve information already saved in memories. 5-HT has some role in neural regulatory control and may be related to the recovery from dissociative amnesia. To examine the role of 5-HT$_{1A}$ receptors in the recovery from dissociative amnesia, we performed two positron emission tomography (PET) scans on a 30-year-old patient of dissociative amnesia using [^{11}C]WAY-100635, the first at amnesic state, and the second at the time he had recovered. Exploratory voxel-based analysis (VBA) was performed using SPM software. 5-HT$_{1A}$ BP$_{ND}$ images were compared between the patient at amnesic and recovery states and healthy subjects (14 males, mean age 29.8 ± 6.45) with Jack-knife analysis. 5-HT$_{1A}$ receptor bindings of the patient at the recovery state were significantly higher than those of healthy subjects in the right superior and middle frontal cortex, left inferior frontal and orbitofrontal cortex and bilateral inferior temporal cortex. The increase in BP$_{ND}$ values of recovery state was beyond 10% of those of amnesia state in these regions except in the right superior frontal cortex. We considered that neural regulatory control by the increase of 5-HT$_{1A}$ receptors in cortical regions played a role in the recovery from dissociative amnesia.

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1. Introduction

Dissociative amnesia is characterized by an inability to retrieve information already saved in memories, and is usually related to a traumatic or stressful autobiographical event that cannot be explained by a physiological condition such as brain damage (Markowitsch, 1995). The diagnosis usually relies on features such as circumstances of onset, loss of personal autographical identity, and whether or not new learning is affected (often spared in dissociative amnesia). Semantic knowledge often remains intact and memories for skills are often preserved (Kopelman, 1987).

The underlying neuronal mechanism of dissociative amnesia has been unclear, and we performed a positron emission tomography (PET) activation study on a dissociative amnesic patient. A task requiring explicit retrograde memory of faces was compared with a control task. During the task, activation of the right anterior medial temporal region including the amygdala was increased in the patient. During recovery, the right anterior medial temporal region became less active while the right hippocampal region became more active (Yasuno et al., 2000). In addition, in a functional magnetic resonance imaging (fMRI study), (Anderson et al., 2004) reported that increased activity of the bilateral dorsolateral prefrontal cortex and decreased activity of the hippocampus were involved in suppression of unwanted memories. In a functional MRI study, (Kikuchi et al., 2010) reported that hyperactivation of the prefrontal regions and deactivation of the hippocampus were observed in a dissociative amnesia patient, but these functional changes vanished at the recovery state. They also found that the patient who did not recover from retrograde amnesia continued to show abnormal activation in the prefrontal regions.

These previous studies suggested that the inhibition of memory retrieval in the dissociative amnesic patients occurred by the suppression of limbic regions such as the hippocampus due to abnormal activity of the prefrontal / temporal regions. Normalization of the abnormal activity of these regions could play an important role in the recovery from dissociative amnesia. On the other hand, the function of 5-hydroxytryptamine (5-HT) has been implicated in neural regulatory control, and 5-HT$_{1A}$ receptor is the main inhibitory serotonergic receptor subtype (Fink et al., 2008; Drueke et al., 2013). The 5-HT$_{1A}$ receptors are in a position to

influence the activity of glutamatergic, cholinergic and possibly GABAergic neurons in the cerebral cortex, hippocampus, and in the septohippocampal projection, thereby affecting memory functions (Ogren et al., 2008). We hypothesized that 5-HT$_{1A}$ receptors had some potential role in the manifestation of, or recovery from dissociative amnesia.

In this study, we described a dissociative amnesia patient who experienced generalized retrograde amnesia, without anterograde amnesia, following a stressful event without organic backgrounds. To examine the role of regional 5-HT$_{1A}$ receptors in the recovery from dissociative amnesia, we performed PET scans for the patient using [^{11}C] WAY-100635 (Pike et al., 1996; Farde et al., 1998). Regional 5-HT$_{1A}$ receptor bindings of the patient at the amnesic and recovery states were compared to those of age-matched healthy controls (HC), and then the changes in regional 5-HT$_{1A}$ receptor bindings of the patient during the recovery were examined.

2. Materials and method

2.1. Patient report and healthy control subjects

A 30-year-old right-handed male patient was admitted to our hospital on referral for treatment of his generalized dissociative amnesia. Before onset, he had been troubled by work-related and possibly legal problems. During his travel to work, he became disorientated and not knowing who or where he was. Then he was referred to a hospital.

He was introduced and referred to our institute one month after onset. A physical examination revealed no abnormalities. Brain computed tomography scan revealed no brain damage. He was alert, attentive and oriented. Spontaneous speech, comprehension, repetition, and naming were normal, as were calculation, mapping, praxis, right-left orientation, and finger naming. All other neurological parameters were normal except for persistent memory disturbance. He met the DSM-IV-TR criteria and was diagnosed as dissociative amnesia. Then, five months after onset, he was referred to our institute again. At the second examination he had already regained his autobiographical memory and he had no difficulties in retrieving long-term memories. The patient did not suffer from a psychiatric disorder before his amnesic phase and he did not develop any psychiatric disorder after the first measurement. He did not take any steady dose of medication during the five-month period.

PET acquisition using [^{11}C] WAY-100635 was performed twice, the first at one month after onset, when the patient was in an amnesic state, and the second five months after onset, when he had recovered from amnesia and regained all his memories. Structural MRI was performed on the day of the initial PET acquisition. The same PET and MRI were performed on HC (14 males, mean age 29.8 ± 6.45 years). The HC group was recruited from the local area by poster advertisement. Exclusion criteria for HC were a history or present diagnosis of any DSM-IV axis I diagnosis or any neurological illness.

This study was approved by the Ethics and Radiation Safety Committee of the National Institute of Radiological Sciences, Chiba, Japan. After complete description of the study, written informed consent was obtained from all subjects.

2.2. Neuropsychological data

Table 1 shows the neuropsychological data of the patient at the amnesic and post-amnesic states. There were no remarkable abnormalities except for retrograde amnesia at the dissociative amnesic state. The patient was alert, attentive, socially appropriate, and had normal digit span performance (seven digits forward, four backward). Intellectual functioning assessed by the WAIS-R (Wechsler, 1987) and RCPM (Raven, 1958) tests was adequate. Word fluency was normal, and all language and language-related functions were intact. The patient had no anterograde memory deficits. Results with RAVLT-R (Spreen and Strauss, 1991) and the Rey-Osterrieth Figure were normal (Lezak, 1983), and performance on WMS-R (Wechsler, 1987) was also excellent.

Autobiographical memory for events before his illness was assessed by a structured interview covering past personal events. The items selected for this interview covered educational experiences, occupational history, births, deaths and other events within his family. In this test, difficulties appeared during the amnesic state. For example, he could not remember his graduated college, his occupation, the name of his employer, and the death of his grandparents. The same interview was also performed with the patient in the recovery state. At this state, the patient had regained his memories and had no difficulties with the structured interview covering past personal events.

Table 1
Neuropsychological test results of the patient.

Neuropsychological test	Amnesia state	Post-amnesia state
General intelligence		
RCPM	31/36	35/36
Frontal function		
Word fluency	11/min	17/min
Memory		
Digit span		
Forward	7	5
Backward	5	6
RAVLT-R		
Trials 1–5	2, 4, 7, 8, 11	3, 7, 11, 13, 14
Postinterference	8	4
30-min delayed recall	12	14
Rey–Osterrieth figure		
copy	36	34
Immediate recall	30	30.5
15-min delayed recall	29.5	31
WMS-R		
General memory index	105	109
Verbal memory index	105	104
Visual memory index	101	119
Attention/concentration index	95	85
Delayed index	104	117

RCPM, Raven's colored progressive matrices; WAIS-R, Wechsler adult intelligence scale revised; RAVLT-R, Rey auditory verbal learning test revised; WMS-R, Wechsler memory scale revised.

2.3. PET and MRI procedures

The PET system ECAT EXACT HR+ (CTI-Siemens, Knoxville, TN) was used to measure radioactivity in the brain in three-dimensional (3D) mode, which provides 63 planes and 15.5-cm field of view (FOV). To minimize head movement, a head fixation device (Fixster, Stockholm) was used. A transmission scan for attenuation correction was performed using a ^{68}Ge-^{68}Ga source. Acquisitions were performed in 3D mode with the interplane septa retracted.

After the transmission scan, a bolus of 176.5 to 237.9 MBq of [^{11}C] WAY-100635 with specific radioactivity of 42.6 to 400.0 GBq/μmol was injected intravenously into a cannula inserted in an antecubital vein. The cannula was then flushed by the rapid injection of 20 ml of saline. Radioactivity was measured for 90 min starting immediately after the injection. All emission scans were reconstructed with a Hanning filter cutoff frequency of 0.4 (full width half maximum [FWHM]=7.5 mm). T1-weighted MRI was acquired by Phillips Intera, 1.5 T (Philips Medical Systems, Best, Netherlands). T1-weighted images of the brain were obtained from all subjects. The scan parameters were 1 mm thick, 3D, T1 images with a transverse plane (repetition time [TR]/echo time [TE] 21/9.2 msec, flip angle 30, matrix 256 × 256, FOV 256 mm × 256 mm).

2.4. Quantification of 5-HT$_{1A}$ receptors and MR gray matter image

Parametric maps of binding potential (BP$_{ND}$) were calculated from dynamic [^{11}C] WAY-100635 PET images using the reference tissue compartment model with the cerebellar gray matter without vermis or venous sinus as reference tissue [simplified reference tissue method (SRTM) (Gunn et al., 1998)] using PMOD software (PMOD Technologies Ltd., Zurich, Switzerland). SRTM yields binding potential (BP$_{ND}$) estimates relative to the non-displaceable (ND) binding, which is denoted by BP$_{ND}$ (Innis et al., 2007). BP$_{ND}$ can also be described by the equation BP$_{ND}$=f$_{ND}$ B$_{Avail}$ (1/K$_D$), where B$_{Avail}$ is the density of receptors available to bind radioligand, 1/K$_D$ is the apparent affinity, and f$_{ND}$ is the fraction of free radiotracer in the non-displaceable tissue compartment. The model also allows the estimation of parametric maps of R1, the ratio of the delivery in the tissue ROI compared with that in the reference region (ratio of influx).

Spatial preprocessing of BP$_{ND}$ and R1 maps was performed using Statistical Parametric Mapping (SPM, Welcome Institute of Neurology, University College of London, UK). The ligand-specific template image (Meyer et al., 1999) was used to define the stereotactic transformation parameters for the binding potential images of [^{11}C]WAY-100635. Normalized binding potential images were smoothed with a Gaussian filter to 6 mm FWHM.

Normalized gray matter image maps were generated from each individual T1-weighted MRI using the VBM8 toolbox with SPM8 software. Normalized maps were spatially smoothed using an isotropic Gaussian filter (6 mm full-width at half-maximum).

2.5. Statistical analysis

Exploratory voxel-based analysis (VBA) was performed using SPM software. BP_{ND} and R1 images were compared between the patient and healthy subjects by Jack-knife analysis. Normalized and smoothed gray matter image maps were also compared with voxel-based analysis between the patient and healthy controls by Jack-knife analysis. Statistical inferences were made with a voxel-level threshold of $p < 0.05$, after family-wise error correction for multiple comparisons, with a minimum cluster size of 150 voxels.

To show the exact individual regional BP_{ND} values of the patient and HC, spherical VOIs of 5 mm radius were placed by using the coordinates of pixel
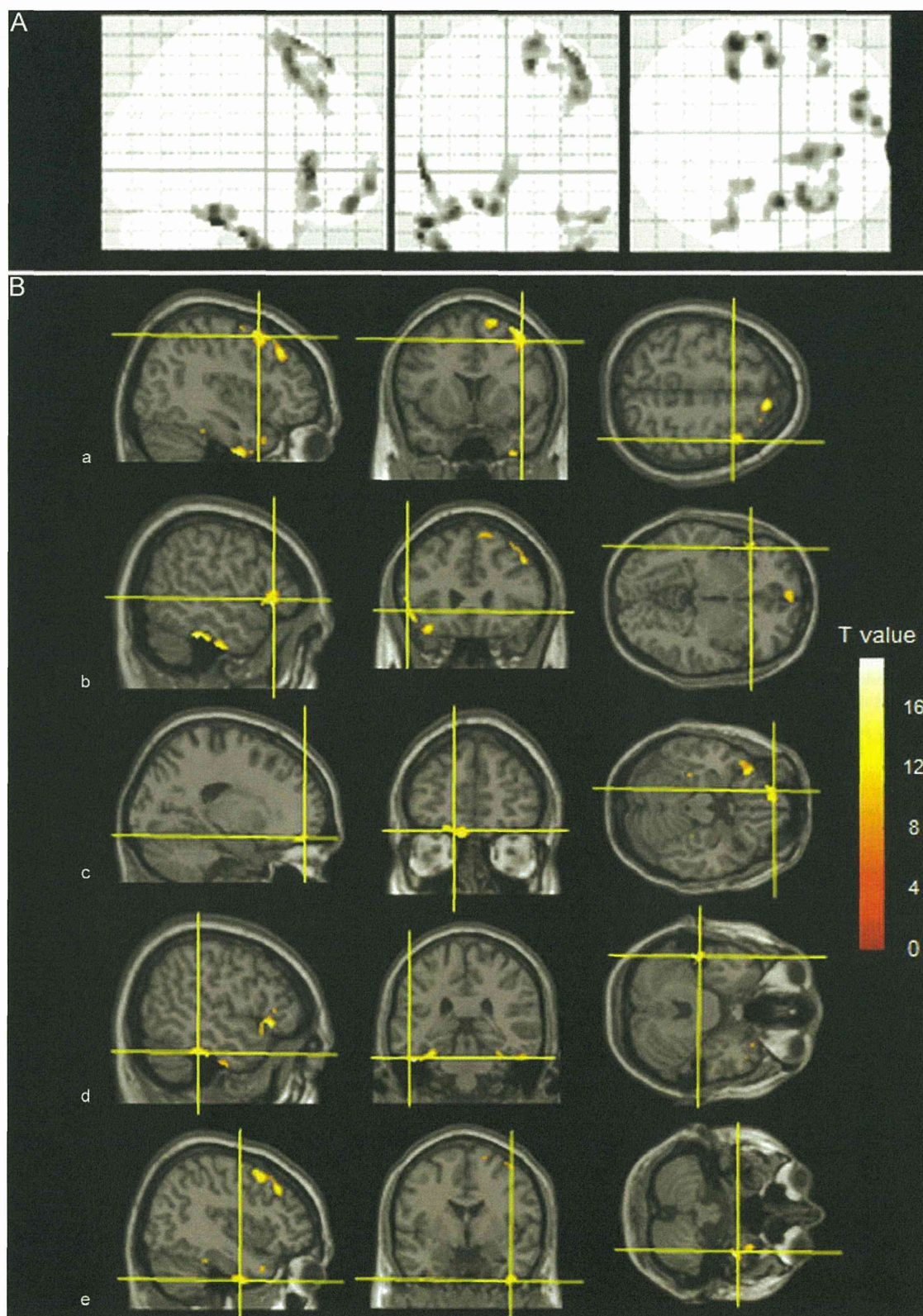

Fig. 1. Significant regional BPND elevation in dissociative amnesia patient of recovery state compared to HC in voxel-based analysis ($p < 0.05$, FWE correction for multiple comparison). Results are projected on three two-dimensional planes (A) and superimposed on representative magnetic resonance images (B). (a) right middle frontal cortex ($x=40$, $y=12$ and $z=56$), (b) left inferior frontal cortex ($x=-52$, $y=26$ and $z=-4$), (c) left orbitofrontal cortex ($x=-18$, $y=50$ and $z=-20$), (d) left inferior temporal cortex ($x=-50$ and $y=-32$).

Table 2
Regional changes in BP_{ND} of [^{11}C]WAY-100635 related to dissociative amnesia.

Comparison	Region	MNI coordinations (x, y, z)			Voxels	t-value	p Value (FWE corrected)
Patient < Controls	None						
Patient > Controls	Rt superior frontal cortex	12	36	56	253	14.6	< 0.001
	Rt middle frontal cortex	40	12	56	346	14.1	< 0.001
	Lt inferior frontal cortex	−52	26	−4	187	15.8	< 0.001
	Lt orbitofrontal cortex	−18	50	−20	307	14.4	< 0.001
	Lt inferior temporal cortex	−50	−32	−30	318	17.5	< 0.001
	Rt inferior temporal cortex	42	−2	−46	184	16.6	< 0.001

BP, binding potential; MNI, Montreal Neurological Institute; FEW, family wise error; Lt, left; Rt, right.

Table 3
BP_{ND} value of [^{11}C]WAY-100635 of the patient and healthy controls.

	Patient		Percent change of BP_{ND} [a]	HC mean (SD)
	Amnesia state	Recovery state		
Rt superior frontal cortex	3.25	3.36	3.2	2.69 (0.33)
Rt middle frontal cortex	3.22	4.05 [b]	25.9	2.69 (0.28)
Lt inferior frontal cortex	2.84	3.45 [b]	21.5	3.00 (0.43)
Lt orbitofrontal cortex	4.04	5.34 [b]	32.3	2.91 (0.45)
Lt inferior temporal cortex	4.85	6.03 [b]	24.3	3.67 (0.67)
Rt inferior temporal cortex	4.65	6.28 [b]	35.1	3.79 (0.56)

BP_{ND}; binding potential, SD; standard deviation, HC; healthy controls, Lt; left, Rt; right.

[a] (BP_{ND} in recovery state – BP_{ND} in amnesic state)∗100/BP_{ND} in amnesic state.

[b] BP values of patients at recovery state were beyond 10% than those of amensia state.

maxima as the centroid in regions where we found significant differences of BP_{ND} in the VBA. The values of BP_{ND} of both the patient and HC were calculated by averaging the values for all voxels within the VOIs. We examined whether the resulting BP_{ND} differences are beyond 10%, around which the test–retest reliability with this radioligand lies.

3. Results

3.1. Comparison of BP_{ND} between the patient and HC

In the VBA, there was no significant difference between the patient with amnesia state and HC. On the other hand, in the post-amnesia state, the patient showed significant BP_{ND} elevation of [^{11}C] WAY-100635 in the right superior frontal cortex, right middle frontal cortex, left inferior frontal cortex, left orbitofrontal cortex and bilateral inferior temporal cortex (Fig. 1, Table 2). There were no significantly lower BP_{ND} values of the patient than those of HC. There were no significant differences of R1 and gray matter volumes between HC and the patient at amnesic and post-amnesic states.

3.2. Difference in patient BP_{ND} values between amnesic and post-amnesic states

Spherical VOIs were placed on the BP_{ND} image by using the coordinates of pixel maxima as the centroid in the resultant regions above, where we found significant differences between the patient with post-amnesic state and HC in the VBA. The exact BP_{ND} values of each area of the patient at amnesic and post-amnesic states and HC are shown in Table 3. Except for the right inferior temporal cortex and right superior frontal cortex, the BP_{ND} values of recovery state were beyond 10% of those of amnesia state.

4. Discussion

In this study, we found that 5-HT$_{1A}$ receptor bindings of the patient in the right superior frontal cortex, left inferior frontal cortex, left orbitofrontal cortex and bilateral inferior temporal cortex at the post-amnesic state were significantly higher than those of HC by PET analysis. We found a significant increase in 5-HT$_{1A}$ receptor bindings of these regions in the patient during recovery from dissociative amnesia. Since [^{11}C] WAY-100635 binding was not sensitive to either acute or chronic changes of endogenous serotonin (Maeda et al., 2001), our finding might be attributable to the density of 5-HT$_{1A}$ receptors. The increase of BP_{ND} of [^{11}C]WAY-100635 is unlikely to be an effect of altered blood flow, since we found no significant increase in R1 values after recovery, and BP_{ND} from the reference tissue method is minimally dependent on tracer delivery over the R1 values obtained in this study (Lammertsma and Hume, 1996). This suggested that the increase in postsynaptic 5-HT$_{1A}$ receptors in these regions might have had an influence on the recovery from dissociative amnesia.

The function of 5-HT has been implicated in neural regulatory control, and 5-HT$_{1A}$ receptor is the main inhibitory serotonergic receptor subtype (Fink et al., 2008; Drueke et al., 2013). Post-synaptic 5-HT$_{1A}$ receptors are predominant on pyramidal neurons (Buhot et al., 2000), and it is reasonable to consider that an increase of postsynaptic 5-HT$_{1A}$ receptors on pyramidal neurons in the frontal and temporal regions in the patient during recovery from dissociative amnesia may have resulted in a decrease in pyramidal cell activity by their inhibitory effect in these regions.

There is an etiological model for dissociative amnesia, in which stress predominantly affects the control and executive functions of the frontal regions (Kopelman, 2000, 2002). In dissociative amnesia, the excessive activity of frontal regions causes interference with memory retrieval, which disappears in the recovery state (Kikuchi et al., 2010). It is considered that there is a possible interaction between prefrontal/orbitofrontal cortex and limbic

regions such as the hippocampus (Lanius et al., 2012; Kumfor et al., 2013), and frontal regions play a critical role in suppression of memory retrieval in dissociative amnesia.

The inferior temporal cortex is related to the visual recognition process, and it has a dense connection with the amygdala (Sabatinelli et al., 2005), which was reported to be activated in a PET activation study of a dissociative amnesic patient during a retrograde memory task (Yasuno et al., 2000). It is reasonable to posit that the hypothesized excessive activity of the amygdala of the patients induced the abnormal activity of the inferior temporal cortex, which may affect the symptom-related abnormality of the recognition of the past object. Taken together, the abnormal activity of frontal and temporal regions may play a role in the pathophysiology of dissociative amnesia.

The hypothesized abnormal activity of frontal and temporal regions might become normalized by the increased effect of 5-HT_{1A} on pyramidal neurons in these regions during the recovery process. The level of postsynaptic 5-HT_{1A} receptor binding in the observed regions was above the normal range five months after the onset, when he had recovered from amnesia and reacquired all his memories. We speculate that up-regulation of the binding is transient and is a compensatory change for the abnormal activity of prefrontal and temporal regions due to the emotional stress. Unfortunately, the patient was lost to follow-up and we cannot determine whether and when 5-HT_{1A} receptors were normalized after his recovery from dissociative amnesia.

In conclusion, our findings suggested that the increase of postsynaptic 5-HT_{1A} receptors in frontal and temporal regions has a role in the recovery from dissociative amnesia. Our findings may give rise to the possibility that drugs that work as agonists on postsynaptic 5-HT_{1A} receptors in the frontal and temporal regions lead to improvement of dissociative symptoms, although this improvement has the risk of being compromised by a negative effect on memory function due to suppression of hippocampal function by the systemic administration of 5-HT_{1A} agonist (Yasuno et al., 2003). Ours is a single-case study and the results cannot be interpreted generally. Further investigations of a larger number of patients and control subjects including the use of functional/molecular imaging with the administration of agonists of 5-HT_{1A} receptors are required to better understand the abnormal functions in dissociative amnesia.

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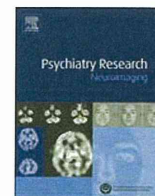
Microstructural changes of the nucleus accumbens due to increase of estradiol level during menstrual cycle contribute to recurrent manic episodes—A single case study

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ARTICLE INFO

ABSTRACT

We examined a rapid-cycling bipolar disorder patient who demonstrated manic episode regularly at around day 7 of the menstrual cycle. We hypothesize that gonadal hormones may induce a state-dependent change in cerebral microstructure and function. Following this hypothesis, the serum levels of estradiol and progesterone were analyzed and diffusion tensor imaging data were examined between the manic and euthymic states of the patient. Estradiol levels increased in the late follicular phase at manic state when compared to the luteal or early follicular phase at euthymic state. DTI results showed that the patient had increased fractional anisotropy values at manic state in the bilateral nucleus accumbens (NAc) and its connected areas, which is a major projection field of the mesolimbic dopamine (DA) system, perhaps reflecting microstructural changes due to neuronal activation related to manic episodes. According to these results, we consider that the mesolimbic DA system of this patient has hypersensitivity to estradiol, and elevation of the estradiol level increases the activity of the dopaminergic system, which in turn may contribute to recurrent manic episodes. Our findings provide a clue for understanding how fluctuations in gonadal hormone may amplify or ameliorate the symptomatology of psychiatric disorders related to the menstrual cycle.

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1. Introduction

Affective fluctuations during the menstrual cycle have been studied (Akdeniz and Karadağ, 2006). A few previous case presentations of patients, including a report of a woman with bipolar disorder (BPD), showed experienced specific mood episodes in certain periods of the menstrual cycle, such as the premenstrual period (Kukopulos et al., 1985; D'Mello et al., 1993) and luteal phase (Becker et al., 2004). However, the mechanisms underlying the illness phases related to the menstrual cycle have not been investigated. In the present study we report a rapid-cycling bipolar disorder patient who regularly demonstrated manic episode starting in the follicular phase and continuing for about 2 weeks.

In our case of recurrent manic episodes related to the phases of the menstrual cycle, we hypothesize that fluctuations of gonadal hormones may induce a state-dependent change in cerebral microstructure and function that result in a recurrence of the manic symptoms. According to this hypothesis, the serum levels of estradiol and progesterone were analyzed at manic and euthymic states of the patient. In order to elucidate the regional microstructural changes related to manic symptoms, we performed exploratory voxel-based analysis and compared DTI images between the patient and healthy subjects. We expected that the patient would show manic state-dependent brain microstructural changes in the regions related to manic symptoms, which were affected by the fluctuations of gonadal hormones related to the phases of the menstrual cycle.

2. Materials and methods

2.1. Patient and healthy control subjects

The patient was a 32-year-old right-handed woman. She had no history of alcohol or illicit drug abuse. Since age 21, she had recurrent manic episodes every

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