Associations of Serum Serotonin Levels with 12-week and 12-month Remission in Patients with Depressive Disorders

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Objective: To investigate associations between baseline serum serotonin levels and short- and long-term treatment outcomes in outpatients with depressive disorders in a naturalistic one-year prospective study design.
Methods: Patients were recruited at a University hospital in South Korea from March 2012 to April 2017. At baseline, blood samples were obtained from 1,094 patients who received initial antidepressant monotherapy (Step 1). After the Step 1 treatment, further treatment steps (at least Steps 2−4) could be administered every 3 weeks during the acute treatment phase (3, 6, 9, and 12 weeks; n = 1,086), and every 3 months during the continuation treatment phase (6, 9, and 12 months; n = 884). In cases showing an insufficient response or intolerable side effects, patients were asked to choose whether to remain at the current step or enter the next treatment step, with alternative strategies including switching, augmentation, combination, and a mixture of these approaches. Remission was defined as a Hamilton Depression Rating Scale score of ≤ 7.
Results: The remission group had significantly higher baseline serum serotonin levels among patients who received Step 1 monotherapy in both acute and continuation treatment phases. These associations remained significant after adjustment for relevant covariates. No associations were found with any other treatment steps.
Conclusion: Baseline serum serotonin levels may be used as a biomarker for predicting short- and long-term treatment outcomes in antidepressant monotherapy-treated patients with depressive disorders in a real-world clinical setting.

KEY WORDS: Depression; Serotonin; Treatment; Antidepressant; Remission induction.

INTRODUCTION

Depressive disorders are one of the most serious public health problems globally, affecting more than 250 million people worldwide [1,2]. Antidepressants are the first-line treatment, particularly for moderate to severe major depressive disorder (MDD) [3]. Currently used antidepressants have similar efficacy to each other and are associated with remission rates of less than one third of patients after 8−12 weeks of first trial [2-5]. Considering its high disease burden and low initial antidepressant remission rate, it is necessary to develop biomarkers that predict antidepressant outcomes that could aid in deciding personalized treatment strategies in depressive disorders. Based on the main hypotheses pertaining to the biological mechanisms of depression, such as the monoamine transmitter hypothesis [6], the inflammatory hypothesis [7], the neurotrophic hypothesis [8], and the hypothalamic-pituitary-adrenal axis dysfunction hypothesis [9], efforts have been made to develop peripheral biomarkers of antidepressant response. However, clinically meaningful biomarkers for differentiation between treatment strategies have not yet been developed [10].

Although the detailed pharmacological mechanisms of antidepressants remain unclear, most types of antidepressants are thought to upregulate serotonin (5-hydroxytryptamine, 5-HT) levels in the synaptic cleft [11]. 5-HT is a monoamine neurotransmitter that regulates various

physiological functions in the central and periphery (gastrointestinal tract, cardiovascular system, immune system, endocrine system, etc.). By acting as a neurotransmitter in the central nervous system (CNS), 5-HT regulates mood [12], sleep-wake behavior [13], and appetite [14].

Taking into account the proposed mechanism of action for antidepressants, there have been efforts to explore peripheral blood 5-HT as a biomarker for antidepressant response. Recent studies have shown that higher baseline plasma 5-HT levels before selective serotonin reuptake inhibitor (SSRI) treatment were associated with a better treatment response [15,16]. They hypothesized that these associations might be related to 5-HT_{1A} autoreceptor hypofunctioning [17,18]. In the CNS, 5-HT negatively regulates the activity of the 5-HT system through 5-HT_{1A} autoreceptor signaling [19]. Because 5-HT_{1A} autoreceptor hypofunctioning leads to increased 5-HT in the synapse and is associated with a better antidepressant response [17,18], they speculated that increased pre-treatment plasma 5-HT levels might be related to 5-HT_{1A} autoreceptor hypofunctioning. Since these studies evaluated the associations between 5-HT levels and treatment responses for up to 8 weeks in MDD patients treated with SSRIs, associations between 5-HT levels and long-term outcomes in patients using various types of antidepressant in real-world clinical situations are not known.

In the present study, by using data from a naturalistic prospective study of Korean patients with depressive disorder, we investigated associations between baseline serum 5-HT levels and short- and long-term remission in patients using various treatment strategies based on early clinical decisions.

METHODS

Study Outline

This study was carried out as a component of the MAKE Biomarker discovery for Enhancing antidepressant Treatment Effect and Response (MAKE BETTER) program. Details of the study have been published as a design paper [20] and registered with cris.nih.go.kr (identifier: KCT0001332). Data on socio-demographic and clinical characteristics were obtained using a structured clinical report form (CRF) by clinical research coordinators, who were blind to treatment modalities. They were trained in CRF implementation and data collection methods by the research psychiatrists. Patients' data were recorded on a CRF, registered in the website of the MAKE BETTER study (http://icreat.nih.go.kr/icreat/webapps/com/hismainweb/jsp/cdc_n2.live) within 3 days, and monitored by data management center personnel. All patients gave written informed consent to participate in the study and use their data. The study was conducted in accordance with the Helsinki Declaration of 1975, as revised in 2013 and approved by the Ethics Commission of the Chonnam National University Hospital Institutional Review Board (CNUH 2012-014) as it uses de-identified data.

Participants

Patients with depressive disorders were consecutively recruited from March 2012 to April 2017 from those who had visited the outpatient psychiatric department of Chonnam National University Hospital. Research psychiatrists assessed and diagnosed depressive disorders using the Mini-International Neuropsychiatric Interview [21], a structured diagnostic psychiatric interview based on the Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition (DSM-IV) criteria. As the aim of the study was to reflect a real-world clinical setting as closely as possible, broad inclusion criteria and minimal exclusion criteria were applied. Inclusion criteria were: i) aged older than 7 years; ii) diagnosed with MDD, dysthymic disorder, or depressive disorder not otherwise specified (NOS); iii) Hamilton Depression Rating Scale (HAMD) [22] score ≥ 14; iv) able to complete questionnaires, understand the objective of the study, and sign the informed consent form. Exclusion criteria were as follows: i) unstable or uncontrolled medical condition; ii) unable to complete the psychiatric assessment or comply with the medication regimen, due to a severe physical illness; iii) current or lifetime DSM-IV diagnosis of bipolar disorder, schizophrenia, schizoaffective disorder, schizophreniform disorder, psychotic disorder NOS, or other psychotic disorder; iv) history of organic psychosis, epilepsy, or seizure disorder; v) history of anticonvulsant treatment; vi) hospitalization for any psychiatric diagnosis apart from depressive disorder (e.g., alcohol/drug dependence); vii) electroconvulsive therapy received for the current depressive episode; viii) pregnant or breastfeeding. All participants reviewed the consent form and written informed consent was obtained. For participants aged under 16, written consent was obtained from a parent or legal

guardian, and written assent was obtained from the participant.

Measurements at Baseline

Socio-demographic characteristics obtained comprised age, sex, years of formal education, marital status (currently married or not), cohabiting status (living alone or not), religion (religious observation or not), occupation (current employed status or not), and income (above or below 2,000 USD). Clinical characteristics evaluated were comprised of diagnoses of depressive disorders as mentioned above with certain specifiers, age at onset and duration of illnesses, history of previous depressive episodes (recurrent or first episode), number of previous depressive episodes, duration of present episode, family history of depression, history of suicide attempt, and number of concurrent physical disorders (applying a questionnaire enquiring about 15 different systems or disorders). Assessment scales for investigating symptoms and function were administered. Depressive symptoms were evaluated by the HAMD, anxiety symptoms by of the Hospital Anxiety Depression Scale-anxiety subscale (HADS-A) [23], quality of life by the EuroQol-5D (EQ-5D) [24], and functioning levels by the Social and Occupational Functioning Assessment Scale (SOFAS).

Blood Sampling and Assays

Participants were instructed to have fasted (except water) since the night before for blood sampling. They were to sit quietly and relax for 25–45 minutes before blood samples were obtained. Among 1,262 study participants, 1,094 subjects agreed to offer blood samples in the baseline evaluation. Serum 5-HT levels were measured using ClinRep high-performance liquid chromatography kit (Recipe, Munich, Germany) at GreenCross LabCell (Yongin, Korea).

Treatment

Details of the treatment in this study have been previously published [25]. Before treatment commencement, a comprehensive review was made of the patients' clinical manifestations (e.g., psychotic and anxiety symptoms), severity of illness, physical comorbidities and medication profiles, and history of previous treatments. Minimal and maximal dosages of pharmacological agents were determined considering existing treatment guide-

lines [26,27]. In the first treatment Step 1, patients received antidepressant treatment, taking into consideration these data and treatment guidelines [27-29], for 3 weeks. Antidepressants used were bupropion, desvenlafaxine, duloxetine, escitalopram, fluoxetine, mirtazapine, paroxetine, sertraline, venlafaxine, and vortioxetine. After Step 1 antidepressant monotherapy, next step pharmacotherapy could be administered every 3 weeks during the acute treatment phase (3, 6, 9, and 12 weeks with a 3-day allowable window) and every 3 months during the continuation treatment phase (6, 9, and 12 months with a 7-day allowable window), whenever needed. At the end of each step, overall effectiveness and tolerability were reviewed for proceeding with measurement-based next-step treatments. In cases of insufficient improvement (a HAMD score reduction of < 30% from the baseline) or intolerable side effects, patients were instructed to choose whether they would prefer to remain in the current step or enter the next step strategies with switching (S), augmentation (A), combination (C), S + A, S + C, A + C, and S + A + C treatment. Patients were also allowed to receive next step treatment if they showed sufficient improvement (a HAMD score reduction of ≥ 30% from the baseline) and absent/tolerable side effects. For determining treatment strategies, each patient's preference was given priority to maximize medication compliance and treatment outcomes [30]. Antidepressants switched or combined were bupropion, desvenlafaxine, duloxetine, escitalopram, fluoxetine, mirtazapine, paroxetine, sertraline, venlafaxine, and vortioxetine. Augmented drugs were buspiron, lithium, triiodothyronine, and atypical antipsychotics including aripiprazole, risperidone, olanzapine, quetiapine, and ziprasidone. Since the number entered into Step 5 or above was small, treatment steps were classified into Step 1, 2, 3, and 4 (including Step 5+) in the analysis.

Definition of the Outcome

Remission was defined as a HAMD score ≤ 7. Remission at 12 weeks and at 12 months was used in order to investigate the associations of serum 5-HT levels with short- and long-term treatment outcomes, respectively.

Statistical Analysis

Baseline data were compared by remission status in the acute treatment phase (up to 12 weeks) and continuation treatment phase (up to 12 months) using independent

t tests or chi-square tests. The unadjusted associations of the initially prescribed antidepressant type with 12-week and 12-month remission were investigated using chi-square tests. Antidepressants were classified as SSRIs (escitalopram, sertraline, paroxetine, fluoxetine) and non-SSRIs, and included a noradrenergic and specific serotonin antidepressant (NaSSA; mirtazapine), serotonin-norepinephrine reuptake inhibitors (SNRIs; venlafaxine, duloxetine, desvenlafaxine), a norepinephrine-dopamine reuptake inhibitor (NDRI; bupropion), and a serotonin modulator and stimulator (vortioxetine). Serum 5-HT levels were compared by the remission status in treatment steps using independent *t* tests. Serum 5-HT levels were compared between the four treatment steps (Step 1, 2, 3, and 4) up to 12 weeks and up to 12 months using analysis of variance with *post-hoc* Scheffe's tests. Moreover, serum 5-HT levels were compared by the remission status in treatment steps using independent *t* tests.Relationships between baseline serum 5-HT levels (categorized as per 20 ng/ml increase) and 12-week and 12-month remission status were analyzed using binary logistic regression after adjustment for covariates that significantly differed (*p* value < 0.05) in the baseline data, including age, monthly income, age at onset, number of depressive episodes, dura-

tion of present episode, HAMD, HADS-A, and SOFAS. All statistical tests were two-sided with a significance level of 0.05. Statistical analysis were performed using IBM SPSS Statistics version 25 (IBM Co., Armonk, NY, USA).

RESULTS

Recruitment and Treatment Flow

Patient flow by treatment steps over a 12-month period is described in Figure 1. Among 1,262 patients evaluated at baseline, blood samples were obtained in 1,094 (86.7%) and 1,086 (86.1%) who were followed up at least once during the 12-week treatment period. Reasons for drop-out were: lack of treatment effect (n = 4) and loss to follow-up (n = 4). A total of 1,086 patients, included for a 12 week analysis, had lower rates of suicide attempt history but higher scores of HADS-A and EQ-5D compared to 176 in the baseline. At the 12-week assessment point, 463 (42.6%) patients received Step 1 antidepressant monotherapy treatment, 360 (33.1%) received Step 2 treatment, 200 (18.4%) received Step 3 treatment, and 63 (5.8%) received Step 4 treatment.

After the acute treatment phase, 884 (81.4%) were followed up at least once, up to the 12-month follow-up and
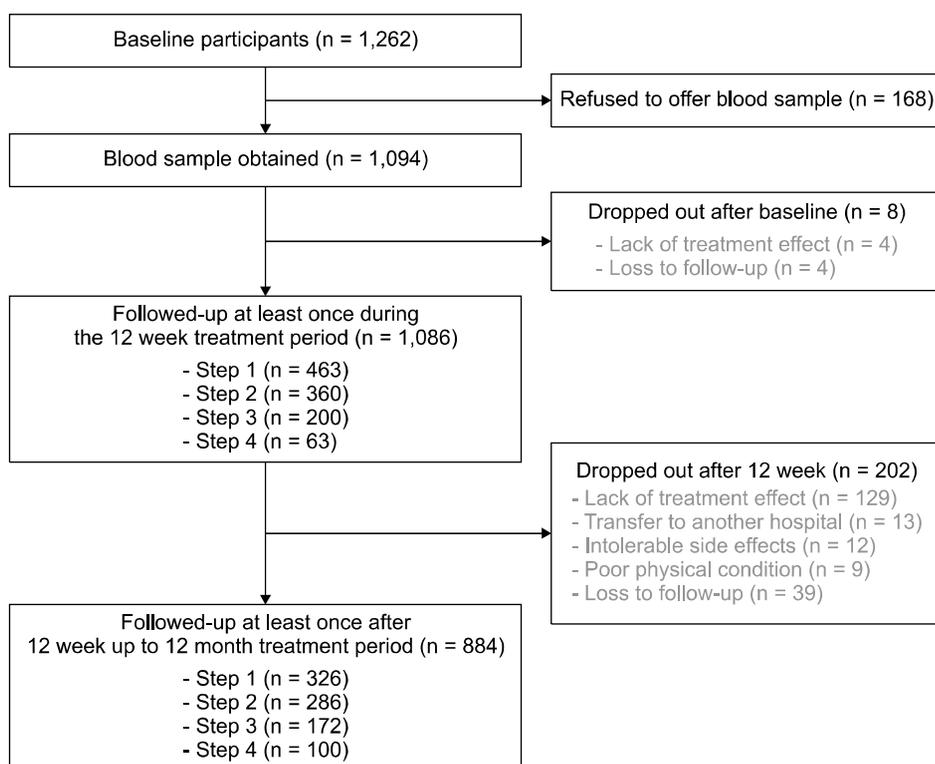

Fig. 1. Participant recruitment and treatment.

an analysis of remission in the continuation treatment phase was performed. Reasons for drop-out were: lack of treatment effect (n = 129), transfer to another hospital (n = 13), intolerable side effects (n = 12), poor physical condition (n = 9), and loss to follow-up (n = 39). Attrition at 12 months was significantly associated with unemployed status, melancholic features, and higher EQ-5D scores at baseline. However, drop-out at 12 months was not associated with 5-HT level ($t = 1.114$, $p = 0.266$). At the 12-month assessment point, 326 (36.9%) patients had received Step 1 treatment, 286 (32.4%) had received Step 2 treatment, 172 (19.5%) had received Step 3 treatment, and 100 (11.3%) had received Step 4 treatment.

Baseline Characteristics by Remission Status

Baseline characteristics by remission status in patients up to 12 weeks of treatment (acute treatment phase) and up to 12 months of treatment (continuation treatment phase) are compared in Table 1. Age ($t = 2.610$, $p = 0.009$), monthly income ($\chi^2 = 5.801$, $p = 0.016$), age at onset ($t = -3.042$, $p = 0.002$), duration of present episode ($t = 3.128$, $p = 0.002$), HAMD ($t = 2.418$, $p = 0.016$), HADS-A ($t = 3.666$, $p < 0.001$), and SOFAS ($t = -4.785$, $p < 0.001$) were significantly different between the 12-week remission group and no remission group. Monthly income ($\chi^2 = 4.465$, $p = 0.035$), number of depressive episodes ($t = 3.589$, $p < 0.001$), and HADS-A ($t = 2.521$, $p = 0.012$) were significantly different between the 12-month remission group and no remission group. There was a general pattern of better clinical presentations associated with the remission group in both the acute and continuation treatment phase. The types of antidepressants

Table 1. Baseline characteristics by 12-week and 12-month remission in blood obtained from patients with depressive disorders

Characteristics	Up to 12-week treatment (n = 1,086)				Up to 12-month treatment (n = 884)			
	No remission (n = 596)	Remission (n = 490)	Statistical coefficients[a]	*p* value	No remission (n = 259)	Remission (n = 625)	Statistical coefficients[a]	*p* value
Socio-demographic characteristics								
Age (yr)	55.9 ± 15.6	58.2 ± 13.9	$t = 2.610$	0.009	56.1 ± 16.0	57.3 ± 14.2	$t = -1.048$	0.295
Sex, female	411 (69.0)	334 (68.2)	$\chi^2 = 0.079$	0.778	173 (66.8)	434 (69.4)	$\chi^2 = 0.595$	0.440
Education (yr)	9.1 ± 4.7	9.1 ± 4.9	$t = -0.065$	0.948	9.3 ± 4.7	9.0 ± 4.8	$t = 1.090$	0.276
Marital status, unmarried	192 (32.2)	134 (27.3)	$\chi^2 = 3.033$	0.082	83 (32.0)	174 (27.8)	$\chi^2 = 1.571$	0.210
Living alone	94 (15.8)	73 (14.9)	$\chi^2 = 0.158$	0.691	43 (16.6)	88 (14.1)	$\chi^2 = 0.923$	0.337
Religious observance	326 (54.7)	281 (57.3)	$\chi^2 = 0.765$	0.382	141 (54.4)	355 (56.8)	$\chi^2 = 0.414$	0.520
Unemployed status	186 (31.2)	130 (26.5)	$\chi^2 = 2.852$	0.091	82 (31.7)	160 (25.6)	$\chi^2 = 3.383$	0.066
Monthly income, < 2,000 USD	375 (62.9)	273 (55.7)	$\chi^2 = 5.801$	0.016	167 (64.5)	355 (56.8)	$\chi^2 = 4.465$	0.035
Clinical characteristics								
Major depressive disorder	510 (85.6)	415 (84.7)	$\chi^2 = 0.164$	0.686	227 (87.6)	534 (85.4)	$\chi^2 = 0.743$	0.389
Melancholic feature	95 (15.9)	67 (13.7)	$\chi^2 = 1.088$	0.297	45 (17.4)	96 (15.4)	$\chi^2 = 0.554$	0.457
Atypical feature	35 (5.9)	34 (6.9)	$\chi^2 = 0.514$	0.473	16 (6.2)	39 (6.2)	$\chi^2 = 0.001$	0.972
Age at onset (yr)	50.5 ± 17.3	53.6 ± 15.7	$t = -3.042$	0.002	50.6 ± 17.7	52.3 ± 16.1	$t = -1.328$	0.185
Duration of illness (yr)	5.4 ± 9.3	4.7 ± 8.7	$t = 1.311$	0.190	5.5 ± 9.0	5.0 ± 9.1	$t = 0.739$	0.460
Recurrent depression	322 (54.0)	248 (50.6)	$\chi^2 = 1.257$	0.262	145 (56.0)	321 (51.4)	$\chi^2 = 1.571$	0.210
Number of depressive episodes	1.2 ± 1.5	1.0 ± 1.4	$t = 1.836$	0.067	1.4 ± 1.8	1.0 ± 1.3	$t = 3.589$	< 0.001
Duration of present episode (mo)	8.3 ± 12.0	6.4 ± 8.0	$t = 3.128$	0.002	8.3 ± 12.2	7.3 ± 9.5	$t = 1.138$	0.256
Family history of depression	82 (13.8)	76 (15.5)	$\chi^2 = 0.664$	0.415	32 (12.4)	93 (14.9)	$\chi^2 = 0.961$	0.327
History of suicide attempt	56 (9.4)	39 (8.0)	$\chi^2 = 0.695$	0.404	27 (10.4)	45 (7.2)	$\chi^2 = 2.545$	0.111
Number of physical disorders	1.6 (1.3)	1.7 (1.2)	$t = -1.212$	0.226	1.7 (1.3)	1.7 (1.3)	$t = 0.021$	0.983
Assessment scales (scores)								
Hamilton Depression Rating Scale	21.0 ± 4.1	20.4 ± 4.1	$t = 2.418$	0.016	20.8 ± 4.2	20.1 ± 4.1	$t = 0.427$	0.669
Hospital Anxiety & Depression Scale-anxiety subscale	12.2 ± 4.0	11.3 ± 4.1	$t = 3.666$	< 0.001	12.3 ± 4.0	11.6 ± 4.0	$t = 2.521$	0.012
EuroQol-5D	9.0 ± 1.6	8.8 ± 1.4	$t = 1.767$	0.078	9.0 ± 1.5	8.8 ± 1.5	$t = 1.607$	0.108
Social and Occupational Functional Assessment Scale	55.0 ± 7.6	57.1 ± 7.1	$t = -4.785$	< 0.001	55.6 ± 7.0	56.3 ± 7.3	$t = -1.028$	0.227

Values are presented as mean ± standard deviation or number (%).
[a]Independent two sample *t* test or χ^2 tests, as appropriate.

Table 2. Baseline serum 5-HT levels by 12-week and 12-month remission and treatment steps

Treatment steps	No remission		Remission		Statistical coefficients[a]	p value
	Number	Value	Number	Value		
Up to 12-week treatment						
All participants	596	76.8 ± 50.2	490	83.4 ± 50.2	$t = -2.083$	0.037
Step 1	291	81.2 ± 50.6	172	92.4 ± 52.6	$t = -2.276$	0.023
Step 2	184	77.8 ± 50.1	176	83.5 ± 52.8	$t = -1.129$	0.259
Step 3	94	67.7 ± 45.8	106	73.6 ± 56.5	$t = -0.812$	0.418
Step 4	27	57.2 ± 54.0	36	69.4 ± 68.6	$t = -0.762$	0.449
Up to 12-month treatment						
All participants	259	75.6 ± 51.7	625	80.3 ± 52.3	$t = -1.216$	0.224
Step 1	112	75.6 ± 50.6	214	89.1 ± 49.4	$t = -2.330$	0.020
Step 2	83	78.7 ± 48.5	203	82.0 ± 51.8	$t = -0.486$	0.627
Step 3	32	82.0 ± 61.4	140	73.0 ± 51.6	$t = 0.774$	0.443
Step 4	32	61.4 ± 53.1	68	62.9 ± 58.6	$t = -0.119$	0.906

Values are presented as mean ± standard deviation.
5-HT, 5-hydroxytryptamine.
[a]Independent two sample t test.

initially prescribed are compared between those with and without remission in Supplementary Table 1 (available online). There was no difference in frequency of the types of antidepressants initially prescribed between the remission and no remission groups in both acute and continuation treatment phase. Considering these findings and potential collinearity between the variables, as well as previous reports on remission predictors [4,31], eight variables (age, monthly income, age at onset, number of depressive episodes, duration of present episode, HAMD, HADS-A, and SOFAS) were included as covariates in the later adjusted analysis.

Baseline Serum 5-HT Levels by Treatment Steps

Baseline serum 5-HT levels by treatment steps are compared in Supplementary Table 2 (available online). Baseline serum 5-HT levels were significantly different between treatment steps in both the acute treatment phase (F = 5.597, p = 0.001) and continuation treatment phase (F = 5.176, p = 0.002). In *post-hoc* comparisons, patients who received Step 1 treatment showed higher baseline serum 5-HT levels compared to patients who received Step 3 or Step 4 treatment in the acute treatment phase. Similar to the acute treatment phase, patients who received Step 1 or Step 2 treatment showed higher baseline serum 5-HT levels than patients who received Step 4 treatment in the continuation treatment phase. Taken together, baseline serum 5-HT levels were higher in the lower step treatment regardless of the treatment period.

Baseline Serum 5-HT Levels by Remission Status and Treatment Steps

Baseline serum 5-HT levels by 12-week and 12-month remission and treatment steps are compared in Table 2. Up to 12 weeks of treatment, the remission group showed higher baseline serum 5-HT levels compared with the no remission group ($t = -2.083$, $p = 0.037$). Among treatment steps in the acute treatment phase, not in patients who received Step 2 ($t = -1.129$, $p = 0.259$), Step 3 ($t = -0.812$, $p = 0.418$), and Step 4 ($t = -0.762$, $p = 0.449$), but among those in Step 1 ($t = -2.276$, $p = 0.023$) treatment, higher baseline serum 5-HT levels in the remission group compared to the no remission group were shown. Up to 12 months of treatment, there was no difference of baseline serum 5-HT levels between the remission group and no remission group ($t = -1.216$, $p = 0.224$). However, in the patients who received Step 1 treatment, baseline serum 5-HT levels were higher in the remission group compared to the no remission group ($t = -2.330$, $p = 0.020$). These data indicate that baseline serum 5-HT levels of the remission group are higher than those of the no remission group in patients who received Step 1 treatment, regardless of the treatment period.

Adjusted Associations of Baseline Serum 5-HT Levels with 12 Week and 12 Month Remission

Unadjusted and adjusted associations of baseline serum 5-HT levels with 12-week and 12- month remission by treatment steps are summarized in Table 3. Baseline

Table 3. Associations of baseline serum 5-HT levels with 12-week and 12-month remission

Treatment steps	Unadjusted OR (95% CI)	*p* value	Adjusted OR (95% CI)	*p* value
12-week remission				
All participants (n = 1,086)	1.050 (1.004−1.099)	0.034	1.061 (1.012−1.112)	0.015
Step 1 (n = 463)	1.097 (1.020−1.180)	0.013	1.135 (1.048−1.230)	0.002
Step 2 (n = 360)	1.041 (0.961−1.128)	0.320	1.040 (0.957−1.131)	0.355
Step 3 (n = 200)	1.037 (0.930−1.155)	0.516	1.065 (0.946−1.198)	0.298
Step 4 (n = 63)	1.073 (0.909−1.266)	0.403	1.095 (0.895−1.339)	0.378
12-month remission				
All participants (n = 884)	1.033 (0.976−1.093)	0.260	1.039 (0.980−1.101)	0.200
Step 1 (n = 326)	1.130 (1.025−1.246)	0.014	1.133 (1.021−1.258)	0.019
Step 2 (n = 286)	1.011 (0.915−1.118)	0.828	1.019 (0.918−1.132)	0.719
Step 3 (n = 172)	0.929 (0.808−1.067)	0.297	0.931 (0.799−1.084)	0.358
Step 4 (n = 100)	1.018 (0.877−1.182)	0.815	1.034 (0.870−1.228)	0.705

5-HT, 5-hydroxytryptamine; CI, confidence interval.
Adjusted odds ratio (OR) after adjustment for age, monthly income, age at onset, number of depressive episodes, duration of present episode, Hamilton Depression Rating Scale, Hospital Anxiety and Depression Scale-anxiety subscale, and Social and Occupational Functional Assessment Scale.

serum 5-HT levels showed positive associations with 12-week remission. Adjusted logistic regression analysis, after adjustment for seven variables mentioned above, showed that high baseline serum 5-HT levels are significantly associated with 12-week remission (odds ratio [OR] = 1.061, $p = 0.015$). Among treatment steps, not in patients who received Step 2 (OR = 1.040, $p = 0.355$), Step 3 (OR = 1.065, $p = 0.298$), and Step 4 (OR = 1.095, $p = 0.378$), but among those in Step 1 (OR = 1.061, $p = 0.015$) treatment, positive associations between baseline serum 5-HT levels and 12-week remission were shown. Unlike the 12-week remission, there was no association between 12-month remission and baseline serum 5-HT levels (OR = 1.039, $p = 0.200$). However, patients who received Step 1 treatment showed positive associations between baseline serum 5-HT levels and 12-month remission in both unadjusted (OR = 1.130, $p = 0.014$) and adjusted (OR = 1.133, $p = 0.019$) logistic regression analysis. These data demonstrate that baseline serum 5-HT levels are positively associated with 12-week and 12-month remission in patients who received Step 1 treatment.

DISCUSSION

In the present study, by using data from a naturalistic prospective study, which reflects real-world clinical practice, we determined that baseline serum 5-HT levels were higher in the remission group, and that high baseline se-

rum 5-HT levels are associated with 12-week and 12-month remission in patients with depressive disorders who are treated with antidepressant monotherapy. In addition, baseline serum 5-HT levels were higher in lower treatment steps regardless of treatment period.

Results from the acute treatment phase, particularly in patients who received Step 1 treatment, showed that baseline serum 5-HT levels were higher in the remission group and high baseline serum 5-HT levels were associated with 12-week remission. These results are similar to previous studies, which observed that baseline plasma 5-HT levels are higher in responders, and that high baseline plasma 5-HT levels are associated with 4- and 8-week outcomes in patients who received SSRI monotherapy [15,16]. Going one step further than previous studies, we demonstrated the utility of baseline 5-HT levels as a biomarker for treatment outcomes not only in patients treated with SSRIs, but also in those treated with other types of antidepressants, such as NaSSAs, SNRIs, NDRIs, and serotonin modulators and stimulators. Since various types of antidepressants are used as initial treatment for depressive disorders in real-world clinical practice, our results shed light on using baseline 5-HT levels as a biomarker for predicting treatment outcomes in patients receiving antidepressant monotherapy. Although our findings are novel, care should be taken in their interpretation because type of antidepressant was not controlled for and there were many drop-outs during the continuation treatment phase. Further large-scale randomized studies are needed

to confirm the results.

In this study, we have newly discovered that high baseline serum 5-HT levels are associated not only with short-term outcomes but also with long-term outcomes in patients who receive Step 1 treatment. Although there were no significant differences of baseline serum 5-HT levels between the remission group and no remission group among all participants in the continuation phase, the remission group had higher baseline serum 5-HT levels in the Step 1 treatment subgroup. Furthermore, high baseline serum 5-HT levels were associated with 12-month remission in patients who received Step 1 treatment. These findings suggest that baseline serum 5-HT levels might be used as a biomarker for predicting long-term outcomes as well as short-term outcomes in patients who receive antidepressant monotherapy.

Based on our results, baseline serum 5-HT levels seem to lose effectiveness as a biomarker for predicting treatment outcomes as the treatment strategy becomes complicated in depressive disorders. Among the treatment steps, only Step 1 treatment showed associations between high baseline serum 5-HT levels and remission in both the acute and continuation treatment phase. In addition, patients who were treated by simple strategies had higher baseline serum 5-HT levels compared to patients treated by complicated strategies in both acute and continuation treatment phases. From these results, we speculate that baseline serum 5-HT level can function as a biomarker for treatment outcomes only when it is sufficiently high. However, further investigations are needed to clarify this idea.

Several issues should be borne in mind before drawing a conclusion. First, central and peripheral 5-HT systems are functionally separated and correlation between peripheral blood and cerebrospinal fluid 5-HT levels are inconclusive [32,33]. However, results from our study and others [15,16] show that baseline peripheral blood 5-HT levels are associated with treatment outcomes in patients who receive antidepressant monotherapy. Further studies are needed to uncover the detailed mechanisms of how high peripheral blood 5-HT levels are connected with better antidepressant outcomes. Second, we measured 5-HT in serum rather than plasma. In the periphery, most of 5-HT is synthesized by enterochromaffin cells in the gut [34,35]. Once released from the gut, most 5-HT is taken up into platelets (> 95%) by 5-HT transporter, with the re-

maining free 5-HT levels very low in circulation [36-38]. While plasma 5-HT levels reflect only the bioactive free 5-HT in the peripheral blood, serum 5-HT levels reflect some portion of platelet 5-HT [39]. Our study showed that, as well as plasma 5-HT levels, serum 5-HT levels could also be used as a biomarker for antidepressant treatment outcomes. From these results, we hypothesize that both the bioactive 5-HT and 5-HT pool in the periphery might predict the antidepressant treatment outcomes in patients with depressive disorders. To verify this hypothesis further, it would be of interest to measure platelet 5-HT levels in patients who receive antidepressant treatment with depressive disorders.

There are several limitations to this study. First, unlike previous studies, which showed that a greater decrease in plasma 5-HT levels as well as higher baseline plasma 5-HT levels are associated with better clinical outcomes [15,16], we were unable to determine whether treatment-related changes in serum 5-HT levels are associated with treatment outcomes, because we did not obtain serum 5-HT levels during the follow-up period. However, since a greater decrease in plasma 5-HT levels was largely attributed to higher baseline plasma 5-HT levels, rather than to different follow-up period plasma 5-HT levels between responders and non-responders in one previous study [16], we speculate that our study would have shown similar results if the follow-up period serum 5-HT levels had been measured. Second, as this study used a naturalistic prospective design, treatment was in accordance with patient preference under a clinician's guidance rather than being determined by a predetermined protocol. Third, follow-up rates were reduced in the continuation treatment phase compared to the acute treatment phase. Study participants who were lost to follow-up had poor prognostic characteristics at baseline such as unemployed status, melancholic features, and higher EQ-5D scores, which might have obscured the results. Fourth, since various antidepressants were initially used and different treatment strategies (switching, augmentation, combination, and mixtures of these approaches) were applied from 3 weeks after the start of antidepressant monotherapy, there were too many variables to evaluate the effect of 5-HT on antidepressant response by the type of antidepressant.

This study has several strengths. It was a naturalistic prospective study; therefore, results reflect actual clinical practice situations. We followed up patients up to 12

months to evaluate the long-term outcomes of treatment. Moreover, since we assessed various factors associated with treatment outcomes in the baseline, including socio-demographic characteristics, clinical characteristics, and assessment scales, we verified the independent role of baseline serum 5-HT levels as a biomarker for short- and long-term treatment outcomes in depressive disorders, considering multiple confounders.

Although antidepressants are the first-line treatment for moderate to severe depressive disorders, less than one third of patients achieve remission after the first trial. However, proper biomarkers predicting outcomes before starting antidepressant treatments are lacking. This study verified that baseline serum 5-HT levels are higher in the remission group, and high baseline serum 5-HT levels are associated with remission in acute and continuation treatment phases in patients who receive antidepressant monotherapy. By these results, we suggest that baseline serum 5-HT levels might be a biomarker for predicting treatment outcomes in patients who receive antidepressant monotherapy. Further studies should evaluate whether treatment-related changes of serum 5-HT levels are associated with short- and long-term outcomes. As this study was a non-randomized trial, effectiveness of baseline serum 5-HT levels as a biomarker for predicting treatment outcomes in patients who receive antidepressants should be evaluated in randomized trials in the future.

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■ Conflicts of Interest

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