



Large-scale network dynamics in neural response to emotionally negative stimuli linked to serotonin 1A binding in major depressive disorder

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Abstract

Serotonergic dysfunction is implicated in major depressive disorder (MDD), but the mechanisms of this relationship remain elusive. Serotonin 1A (5-HT_{1A}) autoreceptors regulate brain-wide serotonin neuron firing and are positioned to assert large-scale effects on negative emotion. Here we investigated the relationship between raphe 5-HT_{1A} binding and brain-wide network dynamics of negative emotion. 22 healthy-volunteers (HV) and 27 medication-free participants with MDD underwent positron emission tomography (PET) using [¹¹C]CUMI-101 (CUMI) to quantify 5-HT_{1A} binding in midbrain raphe nuclei and functional magnetic resonance imaging (fMRI) scanning during emotionally negative picture viewing. Causal connectivity across regions responsive to negative emotion was estimated in the fMRI data using a multivariate dynamical systems model. During negative picture viewing, MDD subjects demonstrated significant hippocampal inhibition of amygdala, basal-ganglia, thalamus, orbital frontal cortex, inferior frontal gyrus and dorsomedial prefrontal cortex (IFG, dmPFC). MDD-related connectivity was not associated with raphe 5-HT_{1A} binding. However, greater hippocampal inhibition of amygdala, thalamus, IFG and dmPFC correlated with hippocampal 5-HT_{1A} binding. Correlation between hippocampal 5-HT_{1A} binding and the hippocampal inhibition network was specific to MDD but not HV. MDD and HV groups also differed with respect to the correlation between raphe and hippocampal 5-HT_{1A} binding which was more pronounced in HV. These findings suggest that increased hippocampal network inhibition in MDD is linked to hippocampal serotonergic dysfunction which may in turn arise from disrupted linkage in raphe to hippocampus serotonergic circuitry.

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Introduction

Several lines of evidence implicate serotonin 1A (5-HT_{1A}) autoreceptors, in the median and dorsal midbrain raphe nuclei (RN), in the pathophysiology of major depressive disorder (MDD). Serotonergic function across the brain modulates responses to emotionally negative stimuli [1–3]. 5-HT_{1A} autoreceptors regulate serotonin neuron firing and release at terminal fields, moderating brain-wide response to emotionally negative stimuli [1, 2, 4]. Animal research links upregulated 5-HT_{1A} autoreceptors to anxious and depression-like behaviors and their reversal [5–11]. However, studies investigating 5-HT_{1A} autoreceptor binding in MDD diagnosis report mixed results. Using positron emission tomography (PET) some studies report upregulated 5-HT_{1A} receptors in MDD [12–15] and others have not [16–18]. Given the phenotypic heterogeneity of MDD, neurocognitive function rather than syndrome diagnoses,

may link more closely with specific pathophysiological mechanisms such as 5-HT_{1A} autoreceptor binding.

Prior studies investigating the relationship between 5-HT_{1A} autoreceptor binding and neurocognitive features of MDD focused primarily on the amygdala. Some studies found decreased amygdala response to negative stimuli related to greater 5-HT_{1A} autoreceptor binding [19, 20] and another showed non-significant results, when controlling for age/sex [21]. The latter study, conducted in the largest sample, indicates limited support for a simple 5-HT_{1A} autoreceptor binding relationship with amygdala response.

However, the amygdala is only part of the emotional response system [22] and one of many brain regions targeted by serotonergic neurons [23]. Converging evidence indicates that connectivity across large-scale brain networks provides a more informative model of cognitive processing compared with localized activations such as amygdala reactivity [24–31]. Causal connectivity, i.e. the degree that activity in one region predicts in time and intensity the subsequent activity in other regions within the network, particularly conveys useful information about the functioning of a large-scale network [32]. When a network incorporates regions that serve functions as diverse as memory, attention, cognitive control, emotion and perception, the pattern of causal connectivity across the network may be indicative of the type of mental process occurring in response to a given stimulus, e.g. top-down control processes vs. bottom-up perceptually driven processes [33, 34]. For this reason, we examined the relationship between 5-HT_{1A} autoreceptor binding and the pattern of causal connectivity within a large-scale brain network involved in responding to negative emotional stimuli.

To do this, we conducted a multimodal PET and functional magnetic resonance imaging (fMRI) study in a sample of medication-free depressed patients with MDD and healthy volunteers (HV). fMRI was used to identify regions involved in responding to emotionally negative stimuli and to delineate causal relationships between the distributed regions in this network. Using PET, we quantified 5-HT_{1A} autoreceptor binding in midbrain raphe nuclei (RN). Finally, we estimated the relationship between depression related changes in network causality and 5-HT_{1A} binding in MDD vs. HV. We expected to find a relationship between 5-HT_{1A} autoreceptor binding and MDD related network causality in MDD subjects specifically.

Methods

Sample

A total of 77 subjects underwent MRI scanning, the composition of this group was as follows MDD: $N = 36$; HV:

$N = 33$; and high familial risk individuals (HFRI, healthy volunteers despite a first- or second-degree relative with depression) $N = 8$. Out of these subjects, 52 also received a PET scan with [¹¹C]CUMI-101: 27 with MDD, 22 HV and 3 HFRI.

Clinical assessments

Eligibility assessment included medical/psychiatric history, physical examination, routine blood tests, urinalysis, and urine toxicology. Psychiatric diagnoses were established using the Structured Clinical Interview for DSM-IV [35], conducted by doctoral- or masters'-level psychologists trained to a criterion level in diagnostic reliability and accuracy. Depression severity was quantified with the 17-item Hamilton Depression Rating Scale (HDRS-17) [36].

Inclusion criteria for the MDD sample: (1) MDD in a current major depressive episode; (2) age 18–65 years; and (3) off all medications likely to interact with 5-HT_{1A} receptors for ≥ 21 days at the time of scan. This three week drug-free period is based on our findings that antidepressant-associated downregulation of 5-HT_{1A} autoreceptor binding reverses within 2 weeks of medication discontinuation [37]. Medication washout was performed as an inpatient at NYSPI or as an outpatient supervised by a psychiatrist in the MIND clinic. It involved approximately one-week medication taper and three weeks off any medication that affects the serotonergic systems.

Exclusion criteria: (1) psychosis (lifetime); (2) substance or alcohol abuse (2-months), or substance or alcohol dependence (1-year); (3) anorexia nervosa or bulimia nervosa (1-year); (4) intravenous drug use (lifetime); (5) 3,4-methylenedioxy-methamphetamine (>3 times, lifetime); (6) first-degree family of schizophrenia for individuals under age 33; (7) significant active physical illness; (8) electroconvulsive therapy (6-months); (9) previous head trauma with loss of consciousness or cognitive impairment. Inclusion criteria for HVs: (1) absence of major psychiatric illness; (2) age 18–65 years; (3) physically healthy. HFRI did not have a current or past MDD, PTSD, psychosis, bipolar or panic disorders despite having a first or second degree relative with a history of a major depressive episode with onset before age 35 years, who died by suicide or had made a suicide attempt. Exclusion criteria for HVs included: (1) substance/alcohol abuse or dependence (lifetime); (2) IV-drug use (lifetime); (3) MDMA (3 times, lifetime). HFRI were also excluded for current drug or alcohol abuse (within past 2 months), or current or past drug or alcohol dependence in remission for less than 1 year; anorexia nervosa (binge-eating/purging type), bulimia nervosa, or binge-eating disorder in remission for less than one year. Nicotine use was not exclusionary in either group.

Details of the PET and MRI acquisition and preprocessing are included in Supplemental Information, Materials.

fMRI task

Participants underwent three runs of fMRI on a GE 3T scanner while being presented with negative and neutral images from the International Affective Picture System (IAPS [38]). Subjects provided ratings of negative affect following each picture on a 5-point scale (1 “weak” to 5 “strong”). Full details of the fMRI task are included in the supplement. Negative and neutral images were selected on the basis of the normative ratings included within the IAPS. For each trial, a cue was presented instructing participants to look at the upcoming image presented, the presentation of a picture for 8 s, followed by a jittered fixation of 2–4 s, a negative affect rating period, and finally a jittered inter-trial fixation interval of 2–4 s (average = 3 s). Participants completed fifteen trials per run, comprising 5 negative and 5 neutral images, in which the instruction was to look at the images, and 5 negative images, with an instruction to reappraise the images. We investigated emotional reactivity in this paper and therefore “reappraise” trials were modeled as a nuisance regressor and not incorporated into subsequent analyses. Other task related nuisance regressors included, instruction periods, 6-DOF motion, probe periods.

Data analysis

To delineate regions responsive to emotionally negative stimuli, we contrasted negative vs. neutral picture presentation. Within-subject fixed effects models combined data across runs; a group level analysis was used to identify clusters (voxel- $p < 0.001$ and cluster- $p < 0.05$).

The goal of this analysis was to identify brain regions to be used in the connectivity analyses. We therefore grouped all subjects together (i.e., MDD, HVs and HFRI) rather than calculating a separate set of regions for each subgroup, in order to maximize the power of this analysis and its ability to detect all relevant regions. Data for HFRI subjects was not used after this initial analysis as all subsequent analyses were done within or between 2 groups only, to focus specifically on MDD and HV.

Causal connectivity

We next sought to determine the causal interactions between clusters responsive to negative images, i.e. the degree to which each cluster influenced activation of the other regions in the system during negative picture viewing. We employed a multivariate dynamical systems (MDS [24]) model. The MDS model is a type of dynamic causal model that is purely data driven, incorporating minimal priors. This is a state-

space model that consists of a state equation and an observation equation. The state equation models the causal dynamics of the latent quasineuronal activity in the presence of modulatory inputs. The observation equation is a linear convolution model that translates the latent quasineuronal activity into BOLD observations. This provides a measure of the degree to which activity in any given region predicts in both time and intensity the activity of a different region in the model during negative picture viewing. For each node, a measure of the strength of connectivity to and from that node vis-à-vis all other nodes in the network is produced.

In this study, 13 nodes were identified as responsive to negative images (see Results). We extracted run-level average timeseries for neural activity within each of the 13 nodes after first regressing out the 6 DOF motion regressors. MDS was applied to these 13 timeseries generating a matrix of 169 connection strengths per subject. The MDS analysis was performed on BOLD data collected during negative picture presentation and neutral picture presentation, separately. We then identified negativity-related connectivity by subtracting neutral picture connectivity from negative picture connectivity. All statistical analyses were two sided.

Evaluating the MDS

We next sought to evaluate the success of MDS in describing causal neural activity. While it is possible to identify the degree of variance in BOLD signal explained by an MDS model, interpretation of variance explanation is only possible when using simulated data to compare with ground truth or comparing two competing models; neither of which was the case for this study. As a result, we used a permutation analysis to evaluate the MDS model. For each subject, we randomly scrambled the phase of the BOLD time series for all nodes 100 times. Each of these permutations within subject was drawn upon randomly to generate 100,000 permutations of the overall data. We then calculated the difference in observed connectivity between negative and neutral trials and computed the same for the permuted data.

Causal connectivity in MDD vs. HV

To identify network patterns for HV and MDD groups separately, we compared mean observed connectivity for each connection to that observed in the permuted data. To compare connectivity patterns between MDD and HV groups we created a distribution of permuted differences between MDD and HV groups and compared the observed differences between MDD and HV groups to the permuted differences. For all analyses we retained connections showing a difference that met a $p < 0.05$ threshold, when corrected for 169 comparisons, i.e. uncorrected $p < 0.0003$.

MDS was not calculated for the healthy but at-risk subjects because we were specifically interested in comparing MDS results for the MDD and HV group.

Identifying a causal network linked to 5-HT_{1A} autoreceptor binding

We next sought to identify the relationship between MDD related causal connectivity and 5-HT_{1A} autoreceptor binding. To do this, we calculated the sum of connectivity per subject in the connections that were significantly different between MDD and HV. This sum was weighted such that connections that were higher in MDD vs. HV were multiplied by -1 , while connections which were lower or inhibitory in MDD vs. HV were not. This was done so that lower scores would reflect greater inhibition in the connections that were more inhibitory in MDD vs. HV as well as greater excitation along the connections that were more excitatory in MDD vs. HV, that is, greater similarity to MDD.

Results

Sample

Sample demographics are presented in Table 1. Groups did not differ in age, sex, smoking or years of education. Based on the average HDRS-17 score the MDD group was moderately depressed [39]. Group contrasts of regional 5-HT_{1A} binding are the subject of a separate manuscript in preparation and are not presented here. Within the MDD group, 5-HT_{1A} RN autoreceptor binding correlated modestly with depression severity as measured by the HDRS-17 ($r = 0.38$, $p = 0.048$) but not the Beck Depression Inventory ($r = 0.06$, $p = 0.64$). Notably the correlation between autoreceptor binding and HDRS scores does not survive correction for two analyses.

Negative emotional response task affect ratings

Affect ratings collected during the fMRI task indicated greater negative affect in response to negative pictures as compared to neutral pictures across both groups (negative: $M(SD) = 3.56(0.76)$, neutral $M(SD) = 1.59(0.53)$, paired $T_{69} = 20.5$, $p < 0.001$). MDD subjects did not show more negative affect in response to *negative* pictures compared with HVs (MDD: $M(SD) = 3.65(0.66)$, HV $M(SD) = 3.47(0.85)$, $t_{69} = -0.99$, $p = 0.32$). MDD subjects showed more negative affect in response to *neutral* pictures compared with HVs (MDD: $M(SD) = 1.77(0.56)$, HV $M(SD) = 1.39(0.42)$, $t_{69} = -3.12$, $p = 0.002$). Within the MDD group, affect ratings to negative images correlated significantly

Table 1 Demographic and Clinical Characteristics of the Sample.

	MDD ($n = 27$)	HV ($n = 22$)	Group difference ^a
Age	30.9 (8.1)	31.7 (8.5)	$t = 0.33$
Education years	15.23 (2.11)	16.19 (1.56)	$t = 1.73$
Males	14	9	$t = 0.75$
BDI	25.5 (9.5)	1.45 (2.63)	$t = -16.13^{**}$
HDRS-17	18.66 (4.9)	1.09 (0.51)	$t = -11.49^{**}$
Prior depressive episodes			
0 episodes	$N = 7$	N/A	
1 episode	$N = 4$	N/A	
2 episodes	$N = 4$	N/A	
3 episodes	$N = 2$	N/A	
4 episodes	$N = 3$	N/A	
>5 episodes	$N = 7$	N/A	
Length of current episode (days)	Range: 1–624 Median: 52.5		
Current anxiety disorder	$N = 5$	N/A	
Smoking	$N = 1$	0	

BDI Beck depression inventory version I, HDRS-17 Hamilton Depression Rating Scale 17 Item.

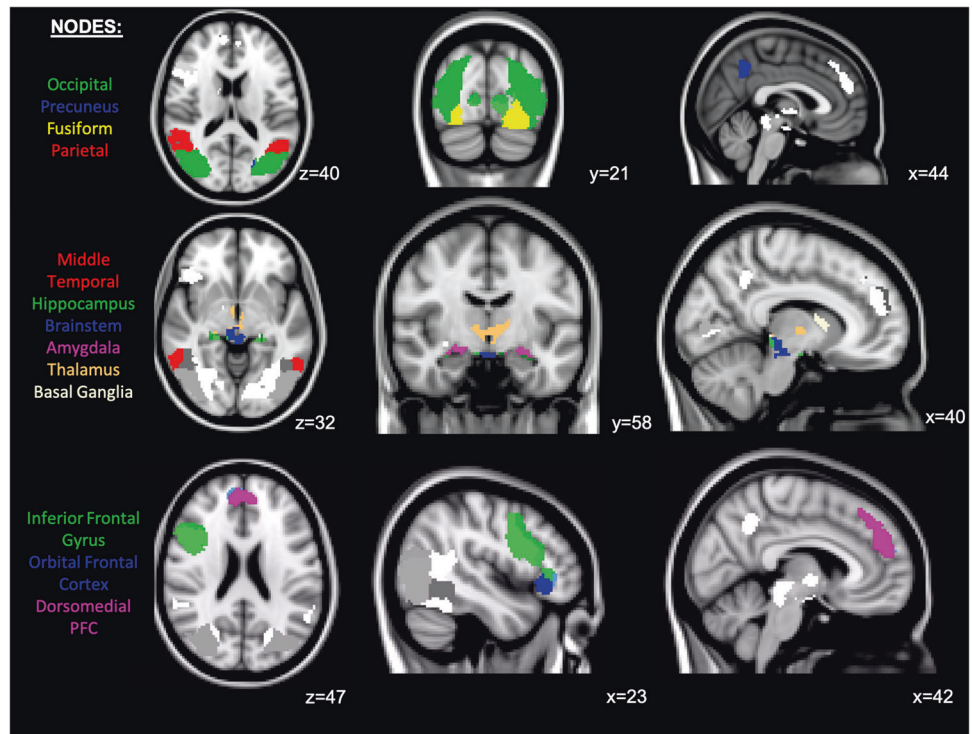
^aAssessed by independent samples t test, $^{**}p < 0.001$.

with depression severity as measured by the self-report BDI but not the clinician-rated HDRS-17 (BDI: $r_{36} = 0.42$, $p = 0.009$, HDRS-17: $r_{36} = 0.166$, $p = 0.34$). Given that the BDI assesses more subjective variables of depression than the HDRS, it is reasonable that affective ratings, which are also subjective, correlated more with the BDI.

Negative images neural response

BOLD activation associated with negative vs. neutral images was widely distributed across the brain (see Fig. 1, Supplementary Table S1). We sought to reduce the broad distribution of activity associated with negative vs. neutral images to a smaller number of nodes for input into the MDS analysis. This was done to simplify the MDS model and also mitigate effects of Bonferroni correction in subsequent analyses. Clusters of significant activity in response to negative vs. neutral images were grouped into 13 nodes based on prior knowledge about the functions of these regions in the context of an emotional perceptual task (Fig. 1). For example, hippocampus and amygdala were grouped separately, while lateral occipital and intracalcarine cortex were grouped together in a broader occipital node. Supplementary Table S1 presents the complete grouping of cluster responses into nodes. Regional delineations followed the Harvard Oxford Cortical and Subcortical Atlases [40].

Fig. 1 Organization of clusters responsive to negative vs. neutral images into 13 nodes. This figure displays all clusters responsive to negative images (voxel- $p < 0.001$, cluster- $p < 0.05$). Color coding is used to indicate the organization of clusters into the 13 nodes that were used as input for the multivariate dynamical systems model. For visual clarity, each row provides color coding for a set of nodes while presenting the other nodes in greyscale. However, all 13 nodes were used in the subsequent analysis.



Causal connectivity in MDD and HV

Inspection of connectivity matrices across the 13-node network in HV and MDD subjects revealed one HV subject to be an outlier who was subsequently removed otherwise variance between groups was similar (Supplementary Fig. S1–S3). Permutation analyses identifying significant connections in the HV group revealed a high degree of excitatory and inhibitory interconnectivity in response to negative pictures across the 13-node network, with the amygdala showing the most excitatory connections and the orbital frontal cortex (OFC) showing the most inhibitory connections (Fig. 2). In MDD subjects, the hippocampus showed the most inhibitory connections and the thalamus showed the most excitatory connections (Fig. 2). Relative to HV, MDD showed greater hippocampal inhibition of dorsomedial prefrontal cortex (dmPFC), inferior frontal gyrus (IFG), amygdala and thalamus, amongst other differences (Fig. 2). Ranges and means for all connection strengths as well as the results of permutation testing are presented in Supplementary Table S2.

Depression related connectivity and 5-HT_{1A} binding

The relationship between raphe 5-HT_{1A} and the pattern of connectivity associated with MDD was not significantly different across MDD and HV groups ($B_{47} = -0.06$, $p = 0.98$, 95%CI: -5.87 to 5.74). Moreover, within each group individually there was no significant relationship between

raphe 5-HT_{1A} and MDD related connectivity (MDD: $r_{27} = 0.11$, $p = 0.61$, HV: $r_{21} = -0.19$, $p = 0.38$).

After finding no effect for raphe 5-HT_{1A} binding on connectivity we conducted a post-hoc analysis modeled on the initially hypothesized autoreceptor analysis. We therefore sought a region with high 5-HT_{1A} binding that influences multiple other emotion-processing regions. In the network analysis, the hippocampus emerged as the region with the most outgoing connections of all nodes in the MDD vs. HV network. Across four connections originating in hippocampus, MDD showed increased inhibition relative to HV and these connections also emerged as significantly inhibitory in the MDD-alone analysis. The hippocampus has the highest 5-HT_{1A} binding outside the raphe nuclei and receives dense afferent fibers from the raphe [41, 42].

Hippocampal inhibition was therefore defined as the sum of causal connections originating in hippocampus that differed significantly between MDD and HV. We calculated the sum of connectivity along these pathways (hippocampus→amygdala, dmPFC, thalamus, IFG) and assessed the interaction effect of diagnosis on the relationship between hippocampal 5-HT_{1A} binding and hippocampal inhibition.

There was a significant interaction of diagnosis on the relationship between hippocampal 5-HT_{1A} and hippocampal inhibition (Fig. 2, Supplementary Table S3). Analysis of main effects revealed a positive correlation between hippocampal 5-HT_{1A} binding and hippocampal inhibition in MDD ($B_{47} = 10.3$, $p = 0.006$, partial $r = 0.41$; 95% CI:

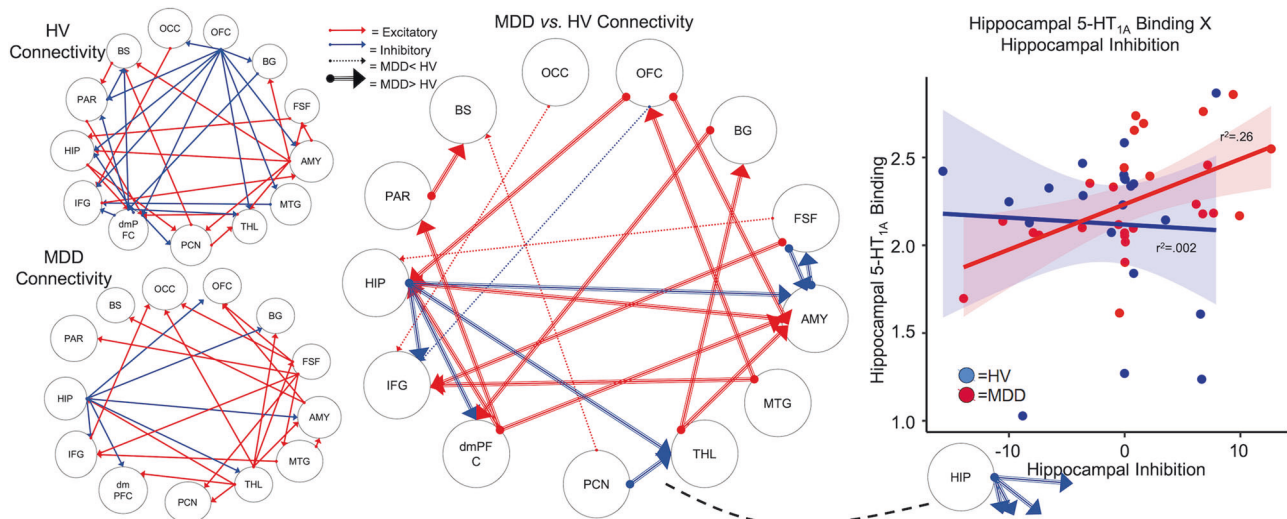


Fig. 2 Causal connectivity in MDD related to hippocampal 5-HT_{1A}. The left most column displays causal connectivity patterns for HV and MDD groups separately. The middle displays connections that differed significantly between depressed and control groups. All analyses employed a threshold $p < 0.05$ Bonferroni corrected for 169 connections (i.e. uncorrected $p = 0.0003$, verified with 100,000 permutations). The right most column shows that hippocampal inhibition

of amygdala, thalamus, dmPFC, and IFG correlates with hippocampal 5-HT_{1A} binding in MDD subjects specifically. Shaded regions show 95% CI. Node HIP hippocampal, PAR parietal, BS brainstem, OCC Occipital cortex, OFC Orbital frontal cortex, BG basal ganglia, FSF fusiform, AMY amygdala, MTG middle temporal gyrus, THL thalamus, PCN precuneus, dmPFC dorsomedial prefrontal cortex, IFG inferior Frontal Gyrus.

3.12–17.49) but not HV ($B_{47} = -0.61$, $p = 0.824$, partial $r = 0.03$; 95% CI: -6.05 to 4.86).

Given the raphe's role in modulating hippocampal serotonergic activity, we computed an interaction examining the relationship between raphe and hippocampal 5-HT_{1A} binding as a function of diagnosis. Raphe and hippocampal 5-HT_{1A} binding were more correlated in the HV compared with MDD group (Fig. 3, Supplementary Table S4). Main effects revealed a positive correlation between raphe and hippocampal 5-HT_{1A} binding in HV ($B_{47} = 1.44$, $p < 0.001$, partial $r = 0.61$; 95% CI: 0.88 to 2.01) but not MDD ($B_{47} = 0.45$, $p = 0.13$, partial $r = 0.22$; 95% CI: -0.14 to 1.04).

Discussion

Individuals with MDD, in comparison to a control group, demonstrated altered patterns of neural inhibition and excitation when viewing emotionally negative pictures. Relative to controls, MDD subjects employed a network of hippocampal inhibition targeting amygdala, thalamus, IFG and dmPFC. Hippocampal inhibition along these pathways correlated with greater hippocampal 5-HT_{1A} binding in MDD specifically. MDD and controls also differed regarding the association between hippocampal and raphe 5-HT_{1A}, which was significant in controls but not in MDD. These findings suggest that increased hippocampal network inhibition in MDD is linked to hippocampal serotonergic dysfunction which may in turn arise from disrupted raphe to hippocampus serotonergic circuitry.

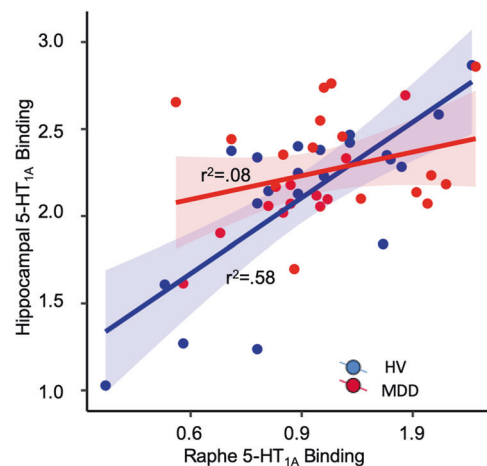


Fig. 3 Hippocampal 5-HT_{1A} in MDD vs. HV. Diagnosis moderated the relationship between raphe and hippocampal 5-HT_{1A} such that they were positively correlated in the HV but not MDD group. Shaded regions show 95% CI.

We initially hypothesized that raphe 5-HT_{1A} binding would correlate with the pattern of emotion processing-neural connectivity linked to MDD. This hypothesis derived from the role of raphe autoregulation in moderating serotonergic activity throughout the brain. However, our data indicate that hippocampal rather than raphe 5-HT_{1A} binding corresponds with broader networked connectivity patterns characteristic of MDD. Even though raphe 5-HT_{1A} binding correlated strongly with hippocampal 5-HT_{1A} in the HV group, it did not correlate in the MDD group. These findings

raise the possibility that alteration of raphe-hippocampus 5-HT_{1A} balance may contribute to the neural response to negative emotion seen in MDD. 5-HT_{1A}-mediated raphe-hippocampal serotonergic circuitry reduces hippocampal theta rhythm, a marker for anxiety [43, 44]. Disconnection of this circuit, as seen in MDD, may deprive the hippocampus of an important modulator of stress responding and contribute to the cascade of downstream hippocampal effects on frontal and limbic targets evidenced here. Future studies with larger sample sizes can test whether the degree of correlation between raphe and hippocampal 5-HT_{1A} moderates the relationship between hippocampal 5-HT_{1A} and downstream hippocampal inhibition.

The network of hippocampal inhibition identified in MDD subjects followed several direct neuroanatomical pathways. Hippocampal projections innervate amygdala, basal ganglia, thalamus and PFC [45, 46]. Non-causal functional connectivity analyses have shown increased inverse correlations between hippocampus and frontal targets in a number of patient groups [47–50]. We add to these findings by showing that the direction of this connectivity in MDD follows an inhibitory path stemming from hippocampus.

Prior directional functional connectivity analyses in healthy subjects demonstrate that prefrontal cortical regions inhibit hippocampal activity during directed suppression of unwanted thoughts [51, 52]. Conversely, in our data MDD subjects showed hippocampal inhibition of frontal and limbic targets. Hippocampal-frontal inhibition may indicate that MDD subjects engaged targeted enhancement of memory encoding and retrieval rather than the memory suppression encoded by frontal-hippocampal inhibition. Supporting this interpretation, the control group in our study showed dmPFC and OFC inhibition of hippocampus.

Hippocampal inhibition of frontal and limbic targets correlated with hippocampal 5-HT_{1A} binding in the MDD group specifically. Animal and human studies show that hippocampal 5-HT_{1A} impairs memory and context encoding [53, 54]. We now provide a potential mechanism for the relationship between hippocampal 5-HT_{1A} binding and impaired memory in the form of hippocampal inhibition. By contributing to hippocampal inhibition, hippocampal 5-HT_{1A} binding may alter the balance of frontal to hippocampal connectivity necessary to encode memories while modulating emotional memory arousal. This lack of modulation may contribute to the lack of specificity in memory that occurs in MDD [55] and generally impaired memory encoding.

Limitations

Causal inference in the relationship between binding and connectivity cannot be made on the basis of this cross-sectional data. The cerebellar gray matter was used as the reference region within the tissue-based model (SRTM) to

quantify [¹¹C]CUMI-101 BP_{ND}, and this may have introduced some underestimation of BP_{ND} due to the presence of some specific binding in the purported reference region. In the absence of blood data, we cannot estimate the tracer volume of distribution in the reference region binding. Systematic differences in reference region volume of distribution between groups could add a potential confound when comparing relationships between connectivity and BP_{ND} across groups. Finally, we note that the hippocampus findings are derived from post-hoc and not hypothesis driven analyses.

Conclusions

This study identified patterns of causal network dynamics across brain regions responsive to emotionally negative stimuli in MDD subjects and healthy volunteers. MDD subjects showed increased hippocampal inhibition of IFG, dmPFC, amygdala and thalamus. This inhibition correlated with increased 5-HT_{1A} binding in hippocampus in MDD but not control subjects suggesting that hippocampal serotonergic dysfunction in MDD may contribute to increased hippocampal inhibition. One potential explanation of serotonergic dysfunction in MDD may be the lack of association between raphe and hippocampal 5-HT_{1A} binding observed specifically in MDD subjects. Taken together, these findings suggest that increased hippocampal network inhibition in MDD is linked to hippocampal serotonergic dysfunction which may in turn arise from disrupted linkage in raphe to hippocampus serotonergic circuitry.

Compliance with ethical standards

Conflict of interest JJM receives royalties for commercial use of the C-SSRS from the Research Foundation for Mental Hygiene and has stock options in Qualitas Health, a start-up developing a PUFA supplement. PS is the majority owner and Chairman of the Board of Neuromatters LLC, a brain computer interface neuromarketing and gaming company. MAO receives royalties for the commercial use of the Columbia-Suicide Severity Rating Scale. Her family owns stock in Bristol Myers Squibb. The remaining authors have no conflicts of interests to declare.

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