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Elevated striatal γ -aminobutyric acid in youth with major depressive disorder

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Abstract

Background: Alterations in γ -aminobutyric acid (GABA) have been hypothesized to play a role in the pathogenesis of psychiatric illness. Our previous work has specifically linked anterior cingulate cortex (ACC) GABA deficits with anhedonia in youth with major depressive disorder (MDD). As anhedonia reflects alterations within the reward circuitry, we sought to extend this investigation and examine GABA levels in another key reward-related region, the striatum, in the same adolescent population.

Methods: Thirty-six youth [20 with MDD and 16 healthy controls; (HC)], ages 12 to 21 years old, underwent J-edited proton magnetic resonance spectroscopy (¹H MRS) whereby GABA levels were measured in striatal and ACC voxels. GABA levels were compared between groups and

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Author contributions

KAB performed statistical analyses, interpreted results, and prepared the manuscript. CMA conducted clinical evaluations of participants. LMM assisted with statistical analyses and manuscript preparation. JX assisted with the methods and manuscript preparation. VG designed and executed the study, interpreted results, and assisted with manuscript preparation. All authors have approved the final article.

Conflict of interest

All authors report no conflict of interest.

Declarations of interest

None.

Ethical statement

This work was carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki). Participants who were 18 years of age or older provided informed consent prior to beginning study procedures, while those under 18 provided assent, with a parent or guardian providing informed consent. Institutional review boards at all participating institutions approved the study protocol.

between voxel positions and were examined in relation to clinical symptomatology, such as depression severity, anhedonia, anxiety, and suicidality.

Results: Depressed youth had unexpectedly higher GABA levels in the striatum compared to HC. In both depressed and healthy youth, GABA levels were higher in the striatum than in the ACC, while the differences in depressed youth were greater. Moreover, in depressed youth, higher striatal GABA above the mean of HCs was correlated with lower ACC GABA below the mean of HCs. Striatal GABA was not correlated with clinical symptomatology in this small sample.

Conclusions: Together, these findings suggest that higher striatal GABA levels may serve some compensatory function as a result of lower ACC GABA in depressed adolescents. It is also possible that, like lower ACC GABA, higher striatal GABA might simply be another pathological feature of adolescent depression.

Keywords

Depression; Adolescence; J-edited magnetic resonance spectroscopy; GABA; Striatum

1. Introduction

Dysfunction in γ -aminobutyric acid (GABA), the main inhibitory neurotransmitter in the brain, has been increasingly reported across psychiatric disorders (Bustillo, 2013; Chiapponi et al., 2016; Iorfino et al., 2016; Schur et al., 2016). Converging evidence in adult (Abdallah et al., 2015; Hasler et al., 2007; Price et al., 2009; Sanacora et al., 2004, 1999) and adolescent (Gabbay et al., 2017, 2012) clinical populations has implicated alterations in GABA in the development of major depressive disorder (MDD). Our work has specifically focused on adolescents with MDD in order to assess neurobiological contributors early on in illness during a critical developmental period when many psychiatric conditions first occur (Kessler et al., 2005).

In a recent publication (Gabbay et al., 2017), we confirmed our previous finding of lower anterior cingulate cortex (ACC) GABA levels in psychotropic medication-free youth with MDD compared to healthy controls [HC; (Gabbay et al., 2012)]. Importantly, GABA levels were only lower in depressed youth categorized as anhedonic, which reflects deficits in reward processes. Furthermore, across all depressed participants, anhedonia severity was negatively correlated with ACC GABA levels, thus implicating lower ACC GABA in the phenomenology of anhedonia in depressed youth.

Multiple reviews have implicated dysfunction within the broader reward network in the phenomenology of anhedonia in support of our laboratory's work (Heshmati and Russo, 2015; Hulvershorn et al., 2011). In our previous investigations of adolescents with MDD, we focused specifically on the ACC due to this prefrontal region's role in reward processing (Der-Avakian and Markou, 2012; Haber, 2011; Heshmati and Russo, 2015). However, the reward network is generally acknowledged as being comprised of cortico-striatal regions, including the orbitofrontal cortex, ACC, striatum (caudate, putamen), ventral striatum (nucleus accumbens), ventral pallidum, and midbrain dopamine neurons (Haber, 2011). As anhedonia reflects alterations within this reward network (Der-Avakian and Markou, 2012;

Heshmati and Russo, 2015), we chose to examine GABA in regions primary to reward function. As an extension of our previous investigations in depressed youth (Gabbay et al., 2012, 2017), here we aimed to further investigate GABA alterations in the striatum, another core region within the reward network.

To date, all studies of GABA levels in mood disorders have focused on cortical regions, such as the prefrontal and occipital cortex (Schur et al., 2016). There have been no investigations of striatal GABA in adolescents with MDD, likely due to the challenge of achieving high quality proton magnetic resonance spectroscopy (^1H MRS) data from this subcortical region (Quetscher et al., 2015). The striatum has been shown to be inextricably linked to reward function and is known to play a role in the pathophysiology of mood disorders (Marchand and Yurgelun-Todd, 2010). Additionally, psychiatric studies have shown opposing findings of neurochemical levels between subcortical and cortical regions. For example, studies of dopamine levels in schizophrenia have indicated increased levels of dopamine in the striatum but decreased levels in the prefrontal cortex (Brisch et al., 2014), further highlighting the importance of examining neurochemical levels across entire functional networks. Therefore, given the paucity of research investigating GABA levels in subcortical reward-related regions in relation to clinical symptomatology such as anhedonia in adolescent depression, we sought to address this gap. Based on our prior findings of lower ACC GABA levels in depressed youth (Gabbay et al., 2012, 2017), we hypothesized that GABA levels would be similarly lower in the striatum of depressed youth, with GABA levels inversely correlated with anhedonia severity. Additionally, since our sample overlapped with those from our previous investigations of ACC GABA, we also explored regional differences in GABA levels between the ACC and striatum in the depressed and HC participants—a topic which has not been previously examined in this clinical population.

2. Materials and methods

2.1. Participants and procedure

Thirty-six participants completed the study, 20 with MDD, and 16 healthy controls (HC). Participants underwent a clinical evaluation using the Kiddie Schedule for Affective Disorders-Present and Lifetime Version [KSADS-PL; (Kaufman et al., 1997)], along with a comprehensive battery of clinical assessments and a magnetic resonance imaging (MRI) scan, including structural MRI and striatal GABA MRS. All 36 participants also completed an ACC GABA MRS scan, of which the data have been previously published (Gabbay et al., 2017). Participants who were 18 years of age or older provided informed consent prior to beginning study procedures, while those under 18 provided assent, with a parent or guardian providing informed consent. Institutional review boards at all participating institutions approved the study protocol.

2.2. Inclusion and exclusion criteria

All participants were youth between the ages of 12 and 21 years old. Participants were required to have an intelligence quotient (IQ) of at least 80 on the Kaufman Brief Intelligence Test (Kaufman and Kaufman, 1990) and could not have a significant medical, neurological, or neurodevelopmental disorder. All participants were given a standard MRI

safety-screening questionnaire, and participants with MRI contraindications were excluded. Furthermore, pregnancy in females and current drug use, as assessed by a urine toxicology test, in any participant on the day of the scan, were also exclusionary.

For inclusion in the MDD group, participants were required to meet *Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition-Text Revision* [DSM-IV-TR; (APA, 2000)] criteria for MDD, with a current episode duration of at least 8 weeks, and a depression severity score of at least 35 on the Children's Depression Rating Scale-Revised [CDRS-R; (Poznanski and Mokros, 1996)]. Due to the inherent heterogeneity of MDD in youth, comorbid diagnoses of attention-deficit/hyperactivity disorder (ADHD) and anxiety disorders were not exclusionary, so as to be representative of the disorder as it generally presents in an adolescent sample. However, diagnoses of psychosis, bipolar disorder, pervasive development disorder, eating disorders, a current substance abuse disorder, or active suicidality requiring immediate medical attention were exclusionary. Additionally, participants with MDD were required to be either medication-naïve or medication-free for at least seven half-lives of the medication. The HC group could not meet criteria for any current or past DSM-IV-TR disorder and had to be psychotropic medication-naïve.

2.3. Clinical assessment instruments

Participants were all evaluated using the KSADS-PL, a clinical assessment validated for use in adolescents up to age 17 (Kaufman et al., 1997). The KSADS-PL was used for all youth in the study, even though the sample extended up to 21 years old, to maintain continuity of assessment and in light of the rather vague and debated definition of the age range of “adolescence” in psychiatric research. A board-certified child/adolescent psychiatrist or licensed clinical psychologist trained in administering the KSADS-PL carried out the diagnostic evaluations. The final clinical report was discussed between the Primary Investigator (a licensed child/adolescent psychiatrist with expertise in adolescent depression) and the assessor to ensure consistency and diagnostic accuracy. A battery of clinical assessments was also given, including the Children's Depression Rating Scale-Revised [CDRS-R; (Poznanski et al., 1984, 1985; Poznanski and Mokros, 1996)], the Beck Depression Inventory [BDI; (Beck et al., 1961, 1988, 1997)], the Multidimensional Anxiety Scale for Children [MASC; (March et al., 1997)], and the Beck Scale for Suicidal Ideation [BSSI; (Beck et al., 1979)].

Anhedonia severity was quantified by combining the anhedonia-specific items from the clinician-rated CDRS-R and the self-rated BDI. Specifically, the “difficulty having fun” item on the CDRS-R, and the “loss of interest” and “loss of pleasure” items on the BDI were summed after converting items to be rated on the same scale, as has been done in our previous investigations (Gabbay et al., 2012, 2017).

2.4. Neuroimaging data acquisition and analysis

2.4.1. Structural MRI—Imaging data was acquired using a 3.0 Tesla MRI system (Excite HD, General Electric) with an 8-channel phased-array head coil. Standard high-resolution axial, coronal, and sagittal T1-, T2-, and spin density-weighted scans were collected in order to prescribe the ¹H MRS voxels of interest (VOI) in the left striatum and

ACC. T1-weighted spoiled gradient-recalled echo (SPGR) volumetric images with the following parameters were also collected for brain tissue segmentation (repetition time [TR] = 30 ms; echo time = 8 ms; flip angle = 45 degrees; field-of-view [FOV] = 24 cm; matrix = 256×256 ; thickness = 1.5 mm; 124 coronal slices). Lastly, an axial fast fluid-attenuated inversion recovery (FLAIR) scan was acquired to rule out brain lesions, which were exclusionary. Full imaging parameters and procedures have been described in detail previously (Gabbay et al., 2012, 2017).

2.4.2. ^1H MRS—As with our prior studies, a standard J-edited MEGA-PRESS sequence (Rothman et al., 1993) was used to acquire the GABA-edited ^1H MRS data, with the same acquisition parameters and processing steps as previously implemented (Freed et al., 2016; Gabbay et al., 2012, 2017; Shungu et al., 2016). Spectra were recorded from a $1.5 \times 2.0 \times 3.0 \text{ cm}^3$ left striatal VOI and a $2.5 \times 2.5 \times 3.0 \text{ cm}^3$ VOI in the ACC. MRS data quality assessment procedures are provided in (Shungu et al., 2016). Metabolite peak areas proportional to the concentrations of the associated metabolites were obtained for cases that met quality assessment criteria. The GABA resonance and the co-edited Glx (i.e., glutamate and glutamine) resonance in the J-edited difference spectra were modeled as a linear combination of pseudo-Voigt line-shape functions and fitted in the frequency domain using a public-domain Levenberg–Marquardt nonlinear least-squares minimization routine (Markwardt et al., 2009). The final GABA and Glx levels were expressed as ratios (GABA/W or Glx/W) of the peak neurometabolite area relative to the area of the unsuppressed voxel tissue water signal (W).

2.4.3. Voxel tissue heterogeneity variables—Due to potential differences in the GABA and Glx signals between brain tissue types (e.g., gray matter and white matter), volumetric structural MRI-based tissue segmentation was conducted using MEDx software (Medical Numerics, Germantown, MD), and relationships between tissue variables and neurometabolites were explored. From each participant's T1-weighted SPGR volumetric scan, brain tissue was segmented based on the signal-intensity histogram and estimates of the proportions of gray matter (GM), white matter (WM), and cerebrospinal fluid (CSF) in each striatal voxel of interest were made; a segmentation mask of each striatal voxel was created from the histogram using in-house software developed in MATLAB (MathWorks, Natick, MA). Voxel tissue heterogeneity was then compared between groups, and tissue variables showing significant differences between groups or specific correlations with neurometabolites were included as covariates in the appropriate statistical models.

2.5. Statistical analyses

Analyses were conducted using SPSS Statistics, version 23 (IBM Corp., Armonk, NY). All variables were examined using descriptive statistics. Normality was assessed via the Shapiro-Wilk test. Groups (i.e., HC and MDD) were compared on normally distributed demographic and clinical variables using independent samples *t*-tests. When data were not normally distributed, nonparametric alternatives were utilized such as the Mann-Whitney *U* test. Group differences in voxel tissue heterogeneity variables (i.e., W, GM, WM, and CSF) were assessed using analysis of covariance (ANCOVA), controlling for age and sex, as these factors have been shown to affect brain tissue makeup (De Bellis et al., 2001).

Striatal GABA and Glx levels were compared between MDD and HC groups using independent samples *t*-tests or ANCOVA models. For group comparison models, relationships between GABA and Glx and demographic and voxel tissue variables were examined. Only variables that were significantly correlated ($p < .05$) with or showed differences with GABA or Glx were included as covariates in ANCOVA models that compared neurometabolites between groups. Additionally, due to potential neuroendocrine variation in the sample, all group comparisons of GABA and Glx explored Tanner stage as a covariate. Finally, as a post-hoc analysis, a mixed model ANCOVA was used to compare striatal GABA and ACC GABA levels in youth with MDD and HC, controlling for GM%, given the inherent differences in tissue makeup of the ACC and striatum.

Relationships between clinical variables (i.e., depression severity, anhedonia, anxiety, and suicidality) and striatal GABA and Glx in the group with MDD were examined using correlations; Pearson's ("r") correlations (two-tailed) were used when data was normally distributed, whereas Spearman's rank-order ("rho" or "p") correlations (two-tailed) were used when data was not normally distributed. Partial Pearson's or Spearman's rank-order correlations were also conducted between anhedonia and GABA, controlling for depression severity, and between depression severity and GABA, controlling for anhedonia.

For all analyses, significance was defined as $p < .05$.

3. Results

3.1. Demographic and clinical characteristics

There were no significant differences between the MDD and HC groups in age [$t(34) = 0.08$, $p = .94$], sex [$\chi^2(1) = 0.36$, $p = .55$], or ethnicity [$\chi^2(3) = 4.13$, $p = .25$]. Additionally, there was no correlation between age and striatal GABA [$\rho = 0.13$, $p = .45$], or differences in striatal GABA between the sexes [$t(34) = 0.67$, $p = .51$] or ethnicities [$F(3,32) = 0.50$, $p = .68$]. Therefore, none of these demographic variables were included in the primary statistical models comparing neurochemical levels between groups. All participants were in an advanced puberty stage (stage 4 or 5) as determined by the Tanner scale, except for one HC male, who was a 3; Tanner staging was not completed on 2 participants, ages 16 and 21 years old.

Within the MDD group, 15 participants were psychotropic medication naïve and 5 were medication free. Demographic and clinical characteristics are presented in Table 1.

3.2. Striatal voxel tissue heterogeneity

There were no differences between the MDD and HC groups in unsuppressed voxel tissue water (W) [$F(1,32) = 3.84$, $p = .06$]; thus, the ratios of GABA/W and Glx/W were used in all analyses and are simply referred to as GABA or Glx throughout the manuscript. Additionally, there were no group differences in GM% [$F(1,32) = 0.49$, $p = .49$], WM% [$F(1,32) = 0.26$, $p = .61$], or CSF% [$F(1,32) = 2.62$, $p = .12$] in the striatal VOI. Moreover, there were no correlations between striatal GABA and GM% [$r = -0.28$, $p = .10$], WM% [$r = 0.27$, $p = .11$], or CSF% [$\rho = -0.01$, $p = .95$]. Therefore, none of these variables were

included as covariates in the statistical models comparing MDD and HC groups. Voxel tissue variables are presented in Table 2.

3.3. Group (MDD, HC) comparison of striatal GABA

Striatal GABA was significantly higher in youth with MDD (mean = 4.61×10^{-3} , SD = 0.66×10^{-3}) compared to HC [mean = 3.88×10^{-3} , SD = 0.45×10^{-3} ; $t(34) = -3.81$, $p = .001$; Table 3; Fig. 1]. Despite the lack of correlation between age and striatal GABA (see Section 3.1 above), an additional ANCOVA, including age as a covariate, was explored to compare striatal GABA levels between groups due to the wide age range of the sample. When controlling for age, results were unchanged, with striatal GABA significantly higher in the MDD group [$F(1,33) = 15.03$, $p < .0005$, $\eta_p^2 = 0.31$]. Lastly, due to potential neuroendocrine variations, Tanner stage was also explored as a covariate. Results remained unchanged when controlling for both age and Tanner stage, [$F(1,30) = 12.52$, $p = .001$, $\eta_p^2 = 0.29$].

3.4. Striatal GABA and clinical symptom correlations in MDD

Within the MDD group ($n = 20$), striatal GABA was not correlated with anhedonia [$r = -0.03$, $p = .90$], even when controlling for depression severity [CDRS-R minus the anhedonia item; $r = 0.11$, $p = .65$]. Striatal GABA was also not correlated with suicidality [BSSI; $r = -0.38$, $p = .10$] or anxiety (MASC) [$r = -0.44$, $p = .06$]. Furthermore, striatal GABA was not correlated with depression severity (CDRS-R minus the anhedonia item), both when controlling for [$r = -0.38$, $p = .11$] and not controlling for anhedonia [$r = -0.36$, $p = .12$].

3.5. Striatal and ACC GABA comparisons between groups (MDD, HC)

A mixed model ANCOVA compared GABA levels across regions (striatum, ACC) and groups (MDD, HC); the difference in GM% between the ACC and striatum was included as a covariate due to the different tissue makeups of each of these regions. The interaction between group (MDD, HC) and region (striatum, ACC) was significant [$F(1,33) = 15.17$, $p < .0005$]. Follow-up simple effects showed that striatal GABA was higher than ACC GABA in both the MDD group ($p < .0005$) and HC ($p < .0005$) (Table 3; Fig. 1). The main effect of region (striatum, ACC) [$F(1,33) = 17.67$, $p < .0005$] was also significant, such that GABA was higher in the striatum than in the ACC. The main effect of group (MDD, HC) was not significant [$F(1,33) = 3.70$, $p = .06$].

3.6. Post-hoc investigation of regional GABA differences in MDD

In order to further examine the relationship between striatal and ACC GABA in youth with MDD, we calculated the Pearson correlation coefficient between GABA levels in these regions. There was a significant negative correlation between striatal GABA and ACC GABA in MDD [$r = -0.46$, $p = .04$]. We additionally calculated the difference between individual MDD striatal GABA levels and the mean HC striatal GABA level, as well as the difference between the mean HC ACC GABA level and individual MDD ACC GABA levels. There was a significant positive correlation between these deviations from the average GABA levels in HC [$r = 0.46$, $p = .04$; Fig. 2]. Together, these results show that individuals with higher striatal GABA above the mean of the HC also had lower ACC

GABA below the mean of the HC. Lastly, ACC GABA was not significantly correlated with anhedonia severity, neither when controlling [$r = 0.04$, $p = .86$] nor not controlling [$r = 0.17$, $p = .46$] for depression severity (CDRS-R minus the anhedonia item).

3.7. Striatal Glx group comparisons and correlations with clinical variables

Striatal Glx levels were not significantly different between groups, either when controlling for age [$F(1,33) = 3.00$, $p = .09$, $\eta_p^2 = 0.08$] or when not [MDD mean = 2.61×10^{-3} , SD = 5.09×10^{-4} ; HC mean = 2.31×10^{-3} , SD = 5.20×10^{-4} ; $t(34) = -1.74$, $p = .09$]. Additionally, the results remained unchanged when including Tanner stage and age as covariates, [$F(1,30) = 2.25$, $p = .14$, $\eta_p^2 = 0.07$].

There were no significant correlations between Glx and anhedonia, either when controlling for [$r = -0.06$, $p = .81$] or not controlling for depression severity [CDRS-R minus the anhedonia item; $r = 0.07$, $p = .77$]. Glx was also not correlated with depression severity, either when controlling for [$\rho = 0.10$, $p = .67$] or not controlling for anhedonia [$\rho = 0.08$, $p = .73$]. Lastly, Glx was not correlated with anxiety [MASC; $r = -0.21$, $p = .40$] or suicidality [BSSI; $\rho = 0.34$, $p = .15$].

4. Discussion

The aims of the current study were to: 1) examine striatal GABA levels in youth with MDD compared to HC; 2) assess relationships between striatal GABA and clinical symptoms of MDD; and 3) explore regional differences (striatum, ACC) in GABA levels. Contrary to our original hypothesis following our initial observation in the ACC, depressed youth exhibited significantly higher striatal GABA levels than HC. Striatal GABA was not correlated with anhedonia, whether controlling or not controlling for depression severity. No other clinical variables (e.g., anxiety, suicidality) were associated with striatal GABA levels either. Additionally, there were no group differences in striatal Glx or any associations between striatal Glx levels and clinical symptomatology. We also found significant regional differences in GABA levels between the striatum and ACC. Both depressed youth and HC showed higher GABA in the striatum compared to the ACC, but the larger differences were in the depressed group. Moreover, higher striatal GABA above the HC mean were significantly correlated with lower ACC GABA below the HC mean in depressed youth. Furthermore, anhedonia was not correlated with ACC GABA in this small sample. Collectively, these results suggest that higher striatal GABA levels are associated with lower ACC GABA levels in adolescent depression. While neither was independently associated with anhedonia severity in this small sample, we speculate that these regional differences reflect some compensatory function against developing anhedonia in depressed youth, based on a clear negative relationship between anhedonia severity and ACC GABA levels in a much larger sample composed of individuals with more severe anhedonia from our prior work (Gabbay et al., 2017). However, it is also possible that, like lower ACC GABA, higher striatal GABA might simply be a pathological feature of adolescent depression. These explanations are discussed below.

4.1. Elevated striatal GABA in depressed youth

Contrary to our original hypothesis that youth with MDD would exhibit lower striatal GABA than HC, we found higher GABA in this region. Conversely, our previous investigations in depressed adolescents demonstrated lower GABA levels in the ACC, with reductions in this region most prominent in the anhedonic subgroup (Gabbay et al., 2017; Gabbay et al., 2012). To the best of our knowledge, only one other study has investigated brain GABA levels in an adolescent sample and found that individuals at high familial risk for depression showed no differences in parieto-occipital GABA levels compared to HC (Taylor et al., 2011). Within the adult literature, findings are mostly consistent, showing lower GABA in both the plasma (Lu et al., 2014; Petty et al., 1990, 1992) and the CSF (Gerner and Hare, 1981; Kasa et al., 1982) of depressed individuals. ¹H MRS investigations in adults similarly show a general trend of lower cortical GABA levels in actively depressed adults (Hasler et al., 2007; Sanacora et al., 1999, 2004). A recent meta-analysis confirmed this trend and found that brain GABA levels were lower in actively depressed, but not remitted depressed adults, compared to HC (Schur et al., 2016); these differences were largely in cortical regions such as the occipital cortex (8 studies), ACC (4 studies), and prefrontal cortex (5 studies). Only one study included in the meta-analysis examined GABA levels in a subcortical voxel that encompassed portions of the striatum, subgenual cingulate, hippocampus, amygdala, and thalamus (Shaw et al., 2013). In this study, there were no differences in either subcortical or cortical GABA levels between remitted depressed adults or HC (Shaw et al., 2013). The differences between the current findings and both the existing pediatric and adult literature may be the result of the specific brain regions examined; most previous investigations have focused on cortical regions, whereas our current study examined the striatum, a subcortical region. In addition, unlike Shaw et al. (2013), our study assessed actively depressed youth and not remitted adults. Most ¹H MRS investigations of GABA levels in both pediatric and adult samples have been in frontal and occipital cortical regions, no doubt due to the convenience of attaining high quality spectra in these regions. However, our finding of higher subcortical GABA levels is unexpected and exemplifies the importance of examining neurochemical levels across entire functional networks in order to better elucidate the biological mechanisms underlying psychopathology.

4.2. GABA and anhedonia severity

Given that anhedonia reflects reward circuitry deficits (Heshmati and Russo, 2015) and the striatum is integral to reward processing (Delgado, 2007), we speculate that higher GABA levels in this region may be related to anhedonia. While our small sample of depressed adolescents in this current investigation did not demonstrate a significant correlation between striatal GABA levels and anhedonia severity, this lack of a relationship does not necessarily imply that subcortical GABA levels are not related to reward circuitry mediated psychopathology. Our prior two reports on ACC GABA levels in depressed youth demonstrated negative correlations between ACC GABA and anhedonia severity (Gabbay et al., 2012, 2017) in the MDD group. However, it is important to note that our prior samples in which we detected a correlation between ACC GABA and anhedonia included patients with melancholic depression, which is characterized by severe anhedonia; these melancholic depressed adolescents had the lowest ACC GABA levels (Gabbay et al., 2012, 2017). We

did not document a correlation between ACC GABA and anhedonia in a smaller sample without these highly anhedonic participants (Gabbay et al., 2017). This decrease of cortical GABA specifically in melancholic depression has also been documented in adults (Sanacora et al., 2004), suggesting that this relationship between GABA and anhedonia may be most apparent in samples of highly anhedonic individuals, which we did not have here. Due to fewer participants with high anhedonia and lower power resulting from the smaller sample size of depressed youth ($n = 20$) compared to our prior report ($n = 44$; Gabbay et al., 2017), these relationships between cortical and subcortical GABA levels and anhedonia may not be apparent in the current investigation.

Research has consistently demonstrated interconnected relationships between GABA levels and dopamine release in reward-related subcortical and cortical brain regions (Brambilla et al., 2003; Jones et al., 1988; Miller et al., 2013; Pycock and Horton, 1979; Reid et al., 1988). The reward circuitry is generally regarded as being comprised of the prefrontal cortex—specifically the orbitofrontal cortex (OFC), ACC, dorsomedial and dorsolateral prefrontal cortex (PFC)—ventral striatum (nucleus accumbens; NAc), dorsal striatum (caudate and putamen), ventral tegmental area (VTA), habenula, amygdala, and hippocampus (Der-Avakian and Markou, 2012; Heshmati and Russo, 2015). Most commonly, animal studies have shown that GABA activation suppresses dopamine release in both subcortical and cortical reward regions (Brambilla et al., 2003; Jones et al., 1988; Pycock and Horton, 1979; Reid et al., 1988). One study specifically examined rat reward-seeking behavior in relation to direct VTA GABA neuron activation and found that activation of VTA GABA neurons inhibited reward consumption but not anticipation, presumably due to decreased dopamine release (van Zessen et al., 2012). In line with this work, one explanation, though controversial, of our current findings is that increased striatal GABA, and possibly the associated striatal glutamate level change, suppresses dopamine release (Avshalumov et al., 2003; Sulzer et al., 2016), inhibiting the function of the striatum and leading to impaired reward function and anhedonic symptomatology in depressed adolescents. In the current study, youth with the highest striatal GABA also had the lowest ACC GABA, and low cortical GABA has been previously associated with anhedonia (Gabbay et al., 2012, 2017; Sanacora et al., 2004). Given this mechanism, higher striatal GABA, like lower ACC GABA, could be a pathological feature of adolescent depression related to impaired reward function.

Alternately, the VTA also has prominent connections to the dorsal striatum and prefrontal cortex, and activation of GABA neurons in these pathways may differentially affect corresponding dopamine levels in these areas (van Zessen et al., 2012). For example, GABA expression has also been found to increase dopamine release (Brambilla et al., 2003; Garbutt and van Kammen, 1983) in the rat striatum and PFC (Bonanno and Raiteri, 1987). Due to the strong role of the striatum in reward processing (Delgado, 2007), it is possible that a higher GABA level in the striatum may increase dopamine release in this important reward-related region in order to serve some neuroprotective or compensatory function in response to lower ACC GABA, which, again, has been associated with anhedonia (Gabbay et al., 2012, 2017). Importantly, our GABA measurements reflect both the intracellular and extracellular GABA levels combined, which does not allow for the assessment of specific synaptic GABA levels or its receptor function. Thus, we cannot truly delineate the specifics

of how striatal and ACC GABA are related to each other and anhedonia in this population. Therefore, the exact mechanisms through which striatal and ACC GABA may be related to reward function in adolescent depression require further investigation.

4.3. GABA differences across regions

Lastly, as our entire sample also underwent a separate ACC GABA scan, from which data were previously published (Gabbay et al., 2012, 2017), we also aimed to investigate regional differences in GABA levels in youth. To the best of our knowledge, there have been no systematic investigations comparing cortical and subcortical GABA levels using ^1H MRS in young psychiatric samples. We documented a significant group by region interaction such that depressed youth had higher GABA levels in the striatum compared to the ACC. While higher striatal GABA levels were also significant in the HC group, this regional difference in GABA was much larger in the depressed subgroup. Given the inherent difference in GM tissue makeup between the ACC and striatum, regional GM difference was controlled for in these analyses. Overall, the striatum (average GM = 71.50%) had a larger GM% than the ACC (average GM = 58.06%) in our sample. In general, it has been documented that GM tissues contain much higher concentrations of GABA than WM (Choi et al., 2006; Jensen et al., 2005). Therefore, our finding of higher GABA in the striatum compared to the ACC in both depressed youth and HC is not surprising, given that this region has a higher GM %.

Importantly, since our previous work showed lower ACC GABA levels in an overlapping depressed cohort compared to HC (Gabbay et al., 2012, 2017), we also investigated whether lower ACC GABA was correlated with higher striatal GABA in the MDD group. We found that striatal and ACC GABA levels were indeed negatively correlated in MDD. Moreover, in the depressed group, higher striatal GABA above the mean of the HCs was also associated with lower ACC GABA below the mean of the HCs. Together, these results support both the theory that increased striatal GABA and lower ACC GABA are dual pathological features of adolescent depression, as well as our speculation that higher subcortical GABA levels in MDD may arise as a way to compensate for lower levels of ACC GABA and subsequent anhedonic symptomatology in depressed youth.

4.4. Limitations

While this is one of the first studies to investigate striatal GABA levels in psychotropic medication-free youth, there are a number of limitations that may affect the results. First, the low concentration of brain GABA requires sampling a relatively large voxel, and while our voxel size was within the standard scope of current ^1H MRS methods at 3 Tesla, partial volume effects from white matter and CSF within the subcortical voxel of interest could confound interpretation. Additionally, despite the limited evidence in literature to support that the concentration of mobile macromolecules in the brain differ in MDD from HC, the GABA peak could have contained up to 50% contribution from mobile macromolecules that co-edit with GABA. Furthermore, while measures of inter-rater reliability could not be conducted for clinical evaluations, such as the KSADS-PL or the CDRS-R, due to the study procedures whereby a single clinician performed the evaluations, these assessments were done by a board-certified child/adolescent psychiatrist or licensed clinical psychologist trained in administering these assessments under our laboratory's standard protocol, and the

clinical reports were reviewed by a licensed child and adolescent psychiatrist specializing in pediatric depression. Lastly, the small sample size is not ideal to make findings generalizable to the broader population of depressed youth and may have limited our ability to investigate relationships between GABA levels and clinical symptomatology. Thus, additional studies conducted in larger samples of primarily anhedonic adolescents are also needed to fully understand the complex relationships between GABA in the striatum and ACC in depressed youth.

5. Conclusion

This study is the first investigation of subcortical GABA levels in a sample of depressed, psychotropic medication-free youth. We unexpectedly found higher striatal GABA levels in depressed youth, with no specific correlations between subcortical GABA levels and clinical symptomatology. Additionally, we found regional GABA differences such that higher striatal GABA was associated with lower ACC GABA in depressed youth. Together, the current results suggest opposite abnormalities in GABA neurotransmission between cortical and subcortical reward-related brain regions in adolescent depression, which highlights the complexity of GABA neurotransmission in this population. Further work is necessary to investigate the mechanisms underlying region-specific GABAergic dysfunction within the reward circuitry of depressed youth.

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References

- Abdallah CG, Jackowski A, Sato JR, Mao X, Kang G, Cheema R, et al., 2015 Prefrontal cortical GABA abnormalities are associated with reduced hippocampal volume in major depressive disorder. *Eur. Neuropsychopharmacol* 25, 1082–1090. [PubMed: 25983019]
- APA, 2000 Diagnostic and Statistical Manual of Mental Disorders: DSM-IV-TR American Psychiatric Association, Washington, DC.
- Avshalumov MV, Chen BT, Marshall SP, Pena DM, Rice ME, 2003 Glutamate-dependent inhibition of dopamine release in striatum is mediated by a new diffusible messenger, H₂O₂. *J. Neurosci* 23, 2744–2750. [PubMed: 12684460]
- Beck AT, Ward CH, Mendelson M, Mock J, Erbaugh J, 1961 An inventory for measuring depression. *Arch. Gen. Psychiatry* 4, 561–571. [PubMed: 13688369]
- Beck AT, Kovacs M, Weissman A, 1979 Assessment of suicidal intention: the scale for suicide ideation. *J. Consult. Clin. Psychol* 47, 343–352. [PubMed: 469082]
- Beck AT, Steer RA, Garbin MG, 1988 Psychometric properties of the beck depression inventory: Twenty-five years of evaluation. *Clin. Psychol. Rev* 8, 77–100.
- Beck AT, Guth D, Steer RA, Ball R, 1997 Screening for major depression disorders in medical inpatients with the beck depression inventory for primary care. *Behav. Res. Therapy* 35, 785–791.
- Bonanno G, Raiteri M, 1987 Coexistence of carriers for dopamine and GABA uptake on a same nerve terminal in the rat brain. *Br. J. Pharmacol* 91, 237–243. [PubMed: 3594080]

- Brambilla P, Perez J, Barale F, Schettini G, Soares JC, 2003 GABAergic dysfunction in mood disorders. *Mol. Psychiatry* 8 (721–737), 715.
- Brisch R, Saniotis A, Wolf R, Bielau H, Bernstein HG, Steiner J, et al., 2014 The role of dopamine in schizophrenia from a neurobiological and evolutionary perspective: old fashioned, but still in vogue. *Front. Psychiatry* 5, 47. [PubMed: 24904434]
- Bustillo JR, 2013 Use of proton magnetic resonance spectroscopy in the treatment of psychiatric disorders: a critical update. *Dialogues Clin. Neurosci* 15, 329–337. [PubMed: 24174904]
- Chiapponi C, Piras F, Piras F, Caltagirone C, Spalletta G, 2016 GABA system in schizophrenia and mood disorders: a mini review on third-generation imaging studies. *Front. Psychiatry* 7, 61. [PubMed: 27148090]
- Choi IY, Lee SP, Merkle H, Shen J, 2006 In vivo detection of gray and white matter differences in GABA concentration in the human brain. *NeuroImage* 33, 85–93. [PubMed: 16884929]
- De Bellis MD, Keshavan MS, Beers SR, Hall J, Frustaci K, Masalehdan A, et al., 2001 Sex differences in brain maturation during childhood and adolescence. *Cereb. Cortex* 11, 552–557. [PubMed: 11375916]
- Delgado MR, 2007 Reward-related responses in the human striatum. *Ann. NY Acad. Sci* 1104, 70–88. [PubMed: 17344522]
- Der-Avakian A, Markou A, 2012 The neurobiology of anhedonia and other reward-related deficits. *Trends Neurosci* 35, 68–77. [PubMed: 22177980]
- Freed RD, Coffey BJ, Mao X, Weiduschat N, Kang G, Shungu DC, et al., 2016 Decreased anterior cingulate cortex gamma-aminobutyric acid in youth with Tourette's disorder. *Pediatr. Neurol* 65, 64–70. [PubMed: 27743746]
- Gabbay V, Mao X, Klein RG, Ely BA, Babb JS, Panzer AM, et al., 2012 Anterior cingulate cortex gamma-aminobutyric acid in depressed adolescents: relationship to anhedonia. *Arch. Gen. Psychiatry* 69, 139–149. [PubMed: 21969419]
- Gabbay V, Bradley KA, Mao X, Ostrover R, Kang G, Shungu DC, 2017 Anterior cingulate cortex gamma-aminobutyric acid deficits in youth with depression. *Transl. Psychiatry* 7, e1216. [PubMed: 28892070]
- Garbutt JC, van Kammen DP, 1983 The interaction between GABA and dopamine: implications for schizophrenia. *Schizophr. Bull* 9, 336–353. [PubMed: 6137869]
- Gerner RH, Hare TA, 1981 Csf GABA in normal subjects and patients with depression, schizophrenia, mania, and anorexia nervosa. *Am. J. Psychiatry* 138, 1098–1101. [PubMed: 7258390]
- Haber, 2011 Chapter 11: neuroanatomy of reward: a view from the ventral striatum. In: Gottfried JA (Ed.), *Neurobiology of Sensation and Reward* CRC Press/Taylor & Francis, Boca Raton, FL.
- Hasler G, van der Veen JW, Tumonis T, Meyers N, Shen J, Drevets WC, 2007 Reduced prefrontal glutamate/glutamine and gamma-aminobutyric acid levels in major depression determined using proton magnetic resonance spectroscopy. *Arch. Gen. Psychiatry* 64, 193–200. [PubMed: 17283286]
- Heshmati M, Russo SJ, 2015 Anhedonia and the brain reward circuitry in depression. *Curr. Behav. Neurosci. Rep* 2, 146–153. [PubMed: 26525751]
- Hulvershorn LA, Cullen K, Anand A, 2011 Toward dysfunctional connectivity: a review of neuroimaging findings in pediatric major depressive disorder. *Brain Imag. Behav* 5, 307–328.
- Iorfino F, Hickie IB, Lee RS, Lagopoulos J, Hermens DF, 2016 The underlying neurobiology of key functional domains in young people with mood and anxiety disorders: a systematic review. *BMC Psychiatry* 16, 156. [PubMed: 27215830]
- Jensen JE, Frederick Bde B., Renshaw PF, 2005 Grey and white matter GABA level differences in the human brain using two-dimensional, j-resolved spectroscopic imaging. *NMR Biomed* 18, 570–576. [PubMed: 16273508]
- Jones MW, Kilpatrick IC, Phillipson OT, 1988 Dopamine function in the prefrontal cortex of the rat is sensitive to a reduction of tonic GABA-mediated inhibition in the thalamic mediodorsal nucleus. *Exp. Brain Res* 69, 623–634. [PubMed: 3371442]
- Kasa K, Otsuki S, Yamamoto M, Sato M, Kuroda H, Ogawa N, 1982 Cerebrospinal fluid gamma-aminobutyric acid and homovanillic acid in depressive disorders. *Biol. Psychiatry* 17, 877–883. [PubMed: 7115838]

- Kaufman AS, Kaufman NL, 1990 Manual for the Kaufman Brief Intelligence Test American Guidance Service, Circle Pines, MN.
- Kaufman J, Birmaher B, Brent D, Rao U, Flynn C, Moreci P, et al., 1997 Schedule for affective disorders and schizophrenia for school-age children-present and lifetime version (k-sads-pl): initial reliability and validity data. *J. Am. Acad. Child Adolesc. Psychiatry* 36, 980–988. [PubMed: 9204677]
- Kessler RC, Berglund P, Demler O, Jin R, Merikangas KR, Walters EE, 2005 Lifetime prevalence and age-of-onset distributions of dsm-iv disorders in the national comorbidity survey replication. *Arch. Gen. Psychiatry* 62, 593–602. [PubMed: 15939837]
- Lu YR, Fu XY, Shi LG, Jiang Y, Wu JL, Weng XJ, et al., 2014 Decreased plasma neuroactive amino acids and increased nitric oxide levels in melancholic major depressive disorder. *BMC Psychiatry* 14, 123. [PubMed: 24767108]
- March JS, Parker JD, Sullivan K, Stallings P, Conners CK, 1997 The multidimensional anxiety scale for children (MASC): factor structure, reliability, and validity. *J. Am. Acad. Child Adolesc. Psychiatry* 36, 554–565. [PubMed: 9100431]
- Marchand WR, Yurgelun-Todd D, 2010 Striatal structure and function in mood disorders: a comprehensive review. *Bipolar Disord* 12, 764–785. [PubMed: 21176024]
- Markwardt SJ, Wadiche JI, Overstreet-Wadiche LS, 2009 Input-specific GABAergic signaling to newborn neurons in adult dentate gyrus. *J. Neurosci* 29, 15063–15072. [PubMed: 19955357]
- Miller AH, Haroon E, Raison CL, Felger JC, 2013 Cytokine targets in the brain: impact on neurotransmitters and neurocircuits. *Depress. Anxiety* 30, 297–306. [PubMed: 23468190]
- Petty F, Kramer GL, Dunnam D, Rush AJ, 1990 Plasma GABA in mood disorders. *Psychopharmacol. Bull* 26, 157–161. [PubMed: 2236451]
- Petty F, Kramer GL, Gullion CM, Rush AJ, 1992 Low plasma gamma-aminobutyric acid levels in male patients with depression. *Biol. Psychiatry* 32, 354–363. [PubMed: 1420649]
- Poznanski EO, Mokros H, 1996 Children's Depression Rating Scale-Revised (CDRS-R) Western Psychological Services, Los Angeles, CA.
- Poznanski EO, Grossman JA, Buchsbaum Y, Banegas M, Freeman L, Gibbons R, 1984 Preliminary studies of the reliability and validity of the children's depression rating scale. *J. Am. Acad. Child Psychiatry* 23, 191–197. [PubMed: 6715741]
- Poznanski E, Freeman L, Mokros H, 1985 Children's depression rating scale-revised. *Psychopharmacol. Bull* 21, 979–989.
- Price RB, Shungu DC, Mao X, Nestadt P, Kelly C, Collins KA, et al., 2009 Amino acid neurotransmitters assessed by proton magnetic resonance spectroscopy: relationship to treatment resistance in major depressive disorder. *Biol. Psychiatry* 65, 792–800. [PubMed: 19058788]
- Pycock CJ, Horton RW, 1979 Dopamine-dependent hyperactivity in the rat following manipulation of GABA mechanisms in the region of the nucleus accumbens. *J. Neural Transm* 45, 17–33. [PubMed: 112217]
- Quetscher C, Yildiz A, Dharmadhikari S, Glaubit B, Schmidt-Wilcke T, Dydak U, et al., 2015 Striatal GABA-mrs predicts response inhibition performance and its cortical electrophysiological correlates. *Brain Struct. Funct* 220, 3555–3564. [PubMed: 25156575]
- Reid M, Herrera-Marschitz M, Hokfelt T, Terenius L, Ungerstedt U, 1988 Differential modulation of striatal dopamine release by intranigral injection of gamma-aminobutyric acid (GABA), dynorphin a and substance p. *Eur. J. Pharmacol* 147, 411–420. [PubMed: 2454197]
- Rothman DL, Petroff OA, Behar KL, Mattson RH, 1993 Localized 1h nmr measurements of gamma-aminobutyric acid in human brain in vivo. *Proc. Natl. Acad. Sci. U. S. A* 90, 5662–5666. [PubMed: 8516315]
- Sanacora G, Mason GF, Rothman DL, Behar KL, Hyder F, Petroff OA, et al., 1999 Reduced cortical gamma-aminobutyric acid levels in depressed patients determined by proton magnetic resonance spectroscopy. *Arch. Gen. Psychiatry* 56, 1043–1047. [PubMed: 10565505]
- Sanacora G, Gueorguieva R, Epperson CN, Wu YT, Appel M, Rothman DL, et al., 2004 Subtype-specific alterations of gamma-aminobutyric acid and glutamate in patients with major depression. *Arch. Gen. Psychiatry* 61, 705–713. [PubMed: 15237082]

- Schur RR, Draisma LW, Wijnen JP, Boks MP, Koevoets MG, Joels M, et al., 2016 Brain GABA levels across psychiatric disorders: a systematic literature review and meta-analysis of (1) h-mrs studies. *Hum. Brain Mapp* 37, 3337–3352. [PubMed: 27145016]
- Shaw A, Brealy J, Richardson H, Muthukumaraswamy SD, Edden RA, John Evans C., et al., 2013 Marked reductions in visual evoked responses but not gamma-aminobutyric acid concentrations or gamma-band measures in remitted depression. *Biol. Psychiatry* 73, 691–698. [PubMed: 23200528]
- Shungu DC, Mao X, Gonzales R, Soones TN, Dyke JP, van der Veen JW, et al., 2016 Brain gamma-aminobutyric acid (GABA) detection in vivo with the j-editing (1) h mrs technique: a comprehensive methodological evaluation of sensitivity enhancement, macromolecule contamination and test-retest reliability. *NMR Biomed* 29, 932–942. [PubMed: 27173449]
- Sulzer D, Cragg SJ, Rice ME, 2016 Striatal dopamine neurotransmission: regulation of release and uptake. *Basal Ganglia* 6, 123–148. [PubMed: 27141430]
- Taylor MJ, Mannie ZN, Norbury R, Near J, Cowen PJ, 2011 Elevated cortical glutamate in young people at increased familial risk of depression. *Int. J. Neuropsychopharmacol* 14, 255–259. [PubMed: 20846462]
- van Zessen R, Phillips JL, Budygin EA, Stuber GD, 2012 Activation of vta gaba neurons disrupts reward consumption. *Neuron* 73, 1184–1194. [PubMed: 22445345]

Fig. 1. ACC and striatal GABA levels.

Depressed youth had higher striatal GABA than HC ($p = .001$). GABA was significantly higher in the striatum than the ACC in both depressed youth ($p < .0005$) and HC ($p < .0005$), although this difference was greater in MDD.

Fig. 2.

A) Line graphs of individual ACC and striatal GABA levels for all depressed participants.

B) Significant correlation between the deviations from the average HC levels for striatal GABA and ACC GABA in depressed youth. Higher striatal GABA above the HC mean was correlated with lower ACC GABA below the HC mean.

Table 1

Demographic and clinical characteristics of the study sample.

Demographic variables	MDD (<i>n</i> = 20)	HC (<i>n</i> = 16)	<i>p</i>
Age [mean ± SD]	15.49 ± 2.46	15.56 ± 2.64	0.94
Sex [<i>n</i> female] (%)	8 (40%)	8 (50%)	0.55
Ethnicity [<i>n</i>] (%)			
Caucasian	12 (60%)	9 (56.25%)	0.25
African American	6 (30%)	5 (31.25%)	
Asian	2 (10%)	0 (0%)	
Other	0 (0%)	2 (12.5%)	
Clinical variables [mean ± SD] (range)			
Anhedonia	6.45 ± 2.82 (2–12)	2.22 ± 0.67 (2–4)	< 0.0005*
CDRS-R	46.95 ± 11.82 (35–79)	18.50 ± 2.76 (17–25)	< 0.0005*
BDI-II	19.70 ± 12.16 (0–40)	2.73 ± 3.72 (0–10)	< 0.0005*
BSSI	4.50 ± 6.41 (0–21)	0.10 ± 0.32 (0–1)	0.015*
MASC total	45.53 ± 17.61 (2–76)	28.53 ± 15.55 (4–57)	0.006*
Illness history			
Current comorbidity [<i>n</i>] (%)			
Attention-deficit/hyperactivity disorder	6 (30)	0 (0)	
Generalized anxiety disorder	8 (40)	0 (0)	
Separation anxiety disorder	1 (5)	0 (0)	
Specific phobia	1 (5)	0 (0)	
Social anxiety disorder	5 (25)	0 (0)	
Obsessive-compulsive disorder	1 (5)	0 (0)	
Post-traumatic stress disorder	1 (5)	0 (0)	
Oppositional defiant disorder	4 (20)	0 (0)	
Enuresis disorder not otherwise specified	2 (10)	0 (0)	

Abbreviations: BDI-II = Beck depression inventory, second edition; BSSI = Beck scale of suicidal ideation; CDRS-R = Children's depression rating scale-revised; HC = healthy controls; MDD = major depressive disorder; MASC = multidimensional anxiety scale for children.

* Significant difference between the MDD and HC groups ($p < .05$)

Table 2

Striatal voxel tissue heterogeneity variables.

	MDD (<i>n</i> = 20)		HC (<i>n</i> = 16)		<i>p</i>[*]
	Mean	SD	Mean	SD	
WM %	27.94	2.85	27.36	4.63	0.49
GM %	71.13	2.71	72.13	5.50	0.61
CSF %	0.84	0.59	0.48	0.44	0.12
W	9.85×10^{11}	9.03×10^{10}	1.07×10^{12}	1.84×10^{11}	0.06

Abbreviations: CSF = cerebrospinal fluid; GM = gray matter; HC = healthy controls; MDD = major depressive disorder; SD = standard deviation; W = unsuppressed voxel tissue water signal; WM = white matter.

^{*} No significant differences between MDD and HC groups ($p < .05$).

Table 3

Striatal and ACC GABA levels.

	MDD (<i>n</i> = 20)		HC (<i>n</i> = 16)		<i>p</i>
	Mean	SD	Mean	SD	
Striatal GABA/W	4.61 × 10 ⁻³ ***	0.66 × 10 ⁻³	3.88 × 10 ⁻³ ***	0.45 × 10 ⁻³	0.001*
ACC GABA/W	2.78 × 10 ⁻³	0.33 × 10 ⁻³	3.02 × 10 ⁻³	0.50 × 10 ⁻³	0.07**

Abbreviations: ACC = anterior cingulate cortex; GABA = γ -aminobutyric acid; HC = healthy controls; MDD = major depressive disorder; SD = standard deviation; W = unsuppressed voxel tissue water signal.

* Significant difference between MDD and HC groups ($p < .05$).

** Approaches significant difference between MDD and HC groups ($p < .10$).

*** Significant difference between striatal and ACC GABA levels ($p < .0005$).