Lower *N*-Acetyl-Aspartate Levels in Prefrontal Cortices in Pediatric Bipolar Disorder: A ^{1}H Magnetic Resonance Spectroscopy Study

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Objective: The few studies applying single-voxel ^{1}H spectroscopy in children and adolescents with bipolar disorder (BD) have reported low *N*-acetyl-aspartate (NAA) levels in the dorsolateral prefrontal cortex (DLPFC), and high myo-inositol / phosphocreatine plus creatine (PCr+Cr) ratios in the anterior cingulate. The aim of this study was to evaluate NAA, glycerophosphocholine plus phosphocholine (GPC+PC) and PCr+Cr in various frontal cortical areas in children and adolescents with BD. We hypothesized that NAA levels within the prefrontal cortex are lower in BD patients than in healthy controls, indicating neurodevelopmental alterations in the former. **Method:** We studied 43 pediatric patients with *DSM-IV* BD (19 female, mean age 13.2 ± 2.9 years) and 38 healthy controls (19 female, mean age 13.9 ± 2.7 years). We conducted multivoxel in vivo ^{1}H spectroscopy measurements at 1.5 Tesla using a long echo time of 272 ms to obtain bilateral metabolite levels from the medial prefrontal cortex (MPFC), DLPFC (white and gray matter), cingulate (anterior and posterior), and occipital lobes. We used the nonparametric Mann–Whitney *U* test to compare neurochemical levels between groups. **Results:** In pediatric BD patients, NAA and GPC+PC levels in the bilateral MPFC, and PCr+Cr levels in the left MPFC were lower than those seen in the controls. In the left DLPFC white matter, levels of NAA and PCr+Cr were also lower in BD patients than in controls. **Conclusions:** Lower NAA and PCr+Cr levels in the PFC of children and adolescents with BD may be indicative of abnormal dendritic arborization and neuropil, suggesting neurodevelopmental abnormalities. J. Am. Acad. Child Adolesc. Psychiatry, 2011;50(1): 85–94. **Key words:** magnetic resonance spectroscopy, bipolar disorder, prefrontal cortex, *N*-acetyl aspartate

Proton magnetic resonance spectroscopy (^{1}H MRS) is a noninvasive neuroimaging technique that measures neurochemicals such as *N*-acetyl aspartate (NAA), as well as the choline-containing compounds glycerophosphocholine plus phosphocholine (GPC+PC) and the compounds phosphocreatine plus creatine (PCr+Cr).

NAA is related to myelin formation and participates in the energy metabolism of neuronal mitochondria.[1,2] During early development, NAA levels increase in parallel with dendritic arborization and the formation of synaptic connections, and NAA is therefore considered a neuroaxonal marker of tissue function.[3,4] Low NAA levels can occur as a consequence of neuropil reduction and axonal metabolic dysfunction.[5]

The peaks in GPC+PC levels include very small amounts of glycerophosphocholine (a metabolite in the catabolic pathway of membrane phospholipids) and phosphocholine (a precursor of membrane phospholipids), and membrane phospholipids in the bilayer structure function as a barrier between cellular components such as neuronal dendrites and synaptic connections. The GPC+PC compound consists of membrane phospholipid metabolites involved in membrane synthesis and breakdown, and products of recep-

tor-mediated lecithin hydrolysis also serve as important second messengers in signal cascades that control cell growth. This compound is also a precursor of the synthesis of acetylcholine, the neurotransmitter involved in memory and cognition.[6]

The PCr+Cr peaks contain mostly phosphocreatine and creatine. The reaction between phosphocreatine and creatine serves as energy storage of phosphates and the adenosine 5′-triphosphate/adenosine diphosphate ratio. Therefore, PCr+Cr is essential for the regeneration of adenosine 5′-triphosphate consumed by the cell, and decreased PCr+Cr concentrations suggest decreased energy metabolism.[7,8]

In adults with bipolar disorder (BD), phosphorus MRS (^{31}P-MRS) that measures high-energy phosphate metabolism, and ^{1}H MRS suggest a mechanism of mitochondrial dysfunction that involves a decrease in total cellular energy production and altered phospholipid metabolism.[1] However, ^{1}H MRS has been underused as a tool in the study of pediatric BD and the underlying neurodevelopmental processes.[9-11]

The few studies applying single-voxel ^{1}H MRS in children and adolescents with BD, compared to healthy controls, have reported lower NAA levels and NAA/PCr+Cr ratios in the dorsolateral prefrontal cortex (DLPFC),[12,13] lower levels of NAA and GP+PC in the orbitofrontal cortex,[14] higher myo-inositol/PCr+Cr ratios in the anterior cingulate,[15,16] and lower glutamate plus glutamine levels and higher glutamate plus glutamine/PCr+Cr ratios in the frontal lobes and basal ganglia.[17] However, other studies demonstrate that NAA levels in the anterior cingulate and DLPFC of pediatric BD patients are comparable to those observed in healthy controls,[16,18] Table 1[12-25] summarizes current available evidence on ^{1}H MRS findings among children and adolescents with BD.

Currently, in a single MRI scan session, multiple in vivo ^{1}H MRS spectra can be acquired to simultaneously measure metabolite levels in multiple brain areas.[7,26] This approach would broaden the neurochemical investigation of specific brain areas that are interconnected and functionally related to the symptomatology of mood disorders, mainly the fronto-limbic areas, is the model proposed by various researchers.[10,11] Specifically in pediatric BD, a comprehensive neurochemical examination of these frontal areas would shed light on how this disorder affects the developing brain.

Previously, we reported lower NAA levels within the left DLPFC of children and adolescents with type I or type II BD compared to healthy controls in a single-voxel H MRS approach.[20] Now, in our current study we enlarged our sample to include those with BD not otherwise specified (NOS) and also adopted a multivoxel ^{1}H MRS approach so as to simultaneously evaluate the following a priori regions of interest (ROIs): the medial prefrontal cortex (MPFC), the DLPFC (white and gray matter), and the cingulate (anterior and posterior), to obtain a neurochemical profile of these various frontal areas, all of which are involved in mood regulation. Indeed, PFC and cingulate functions, such as decision making, executive functions, and emotion integration, have been reported as abnormal in BD youth.[27] We hypothesized that NAA levels in the DLPFC, MPFC and anterior cingulate are lower in children and adolescents with BD compared with healthy controls, which could be indicative of neurodevelopmental abnormalities. As a control region, we included the occipital cortex in our study, as it is not believed involved in any neural pathway that regulates mood and therefore was expected to present normal levels of the neurochemicals evaluated.

METHOD

Subjects

Children and adolescents to be included in the control group were recruited from newspaper and television advertisements and flyers posted in the community. Children and adolescents with BD were referred by local psychiatrists. In both cases, only those individuals between 8 and 17 years of age were included. Individuals with serious medical problems or contraindications to magnetic resonance imaging (MRI) were excluded. Patients were included if they had been diagnosed with BD according to *DSM-IV* criteria (1994). Patients with BD presenting with substance abuse or dependence in the 6 months preceding study enrollment were excluded, as were those with a history of schizophrenia, developmental disorders, eating disorders, Tourette syndrome, or mental retardation. For healthy controls, exclusion criteria were any lifetime Axis I *DSM-IV* psychiatric disorder and history of any Axis I psychiatric disorder in first-degree relatives.

A total of 43 pediatric patients with *DSM-IV*–diagnosed BD (27 with BD type I, 10 with BD type II, and 6 with BD NOS) were compared with 38 healthy controls. The two groups were comparable in terms of