Available online at www.sciencedirect.com
SCIENCE DIRECT®
Neuroscience Letters 384 (2005) 321–326
Neuroscience Letters
www.elsevier.com/locate/neulet

Proton spectroscopy study of the left dorsolateral prefrontal cortex in pediatric depressed patients

Sheila C. Caetano [a,b,c], Manoela Fonseca [a,b,d], Rene L. Olvera [e], Mark Nicoletti [a], John P. Hatch [a,f], Jeffrey A. Stanley [g], Kristina Hunter [a], Beny Lafer [c], Steven R. Pliszka [e], Jair C. Soares [a,b,h,*]

[a] *Division of Mood and Anxiety Disorders, Department of Psychiatry, The University of Texas Health Science Center at San Antonio, 7703 Floyd Curl Drive, San Antonio, TX 78229, USA*
[b] *Psychiatry Service, South Texas Veterans Health Care System, Audie L. Murphy Division, San Antonio, TX, USA*
[c] *Department of Psychiatry, Institute of Psychiatry, University of São Paulo School of Medicine, São Paulo, Brazil*
[d] *Psychiatry Research Unit, Federal University of Rio Grande do Sul, School of Medicine, Porto Alegre, Brazil*
[e] *Division of Child and Adolescent Psychiatry, Department of Psychiatry, The University of Texas Health Science Center at San Antonio, San Antonio, TX, USA*
[f] *Department of Orthodontics, The University of Texas Health Science Center at San Antonio, San Antonio, TX, USA*
[g] *Department of Psychiatry and Behavioral Neurosciences, Wayne State University School of Medicine, Detroit, MI, USA*
[h] *Department of Radiology, The University of Texas Health Science Center at San Antonio, San Antonio, TX, USA*

Received 22 February 2005; received in revised form 28 April 2005; accepted 29 April 2005

Abstract

The dorsolateral prefrontal cortex (DLPFC) plays an essential role in mood regulation and integration of cognitive functions that are abnormal in major depressive disorder (MDD). Few neuroimaging studies have evaluated the still maturing DLPFC in depressed children and adolescents. We conducted single voxel proton magnetic resonance spectroscopy (^{1}H MRS) of the left DLPFC in 14 depressed children and adolescents (13.3 ± 2.3 years old, 10 males) and 22 matched healthy controls (13.6 ± 2.8 years old, 13 males). Depressed subjects had significantly lower levels of glycerophosphocholine plus phosphocholine (GPC + PC; or choline-containing compounds) and higher myo-inositol levels in the left DLPFC compared to healthy controls. In the depressed subjects, we found significant inverse correlations between glutamate levels and both duration of illness and number of episodes. In healthy controls there was a significant direct correlation between age and glutamine levels, which was not present in the patient group. Lower GPC + PC levels in pediatric MDD may reflect lower cell membrane content per volume in the DLPFC. Increased myo-inositol levels in MDD may represent a disturbed secondary messenger system. GPC + PC and myo-inositol abnormalities further demonstrate the involvement of DLPFC in pediatric MDD.

Keywords: Choline-containing compounds; Myo-inositol; Major depression; Children; Dorsolateral prefrontal cortex; Proton spectroscopy

Major depressive disorder (MDD) has a lifetime prevalence of 15% in childhood and adolescence [19] and often presents a chronic course associated with functional impairment [32]. The investigation of the pathophysiology of MDD has lead to the discovery of abnormalities in the neural structures involved in mood regulation [36]. In particular, the dorsolateral prefrontal cortex (DLPFC, Brodmann areas 9/46) plays an essential role in mood regulation and working memory [10]. DLPFC abnormalities have been consistently found in post-mortem [7,29,40], neuroanatomical [12], neurochemical [3,16,23] and functional [2,8,18,20] studies conducted in depressed adults. Interestingly, the DLPFC is one of the last regions to mature in the human brain, probably because of its integrative role in cognitive functions [11,37]. Nevertheless, the few in vivo anatomical [24] and neurochemical [9] studies that have evaluated the DLPFC in children and adolescents suffering from MDD noted abnormalities in the left side.

* Corresponding author. Tel.: +1 210 567 5492; fax: +1 210 567 3759.
E-mail address: soares@uthscsa.edu (J.C. Soares).

0304-3940/$ – see front matter © 2005 Elsevier Ireland Ltd. All rights reserved.
doi:10.1016/j.neulet.2005.04.099

Proton (^{1}H) spectroscopy is a non-invasive and non-radioactive neuroimaging tool, which allows measurement of major brain chemicals such as choline-containing compounds glycerolphosphocholine + phosphocholine (GPC + PC), myo-inositol (Ino), *N*-acetyl aspartate (NAA), phosphocreatine + creatine (PCr + Cr), glutamate (Glu) and glutamine (Gln) [38]. ^{1}H spectroscopy studies of MDD adults have demonstrated multiple brain chemical abnormalities. For instance, MDD adults show lower GPC + PC/PCr + Cr ratios in the left amygdala [17] and in the basal ganglia [30], lower Glu + Gln in the cingulate [1], lower NAA/PCr + Cr ratios in the caudate, higher GPC + PC/PCr + Cr ratios in the putamen [41] compared to healthy controls. Specifically in the *left* DLPFC, MDD patients show higher GPC + PC/PCr + Cr and Ino/PCr + Cr ratios [16] and lower NAA/PCr + Cr ratios [23].

Post-mortem studies in adult depressed patients suggest reduced number and density of DLPFC glial cells [7,29,40]. This finding is supported by functional studies showing reduced glucose metabolism and blood flow in the DLPFC of adult depressed patients [2,8,18,20]. Interestingly, the choline-containing compounds have been associated with membrane synthesis and repair, intracellular signal transduction and the myelination process [38], which leads us to expect a reduction of GPC + PC in MDD.

We used ^{1}H spectroscopy to compare the neurochemistry of the left DLPFC of children and adolescents suffering from MDD with that of age- and gender-matched healthy controls. Based on the previous postmortem and functional findings, we expected to find lower GPC + PC in pediatric depressed patients compared to controls. We also hypothesized that we would replicate higher Ino in pediatric depressed patients compared to healthy controls [9].

MDD subjects ($N = 14$) were included if they met DSM-IV diagnostic criteria for MDD and excluded if they met lifetime diagnostic criteria for psychotic disorders, bipolar disorder, developmental disorders, substance abuse/dependence, eating disorders, Tourette' Disease, or mental retardation (demographic and clinical characteristics of pediatric MDD patients and healthy subjects are presented in Table 1).

Table 1
Demographic and clinical characteristics of pediatric MDD patients and healthy subjects

	Pediatric MDD patients ($N = 14$)	Healthy controls ($N = 22$)
Age (years)	13.3 ± 2.3 (9.5–17.2)	13.6 ± 2.8 (8.5–17.7)
Gender (males)	10	13
Education (years)	7.1 ± 2.5	7.6 ± 2.9
SES	44.6 ± 12.7	43.4 ± 12.9
GAF	52.6 ± 6.9	87.9 ± 10.6
Age of onset (years)	10.4 ± 2.3	
Duration of illness (months)	29.13 ± 13.7	
Number of episodes	1.7 ± 1.6	
CDRS	46.6 ± 16.5	

Data are presented as mean ± standard deviation (range).

Of the 14 depressed patients, 4 had attention deficit hyperactivity disorder (ADHD), 5 had generalized anxiety, 3 had separation anxiety, 2 had social phobia, and 1 had panic disorder. Eleven patients had a positive family history of mood disorders among their first degree-relatives (*MDD* ($N = 11$) and bipolar disorder ($N = 3$)), and all patients had a positive family history of Axis I psychiatric disorders in a first degree-relative. Eight patients were free of psychotropic medication (6 were medication-naïve, 1 had last taken medication 1 year prior to the study and 1 was off medication for 2 years) and six patients were on psychotropic medication (antidepressants: sertraline ($N = 2$), escitalopram ($N = 2$), paroxetine ($N = 1$); and/or stimulants: methylphenidate ($N = 1$) and atomoxetine ($N = 1$)).

Healthy controls ($N = 22$) were matched to the MDD subjects for age, gender and puberty status. Exclusion criteria for healthy controls were history of or current psychiatric disorder, and history of any Axis I psychiatric disorder in first-degree relatives.

Patients and controls were also required to be between 8 and 17 years old, not to have serious medical problems, and to be magnetic resonance compatible. All subjects and their parents or legal guardians were assessed using the Schedule for Affective Disorders and Schizophrenia for School-Age Children—Present and Lifetime version (K-SADS-PL) interview [15]. The severity of depression was rated using the Children's Depression Rating Scale, revised (CDRS) [27]. Puberty status was assessed through the Pubertal Development Scale–Petersen Scale [25]. Scores on the Global Assessment of Functioning Scale (GAF) were recorded. Paternal socioeconomic status (SES) was scored according to the Hollingshead Socioeconomic status [13]. This study was approved by the Institutional Review Board of The University of Texas Health Science Center at San Antonio. After the study was fully explained, written informed consent was obtained from all subjects and their parents or legal guardians.

All ^{1}H spectroscopy scans were performed on a 1.5 T Philips Intera 8.1.1. scanner at the South Texas Veterans Health Care System (Audie Murphy Division). The MRI studies were performed with a T1-weighted fast field echo sequence (3D T1-FFE), with repetition time (TR) of 25 ms, echo time (TE) of 5 ms, field of view (FOV) of 240 mm × 220 mm, slice thickness of 1.0 mm, gap = 0, number of excitations (NEX) of 2, and matrix size of 256 × 192.

A single-voxel ^{1}H spectroscopy approach [PRESS sequence (point-resolved spectroscopy), TR of 6 s, TE of 30 ms, bandwidth of 4 kHz, 4096 complex data points] was used to localize a 2 cm × 2 cm × 2 cm voxel placed in the left DLPFC. Based on high-resolution anatomical images, the superior frontal sulcus, the lateral fissure, and the genu of corpus callosum were used as anatomical landmarks for placement of the MRS voxel [14] (Fig. 1). We also collected water-unsuppressed spectra for absolute quantification. The LC Model software package was used to quantify the ^{1}H metabolites which include: NAA, Gln, Glu, Ino, GPC + PC, PCr + Cr, taurine, alanine, aspartate, γ-amino-butyric acid (GABA),