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Early Metabolic Development of Posteromedial Cortex and Thalamus in Humans Analyzed Via In Vivo Quantitative Magnetic Resonance Spectroscopy

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Abstract

The posteromedial cortex (PMC) including the posterior cingulate, retrosplenial cortex, and medial parietal cortex/precuneus is an epicenter of cortical interactions in a wide spectrum of neural activity. Anatomic connections between PMC and thalamic components have been established in animal studies, but similar studies do not exist for the fetal and neonatal period. Magnetic resonance spectroscopy (MRS) allows for noninvasive measurement of metabolites in early development. Using single-voxel 3-T MRS, healthy term neonates (n 5 31, mean postconception age 41.5 weeks $\pm$ 3.8 weeks) were compared with control children (n 5 23, mean age 9.4 years $\pm$ 5.1 years) and young adults (n 5 10, mean age 24.1 years $\pm$ 2.6 years). LCModel-based calculations compared metabolites within medial parietal gray matter (colocalizing to the PMC), posterior thalamus, and parietal white matter voxels. Common metabolic changes existed for neuronal–axonal maturation and structural markers in the PMC, thalamus, and parietal white matter with increasing NAA and glutamate and decreasing myoinositol and choline with age. Key differences in creatine and glucose metabolism were noted in the PMC, in contrast to the thalamic and parietal white matter locations, suggesting a unique role of energy metabolism. Significant parallel metabolite developmental changes of multiple other metabolites including aspartate, glutamine, and glutathione with age were present between PMC and parietal white matter but not

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ROLE OF AUTHORS

AJD participated in study design, compiled data, performed statistical analysis, interpreted data, authored graphics, and wrote and edited the manuscript; RC participated in data collection and statistical analysis; VL participated in data analysis and authored graphics; VJS performed statistical analysis and participated in interpretation; SB participated in study design, performed supplemental T1/T2 effect calculations, participated in data interpretation, and provided critical revision of the manuscript; AP participated in study design, data interpretation, and critical revision of the manuscript.

CONFLICT OF INTEREST STATEMENT

The authors have no conflicts of interest to declare.

between PMC and thalamus. These findings offer insight into the metabolic architecture of the interface between structural and functional topology of brain networks. Further investigation unifying metabolic changes with functional and anatomic pathways may further enhance the understanding of the PMC in posterior default mode network development.

Keywords

magnetic resonance spectroscopy; brain development; metabolism; posteromedial cortex; thalamus

The posteromedial cortex (PMC), consisting of the posterior cingulate, retrosplenial cortex, and medial parietal cortex/precuneus, is an epicenter of cortical interactions among many areas of the brain in a wide spectrum of neural activity (Parvizi et al., 2006; Buckwalter et al., 2008). From a neuronal connectivity standpoint, the PMC retains a vital role within the resting-state neural networks identified by functional connectivity or resting-state MRI (RS-fMRI) as part of the posterior default mode network (DMN). The structures of the PMC appear to function uniformly as particularly robust components within the posterior DMN involved in self-referential thought and to be downregulated in during-task activities (Tzourio-Mazoyer et al., 2002; Pfefferbaum et al., 2011).

Rudimentary elements of the posterior DMN including the PMC are present during early infancy as indicated by RS-fMRI investigations (Fransson et al., 2007). More recent structural–functional covariance research reinforces the concept of limited posterior DMN precursors lacking covariance of anatomical structural and functional activity between the PCC and frontal regions in infancy and early childhood, consistent with RS-fMRI and EEG investigations demonstrating increasing local functional connectivity in early development and limited anterior–posterior interactions that progressively mature across childhood (Srinivasan, 1999; Fransson et al., 2007; Fair et al., 2008; Smyser et al., 2010; Zielinski et al., 2010). Graph theory analysis demonstrates strong centrality of the PCC and retrosplenium components of the PMC for both infants and young children, whereas prefrontal cortex centrality emerges later between 1 and 2 years of age (Gao et al., 2009). These studies allude to a critical period of development within the PMC that proliferates during the first few years of life and extends during childhood involving maturation of both neural structure and functional interactions to form an essential, primary cortical hub.

Although neuroanatomically validated connections exist between the PMC and thalamus as integral mature gray matter structures (Buckwalter et al., 2008), similar data do not exist for fetal and neonatal primates or nonprimates. Both seed-based and independent components analysis (ICA) studies of neonatal resting-state networks have detected the presence of separate thalamic and PMC networks without evidence of definitive functional connections between them.

As a major subcortical structure, the thalamus constitutes a critical integrator of neural connections, with dendritic arborization occurring earlier within the thalamus compared with cortex (Erecinska et al., 2004). This theory of early thalamic development is also supported by functional imaging evidence of robust thalamic metabolism during infancy (Kinnala et

al., 1996; Chugani, 1999). Understanding the maturation of the PMC with closely linked structures requires a comprehensive map of physiological, neuroanatomic, and metabolic changes across development. Development of these connections requires the generation of white matter and cortical pathways dependent on and reflective of metabolic maturation. In fact, the posterior cortical elements of the DMN were first ascertained on the basis of metabolic PET imaging and were, therefore, based at least partially on glucose metabolism differences (Raichle et al., 2001). Despite their potential roles in early brain network development, very little is known about the normal metabolic development of the PMC and the thalamus.

Magnetic resonance spectroscopy (MRS) offers valuable information regarding the metabolic basis of these neuroanatomical structures. Another distinct benefit of metabolic evaluation of development is the robustness of this technique to detect developmental changes in metabolites over time without being prone to activity-related variations, because temporal deviations in metabolism are very small (Hyder et al., 2013). Quantitative assessment of metabolite changes within the developing brain extends the understanding of normal brain development beyond morphological or connectional patterns, and this method is capable of investigating neuronal or axonal density and biochemical composition noninvasively.

Studies of neural metabolism in the developing brain demonstrate strikingly different metabolite concentrations between the neonatal and the adult brain. In the preterm brain, N-acetylaspartate (NAA; indicative of neuronal–axonal density) is particularly low in concentration and subsequently rapidly increases throughout much of brain development, as reviewed at length elsewhere (Vigneron, 2006). With development, certain metabolites including myoinositol (mI) and choline (Cho) decrease from infancy to adulthood in normal individuals (Panigrahy et al., 2010; Bluml et al., 2012). In contrast, NAA is known to increase during this same interval (Bluml et al., 2012). Although most brain regions demonstrate common metabolite trends, some regional differences have been noted even within similar brain structures, including earlier NAA increases within the parietal vs. frontal white matter (Xu et al., 2011). However, little is known regarding relative metabolite changes within the PMC as part of the posterior DMN in relation to other brain structures during early development.

In this study, 3-T proton MRS was employed with voxels located within the PMC gray matter, posterior thalamus, and parietal white matter to determine both similarities and differences in the development of selected metabolites related to structure (i.e. axonal–neuronal integrity, astroglia, and premyelination elements), neurotransmission (glutamate), and energy (glucose and phosphocreatine) among these structures, as summarized in Figure 1. We propose that the PMC acts as a site of neural maturation early in brain development as a key component of the posterior DMN and that development of the PMC would occur in close association with white matter tract metabolic development. It is expected that structural metabolites would change similarly, indicative of common structural development, but that the PMC might demonstrate a distinct metabolite profile relative to the posterior thalamus and parietal white matter reflective of developmental differences in biochemical or energy properties.

MATERIALS AND METHODS

Research participants

Neonates and children in this study were selected from a database of patients aged from birth to 18 years who underwent combined research and clinical magnetic resonance imaging (MRI) and MRS studies over a 4-year period (2008–2012) at Children's Hospital of Pittsburgh of UPMC. Pediatric and adult subjects were recruited as part of a healthy control study and had no pertinent medical history. The neonates were prospectively recruited from both a normal newborn nursery and clinical services if a neonatal scan was performed for minor indication (nonneurological). An experienced, board-certified pediatric neuroradiologist (AP) reviewed all studies. Only participants with unremarkable medical histories (without any neurological disorders or developmental delay) were included and enrolled in longitudinal developmental cognitive studies. The University of Pittsburgh Internal Review Board approved this study.

In total, there were 31 neonates (18 male, 13 female) who met inclusion criteria for this study, with a mean postconceptional age (gestational age + age on date of scan) of 41.5 weeks (SD 3.8 weeks). In the pediatric control group, there were 23 subjects (14 female, nine male) in total with a mean age of 9.4 years (SD 5.1 years) at the time of scan. In the young adult control group, there were 10 subjects (eight female, two male) in total with a mean age of 24.1 years (SD 2.6 years) at the time of scan. Representative spectra from the infant and older age groups are depicted in Figure 2 for each of the voxels analyzed.

MRS acquisition

Single-voxel ¹H spectra from a 3-T GE Signa HDxt scanner (GE Healthcare, Milwaukee, WI) were acquired using point-resolved spectroscopy with a short echo time of 35 msec, a repetition time of 1.5 seconds, 128–256 signal averages, and a total acquisition time for each spectrum of approximately 5–7 minutes, including scanner adjustments. Gray matter and white matter regions of interest (ROIs) increased typically from 2 cm³ in newborns to 5 cm³ in adults to reflect the increase in brain size with increasing age. An eight-channel dedicated pediatric head coil was used for acquisition for all subjects.

Selective spectra were acquired in three ROIs, left parietal white (p-WM) matter, parietal cortical gray matter (p-GM), and left posterior thalamus. Parietal voxels were chosen because others previously showed that parietal WM had higher NAA concentration than frontal regions, implying that the parietal regions associated with sensory function mature earlier (Xu et al., 2011). The left thalamus was chosen because previous work suggests stronger connectivity between the left thalamus and the cortex (Alkonyi et al., 2011). Biochemical profiles of three distinct brain regions (Fig. 2) were analyzed in detail.

1. An ROI contained parietal white matter, dorsolateral to the trigone of the lateral ventricle. Generally included within this ROI are the longitudinal association tracks including the superior longitudinal fasciculus (anterior–posterior), cingulum (anterior–posterior; medially), and centrum semiovale and also short U-fibers and white matter extending into parietal gyri. This ROI would be expected to contain long-range fibers connecting the PCC and the MPFC components of the DMN that

would increase with age on the basis of functional connectivity studies (Fair et al., 2008; Thomason et al., 2008).

2. An ROI contained mostly parietal gray matter, in most cases including the precuneus and posterior cingulate and extending inferiorly into retrosplenial cingulate and posteriorly into cuneus (medial occipital lobe; supracalcarine) encompassing the PMC (Brodmann areas 23, 29, 30, 31, and 7m; Parvizi et al., 2006). This region putatively contains the central hub of the DMN (Bonnelle et al., 2012).
3. An ROI was positioned in the posterior one-third of the thalamus, including the lateral posterior nucleus of the thalamus and pulvinar. The thalamus, particularly its posterior portions such as the pulvinar, appears to be highly connected with the PCC from multiple RS-fMRI studies (Greicius et al., 2003; Fransson, 2005; Thomason et al., 2008).

MRS analysis

All MRS processing was performed using fully automated LCModel software (Stephen Provencher Inc., Oakville, Ontario, Canada; LCModel version 6.1–4F). Data quality was ensured by using established criteria (Kreis, 2004), excluding entire spectra with a line width full width at half-maximum (FWHM) exceeding 0.1 ppm. Visual inspection confirmed quality of spectra by examining for the presence of abnormal features such as asymmetric line shapes. Metabolites were left out of analysis if the Cramer-Rao minimum variance bound was larger than 50% (Kreis, 2004). If more than one-quarter of the data set was excluded for any particular metabolite, analysis for that metabolite was not performed; only those metabolites meeting these requirements are included in the analyses described in this article. Water-normalized absolute metabolite concentrations were utilized for analysis and comparison. Other methodological details of the postprocessing of the metabolite concentrations have been previously described (Bluml et al. 2012). In keeping with prior studies, choline (Cho) was analyzed as the aggregate of PCH and GPC.

Statistical analysis

Statistical analysis in SPSS (version 20; IBM Corp.) examined group differences between the neonates and older controls by using a Student's *t*-test for metabolite concentrations. Logarithmic regression analysis of metabolites with age was performed. For all analyses, metabolic changes were considered to be significant when the regression analyses yielded slopes that were significantly different from zero at a level of $P < 0.05$.

RESULTS

Metabolite regression analysis with age

The estimated values for each metabolite by age group are listed in Table 1. Table 2 displays regression analyses identifying several metabolite trends observed with age (from infancy to young adulthood). Only those metabolites that fulfilled quality assurance criteria for statistical analysis are presented, which include Asp, total Cho (GPC and PCH), total creatine (Cr and PCr), Glc, Gln, Glu, GSH, mI, and NAA.

For all three voxels (PMC, posterior thalamus, parietal white matter), it was observed that Glu and NAA increase with age, whereas mI and Cho decrease with age. Within the PMC voxel, total creatine (Cr and PCr) is noted to increase with age, whereas Cho (combined GPC and PCh) decreased with age. Total creatine also increased significantly within the parietal WM voxel although to a lesser degree than in the PMC. Regression analyses were not significant for age-related changes for Asp, Glc, Gln, and Tau in any voxel location in this study.

Correlation analysis of PMC metabolite trends with thalamic and parietal WM changes

Adjusting for age, the partial correlation analyses were performed between metabolites for the PMC voxel compared with values for thalamic and parietal–occipital WM voxels as summarized in Table 3. These analyses demonstrate more significant correlations between PMC and parietal WM voxels for important metabolites including Asp, Cho, Gln, Glu, GSH, mI, and NAA in comparison with the far fewer and less substantial associations between PMC and thalamus metabolites, which were significant only for Cho, mI, and NAA. This suggests greater similarity between PMC and parietal WM metabolite changes with age.

Structural metabolite changes occur similarly in all voxels with age

Overall, developmental metabolite changes occurred in a similar fashion in most voxels for metabolites associated with structural neuronal and axonal development (NAA), neurotransmission (Glu), and myelin maturity (mI), suggesting similar neuronal structural and integrity processes in all three voxel locations examined with age. mI is thought to be a marker of immature myelination and an indicator of glial cells as part of signaling pathways, a precursor for the phosphoinositide second messenger system. mI decreased with age significantly within all voxel groups and closely corresponded among voxel locations on correlational analyses. The visual depiction of these changes with age (Fig. 3) demonstrates consistent mI decreases following the neonatal epoch, suggesting similar maturation of myelin in these structures.

Similar to mI, Cho is abundant in cell membrane and de novo myelin synthesis, and Cho also serves as a precursor for acetylcholine. Phosphocholine (PCh) and glycerophosphocholine (GPC) treated in aggregate by convention significantly decreased with age for all voxel groups. In addition, age-related decreases in Cho were similarly correlated between voxels. Cho changes were similar for the PMC, parietal WM, and thalamus.

NAA is stored within mature neurons and axons and is thought to indicate the presence of adult axons and neurons (Ross and Bluml, 2001). In this study, NAA demonstrated statistically significant increases with age for all three voxels examined (PMC, posterior thalamus, and parietal white matter) as demonstrated in Figure 3.

Glu is the most abundant excitatory neurotransmitter and is essential to brain function. Glu can be converted into glutamine and vice versa within astrocytes, and there exists a balance between glutamatergic and glutaminergic biochemical pathways (Ward et al., 1983). Glu has also been more recently posited to promote neural synchronization (Rodriguez et al., 2013).

In this study, there were statistically significant increases in Glu with age for all voxels (PMC, posterior thalamus, and parietal white matter), although intervoxel correlation between the PMC and thalamus was not statistically significant. Metabolite changes are essentially consistent between the three voxels but are most similar between the PMC and WM, whereas thalamic Glu plateaus earlier in development (Fig. 3). Our study provides a reliable measurement of Glu separate from Gln as demonstrated by a small cross-correlation coefficient. For Glu and Gln, an average cross-correlation of $-0.194 (\pm 0.132)$, maximum 0.012, minimum -0.373 was observed. A negative cross-correlation is expected (i.e., if Glu increases, Gln decreases and vice versa). This cross-correlation is small, supporting an accurate separation between glutamate and glutamine at 3 T compared with 1.5 T. In individual subjects, there is still a risk of overestimation or underestimation of the Glu and Gln concentrations. Nevertheless, our Glu concentrations in the PMC are consistent with previously published values of Glu in gray matter voxels (Pouwels and Frahm, 1998; Schubert et al., 2004).

Energy metabolite changes are divergent between the PMC and other locations

Creatine is a major contributor to the brain's energy supply and is represented by both free Cr and PCr. The PMC voxel was the only location to demonstrate significant age-related increases in total creatine that is suggestive of more significant changes in energy metabolites than in parietal WM and thalamic voxels. The developmental metabolite changes (Fig. 4) show a consistent upward slope of creatine values with age in juxtaposition to the less substantial increase with age observed in thalamic and parietal WM voxels.

Glucose is the predominant energy source for neurons and serves as the basis of FDG-PET examination of neural functional activity (Erecinska et al., 2004). Glc decreases with age in the selected voxels were not statistically significant, but the age-related decrease in Glc approached statistical significance ($P = 0.07$), and the metabolite concentration change within the PMC voxel demonstrated a much greater decrease in Glc suggestive of greater early Glc concentrations within the PMC, as can be seen in Figure 4. Therefore, energy metabolite changes appear more prominent within the PMC and earlier in development.

Other metabolites demonstrated no significant developmental trends

Gln serves as a major precursor for Glu in synapses (Bradford et al., 1978; Daikhin and Yudkoff, 2000). More recently, however, Glu has been implicated in a neuro-protective role and also serves as an energy metabolite (Chen and Herrup, 2012). No significant age-related relationship was demonstrated for Gln in any of the three voxel locations, but there was a statistically significant correlation between Gln in PMC and parietal WM voxels.

Asp is an excitatory neurotransmitter like Glu, but with less defined importance in the brain. It exists in close association with Glu and Gln within neurons and glial cells (Dingledine and McBain, 1999). There were no statistically significant patterns of Asp measurements with age in any of the three voxels but a significant correlation between values in the PMC and parietal WM.

Glutathione (GSH) is an antioxidant and free radical scavenger prominently affected in hypoxic–ischemic injury although with an unclear role in normal development (Janaky et al., 1999). Authors have suggested that GSH potentially modulates neurotransmission of Glu, GABA, and dopamine in the brain (Oja et al., 2000). Although expected to decrease with age, GSH increased in the thalamus with age with statistical significance. There was a correspondence between parietal WM and PMC GSH concentrations despite no statistically significant change with age in the PMC.

Measurement reliability statistics

The Cramer-Rao lower bound (CRLB) mean values (percentage), standard deviation, and ranges for each metabolite within voxel location and age group are detailed in Table 4. Only those metabolites that met inclusion criteria are included in this analysis.

Comparison with previously published metabolite concentrations

Most metabolite concentrations were similar to those previously reported by others despite differences in methodology as summarized in Table 5 (Kreis et al., 1993; Pouwels and Frahm, 1998; Pouwels et al., 1999; Sarchielli et al., 1999; Horska et al., 2002; Schubert et al., 2004; Minati et al., 2010; Tomiyasu et al., 2013).

Estimation of T1 and T2 metabolite concentration correction

Estimates of T1 and T2 correction for metabolite quantification were extrapolated using available T1 saturation and T2 relaxation times from the literature at 3 T where possible, as detailed in Table 6 (Wansapura et al., 1999; Mlynárik et al., 2001; Ethofer et al., 2003; Zaaraoui et al., 2007). A correction value greater than 1.00 indicates possible overestimation of metabolite concentration in the absence of T1 and T2 correction (e.g., a total correction of 1.19 indicates possible overestimation by 19%), whereas a value less than 1.00 indicates underestimation.

DISCUSSION

The PMC is a prominent cortical hub within the posterior DMN, but there is presently limited information regarding the normal metabolic development of the PMC in relation to other structures. This study utilized 3-T MRS in neonates, children, and young adults to identify metabolic patterns of development. MRS analysis of the underpinnings of these neuroanatomical structures complements what is known from the functional imaging literature. We specifically examined voxels including both precuneus and PCC (together, the PMC) with parietal WM as projection fibers concordant with previously described long-range DMN connections (Fig. 1). These findings argue for a common substrate of metabolic development indicated by neuronal integrity and maturation markers among PMC, parietal WM, and thalamus. Thus, commonality of age-related myoinositol and choline decreases with simultaneous increases in NAA and glutamate suggests similar development of neuronal maturation and myelination. On the other hand, the PMC appears to have unique energy metabolism indicated by divergent creatine and glucose changes that do not correspond to the thalamus or parietal white matter. In addition, there was more similarity of development in terms of the overall number of metabolite changes shared between the PMC

and parietal white matter compared with the PMC and the thalamus, which may provide insight into the metabolic basis of some of the known differences in functional and structural topological network properties of the PMC and thalamus.

Overall, these results suggest a unique pattern of development for the PMC independent of the thalamus consistent with the absence of significant associations between these structures on RS-fMRI investigations, which serve as surrogate indicators of functional connectivity. The PMC appears to demonstrate high metabolic activity early in development with elevated glucose concentrations and greater increases in the mature energy metabolite, total creatine. This study also argues for a closer metabolic resemblance of the PMC to parietal WM, which could potentially be explained by the integration of resting state networks with the PMC via parietal white matter tracts.

Metabolic information supports the posteromedial cortex as an early DMN component

Metabolic information provides intrinsic characteristics of neural development that may not be well represented in structural and functional data. Functional data may represent an admixture of brain maturation, performance-related changes, and subject variability that may be greater in children (Fair et al., 2008; Uddin et al., 2010). Prior work by our group (Bluml et al., 2012) demonstrated the rapid transition of neural metabolites within the first 3 months of life, which coincides with the temporal order of posterior DMN activations on PET (Kinnala et al., 1996; Chugani, 1999). We conceptualize the PMC as a predominant hub in neural activity supported by early metabolic maturation similar to that of parietal long-range WM tracts known from pathways in brain networks.

The present study identifies key metabolite differences suggesting that the PMC has a distinct profile with prominence of energy metabolite changes early in development not seen in the thalamus or parietal WM. Therefore, the PMC may be thought of as a more metabolically active and central metabolic hub of early maturation. The posterior cingulate and precuneus together as the PMC have been thought of as integrators of information and the hub of self-referential thought since Raichle first described the DMN (Raichle et al., 2001). Subsequent studies identify the precuneus and PCC as the largest component of the DMN and likely earliest component to emerge as indicated by high centrality that is present even during infancy, with later centrality of frontal regions in childhood (Thomason et al., 2008; Gao et al., 2009). Elaboration of the PMC within the DMN earlier in development is not surprising in light of its central role in mature DMN on functional network analysis (Hagmann et al., 2008) and the general sequence of brain development, in which prefrontal cortex develops much later (Chu-Shore et al., 2011).

In addition, more recent work examining functional connectivity density indicates that posterior regions, especially the posterior cingulate cortex and precuneus, demonstrate the greatest density of connections, implying their significance in integrating brain networks (Tomasi and Volkow, 2011). This particular segment of the DMN along with other posterior components has been implicated in autobiographical thought, episodic memory retrieval, spontaneous cognition, and self-projection (Buckner et al., 2008; Fair et al., 2008). This study contributes to understanding the early metabolic changes within this vital structure of the DMN (visually depicted in Fig. 1), reinforcing our conceptualization of the PMC as a

metabolically robust region early in development as indicated by the prominent glucose concentrations in infancy and progressive increase in the energy metabolite total creatine across infancy into young adulthood.

Glutamatergic metabolism may provide a metabolic backbone for PMC–white matter pathways

Our study provides a reliable measurement of Glu separate from Gln with glutamate concentrations in the PMC consistent with previously published values for gray matter voxels (Pouwels and Frahm, 1998; Schubert et al., 2004). The basis for the early development of neural networks involving the PMC may first originate with greater metabolic activity that is distinct with unique energy metabolism with a key role of Glu, but subsequent elaboration of the PMC as a cortical hub likely is related to extension of cortical connections via white matter fiber tracts. Our study sought to identify similarity between the PMC and parietal WM as the likely site of such integrating white matter tracts as part of known resting state networks. In this study, the PMC bore closest similarity in terms of metabolic changes with the parietal white matter, as expected. Metabolic developmental patterns statistically significantly covaried for Asp, Cho, Gln, Glu, GSH, mI, and NAA between the PMC and parietal WM. On the other hand, the thalamus had significant covariance only for Cho, MI, and NAA; these metabolites are thought of as a common neuronal structure shared by all structures.

The similarities of Glu and Gln between parietal WM and PMC are particularly salient in conceptualizing these structures as integrated components in resting state networks. As the major excitatory neurotransmitter essential to brain function, Glu facilitates neuronal synchronization according to recent research, expanding the functions assigned to Glu (Rodriguez et al., 2013). Using J-resolved MRS and RS-fMRI, Kapogiannis et al. (2013) identified a positive correlation between Glu within the PMC and DMN functional connectivity, arguing for a key role of Glu in neural networks. Evidence from a combined DTI and RS-fMRI study looking at PCC and hippocampal connectivity demonstrated close association between anatomical and functional connectivity via the connecting WM tracts (Teipel et al., 2010). This finding argues for the importance of a metabolic convergence between connected cortical and subcortical structures via WM. Glu similarities also are important in that Gln can be thought of as a neuroprotective agent required for healthy neurons, and its increase across age within the PMC and parietal white matter in normal development may reflect the development of a normal neurometabolite homeostasis as part of the glutamate–glutamine cycle and may represent the supportive neural substrate of this vital functional network (Daikhin and Yudkoff, 2000; Chen and Herrup, 2012).

These findings suggest an integrated development of PMC neurons with extension of integrated WM tracts with age supported by common development of neuronal structural, neuroprotective, and synchronizing neurometabolites suggested by the greater similarity between these structures not shared by the thalamus. The glutamatergic–glutaminergic pathways strongly argue for formation of synchronized neural networks in these early resting-state networks along the neuronal–axonal pathways incorporating the PMC with cortical structures via the parietal WM.

Comparative anatomical connections between PMC and thalamus are divergent from metabolic and functional interactions

Comparative neuroanatomical studies in primates have established the PMC as an anatomically rich hub of neural connections (Vogt et al., 1979). Early studies in the monkey evidenced afferents from the anterior thalamus to the PMC (Vogt et al., 1979), and subsequent investigation demonstrated that the PMC possesses connections to the posterior thalamus in the macaque (Baleydier and Mauguier, 1987; Tzourio-Mazoyer et al., 2002). A histopathological tracer examination in the macaque observed migration from different regions of the PMC to distinct portions of the thalamus (Buckwalter et al., 2008). They described connections between PCC and retrosplenium with anterior thalamic nuclei, medial parietal cortex with lateral posterior thalamus and pulvinar, posterior cingulate with ventral thalamus (Buckwalter et al., 2008). Therefore, the PMC contains a complex cytoarchitectural relationship between different thalamic nuclei that defies the common treatment of these grouped structures as a common functional unit (Parvizi et al., 2006).

However, despite these anatomical connections with the thalamus, there is little evidence to support functional connectivity of the thalamus to the posterior DMN in early development in humans. Notably, very little comparative neuroanatomical information exists on the PMC in relation to early fetal and neonatal primate or nonprimate development, except for examinations of the role of parietal lesions in early development in rats (Alexinsky, 2001). The functional MRI literature has yet to identify prominent networks involving the thalamus with the PMC despite a wealth of literature reporting the PMC as a major cortical hub within the DMN. For this reason, we sought to examine whether the divergent information provided by the RS-fMRI literature regarding the interactions between the thalamus and the PMC might be explained by discordant metabolic substrate between these anatomically associated areas.

The distinct metabolic pattern that we observe within the PMC compared with the thalamus may reflect the divergent roles of these structures. The PMC is a prominent portion of the posterior DMN, whereas the role of the thalamus is unclear in resting-state networks with only homotopic interactions (i.e., those between matching structures in right and left cerebral hemispheres) demonstrated in an RS-fMRI study of human infants (Smyser et al., 2010). Prominence of energy metabolites such as creatine may confer greater metabolic activity within the PMC as a major hub of resting-state networks. In addition, the PMC demonstrated continued increases in Glu, as displayed in Figure 3. Glu has been thought to play a role in neural synchronization in recent literature (Rodriguez et al., 2013), so it may be that glutamatergic activity with similar developmental changes in the parietal WM and PMC is the metabolic driver of the synchronization of posterior DMN components. This finding is not shared by the thalamus, in which Glu plateaus by young adulthood and age-related changes are not statistically similar to PMC and parietal WM.

Another possible explanation for the lack of functional connectivity and metabolic covariance of the thalamus and the PMC may be the diverse cytoarchitecture of both the PMC and the thalamus. The distinct anatomical relationships delineated in the macaque imply that humans would also have complex differentiation of connections among different components of the PMC with distinct thalamic nuclei. The spatial sensitivity of the present

RS-fMRI and MRS techniques cannot accurately evaluate these relationships. Thus, the lack of functional and anatomical covariance and now, in the present study, absent metabolic covariance may equally be explained by technical limitations or an incoherent relationship between the thalamus and PMC.

General metabolic trends observed are consistent with normal maturation

This study replicates some general metabolic trends noted in previous studies while additionally illustrating these trends within a posterior thalamic voxel. The finding of increases in Glu and NAA and decreased mI and Cho over the developmental spectrum is unsurprising; others have demonstrated similar trends in other voxels (Girard et al., 2006; Bluml et al., 2012). The increases in NAA are thought to be related to increased neuronal function and progressive myelination; these increasing levels of NAA occur even prior to term infancy as evidenced by fetal MRS (Heerschap et al., 2003). Previous work suggests that NAA peaks during young adulthood (between 20 and 30 years of age) and subsequently declines with aging (Brooks et al., 2001; Kadota et al., 2001); our trends in NAA from infancy until young adulthood are in keeping with these observations.

Similarly, the observed decreases in mI over normal development agree with concepts of brain development. mI in particular reflects astrocyte or glial cell density and is altered in conditions affecting osmolality (Pugash et al., 2009); it also is present within immature myelin and subsequently declines with mature myelination. Thus, the normal relative decrease in glial cells and loss of immature myelin after the neonatal period would account for concomitant reductions in mI (Girard et al., 2006; Vigneron, 2006). Cho decreases similarly are thought to correspond to maturation of myelination and loss of immature myelin, because Cho is present in de novo myelin synthesis and would therefore be expected early in development, but not in older populations.

Limitations

As with many studies conducted with MRS, our results are limited by the nature of a cross-sectional study with modest participant numbers, particularly with few older controls for comparison. Others have alluded to the significance of covering different epochs of development throughout childhood. In light of limitations of pediatric MRI study compliance among certain ages of participants, these data do not cover substantial numbers of children during middle childhood, which many would argue to be an important time based on developmental milestones (Thomason et al., 2008). Further work must be undertaken with 3-T MRS in a larger group of healthy individuals to strengthen the validity of these interesting results, particularly inasmuch as 3-T MRS may improve separation of signal maxima for metabolite differentiation (Gussew et al., 2008). Longitudinal analyses may offer additional insight into brain development, and MRS is well-suited for longitudinal studies on the individual level by consistently tracking metabolite changes with age (Currie et al., 2013). Moreover, future work should integrate preterm infant and fetal data sets to extend the developmental spectrum sampled in our study, especially because most rapid changes occur during prematurity and the first few months of term infancy (Vigneron, 2006; Bluml et al., 2012). Our failure to detect certain developmental changes reported from

studies including preterm infants may reflect changes that occur at earlier postconceptional ages not included in our study (Volpe, 2009).

Technical considerations

The metabolite concentrations measured in this study represent estimates of metabolite concentration and were not corrected for T2 relaxation, T1 saturation, or partial volume effects of voxels including both white matter and gray matter within a given region. Although these corrections were not performed, the overall measurements and relative concentrations are similar to those previously reported for multiple metabolites (Table 5; Kreis et al., 1993; Pouwels and Frahm, 1998; Pouwels et al., 1999; Horska et al., 2002; Kreis et al., 2002; Schubert et al., 2004; Minati et al., 2010; Tomiyasu et al., 2013). Predominantly because of T2 relaxation effects, the metabolite concentrations may be overestimated by up to approximately 25% for certain metabolites (e.g., 25% and 26% for NAA and total Cho, respectively, in parietal white matter) as estimated from previously published saturation and relaxation values (Table 6). For other metabolites and locations, estimation of combined T1 and T2 effects is not substantial (e.g., no combined effect for total Cr and 7% overestimation for NAA in the thalamus). In addition, interindividual variability in metabolite concentrations may exist. However, all subjects were healthy, so the interindividual variability is dominated by the limitations of the MRS methodology and assumed to be similar among all age groups.

Another key limitation (although a simultaneous strength) of this study relates to the rigorous quality assurance approach chosen. Exclusion based on the Cramer-Rao number is commonplace within the MRS literature, and we chose to adhere to these technical requirements for reproducibility, which may limit the number of metabolites analyzed (Kreis, 2004). Not all metabolites could be analyzed. Spectral editing techniques for GABA were not available for this study. Inclusion of approaches that utilize two-dimensional uncoupling and spectral editing such as J-difference editing point-resolved MRS may be of benefit in future studies (Levy and Degnan, 2013). Like Glu, GABA has been implicated in primate studies as a key component of subplate neurons thought to be of great importance in thalamocortical connectivity (Kostovic and Judas, 2010) and might be a valuable target for further investigation in developmental analyses.

As is true of quantitative MRS studies, metabolite concentration determination relies on the fitting of curves to metabolite spectra. Inherent in these analyses are measurement uncertainties and intersubject variation dependent on the quality of individual spectra. Nevertheless, our application of rigorous data analysis criteria to this data set ensures that these findings are reported in a replicable fashion (Kreis, 2004). Still to be addressed is the fundamental reproducibility of MRS and the biological underpinnings of the metabolite measurements reported (Currie et al., 2013). Several technical aspects and interpretative differences preclude correlations of concentrations between different sites and are further complicated by unique aspects of analysis criteria employed. Investment in additional research should seek to unify the acquisition of robust, reproducible MRS spectra. Use of three-dimensional short TE metabolic spectroscopic imaging may offer additional advantages of comparing metabolites across the whole brain, rather than within selected

regions as in single voxel MRS performed in this study, and could thus allow for additional insights into anatomic regions not previously considered. The application of these advances in MRS will greatly add to the understanding of the metabolic correlates of neural development.

CONCLUSIONS

In concordance with identified anatomic connections from prior comparative neuroanatomical studies between the PMC and the thalamus, these structures demonstrated similar development of structural metabolites (i.e., neuronal–axonal). However, the PMC and thalamus demonstrated divergent metabolite profiles, particularly with differences in energy metabolism, that may explain the prior lack of consistent functional covariance on resting-state network analyses. Similarity of the developmental changes of multiple metabolites between the PMC and the parietal WM, compared with between the PMC and the thalamus, may provide insight into the possible metabolic basis of functional and structural topology of PMC vs. thalamic networks. Further investigation integrating metabolic, functional, and anatomic information regarding the PMC in relation to the closely linked posterior DMN may provide further clarification of these findings.

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Figure 1.

Graphic representation of MRS voxel location to default mode network components with representative spect. **A:** Left: A MNI template was used to generate an anatomic mask within a cerebral hemisphere (viewed in an oblique angle) based on automated anatomical labeling (Tzourio-Mazoyer, et al. 2002) for both of the PMC structures, including the precuneus (blue) and the posterior cingulate (red), and the thalamus (green). Right: FACT-based tractography was used to generate a comparable image (oblique) of the intersection (crossing fiber region) of the relevant white matter fiber bundles that transverse the parietal white matter voxel including the cingulum (connecting anterior and posterior default mode networks), the splenium of the corpus callosum (interhemispheric connectivity), and the posterior thalamic radiations (thalamocortical connectivity). White boxes indicate the approximate location of MRS voxels within the PMC, thalamus, and parietal white matter (n.b., parietal white matter voxel was performed in the parasagittal plane, whereas on this diagram the voxel is displayed in the midsagittal image for simplicity). Representative adult spectra are included below for the PMC (**B**), thalamus (**C**), and parietal white matter (**D**).

Figure 2. Representative voxel locations and MR spectra. Representative magnetic resonance spectra and region-of-interest/voxel placement displayed on structural isotropic 3D T2-weighted neonatal MRI of left posterior thalamus (A), parietal white matter (B), and parietal cortical gray matter (C) in neonate (left) and adolescent (right).

Figure 3.

Similarities of structural metabolite changes. Error plots depict mean metabolite concentrations for NAA (A), Cho (B), Glu (C), and mI (D) within the PMC, thalamus, and parietal WM for each age group. mI and Cho demonstrate similar patterns of reduction with age in all voxels (PMC, thalamus, and parietal WM). These metabolite reductions are thought to reflect global maturation of myelin, with decreases in de novo myelin synthesis (Cho) and reduction in mI. Both NAA and Glu increase similarly, and these findings likely represent increased neuronal density and maturation. Error bars represent the standard error of the mean for metabolite concentrations.

Figure 4.

Divergent energy metabolite changes. Error plots depict mean metabolite concentrations for total creatine (A) and glucose (B) within the PMC, thalamus, and parietal WM for each age group. The energy metabolite total creatine increases consistently with age only within the PMC, whereas the thalamus and parietal WM voxels demonstrate concentration plateau with age. Glucose demonstrates a sharp decline from its greater initial concentration in the PMC compared with other voxels, suggesting a disparate trend with the PMC demonstrating greater immature energy metabolite usage earlier in development. Error bars represent the standard error of the mean for metabolite concentrations.

TABLE 1
Metabolite Values by Voxel and Age

Metabolite	Voxel location	Neonate (mmol $\pm$ SD)	Pediatric (mmol $\pm$ SD)	Adult (mmol $\pm$ SD)
Asp	Thalamus	4.53 $\pm$ 1.80	4.44 $\pm$ 1.94	5.18 $\pm$ 2.85
	PMC GM	6.41 $\pm$ 2.79	4.43 $\pm$ 2.09	6.89 $\pm$ 2.28
	Parietal WM	5.39 $\pm$ 2.65	3.78 $\pm$ 0.68	5.59 $\pm$ 2.17
Cho(GPC + PCH)	Thalamus	2.84 $\pm$ 0.47	1.99 $\pm$ 0.36	1.99 $\pm$ 0.14
	PMC GM	2.32 $\pm$ 0.58	1.04 $\pm$ 0.21	1.29 $\pm$ 0.19
	Parietal WM	2.23 $\pm$ 0.55	1.71 $\pm$ 0.19	1.79 $\pm$ 0.19
Total Cr	Thalamus	7.11 $\pm$ 0.93	7.04 $\pm$ 1.63	7.46 $\pm$ 0.90
	PMC GM	5.48 $\pm$ 0.85	7.25 $\pm$ 1.08	7.69 $\pm$ 1.08
	Parietal WM	4.62 $\pm$ 1.10	5.20 $\pm$ 0.58	5.72 $\pm$ 0.99
Glc	Thalamus	3.15 $\pm$ 1.32	1.92 $\pm$ 1.45	2.27 $\pm$ 1.63
	PMC GM	4.69 $\pm$ 4.59	1.29 $\pm$ 0.62	1.62 $\pm$ 0.51
	Parietal WM	3.50 $\pm$ 1.43	2.23 $\pm$ 1.28	2.18 $\pm$ 1.30
Gln	Thalamus	2.47 $\pm$ 1.01	2.53 $\pm$ 0.91	2.99 $\pm$ 1.57
	PMC GM	2.57 $\pm$ 1.20	2.10 $\pm$ 0.92	2.61 $\pm$ 1.51
	Parietal WM	2.70 $\pm$ 1.00	2.12 $\pm$ 1.43	-
Glu	Thalamus	6.42 $\pm$ 1.50	9.62 $\pm$ 2.63	9.88 $\pm$ 2.08
	PMC GM	6.80 $\pm$ 2.21	11.58 $\pm$ 2.53	11.93 $\pm$ 2.44
	Parietal WM	5.84 $\pm$ 2.39	8.28 $\pm$ 1.93	9.46 $\pm$ 2.57
GSH	Thalamus	2.59 $\pm$ 0.74	2.52 $\pm$ 0.62	3.18 $\pm$ 1.14
	PMC GM	3.16 $\pm$ 1.17	2.58 $\pm$ 0.71	2.65 $\pm$ 0.64
	Parietal WM	2.79 $\pm$ 1.13	2.04 $\pm$ 0.35	2.84 $\pm$ 1.29
ml	Thalamus	6.92 $\pm$ 1.43	3.78 $\pm$ 0.96	4.28 $\pm$ 1.09
	PMC GM	9.12 $\pm$ 3.68	4.66 $\pm$ 0.50	5.27 $\pm$ 0.63
	Parietal WM	7.26 $\pm$ 1.84	4.04 $\pm$ 0.45	4.29 $\pm$ 0.67
NAA	Thalamus	5.99 $\pm$ 1.19	10.10 $\pm$ 1.00	9.82 $\pm$ 1.48
	PMC GM	4.00 $\pm$ 1.46	10.05 $\pm$ 1.37	11.24 $\pm$ 1.94
	Parietal WM	3.95 $\pm$ 1.38	9.49 $\pm$ 1.33	10.00 $\pm$ 1.23

TABLE 2
Metabolite Regression Analysis With Age

Metabolite	Voxel location	R value	Significance (<i>P</i>)
Asp	Thalamus	0.257	0.059
	PMC GM	−0.004	0.982
	Parietal WM	−0.043	0.811
Cho(GPC + PCH)	Thalamus	−0.581	<0.001
	PMC GM	−0.602	<0.001
	Parietal WM	−0.205	0.002
Total Cr	Thalamus	0.001	0.818
	PMC GM	0.570	<0.001
	Parietal WM	0.157	0.006
Glc	Thalamus	−0.248	0.096
	PMC GM	−0.435	0.071
	Parietal WM	−0.324	0.075
Gln	Thalamus	0.222	0.126
	PMC GM	0.071	0.680
	Parietal WM	0.236	0.289
Glu	Thalamus	0.579	<0.001
	PMC GM	0.674	<0.001
	Parietal WM	0.528	<0.001
GSH	Thalamus	0.343	0.004
	PMC GM	−0.226	0.151
	Parietal WM	−0.073	0.635
ml	Thalamus	−0.552	<0.001
	PMC GM	−0.499	0.001
	Parietal WM	−0.607	<0.001
NAA	Thalamus	0.684	<0.001
	PMC GM	0.852	<0.001
	Parietal WM	0.846	<0.001

TABLE 3
Correlations of PMC Metabolites With Thalamic and WM Voxels

Metabolite	Thalamic correlation	Significance (P)	Parietal WM correlation	Significance (P)
Asp	0.092	0.708	0.559	0.013
Cho (GPC + PCH)	0.453	0.004	0.724	<0.001
Total Cr (Cr + PCr)	-0.130	0.470	0.107	0.554
Glc	0.241	0.306	0.194	0.413
Gln	0.209	0.514	0.661	0.019
Glu	0.259	0.116	0.328	0.044
GSH	0.051	0.767	0.491	0.002
ml	0.493	0.001	0.515	0.001
NAA	0.477	0.002	0.823	<0.001

TABLE 4
Cramer-Rao Lower Bound (CRLB) for the Metabolites Included in This Analysis¹

Metabolite	Voxel location	Neonate (% CRLB $\pm$ SD, range)	Pediatric (% CRLB $\pm$ SD, range)	Adult (% CRLB $\pm$ SD, range)
Asp	Thalamus	28.1 $\pm$ 7.4, 18–40	34.1 $\pm$ 10.8, 17–50	32.3 $\pm$ 10.7, 19–50
	PMC GM	22.4 $\pm$ 6.8, 12–39	22.6 $\pm$ 10.1, 13–39	19.8 $\pm$ 5.5, 14–30
	Parietal WM	33.2 $\pm$ 9.4, 13–50	34.1 $\pm$ 9.2, 24–50	29.7 $\pm$ 6.7, 19–36
Cho (GPC + PCH)	Thalamus	3.2 $\pm$ 0.7, 2–6	4.0 $\pm$ 1.0, 3–6	4.7 $\pm$ 1.4, 3–8
	PMC GM	5.2 $\pm$ 3.4, 3–19	5.0 $\pm$ 1.6, 3–8	4.9 $\pm$ 1.2, 3–7
	Parietal WM	4.4 $\pm$ 0.8, 3–7	4.3 $\pm$ 0.8, 3–5	5.4 $\pm$ 1.9, 3–8
Total Cr	Thalamus	3.4 $\pm$ 0.8, 2–6	3.4 $\pm$ 0.8, 3–6	3.9 $\pm$ 1.4, 2–7
	PMC GM	5.3 $\pm$ 3.1, 2–16	2.4 $\pm$ 0.8, 2–4	2.5 $\pm$ 0.8, 2–4
	Parietal WM	5.1 $\pm$ 1.5, 3–8	3.9 $\pm$ 0.9, 3–5	4.9 $\pm$ 2.1, 3–9
Glc	Thalamus	24.5 $\pm$ 8.9, 9–50	37.5 $\pm$ 10.1, 18–50	24.0 $\pm$ 8.0, 16–32
	PMC GM	20.3 $\pm$ 6.7, 6–34	32.0 $\pm$ 7.0, 27–37	36.3 $\pm$ 7.2, 27–44
	Parietal WM	23.7 $\pm$ 8.2, 7–46	39.3 $\pm$ 13.9, 24–50	37.3 $\pm$ 13.3, 26–50
Gln	Thalamus	26.9 $\pm$ 8.4, 10–41	28.6 $\pm$ 7.3, 19–42	32.4 $\pm$ 9.0, 21–43
	PMC GM	27.9 $\pm$ 7.3, 18–38	25.0 $\pm$ 5.8, 18–32	29.8 $\pm$ 3.5, 26–34
	Parietal WM	31.8 $\pm$ 10.2, 13–50	31.5 $\pm$ 17.7, 19–44	31.8 $\pm$ 10.5, 10–50
Glu	Thalamus	13.5 $\pm$ 3.2, 9–25	10.2 $\pm$ 3.3, 7–20	11.1 $\pm$ 3.5, 8–21
	PMC GM	14.7 $\pm$ 5.1, 8–31	7.1 $\pm$ 1.5, 5–9	7.8 $\pm$ 1.9, 6–10
	Parietal WM	17.3 $\pm$ 6.9, 9–42	10.1 $\pm$ 0.9, 9–11	11.8 $\pm$ 3.1, 8–16
GSH	Thalamus	14.6 $\pm$ 3.2, 10–23	16.8 $\pm$ 7.3, 10–47	15.6 $\pm$ 5.2, 10–28
	PMC GM	15.9 $\pm$ 8.3, 9–50	11.6 $\pm$ 2.4, 9–16	13.8 $\pm$ 3.5, 9–19
	Parietal WM	16.9 $\pm$ 6.3, 10–35	21.1 $\pm$ 11.9, 13–47	16.0 $\pm$ 2.5, 13–20
mI	Thalamus	6.7 $\pm$ 1.7, 5–14	10.2 $\pm$ 3.0, 7–18	12.2 $\pm$ 5.9, 6–30
	PMC GM	6.5 $\pm$ 2.6, 4–14	6.3 $\pm$ 1.0, 5–8	6.9 $\pm$ 1.7, 5–10
	Parietal WM	7.1 $\pm$ 1.8, 4–12	9.1 $\pm$ 2.5, 7–14	10.4 $\pm$ 3.9, 6–17
NAA	Thalamus	4.5 $\pm$ 0.6, 4–6	3.8 $\pm$ 0.9, 3–6	4.4 $\pm$ 1.2, 3–7
	PMC GM	7.6 $\pm$ 3.6, 3–22	3.3 $\pm$ 1.0, 2–5	3.3 $\pm$ 0.9, 2–4
	Parietal WM	7.0 $\pm$ 2.0, 4–12	3.7 $\pm$ 1.1, 3–6	4.5 $\pm$ 1.4, 3–7

¹The CRLBs are objective measurements of reliability of individual metabolite quantitation provided by LCModel analysis.

TABLE 5
Comparison of Metabolite Concentrations With Previously Published Literature¹

	Concentration (mmol/liter) in this study (range of means across age groups)	Previously published concentration values (mmol/liter)
Glutamate		
Parietal/occipital ² GM	6.8–11.9	7.1–12.5
Parietal/occipital WM	5.8–9.5	6.7
Thalamus	6.4–9.9	6.9
NAA		
Parietal/occipital GM	4.0–11.2	7.9–14.1
Parietal/occipital WM	4.0–10.0	3.8–14.0
Thalamus	6.0–10.1	6.9–16.3
Choline		
Parietal/occipital GM	1.0–2.3	1.3–3.6
Parietal/occipital WM	1.7–2.2	1.7–2.9
Thalamus	2.0–2.8	1.9–3.4
Creatine		
Parietal/occipital GM	5.5–7.7	6.8–12.0
Parietal/occipital WM	4.6–5.7	4.3–10.7
Thalamus	7.0–7.5	5.7–12.0
Myoinositol		
Parietal/occipital GM	4.7–9.1	6.2–7.9
Parietal/occipital WM	4.0–7.3	5.8–7.0
Thalamus	3.8–6.9	6.6

¹ Representative previously published concentrations vary in terms of age group examined (range 0–60 years), precise MRS voxel site, and technique but provide an approximation of normative values for these metabolites in the vicinity of the PMC, parietal white matter, and thalamus.

² For comparison, PMC and parietal white matter metabolite concentrations in this study were compared with parietal and occipital gray matter and white matter concentrations because of a lack of availability of comparable voxel locations in previously published literature.

TABLE 6
Estimates of T1 and T2 Correction Based on Literature Values

		T1 (s)	T1 saturation	T2 (msec)	T2 relaxation	T1 water (sec)	T1 saturation water	T2 water (msec)	T2 relaxation water	T1 correction	T2 correction	Total correction
Parietal/occipital ¹ grey matter	NAA	1.47 ± 0.08	0.74	247 ± 19	0.87	1.47 ± 0.05	0.74	112 ± 4	0.73	1.00	1.19	1.19
	Cho	1.25 ± 0.22	0.80	207 ± 16	0.84	1.47 ± 0.05	0.74	112 ± 4	0.73	1.07	1.15	1.24
	Cr	1.33 ± 0.13	0.78	152 ± 7	0.79	1.47 ± 0.05	0.74	112 ± 4	0.73	1.05	1.09	1.14
	ml	1.12 ± 0.25	0.83	–	–	1.47 ± 0.05	0.74	112 ± 4	0.73	1.12	–	–
	Glu	1.27 ± 0.10	0.79	–	–	1.47 ± 0.05	0.74	112 ± 4	0.73	1.07	–	–
Parietal/occipital white matter	NAA	1.40 ± 0.15	0.76	295 ± 29	0.89	1.11 ± 0.04	0.83	80 ± 1	0.65	0.91	1.38	1.25
	Cho	1.17 ± 0.15	0.82	187 ± 20	0.83	1.11 ± 0.04	0.83	80 ± 1	0.65	0.98	1.28	1.26
	Cr	1.31 ± 0.13	0.78	141 ± 16	0.78	1.11 ± 0.04	0.83	80 ± 1	0.65	0.94	1.21	1.13
	ml	0.91 ± 0.13	0.89	–	–	1.11 ± 0.04	0.83	80 ± 1	0.65	1.06	–	–
	Glu	1.17 ± 0.08	0.82	–	–	1.11 ± 0.04	0.83	80 ± 1	0.65	0.98	–	–
Thalamus	NAA	1.57 ± 0.08	0.72	229 ± 15	0.86	1.15 ± 0.08	0.82	98 ± 21	0.70	0.87	1.23	1.07
	Cho	1.38 ± 0.22	0.77	198 ± 14	0.84	1.15 ± 0.08		98 ± 21		0.93	1.20	1.11
	Cr	1.45 ± 0.16	0.75	135 ± 10	0.77	1.15 ± 0.08		98 ± 21		0.91	1.10	1.00
	ml	1.11 ± 0.32	0.83	–	–	1.15 ± 0.08		98 ± 21		1.01	–	–

¹Occipital values were used in lieu of parietal values in many instances because of the limited availability of published data for parietal voxels.