

# Proton magnetic resonance spectroscopic imaging as a cancer biomarker for pediatric brain tumors (Review)

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**Abstract.** Magnetic resonance (MR) techniques offer a non-invasive, non-irradiating yet sensitive approach to diagnose and monitor cancer, which encompasses diverse processes affecting various aspects of pathophysiology. Techniques such as MR spectroscopy (MRS) have been developed and applied to monitor the metabolic aspects of cancer. Given that cancer is such a variable disease, biomarkers identified using MRS represent a promising advance and may suggest appropriate therapy, especially when diagnostic biopsies are not feasible. This article will focus on proton MRS, which appears to be the most promising MR method and is complementary to existing diagnostic methods that may be used to characterize and monitor cancer processes. We further focus on applying the MRS technology to pediatric brain tumors, the leading cause of pediatric cancer mortality.

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## 1. Introduction

The incidence of cancer in children under 15 years of age in the United States has risen in recent years (1). This increase can largely be attributed to the increased incidence of lymphoblastic leukemia and tumors of the nervous system, including the brain, rather than other childhood cancers such as Wilms' tumors, soft tissue and bone sarcomas, lymphomas, and Hodgkin's disease. Between 1973 and 1988, the incidence of nervous system tumors in children jumped by 30% (2). Every year, >1,500 children are diagnosed with brain tumors (3). Given that such cancers generally develop during the first five years of life, their etiology is likely to be different from those seen in later life, and different factors are probably involved. Although childhood tumors are generally more aggressive than those seen in later life, long-term control of such cases is often possible (4). As reported in *Cancer Statistics Review*, mortality from childhood cancer has shown an overall decrease, although deaths associated with brain and nervous system cancers in children have shown a smaller decline than those due to other malignancies (1). Brain cancer is currently the leading cause of death from cancer in children under the age of 15 and the second leading cause of death from cancer from age 15 to 34 (5).

Brain tumor treatment in most modern centers is managed through a tumor board, which typically relies, at least in part, on magnetic resonance spectroscopic imaging (MRSI), especially for inoperable tumors that can be difficult to biopsy. Additionally, in cases where tumor progression or treatment response is in question, serial *in vivo* MRSI is preferred over serial biopsy or positron emission tomography (PET) and single photon emission tomography (SPECT), which are irradiating, expensive and not widely available. Clearly there is a need for *in vivo* biomarkers to allow clinical evaluation of treatment protocols for pediatric brain tumors. Non-invasive and non-irradiating methods, such as MRSI, are certainly preferable to the use of sequential biopsies to monitor therapeutic response in children with brain tumors. In addition, MRSI not only measures tumor volume, but also provides diagnostic indices or biomarkers useful in clinical settings. Furthermore, *in vivo* MRSI can be performed as an adjunct to magnetic resonance imaging (MRI), making it a cost-effective method for use in children under 5 years of age, in whom radiation is a serious concern.

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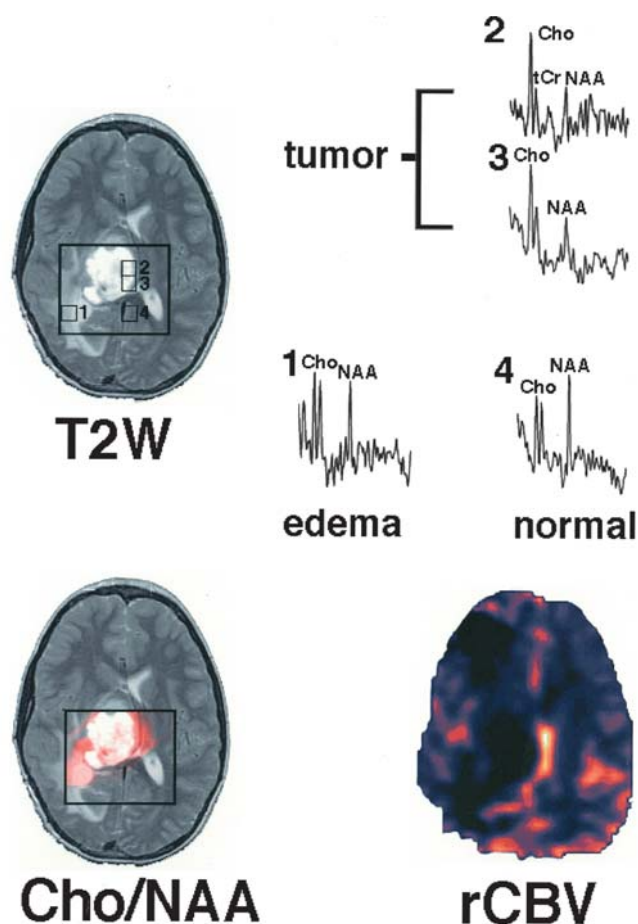


Figure 1. Conventional T2-weighted magnetic resonance image (T2W) and selected MR spectra from the brain of an 8-year-old boy with a large anaplastic astrocytoma of the right thalamus with a collapsed necrotic cystic central component. A metabolite map of the ratio of choline over N-acetylaspartate (Cho/NAA) and a hemodynamic or relative cerebral blood volume (rCBV) image are also shown. The metabolite map was calculated from a multivoxel proton MRS data set and was overlaid on the T2W image to show the distribution of Cho/NAA within the volume of interest (large box on T2W image). The rCBV image shows a necrotic region with low rCBV. The peripheral enhancement indicating an active tumor is consistent with the increased Cho/NAA shown by spectra 2 and 3 and by the distribution of intensities in the Cho/NAA map. Finally, there is evidence of edema around the right temporal horn (T2W image and spectrum 1).

Although MRSI does not obviate the utility of biopsy, it does have the potential of replacing serial biopsies in addition to serving as an excellent alternative to biopsy in inoperable or unbiopsied tumors. When combined with anatomical or other types of imaging, such as MRI, MRSI data provide unique information regarding biochemistry in inoperable brain tumors and may complement neuropathologic data, guide biopsies and inform therapeutic options. The combination of non-invasively acquired prognostic information and the high-resolution anatomical imaging provided by conventional MRI is expected to surpass molecular analysis or DNA microarray gene profiling, both of which, although promising, depend on invasive biopsy.

## 2. Application of brain tumor proton MRS in children

Localized MR spectroscopy (MRS) studies in children with cancer are increasing (6). Brain tumors are usually

heterogeneous, complicated by edema and necrosis of the adjacent parenchyma, and their spectra are critically affected by voxel size and position. Single-voxel MRS has the inherent disadvantage that spectral data are not simultaneously collected from the tumor and its surrounding tissue, meaning that valuable information is lost for assessment of therapeutic response (7). Technical difficulties have limited the number of tumor studies in the adult brain that involve the use of advanced localized MRS (8), and even fewer such studies have been carried out in children (9).

## 3. Biological aspects of selected metabolites detected by proton MRS

Proton MRS of the brain has identified several metabolites that are biomarkers of tumor growth and apoptosis (10). As shown in Fig. 1, these studies consistently demonstrate reduced or absent n-acetylaspartate (NAA) and total creatine (tCr) attributed to edema and necrosis; increased choline (Cho)-containing compounds, possibly due to cell membrane disruption (11) and altered phospholipid metabolism (12); and increased lactate due to metabolic acidosis (13). Low NAA levels are expected in glial tumors because it is primarily localized in neurons. Therefore, NAA detection within glial tumors corresponds to either partial volume averaging with adjacent normal tissue or tumor infiltration of normal tissue. Because NAA occurs in cell cultures of oligodendroglia progenitors (14), NAA in childhood tumors may also reflect immature oligodendroglia. A reduction in tCr resonance may indicate cell loss due to necrosis, and corresponds to exhausted energy reserves resulting from rapid cell proliferation and ischemia. It is also possible that tCr may be a valuable independent predictor of tumor response to therapy (15).

The Cho peak represents water-soluble Cho-containing compounds, such as phosphocholine (PCho) and glycerophosphocholine (GPC), and free choline (not membrane-bound phosphatidylcholine) (16). *In vivo* MRS reveals that phosphomonoesters (PMEs), such as PCho and phosphoethanolamine (PEth), are elevated in tumors and rapidly proliferating tissues (17). Furthermore, elevations in PCho and PEth correlate with increased cell growth or increased cell degradation, and have been shown to occur in human tumors as well as in animal tumor models and cell lines. For example, actively proliferating cultures show a dramatically lower PCho/PEth ratio compared with stationary cultures (18). Mahmood *et al* found a strong radiation dose-dependent response in the relative PCho/PEth ratio, suggesting that changes in the PCho levels may be related to cell proliferation and/or radiation-induced membrane damage (19). Indeed, *in vivo* proton MRS studies suggest the Cho peak reflects proliferative activity in gliomas (20,21), and the PCho/GPC ratio corresponds to oncogenic transformation (22). Together, these data suggest that PCho, which can be measured with either phosphorous or proton MRS, is elevated in actively proliferating cells.

The PCho-produced Cho signal has been proposed to also depend on local cellularity (16). Recently, using a high-resolution magic angle spinning proton MRS technique, PCho levels in glioblastoma multiforme were shown to correlate

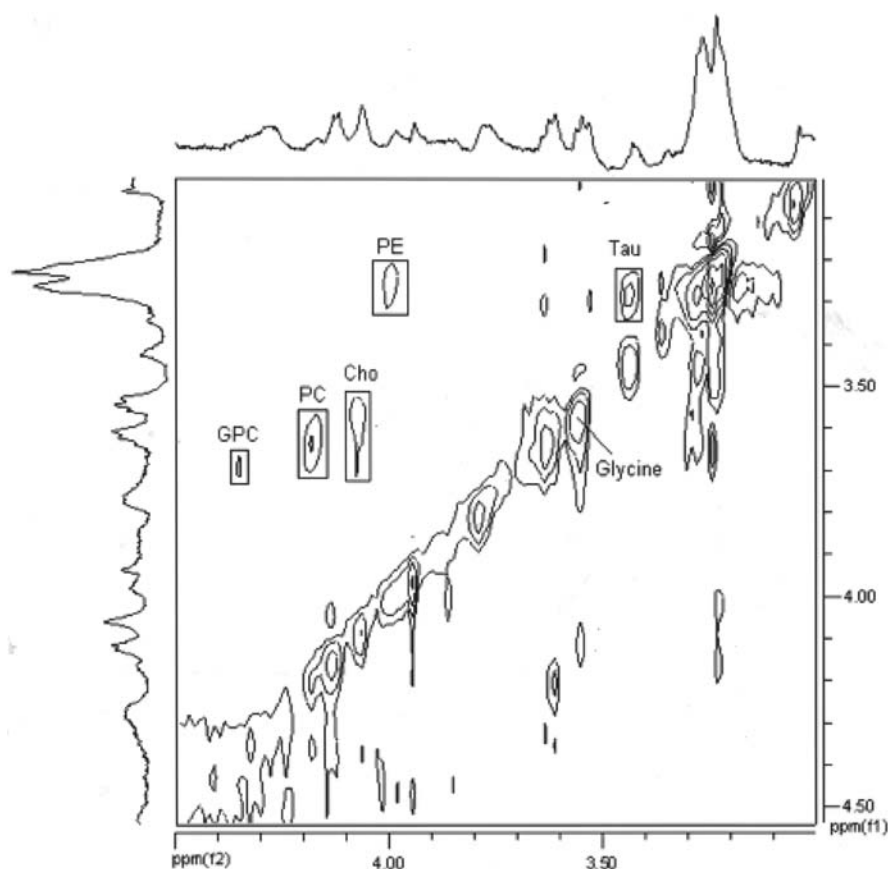


Figure 2. TOCSY (total correlation spectroscopy) using high resolution magic angle spinning (HRMAS) on a specimen from a patient with an anaplastic ganglioglioma (mixing time was 80 msec). GPC, glycerophosphocholine; PC, phosphocholine; Cho, choline; PE, phosphoethanolamine; Tau, taurine.

with the percentage of highly cellular malignant glioma (23). According to other studies, altered phospholipid metabolism, such as PCho and GPC accumulation, reflects early stages of growth arrest or apoptosis (21). In addition, GPC levels were found to increase in cultured mammalian cells exhibiting perturbed energetic metabolism during acidosis (24). There is a general consensus that tissues with high proliferative potential or tissues that are oncogenically transformed are highly cellular in the absence of compensating apoptotic mechanisms or when vascular supply is limited. Consistent with this assumption, many studies strongly suggest that the Cho peak detected by *in vivo* MRS may be elevated because the target region is highly cellular and includes cells with high PCho due to increased proliferative potential (19,25) or because it includes cells that have been oncogenically transformed (26).

It was recently reported that in *ex vivo* high-resolution MR spectra (much higher resolution than the spectra obtained *in vivo*) of biopsied tissue from a cerebellar primitive neuroectodermal tumor, myo-inositol, taurine, and phosphorylethanolamine contribute to the *in vivo* signal at 3.2 ppm, a signal usually attributed to Cho-containing compounds (27). In addition, we have used two-dimensional *ex vivo* high-resolution MRS with magic angle spinning and performed gene expression analysis in the same biopsies to further identify and validate the biological importance of MRS-detected metabolites (28). Fig. 2 shows the detail of two-dimensional MRS. Metabolites such as free Cho,

PCho and GPC, not separable by one-dimensional MRS, are separated here. We anticipate localized *in vivo* two-dimensional (2D) proton MRS will be clinically applied to analyze the metabolic profiles especially of non-operable tumors; technologic development toward implementation of localized 2D J-resolved, 2D zero-quantum, 2D double-quantum, zero-quantum-filtered COSY and SECSY, and double-quantum-filtered COSY and SECSY  $^1\text{H}$  MRS is very promising (29-34).

Cancer cells are apoptotic, and typically die upon conventional chemotherapy or radiation and other experimental approaches such as antiangiogenic and ganciclovir treatments (35). Previously, *in vivo* proton MRS detected a substantial accumulation of polyunsaturated fatty acids associated with gene therapy-induced apoptosis (36). Furthermore, depletion of PCho, a major constituent of the Cho peak detected *in vivo* in tumors, coincides with growth arrest (21). Based on these findings, it is reasonable to expect that facilitating apoptosis by means of selective chemotherapeutic agents or gene therapy could be a future strategy for treatment of human cancers (35). Importantly, for the purposes of this review, we should emphasize here that MRS can be used to monitor and detect the effectiveness of such treatments.

Other promising studies have indicated changes in the diffusion properties of cancerous tissue in response to therapy. For example, prior to the tumor's loss of volume when treated, response to treatment has been associated with an increase in tissue water diffusion and T2 relaxation



time (37), which suggests increased water content and bulk diffusibility. Reduced diffusion of Cho-containing compounds in gliomas undergoing apoptosis has also been reported (38). These observations suggest increased viscosity and restriction within cells, reflecting cell shrinkage.

Flow cytometric studies demonstrate that gene therapy-induced apoptosis (39) begins with an irreversible arrest in the late S- or G2-phase of the cell cycle (40). The MRS-visible lipids not only correlate with apoptosis or necrosis (24,27), but also with the proportion of cells in these S- or G2 stages (41). Finally, the ceramide resonance region has been associated with the differential diagnosis of high and low malignancy of brain gliomas (42). This observation deserves further investigation because apoptotic stimuli such as ceramide, a second messenger related to apoptosis, disrupt electron transport in mitochondria (43), which are important sites for apoptosis initiation (44,45).

#### 4. Methodology for acquiring proton MRSI data in a clinical setting

The methodological aspects of MRSI are not standardized; they tend to vary among investigators and clinical sites. Typically, a large volume of interest can be selected and followed by phase encoding to obtain multiple voxels in a single plane (10) or in three dimensions (8). The advantage of obtaining multivoxel data is that it allows us to observe heterogeneity within the lesion as well as to examine surrounding tissue that may appear normal on MR images. With these data, we then have a reference for comparing metabolite levels in the tumor, and can identify regions of abnormal metabolism outside the morphological lesion (46).

Proton MRSI is more demanding with regard to magnetic field homogeneity than MRI. In many circumstances (e.g., close to the sinus, cavities of resected tumors, or permanent radioactive seeds) shimming fails and water and lipid suppression becomes inadequate, thus compromising the quality of the data obtained. In addition, because of the low spatial resolution of MRSI ( $\sim 1 \text{ cm}^3$  per voxel at 1.5 T) the spectra may reveal a mixed metabolic profile of tumor, necrosis and normal brain tissue. Finally, MRSI sequences are time-consuming because they usually lack the most rapid form of gradient spatial encoding, the frequency encoding performed by the readout gradient.

Our experience with MRSI in children has demonstrated that multilevel, two-dimensional MRS data acquisitions with no gap may be used in place of three-dimensional methods. This approach improves the signal-to-noise ratio because adjustments for magnetic field homogeneity and water suppression can be performed in each section (46). The two most common methods used for volume pre-selection are point-resolved spectroscopy (PRESS) (47) and stimulated echo acquisition mode (STEAM) (48). PRESS is preferred, when the echo time allows, because of its intrinsically higher signal-to-noise ratio (49). After a 50-to-100-ml volume has been selected and shimming and water suppression adjustments have been made, a large data set is obtained by using phase-encoding gradients in two or three directions. The following parameters may be used for two-dimensional acquisitions: 1000/65 msec (repetition time/echo

time), 16x16 phase-encoding matrix, 160-mm FOV, section thickness of 10 mm, 1250-Hz spectral width, two averages and 512 points. Using these parameters, data sets with resolution of  $1\text{-}1.2 \text{ cm}^3$  are acquired.

The decision to use an echo time (TE) of 65 msec is appropriate when one is more interested in the detection of lipids than lactic acid. Lipids, because they are related to tumor necrosis and apoptosis, serve as indicators of tumor grade. Our current notion is that a TE of 65 msec provides us with the opportunity to null lactic acid, increase sensitivity to lipid detection, and prevent diffusion artifacts and water-suppression failures that occur when PRESS is performed at TEs shorter than 65 msec. Thus, the four prominent peaks of biological importance in our studies were those of NAA, Cho, tCr and lipids (and/or lactate) (Fig. 3).

As stated, an advantage of the approach described is that the volume of interest can be selected to eliminate most subcutaneous lipids and to avoid regions likely to cause large variations in susceptibility, such as the sinuses. It also permits improved shimming and provides spectra with narrower peaks and higher signal-to-noise ratios. Figs. 1 and 3 show examples of multivoxel spectra from normal brain tissue, necrosis and different regions of brain tumors (10). In the normal brain, NAA has approximately twice the signal intensity of Cho and tCr. Tumors generally have decreased NAA and increased Cho and are associated with variable levels of tCr. Peaks corresponding to lipid and/or lactate may be present in regions of necrosis (10,46). This is apparent in pilocytic astrocytomas where necrosis is present (Fig. 3).

Many new approaches have been developed to improve the performance of conventional MRSI (50). Special alternative radiofrequency pulses provide improved spatial and frequency selection (51,52) and better volume selection can be achieved with spatially selective saturation bands (53). Multislice and multiple echo time techniques (54,55) can be used to acquire multivoxel MRSI data in a time-efficient manner; and fast, three-dimensional multivoxel MRSI can be achieved with echo planar methodology yielding good-quality data (56). Magnetic resonance spectroscopists look forward to the development of hybrid PRESS-echo planar spectroscopic imaging techniques with spatially selective saturation bands that may speed up MRSI and overcome its present limitations in water suppression, volume selection and susceptibility artifacts (57).

#### 5. Processing and analysis of proton MRS data in a clinical setting

The processing and analysis of the resulting proton magnetic resonance spectra combines Fourier transforms and apodization with automated methods of spectral processing to provide data that can be interpreted by visual inspection or quantified to generate maps showing the spatial distribution of different metabolites (58). Different magnetic resonance system manufacturers offer different packages for proton MRS analysis. Typically, the data are transferred off-line to a remote workstation, converted into a standard data format, Fourier-transformed and phased using appropriate spectroscopic packages. The quantification of *in vivo* spectra in a

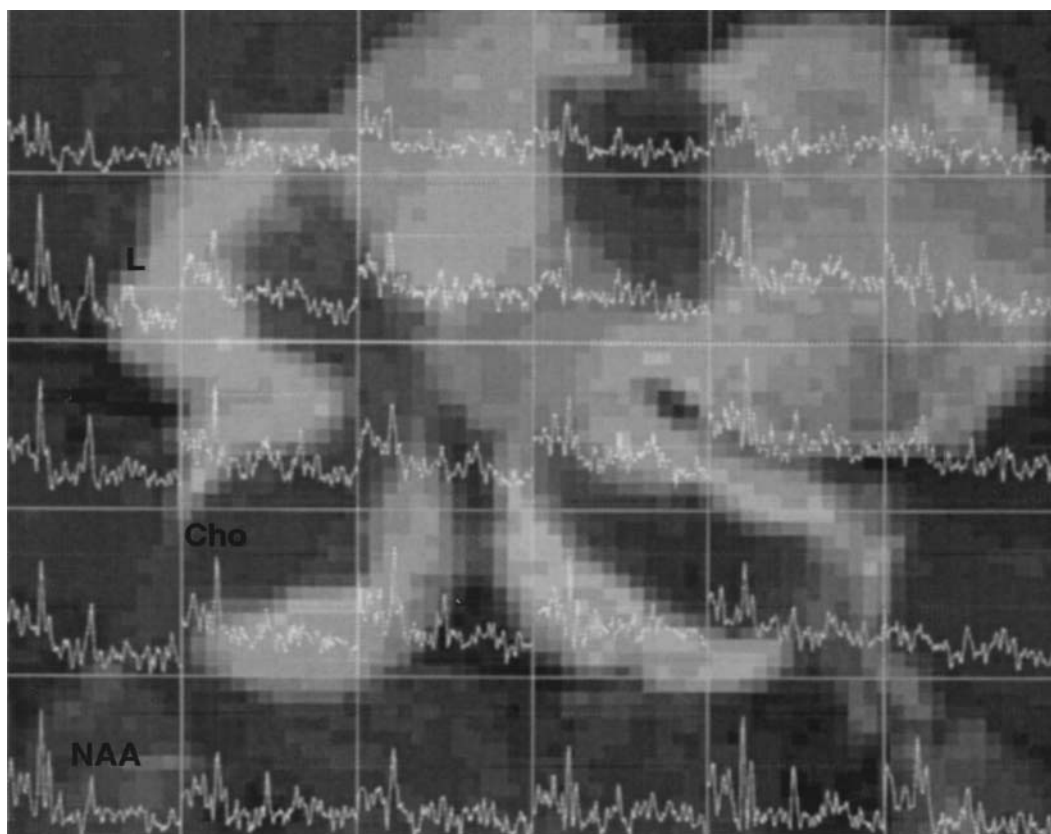


Figure 3. Magnetic resonance spectra from a multivoxel MRS data set superimposed on an T1-weighted gadolinium-enhanced magnetic resonance image of a well-differentiated pilocytic astrocytoma (NAA, N-acetylaspartate; Cho, choline-containing compounds; L, lipids and/or lactate).

reliable and reproducible manner requires removal of baseline components, identification of peaks, and estimation of peak parameters, which can be accomplished using several different approaches (59-61). Characteristics of the proton MRS data that inform the choice of methodology include the number of spectra that need to be considered, and the sensitivity of the method to differences in signal-to-noise ratios and peak configurations corresponding to different tissue types. Additionally, more sophisticated fitting algorithms can be applied to spectra whose signal-to-noise ratios are sufficient to ensure that optimization routines are reliable (62). The output of the analysis is a number of spatial maps of metabolite parameters that can be applied to identify regions of normal and abnormal metabolism.

We have used a software application in interactive data language (IDL). It employs the PIQUABLE algorithm, which has the advantages of being automated, of using non-parametric methods for objective identification of peaks, and of being capable of removing broad baseline components. This algorithm has been tested using simulated data (63) and data from human volunteers and patients (64). We have calibrated our software with simulated spectra and spectra from patients, and found reliable and reproducible results within the accuracy of random noise. Additional corrections for spatial variations in intensity caused by the data acquisition procedures may be required when comparing relative intensities of metabolites such as Cho, creatine, NAA, lactate and lipids (58). When the PIQUABLE algorithm fails, we use

alternative quantitation algorithms in the magnetic resonance user interface (MRUI) quantitation package.

Several approaches may be used to display the information from multivoxel MRSI data sets and to correlate anatomy with spatial variations in metabolites. Superimposing a grid on the magnetic resonance image and plotting the corresponding array of spectra is one method. This approach does not require quantification and can be quickly performed after data collection. Another approach is to form metabolite images from arrays of estimated peak parameters. The primary resonances of interest are NAA, Cho and tCr, as well as lipid resonances at 0.9 ppm (methyl groups), 1.3 ppm (methylene groups and lactate), 2.8 ppm (bisallylic methylene fatty acids); and at 5.4 ppm (vinyl protons, including ceramide). In addition, other metabolites, such as glutamate, glutamine,  $\gamma$ -aminobutyric acid, scyllo-inositol, aspartate, taurine, N-acetylaspartylglutamate, glucose and branched amino acids may be detected (65). To visualize the spatial distributions that correspond to the metabolites of interest, gray-level images mapping the peak area of these metabolites may be obtained. A third method is overlaying colored metabolite images on gray-level MR images to facilitate estimation of the anatomic correlation of the varying levels of color metabolite images. Finally, it is also possible to extract selected spectra from the MRSI data sets for correlation with data from other modalities or with *ex vivo* high-resolution magic angle spinning proton MR spectra of tumor biopsies that correspond to the same anatomic location.

## 6. Contribution of proton MRSI in tumor grading in a clinical setting

Whether proton MRSI is able to contribute to defining tumor type and grade remains an open question. Although elevated Cho and low NAA levels before therapy may be a reliable indicator of malignancy in pediatric brain tumors (6,10), the consensus is that proton MRS results should not be used alone to classify tumors, as there can be considerable overlap in proton MRSI results among different tumor types (66,67). In our experience, the different MR spectral patterns suggest that proton MRSI can be used to distinguish at least three different tissue types - normal, tumorous, and necrotic (10,68) whereas mixed MR spectral patterns can be used to map the known heterogeneity of tumors, as confirmed by histo-pathologic features (46). This notion is consistent with other reports (69,70). MR spectral patterns with elevated Cho and lipid levels in the absence of NAA, histologically verified to represent regions of active tumors with extensive areas of necrosis, suggest that such MR spectral patterns contribute additional information not available from conventional MRI.

Because glial tumors are graded according to their cellularity, proliferative activity, and degree of necrosis, identification of increased cellularity and proliferative activity through Cho mapping may contribute to the value of MRI in patients with brain tumors, especially when combined with identification of necrosis and/or apoptosis through lipid mapping. Indeed, contrast-enhanced regions with high lipid levels and low or no Cho may represent areas of high neoplastic potential intermingled with microscopic necrosis. This was previously found in 11 patients with malignant or inoperable tumors and was verified at biopsy (46). Low-grade tumors not showing enhancement on T1-weighted gadolinium-enhanced images exhibit prominent peaks corresponding to Cho. Although tCr and NAA peaks are occasionally detected, the absence of lipids and lactate is noted (Fig. 3).

Successful classification of tumors in pediatric patients with posterior fossa tumors was reported by Arle *et al*, who used single-voxel proton MRS and a computer-based neural network (71). The network combined MRS data (ratios of NAA, Cho and tCr) with 10 characteristics of tumor tissue obtained from MR images, tumor size and the patient's age and sex, which improved diagnostic accuracy to identify 95% of the tumors correctly. A similar approach was used by Poptani *et al* (72). Given the differences in spatial extent of tumors, however, the question arises as to whether single-voxel MRS is able to contribute to defining tumor type and grade. To this end, Preul *et al* reported excellent classification of brain tumors in adult patients using two-dimensional proton MRSI and a multivariate pattern-recognition analysis of peaks corresponding to Cho, tCr, NAA, lactate, lipids and alanine (73). In this study, grade 2 gliomas tended to have low lactate and lipids, some NAA, and some creatine while grade 3 gliomas had less NAA and creatine and higher Cho. Grade 4 gliomas, in contrast, had high lactate and lipids and very low NAA. Upon visual inspection of spectral patterns and metabolite levels in each class, meningiomas were distinguishable as the only lesions that contained alanine.

Although these initial results are very promising, there has not yet been a prospective study using the statistical classification that these authors derived. One of the complications in analyzing data obtained with a multivoxel data acquisition technique is determining which spectrum to consider for each lesion. Suggestions include using the most abnormal voxel or the average of all voxels within the lesion. Both of these approaches involve a subjective decision that takes into account the anatomical appearance of the lesion. For example, does the lesion include the entire T2 abnormality or is it restricted to the enhancing volume? As seen in a study by Li *et al* (70), the spectral characteristics of these regions may be quite different. The same issue is present with single-voxel analysis, but in that case the decision is made implicitly at the time of data acquisition by the choice of the selected volume. The studies from Nelson's group at UCSF have suggested that although it may be possible to detect mean differences between populations of gliomas of different grades based upon metabolite levels, there is considerable overlap, both for mean metabolite levels and the most abnormal voxels within the T2 lesion (70).

In our experience, information such as relative cerebral blood volume (rCBV) or the apparent diffusion coefficient (ADC) may also help in grading tumors and in distinguishing between tumors and other types of mass lesions (74). Because brain tumors can be characterized according to their physiological parameters, including proton MR spectral metabolites (NAA, Cho, tCr and lipids), hemodynamic indices (e.g., rCBV), and physicochemical measures (e.g., ADC), relationships among these parameters may reflect the biochemical state of tumors; this view is supported by our findings to date (74). Finally, patient age and tumor location in addition to anatomic factors seem likely to be relevant for proper classification. Whichever procedure is used, the influence of all factors should be explicitly considered in the analysis. Recently, we also proposed the combination of high-resolution magic angle spinning proton magnetic resonance spectroscopy and microscale genomics to type operable brain tumors (28). Additionally, localized *in vivo* two-dimensional (2D) proton MRS is clinically promising (29-34).

## 7. Role of proton MRSI in predicting response to therapy in a clinical setting

A number of studies suggest that proton MRS promises an early prediction of whether a lesion has responded to therapy (6,15,66,75,76). In our experience, although Cho is found to be the strongest metabolite signal detected in tumors, the only significant independent predictor of active tumor growth is tCr (15). We have also found the percent change in the Cho/NAA ratio to be the most promising prognostic index in children with brain tumors (6). Fig. 1 shows how this index can be used in conjunction with perfusion imaging indices (i.e., rCBV) for diagnostic and possibly prognostic evaluations.

For proton MRSI to be included in the clinical management of pediatric patients, it is important that the independent information gained from MRSI improves the assessment of tumors. In gadolinium-enhanced MR images, for example, the relationship between the extent of the tumor



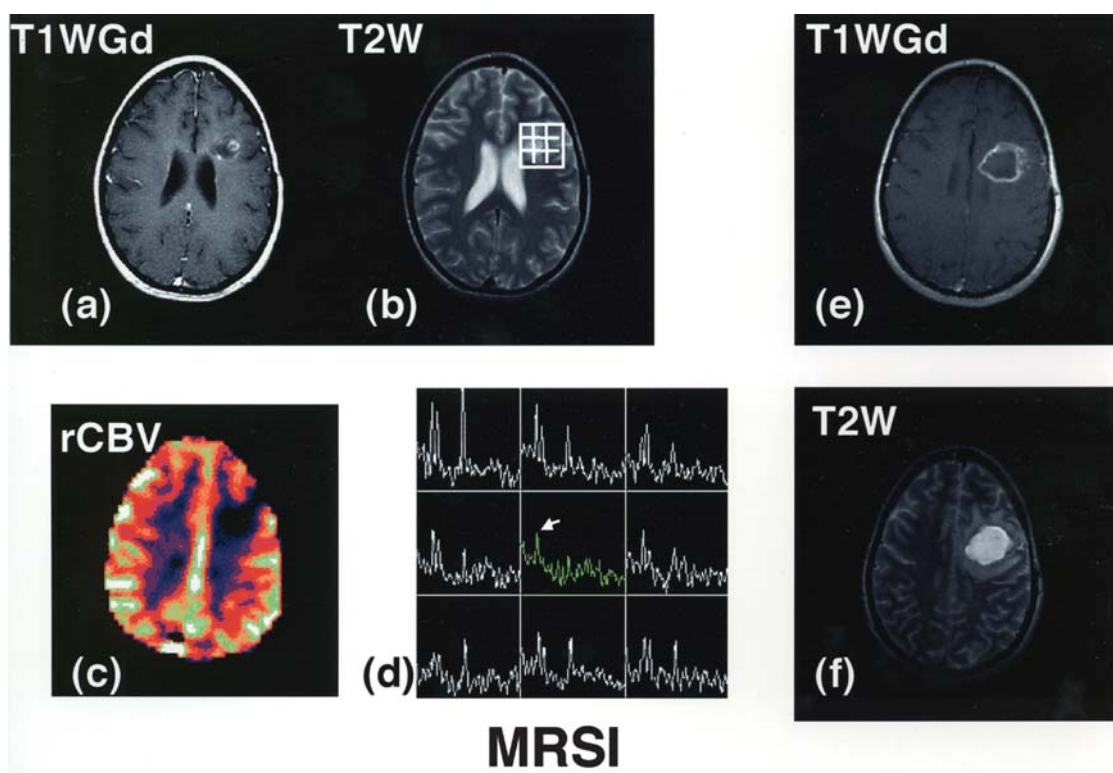


Figure 4. Baseline magnetic resonance images and MRSI (a-d) and 1-month follow-up magnetic resonance images during therapy (e and f) from a 13-year-old female patient with an anaplastic ependymoma.

cells and the contrast-enhanced regions is unclear. If proton MRS were capable of defining tumor borders, it would allow an ineffective treatment strategy to be modified before the tumor progresses further. In pursuit of this goal, we analyzed MRI and MRSI data in 31 children with brain tumors and found that certain spectral patterns were detected in regions of the tumor and outside enhancing tumor beds in patients with clinically determined progression. These findings were confirmed by neuropathological analysis (46).

This study demonstrates the importance of mapping both the temporal and spatial distribution of metabolite changes in response to therapy. Such mapping requires the use of two- or three-dimensional proton MRSI, and is most easily achieved with focal therapies such as surgery or radiation. The incorporation of multiple imaging modalities into therapy planning offers the potential to better identify regions of pathology. On this basis, multiparametric and/or multimodality imaging has been proposed (74,77). Registration of MR images and proton MRS data is critical for correlating data from such examinations. Excellent results from studies in adults undergoing brain tumor therapy at the University of California, San Francisco have already been reported (69,77-79). In our opinion, multivoxel proton MRSI is more powerful than conventional or more novel types of MRI, such as perfusion MRI, for predicting tumor progression (6). Although proton MRSI results generally correspond with perfusion MRI, they may be superior to perfusion MRI in cases where perfusion MRI is limited (Figs. 1 and 4). More importantly, our analysis in 76 children has shown that brain proton MRSI biomarkers predict survival of children with central nervous system (CNS) tumors better than standard

histopathology (80). More accurate prediction of survival using MRSI represents an important advance and may be used to suggest more appropriate therapy, especially when diagnostic biopsy is not feasible.

## 8. Conclusion

*Use of proton MRSI in the clinical management of pediatric patients with brain tumors.* The therapeutic approach to pediatric patients with malignant brain tumors is multifaceted and varies depending on the location and resectability of the tumor, as well as the patient's age (81). Surgery continues to be the treatment of choice for most patients, although overall effectiveness is often limited by the primary location of the tumor and the extent to which the disease has disseminated (82,83). Radiation therapy has a documented role in the treatment of children with brain tumors (84,85) despite the finding that in most high-grade glial tumors any amelioration is temporary. Furthermore, as stated previously, the deleterious effects of radiation therapy on the developing nervous system militate against use of this modality. Finally, chemotherapy is effective predominantly in neural tumors and low-grade gliomas, not in patients with high-grade gliomas or recurrent/ progressive disease (86).

Given the difficulties inherent in performing sequential biopsies of brain tumors in children, non-invasive and non-irradiating methods are needed to provide diagnostic/prognostic indices or biomarkers beyond simple tumor volume measurements. Moreover, an important unresolved issue in brain tumor therapy is the difficulty in differentiating dying or necrotic CNS tissue from a viable, recurring tumor

(64). By both clinical and standard CT or MRI scan criteria, necrosis and recurring tumors can appear to be identical, thus the ability to differentiate between a growing tumor and necrotic tissue at an early point in time is of great importance.

Effective patient management requires access to the biological activity of tumorigenesis inhibitors and/or chemotherapeutic drugs. Advanced neuroimaging MR techniques, such as proton MRSI, promise to help differentiate between these at an early stage. In addition, the spatial extent of the metabolic lesion by MRSI is different from the gadolinium-enhancing region and hyperintensity on T2-weighted images (46). Given such a distinction, there may be added value in having access to proton MRSI data over and above what is obtained from conventional MRI. Of key importance, however, is the general consensus that proton MRSI may be able to predict early on whether a lesion will respond to therapy (6). If so, that would allow tailoring therapy to each individual patient or modifying an ineffective treatment strategy before the tumor progresses. It would also allow clinicians to avoid giving unnecessary treatment in a case where increase in tumor volume is attributable to treatment-induced necrosis as opposed to a recurrent or residual tumor.

Although the clinical relevance of proton MRSI has yet to be determined, it is clear that this technique improves the assessment of pediatric brain tumors by adding independent biochemical information regarding tumor type, grade and involvement and by depicting residual or recurrent tumors outside the gadolinium-enhancing tumor bed. MRSI is an invaluable adjunct to MRI and other modalities currently in clinical use. Of key importance to clinicians, proton MRSI promises to provide biomarkers capable of predicting tumor response earlier than is possible with conventional MRI. Furthermore, since higher field magnetic resonance systems have been approved by the FDA and are already being introduced in the clinical setting, it is likely that high-field, higher resolution proton MRSI will be used in inoperable tumors in the near future to complement neuropathology, guide biopsies, and monitor the success and failure of therapy for operable brain tumors. Correlative studies with genomic biomarkers (work which is under way in our laboratory) will strengthen the biological and clinical relevance of proton MRSI.

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