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Magnetic Resonance in Preterm and Term Newborns: ^{1}H-Spectroscopy in Developing Human Brain

PETRA S. HÜPPI, STEFAN POSSE, FRANÇOIS LAZEYRAS, ROLAND BURRI, EMILIO BOSSI, AND NORBERT HERSCHKOWITZ

Department of Pediatrics, University of Berne, Berne, Switzerland

ABSTRACT. Localized proton magnetic resonance spectra were recorded from human cerebellum *in vivo* with a 1.5-T magnet. The spectra from healthy adults and preterm and term babies showed resonances from N-acetylaspartate, creatine and phosphocreatine, choline-containing compounds such as phosphocholine and glycerophosphocholine, taurine, and inositol. The age-dependent changes of *in vivo* molar concentrations of N-acetylaspartate, choline, taurine, and inositol were estimated in preterm babies, babies at term, and adults. The range of postconceptional age in the studied babies was 31 to 45 wk. Taking the biochemically measured creatine concentrations in age-corresponding autopsy material as an internal standard, the *in vivo* concentrations of the other metabolites were calculated from the proton spectra. N-acetylaspartate showed an increase from 1.9 mM in preterm babies to 3.1 mM in term babies and to 6.5 mM in adult brain. Taurine was noted to increase from 1.1 mM in preterm infants to 2.3 mM in term infants and did not decrease significantly in adult brain. Choline and inositol concentrations did not change significantly throughout the studied age groups. These new data on *in vivo*, localized ^{1}H-spectroscopy show that it is a sensitive method for studying early metabolic brain development in humans. (*Pediatr Res* 30: 574–578, 1991)

Abbreviations

^{1}H-MRS, proton magnetic resonance spectroscopy
NAA, N-acetylaspartate
Cr, creatine and phosphocreatine
Cho, choline
Tau, taurine
Ino, inositol
PCA, postconceptional age
NAAG, N-acetylaspartyl-glutamate
GABA, γ-aminobutyric acid
NMR, nuclear magnetic resonance
MR, magnetic resonance

In vitro and *in vivo* ^{1}H-MRS is a powerful noninvasive method used to identify and quantitate chemical compounds (1). The ^{1}H nucleus has several important advantages (2, 3). It has the highest sensitivity of any stable nucleus, 100% natural abundance, and is found in virtually all metabolites. It therefore provides access to a large number of metabolites, and detailed biochemical information may be obtained not only on energy metabolism (lactate production) but also on free amino acids, fatty acids, and neurotransmitters. By *in vitro* ^{1}H-MRS, Arus *et al.* (4) were able to distinguish more than 15 different compounds in rat brain perchloric acid extract. In addition, Cerdan *et al.* (5) detected cerebral *myo*-inositol in rat brain perchloric acid extracts. Behar *et al.* (6) were the first to record ^{1}H-NMR spectra of *in vivo* rat and rabbit brain. Avison *et al.* (7) recently detected phenylalanine in experimental hyperphenylalaninemic rabbit brain by *in vivo* ^{1}H-MRS and compared these data with column chromatographic estimation of phenylalanine in brain and serum. This opens up a new possibility of monitoring the treatment of phenylketonuria. Luyten and den Hollander (8) presented the first human brain spectra identifying the resonances of NAA and Cr. Barany *et al.* (9) added the detection of glutamate, choline-containing resonances, taurine, and ring protons of amino acids. Frahm *et al.* (10) presented a reliable method to detect the above-mentioned metabolites *in vivo* in localized brain areas.

Very recently, data were presented on developmental changes in ^{1}H-MRS and ^{31}P-MRS in infants aged 1 mo to 16 y (11). Peden *et al.* (12) presented data on spectroscopic abnormalities in infants with perinatal ischemic and hemorrhagic brain injury. We were interested in using ^{1}H-MRS to study age-dependent changes in preterm and term infants' brain. The cerebellum was chosen for its well-defined and homogenous area and its maturational changes throughout fetal and postnatal life with a rapid developmental change in the perinatal period (13). We report here the first data on localized ^{1}H-MRS in preterm and term newborns in comparison to ^{1}H-MRS in adults.

PATIENTS AND METHODS

Twenty infants and six healthy young adult volunteers were included in the study. The mean gestational age of the babies at birth was 33 ± 2 wk (range: 27–42 wk). The mean PCA at the MR examination was 37 ± 4 wk, with a range of 31 to 45 wk. The babies were divided into two groups. One group consisted of 12 prematurely born infants with a PCA at MR examination below 37 wk (mean ± SD: 34.7 ± 1.8 wk; range: 31–37 wk) (preterm), the second group consisted of 8 prematurely born and term infants with a PCA at MR examination of over 37 wk (mean ± SD: 40.8 ± 2.2 wk; range: 39–45 wk) (term). Fifteen newborns were hospitalized for close prophylactic monitoring, two had periventricular leukomalacia (one in each group), two had neonatal seizures (one in each group), and one had mild birth asphyxia (included in the term group); none had to be mechanically ventilated. All the babies were fed enterally, mainly mother's milk with an additional supply of calories for preterm infants (120–140 kcal/kg).

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Correspondence: Petra S. Hüppi, M.D., Department of Pediatrics, University of Berne, Inselspital, 3010 Bern, Switzerland.
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For MR examination, the babies were kept in a specially designed MR incubator with continuous temperature control and in some cases oxygen application. Cardiorespiratory function was monitored by pulse oximetry (Nonin, Plymouth, MN) and capnograph (Odam, Wissembourg). No sedation was needed in the neonates. A neonatologist was present inside the magnet room during the examination. The study had previously been accepted by the clinic's ethical committee and informed consent was obtained from parents of patients and volunteers before entering the study.

A 1.5-T MR unit was used for imaging and spectroscopy (Signa Performance Plus, General Electric, Milwaukee, WI). A quadrature head coil served for spectroscopy in adults. A 3-inch diameter receive-only surface coil (with body coil transmit) or a 20-cm diameter extremity coil (birdcage) was used for the babies. A localization image was obtained to position to voxel of 1.5 × 1.5 × 1.5 cm within the cerebellum in both adults and babies. The homogeneity of the magnetic field was optimized by shimming with 3–4 Hz, which corresponds to 0.05 ppm. For the acquisition of the spectra, a stimulated echo pulse sequence as described by Posse *et al.* (14) was used, with sinc water suppression, pulses of 100 Hz band width, an eight-step phase cycle of the STEAM pulses, and complete outside volume presaturation: repetition time was 2000 ms and echo time was 6 ms. Automatic zero order phase correction was applied.

Data processing was done by exponential multiplication enhancement and without baseline correction. A manual line-fitting routine, considering purely Lorentzian line shape in the frequency domain, was carried out, whereby the line integrals represent signal strength.

In vivo concentrations of four major cerebral metabolites were calculated using a biochemically measured "internal standard."

These calculations were based on Cr concentrations measured by HPLC in age-corresponding human autopsy material of fetal, neonatal, and adult brain tissue as described by Burri *et al.* (15): HPLC parameters were Aquapore (Brownlee Labs, Santa Clara, CA) RB-300 column, 220 × 4.6 mm, 7-μm particle size; mobile phase to 930 mL of a 2:1 (vol/vol) mixture of 0.02 M citric acid and 0.02 M Na_2PO_4, 0.58 g octylsulfonic acid (final: 2.5 mM), 18.6 mg EDTA (final: 0.05 mM), and 70 mL methanol were added. The pH of this solution was between 3.10 and 3.20, flow rate was 0.75 mL/min, and detection was at 210 nm (16).

Autopsy data were taken from adult patients with an age range of 23–84 y. Cause of death was other than brain pathology. Medical history was free of CNS disease, and none of the patients was recorded to be mentally retarded. Autopsy data for the infants included fetuses and babies at term who died from various causes at birth or soon thereafter because of heart malformation. Time between death and autopsy was kept as short as possible and was usually a few hours in a cooled room (3–4°C); at autopsy brain tissue was immediately frozen.

The mean values obtained for a certain age group were used to define the peak area of Cr in the spectra. Using this internal standard, the *in vivo* concentrations of other metabolites were calculated, taking into account spectral area and number of detectable protons per molecule.

Comparison between groups was done by statistical analysis using two-tailed *t* test for independent samples.

RESULTS

The brain ^{1}H-spectra of newborns obtained from the voxel in the cerebellum showed the same resonances of the metabolites as those known from adult data. With the achieved spectral resolution, NAA, Cr, Cho, Tau, and Ino could clearly be assigned. However, when the single signal strength of the cerebellar spectra in two groups of newborns and adults were compared, clear differences in relative peak heights were noted (Figs. 1 and 2).

To estimate *in vivo* concentrations of the observed metabolites in human brain, total Cr concentrations from human cerebellum were used as an internal endogenous marker. The values presented in Table 1 are the mean ± SD for a group of fetuses and term babies with similar gestational age as the spectroscopically examined group. By taking these chromatographically measured Cr concentrations as an internal standard for the Cr peak area, *in vivo* concentrations (mM) of NAA, Cho, Tau, and Ino were obtained in the three age groups. NAA (Fig. 3*A*) significantly increased from 1.8 mM in preterm infant cerebellum to 3.1 mM in term infants and to 6.5 mM in adults. The Cho concentration (Fig. 3*B*) was the same in the two infant groups and showed nonsignificant lower levels in adults. Tau (Fig. 3*C*) increased significantly from 1.1 mM in preterm infants to 2.3 mM in term infants, but no differences between term infants and adult values were observed. Ino concentration (Fig. 3*D*) remained stable throughout the three age groups.

DISCUSSION

Until now, the study of the metabolites shown was restricted to either animal work or postmortem analysis of human autopsy brain tissue (17–22). Both models have obvious restrictions. Animal studies show considerable species and regional differences, and postmortem analysis bears the uncertainty of autolytical alterations. Comparison between data on concentrations of the metabolites as published in the literature and results obtained from *in vivo* localized ^{1}H-spectroscopy is therefore difficult. Our own results with ^{1}H-MRS showed higher levels of neonatal NAA than the NAA concentrations obtained from classic biochemical analysis of autopsy material from the same age group (2.3 *versus* 1.3 mM). It is known that NAA concentration is reduced in brain by autolytical processes beginning after 15 min at room temperature (17). Tau, on the contrary, seems to increase with autolytical processes. Regional differences in metabolite concentration are also important. The obtained chromatographic results in a term born infant cerebellum, which was frozen immediately, and the spectroscopically obtained values in the cerebella of the group of babies at term are in good agreement for NAA (3.1 *versus* 2.5 mM) and Tau (2.3 *versus* 2.1 mM). A comparison of our spectroscopic results with earlier published data from *in vivo* ^{1}H-spectroscopy by Frahm *et al.* (23) in adult brain show that our concentration levels are lower. Frahm's results were based on an assumed total Cr of 10 mM, as in data from *in vitro* rat brain (24). Our measurements in adult human brains at autopsy, however, yielded a mean total Cr concentration of only 6.2 mM. Taking this value as internal standard, Frahm's spectroscopic results are very similar to ours (NAA 7.8 mM, Cho 1.4 mM, Tau 0.9–1.2 mM, Ino 4.4 mM). Differences in the concentration of the internal standard have to be taken into account when comparing the results of different research groups. Spectroscopic measurements of NAA in gray and white matter perchloric acid extracts of human brain by Petroff *et al.* (25) showed excellent agreement with our results obtained by *in vivo* ^{1}H-MRS. To reduce the alteration of metabolite concentration in autolytical processes, Cr was chosen for internal standard, because it is least affected by autolytical processes. Large interindividual differences would affect the calculation. SD in our measurements were 10% or less.

Further problems that have to be taken into account when quantifying *in vivo* proton spectra are J-modulation effects on spin-coupled resonances, which were almost excluded in this study by using very short echo times. Overlap of multiplet resonances, though, are still critical for resonances such as Ino and Tau and even more so for glutamine, glutamate, and GABA. Because of the very short echo time, we were able to measure resonances with short T_2 relaxation times. We agree that this leads to the detection of a variety of substances in the spectra causing overlap of resonances. Such substances are carnitine, ethanolamine derivatives, glycine, glucose, threonine, betaine, trimethylamine, GABA, glutamate, and NAAG.

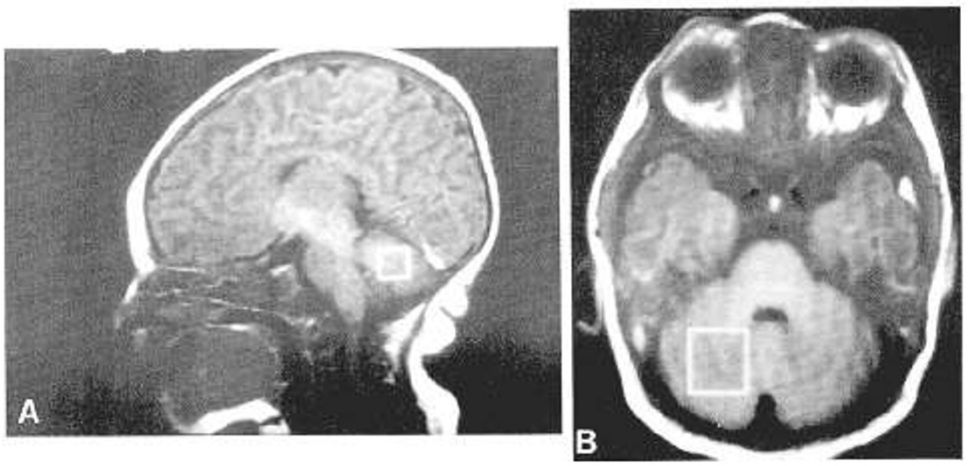

Fig. 1. *A,* Sagittal MRI with voxel, localized in the cerebellum, of a preterm infant examined at term (repetition time 400 ms, echo time 20 ms). *B,* Axial MRI with voxel, localized in the cerebellum, of a preterm infant examined at term (repetition time 400 ms, echo time 20 ms).

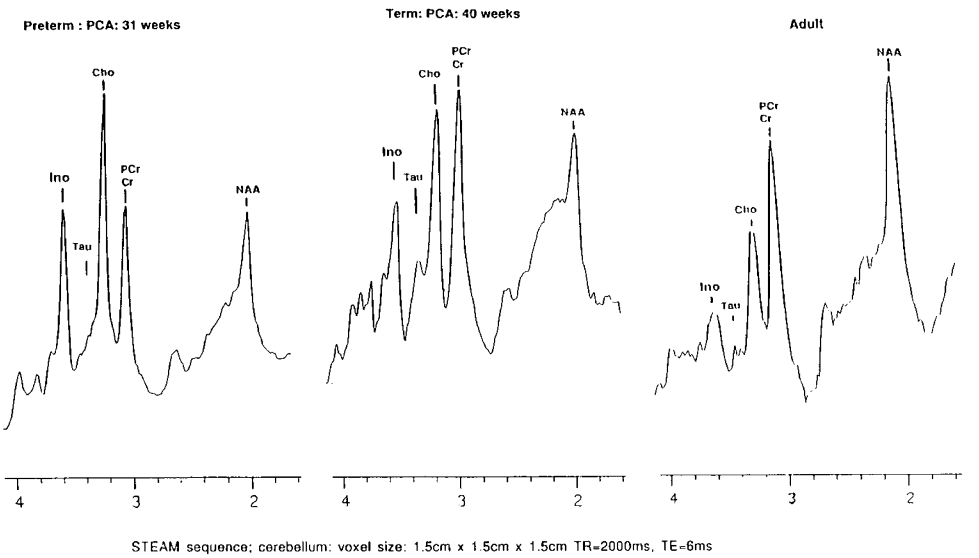

Fig. 2. Localized *in vivo* ^{1}H-spectra of a preterm infant (31 wk PCA at examination), a term infant (42 wk PCA at examination), and an adult.

Table 1. *Cr concentration in human brain autopsy material, used as internal standard for ^{1}H-MRS (mean ± SD)*

	Cr (mM)
Preterm ≤ 37 wk (n = 3)	3.78 ± 0.03
Term ≥ 38 wk (n = 3)	4.69 ± 0.58
Adult (n = 5)	6.22 ± 0.62

Reported brain concentrations of carnitine, ethanolamine derivatives such as diacylglycerophosphoethanolamine and alkenylacylglycerophosphoethanolamine, glycine, threonine, GABA, and NAAG are between 0.1 and 0.5 mM, a range of concentrations that is beyond detectability by *in vivo* NMR (19, 25–28). These substances will therefore not affect area measurements of detected metabolites significantly. In patients with diabetes and high glucose levels, resonances of glucose with a partial overlap with Tau were detected. However, intracellular brain glucose in physiologic levels could not be detected by *in vivo* ^{1}H-MRS. Michaelis *et al.* (28) claim the separate observation of Tau in proton spectra of normoglycemic patients. Neither of our patients showed hyerglycemia. The two degradation products of Cho, betaine and trimethylamine, are produced by intestinal bacterial flora, which in preterm and term newborns are not yet developed; therefore, production of these substances is minimal.

In analytical high-field NMR spectroscopy, glutamate can be detected with resonances at 2.35, 2.11, and 3.76 ppm. At lower field strength, as used in *in vivo* spectroscopy, coupling effects are so strong that all glutamate resonances are considerably split and therefore reduced. For measuring glutamate this is a great disadvantage; however, for the problem of overlap and peak area measurements of the interfering resonance (NAA), this minimizes erroneous calculation of NAA due to glutamate overlap.

The babies with pathologic clinical findings were included in the study because their cerebella showed no defect at MR imaging and the obtained spectroscopic results showed no statistical differences from those of the healthy population.

With this method we were able to show a 2-fold increase of NAA in preterm brain (31–37 wk PCA) to term infants' brain (38–45 wk PCA) and a 2-fold increase of NAA from term to adult brain in humans. This is less than the observed 5- to 7-fold increase of NAA from newborn to adult brain in rats and rabbits (29, 30). The significance of these findings is still unclear because the functional role of NAA is not yet fully known (31). It has been shown that the acetyl group of NAA is very effectively incorporated into brain lipids (32, 33). A correlation between metabolite concentration and extent of myelinogenesis assessed by MR spectroscopy and imaging might clarify the possible role of NAA in myelination. Other still speculative functions of NAA are its role in neuronal protein synthesis (34) and its function as precursor of the putative neurotransmitter NAAG (35). When the age-corresponding, normal *in vivo* NAA values for different brain areas are known, the studies can be extended to pathologic

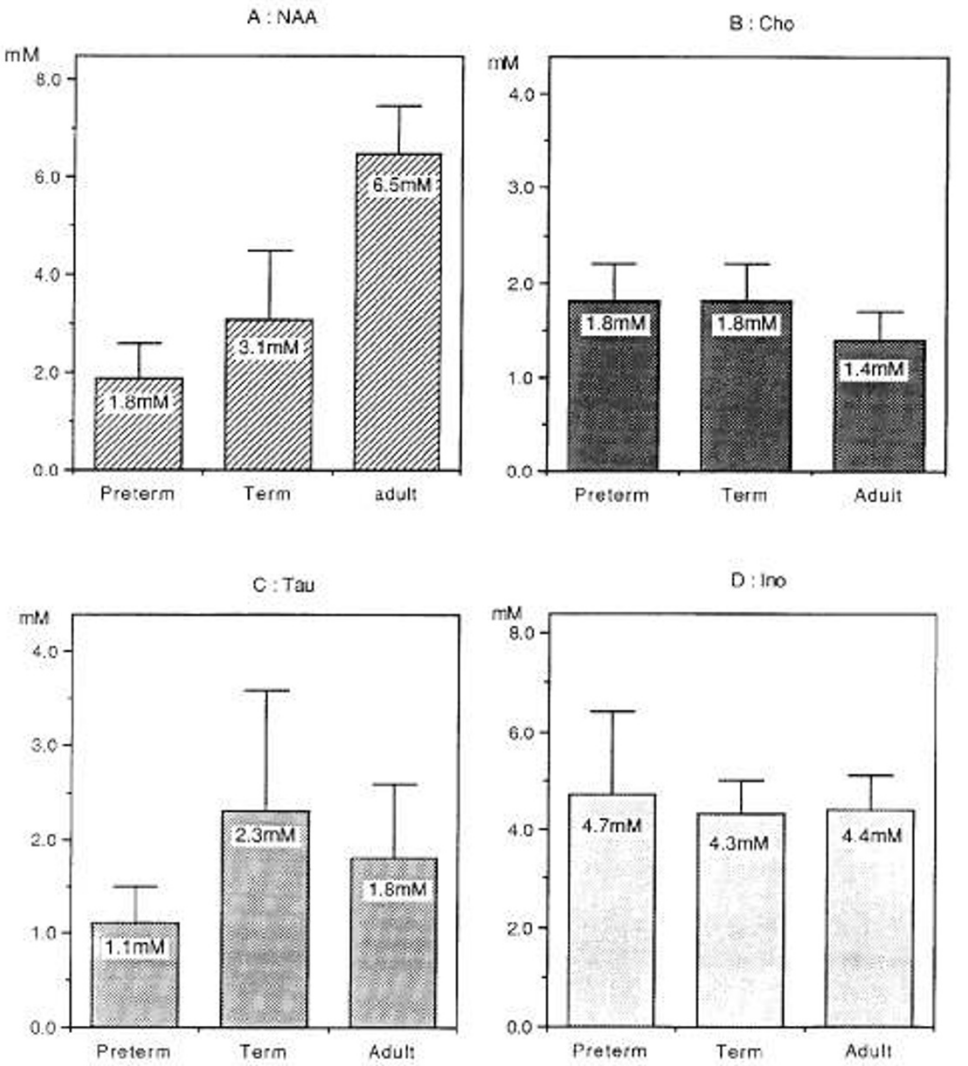

Fig. 3. Developmental changes of NAA, Cho, Tau, and Ino concentration in the cerebellum of preterm and term infants and adults, measured with *in vivo* ^{1}H-MRS (group mean and SD). *A,* Significant increases of NAA in the three age groups: preterm/adult, $p < 0.001$; term/adult, $p < 0.01$; preterm/term, $p < 0.05$. *B,* Cho concentration was unchanged in the three age groups. *C,* Significant increase of Tau from preterm to term infants and from preterm infants to adults: preterm/term, $p < 0.01$; preterm/adult, $p < 0.05$. *D,* Ino concentration was unchanged in the three age groups.

conditions, where localized alterations in NAA content in tumors or in areas of neuronal damage and demyelination can be evaluated (14, 36, 37).

The cerebellum of term infants showed the highest Tau level throughout the studied age groups. Our mean value of 2.3 mM is in agreement with early biochemical studies by Okumura (38), who showed levels of 1.8 mM in human fetal brain of 8 mo gestation. Tau is the most abundant free amino acid in neural tissue and is not usually incorporated into proteins. The importance of Tau in brain development has recently been shown: kittens that were deprived of Tau prenatally and postnatally had severe cerebellar dysfunction with delay in cell migration and failure to form synaptic connections (39, 40). The possible role of Tau in facilitating synaptic connections in the developing brain could explain the high concentration at a time of intense synaptic network formation (41). Because it is known that the activity of the rate-limiting enzymes of Tau biosynthesis in fetal brain and liver is low or even absent in the neonatal period, nutrition should provide sufficient Tau during the neonatal period (42). Human milk is known to contain high concentrations of Tau in contrast to cow's milk (43). Nutritional effects on cerebral Tau concentrations are currently under investigation.

The sum of Cho-containing compounds, contributing to the Cho peak in ^{1}H-MRS, showed no changes in its cerebellar concentration throughout the studied late fetal and newborn period. The slight, nonsignificant decrease toward adult levels has to be studied further.

Unlike *in vitro* animal studies that showed increasing concentrations of *myo*-inositol during brain development (5), we found stable values for Ino in human brain development. If this can be further confirmed, Ino might serve as a reliable internal standard for ^{1}H-MRS during development.

The possibility of measuring these metabolites noninvasively from small localized brain volumes allows us to obtain previously inaccessible information on *in vivo* age-dependent regional brain biochemistry. These results are a basis for the further understanding of normal and abnormal brain development.

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REFERENCES

1. Radda GK, Rajagopalan B, Taylor DJ 1989 Biochemistry *in vivo*: an appraisal of clinical magnetic resonance spectroscopy. Magn Reson Q 5:122–151
2. Petroff OAC 1988 Biological ^{1}H NMR spectroscopy. Comp Biochem Physiol 90B:249–260
3. Barany M, Langer BG, Glick RP, Venkatasubramanian PN, Wilbur AC, Spigos

DG 1988 *In vivo* ^{1}H spectroscopy in humans at 1.5T. Radiology 167:839–844
4. Arus C, Chang Y, Barany M 1985 Proton nuclear magnetic resonance spectra of excised rat brain. Assignment of resonances. Phys Chem Phys Med NMR 17:23–33
5. Cerdan S, Parrilla R, Santoro J, Rico M 1985 ^{1}H NMR detection of cerebral *myo*-inositol. FEBS Lett 187:167–172
6. Behar KL, Den Hollander JA, Srtomski ME, Ogino T, Shulman RG, Petroff OAC, Prichard JW 1983 High-resolution ^{1}H nuclear magnetic resonance study of cerebral hypoxia *in vivo*. Proc Natl Acad Sci USA 80:4945–4948
7. Avison MJ, Herschkowitz N, Novotny EJ, Petroff OAC, Rothman DL, Colombo JP, Bachmann C, Shulman RG, Prichard JW 1990 Proton NMR observation of phenylalanine and aromatic metabolite in the rabbit brain *in vivo*. Pediatr Res 27:566–570
8. Luyten PR, den Hollander JA 1986 Observation of metabolites in the human brain by MR-spectroscopy. Radiology 161:795–798
9. Barany M, Spigos DG, Mok E, Venkatasubramanian PN, Wilbur AC, Langer BG 1987 High resolution proton magnetic resonance spectroscopy of human brain and liver. Magn Reson Imaging 5:393–398
10. Frahm J, Bruhn H, Gyngell ML, Merboldt KD, Hänicke W, Sauter R 1989 Localized high-resolution proton NMR spectroscopy using stimulated echoes: initial applications to human brain *in vivo*. Magn Reson Med 9:79–93
11. Van der Knaap MS, Van der Grond J, Van Rijen PC, Faber JAJ, Valk J, Willemse K 1990 Age-dependent changes in localized proton and phosphorus MR spectroscopy of the brain. Radiology 176:509–515
12. Peden CJ, Cowan FM, Bryant DJ, Sargentoni J, Cox J, Menon DK, Gadian DG, Bell JD, Dubowitz LM 1990 Proton spectroscopy of the brain in infants. J Comput Assist Tomogr 14:886–894
13. Verbitskaya LB 1969 Some aspects of the ontophylogenesis of the cerebellum. In: Linas R (ed) Neurobiology of the Cerebellar Evolution and Development. American Medicine Association, Chicago, pp 859–874
14. Posse S, Lazeyras F, Schuknecht B, Herschkowitz N, Moonen CTW 1990 Brain tissue differentiation using short time proton spectroscopic imaging. Society of Magnetic Resonance in Medicine 9th Annual Scientific Meeting, August 1990, New York, abstr 1210
15. Burri R, Bigler P, Straehl P, Posse S, Colombo JP, Herschkowitz N 1990 Brain development: ^{1}H-magnetic resonance spectroscopy of rat brain extracts compared with chromatographic methods. Neurochem Res 15:1009–1016
16. Jüngling E, Kammermaier H 1980 Rapid assay of adenine nucleotides or creatinine compounds in extracts of cardiac tissue by paired-ion reverse-phase high performance liquid chromatography. Anal Biochem 102:358–361
17. Petroff OAC, Ogino T, Alger JR 1988 High resolution proton magnetic resonance spectroscopy of rabbit brain: regional metabolite levels and postmortem changes. J Neurochem 51:163–171
18. Agrawal HC, Davis JM, Himwich WA 1966 Postnatal changes in free amino acid pool of rabbit brain. Brain Res 3:374–380
19. Perry TL, Hansen S, Gandham SS 1981 Postmortem changes of amino compounds in human and rat brain. J Neurochem 36:406–412
20. Petroff OAC, Prichard JW, Ogino T, Shulman RG 1988 Proton magnetic resonance spectroscopic studies of agonal carbohydrate metabolism in rabbit brain. Neurology 38:1569–1574
21. Tallan HH 1957 Studies on the distribution of N-acetyl-L-aspartic acid in brain. J Biol Chem 224:41–45
22. Lowry OH, Berger SJ, Chi MMY, Carter JG, Blackshaw A, Outlaw W 1977 Diversity of metabolic patterns in human brain tumors—I. High energy phosphate compounds and basic composition. J Neurochem 29:959–977
23. Frahm J, Bruhn H, Gyngell ML, Merboldt KD, Hänicke W, Sauter R 1989 Localized proton NMR spectroscopy in different regions of the human brain *in vivo*. Relaxation times and concentrations of cerebral metabolites. Magn Reson Med 11:47–63
24. Frahm J, Michaelis T, Merboldt K, Hänicke W, Gyngell ML, Chien D, Bruhn H 1989 Localized NMR spectroscopy *in vivo*. NMR Biomed 2:188–195
25. Petroff OAC, Spencer DD, Alger JR, Prichard JW 1989 High-field proton magnetic resonance spectroscopy of human cerebrum obtained during surgery for epilepsy. Neurology 39:1197–1202
26. Nakano C, Takashima S, Takeshita K 1989 Carnitine concentration during the development of human tissues. Early Hum Dev 19:21–27
27. Leray C, Pelletier A, Massarelli R, Dreyfus H, Freysz L 1990 Molecular species of choline and ethanolamine phospholipids in rat cerebellum during development. J Neurochem 54:1677–1681
28. Michaelis T, Merboldt KD, Hänicke W, Gyngell ML, Bruhn H, Frahm J 1991 On the identification of cerebral metabolites in localized ^{1}H NMR spectra of human brain *in vivo*. NMR Biomed 4:90–98
29. Miyake M, Kakimoto Y, Sorimachi M 1981 A gas chromatographic method for the determination of N-acetyl-L-aspartic acid, N-acetyl-a-aspartylglutamic acid and b-citruyl-L-glutamic acid and their distributions in the brain and other organs of various species of animals. J Neurochem 36:804–810
30. Bates TE, Williams SR, Gadian DG, Bell JD, Small RK, Iles RA 1989 ^{1}H NMR study of cerebral development in rat. NMR Biomed 2:225–229
31. Birkin DL, Oldendorf WH 1989 N-acetyl-L-aspartic acid: a literature review of a compound prominent in ^{1}H-NMR-spectroscopic studies of brain. Neurosci Biobehav Rev 13:23–31
32. D'Adamo AF, Yatsu FM 1966 Acetate metabolism in the nervous system. N-acetyl-L-aspartic acid and the biosynthesis of brain lipids. J Neurochem 13:961–965
33. Burri R, Steffen C, Herschkowitz N 1991 N-acetyl-aspartate is a major source of acetyl groups for lipid synthesis during rat brain development. Dev Neurosci (in press)
34. Clarke D, Greenfield S, Dicker E, Tirri LJ, Ronan EJ 1975 A relationship of N-acetyl-aspartate biosynthesis to neuronal protein synthesis. J Neurochem 24:479–485
35. Cangro CB, Namboodiri MAA, Sklar LA, Coigliano-Murphy A, Neale JH 1987 Immunohistochemistry and the biosynthesis of N-acetylaspartylglutamate in spinal sensory ganglia. J Neurochem 49:1579–1588
36. Gill SS, Thomas DGT, Van Bruggen N, Gadian DG, Peden CJ, Bell JD, Menon DK, Iles RA, Bryant DJ, Coutts GA 1990 Proton MR spectroscopy of intracranial tumours: *in vivo* and *in vitro* studies. J Comput Assist Tomogr 14:497–504
37. Ott D, Ernst H, Ernst T 1990 *In vivo* ^{1}H spectroscopy of myelin disorders. Society of Magnetic Resonance in Medicine, 9th Annual Scientific Meeting, August 1990, New York, abstr 1012
38. Okumura N, Otsuki S, Kameyama A 1960 Studies on free amino acids in human brain. J Biochem 47:315–320
39. Sturman JA, Moretz RC, French JH, Nisniewski HM 1985 Taurine deficiency in the developing cat: persistence of the cerebellar external granule layer. J Neurosci Res 13:405–416
40. Sturman JA, Moretz RC, French JH, Nisniewski HM 1985 Postnatal taurine deficiency in the kitten results in a persistence of the cerebellar external granule cell layer. J Neurosci Res 13:521–528
41. Herschkowitz N 1988 Brain development in the fetus, neonate and infant. Biol Neonate 54:1–19
42. Huxtable RJ 1981 Insights on function: metabolism and pharmacology of taurine on the brain. In: Lombardini JB, Kenny AD (eds) The Role of Peptides and Amino Acids as Neurotransmitters. Alan R Liss, New York, pp 53–97
43. Gaull GE 1989 Taurine in pediatric nutrition: review and update. Pediatrics 83:433–442