Neuroradiology (2005) 47: 562–565
DOI 10.1007/s00234-005-1387-3

K. Sarac
A. Akinci
A. Alkan
M. Aslan
T. Baysal
C. Özcan

Brain metabolite changes on proton magnetic resonance spectroscopy in children with poorly controlled type 1 diabetes mellitus

Received: 27 October 2004
Accepted: 4 February 2005
Published online: 23 June 2005

K. Sarac (✉) · A. Alkan · T. Baysal
Department of Radiology,
Inonu University School of Medicine,
Turgut Özal Tip Merkezi, Malatya, Turkey
E-mail: ksarac@inonu.edu.tr
Fax: +90-422-3410787

A. Akinci · M. Aslan
Department of Paediatric Endocrinology,
Inonu University School of Medicine,
Malatya, Turkey

C. Özcan
Department of Neurology,
Inonu University School of Medicine,
Malatya, Turkey

Abstract The metabolite changes in the brains of children with poorly controlled type 1 diabetes mellitus (DM) were investigated by proton magnetic resonance spectroscopy (MRS). A total of 30 subjects and 14 age-matched healthy volunteers underwent single-voxel MRS (TE: 136). The duration of disease, medication, presence of hypoglycaemia episodes and the level of haemoglobin A1C (HbA1C) in the patients were noted. Voxels were placed in the pons, left basal ganglion (LBG) and left posterior parietal white matter (PPWM). *N*-acetylaspartate (NAA)/creatinine (Cr) and choline (Cho)/Cr ratios were calculated. The average HbA1c level was 11.9 ± 3.4 (8.2–19.4). The average number of keto-acidosis episodes was 1.9 ± 2.2 (0–9) and the average number of daily insulin injections was 2.8 ± 0.97 (2–4). MRS revealed lower NAA/Cr and Cho/Cr ratios in the pons and lower NAA/Cr ratio in the PPWM of patients with DM than in control subjects. No significant correlation was observed between the number of hypoglycaemia episodes and metabolite ratios. Metabolic abnormalities have been observed by MRS in the brain of poorly controlled type 1 DM children. These metabolic changes, in particular in the pons region, include a decrease in NAA, indicating neuronal loss or functional impairment, and likely explanations for a decrease in Cho may be dynamic changes in membrane lipids and/or decreased membrane turnover.

Keywords Type 1 diabetes mellitus · Brain · MR spectroscopy

Introduction

Type 1 diabetes mellitus (DM) is a complex metabolic disorder and is often associated with complications secondary to central nervous system involvement [1, 2]. Other than acute changes that developed during hypoglycaemia or hyperglycaemia episodes, there is long-term impairment of cognitive functions in type 1 DM patients with poor gylcaemic control [3–5]. These cerebral complications may be referred to as ''diabetic encephalopathy'', which is diagnosed by clinical, psychological and electrophysiological tests [6]. The underlying mechanisms causing brain damage in diabetes are not clear. Experimental studies showed that diabetic rodents develop cerebral deficits similar to those observed in diabetic patients [7]. Fluctuations in blood glucose level secondary to exogenous insulin use, and acute and/or chronic metabolic and vascular impairments, may cause functional and structural changes in diabetic patients [6, 8].

In vivo proton magnetic resonance spectroscopy (MRS) is a non-invasive imaging method for monitoring chemical and metabolic changes in areas of interest within the brain. MRS can provide information about

the concentration and relative levels of proton-including metabolites and may assist in discriminating normal and pathological tissues. Although several articles have appeared on the use of the proton MRS in different brain disorders recently, brain MRS data for DM is still limited [9–11]. In the present study, we report our MR spectroscopic data of the brain to study metabolic disturbances in a childhood population with poorly controlled type 1 DM.

Methods

We performed MR spectroscopic analysis on 30 patients with type 1 DM. Mean age of the patients was 13.1 ± 3.5 years (range 8–19 years). Mean follow-up period was 58 ± 42.6 months (range 3–186 months). We noted the duration of disease, medication, presence of hypoglycaemia episodes and the level of haemoglobin A1C (HbA1C). Fourteen patients had diabetes-related retinopathy, whereas the other 16 patients had no complications except elevated HbA1C blood levels. The control group included volunteer, healthy subjects who were admitted to healthy child polyclinics. Fourteen apparently healthy subjects (mean age 12.1 ± 3.3 years, range 8–20 years) without known neurological or biochemical disease, who were approximately age- and gender-matched to the group of patients with DM, were selected. The healthy subjects underwent identical examination by means of proton MRS. The patients and their families gave informed consent to participate in the study.

MRS was carried out on a 1.5-T whole-body imager (Philips, Gyroscan Intera Master, Best, The Netherlands), using a combined magnetic resonance imaging (MRI) and spectroscopic imaging protocol. We used a transit/receive quadrature birdcage head coil. The protocol consisted of axial and sagittal T1-weighted images (TR: 560, TE: 15), sagittal, coronal and axial T2-weighted images (TR: 4,530, TE: 100), plus MRS. The routine images were used to identify anatomical structures and to confirm the absence of any structural or signal abnormality. In the same session, the pons, left basal ganglion (LBG) and left posterior parietal white matter (PPWM) were studied in patients and control subjects. Single-voxel spectroscopy was performed on all patients using a point-resolved spectroscopy sequence (TR: 2,000/TE: 136 ms) with 128 averages. Voxels were placed to the locations in the pons, LBG and PPWM. Voxel sizes were selected as 15×15×15 mm in the LBG and PPWM, 13×13×13 mm in the pons. Prior to MRS, shimming was performed to optimize field homogeneity, and water suppression was optimized with automated routines provided by the manufacturer. The water signal was suppressed by a chemical-shift selective saturation pulse. A spectral sweep width of 1,000 Hz was used, with a data size of 1,024 points. The magnitude spectra were processed automatically by baseline correction and curve-fitting procedures to determine the resonance areas of *N*-acetylaspartate (NAA), creatine (Cr) and choline (Cho). Analysis of the spectra was performed with the manufacturer's supplied spectroscopy software package of the MRI system. Resonances were assigned as follows: NAA, 2.0 ppm; Cr 3.02 ppm; Cho 3.2 ppm. Peak area metabolite ratios (NAA/Cr and Cho/Cr,) were calculated. For each patient, two authors assessed whether the spectra were diagnostic.

All statistical analyses were performed with a commercially available SPSS release 10.0 software package (SPSS, Chicago, Ill., USA). That was the rational for the application of the non-parametric Mann–Whitney *U*-test to assess the metabolite differences among these groups. A P value of <0.05 was considered to be significant.

Results

No significant differences between the mean ages of the groups were observed. Neurological examination of the patients was within normal limits. The patients had a history of DM for 2–8 years. The average HbA1c level was 11.9 ± 3.4 (8.2–19.4). The average number of ketoacidosis episodes was 1.9 ± 2.2 (0–9) and the average number of daily insulin injections was 2.8 ± 0.97 (2–4). MRI of the patients dnnot reveal any anatomical or morphological abnormalities. In the 44 subjects, a total of 132 spectra were sampled. Nine spectra were rejected due to insufficient resolution. Cerebral metabolite concentration and ratios are given in Table 1. MRS revealed lower NAA/Cr ratio in the PPWM and lower NAA/Cr

Table 1 The mean values of NAA/Cr and Cho/Cr in the pons, left basal ganglion and posterior parietal white matter in the patient and control groups (*NS* not significant.)

Location	Ratio	Patient group	Control group	*P*
Pons	NAA/Cr	1.77 ± 0.90	2.54 ± 1.23	0.005
	Cho/Cr	1.53 ± 0.77	2.01 ± 0.63	0.006
LBG	NAA/Cr	1.23 ± 0.20	1.28 ± 0.13	NS
	Cho/Cr	0.89 ± 0.21	0.82 ± 0.15	NS
PPWM	NAA/Cr	1.99 ± 0.53	2.42 ± 0.61	0.04
	Cho/Cr	1.07 ± 0.36	1.31 ± 0.43	NS

and Cho/Cr ratio in the pons of DM patients than in control subjects. No significant correlation was observed between number of hypoglycaemia episodes and metabolite ratios.

Discussion

9The pathogenesis of cerebral dysfunction shown by neuroradiological, electrophysiological and cognitive tests in DM seems to be a multifactorial process [8]. In poorly controlled DM, exposure to the extremes of blood glucose levels, chronic hyperglycaemia and repeated episodes of severe hypoglycaemia may adversely affect the brain [6, 12]. While acute hypoglycaemia alters regional cerebral blood flow and leads to microvascular changes, the mechanisms of hyperglycaemia are likely to be complex interactions of hyperglycaemia, ketonaemia, acidosis and hyperosmolality. During hyperglycaemia, enhanced formation of oxygen free radicals occurs in the tissues. These oxidant radicals contribute to increased neuronal death by oxidizing proteins, damaging DNA and inducing the lipoperoxidation of cellular membranes. Neuronal abnormalities may also occur secondary to a possible marginal deficiency of long-chain polyunsaturated fatty acids [13–15].

Neuroimaging techniques, such as MRI and computed tomography, have provided data on structural changes in the brain that may be more relevant to the general population of the diabetic patients [8]. MRS may demonstrate metabolic changes in normal-appearing MRI examinations. The prominent resonances detected on MRS in normal brains include NAA, Cho and Cr. Cr participates in cerebral energy metabolism and reserves. Since Cr is relatively constant throughout the normal brain tissue and in different pathological conditions, it is often used as a reference resonance for the measurement of relative changes in NAA or Cho or both. NAA is the most sensitive central nervous system metabolite. Since it is a neuro-axonal marker, abnormalities of neuronal structures, such as reduced neuronal density or viability, lead to the reduction of NAA [16]. More recently, reduction of cerebral NAA, however, was shown to be potentially reversible, indicating that NAA levels not only reflect neuronal density and viability but can also be used as a dynamic marker of neuronal metabolic dysfunction and integrity [17]. Only limited proton MR spectroscopic data are currently available for DM. Kreis and Ross [10] showed a significant reduction of *N*-acetyl metabolites in the parietal white matter, but no change in the occipital cortex, in subacute and chronic DM patients. Lai et al. [18] revealed NAA decreases in the basal ganglia of hyperglycaemic diabetic patients with chorea-ballismus. Biessels et al. [9] found a reduction in NAA/Cr ratios in streptozotocin-diabetic rats. In the present study, we found a decrease in NAA/Cr in the PPWM, and particularly in the pons, in the patients with poorly controlled type I DM. This finding can be attributed to neuronal loss or neuronal dysfunction in children with type 1 DM.

Cho/Cr was the other important metabolite ratio that was analysed. Choline includes metabolites involved in the composition of myelin, such as phosphorylcholine, glycerophosphorylcholine and a relatively negligible amount of free choline [16, 19]. It can be viewed as an indirect marker of myelination and cell membrane metabolism. An increase in the Cho peak is associated with conditions such as brain tumours and demyelinating disease [20]. Decreased concentration of Cho may be due to changes in dynamic behaviour of the membrane lipids and/or decreased membrane turnover [21, 22]. Biessels et al. [9] found no differences of Cho/Cr ratios in streptozotocin-diabetic rats. Kreis and Ross [10] showed that Cho was, on average, unchanged, but that there were large increases in some individuals, especially in diabetic keto-acidosis and hyperglycaemic coma. They concluded that there is a correlation between hyperosmolality and choline, which is an osmolite substance. Also, they reported that a 10% higher level of Cho in both white and grey matter characterized the small group of patients with type 2 DM compared with patients with type 1 DM [10]. In another study, diabetic patients with non-ketotic hyperglycaemia and chorea-ballismus showed increased Cho levels in the basal ganglia [18]. In our study, Cho/Cr was found to be normal in other brain regions, while there was a significant Cho/Cr decrease in the pons region. The discrepancy between previous studies and our study may be explained by difference in diabetic populations and blood sugar levels. The likely explanations for a decrease in Cho may be dynamic changes in membrane lipids and/or decreased membrane turnover.

Lactate is usually detected only under pathological condition, when energy metabolism is affected severely and it could indicate anaerobic metabolism [19, 20]. With ^{31}P and ^{1}H MRS studies, Biessels et al. [9] reported that diabetes did not cause alterations in cerebral energy metabolism in streptozotocin-diabetic rats. To the contrary, Lai et al. [18] reported a lactate peak in the basal ganglia of diabetic hyperglycaemic patients with chorea-ballismus in the acute stage. However, in our study, we did not detect lactate. The possible explanation for this may be absence of acute stage of keto-acidosis in our patients.

To our knowledge, there is no MRS study of the pons region in DM patients. In type 2 DM, pontine lesions and cranial nerve involvement have been attributed to an ischaemic lesion in this region [23]. Electrophysiological studies on type 1 DM patients depict pons and midbrain involvement [24, 25]. Pontine involvement that we have shown by MRS may explain this electrophysiologically proven disorder.

In DM patients, the effect of recurrent severe hypoglycaemia episodes on cognitive function was investigated but a correlation could not be found [26, 27]. Perros at al. [11] found no differences in metabolite ratios in DM patients with and without a history of recurrent severe hypoglycaemia. Similarly, we did not find a relationship between hypoglycaemia and metabolite ratios in DM.

In conclusion, type 1 DM causes metabolic changes in the brain that can be shown by MRS. These metabolic disorders, in particular in the pons region, include a decrease in NAA, indicating neuronal loss or functional impairment, and a decrease in Cho, indicating dynamic changes in membrane lipids and/or decreased membrane turnover. Further studies with follow-up are needed to determine the value of MRS in DM.

References

1. Harati Y (1996) Diabetes and nervous system. Endocrinol Metab Clin North Am 25:325–359
2. Gispen WH, Biessels GJ (2000) Cognition and synaptic plasticity in diabetes mellitus. Trends Neurosci 23:542–549
3. Franceschi M, Cecchetto R, Minicucci F, Smizne S, Baio G, Canal N (1984) Cognitive processes in insulin-dependent diabetes. Diabetes Care 7:228–231
4. McCarthy AM, Lindgren S, Mengeling MA, Tsalikian E, Engvall JC (2002) Effects of diabetes on learning in children. Pediatrics 109:E9
5. Widom B, Simonson DC (1990) Glycemic control and neuropsychologic function during hypoglycemia in patients with insulin-dependent diabetes mellitus. Ann Intern Med 112:904–912
6. Brands AM, Kessels RP, de Haan EH, Kappelle LJ, Biessels GJ (2004) Cerebral dysfunction in type 1 diabetes: effects of insulin, vascular risk factors and blood-glucose levels. Eur J Pharmacol 490:159–168
7. Biessels GJ, Cristino NA, Rutten GJ, Hamers FP, Erkelens DW, Gispen WH (1999) Neurophysiological changes in the central and peripheral nervous system of streptozotocin-diabetic rats. Course of development and effects of insulin treatment. Brain 122:757–768
8. Biessels GJ, van der Heide LP, Kamal A, Bleys RL, Gispen WH (2002) Ageing and diabetes: implications for brain function. Eur J Pharmacol 441:1–14
9. Biessels GJ, Braun KP, de Graaf RA, van Eijsden P, Gispen WH, Nicolay K (2001) Cerebral metabolism in streptozotocin-diabetic rats: an in vivo magnetic resonance spectroscopy study. Diabetologia 44:346–353
10. Kreis R, Ross BD (1992) Cerebral metabolic disturbances in patients with subacute and chronic diabetes mellitus: detection with proton MR spectroscopy. Radiology 184:123–130
11. Perros P, Deary IJ, Sellar RJ, Best JJ, Frier BM (1997) Brain abnormalities demonstrated by magnetic resonance imaging in adult IDDM patients with and without a history of recurrent severe hypoglycemia. Diabetes Care 20:1013–1018
12. Biessels GJ, Kappelle AC, Bravenboer B, Erkelens DW, Gispen WH (1994) Cerebral function in diabetes mellitus. Diabetologia 37:643–650
13. Tallroth G, Ryding E, Agardh CD (1993) The influence of hypoglycaemia on regional cerebral blood volume in type 1 (insulin-dependent) diabetes mellitus. Diabetologia 36:530–535
14. Vincent AM, Brownlee M, Russell JW (2002) Oxidative stress and programmed cell death in diabetic neuropathy. Ann N Y Acad Sci 959:368–383
15. Kumar JS, Menon VP (1993) Effect of diabetes on levels of lipid peroxides and glycolipids in rat brain. Metabolism 42:1435–1439
16. Miller BL (1991) A review of chemical issues in 1H NMR spectroscopy: *N*-acetyl-L-aspartate, creatine and choline. NMR Biomed 4:47–52
17. Demougeot C, Garnier P, Mossiat C, Bertrand N, Giroud M, Beley A, Marie C (2001) *N*-acetylaspartate, a marker of both cellular dysfunction and neuronal loss: its relevance to studies of acute brain injury. J Neurochem 77:408–415
18. Lai PH, Chen PC, Chang MH, Pan HB, Yang CF, Wu MT, Li JY, Chen C, Liang HL, Chen WL (2001) In vivo proton MR spectroscopy of chorea-ballismus in diabetes mellitus. Neuroradiology 43:525–531
19. Cox IJ (1996) Development and applications of in vivo clinical magnetic resonance spectroscopy. Prog Biophys Mol Biol 65:45–81
20. Ross B, Michaelis T (1994) Clinical applications of magnetic resonance spectroscopy. Magn Reson Q 10:191–247
21. Jagannathan NR, Tandon N, Raghunathan P, Kochupillai N (1998) Reversal of abnormalities of myelination by thyroxine therapy in congenital hypothyroidism: localized in vivo proton magnetic resonance spectroscopy (MRS) study. Brain Res Dev Brain Res 109:179–186
22. Bhatara VS, Tripathi RP, Sankar R, Gupta A, Khushu S (1998) Frontal lobe proton magnetic-resonance spectroscopy in Graves' disease: a pilot study. Psychoneuroendocrinology 23:605–612
23. Thomke F, Urban PP, Marx JJ, Mika-Gruttner A, Hopf HC (2002) Seventh nerve palsies may be the only clinical sign of small pontine infarctions in diabetic and hypertensive patients. J Neurol 249:1556–1562
24. Khardori R, Soler NG, Good DC, DevlescHoward AB, Broughton D, Walbert J (1986) Brainstem auditory and visual evoked potentials in type 1 (insulin-dependent) diabetic patients. Diabetologia 29:362–365
25. Toth F, Varkonyi TT, Rovo L, Lengyel C, Legrady P, Jori J, Czigner J, Kiss JG (2003) Investigation of auditory brainstem function in diabetic patients. Int Tinnitus J 9:84–86
26. Ferguson SC, Blane A, Perros P, McCrimmon RJ, Best JJ, Wardlaw J, Deary IJ, Frier BM (2003) Cognitive ability and brain structure in type 1 diabetes: relation to microangiopathy and preceding severe hypoglycemia. Diabetes 52:149–156
27. The Diabetes Control and Complications Trial Research Group (1993) The effect of intensive treatment of diabetes on the development and progression of the long-term complications in insulin-dependent diabetes mellitus. The Diabetes Control and Complications Trial Research Group. N Engl J Med 329:977–986