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24 month-treatment with miglustat of three patients with Niemann-Pick disease type C: Follow up using brain spectroscopy

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ABSTRACT

Niemann-Pick C (NPC) is a fatal progressive neurolipidosis. Miglustat, an inhibitor of glycosphingolipid synthesis, has been proposed to treat patients but questions remain regarding its efficacy. A major problem has been the lack of suitable objective efficacy endpoints. Three adults with NPC were treated with miglustat for 24 months. Efficacy of treatment was assessed clinically and using brain magnetic resonance spectroscopy. All patients reported mild clinical improvement or stabilization. Furthermore, a sustained decrease in the choline/creatine ratio was observed in all three patients over time. Although these preliminary results require confirmation on a larger cohort of patients, they suggest that miglustat has some beneficial effect on brain dysfunction in NPC and that MRS could be used routinely as a non invasive surrogate marker of treatment efficacy.

Introduction

Niemann-Pick disease type C (NPC) is a progressive neurological lysosomal storage disease of autosomal recessive inheritance. NPC arises from mutations in two genes, NPC1 (95% of patients) and NPC2, that play a role in intracellular cholesterol and glycolipid trafficking [1]. While large amounts of free cholesterol accumulate in peripheral organs, the most conspicuous storage in brain concerns glycosphingolipids, essentially GM2 and GM3 gangliosides. Mechanisms by which gangliosides accumulate are probably multiple and the relationship between glycolipid storage and abnormalities in cholesterol transport are still controversial. Neurological signs of NPC arise both from neuronal loss and neuronal dysfunction [2]. While symptoms related to cell loss are less likely to be improved by treatments, restoring cellular homeostasis by any therapeutic approach could potentially reverse cellular dysfunction and then provide clinical stabilization or improvement.

Adult forms of NPC are characterized by psychiatric disorders, vertical supranuclear gaze palsy, cerebellar ataxia, cognitive deficits, movement disorders, gelastic cataplexy and hepatosplenomegaly [3]. Death occurs on average 12 years after the onset of psychiatric or neurological signs. *N*-butyl-deoxynojirimycin(NB-DNJ, OGT 918, miglustat) is an inhibitor of glucosylceramide synthetase, currently approved for the treatment of Gaucher type I disease [4]. It also inhibits the biosynthesis of all glycolipids derived from the glucosylceramide precursor, among which most gangliosides. Consequently, this compound has been tested in animal models of NPC: treatment with miglustat delayed onset of neurological symptoms, prolonged survival and improved brain neuropathology [5]. However, in humans, results are equivocal. Several case reports described some stabilization or improvement in adult patients but not in children [6–8]: as in Gaucher disease type II, early onset forms of NPC are usually more severe, rapidly

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progress and are therefore less accessible to treatments. A recent open-randomized controlled trial in patients with a late onset disease, reported stabilization or improvement of horizontal saccade velocity, swallowing and ambulatory index, however, statistical difference was weak [9].

A major problem in previous trials with miglustat in NPC has been the lack of quantifiable objective markers that could be predictive of clinical efficacy. Neurological examination can be quantified through clinical scales but these are not fully appropriate to NPC: given the slow mechanism of action of miglustat, as observed in Gaucher disease [10] and given the probable irreversible neuronal damage, a rapid clinical improvement is not expected.

Here we report the follow up with MRS of three patients treated with miglustat for 24 months. Interestingly, we observed a decrease of the choline/creatine ratio in all three patients.

Methods

From March 2006 to December 2008, nine patients with NPC were treated with miglustat off license in France. Patients who accessed to treatment had relative late-onset-diseases (juvenile or adult forms) and were relatively autonomous i.e., without gastric tube, dementia or severe psychiatric problems. Follow up with serial brain MRS was obtained in three patients followed at the Pitié-Salpêtrière Hospital (Paris). Only these three patients are reported here. All patients were treated initially with a dose of 600 mg/day that was decreased to 300 mg after 3–6 months in two patients (patients 1 and 3) because of adverse effects (severe weight loss in patient 1, increased tremor in patient 3). Clinical records were performed by the same investigator (FS). The clinical disability scale was calculated as previously reported [11]: ambulation score (A) varies from 1 (normal) to 5 (Wheelchair bond); manipulation (M) from 1 (normal) to 4 (requires assistance in all activities); language (L) from 1 (normal) to 5 (absence of communication); swallowing (S) from 1 (normal) to 4 (nasogastric tube or gastric button).

Magnetic resonance (MR) imaging and spectroscopy were performed on a 1.5 MR unit (General Electric, WI) and included T1, FLAIR, T2-weighted sequences, and single voxel MRS acquisitions at long echo time (TR = 1500 ms, TE = 135 ms) in the white matter (centrum ovale or semi ovale), basal ganglia and cerebellum. Because of variability of the quality of spectra in these two last regions, only results in the white matter could be reliably analysed. Resonance of choline (Cho), creatine (Cr), *N*-acetyl aspartate (NAA) as well as Cho/Cr and NAA/Cr ratios were automatically quantified by the software provided by the manufacturer (Probe Q, General Electric Medical Systems, Milwaukee, WI).

Results

Clinical data

The clinical, biological and genetic characteristics of the three patients have been previously reported [3]. Patients 1–3 in the present study correspond to cases 11, 13 and 7, respectively, in Sevin et al. [3]. Main clinical characteristics, genotypes, and clinical evolution under treatment with miglustat are summarized in Table 1. Clinical evolution under treatment with miglustat is summarized in Table 2. In brief, after 24 months of treatment, all three patients exhibited mild improvement or stabilization regarding swallowing, dysarthria, awareness or ambulation. Improvement was the most obvious in patient 1: after 18 months, this patient did not present any paranoid delusion (acute episodes of delusion occurred around twice a year before treatment, and she had mild paranoid delusion in between these attacks), she did not fall anymore (falls were frequent before treatment) and she did not present any dysphagia whereas dysphagia occurred once daily before treatment. Patient 2 remained essentially stable after 12 months (last follow up) with some improvement of dysphagia: dysphagia occurred at least three times a week before treatment as compared to once a month after treatment. Clinical improvement was more difficult to establish in patient 3: although this patient reported some improvement of dysarthria and attention after 18 months, his gait worsened during the same period of time (Table 1).

MRI and MR spectroscopy (MRS)

MRS and MRI were obtained at baseline and after 12 (M12), 18 (M18) and 24 (M24) months in all patients except in patient 2 for whom the MRS at M18 could not be performed.

Table 1
Characteristics of patients at baseline and clinical evolution with treatment.

Cases	Gender	Current age (years)	Age at neurological onset (y)	Genotype	Major Clinical signs at treatment onset	Dosage of miglustat	clinical evolution at last follow up	Adverse effects	Disability scale[a] at baseline (total)	Disability scale[a] at month 24 (total)
1	F	23	19	p.V950M/ p.I1061T	Psy, Cog, At, Dys, VSO, Sm	200 mg tid M0 to M6 100 mg tid M6 to M24	Improvement of walking, swallowing, dysarthria and awareness	Weight loss, diarrhea	A3 M2 L2 S3 (10/18)	A2 M2 L2 S2 (8/18)
2	M	21	17	p.I1061T/ p.G538R	Cog, Dys, VSO, Hea, Sm	200 mg tid M0 to M24	Slight improvement of swallowing and dysarthia	None	A2 M2 L2 S3 (9/18)	A2 M2 L2 S2 (8/18)
3	M	38	16	p.G992R/ p.N452fs	Cog, Md, At, VSO, Sm	200 mg tid M0 to M3 100 mg tid M3 to M24	Improvement of dysarthria and awareness. worsening of gait	Worsening of myoclonus reversible after decreased dosage	A4 M4 2 S2 (12/18)	A5 M4 L2 S2 (13/18)

Abbreviations: At, ataxia; Cog, cognitive disorder (excluding psychiatric symptoms); Dys, dystonia, F, female; Hea, hearing loss; M, male; Psy, psychiatric signs; Sm, splenomegaly; VSO, vertical supra nuclear ophthalmoplegia.
[a] See the methods section.

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Table 2
Measurements of main metabolic peak ratios in the centrum semi ovale in three patients treated with miglustat for 24 months.

Patients	Cho/Cr				NAA/Cr			
	Baseline	M12	M18	M24	Baseline	M12	M18	M24
Patient 1	1.2414	1.2609	0.9383	0.762	2.069	2.304	2.136	2.143
Patient 2	1.4167	1.2466	NA	1.0545	2	1.973	NA	1.982
Patient 3	1.2759	1.27	1.0727	1.12	2.3	2.26	2.127	2.576
Mean values	1.31	1.26	1.01	0.98	2.12	2.18	2.13	2.23

M12, M18, M24 values after 12, 18 and 24 months of treatment with miglustat, NA, non available. Cho/Cr and NAA/Cr ratios were calculated automatically from the raw data using the Probe Q software. Position of the voxel of interest is shown in Fig. 1.

Brain MRI at baseline did not show any abnormalities in patients 1 and 2 but showed cerebellar atrophy in patient 3 (not shown). The MRS Choline (Cho)/creatine (Cr) and Nacetyl aspartate (NAA)/Cr were automatically calculated in the brain white matter (centrum ovale or semiovale). Positions of the voxels and quality of spectra are shown in Fig. 1. Cho is considered as a marker of membrane destruction or gliosis whereas NAA is believed to reflect neuronal viability. In all patients, MRS follow up during the first year of treatment (M12) showed an initial stabilization of these ratios (Table 2 and Fig. 1). However, after 18 and 24 months, a decrease in the Cho/Cr ratio was observed in all patients (Table 2 and Fig. 1). More precisely, in patient 1, the Cho/Cr ratio dropped from 1.24 at baseline to 0.94 after 18 months and 0.76 after 24 months. In patient 2; this ratio dropped from 1.42 at baseline to 1.05 after 24 months and in patient 3 it dropped from 1.28 at baseline to 1.07 after 18 months and 1.12 after 24 months. Overall, the mean Cho/Cr value, dropped from 1.31 at baseline to 1.01 after 18 months and 0.98 after 24 months. Cho/Cr values after 24 months of treatment were close to values usually obtained in control subjects in the brain white matter that is around 1.1 [12, our unpublished data]. During the same period, the mean NAA/Cr ratio remained stable (Table 2 and Fig. 1).

Discussion

We observed a sustained decrease in the Cho/Cr ratio in three NPC patients treated with miglustat for up to 24 months. This ratio decreased after 18 months of treatment and was observed whatever the dose of miglustat (600 mg vs 300 mg/day). Since Cho/Cr is considered as a marker of brain dysfunction, it is suggested that miglustat has a beneficial effect over time. Interestingly, the same effect was observed in patient 3 who had 22 years of symptomatic disease versus 4 years in patients 1 and 2 suggesting that even in advanced stages, some brain dysfunction may still be reversible.

This study has important methodological limitations.

First, the small number of patients did not allow valid statistical analysis. It is assumed that all results reported here need to be confirmed in a larger cohort of patients.

Second, this is an uncontrolled study. We did not perform sequential brain MRI in untreated NPC patients so that we do not have a precise idea of the natural evolution of MRS in NPC. In the worst case, the change in Cho/Cr ratios observed in our patients could simply represent progressive changes in the white matter of patients as getting older and sicker over a 2-year period. Although we can not exclude formally this possibility, we do not favor it for two reasons: (1) Cho/Cr was about the same in patient 3 with a more advanced disease as compared to patients 1 and 2. (2) Disease progression is expected to cause a decrease in NAA (considered as a marker of neuronal viability) and an increase in choline (considered as a marker of membrane destruction or gliosis) [13,14]. However, NAA remained stable during 24 months whereas choline decreased during the same time suggesting a direct effect of miglustat.

Third, some variations in the position and size of the volume of interest could account for Cho/Cr decrease. This may be the case for the last examination of patient 1, and such a variation is diffi-

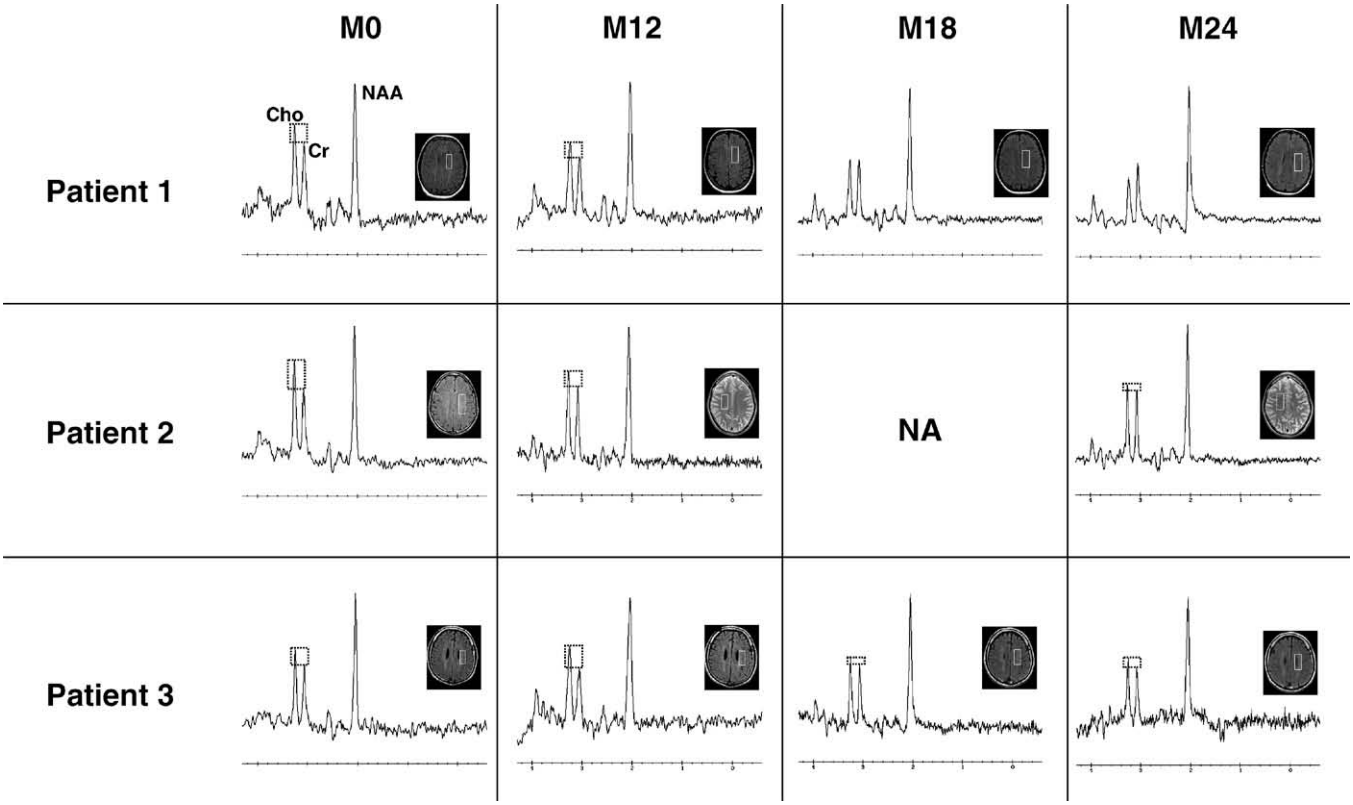

Fig. 1. MRS spectra and voxel positions. M0, M12, M18, M24: MRS spectra at baseline and after 12, 18 and 24 months of treatment with miglustat. The Cho/Cr ratio is materialized by a dotted rectangle. Note that Cho/Cr decreases over time in all patients. Cho, choline peak; Cr, creatine peak; NA, non available; NAA, *N*-acetyl aspartate peak.

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cult to avoid in disabled patients, even with careful repositioning in the magnet. However, it is unlikely that variation in the voxel size or position is responsible for the decrease in the Cho/Cr ratio observed at all time points in all patients.

Fourth, the choice of the cerebral white matter (centrum ovale or semi ovale) to follow disease's progression seems paradoxical since NPC mainly affects basal ganglia, cerebellum or brain stem [1]. This choice was motivated by practical considerations: MRS spectra were of better quality in this region compared to cerebellum or basal ganglia.

Fifth, it is still not known, to what extent spectroscopic changes can be translated into clinical improvement. While the MRS changes are encouraging, their clinical and pathological significance is elusive. Indeed, the clinical outcomes in our patients as in previous studies were at best modest.

MRS has been used for many years in the diagnosis and follow up of nervous system diseases including inborn errors of metabolism [14]. Although abnormalities in metabolite peaks have limited diagnostic value, they may constitute useful quantitative surrogate markers to monitor treatment efficacy. For instance, in patients with adrenoleukodystrophy, the efficacy of bone marrow transplantation is correlated in individual cases with a decrease in Cho and an increase in NAA [15]. While MRS has been proposed to measure neuronal dysfunction *in vivo* in NPC it has not been used in therapeutic trials so far [13]. The procedure used in our study is however, very simple: it is based on a single voxel acquisition in the centrum ovale or semiovale at long echo time with a simple automatic ratio calculation by the machine.

Although results reported here need to be confirmed, they suggest that (1) MRS could be used routinely as a non invasive quantitative and objective surrogate marker of treatment efficacy and (2) treatment with miglustat should be efficacious in NPC after a relative long period of time and this effect would exist even in advanced stages of the disease. The need for comprehensive studies of the course of untreated NPC in a large multicenter series of patients with the different categories of clinical presentation is increasingly pressing if we are to gain further useful therapeutic insight into this disorder.

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