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Clinical experience with miglustat therapy in pediatric patients with Niemann–Pick disease type C: A case series

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ABSTRACT

Niemann–Pick disease type C (NP-C) is an inherited neurovisceral lysosomal lipid storage disease characterized by progressive neurological deterioration. Different clinical forms have been defined based on patient age at onset: perinatal, early-infantile (EI), late-infantile (Li), juvenile and adult. We evaluated the efficacy and tolerability of miglustat in 16 symptomatic NP-C patients, with comparative reference to one neurologically asymptomatic, untreated patient. All patients were categorized according to age at neurological disease onset, and were assessed using a standardized clinical assessment protocol: disability and cognitive function scales, positron emission tomography (PET), and biochemical markers. PET and disability scale evaluations indicated that cerebral hypometabolism and neurological symptoms were stabilized during treatment in juvenile-onset NP-C patients. EI and Li NP-C patients, who had higher disease severity at baseline (treatment start), showed increased disability scores and progressive cerebral hypometabolism during follow up. Similarly, while cognitive scale scores remained relatively stable in patients with juvenile NP-C, cognition deteriorated in EI and Li patients. Plasma chitotriosidase (ChT) activity was lower in the juvenile NP-C subgroup than in EI and Li patients, and generally increased in patients who discontinued treatment. Plasma CCL18/PARC and ChT activities indicated greater macrophagic activity in EI and Li patients *versus* juveniles. Miglustat was generally well tolerated; frequent adverse events included diarrhea and flatulence, which were managed effectively by dietary modification and loperamide. Overall, miglustat appeared to stabilize neurological status in juvenile-onset NP-C patients, but therapeutic benefits appeared smaller among younger patients who were at a more advanced stage of disease at baseline.

Introduction

Niemann–Pick disease type C (NP-C) is a neurovisceral lysosomal lipid storage disease characterized by progressive neurological deterioration. NP-C is caused by mutations in either one of the two genes, *NPC1* or *NPC2*, which encode proteins involved in the regulation of normal intracellular lipid trafficking through sequential activities within a common pathway [1–4]. Expression of the mutant genes leads to severely impaired intracellular lipid transport and marked accumulation of both unesterified cholesterol and several glycosphingolipids in a variety of tissues and organs, in particular the brain [2,3,5,6].

NP-C has a highly variable clinical presentation. The symptomatology and rate of disease progression are strongly influenced by age at disease onset [7,8], and different clinical forms have been described on this basis. In the perinatal form, patients die from liver failure within the first months of life. Other forms are defined based

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on the following ages at onset: early-infantile (EI) form, <2 years; late-infantile (Li) form, 3–5 years; juvenile form, 5–16 years; adult form, >16 years. Clinical symptoms include progressive neurological deterioration and visceral organomegaly. Neurodegeneration begins with clumsiness and progressive ataxia followed by a range of symptoms that can generally include dysmetria, vertical supranuclear ophthalmoplegia, cataplexy, seizures, dystonia, pyramidal signs, dysphagia and dementia [8,9].

The biochemical diagnosis of NP-C is currently based on the demonstration of impaired low-density lipoprotein (LDL) cholesterol trafficking in cultured fibroblasts from patients, by cytochemical visualization of accumulated free cholesterol after filipin staining [9]. Recently, CCL18 pulmonary and activation-regulated chemokine (PARC), termed hereinafter as 'CCL18', has been reported as a potential new surrogate marker for monitoring symptomatic patients with Gaucher disease (GD) [10]. On average, this protein is elevated 29-fold in GD patients, without overlap between patient and control values. Chitotriosidase (ChT) is a human chitinase that shows markedly elevated activity in a variety of lysosomal storage disorders [11]. It is secreted by activated macrophages and is thought to play a role in defense against chitin-containing pathogens, in tissue remodeling and cell migration, as well as during atherogenesis. Plasma ChT is considered a useful surrogate marker in the lysosomal work-up of GD and NP-C patients with organomegaly, as it is relatively inexpensive and is easily assayed [12]. However, the use of plasma ChT as a marker of disease progression can be problematic in some patients who have no ChT activity due to possession of a 24-base pair (bp) duplication in the ChT gene; this mutation is inherited as an autosomal recessive trait [11]. Nevertheless, plasma ChT is considered also to be of possible use as a screening marker in pediatric patients [12].

Currently, there is no cure for NP-C, although palliative therapy can alleviate some symptoms of the disease [13]. Miglustat (*N*-butyldeoxynojirimycin; NB-DNJ; OGT-918) is a small iminosugar molecule that reversibly inhibits glucosylceramide synthase, the enzyme that catalyses the first committed step in glycosphingolipid synthesis [14]. The ability of miglustat to cross the blood–brain barrier indicated its potential use as a therapy for lysosomal storage diseases affecting the central nervous system. In animal NP-C models, miglustat delayed the onset of neurological symptoms and increased life span [15]. Evidence suggests that miglustat might also have beneficial effects on pathogenetic NP-C cellular pathways associated with calcium homeostasis [16]. Based on findings from a randomized, controlled clinical trial and a retrospective observational cohort study [17,18], miglustat was approved in the European Union for the treatment of progressive neurological manifestations in adult patients and pediatric patients with NP-C in January 2009.

We report an evaluation of 17 patients with NP-C (16 symptomatic and one neurologically asymptomatic) from Spain and Portugal who were treated with miglustat for up to 4 years. We applied a standardized clinical, biochemical and neuroimaging protocol in order to establish the effect of miglustat on several markers of NP-C severity.

Methods

Patients and dosing

During the last 4 years, we evaluated 17 children from Spain and Portugal who had clinical and biochemical diagnoses of NP-C. In all patients, biochemical diagnoses were confirmed by fibroblast assays for cholesterol esterification as well as filipin staining. Four of these patients have also been described in a retrospective observational cohort study of miglustat in patients with NP-C [18].

Clinical records for all patients were collected by a single investigator, and clinical NPC1 phenotypes were categorized according to age at onset of neurological symptoms. All 16 symptomatic patients received miglustat at doses based on body surface area (BSA):(BSA patient (m^2)/1.8) × adult dose (200 mg t.i.d.). The neurologically asymptomatic patient, who was diagnosed at 8 months of age due to splenomegaly, was not receiving miglustat and was included as a control at 8 years old, following all the protocol.

Clinical assessments

A standard assessment protocol was applied to all patients, including: clinical assessment (neurological examination, modified functional disability scale and cognitive development evaluation), biochemical analyses (plasma ChT and CCL18 activities), and imaging studies (abdominal ultrasound and cerebral positron emission tomography [PET] with radiolabelled [^{18}F]-2-fluoro-deoxy-D-glucose). The full assessment battery was applied at baseline (treatment start), 6 months, 12 months and every year thereafter. Neurological examinations and biochemical analyses were also performed at screening and Months 4 and 8. Disability scale assessments were performed every 4 months. In addition, all treatment-emergent adverse events were recorded at each post-screening visit.

The modified disability scale, assessing four key functional domains (ambulation, manipulation, language and swallowing) relating to disease severity, was calculated as reported previously [8]. We modified our disability scale with the following scores: ambulation score ranged from 1 (clumsiness) to 5 (wheelchair-bound); manipulation ranged from 1 (mild tremor) to 4 (severe dysmetria/dystonia); language ranged from 1 (delayed acquisitions) to 5 (absence of communication); swallowing ranged from 1 (abnormal chewing) to 4 (nasogastric/gastric button feeding); epilepsy ranged from 1 (occasional seizures) to 3 (seizures resistant to antiepileptic drugs); ocular movements ranged from 1 (slow ocular pursuit) to 3 (complete ophthalmoplegia) (Table 1). Scores on each of these domains were used to calculate an overall (composite) disability score, which are referred to simply as 'disability scale scores throughout the remainder of this report.

The Denver developmental screening test (DDST) and Wechsler intelligence scale for children (WISC–R) were used to assess cognitive development and function. Cranial PET was used to assess brain metabolism in affected cerebral areas including the frontal and temporo–parietal regions, as well as in the thalamus, basal ganglia and cerebellum. Impaired brain function was rated according to scores ranging from 1 (mild) to 5 (severe).

Written informed consent for participation in this study was obtained from all parents or their legal representatives.

ChT studies

Plasma ChT activity was measured using the fluorogenic substrate, 4-methylumbelliferyl-β-D-*N*,*N*′,*N*″-triacetylchitotrioside (4-MU-chitotrioside; Sigma Chemical Co, St Louis, MO, USA), as described previously by Hollak et al. [19]. Samples with high ChT activities were diluted to bring them into the linear range of the assay. Measured enzyme activities were doubled in patients carrying the mutated ChT gene.

A 24-bp duplication polymorphism in the ChT gene in some patients leads to a null allele, producing a defective protein product and a subsequent inherited deficiency in ChT activity. DNA analysis of ChT gene polymorphisms was undertaken in all but one patient using PCR followed by agarose gel electrophoresis of the amplified fragment as described previously [20].

Table 1
Modified disability scale for patients NP-C.

Ambulation	Score	Language	Score
Clumsiness	1	Delayed acquisitions	1
Autonomous ataxia gait	2	Mild dysarthria (understandable language)	2
Outdoor assisted ambulation	3	Severe dysarthria (only understood by some members of the family)	3
Indoor assisted ambulation	4	Non-verbal communication	4
Wheelchair-bound	5	Absence of communication	5
Manipulation	**Score**	**Swallowing**	**Score**
Tremor	1	Abnormal chewing	1
Slight dysmetria/dystonia (allows autonomous manipulation)	2	Occasional dysphagia	2
Mild dysmetria/dystonia (requires help for several task bur is able to feed himself)	3	Daily dysphagia	3
Severe dysmetria/dystonia (requires assistance in all activities)	4	Nasogastric/gastric button feeding	4
Seizures	**Score**	**Ocular movements**	**Score**
Occasional seizures	1	Slow ocular pursuit	1
Seizures with antiepileptic drugs	2	Vertical ophthalmoplegia	2
Seizures resistant to antiepileptic drugs	3	Complete ophthalmoplegia	3

CCL18 protein assay

CCL18 protein assays were conducted using a CCL18-specific, enzyme-linked immunosorbent assay (ELISA) system according to the manufacturer's instructions (CytoSet, Biosource International, Camarillo, CA). Dilution series of recombinant human CCL18 protein (Biosource International, Camarillo, CA) were used as controls to produce standard curves.

Results

Patients and disposition

A total of 17 NP-C patients (9 male and 8 female) were included. No patients had a family history of NP-C, but all had a confirmed diagnosis. Among all 17 patients, 16 showed mutations in the *NPC1* gene (data not shown and partially published [21,22]). Table 2 summarizes patient baseline characteristics. Categorization of patients on clinical grounds according to age at onset of neurological symptoms identified five patients with the EI form of the disease, four patients with the Li form, and seven patients with the juvenile form. The duration of miglustat treatment ranged between 6 months and 4 years; therapy was initiated at different patient ages, depending on the clinical form.

Discontinuations due to death were reported in the following patients: patient 2 (at 6 months of treatment), patient 5 (at 14 months of treatment due to respiratory infection as a complication of immunosuppressive therapy following liver transplantation) and patient 6 (at 2 years and 7 months of treatment, due to disease progression). Discontinuations due to family decisions were reported in two patients: patient 7 after 2 years of treatment and patient 10 after 3 years of treatment.

Splenomegaly

Splenomegaly showed a high degree of variability in patients with EI NP-C and in the neurologically asymptomatic patient. The median spleen volume ranged from 140 to 200 mm (normal spleen size, 100 mm). Patients with Li and juvenile NP-C showed relatively stable spleen size throughout treatment.

Disability scale scores

In the EI patient subgroup (Fig. 1a), patients 1 and 4, who started treatment at the youngest ages, showed lower baseline disability scores than patients 2, 3 and 5. Patient 4 had the lowest disability score throughout treatment, possibly due to early diagnosis of the disease due to splenomegaly investigations. The disability score of patient 1 increased over a period of approximately 2 years, but remained stable thereafter. Patient 5 had the highest disability score at the onset of treatment, and died at 14 months of age.

In the Li patient subgroup (Fig. 1b), patient 7, who started treatment at 2 years and 7 months of age, showed a low initial degree of disability and slow disease progression during 2 years of follow up. Patient 6, who started treatment later than other patients (at 8 years and 7 months of age), died after 2 years and 7 months on treatment.

In the juvenile patient subgroup (Fig. 1c), an initial decrease in disability score was observed in patients 10 and 11. The disability score for patient 10 started to increase at 32 weeks, which coincided with the onset of epilepsy that continued between Months 33 and 36; the patient discontinued treatment after this, and his disability score increased further, up to Month 44. In contrast, while patient 11 commenced treatment with the highest disability score, he showed stable disease throughout 3 years of treatment.

PET studies

In the EI patient subgroup, cranial PET studies in patient 1 showed normal metabolism in the cerebellum, thalamus and basal ganglia throughout 2 years of treatment, but there were notable changes in frontal and temporo-parietal cerebral metabolism (Fig. 2a). Substantial and generalized effects on cerebral metabolism were also seen in Patient 2 at baseline, but no follow-up cranial PET assessments could be performed because the patient died 6 months after the baseline assessment. Patient 4 showed mild disruption of thalamic metabolism at baseline, which progressed to 'moderate' at 1-year follow up; this condition appeared stabilized at 2-year follow up, with slight cerebellar hypometabolism. Patient 5 showed frontal-region impairment at baseline, which appeared stabilized at 1-year follow up.

In the Li patient subgroup, cranial PET showed an increased degree of hypometabolism in the frontal and temporo-parietal regions as well as in the thalamus and basal ganglia at 1 year of treatment in patient 6, but cerebellar function remained stable (Fig. 2b). This condition was unchanged at 2-year follow up. Patient 8 showed progressive increases in thalamic hypometabolism during the first and second years of treatment, and cerebellar changes also became apparent at 2-year follow up. Patient 9, who had the highest disability score at baseline and throughout treatment in the Li subgroup, showed improved frontal and temporo-parietal metabolism at 1-year follow up.

Table 2
Patient characteristics.

	Gender	ChT genotype[a]	NPC1 genotype	Age at treatment start	Treatment duration[b]	Dosage
Early-infantile						
Patient 1	Male	(dup/wt)	NPC40[c]	2 Years 6 months	4 Years	100 mg b.i.d.
Patient 2	Female	(wt/wt)	NPC17[c]	4 Years 6 months	6 Months	200 mg b.i.d.
Patient 3	Female	n.d.		3 Years 1 month	1 Year	50 mg t.i.d.
Patient 4	Female	(wt/wt)	NPC43[d]	1 Year 4 months	2 Years	50 mg t.i.d.
Patient 5	Male	(wt/wt)	–[e]	4 Years	1 Year 2 months	30 mg t.i.d.
Late-infantile						
Patient 6	Female	(wt/wt)	NPC22[c]	8 Years 7 months	2 Years 7 months	200 mg b.i.d.
Patient 7	Male	(wt/wt)	NPC31[c]	2 Years 7 months	2 Years	50 mg b.i.d.
Patient 8	Female	(dup/wt)	NPC36[c]	5 Years 7 months	4 Years	100 mg t.i.d.
Patient 9	Male	(wt/wt)	–[e]	6 Years 2 months	2 Years	100 mg b.i.d.
Juvenile						
Patient 10	Male	(wt/wt)	NPC34[c]	15 Years 7 months	3 Years	200 mg t.i.d.
Patient 11	Male	(dup/wt)	NPC32[c]	14 Years 1 month	4 Years	200 mg b.i.d.
Patient 12	Female	dup/19E10 + 43		11 Years	4 Years	100 mg t.i.d.
Patient 13	Male	(dup/wt)	–[e]	14 Years	3 Years	200 mg b.i.d.
Patient 14	Male	(dup/wt)	–[e]	13 Years 10 months	21 Months	200 mg t.i.d.
Patient 15	Female	(wt/wt)	–[e]	6 Years	1 Year	100 mg t.i.d.
Patient 16	Male	(wt/wt)	–[e]	10 Years	1 Year	100 mg b.i.d.
Asymptomatic						
Patient 17	Female	(dup/wt)	NPC24[c]	_	No treatment	_

[a] wt, wild-type allele; (dup/wt), 24-bp duplication in one allele; wt/wt, no duplication.
[b] Treatment duration at observation cut-off; ChT, chitotriosidase; n.d., not determined.
[c] Ref. [21].
[d] Ref. [22].
[e] Publication in draft.

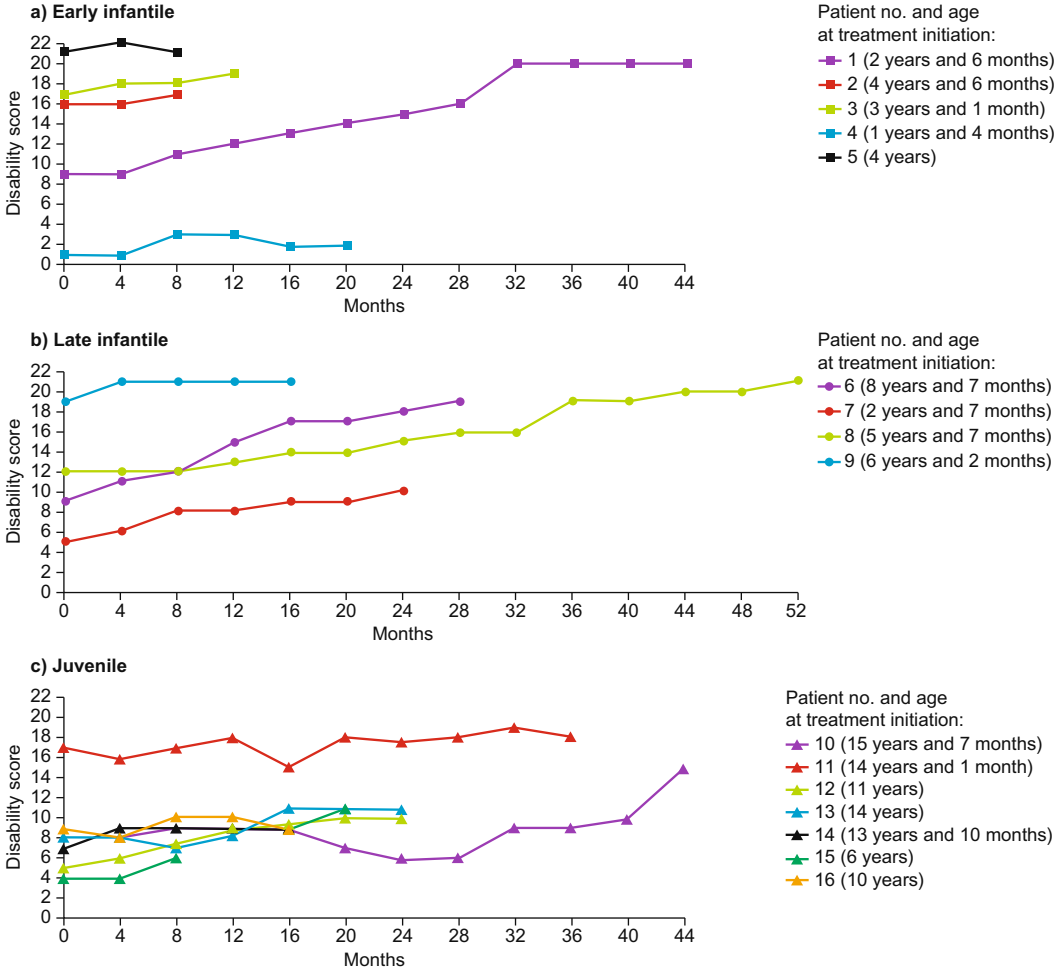

Fig. 1. Composite disability score in (a) EI patients, (b) LI patients, and (c) juvenile patients with NP-C.

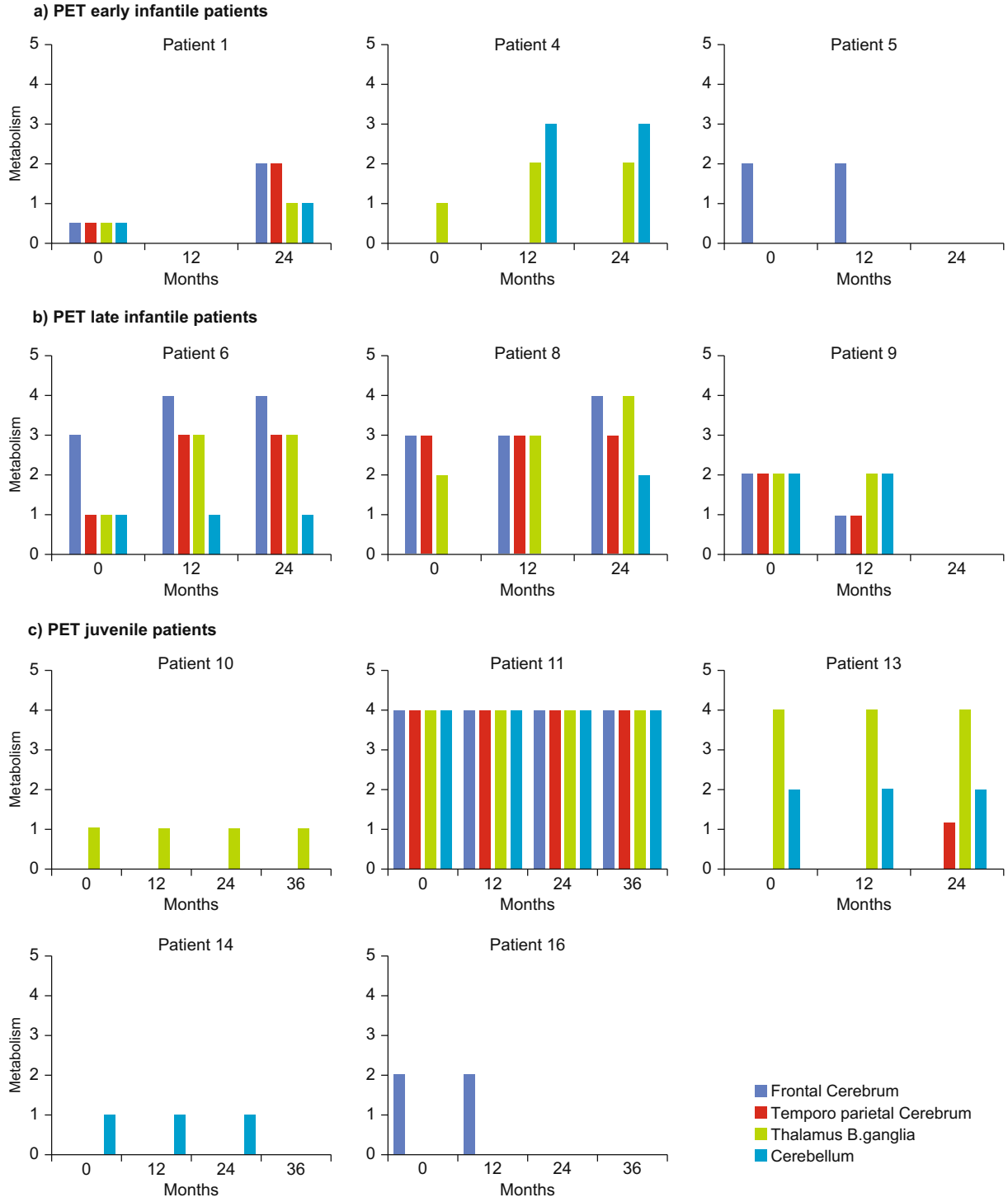

Fig. 2. PET in (a) EI patients, (b) Li patients, and (c) juvenile patients with NP-C. Findings at last PET evaluation are shown. Treatment periods may have extended past the last evaluation point. Scores range from 0 (normal cerebral metabolism) to 5 (severe impairment).

In the juvenile patient subgroup, cranial PET indicated stabilization throughout 3 years of treatment in patient 10 (Fig. 2c). Patient 11, who had the highest disability score in this subgroup, showed a generalized hypometabolism in all cerebral and cerebellar regions at baseline, which remained stable during 3 years of treatment. Patient 14 showed no changes on PET during 2 years of follow up, which appears in agreement with a lack of change in this patient's disability score throughout observation. Brain metabolism was affected in the thalamic and cerebellar regions and in the basal ganglia in Patient 13 at baseline and follow up at 2 years. There was slight hypometabolism in the temporo-parietal cerebrum at 2 years, whereas there had been none at baseline or 1-year follow up.

Findings from cranial PET assessments in the asymptomatic patient showed stable cerebral, thalamic and cerebellar function throughout 4 years of follow up (data not shown).

Cognitive development and function

In the EI patient subgroup, patient 1 experienced substantial worsening of developmental cognitive function during follow up, which paralleled increases in his disability scores; his IQ score was 71 at baseline, which fell to 20 at both 1 and 2 years of treatment. Patient 4 showed stable developmental cognitive function at 2 years of treatment, with very slight retardation. No follow-up information on cognitive function was available for patients 2 and 5, as both patients died. It was not possible to evaluate cognitive development and function in patient 3 due to progressive neurological manifestations of the disease. Cognitive development and function data were not available from patients 9, 15 and 16.

In the Li patient subgroup, patients 6, 7 and 8 experienced progressive reductions in cognitive function during 2 years of treatment. It was not possible to evaluate cognition beyond 2 years due to progressive cognitive impairment (IQ: <20) and the evolution of physical conditions of the disease.

In the juvenile patient subgroup, patient 13 showed worsening of cognitive function between baseline (IQ score 78) and 1-year follow up (IQ score 55), but cognition has since remained stable throughout years 2 and 3. Cognitive function decreased during the first year of treatment in patient 11 (IQ score 35 at baseline and 20 at 1-year follow up), which reflected a parallel increase in disability scale score. Patients 10, 12 and 14 showed stable cognitive function (IQ score 40–50) during the first year of treatment. IQ scores for patient 10 ranged from 50 at 2 years of treatment to 40 at 3 years. Patient 14 had an IQ score < 40 at 2 years of treatment.

Cognitive function has remained stable in the asymptomatic patient throughout 4 years of follow up. IQ scores ranged between 70 and 80 throughout the observation period.

Plasma ChT and CCL18 activities

Genotyping analyses for the 24-bp duplication polymorphism in the ChT gene was conducted in 16/17 patients. Plasma ChT 'pseudo-deficiency' was confirmed for seven patients (i.e. those with the 24-bp duplication in one ChT gene allele): patients 1, 8, 11, 12, 13, 14 and 17. Overall, plasma ChT activity was lower in the juvenile patient subgroup (value range; 52–474 nmol/ml h) than in the EI (value range; 316–3245 nmol/ml h) and Li subgroups (value range; 323–2744 nmol/ml h) (Fig. 3), and was increased in patients who discontinued treatment (e.g. patients 3 and 10, who both discontinued based on family decisions). The asymptomatic patient showed the lowest plasma ChT activity during 4 years of follow up (values range; 60–47 nmol/ml h).

Similar to findings with plasma ChT (overall value range; 52–3245 nmol/ml h), patients in the EI (value range; 851–1691 ng/ml) and Li subgroups (value range; 176–950 ng/ml) had higher plasma CCL18 activities compared with both the juvenile patient subgroup (value range; 112–874 ng/ml) and the asymptomatic patient (value range; 213–423 ng/ml) (Fig. 4).

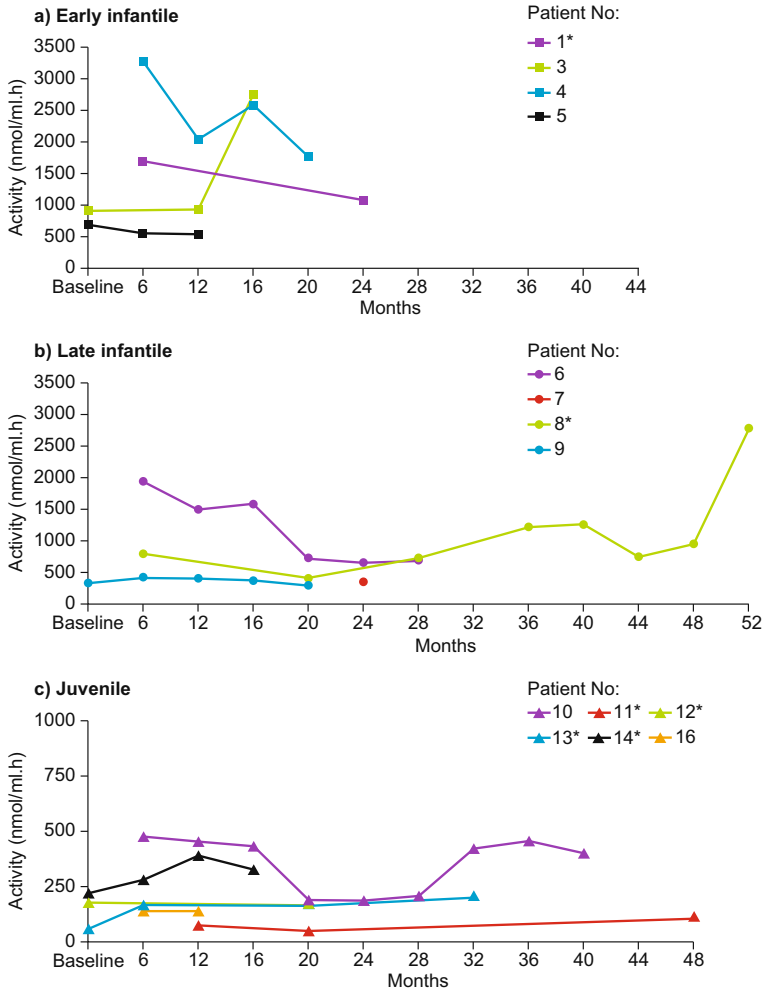

Fig. 3. Plasma ChT activity in (a) EI patients, (b) Li patients, and (c) juvenile patients with NP-C. *Pseudo-deficiency patients, possessing 24-bp duplication in one ChT gene allele; ChT, chitotriosidase. *Note*: plasma ChT activity in normal, healthy subjects is < 50 nmol/ml h. Data not available for patient 2 and patient 15.

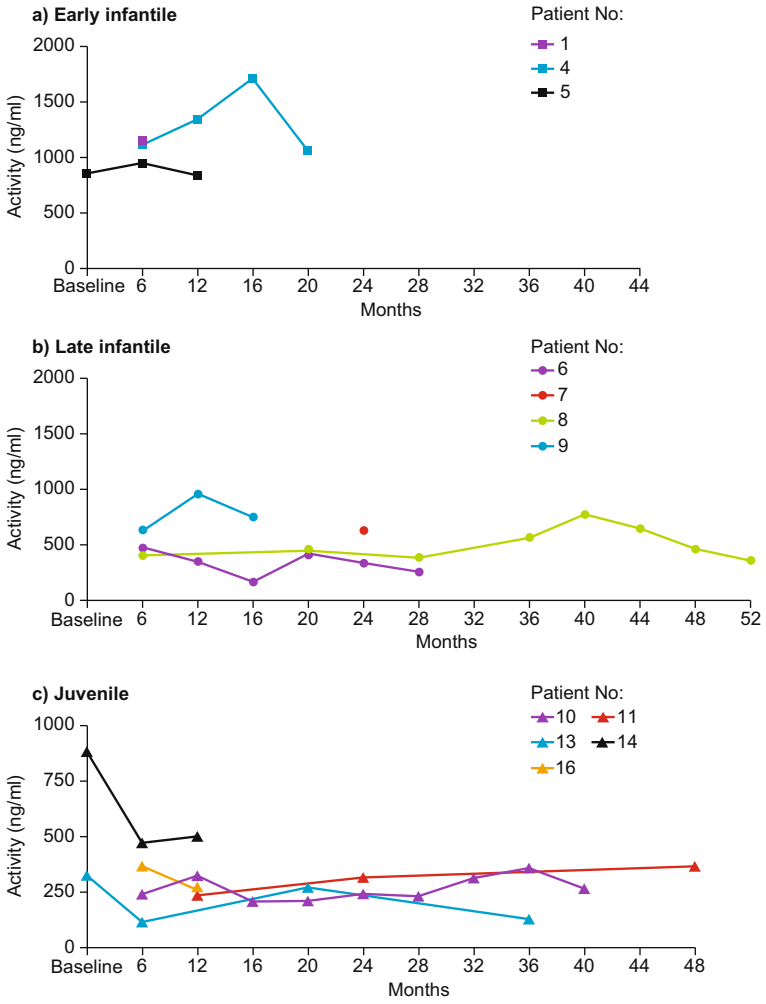

Fig. 4. Plasma CCL18 activity in (a) EI patients, (b) Li patients, and (c) juvenile patients with NP-C. *Note*: plasma CCL18 activity in normal, healthy subjects is zero. Data not available for patients 2, 3, 12 and 15.

Tolerability and safety

Miglustat was well tolerated, and no serious adverse events were recorded. The most frequently occurring adverse events were diarrhea and flatulence, which were managed satisfactorily using proper dietary and nutritional care such as the 'bland diet', an oral re-hydration solution, and loperamide. No patients showed clinically significant weight loss during the observation period.

Discussion

There are limited published case reports regarding the use of miglustat to treat pediatric patients with NP-C. Our case series assessed the effects of miglustat on disease progression in pediatric patients with different clinical forms of the disease. Our patients were classified on clinical grounds according to the age at onset of neurological symptoms. Splenomegaly did not appear to be a good marker of response to treatment, as there was no apparent effect of miglustat on spleen size; splenomegaly may be best considered as a disease hallmark for diagnosis. The disease-specific functional disability scale was a practical tool that provided valuable information from clinical assessments conducted according to a standard protocol. We modified our previous disability scale by adding epilepsy, which is a severe symptom that worsens the disease, and ocular movements, as it is a hallmark of the disease. PET is expensive and is unlikely to be of practical use for routine clinical monitoring, but it has the potential to be used as a quantitative and objective marker of treatment efficacy in the early stages of disease. Further PET data on untreated patients would be valuable. Plasma ChT and CCL18 activities may serve as biochemical markers of therapeutic response in NP-C, although possible genetic deficiency should be taken into account in the case of ChT. However, ChT and CCL18 do not necessarily reflect the evolution of neurological disease.

Our findings regarding changes in neurological symptom progression, brain metabolism, cognitive status and plasma disease markers add to existing published data on the efficacy of miglustat from previous clinical trials and case reports. Pivotal efficacy data were reported from a 12-month randomized, controlled, clinical trial involving 29 juvenile and adult patients, and a parallel, non-controlled sub-study, involving 12 patients aged 4–12 years [17]. The primary study end point – horizontal saccadic eye movement velocity (HSEM-α) – was improved with miglustat *versus* standard care in adult and juvenile patients; similar improvements were seen in children included in the pediatric sub-study [17]. Improved swallowing capacity, stable auditory acuity, and slower deterioration of ambulation were also seen in miglustat-treated patients aged over 12 years. Further data, indicating stabilization of key parameters of neurological disease progression in NP-C, were reported in a retrospective observational cohort study in 66 patients

with a mean (standard deviation) age of 9.7 (7.6) years [18]. A published case series reported the efficacy of 24 months' miglustat therapy in three adult patients with NP-C, based on clinical evaluations and brain magnetic resonance spectroscopy (MRS) [23]. This study reported mild clinical improvement or stabilization in all patients. However, the findings were limited by the small number of patients and the choice of cerebral white matter to follow disease progression.

To date, cerebral PET scan data have not been reported from longitudinal follow up of patients with NP-C. Our PET imaging studies indicated that cerebral hypometabolism was stabilized in patients with juvenile-onset NP-C; miglustat appeared to slow progression of neurological symptoms. Patients with EI and Li forms of NP-C who started treatment in the advanced stages of the disease showed increased disability scores and progressive cerebral hypometabolism. Patient 9, who also had a high baseline disability score, showed an improvement on PET evaluation after 1 year of treatment. Control cranial PET and disability scale data indicated stable function over 4 years of follow up in the asymptomatic patient.

A published case series of two male Taiwanese patients with NP-C, who started miglustat therapy aged 14 and 9 years, reported substantial improvements in swallowing and ambulation by Month 6 of treatment, followed by stabilization of neurological symptoms between Months 6 and 12 [24]. Spleen size remained approximately stable throughout treatment in both patients and, predictably, there were no overt changes in plasma ChT activities. In our series, splenomegaly varied greatly between patients and over time, and did not appear related to treatment response or to neurological status. However, plasma CCL18 and ChT levels were higher in both the EI and Li patients compared with the juvenile group and the asymptomatic patient. Further, in patients with the EI and juvenile forms of NP-C, plasma ChT activities increased when patients discontinued therapy, suggesting that this marker is reflective of therapeutic response. The levels of plasma ChT and CCL18 indicated the presence of quantifiable, active disease in the asymptomatic patient.

Previous studies have demonstrated that both plasma ChT and CCL18 can serve as markers for the extent of pathological formation of lipid-laden macrophages in GD [10]. Further, reductions in plasma CCL18 levels have been shown able to reflect therapeutic corrections of the total-body burden of activated macrophages. It should also be noted that plasma CCL18 levels are affected by immune system activity, and it is possible that CCL18 levels might reflect specific immune processes during NP-C. Zimran et al. [25] characterized the inflammatory profile of patients with GD, and it might be that similar factors and processes are involved in the modulation of plasma CCL18 concentration in patients with NP-C.

Santos et al. [26] studied the effects of miglustat treatment in a 9-year old Brazilian patient, reporting a rapid and positive impact of therapy on cognitive function, ataxia, dysarthria and ophthalmoplegia. In addition, functional disability (assessed on the disability scale published by Iturriaga et al. [8]) was reduced from a pretreatment score of 15 down to a score of 8, after treatment. These findings are comparable with data from our series. Although disability scale scores increased in patients with EI disease (e.g. in patient 1 at 2-year follow up), progression was slow compared with the natural evolution of this clinical form. In addition, patient 4 remained stable throughout 2 years of treatment, and a decrease in disability scores was observed in patients with the juvenile form of the disease (although this trend reversed at the onset of epilepsy).

With regard to cognitive function and developmental level, the severity of cognitive impairment appears strongly related to the age at which treatment is commenced; in general, cognitive outcomes were worse in younger patients. There was a clear worsening of cognitive scores in patients with EI and Li disease during follow up, but scores appeared to remain stable in patients with juvenile-onset disease.

Miglustat was generally well tolerated in our patient series. Some patients experienced episodes of diarrhea and flatulence at the onset of treatment, but there was no weight loss during the observation period. There were no reports of insomnia, paresthesia or fine tremors, which have been described in previous studies with miglustat [17,24,26].

In summary, the patients in our series who showed deterioration during miglustat therapy were those who were at a more advanced stage of the disease. This seems in agreement with a previous case of a male patient aged 3 years, where little therapeutic response was reported after 12 months' treatment with miglustat [27]. In general, patients with infantile NP-C generally exhibit greater symptom severity and more rapid disease progression than those with juvenile-onset disease [28], and therefore seem less likely to show appreciable therapeutic responses to miglustat therapy [9]. We consider that miglustat therapy should be commenced at or just before neurological signs start to appear. However, more data are necessary to define further the stages at which treatment is best initiated within the different clinical forms of NP-C.

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