Molecular Genetics and Metabolism xxx (2014) xxx–xxx

Contents lists available at ScienceDirect

Molecular Genetics and Metabolism

journal homepage: www.elsevier.com/locate/ymgme

Efficacy of pyruvate therapy in patients with mitochondrial disease: A semi-quantitative clinical evaluation study

Tatsuya Fujii [a,*], Fumihito Nozaki [a], Keiko Saito [a,1], Anri Hayashi [a], Yutaka Nishigaki [b,2], Kei Murayama [c], Masashi Tanaka [b], Yasutoshi Koga [d], Ikuko Hiejima [a], Tomohiro Kumada [a]

[a] Department of Pediatrics, Shiga Medical Center for Children, 5-7-30 Moriyama, Shiga 524-0022, Japan
[b] Department of Genomics for Longevity and Health, Tokyo Metropolitan Institute of Gerontology, 35-2 Sakane-cho, Itabashi, Tokyo 173-0015, Japan
[c] Department of Metabolism, Chiba Children's Hospital, 579-1 Heta-cho, Midori, Chiba 266-0007, Japan
[d] Department of Pediatrics and Child Health, Kurume University Graduate School of Medicine, 67 Asahi-machi, Kurume, Fukuoka 830-0011, Japan

ARTICLE INFO

Article history:
Received 26 February 2014
Received in revised form 25 April 2014
Accepted 25 April 2014
Available online xxxx

Keywords:
Pyruvate
Therapy
Mitochondrial disease
NAD^+
Lactate-to-pyruvate ratio

ABSTRACT

Background: Disorders of oxidative phosphorylation (OXPHOS) cause an increase in the NADH/NAD^+ ratio, which impairs the glycolysis pathway. Treatment with pyruvate is expected to decrease the ratio and thereby restore glycolysis. There are some case reports on the efficacy of pyruvate treatment for mitochondrial diseases. However, few of these reports assessed their results using a standardized scale.

Methods: We monitored 4 bedridden patients with OXPHOS disorders who continued therapies of 0.5–1.0 g/kg/day of sodium pyruvate for more than 12 months. The efficacies of these treatments were evaluated with the Newcastle Pediatric Mitochondrial Disease Scale and the Gross Motor Function Measure with 88 items.

Results: The ages of the patients at the treatment initiation ranged from 8–100 months. Of the 4 patients, 3 exhibited improvements within 1–3 months from the initiation of treatment. Among these 3 patients, one maintained the improvement for over 2 years. The remaining 2 regressed 3–6 months after the initiation of treatment. The blood lactate/pyruvate ratios did not correlate with the efficacy of treatment.

Conclusion: Pyruvate was effective even in bedridden patients with OXPHOS disorders, at least in the short term. Clinical trials with more patients and less severe disabilities are necessary to evaluate the long-term efficacy of this treatment. Biomarkers other than lactate and pyruvate need to be identified to biochemically monitor the efficacy of this treatment.

1. Introduction

Tanaka et al. [1] proposed that pyruvate has therapeutic potential for patients with oxidative phosphorylation (OXPHOS) disorders in which the intracellular NADH/NAD^+ ratio is increased. Such an increased ratio impairs the activity of glyceraldehyde-3-phosphate dehydrogenase (GAPDH) in the glycolysis pathway. Theoretically, with lactate dehydrogenase, pyruvate provides NAD^+ and decreases this ratio and thereby restores the activity of GAPDH, which produces ATP. Additionally, pyruvate activates pyruvate dehydrogenase and non-enzymatically eliminates hydrogen peroxide.

There are several case reports on the efficacy of pyruvate in patients with OXPHOS disorders [2–4]. However, few of these reports have evaluated the clinical outcomes using a standardized clinical assessment scale. We semi-quantitatively evaluated the efficacy of pyruvate therapy in 4 patients with OXPHOS disorders using standardized scales. This study was approved by the Ethical Committee of our institution. Written informed consent was obtained from the parents of every patient.

2. Patients and methods

2.1. Patients

Four patients who had been on pyruvate for more than 12 months were studied (Table 1). Two patients had Leigh syndrome associated with m.8993 T>G or m.9176 T>C mutations. One patient had non-specific encephalomyopathy associated with complex I and IV combined deficiency. Another patient had myopathic mitochondrial DNA depletion syndrome. All patients were bedridden, and all but one

Abbreviations: NPMDS, Newcastle Pediatric Mitochondrial Disease Scale; GMFM-88, Gross Motor Function Measure with 88 items; JMDRS, Japanese Mitochondrial Disease Rating Scale; OXPHOS, Oxidative phosphorylation; MELAS, Mitochondrial myopathy, encephalopathy, lactic acidosis and stroke-like episodes; FGF-21, Fibroblast growth factor 21.

* Corresponding author at: Department of Pediatrics, Shiga Medical Center for Children, 5-7-30 Moriyama, Moriyama-City, Shiga 524-0022, Japan.
E-mail address: tatsufu@gmail.com (T. Fujii).
[1] Present address: Department of Pediatrics, Graduate School of Medicine, Kyoto University, 54 Shogoinkawahara-cho, Sakyo, Kyoto, Kyoto 606-8507, Japan.
[2] Present address: Nishigaki Clinic & Research Laboratory, 1-177 Uchinaka, Nakagawa, Nagoya 454-0927, Japan.

http://dx.doi.org/10.1016/j.ymgme.2014.04.008
1096-7192/

Table 1
Profiles of the patients.

Patients	Clinical Dx	Molecular or biochemical Dx	Age at the start of the Tx	ADL at the start of the Tx	Dose of sodium pyruvate (g/kg/day)	Duration of the Tx
Patient 1	Leigh syndrome	m.8993 T>G	8 y 4 m	Bedridden Unable to roll over Tube fed	0.5	27 m
Patient 2	Leigh syndrome	m.9176 T>C	8 m	Bedridden Unable to roll over Tube fed	0.5	66 m
Patient 3	Non-specific encephalomyopathy	Complex I + IV deficiency	1 y 8 m	Able to roll over to one direction Unable to creep Orally fed	0.5 then 1.0	17 m
Patient 4	Myopathic mitochondrial depletion syndrome	mtDNA depletion	1 y 7 m	Bedridden Unable to roll over On a respirator Tube fed	0.5	41 m

Dx, diagnosis; Tx, treatment; mt, mitochondrial; ADL, activities of daily living.

(namely, the patient with combined deficiencies of complex I and IV) were tube fed. The ages at the initiation of pyruvate therapy were 8–100 months (median 20 months). The durations of therapy were 17–66 months (median 34 months). During the pyruvate therapy monitoring period, all other concomitant mitochondrial disease medications were maintained unchanged.

2.2. Pyruvate

Sodium pyruvate was obtained from Musashino Chemical Laboratory (Tokyo). Sodium pyruvate was administered at 0.5 g/kg/day orally or through a feeding tube in 2 divided doses. This dose was increased to 1.0 g/kg/day in one patient. To avoid osmotic diarrhea, the pyruvate was dissolved in water at concentrations of approximately 2%–10%. Higher concentrations were utilized if the dilution caused overhydration or the volume was too large to drink.

2.3. Clinical evaluation

The efficacy of the pyruvate therapy was clinically evaluated with 3 standard scales: the Newcastle Pediatric Mitochondrial Disease Scale (NPMDS) [5], the Gross Motor Function Measure with 88 items (GMFM-88) [6], and the Japanese Mitochondrial Disease Rating Scale (JMDRS) [7]. The NPMDS is composed of 4 domains: Section I, current function; Section II, systemic specific involvement; Section III, current clinical assessment; and Section IV, quality of life. Sections I–III are scored based on objective observations, and Section IV takes the subjective views of the parents into account. Higher scores indicate more severe clinical situations. There are 3 sets of age-specific NPMDSs. Depending on the patient's age at the time of the evaluation, the NPMDS for 0–24 months or that for 2–11 years was used. The GMFM-88 is composed of 5 dimensions: A, lying and rolling; B, sitting; C, crawling and kneeling; D, standing; and E, walking, running and jumping. The scores are expressed in percentages relative to the maximum score in each dimension. The total score is expressed as the mean of percentages across all 5 dimensions. As the patients were bedridden, only dimensions A and B could be assessed, and the scores for the dimensions C to E were considered to be zero %. Higher scores indicate better motor abilities. The JMDRS is the modified Japanese version of the European Neuromuscular Conference (ENMC) Mitochondrial Disease Rating Scale [8]. Higher scores in this scale indicate more severe symptoms. With the exception of Patient 4, who was only assessed with the NPMDS, all other patients were evaluated with the NPMDS and the GMFM at the same time. Patient 2 was initially monitored with the JMDRS. Then, after a 4-week-washout period, the patient was reassessed with the NPMDS and GMFM. Changes in motor functions that were too subtle to be detected with these scales were descriptively recorded. Serum lactate and pyruvate levels as well as plasma amino acids were monitored.

2.4. Statistical analysis

Statistical analysis of the biochemical data was performed using Mann–Whitney *U*-test. A value of $p < 0.05$ was considered as statistically significant.

3. Results

The changes in motor function and assessment scores are summarized in Table 2.

3.1. Patient 1 (m.8993 T>G Leigh syndrome)

The therapy was initiated at the age of 8 years and 4 months, and at this time, this female patient was unable to roll over. In the supine position, she could not raise her legs more than 45 degrees from the floor (as measured at the hip joint). One month after the initiation of therapy, the patient gained the abilities to roll over and raise her legs vertically from the floor. The movement of her arms became more active and rapid. The overall NPMDS score changed from 42.3 to 38.6. The sum of the scores for sections I–III changed from 31 to 29, which indicates that the objective findings improved by 2 points over one month. Dimension A of the GMFM-88 also changed from 31.4% to 47.1%, which resulted in a change from 6.3% to 9.4% in the total score. Thus, this patient's improvement was confirmed semi-quantitatively with 2 scales. Next, pyruvate was withdrawn to confirm the effect of the pyruvate treatment. Within 1 to 2 weeks, the patient became lethargic and less active. After 19 days of washout, she developed status epilepticus. Resumption of pyruvate therapy restored her clinical status to the pre-washout state. Upon re-evaluation at the age of 10 years and 7 months (after 26 months of treatment excluding the washout period), the patient exhibited maintained improved motor ability as confirmed by the unchanged GMFM-88 score. The NPMDS was not administered at this point.

Blood lactate levels and lactate/pyruvate ratios measured twice during the pre-treatment period and once after the 19-day-washout were from 1.2 mM to 1.5 mM (median 1.2 mM), and from 14.2 to 25.6 (median 19.7), respectively. Those measured at 1, 4, 18 and 20 months after the treatment resumption following the washout period ranged from 0.81 mM to 1.2 mM (median 0.85 mM), and from 15.7 to 27.3 (median 20.0), respectively (Table 3). Thus, lactate levels decreased with pyruvate therapy, but the difference was not significant. Lactate/pyruvate ratio was not reduced. Plasma alanine, valine and lysine levels were measured after the washout and 1 month after the treatment resumption. None of these decreased with the therapy (Table 3).

Please cite this article as: T. Fujii, et al., Efficacy of pyruvate therapy in patients with mitochondrial disease: A semi-quantitative clinical evaluation study, Mol. Genet. Metab. (2014), http://dx.doi.org/10.1016/j.ymgme.2014.04.008