EPI-743 reverses the progression of the pediatric mitochondrial disease—Genetically defined Leigh Syndrome

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A B S T R A C T

Background: Genetically defined Leigh syndrome is a rare, fatal inherited neurodegenerative disorder that predominantly affects children. No treatment is available. EPI-743 is a novel small molecule developed for the treatment of Leigh syndrome and other inherited mitochondrial diseases. In compassionate use cases and in an FDA Expanded Access protocol, children with Leigh syndrome treated with EPI-743 demonstrated objective signs of neurologic and neuromuscular improvement. To confirm these initial findings, a phase 2A open label trial of EPI-743 for children with genetically-confirmed Leigh syndrome was conducted and herein we report the results.

Methods: A single arm clinical trial was performed in children with genetically defined Leigh syndrome. Subjects were treated for 6 months with EPI-743 three times daily and all were eligible for a treatment extension phase. The primary objective of the trial was to arrest disease progression as assessed by neuromuscular and quality of life metrics. Results were compared to the reported natural history of the disease.

Results: Ten consecutive children, ages 1–13 years, were enrolled; they possessed seven different genetic defects. All children exhibited reversal of disease progression regardless of genetic determinant or disease severity. The primary endpoints—Newcastle Pediatric Mitochondrial Disease Scale, the Gross Motor Function Measure, and PedsQL Neuromuscular Module—demonstrated statistically significant improvement (p<0.05). In addition, all children had an improvement of one class on the Movement Disorder–Childhood Rating Scale. No significant drug-related adverse events were recorded.

Conclusions: In comparison to the natural history of Leigh syndrome, EPI-743 improves clinical outcomes in children with genetically confirmed Leigh syndrome.

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1. Introduction

Subacute necrotizing encephalopathy, or Leigh syndrome, is a rare, fatal inherited neurodegenerative disorder that predominantly affects children. Originally described in 1951 by Leigh, the histopathological findings consist of necrotizing focal bilateral lesions in the brain stem, thalamus, and/or basal ganglia [1]. Leigh syndrome arises from DNA defects in mitochondrial [2], nuclear [3–5], or X-linked genes [6–9].

The estimated incidence of Leigh syndrome is 1:30,000 [4,10,11]. There are no approved drugs for Leigh syndrome.

EPI-743 is a small-molecule therapeutic being developed for the treatment of inherited respiratory chain diseases, including Leigh syndrome. EPI-743 is a para-benzoquinone targeting repletion of reduced intracellular glutathione [12,13].

Given EPI-743’s favorable preclinical safety and efficacy profile, the lack of approved therapies and the significant morbidity and mortality associated with inherited respiratory chain diseases, the United States Food and Drug Administration granted approval for the study of EPI-743 in an Expanded Access Protocol for children with genetically confirmed inherited respiratory chain diseases within 90 days of end of life care (NCT01370447). Children with Leigh syndrome.
treated under Expanded Access have consistently demonstrated arrest of disease progression and neurologic and neuromuscular improvement on clinical and radiographic examination. In addition, none of these children experienced drug-related serious adverse events [12].

Based on these data, a phase 2A study was undertaken to further study the efficacy and safety effects of EPI-743 in children with Leigh syndrome in a controlled trial. Herein we report on the clinical response of 10 consecutive children treated with EPI-743 for 6 months in a single-site, open-label phase 2A study.

2. Methods

2.1. Study overview and subjects

We performed a prospective single arm subject-controlled trial of EPI-743 in children with genetically confirmed Leigh syndrome. This study was conducted at the Bambino Gesù Children’s Hospital, Rome, Italy. Institutional Review Board approval was obtained prior to study initiation (Vatican Ethics Committee Reference Number: EPI-2011-004) and registered in Europe (EudraCT Number: 2012-001294-84).

Subjects were children with a genetically confirmed diagnosis of Leigh syndrome who had a Newcastle Pediatric Mitochondrial Disease Scale (NPDMS) score on sections I through III greater than 15—signifying at least moderately severe disease. All participants were required to have MRI confirmation of necrotizing encephalopathy. In addition, all children were required to discontinue the use of CoQ10 and any other antioxidant supplements for the duration of the trial. Informed consent was obtained from the parents of each child.

2.2. Treatment

Subjects were treated for 6 months with 100 mg of EPI-743 three times daily orally or via gastrostomy tube and evaluated using disease-relevant functional, neurologic and physiologic assessments. All adverse and serious adverse events were tracked in the trial database. In addition, all held doses due to intolerance of EPI-743 or any other reason were recorded.

2.3. Endpoints

The primary endpoints of this study were the Newcastle Pediatric Mitochondrial Disease Scale (NPDMS), the Gross Motor Function Measure (GMFM) and the PedsQL Neuromuscular Module (PedsQL). The NPDMS is a scale developed for and validated in children with inherited mitochondrial diseases to assess disease severity. Sections I–III of the NPDMS assess organ-specific function and section IV is a quality of life assessment. In this study we utilized serial NPDMS measurements to assess EPI-743 effect on disease progression. Appropriate age-specific versions of the NPDMS were utilized: 0–24 months; 2–11 years; and 12–18 years. The GMFM is an observational tool used to assess gross motor function over time in children with neuromuscular disorders [14]. The GMFM has been validated as an outcome measurement instrument in intervention trials of children with neuromuscular disorders [14]. The PedsQL is a validated measurement of pediatric quality of life and the neuromuscular module utilized in this study is specific to children with neuromuscular disorders [15].

In addition, clinical response was measured using the Movement Disorder-Childhood Rating Scale (MD-CRS), a validated instrument for assessing movement disorders and their impact on development in children [16,17].

All assessments at all time points were performed by a single physician with expertise in pediatric neurology and mitochondrial disease (DM).

2.4. Pharmacokinetic data

Complete sample sets (T0 to T18h) for analysis of plasma concentrations following the first dose of EPI-743 at 100 mg were obtained from 5 of the patients selected randomly. Plasma concentrations were analyzed using a liquid chromatography-tandem mass spectroscopy method.

2.6. Review of natural history

Given the open label nature of this trial, treatment response was also assessed against the natural history of Leigh syndrome. A search of the PubMed database was conducted using the following criteria: i) case reports or series describing children with Leigh syndrome; ii) written in English; iii) published from 2000 to 2012. All identified publications were reviewed and those that included case histories of children with one of the seven mutations evaluated in this study (see Table 1) formed the basis of the analysis. Each child’s clinical outcome was categorized as improved, stable, progressing, or death based on the description in the paper. When available, patient sex, age at diagnosis, and age at last follow-up were recorded.

2.7. Statistical analyses

Descriptive statistics were performed on the entire cohort of children enrolled in this study. Changes in outcome measurement were calculated for each subject and overall mean changes from baseline were analyzed using a Wilcoxon signed rank test. Given that different versions of the NPDMS were used based on patient age, the Wilcoxon test was used to determine overall significance of treatment effect across the entire population and mean changes were calculated separately for each age group. Statistical significance was defined as p < 0.05.

3. Results

A total of 10 children with seven forms of genetically confirmed Leigh syndrome were enrolled in this study and treated with EPI-743 for at least 4 months. Nine of the 10 subjects completed 6 months of EPI-743 treatment. The parents of one patient discontinued treatment after 111 days of therapy due to (parent) concerns related to patient somnolence. This symptom was deemed by the investigators to be related to the underlying disease.

The baseline patient characteristics are shown in Table 1. Mean patient age was 6.3 years (range 1–13) and six of the 10 children were male. The average baseline NPDMS score was 45.4 (range 19.6–62), signifying that all children enrolled in the study had advanced disease.

3.1. Clinical outcome

The clinical outcome results are summarized in Table 2 and Fig. 1. All children—regardless of age, genotype, and starting NPDMS score—demonstrated arrest of disease progression and/or reversal. Mean

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Changes in NPMDS scores for sections I–III (organ function) and section IV (quality of life) were calculated based on patient age group given that different versions of the measurement are used for different age groups. Eight subjects were assessed with version for children ages 2–11. Sections I–III improved by a mean of 7.5±3.9 (p=0.01) and for section IV (quality of life), scores improved an average of 5.0±5.3 (p=0.02). For subject 3 (age 1) and subject 6 (age 3), scores on sections I–III and section IV also decreased following treatment with EPI-743.

There was also significant increase in GMFM scores over the treatment period with a mean improvement of 10.1±11.1 (p=0.006). On the MD-CRS—a validated measurement of pediatric movement disorders and dystonia—each child had improvement of one class, indicating significant improvement in dystonia and spasticity symptoms [16,17]. The subject who had treatment discontinued at 16 weeks had initially improved one level on the MD-CRS but returned to baseline following discontinuation of treatment.

Table 2
Summary of outcome measurements.

<table>
<thead>
<tr>
<th>Subject</th>
<th>NPMDS</th>
<th>GMFM</th>
<th>PedsQL</th>
<th>MD-CRS class score</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sections I–III</td>
<td>Section IV</td>
<td>Sections I–IV</td>
<td>Baseline 6 months</td>
</tr>
<tr>
<td>1</td>
<td>29</td>
<td>18</td>
<td>20.4</td>
<td>3.1</td>
</tr>
<tr>
<td>2</td>
<td>42</td>
<td>32</td>
<td>20</td>
<td>14</td>
</tr>
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<td>23</td>
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<td>8.8</td>
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<tr>
<td>4</td>
<td>25</td>
<td>18</td>
<td>10.8</td>
<td>6.0</td>
</tr>
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<td>7</td>
<td>15</td>
<td>9</td>
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<td>3.2</td>
</tr>
<tr>
<td>8a</td>
<td>42</td>
<td>43</td>
<td>17.5</td>
<td>16.2</td>
</tr>
<tr>
<td>9</td>
<td>33</td>
<td>26</td>
<td>12</td>
<td>7.1</td>
</tr>
<tr>
<td>10</td>
<td>25</td>
<td>14</td>
<td>7.1</td>
<td>6.7</td>
</tr>
</tbody>
</table>

Mean change: 6.8±3.8 (p=0.007)
Mean change: 5.1±4.7 (p=0.005)
Mean change: 11.6±7.3 (p=0.005)
Mean change: 10.0±11.1 (p=0.006)
Mean change: 15.0±20.8 (p=0.02)
Response rate: 90%

a Test not performed.
b Therapy discontinued at week 16.
c Pooled mean for all subjects.

Fig. 1. Clinical outcome measurements. Clinical outcomes prior to and following 6 months of EPI-743 treatment are shown. There was a significant improvement on NPMDS sections I–III (organ function) and NPMDS section IV (quality of life) as shown in panels a and b, respectively. Panel c demonstrates the overall percentage improvement in each subject following 3 months of EPI-743 treatment. Panels d and e demonstrate the change from baseline to 3 months in neuromuscular function as measured by the GMFM (D) and health related quality of life as measured by the PedsQL (E). Panel f demonstrates the one class improvement in each patient on the MD-CRS.
Finally, there was a significant improvement (15.0 ± 20.8; \( p = 0.02 \)) in quality of life score on the PedsQL neuromuscular module [15]. This significant improvement is consistent with the improvement observed in section IV of the NPMDS, which is focused on quality of life.

The patient (#8) who discontinued treatment at 16 weeks had an initial improvement in all clinical outcome measurements, but returned to baseline levels following cessation of EPI-743 therapy (Fig. 2) on the NPMDS, GMFM and MD-CRS.

Fig. 2. Patient 8 clinical outcome. This patient was treated for 16 weeks with EPI-743 prior to therapy cessation by the child’s parents. The results of the NPMDS (a and b), the GMFM (c), the MD-CRS (d) demonstrate initial improvement followed by return to baseline following cessation of therapy. The PedsQL score improved following cessation of therapy (Fig. 2e).

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3.2. Pharmacokinetic data

Pharmacokinetic data are summarized in Table 3. EPI-743 was rapidly absorbed ($T_{\text{max}}$ 2–4 hours) and was distributed or eliminated rapidly with an estimated half-life of 3.9 to 5.6 hours. Plasma exposure to EPI-743 appeared to be dependent on body weight of the patients. Two patients with body weight of 7.9 and 9 kg showed $C_{\text{max}}$ and AUC 0–8 hour values of 1377 and 1167 ng/mL and 3011 and 4353 ng h/mL, respectively, whereas the patients with higher body weights (21–28.6 kg) had $C_{\text{max}}$ values of 144–215 ng/mL and AUC 0–8 values of 771–894 ng h/mL. Correcting the $C_{\text{max}}$ and AUC 0–8 for dose (mg/kg) resulted in a substantially smaller variability ($C_{\text{max}}$/dose or $AUC_{0-8}$/dose mg/kg range 10.7 to 36.25 ng/mL; AUC/dose mg/kg range 84.6 to 168.6 ng h/mL).

3.3. Safety and tolerability

Treatment with EPI-743 was found to be safe and well tolerated other than the one subject who had therapy discontinued for what parents reported to be increased somnolence.

Eight serious adverse events requiring hospitalization occurred in four patients during the 6-month trial. They were all due to underlying disease and not associated with drug: three cases of upper respiratory tract infection, one case of bronchopneumonia, one case of salmonellosis, one case of metabolic acidosis, and one case of an apneic spell. All adverse events resolved without sequelae. Two non-serious adverse events, one case of metabolic acidosis, and one case of an apneic spell. There were no clinical chemistry or laboratory abnormalities attributed to the drug.

3.4. Natural history

A total of 71 publications were identified and reviewed that described clinical data on children affected by six of the seven genetic causes of Leigh syndrome in our series. No ND 1 publications were identified. 44 of these 71 publications provided specific data on patient clinical course. Demographic and clinical outcome data were ascertainable on 180 children (Table 4 and Fig. 3). 99.4% of children either had progressive neurologic deterioration or died. There was report of only one child demonstrating signs of clinical improvement over time.

4. Discussion

The natural history of Leigh syndrome was recently reviewed by Finsterer and characterized as a pediatric progressive neurodegenerative disorder in which children rapidly deteriorate, resulting in death by five years of age [18]. Given the absence of any approved drugs, we undertook a therapeutic development program for this devastating medical condition.

EPI-743 is a rationally designed, catalytic, 2-electron transfer NQO1 cofactor [13]. The 2,3-dimethyl para-benzoquinone chemical nucleus was selected for lead optimization based on its favorable electrochemical coupling between NADPH, NQO1, and glutathione reductase. It possesses a redox potential of $-175$ mV that exists between the flavin redox potentials of NQO1 and glutathione reductase. Final structure optimization was directed at blood–brain-barrier penetration and oral bioavailability.

In this open label trial, 10 children with seven different genetic mutations consistent with Leigh syndrome were enrolled at a single European site. In order to ensure that these children all had evidence of moderate disease progression, a threshold of an NPMDS of greater than 15 was required for enrollment. Four outcome measures were used to capture the clinical spectrum of Leigh syndrome and response to EPI-743 treatment. These included the mitochondrial disease-specific NPMDS, the more generalized pediatric GMFM, the MD-CRS, and the PedsQL. A statistically significant improvement was shown in all four of these outcome measurements. Central nervous system and neuromuscular function improved following treatment with EPI-743 as assessed by the NPMDS Sections 1–3, the GMFM, and the MD-CRS. Consistent with improvement in physician-recorded outcome measures, patient families reported improvement in quality of life measures (Section 4 of NPMDS and the PedsQL), both of which were statistically significant. Improvement was independent of genetic determinant, age, sex, and disease severity. These results confirm the initial findings of Leigh syndrome subjects treated with EPI-743 under FDA-approved expanded access and physician-sponsored IND protocol in the United States and compassionate treatment in Europe. [12]

The clinical results obtained in this study were compared to an historical cohort obtained from the published natural history of Leigh syndrome over the last 12 years. In contrast to the frequency of the combined disease progression and mortality (179/180 = 99.4%), 100% of the EPI-743 treated subjects reversed disease progression and improved. These results occurred across each of the major Leigh syndrome genetic determinants (Table 4 and Fig. 3). While these cohorts are not directly comparable due to the fact that children in the study were followed for only 6 months, the results do confirm the progressive natural history of Leigh syndrome and the fact that there is only one documented case of spontaneous clinical improvement over time.

In addition to efficacy parameters, both clinical and standard laboratory metrics of drug safety were prospectively assessed. No significant drug-related adverse events were recorded. Coupled with pharmacodynamic data, the safety data established herein demonstrate a favorable therapeutic index. Separately, as of August 1, 2012 an estimated 125 subjects with mitochondrial disease have been treated with EPI-743 with a cumulative patient exposure of $>42,000$ days, with only one reported possibly drug-related adverse event. Based on the collective experience of the treatment of subjects with genetically defined mitochondrial disease and data from non-clinical safety assessment studies, a safety margin of at least 10-fold for the 100 mg three times daily dosing has been assigned to EPI-743 [12,19,20].

One child in this study had EPI-743 discontinued following 16 weeks of therapy due to parental concerns over increased somnolence. The investigators deemed this symptom disease related. Review of this patient’s clinical data demonstrates an improvement in all clinical outcome measurements up to the point of therapy discontinuation, with a return to baseline (i.e. pretreatment) levels shortly after therapy cessation. These findings not only confirm the potential EPI-743 treatment benefit, but also suggest a change in clinical signs and symptoms that are drug-dependent.

There are several limitations to this study principally related to its open label design. First, given the lack of placebo group it is difficult to determine the rate of disease progression absent treatment over a 6-month period. However, while there are sporadic reports in the literature of
children with Leigh syndrome demonstrating signs of clinical improvement, the observation that all children on treatment demonstrated improvement on all outcome variables suggests a bona fide treatment effect. In addition, the physician performing the clinical outcome measurement assessments knew that all children were receiving treatment which potentially biases the results. Finally, given the rarity of Leigh syndrome and the absence of previous clinical trials in this population, there is a lack of well instantiated outcome measurements that have been appropriately validated to assess treatment effect.

5. Conclusion

In conclusion, EPI-743 treatment resulted in neurological and neuromuscular improvement in genetically defined Leigh syndrome subjects independent of genetic determinant and reverses disease progression—results that have not previously been recorded in a progressive mitochondrial disease. While these results are promising, they must be considered in the context of the study’s open label design. A randomized placebo controlled trial will be conducted to confirm the efficacy and safety findings.

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