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CASE REPORT

# Infantile Progressive Hepatoencephalomyopathy with Combined OXPHOS Deficiency due to Mutations in the Mitochondrial Translation Elongation Factor Gene *GFM1*

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**Abstract** Mitochondrial disorders are a heterogeneous group of often multisystemic and early fatal diseases caused by defects in the oxidative phosphorylation (OXPHOS) system. Given the complexity and intricacy of the OXPHOS system, it is not surprising that the underlying molecular defect remains unidentified in many patients with a mitochondrial disorder. Here, we report the clinical features and diagnostic workup leading to the elucidation of the genetic basis for a combined complex I and IV OXPHOS deficiency secondary to a mitochondrial translational defect in an infant who presented with rapidly progressive liver failure, encephalomyopathy, and severe refractory lactic acidemia. Sequencing of the GFM1 gene revealed two inherited novel, heterozygous mutations: a.539delG (p.Gly180AlafsX11) in exon 4 which resulted in a frameshift mutation, and a second c.688G > A (p.Gly230Ser) mutation in exon 5. This missense mutation is likely to be pathogenic since it affects an amino acid residue that is highly conserved across species and is absent from the dbSNP and 1,000 genomes databases. Review of literature and comparison were made with previously reported cases of this recently identified mitochondrial disorder encoded by a nuclear gene. Although limited in number, nuclear gene defects causing mitochondrial translation abnormalities represent a new, rapidly expanding field of mitochondrial medicine and should potentially be considered in the diagnostic investigation of infants with progressive hepatoencephalomyopathy and combined OXPHOS disorders.

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## Introduction

Mitochondria are ubiquitous intracellular organelles present in virtually all eukaryotic cells. They are relics of an ancestral alpha-proteobacterial endosymbiont (Gray et al. 1999) that took permanent residence in our cells. Genetic disorders of mitochondrial respiratory chain are the most common group of inborn errors of metabolism, collectively affecting approximately 1 in 5,000 births (Skladal et al. 2003). Mitochondrial dysfunction encompasses an extraordinary assemblage of clinical phenotypes, commonly

manifesting in tissues with high energy requirements, such as brain, retina, heart, muscle, liver, and endocrine systems (Cwerman-Thibault et al. 2011). It has also been implicated in a variety of diseases, including common multifactorial disorders such as diabetes (Gerbitz et al. 1996), Parkinson's disease (Mizuno et al. 1995; Mandemakers et al. 2007), and cancer (Brandon et al. 2006).

The complex and intricate nature of the oxidative phosphorylation (OXPHOS) system, which consists of about 90 proteins encoded by both the nuclear and the mitochondrial genome, explains the clinical heterogeneity associated with genetic defects in OXPHOS (Munnich et al. 1992). In this report, we describe an infant presenting with early fatal hepatoencephalopathy resulting from combined deficiencies of complex I and IV due to mutations in a nuclear gene encoding mitochondrial translational factor EFG1.

## Case Report

The patient was the second child of a healthy, non-consanguineous Chinese couple, born at term via Cesarean section for intrauterine growth retardation and abnormal lie. The Apgar scores were good. Her weight of 2.03 kg and her head circumference of 30 cm were both below the third percentile, while her length was 49 cm, at the 25th percentile. On day 2 of life, she was noted to be lethargic, tachypnoeic, and hypoglycemic, with a venous blood sugar of 1.7 mmol/L. She had mild hyperammonemia, 167 μmol/L (normal <50), refractory raised anion gap metabolic acidosis (~22–30) with a pH of 6.8, greatly elevated serum lactate of 17–25 mmol/L (normal <2.4), and CSF lactate of 12 mmol/L (normal <2.1). She was ventilated and had single volume exchange transfusion followed by peritoneal dialysis performed in an effort to control the acidosis. Her urine organic acids showed excessive excretion of lactate, 3-OH butyric, 2-OH butyric acids, 4-OH phenyllactate, and ketonuria. Her acylcarnitines were normal and her serum amino acids revealed elevated alanine at 1,228 μmol/L (normal 122–546), glutamine 1,114 μmol/L (normal 59–561), methionine 128 μmol/L (normal 10–79), phenylalanine 165 μmol/L (normal 31–157), and tyrosine 569 μmol/L (normal 5–167). Liver function test showed hypoproteinemia (protein 42 and albumin 24 g/L), raised alkaline phosphatase 516 IU/L (age-related reference range 0–341), gamma-glutamyl transferase 523 IU/L (normal 11–50), mildly elevated alanine aminotransferase 66 U/L (normal 0–54), aspartate aminotransferase 86 U/L (normal 0–82), and total bilirubin of 108 μmol/L (normal 0–17). She responded to peritoneal dialysis with normalization of acidosis and a reduction of serum lactate to 6 mmol/L. Echocardiogram was normal and ultrasound brain showed dilated ventricles. She had notable generalized hypotonia with myopathic facies and subtle dysmorphism including flat nasal bridge, low-set ears, high, broad forehead, and smooth philtrum. She was suspected to have a possible mitochondrial DNA (mtDNA) depletion syndrome and commenced on oral Coenzyme Q 10, oral thiamine, riboflavin, biotin, and vitamin E. She was discharged home at 2 weeks of age on breast feeding with normal blood gas and lactate.

Oral sodium bicarbonate was supplemented at 4 weeks of life with the recurrence of systemic acidosis and lacticacidemia. She developed persistent vomiting and steatorrhoea. Clinically an enlarged liver was palpable 5 cm below the right costal margin. Progressive deterioration of liver function occurred with conjugated hyperbilirubinemia (direct bilirubin 70 μmol/L), total 134 (normal 0–17), worsening hypoalbuminemia, and transaminemia.

In the following months, she remained hypotonic with failure to thrive and globally delayed developmental milestones. She was readmitted at 10 weeks of age with escalating lethargy and inactivity precipitated by recurrent vomiting and loose stools. Clinically, she demonstrated features of circulatory compromise with cold, cyanosed peripheries. Biochemical parameters included markedly raised lactate at 15 mmol/L and compensated metabolic acidosis (pH 7.35, bicarbonate 12.7, base excess −13). Brain MRI showed global cystic changes in the subcortical white matter, T2 hyperintensities in the putamen, globus pallidi, and ventricular dilatation with septatation (Fig. 1a–c). She was discharged home after several days with anti-reflux infant formula, oral ranitidine, and domperidone.

Shortly thereafter, she was readmitted with a similar episode of recurrent vomiting and systemic acidosis, and was subsequently referred to a tertiary center for further management. Features noted at initial assessment here showed an emaciated, pale, and mildly jaundiced infant with a weight of only 2.9 kg at 4 months of age. She had reduced spontaneous movements, hypotonia with severe head lag, myopathic facies, and reduced deep tendon reflexes. Intermittent roving nystagmus and mild ptosis were also noted. A soft ejection systolic murmur was auscultated over her upper left sternal edge. Echocardiography showed a small, 0.21 cm, atrial septal defect, with good contractility and ejection fraction of 65.8%. Her liver was palpable 6 cm below the right costal margin. Her hemoglobin was 9.5 g/dL with normochromic normocytic cells on film and adequate reticulocyte response. The other cell lines were normal. Hepatic dysfunction was evident with mild coagulopathy; activated partial thromboplastin time 50 s (control 30.9–45.9 s), prothrombin time 19 s (control 11.9–14 s), serum albumin 27 g/L (normal 30–54), and mildly elevated transaminases; alanine aminotransfer-

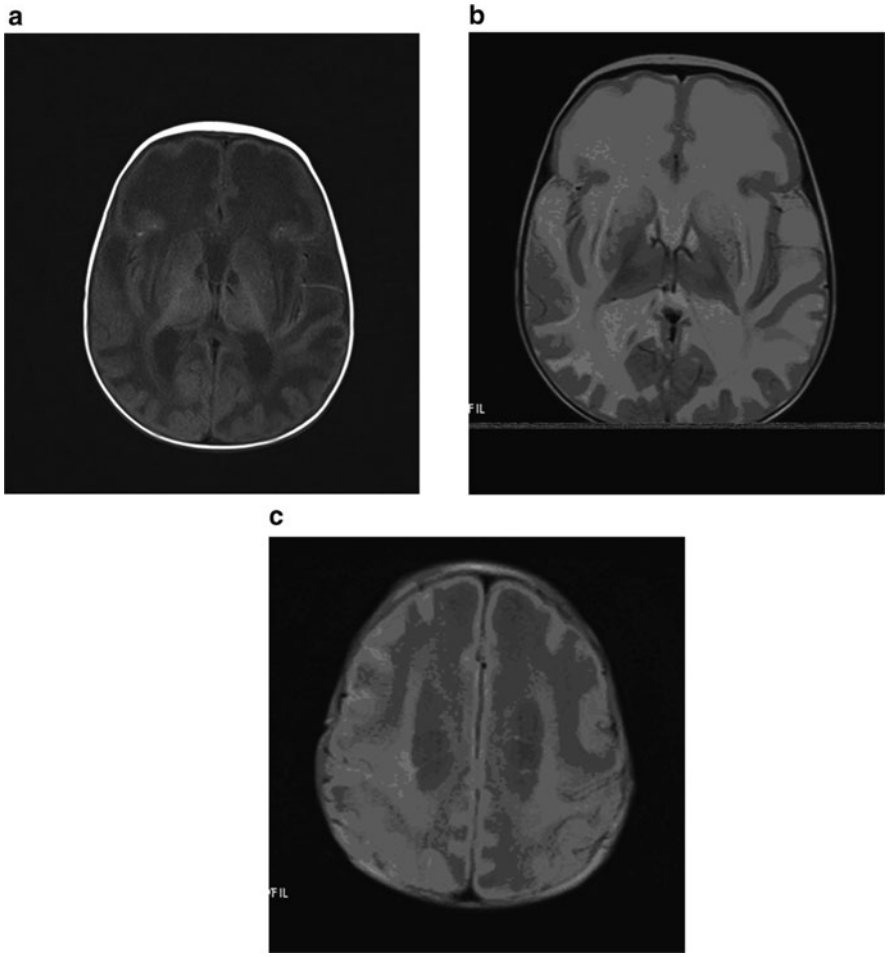


**Fig. 1** (**a**, **b**) MRI brain T1 and T2 weighted axial images showing global, cystic changes and hyperintensities in putamen and glubus pallidi. (**c**) MRI brain showing subcortical cystic changes and dilated lateral ventricles with ventricular septae

ase 170 U/L (normal 0–33), aspartate aminotransferase 116 (normal 0–82). Total bilirubin was 115 μmol/L (normal 0–17) with conjugated bilirubin 90 μmol/L, raised alkaline phosphatase 954 U/L (age-related reference range 0–356), gamma-glutamyl transferase 289 U/L (normal 11–50), and ammonia 65 μmol/L (normal <50). Serum lactate was 17.7 mmol/L, pyruvate 445.6 μmol/L (normal 30–80), blood pH 7.151, $pCO_2$ 12.1 mmHg, $pO_2$ 168.8 mmHg, bicarbonate 4, and base excess −24.5. Serum creatine kinase, very long chain fatty acids, carnitine, and transferrin isoforms were normal. Plasma alanine and proline were raised at 1,116 μmol/L (normal 132–455) and 508 μmol/L (normal 77–329), respectively. Urine organic acids showed increased excretion of lactate, ketones, citramalic, and fumaric acid.

Abdominal ultrasound showed a left multicystic kidney with a dilated distal left ureter and enlarged right kidney with bipolar length of 5.5 cm. The liver was enlarged with a smooth outline and absence of any focal lesions. Repeat brain ultrasound revealed multiple cystic changes predominantly observed in bifrontal lobes, and periventricular regions with ex-vacuo dilatation of the lateral ventricles. Her thyroid function studies were abnormal; free T4 12.1 pmol/L (14.1–19.2), TSH 7.55 mU/L (0.98–5.3). Ophthalmological assessment reported the presence of alternating exotropia with poor fixation, but normal fundus, possibly indicating cortical visual impairment. Treatment included attempts to correct electrolyte imbalances, metabolic derangements with oral sodium bicarbonate, and addressing feeding difficulties with nasogastric feeding of anti-reflux infant formula with supplemented medium chain triglycerides (MCT). Nevertheless her weight failed to pick up and at 8+ months of age, she only weighed 4.2 kg, well below the third percentile. She developed increasingly severe liver impairment, systemic metabolic and lactic acidosis. Her parents opted for a conservative management and she succumbed at home from respiratory failure at 8 months of age.

Meanwhile, further investigations were carried out to identify the precise cause of her illness. Biochemical examination cultured fibroblasts had demonstrated reduced respiratory chain enzyme activities (OXPHOS) of complex

I and IV with 68% and 47% residual activities expressed as a percent of the lowest control value respectively. The other OXPHOS enzymes complex II, complex III, and complex V showed a normal activity. Assays to quantify OXPHOS enzyme activities were based on spectrophotometry (Janssen et al. 2007; Mourmans et al. 1997; Cooperstein and Lazarow 1951; Jonckheere et al. 2008; Srere 1969). Muscle and liver OXPHOS assays would perhaps have yielded more representative results of the tissue-specific involvement observed in mitochondrial respiratory chain disorders; however, her parents had declined these tests due to concerns over her fragile state. Molecular genetic testing of a total of 12 point mutations in mtDNA isolated from blood for five genes associated with Leigh disease and seven genes associated with MELAS failed to demonstrate any pathogenic mutations. Postmortem liver biopsy (Figs. 2 and 3) and histopathology displayed lobular disarray, portal fibrosis, micro and macrovesicular steatosis, and intrahepatic and intracanicular cholestasis. Due to financial constraints, we were unable to analyze OXPHOS assays on postmortem liver tissue or perform direct sequencing of mtDNA. mtDNA depletion studies were not performed as the finding of reduced enzyme deficiencies in fibroblasts make a depletion syndrome less likely. Her parents had not consented for postmortem muscle biopsy. The associated biochemical evidence of a combined enzyme deficiency in cultured fibroblasts, in addition to the early, severe, and rapid progression of a predominant hepatocerebral disease in our patient, suggested a possible candidate nuclear gene defect such as *GFM1* mutation which had only recently been described in a handful of patients with a similar phenotype. Subsequent sequencing of *GFM1* gene was performed (Smits et al. 2010a) and revealed the heterozygous changes c.539delG (p.Gly180AlafsX11) in exon 4 which was inherited from the father, and c.688G > A (p. Gly230Ser) in exon 5, inherited from the mother.

## Discussion

OXPHOS disorders may occur as isolated enzyme deficiencies and have been reported in approximately 67% cases, while combined enzyme deficiencies account for 33% of all respiratory chain disorders (Smits et al. 2010b). Isolated OXPHOS deficiencies are generally caused by mutations in structural genes which encode subunits of the OXPHOS system, or in genes encoding proteins involved in the assembly of a specific OXPHOS enzyme complex (Zeviani and Di Donato 2004). Combined OXPHOS defects tend to involve genes required for mtDNA maintenance, mitochondrial transcription, or translation including posttranscriptional or posttranslational processes, import of nDNA-encoded proteins into the mitochondrion or mitochondrial membrane biogenesis (Smits et al. 2010b). Approximately 40% of all combined respiratory chain deficiencies occur as a result of mtDNA deletions and point mutations in mitochondrial transfer RNA genes, which more frequently affect adult patients (Kemp et al. 2011). Another 40% of

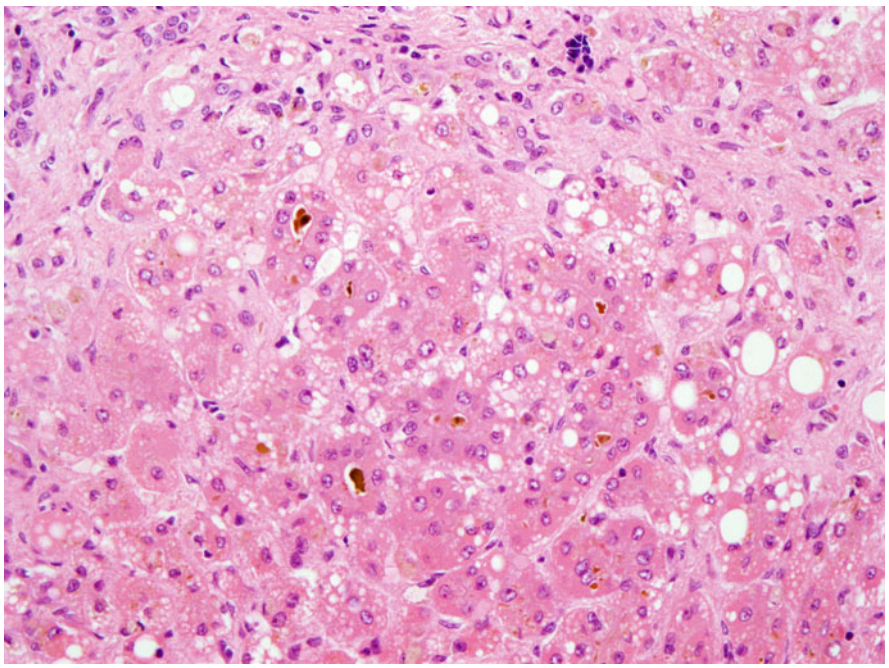

**Fig. 2** Liver H&E X40: Liver tissue displaying moderate microvesicular steatosis and patchy macrovesicular steatosis with intrahepatic and intracanicular cholestasis. The portal tracts are expanded with accompanying portal fibrosis and portal to portal bridging fibrosis

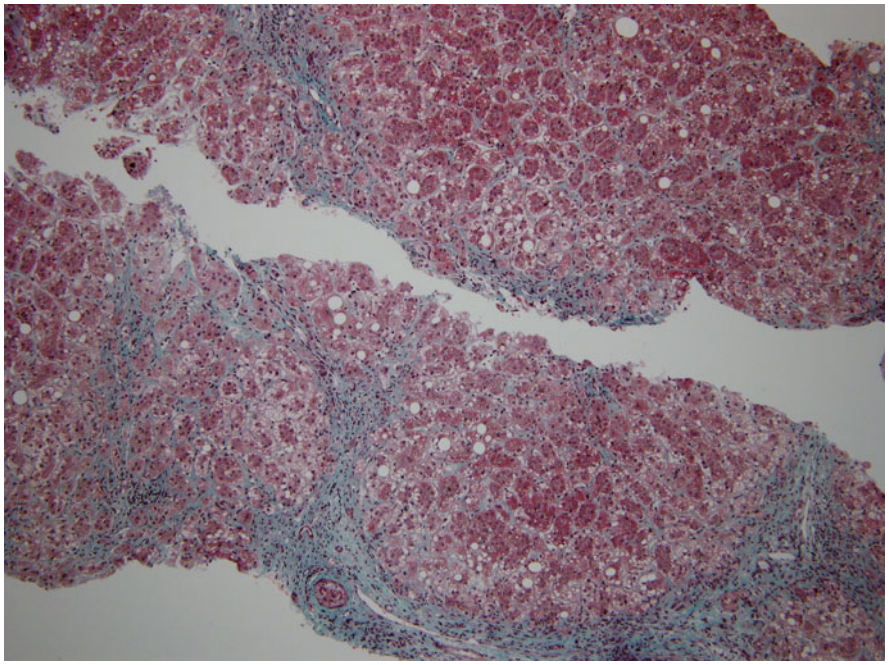

**Fig. 3** Liver MT X10: Lobular disarray of liver tissue is evident with the nodular pattern emphasized by Masson's Trichrome stain

combined OXPHOS deficiencies are related to mtDNA depletion, which predominantly affect young children and are caused by autosomal recessive mutations in nuclear genes (*DGUOK*, *MPV17*, *POLG*, *TYMP*, *TK2*, *SUCLA2*, *SUCLG1*, *RRM2B*, *PEO1*) influencing mtDNA replication and maintenance (Spinazzola et al. 2009). In the remaining 20% of combined respiratory chain deficiencies, after excluding mtDNA deletions, depletion, and point mutations, no clear diagnostic pathway is currently available to determine the cause of disease. The reduced enzyme activities in our patient's cultured fibroblasts made the possibility of mtDNA depletion less likely as it has been reported that exponentially growing cells may not manifest any OXPHOS enzyme deficiencies, but instead have to be kept in a quiescent state in order to clearly demonstrate their phenotype (Pontarin et al. 2011; Gonzalez-Vioque et al. 2011). For several types of depletion syndromes, it has been shown that nucleotide metabolism plays an important role in this phenomenon (Gonzalez-Vioque et al. 2011). We used growing fibroblasts with a low passage number, and under these conditions fibroblasts from depletion syndrome patients usually do not show OXPHOS enzyme deficiencies.

A thorough clinical characterization of patients is imperative in identifying homogeneous patient groups and analyzing the complex molecular mechanisms behind combined respiratory chain deficiencies. When facilitated by linkage studies in consanguineous families and functional cell culture investigations conducted at various regulatory levels of mitochondrial function, transcription, translation, ribosome function, protein stability, subcomplex formation, it may provide a potentially promising approach in selecting novel candidates in combined respiratory chain deficiencies (Kemp et al. 2011).

Eukaryotes contain two translational systems, one in the cytosol and the other in the mitochondria. The mitochondrial translation machinery comprises mtDNA-encoded rRNAs and tRNAs, in addition to various proteins encoded by the nuclear genome including two initiation factors, IF2 (*MTIF*) (Ma and Spremulli 1995), IF3 (*MTIF3*) (Koc and Spremulli 2002); three elongation factors, EFTu (*TUFM*) (Ling et al. 1997), EFTs (*TSFM*) (Xin et al. 1995), EFG1 (*GFM1*) (Gao et al. 2001); four release factors, RF1 (*MTRF1*) (Zhang and Spremulli 1998), RF1a (*HMRF1L*) (Zhang and Spremulli 1998), *C12ORF65* (Antonicka et al. 2010), *ICT1* (Richter et al. 2010); and two recycling factors, RRF (*MRRF*) (Soleimanpour-Lichaei et al. 2007) and EFG2, which has been renamed RRF2 (*GFM2*) (Zhang and Spremulli 1998; Rorbach et al. 2008; Tsuboi et al. 2009); mitochondrial ribosomal proteins (MRPs); mitochondrial aminoacyl-tRNA synthetases, and methionyl-tRNA transformylase (Smits et al. 2010b). Although most components of the mitochondrial translation system are nuclear encoded, the majority of patients harboring mutations in nuclear genes are very limited. Most mutations associated with mitochondrial protein synthesis defects to date have been reported in mtDNA.

Recently, mutations in *GFM1*, previously known as *EFG1*, a nuclear factor of the mitochondrial translation machinery, have been described (Smits et al. 2010a;

Coenen et al. 2004; Antonicka et al. 2006; Valente et al. 2007). Mitochondrial *GFM1* is a five-domain GTPase that catalyzes the translocation step of mitochondrial protein synthesis, during which peptidyl-tRNA moves from the ribosomal acceptor (amino acyl or A) site to the ribosomal peptidyl (P) site following removal of the deacylated tRNA from the ribosomal (P) site to the exit (E) site. This results in the concomitant advancement of the mRNA by one codon and exposure of the next codon in the A site preparing for a new elongation cycle (Wintermeyer et al. 2004). The compilation of biochemical evidence of a combined enzyme deficiency in cultured fibroblasts of complex I and IV, which is a clear and well-known functional consequence of *GFM1* mutations (Smits et al. 2010a; Coenen et al. 2004; Antonicka et al. 2006; Valente et al. 2007), in addition to the severe and early onset presentation with rapidly progressive hepatocerebral disease in our patient suggested a possible candidate nuclear gene defect such as *GFM1* mutation which had only recently been described in a handful of patients with a predominant hepatic and/or neurological phenotype.

Sequence analysis of the complete coding region of the *GFM1* gene in our patient revealed the heterozygous changes c.539delG (p.Gly180AlafsX11) in exon 4 which was inherited from the father had resulted in a frameshift mutation, while a second c.688G > A (p.Gly230Ser) mutation in exon 5 was inherited from the mother. The pathogenicity of the second novel missense mutation was suggested by its highly conserved nature in all nine organisms of which a *GFM1* sequence could be found, and its absence in the dbSNP database. In addition, both these mutations were not present in the 1,000 genomes database which includes 60 Chinese genomes and 194 South-East Asian genomes.

Previously, four other *GFM1* defects have been described. A review of their clinical features and investigations has been tabulated in Table 1. The first report of two siblings of consanguineous Lebanese parents harbored homozygous mutations, c.521A > G in the GTP-binding domain I (Coenen et al. 2004). The index patient had presented with intrauterine growth retardation, mild microcephaly, hypertonicity with reduced spontaneous movements, and she soon developed profound metabolic acidosis and lactic academia with raised lactate: pyruvate ratio of 38 (normal 12–18) by day 10 of life. Rapidly progressing liver failure occurred leading to death on day 27 of life.

Antonicka and colleagues described similar clinical phenotypes in two siblings with significant growth retardation, lactic acidosis, fatal hepatopathy, small or undeveloped corpus callosum, and severe respiratory deficiency in skeletal muscle and liver, without any evidence of clinical cardiac dysfunction (Antonicka et al. 2006). The molecular basis for the novel mutations in *GFM1* demonstrated that the translation defect resulting from these mutations was associated with unique, tissue-specific patterns of OXPHOS deficiency. The severity of the OXPHOS defect correlates with the residual levels of the mutant EFG1 protein in different tissues. In addition, there appears to be an adaptive response that involves transcriptional upregulation of EFTu, another translation elongation factor, with significant quantitative differences in the ratios of the translation elongation factors. This may reasonably reflect the different tissue-specific demands for the mitochondrially encoded polypeptides and explain how the heart uniquely possesses mechanisms to circumvent the disruption of mitochondrial translation caused by mutations in the nuclear-encoded components of the translation machinery.

A subsequent report by Valente and colleagues on an infant affected by neonatal lactic acidosis, rapidly progressive encephalopathy, severely decreased mitochondrial protein synthesis, and combined OXPHOS deficiency was a compound heterozygote for two novel *GFM1* mutations (Valente et al. 2007). No data are available concerning the expression of mitochondrial *GFM1* in human brain, but the predominant neurological involvement with neuroradiological hallmarks of early onset Leigh syndrome including bilateral necrotizing lesions in the basal ganglia and brain stem overwhelmed, that of other organs notably, skeletal muscle, liver, and heart in this patient. It is postulated that the clinical variability may be attributed to various mechanisms, including the different sites of mutations in the EFG1 protein which may subsequently have tissue-specific effects on mitochondrial translation, adaptive processes (e.g., compensatory changes in other translational factor) which may act differently in different patients, and finally partial compensation by the second isoform of mitochondrial *EFG*, namely *EFG2* which is highly expressed in energy-consuming tissues, such as skeletal muscle, heart, and fetal liver. The functional role of *EFG2*, however, is uncertain (Smits et al. 2010a).

Strikingly, in the recent report by Smits et al., a mutation in *GFM1* was observed in a patient affected by severe, rapidly progressive mitochondrial encephalopathy; the decrease in enzyme activities of complex I, III, and IV detected in fibroblasts was not found in muscle tissue (Smits et al. 2010a). Reduced respiratory chain activities in fibroblasts with normal values in muscle tissue are an uncommon finding in mitochondrial disorders, whereas the opposite is often observed. The capacity of the mitochondrial energy-generating system, however, was clearly reduced in muscle tissue, indicated by impairments in pyruvate oxidation and ATP production rates. This evinces the importance of a thorough diagnostic biochemical analysis of muscle tissue and fibroblasts in patients as OXPHOS defects may be selectively expressed.

The clinical symptomatology in our patient is reminiscent of the severe phenotype of the first two reports

**Table 1** Clinical features, biochemical analysis of OXPHOS assay on liver tissue, skeletal muscle and fibroblasts, mutation testing of *GFM1* gene and neuroradioimaging of the six patients diagnosed to date in comparison to our patient

| | Clinical features | OXPHOS assay on fibroblasts | OXPHOS assay on liver tissue | OXPHOS assay on muscle tissue | *GFM1* mutation (cDNA level) | Brain imaging |
|---|---|---|---|---|---|---|
| Patient 1[a] | Female index patient, born to consanguineous parents with intrauterine growth retardation, mild microcephaly, and hypertonicity. Rapidly progressive liver failure with initial liver dysfunction from day 7, profound metabolic and lactic acidosis from day 10, and early death on day 27 of life.Blood lactate – 17.1 mmol/L, Lactate:pyruvate-38 | Complex I – 40%<br>Complex III – 69%<br>Complex IV – 18% | Not done | Complex I – 52%<br>Complex IV – 54% | Homozygousc.521A > G mutation | MRI-hypoplasia of the corpus callosum with several symmetrical cystic lesions in the white matter in the area of the basal ganglia |
| Patient 2[a] | Male sibling with extremely delayed growth and development, as well as increased muscle tone in his upper extremities. Signs of liver failure were present at week 7, leading to death at 5 months of ageBlood lactate – 9.3 mmol/L, Lactate:pyruvate-84 | Complex I – 13%<br>Complex IV – 31% | Not done | Not done | Homozygousc.521A > G mutation | Ultrasound brain demonstrated generalized atrophy and a small corpus callosum |
| Patient 3[b] | Female neonate with growth retardation, lactic and metabolic acidosis, rapidly progressive fatal hepatopathy on day 9 of life from pulmonary hemorrhage. Patent ductos arteriosus presentBlood lactate – 34 mmol/L | Complex I – 20%<br>Complex III – 40–60%<br>Complex IV – 20%<br>Complex V – 40–60% | Complex I <10%<br>Complex IV < 10%<br>Complex V – 50% | Complex I – 50%<br>Complex IV – 20%<br>Complex V – 20% | c.1068T > C;<br>c.1872-2 del AG | Brain MRI was normal on day 5 but MRS showed marked elevation of lactate |
| Patient 4[b] | Termination of pregnancy for IUGR and significant oligohydramnios. The baby died 45 min after birth. Amniocentesis 15 weeks – 45X (4)/46XX (16) karyotype in cultured cells, with inconclusive analysis of lactate:pyruvate ratio, COX activity, immunocytochemistry, and western blots | Complex I – 30%<br>Complex III – 60%<br>Complex IV – 10%<br>Complex V – 50% | Not done | Not done | Not done | Not done |
| Patient 5[c] | Dysmorphic features, including flat nasal bridge, low-set ears, small hands and feet, epicanthus, and high, arched palate noted at day 7 of life. At age 3 weeks, she started having feeding difficulties, weight loss, and subsequently had rapidly progressive encephalopathy. She passed away at 16 months from respiratory insufficiency | Complex I – 10%<br>Complex II – 25%<br>Complex IV – 53% | Not done | Complex I – 4%<br>Complex II – 17%<br>Complex III – 58%<br>Complex IV – 17%<br>Complex V – 35% | c.139C > T;<br>c.1478T > G | MRI brain showed two large, bilateral, and symmetrical areas of increased T2 involving the putamen and the globus pallidus |

**Table 1** (continued)

| | Clinical features | OXPHOS assay on fibroblasts | OXPHOS assay on liver tissue | OXPHOS assay on muscle tissue | *GFM1* mutation (cDNA level) | Brain imaging |
|---|---|---|---|---|---|---|
| Patient 6[d] | Female patient, second of a dizygotic twin, small for gestational age presented at day 2 of life with feeding problems, encephalopathy, and hypotonia. Seizure onset at 8 weeks of life with microcephaly, delayed visual maturation. Progressive neurological deterioration occurred and succumbed at 2 years of age from pneumoniaBlood lactate – 4.9 mmol/L, Lactate:pyruvate-22 | Complex I – 34 mU/U CS (reference range 100–310 mU/U CS)Complex III – 1,038 mU/U CS (reference range 1,320–2,610 mU/U CS) Complex IV – 168 mU/U CS (reference range 1,320–2,610 mU/U CS) | Not done | Complex III – 2,151 mU/U CS (reference range 2,200–6,610 mU/U CS) | Homozygous c.748C > T | Brain MRI revealed a small frontal cortex, a thin corpus callosum and delayed myelination |
| Patient 7[e] | Intrauterine growth retardation, severe metabolic and lactic acidosis day 2 of life, with progressive liver failure. Some dysmorphic features noted, atrial septal defect, abnormal kidneys | Complex I – 68% 112 mU/U CS (reference range 163–599 mU/U CS)Complex IV – 47% 136 mU/U CS (reference range 288–954 mU/U CS) | | | c.539delG; c.688G > A | Brain MRI showed globally extensive cystic changes in subcortical white matter. Increased T2 of putamen, globus pallidi, and ventricular dilatation with septations |

[a] Siblings described by Coenen et al. 2004

[b] Siblings described by Antonicka et al. 2006

[c] Valente et al. 2007

[d] Smits et al. 2010a

[e] Our patient

(Coenen et al. 2004; Antonicka et al. 2006), characterized by intrauterine growth retardation, profound lactic acidosis, and progressive liver dysfunction, resulting in liver failure and death within the first weeks or months. In addition, our patient had a multicystic kidney and an atrial septal defect. Neuroradiological evidence of cystic changes in temporal lobes with white matter abnormalities and biochemical findings of high blood lactate levels of 25 mmol/L and lactate: pyruvate ratios of 38 suggest a more global and deleterious disturbance of mitochondrial functioning.

In summary, unique patterns of OXPHOS deficiency result when one of the nuclear-encoded components of the translation system fail. The clinical heterogeneity associated with mutations in the mitochondrial translation apparatus may reflect different tissue-specific demands for the mitochondrially encoded polypeptides with significant quantitative differences in the ratios of the translation elongation factors. Despite rapid advancements in our understanding of the mechanisms implicated in mitochondrial disease, it is far from complete. The intricacy and complexities of the OXPHOS system renders the identification of the genetic defect an arduous task. Rapid advances in technologies involving high-throughput sequencing which are much cheaper and faster than the conventional approach of polymerase chain reaction followed by capillary sequencing (Tucker et al. 2009) or exome sequencing (Ng et al. 2009) which is the targeted sequencing of all protein-coding regions when applied with appropriate bioinformatics tools potentially offer a promising approach in elucidating the genetic etiology of OXPHOS deficiencies. Clearly, much work remains to be done to fully comprehend the processes involved in mitochondrial translation and the biogenesis of the OXPHOS system and their roles in the pathogenesis of combined OXPHOS deficiencies.

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## Contributions of Authors

1. Balasubramaniam S – Clinical management of the patient, draft of manuscript, and completed version.
2. YS Choy – Clinical management of patient, review of the draft, and contribution to the completed article.
3. Talib A – Histopathological analysis of liver biopsy, review of the draft, and contribution to the completed article.
4. Norsiah MD – Molecular testing of parental DNA for GFM1 mutation, review of the draft, and contribution to the completed article.
5. van den Heuvel LP – Biochemical analyses of OXPHOS assay and molecular testing in the proband, review of the draft, and contribution to the completed article.
6. Rodenburg RJ – E Biochemical analyses of OXPHOS assay and molecular testing in the proband, review of the draft, and contribution to the completed article.

## One Sentence Take Home Message

This report demonstrates a combined OXPHOS deficiency detected in patient fibroblasts, occurring as a result of a nuclear-encoded mitochondrial translational defect secondary to *GFM1* mutation, ultimately leading to mitochondrial hepatoencephalomyopathy and death at 8 months of age.

All authors declare that the answers to all questions on the JIMD competing interest form are “no” and therefore have nothing to declare.

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