Somatic Mosaicism for a Novel *PDHA1* Mutation in a Male with Severe Pyruvate Dehydrogenase Complex Deficiency: Case Report and Literature Review

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ABSTRACT and BACKGROUND

The mitochondrial multienzyme pyruvate dehydrogenase complex (PDC) is the gateway for oxidative metabolism of carbohydrate, catalyzing oxidative decarboxylation of pyruvate into acetyl-CoA as the primary substrate for the tricarboxylic acid cycle and oxidative phosphorylation. Mutations of the PDH-E1α subunit, encoded by the X-linked *PDHA1* gene, are responsible for the majority of PDC deficiency cases. Hemizygous males with a deleterious *PDHA1* gene mutation may be clinically more severely affected (often with lethality in infancy), while females with the same mutation may have more variable clinical manifestations and greater survival. Somatic *PDHA1* mosaicism has been documented only in a few patients, who usually manifest milder or attenuated phenotypes.

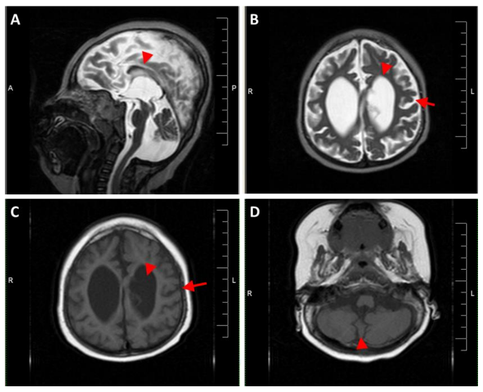
The patient is a 4 ½ year old boy with a clinical history of congenital microcephaly, significant brain anomalies including agenesis of the corpus callosum, colpocephaly, hypoplasia of the vermis and Dandy-Walker variant, persistent seizures, with profound developmental delay and failure to thrive. He exhibited persistently elevated blood lactate, pyruvate, and alanine with normal lactate:pyruvate ratio, suggestive of PDC deficiency. Family history was non-contributory.

Activity of PDC in cultured skin fibroblasts (SF) from this patient (0.63 and 0.76 nmol/min/mg protein) was below the reference range (mean 2.42; range 1.26-4.42, n = 329), corresponding to 26% and 31% of the mean, respectively. Mutational analysis by Sanger sequencing of the entire *PDHA1* gene from fibroblast DNA of the patient confirmed the clinical and biochemical findings of PDC deficiency. The patient was found to be mosaic for a novel, unclassified missense variant, c.523G>A (p.A175T). The c.523G>A missense mutation results in a substitution of a highly conserved alanine to threonine at position 175 of the E1α subunit. Interestingly, both the mutant adenine (A) allele and the wild-type guanine (G) allele were observed at the c.523 position, suggesting mosaicism in the fibroblasts of this patient. Subsequent sequencing of DNA from peripheral blood and buccal cells from this patient also revealed the same mosaicism for c.523G>A, but of varying degree depending on the cell-type analyzed. Molecular analysis of the *PDHA1* gene from the peripheral blood and buccal cells of the proband's mother did not reveal the same change, suggesting a *de novo* event in this patient.

We reviewed reported cases of somatic *PDHA1* mutation mosaicism in both males and females. This is the seventh reported case of male somatic mosaicism for a *PDHA1* mutation, but notably with a significantly severe clinical phenotype.

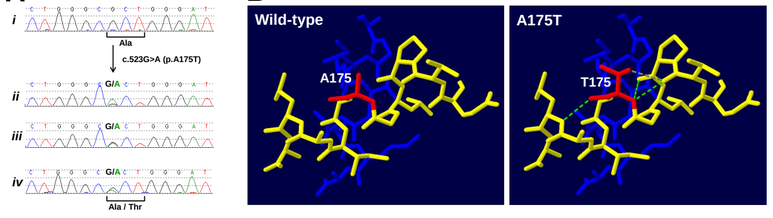
RESULTS

1 MRI showing several brain abnormalities



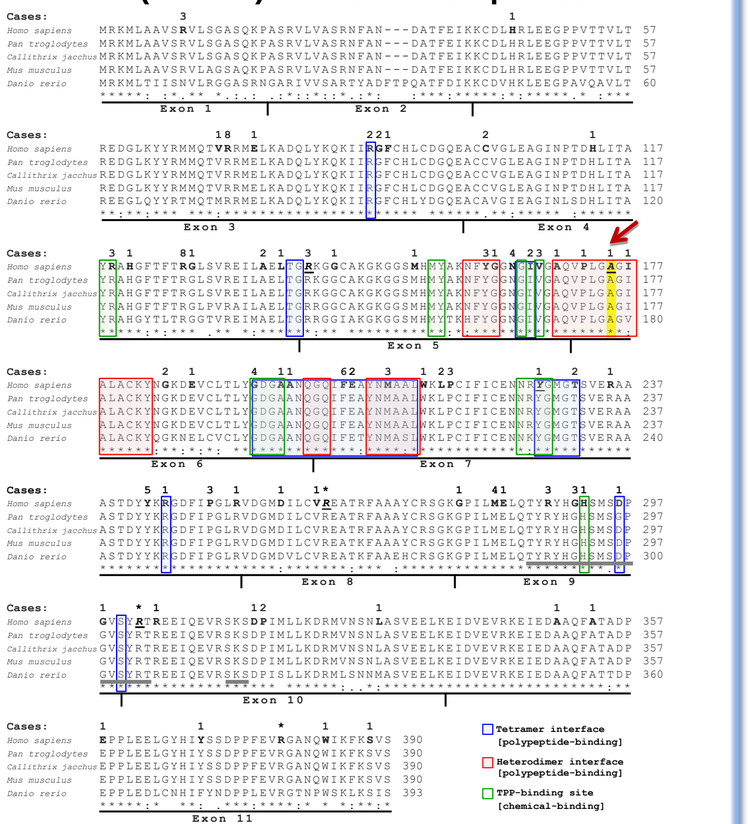
A. T2 sagittal image: severely hypoplastic corpus callosum (arrowhead). **B and C.** T2 and T1 axial images, respectively: ventriculomegaly (arrowhead) and loss of volume of the brain parenchyma with marked prominence of the cortical sulci (arrow). **D.** T1 axial image: component of vermian hypoplasia (arrowhead), consistent with Dandy-Walker variant. No abnormalities were noted in the basal ganglia and there was absence of the cavum septum pellucidum (not shown here).

2 Sequence analysis and protein structure prediction of *PDHA1* gene with c.523G>A (p.A175T) mutation



A. Sequence chromatograms of *PDHA1* showing a normal sequence in the proband's mother's blood (*i*); mosaicism for c.523G>A (p.A175T) in the proband's cultured skin fibroblasts (*ii*); the proband's blood (*iii*); and the proband's buccal cells (*iv*). **B.** *In silico* prediction of altered protein structure of *PDHA1* c.523G>A (p.A175T) mutation (red) based on Protein Data Bank entry 3EXE (Swiss-pdbViewer 4.1.0, http://spdbv.vital-it.ch). Side-chains and polypeptide backbone of E1α (yellow) and E1β (blue) subunits. Hydrogen bonds are shown in green.

3 Pyruvate dehydrogenase E1α subunit (PDHA1) amino acid sequence



The pyruvate dehydrogenase E1α-subunit (PDHA1) is highly conserved through evolution (*Pan troglodytes*, chimpanzee; *Callithrix jacchus*, marmoset; *Mus musculus*, mouse; and *Danio rerio*, zebrafish). Amino acids with reported missense/nonsense mutations are shown (**bold**) with the number of reported cases noted; greater than 10 reported cases are noted with an asterisk. Amino acids in which ***mosaicisms*** are reported are italicized and underlined. The tetramer (**blue**) and heterodimer (**red**) peptide binding interfaces, TPP-binding site (**green**) and phosphorylation loop (underlined **gray**) are highlighted. The c.523G>A mutation in this proband (**red arrow**) results in a substitution of a highly conserved alanine (**yellow**) to threonine at position 175 in the heterodimer interface region of E1α subunit.

TABLE 1. Cases of *PDHA1* Somatic Mosaicism

Patient	Sex	Clinical Phenotype	Lactic Acidemia	PDC enzyme SF (%) §	PDC enzyme SM (%) §	PDC enzyme lymphocytes (%) §	PDHA1 mutation	Samples evaluated for PDHA1 mutation
Sedya *et al.* [3]	M	Neonatal lactic acidosis (neonatal lethality)	Severe	25-30	NA	NA	c.422G>T (p.R141L)	SF, liver, muscle
Biochard *et al.* [4]	M	Failure to thrive and axial hypotonia	Mild	62	NA	67	c.483C>T (p.Y161Y) del exon 5	Blood, hair
Mine *et al.* [5]	M	**MRI findings**: cortical atrophy and ventricular enlargement	Mild	55	NA	NA	c.787C>T (p.R263X)	Blood, SF, hair
Okijima *et al.* [6]	M	Delayed motor and cognitive development **MRI findings**: normal	Mild	33-53	37	68	c.592G>A (c.511_603del) del exon 6	Blood, SF, hair, buccal swab
Soares-Fernandez *et al.* and Quintana *et al.* [7]	M	Dysmorphic features **MRI findings**: colpocephaly, corpus callosum dysgenesis, increased diffusion in the white matter, and bilateral subependymal cysts.	Significant	169	28	NA	c.904C>T (p.R302C)	Blood, SF, muscle
Ridout *et al.* [8]	F	Able to walk and reasonable comprehension with limited speech **MRI findings**: ventricular dilatation, sulcal widening, and relative course gyral pattern	Mild	51*	NA	NA	c.900-1G>A (p.S300Rfs*32)	SF
Coughlin *et al.* [9]	M	Failure to thrive and developmental delay **MRI findings**: periventricular white matter volume loss, with ex vacuo dilatation of lateral and third ventricules, thinned corpus callosum, and slightly small brainstem	Significant	148	24,45	NA	c.679T>C (p.Y227H)	Blood, SF, muscle
Current proband	M	Seizures, failure to thrive and developmental delay **MRI findings**: hypoplastic corpus callosum, ventriculomegaly, volume loss of the brain parenchyma with marked prominence of the cortical sulci, and vermian hypoplasia	Significant	26,31	NA	NA	c.523G>A (p.A175T)	Blood, SF, buccal swab

SF, skin fibroblasts; SM, smooth muscle; NA, not available.
* Ridout *et al.* reported an overall PDC activity of 0.46 nmol/min/mg protein (normal range 0.7-1.1) [8].
§ PDC activity is the percent of the mean for the respective diagnostic laboratories.

CONCLUSIONS

- This case expands the mutation spectrum of mosaicism in *PDHA1* (Table 1).
- This patient has a severe clinical phenotype – congenital microcephaly, significant brain abnormalities (Fig. 1), persistent seizures, profound developmental delay, and failure to thrive.
- This patient is mosaic for a novel, missense mutation, c.523G>A (p.A175T) (Fig. 2), confirming the clinical and biochemical findings of PDC deficiency.
- The mutation burden is ~70% in the skin fibroblasts, and a lower mutation burden in blood or buccal cells (Fig. 2). The neurological severity is likely related to the mutational prevalence in brain cells.
- Alanine-175 residue is located in the α-helix containing the heterodimer interface of the E1α subunit (Figs. 2 and 3). Threonine at residue 175 may cause altered hydrogen bonding patterns which alters the E1α subunit protein backbone and impedes its interaction with the E1β subunit.

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