Coincidental omega-amidase deficiency: a novel inborn error of glutamine metabolism in a child with congenital generalised lipodystrophy type 2

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Introduction
A 3 month old boy was investigated for severe failure to thrive. A urine metabolic screen was requested as part of routine investigations. First-tier testing by tandem mass spectrometry revealed an apparent increase in Δ¹-piperideine-6-carboxylic (P6C), a marker for pyridoxine dependent epilepsy (PDE, OMIM 266100). This was puzzling as there was no history of seizures and neurodevelopment was age appropriate. Follow-up investigations are summarised in Results (next slide).

Video presentation:

A 3-month old boy with severe failure to thrive
A urine metabolic screen was requested as part of routine investigations:

Urine metabolic screen results: tandem mass spectrometry, arbitrary units, log-log scale:

- All samples, n =3086
- Patient
- PDE
Results

Clinical
- Fourth child of first cousin parents
- Weight <5th centile, length and head circumference ~75th centile.
- Pronounced musculature with little subcutaneous fat, a liver edge 9 cm below the right costal margin and a spleen 6 cm below the left costal margin, large hands and feet, hypertrichosis.
- Abdominal U/S: non-specific hepatic enlargement

Biochemical
P6C and pipecolic measured by LC-MSMS were normal, excluding PDE and suggestive of an interference in the initial urine metabolic screen.

Urine organic acids (Fig 1): Grossly increased 2-hydroxy-5-oxoproline (2H5OP), the major lactam form of 2-ketoglutaramic (2KGM) present in urine under physiological conditions, and a related dehydration product. Other peaks corresponded to 2-hydroxyglutaramic and 2-hydroxysuccinamic. These metabolites were undetectable or present at much lower levels in urines from control subjects.

Plasma: glutamine and ammonia levels were normal

Omega-amidase enzyme activity measured in the patient’s plasma was decreased (19% of controls).

Molecular
Targeted sequencing of NIT2 (NM_020202.4): homozygous exon 6 insertion/deletion c.449_467delinsGTA; p.(Leu150Argfs*27) variant. This causes a frameshift and downstream introduction of a stop codon, removing 101 amino acid residues from the omega-amidase protein. The protein truncation removes part of the omega-amidase enzyme active site (Cys153) and alters the dimer interface interactions. The patient’s variant is therefore predicted to be deleterious. The parents were each heterozygous for the same insertion/deletion.

Targeted sequencing of BSCL2 (NM_001122955.3): homozygous c.346_347dupTT; p.(Tyr117Serfs*40) variant, previously reported as pathogenic in congenital generalised lipodystrophy type 2.

FIG 1: Urine organic acid profile of the patient. Normal urine components are indicated in italics. Abnormal peaks are numbered: 1= unknown M=171; 2 = 2-hydroxy-5-oxoproline TMS; 3 = 2-hydroxy-5-oxoproline - H_2O TMS; 4= 2-hydroxy-5-oxoproline TMS; 5 = 2-hydroxysuccinamic TMS; 6 = 2-hydroxy-5-oxoproline - H_2O TMS; 7 = 2-hydroxyglutaramic TMS; 8 = unknown M=385
Discussion
On referral it was clear that the patient had a lipodystrophy phenotype which prompted targeted testing of BSCL2. This gene is associated with congenital generalised lipodystrophy type 2 and a homozygous, pathogenic variant was detected which accounted for his phenotype.

Further investigations of the biochemical abnormalities showed metabolites related to accumulating 2-ketoglutaramic, 2KGM. One of the metabolites has the same molecular weight as P6C, accounting for the interference in the first-tier urine metabolic screen.

2KGM is a substrate for omega-amidase (encoded by NIT2). This enzyme converts 2KGM to 2-ketoglutaric and ammonia in the glutaminase II pathway, a secondary pathway of glutamine metabolism1 (Fig 2). Targeted sequencing of NIT2 revealed a homozygous, likely pathogenic variant. Pathogenicity was confirmed by the finding of decreased omega-amidase activity in plasma. No patient with omega-amidase deficiency has been previously described to our knowledge.

The occurrence of two genetic disorders in our patient confounds a determination of the phenotype of omega-amidase deficiency. However, the fact that his phenotype is entirely accounted for by CGL2 suggests that omega-amidase deficiency has a mild, possibly benign, phenotype.

2KGM is a reactive molecule which has previously been suggested to play a role in the neurologic sequelae of ammonia intoxication syndromes.3 However, our patient was neurodevelopmentally normal. We did not have the opportunity to analyse CSF samples to check for CNS accumulation of 2KGM.

FIG 2: The glutaminase II and asparaginase II metabolic pathways. 1 = glutamine synthase; 2 = glutaminase. Several enzymes can catalyse the transaminase reaction.

Take-home messages:
- We describe omega-amidase deficiency, a novel inborn error of glutamine metabolism, caused by a NIT2 variant and readily recognised by an abnormal organic acid profile.
- The phenotypic description of omega-amidase deficiency awaits the description of further patients but our observations suggest it may be a mild condition.
- The occurrence of two genetic disorders in a consanguineous individual is not unusual — abnormalities that are not explained by a single gene defect are worthy of additional investigation.

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References