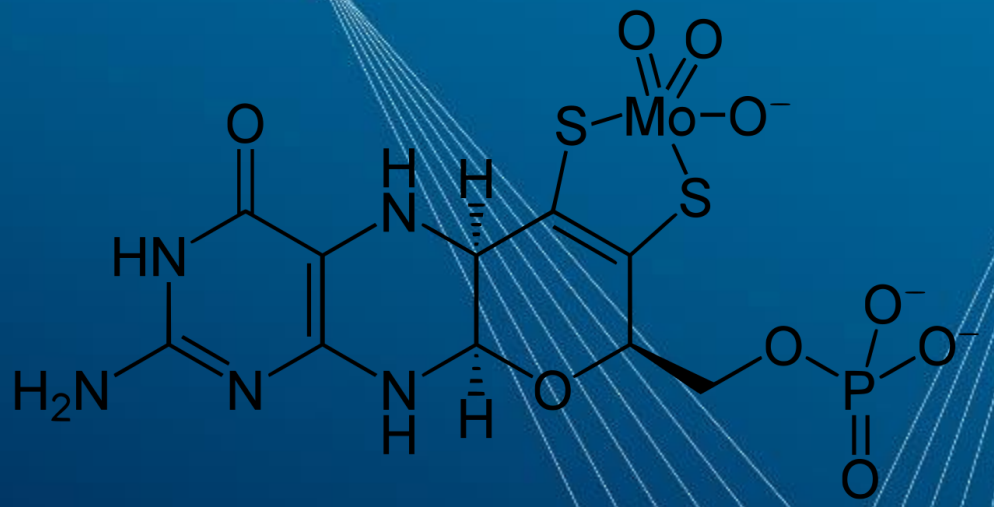
Molybdenum Cofactor deficiency – from diagnosis to clinical trial



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What we'll cover

1 | Case example
2 | Clinical presentation
3 | Biochemical diagnosis
4 | Analytical methods
5 | Genetics
6 | Management and outcome
7 | Treatment

Case presentation

- Male infant presented at 36 hours old to local A&E
    - Focal seizures
    - Limb cycling movements
    - High pitched cry
    - Parents noted he had not been feeding well
- History – no antenatal concerns, born 39 weeks gestation, no issues at delivery apart from hypospadia noted
- Two siblings, no concerns
- No parental consanguinity

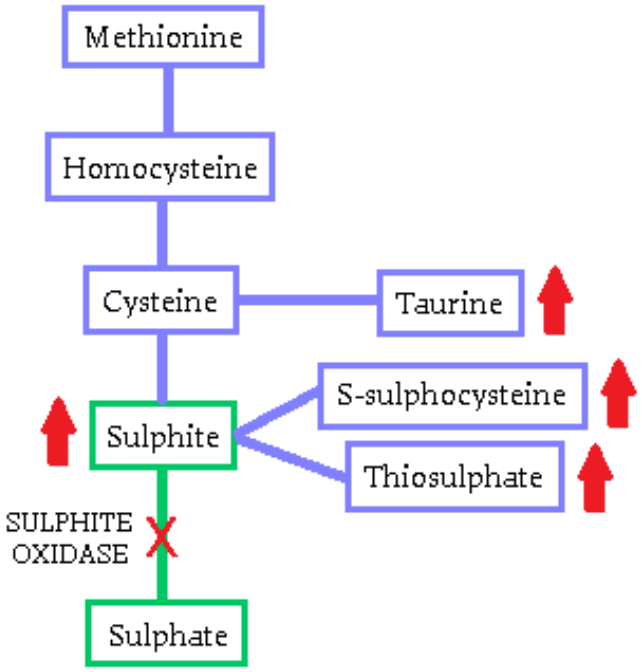
Initial management

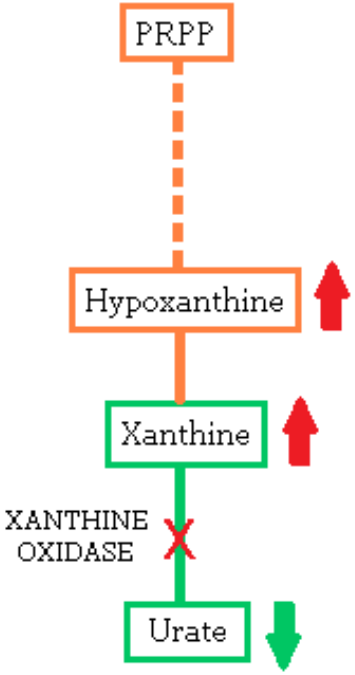
- Periodic apnoeas so intubated
- NG tube
- Prolonged EEG
    - Focal status epilepticus
    - Neonatal epileptic/developmental encephalopathy with burst-suppression
- Levetiracetam, phenytoin, pyridoxal phosphate, antibiotics, antivirals
- Transferred to PICU at the Evelina London on day 3

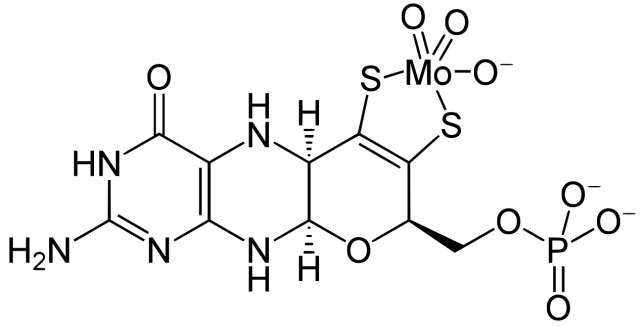
Investigations

- Head MRI –
    - Extensive abnormality with severe cytotoxic oedema throughout much of the cerebral cortex/subcortical white matter, and to a lesser extent deep grey matter involvement
    - Imaging differential includes a severe hypoxic ischaemic insult, sepsis and an underlying neurometabolic condition
- No immediately apparent cause on review of routine bloods and cultures
- Further samples included serum urate:
    - Urate = 0.06 mmol/L (RR 0.20 – 0.42)
- Clinical history + hypouricaemia → suspicion of MoCoD

Molybdenum Cofactor (MoCo)

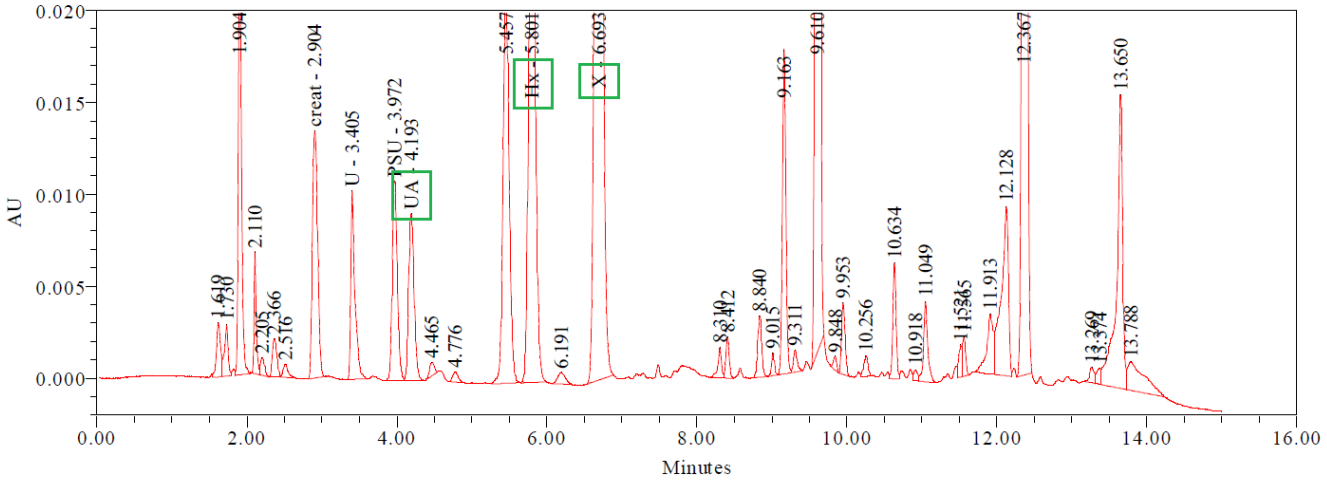






Metabolic results 1

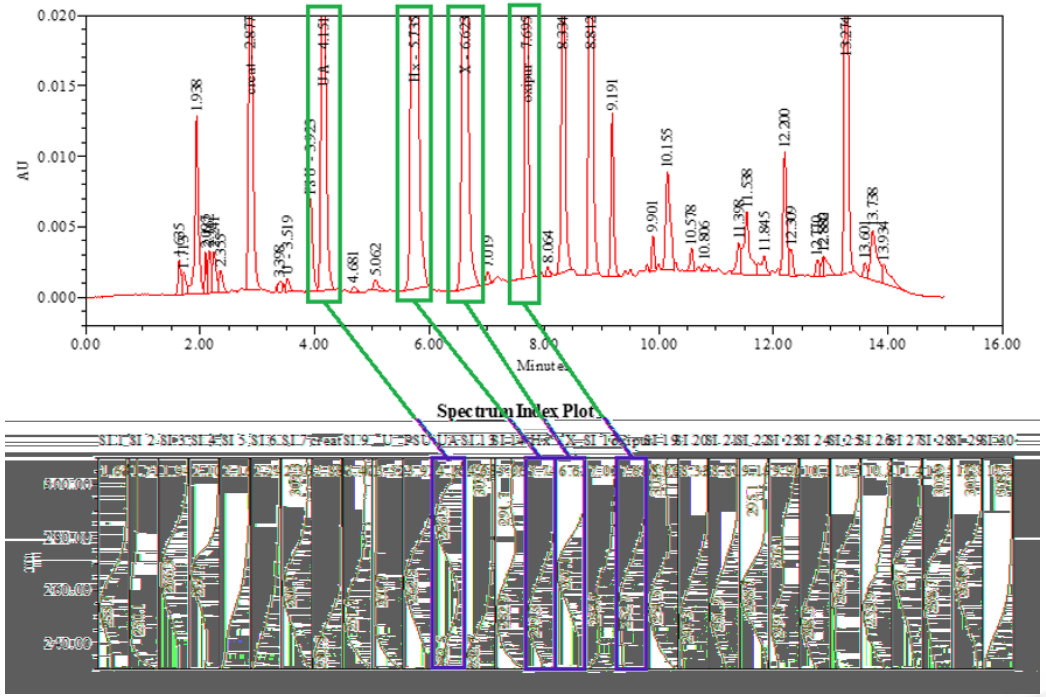
- Urine Purine and Pyrimidine results available on day 5

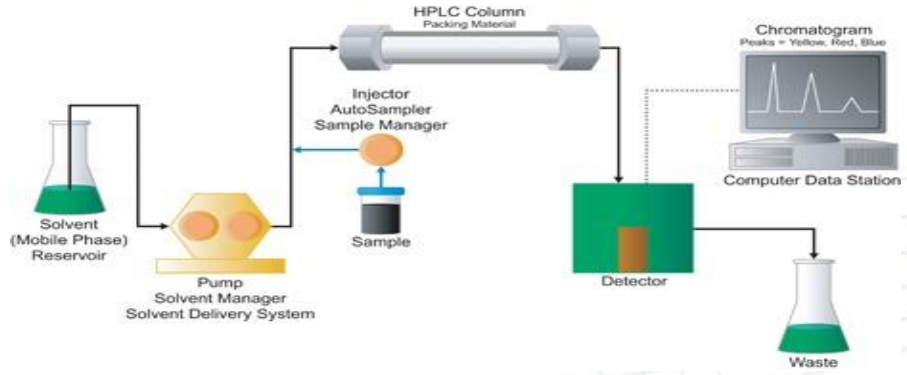


Test	Analyte	Result	Ref. range
Urine P&P screen	UA/Cr	0.23	0.30 – 1.50
	Xanthine	1.390 mmol/L	
	Hypoxanthine	0.385 mmol/L	
Sulphite dipstick		+++	
Urine Scys		73.0 uM/mM Cr	0.0 – 10.0

Purine analysis

- Urine, plasma, RBC and enzyme extracts run by Ultra Performance Liquid Chromatography (UPLC)
- Reverse phase (non-polar stationary phase, polar mobile phase)
- Photodiode array (based on UV/Vis spectrophotometry)
- Analytes identified based on retention time and characteristic absorption profile





Metabolic results 2

- Amino acids

Test	Analyte	Result	Ref. range
Urine AA (LC-MS/MS)	Scys	75 umol/mmol Cr	
	Taurine	101	<54
Plasma AA (LC-MS/MS)	Scys	20 umol/L	*(<20)*
	Taurine	163 umol/L	19 - 173
CSF AA (IEC)	Scys	Haemolysed Small peak	

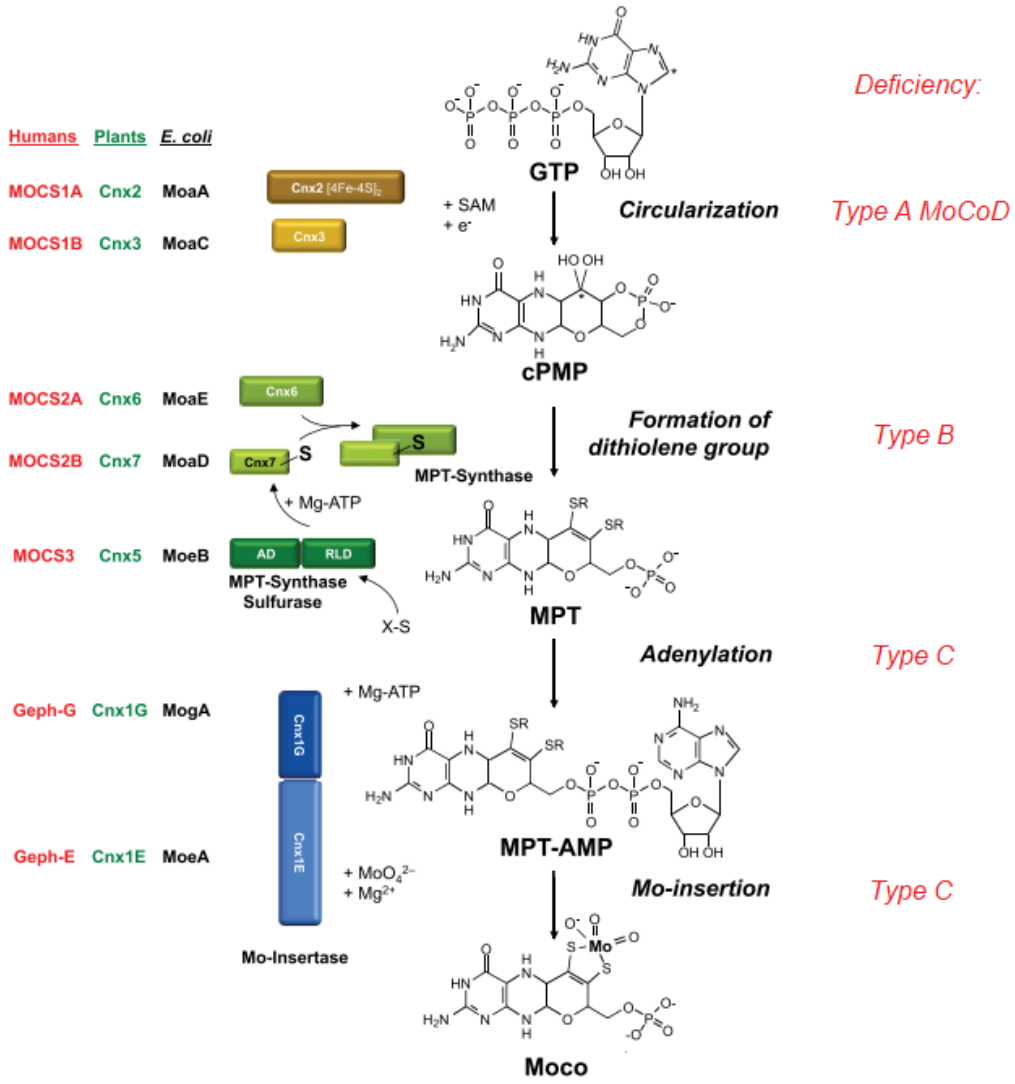
Amino acid (plasma)	Non-pathological decreases	Non-pathological increases
Taurine		Leukocyte/platelet contamination Sample deterioration Haemolysis Prolonged storage
Sulphocysteine	Delayed separation Delayed deproteinisation Serum sample	Sodium metabisulphite (IV/tube) Dietary
Cystine	Delayed separation Delayed deproteinisation Serum sample Sodium metabisulphite (IV/tube)	

Amino acid analysis

- LC-MS/MS and ion-exchange chromatography (IEC)

	Advantages	Disadvantages
LC-MS/MS	- 20 minute run time - Specificity – identification by mass and RT plus use of stable isotope internal standards - With improvements in analyser performance and availability of commercial kits – new 'gold standard' for *plasma* amino acid analysis	- Have to select what you're looking for - ? precision - Depending on model, not sensitive enough for CSF glycine
IEC	- Traditional 'gold standard' as in use for 40+ years - High sensitivity (e.g. CSF glycine) plus wide linear range - Stable and precise - Identifies all analytes of interest	- Unchanged for 40+ years - 2.5 hour run time - Lack of specificity – identification by RT only - Co-eluting compounds – phe/5-ALA, met/hcit, scys/phosphoserine - Interferences from drugs/exogenous sources - Single point calibration run infrequently

MoCo synthesis



- The unique tricyclic pterin molybdopterin is synthesised from GTP
- Molybdopterin + Mo → Molybdenum cofactor (MoCo)

Diagram from: The Molybdenum Cofactor, Ralf R Mendel (2013). *JBC*, **288(19)**, 13165-72

Genetic results

- Result available on day 6
- All exons and flanking sequences of *MOCS1* and *MOCS2* genes sequenced
- Homozygous for *MOCS2* c.413 G>A, p.G76R mutation in exon 4

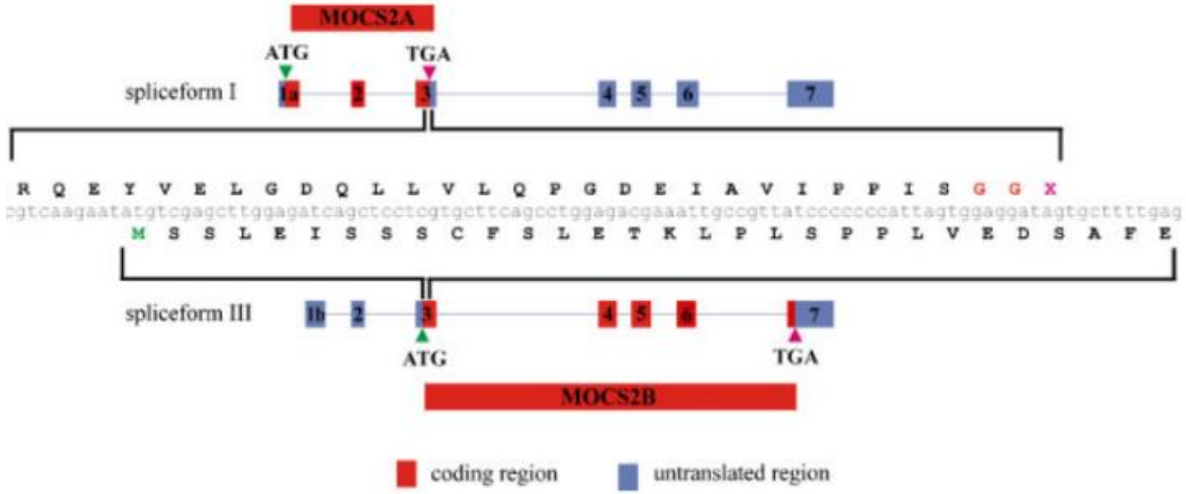


Diagram from: Molybdenum Cofactor Deficiency: Mutations in GPHN, MOCS1, and MOCS2, J Reiss and R Hahnewald (2011). *Human Mutation*, **32**, 10-18

Management and outcome

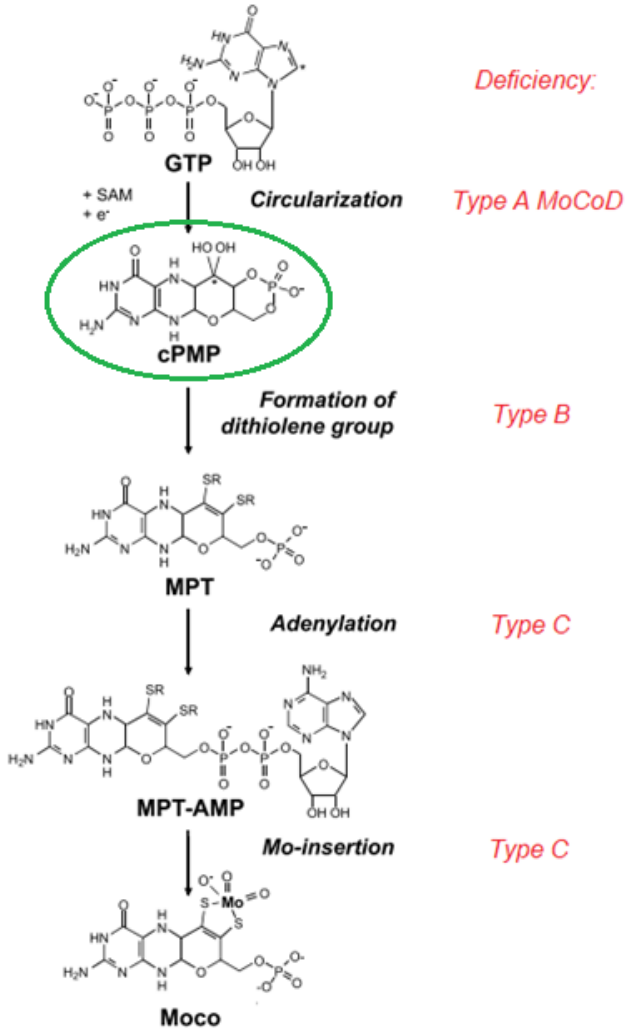
- Evelina PICU for 10 days
- Extubated after one week, no further ventilation issues
- Symptom management and DNAR plans put in place
- Medication review and NG tube training for parents before discharge
- Referral to palliative care team

- Has been at home for 18 months and doing well
- Seizures well controlled, but episodes of dystonia cause agitation and distress
- Poor sleep and hypotonia worsening
- PEG tube inserted to better manage gastro issues

Potential treatment

- Our case had MoCoD type B
- Potential treatment for MoCoD type A is to replace cPMP
- Approved by the FDA in the US. Ongoing trials in the UK
- Reduces risk of mortality
- Cannot reverse any neurological damage that has already occurred

Nulibry® (fosdenopterin) for injection

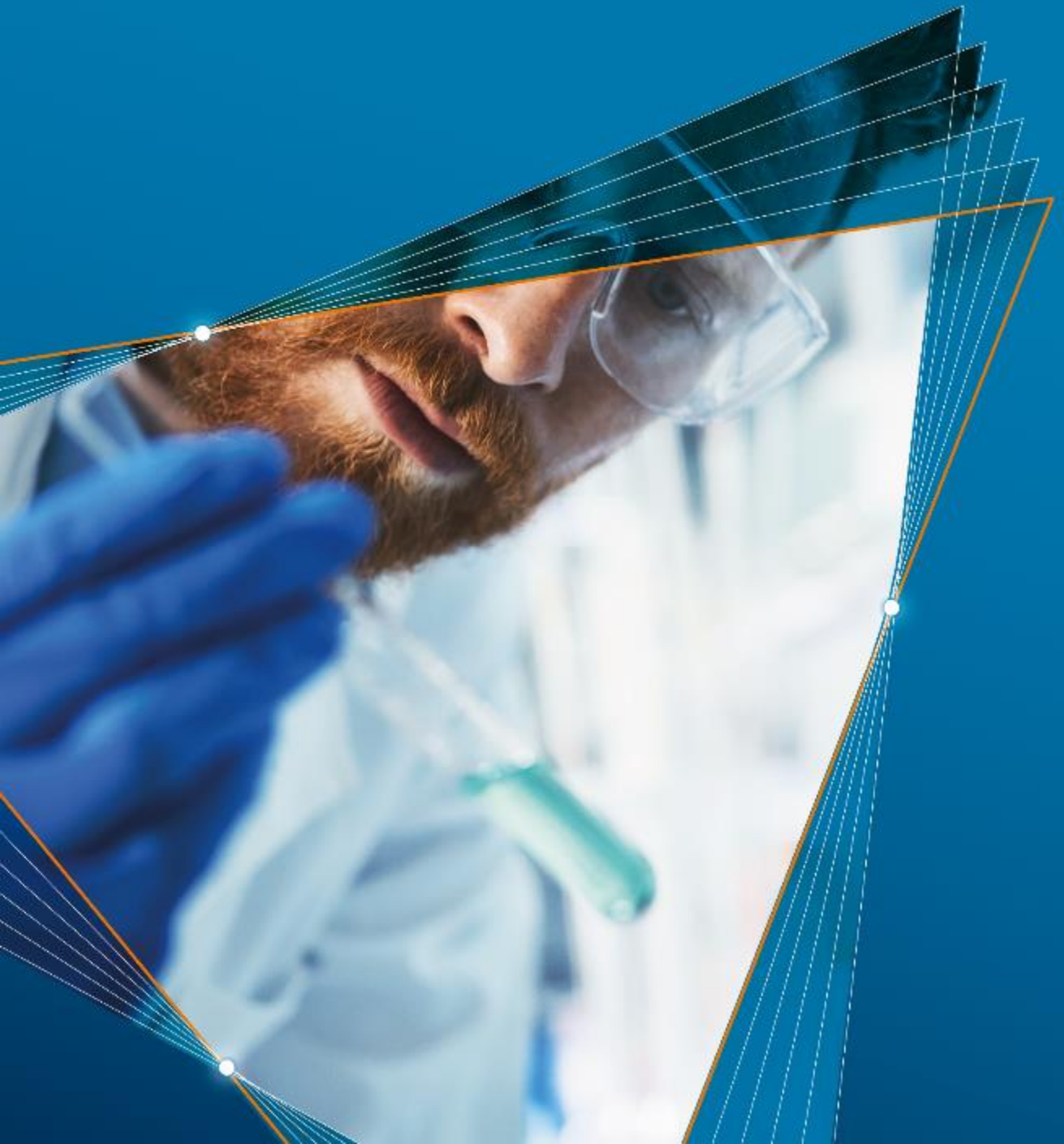


Efficacy and safety of cPMP substitution in severe molybdenum cofactor deficiency type A: a prospective cohort study, B Schwahn *et al* (2015). *Lancet*, **386**, 1955-63
Fosdenopterin: a first-in-class synthetic cyclic pyranopterin monophosphate for the treatment of molybdenum cofactor deficiency type A, B Schwahn (2021), *Paed Neur*, **17(2)**, 85-91

SYNLAB
NHS Guy's and St Thomas' NHS Foundation Trust
NHS King's College Hospital NHS Foundation Trust

Summary of key points

- The molybdenum cofactor (MoCo) is synthesised via action of six enzymes in four steps. The majority of cases of MoCo deficiency are due to *MOCS1* mutation (MoCoD type A), which can be treated with recombinant cPMP
- MoCo deficiency results in a neurological presentation including seizures and hypotonia
- A diagnosis of MoCo deficiency can be made, or at least suspected, from a combination of (in order of routine availability):
    - Plasma urate
    - Sulphite dipstick
    - Plasma/CSF/urine amino acids
    - Urine sulphocysteine
    - Urine/plasma xanthine and hypoxanthine (Purine screen)
    - Genetic confirmation
- Causes of ‘false’ increases and decreases in analytes associated with MoCo deficiency should be taken into account and results interpreted in light of the clinical picture



Thank you for listening
Any questions?