CBS Deficient Homocystinuria.

Kenneth N. Maclean PhD
The methionine cycle
Alternative metabolic fates for Hcy

• Extrusion into the extracellular space- plasma and urine
• Remethylation to methionine with either N-5-methylytetrahydrofolate or betaine as the methyl donor. These reactions are catalyzed by methylene tetrahydrofolate reductase/methionine synthase or betaine-homocysteine methyltransferase respectively.
• Conversion to cystathionine and ultimately cysteine catalyzed by Cystathionine beta-synthase (CBS) and cystathionine gamma-lyase (CGL) respectively.
Cystathionine beta-synthase

- Pyridoxal L-phosphate (PLP) dependent hemeprotein that catalyzes the condensation of homocysteine (Hcy) and serine to form cystathionine which is subsequently processed to cysteine by another PLP dependent enzyme cystathionine gamma lyase (CGL).
- CBS deficiency is inherited as an autosomal recessive trait, as shown by both pedigree studies and by enzyme assays in samples derived from parents of affected children.
Metabolic Sequelae of CBS deficiency

- Increased in plasma
  - Hcy (155-471 uM: normal is 8-13.9 uM) 
  - Methionine (353-1891uM: normal is 13-45 uM) 
  - AdoMet 888-2030 nM normal is 59-120 nM 
  - Adohcy 147-1700 nM: normal is 9-21nM)

- Decreased in plasma
  - Cysteine  40-140 uM: normal is 200-361uM 
  - Cystathionine 0-7 nM : normal is 50-342 nM

- Abnormal accumulations of Hcy and methionine have also been shown to occur in cerebrospinal fluid, aqueous humor and tissues
<table>
<thead>
<tr>
<th>Clinical Abnormalities in CBSDH.</th>
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The pyridoxine effect.

- Pyridoxine (vitamin B6) is the precursor of the coenzyme PLP which forms Schiff's bases of substrate amino acids during catalysis of transamination, decarboxylation and racemisation reactions.

- Approximately 50% of individuals with CBSDH are defined as “pyridoxine responsive” in that treatment (250-500mg daily) lowers plasma Hcy and methionine levels.
• The exact mechanism of this response is not yet clear but genetic studies indicate that pyridoxine responders have CBS mutations that are deficient in binding PLP and provision of extra PLP helps to stabilize residual levels of activity.

• Pyridoxine responding patients are not uniform. Some individuals respond by lowering Hcy and methionine to normal levels. Other patients retain slight elevations while some patients show little or no response and are thus referred to as “pyridoxine non-responsive”.

• Pyridoxine non-responsive patients are treated with betaine and a methionine restricted diet.

• Those patients that respond maximally to pyridoxine are still compromised in terms of transsulfuration and struggle to cope with methionine loads.
The methionine cycle
Mental retardation in CBSDH

• The most frequent abnormality of the CNS in CBSDH is MR. Often the first recognized sign of CBS deficiency and instrumental in initial diagnosis, presenting as developmental delay during the first and second year of life.
• IQ of untreated patients varies from 10 to 138 with the median of the cumulative frequency curve at approximately 64
• The distribution for pyridoxine responsive patients is shifted towards higher IQs with a median of 78
• Pyridoxine non-responsive individuals are the most severely affected with a median value of 56
Neurological Abnormalities In CBSDH

• Approximately 21% of individuals with CBSDH not treated from early infancy have had seizures most often of the grand mal type
• CBS deficiency is also associated with deficits in the extrapyramidal motor system. Gait disturbances are common in CBSDH
• Abnormal electroencephalograms have been reported in patients with and without seizures
• Dystronia (disordered tonicity of muscle) has been reported in a few patients.
• Although Hemiparesis (Paralysis affecting only one side of the body) or focal neurological signs suggest the presence of cerebrovascular occlusion in many patients a cerebrovascular origin for the neurological symptoms seems unlikely
The graph illustrates the cumulative frequency of patients, percent, for different categories: B6 - responsive, B6 - non-responsive, and all types combined. The x-axis represents the cumulative frequency of patients, while the y-axis shows the decile values.
Psychiatric abnormalities in CBSDH

• Mental illness has been reported frequently amongst individuals with CBSDH.
• In an investigation of 63 individuals with CBSDH clinically significant psychiatric disorders were found in 51%.
• Four diagnostic categories predominate: Episodic depression –10%, Chronic disorders of behaviour-17%, Chronic obsessive compulsive disorder-5%, Personality disorders-19%
• Aggressive behavior and other disorders of conduct were particularly common among patients with mental retardation and among pyridoxine-nonresponsive patients.
• Anecdotal reports of schizophrenia in CBSDH. Insufficient evidence to suggest it is anything other than rare.
Neural Pathology in CBSDH

- Within the CNS lesions in the brain are the most obvious sign of neurological damage.
- Infarcts, secondary to cerebrovascular occlusions are common.
- No evidence of demyelination
- Lipid composition of the brain ostensibly normal
- Apart from stroke, specific neurological findings have generally not been striking.
Putative Pathological Mechanisms

• Homocysteinylation of cysteine residues in cysteine-rich long-lived structural proteins
• Elevations in homocysteine thiolactone
• Perturbation of SAM/SAH ratio
• Accumulation of homocysteic and homocysteine sulfinic acids
• Inhibition of endothelial cell proliferation
• Induction of vascular smooth muscle cell proliferation
• Decrease in the synthesis and bioavailability of NO
• Induction of ER stress
• Oxidative stress through the endogenous production of H$_2$O$_2$
Evidence For a Downstream Contribution to Certain Aspects of CBSDH Pathology

- Total Hcy of 160µM/L
- Precocious thrombosis
- No connective tissue disorders or MR
- Suggestive that Hcy is toxic to the vasculature but that other aspects of CBSDH pathology require a concomitant block in cysteine biosynthesis.
The methionine cycle

[Diagram showing the methionine cycle with various reactions and compounds labeled, including dUMP, dTMP, Folic Acid, Protein, ATP, PPI + Pi, CO₂, dSAM, Methionine, SAM, CH₂THF, CH₃THF, THF, NADPH, NADP⁺, FAD, PLP, MeCbl, AdoMet, DMPG, Betaine, Choline, Ado, SAH, Homocysteine, thiolactone, Cystathionine, Transsulfuration Pathway, Glutathione, Protein, Cysteine, SO₄²⁻, Thymidylate Cycle, Metabolism, and other biochemical interactions.]
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Evidence that Hcy is not the sole pathological determinant in CBSDH

- Homocystinuria due to MTHFR or MS deficiency lack connective tissue defects and possibly MR
- Occult homocystinuria associated with C-terminal regulatory mutations of CBS. These patients have residual activity, have very high Hcy but retain the capacity for low-level activity-increased risk of precocious thrombosis but no MR or connective tissue disturbances.
- Discrepancy between the incidence of the pyridoxine-responsive CBS I278T mutant allele and the number of patients presenting as classical homocytinurics. Some of these patients have been observed after a thrombotic event but lack the usual developmental delay and connective tissue disorders.