Urea Cycle Defects

Dr Mick Henderson

Biochemical Genetics
Leeds Teaching Hospitals Trust
The Urea Cycle

The urea cycle enables toxic ammonia molecules to be converted to the readily excreted and non-toxic urea. The urea cycle has other metabolic benefits. It is an important source of arginine, used in a variety of metabolic reactions. The enzymes of the urea cycle are predominantly located in the liver and to a lesser extent in the renal cortex.

Ammonia is generated from a variety of sources in the body. It is a waste product of the deamination of amino acids. It is also produced in large quantities by gut bacteria. It is absorbed across the intestinal wall and found in high concentrations in hepatic portal blood. It is produced by the metabolism of muscles and venous concentrations are higher than arterial.

Defects of enzymes involved with the urea cycle lead to hyperammonaemia and arginine deficiency, except in the case of arginase deficiency. Ammonia is neurotoxic and damages the central nervous system causing a variety of symptoms from drowsiness to death.

Treatments are available for most of the disorders, so early diagnosis and institution of therapy is vital.

There is considerable phenotypic variation. Clinical presentations vary from severe neonatal onset to more mild adult forms following catabolic episodes.

Diagnosis of urea cycle defects is usually based initially on patterns of metabolites in plasma and urine. Enzyme confirmation is not simple and requires a liver biopsy for carbamoyl phosphate synthetase, ornithine transcarbamylase or N-acetylglutamate synthetase deficiencies. The other disorders can be diagnosed on skin fibroblasts or in the case of arginase deficiency in red blood cells. Genetic mutations can be helpful in confirming a diagnosis and offering a method for prenatal diagnosis.
Blood ammonia
What is it?

Ammonia is a weak base in equilibrium with the ammonium ion
At physiological pH 95% is NH$_4^+$
Clinical chemistry methods measure total NH$_3$ + NH$_4^+$
Normally in venous blood it is < 40 µmol/L
  neonates < 100 µmol/L
  premature neonates < 200 µmol/L
Blood ammonia
Where does it come from?

The **deamination** of amino acids

From **gut bacteria** *hepatic portal venous NH₃ up to 20x higher than systemic*

From muscle metabolism, particularly the deamination of AMP

*Venous NH₄⁺→arterial*

Renal metabolism, renal tubular generation from glutamine

*Renal venous NH₄⁺→renal arterial*
Symptoms of ammonia toxicity

- Poor feeding
- Lethargy
- Irritability
- Cognitive impairment
- Vomiting
- Hyperventilation, *respiratory alkalosis*
- Mental retardation
- Ataxia
- Convulsions
- Coma
- Death
Causes of elevated ammonia

Factitious

A struggling infant, or a difficult venepuncture

Delayed analysis

Smoking

Liver disease e.g. Reye’s syndrome

Mitochondrial poisoning, chemotherapy, Valproate

Mitochondrial disease (Respiratory chain disorders)

Organic acidaemias

Inherited defects of the urea cycle

More rarely:

triple ‘H’ syndrome (HHH)

Lysinuric protein intolerance

Hyperinsulinism due to glutamate dehydrogenase deficiency
The Urea cycle

- HCO₃
- NH₃
- carbamoyl phosphate
- orotic acid
- pyrimidines
- ornithine
- citrulline
- arginine
- argininosuccinic acid
- aspartate
- fumarate
- urea
The urea cycle is split between two compartments

Part of the urea cycle takes place within mitochondria.
Carbamoyl phosphate is generated from ammonia and bicarbonate within mitochondria.
The enzyme OTC (ornithine transcarbamylase) is synthesised in the cytoplasm and then imported into mitochondria.
Ornithine is generated in the cytoplasm but enters mitochondria to form citrulline.
Citrulline has to be exported to the cytoplasm for conversion to argininosuccinate.
These transport steps facilitate control but also are potential sites for genetic disease.
Carbamoyl phosphate synthetase (CPS) and N-acetylglutamate synthetase (NAGS) deficiencies

- Glutamic acid and acetyl CoA 
- Carbamoyl phosphate synthetase
- N-acetyl glutamate 
- Carbamyl phosphate synthetase
- Ornithine
- Citrulline
- Arginine
- Argininosuccinic acid
- Fumarate
- Urea
- Aspartate
- Orotic acid
- Pyrimidines
- NH$_3$
- HCO$_3$
CPS and NAGS deficiencies

They have a similar clinical presentation usually with severe hyperammonaemia. NAGS can suffer competitive inhibition by metabolites that accumulate in some organic acidaemias thus leading to secondary hyperammonaemia.
Ornithine Transcarbamylase (OTC) deficiency

\[
\begin{align*}
\text{HCO}_3^- & \quad \text{NH}_3 \\
\downarrow & \quad \downarrow \\
\text{carbamoyl phosphate} & \\
\downarrow & \\
\text{ornithine transcarbamylase} & \\
\text{ornithine} & \text{citrulline} \\
\downarrow & \downarrow \\
\text{arginine} & \text{argininosuccinic acid} \\
\downarrow & \downarrow \\
\text{fumarate} & \text{urea} \\
\text{aspartate} & \text{orotic acid} \\
\text{pyrimidines} & \text{ornithine}
\end{align*}
\]
OTC deficiency

It is the most common urea cycle defect
It is X linked, thus there is a variable phenotype in female heterozygotes depending on pattern of random X chromosome inactivation. Males are usually more severely affected.
OTC is characterised by orotic aciduria and hyperammonaemia
The amino acid abnormalities are mainly non-specific, i.e. increased glutamine and alanine and decreased ornithine, arginine and citrulline
Citrullinaemia

HCO$_3^-$ → NH$_3$

→ carbamoyl phosphate

→ ornithine → citrulline → argininosuccinic acid synthetase

→ arginine → argininosuccinic acid

→ aspartate → fumarate → urea

→ orotic acid → pyrimidines
Citrullinaemia

It is characterised by elevated citrulline in plasma and urine, and orotic acid in urine and hyperammonaemia. Citrulline has relatively poor renal clearance, so proportionately greater elevations are observed in plasma.
Argininosuccinic aciduria

- HCO₃⁻
- NH₃
- carbamoyl phosphate
- orotic acid
- pyrimidines
- ornithine
- citrulline
- arginine
- argininosuccinic acid
- urea
- aspartate
- argininosuccinic acid lyase
- fumarate
Argininosuccinic aciduria

It is characterised by elevations of argininosuccinate in plasma and urine. Argininosuccinate has high rate of renal clearance hence is much more readily detected in urine.

Renal excretion also provides the body with route to excrete nitrogen, so hyperammonaemia is often mild and may be absent.
Arginase deficiency

Enzyme defect
confirmed in red cells

- HCO₃
- NH₃
- carbamoyl phosphate
- ornithine
- citrulline
- argininosuccinic acid
- aspartate
- orotic acid
- arginine
- arginase
- urea
- fumarate
Arginase deficiency

It is characterised by elevations of arginine in plasma and urine and orotic aciduria. Hyperammonaemia is variable and may only be mild or intermittent. However the clinical picture is usually severe. Patients may present with neonatal seizures and frequently suffer progressive neurological symptoms as they grow including spastic diplegia.
Principles of treatment of urea cycle defects

- Alternative pathway stimulation; oral drugs that cause an increase in the excretion of glycine thereby depleting ammonia by stimulating the replacement synthesis of glycine
  
  Most commonly:
  - Benzoate

Can also involve using:
  - Phenylbutyrate
  - Phenylacetate

(See Treatment and Monitoring Module)

- Haemodialysis, in cases of acute, extreme hyperammonaemia
- Stimulation of CPS by a synthetic co-factor
- A low protein diet is a very common strategy to control the chronic hyperammonaemia
- Arginine supplementation, in relevant disorders
Self assessment questions

1. What are the early symptoms of hyperammonaemia?
2. What is a 'normal' ammonia for a neonate?
3. In which organ is the urea cycle principally located?
4. What part of the urea cycle takes place within mitochondria?
5. Which metabolite is most characteristic of OTC deficiency?
6. Which disorder is x-linked?
7. Which disorder is can cause spastic diplegia?
8. Which metabolite is most characteristic of argininosuccinic acid lyase deficiency?
9. Which drug is commonly used to treat hyperammonaemia?
10. Which treatment can be used for acute severe hyperammonaemia?
Self assessment answers

1. Irritability, lethargy, poor feeding, cognitive impairment, respiratory alkalosis, vomiting.
2. Less than 100 umol/L
3. The liver
4. The formation of carbamoyl phosphate and the formation of citrulline
5. Orotic acid
6. OTC deficiency
7. Arginase deficiency
8. Argininosuccinic acid
9. Benzoate
10. Haemodialysis