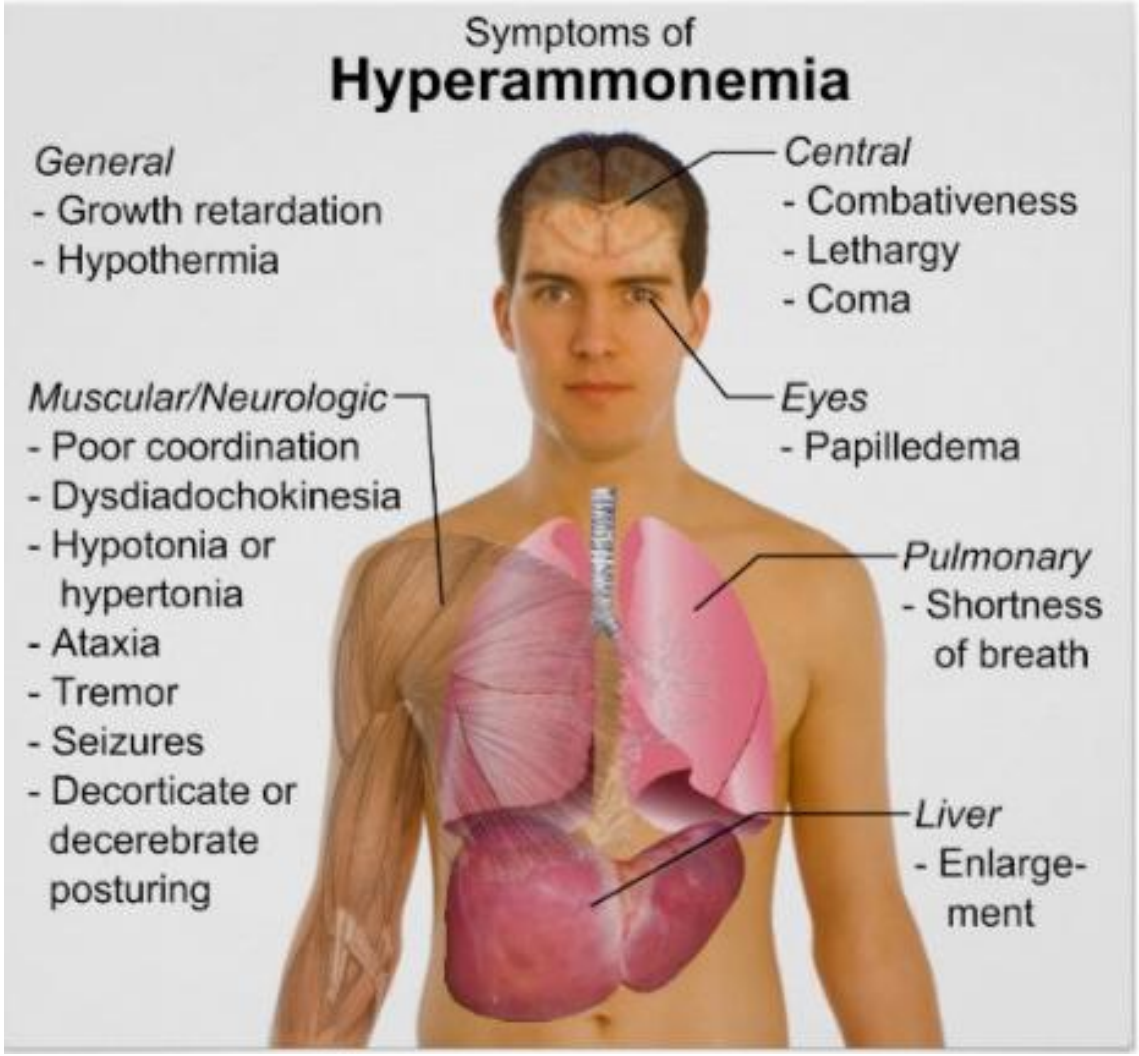
Determination of Ammonia in Plasma



Amal Alamri

Sources of ammonia

However, substantial amounts of ammonia can be obtained from other sources.

- **From glutamine:** The kidneys generate ammonia from glutamine by the actions of renal *glutaminase* and *glutamate dehydrogenase*.
- **From bacterial action in the intestine:** Ammonia is formed from urea by the action of bacterial *urease* in the lumen of the intestine.
- **From amines:** Amines obtained from the diet, and monoamines that serve as hormones or neurotransmitters, give rise to ammonia by the action of *amine oxidase*
- **From purines and pyrimidines:** In the catabolism of purines and pyrimidines, amino groups attached to the rings are released as ammonia

FATE OF AMMONIA

Although ammonia is constantly produced in the tissues, it is present at very low levels in blood. This is due both to the ***rapid removal of blood ammonia by the liver***, and the fact that many tissues, particularly ***muscle, release amino acid nitrogen in the form of glutamine or alanine, rather than as free ammonia***

1. **Formation of Urea:** in the liver is quantitatively the most important *disposal route for ammonia*.
2. **Formation of Glutamine :** this is an ATP-requiring formation of glutamine from glutamate and ammonia by ***glutamine synthetase*** occurs primarily in the ***muscle*** and ***liver***, but is also important in the CNS where it is the major mechanism for the removal of ammonia in the brain.

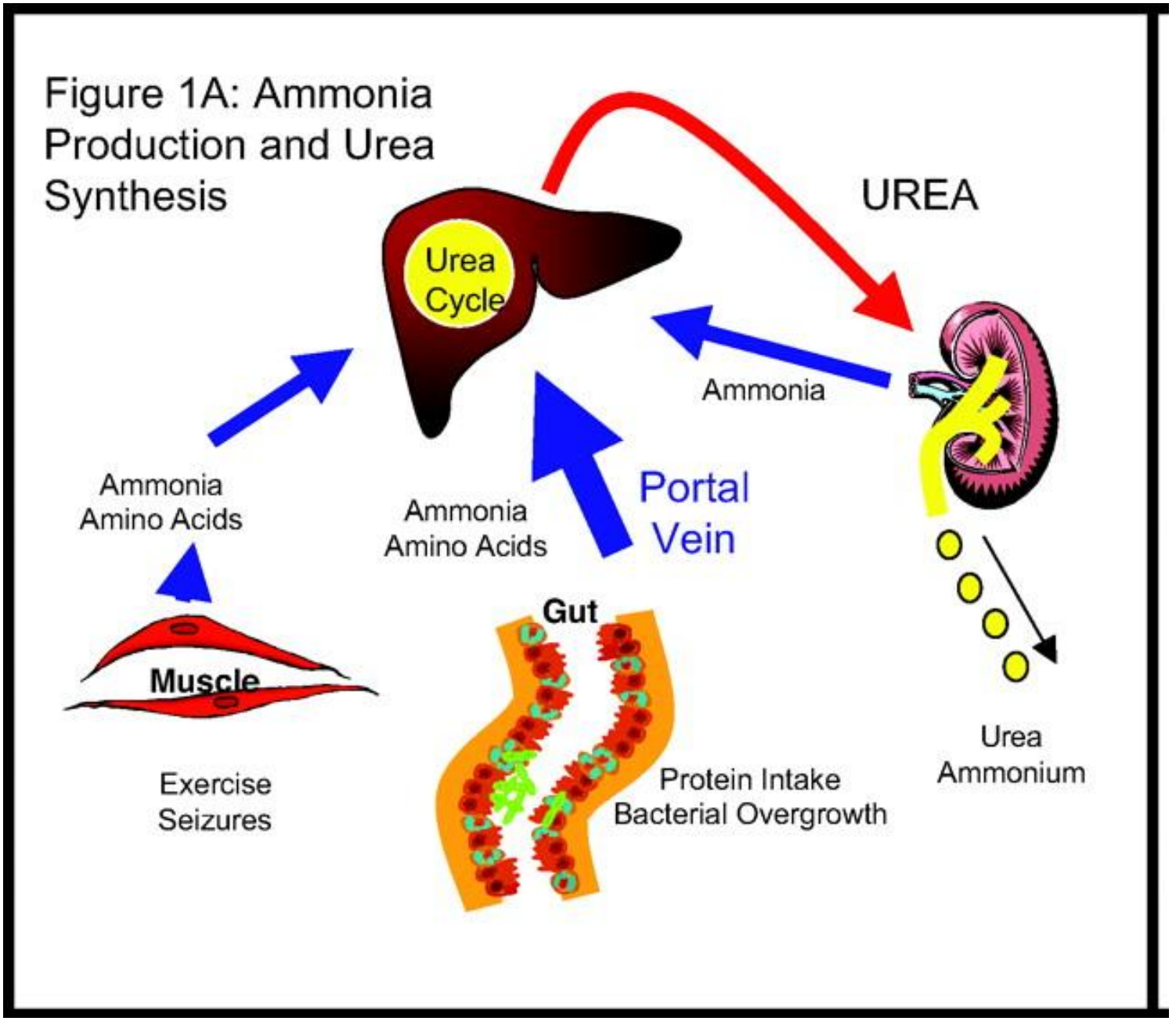


Figure 1A: Ammonia Production and Urea Synthesis
UREA
Urea Cycle
Ammonia
Ammonia Amino Acids
Ammonia Amino Acids
Portal Vein
Gut
Muscle
Exercise Seizures
Protein Intake Bacterial Overgrowth
Urea Ammonium

Hyperammonemia

hyperammonemia is a medical emergency, because ammonia has a direct neurotoxic effect on the CNS.(central nervous system).

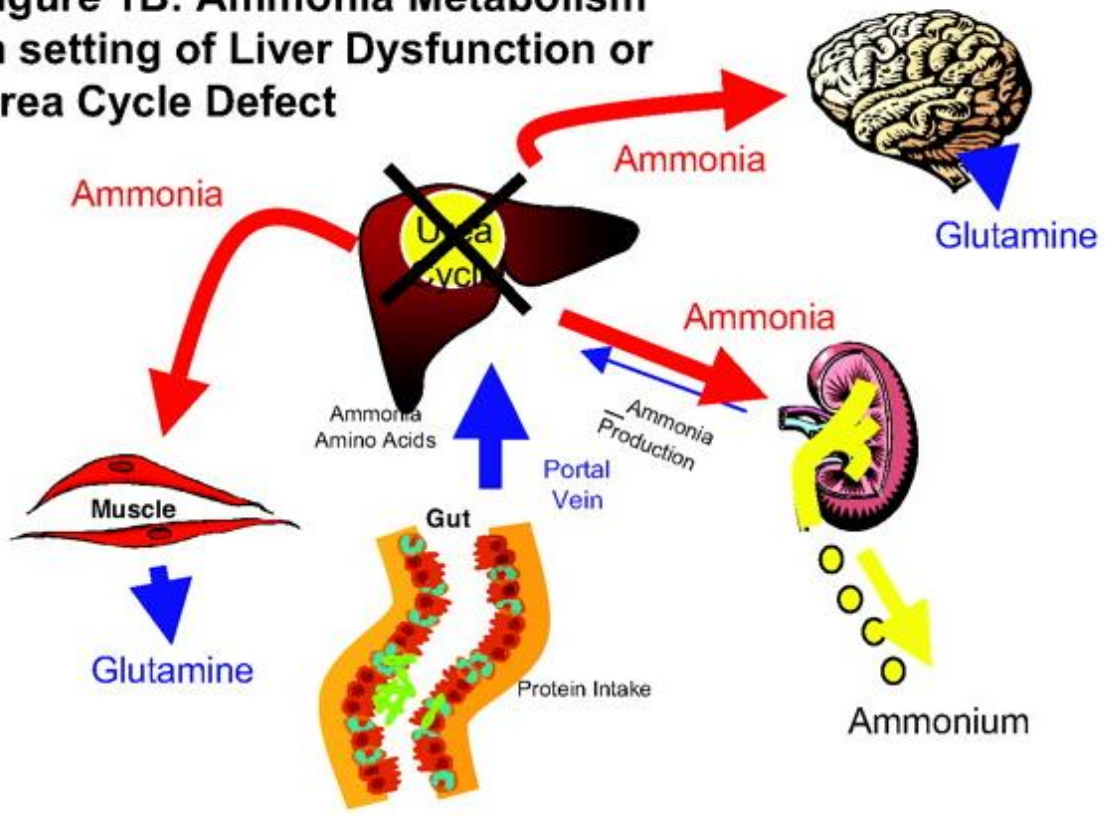
Symptoms of ammonia intoxication, which include tremors, slurring of speech, somnolence, vomiting, cerebral edema, and blurring of vision. ammonia can cause coma and death.

The two major types of hyperammonemia are:

1-Congenital hyperammonemia:

2-Acquired Hyperammonemia:

Figure 1B: Ammonia Metabolism in setting of Liver Dysfunction or Urea Cycle Defect



HOW TO COLLECT THE SAMPLES?

1-Blood is collected from patients fasted at least 6 hours.

2- Donor should not clench fist during collection as muscular exertion often increases venous Ammonia levels. Since erythrocytes contain larger amounts of ammonia than plasma, ***hemolysis may increase results***.

3-Heparin is the preferred anticoagulant because it reduces red cell ammonia production. Other anticoagulants, such as sodium citrate, ***Potassium Oxalate or sodium fluoride*** are reportedly produce high results.

HOW TO COLLECT THE SAMPLES?

4- After drawing blood is placed in an ice bath and Plasma is separated within 30 minutes.

5- venous blood is preferred to capillary blood since the later may yield higher ammonia level due to ammonia release through platelet activation.

6- heparinized blood specimens should be centrifuged immediately and at speeds sufficient to yield platelet poor plasma.

7- Storage of specimens at -70 °C is recommended, if assays cannot be performed promptly

WHY THE TEST PERFORMED?

- **newborn** when symptoms such as irritability, vomiting, lethargy, and arise in the first few days after birth.
- It may be performed when a **child** develops these symptoms about a week following a viral illness, such as influenza
- When **adults** experience mental changes, disorientation, sleepiness, or lapse into a coma, an ammonia level may be ordered to help evaluate the cause of the change in consciousness.
- As indicators of liver cells damage (e.g. Reye's Syndrome)

EXPECTED VALUES

Plasma Ammonia Range = 0.17 - 0.80 mg/ml

10-47 mmol/L

OBJECTIVES

For the quantitative determination of ammonia in plasma by enzymatic UV / end point method.

METHOD PRINCIPLE

NH_3 + 2-Oxoglutarate + NADH+ H+ $\xrightarrow{\text{GLDH}}$ L-Glutamate + NAD+

The Ammonia reacts with 2-Oxoglutarate, in the presence of L-GLDH and the co-enzyme NADH, to produce L-glutamate. The resulting decrease in absorbance of NADH at 340 nm is proportional to the level of ammonia in the sample.

Methylated amines which interfere with other conventional procedure, do not react in the described method

GLDH: *glutamate dehydrogenase*
NADH+ H+: *nicotinamide adenine dinucleotide*

QUESTIONS:

What are the defects that causing congenital Hyperammonemia?

What are the defects that causing acquired Hyperammonemia?

What causes reye's syndrome?

References

- https://www.inkling.com/read/illustrated-reviews-biochemistry-harvey-5th/chapter-19/metabolism-of-ammonia
- http://labtestsonline.org/understanding/analytes/ammonia/tab/test/

- UDI ammonia reagent kit.