Protein:

Protein is a macromolecule that presented in food, and with digestion it broken to amino acids.

In the body or living organisms: Protein have different functions depending on the class of the protein: (**Proteins display an incredible diversity of functions)**

1. enzymes and polypeptide hormones direct and regulate metabolism in the body
2. contractile proteins in muscle permit movement
3. In bone, the protein collagen forms a framework for the deposition of calcium phosphate crystals acting like the steel cables in reinforced concrete.
4. In the bloodstream, proteins hemoglobin and plasma albumin, shuttle molecules essential to life.
5. immuno globulins fight infectious bacteria and viruses.

Common structure of protein, they are a polymer of amino acid(amino acids bounded with peptide bond.fig 1,B)(20 amino acids) for animal and human amino acid fiog1, A . Not all plants protein have the complete 20 amino acid.

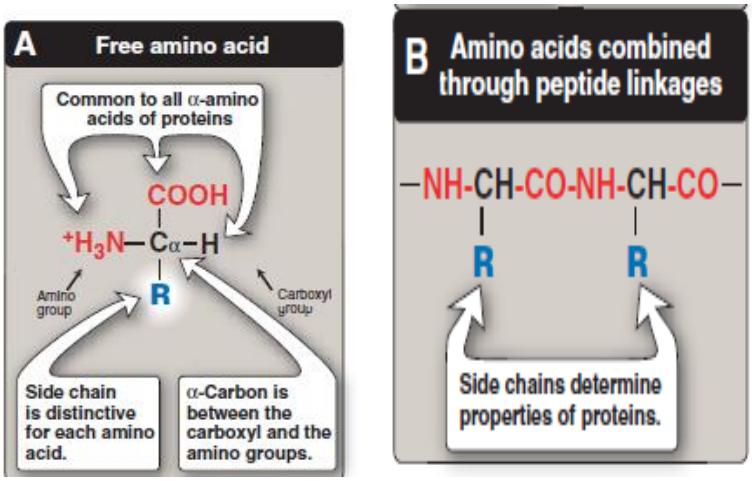

Figure 1
Structural features of amino acids (shown in their fully protonated form)

How Much Protein Do We Need?

The amount of protein that we need is dependent in part on our age, weight and levels of activity. Children and adolescents who are still growing and developing need proportionately more protein in their diets than adults. People with high levels of activity

may need slightly more protein than those who lead more sedentary lifestyles – as protein is essential in building and repairing muscle and other tissues slightly more is needed for those actively trying to develop muscle.

To calculate roughly how much protein you need to consume daily: multiply your weight in kilograms by 0.8. The answer is the number of grams of protein you should consume every day.

Therefore, if you weigh 100kg you should be consuming around 80grams of protein a day.

Food Rich in Protein:

Meat, Fish, Egg, Dairy Products, Beans, Nuts and Seeds, Other Protein Sources (Whole grain, asparagus, broccoli, Brussels sprouts, cauliflower),

Supplement :

(commonly powdered milk (whey) and soya based proteins), pill form, either individually or combining two or more of the essential amino acids (be prescribed to patients who cannot, thorough various health complaints, synthesis the amino acids they need from protein) .

Amino Acids: Disposal of Nitrogen

Amino acids are not like carbohydrate of lipids, they can't be stored in the body . there are no protein which is sole function to maintain a supply of amino acid for future use .

Amino acids are obtained from:

1. diet
2. synthesized de novo
3. produced from normal protein degradation

Excesss of amino acid for the biosynthesis needs of the cell will be rapidly degraded:

1. **The first phase of catabolism involves** :
   a. the removal of the α-amino groups (transamination)
   b. subsequent oxidative deamination
   this will lead to forming ammonia(part of it will be excreted in urine and most of it will be used in urea biosynthesis) and the corresponding, α-keto acid—the "carbon skeletons" of amino acids (Fig 2)

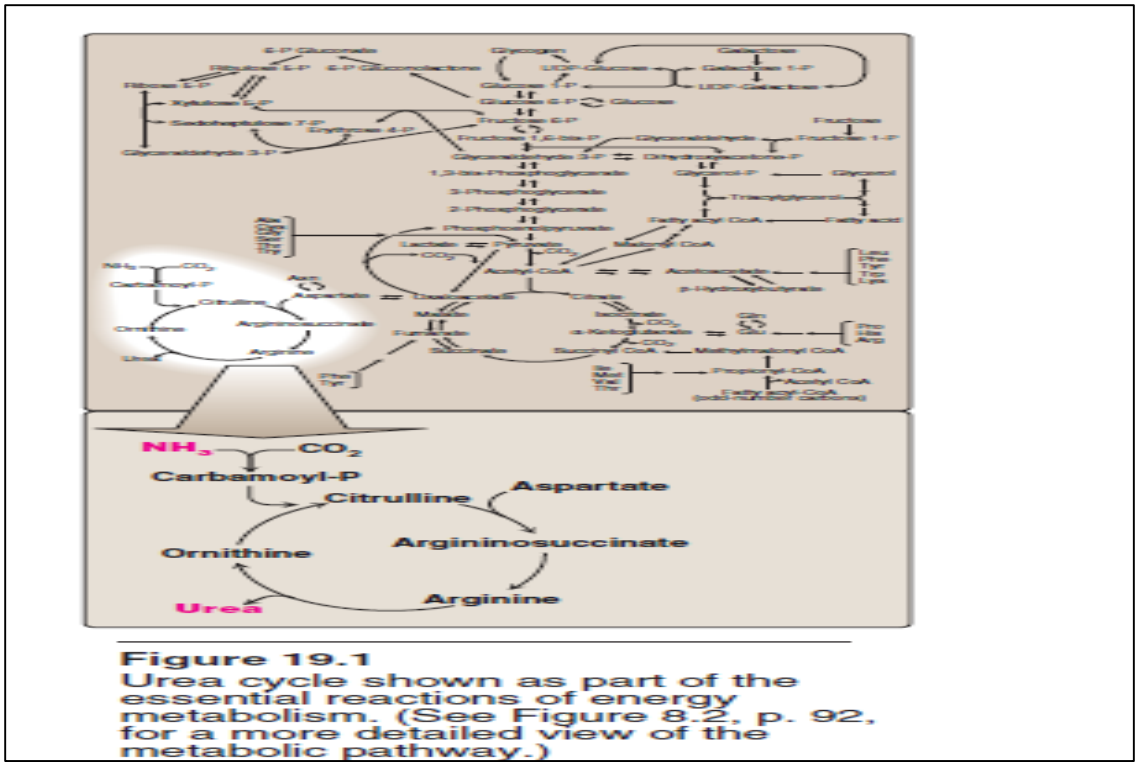

Figure 19.1
Urea cycle shown as part of the essential reactions of energy metabolism. (See Figure 8.2, p. 92, for a more detailed view of the metabolic pathway.)

Fig 2 : urea cycle

2. . In the second phase of amino acid catabolism, the carbon skeletons of the, α-ketoacids are converted to common metabolic pathways. These compounds can be metabolized to CO2 and water, glucose, fatty acids, or ketone bodies by the central pathways of metabolism.

OVERALL NITROGEN METABOLISM;

The role of body proteins in these transformations involves two important concepts: the amino acid pool and protein turnover

Amino acid pool

Free amino acids are present throughout the body, for example, in cells, blood, and the extracellular fluids. Amino acids are exist in one single entity, called the amino acid pool.

This pool is supplied by three sources:
1) amino acids provided by the degradation of body proteins,
2) amino acids derived from dietary protein,
3) synthesis of nonessential amino acids from simple intermediates of Metabolism

Conversely, the amino pool is depleted by three routes:

1) synthesis of body protein

2) amino acids consumed as precursors of essential nitrogen-containing small molecules

3) conversion of amino acids to glucose, glycogen,
fatty acids, ketone bodies, or $CO_2 + H_2O$.

Although the amino acid pool is small (comprised of about 90–100 g of amino acids) in comparison with the amount of protein in the body (about 12 kg in a 70-kg man), it is conceptually at the center of whole-body nitrogen metabolism.

Protein turnover:

Most proteins in the body are constantly being synthesized and then degraded, permitting the removal of abnormal or unneeded proteins. For many proteins, regulation of synthesis determines the concentration of protein in the cell, with protein degradation assuming a minor role. For other proteins, the rate of synthesis is constitutive, that is, relatively constant, and cellular levels of the protein are controlled by selective degradation.

In healthy adults, the total amount of protein in the body remains constant, because the rate of protein synthesis is just sufficient to replace the protein that is degraded. This process, called protein turnover, leads to the hydrolysis and resynthesis of 300–400 g of body protein each day. The rate of protein turnover varies widely for individual proteins. Short-lived proteins (for example, many regulatory proteins and misfolded proteins) are rapidly degraded, having half-lives measured in minutes or hours. Long-lived proteins, with half-lives of days to weeks, constitute the majority of proteins in the cell. Structural proteins, such as collagen, are metabolically stable, and have half-lives measured.

DIGESTION OF DIETARY PROTEINS:

Most of the nitrogen in the diet is consumed in the form of protein, Proteins are generally too large to be absorbed by the intestine. [Note: An example of an exception to this rule is that newborns can take up maternal antibodies in breast milk]. They must, therefore, be hydrolyzed to yield di- and tripeptides as well as individual amino acids, which can be absorbed. Proteolytic enzymes responsible for degrading proteins are produced by three different organs: the stomach, the pancreas, and the small intestine. Fig 3.

1. **Digestion of proteins by gastric secretion**
   The digestion of proteins begins in the stomach, which secretes

gastric juice—a unique solution containing **hydrochloric acid** (too dilute (pH 2–3) , functions instead to kill some bacteria and to denature proteins. **Proenzyme, pepsinogen** (This acid-stable endopeptidase). Pepsinogen is activated to pepsin, either by HCl, or autocatalytically by other pepsin molecules that have already been activated. Pepsin releases peptides and a few free amino acids from dietary proteins

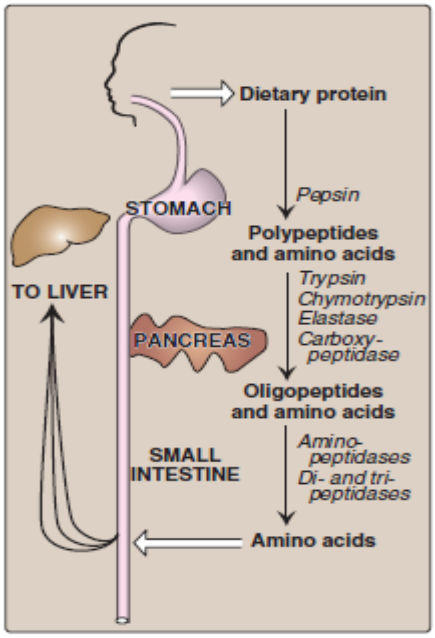

Figure 3
Digestion of dietary proteins by the proteolytic enzymes of the gastro-intestinal tract.

2. **Digestion of proteins by pancreatic enzymes**
   On entering the small intestine, large polypeptides produced in the and amino acids by a group of pancreatic proteases:
   1. **Specificity**: Each of these enzymes has a different specificity for the amino acid R-groups adjacent to the susceptible peptide bond (fig 4)
   These enzymes, like pepsin described above, are synthesized and secreted as inactive zymogens.
   (Abnormalities in protein digestion: In individuals with a deficiency in pancreatic secretion (for example, due to chronic pancreatitis, cystic fibrosis, or surgical removal of the pancreas), the digestion and absorption of fat and protein are incomplete. This results in the abnormal appearance of lipids (called steatorrhea steatorrhea,) and undigested protein in the feces

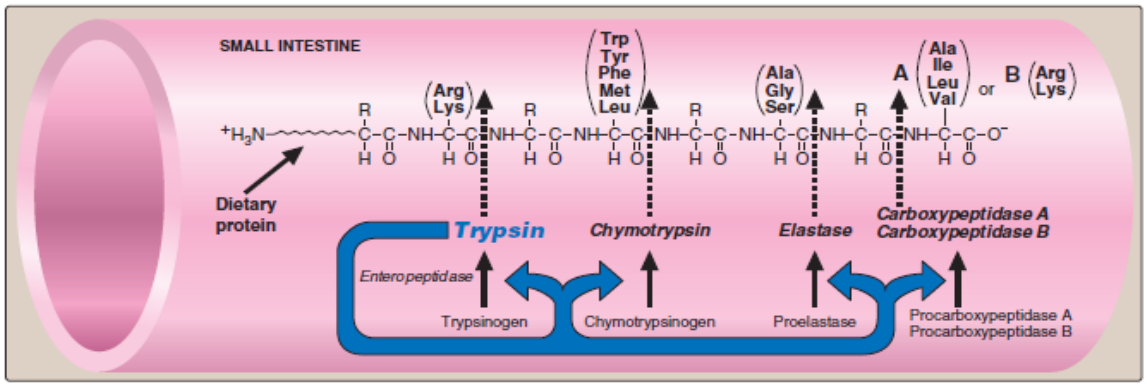

Figure 4
Cleavage of dietary protein by *proteases* from the pancreas. The peptide bonds susceptible to hydrolysis are shown for each of the five major pancreatic *proteases*. [Note:The first three are serine *endopeptidases*, whereas the last two are *exopeptidases*.]

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> Celiac disease (celiac sprue) is a disease of mal-absorption resulting from immune-mediated damage to the small intestine in response to ingestion of gluten (or gliadin produced from gluten), a protein found in wheat, barley and rye.

3. **Digestion of oligopeptides by enzymes of the small intestine**
   The luminal surface of the intestine contains aminopeptidase—an exopeptidase that repeatedly cleaves the N-terminal residue from oligopeptides to produce even smaller peptides and free amino acids.
4. **Absorption of amino acids and small peptides**
   Free amino acids are taken into the enterocytes by a Na+-linked secondary transport system of the apical membrane. Di- and tri -peptides, however, are taken up by a H+-linked transport system. The peptides are hydrolyzed in the cytosol to amino acids that are released into the portal system by facilitated diffusion. Thus, only free amino acids are found in the portal vein after a meal containing protein. These amino acids are either metabolized by the liver or released into the general circulation. [Note: Branched-chain amino acids are important examples of amino acids that are not metabolized by the liver, but instead are sent from the liver primarily to muscle via the blood.]

REMOVAL OF NITROGEN FROM AMINO ACIDS:

The presence of the α-amino group keeps amino acids safely locked away from oxidative breakdown. Removing the α-amino group is essential for producing energy from any amino acid, and is an obligatory step in the catabolism of all amino acids. Once removed, this nitrogen can be

incorporated into other compounds or excreted, with the carbon skeletons being metabolized.

A. Transamination: the funneling of amino groups to glutamate:
The first step in the catabolism of most amino acids is the transfer of their α-amino group to α-ketoglutarate (Fig5).

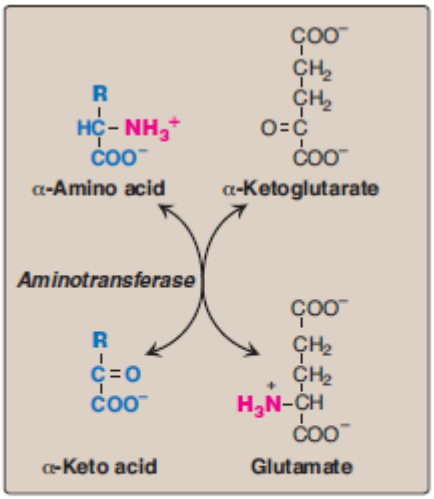
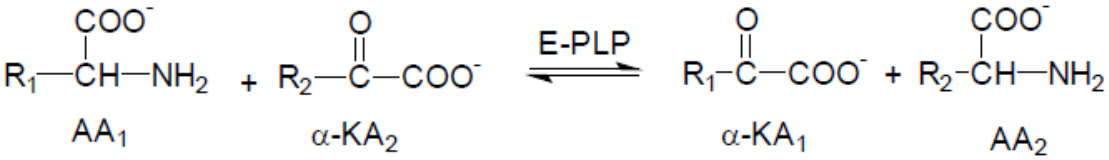


Figure [redacted] 5
Aminotransferase reaction using α-ketoglutarate as the amino-group acceptor.

This transfer of amino groups from one carbon skeleton to another is catalyzed by a family of enzymes called aminotransferases (formerly called transaminases). These enzymes are found in the cytosol and mitochondria of cells throughout the body—especially those of the liver, kidney, intestine, and muscle. All amino acids, with the exception of lysine and threonine, participate in transamination at some point in their catabolism. [Note: These two amino acids lose their α-amino groups by deamination].



The two most important aminotransferase reactions are catalyzed by alanine aminotransferase(ALT) and aspartate aminotransferase(AST).

a. Alanine aminotransferase (ALT): ALT is present in many tissues. The enzyme catalyzes the transfer of the amino group of alanine to α-ketoglutarate, resulting in the formation of pyruvate and glutamate. The reaction is readily reversible. However, during amino acid catabolism, this enzyme (like most

aminotransferases) functions in the direction of glutamate synthesis. Thus, glutamate, in effect, acts as a "collector" of nitrogen from alanine. (fig 6, 7)

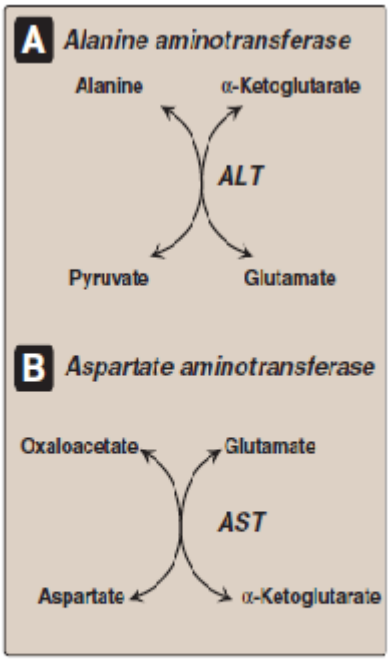


Figure 6
Reactions catalyzed during amino acid catabolism. A. *Alanine aminotransferase (ALT).* B. *Aspartate aminotransferase (AST).*

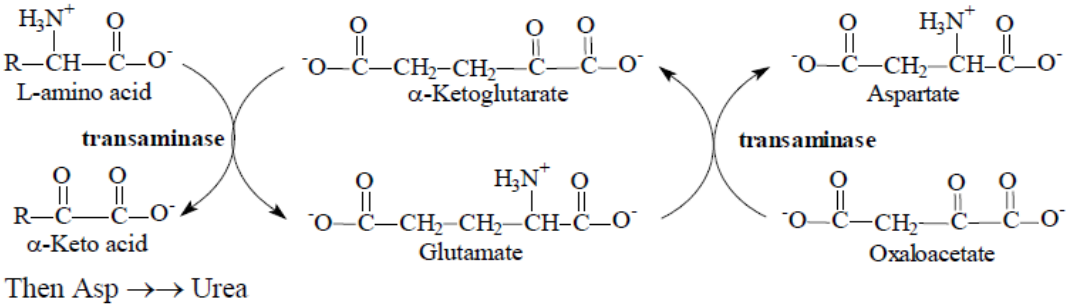


Fig 7 : Transaminase Reactions.

Aspartate aminotransferase (AST): AST is an exception to the rule that aminotransferases funnel amino groups to form glutamate. During amino acid catabolism, AST transfers amino groups from glutamate to oxaloacetate, forming aspartate. which is used as a source of nitrogen in the urea cycle. [Note: The AST reaction is also reversible.] fig 6,7

Mechanism of action of aminotransferases:

All aminotransferases require the coenzyme **p**yridoxa**l p**hosphate (a derivative of vitamin B6 which is covalently linked to the ε-amino group of a specific lysine residue at the active site of the enzyme. Aminotransferases act by transferring the amino group of an amino acid to the pyridoxal part of the coenzyme to generate pyridoxamine phosphate. The pyridoxamine form of the coenzyme then reacts with an α-keto acid to form an amino acid, at the same time regenerating the original aldehyde form of the coenzyme. Figure 8. shows these two component reactions for the reaction catalyzed by AST.

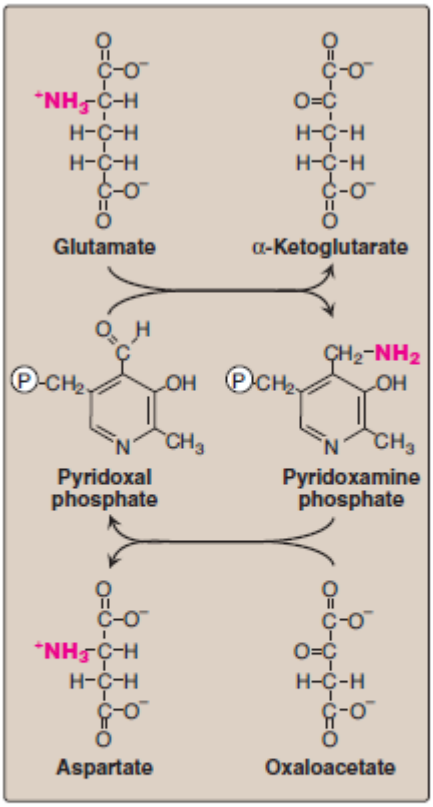


Figure 8
Cyclic interconversion of pyridoxal phosphate and pyridoxamine phosphate during the *aspartate aminotransferase* reaction.
[Note: Ⓟ = phosphate group.]

B. Glutamate dehydrogenase: the oxidative deamination of amino acids :

In contrast to transamination reactions that transfer amino groups, oxidative deamination by glutamate dehydrogenase results in the liberation of the amino group as free ammonia (NH3) (Figure 9). These reactions occur primarily in the liver and kidney. They provide α-keto acids that can enter the central pathway of energy metabolism, and ammonia, which is a source of nitrogen in urea synthesis.

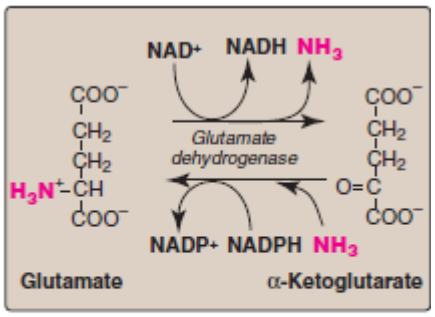


Figure [redacted] 9
Oxidative deamination by *glutamate dehydrogenase*.

Direction of reactions: The direction of the reaction depends on the relative concentrations of glutamate, α-keto glutarate, and ammonia, and the ratio of oxidized to reduced co - enzymes. For example, after ingestion of a meal containing protein, glutamate levels in the liver are elevated, and the reaction proceeds in the direction of amino acid degradation and the formation of ammonia (see Figure 10A). [Note: the reaction can also be used to synthesize amino acids from the corresponding α-keto acids (see Figure 10 B).]

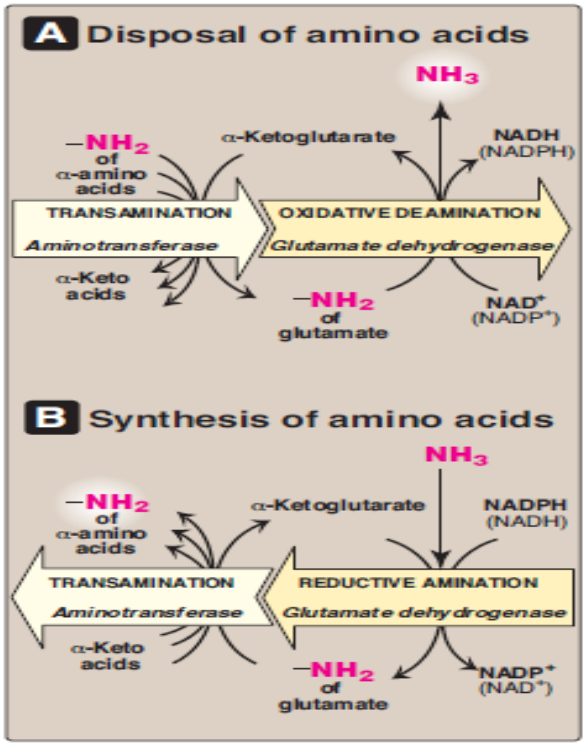


Figure [redacted] 10
Combined actions of *aminotransferase* and *glutamate dehydrogenase* reactions.

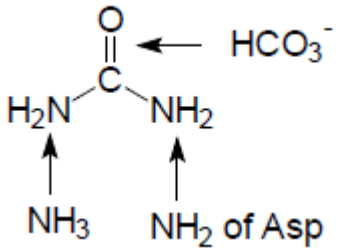
C. Transport of ammonia to the liver

Two mechanisms are available in humans for the transport of ammonia from the peripheral tissues to the liver for its ultimate conversion to urea. The first, found in most tissues, uses glutamine synthetase to combine ammonia (NH3) with glutamate to form glutamine— a nontoxic transport form of ammonia (Figure 11). The glutamine is transported in the blood to the liver where it is cleaved by glutaminase to produce glutamate and free ammonia

The second transport mechanism, used primarily by muscle, involves transamination of pyruvate (the end product of aerobic glycolysis) to form alanine. Alanine is transported by the blood to the liver, where it is converted to pyruvate, again by transamination. In the liver, the pathway of gluconeogenesis can use the pyruvate to synthesize glucose, which can enter the blood and be used by muscle—a pathway called the glucose-alanine cycle.

UREA CYCLE:

Urea is formed from ammonia (NH3), amino group (NH2) of Asp, and bicarbonate (HCO3-) by urea cycle in liver.



- Five enzyme s are involved in urea synthesis in urea cycle.
Two enzymes are in mitochondrion. Three enzymes are in cytosol.
- Therefore, the urea cycle occurs partially in the mitochondrion and partially in the cytosol.(Fig12)

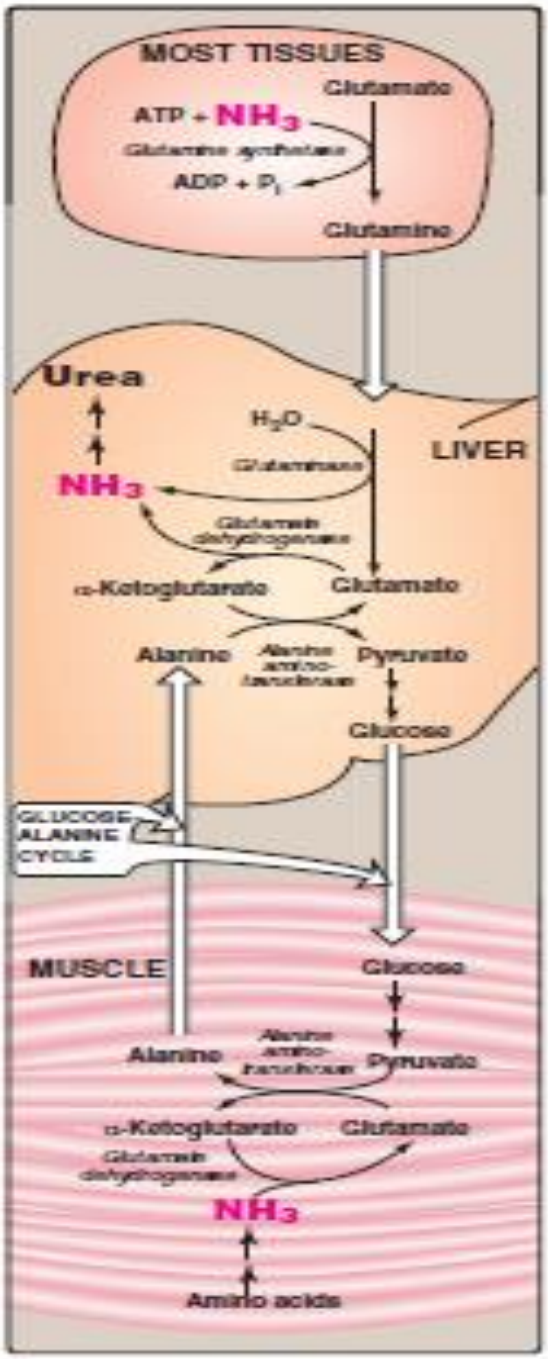


Figure 11
Transport of ammonia from peripheral tissues to the liver.

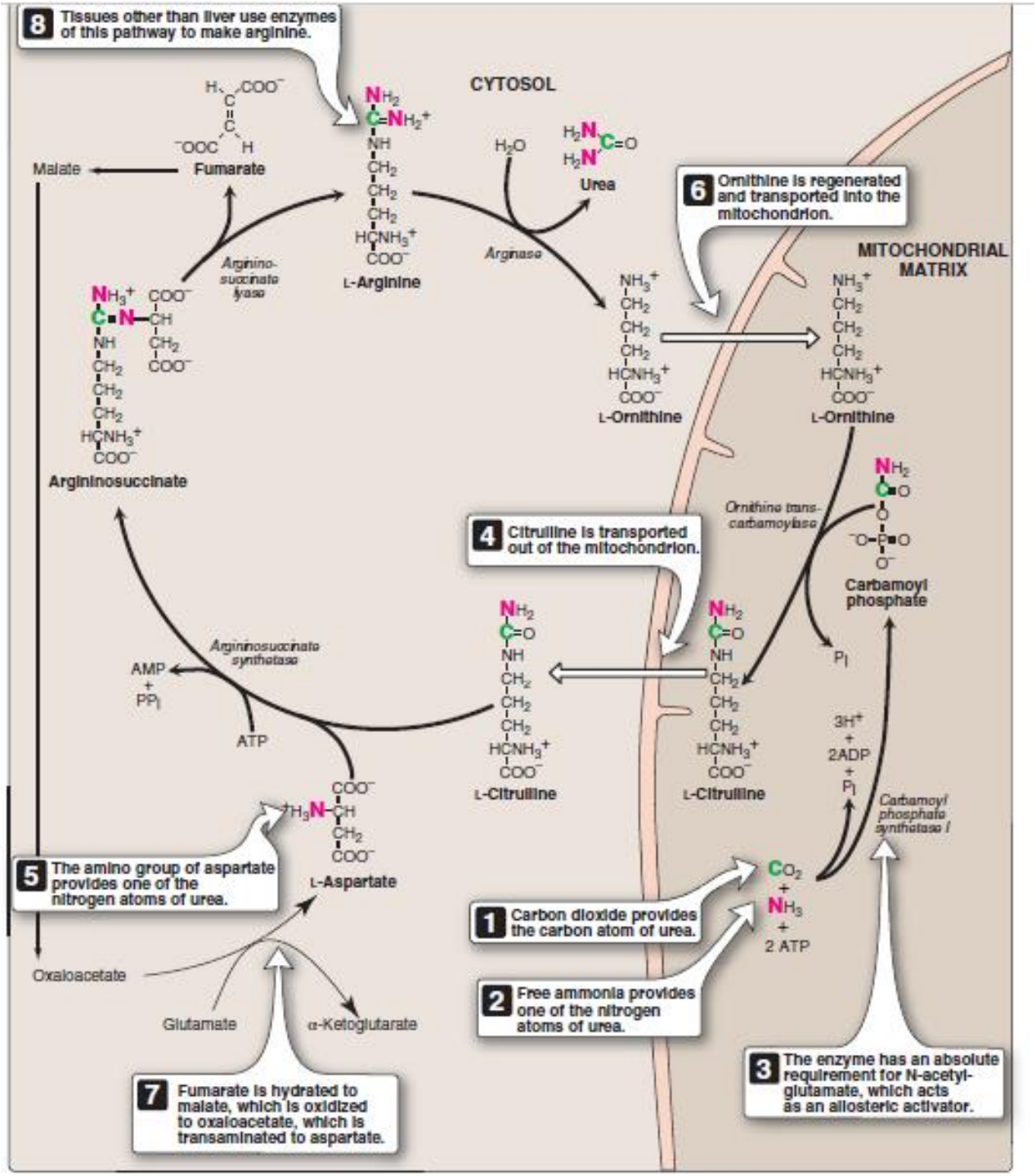
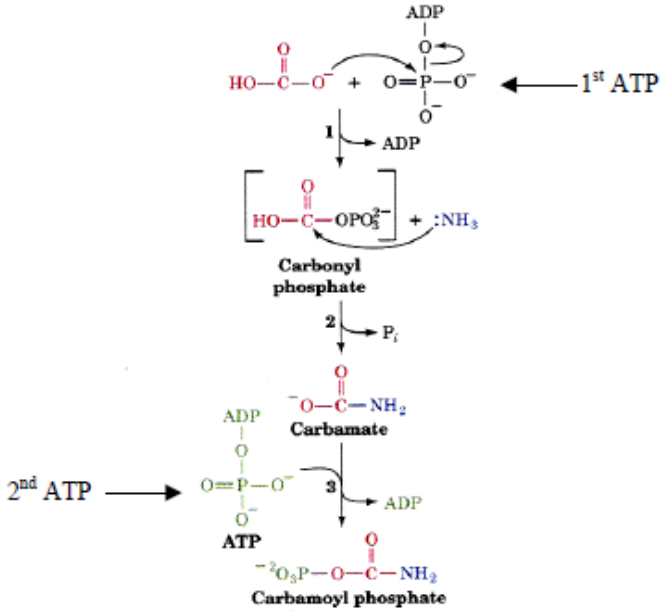


Fig. 12 Urea Cycle

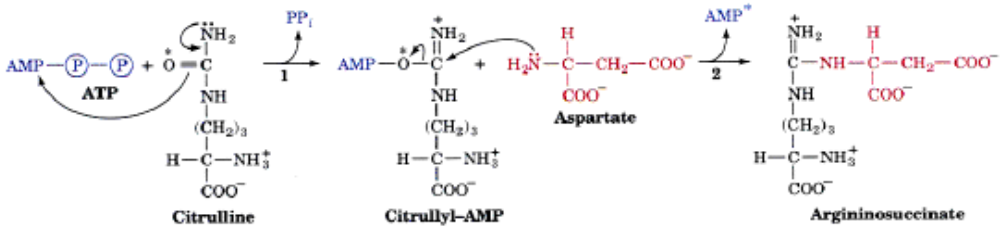
Reactions in urea cycle

1. Carbamoyl phosphate synthetase (Regulating enzyme)
   Formation of carbamoyl phosphate from NH_3 and HCO_3^- (bicarbonate) using ATP as energy source.
   $HCO_3^- + NH_3 + 2ATP \rightarrow H_2N\text{-}CO(OPO_3^{2-}) + 2ADP + P_i$



2. Ornithine transcarbamoylase
   Transfer carbamoyl group ($O{=}C\text{-}NH_2$) to ornithine to produce citrulline.
   Ornithine + $O{=}C\text{-}NH_2(PO_3^{2-})$ → Citrulline + P_i

3. Argininosuccinate synthetase
   Acquisition of the second urea nitrogen atom from Asp.
   Citrulline + Asp → Argininosuccinate



4. Argininosuccinase
   Elimination of arginine from the aspartate carbon skeleton to form fumarate.
   Argininosuccinate → Fumarate + Arginine

5. Arginase
   Hydrolysis of arginine to yield urea and regenerate ornithine.
   Arginine → Urea + Ornithine

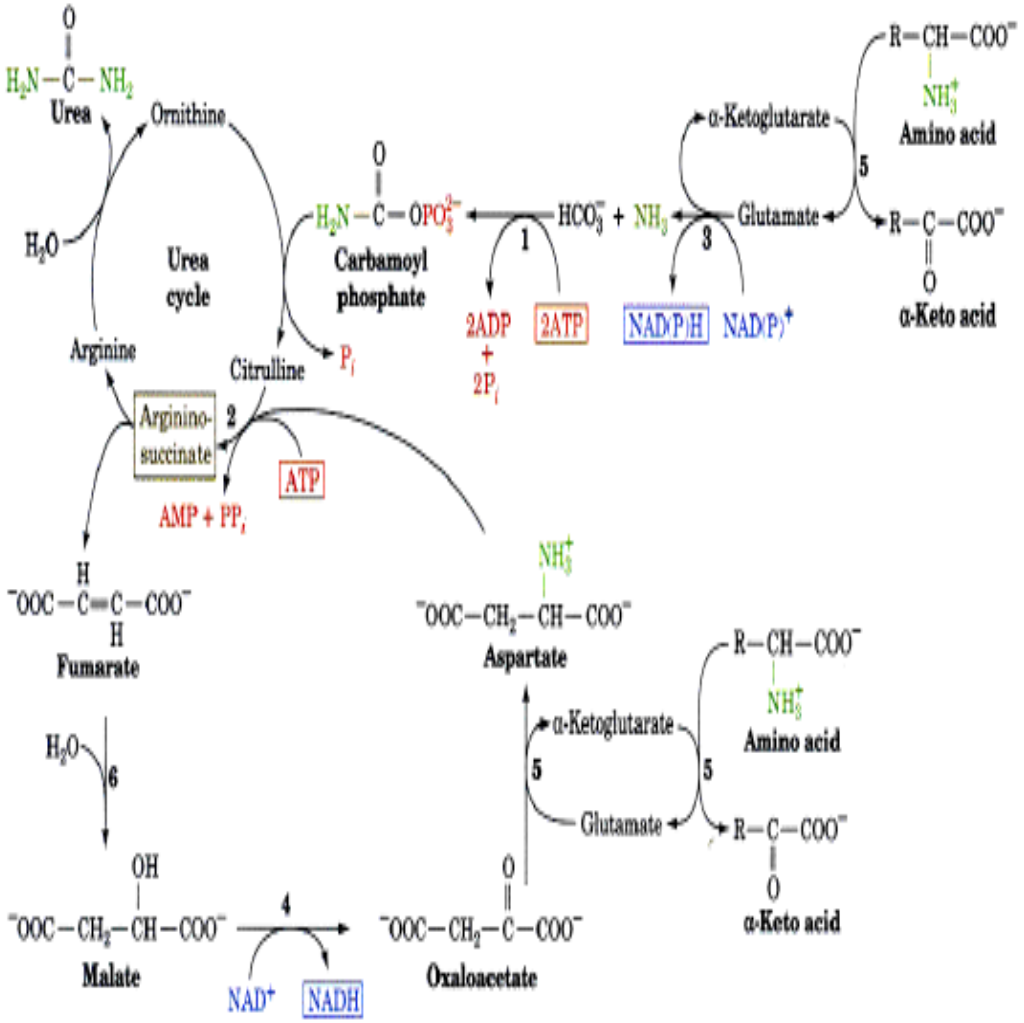
Metabolism 2 Nitrogen compounds metabolism/ protein& Amino Acids Dr.Perry Habib (Lecture :1)

Overall reaction of urea cycle is:

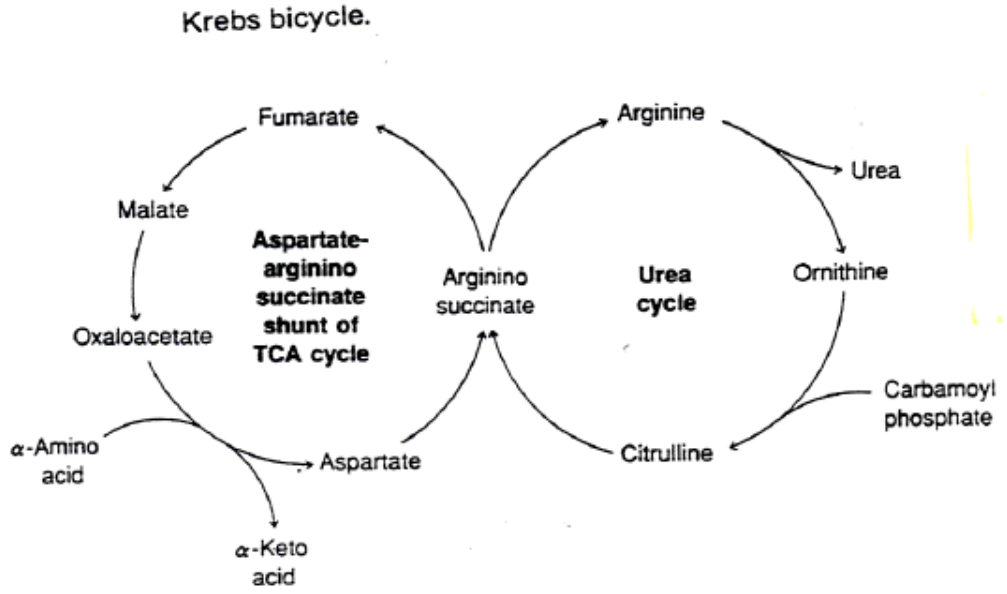
$$CO_2 + NH_3^+ + 3ATP + Asp + 2H_2O \rightarrow Urea + 2ADP + 2P_i + AMP + PP_i (\rightarrow 2P_i) + Fumarate$$

The urea cycle converts two amino groups (one from NH_3 and one from Asp) and a carbon atom (HCO_3^-) to non-toxic excretion product, urea, at the cost of 4 "high-energy" phosphate bonds (i.e., 4ATP).

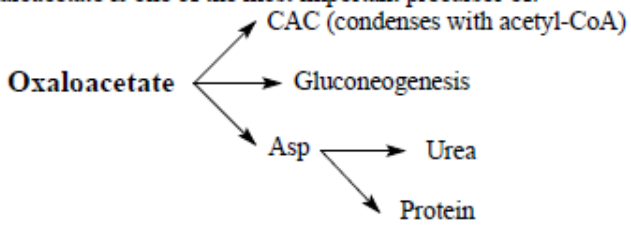
However, oxidations of urea cycle's substrate (Glu) and product (malate) produce 2 NADH (= 6 ATP) as shown in Fig. 24-7.



The urea cycle is conjunct with apartate-argininosuccinate shunt of tricarboxylic acid (TCA) cycle as shown below. This is called "Krebs bicycle". Note: tricarboxylic acid cycle = citric acid cycle = Krebs cycle.

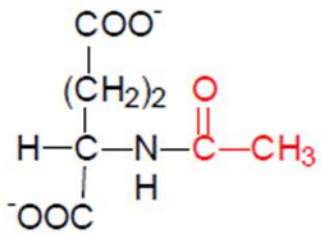


Oxaloacetate is one of the most important precursor of:



Regulation of the urea cycle

- is regulated by carbamoyl phosphate synthetase.
- Carbamoyl phosphate synthetase is allosterically activated by N-acetylglutamate. Thus, N-acetyl-glutamate plays an important role in urea cycle regulation.



N-Acetyl-glutamate

- Acetyl-Glu is synthesized by acetyl-glutamate synthase
  Glu + Acetyl-CoA → N-acetyl-Glu.
- N-acetyl-Glu formation can be as follows:
  1. Breakdown of protein produces amino acids including Glu (i.e., [Glu] ↑).
  2. Need urea cycle to be activated since amino acid degradation produces amines.
  3. In the mean time, ↑[Glu] causes [N-acetyl-Glu] ↑
  4. ↑[N-acetyl-Glu] increases the activity of carbamoyl phosphate synthetase. Thus, urea cycle is activated.