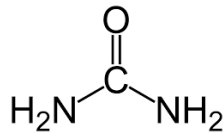


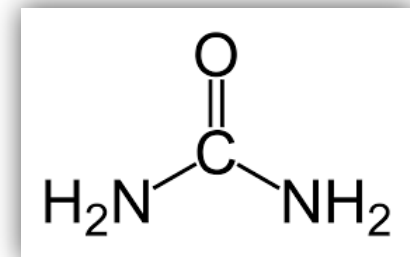
Estimation of Serum Urea

BCH 472



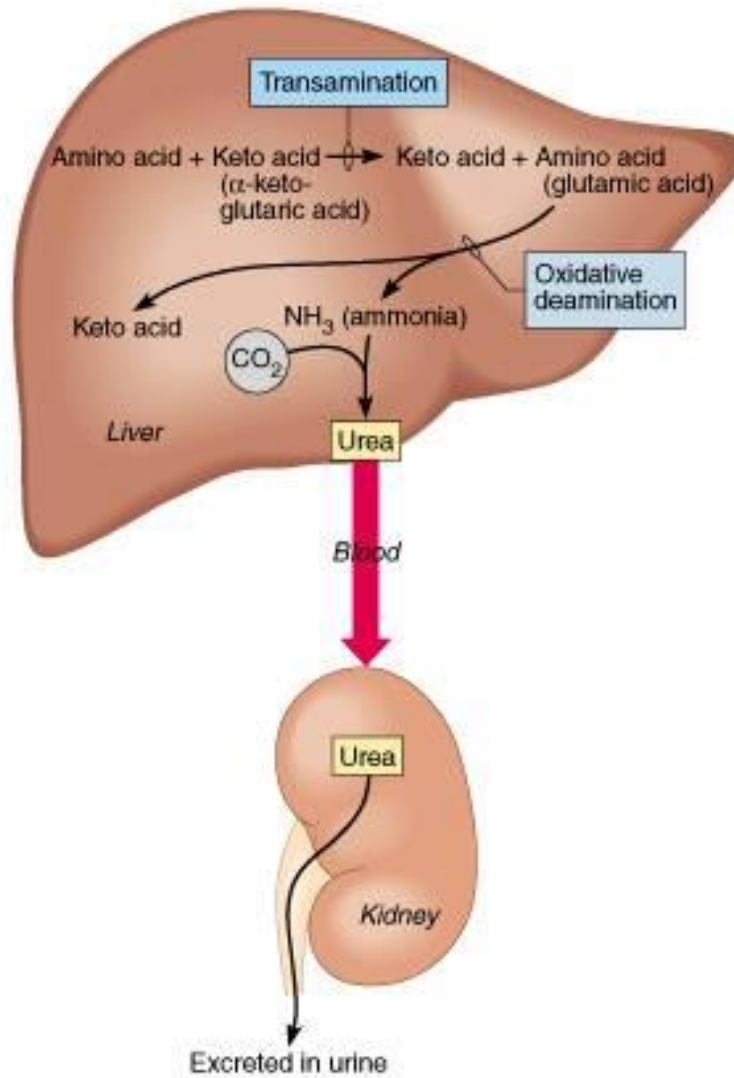
Urea:

- Urea is the highest non-protein nitrogen compound in the blood.
- Urea is the major excretory product of protein metabolism.
- It is formed in the liver from free ammonia generated during protein catabolism.
- the most important catabolic pathway for **eliminating excess nitrogen** in the human body.
- Since historic assays for urea were based on measurement of nitrogen, the term **blood urea nitrogen** (BUN) has been used to refer to urea determination.



Urea synthesis :

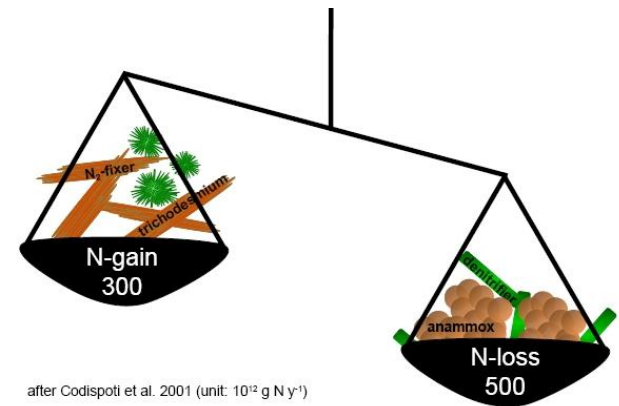
- Protein metabolism produces amino acids that can be oxidized, this result in the release of **ammonia** which is converted to urea (via **urea cycle**) and excreted as a waste product.
 - Following synthesis in the liver, urea is carried out in the blood to the kidney which is readily filtered from the plasma by glomerulus.
 - **Most** of the urea in the glomerular filtrate **excreted** in the urine, and **some** urea is **reabsorbed** through the renal tubules.
 - The amount reabsorbed depends on urine flow rate and extent of hydration .
 - The **concentration** of urea in the plasma is determined by renal function, the protein content in diet and the rate of protein catabolism .
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Clinical Application:

Measurement of urea used in :

- Evaluate renal function
- To assess hydration status
- To determine nitrogen balance
- To aid in the diagnosis of renal diseases
- To verify adequacy of dialysis
- Check a person's protein balance



Urea Concentration-plasma:

- Measurement of Blood urea nitrogen (**BUN**) alone is **less useful** in **diagnosing kidney diseases** because it's blood level is influenced by dietary protein and hepatic function .
- But its diagnostic value improves **with serum creatinine values**.

	Type	Cause	Note
High urea (High urea concentration in plasma is called (azotemia))	Pre-renal	<ul style="list-style-type: none"> • Cognitive heart failure • Dehydration • High protein diet • Increased protein catabolism 	Caused by reduced renal blood flow, less blood is delivered to kidney , then less urea is filtered.
	Renal	<ul style="list-style-type: none"> • Renal failure 	
	Post renal	<ul style="list-style-type: none"> • Urinary tract obstruction 	
Low urea		<ul style="list-style-type: none"> • Low protein intake (Starvation, anorexia) • Liver disease • Pregnancy 	

Urea Concentration-Urine:

- The **urine urea nitrogen test (UUN)** determines how much urea is in the urine to assess the amount of protein breakdown. The test can help determine how well the kidneys are functioning, and if the intake of protein is too high or low.
- Specimen: The urine urea nitrogen test is performed by collecting a 24-hour urine sample urine sample.

	Cause
High urea in urine	<ul style="list-style-type: none">- too much protein in the diet- too much protein breakdown in the body
Low urea in urine	<ul style="list-style-type: none">- malnutrition- too little protein in the diet- <u>kidney issues</u>

Practical Part

Experiments

Estimation of Blood urea

Objective:

- Estimation of Blood urea nitrogen (BUN)
-

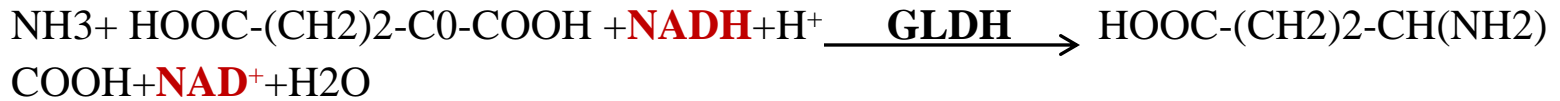
Principle(of the kit used):

The Reagent used contains: Urease, Glutamate Dehydrogenase, NADH, α -ketoglutaric acid, buffers and stabilizers

1. **Urea** is hydrolyzed in the presence of urease enzyme and water to yield ammonia and carbon dioxide.



2. The ammonia reacts with α -ketoglutaric acid and reduced nicotinamide adenine dinucleotide (NADH) in the presence of glutamate dehydrogenase (GLDH) to yield glutamic acid and nicotinamide adenine dinucleotide (NAD).



α -KETOGLUTARIC ACID

GLUTAMIC ACID

Note:

The rate of oxidation of NADH to NAD is measured at 340 nm over a limited urea concentration range and limited time period, and is **proportional** to the concentration of urea

Method:

	Standard	Serum
Reconstituted Reagent	3ml	3ml
Pre-warm at 37°C for 2 min. and add:		
Standard	0.025/25 μ l	-
serum	-	0.025/25 μ l

- After exactly 30 seconds . read and record absorbance A1 against **distilled water** at 340 nm.
 - At exactly 60 seconds, read and record the absorbance A2 and determine ΔA
-

Calculations of the Results :

UREA NITROGEN	UREA
SERUM OR PLASMA	
$\text{Urea Nitrogen (mg/dL)} = \frac{\Delta A (\text{Sample}) \times 25}{\Delta A (\text{Standard})}$	$\text{Urea (mg/dL)} = \frac{\Delta A (\text{Sample})}{\Delta A (\text{Standard})} \times 53.57$

Discussion:

Comment on the level of Urea in serum .

Reference Value:

SPECIMEN	UREA NITROGEN	UREA
Serum/Plasma	5-23 mg/dL	10-50 mg/dL
Urine 24 h	9-16g/24h	20-35 g/24 h

Questions:

Why liver disease cause a decrease in blood urea?

What is positive nitrogen balance ? And when it occur?
