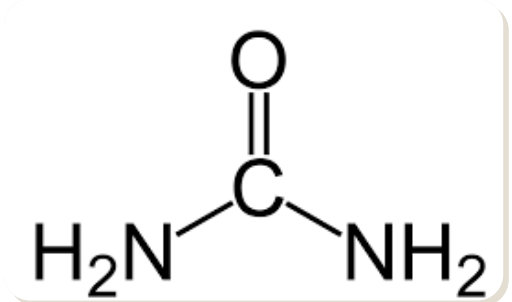


# Estimation of Serum Urea

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## -Urea:

- Urea is the highest non-protein nitrogen compound in the blood.
- Urea is the major excretory product of protein metabolism.
- It is formed by urea cycle in the liver from **free ammonia** generated during **protein catabolism**.
- 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.

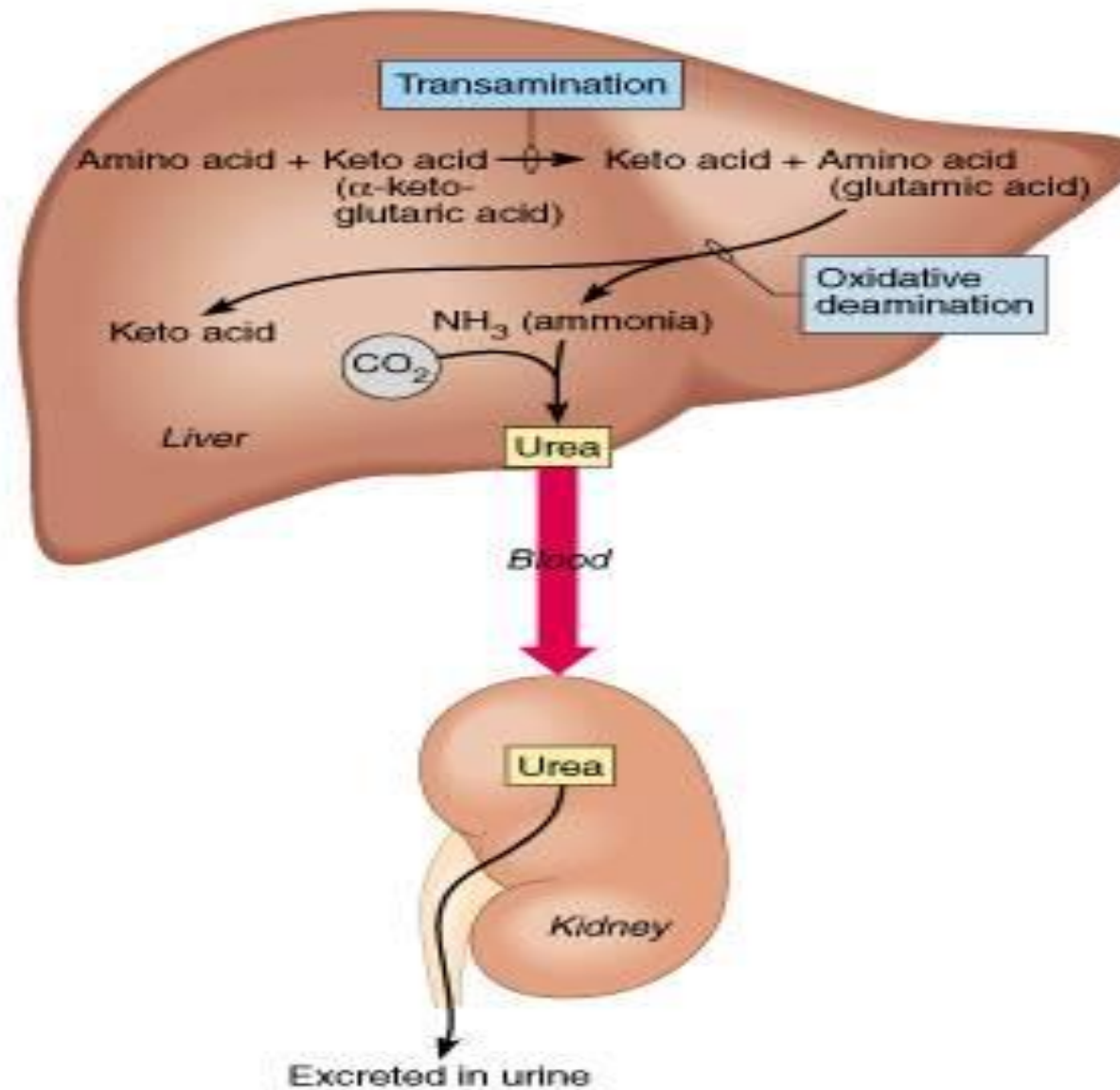


## -Urea synthesis:

- **Protein metabolism** produces amino acids that can be oxidized.
- This results 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 amount of urea reabsorbed increases with dehydration).

- The concentration of urea in the plasma is determined by renal function, the protein content in diet and the rate of protein catabolism.

## -Urea synthesis:



## - 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.
- Check a person's protein balance.

# 1-Plasma urea Concentration:

- 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**.

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

## 2-Urine urea Concentration:

- 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.

	Cause
High urea in urine	<ul style="list-style-type: none"><li>• Too much protein in the diet.</li><li>• Too much protein breakdown in the body.</li></ul>
Low urea in urine	<ul style="list-style-type: none"><li>• Malnutrition.</li><li>• Too little protein in the diet.</li><li>• Kidney issues.</li></ul>



# Practical Part

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## -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. **Reaction one:** Urea is hydrolysed in the presence of urease enzyme and water to yield ammonia and carbon dioxide.



2. **Second reaction:** 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).



## -Reference Value:

SPECIMEN	UREA
Serum/Plasma	10-50 mg/dL
Urine 24 h	20-35 g/24 h

## -Materials:

- BUN-ZYME Reagent: UREASE, GLDH, NADH,  $\alpha$ -KETOGLOUTARIC ACID , buffers and stabilizers.
- BUN-ZYME Standard solution 25 mg/dl ( nitrogen = 53.57 mg/dl).
- BUN-ZYME Serum sample.

# -Method:

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

- After exactly 30 seconds . read and record absorbance A1 against distilled water at 340 nm.
- At exactly 60 seconds after A1, read and record the absorbance A2 and determine  $\Delta A$  (A1-A2).

## -Calculations of the Results :

### - Concentration of urea in serum sample:

- Standard concentration= 53.57 mg/dl

$$\text{- Urea (mg/dL)} = \frac{\Delta A (\text{Sample})}{\Delta A (\text{Standard})} \times 53.57$$

## -Discussion:

- Comment on the level of Urea in serum .