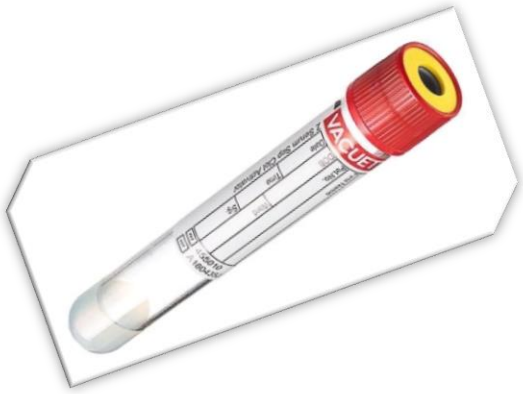
Ministry of Higher Education and Scientific Research

Al- Mustaqbal University- College of Science

Department of Medical Biotechnology

Laboratory Practical: Blood Urea Test (BUN Test)



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1. Introduction

Urea is the final product of protein metabolism in the liver, and it is excreted by the kidneys.
Measuring blood urea levels helps in assessing kidney function, hydration status, and some metabolic disorders.

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2. Purpose of the Test

- Evaluate kidney function.
- Detect acute or chronic renal failure.
- Assess dehydration or high-protein intake.
- Monitor patients in intensive care units.

3. Principle of the Test

The most commonly used method is the **Urease Enzymatic Method**:

1. The enzyme urease converts urea into:
$NH_3 + CO_2$

2. The released ammonia reacts with a color reagent to produce a color proportional to the urea concentration.

3. Absorbance is measured using a **spectrophotometer**.

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4. Specimen

- **Serum** or **Heparinized Plasma**
- No special preparation is required.
- Serum should preferably be separated within 30 minutes.
- Store the sample in the refrigerator if testing is delayed.

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5. Reagents & Equipment

Equipment:

- Spectrophotometer
- Micropipettes
- Incubator (optional depending on method)
- Test tubes
- Timer

Reagents:

- Urease reagent
- Color developer (phenol–hypochlorite or other commercial reagent system)
- Standard urea solution
- Distilled water

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6. Procedure

Prepare three tubes:

Blank .1

Standard .2

Test (patient sample) .3

Example for a urease enzymatic method:

Tube	Reagent	Standard	Sample
Blank	1.0 ml	–	–
Standard	1.0 ml	10 µl	–
Test	1.0 ml	–	10 µl

Steps:

(1)Mix all tubes well.

(2)Incubate for **10 minutes at 37°C** or **20 minutes at room temperature** (according to kit instructions).

(3)Place tubes in the spectrophotometer.

(4)Zero the instrument using the **Blank**.

(5)Read absorbance at **580–600 nm** (varies by manufacturer).

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7. Normal Range

Serum Urea: 15–45 mg/dL

BUN: 7–20 mg/dL

8. Causes of High Urea

Acute or chronic renal failure

- Dehydration
- Gastrointestinal bleeding
- Fever and infections
- High-protein diet
- Use of some medications (e.g., steroids)

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9. Causes of Low Urea

- Liver diseases
- Malnutrition
- Pregnancy
- Overhydration

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10. Sources of Error

- Delay in serum separation, increasing ammonia levels
- Contamination with urease from external sources
- Using unclean tubes
- High bilirubin or hemoglobin may interfere with color reaction

11. Important Notes for Students

- Check reagent expiry before starting.
- Record every step on the worksheet.
- Read absorbance within the recommended time only.
- Compare results with normal reference ranges.

Thank You

"ربما تحصل على مبتغاك في مرحلة متأخرة جدًا، لكن من خلال هذا التأخر تكون قد قطعت صلتك بأفكارك اليائسة، وتدرّبت على الصبر حتى صار جزءًا متأصلًا من صفاتك، واكتسبت نفسًا قوية اعتادت مشقة الاحتياج. فإن ثمرة التأخر أن تفرح بنفسك القوية أكثر من فرحك بحصولك على مبتغاك."