

This reagent kit is for quantitative estimation of urea in serum, plasma or urine.

PRINCIPLE:

Urease breaks down urea into ammonia and carbon dioxide. In alkaline medium, ammonia reacts with hypochlorite and salicylate to form dicarboxyindophenol, a coloured compound. The reaction is catalyzed by sodium nitroprusside. The intensity of colour produced is measured photometrically at 578 nm (570-620 nm with RED filter)



CLINICAL SIGNIFICANCE:

Increased urea levels can occur in liver disease, congestive heart failure, diabetes, infections, in diseases which impair kidney function and with dietary changes. It is also increased in adrenocortical insufficiency, acute intestinal occlusion, various poisonings, shock, urine retention and raised protein break down. Decreased levels are seen in malnutrition, hepatic failure and pregnancy.

SPECIMEN COLLECTION AND STORAGE:

- Fresh non hemolysed serum is preferred.
- Heparinised, EDTA or oxalated plasma may be used.

PRECAUTION:

- UREA reagent is for invitro diagnostic use only.

REAGENTS:

All the reagents are to be stored at 2-8°C

| | No. of Bottles | |
|--------------------------|----------------|--------|
| | 100 ml | 200 ml |
| Reagent 1 (Enzyme) | 5 | 4 |
| Reagent 2 (Hypochlorite) | 1 | 2 |
| Standard (50mg/dl) | 1 | 1 |
| Distilled Water | 1 | 4 |

PREPARATION OF ENZYME REAGENT (E):

A) 100ml: Dissolve the contents of Reagent 1 using 10 ml. distilled water.

B) 200 ml: Dissolve the contents of Reagent 1 using 25 ml distilled water.

Mix well and wait for 10 minutes before use.

REAGENT STORAGE AND STABILITY:

All the reagents included in the kit are stable at 2-8°C until the expiry date stated on the label. Working Enzyme Reagent is stable for 3 weeks at 2-8°C.

Protect the working reagents from light.

GENERAL INSTRUMENT PARAMETERS:

| | |
|----------------------|--|
| Reaction Type | : Two Step / End Point |
| First Incubation | : 3 min. at 37°C |
| Slope of Reaction | : Increasing |
| Reagent (2) Volume | : 1.0 ml |
| Wavelength | : 578 (570-620nm) for 2 ml. procedure 600 (580-650nm) for 3 ml. procedure |
| Second Incubation | : 5 min. at 37°C |
| Flowcell Temperature | : 37°C |
| Std. Concentration | : 50 mg / dl |
| Sample Volume | : 10 µl |
| Units | : mg / dl |
| Working E Reagent | : 1.0 ml |
| Zero setting | : Reagent Blank |

PROCEDURE FOR 2 ML at 578 nm

| Dispense into Test Tubes | Blank | Std. | Test |
|--------------------------------|-------|------|------|
| Working Enzyme E Reagent in ml | 1.0 | 1.0 | 1.0 |
| Standard in ml | - | 0.01 | - |
| Sample in ml | - | - | 0.01 |

Mix incubate at 37° C for 3 minutes, Add

| Reagent 2 volume in ml | 1.0 | 1.0 | 1.0 |
|------------------------|-----|-----|-----|
|------------------------|-----|-----|-----|

Incubate for 5 minutes at 37° C. Read at 578 (570-620 nm)

PROCEDURE FOR 3 ML at 600 nm

| Dispense into Test Tubes | Blank | Std. | Test |
|--------------------------------|-------|------|------|
| Working Enzyme E Reagent in ml | 1.0 | 1.0 | 1.0 |
| Standard in ml | - | 0.01 | - |
| Sample in ml | - | - | 0.01 |

Mix incubate at 37° C for 3 minutes, Add

| Reagent 2 volume in ml | 1.0 | 1.0 | 1.0 |
|------------------------|-----|-----|-----|
|------------------------|-----|-----|-----|

Incubate for 5 minutes at 37° C.

| Distilled water volume in ml | 1.0 | 1.0 | 1.0 |
|------------------------------|-----|-----|-----|
|------------------------------|-----|-----|-----|

Mix, read at 600 (580-650 nm)

STABILITY OF REACTION MIXTURE:

The color of final reaction mixture is stable for one hour.

LINEARITY:

This method is linear up to 250 mg/dl. For Urea concentration higher than linearity limit, mix one volume of sample with one volume of 0.9% saline and multiply the results obtained by two.

CALCULATION:

$$\text{Urea Concentration (mg/dl)} = \frac{\text{Absorbance of Test}}{\text{Absorbance of Std.}} \times 50$$

$$\text{BUN Concentration (mg/dl)} = 0.467 \times \text{Urea Concentration (mg/dl)}$$

$$\text{Urea Concentration (mg/dl)} \times 0.167 = \text{Urea Concentration mmol/lit.}$$

NORMAL VALUES:

| | |
|------------|---|
| Serum Urea | : 10 to 45 mg/dl (1.75 to 7.5 mmol/lit) |
| Serum BUN | : 5 to 21 mg/dl |

PROCEDURE FOR ESTIMATION OF UREA IN URINE:

Dilute the sample 1:50 with distilled water, follow the procedure given for serum urea estimation and calculate the test results as follows:

$$\text{Urea Concentration (Gmg/liter)} = \frac{\text{Abs. of Test}}{\text{Abs. of Std.}} \times 50 \times \frac{\text{Dilution factor}}{100} \times 1$$

NORMAL VALUES:

Urine Urea : 20-30 gm/24hrs.

BIBLIOGRAPHY:

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| | Attention, see instructions for use | | Consult Instructions For Use |
| | For in vitro diagnostic use only | | Catalog # |
| | Store between 2-8°C | | Lot Number |
| | Do not use if package is damaged | | Date of Manufacturing |
| | Manufacturer | | Use by |