Urine & stool examination: Routine & Microscopy

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Urine and stool analysis

- Definition of Urinalysis
- Composition of urine
- Specimen collection and handling
- Physical examination
- Chemical examination
- Microscopic examination
- Fecal analysis

Definition and purpose of Urinalysis

Clinical Laboratory Standards Institute defines **Urinalysis** as

- "Testing of urine with procedures commonly performed in an **expeditious**, **reliable**, **accurate**, **safe** and **cost effective** manner".

Purpose of Urinalysis:

- Diagnosis of disease
- Monitoring disease progress & therapy
- Screening of asymptomatic population for undetected disorders

Composition of urine

Water - 95%

Solutes - 5%

Solutes:

- **Urea**: product of protein and amino acid metabolism.
- **creatinine**: product of creatine metabolism
- **Uric acid**: product of nucleic acid breakdown
- **Inorganic substances**: chloride, sodium, calcium, potassium, phosphate
- **Other substances**: hormones, vitamins, medications, cells, casts, crystals, mucus and bacteria

Normal Volume: 600 to 2000ml/day.

Specimen collection

Types of Urine specimen:

- Random
- First morning
- Midstream clean-catch
- Timed (24 Hr Urine)
- Catheterized specimen

Container:

- Clean leak proof
- Disposable
- Screw top lids
- wide mouth with flat bottom
- Transparent material
- capacity 50 ml
- sterile for Culture and sensitivity

Label and Requisition

Label must be **attached** to container.

Should have:

- Name
- Identification number
- patients age and sex
- Location
- clinicians name
- date and time of collection.

Requisition:

- Details of label
- Test required
- clinical information
- Medication details
- specific preparation (fasting)

Criteria for rejection of specimen

- Specimen improperly labeled or unlabeled
- Insufficient sample quantity
- Leaking or broken container
- Wrong container
- Duplicate specimen
- Wrong requisition
- Outside of container grossly contaminated with sample

Urinalysis

Physical Examination:

- Color
- Clarity
- Specific Gravity

Chemical examination:

- pH
- Protein
- Glucose
- Ketones
- Blood
- Bilirubin
- Urobilinogen
- Nitrite
- Leukocyte esterase

Microscopic examination:

- Urine sediment constituents
- Urinary crystals

Physical examination : color

- Normal color : colorless to pale yellow
- Normal Pigments: Urochrome, Uroerythrin and Urobilin

Dark yellow	**Biliverdin, Dehydration, Multivitamin Tablets**
Orange	Phenazopyridine
Red	Blood, Beet, Rifampicin
Brown	Malignant melanoma, Alkaptonuria, Metronidazole
Bluish green	*Pseudomonas aeruginosa* infection
Purple	Due to Indican by *Kleibsiella pneumoniae* and *Providencia spp*

Physical examination: Clarity

- Normal: Clear or transparent
- Hazy: Print can be read (few particles present)
- Cloudy: Print blurred (many Particles present)
- Turbid: Print can not be seen
- Milky: May precipitate or clot
- Causes of turbidity

Non-pathogenic causes	Pathogenic causes
Phosphates Urates Fecal contamination Radiographic contrast media Semen	RBC and/or WBC Bacteria Yeast Abnormal crystals

Physical examination: Specific Gravity

- **Definition:**

  "Density of a solution as compared to distilled water at a similar temperature".

  i. e. Higher the concentration of urine, Higher is Specific gravity.

- Measured by: Refractometer
  Reagent strips
  Osmolality
- Normal value: 1.015 to 1.030
- Isosthenuric: 1.010
- Hyposthenuric: <1.010
- Hypersthenuric: >1.010

Chemical examination: Reagent strips

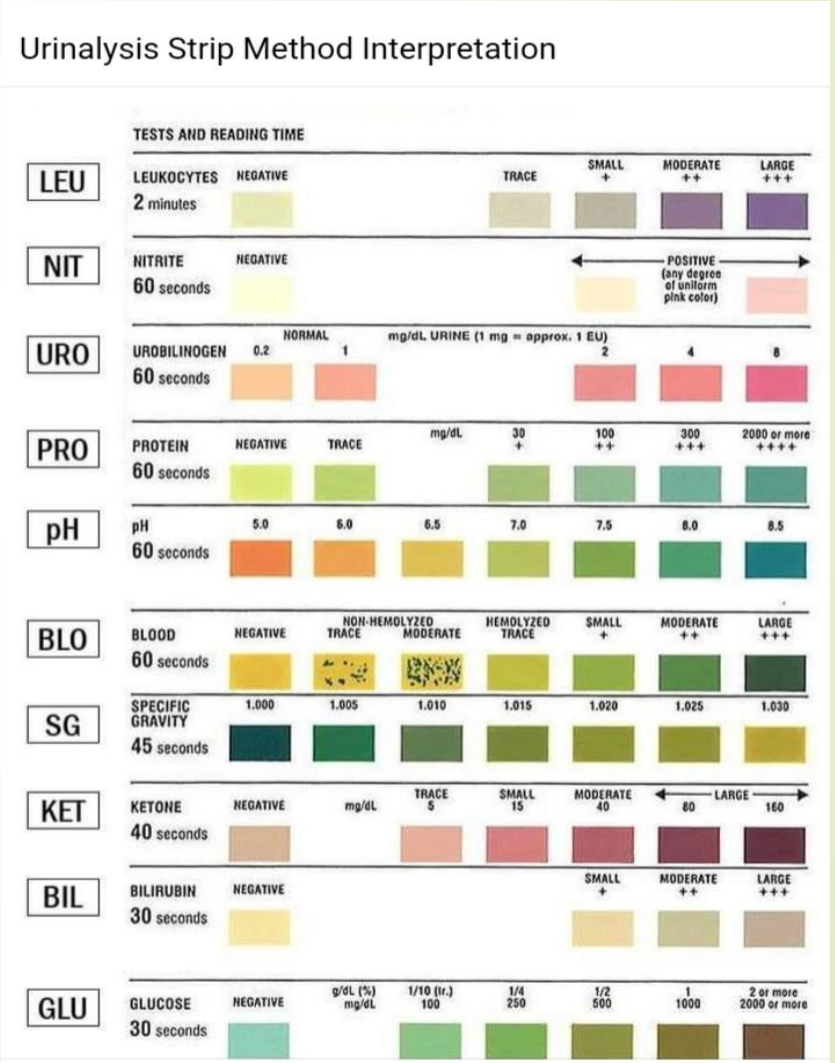
- Reagent strips: Semi-quantitative method
- Has chemical impregnated absorbent pads, attached to plastic strip
- Reported on the basis of comparison of color
- **Method:**

1. Dip the strip completely in well mixed specimen

2. For 60 to 120 seconds

3. Remove excess fluid

4. Compare color with Color chart of manufacturer



Chemical examination: pH

- pH detects existence of systemic acid-base disorders
- Normal value: 4.5 to 8.0
- Reagents: Methyl red- turns yellow to red in acidic medium

  Bromothymol blue- turns blue to green in alkaline medium
- Causes:

Acidic urine	Alkaline urine
Diabetes mellitus Starvation Dehydration Diarrhea Bacteria like *Escherichia coli* Drugs like Fosfomycin	Hyperventilation Vomiting Bacteria like *Proteus mirabilis*

Chemical examination: Proteins

- Normal value: < 10 mg/dL or 100 mg/L
- Indicates early renal disease
- Normal proteins in Urine: Albumin & Uromodulin (Tamm-Horsfall protein)
- Clinical Proteinuria: 30 mg/dL or 300 mg/L

Pre-renal Proteinuria	Renal Proteinuria	Post-renal Proteinuria
Haemoglobin or Myoglobin Not detected by Reagent strips Multiple myeloma (Bence-Jones proteins)	Diabetes mellitus- microalbuminuria Streptococcal Glomerulonephritis Dehydration Hypertension Toxic agents	Bacterial & fungal infections Injury / Trauma

- Color change in Reagent strip from yellow - green – blue.

Chemical examination: Glucose

- Most frequently performed test.
- Useful in detection and monitoring of Diabetes mellitus.
- Fasting is recommended before test.
- Clinical significance:

Hyperglycemia associated	Renal associated
Diabetes mellitus Pancreatitis Pancreatic cancer Cushing syndrome Acromegaly	Fanconi syndrome Osteomalacia Pregnancy Advanced renal disease

- Color indicator: Potassium iodide or tetramethyl benzidine.

Chemical examination: Ketones

- Ketones: intermediate products of fat metabolism.
- consists Acetone, Acetoacetic acid and β-hydroxybutyrate.
- Normally do not appear in urine.
- Clinical significance: Diabetes mellitus
  Vomiting
  Starvation
  Malabsorption
- Reagent strips use Sodium nitroprusside to produce purple color.

Chemical examination: Blood

- Blood in urine: RBC or hemoglobunuria.
- 5 RBC/ ml is significant.
- Clinical significance:

Hematuria	Hemoglobinuria	Myoglobinuria
Renal calculi Glomerular disease Tumours Trauma Pyelonephritis	Transfusion reactions Hemolytic anaemia Severe Burns Malaria	Muscular trauma Prolonged coma Muscle wasting disease Alcoholism Drug abuse

- Reagents strips use pseudoperoxidase activity of hemoglobin.
- Chromogen is oxidised to produce green-blue color.

Chemical examination: Bilirubin & Urobilinogen

- Bilirubin: degradation product of Hemoglobin
- Urobilinogen: degradation product of Bilirubin.
- Clinical significance:

Bilirubin	Urobilinogen
Hepatitis Cirrhosis Biliary obstruction Detected by Diazo reaction Pink to violet	Hemolytic disorders Constipation Ehrlich’s Reaction or Azo coupling reaction Dark pink to red color

Chemical examination: Nitrite

- Rapid test for detection of Urinary tract infection.
- Positive in infections due to Nitrate reducing bacteria (Enterobacterales)
- Detected by Greiss reaction.
- Positive test indicates significant Bacteriuria (100000bacilli/ml)
- Pink color is positive.

Chemical examination: Leukocyte esterase

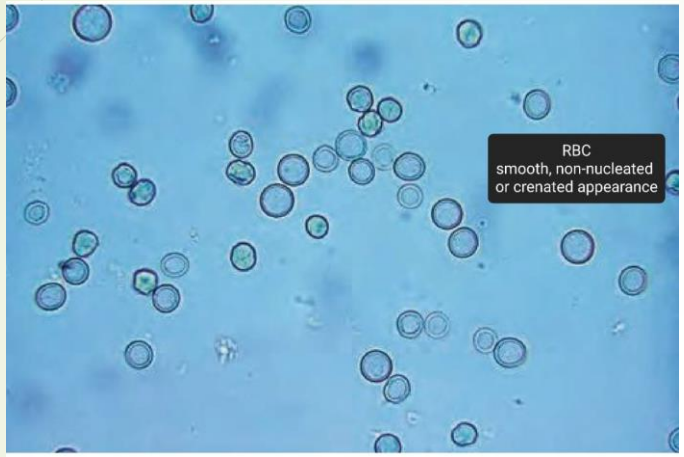
- Detects lysed leukocyte.
- Normal values: 0 to 5 per High power fields.
- Increased urinary leukocytes indicates Urinary tract infection.
- Reagent strip uses hydrolysis of ester.
- positive is purple color.
- clinical significance: Bacterial urinary tract infections
  screening of urine culture specimens

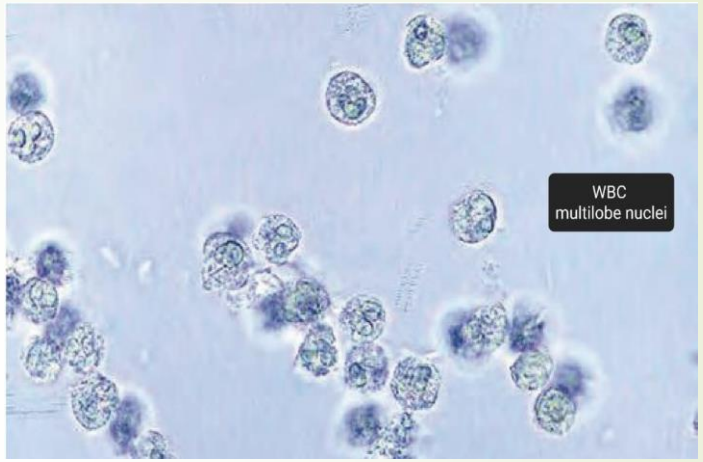
Microscopic examination

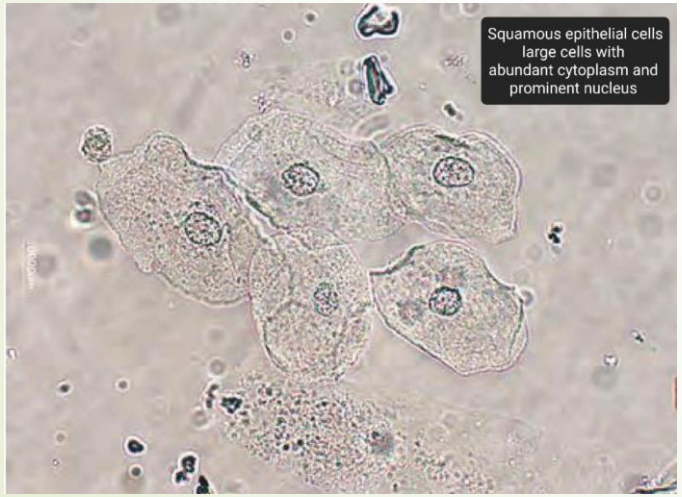
- Specimen preparation:
- volume : 10 12 ml in conical tube
- centrifuge at Relative centrifugal force (RCF) of 400 for 5 min.
- Take 20 µl of sample and cover with 22 x22 cover slip.
- Examine 10 fields under low(10x) and high(40x) power.
- Low power field detects casts.
- high power field detects cells.
- Report: **Urine sediment constituent.**

  Urinary crystals.

Urine sediment constituents







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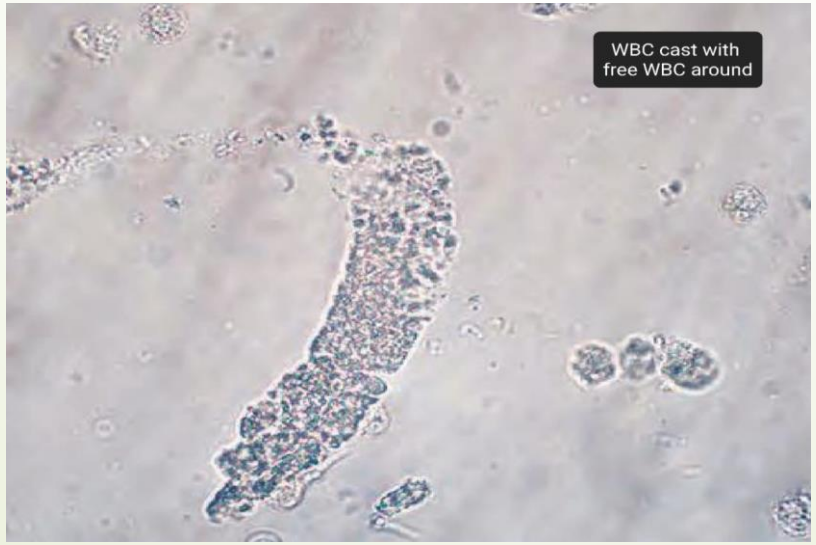
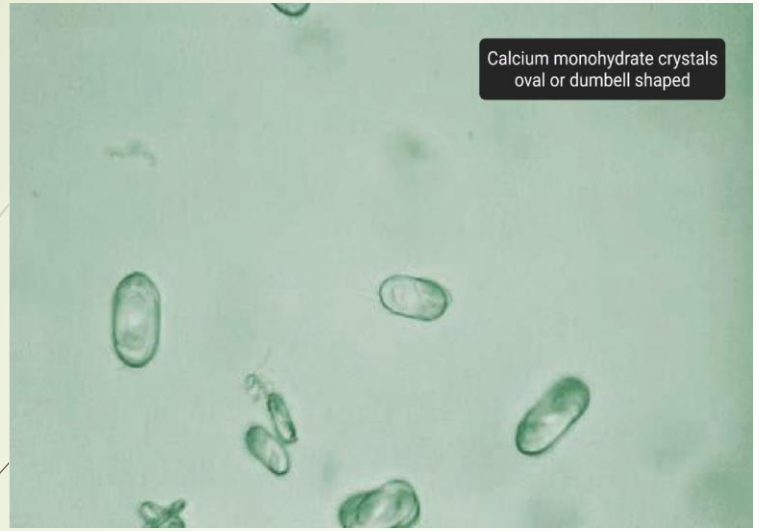


Urinary crystals



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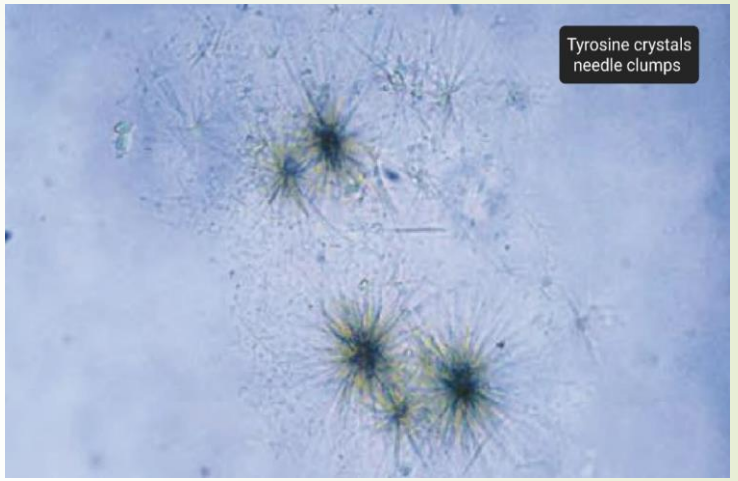


Stool examination

- Macroscopic examination
  - Color
  - Appearance
- Microscopic examination
  - Fecal leukocytes
  - Muscle fibers
- Chemical examination
  - Occult blood

Stool examination: macroscopic

- Color: normally brown

Black	**Upper GI bleeding** **Iron therapy**
Red	Lower GI bleeding Rifampicin
Pale yellow or white	Obstructive Jaundice
Green	Biliverdin

- Appearance:

Bulky	**Bile duct Obstruction**
Mucus	Colitis
Mucus with blood streaks	Dysentery Malignancy

Stool examination: microscopic

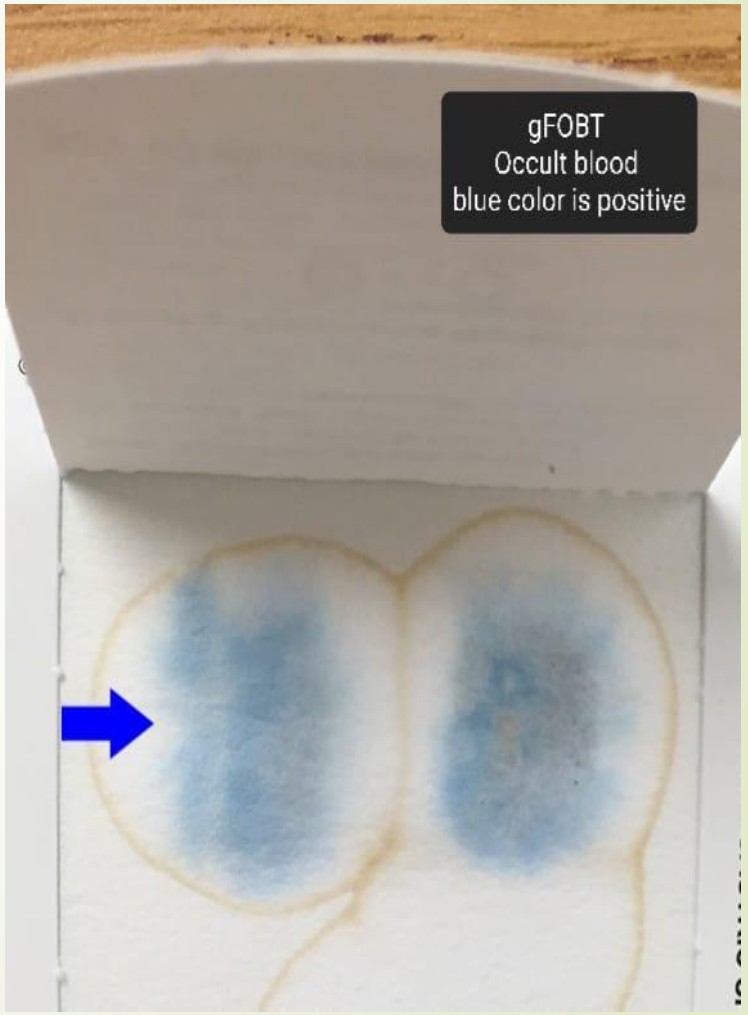
- Leukocytes: Screening for culture specimens

  Presence indicates bacterial infection
- Three WBC per high power field (40x) is significant.
- Muscle fibers:

  Present in Biliary obstruction and gastro-colic fistula.

Stool examination: Occult blood

- Most frequently performed test.
- Bleeding > 2.5 ml per 150 gm of stool is significant.
- Detects blood when no visible bleeding.
- Guaiac based test for Occult blood (gFOBT).
- Based on pseudoperoxidase reaction of hemoglobin.
- Sample is placed on front side of filter paper with applicator stick.
- Hydrogen peroxide added on back side of filter paper.
- Blue color indicates positive test.



Thank you