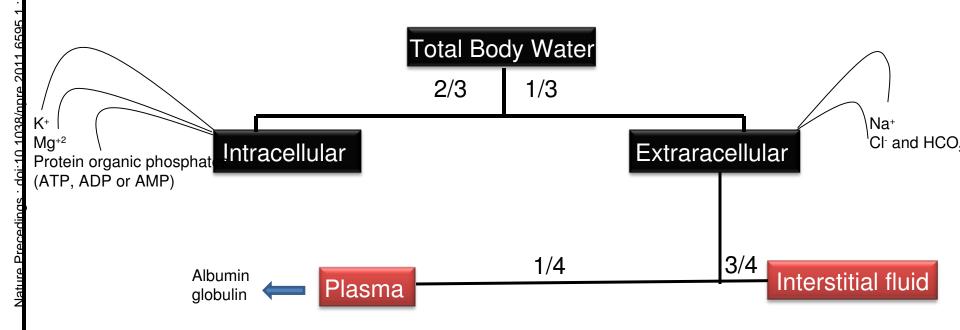
# Biochemical composition of normal urine



Atif Amin Baig Lecturer, Faculty of Medicine and Health Sciences Universiti Sultan Zainal Abidin Highest in newborns and adult males lowest in adult females and in adults with a large amount of adipose tissue



# Function of kidney

Controlling the blood volume and composition

Eliminating the wastes by filtering the blood plasma

Regulating blood volume/ blood pressure

Secretion and hormones

- 1. Rennin
- 2. Erythropoietin
- 3. Calcitrol (Vit D)
- 4. gluconeogenesis

Acid-Base balance
By regulating
pCO<sub>2</sub>

osmolarity

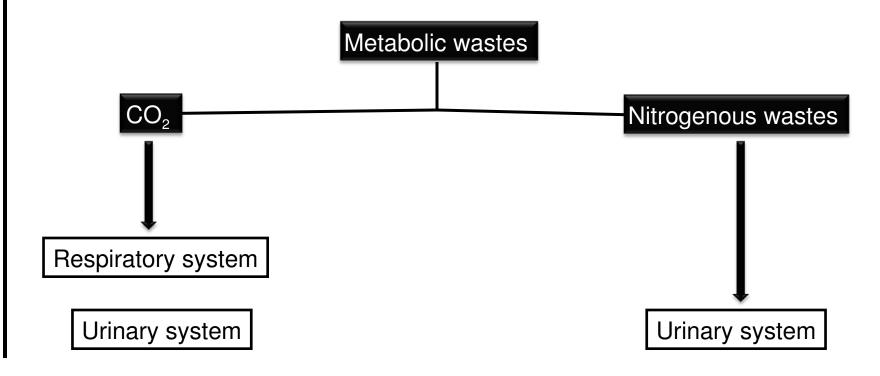
Regulating fluid

#### Detoxification

- Free radicals
- Drugs

### **Human Metabolic Wastes**

 The by product of all chemical reactions in the body that needs to be excrete or else the build up of these compounds will sicken and eventually kill the person.



# Nitrogenous wastes

Protein metabolism

 $2NH_3 + CO_2 \rightarrow H_2NC0NH_2$ 

Urea

Nucleic acid metabolism

Major portion from purines

Uric acid



Creatine

Creatinine

Urea comprised 50% of total nitrogenous wastes in the humans

# **Urine**

Definition
 By definition "urine is a ?"

A liquid containing multiple waste products of metabolism, especially urea and other nitrogenous compounds, that are filtered from the blood by the kidneys. Urine is stored in the urinary bladder and is excreted from the body through the urethra.

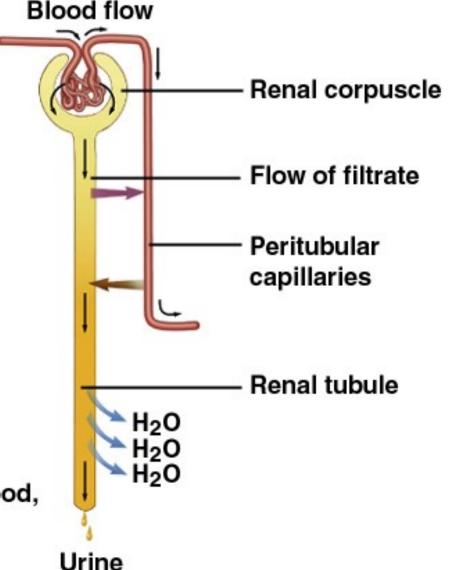
#### **Urine Formation**

Glomerular filtration
 Creates a plasmalike
 filtrate of the blood

2 Tubular reabsorption Removes useful solutes from the filtrate, returns them to the blood

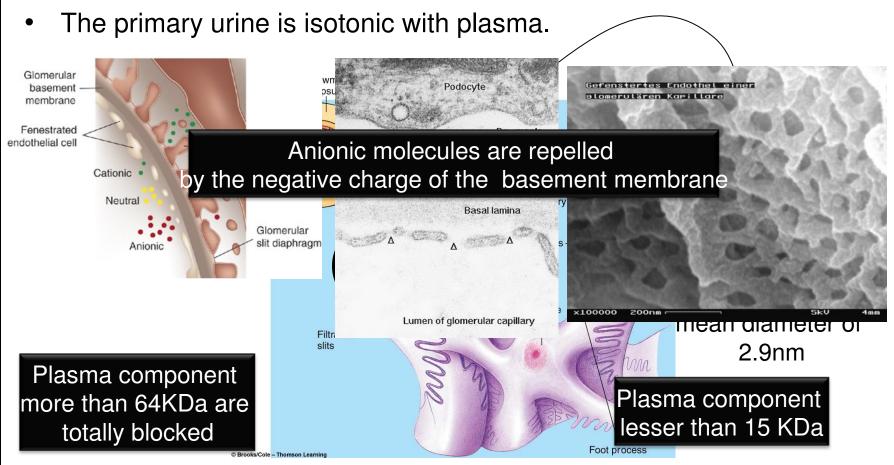
3 Tubular secretion
Removes additional
wastes from the blood,
adds them to the filtrate

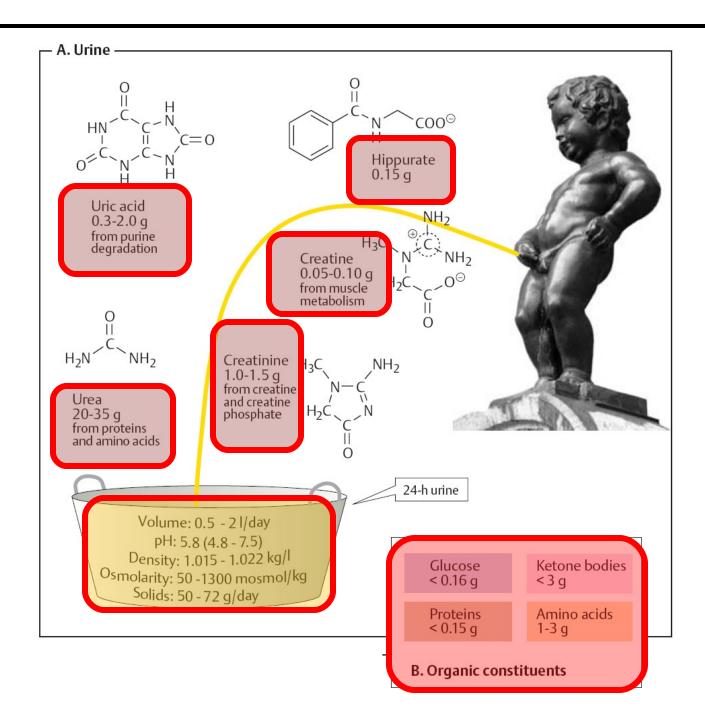
4 Water conservation
Removes water from the
urine and returns it to blood,
concentrates wastes



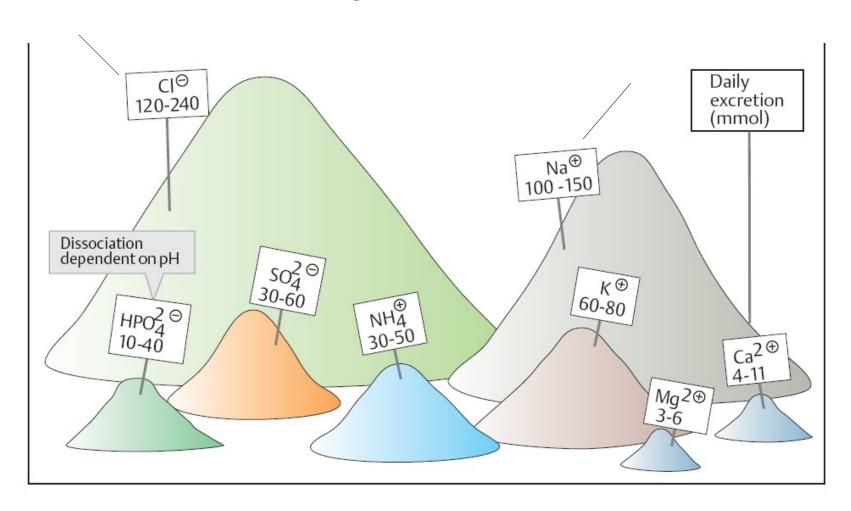
# Ultrafiltration of plasma

Ultrafiltration occurs through glomerulus giving rise to primary urine





#### C. Inorganic constituents



# Urinalysis

- The routine urine examination.
- Most useful tool for the clinicians as an indicator or health or disease.
- Particularly, used in renal metabolic disorders.
- Often done for patients admitted to the hospital.
- The routine urine analysis is divided into four main groups:
- 1. Physico-Chemical properties.
- 2. Chemical examination.
- 3. Microscopic examination.
- 4. Bacterial screening

#### 1. Physico-Chemical properties

Color
Appearance
Volume
Specific gravity
Odor

#### 3. Microscopic examination

WBCs

Epithelial cells

**RBCs** 

Casts

Crystals

Bacteria

Yeasts

Nonbacterial organisms

#### 2. Chemical examination

pH
Sugar
Protein
Ketone bodies
Bilirubin
Urobilinogen
Occult blood
Nitrite
Ascorbic acid

#### 4. Bacterial screening

Gram staining Colony count

### **Urine collection**

- Urine must be collected in a sterile bottle.
- The routine urine examination must be conducted within 30 min.
- In case of delay, urine must be refrigerated



- Decreased pH by the utilization of glucose by bacteria.
- Increased pH by the conversion of urea to ammonia by bacteria.
- In increased pH, the tendency of phosphates to precipitate will increase.
- Oxidation of urobilinogen to urobilin. It may give false negative result for jaundice.

# Types of specimen

Random	First morning	Afternoon	24 hours
Chemical & microscopic examination  Taken any time after first morning	Specimen of choice  Most concentrated	Taken between 2 → 4  pm  Best for detecting  urobilinogen	Used for clearance test of kidney  Samples are collected for 24 hours (for eg,
urine	Microscopia and	ar ozumogom	8am to next day 8 am).
Night	Day	Postprandial	It is to avoid any changes in the
All urine samples are collected in 12 hour of night (for eg, 8pm to 8am)	All urine samples are collected in 12 hour of day (for eg, 8am to 8pm)	Taken after 2 hours of meal	dilution of various suspended constituents of urine

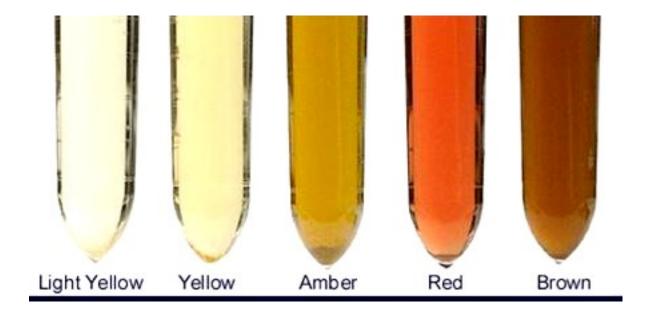
# Physico-Chemical properties

#### Color of normal urine

- Normal urine color ranges from pale yellow to deep amber.
- it is the result of a pigment called urochrome.
  - B vitamins turn urine an eye-popping neon yellow BUT may also indicate liver disease.
  - porphyria, a disease that affects your skin and nervous system, turns urine the color of port wine.

#### Color of normal urine

- Most changes in urine color are harmless and temporary and may be due to:
  - Certain foods beets may turn urine red
  - Dyes in foods/drinks
  - Supplements vitamins
  - Prescription drugs
- Unusual urine color can indicate an infection or serious illness.

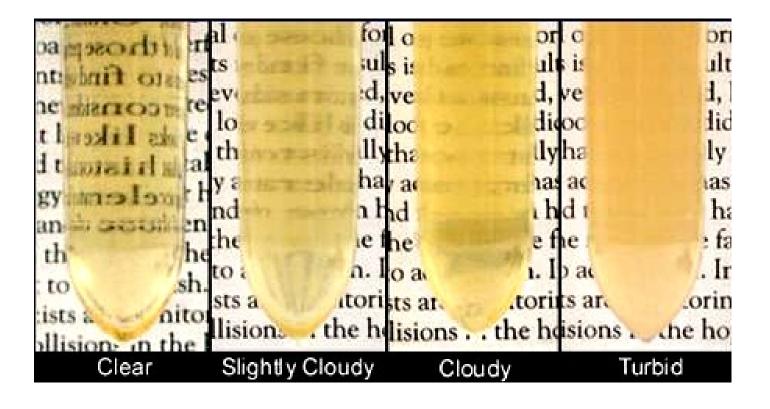


#### **Different colors of urine**

pale yellow (straw)
light yellow
yellow
green-yellow (olive)
red-yellow
red
red-brown
brown-black
black
milky

# Appearance of normal urine

Clarity of urine



# Appearance of urine

- Substances that cause cloudiness but that are not considered unhealthy include:
  - mucous,
  - sperm and prostatic fluid,
  - cells from the skin,
  - normal urine crystals, and
  - contaminants (like body lotions and powders).
- Other substances that can make urine cloudy (such as red blood cells, white blood cells, or bacteria) indicate a condition that requires attention.

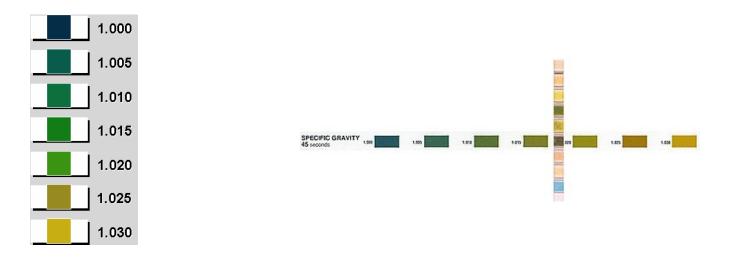
### Volume of urine

- Normal volume is 750 –2000ml/24H
- Under 750 ml (oligouria) dehydration, infection, obstruction, renal stones, kidney, failure, etc).
- Over 2000ml (polyuria) diabetes insipidus, hypertension, nephroticsyndrome, ingestion of alcohol or drugs, endocrinal disorders.
- Absent of urine (anuria) obstruction, kidney failure, stenosis.

#### Odor of urine

- Healthy urine may have a mild smell but generally does not have a foul odor.
- In some cases, an unusual or strong urine odor may be due to benign conditions that are not harmful, such as eating certain foods or taking certain medications and volatile acids.
- In some cases, an unusual or strong urine odor may be due to benign conditions that are not harmful, such as eating certain foods or taking certain medications.

# **Specific gravity**



- Specific gravity reflects kidney's ability to concentrate.
- Want concentrated urine for accurate testing, best is first morning sample.
- Low specimen not concentrated, kidney disease.
- High first morning, certain drugs
- Random specimen (1.003 to 1.032)
- 24 hours specimen (1.015 to 1.025)
- Severe renal tubular damage, sp. Gravity is fixed at 1.010  $\rightarrow$  isothenuric

# Osmolality of urine

- Determination of urinary osmolality is more important than specific gravity because it gives more accurate reflection of the concentration of dissolved substances.
- It is the measure of the moles of dissolved particles (un dissociated molecules and ions) contained in a kilogram (Kg) of a solvent.
- The normal osmolality of random urine specimen varies from 40-1350 m,osmol/kg.
- The normal osmolality of 24 hours urine specimen varies from 500-800 m,osmol/kg.

#### **Chemical Examination**

- Reagent strips are used only once and discarded.
- Testing
  - Perform within 1 hour after collection
  - Allow refrigerated specimens to return to room temperature.
  - Dip strip in fresh urine and compare color of pads to the color chart after appropriate time period.
  - Instruments are available which detect color changes electronically

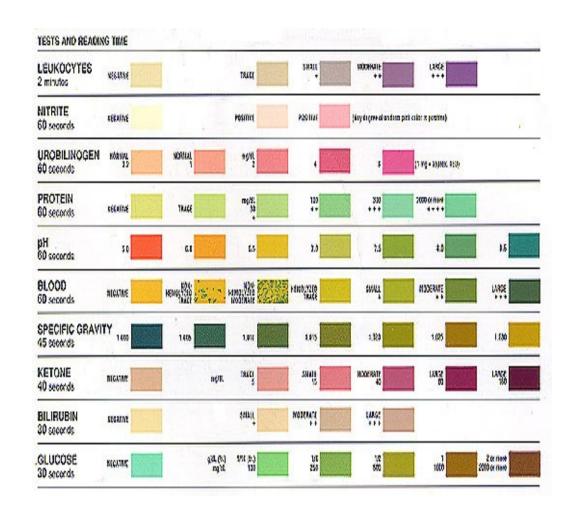
# **Using Reagent Strips**

- BRIEFLY dip the strip in urine.
- Colors are matched to those on the bottle label at the appropriate times.
- Timing is critical for accurate results.

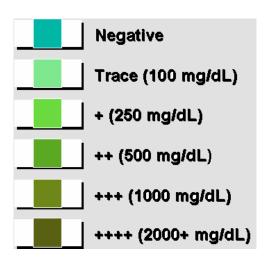


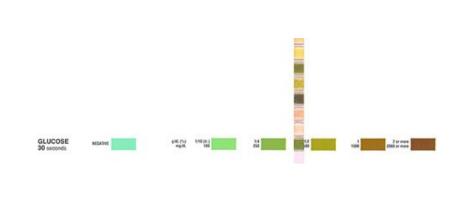
# Reagent Strips





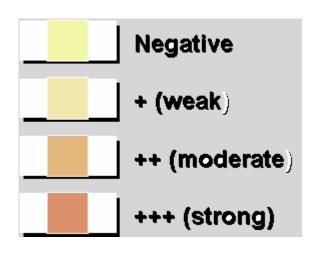
### Glucose

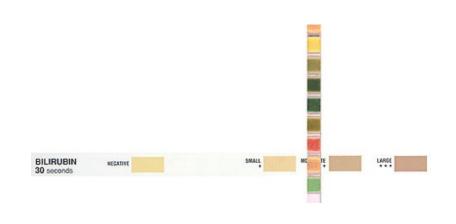




- Presence of glucose (glycosuria) indicates that the blood glucose level has exceeded the renal threshold.
- Useful to screen for diabetes.

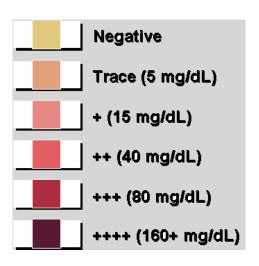
### Bilirubin

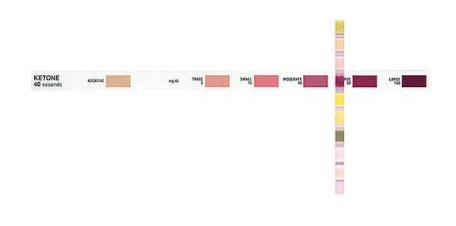




- Bilirubin is a byproduct of the breakdown of hemoglobin.
- Normally contains no bilirubin.
- Presence may be an indication of liver disease, bile duct obstruction or hepatitis.
- Since the bilirubin in samples is sensitive to light, exposure of the urine samples to light for a long period of time may result in a false negative test result.

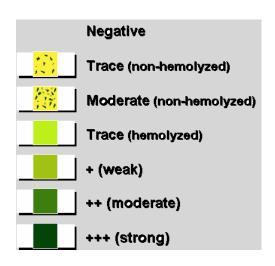
### Ketones

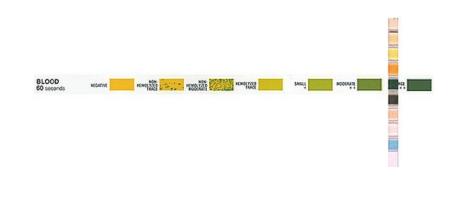




 Ketones are excreted when the body metabolizes fats incompletely (ketonuria)

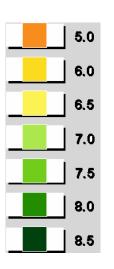
### **Blood**

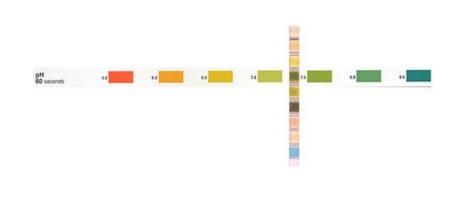




- Presence of blood may indicate infection, trauma to the urinary tract or bleeding in the kidneys.
- False positive readings most often due to contamination with menstrual blood.

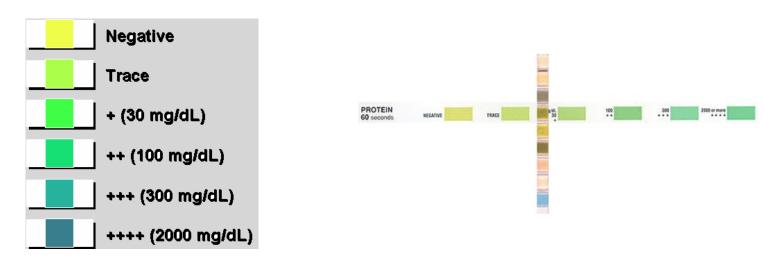
### Ph





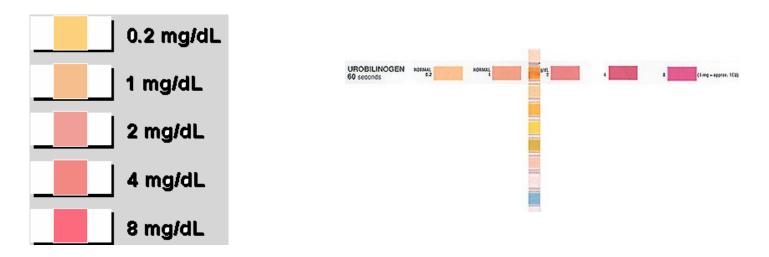
• pH measures degree of acidity or alkalinity of urine

### **Protein**



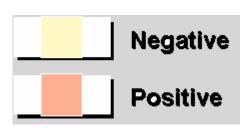
- Presence of protein (proteinuria) is an important indicator of renal disease.
- False negatives can occur in alkaline or dilute urine or when primary protein is not albumin.

# Urobilinogen



- Urobilinogen is a degradation product of bilirubin formed by intestinal bacteria.
- It may be increased in hepatic disease or hemolytic disease

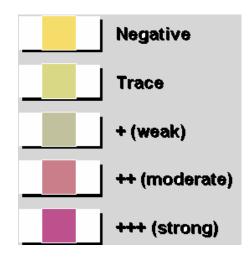
# **Nitrite**





 Nitrite formed by gram negative bacteria converting urinary nitrate to nitrite

# Leukocytes



- Leukocytes (white blood cells) usually indicate infection.
- Leucocyte esterase activity is due to presence of WBCs in urine while nitrites strongly suggest bacteriuria.

### **Normal Values**

- Negative results for glucose, ketones, bilirubin, nitrites, leukocyte esterase and blood.
- Protein negative or trace.
- pH 5.5-8.0
- Urobilinogen 0.2-1.0 Ehrlich units

# Handling and Storage of Strips

- Handling and Storage
  - Keep strips in original container
  - Do not touch reagent pad areas
  - Reagents and strips must be stored properly to retain activity
    - Protect from moisture and volatile fumes
    - Stored at room temperature
  - Use before expiration date

### **Procedure**

- Dip strip briefly, but completely into well mixed, room temperature urine sample.
- Withdraw strip.
- Blot briefly on its side.
- Keep the strip flat, read results at the appropriate times by comparing the color to the appropriate color on the chart provided.





### Sources of Error

- **Timing** Failure to observe color changes at appropriate time intervals may cause inaccurate results.
- **Lighting** Observe color changes and color charts under good lighting.
- QC Reagent strips should be tested with positive controls on each day of use to ensure proper reactivity.
- Sample Proper collection and storage of urine is necessary to insure preservation of chemical.

### Sources of Error

- Testing cold specimens would result in a slowing down of reactions; test specimens when fresh or bring them to RT before testing
- Inadequate mixing of specimen could result in false reduced or negative reactions to blood and leukocyte tests; mix specimens well before dipping
- Over-dipping of reagent strip will result in leaching of reagents out of pads; briefly, but completely dip the reagent strip into the urine

Parameter	Normal	Abnormal
Colour	Yellow straw,	Dark amber - concentrated urine insufficient of fluid intake
	amber,	•Cloudy - infectious process
	transparent	Dark orange – drug, eg: pyridium
		•Red or dark brown - disease process causing blood in
Consistency	Clear liquid	Mucous plug, viscid, thick - infectious process
Odor	Faint aromatic	Offensive - infectious process
Sterility	No	Microorganism present - infectious of UT
	microorganism	
рН	4.5-8.0	Over than 8.0 - UT infection
		•Under 4.5 - uncontrolled diabetes, starvation, dehydration
Specific gravity	1.010 to 1.025	•Over 1.025 – diabetes mellitus, under hydration
		•Under 1.01 – diabetes insipidus, kidney disease, over
Urine glucose	Not present	Present – diabetes mellitus
Urine ketone	Not present	Present – diabetic coma, starvation, prolonged vomiting
bodies (eg:		
Urine blood	Not present	Occult -kidney disease
		Bright red - hemorrhage

- White blood cells
- Pyuria refers to the presence of elevated number of leukocytes (granulocytes):
  - Upper or lower UT infection
  - Glomerulonephrotis
  - Vaginal & cervical infections
  - External urethral meatus (men & women)

#### Normal range:

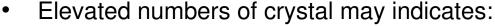
0-2 /HPF (5+ indicates an infection and 10+ indicates more severe conditions).

#### **Epithelial cells**

- Normal urine not present/unquantified
- Renal tubular epithelial cells, usually larger than granulocytes, contain a large round or oval nucleus are normally slough into the urine in small amounts.
- Positive nephrotic syndrome, condition leading to tubular degeneration.

#### **Urinary crystals**

- Normal urine present but unquantified.
- Type of crystals:
  - Calcium oxalate
  - Triple phosphate
  - Amorphous phosphates
  - Uric acids
- Uric acid crystals (hyoeruricemia)
  - Cyctine
  - **Tyrosine**
  - Leusine



- Hipercalcemia
- Cystine crystal -cystinuria
- May due to renal failure or other renal disease.











Oxalate

Triple Phosphate

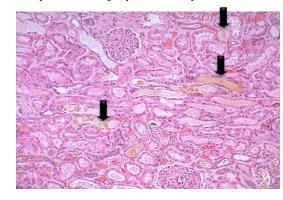
Cystine

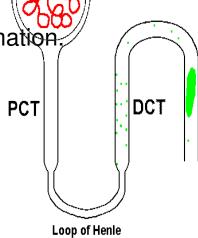
#### **Casts**

- Formed at the distal convoluted tubule or the collecting duct (distal nephron).
- Protein based cylindrical molds of the renal tubule
- Result of damage to the renal tubule
- Can contain cells and other material
- Dehydration and acidic urine especially predisposes to cast formation

#### Types of casts

- Protein
- RBC
- WBC
- Other





### **Urinary bacteria**

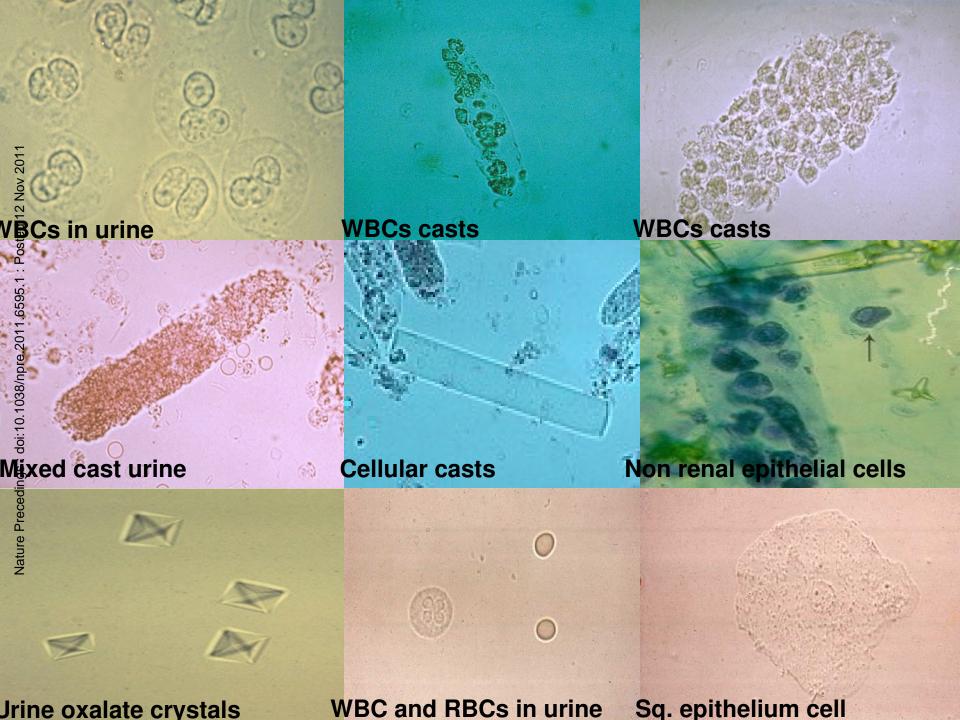
- Abundant normal microbial flora of the vagina or external urethral meatus
- Ability to rapidly multiply in urine standing room temperature.
- However, it should be interpret in view of clinical symptoms.
- Bacterial culture must be done in case of bacteriuria

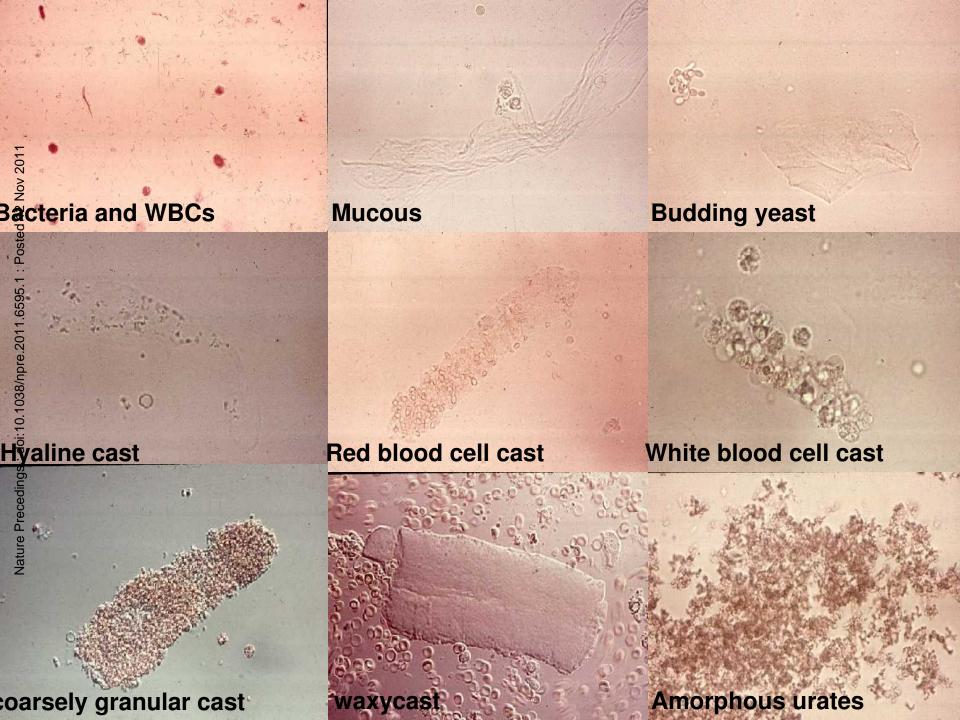
### **Urinary Yeast**

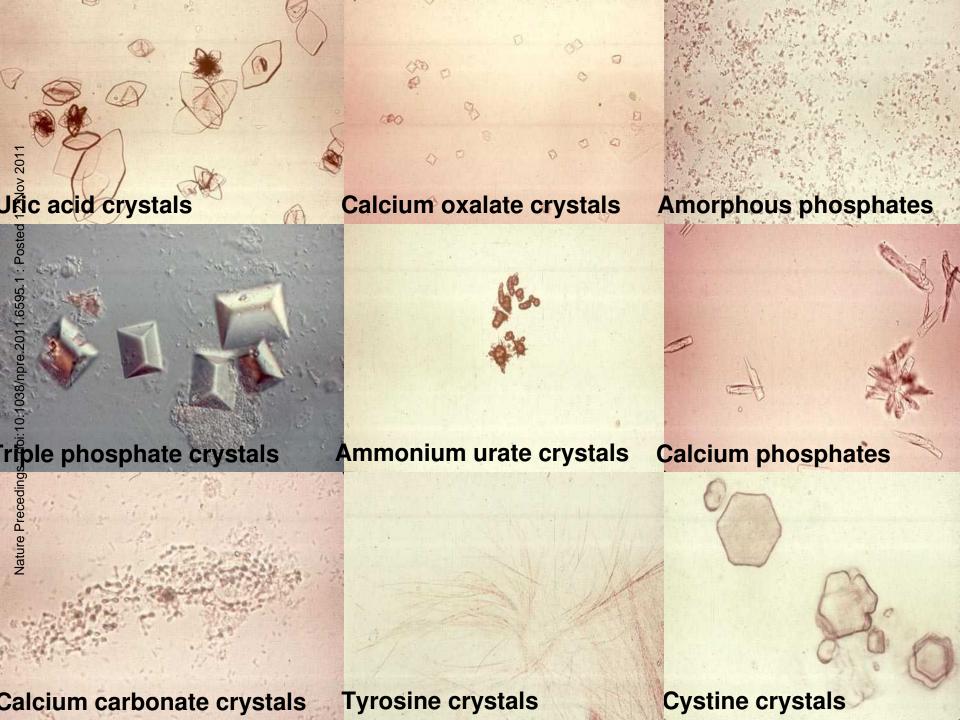
- Yeast cells may be contaminants or represent a true yeast infection.
- Often difficult to distinguish from red cells and amorphous crystals but can be distinguished by their tendency to bud.
- Most often they are candida, which may colonize bladder, urethra or vagina.

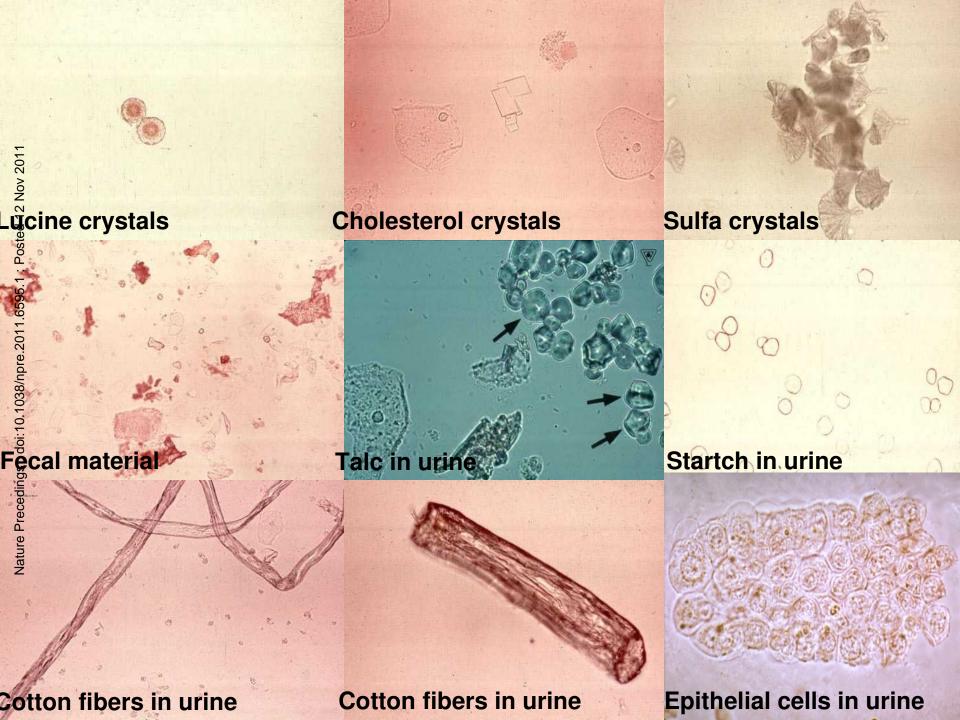
### Non bacterial organisms

- General "crud" or unidentifiable objects may find their way into a specimen, particularly those that patients bring from home.
- Spermatozoa can sometimes be seen.
- Rarely, pinworm ova may contaminate the urine.
- ova from bladder infestations with schistosomiasis may be seen.









# Requesting a urinalysis

#### Signs and symptoms

- Burning urination, difficulty in urination & painful urination infectious and obstruction.
- Semen in urine with great exhaustion (semonorrhoea) prostate problem.
- Backache early in the morning infection, renal stones (fullnes of kidney, pelvis).
- UTI (leucorrhoea in females) spread of infection into UT (honeymoon cystitis).
- Fatigue and exhaustion renal problems.
- Lower abdominal pain or supra pubic pain due to infection or stones.

# Thank you