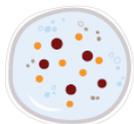


URINE DIPSTICK AND SULPHOSALICYLIC ACID TEST

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A Theoretical & Practical International Course

URINE EXAMINATION

MASTERING THE BASICS

KIDNEY DISEASE?

- severity of kidney disease = estimating GFR
- cause of kidney disease = urinalysis

URINE EXAMINATION

1. macroscopic urine examination
2. dipstick test and sulphosalicylic acid test
3. quantitative and qualitative measurement of proteinuria
4. urine sediment examination - urine microscopy

DIPSTICK TESTING

- reagent strip – dipstick
- rapid semi quantitative assessment of urinary characteristics
- manual method
- method with automatic machines

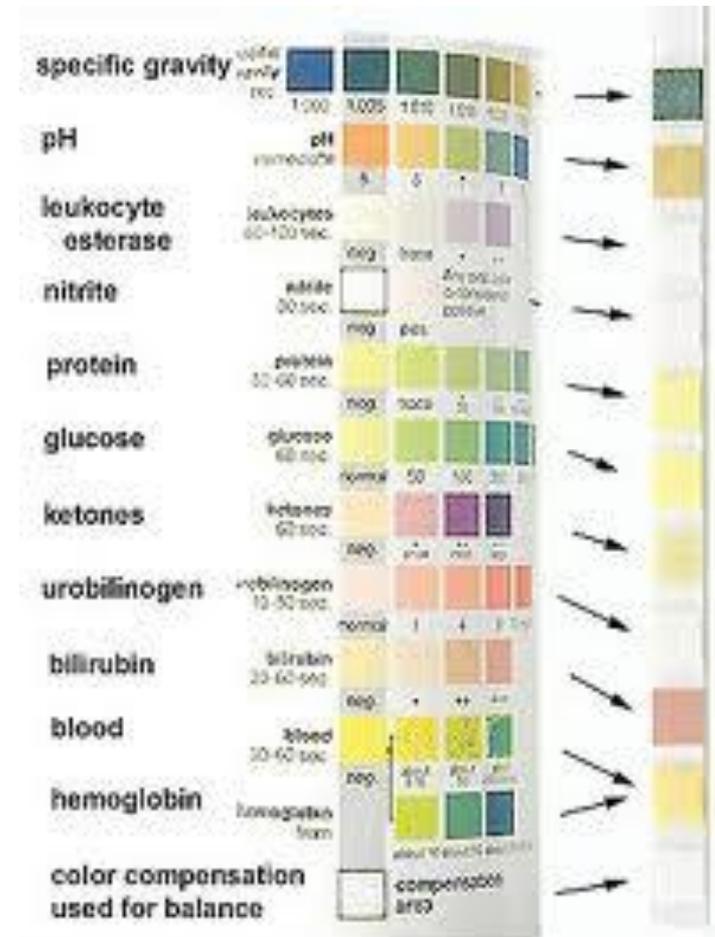
- different dipsticks – be aware





DIPSTICK TESTING

- specific gravity
- pH
- leukocytes
- nitrite (bacteria)
- protein (albumin)
- glucose
- ketones
- pigments (bilirubin, urobilinogen)
- hemoglobin (erythrocytes)



SPECIFIC GRAVITY

- defined as the weight of the solution compared with the weight of an equal volume of distilled water
- gives information about **concentration ability of the kidney**
- dipstick detects only ionized substances and not glucose or X ray contrast agent → measured specific gravity of urine is lower
- when is low (1.005) → cells can burst → the dipstick can detect cells, but under the microscope they can not be seen
- we can measure: 1.000, 1.005, 1.010, 1.015, 1.020, 1.025, 1.030

pH

- reflects the degree of acidification of the urine
- pH: 4.5 – 8.0, average 5.0 - 6.0 – slightly acidic
- we can measure: 5, 6, 7, 8, 9
- **Why is important to know pH of the urine?**
 - in metabolic disorders (metabolic acidosis)
 - in renal disorders
 - in patient with kidney stones
 - in patient with urinary tract infection

LEUKOCYTES - LEUKOCYTE ESTERASE

- presence of neutrophils and macrophages is indicated by a **positive leukocyte esterase test**
- we can measure: neg, 1+, 2+, 3+

LEUKOCYTES - LEUKOCYTE ESTERASE

- **false-positive results:**

- strong oxidizing agents (strong detergents)
- vaginal discharge
- some preservatives (formalin)
- drugs: nitrofurantoin, imipenem, meropenem, clavulanic acid
- excessively dilute urine

LEUKOCYTES - LEUKOCYTE ESTERASE

- **false-negative results**

- concentrated urine
- proteinuria
- glycosuria
- drugs (ascorbic acid, oxalic acid, gentamicin, tetracycline)

NITRITE

- **nitrite test** is a rapid, indirect method for detection of significant and asymptomatic **bacteriuria**
- some bacteria: Enterobacter, Citrobacter, E. Coli, Klebsiella and Proteus **reduce nitrate to nitrite**
- analyze first morning urine
- we can detect: positive (any degree of uniform pink color) or negative

NITRITE

- **false-positive results**
 - urine stands too long before testing
 - red urine

NITRITE

- **false-negative results**

a negative test should never be interpreted as indicating the absence of bacterial infection!!!

Why?

- presence of pathogens in the urine that do not form nitrite
- the urine was not in the bladder long enough
- the urine does not contain any nitrate
- the bacterial enzymes may have reduced nitrate to nitrite and then converted nitrite to nitrogen

- urobilinogen, ascorbic acid, acidic urine

GLUCOSE

- the presence of glucose in the urine – **glycosuria – glucosuria**
- **causes:**
 - inability of kidney to reabsorb filtered glucose despite normal plasma glucose levels – **renal glycosuria**
 - urinary spillage because of abnormally high plasma glucose concentration
- **two types of test:**
 - with the enzyme glucose oxidase - specific for glucose
 - copper reduction test – detects any reducing substance
- we can detect: negative, 1+, 2+, 3+, 4+ - be aware with interpretation !!

GLUCOSE

- **false-positive results:** not likely
 - a reading taken after the prescribed time
 - urine contamination with strong oxidizing cleaning agents

GLUCOSE

- **false-negative results**
 - sensitivity for glucose may be affected by temperature, specific gravity and pH
 - ascorbic acid: ingestion, parenteral administration of vitamin C or antibiotics that contain ascorbic acid as a stabilizing agent (tetracycline)
 - moderately high ketone levels may reduce sensitivity

KETONES

- **ketones or ketone bodies** – formed during the catabolism of fatty acids
- acetoacetic acid, β -hydroxybutyric acid and acetone
- some reagents strips are sensitive only to acetoacetic acid, some also detect acetone, none detect β -hydroxybutyric acid
- normal no ketones in the urine, except in starvation, extreme exercise, diabetic ketoacidosis

- we can detect: neg, 1+, 2+, 3+

KETONES

- **false-positive results**

- highly pigmented urine specimen
- large amounts of levodopa metabolites
- specimens with high specific gravity and a low pH

- **false-negative results**

- delayed analysis of urine

PIGMENTS - BILIRUBIN

- **bilirubin:** no detectable amount of bilirubin in urine
- **high levels of bilirubin in plasma:**
 - ↑ unconjugated bilirubin → no bilirubin in the urine
 - ↑ conjugated bilirubin → bilirubin in the urine (**early detection!**)
- **bilirubin** - light sensitive – protect the urine from the light, quickly examination
- **bilirubin** - yellow brown → **biliverdin** – green

PIGMENTS - BILIRUBIN

- we can detect: neg, 1+, 2+, 3+
- **false-positive results:**
 - chlorpromazine, metabolites of some drugs
- **false-negative results:**
 - ascorbic acid
 - high levels of nitrite
 - exposure to room temperature and light

PIGMENTS - UROBILINOGEN

- **urobilinogen:** a very small amount of urobilinogen is excreted by the kidney into the urine
- **altered levels of urobilinogen in urine:**
 - liver disease
 - antibiotics (alter normal bacterial flora)
 - intestinal obstruction
- **instability:** urobilinogen → urobilin

PIGMENTS - UROBILINOGEN

- most dipsticks show block(s) for normal levels and blocks for elevated levels
- we can detect: norm, 1+, 2+, 3+, 4+
- **false-positive results:**
 - substances that react with urobilinogen test pad (sulfonamides,..)
 - urine from patients receiving phenzopyridine
- **false- negative results:**
 - urine containing nitrite or those preserved with formalin
 - improperly stored samples

HEMOGLOBIN - ERYTHROCYTES

- **heme** – acts like pseudoperoxidase and reacts with peroxide and a chromogen on the test pad
- positive results in **hematuria**, presence of free **hemoglobin** (intravascular hemolysis - hemoglobinemia) **or myoglobin**
- the diagnosis of hematuria → **conformation with microscopy (be aware in high pH and low SG)**
- two separate color scales: for erythrocytes and hemoglobin
- we can detect: ery: neg, 1+, 2+, 3+, 4+
hb: 1+, 2+, 3+, 4+.

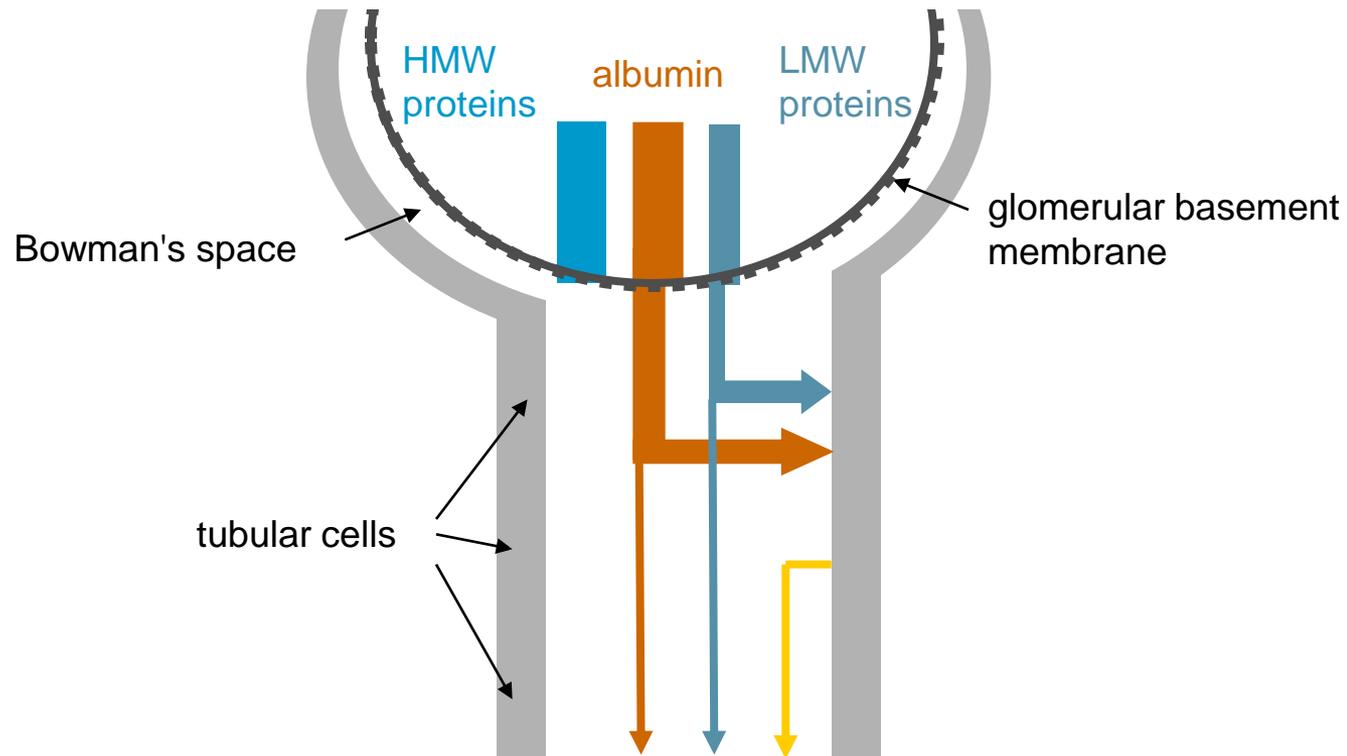
HEMOGLOBIN - ERYTHROCYTES

- **false-positive results:**
 - oxidizing contaminants (hypochlorites)
 - high bacterial content
 - semen in the urine
 - menstrual blood

HEMOGLOBIN - ERYTHROCYTES

- **false-negative results:** unusual → negative dipstick for heme theoretically excludes hematuria
- the urine is not mixed well
- ascorbic acid

PROTEINURIA < 150 mg/day



PROTEINS

- white foam – indicator of proteinuria



PROTEINS

- urine dipstick test is **sensitive to albumin – semi quantitative means of assessing albuminuria**
- albuminuria in the range of 30 – 300 mg/L in most cases cannot be detected
- detection of albuminuria more than 300 mg/L
- positive dipstick → proteinuria should be quantified
- dipstick is insensitive to non-albumin proteins (be aware of immunoglobulin light chains)

- we can detect: 0, 1+, 2+, 3+, 4+

PROTEINS

- **false-positive results:**
 - very concentrated urine
 - highly alkaline urine
 - ammonium compounds (for cleaning urine containers)
 - contamination with vaginal discharge, semen, heavy mucus, pus and blood
 - chlorhexidine gluconate, phenazopyridine,..

PROTEINS

- **false-negative results:**
 - very dilute urine
 - **proteins other than albumin!!!**

SULFOSALICYLIC ACID (SSA) TEST

- SSA detects all proteins in urine (including light chains and albumin)
- the test – mixing urine supernatant with SSA
- if becomes turbid, this indicate the presence of proteinuria

SULFOSALICYLIC ACID (SSA) TEST

0 = no turbidity (0 mg/dL)

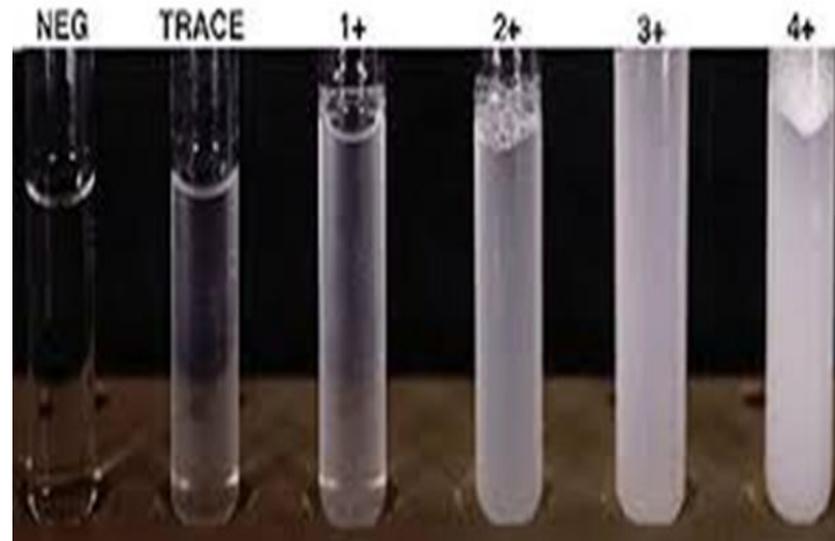
trace = slight turbidity (1 to 10 mg/dL)

1+ = turbidity through which print can be read (15 to 30 mg/dL)

2+ = white cloud without precipitate - heavy black lines can be seen (40-100 mg/dL)

3+ = white cloud with fine precipitate - heavy black lines cannot be seen (150-300 mg/dL)

4+ = flocculent precipitate – cloud is dense with large clumps (more than 500 mg/dL)



SULFOSALICYLIC ACID (SSA) TEST

- **false-positive results**

- therapy with tolbutamid, penicilin, cephalosporine, sulphonamide
- investigation with iodine radiocontrast agent (up to three days)

- **false-negative results**

- highly alkaline urine
- very diluted sample

Dipstick testing + precipitation test

albumin



all proteins



Dipstick testing ≥ 1 and precipitation test **positive** → **U-proteins/U-creatinine**

Dipstick testing **0** and precipitation test **positive** → **S-light chains?**
tubular proteinuria ?

Dipstick testing **0** and precipitation test **0**

WORKSHOP tomorrow

- URINE DIPSTICK, SULPHOSALICYLIC ACID AND SEDIMENT PREPARATION – HANDS-ON SESSION