

Clinical Laboratory Science:

Urinalysis

Urine is produced by the kidney to maintain constant plasma osmotic concentration; to regulate pH, electrolyte and fluid balances and to excrete some 50 grams of waste solids (mostly urea and sodium chloride). Texts on human anatomy and physiology describe in detail the function and mechanism by which the kidney's nephrons accomplish this.

Some normal urine constituents excreted (in g/24 hours):

Urea	25-30
Uric acid	0.6-0.7
Creatinine	1.0-1.2
Hippuric acid	0.7
Ammonia	0.7
Amino acids	3
Sodium	1-5 (NaCl 15.0)
Potassium	2-4
Calcium	0.2-0.3
Magnesium	0.1
Chloride	7
Phosphate	1.7-2.5
Sulfate	1.8-2.5

Routine urinalysis is composed of two examinations:

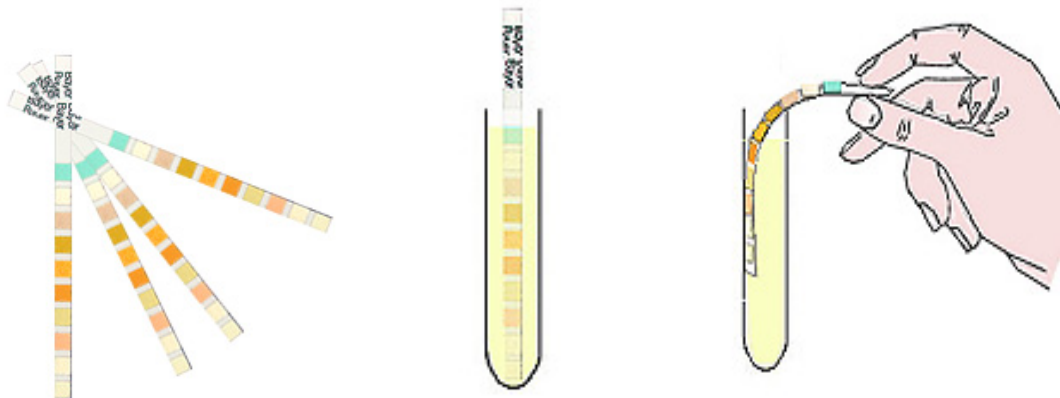
- 1) Chemical tests for abnormal chemical constituents
- 2) Microscopic exam for abnormal insoluble constituents

PROCEDURES

The color and appearance of the urine specimen is recorded. Usual colors are colorless, straw, yellow, amber; less commonly pink, red, brown. Usual appearances (opacity) are clear or hazy; less commonly turbid, cloudy and opaque, unless the specimen has remained at room or refrigerated temperatures.

CHEMICAL

The common chemical testing of urine utilizes commercial disposable test strips. Bayer's *Multistix 10 SG* test strips test for Glucose, Bilirubin, Ketone, Specific Gravity, Blood, pH, Protein, Urobilinogen, Nitrite, and Leukocyte Esterase. The result of this testing is regarded as semiquantitative.



A fresh urine specimen is collected in a clean, dry container. A Multistix strip is briefly immersed in the urine specimen, covering all reagent areas.

The edge of the Multistix strip is run against the rim of the urine container to remove excess urine. The strip is held in a horizontal

METHODOLOGIES AND INTERPRETATIONS

Glucose 30 seconds Negative g/dl (%) 1/10 (tr.) 1/4 1/2 1 >=2
mg/dl 100 250 500 1000 >=2000

Glucose:

This test is based on a double sequential enzyme reaction. One enzyme, glucose oxidase, catalyzes the formation of gluconic acid and hydrogen peroxide from the oxidation of glucose. A second enzyme, peroxidase, catalyzes the reaction of hydrogen peroxide with a potassium iodide chromogen to oxidize the chromogen to colors ranging from green to brown.

In general the presence of glucose indicates that the filtered load of glucose exceeds the maximal tubular reabsorptive capacity for glucose. In diabetes mellitus, urine testing for glucose is often substituted for blood glucose monitoring.



Bilirubin:

This test is based on the coupling of bilirubin with diazotized dichloroaniline in a strongly acid medium. The color ranges through various shades of tan.

Bilirubin in the urine indicates the presence of liver disease or biliary obstruction. Very low amounts of bilirubin can be detected in the urine, even when serum levels are below the clinical detection of jaundice.



Ketone:

This test is based on the development of colors ranging from buff-pink, for a negative reading, to purple when acetoacetic acid reacts with nitroprusside.

Urine testing only detects acetoacetic acid, not the other ketones, acetone or beta-hydroxybuteric acid. In ketoacidosis (insulin deficiency or starvation), it can be present in large amounts in the urine before any elevation in plasma levels.



Specific Gravity:

This test is based on the apparent pKa change of certain pretreated polyelectrolytes, poly(methyl-vinyl-ether/maleic anhydride), in relation to ionic concentration. In the presence of bromthymol blue, colors range deep blue-green in urine of low ionic concentration through green and yellow-green in urines of increasing ionic concentration.

The specific gravity is a convenient index of urine concentration. It measures density and is only an approximate guide to true concentration. A specific gravity of <1.010 is consistent with a concentrating defect. A specific gravity of >1.025, in the absence of protein, glucose and other large molecular weight substances such as contrast media, usually indicates normal renal concentration and makes chronic renal insufficiency unlikely.



Blood:

This test is based on the peroxidase-like activity of hemoglobin, which catalyzes the reaction of diisopropylbenzene dihydroperoxide and 3,3',5,5'-tetramethylbenzidine. The resulting color ranges from orange through green; very high levels of blood may cause the color development to continue to blue.

The presence of large numbers of RBCs in the urine sediment establishes the diagnosis of hematuria. If the dipstick is more strongly positive than would be expected from the number of RBCs, then the possibility of hemoglobinuria or myoglobinuria should be considered.



pH:

The test is based on the double indicator (methyl red/bromthymol blue) principle that gives a broad range of colors covering the entire urinary pH range. Colors range from orange through yellow and green to blue.

The urine pH should be recorded, although it is seldom of diagnostic value. Phosphates will precipitate in an alkaline urine, and uric acid will precipitate in an acidic urine.



Protein:

This test is based on the protein-error-of-indicators (tetrabromphenol blue) principle. At a constant pH, the development of any green color is due to the presence of protein. Colors range from yellow for negative through yellow-green and green to green-blue for positive reactions.

Heavy proteinuria usually represents an abnormality in the glomerular filtration barrier. The test is more sensitive for albumin than for globulins or hemoglobin.



Urobilinogen:

This test is based on the modified Ehrlich reaction, in which para-diethylaminobenzaldehyde in conjunction with a color enhancer reacts with urobilinogen in a strongly acid medium to produce a pink-red color.

Urine urobilinogen is increased in any condition that causes an increase in production or retention of bilirubin.

Nitrite
60 seconds

Negative		Positive		Positive		(Any degree of uniform pink colour is positive)
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Nitrite:

This test depends upon the conversion of nitrate (derived from the diet) to nitrite by the action of Gram negative bacteria in the urine. At the acid pH of the reagent area, nitrite in the urine reacts with para-arsanilic acid to form a diazonium compound. This diazonium compound in turn couples with 1,2,3,4-tetrahydrobenzo(h)quinoline-3-ol to produce a pink color.

Bacteriuria caused by some Gram negative bacteria which produce the nitrate reductase enzyme give a positive test.

Leukocytes
2 minutes

Negative		trace		small +		mod. ++		Large +++	
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Leukocytes:

Granulocytic leukocytes contain esterases that catalize the hydrolysis of the derivatized pyrrole amino acid ester to liberate 3-hydroxy-5-phenyl pyrrole. This pyrrole then reacts with a diazonium salt to produce a purple product.

A positive leukocyte esterase test provides indirect evidence for the presence of bacteriuria.