

Urinalysis

PLEASE NOTE: The actual urinalysis goes rather quickly. You may want to skip directly to the "Lab Exercise", collect data, and read the Introduction outside of lab.

I. Introduction

This document provides instruction for performing a chemical analysis using a urine dipstick. The testing may consist of any or all of the following: pH, protein (as a semi-quantitative analysis), glucose, bilirubin, ketones, hemoglobin (blood), urobilinogen, nitrite, leukocytes (leukocyte esterase) and specific gravity.

These test require an order from primary care provider (Physician, Nurse Practitioner, Physician Assistant). At the discretion of the provider, test results where clinical indication or departmental protocol suggest confirmatory testing, the urine specimen may be sent to the laboratory.

Urine test depend on these 7 principles:

- Urine is a physiological fluid of widely varying composition formed by the kidney from the blood. The principle constituent of urine is water (92 -99% in normal urine).
- Thousands of compounds have been identified in normal urine and the vast majority are derived from the blood. The major urine components are creatinine, urea, uric acid, sodium, potassium, chloride, calcium, magnesium, phosphates, sulfates, and ammonia.
- A considerable portion of the urine solids is waste products which have been filtered through the kidneys and are being excreted from the body. Some of the constituents are considered to be regulatory substances, which may be either retained or excreted by the kidney. Urine is a principle pathway for excretion of body waste and for homeostatic regulations of body water and body electrolytes.
- A fresh early morning specimen after overnight fasting gives the most valuable information concerning renal function and Genitourinary disease. Therefore, whenever feasible, collect specimen routinely in early A.M.
- A mid-stream collection enhances the stability of the specimen.
- A randomly voided specimen may also be tested.
- The urine specimen should be examined within 1 hour of collection, or it should be refrigerated.

- If urine is unusually dark (brown or blackish), send laboratory for testing. Most likely specimen contains one or more of the following metabolites: myoglobin, bile porphyrins, homogentisic acid. This should be brought to the attention of the attending physician.

The chemical principles of the reagent pads on the strips are (do not memorize the colors):

1. pH - this test is based on a double indicator principle that gives a broad range of colors covering the urinary pH range from 5 to 9. Colors range from orange through yellow and green to blue.
2. Protein - This test is based on the protein-error-of-indicators principle. At a constant buffered 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.
3. Glucose - This test is based on a double sequential enzyme reaction to colors ranging from green to brown
4. 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.
5. Bilirubin - This test is based on the coupling of bilirubin with diazotized dichloraniline in a strongly acid medium. The color ranges through various shades of tan.
6. Blood - This test is based on the peroxidase-like activity of hemoglobin with resulting color ranges from orange through green to dark blue.
7. Urobilinogen - This test is based on the Ehrlich reaction in which p-diethylaminobenzaldehyde reacts with urobilinogen in a strongly acid medium to produce a pink-red color.
8. 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 to produce a pink color.
9. Leukocytes – Granulocytic leukocytes contain esterases that produce a purple product.
10. Specific gravity-- In the presence of cations, protons are released by a complexing agent in the test and produce a color change of the indicator, (bromthymol blue) from blue to yellow.

II. Lab Exercises

A. Using the strips

Take a strip from each jar. The following is a list of what we might have in the lab:

(NOTE: as of the writing of this document, all we have available is Chemstrips 6, 7, 8, and 9 with SG. Ignore any that we do not have in the list below)

1. ChemStrip UGK strip for Diabetes Testing, MM # 32141, (glucose and ketones)
2. ChemStrip 4OB strip, Fisher, Cat. No. BC-417144, (blood, glucose, leukocytes, and protein)
3. ChemStrip 7 strip, MM# 32150, (Blood, Nitrite, Glucose, Ketones, Leukocytes, Protein, pH)
4. ChemStrip 9 strip, Fisher, Cat. No. BC-417109, (Blood, Bilirubin, Glucose, Ketones, Leukocytes, Protein, pH, Nitrite, Urobilinogen)
5. ChemStrip 10 strip, MM# 32230, (Blood, Bilirubin, Glucose, Ketones, Leukocytes, Protein, pH, Nitrite, Urobilinogen, Specific Gravity)
6. Multistix 10SG, PSS# 2161 (Multistix 9 plus specific gravity)

- Careful attention must be paid to time limits set for each reagent pad.
- Be aware that darkly colored urines may have substances that interfere with reading color test pads. Avoid testing of samples heavily pigmented by pyridium drugs.

Follow these steps:

1. Thoroughly mix urine specimens by inverting 10X.
2. Remove one strip from the vial and replace cap.
3. Hold the strip against vial to observe proper reading format.
4. Completely immerse reagent areas of the strip in the urine specimen and remove immediately. Start the timer and touch (blot) the edge of the strip on an absorbent material to remove the excess urine. This prevents the 'run-off' phenomenon which can lead to erroneous or inaccurate results.
5. Hold the strip in a horizontal position to prevent possible mixing of chemicals from adjacent reagent areas and/or contaminating the hands with urine.
6. Compare reagent areas to corresponding Color Chart on the bottle label at the time

specified. HOLD STRIP CLOSE TO COLOR BLOCKS AND MATCH CAREFULLY. Avoid laying the strip directly on the Color Chart as this will result in the urine soiling the chart.

7. After dipping the strip, check the pH area. If the color on the pad is not uniform, read the pH reagent area immediately, comparing the darkest color to the appropriate Color Chart.

8. Refer to specific manufacturer's instructions on package insert or on vial for permitted reading time.

A positive reaction (small or greater) at or less than 2 minutes on the leukocyte test may be regarded as a positive indication of leukocytes in urine.

Color changes that occur after 2 minutes are of no diagnostic value.

9. Record results on patient chart (Analysis Sheet below).

10. Discard used reagent strip. Reseal specimen & set aside for possible further testing.

CAUTION doing the pH test:: If proper procedure is not followed and excess urine remains on the strip, a phenomenon known as "runover" may occur in which the acid buffer from the protein reagent area will run onto the pH area, causing a false lowering in the pH result.

ANOTHER CAUTION: Urobilinogen: The absence of urobilinogen cannot be determined with this test. For best results, samples should be at room temperature when tested since reactivity is influenced by specimen temperature.

B. Microscopic Exam

There is a chart in lab showing common bodies found in a urine specimen.

Red Blood Cells

Hematuria is the presence of abnormal numbers of red cells in urine due to: glomerular damage, tumors which erode the urinary tract anywhere along its length, kidney trauma, urinary tract stones, etc. This will be confirmed with the urinalysis sticks.

White Blood Cells

Pyuria refers to the presence of abnormal numbers of leukocytes that may appear with infection in either the upper or lower urinary tract or with acute glomerulonephritis.

Usually, the WBC's are granulocytes. White cells from the vagina, especially in the presence of vaginal and cervical infections, or the external urethral meatus in men and women may contaminate the urine.

Casts

LOTS of substances can clump together or crystalize and form "casts" in the urine. Urinary casts are formed only in the distal convoluted tubule (DCT) or the collecting duct (distal nephron). The proximal convoluted tubule (PCT) and loop of Henle are not locations for cast formation.

An absence of casts does not rule out renal disease. Casts may be absent or very few in cases of chronic, progressive, generalized nephritis. Even in cases of acute renal disease, casts can be few or absent in a single sample since they tend to be shed intermittently. Furthermore, casts are unstable in urine and are prone to dissolution with time, especially in dilute and/or alkaline urine.

Although the presence of numerous casts is solid evidence of generalized (usually acute) renal disease, it is not a reliable indicator of prognosis. If the underlying cause can be removed or diminished, regeneration of renal tubular epithelium can occur (provided the basement membrane remains intact).

Look at these cast on the wall charts:

RBC: The presence of this red blood cell cast in on urine microscopic analysis suggests a glomerular or renal tubular injury.

WBC: Pus in the urine. Possibly from a reproductive tract infection in women.

Hyaline or Protein casts:

The factors which favor protein cast formation are low flow rate, high salt concentration, and low pH, all of which favor protein denaturation and precipitation, particularly that of the Tamm-Horsfall protein. Protein casts with long, thin tails formed at the junction of Henle's loop and the distal convoluted tubule are called cylindroids. Hyaline casts can be seen even in healthy patients.

DATA/ANALYSIS SHEET

Name _____

Format **CHEMSTRIP TestStrip ONLY** (Ignore Multistix)

Results: There are 5 data sheets for 5 samples. Then, do the questions in the ANALYSIS section.

HOW TO READ THE "Specific Gravity" reader:

The reader has 3 graphs...the 1st 2 on the left are not to be used. Just use the one that says "UG". It reads sort of awkwardly:

1.0
-10
-20
-30

So..if the reading goes to "-20", that reads as 1.020 (added together).

SAMPLE 1

1. Color: See Strip Jar for chart

Colorless	pale yellow	yellow	amber	red	brown
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2. Turbidity: Clear slightly hazy hazy slightly cloudy cloudy

3. Glucose (mg/dL)

Chemstrip:	Normal	50	100	250	500	1000
Multistix:	Negative	100	250	500	1000	≥2000

4. Bilirubin

Chemstrip:	Negative	1+	2+	3+
Multistix:	Negative	1+	2+	3+

5. Ketone (mg/dL)

Chemstrip:	Negative	small	moderate	large		
Multistix:	Negative	5	15	40	80	160

6. Blood

BloodChemstrip:	Negative	10	50	250	
Multistix:	Negative	Trace	1+	2+	3+

7. pH

Chemstrip: Normal Range: 5 – 9 _____

Multistix: Normal Range: 5 - 8.5 _____

8. Protein (mg/dL)

Chemstrip:	Negative	trace	30	100	500	
Multistix:	Negative	trace	30	100	300	2000

9. Uro-bilinogen

Chemstrip:	normal	1	4	8	12
Multistix:	0.2	1.0	2.0	4.0	8.0

10. Nitrite: Negative Positive

11. Leukocytes

Chemstrip:	Negative	trace	1+	2+	
Multistix:	Negative	trace	1+	2+	3+

12. Specific gravity

Chemstrip:	1.00	1.005	1.010	1.015	1.020	1.025	1.030
Multistix:	1.00	1.005	1.010	1.015	1.020	1.025	1.030

SAMPLE 2

1. Color: See Strip Jar for chart

Colorless	pale yellow	yellow	amber	red	brown
-----------	-------------	--------	-------	-----	-------

2. Turbidity: Clear slightly hazy hazy slightly cloudy cloudy

3. Glucose (mg/dL)

Chemstrip:	Normal	50	100	250	500	1000
Multistix:	Negative	100	250	500	1000	≥2000

4. Bilirubin

Chemstrip:	Negative	1+	2+	3+
Multistix:	Negative	1+	2+	3+

5. Ketone (mg/dL)

Chemstrip:	Negative	small	moderate	large	80	160
Multistix:	Negative	5	15	40		

6. Blood

BloodChemstrip:	Negative	10	50	250	
Multistix:	Negative	Trace	1+	2+	3+

7. pH

Chemstrip: Normal Range: 5 – 9 _____

Multistix: Normal Range: 5 - 8.5 _____

8. Protein (mg/dL)

Chemstrip:	Negative	trace	30	100	500	2000
Multistix:	Negative	trace	30	100	300	

9. Uro-bilinogen

Chemstrip:	normal	1	4	8	12
Multistix:	0.2	1.0	2.0	4.0	8.0

10. Nitrite: Negative Positive

11. Leukocytes

Chemstrip:	Negative	trace	1+	2+	3+
Multistix:	Negative	trace	1+	2+	

12. Specific gravity

Chemstrip:	1.00	1.005	1.010	1.015	1.020	1.025	1.030
Multistix:	1.00	1.005	1.010	1.015	1.020	1.025	

SAMPLE 3

1. Color: See Strip Jar for chart

Colorless	pale yellow	yellow	amber	red	brown
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2. Turbidity: Clear slightly hazy hazy slightly cloudy cloudy

3. Glucose (mg/dL)

Chemstrip:	Normal	50	100	250	500	1000
Multistix:	Negative	100	250	500	1000	≥2000

4. Bilirubin

Chemstrip:	Negative	1+	2+	3+
Multistix:	Negative	1+	2+	3+

5. Ketone (mg/dL)

Chemstrip:	Negative	small	moderate	large	80	160
Multistix:	Negative	5	15	40		

6. Blood

BloodChemstrip:	Negative	10	50	250	
Multistix:	Negative	Trace	1+	2+	3+

7. pH

Chemstrip: Normal Range: 5 – 9 _____

Multistix: Normal Range: 5 - 8.5 _____

8. Protein (mg/dL)

Chemstrip:	Negative	trace	30	100	500	
Multistix:	Negative	trace	30	100	300	2000

9. Uro-bilinogen

Chemstrip:	normal	1	4	8	12
Multistix:	0.2	1.0	2.0	4.0	8.0

10. Nitrite: Negative Positive

11. Leukocytes

Chemstrip:	Negative	trace	1+	2+	
Multistix:	Negative	trace	1+	2+	3+

12. Specific gravity

Chemstrip:	1.00	1.005	1.010	1.015	1.020	1.025	1.030
Multistix:	1.00	1.005	1.010	1.015	1.020	1.025	1.030

SAMPLE 4

1. Color: See Strip Jar for chart

Colorless	pale yellow	yellow	amber	red	brown
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2. Turbidity: Clear slightly hazy hazy slightly cloudy cloudy

3. Glucose (mg/dL)

Chemstrip:	Normal	50	100	250	500	1000
Multistix:	Negative	100	250	500	1000	≥2000

4. Bilirubin

Chemstrip:	Negative	1+	2+	3+
Multistix:	Negative	1+	2+	3+

5. Ketone (mg/dL)

Chemstrip:	Negative	small	moderate	large		
Multistix:	Negative	5	15	40	80	160

6. Blood

BloodChemstrip:	Negative	10	50	250	
Multistix:	Negative	Trace	1+	2+	3+

7. pH

Chemstrip: Normal Range: 5 – 9 _____

Multistix: Normal Range: 5 - 8.5 _____

8. Protein (mg/dL)

Chemstrip:	Negative	trace	30	100	500	
Multistix:	Negative	trace	30	100	300	2000

9. Uro-bilinogen

Chemstrip:	normal	1	4	8	12
Multistix:	0.2	1.0	2.0	4.0	8.0

10. Nitrite: Negative Positive

11. Leukocytes

Chemstrip:	Negative	trace	1+	2+	
Multistix:	Negative	trace	1+	2+	3+

12. Specific gravity

Chemstrip:	1.00	1.005	1.010	1.015	1.020	1.025	1.030
Multistix:	1.00	1.005	1.010	1.015	1.020	1.025	1.030

SAMPLE 5

1. Color: See Strip Jar for chart

Colorless pale yellow yellow amber red brown

2. Turbidity: Clear slightly hazy hazy slightly cloudy cloudy

3. Glucose (mg/dL)

Chemstrip:	Normal	50	100	250	500	1000
Multistix:	Negative	100	250	500	1000	≥2000

4. Bilirubin

Chemstrip:	Negative	1+	2+	3+
Multistix:	Negative	1+	2+	3+

5. Ketone (mg/dL)

Chemstrip:	Negative	small	moderate	large		
Multistix:	Negative	5	15	40	80	160

6. Blood

BloodChemstrip:	Negative	10	50	250	
Multistix:	Negative	Trace	1+	2+	3+

7. pH

Chemstrip: Normal Range: 5 – 9 _____

Multistix: Normal Range: 5 - 8.5 _____

8. Protein (mg/dL)

Chemstrip:	Negative	trace	30	100	500	
Multistix:	Negative	trace	30	100	300	2000

9. Uro-bilinogen

Chemstrip:	normal	1	4	8	12
Multistix:	0.2	1.0	2.0	4.0	8.0

10. Nitrite: Negative Positive

11. Leukocytes

Chemstrip:	Negative	trace	1+	2+	
Multistix:	Negative	trace	1+	2+	3+

12. Specific gravity

Chemstrip:	1.00	1.005	1.010	1.015	1.020	1.025	1.030
Multistix:	1.00	1.005	1.010	1.015	1.020	1.025	1.030

Analysis

Findings in Disease:

Answer the questions below. There is a sheet with each strip jar that describes what might be expected with certain readings. Also, I have added some notes of my own:

Glucose: Diabetes mellitus is the chief cause of glucosuria. The sugar content of a diabetic urine may reach as high as 10%, but 2 - 5% values are more commonly found.

Bilirubin: Bilirubin may often appear in the urine before other signs of liver dysfunction are apparent.

Blood:

Hemoglobinuria may indicate a hematologic disorder such as haemolytic anemia, hemolytic transfusion reaction, etc. In these, the RBCs are destroyed and release their hemoglobin.

It is also found in poisonings with strong acids or mushrooms, following burns, or in renal infection. A significant amount of free hemoglobin is found in the urine whenever red blood cells are present in excessive numbers.

Another cause may be bleeding in the urinary tract.

Protein: Please Note: the test strips are giving their readings in mg; 500 mg = .5 grams

1. Marked proteinuria (more than 4 gm per day) is typical of the several renal disease: nephrotic syndrome, glomerulonephritis, nephrosclerosis, amyloid disease, systemic lupus erythematosus, and severe venous congestion of the kidney produced by renal vein thrombosis, congestive heart failure, or constrictive pericarditis.

2. Moderate proteinuria (0.5 to 4.0 gm per day) is found in the vast majority of renal diseases (see above) as well as preeclampsia and any inflammatory, malignant, degenerative, or irritative condition of the lower urinary tract, including the presence of calculi.

3. Minimal proteinuria (less than 0.5 gm per day) is associated with chronic glomerulonephritis, renal tubular disorders and various disorders of the lower urinary tract leading to bleeding.

Urobilinogen:

Urinary bilinogen is increased by any condition that causes an increase in the production of bilirubin (that is, increased destruction of RBCs: hemolytic anemia, pernicious anemia, and malaria are examples) or by any disease that prevents the liver from normally removing the reabsorbed urobilinogen from the portal circulation (infectious hepatitis, toxic

hepatitis, portal cirrhosis, congestive heart failure, and infectious mononucleosis being examples).

1. Describe what diseases, disorders or conditions you expect with each sample:

2. Name all the substances we tested for:

3. Describe, in your own words, what "runover" is. Why would the pH be lowerd?

4. Would you expect a lot of lipid-soluble substance in urine? Why or why not?

5. When should a urine analysis be performed for optimal conditions? Which other conditions are optimal?

6. Draw a hyaline, RBC, and WBC cast in the space below: