Urinalysis Review

2016 ASCLS-MO/CLMA Spring Meeting

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Objectives

• Demonstrate an understanding of concepts in urinalysis by answering questions and/or discussing the theory or principles of urinalysis testing.

• Interpret urinalysis test results.

• Correlate urinalysis results with disease states.
Case #1

- 25-year-old pregnant woman comes to the outpatient clinic with symptoms of lower back pain, urinary frequency, and a burning sensation when voiding. Her pregnancy has been normal up to this point. She is given a sterile container and asked to collect a mid-stream-clean-catch urine specimen.
Her urinalysis results are as follows:

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Result</th>
<th>Interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Color</td>
<td>Pale Yellow</td>
<td></td>
</tr>
<tr>
<td>Clarity</td>
<td>Cloudy</td>
<td>Blood, Small</td>
</tr>
<tr>
<td>Sp. Gravity</td>
<td>1.005</td>
<td>Bilirubin, Negative</td>
</tr>
<tr>
<td>pH</td>
<td>8.0</td>
<td>Urobilinogen, Normal</td>
</tr>
<tr>
<td>Protein</td>
<td>Trace</td>
<td>Nitrate, Positive</td>
</tr>
<tr>
<td>Glucose</td>
<td>Negative</td>
<td>Leukocyte, 2+</td>
</tr>
<tr>
<td>RBCs</td>
<td>6-10/hpf</td>
<td>SECs, Few</td>
</tr>
<tr>
<td>WBCs</td>
<td>40-50/hpf</td>
<td></td>
</tr>
<tr>
<td>Bacteria</td>
<td>Many</td>
<td></td>
</tr>
</tbody>
</table>
What is the correlation between the color and the specific gravity?

• Because urochrome is excreted at a constant rate, the intensity of color in a fresh urine sample can give a rough estimate of urine concentration. A dilute urine will be pale yellow and a concentrated urine will be dark yellow.
Specific Gravity

- A measure of dissolved substances present in solution.

- Measurement that helps to assess the kidney’s concentrating ability.

- Expressed as a numerical value reported to the third decimal place – it has NO associated units.
Specific Gravity

- Water serves as the “control”
  - Specific gravity: 1.000

- Protein-free filtrate
  - Specific gravity: 1.010

- Plasma
  - Specific gravity: 1.010
Specific Gravity

• Specific gravity of normal urine: 1.003-1.035

• Hyposthenuria
  – ≤ 1.010
  – Dilute urine
  – Hypotonic

• Hypersthenuria
  – ≥1.010
  – Concentrated urine
  – Hypertonic

• Isosthenuria
  – Fixed at 1.010 (same as plasma)
  – Renal tubules lost absorption and secreting capability
Urine Color

• Varies from colorless to almost black
  – Metabolic functions, physical activities, food/drink, pathologic conditions.
Urine Color

- Normal Urine Color
  - Straw to dark yellow
Urine Color

Light Yellow  Yellow  Amber  Red  Brown
Urine Clarity

- What is the significance of the urine’s clarity?

http://uoitclinicalbiochemistry.weebly.com/urinalysis.html
Urine Clarity

• Clear
  – No particulate matter
  – Normal urine and dilute samples
  – Can read newsprint through it

– Suggests that minimal, if any, formed elements (cells, crystals, cast) will be present upon microscopic examination of the specimen
Urine Clarity

- Slightly Cloudy/Hazy
  - Suggests the presence of small amounts of formed elements present upon microscopic examination of the specimen
  - Defined as visible particulate matter present
  - Newspaper NOT distorted or obscured when viewed through the urine
  - Indicates presence of mucus, crystals, bacteria, pus (white blood cells), other cells, fat, casts..
Urine Clarity

• Cloudy
  – Suggests the presence of numerous amounts of formed elements upon microscopic examination of the specimen
  – Defined as newsprint can be seen through the urine but the letters are distorted or blurry
  – Indications- same as for slightly cloudy/hazy
Urine Clarity

- **Turbid**
  - Suggests the presence of "loads" of formed elements upon microscopic examination of the specimen
  - Defined as unable to see newsprint through the urine
  - Indications - same as for hazy
Urine Clarity

• Milky
  – May indicate the presence of fats or chyle (emulsified fats and lymph)

http://www.physio-pedia.com/Filariasis
Urine Clarity

• Brick Dust
  – Suggests the presence of amorphous urates or uric acid crystals upon microscopic examination of the urine

  – Sometimes seen in newborns. Usually go away on their own.
Urine Clarity

• Non-pathologic Turbidity:
  – Women: SECs (squamous epithelial cells), mucus vaginal creams
  – Talcum powder
  – Semen
  – Radiographic contrast media

  – Refrigeration: precipitation of amorphous solids
    • Amorphous phosphates (white precipitate in alkaline pH)
    • Amorphous urates ("brick dust" precipitate in acidic pH)

http://library.med.utah.edu/WebPath/TUTORIAL/URINE/URIN066.html

https://quizlet.com/31381272/microscopic-ua-flash-cards/
Urine Clarity

• Pathologic Turbidity:
  – RBCs, WBCs, bacteria, yeast
    – High amounts of non-squamous epithelial cells
  – Abnormal crystals
  – Lipids
• What is the significance of the positive nitrate result?
Nitrite: Description and Purpose

• Results from conversion of nitrate to nitrite.

• Some bacteria have the ability to convert nitrate to nitrite.

• Dietary nitrate is usually excreted in some form.
Nitrite: Clinical Significance

• Rapid screening test for a UTI
  – Detects initial bladder infection (cystitis).
  • Often have vague symptoms at that time
  • If untreated, can lead to renal tissue damage
• Examples of nitrate converting bacteria (note these are all gram negative bacilli):
  – E. coli
  – Klebsiella
  – Proteus
  – Enterobacter
  – Citrobacter

• Gram positive organisms do not typically convert nitrate to nitrite
• There are three (3) criteria necessary for a bacterial infection with a positive nitrite bacteria to occur:
  – 1. Bacteria must be present & capable of making the conversion
  – 2. Bacteria must have the opportunity to live in the bladder at least 4 hours
  – 3. A minimal amount of nitrate must be present
Nitrite: Results

• Normal: negative
• Any shade of pink is positive
  – Should correlate with a positive leukocyte esterase test.

• Note:
  – A negative reaction does NOT r/o a UTI (remember, only select bacteria make the conversion)
Nitrite: Interfering Substances

• False Positives:
  – Bacterial contamination (improperly preserved specimens)
  – Any substance/medication that contains azo-compound
  – Any substance that colors the urine red in an acid medium
• False Negatives:
  – Nonreductase-containing bacteria
  – In sufficient contact time between bacteria and urinary nitrate
  – Lack of urinary nitrate
  – Large #s of bacteria converting nitrite to nitrogen
  – Antibiotics
  – High concentrations of ascorbic acid (≥ 30 mg/dL)
Nitrite: Confirmatory Tests

• Bacterial culture and sensitivity to grow and identify causative microbial (bacterial) agent
What is the significance of the positive leukocyte esterase?
Leukocyte Esterase: Description and Purpose

• Leukocytes (white blood cells) assist the body in fighting off infections.

• The enzyme leukocyte esterase is found in the primary granules of select types of white cells.

• Does not give a quantified result
  – Quantitation is done by microscopic examination
Leukocyte Esterase: Clinical Significance

• Associated with bacterial and nonbacterial infections

• Positive test is mostly associated with bacterial infection, however, infections caused by *Trichomonas, Chlamydia*, and yeast cause leukocyturia without bacteria.
• When cells lyse, the leukocyte esterase enzyme is present and can be detectable.

• Therefore, it is not uncommon to have a positive dipstick result and little or no intact white cells on the microscopic.
Leukocyte Esterase: Results

• Test is specific for leukocyte esterase

• Normal: Negative
Leukocyte Esterase: Confirmatory Tests

- Microscopic examination for intact white blood cells (WBCs)
- Gram Stain for WBCs
- Culture and sensitivity

What is the most probable diagnosis for this patient?

- UTI

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<th><strong>Urobilinogen</strong></th>
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<th><strong>Glucose</strong></th>
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Development of UTIs:

- Usually result of bacterial (or yeast) infection
- Often bacteria involved are from the intestinal tract & become introduced into the urinary tract.
Lower UTIs:

- When it involves the urethra = urethritis
- When it involves the bladder = cystitis

– Since these infections do NOT involve the kidneys (specifically the renal tubules), casts are NOT present

– Urinalysis Findings:
  • Positive LE
  • Positive Nitrite?
  • Increased pH
  • Increased protein
  • Bacteria
  • WBCs
  • Microscopic hematuria
– Upper UTIs:
  • When it involves the renal pelvis = pyelitis

http://pmgbiology.com/2015/02/20/kidney-part-i-a-understanding-of-kidneys-role-in-excretion/
– When it involves the renal pelvis & interstitium = pyelonephritis
  • Can be acute or chronic
    – Acute:
      » bacteria from lower UTI moves up the urinary tract
      » Renal calculi, pregnancy, and reflux of urine from bladder into ureters
      » Rapid onset of symptoms

  » Urinalysis findings:
    • Leukocytes
    • Bactiuria
    • **WBC casts**
    • Bacterial casts
    • Microscopic hematuria
    • proteinuria
Case Study #2

An eight-year-old boy was taken to the emergency room (ER) after his mother noticed that his urine was red. The physician on duty ordered a battery of tests including a urinalysis (UA), chemistry profile & a complete blood count (CBC). Upon further questioning Jack’s mom revealed that the boy had a sore throat recently but other than that was an overall healthy child.
The physical & chemical exam results of the UA were as follows:

<table>
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<tr>
<th>TEST</th>
<th>RESULT</th>
<th>REFERENCE RANGE</th>
</tr>
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<tbody>
<tr>
<td><strong>PHYSICAL EXAM</strong></td>
<td>******************</td>
<td>******************</td>
</tr>
<tr>
<td>Color</td>
<td>Red</td>
<td>Yellow</td>
</tr>
<tr>
<td>Appearance (Clarity)</td>
<td>Cloudy</td>
<td>Clear</td>
</tr>
<tr>
<td><strong>CHEMICAL DIPSTICK EXAM</strong></td>
<td>******************</td>
<td>******************</td>
</tr>
<tr>
<td>Glucose</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>Bilirubin</td>
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<td>Negative</td>
</tr>
<tr>
<td>Ketone</td>
<td>Negative</td>
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</tr>
<tr>
<td>Specific Gravity</td>
<td>1.040</td>
<td>1.003-1.030</td>
</tr>
<tr>
<td>Blood</td>
<td>Moderate</td>
<td>Negative</td>
</tr>
<tr>
<td>pH</td>
<td>7.5</td>
<td>5.0-8.0</td>
</tr>
<tr>
<td>Protein</td>
<td>2+ (100 mg/dL)</td>
<td>Negative-Trace</td>
</tr>
<tr>
<td>Urobilinogen</td>
<td>0.2 mg/dL</td>
<td>0.2-1.0 mg/dL</td>
</tr>
<tr>
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<td>Negative</td>
</tr>
<tr>
<td>Leukocyte Esterase</td>
<td>Negative</td>
<td>Negative</td>
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</table>
• What is the most likely explanation for the color of little Jack’s urine?
  - **Blood or other heme-derived products**
Abnormal Urine Color

• Red- Red/Brown:
  – Most likely associated with blood or heme-derived products
  – Porphyrins
    • Negative chemical test for blood
Red Urine

Clear

Hemoglobin (intravascular hemolysis)

Red Plasma

Myoglobin (muscle damage)

Clear Plasma

Cloudy

RBCs present (hematuria)

These will also have positive chemical tests for blood
Non-pathologic causes include:

- Beets, blackberries (in acidic urine)
- Rifampin (tuberculosis medication)
- Menstrual contamination (cloudy, RBCs, mucus, and clots)
• What is the most likely explanation for the clarity of Jack’s urine?
  – Formed elements. Specifically, we should expect to see red blood cells.
• What confirmatory test could be done to confirm the dipstick protein results?
  – SSA
Confirmatory Testing

- Protein dipstick of $>30$ mg/dL AND pH of $>7.0$
  - do a sulfosalicylic acid (SSA) test
SSA test

• Cold precipitation test
• Reacts with all forms of protein
• Centrifuge urine
• Add 3mL of 3% SSA reagent to 3 mL of urine
• Mix by inversion
• Observe for cloudiness
• Grade the degree of turbidity by comparing against standards
What condition is this patient most likely suffering from? Justify your answer.

- Hematuria caused by some kind of damage to the glomerulus as evidenced by the intact red blood cells and elevated protein. The patient is most likely suffering from post-streptococcal glomerulonephritis.

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</tr>
<tr>
<td>Leukocyte Esterase</td>
<td>Negative</td>
<td>Negative</td>
</tr>
</tbody>
</table>
Glomerular Diseases

- Major affected component - Glomeruli (duh!)
• Primary disease causing mechanism:
  – Immune complexes circulating in the bloodstream are deposited on glomerular membrane (IgA)
    Leads to...
  – Complement, neutrophils, lymphocytes, monocytes, cytokines are attracted to the area
    Produces...
  – Changes to and damage of the membranes
    Results in...
  – Thickening of glomerular membrane and/or damage to the capillaries
Acute Glomerulonephritis

• Acute Poststreptococcal Glomerular Nephritis:
  – Occurs after a respiratory infection caused by group A streptococcus.
  – Sudden onset of symptoms:
    • Fever
    • Edema (around the eyes)
    • Oliguria
    • Hematuria
• During infection streptococci form immune complexes with Abs that become deposited on glomerular membranes
  – Followed by inflammatory reaction that affects glomerular function
• Urinalysis findings:
  – Hematuria
  – Proteinuria
  – Oliguria
  – RBC casts
  – Dysmorphic RBCs
  – Hyaline and granular casts
  – WBCs
• Confirm with antistreptolysin O (ASO titer)
Case #3

- A 24-year old female went to the local clinic with complaints of constant fatigue & nausea. The physician ordered a battery of tests including a urinalysis (UA) [physical exam & chemical dipstick] to assess the situation.
The following results were obtained:

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</tr>
</thead>
<tbody>
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<td>PHYSICAL EXAM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Color</td>
<td>Dark yellow</td>
<td>Yellow</td>
</tr>
<tr>
<td>Appearance (Clarity)</td>
<td>Clear, yellow foam present</td>
<td>Clear</td>
</tr>
<tr>
<td>CHEMICAL DIPSTICK EXAM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glucose</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>Bilirubin</td>
<td>Moderate (2+)</td>
<td>Negative</td>
</tr>
<tr>
<td>Ketone</td>
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</tr>
<tr>
<td>Specific Gravity</td>
<td>1.015</td>
<td>1.003-1.030</td>
</tr>
<tr>
<td>Blood</td>
<td>Negative</td>
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</tr>
<tr>
<td>pH</td>
<td>6.0</td>
<td>5.0-8.0</td>
</tr>
<tr>
<td>Protein</td>
<td>Negative</td>
<td>Negative-Trace</td>
</tr>
<tr>
<td>Urobilinogen</td>
<td>4.0 mg/dL</td>
<td>0.2-1.0 mg/dL</td>
</tr>
<tr>
<td>Nitrite</td>
<td>Negative</td>
<td>Negative</td>
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<tr>
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<td>Negative</td>
<td>Negative</td>
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</tbody>
</table>
• What does the color and foam present upon inspection of this indicate?

– Bilirubin
Abnormal Urine Color

• Dark Yellow (Amber):
  – Abnormal when due to bilirubin (confirmed in chemical exam by Ictotest)
    • Suspected if there is a yellow foam when shaken
  – May also contain Hepatitis virus
Foam

- **White:**
  - Small Amount: Normal
  - Large Amount: Protein

- **Yellow:**
  - Large Amount: Bilirubin

Photo by Amanda Reed
• What two confirmation tests could be done to confirm the dipstick results? [indicate what dipstick test is being confirmed for each]
  – Ictotest for bilirubin
  – Watson-Schwartz for urobilinogen
Ictotest

- Brown Urine and/or positive dipstick bilirubin (small, moderate or large)- Ictotest

Ictotest

- Less subject to interference

- Sensitive to 0.05-0.10 mg/dL (reagent strips sensitive to 0.40 mg/dL)

- Used to detect early stages of liver disease
Ictotest

Photo by Amanda Reed
Bilirubin: Description and Purpose

• Breakdown product of hemoglobin

• In normal, healthy individuals there is such a minute amount of bilirubin present that its undetectable. (0.4-1.0 mg/dL)
Bilirubin: Clinical Significance

- Liver disease (Hepatitis, Cirrhosis)
- Biliary Obstruction (Gallstones)
- Carcinoma

- Can detect these before the development of jaundice
Bilirubin: Results

• Normal: negative
• Result Choices:
  – Trace, small, moderate, large
Urobilinogen: Description and Purpose

• 2 substances of importance with this dipstick test:
  – Urobilinogen
  – Porphobilinogen
• Urobilinogen:
  – Breakdown product of bilirubin that circulates in the blood.
  – Some gets filtered through the glomerulus, therefore, a small amount $\leq 1 \text{mg/dL}$ is normally found in the urine

• Porphobilinogen:
  – Substance involved in formation of heme
Urobilinogen: Clinical Significance

• Early detection of liver disease
• Liver disorders (hepatitis, cirrhosis, carcinoma)
• Hemolytic disorders

• 1-9% have elevated urobilinogen results due to constipation.
Urobilinogen: Results

• Normal: 0.2 – 1 mg/dL
• Result Choices:
  – 0.2, 1, 2, 4, 8 mg/dL
<table>
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<tr>
<th>Condition</th>
<th>Urine Bilirubin</th>
<th>Urine Urobilinogen</th>
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<tbody>
<tr>
<td>Bile Duct Obstruction</td>
<td>+++</td>
<td>Normal</td>
</tr>
<tr>
<td>Liver Damage</td>
<td>+ OR -</td>
<td>++</td>
</tr>
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- What condition is this patient most likely suffering from?
  - Liver problems

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• What would the diagnosis be if the urobilinogen remained at 4.0 mg/dL but the bilirubin result was negative?
  – Hemolytic disease