Urinalysis

Chemical, Physical and Microscopic Examination of Urine

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I. Introduction and Overview

> *"A urinalysis is a simple, inexpensive test that can help to detect problems in many parts of your body including your kidneys and urinary tract, your heart and your liver. A urinalysis can help to detect many diseases before symptoms occur. Early detection and treatment can often prevent serious diseases from getting worse. More than 20 million Americans – one in nine adults – have chronic kidney disease, and most don't even know it. More than 20 million others are at increased risk for chronic kidney disease. Yet, a urinalysis can detect protein in the urine – one of the earliest signs of kidney disease – years before symptoms appear and at a time when treatment can make all the difference."* -National Kidney Foundation

Urinalysis is the chemical, physical and microscopic examination of urine via procedures performed in an expeditious, reliable, accurate and safe manner. In ancient times, urine was poured on the ground and the attraction of insects to it indicated an abnormal urine sample. For centuries, the utility of urinalysis as a diagnostic tool is well known in medicine. Since the kidney is involved in maintenance of homeostasis in the body, any deviation from the normal urine composition can be of great diagnostic significance to the healthcare provider. Furthermore, it can also imply pathology.

Historically, urinalysis involved an evaluation of color, transparency (cloudiness or turbidity), odor and taste. In modern medicine, while all of these (except taste) are still evaluated, the evaluation is generally done via test strips. Specifically, the strips are interpreted by watching the various sections (pads or reagent areas) on the strip for color changes and comparing to them to the color-coded key on the bottle label. The specific gravity and pH of urine are additional parameters measured separately. In addition, a microscopic examination of the solids or sediment in urine is critical in pathological diagnosis as it may or may not reflect injury, diseases, infections, etc.

NOTE: Review urine composition and formation in the latest edition of *Hole's Anatomy and Physiology*.

Urine pH:
Normal range: 4.6-8 (average, 6)
Elevated in: Bacteriuria, vegetarian diet, renal failure with inability to form ammonia, drugs (antibiotics, sodium bicarbonate, acetazolamide)
Decreased in: Acidosis (metabolic, respiratory), drugs (ammonium chloride, methenamine mandelate), diabetes mellitus, starvation, diarrhea

II. Chemical Analysis of Urine

- Safety gloves and glasses are required for this exercise. Use only your own urine and place all glassware in a disinfectant bleach bath after use. Wash your hands before and after this exercise.
- Handling of reagent strips: Use only one reagent strip at a time. Do not touch the reagent area and hold the test strip by the plastic end only. Keep the bottle tightly closed after removing a strip.

Water from the air, light and contaminants from your hands can decompose the chemicals on the reagent areas. Check the expiration date on the bottle. In a clinical setting, never use any material or chemicals with a passed expiration date.

- Obtain 50 ml from each of the three simulated urine samples provide by Dr. Bawa and transfer them into a beaker. Label each beaker accordingly. Read the directions (if any) on the strip container. Dip a urinalysis test strip into each beaker and record your results in table A below.
- Collect your urine in a clean or sterile specimen container (paper cup) by the clean-catch method of collecting a urine sample. This method is used to prevent bacteria from the penis or vagina from getting into a urine sample. After cleaning the genital area, urinate a small amount into the toilet bowl to clear the urethra of any contaminants. Then collect the sample of urine. Remove the specimen container from the urine stream without stopping the flow.
- Using a new urine test strip, measure pH, presence or absence of glucose, albumin, blood and ketone bodies. Dip a new urinalysis test strip into the specimen container (paper cup) and record the results in the table B below.

Table A

Test	**Normal values**	**Abnormal values**	**Sample 1**	**Sample 2**	**Sample 3**
Color	Colorless	Milky			
	Straw	Brownish yellow			
	Amber	Green			
	Smoky	Brown			
Transparency	Clear	(+) Slight			
		(++) Moderate			
		(+++) Cloudy			
		(++++) Very Cloudy			
pH	(4.5-8.0; av 6.0)	<4.5, >8.0			
Protein	None to trace	see color chart			
Glucose	none	see color chart			
Ketones	none	see color chart			
Hemoglobin/Occult Blood	none	see color chart			

Table B

pH	Protein	Glucose	Ketone	Hb/Occult Blood	Color	Transparency

Color or Appearance of Urine	Possible Cause
Colorless	Highly dilute, recent fluid consumption
Pale yellow	Normal
Yellow	Normal
Amber	Normal, concentrated urine
Deep yellow	Concentrated sample, riboflavin, dehydration
Orange	Antibiotic, pyridium, bilirubin, anticoagulant
Pink	Porphyrin, myoglobin, hemoglobin, beet pigment, rhubarb
Red	Porphyrin, myoglobin, hemoglobin, beet pigment, uroerythrin
Green	Oxidized bilirubin (biliverdin), clorets, pseudomonas infections
Blue	Diagnex, methylene blue, indicant
Brown	Bilirubin, hematin, methemoglobin
Gray	Furazolidone, nitrofurantoin
Black	Melanin, homogenistic acid

Q. What compound is responsible for giving urine its color?
Q. What is the color of urine that contains bilirubin?

III. Specific Gravity Measurement – The Urinometer Method

The specific gravity of a urine sample (i.e., urine density) indicates the solute concentration in urine – it reveals how concentrated or dilute the urine is. Normal values are 1.003 to 1.035 (normal value ranges may vary slightly among laboratories).

Q. What is specific gravity?
A. It is defined as the ratio of the weight of something to the weight of an equal volume of pure water. Obviously, urine is slightly heavier than an equal volume of water because it contains dissolved solutes.

Procedure:

- Pour urine into a urinometer cylinder (75% full)
- Removed any foam at top with a blotting paper
- Float the hydrometer on top of the urine without touching the sides of the urinometer cylinder
- Position your eye to see the concave meniscus intersect the line on the hydrometer
- Do a temperature correction (if needed)
- Clean the urinometer cylinder and the hydrometer by rinsing them under running tap water and then drying it carefully using a tissue

Q. What does a specific gravity value higher than the normal range indicate? What does a specific gravity value lower than the normal range indicate? Is there any risk in an individual of concentrated urine over an extended period?

A. Increased urine specific gravity may be due to Addison's disease (rare); dehydration; diarrhea that causes dehydration; glucosuria; heart failure (due to decreased blood flow to kidneys); renal arterial stenosis; shock; syndrome of inappropriate antidiuretic hormone secretion (SIADH)
Decreased urine specific gravity may be due to Aldosteronism (very rare); Excessive fluid intake; Diabetes insipidus (central or nephrogenic); renal failure; renal tubular necrosis; severe kidney infection (pyelonephritis)

IV. Microscopic Examination – The Sedi-Stain Method

Normal urine is sterile. Urine may contain one or many "formed elements" which must be reported quantitatively. As part of a urinalysis, the urine sediment is centrifuged and examined microscopically for various crystals, casts, cells, threads, microbes (protozoa, bacteria, yeast, etc.). Examination of sediment provides a direct sampling of urinary tract morphology. In a healthy individual, urine contains some cells (e.g., epithelial cells from the kidney, ureter, bladder, and urethra) and other formed elements from the entire urinary tract. However, in renal disease, the urine often contains increased numbers of substances discharged from the kidney.

The specimen used for microscopic examination should be as fresh as possible. For example, red blood cells and many formed solids tend to disintegrate upon standing, particularly if the specimen is warm or alkaline. Additionally, microbes may grow in urine stored at room temperature.

Hematuria is defined as the abnormal presence of red blood cells (RBCs) in the urine. The American Urological Association (AUA) defines microscopic hematuria as 3 RBCs/high-power field on microscopic examination of the centrifuged urine specimen in two of three freshly voided, clean catch, midstream urine samples It may be due to: glomerular disease; tumors; kidney trauma; transfusion reaction; menstrual blood; renal infarcts; acute tubular necrosis; recent strenuous exercise; upper and lower urinary tract infections; nephrotoxins; traumatic catheterization; passage of renal stones; etc.

Casts are urinary sediments formed by coagulation of albuminous material in the kidney tubules. Their shape is generally cylindrical with varying diameter. They always indicate some form of kidney disorder.

The presence of numerous leucocytes and bacteria is characteristic of a urinary tract infection (UTI). However, since urine specimens for routine examination are not usually obtained via sterile techniques old specimens may contain many bacteria with few leukocytes. The presence of many squamous cells may indicate an external genital source for the bacteria. A positive nitrite test result may indicate a UTI but a safe diagnosis is only possible by a positive culture obtained with a midstream urine specimen. Bacteria associated with UTIs are mostly bacilli (*E. coli*).

Microscopic examination:
Red blood cells: 0-5 (high-power field)
White blood cells: 0-5 (high-power field)
Bacteria (spun specimen): absent
Casts: 0-4 hyaline (low-power field)

Procedure:

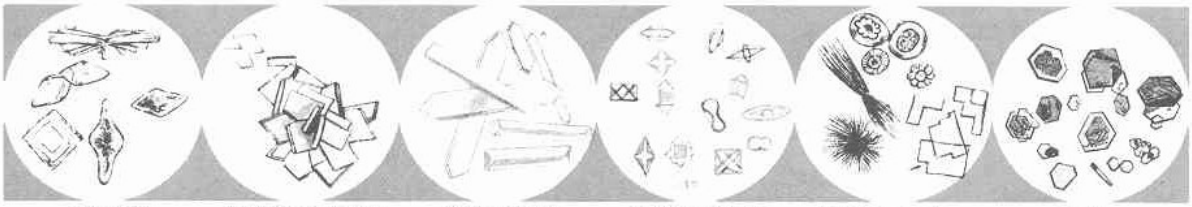
- Shake your urine sample to resuspend any sediment and pour approximately 5-10 ml into a centrifuge tube. Conical bottom test tubes are preferred as they allow for better pellet formation. Be sure the centrifuge is balanced with an even number of loaded tubes.
- Centrifuge at 1,500 rpm for 5-7 minutes (speed varies depending on the centrifuge's characteristics). You should have white sediment at the bottom of the tube. Carefully decant or aspirate off the supernatant urine carefully. Resuspend the white sediment in the residual urine that appears at the bottom of the test tube by tapping the tube against a hard surface. Add a drop of the commercial stain, Sedistain, and mix gently.
- Place a drop of this stained sediment onto a clean slide and place a coverslip on top.
- Examine under a microscope at low magnification (10X) first. Then switch to high dry magnification and then observe under oil immersion.
- Compare the microscopic structures found in your urine sediment with those in the charts provided by Dr. Bawa.
- When you have finished with all the tests, discard your urine directly down the sink and place all disposable materials in the container provided. Wash all glassware with soap and water.

V. Additional Questions

What is the correct technique for collecting and handling a urine sample for analysis?
How do you interpret the findings of a typical routine urinalysis?
Define: (a) purple urine bag syndrome; (b) blue diaper syndrome; (c) red diaper syndrome; and (d) black urine disease?
Does the odor of the urine have any diagnostic or clinical significance?
What is the significance of (a) gross hematuria and (b) or microscopic hematuria?
What is the "three tube test" for hematuria?
What is the nutcracker syndrome?
What is the significance of proteinuria?
What is a urinary tract infection (UTI) and who gets them?
What is the pathogenesis and what are the most common microbes involved in UTIs?
What are the usual symptoms of UTIs?
How do you make a laboratory diagnosis for a UTI?
How should acute uncomplicated UTI be treated? How do you prevent them?
How is a UTI in pregnancy managed?
How to control complicated UTI and how to prevent it?
What is suppressive antimicrobial therapy, and when is this appropriate?
What are the current management strategies for community-acquired UTIs?
How to control or treat UTIs in older patients who reside in nursing homes?
How to diagnose and treat fungal UTIs?

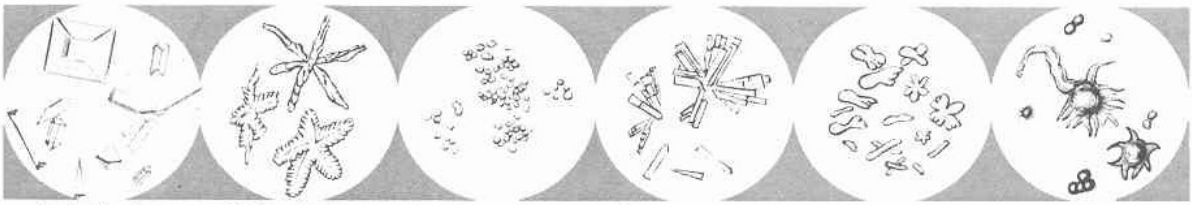
(Modified from: E. Lerma and A. Nissenson (2012). Nephrology Secrets, Mosby/Elsevier, Philadelphia, PA)

CRYSTALS FOUND IN ACID URINE 400 X



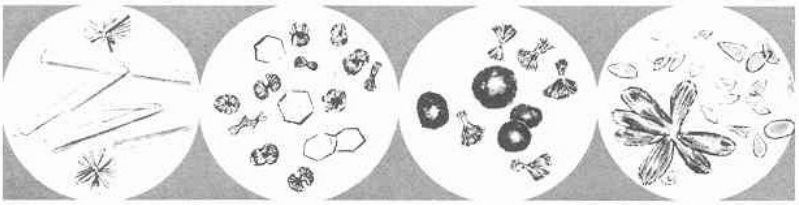
Uric acid | Amorphous urates and uric acid crystals | Hippuric acid | Calcium oxalate | Tyrosine needles Leucine spheroids Cholesterin plates | Cystine

CRYSTALS FOUND IN ALKALINE URINE 400 X



Triple phosphate Ammonium and magnesium | Triple phosphate going in solution | Amorphous phosphate | Calcium phosphate | Calcium carbonate | Ammonium urate

SULFA CRYSTALS



Sulfanilamide | Sulfathiazole | Sulfadiazine | Sulfapyridine

Ames Atlas of Urine Sediment

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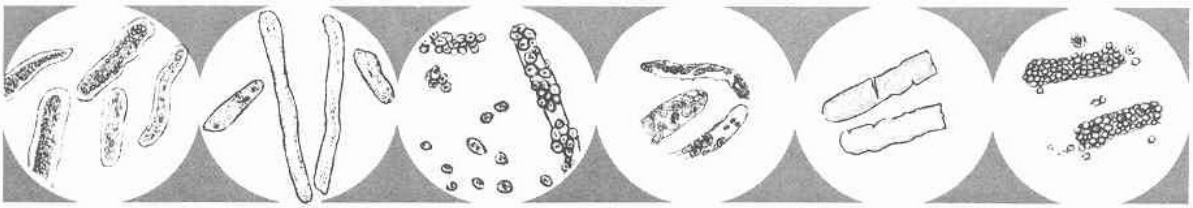
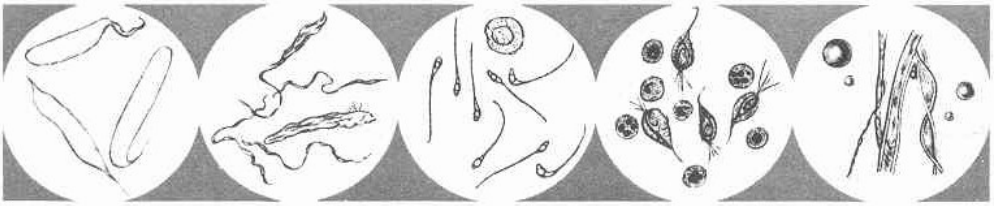
CELLS FOUND IN URINE

RBC and WBC | Renal epithelium | Caudate cells of Renal Pelvis | Urethral and bladder epithelium | Vaginal epithelium | Yeast and bacteria

CASTS AND ARTIFACTS FOUND IN URINE 400 X



Granular casts fine and coarse | Hyaline cast | Leukocyte cast | Epithelial cast | Waxy cast | Blood cast



Cylindroids | Mucous thread | Spermatozoa | Trichomonas vaginalis | Cloth fibers and bubbles