Chapter 7

MICROSCOPIC EXAMINATION OF URINE SEDIMENT

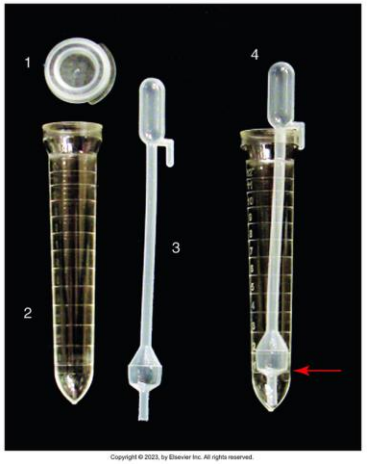
Commercial Urinalysis Systems

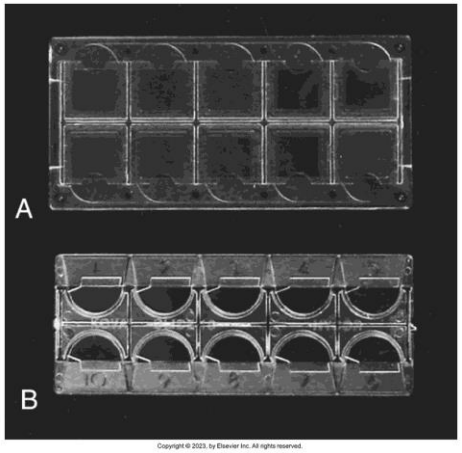
Standardizing urine microscopics by controlling techniques used to prepare and view sediment

All laboratory personnel performing urine microscopics need to adhere to established protocols

Recommended volume to centrifuge at 400 to 450 g for 5 minutes to obtain sediment is 12 mL

1 mL of sediment is obtained (in manual methods) and gently mixed to resuspend





Viewing and Reporting Formats

Laboratory procedure needs to be followed exactly

Same microscope should be used for all microscopics; if not possible, then diameters of field of view (FOV) must be identical on all

Urine components assessed using at least 10 low-power fields (lpf) or 10 high-power fields (hpf), depending on specific component

Red blood cells (RBCs), white blood cells (WBCs), and casts should be enumerated

Crystals and bacteria assessed qualitatively

Staining Techniques

Supravital stains to enhance visualization

- Most common is crystal-violet and safranin
  - Also known as Sternheimer-Malbin stain
- Toluidine blue stains various cell components differently to help in identification

Acetic acid brings out nuclear detail of WBCs and lyses RBCs

Fat stains such as Sudan III or oil red O identify fats inside cells or free floating; cholesterol does not stain and must be confirmed with polarizing microscopy

Staining Techniques (Cont.)

Gram stain

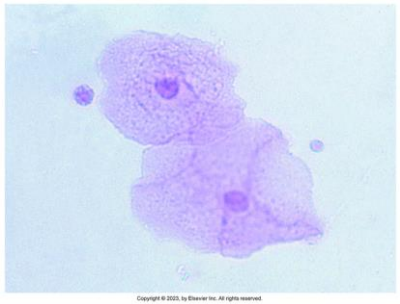
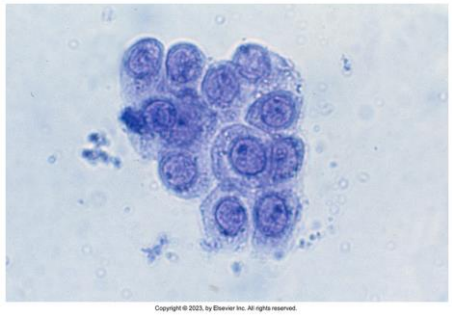
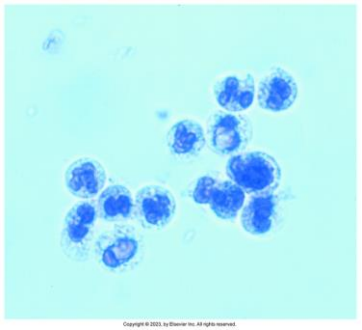
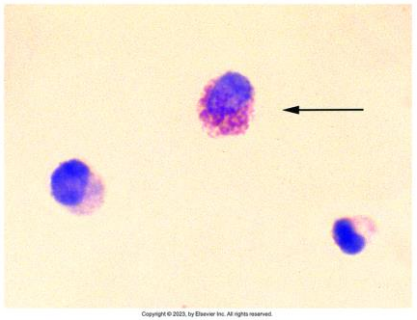
- Used to identify bacteria and yeast; rarely used in urinalysis

Prussian blue

- Stains iron in hemosiderin granules blue; can be free floating or inside cells

Hansel stain

- Methylene blue and eosin-Y; for identification of eosinophils

Microscopy Techniques

Brightfield

- Most commonly used

Phase-contrast

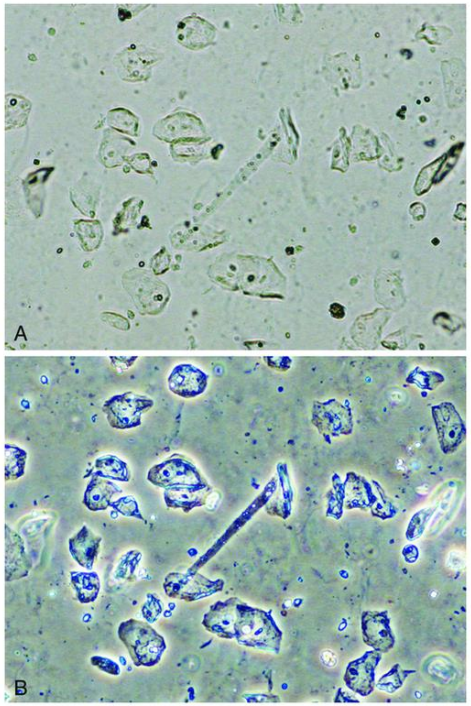
- Ideal for urine sediments; allows more detailed visualization of translucent or low-refractile components and living cells

Polarizing

- Confirms presence of cholesterol, which forms a Maltese cross pattern with polarized light; also used on crystals

Interference contrast

- Gives three-dimensional images, but high cost prevents use by most laboratories



Formed Elements

Originate throughout urinary tract or from contamination

Not all formed elements indicate an abnormal or pathologic process

Presence of large numbers of abnormal formed elements is diagnostically significant

Technologist must be familiar with normal ranges of each component

TABLE 7.4 Reference Intervals for Microscopic Examination*

Component	Number	Magnification
Red blood cells	0–3	Per HPF
White blood cells	0–8	Per HPF
Casts	0–2 hyaline (or finely granular†)	Per LPF
Epithelial cells:		
Squamous	Few	Per LPF
Transitional	Few	Per HPF
Renal	Few (0–1)	Per HPF
Bacteria and yeast	Negative	Per HPF
Abnormal crystals	None	Per LPF

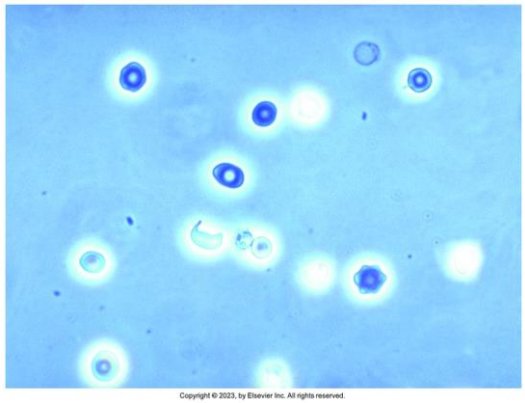
Red Blood Cells (Erythrocytes)

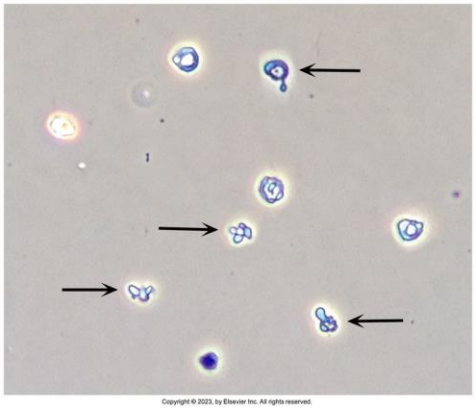
Small, biconcave disks, approximately 8 µm in diameter and 3 µm deep

Can also be crenated, distorted, or ghost cells (ghost cells have no hemoglobin and appear as colorless empty circles)

Viewed and enumerated on high power

Normally, RBCs in healthy adults are less than or equal to 3 per hpf





RBCs

Increased RBCs:

- Along with RBC casts—renal bleeding
- No casts or protein—bleeding below the kidney or caused by contamination (menstruation)

Positive strip test for blood; no RBCs seen:

- RBCs lysed; released hemoglobin tests positive
- False-positive test—detects myoglobin, peroxidases

Negative strip test for blood; RBCs seen:

- Ascorbic acid interference causes false-negative result
- RBC look-alikes present, such as yeast or crystals

White Blood Cells (Leukocytes)

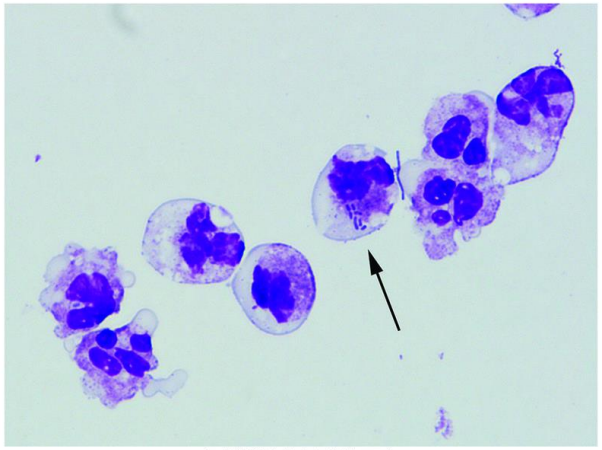
Any type of WBCs from blood can be found in urine

- Neutrophil is most common in both blood and urine

Viewed and enumerated on at least 10 hpf

WBCs in healthy adults usually less than or equal to 8 WBC/hpf

Since WBCs are motile, they can enter urinary tract at any point



WBCs

WBC casts are evidence of an upper urinary tract infection, as are cellular casts and coarsely granular casts; proteinuria will also be present

Increased WBCs but no cellular casts and no or low level of protein are evidence of lower urinary tract infection

Discrepancies between WBCs and leukocyte esterase test:

- WBCs tend to lyse but would still be positive for leukocyte esterase (LE)
- Lymphocytes do not contain LE
- May not be WBCs but look-alikes, such as renal tubular epithelial cells and at times even RBCs

Clinical Significance of White Blood Cells (WBCs)

Increased WBCs in urine termed *leukocyturia*

Inflammatory urinary tract conditions and almost all renal diseases show increased WBCs, particularly neutrophils

Eosinophiluria is a good predictor of acute interstitial nephritis due to drug sensitivity, usually to penicillin and its derivatives

Lymphocyturia—seen in kidney transplant rejection

Epithelial Cells

Some epithelials in urine result from normal cell turnover of aging cells; others represent damage and sloughing from inflammation or disease

Large numbers of some epithelials can indicate improperly collected urine; others indicate a severe pathologic process

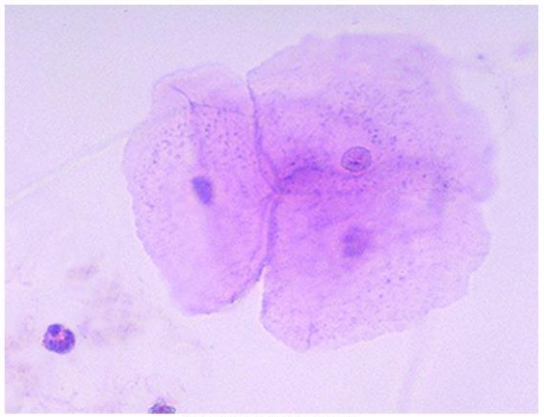
Three basic types of epithelials seen in urine:

- Squamous
- Transitional (urothelial)
- Renal tubular

Epithelial Cells (Cont.)

Squamous

- Most common and largest; use low power
- Line entire urethra in female; distal portion in males
- Rarely significant; usually from contamination



Transitional

- Line renal calyces, pelvis, ureters, and bladder
- Vary in size due to separate layers
- Seen in urinary tract infection (UTI), urinary procedures, carcinoma (in sheets)

Renal tubular

- Each portion of tubule lined with different types of cell
- Convoluted tubular and collecting duct cells enumerated
- Seen in acute ischemic or toxic renal tubular disease from heavy metals or drug (aminoglycosides) toxicity

Casts

Formed in distal and collecting tubules with a core matrix of uromodulin (formerly Tamm-Horsfall protein) secreted by renal tubular cells

Cylindrical, thick in middle, parallel sides

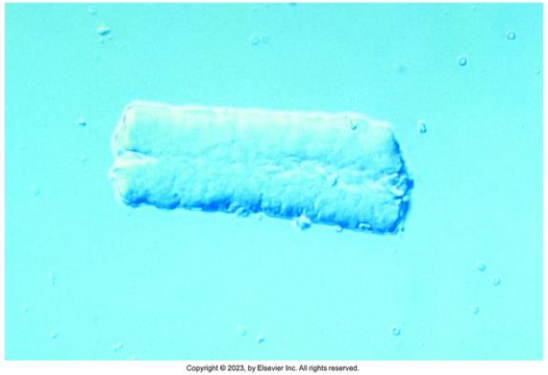
Sizes and shapes vary

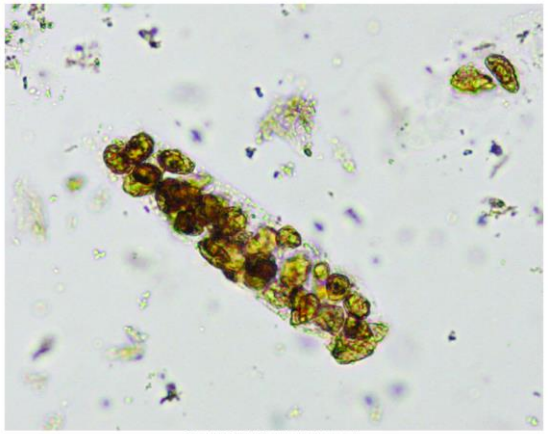
Acid pH, increased solutes, urine stasis, and increased albumin enhance cast formation

A few hyaline or finely granular casts normal; increased numbers and other types seen in renal disease, extreme exercise, and diuretics

Types of Casts

- Hyaline
- Waxy
- WBC
- RBC
- Renal tubular cell
- Mixed cell
- Granular
- Fatty
- Bacterial
- Others





Crystals

Can form in urine on standing

Significant in freshly voided urine

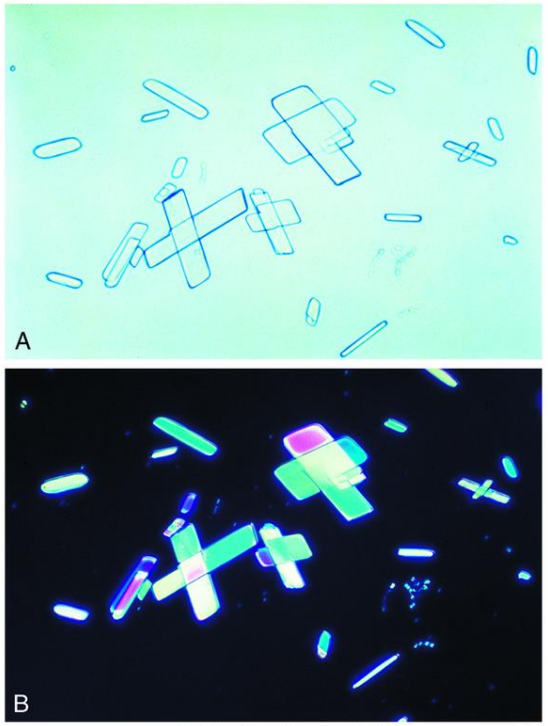
- Formed in vivo and can cause tubular damage

Identified by microscopic appearance and pH

Some indicate a pathologic process

Factors influencing crystal formation:

- Concentration of urine solute
- Urine pH
- Slow flow of urine through tubules



Types of Crystals

Amorphous urates

Acid urates

Monosodium urate

Drugs

Uric acid

Calcium oxalate

Bilirubin

Cystine

Tyrosine

Leucine

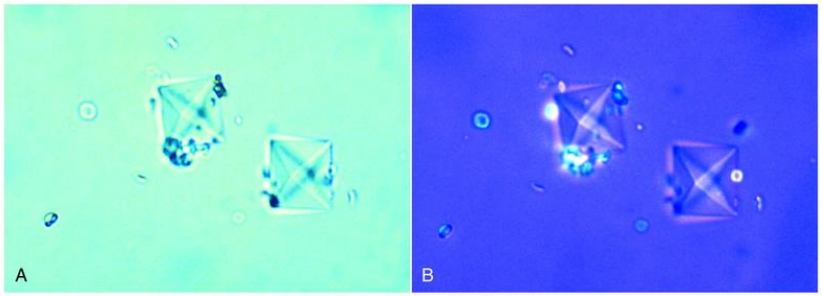
Cholesterol

Triple phosphate

Calcium phosphate

Ammonium biurate

Calcium carbonate



Other Components in Sediment

Microorganisms

- Bacteria
- Yeast
- Trichomonas
- Clue cells
- Parasite eggs or cysts

Other substances

- Mucus
- Fat
- Hemosiderin

Contaminants

- Fecal matter
- Sperm
- Starch
- Fibers