Urinalysis Part II
Microscopic Examination

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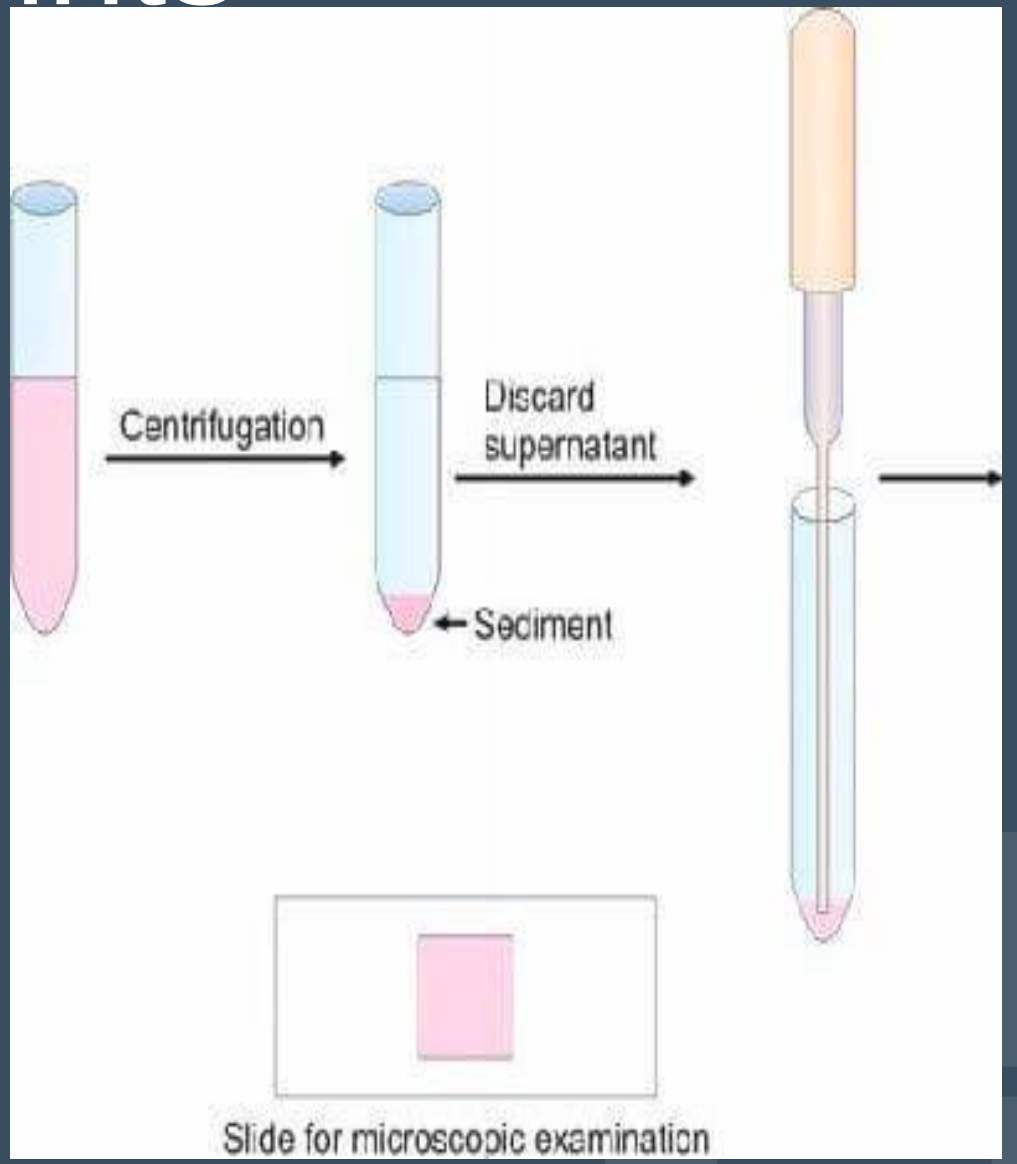
General points

- Aliquot centrifuged for 5 minutes at relative centrifugal force of 400
- Aspirate off urine so there is uniform amount of urine and sediment
  - Resuspend specimen
  - 20µL sample onto slide (with coverslip)



Microscopic Examination

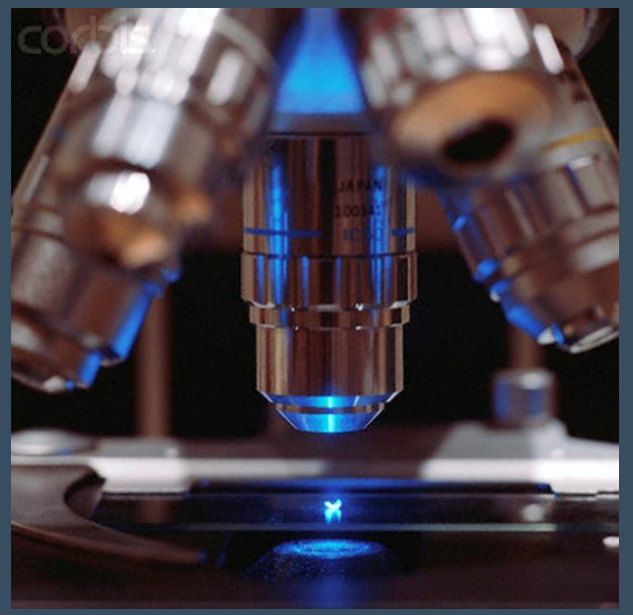
- Review at 10x (lpf) and 40x (hpf)
  - Cast – average per low power field
  - RBCs and WBCs- average per high power field
  - Epithelial cells, crystals and others- few, moderate, or many

Cellular Elements

- Red blood cells
- White blood cells
- Epithelial cells
- Bacteria
- Crystals
- Casts

Types of microscopy

- Bright-field
- Phase-contrast
- Polarization
- Interference-contrast
- Dark-field
- Fluorescence



Sediment appearance

- Cells and casts in various stages of development and degeneration
- Distortion of cells and crystals by the chemical content of the specimen
- Contamination by artifacts
- Low refractive index elements are often difficult to see under bright field microscopy

Stains

- Sternheimer-Malbin
  - Most frequently used stain and increases refractive index
  - Crystal violet and safranin O
  - WBCs, epithelial cells, casts
  - Outlines cytoplasmic and nuclear details
- Toluidine blue
  - Enhances nuclear detail
  - Helps differentiate WBCs and renal tubular epithelial cells

Stains

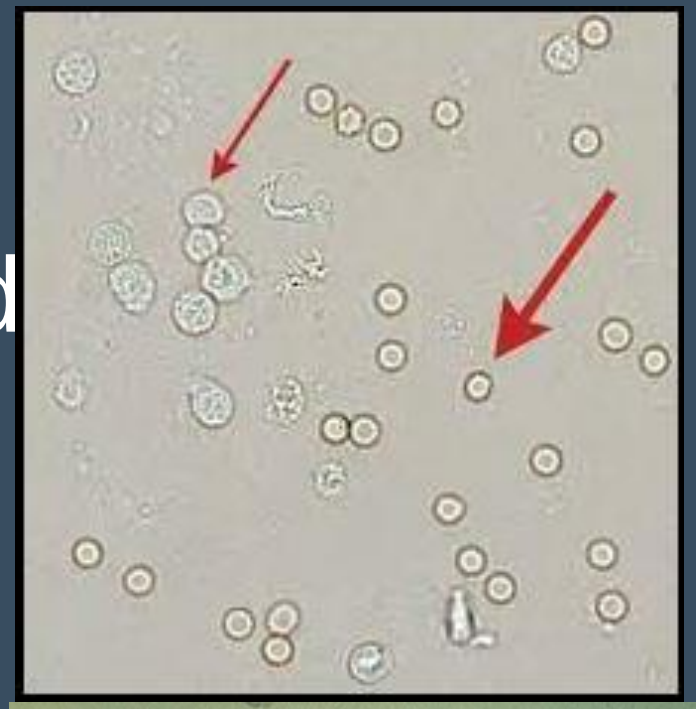
- Acetic acid (2%)
  - Lyses RBCs, enhances WBC nuclei
- Lipid stains
  - Oil red O and Sudan III
  - Stains triglycerides and fats orange-red
  - Cholesterol does not stain

Stains

- Gram stain
  - Differentiates bacterial casts from granular casts
- Hansel Stain
  - Eosinophils
  - Drug induced allergic reaction
- Prussian blue stain
  - Hemosiderin granules seen with hemogloinuria

RBC

- Appear as smooth, non-nucleated
- Normal range : 0-3 to 5/ hpf
- Crenated cells in hypersthenuric
- Ghost cells in hyposthenuric
- Dysmorphic in glomerular disease



RBC (cont’d)

- Identification difficulties
  - Yeast: look for buds
  - Oil droplet : refractility
  - Air bubbles: refractility and possibly in a different plane
  - Starch: refractile and polarizes
  - Reagent strip correlation

Clinical Significance

- Macroscopic versus microscopic hematuria
  - Cloudy red urine, acute infection, damage to glomerular membrane or vascular injury to the genitourinary tract
  - Clear urine, early glomerular diseases, malignancy of urinary tract and to confirm the presence of renal calculi

WBC

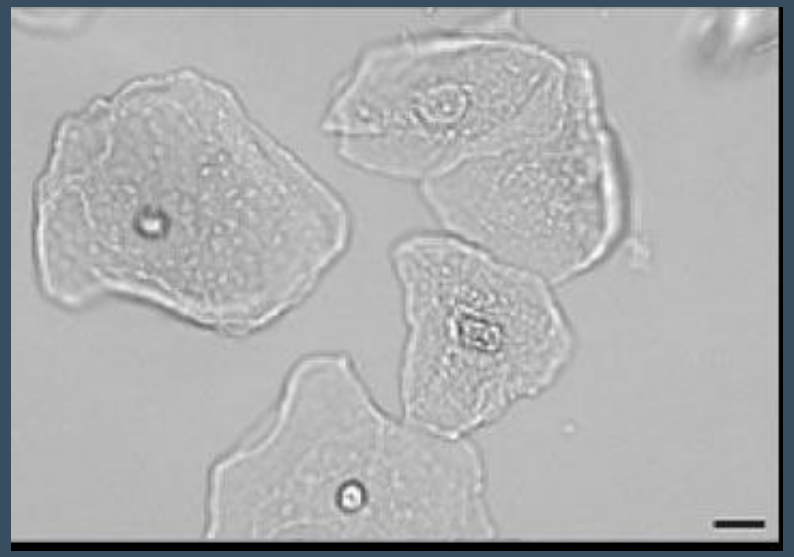
- Neutrophils are most predominant
  - Granular cell with visualized nuclei
- Larger than RBCs & smaller than epithelial
- ↑numbers indicate inflammation or infection somewhere along urinary tract
- Glitter cells
  - Hypotonic urine
  - Brownian movement
  - Swell; granules sparkle

Clinical Significance

- Normal range: 0-5/hpf
- Increased WBCs= pyuria
- Infection : pyelonephritis, urethritis , cystitis, and prostatitis
- Reporting the presence of bacteria in specimens containing WBCs is important

Epithelial cells

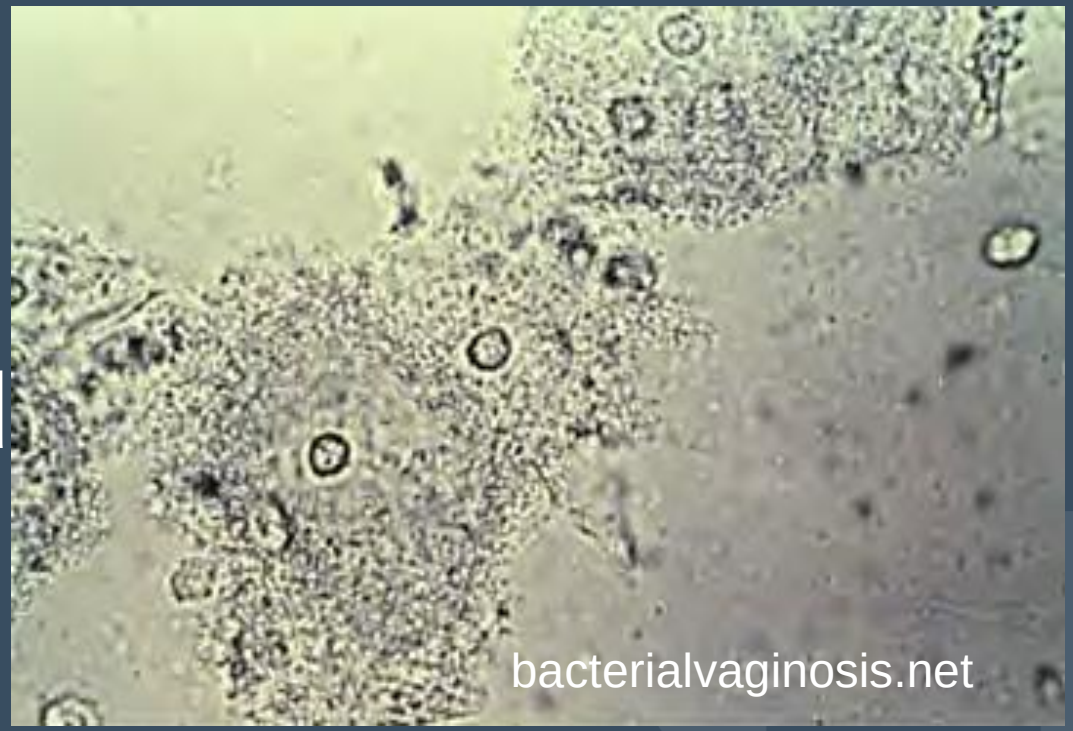
- Squamous epithelial cells
  - largest cell appearing in urine sediment
  - Irregular outline, prominent nucleus & occur singly or in sheets



- Significance:
  - Certain number of epithelial cells in urine is normal
  - Most often contaminant when present in large number
  - Pathologic condition : cystitis, pyelonephritis

Clue cells

- Squamous cells with pathological significance
- Infection by Gardnerella vaginalis
- Coccobacillus sp. covers most of the cell and extend over the edges
- Vaginal wet prep or urine sediment



Transitional (urothelial)

- Renal pelvis, ureter, bladder
  - Smaller overall size
  - Larger nucleus (central)
  - Spherical to spindled shape
- Causes
  - Catheterization
  - If atypical, malignancy or viral infection

Renal Tubular Epithelial Cells (RTE)

- RTE cells are the most clinically significant urine epithelial cells; indicate tubular necrosis
- Oval fat bodies: RTE cells that have absorbed lipid in the filtrate
  - Maltese cross formation with polarized light
  - Sudan III or oil red O stain
- Also free floating refractile droplets
- Lipiduria: nephrotic syndrome, acute tubular necrosis, diabetes

Transitional Epithelial and RTE Cells

Figure 7–22 Transitional epithelial cells.

Figure 7–24 Caudate transitional epithelial cells (×400).

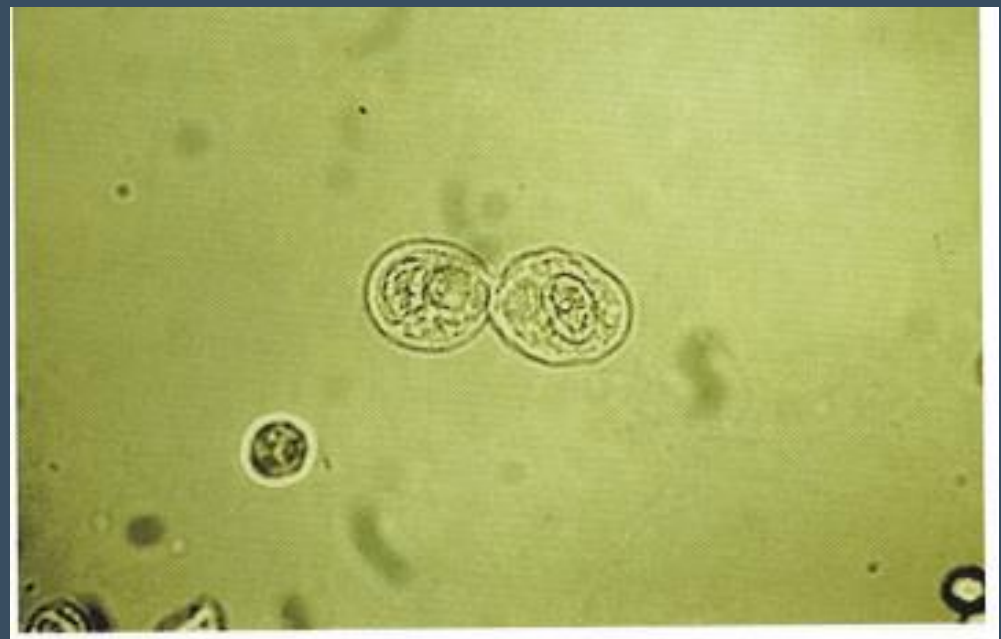


Figure 7–27 RTE cells. Oval distal convoluted tubule cells. Notice the eccentrically placed nuclei (×400).

Oval fat bodies

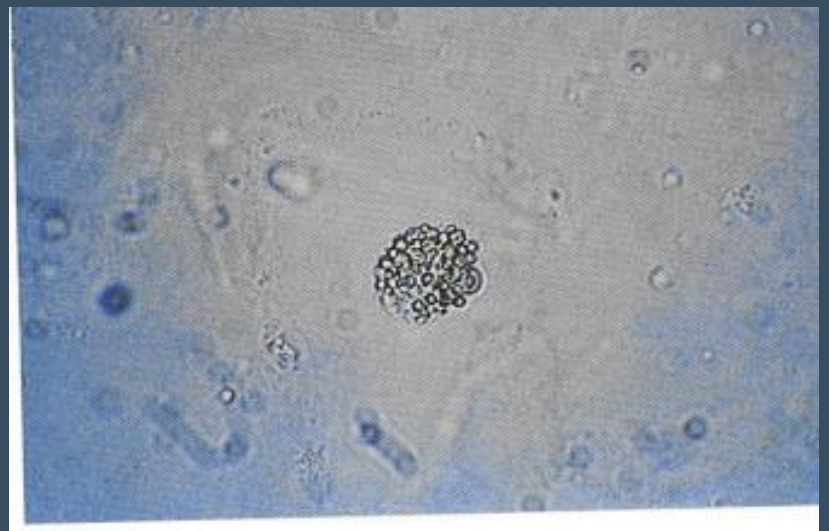
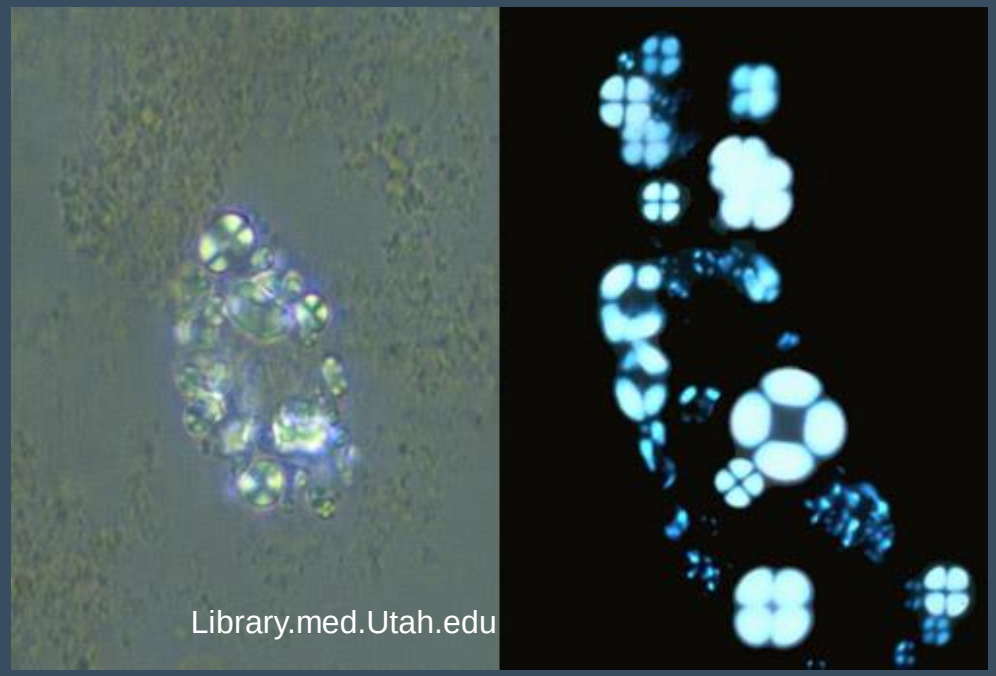


Figure 7–31 Oval fat body (×400).



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Casts

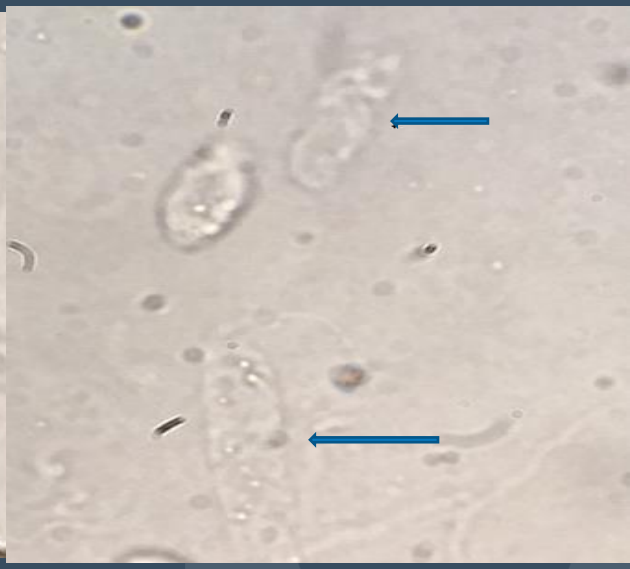
- Hyaline
- RBC
- WBC
- Epithelial cell
- Bacterial
- Mixed cell
- Fatty
- Granular
- Waxy
- Broad

Casts

- Tamm-Horsfall protein
  - Found in normal urine
  - Not detected by reagent strip
- Precipitation and denaturation of protein
  - Low flow rate (stasis)
  - High salt concentration
  - Acidity (low pH)
- Formed in distal tubules and collecting ducts
- Average # per 10 lpf

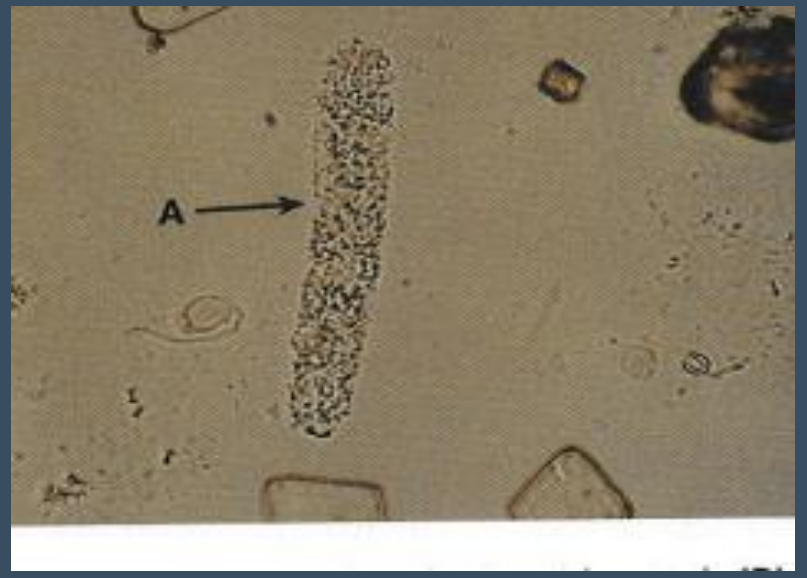
Hyaline

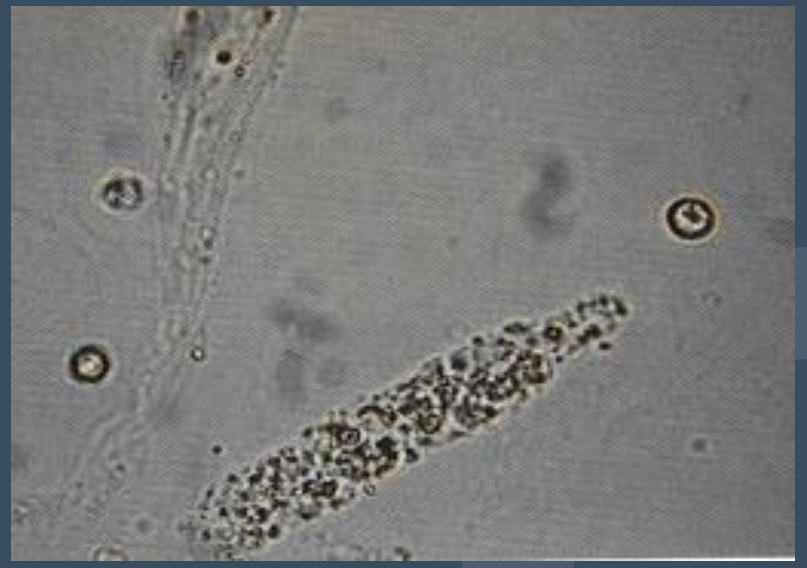
- T-H protein matrix
  - Most frequent type
  - Colorless, transparent
  - 0-3 per lpf normal finding
    - Exercise, stress, dehydration
  - More than 3 per lpf
    - Glomerulonephritis
    - Pyelonephritis
    - Chronic renal disease
    - Congestive heart failure



Granular

- Disintegration of cellular casts
  - Coarse to fine granulation
  - Occasionally cell remnants present
- Can rarely be seen for non-pathologic reasons
  - Lysosome secretion by tubular cells during extreme exercise or stress
  - Seen in association with hyaline casts





Cell Casts

RBC Cast

- Matrix containing RBCs
  - Orange-red color
- Nephron hemorrhage
  - Glomerulonephritis
    - Most common cause
  - Rarely with strenuous exercise
- Hemoglobin cast
  - Orange-red color, no RBCs

WBC Cast

- Matrix containing WBCs
- Nephron infection or inflammation
  - Pyelonephritis
    - Most common cause
  - Acute interstitial nephritis
  - Glomerulonephritis (with RBC casts)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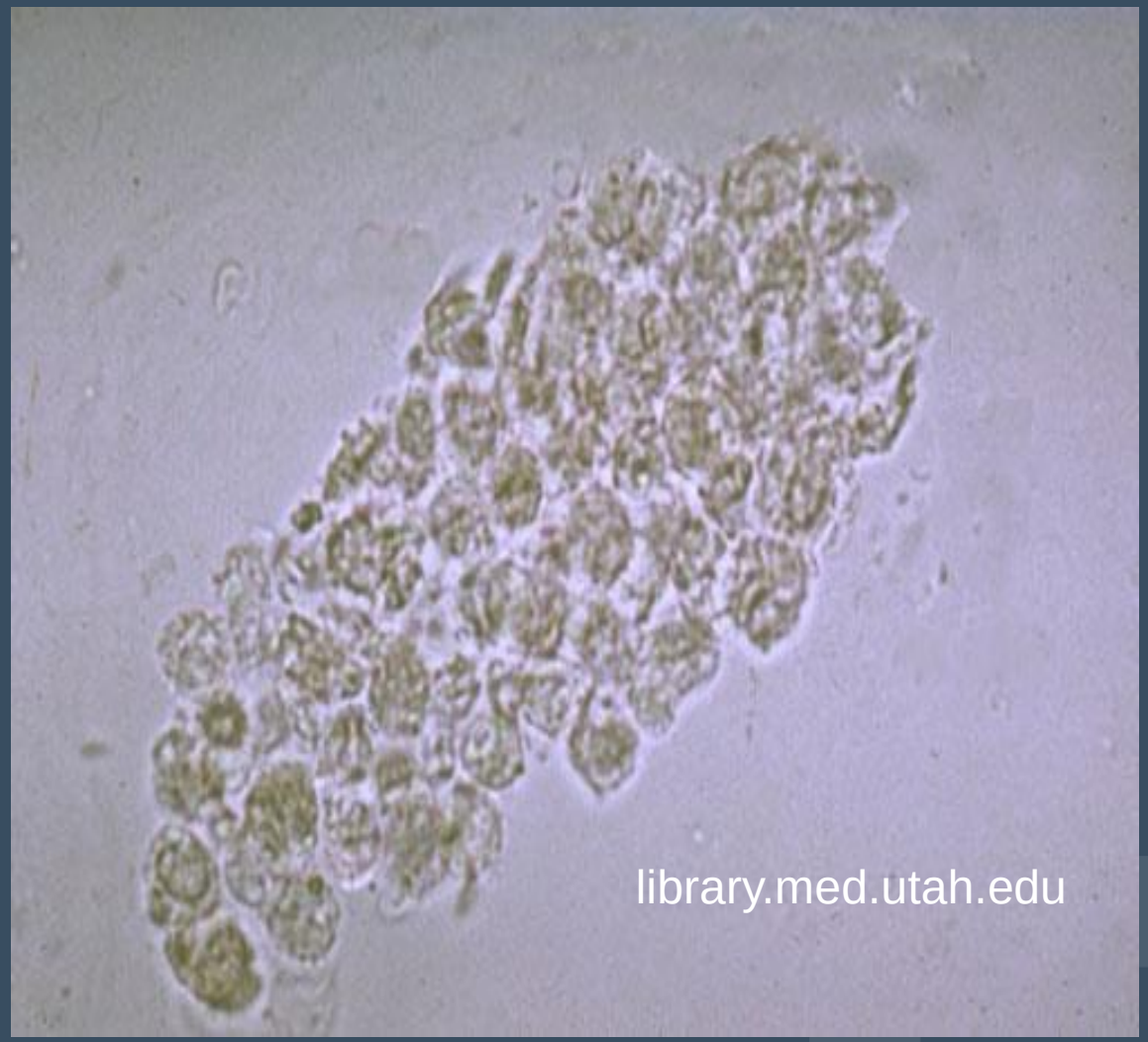
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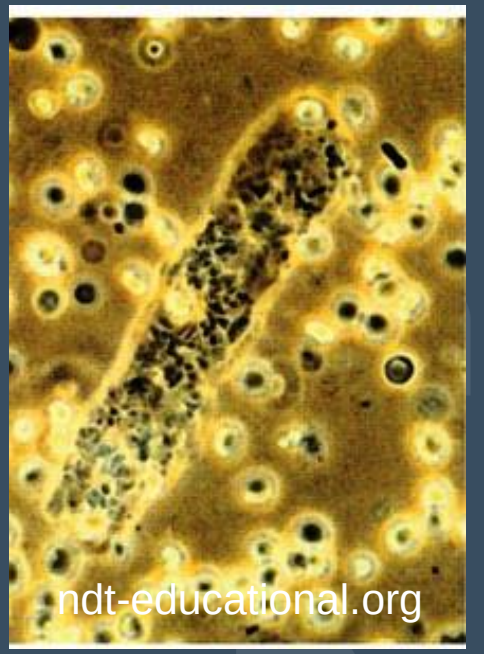
Cast

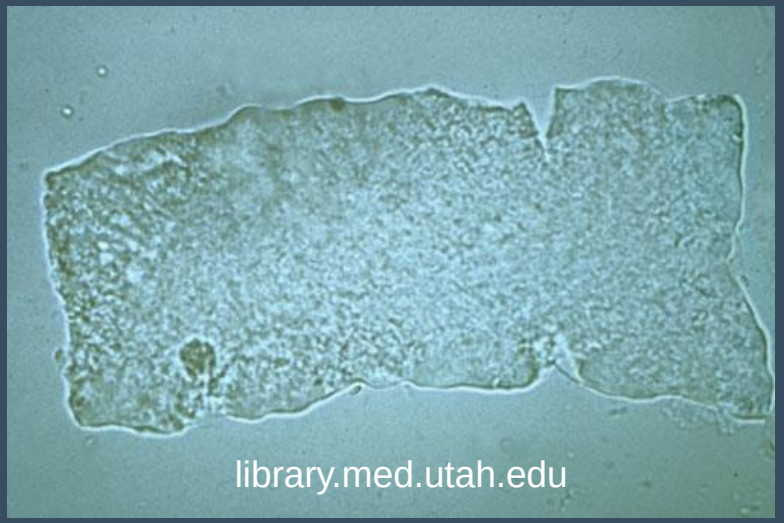
- Epithelial cell
  - Matrix containing renal tubular cells
  - Tubular disease
  - Pyelonephritis (with WBC casts)
    - Staining can help discriminate nuclear details
- Bacterial
  - Rarely seen
  - WBC casts often present
  - Can be confused with granular casts
    - Confirm with Gram stain



Cast

- Fatty
  - Matrix with fat droplets
    - Few to many droplets present
    - Same properties as oval fat bodies
  - Causes
    - Nephrotic syndrome, Tubular necrosis
    - Diabetes mellitus, Trauma

Cast



- Waxy
  - Prolonged urinary stasis
    - Chronic renal failure
  - Further disintegration of granular casts
  - Higher refractive matrix than hyaline cast
  - Generally broad and stubby w/blunt ends
  - Well defined parallel margins that may be notched

Crystals

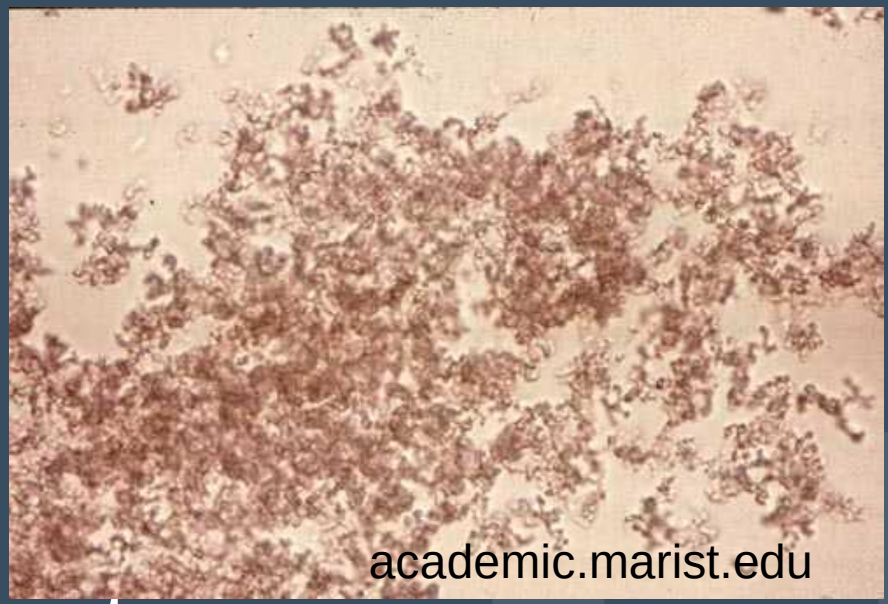
- Formed from precipitation of urine solutes: salts, organic compounds, and medications
- Formation based on temp, solute concentration and pH
- High specific gravity needed in fresh specimens
- Most valuable ID is urine pH

Crystals

Normal		Abnormal	
Acidic Urine	Alkaline Urine	Acidic Urine	Alkaline Urine
Amorphous Urates	Amorphous Phosphates	Cystine	None
Uric Acid	Triple Phosphates	Tyrosine	
Calcium Oxalate	Ammonium Biurates	Leucine	
	Calcium Phosphates	Sulfonamide (Sulfadiazine)	
	Calcium Carbonates		

Amorphous urates

- Yellow-brown to pink granules
- Urine sediments has pink color due to the pigment uroerythrin attaching on surface of granules
- Frequently form in refrigerated specimen
- Granular appearance
  - Heat soluble
- Often in clumps; may resemble casts

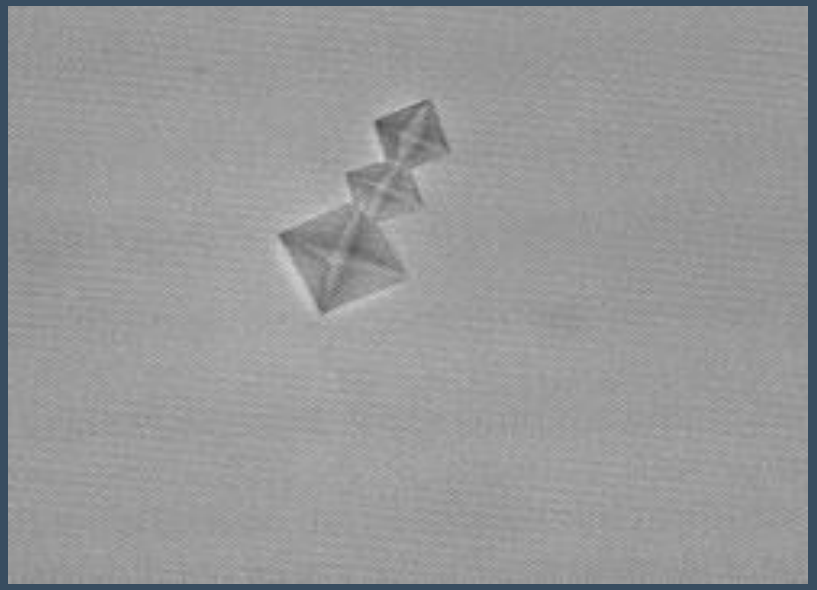


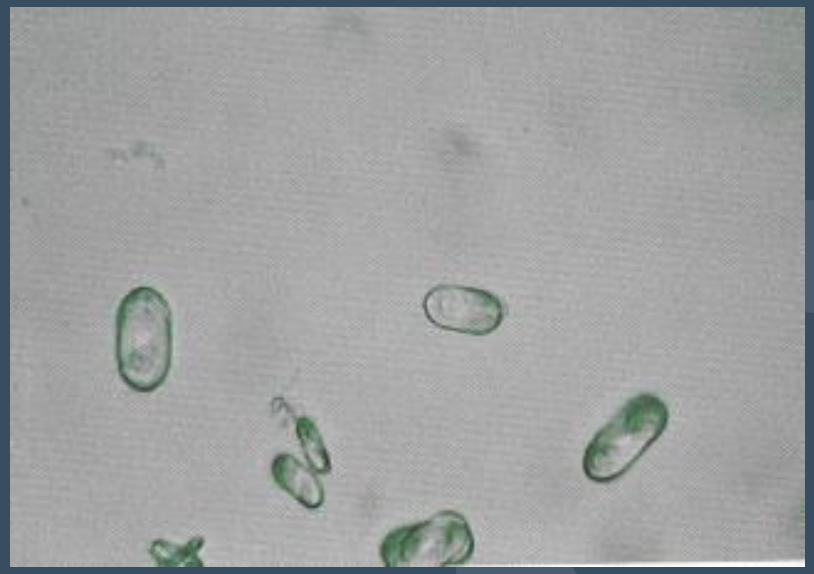
Uric acid

- Rhombus, flat plates, wedges, rosettes
- Colorless to yellow-brown
- Birefringent under polarized light
- If many present, consider…
  - Gout
  - Tumor lysis syndrome
  - Certain metabolic disorders

Calcium oxalate

- Dihydrate (most common)
  - envelope or two pyramid-shaped
- Monohydrate is oval or dumbbell shaped
- Calcium oxalate is a major component of renal calculi



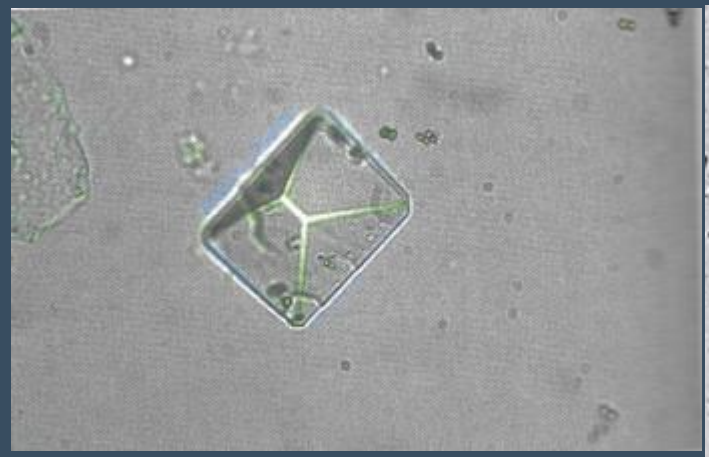


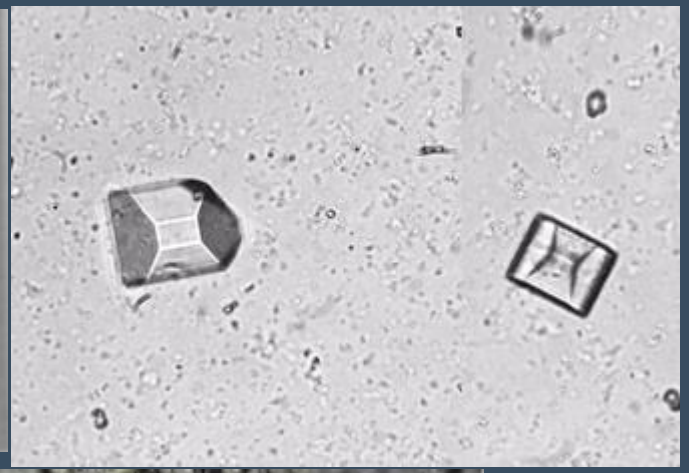
Ammonium biurate

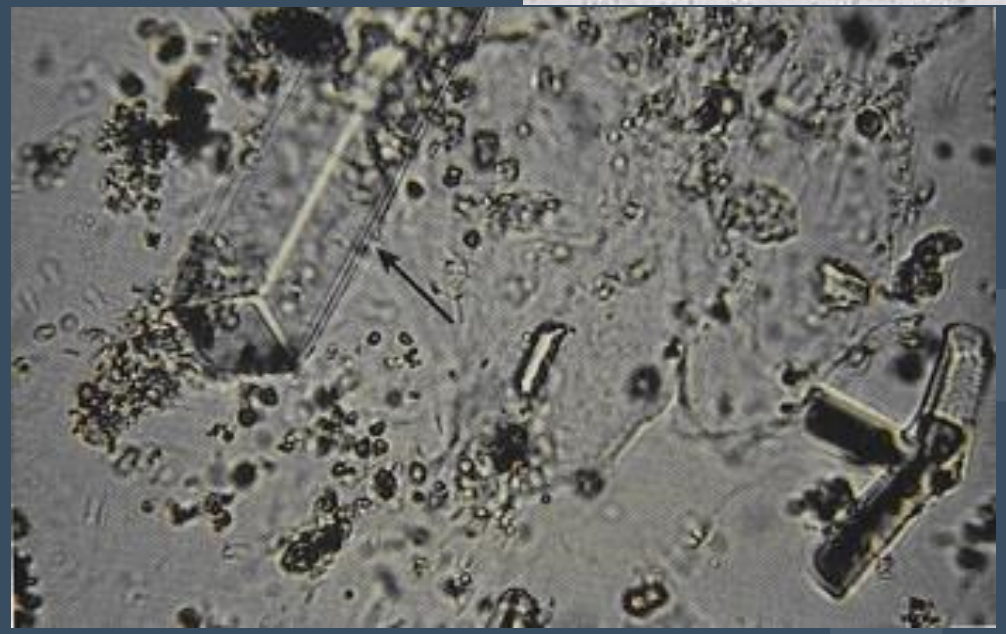
- Yellow-brown, spicule covered spheres; “Thorny apple”
- May be associated with urea-splitting bacteria or old specimens

Triple phosphate

- Colorless, prism, or coffin lid shaped
- Polarize
- Associated with urea-splitting bacteria
- No clinical significance

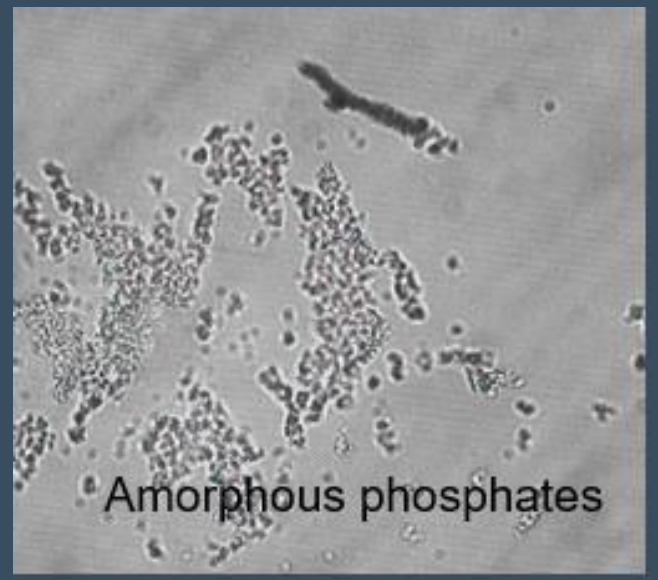


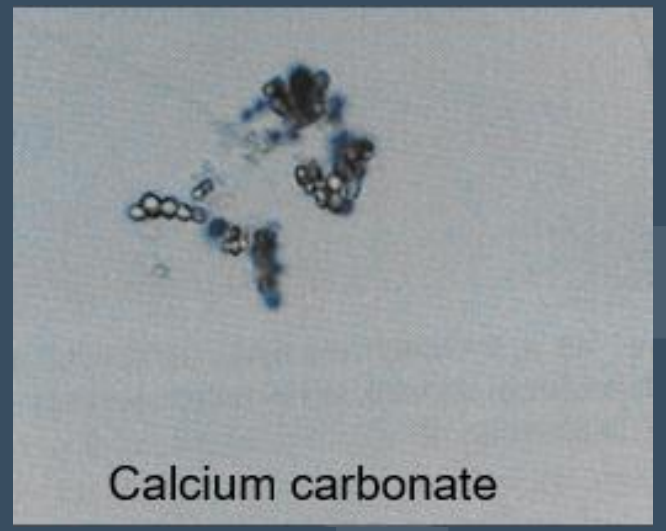




Crystal

- Amorphous phosphates
  - White , granular appearance
  - Not heat soluble
- Calcium phosphate
  - Colorless, not commonly seen
  - Flat rectangular plates or rosettes
- Calcium carbonate
  - Small, spherical to dumbbell-shaped
  - Gas produced with addition of acetic acid



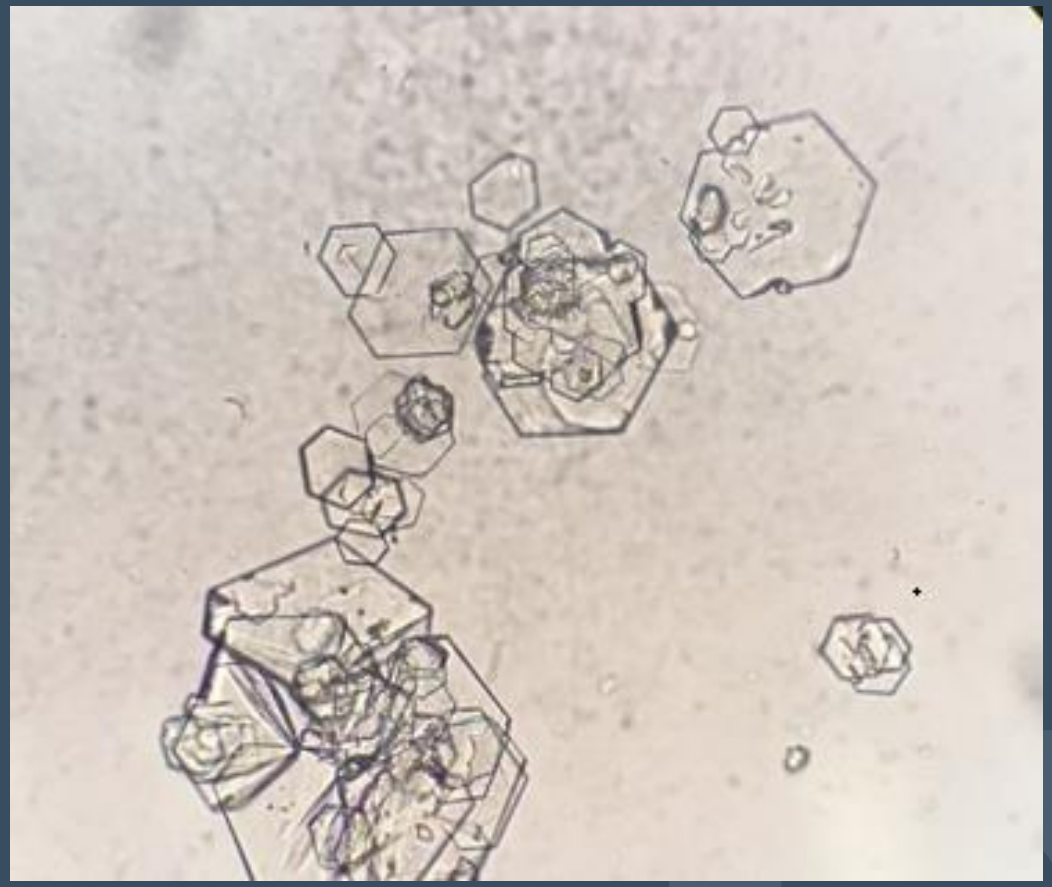


Abnormal urinary crystals

- Cystine
- Cholesterol
- Leucine
- Tyrosine
- Bilirubin
- Sulfonamides
- Radiographic dye
- Ampicillin

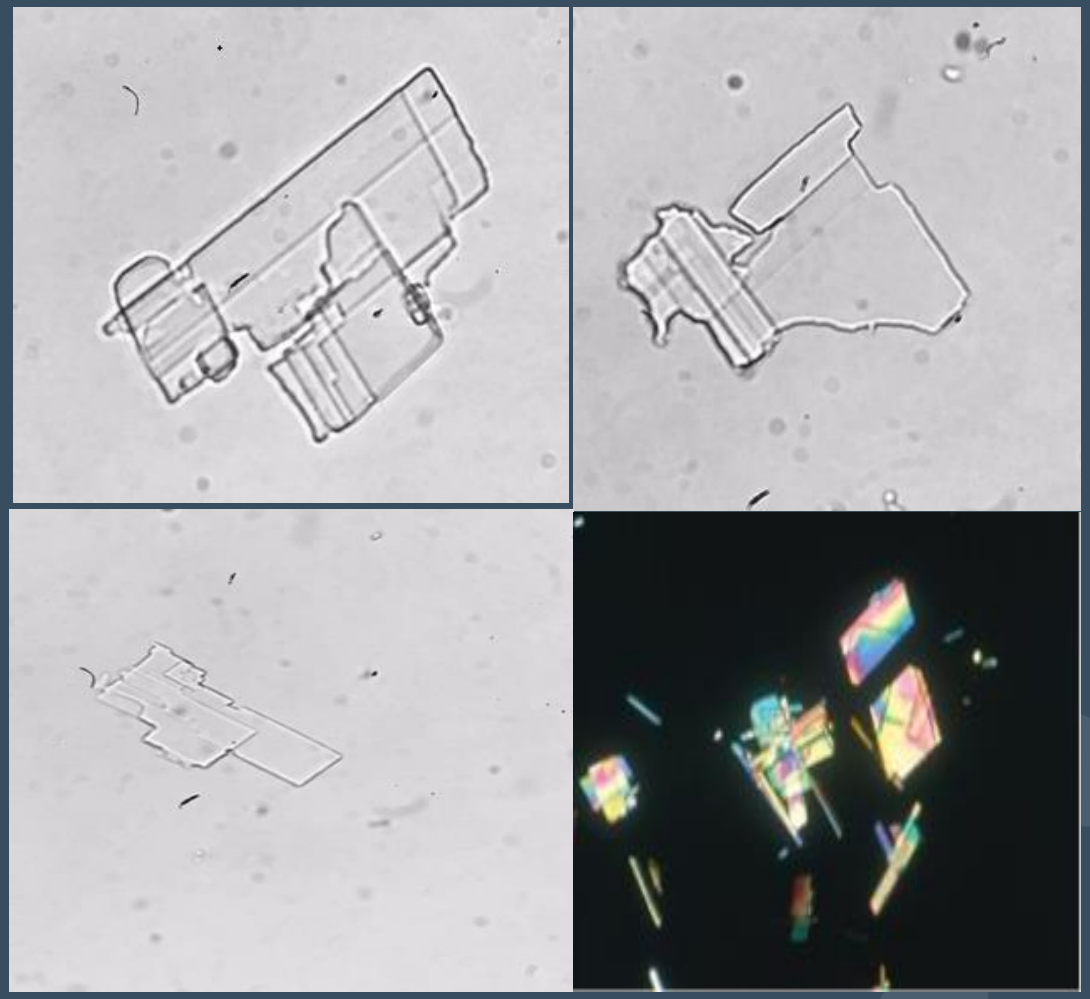
Cystine

- Colorless
- Hexagonal plates
- Seen in cystinuria: inability to reabsorb cystine
- Confirm: cyanide nitroprusside



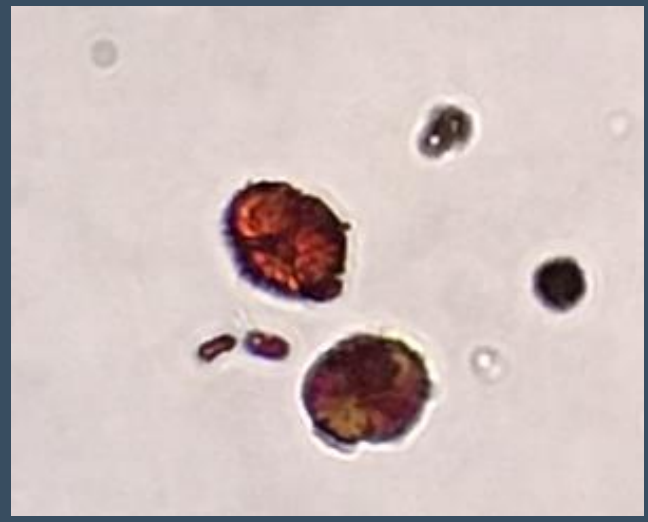
Cholesterol

- Refrigerated specimens
- Colorless, notched rectangular plates
- Highly birefringent
- Lipiduria
  - Nephrotic syndrome
  - Seen in conjunction with fatty casts and oval fat bodies



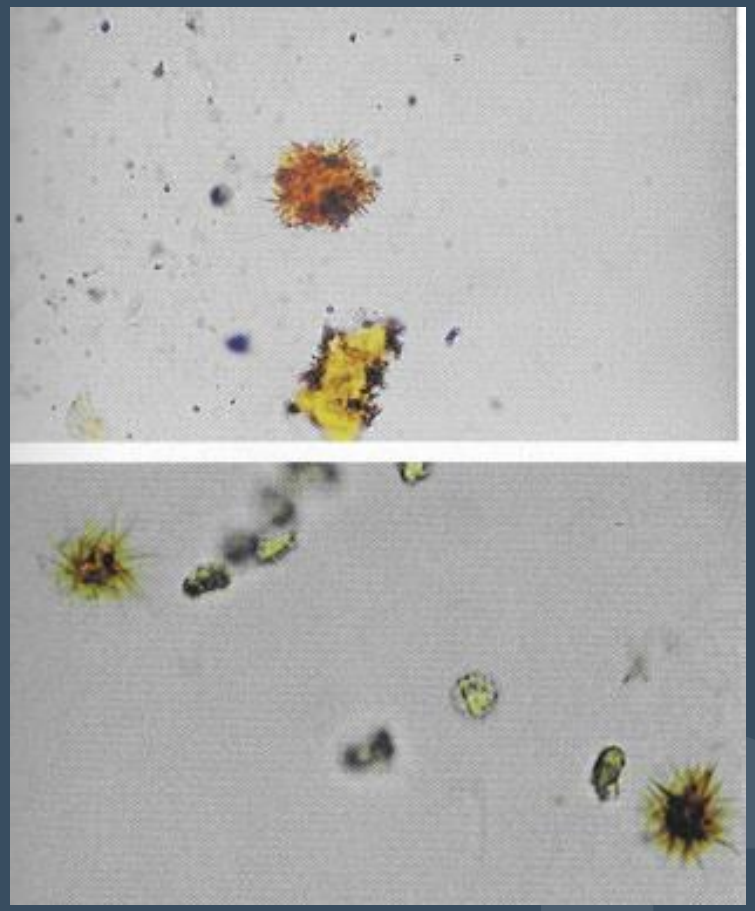
Liver Disease Crystals

- Leucine crystals
  - Yellow-brown spheres with concentric circles and radial striations
  - Maltese cross
- Tyrosine crystals
  - Fine yellow needles in clumps or rosettes
  - Seen with leucine crystals
  - Inherited AA disorders



Liver Disease Crystals

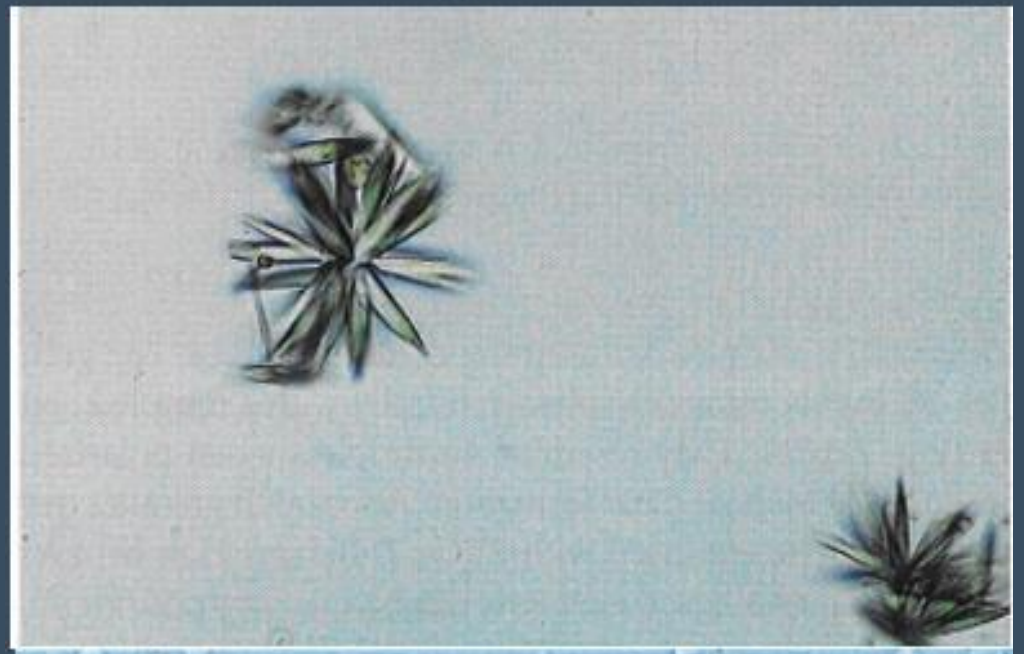
- Bilirubin crystals
  - Characteristic yellow color
  - Clumped needles or granules
  - Positive bilirubin on UA dipstick



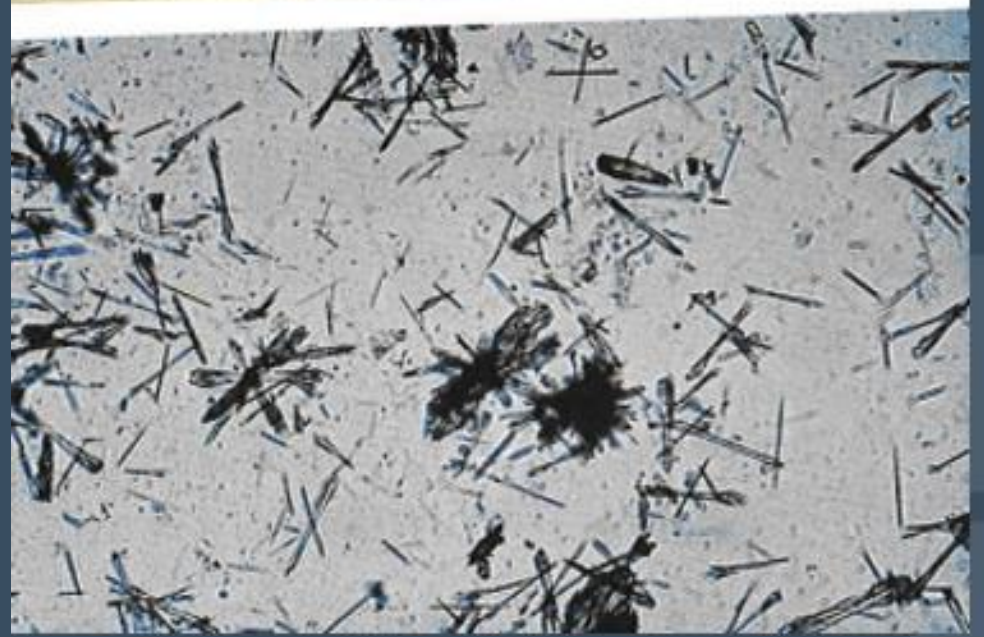
Radiographic dye

- Radiographic dye
  - Similar to cholesterol crystals, polarize
  - Patient history
  - Markedly increased SG with refractometer
- Sulfonamide crystals
  - Variety of colors and shapes
    - Colorless to yellow-brown
    - Needles, rhombi, fan-shaped
  -

Sulfa crystals

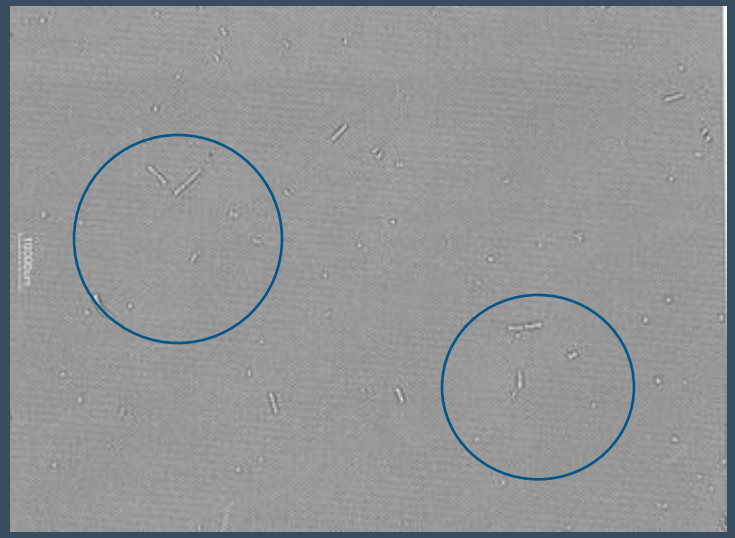


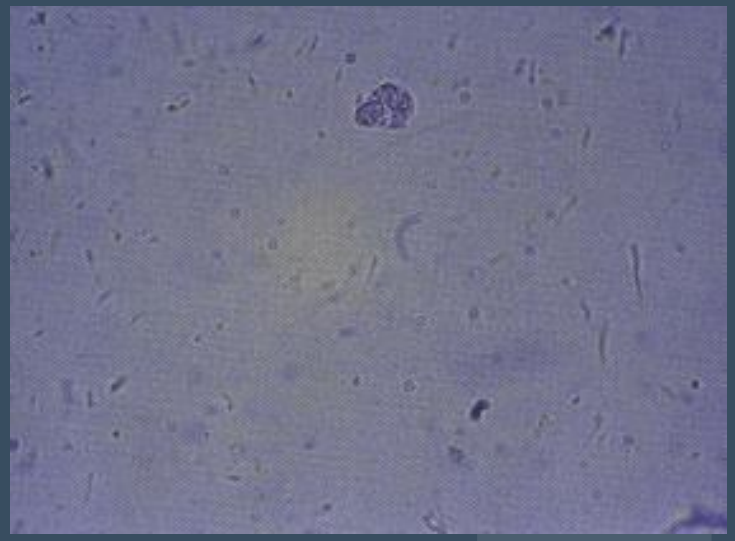
Ampicillin crystals



Bacteria

- Normally not present unless not collected in sterile condition
- May indicate contamination
- Bacteria should be accompanied by WBCs to be considered for UTI
- Nitrite helps to confirm bacilli not cocci
- Report few, moderate , many per hpf





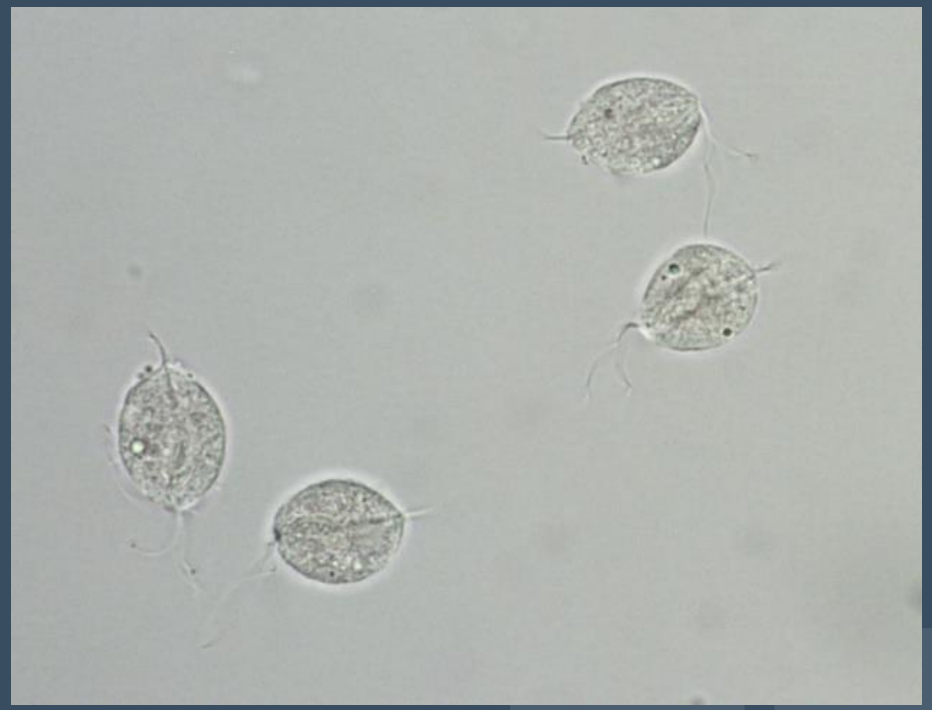
Yeast

- Appear as small, refractile, oval structure with may or may not contain a bud
- Mycelial forms or branched in severe infection
- A true yeast infection should be accompanied by the presence of WBCs
- Diabetic urine: ↑ glucose and acid ideal for yeast growth

Budding and Hyphenated Yeast

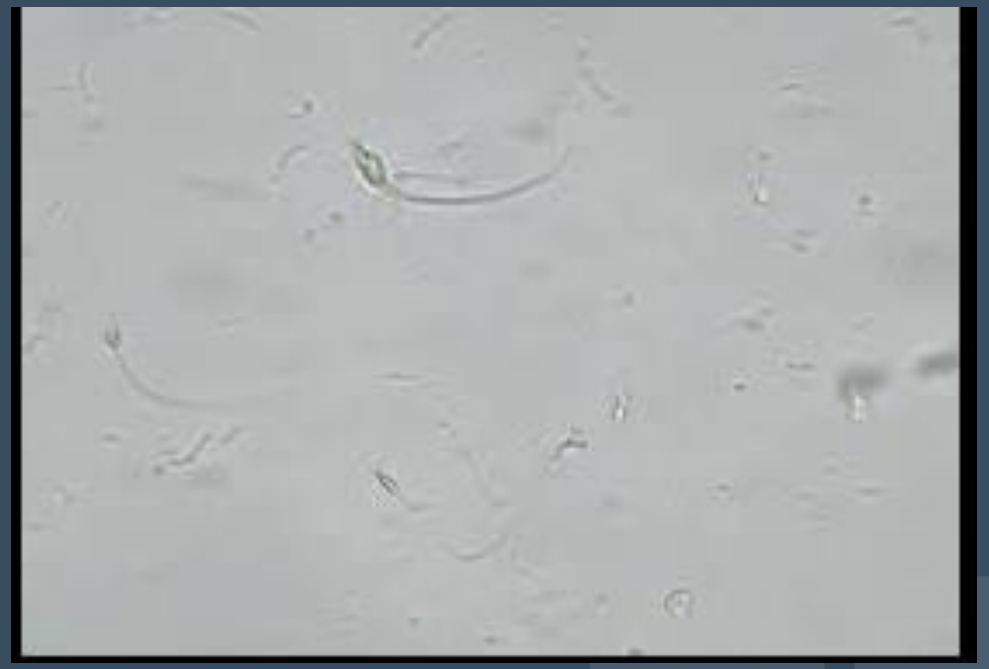
Parasites

- Trichomonas vaginalis is most frequently encountered
- Darting movement in wet prep of sediment
- Phase microscopy enhance visualization of flagella or undulating membrane



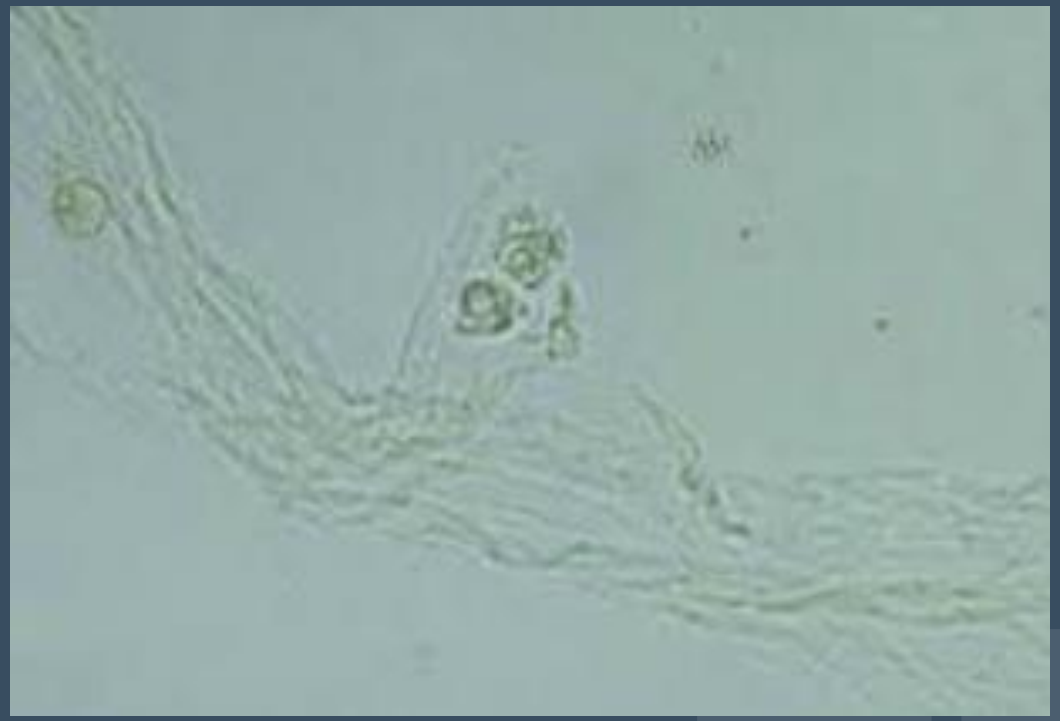
Spermatozoa

- Oval, tapered heads and long flagella like tails
- Rarely significant, infertility: sperm expelled into bladder instead of urethra
- Lack of clinical significance , legal consequences



Mucus

- Protein from RTE, glands, squamous cells
- Threadlike, low refractive index
- Confuse with casts
- No clinical significance



Artifacts