Wildlife under the Microscope

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Blue Ridge Community College
Florida wildlife Symposium

Making a diagnosis

- Need a history
- Perform a physical exam on the patient
- Develop a list of rule outs
- Use diagnostic aids to confirm or eliminate them
  - or to assess patient’s condition

Diagnostic aids

- Blood tests
  - CBC, chemistrys, toxins
- Cytology and/or biopsy
- Urinalysis
- Fecal analysis
- Radiography
- Diagnostic ultrasound
- Catscan
- MRI
- Endoscopy
- Laparoscopy
- Necropsy

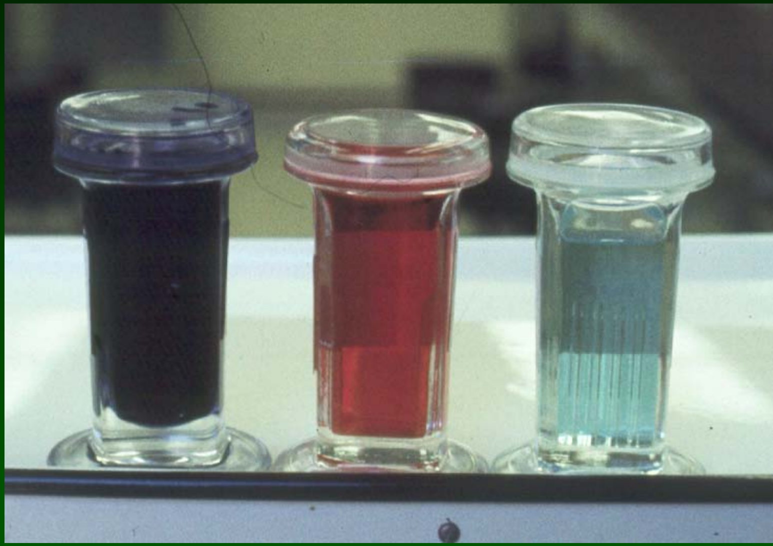
The microscope

- A useful diagnostic tool
- Used in many tests
- Many types and models available
- Magnify 40-1000X

Lab Equipment

- Microscope
- Slides
- Stains
- Immersion oil
- Leukopets
- Hemocytometer

Stains



- Diff Quik
  - general purpose stain
- New Methylene Blue
  - supravital stain
- Gram stain for bacteria
- Acid fast stain
  - for certain organisms
- Many others available

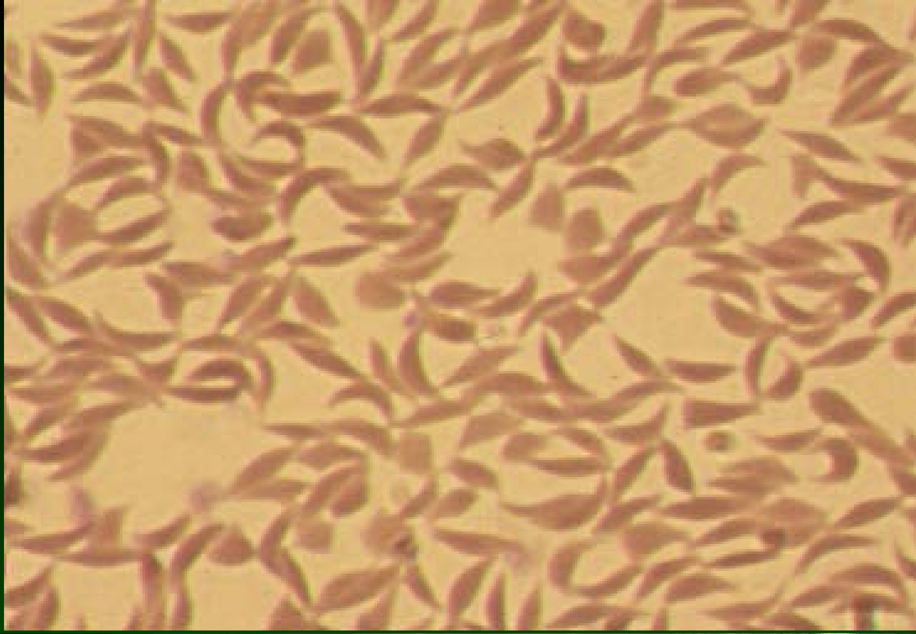
Lab samples

- Blood
- Urine
- Feces
- Tissues
- Ear swabs/skin scrapings
- Discharges, effusions

Using the scope

- Scan slide on low power
  - evaluate staining, cellularity
  - if ok, find place to start
- Move 40x objective into place
  - readjust light
- If needed, place drop of oil on spot
  - move 100x objective into place

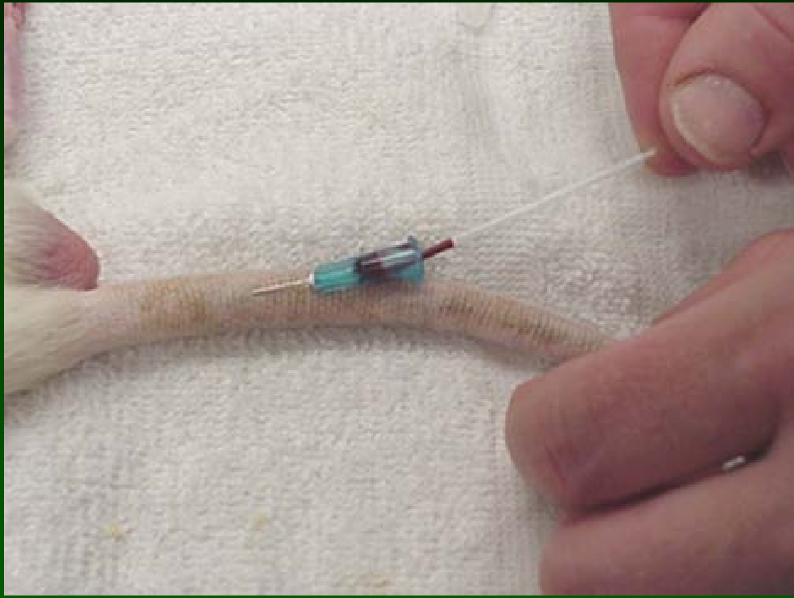
Blood






Ear vein

Tail Veins



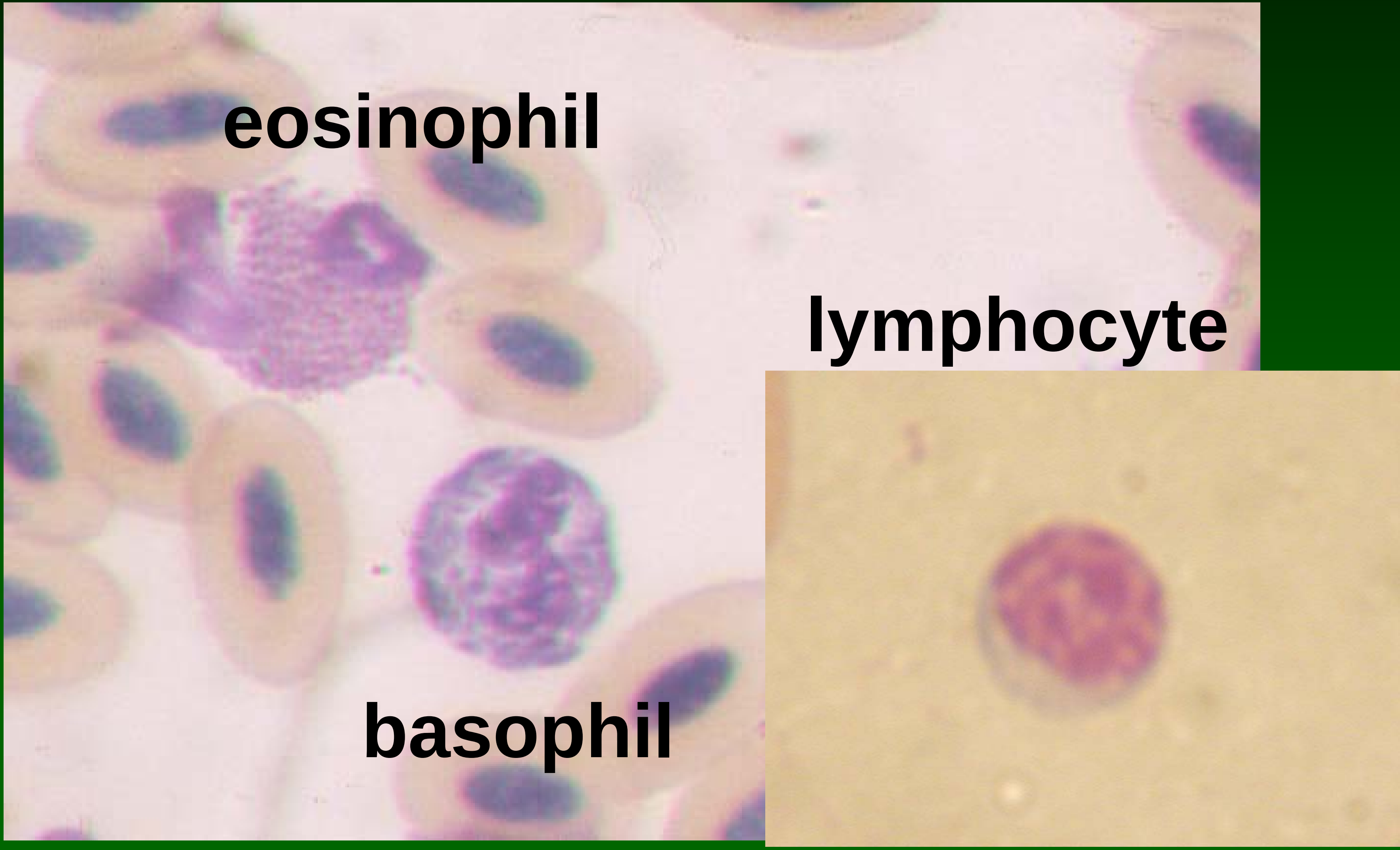
Looking at blood

- Similar cells in all animals
- Parasites do vary
- Need to learn differences
- Blood reveals a lot

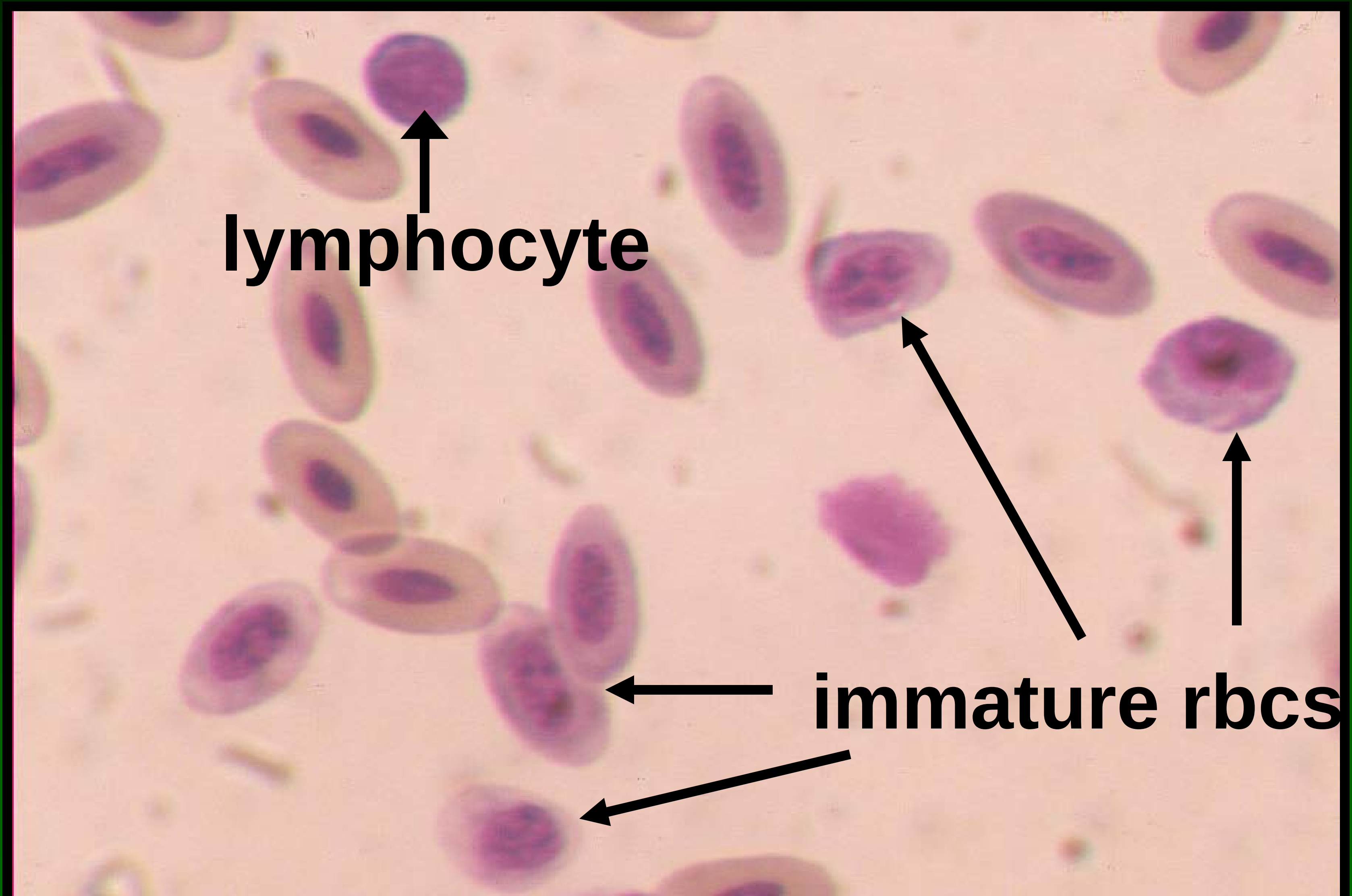
Microfilaria, D. immitis

Can id cell types

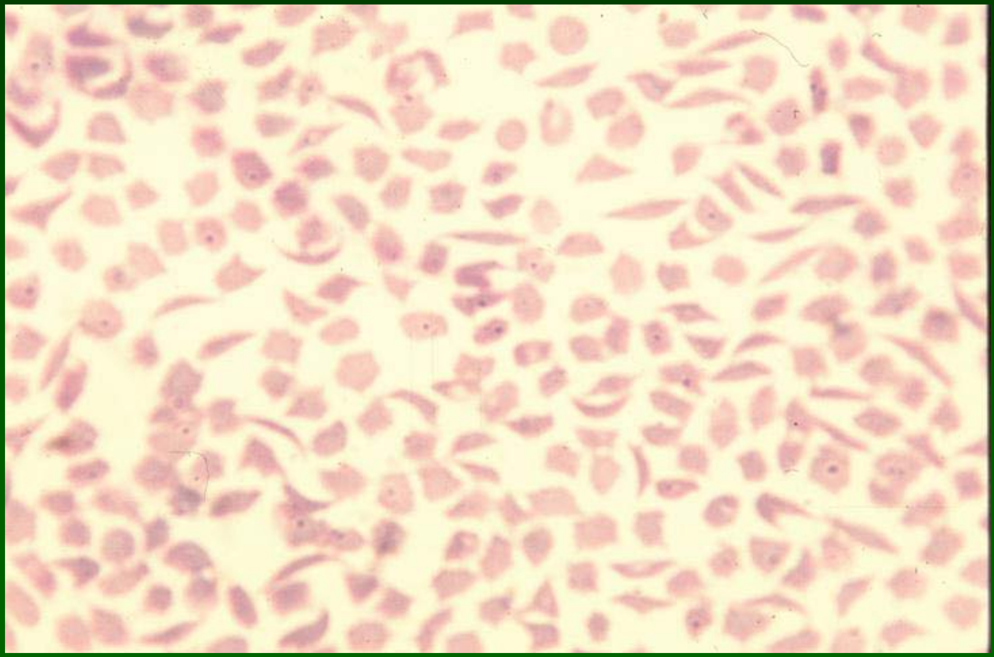
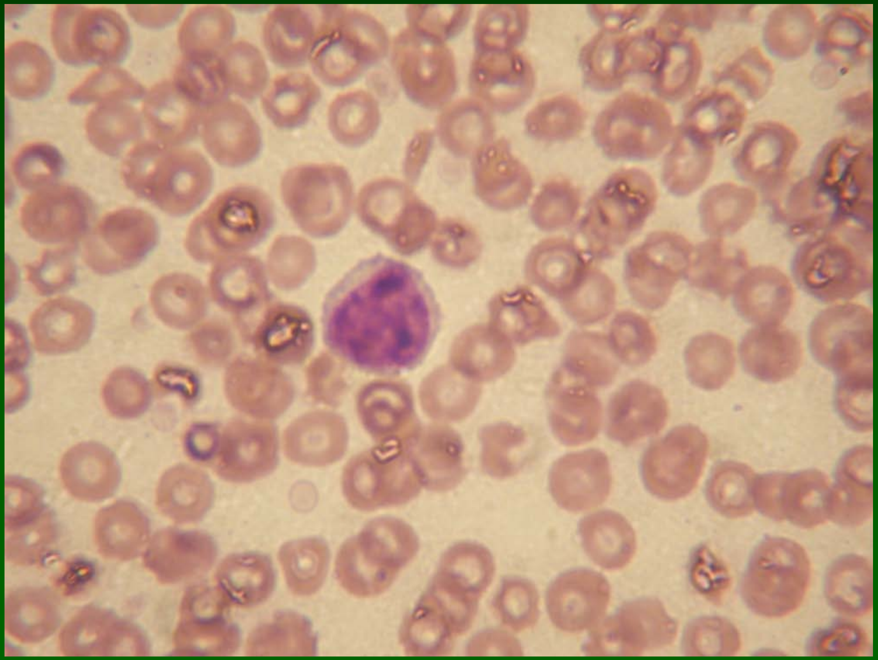
Bird blood cells



Can see immature cells

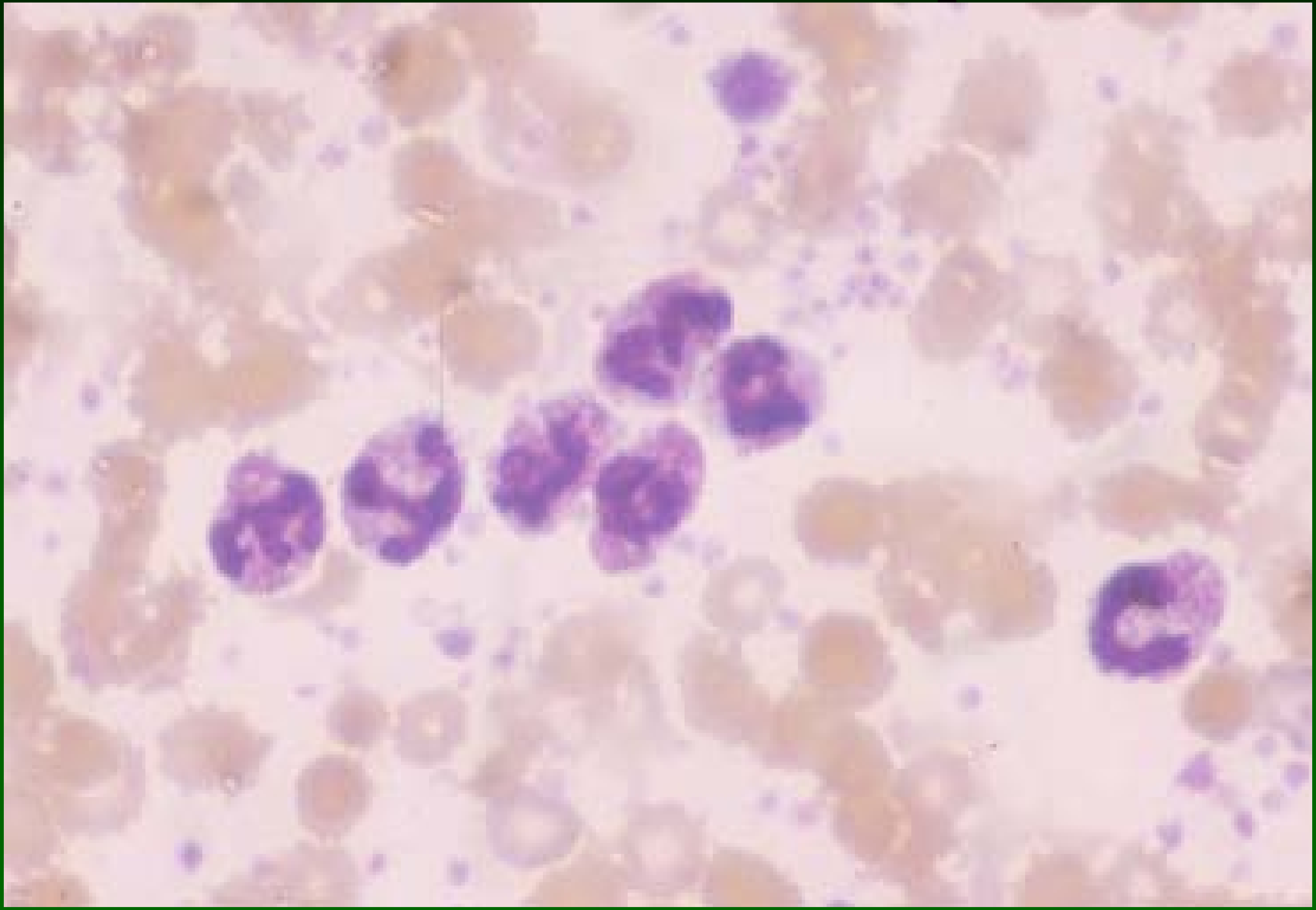


White Tail Deer- sickled erythrocytes & typical lymphocyte

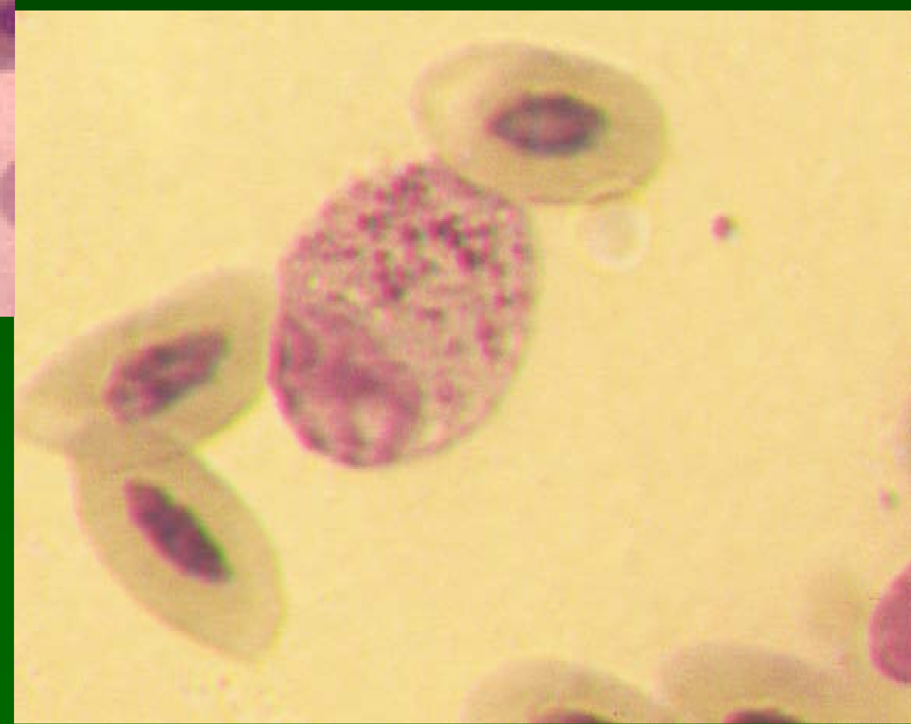
Black bear segs

Rabbit heterophils

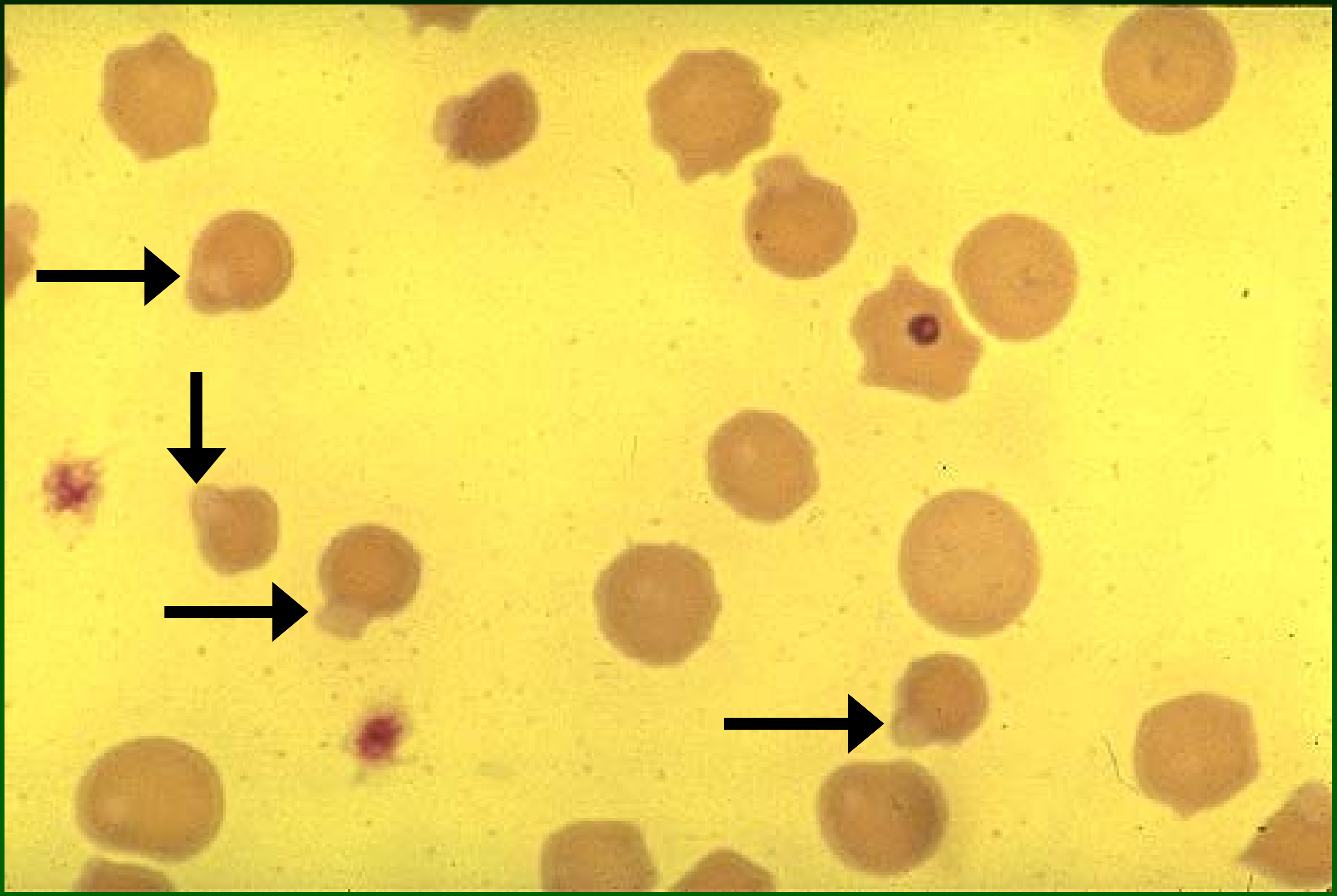


Can id toxic cells

Toxic heterophils

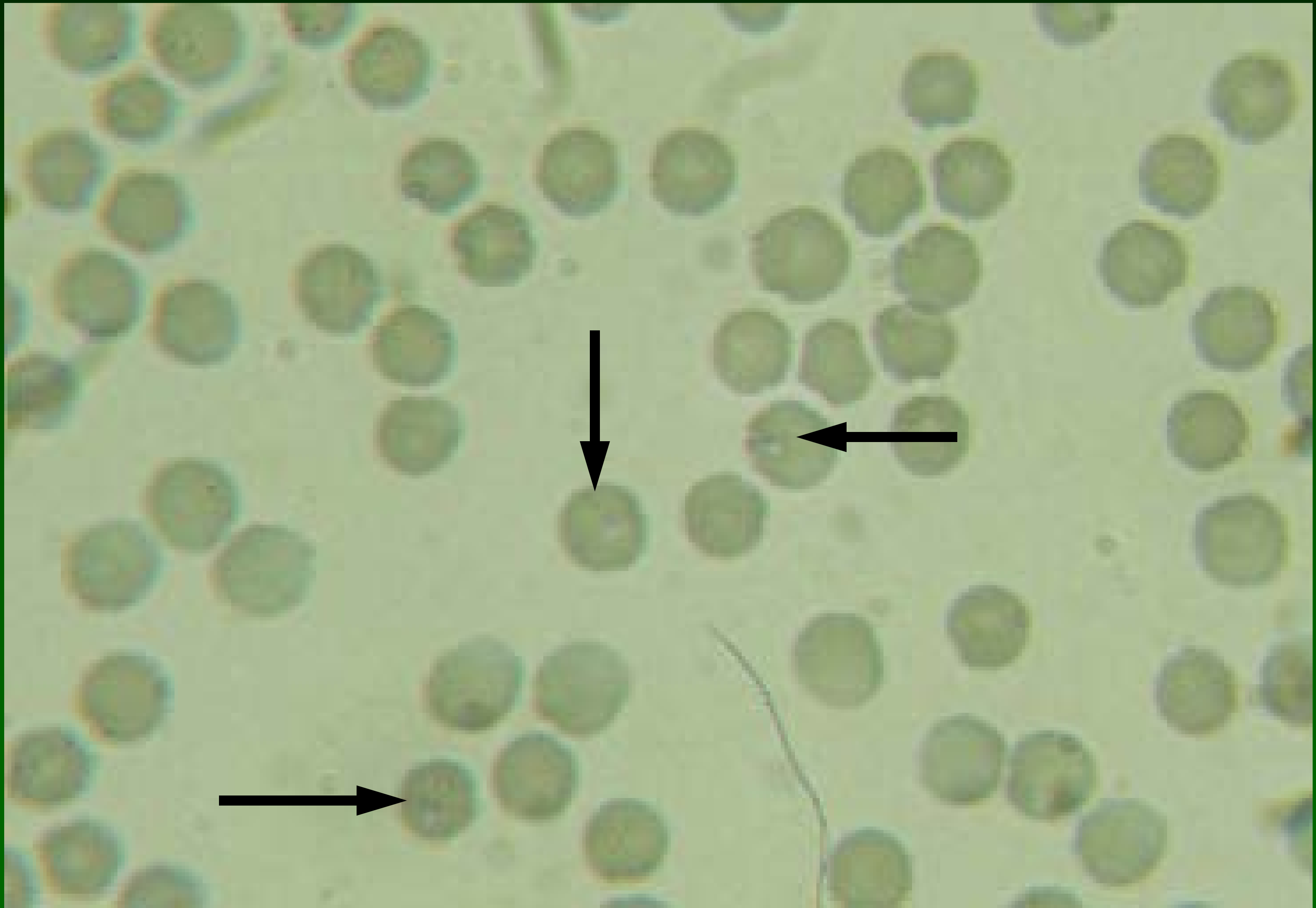


Heinz bodies

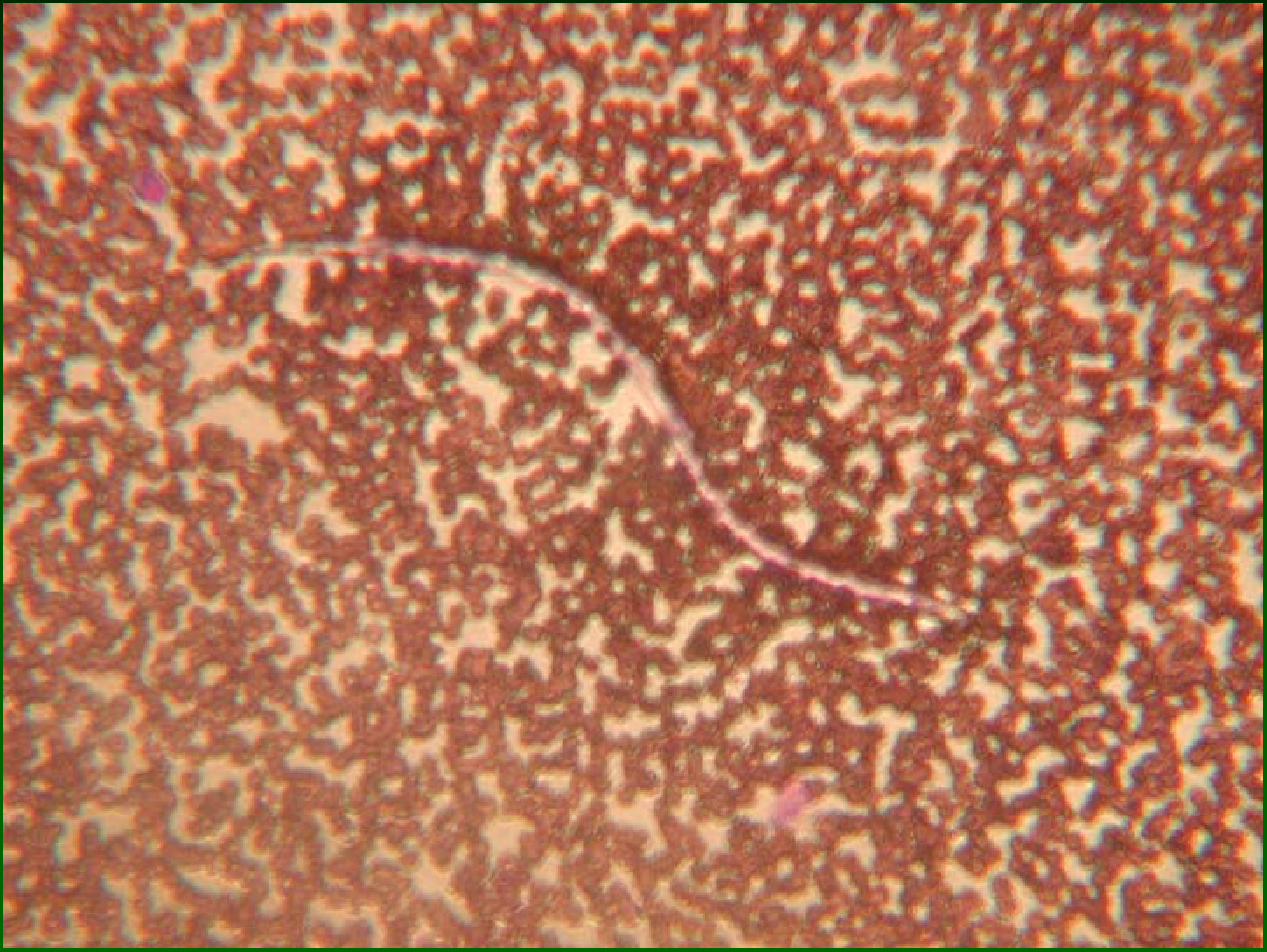


Due to toxicity

Cytauxzoan felis

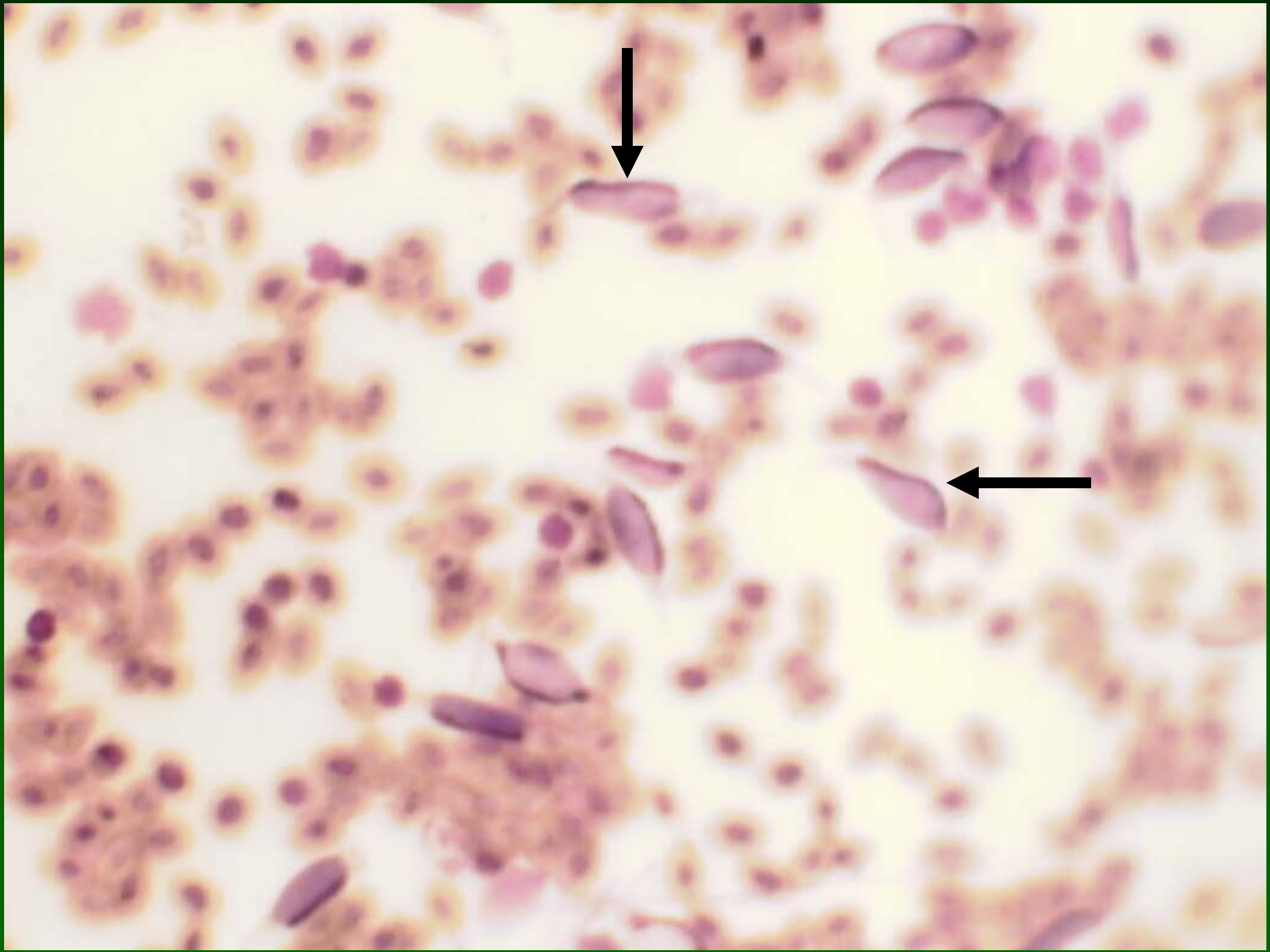


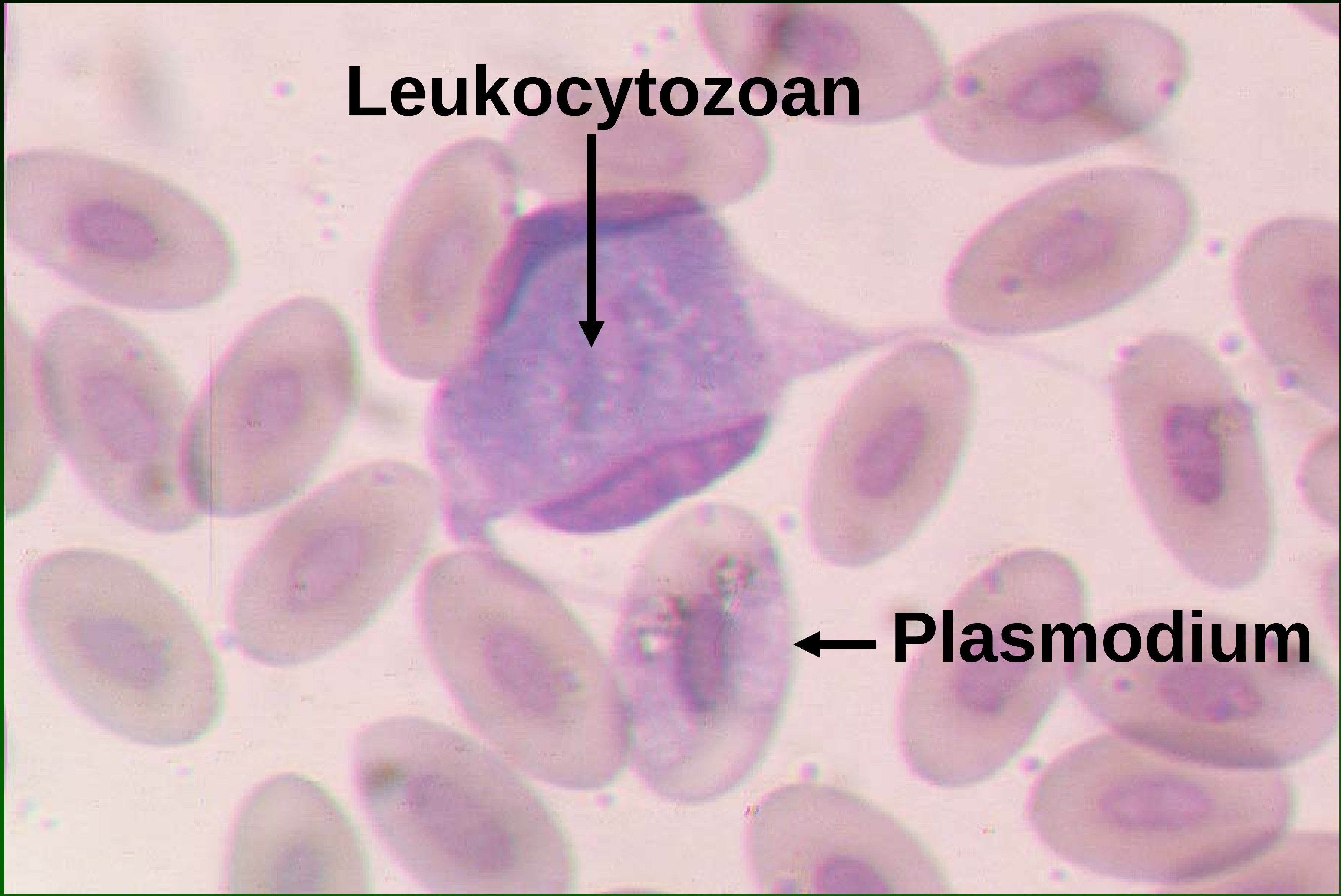
Microfilaria

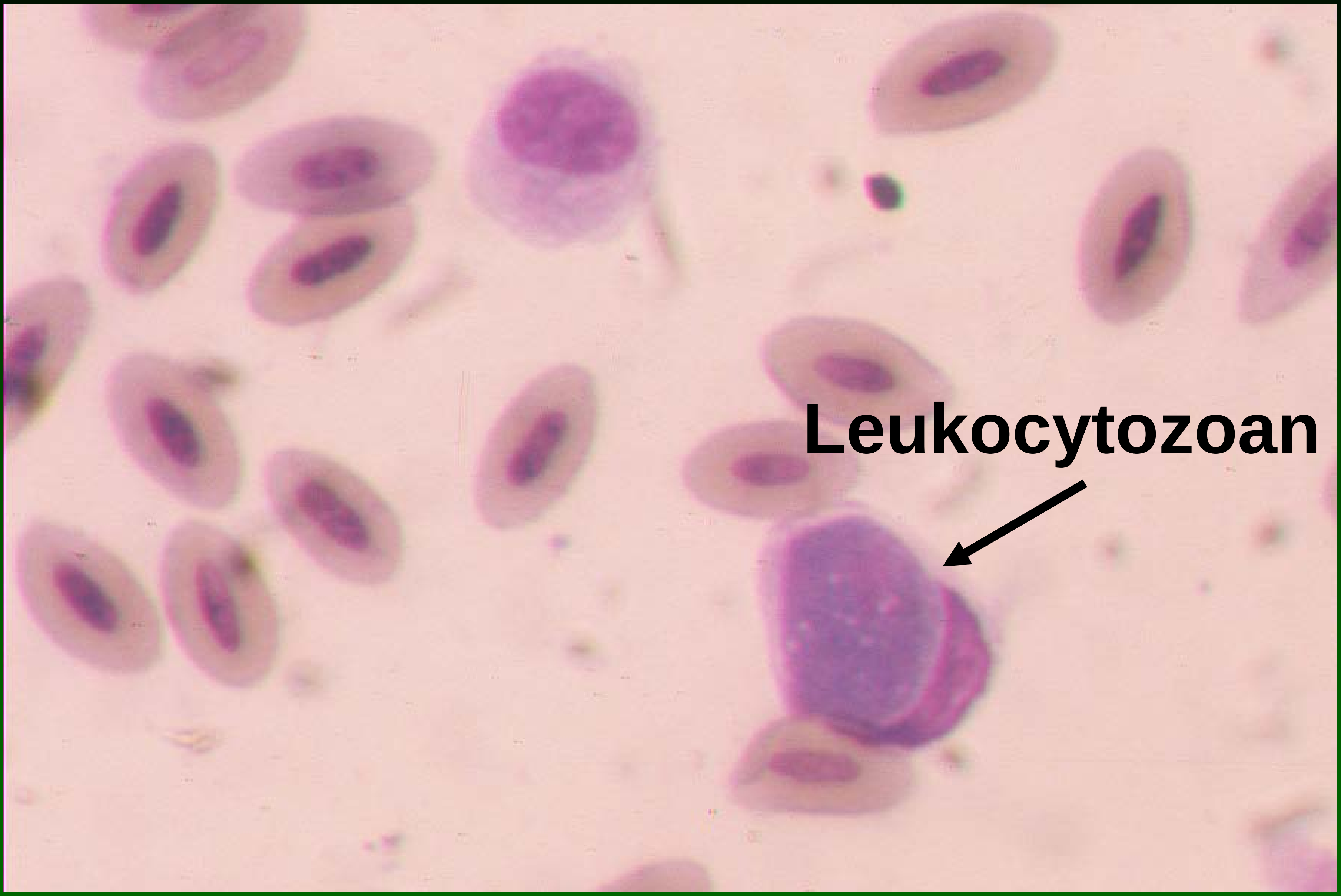


Trypanosoma cruzi

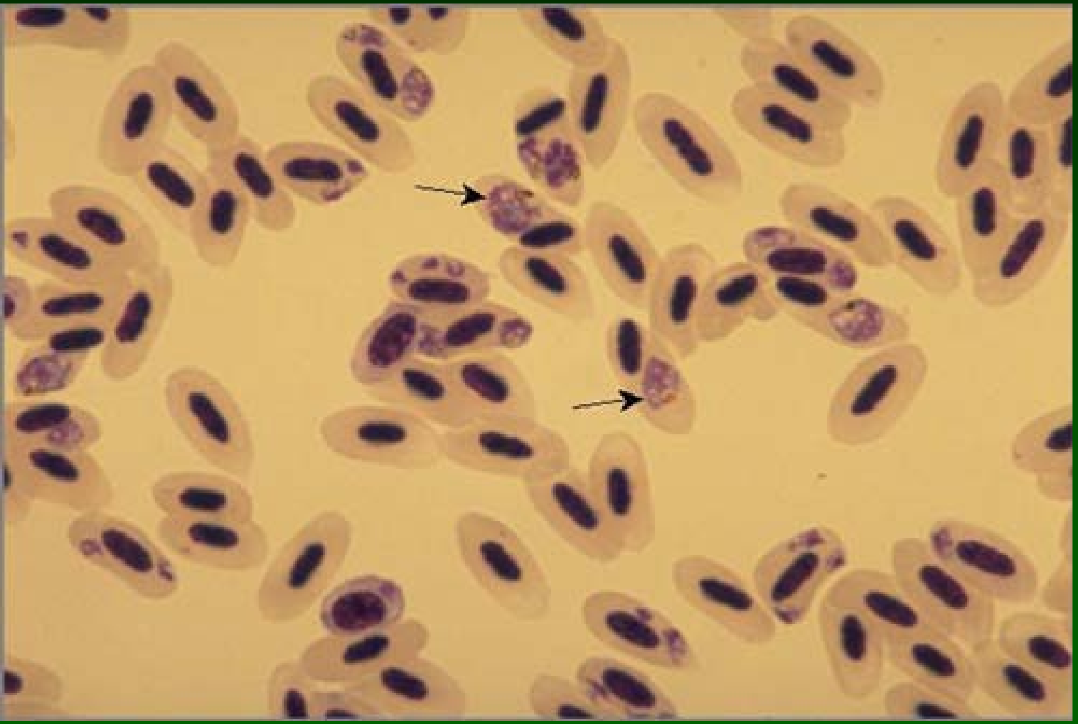
Leukocytozoan 200x

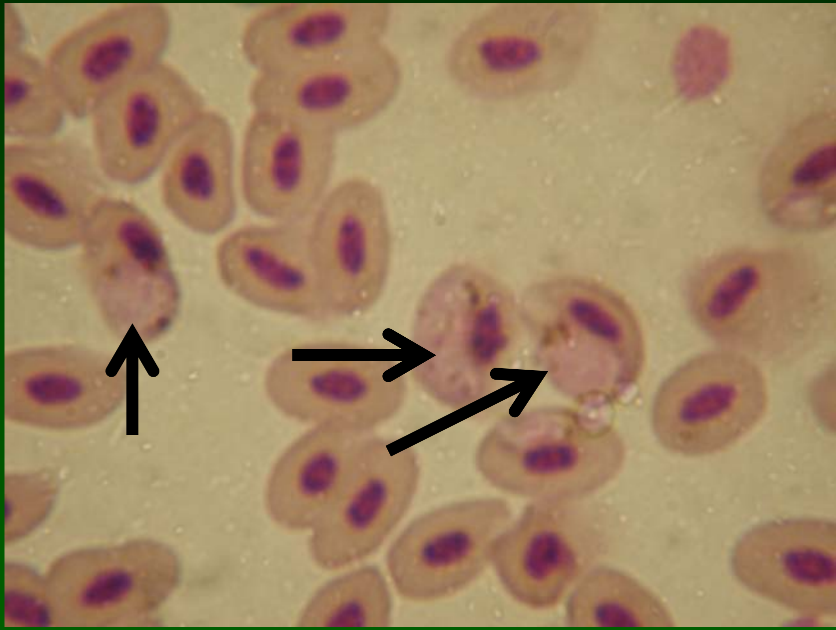



Leukocytozoan
Plasmodium


Leukocytozoan

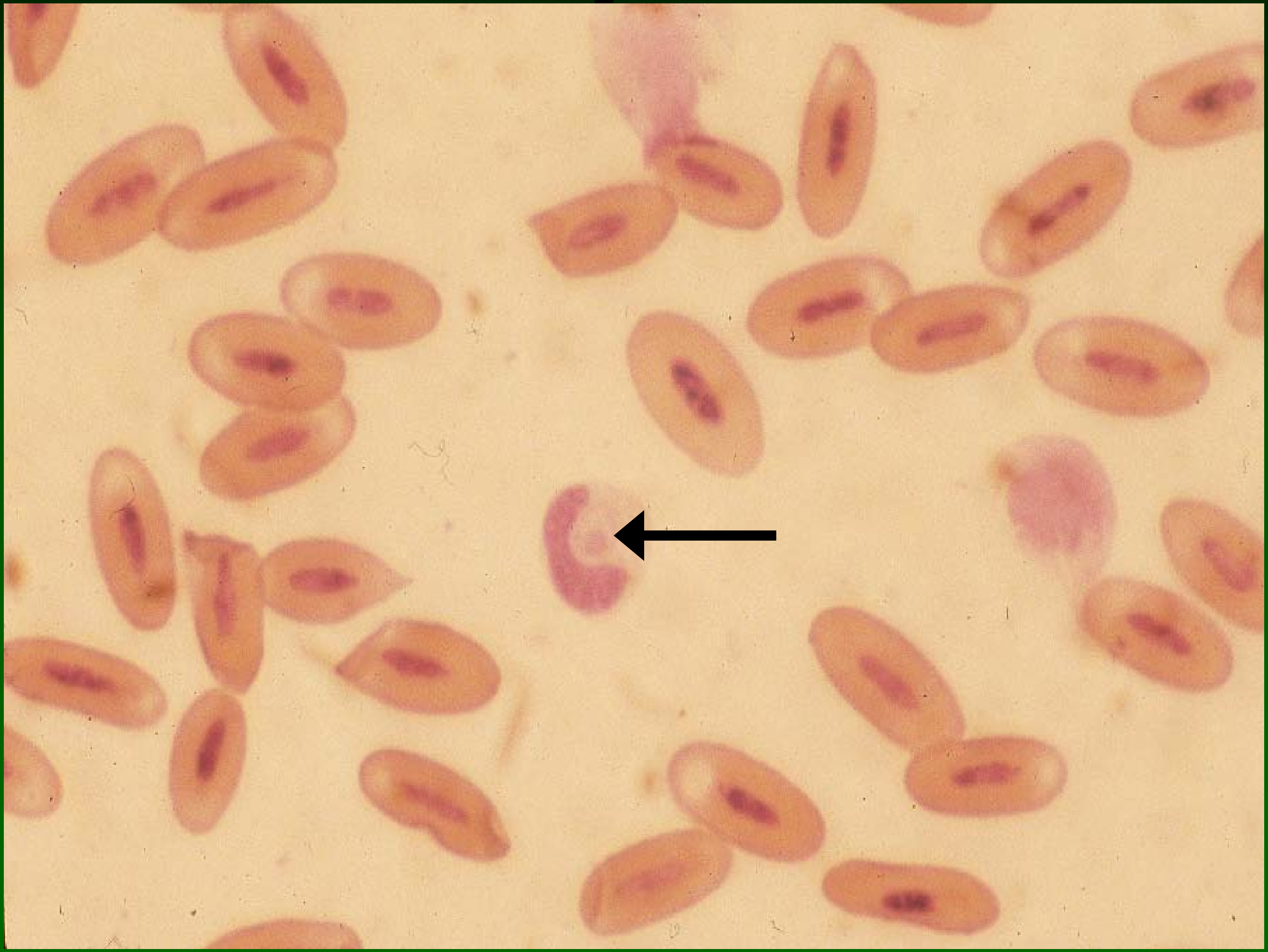
Plasmodium





Hemoproteus

Atoxoplasma



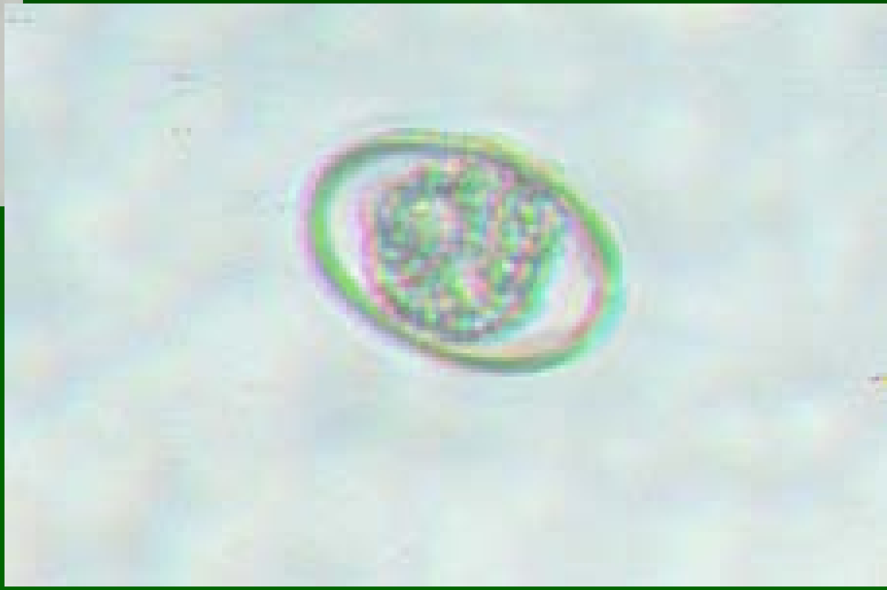
Fecal examinations

Direct smear, flotation, centrifugation

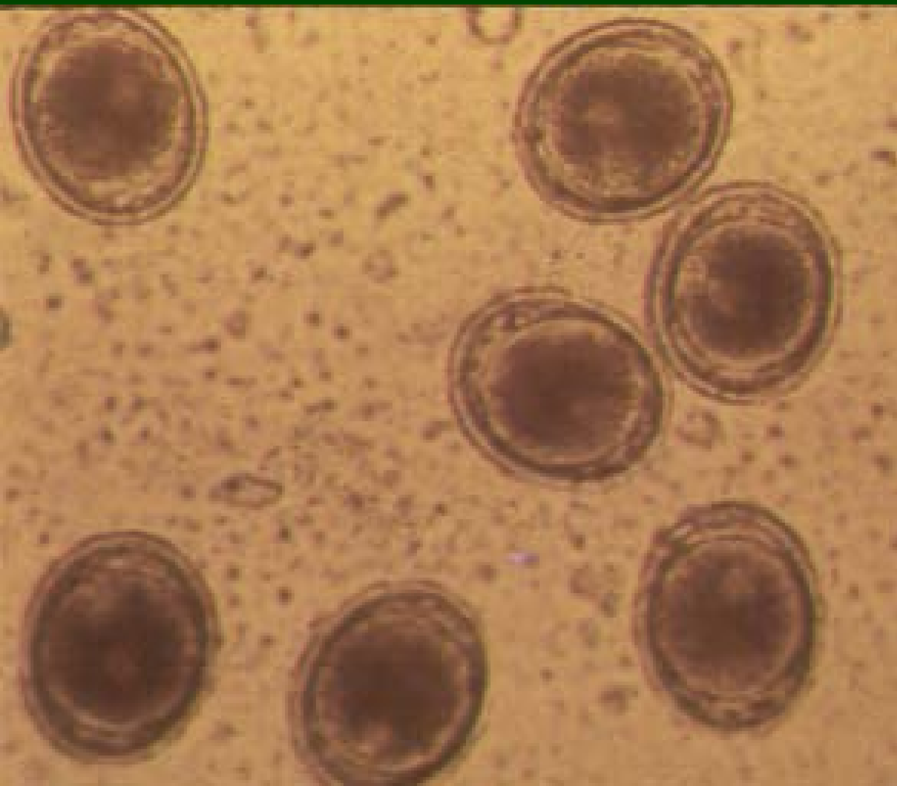
Mammal parasites

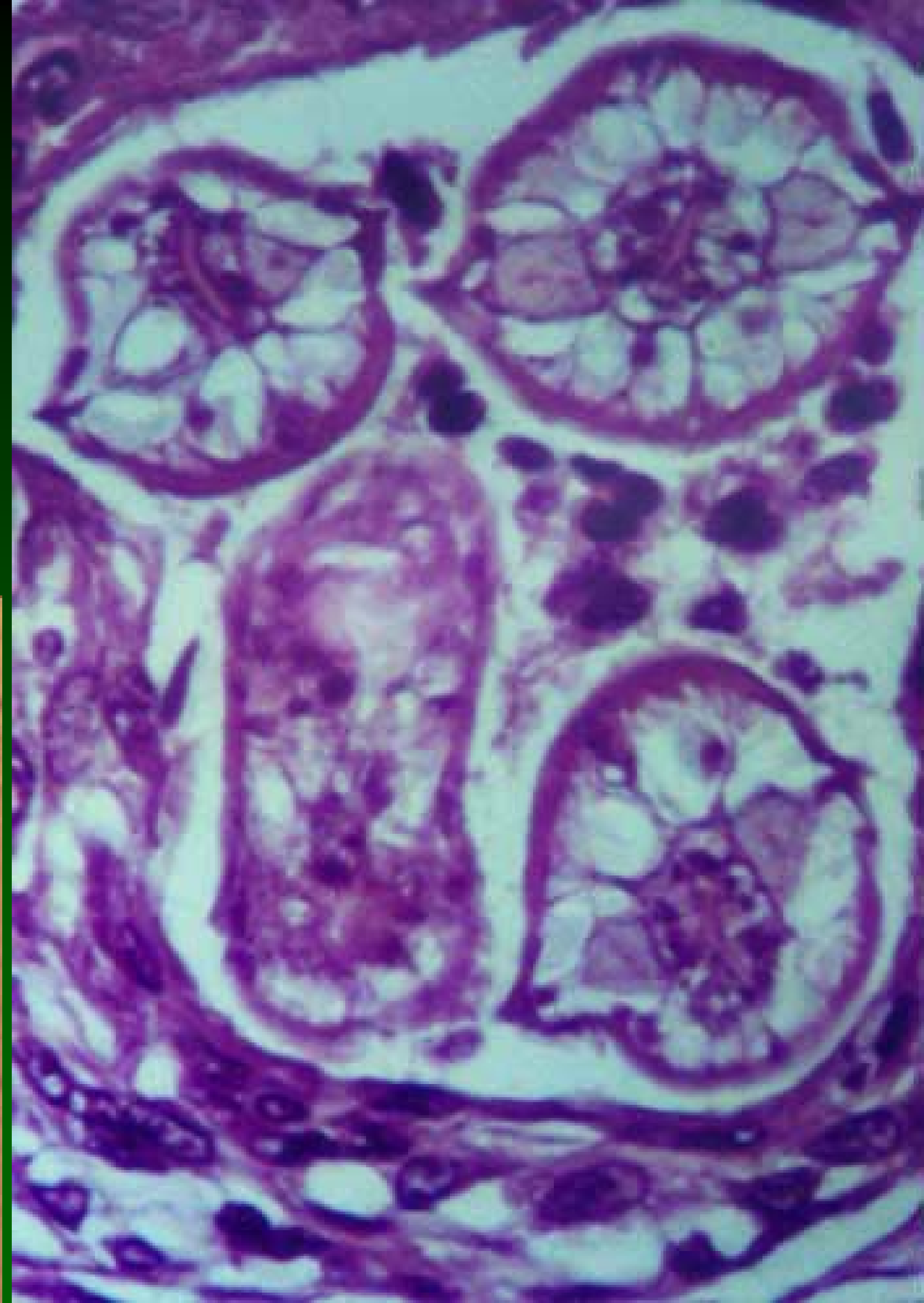
Coccidia

Ascarid



Baylisascaris procyonis

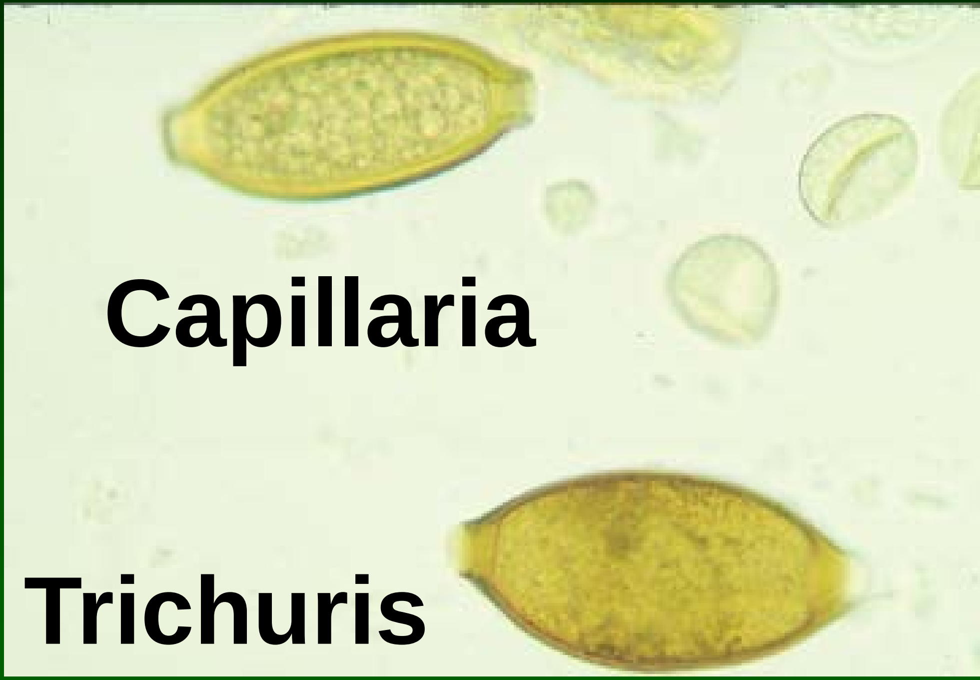




Hookworm ova- 250x

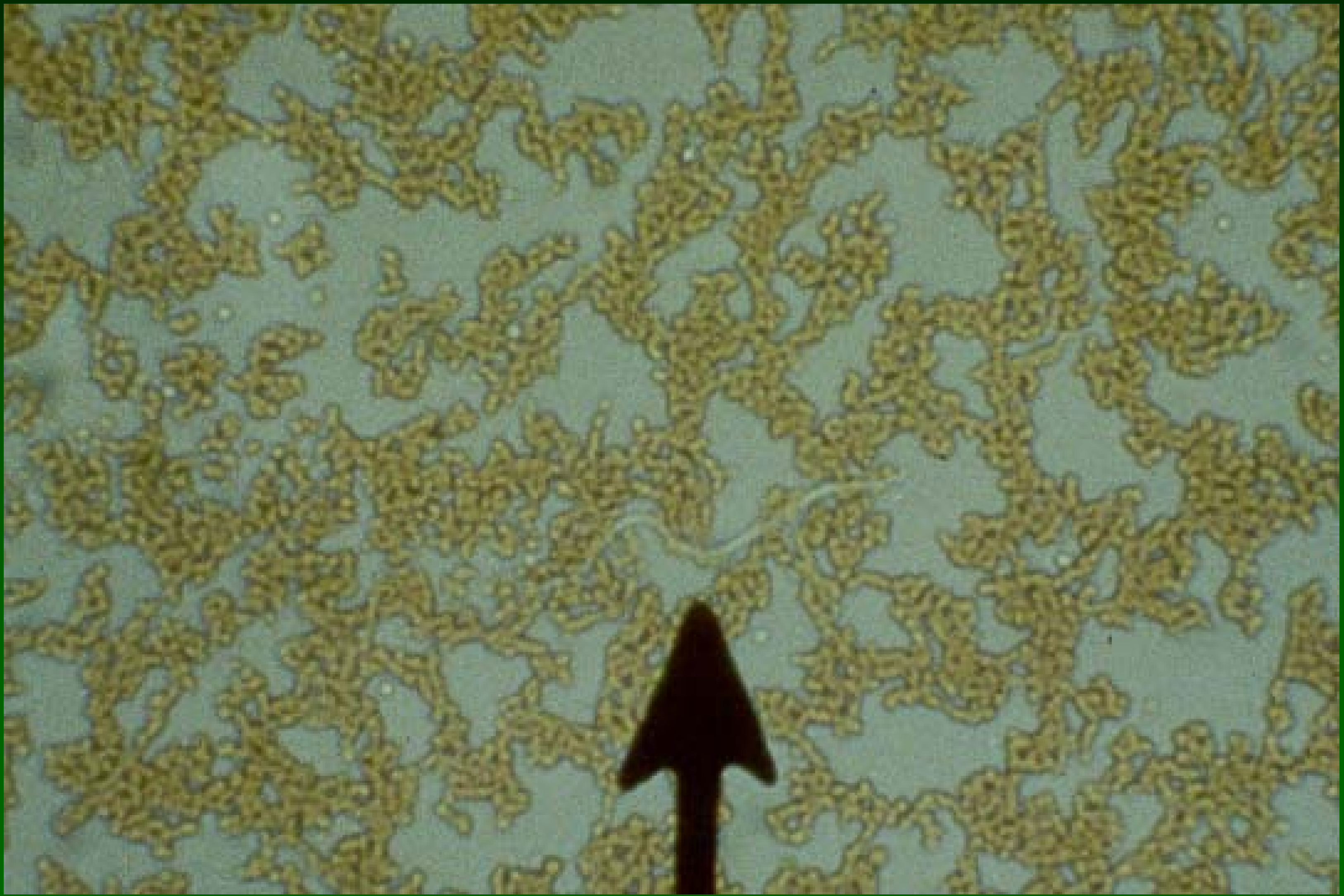
Lungworms

Aelurostrongylus from a fox



raccoon

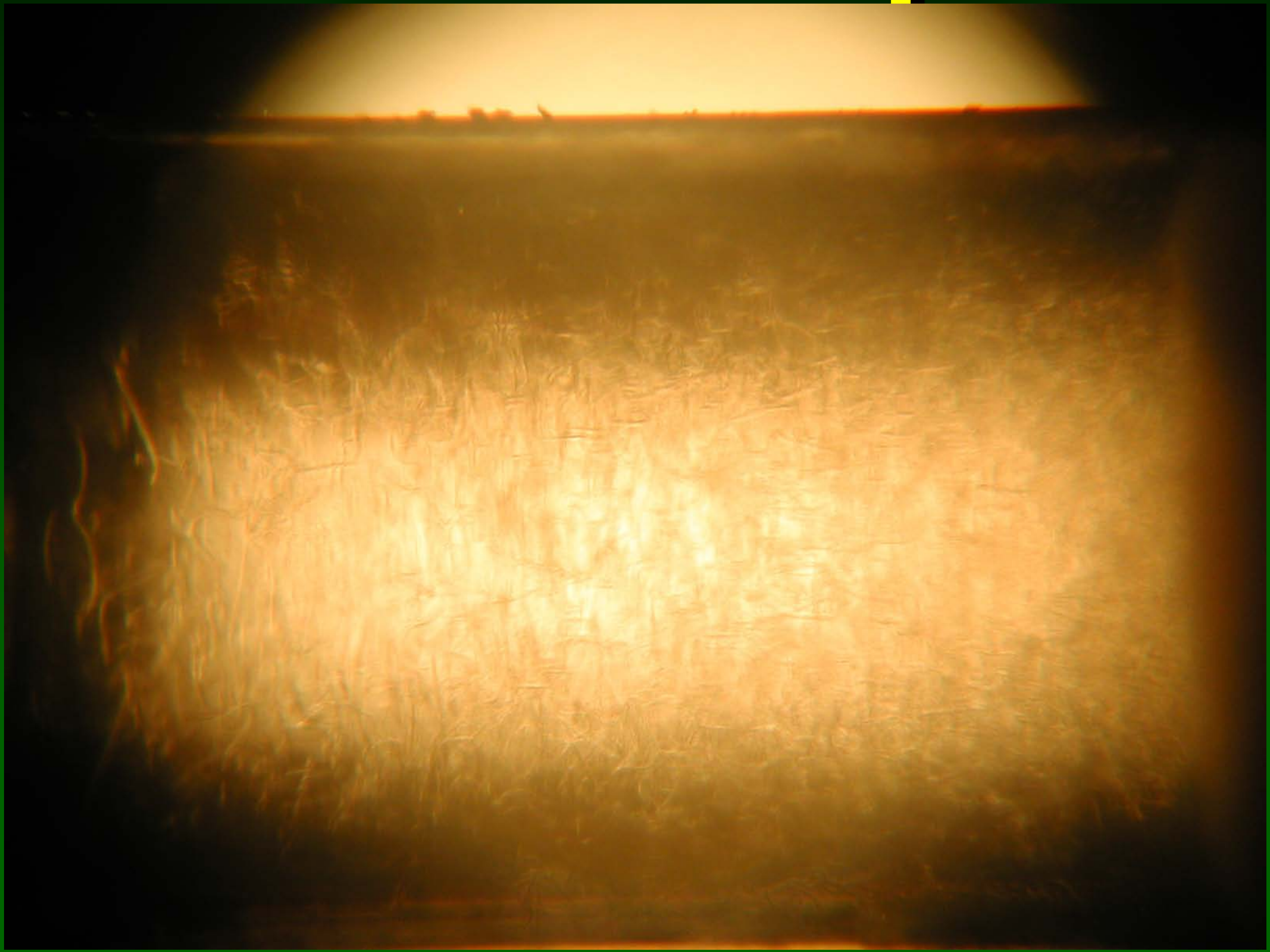
Microfilaria unstained



Microfilaria w/ DQ

Microfilaria in plasma

Microfilaria in plasma



Flukes and tapes

Alaria from jaguarundi

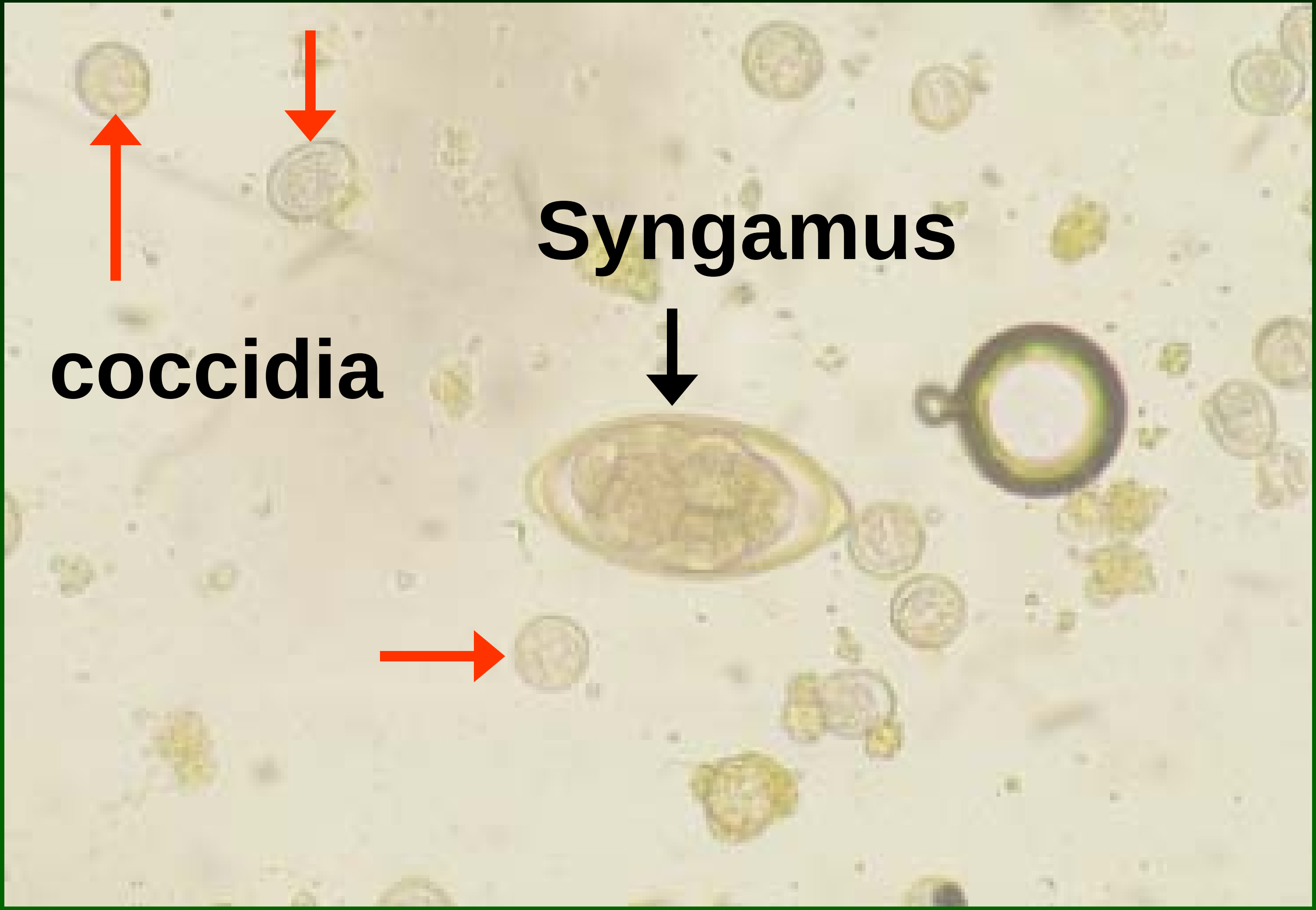
Moniezia from deer

Isospora caninum

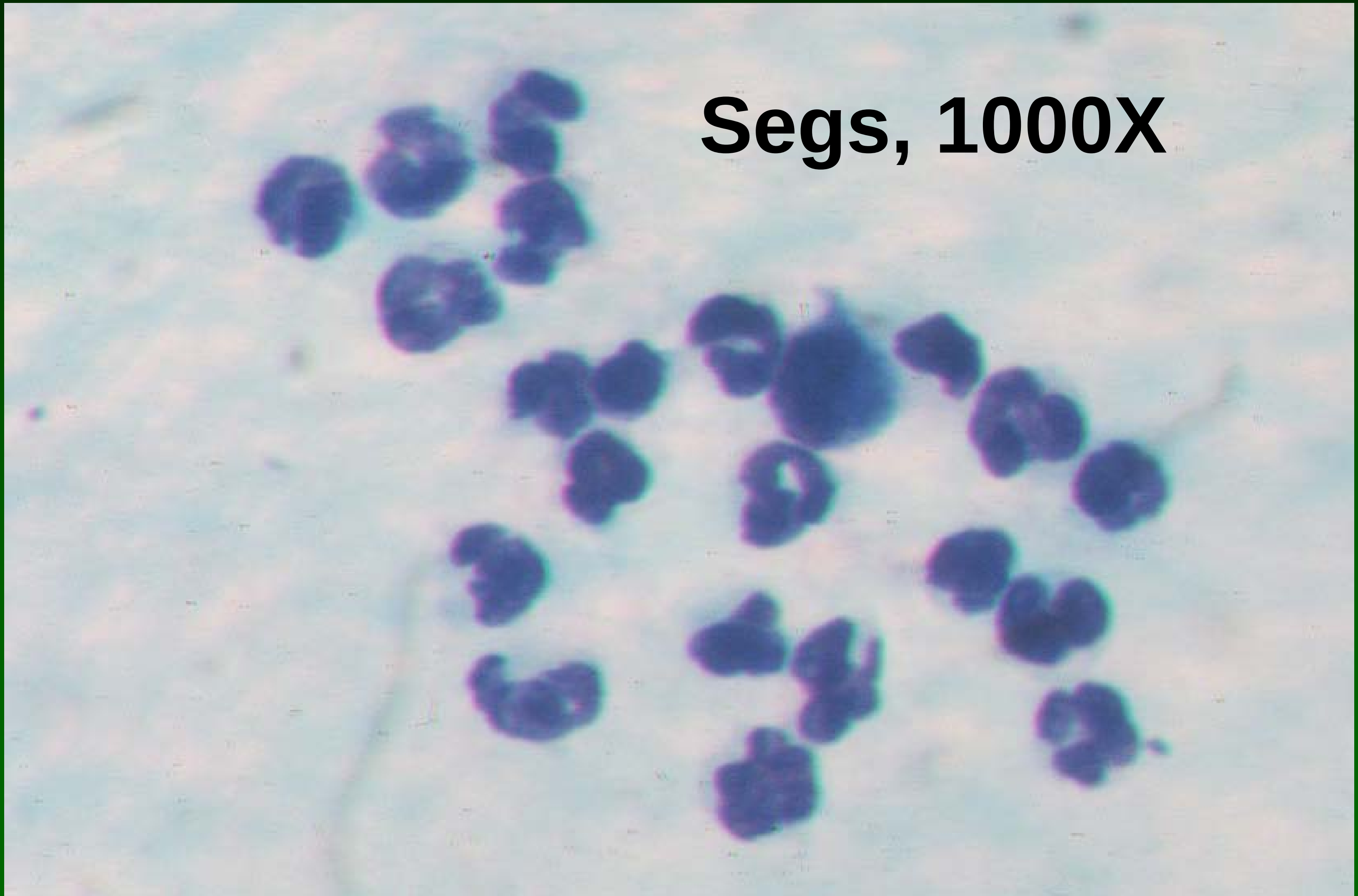
Sporulated oocyst

Avian GI parasites

Parasites in birds



Urine Sediment



Cytology

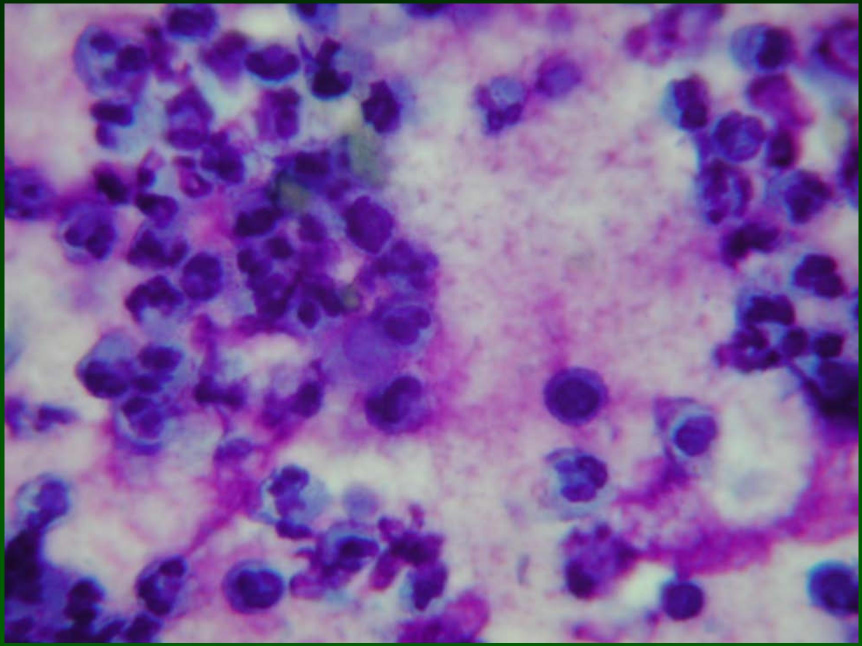
- Needle aspirate
- Touch tissue to slide
- Swab tissue
- Flush tissue
- Touch slide to lesion
- Discharge to slide

Specific procedures

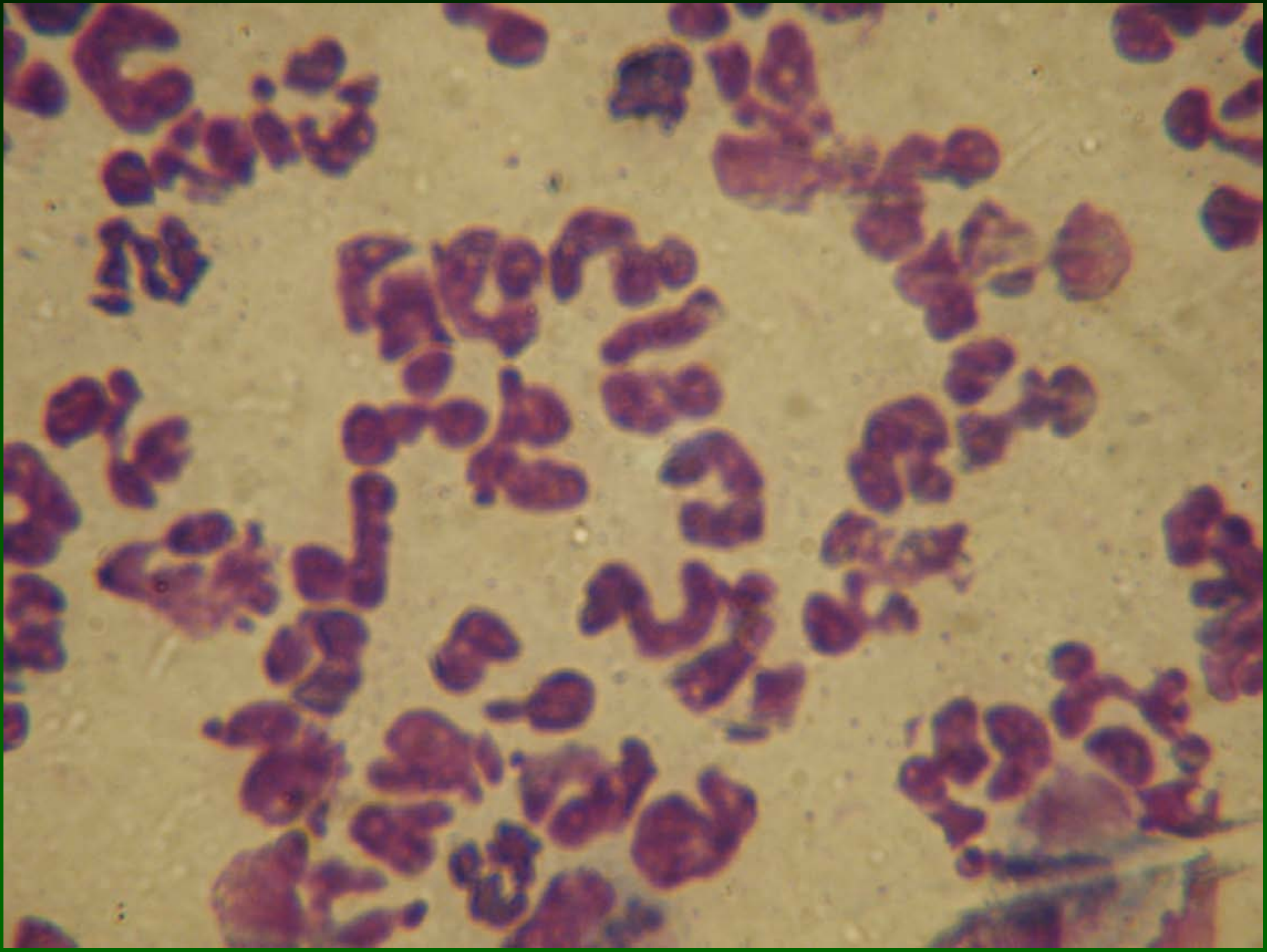
- Transtracheal wash
- Cystocentesis
- Thoracocentesis
- Abdominocentesis
- Arthrocentesis
- Skin scraping

Acute inflammation

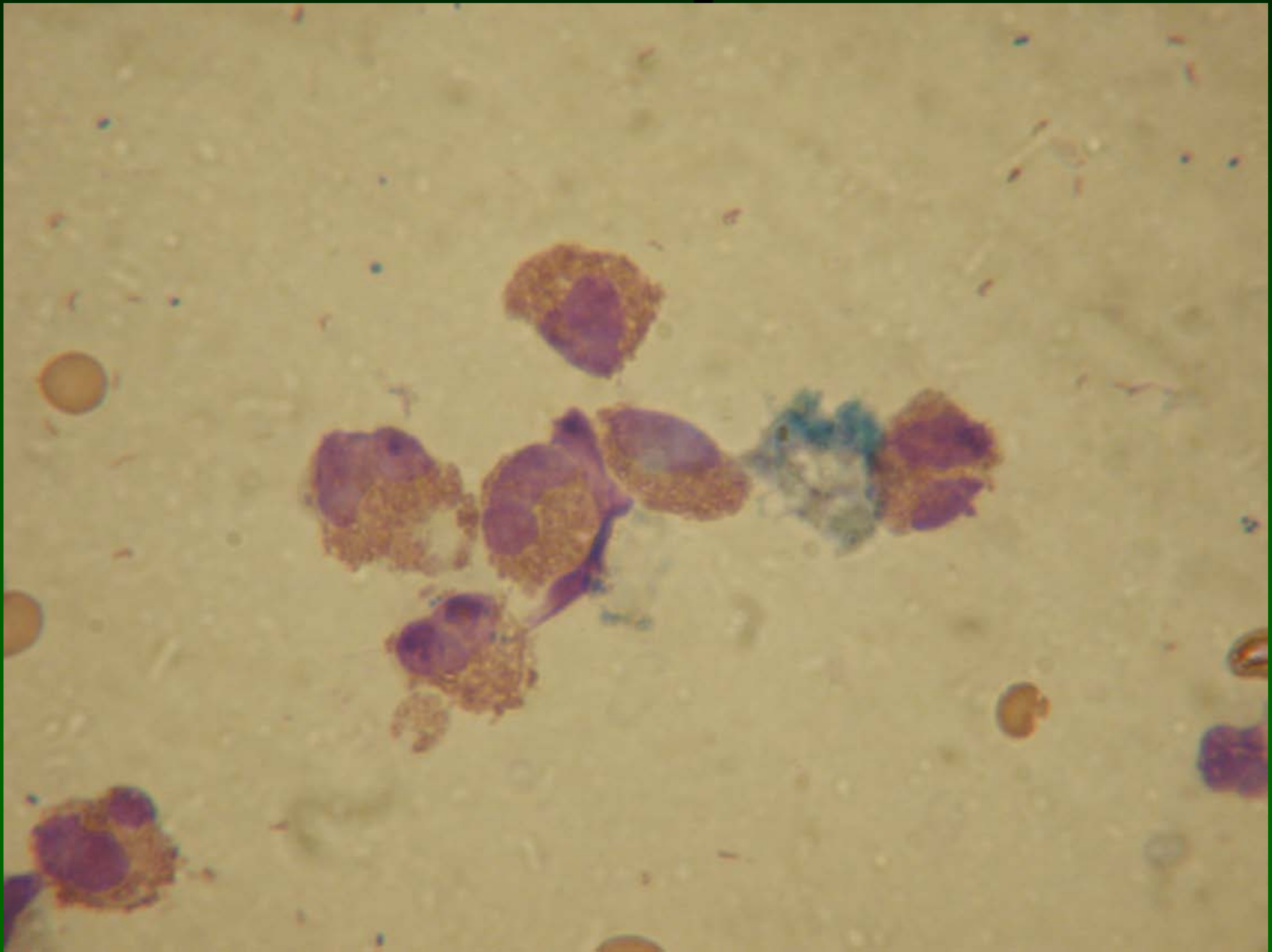
- When present look for organisms
  - Direct Gram stain



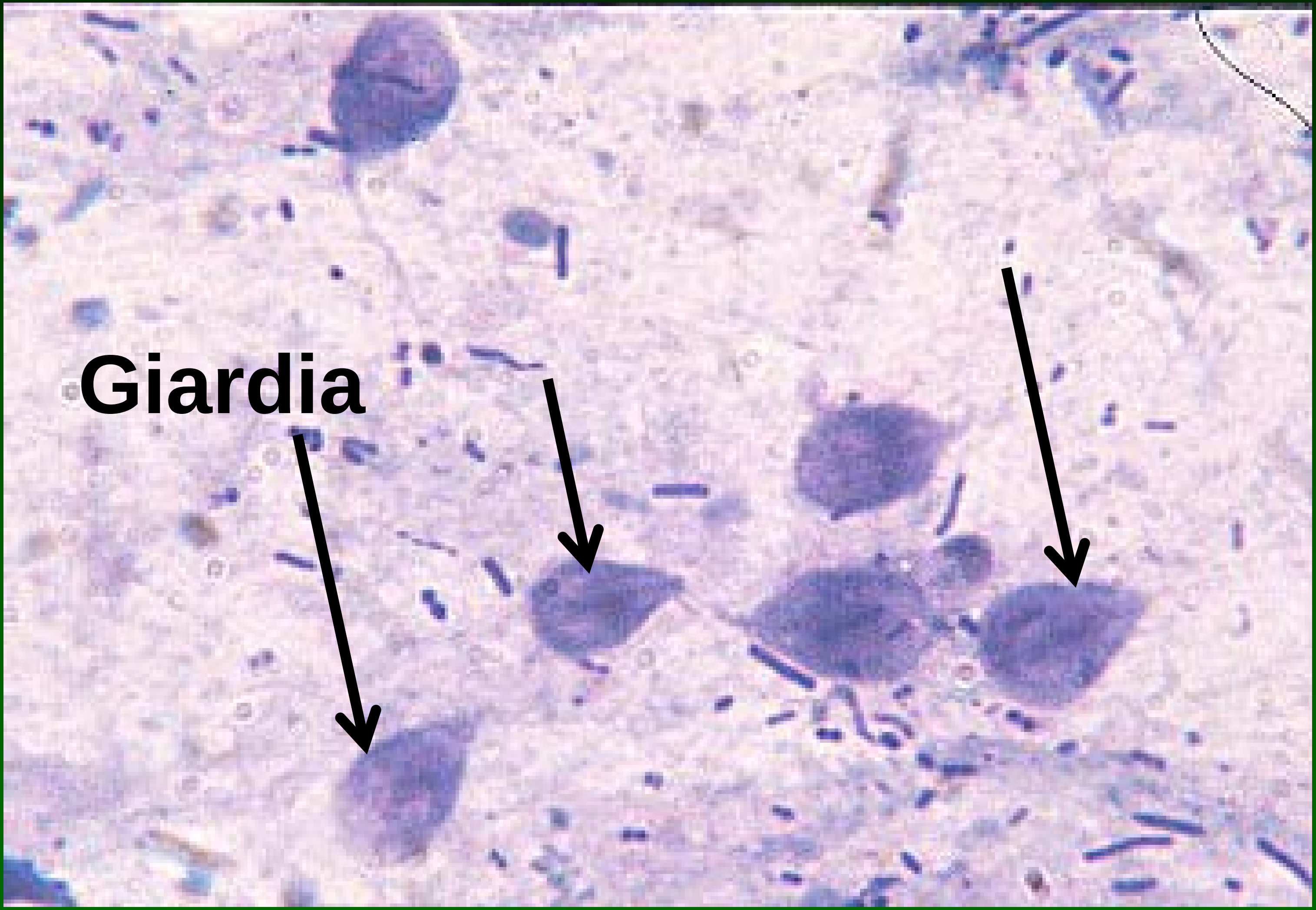
Abdominocentesis



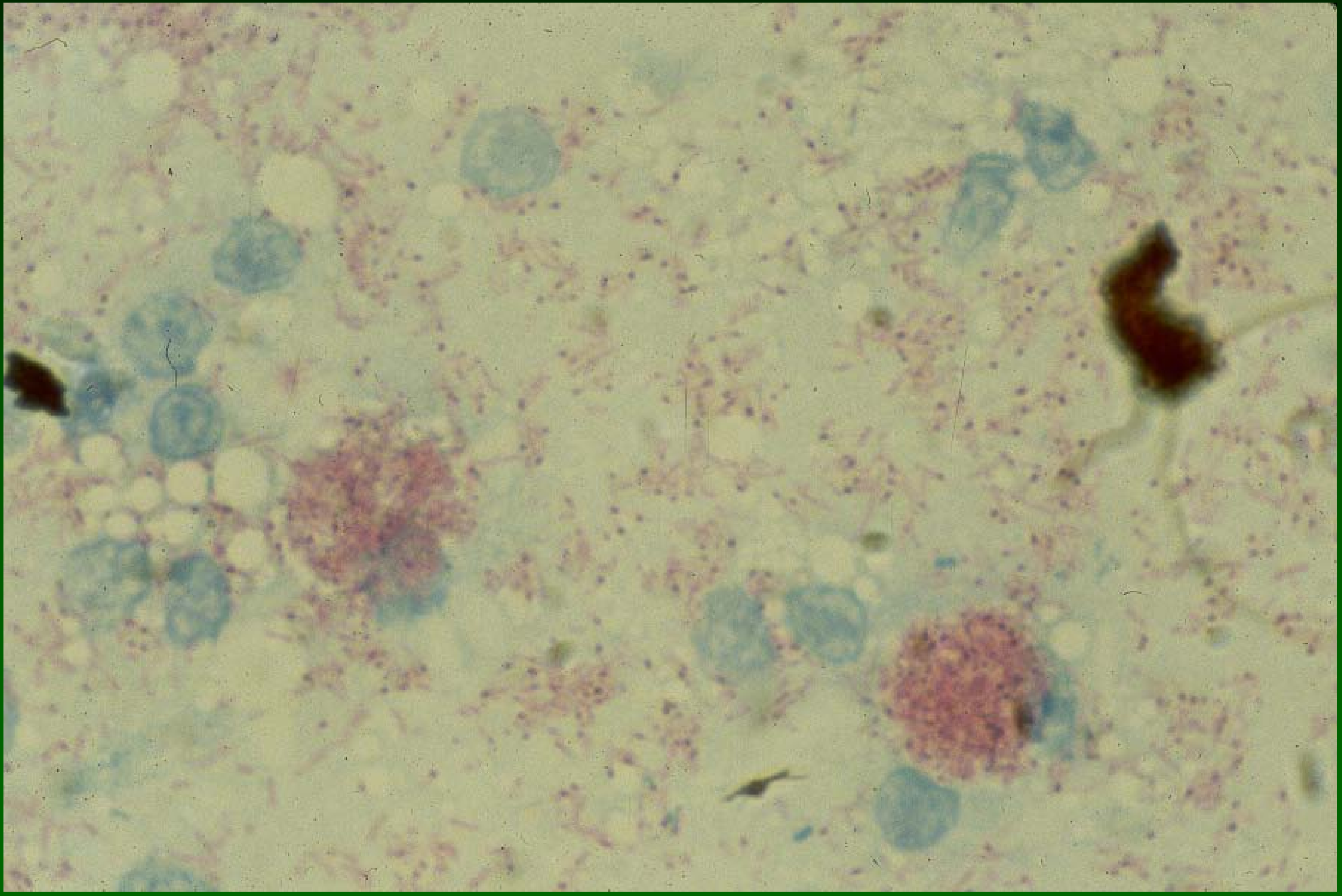
Eosinophils



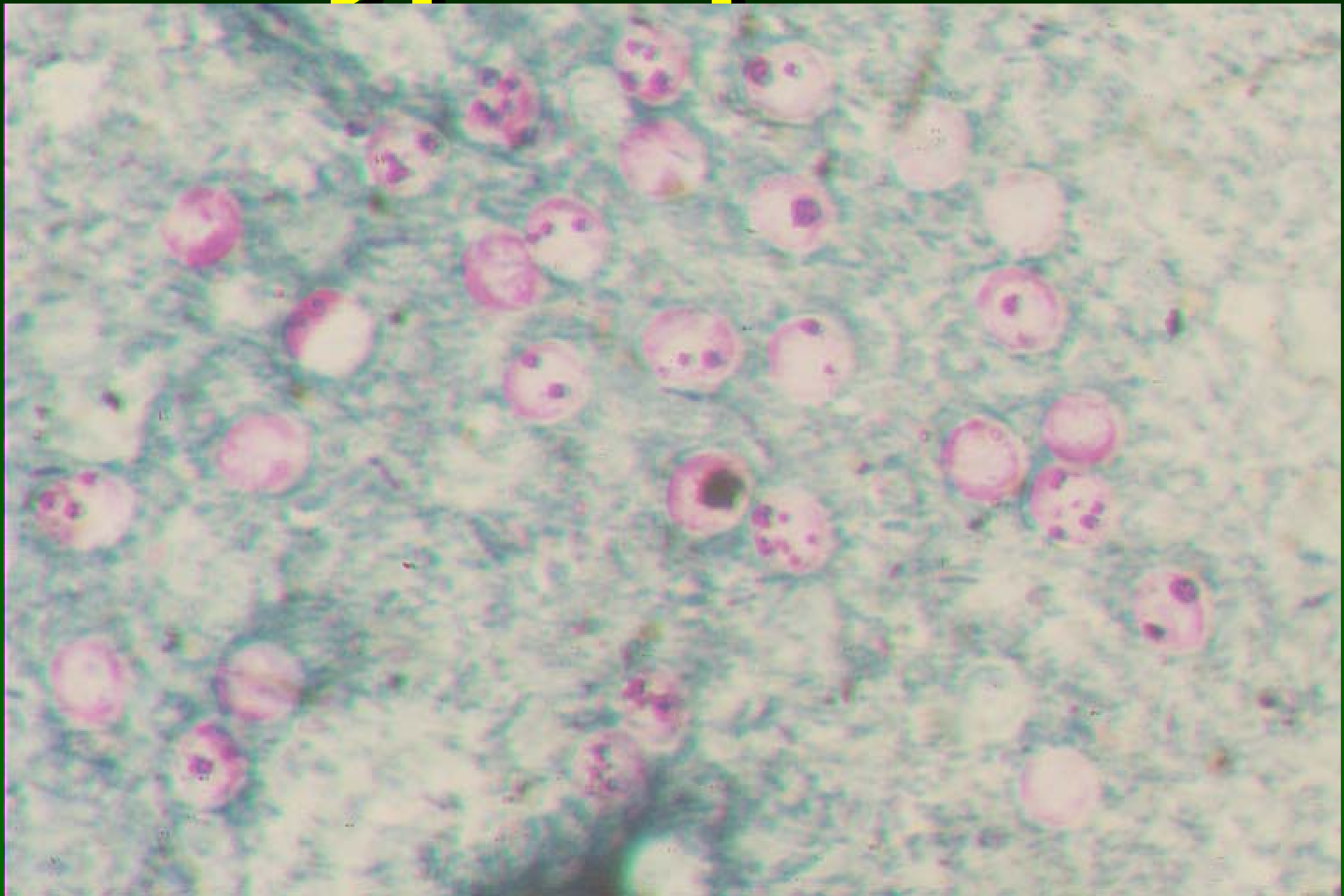
Stained fecal smear



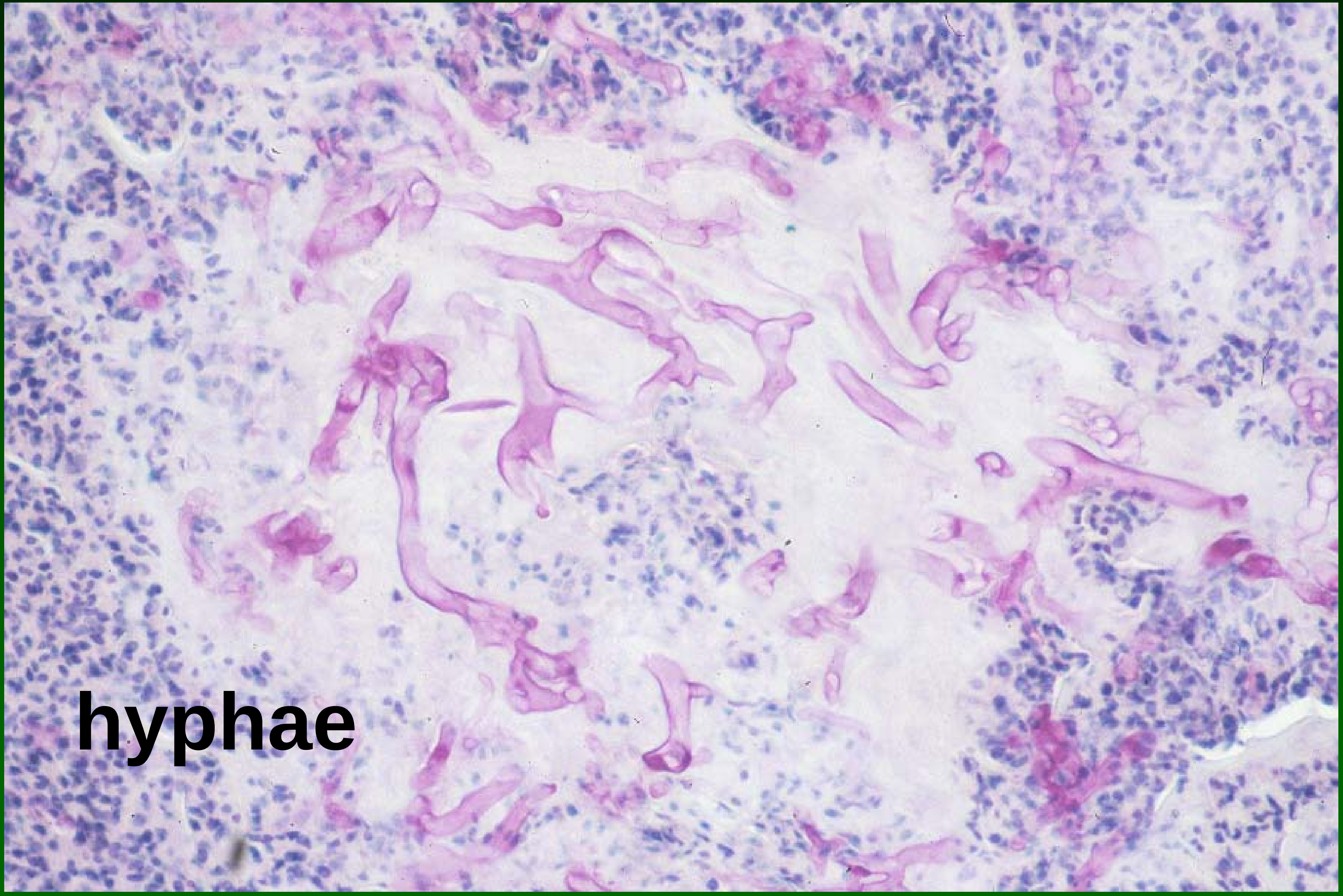
Acid fast stain



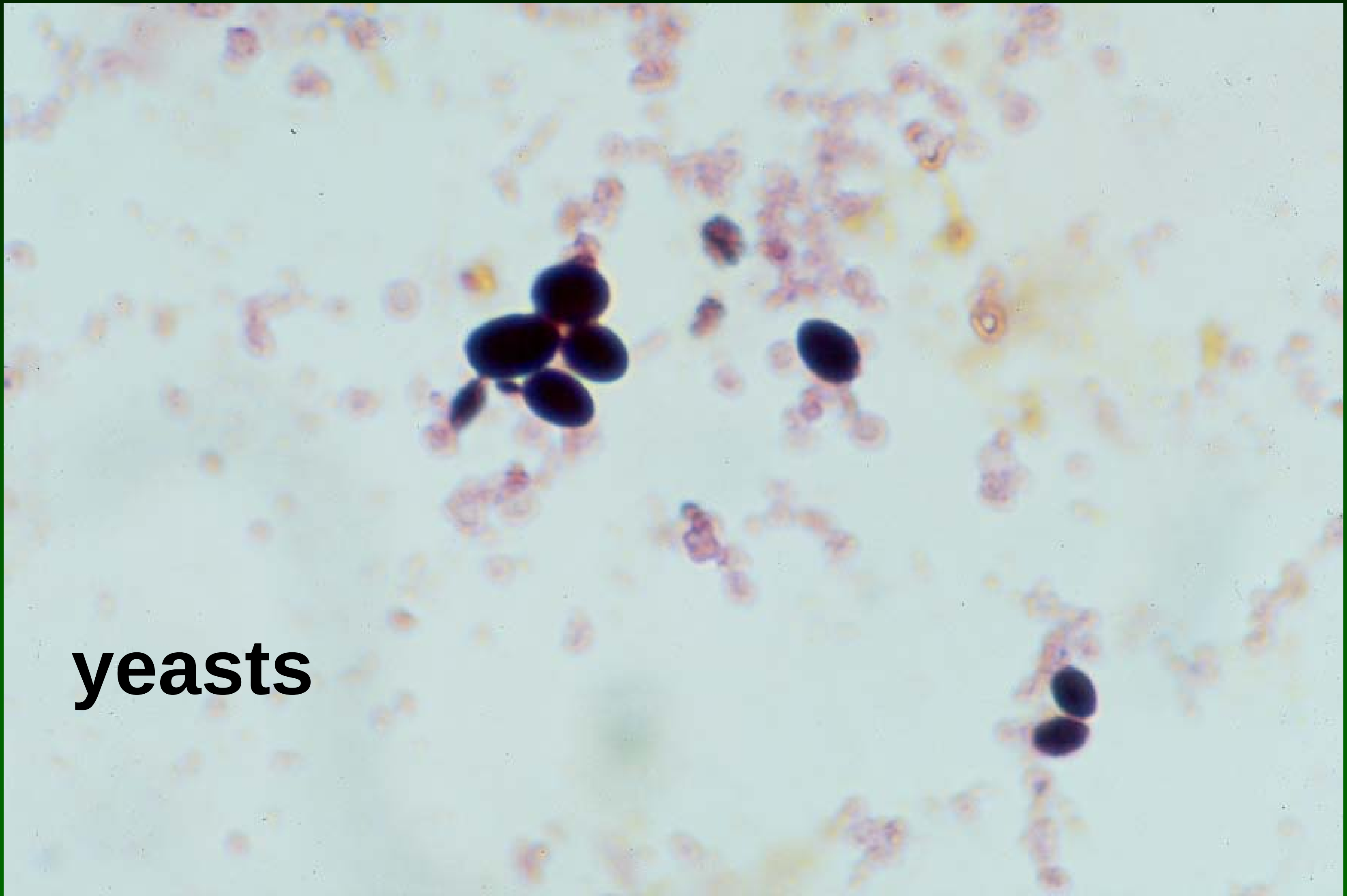
Acid fast stain-feces Cryptosporidia



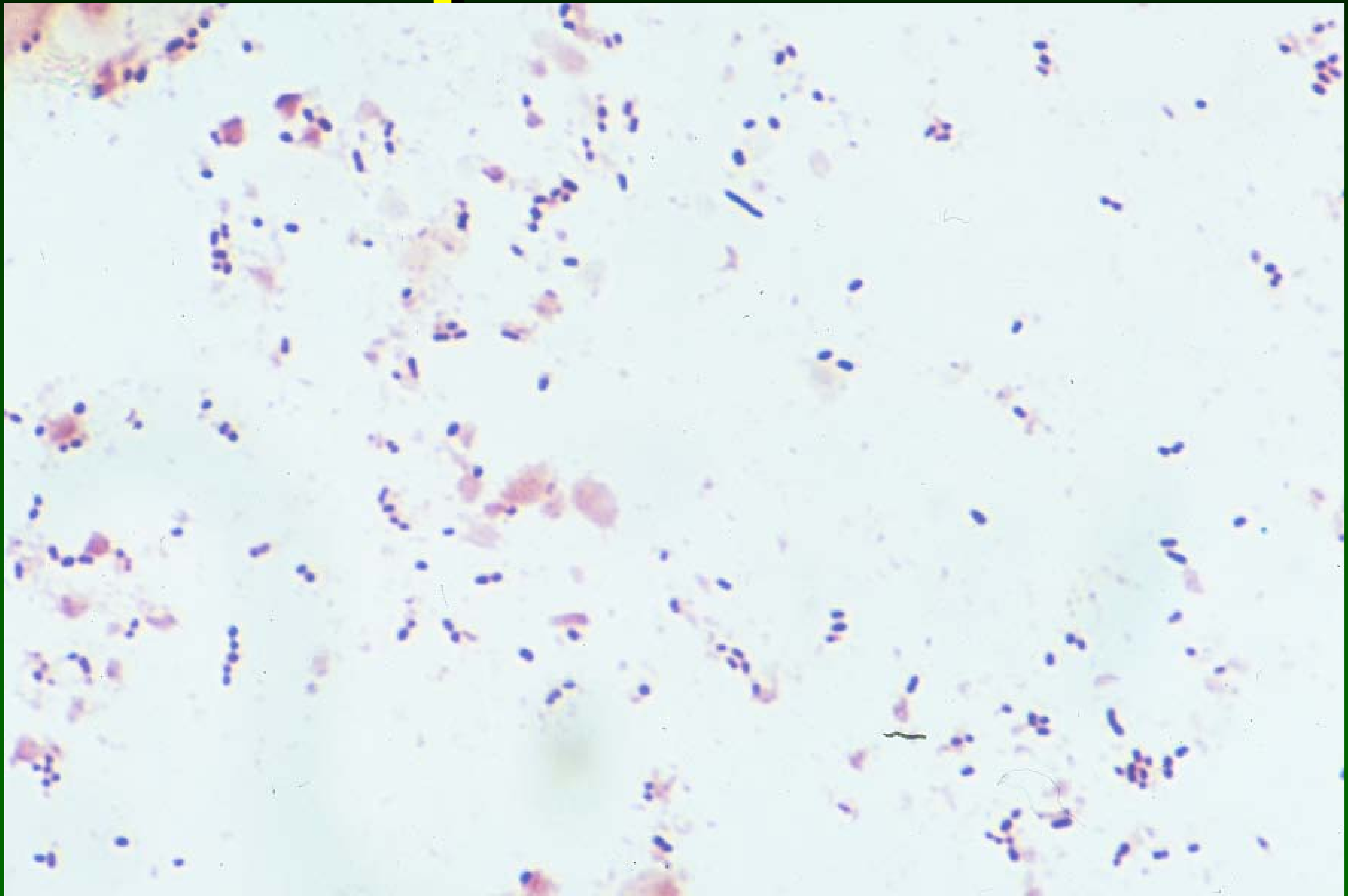
PAS stain



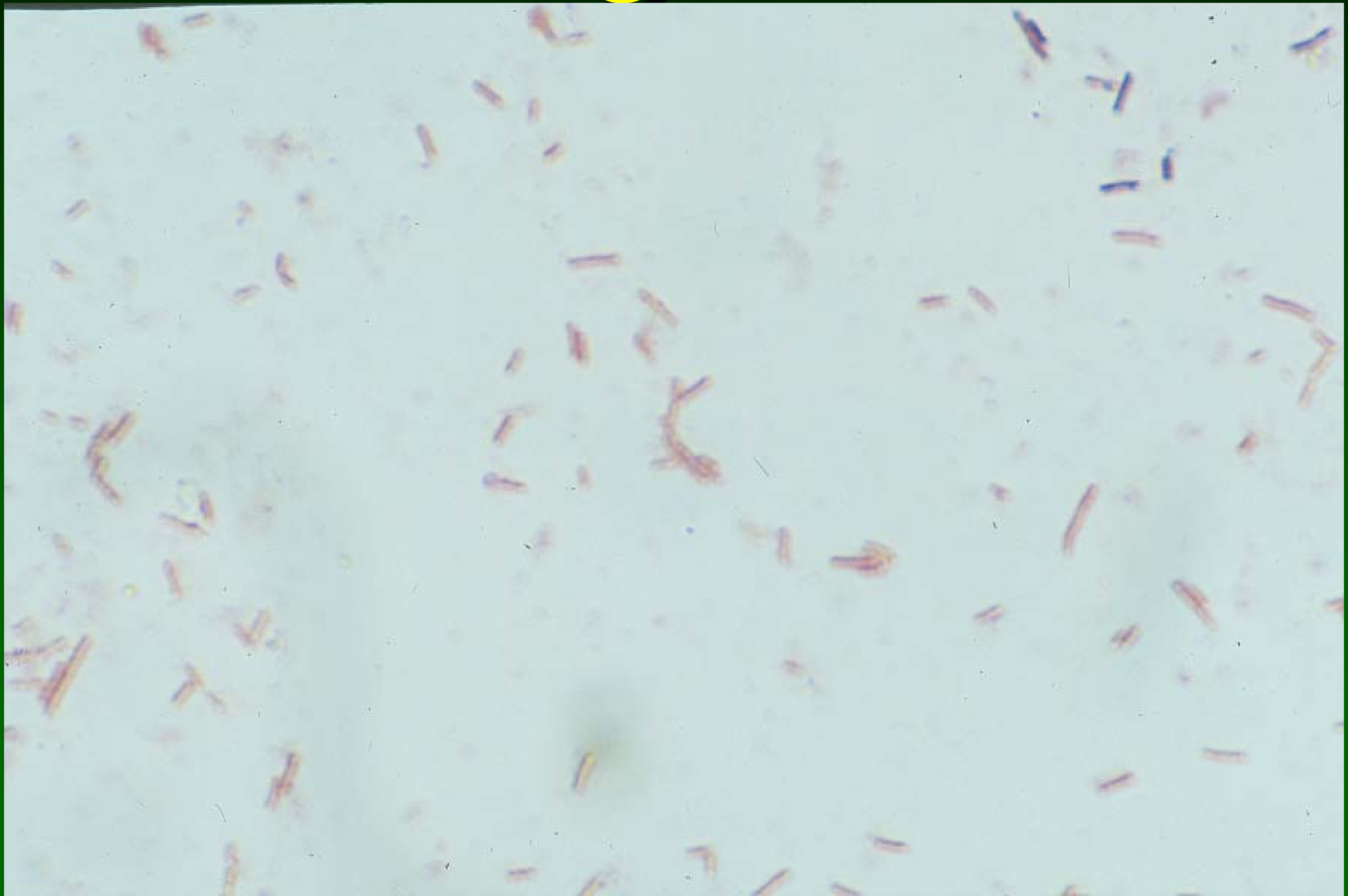
Gram stain



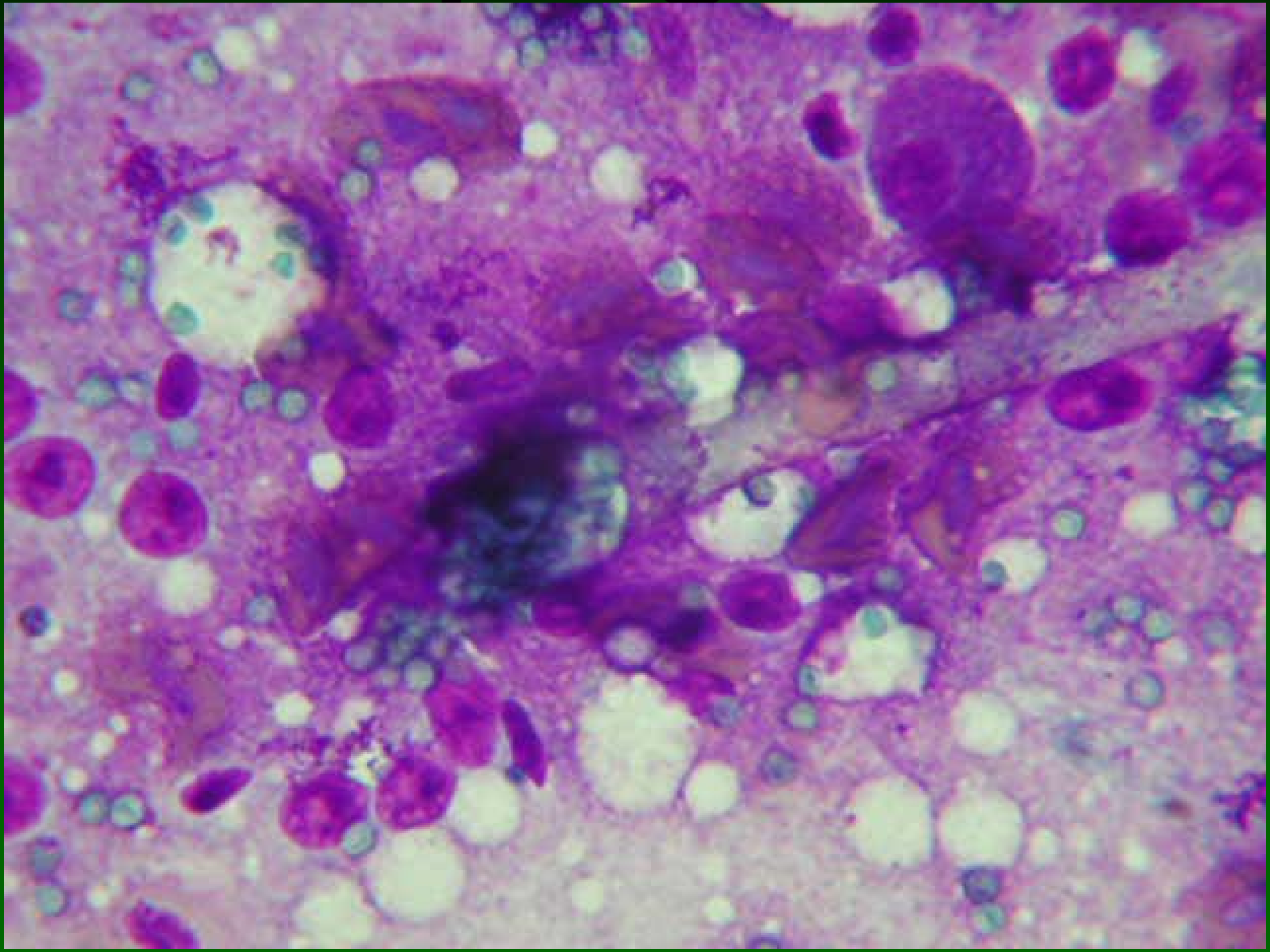
Gram positive cocci



Gram negative rods

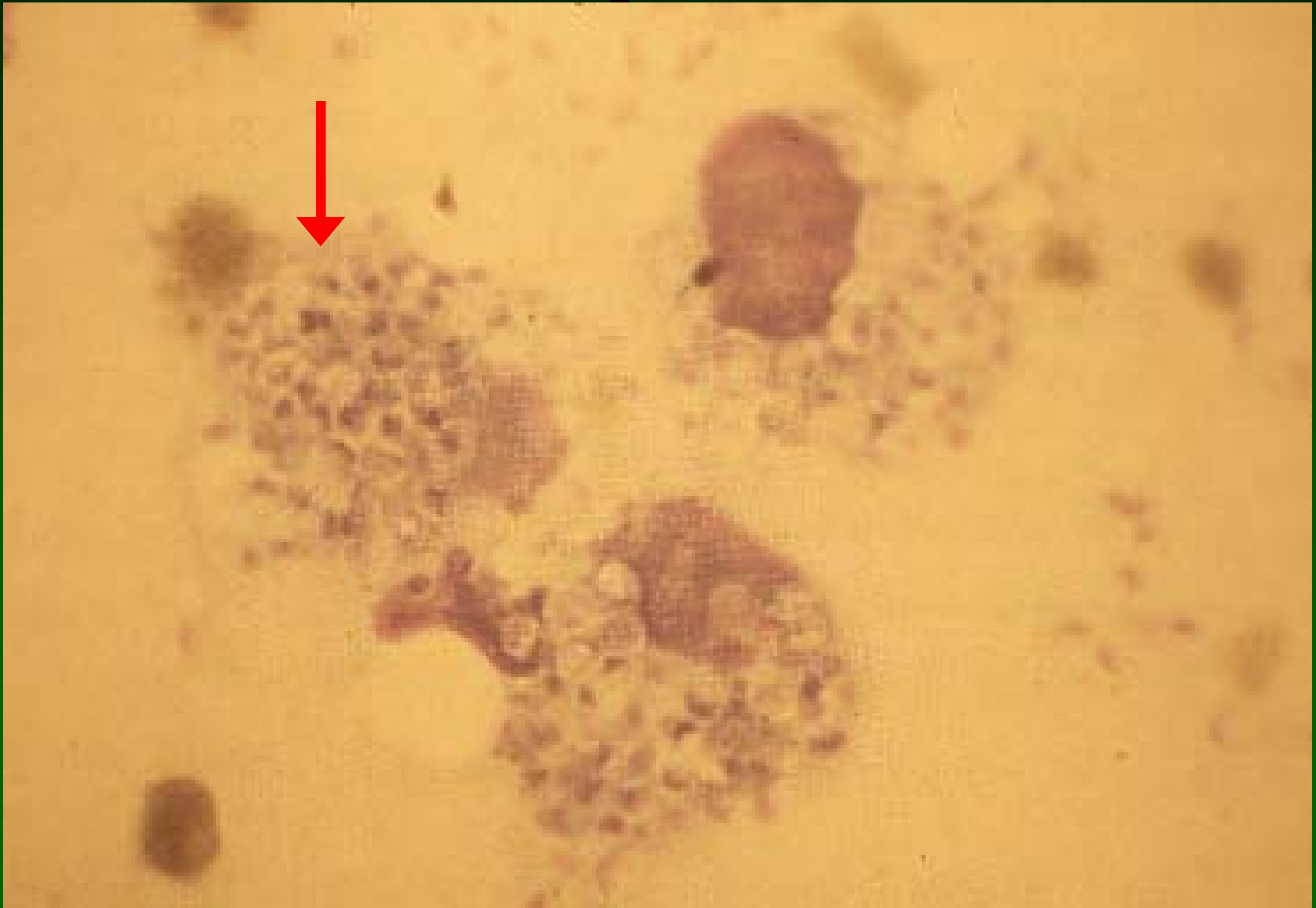


Aspergillus

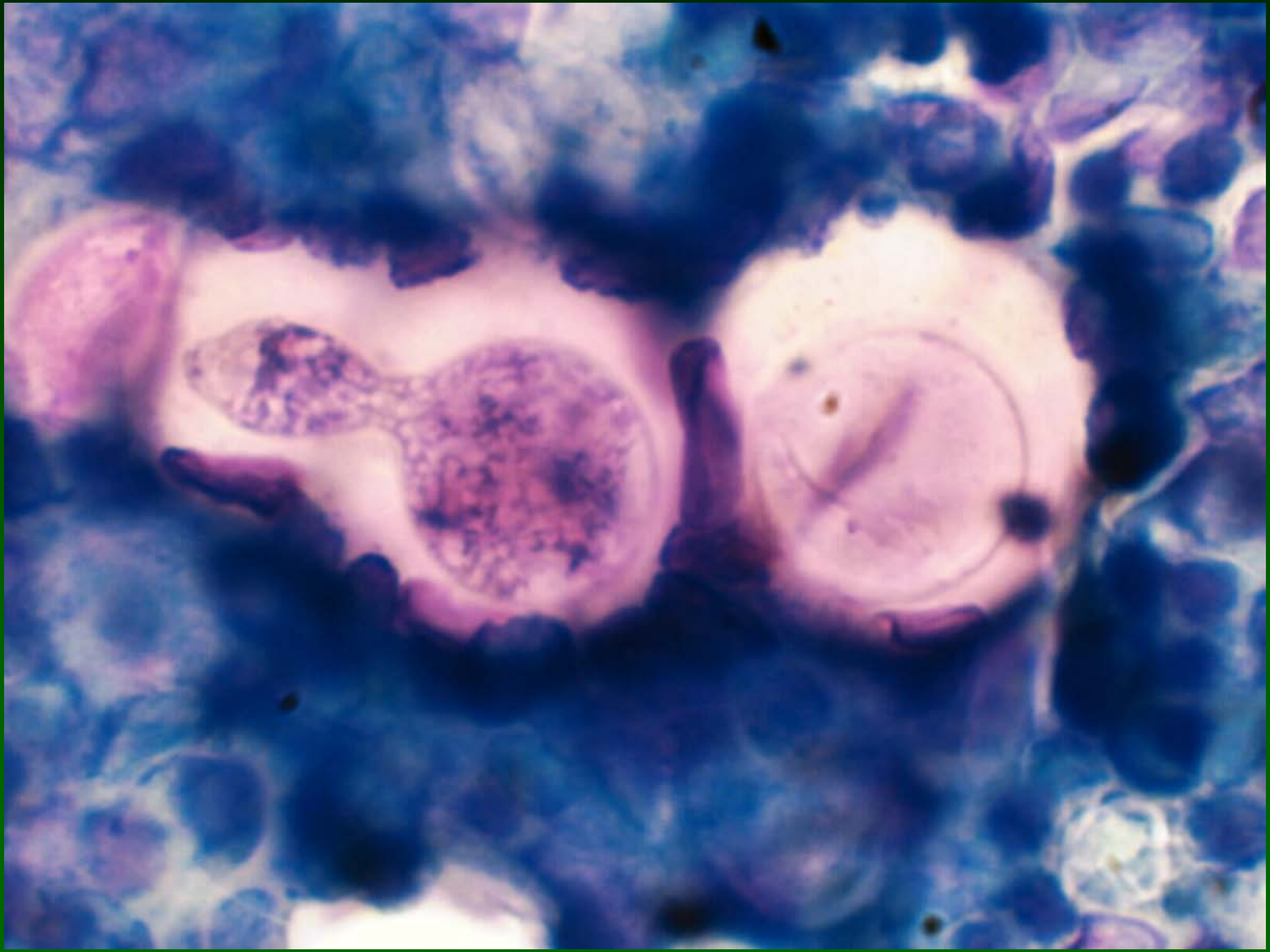


Blastomyces

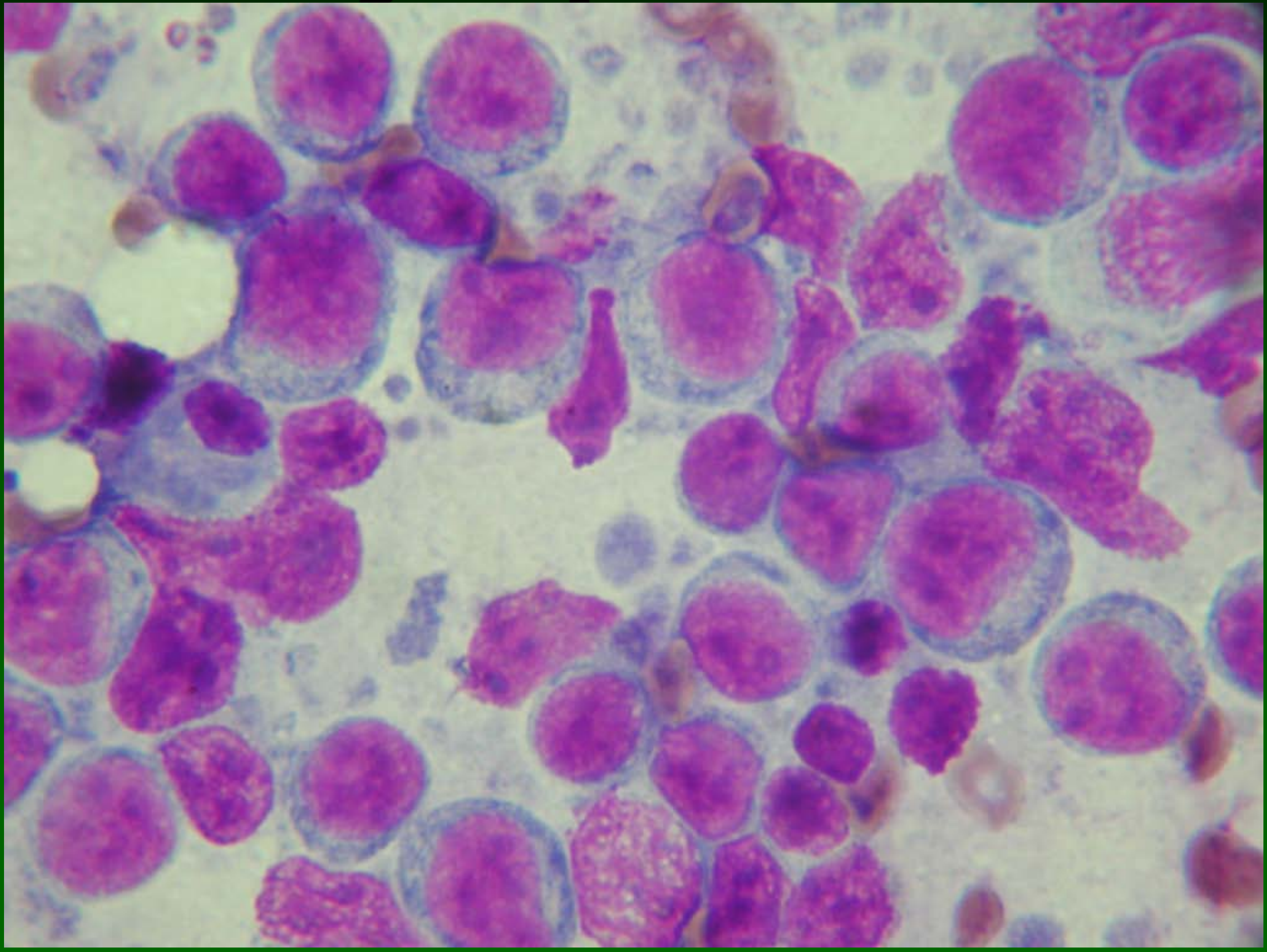
Histoplasma



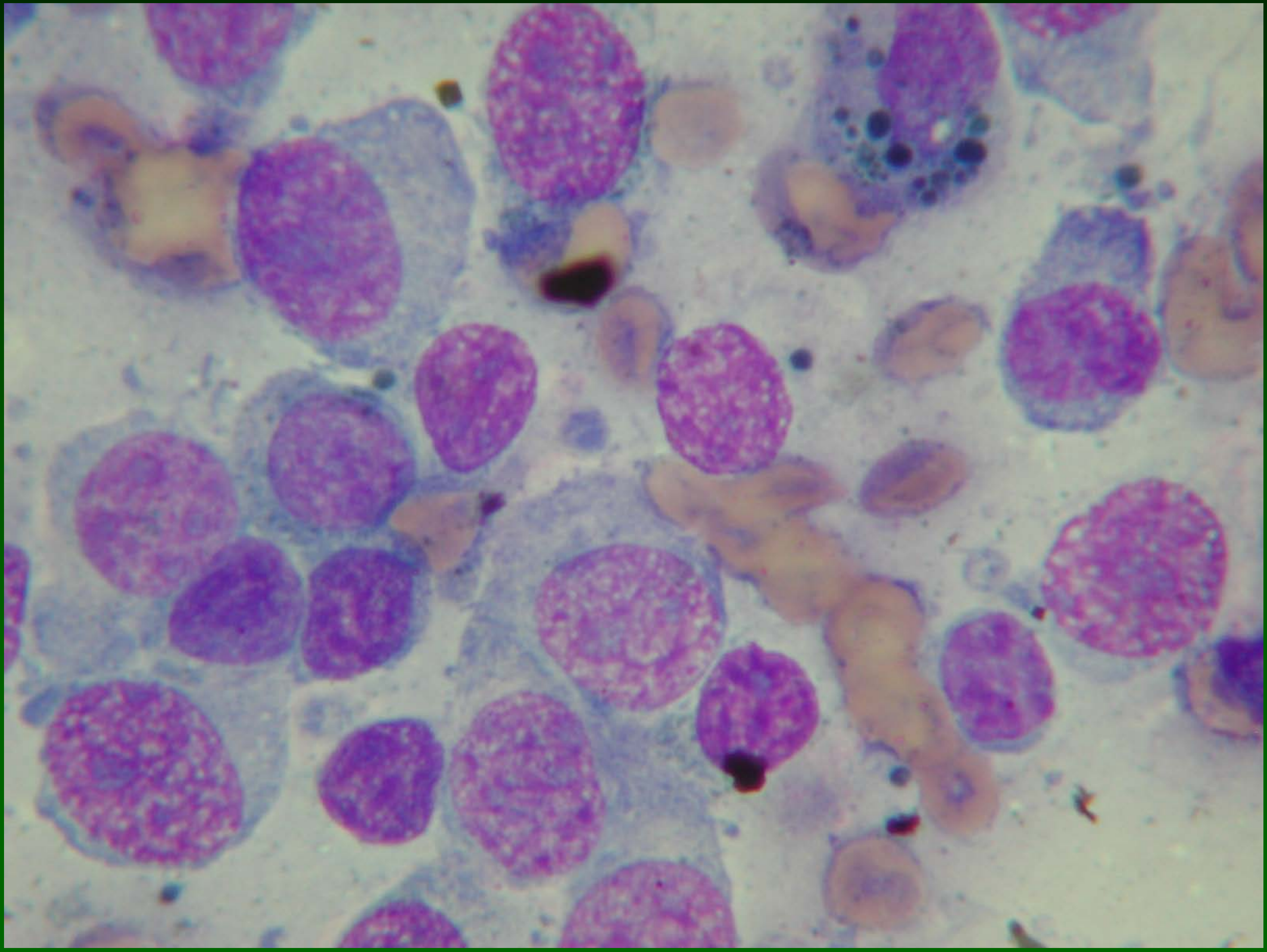
Cryptococcosis



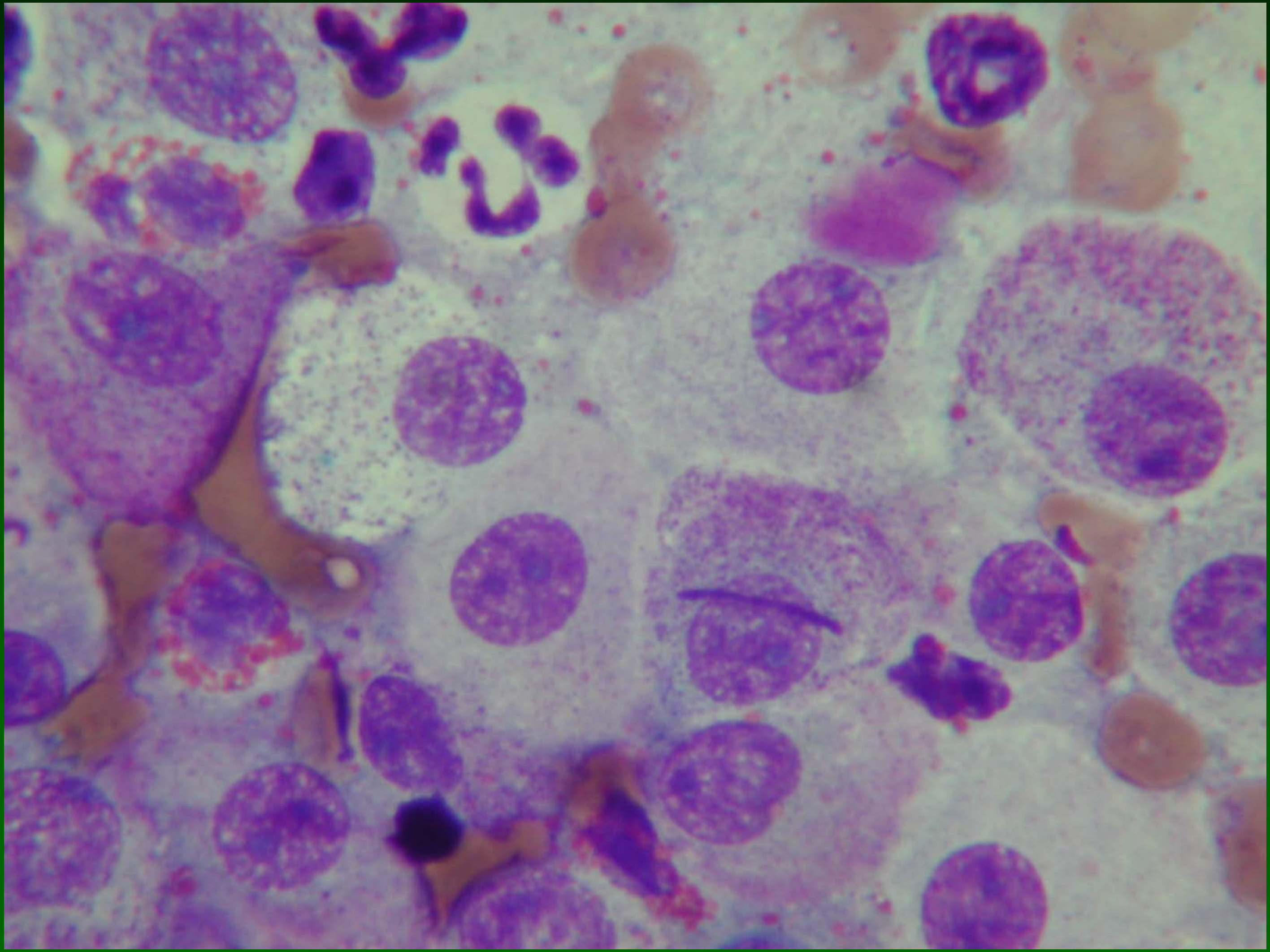
Lymphoma



Malignant melanoma

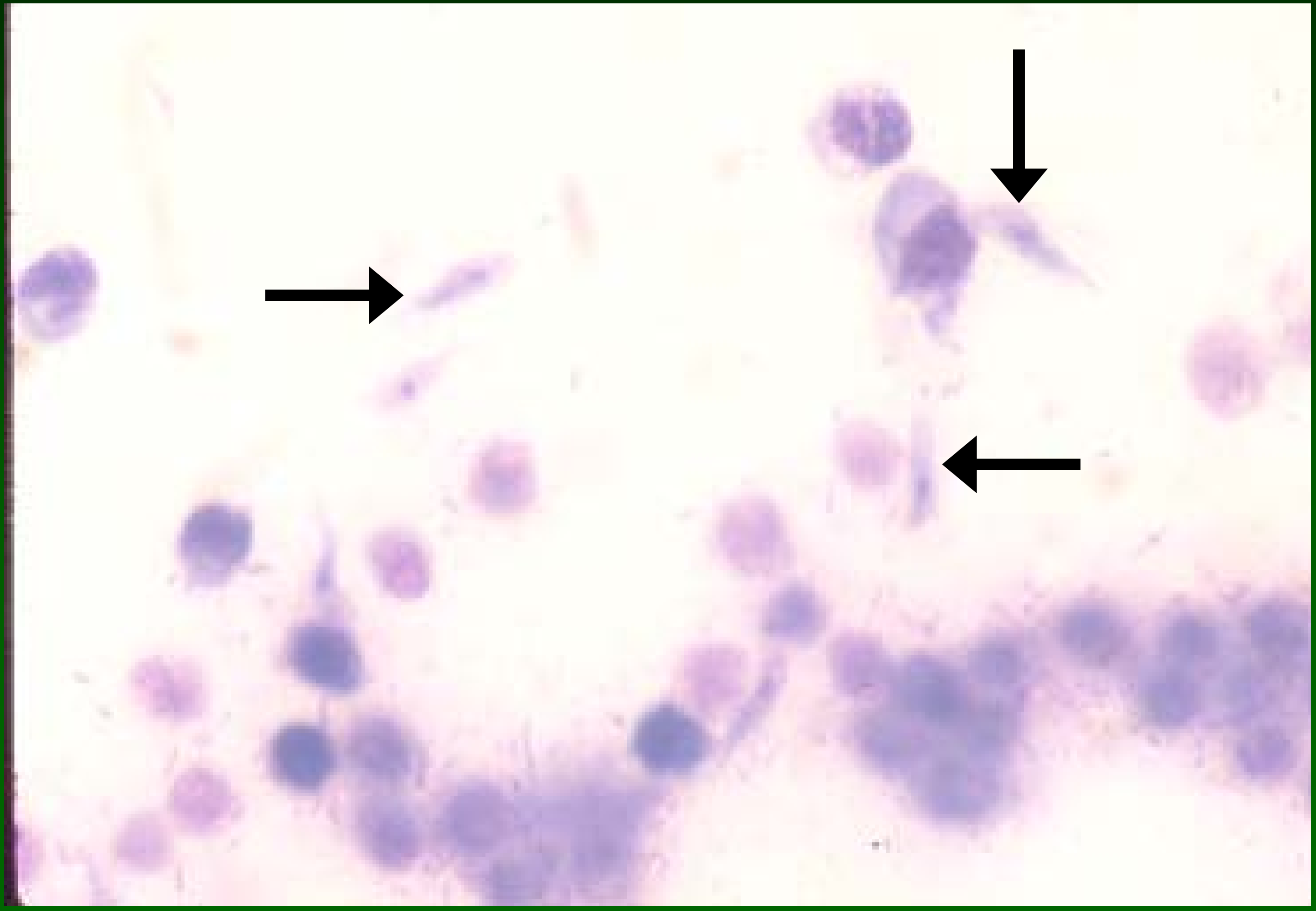


Mast cell tumor



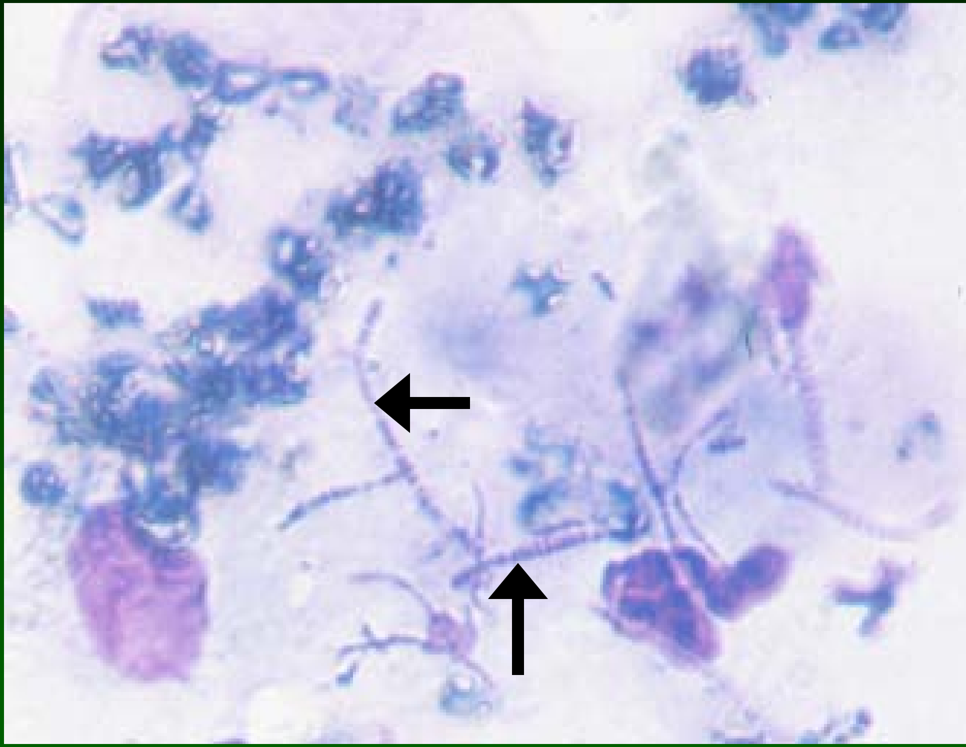
Owl with mouth lesion

Trichomoniasis



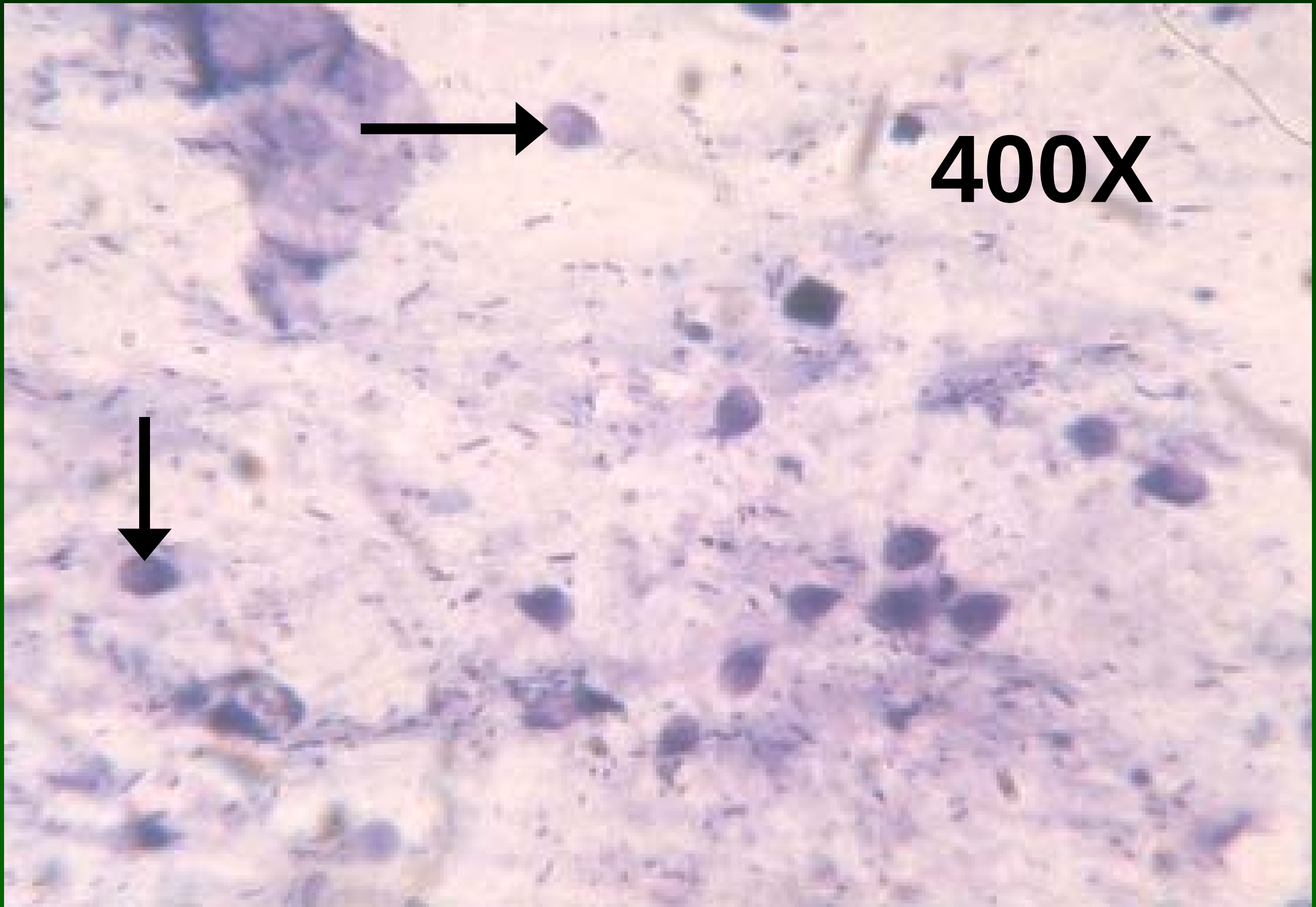
Candida with hyphae

Deer dermatitis

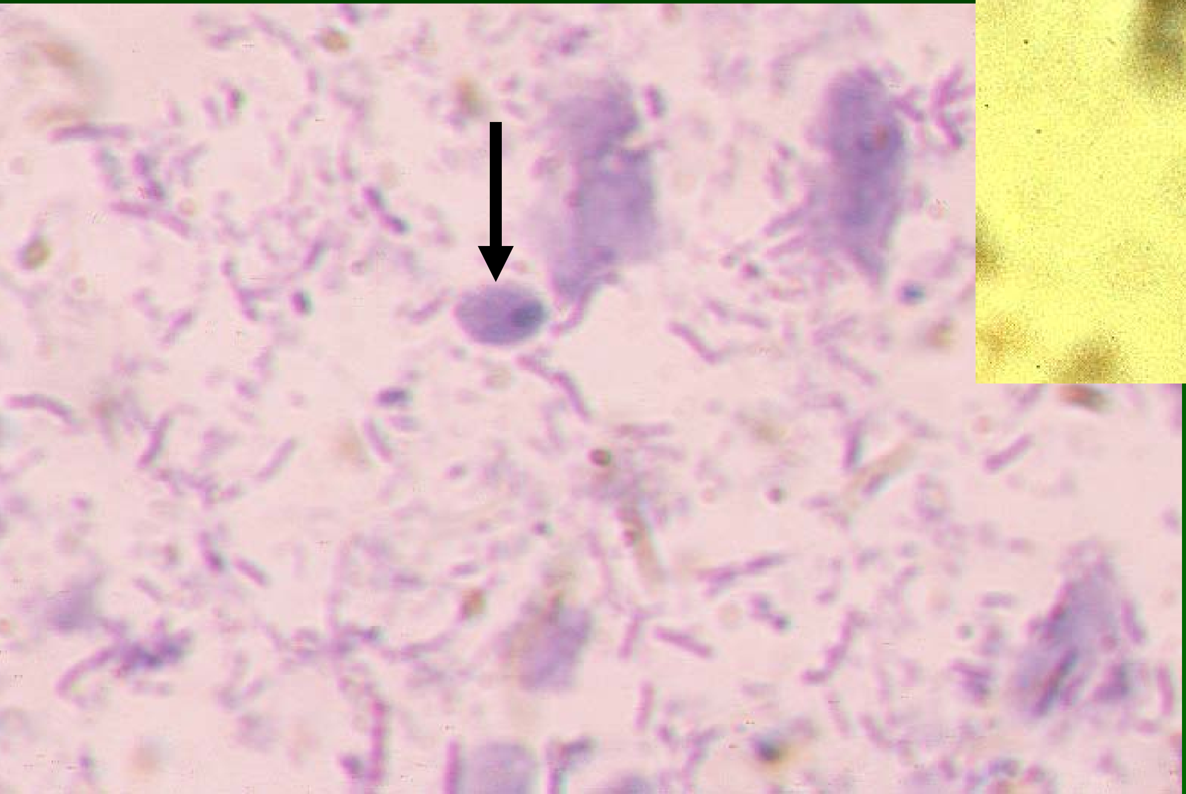


Dermatophilus

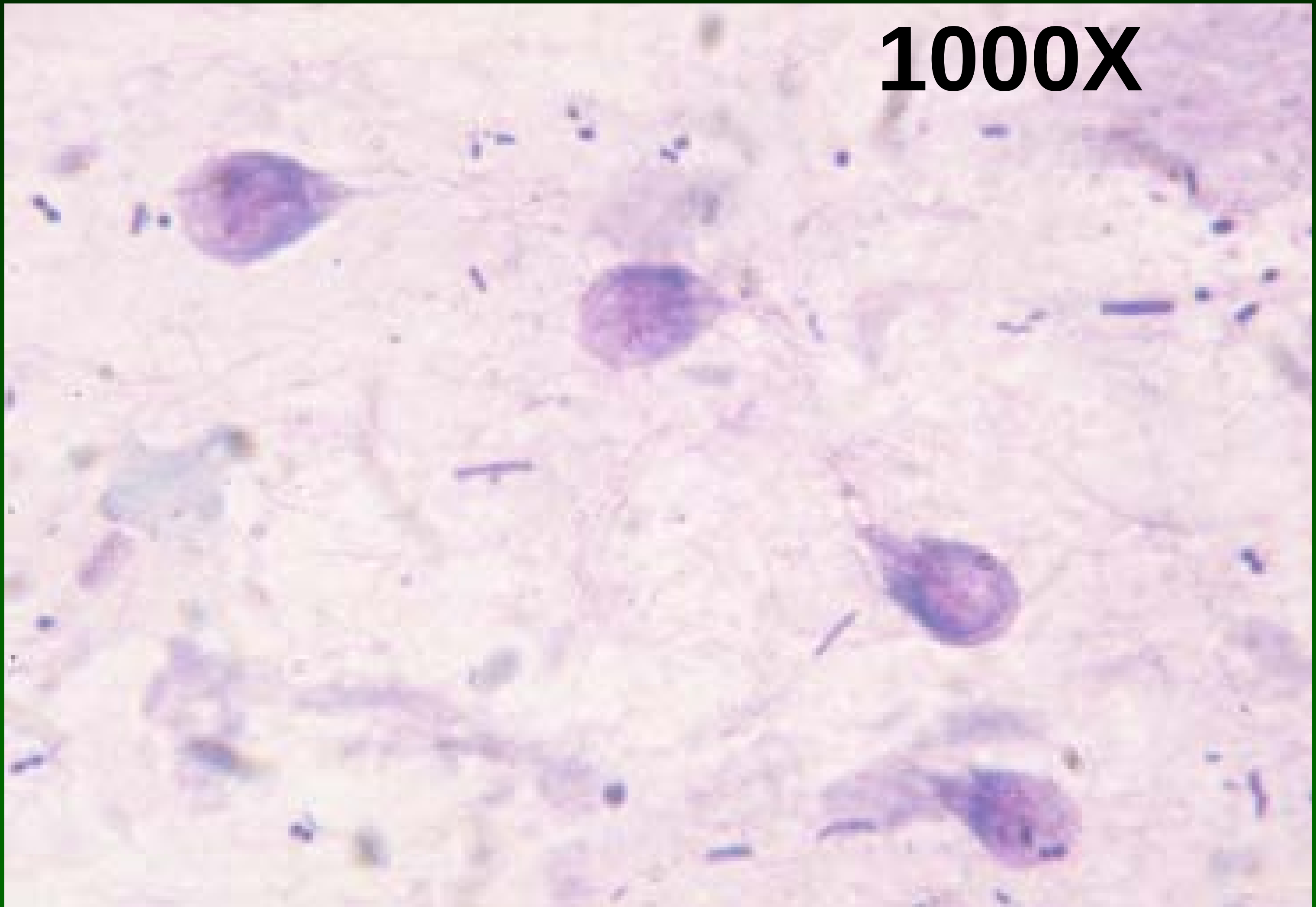
Giardia on DQ stained fecal smear



Giardia cyst- DQ oil

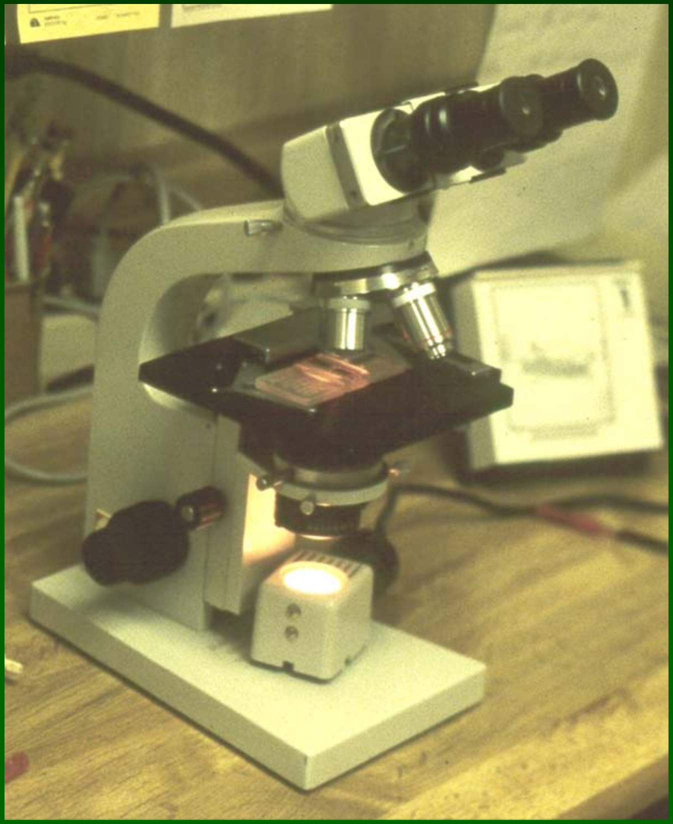


Trophozoites- DQ



Cell counting

- Automated counters used in mammals
- Need Leukopet® & hemocytometer in birds
  - to count granulocytes
  - Estimate mononuclear cells
  - or Natt-Pette
    - to count erythrocytes, leukocytes, thrombocytes

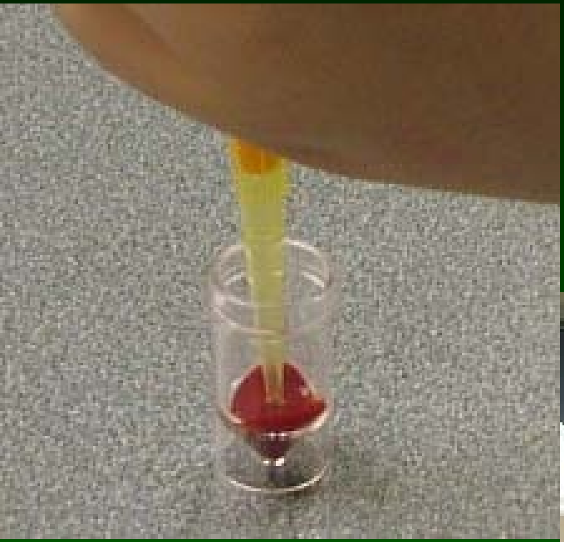


Avian Cell Counting

- Vetlab Supply
  - 800-330-1522
- Use 25µl minipet
  - Eppendorf pipet tip
  - premeasured phloxine tubes
- Draw blood into tip, wipe, add to phloxine
- Counts only granulocytes
- Use diff results to estimate monos, lymphs

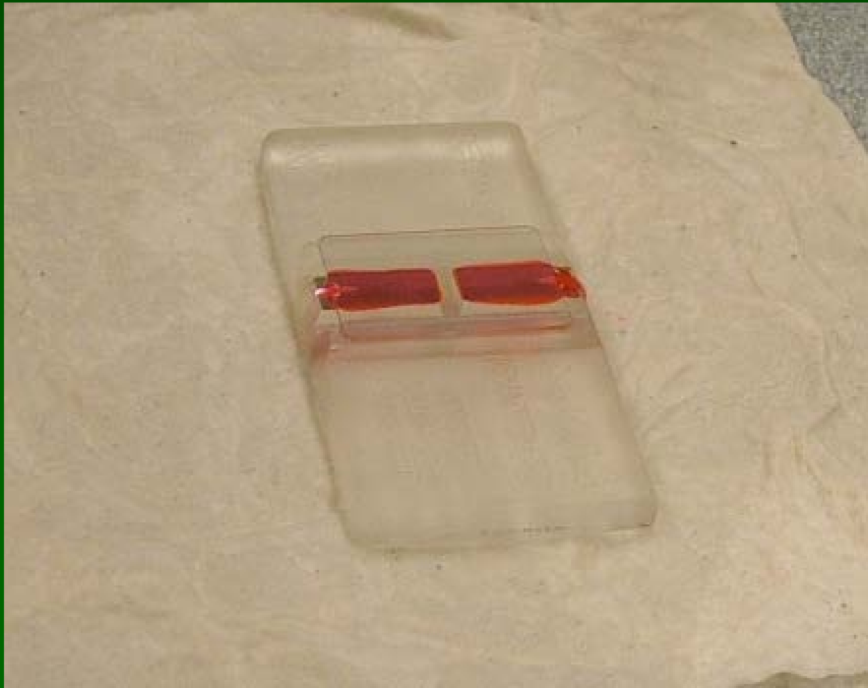
Avian Leukopets

Leukopette use



Procedure

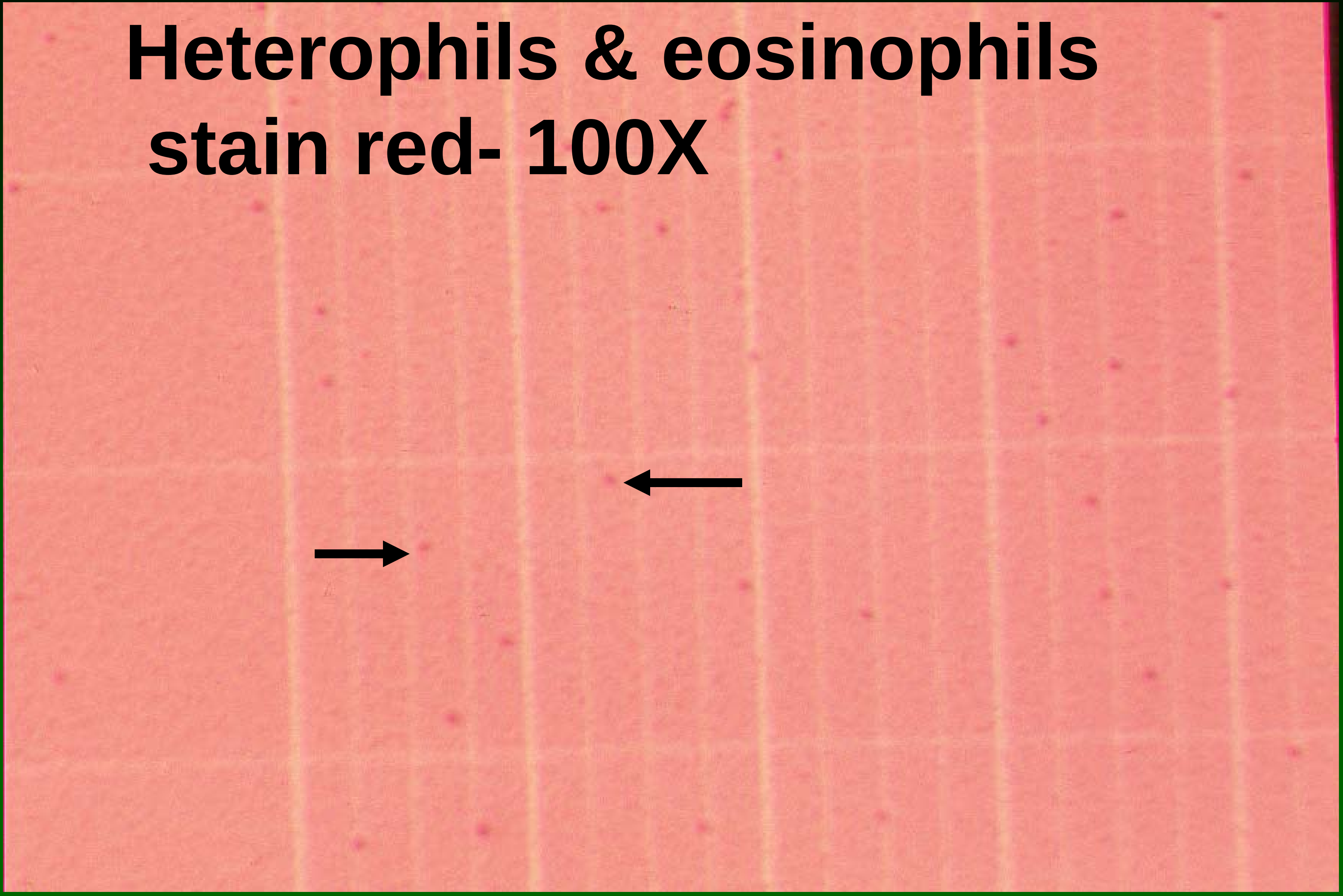
- Use 25 μl well mixed blood
- Load into diluent
- Let sit 5 minutes
- Load both sides of hemocytometer
- Let sit 5 minutes

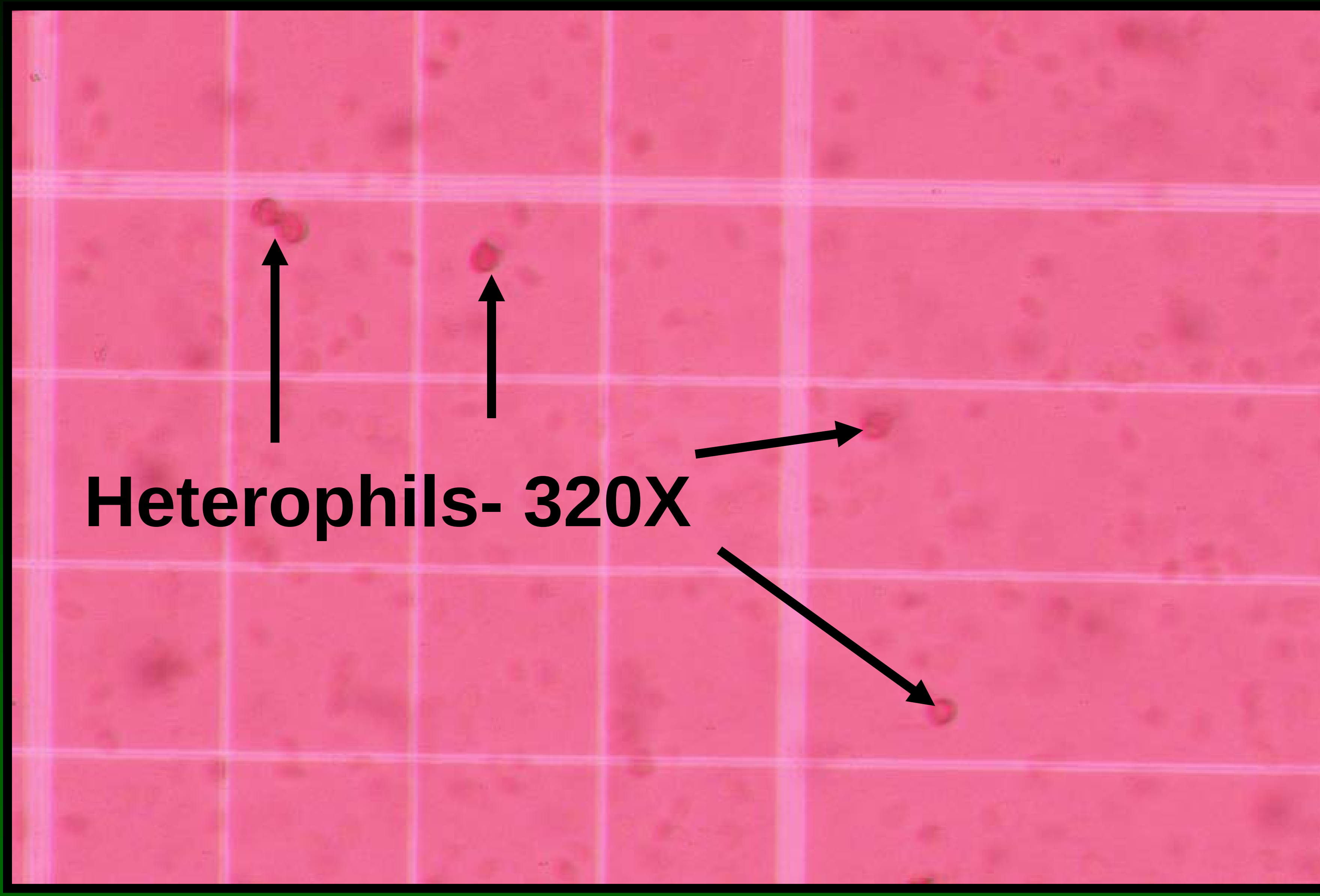


Unopette technique

- Count all cells in all 9 primary squares on both sides
- Take 10% and add it to count
- Multiply by 16
- Get total granulocyte count
- Estimate mononuclear cells

Heterophils & eosinophils stain red- 100X




Heterophils- 320X

To calculate mononuclear cell count

- % lymphs & monocytes / % granulocytes
  - from differential
- Multiply by granulocyte count
  - from hemocytometer
- Add granulocyte count to mononuclear cell count

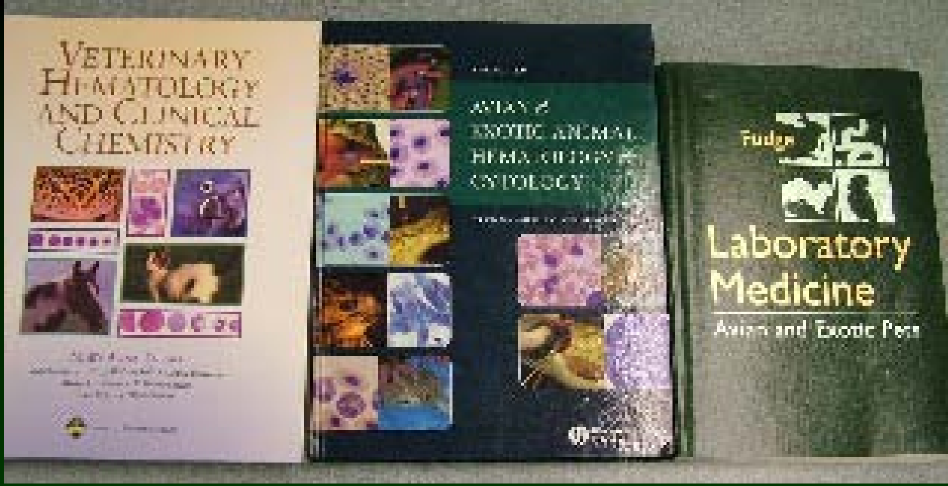
Example

- H-60, E-8, L-27, M-5
  - 12,000 granulocytes (unopette)
- 27 + 5= 32, 60 + 8= 68
- 32/68 X 12,000 = 5,647
- 5,647 is mononuclear cells
- total count= 17,647

Another formula

$$\frac{\text{cells counted} \times 1.1 \times 16 \times 100}{\%\ \text{granulocytes from diff}}$$

Useful References



- Avian & Exotic Mammal Hematology and Cytology by T. Campbell
- Laboratory Medicine: Avian and Exotic Pets by Fudge
- Veterinary Hematology & Clinical Chemistry by M. Thrall
- Veterinary Parasitology: A reference manual by W. Foreyt
- Color Atlas of the Cytology of the Dog and Cat by R. Baker and J. Lumsden
- Diagnostic Cytology & Hematology of the Dog & Cat by R. Cowell and R. Tyler