Examination of the Blood Film in the Dog and the Cat

Procedure for Blood Film Evaluation

- Scan slide at 20x magnification
  - Predominant leukocyte
  - Erythrocyte arrangements
  - Platelet clumps
  - Abnormal cells
    - Nucleated red blood cells (NRBC)
    - Reactive, left-shifted, neoplastic
- Erythrocyte morphology (100x)
- Platelet numbers and morphology (100x)
- Leukocyte numbers and morphology (100x)

Erythrocyte Evaluation

100x Oil Objective

- Erythrocyte morphology
  - Changes in size, shape, or color
- Evaluation for presence of hemoparasites
The Saline Test

- Distinguishes rouleaux from agglutination
- One drop of patient blood and 1 ml saline
- Examine as unstained, wet-prep on 20X with low light

Changes in Shape

- Nonpathological
  - crenation
- Pathological
  - Polychroms.
  - Acanthocytes
  - Schistocytes
  - Keratocytes
  - Spherocytes
Changes in Color

• Related to amount of hemoglobin in the cell
• Two changes recognized
  – Polychromasia
  – Hypochromasia
New Methylene Blue Stain  
(Reticulocyte Stain)

Erythrocyte Inclusions

- Stain precipitation
- Drying artifacts
- Hemoglobin abnormalities
- Erythrocyte nuclear material
- Hemoparasites
Hemotrophic Mycoplasmas  
Formerly Haemobartonella

- Extracellular (surface) RBC parasites
  - Causes immune-mediated (primarily extravascular) destruction of RBCs by host
  - Coombs’-positive anemia

- 2 species in cats
  - *M. haemofelis* (formerly large *H. felis*)
    - Most pathogenic (anemia)
  - *M. haemominutum* (small form)
    - Mild clinical signs if any (many infections unapparent)

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*M. haemofelis*

- Believe to be vector borne, primarily transmitted by fleas
  - Mechanical transmission is also possible
  - Risk factors – outdoor cats, cat-bite abscesses, FeLV positive status
  - Clinical signs vary from unapparent disease to severe anemia and death
    - Fever, pale mucus membranes, icterus, splenomegaly

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*M. haemofelis* in Peripheral Blood

- Attaches to erythrocyte surface and indents membrane
- Causes membrane damage and pock-like lesions even upon detachment
- May contribute to immune-mediated component of red cell destruction
- Cyclic parasitemia
Laboratory Findings

- Regenerative anemia – MCV, MCHC
  - unless complicated by FeLV infection
  - Combination may promote myeloproliferative disorders (erythrodysplasia / erythroleukemia)
- Autoagglutination and Coombs’ + anemia
- Hyperbilirubinemia (extravascular hemolysis)

Diagnosis

- Serology not available
- Identification of organism or organism DNA
  - Peripheral blood film: Fresh smear not in EDTA
  - Collect blood in heparinized capillary tube
  - PCR analysis of peripheral blood

PCR Analysis

- Highly sensitive PCR for *M. haemofelis* and *M. haemominutum*
  - 14% healthy control cats positive for haemoplasmas (subclinical infections)
  - May also get false negatives: cyclic parasitemia
Platelet Evaluation

- Scan slide for platelet clumps
- Estimation of platelet numbers
  - Dogs (# per 100x field) x (15,000 cells/ul)
  - Cats (# per 100x field) x (20,000 cells/ul)
- Platelet size
  - Megaplatelets
- Parasite
  - *Anaplasma platys*

Leukocyte Evaluation

- Estimation of leukocyte numbers
  - (# per 100x field) x (10,000 cells/ul)
- Differential cell count
- Morphologic changes in leukocytes
  - Toxicity
  - Reactivity
  - Parasites
  - Neoplastic changes
Neutrophils
- Predominant leukocyte in dog and cat
- Indicators of inflammatory/infectious disease
  - Corticosteroid induced stress leukogram
- Left-shifting
  - Degree indicates magnitude of disease process
    - Degenerative vs. regenerative

Eosinophils and Basophils
- Neoplasia
- Allergic / hypersen.
- Addison’s
- Collagen / vascular
- Parasitism

Monocytes and Lymphocytes
- Left-shifting
  - Response to increased demands for neutrophils in peripheral tissues
  - Regenerative, degenerative and transitional shifts

Left-Shifting
Toxic Changes

- Developmental abnormality seen only in the neutrophil cell line
  - Cytoplasmic changes
- Different from degenerative changes
  - Nuclear changes
- Typically associated with infectious diseases or exposure to endogenous or exogenous toxins
- Variable stages of severity
  - +1, +2, +3, +4

+1 Toxicity
Arrows Indicate Doehle Bodies

+2 Toxicity

Doehle Bodies
Cytoplasmic Basophilia

+3 Toxicity

Doehle Bodies
Cytoplasmic Vaculation

+4 Toxicity
Arrows Indicate Doehle Bodies
Hypersegmented Neutrophils

- Prolonged maturation in circulation
  - Corticosteriod therapy
- Bone marrow disease
  - Dysplasia
  - Neoplasia

Reactive Lymphocytes & Monocytes

- Increased cytoplasmic basophilia
  - Monocytes & lymphocytes
- Cytoplasmic vacuolation
  - Monocytes

Neoplastic Cells

- Non-hematopoietic cells in circulation
  - Mast cells
- Hematopoietic cells
  - 3 or more times the size of erythrocytes
  - Increased cytoplasmic basophilia
  - Diffuse chromatin pattern in nuclei
  - Other anaplastic features (nucleoli)
  - Lymphoproliferative vs. myeloproliferative
Summary

- The systematic evaluation of a blood film is the most important part of the hemogram.
- Estimates of the complete blood count can be quickly obtained from the blood film.
- The morphological evaluation of blood cells can yield clinically important information that cannot be obtained by any other laboratory testing.