
Quantitative Analysis of Anemia

- Anemia (reductions in erythrocyte mass)
  - Hemorrhage
  - Hemolysis
  - Decreased production
- Two areas of quantitative analysis
  - Evaluation of red blood cell mass (PCV and HCT)
  - Red blood cell indices

Analysis of Red Cell Mass

- Hematocrit, RBC, PCV
  - Red cell mass in relation to plasma volume
  - PCV vs. HCT via automated cell count
    - HCT calculated based on RBC and MCV
    - HCT Affected by agglutination

Red Blood Cell Indicies

- MCV, MCH, MCHC
- Assist in the classification of anemia
  - Regenerative
    - Hemorrhage
    - Hemolysis
  - Nonregenerative
    - Decreased production
- Identification of specific diseases or pathological processes
Mean Cell Volume (MCV)

- An evaluation of the average size (volume) of a single RBC
- Can be measured directly (cell counters)
- Calculation: \( \frac{PCV}{RBC} \times 10 \)

Increased (larger than normal RBCs)
- Regenerative anemias (requires a week or more)
- FeLV
- Leukemia / preleukemia (Myeloproliferative disorders)

Decreased (smaller than normal RBCs)
- Iron deficiency (requires 1 or more months)
- Fragmentation of erythrocytes
- Portosystemic shunts
- Normal in Akita dogs

Mean Cell Hemoglobin Concentration (MCHC)

- The average Hb concentration
- Calculation: \( \frac{Hb}{PCV \ or \ HCT} \times 100 \)
- Decreases
  - Regenerative anemia
  - Iron deficiency anemia
- Increases (artifact)
  - Intravascular hemolysis (also in vitro)
  - Heinz body formation
  - Lipemia

Concurrent Evaluation of Indicies

- Necessary for interpretation
- MCV
  - Normocytic
  - Macrocytic
  - Microcytic
- MCHC
  - Normochromic
  - Hyperchromic
  - Hypochromic
Common Abnormalities in the Anemic Pet

- Normocytic, Normochromic
- Macrocytic, Hypochromic
- Microcytic, Hypochromic

Normocytic, Normochromic

- Normal MCV and MCHC
- Nonregenerative anemia
  - Decreased production problem
    - Anemia of Chronic Inflammation
    - Chronic Renal Failure
    - Bone marrow disease
  - Peracute hemorrhage or hemolysis
    - 3 to 5 days for peripheral response
    - May take a week or more to change indices
    - RDW (Red Cell Distribution width)
    - Blood film evaluation

The Red Cell Distribution Width

- A measurement of anisocytosis
  - Variability in red cell size
- The more variability in size (smaller or larger red cells) the higher the RDW
- Does not depend on numbers of cells to increase an average as does MCV
- More sensitive indicator of regeneration
- May also be increased with red cell fragmentation

Macrocytic, Hypochromic

- Elevated MCV, decreased MCHC
- Regenerative anemia
  - Hemolysis (most dramatic response)
  - Hemorrhage (less dramatic)
    - Temporal
      - Acutely nonregenerative
      - Regenerative
      - Poorly regenerative
Red Cell Indices and the Regenerative Anemia

- Study of over 4,000 dogs with anemia
  - (JAVMA, 138:1452-1458, 2011)
- 32.5% had regenerative anemia
  - Of those, only 11.8% had macrocytosis and hypochromasia (11% sensitivity; 98% specificity)
  - Polychromasia on blood smear alone, or with high RDW had 77% and 79% accuracy, respectively

Microcytic, Hypochromic

- Iron deficiency
- Chronic blood loss
- Diameter may be normal
- Cells are flat with reduced hemoglobin

Other Abnormalities of Indices

- Macrocytic, normochromic (Inc. MCV)
  - Leukemia / preleukemia
    - Dysplastic change (preneoplastic)
    - Neoplastic (leukemia)
  - FeLV infection
- Microcytic, normochromic (Dec. MCV)
  - Portosystemic shunt
  - Normal in Akita and Saluki dogs

The Platelet Count

- Low platelet counts should always be confirmed
  - Check tube and stopper for clots
  - Evaluate platelet numbers on blood smear (clumps)
- Beware of the thrombocytopenic cat!
- Very difficult to obtain accurate counts for cats
- Impedence counters vs. Laser technology
  - Size of platelets vs. RBC
  - Platelet clumps
Thrombocytosis

- Iron deficiency and chronic blood loss
- Nonregenerative (Central) IMHA
- Essential thrombocythemia (leukemia)
- Vincristine
- Hyperactive bone marrow (severe regenerative anemias)
- Cushing’s disease (increased glucocorticoids)
- Diabetes mellitus
- Splenectomy
- Inflammatory diseases

Thrombocytopenia

- Bone marrow disease: typically will involve other cell lines as well
  - Infectious, drug-induced, neoplastic
  - IMT directed against megakaryocytes
- Infectious agents: tick-borne diseases
- Numerous drugs (Immune mediated)
- Vascular disease / neoplasia (HSA / Thyroid)
- IMT alone
  - Most severe decreases
  - Leukopenia and red cell changes typically absent

Mean Platelet Volume (MPV)

- Machine-calculated measurement of the average size of platelets found in blood
  - Requires accurate identification
- Increased in regenerative responses to increased platelet demands
- Severe thrombocytopenia (<10,000 / µl) with increased MPV → IMT
  - In the absence of other cytopenias
  - Blood loss anemia being one exception

Thrombocytosis

- Response to inflammation
  - Low grade inflammatory responses may have high MPV (vascular disease, cancer)
    - May be associated with conditions promoting vascular thrombosis
    - Negative prognostic indicator for thrombosis in people
    - Cardiovascular and cerebrovascular diseases
  - High grade inflammatory responses may have low MPV
    - Resolve after appropriate therapy
The Leukogram

WBC (it’s not enough!)
- Analyzers that perform an accurate differential are superior
- Clinically significant changes with normal Leukogram
  - Increased numbers of immature neutrophils
  - Toxicity
  - Atypical leukocytes (reactive or neoplastic)
  - NRBCs
- Laser technology vs. impedance counters
- “Flagging” atypical cells (Reactive or Neoplastic?)

Normal Neutrophil Numbers

- Left shifts not often reported on the hemogram
- Major problem when numbers are normal
- Degenerative left shift
  - More immature neutrophils than mature in a patient with normal or low neutrophil numbers
  - Guarded prognostic factor especially in cats!
  - Up to 50% mortality rate due to death or euthanasia (Burton et al. 2013. JVIM 27(6) 1517-1522)

Nucleated Red Blood Cells

- Regenerative anemia if polychromasia is present (reticulocytosis)
- Bone marrow disease or damage in patients without anemia or polychromasia
  - Lead poisoning
  - Bone marrow neoplasia (leukemia)
  - Bone marrow damage (septicemia)
- Can alter the leukogram
  - Mistaken for lymphocytes (lymphocytosis)
  - Falsely increased WBC

Correction for NRBCs

- 100/100 + NRBCs X WBC = corrected WBC
- Must perform a differential and record the # NRBCs per 100 WBCs
- Significant when there are >5-7 NRBCs / 100 WBCs.
Microscopic Examination of a Blood Film (When?)

- Any quantitative abnormalities
  - RBC
  - WBC
  - Platelets
- Evaluation of sick patients
- Quality assurance for hemogram

Slide Preparation

- Small drop of blood
- Push Smear
- Quick drying to reduce artifacts
- Staining
Staining the Blood Smear

- Three-step staining set
  - Diff Quik®
  - Fix 2 min. or as long as you like
  - Red – 1 min.
  - Blue – 45 sec