Evaluating avian hematology: A clinical perspective

Interpretation of the avian hemogram can be challenging for clinical pathologist and the avian clinician. Provided here are a series of steps which may be useful in interpreting avian hematology from a laboratory and clinical perspective. Foremost should be the realization that few true reference intervals or normal ranges are available in avian medicine. By convention, the normal range for a particular test (For example total white blood cell count) is set to cover ninety-five percent (95%) of all values from the general population. Because 5% of results fall outside the normal range, values that prove normal can therefore sometimes be outside the normal range. In human medicine ranges of normal for a single value (like total white blood cells for example) are created for a single instrument or laboratory from 100s of apparently healthy people and may be further modified based on gender, age, race, or other factors. Results are statistically evaluated to create a reference or normal range (Figure 1). However exotic animal practitioners and scientists seldom have access to these numbers of healthy animals of the species in question. Therefore it is helpful to develop a series of rules of thumb to evaluate avian hematology in the absence of a reference range.

1. Evaluate packed cell volume
a. For flighted birds this may easily range to 55% without concern
b. In couch potato birds, the dog cat range is very acceptable 35-45%
c. Anemia of chronic disease is common in birds
d. I seldom consider transfusion in birds until 12% as they have very efficient respiratory systems to compensate
e. 1+ polychromasia is normal in birds and all red blood cells are normally nucleated
f. A reticulocyte count is rarely done, but can be requested
g. Basophilic stippling can be seen with lead toxicity in birds

2. Evaluate the total solids
   a. Consider which method has been used to determine total solids
   b. Dry slide or automated analyzer is more appropriate, EPH is most accurate
   c. Refractometric determination is often higher than actual based on the normal glucose (~300) of birds
   d. Birds generally have total solids of 3-5
   e. However 2-3 is not uncommon in juvenile birds which appear to suffer no ill effects

3. Obtaining an accurate white blood cell of quality and accuracy in birds is heavily dependent on the quality of the blood smear
   a. Anticoagulants used in avian medicine include: lithium heparin, sodium citrate and edetate dipotassium sodium (EDTA)
   b. Adding bovine albumen may stabilize cells and reduce skip-o-cytes
   c. A fresh blood smear, quickly air dried, remains the gold standard
   d. Cover slop to cover slip or standard wedge techniques may be used. The correct technique is the one which results in the fewest damaged cells.

4. Obtain the White Blood Cell Count & Differential; Indirect method
   This method is most commonly used in reference laboratories and is MUCH MORE accurate than a WBC estimate. However all results heavily depend on cell quality in the sample and smear

5. Obtain the White Blood Cell Count & Differential; Estimate method
   a. Can be done in house but is time consuming and a poor estimate
   b. Method
      i. Count white blood cell number at the cell monolayer
      ii. Take average number from 10 20 x fields x 1500 = Total White Blood Cell Count Estimate
      iii. However this method provides only an estimate and is unlikely to provide the information quality that a cell counting chamber and a differential can.
         1. At the cell monolayer count 10 hpf (40x) fields = W
         2. W/10 x 1500 – estimated white blood cell count
   c. Most birds have a white blood cell count which ranges from 5,000-15,000 cells/μl
   d. Birds do have a stress leukogram which can elevate the total white count up to 20,000 (IME) without other apparent causes of inflammation
      i. Small birds tend to have lower white blood cell counts (3,000-4,000 cells/μl) without apparent effect: Canaries, Finches, small passerines, Old cockatiels
   e. White blood cell count > 30,000?
6. Evaluate the Heterophil to Lymphocyte ratio (H:L)
   a. In most birds this tends to range from 1.5:1 to 3:1
   b. However some birds are 1:1 (lymphocytic)
      i. Examples: Canaries, Amazons, Macaws, Quaker parrots, Owls, Bald eagles
   c. Abnormalities in this ratio results in a relative heterophilia, relative lymphopenia etc.
      i. Heterophil form: The most common inflammatory cell of most birds is quite different from the neutrophil of mammals. This cell has a multilobed nucleus bit is characterized by its rod shaped granules which may stain blue or pink in different species or may only barely uptake stain.
      ii. Lymphocyte form: Small and large lymphocytes are present in most species and have a similar form to that of mammals
      iii. Relative Heterophilia (increased heterophils) – most likely this is due to acute inflammation. Differentials for heterophilia are the same as those which would cause a neutrophilia in mammals.
      iv. Causes of inflammation include trauma, neoplasia, allergy/immune, infection
      v. Relative Lymphocytosis (increases lymphocytes)- chronic inflammation which may be viral
      vi. Heteropenia is rare in birds but is of clinically indicative of a poor prognosis and results in a leucopenia. This may be associated with young cells (band heterophilia, left shift) which are associated with severe inflammation which is overwhelming the body.
      vii. A stress leukogram is defined as a heterophilia, lymphopenia and monocytosis IME common in birds.

d. Evaluate the monocytes
   i. Form: similar to mammals
   ii. In most birds < 1,000 monocytes is acceptable.
   iii. 1,000~1,500 cells gray zone which can be associated with a stress leukogram
   iv. > 1,500 cells I consider a real monocytosis, associated with chronic inflammation

e. Evaluate the eosinophils
   i. Form: smaller cell round with nucleus which may be bilobed, this cell is characterized by round granules which are often pink/orange; however in certain species, granules may be bluish in color
   ii. This is a rare cell in many species
      1. common in wading birds, barred owl and red shouldered hawks
      2. Eosinopenia does not occur
   iii. Function remains elusive but it may be involved in allergic response, fungal disease and parasitism as in mammals
f. Evaluate the basophils
   i. Form: small cell with eccentric ovoid dark purple nucleus. Intensely stained blue purple granules which may obscure the nucleus and often make the cell appear
as a cluster of grapes. Diff-Quick stain may degranulate this cell resulting in a small “ghost cell” with a pinkish cytoplasm

ii. Basophils are rare in most birds thus basopenia does not occur

iii. Basophilia has been associated with chronic respiratory disease in cockatiels

g. USEFUL BIOCHEMICAL TESTS

i. Uric acid, Bile Acids

ii. TP, Alb, Glob

iii. Glucose, AST, GGT, ALKP, GLDH

iv. Electrolytes, Lactate, blood gases