

Hematology?! Using the Stuff You Forgot (Blocked) From School

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Introduction

As technicians, one of the many valuable skills we can provide for our practices is blood smear evaluations. The goals of the lecture are to review: 1) review slide preparation; 2) review basic blood cells; and 3) what abnormalities to watch for.

Review of Slide Preparation

To prepare a sample for cytology use a sample in EDTA, lavender top tube, and gently invert the sample ten times. Using a syringe, either 3 mL or 1 mL, gently aspirate a small sample from the blood tube. Drop a small blood sample onto a clean, dry microscope slide. I prefer a 25 g needle as the drop is small enough to allow for a good blood smear. If the drop of blood is too large, the smear may not provide a monolayer for cytologic evaluation. After the sample has been smeared on the slide it is important to look for even spread with no holes and a rainbow sheen indicating a monolayer of blood cells on the slide. Allow the sample to air dry prior to staining.

For general hematology the stain used is the Romanowsky stain, or Diff-Quik®, with the slides dipped in fixative reagent, eosinophilic solution, and basophilic solution for 10-15 seconds in each followed by rinsing with distilled water. Some preparations require 30 seconds such as lymph node aspirates, but blood smears only require 15 seconds.

It is recommended to change the stain every 1-2 weeks and clean the jars to prevent artifacts on slides. Precipitate crystals can form in older stain or stain that has evaporated. To prevent evaporation, make sure to keep the lids on the jars between uses.

Review of Hematologic Cytology

It is important to review a blood smear any time there are abnormalities noted on the in-house CBC machine, there is a suspicion of an autoimmune disease, transfusion reaction, snake envenomation, or a variety of other conditions.

Ideally a clinical pathologist would review the blood microscopically, however, this is not readily available for most and a turnaround time of 12 to 24 hours is expected. Because of this, technicians should have the training and be able to review a blood smear slide to look for classic cytological changes.

When starting a blood smear evaluation, it is recommended to start by viewing the stained slide in the monolayer at the lowest microscope setting, typically 4x magnification, for gross changes like platelet clumping. After a quick scan view the slide under 100x magnification. Using the battlement method perform a differential count and platelet count. While performing the count look at the cells for any abnormalities. The more you perform slide evaluations and compare them to the results from the in-house CBC machine and what the clinical pathologist reports, the better you become.

Abnormalities to look for include:

- Red Blood Cells:
 - Changes in colors of the red blood cells: polychromasia
 - Different sized red blood cells: anisocytosis

- nRBC's: nucleated red blood cells (usually counted per high power field)
- Are there other abnormalities such as echinocytes or spherocytes present?
- White Blood Cells:
 - Increases or decreases in cell numbers
 - Hypo- or Hyper- segmentation of the neutrophils
 - Do the neutrophils appear toxic?
- Platelets:
 - Are there adequate numbers of platelets?
 - Estimate by counting platelets in 10 high power fields, adding together and dividing by 10, and then multiplying by 15,000-20,000
 - Is there any platelet clumps present?
 - Are there micro- or macroplatelets present?
- Blood parasites:
 - Common ones include microfilaria and babesia.

To learn more about different abnormalities, a great reference is Cornell University College of Veterinary Medicine's eClinPath website (Cornell University College of Veterinary Medicine, 2013-2020). It is updated frequently and has everything from beginner to advanced level concepts. Idexx Learning Center has some good CE as well on hematology and cell identification as well as a book to use as a reference in the clinic (Idexx Laboratories, 2002).

It is important to keep track of what you see and record it in the medical record. A form such as the one in Table 1 can be used to record the results and keep within the medical record.

Table 1 Differential Form (Internal Medicine For Vet Techs, LLC, 2023)

Complete all the cells in grey

Patient Name & number: _____

Manual blood smear differential

Platelet estimate:	0 /ul	
Count # of platelets per high power field (hpf)		
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
Total # seen in 10 hpf	Ave. # of platelets	0

RBC morphology: _____

WBC morphology: _____


Platelet morphology: _____

Differential Count

WBC count from ProCyle	0 /ul	
basophil	0%	0.000 /ul
eosinophil	0%	0.000 /ul
neutrophil	0%	0.000 /ul
lymphocyte	0%	0.000 /ul
monocyte	0%	0.000 /ul

Date: _____

Performed by: _____



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Conclusion

Basic hematologic evaluation is an important skill technicians can perform that can provide valuable information about pets to help doctors provide a treatment plan. Proper preparation and evaluation is key to providing accurate results. Knowing the common blood cell abnormalities to monitor for can assist veterinarians in creating a treatment plan for patients.

References

Cornell University College of Veterinary Medicine. (2013-2020). Retrieved from EClinPath:
<https://eclinpath.com/>

Idexx Laboratories. (2002). *Guide to Hematology in Dogs and Cats*. Jackson, Wyoming: Teton NewMedia.

Internal Medicine For Vet Techs, LLC. (2023, August 29). *CE Handouts: Hematology?! Using the Stuff You Forgot (Blocked) From School*. Retrieved from Internal Medicine For Vet Techs:
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