

## Hematology of Infectious Disease

Mike Rosser, DVM, MS, DACVP  
Clinical Assistant Professor  
University of Illinois at Urbana-Champaign  
Consultant to Heska



1

## To Recap...



Submit **questions** anytime. They will be answered during Q&A



You can download the **CE Certificate** during Q&A. Other handouts can be downloaded now.

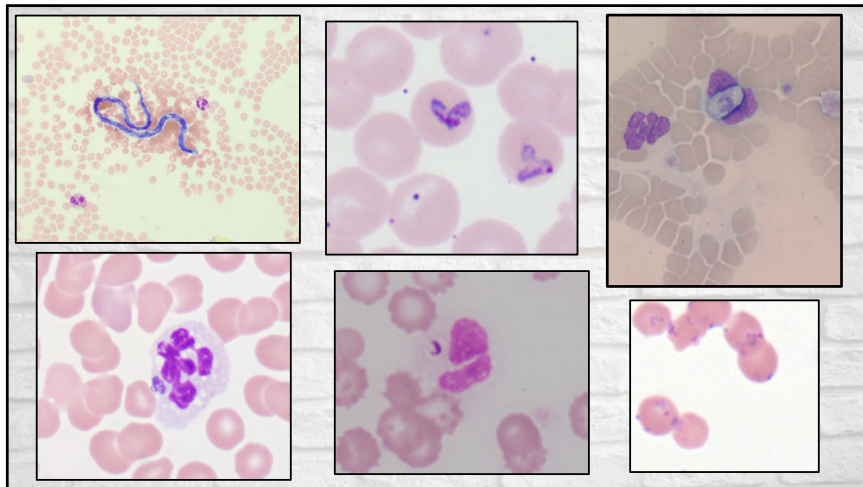


You'll get the **recording** via email within a few days



You can tell us what you thought in the **survey**

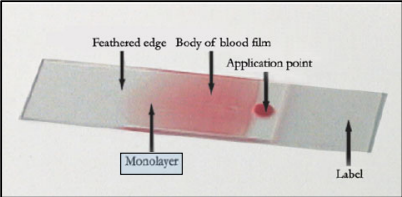
2



3

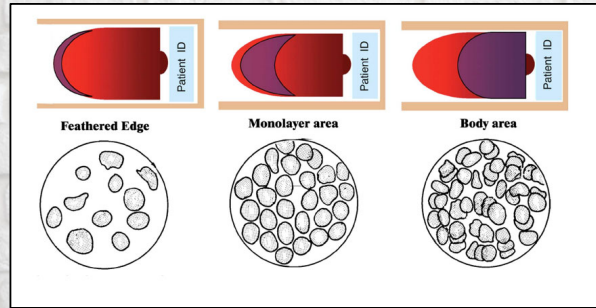
## Blood smear basics: Preparation

Smear a drop of blood on a glass slide to create a **monolayer**, in which individual cell morphology can be appreciated



4

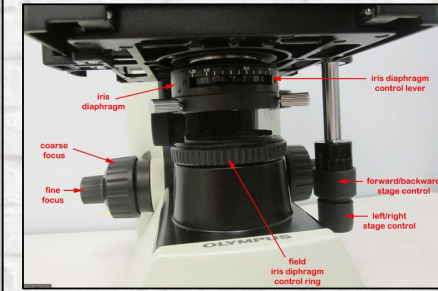
### Blood smear basics: Parts of a blood smear



5

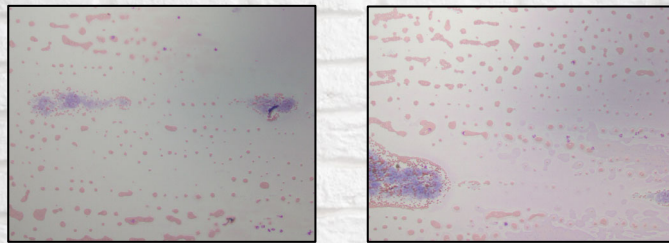
### Blood smear basics: Microscope setup

- Objective lenses:
  - 10x (Low power)
  - 40x dry OR 50x oil (Mid power)
  - 100x oil (High power)
- Iris diaphragm fully open
- Condenser raised to highest position



6

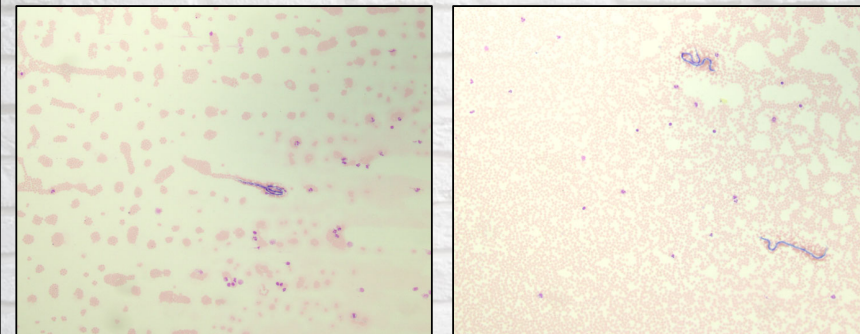
### Blood Smear Approach 10x Evaluation



- Briefly scan entire length of the feathered edge for:
- Platelet clumps
  - Microfilariae
  - Large/atypical cells

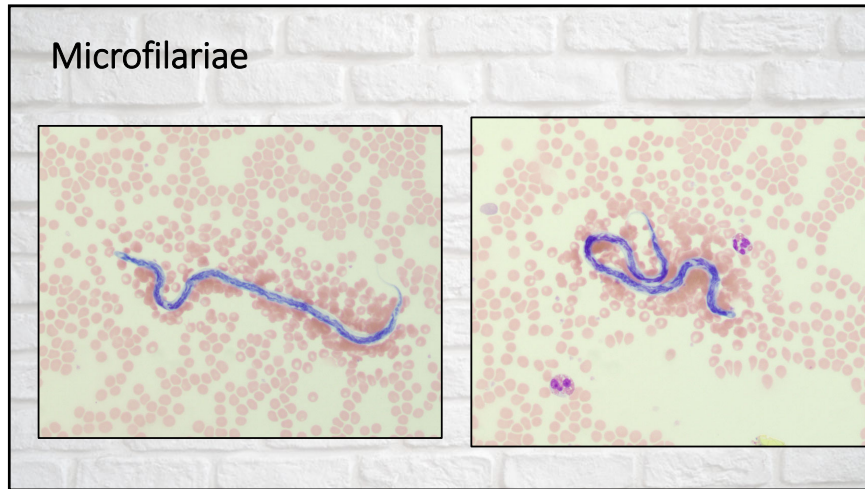
7

### Microfilariae



8





9

### *Dirofilaria immitis*

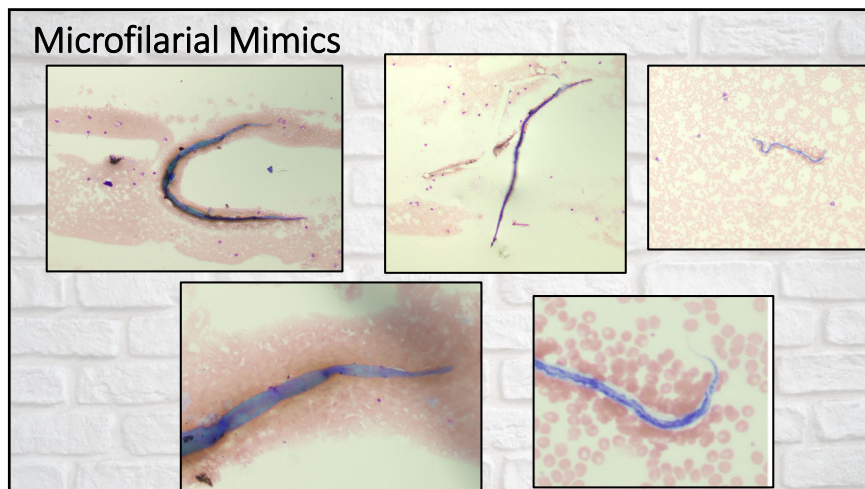
- Transmitted by mosquito vectors
- Antigen testing preferred
  - Some limitations
  - Recommended as follow-up to identification of microfilariae
- Other blood smear findings:
  - Eosinophilia and/or basophilia

**2019 HEARTWORM INCIDENCE**  
AMERICAN HEARTWORM SOCIETY

Average number of cases per reporting clinic:  
 □ 1-5 cases/clinic  
 □ 6-25 cases/clinic  
 □ 26-50 cases/clinic  
 □ 51-99 cases/clinic  
 □ 100+ cases/clinic

Image: American Heartworm Society  
<https://www.heartwormsociety.org/pet-owner-resources/incidence-maps>

10



11

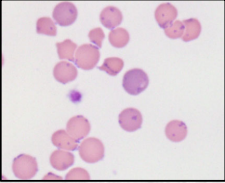
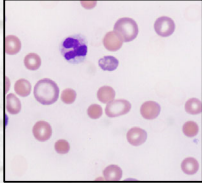
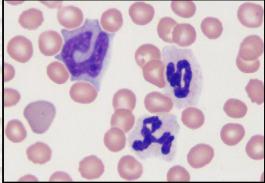
### Blood Smear Approach 40x/50x Evaluation

Focus on the monolayer:

- ~50% of RBC should contact each other in normal monolayer

12

### Blood Smear Approach 100x Evaluation

Goals:

- Stay within the monolayer!
- Identify & quantify RBC/WBC morphology changes
- Most infectious organisms are best seen at 100x**

13

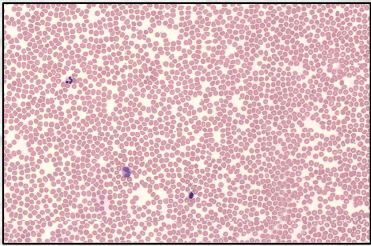
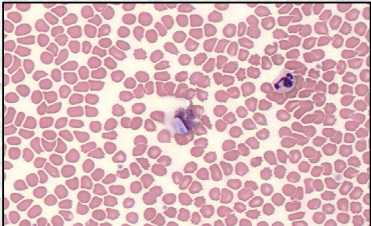
### Case 1: "Maya"

- 1.5 yr FS Border Collie Mix
- Adopted from shelter in Memphis, TN and brought to Chicago area
- Since adoption, waxing and waning fever
- New onset of skin lesions
- Presented for internal medicine consultation

14

### Case 1: "Maya"

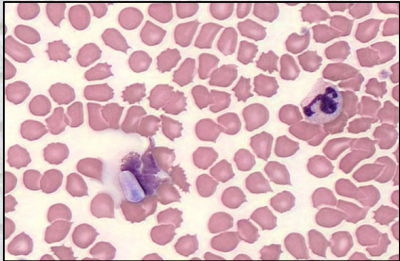
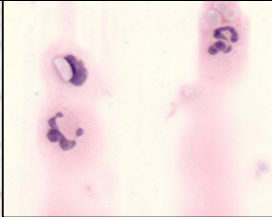
Test	Results	Reference Interval
Hematocrit	43.0	35.0 – 52.0%
WBC	6.71	6.00 – 17.00 x 10 <sup>3</sup> /μL
PLT	206	200 – 700 x 10 <sup>3</sup> /μL

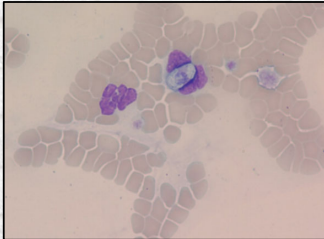
Is it repeatable?

15

### Case 1: "Maya"

PCR: *Hepatozoon canis*



Images courtesy of Dr. Ashlee Urbasic

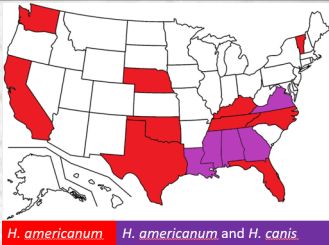
16





### *H. americanum*

- Endemic in southern USA
- Ingestion of **gulf coast tick**
- Clinical signs:
  - Fever, weight loss, lethargy, muscle atrophy
  - Hyperesthesia
  - Neutrophilic leukocytosis
    - May be extreme!
  - Periosteal reactions resembling hypertrophic osteoarthropathy



*H. americanum*    *H. americanum* and *H. canis*

### *H. canis*

- Global distribution
- Ingestion of **brown dog tick**
- Often subclinical
- Clinical signs may include fever, weight loss, lethargy, splenomegaly, and anemia

17

### Case 1: "Maya"

- Follow-up
  - Treated with imidocarb dipropionate protocol
  - Parasitemia relapsed after 3 months
  - Therapy ongoing

18

### Case 2:

#### "Sadie"

- 5 yr FS Golden Retriever
- Undergoing CHOP chemotherapy protocol for multicentric large-cell lymphoma
- CBC prior to chemotherapy treatment

#### "Oscar"

- 14 yr MC Puggle
- Presented on emergency basis for acute onset of lethargy and decreased appetite
- CBC/Serum Chemistry/Urinalysis performed

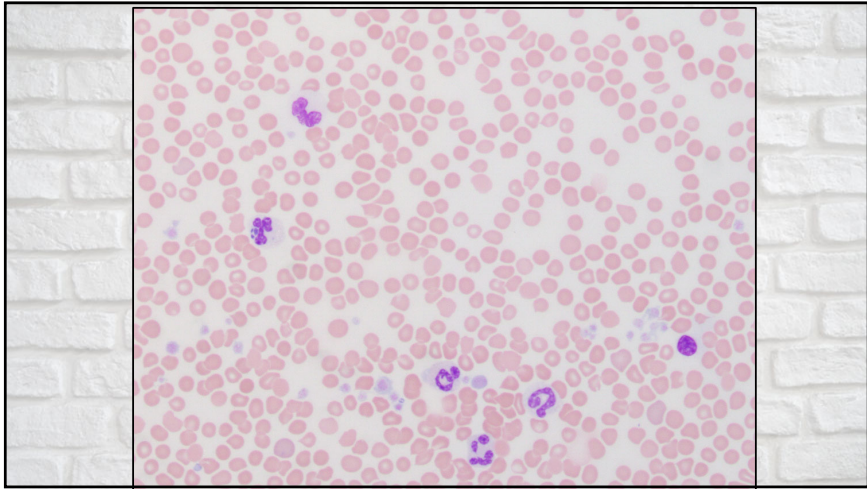
19

"Sadie"

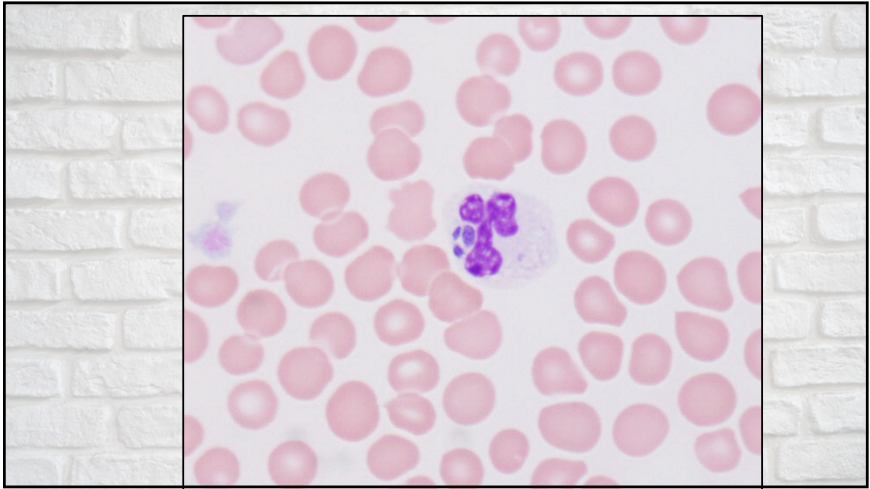
"Oscar"

Test	Results	Reference Interval	Test	Results	Reference Interval
Hct	36.1	35.0 – 52.0%	Hct	36.4	35.0 – 52.0%
WBC	3.35 L	6.00 – 17.00 x 10 <sup>3</sup> /μL	WBC	8.40	6.00 – 17.00 x 10 <sup>3</sup> /μL
PLT	5 L	200 – 700 x 10 <sup>3</sup> /μL	PLT	72 L	200 – 700 x 10 <sup>3</sup> /μL

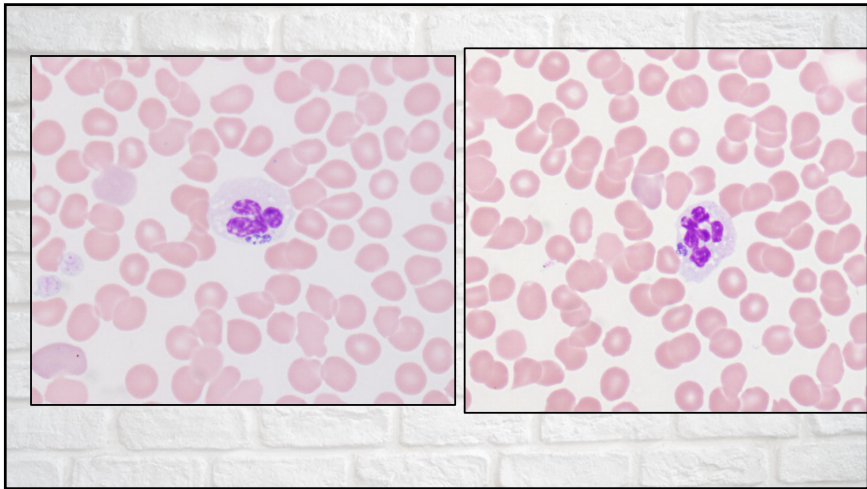
20



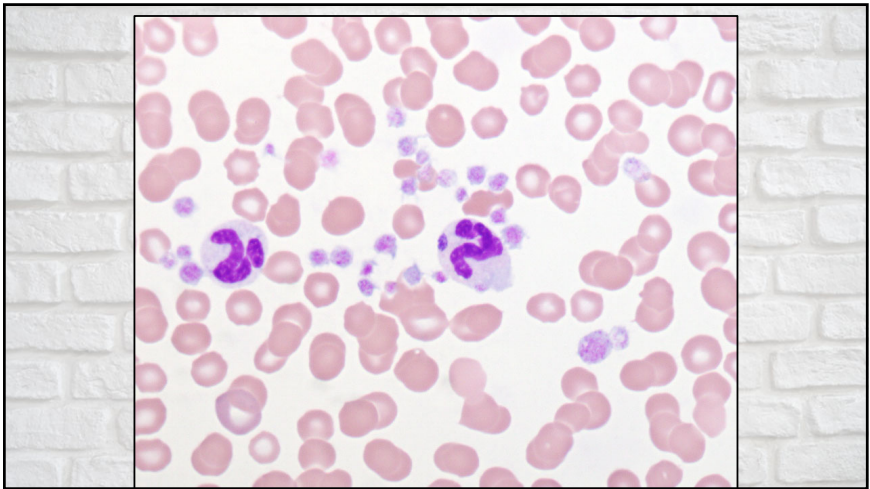
21



22



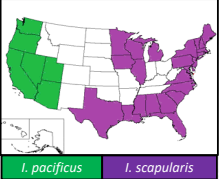
23




24



## E. ewingii & Anaplasma phagocytophilum



*I. pacificus*    *I. scapularis*



**E. ewingii and A. phagocytophilum are cytologically indistinguishable**

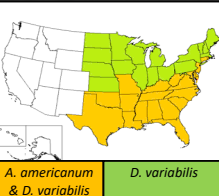
- Morulae within granulocytes (mainly neutrophils)
- Antibody testing or PCR for differentiation

**Vectors:**

*A. phagocytophilum*: Ixodes spp. ticks  
*E. ewingii*: lone star tick (*A. americanum*) and American dog tick (*D. variabilis*)

**Clinical signs:**

- May be clinically silent
- Fever, lymphadenomegaly, splenomegaly, **thrombocytopenia**
  - Petechiae or clinical bleeding uncommon
- Variable anorexia, depression, arthritis, hindlimb or scrotal edema



*A. americanum & D. variabilis*    *D. variabilis*

25

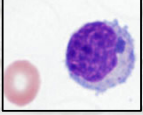
## Ehrlichia canis

**Morulae within mononuclear cells**

**Vector: Brown dog tick (*R. sanguineus*)**

**Clinical signs:**

- Similar acute signs as *E. ewingii* or *A. phagocytophilum*, with potential for more severe clinical disease
- Subclinical phase
- Chronic phase
  - Pancytopenia
    - Thrombocytopenia with risk for clinical bleeding
  - Polyclonal or monoclonal gammopathy
  - Glomerulonephritis/Renal failure
  - Marked splenomegaly
  - Other signs may vary based on predominant organs affected



26

### Follow-up diagnostics

Test Name	Sadie's result	Oscar's Result
Heartworm Antigen	Negative	Negative
Ehrlichia Antibody	Negative	Negative
Lyme disease Antibody	Negative	Negative
Anaplasmosis Antibody	Negative	Negative

Test Name	Sadie's result	Oscar's Result
<i>Babesia</i> spp. PCR	Negative	Negative
<i>Ehrlichia</i> spp. PCR	Negative	Negative
<i>Anaplasma</i> spp. PCR	<b>POSITIVE</b> ( <i>A. phagocytophilum</i> )	<b>POSITIVE</b> ( <i>A. phagocytophilum</i> )
<i>Rickettsia</i> spp. PCR	Negative	Negative
<i>Hepatozoon</i> spp. PCR	Negative	Negative
<i>Leishmania</i> spp. PCR	Negative	Negative
<i>Neorickettsia</i> spp. PCR	Negative	Negative
<i>Bartonella</i> spp. PCR	Negative	Negative

27

## Case 2: Follow-up

- "Oscar"
  - Infection likely peracute
    - Antibodies not yet detectable
- "Sadie"
  - Multiple follow-up antibody tests remained negative
    - Likely immunocompromised secondary to lymphoma and chemotherapy
    - Unable to mount antibody response

**Both patients currently undergoing treatment with doxycycline**

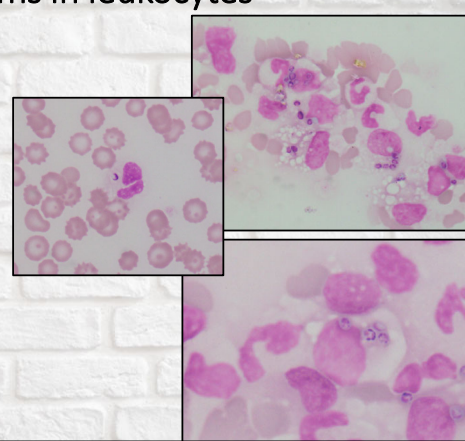
28



### Other infectious organisms in leukocytes

*Histoplasma capsulatum*

- Uncommonly identified on blood smears
- Most often detectable in cases of bone marrow infiltration
- Best identified at feathered edge

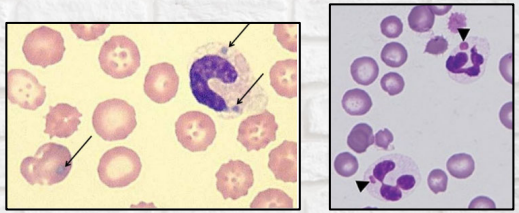


29

### Other infectious organisms in leukocytes

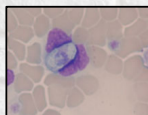
- Canine distemper virus inclusions
- Transient and rarely encountered, but pathognomonic

- Bacteremia
- Intracellular bacteria extremely rare

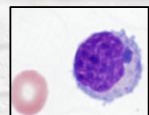


30


### Infectious organisms of leukocytes



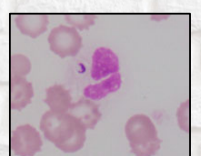
Hepatozoon spp.



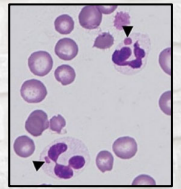
Ehrlichia canis



Ehrlichia ewingii or Anaplasma phagocytophilum



Histoplasma capsulatum



Canine distemper virus inclusions

31

### Case 3: "Cora"

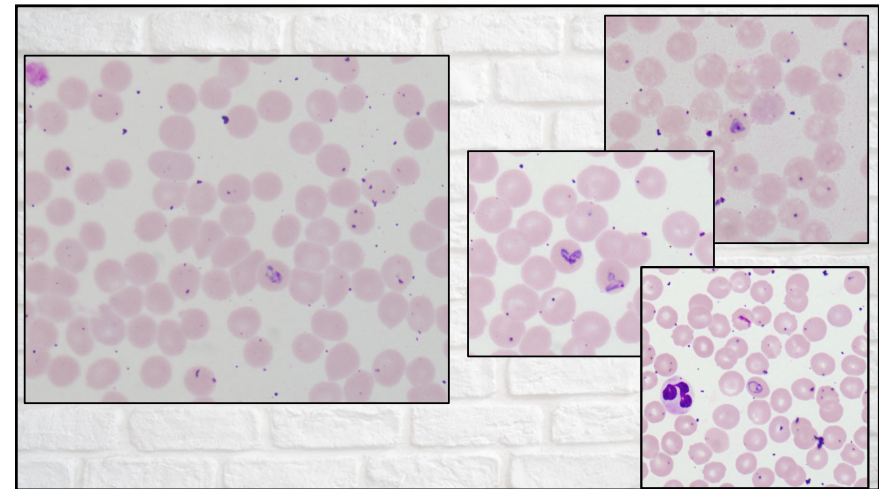
- 5 yr FS Collie
- Increased respiratory effort
- Persistent hypercalcemia
- Treated with prednisone – no clinical improvement

32

### Case 3: "Cora"

Test	Results	Reference Interval
Hematocrit	44.7	35.0 – 52.0%
WBC	19.8 <b>H</b>	6.00 – 17.00 x 10 <sup>3</sup> /μL
Neut	18.41 <b>H</b>	3.00 – 11.50 x 10 <sup>3</sup> /μL
Lymph	0.40 <b>L</b>	1.00 – 4.80 x 10 <sup>3</sup> /μL
PLT	14 <b>L</b>	200 – 700 x 10 <sup>3</sup> /μL

33



34

### *Babesia canis*

"Large form"

Subspecies	Endemic region	Pathogenicity	Clinical Signs	Tick vector
<i>B. canis vogeli</i>	Worldwide, including USA (mostly southern USA)	Low	Often subclinical +/- thrombocytopenia	Brown dog tick
<i>B. canis canis</i>	Europe and Asia	Intermediate	Hemolytic anemia & thrombocytopenia	Ornate cow tick
<i>B. canis rossi</i>	South Africa	High	Peracute to acute hemolytic anemia & SIRS ARDS/MODS in severe cases	Yellow dog tick

Traditional classification based on pathogenicity and endemic region...

35

### ...but it's not that simple

J Vet Intern Med 2010;24:127-131

**Babesiosis Caused by a Large *Babesia* Species in 7 Immunocompromised Dogs**

L.E. Sikorski, A.J. Birkenheuer, M.K. Holowaychuk, A.L. McCleary-Wheeler, J.M. Davis, and M.P. Littman

Detection and molecular characterization of a novel large *Babesia* species in a dog

A.J. Birkenheuer<sup>a,\*</sup>, J. Neel<sup>b</sup>, D. Ruslander<sup>a</sup>, M.G. Levy<sup>a</sup>, E.B. Breitschwerdt<sup>a</sup>

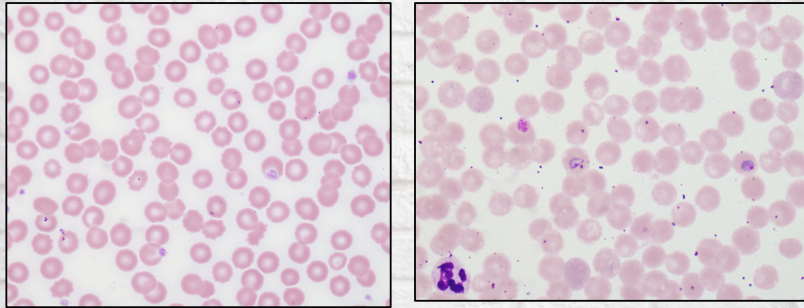
<sup>a</sup>Department of Clinical Sciences, College of Veterinary Medicine, North Carolina State University, 4700 Hillsborough St., Raleigh, NC 27606, USA  
<sup>b</sup>Department of Molecular and Biomedical Sciences, College of Veterinary Medicine, North Carolina State University, 4700 Hillsborough St., Raleigh, NC 27606, USA  
Received 4 July 2006; accepted 15 July 2006

- At least one other large-form *Babesia* subspecies has been identified in immunocompromised dogs in the USA via PCR and sequencing
- Most cases previously splenectomized or undergoing chemotherapy
- Associated with anemia and thrombocytopenia
- Specific tick vector unknown
- "*Babesia sp. (coco)*"

36



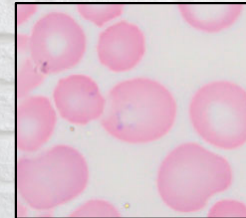
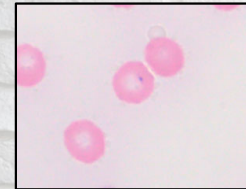
### Piroplasm or platelet?



37

### *Babesia gibsonii*

#### “Small form”



- Most commonly affects Pit Bull terriers
- Pathogenesis not well established
  - Brown dog tick likely vector in USA
  - Several reports of transmission via bite wounds from infected dogs
- Often subclinical
- Severe hemolytic anemia may occur in young or immunocompromised dogs
  - Many symptomatic dogs have positive Coombs' test +/- autoagglutination
  - +/- moderate to marked thrombocytopenia
- Challenging to identify on blood smear
  - PCR widely available

38

### Case 3: “Cora”

#### Follow-up:

- Mediastinal mass
  - Presumed mediastinal lymphoma vs. less likely thymoma
- Additional diagnostics not pursued due to financial constraints
- Babesiosis
  - Immunosuppression?
  - Doxycycline and azithromycin prescribed
- Returned to rDVM for continued care, lost to follow-up

39

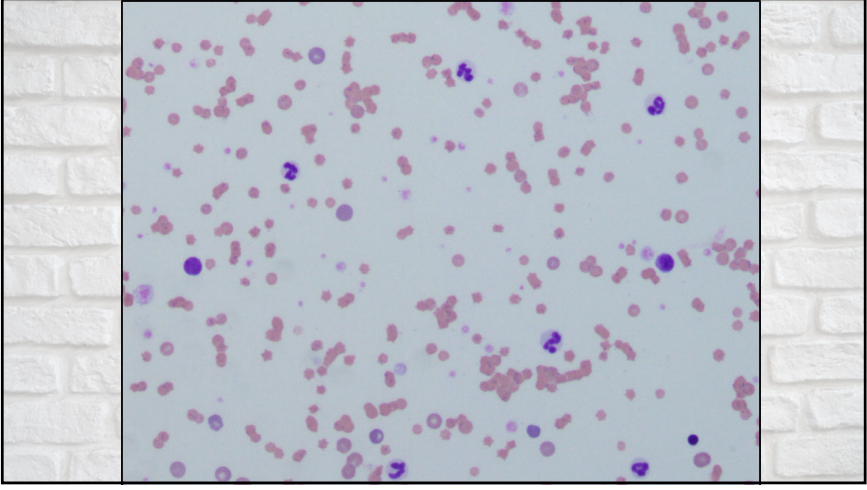
### Case 4: “Biscuit”

- 1 yr MI DSH Cat
- Primarily outdoor cat, lives with multiple other outdoor cats and often fights with feral cats
- Presented after being found nonresponsive in yard
- Laterally recumbent on presentation, live fleas noted
  - FeLV snap +
  - PCV 15%

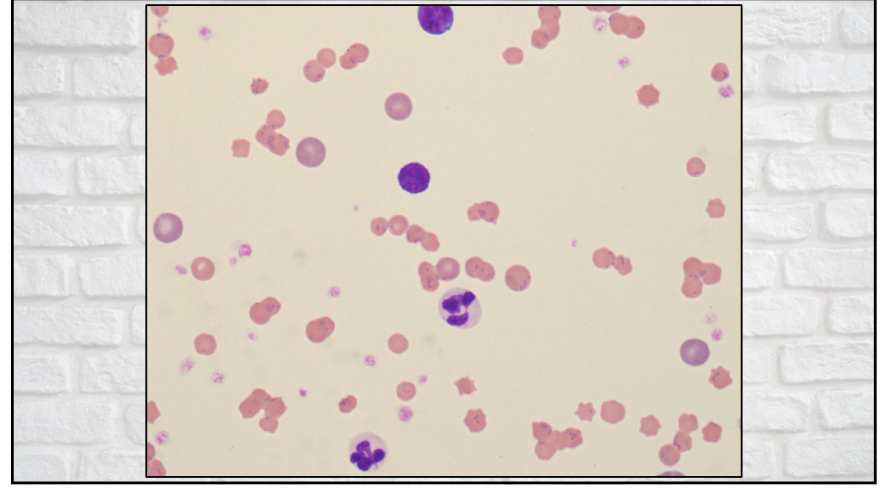
Test	Results	Reference Interval
Hematocrit	12.5 <b>L</b>	30.0 – 45.0%
WBC	20.6 <b>H</b>	5.50 – 19.50 x 10 <sup>3</sup> /μL
Neut	15.47 <b>H</b>	2.50 – 12.50 x 10 <sup>3</sup> /μL
Band	2.47 <b>L</b>	0.00 – 0.30 x 10 <sup>3</sup> /μL
PLT	196 <b>L</b>	300 – 700 x 10 <sup>3</sup> /μL

40

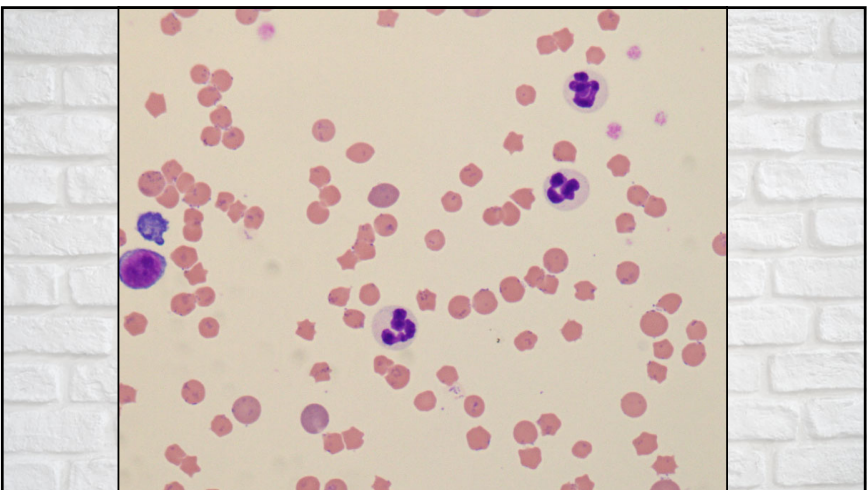




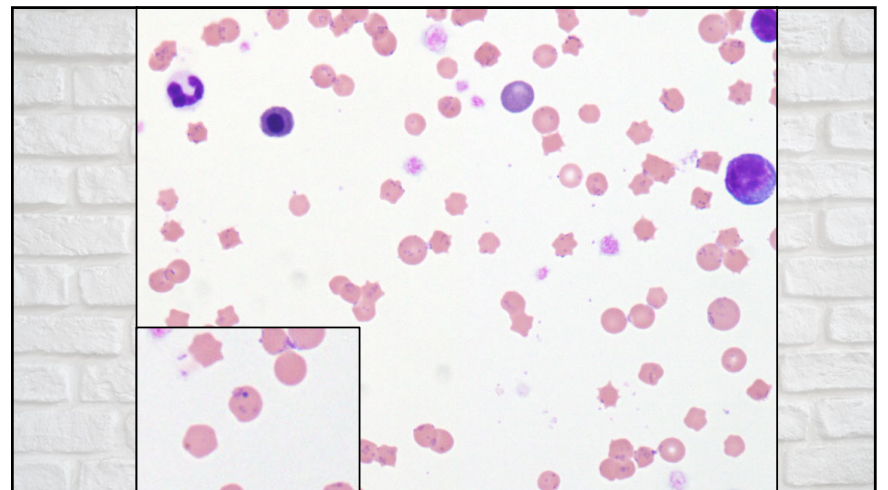
41



42



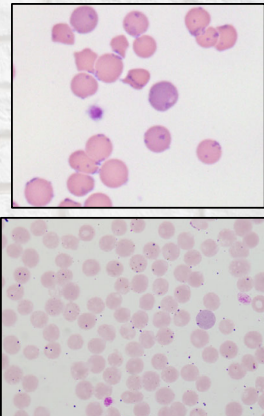
43



44

### *Mycoplasma* spp.

- *Epicellular* organisms
- Transmitted by arthropod vectors
  - Commonly fleas
  - Direct transmission via fighting and vertical transmission also documented in cats
- *M. hemofelis* causes acute hemolytic anemia in immunocompetent cats
- PCR for confirmation
  - Can be challenging to differentiate from stain debris

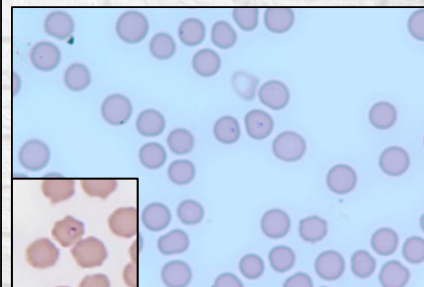


45

### Another important hemoparasite of feline RBC...

#### *Cytauxzoon felis*

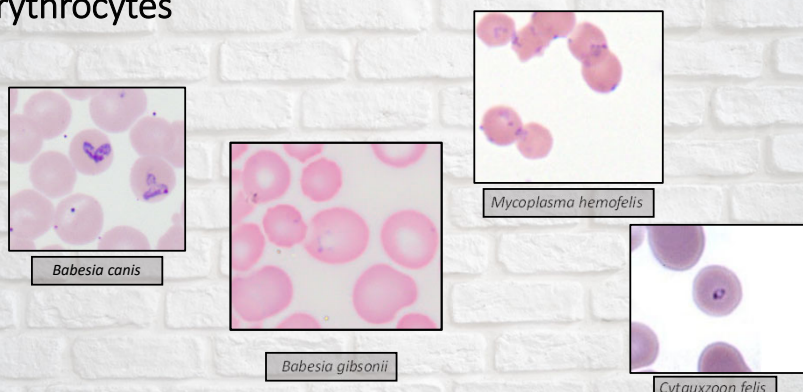
- Transmitted by lone star tick
- Bobcat reservoir host
- Causes severe clinical disease
  - Pyrexia, icterus, lymphadenomegaly, severe hemolytic anemia
- Often fatal
  - Piroplasms on blood smears apparent 1-3 days before death
  - Tissue schizonts rarely seen at feathered edge



Images courtesy of Dr. Anne Barger

46

### Infectious organisms of canine and feline erythrocytes



*Babesia canis*

*Babesia gibsonii*

*Mycoplasma hemofelis*

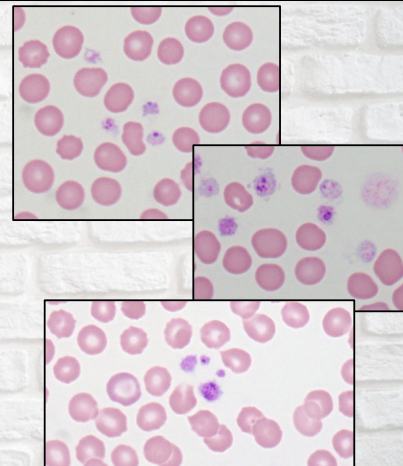
*Cytauxzoon felis*

47

### Don't forget the platelets!

#### *Anaplasma platys*

- Transmitted by brown dog tick
- Infectious cyclic thrombocytopenia
  - May manifest as clinical bleeding, especially as epistaxis
- Otherwise typically subclinical
- May be challenging to identify and differentiate from aggregated platelet granules
- PCR for confirmation
  - Some antibody tests cross-react with *A. phagocytophilum*



48

# Questions?

Remember to **download the CE certificate** in the handouts panel of the webinar control panel.

NOTE: CE certificate not available for watching the recording.

Questions about CE? [events@heska.com](mailto:events@heska.com)

Thank you for joining us!

©2021 Heska Corporation. All rights reserved. HESKA, Element POC are registered trademarks of Heska Corporation in the United States and other Countries. All other trademarks are the property of their respective owners.

