University of Saskatchewan

Lindee Climo

Clinical Pathology in Practice

Nicole Fernandez, DACVP

November 3, 2018

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My Desk

University of Saskatchewan

Clinical Pathology in Practice

- Ask the audience
- Sample handling
- Cytology
- Hematology

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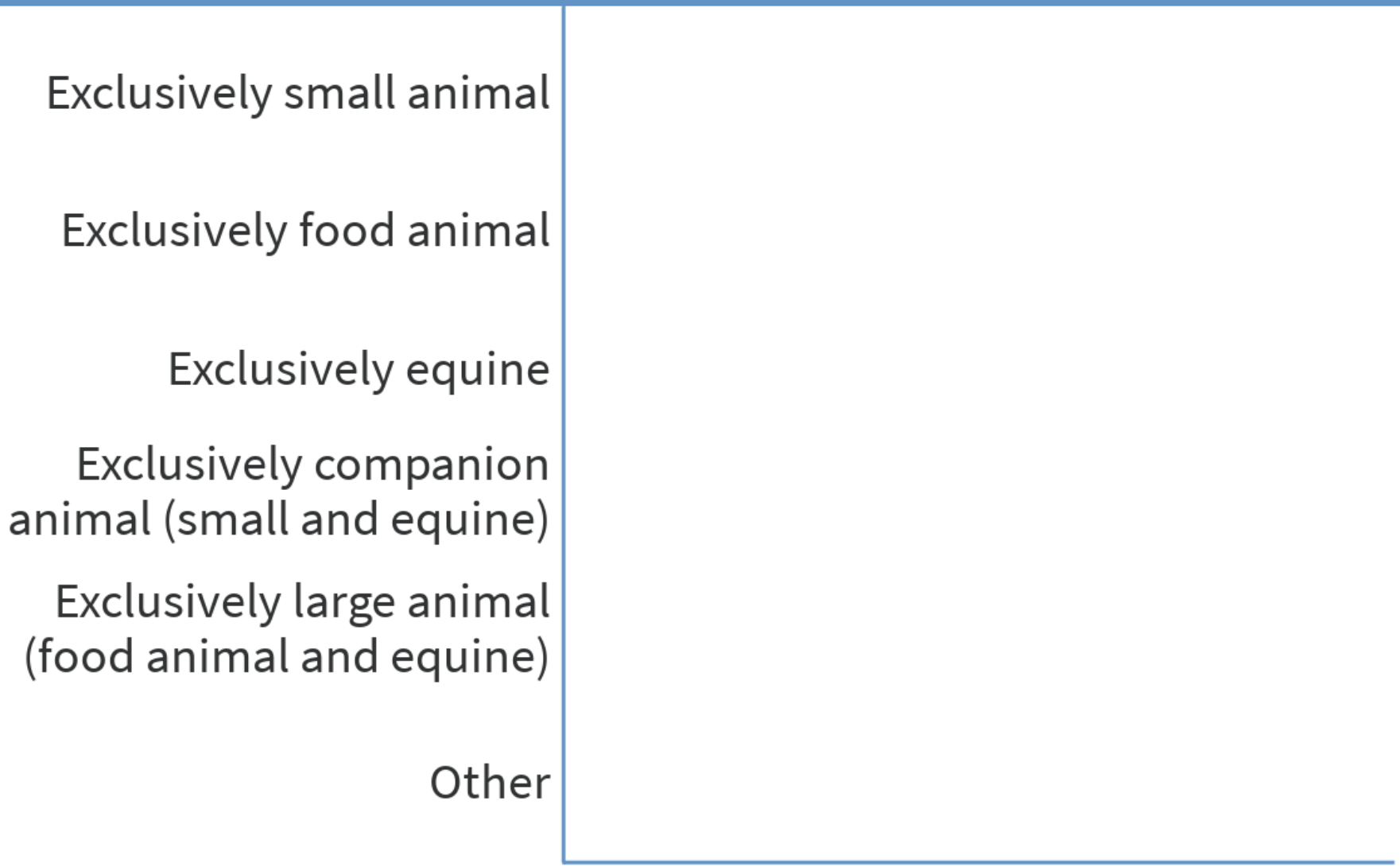
Ask the Audience

- From your smart phone, tablet, or laptop

What is your practice type?



Why did you decide to attend this session?

Clin path is AWESOME

I am comfortable with cytology and hematology but could use a review

I have forgotten all my cytology and hematology from vet school

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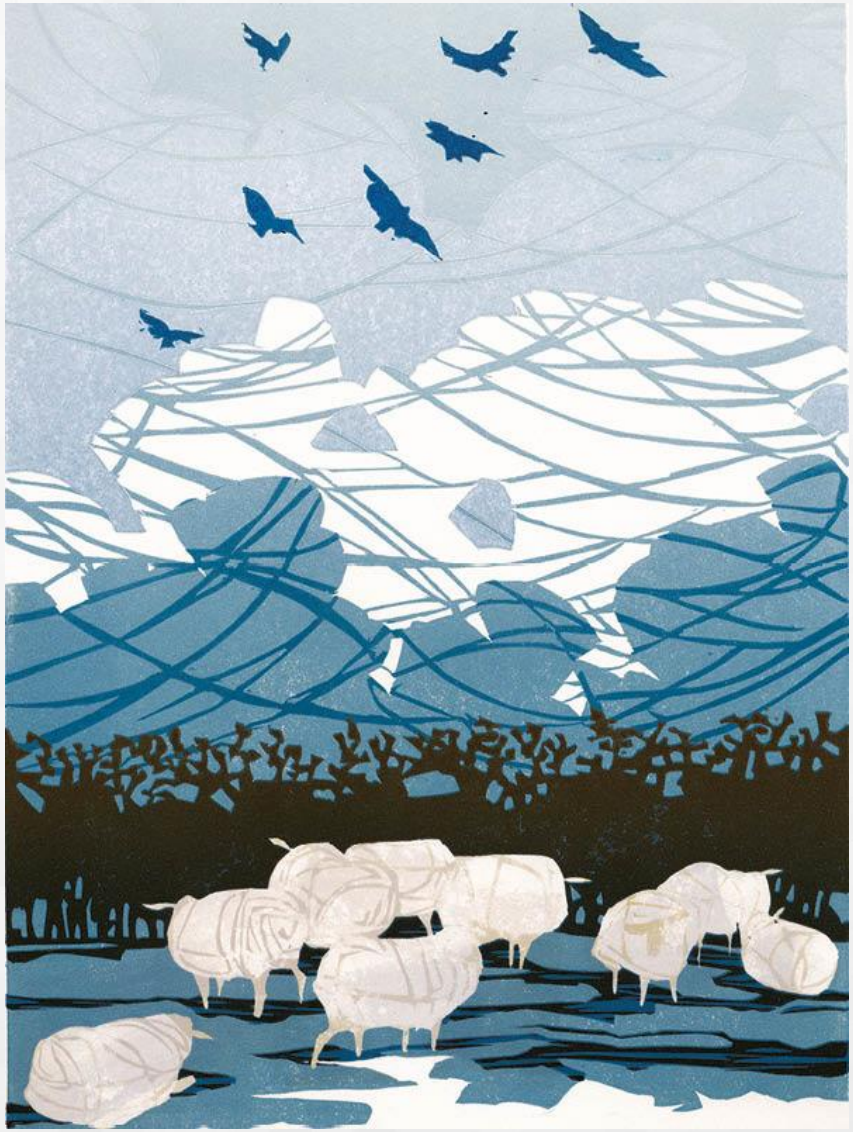
Sample Handling

Resources

Cytology samples

Hematology samples



Laura Boswell

Sample Handling

- PDS Protocol Manual is online
- Search “pds protocol manual”
- http://pdsinc.ca/Portals/0/Protocol%20Manual%202017%20with%20bookmarks.pdf

Sample Handling

PDS lab techs recommend:

- Separate bag for each patient
- Label containers with patient name, date, sample type
  - Is this serum or urine?
- Wipe off outside of containers
- More padding than you think

Sample Handling

Cytology smears

- Air dry slides
- Adequate padding/proper containers
- Pack separately from histopath samples
  - Applies to blood smears too

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Sample Handling

Fluid samples

- Submit in EDTA
- Make direct smears
- Keep cool

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Sample Handling

Hematology samples

- CBC- EDTA tube
- Make blood smears

- Chemistry- red top tube
- Separate serum from cells if possible

- Keep cool

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Cytology

Sample collection

Approach to the slide

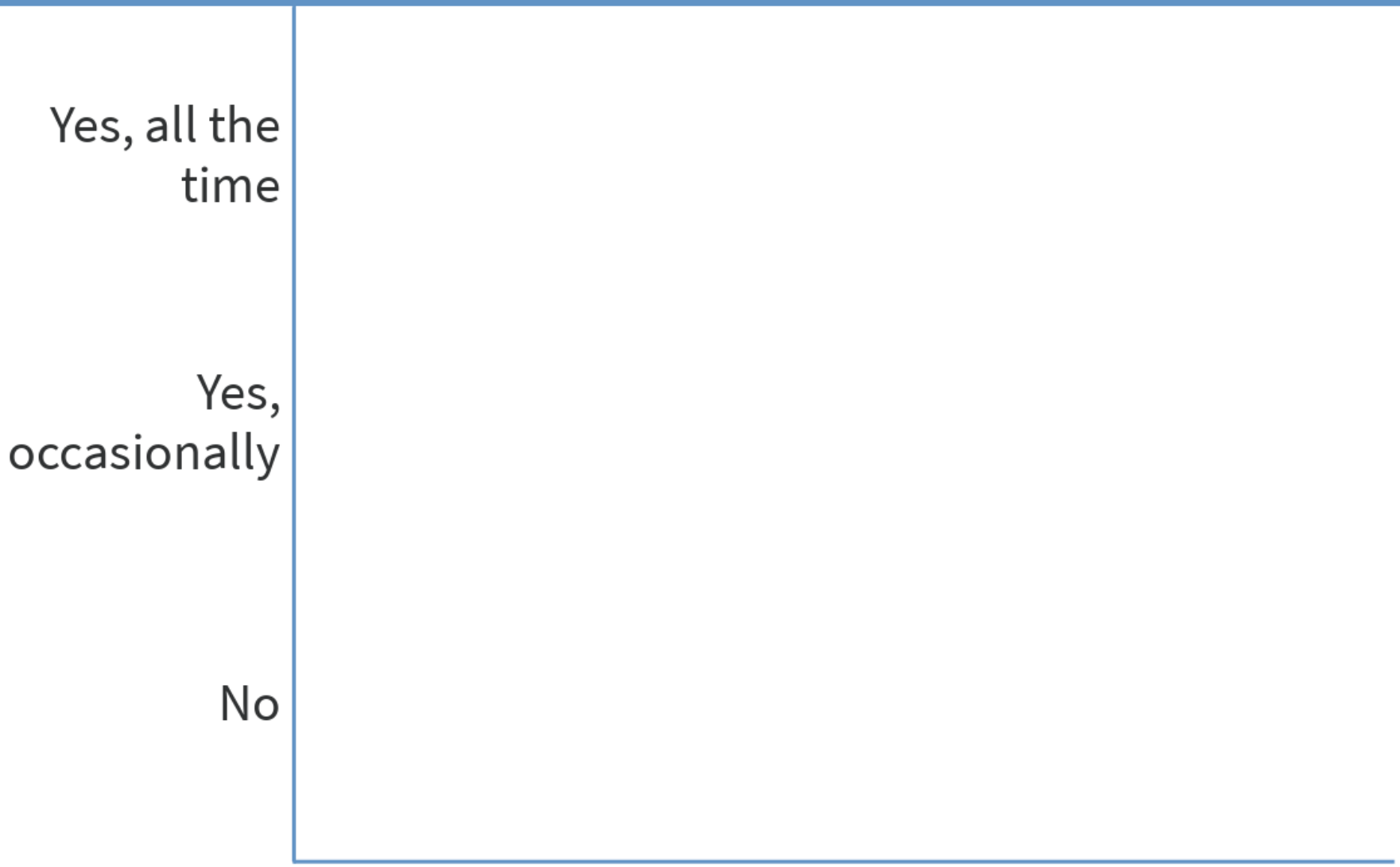
A few lumps and bumps

Owen Fink

Sample Collection

- FNA and/or FNNA
  - Videos in PDS Protocol Manual
- Make smears quickly
- Air dry
- Stain with Diff-Quik

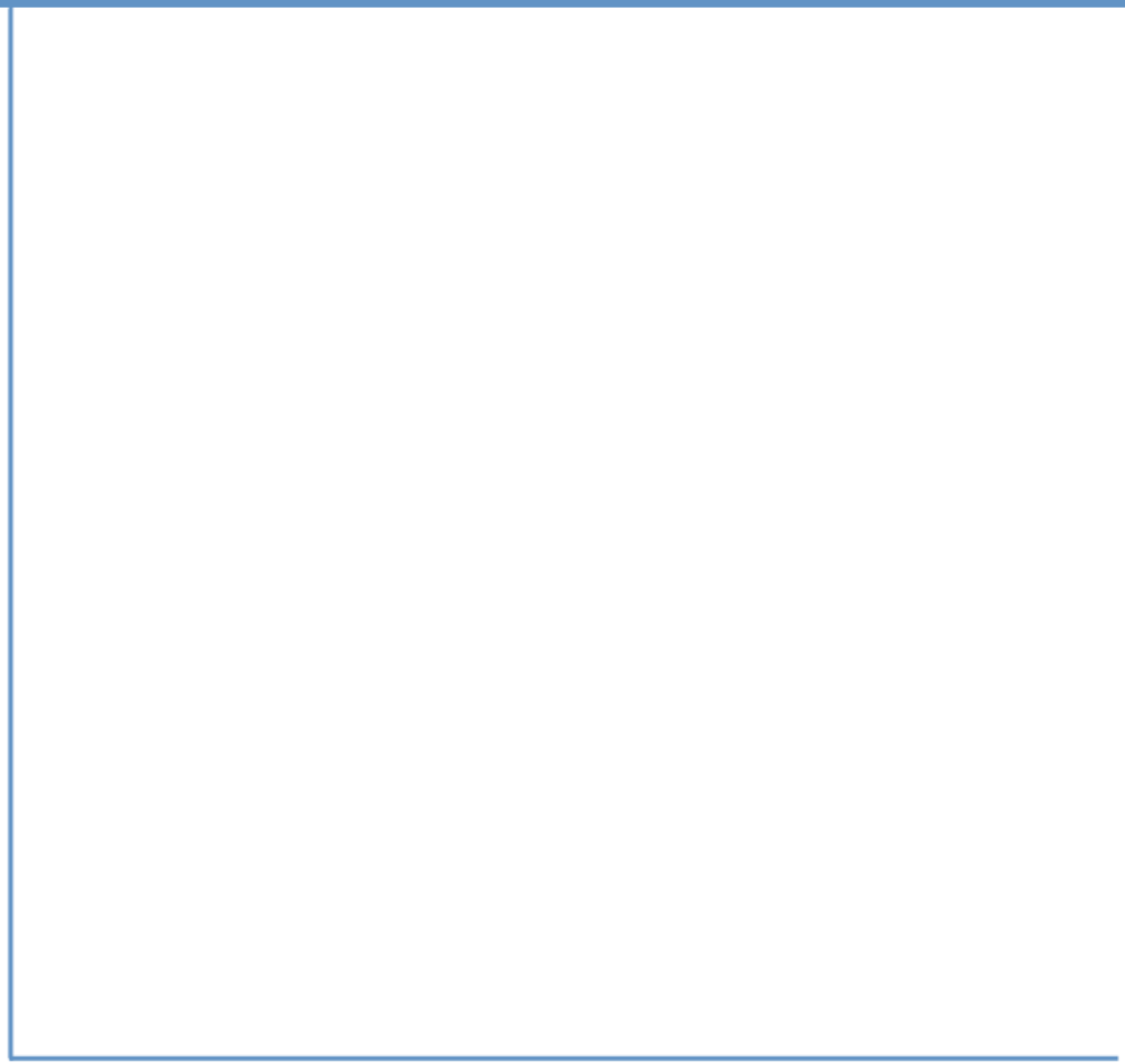
Do you collect cytology samples in-house?



Respond at **PollEv.com/nicolefernan414**

Text **NICOLEFERNAN414** to **37607** once to join, then **A, B, or C**

Do you read your own cytology samples?



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Approach to the Slide

1. Gross Examination
   - How cellular is the slide?
   - Where on the slide is the material distributed?
   - Is blood present?
2. 4x objective
   - Scan entire smear
   - Find areas to evaluate at higher magnification
     - ➢ Intact cells spread in a thin layer
3. 10x-40x objective
   - Evaluate several areas identified at 4x
   - Identify cell types present
   - Classify the lesion (see Figure 1)
4. 100x objective (oil)
   - Confirm identification of cells
   - Evaluate cellular details
   - Identify bacteria

About the images

- Cases from WCVM and UCVM
- Many taken by Dr Galezowski

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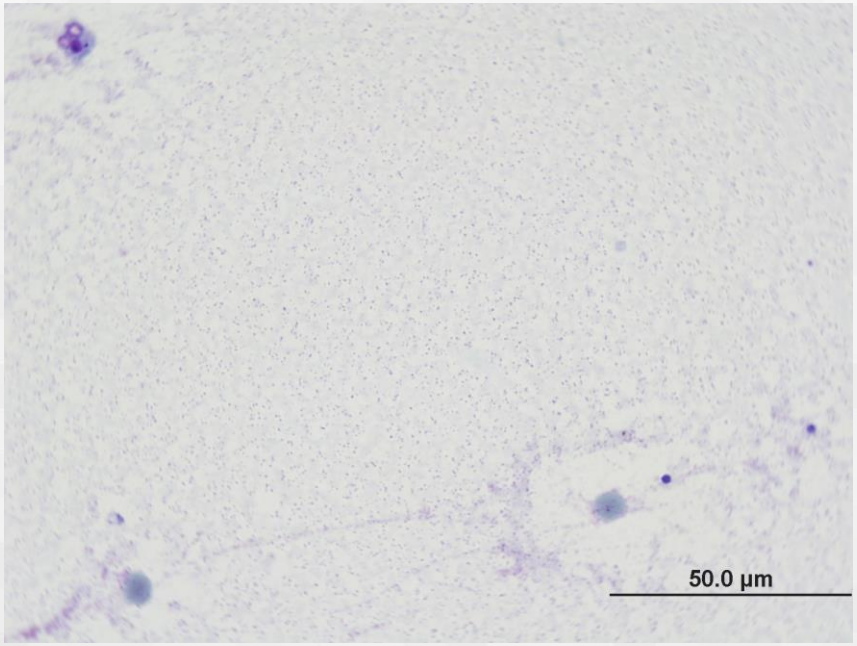
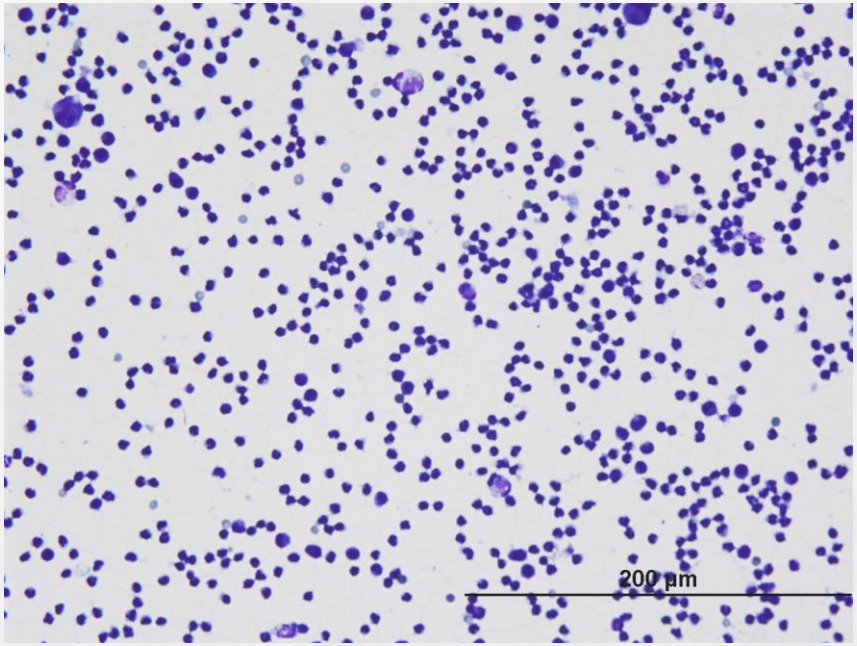
Cells needed…





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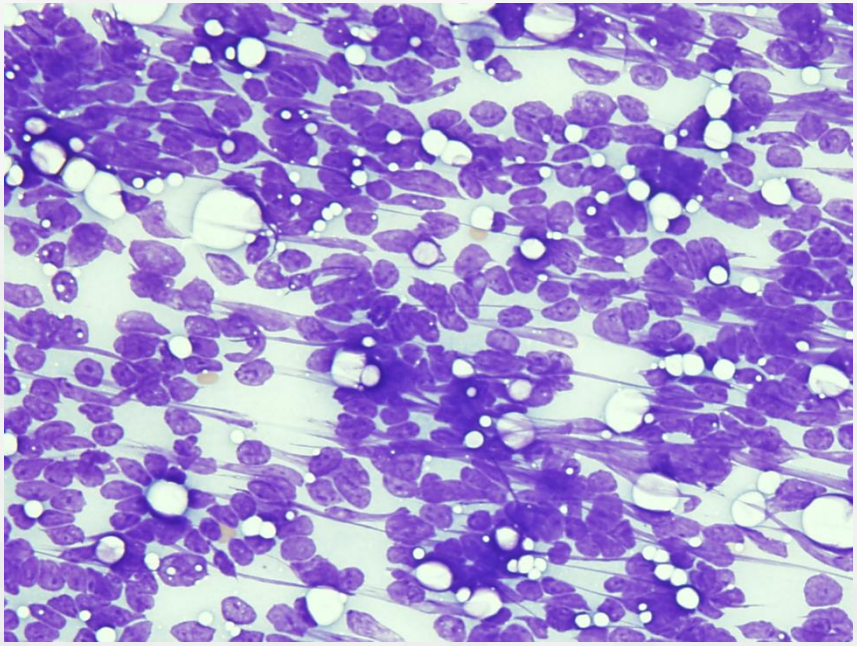
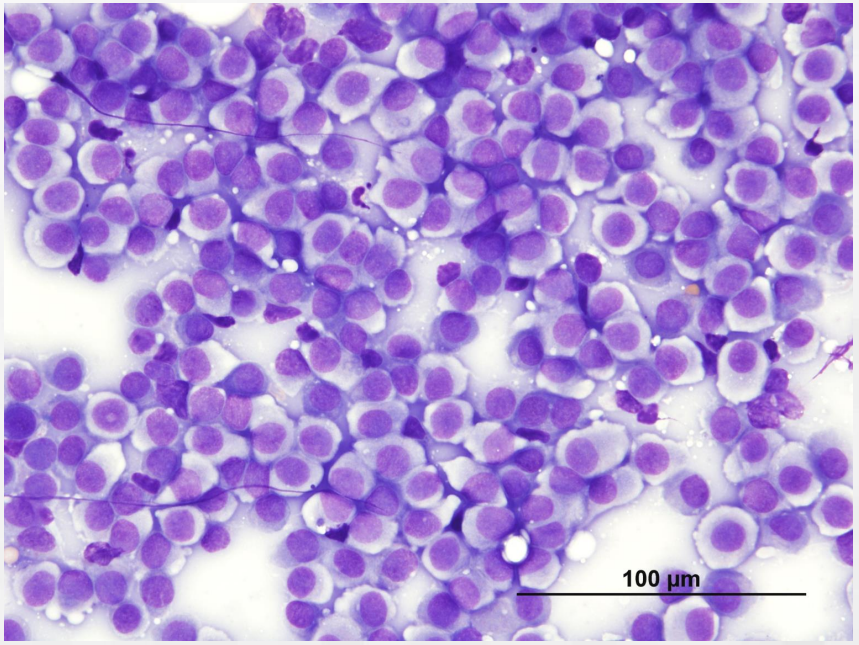
Intact cells needed…





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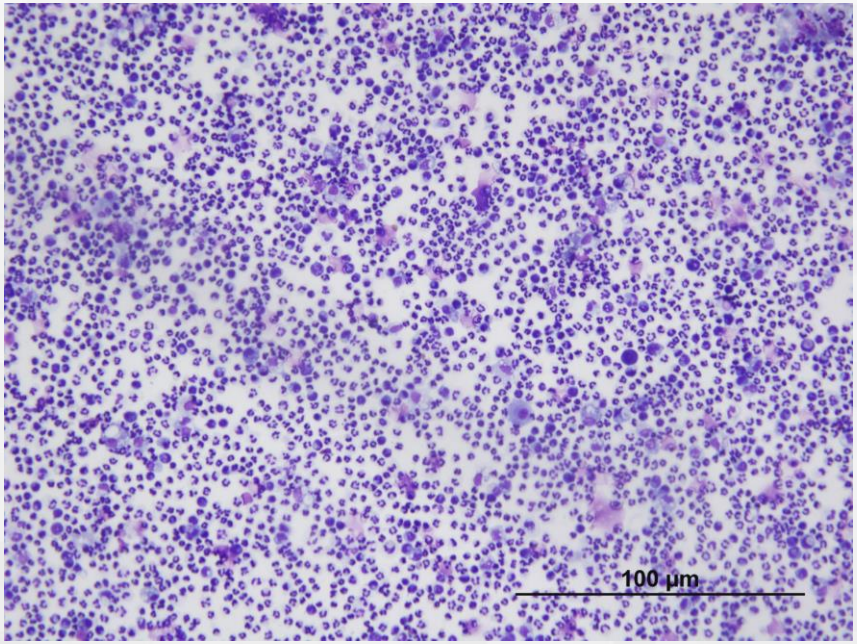
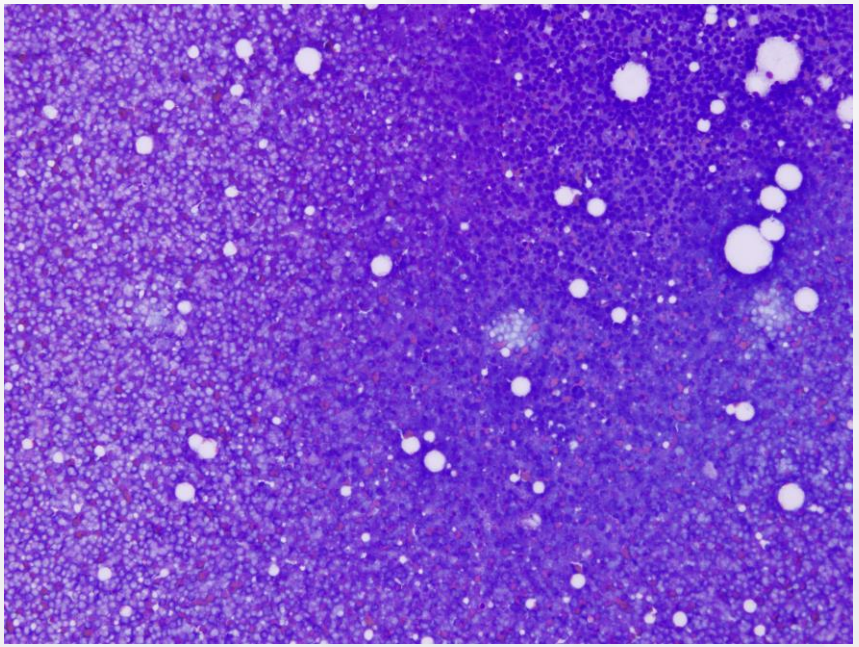
In a thin layer…





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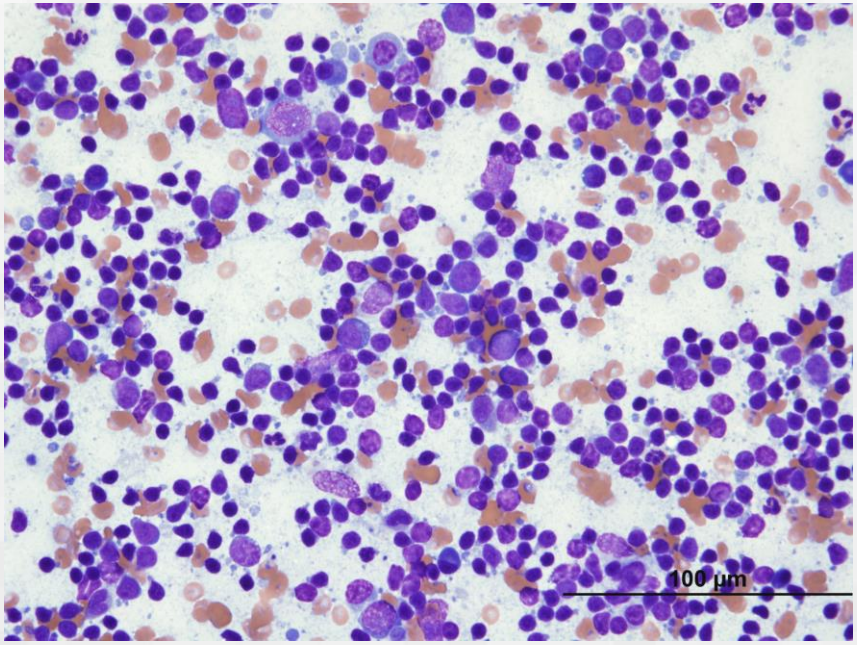
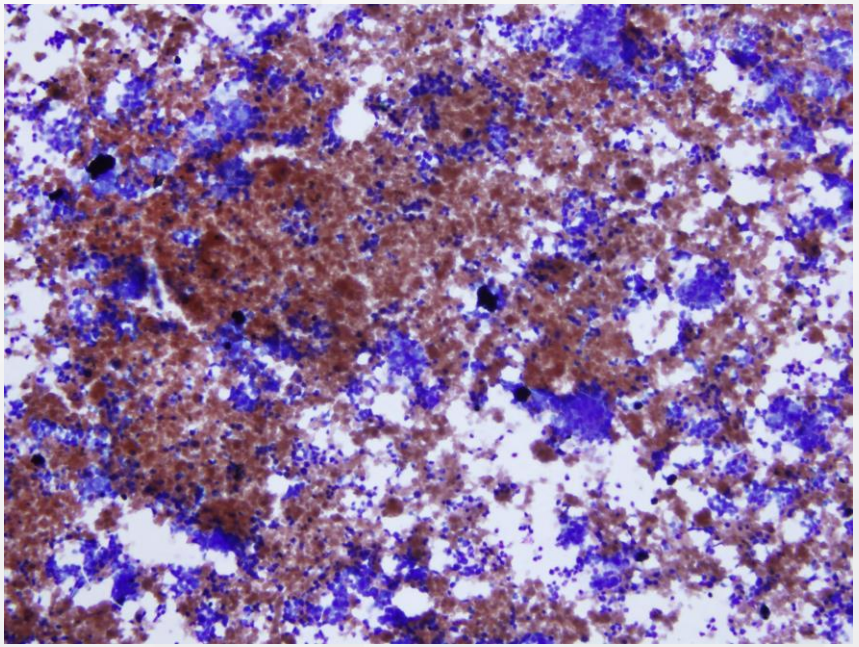
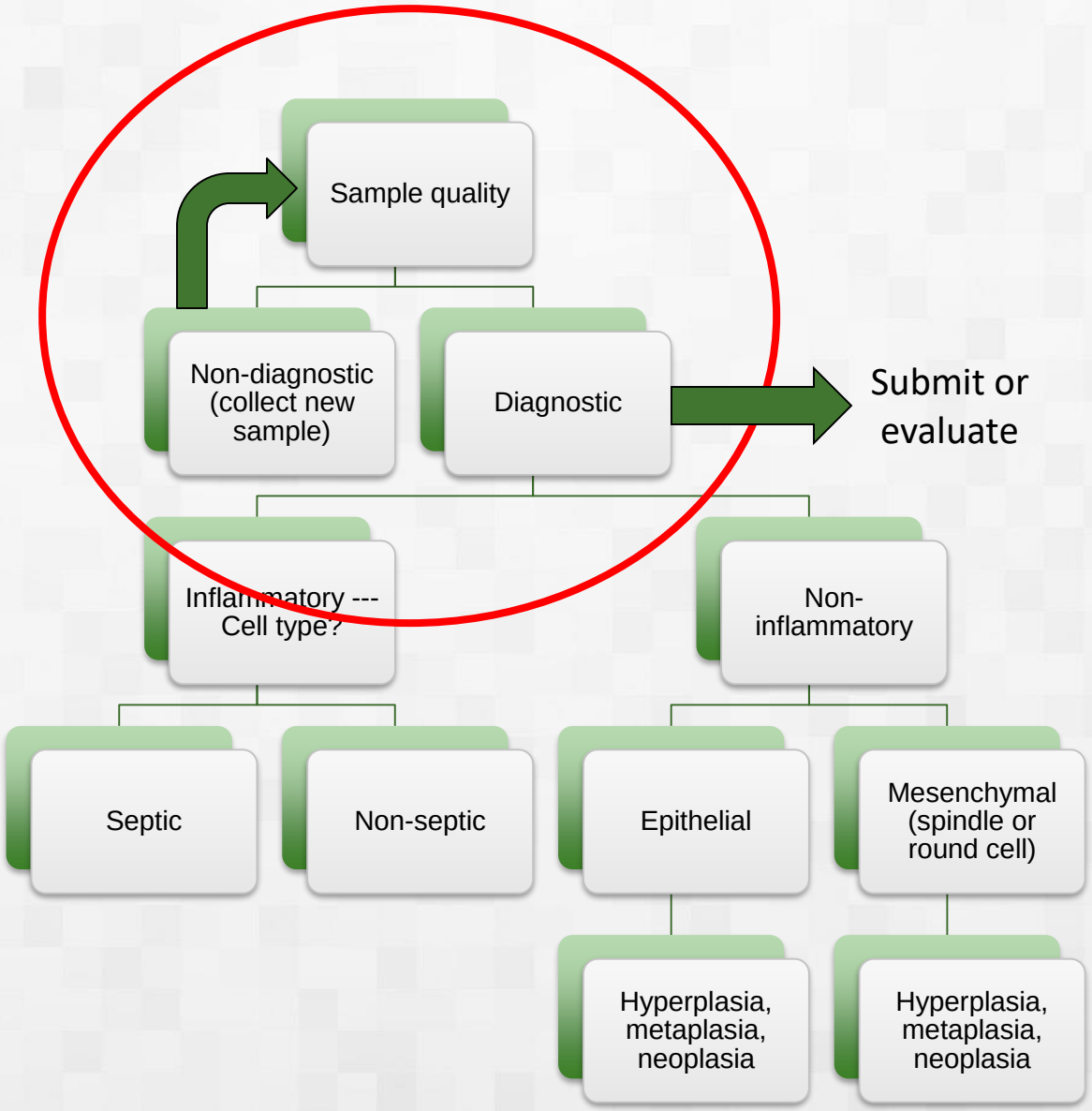
Without too much blood






Sample quality
Non-diagnostic (collect new sample)
Diagnostic
Submit or evaluate
Inflammatory --- Cell type?
Non-inflammatory
Septic
Non-septic
Epithelial
Mesenchymal (spindle or round cell)
Hyperplasia, metaplasia, neoplasia
Hyperplasia, metaplasia, neoplasia

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In-house Cytology: The Short List

Melanoma

Mast Cell Tumor

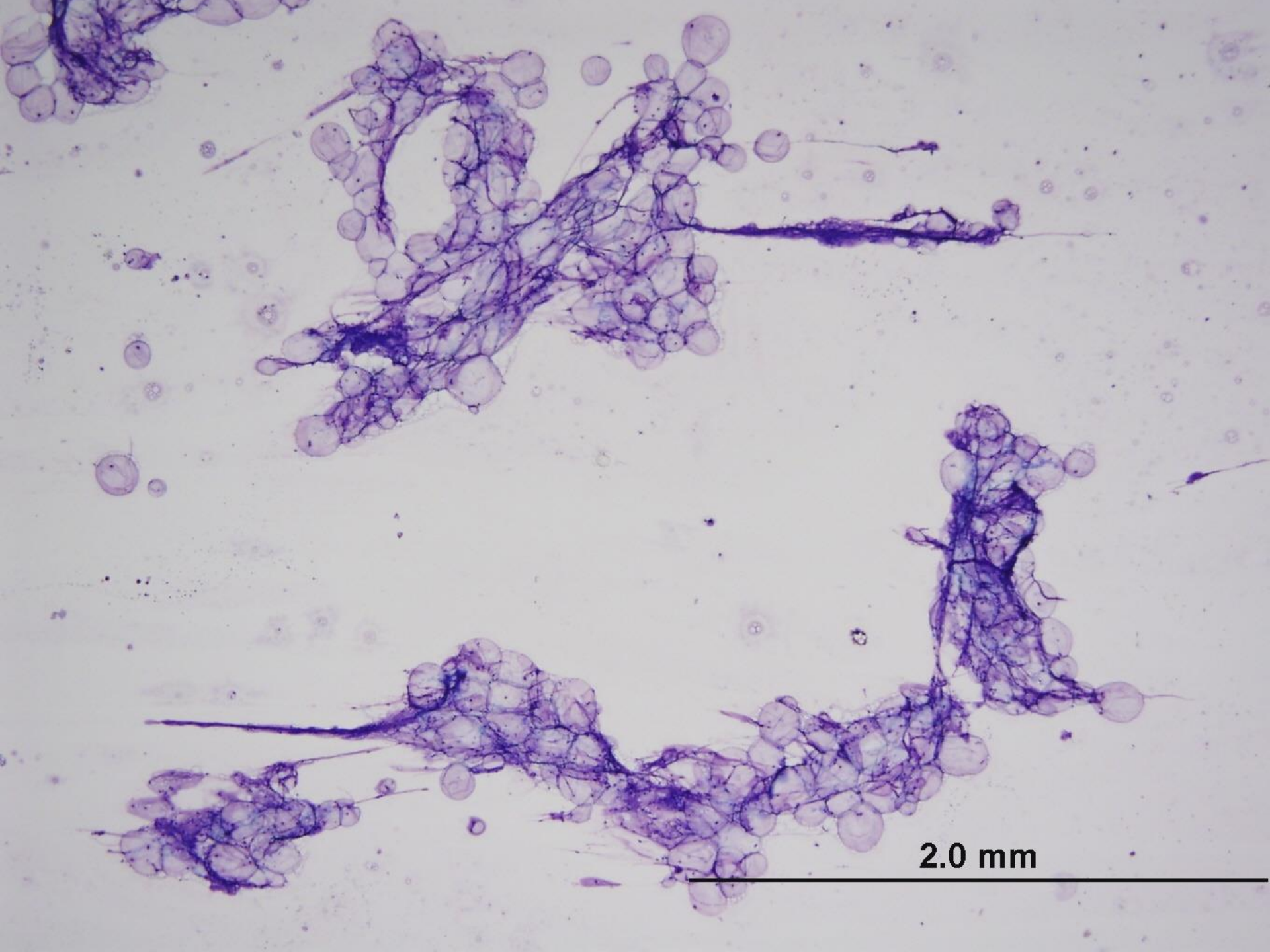
Lipoma

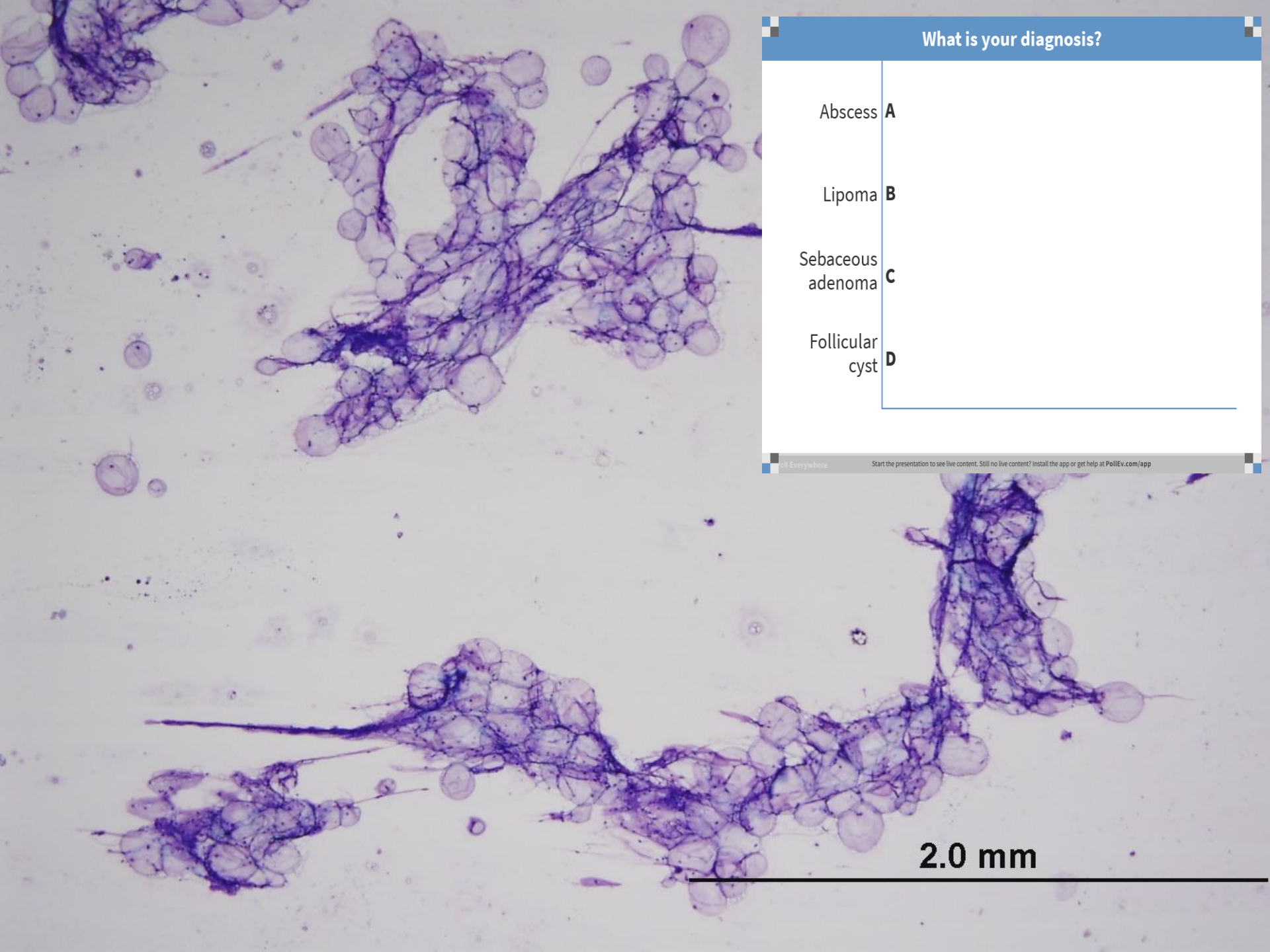
Sebaceous adenoma

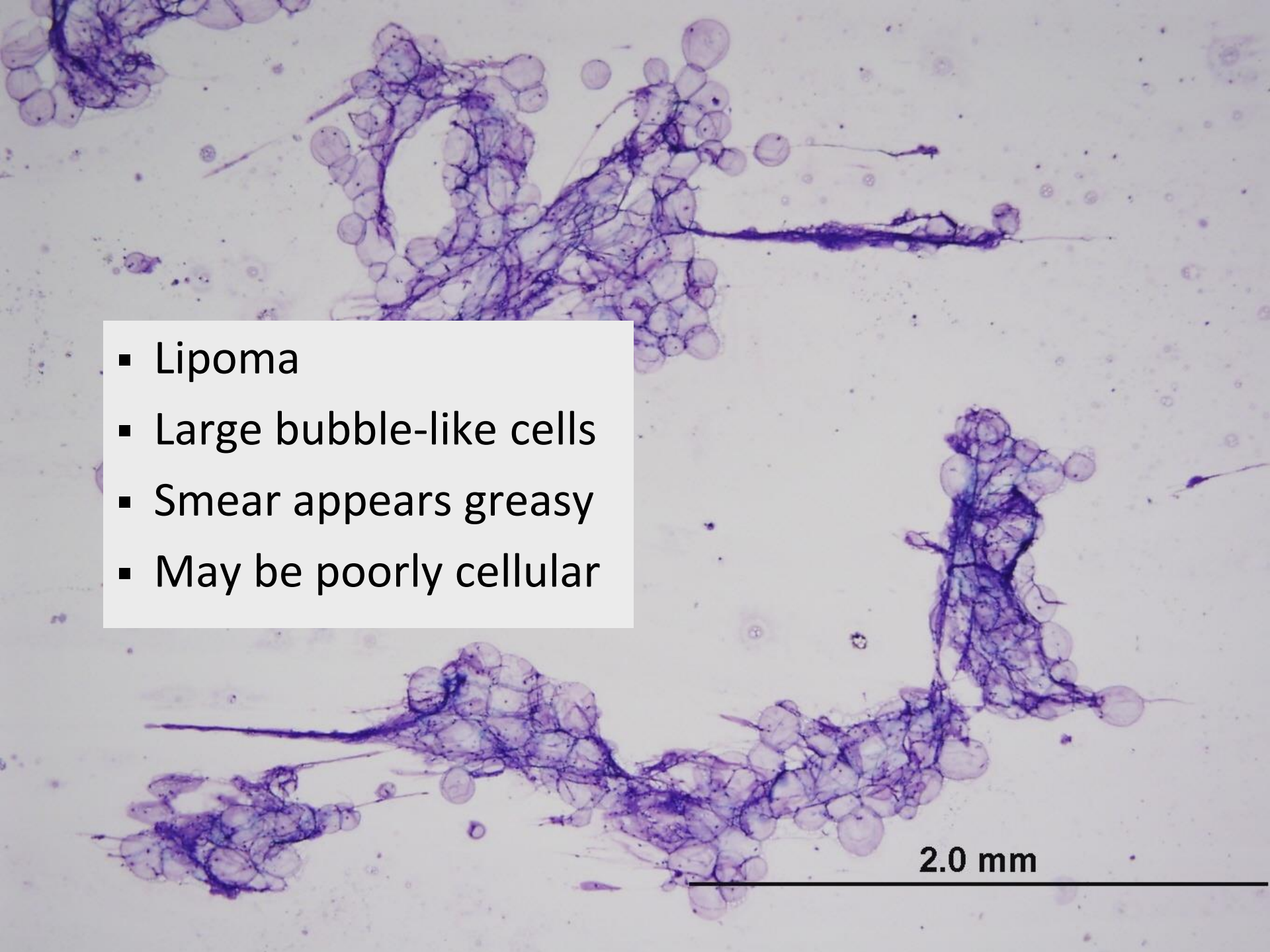
Follicular Cyst

Abscess

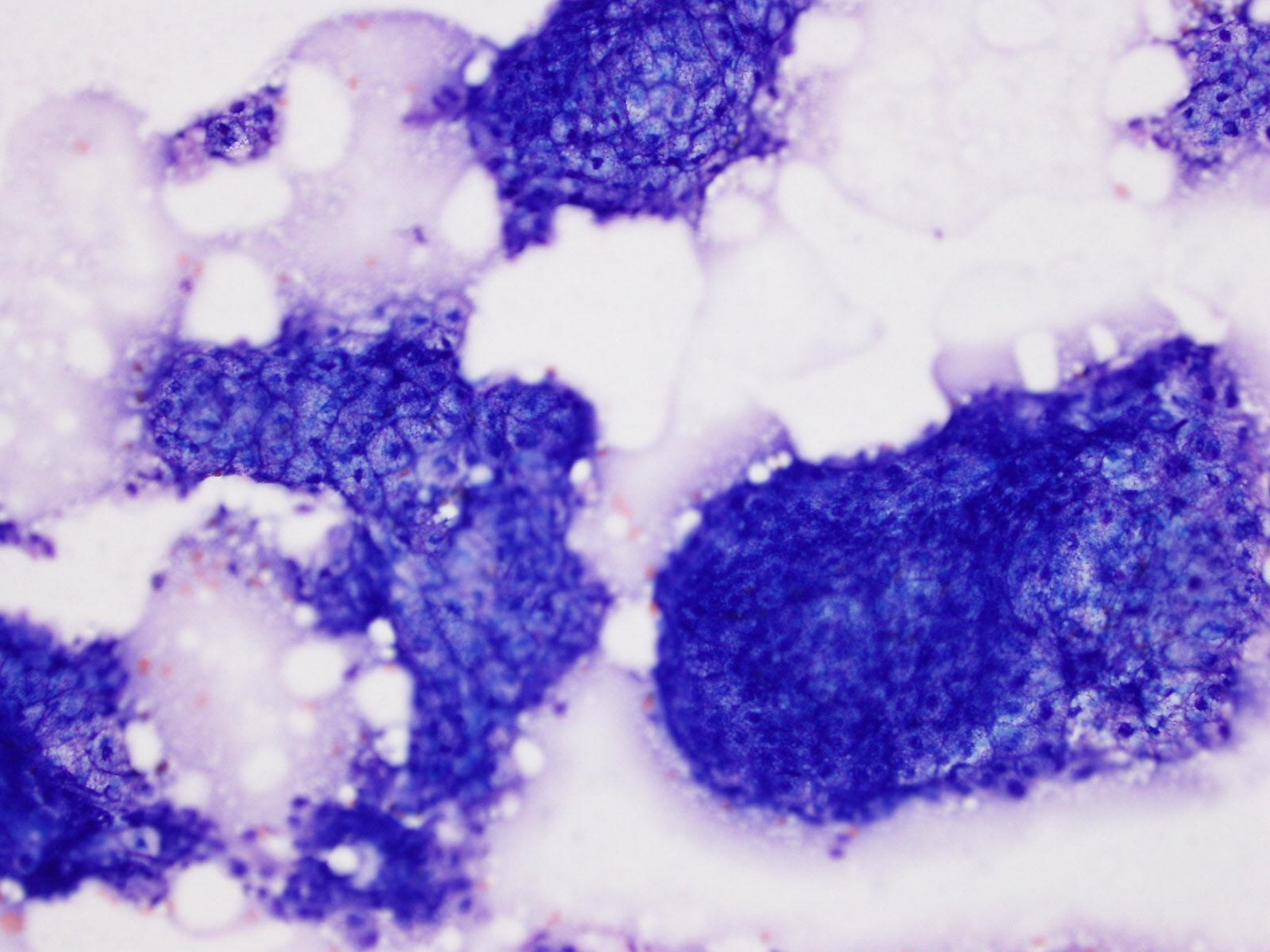
Septic Inflammation

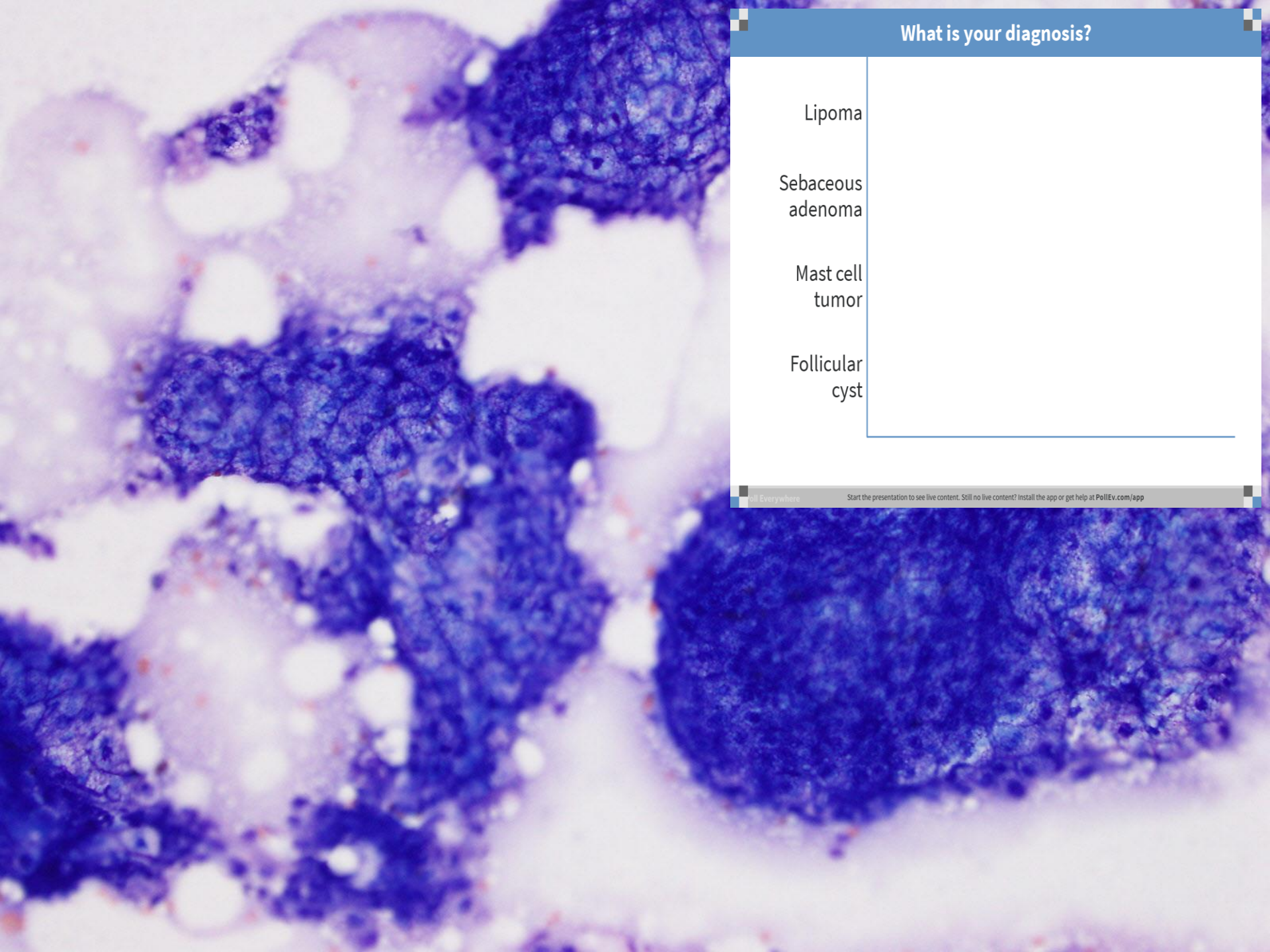

2.0 mm

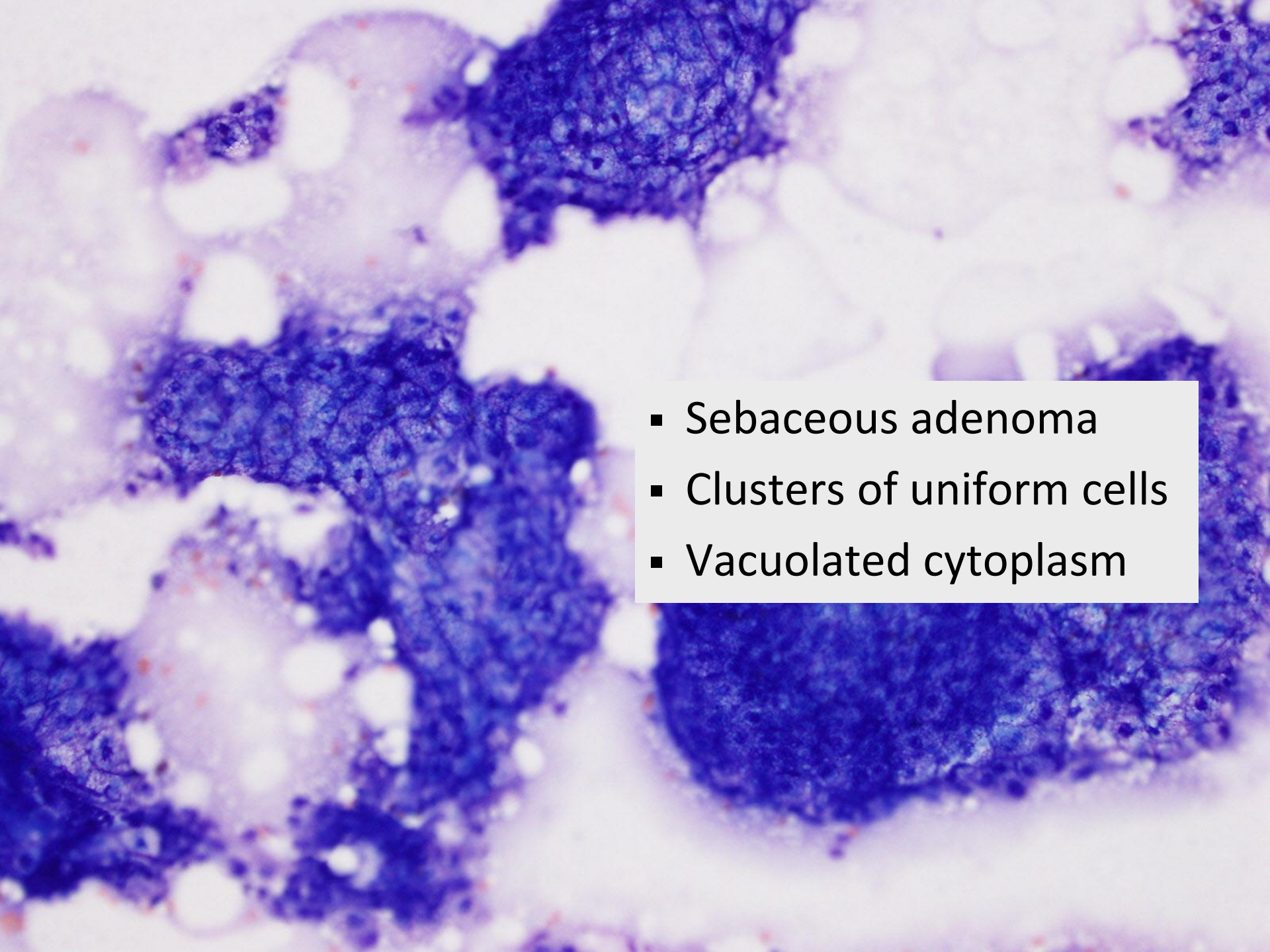

What is your diagnosis?
Abscess A
Lipoma B
Sebaceous adenoma C
Follicular cyst D
Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app
2.0 mm

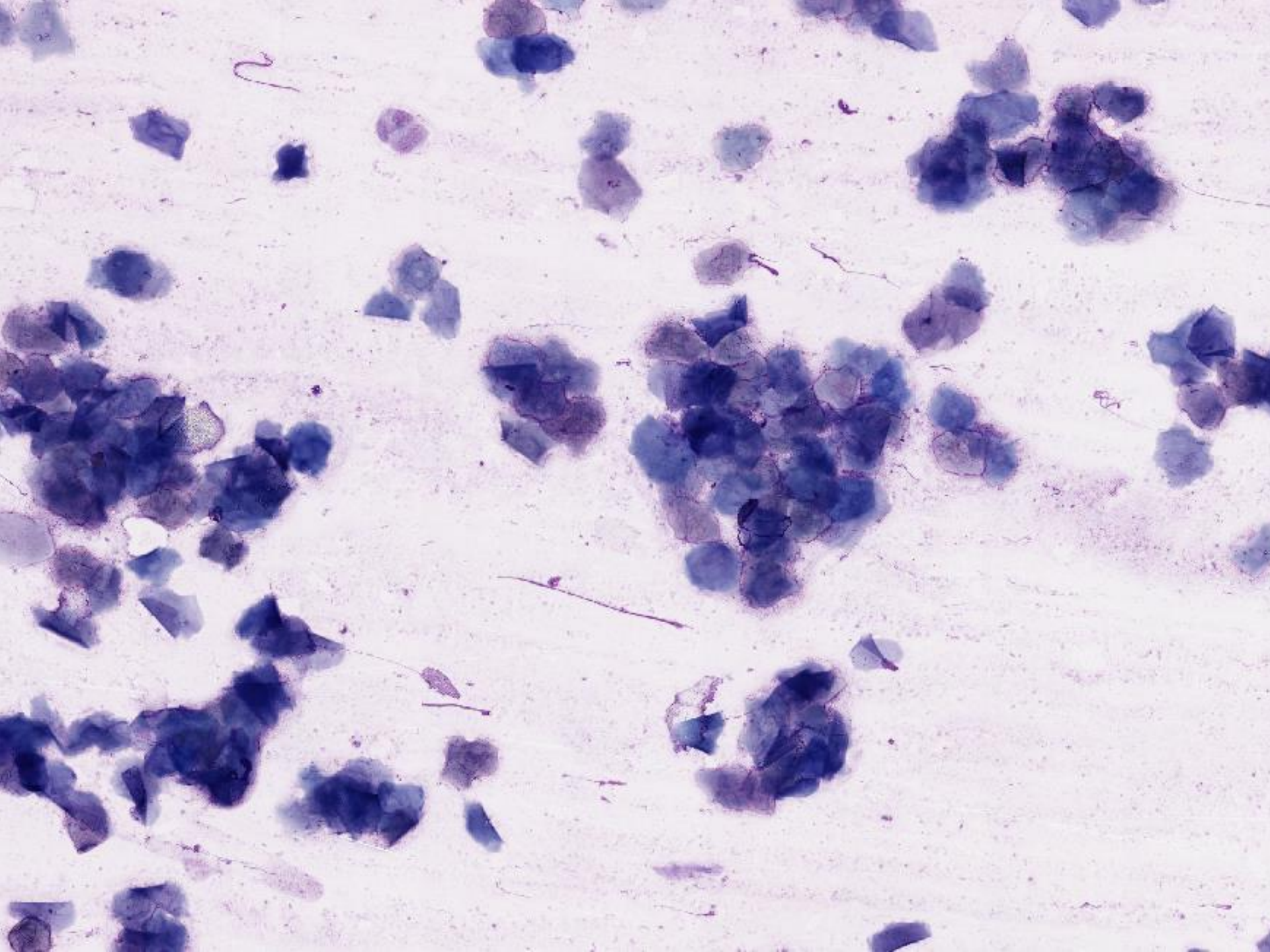


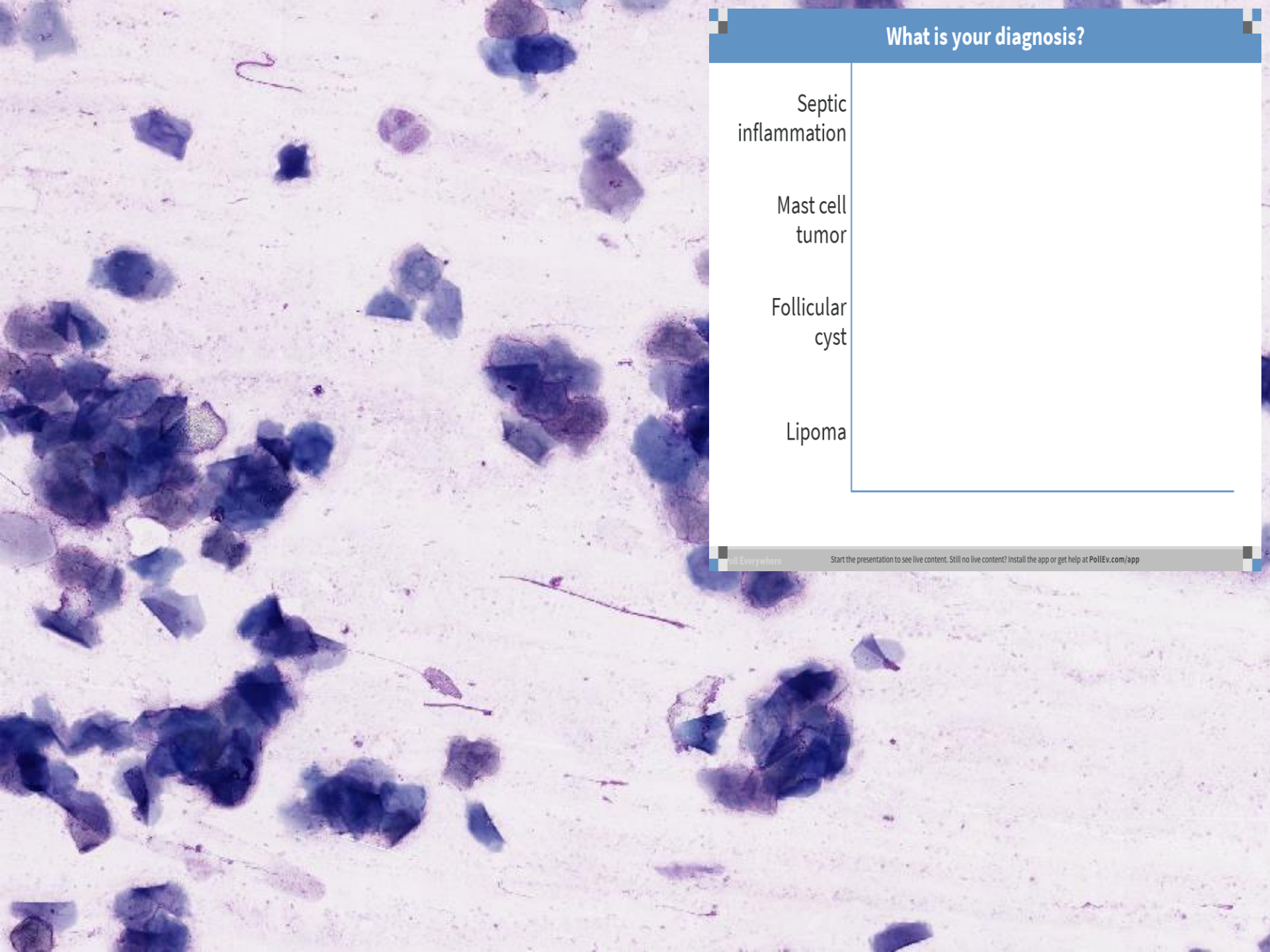
- Lipoma
- Large bubble-like cells
- Smear appears greasy
- May be poorly cellular

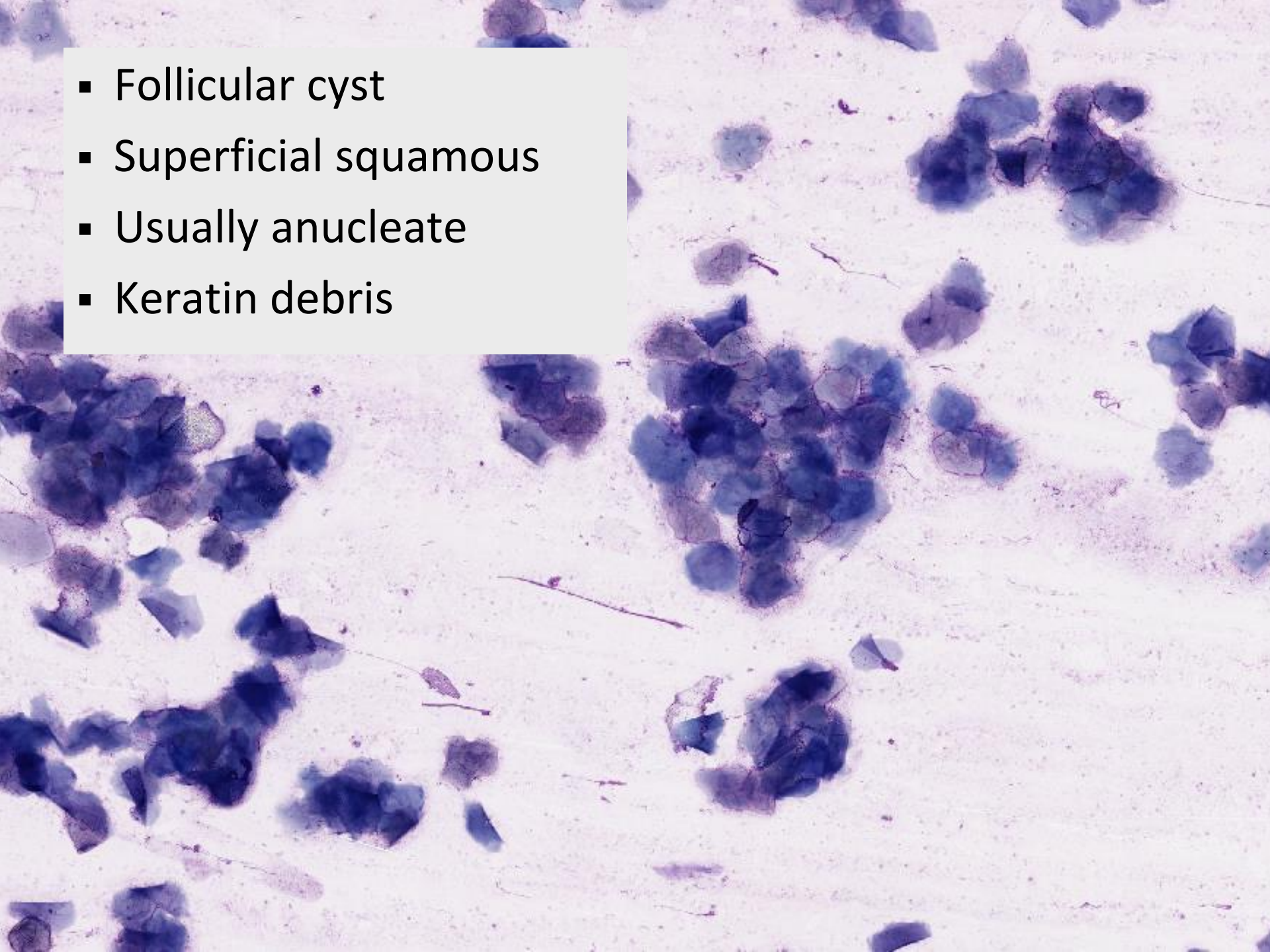


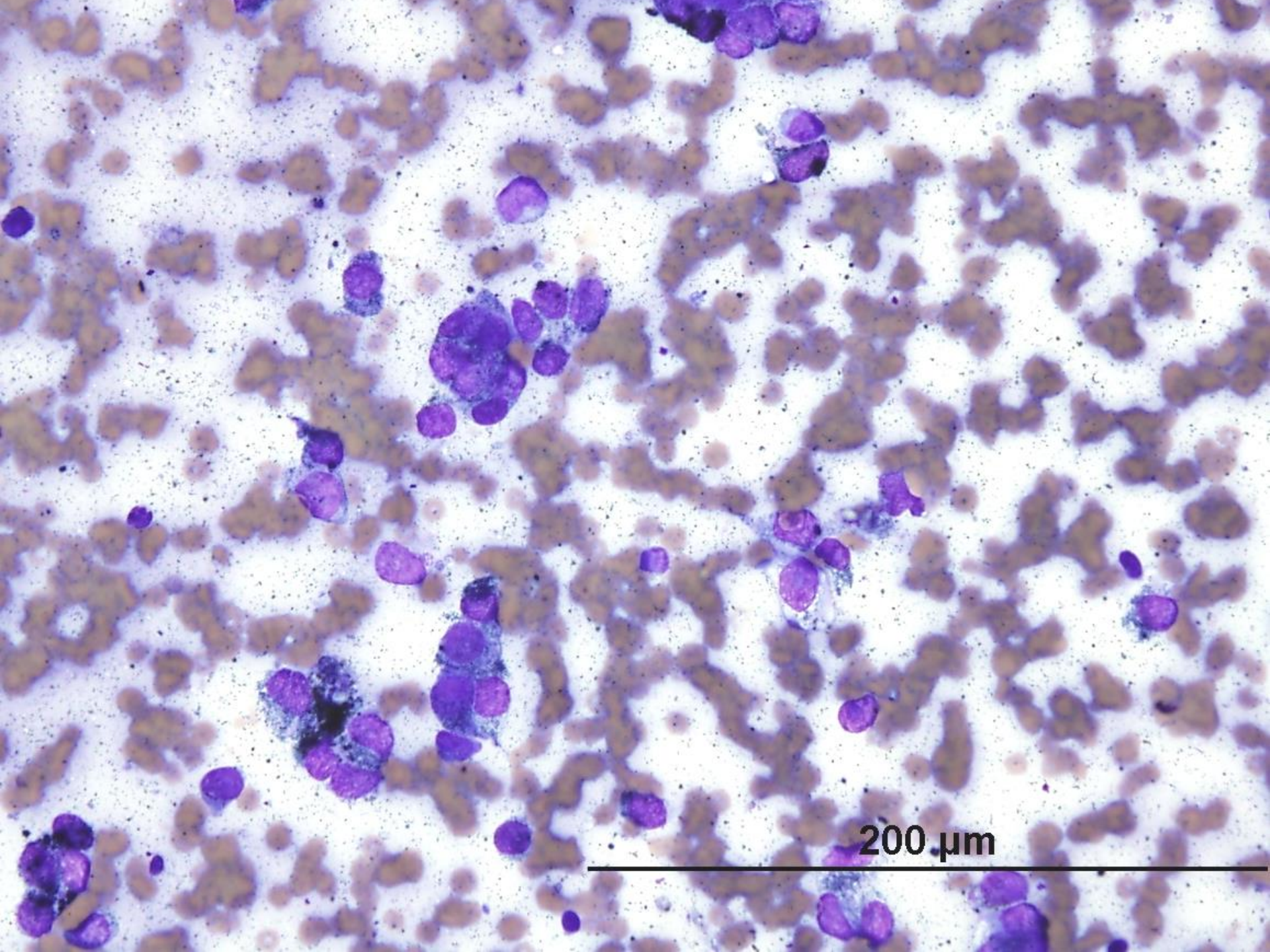

What is your diagnosis?
Lipoma
Sebaceous adenoma
Mast cell tumor
Follicular cyst
Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app

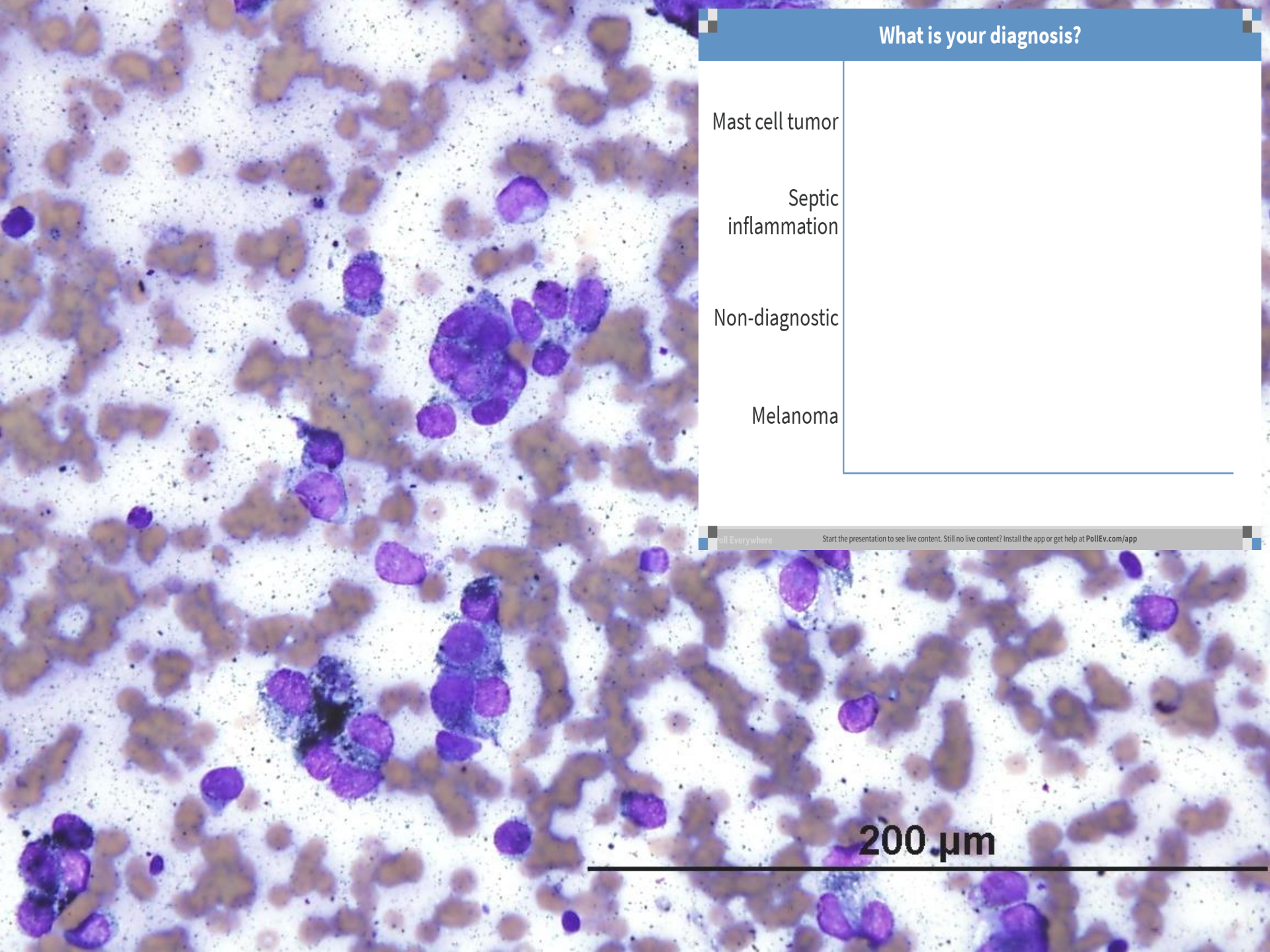

Sebaceous adenoma
Clusters of uniform cells
Vacuolated cytoplasm

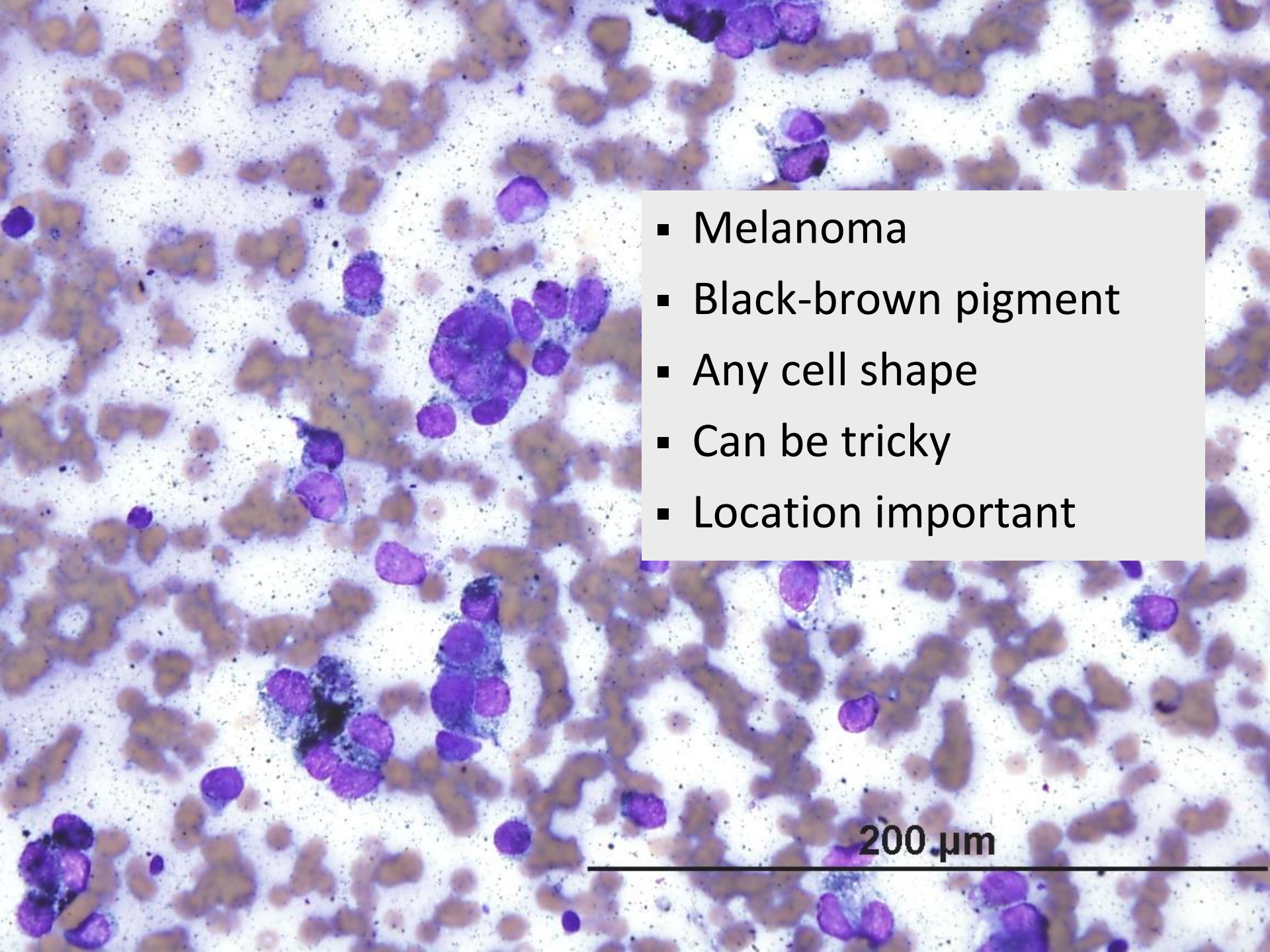



What is your diagnosis?
Septic inflammation
Mast cell tumor
Follicular cyst
Lipoma
Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app

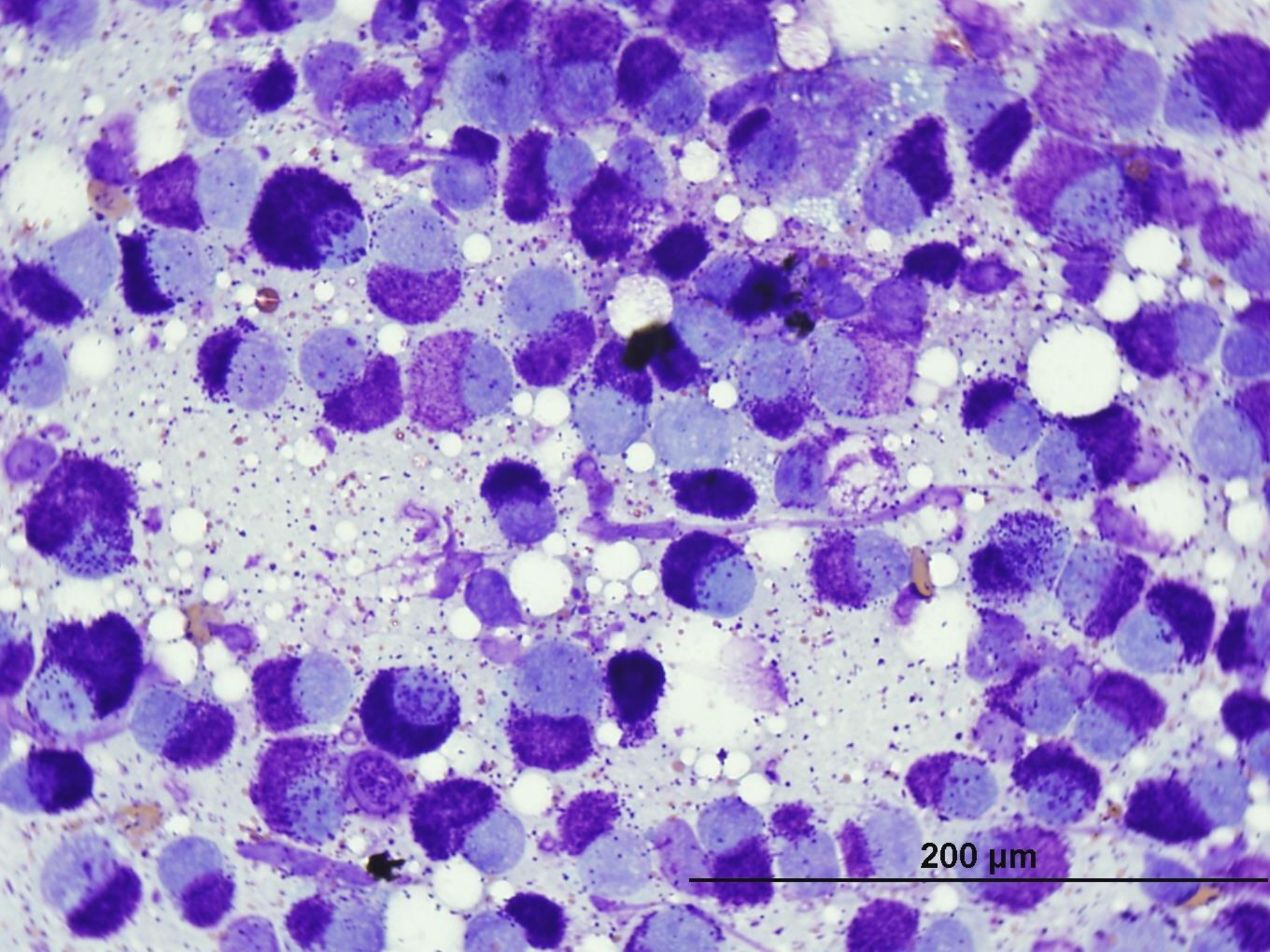

Follicular cyst
Superficial squamous
Usually anucleate
Keratin debris

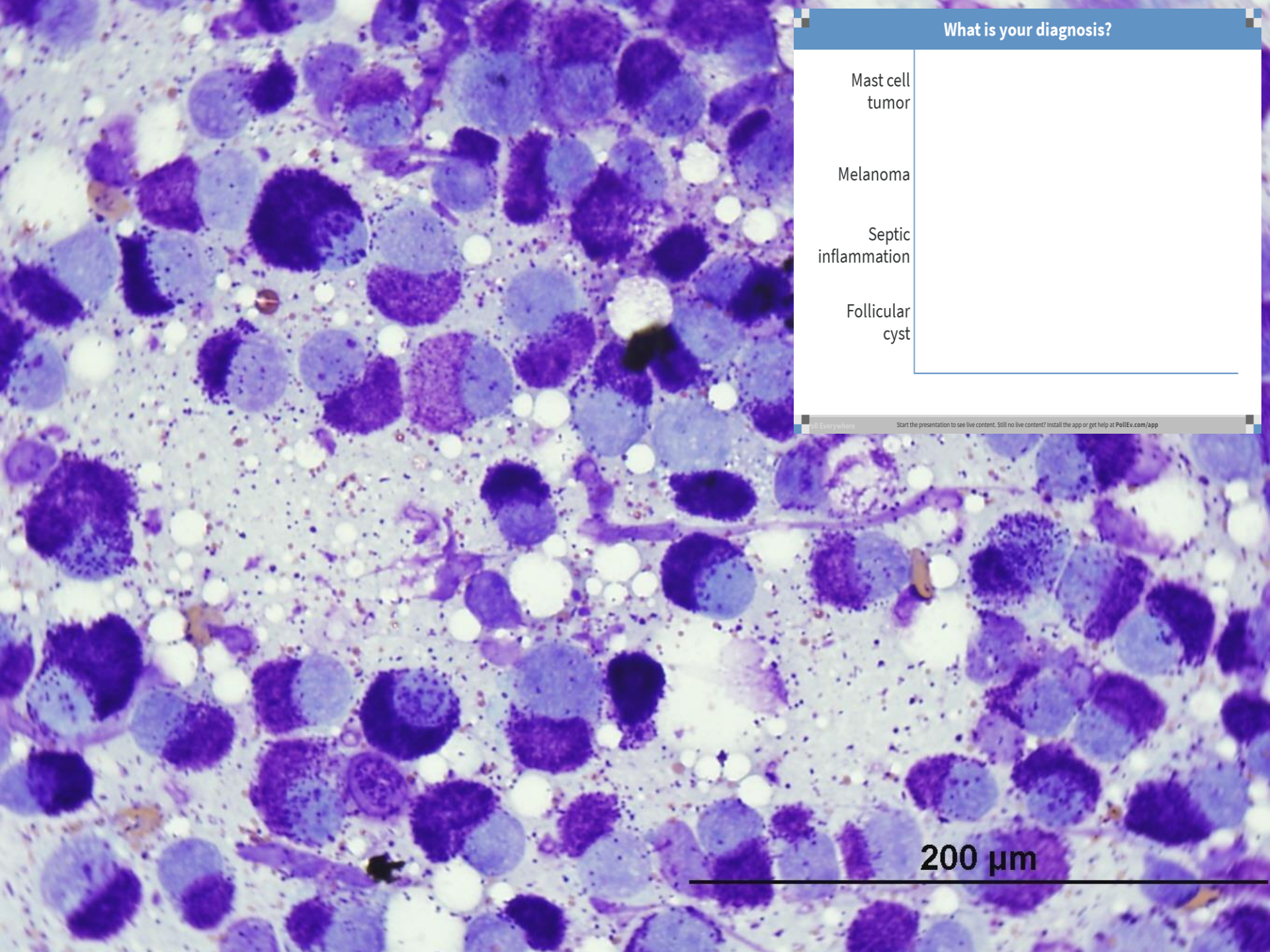

200 µm

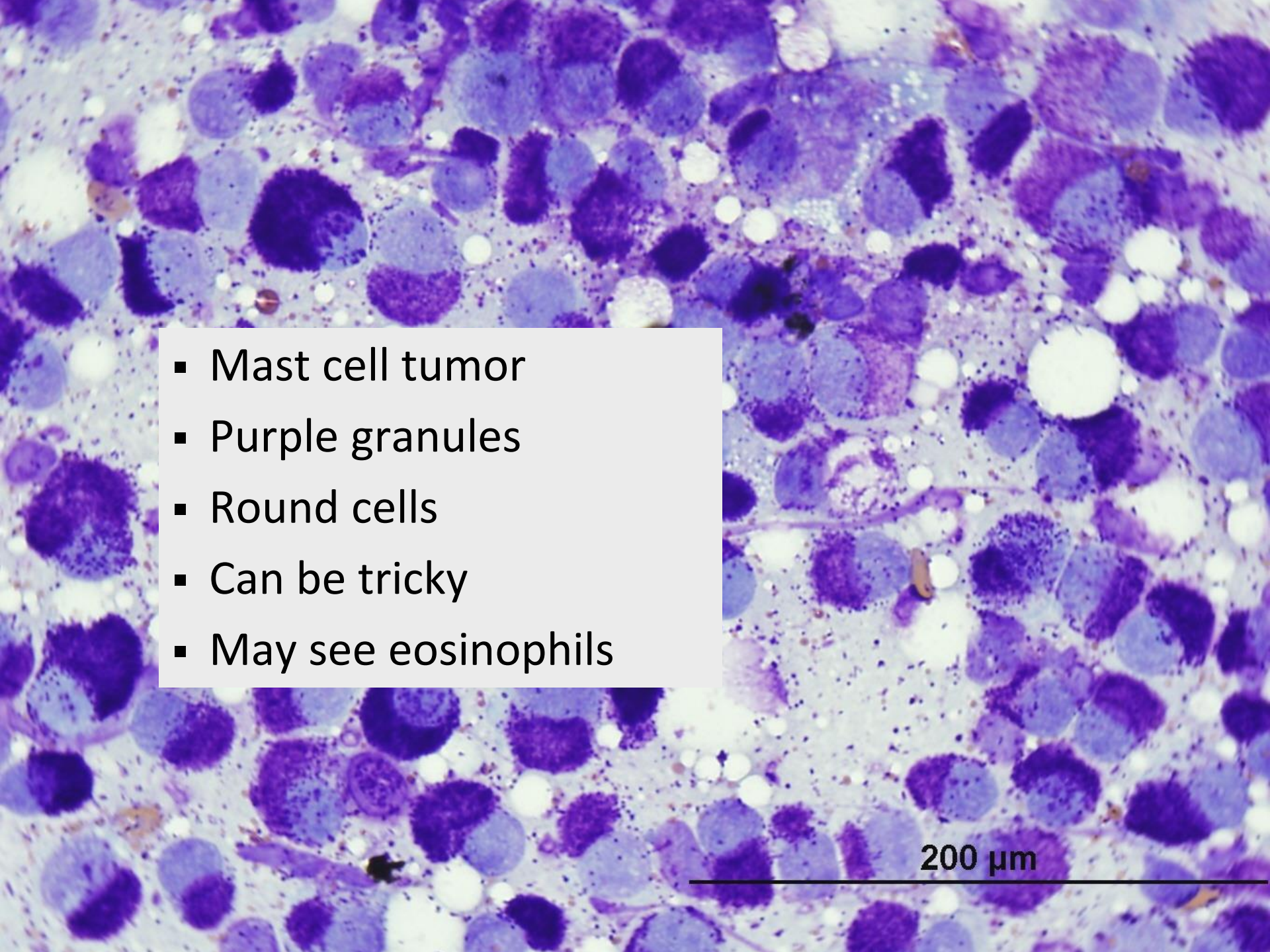

What is your diagnosis?
Mast cell tumor
Septic inflammation
Non-diagnostic
Melanoma
Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app
200 µm



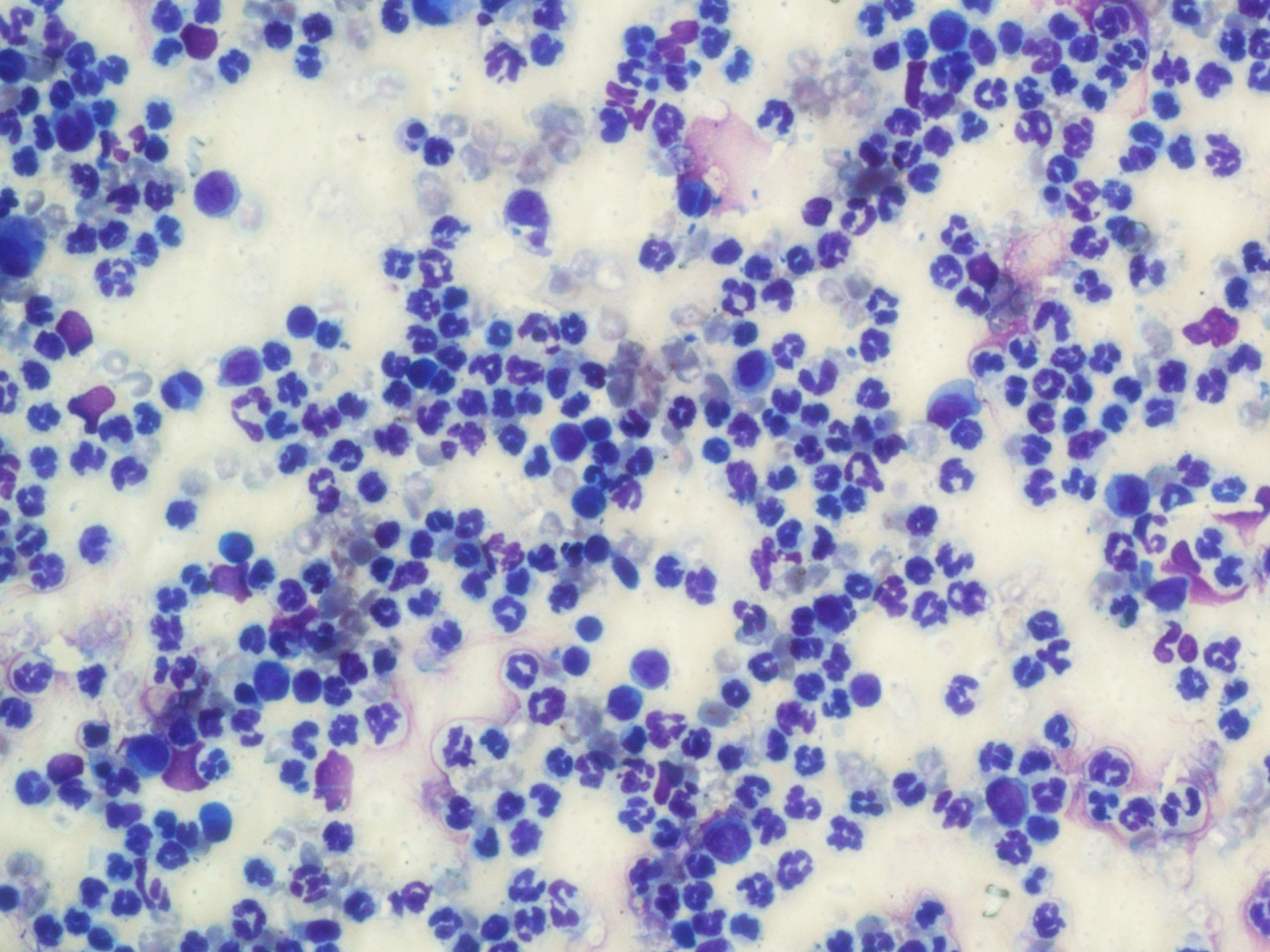
- Melanoma
- Black-brown pigment
- Any cell shape
- Can be tricky
- Location important

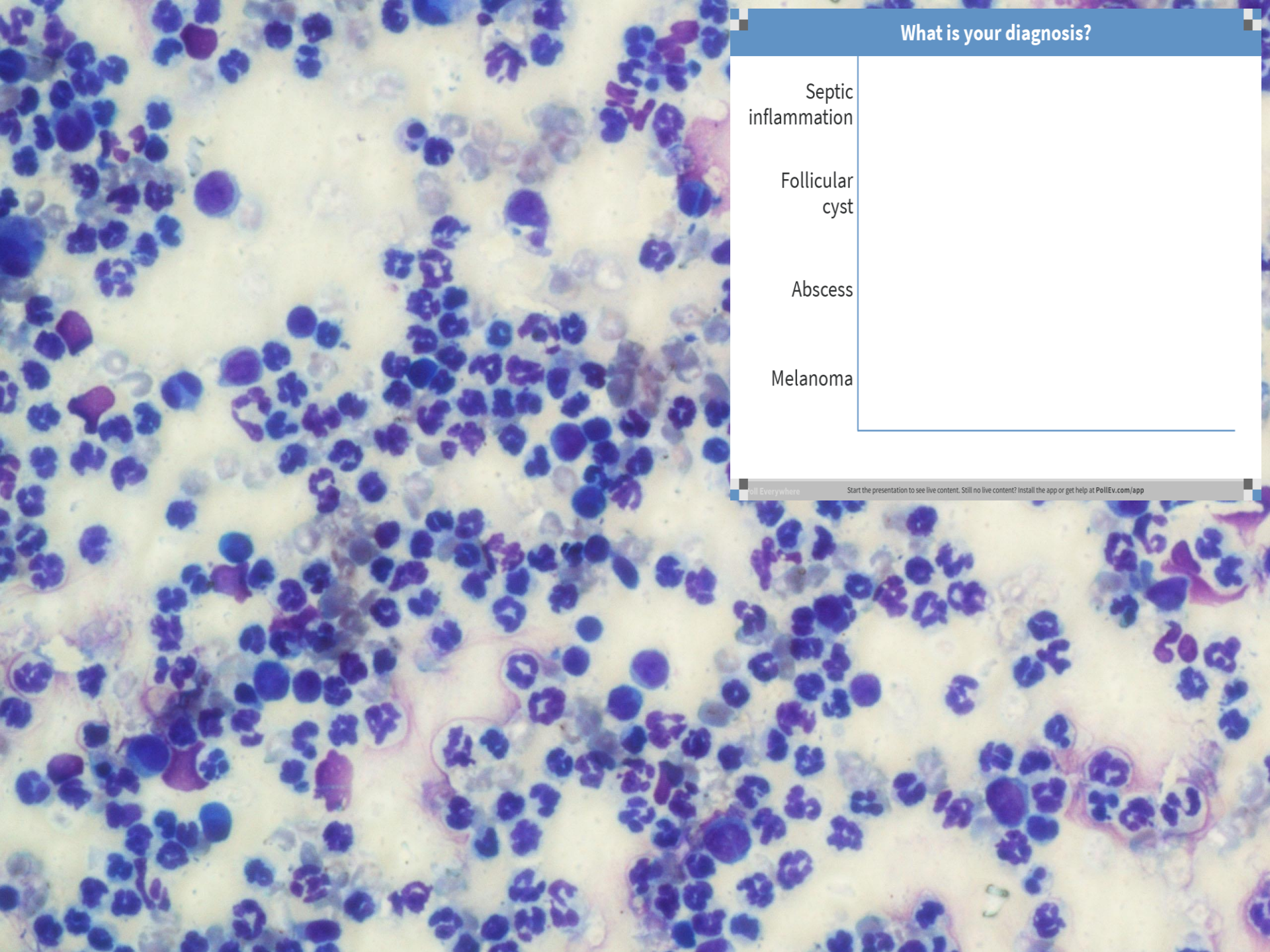

200 µm

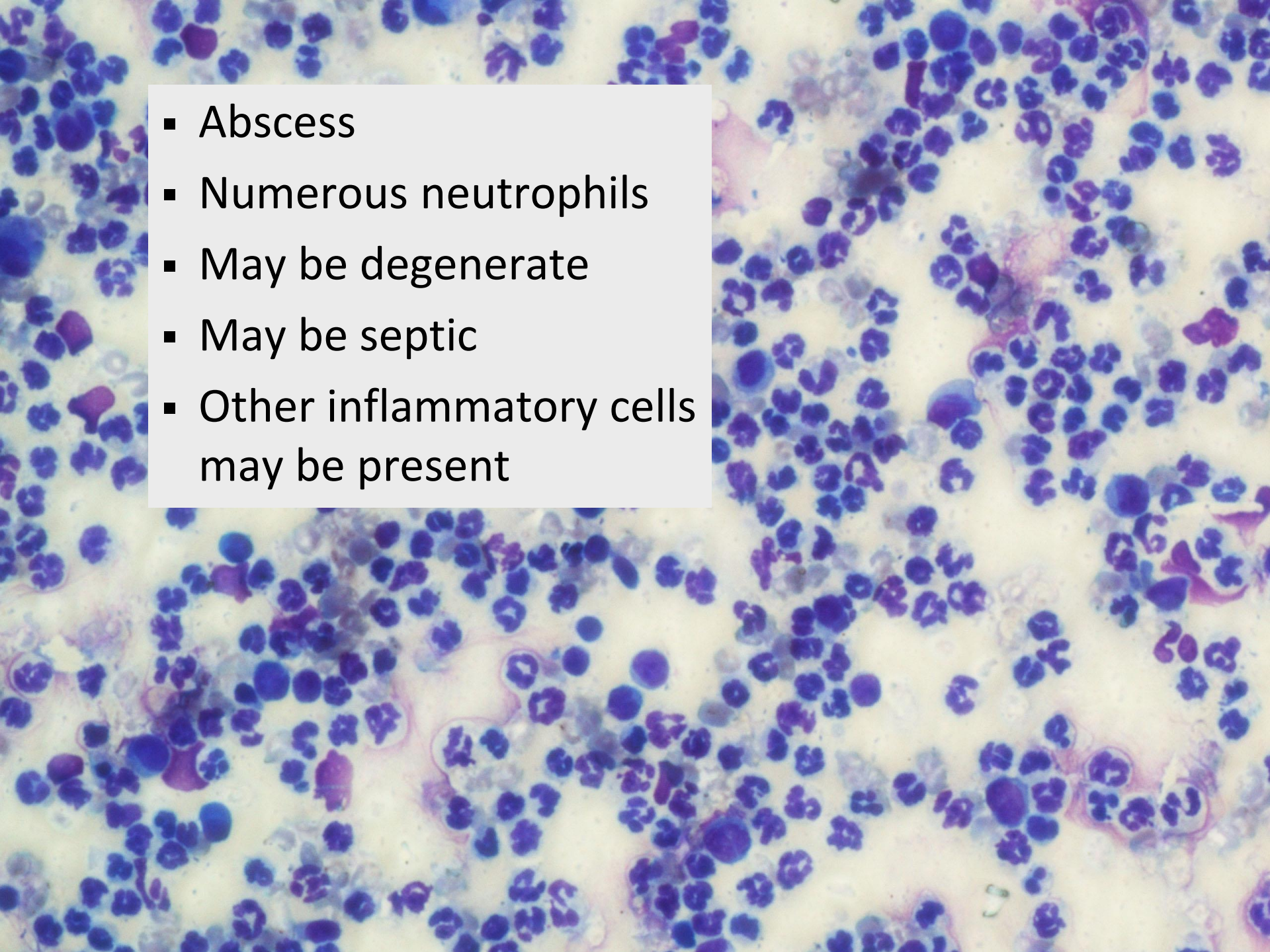

What is your diagnosis?
Mast cell tumor
Melanoma
Septic inflammation
Follicular cyst
Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app
200 µm



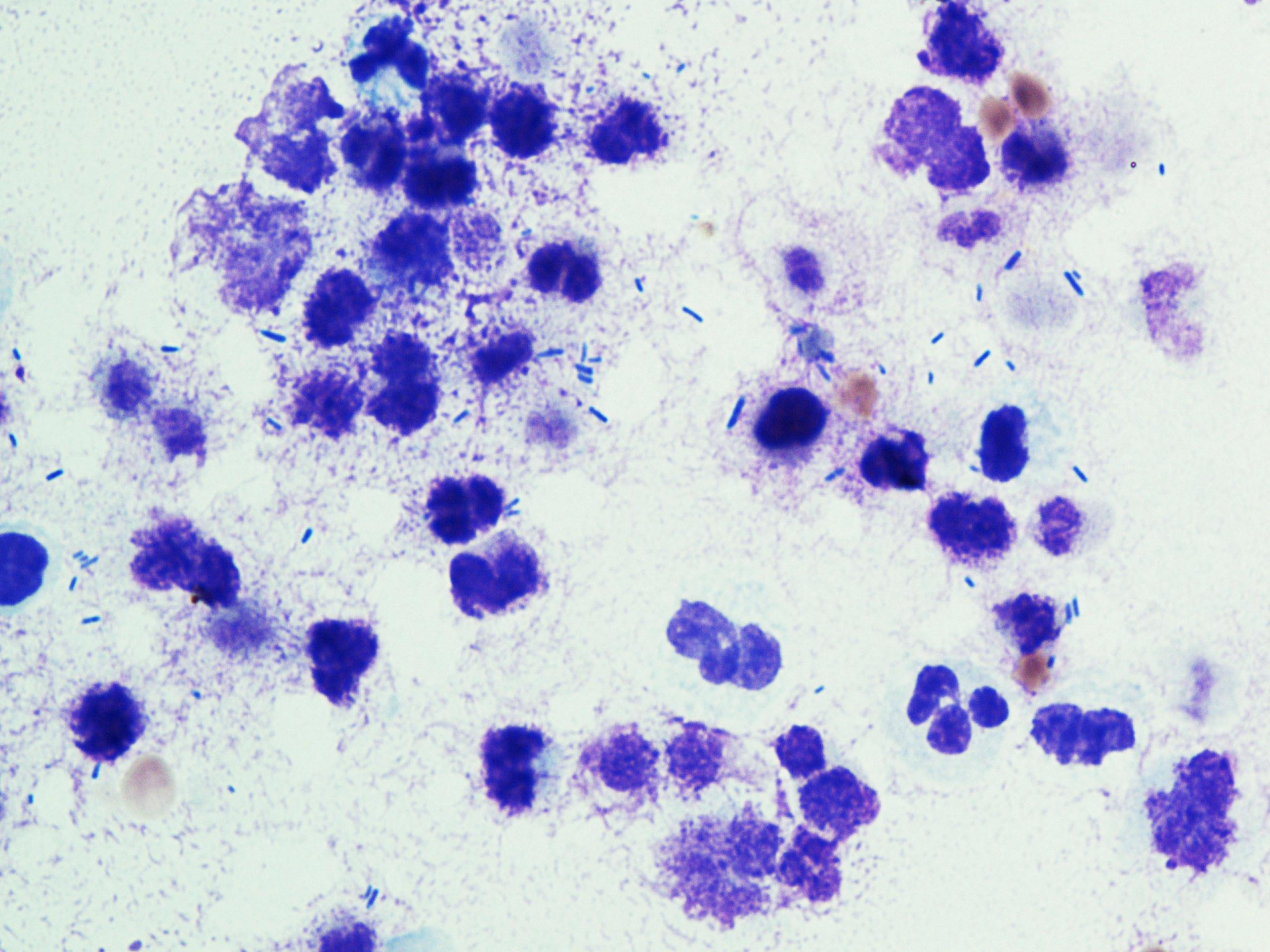
- Mast cell tumor
- Purple granules
- Round cells
- Can be tricky
- May see eosinophils

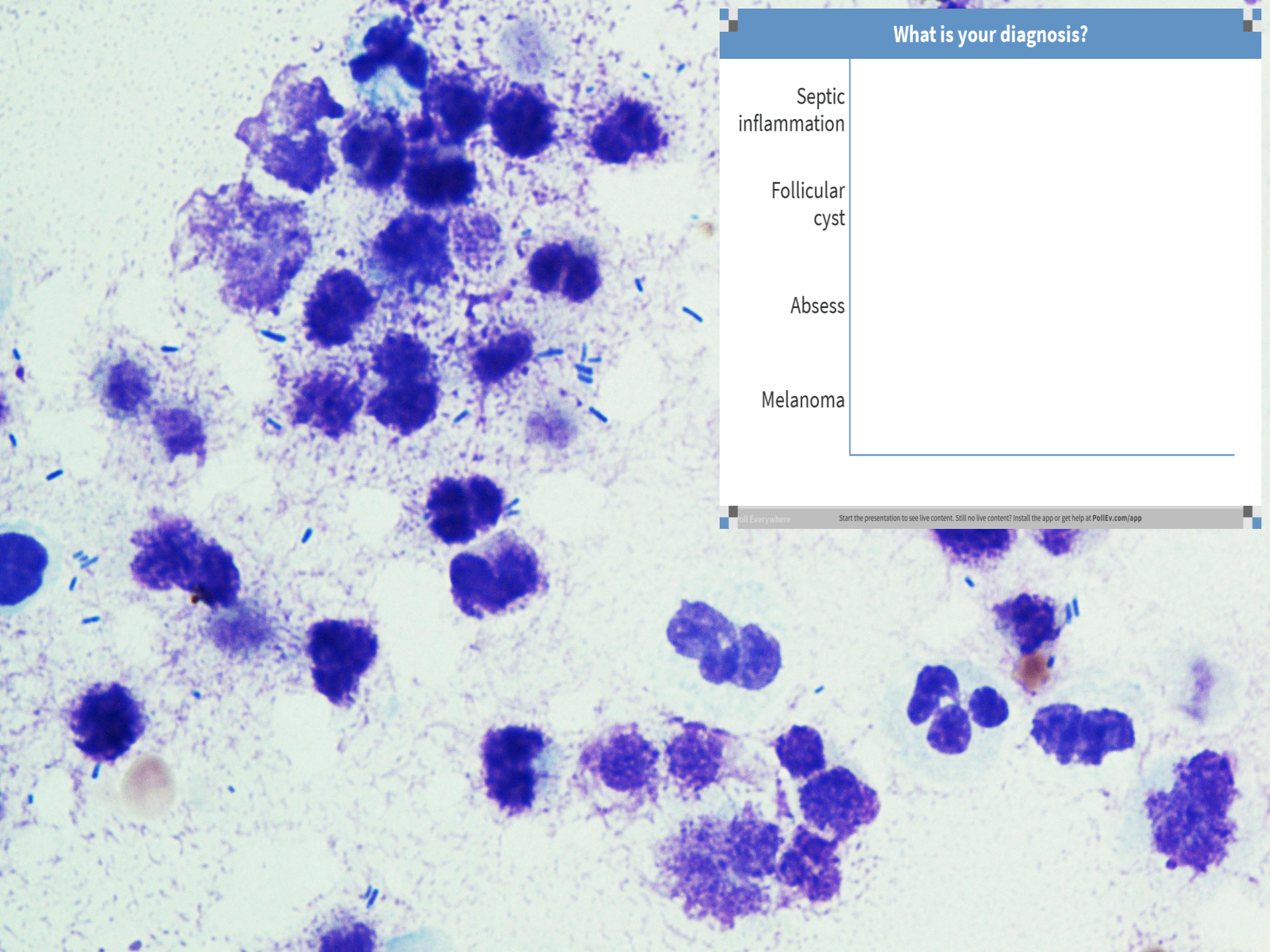


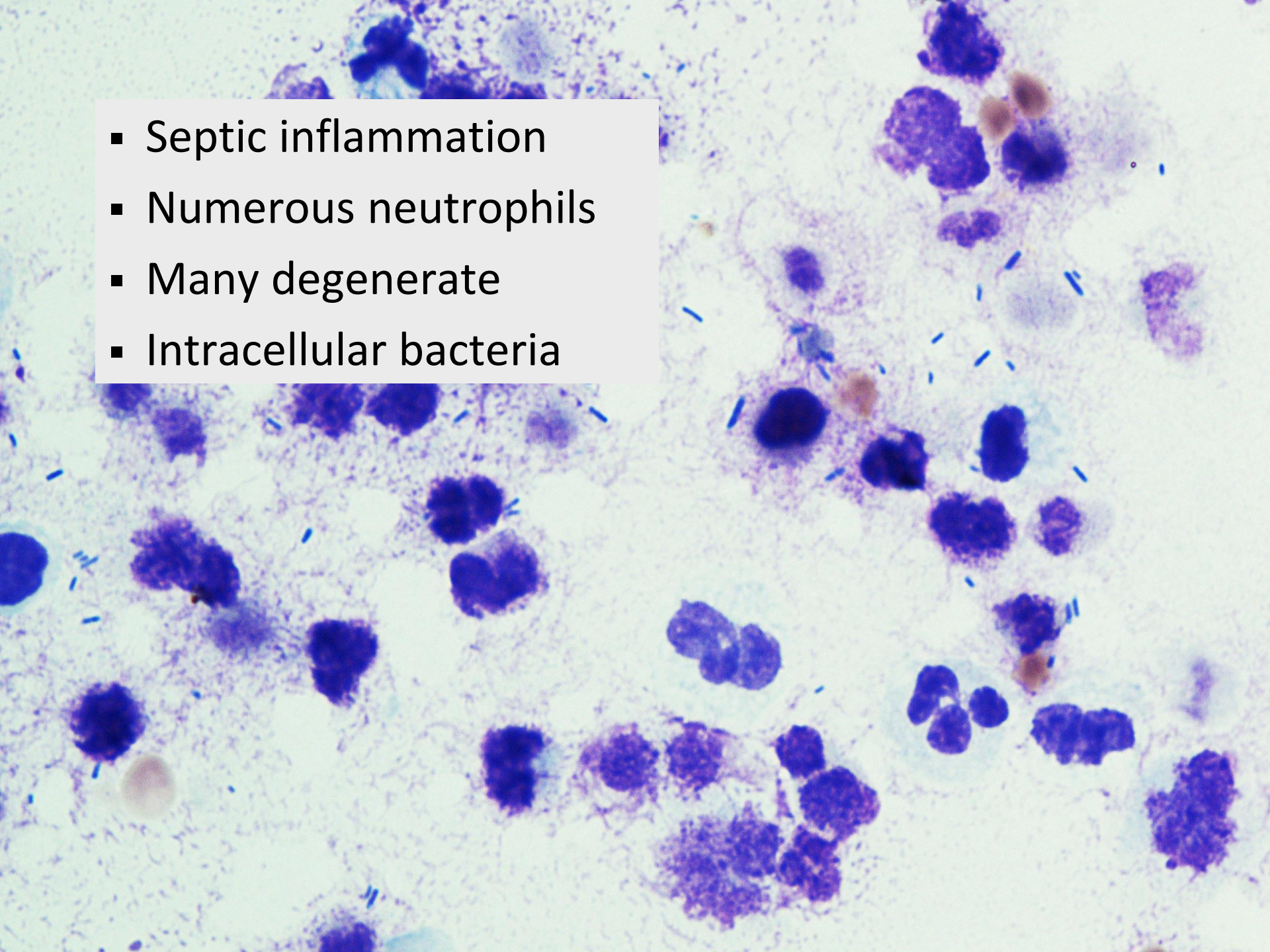

What is your diagnosis?
Septic inflammation
Follicular cyst
Abscess
Melanoma
Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app



- Abscess
- Numerous neutrophils
- May be degenerate
- May be septic
- Other inflammatory cells may be present




What is your diagnosis?
Septic inflammation
Follicular cyst
Absess
Melanoma
Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app


Septic inflammation
Numerous neutrophils
Many degenerate
Intracellular bacteria

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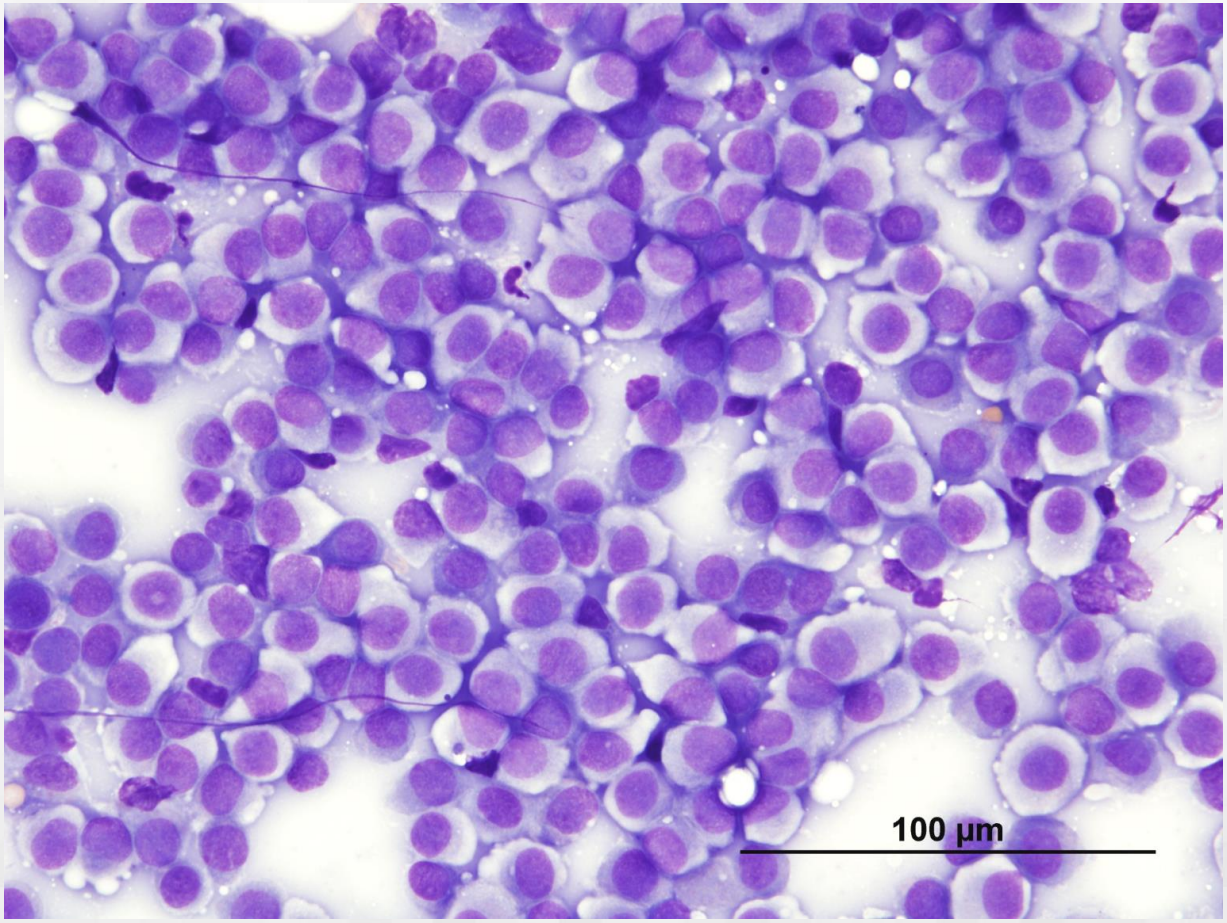
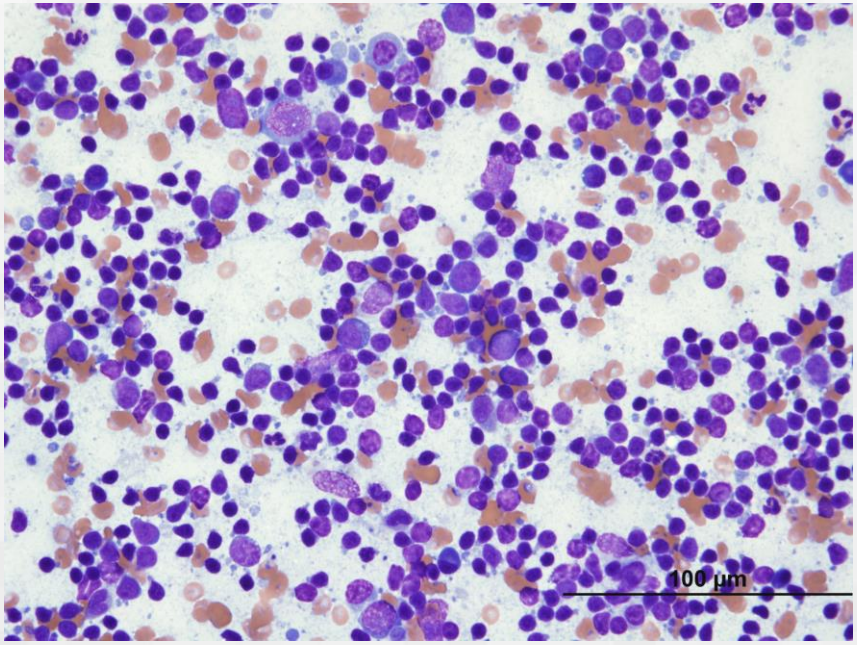
Special Considerations

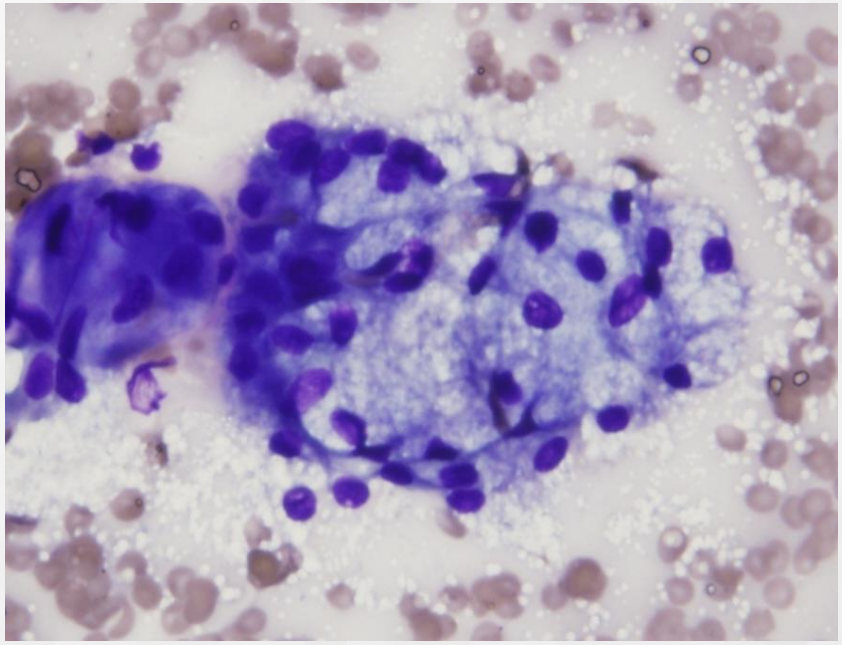


Sub-mandibular mass

Lymph node

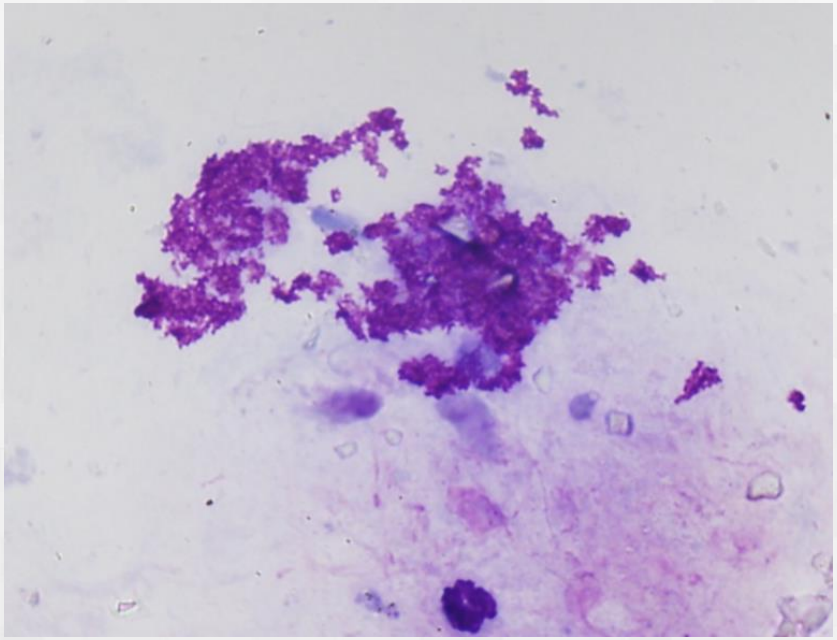


Salivary gland

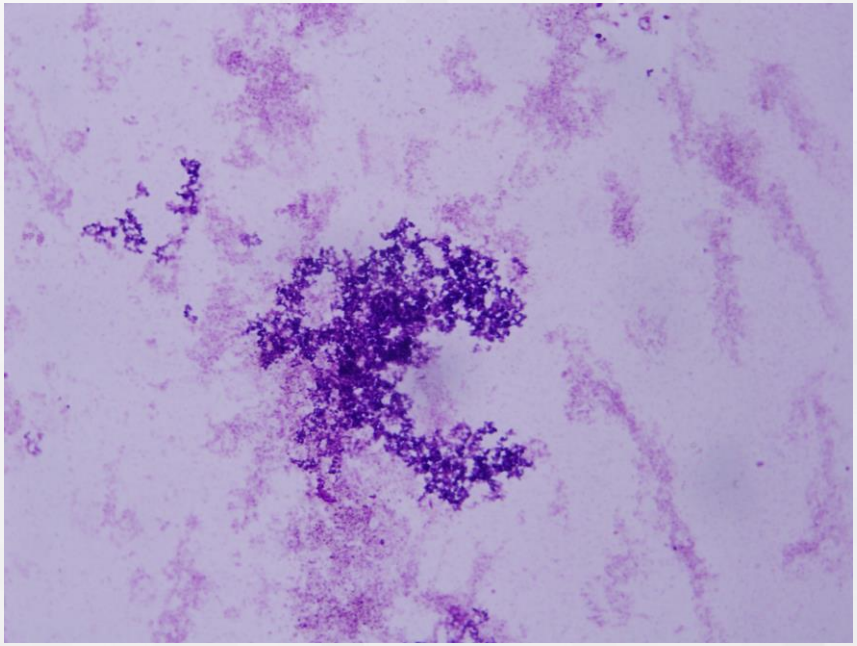


Artifacts

US gel



Stain precipitate



Questions?

Stay tuned for hematology…

Anna Lee Larimore

Part 2: Hematology

Red cells

White cells

Platelets

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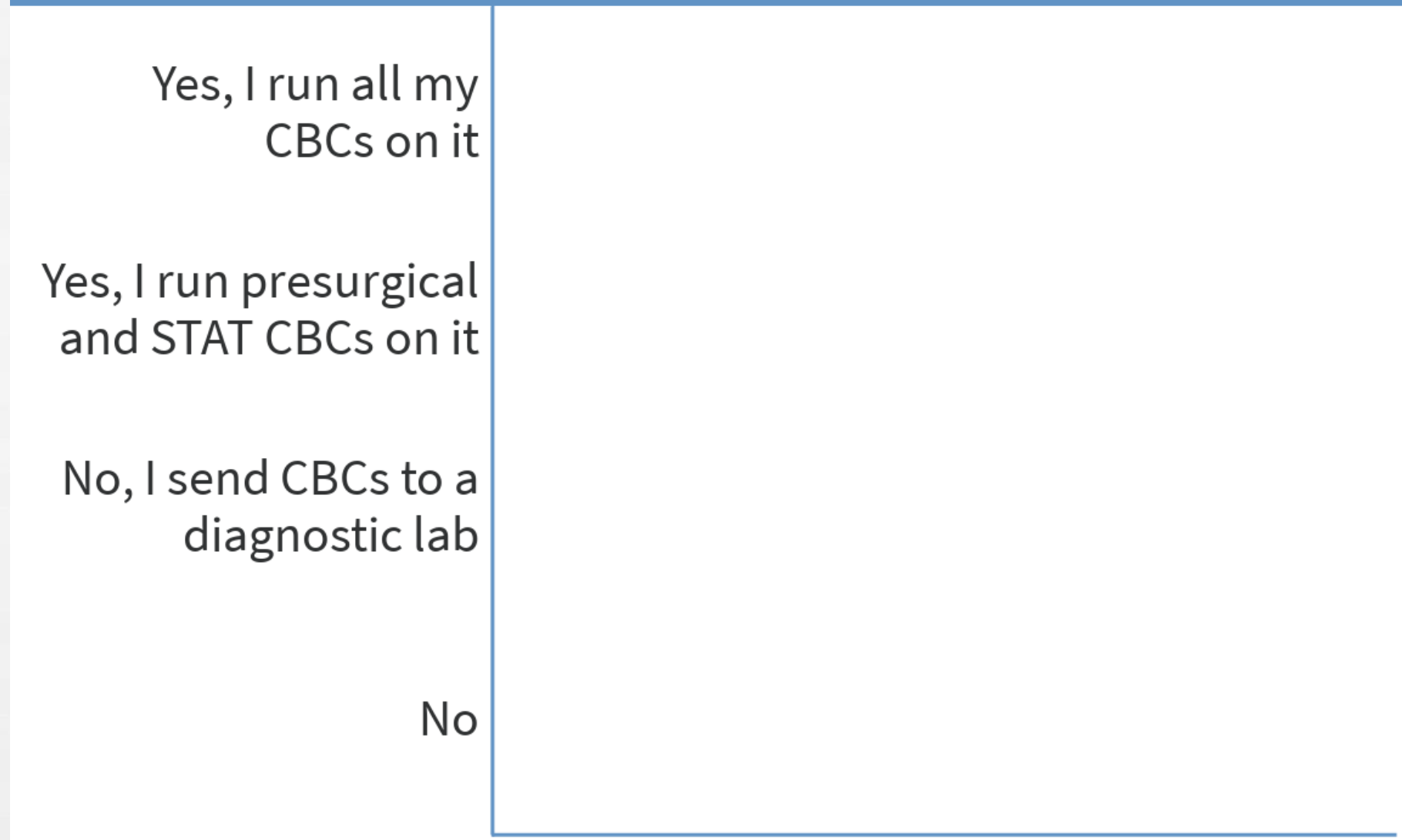
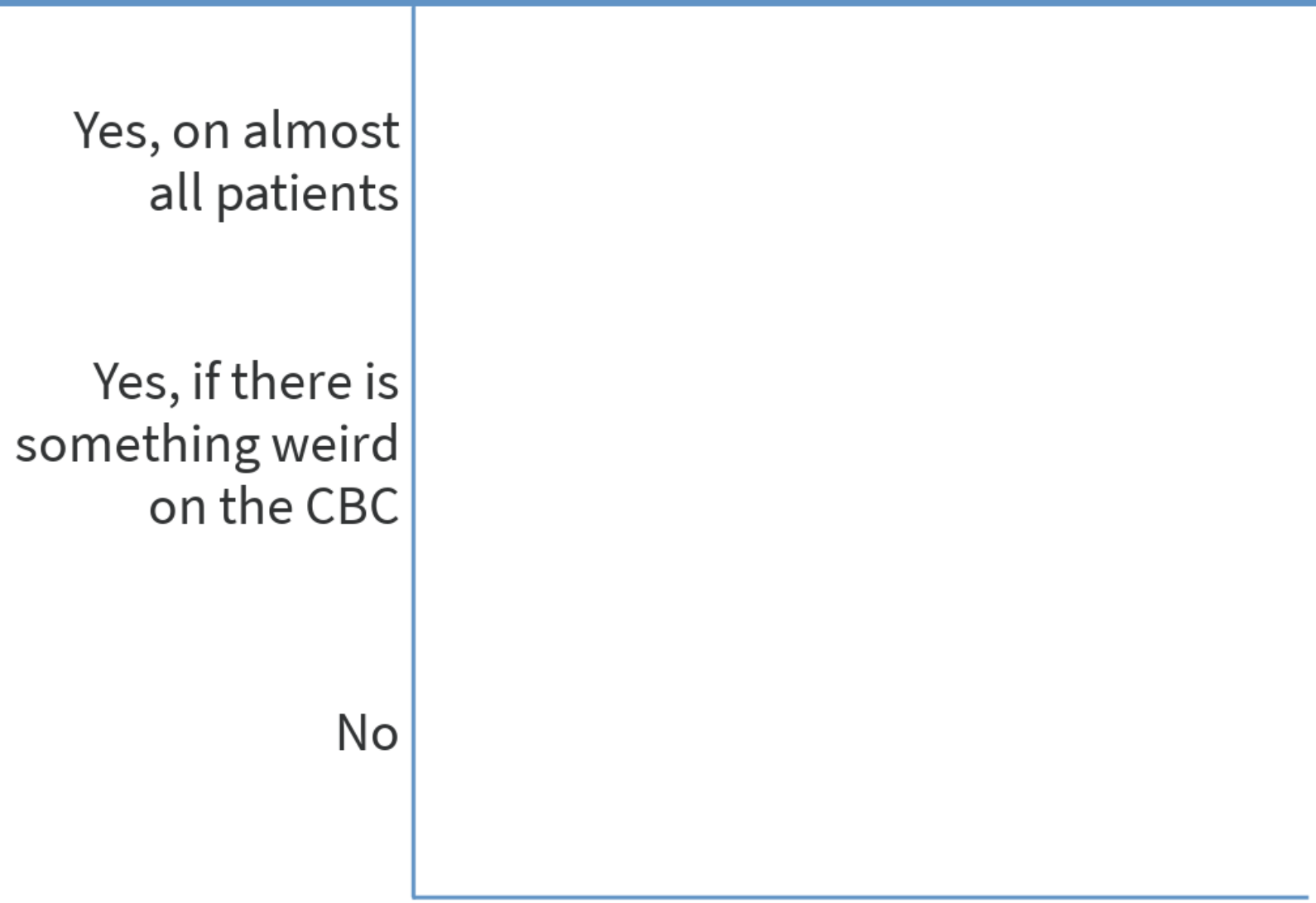
Part 2: Hematology

- Ask the audience:

Do you have an in-house hematology analyzer?



Do you routinely examine blood smears from in-house CBCs?



Hematology

- Your analyzer can't tell you everything
- RBC morphology
  - Regeneration
  - Causes of anemia
- WBC morphology
  - Left shift and toxic change
  - Leukemia
- Platelet clumping

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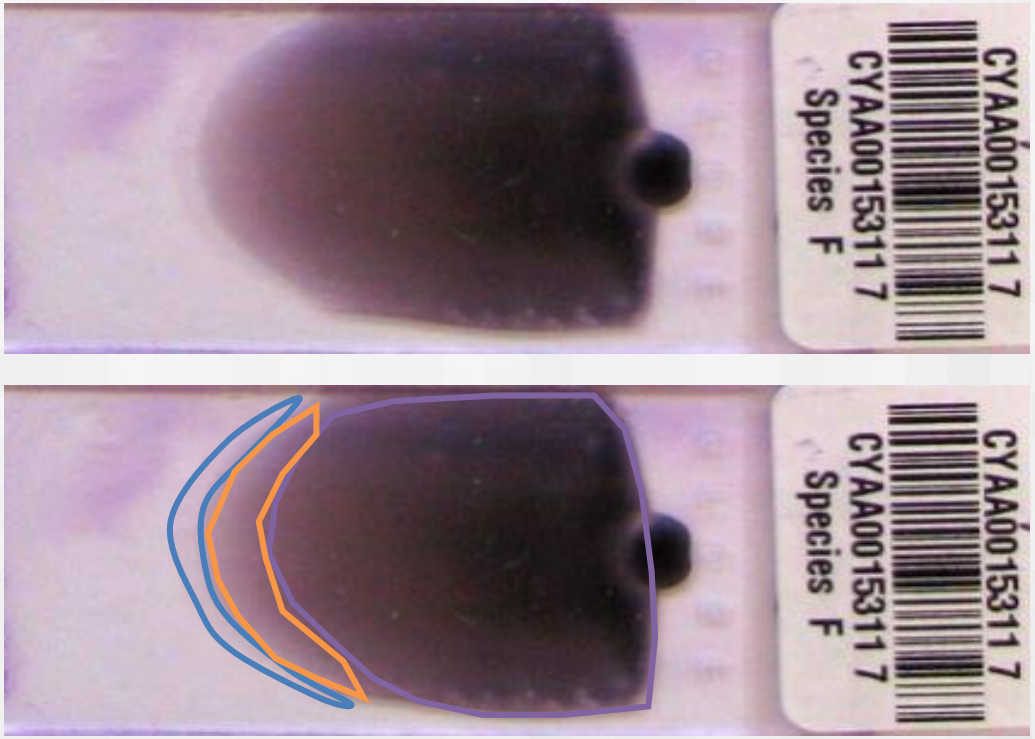
The Monolayer

Feathered Edge

Monolayer

Body



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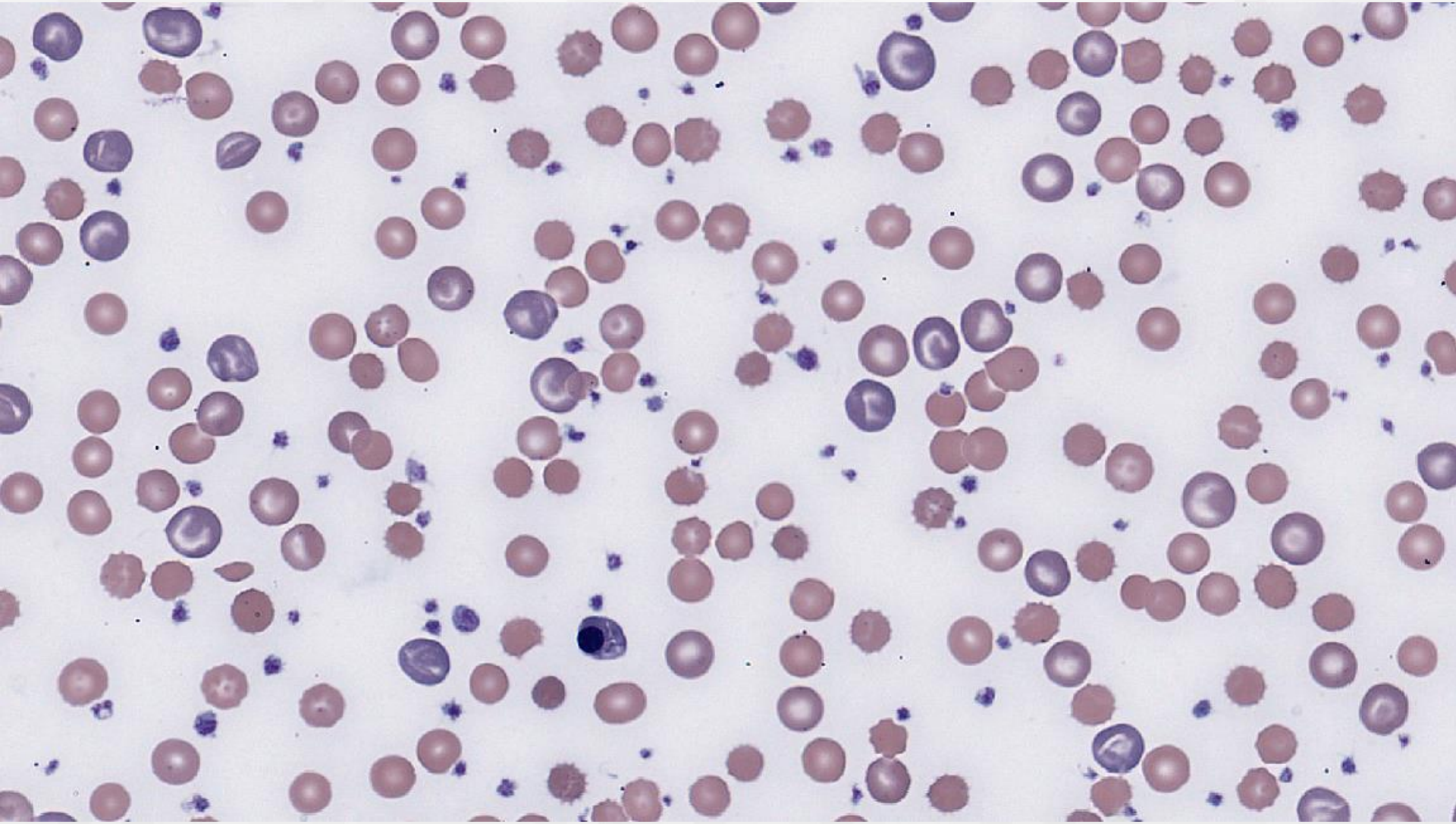
RBC Case 1



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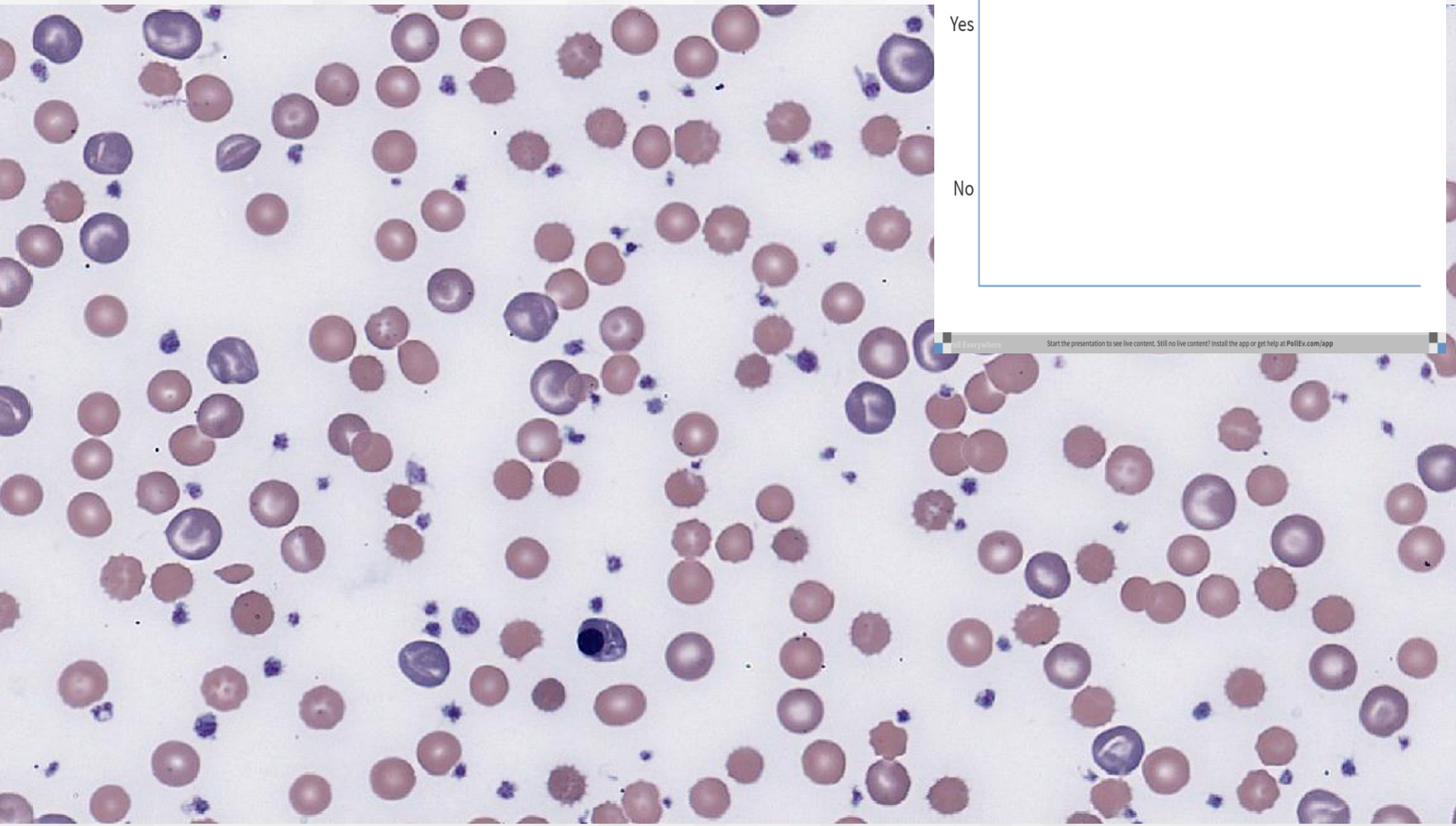

Is this anemia regenerative?

Yes

No

Start the presentation to see live content. Still no live content? Install the app or get help at PollEv.com/app

What is the evidence in support of regeneration?

Respond at **PollEv.com/nicolefernan414**

Text **NICOLEFERNAN414** to **37607** once to join, then **A, B, or C**

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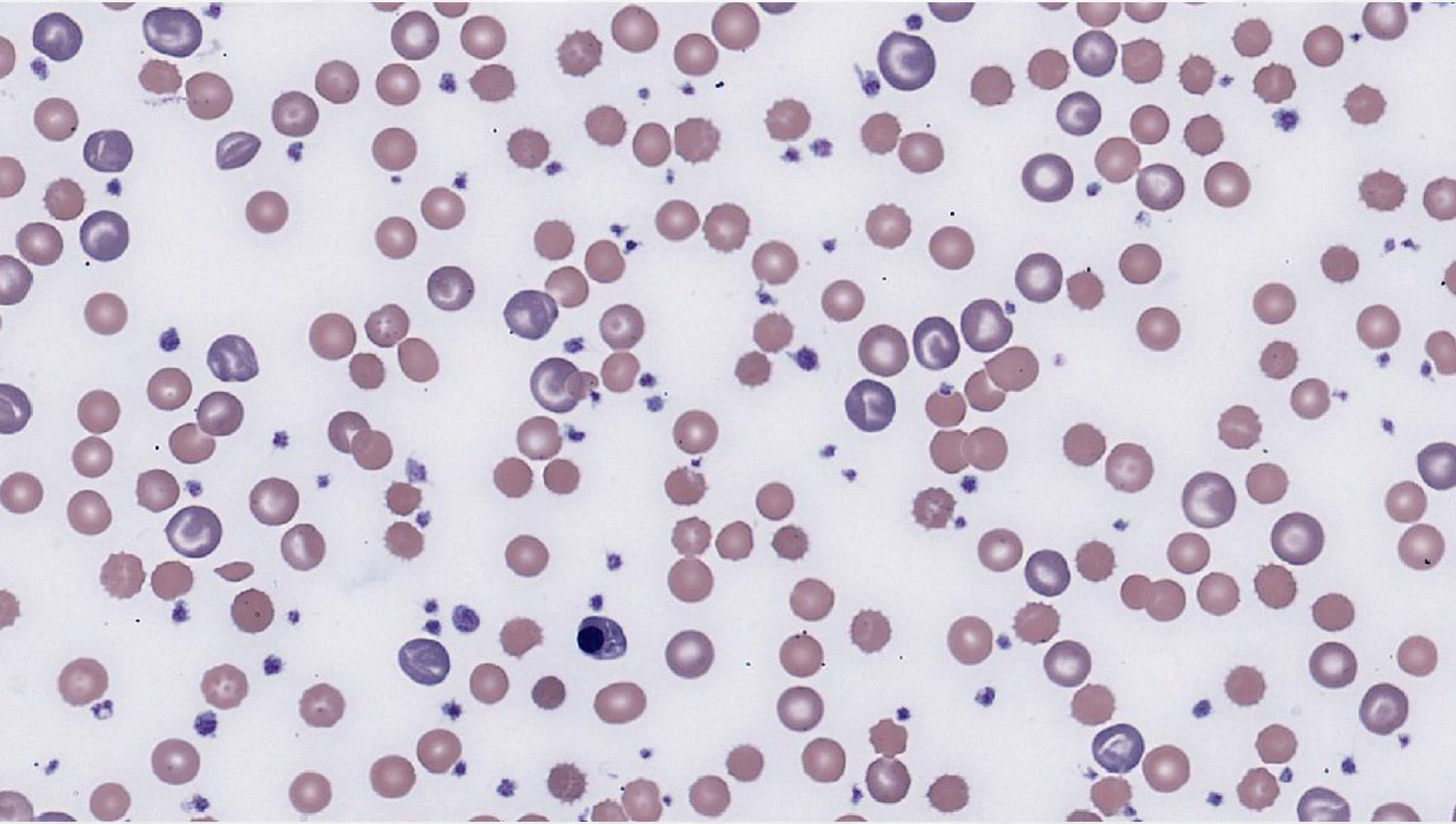
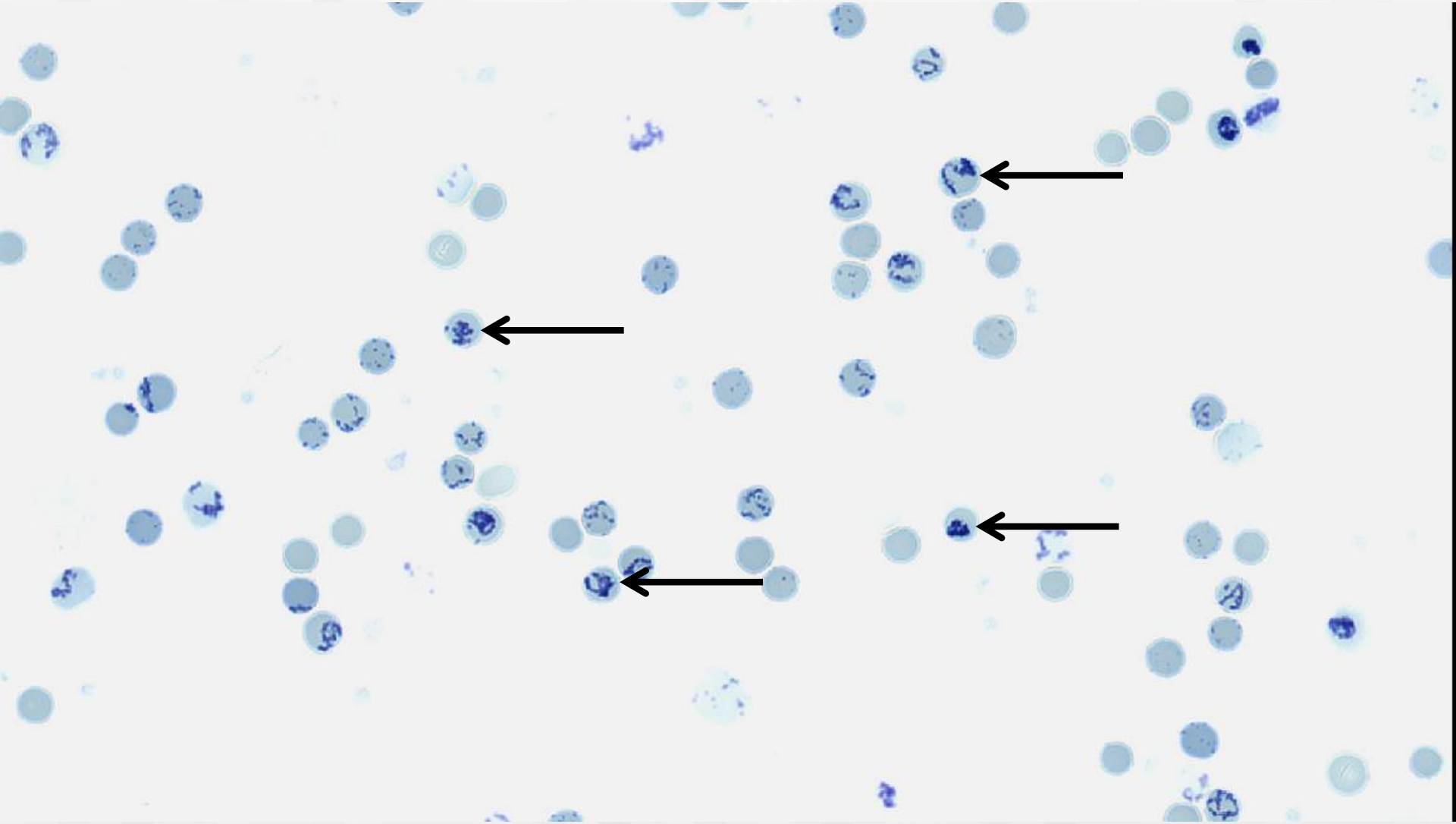
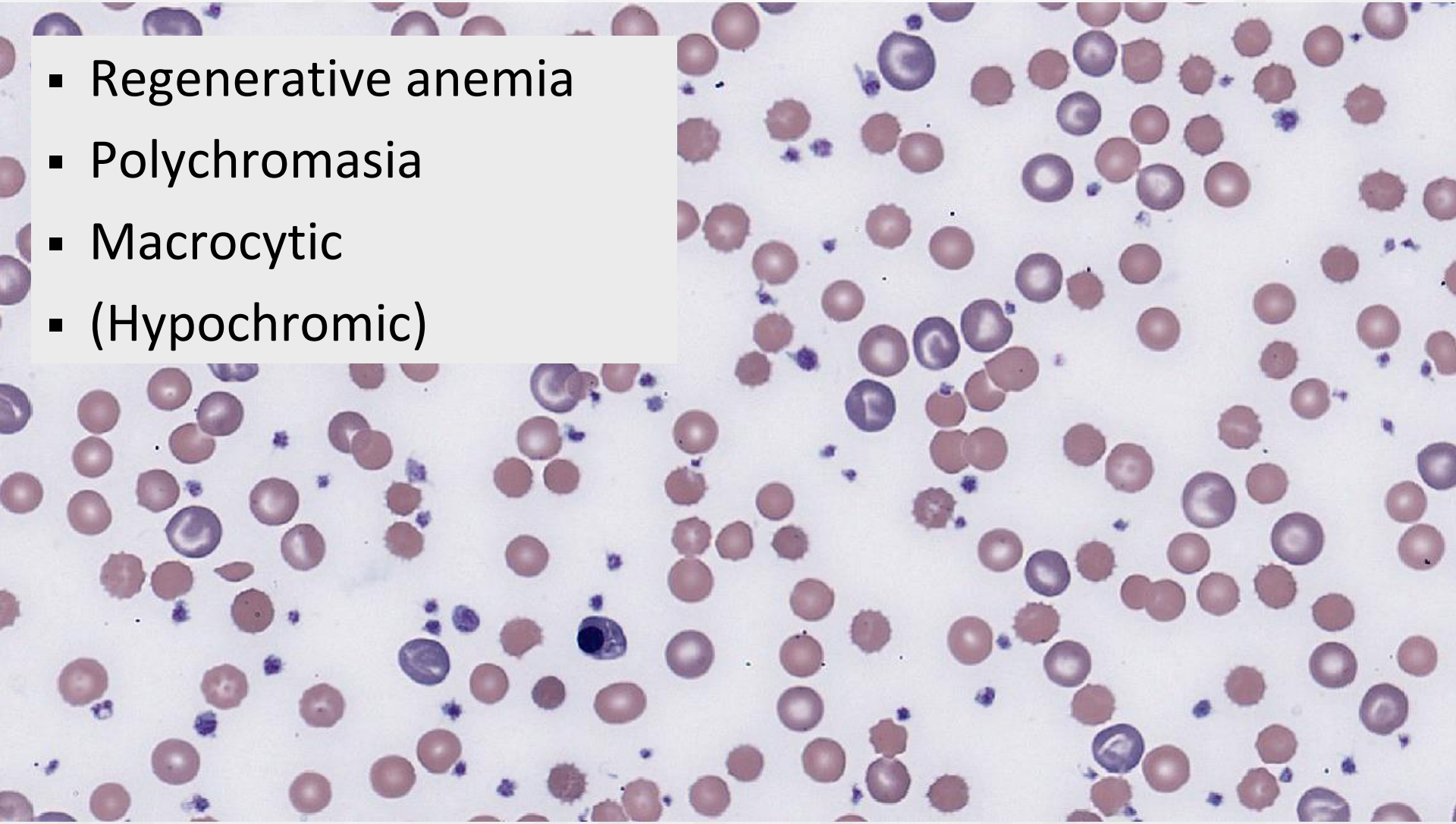
RBC Case 1





RBC Case 1



- Regenerative anemia
- Polychromasia
- Macrocytic
- (Hypochromic)

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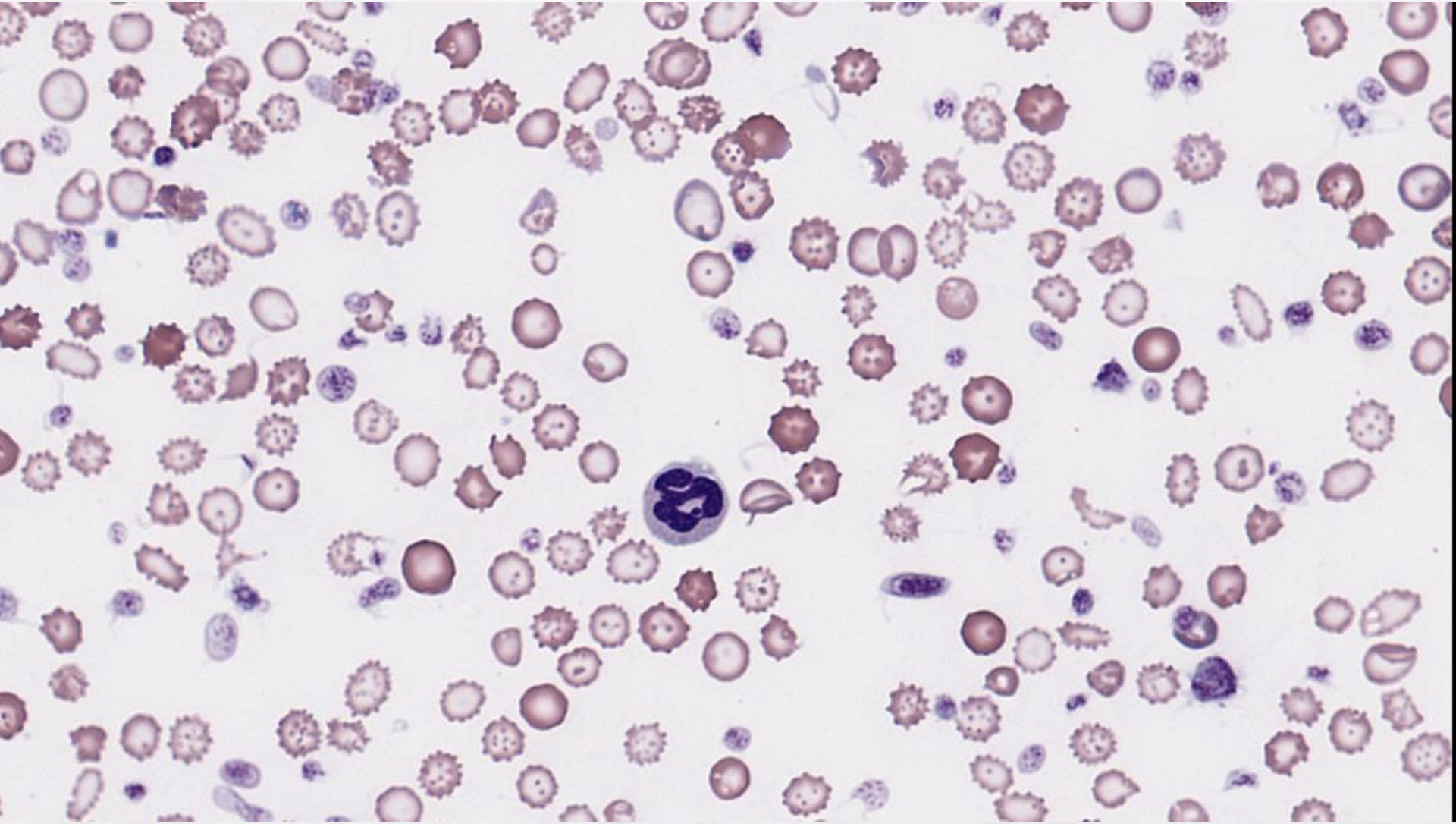
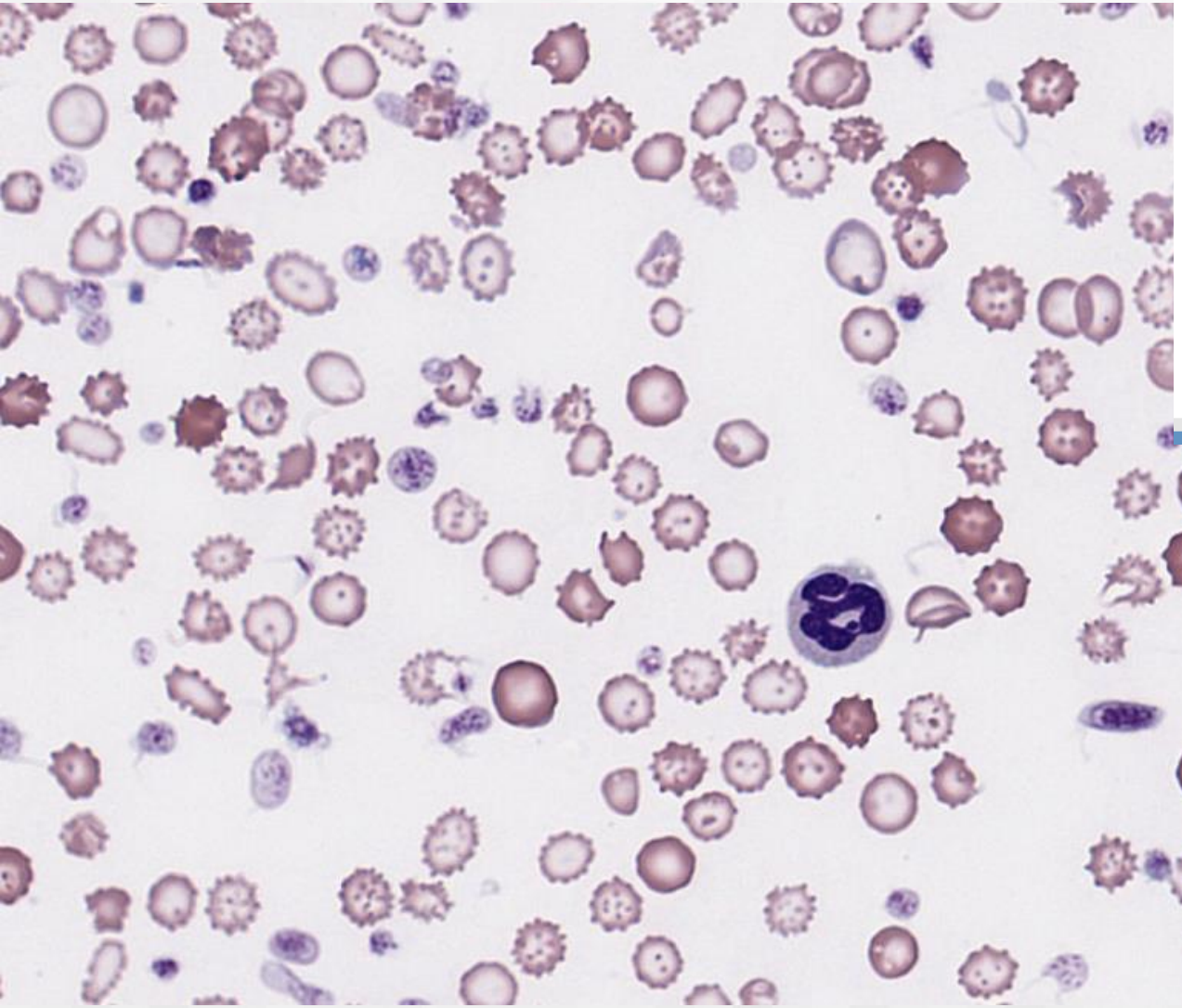
RBC Case 2





Is the anemia regenerative?

Yes

No

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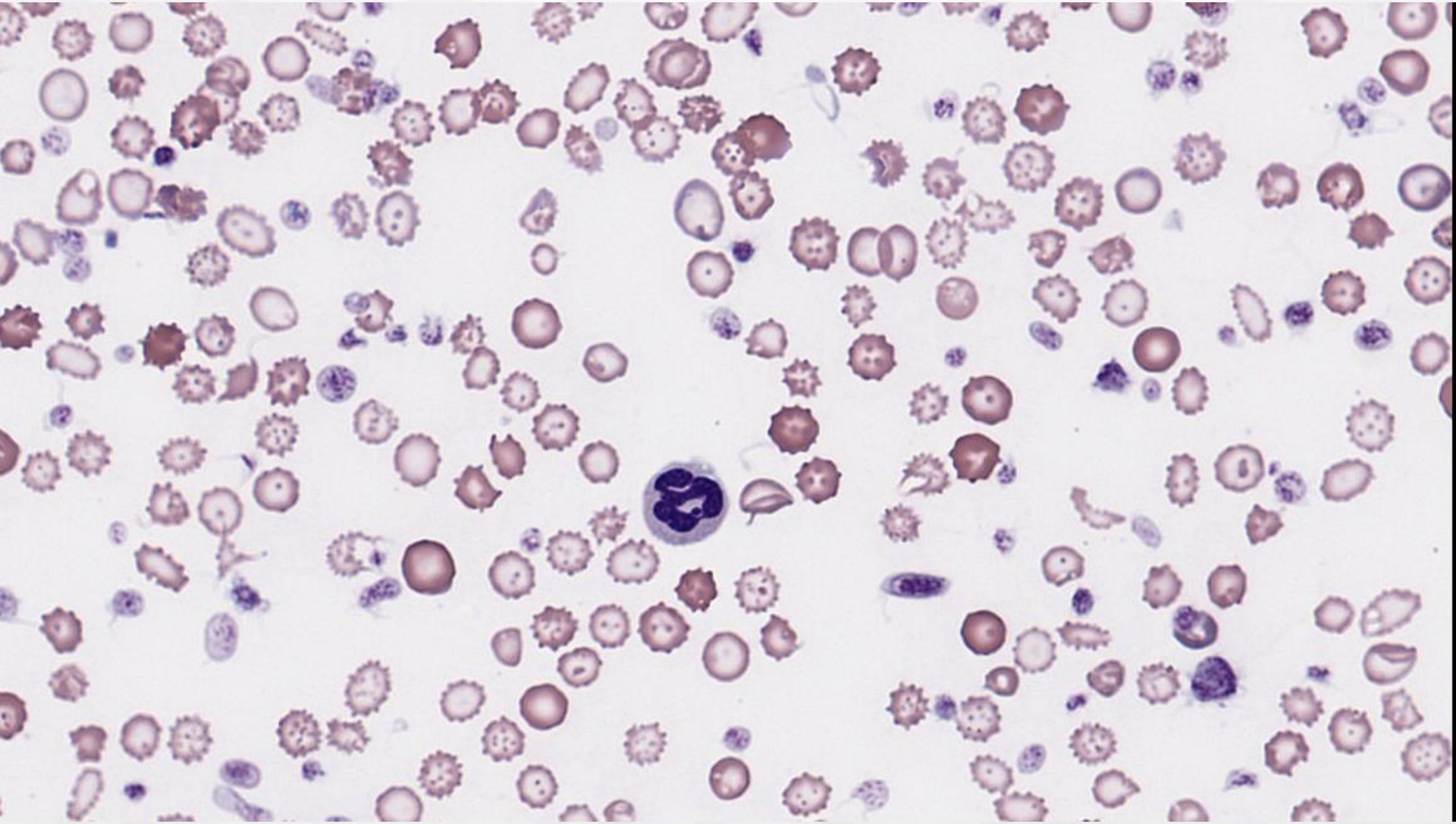
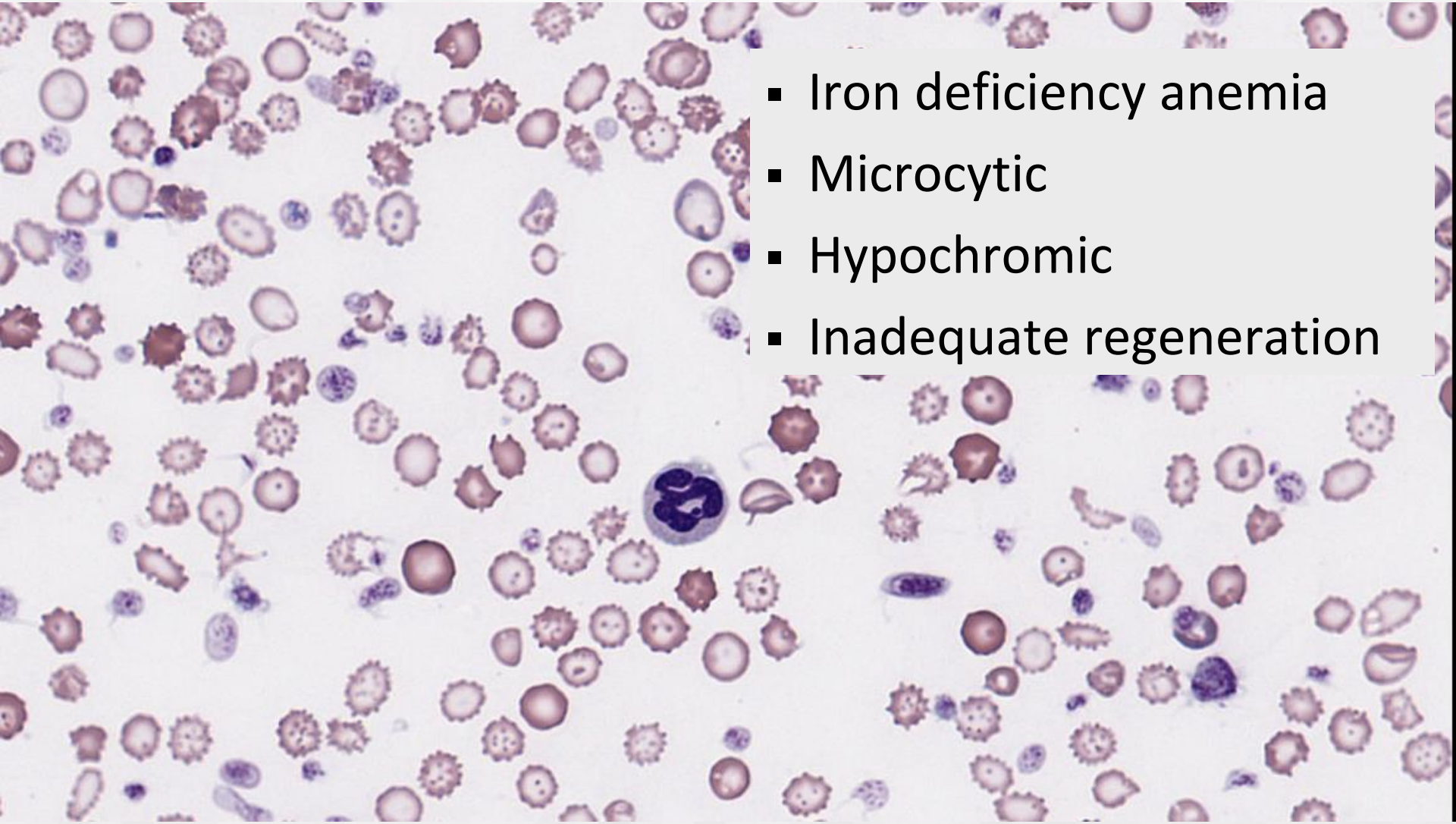
RBC Case 2

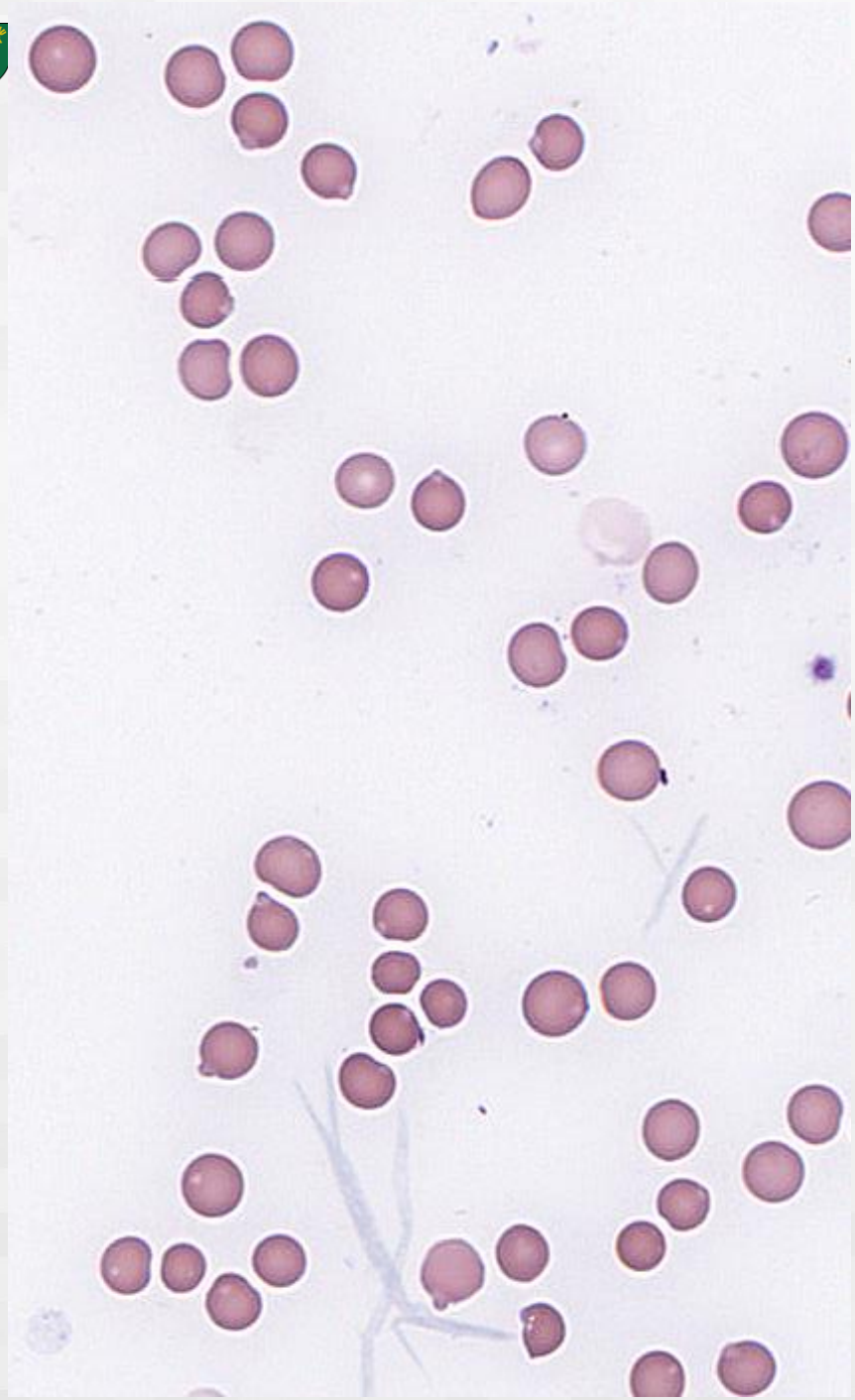


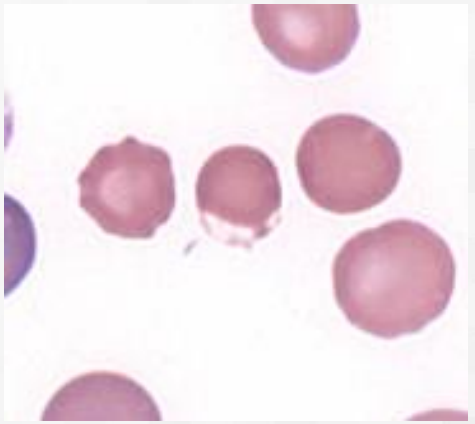
RBC Case 2

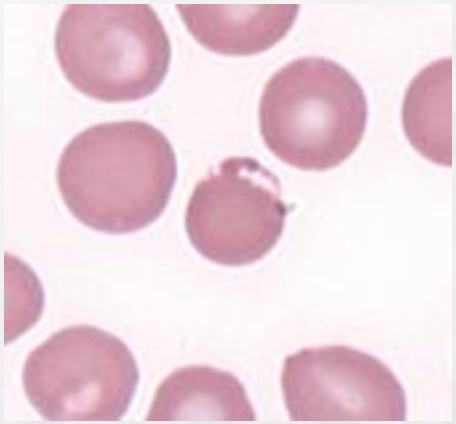


- Iron deficiency anemia
- Microcytic
- Hypochromic
- Inadequate regeneration

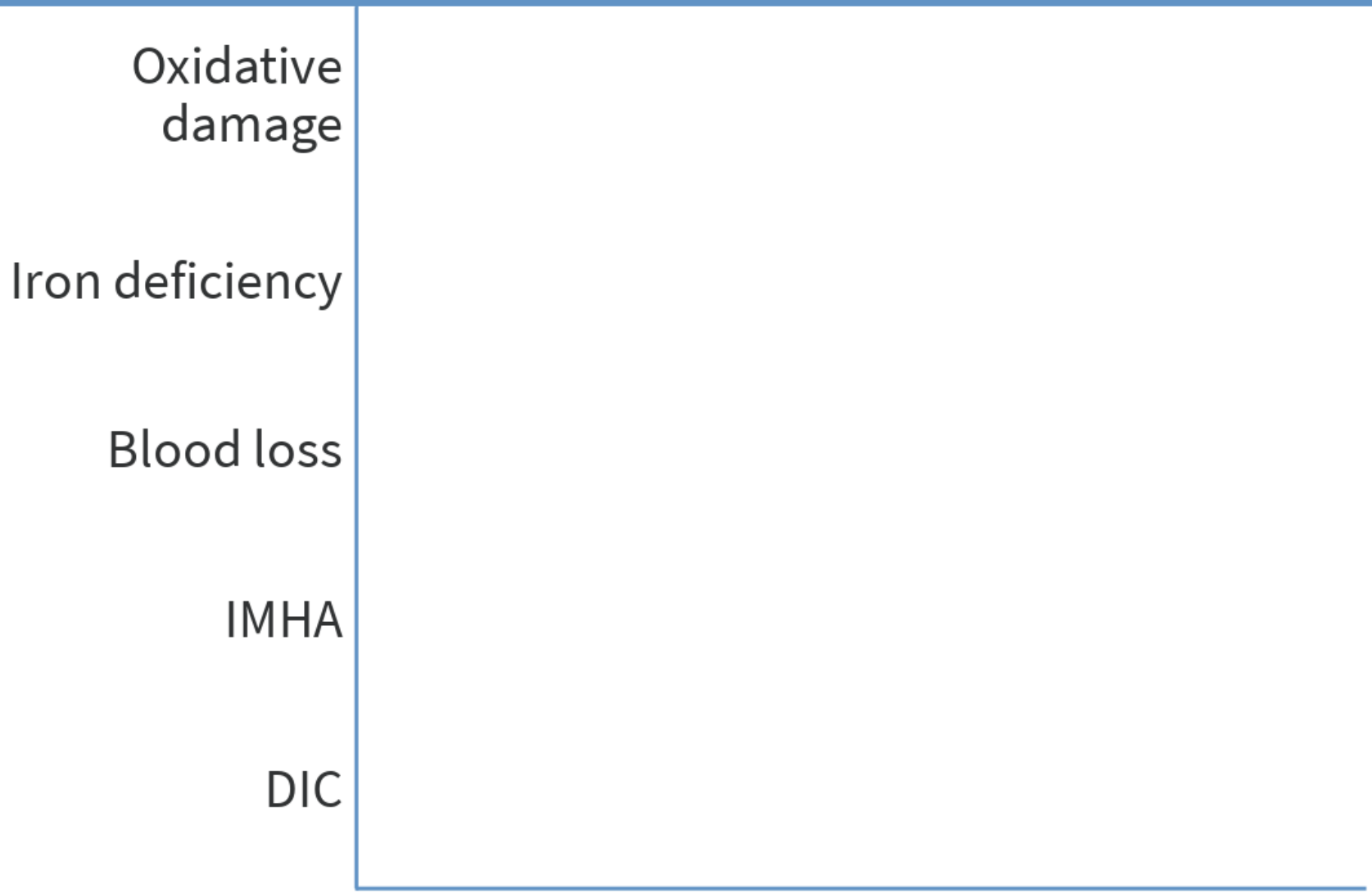
RBC Case 3







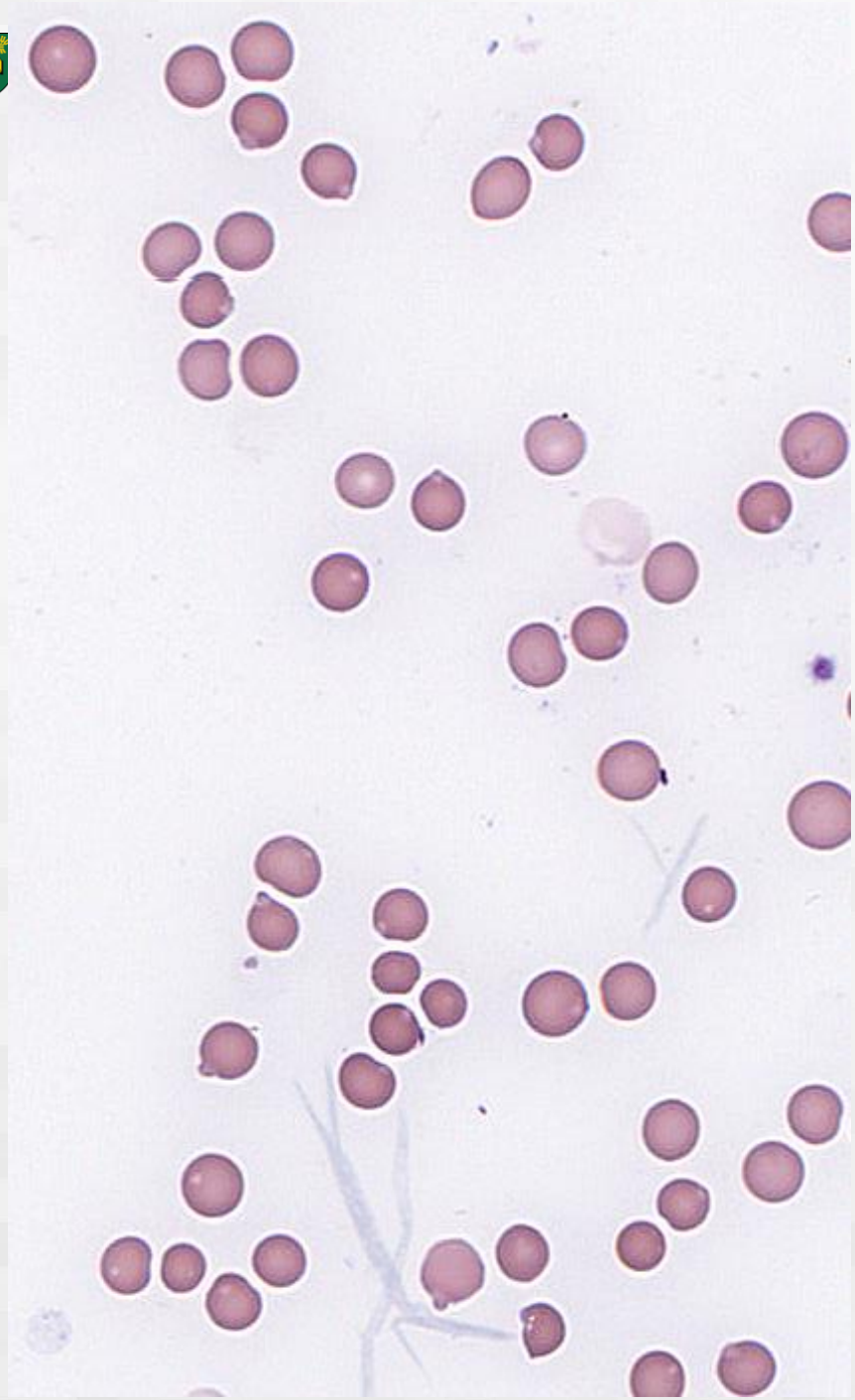
What is the cause of the anemia?

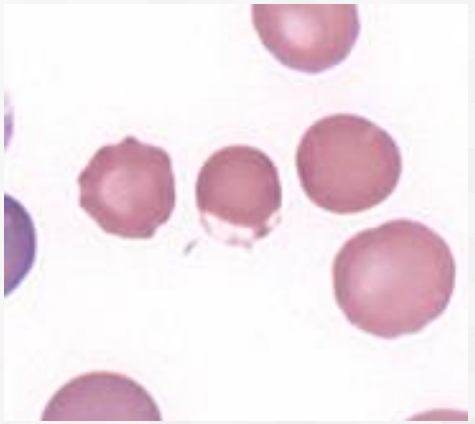


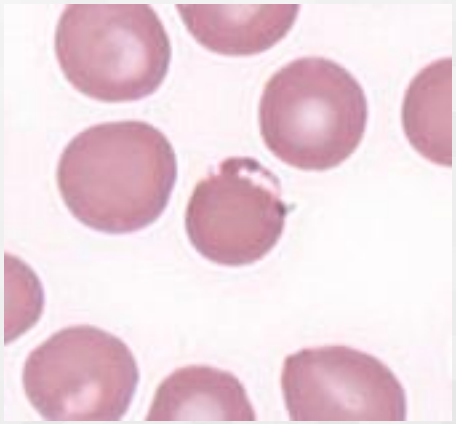
Respond at **PollEv.com/nicolefernan414**

Text **NICOLEFERNAN414** to **37607** once to join, then **A, B, or C**

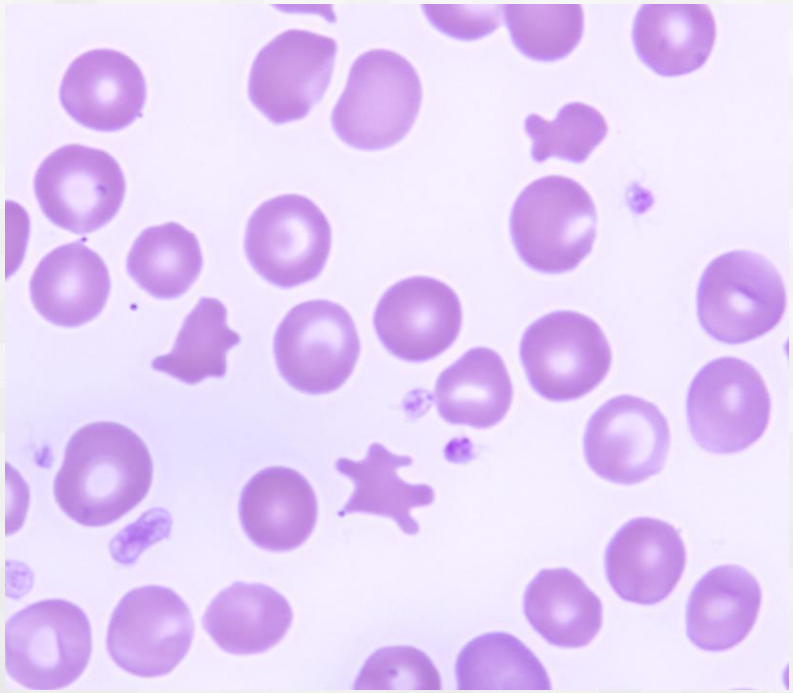
Oxidative Damage



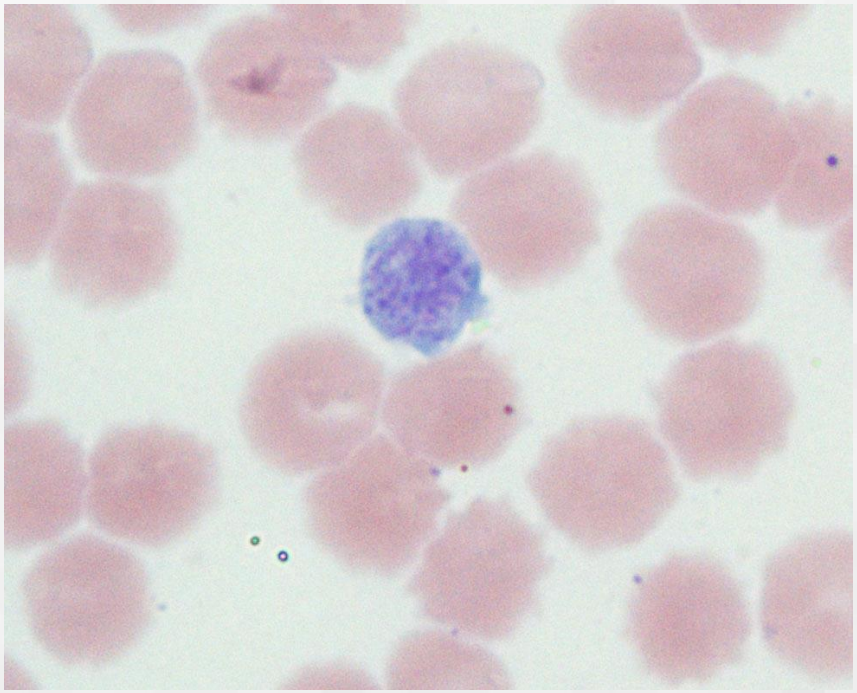




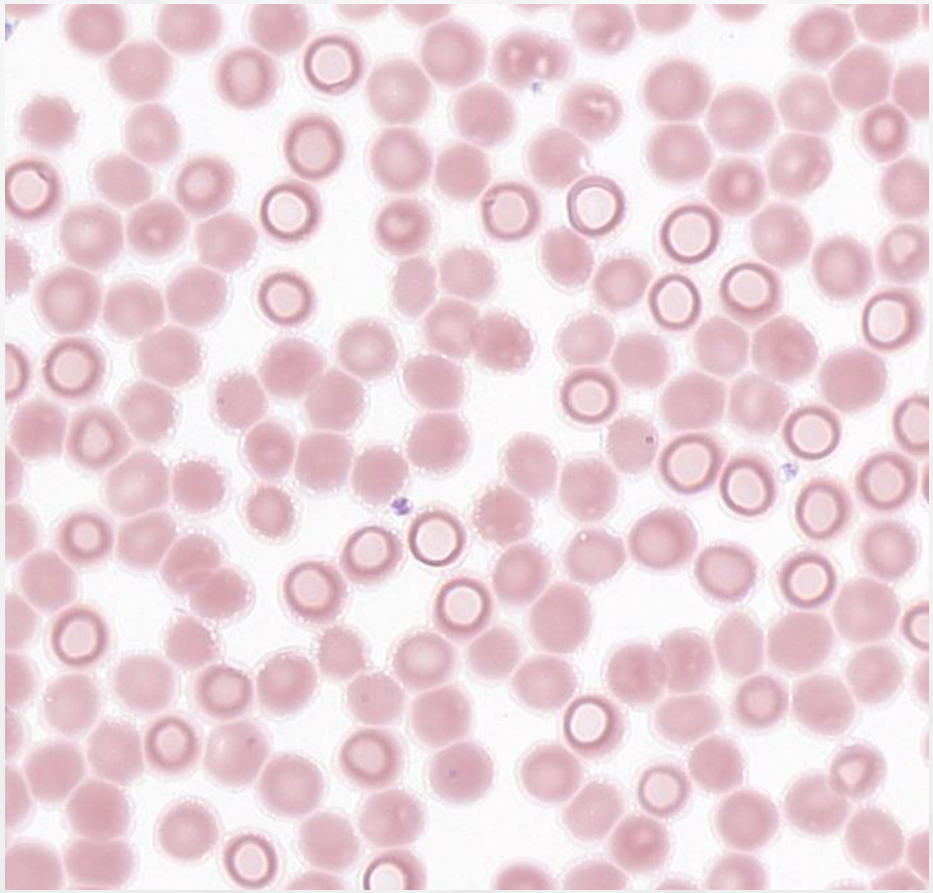
Acanthocytes



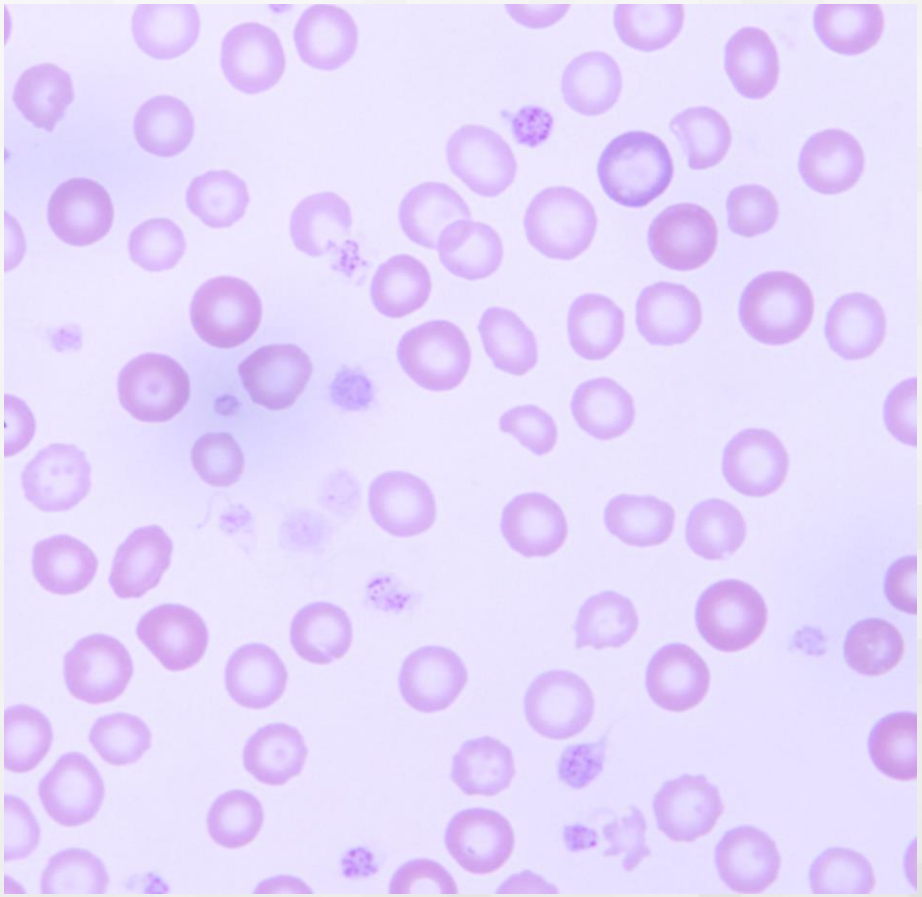
Echinocytes



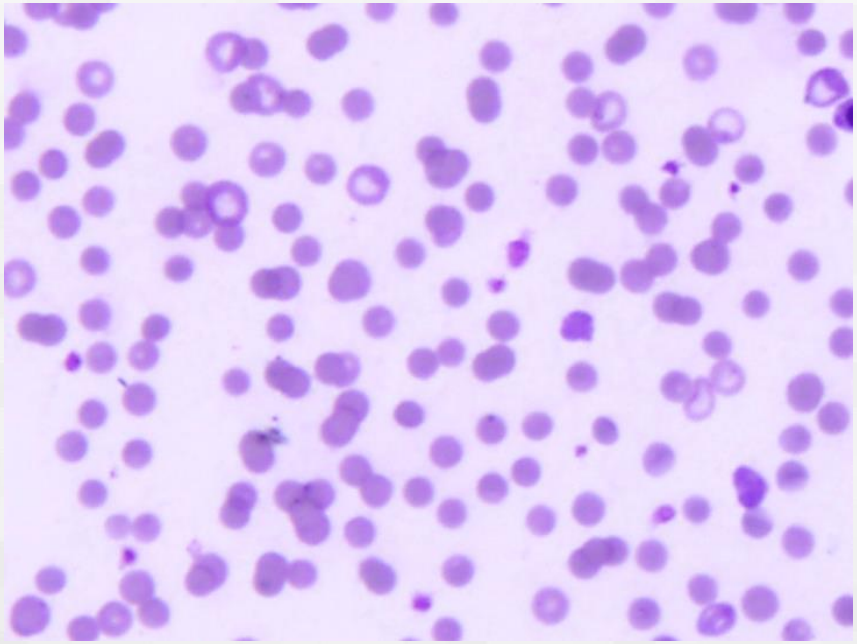
Torocytes



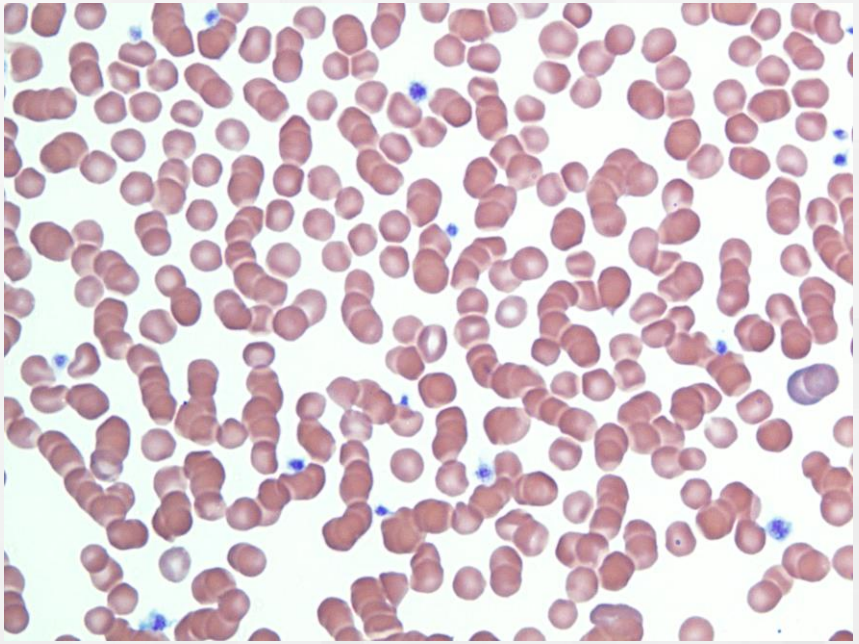
Hypochromasia



Agglutination



Rouleaux



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Questions?

WBC coming up…

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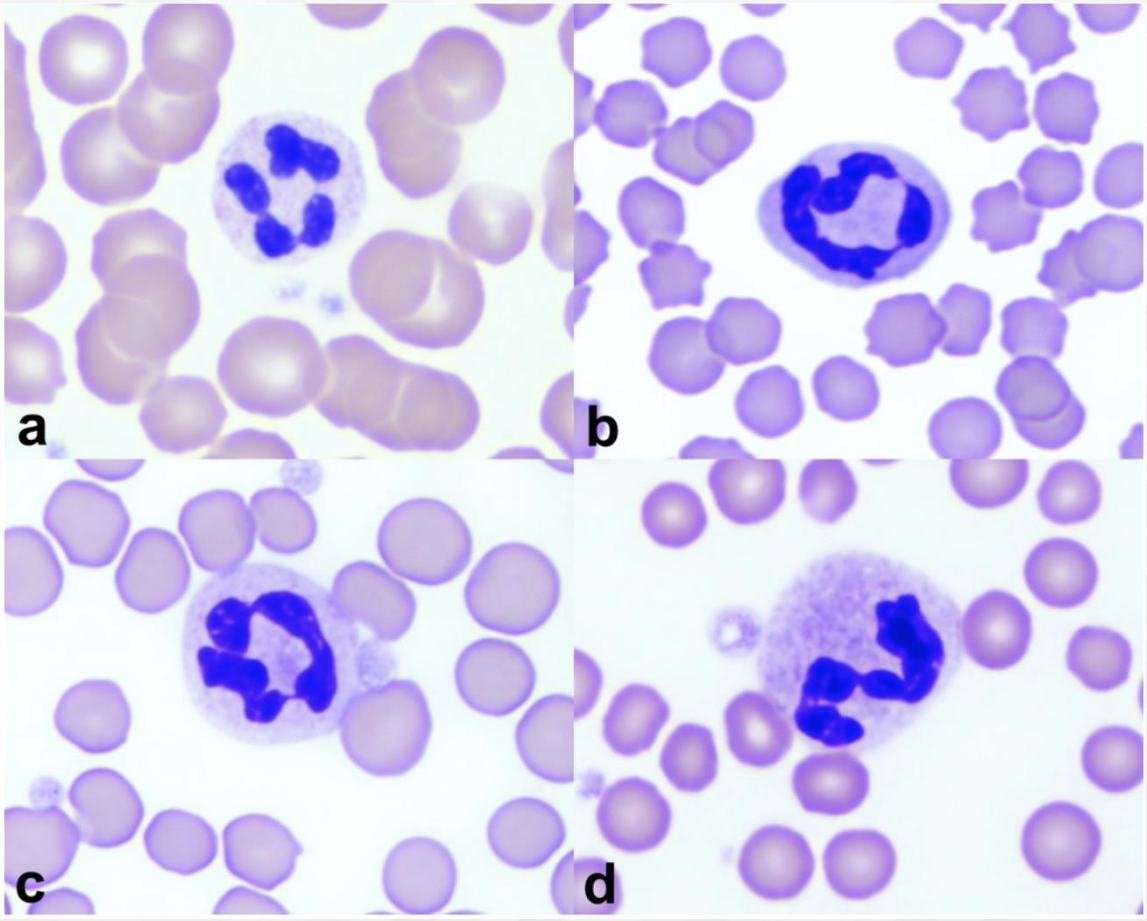
Leukocyte Review

Dog

Cat



Horse

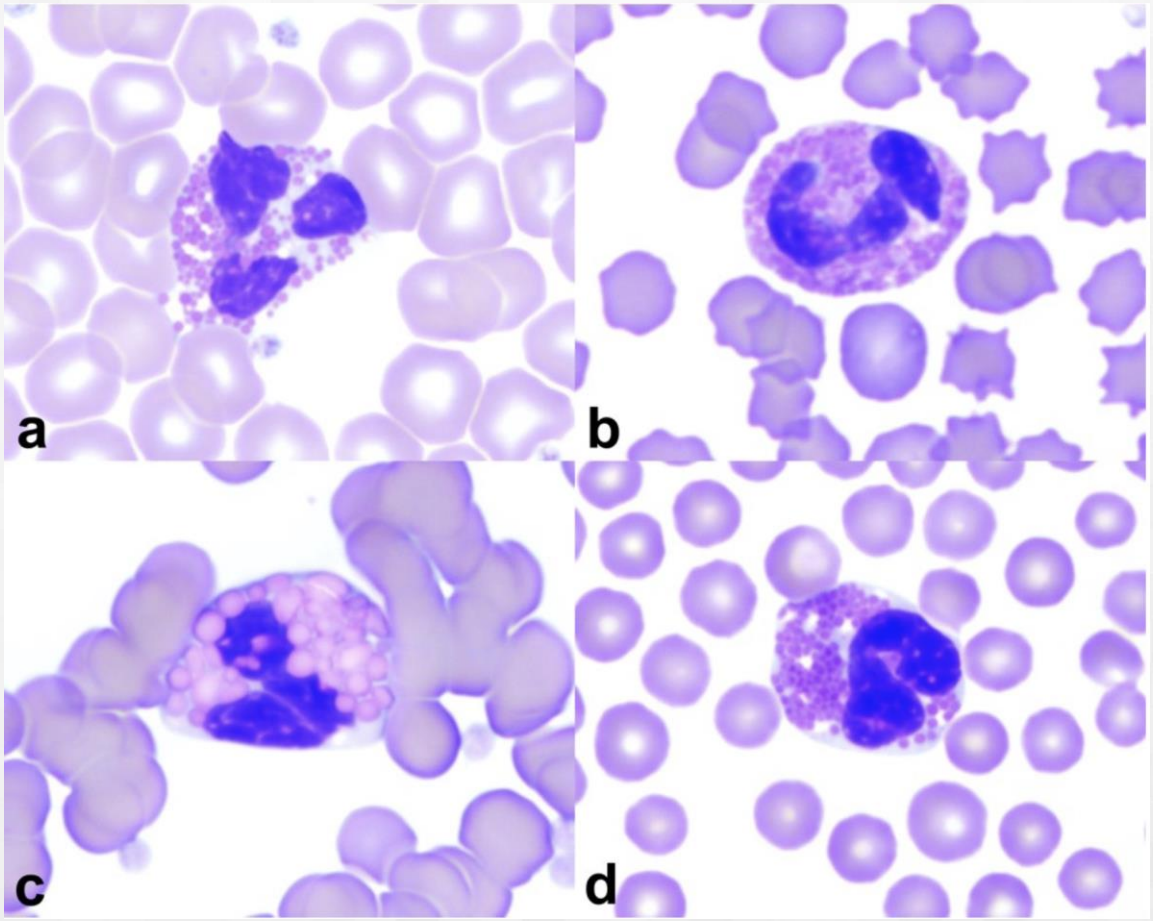
Cow

UNIVERSITY OF SASKATCHEWAN

Leukocyte Review

Dog

Cat



Horse

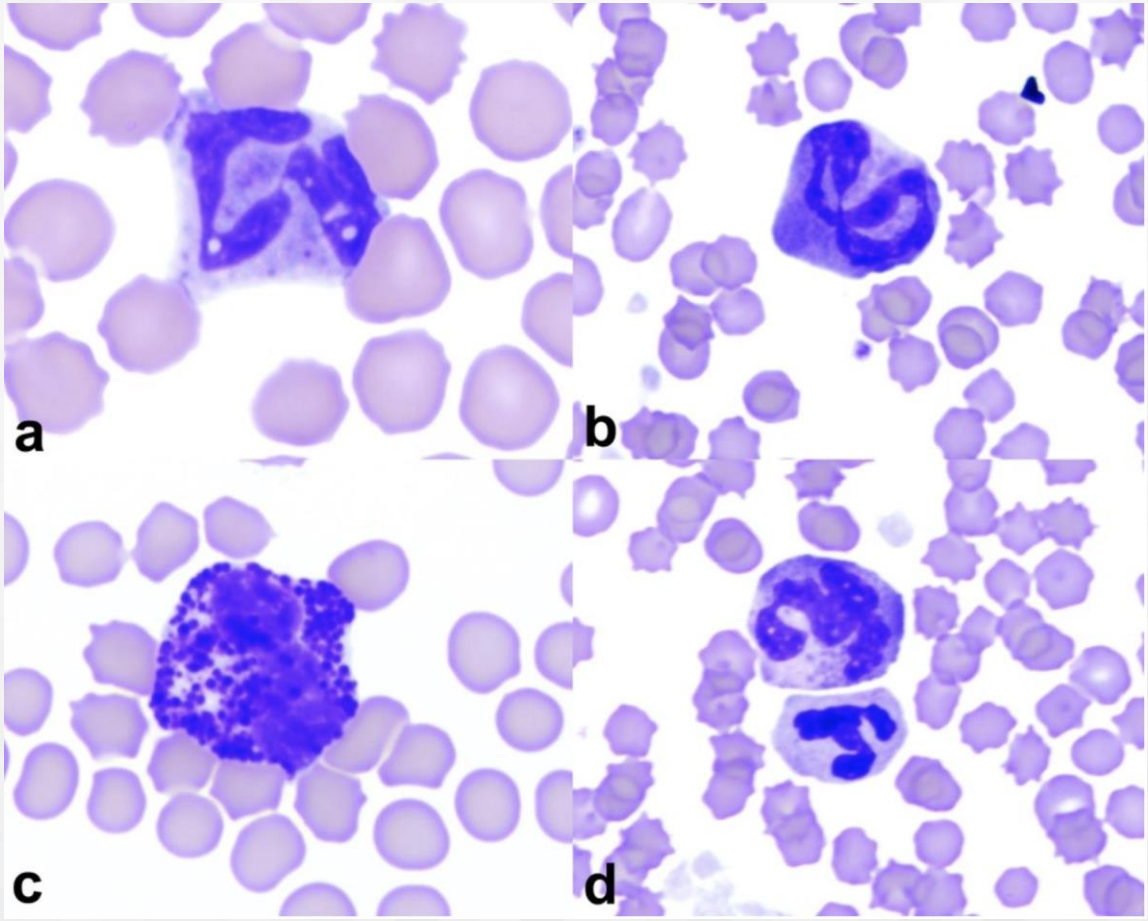
Cow

UNIVERSITY OF SASKATCHEWAN

Leukocyte Review

Dog

Cat



Horse

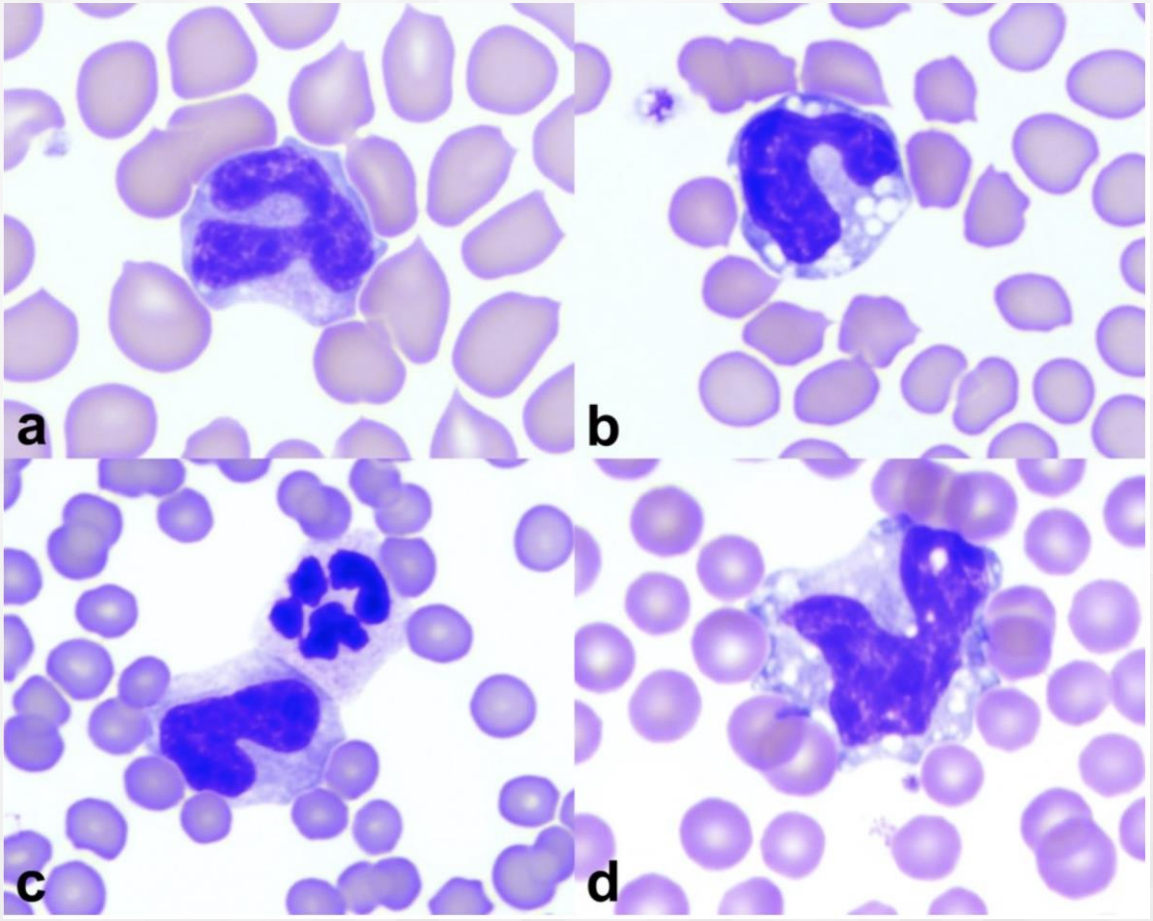
Cow

UNIVERSITY OF SASKATCHEWAN

Leukocyte Review

Dog

Cat



Horse

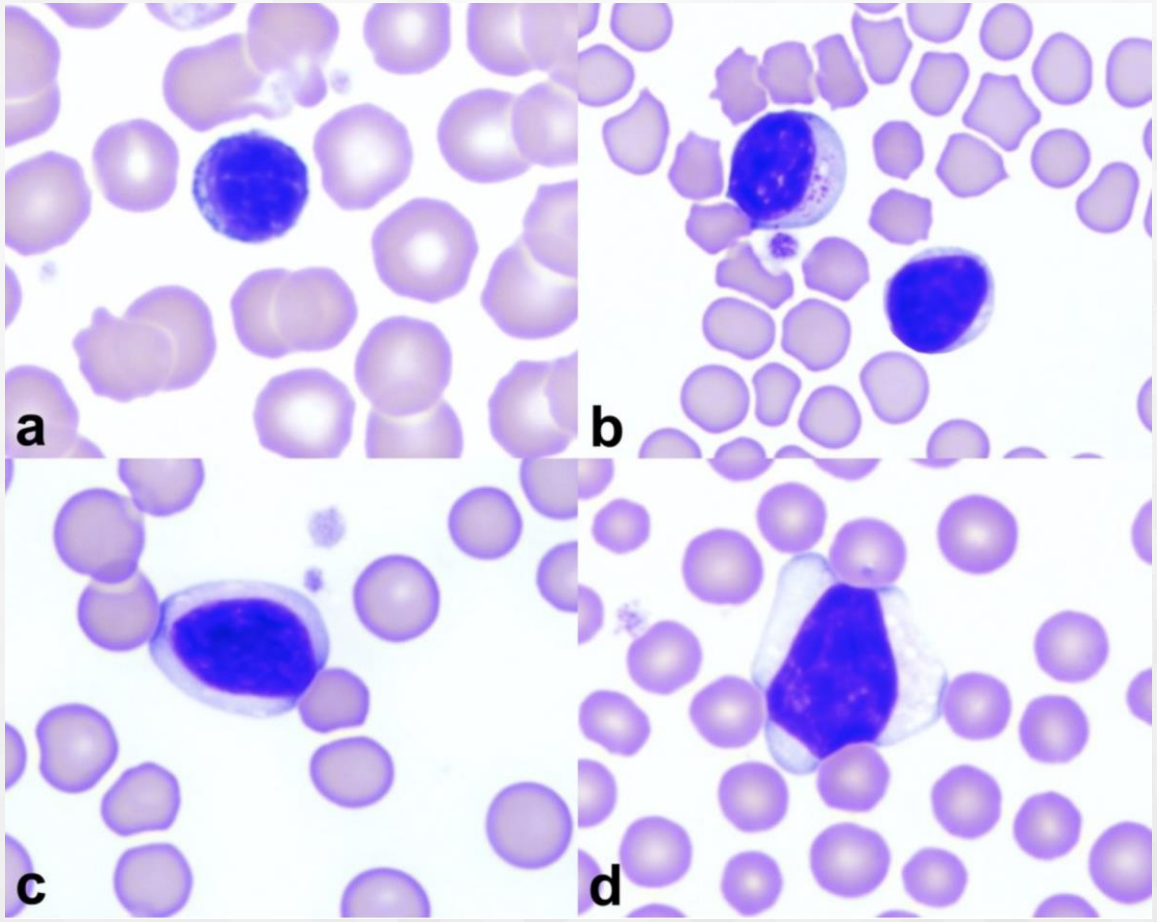
Cow

UNIVERSITY OF SASKATCHEWAN

Leukocyte Review

Dog

Cat

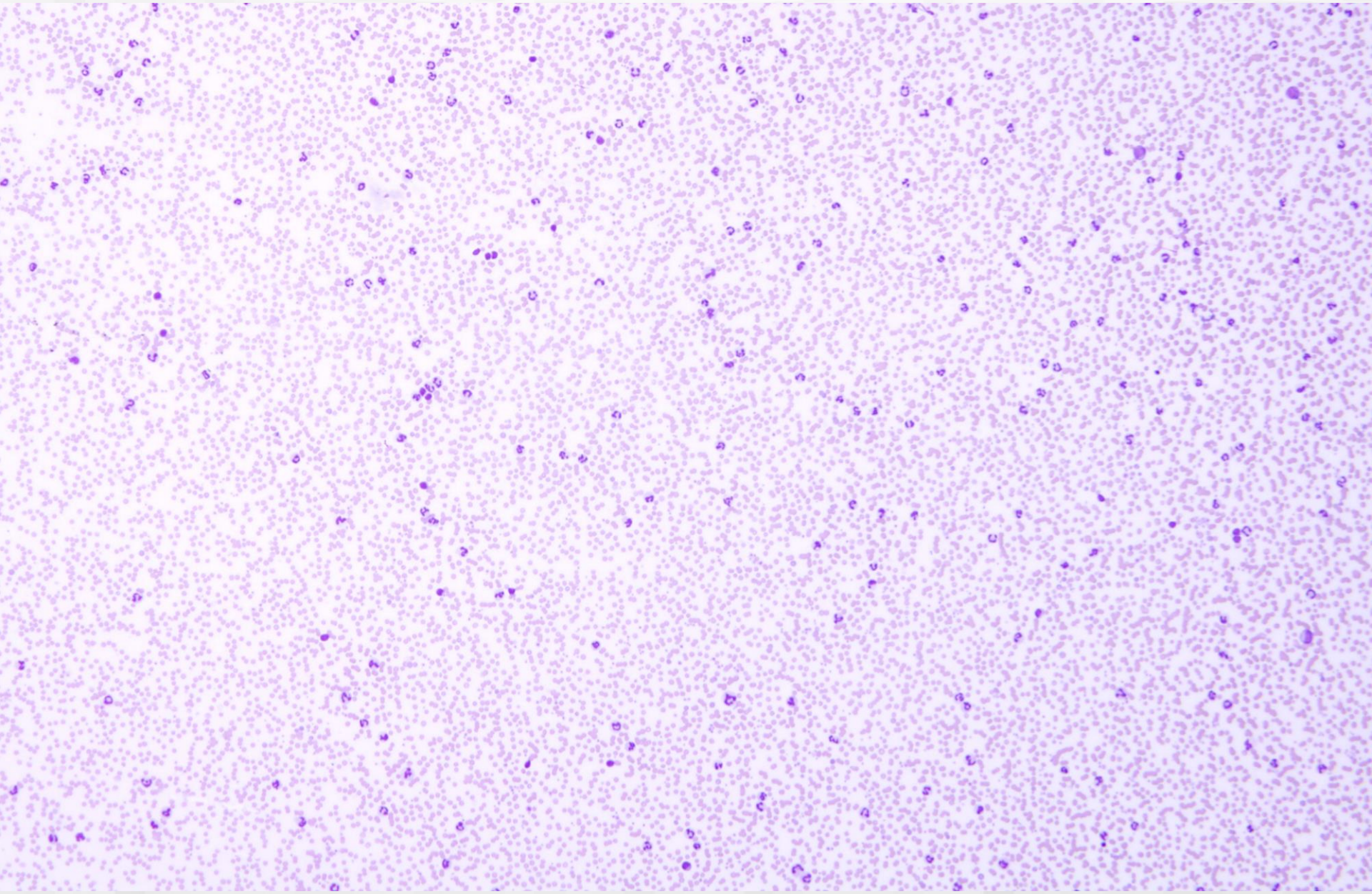


Horse

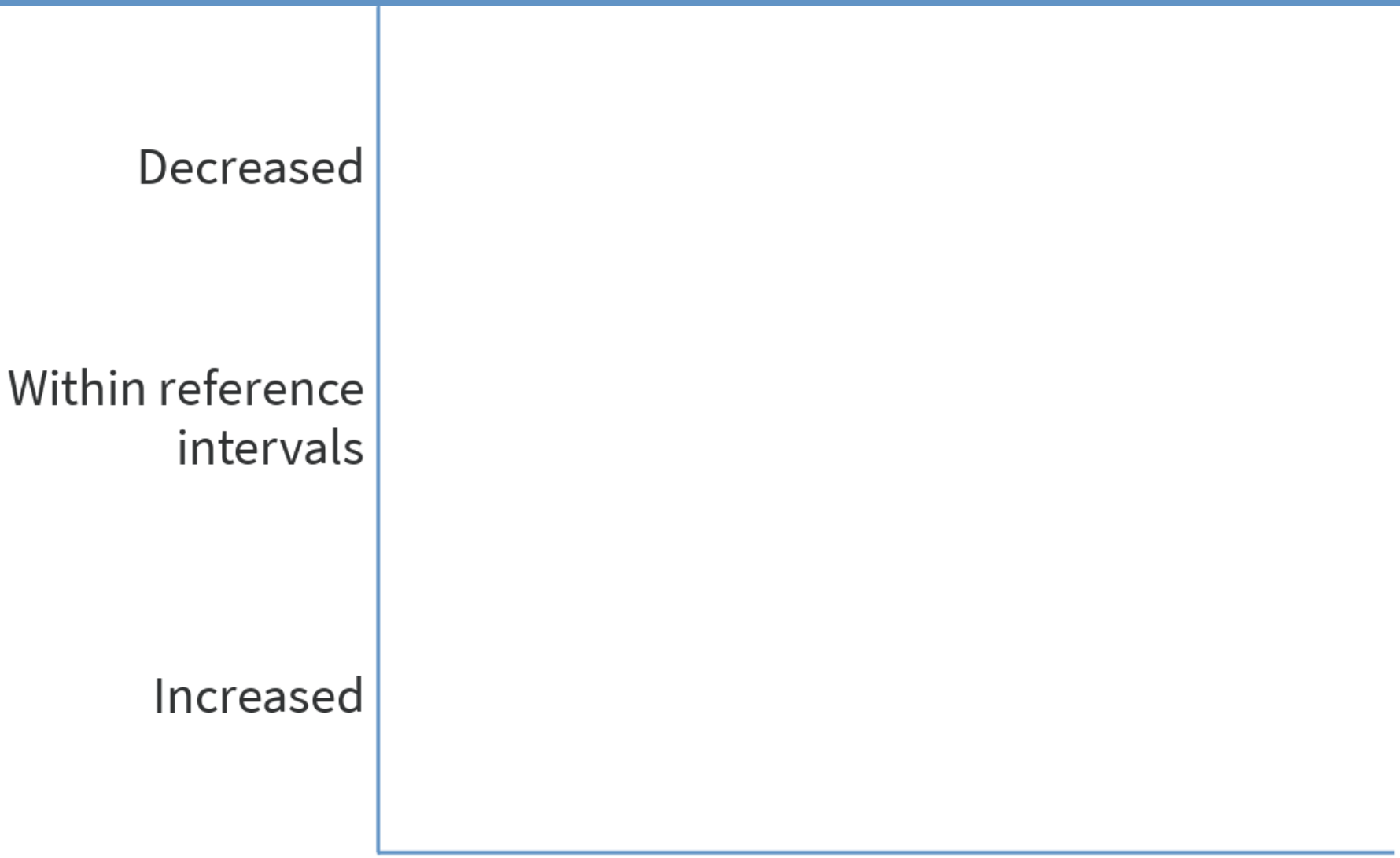
Cow

University of Saskatchewan

WBC Case 1



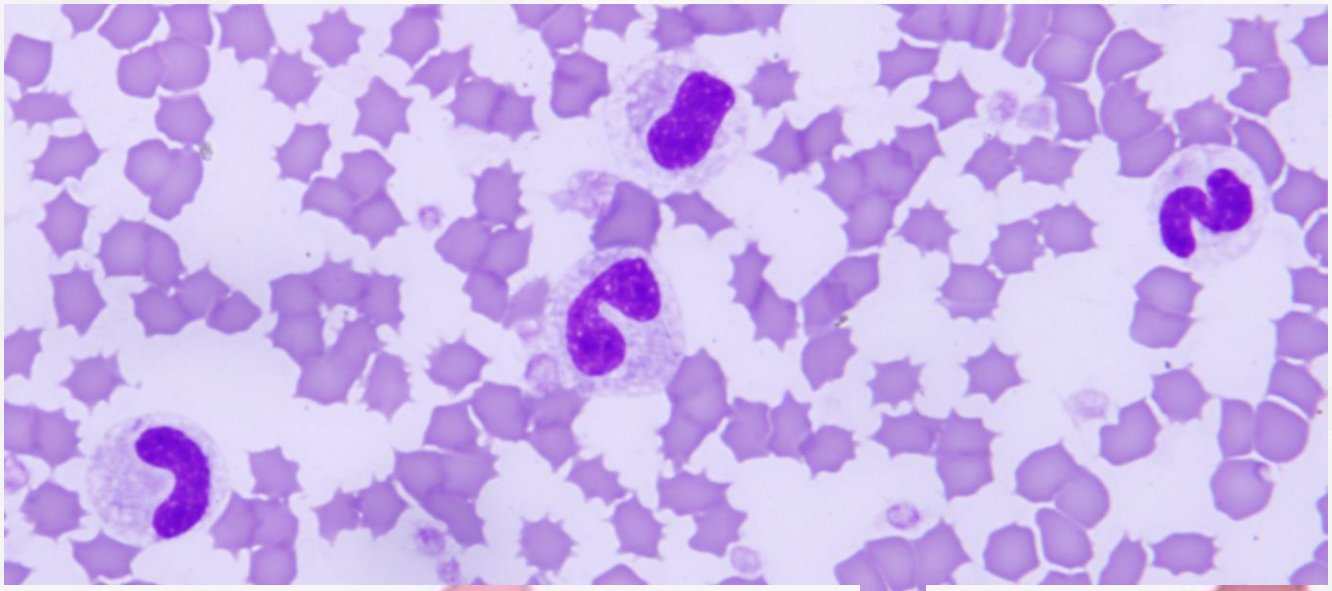
Are WBC numbers...

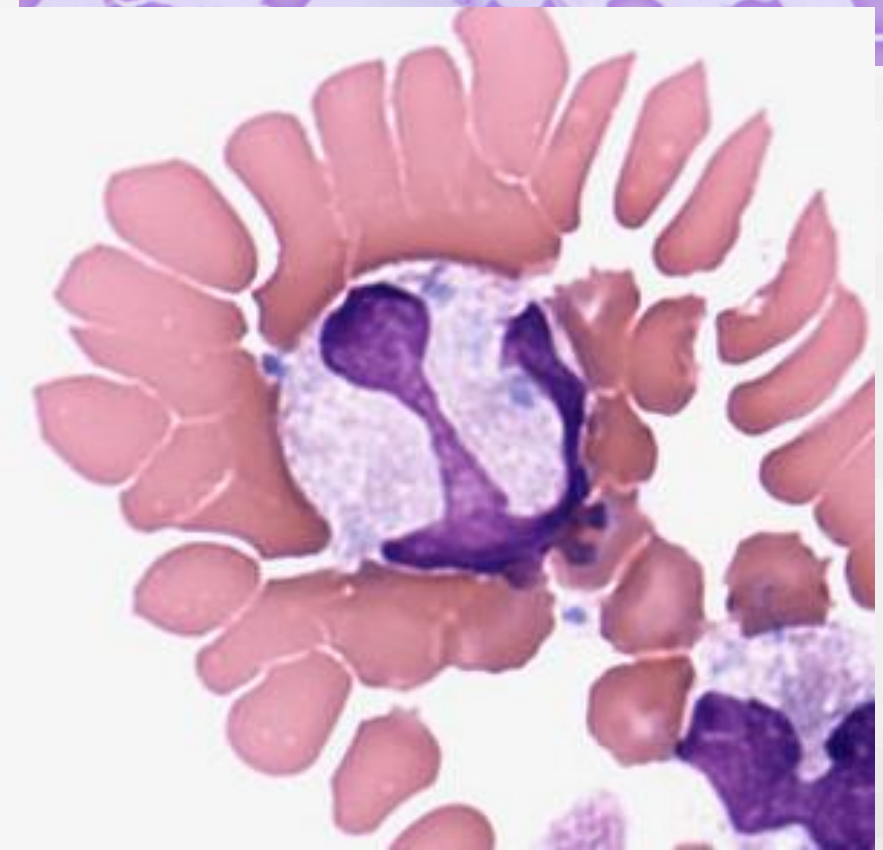


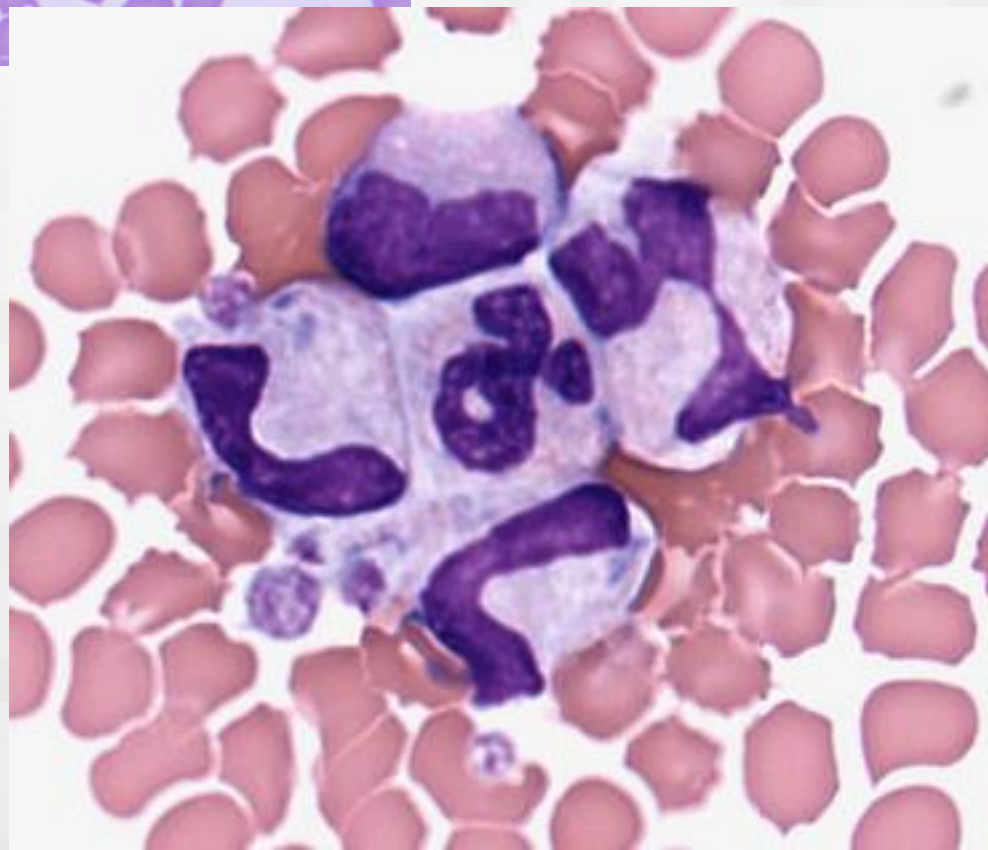
WBC Estimate 10x

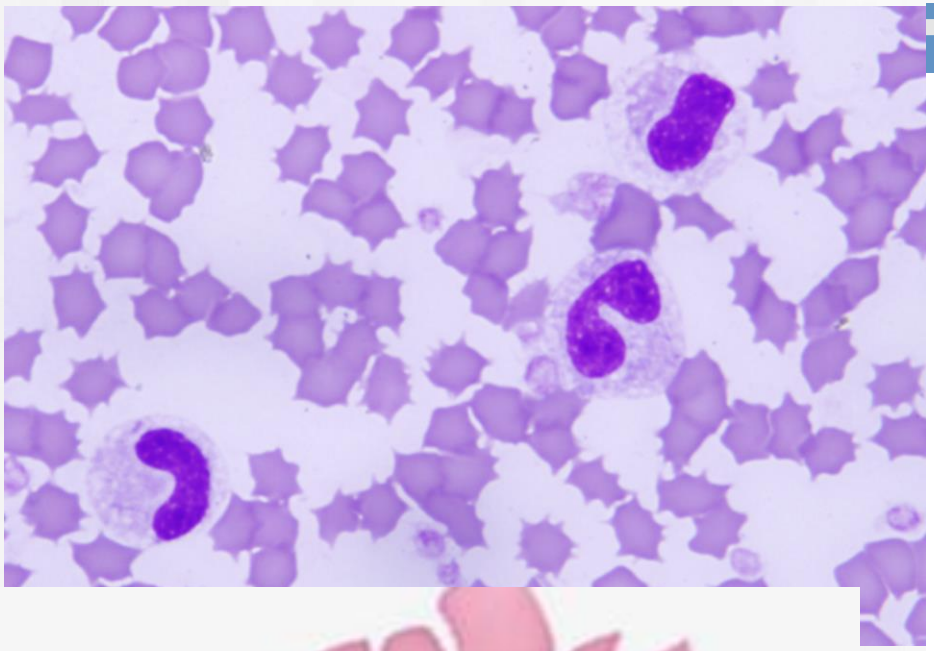
- Count the leukocytes in at least 3 fields in the **monolayer**
- Divide the total number of leukocytes counted by the number of fields to obtain the average number per field
- Divide the average number of leukocytes counted by 4 in order to approximate the number of leukocytes $x10^9/L$

WBC Case 1









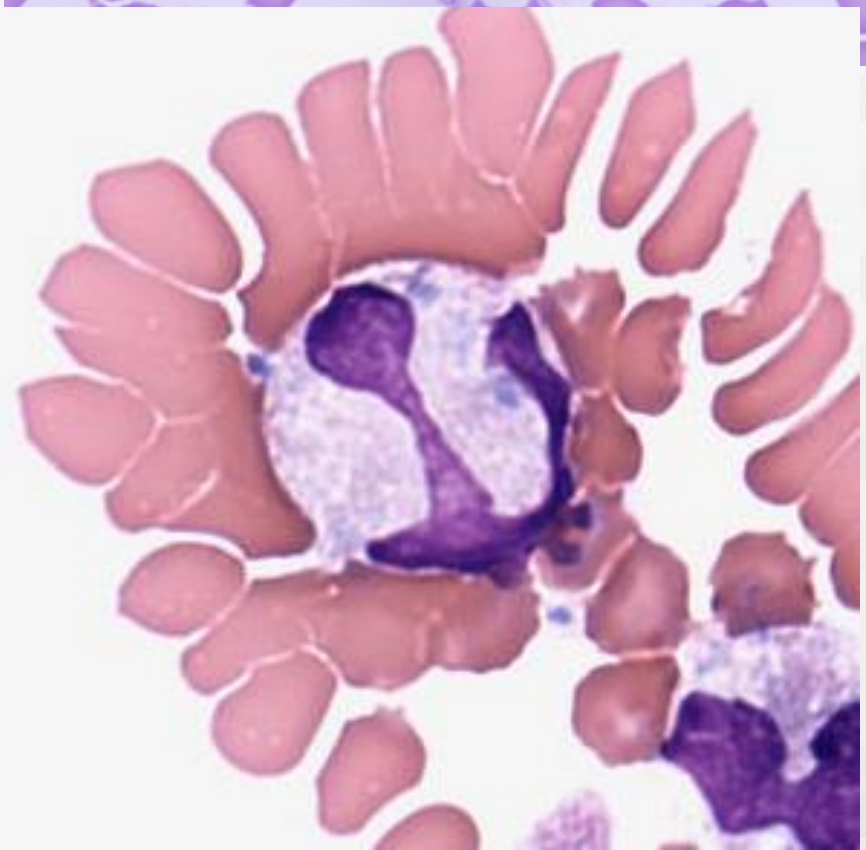
What does the WBC morphology indicate?

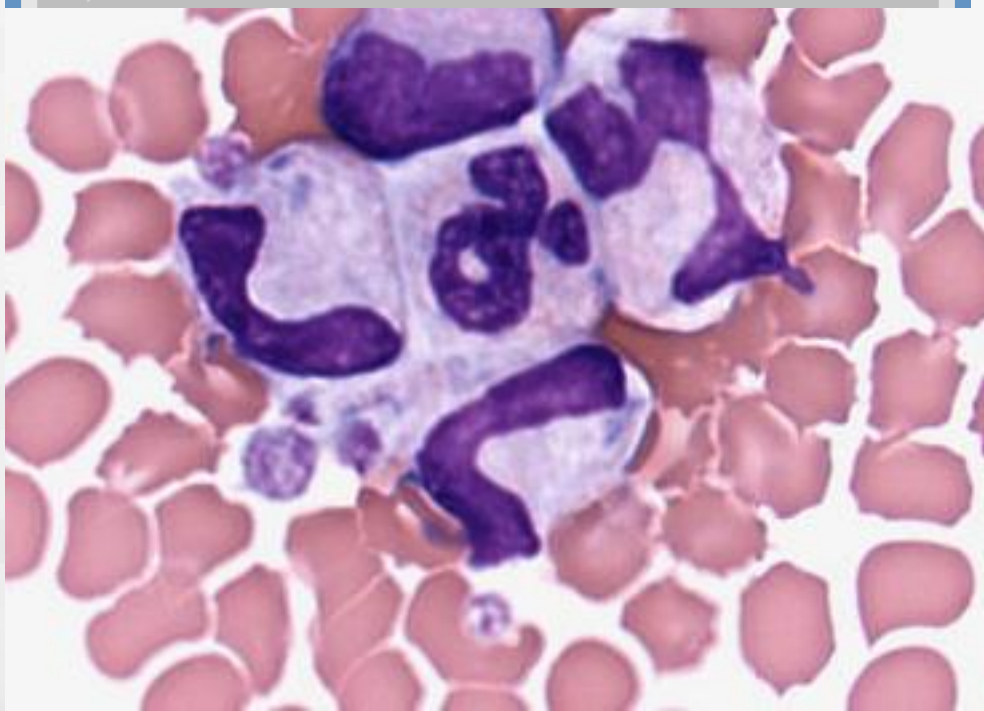
There is a left shift

There is toxic change

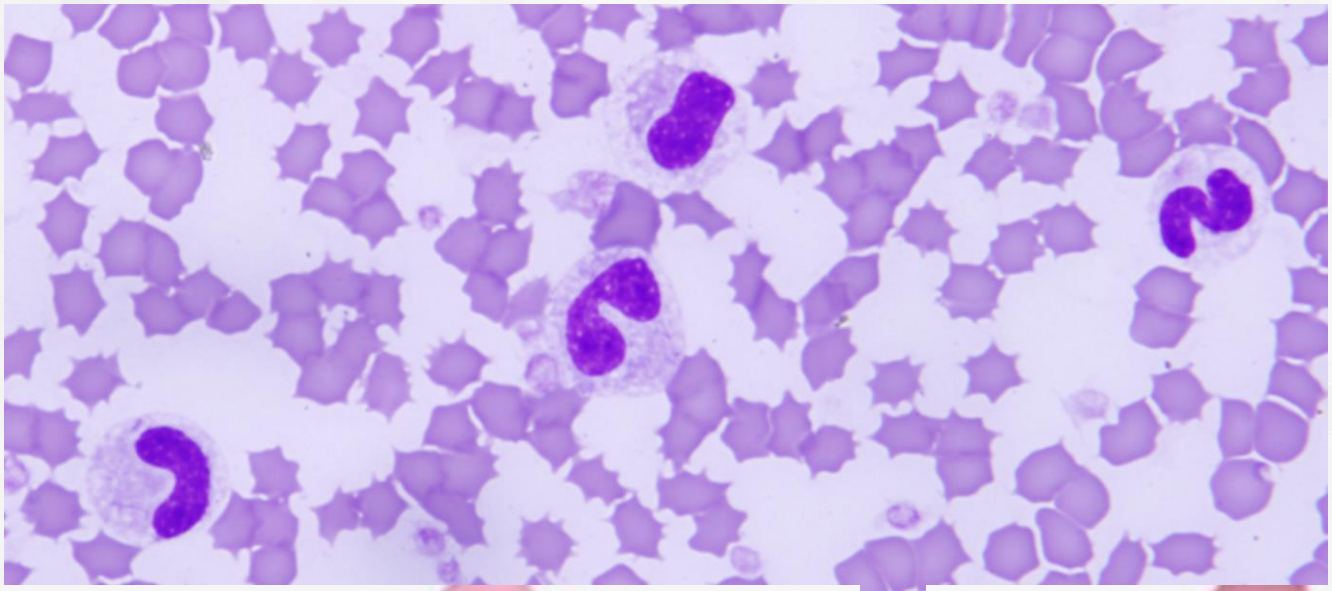
There is both a left shift and toxic change

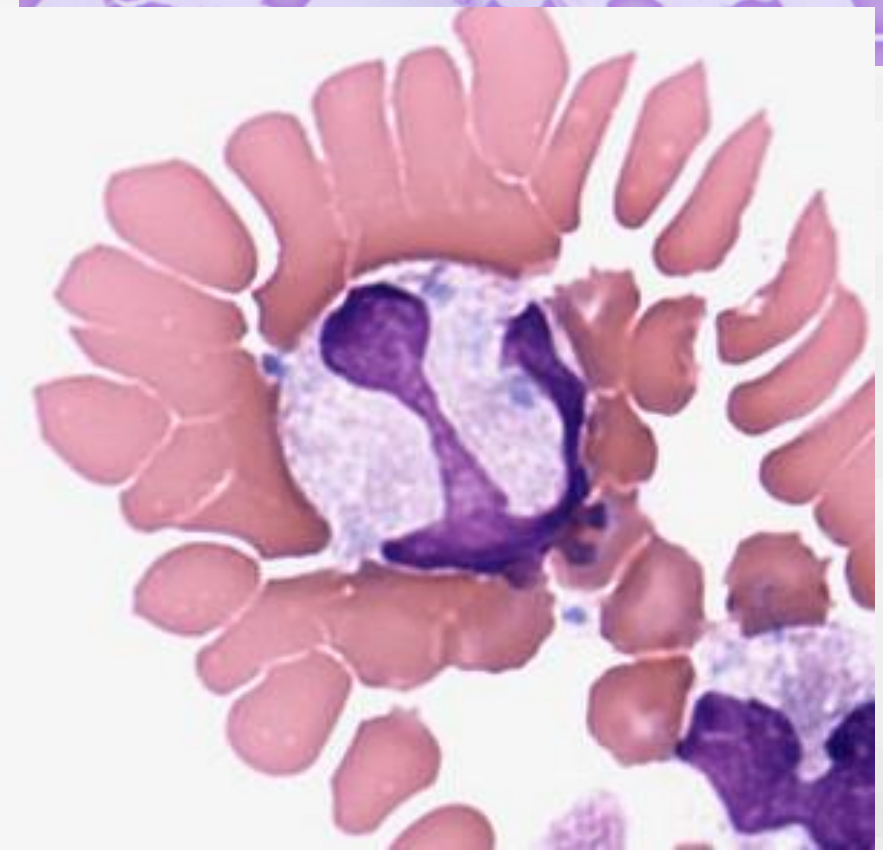
There is neither a left shift nor toxic change

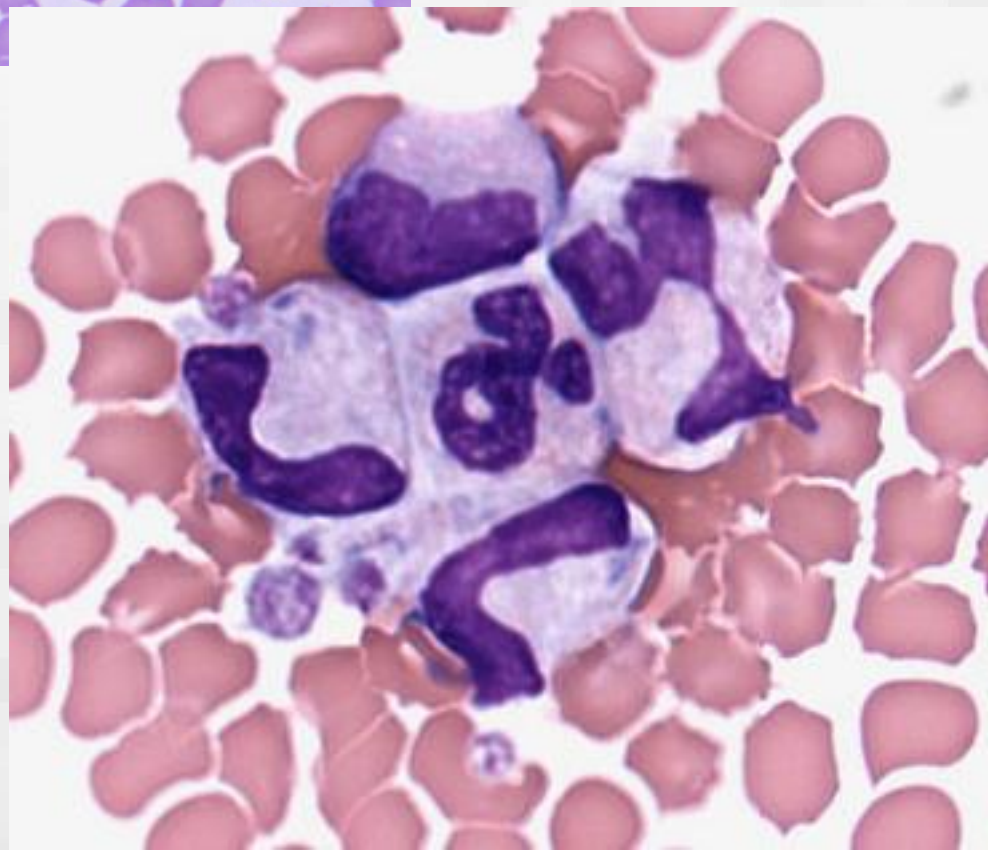


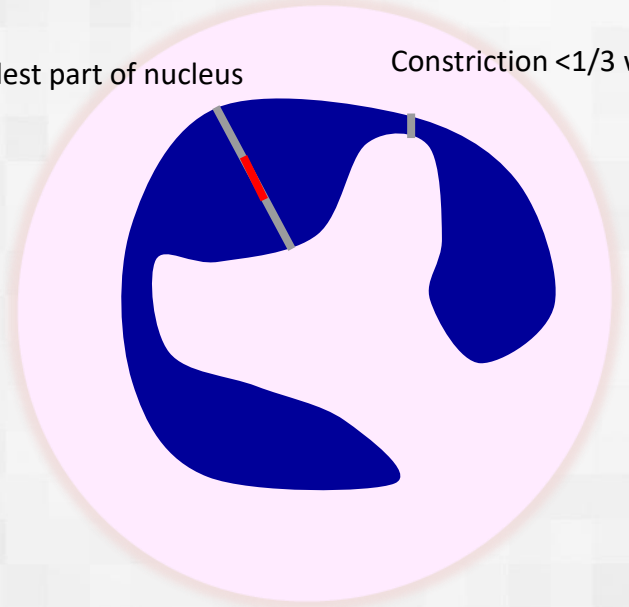


WBC Case 1
Left Shift

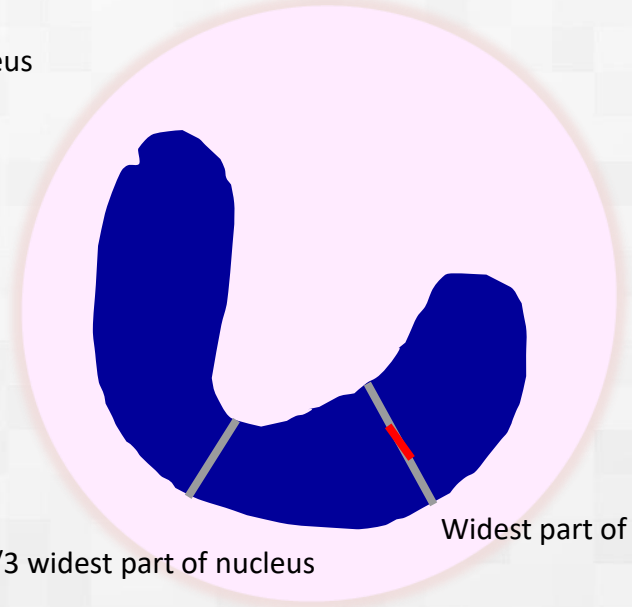






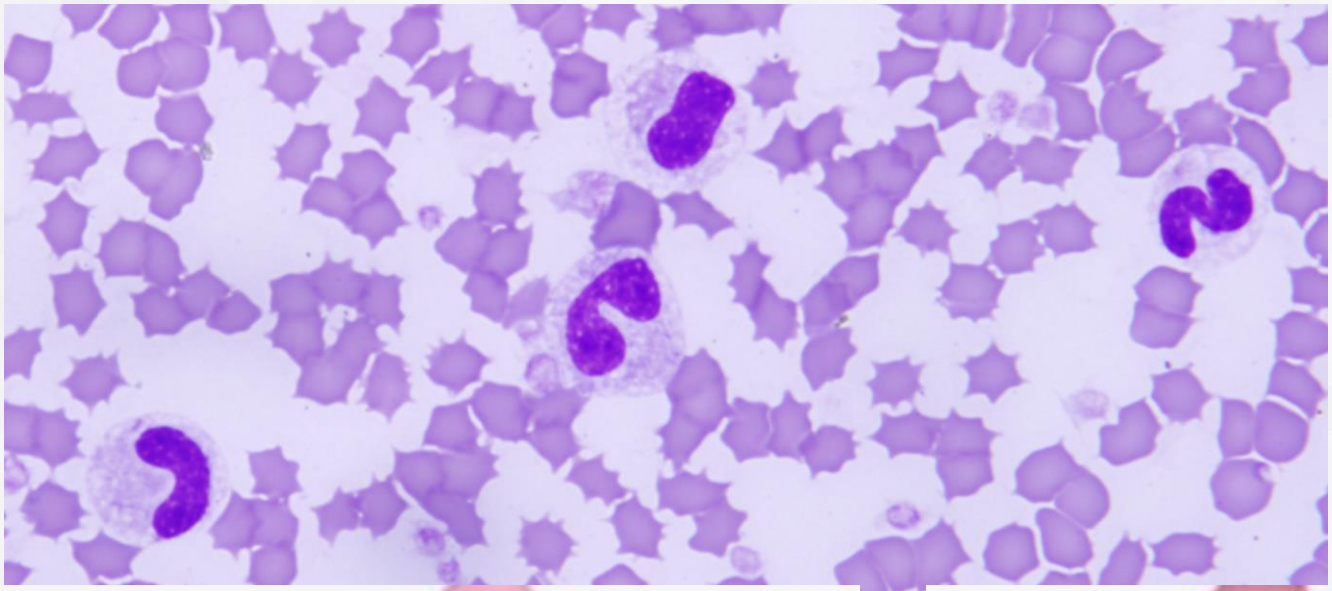


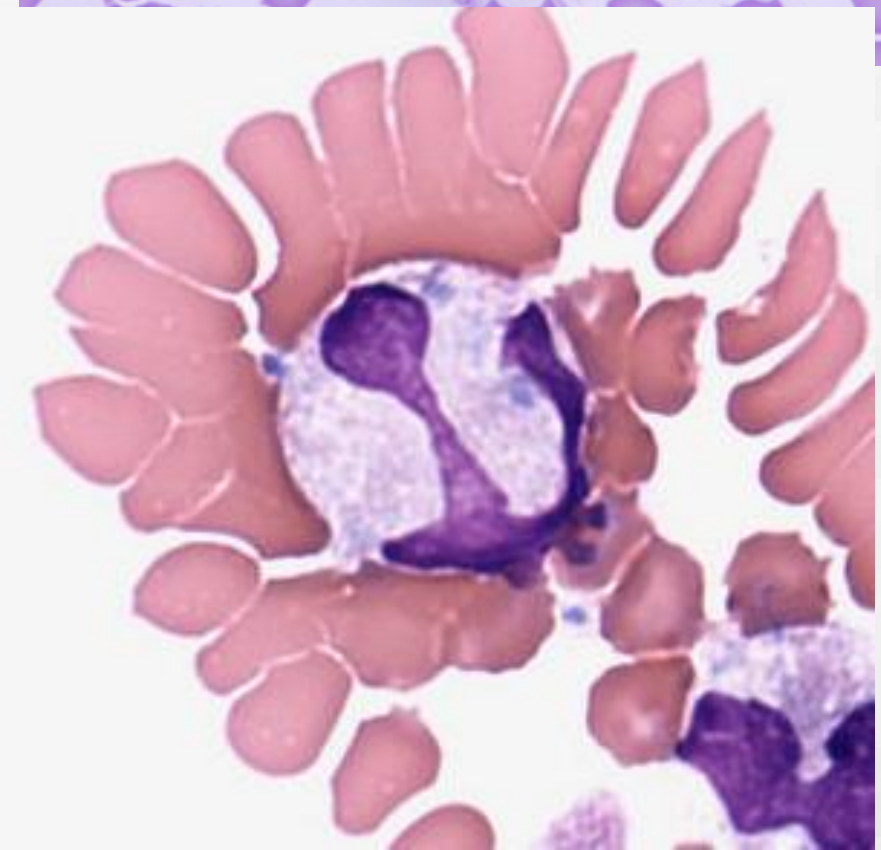
Mature seg

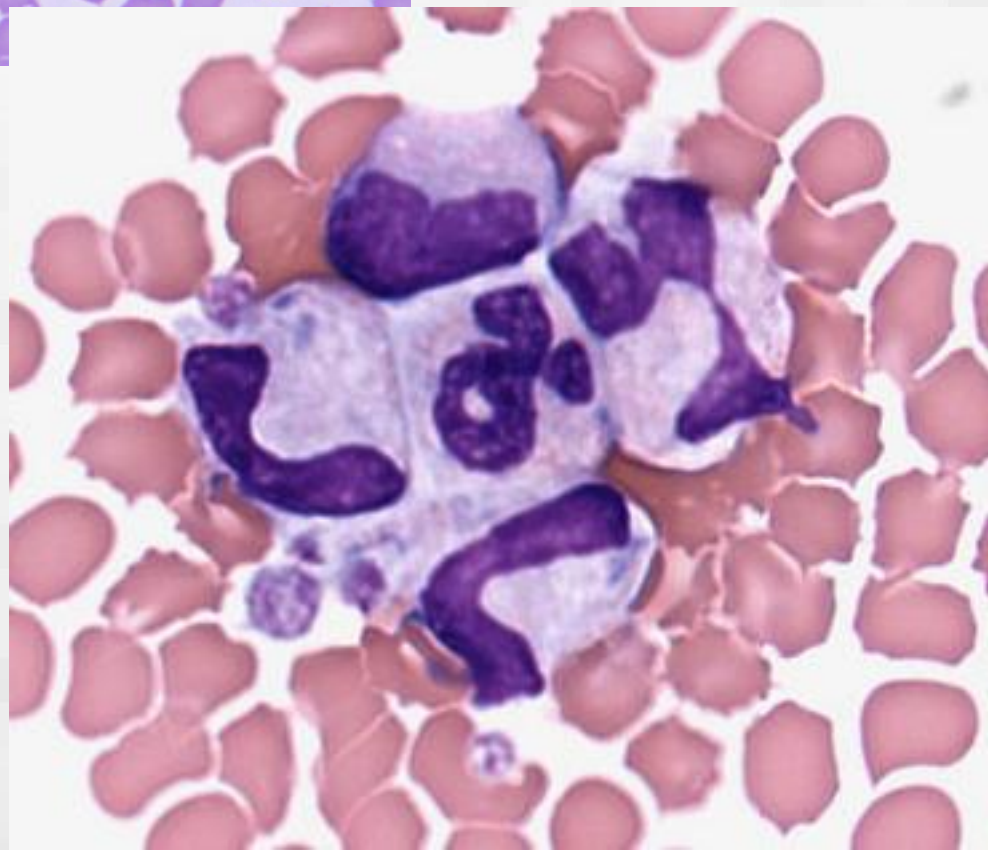


Band

WBC Case 1
Toxic Change



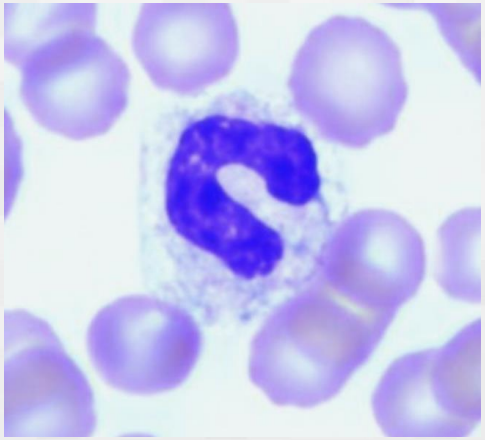




Source of Confusion

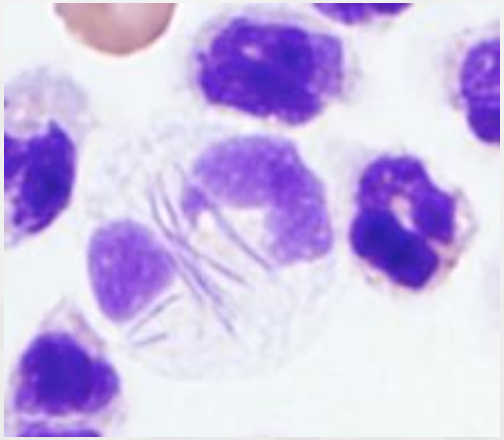
Toxic change

- In blood (blood smears)
- Cytoplasmic changes
  - Basophilia
  - Foaminess
  - Dohle bodies
  - Toxic granulation



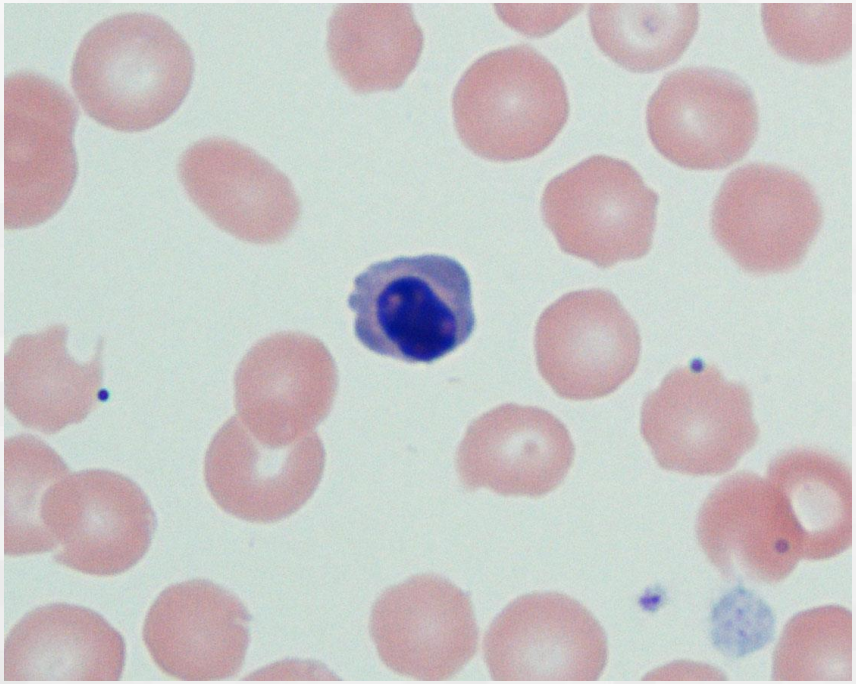
Degenerate neutrophils

- In tissues (cytology smears)
- Nuclear changes
  - Swelling
  - Pale-staining
  - Lysis
  - Hunt for bacteria

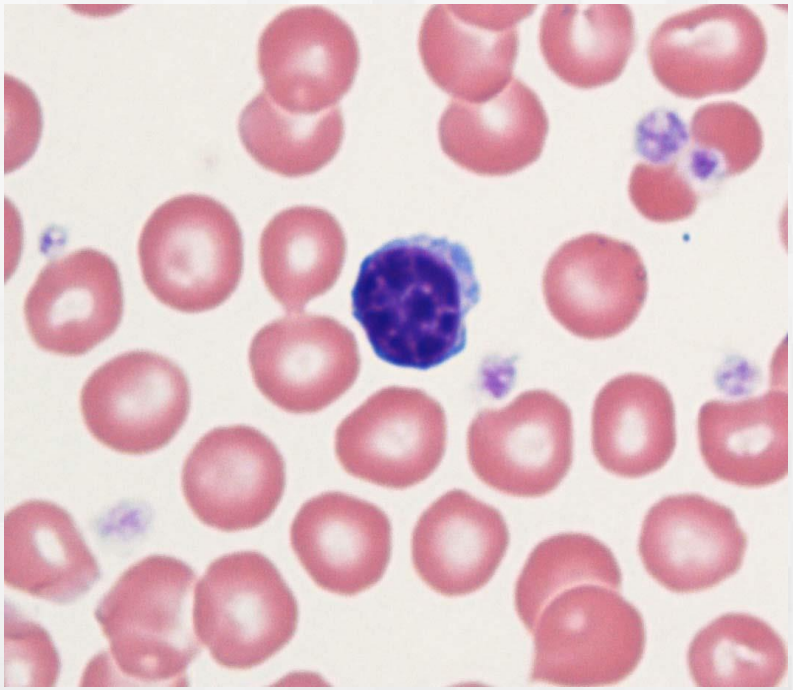


Source of Confusion

Nucleated RBC



Lymphocyte



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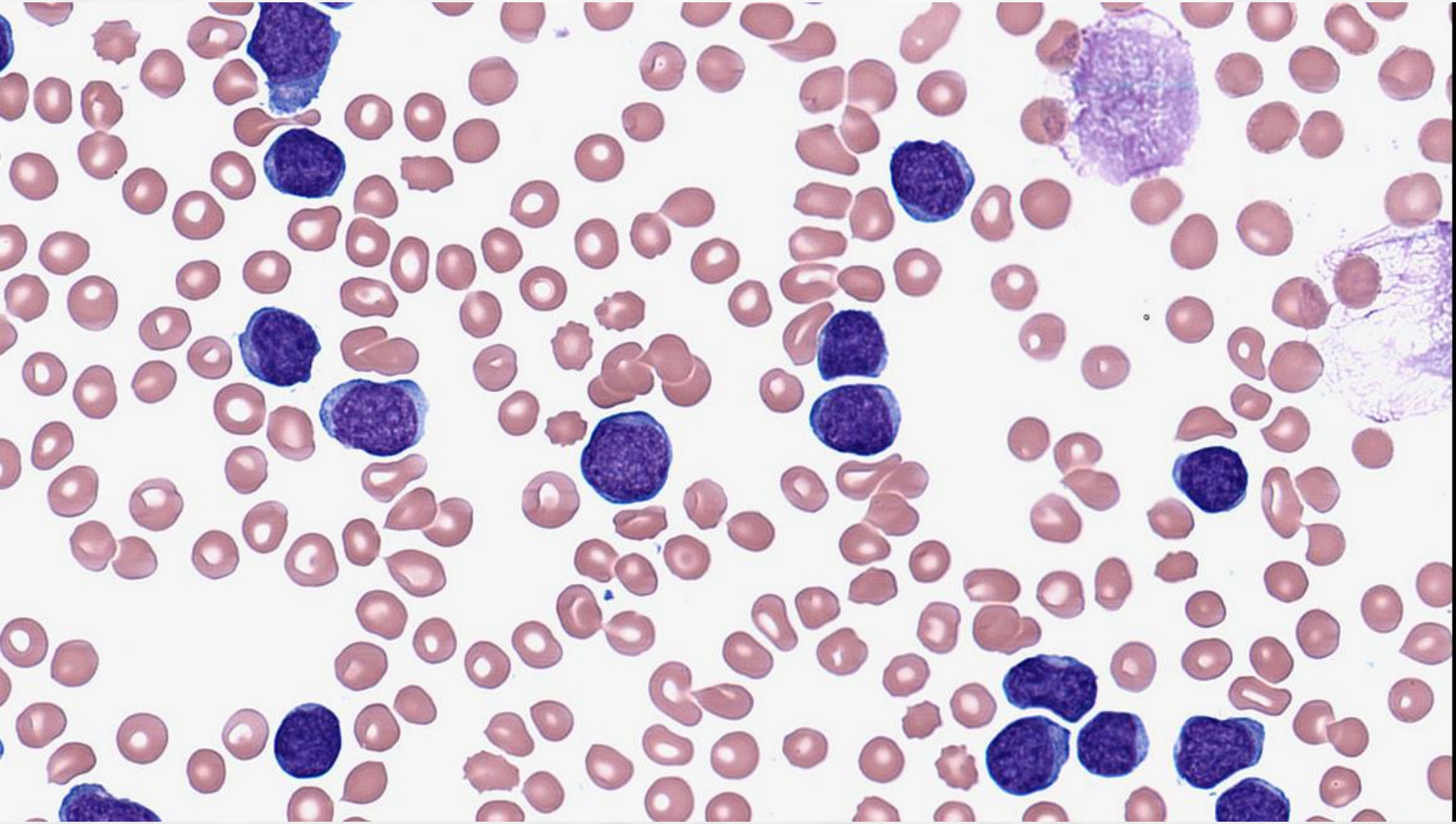
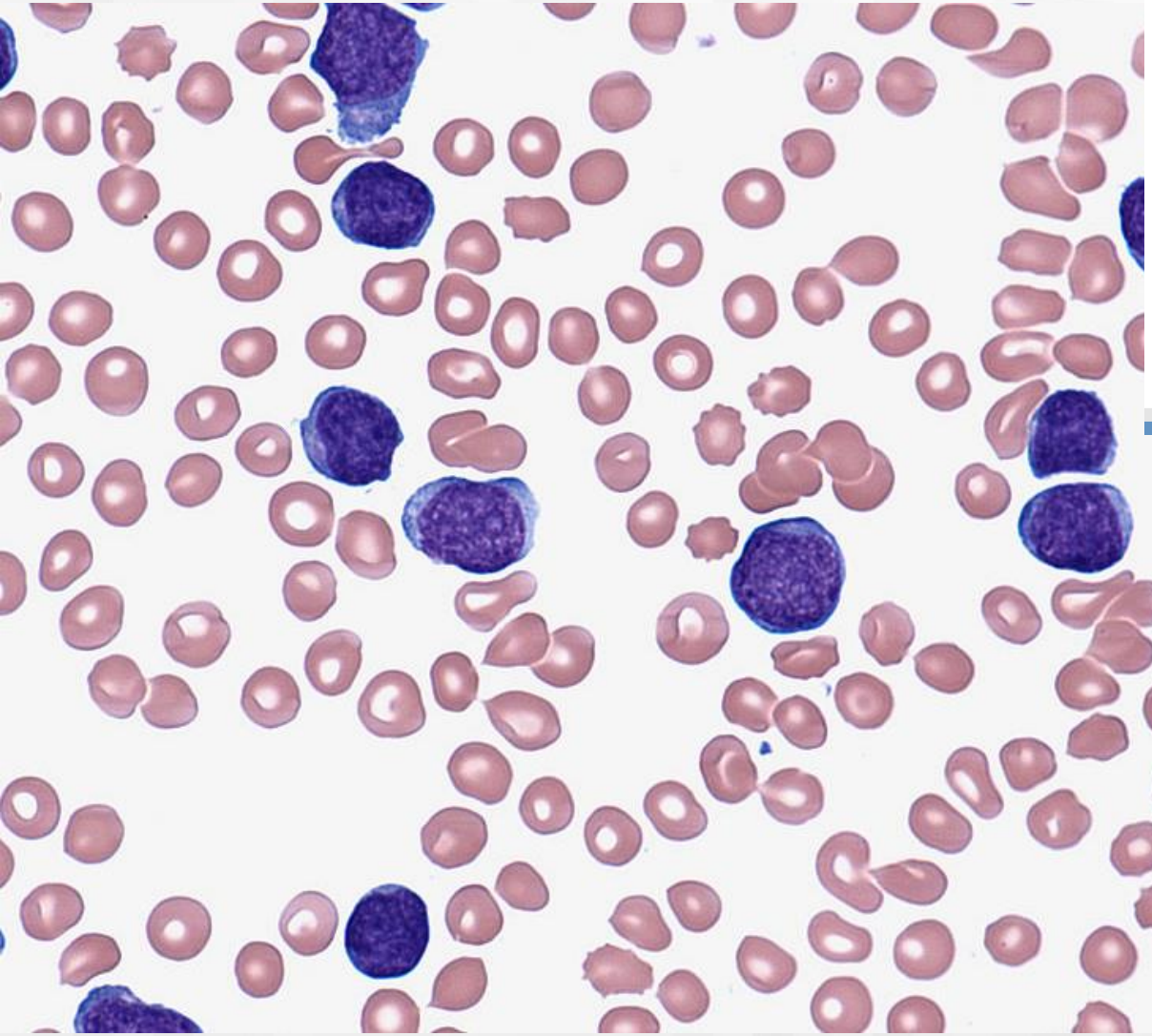
WBC Case 2





What is the most likely diagnosis?

Reactive lymphocytosis

Lymphocytic leukemia

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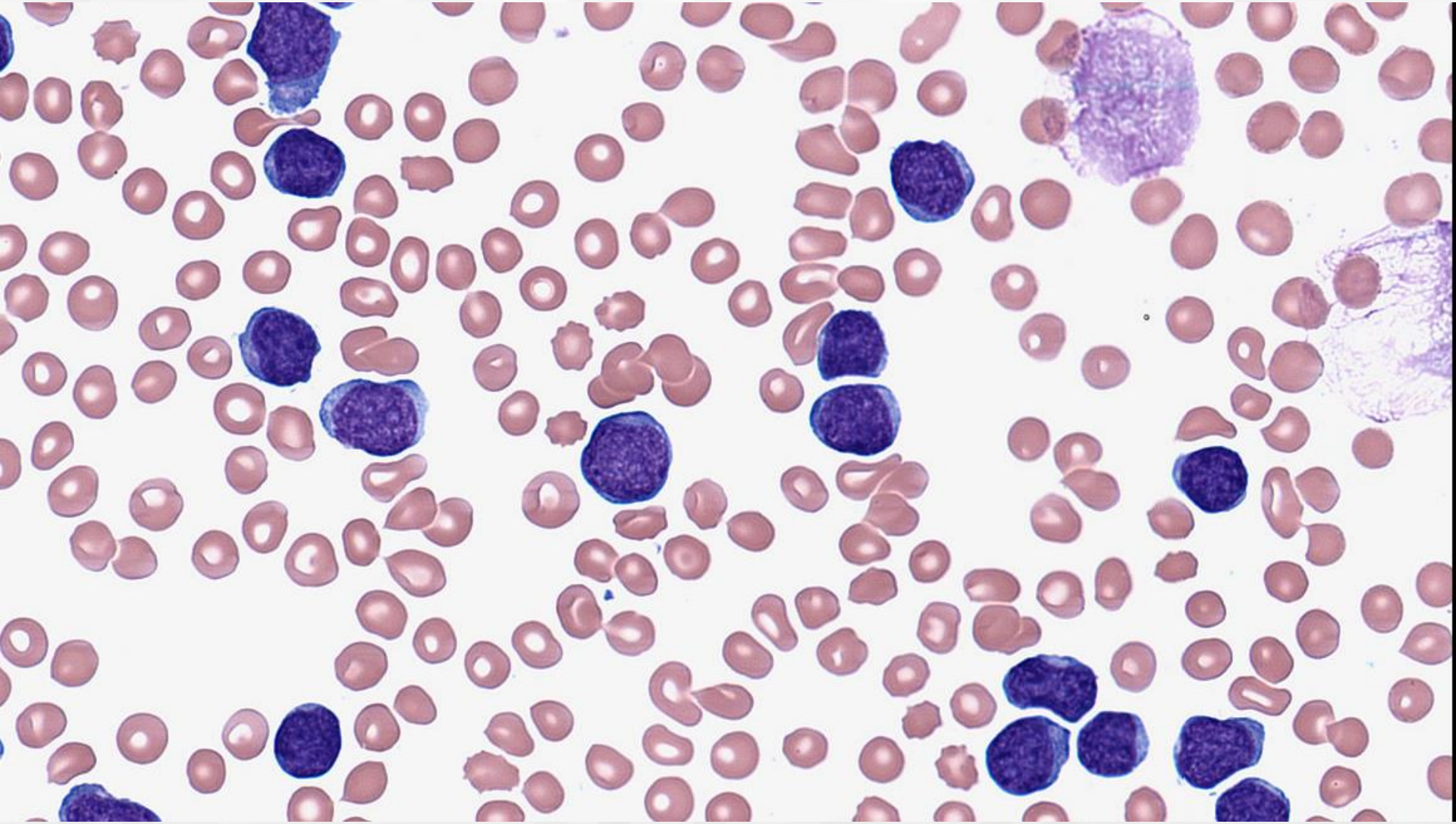
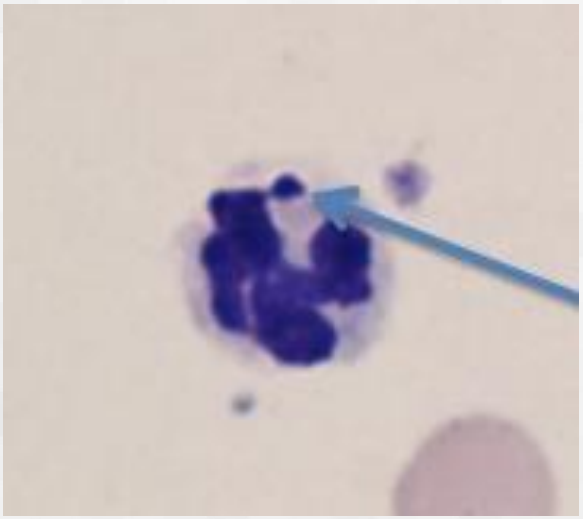
WBC Case 2



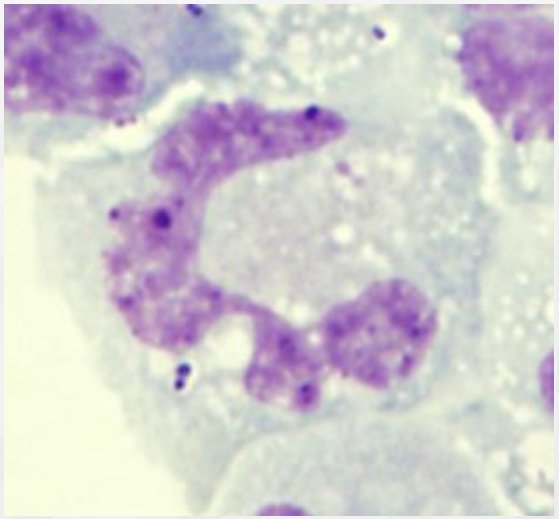
University of Saskatchewan

Inclusion Confusion

Barr Bodies

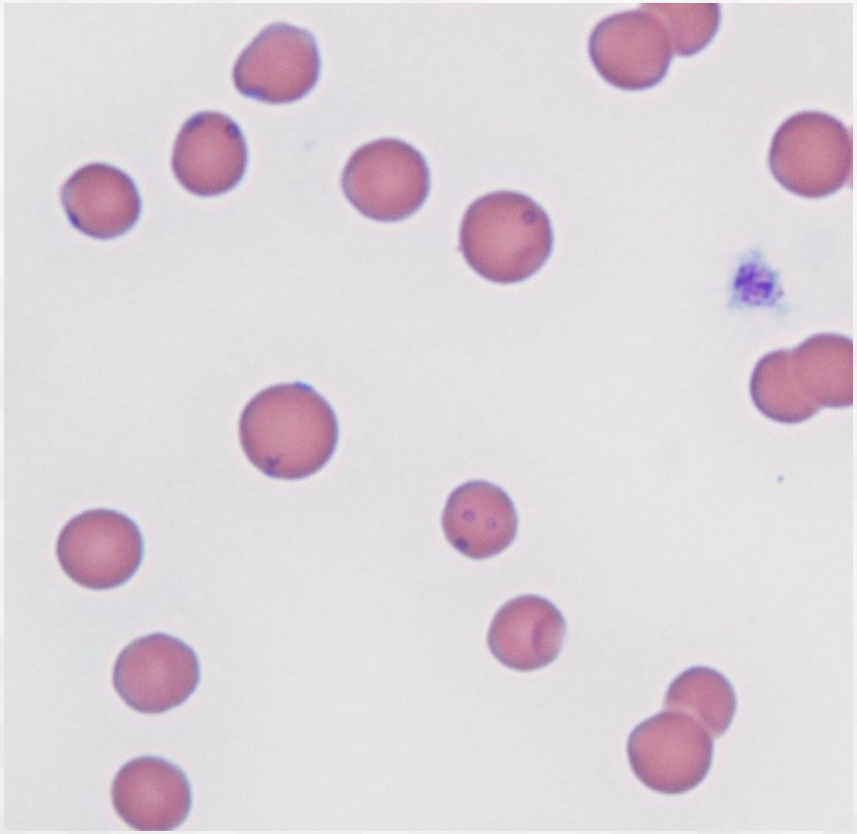


Bacteria

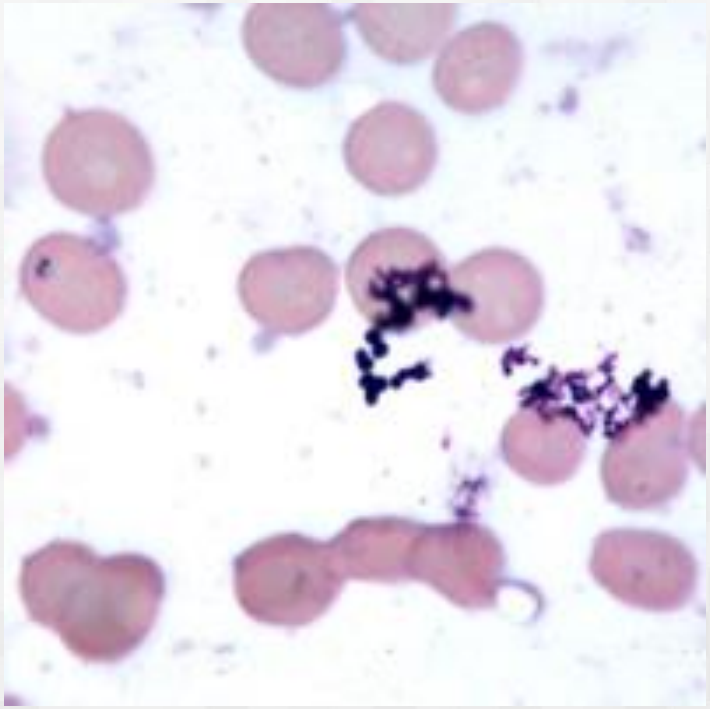


Inclusion Confusion

Mycoplasma hemofelis



Stain precipitate



http://www.eclinpath.com/hematology/sample-collection-heme/stain-precipitate/

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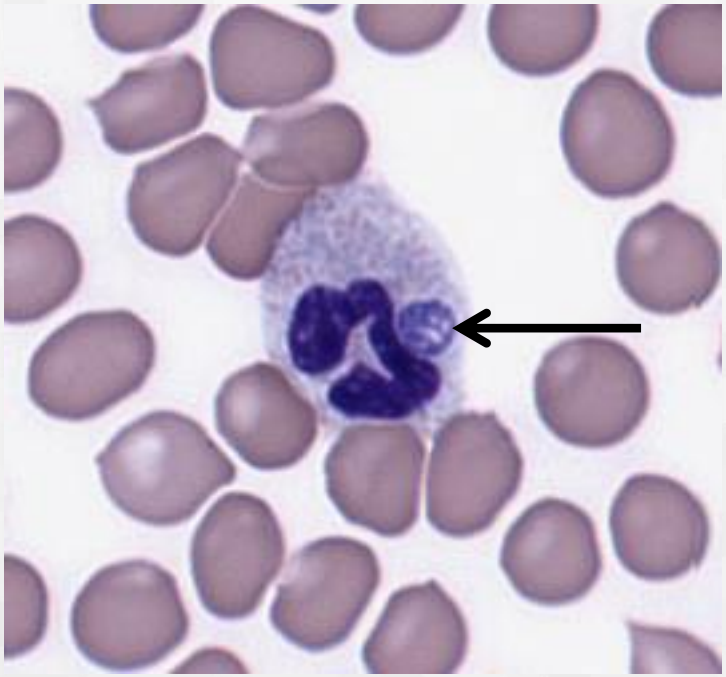
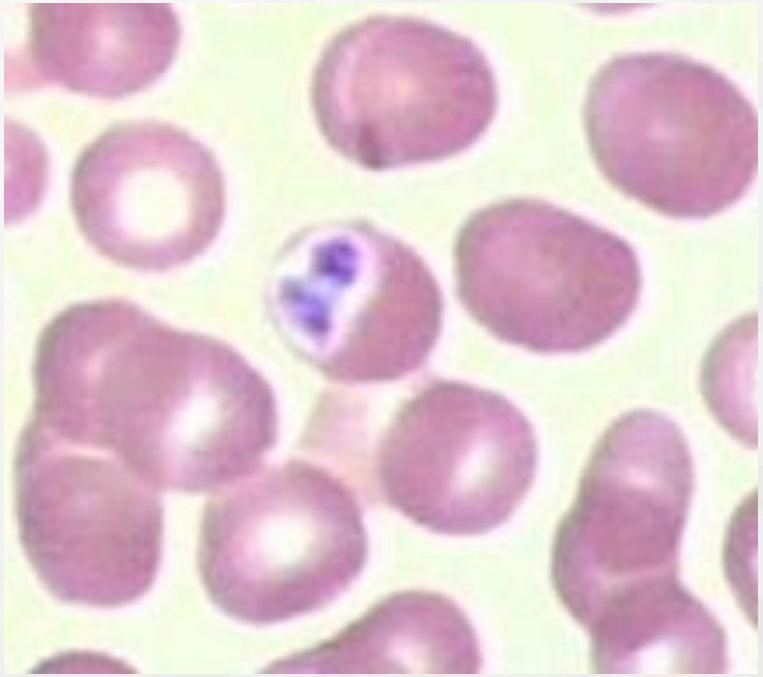
Inclusion Confusion

Anaplasma phagocytophilum



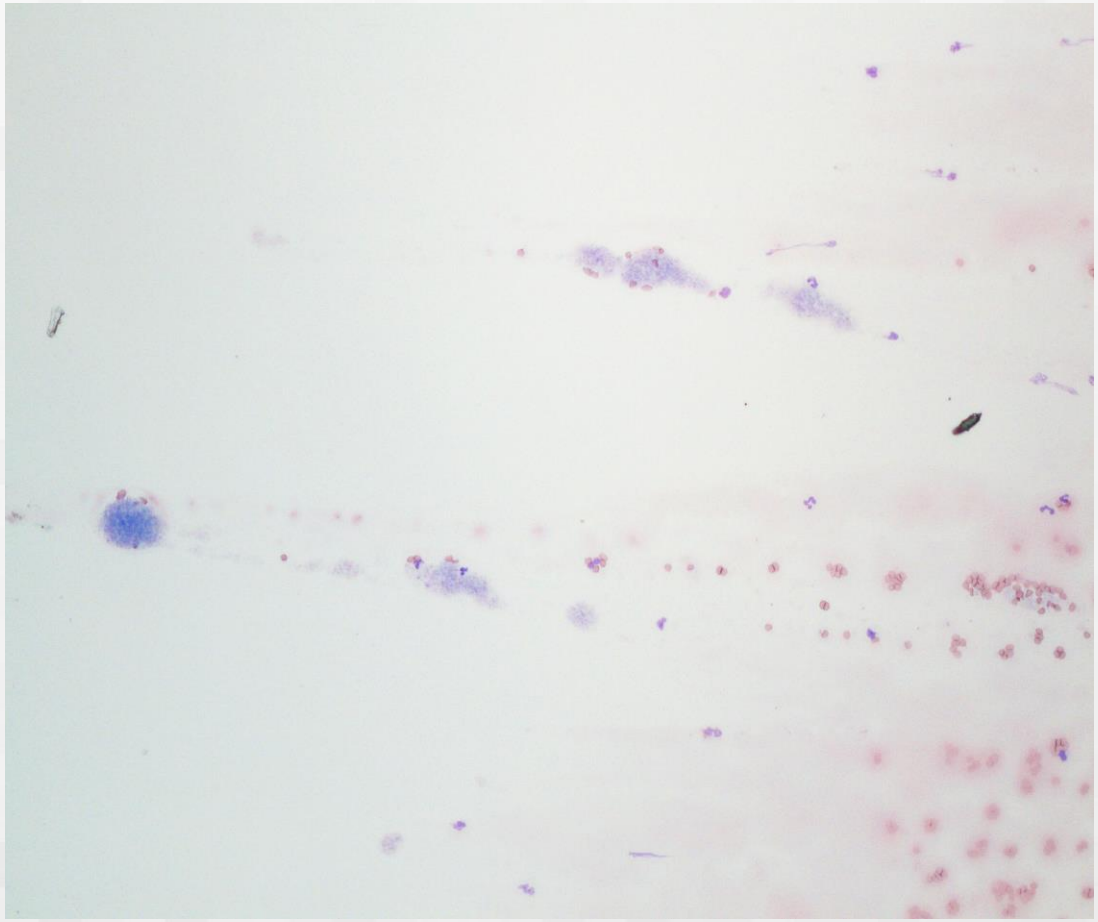
Overlapping platelet



Jacob EA. American Journal of Laboratory Medicine
Volume 1, Issue 3, November 2016, Pages: 34-57

Platelets

- Feathered edge
- 10x

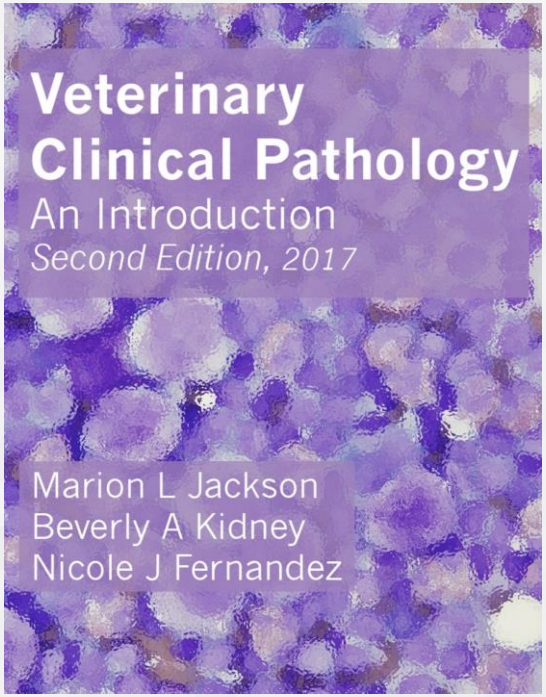


Platelet Estimate

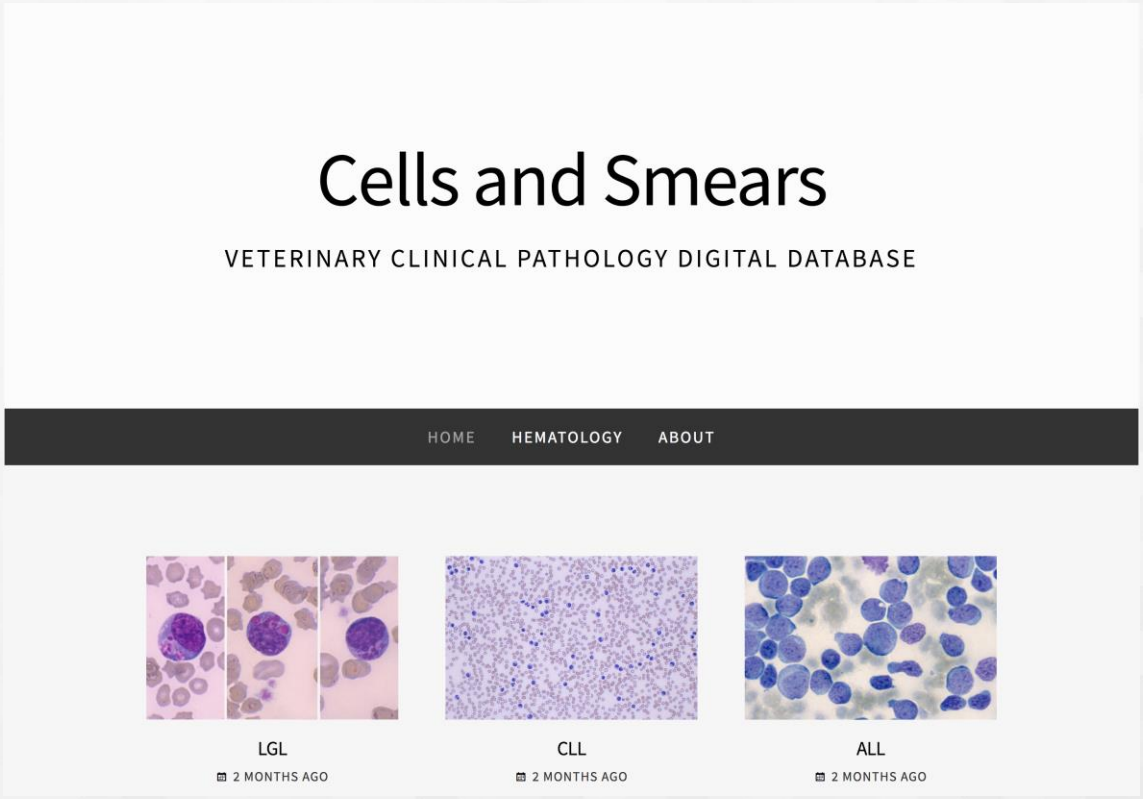
- 10x Scan the feathered edge for platelet clumps
- 100x Count the number of platelets in at least 5 fields of the monolayer and calculate the average for 1 field. Multiply the average for 1 field by 20 x 10^9/L.
- If platelets are clumped the formula will **underestimate** the platelet count.
- Less than 3 - 4 per 100x field (60 - 80 x 10^9/L) represents a significant thrombocytopenia

Digital Resources

- Veterinary Clinical Pathology ebook
  - iBooks
  - Google drive link
- Cornell e-ClinPath
  - http://www.eclinpath.com/



Digital Resources



- https://vetclinpathimages.com/

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Digital Resources

- PDS Protocol Manual
  - Search “pds protocol manual” for most recent version
- Videos
  - FNA & smear making: https://youtu.be/DkXDBom_3JI
  - Fine needle nonaspiration: https://youtu.be/jgTnqCWewI4
  - Impression smear making: https://youtu.be/prggfKrNlbI
  - Cytology fluid handling: https://youtu.be/Dl_Adsa7mpc
  - Cytology microscopy: https://youtu.be/vWSD7bjskjs
  - Urinalysis: https://www.youtube.com/playlist?list=PLm-jtvx5oGMTmnz0GJvPDzK0O1_AIFz5z

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Digital Resources

- Hematology Videos
  - Manual WBC count: https://youtu.be/WwHI1d5pqSc
  - PCV and total protein: https://youtu.be/cau5wWe4Uds
  - Making/staining blood smears: https://youtu.be/nbRUiWl2Qrs
  - Blood smear evaluation: https://youtu.be/wIZtvTGJL6M

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