Interactive Case Presentations: From New England to Beyond

Northeast Association for Clinical Microbiology and Infectious Disease (NACMID)

Portsmouth NH

September 12 & 13, 2022

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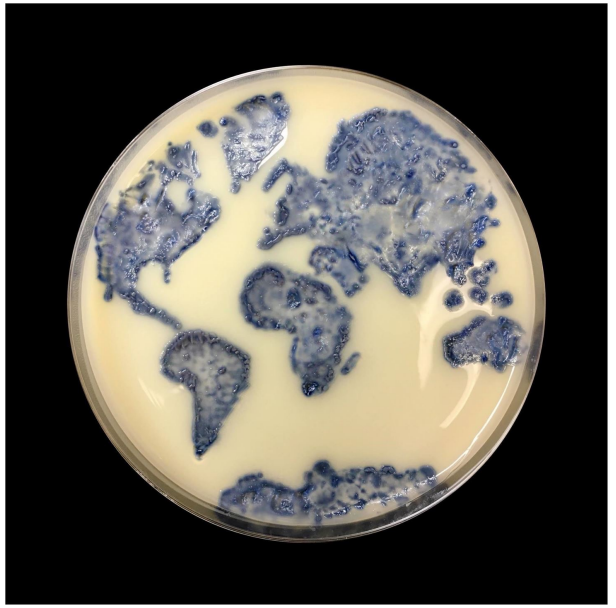


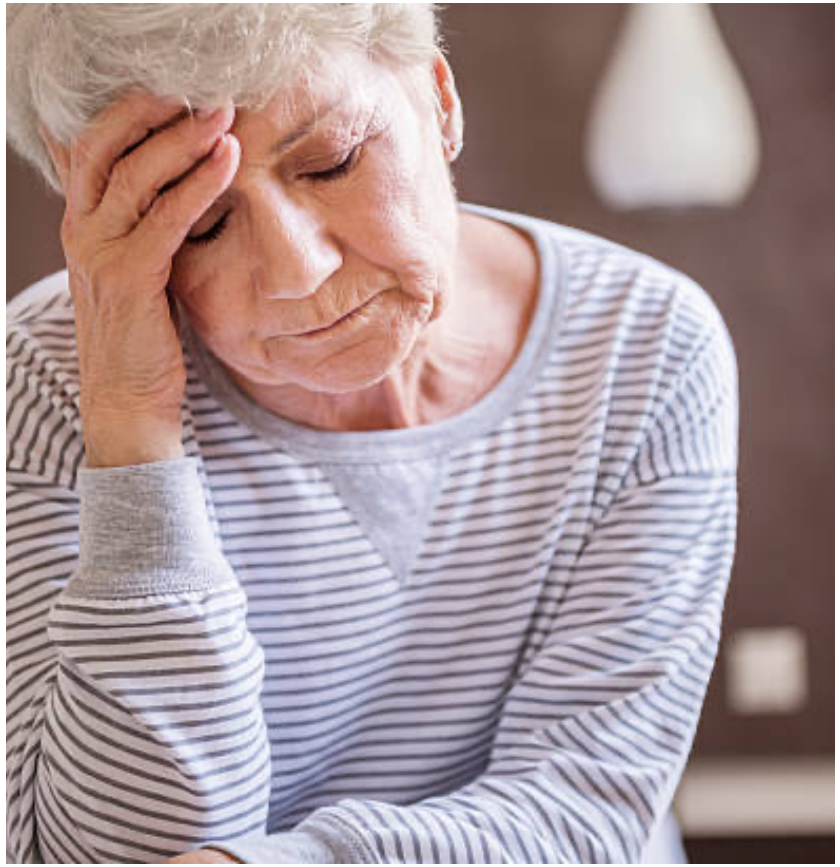
HARVARD MEDICAL SCHOOL

Mass General Brigham

The University of Vermont
LARNER COLLEGE OF MEDICINE

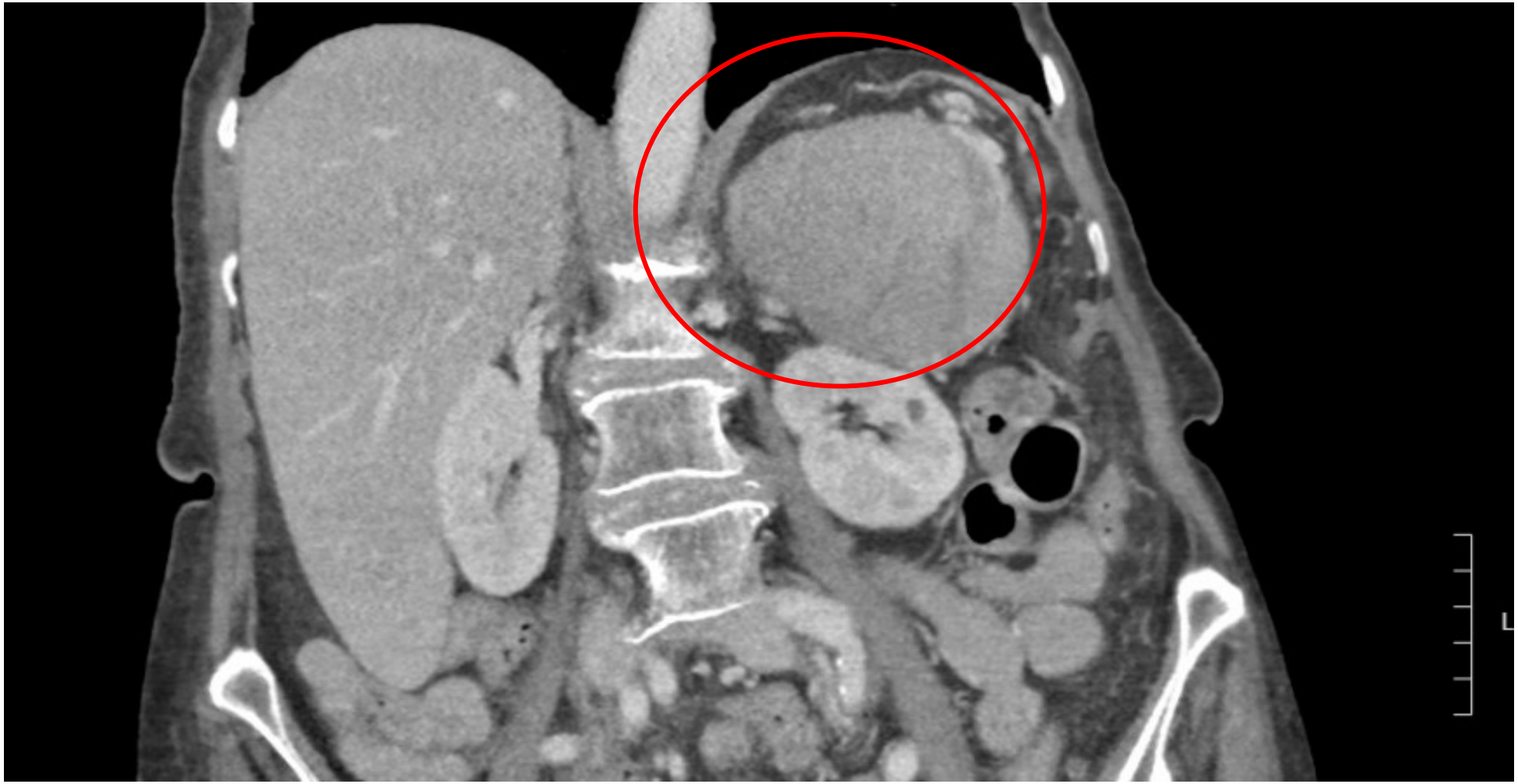
Case # 1

- 90 yo woman presents with 2 weeks of diarrhea and 2-3 days of fevers and chills, poor PO intake, worsening weakness and fatigue
- She takes care of her husband who has dementia and finds she can't meet his needs.
- No travel. Her son sometimes visits.
- Her PCP checks covid (neg) and sends basic labs which reveal significant Hgb drop
- Referred to ED:
  - Afebrile and clinically stable
  - CBC: elevated WBC 12.93, RBCs 3.36, Hgb 9.7 (2 pts lower than at last check), platelets 62.
  - BMP:
    - Creatinine 1.39 from baseline of 0.86
    - AST/ALT/bili 111/25/1.9 (nml last check)



Case # 1 CT A/P

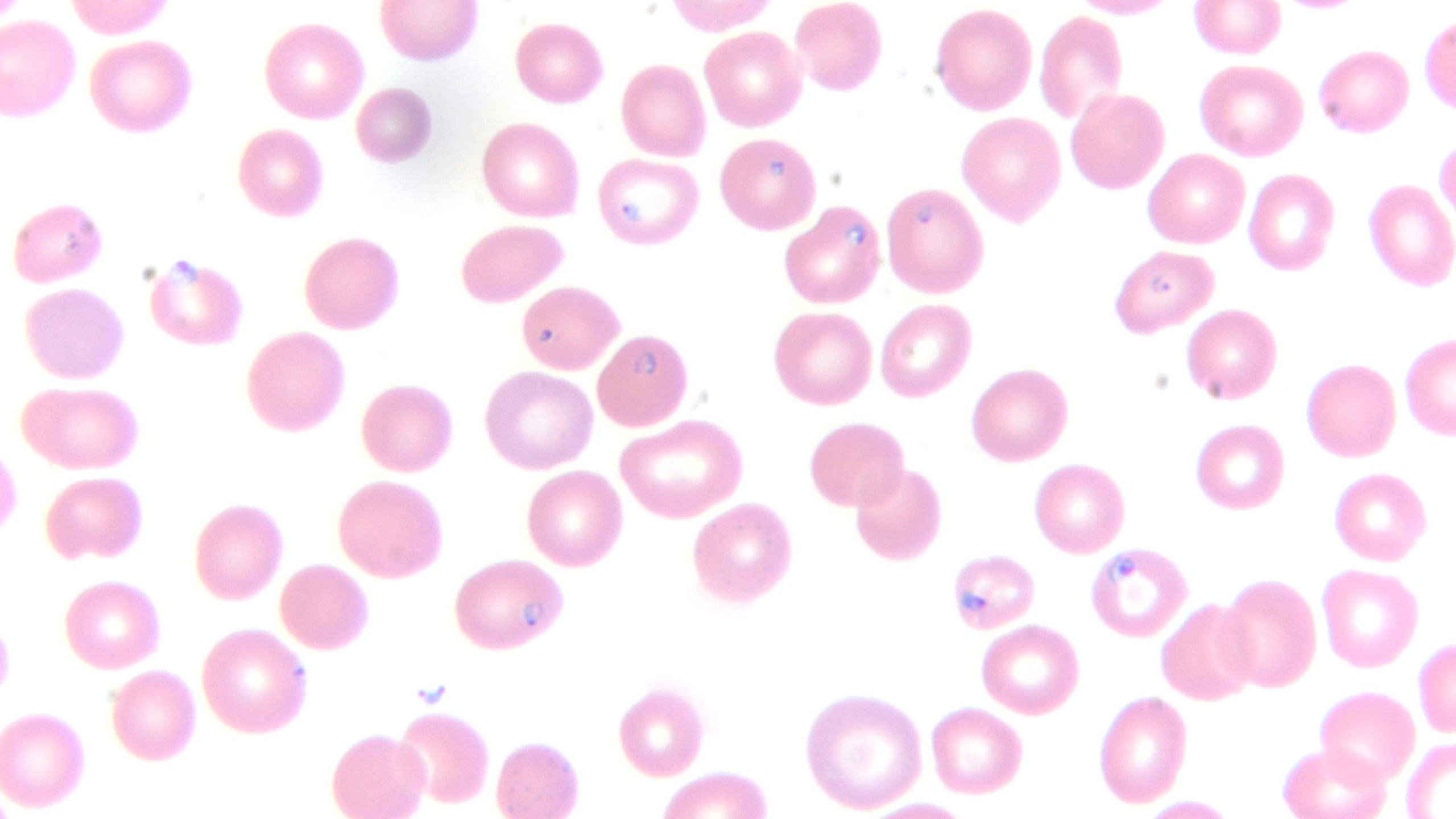
"Abnormal heterogeneous and globular appearance of the spleen. Findings raise concern for splenic rupture...No evidence of Active extravasation...No hemoperitoneum"

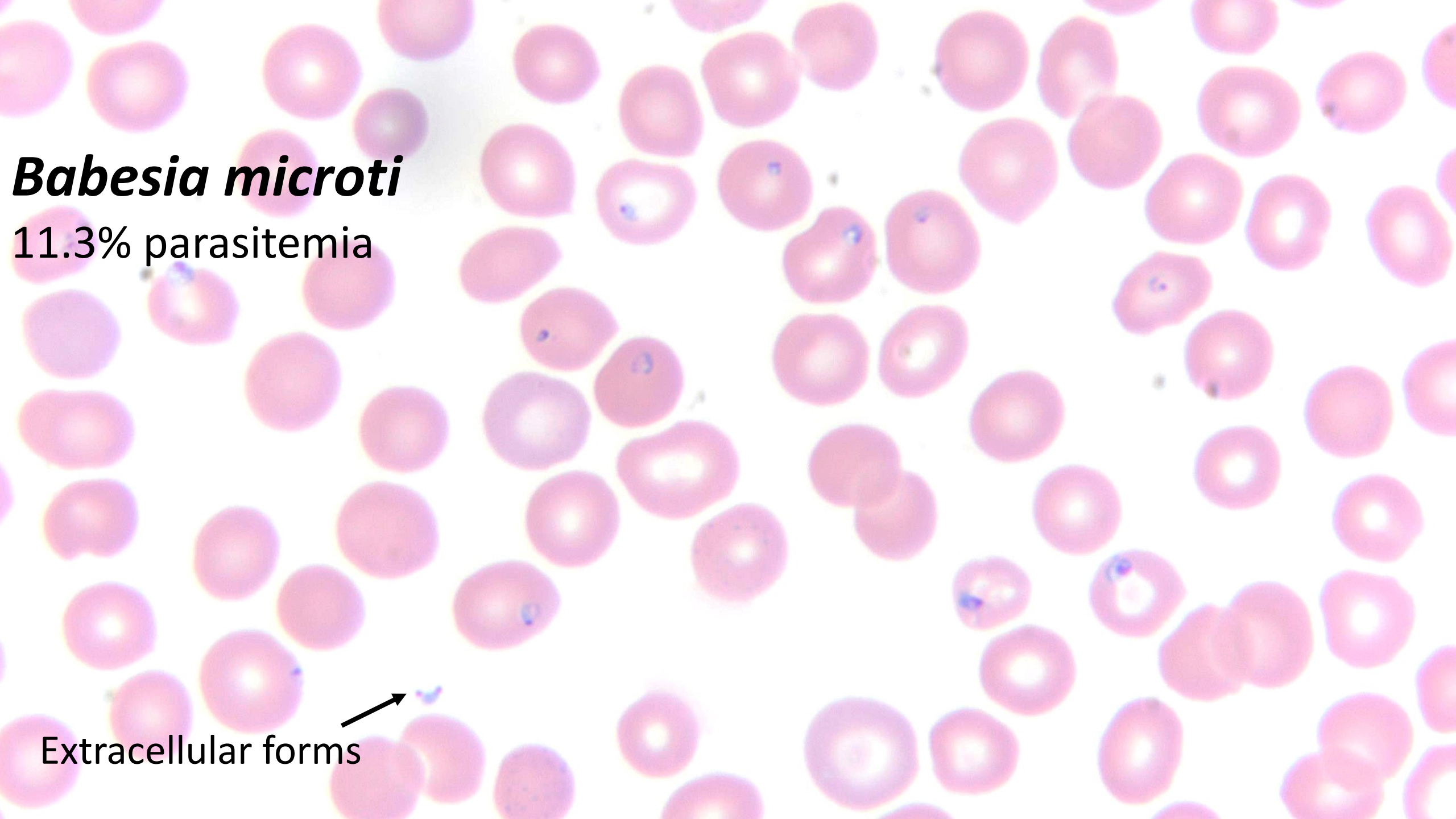


Case # 1: More Labs….

- Lyme Ab negative
- COVID negative
- Ehrlichia and Anaplasma PCR negative

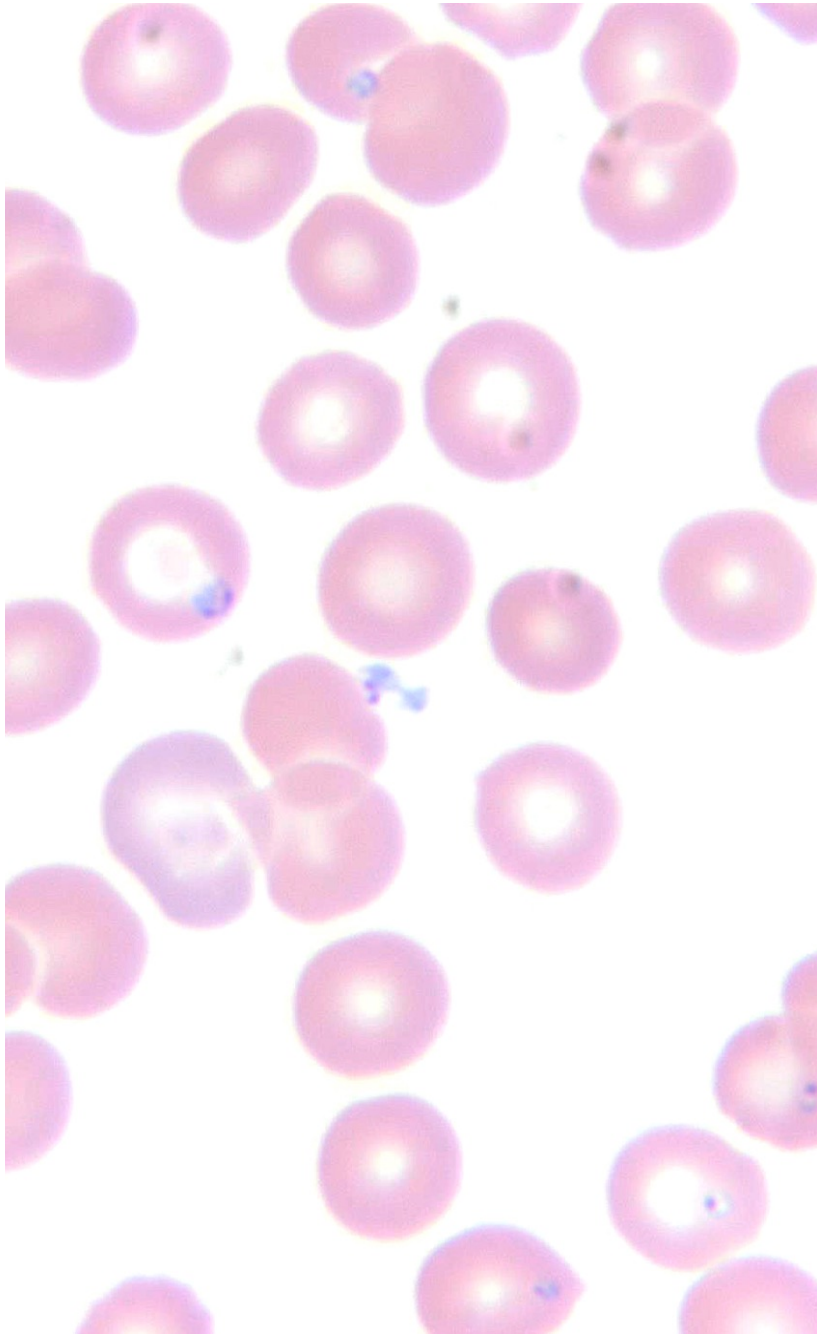
- Blood parasite sent……




Babesia microti
11.3% parasitemia
Extracellular forms

Hospital Course

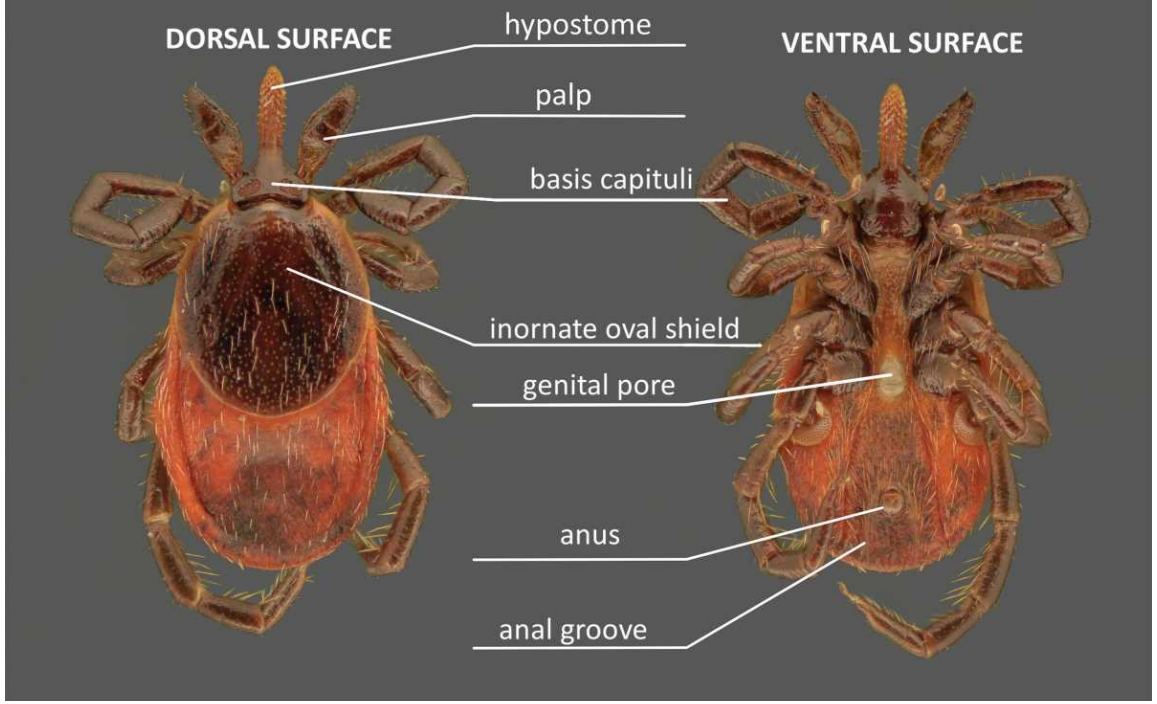
- Started on azithromycin and atovaquone
- Hematology advises no exchange transfusion
- Parasitemia monitored daily and 11.3-->9-->7.5 --> 0.5% HD 4
- Hgb nadir is 6.6 (hospital day 1)
- CXR hospital day 2 shows pulmonary edema
- Discharged HD 9 with no parasites seen on blood smear, Hgb 8



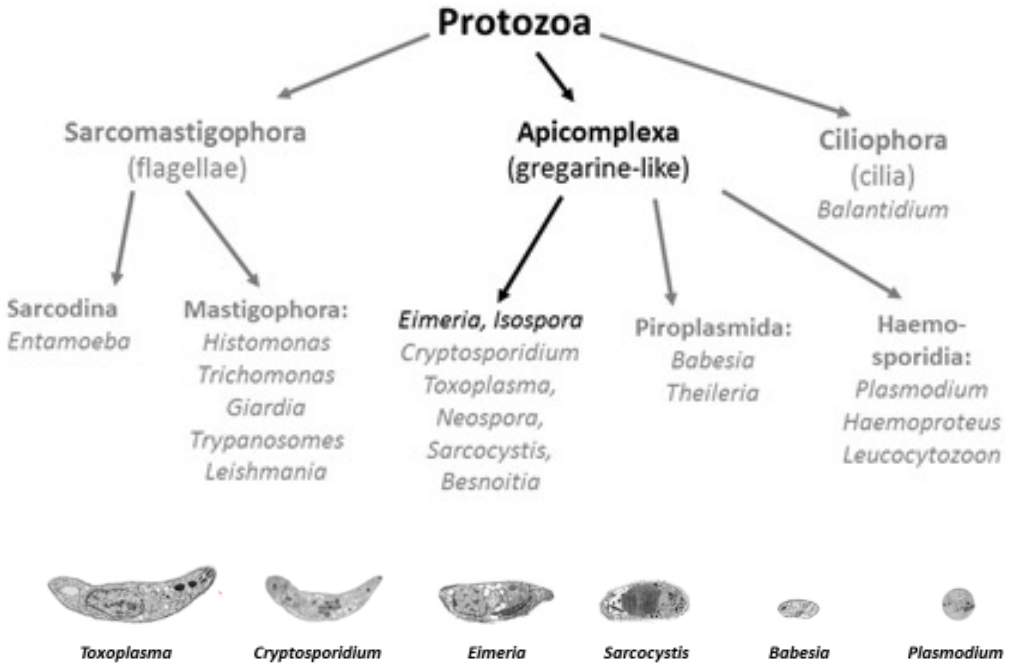
Ixodes scapularis

- *Borrelia burgdorferi, Borrelia mayonii, Borrelia miyamotoi*
- *Anaplasma phagocytophilum*
- *Babesia microti*
- deer tick virus (Powassan virus),
- *Ehrlichia chaffeensis*, *E. ewingii*, or *E. muris eauclairensis*

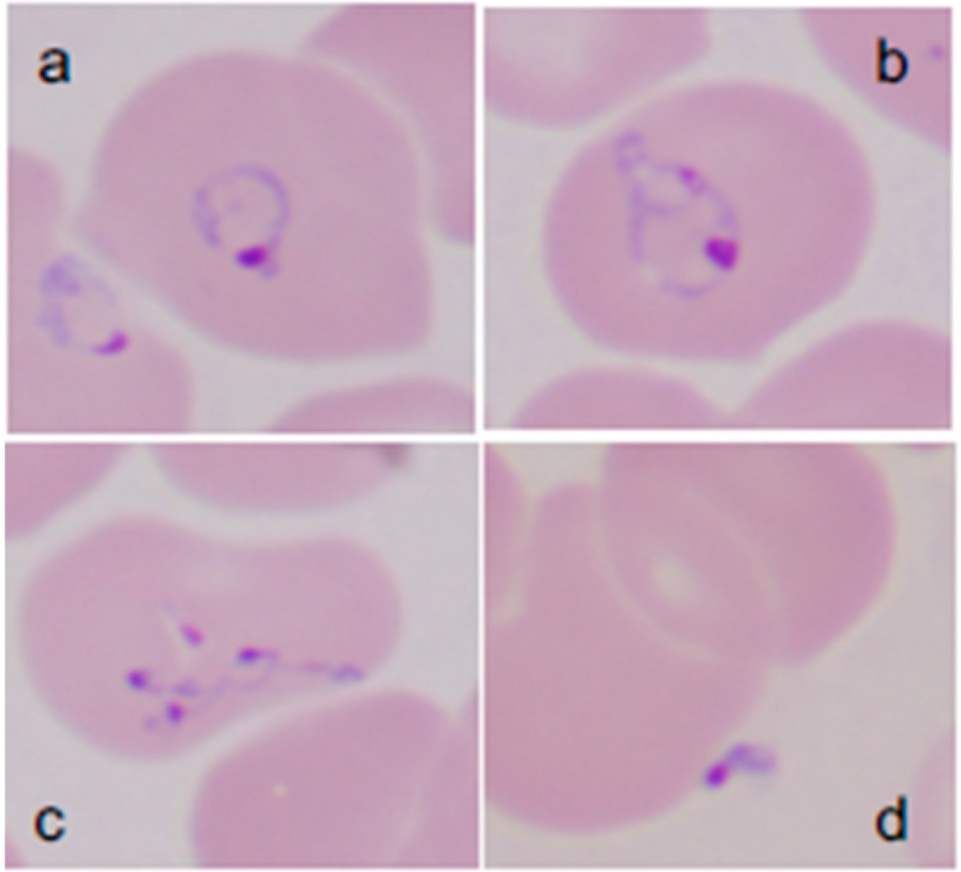
These pathogens differ in their geographic range within the Northeast and northern Midwest



Parasitic Protozoa- Classification



http://morrissettelab.bio.uci.edu/background%20information%20FINAL.html
http://ec.asm.org/cgi/content/full/3/2/483#F7

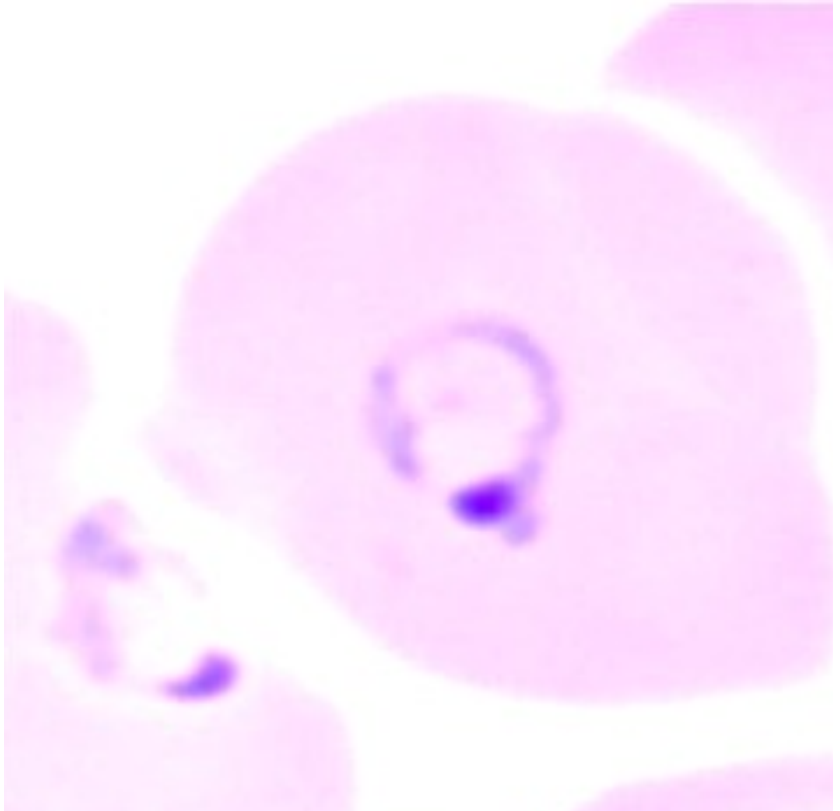


Giemsa-stained thin blood films showing *Babesia microti* parasites. *B. microti* are obligate parasites of erythrocytes. Trophozoites may appear as ring forms (**A**) or as ameboid forms (**B**). Merozoites can be arranged in tetrads and are pathognomonic (**C**). Extracellular parasites can be noted, particularly when the parasitemia level is high (**D**).

Block et al. Pathogens **2019**

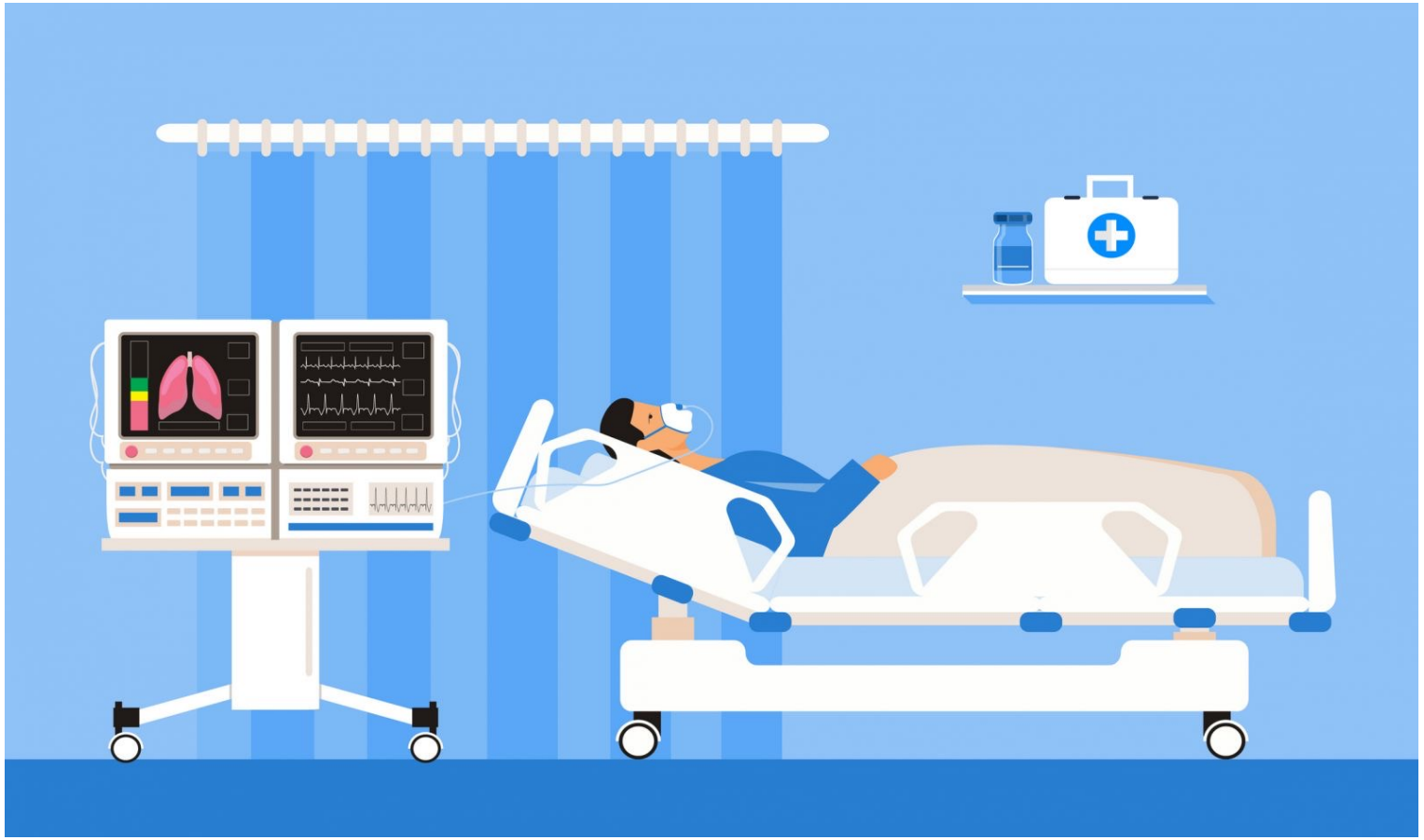
Babesiosis

- Illness ranges from subclinical illness to fulminating disease resulting in death
- Majority - mild illness or are asymptomatic (25% adults; 50% children)
- Symptoms within several weeks after exposure
  - Fevers + chills, myalgias, arthralgia, headache, sweats, extreme fatigue, anorexia and nausea
  - splenomegaly, hepatomegaly
  - hemolytic anemia (Jaundice, Dark urine), elevated renal function and liver enzyme levels, and thrombocytopenia
  - Severe: disseminated intravascular coagulation, hemodynamic instability, acute respiratory distress, renal failure, hepatic compromise, altered mental status, and death.
- Rare cases of chronic or reoccurring parasitemia can occur (immunocompromised or post splenectomy).



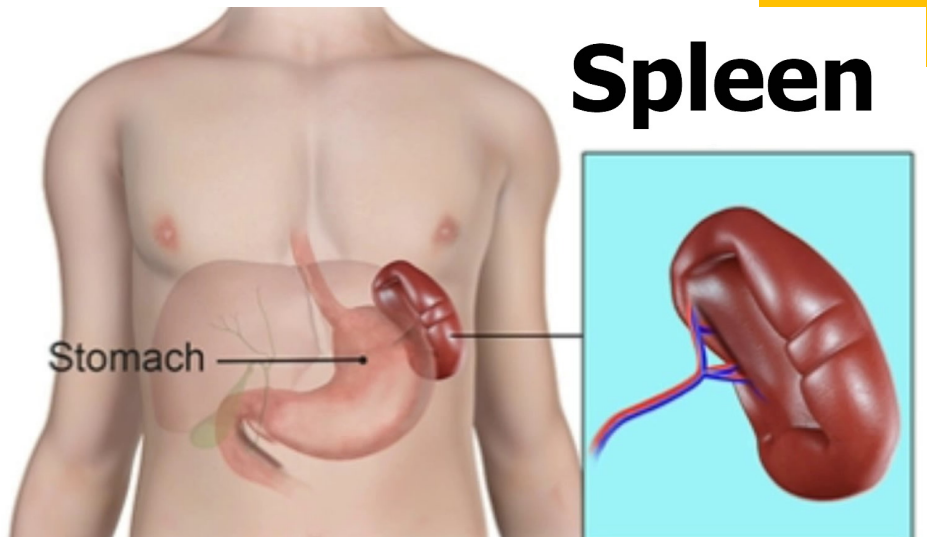
Adapted from Vannier and Krause, 2012. N Engl J Med 366, 239

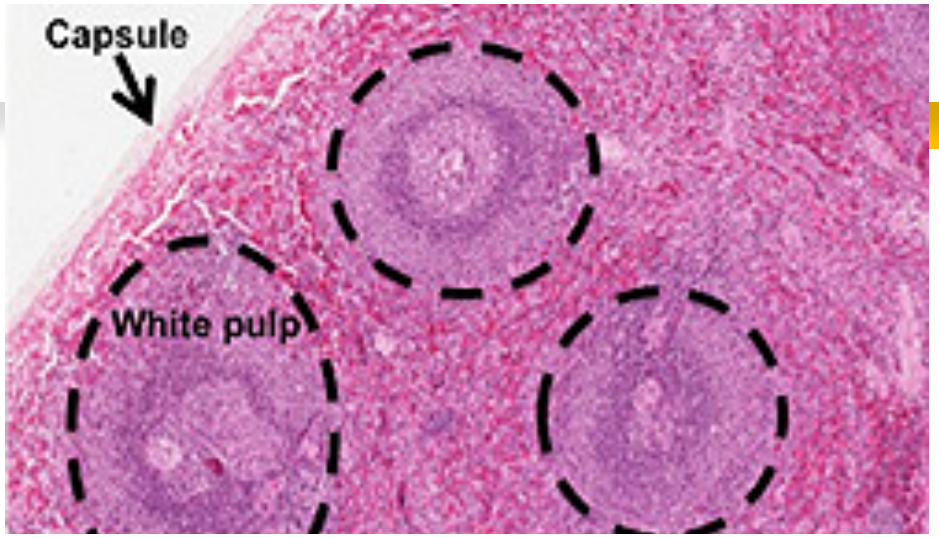
When/How do you decide to admit someone with a blood parasite?



Babesia - Vulnerable Populations

- Mortality may be as high as 5–9 % in hospitalized patients who are:
  - Asplenic individuals
  - >50 years of age
  - impaired immune function



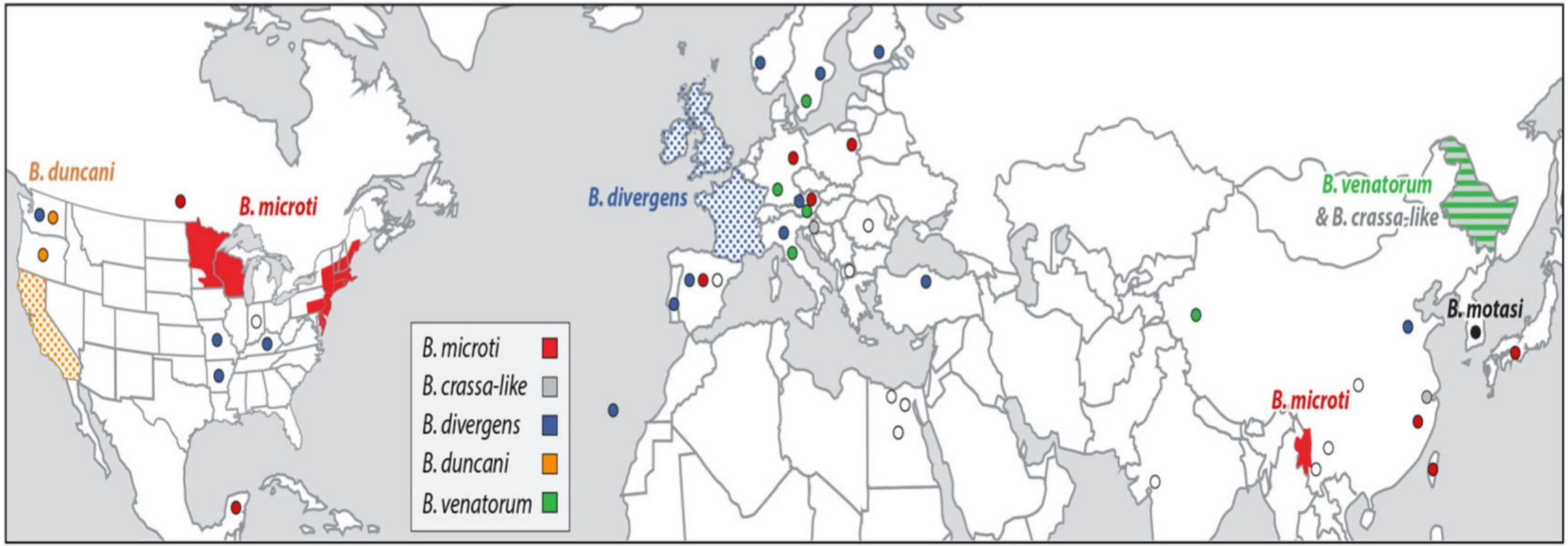


- Largest lymphoid organ in the body.
- Filters blood, allowing B and T cells to be activated in response to antigens in the blood
- Remove old/infected RBCs from the circulation

Babesia spp.

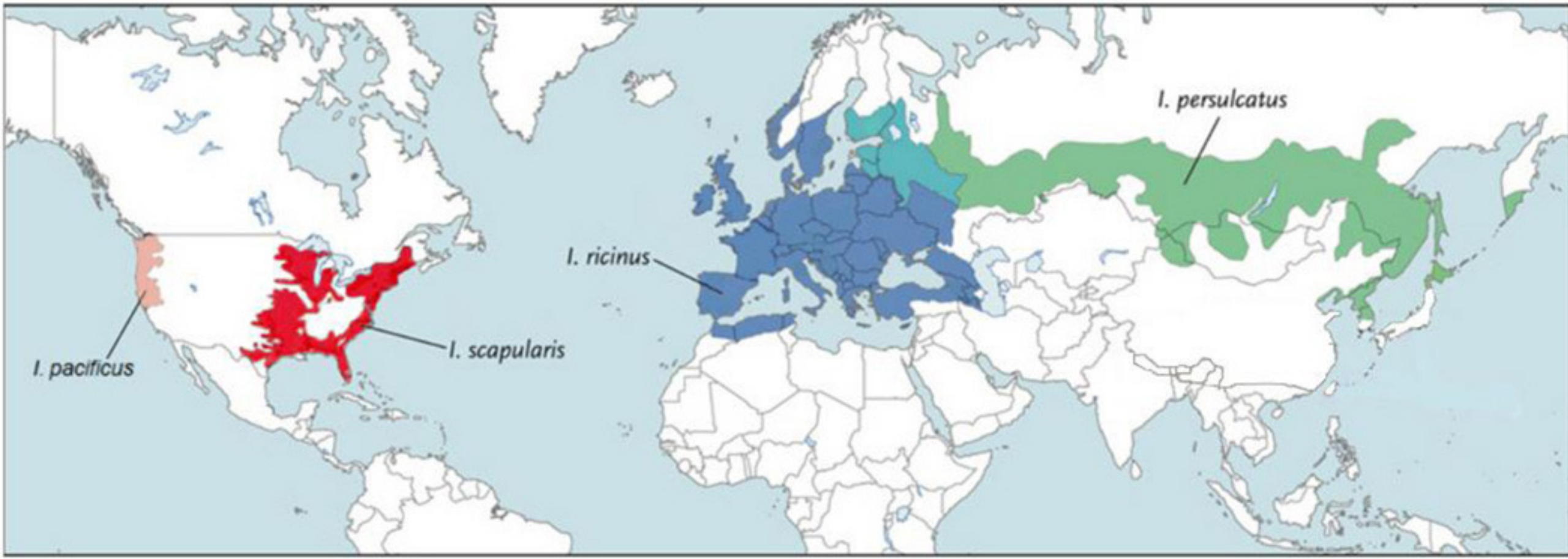
- 1888 - first described by Victor Babes in Romanian cattle
- 1957 - first human case of babesiosis (Yugoslavia)
- 1968 - second case (California)
- 1969 - third case - identified as *B. microti.* (Nantucket Island)
- > 100 Babesia species infect a wide spectrum of wild and domestic animals worldwide; six identified as human pathogens
  - *Babesia microti* -**endemic in US & China.**
  - *B duncani* - Pacific Coast
  - *B. venatorum – China, Europe*
  - *B. crassa-like- China*
  - *B.crassa*s-heep in Iran and Turkey
  - *B divergens* - Europe, reported in US

Babesia: Global Distribution



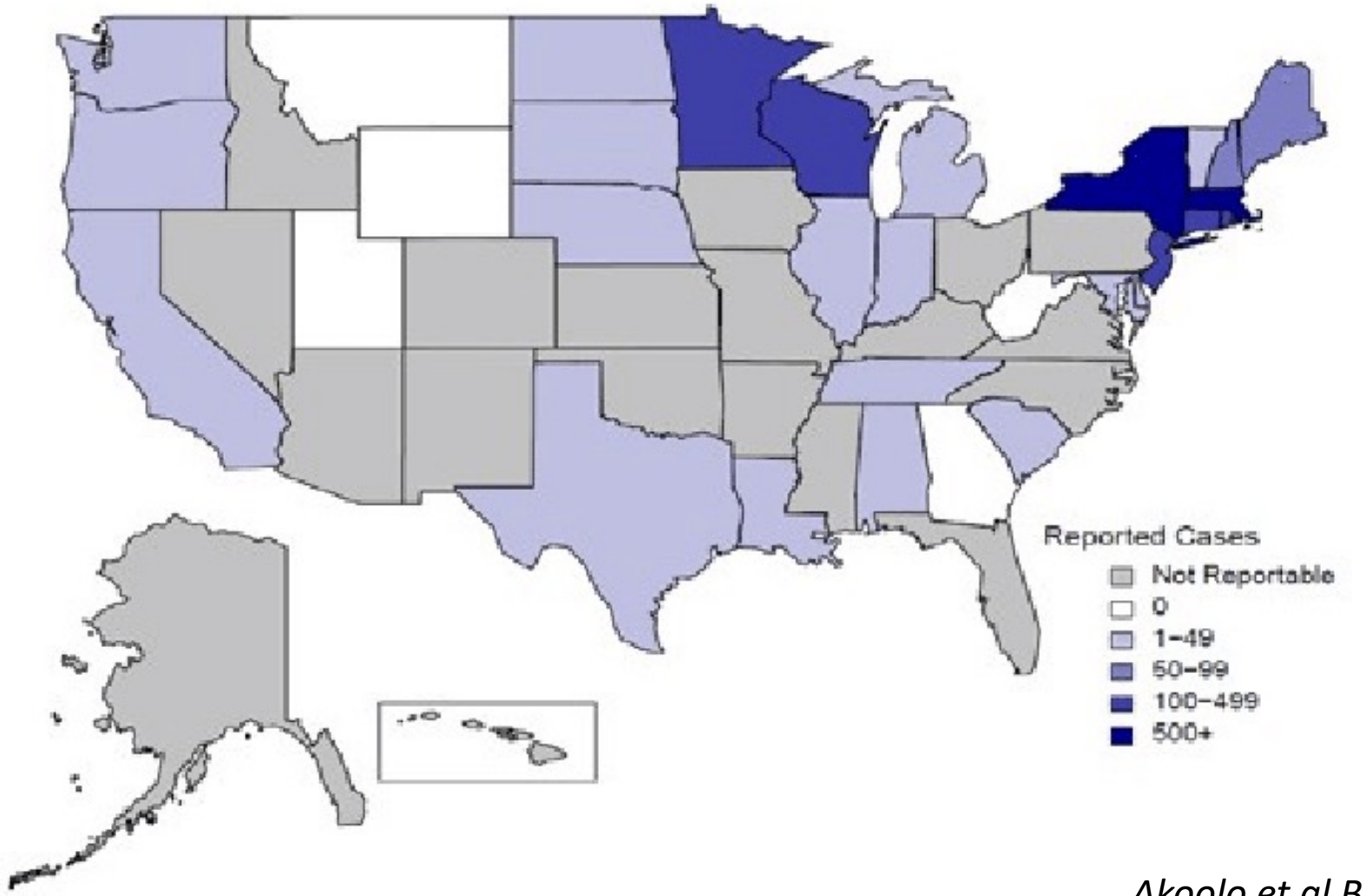
Diuk-Wasser MA, et al. Trends Parasitol. 2016

Global Tick Vector Distribution



Diuk-Wasser MA, et al. Trends Parasitol. 2016

Babesia: US Distribution

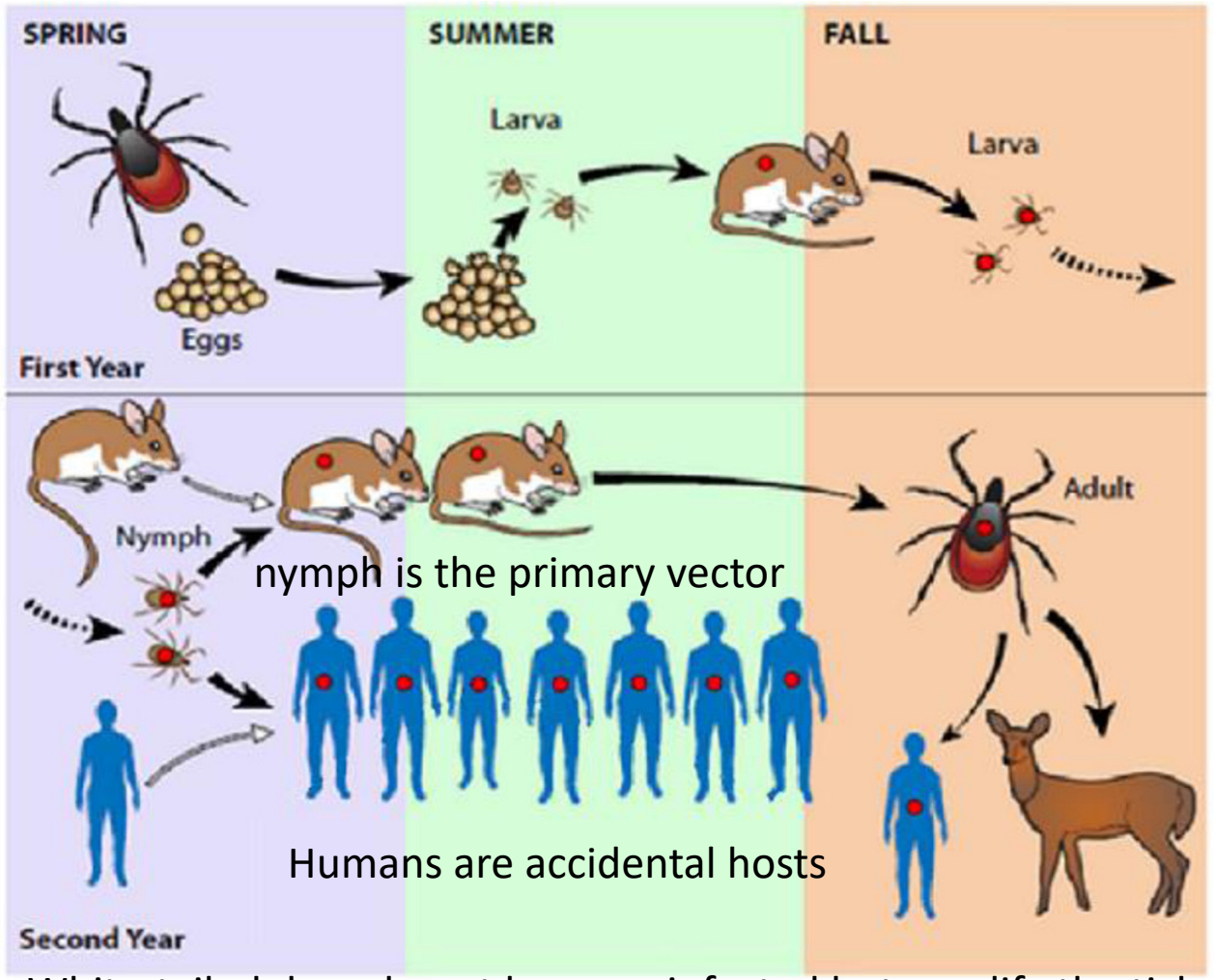


Akoolo et al BMC Microbiology 2017

Babesia Transmission

- **Tick Vectors** –
  - *Ixodes scapularis, Ixodes pacificus (US)*
  - *Ixodes ricinus* and others (Europe & Asia)
  - Incubation: 1-4 wks
- **Blood transfusion**
  - Incubation: 1-9 wks (up to 24 eks)
- **Organ transplantation**
- **Congenital –** Vertically transmitted across the placenta from mother to fetus

White-footed mice are the primary reservoir host



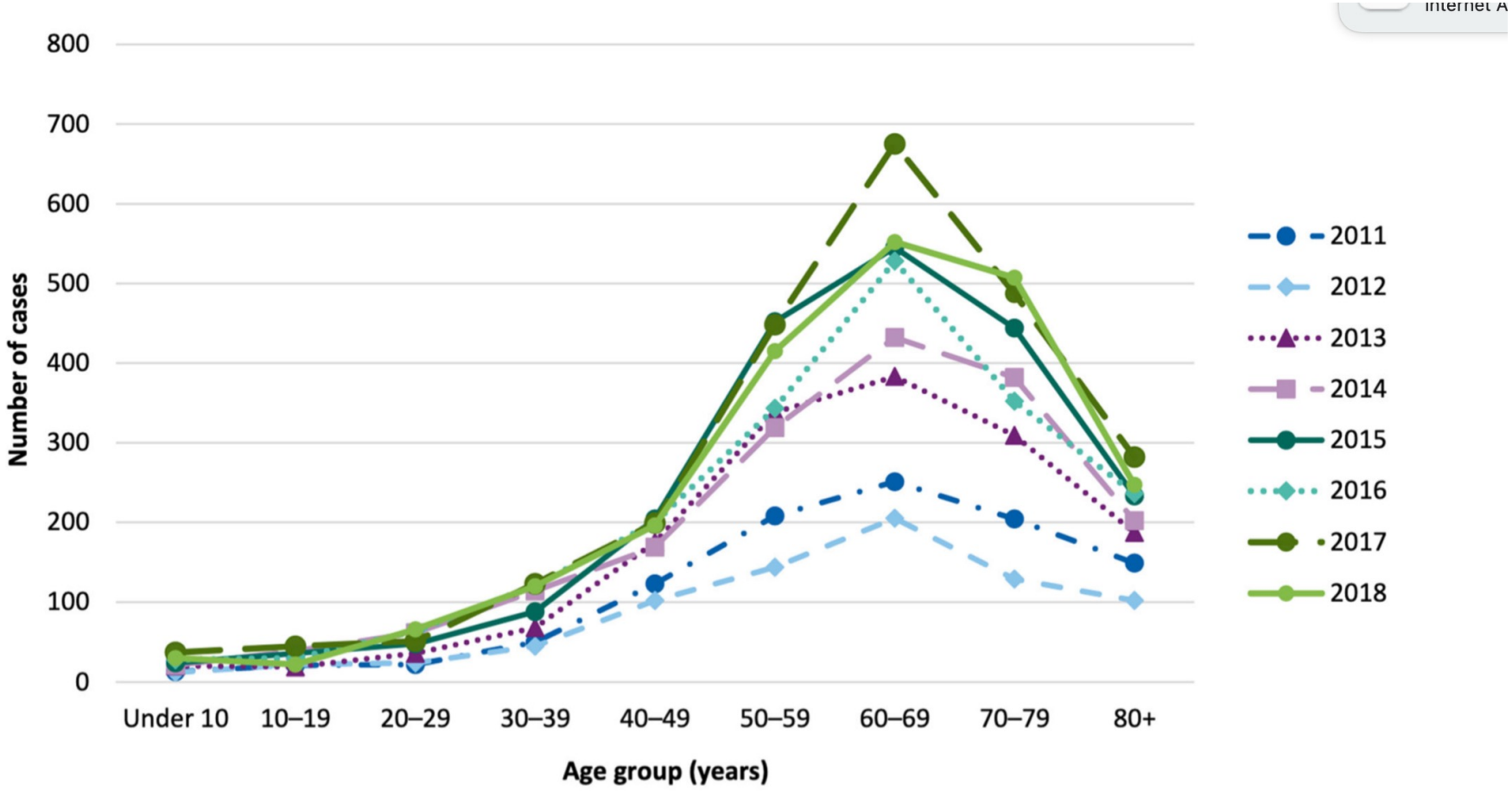
White-tailed deer do not become infected but amplify the tick population by providing a blood meal for adult ticks

Babesia as an Emerging Disease

Late 19th : US White Tailed Deer population: 300,000

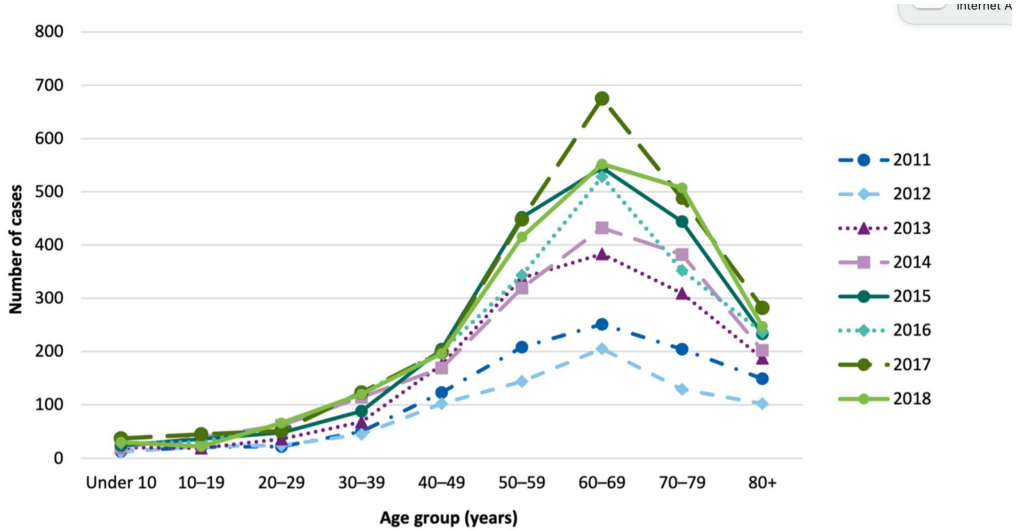
=> 3 million (2012)

Babesia Incidence Overtime by Age Group



Kumar et al Pathogens 2021

Babesia Incidence Overtime by Age Group



Kumar et al Pathogens 2021

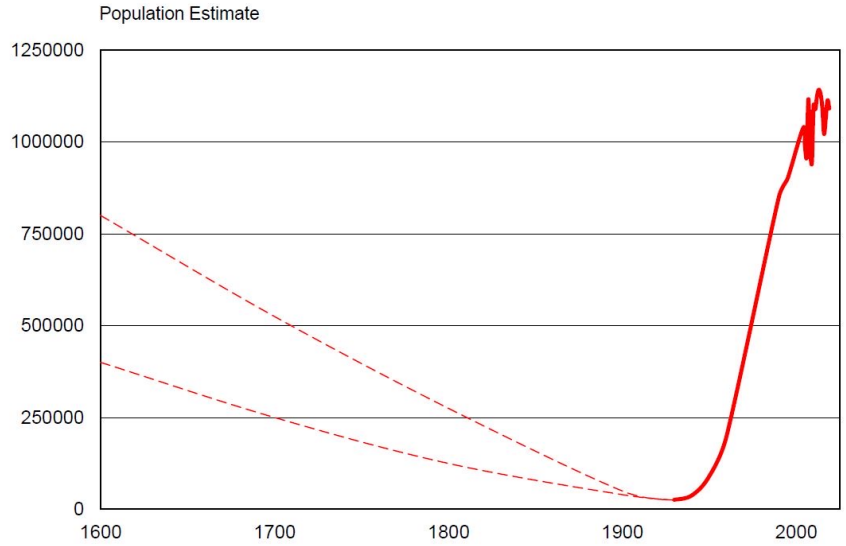


Figure 2. Hypothetical population curve for Virginia's deer herd, 1600-present.

White Tail Deer Population

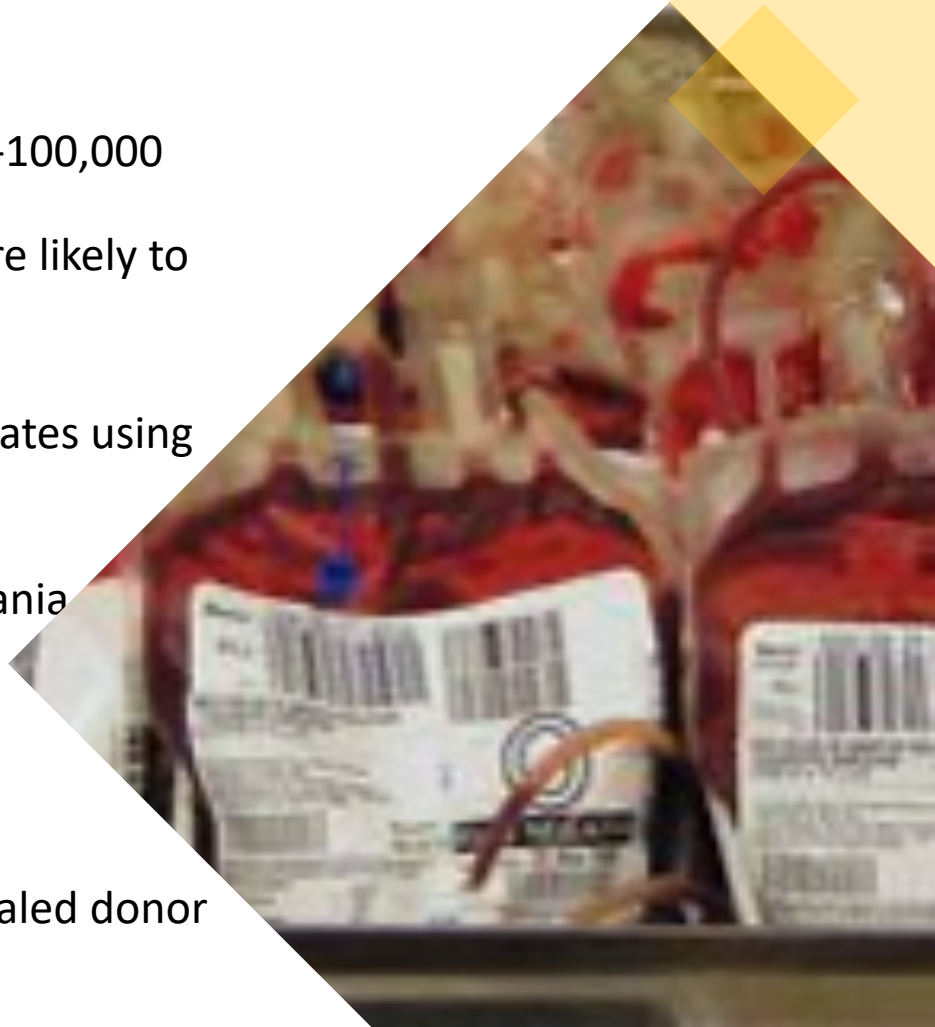
Arlington Regional Master Naturalists

Babesia Screening and the Blood Supply

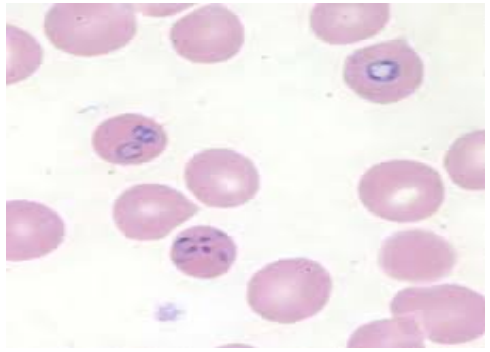
- Leading causes of transfusion transmitted infection in the United States
  - 250 cases reported (~ 1/5th fatal)
  - Frequency 1 per 250 donations using both antibody and NAT (0.4%)
  - Estimated risk of transfusion-transmitted babesiosis = 1 per 18,000 -100,000 unscreened donations in an (highly) endemic regions
  - Unscreened blood collected in highly endemic areas is 16 times more likely to be infectious than screened blood

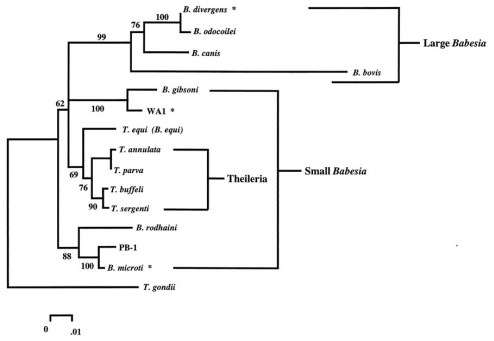
- 2020: US FDA recommended donor screening in 14 *B. microti* endemic states using approved PCR technologies.
  - Maine, Vermont, New Hampshire, Connecticut, Massachusetts,
  - New York, New Jersey, Delaware, Rhode Island, Maryland, Pennsylvania
  - Virginia, Minnesota, and Wisconsin and in the District of Columbia
  - mini-pool testing can be performed (MPs of 16)

- China – screening not yet available.
  - 2016 pilot serosurvey of blood donors in Heilongjiang Province revealed donor prevalence of **1.3%**



How much do you take specific occupation/exposure into account?





Diagnosis of Babesiosis

- **Animal Sub-inoculation** - Identification of the organism following the injection of patient blood into laboratory animals

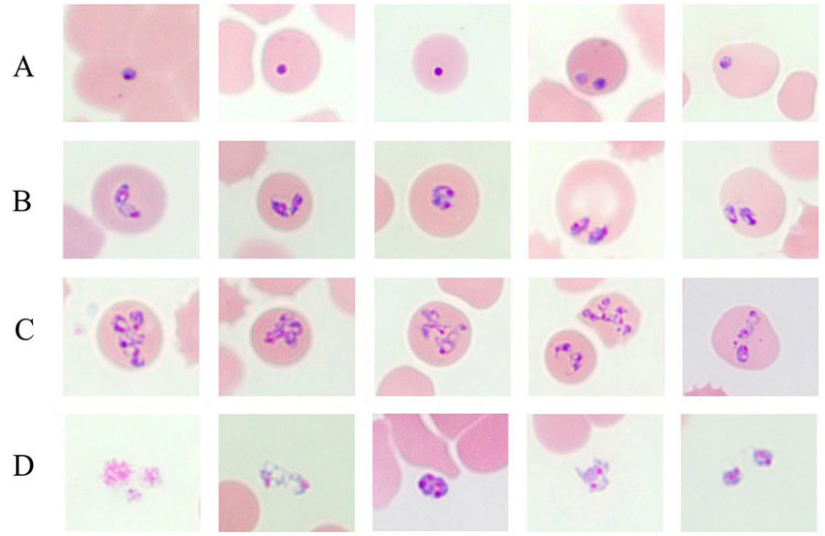
- **Serology** - babesial antibody in acute and convalescent-phase sera

- **Microscopic** identification of the organism by Giemsa or Wright staining of thick or thin blood smears
  - Multiple thick and thin blood smears should be examined

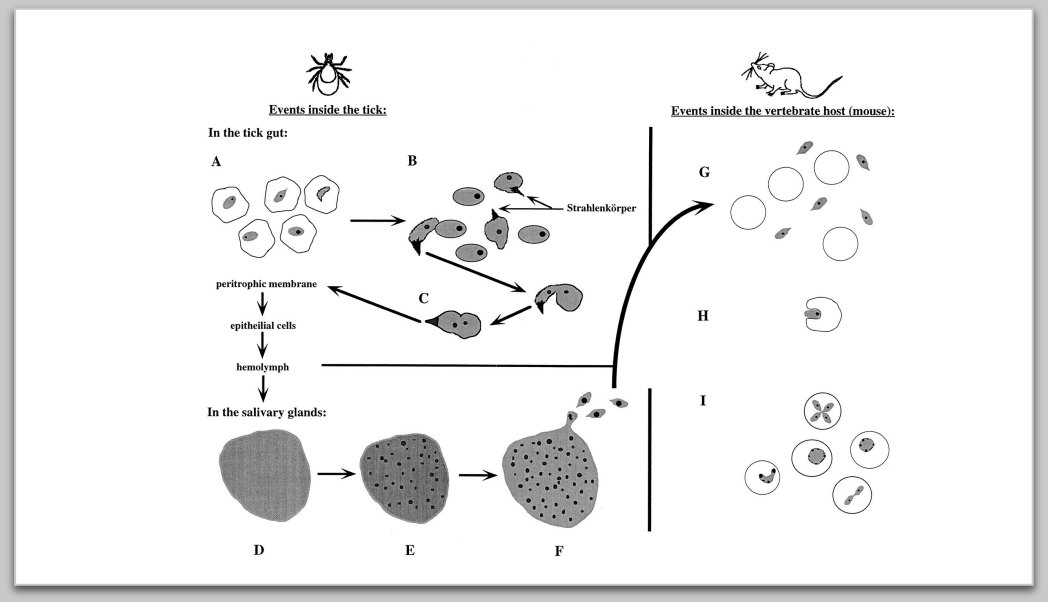
- **Molecular** identification of organism in blood sample

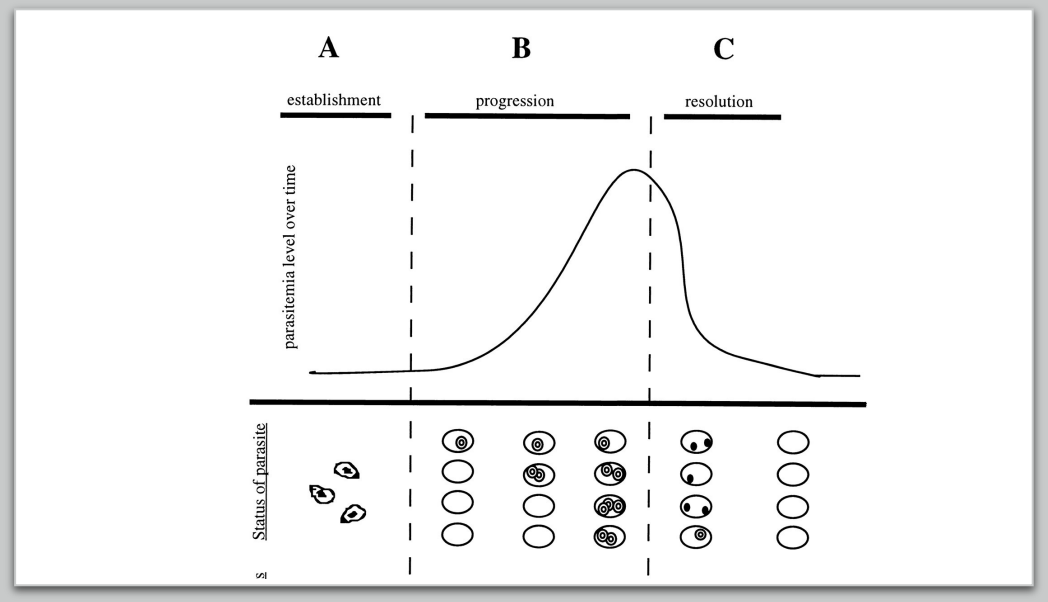
Blood Smears

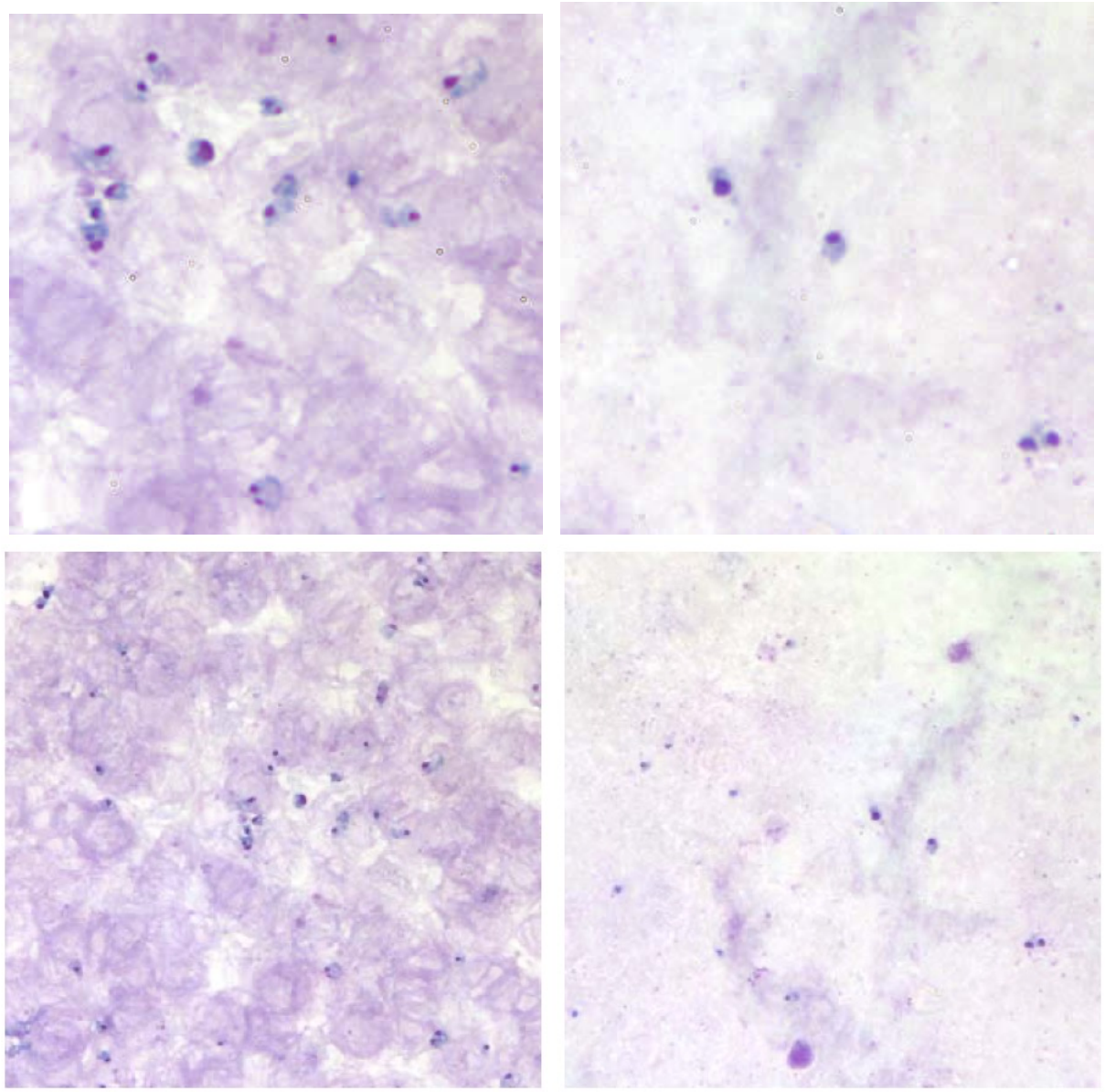
- Typically appearing in infected cells as small (1–2.0 µm in diameter) basket-shaped rings with extended chromatin



- The organisms require several weeks or longer to establish a detectable infection
  - <1% of RBCs are parasitized in the first week of the illness, when most people seek medical attention.

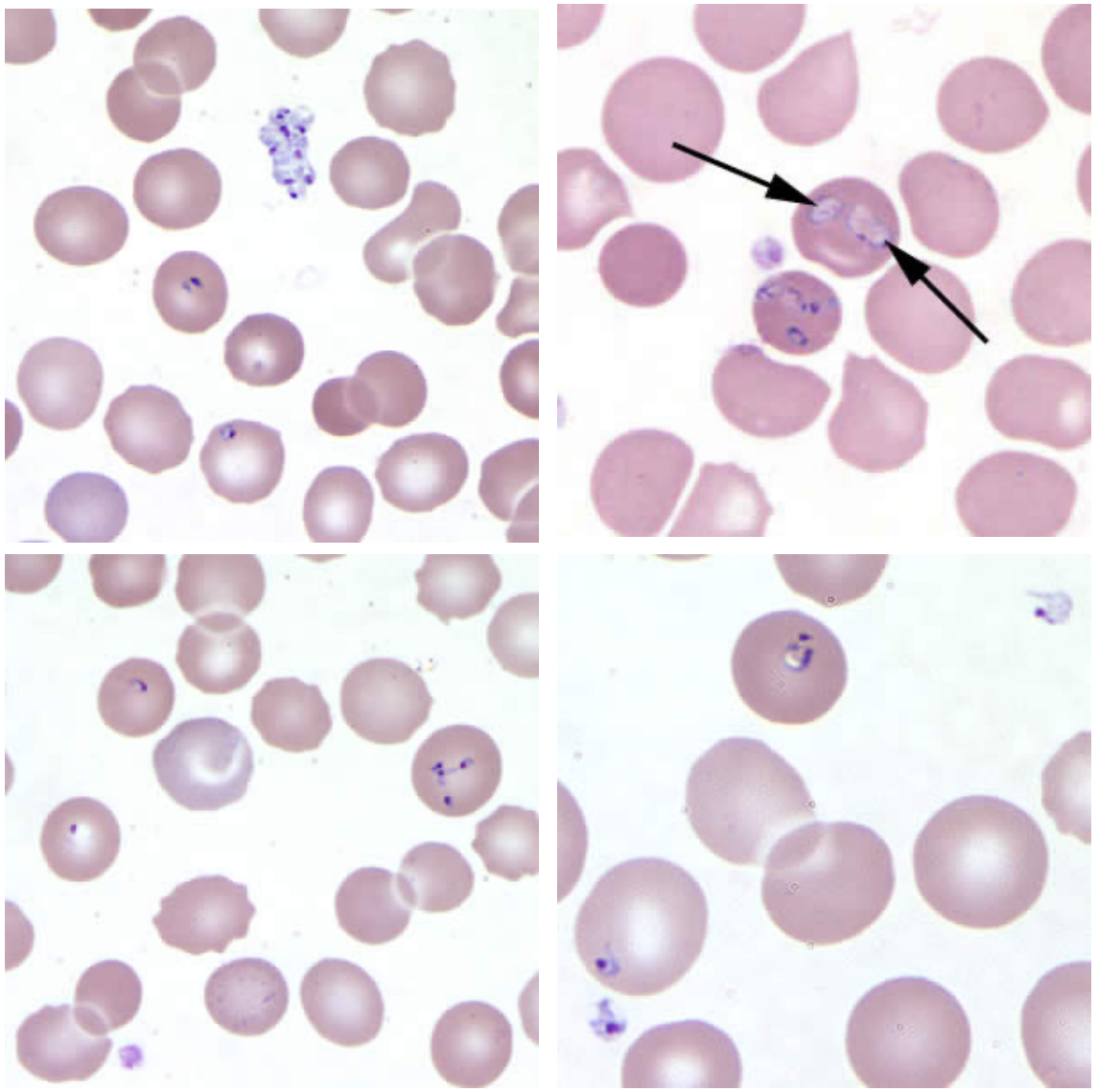






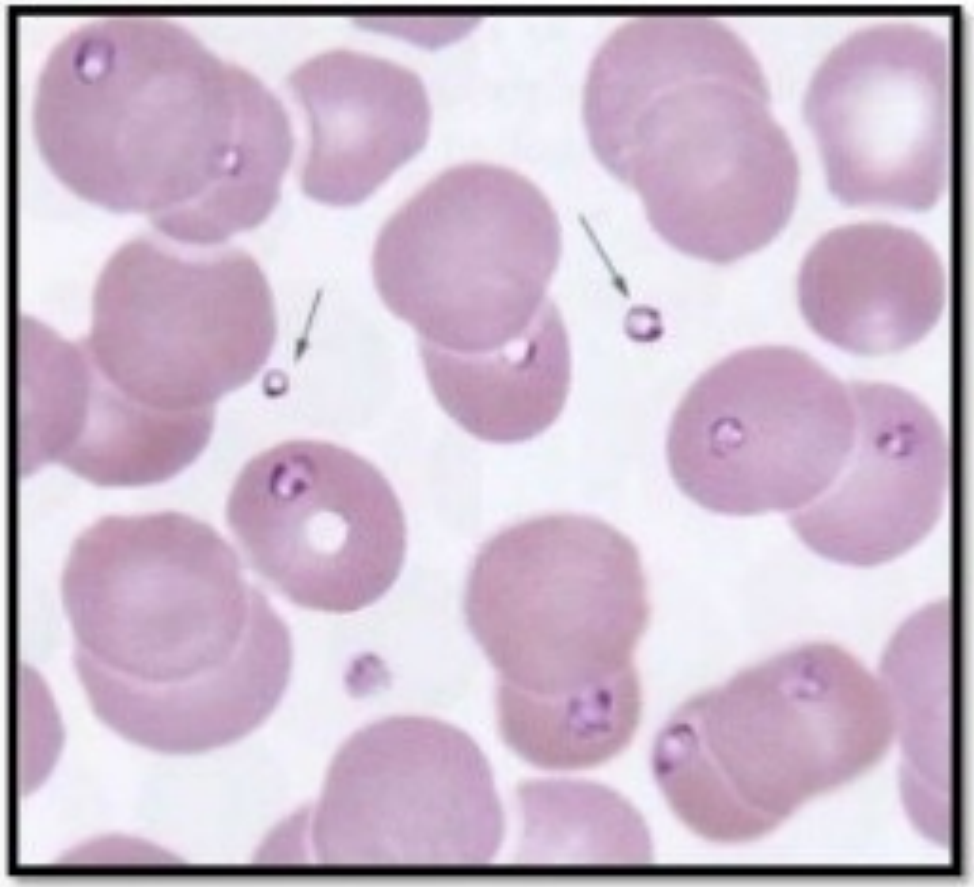
Thick Smears

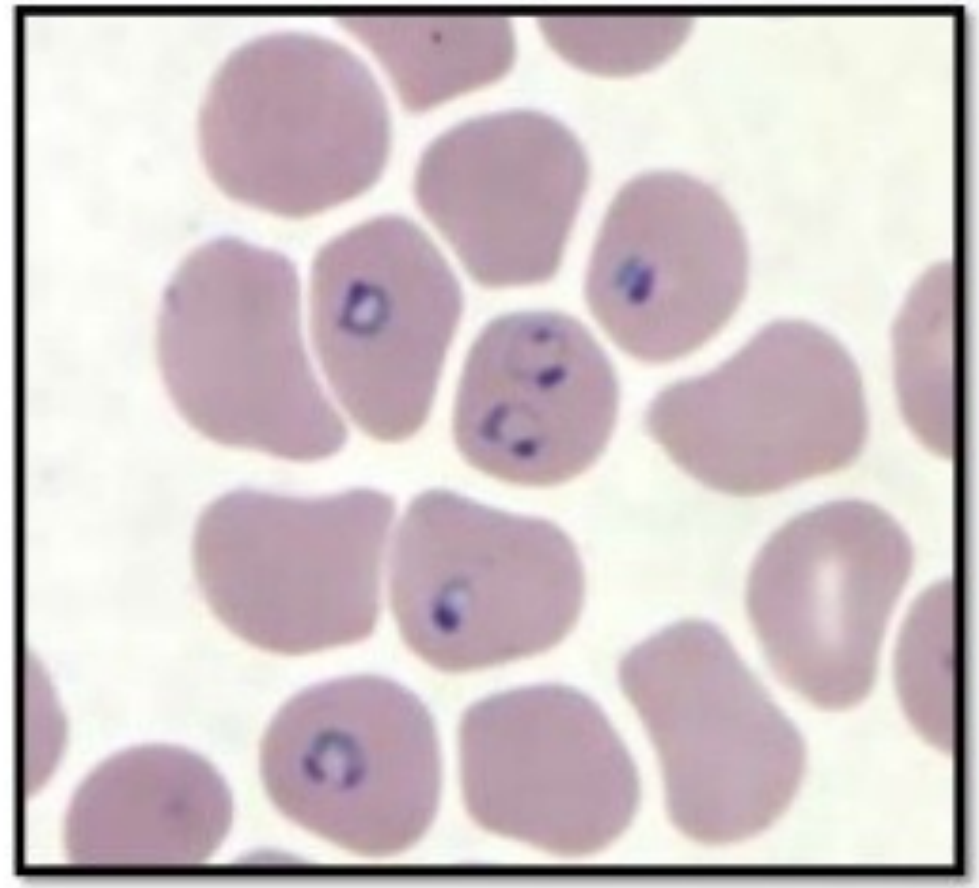
- Sensitivity

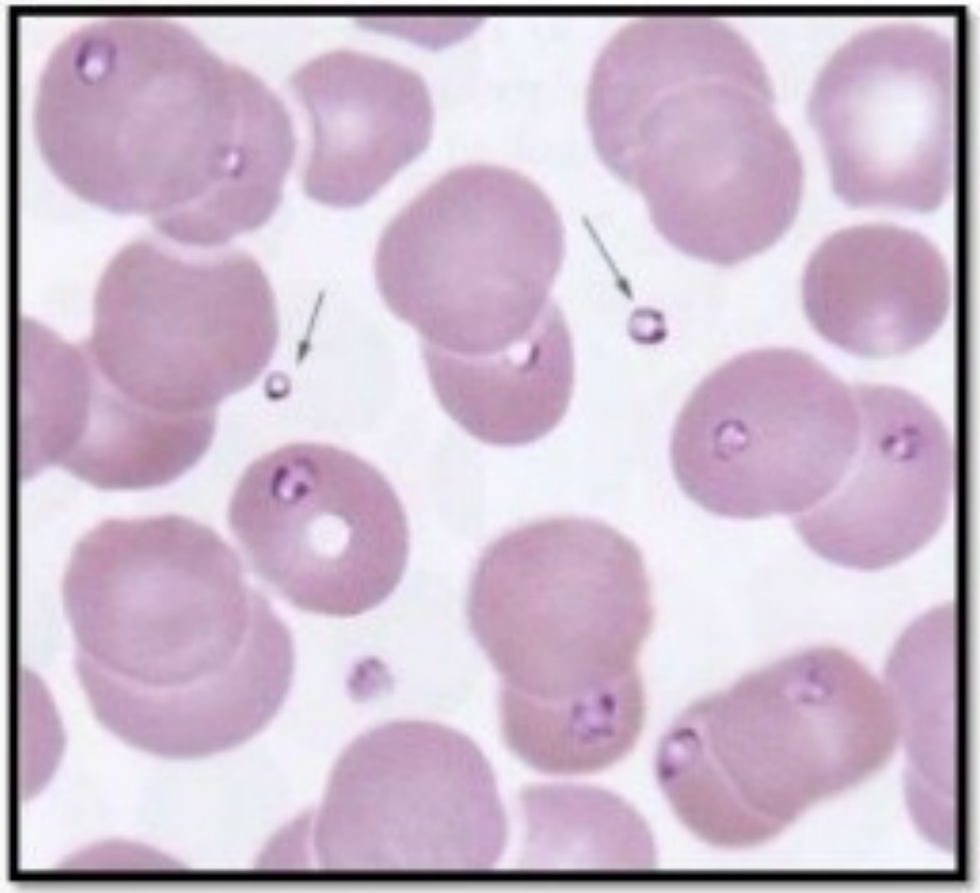


Thin Smears

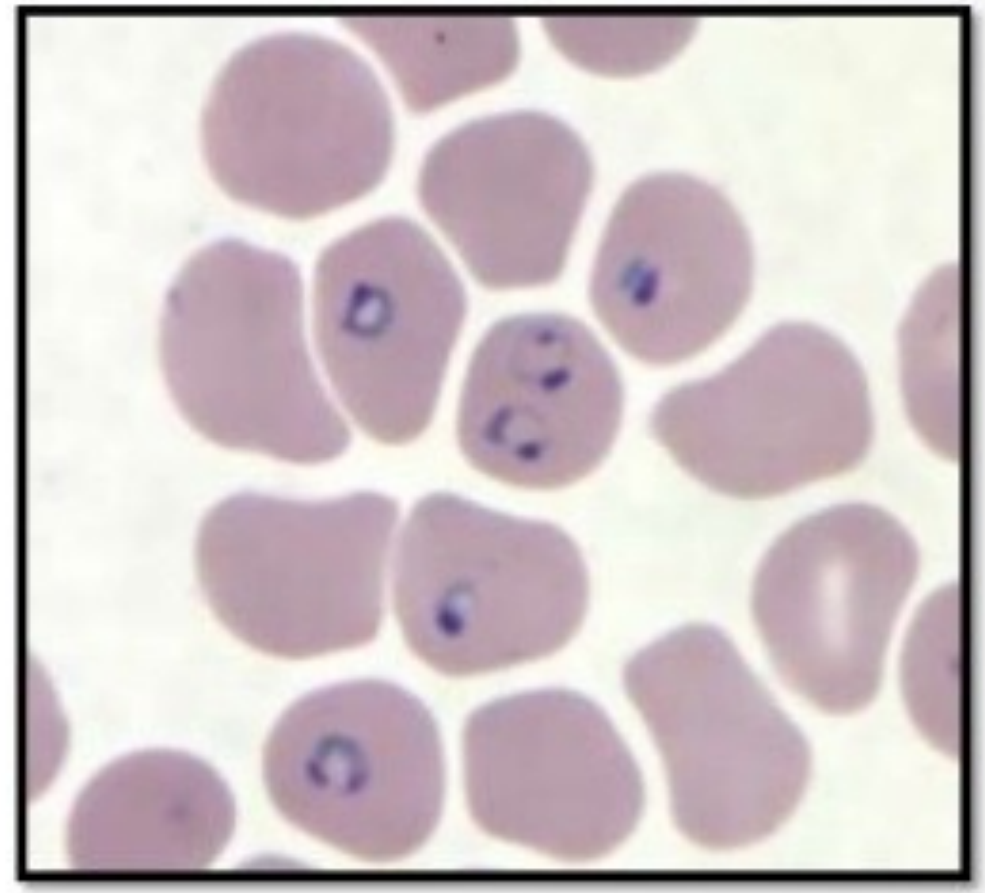
- Specificity
- Morphology
- Quantitative (% parasitemia)





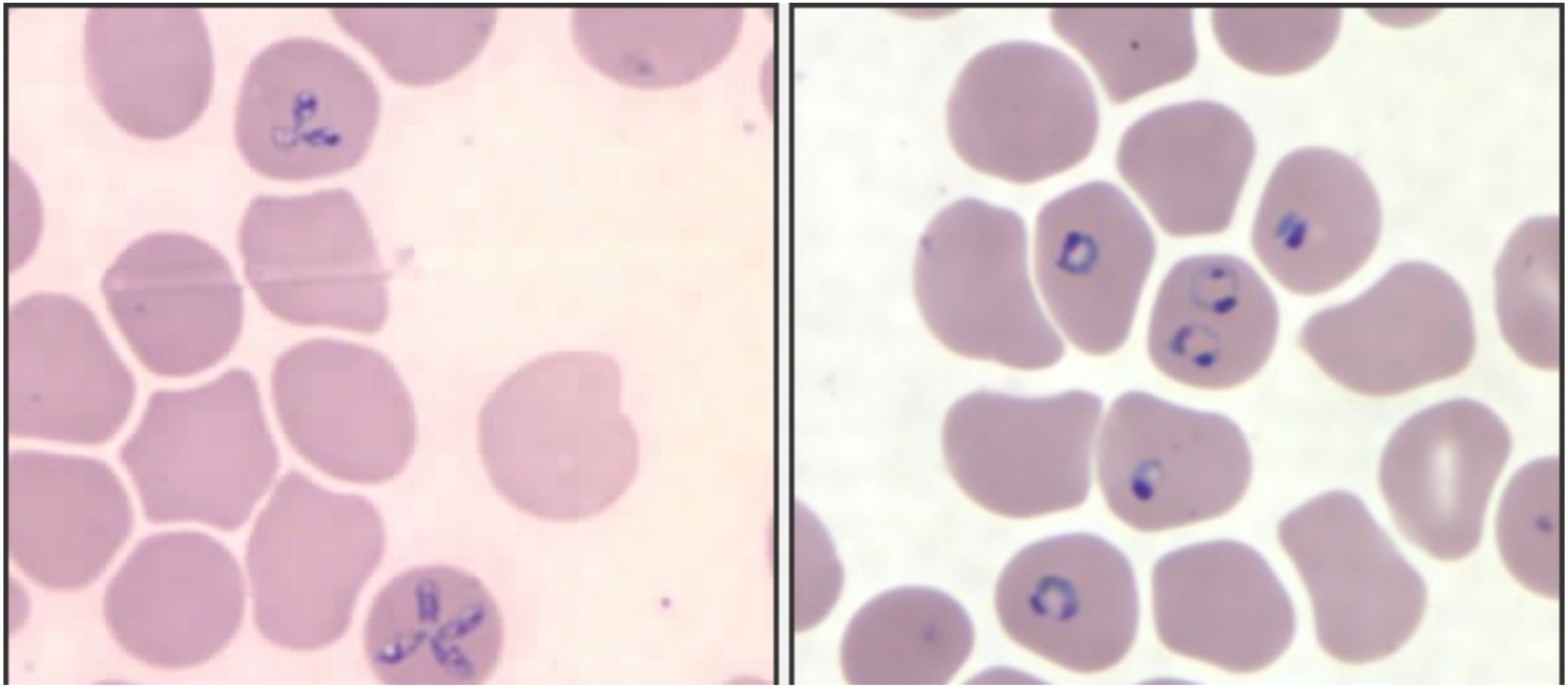


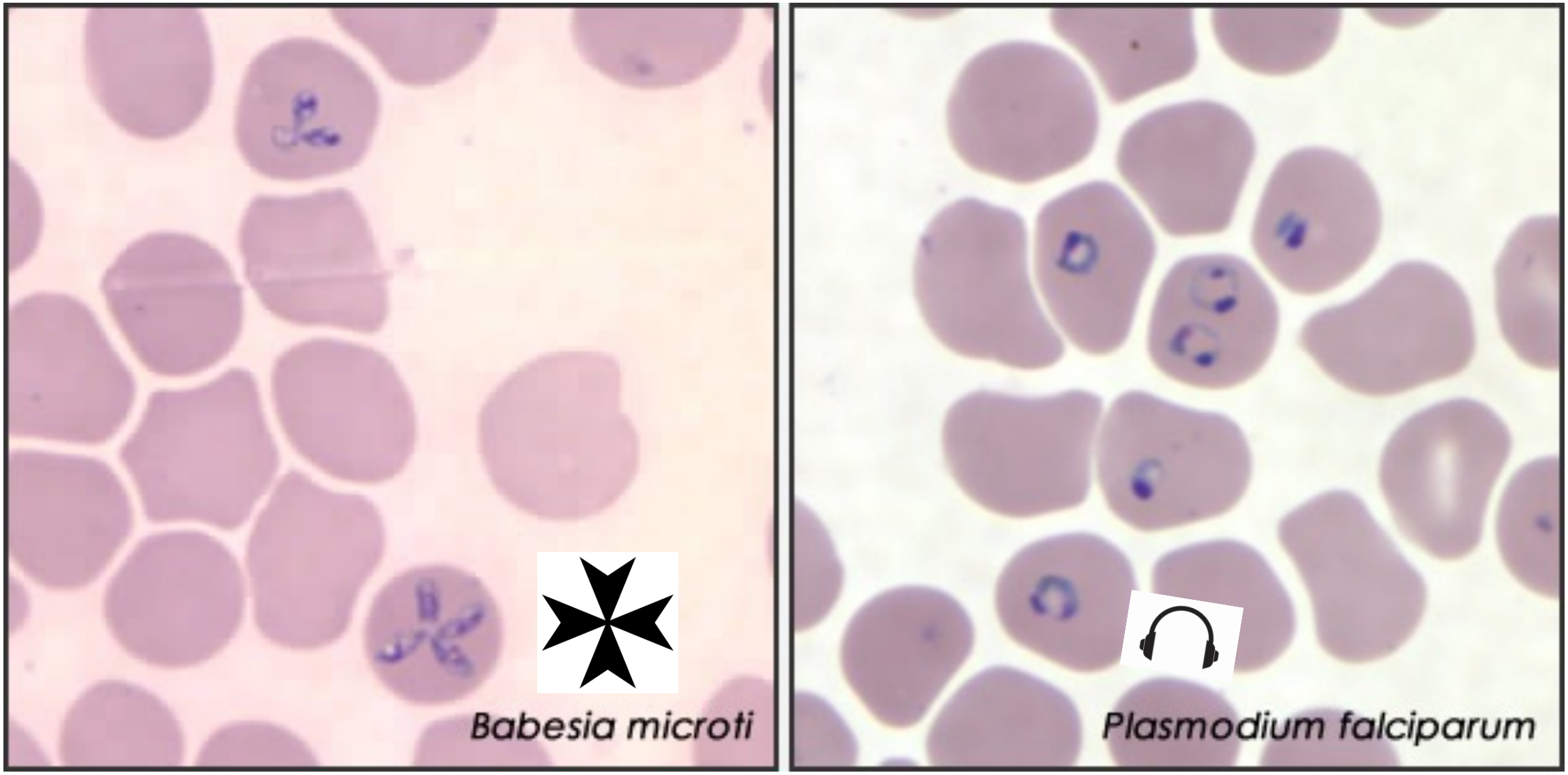
Babesia microti



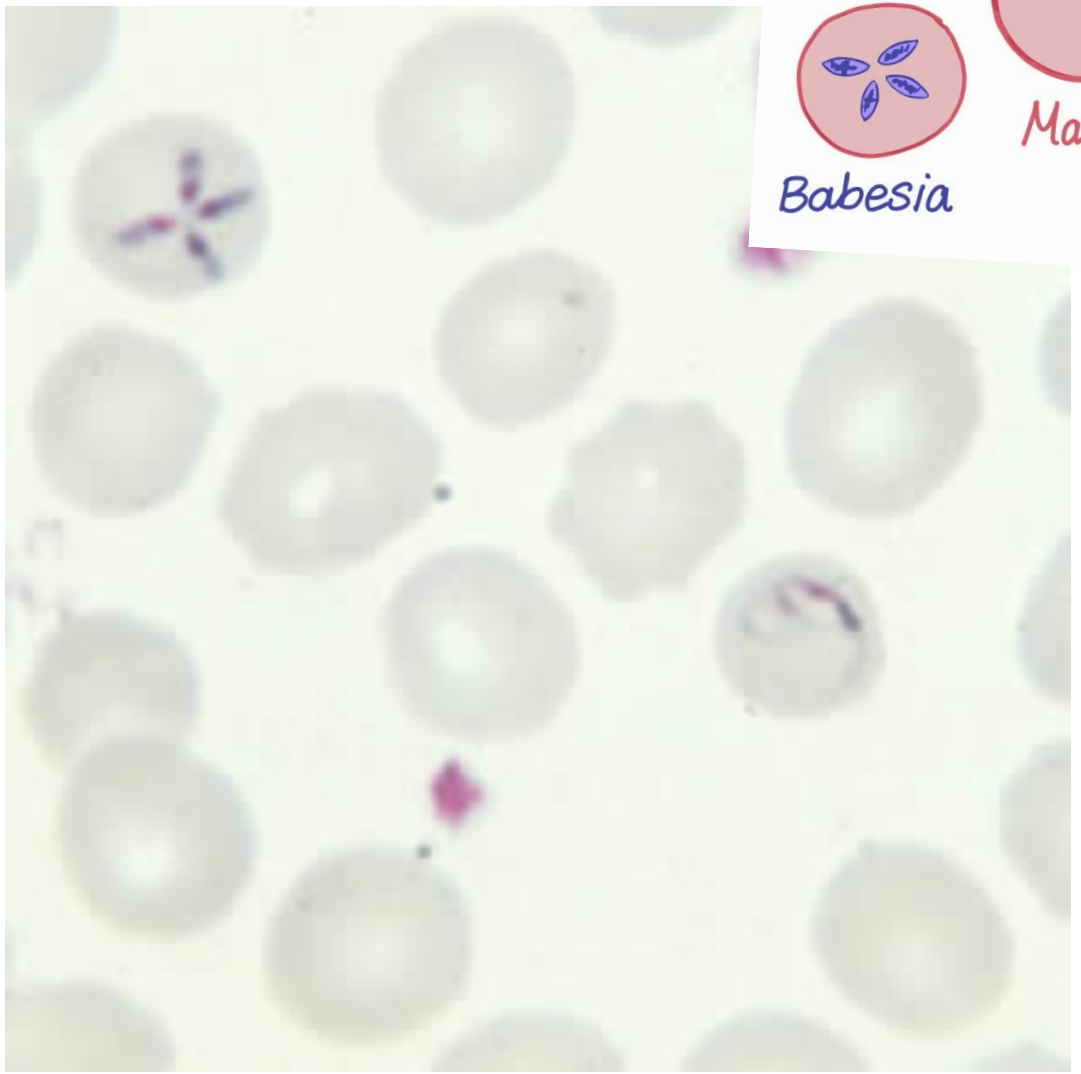
Plasmodium falciparum

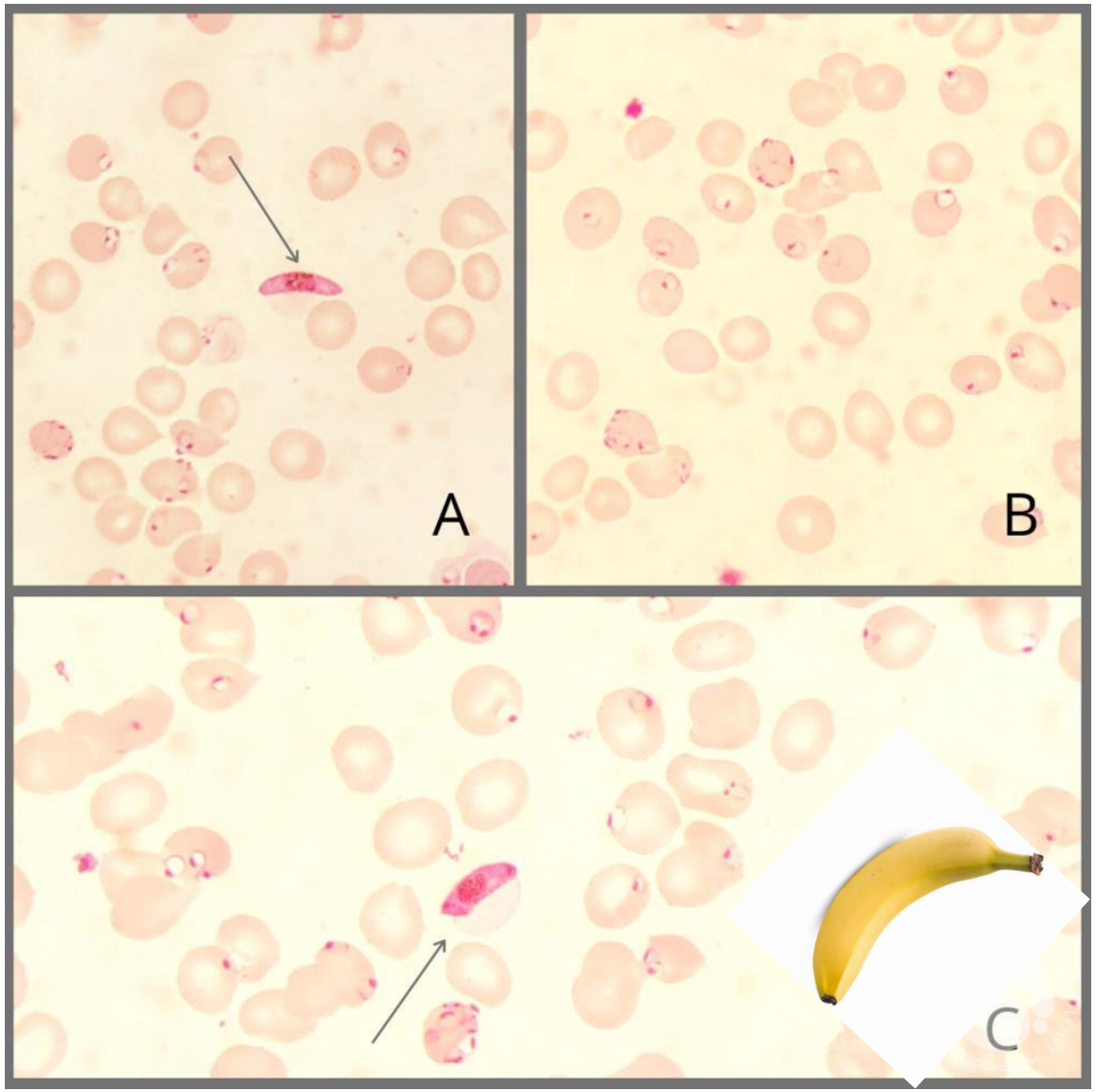
Holtry; https://www.antiinfectivemeds.com/parasitic-infections/malaria-and-babesia/





CDC Blog: https://blogs.cdc.gov/global/2014/02/24/dpdx-15-years-of-strengthening-laboratory-capacity-for-parasitic-disease-diagnosis/b_microti_vs_p_falciparum-2/





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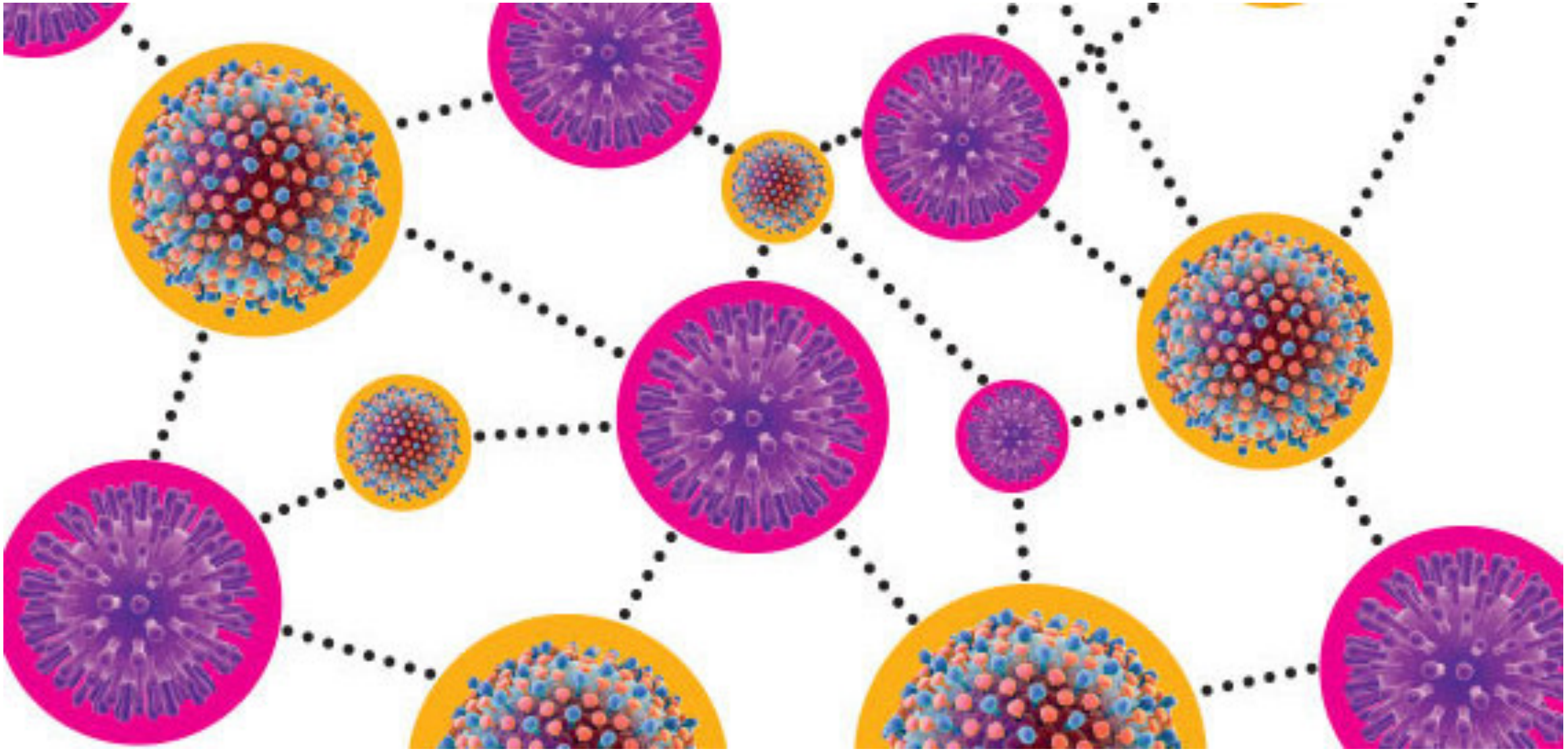
What are the pros and cons of Blood Smears vs. Molecular Detection for blood-borne parasites?

Morphology vs. Molecular

- Unbiased
- Rapid TAT
- Need for % parasitemia
  - predict prognosis, guide patient management, and monitor response to treatment
- Need for experience
  - Testing Capacity?!
  - Specificity- Malaria!?
- Cannot speciate Babesia
- Decrease sensitivity
  - Low grade parasitemia/early in infection

- Increased sensitivity
- Species-level identification
  - Need for informed primer targets
  - Biased/primer selection (vs multiplex)
- Testing Capacity
- TAT +/-
- Specificity (r/o malaria)
- ?! DNA copy number to quantitate

How do you manage the possibility of co-infections in patients with vector-borne illnesses?



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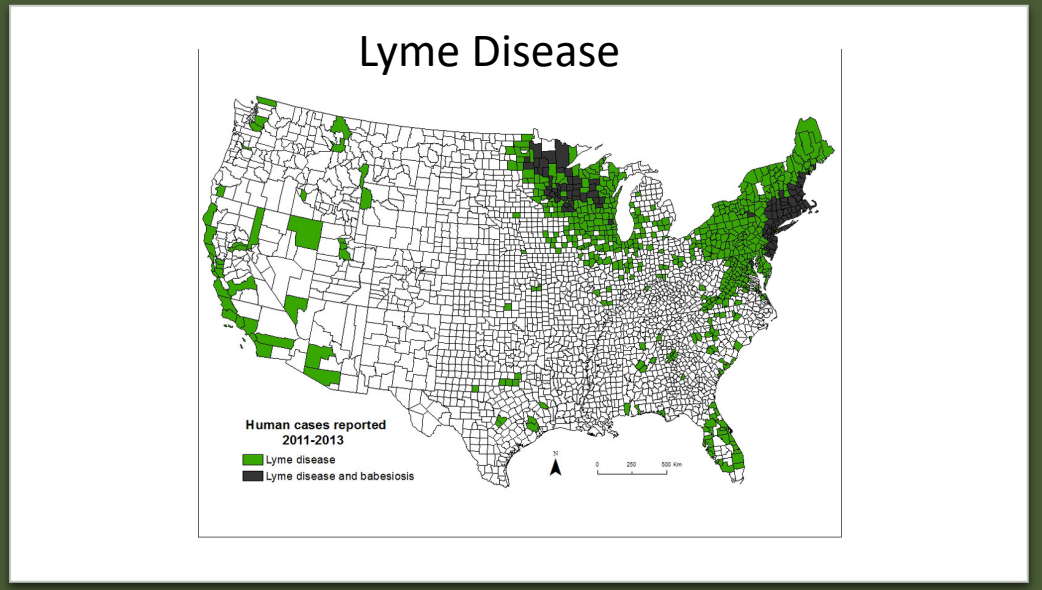
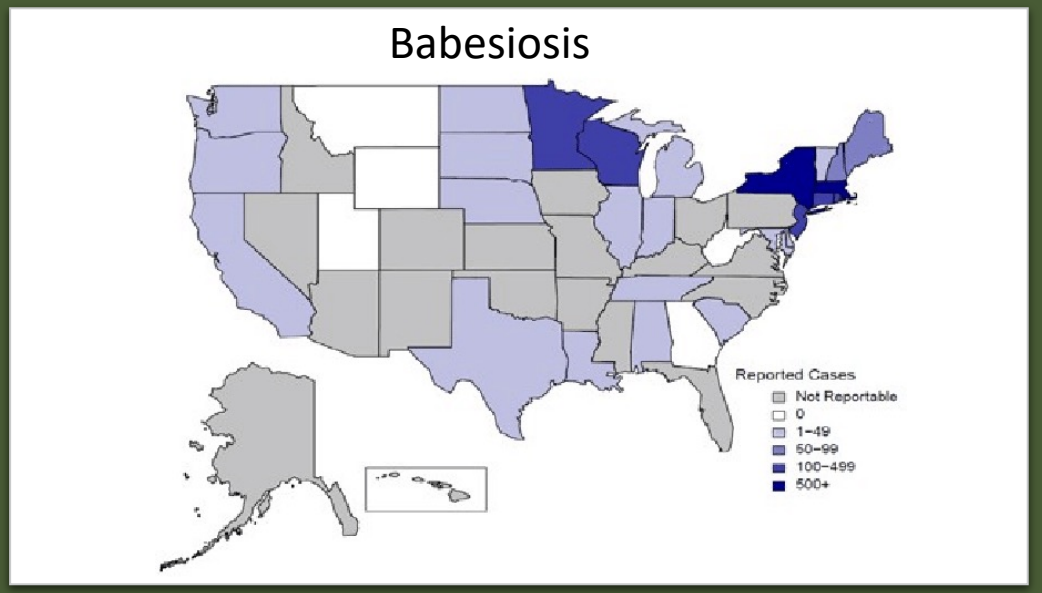




Diuk-Wasser MA, et al. Trends Parasitol. 2016

CASE HISTORY #2:

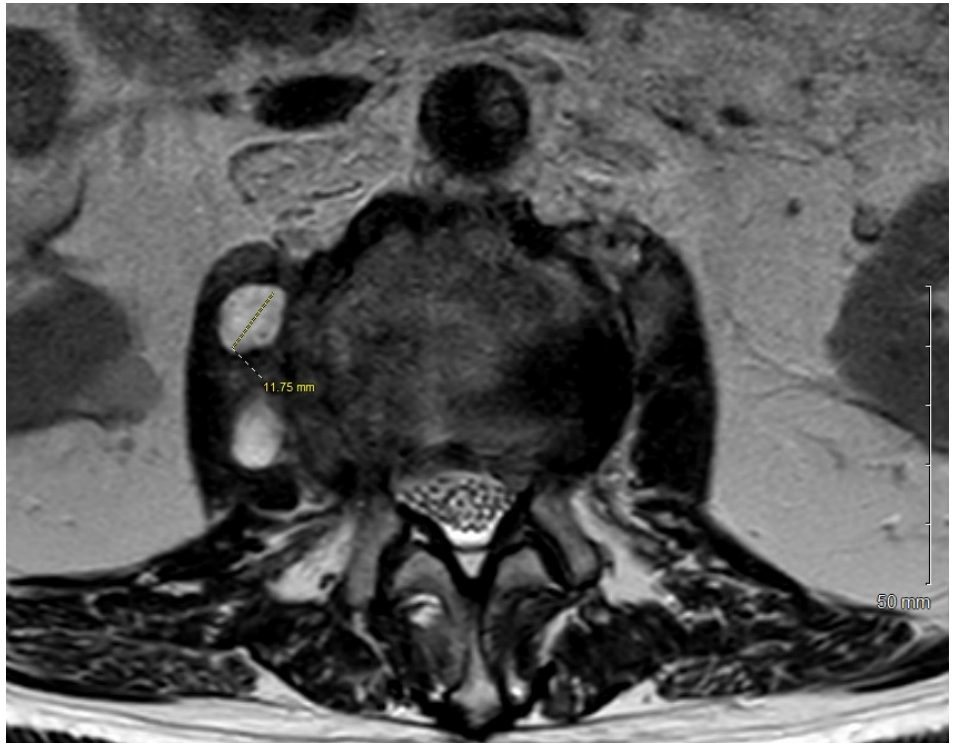
- A 75 y/o previously healthy man, presents to the ED with low-back pain.
  - Lifelong Vermonter currently lives with wife in Colchester. Briefly lived in Delaware at age 18.
  - Retired mechanic, 32 years in Air National Guard, remote travel to Panama.
  - Now works as a handyman, carpentry as a hobby.
  - Former smoker, quit 30 years ago. Rare alcohol. No recreational drugs.
  - Avid hunter – October 2020 hunting deer in central PA

CASE HISTORY #2:

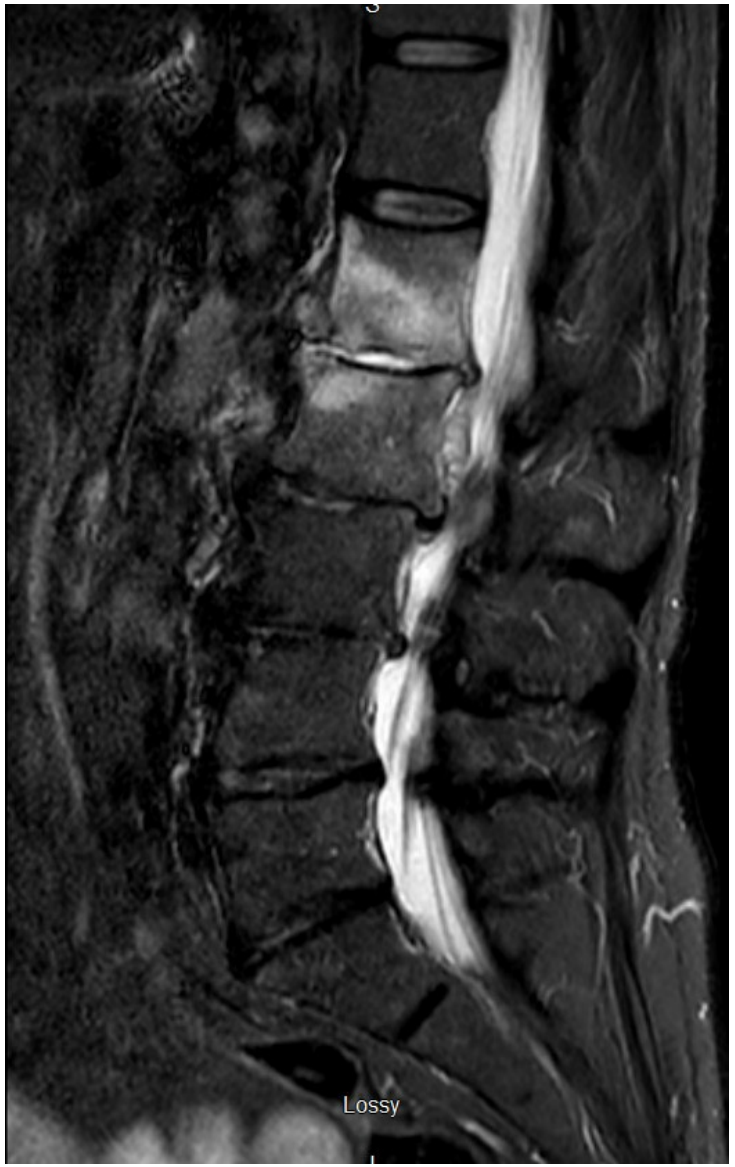
- 7 months prior presented to PCP w/ acute onset fever to 101.3 with chills and urinary frequency
    - Diagnosed clinically w/ prostatitis
    - Rx'd 10 days of TMP-sulfa DS BID
    - Negative urine culture
- 6 days after completion of antibiotics, fever to 102 and return of LUT symptoms
    - Tenderness on DRE, PSA of 10
    - Prescribed 4 weeks of cipro 500 mg BID w/ resolution
    - Urine culture negative
- urology referral - 2 months later: symptoms resolved, normal DRE, normal PSA
- 1 month later, new low-back pain radiating to groin
    - Attributed to rental car with poor lumbar support and strenuous yardwork
    - Trips to urgent care, Rx'd muscle relaxants and methylprednisolone with temporary relief.
- Another month later –
    - epidural steroid injections
    - PCP orders an MRI
        - discitis with multiple paraspinal/psoas muscle abscesses
        - Admitted for IR biopsy and ID consult

CASE HISTORY #2

- MRI of the lumbar spine concerning for paraspinal abscess.
- Admitted for discitis with **vertebral osteomyelitis, epidural phlegmon, and psoas abscesses.**
- possible staph bacteremia



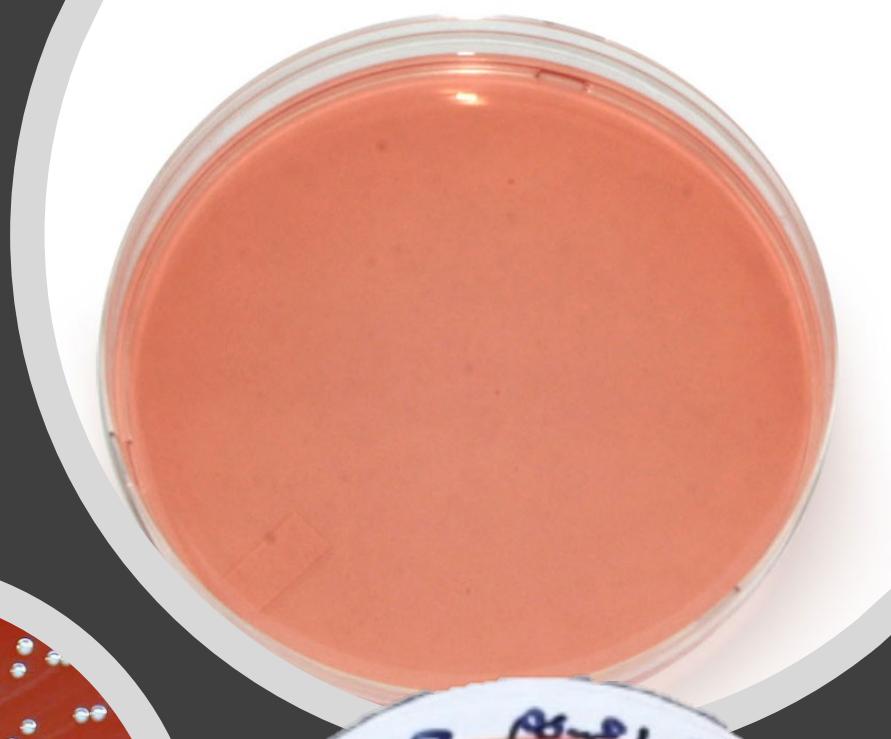
Axial: multiple abscesses in the psoas muscles at the level of L1/L2…

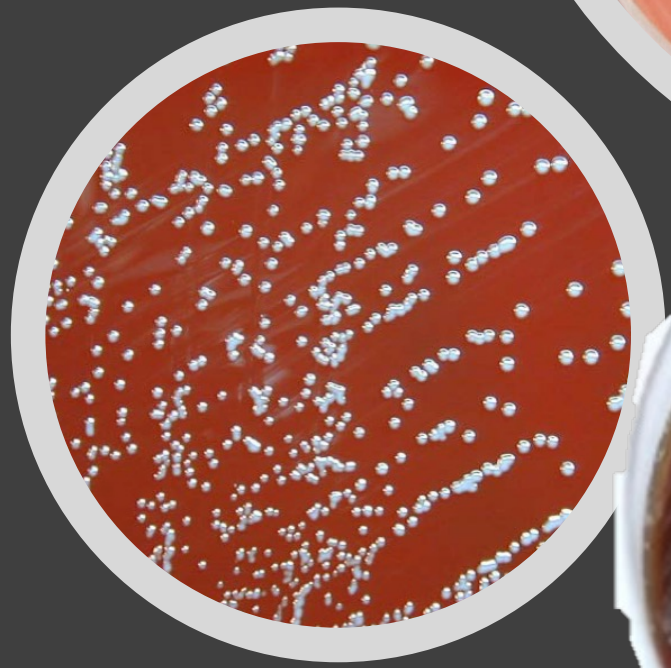


Sagittal: discitis osteomyelitis at L1/L2 with a small amount of epidural phlegmon

CASE HISTORY #2: Lab Results

- Elevated CRP
- Normal PSA
- Admission Blood cultures:
  - positive for staphylococcus capitis and staphylococcus epidermidis
- IR biopsy specimen:
  - Histology: necrotic fibroconnective tissue and collagenous tissue with acute inflammation.
  - Cultures:
    - AFB negative
    - Anaerobic cultures positive for……

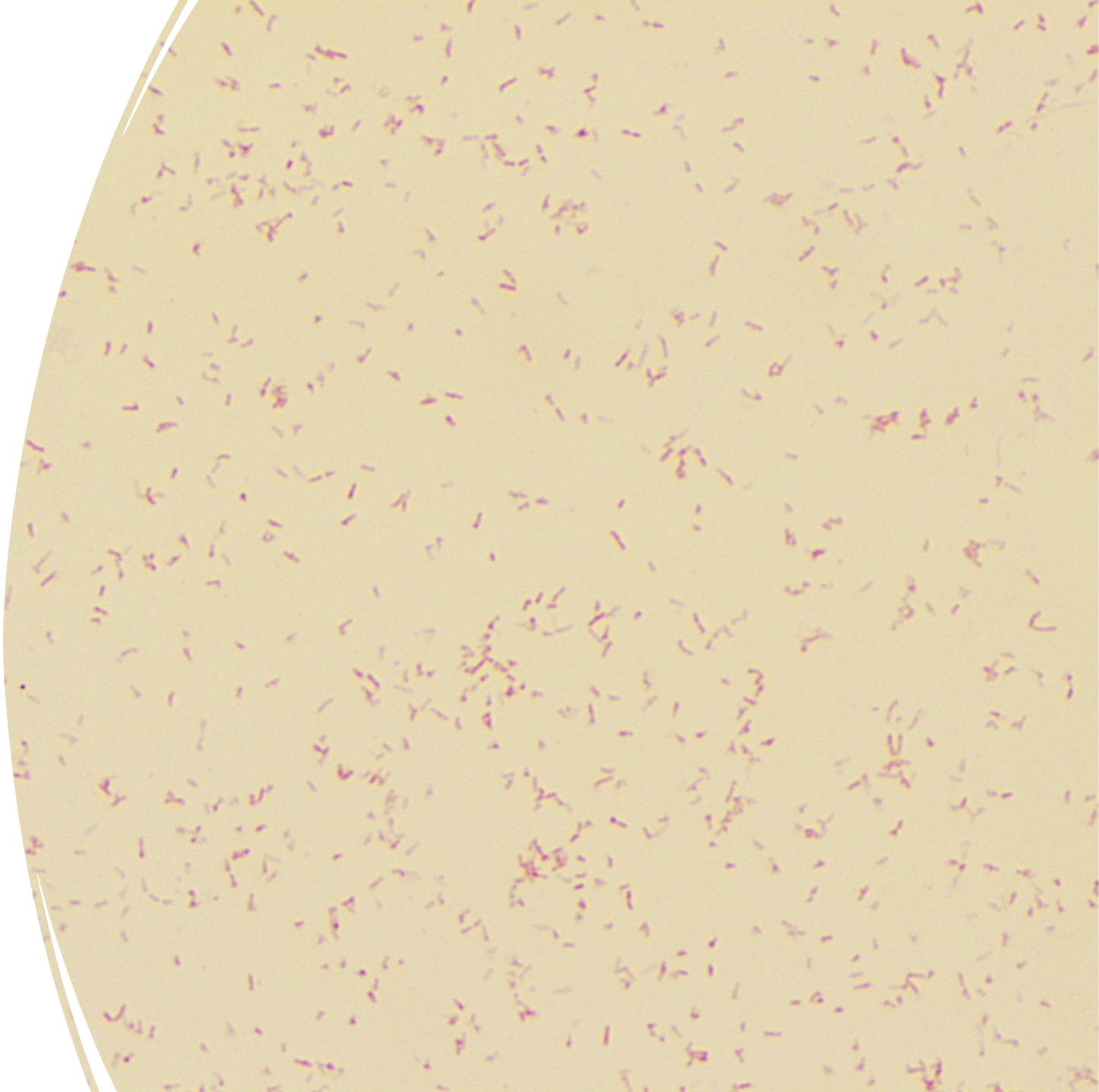




MALDI ID is…….

Brucella spp.

- Incredibly low infectious dose
  - (10-100 organisms)
- Most commonly reported laboratory-associated bacterial infection
- Is this real?
  - Gram stain (in BSC)
  - Clinical History….
- Call Clinical Team
  - Call Department of Health



More case history…

- Feral bore hunting near Lake Okeechobee, FL 8 months prior

- Grandson had killed the animal, patient accidentally cut his finger while assisting in processing of the animal

CDC Home | Search | Health Topics A-Z

MMWR Weekly

June 12, 2009 / 58(22);618-621

Brucella suis Infection Associated with Feral Swine Hunting --- Three States, 2007--2008

- Feral hog data collected by USDA indicate seroprevalences as high as 50% in some feral swine herds in central Florida, and statewide prevalence is estimated to be 10-20%" (Florida DOH)

JCM
Journals.ASM.org

A Literature Review of Laboratory-Acquired Brucellosis

Rita M. Traxler,[a] Mark W. Lehman,[a,b] Elizabeth A. Bosserman,[a] Marta A. Guerra,[a] Theresa L. Smith[a]

Division of High-Consequence Pathogens and Pathology, Centers for Disease Control and Prevention, Atlanta, Georgia, USA[a]; Epidemic Intelligence Service, Centers for Disease Control and Prevention, Atlanta, Georgia, USA[b]

- Between 1982 – 2007, 167 potential lab exposures, 71 developed disease
- 88% aerosolization of organisms during routine identification activities, 11% lab accidents, 1% unknown
- High-risk exposures were 9.3 times more likely to develop LAB than low-risk exposures (95% CI, 3.0 to 38.6; P < 0.0001)
- 0.009 times likelier to develop LAB w/ PEP vs no PEP (95% CI, 0 to 0.042; P < 0.0001).
- Median incubation of 8 weeks (range 1 to 40 weeks)
- No LAB developed w/ PEP use

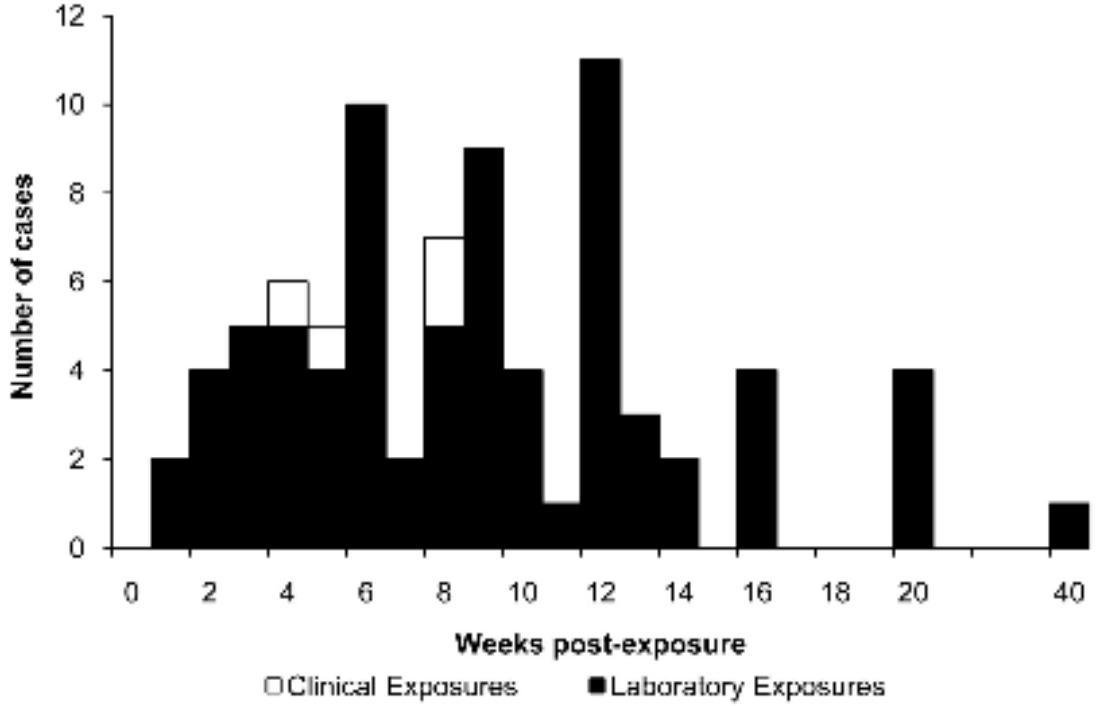


FIG 1 Time course of disease onset following occupational exposure to *Brucella* spp. ($n = 80$).

Immediate Next Steps:

- Stop work-up (do not attempt further testing)

- Laboratory Risk Assessment (High/Low/Not Zero)
  - Enriched vs. Non-enriched
  - Aerosolizing procedures
  - Open Bench vs. BSC
  - <5’ vs >5’
  - Risk Factors
    - pregnancy, immune suppressed
  - Serologic Monitoring +/- Post Exposure Prophylaxis (PEP)

- Immediately notify:
  - Treating team of presumptive diagnosis
  - LRN Reference Laboratory
  - Infection preventionists
    - Identify clinical high risk exposures

- To take blood cultures off incubator?
  - 5pm Friday
  - Laboratory risk assessment (working up positive bottles)
  - Bacteremia – prognosis/patient management

Brucellosis

- “Ancient” Disease
  - Animal Domestication, diary consumption
  - Lytic spine lesions
- 1799: 25k British soldiers occupied Malta
  - Malta fever, undulant fever, Rock of Gibraltar fever, and Bang’s disease.
  - Between 1901 – 1907, the Navy observed 1705 cases with 30 deaths; Army observed 1947 cases with 55 deaths
  - mortality rate of **6.9%**

Table 2.1. Number of cases of “Malta fever” admitted to Valletta Hospital from 1876 to 1888

Year	Cases
1876	28
1877	43
1878	56
1879	33
1880	27
1881	5
1882	5
1883	27
1884	44
1885	11
1886	91
1887	109
1888	45

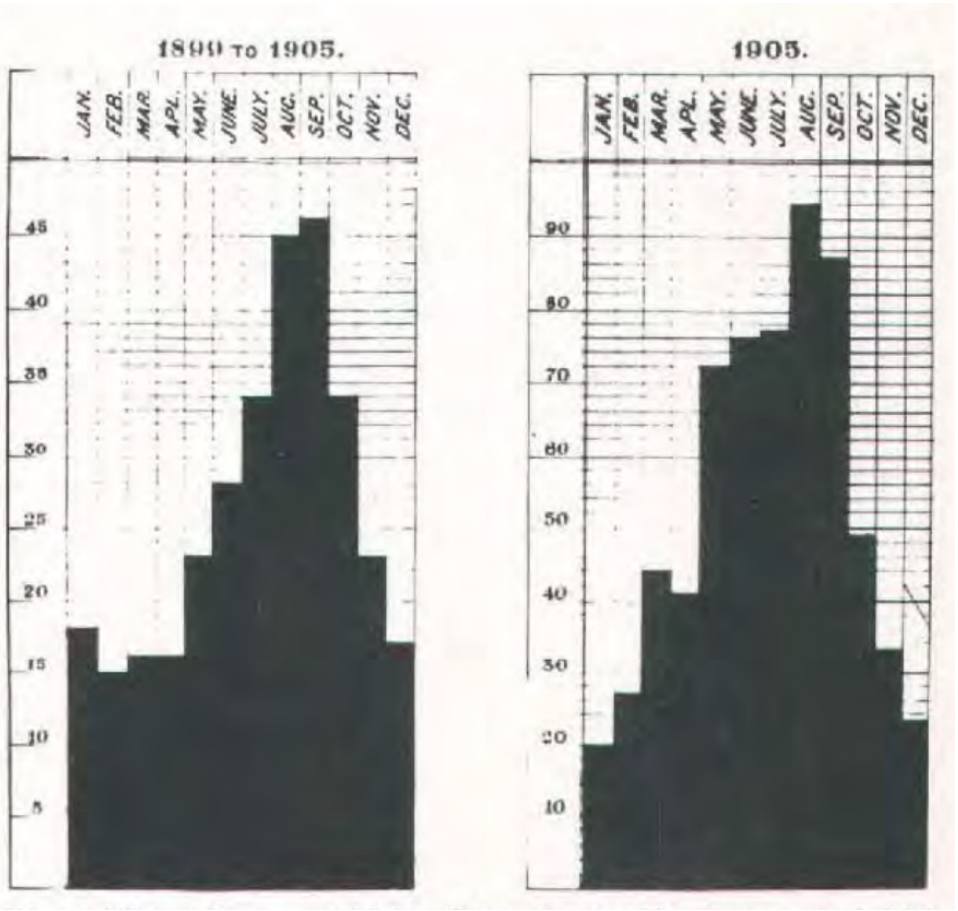
Brucellosis: Discovery

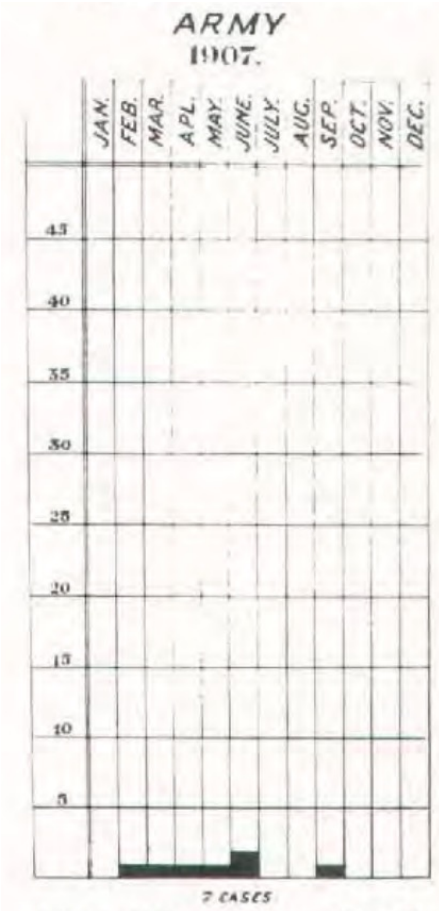
- David Bruce (1855-1931)
  - Australian-born British pathologist and microbiologist and British army medical doctor
- Mary Elizabeth Steele
  - Microbiologist

- In 1887, inspired by Koch's discovery of the Tubercle bacillus (1882), Bruce conducted an autopsy on a soldier who died 15 days after developing "Malta fever" and noted in the spleen:
  - *" ...the field of the microscope was literally crowded with a myriad of micrococci dancing about in the most active manner"*
  - Isolated and named *Micrococcus melitensis* (latin for "Malta")
  - Later isolated organism from 6 other fatal cases (spleen, liver, kidney)

- 1897: Wright and Smith successfully adapted the typhoid agglutination assay developed by Gruber and Durham to diagnose Malta Fever

- Bruce chaired the Mediterranean Fever Commission (MFC) in 1904 to identify the source of infection and mode of transmission
- Goat farmers noted to be disproportionately affected
- In 1904, this prompted Dr. Themistocles Zammit to feed 6 goats agar cultures of *M. melitensis*, and observe (+) agglutination assays but no disease
- Within weeks, discovered that up to 50% of goats on Malta were positive, and 10% secreted bacteria into milk
- In 1906, consumption of goats' milk prohibited by the British Army





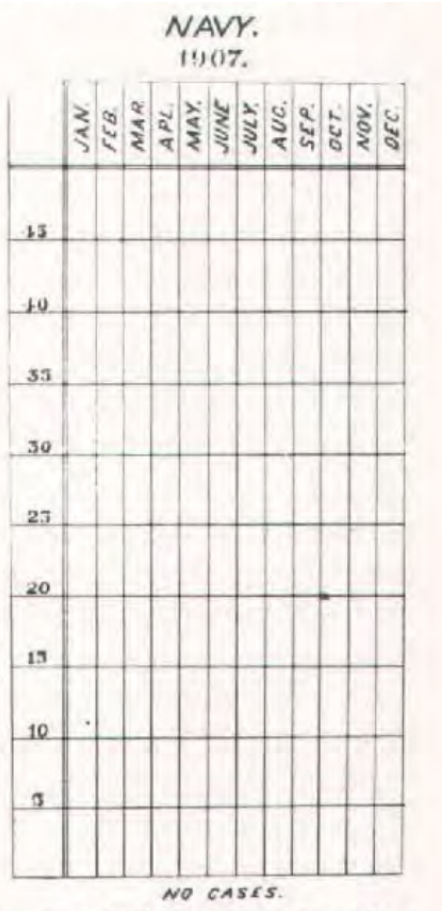


Fig 5. "Prevalence of Malta Fever, per 1,000 Servicemen, after Banning of Goats' Milk"; by David Bruce.
Acknowledgement to Wellcome Institute Library, London.

Vella EE. Brucellosis (the Corps disease). 1983 PMID: 6352932

David and Mary Elizabeth Bruce

- **1894:** discovered *Trypanosoma brucei* as the causative agent of African sleeping sickness

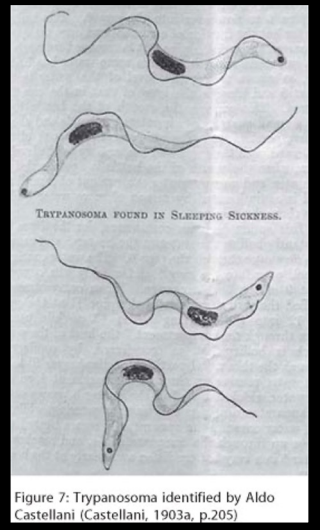
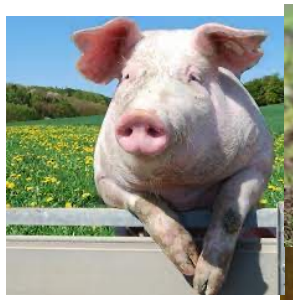


Figure 7: Trypanosoma identified by Aldo Castellani (Castellani, 1903a, p.205)

Brucellosis: Transmission

- Zoonotic infection
  - Consuming milk products, traumatic inoculation, airborne
  - 4 species being causing human infx:







Pig	→ B. Suis	→	High Pathogenecity
Dogs	→ B. Canis	→	Modertae Pathogenecity
Cattle	→ B. Abortus	→	Modertae Pathogenecity
Sheep	→ B. Mellitensis	→	High Pathogenecity

*Most common

labpedia.net

- Case repots:
  - *B pinnipediae and B cetaceae (*marine animals*)*
  - *Brucella inopinata (*breast implant infection)

Brucellosis: Transmission

- Human to human transmission unusual
  - Bone marrow transplant, Blood transfusion
  - Sexually Transmitted Disease
  - Neonatal brucellosis
    - transplacental transmission of *Brucella spp.* during a maternal bacteremic phase, from exposure to blood, urine, or vaginal secretions during delivery, or through breastfeeding
    - risk of causing spontaneous abortion, particularly during 1st and 2nd trimesters

Brucellosis: Clinical

- Average incubation = 2 - 10 weeks (-6 months)
- Among the “great imitators”
  - spectrum of asymptomatic to fulminant disease
- Symptoms are non-specific and systemic
  - intermittent fever, sweats, headache, anorexia, fatigue, weakness, malaise, back pain, and weight loss being frequent.
  - GI symptoms in 50% of patients
- Acute and chronic infections
  - Chronic form can mimic miliary tuberculosis.
  - abscesses in the liver, spleen, heart valves, brain, or bone; osteoarticular complications
- Mortality ~ 5% in untreated individuals
  - endocarditis

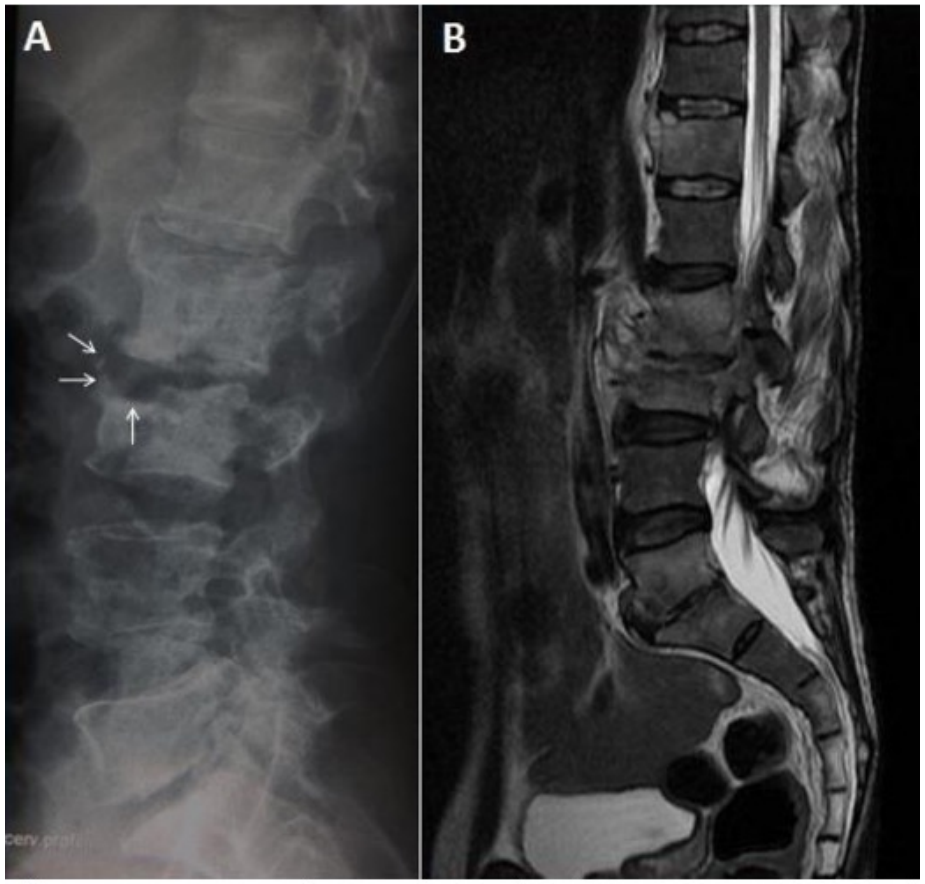
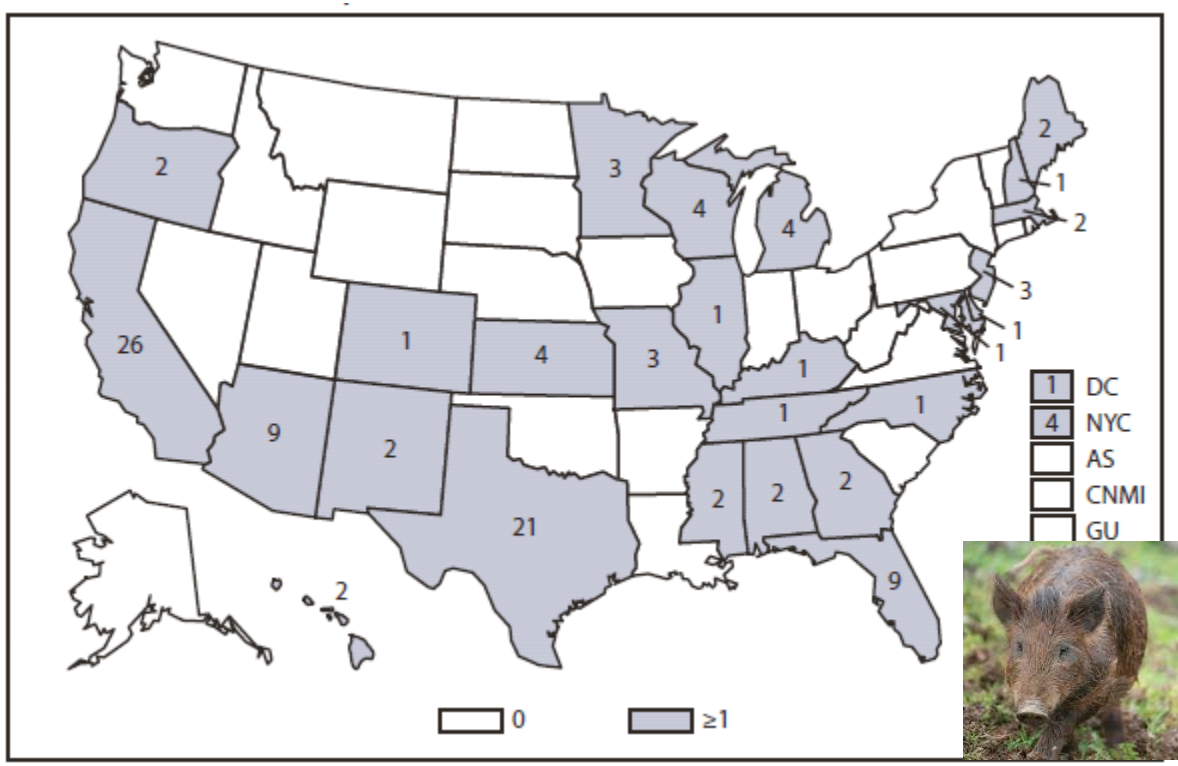


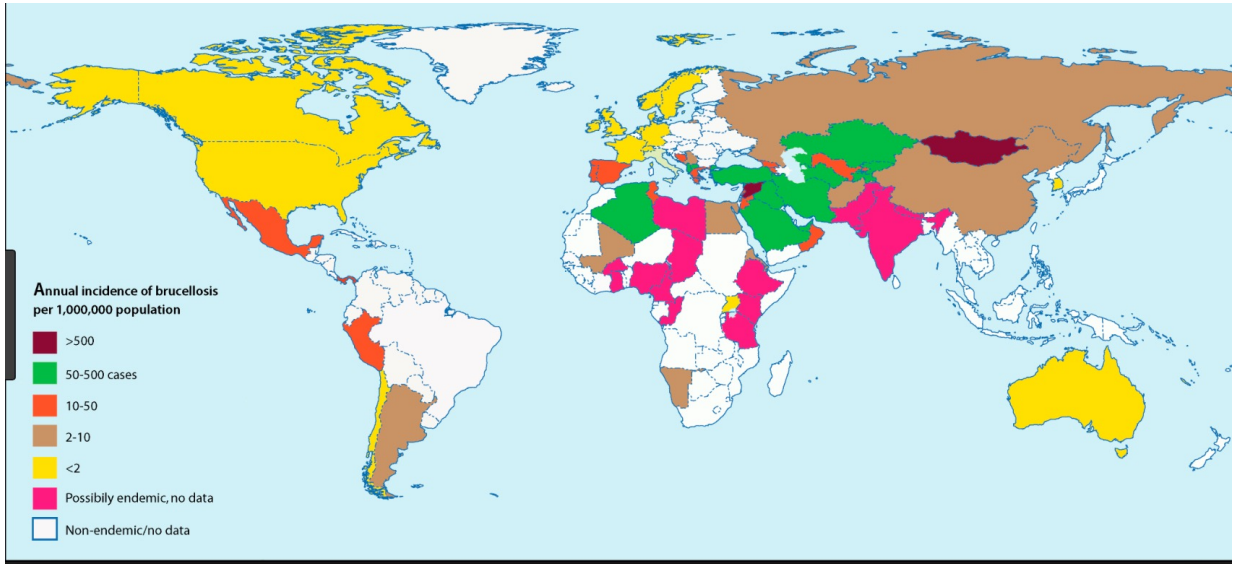
Figure 1: Radiography of the spine revealed

“Pedro-Pons sign” of Brucella spondylitis:
Steplike erosion at anterosuperior portion of a vertebral body

Brucellosis: Epidemiology

- 80 - 120 human cases of Brucella infection diagnosed each year in the US
  - highest reported incidence from California
- IP efforts: vaccination of cattle herds
  - In 2008, the USDA reported that all 50 states were Brucella Class Free
  - sporadic cases con't to be reported
  - Continues to be present in US ferel swine, bison and the elk populations

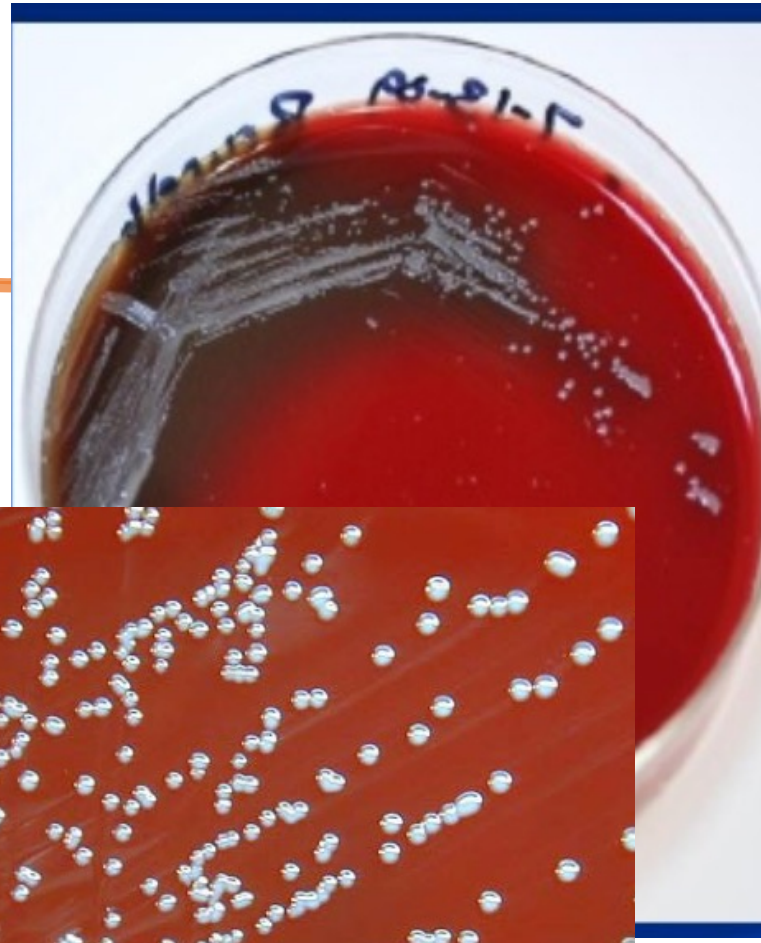




Brucellosis: Diagnosis

- A definitive diagnosis requires that bacteria be cultured from clinical specimens.
- Clinically, rapid identification to the genus level is adequate to initiate therapy, and the type of Brucella species involved does not alter the therapy

Microbiologic Identification

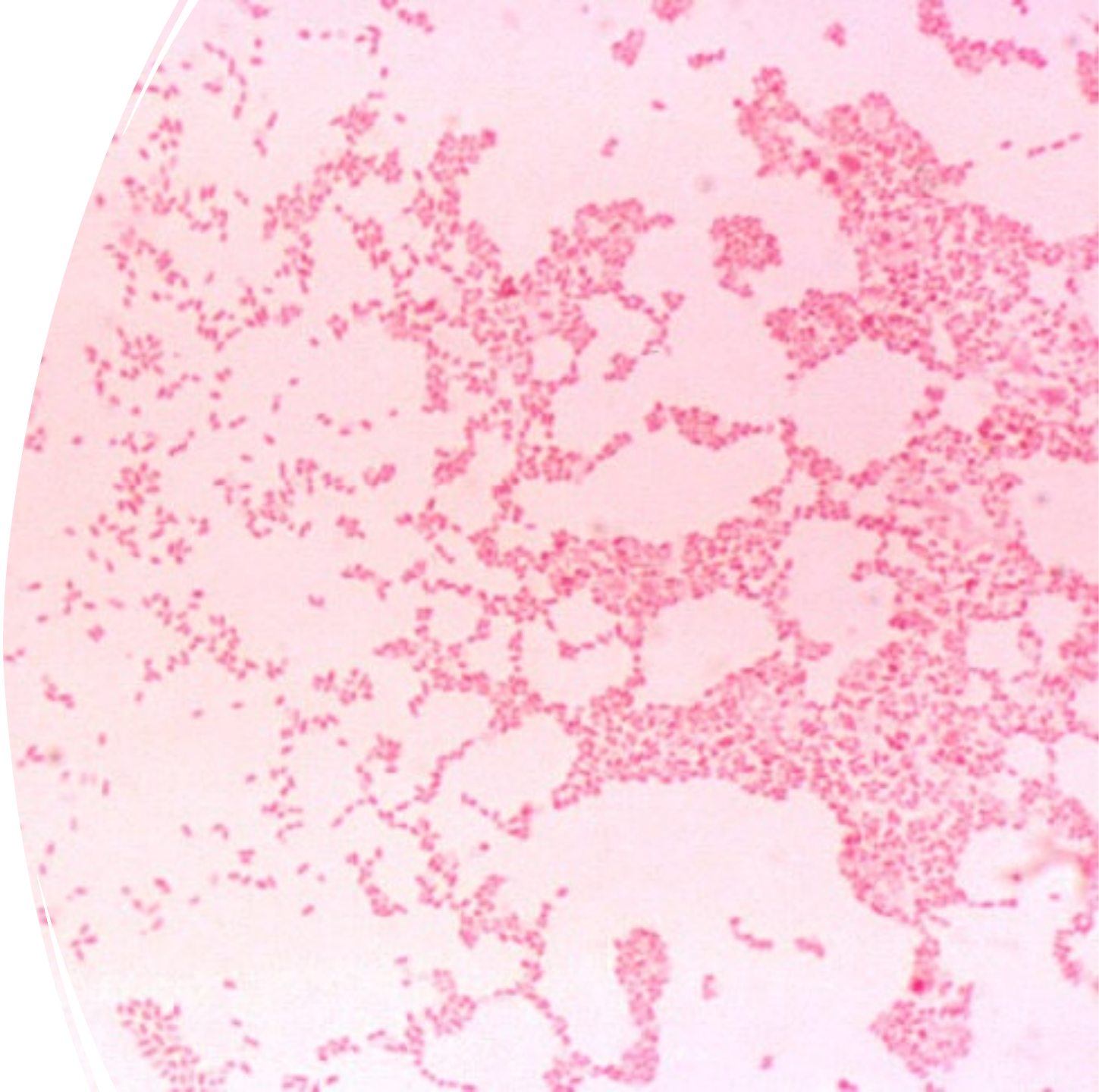


- fastidious, aerobic, small, gram-negative coccobacillus that is neither motile nor spore-forming
- Grows on BAP, CHOC
  - Pinpoint colonies have been infrequently observed on MAC after extended incubation times (7 days).
  - Best growth on TSA (Trypticase Soya Agar + Polymixin, Cyclohexamide)

- Hold plates for 10 days
  - Pinpoint at 24 h and are easily visible as 0.5 to 1 mm white, non-hemolytic colonies at 48 hrs
  - peak isolation occurring at 3 to 4 days (vs. 6 - 36 h for most pathogens)
- Small, raised, convex white colonies with an entire edge and shiny surface, moist, translucent glistening
- Not mucoid, non-pigmented, anon-hemolytic and have no distinct odor.

Gram Stain

- Tiny (0.4 by 0.8 µm), gram-negative coccobacilli that stain faintly.
- Unique in that the organisms are small, stain poorly and not clustered
- Confused with poorly staining gram positive cocci, because of their tiny size.



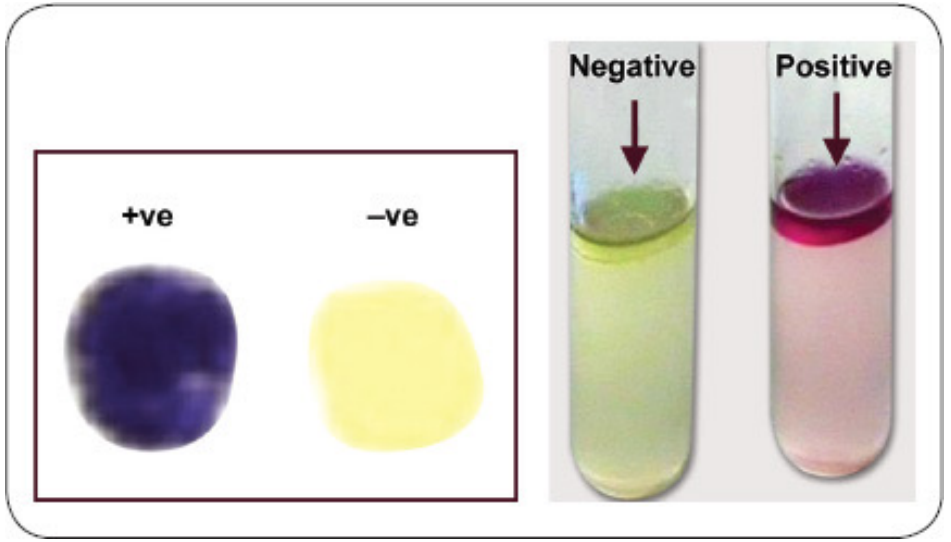
Micrococcus melitensis

- *"When a minute portion taken from one of these colonies is placed in a drop of sterilized water and examined under a high power, innumerable small micrococci are seen. They are very active, and dance about—as a rule singly, sometimes in pairs, rarely in short chains".*

MAJOR-GENERAL SIR DAVID AND LADY BRUCE.

Brucella spp: Presumptive Identification Criteria

- Faintly staining tiny gram negative coccobacillus
- Growing on BAP without the addition of a staphylococcus streak (satellite test), as nonhemolytic, non-pigmented, odorless, white colonies.
- Not growing on MAC in 48 h
- Oxidase-positive
- Catalase-positive
- Urease-positive



Brucella Identification Flowchart

SAFETY: As soon as *Brucella* is suspected, perform ALL further work in a Class II Biosafety Cabinet (BSL3)

Major Characteristics of *Brucella* species
Gram Stain Morphology: Small (0.4 x 0.8 µm), Gram-negative coccobacillus
THINK BRUCELLA

Growth: Subculture positive aerobic blood culture bottle to:
BAP, CHOC
Incubate in 5-10% CO_2 at 35°C

Spot BAP with *S. aureus* ATCC 25923 for satellite test.

Note poorly growing colonies after 24 hours incubation on BAP and CHOC.
Incubate plates for at least 2 additional days if no growth in 24 hours.
Organism does **not** grow on MAC.

Is the organism only growing on BAP without the need to satellite around the *S. aureus* at 24-48 hours?

No → Consider *Haemophilus*

Yes

Oxidase positive and catalase positive?

No → Think *Francisella* (see procedure)

Yes

Urea positive?

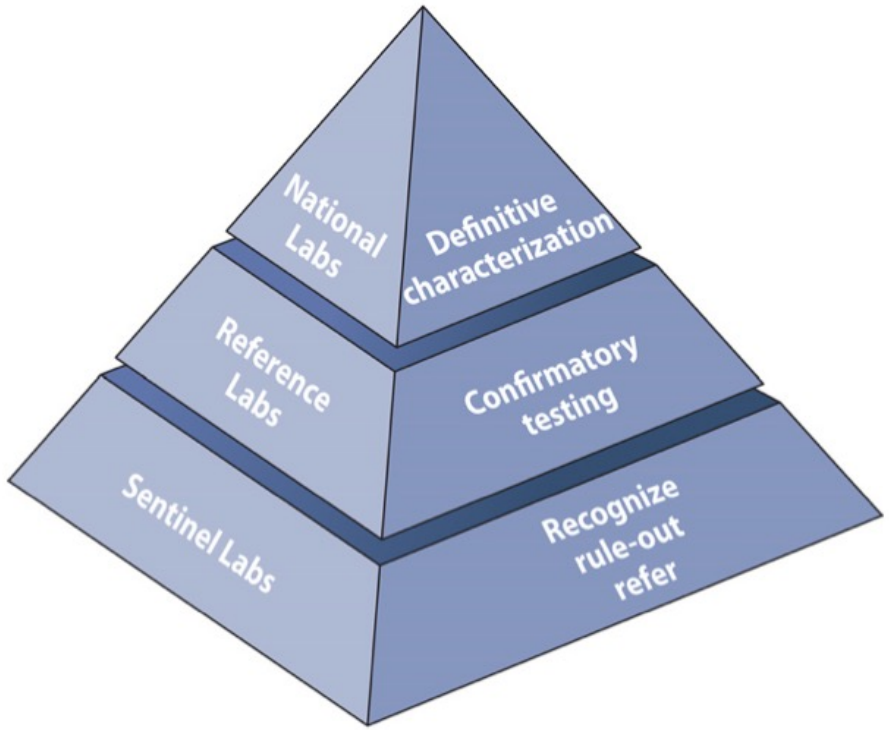
No → Reincubate and see written procedure

Yes

Brucella not ruled out.
Send to LRN Reference Level Laboratory.
Inform physician that *Brucella* species cannot be ruled out.

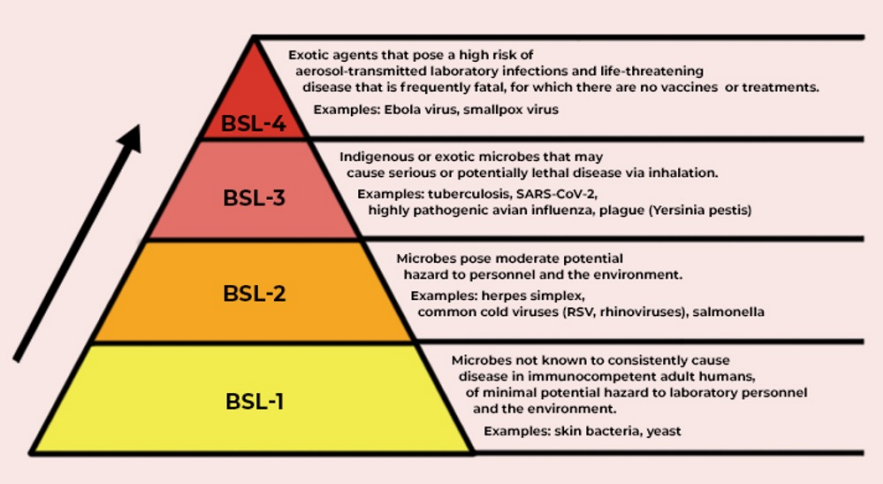
Antimicrobial therapy: Rifampin or Streptomycin plus Doxycycline

Note: Biochemical test procedures and quality control instructions can be found at the end of the *General Recommendation and Biochemical Testing Procedures* document.



Special Safety Considerations

- Avoid aerosolizing procedures
- Do not use multi-test kit or automated system for further ID
- Tape plates
- All further testing in BSC
  - BSL-3 or BSL-2 with BSL-3 precautions
- Precautions for manipulations of primary specimens
  - Enriched vs. non-enriched
- Plates and specimens should be destroyed as directed by the LRN reference laboratory when the identification is confirmed.



Agents of Bioterrorism

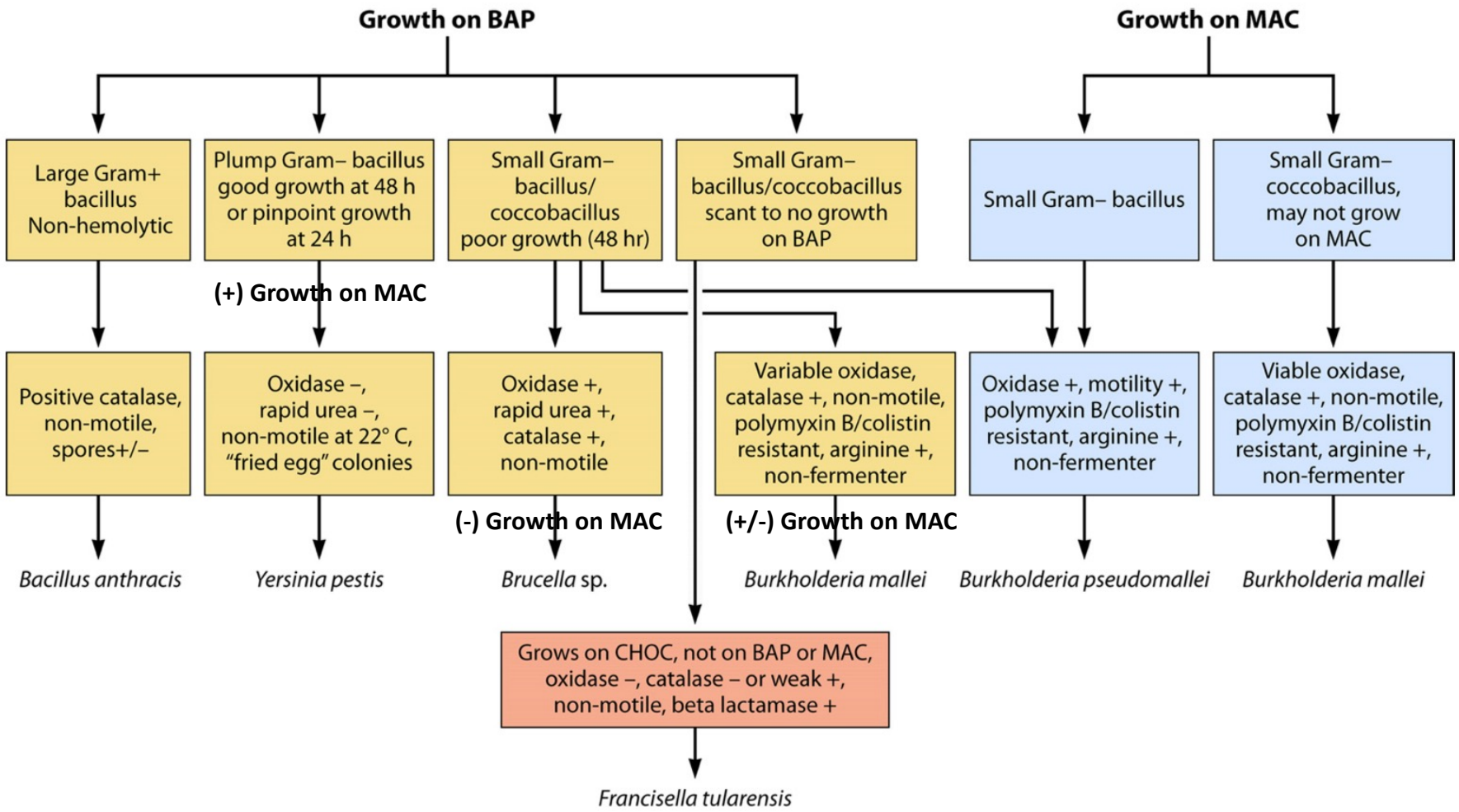
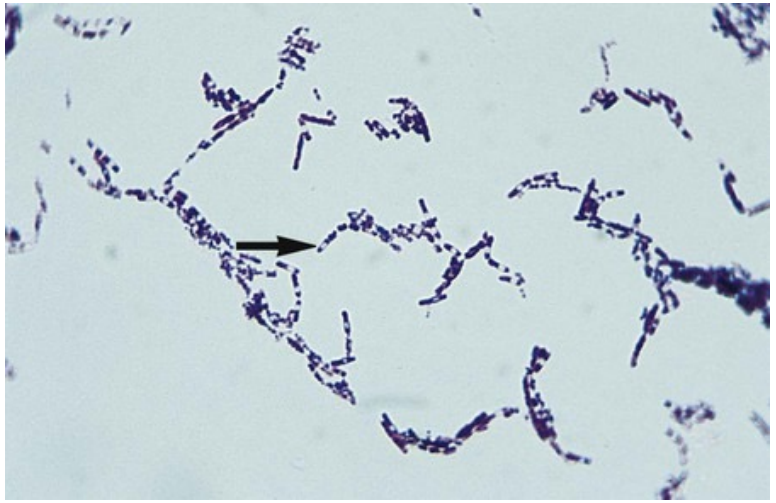
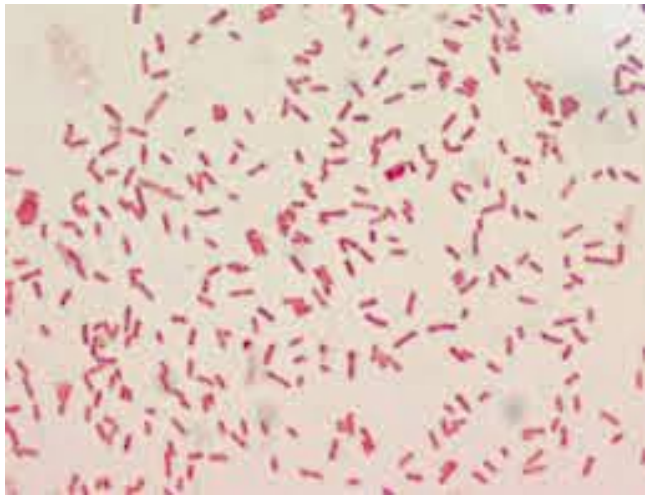


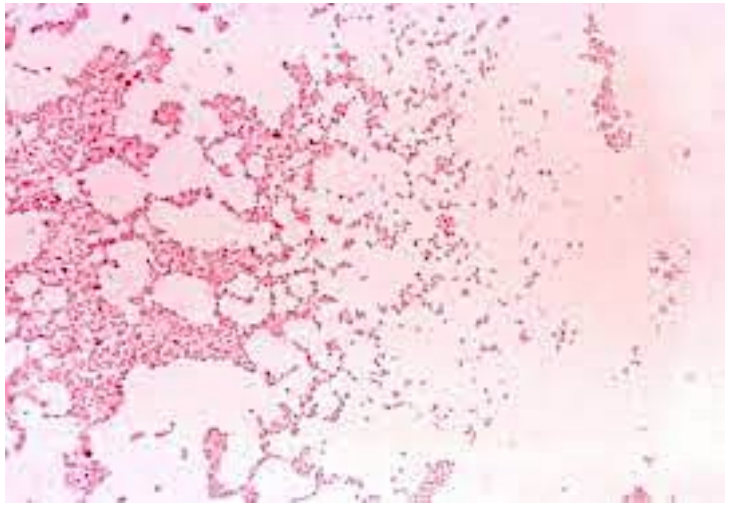
FIG 2 Cultivable bioterrorism agents. BAP, blood agar plate; MAC, MacConkey plate.



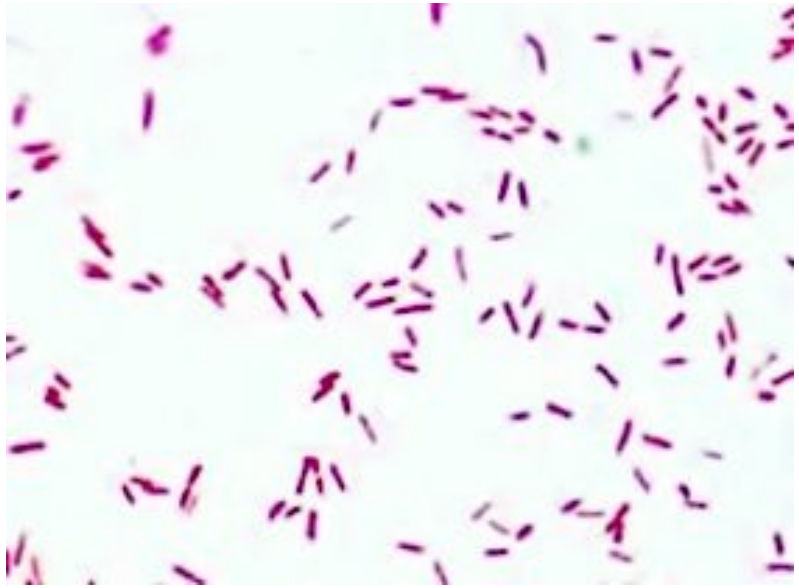
Bacillus anthracis

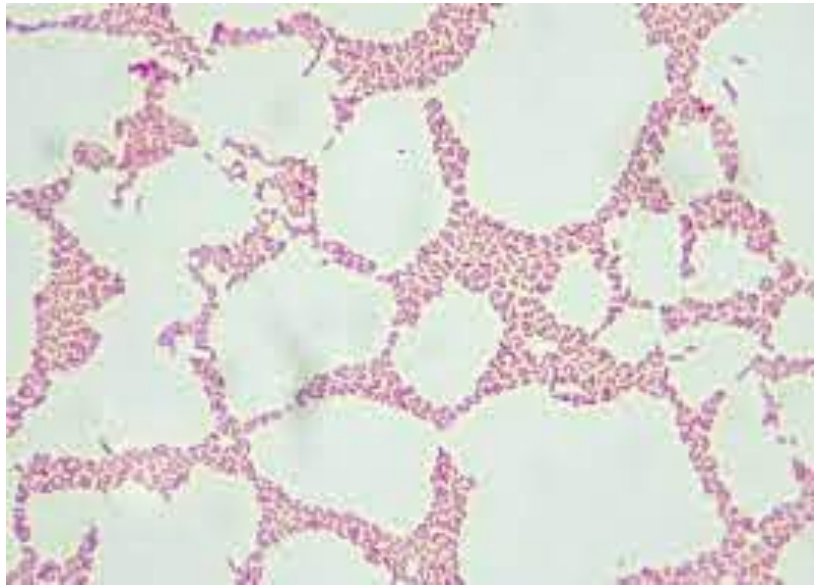


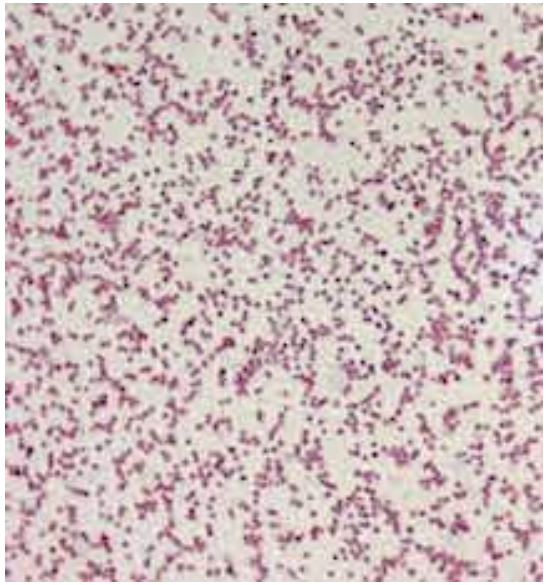
Yersinia pestis



Brucella sp.



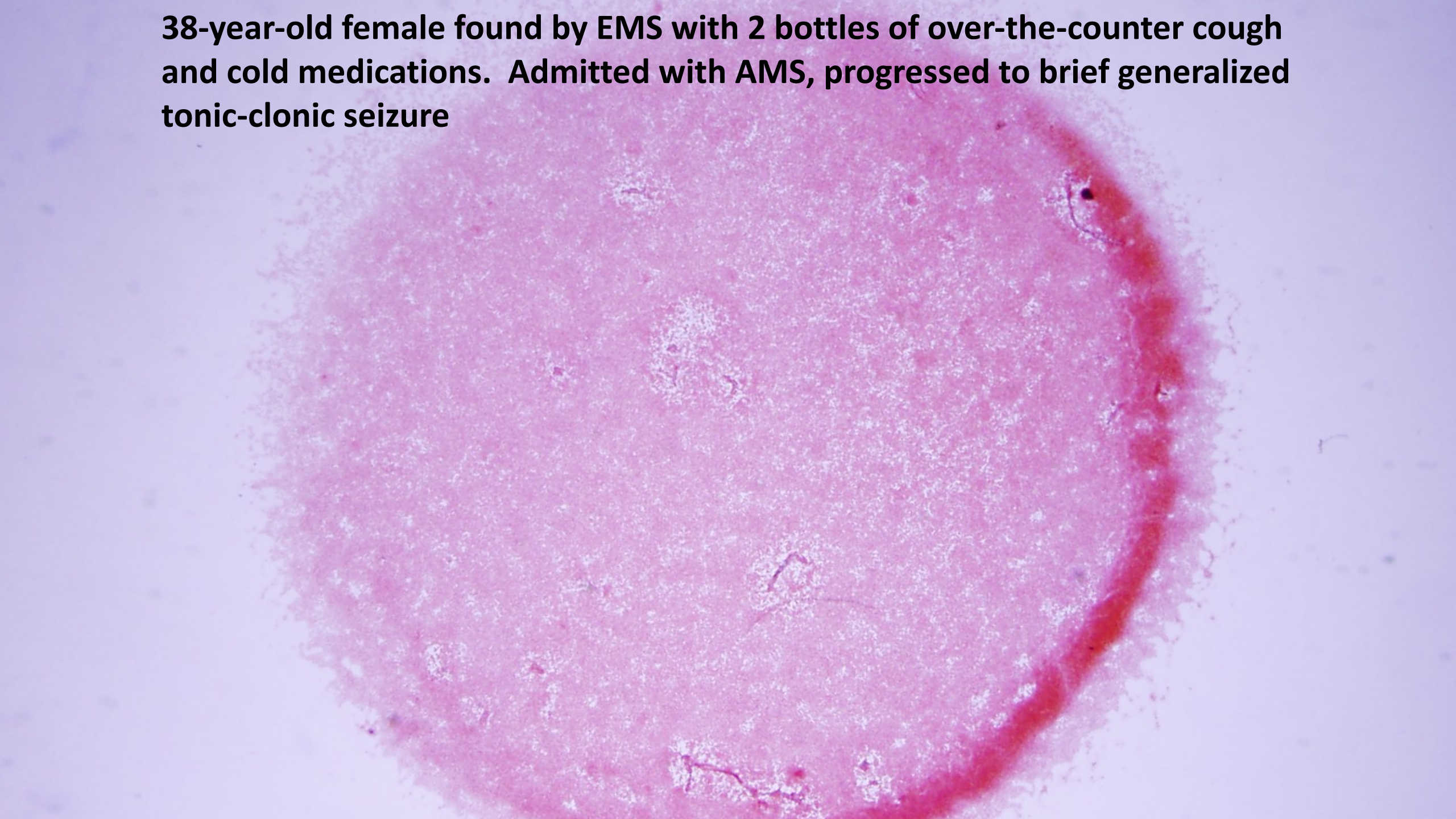


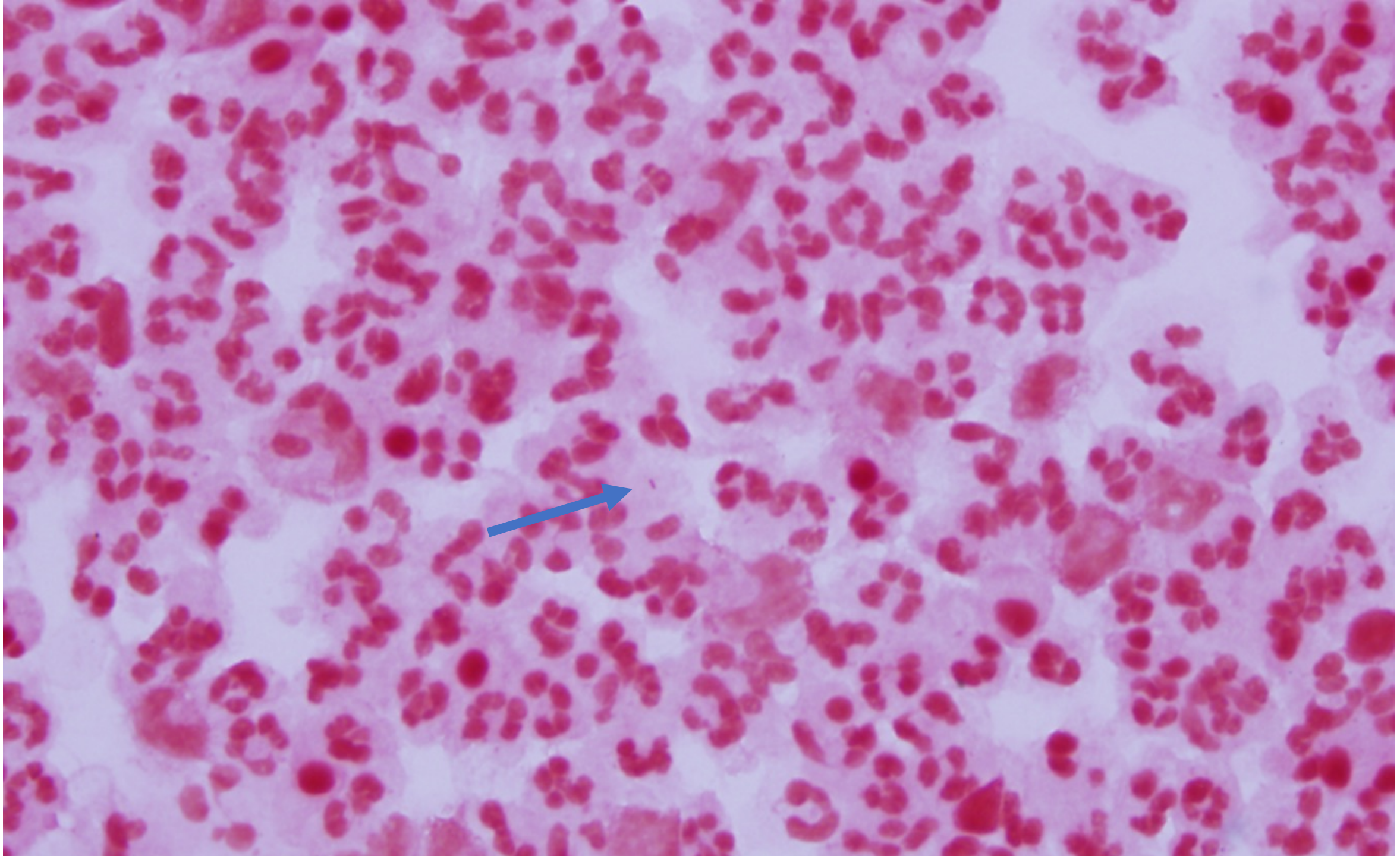


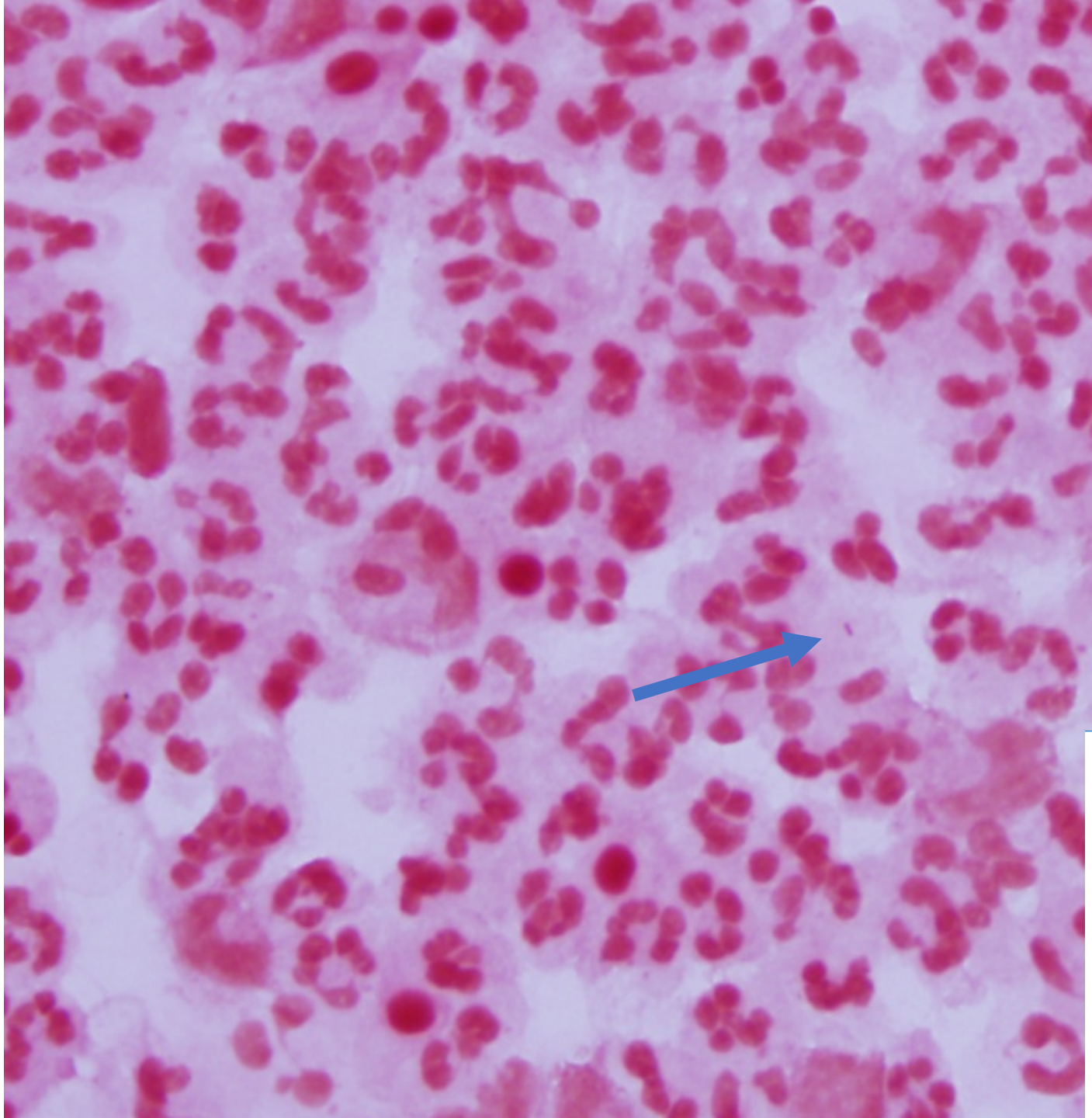
Burkholderia mallei *Burkholderia pseudomallei* *Burkholderia mallei*

Francisella tularensis

38-year-old female found by EMS with 2 bottles of over-the-counter cough and cold medications. Admitted with AMS, progressed to brief generalized tonic-clonic seizure

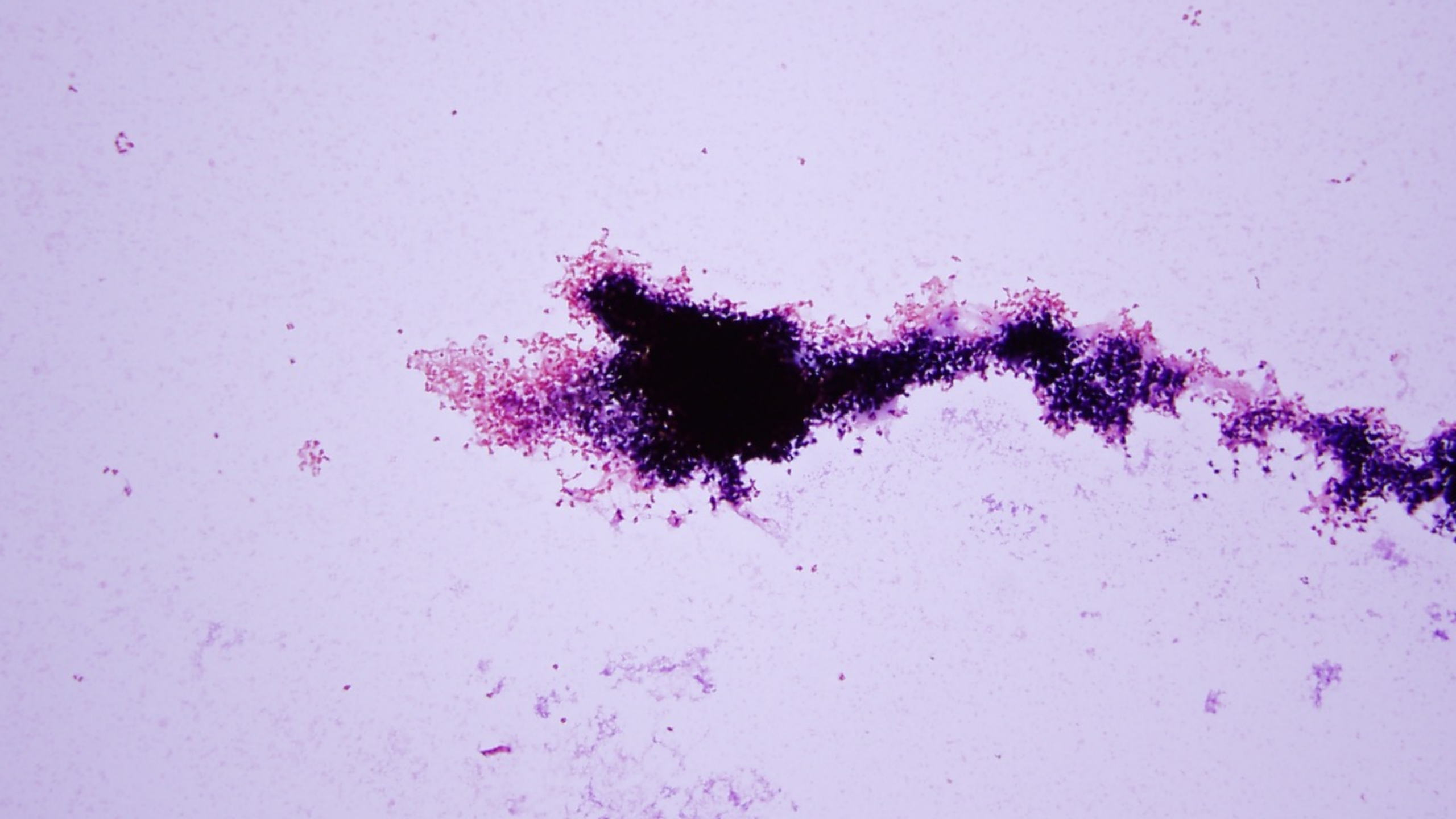


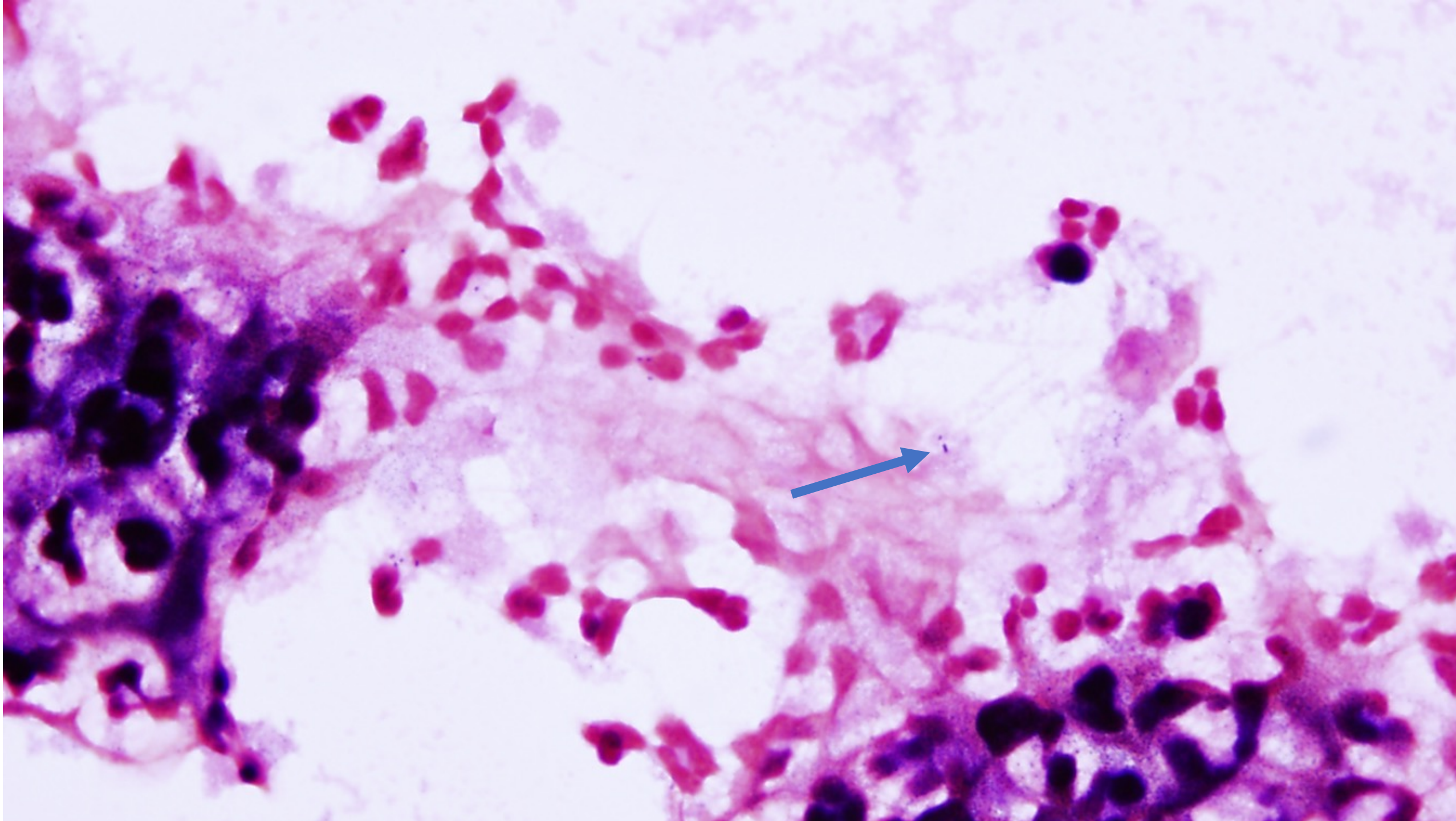


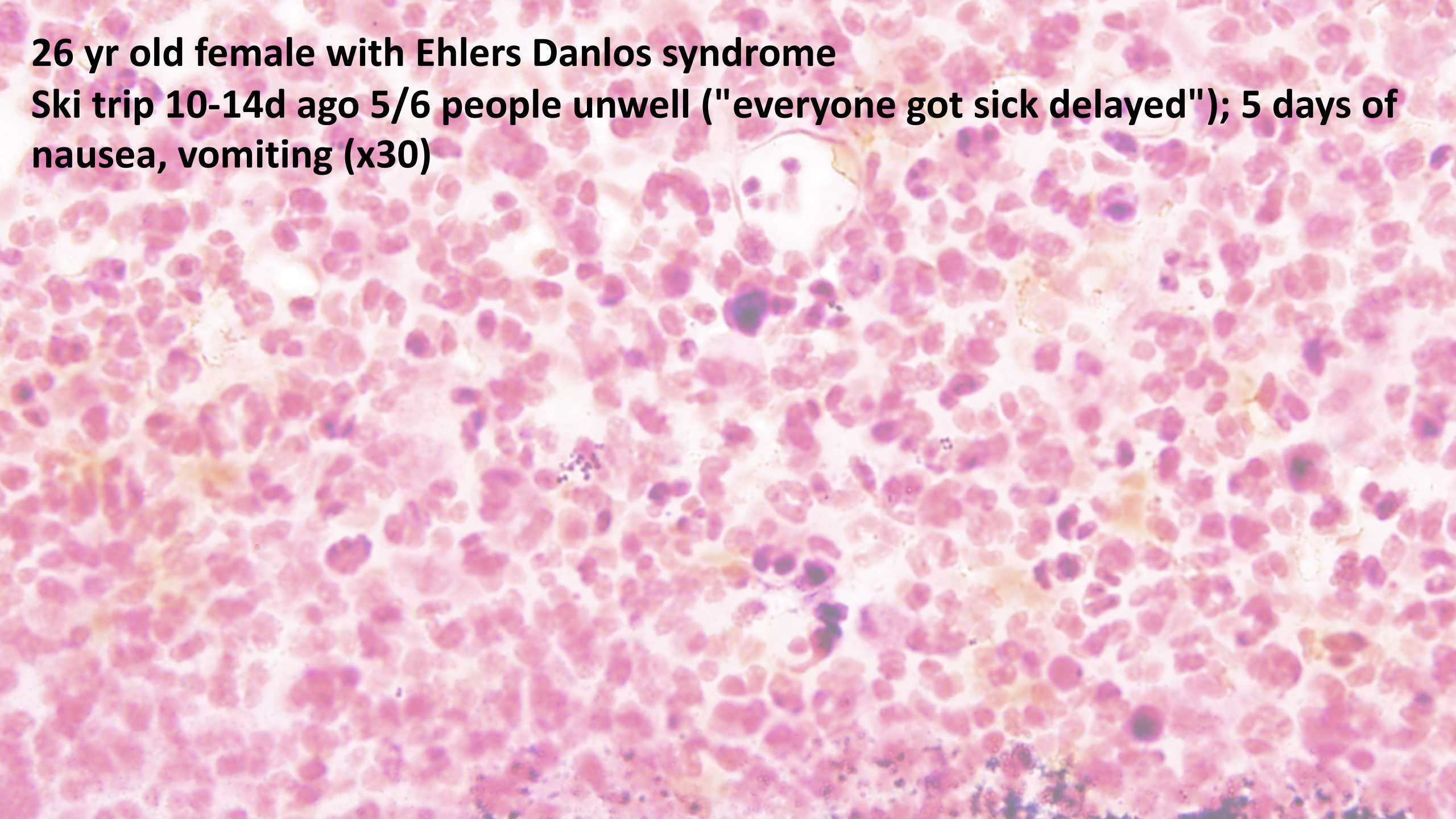


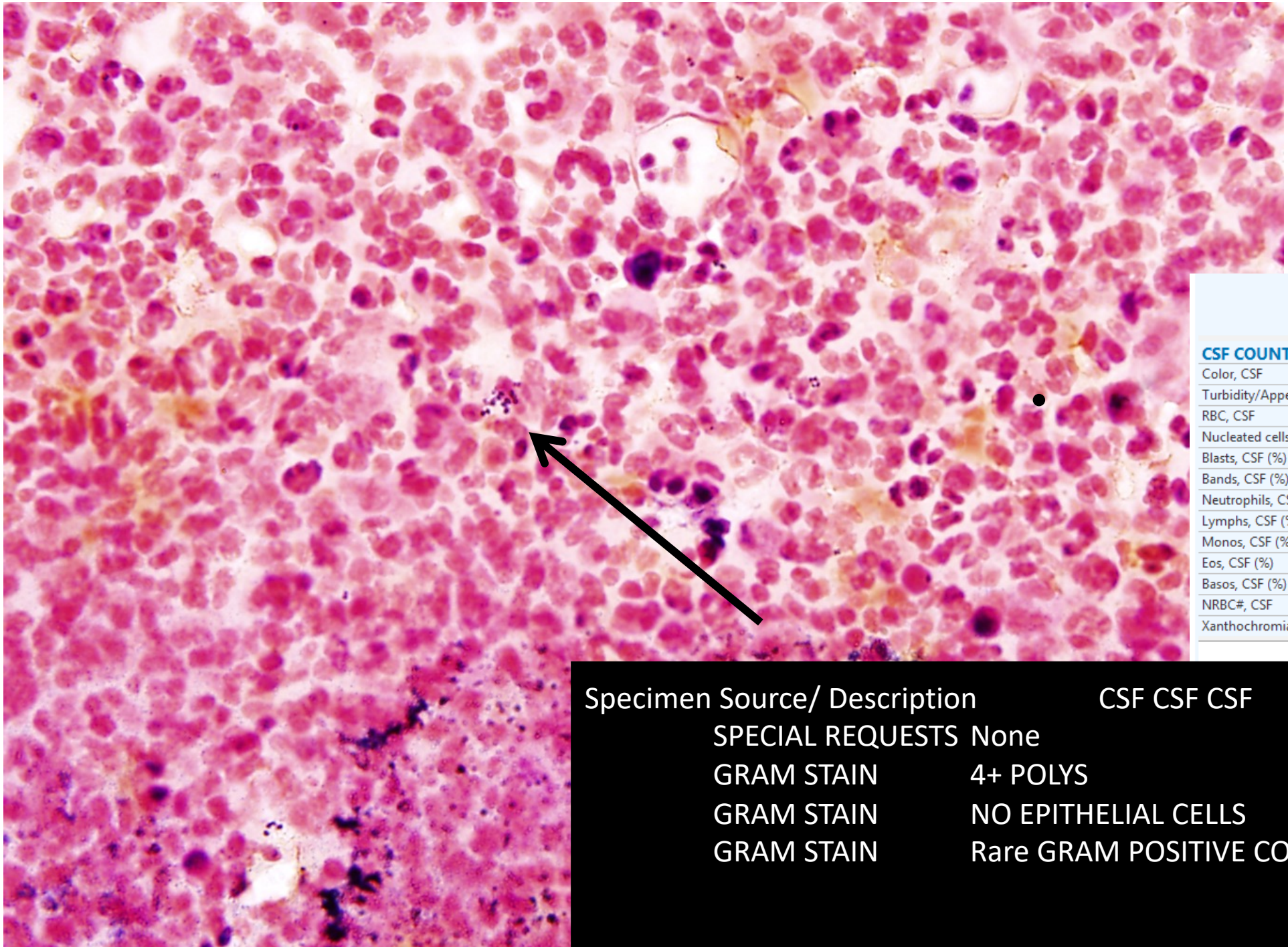
CSF COUNTS AND DIFF				
Color, CSF	Yellow	!	Yellow	!
Turbidity/Appearan...	HAZY	!	HAZY	!
RBC, CSF	460	!	220	!
Nucleated cells, CSF	321 *	!!	156 *	!!
Blasts, CSF (%)	0		0	
Bands, CSF (%)	0		0	
Neutrophils, CSF (%)	95		91	
Lymphs, CSF (%)	5		5	
Monos, CSF (%)	0		4	
Eos, CSF (%)	0		0	
Basos, CSF (%)	0		0	
NRBC#, CSF	0		0	
Xanthochromia, CSF	PRESENT	!	PRESENT	!

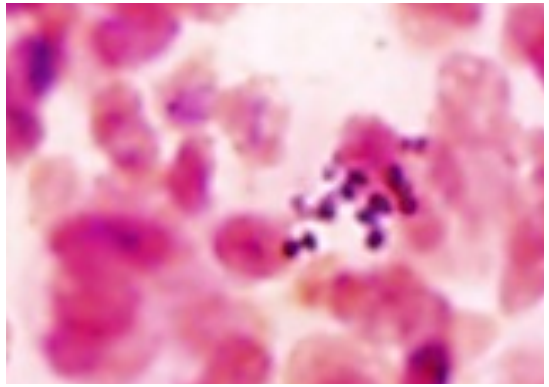
	Ref Range & Units	10/4/18 11:36 PM
ESCHERICHIA COLI K1	Not Detected	Not Detected
H. INFLUENZAE	Not Detected	Not Detected
L. MONOCYTOGENES	Not Detected	Not Detected
N. MENINGITIDIS	Not Detected	Not Detected
S. AGALACTIAE(GRP B)	Not Detected	Not Detected
STREP. PNEUMO	Not Detected	Detected (*)
CYTOMEGALOVIRUS	Not Detected	Not Detected
ENTEROVIRUS	Not Detected	Not Detected
HSV 1	Not Detected	Not Detected
HSV 2	Not Detected	Not Detected
HUMAN HERPESVIRUS 6	Not Detected	Not Detected
HUMAN PARECHOVIRUS	Not Detected	Not Detected
VZV	Not Detected	Not Detected
CRYPTOCOCCUS NEO/GAT	Not Detected	Not Detected






26 yr old female with Ehlers Danlos syndrome
Ski trip 10-14d ago 5/6 people unwell ("everyone got sick delayed"); 5 days of nausea, vomiting (x30)





	2 3/26/2019 2244	1 3/26/2019 2244
CSF COUNTS AND DIFF		
Color, CSF	WHITE !	WHITE !
Turbidity/Appearan...	HAZY !	HAZY !
RBC, CSF	172 !	233 !
Nucleated cells, CSF	17,950 * ⇈	18,600 * ⇈
Blasts, CSF (%)	0	0
Bands, CSF (%)	0	0
Neutrophils, CSF (%)	89 *	91 *
Lymphs, CSF (%)	4	3
Monos, CSF (%)	7	6
Eos, CSF (%)	0	0
Basos, CSF (%)	0	0
NRBC#, CSF	0	0
Xanthochromia, CSF	NOTIFY LAB WITH... !	NOT PRESENT

Specimen Source/ Description CSF CSF CSF

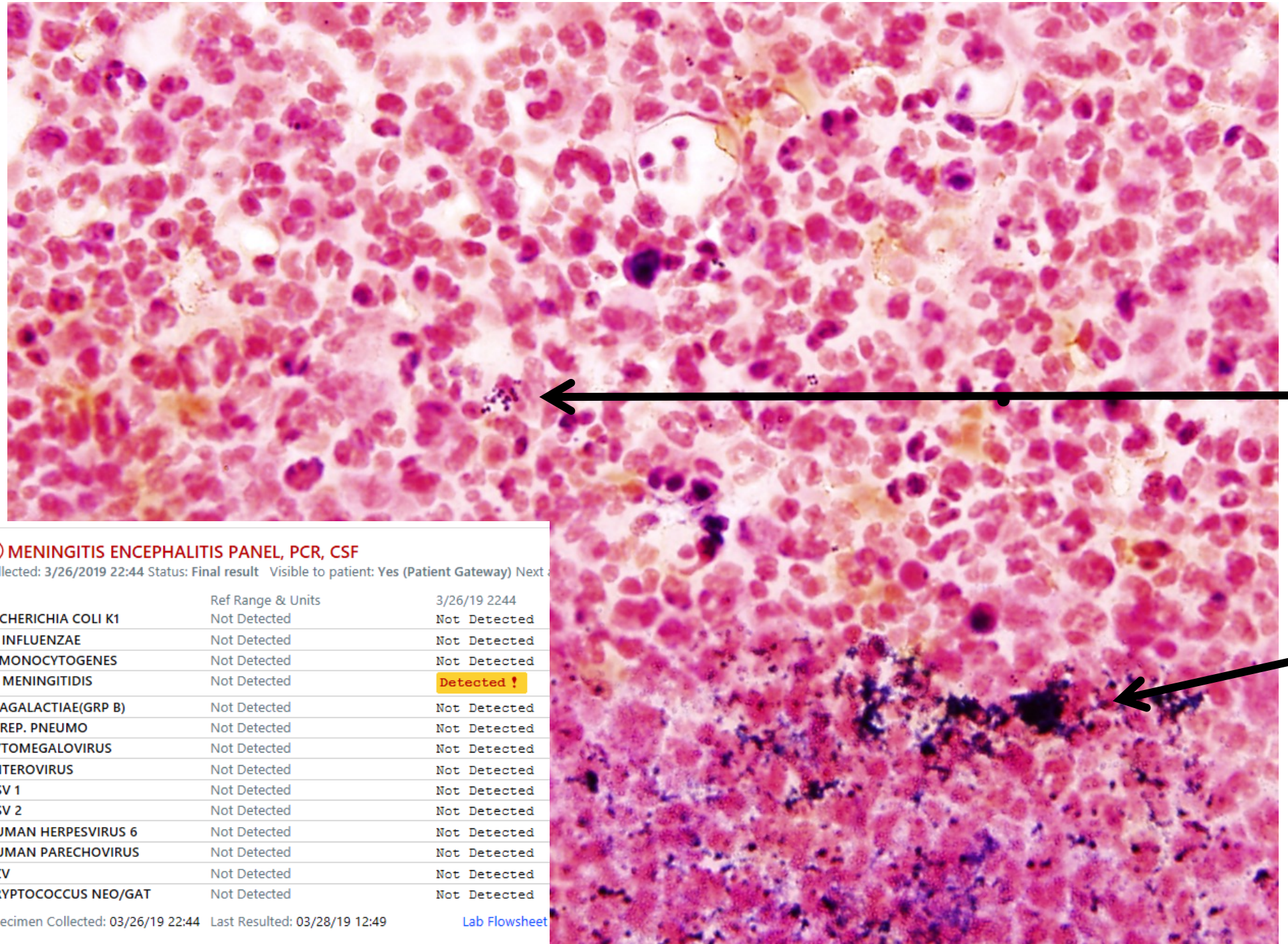
SPECIAL REQUESTS None

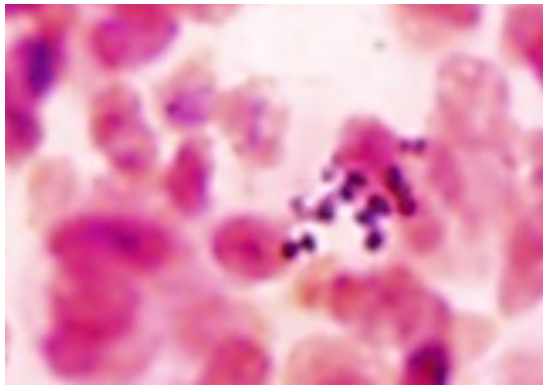
GRAM STAIN 4+ POLYS

GRAM STAIN NO EPITHELIAL CELLS

GRAM STAIN Rare GRAM POSITIVE COCCI in PAIRS and CLUSTERS

Initial gram stain read





"Gram positive cocci"
- good morphology
But underdecolorized
-truly gram negative cocci

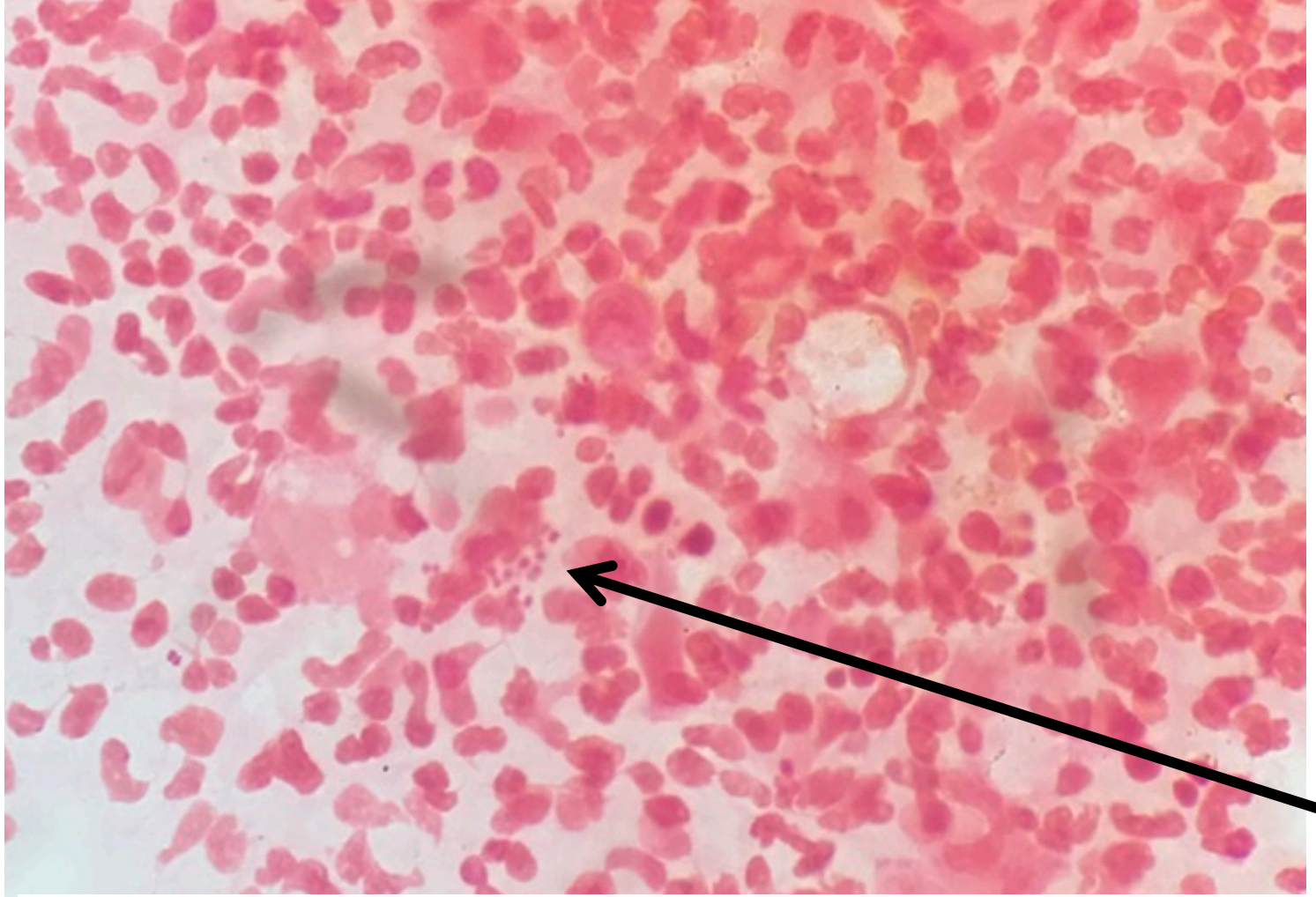
Stain precipitate
Can be confused with
Gram positive cocci

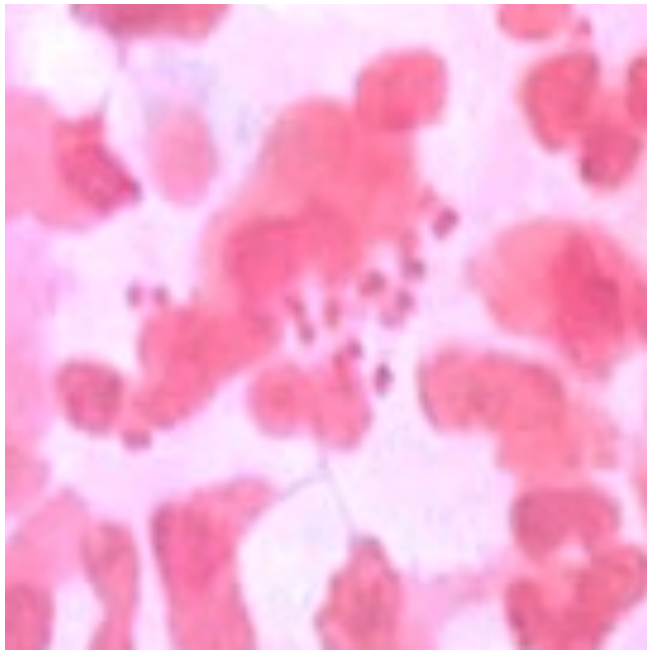
MENINGITIS ENCEPHALITIS PANEL, PCR, CSF

Collected: 3/26/2019 22:44 Status: **Final result** Visible to patient: **Yes (Patient Gateway)** Next

	Ref Range & Units	3/26/19 2244
ESCHERICHIA COLI K1	Not Detected	Not Detected
H. INFLUENZAE	Not Detected	Not Detected
L. MONOCYTOGENES	Not Detected	Not Detected
N. MENINGITIDIS	Not Detected	Detected !
S. AGALACTIAE(GRP B)	Not Detected	Not Detected
STREP. PNEUMO	Not Detected	Not Detected
CYTOMEGALOVIRUS	Not Detected	Not Detected
ENTEROVIRUS	Not Detected	Not Detected
HSV 1	Not Detected	Not Detected
HSV 2	Not Detected	Not Detected
HUMAN HERPESVIRUS 6	Not Detected	Not Detected
HUMAN PARECHOVIRUS	Not Detected	Not Detected
VZV	Not Detected	Not Detected
CRYPTOCOCCUS NEO/GAT	Not Detected	Not Detected

Specimen Collected: 03/26/19 22:44 Last Resulted: 03/28/19 12:49 Lab Flowsheet





Gram negative cocci
Appropriately stained area, good for morphologic assessment

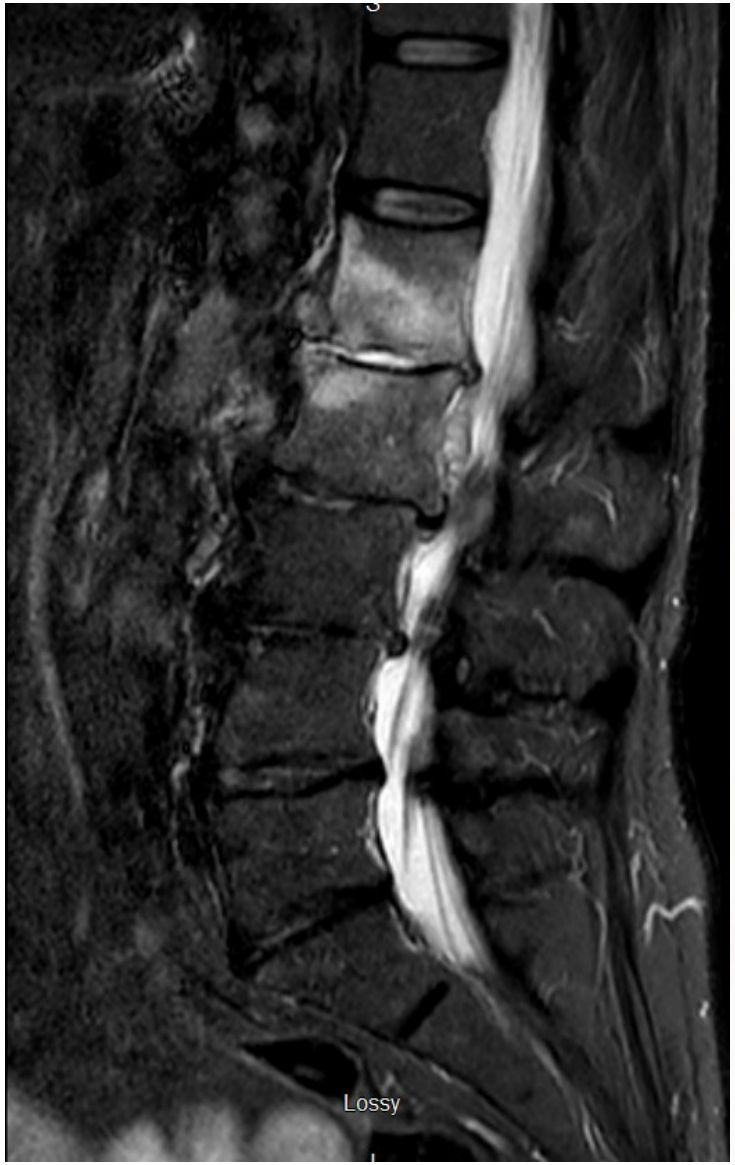
Component	
Specimen Source/ Description	CSF CSF CSF
SPECIAL REQUESTS	None
GRAM STAIN	4+ POLYS
GRAM STAIN	NO EPITHELIAL CELLS
GRAM STAIN	1+ GRAM NEGATIVE DIPLOCOCCI Corrected On: 03/27 AT 1454: Previously Reported as: NO ORGANISMS SEEN Corrected On: 03/27 AT 1422: Previously Reported as: Rare GRAM POSITIVE COCCI in PAIRS and CLUSTERS
GRAM STAIN	CALLED TO Dr Dadabhoy,Farah 30778 3/27/2019 @0027
GRAM STAIN	CORRECTED REPORT CALLED TO Hemen Muleta MD (35599) at 14:28 on 3/27/19
CULTURE / TEST	NO GROWTH 7 DAYS

Gram Stain Paralysis

1. Make a second smear, re-stain
2. Phone a friend
   - ask for a second read
   - Ask for clinical communication

3. Hedge… “gram variable….”

Gram Stain Paralysis



Isolate confirmed as *Brucella spp. by* PCR at Vermont Health Department => CDC *B. suis*

- ***Brown & Brenn (100x)***