Interesting Cases in Infectious Disease Pathology

Tales from the Crypts of the CDC

34th Annual Park City Update

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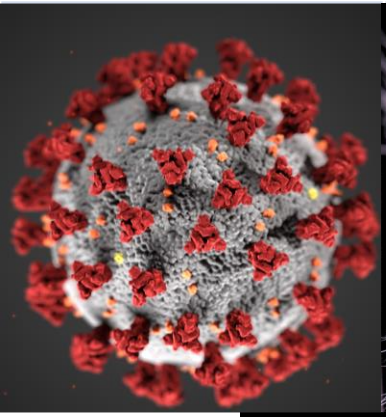
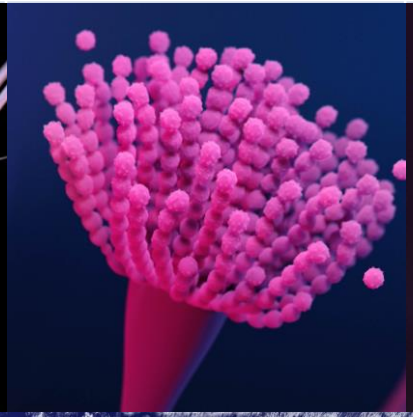
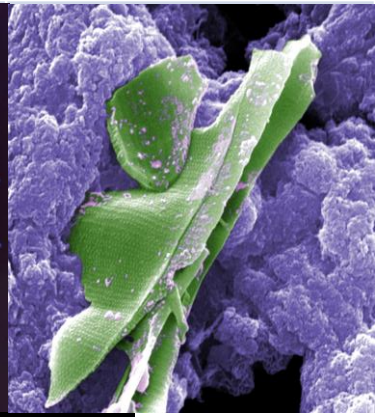
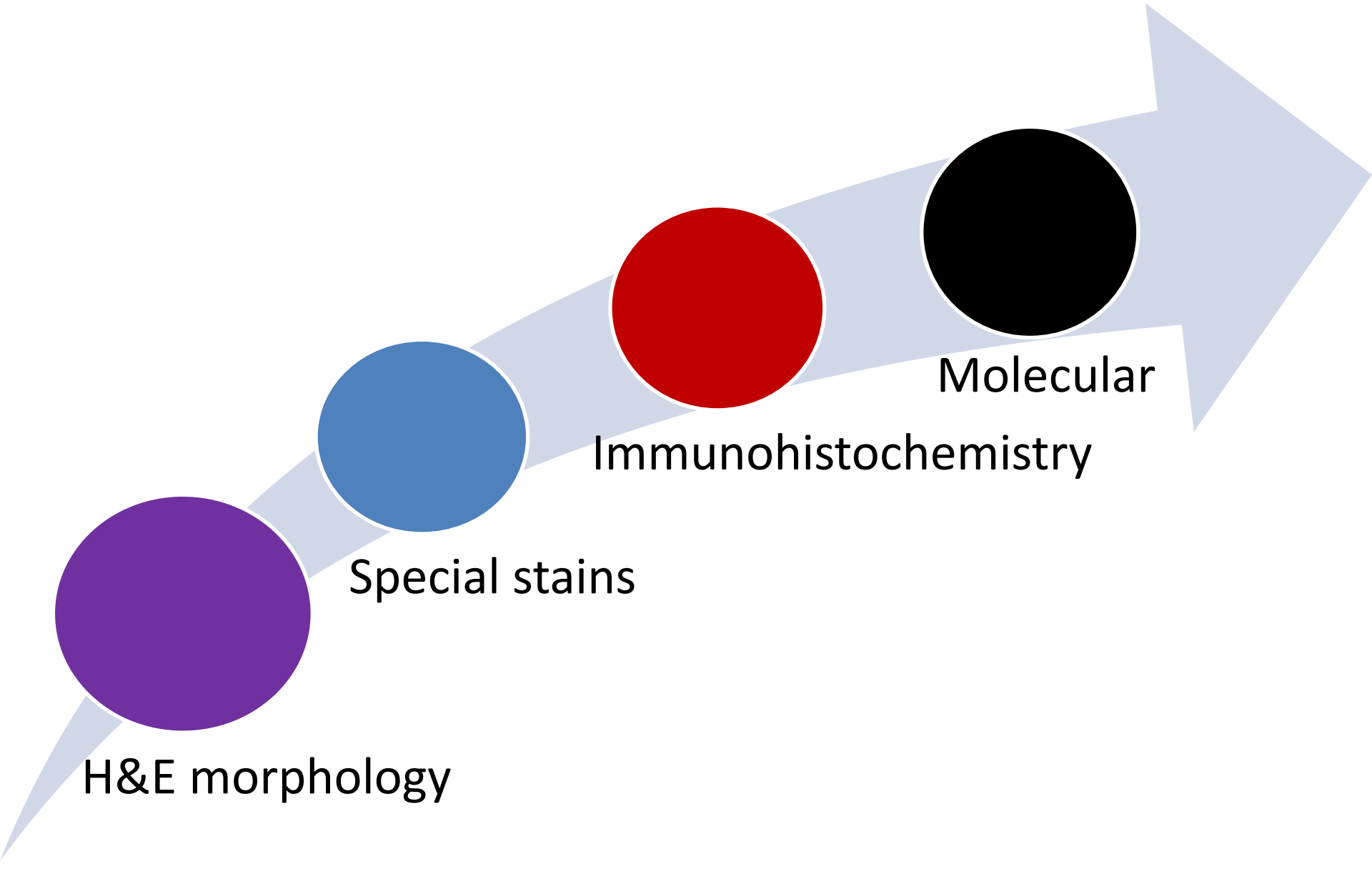
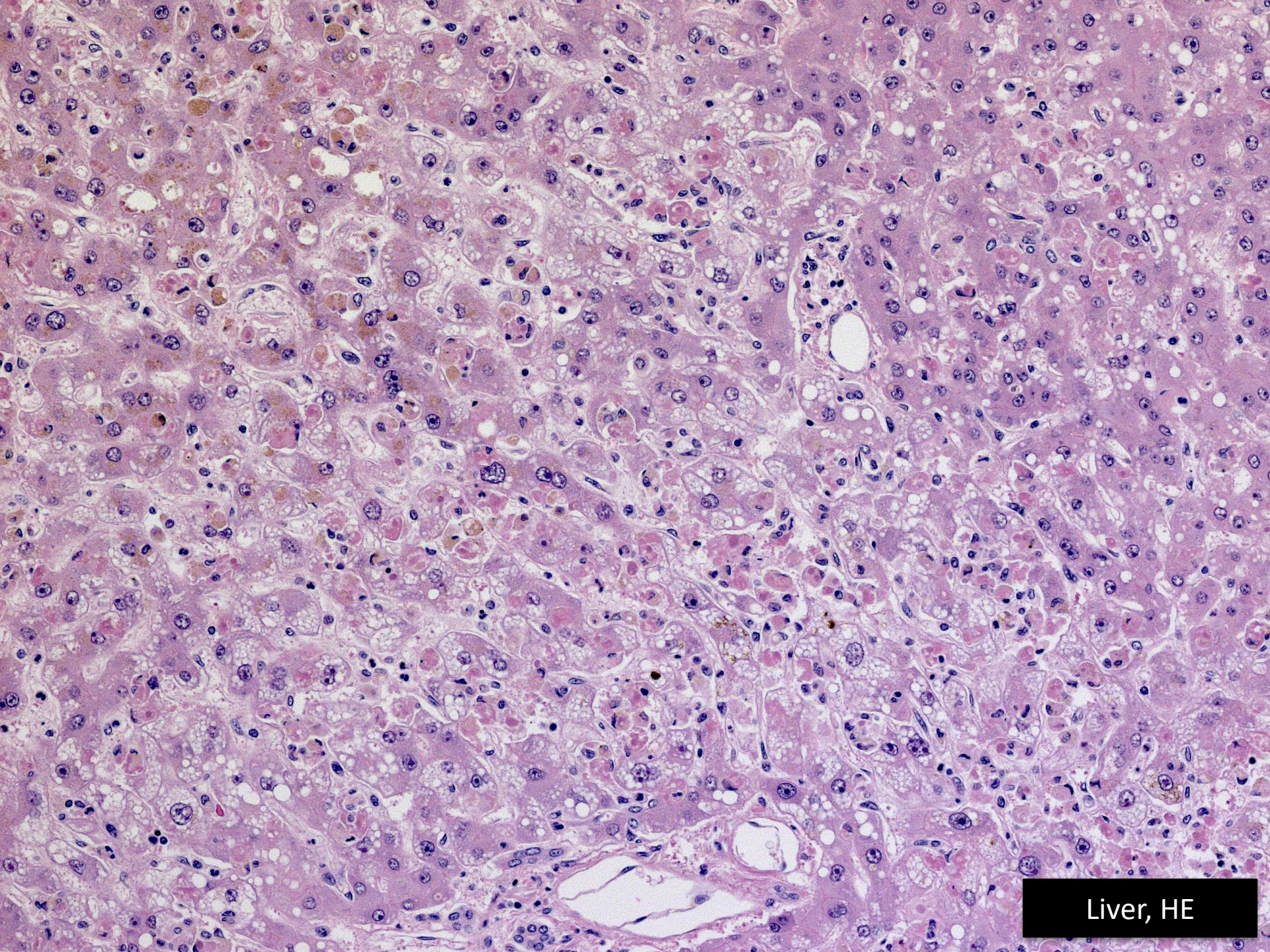
Viruses	Bacteria	Fungi	Parasites	Algae
				

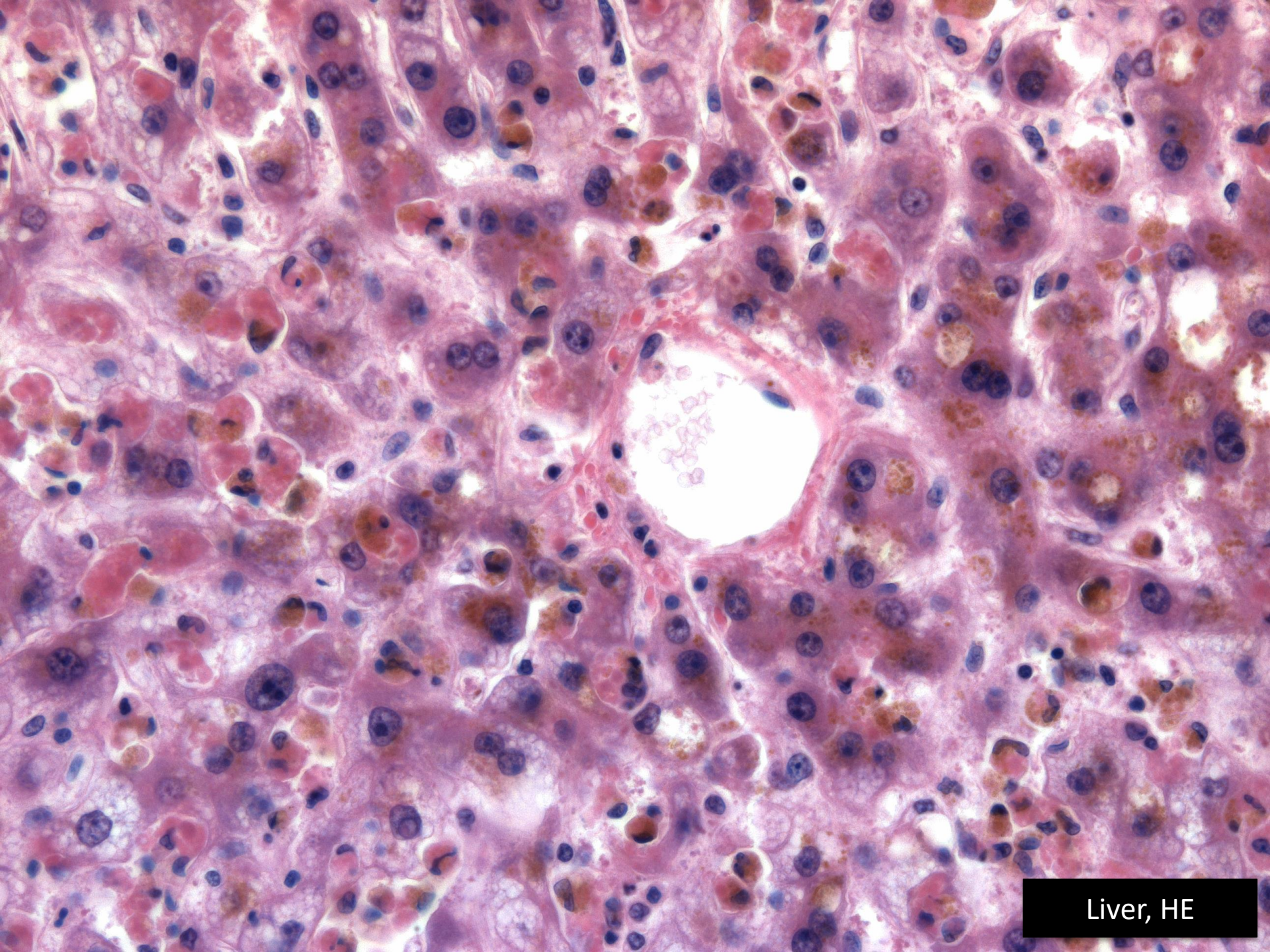
Approach to Diagnosis

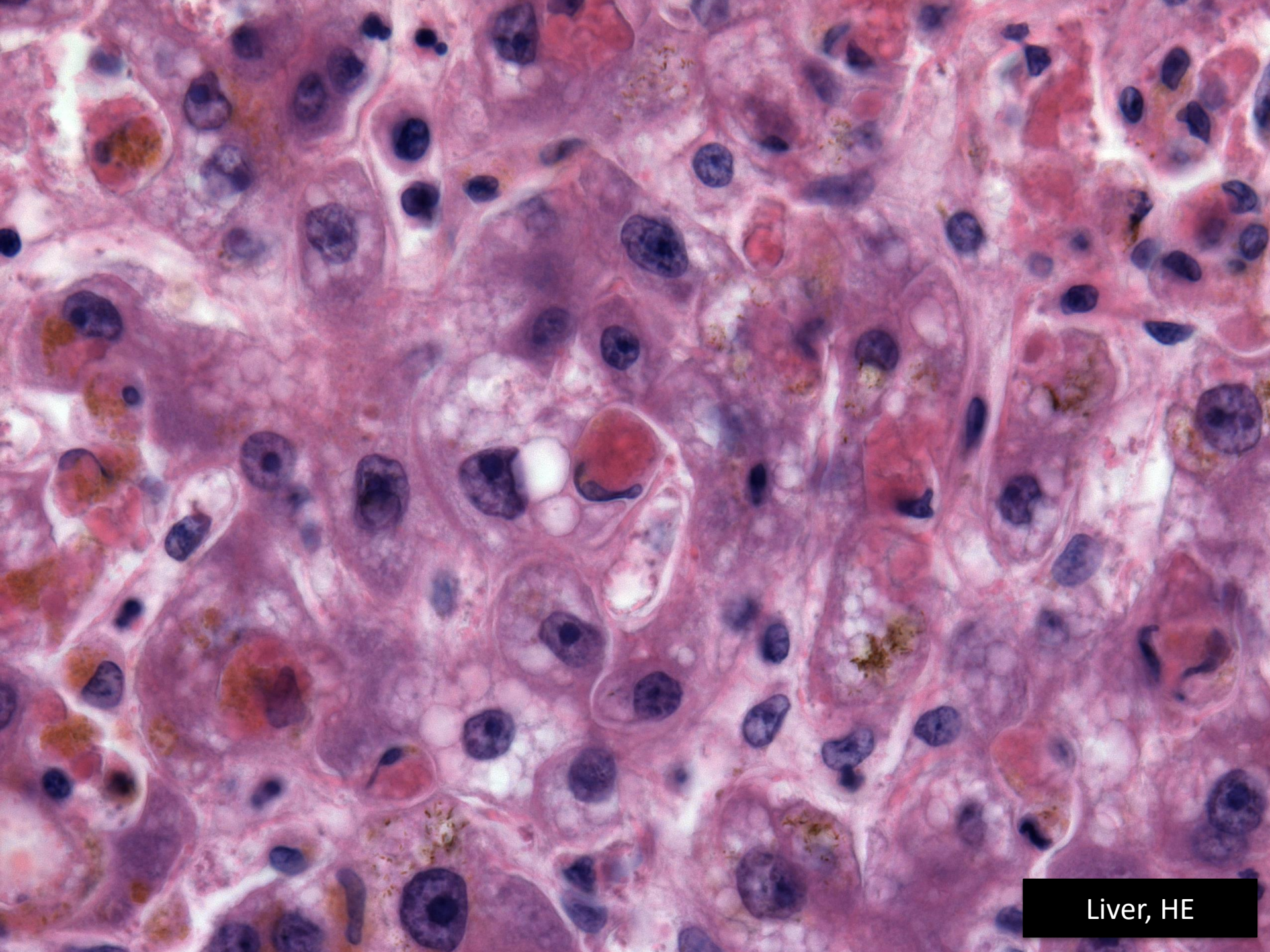


Case 1

- 74 yo M from NY
- Traveled to Peru and Amazon rainforest
- Developed fever, vomiting, malaise, and anorexia on the trip
- Symptoms persisted; he returned to NY where he was hospitalized
- Died 3 days after his return to the U.S. with jaundice, hepatic failure, renal failure, and disseminated intravascular coagulation
- Vaccination history unknown


Liver, HE

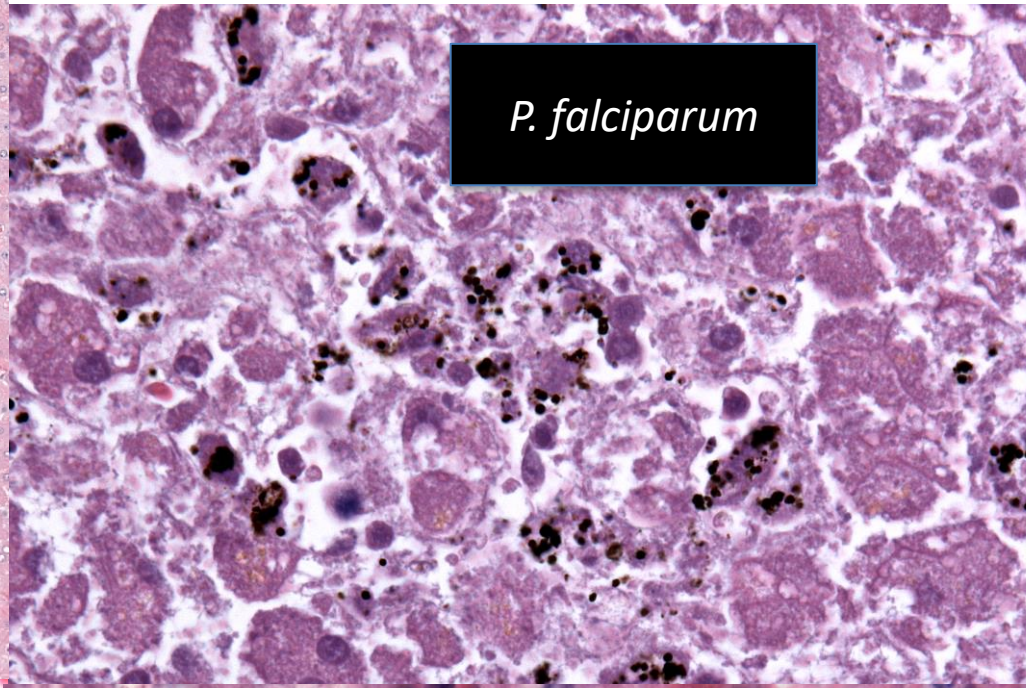

Liver, HE


Liver, HE

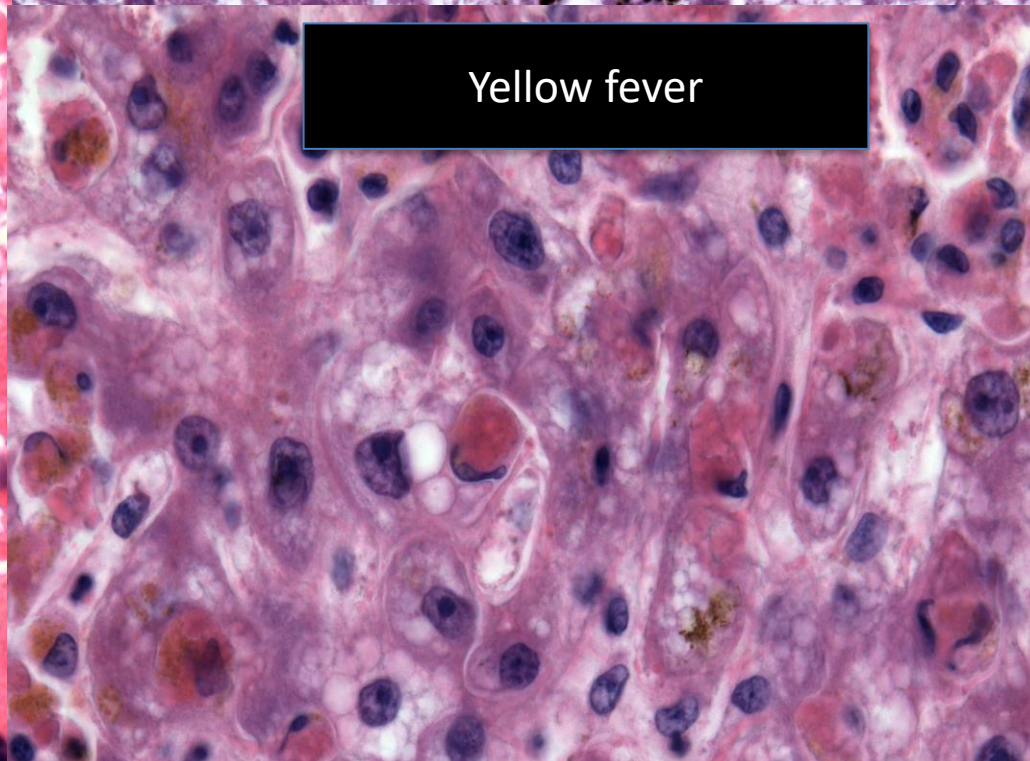
Case 1 Workup

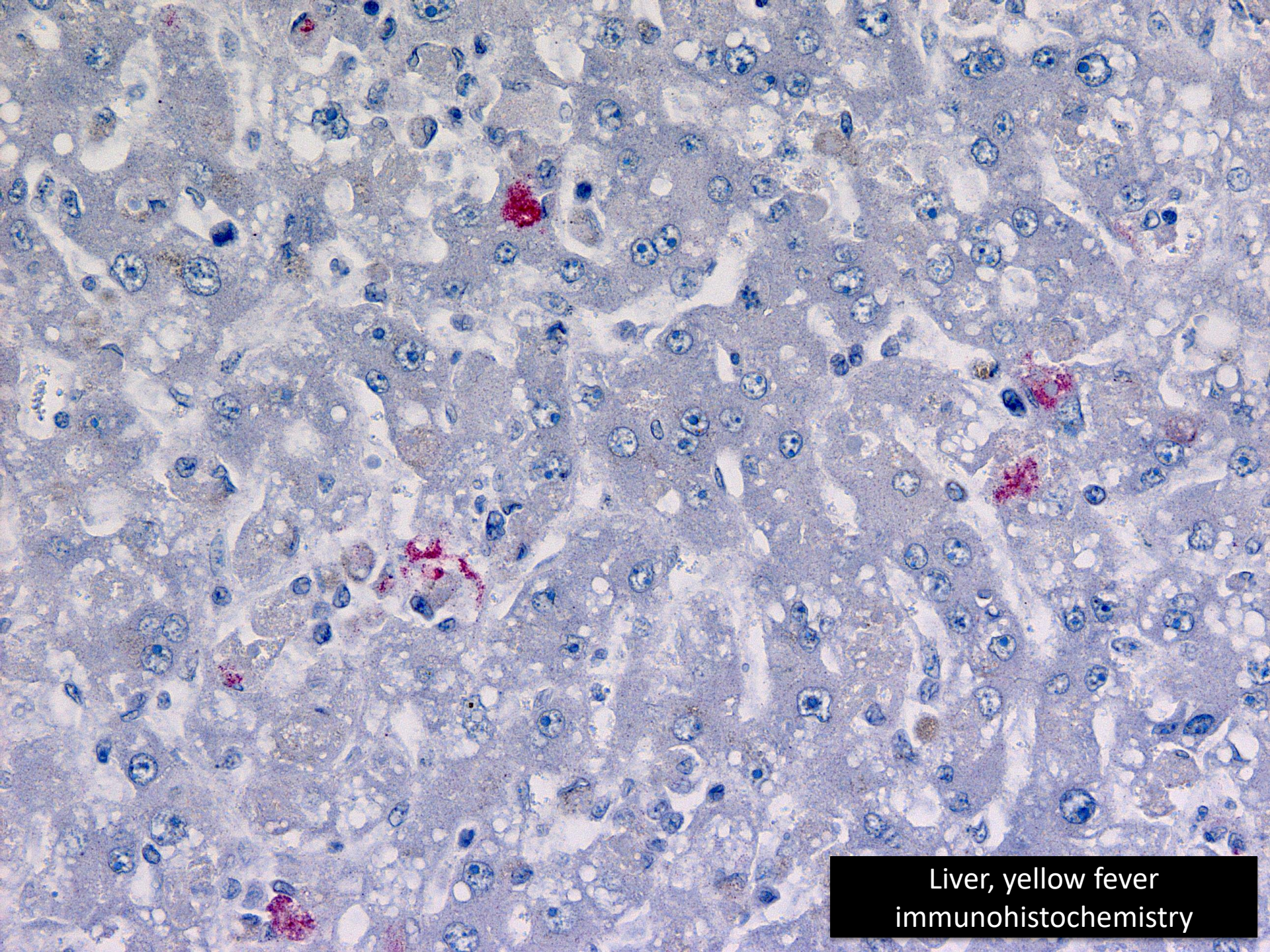
- **Yellow fever virus**
  Midzonal necrosis, Councilman-Rocha Lima bodies, rarely Torres bodies
- **Dengue virus**
  Hepatocyte necrosis, Councilman-Rocha Lima bodies, hemorrhage
- **Malaria**
  Parasitized RBCs, Hemozoin pigment
- **Human herpesviruses (VZV)**
  Characteristic inclusions
- **Viral hepatitis (A, B, C, E)**
  Evaluate clinically
- **Toxin**
  Exposure history

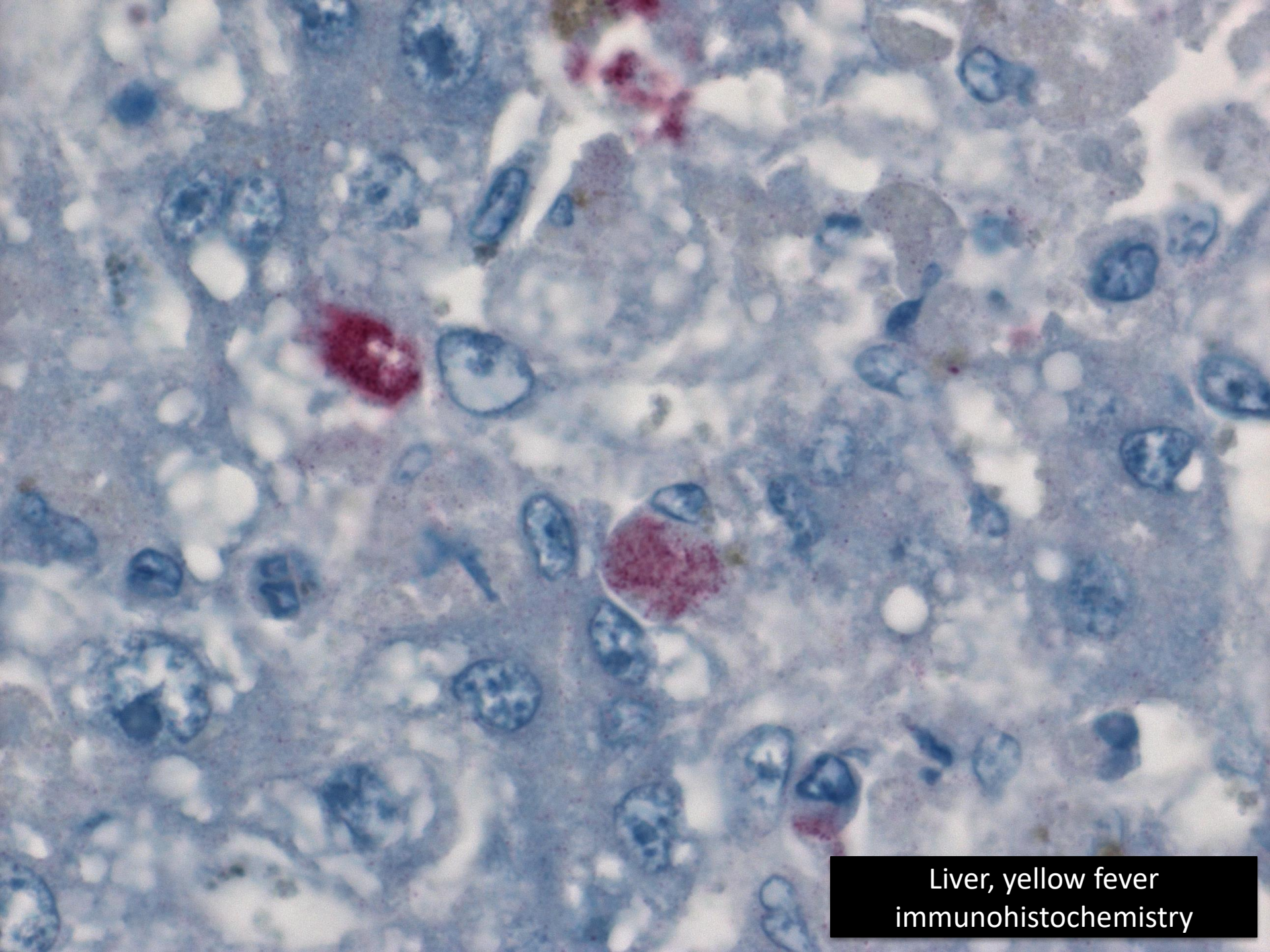

Dengue virus


P. falciparum


Varicella zoster virus


Yellow fever


Liver, yellow fever
immunohistochemistry



Liver, yellow fever
immunohistochemistry

Yellow Fever

Disease	Yellow Fever
Vector	Mosquito
Epidemiology	Sub-Saharan Africa, tropical areas of South America 200,000 cases, 30,000 deaths annually **Imported infections:** - **1970 - 2015:** 11 cases reported in U.S. and Europe, 8 patients died (73%) - **2016 - mid 2018: More than 35 travel-associated cases reported in unvaccinated travelers who were residents of nonendemic areas (*Source: CDC, Traveler's Health*)**
Clinical symptoms	Variable: mild to fulminant fatal infections Abrupt onset fever, HA, chills, nausea; remission with recovery; or fever vomiting, jaundice, renal failure, hemorrhage
Pathology	Classically mid-zonal necrosis with apoptotic hepatocytes, Councilman-Rocha Lima bodies
Diagnosis	PCR, serology, IHC
Treatment	Supportive; prevention is key through vaccination

Was the patient vaccinated for yellow fever?

Yellow Fever

wild type or vaccine-associated?

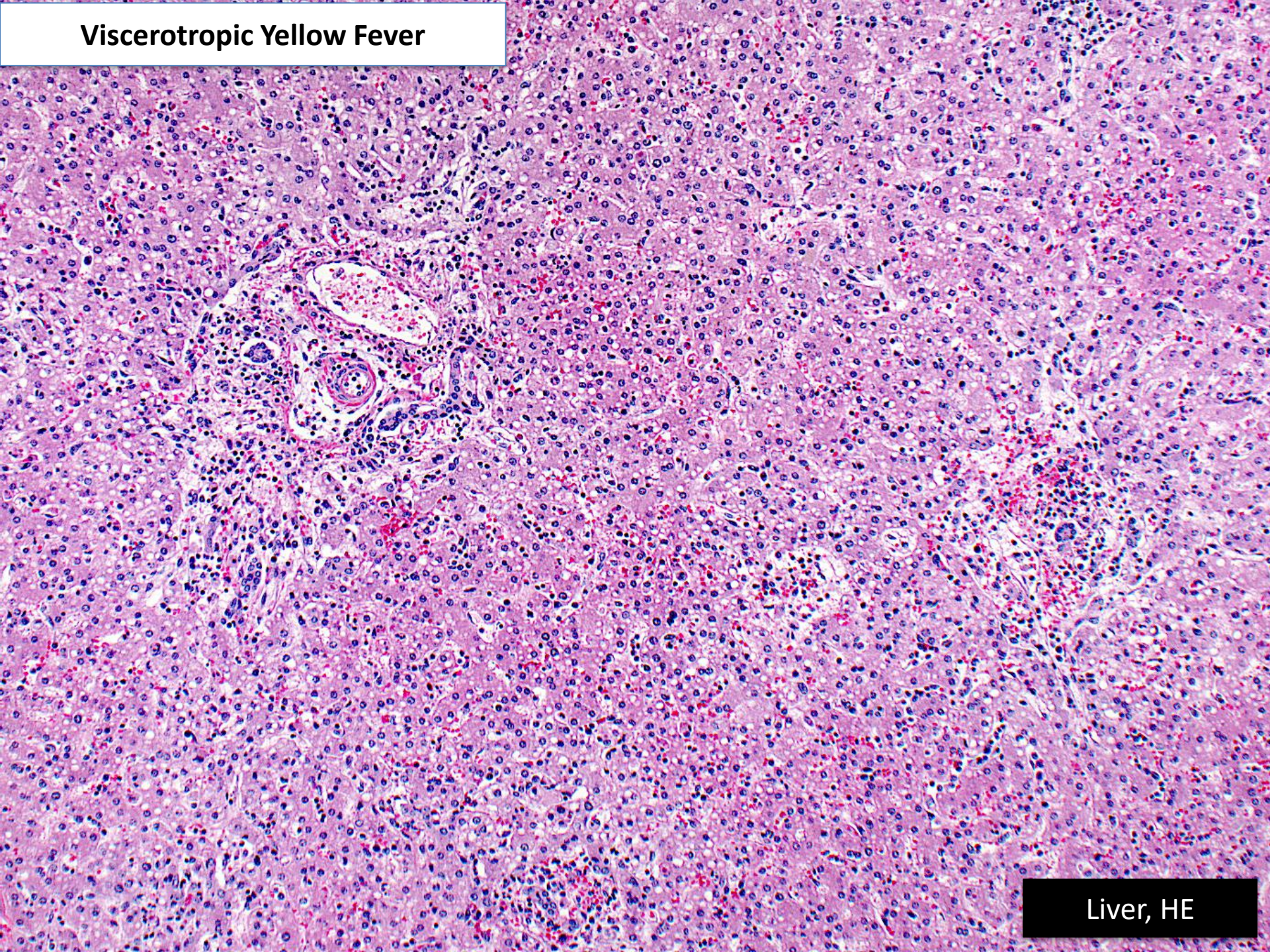
Vaccine-associated Viscerotropic Disease

- Current vaccines contain live-attenuated virus
  - 17D strain of virus contained in the vaccine protects against all strains of yellow fever virus circulating in nature
- Rare adverse event following vaccination
- Vaccine virus proliferates throughout multiple organs

Pathology: <u>Portal based mononuclear inflammation</u> predominates in liver, and multiple organs involved in inflammatory response

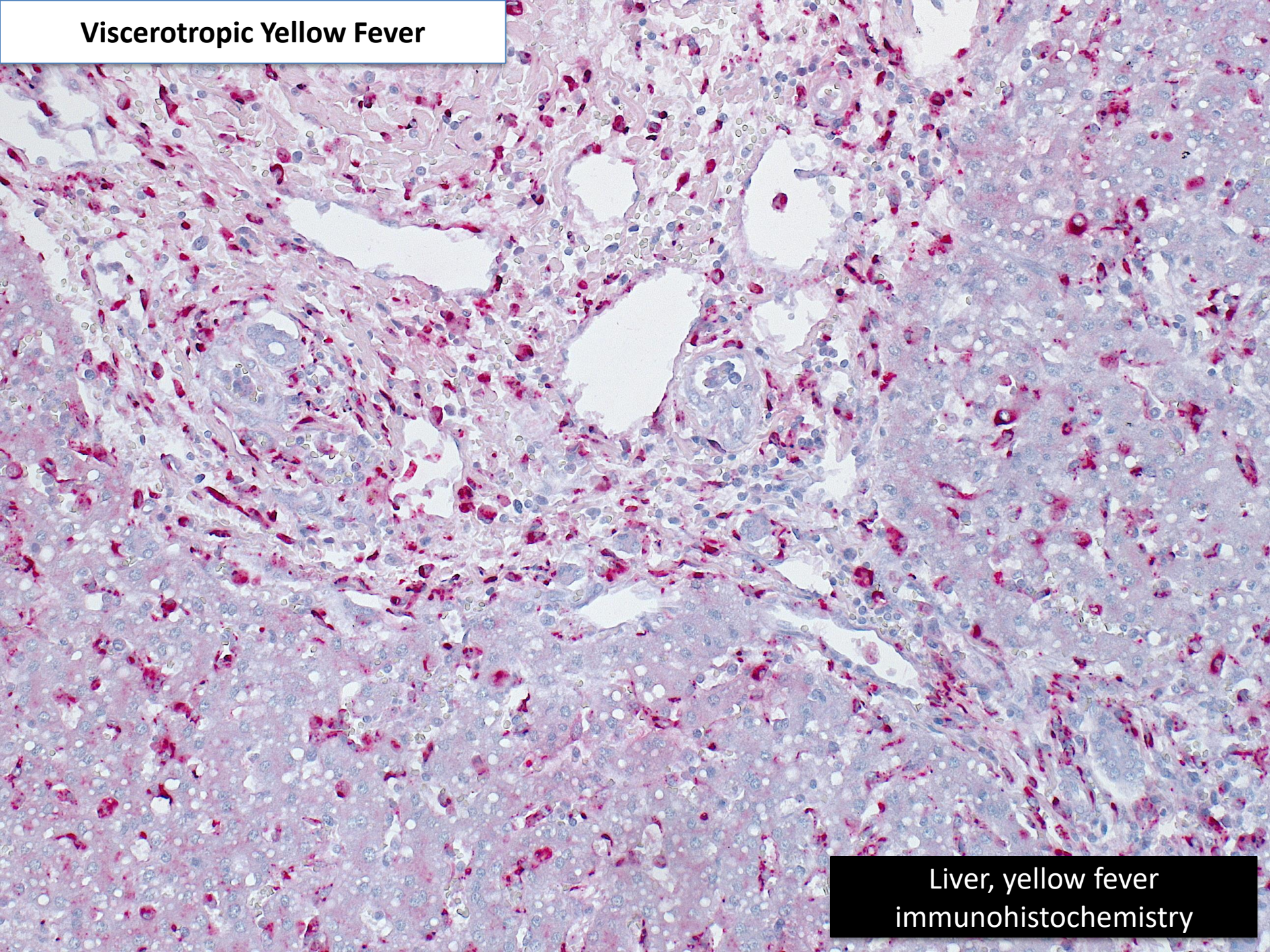
IHC: YF viral antigens <u>portal based within mesenchymal cells</u> and Kupffer cells

Viscerotropic Yellow Fever



Liver, HE

Viscerotropic Yellow Fever



Liver, yellow fever immunohistochemistry

Case 1 Summary

- Immunohistochemical evidence of yellow fever (**limited to the liver**)
- PCR testing on RNA extracted from the paraffin-embedded tissue block confirmed infection by **wild type yellow fever virus**
- **Increasing risk of imported cases**

Areas with Risk of Yellow Fever Virus Transmission in South America

CDC Warns Of Deadly Outbreak of Yellow Fever in Brazil

Travelers to Brazil should get Yellow Fever vaccine

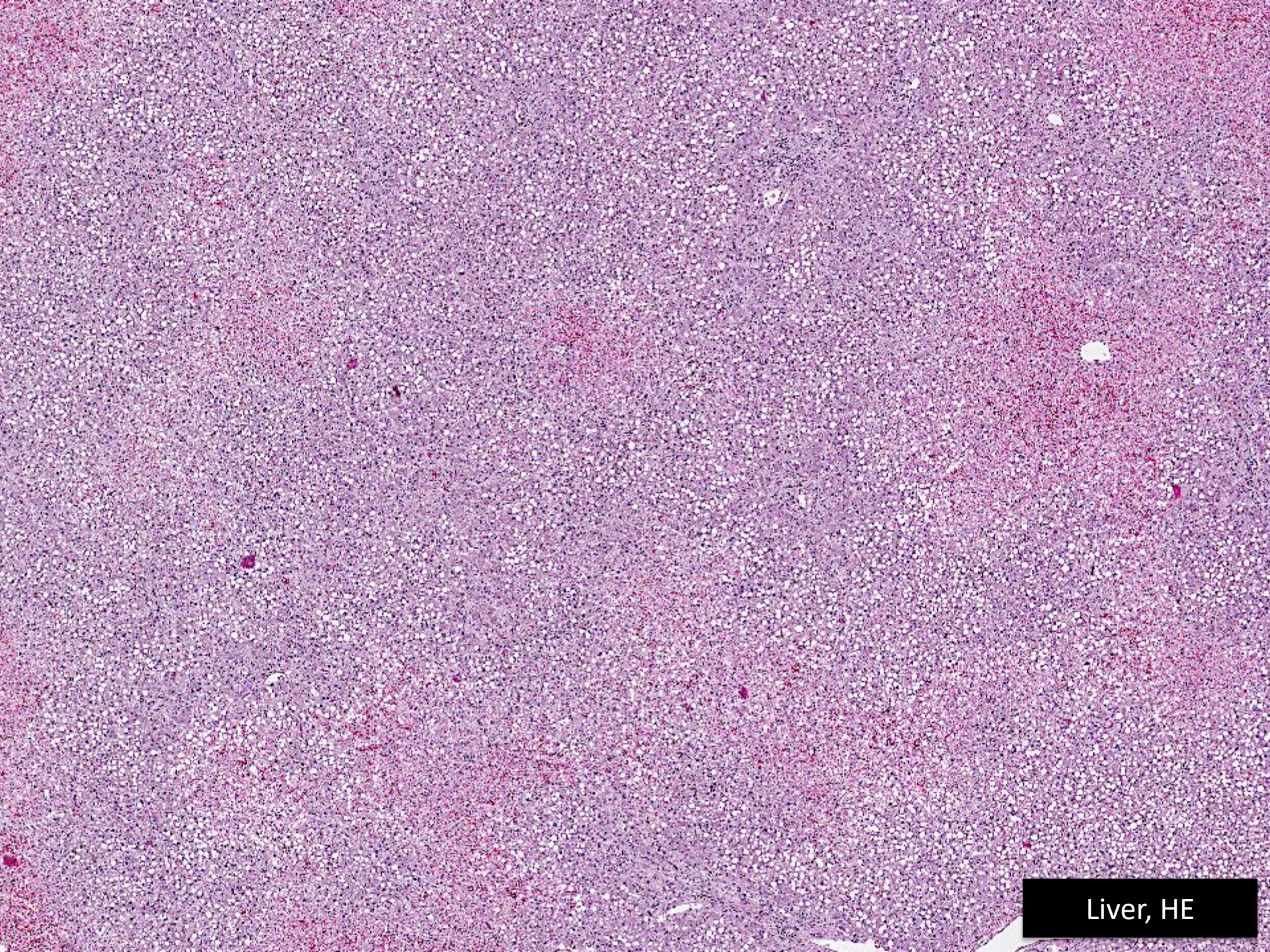
Media Statement
For Immediate Release: Friday, March 16, 2018
Contact: Media Relations,
(404) 639-3286

In response to a large, ongoing outbreak of yellow fever in multiple states of Brazil, including near large urban areas and popular tourist destinations, CDC is recommending travelers to the country protect themselves from yellow fever by getting the yellow fever vaccine at least 10 days before travel, and taking steps to prevent mosquito bites during their travel.

CDC recommends that people who are unable to get yellow fever vaccine or aren't recommended to get it should avoid traveling to areas of Brazil where yellow fever vaccination is recommended**.** Travelers going to areas with ongoing outbreaks may consider getting a booster dose of yellow fever vaccine if it has been 10 or more years since they were vaccinated.

Case 2

- 47-year-old F from Spain
- Presents with fever, myalgias, nausea and vomiting
- Admitted to the hospital and required mechanical ventilation
- Developed multiple organ failure and died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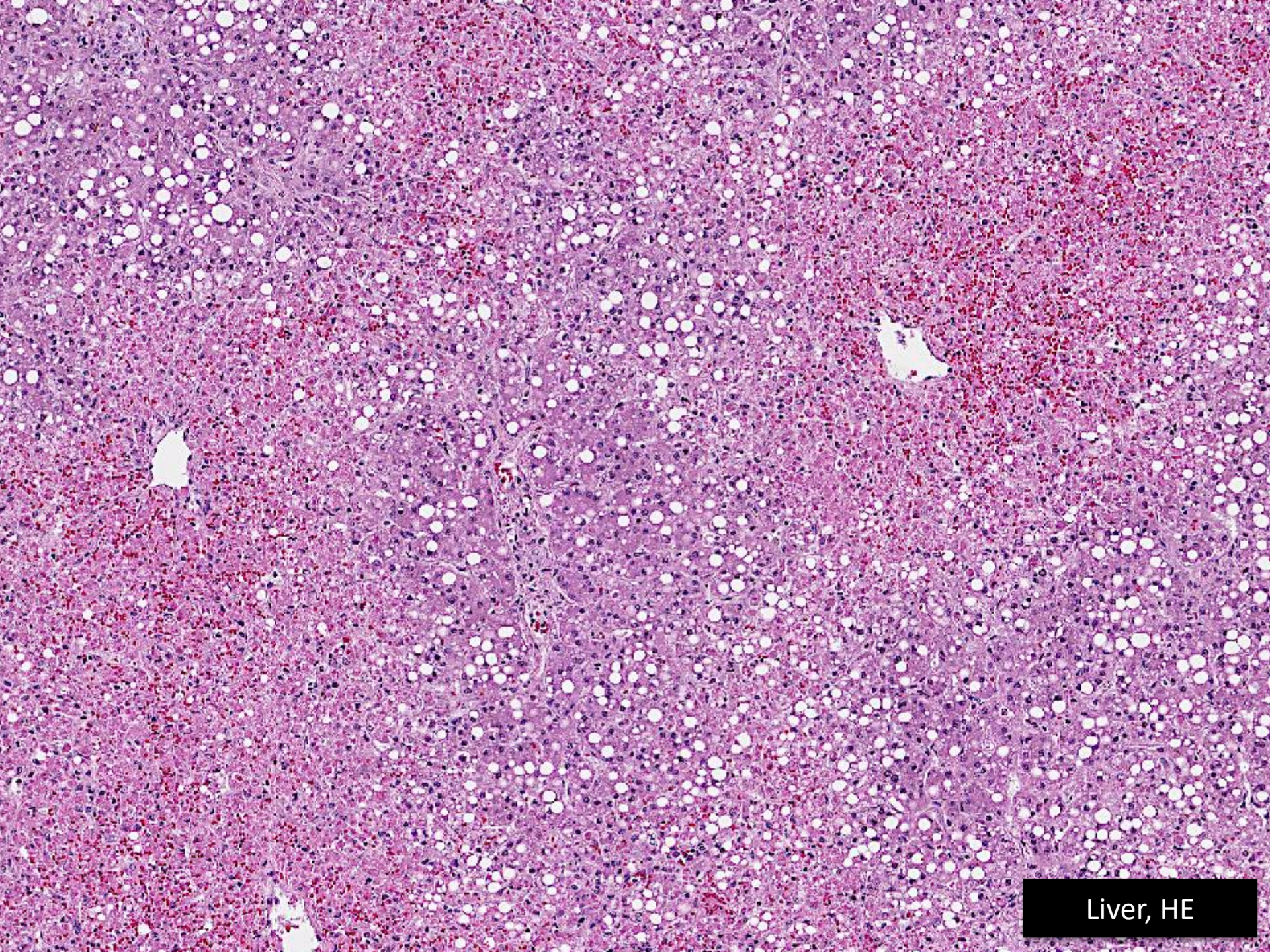
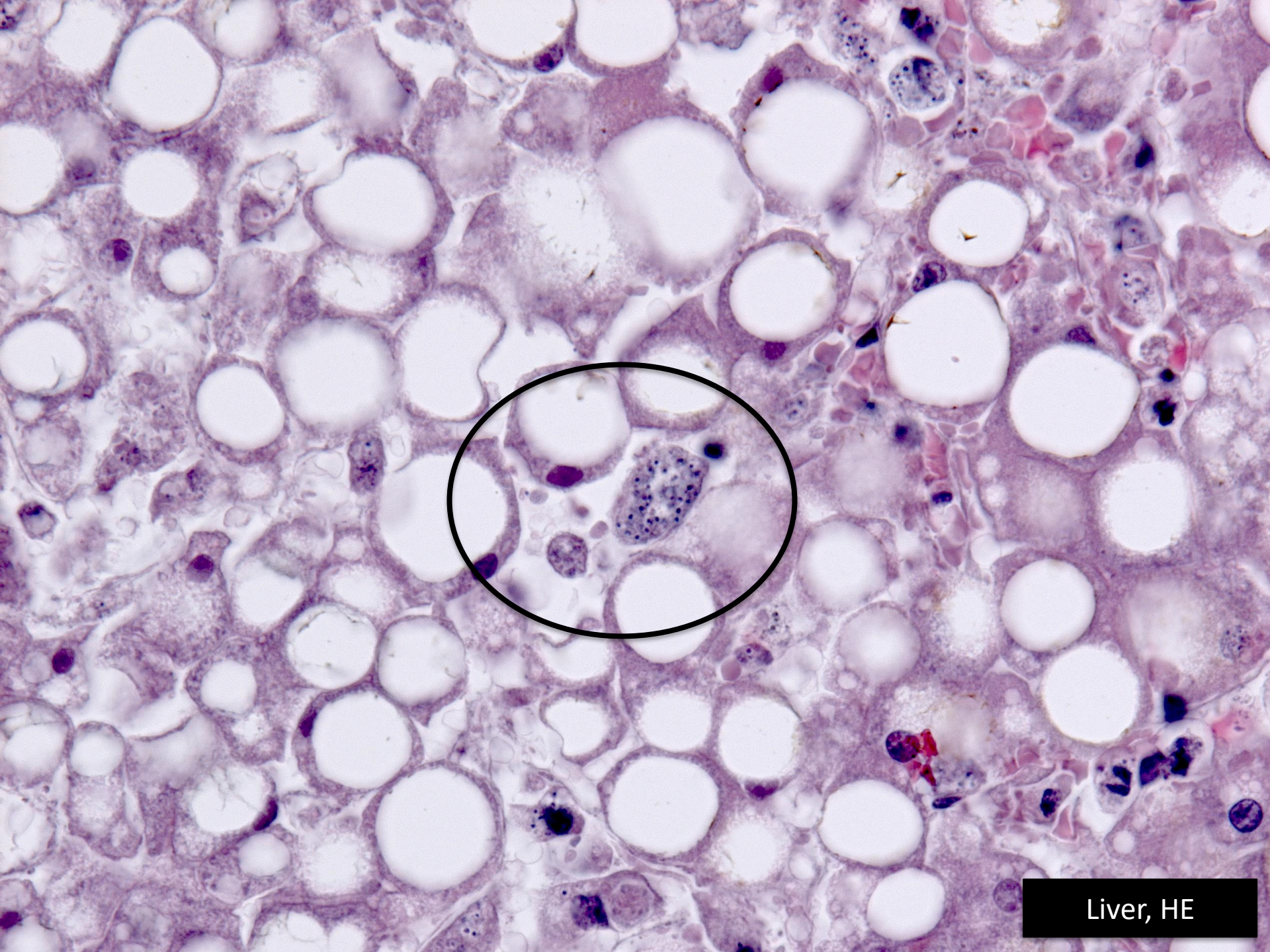
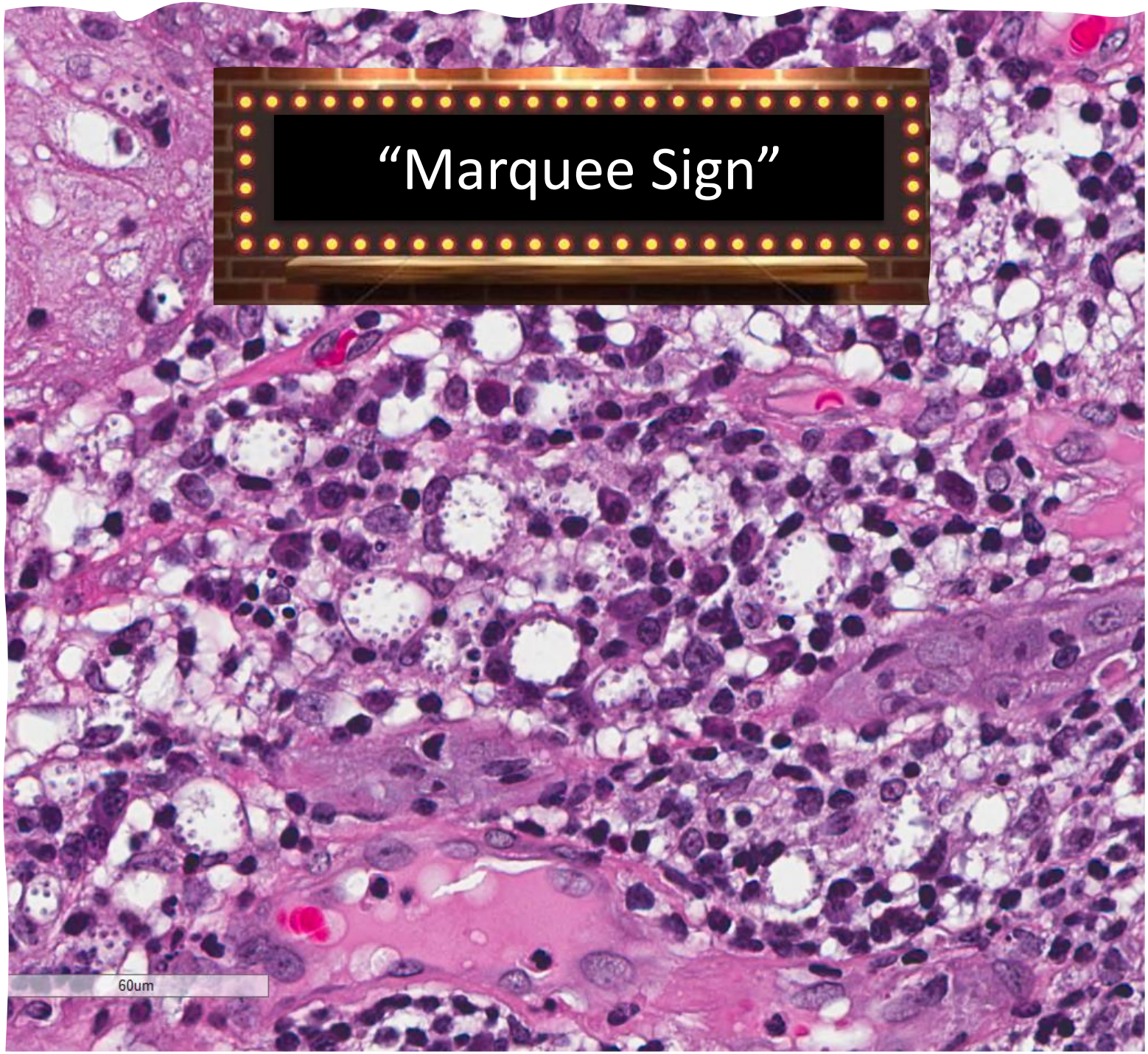

Liver, HE

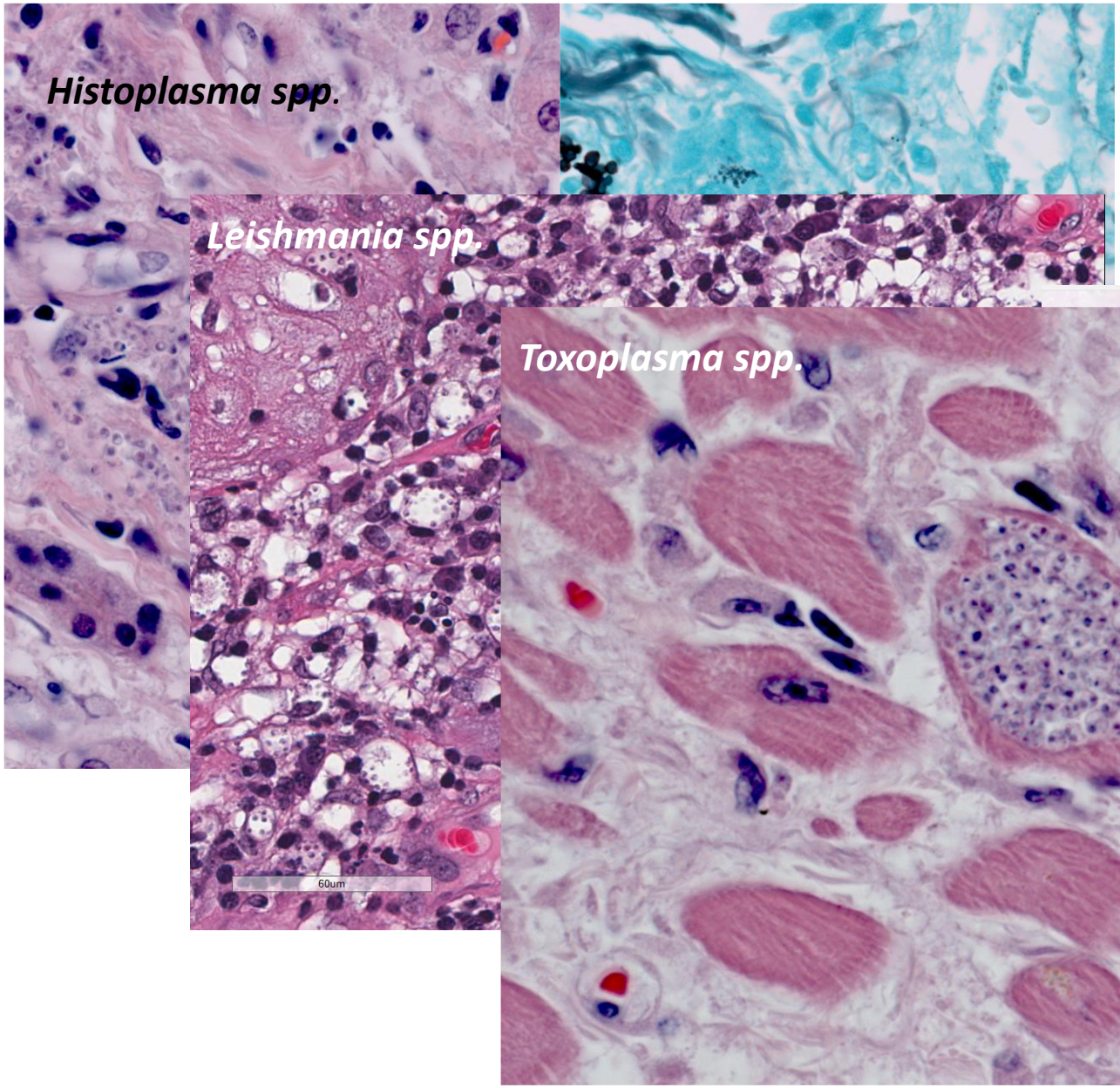

Liver, HE

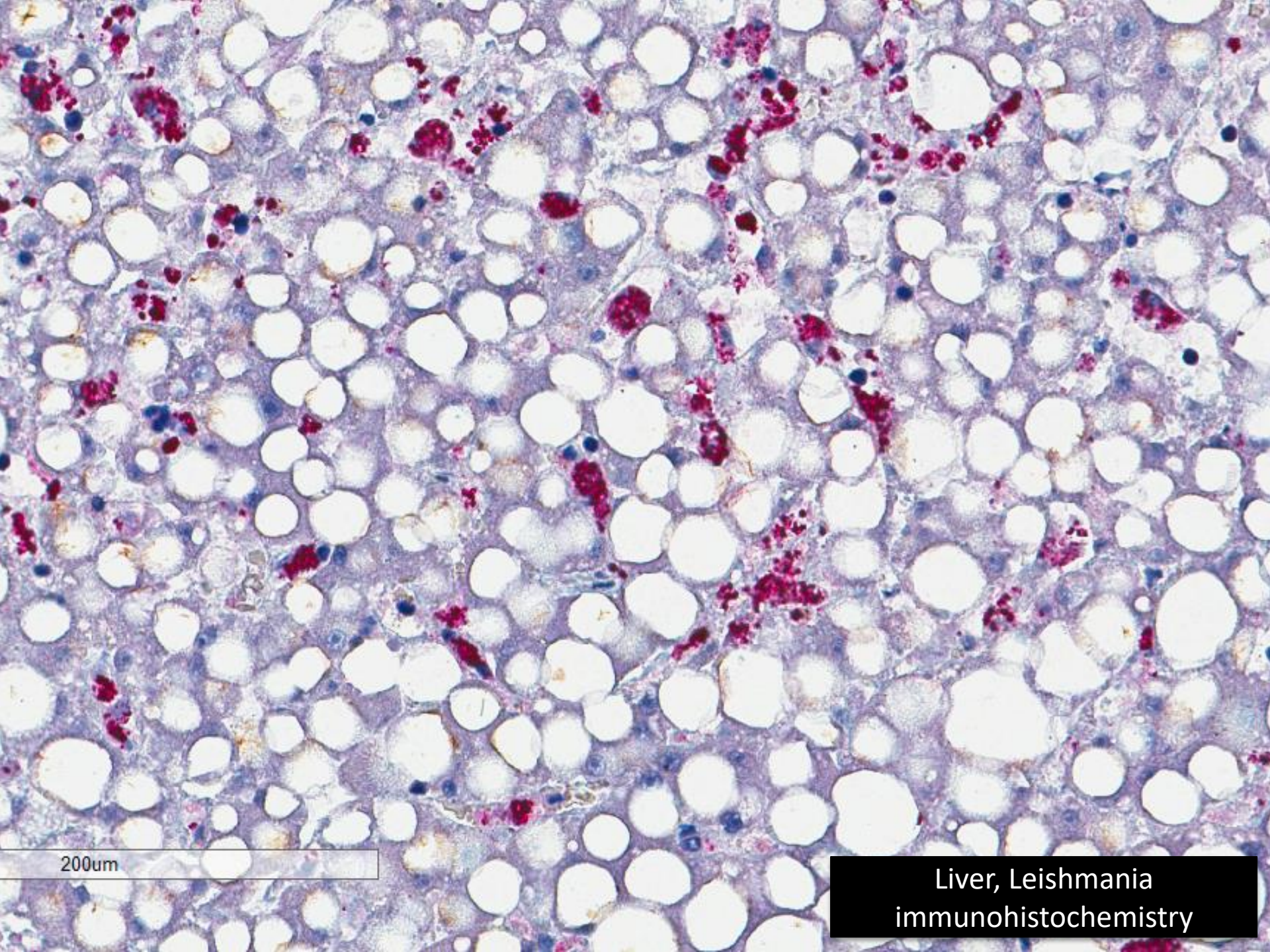

“Marquee Sign”
60um

Case 2 Workup

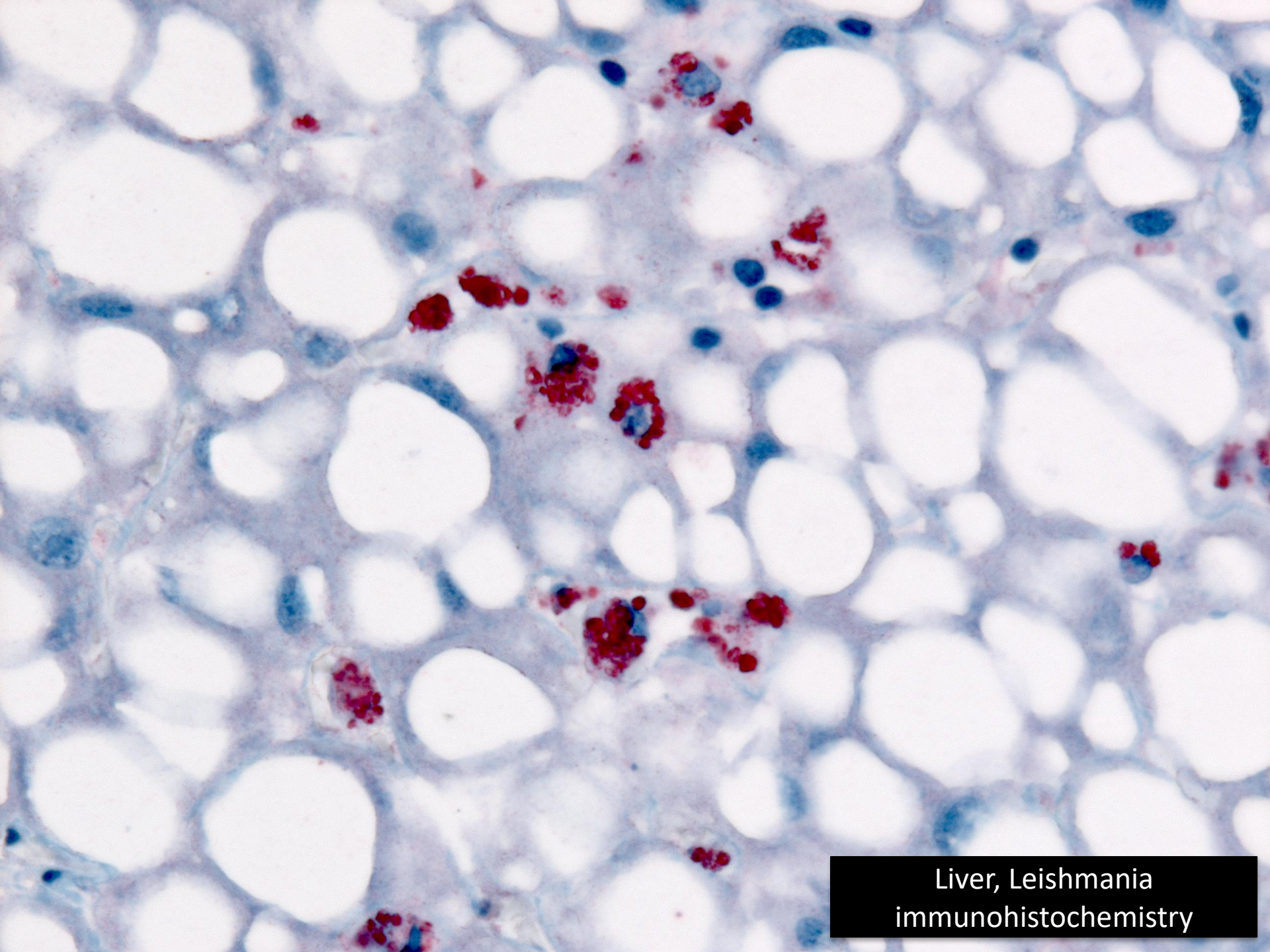
- **Differential diagnoses for intracytoplasmic, small round structures:**
  - *Histoplasma capsulatum*
  - *Leishmania spp*
  - *Toxoplasma gondii*
  - *Trypanosoma cruzi (tropism for striated cardiac and smooth muscle cells)*

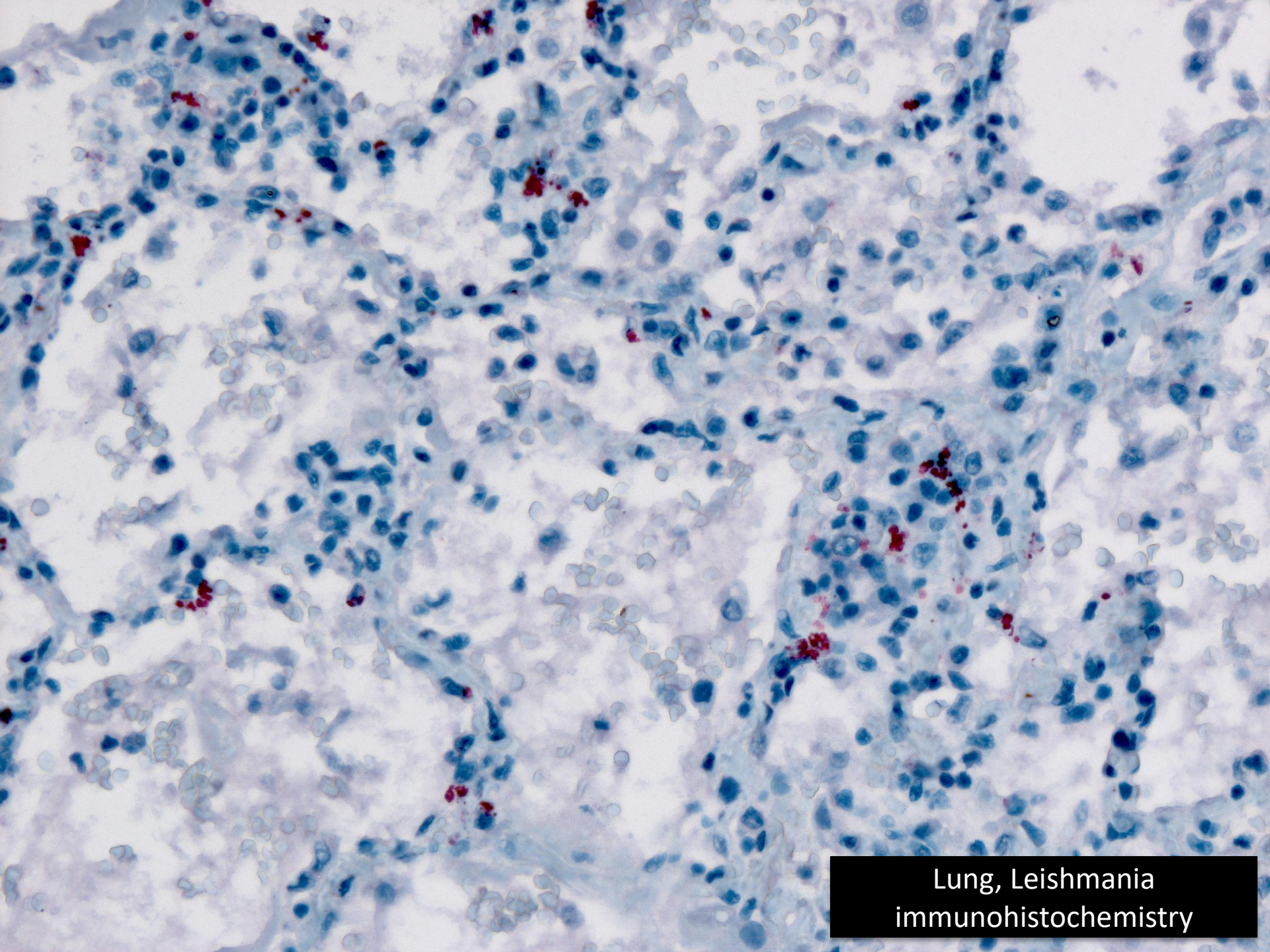
Differential Diagnosis for Small, Intracytoplasmic Organisms

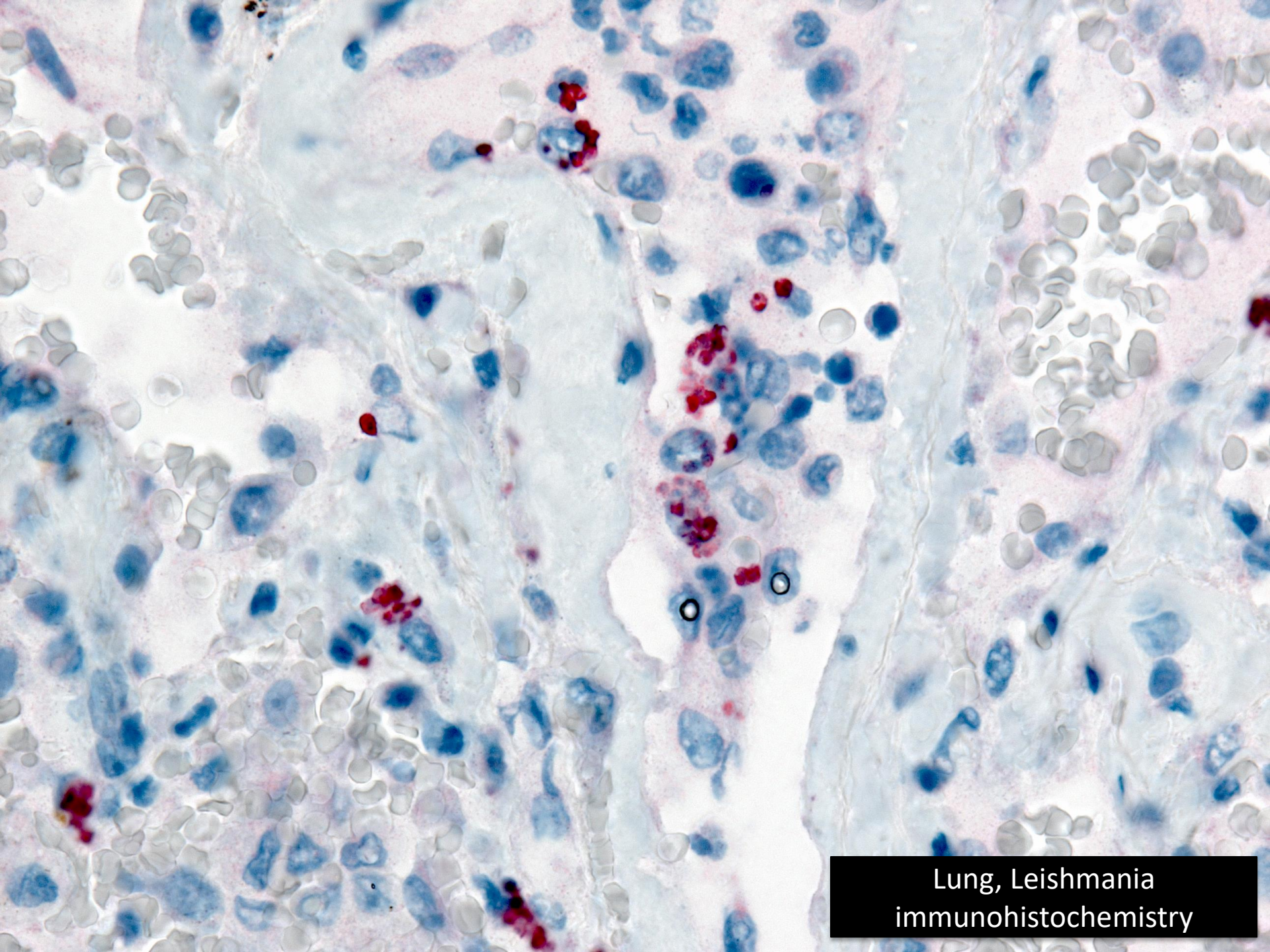


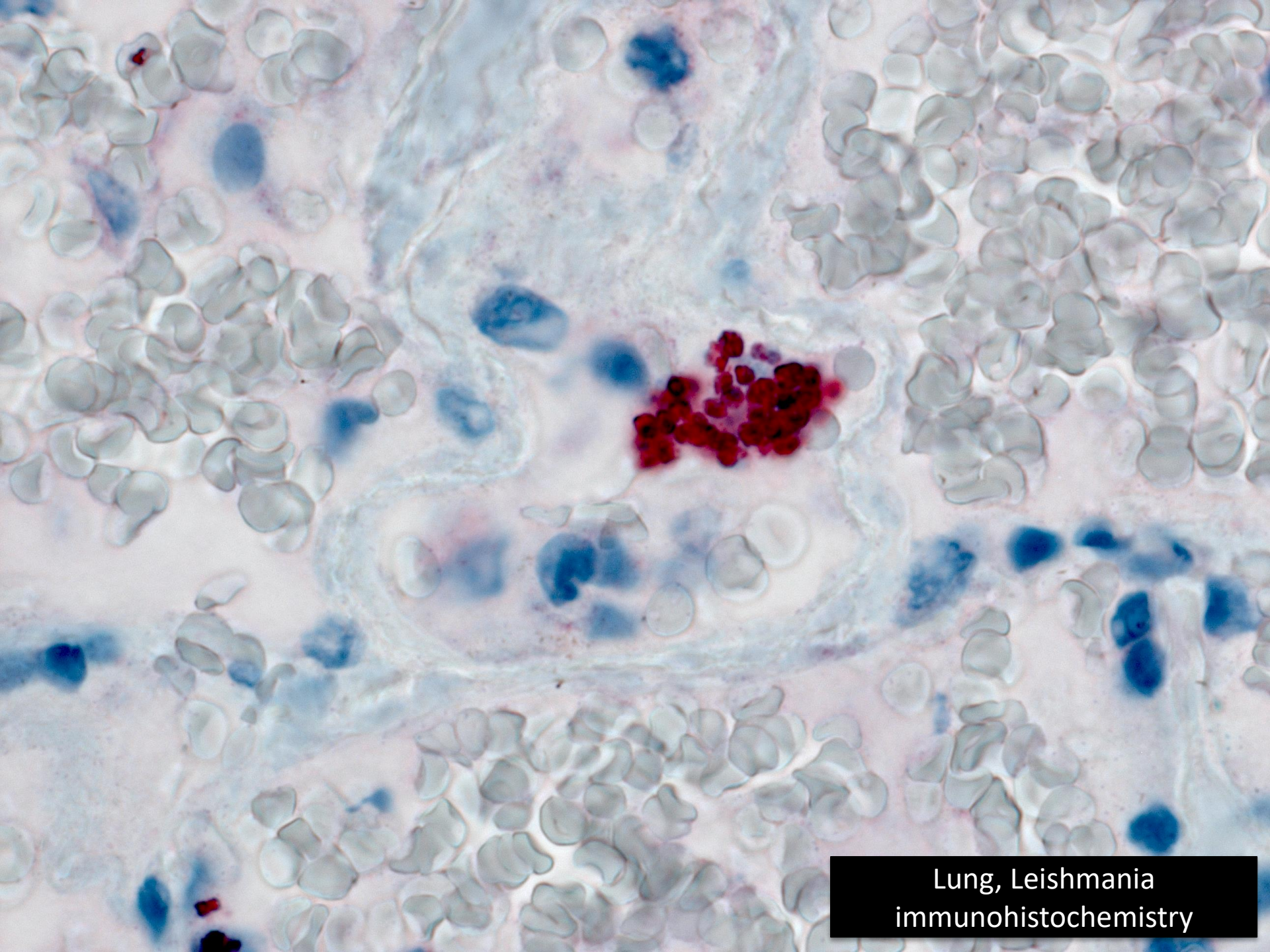


Liver, Leishmania immunohistochemistry

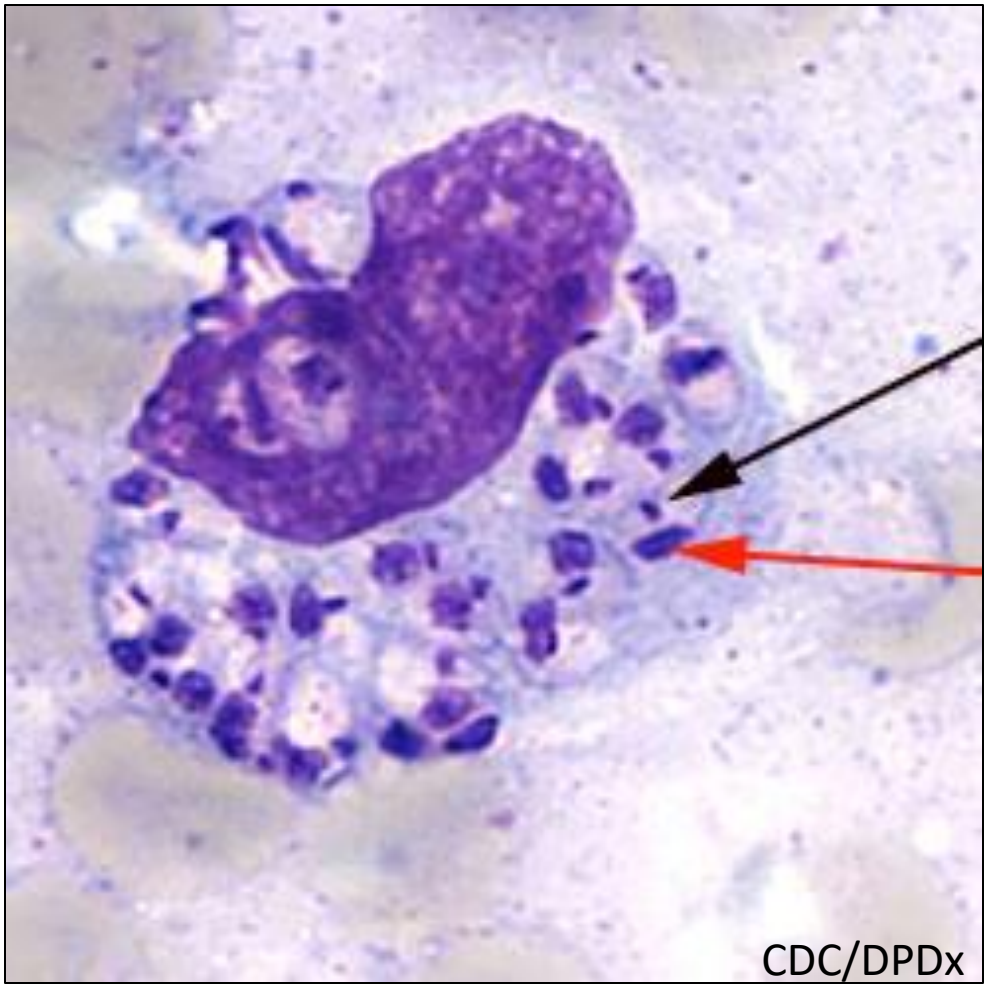

Liver, Leishmania
immunohistochemistry


Lung, Leishmania
immunohistochemistry


Lung, Leishmania
immunohistochemistry


Lung, Leishmania
immunohistochemistry

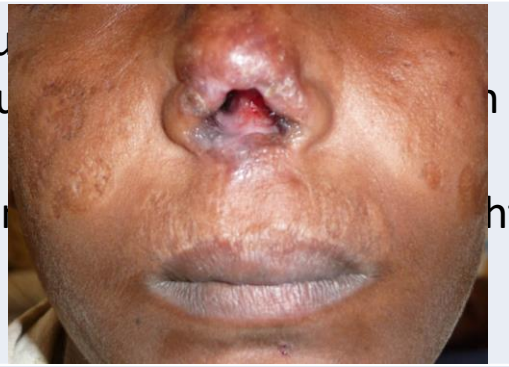
Leishman-Donovan Bodies
Leishmaniasis



- **Amastigote**
  - Nucleus and Kinetoplast
  - 2-4 micrometers
- 1900 Sir William Leishman and Charles Donovan
  - “Oval bodies” in patients from India

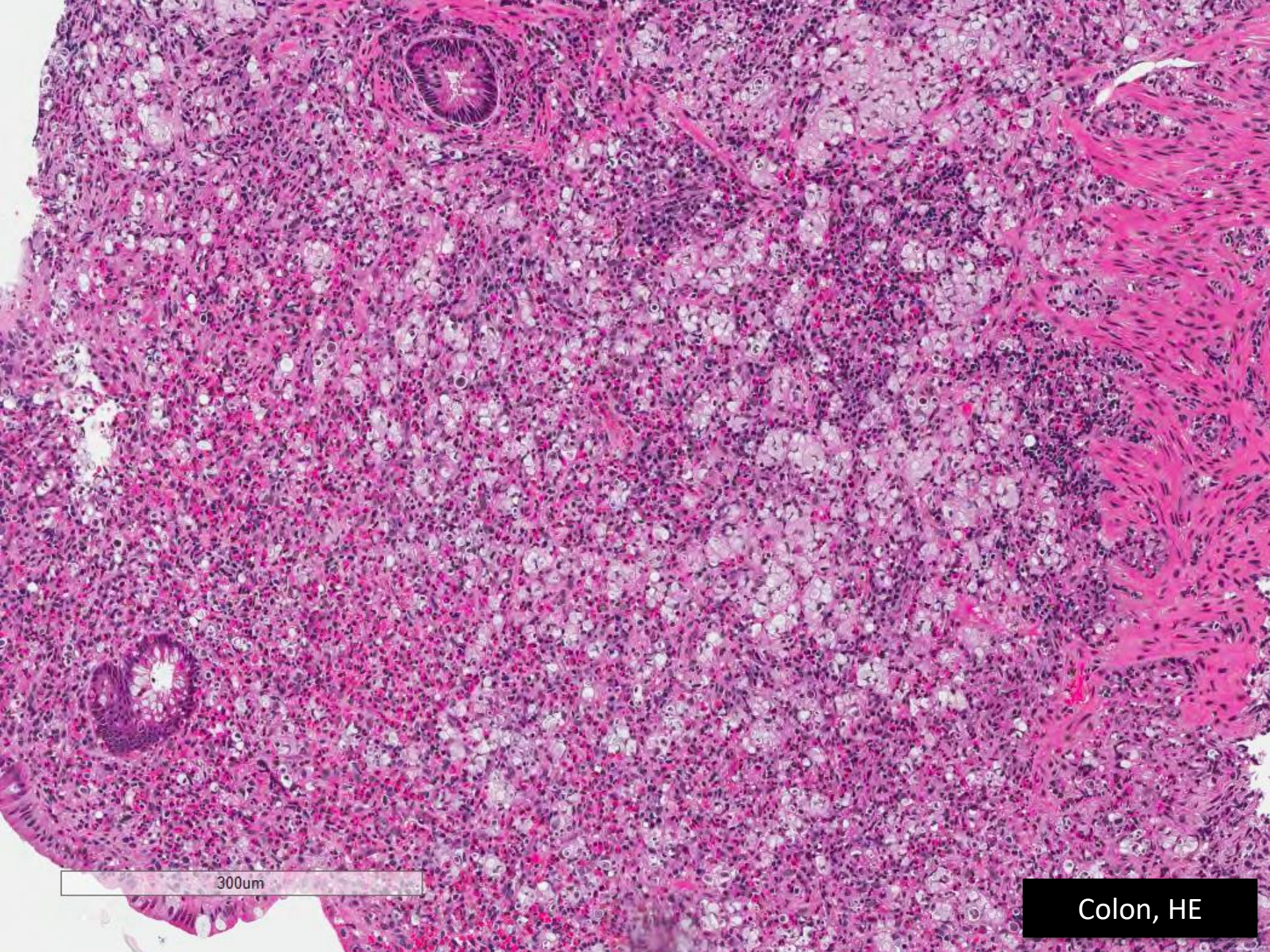
Leishmaniasis

Neglected Tropical Disease

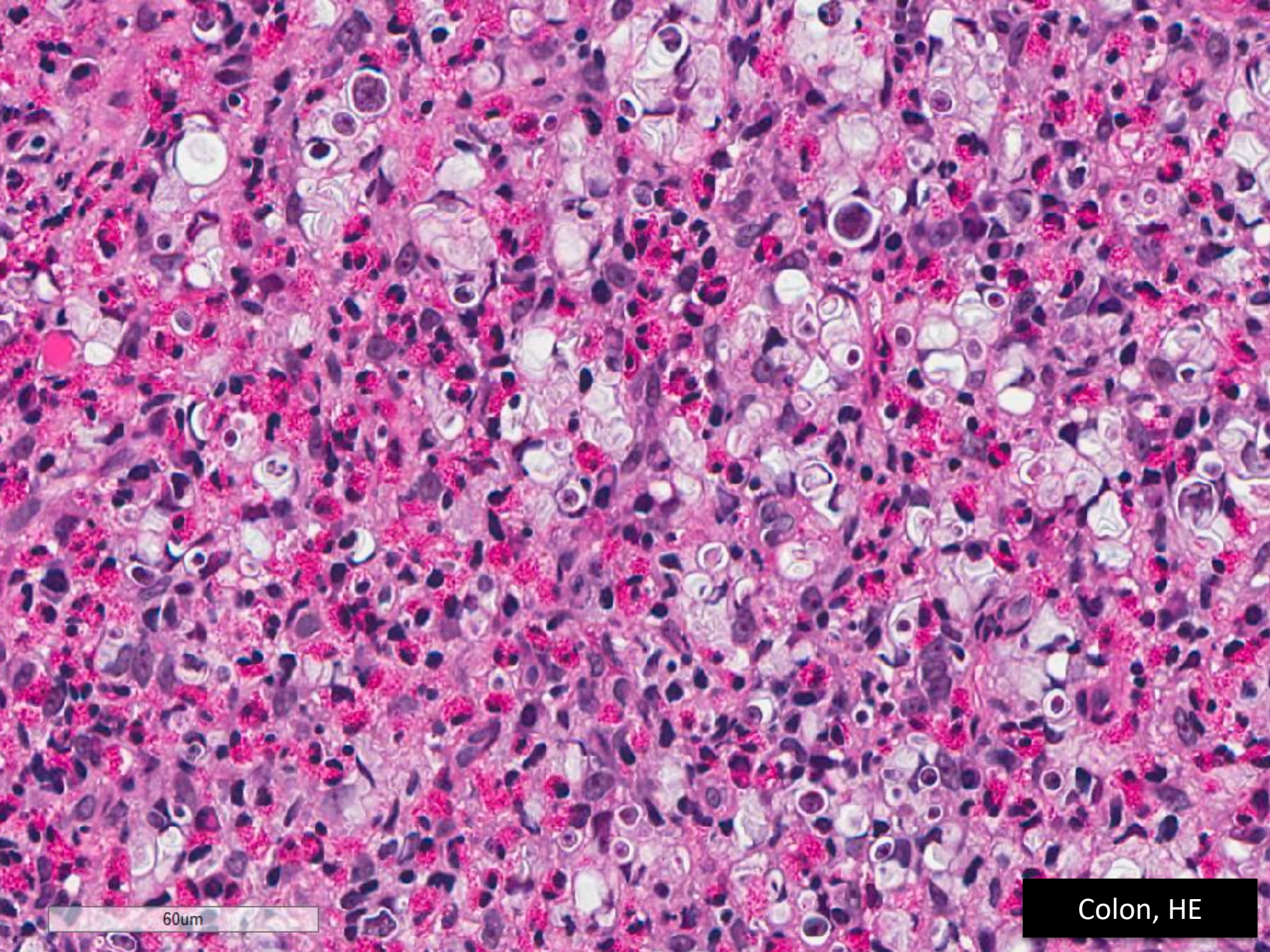
Disease	Leishmaniasis (cutaneous, mucocutaneous, visceral – kala-azar)
Agent	Intracellular protozoa; more than 20 species
Epidemiology	Worldwide; tropical and subtropical
Transmission	Sandflies (*Lutzomyia spp*)
Clinical features	  
Pathology	Granulomatous and lymphoplasmacytic inflammation in skin; histiocytes with intracytoplasmic amastigotes (dot-nucleus and dash-kinetoplast morphology)
Diagnosis	Culture, biopsy (PCR on formalin-fixed tissues to speciate)
Treatment	Depends on species, includes amphotericin B for visceral leishmaniasis

Case 3

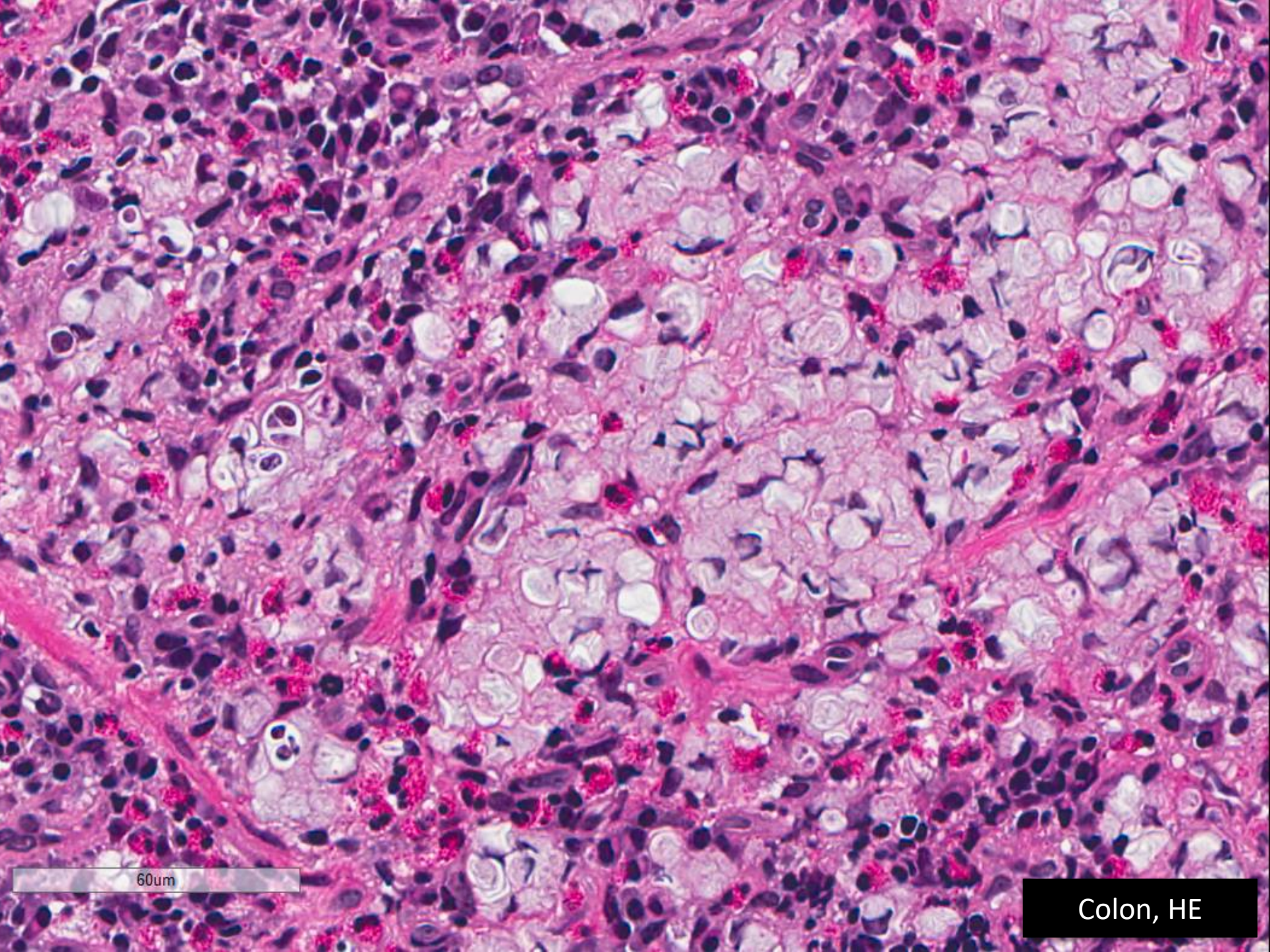
- 9 yo F from Turkey
- 3-months of fatigue, oral ulceration, abdominal pain and diarrhea
- Symptoms unresponsive to steroids, elementary diet, anti-amebic medication, antibiotics
- Abundant fecal leukocytes in stool
- Testing for celiac disease, HIV, and CMV was negative
- Endoscopic evaluation showed **pancolitis**
- Diagnosed with **“non-specific colitis”** based on endoscopic and histopathologic findings

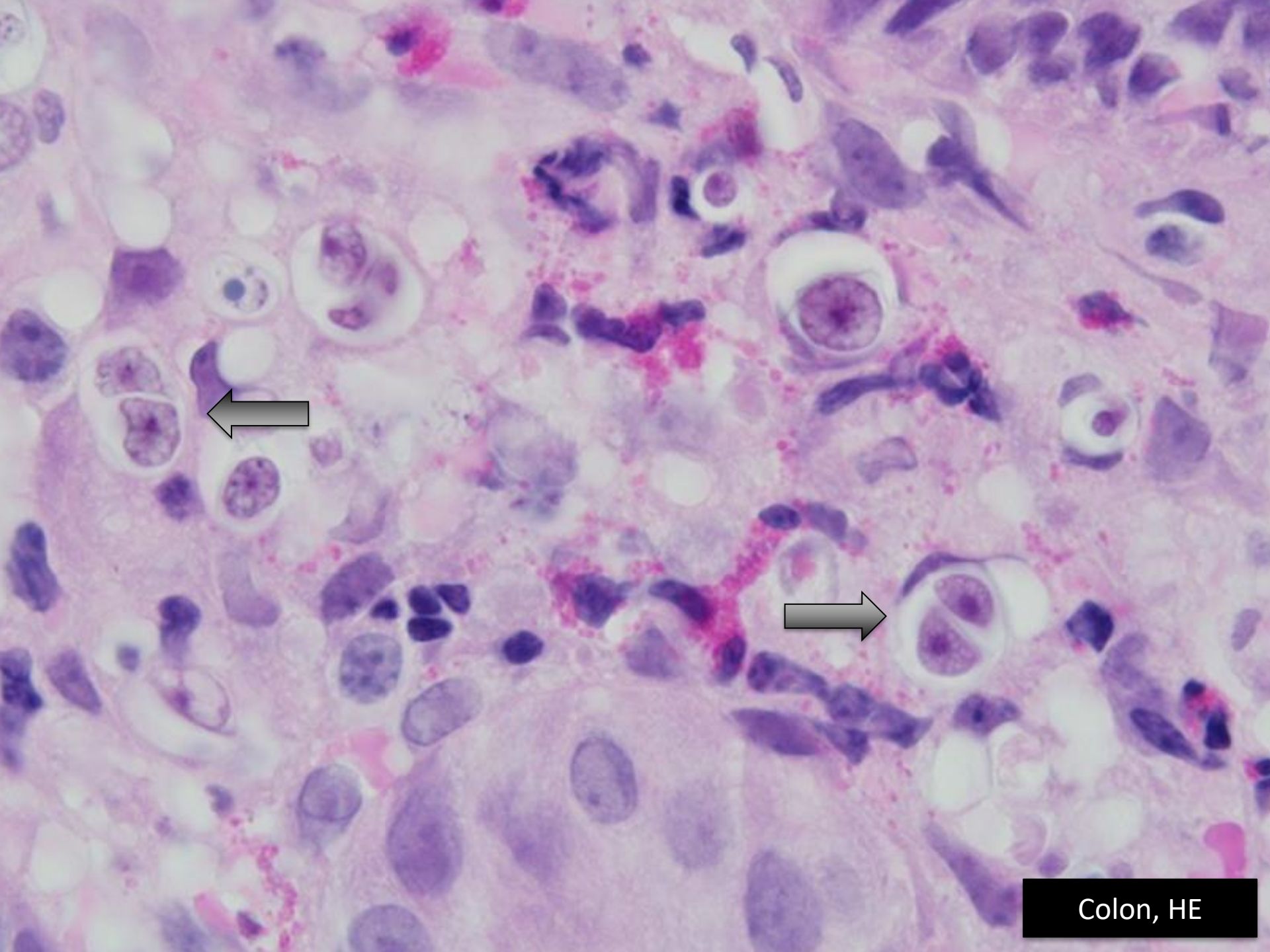


Colon, HE



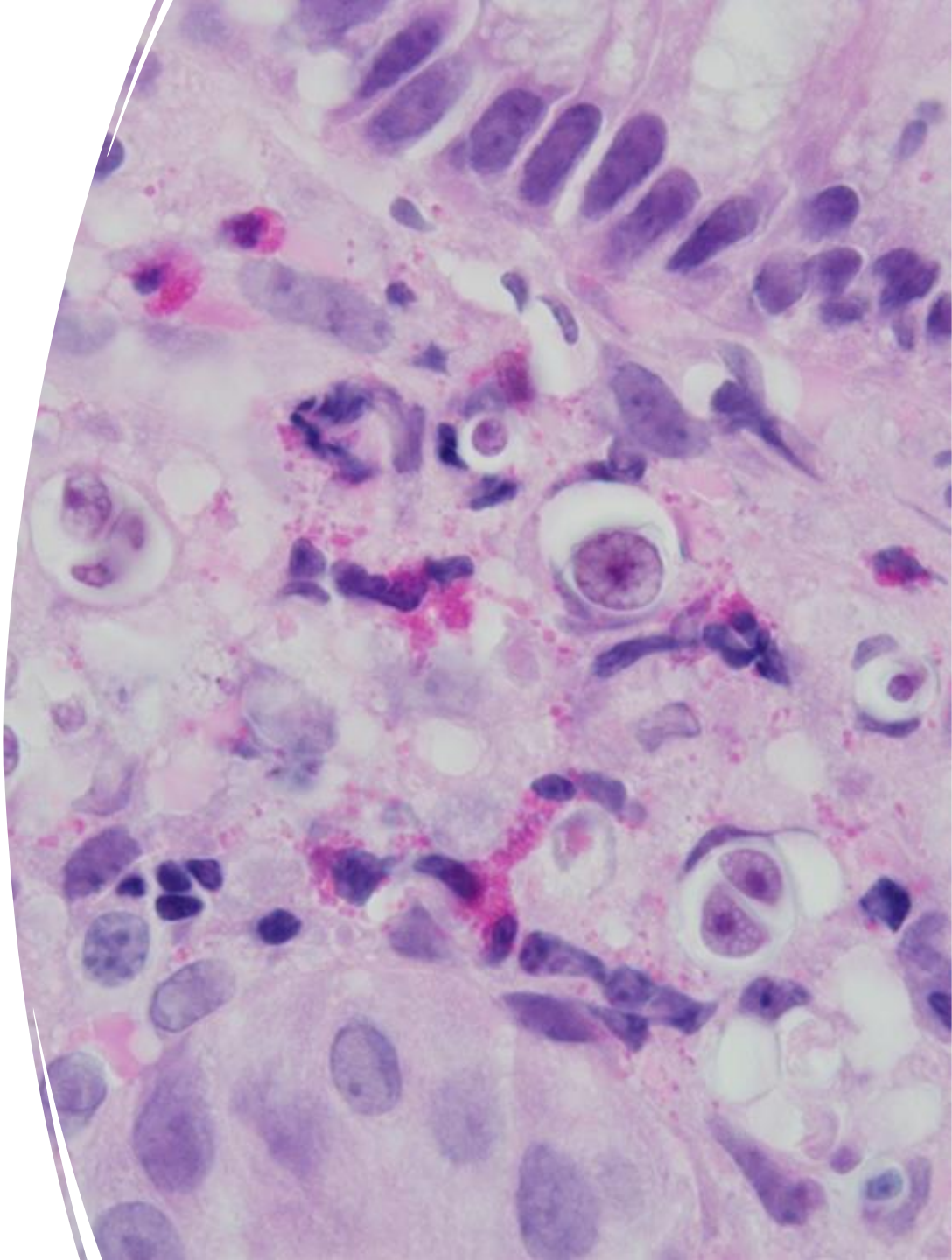
Colon, HE

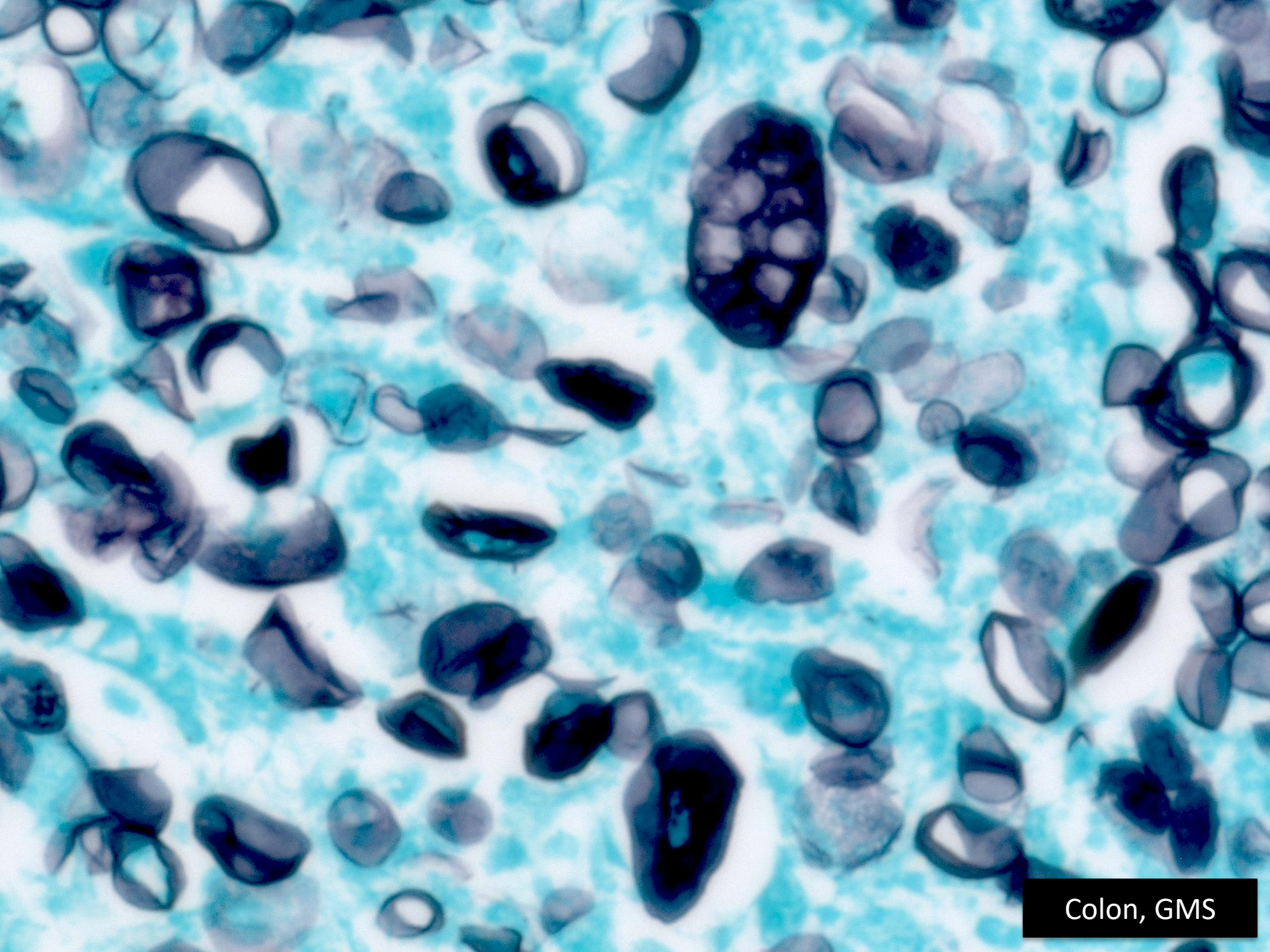

60um
Colon, HE

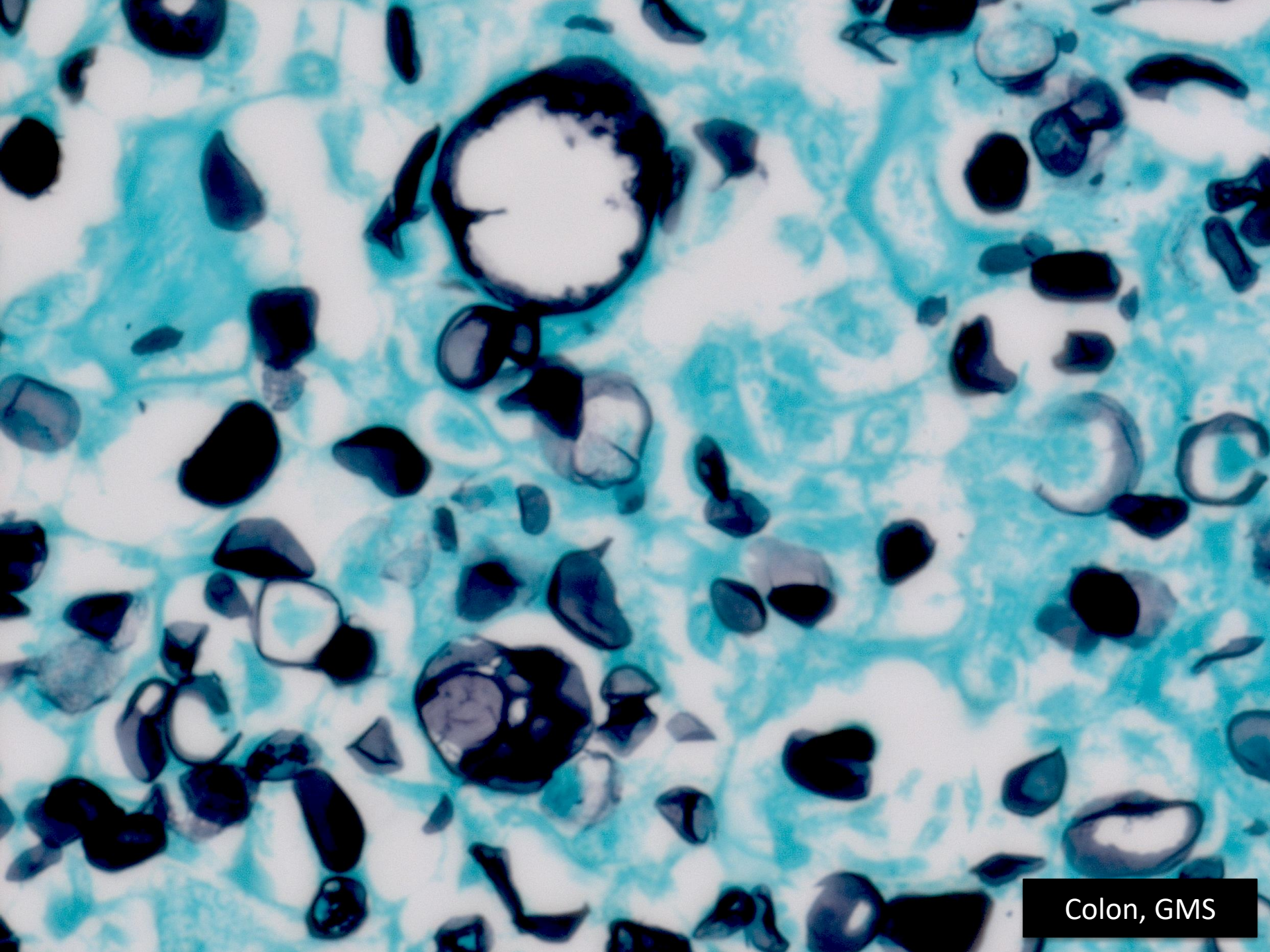

Colon, HE

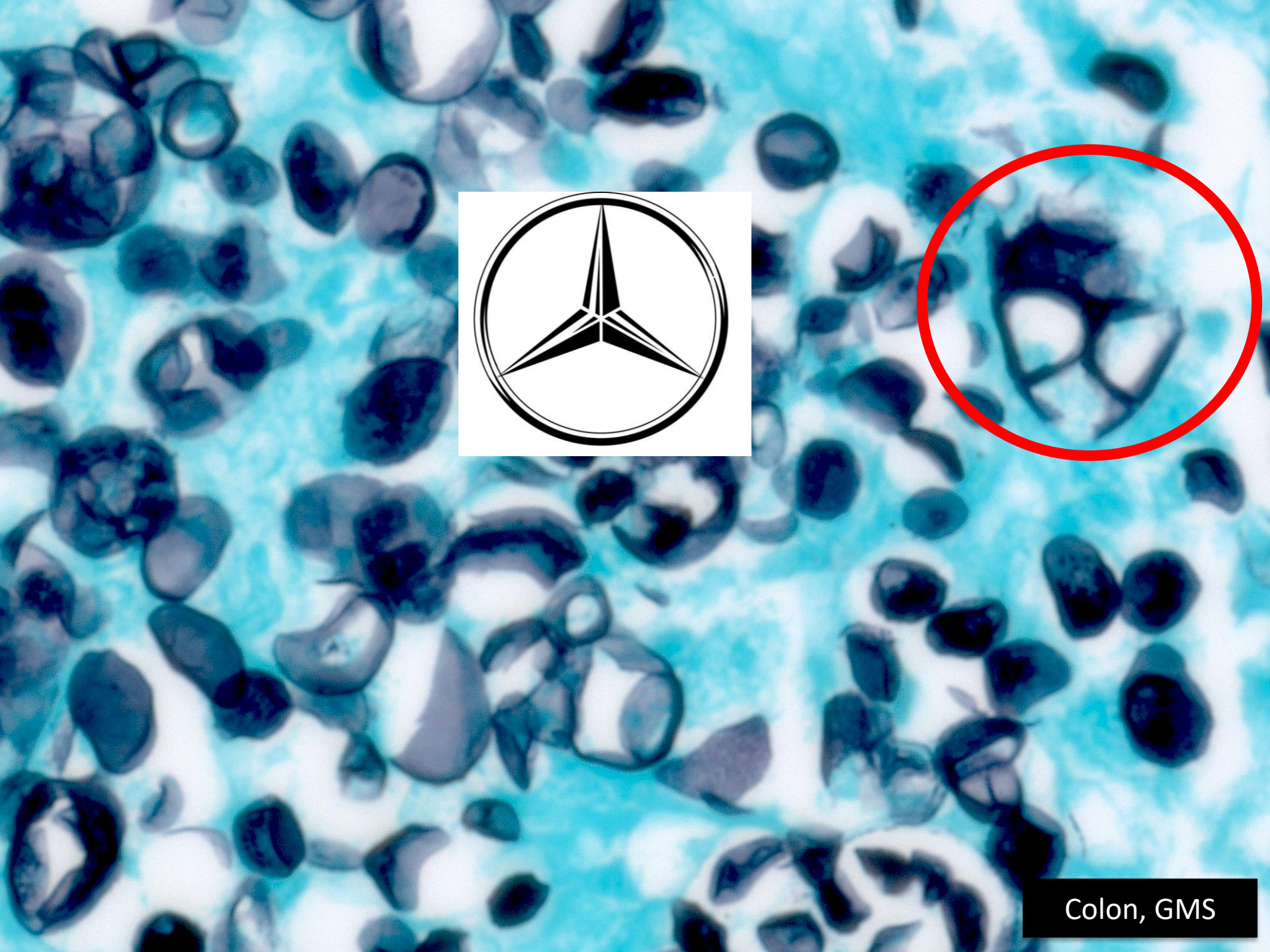
Case 3 Workup

- Fungus
  - GMS stain
- Ameba
- Malignancy
- Artifact



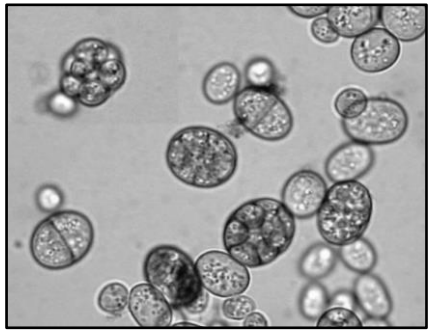

Colon, GMS


Colon, GMS



Colon, GMS

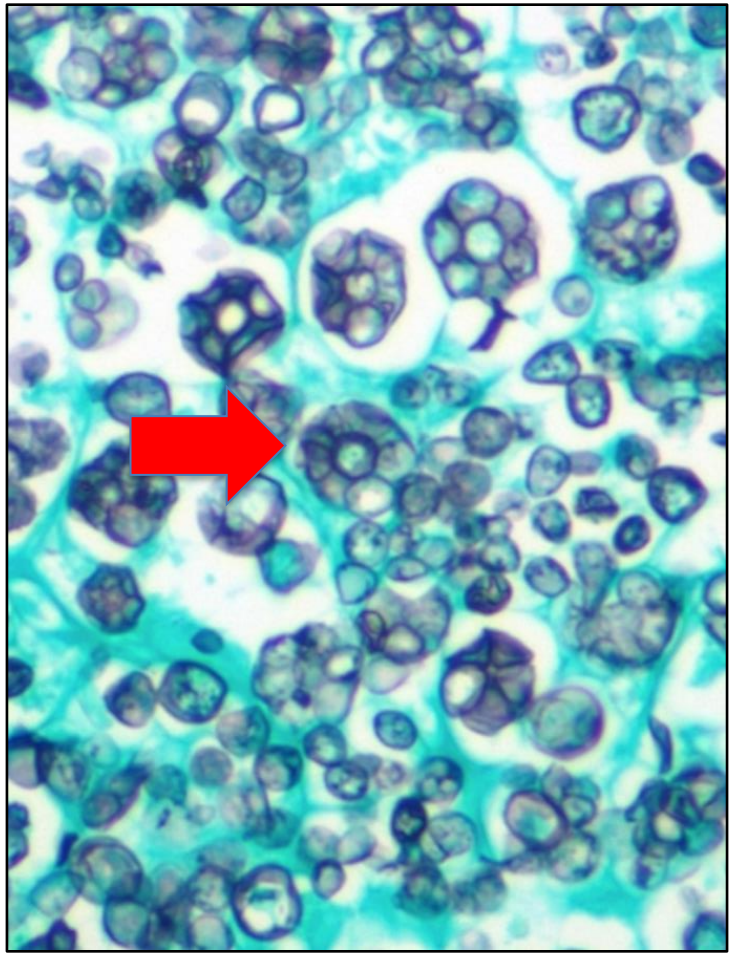
Prototheca



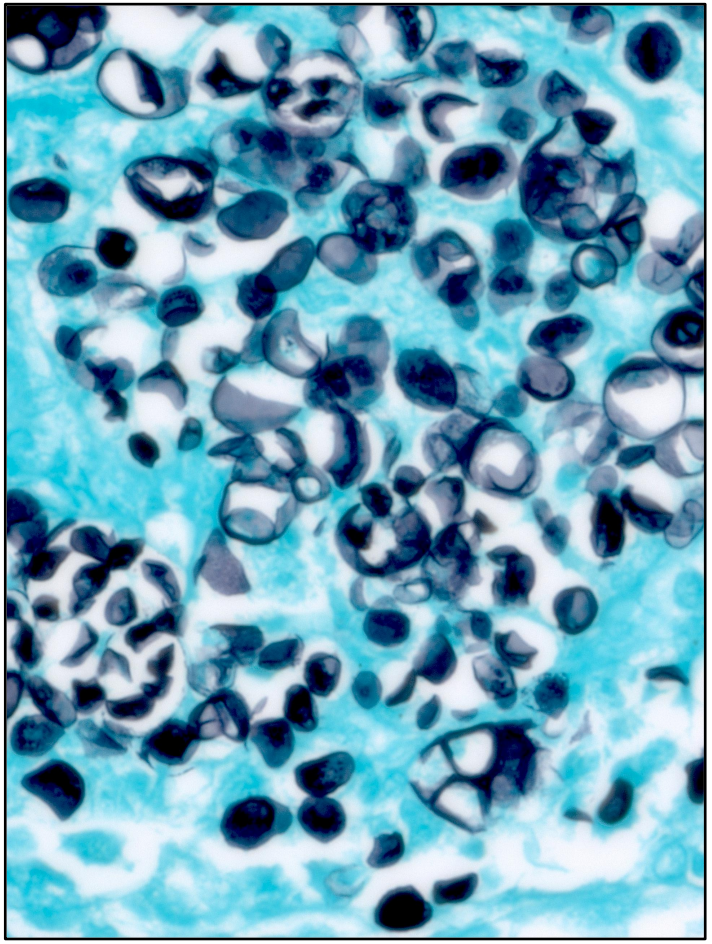
Disease	Protothecosis
Origin	First reported human infection in Sierra Leone (1964), farm worker
Habitat	**Ubiquitous** achlorophyllous algae; raw or treated sewage, animal waste, soil, food, flowing or standing water
Human pathogens	***P. wickerhamii*** (more common in humans); ***P. zopfii*** (more common in animals); morphologically difficult to differentiate
Human infections	Cutaneous/subcutaneous (ulceration) Olecranon bursitis (repeated trauma to elbow) Disseminated disease (immunosuppressed)
Diagnosis	Morphology: **3-30 um**, spherical to oval, asexual reproduction by **internal septation**. UNLIKE fungi, NO glucosamine in cell well and UNLIKE bacteria, NO muramic acid; can be cultured on standard media
Treatment	Difficult! Infections can persist. Amphotericin B (+/- excision)

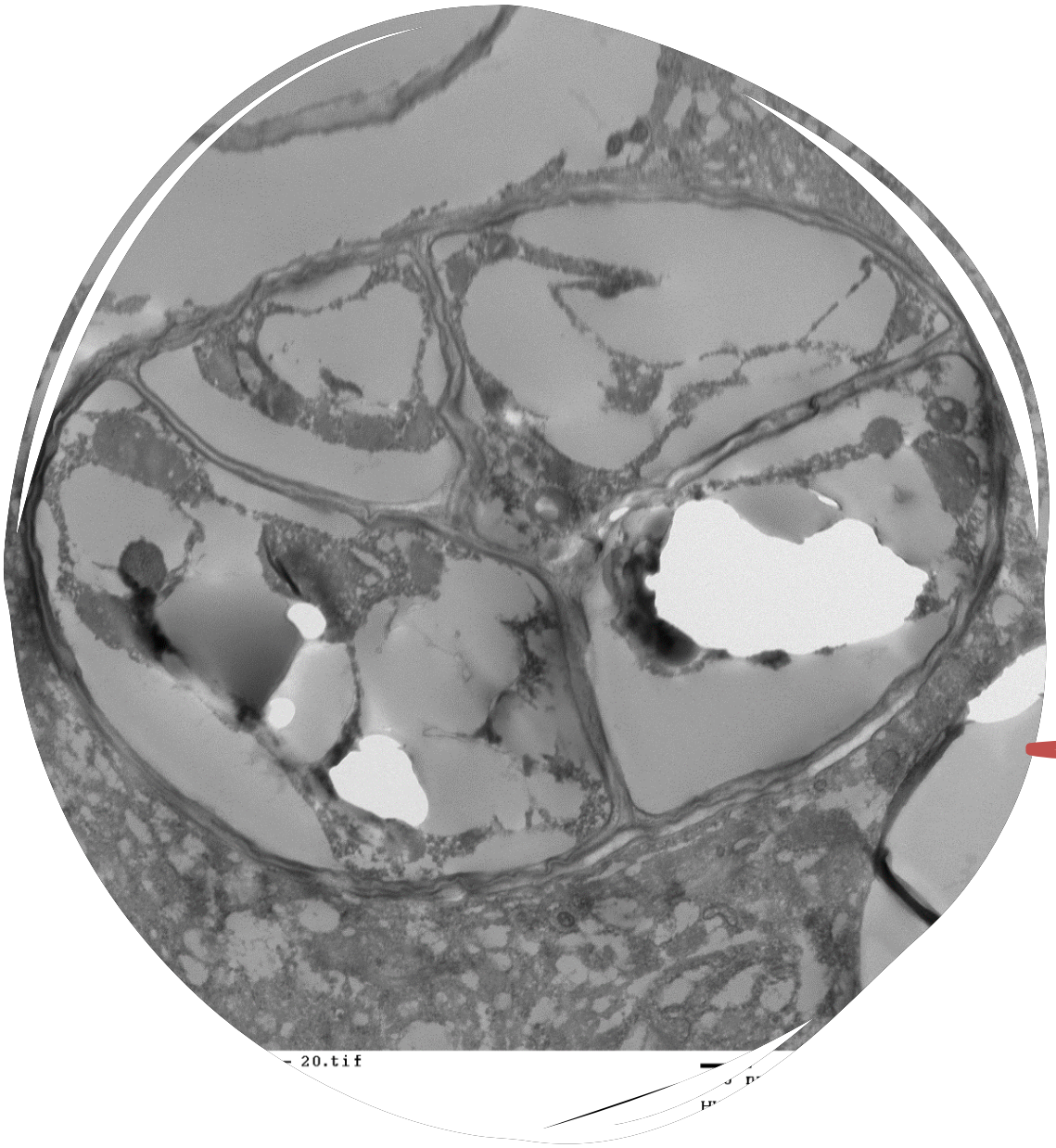
Prototheca species

P. wickerhamii



P. zopfii



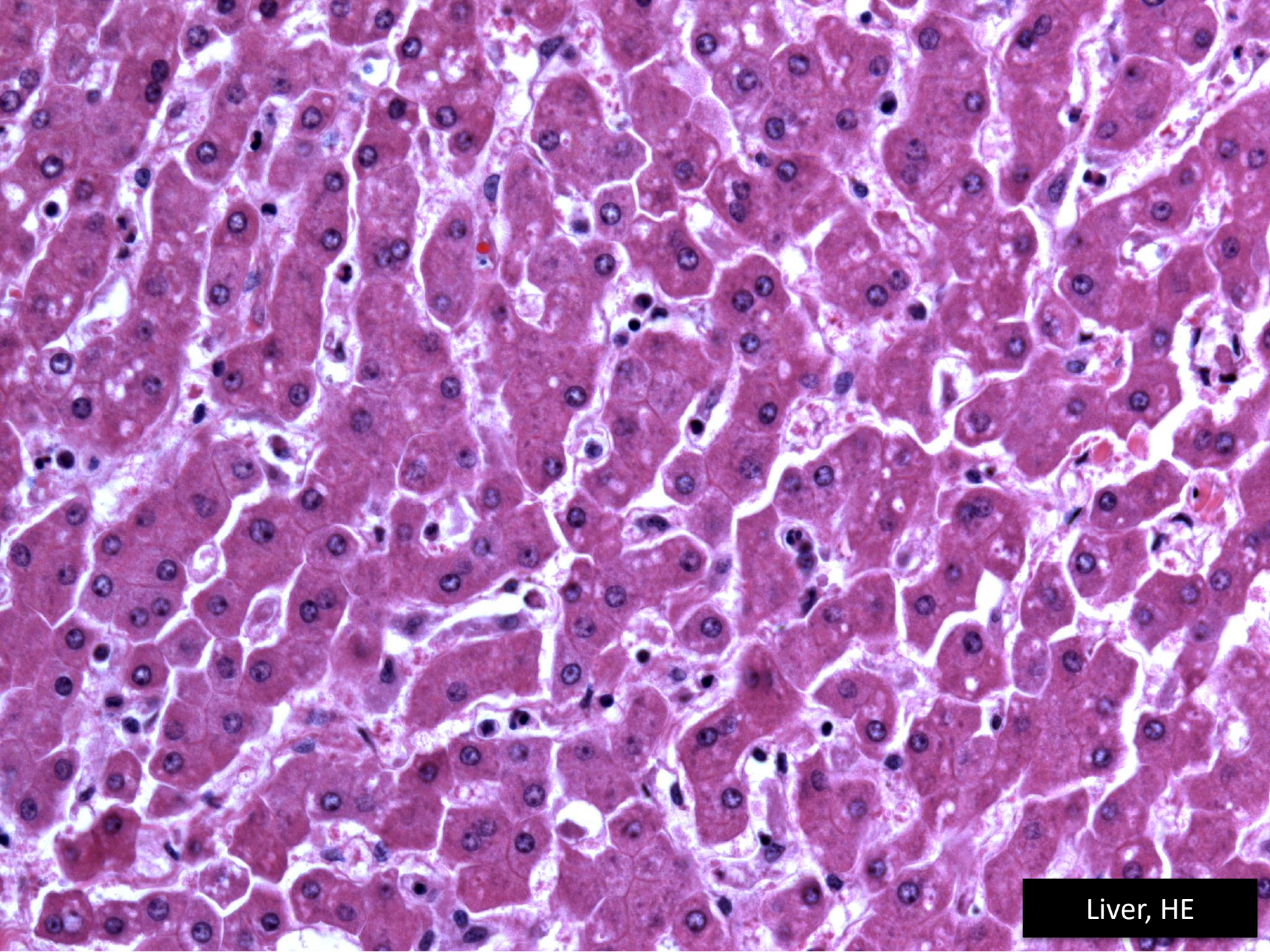


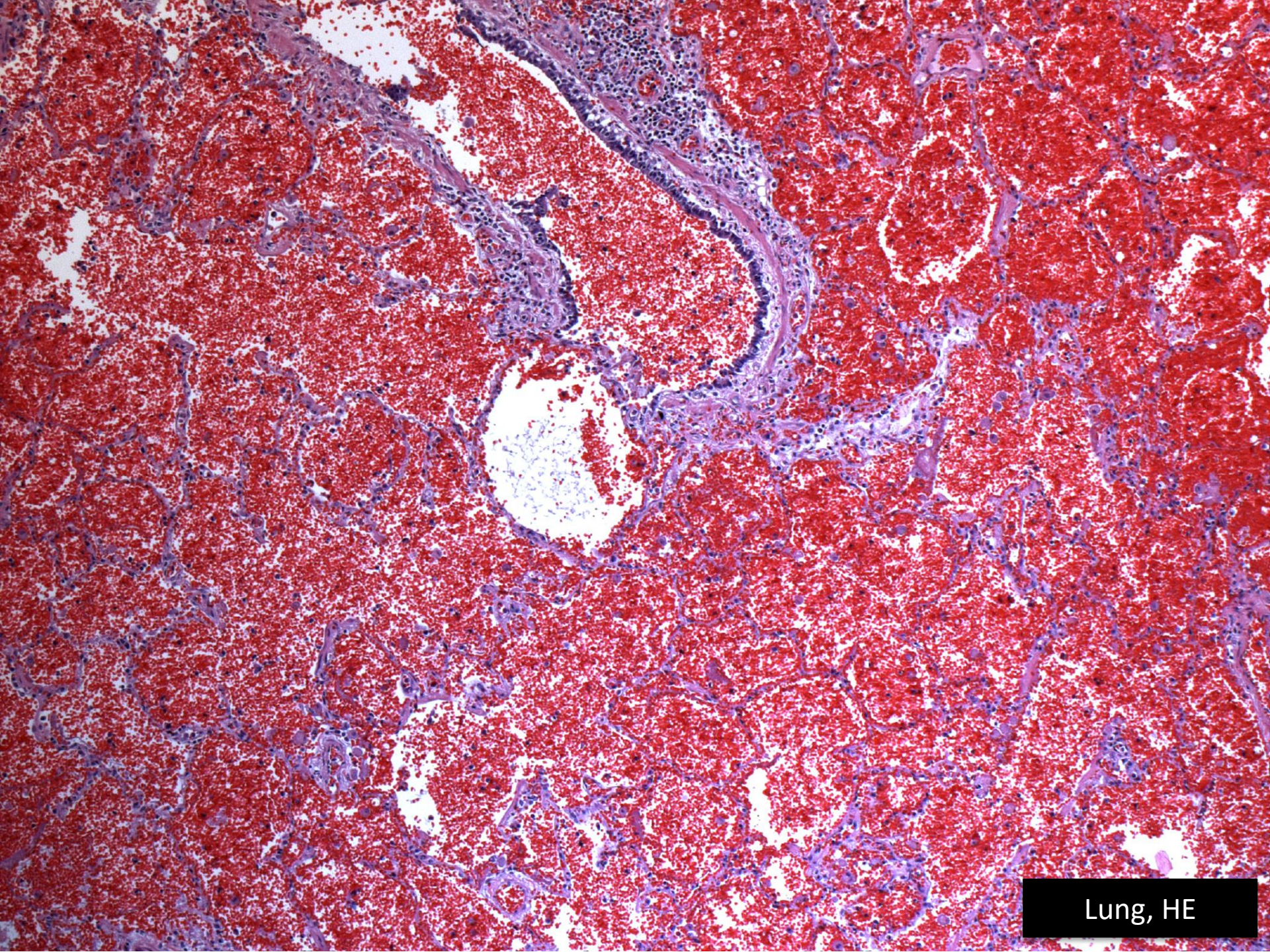
Electron micrograph of ***Prototheca zopfii*** - 4 sporangiospores can be seen

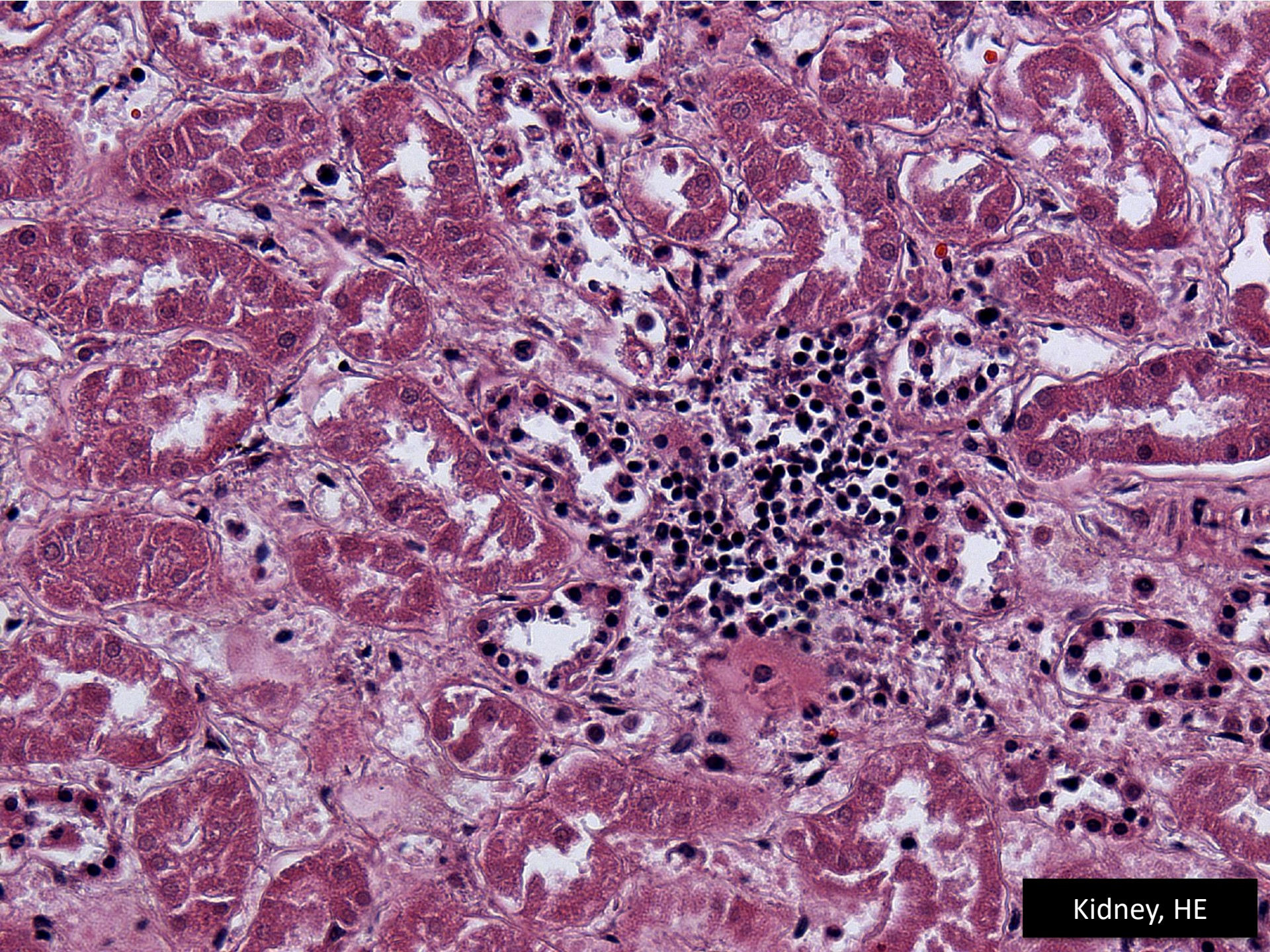
Case 4

- 18 yo M from Puerto Rico
- Developed fever, hematemesis, cough, sore throat, headache, myalgia
- Presented to the ED and was found to have thrombocytopenia
- Precipitous clinical course and died 1 week after onset of symptoms
- Pulmonary hemorrhage seen at autopsy

Liver, HE

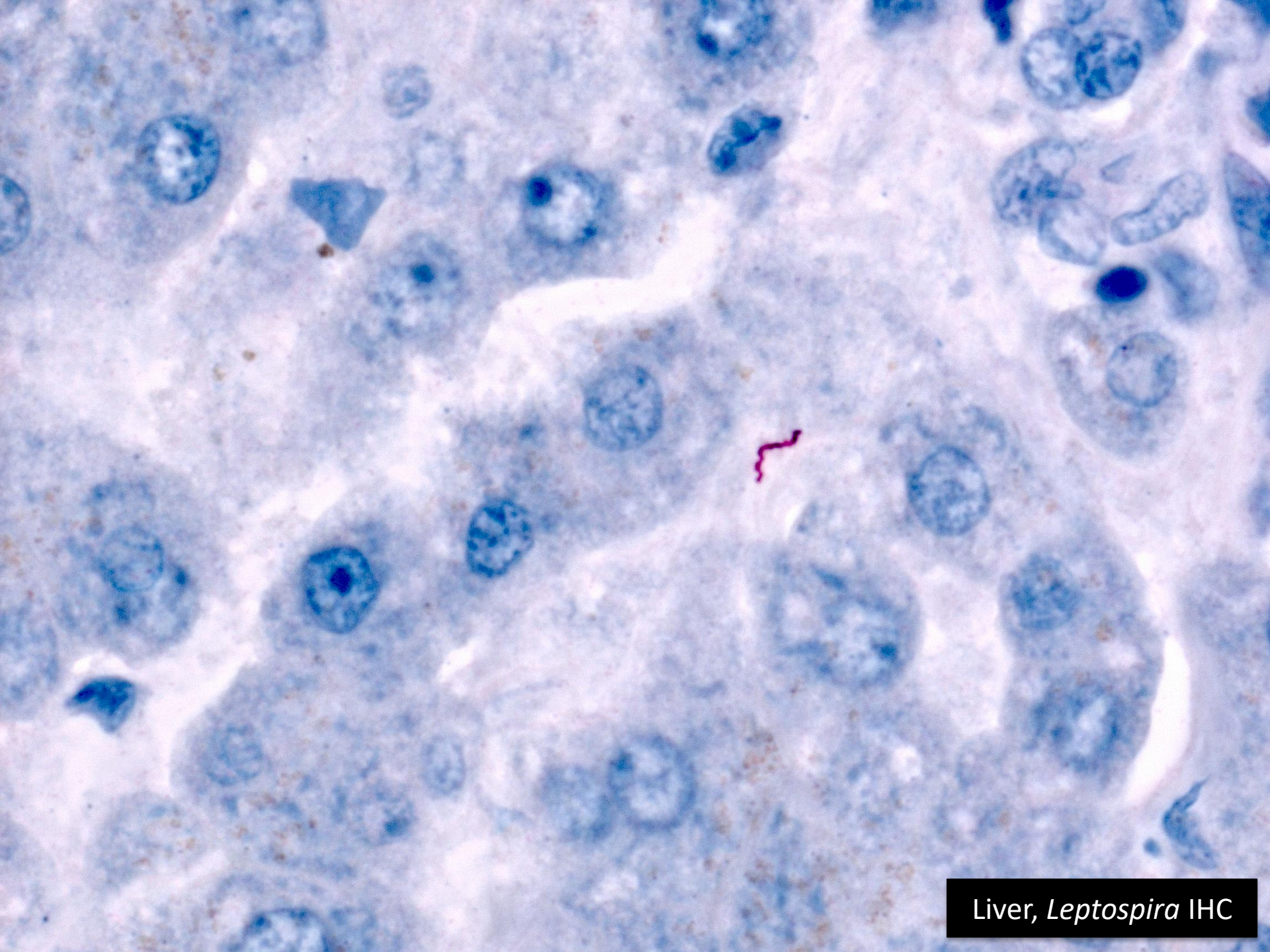

Liver, HE


Lung, HE


Kidney, HE

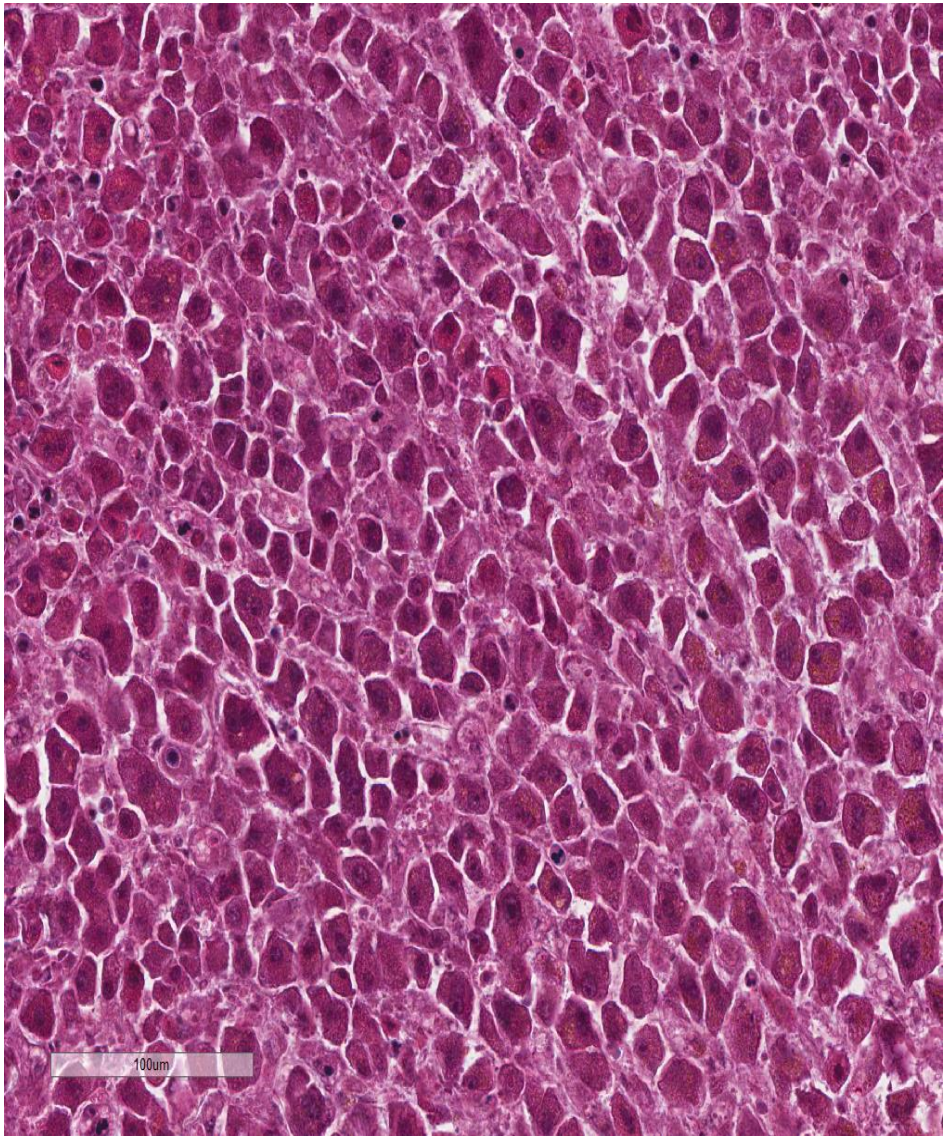
Case 4 Workup

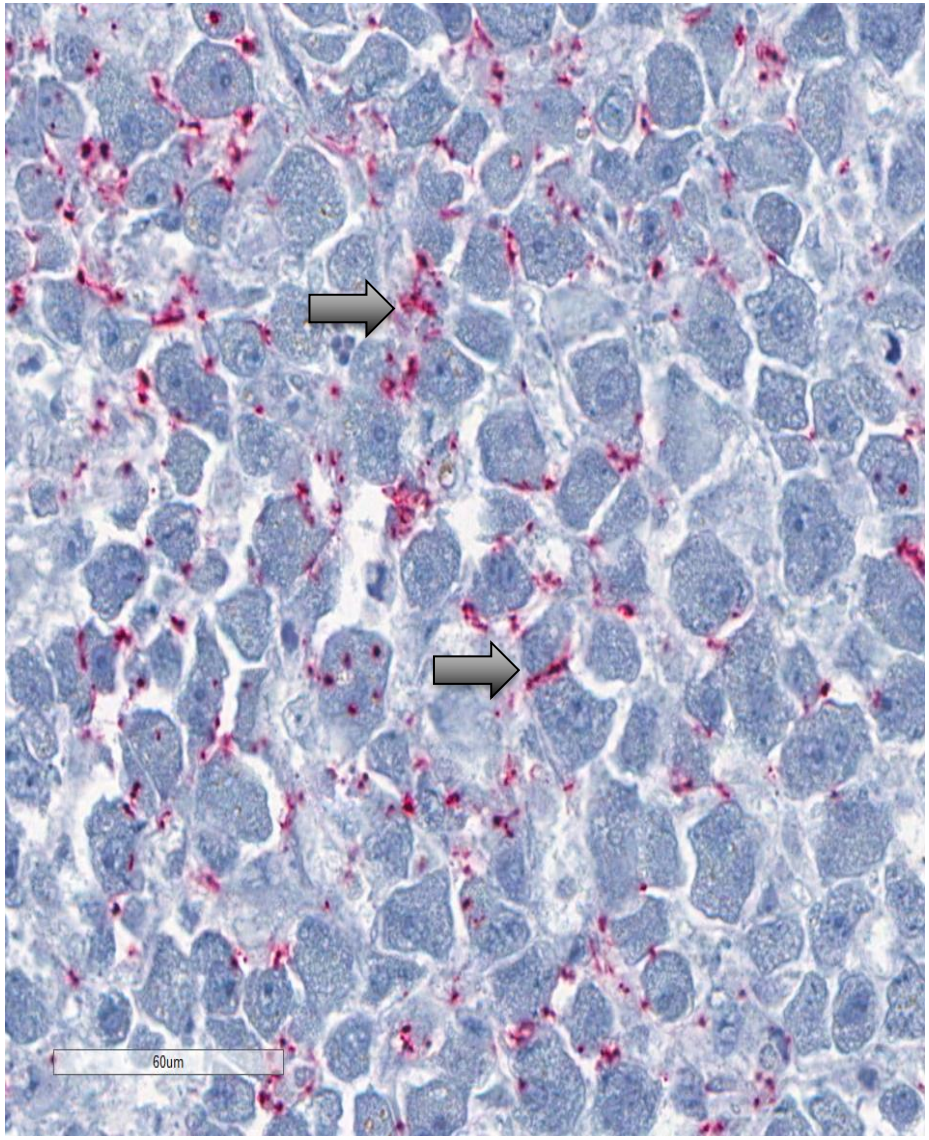
- **Differential diagnosis**
  - Leptospirosis
  - Dengue virus
  - *Rickettsia spp.*
  - Influenza viruses


Liver, *Leptospira* IHC

Hepatocellular dissociation in leptospirosis

studies show preferential leptospiral attachment to and invasion of the perijunctional region between hepatocytes (*Miyahara et al. 2014*)





Leptospirosis

Disease	Leptospirosis
Agent	*Leptospira interrogans*, tightly coiled spirochetal bacteria
Epidemiology	Worldwide; most common in tropical and subtropical areas with abundant rainfall; peak incidence in summer or fall; **100-200 cases reported annually in the United States**
Reservoir	Rodents such as rats, wild animals, pets such as dogs
Transmission	Direct or indirect exposure to the urine of an infected animal; entry through skin; large outbreaks can occur during flooding
Clinical features	Range in severity, from mild febrile illness to fulminant life-threatening; sudden onset fever, chills and headache, muscle pain (calves); **jaundice, renal failure, hemorrhage (Weil's disease)**
Pathogenesis	Penetrate skin, hematogenous dissemination (contrasts *with B. burgdorferi* and *T. pallidum*); hepatocellular dissociation, pulmonary hemorrhage, interstitial nephritis
Diagnosis	Serology, detection in tissues

Case 5

- 56 yo M from CA
- 14 mos s/p aortic valve replacement and root repair
- Admitted to hospital with 4 mos of fever, chills, weight loss, pancytopenia
- AST 85, ALT 105, ALK 223, tbili 0.7
- Liver bx was performed
- Patient treated w/Abx but died 1 month after presentation

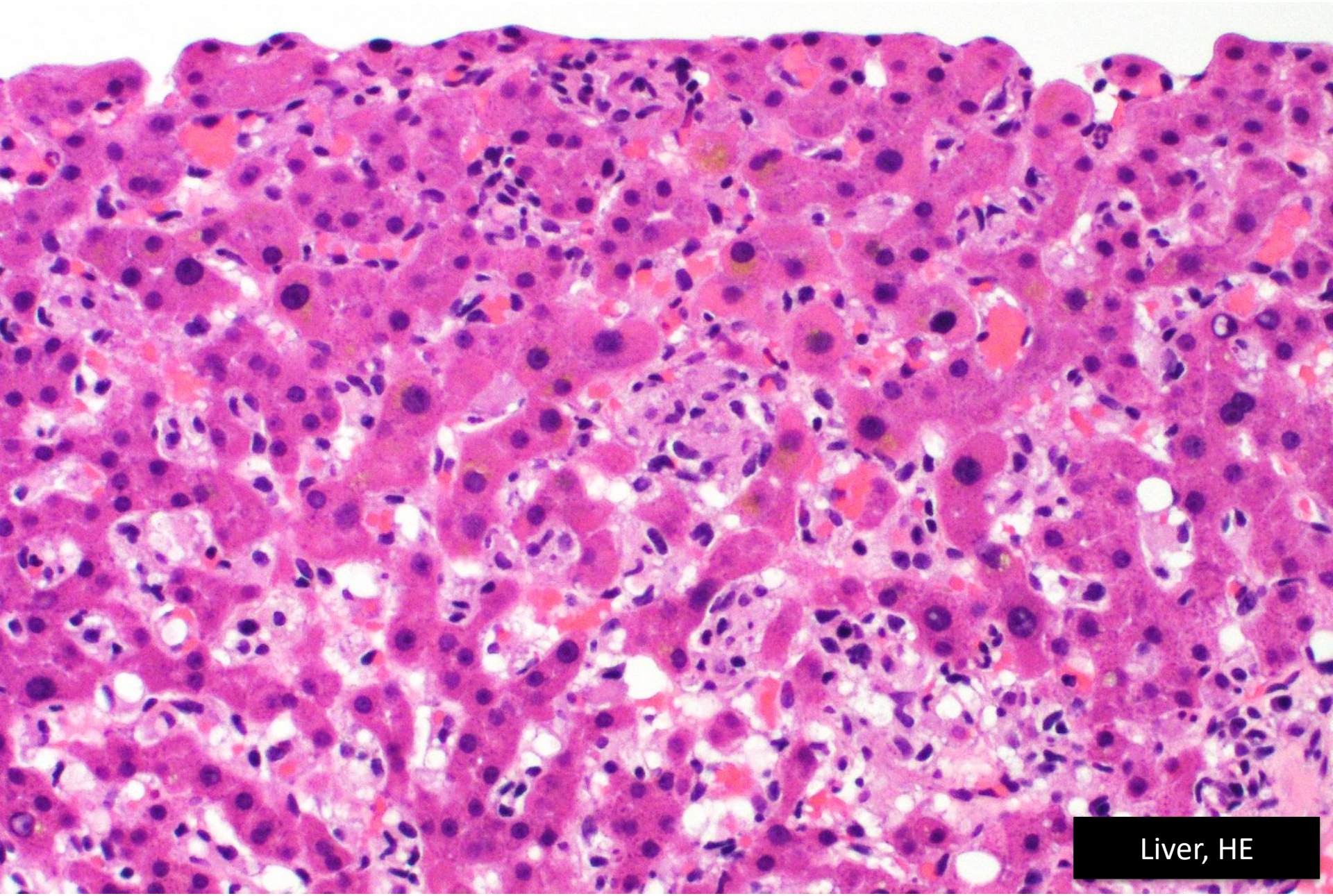


Photo credit: Dr. Nafis Shafizadeh

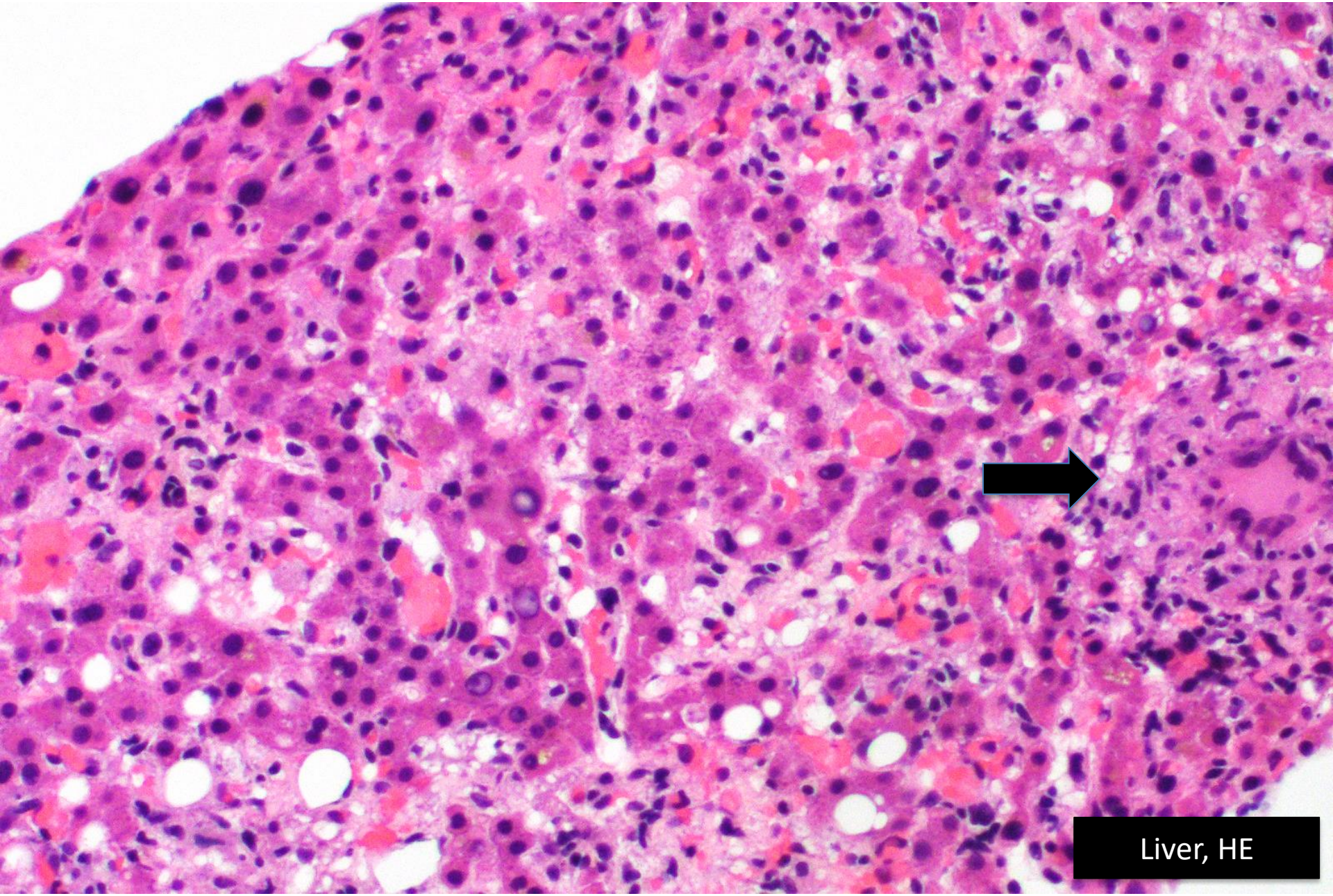


Photo credit: Dr. Nafis Shafizadeh

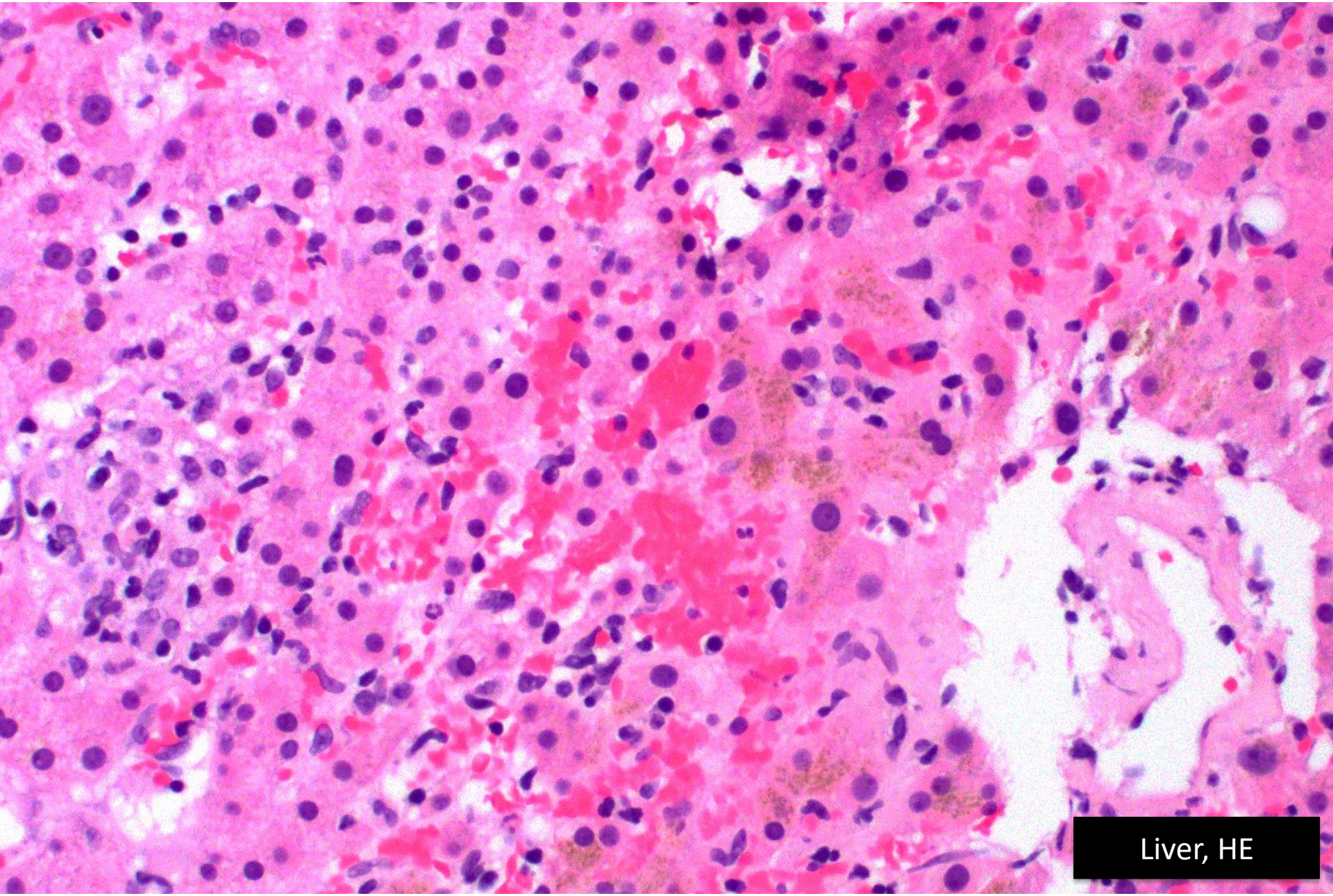


Photo credit: Dr. Nafis Shafizadeh

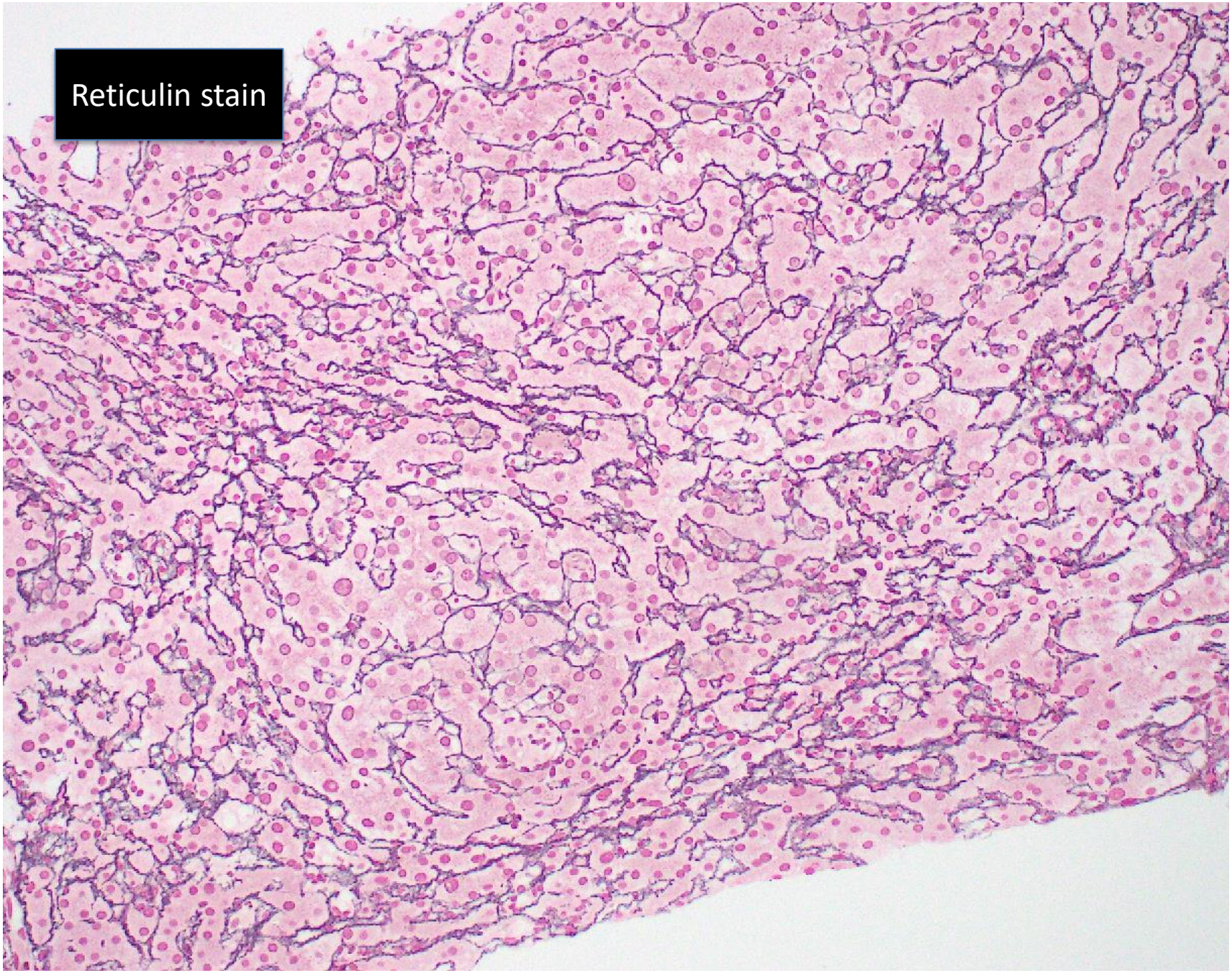
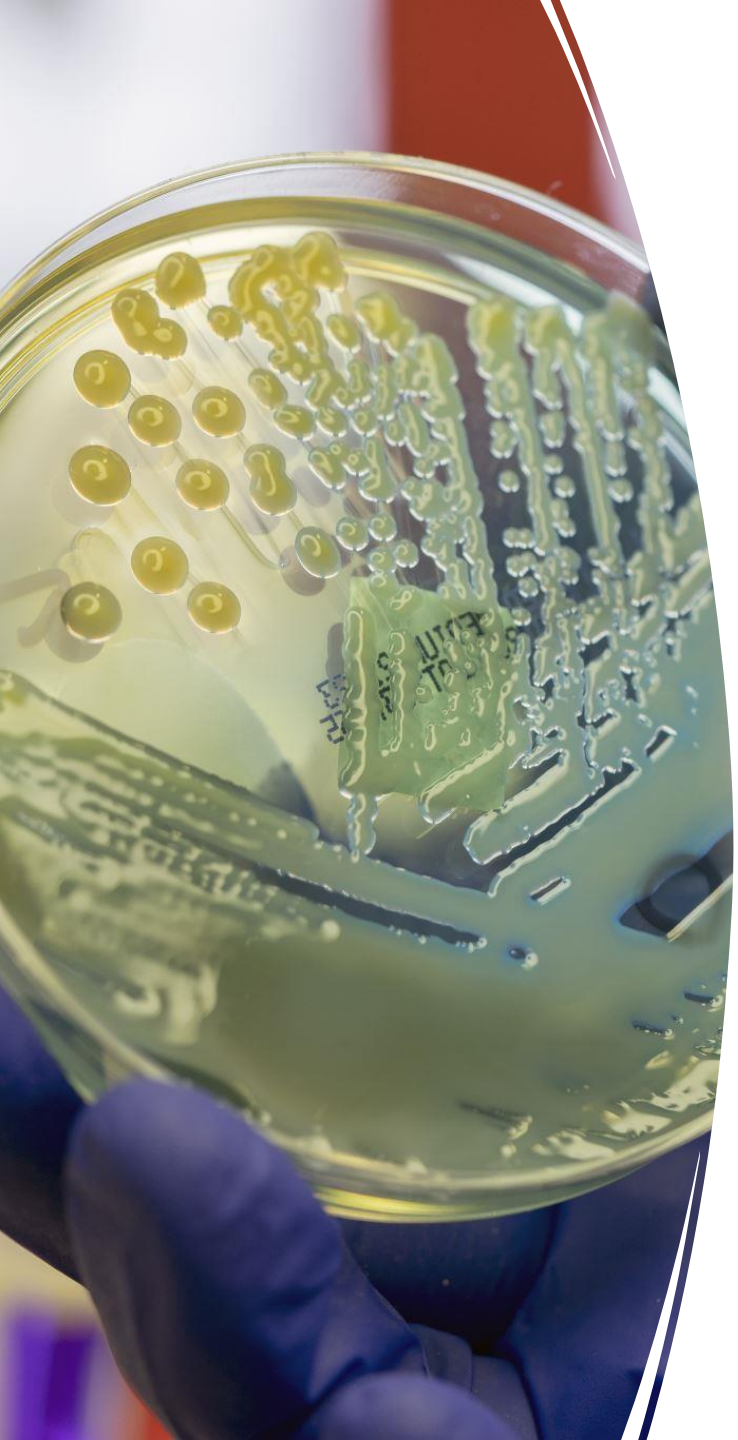


Photo credit: Dr. Nafis Shafizadeh

Case 5 Workup

- Fungal infection (GMS stain)
- Mycobacteria (AFB stain)
- Drug
- Sarcoid



Microbiology laboratory

Cultures from liver tissue revealed ***Mycobacterium chimaera***

Mycobacterium chimaera Hepatitis

A New Disease Entity

Nafis Shafizadeh, MD, Gillian Hale, MD, MPH,† Julu Bhatnagar, PhD,†*
Najeeb S. Alshak, MD, and Jim Nomura, MD‡*

- Case series: 7 patients with liver biopsies, all with sinusoidal histiocyte collections
- Poorly-formed sinusoidal granulomas and features of venous outflow obstruction
- 5/7 patients died (71%)

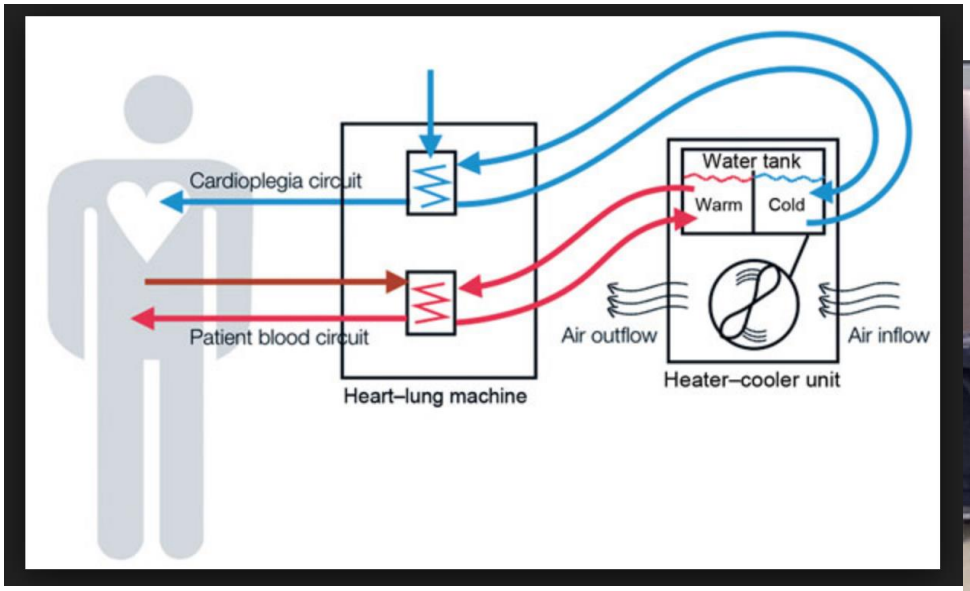
TABLE 2. Pathologic Features of *Mycobacterium chimaera* Hepatitis

Histologic Findings	Cases (n = 7)
Sinusoidal histiocyte collections	7 (100)
Mild	1 (14)
Moderate	5 (72)
Marked	1 (14)
Multinucleated giant cells	6 (86)
Rare (≤2)	5 (72)
Occasional (3-5)	1 (14)
Many (>5)	0
Sinusoidal dilatation and congestion	7 (100)
Patchy (≤20% of the biopsy)	1 (14)
Obvious (>20% of the biopsy)	6 (86)
Fibrosis	2 (29)
Pericellular	2 (29) (mild)
Portal	0
Bridging	0
Large and/or necrotizing granulomas	0
Foamy macrophage collections	0
Portal granulomas	0
Fibrin ring granulomas	0
Suppurative granulomas	0
NRH	2 (29)
Acid-fast stain positive	1 (14)

Values represent number (percentage).

Shafizadeh N, Hale G, Bhatnagar J, Alshak NS, Nomura J. Mycobacterium chimaera Hepatitis: A New Disease Entity. Am J Surg Pathol. 2019 Feb;43(2):244-250.

M. chimaera transmission

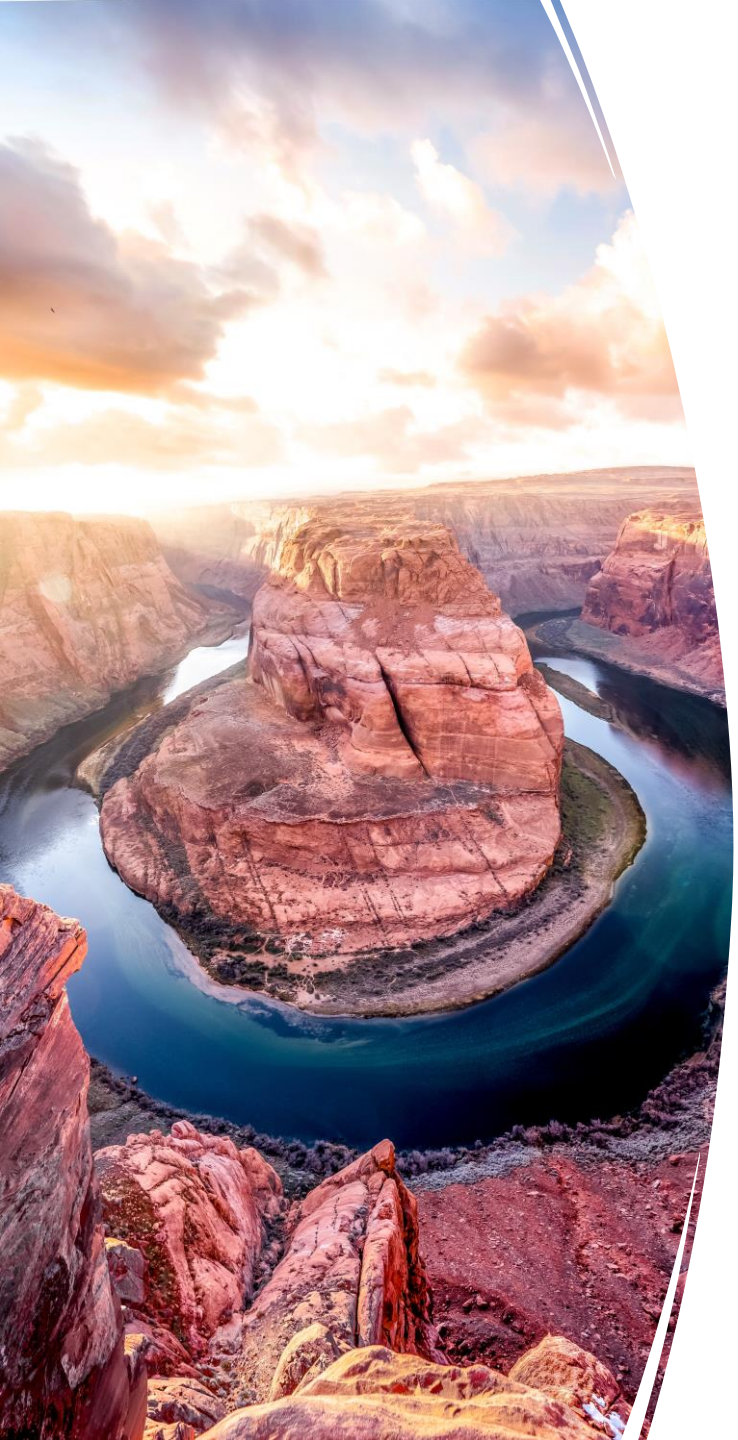


Sommerstein R et al. Transmission of Mycobacterium chimaera from Heater-Cooler Units during Cardiac Surgery despite an Ultraclean Air Ventilation System. Emerg Infect Dis. 2016 Jun;22(6):1008-13.

Chand M et al. Insidious Risk of Severe Mycobacterium chimaera Infection in Cardiac Surgery Patients. Clin Infect Dis. 2017 Feb 1;64(3):335-342.

Mycobacterium chimaera

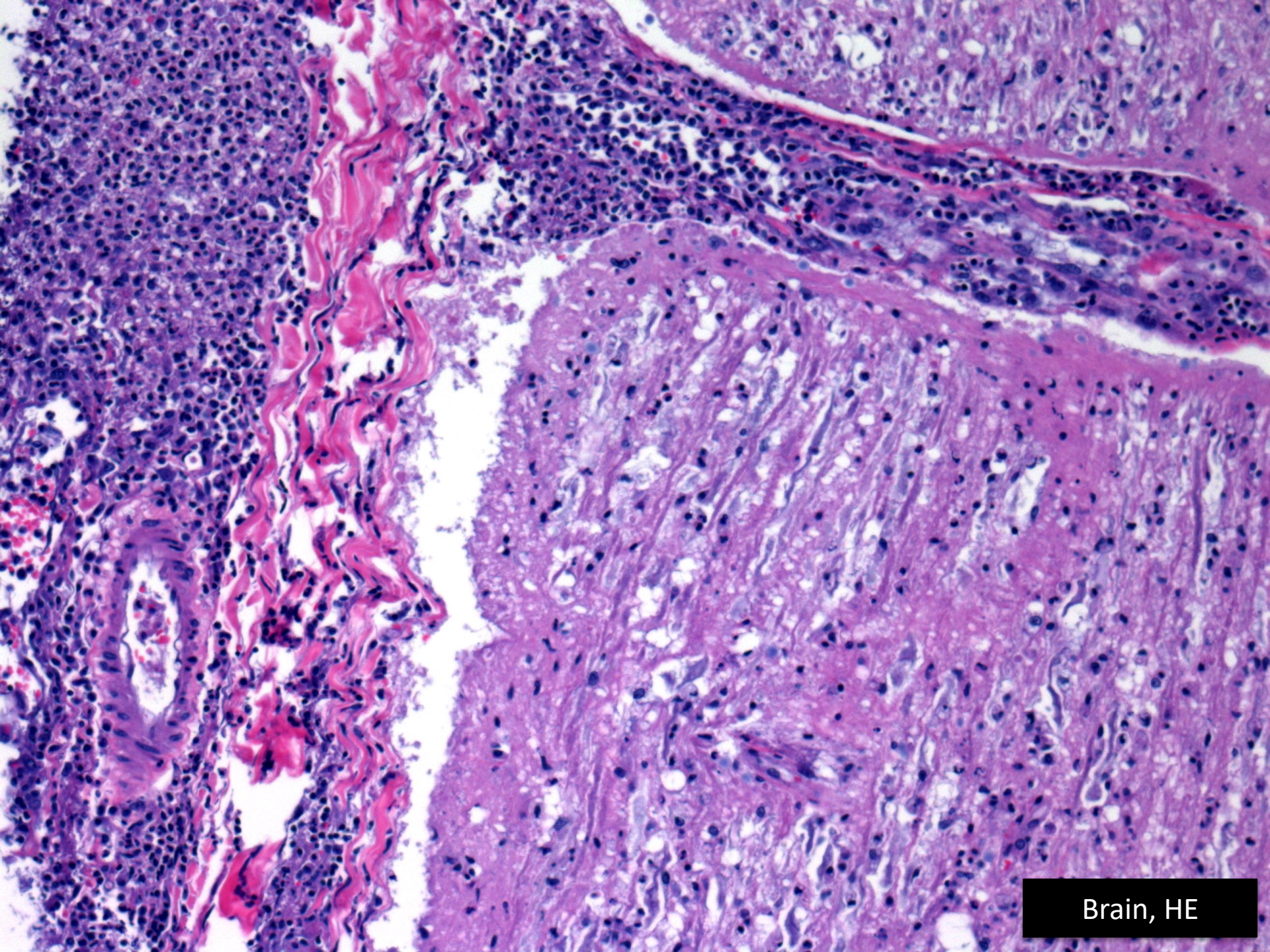
Disease	***M. chimaera* hepatitis, endocarditis, septsis**
Features	Slow-growing non-tuberculous mycobacteria within the Mycobacterium avium complex, originally associated with pulmonary infections
Epidemiology	120 cases of systemic *M. chimaera* infection have been reported around the world; identified in used and new LivaNova Stockert 3T HCU systems implicating the manufacturing process
Transmission	Aerosolization of bacteria into surgical field from contaminated heater cooler units (exhaust fan) used during open heart surgeries
Clinical features	Long latency between manifestation of symptoms and initial exposure; nonspecific symptoms, ie. fever, weight loss Mortality rates associated with M. chimaera are high (50-60%) and carry a 45% 5-year survival rate
Pathology	Poorly-formed sinusoidal granulomas, rare multinucleated giant cells, nodular regenerative hyperplasia changes, sinusoidal fibrosis c/w sinusoidal obstructive etiology
Diagnosis	Culture fresh tissue, PCR on the culture specimen and/or formalin fixed paraffin-embedded tissue samples with 16S riboxomal RNA (rRNA) gene sequencing.

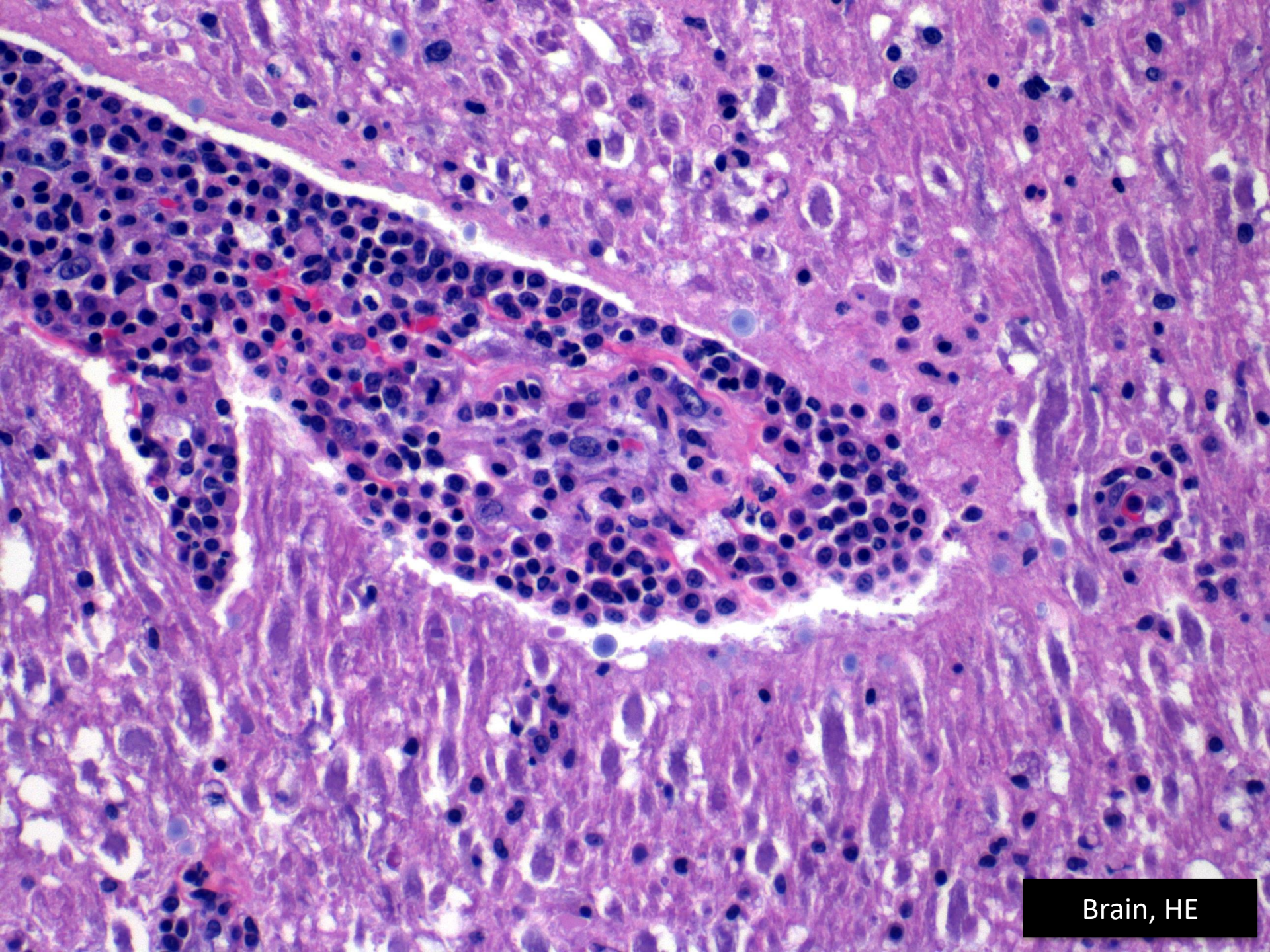


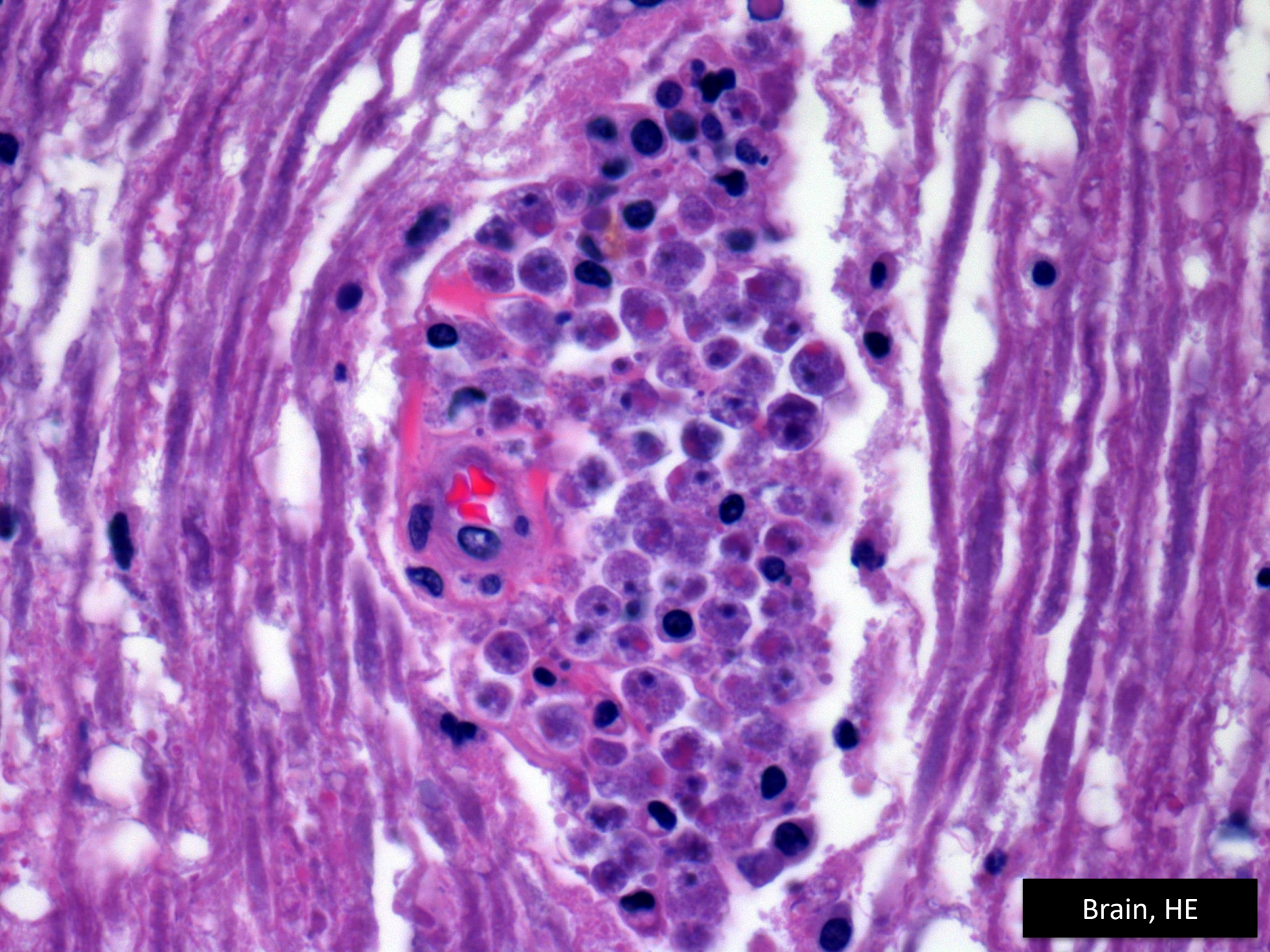
Case 6

- 24 yo F presented to ED with headache in August, one week after swimming in the Colorado River
- CT scan unremarkable; sent home
- Returned to ED the next day with more severe headache, fever, photophobia & vomiting
- Symptoms, LP and CSF analysis all suspicious for bacterial meningitis
- All cultures – no growth
- Died 4 days after presentation

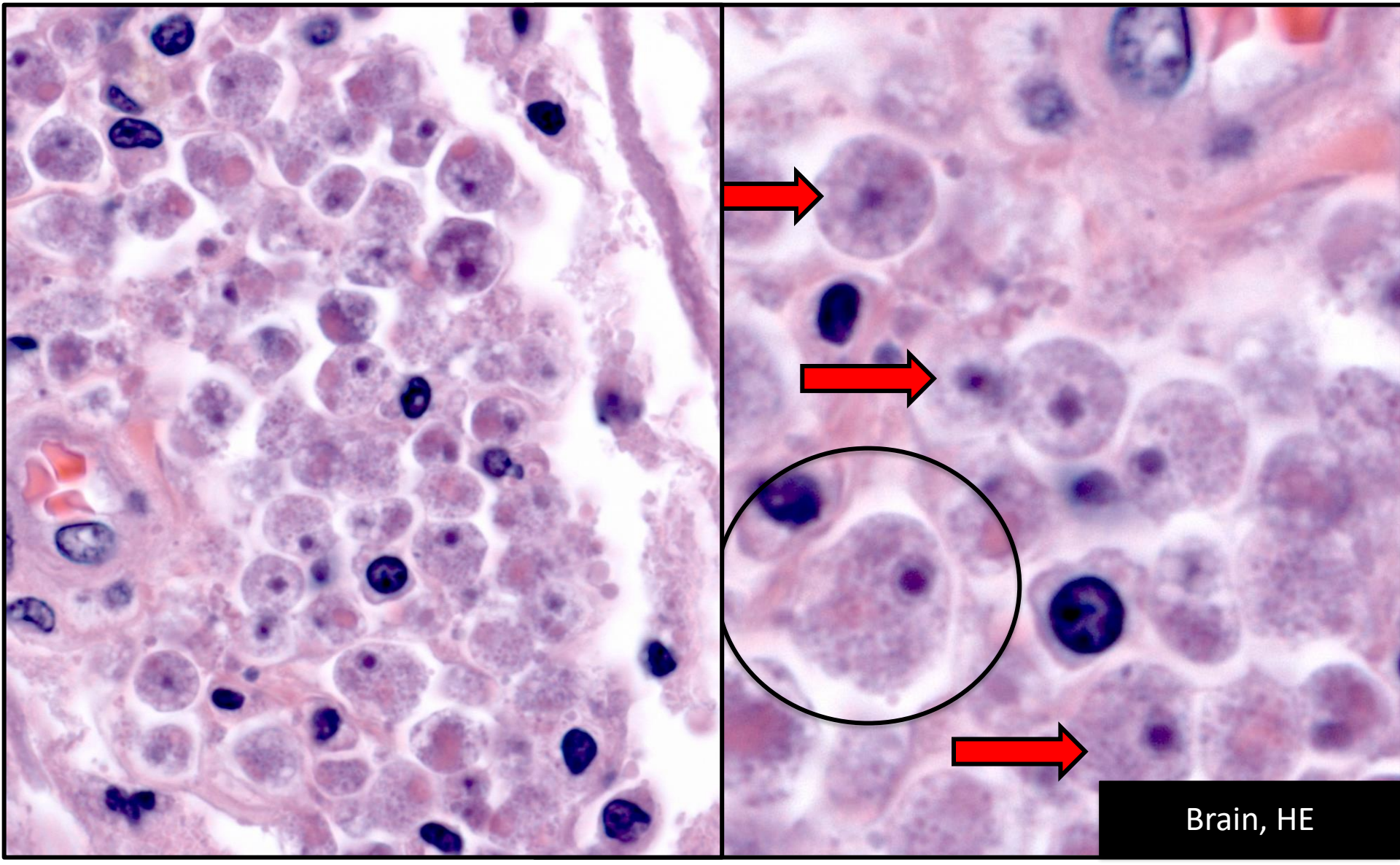
Gross images courtesy of S.D.

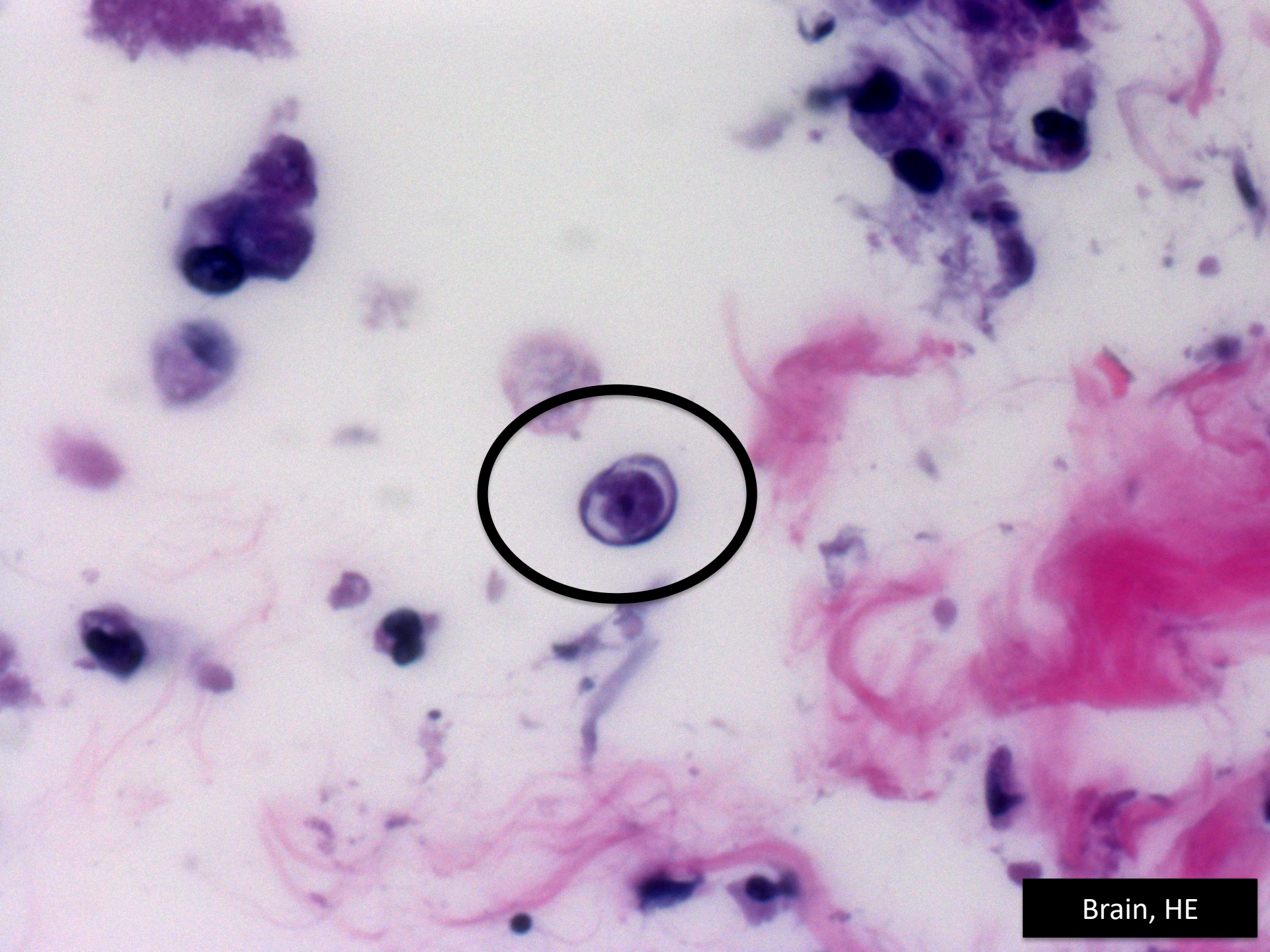

Brain, HE


Brain, HE


Brain, HE

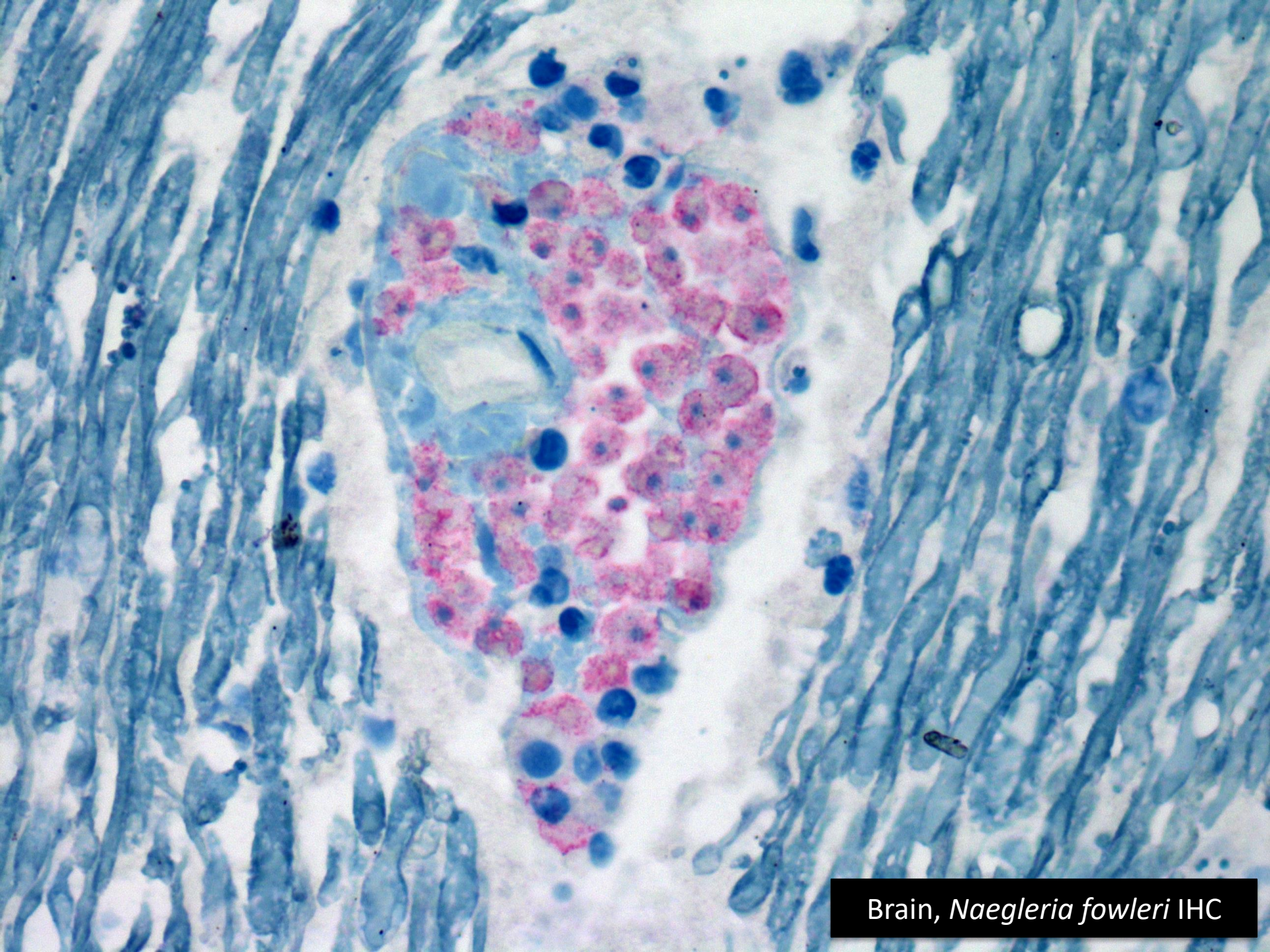
Case 7




Brain, HE

Free Living Ameba

	Naegleria fowleri	*Acanthameba spp &* *Balamuthia mandrillaris*
Disease pathology	**Primary amebic meningoencephalitis**	**Granulomatous amebic encephalitis, Acanthameba keratitis**
Route of infection	Olfactory neuroepithelium	Enter through lungs, skin, or mucous membranes, disseminate to the brain hematogenously
Organs targeted	Brain	Brain, skin, eyes, lungs
Epidemiology	Immunocompetent; warm freshwater and soil	Immunocompromised; soil, fresh and seawater, sewage, pools
Clinical presentation	Rapid; typically fatal within 10 days	Slow, insidious
CNS forms	Trophozoites (10-15 um), rare cysts	Trophozoites (25-40 um) and cysts (double walled)
Diagnostic Testing	PCR (CSF), IHC and PCR on tissues	PCR (CSF), IHC and PCR on tissues



Brain, *Naegleria fowleri* IHC

Case 7 Workup

Pathology	Necrotizing meningo-encephalitis in an *immunocompetent* patient
Diagnostic features	Numerous amebic trophozoites and rare cyst forms seen
Special stains	Can be helpful (GMS) to identify cyst forms; not performed in this case
Immunohistochemistry	*Naegleria fowleri*. Does not cross react with Acanthamoeba or Balamuthia
Molecular	DNA extracts from formalin-fixed CNS tissues sent to Free Living Ameba Laboratory; *Naegleria fowleri* confirmed

Case Summary

1. **Yellow fever**
- Pauci-inflammatory hepatitis with Councilman bodies in travel to SA

2. **Leishmaniasis**
- Differentiate from *Histoplasma* and *Toxoplasma*

3. **Prototheca**
- GMS stain, endosporulation, “Mercedes Benz” morphology

4. **Leptospirosis**
- Triad: hepatocellular dissociation, pulmonary hemorrhage, interstitial nephritis

5. ***M. chimaera* hepatitis**
- History of open-heart surgery

6. ***Naegleria fowleri***
- Immunosuppression vs. immunocompetent

QUARANTINE

CONTAGIOUS DISEA

NO ONE SHALL ENTER OR L EN PERMISSION
OF THE LOCAL H .C.S.)
NO PE TEE OF THE HEALTH DISTRICT SHALL
ALTE HIS CARD. (Art. 4477 - V.A.C.S.)

ONE VIOLATING THIS REGULATION WILL BE FINED NOT LESS THAN $10.00 NOR MORE THAN $1,000.00 FOR EACH VIOLATION. (ART. 770 Texas-Penal Code)

BY ORDER OF

DIRECTOR OF HEALTH

SAN ANTONIO METROPOLITAN HEALTH DISTRICT

Thank you!