

13 Protozoa That Could Be Tested

Speaker: Edward Mitre, MD

IDBR

INFECTIOUS

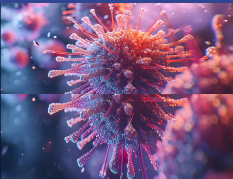
DISEASE

BOARD REVIEW

DISEASE

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AUGUST 16-20, 2025



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Edward Mitre, MD
Rockville, MD

6/30/2025

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Disclosures of Financial Relationships with Relevant Commercial Interests

- None

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Protozoa

| <u>Protozoa - Extraintestinal</u> | <u>Protozoa - Intestinal</u> |
|---|--|
| Apicomplexa Plasmodium (Babesia) (Toxoplasma) | Apicomplexa Cryptosporidium Cyclospora Cystoisospora |
| Flagellates Leishmania Trypanosomes (Trichomonas) | Flagellates Giardia Dientamoeba |
| Amoebae Naegleria Acanthamoeba Balamuthia | Amoebae Entamoeba |
| | Ciliates Balantidium |

Maybe Not Protozoa **Fungi Kingdom:** Microsporidiosis agents
SAR supergroup: Blastocystis

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Question #1

PREVIEW QUESTION



A 54-year-old woman presents with fever, chills, and oliguria one week after travel to Malaysia.

Vitals: **39.0°C**, HR 96/min, RR 24/min, **BP 86/50**

Labs: Hct 31%, platelets 14,000/μl, Cr of 3.2 mg/dL.

Peripheral blood smear has intraerythrocytic forms that are morphologically consistent with *Plasmodium malariae*.

What is the most likely infectious agent causing the patient's illness?

- A. *Plasmodium malariae*
- B. *Plasmodium knowlesi*
- C. *Plasmodium vivax*
- D. *Plasmodium falciparum*
- E. *Babesia microti*

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P. knowlesi

Morphologically similar to *P. malariae*

Usually a parasite of long-tailed macaques



Increasingly recognized in Myanmar, Philippines, Indonesia, and Thailand

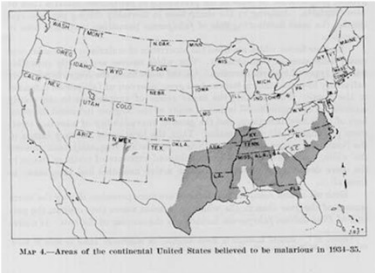
Causes high parasitemia

Highly morbid and can be lethal

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Malaria

One of the most important pathogens in the history of the world

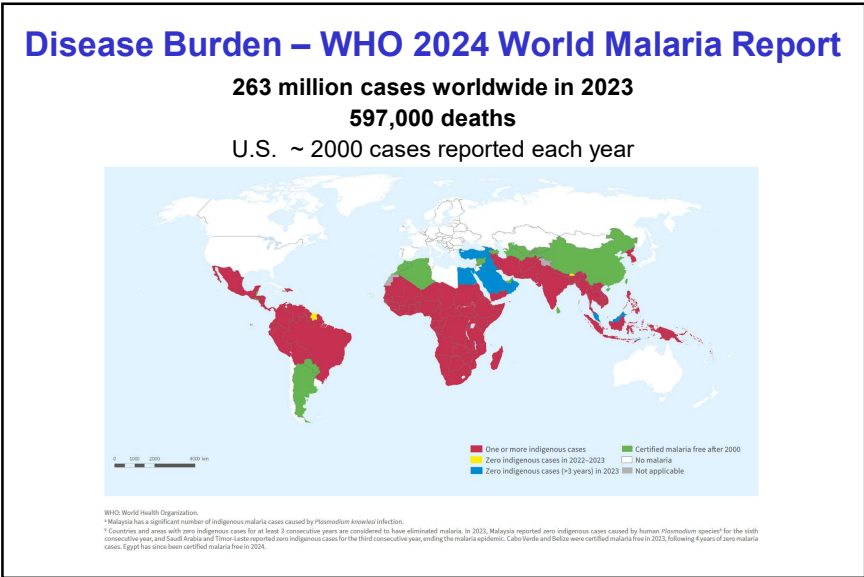


National Malaria Elimination Program: 1947- 1951
→ DDT spraying, drainage of wetlands
→ Atlanta was chosen for the Office of Malaria Control in War Areas (the predecessor agency of the CDC) in part because of its location in a malaria-endemic region

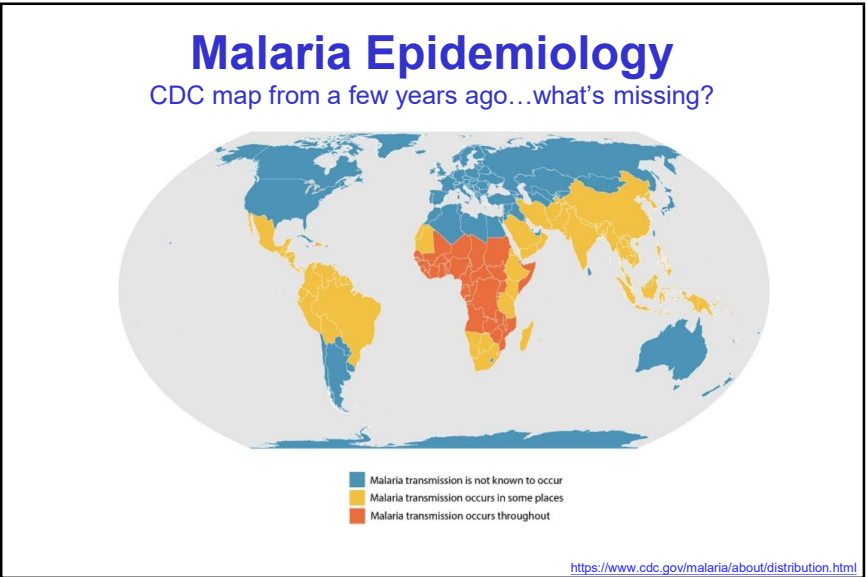


In 1775 the Continental Congress purchased quinine for George Washington's troops

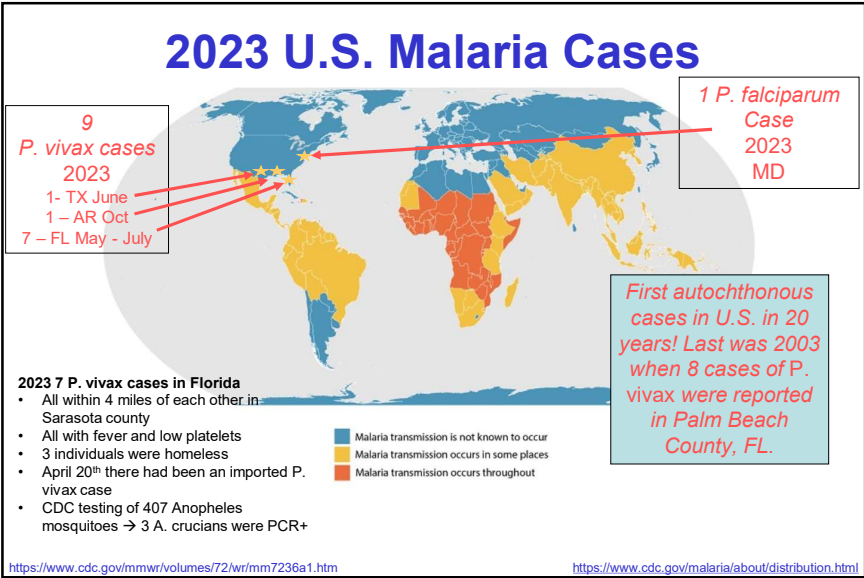
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In non-immune patients, falciparum malaria is a medical emergency!!

One of the most common causes of fever in a returned traveler

Infected individuals can rapidly progress from appearing well to being critically ill

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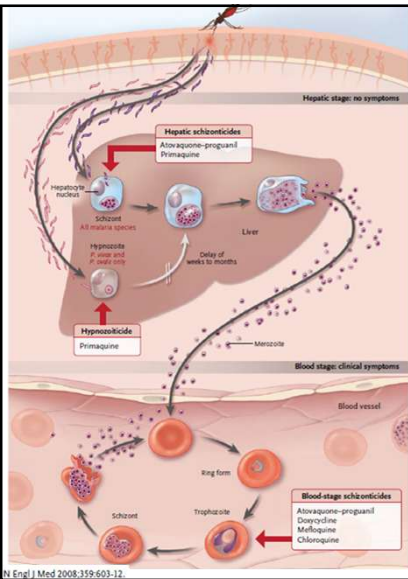
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Some Helpful Heuristics

| If patient has | Make sure patient doesn't have |
|------------------------------------|--------------------------------|
| Fever and freshwater contact-----> | Malaria |
| Fever and unpasteurized milk-----> | Malaria |
| Fever and undercooked meat-----> | Malaria |
| Fever and raw vegetables-----> | Malaria |
| Fever and untreated water-----> | Malaria |
| Fever and wild dog bite-----> | Malaria |
| Fever and abdominal pain-----> | Malaria |
| Fever and headache-----> | Malaria |
| Fever and diarrhea-----> | Malaria |
| Fever and cough-----> | Malaria |
| Fever and dysuria-----> | Malaria |

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- Sporozoites**
- Infective stage
 - Come from mosquito
- Liver schizont**
- Asymptomatic replicative stage
 - Become 10,000 to 30,000 merozoites
- Hypnozoite**
- Dormant liver stage in vivax and ovale
 - Release merozoites weeks to months after primary infection
- Merozoites**
- Infect RBCs and develop into ring-stage trophozoites
 - Mature into schizonts, which release merozoites which infect more RBCs
- Gametocytes**
- Infective stage for mosquitoes

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Characteristics of Human Malaria Species

| | P. falciparum | P. knowlesi | P. vivax | P. ovale | P. malariae |
|-------------|---------------|-------------|----------|----------|-------------|
| incubation | 8 - 25 d | prob 8-25 d | ~ 2 wks | ~ 2 wks | ~ 3-4 wks |
| hypnozoite | no | no | yes | yes | no |
| RBC age | any | any | young | young | old |
| parasitemia | high | high | < 2% | < 2% | < 1% |
| morbidity | high | high | high | moderate | low |
| mortality | high | moderate | low | low | low |

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Possible Evolutionary Defenses Against Malaria

- Duffy antigen negative**
(P. vivax uses Duffy Ag to enter RBCs)
- Sickle cell trait**
(increases survival during P. falciparum infection, perhaps by selective sickling of infected RBCs)
- Glucose-6-phosphate dehydrogenase deficiency**
(malaria parasites grow poorly in G6PD deficient RBCs, perhaps b/c this results in an overall increase in reactive oxygen species in RBCs)

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Uncomplicated (Mild) Malaria

Symptoms:

- Fevers, chills, headache, fatigue
- *NOTE: abdominal pain presenting symptom in 20%

→ Periodicity of fevers not common when patients seen acutely

Labs:

- Thrombocytopenia in 50%
- Mild anemia in 30%
- Typically, no leukocytosis
- May see evidence of hemolysis with mild increase T bili and LDH

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Complicated (Severe) Malaria

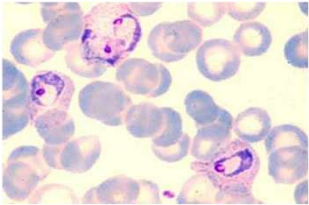
- Cerebral malaria (altered mental status, seizures)
- Respiratory distress/pulmonary edema
- Severe anemia (hct <15% in children, <20% in adults)
- Renal failure
- Hypoglycemia
- Shock (SBP < 80 mm Hg or capillary refill > 3 seconds)
- Acidosis (often lactic acidosis)
- Jaundice (total bilirubin > 3 mg/dL)
- Bleeding disorder (spontaneous bleeding or evidence of DIC)

Often seen in children of endemic countries. Adults more often get multiorgan failure.

These complications primarily occur with *Plasmodium falciparum*, usually when parasitemia ≥ 2%.
NOTE: in the absence of end organ damage, parasitemia ≥ 5% is often used as the cut-off to treat for severe malaria in the U.S.

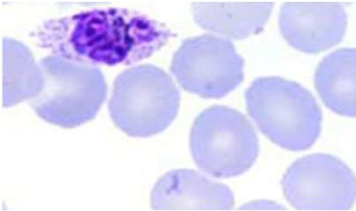
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P. vivax or *ovale*



- Intracellular schüffner's dots
- Enlarged infected cells

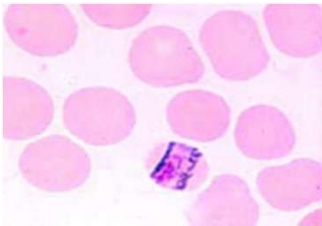
P. ovale



- Elongated or oval
- 6-12 merozoites (vs 12-24 for vivax)

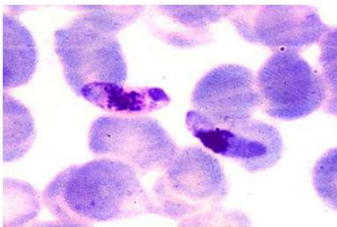
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P. malariae



- Band form (also seen in *P. Knowlesi*)

P. falciparum



- Banana shaped gametocyte

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Malaria: Diagnosis

Rapid diagnostic antigen tests

→ sensitivity > 90% for *P. falciparum*
(about 85% for *P. vivax*, lower for *P. knowlesi* and *P. ovale*)



Binax Now® ICT assay for the detection of *Plasmodium falciparum* malaria according to the level of parasitemia

| Parasitemia (no. of parasites/μL of whole blood) | Microscopy (no. positive) | NOW ICT (no. positive) | Sensitivity (%) |
|---|------------------------------|---------------------------|--------------------|
| 1–100 | 4 | 3 | 75.0 |
| 101–1,000 | 26 | 25 | 96.2 |
| 1,001–10,000 | 37 | 36 | 97.3 |
| >10,000 | 34 | 33 | 97.1 |

Am. J. Trop. Med. Hyg., 69(6), 2003, pp. 589–592

For *P. falciparum* (T1) → tests for histidine-rich protein 2
For all species (T2) → tests for aldolase

*Most false-negative antigen tests are due to low parasite burden
→ Retest suspected patients that initially test negative

*Increasing false negative cases occurring worldwide due to mutations in HRP2

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Question #2

PREVIEW QUESTION



A 33-year-old woman is traveling to Uganda to do field studies in anthropology. She is two months pregnant.

Which of the following do you prescribe for malaria prophylaxis?

- A. Doxycycline
- B. Chloroquine
- C. Mefloquine
- D. Atovaquone/proguanil
- E. No prophylaxis

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Question #2

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- C. Mefloquine
- D. Atovaquone/proguanil
- E. No prophylaxis

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Malaria Chemoprophylaxis

(note: no malaria vaccine approved for U.S. travelers)

SOME REGIONS IN CENTRAL AMERICA and the MIDDLE EAST

| | Pre-Exposure | During | Post-Travel |
|-----------------------------------|-------------------|----------|------------------|
| Chloroquine 500mg tabs | 1 tab/wk x 2 wks | 1 tab/wk | 4 weeks |
| EVERYWHERE | | | |
| Atovaquone/proguanil 250/100mg | 1 tab daily x 2 d | 1 daily | 7 days |
| Doxycycline 100mg tabs | none | 1 daily | 4 weeks |
| Tafenoquine* | 2 tab daily x 3 d | 2 tab/wk | 2 tab after 1 wk |
| 100mg tabs | | | |
| Mefloquine (not SE Asia)** | 1tab/wk x 2-3 wks | 1 tab/wk | 4 weeks |
| 250mg tabs | | | |

* **Tafenoquine can precipitate severe hemolytic anemia in individuals that are G6PD deficient**

** **FDA black box warning mefloquine can cause neurologic symptoms, hallucinations, and feelings of anxiety, mistrust, and depression. Can also cause QT prolongation. Thus, many U.S. practitioners now reserve mefloquine for pregnant travelers to areas with chloroquine resistance**

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P. falciparum Treatment

Excellent review → 2022 JAMA, 328(5):460-47, PMID: 3591684

Uncomplicated P. falciparum malaria (no organ dysfunction, low parasitemia, able take po)

If chloroquine sensitive area → Chloroquine or hydroxychloroquine

If not chloroquine sensitive area (most cases) → **Artemether/lumefantrine (Coartem)**
ACTs are treatment of choice, WHO 2022 guidelines
Alternatives if artemether/lumefantrine not available:
Atovaquone/proguanil (Malarone), quinine + doxycycline, mefloquine

Severe Malaria

→ IV artesunate (CDC malaria hotline: 770-488-7788)

NOTES

1. Treatment failures can occur with artemether/lumefantrine, especially when > 65 kg
Sonden K. et al, *Clinical Infectious Diseases* 2017 PMID: 27986683
2. Artemisinin resistance reported in SE Asia (Cambodia, Laos, Myanmar, Thailand, Vietnam), parts of Africa (Uganda, Rwanda), and in S. America (Guyana)
3. Delayed-onset anemia in 2.7% of U.S. patients after treatment with artesunate
Abanyie F. et al, *Clinical Infectious Diseases* 2022 PMID: 36052468
4. Hypoglycemia and ARDS are complications that can occur during treatment of malaria.
ARDS sometimes develops even as pt improving from malaria.

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P. vivax/P. ovale Treatment

Chloroquine x 3 days, or ACT (artemether/lumefantrine in U.S.)

Note: PNG, Indonesia, Oceania have CLQ R P. vivax → use ACT

Then ANTIRELAPSE THERAPY with primaquine or tafenoquine

→ Need to check G6PD status before administering primaquine OR tafenoquine
(as both can cause severe hemolysis in patients with G6PD deficiency)

→ Both primaquine and tafenoquine contraindicated during pregnancy

• **Primaquine – weight-based dosing and duration as determined by G6PD activity**

ALWAYS LOOK UP DOSING BEFORE ADMINISTERING

→ Usually 30 mg primaquine base per day x 14 days if normal G6PD activity

→ Do not exceed 30 mg primaquine base per day

→ If over 70 kg, can calculate total dose 6 mg/kg and then extend duration of 30 mg daily doses until total goal met

→ if G6PD deficient consider weekly chloroquine x 1 year

• **Tafenoquine (two 150 mg tabs once, given on 1st or 2nd day of chloroquine therapy)**

(Tafenoquine was approved for radical cure of P. vivax in 2018, P. ovale treatment is off-label)

2020: Company (GSK) reported some failures when tafenoquine was used after ACT treatment of P. vivax.

NEW FDA LABELING: **Tafenoquine now only approved and recommended after chloroquine treatment**

<https://www.cdc.gov/malaria/hcp/clinical-guidance/treatment-uncomplicated.html>

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Suggestions for All ID Practitioners

1. Make sure the facility where one works has the means to rapidly test for malaria
2. Ensure that hospital pharmacy has access to appropriate medications for treatment of malaria

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Babesia

Transmission

- Ixodes ticks in Northeast and upper midwest
→ co-infection with Lyme and Anaplasma

Transfusion

(Ab screening tests approved by FDA in 2018)

Symptoms:

 fever, headache, chills, myalgias

less common: nausea, dry cough, neck stiffness, vomiting, diarrhea, arthralgias

→ severe disease: in HIV, asplenia

Labs: anemia, thrombocytopenia, mild increase LFTs, normal/low/high WBC

Diagnosis: small ring forms in RBCs, PCR, Ab
merozoites can make tetrad ("Maltese cross")

Treatment: azithromycin + atovaquone
(clindamycin + quinine is alternative)

→ Exchange transfusion for severe disease



CDC DpDx

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13 Protozoa That Could Be Tested
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Protozoa

Protozoa - Extraintestinal

Apicomplexa
Plasmodium (Babesia) (Toxoplasma)

Flagellates
Leishmania
Trypanosomes (Trichomonas)

Amoebae
Naegleria
Acanthamoeba
Balamuthia

Protozoa - Intestinal

Apicomplexa
Cryptosporidium
Cyclospora
Cystoisospora

Flagellates
Giardia
Dientamoeba

Amoebae
Entamoeba

Ciliates
Balantidium


Maybe Not Protozoa Fungi Kingdom: Microsporidiosis agents
SAR supergroup: Blastocystis

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Leishmaniasis

→ Obligate intracellular protozoan infection
→ Transmitted by sand flies (noiseless, active in evenings)

Lutzomyia
New world leishmaniasis



Phlebotomus
Old world leishmaniasis




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Leishmaniasis Life Cycle – Two Stages

Promastigote

Extracellular, in sand fly
2 µm wide x 20 µm long

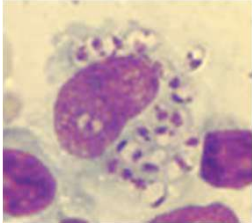
- Flagella
- Large central nucleus
- Band shaped kinetoplast



Amastigote

Intracellular (macrophages)
Round or oval
Wright-Giemsa:

- Dark-purple nucleus
- Small rod shaped kinetoplast



CDC DpDx

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Question #3

A 42-year-old man from Bolivia presents with nasal stuffiness and is found to have nasal septal perforation. Biopsy demonstrates intracellular amastigotes consistent with Leishmania.

Which is the most likely species?

A. *L. mexicana*

B. *L. braziliensis*

C. *L. peruviana*

D. *L. infantum chagasi*

E. *L. major*

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- C. *L. peruviana*
- D. *L. infantum chagasi*
- E. *L. major*

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Leishmania Taxonomy and Disease Simplified

| | Cutaneous | Mucosal | Visceral |
|----------------------------|-----------|---------|----------|
| NEW WORLD | | | |
| <i>L. mexicana complex</i> | X | | |
| <i>L. braziliensis</i> | X | X | |
| <i>L. infantum chagasi</i> | | | X |
| OLD WORLD | | | |
| <i>L. tropica</i> | X | | |
| <i>L. major</i> | X | | |
| <i>L. donovani</i> | | | X |
| <i>L. infantum chagasi</i> | | | X |

*Note: *L. braziliensis* is in the Viannia subgenus. *L. V. guyanensis* and *L. V. panamensis* also cause mucosal disease. *L. peruviana* DOES NOT

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Cutaneous Leishmaniasis:
Clinical Presentation

- Papule → nodule → ulcerative lesion → atrophic scar
 - Ulcerative lesion may have:
 - Induration
 - Scaliness
 - Central depression
 - Raised border
- Takes weeks to months to develop
- Usually painless, unless superinfected
- Most lesions will eventually resolve on their own



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**Cutaneous Leishmaniasis:
Diagnosis**

Definitive diagnosis is very helpful because:

1. Allows you to rule out other possibilities
2. May help in deciding whether and how to treat

Diagnostic Tools:

Edge of ulcer skin: scraping, aspirate, punch

- Touch prep with examination under oil looking for amastigotes
- Culture on triple N media (may take weeks to grow)
 - (Nicolle's modification of Novy and MacNeal's medium – biphasic)
- Histology
- PCR

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Cutaneous Leishmaniasis: Treatment Recommendations

- Treat systemically if *L. (V.) braziliensis, guyanensis, panamensis*
- If not, ok to observe if there are:
 - Few lesions, they are < 5 cm, not on face/fingers/toes/genitals, normal host, no subcutaneous nodules

Treatment Options

- Local: Heat with radiotherapy (FDA approved), cryotherapy, intralesional therapy systemic
- Oral: Miltefosine for certain species, especially New World CL species
Ketoconazole, fluconazole (off-label)
- IV: Liposomal amphotericin B (off-label)
pentavalent antimony (*meglumine antimoniate*, *ASTMH website has instructions for obtaining on IND from Sanofi*)

2016 IDSA GUIDELINES FOR TREATMENT OF LEISHMANIA
http://www.idsociety.org/Guidelines/Patient_Care/IDSA_Practice_Guidelines/Infections_by_Organism/Parasites/Leishmaniasis/

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Mucosal Leishmaniasis

Leishmania (Viannia) braziliensis, guyanensis, panamensis

- Dissemination to nasal mucosa
- Slow, progressive, destructive
- Can occur months or years after cutaneous ulcer



Treatment:

- Oral miltefosine (FDA approved for *L. braziliensis*)
- IV lip. amphotericin (off-label)
- IV antimony (no longer commercially available)

Miltefosine notes

Side effects: nausea, vomiting, diarrhea, increased AST/ALT
Contraindicated in pregnancy, use contraception for 5 months after treatment ($t_{1/2} = 30\text{ d}$)

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Visceral Leishmaniasis

- *L. donovani* (South Asia, East Africa)
- *L. infantum chagasi* (Middle East, Central Asia, Mediterranean, Central and S. America)
- Amastigotes in macrophages go to local LNs then hematogenously to liver, spleen, bone marrow

A persistent disease that can reactivate
TNF blockade, HIV CD4 < 200

- Wks/months of fevers, chills, hepatosplenomegaly
- Pancytopenia & hypergammaglobulinemia



Diagnosis: PCR, culture, or histopathology for intracellular amastigotes
→ bone marrow aspirate preferred, can also check LN, spleen, or buffy coat
Antibody to rK39 recombinant Ag (dipstick test)

Treatment: Liposomal amphi B (FDA approved)
Miltefosine (oral) FDA approved for *L. donovani*
(Combination treatment for *L. donovani* in people living with HIV in SE Asia)

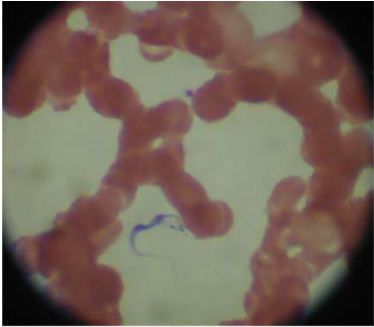
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Question #4

A 41-year-old woman presented to a local emergency department with a one-day history of fever associated with swelling and redness in her groin four days after returning from safari in Tanzania. Peripheral blood smear is obtained.

What is the most likely diagnosis?

- A. *Leishmania donovani*
- B. *Plasmodium vivax*
- C. *Trypanosoma brucei*
- D. *Wuchereria bancrofti*
- E. *Leptospira interrogans*



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Question #4

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- B. *Plasmodium vivax*
- C. *Trypanosoma brucei*
- D. *Wuchereria bancrofti*
- E. *Leptospira interrogans*



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African Trypanosomiasis

(Sleeping Sickness)

Vector = Tsetse fly (*Glossina* sp)

Trypanosoma brucei gambiense (W. Africa)

- Humans as reservoirs
- Progression over many months

Trypanosoma brucei rhodesiense (E. Africa)

- Cattle and game park animals as reservoirs
- Progression over weeks

Disease

Within 5 days: Chancre at Tse Tse fly bite
Regional lymphadenopathy

For weeks: Fever, hepatosplenomegaly,
lymphadenopathy, faint rash, headache

Late: Mental status changes, terminal somnolent state



©CDC 1996



W.H.O.

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African Trypanosomiasis:
Lab Findings

Non-specific Lab Findings

- Anemia
- Elevated IgM
- Thrombocytopenia
- Hypergammaglobulinemia

Diagnostic Lab Findings

- Detection of parasite in lymph node, circulating blood, or CSF
 - Do FNA of lymph node while massaging node, then push out the aspirate onto a slide and immediately inspect under 400x power. Trypanosomes can be seen moving for 15-20minutes, usually at edge of the coverslip
- A card agglutination test that detects *T.b. gambiense* sp. Antibodies
 - V. sensitive (94-98%), but poor specificity
 - Can get false +s in pts with Schisto, filaria, toxo, malaria

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African Trypanosomiasis:
Life Cycle

Question

Why are *Trypanosoma brucei* infections associated with persistently elevated IgM levels?

- Because they keep changing their outer surface protein
 - *T. brucei* contains as many as 1000 genes encoding different VSGs (VSG = variant surface glycoprotein)
 - Each trypanosome expresses one, and only one, VSG at a time
 - Individual parasites can spontaneously switch the VSG they express

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African Trypanosomiasis:
The Lady Gaga of the Microbial World



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African Trypanosomiasis: Treatment

July 16, 2021: Oral fexinidazole FDA approved for *T. gambiense*

West African (*T. gambiense*)

If < 6 yo or < 20 kg: lumbar puncture

CSF < 5 WBC/ul → iv pentamidine

CSF > 5 WBC/ul → iv eflornithine + nifurtimox

If > 6 yo and > 20kg: confusion, ataxia, anxiety, abnl speech, motor weakness, abnl gait?

No suspicion of late disease → oral fexinidazole

If suspicion of CNS disease → obtain lumbar puncture

CSF < 100 cells/ul (non-severe 2nd stage) → oral fexinidazole

CSF > 100 cells/ul → iv eflornithine+ nifurtimox

East African (*T. rhodesiense*)

If < 6 yo or < 20 kg: lumbar puncture

CSF < 5 WBC/ul → suramin

CSF > 5 WBC/ul → melarsoprol

If > 6 yo and > 20kg → oral fexinidazole (IV melarsoprol or suramin depending on CSF counts if unable to take po)

Notes: 1. Melarsoprol associated with ~5% death rate due to reactive encephalopathy
2. This is reduced by co-administration of corticosteroids

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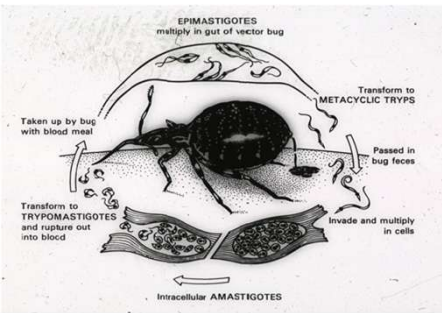
Chagas Disease

- Caused by *Trypanosoma cruzi*
(Also blood transfusion and congenitally)
- Vector: Reduviid (triatomine) bugs
- Reservoirs: Opossums, rats, armadillos, raccoons, dogs, cats
- Autochthonous cases in the U.S.:

Texas
Louisiana
Mississippi
Missouri
California



- Oral ingestion of food and drinks contaminated with reduviid bugs or the feces of those bugs is a major route of infection (acai and sugar cane juice)



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Chagas: Clinical Disease

Acute (starts 1 week after infection, can persist for 8 weeks)

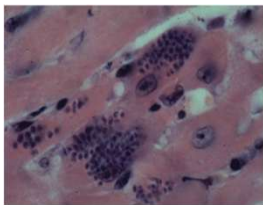
- Fever
- Local lymphadenopathy
- Unilateral, painless periorbital edema



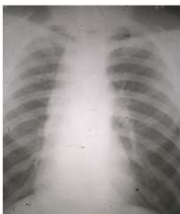
Indeterminate stage

- Serology positive, no evidence of disease

Chronic



Dilated cardiomyopathy, R>L
(CHF, syncope, arrhythmia)



Megaesophagus

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13 Protozoa That Could Be Tested

Speaker: Edward Mitre, MD

Chagas: Diagnosis & Rx

Acute disease

- Identification of parasites in blood

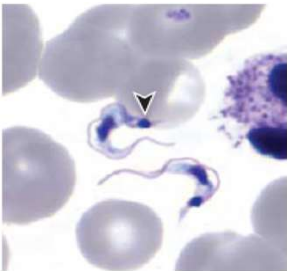
Chronic disease

- *T. cruzi* specific IgG antibodies in serum
- Two antibody tests using different antigens and different techniques recommended for dx (research: xenodiagnosis, hemoculture, PCR)

NOTE: U.S. blood supply screened for 1st time donors

Treatment

- Benznidazole for 30 – 60 d, alternative: Nifurtimox (both FDA approved)
- **Benznidazole AEs:** peripheral neuropathy, granulocytopenia, rash
- **Nifurtimox AEs:** abdominal pain/vomiting, tremors, peripheral neuropathy
- **Always offer:** acute infection, congenital, < 18 yo, reactivation disease
- **Usually offer:** 19-50 years old and no advanced cardiac disease
- **Individual decision:** > 50 years old and no advanced cardiac disease



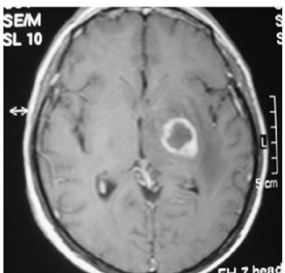
53

Chagas in Immunosuppressed Patients

T. cruzi and AIDS

Primarily reactivation neurologic disease

- Acute, diffuse, necrotic meningoencephalitis
- Focal CNS lesions (similar to Toxo)**



2008 Int J Infectious Diseases

T. cruzi and solid organ transplant

- Recipient of infected organ: fevers, hepatosplenomegaly, myocarditis
- Disease often does not occur until months after transplant

ALSO...reactivation myocarditis occurs in ~40% of patients that receive heart transplant because of Chagas cardiomyopathy

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Protozoa

Protozoa - Extraintestinal

Apicomplexa

- Plasmodium (Babesia) (Toxoplasma)

Flagellates

- Leishmania Trypanosomes (Trichomonas)

Amoebae

- Naegleria Acanthamoeba Balamuthia

Protozoa - Intestinal

Apicomplexa

- Cryptosporidium Cyclospora Cystoisospora

Flagellates

- Giardia Dientamoeba

Amoebae

- Entamoeba

Ciliates

- Balantidium

Maybe Not Protozoa Fungi Kingdom: Microsporidiosis agents
SAR supergroup: Blastocystis

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Free-living Amoebae

Naegleria fowleri

- Warm freshwater exposure
- Enters through olfactory neuroepithelium
- Fulminant meningoencephalitis
- Immunocompetent children/young adults

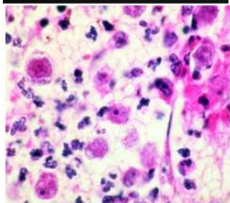
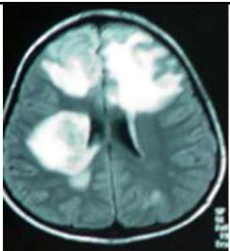
Acanthamoeba

- Found in soil and water
- Enter through lower respiratory tract or broken skin
- Subacute granulomatous encephalitis
- Immunocompromised hosts
- Chronic granulomatous keratitis (contact lens, LASIK)

Balamuthia mandrillaris

- Likely enters through lower respiratory tract or broken skin
- Transmission by solid organ transplantation has been reported
- Subacute granulomatous encephalitis
- Normal and immunocompromised hosts

Outcome → often fatal (amphotericin B, azoles, pentamidine, others tried)



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Protozoa

Protozoa - Extraintestinal

Apicomplexa

Plasmodium
(Babesia)
(Toxoplasma)

Flagellates

Leishmania
Trypanosomes
(Trichomonas)

Amoebae

Naegleria
Acanthamoeba
Balamuthia

Protozoa - Intestinal

Apicomplexa

Cryptosporidium
Cyclospora
Cystoisospora

Flagellates

Giardia
Dientamoeba

Amoebae

Entamoeba

Ciliates

Balantidium

Maybe Not Protozoa Fungi Kingdom: Microsporidiosis agents
SAR supergroup: Blastocystis

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When to Suspect an Intestinal Protozoan Infection

Patient has: Protracted watery diarrhea (weeks to months)

AND/OR:

- History of travel [domestic (esp. camping) or foreign]
- Recreational water activities
- Altered immunity (HIV infection)
- Exposure to group care (daycare)

Note: Discussion will focus on intestinal protozoa as they occur in patients seen in the U.S. These are leading causes of diarrhea, morbidity, and mortality worldwide, especially in young children.

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Intestinal Apicomplexa Parasites

Cryptosporidium

- *C. parvum*: Cows
- *C. hominis*: Humans

Cyclospora cayentanensis

Cystoisospora belli

- All have worldwide distribution
- All transmitted by water or food contaminated with oocysts
- Organisms invade enterocytes
- All cause watery diarrhea that can be prolonged & severe in immunocompromised



Cryptosporidium in enterocyte. CDC DpDx

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Intestinal Apicomplexa: Clinical Clues

Cryptosporidium

- Watery diarrhea of several weeks
- Cattle workers and daycare outbreaks
- Cysts are resistant to chlorine (water supply outbreaks)
 - #1 cause of water park/swimming pool outbreaks

Cyclospora cayentanensis

Self-limited immunocompetent BUT can last up to 10 weeks!



- Abrupt onset with nausea, vomiting, and fever early
- Anorexia, weight loss, fatigue late in course
- Food associated outbreaks: raspberries, lettuce, herbs
- Esp. Nepal, Peru, Guatemala

Cystoisospora belli

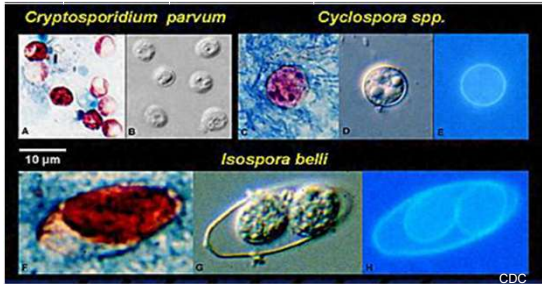
- No animal reservoirs known
- Watery diarrhea
- May be associated with a peripheral eosinophilia!
(the ONLY intestinal protozoa that does this)



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Intestinal Apicomplexa Characteristics

| Pathogen | Size | Stain | Treatment |
|-----------------|-------|-------------|------------------------------------|
| Cryptosporidium | 4 μm | m acid-fast | (None) Nitazoxanide or Paromomycin |
| Cyclospora | 10 μm | m acid-fast | TMP/SMX |
| Cystoisospora | 20 μm | m acid-fast | TMP/SMX |



- Molecular tests
- Most stool multiplex PCR assays detect cryptosporidium AND Cyclospora but NOT Cystoisospora
 - Stool Ag tests commercially available for cryptosporidium

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2:15 [social media icons] 71%

← Grandpa [phone, search, menu icons]

Friday, Jun 28 • 8:08 AM



Shall I add crptosporidium to my list of worries now that I swim frequently in our condo pool. ...chemistry is checked 3 times daily ...thx😎

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Morbidity and Mortality Weekly Report

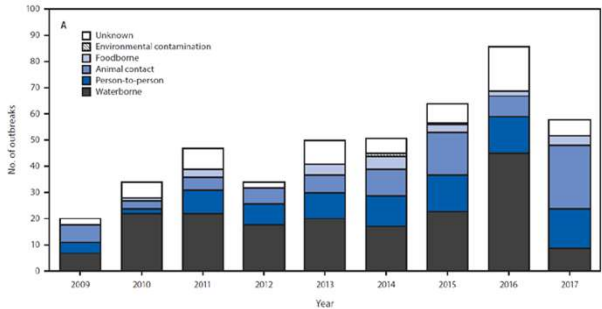
Cryptosporidiosis Outbreaks — United States, 2009–2017

MMWR / June 28, 2019 / Vol. 68 / No. 25

Morbidity and Mortality Weekly Report

Cryptosporidiosis Outbreaks — United States, 2009–2017

MMWR / June 28, 2019 / Vol. 68 / No. 25



“The number of reported outbreaks has increased an average of approximately 13% per year.”

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13 Protozoa That Could Be Tested

Speaker: Edward Mitre, MD

Question #5

A 28-year-old woman returns after studying mosquito breeding habits in Honduras for one year. She reports intermittent abdominal pain and diarrhea for several months. Stool ova and parasite exam is positive for the presence of a ciliated single cell organism.

What is the most likely diagnosis?

- A. *Balantidium coli*
- B. *Entamoeba histolytica*
- C. *Giardia lamblia*
- D. *Dientamoeba fragilis*
- E. *Endolimax nana*

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Question #5

A 28-year-old woman returns after studying mosquito breeding habits in Honduras for one year. She reports intermittent abdominal pain and diarrhea for several months. Stool ova and parasite exam is positive for the presence of a ciliated single cell organism.

What is the most likely diagnosis?

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- C. *Giardia lamblia*
- D. *Dientamoeba fragilis*
- E. *Endolimax nana*

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Balantidium coli

- The only ciliated pathogen of humans!
- Largest protozoan pathogen of humans! (about 70 µm wide and up to 200 µm long)
- Found worldwide, especially Central and S. America, S.E. Asia, and Papua New Guinea
- Associated with eating food/water contaminated with pig feces
- Symptoms: Most people asymptomatic
Can cause colitis with abdominal pain, weight loss, +/- diarrhea (Especially in malnourished and immunocompromised)
- Treatment: Tetracycline (!) or metronidazole



CDC DpDx

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Entamoeba histolytica

- Strictly human pathogen
 - Fecal/oral (contaminated food/water)
 - Cysts = infective stage
 - Trophozoites = active form, tissue-destructive
- Clinical Presentations
- Asymptomatic
 - Traveler's diarrhea
 - Colitis
 - Sharp abdominal pain
 - Bloody diarrhea
 - Fever
 - Flask-shaped ulcerations
 - Onset can occur weeks to months after travel
 - Ameboma
 - Liver and brain abscesses, esp. in young men, usually 2-5 months after travel



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13 Protozoa That Could Be Tested

Speaker: Edward Mitre, MD

Entamoeba histolytica

Diagnosis

Stool PCR (multiplex or single)

- Close to 100% sensitivity and specificity

Stool O/P

- Only 50% sensitive for colitis and abscess
 - Poor specificity b/c unable to differentiate E.histolytica from non-pathogenic E. dispar and the diarrhea-only causing E. moshkovskii
- (note: ingested RBCs suggestive of Eh, but not 100%)

Stool antigen testing > 85% sensitive for intestinal disease

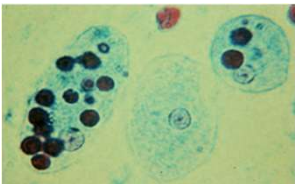
Serology 95% sensitive for liver abscess, 85% sensitive for intestinal infection

Treatment

Asymptomatic: Luminal agents such as paromomycin

Symptomatic: Tissue agents such as metronidazole or tinidazole THEN luminal agent

Liver abscess: Medical therapy (tissue agent then luminal agent) usually sufficient!
Drainage if no response to medical therapy or dx unclear or v large abscess



E. histolytica trophozoites with ingested RBCs.

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Giardia duodenalis → described by Antony van Leeuwenhoek

Flagellated protozoan

- Fecal/oral via ingestion of cyst form in food/water
- Cyst is chlorine resistant
- Cysts from humans (beavers, muskrats)

Disease in U.S.

- Most common parasitic infection in the U.S (20k cases reported/year, likely 2M)
- U.S-acquired cases peak in the late summer/early fall
- A leading cause of traveler's diarrhea

Symptoms

- Intermittent watery diarrhea weeks to months
- Foul smelling stools, flatulence, "sulfur burps"



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Giardia

At risk populations

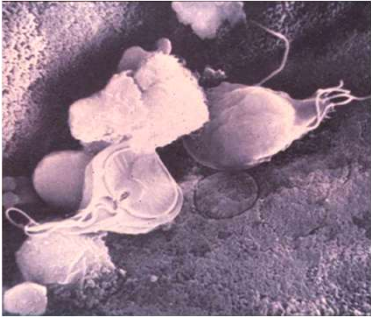
- International travelers
- Swimming in lakes/streams, outdoor survival/camping
- Infants in daycare
- Child-care workers
- Immunoglobulin deficiencies (esp CVID)
- HIV when CD4 < 100

Diagnosis

- Stool antigen test
- Stool multiplex PCR

Treatment

- Tinidazole (FDA approved)
- Metronidazole (off-label), nitazoxanide (FDA-approved), and albendazole (off label)



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Other Intestinal Protozoa

Non-pathogens

Amoebae

- Entamoeba dispar
- Entamoeba hartmanni
- Entamoeba coli
- Endolimax nana
- Iodamoeba bütschlii

Flagellates

- Chilomastix mesnili
- Trichomonas hominis

Treat if symptomatic: Dientamoeba fragilis (implicated in IBS)

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13 Protozoa That Could Be Tested

Speaker: Edward Mitre, MD

Protozoa

Protozoa - Extraintestinal

Apicomplexa

- Plasmodium (Babesia) (Toxoplasma)

Flagellates

- Leishmania
- Trypanosomes (Trichomonas)

Amoebae

- Naegleria
- Acanthamoeba
- Balamuthia

Protozoa - Intestinal

Apicomplexa

- Cryptosporidium
- Cyclospora
- Cystoisospora

Flagellates

- Giardia
- Dientamoeba

Amoebae

- Entamoeba

Ciliates

- Balantidium

Maybe Not Protozoa Fungi Kingdom: Microsporidiosis agents
SAR supergroup: Blastocystis

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Microsporidia – Obligate Intracellular Fungi!

- Produce extracellular, 1-2 micron, infective spores
- Spores have a coiled organelle called a polar tubule
- After ingestion, the spore germinates, and the polar tubule is used to inject sporoplasm into a host cell

Enterocytozoon bienersi

- Watery diarrhea
- Biliary disease (cholangitis, acalculous cholecystitis)

Encephalitozoon intestinalis

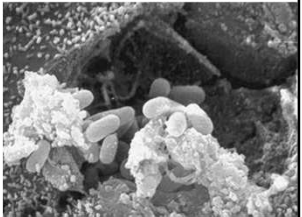
- Watery diarrhea
- Biliary disease
- Disseminated disease (liver, kidney, lung, sinuses)

Encephalitozoon cuniculi, hellem

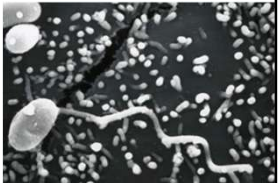
- Can cause disseminated disease of multiple organs, plus eye

Many species (including Vittaforma corneae)

- Punctate keratoconjunctivitis (contact lens use, after eye surgery, bathing in hot springs)



Spores of E. hellem bursting out of a cell (CDC DpDx)



Polar tubule inserted into a eukaryotic cell (CDC DpDx)

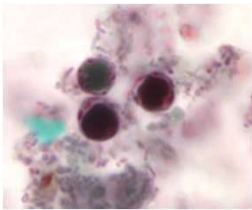
DIAGNOSIS: Modified trichrome stain, Calcofluor white, IFA to detect spores in stool
TREATMENT: Albendazole (not effective for E. bienersi)

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Blastocystis

What is it?

- Currently classified as a protozoa
- Forms are 5-40 microns wide
- Anaerobic
- Eukaryotic
- Cystic, ameboid, granular, and vacuolar forms



Blastocystis cyst-like forms, trichrome (CDC DpDx)

Often the most common eukaryotic organism found in human stool samples

Does it cause disease?

- Maybe
- Associated with watery diarrhea, abdominal discomfort, nausea, and flatulence

Diagnosis

- Light microscopy of stool samples

Treatment

- Metronidazole, tinidazole, TMP/SMX, or nitazoxanide (none FDA-approved)

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Protozoan Infections that can Reactivate in the Severely Immunocompromised

- Toxoplasmosis
 - Encephalitis with mass lesions
 - Pneumonitis
 - Retinitis
 - Leishmania
 - Reactivation of visceral and cutaneous reported
 - Visceral with fever, hepatosplenomegaly, pancytopenia
 - Chagas
 - Encephalitis with mass lesions
 - Hepatosplenomegaly and fevers
 - Myocarditis in 40% that receive heart transplant b/c Chagas disease
 - Malaria
- Some other protozoa that can case severe disease in immunocompromised
- Cryptosporidium
 - Giardia
 - Microsporidia
 - Babesia
 - Acanthamoeba

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13 Protozoa That Could Be Tested

Speaker: *Edward Mitre, MD*

Good luck on the exam!

Edward Mitre, M.D.
edwardmitre@gmail.com