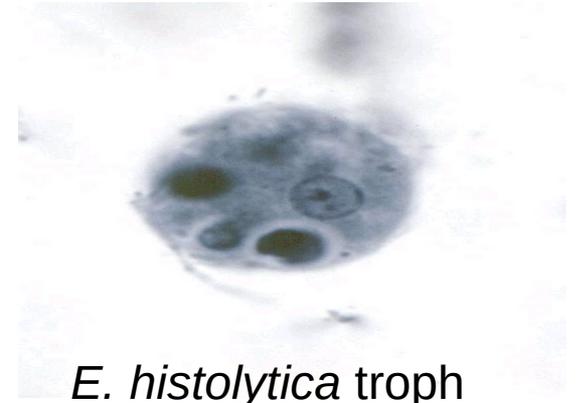


# Protozoan: Intestinal Amoeba

- *Entamoeba histolytica*
- *E. Dispar*
- *E. hartmanii*
- *E. coli*
- *Endolimax nana*
- *Iodamoeba butschlii*
- *Blastocystis hominis.*

# Amebae

- Intestinal Amebae: pathogenic
  - *Entamoeba histolytica*
- Intestinal Amebae: nonpathogenic
  - *Entamoeba dispar*
  - *Entamoeba hartmanni*
  - *Entamoeba coli*
  - *Entamoeba polecki*
  - *Endolimax nana*
  - *Iodamoeba butschlii*
  - *Blastocystis hominis*: may be pathogenic if found as sole parasite in large numbers in symptomatic patients



*E. histolytica* troph

# Amebae

- Oral Amebae: nonpathogenic
  - *Entamoeba gingivalis*: nonpathogenic amebae found in the mouth between teeth or tonsillar crypts , occasionally found in sputum; must be differentiated from *E. histolytica*
- Free-living Amebae: pathogenic
  - *Naegleria fowleri*
  - *Acanthamoeba spp*



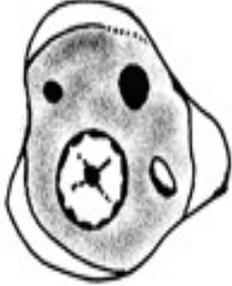
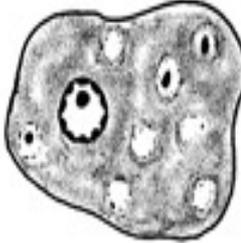
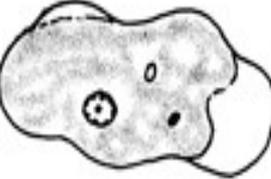
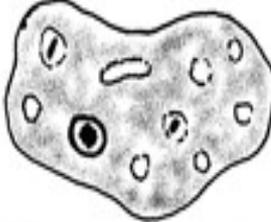
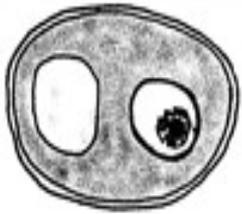
*E. histolytica* troph

# Intestinal Amebae

- General Characteristics of Amebae
  - most primitive of protozoa; smaller than other parasites
  - multiply by binary fission
  - mode of transmission: fecal-oral route
  - 2 distinct stages: trophozoite and cyst
  - trophozoite: motility by pseudopodia
  - ingestion of infective cyst stage in contaminated water and food
  - worldwide distribution with high incidence in developing countries

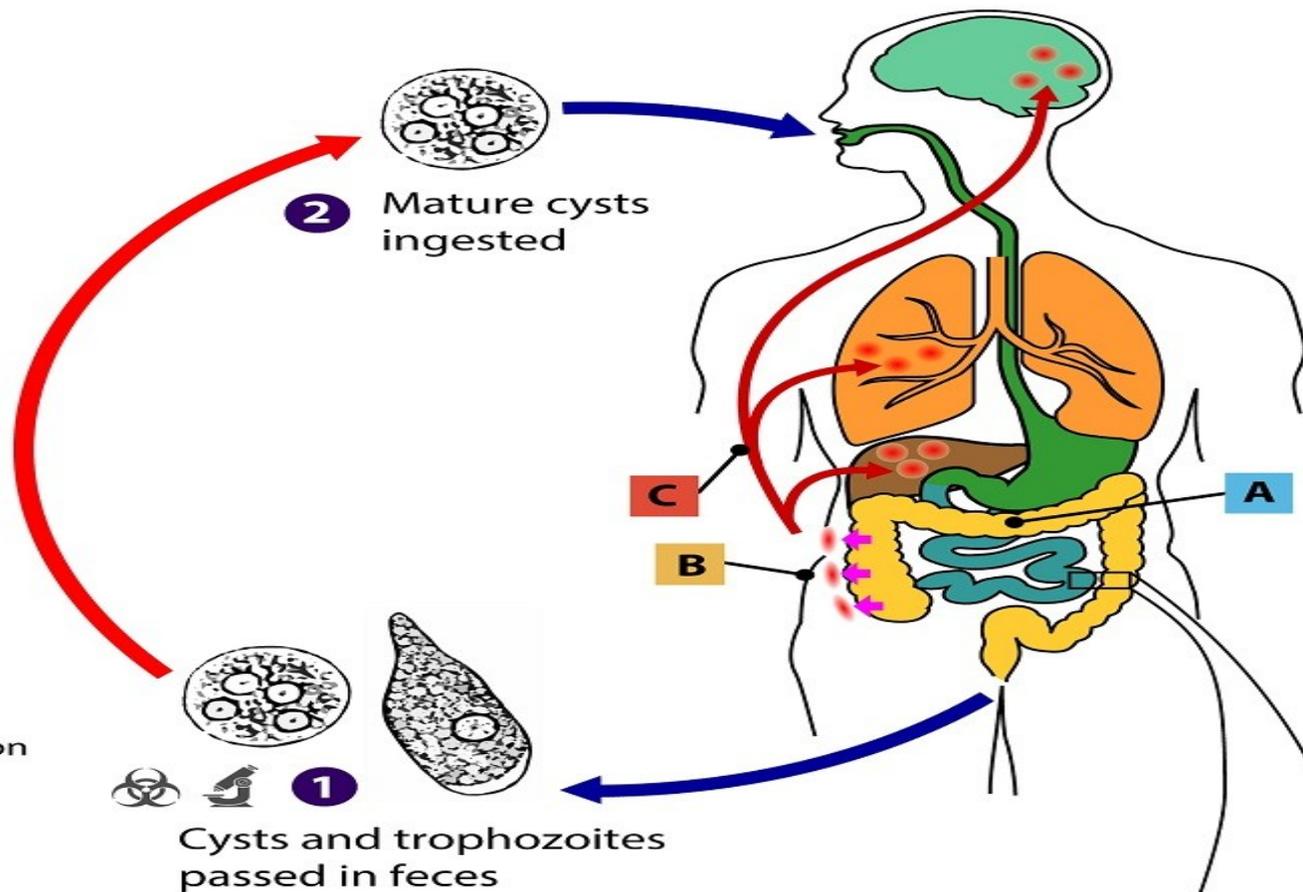
# Intestinal Ameba

- Species Identification rests on the following:
  - size of cyst / trophozoite
  - number of nuclei in cyst and trophozoite
  - nuclear structure
  - presence of specific internal structures
  - motility of trophozoite in saline wet mount

Organism	Trophozoite	Precyst	Cyst
<i>E. histolytica</i> <i>E. dispar</i> <i>E. moshkovskii</i>			
<i>E. coli</i>			
<i>E. hartmanni</i>			
<i>I. bütschlii</i>			

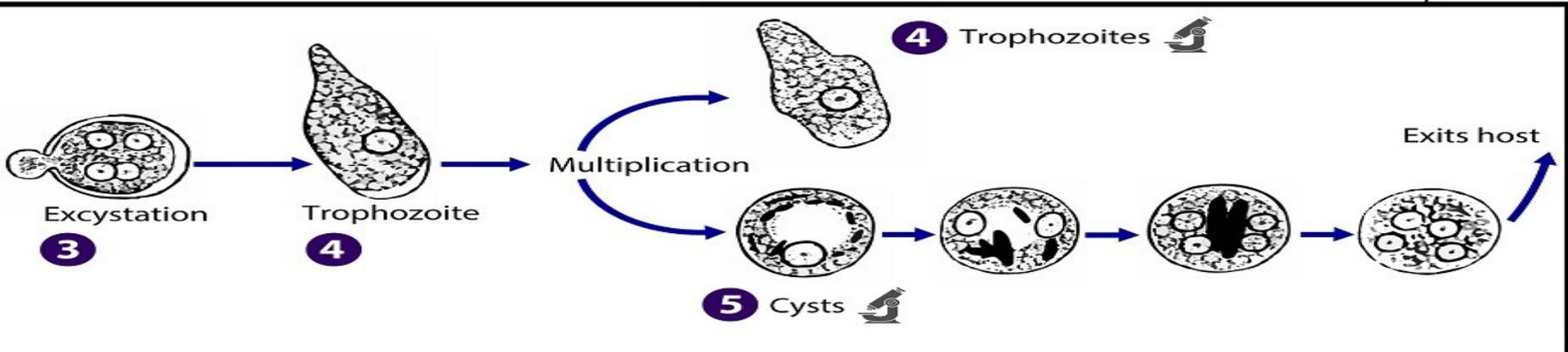
# E. histolytica lifecycle

- Cysts and trophozoites are passed in feces
- Cysts are typically found in formed stool, whereas trophozoites are typically found in diarrheal stool
- Infection with *Entamoeba histolytica* occurs via ingestion of mature cysts from fecally contaminated food, water
  - Exposure to infectious cysts and trophozoites in fecal matter during sexual contact may also occur
- Excystation occurs in the small intestine and trophozoites are released, which migrate to the large intestine
  - Trophozoites can remain in intestinal lumen (non-invasive) or spread into intestines or cause extra-intestinal diseases affecting liver, brain, and lungs
- Infectious cysts can survive weeks in the external environment due to the protection conferred by their thick walls. Trophozoites passed in the stool are rapidly destroyed once outside the body, and if ingested would not survive exposure to the gastric environment.



Infective stage  
 Diagnostic stage

**A** Noninvasive colonization  
**B** Intestinal disease  
**C** Extraintestinal disease



# Intestinal Amoeba Workup

## HISTORY

Subacute onset, blood /mucous in stool, travel /residence in endemic area

## CLINICAL SIGNS AND SYMPTOMS

Diarrhea/dysentery, abdominal pain in most, weight loss, fever ( $> 38^{\circ}\text{C}$ ) in  $< 20\%$

## DIAGNOSTIC TESTS TO SEPARATE INVASIVE FROM SECRETORY DIARRRHEA

Lactoferrin, fecal leucocytes, occult blood

## UTILIZATION OF *E. histolytica* DIAGNOSTIC TESTS

A combination of  
Stool *E.histolytica* Ag-detection test  
or  
Stool PCR  
PLUS  
serology

# Intestinal Amebae

- *E. histolytica* is the only true pathogen
- The other intestinal amoebas are non-pathogenic.
  - *Blastocystis hominis* may cause diarrhea.
- Identification of each species is important:
  - Ensures proper therapy and avoids unnecessary and inappropriate treatment because of misdiagnosis
    - *Endolimax nana*, *Iodamoeba buetschlii*, and other *Entamoeba* species) are important to ID because they may be confused with *E. histolytica*

# Stool Specimen Collection

- Collect the stool in a dry, clean, leakproof container. Make sure no urine, water, soil or other material gets in the container
- Fresh stool should be examined, processed, or preserved immediately
  - Exception for refrigerated specimens when preservatives are not available
    - These specimens are suitable for antigen testing only; follow individual Hospital SOPs
- Ensure that the specimen is mixed well with the preservative
- Specimen Frequency
  - Minimum of 3 stool specimens for O&P exam within 10 days

# Laboratory Diagnosis of Amebae: Microscopy

## 1. Direct Wet Mount

- Fresh liquid stool: examine for motile trophs within 30 min
- Small sample of stool + saline: look for extending pseudopods
- Small sample of stool + dilute iodine: contrast
- Low power (x100) entire coverslip; high power (x400)

## 2. Concentration Technique

- Stool sample in Proto-Fix or comparable solution
- Examine sediment for cysts, add dilute iodine
- Examine under low and high power like wet mount

## 3. Permanent Stained Smear

- Preserve stool sample in PVA or Proto-Fix
- Excellent preservation of cysts and trophs
- Trichrome stain: contrasting colors; confirmation of direct mounts
- Examine under oil immersion (x1000)

# Laboratory Diagnosis of Amebae

- Immunoassay: lateral flow immunoassay for *E. histolytica* Antigens
- PCR: syndromic panel for GI infections (viruses, bacteria, parasites)
  - Conventional or real-time PCR



# Antibody Detection for *E. histolytica*

- The indirect hemagglutination (IHA) test has been replaced by commercially available EIA test kits for routine testing
- If antibodies are not detectable in patients with an acute presentation of suspected amebic liver abscess, a second specimen should be drawn 7-10 days later
- If the second specimen does not show seroconversion, other agents should be considered
- Detectable *E. histolytica*-specific antibodies may persist for years after successful treatment
  - The presence of antibodies does not necessarily indicate acute or current infection
  - Patients who have lived in highly endemic areas are likely to be seropositive due to past exposures.

TROPHOZOITE Characteristics	<i>E. histolytica</i> , <i>E. Dispar</i> <i>E. moshkovskii</i>	<i>E. hartmanni</i>	<i>E. coli</i>	<i>E. polecki</i>
Trophozoites (size, nucleus, and movement)	15-20 $\mu\text{m}$ 1 nucleus actively motile cytoplasmic protrusions pseudopodium	8-10 $\mu\text{m}$ 1 nucleus Non-successive	20-25 $\mu\text{m}$ 1 nucleus slow movement short and blunt pseudopodium	15-20 $\mu\text{m}$ 1 nucleus motility like <i>E. coli</i>
Appearance of trophozoites	Stained trophozoites with fine, uniform granules of peripheral chromatin Small central karyosome in nucleus  Ingested RBC - rules in <i>E. histolytica</i>  <i>E. Dispar</i> - no ingested RBCs	Nuclear structure similar to <i>E. histolytica</i> ; ingested bacteria; cytoplasm finely granular	Nuclear with irregular cluster of peripheral chromatin; large, irregular, eccentric karyosome	Nucleus with minute central karyosome, with fine granules of peripheral chromatin, finely granular cytoplasm; ingested bacteria
Pathogenicity	Pathogen - <i>E. histolytica</i> Non-pathogens: <i>E. dispar</i> and <i>E. moshkovskii</i>	Non-pathogen	Non-pathogen	Non-pathogen

CYST Characteristics	E. histolytica, E. dispar E. moshkovskii	E. hartmanni	E. coli	E. polecki
Cysts (size, nucleus)	12-15 $\mu\text{m}$ ; mature cyst has 4 nuclei, immature cyst has 1 or 2 nuclei	6-8 $\mu\text{m}$ ; mature cyst has 4 nuclei; immature cyst has 1 or 2 nuclei; 2 nucleated cysts very common	15-25 $\mu\text{m}$ ; mature cyst has 8 nuclei, rarely 16 or more nuclei	10-15 $\mu\text{m}$ ; 1 nucleus, very rarely binucleate or quadrinucleate
Appearance of cysts	Uniform size in having both karyosome and peripheral chromatin, typical nuclear structure, chromatodial bars with squared or rounded ends	Typical nuclear structure, chromatodial bars with rounded or squared ends	Typical nuclear structure, sliver-shaped or irregular chromatoidals	Mononucleate; large central karyosome; inclusion masses, chromatoid bars with pointed or angular ends
Pathogenicity	Pathogen  (E. dispar and E. moshkovskii are nonpathogens)	Nonpathogen	Nonpathogen	Nonpathogen

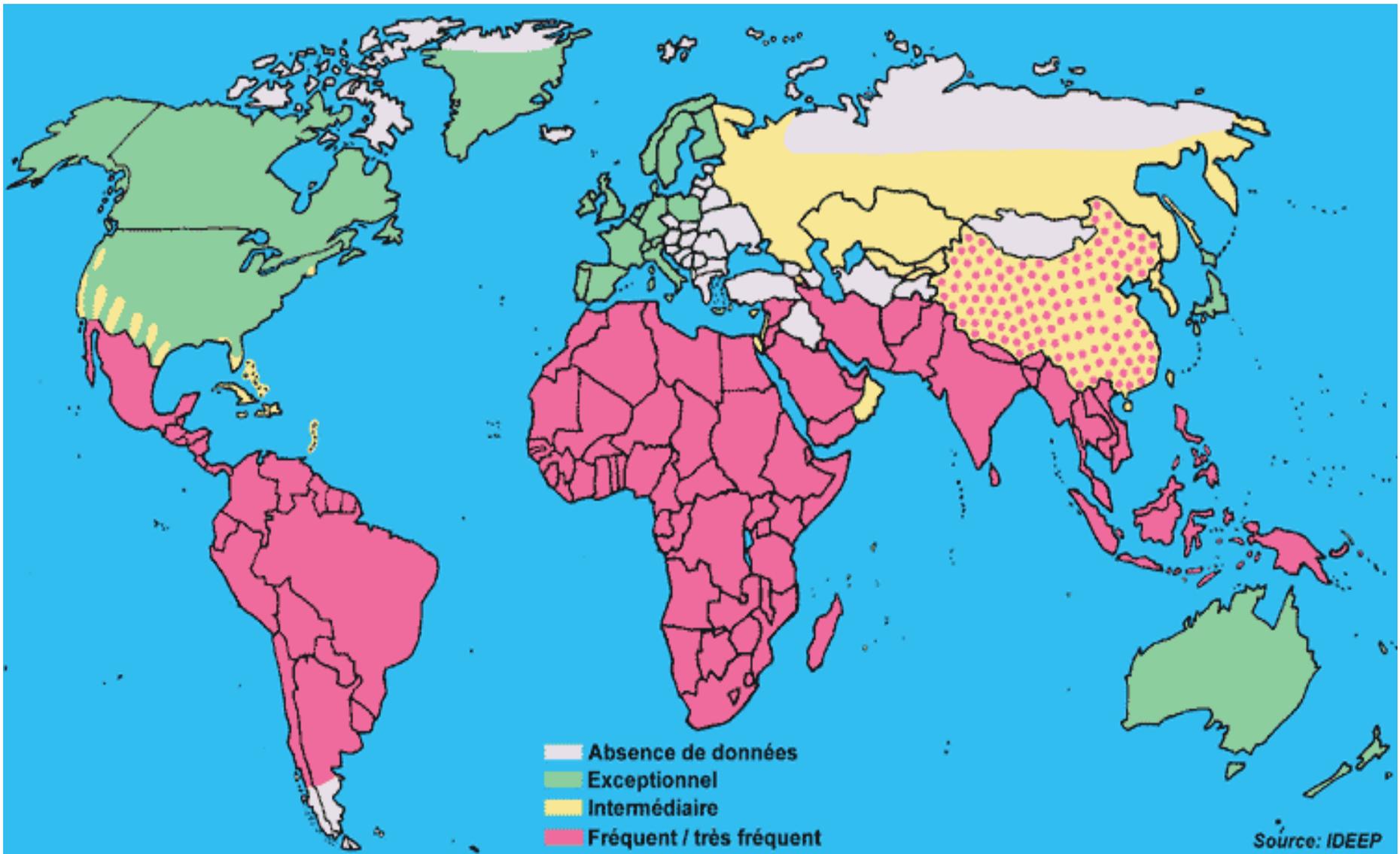
# Entamoeba histolytica: Epidemiology

- Infection rate is highest in tropical and subtropical countries
  - 2nd leading cause of mortality due to parasitic disease in humans as about 12% of world's population is infected
  - > 50% in areas of poor sanitation
  - Mexico, Caribbean, western S. America
  - West & South Africa, Middle East, India, Southeast Asia
  - Colder climates: Russia, Canada, Alaska
- Symptomatic infection (10-20% of infected population)
  - 80-98% of symptoms: intestinal mucosa (diarrhea)
  - 2-20%: extraintestinal infections (liver abscess)

# Entamoeba histolytica: Epidemiology

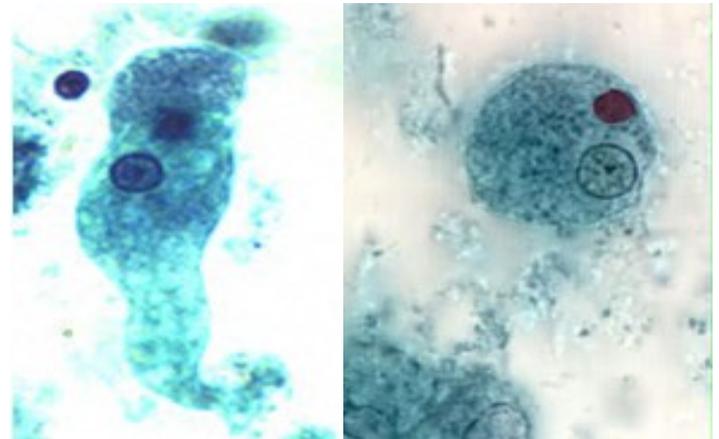
- Risk factors for transmission of amebiasis
  - crowding, poor sanitation, poor nutrition, alcoholism
  - protein malnutrition, high carbohydrate diet
  - stress & suppressed immune response – extraintestinal disease
  - reduced colonic bacterial flora
- Risk groups in industrialized countries
  - travelers, immigrants
  - institutionalized population
  - male homosexuals: sexually transmitted, gay bowel syndrome
- Prevalence of 1-5% in US
  - most cases acquired outside of US

# E. histolytica Epidemiology



# E. histolytica: Diagnosis and Risk Factors

- Risk factors for transmission of amebiasis
  - Untreated drinking water supply; poor sanitation
  - Exposure to household members, sexual partners (human reservoir)
  - Use of human feces for fertilizer
  - 5 Fs: feces, fingers, food, fomites, flies
- Disease names:
  - Amebiasis, amebic dysentery
  - Amebic hepatitis (if liver is involved)



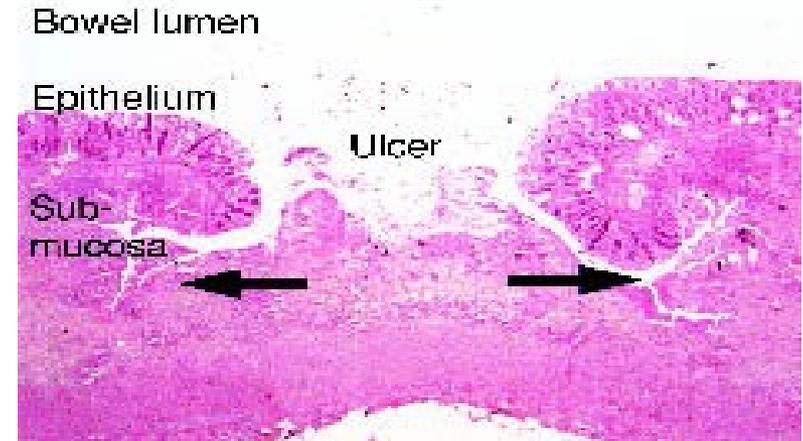
*Entamoeba histolytica* trophs

# *E. histolytica*: Pathogenesis + Life Cycle

- Ingestion of cyst (metacyst) with excystation in stomach & duodenum
- Metacyst divides to produce 4 amebulae (1 for each cyst nucleus)
  - Each amebulae divides again producing 8 trophozoites per cyst
- Trophozoites pass to cecum
  - *E. histolytica*: invade intestinal epithelium; may ingest RBCs
  - *E. dispar* looks similar but remains in lumen; feed on bacteria
- Some trophozoites encyst and cysts are passed out in stool
  - Survival of cysts in feces-contaminated water for up to 1 month

# E. histolytica: Pathogenesis

- Mucosal invasion by trophozoites
  - Adherence
    - trophozoites adhere to colonic mucins mediated by a galactose/N-acetyl-D-galactosamine adherence lectin – virulence factors
  - Invasion
    - trophozoites secrete proteolytic enzymes that disrupt colonic epithelial cells causing cell lysis
  - Ulcer formation
    - trophozoites break through muscularis layer into submucosa
    - rapid lateral spread
    - “flask-shaped” ulceration of the colonic wall
    - bloody dysentery



# E. histolytica: Intestinal Disease

- Asymptomatic Carrier State (80% of exposed persons)
  - Low virulence strain of E. histolytica
  - Low inoculation into human host; intact immune system
  - Cysts found in stool specimens; if trophozoites, no ingested RBCs
  - Cyst shedding period: 1-9 months
- Noninvasive Disease
  - Gradual onset of abdominal pain, frequent bowel movements
  - Tenesmus: urgency to defecate
  - Onset of symptoms: 4 days after exposure to 1 year

# E. histolytica: Intestinal Disease

- Invasive Disease
  - Symptoms: range from mild diarrhea to severe ulcerative colitis
  - Serious disease: extreme abdominal tenderness, fulminating dysentery (>10 stools/day), fever, dehydration, electrolyte imbalance, weight loss with general malaise
  - Dysentery: frequent watery stools with blood-tinged mucous
  - Amebic colitis: segmentary ulceration of the colon

# E. histolytica: Extra-intestinal Disease

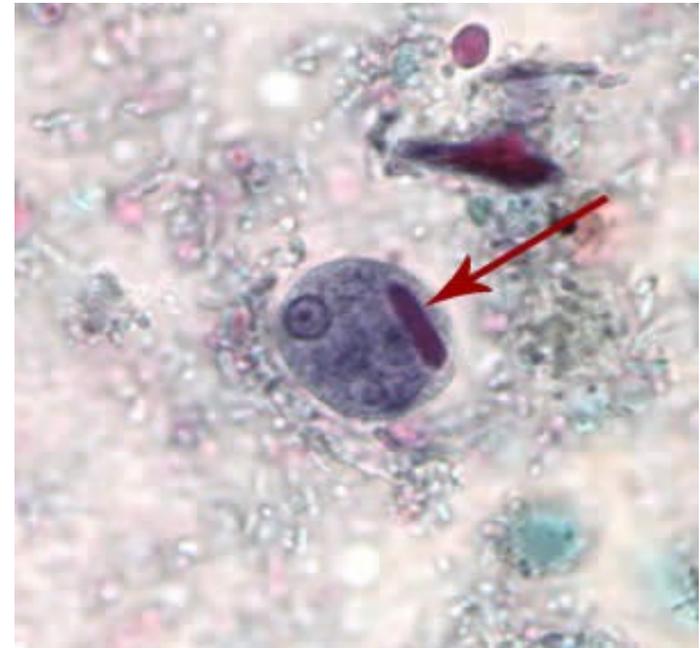
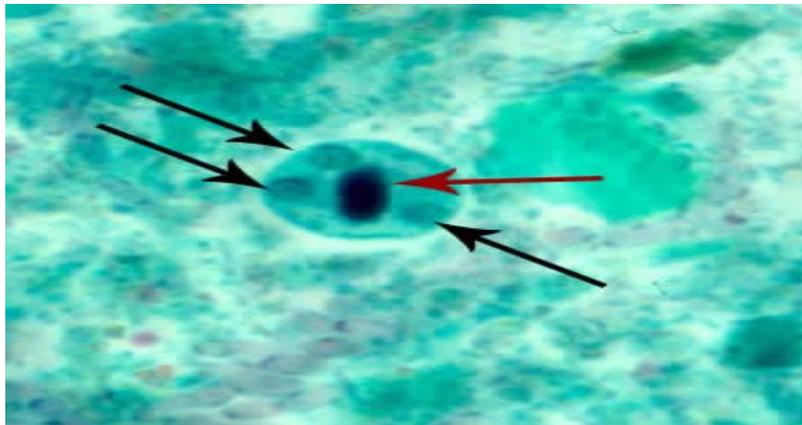
- Amebic Hepatitis (liver abscess)
  - Hematogenous spread of trophozoites from colonic submucosa to liver
  - 4% of clinical infections involve the liver
  - Upper right abdominal pain, fever, hepatomegaly, jaundice is rare
  - Liver function tests may be normal or slightly elevated
  - Amebic abscess: progressive, nonsuppurative, destructive
  - >50% of patients: no history of intestinal infection
  - Symptoms: weakness, weight loss, sweating, nausea, vomiting
  - Constipation with or without alternating diarrhea

# E. histolytica: Extra-intestinal Disease

- Amebic Pneumonitis
  - Trophozoite erosion of liver abscess
  - Trophozoite extension from liver migrate through diaphragm into lung
- Other Extraintestinal Sites
  - Hematogenous dissemination of trophozoites
  - Lung, brain, spleen, pericardium, skin, genital areas

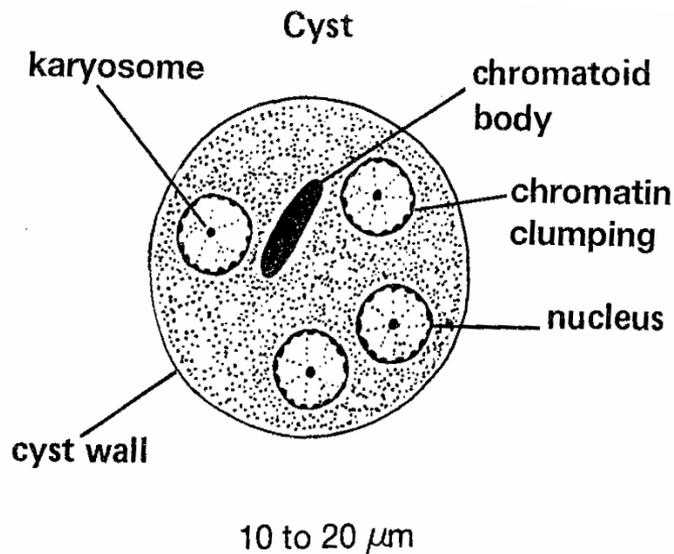
# *E. histolytica* cysts

- Nearly impossible to differentiate from *E. dispar*, find trophozoites with ingested RBCs
- Cysts have 4 nuclei with centrally located karyosomes with fine, even peripheral chromatin
- Cysts are usually 12-15  $\mu\text{m}$  in size
- Cytoplasmic inclusions include chromatoid bodies with blunted/rounded ends



# *E. histolytica*: Cyst

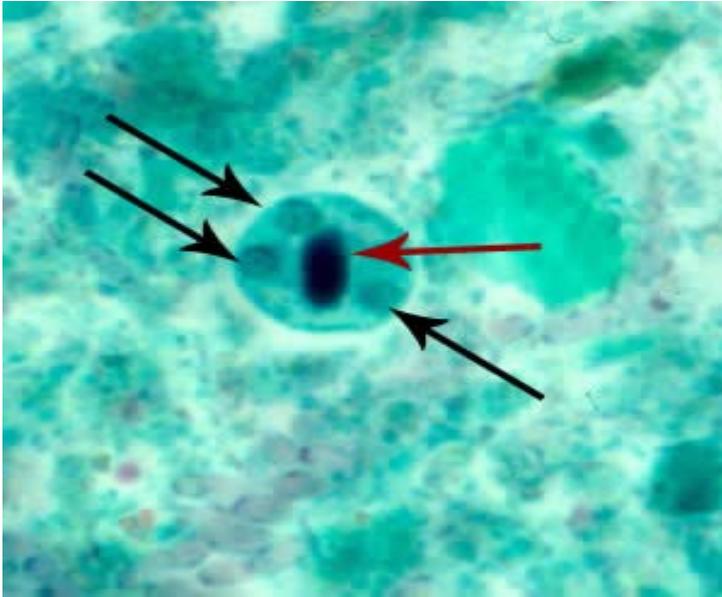
- Size : 12 - 15  $\mu\text{m}$
- Number of nuclei: 1, 2 or 4
- Nuclear appearance: small central karyosome with even peripheral chromatin
- has cigar-shaped chromatoid bars ( composed of RNA)
- trichrome stained smear: cysts are light green-gray and the nuclear material are purple-red



# E. histolytica Cyst Morphology

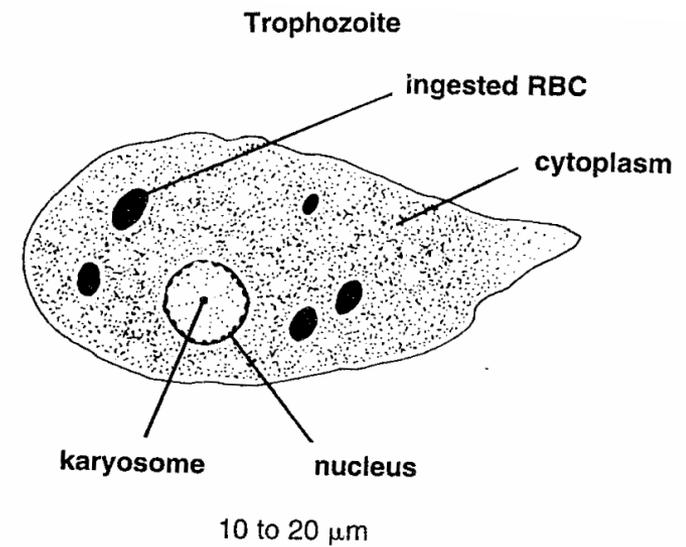
			NUCLEUS			CYTOPLASM	
Species	Size (Diameter or Length)	Shape	Number	Peripheral Chromatin	Karyosomal Chromatin	Inclusions	Appearance
<i>Entamoeba histolytica</i>	10-20 $\mu\text{m}$ Usual range, 12- 15 $\mu\text{m}$ .	Usually spherical.	4 in mature cyst. Immature cysts with 1 or 2 occasionally seen.	Peripheral chromatin present. Fine, uniform granules, evenly distributed.	Small, discrete, usually centrally located.	Present. Elongated bars with bluntly rounded ends.	Usually diffuse. Concentrated mass often present in young cysts. Stains reddish brown with iodine.

# *E. histolytica*: Cyst

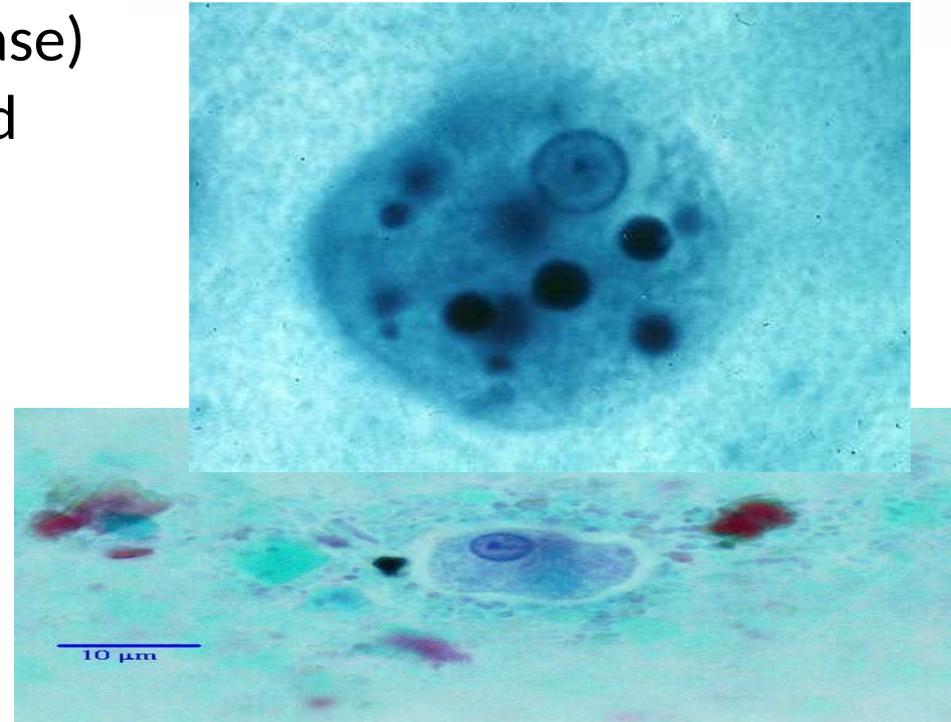
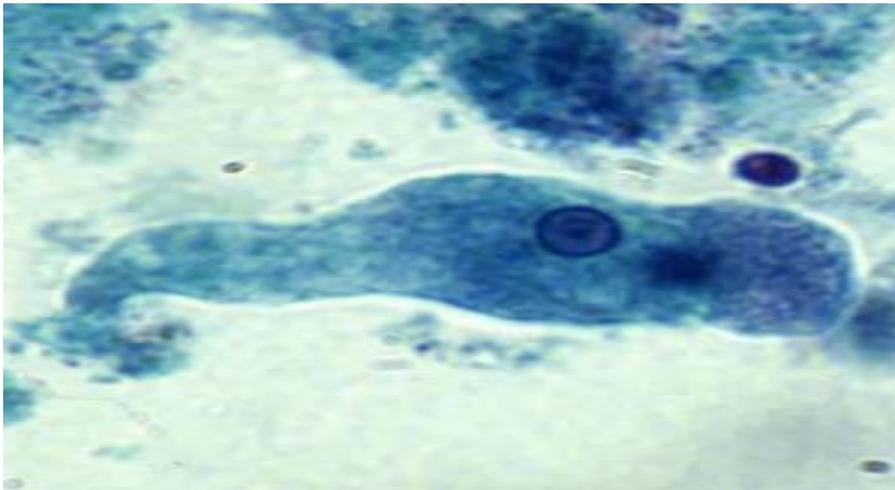


# E. histolytica: Trophozoite

- Size: 15 to 20  $\mu\text{m}$  (may be up to 60  $\mu\text{m}$ )
- Number of nuclei: 1 with centrally located karyosome with a “bull’s eye appearance,” and fine even peripheral chromatin
- Cytoplasm: ground-glass appearance and may contain ingested RBC (diagnostic of active invasive disease)
  - May have ingested bacteria and vacuoles but not diagnostic



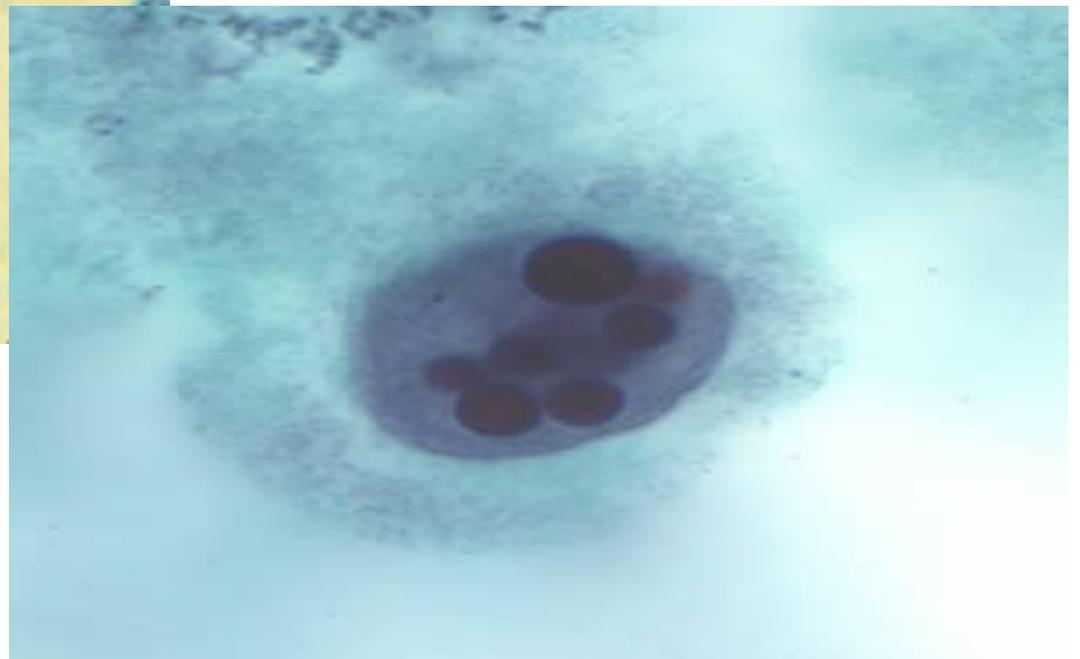
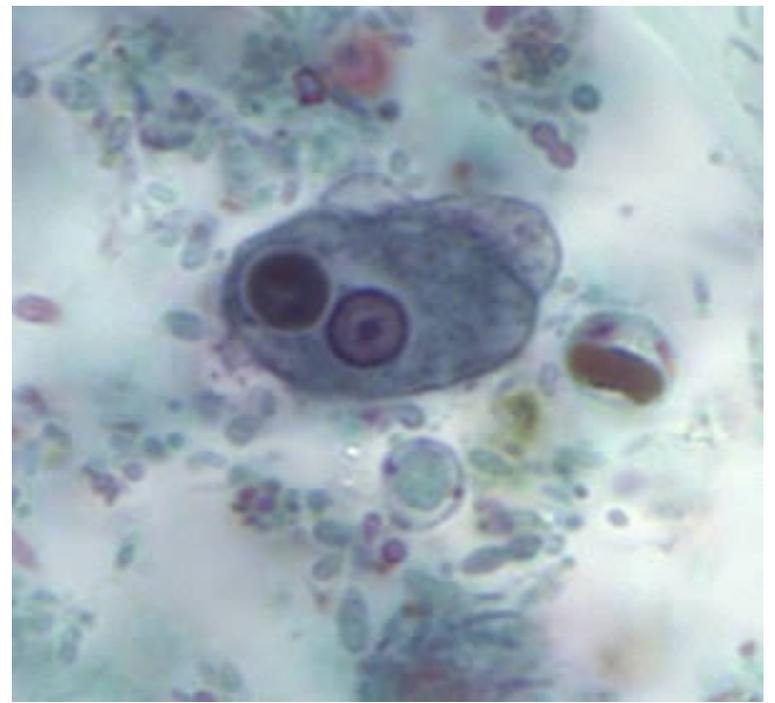
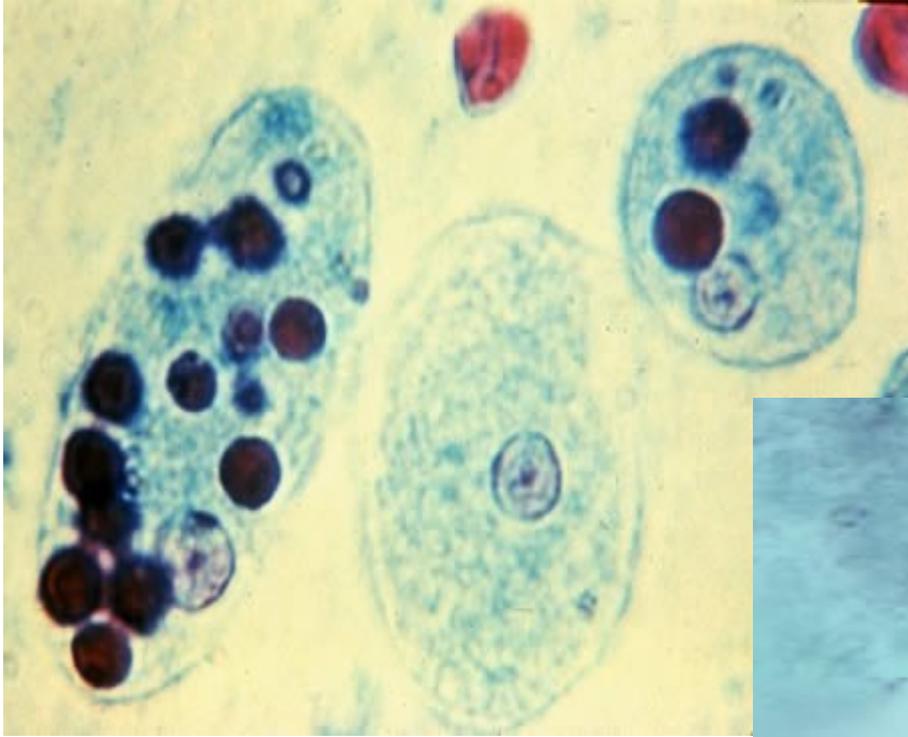
*Note:* Even pattern of nuclear chromatin on the nuclear rim with spoke-like pattern from the karyosome.



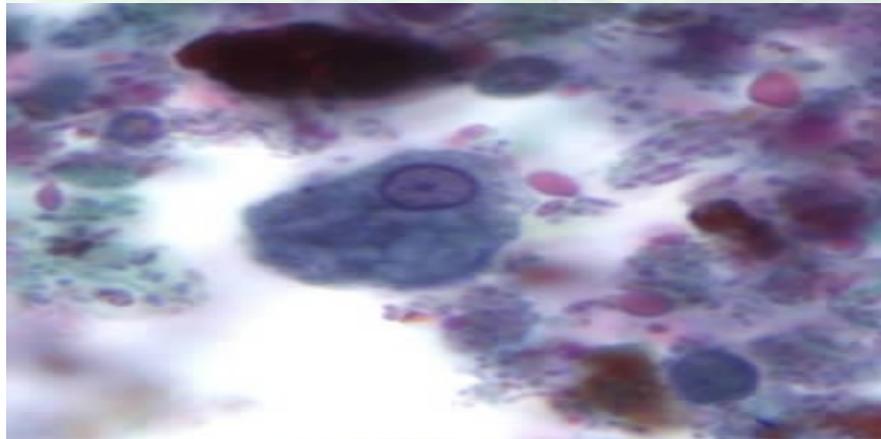
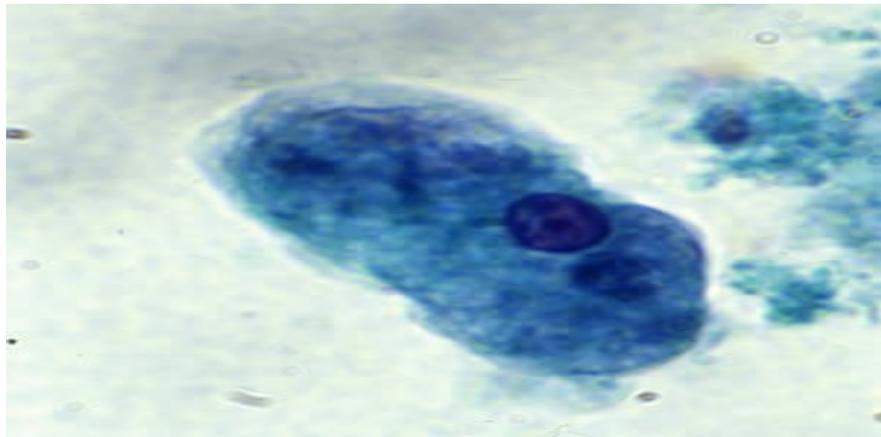
# E. histolytica Trophozoite Morphology

			NUCLEUS			CYTOPLASM	
Species	Size (Length)	Motility	Number	Peripheral Chromatin	Karyosomal Chromatin	Appearance	Inclusions
<i>Entamoeba histolytica</i>	10-60 $\mu\text{m}$ . Usual range, 15-20 $\mu\text{m}$ commensal form. <sup>1</sup> Over 20 $\mu\text{m}$ invasive form. <sup>2</sup>	Progressive with hyaline, finger-like pseudopods.	1 Not visible in unstained preparations.	Fine granules. Usually evenly distributed and uniform in size.	Small, discrete. Usually centrally located, but occasionally is eccentric.	Finely granular.	Red blood cells occasionally. Noninvasive organisms may contain bacteria.

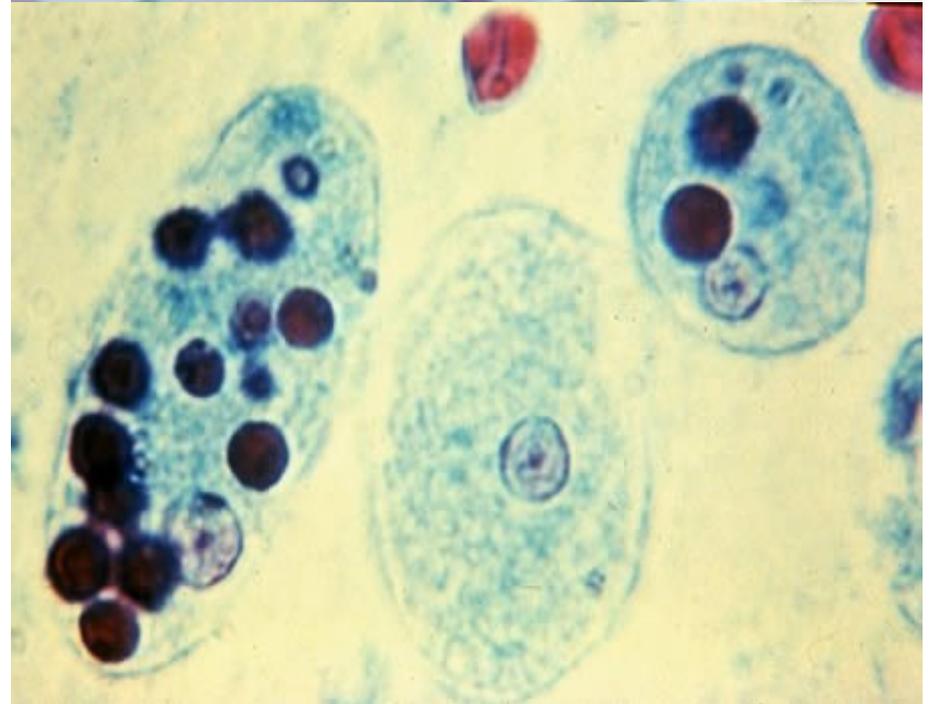
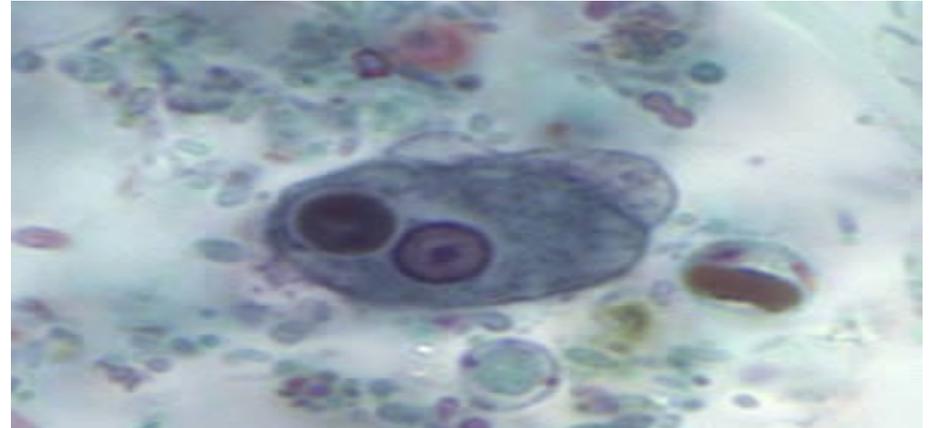
# E. Histolytica with ingested RBCs



# *E. histolytica*: Trophozoite



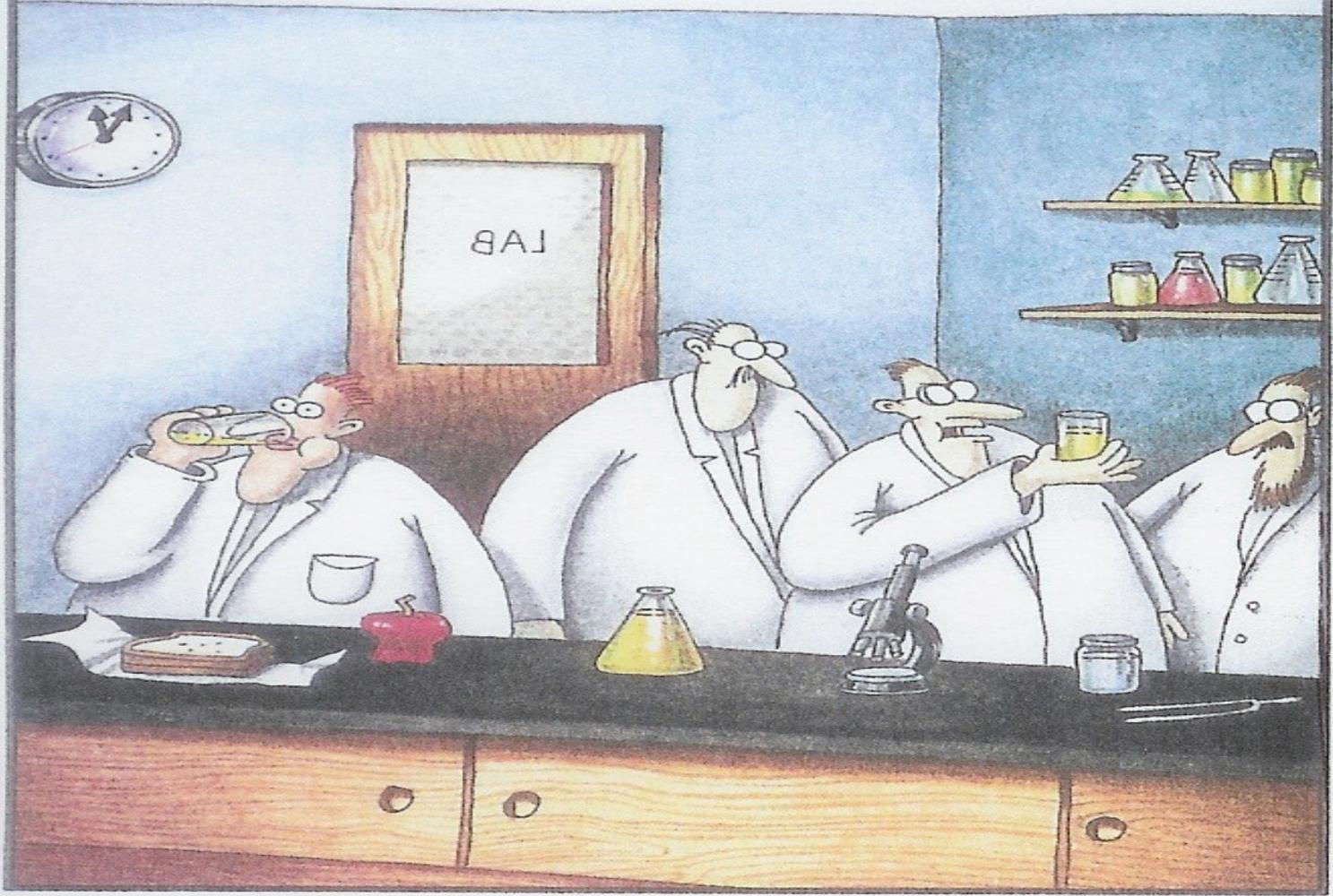
Wet Mount - Iodine prep



# E. histolytica Treatment

- Requires treatment of patient regardless of symptoms
- Treatments to avoid (worsens symptoms)
  - corticosteroids and antimotility drugs
- Asymptomatic patients with cysts only
  - Luminal amebicides for cysts (intraluminal infections)
  - 1) paromomycin (Humatin), 2) iodoquinol (Yodoxin), or 3) diloxanide furoate (Furamide, safe in children)
- Asymptomatic patients with cysts and trophozoites
  - Tissue amebicides for trophozoites
  - 1) metronidazole, 2) tinidazole, or 3) dehydroemetine followed by iodoquinol (for cysts)
- Symptomatic invasive disease: mild to severe
  - Metronidazole followed by iodoquinol or paromomycin
  - For treatment failures use dehydroemetine (higher toxicity) followed by iodoquinol or paromomycin

Louison



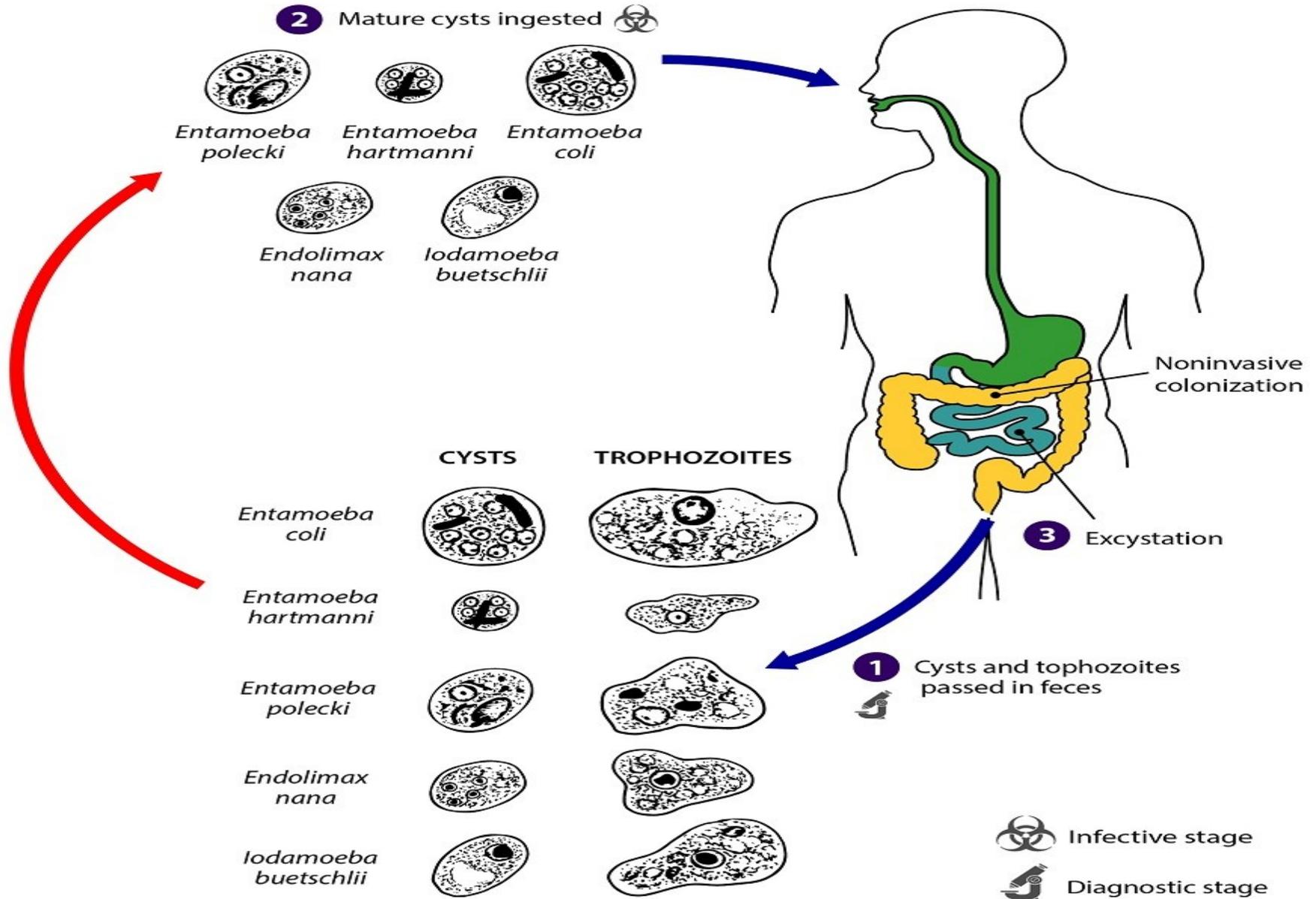
“What the? ... This is lemonade! Where’s my culture of amoebic dysentery?”

# Non-pathogenic Intestinal Amebae

- These amebae are found worldwide
  - Prevalence is highest in areas with inadequate sanitation
- None of these amebae cause symptomatic disease in humans; colonization is non-invasive
  - *Entamoeba dispar*
  - *Entamoeba hartmanii*
  - *Entamoeba coli*
  - *Endolimax nana*
  - *Iodamoeba butschlii*
  - *Blastocystis hominis*

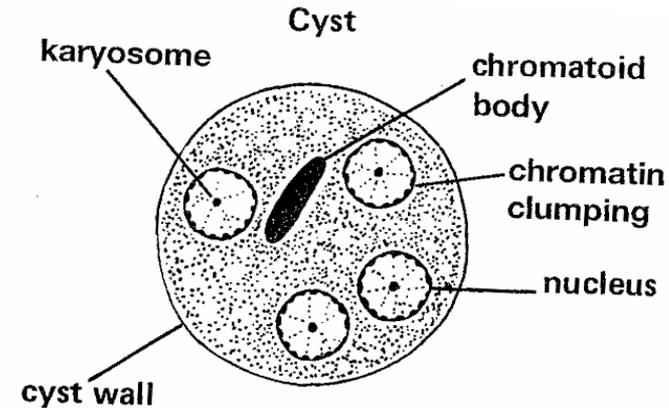
# Non-Pathogenic Entamoeba life cycles

- *Entamoeba coli*, *E. hartmanni*, *E. polecki*, *Endolimax nana*, and *Iodamoeba buetschlii*
- Both cysts and trophozoites are passed in stool and are diagnostic
  - Cysts are typically found in formed stool, whereas trophozoites are typically found in diarrheal stool
- Intestinal colonization with nonpathogenic amebae occurs after ingestion of mature cysts in fecally contaminated food, water, or fomites
- Excystation occurs in the small intestine releasing trophozoites that migrate to the large intestine. The trophozoites multiply by binary fission and produce cysts
- Cysts can survive weeks in the external environment and are responsible for transmission
- Trophozoites passed in the stool are rapidly destroyed once outside the body and, if ingested, would not survive exposure to the gastric environment.

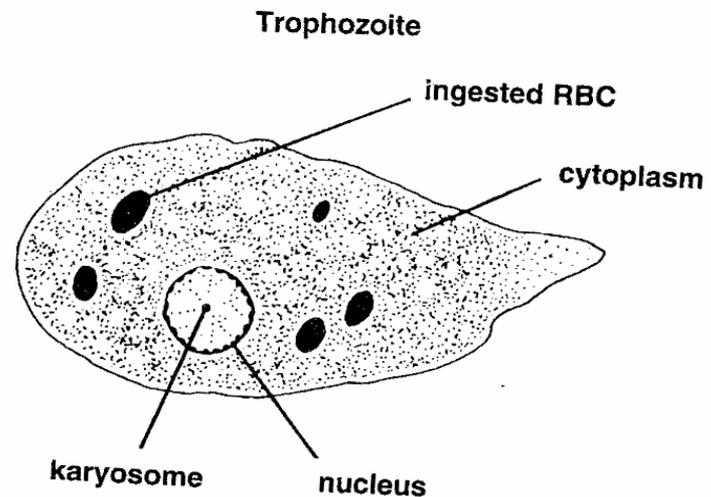


# Entamoeba dispar: non-pathogenic

- Human host, noninvasive, nonpathogenic (commensal)
- never ingest RBCs
- similar morphologically to *E. histolytica*
- formerly referred to as nonpathogenic *E. histolytica*
- Species identified by different antigens or DNA sequences



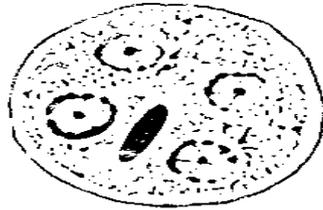
10 to 20  $\mu\text{m}$



10 to 20  $\mu\text{m}$

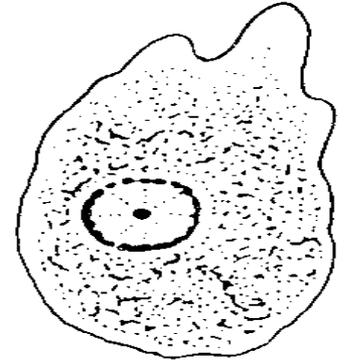
# Entamoeba hartmanni: nonpathogenic

*Entamoeba hartmanni* (small race of  
*E. histolytica*; nonpathogenic)



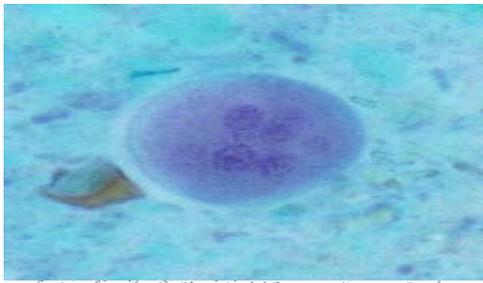
diameter less than 10  $\mu\text{m}$

A

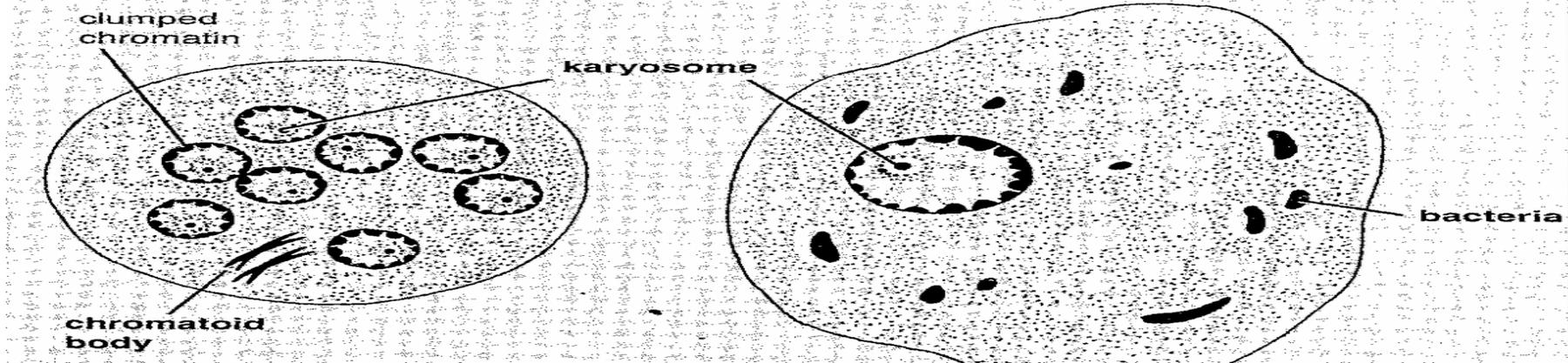
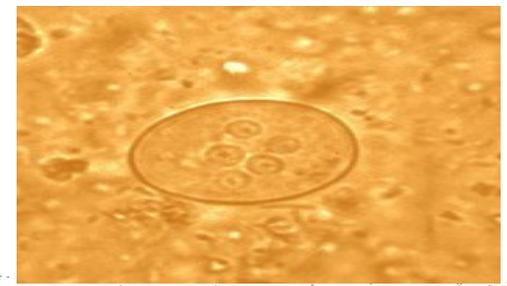


B

- Cyst
  - formerly known as small race of *E. histolytica*
  - Size: 5-10  $\mu\text{m}$ ;
  - Nuclei : 4 with small discrete centrally located karyosome and evenly distributed peripheral chromatin
  - Cytoplasmic inclusion: cigar-shaped chromatoidal bars/bodies
- Trophozoite
  - Size : 5-15  $\mu\text{m}$ ; number of nuclei : 1 with small, compact, central karyosome w/ even peripheral chromatin
  - Cytoplasmic inclusions : bacteria, no RBC



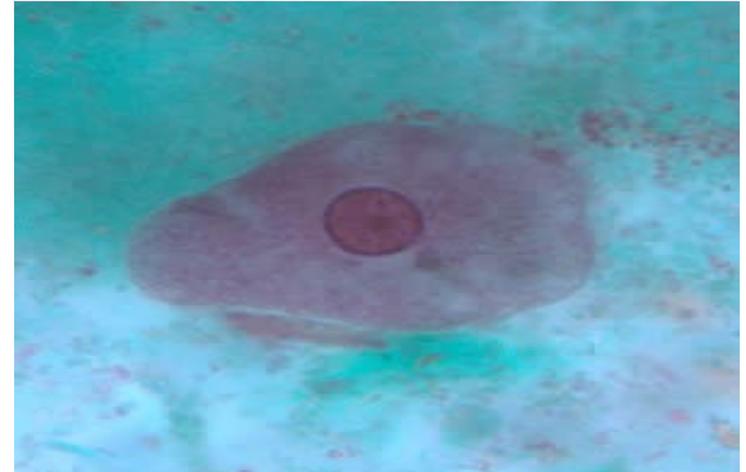
# Entamoeba coli: nonpathogenic



- Cosmopolitan in distribution, nonpathogenic
- 20 to 30 % rate of infection in US population
- Cysts – usually spherical
  - size : 10-35  $\mu\text{m}$ ; number of nuclei: 4-8; sometimes up to 16
  - Nuclear appearance: eccentric karyosome w/ irregular, coarse peripheral chromatin
  - Cytoplasmic inclusions: diffuse glycogen and chromatoid bodies are rarely seen, but are splinter-shaped with pointed rough edges

# E. coli

- Trophozoite
  - size: 15 to 50  $\mu\text{m}$  (average 25)
  - Number of nuclei : 1
  - Nuclear appearance: eccentric karyosome w/ irregular, clumped chromatin
  - Cytoplasmic inclusions: ingested bacteria , vacuoles, coarse and granular
  - Motility: sluggish, nondirectional



# Differential of Entamoeba spp

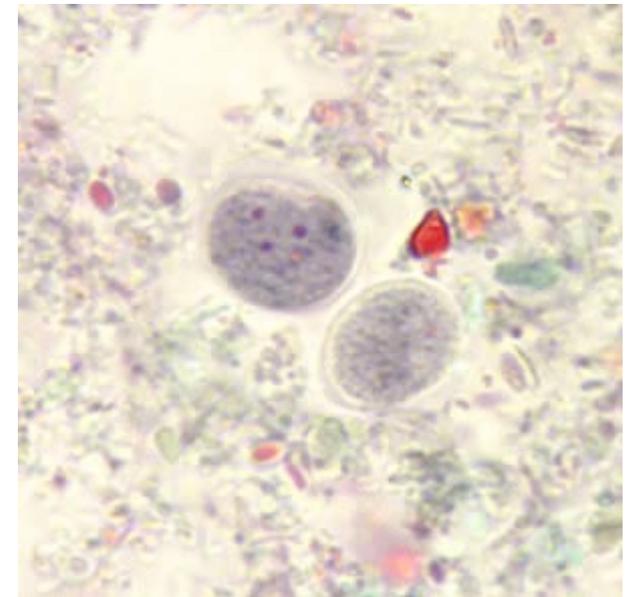
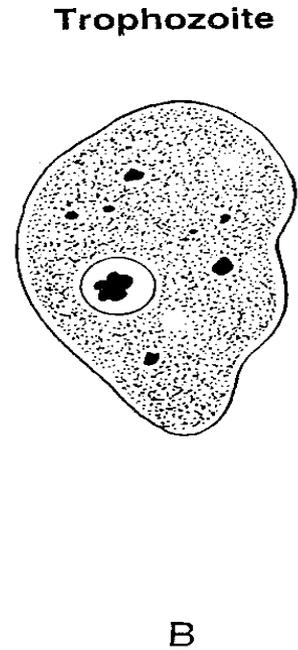
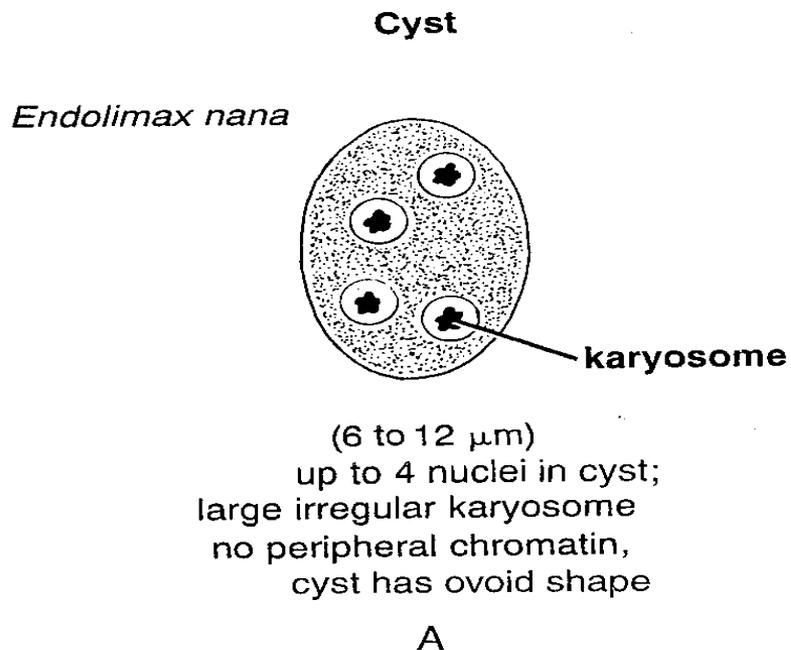
Characteristics	<i>E histolytica</i>	<i>E hartmanii</i>	<i>E coli</i>
<b>Cysts</b>	Larger	Smaller	Larger
size	>10 $\mu\text{m}$ (12-15)	<10 $\mu\text{m}$ (6-8)	>10 $\mu\text{m}$ (15-25)
chromatoidal bars	rods	rods	splinters
nuclei	1-4	1-4	1-8
peripheral chromatin	<b>fine, even</b>	fine, even	<b>coarse, irregular</b>
karyosome	small, central	small, central	<b>large, eccentric</b>
<b>Trophozoites</b>			
size	>10 $\mu\text{m}$ (12-15)	<10 $\mu\text{m}$ (6-8)	>10 $\mu\text{m}$ (15-25)
Ingested bacteria	few	few	<b>many</b>
Ingested RBCs	yes	no	no

1 nucleus each; peripheral chromatin & karyosomes like cysts

# Endolimax nana.

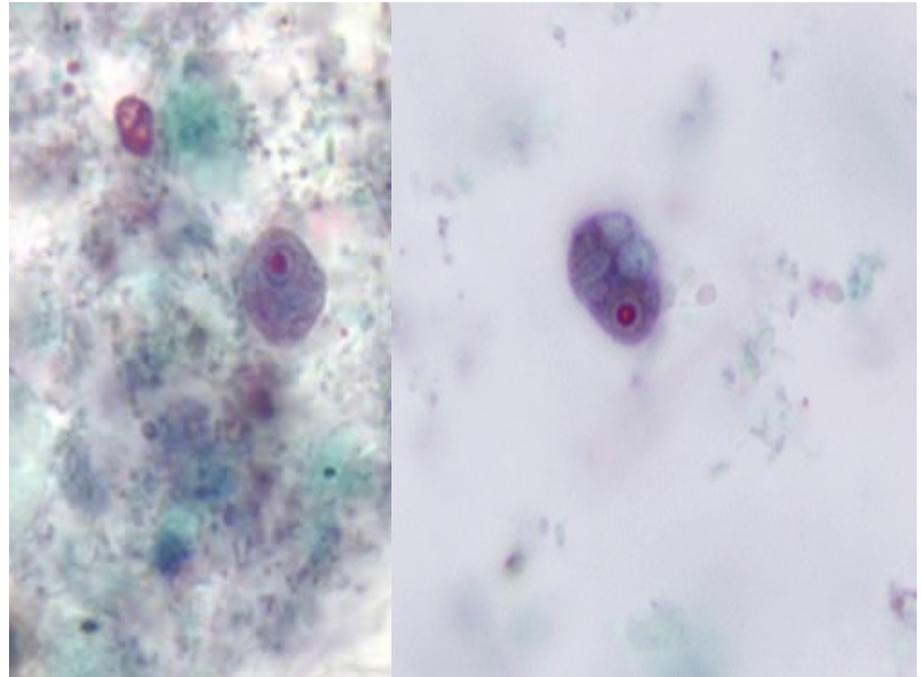
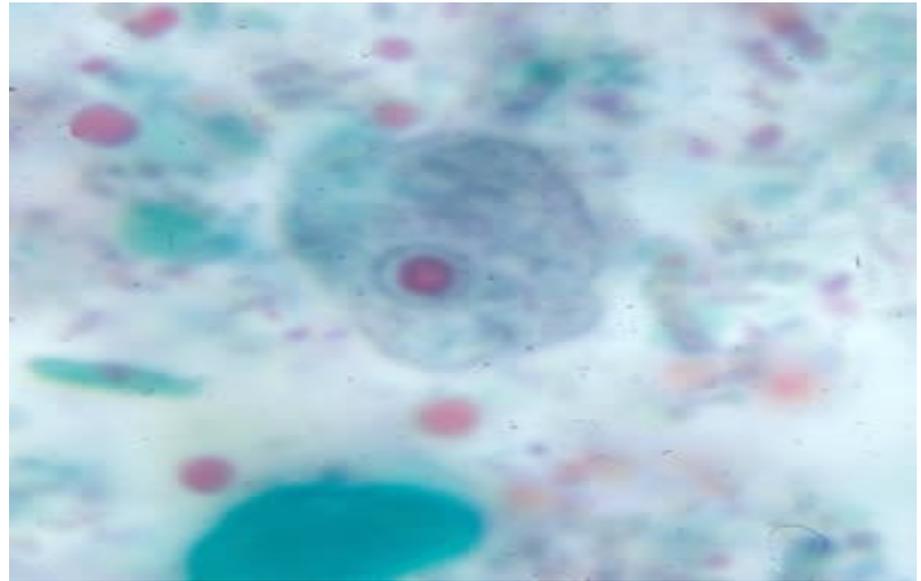
## nonpathogenic

- Cyst: ovoid or spherical → size: 5 to 12  $\mu\text{m}$  ( usually 8-10)
  - Number of nuclei: 4 with large centrally located karyosome and no peripheral chromatin “4 raisins in rice pudding”
  - Nuclear appearance: not visible with unstained wet mounts, but are visible with iodine and trichrome
  - Cytoplasmic coarsely granular with inclusions but no chromatoid bodies exist

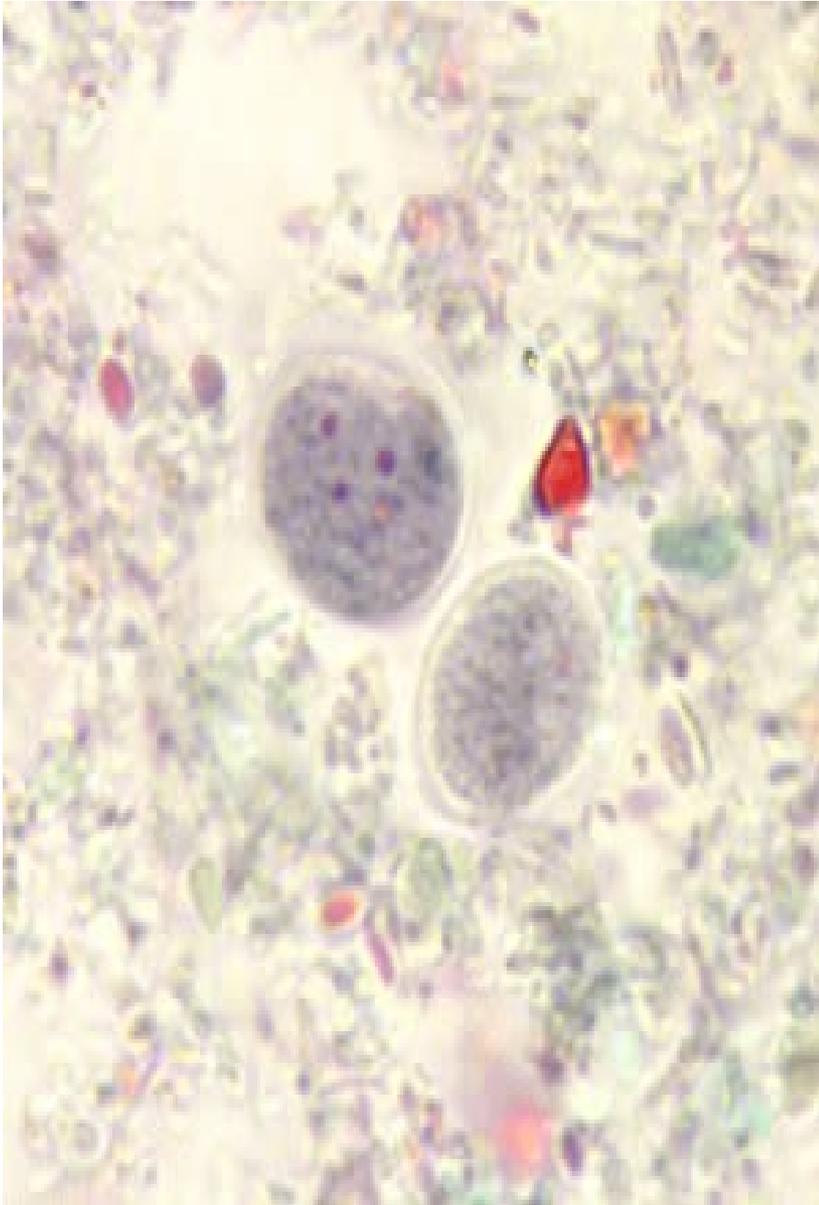


# E. nana

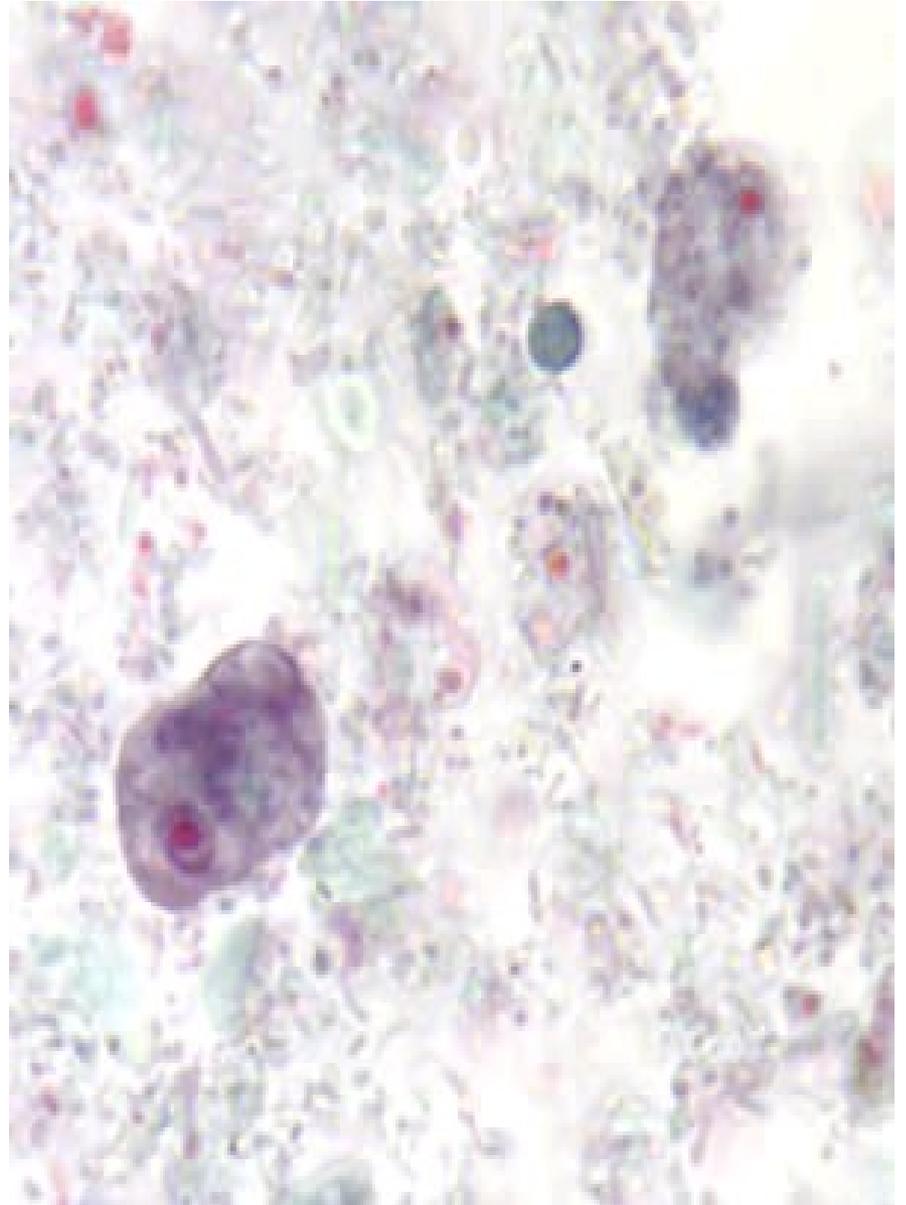
- Trophozoite:
  - size: 5 to 12  $\mu\text{m}$  (average <10)
  - Number of nuclei : 1 with irregularly shape, “blot-like” karyosome
  - Motility : sluggish
  - Cytoplasmic inclusions : vacuoles w/ ingested debris and bacteria
  - May be hard to differentiate from *Iodamoeba buetschlii*



Cyst of *E. nana*

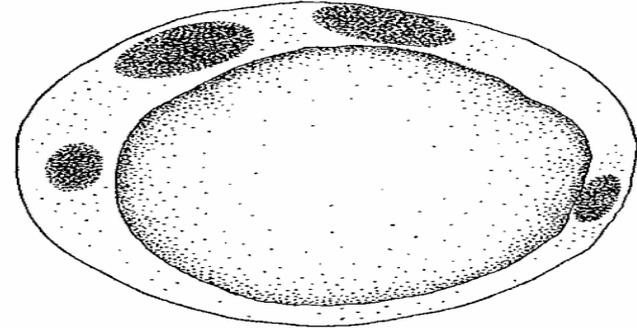


Trophozoite of *E. nana*

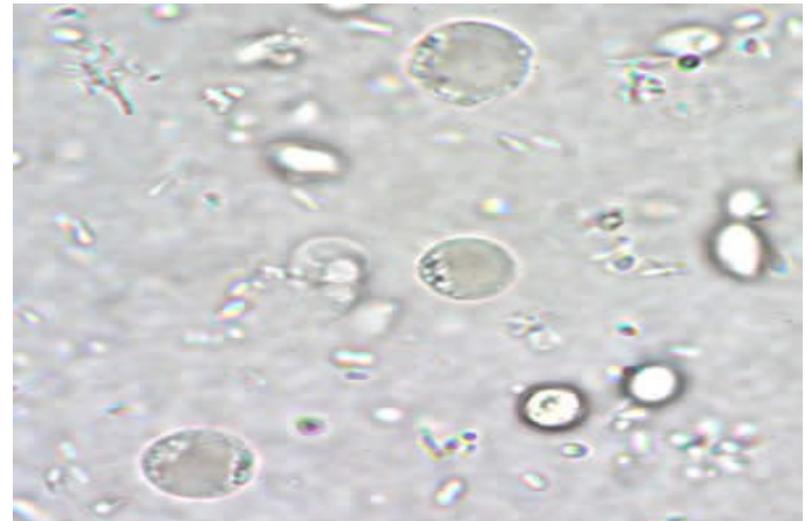
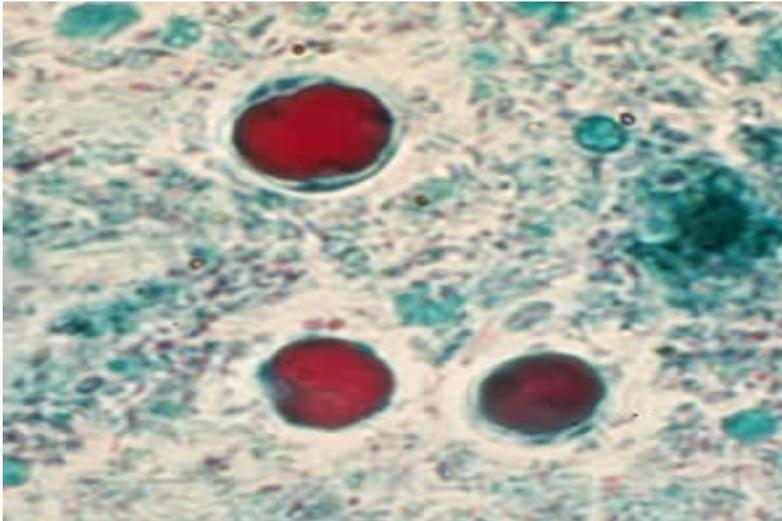


# Blastocystis hominis

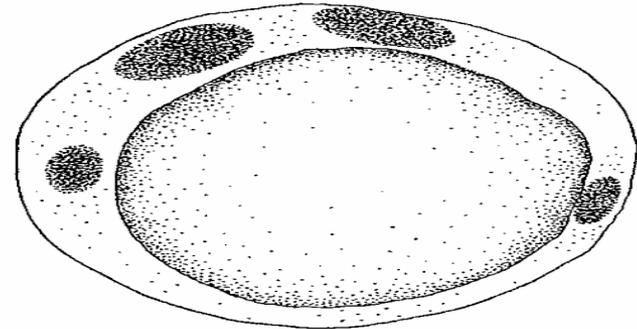
- worldwide distribution
- strictly anaerobic
- multiply by binary fission or sporulation
- considered pathogen in symptomatic patients (diarrhea, cramps, fever, nausea ) and present in counts of  $> 5/ \text{ hpf}$  in absence of other parasites
- Pleomorphic, but classic stage is circular, spherical cyst (vacuolar form)



*Blastocystis hominis*  
5 to 30  $\mu\text{m}$



# Blastocystis hominis

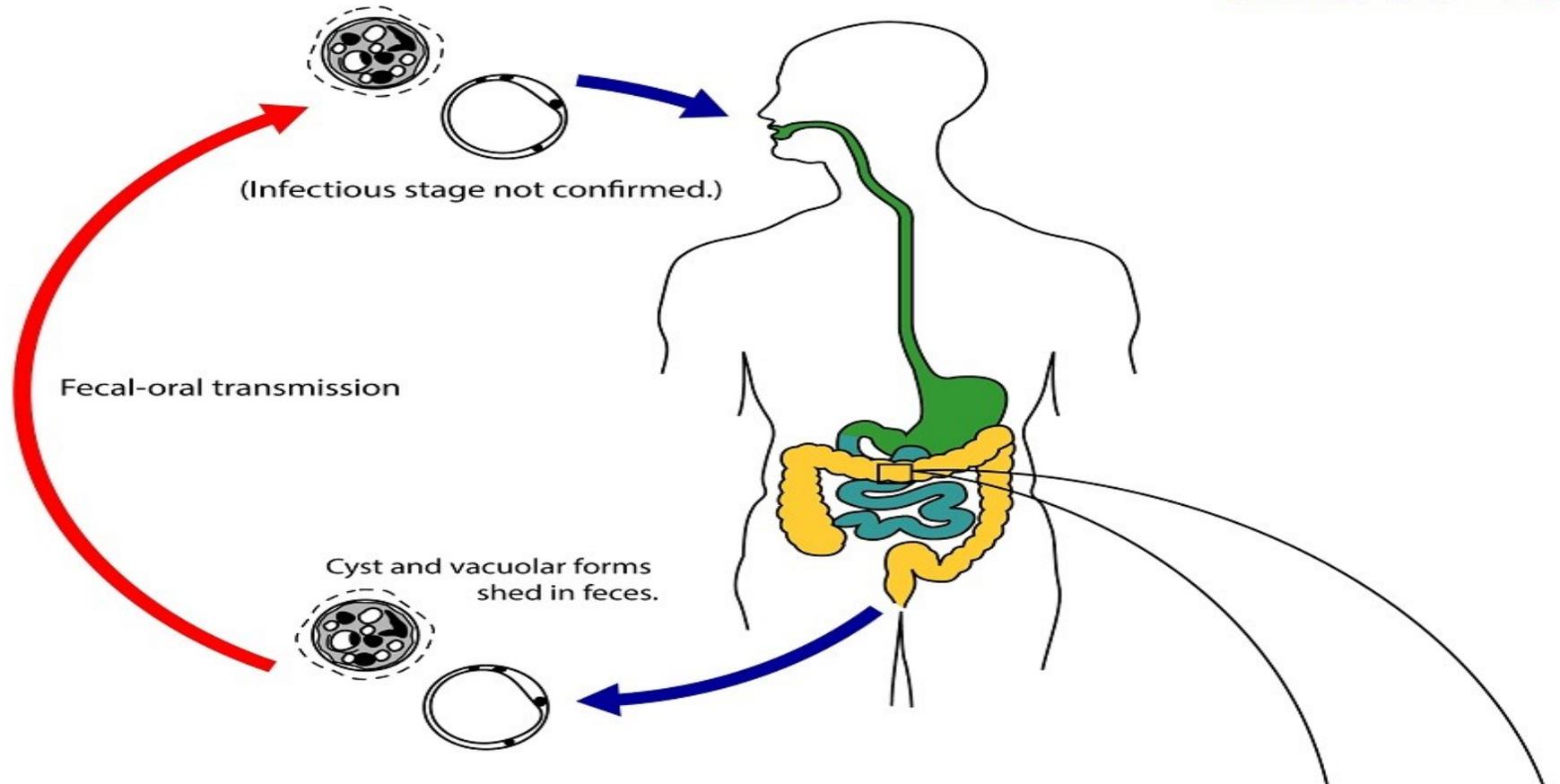


*Blastocystis hominis*  
5 to 30  $\mu\text{m}$

- Morphology of classic form\* (thick-walled cyst)
  - Size: range from 5 to 50  $\mu\text{m}$  (most are 10  $\mu\text{m}$ )
  - round w/ large central body ( large vacuole)
  - multiple, small , peripheral nuclei
  - in iodine wet mounts, cytoplasm is brown, central body does not stain
  - in trichrome , cytoplasm is dark green, central body may stain pale to intense green and the nuclei are dark purple
- Metronidazole is the drug of choice if treatment is needed

# *Blastocystis Hominis* Life Cycle

- Life cycle is not fully understood, as various morphologic forms of this polymorphic organism have been identified in stool or culture
- The cyst form (3–5  $\mu\text{m}$ ) is postulated to be an infectious stage, but not confirmed
- The predominant form found in human stool specimens is referred to as the vacuolar (or central body) form and is of variable size
  - (5–40  $\mu\text{m}$ , occasionally much larger)
- Other morphologic forms (e.g., ameboid and granular forms) also have been noted in stool samples and/or culture, but have unknown significance
- Replication appears to occur via binary fission

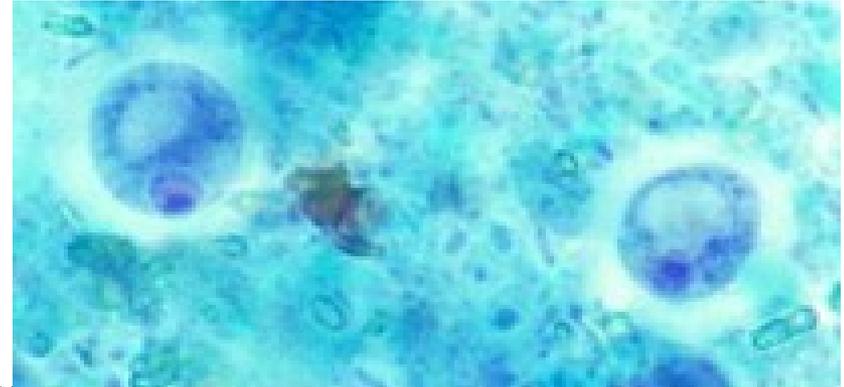
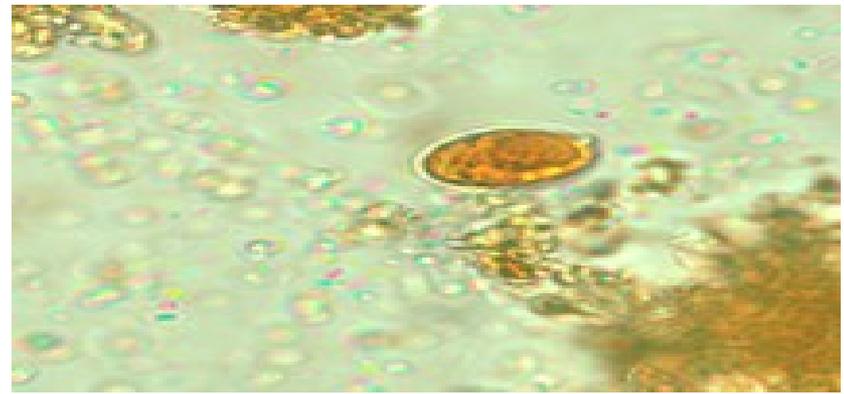


Cyst form	Vacuolar form	Binary fission / Mitotic forms	Granular form*	Amoeboid form*

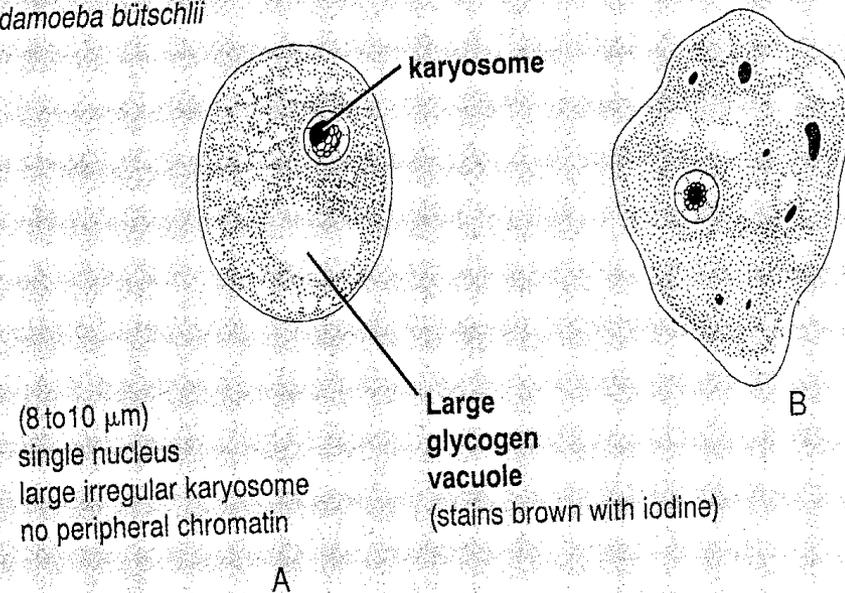
\*Various forms that may occasionally be seen in human stool samples and in culture. Their biological significance is not well understood.

# Iodamoeba butschlii: nonpathogenic

- Cyst: nearly spherical to ellipsoidal in shape
  - size: 5 to 20  $\mu\text{m}$
  - Single nucleus with a thin nuclear membrane
  - Nuclear appearance: large, eccentric, irregular karyosome w/o peripheral chromatin
- Cytoplasmic inclusions : large glycogen vacuole (unique & identifiable)
  - vacuole stains dark brown in iodine wet mount and empty in permanently stained smear

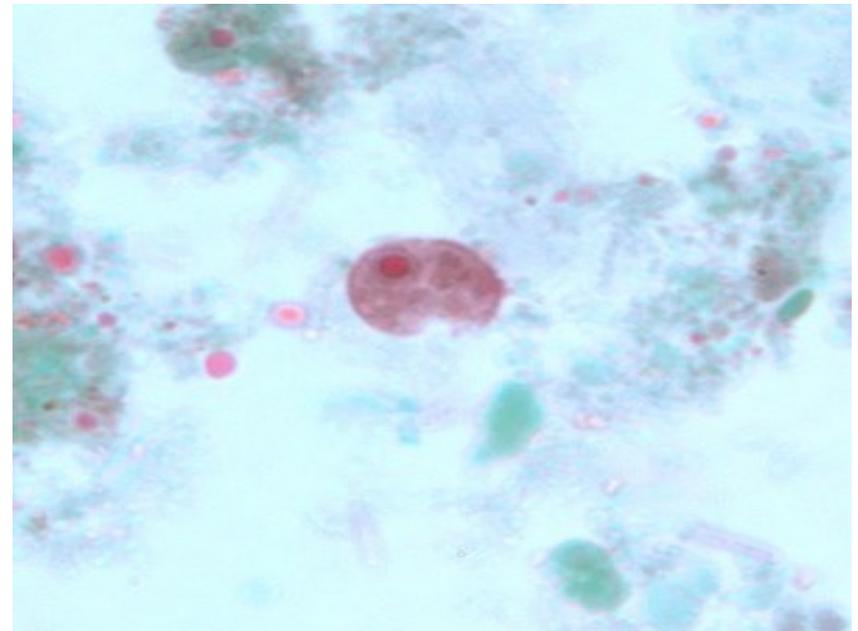
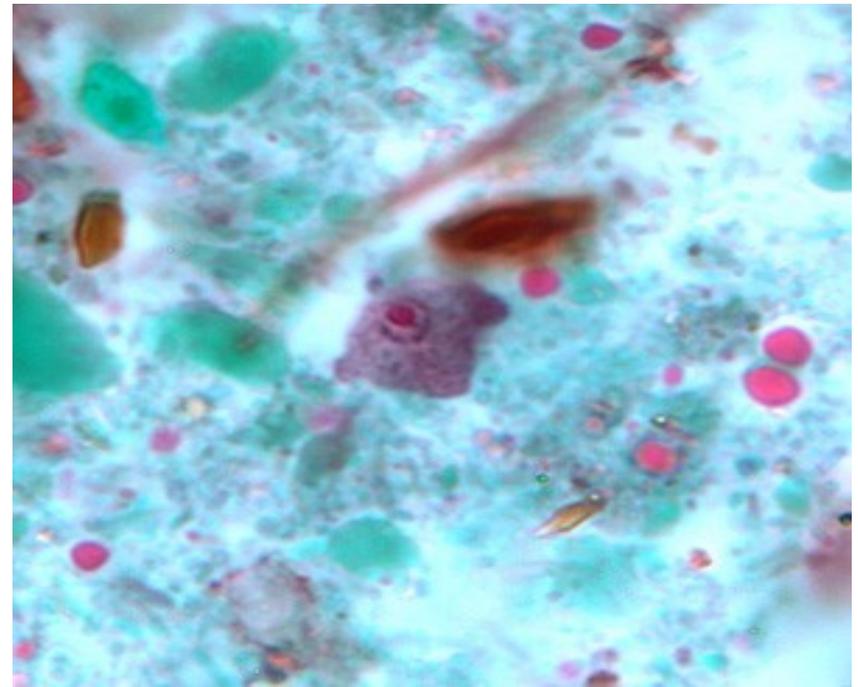


*Iodamoeba butschlii*



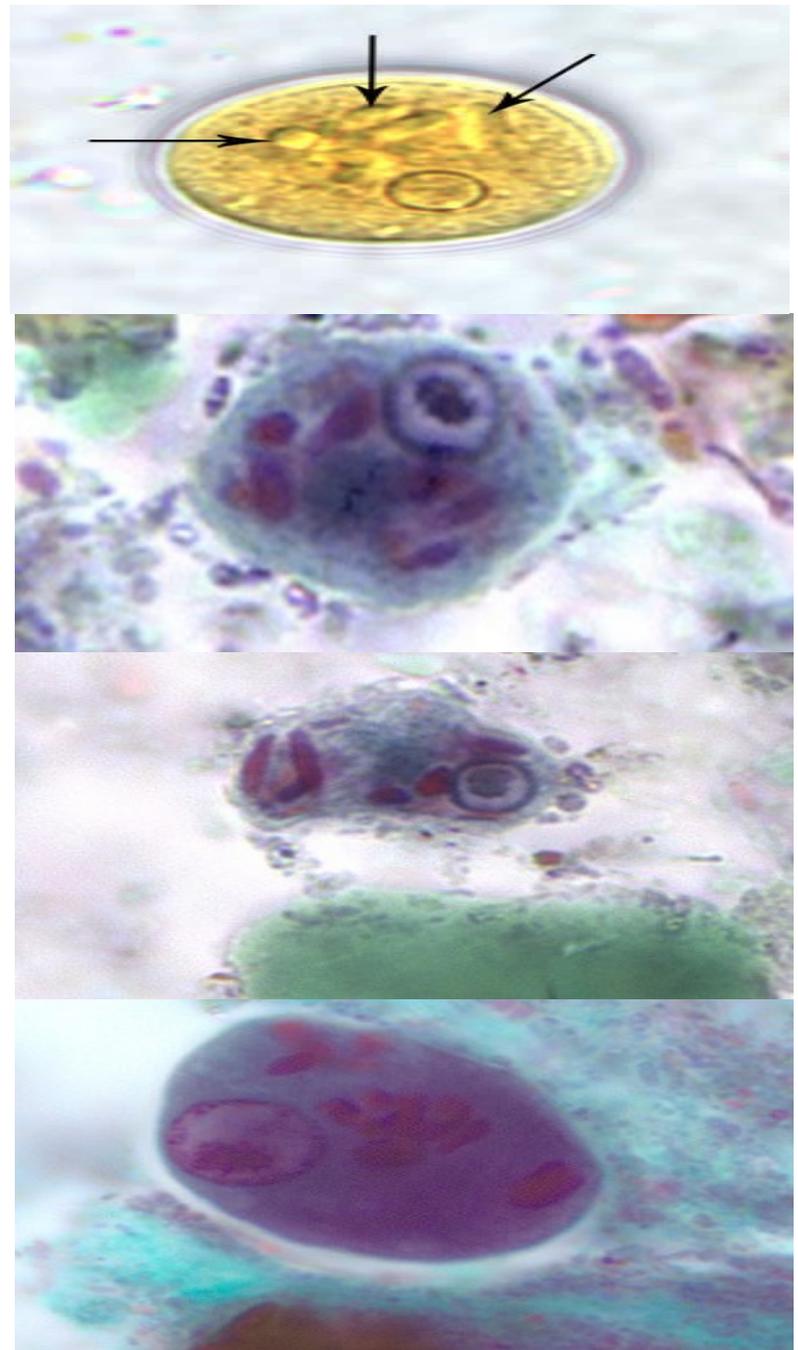
# Iodamoeba butschlii

- Trophozoite:
  - size: 8 to 20  $\mu\text{m}$
  - Number of nucleus: 1
  - Nuclear appearance: Centrally located karyosome with refractile, achromatic granules
  - Motility : sluggish, nonprogressive movement
  - Cytoplasmic inclusions : coarsely granular with vacuoles w/ ingested debris/bacteria
- Trophozoite may be confused with *Endolimax nana*



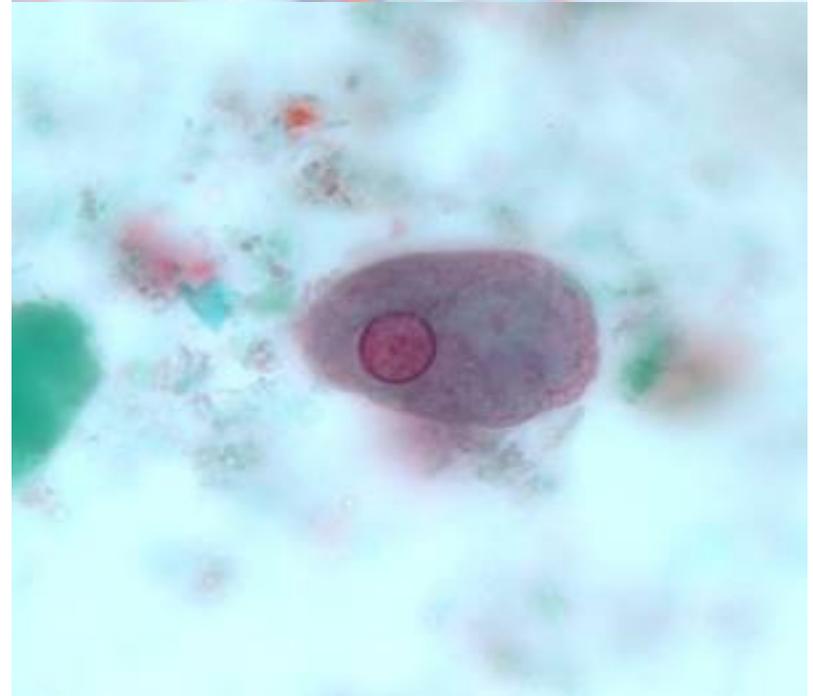
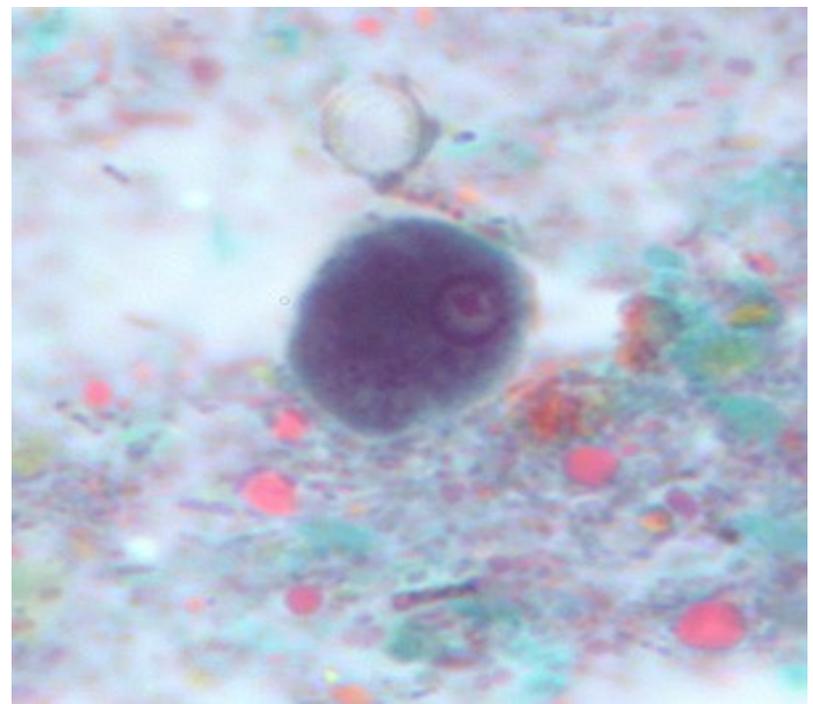
# Entamoeba polecki: nonpathogenic

- Primarily found in pigs and monkeys, but occasionally found in humans
- Closely resembles *E. coli* and *E. histolytica*
  - Must be identified to prevent unnecessary treatment
- Cyst form:
  - 9x25  $\mu\text{m}$  in size with one large nucleus (1/4 to 1/3<sup>rd</sup> of cell) with evenly distributed chromatin, and variable karyosome size
  - Large inclusion mass and an abundance of pointed chromatoidal bodies **unlike** the rounded ones of *E. histolytica*/*E. dispar*



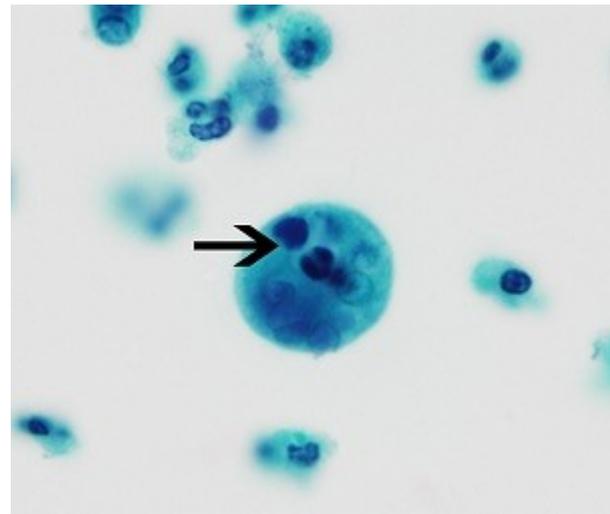
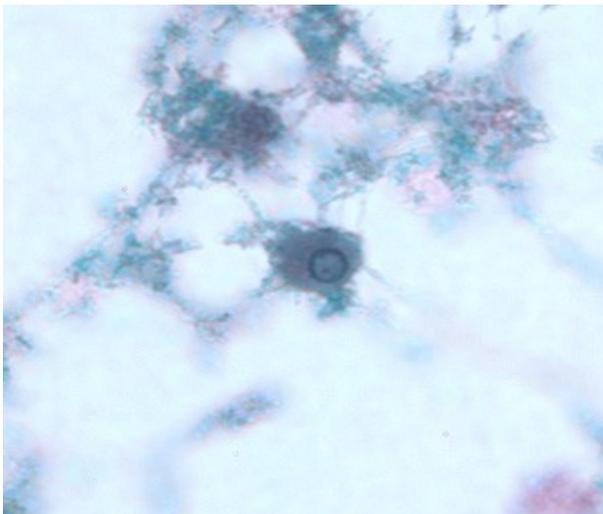
# Entamoeba polecki: nonpathogenic

- Trophozoites form:
  - Often rounded, measuring 10–25  $\mu\text{m}$
  - A single nucleus is often distorted and irregularly shaped
  - A small-to-minute centrally located karyosome
  - The peripheral chromatin is usually delicate and uniform
  - The cytoplasm is often vacuolated with a hyaline border
  - Blunt pseudopodia may be seen



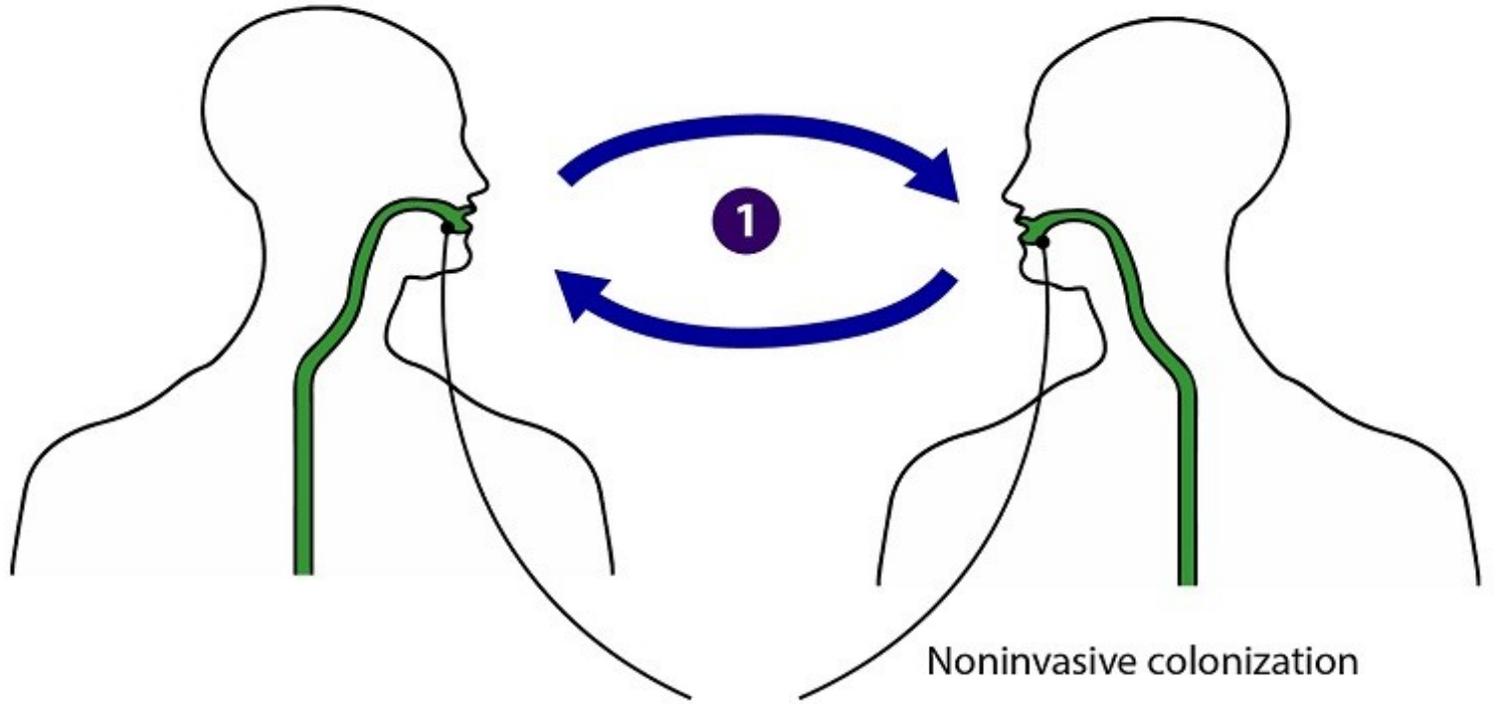
# Entamoeba Gingivalis: nonpathogenic

- found in the mouth, in soft tartar between teeth or in tonsillar crypts; occasionally found in sputum
- has no cyst stage; must be differentiated from *E. histolytica*
- the only species known to ingest WBC
- nuclear fragments of WBC can be seen in large vacuoles in permanently stained smears



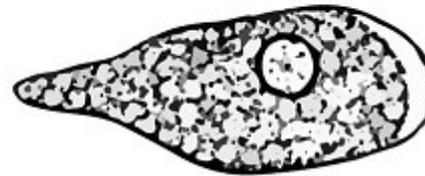
# Entamoeba gingivalis Life Cycle

- There is no known cyst stage for *Entamoeba gingivalis*
- Trophozoites live in the oral cavity of humans, residing in the gingival pockets near the base of the teeth
- They are not considered pathogenic and feed on bacteria and other debris
- Trophozoites are transmitted person-to-person orally by kissing or fomites (such as eating utensils)
- The trophozoite stage is morphologically similar to *E. histolytica* and requires differentiation
  - Both can be coughed up in sputum specimens (if *E. histolytica* is present in pulmonary abscesses)



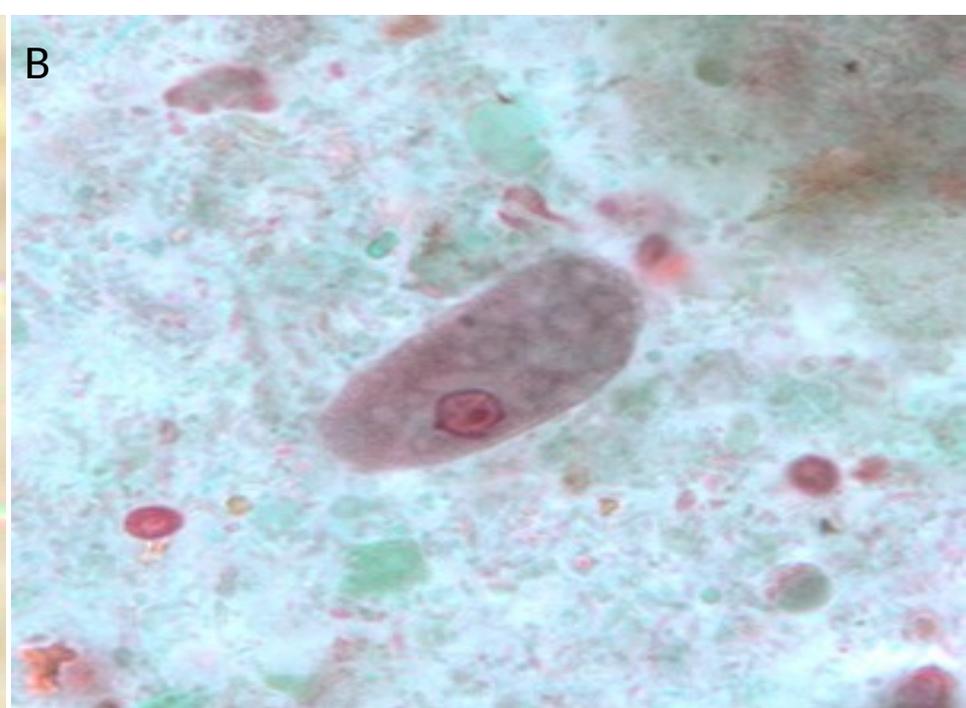
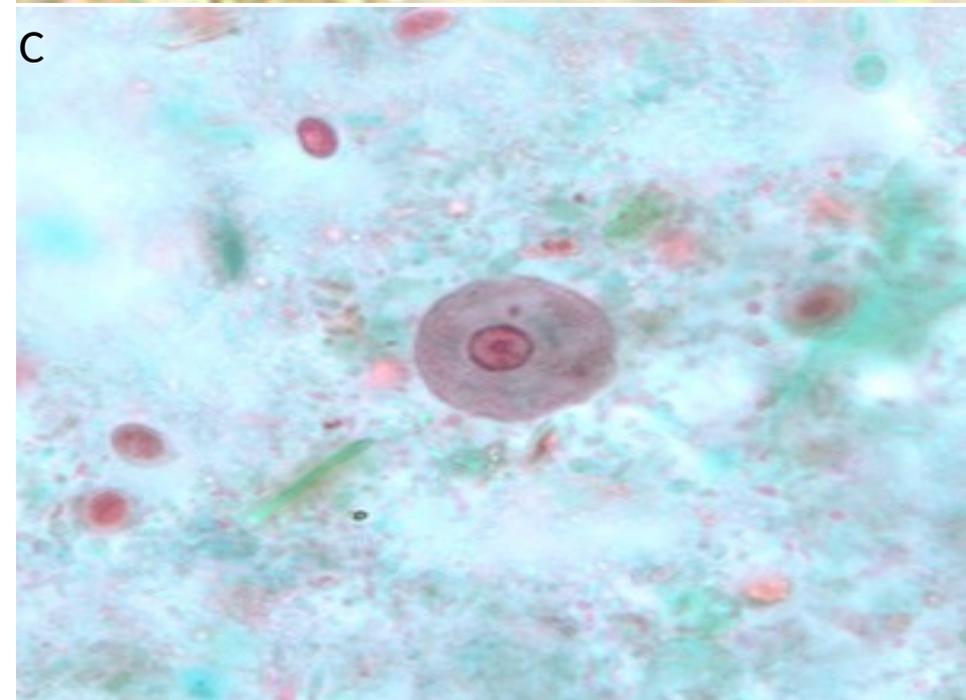
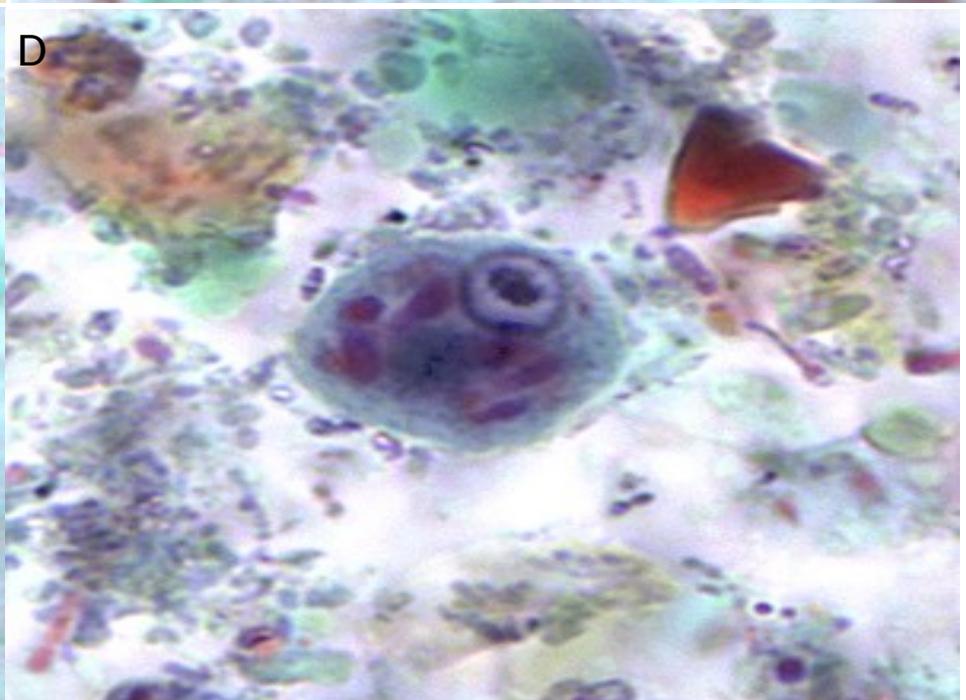
 Infective stage

 Diagnostic stage



Trophozoite



**A****B****C****D**

# Case Question 1

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A 34 year-old female presented to a Primary Care Clinic with bloody diarrhea, abdominal pain, loss of appetite, and dehydration. She is a product manager for a US company and had returned to Philadelphia 4 days ago from a 2-month business trip to Mexico. At the Clinic she states that she is passing 5 to 8 watery, bloody stools per day.

Her workup includes a blood culture, stool culture, and stool parasite exam. Her trichrome permanent smear is positive for an amoebic trophozoite, and the organism would most likely show the following nuclear morphology:

- a. Irregular, clumped, bot-like karyosome in nucleus without peripheral chromatin
- b. Eccentric karyosome in nucleus with irregular peripheral chromatin
- c. Central karyosome with eccentric nucleus with fine, regular peripheral chromatin
- d. Large, irregular karyosome in nucleus without peripheral chromatin, and with a glycogen vacuole in the cytoplasm