ENDEMIC SYSTEMIC MYCOSES

Systemic= throughout the body, in deep tissues

Disseminated = present in an organ other than at the original site of infection

Soil fungi are causative

Inhalation mode of acquisition

Asymptomatic $\rightleftharpoons$ symptomatic

Dimorphic 37^0 yeast pnase

30^0 mould (hyphal) phase

Patients with **normal immunity** mainly affected, but serious disease in immunocompromised occurs

Endemic systemic mycoses dimorphic fungi

Geographic variation in incidence

- *Histoplasma capsulatum*
- *Blastomyces dermatitidis*
- *Coccidioides imitis*
- *Paracoccidioides brasiliensis*
- *Penicillium marneffei*

Other dimorphic

- *Sporothrix schenckii*

HISTOPLASMOSIS

H. capsulatum

Dimorphic

Has sexual stage – *Ajellomyces caspsulatus*

Intracellular infection

World-wide, U.S. focus

N. America

- Mississippi Valley
- St. Lawrence Valley

Heavy soil contamination by bird or bat excrement

- birds not infected, bats may be symptomatic

Most infections asymptomatic

HISTOPLASMOSIS

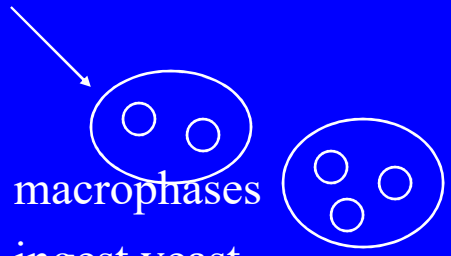
Pathogenesis

Respiratory Infections

inhale

spores

yeast form



macrophages in lung

macrophases ingest yeast

Lymph nodes
Liver
Spleen
Adrenal glands
Intestine
Bone Marrow

Proliferation halted by onset of acquired CMI at 10-14 d.

Vasculitis, tissue necrosis, **caseating granulomata**. Killing by macrophages, healing, calcification

Histoplasmosis
Clinical

- Primary respiratory infection
- Most initial infections are asymptomatic to mild (90-95%)
  - Flu-like illness (fever, headache, chills, chest pain, weakness, weight loss, muscle pain, fatigue, non-productive cough 3->10 d)
  - CXR
    - » normal to patchy infiltrates (lower lung), hilar and mediastinal lymphadenopathy
- +/- Pericarditis, arthralgias, arthritis, EM, EN
- May have severe pneumonia (ARDS) associated with hepatomegaly & splenomegaly
- May disseminate widely (1/2000 adults)

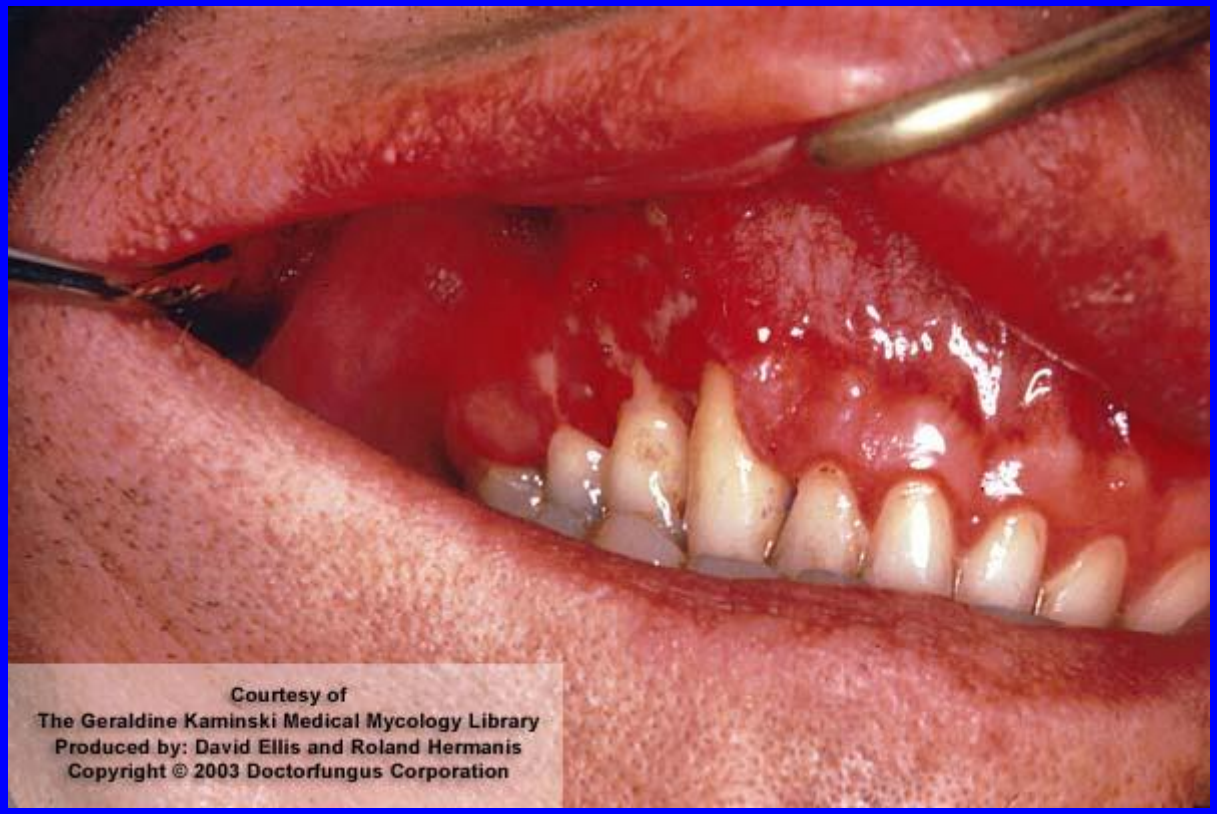
Histoplasmosis
Clinical

- **Progressive disseminated histoplasmosis**
  - **Defects in host immunity**
    - » **Infants, immuncompromised, HIV**
  - Acute, subacute, chronic
  - Failure of macrophages to kill fungus
  - Diffuse spread throughout MPS
    - » Oropharyngeal ulcers
    - » Hepatosplenomegaly
    - » Adrenal
    - » GI
    - » Endocarditis
    - » Meningitis
    - » Brain abscess
    - » Lymphadenopathy
    - » Coagulopathy
    - » Bone marrow suppression **(pancytopenia)**

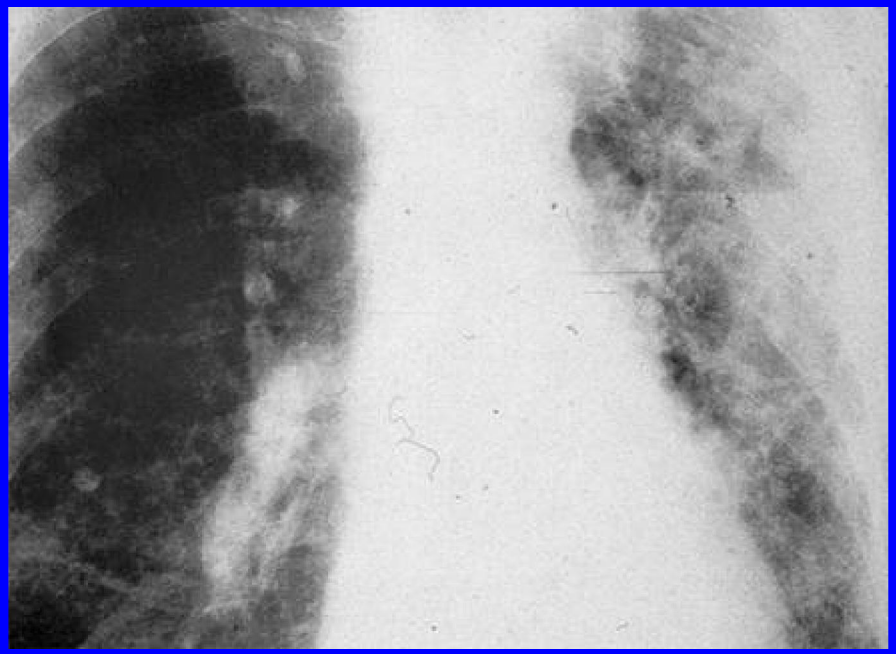
Histoplasmosis
Clinical

- **Chronic pulmonary histoplasmosis** (1/100,000)
  - pre-existing structural lung defect, i.e. COPD, emphysema
  - chronic pneumonia or infection in cavities, increased sputum
  - reactivation or reinfection
  - apical infection, may be cavitary
- **Mediastinal granulomatosis** and fibrosis
  - fibrosis, traction, occlusion of mediastinal structure
- **Histoplasmoma**
  - Fibrocaseous nodule
  - Concentric caseation and calcification
- **Presumed ocular histoplasmosis syndrome**
  - choroiditis - active or inactive
    - » may result in visual loss due to macular involvement

Oral histoplasmosis



Histoplasmosis



Chronic fibrocavitary histoplasmosis

Histoplasmoma

Histoplasmosis

Diagnosis

1. Obtain appropriate specimens

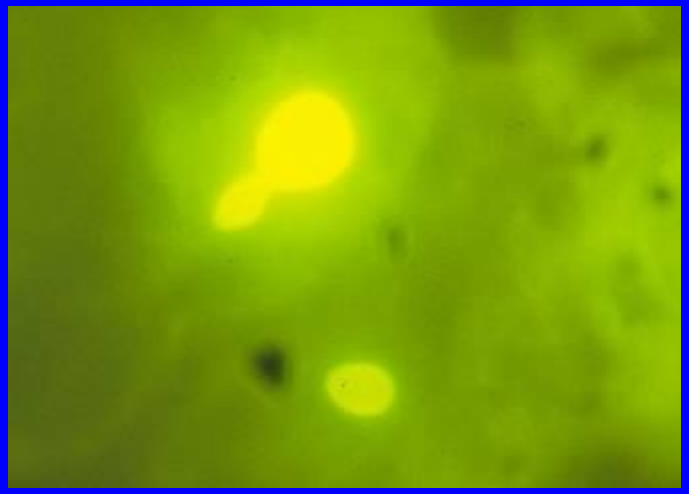
sputum	bone marrow
blood	lesion scrapings
urine	biopsy specimens

2. Direct Examination

- Tissue Specimens
  - stains for fungi - PAS, GMS, Giemsa
  - routine histology - H & E

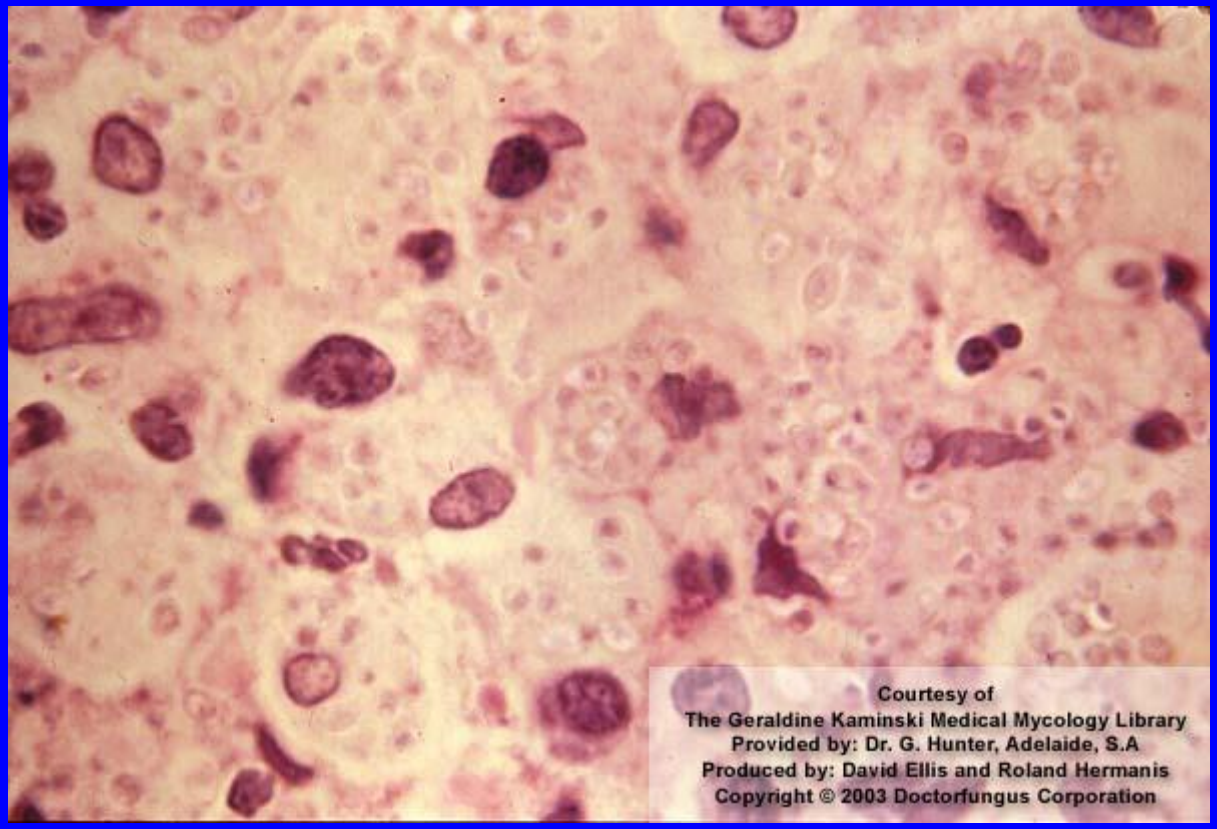
  - small yeast (2-4 μ) intracellular in macrophages
  - granulomas - non-caseating
    - caseating
- Sputum - KOH or calcofluor

Calcofluor stain x400



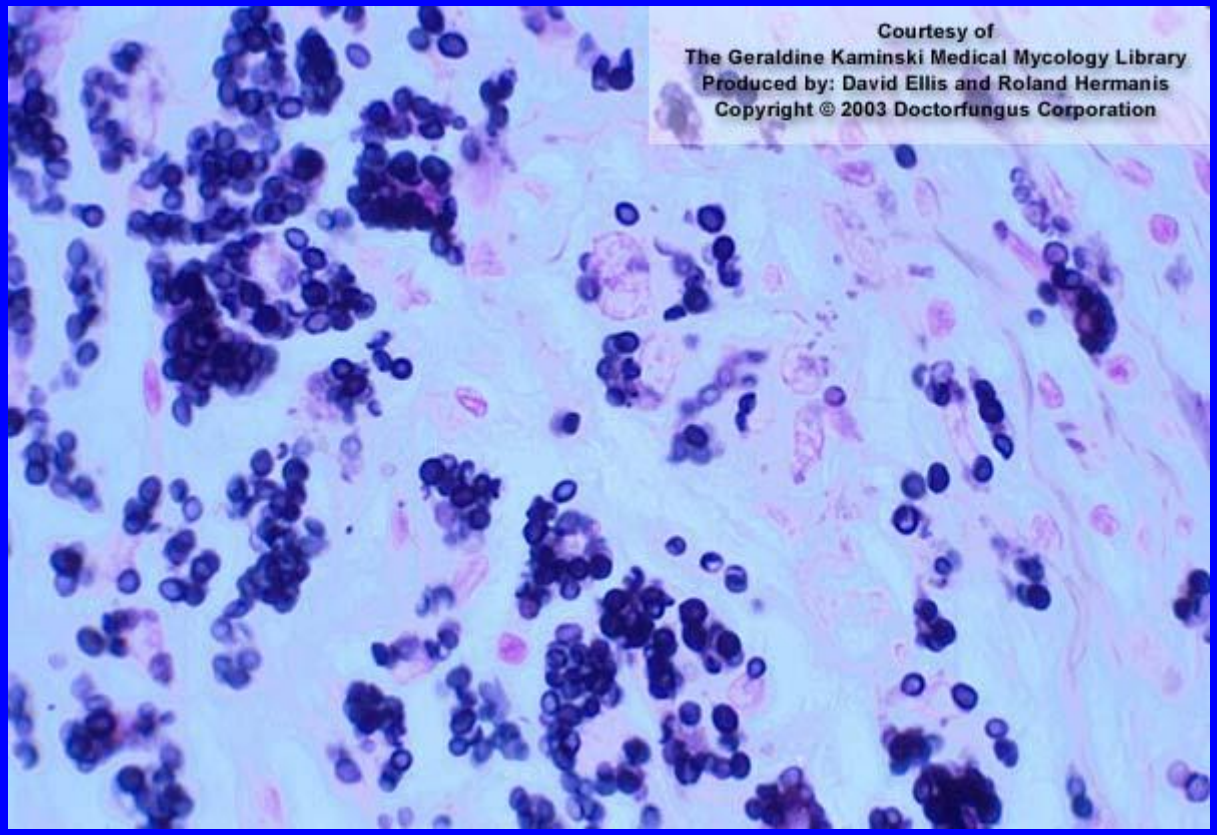
Narrow-neck bud

H&E mouth biopsy



Yeast in macrophages

GMS lung biopsy



Histoplasmosis (diagnosis cont.)

3. Culture

Sabouraud's agar

- White - brown mould
- Typical microscopic morphology

Slow growth 2-8 weeks

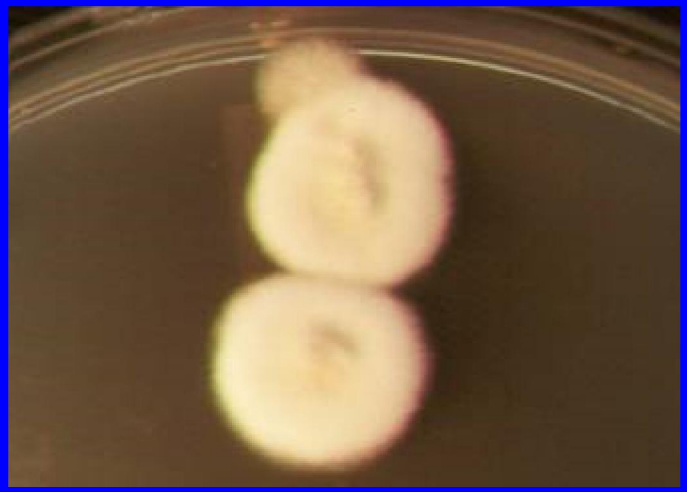
Rapid ID confirmation

- Exo-antigen
- Molecular probe

Traditional ID confirmation

- Conversion mould to yeast
- Animal inoculation

Macroscopic morphology
Sabouraud's dextrose agar



Mould at RT

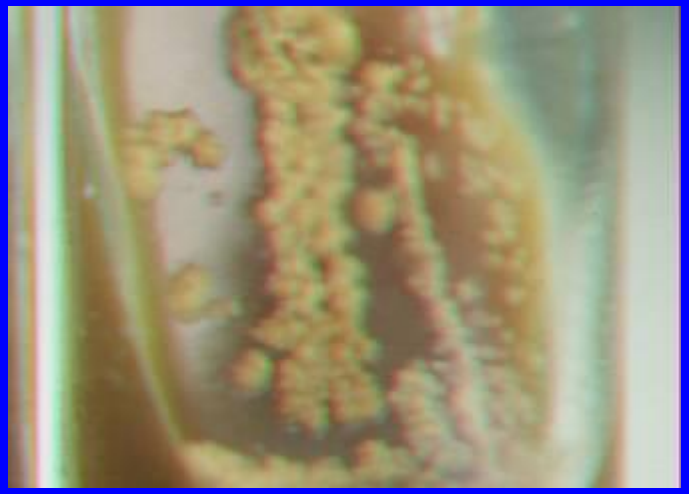
Microscopic morphology

Tuberculate macroconidia

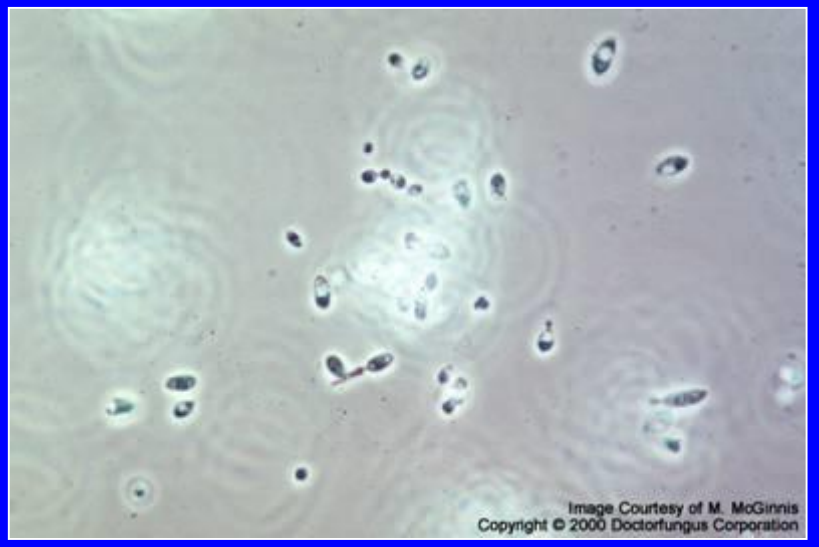
Microscopic morphology

Tuberculate macroconidia and microconidia

Hyphal to yeast conversion at 37ºC



Yeast-like colonies



Yeast cells

Diagnosis (cont.)

- 4. Serology
  - Sensitivity and specificity vary according to stage and form of disease
    - » Lowest for early acute pulmonary and disseminated (sensitivity 5-15% at 3 weeks)
    - » Highest for chronic pulmonary and disseminated (sensitivity 70-90% at 6 weeks)
  - Complement fixation test (CFT)
    - » Yeast (more sensitive) and mycelial (histoplasmin) phase antigens required
    - » ≥1:32 or 4-fold rise suggests recent infection
    - » X-reactions with *B. dermatitidis* and *C. immitis*

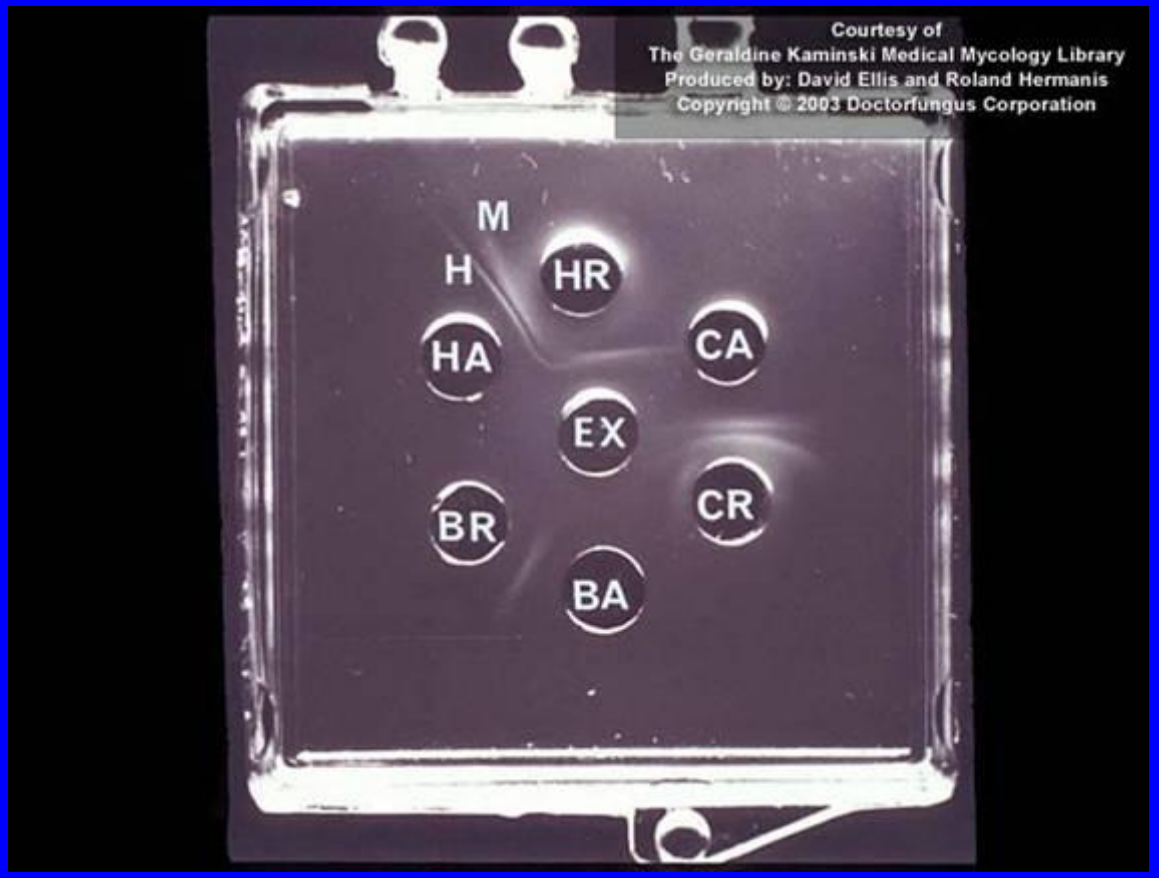
Diagnosis (cont.)

- Immunodiffusion
  - More specific, less sensitive
  - M bands
    - » Prior exposure
    - » Acute and chronic diseases
    - » X-reactions occur with other fungi
  - H bands
    - » Diagnostic of acute disease
    - » Revert to negative in 6 months
    - » Acute or chronic
    - » Little cross-reaction with other fungi
    - » Appear later than CFT Abs
- ELISA/RIA
  - Increased sensitivity (90% active pulmonary histo)
  - Decreased specificity compared to CFT

Diagnosis (cont.)

- Ag detection
  - Urine
  - Most useful in patients with large fungal burden
    - » Acute pulmonary histo (80% sensitive)
    - » Progressive disseminated histo (90% sens)
  - Less useful with lower fungal burdens
    - » Chronic pulmonary (15% sensitive)
    - » Subacute pulmonary (30% sensitive)
  - Serum sensitivity is lower
  - Cross-reactions with *B. dermatitidis* and recipients of anti-thymocyte globulin
  - Joe Wheat, MiraVista Diagnostics, Indianapolis

Exoantigen test



H and M bands

Histoplasmosis treatment

1. Immunocompetent (acute pulmonary, localized, disseminated, including meningitis)

- Mild – none
- Moderate - Itraconazole 200 mg pO OD x 9 mo or increased oral dose until response or IV
- Severe - IV amphotericin B

2. Immunocompromised

- Moderate to severe
  - Ampho B IV (total 10-15 mg/kg) or liposomal AMB
  - Itraconazole suppressive 200 mg OD
- Less severe
  - Itraconazole 300 mg pO BIDx 3d, then 200 pO BID x 12 wk, then 200 mg pO OD

BLASTOMYCOSIS

Blastomyces dermatitidis

Dimorphic

Has sexual stage – *Ajellomyces dermatitidis*

Not intracellular infection

Mainly N. America (also S. America, Africa, Mid-East)

N. America

- Mississippi, Missouri and Ohio
- Great Lakes and St. Lawrence River

Exposure to soil ↑↑ risk of infection

Warm, moist soil of wooded areas

- Rich in organic debris

Hard to isolate from environmental sources

Blastomycosis

Pathogenesis

Primary pulmonary infection

↓

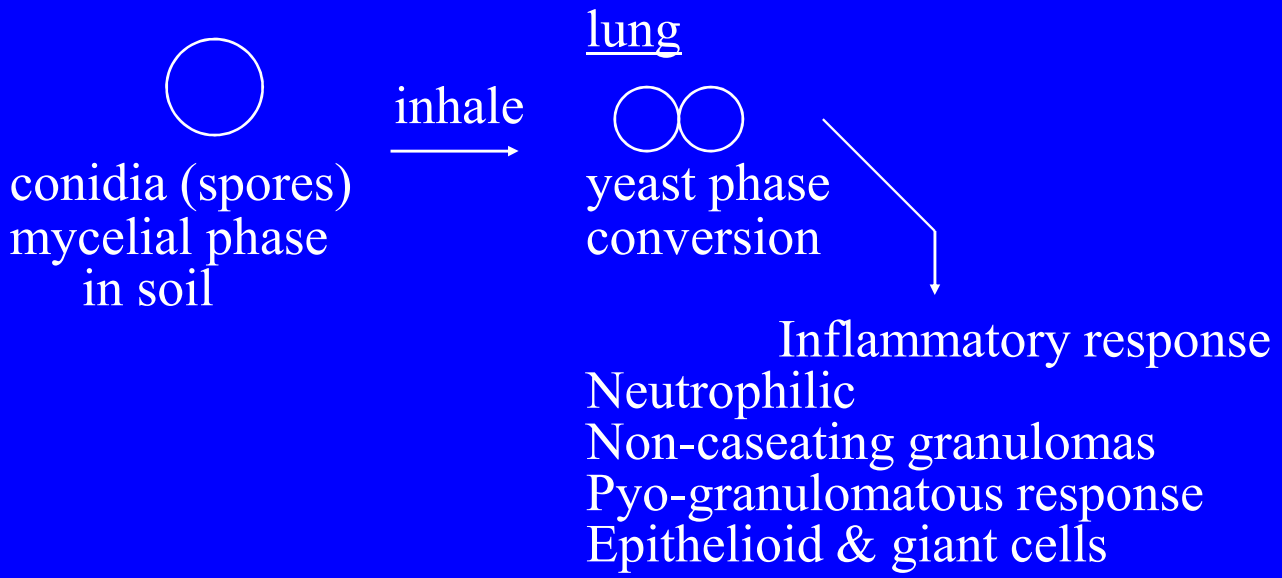
Dissemination

↓

Broad clinical manifestations

Especially bone, skin, lung, genito-urinary system

Chronic granulomatous and suppurative infection



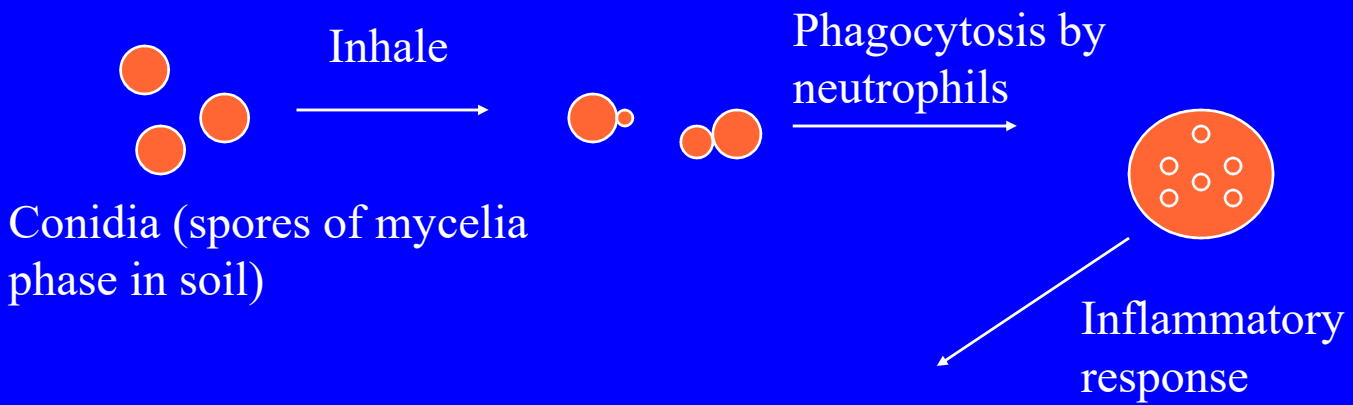
Blastomycosis

Pathogenesis

- Primary pulmonary infection
  - Conidia phagocytosed by neutrophils, but not killed. Macrophages may kill conidia
  - Condia germinate into yeast forms in lung, grow and disseminate via bloodstream
    - » Bone, skin and genito-urinary system
  - Development of CMI creates pyogranulomatous response (neutrophils and macrophages)
    - » Non-caseating granulomata

Histoplasmosis

Pathogenesis



Neutrophils and macrophages
Pyogranulomatous response
Non-caseating granulomata
Epithelioid and giant cells

Blastomycosis
Clinical

Spectrum of Infection

Asymptomatic ⟷ Symptomatic (most)*

1. **<u>Acute</u>**

   Flu-like symptoms (fever, muscle pain, joint pain, chills, chest pain, cough)

   Pneumonia

   Spontaneous resolution is rare

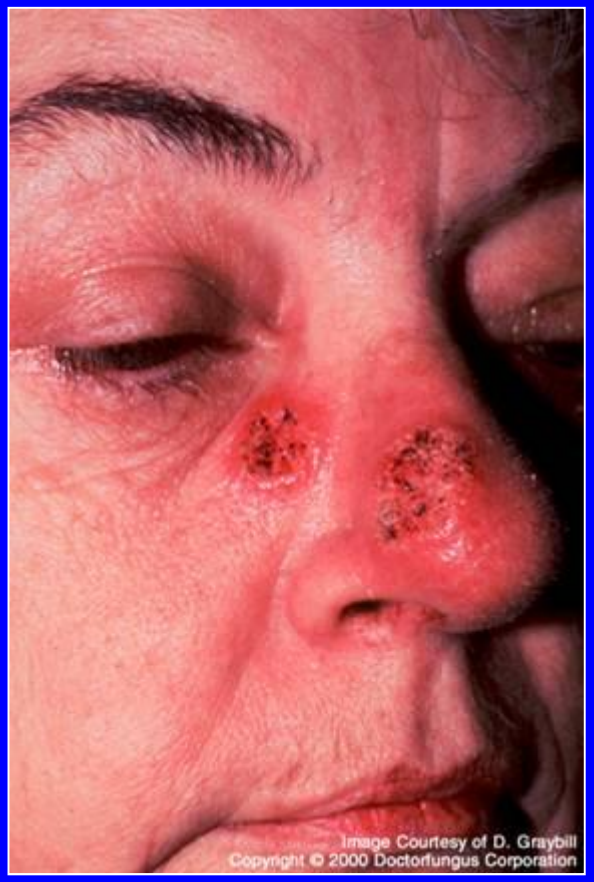
   Most patients go on to chronic or recurrent infection

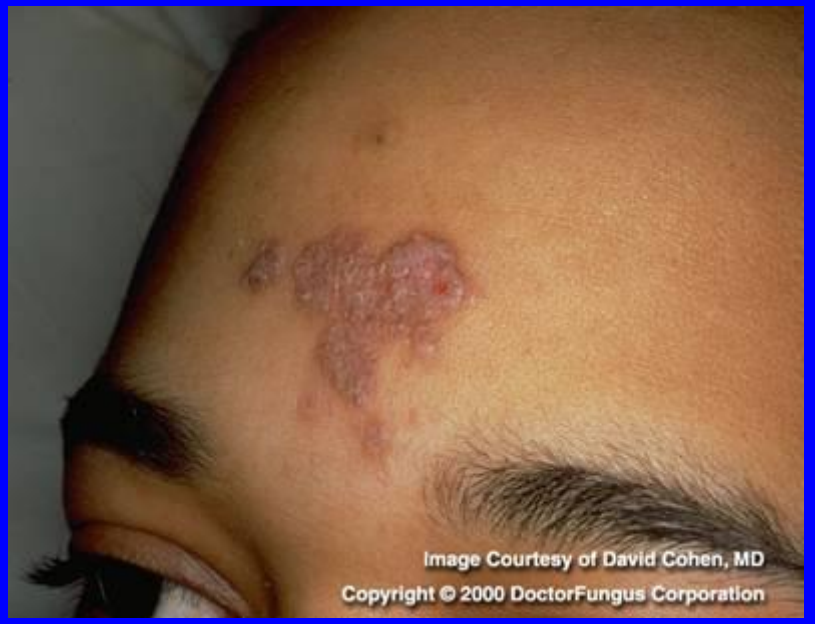
   CXR – lower lobe consolidation

Blastomycosis
Clinical

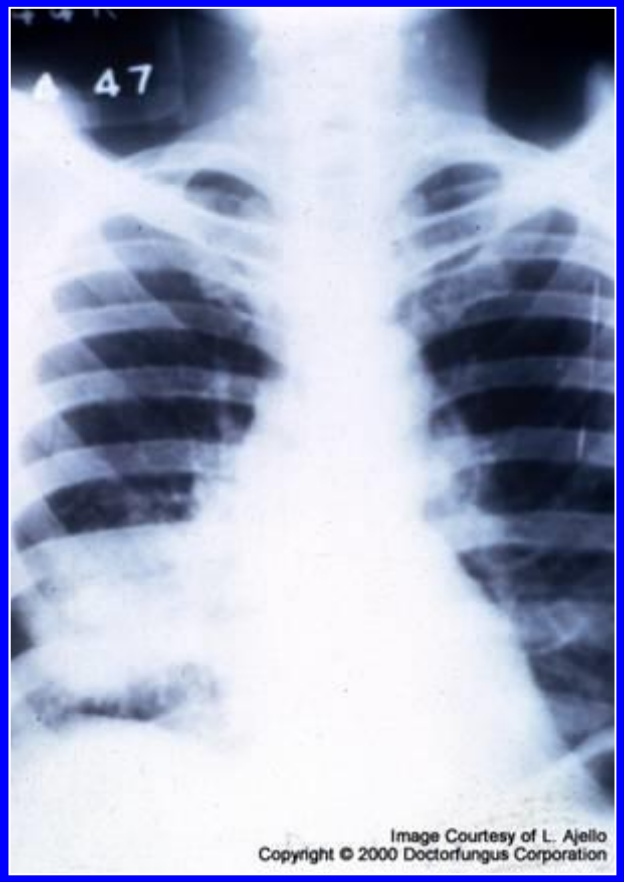
2. **Chronic/Recurrent**
   - Pulmonary - chronic pneumonia
     - cavitation
     - pleural involvement
   - Skin 40-80% skin and mucosa
     - pustular (verrucous), heaped-up
     - ulcerated lesions
   - Subcutaneous nodules
   - Bone/joint infection
   - GU tract - prostate, epididymis

Blastomycosis - skin

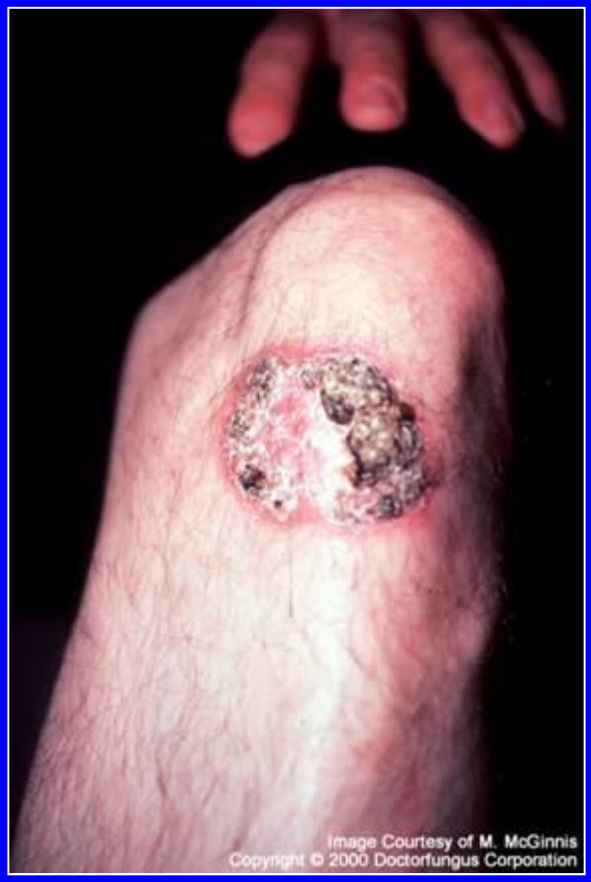




Blastomycosis

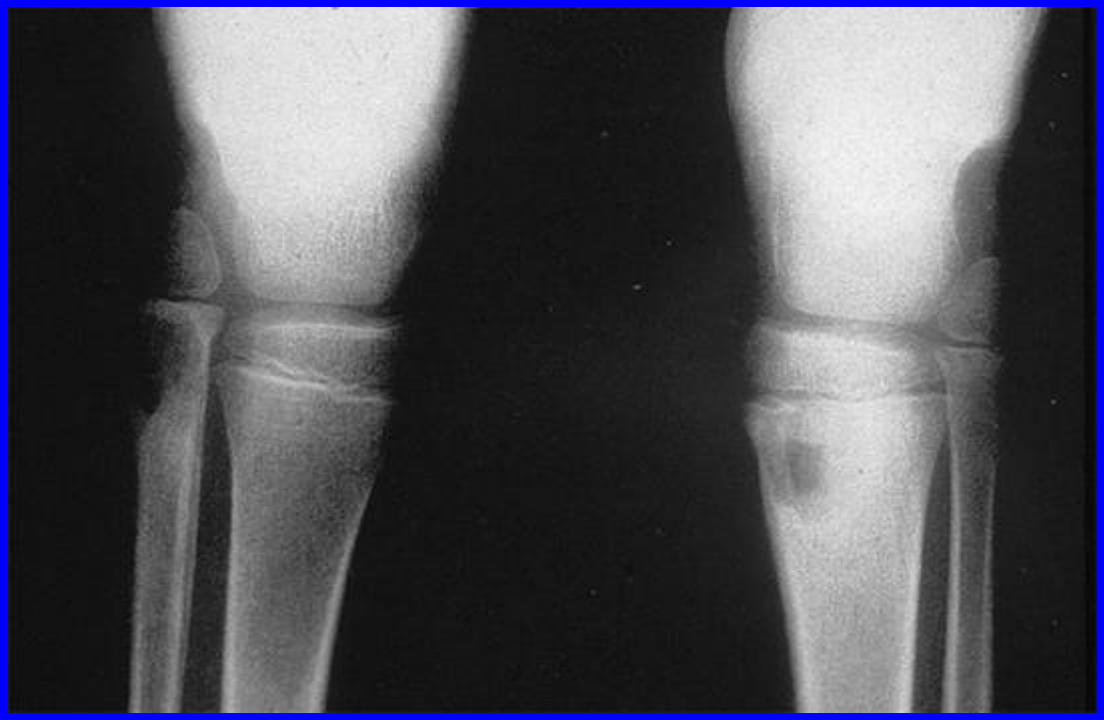


Lobar pneumonia



Verrucous knee lesion

Blastomycosis



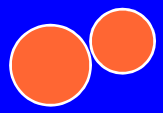
Osteomyelitis

BLASTOMYCOSIS
DIAGNOSIS

1. Direct Examination

Sputum	Prostatic fluid
Pus (skin, etc.)	Urine
Biopsy	

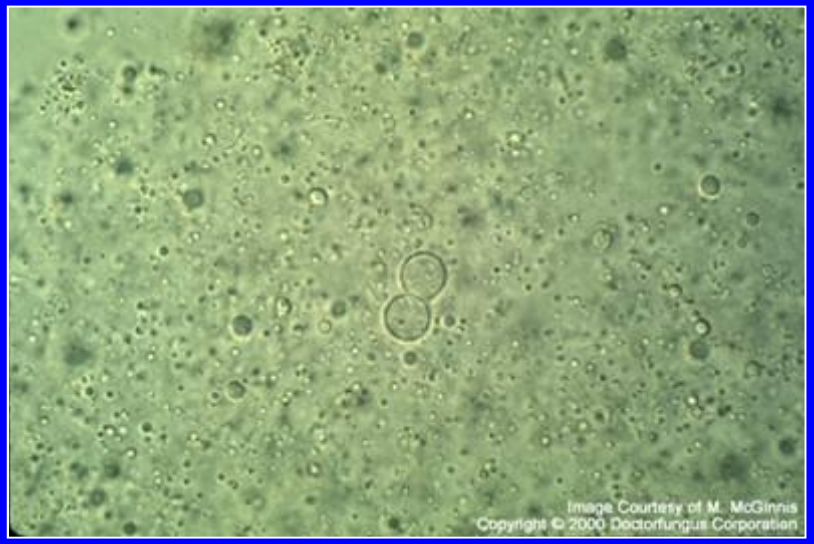
- KOH, calcofluor - yeast
- PAS, GMS, H & E



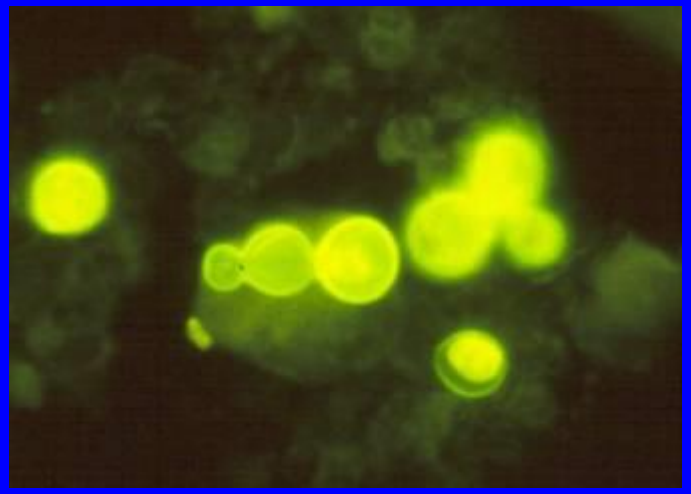
8 - 15 μ

Pyogranulomas
Thick walls
Broad-based buds

Direct examination
KOH and calcofluor

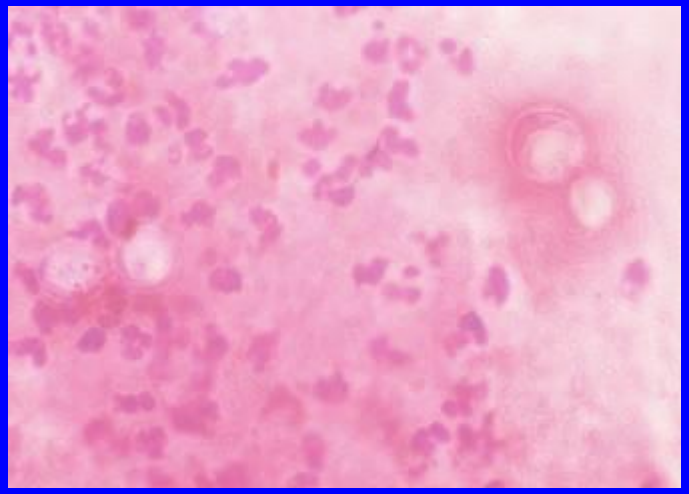


KOH

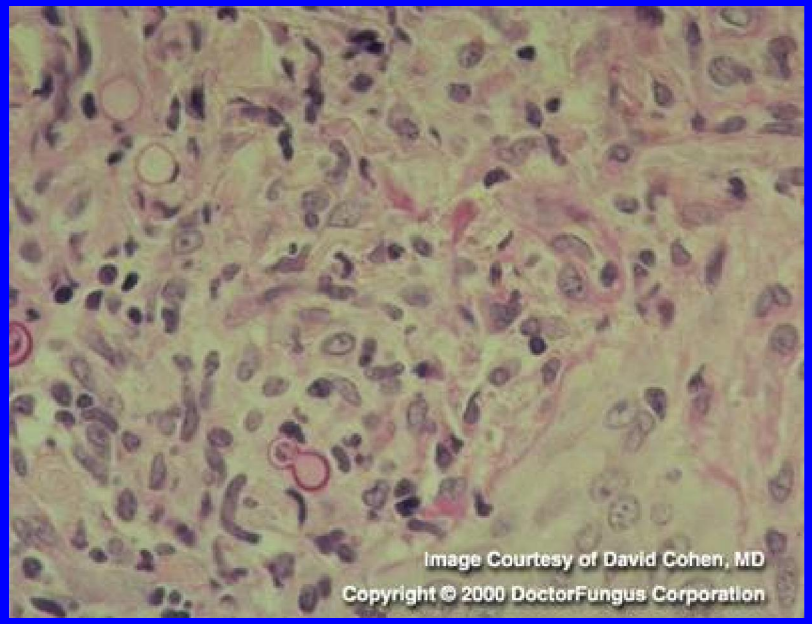


Calcofluor

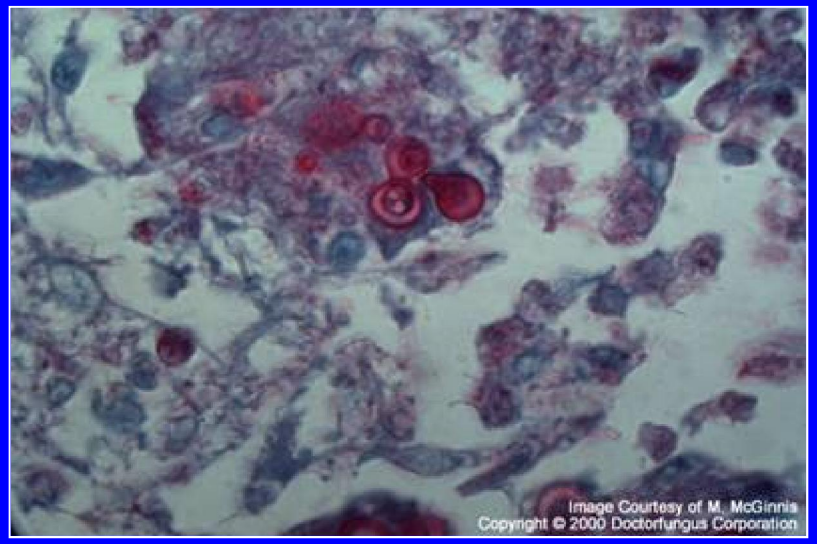
Gram stain



Tissue stains



Skin biopsy PAS



Brain biopsy PAS

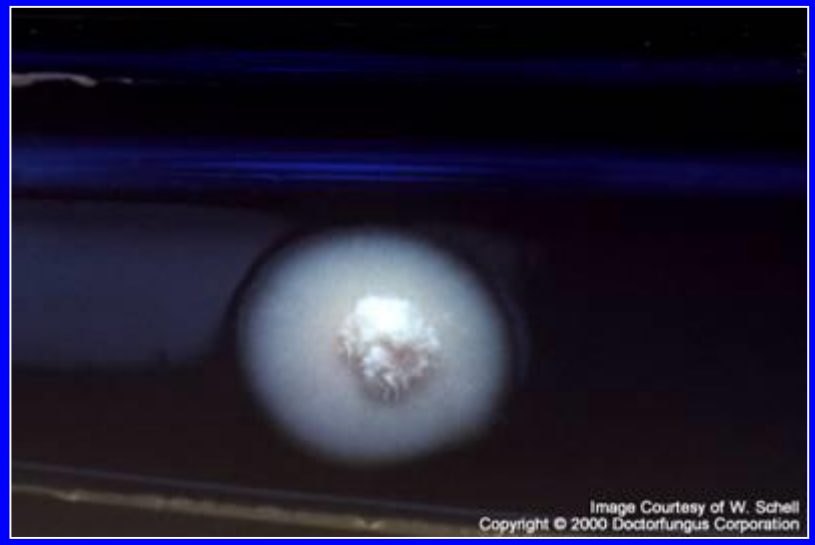
BLASTOMYCOSIS
DIAGNOSIS (cont.)

2. Culture

   - slow growth ~ 14 days up to 8 weeks
   - mycelial form at 30^0C white to light brown

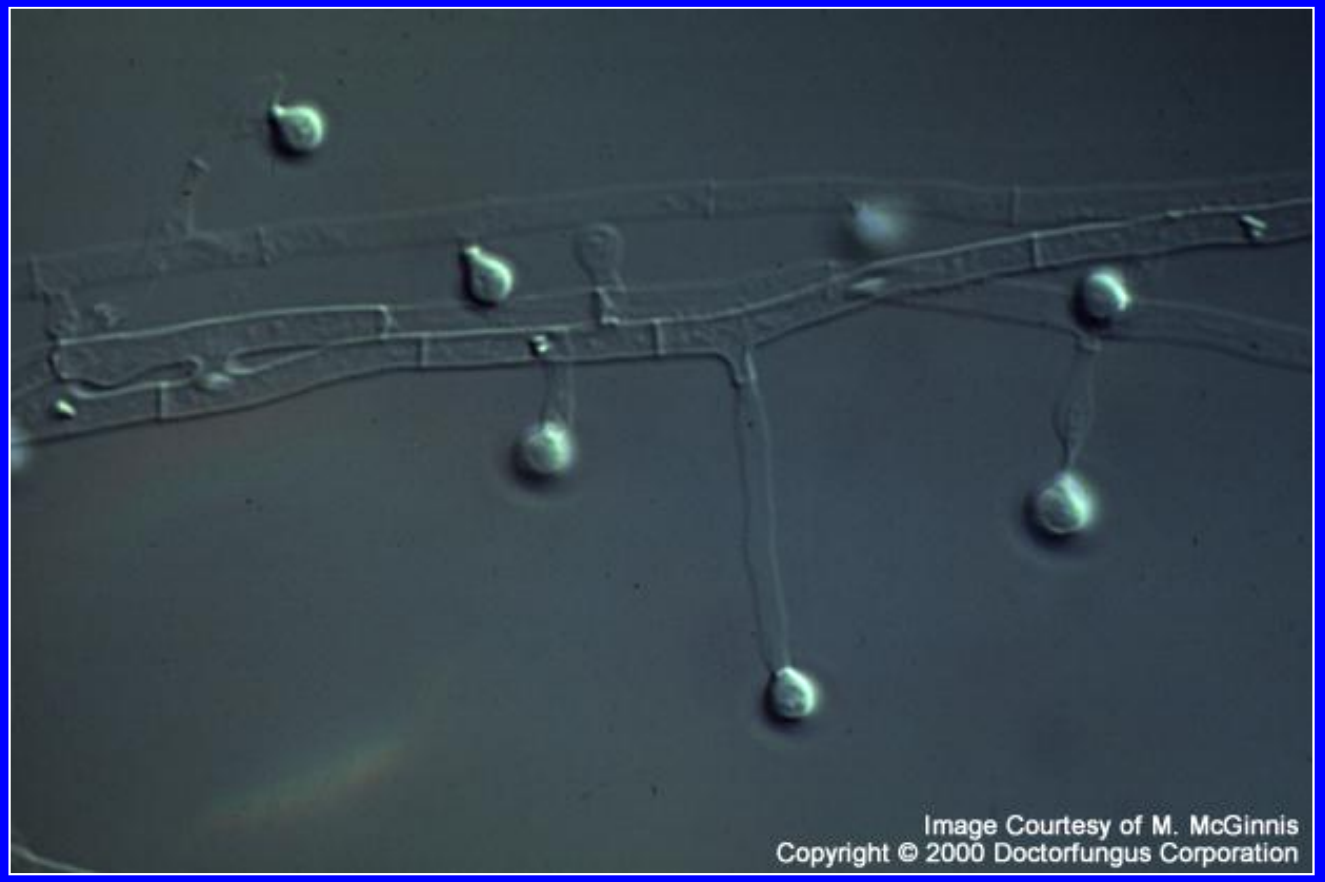
- Conversion to yeast form at 37^0C necessary for I.D. or exo - Ag or molecular probe

Mould phase at RT



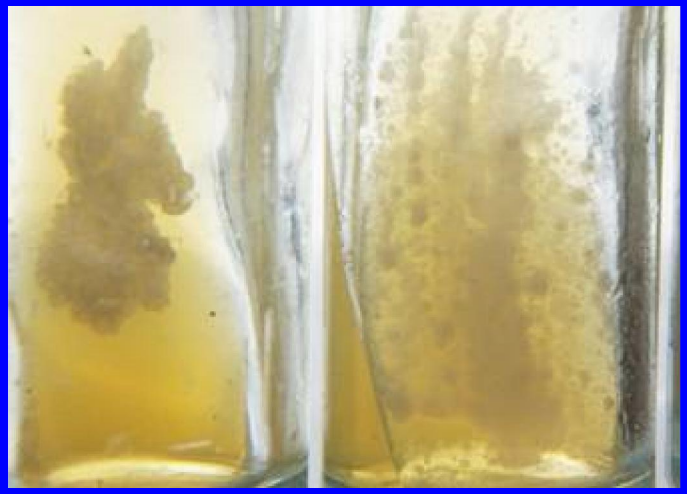
Reverse

Microscopic morphology of mould phase

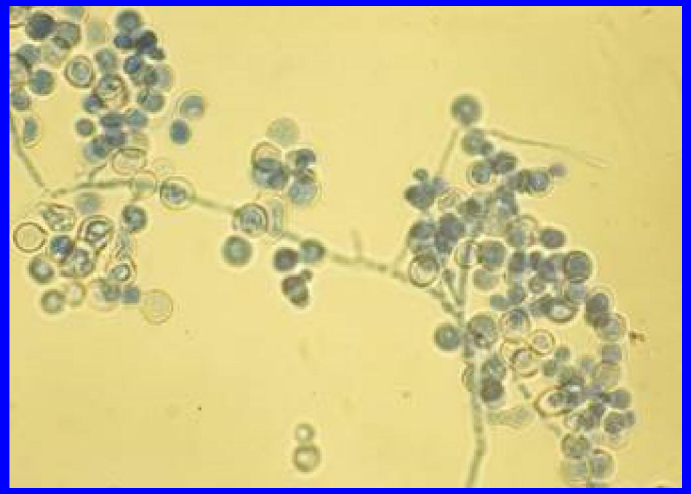


Lollypop-like conidia attached directly to hypha
Via conidiophore. No macroconidia

Conversion to yeast phase at 37ºC



macroscopic



microscopic

BLASTOMYCOSIS
DIAGNOSIS (cont.) - **SEROLOGY**

More likely to be positive later in disease (>6 wk)

- **Complement Fixation Test (CFT)**
  - Insensitive (<50%)
  - Non-specific (X-reactions)
- **Immunodiffusion**
  - Sensitivity 52-80% using A Ag
  - Good specificity >90%
  - Disseminated disease 88% positive
  - Local disease 33% positive
- **ELISA**
  - Best with A Ag
  - Sensitivity 80%
  - Specificity 80-92%
  - Single titre of ≥1:32 strongly supports diagnosis, 1:8 – 1:16 suggestive

BLASTOMYCOSIS
TREATMENT

Treat all active cases

1. *Itraconazole* (200-400 mg OD x 6 months)
   Except CNS blastomycosis

2. *Amphotericin B IV* (x 6-10 weeks)
   Life-threatening disease
   CNS disease
   Lack of response to ICZ
   ICZ toxicity

COCCIDIOIDOMYCOSIS
COCCIDIOIDES IMITIS

Regional mycosis - N. American significance

- Desert Southwest
- "San Joaquin Valley fever"
- Arid, rare freeze, low altitude, alkaline soil and sparse flora

Prevalence Endemic Areas

- 1/3 infected
- Annual incidence (symptomatic) 0.43%

Pathogenesis

- Inhalation of arthroconidia (spores)
- Present in soil (mycelial phase)
- Arthrospore → lower airway
  ↓
  disease

CLINICAL COCCIDIOIDOMYCOSIS

1. Primary Infection

   Pulmonary - 40% symptomatic

   1-3 weeks incubation

   1) Acute Valley Fever

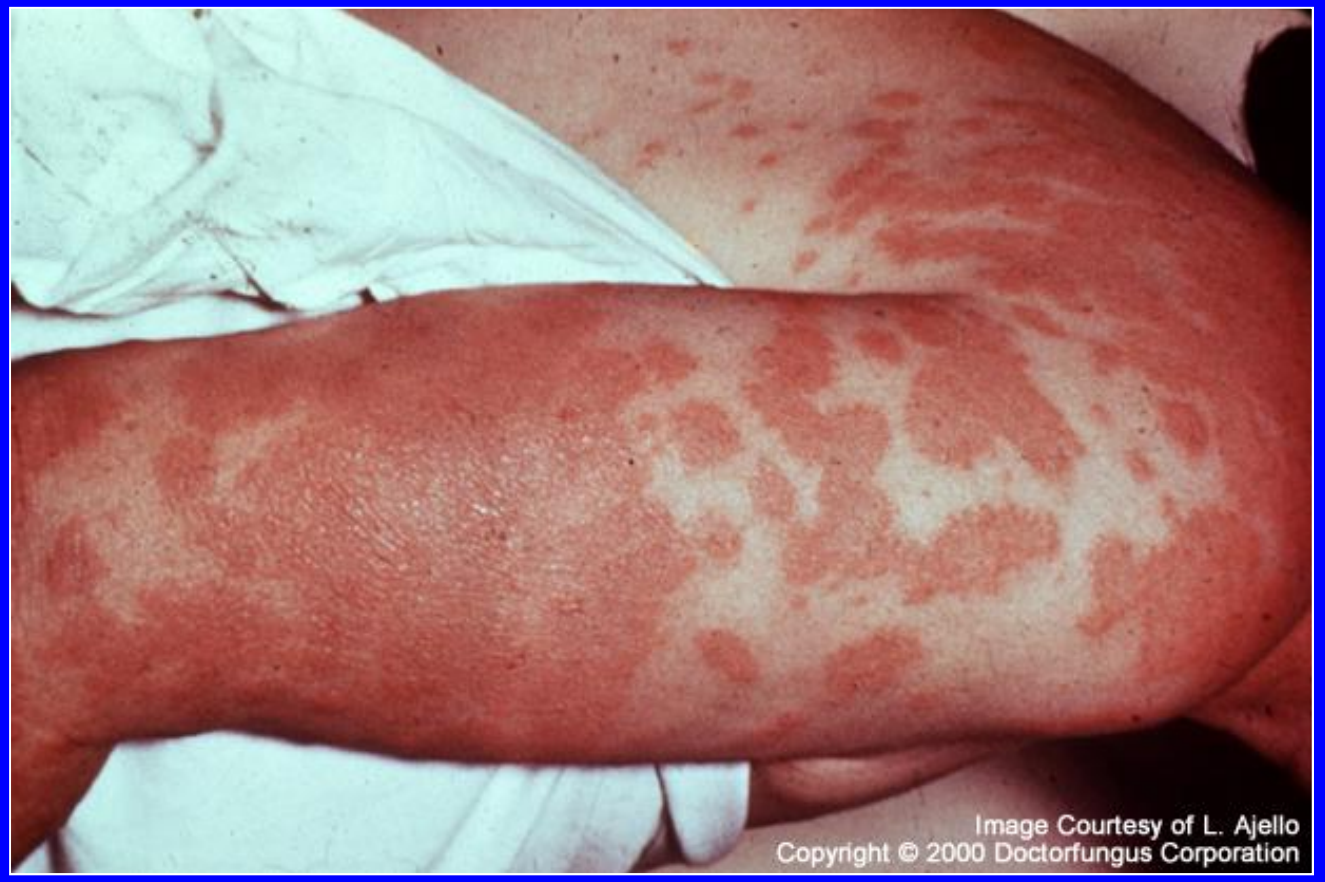
      EN or EM
      Arthralgia } 1/4
      Fever

   2) Skin Rash

      Erythroderma

      Maculopapular rash

Primary coccidioidomycosis



Acute allergic cutaneous lesions

CLINICAL COCCIDIOIDOMYCOSIS
(cont.)

3) Chest Pain
   - Pleuritic
   - Cough, sputum

4) Eosinophilia
   - Peripheral and tissue

CLINICAL COCCIDIOIDOMYCOSIS
(cont.)

X-RAY

- Hilar adenopathy
- Alveolar infiltrate - fleeting
- Pleural effusion
- Cavity
- Pneumothorax/pyo-pneumothorax

spontaneous resolution the rule in 6-8 weeks

Coccidioidomycosis

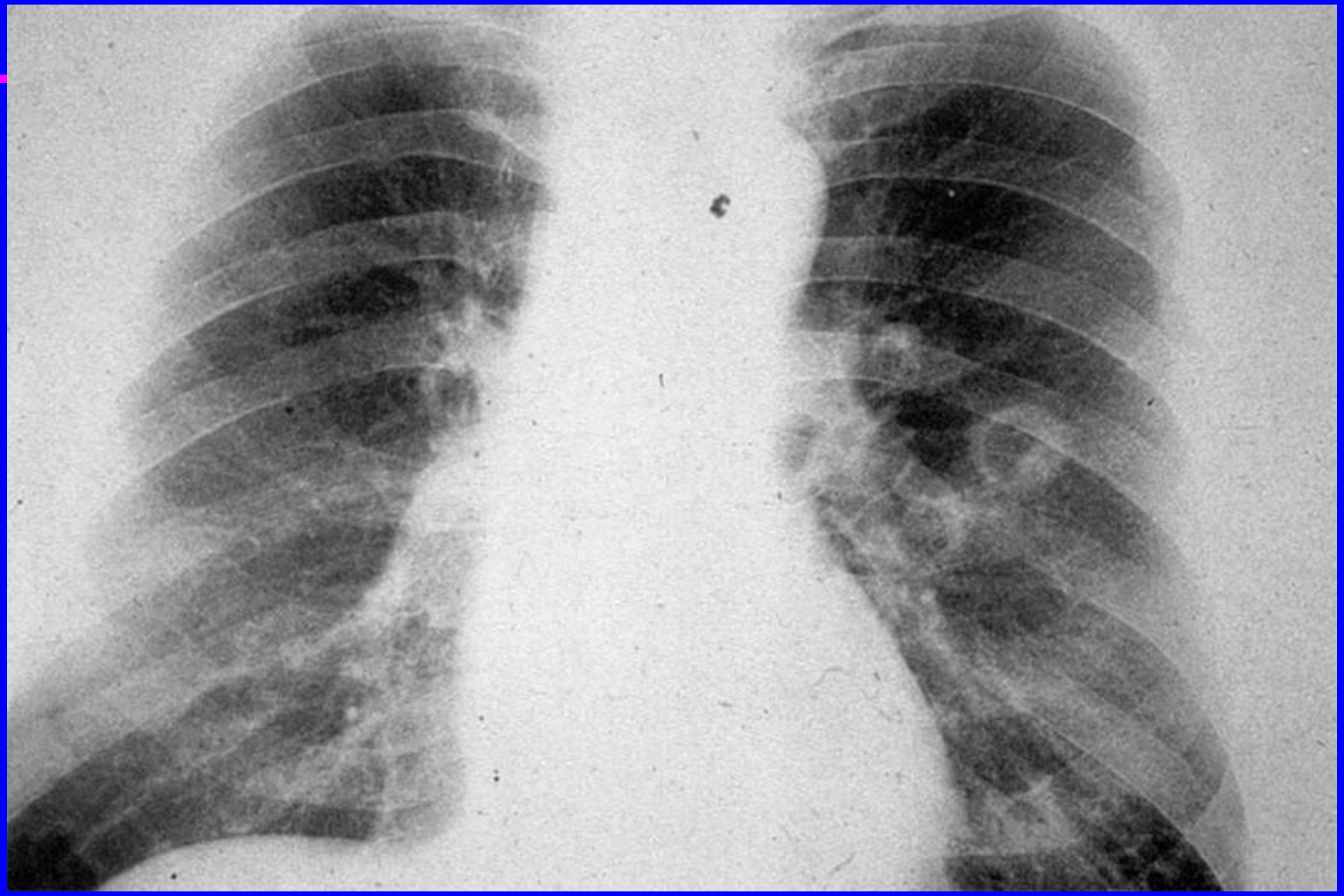
2. Chronic Pulmonary Infection/Acute Progressive 5%

Asymptomatic ⟷ Symptomatic

Asymptomatic:
- nodule
- cavity

Symptomatic:
- chronic pneumonia
- scarring
- cavities
- bronchiectasis
- mycetoma
- hemoptysis
- empyema/B-P fistula

Coccidioidomycosis - chronic



CLINICAL COCCIDIOIDOMYCOSIS
(cont.)

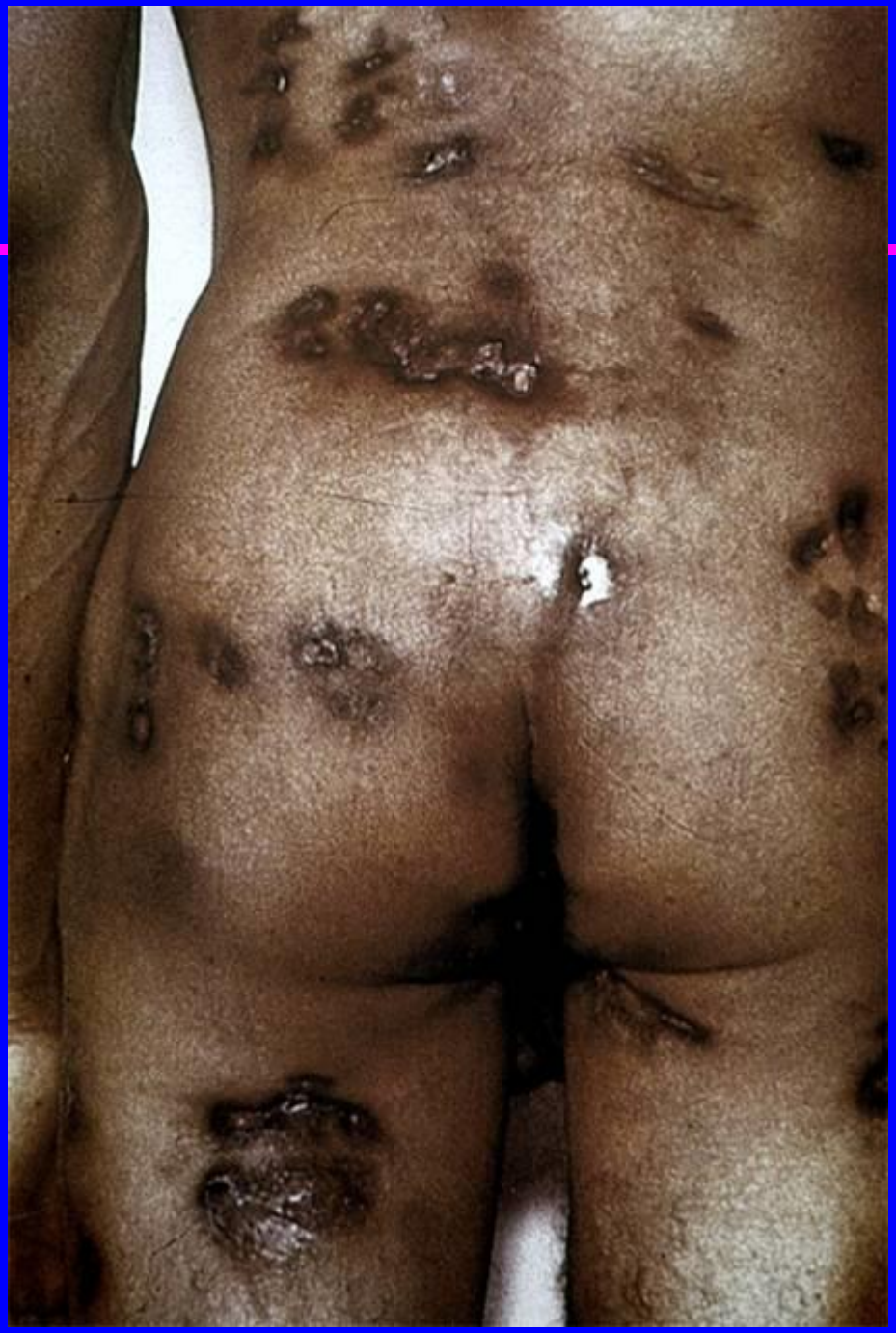
3. Disseminated Infection 0.5%

a) Skin — verrucous granulomas
erythematous plaques
nodules

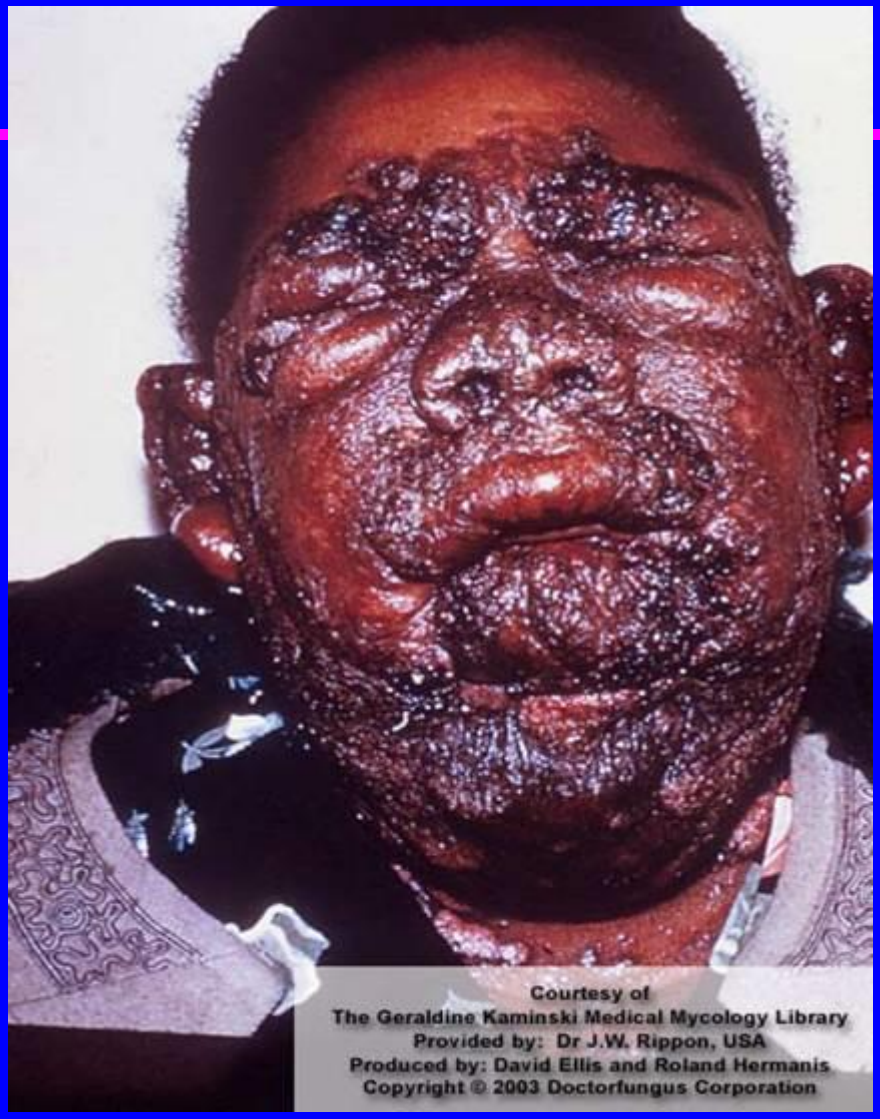
b) Musculoskeletal

Bone — (40% disseminated)
chronic presentation
skull, metatarsals, spine, tibia

Joints — monoarticular
knee, wrist, ankle
subcutaneous, muscle extension

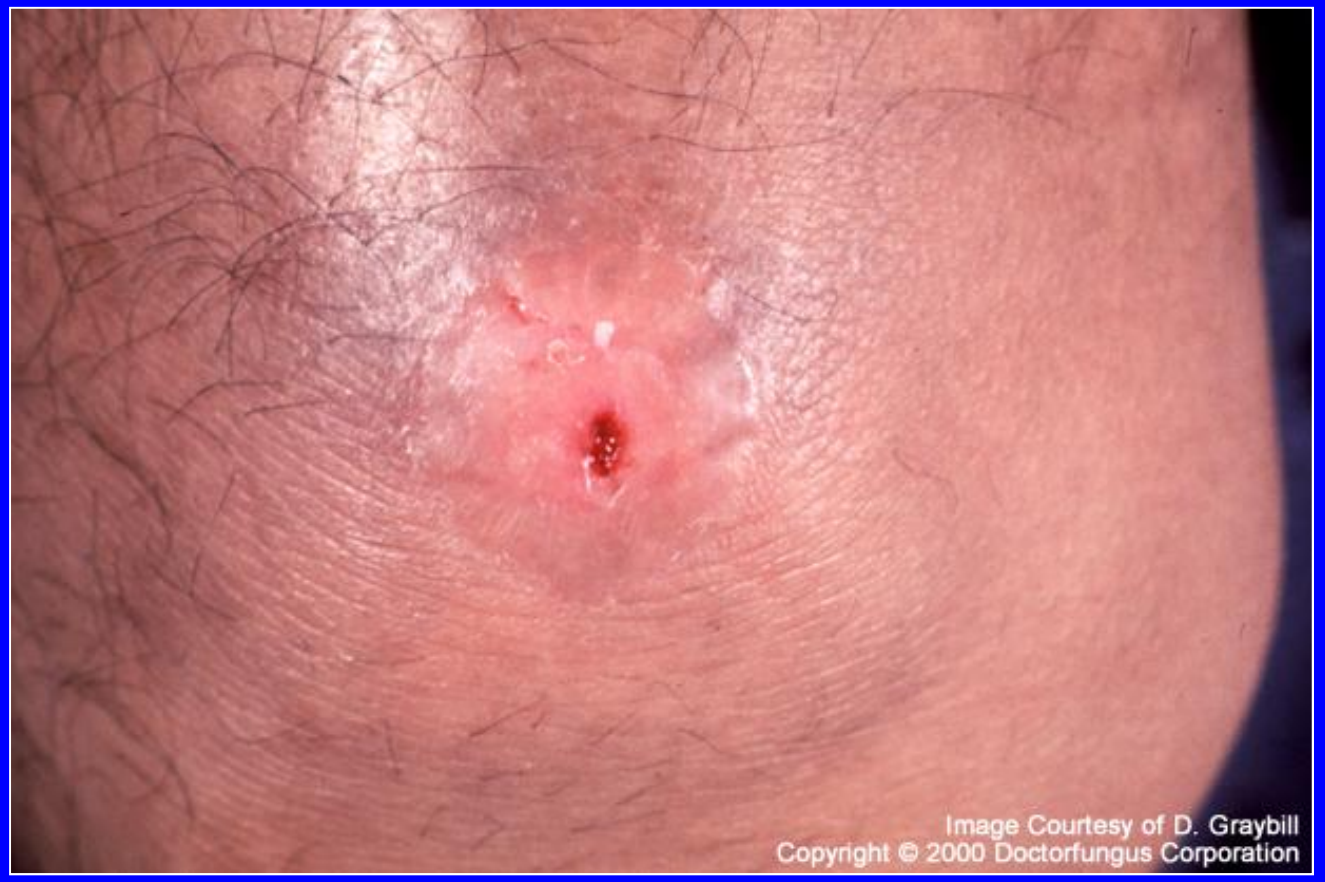


Chronic skin



Chronic granulomatous coccidioidomycosis

Disseminated lesion to knee



CLINICAL COCCIDIOIDOMYCOSIS
(cont.)

3. Disseminated Infection (cont.)

c) <u>CNS</u> presents with 1^0 or up to 6 months afterwards
fatal within 2 years of prognosis
basilar meningitis

subtle, nonspecific presentation
H/A, lethargy, confusion, fever, weight loss, weakness, seizures, behaviour change, ataxia, vomitting, focal deficit

<u>CSF</u>
↑ cells (lymphs) (eosinos)
↑ protein
↓ glucose

CLINICAL COCCIDIOIDOMYCOSIS
(cont.)

3. Disseminated Infection (cont.)

Peripheral eosinophilia		
Serology (CF)	+	
Skin test	-/(+)	
CSF Ab (CF)	+	83%
Cult	+	25%

d) GU system

e) GI - peritonitis

f) Miliary

g) Neonatal - severe

COCCIDIOIDES IMITIS

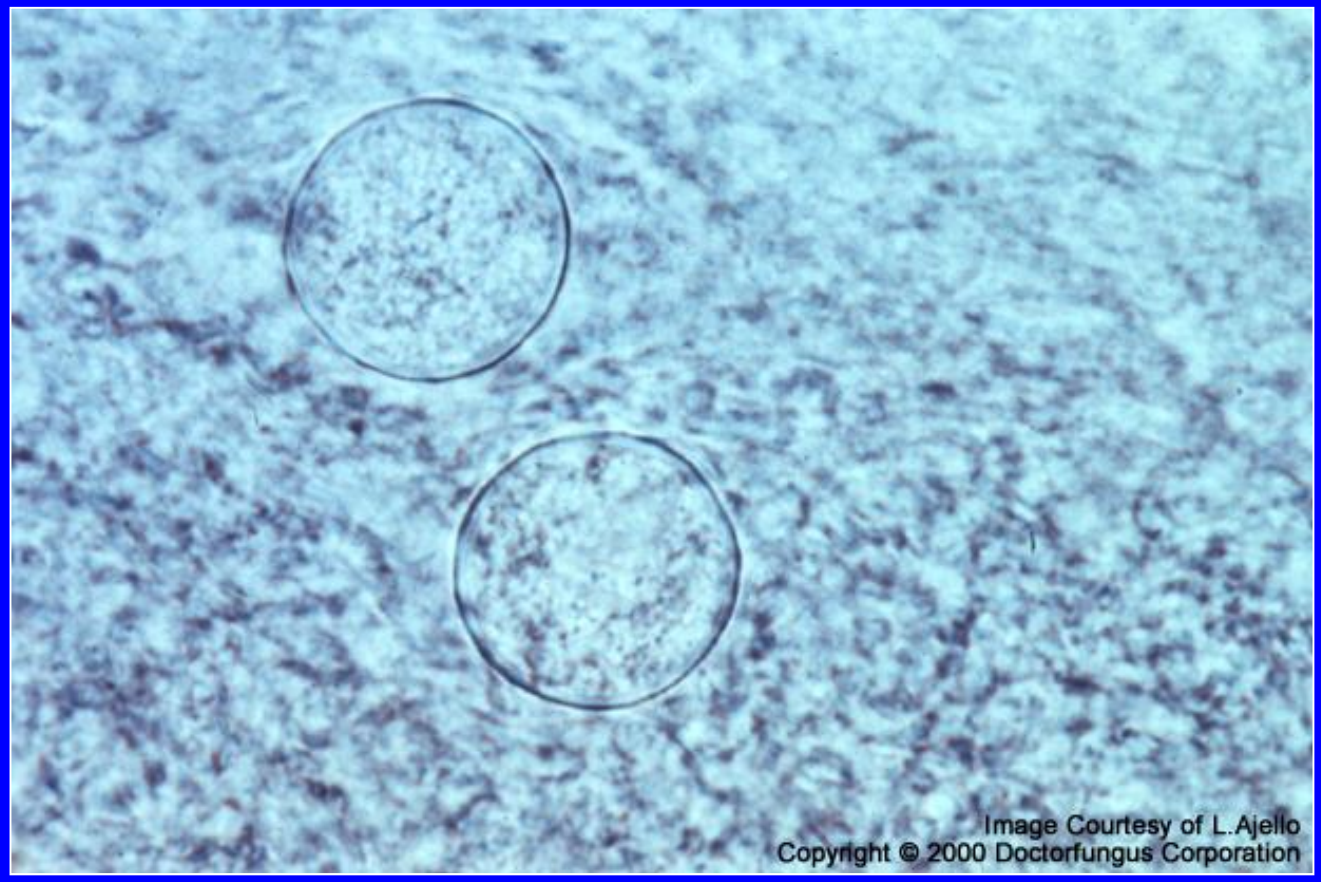
A) Laboratory Diagnosis

1. Direct Examination - sputum
   - tissue biopsy
   - skin
   - CSF

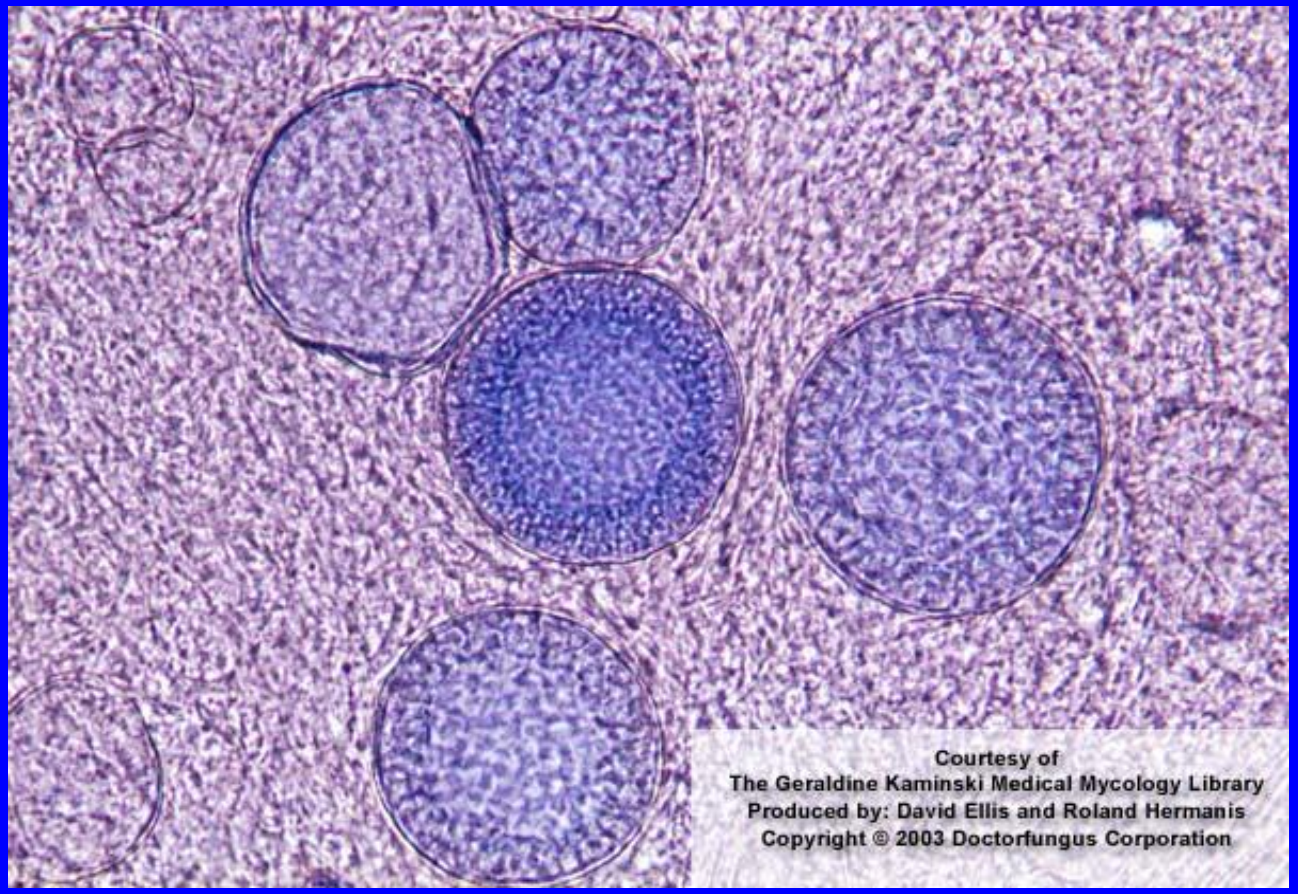
KOH
Calcofluor
Histopathology (GMS, PAS)

Spherule (yeast) form
i. immature spherule (5-30 μ)
*ii. mature spherule (30-100 μ)
iii. endospores (2-5 μ)

KOH, spherules and endospores

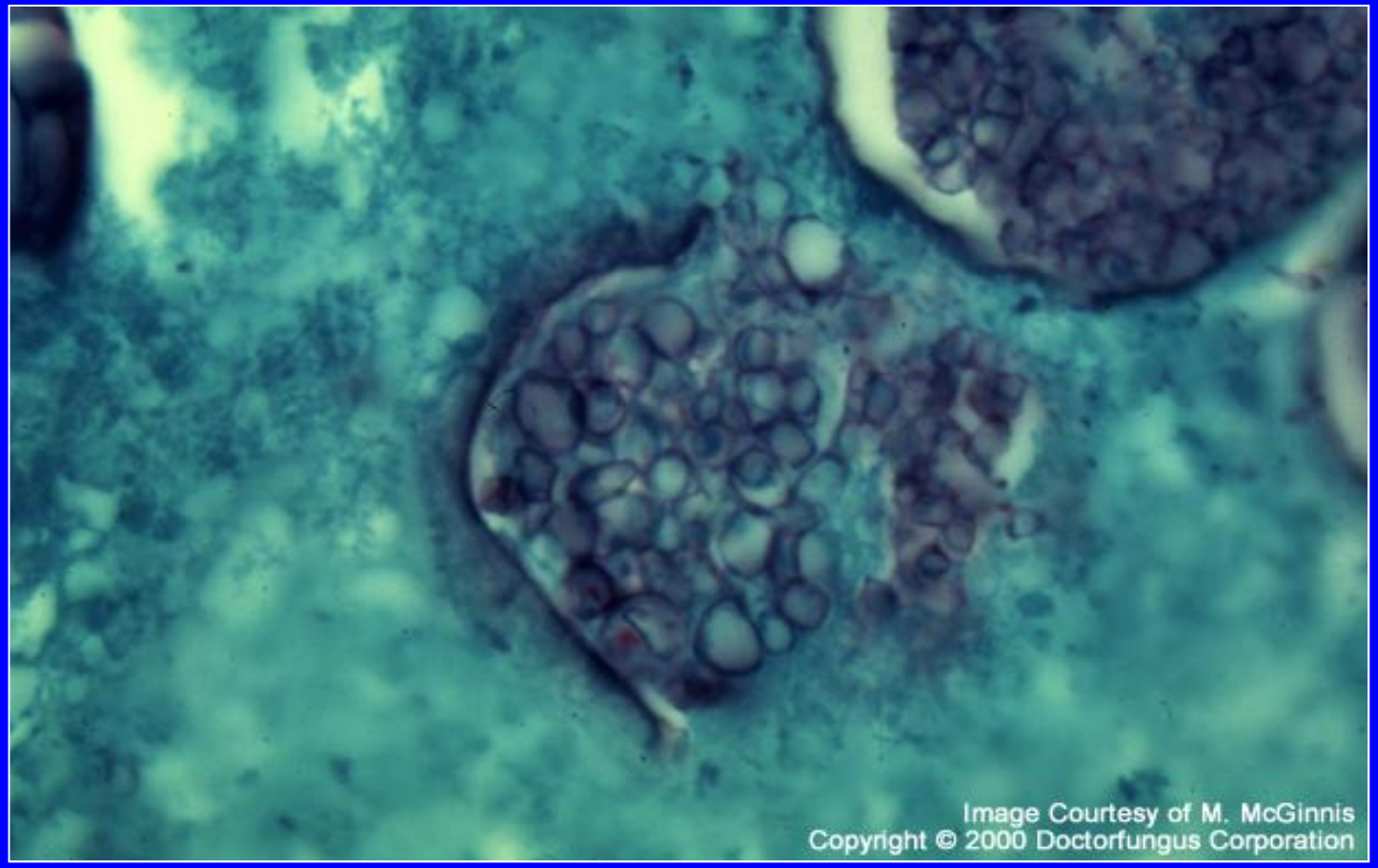


Spherules and endospores



KOH and Parker ink

GMS lung, spherules and endospores



COCCIDIOIDES IMITIS (cont.)

A) Laboratory Diagnosis (cont.)

2. Culture - 30^0C

Mould form (mycelial phase)

while colony, rapid growth

septate hypae (2-4 μ)

arthroconidia - alternating

- barrel-shaped

Macroscopic cocci

Mould cocci

Lactophenol

Mould cocci

Phase contrast

Alternating barrel-shaped arthroconidia

COCCIDIOIDES IMITIS (cont.)

A) Laboratory Diagnosis (cont.)

3. Conversion Test

Mould ⟶ spherule

40^0, special media

or Exo antigen test

Molecular probe

4. Serology

IgM available

IgG - prognosis

- monitoring treatment (CSF level)

COCCIDIOIDES IMITIS (cont.)

B) Other Diagnostic Tests

1. CXR

2. Skin Test

"coccidioidin"	"spherulin"
mycelial phase Ag	spherule Ag

*primary infection - positive by 4-6 weeks

* disseminated infection - may be negative

COCCIDIOIDES IMITIS (cont.)

Treatment

< 5% of patients need treatment

- severe 1^0 pulmonary
- ↑ CF titer > 1:16 - 31
- worsening clinical status at 6 weeks
- immunocompromised patients
- disseminated infection

COCCIDIOIDES IMITIS (cont.)

Treatment (cont.)

Amphotericin B
Azoles
Fluconazole
Itraconazole
Non-meningeal disease
FLU/ITRA

Meningitis
- Fluconazole
- AMB (IT) ± azole
± IV AMB