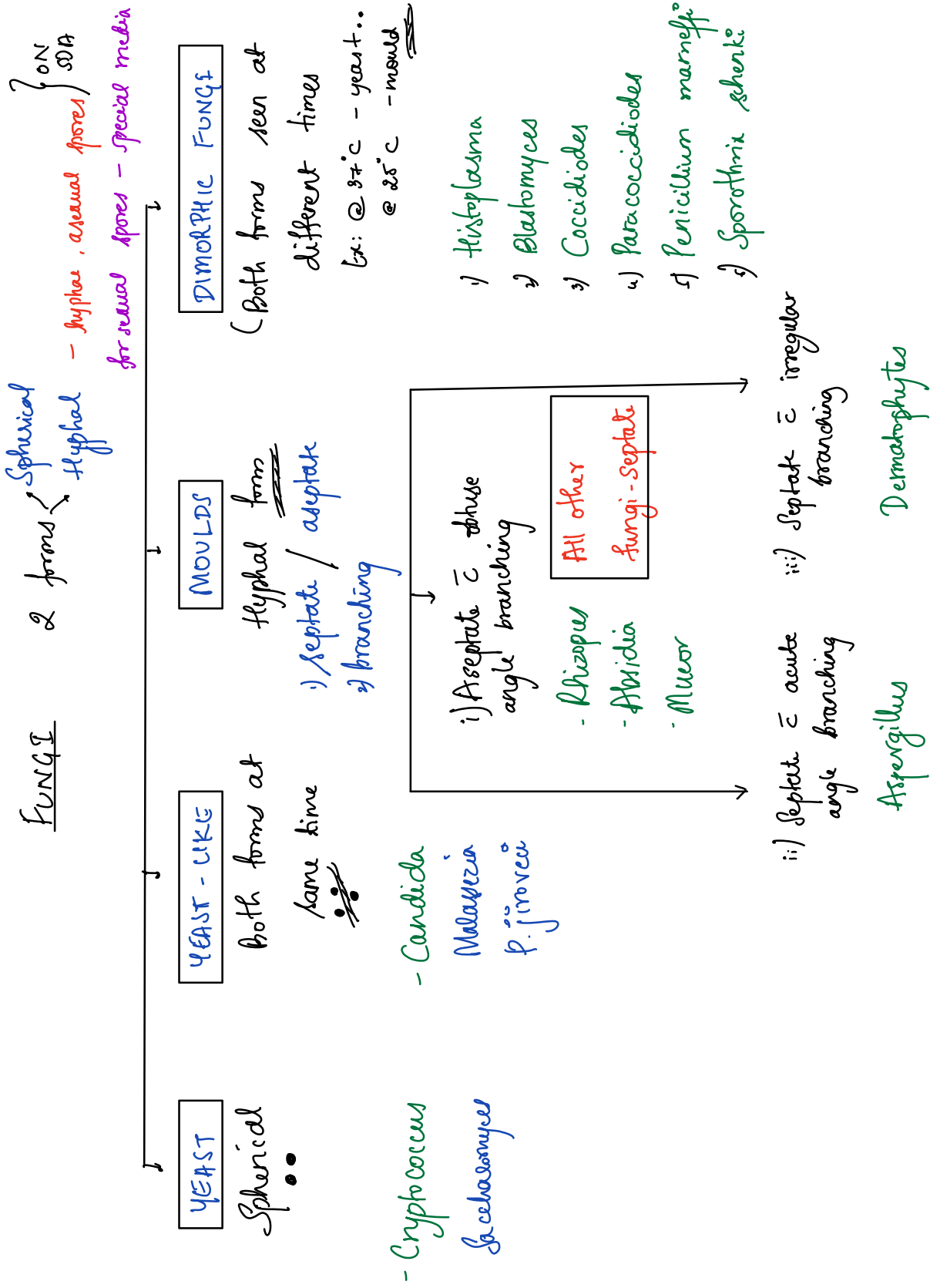
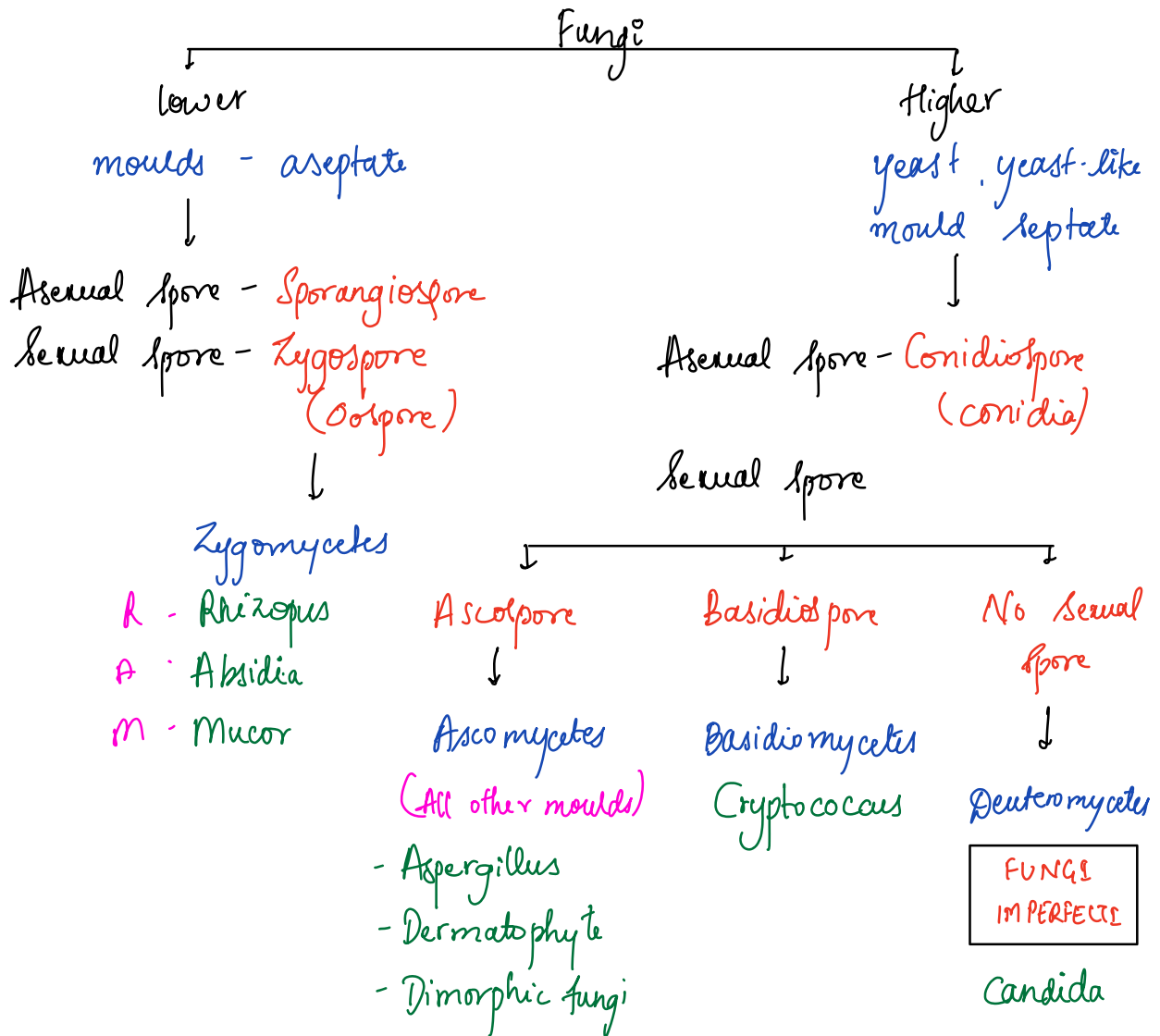






Sexual Spore	Asexual Spore
- Zygosporangium - Ascospore - Basidiospore	- Sporangiospore - lower : RAM - Conidiospore - All higher fungi - Chlamydospore ← candida albicans candida dublinensis - Arthrospore - Ectothrix & endothrix - Blastospore - yeast cells

As & Base } Sexual

Fungal diseases (Mycosis)

1) Superficial mycosis
(epidermis)

- Tinea versicolor / pityriasis versicolor
- Piedra
- Dermatophytoses

2) Deep mycosis
(subcutaneous tissue)

- Madura mycosis
- Sporotrichosis
- Chromoblastomycosis
- Phaeohyphomycosis

3) Systemic
(endemic)

- Histoplasmosis
- Blastomycosis
- Coccidiomycosis
- Paracoccidiomycosis

4) Opportunistic

- Candidiasis
- Cryptococcosis
- Aspergillosis
- Mucormycosis
- Pneumocystosis

Lab diagnosis

i) Microscopy

i) KOH mount

yeast / hyphae seen

KOH - clearing agent

dissolves proteinaceous material

ii) Staining :

- PAS Periodic Acid Schiff
- GMS Gomori Methanamine Silver
- CFW Calcofluor White - Fluorescent stain

} Histopathological stains

Fungi cell wall — chitin
↑
stain

iii) Culture

- SDA - Sabouraud's Dextrose Agar
- PDA - Potato Dextrose Agar
- CMA - Cornmeal Agar
- Bird Seed Agar

Non cultivable fungi

		Earlier	Later	Now
1) PNEUMOCYSTIS	JEKOVECI / CARINEL	Protozoa	Fungi	Fungi
"		Trophocyst	∴ chitin in cell wall	
		Cyst		
2) RHINOSPORIDIUM	SEEBERS	Earlier aquatic fungi	later aquatic protozoa	Meso mycetozoa

iv) Identification after culture

- yeast / yeast like — Gram stain
- moulds — LPCB mount

(Lactophenol Cotton Blue)
hyphae & asexual spores
↓

Slide culture technique (best)
morphology better seen

TINEA VERSICOLOR / PITYRIASIS VERSICOLOR

Cfr : hypo / hyper pigmented patches (neck & trunk)

Causative agent : *Malassezia furfur* (yeast like)

* difficult to cultivate

(supplement = 1% olive oil to cultivate)

∴ LIPOPHILIC FUNGI

Δ^{fu} : Microscopy -



budding yeast cells
septate hyphae

* SPAGHETTI & MEATBALL APPEARANCE

Culture - Fried egg colonies



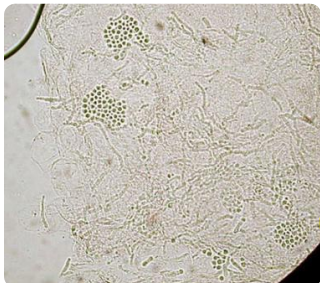
Wood's lamp -
(UV source)

Golden yellow colour

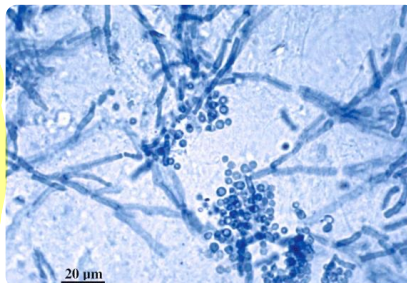
fluorescent - pteridine pigment

Rx: Topical azoles & terbinafine x 2wks
(resistant to griseofulvin)

mc fungi to cause Seborrheic dermatitis (dandruff) - *Malassezia furfur*



Spaghetti & meatball appearance



Wood lamps : Golden yellow fluorescence

PIEDRA

C/F : Nodule formation on hairshaft

Cautative :
agent

white piedra

Trichosporon
beijii

Black piedra

Piedra hortae

Dis : Microscopy



septate hyphae +
arthrospore



septate hyphae +
arthrospore

Culture : white color
colonies

Brown-black
colonies

pubic hair, axilla
facial hair

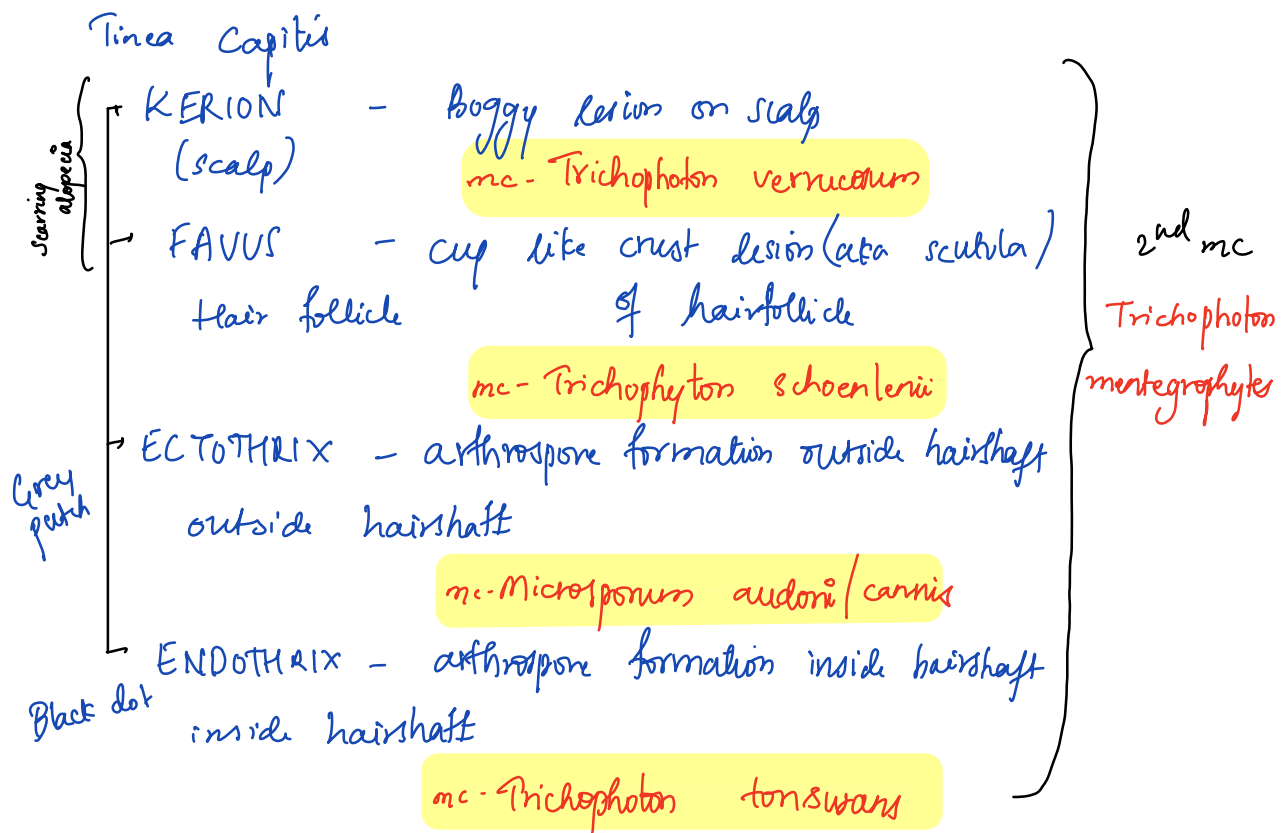
scalp

DERMATOPHYTOSSES (Tinea infection)

↳ infect & survive only in dead keratinocytes
stratum corneum

C/F: skin scaly, itchy, circular rash
hair
nails

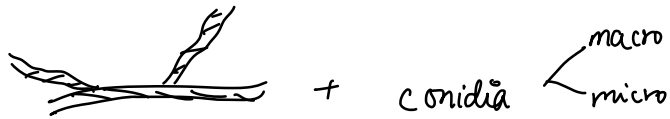
Causative agent:	Trichophyton	skin, nail, hair	All 3 inv
	Microsporum	skin, hair	N not affected
	Epidermophyton	skin, nail	H not affected



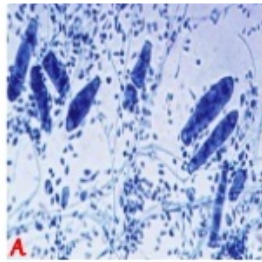
all other tinea infection → Trichophyton rubrum (mc)

Δ^{no} :

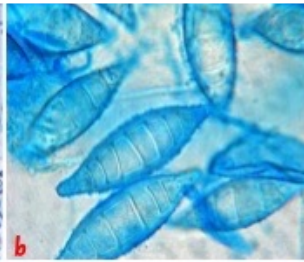
1) Microscopy



	T	M	E
macro conidia	 pencil / cigar shaped	 spindle shaped	 club shaped
microconidia	abundant	few	absent



A. *Trichophyton* Species,



B. *Microsporum* Species,



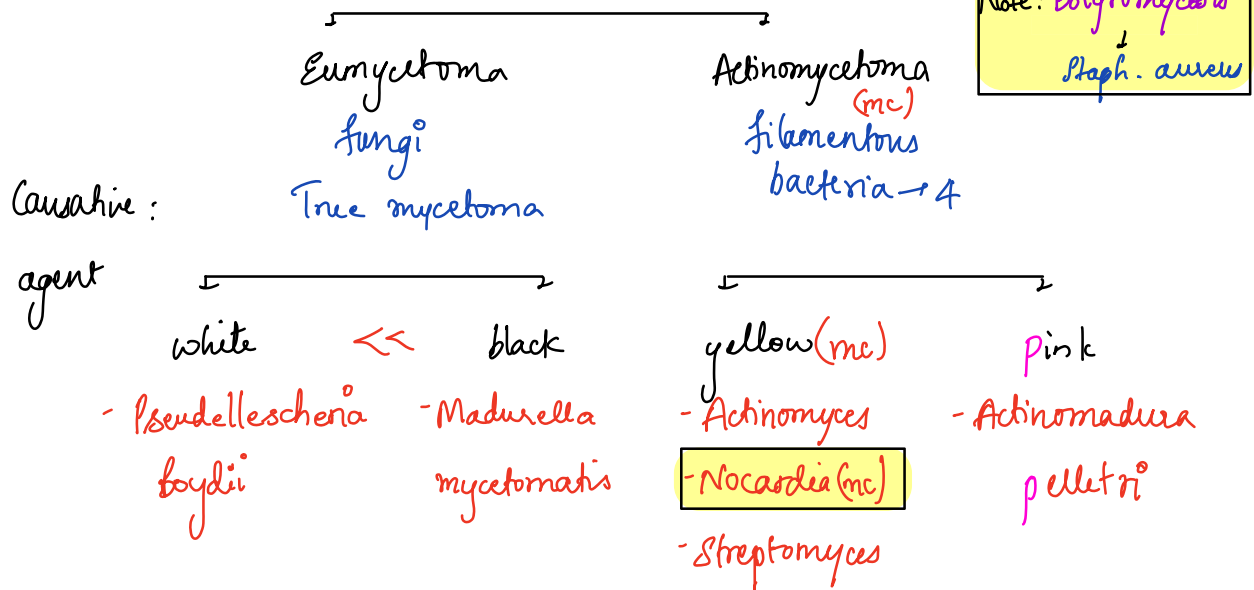
C. *Epidermophyton* Species

2) Culture - white cottony growth

3) Wood's lamp - Apple green fluorescence
(only few)

MADURA MYCOSIS / MADURA FOOT / MYCETOMA

C/F : s/c swelling → sinus → discharge of granules



Note: Botryomycosis
↓
Staph. aureus

Asia:  + asexual spore


Gram positive filamentous bacilli



SPOROTRICHOSIS (Rose gardener's disease)
↳ thorn prick

mc in sub Himalayan region

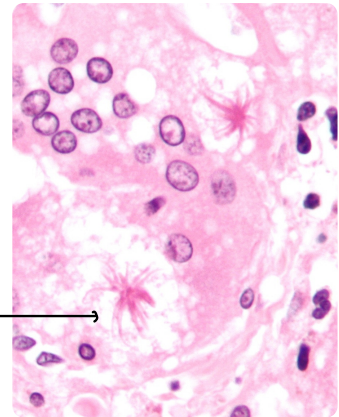
C/F: 1/c nodules along lymphatics



Causative agent: *Sporothrix schenckii*

Di:

1) Microscopy - asteroid bodies



2) Culture - Flower like pattern

**

Sporotrichoid like pattern → Nocardia (mc)
Seen in
i.e. nodules along lymphatics

CHROMOBLASTOMYCOSIS

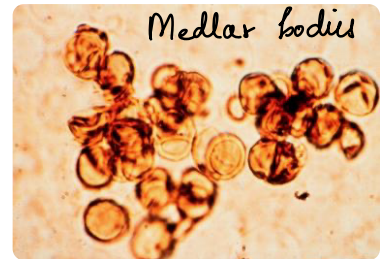
C/F: s/c nodules , ulcer , verrucose lesions
(mc)

Causative agent: Pheoid / darkly pigmented fungi (brown-black)
= sclerotic bodies

Δ^{ru}: 1) Microscopy : sclerotic bodies
aka Medlar bodies

round brown golden yellow color

2) culture : Brown black pigment



PHEOHYPOMYCOSIS

C/F: s/c nodules , ulcer , verrucose lesions
(mc)

Causative agent: Pheoid / darkly pigmented fungi (brown-black)
= out sclerotic bodies

Δ^{ru}: 1) Microscopy : No sclerotic bodies

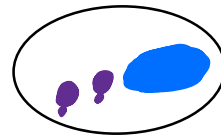
2) culture : Brown black pigment

SYSTEMIC INFECTIONS

C/F: cutaneous, lung manifestations more common
others

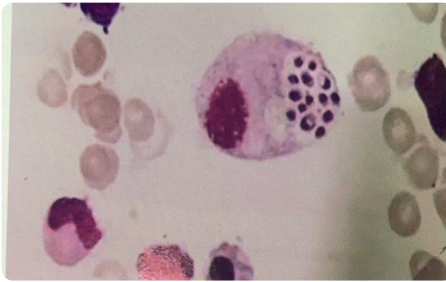
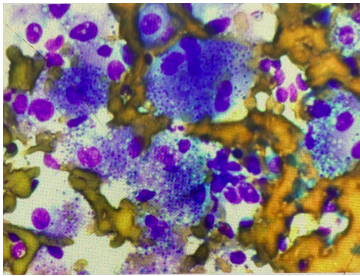
i) Histoplasmosis / Darling's disease
involve RES (mac)

Δ^{su} : Microscopy (sample) - yeast form
(37°C)



Narrow base budding

always intracellular
= in macrophage



culture (SDA)
(25°C) · mould form



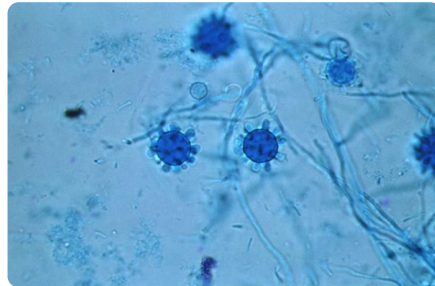
septate hyphae +



tuberculate

conidia

asexual spore



2) Blastomycosis

Δ^{SH} : Microscopy (Sample) - yeast form
(37°C)

● ●
Broad based
budding

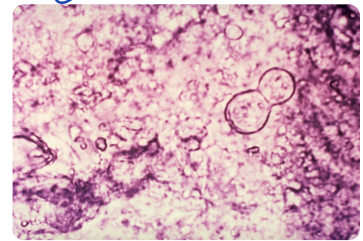
FIGURE OF
8 APPEARANCE

culture (SDA)
(25°C) · mould form



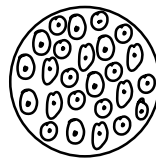
septate hyphae ±

pear shaped conidia



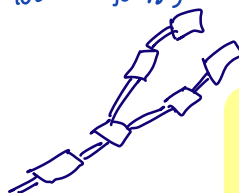
3) Coccidiomycosis

Δ^{SH} : Microscopy (Sample) - yeast form
(37°C)

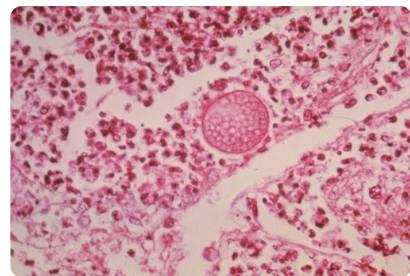


Spherules
containing endospores → (also seen in
Rhizopus
seeberi)

culture (SDA)
(25°C) · mould form

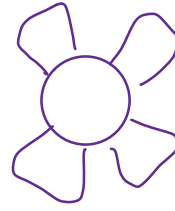


Fragmented hyphae
± barrel shaped conidia

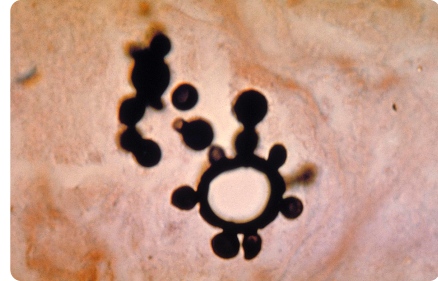


4) Paracoccidioidomycosis

Δ^{SN} : Microscopy (Sample) - yeast form
(37°C)



MICKEY MOUSE/
PILOT WHEEL
APPEARANCE



Culture (SDA) · mould form
(25°C)



septate hyphae ±

pear shaped conidia

OPPORTUNISTIC INFECTIONS

CANDIDIASIS

CF: cutaneous, mucocutaneous, systemic (Invasive fungal infection)

Causative:

Agent

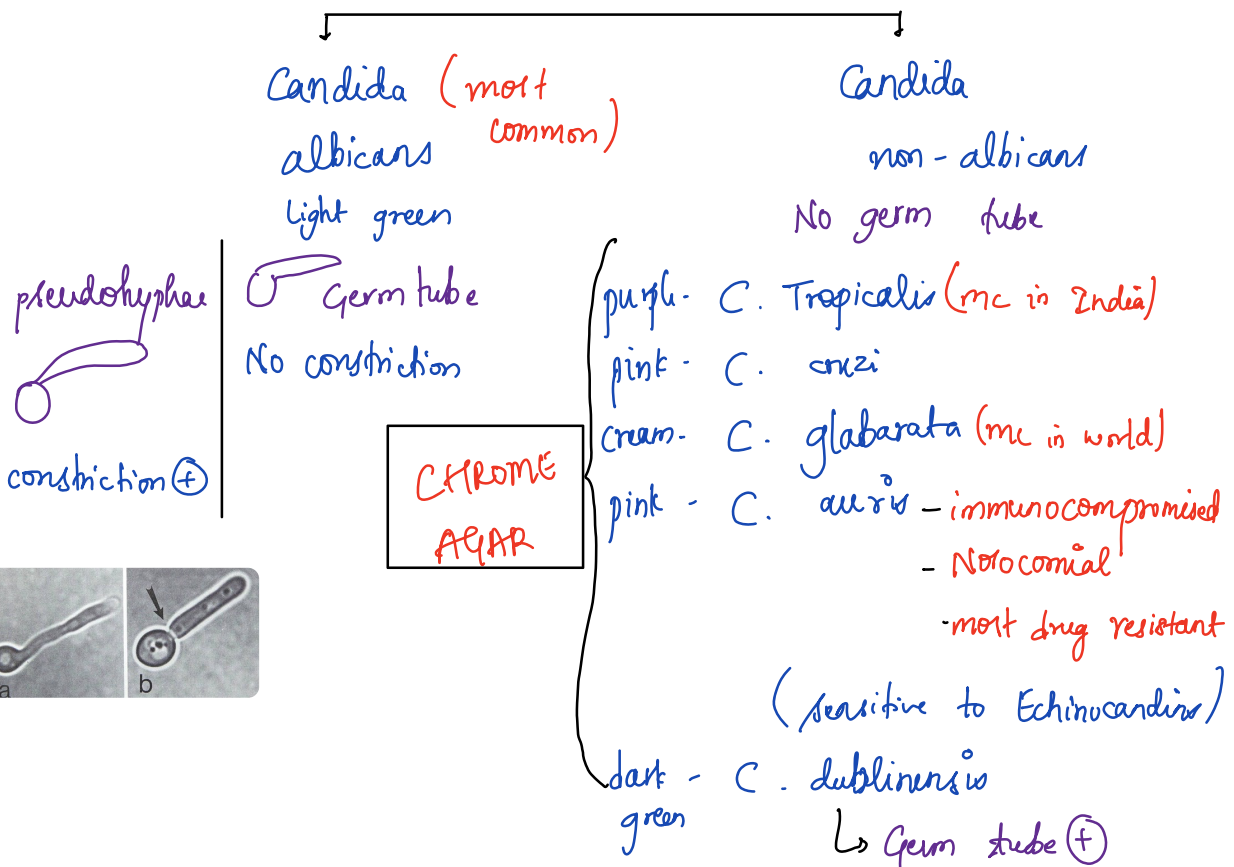
Candida

to differentiate:

Germ tube test

@ 37°C 90 mins
(colony + serum)

aka Reynolds's braude phenomenon



<i>C. albicans</i>	<i>C. dublinensis</i>
- budding yeast cells - chlamydospores - germ tube test ⊕	
@ 42°C on chrome agar :	@ 42°C no growth dark green
growth ⊕ light green 	

Δ^{SN} : Microscopy -  oval budding yeast cells
 ⊕
 pseudohyphae (constriction ⊕)
 (elongated budding yeast cells)
 culture - chrome agar

mc fungal infection in immunocompetent
 mc fungal infection in immunocompromised
 mc fungal infection in neutropenic pts

} CANDIDA

✱

Invasive candidiasis marker

BDG - β -D Glucan

seen in .candida

.aspergillus

.pneumocystis
jieroveci

C. gatti

immuno competent &
immuno compromised

Eucalyptus tree

Eucalyptus tree

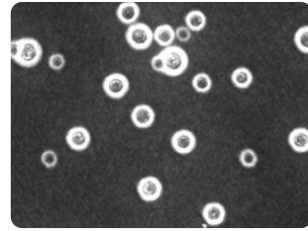
does not produce blue color colonies ← **CGB agar** → produces blue color colonies
(Canavanine Glycine Bromothymol Agar)

Δ_{sis} : Microscopy Gram :  - round, budding yeast cells
(bigger than candida)

Mucicarmine stain
(everything stained except target) Negative stain : to demonstrate capsule

urease +ve

- India Ink
- Nigrosin



Culture - cream colour
yeasty growth

Immunological - Rapid test : Latex Agglutination Test
(for all capsulated organisms + CRP)

mc CNS fungal infection in immunocompromised } CRYPTOCOCCUS
mc fungal infection in post transplant patient }

No BDG in sample $\therefore$ capsule prevents release

mc fungi causing brain abscess - *Cladophialophora* (Chromoblastomycosis)
barteliana

ASPERGILLOSIS

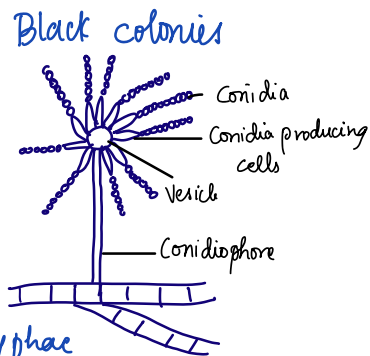
Septate hyphae =
acute branching

Dichotomous - ends in two
branching

C/F: Ear, PNS, Respiratory system

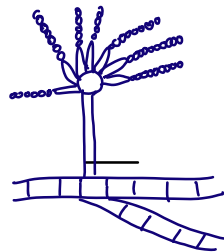
Causative Agent	<u>A. Niger</u> (Ear)	<u>A. flavus</u> (PNS > Resp) mc in India	<u>A. fumigatus</u> (Resp > PNS) mc in world
	Black colonies	Yellow colonies	Green colonies

Δ^{SW}:

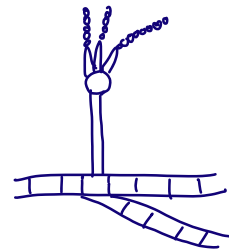


septate hyphae
= acute branching

vesicle completely surrounded
by conidia



2/3rd of vesicle
has conidia



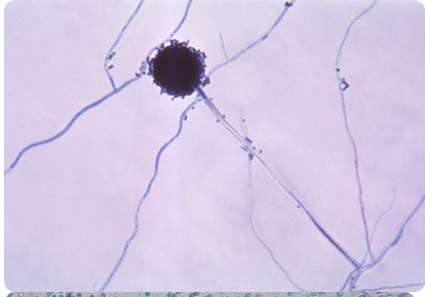
1/3rd of vesicle
has conidia

Aflatoxin
(mycotoxin)

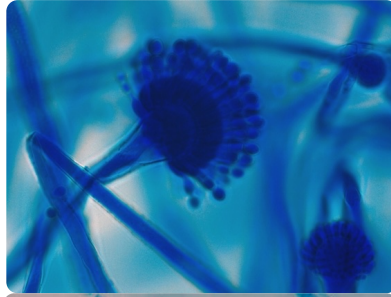
ABPA
Δ^{SW} by IgG

Aspergilloma

Invasive aspergillosis marker : Galactomannan



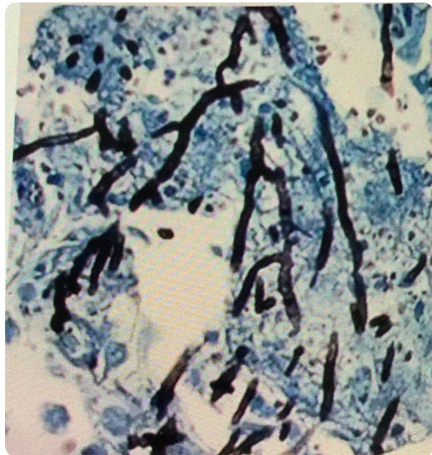
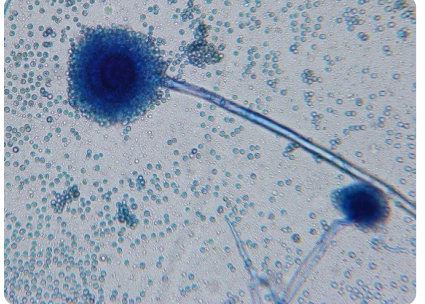
A. Niger



A. flavus



A. fumigatus



Dichotomous branching
 $\therefore$ Aspergillus



Mucormycosis

Diabetic, DKA, Iron therapy (∴ growth factor)
ANGIOINVASION

C/F : mc site: **Rhinocerebral**

LID LIFTERS

Causative agent

Rhizopus
(mc)

Absidia

Mucor

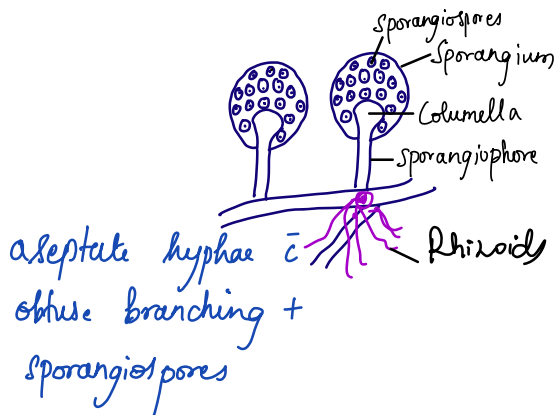
Asia :

white growth
 ∴ black in b/w

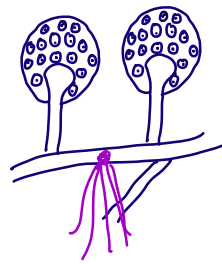
white
 growth

white
 growth

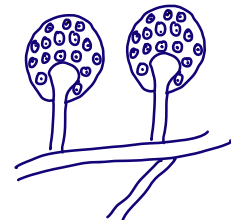
**SALT & PEPPER
 APPEARANCE**



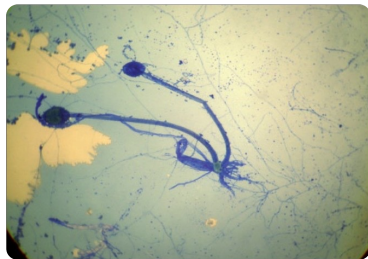
Nodal rhizoids



Internodal rhizoids

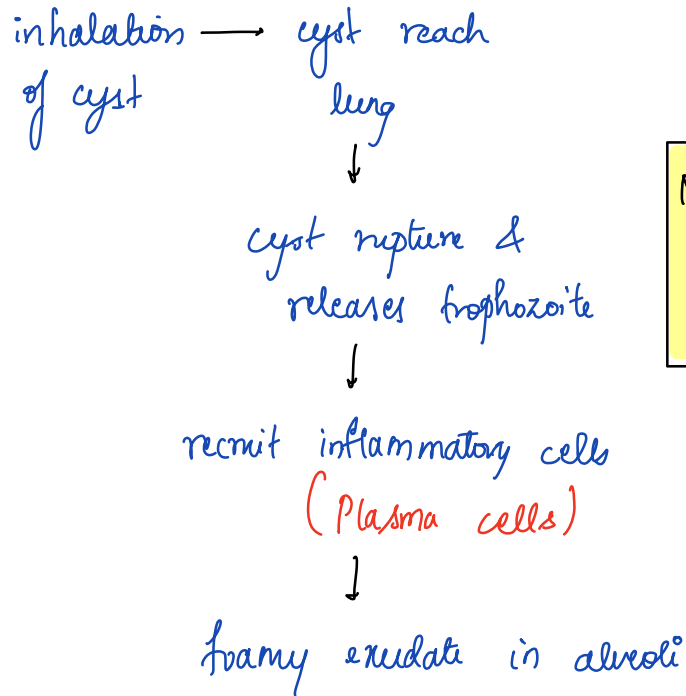


No rhizoids



PNEUMOCYSTOSIS PCP - PNEUMOCYSTIS CARINII PNEUMONIA
Plasma cell pneumonia

C/F : Pneumonia



Note: ↑ risk of *Pneumocystis jirovecii* in Hyper IgM syndrome

Causative agent: *Pneumocystis jirovecii* (carinii)

- Diagnosis :
- 1) Sputum - very low sensitivity
 - 2) BAL - mc used
 - 3) percutaneous lung Bx
 - 4) Open lung Bx - best sample

@ risk individuals : - HIV
- premature infants
- immunosuppressed

BAL → Direct IFA → CFH
Calcofluor white

✓

PCR

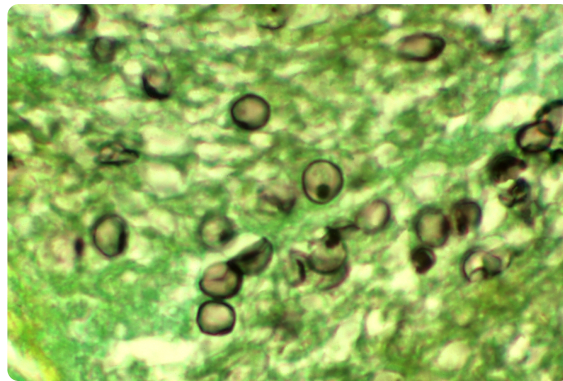
✓

GMS stain:



cyst

PING PONG BALL
APPEARANCE



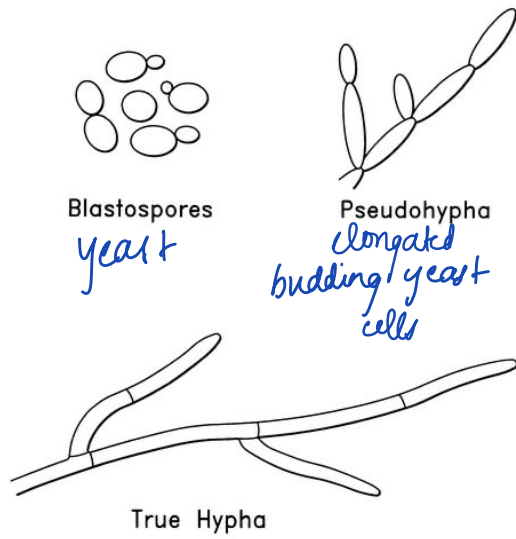
Rx: DOC: Cotrimoxazole

Glucocorticoids if $PaO_2 < 70$ mmHg

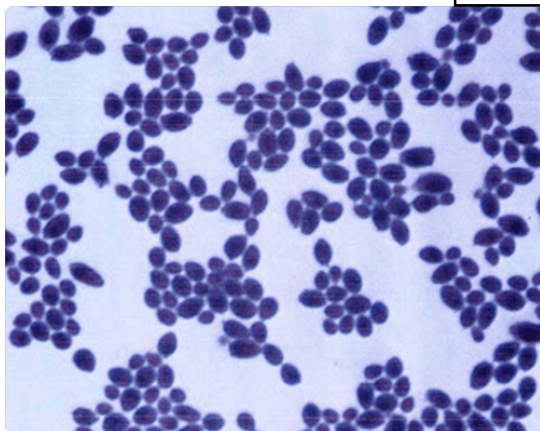
Prophylaxis:

aerosolized pentamidine

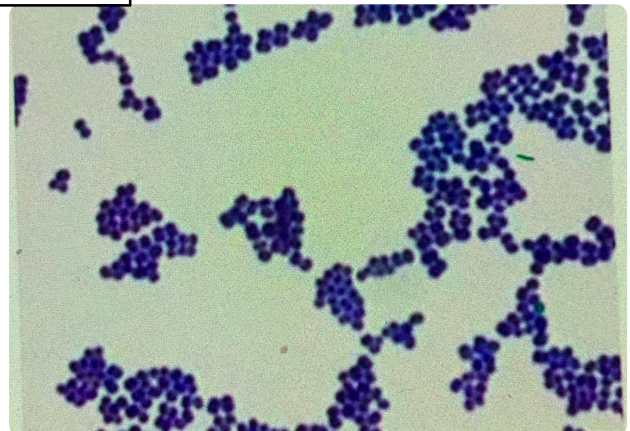
Indications: $CD4 + T_{cell} < 250/\mu L$
recent oropharyngeal candidiasis
prior PCP infection



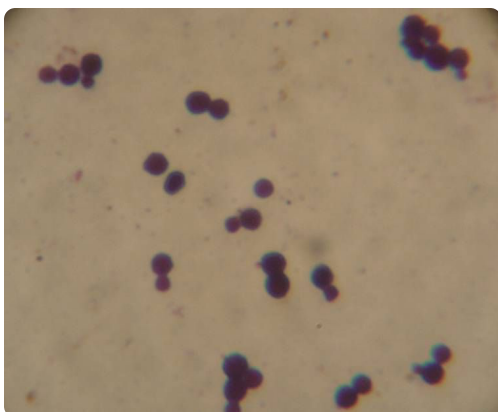
CANDIDA



oval, small size
many in number

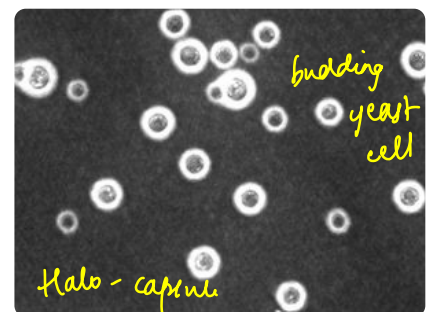


- small size, many in number



CRYPTOCOCCUS

round
bigger in size
less in no



MUCORMYCOSIS (Black fungus)

Mortality : 40 - 80% Avg 54% (CDC)

Phylum - Mucormycota

Order - Mucorales

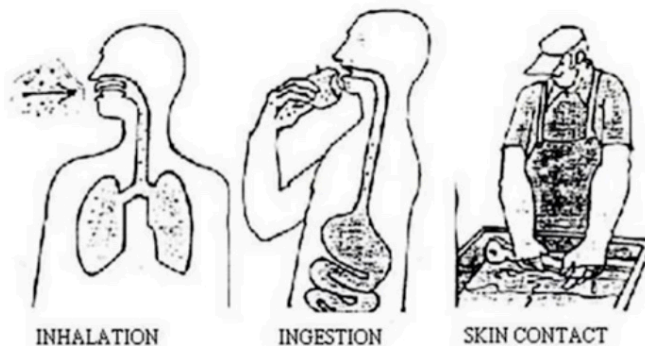
Group - Mucormycetes

Fungi: - *Rhizopus* (mc) - *Rhizomucor* - *Apophysomyces*
- *Absidia* - *Laksanaea* - *Coeteromyces*
- *Mucor* - *Syncephalastrum* - *Cunninghamella*

moulds $\begin{cases} \nearrow \text{hyphal} \\ \searrow \text{spore} \end{cases}$

Habitat : soil & decaying vegetative matter

Modes of infection :
Inhalation of spores
Ingestion of spores
Direct contact of injured skin $\pm$ spores



Risk factors

- Uncontrolled DM / DKA

↳ ∴ macrophages are dysfunctional → ∴ impaired Glutathione pathway

but Rhizopus has ketone reductase system

∴ overcomes ketoacidosis & survives

- Iron chelators (Deferriamine)

↓
utilize it as siderophore (iron carrier)

- Steroids

- Solid organ / BM transplant

- Hematological malignancies

} ↓ immunity

- COVID 19 in diabetes

→ ↓ immunity

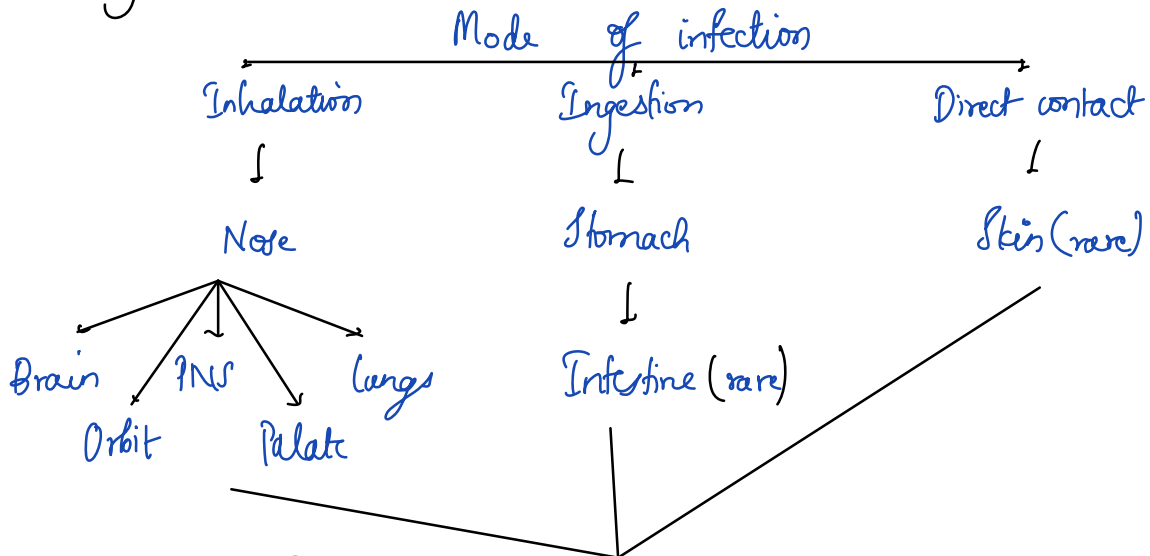
↓
Steroids

↓
↑ sugar

- Trauma & burns

cutaneous mucormycosis

Pathogenesis :



Normally : Phagocytosis by macrophages & oxidative burst by neutrophils

RISK FACTORS PRESENT

Spores germinate into hyphal forms

Inflammation

Clinical manifestation

Angioinvasion

Thrombosis

Ischemia

Necrosis (Black crust)

Types: (Clinical manifestation)

- Rhinocerebral
 - Rhino orbitocerebral
 - Pulmonary
 - Gastrointestinal
 - Cutaneous
 - Disseminated (blood)
- } inhalation
- ingestion
- Direct contact

Clinical features:

Nose, PNS, palate: Nasal congestion, blockade, black crusting
PNS pain, swelling, paraesthesia

Brain : headache, vomiting, seizures

Orbit : diplopia, ptosis, periorbital cellulitis
Abrupt vision loss

Lungs : Chest pain, dyspnoea, haemoptysis

Stomach & intestine : ulcers, haemorrhage

Skin : papules, pustules (chainy pus), black crusting



Diagnosis :

1. Sample collection

- tissue biopsy (best sample)
- Deep nasal swab / nasal secretions

Note: Do not grind/homogenize the sample

∵ hyphae are very delicate

may get damaged → No growth on culture

2. Sample microscopy

- KOH mount

- HPE

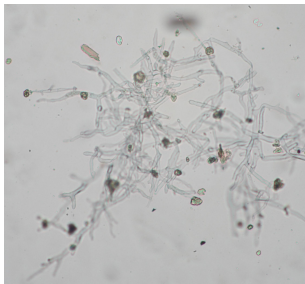
· PAS stain

· GMS stain

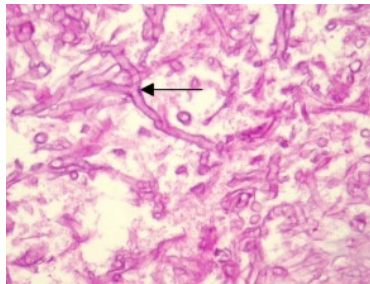
- Fluorescent staining

· Calcofluor white stain

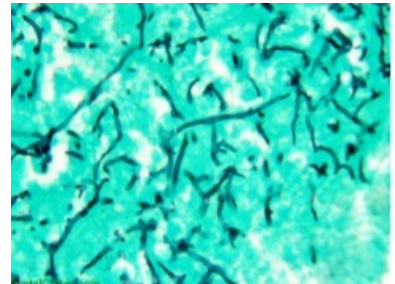
Broad aseptate obtuse angle
branching hyphae
(ribbon like appearance)



KOH



PAS



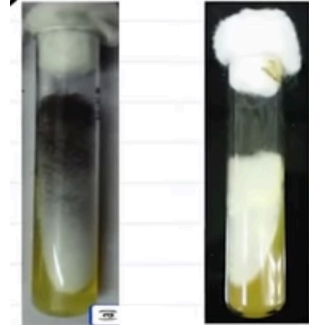
GMS

3. Culture - SDA

- Both vegetative & aerial growth
- cotton candy like growth
- initially - white colour
- later - grey colour

Rhizopus : salt & pepper appearance

LID LIFTERS



4. Culture microscopy

LPCB - Lactophenol cotton blue

- broad aseptate obtuse angle branching hyphae
- sporangiophore ends in sporangium containing **sporangiospores** (asexual)
- Rhizoids

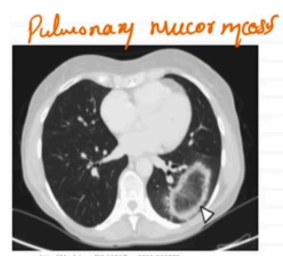
Note: Sexual spores are produced only on special media
Zygospores

5. Molecular PCR (detect specific gene)

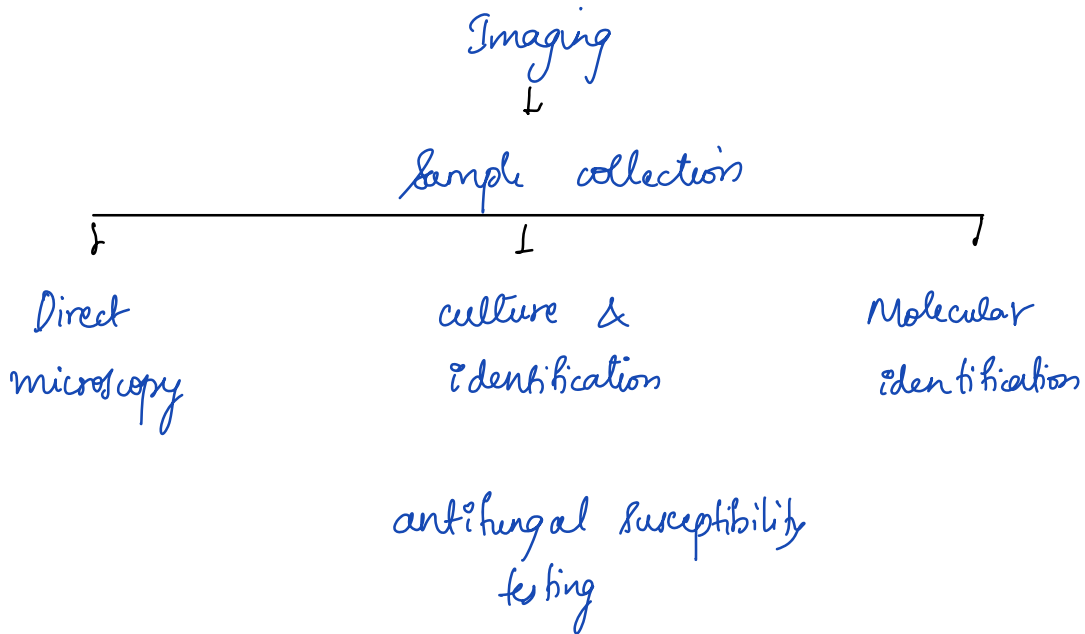
c. Imaging

CT/MRI - to see extent of spread

Reverse halo sign



ECMM guidelines for diagnosis
(European confederation of medical mycology)



Treatment -

- i) Surgical debridement
- ii) Medical

Liposomal Amp B
5-10mg/kg/day
4-6 wks
Renal toxicity

> Itraconazole >
200 mg TID - 2d
200 mg OD - 3rd day onwards
4-6 months

> Posaconazole
300 mg BD - 1d
300 mg OD - 2nd day onwards
4-6 months

ECMM Guidelines for treatment. (Surgical debridement)

Pre-existing
Renal Compromise

Absent

Liposomal
Amp-B
(i.v.)

Any type

Amp-B
Lipid complex
(i.v.)

Any type - without
CNS involvement

Amp-B
lecitholate
(i.v.)

Avoid

Present

Ivavuconazole
i.v.
(or)

Posaconazole
i.v.

Treatment Response
Assessment
(Weekly imaging)

Good response

Shift to Oral
ivavuconazole
(or)
Posaconazole

Stable disease

Continue amp-B
(or)
change it to i.v.
ivavuconazole
(or)
Posaconazole

Progressive disease

Shift to i.v.
ivavuconazole
(or)
Posaconazole

Toxicity (Nephrotoxicity)

Shift to i.v.
ivavuconazole
(or)
Posaconazole

Other Treatment modalities

→ The usefulness of these interventions are unclear

a) Hyperbaric Oxygen

- improves neutrophil function
- inhibit mucormycetes
- improves wound healing

b) CSF → (Colony Stimulating Factors)

- enhance the immune responses

Prophylaxis →

a) Immunoprophylaxis

→ No Vaccine

b) Chemoprophylaxis → Posaconazole oral

1° → hematological malignancies
± neutropenia

2° → Immunocompromised patients
± previous mucormycosis

c) Preventive measures -

- Strict control of diabetes
 - Avoid coming in contact
± soil & decaying matter
 - Wear/stop corticosteroids &
immunosuppressive drugs
 - Treat neutropenia
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