وزارة التعليم العالي والبحث العلمي
الجامعة التقنية الجنوبية
المعهد التقني العمارة
قسم تقنيات المختبرات الطبية

الحقيبة التدريسية لمادة

Medical Mycology

الصف الثاني

.....

تدريسي المادة

م. م ختام جبر حسن

.....

الفصل الدراسي الثاني

جدول مفردات مادة الفطريات الطبية

الاسبوع	المفردات
1	Fungus isolation in general
2	Equipment, chemicals needed for fungal media
3	Type of Fungal culture media
4	Isolation and cultured pathogenic Fungi
5	Using biochemical tests for identification
6	Macroscopic examination of fungal colonies
7	Microscopic examination
8	Dermatophyte identification
9	*Candida* identification
10	*Pencillium* identification
11	*Aspergillus* identification
12	Actinomyces identification
13	Antibiotic producing by fungus.
14	Anti fungal agents
15	Review

• The purpose of studying laboratory instruments:

acquaint students about medical mycology and diseases caused by how to diagnose and treat .

• Target group:

Second - year students/ Medical Laboratory Technology.

• Educational techniques used:

1. Whiteboard and pens.
2. Interactive whiteboard.
3. Data show.
4. Laptop.
5. Instruments required by the curriculum .

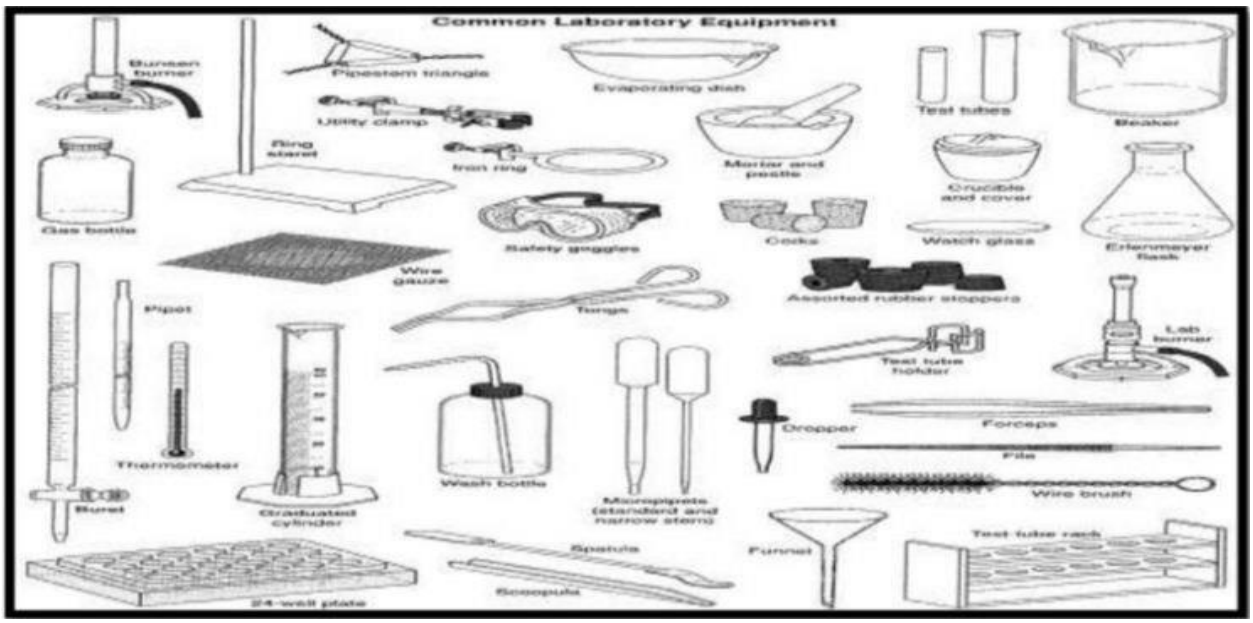
Week - 1 -

Fungus isolation in general

Fungus spread in many environments heavily where there is no place free Which can be ،of the presence of one or more type of fungus or spores isolated from soil, air or water, fungi parasitize humans and plants and less on animal causing diseases and economic losses, so it is necessary to isolation and Diagnosis these fungi to reduce the danger.

Materials and tools:

Slides and cover slips
Petri dishes Burner
Gloves
Lactophenol cotton blue (LPCB)
Pipettes Inoculating
Needle
Flask and bakers
Test tube
Media
Microscope , Autoclave ,Hood UV radiation lab , Hoot air oven , Incubater ……

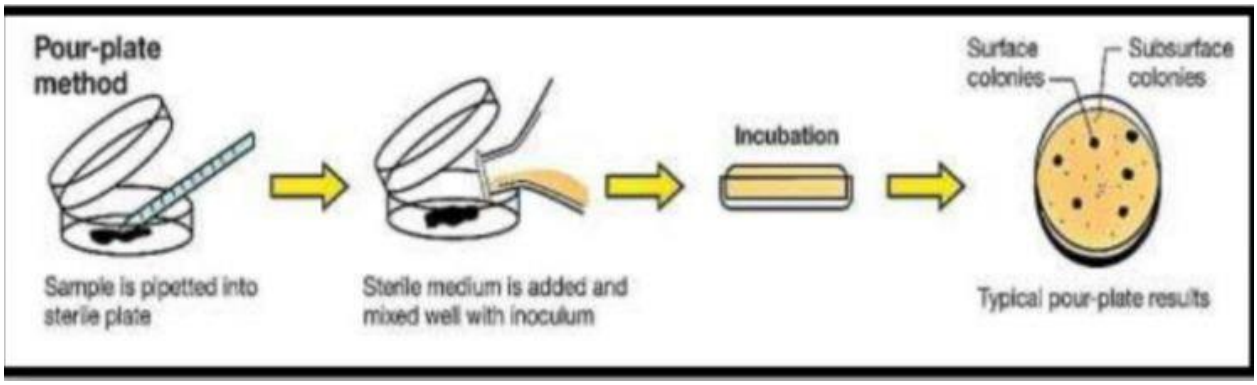


1-Isolation fungi from soil

a-Pour plate methods (Direct Isolation)

- Weighted 0.1 gm of soil and placed it in Petri dish.
- Poured the culture media in Petri dish near flame .
- Moved the Petri dish with a circular movement right and left for mixing the soil with media .
- Leave the plate until it solid , then incubated for 5 days at 25-28 °C.

Not: Can distribution the soil sample on the surface of solid culture media .



Pour plate method

b- Indirect isolation :

It's a modified method of poured plate methods, use to Purification fungi from bacteria .

- Add 10 ml of culture media in Petri dish and left to solid.
- Placed 0.1 gm of soil sample above the culture media .
- Poured 5 ml of culture media on the plate and left to solid.

In this case hyphal fungi grow to high because it **Aerobic** organisms while the bacteria (**anaerobic**) stay within the culture media .

c-Dilution method:

•Weighted 1 gm of soil and placed it in test tube contain 10 ml Distilled water(DW) (stoke), Mixed it well. •Prepare 5 test tube each contain 9 ml of (DW).

•Take 1ml of stoke to the first tube that will be first dilution 1/ 11 (10-1).

•Take 1ml of the first dilution to the second tube that will be the second dilution 1/ 111 (10-2).

•Take 1ml of second dilution to the third tube that will be the third dilution 1/ 1111 (10-3).

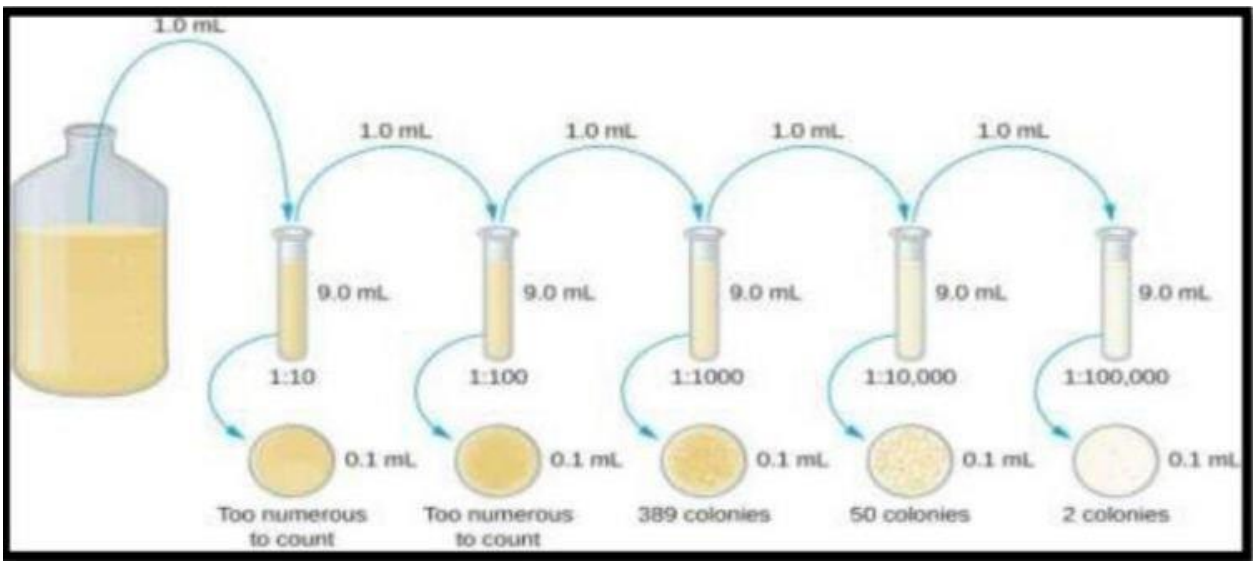
•Take 1ml of third dilution to the fourth tube that will be the fourth dilution 1/ 11111 (10-4).

•Take 1ml of fourth dilution to the fifth tube that will be the fifth dilution 1/ 111111 (10-5).

•Placed 1 ml of each filtrate dilution in a Petri dish and then poured the culture media, moved the dish for mixing the sample with the culture media.

•Leaf it until solid , then incubated for 5 days at 25-28°C.

Not: The dilution (1ml) can be distributed on surface of solid culture media .



2-Isolation fungi from air:

Poured the media on Petri dishes and leave it until solid . Open the dished in several different places for 30 min. Incubated for 5 days at 25-28 °C.

3-Isolation fungi from water:

a-Dilution method:

1-Take 1ml of water sample placed it in test tube contain 9ml of (D.W) , that will be first dilution 1/ 11 (10-1).
2- Take 1ml of the first dilution to the second tube that will be the second dilution 1/ 111 (10-2).
3--Take 1ml of second dilution to the third tube that will be the third dilution 1/ 1111 (10-3).
4-Take 1ml of third dilution to the fourth tube that will be the fourth dilution 1/ 11111 (10-4).
5-Take 1ml of fourth dilution to the fifth tube that will be the fifth dilution 1/ 111111 (10-5).
6- Placed 1 ml of each filtrate dilution in a Petri dish and then poured the culture media, moved the dish for mixing the sample with the culture media.
7- Leaf it until solid , then incubated for 5 days at 25-28°C.

Not: The dilution (1ml) can be distributed on surface of solid culture media .

b-Filtration methods:

Filtrate the water sample using cellulose filter Placed in a sterile suppression.

- After the passage the sample, Filter paper taken by sterile forceps and placed on surface of culture media then incubated the dishes for 5 days at 25-28 °C.

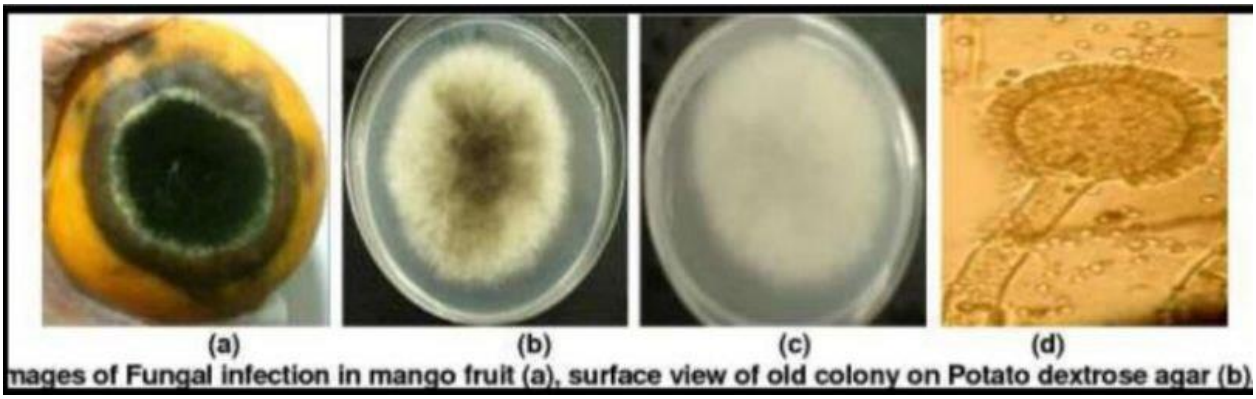
4-Isolation fungi from fruits and vegetables:

- Placed the media into the plate and left it until solid .
- Surface sterilization to rotten part by Sodium Hypochlorite or Potassium Permanganate (2%) .
- Washing the sample by (D.W) 2-3 times .
- Transfer part rotten to the Petri dish by a sterile needle.
- Incubated the dishes for 5 days at 25-28 °C.

5-Isolation of Medically Important Fungi

Laboratory diagnosis of mycoses (fungal infections of Human and other vertebrates) is carried out by demonstration of the fungus in the skin, exudates or deeper tissues. Isolation and identification of the causative agent in culture is usually necessary to confirm this. Proper collection and rapid transport of clinical specimens are very important for successful diagnosis of mycotic infections and the recovery of etiological agents. Prompt delivery of specimens to the laboratory is important to avoid overgrowth of bacteria or rapidly growing saprophytic fungi. Specimens should be transported in a sterile container that provides a moist environment. Sterile saline can be added if necessary. If storage of specimens is necessary, they should be stored at 4˚ C for no longer than 24 hours. However, some loss of specimen viability may occur. Sufficient material of specimens should be collected for both direct examination and culture. Specimens may include: skin scraping, nails, scraping from ulcers, pus, cerebrospinal or other body fluids, urine, sputum, blood, bone marrow, and stools.

Filtration method



(a) (b) (c) (d)

mages of Fungal infection in mango fruit (a), surface view of old colony on Potato dextrose agar (b)

Week - 2,3-

Equipment, chemicals needed for fungal media and Type of Fungal culture media

Culture media for Fungi

Culture media: Balanced mixture of different nutrients necessary for the growth of microorganisms, it may be simple or complex composition in each case serves to provide the energy and basic units for building cells.

The purpose of using Culture media

- Growing and preserving fungi.
- Study the effect of single nutrients found in media on the growth of fungus.
- Inducing fungi to produce and forming some material.
- Classification of fungi and study the cultural characteristics.

Common ingredients of culture media

• Peptone- source of carbon and nitrogen
. • Beef extract- source of amino acid, vitamins, minerals.
• Yeast extract- source of vitamin, carbon, nitrogen.
• Distilled water
• Agar- solidifying agent Common ingredients of culture media
• Peptone- source of carbon and nitrogen.
• Beef extract- source of amino acid, vitamins, minerals.
• Yeast extract- source of vitamin, carbon, nitrogen.
• Distilled water • Agar- solidifying agent

Division of Culture media

a- According to the chemical composition:

- Chemically defined media: Must be known composition, consists of metal salts have added some sources of carbon or nitrogen can be prepared each time the same precision ex: Czapek's Agar (CZ).

♦Chemically no defined media: Not have a specific composition, composition changed depending on the nature of the material prepared, Difficult prepared each time the same precision ex: Potato Dextrose Agar (PDA), Corn Meal Agar (CMA), Malt extract agar.

♦ Natural media: Use of natural materials without additions, ex: Extracts of the roots of potatoes or carrots, Prepared from wheat or barley or corn.

b- According to the Textures:

♦ Solid media: It may be natural such as potato chips, or it may be artificial, such as (PDA)Containing (Agar).

♦ Semi solid media: Contains a half or a quarter of the amount Agar added to solid media.

♦Liquid media: Not contains Agar such as (PD) artificial, (Milk) natural.

c- According to the purpose:

♦ General purpose media: Media are used to growth different types of fungi, such as :

1. Water Agar (WA).
2. Potato Dextrose Agar (PDA).
3. Carrot Agar.
4. Malt extract agar.
5. Czapek′s Agar (CZ).
6. Corn Meal Agar (CMA).

♦ Selective media: Contains a substance inhibits the growth of some fungi while helping growth another kind , such as add some antibiotics or modify the value of (PH) , or add salt , or use Rose Bengal ex:

1- Selective Fusarium Agar.
2- Phytophthora selective medium.

•Notes: Some additives to the culture medium to get a pure Culture, contamination-free:

1. Media with cyclohexamide (cycloheximide is added to inhibit the growth of rapidly growing contaminating molds)

2. Media with or without an antibacterial agent (chloramphenicol, gentamicin and ciprofloxacin are commonly used antibacterial for this purpose).

♦Types of culture media based on chemical composition /application: There are seven routine laboratory media.

1. Basal media
2. Enriched media
3. Selective media
4. Enrichment media
5. Indicator media or differential media.
6. Transport media
7. Storage media.

• Preparation of Culture Media General

1- Broth & agar media are prepared by dissolving specified amount of powder in distilled water.
2- Boiling is often required to dissolve the powder by autoclave in 121 C° for 15-20 min.
3- Cool the flask containing the culture media to about 50 C°
4- Pour the culture media on the Petri dishes let it until Solidify.

• Application of culture media

1. To culture microbes.
2. To identify the cause of infection.
3. To identify characteristics of microorganisms.
4. To isolate pure culture.
5. To store the culture stock.
6. To observe biochemical reactions.
7. To test microbial contamination in any sample.
8. To check antimicrobial agents and preservatives effect.
9. To observe microbe colony type, its color, shape, cause.
10. To differentiate between different colonies.
11. To create antigens for laboratory use.
12. To estimate viable count.
13. To test antibiotic sensitivity.

♦ Limitations of culture media

1. Risk of cross-contamination.
2. High skill required for optimal results.
3. Increased drying out of media can occur.

Week - 4-

Isolation and cultured pathogenic Fungi

- To confirm clinical suspicion to establish fungal cause of disease
- To help in :

1- Chooeing a therapeutic agent .
2- Monitoring the course of disease .
3- Confirming mycological cure .

Specimen Collection

a) Superficial Mycosis

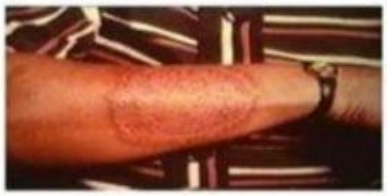
- clean the part with 70 % alcohol .
- collect the material in a sterile paper or a sterile petridish to :

1- Allow drying of the specimen .
2- Reduce bacterial contamination .
3- Maintain viability .

(a) Superficial Mycosis

- **Dermatophytic lesion** – spreads outward in a concentric fashion with healing in the center – scrape outwards from the edge of the lesion with a scalpel blade or use Cellophane tape

- **Scalp lesion –** scraping with a blunt scalpel, including hair stubs, scales & contents of plugged follicles.



- **Scalp lesion –** Wood's lamp examination of infected hair – fluorescence- ring worm infection.

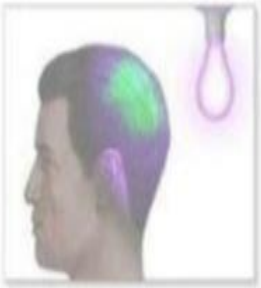
  Hairbrush sampling technique – esp. for culture.

- **Onychomycosis** – Fungal infection of nail.

- Stop antifungals one week prior to collection.

- Sample collected from base of the nail as fungus in distal end is non-viable, include full thickness of nail.

- **Mucosal infections** – Mucosal scrapings are preferred over swabs.



Infectious organisms glowing under Wood's lamp illumination

b) Subcutaneous mycosis

- Scraping or crusts from the superficial parts of lesions.
- Collect carefully usually contaminants are more in these area .
- Pus aspirates & Biopsy are valuable .

C- Systemic mycosis

Pus , Biopsy , Feces , Urine , Sputum, CSF, Blood , scraping or swabs from the edge of lesions .

Collection & Transport of specimen

- Proper collection of specimen and in Adequate quantity .
- Early transport to the lab to avoid Overgrowth of contaminant .

- Sputum early morning sample , after mouth wash flakes to be used for culturing.

Blood

- In biphasic Brain Heart Infusion agar
- Inoculated in 2 bottles.for dimorphic fungi.
- Subculture is done after 2 days and 7 days

Cerebrospinal fluid

else stored Should be immediately processed at RT or at 30°C . in an incubator .

Centrifuge&use sediment for culture.

Skin, Hair & Nail

Taken for Dermatophytic infections

Hair: plucked with forceps.

Tissue, BM & Body fluids

Tissues:grind or mince before culturing.

Body fluid : centrifuge & use sediment for culture .

Urine : Centrifuge & use sediment for culture.

Stool : Not suitable, lntestinal biopsy better .

Eye : ln Keratomycosis , scraping from base and margins of ulcer are taken using Kimura spatula .

Culture Media

- Fungal Culture is frequently performed for isolation and correct identification of the fungi.

- **CULTURE MEDIA –**
- **SABOURADU'S DEXTROSE AGAR (SDA) –**
- Most commonly used medium in diagnostic mycology.
- Contains – Peptone - 1%.
- Dextrose – 4%.
- pH – 5.6.
- Antibiotics (gentamicin, chloramphenicol) and cycloheximide
- This may not support some pathogenic fungi as
- Cycloheximide is not used when *Cryptococcus, Aspergillus or Penicillium* is suspected

- RICE STARCH AGAR CORN MEAL AGAR AND

They are nutritionally deficient media used for stimulation of chlamydospores production

- BRAIN HEART INFUSION AGAR (BHI) & BLOOD AGAR

Enriched medias used for cultivation of fastidious fungi like *Histoplasma* & *Cryptococcus* .

- NIGER SEED AGAR & BIRD SEED AGAR

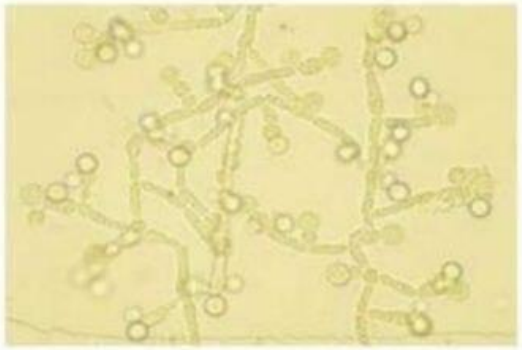
Used as selective media for *Cryptococcus* forms brown color colonies

-CHROM AGAR CANDIDA MEDIUM

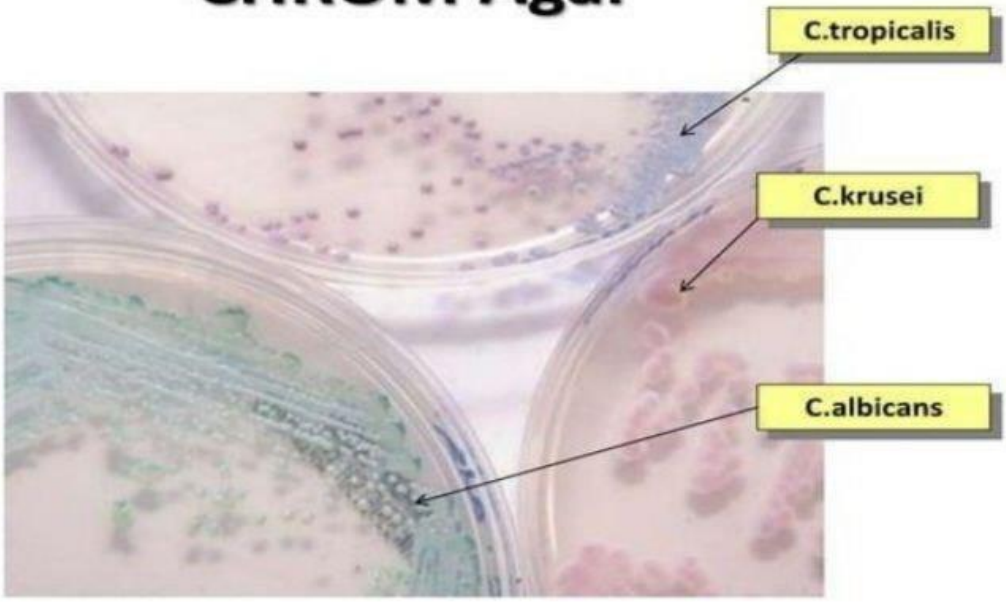
Used as selective as well as differential for speciation of *Candida* .

Corn Meal Agar

Bird Seed Agar



CHROM Agar



Fungal Culture

- **Specimens should be cultured on agar slants:**
  - Safe
  - Require less space
  - More resistant to drying during prolonged incubation
  - Blood cultures should be inoculated in to biphasic blood culture bottles

Temperature requirement

- of fungi Majority 37 C°
- Superficial mycosis30 C°
- Dimorphic fungi: 25 C°& 37 C°

Incubation time

- At least 4 Weeks .

-Usually positive cultures are obtained in 7-10 days .

- Candida & Aspergillus-24 to 72 hrs .

Week - 5-

Using biochemical tests for identification

biochemical tests for identification Fungi

- **Biochemical tests :are among the most important methods for microbial identification .**
  - **Role of Biochemical tests of fungi**

Biochemical testing of fungi involve the use of various laboratory techniques to identify and characterize fungi based on their metabolic activities and biochemical properties .

These tests help microbiologists distinguish between different fungal species and genera.

- **Some commonly used biochemical tests and their significance :**

1- Urease test

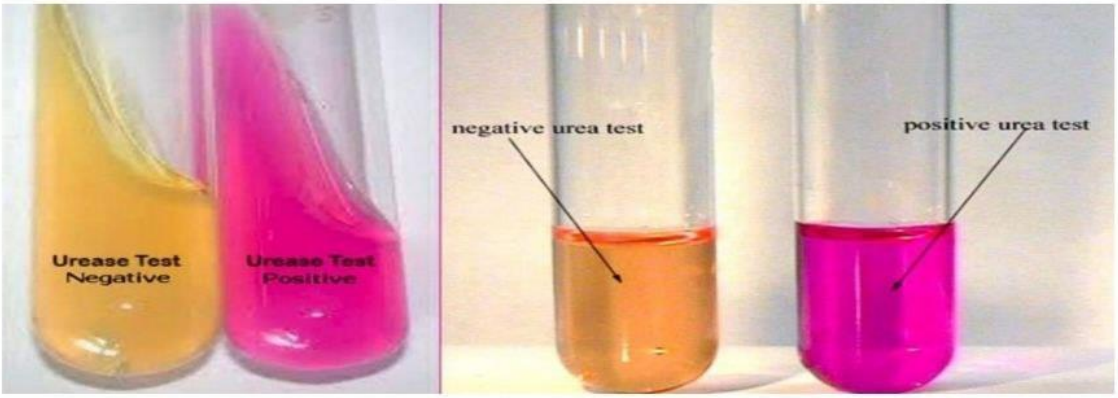
Purpose :

Detects the ability of a fungus to produce the the enzyme urease , which hydrolysis urea to ammonia and carbon dioxide .

Procedure :

Inoculate the fungus on urea agar medium and observe for a color change (pink to magenta) due to the production of ammonia .

Ex : *Cryptococcus neoformans* is urease positive .



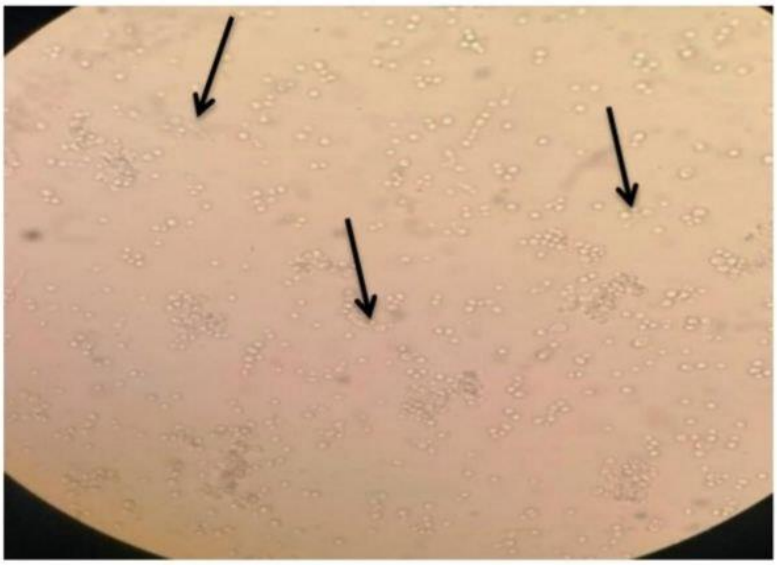
2- Germ Tube test

Purpose :

Used to differentiate *Candida albican* from other *Candida* species .

Procedure:

Inoculate a small amount of yeast in to human serum or plasma and incubate at 37 C . Candida produces germ tubes (hyphae – like projections) within 2-3 hours .



Germ tube in *Candida albicans*

3- Nitrate Reduction Test

Purpose :

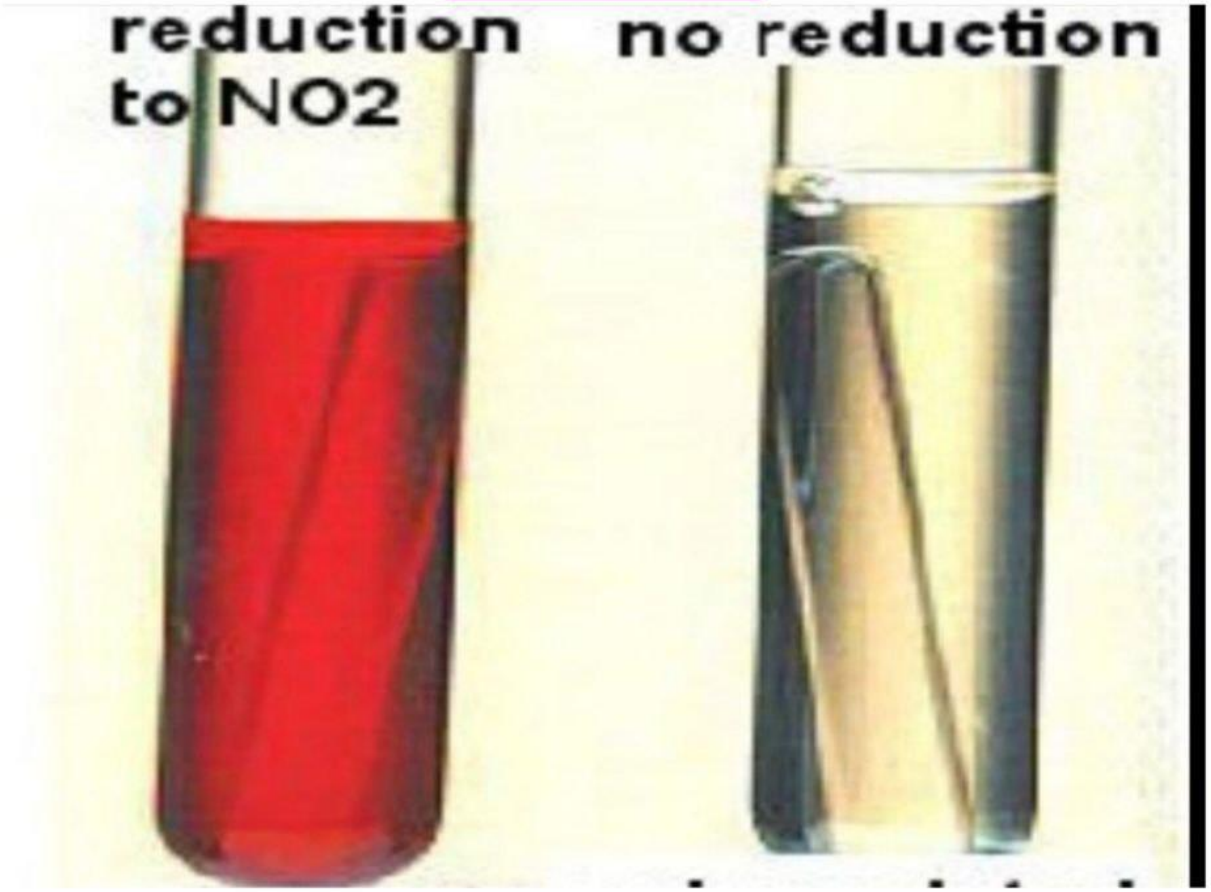
Determines the ability of a fungus to reduce nitrate to nitrite to or other nitrogenous compounds .

Procedure :

Inoculate the fungus in to a nitrate broth and add nitrate reagents. Observe for color changes . further testing may be required to confirm the results .

Ex : *Aspergillus* species are nitrate reducers .

Nitrate reduction



4- Catalase test

Purpose :

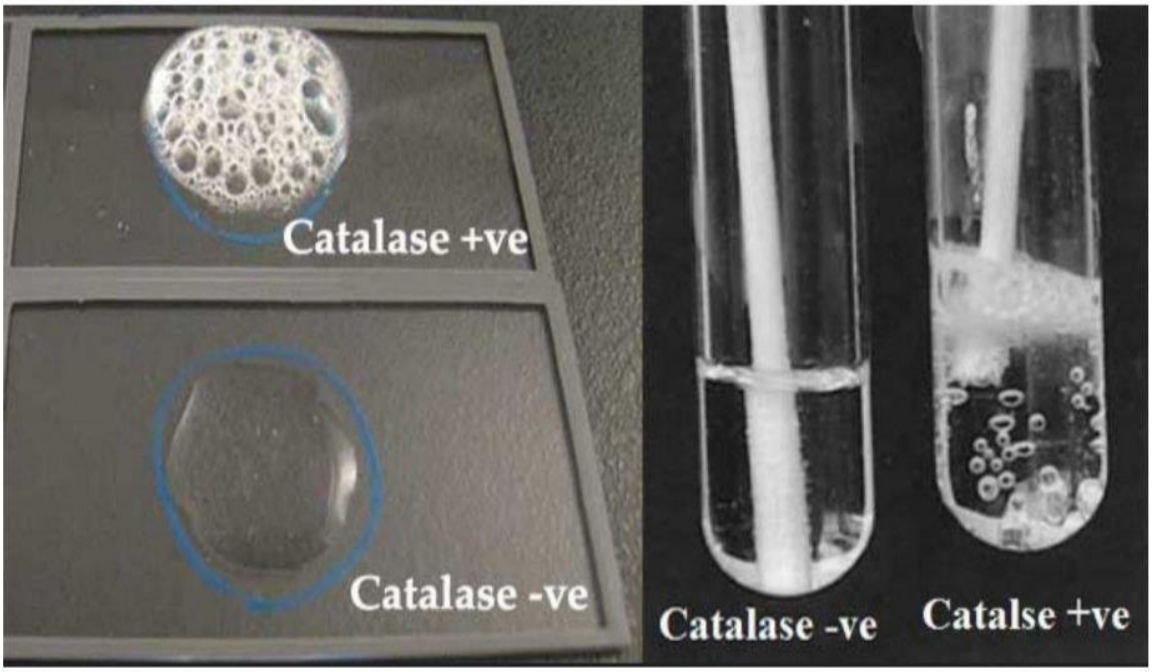
Identifies fungi that produce the enzyme catalase, which breaks down hydrogen peroxide in to water and oxygen .

Procedure :

Apply adrop of 3% hydrogen peroxide to fungal culture .

The release of oxygen bubbles indicates a positive catalase reaction .

Ex : *Candia* species are catalase positive .



5- Starch Hydrolysis test

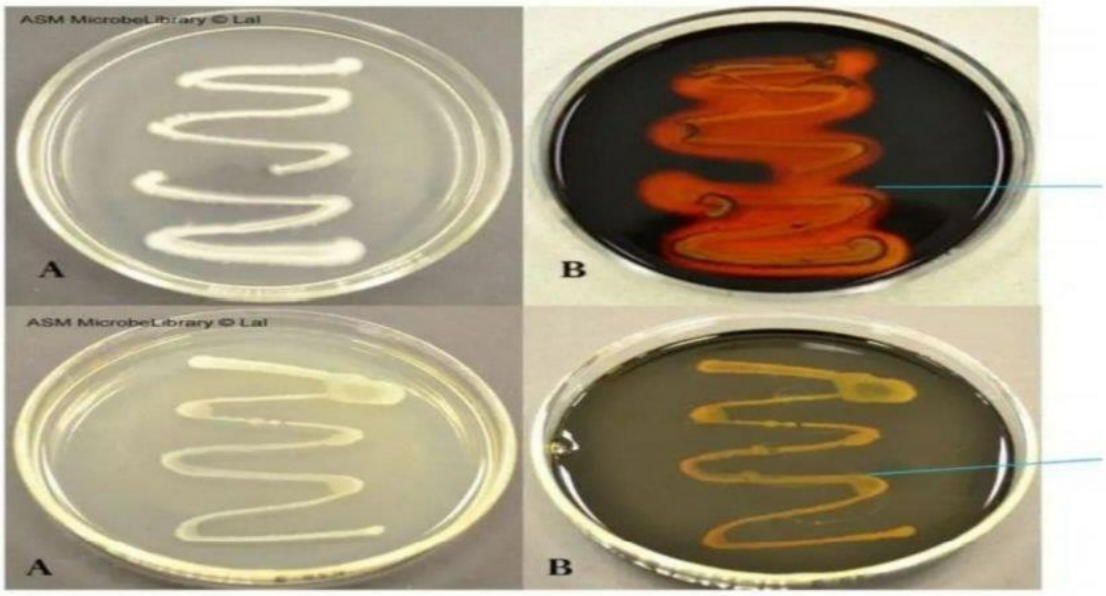
Purpose :

Determines the ability of a fungus to hydrolysis starch using the enzyme amylase .

Procedure :

Inoculate the fungus on a starch agar plate and flood the plate with iodine after incubation . clear zone around the fungal growth indicate starch hydrolysis .

Ex: Tricophyton species typically exhibit starch hydrolysis .



Before addition of the iodine solution

After addition of the iodine solution

6- Lipase test

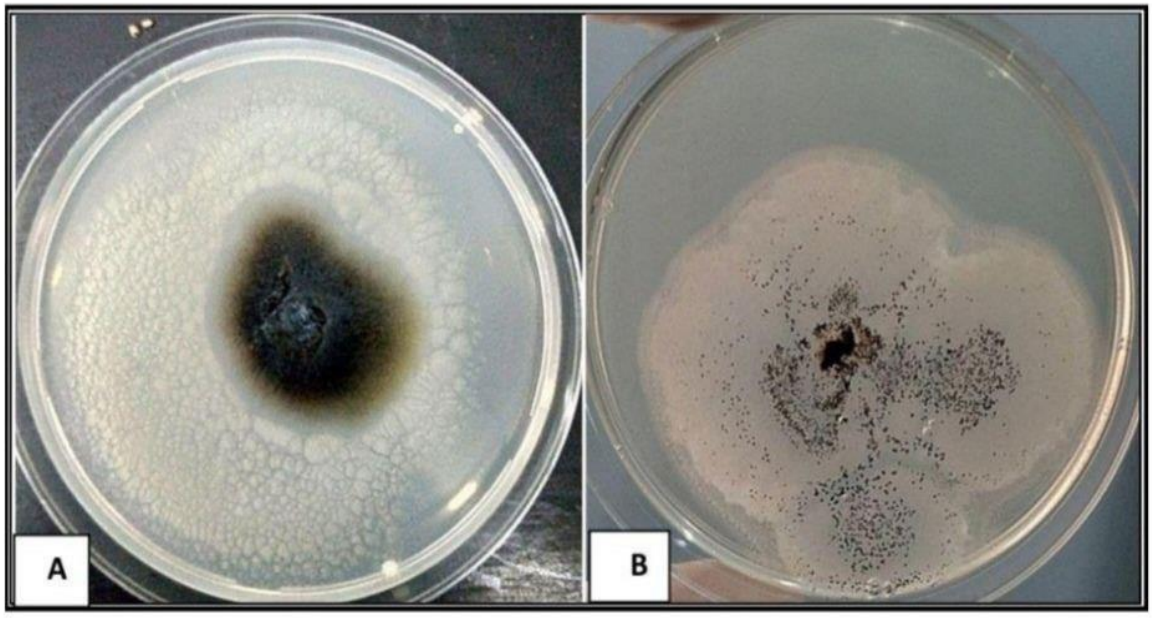
Purpose:

Detects the production of lipase , an enzyme that hydrolysis lipids .

Procedure :

Inoculate the fungus on medium containing lipids and observe for the formation of a precipitate or clearing around the fungal growth.

Ex: Malassesia furfur produced lipase .



Lipase test

7- Lactase test

Purpose :

Identifies fungi capable of hydrolysis lactose .

Procedure :

Inoculate the fungus on a medium containing lactose , observe for acid production (change in pH) and gas formation .

8- Carbohydrate fermentation test

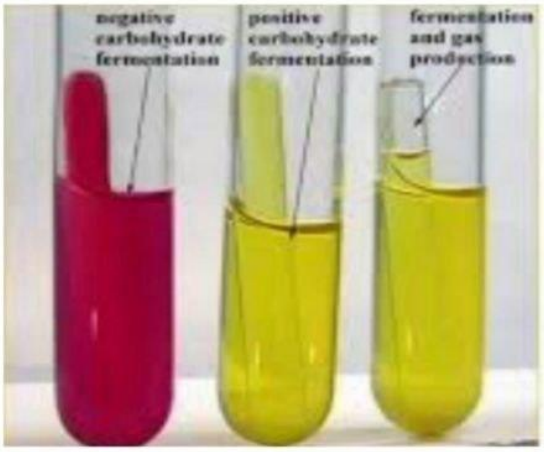
Purpose :

Identifies fungi capable of fermenting specific carbohyrates.

Procedure :

Inoculate the fungus on medium containing a specific carbohydrate(ex : glucose , lactose) . observe for acid production (change in pH) or gas formation .

Ex : *Candida tropicalis* ferments glucose .



Week - 6,7-

Macroscopic examination of fungal colonies and Microscopic examination

Identification of the isolated fungi on culture is done by :

For molds : identification is done by :

- ✓ **Macroscopic examination .**
- ✓ **Microscopic examination .**

For Yeasts : identification is done by :

- ✓ **Microscopic examination .**
- ✓ **Biochemical tests .**

MACROSCOPIC APPEARANCE OF COLONIES

- Rate of growth

Rapid growth (<5 days): Seen in Saprophytes, Yeasts and agents of opportunistic mycocses .

- Pigmentation

Seen on erverse side of the culture media slant .

- Texture

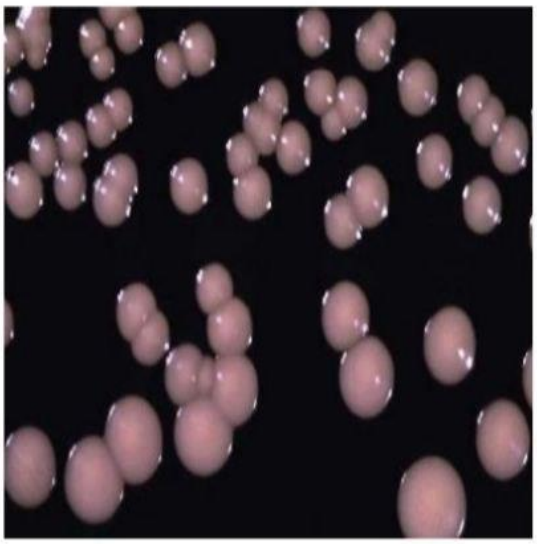
Refers to if allowed to touch how colony fell .

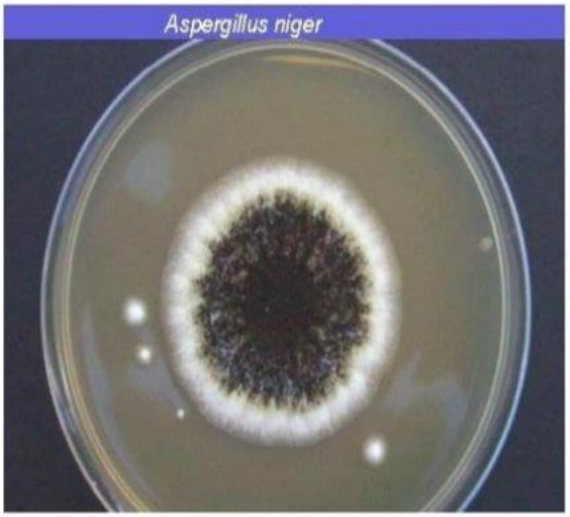
For e.g. Waxy/leathery, velvety, yeast like , cottony , or granular Powdery.

- Colony Surface .

Rugose (radial grooves), folded, verrucous or cribriform (Brain like).

Colony morphology – colour, texture, pigment production





Microscopic examination

- ***According to the site of infection.***
  - ***For example, skin scales, nails, hair clippings for dermatophyte examination.***

Microscopic examination of these specimens using KOH 10%:

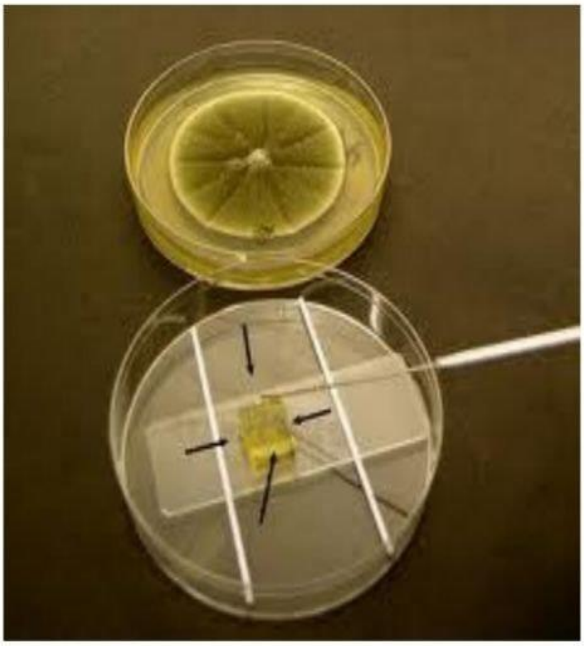
- ***KOH dissolves keratin but does not***

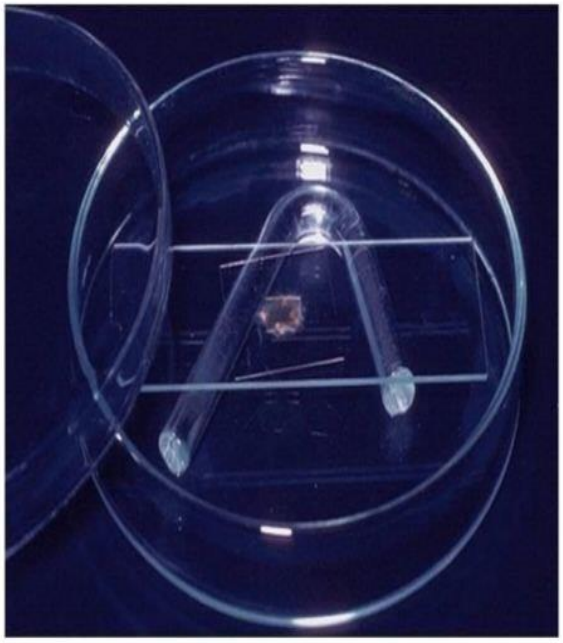
***affect** fungi so* **hyphae seen fungal clearly under microscope.**

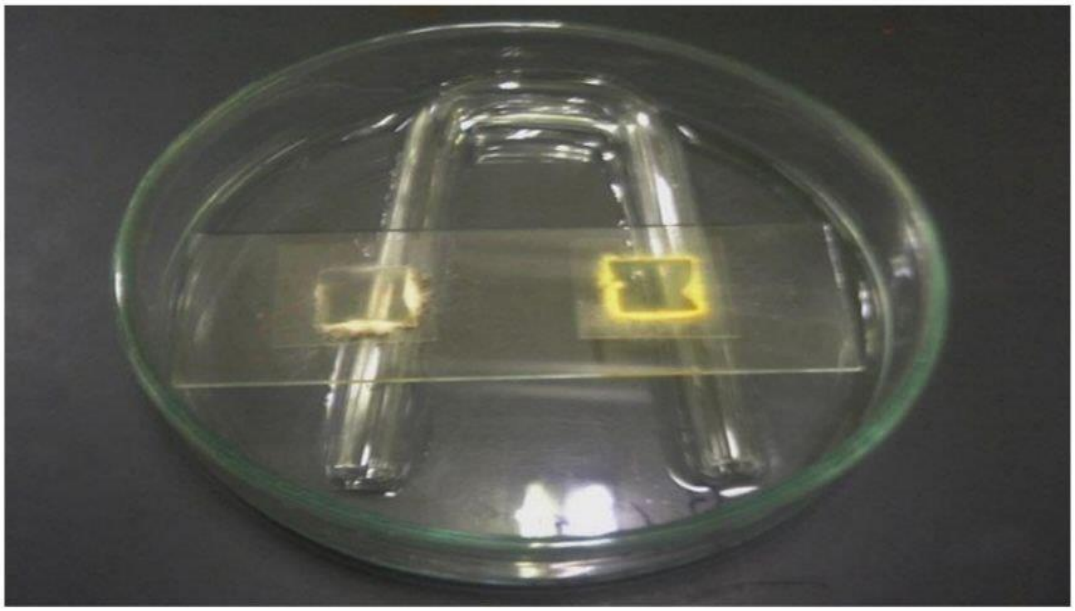
- ***Fungal stains such as lactophenol cotton blue could be used.***

Slide culture

- **In** order to avoid disturbing the arrangement of the fungus structures ,slide techniques can be utilized& permanently stained mounts made from the slide and cover glass



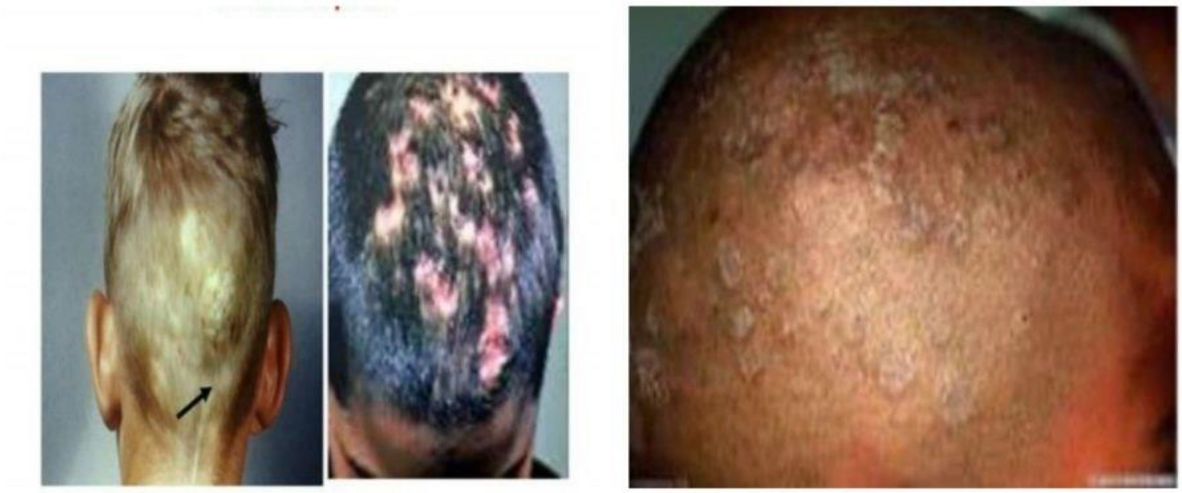




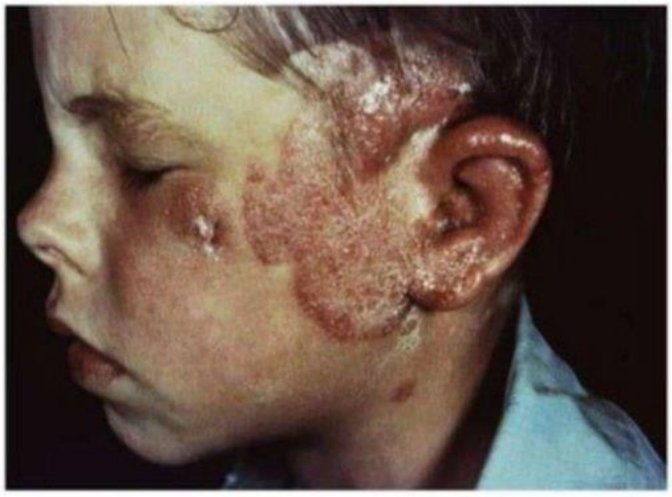
Slid culture technique

Week -8 -

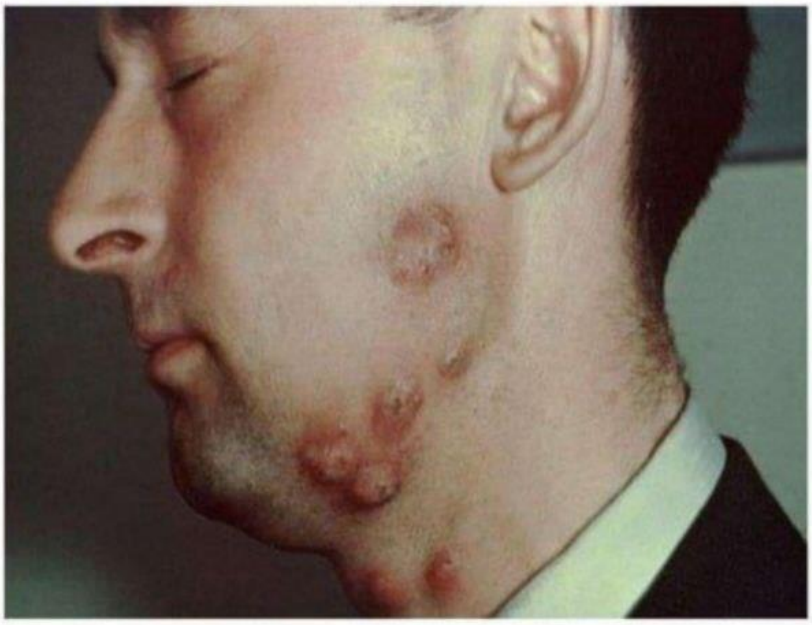
Dermatophyte identification



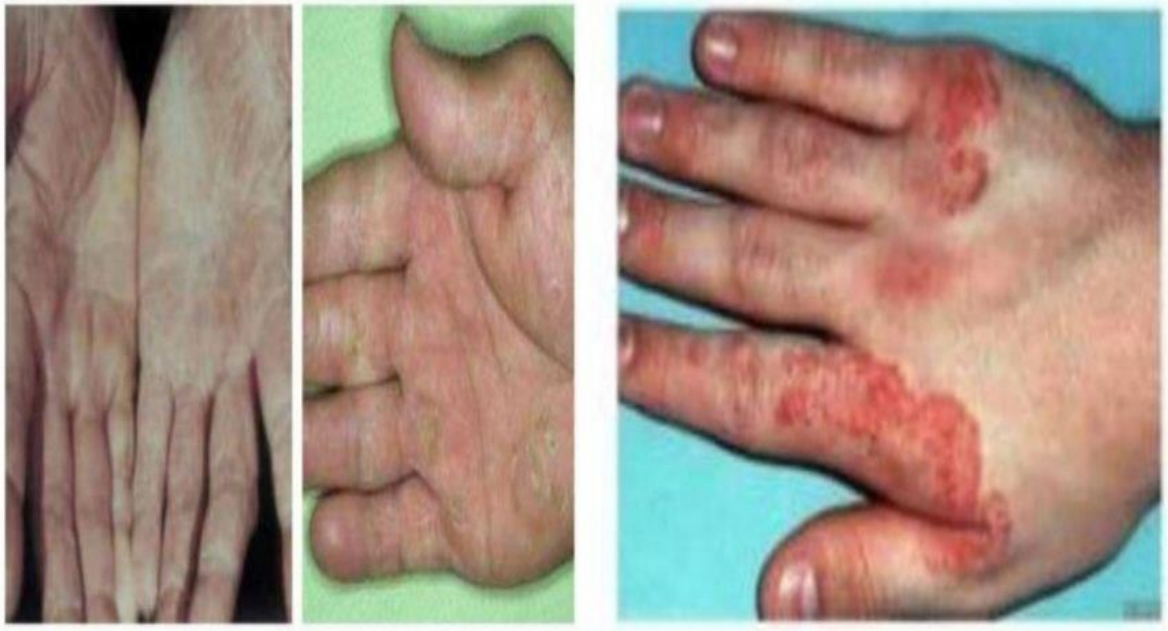
Tinea capitis



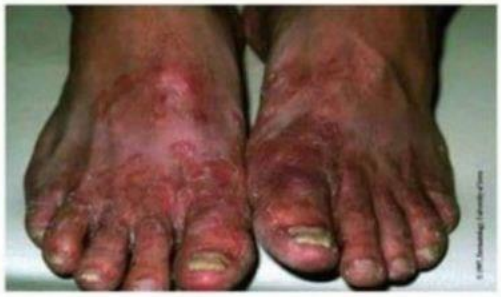
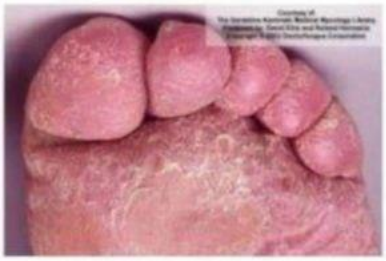
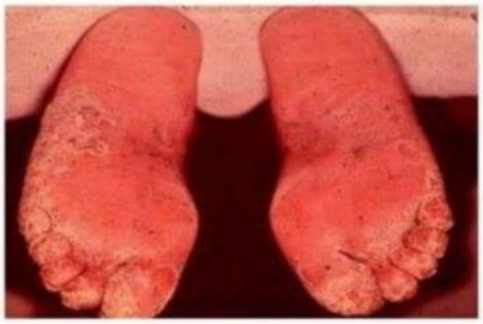
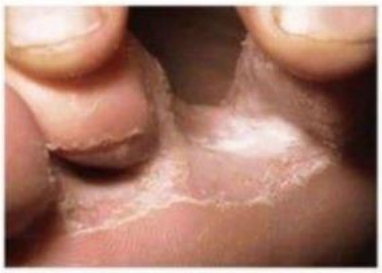
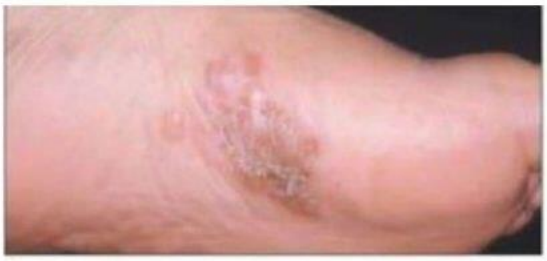
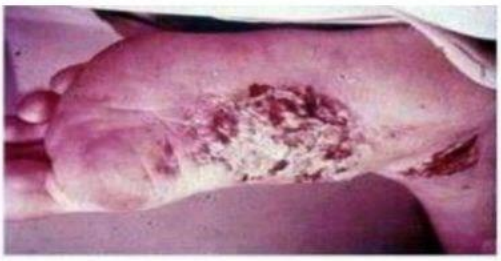
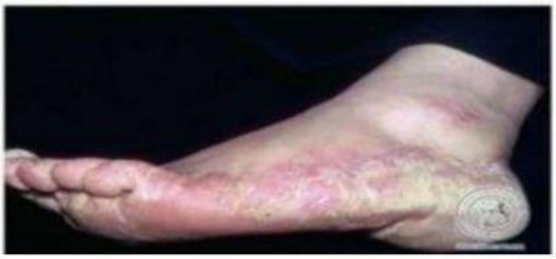
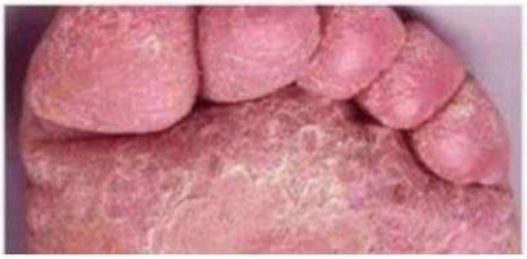
Tinea Faciei



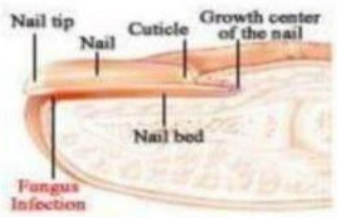
Tinea Barbae

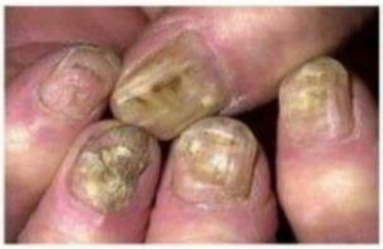


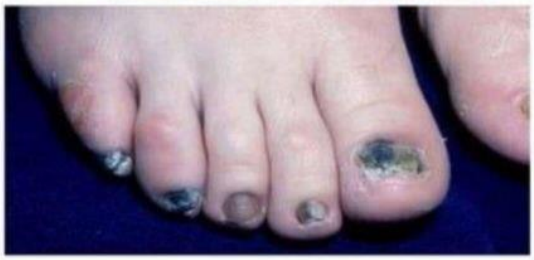
Tinea Manum

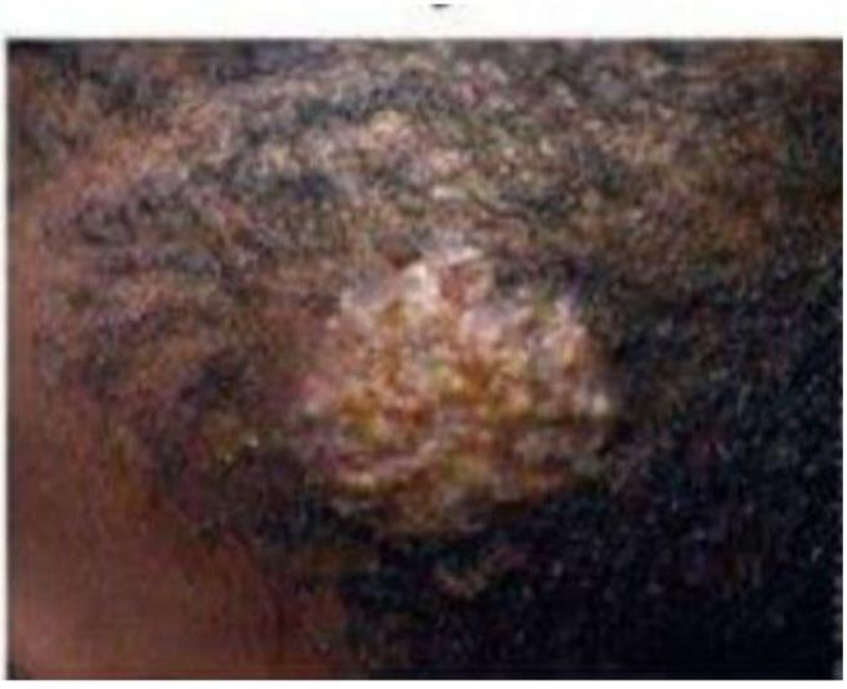
Tinea Pedis



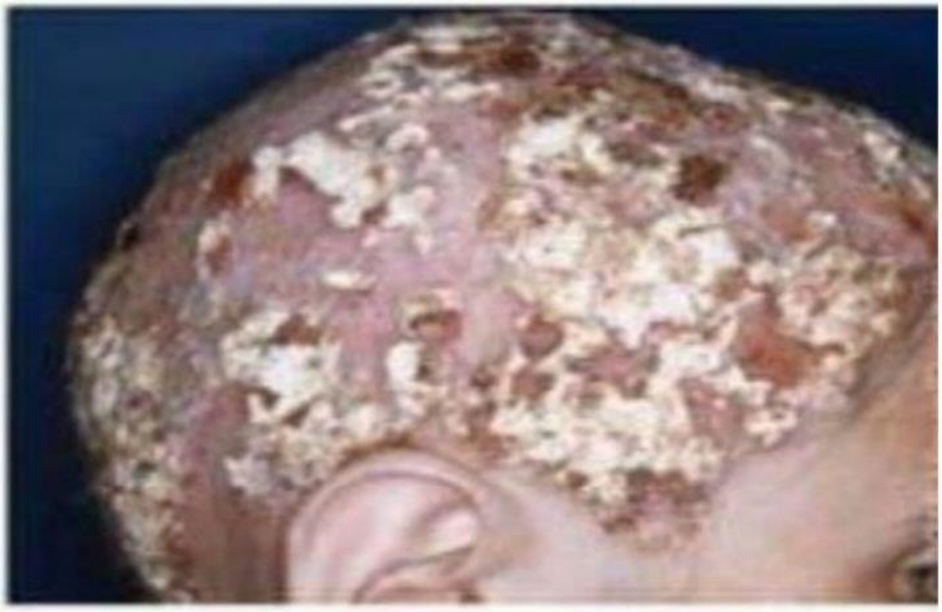




Tinea Unguium

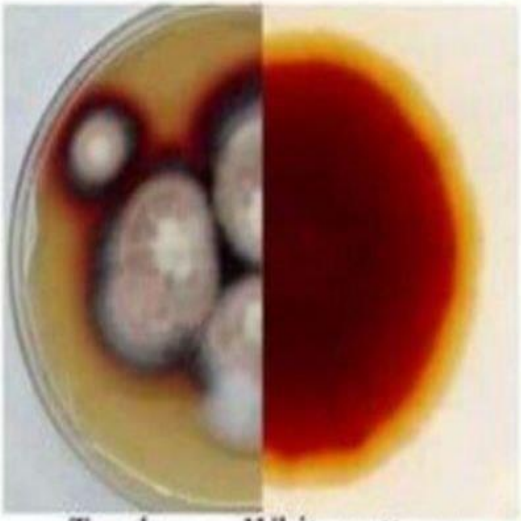


Zoophilic species : induce combined inflammatory and hypersensitivity reaction – kerion

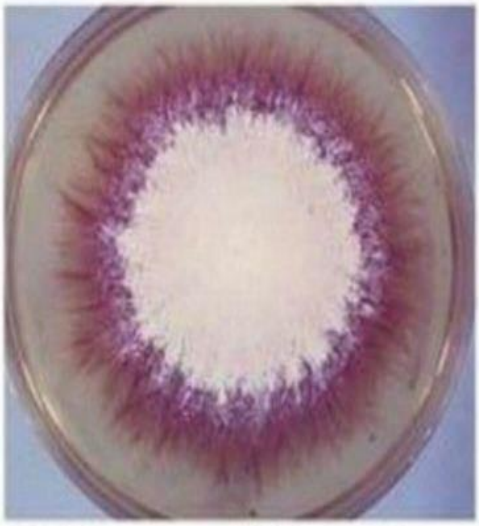


Trochophyton schoenleinii : acute inflammatory reaction of hair follicle leading to formation of scutula (crust) favus.

T. metagrophytes : cottony to granular colony



T. rubrum : White cottony surface and a deep red nondiffusible pigment from reverse side



T. tonsurans : flat, powdery, velvety colony.

Microsporum : white cottony sarface with deep yellow from revese.

Epidermophyton : flat, velvety with a tan to olive green tinge.

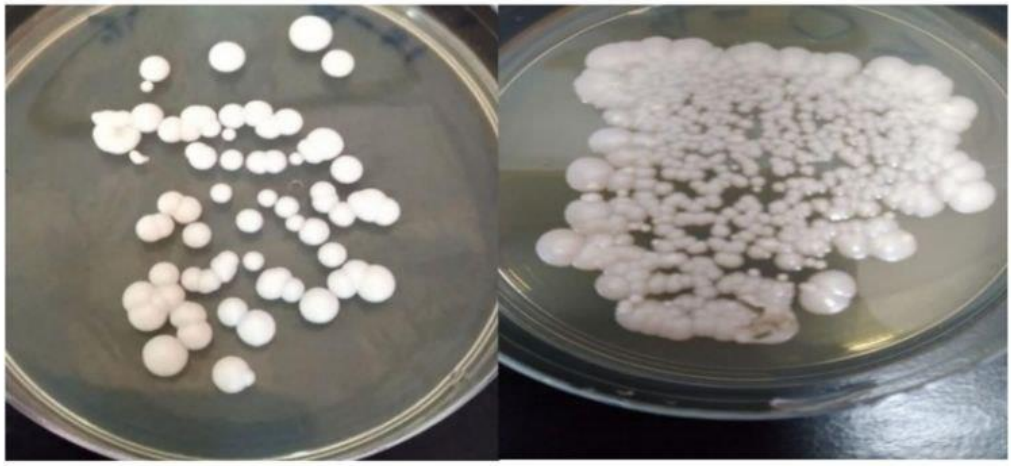
Week -9 -

Candida identification

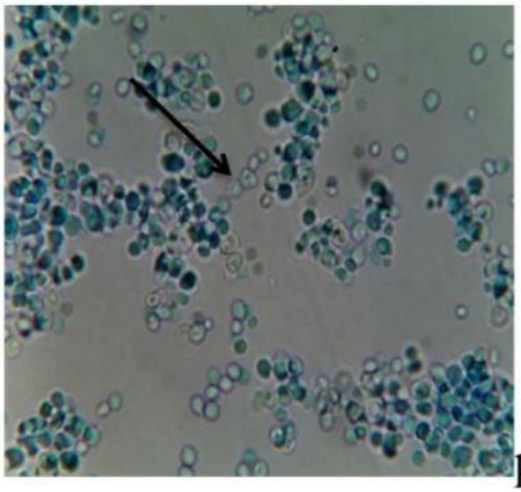
Laboratory Diagnosis of *Candida*

- Direct microscopy : Gram positive , oval budding yeast cells with pseudohyphae.
- Culture on SDA : produces creamy white and pasty colony .
- Tests for species identification :

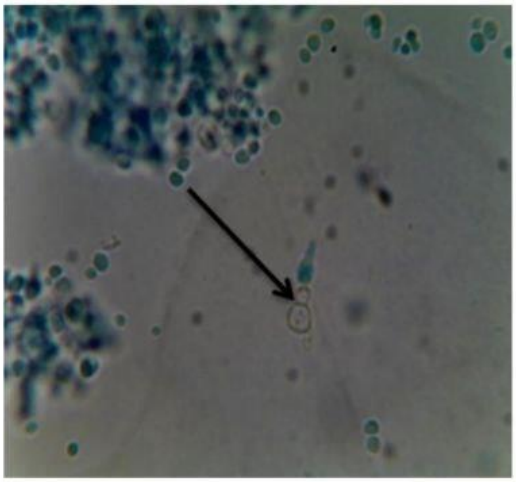
✓ Germ tube test (positive for *C . albicans*).
✓ Dalmau plate culture for chlamydospore production .
✓ CHROM agar .
✓ Growth at 45C° (positive for *C. albicans*).
✓ Sugar assimilation test and sugar fermentation test.
✓ Molecular methods such as PCR.



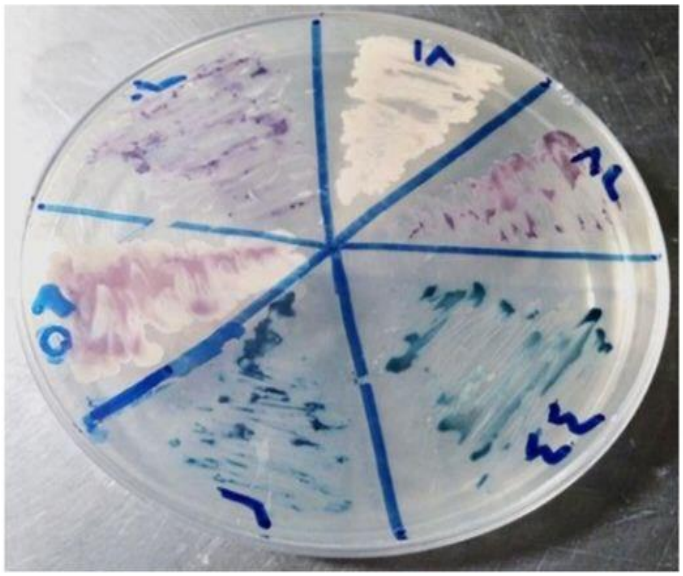
Candida Colony on SDA



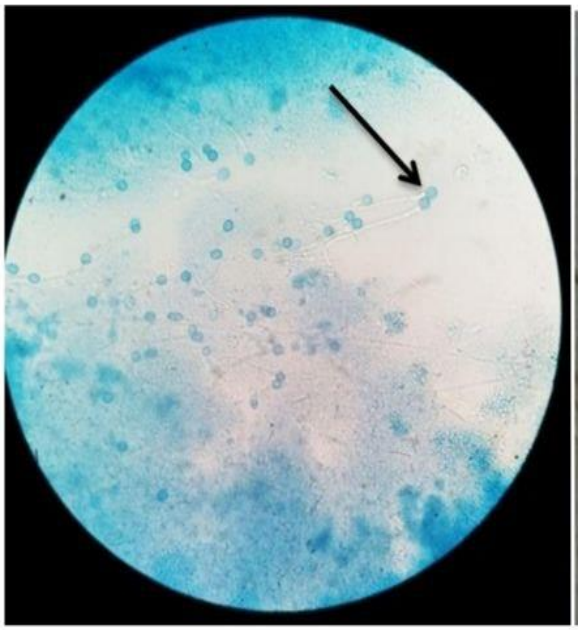
pseudohyphae of *Candida*



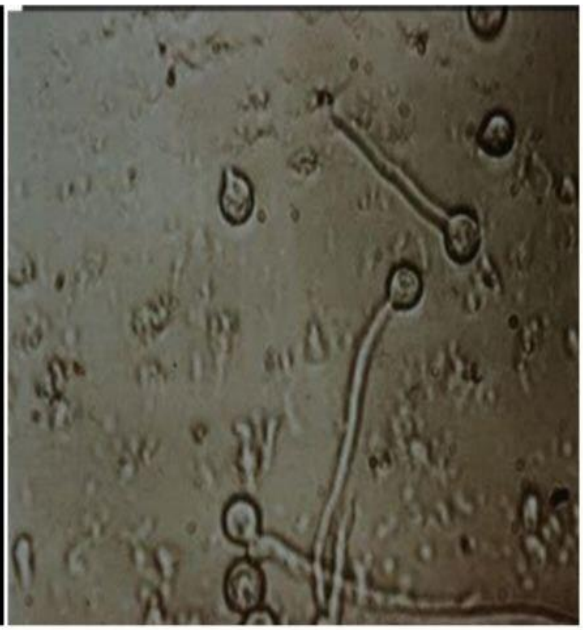
Budding cell for *Candida*



Candida. sp on CHROM agar



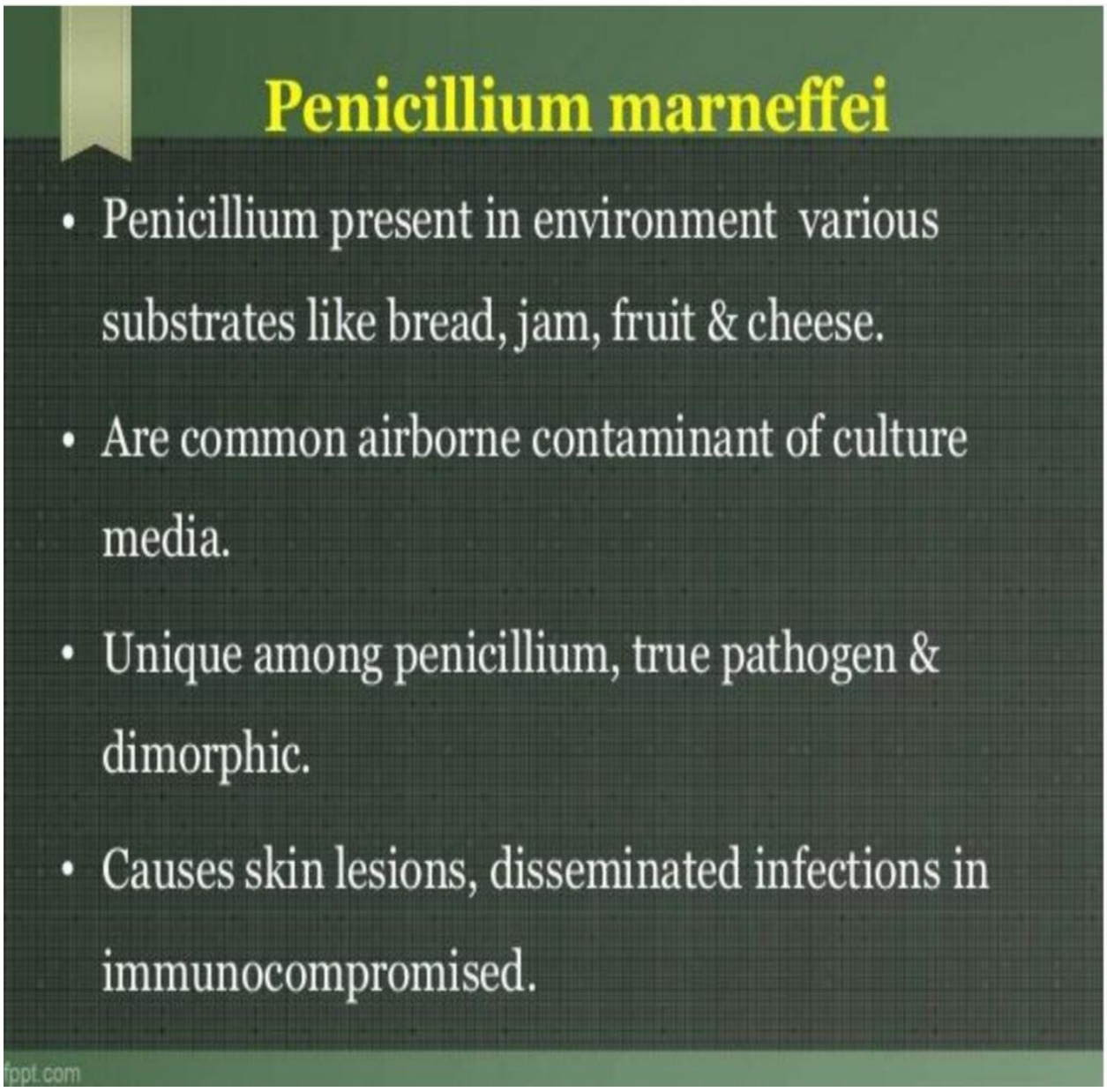
Chlamydospore of *C . albicans*



Germ tube of *C. albicans*

Week- 10 -

Pencillium identification



Penicillium marneffei

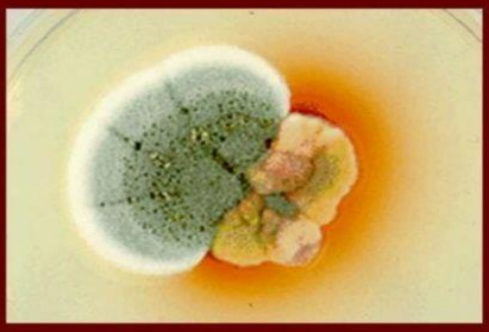
Laboratory diagnosis

- Direct examination
  - Wright or Giemsa stain, PAS, H&E
  - Oval yeastlike cells and multiply within histiocytes in tissue (or within monocytes in blood or BM)

Penicillium marneffei

Mycelial form

- Colony is flat powdery to velvety, gray-green in the center
- A deep-reddish soluble pigment diffuses into the medium



Culture showing a common green saprophytic Penicillium sp. and the typical reddish yellow with a yellow or white edge colony with distinctive red diffusable pigment of *Penicillium marneffei*

Week- 11-

Aspergillus identification

The symptoms of aspergillosis depend on where in the body the fungus is growing. aspergillosis most commonly affects the sinuses or lungs. Symptoms of sinus infections include fever, headache, and sinus pain. Lung infections with the fungus can cause fever and cough.

➢ Sources of Aspergillosis

Aspergillus **is common in the environment, so most people breathe in the fungal spores every day. It is probably impossible to completely avoid breathing in some *Aspergillus* spores. For people with healthy immune systems, this does not cause harm, and the immune system is able to get rid of the spores. But for people with weakened immune systems, breathing in *Aspergillus* spores can lead to infection.**

Studies have shown that invasive aspergillosis can occur during building renovation or construction.

❖ How is Aspergillosis Diagnosed?

Healthcare providers consider risk factors, symptoms, and physical examination when diagnosing aspergillosis. They may also perform imaging tests when necessary, such as chest x-rays or CT scans of the lungs and other organs. Biopsies of affected tissue or samples of respiratory secretions might be analyzed in a laboratory for evidence of the fungus under a microscope or through fungal culture. Other tests are available to monitor high-risk people for invasive aspergillosis, such as those with severely weakened immune systems

- **Treatment of Aspergillosis**
- **How is Aspergillosis Treated?**

Aspergillosis requires treatment with antifungal medication prescribed by a doctor. Voriconazole is currently the first-line treatment for invasive aspergillosis. There are other medications that can be used to treat invasive aspergillosis in patients who cannot take voriconazole or who have not responded to voriconazole. These include itraconazole, lipid amphotericin formulations, caspofungin, micafungin, and posaconazole. Whenever possible, immunosuppressive medications should be discontinued or decreased

Week- 12-

Actinomyces identification

- **General Characteristics**

1- Gram-positive, filamentous bacteria

2- Anaerobic or microaerophilic (does not thrive in oxygen-rich environments)

3- Non-acid-fast (does not retain stain in acid-fast tests)

4- Normal flora of the human oral cavity, gastrointestinal tract, and female genital tract .

- **Pathogenesis and Diseases**

Actinomyces causes actinomycosis, a chronic, slow-growing infection characterized by abscess formation, fibrosis, and sulfur granules (yellowish granules in pus).

- **Common Types of Actinomycosis**

1-Cervicofacial actinomycosis ("Lumpy Jaw") – Most common form, associated with dental infections, oral trauma, or poor oral hygiene

2- Thoracic actinomycosis – Affects the lungs and chest wall, often from aspiration

3- Abdominal actinomycosis – Can occur after bowel perforation or surgery

4- Pelvic actinomycosis – Linked to prolonged use of intrauterine devices (IUDs)

- **Diagnosis**

Microscopy & Culture:

Filamentous, branching rods; presence of sulfur granules Gram staining: Shows gram-positive branching filaments , Anaerobic culture: Grows slowly over 1-2 weeks

- **Treatment**

Penicillin G (high-dose, prolonged therapy) , Surgical drainage if necessary.

Week- 13,14 -

Antibiotic producing by fungus and Anti fungal agents

Antifungal agent

- 1-Polyene Antifungal Drugs
- Amphotericin, nystatin, and pimaricin
- interact with sterols in the cell membrane to form channels through which small molecules .
- leak from the inside to the outside.
- 2-Azole Antifungal Drugs
- Fluconazole, itraconazole, and ketoconazole
- inhibit cytochrome P450-dependent enzymes involved in the biosynthesis of ergosterol, which is required for fungal cell membrane structure and function
- Antifungal Azoles are synthetic drugs

3- Allylamine and Morpholine Antifungal Drugs

inhibit ergosterol biosynthesis at the level of squalene epoxidase.

4- Antimetebolite antifungal drugs

Fluorocytosine : inhibitor of both DNA and RNA synthesis.

Week - 15 -

Review