



Lab-3

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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.

Culture media are divided According to the chemical composition into 3:

Natural media

1. Synthetic media
2. Semi Synthetic media

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

Synthetic 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).

Semi Synthetic 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.

Culture media are divided According to the physical condition into3:

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

B_ Semi solid media: Contains a half or a quarter of the amount Agar addedtosolid media.

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

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.

CHROMagar Media :

This test is performed by inoculating CHROM agar Candida medium which is prepared previously from Candida isolate culture grown on SDA for 24 h, and then incubated at 30°C for 24-48 h. CHROM agar test is used for the presumptive identification of *Candida* species by production of different colors on this medium (*C. albicans* = green/ blue green, *C. dubliniensis* = dark green, *C. tropical* = blue, *C. parapsilosis* = cream white, and *C. krusei* = pink)



Serological and Skin tests.

Serological tests have now gained importance in mycology because of the rapidity of results and these tests can serve as a prognostic indicator.

Serological methods utilise the reactions and properties of the serum. The serological tests are done either to demonstrate antigen or antibody in serum or body fluids of suspected fungal infection

Serological tests in mycology

The specific immune response that results from exposure to cell wall, cytoplasmic extracellular fungal antigen during infection can be used for diagnosis. By monitoring this response, prognosis of disease and outcome of therapy can be assessed.

There are different serological tests :

- Agglutination
- Immunodiffusion (ID)
- Complement fixation test (CFT)
- Enzyme linked immunosorbent assay (ELISA)
- Lateral flow assay (LFA)
- Counter immuno-electrophoresis (CIE)
- Radio immunosorbent assay (RIA)

The advantages of serological tests in mycology are :

- To interpret the clinical significance of positive cultures – to rule out lab contamination
- To identify new isolate when the Antibody is demonstrated against that particular antigen
- Rapid diagnosis
- Prognostic marker

Types of Specimens for Fungal Infections:

1. Scrapings from the superficial parts of damage.
2. Biopsy - Feces – Urine – Sputum – CSF - Blood.

Isolation of fungi from hair, nail and skin

1. The direct plate method is used in isolating fungi.
2. Transfer sample (hair, nail and skin) after being flooded in 10% of KOH
3. solution for 1-2 min to SDA .
4. Incubate petri dish in 28°C for 7-21 days depending on the growth of fungus.



Isolation of fungi from Scrapings:

Isolation of fungi from scraping samples were directly cultured by putting scraping pieces over the agar plates. These plates then incubated at 30°C **Laboratory Diagnosis:**

1. Direct examination
2. Fungal culture
3. Serological tests
- 4- Molecular methods (PCR).