



Chlamydospore Agar

M113

Chlamydospore Agar is used for differentiating *Candida albicans* from other species of *Candida* on the basis of chlamydospore formation.

Composition**

Ingredients	Gms / Litre
Ammonium sulphate	1.000
Monopotassium phosphate	1.000
Biotin	0.000005
Trypan blue	0.100
Purified polysaccharide	20.000
Agar	15.000
Final pH (at 25°C)	5.1±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 37.1 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Candida albicans is a diploid sexual fungus (a form of yeast), and the causative agent of opportunistic oral and vaginal infections in humans (1). *C. albicans* is a commensal of skin, gastrointestinal and genitourinary tract. However, under certain conditions overgrowth of this results into oesopharyngeal candidiasis, vulvovaginal candidiasis and candidemia. Chlamydospores formation is the most differential characteristic of *C. albicans* (1). Chlamydospore Agar was specially designed for the differentiation of *C. albicans* from other species on the basis of chlamydospores formation. It is prepared according to the formula of Nickerson and Mankowshi (2).

Ammonium sulphate acts as sources of ions that simulate metabolism. Monopotassium phosphate provides buffering to the medium. Biotin provides the necessary vitamins required for metabolism. Purified polysaccharide acts as a source of carbon. Trypan blue is a vital dye absorbed selectively by the chlamydospores and imparts blue colour to chlamydospores, whereas the filaments are colourless.

Test for chlamydospores: Scratch cut mark like X onto the agar surface with inoculum using sterile needle. Aseptically place an alcohol-flamed and cooled cover slip onto the agar surface over the intersecting lines of the cut marks of X. Incubate plates at 20-25°C for 2-6 days. Temperature should not be higher than 25°C since it will not permit chlamydospore formation. Observe the plates under low power of microscope. After incubation, most strains of *C. albicans* and *C. stellatoide* will form typical chlamydospores. Chlamydospores will be seen along the edge of the cover slip. Chlamydospores are round, thick walled, blue coloured and at the terminal ends of hyphae.

Some *C. albicans* strains may lose their ability to produce chlamydospores after repeated subculturing.

Quality Control

Appearance

Cream to blue homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Blue coloured, opaque gel forms in Petri plates

Reaction

Reaction of 3.71% w/v aqueous solution at 25°C. pH : 5.1±0.2

pH

4.90-5.30

Cultural Response

M113: Cultural characteristics observed after an incubation at 20-25°C for 2-6 days.

Organism	Growth	Chlamyospores
<i>Candida albicans</i> ATCC 10231	good-luxuriant	positive
<i>Candida kruisei</i> ATCC 24408	good-luxuriant	negative
<i>Candida minosa</i>	good-luxuriant	negative
<i>Candida tropicalis</i> ATCC 1369	good-luxuriant	negative

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference

1. Ryan K. J., Ray C. G., (Eds.), 2004, Sherris Medical Microbiology, 4th Ed., McGraw Hill.
2. Nickerson, 1953, J. Infect. Dis., 92:20

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