

Technical Information

Asparagine Broth (Coccidioidin and Histoplasmin Broth)

Product Code: DM 1672

Application: Asparagine Broth is used for the preparation of Coccidioidin and Histoplasmin antigens for immunodiagnostic work.

Composition**

Ingredients	Gms / Litre
L-Asparagine	7.000
Ammonium chloride	7.000
Dipotassium phosphate	1.3 10
Sodium citrate	0.900
Magnesium sulphate	1.500
Ferric citrate	0.300
Dextrose	10.000
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Histoplasma capsulatum, a dimorphic fungus causes histoplasmosis, a systemic fungal disease. *H. capsulatum* is an obligate intracellular organism present in macrophages of the reticuloendothelial system. Recently there has been increased in the incidence of histoplasmosis in patients with AIDS ⁽¹⁾. *Coccidioides immitis*, the causative agent of coccidioidomycosis (Valley fever) is endemic in hot regions with dry climate and alkaline soil. Patients with AIDS are at a risk of developing coccidioidomycosis.

Asparagine Broth is a chemically defined medium used for the preparation of Coccidioidin and Histoplasmin antigens. *Histoplasma capsulatum* or *Coccidioides immitis* are cultured in this medium for 1-3 months at 37°C till the static phase is obtained. At this stage, cells are autolyzed and a mixture of antigen haptens is prepared. Cell free filtrate from this medium is sterilized by filtration and used as the antigen (2, 3). Preparation, standardization and administration of histoplasmin and the interpretation of delayed cutaneous hypersensitivity tests are identical to those for Coccidioidin (4). The amino acid asparagine, favours the synthesis of antigens from *Histoplasma* and *Coccidioides*. Salts included in the medium buffer the medium well. Dextrose and slightly acidic pH of the medium helps for the luxuriant growth of the fungi

Methodology

Suspend 28.01 grams of powder media in 1000 ml distilled water containing 25 ml glycerol. Mix thoroughly and then dispense in a wide bottom flask, to give a depth of 1 to 1.5 inches. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Quality Control

Physical Appearance

Off-white to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Yellow coloured clear solution with brownish precipitate.

Reaction

Reaction of 2.8% w/v aqueous solution containing 2.5 ml glycerol at 25°C. pH : 6.8±0.2

pH range 6.60-7.00



Dehydrated Culture Media
Bases / Media Supplements

Cultural Response/Characteristics

DM1672: Cultural characteristics observed with added glycerol, after an incubation at 35-37°C for 1 week.

Organism	Inoculum (CFU)	Growth
<i>Coccidioides immitis</i>	50-100	Luxuriant
<i>Histoplasma capsulatum</i> ATCC 10230	50-100	luxuriant

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.

Prepared Media: 2-8⁰ in sealable plastic bags for 2-5 days.

Further Reading

1. Jone P. G., Cohen R. L., Bates D. H., et al., 1983, Six Transm. Dis, 10: 202-204.
2. Smith C. E., Pappagianis D., Levine H. B., and Saito M., 1961, Bact. Rev., 25:3 10.
3. Emmons C. W., Olson B. J., and Eldridge W. W., 1945, Pub. Hlth. Rept., 60:1383.
4. Emmons W. W., Binford C. H., Utz J. P., and Kwon-Chung K. J., (Eds.),1977, Medical Mycology, 3rd Ed., Lea and Febiger, Philadelphia.

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