73142 Canavanine Glycine Bromothymol blue Agar (CGB Agar)

CGB Agar is recommended for the differentiation of *Cryptococcus neoformans var. neoformans* and *Cryptococcus gattii.*

Composition:

Ingredients	Grams/Litre
Glycine	10.0
Potassium phosphate	1.0
Magnesium sulphate	1.0
Bromothymol Blue	0.08
L-Canavanine Sulfate	0.03
Thiamine Hydrochloride	0.001
Agar	20.0

Final pH 5.8 +/- 0.2 at 25°C

Store granulated media between 10-30°C in tightly closed container and the prepared medium at 20-30°C. Avoid freezing and overheating. Once opened keep powdered medium closed to avoid hydration. Use before expiry date on the label.

Appearance(color):	Light yellow to dark yellow, free flowing powder
Gel strength:	Firm, comparable with 2 % AGAR GEL
Color and Clarity:	Yellow to dark green-yellow coloured, clear to opalescent gel forms in Petri plates

Directions:
Suspend 32.11 g 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 use as desired.

Principle and Interpretation:
Cryptococcus is the cause of cryptococcosis, a systemic mycosis of humans and animals with a worldwide distribution. Cryptococcosis (earlier called European blastomycosis) commonly starts following inhalation of the organism, which is considered opportunistic infections as it affects mainly immunocompromised individuals. (1)
Cryptococcosis is typically caused by *Cryptococcus neoformans*, an encapsulated yeast found in soil, specifically in soil containing manure from avians. The increased pH and concentration of nitrogen promote the growth of this organism. The yeast cells can easily become airborne.
Glycine serves as a sole source of carbon and nitrogen which is utilized by *Cryptococcus gaitti*, *Cryptococcus laurentii* but not by *Cryptococcus neoformans*. *C. gaitti* can degrade L-canavanine to ammonia and leads to a pH shift which triggers the blue color reaction in combination with the bromothymol indicator in the medium. Potassium phosphate and magnesium sulphate provide essential ions for growth and thiamine is an important vitamin for the growth of *C. gaitti.*

Cultural characteristics observed after an incubation at 25-30°C for 5-6 days.

Organisms (ATCC)	Growth	Colony characteristics
Cyptococcus neoformans (204092)	+	Medium yellow or green colony
Cryptococcus gattii (MYA- 4560)	+++	Deep blue colony

References:

1. Misral, V.C, and Randhawa. H.S, Occurrence and Significance of Cyptococcus neoformans in Vegetables and Fruits. The Indian Journal of Chest Diseases 6 Allied Sciences. 42:317-322 (2000)
2. Hensyl, William R., et al., Stedman's Medical Dictionary, 25th ed. Williams & Wilkins, Baltimore, MD (1990)
3. St. Germain, Guy, et al., Identifying Filamentous Fungi. Star Publishing Company, Belmont, CA.
4. Larone, D.H. Medically Important Fungi: A Guide to Identification, American Society for Microbiology. Washington, D.C. (1996)
5. The Oxoid Vade-Mecum of Microbiology., Unipath Ltd., Basingstoke, UK (1993)
6. Koneman, E.W., et al. Color Atlas and Textbook of Diagnostic Microbiology, J.B. Lippincott Company, Philadelphia, PA.
7. Kwon-Chung, K. J., I. Polacheck, and J.E. Bennett, Improved Diagnostic Medium for Separation of Cryptococcus neoformans var. neoformans (serotypes A and D) and Cryptococcus neoformans var. gattii (serotypes B and C). Journal of Clinical Microbiology, 15:525-537 (1982)
8. Polacheck, I. and K. J. Kwon-Chung, Canavanine Resistance in Cryptococcus neoformans. Antimicrobial Agents and Chemotherapy; 29.3:468-473 (1985)
9. Chakrabarti, Arunaloke, et al., Isolation of Cryptococcus neoformans var. gattii from Eucalyptus camaldulensis in India. Journal of Clinical Microbiology; 35.12:3340-3342 (1997)
10. Gionfriddo, J. R., Feline Systemic Fungal Infections. Vet Clin North Am Small Anim Pract. 30(5):1029-50 (2000)
11. Gerds-Grogan S. and B. Dayrell-Hart B., Feline Cryptococcosis: A Retrospective Evaluation. J. Am. Anim. Hosp. Assoc. 33(2):118-22 (1997)
12. Malik R., D.I. Wigney, D.B. Muir, D.J. Gregory and D.N. Love, Cryptococcosis in Cats: Clinical and Mycological Assessment of 29 Cases and Evaluation of Treatment Using Orally Administered Fluconazole. J. Med. Vet. Mycol. 30(2):133-44 (1992)
13. Klein, K.R., L. Hall, S.M. Deml, J.M. Rysavy, S.L. Wohlfiel and N.L. Wengenack, Identification of Cryptococcus gattii by Use of L-Canavanine Glycine Bromothymol Blue Medium and DNA Sequencing. J. Clin. Micro. 47(11):3669-3672 (2009)

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.