Carrot Agar

M1987

Intended Use:

Recommended for sporangial production and study of mating techniques of *phytophthora* sp.

Composition**

Ingredients	Gms / Litre
Carrot, infusion from	200.000
Agar	15.000
Final pH (at 25°C)	6.5±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 19 grams in 1000 ml purified / 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. Also if desired the medium can be allowed to stand and solidify overnight. It can be reautoclaved again for 10 minutes and poured in 10 ml quantity to prepare thin carrot agar for mating studies.

Principle And Interpretation

Carrot Agar is widely used in mycological research. Different combinations of carrot and agar are known to support proliferation and sporulation of plant pathogens (4,5). It can also be used to enhance sporulation of a variety of fungi such as *Alternaria, Cercospora,* and *Thielaviopsis(*4). This medium has been recommended for studies determining mating type of *Phytophthora ramorum* isolates by mycelial mixing (1). Different isolates under study can be subcultured onto this medium until mycelial growth is seen but with no chlamydospores. Such isolates can be grown or subcultured to carry out mating studies at room temeparture (20-30°C) for 3 days as per standard protocol (1).

Carrot provides the natural nutritional contents required by fungii. It has necessary nutrients, minerals and vitamins which limits the growth of organisms, providing an environment only for the existence rather than their growth. Agar acts as a solidifying agent.

Type of specimen

Roots of plant.

Specimen Collection and Handling

For root samples follow appropriate techniques for handling specimens as per established guidelines (5).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. This medium is general purpose medium and may not support the growth of fastidious organisms.
2. Further biochemical test must be carried out for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Yellow coloured clear to slightly opalescent gel forms in Petri plates

Reaction
Reaction of 1.9% w/v aqueous solution at 25°C. pH : 6.5±0.2

pH
6.30-6.70

Cultural Response
Cultural characteristics observed after an incubation at 20-25°C for 48-72 hours.

Organism	Growth
# Aspergillus brasiliensis ATCC 16404 (00053*)	good
Phytophthora ramorum	good

Key : (*) - Corresponding WDCM numbers. (#) - Formely known as *Aspergillus niger*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use.
Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

Reference

1. Brasier CM &Kirk SA,2004) Production of gametangia by Phytophthora ramorum in vitro. Mycological Research 108: 823-827.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
4. Leslie, J.F. and Summerell, B.A. 2006. The Fusarium Laboratory Manual. Blackwell Publishing Ames, IA p. 12-13 .
5. Protocols for Susceptibility testing Protocol 9: Determination of Mating Type of *Phytophthora ramorum* Isolates by Mycelial Mixing.

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