



Jensen Seedling Agar

M718

Intended Use

Recommended for germinating seeds of leguminous plants while studying the nodulating ability of *Rhizobium* isolates.

Composition**

Ingredients	Gms / Litre
Calcium phosphate	1.000
Dipotassium hydrogen phosphate	0.200
Magnesium sulphate	0.200
Sodium chloride	0.200
Ferric chloride	0.100
Agar	15.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 16.7 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. Cool to 45-50°C. Mix well and dispense as desired.

Principle And Interpretation

Rhizobium is a soil bacterium that has great environmental and agricultural importance because of their symbiotic association with leguminous plants. They are responsible for most of the atmospheric nitrogen fixed on the earth (1). *Rhizobium* is a free-living bacterium, which grow well on a nitrogen free medium. These bacteria utilize atmospheric nitrogen gas for their cell protein synthesis. This cell protein is then mineralised in soil after the death of the cells thereby contributing towards the nitrogen availability to the crop plants (5). Jensen Seedling Agar, a nitrogen free medium, is used for germinating seeds of leguminous plants while studying the nodulating ability of *Rhizobium* species (3). Calcium stimulates nodulation when present as chloride or sulphate. Sodium chloride maintains the osmotic balance of the medium. Dipotassium phosphates provide buffering to the medium. Magnesium sulphate and ferric chloride are sources of ions that simulate metabolism.

Type of specimen

Soil sample- root system of the leguminous plant

Specimen Collection and Handling

For soil 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. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.

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 beige homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light cream coloured, clear to slightly opalescent gel with a slight precipitate.

Reaction

Reaction of 1.67% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH

6.80-7.20

Cultural Response

Cultural characteristics observed after an incubation at 30°C for 7 days.

Organism	Growth
<i>Rhizobium japonicum</i> ATCC 10324	luxuriant
<i>Rhizobium leguminosarum</i> ATCC 10004	luxuriant
<i>Rhizobium meliloti</i> ATCC 9930	luxuriant

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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,4).

Reference

1. Clemence Chaintrevil, Eric Giraud, Yves Prin et al, Appl. Environ. Microbiol., 2000, December; 66 (12): 5437 - 5447.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3. Jensen H. L., Nitrogen fixation in leguminous plants. I., Proc. Int. Soc. NSW, 1942; 66:68 - 108.
4. 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.
5. Subba Rao N. S., 1977, In: Soil Microorganisms and Plant Growth, Oxford and IBH Publishing Co., New Delhi, Pages 254-255.

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Disclaimer :

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