Phenyl Alanine Agar Slant

M281

Intended Use:

Recommended for the differentiation of *Proteus* and *Providencia* group of organisms from other members of *Enterobacteriaceae* on the basis of their ability to form phenyl pyruvic acid from phenylalanine.

Composition**

Ingredients	Gms / Litre
Yeast extract	3.000
Sodium chloride	5.000
DL-Phenylalanine	2.000
Disodium hydrogen phosphate	1.000
Agar	15.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 26.0 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Dispense in tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Allow the tubed medium to cool in slanting position.

Principle And Interpretation

The ability of *Proteus* species to convert phenylalanine to phenylpyruvic acid is an important reaction in the differentiation of *Enterobacteriaceae* (1,2). Based on this criterion, Buttiaux developed Phenylalanine Agar for differentiation of *Proteus* and *Providencia* group from other members of *Enterobacteriaceae* (3,4) by the ability of organism in the genera within *Proteus* to deaminate phenylalanine. Phenylalanine Agar is the modification of the medium originally developed by Ewing et al (5). Yeast extract in the medium supports the growth of the organisms. Sodium chloride maintains osmotic equilibrium. The phenylalanine serves as the substrate for enzymes, which are able to deaminate it to form phenylpyruvic acid. A recommended technique is to inoculate the slant surface with plenty of inoculum and incubate it for 12-16 hours. After incubation, add 0.2 ml of 10% ferric chloride solution so that the solution floods all over the growth. The addition of (0.2 ml 3-5 drops) of a 10%aqueous ferric chloride solution (or a 12% aqueous ferric chloride solution acidified with 2.5 ml of concentrated HCl per 100 ml of reagent) to the cultures following incubation results in the appearance of a light to deep green color (positive reaction) or no color change (negative reaction). In a positive reaction, any phenylpyruvic acid present will react with the ferric salt in the reagent to give a green color. Interpret the results within 5 minutes upon addition of reagent as the green colour fades quickly (2,4).

Type of specimen

Isolated Microorganisms

Specimen Collection and Handling:

A recommended technique is to inoculate the slant surface with plenty of inoculum and incubate it for 12-16 hours. After incubation, add 0.2 ml of 10% ferric chloride solution so that the solution floods all over the growth. The addition of (0.2 ml 3-5 drops) of a 10%aqueous ferric chloride solution (or a 12% aqueous ferric chloride solution acidified with 2.5 ml of concentrated HCl per 100 ml of reagent) to the cultures following incubation results in the appearance of a light to deep green color (positive reaction) or no color change (negative reaction). In a positive reaction, any phenylpyruvic acid present will react with the ferric salt in the reagent to give a green color. Interpret the results within 5 minutes upon addition of reagent as the green colour fades quickly (2,4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic use only. For professional use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations:

1.Some organism may show poor growth due to nutritional variation.
2.Other biochemical tests must be carried out in conjunction 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
Light amber coloured slightly opalescent gel forms in tubes as slants
Reaction
Reaction of 2.6% w/v aqueous solution at 25°C. pH : 7.3±0.2
pH
7.10-7.50
Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 12-16 hours

Organism	Growth	Phenylalanine deaminase
# *Klebsiella aerogenes* ATCC 13048 (00175*)	luxuriant	negative reaction
Escherichia coli ATCC 25922 (00013*)	luxuriant	negative reaction
Proteus mirabilis ATCC 25933	luxuriant	positive reaction, green colouration after addition of 10% ferric chloride
## *Proteus hauseri* ATCC 13315	luxuriant	positive reaction, green colouration after addition of 10% ferric chloride
Providencia alcalifaciens ATCC 9886	luxuriant	positive reaction, green colouration after addition of 10% ferric chloride

Key : *Corresponding WDCM numbers,## Formerly known as *Proteus vulgaris*
Formerly known as *Enterobacter aerogenes*

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 (6,7).

Reference

1. Henrikson S. D., 1950, J. Bacteriol., 60:225.
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5. Ewing W. H., Davis B. R. and Reavis R. W., 1957, Public Health Lab., 15:153.
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook2nd Edition.
7. 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.

Revision : 04/ 2024

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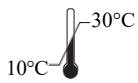
IVD ***In vitro*** **diagnostic medical device**



Storage temperature

EC REP

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CE Marking

Do not use if package is damaged

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