

Technical Data

Andrade Peptone Water w/HM Extract

Intended Use

Recommended as a basal medium to which various carbohydrates can be added to study fermentation reactions, particularly of members of the *Enterobacteriaceae*.

Composition**

Ingredients	g / L
Peptone	10.000
HM extract#	3.000
Sodium chloride	5.000
Andrade indicator	0.100
Final pH (at 25°C)	7.1±0.2
**Formula adjusted, standardized to suit performance parameters	
# Equivalent to Meat extract	

Directions

Suspend 18.1 grams in 1000 ml purified/distilled water. Heat if necessary to dissolve the medium completely and dispense in test tubes containing inverted Durham's tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to room temperature and aseptically add sterile stock solution of carbohydrate to a final concentration of 0.5% to 1.0% (w/v).

Principle And Interpretation

Bacteria differ widely in their ability to metabolize carbohydrates and related compounds. Carbohydrate fermentation reactions aids in the differentiation and identification of various bacteria. Andrade Peptone Water w/HM Extract is the most commonly used media for carbohydrate fermentation (1).

Peptone used is in the medium is free from fermentable carbohydrates (1,2) and the medium is also free from nitrates which may interfere with gas production. HM extract is an additional source of nutrients. Andrade indicator is a solution of acid fuchsin which when titrated with sodium hydroxide; changes colour from pink to yellow. The Andrade indicator changes colour from yellow to pink as the pH decreases (1). The medium is pink when hot but becomes straw coloured on cooling. Test carbohydrate solutions should be sterilized separately and aseptically added to sterile Andrade Peptone Water w/HM Extract. The biochemical identification of organisms capable of growing in this medium is made by various sugar fermentation results (1,3,4).

Use fresh cultures of organisms only which have been presumptively identified by Gram staining and colony morphology. For final identification further biochemical tests are required. Desired carbohydrate is added to the medium, which is inoculated with the test organism. If the test organism metabolizes the added carbohydrate, acids are produced, thereby lowering the pH of the medium. This causes a subsequent colour change of the indicator, from colourless to pink to red. If the added carbohydrate is not metabolized, the medium remains pale tan to straw coloured. Gas produced during fermentation is collected in the Durhams tube.

Type of specimen

Clinical samples - faeces, urine, sputum and wound exudates; Pure isolate

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5,6). 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.Use fresh cultures of organisms only which have been presumptively identified by Gram staining and colony morphology.

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2.Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.2.Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.3.For final identification further biochemical tests are required.

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 coloured with pink tinge homogeneous free flowing powder

Colour and Clarity of prepared medium

Light pink to straw coloured clear solution without any precipitate

Reaction

Reaction of 1.81% w/v aqueous solution at 25°C. pH : 7.1±0.2

pН

6.90-7.30

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Growth	Acid in absence of dextrose	Gas in absence of dextrose	e Acid with added dextrose	Gas with added dextrose
Escherichia coli ATCC 25922 (00013*)	luxuriant	negative reaction	negative reaction	positive reaction,colour changes to pink-red	positive reaction
Klebsiella pneumoniae ATCC 13883 (00097*)	luxuriant	negative reaction	negative reaction	positive reaction, colour changes to pink-red	positive r reaction
## Proteus hauseri ATCC 13315	luxuriant	negative reaction	negative reaction	positive reaction	positive reaction
<i>Salmonella</i> Typhi ATCC 6539	luxuriant	negative reaction	negative reaction	positive reaction	negative reaction
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	luxuriant	negative reaction	negative reaction	positive reaction	positive reaction
Shigella flexneri ATCC 12022 (00126*)	luxuriant	negative reaction	negative reaction	positive reaction	negative reaction
Shigella sonnei ATCC 25931	luxuriant	negative reaction	negative reaction	positive reaction	

Key : (*) Corresponding WDCM numbers. ## Formerly known as *Proteus vulgaris*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-30°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 (5,6).

Reference

1. Cowan S. T. and Steel K. J., 1974, Manual of Identification of Medical Bacteria, 2nd Ed., Cambridge United Press.

2.MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.

3.Finegold S. M. and Baron E. J., 1986, Bailey and Scotts Diagnostic Microbiology, 7th Ed., The C.V. Mosby Co., St. Louis.

4.Murray P. R., Baron E. J., Jorgensen J. H., Pfaller M. A., Yolken R.H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.

5. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.

6.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.

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