ONPG Broth

M1930

Intended Use:

Recommended for the differentiation of microorganisms on the basis of beta-galactosidase activity.

Composition**

Ingredients	g / L
Casitose ▲	7.500
Disodium hydrogen phosphate	0.350
O-Nitrophenyl-β-D-galactopyranoside	1.500
Sodium chloride	3.750
Final pH (at 25°C)	7.5±0.2

**Formula adjusted, standardized to suit performance parameters

▲- Equivalent to Casein Peptone

Directions

Suspend 13.10 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. **DO NOT AUTOCLAVE**. Dispense into tubes or flasks as desired .

Principle And Interpretation

ONPG (Ortho-nitrophenyl beta-D-galactopyranoside) is a synthetic colourless compound (galactoside) structurally similar to lactose(1). β-galactosidase positive organisms cleaves ONPG to galactose and o-nitrophenyl, a yellow compound. This test is specially useful in the rapid identification of cryptic lactose fermenters (late fermenters). Since members of family *Enterobacteriaceae* are routinely grouped according to their lactose fermenting ability the ONPG test is significant here.

ONPG Broth is used for differentiation of organisms based on their ability to utilize ONPG, which is similar in structure to lactose. The presence of two enzymes is required to demonstrate lactose fermentation in a conventional test. The first enzyme permease, facilitates the entry of lactose molecules into the bacterial cell while the second enzyme, β-galactosidase, hydrolyzes the lactose to yield glucose and galactose. True non-lactose fermenters lack both enzymes; however some organisms lack permease but posseses β-galactosidase. These organisms are late lactose fermenters.

Casitose serves as a source of nitrogeneous compounds, long chain amino acids and other growth factors. Phosphate buffers the medium. Sodium chloride maintains the osmotic balance. Ortho-nitrophenyl beta-D-galactopyranoside serves as a substrate for the presence of β-galactosidase enzyme.

Type of specimen

Isolated Microorganism from clinical and non-clinical samples

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (2,3).

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 organisms may show delay in lactose fermentation thus, need further incubation.
2. Other biochemical test must be performed 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

Colour and Clarity of prepared medium

Light amber coloured clear solution in tubes

Reaction

Reaction of 1.3% w/v aqueous solution at 25°C. pH : 7.5±0.2

pH

7.30-7.70

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Growth	ONPG test
Salmonella Choleraesuis ATCC 12011	luxuriant	Positive reaction: yellow colour
Citrobacter freundii ATCC 8090	luxuriant	Positive reaction: yellow colour
# *Klebsiella aerogenes* ATCC 13048 (00175*)	luxuriant	Positive reaction: yellow colour
Escherichia coli ATCC 25922 (00013*)	luxuriant	Positive reaction: yellow colour
## *Proteus hauseri* ATCC 13315	luxuriant	Negative reaction, no colour change
Salmonella Typhimurium ATCC 14028 (00031*)	luxuriant	Negative reaction, no colour change

Key : (*) Corresponding WDCM numbers, (#) Formerly known as *Enterobacter aerogenes*

Formerly known as *Proteus vulgaris*

Storage and Shelf Life

Store dehydrated and prepared medium at 2-8°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.Biochemical tests for Identification of Medical Bacteria, 3rd Edition, Jean F. MacFaddin.

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.

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

2°C 8°C **Storage temperature**

EC REP

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CE Marking

Do not use if package is damaged

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