Bile Esculin Discs

DD024

Bile Esculin Discs are used for detection of esculin hydrolysis in the presence of bile, for differentiating Group D streptococci from other Streptococcal groups.

Directions

Esculin impregnated disc is placed on the seeded Bile Esculin Agar Base (M340) plate and is incubated at 35-37°C for 18-24 hours.

Principle And Interpretation

Group D streptococci hydrolyze esculin to esculetin and dextrose. Esculetin reacts with an iron salt such as ferric citrate to form a blackish brown coloured complex (4).

Rochaix found that esculin hydrolysis is an important criteria in the identification of enterococci (1). Meyer and Schonfeld (2) observed that when bile was added to esculin medium, around 60% enterococci were able to grow and split the esculin while other streptococci could not. When a comparative study was performed by Facklam and Moody (3) for presumptive identification of Group D streptococci, they found the bile esculin test as a reliable means of identifying Group D streptococci and differentiating them from other streptococci groups.

Quality Control

Appearance

Plain filter paper discs of 6mm diameter

Cultural response

Cultural response observed by placing Bile Esculin disc (DD024) on seeded Bile Esculin Agar Base(M340) plate, incubated at 35-37°C for 18-24 hours.

Organism	Growth	Esculin hydrolysis
Enterococcus faecalis ATCC 29212	luxuriant	positive: blackening of media around the disc.
Streptococcus agalactiae ATCC 13813	luxuriant	negative: no blackening
Listeria monocytogenes ATCC 19118	luxuriant	positive: blackening of media around the disc.
Streptococcus pyogenes ATCC 19615	luxuriant	negative: no blackening

Storage and Shelf Life

Store at 2 - 8°C. Use before expiry date on the label.

Reference

1. Rochaix, 1924, C. R. Soc. Biol., 90:771.
2. Meyer and Schonfeld, 1926, Zentralbl. Bacteriol. Parasitenkd. Infectionskr. Hyg. Abt. I Orig., 99:402.
3. Facklam and Moody, 1970, Appl. Microbiol., 20:245.
4.MacFaddin J. F., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd ed., Philadelphia: Lippincott Williams and Wilkins.

Revision : 1 / 2011

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CC Selective Supplement I

FD010

An antibiotic supplement recommended for the selective isolation of *Clostridium difficile*.

Composition

Per vial sufficient for 500 ml medium

*Ingredients	Concentration
D-Cycloserine	250mg
Cefoxitin	8mg

Directions:

Rehydrate the contents of one vial aseptically with 2 ml sterile distilled water. Mix well and aseptically add along with 7% v/v defibrinated horse blood to 500 ml sterile, molten, cooled (45-50°C) Clostridium Difficile Agar Base M836 / Clostridium Difficile HiVeg™ Agar Base MV836 / Clostridium Brazier Agar Base M1803. Mix well and pour into sterile petri plates. Sheep blood may be used in place of horse blood but some strains of the organism will show a slightly reduced growth.

Type of specimen

Clinical samples : stool, abscess, etc.; Food samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,2). For Food samples follow appropriate techniques for handling specimens as per established guidelines (3). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning & Precautions

In Vitro diagnostic use. 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.

Storage and Shelf Life

Store at 2 - 8°C. Use before expiry date on the label.

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 (1,2).

Reference

1. Isenberg (Ed.),2004, Clinical Microbiology Procedures Handbook, Vol.3, American Society for Microbiology, Washington. D.C.
2. 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.
3. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

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Revision : 03/2022

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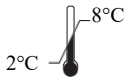
IVD ***In vitro*** **diagnostic medical device**

CE Marking



Storage temperature

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V.C.N. Supplement

FD023

An antibiotic supplement, recommended for the selective isolation of *Neisseria gonorrhoeae* and *Neisseria meningitidis*.

Composition

Per vial sufficient for 500 ml medium

*Ingredients	Concentration
Vancomycin	1.500mg
Colistin methane sulphonate	3.750mg
Nystatin	6250Units

Directions:

Rehydrate the contents of 1 vial aseptically with 2 ml sterile distilled water. Mix well and aseptically add it to Thayer Martin Medium Base M413 / Thayer Martin HiVeg™ Medium Base MV413- for 440 ml of medium aseptically add 50ml sterile lysed blood and one vial of V.C.N. Supplement FD023 along with one vial of Vitamino Growth Supplement FD025. FO Growth Supplement (250ml) FD022 can be used instead of sterile lysed blood in 250ml of medium. In GC Agar Base M434/ GC HiVeg™ Agar Base MV434 for 250 ml of can be used instead of sterile lysed blood in 250 ml of FO Growth Supplement FD022 and GC Selective Supplement FD021, one vial of GC Selective Supplement FD021 for additional selectivity. If desired V.C.N. Supplement FD023 can be used along with GC Selective Supplement FD021 for additional selectivity.

In Transgrow Medium Base M1149 for 440 ml of medium aseptically add 50 ml of sterile FO Growth Supplement FD022 and one vial of V.C.N. Supplement FD023 along with one vial of Vitamino Growth Supplement FD025.

Type of specimen

Clinical samples - Stool, urine, respiratory exudates, etc.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,2). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning & 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.

Storage and Shelf Life

Store at 2-8°C. Use before expiry date on the label.

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 (1,2).

Reference

1.Isenberg (Ed.),2004, Clinical Microbiology Procedures Handbook, Vol.3, American Society for Microbiology, Washington. D.C.

2.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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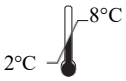

Storage temperature

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Technical Data

MeRS Selective Supplement

FD029

An antibiotic supplement recommended for the selective isolation of *Pseudomonas* species.

Composition

Per vial sufficient for 500 ml medium

*Ingredients	Concentration
Cetrimide	100mg
Nalidixic acid	7.500mg

Directions:

Rehydrate the contents of 1 vial aseptically with 2 ml of sterile distilled water. Mix well and aseptically add it to 500 ml of sterile, molten, cooled (45-50°C) Pseudomonas Agar Base M085 / Pseudomonas HiVeg™ Agar Base MV085. Pseudomonas Agar Base, Granulated GM085. Mix well and pour into sterile petri plates.

Type of specimen

Clinical samples - pus, urine, body fluids, etc; Water samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,2).
For water samples follow appropriate techniques for handling specimens as per established guidelines (3).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning & Precautions

In Vitro diagnostic use. 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.

Storage and Shelf Life

Store at 2 - 8°C. Use before expiry date on the label.

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 (1,2).

Reference

1. Isenberg (Ed.),2004, Clinical Microbiology Procedures Handbook, Vol.3, American Society for Microbiology, Washington. D.C.
2. 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.
3. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

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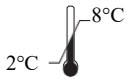

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Technical Data

KL Virulence Enrichment (20 ml per vial)

FD072

Recommended for cultivation and in vitro toxicity testing of *Corynebacterium diphtheria*.

Composition

Per vial sufficient for 100 ml medium

Ingredients	Concentration
Acicase™#	1.00g
Glycerol	1.00ml
Polysorbate 80	1.00ml

Equivalent to Casein acid hydrolysate

Directions:

Warm up the refrigerated contents of 1 vial to 50°C and aseptically add 2 ml in 100 mm sterile petri plate along with 0.5 ml of 1% PTe Selective Supplement FD052. Quickly add 10 ml sterile molten, cooled (45-50°C) Diphtheria Virulence Agar Base M882/ Diphtheria Virulence HiVeg™ Agar Base MV882 . Mix well and pour into sterile Petri plate.

Type of specimen

Clinical samples- Throat swab, nasal swab, wound swab, pus, etc.; Food samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,2).
For food samples follow appropriate techniques for handling specimens as per established guidelines (3).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning & Precautions

In Vitro diagnostic use. 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.

Storage and Shelf Life

Store at 2 - 8°C. Use before expiry date on the label.

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 (1,2).

Reference

1. Isenberg (Ed.),2004, Clinical Microbiology Procedures Handbook, Vol.3, American Society for Microbiology, Washington. D.C.
2. 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.
3. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

*** Not For Medicinal Use**

Revision : 03/2023

HiIndicator™ pH papers **LA310, LA312, LA315, LA318, LA321, LA323, LA334, LA335.**

The convenience of using HiIndicator papers for the rapid determination of pH values has led to many applications in laboratories and industry. These pH papers are made with special indicator dyes that change color at specified pH value.

Somewhat uneven colour of the strips is of no consequence. The colour obtained on use is indicative of the correct pH.

Application : Analytical chemistry, biology & various laboratories and industries etc.

Product Name	Product Code	Description	pH Range
HiIndicator™ pH papers.	**LA310**	HiIndicator pH paper	2.00 – 10.50
	LA312	HiIndicator pH paper	3.50 – 6.00
	LA315	HiIndicator pH paper	3.80 – 5.30
	LA318	HiIndicator pH paper	5.00 – 7.50
	LA321	HiIndicator pH paper	6.50 – 9.00
	LA323	HiIndicator pH paper	8.00 – 10.50
	LA334	HiIndicator pH paper	2.00 - 4.50
	LA335	HiIndicator pH paper	1.00 - 14.00

Direction for use : Tear off strip of indicator paper and insert it for a few seconds into the solution to be tested. With highly viscous or stained liquids and with suspensions, drip the substance onto the indicator paper. Compare the wet paper with the colour scale. For papers where liquids are dripped, compare the reverse side. Possible discolouration of the dry new papers may be caused by their high sensitivity. This does not impair the efficacy of the Indicator papers for pH determinations.

The so-called indicator error may occur with very weakly buffered or unbuffered solution and can be compensated for up to a point in the following manner. : - The strip can be made to adhere to the inner wall of the a test tube, which can then filled to the upper edge of the paper with the fluid to be tested. After 1/2 to 1 minute, the colour of the paper may be compared with the scale through the glass of test tube.

Please refer disclaimer Overleaf

Product Features:

- Instant pH readings.
- Accurate for a wide range of routine pH testing.
- Convenient and portable for field use.
- Pack Size : 1 pack-200 Nos.

Nutrient Agar

M001

Intended use

Nutrient Agar is used as a general purpose medium for the cultivation of less fastidious microorganisms, can be enriched with blood or other biological fluids.

Composition**

Ingredients	Gms / Litre
Peptone	5.000
Sodium chloride	5.000
HM peptone B#	1.500
Yeast extract	1.500
Agar	15.000
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

- Equivalent to Beef extract

Directions

Suspend 28.0 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. If desired ,the medium can be enriched with 5-10% blood or other biological fluids. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Nutrient media are basic culture media used for maintaining microorganisms, cultivating fastidious organisms by enriching with serum or blood and are also used for purity checking prior to biochemical or serological testing (1,2). Nutrient Agar is ideal for demonstration and teaching purposes where a more prolonged survival of cultures at ambient temperature is often required without risk of overgrowth that can occur with more nutritious substrate. This relatively simple formula has been retained and is still widely used in the microbiological examination of variety of materials and is also recommended by standard methods. It is one of the several non-selective media useful in routine cultivation of microorganisms (3,4). It can be used for the cultivation and enumeration of bacteria which are not particularly fastidious. Addition of different biological fluids such as horse or sheep blood, serum, egg yolk etc. makes it suitable for the cultivation of related fastidious organisms. Peptone, HM peptone B and yeast extract provide the necessary nitrogen compounds, carbon, vitamins and also some trace ingredients necessary for the growth of bacteria. Sodium chloride maintains the osmotic equilibrium of the medium.

Type of specimen

Clinical samples - faeces, urine ; Food and dairy samples; Water samples

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5,6).
For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3,4,7).
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (8).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic use. 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. 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.

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 yellow coloured clear to slightly opalescent gel forms in Petri plates

Reaction
Reaction of 2.8% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH
7.20-7.60

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

Organism	Inoculum (CFU)	Growth	Recovery
Escherichia coli ATCC 25922 (00013*)	50-100	good-luxuriant	>=70%
Pseudomonas aeruginosa ATCC 27853 (00025*)	50-100	good-luxuriant	>=70%
Salmonella Typhi ATCC 6539	50-100	good-luxuriant	>=70%
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	50-100	good-luxuriant	>=70%
Streptococcus pyogenes ATCC 19615	50-100	good-luxuriant	>=70%
Salmonella Enteritidis ATCC 13076 (00030*)	50-100	good-luxuriant	>=70%
Salmonella Typhimurium ATCC 14028 (00031*)	50-100	good-luxuriant	>=70%
Yersinia enterocolitica ATCC 9610 (00038*)	50-100	good-luxuriant	>=70%
Yersinia enterocolitica ATCC 23715 (00160*)	50-100	good-luxuriant	>=70%

Key : (*) Corresponding WDCM numbers.

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 (5,6).

Reference

1.Lapage S., Shelton J. and Mitchell T., 1970, Methods in Microbiology', Norris J. and Ribbons D., (Eds.), Vol. 3A, Academic Press, London.
2.MacFaddin J. F., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd Ed., Lippincott, Williams and Wilkins,Baltimore.
3.American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
4.Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, 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.
7.Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed.,APHA Inc., Washington, D.C.
8.Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Revision : 06/2022

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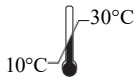
IVD ***In vitro* diagnostic medical device**



Storage temperature

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Bile Broth Base

M071

Intended Use:

Recommended for cultivation of members of the *Enterobacteriaceae* and in culture of blood clots from patients with suspected enteric fever.

Composition**

Ingredients	Gms / Litre
Peptone	20.000
Sodium taurocholate	5.000
Sodium chloride	5.000
Final pH (at 25°C)	7.6±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 30.0 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and add 1 ml of Streptokinase solution (100000 units/ml). Mix well and dispense into sterile tubes or flasks as desired.

Principle And Interpretation

Enterobacteriaceae inhabit a wide variety of niches that include the human gastrointestinal tract and various environmental niches. When blood samples from a patient with suspected enteric fever is submitted for the widal test, it is useful as a routine to culture the clot after separation of serum (1). If it is known that the blood has been withdrawn with strict aseptic precautions, the clot may be placed in a wide tube half-filled with broth, or in a wide mouth screw-capped bottle containing 80 ml of broth. When there is any doubt regarding the presence of contaminating organisms, and this is always a possibility when blood specimens are sent to the laboratory from a distance, the clot should be transferred directly to a tube of sterile ox bile and disintegrated with aseptic precautions. After overnight incubation the bile culture is examined for enteric organism in the usual manner. A method of clot culture with Streptokinase has been recommended (4). Blood is allowed to clot in 5 ml quantities in sterile screw-capped universal containers. The separated serum is removed and 15 ml of 0.5% Bile Broth Base with Streptokinase 100 units/ml is added to each bottle. The streptokinase causes rapid clot lysis with release of bacteria trapped in the clot (4)

Peptone serves as a source of nitrogen, carbon, long chain amio acids and other essential amino acids. Sodium taurocholate inhibits majority of Gram-positive species. Sodium chloride maintains the isotonicity of the medium whereas addition of streptokinase solution causes rapid clot lysis with release of bacteria trapped in the clot (4).

Type of specimen

Clinical samples - Blood clot

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. 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. Further biochemical and serological tests must be carried out for further identification.

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

Yellow coloured, clear solution without any haziness

Reaction

Reaction of 3.0% w/v aqueous solution at 25°C. pH : 7.6±0.2

pH

7.40-7.80

Cultural Response

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

Organism	Inoculum (CFU)	Growth
Escherichia coli ATCC 25922 (00013*)	50-100	luxuriant
# *Klebsiella aerogenes ATCC 13048* (00175*)	50-100	luxuriant
Salmonella Typhi *ATCC 6539*	50-100	luxuriant
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	>=10^4	inhibited

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

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the 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 inorder 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. Colle, J.G., Duguid J.P., Fraser A.G. and Marmion, B.P. (Eds.) 1989 Mackie and McCartney Practical Medical Microbiology, Vol. 2, p:134 Longman Group, UK.

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

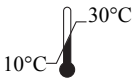
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

4. Watson, K.C. 1955, Isolation of *Salmonella* Typhi from the blood stream. J. of ,Lab and Clinical Medicine 46:128-134.

Revision : 04/ 2019

IVD — In vitro diagnostic medical device

CE Marking



Storage temperature

Do not use if package is damaged

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MacConkey Broth Purple w/ BCP

M083I

Intended Use:

Recommended for presumptive identification of coliforms from water. The composition and performance criteria of this medium are as per the specifications laid down in ISO 9308-2:2012 & ISO 4832:2006.

Composition**

Ingredients	Gms / Litre
Peptone	20.000
Lactose	10.000
Bile salts	5.000
Sodium chloride	5.000
Bromocresol purple	0.010
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 40.01 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into test tubes with inverted Durham tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the tubes to 45-50°C before inoculation.

Principle And Interpretation

MacConkey Broth Purple w/ BCP is a modification of MacConkey Medium (6). Childs and Allen (2) demonstrated the inhibitory effect of neutral red and therefore substituted it by the less inhibitory bromocresol purple dye. BCP is more sensitive in recording pH variation in the medium. MacConkey Broth Purple w/ BCP is recommended by ISO committee (3) with the inclusion of bile salts, as a presumptive test medium for identification of coliforms from water and other materials of sanitary importance.

Peptone provides essential growth nutrients. Lactose is the fermentable carbohydrate. Bile salts or sodium taurocholate inhibits gram-positive organisms. Sodium chloride maintains the osmotic balance of the medium. Bromocresol purple is the pH indicator in the medium, which turns yellow under acidic condition. Lactose fermenting organisms turn the medium yellow due to the acidity produced on lactose fermentation. The colour change of the dye is observed when the pH of the medium falls below 6.8. Lactose non-fermenting organisms like *Salmonella* and *Shigella* do not alter the appearance of the medium. Liquid specimens are directly inoculated while solids have to be homogenized in appropriate diluents such as physiological saline, phosphate buffers, etc. If the inoculum is greater than 1 ml, it is necessary to use the medium at double strength, inoculating equal volumes of specimen and medium.

Type of specimen

Water samples

Specimen Collection and Handling:

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(1) 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. Liquid specimens are directly inoculated while solids have to be homogenized in appropriate diluents such as physiological saline, phosphate buffers, etc.
2. If the inoculum is greater than 1 ml, it is necessary to use the medium at double strength, inoculating equal volumes of specimen and 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 yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Purple coloured clear to slightly opalescent solution in tubes

Reaction

Reaction of 3.45% w/v aqueous solution at 25°C.pH:-7.4±0.2

pH

7.20-7.60

Cultural Response

Growth Promotion is carried out in accordance with the harmonized method of IP. For organisms not specified in pharmacopoeia, cultural response was observed after an incubation at 30-35°C for 18-48 hours.

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating <=100 cfu (at 42-44°C for 24 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temperature for not less than longest period of time specified inoculating >=100cfu(at 42-44°C for >= 48 hours).

Organism	Inoculum (CFU)	Growth	Acid	Gas	Incubation temperature	Incubation period
Escherichia coli ATCC 8739 (00012*)	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	42 -44 °C	<=24 hrs
Inhibitory						
Staphylococcus aureus subsp. aureus ATCC 6538 (00032*)	>=10^4	inhibited			42 -44 °C	>=48 hrs
Additional Microbiological testing						
Escherichia coli ATCC 25922 (00013*)	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
Escherichia coli NCTC 9002	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
# Klebsiella aerogenes ATCC 13048 (00175*)	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
Salmonella Choleraesuis ATCC 12011	50 -100	fair-good	negative reaction	negative reaction	30 -35 °C	18 -24 hrs
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	>=10^4	inhibited			30 -35 °C	>=48 hrs

Key : *Corresponding WDCM numbers.

\# Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°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 (4,5).

Reference

1. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
2. Childs E. and Allen, 1953, J. Hyg: Camb. 51:468-477.
3. International Organization for Standardization (ISO), 1990, Draft ISO/ DIS 9308-2.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
5. 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.
6. MacConkey A. T., 1900, The Lancet, ii: 20.

Revision : 02 / 2019

Pseudomonas Agar Base

M085

Intended use :

For selective isolation of *Pseudomonas* species.

Composition**

Ingredients	Gms / Litre
Tryptone	10.000
Gelatin peptone	16.000
Potassium sulphate	10.000
Magnesium chloride, anhydrous	1.400
Agar	11.000
Final pH (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 24.2 grams in 500 ml purified/distilled water containing 5 ml glycerol. 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 and aseptically add sterile rehydrated contents of either CetriNix Supplement (FD029) or CFC Supplement (FD036) as desired. Mix well and pour into sterile Petri plates. *Note : Do not keep the molten agar for longer than 4 hours.*

Principle And Interpretation

Pseudomonas Agar Base is a modification of Kings A medium (1) which contains magnesium chloride and potassium sulphate to enhance pigment production. Goto and Enomoto (2) formulated CetriNix supplement for the selective isolation of *Pseudomonas aeruginosa* from clinical specimens. Lowbury and Collins (3) studied cetrimide as a selective agent. CetriNix supplement suppresses *Klebsiella, Proteus* and *Providencia* species.

Tryptone and gelatin peptone supplies nitrogenous and carbonaceous compounds, long chain amino acids, and other essential growth nutrients.

C-F-C Supplement was formulated by Mead and Adams (4) making the medium specific for isolation of *Pseudomonas* from chilled foods and processing plants, environmental samples and water. This medium is recommended for enumeration of *Pseudomonas* species from meat and meat products. It can also be used for clinical samples.

Examine inoculated plates after 24 hours and 48 hours using both white and UV light. The presence of blue-green or brown pigmentation may be considered as presumptive evidence of *Pseudomonas aeruginosa. Alteromonas* species may form brown or pink colonies on the medium.

Type of specimen

Clinical samples - pus, urine, body fluids, Food samples; Water samples.

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5,6).

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (7).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (8).

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. 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.Further biochemical and serological tests 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

Gelling

Firm, comparable with 1.1% Agar gel.

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 4.84% w/v aqueous solution containing 1% v/v glycerol at 25°C. pH : 7.1±0.2

pH

6.90-7.30

Cultural Response

Cultural characteristics observed after an incubation for 40-48 hours. Recovery rate is considered as 100% for growth on Soyabean Casein Digest Agar

Organisms	Inoculum (CFU)	Growth (at 34-38°C with FD029)	Recovery 34-38°C with FD029)	Growth (at 24-26°C with FD036)	Recovery (at 24-26°C with FD036)	Colour/ Fluorescence under uv
Proteus vulgaris ATCC 13315	>=10^4	inhibited	0%	-	-	-
Pseudomonas aeruginosa ATCC 27853 (00025*)	50-100	good-luxuriant	>=50%	-	-	blue-green /positive
Pseudomonas aeruginosa ATCC 9027 (00026*)	50-100	good-luxuriant	>=50%	-	-	blue-green /positive
Pseudomonas aeruginosa ATCC 10145 (00024*)	50-100	good-luxuriant	>=50%	-	-	blue-green /positive
Pseudomonas cepacia ATCC 10661	50-100	-	-	good-luxuriant	>=50%	
Pseudomonas fluorescens ATCC 13525 (00115*	50-100	-	-	good-luxuriant	>=50%	
Pseudomonas fragi ATCC 4973 (00116*	50-100	-	-	good-luxuriant	>=50%	
Enterococcus faecalis ATCC 29212 (00087*	>=10^4	inhibited	0%	-	-	-
Enterococcus faecalis ATCC 19433 (00009*	>=10^4	inhibited	0%	-	-	-
Escherichia coli ATCC 25922 (00013*)	>=10^4	inhibited	0%	inhibited	0%	
Escherichia coli ATCC 8739 (00012*)	>=10^4	inhibited	0%	inhibited	0%	

Key : * - Corresponding WDCM numbers

Storage and Shelf Life

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

References

1.King E.O., Ward M.K. and Raney D.E., 1954, J.Lab and Clin. Med., 44:301.

2.Goto S. and Entomoto S., 1970, Jap. J. Microbiol., 14:65.

3.Lowbury E.J. and Collins A.G., 1955, Clin. Path., 8:47.

4.Mead G.C. and Adams B.W., 1977, Br. Poult. Sci., 18:661.

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.

7.Salfinger Y., and Tortorello M.L. , 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

8.Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Revision : 07/2023

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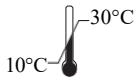
IVD ***In vitro* diagnostic medical device**



Storage temperature

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Brilliant Green Bile Broth 2%

M121

Intended Use:

Recommended for detection and confirmation of coliform bacteria in water, waste water, food, milk and dairy products.

Composition**

Ingredients	Gms / Litre
Peptone	10.000
Lactose	10.000
Bile#	20.000
Brilliant green	0.0133
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

- Equivalent to Oxgall

Directions

Suspend 40.01 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Distribute in fermentation tubes containing inverted Durhams tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates. For preparation of double strength it is recommended to heat the dissolved broth (80.02 grams per litre) at 100°C for 30 minutes.

Principle And Interpretation

Brilliant Green Bile Broth 2% is one of the most widely used medium for the detection of coliform bacteria in water, wastewater, foods, and milk and dairy products. This medium is formulated as per APHA (1,2,3) for the presumptive identification and confirmation of coliform bacteria (4,5). This medium is also recommended by the ISO Committee for enumeration of coliforms by most probable number technique (6).

Peptone serves as a source of essential nutrients. Lactose is the fermentable carbohydrate. Bile inhibits gram-positive bacteria whereas the gram-negative bacteria are inhibited by brilliant green. Production of gas from lactose fermentation is detected by incorporating inverted Durham's tube, which indicates the positive evidence of faecal coliform since non faecal coliforms growing in this medium do not produce gas. Further gas production in EC broth (M127) at 45°C used as a confirmation of faecal coliform. Gram-positive spore formers may produce gas if the bile or brilliant green inhibition is weakened by reaction with food material.

During examination of water samples, growth from presumptive positive tubes showing gas in Lactose Broth (M026) or Lauryl Tryptose Broth (M080) is inoculated in Brilliant Green Bile Broth 2% (M121). Gas formation within 48 ± 2 hours confirms the presumptive test (1).

Type of specimen

Food and dairy samples; Water samples

Specimen Collection and Handling:

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3,7,8). For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (9). 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. Do not autoclave double-strength broth.
2. Gram-positive sporing organisms may produce gas if the bile/brilliant green inhibition is attenuated by foodmaterial.

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 pale green homogeneous free flowing powder
Colour and Clarity of prepared medium
Emerald green coloured, clear solution without any precipitate.
Reaction
Reaction of 4.0% w/v aqueous solution at 25°C. pH : 7.2±0.2
pH
7.00-7.40
Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Gas
Bacillus cereus ATCC 10876	>=10^4	inhibited	
Escherichia coli ATCC 25922 (00013*)	50-100	good-luxuriant	positive reaction
# *Klebsiella aerogenes* ATCC 13048 (00175*)	50-100	good-luxuriant	positive reaction
Enterococcus faecalis ATCC 29212 (00087*)	50-100	none-poor	negative reaction
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	>=10^4	inhibited	

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

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°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 (10,11).

Reference

1. Greenberg A. E., Eaton A. D. and Clesceri L. S., (Eds.), 1998, Standard Methods for the Examination of Water and Wastewater, 20th ed., APHA, Washington, D.C.
2. Richardson G., (Ed.), 1985, Standard Methods for the Examination of Dairy Products, 15th Ed, APHA, Washington, D.C.
3. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
4. McCrady and Langerin, 1932, J. Dairy Science, 15:321.
5. McCrady, 1937, Am. J. Publ. Health, 27:1243.
6. International Organization for Standardization (ISO), 1991, Draft ISO/DIS 4831.
7. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
8. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
9. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
10. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
11. 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 : 04/2022

Malonate Broth

M382

Malonate Broth is recommended for the differentiation of *Enterobacter* and *Escherichia* on the basis of malonate utilization.

Composition**

Ingredients	Gms / Litre
Ammonium sulphate	2.000
Dipotassium phosphate	0.600
Monopotassium phosphate	0.400
Sodium chloride	2.000
Sodium malonate	3.000
Bromothymol blue	0.025
Final pH (at 25°C)	6.7±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Dissolve 8.02 grams in 1000 ml distilled water. Dispense and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Avoid the addition of carbon and nitrogen from other sources.

Principle And Interpretation

Leifson developed a synthetic liquid medium, which differentiated *Aerobacter* (now *Enterobacter*) from *Escherichia* species based on their ability to utilize malonate (1) where *Enterobacter* utilizes malonate and *Escherichia* does not.

An organism that can simultaneously utilize sodium malonate as its carbon source and ammonium sulfate as its nitrogen source produces alkalinity due to the formation of sodium hydroxide (2). The alkali changes the color of the bromothymol blue indicator in the medium to light blue and finally to prussian blue. The color of the medium remains unchanged in the presence of an organism that cannot utilize these substances. Also some malonate-positive organisms produce only a slight alkalinity that causes the results to be difficult to interpret. Therefore these tubes should be compared with an un-inoculated malonate tube (2).

Quality Control

Appearance
Light yellow to light green homogeneous free flowing powder

Colour and Clarity of prepared medium
Bluish green coloured clear solution without any precipitate

Reaction
Reaction of 0.8% w/v aqueous solution at 25°C. pH : 6.7±0.2

pH
6.50-6.90

Cultural Response
M382: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours .

Organism	Inoculum (CFU)	Growth	Malonate Utilization
Enterobacter aerogenes ATCC 13048	50-100	luxuriant	positive reaction, dark blue colour
Escherichia coli ATCC 25922	50-100	poor-fair	negative reaction
Klebsiella pneumoniae ATCC 13883	50-100	luxuriant	positive reaction, dark blue colour

Salmonella Arizonae ATCC 13314	50-100	luxuriant	positive reaction, dark blue colour
Salmonella Typhimurium ATCC 14028	50-100	fair-good	negative reaction

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1. Leifson, 1933, J. Bact., 25:329.
2. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore

Revision : 2 / 2015

Decarboxylase Broth Base, Moeller(Moeller Decarboxylase Broth Base)

M393

Intended Use:

Recommended to differentiate bacteria on the basis of their ability to decarboxylate the amino acids.

Composition**

Ingredients	Gms / Litre
Peptone	5.000
HM peptone B	5.000
Dextrose (Glucose)	0.500
Bromocresol purple	0.010
Cresol red	0.005
Pyridoxal	0.005
Final pH (at 25°C)	6.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 10.52 grams in 1000 ml purified / distilled water. Add 10 gm. of L-Lysine, L-Arginine, L-Ornithine or other L-amino acids. When using DL-amino acids, use 2% concentration. Heat if necessary to dissolve the medium completely. When L-Ornithine is added,readjustment of the pH is required. Dispense in 5 ml amount in screw-capped tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 10 minutes.

Principle And Interpretation

Moeller Decarboxylase Broth Base is used for differentiating gram-negative enteric bacilli on the basis of their ability to decarboxylate amino acids. Moeller introduced the Decarboxylase Broth for detecting the production of lysine and ornithine decarboxylase and arginine dihydrolase (8). Prior to Moellers work, bacterial amino acid decarboxylases were studied by Gale (3) and Gale and Epps (4). Production of ornithine decarboxylase is a helpful criterion in differentiating *Klebsiella* and *Enterobacter* species. *Klebsiella* are nonmotile and do not produce ornithine decarboxylase while *Enterobacter* are motile and produce ornithine decarboxylase except *Enterobacter agglomerans* (7).

This medium contains HM peptone B and peptone which provides nitrogenous and cabonaceous compounds, long chain amino acids and other essential nutrients for the growth of bacteria. Dextrose is the fermentable carbohydrate and pyridoxal is the co-factor for the decarboxylase enzyme. Bromo cresol purple and cresol red are the pH indicators in this medium. When the medium is inoculated with the dextrose fermenting bacteria, the pH is lowered due to acid production, which changes the colour of the indicator from purple to yellow. Acid produced stimulates decarboxylase enzyme. Decarboxylation of lysine yields cadaverine while putrescine is produced due to ornithine decarboxylation. Arginine is first hydrolyzed to ornithine which is then decarboxylated to form putrescine. Formation of these amines increases the pH of the medium, changing the colour of the indicator from yellow to purple. If the organisms do not produce the appropriate enzyme, the medium remains acidic, yellow in colour. Each isolate to be tested should also be inoculated into Moeller Decarboxylase Broth Base medium tube lacking the amino acid.

Inoculated tubes must be protected from air with a layer of sterile mineral oil. Exposure to air may cause alkalinization at the surface of the medium which makes the test invalid.

Type of specimen

Clinical samples - Blood; Food and dairy samples; Water samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5,6).
For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,9,10).
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(2)
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic Use. 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 guidleines should be followed while handling clincal specimens. Saftey guidelines may be referred in individual safety data sheets

Limitations :

1.Some fastidious organisms may show delayed reaction.

2. Overlaying with mineral oil is essential for appropraite results.

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

Light yellow to greenish yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Purple coloured, clear solution without any precipitate in tubes

Reaction

Reaction of 1.05% w/v aqueous solution at 25°C. pH : 6.0±0.2

pH

5.80-6.20

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for upto 4 days with addition of appropriate amino acids and overlaying with sterile mineral oil.

Organism	Inoculum (CFU)	Arginine decarboxylation	Ornithine decarboxylation	Lysine decarboxylation
Citrobacter freundii ATCC 8090	50-100	variable reaction	variable reaction	negative reaction, yellow colour
# Klebsiella aerogenes ATCC 13048 (00175*)	50-100	negative reaction, yellow colour	positive reaction, purple colour	positive reaction, purple colour
Escherichia coli ATCC 25922 (00013*)	50-100	variable reaction	variable reaction	positive reaction, purple colour
Klebsiella pneumoniae ATCC 13883 (00097*)	50-100	negative reaction, yellow colour	negative reaction, yellow colour	positive reaction, purple colour
Proteus mirabilis ATCC 25933	50-100	negative reaction, yellow colour	positive reaction, purple colour	negative reaction, yellow colour
Proteus vulgaris ATCC 13315	50-100	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour
Salmonella Paratyphi A *ATCC 9150*	50-100	delayed positive reaction/positive reaction,purple colour	positive reaction, purple colour	negative reaction, yellow colour

Salmonella Typhi *ATCC 6539*	50-100	delayed positive reaction / negative reaction	negative reaction, yellow colour	positive reaction, purple colour
Serratia marcescens ATCC 8100	50-100	negative reaction, yellow colour	positive reaction, purple colour	positive reaction, purple colour
Shigella dysenteriae ATCC 13313	50-100	negative reaction/ delayed positive reaction	negative reaction, yellow colour	negative reaction, yellow colour
Shigella flexneri ATCC 12022 (00126*)	50-100	negative reaction/ delayed positive reaction	negative reaction, yellow colour	negative reaction, yellow colour
Shigella sonnei ATCC 25931	50-100	variable reaction	positive reaction, purple colour	negative reaction, yellow colour

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

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder 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. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
2. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Inc., Washington, D.C.
3. Gale G. F., 1940, Biochem. J., 34:392.
4. Gale and Epps, 1943, Nature, 152:327.
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.
7. MacFaddin J., 1980, Biochemical Tests for Identification of Medical Bacteria, 2nd ed., Williams and Wilkins, Baltimore.
8. Moeller V., 1955, Acta Pathol. Microbiol. Scand. 36:158.
9. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
10. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

Revision : 03 / 2019

IVD — In vitro diagnostic medical device

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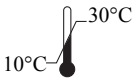

Storage temperature

Do not use if package is damaged

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Thayer Martin Medium Base

M413

Thayer Martin Medium Base used for selective isolation of Gonococci from pathological specimens.

Composition**

Ingredients	Gms / Litre
Peptone, special	23.000
Starch	1.000
Sodium chloride	5.000
Agar	13.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 21.0 grams in 450 ml 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°C. Aseptically add 50 ml of sterile lysed blood and rehydrated contents of one vial of Vitamino Growth Supplement (FD025) and V.C.N Supplement (FD023) or V.C.N.T Supplement (FD024). If desired GC Supplement with Antibiotics (FD021) can be used as a single supplement. Mix well before pouring into sterile Petri plates. If Hemoglobin (FD022) is used suspend 21.0 grams of Thayer Martin Medium Base in 250 ml distilled water.Heat to boiling to dissolve the medium completely. Prepare 250 ml of 2% hemoglobin solution. Sterilize separately by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45°C. Mix both and add the supplements as above.

Principle And Interpretation

Carpenter and Morton reported an improved medium to isolate Gonococci in 24 hours (1). Later on the efficiency of GC medium supplemented with haemoglobin and yeast concentrate was demonstrated for isolating gonococci (2). Subsequently Thayer and Martin Medium was developed for the primary isolation of *Neisseria gonorrhoeae* and *Neisseria meningitidis* from specimens containing mixed flora collected from throat, vagina, rectum an d urethra (3, 4). Thayer and Martin (4) used Vancomycin, Colistin and Nystatin. Martin and Lester (5) used an additional antibiotic Trimethoprim to make the medium selective.

Special peptone provides nutrients to the organisms while starch neutralizes the toxic fatty acids if present in the agar. Haemoglobin provides the X factor whereas the V factor (N.A.D.) is provided by the added supplement. Supplement (FD025) also supplies vitamins, amino acids, coenzymes etc. which enhances the growth of pathogenic *Neisseria* . Vancomycin and colistin inhibits gram-positive and gram-negative bacteria respectively (6). Nystatin inhibits fungi. This medium may inhibit *Haemophilus* species. Some strains of *Capnocytophaga* species may grow on this medium when inoculated with oropharyngeal specimens

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.3% Agar gel.

Colour and Clarity of prepared medium
Basal Medium : Yellow coloured clear to slightly opalescent gel. After addition of haemoglobin or sterile lysed blood and supplements: chocolate coloured opaque gel forms in Petri plates.

Reaction
Reaction of 4.2% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH
6.80-7.20

Cultural Response

M413: Cultural characteristics observed with added sterile lysed blood/Haemoglobin solution (FD022), Vitamino Growth Supplement (FD025) and V.C.N. Supplement (FD023)/V.C.N.T. Supplement (FD024) after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Escherichia coli ATCC 25922	>=10^3	inhibited	0%	
Neisseria gonorrhoeae ATCC 19424	50-100	good-luxuriant	>=50%	small, grayish-white to colourless, mucoid
Neisseria meningitidis ATCC 13090	50-100	good-luxuriant	>=50%	medium to large, blue-gray, mucoid
Proteus mirabilis ATCC 25933	>=10^3	inhibited	0%	

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference

1. Carpenter and Morton, 1947, Proc. N.Y. State Assoc. Public Hlth. Labs., 27:58.
2. Carpenter et al, 1949, Am. J. Syphil. Gonorrh. Vener. Dis., 33:164.
3. Martin, Billings, Hackney and Thayer, 1967, Public Hlth. Rep., 82:361.
4. Thayer J. and Martin J.E. Jr., 1966, Public Health Rep., 81:559.
5. Martin J.E. Jr. and Lester A., 1971, HSMHA Hlth. Service Rep., 86(1):30.
6. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.

Revision : 1 / 2011

Leptospira Medium Base, Korthof, Modified

M457

Intended Use:

Recommended for isolation, cultivation and maintenance of *Leptospira* species.

Composition**

Ingredients	Gms / Litre
Peptone	0.800
Sodium chloride	1.400
Sodium bicarbonate	0.020
Potassium chloride	0.040
Calcium chloride	0.040
Potassium dihydrogen phosphate	0.240
Disodium hydrogen phosphate	0.880
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

1)Preparation of Base: Suspend 3.42 grams in 1000 ml purified/distilled water. Heat if necessary to dissolve the medium completely. Distribute in 100 ml amounts in flasks. Sterilize by autoclaving at Δ 115°C for 15 minutes. Cool to 45-50°C.

2)Preparation of Haemoglobin Solution: To the rabbit blood clot, after removing serum, add equal volume of purified/distilled water. Freeze and thaw repeatedly to haemolyse the corpuscles. Sterilize by Seitz or millipore filtration.

3)Complete Medium: To 100 ml sterile base, add sterile 8 ml inactivated blood serum and 0.8 ml sterile haemoglobin solution. Mix thoroughly. Distribute if desired in 2-3 ml amount in sterile screw capped Bijou bottles / tubes. Test for sterility by incubating at 37°C.

Note: Δ Corresponds to 10 lbs pressure

Principle And Interpretation

Leptospirosis is an acute febrile disease caused by members of the genus *Leptospira* (1,2). Direct culture of blood is the most reliable way to detect *Leptospira* during the first week of illness. After the first week of illness and for several months thereafter, *leptospires* may be isolated by direct culture of undiluted urine specimens. By autopsy, *leptospires* may be isolated from kidney and liver tissues as well as from blood and urine. Leptospira Medium Base, Korthof, Modified is formulated as described by Korthof (3,4) for cultivation and maintenance of *Leptospira* species.

Peptone provide amino acids and other nitrogenous substances to support bacterial growth. Haemoglobin solution and inactivated blood serum provide additional sources of nutrients to the *Leptospires*. The salts supply essential nutrients for the growth of the organisms. Phosphates form buffering system while sodium chloride maintains osmotic equilibrium and also provides essential ions.

All cultures are incubated at room temperature in the dark for up to 6 weeks. The organisms grow below the surface. Material collected from a few centimeters below the surface of broth cultures should be examined weekly for the presence of growth using a direct wet preparation under dark field illumination. *Letpospires* will exhibit corkscrew like motility (1). Examine the tubes for growth every 5-7 days. Growth occurs as a ringed area (disc) 1-3 cm below the surface of the medium. The absence of a ringed area of growth doesnt necessarily mean *leptospires* are not present. Remove a small amount of growth from the disc area and examine microscopically (gram stain is not satisfactory). Microcolonies can be fixed with methanol and stained with Giemsa's stain to show rod forms (3).

Type of specimen

Clinical samples - urine

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. Successive specimens cultured at least 1 day apart increase the likelihood of positive culture, since Leptospira may be shed sporadically (4).

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

Off-white to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Yellowish brown coloured, clear to slightly opalescent solution after addition of serum and haemogloin

Reaction

Reaction of 0.342% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH

7.00-7.40

Cultural Response

Cultural characteristics observed with added inactivated blood serum and sterile haemoglobin solution, after an incubation at 30°C for upto 2-7days.

Organism	Growth
Leptospira interrogans sero. *grippotyhosa*	luxuriant
Leptospira interrogans sero. *Australis*	luxuriant
Leptospira interrogans sero. *Canicola*	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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

1. Forbes B. A., Sahm A. S., and Weissfeld D. F., Bailey & Scotts Diagnostic Microbiology, 10th Ed., 1998, Mosby, Inc., St. Louis, Mo.
2. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
3. Korthof G., 1932, Zentralbl. Bakteriol. Parasitenkd. Infektionskr. Hyg. Abt. I. Orig., 125:429.
4. Rechcigl M. Jr. (Ed.), 1978, Handbook Series in Nutrition and Food, Vol. III, CRC Press.
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.

Revision : 03/2023

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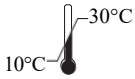
IVD ***In vitro*** **diagnostic medical device**



Storage temperature

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