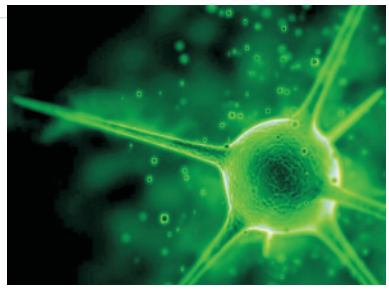
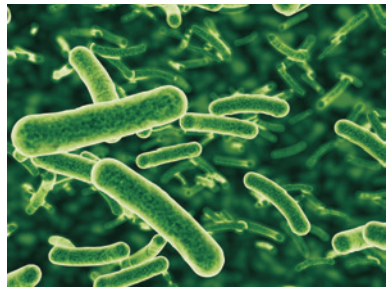
Bacteriological Differentiation Discs

For Rapid Differentiation





HiMediaLaboratories™
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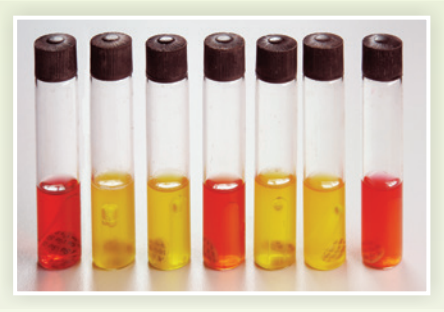
For life is precious

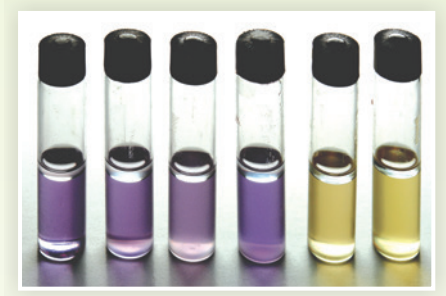
RAPID Identification, Differentiation & Biochemical Characterization

HiMedia Provides an assorted range of differential disc for

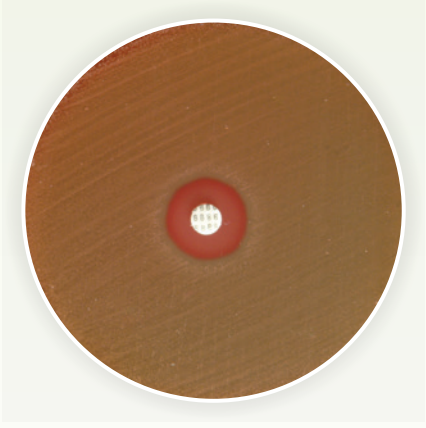
- **Carbohydrate fermentation test**



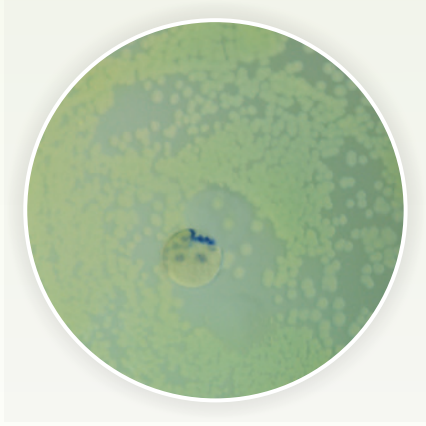
- **Amino Acids Decarboxylation test**



- **Identification & Differentiation**
  of various microorganisms like *Streptococcus* species, *Haemophilus* species, *Vibrio* species etc.



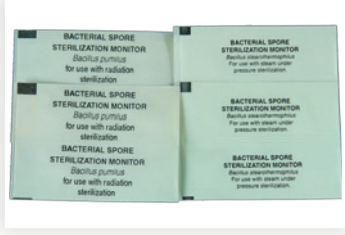
- **Biochemical tests**
  like Indole test, Hippurate hydrolysis test, ONPG test, Oxidase test, H_2S production test, Nitrate reduction test & Esculin hydrolysis



Also available

Sterilization Monitoring Strips

for Monitoring Steam sterilization & Radiation sterilization



For more details browse the following pages ....

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Introduction

Identification and differentiation of microorganisms is of utmost importance when dealing with bacteria associated with infections. To ascertain the findings of any clinical samples it is necessary to identify the causative agents till the species level. This identification cannot be solely carried out on the basis of staining and colony characteristics. Biochemical testing has to be performed as each organism has a different set of biochemical tests which would help in its identification and differentiation from others. There are certain biochemical characteristics which are specific to few organisms aiding it in their rapid identification.

Biochemical differentiation of has to be carried out using an array of biochemical tests starting from carbohydrate fermentation tests & amino acid decarboxylation tests to certain specific tests like ONPG, Indole production or Oxidase. The sensitivity or resistance to certain antibiotics can also help in differentiation of bacteria from the same genus. However, carrying out these tests is a tedious and time consuming process requiring preparation of various media and reagents.

To ease out this process HiMedia provides a wide range of discs and strips impregnated with biochemicals in form of Differentiation Discs (DD).These discs are rapid, economical and user-friendly and can be used for effectual screening of bacteria. These discs help in identification and differentiation of microorganisms when placed on Agar surfaces, in culture media or culture suspensions. Also available are spores strips that are useful in the validation studies of sterilization (steam and radiation).

The range of products available are as follows:

HiMedia's Range of Bacteriological Differential Discs (DD)

Discs for Carbohydrate Fermentation Test

Product		Code	*Packing
Adonitol	Ad	DD025-1VL	1vl
Arabinose	Ar	DD001-1VL	1vl
Cellobiose	Ce	DD028-1VL	1vl
Dextrose	De	DD002-1VL	1vl
Dulcitol	Du	DD003-1VL	1vl
Galactose	Ga	DD016-1VL	1vl
Fructose	Fc	DD017-1VL	1vl
Inositol	Is	DD027-1VL	1vl
Inulin	In	DD026-1VL	1vl
Lactose	La	DD004-1VL	1vl
Maltose	Ma	DD005-1VL	1vl
Mannitol	Mn	DD006-1VL	1vl
Mannose	Mo	DD007-1VL	1vl
Melibiose	Mb	DD030-1VL	1vl
Raffinose	Rf	DD029-1VL	1vl
Rhamnose	Rh	DD010-1VL	1vl
Salicin	Sa	DD011-1VL	1vl
Sorbitol	Sb	DD012-1VL	1vl
Sucrose	Su	DD013-1VL	1vl
Trehalose	Te	DD031-1VL	1vl
Xylose	Xy	DD014-1VL	1vl

Amino Acid Discs

Product	Code	*Packing
Lysine Hydrochloride	DD049-1VL	1vl
Arginine Hydrochloride	DD050-1VL	1vl
Ornithine Hydrochloride	DD051-1VL	1vl
Proline	DD052-1VL	1vl
Serine	DD053-1VL	1vl
Histidine	DD054-1VL	1vl
*1Vl contains 25 Discs		

Differentiation Discs and Strips

Product	Code	Packing
***Bacitracin (50 discs / vl) B** for identification of *Streptococcus pyogenes*.	DD015-1VL	1vl
***Bile Esculin (50 discs / vl)** for detection of esculin hydrolysis in the presence of bile.	DD024-1VL	1vl
***DMACA Indole Discs Dm (50 discs / vl)** for detection of indole formation by microorganisms.	DD040-1VL	1vl
Hippurate (25 Discs / vl) Hp for hippurate hydrolysis testing.	DD035-1VL	1vl
***Kovac's Reagent Strips (25 Strips / vl)** for Indole testing.	DD019-1VL	1vl
***Lead Acetate Paper Strips (25 strips / vl)** for H_2S testing.	DD034-1VL	1vl
***Nitrate Discs (50 discs / vl) N** substrate for detection of nitrate reduction by microorganisms.	DD041-1VL	1vl
***Nitrate Reagent Discs Nr (Twin Pack)**	DD042-1VL	1vl
Part A : 50 discs / vl Part B : Rehydrating fluid (5ml / vl) for detection of nitrate reduction by microorganisms.		
***ONPG (50 discs / vl) On** for ONPG testing.	DD008-1VL	1vl
***Optochin (5 mcg) Op (50 discs / vl)** for identification of *Streptococcus pnuemoniae*.	DD009-1VL	1vl
***Oxidase Discs (50 discs / vl)** for Oxidase testing (10 mm).	DD018-1VL	1vl
****Spore Strips (25 strips/pack)** steam sterilization monitor strips, *Bacillus stearothermophilius*, 10^6 spores per strip.	DD032-1PK	1pk
****Spore Strips (B. pumilus) (25 strips / pack)** radiation sterilization monitor strips, *Bacillus pumilus*, 10^6 spores per strip.	DD039-1PK	1pk
Sterile Discs, 10mm	DD036-1VL	1vl
***X Factor (50 discs / vl) X** for presumptive identification of *Haemophilus* species.	DD020-1VL	1vl
▼V Factor (50 discs / vl) V for presumptive identification of *Haemophilus* species.	DD021-1VL	1vl
▼X+V Factor X+V (50 discs/vl) for presumptive identification of *Haemophilus* species.	DD022-1VL	1vl
***Vibrio O129 Differential Disc (10 mcg) (50 discs/vl)** For differentiation of *Vibrio* species based on sensitivity to Vibriostatic agent O129.	DD047-1VL	1vl
***Vibrio O129 Differential Disc (150 mcg) (50 discs/vl)** For differentiation of *Vibrio* species based on sensitivity to Vibriostatic agent O129	DD048-1VL	1vl

▼ : Store below (-10°C)

* : All products to be stored between 2 to 8° C. For prolonged use, store at (-20°C).

** : Store between 15 to 27°C.

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Carbohydrate Differentiation Discs

Application

Carbohydrate Differentiation Discs are used to differentiate bacteria on the basis of carbohydrate fermentation abilities.

Directions

A Sugar free medium base is prepared as desired, dispensed and sterilized. Following media are recommended for this test.

Liquid Media

M885	Andrade Peptone Water
MV885	Andrade HiVeg™ Peptone Water
M909	Andrade Peptone Water with Meat Extract
MV909	Andrade Peptone Water w/ HiVeg™ Extract No. 1
M054	Phenol Red Broth Base
MV054	Phenol Red HiVeg™ Broth Base
M279	Phenol Red Broth Base w/ Meat Extract
MV279	Phenol Red Broth Base w/ HiVeg™ Extract No. 1
M284	Purple Broth Base
MV284	Purple HiVeg™ Broth Base
M676	Yeast Fermentation Broth
MV676	Yeast Fermentation HiVeg™ Broth Base

Semisolid Media

M159	Cystine Tryptone Agar
MV159	Cystine Tryptone Agar, HiVeg™
M395	OF Basal Medium
MV395	OF Basal HiVeg™ Medium
M319	Tryptone Agar Base
MV319	Tryptone Agar Base, HiVeg™

Solid Media

M053	Phenol Red Agar Base
MV053	Phenol Red HiVeg™ Agar Base
M098	Purple Agar Base
MV098	Purple HiVeg™ Agar Base

Any medium-liquid, semisolid or solid can be used as per choice. Liquid and semisolid media are dispensed in 5 ml amounts in test tubes and sterilized. On cooling to 45-50°C a single Carbohydrate disc is aseptically added to each tube and inoculated with the test organisms. In semisolid medium the disc is pushed in the medium along with the inoculum just below the surface of the medium, so that the medium at the bottom can serve as control while fermentation can be detected at the surface level. Using solid media it is possible to detect fermentation of number of sugars on the same plate. Sterile plates containing the agar medium of choice are surface seeded with test organism(s) and required Carbohydrate discs are aseptically placed and pressed gently on the surface of the plate at sufficient distance (2 cm) from each other. Incubation is carried out at 36 ± 1.0°C for 18-48 hours. Results are recorded at 18-24 hours and again at 48 hours. The results should be frequently observed since reversal of fermentation reaction can take place. In case of liquid medium gas produced during fermentation is collected in the inverted Durham's tube while acid produced changes the colour of the medium. In semisolid media gas produced is trapped and seen as bubbles. On agar plates fermentation is visualised by the change in colour around the disc.

M054 Phenol Red Broth Base (With added DD013 Sucrose)

1. Uninoculated control
2. *Citrobacter freundii* ATCC 8090 +
3. *Enterobacter aerogenes* ATCC 13048 +
4. *Escherichia coli* ATCC 25922 —
5. *Klebsiella pneumoniae* ATCC 13883 +
6. *Serratia marcescens* ATCC 8100 +
7. *Salmonella* Typhimurium ATCC 14028 —

Key : + = Acid and gas production — = no fermentation

Principle and Interpretation

Ability of an organism to ferment a specific carbohydrate added in the basal medium, results in the production of acid or acid and gas. This ability has been used to characterize a specific species of bacteria (2, 3). When carbohydrate impregnated disc is added to a culture medium the carbohydrate diffuses through the medium and is fermented by the microorganism. The acid (or acid and gas) produced lowers the pH of the medium and the indicator in the basal medium thus changes colour (e.g. phenol red changes from red to orange to yellow).

Bacteria capable of fermentation grow in Andrade Peptone Water (M885) and produce acid due to fermentation of the added carbohydrate and changes the colour of the indicator from light straw coloured to pink (1). Fermentation reaction can also be checked in Phenol Red Broth Base (M054) and Bromo Cresol Purple Broth Base (M676) where the colour change is from red to yellow and purple to yellow respectively.

Quality Control

Code	Product	Appearance	Cultural Response
DD001	Arabinose	Filter paper discs of 10 mm diameter bearing letters "Ar" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Arabinose Differentiation discs tested using Phenol Red Broth Base (M054).
DD002	Dextrose	Filter paper discs of 10 mm diameter bearing letters "De" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Dextrose Differentiation discs tested using Phenol Red Broth Base (M054).
DD003	Dulcitol	Filter paper discs of 10 mm diameter bearing letters "Du" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Dulcitol Differentiation discs tested using Phenol Red Broth Base (M054).
DD004	Lactose	Filter paper discs of 10 mm diameter bearing letters "La" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Lactose Differentiation discs tested using Phenol Red Broth Base (M054).
DD005	Maltose	Filter paper discs of 10 mm diameter bearing letters "Ma" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Maltose Differentiation discs tested using Phenol Red Broth Base (M054).
DD006	Mannitol	Filter paper discs of 10 mm diameter bearing letters "Mn" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Mannitol Differentiation discs tested using Phenol Red Broth Base (M054).
DD007	Mannose	Filter paper discs of 10 mm diameter bearing letters "Mo" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Mannose Differentiation discs tested using Phenol Red Broth Base (M054).
DD010	Rhamnose	Filter paper discs of 10 mm diameter bearing letters "Rh" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Rhamnose Differentiation discs tested using Phenol Red Broth Base (M054).
DD011	Salicin	Filter paper discs of 10 mm diameter bearing letters "Sa" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Salicin Differentiation discs tested using Phenol Red Broth Base (M054).
DD012	Sorbitol	Filter paper discs of 10 mm diameter bearing letters "Sb" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Sorbitol Differentiation discs tested using Phenol Red Broth Base (M054).
DD013	Sucrose	Filter paper discs of 10 mm diameter bearing letters "Su" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Sucrose Differentiation discs tested using Phenol Red Broth Base (M054).
DD014	Xylose	Filter paper discs of 10 mm diameter bearing letters "Xy" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Xylose Differentiation discs tested using Phenol Red Broth Base (M054).
DD016	Galactose	Filter paper discs of 10 mm diameter bearing letters "Ga" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Galactose Differentiation discs tested using Phenol Red Broth Base (M054).
DD017	Fructose	Filter paper discs of 10 mm diameter bearing letters "Fc" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Fructose Differentiation discs tested using Phenol Red Broth Base (M054).
DD025	Adonitol	Filter paper discs of 10 mm diameter bearing letters "Ad" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Adonitol Differentiation discs tested using Phenol Red Broth Base (M054).
DD026	Inulin	Filter paper discs of 10 mm diameter bearing letters "In" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Inulin Differentiation discs tested using Phenol Red Broth Base (M054).
DD027	Inositol	Filter paper discs of 10 mm diameter bearing letters "Is" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Inositol Differentiation discs tested using Phenol Red Broth Base (M054).
DD028	Cellobiose	Filter paper discs of 10 mm diameter bearing letters "Ce" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Cellobiose Differentiation discs tested using Phenol Red Broth Base (M054).
DD029	Raffinose	Filter paper discs of 10 mm diameter bearing letters "Rf" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Raffinose Differentiation discs tested using Phenol Red Broth Base (M054).
DD030	Melibiose	Filter paper discs of 10 mm diameter bearing letters "Mb" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Melibiose Differentiation discs tested using Phenol Red Broth Base (M054).
DD031	Trehalose	Filter paper discs of 10 mm diameter bearing letters "Te" in continuous printing style.	The carbohydrate fermentation reactions observed after an incubation at 35-37°C for 18-48 hours (§) of various bacteria with Trehalose Differentiation discs tested using Phenol Red Broth Base (M054).

Organism (ATCC)	Growth	Adonitol (DD025)		Arabinose (DD001)		Cellobiose (DD028)		Dextrose (DD002)		Dulcitol (DD003)		Galactose (DD016)		Inositol (DD027)		Lactose (DD004)		Maltose (DD005)		Mannitol (DD006)	
		Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas
Citrobacter freundii (8090)	luxuriant	-	-	+	+	+	-	+	+	-	-	+	+	-	-	+	+	+	+	+	+
Enterobacter aerogenes (13048)	luxuriant	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	+	+
Escherichia coli (25922)	luxuriant	-	-	+	+	-	-	+	+	-	-	+	+	-	-	+	+	+	+	+	+
Klebsiella pneumoniae (13883)	luxuriant	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	+	+
Proteus vulgaris (13315)	luxuriant	-	-	-	-	-	-	+	+	-	-	+	+	-	-	-	-	+	+	-	-
Salmonella Typhimurium (14028)	luxuriant	-	-	+	+	-	-	+	+	+	+	+	+	+	+	-	-	+	+	+	+
Salmonella Typhi (6539)	luxuriant	-	-	-	-	-	-	+	-	-	-	+	-	-	-	-	-	+	-	+	-
Serratia marcescens (8100)	luxuriant	-	-	-	-	-	-	+	+	-	-	+	-	+	-	-	-	+	-	+	-
Shigella flexneri (12022)	luxuriant	-	-	-	-	-	-	+	-	-	-	+	-	-	-	-	-	+	-	+	-

Organism (ATCC)	Growth	Mannose (DD007)		Melibiose (DD030)		Raffinose (DD029)		Rhamnose (DD010)		Salicin (DD011)		Sorbitol (DD012)		Sucrose (DD013)		Trehalose (DD031)		Xylose (DD014)	
		Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas
Citrobacter freundii (8090)	luxuriant	+	+	-	-	-	-	+	+	-	-	+	+	+	+	+	+	+	+
Enterobacter aerogenes (13048)	luxuriant	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Escherichia coli (25922)	luxuriant	+	+	+	+	-	-	+	+	-	-	+	+	-	-	+	+	+	+
Klebsiella pneumoniae (13883)	luxuriant	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Proteus vulgaris (13315)	luxuriant	-	-	-	-	-	-	-	-	+	+	-	-	+	+	+	+	+	[+]
Salmonella Typhimurium (14028)	luxuriant	-	-	+	+	-	-	+	+	-	-	+	+	-	-	+	+	+	+
Salmonella Typhi (6539)	luxuriant	+	+	+	+	-	-	-	-	-	-	+	-	-	-	+	-	+	-
Serratia marcescens (8100)	luxuriant	+	+	-	-	-	-	-	-	+	[+]	+	-	+	+	+	[+]	-	-
Shigella flexneri (12022)	luxuriant	+	+	-	-	-	-	-	-	-	-	+	-	-	-	+	-	-	-

Organism (ATCC)	Growth	Fructose (DD017)		Inulin (DD026)	
		Acid	Gas	Acid	Gas
Enterobacter aerogenes (13048)	luxuriant	+	+	NA	NA
Escherichia coli (25922)	luxuriant	+	+	NA	NA
Shigella flexneri (12022)	luxuriant	+	+	NA	NA
Streptococcus pneumoniae (6303)	luxuriant	+	+	+	-
Neisseria meningitidis (13090)	luxuriant	-	-	NA	NA
Streptococcus pyogenes (19615)	luxuriant	NA	NA	-	-

Key :

(§) : longer if necessary,
+ : positive reaction, yellow colour/gas production
— : negative reaction, no colour change or red / no gas production
NA : Not Applicable
[+] : weak / slight
* for more details refer,

1. Bergey's Manual of Systematic Bacteriology, 1984, Vol. I, Williams and Wilkins, Baltimore
2. Bergey's Manual of Systematic Bacteriology, 1994, 9th Ed. Williams and Wilkins, Baltimore

Reference

1. Maxted W. R., 1953, J. Clin. Path., 6:234.
2. Eaton A.D, Clesceri L.S., Greenberg. A.E., Rice E. W., (Eds) 2005, Standard Methods for the Examination of Water and wastewater, 21st edn, APHA. Washington. DC.
3. Mackie and McCartney, 1996, Practical Medical Microbiology, 14th ed., Vol. 2, Collee J. G., Fraser A. G., Marmion B. P., Simmons A (Eds.), Churchill Livingstone, Edinburgh.

Storage and Shelf-Life

Store at 10-30° C. Use before expiry date on the label.

Amino Acid Discs DD049-DD054

Application

Amino acid discs are used for amino decarboxylation test to differentiate bacteria.

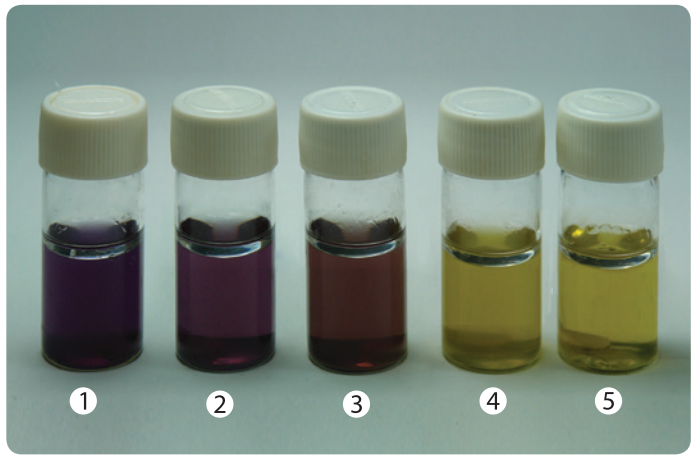
Directions

To determine amino acid decarboxylation, the respective discs (DD) is added in the Decarboxyalse Broth Base, Moeller (M393) which is used as a negative control for studying decarboxylation or as a base for the addition of amino acids. The test organism is inoculated into the broth containing the disc (DD). The inoculated tubes are overlaid with sterile mineral oil and incubated at 35-37°C for up to 4 days. A purple colour indicates positive decarboxylation reaction.

Principle and Interpretation

Amino acid discs are used to differentiate the microorganisms on the basis of their ability to decarboxylate the amino acids Moeller introduced the Decarboxylase Broth for detecting the production of lysine and ornithine decarboxylase and arginine dihydrolase (1). Prior to Moellers work, bacterial amino acid decarboxylases were studied by Gale (2), Gale and Epps (3). Moeller Decarboxylase Broth Base (M393) contains dextrose which 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 amino acids yields amine. Lysine yields cadaverine, while putrescine is produced due to ornithine decarboxylation. Arginine is first hydrolyzed to ornithine which is then decarboxylated to form putrescin. Formation of the amine 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. 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. Each isolate to be tested should also be inoculated into Moeller Decarboxylase Broth Base medium tube lacking the amino acid.

Positive Test: Colour of the medium changes from yellow to purple
Negative Test: Colour of the medium changes to yellow or there is no change



DD049 Lysine Hydrochloride discs-incorporated in Moeller decarboxylase Broth Base (M393)

1. *Enterobacter aerogenes* ATCC 13048 '+'
2. *Salmonella* Typhi ATCC 6539 '+'
3. Control
4. *Proteus vulgaris* ATCC 13315 '-'
5. *Shigella flexneri* ATCC 12022 '-'

Key -

\+ : Positive reaction
\- : Negative reaction

Quality Control

Appearance

Filter paper discs of 10 mm diameter

Cultural Response

Cultural characteristics observed in Moeller Decarboxylase Broth Base (M393) with added respective amino acid discs (DD049-DD054) after an incubation at 35-37°C upto 4 days (Inoculated tubes are overlaid with sterile mineral oil) .

DD049 - Lysine Hydrochloride discs
DD050 - Arginine Hydrochloride discs
DD051 - Ornithine Hydrochloride discs
DD052 - Proline discs
DD053 - Serine discs
DD054 - Histidine discs

Organism	Inoculum (CFU)	Lysine (DD049)	Arginine (DD050)	Ornithine (DD051)	Proline (DD052)	Serine (DD053)
Citrobacter freundii ATCC 8090	50-100	negative reaction, yellow colour	variable reaction	variable reaction	variable reaction	variable reaction
Enterobacter aerogenes ATCC 13048	50-100	positive reaction, purple colour	negative reaction, yellow colour	positive reaction, purple colour	positive reaction, purple colour	positive reaction, purple colour
Escherichia coli ATCC 25922	50-100	variable reaction	variable reaction	variable reaction	variable reaction	variable reaction
Klebsiella pneumoniae ATCC 13883	50-100	positive reaction, purple colour	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour
Proteus mirabilis ATCC 25933	50-100	negative reaction, yellow colour	negative reaction, yellow colour	positive reaction, purple colour	positive reaction, purple colour	positive reaction, purple colour
Proteus vulgaris ATCC 13315	50-100	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour
Salmonella Paratyphi A ATCC 9150	50-100	negative reaction, yellow colour	delayed positive reaction/ positive reaction,purple colour	positive reaction, purple colour	positive reaction, purple colour	positive reaction, purple colour
Salmonella Typhi ATCC 6539	50-100	positive reaction, purple colour	delayed positive reaction / negative reaction	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour
Serratia marcescens ATCC 8100	50-100	positive reaction, purple colour	negative reaction, yellow colour	positive reaction, purple colour	positive reaction, purple colour	positive reaction, purple colour
Shigella dysenteriae ATCC 13313	50-100	negative reaction, yellow colour	negative reaction/ delayed positive reaction	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour
Shigella flexneri ATCC 12022	50-100	negative reaction, yellow colour	negative reaction/ delayed positive reaction	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour
Shigella sonnei ATCC 25931	50-100	negative reaction, yellow colour	variable reaction	positive reaction, purple colour	positive reaction, purple colour	positive reaction, purple colour
Pseudomonas aeruginosa ATCC 9027	50-100	negative reaction, yellow colour	positive reaction, purple colour	negative reaction, yellow colour	negative reaction, yellow colour	negative reaction, yellow colour

Organism	Inoculum (CFU)	Histidine (DD054)
Pseudomonas aeruginosa ATCC 27853	50-100	positive reaction, purple colour
Vibrio parahaemolyticus ATCC 17802	50-100	positive reaction, purple colour
Vibrio fischeri ATCC 7744	50-100	Negative reaction

Reference

1. Moeller V., 1955, Acta Pathol. Microbiol. Scand. 36:158.
2. Gale G. F., 1940, Biochem. J., 34:392.
3. Gale and Epps,1943,Nature, 152:327.

Storage and Shelf-Life

Store the discs at 10-30°C. Use before expiry date on the label.

Bacitracin Susceptibility Test Discs DD015

Application

Bacitracin Susceptibility Test Discs are used for the identification and differentiation of Group A streptococci (especially *S.pyogenes*) from other ß-haemolytic streptococci.

Directions

Pure Cultures :

Evenly inoculate the surface of Tryptose Blood Agar Base (M097) with pure culture of ß-haemolytic streptococci to be tested. Aseptically place a Bacitracin disc on the inoculated surface and incubate the inverted plate at 35-37°C for 18-24 hours in 10% CO_2. Observe for the presence of zone of inhibition around the Bacitracin disc. A zone indicates that the *Streptococcus* is presumptively of Group A. If desired further confirmation can be obtained by serological grouping.

Clinical Materials :

Incubate Tryptose Blood Agar Base (M097) plate with throat swab or other material. Spread the inoculum to obtain discrete colonies on some portion of the plate, so as to determine the species in mixed growth. Aseptically place a Bacitracin disc on the secondary area of inoculation and incubate the inverted plates at 35-37°C for 18-24 hours in 10% CO_2. Examine for zones of inhibition. Bacitracin is inhibitory to many organisms except ß-haemolytic streptococci, however the presence of a zone of inhibition does not essentially indicate Lancefield Group A streptococci. If the colonial morphology is carefully observed, it is possible to select presumptive Group A streptococci. By serological grouping, further confirmation can be obtained.

Precautions

Use known Group A and non-Group A streptococci to determine the accuracy of the discs and inoculum.

Principle and Interpretation

The growth of Group A ß-haemolytic streptococci on blood agar is inhibited by 0.04 units Bacitracin disc. Micrococci and streptococci are also inhibited by 0.04 units disc, while all coagulase-negative staphylococci are resistant (4). Bacitracin susceptibility test discs are filter paper discs impregnated with 0.04 units of Bacitracin. Bacitracin discs can save considerable time, labour and materials if used as a screening test before serological grouping. Maxted showed that Group A streptococci were more sensitive to Bacitracin than ß-haemolytic strains of other groups (1). Hence he suggested that Bacitracin might be used as a rapid diagnostic agent for Group A streptococci. Levinson and Frank (2) who employed Bacitracin impregnated filter paper discs for this purpose, observed that many sensitive ß-haemolytic streptococci were of Group A. Steamer *et al* compared Bacitracin disc, fluorescent antibody technique and Lancefield precipitin technique and found that the Bacitracin disc technique was most convenient for routine clinical laboratory (3). Bacitracin sensitivity test along with Furacin and Optochin tests are useful for distinguishing *Aerococcus viridans* and *S. milleri* from enterococci and *Streptococcus mitis* (2).

Bacitracin Susceptibility Test Discs (DD015)

Streptococcus pyogenes ATCC 19615

Quality Control

Appearance

Filter paper discs of 6 mm diameter bearing letters "B" in continuous printing style

Cultural Response

DD015: Average diameter of zone of inhibition for *S.pyogenes* observed on Tryptose Blood Agar (M097) after an incubation at 35-37°C for 18-24 hours.

Organism	Diameter of zone of inhibition (mm)
Streptococcus pyogenes ATCC 19615	15 mm

Reference

1. Maxted W. R., 1953, J. Clin. Path., 6:234.
2. Levinson M. L. and Frank P.F., 1955, J. Bact., 69:234.
3. Streamer C.W et al, 1962, Am. J. Dis. Children, 104:157.
4. Guthof O.,1960, Ztschr. F hyg. U. Infektionskr.,146:425

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

Bile Esculin Discs DD024

Application

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

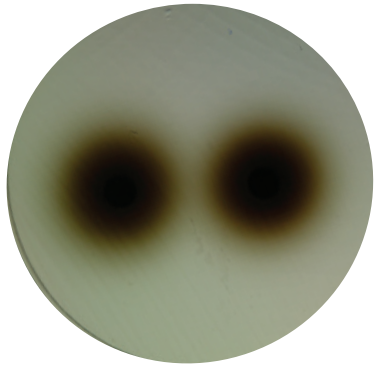
Quality Control

Appearance

Plain filter paper discs of 6mm diameter

Cultural Response

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



M340 Bile Esculin Agar Base with Bile Esculin Discs (DD024)

Enterococcus faecalis ATCC 29212

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

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.

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

DMACA Indole Discs DD040

Application

DMACA Indole discs are used for Indole test to determine the ability of an organism to split indole from the tryptophan molecule, and thus to aid differentiation between Organisms on the basis of Indole formation.

Directions

Place the DMACA Indole Disc on suspected colony from HiCrome UTI Agar (M1353) or HiCrome UTI Agar, Modified (M1418) plate. Observe for appearance of blue-purple colour within 10-30 seconds.

Principle and Interpretation

In the presence of oxygen, some bacteria are able to split tryptophan into indole and alpha-aminopropionic acid. The presence of indole can be detected by the addition of DMACA (p-Dimethylaminocinnamaldehyde) reagent indicated by formation of bluish-purple colour within 10-30 seconds.

Quality Control

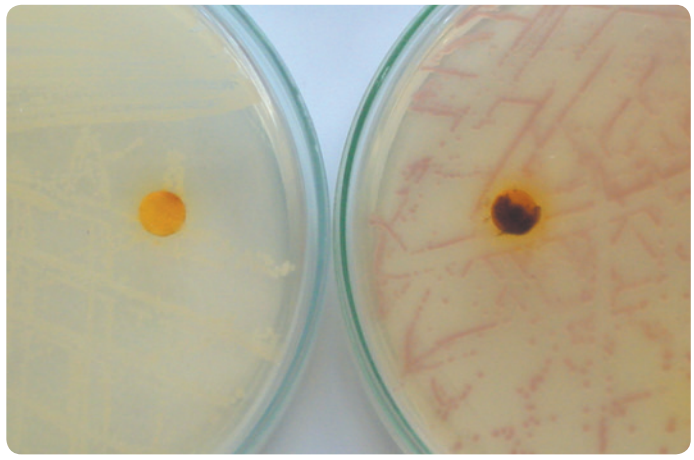
Appearance

Filter paper discs of 6 mm diameter bearing letters 'Dm' in continuous printing style

Cultural Response

The indole production reaction was observed within 10-30 seconds by organisms grown on HiCrome UTI Agar (M1353) incubated at 35-37°C for 18-24 hours.

Organism	Indole production
Escherichia coli ATCC 25922	positive reaction, blue to purple colour.
Staphylococcus aureus ATCC 25923	negative reaction.
Klebsiella pneumoniae ATCC 13883	negative reaction.
Pseudomonas aeruginosa ATCC 27853	negative reaction.



DMACA Indole Discs (DD040)

1. *Staphylococcus aureus* ATCC 25923
2. *Escherichia coli* ATCC 25922

Reference

1. MacFaddin J. F., 1980, Biochemical Tests for Identification of Medical Bacteria, 2nd ed., Williams and Wilkins, Baltimore

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

HIMEDIA®

Hippurate Hydrolysis Test DD035

Application

Hippurate Hydrolysis Test is used for detection of hippurate hydrolyzing bacteria, mainly Streptococcal species.

Directions

Aseptically place hippurate disc in Brain Heart Infusion Broth (M210) inoculated with ß haemolytic streptococci. Incubate at 35-37°C for 48 hours. Separate out the growth by centrifuging the broth. Add 2 ml of ferric chloride reagent to 2 ml of freshly prepared supernatant from the centrifuged culture tubes. Shake well and observe persistence of the precipitate formed even after 10 minutes.

Preparation of ferric chloride reagent :

Ingredients	Grams/100ml
Ferric chloride	12.0 gm
Distilled water	94.6 ml
Concentrated hydrochloric acid	5.4 ml

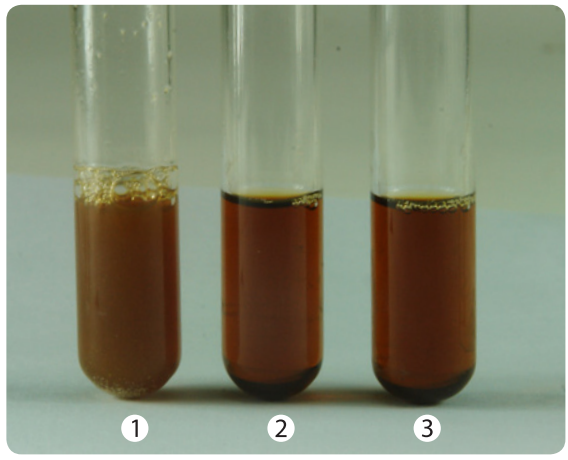
Principle and Interpretation

Group B streptococci (*Streptococcus agalactiae*) and some enterococci can hydrolyze 1% aqueous sodium hippurate to produce glycine and sodium benzoate. Glycine is deaminated by the oxidizing agent ninhydrin which gets reduced and becomes purple. The test medium must contain only hippurate, since ninhydrin reacts with any free amino acids present (5, 6). Group B streptococci can thus be distinguished from Groups A, C, F and G which can not hydrolyze sodium hippurate. Some Group D and very few viridans streptococci can also hydrolyze sodium hippurate. Ayers and Rupp (1) discovered that haemolytic streptococci from human and bovine sources could be differentiated by their ability to hydrolyze sodium hippurate (2). Facklam et al (3) modified the procedure for the presumptive identification of Group A, B and D streptococci. The ability of an organism to hydrolyze sodium hippurate is one of the tests that aid in the differentiation of bovine ß-haemolytic Group B streptococci, from human ß-haemolytic Group B *Streptococcus* species (2). Differentiation of ß-haemolytic Group B streptococci from ß-haemolytic Group A streptococci and non-enterococcal Group D streptococci is also aided by the determination of hippurate hydrolysis by enzymatic activity to form benzoic acid as the end product (4).

Quality Control

Appearance

Filter paper discs of 10 mm diameter bearing letters 'Hp' in continuous printing style.



1. *S. agalactiae* (ATCC 4768) Showing positive reaction (Formation of Brown precipitate even after 10 minutes of addition of 12% ferric chloride)
2. *S. pyogenes* (ATCC 19615) Showing negative reaction (No precipitate after 10 minutes of addition of 12% ferric chloride)
3. Negative control (Uninoculated tube)

Cultural Response

The Hippurate hydrolysis reaction is observed after an incubation at 35-37°C for 24-48 hours, of various bacteria with Hippurate differentiation discs, tested using Brain Heart Infusion Broth (M210).

Organism	Indole production
Enterococcus faecalis ATCC 29212	negative, precipitate if any, dissolves on shaking
Streptococcus agalactiae ATCC 4768	positive, brown flocculant precipitate persisting on shaking after 10 minutes.
Streptococcus pyogenes ATCC 19615	negative, precipitate if any, dissolves on shaking

Reference

1. Ayers S.H. and Rupp P. (1922), J. Infect. Disease., 30:388.
2. Eaton A.D, Clesceri L.S., Greenberg. A.E, Rice E. W., (Eds) 2005, Standard Methods for the Examination of Water and Wastewater, 21st edn, APHA. Washington. DC.
3. Facklam and Moody, 1970, Appl. Microbiol., 20:245.
4. Mackie and McCartney, 1996, Practical Medical Microbiology, 14th ed., Vol. 2, Collee J. G., Fraser A. G., Marmion B. P., Simmons A (Eds.), Churchill Livingstone, Edinburgh.
5. Facklam, R.R. et al, 1974, Appl. Microbiol.,27:102-1136. Forbes. A.B, Sahm. F.D, 2002, Bailey and Scott's Diagnostic Microbiology, 11th ed., The C.V. Mosby Co., St. Louis.

Storage and Shelf-Life

Store at 10-30°C. Use before expiry date on the label.

Kovac's Reagent Strip DD019

Application

Kovac's Reagent Strips are used to detect indole producing bacteria.

Directions

Indole production by organisms is observed by inserting the Kovac's reagent strip between the plug and inner wall of the tube, above the inoculated Peptone Water (M028) and incubating at 35-37°C for 18-24 hours.

Preparation of Kovac's reagent

Kovac's reagent is prepared by dissolving 10 gm of p-dimethyl aminobenzaldehyde in 150 ml of isoamyl alcohol and then slowly adding 50 ml of concentrated hydrochloric acid.

Principle and Interpretation

The various enzymes involved in the degradation of tryptophan to indole are collectively called as tryptophanase, a general term used to denote the complete system of enzymes (2). The presence of indole is detected by the Kovac's reagent strip which turns pink in the presence of indole. Kovac's Reagent Strips are sterile filter paper strips impregnated with Kovac's reagent. Peptone is used in the preparation of Peptone Water because of its high tryptophan content. When tryptophan is degraded by bacteria, indole is produced. Tryptone Water (M463) can also be used to detect indole production in the identification of members of coliform group (1).

Quality Control

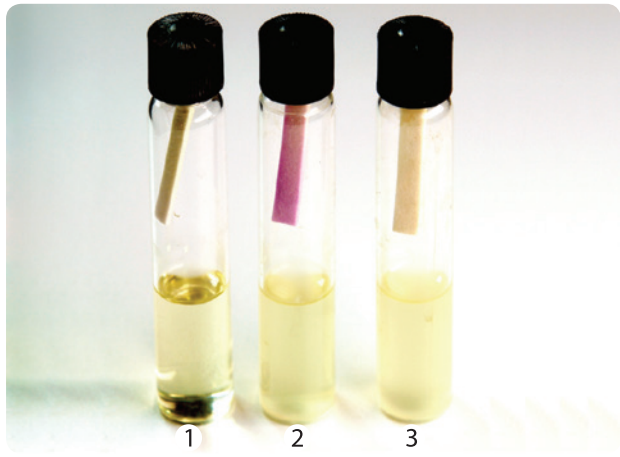
Appearance

Filter paper strips of 70 mm x 5 mm.

Cultural Response

DD019: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours by inserting Kovac's Reagent Strips between the plug and inner wall of the tube, above the inoculated Peptone Water (M028).

Organism	Growth	Indole
Escherichia coli ATCC 25922	luxuriant	positive reaction, pink colour at the lower portion of the strip.
Enterobacter aerogenes ATCC 13048	luxuriant	negative reaction, no colour change.



Peptone Water (M028) with Kovac's Reagent Strip (DD019)

1. Control
2. *Escherichia coli* ATCC 25922
3. *Enterobacter aerogenes* ATCC 13048

Reference

1. Eaton A.D, Clesceri L.S., Greenberg. A.E, Rice E. W.(Eds) 2005, Standard Methods for the Examination of Water and wastewater, 21st ed., APHA, Washington DC.
2. MacFaddin J. F., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd ed., Philadelphia: Lippincott. Williams and Wilkins.St

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

HIMEDIA®

Lead Acetate Paper Strips DD034

Application

Lead Acetate Paper Strips are used for detection of hydrogen sulphide production by microorganisms.

Directions

Inoculate Peptone Water (M028) with the test organism. Insert a Lead acetate paper strip between the plug and inner wall of tube, above the inoculated medium and incubate at 35-37°C for 18-24 hours.

Principle and Interpretation

The lead acetate procedure is more sensitive than any other method for detecting H_2S production. It detects even traces of H_2S. H_2S is a colourless gas which on contact with lead acetate produces lead sulphide, a black precipitate, indicated by a visible black coloured reaction on the Lead acetate paper strip (2). Lead Acetate Paper strips are sterile filter paper strips impregnated with lead acetate reagent. Certain organisms are capable of enzymatically liberating sulphur from sulphur containing aminoacids or inorganic sulphur compounds. Hydrogen sulphide can be produced in small amounts from sulphur containing amino acids like Cysteine by a large number of bacteria in a carbohydrate media (1). This test is used mainly for identification and differentiation of organisms like *Salmonella* species.

Quality Control

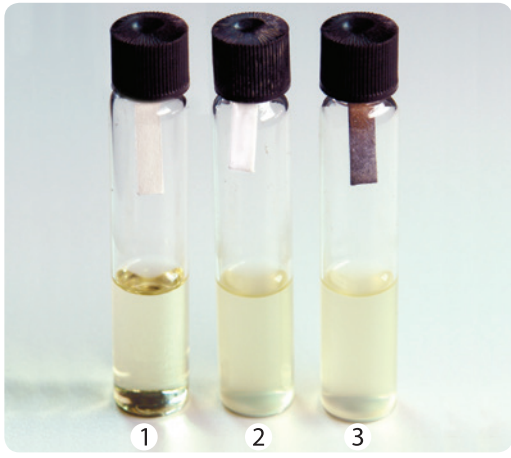
Appearance

Filter paper strips of 70 mm x 5 mm.

Cultural Response

Hydrogen sulphide production by various test organisms is observed after an incubation at 35-37°C for 18-24 hours, by inserting Lead Acetate Paper Strips between the plug and inner wall of tube, above the inoculated Peptone Water (M028).

Organism	Growth	H_2S production
Escherichia coli ATCC 25922	luxuriant	negative reaction, no blackening.
Salmonella Enteritidis ATCC 13076	luxuriant	positive reaction, blackening of the lower portion of the strip.
Salmonella Typhimurium ATCC 14028	luxuriant	positive reaction, blackening of the lower portion of the strip.



M028 Peptone Water with Lead Acetate Paper Strips (DD034)

1. Control
2. *Escherichia coli* ATCC 25922
3. *Salmonella* Typhimurium ATCC 14028

Reference

1. Mackie and MaCartney, 1996, Practical Medical Microbiology, 14th ed., Vol. 2, Collee J.G., Fraser A. G., Marmion B. P., Simmons A. (Eds.), Churchill Livingstone, Edinburgh.
2. MacFaddin JF, (Ed). 2000. Biochemical Tests for Identification of Medical Bacteria. 3rd ed. Philadelphia: Lippincott. Williams & Wilkins.

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

Nitrate Discs DD041

Application

Nitrate discs are used as substrate for detection of nitrate reduction by microorganisms.

Directions

Aseptically put nitrate discs in 5 ml sterile Peptone Water (M028) inoculated with the test microorganisms. Incubate at 35-37°C for 18-24 hours. Add few drops of Nitrate reagents i.e. α-naphthylamine (R009) and Sulphanilic acid (R015). A distinct red or pink colour indicates nitrate reduction. A control (uninoculated) tube should also be tested. If there is no pink colour formation, add a pinch of zinc dust to confirm the absence of nitrate in the medium (3).

Principle and Interpretation

The test involves detection of the enzyme nitrate reductase which causes the reduction of nitrate in the presence of a suitable electron donor to nitrite, which can be tested by an appropriate colorimetric reagent. Almost all *Enterobacteriaceae* reduce nitrate. Nitrate disc contains potassium nitrate as substrate which is broken down to nitrite when nitrate reductase positive culture is grown in presence of these discs. Nitrite production can be detected by using Nitrate Test Reagents-α-naphthylamine (R009) and Sulphanilic acid (R015). Reduction of nitrate (NO_3) to nitrite (NO_2) and subsequently to nitrogen gas (N_2) usually takes place under anaerobic conditions, in which an organism derives its oxygen from nitrate (1). Most facultative anaerobes can reduce nitrate in the absence of oxygen. This anaerobic respiration is an oxidation process in which inorganic substances furnish oxygen to serve as an electron acceptor to provide energy (2). The end product possibilities of nitrate reduction are many depending upon the bacterial species. The more common end product via nitrite reduction is molecular nitrogen (2). Depending upon environmental conditions, these products are usually not further oxidized or assimilated into cellular metabolism, but are excreted into the surrounding medium.

Quality Control

Appearance

Filter paper discs of 6 mm diameter bearing letters 'N' in continuous printing style.

Cultural Response

The Nitrate reduction reaction of various bacteria with Nitrate discs, was observed after an incubation at 35-37°C for 18-24 hours using Peptone Water (M028).

M028 Peptone Water with added Nitrate Discs (DD041)

1. Control
2. *Salmonella* Typhimurium ATCC 14028
3. *Enterobacter aerogenes* ATCC 13048
4. *Escherichia coli* ATCC 25922
5. *Acinetobacter calcoaceticus* ATCC 23055

Organism	Growth	Nitrate Reduction
Escherichia coli ATCC 25922	luxuriant	positive reaction, red or pink colour formation on addition of nitrate test reagents
Enterobacter aerogenes ATCC 13048	luxuriant	positive reaction, red or pink colour formation on addition of nitrate test reagents.
Salmonella Typhimurium ATCC 14028	luxuriant	positive reaction, red or pink colour formation on addition of nitrate test reagents.
Acinetobacter calcoaceticus ATCC 23055	luxuriant	negative reaction

Reference

1. Pelczar M.J. Jr., Reid R.D. (1965), Microbiology, 2nd edn., McGraw-Hill, New York, 567.
2. Stanier R.Y., Douderoff M., Adelberg E.A. (1963), The Microbial World, 2nd edition, Prentice - Hall, 116-117.
3. MacFaddin J. F., (Ed) 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd ed., Philadelphia: Lippincott. Williams and Wilkins

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

HIMEDIA®

Nitrate Reagent Discs (Twin Pack) DD042

Application

Nitrate reagent discs are used for detection of nitrate reduction by microorganisms.

Directions

Grow test culture on a suitable Agar medium plate containing nitrate substrate. Place Part A (disc) on suspected colony. Add a drop or two of Part B (Rehydrating fluid) on the disc. When used in Nitrate Broth, a single disc (part A) is moistened with one or two drops of Part B and added to the tube containing culture incubated for 18-24hours at 35-37°C.

Principle and Interpretation

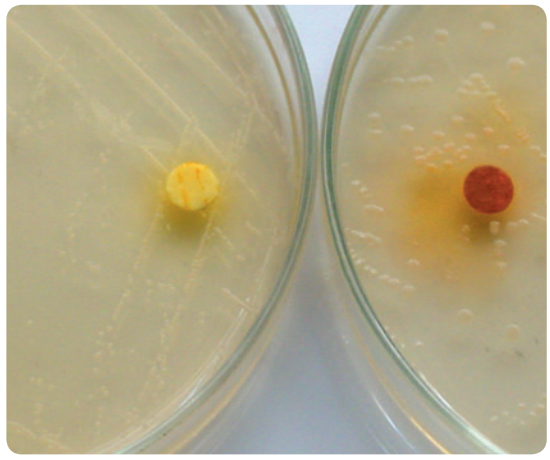
The test involves detection of the enzyme nitrate reductase which causes the reduction of nitrate in the presence of a suitable electron donor to nitrite, which can be tested by an appropriate colorimetric reagent. Almost all Enterobacteriaceae reduce nitrate. Nitrate reagent discs when placed on suspected colony turn red-pink in case of nitrate reduction (positive reaction), when a drop or two of Part B (Rehydrating fluid) is added to the disc. Reduction of nitrate (NO_3) to nitrite (NO_2) and subsequently to nitrogen gas (N_2) usually takes place under anaerobic conditions, in which an organism derives its oxygen from nitrate (1). Most facultative anaerobes can reduce nitrate in the absence of oxygen. This anaerobic respiration is an oxidation process in which inorganic substances furnish oxygen to serve as an electron acceptor to provide energy (2). The end product possibilities of nitrate reduction are many depending upon the bacterial species. The more common end product via nitrite reduction is molecular nitrogen (2). Depending upon environmental conditions, these products are usually not further oxidized or assimilated into cellular metabolism, but are excreted into the surrounding medium.

Quality Control

Appearance

Part A : Filter paper discs of 6 mm diameter bearing letters 'Nr' in continuous printing style.

Part B : Light brown coloured solution, may have black suspended particles



M072 Nitrate Agar with Nitrate Reagent Discs (DD042)

1. *Acinetobacter calcoaceticus* ATCC 23055
2. *Salmonella* Typhimurium ATCC 14028

Cultural Response

The Nitrate reduction reaction was observed after an incubation at 35-37°C for 18-24 hours, for various bacteria with Nitrate Reagent discs (Part A), soaked with a drop of Part B, using Nitrate Broth (M439) / Nitrate Agar (M072).

Organism	Growth	Nitrate Reduction
Escherichia coli ATCC 25922	luxuriant	positive reaction, red or pink colour formation on addition of nitrate test reagents
Enterobacter aerogenes ATCC 13048	luxuriant	positive reaction, red or pink colour formation on addition of nitrate test reagents.
Salmonella Typhimurium ATCC 14028	luxuriant	positive reaction, red or pink colour formation on addition of nitrate test reagents.
Acinetobacter calcoaceticus ATCC 23055	luxuriant	negative reaction

Reference

1. Pelczar M.J. Jr., Reid R.D. (1965), Microbiology, 2nd edn.,McGraw-Hill, New York, 567.
2. Stanier R.Y., Douderoff M., Adelberg E.A. (1963), The Microbial World, 2nd edition, Prentice - Hall, 116-117

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

ONPG Discs DD008

Application

ONPG Discs are used for the rapid detection of ß-galactosidase activity in microorganisms, specially to identify late lactose fermenters quickly.

Directions

Place one ONPG disc in a sterile test tube. Add 0.1 ml of sterile 0.85% w/v sodium chloride solution (physiological saline). Pick up the colony under test with a sterile loop and emulsify it in physiological saline in the tube containing the disc. Incubate at 35-37°C. To detect active lactose fermenters observe the tube at an interval of one hour, upto 6 hours. To detect late lactose fermenters, incubate the tubes upto 24 hours.

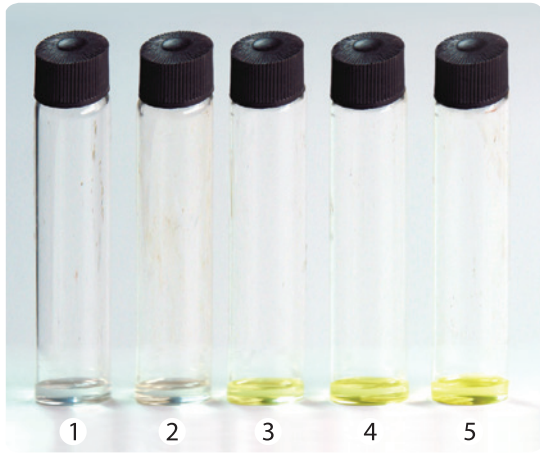
Precautions The reaction speed depends upon the size of inoculum. Use known positive and negative ß-galactosidase producing organisms to monitor the disc reactions.

Principle and Interpretation

ONPG (Ortho-nitrophenyl ß-D-galactopyranoside) is a synthetic colourless compound (galactoside) structurally similar to lactose (1).

ß-galactosidase cleaves ONPG to galactose and o-nitrophenyl, a yellow compound. The ONPG 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 discs are sterile filter paper discs impregnated with ONPG. ONPG 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, ß-galactosidase, hydrolyzes the lactose to yield glucose and galactose. True non-lactose fermenters lack both enzymes; however some organisms lack permease but posses ß-galactosidase. These organisms are late lactose fermenters.



Physiological Saline with ONPG Discs (DD008)

1. Control
2. *Proteus vulgaris* ATCC 13315
3. *Citrobacter freundii* ATCC 8090
4. *Enterobacter aerogenes* ATCC 13048
5. *Escherichia coli* ATCC 25922

Quality Control

Appearance

Filter paper discs of 6 mm diameter bearing letters "On" in continuous printing style.

Cultural Response

DD008: ONPG reaction observed in 0.85% sodium chloride solution of following culture containing ONPG (DD008) disc after an incubation of upto 4 hours at 35-37°C).

Organism	ONPG
Citrobacter freundii ATCC 8090	positive reaction, yellow colour
Enterobacter aerogenes ATCC 13048	positive reaction, yellow colour
Escherichia coli ATCC 25922	positive reaction, yellow colour
Salmonella Choleraesuis ATCC 12011	positive reaction, yellow colour
Proteus vulgaris ATCC 13315	negative reaction, no colour change
Salmonella Typhimurium ATCC 14028	negative reaction, no colour change

Reference

1. Lowe G.H., 1962., J. Med. Lab. Technol., 19:21

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

Optochin Discs DD009

Application

Optochin Discs are used for identification and differentiation of *Streptococcus pneumoniae* and viridans streptococci.

Directions

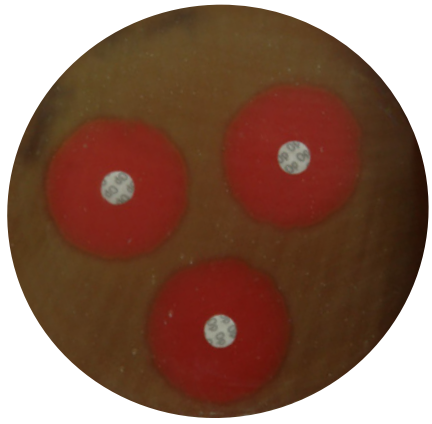
Prepare Soyabean Casein Digest Agar (M290) w/blood or Blood Agar Base (M073) plates and streak pure culture of organism to be tested across one half of the plate. Streak a known *Pneumococcus* culture across the other half of the plate as positive control. Immediately place Optochin discs in the centre of the two halves of the plate and incubate at 35-37°C for 18-24 hours. Following incubation observe for zone of inhibition around the discs.

Principle and Interpretation

Alpha haemolytic (viridans) streptococci and *Pneumococcus* (*Streptococcus pneumoniae*) cannot be easily distinguished on Blood Agar plates as pneumococci strain shows partial clearing of blood and greenish discolouration (α-haemolysis). Optochin is inhibitory for pneumococcal growth whereas other streptococci strains show good growth or a very small zone of inhibition. Bowers and Jeffries have shown a correlation between bile solubility and full Optochin susceptibility for the differentiation of *Streptococcus pneumoniae* from other streptococci (1).

Hence optochin test is a useful diagnostic aid for identification / differentiation of pneumococci and viridans streptococci.

Optochin discs are filter paper discs impregnated with 5 µg of optochin. The test is based on the property of viridans streptococci to grow in the presence of Optochin (ethyl hydrocuprein hydrochloride) which inhibits pneumococci. This test is performed for the diagnosis of penumococcal infections. Specimens of sputum, lung aspirate, pleural fluid, CSF, urine or blood are first examined by Gram's stain, cultured and the isolates are then subjected to Optochin Sensitivity Test.



Optochin Susceptibility Test Discs (DD009)

Streptococcus pneumoniae ATCC 6303

Quality Control

Appearance

Filter paper discs of 6 mm diameter bearing letters "Op" in continuous printing style.

Cultural Response

DD009 : Cultural response observed after an incubation at 35-37°C for 18-24 hours on seeded Soyabean Casein Digest Agar (M290) with added sterile defibrinated sheep blood, using Optochin discs.

Organism	Diameter of zone of inhibition
Streptococcus pneumoniae ATCC 6303	More than or equal to 15mm

Reference

1. Bowers E.F. and Jeffries L.R., 1995, J. Clin. Path., 8:58.

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

Oxidase Discs DD018

Application

Oxidase Discs are used for detection of oxidase production by microorganisms like *Neisseria, Alcaligenes, Aeromonas, Vibrio Campylobacter* and *Pseudomonas*, which give positive reactions and for excluding *Enterobacteriaceae*, which give negative reactions.

Directions

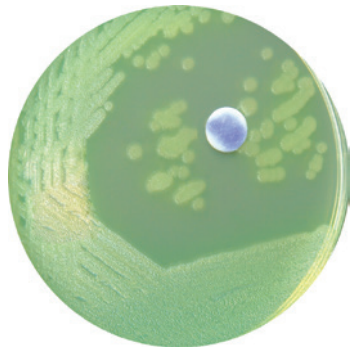
Oxidase reaction is carried out by touching and spreading a well isolated colony on the oxidase disc or by placing the oxidase disc on an isolated colony grown on a non-selective & non-haemoglobin containing medium. The reaction is observed within 5-10 seconds at 25-30°C. A delayed positive reaction appears in 10-60 seconds while a change later than 60 seconds or no change at all is considered negative reaction.

Precautions

1. Do not use stainless steel or nichrome inoculating wires, as false positive reaction may result from surface oxidation products formed during flame sterilization.
2. Growth from media containing dyes, blood or Haemoglobin is not suitable for testing.
3. Timing is critical (5-10 sec) for interpretation of results.
4. Perform oxidase test on all gram-negative bacilli.
5. Cytochrome oxidase production may be inhibited by acid production. False negative reactions may be exhibited by *Vibrio, Aeromonas* and *Plesiomonas* species when grown on a medium containing fermentable carbohydrate e.g. MacConkey Agar (M081). Colonies taken from media containing nitrate may give unreliable results. The loss of activity of the oxidase reagent is caused by auto-oxidation which may be avoided by adding 0.1% ascorbic acid (3).

Principle and Interpretation

Certain bacteria posses either cytochrome oxidase or indophenol oxidase (an iron-containing haemoprotein), which catalyzes the transport of electrons from donor compounds (NADH) to electron acceptors (usually oxygen). In the oxidase test, a colourless dye such as N, N-dimethyl-p-phenylenediamine serves as an artificial electron acceptor for the enzyme oxidase. The dye is oxidized to form indophenol blue, a coloured compound. The test is useful in the initial characterization of aerobic gram-negative bacteria of the genera *Aeromonas, Plesiomonas, Pseudomonas, Campylobacter* and *Pasteurella*. Oxidase discs are sterile filter paper discs impregnated with N, N-dimethyl-p-phenylenediamine oxalate, ascorbic acid and α-naphthol. These discs overcome the neccessity of daily preparation of fresh reagent. Gordon and McLeod (1) introduced oxidase test for identifying gonococci based upon the ability of certain bacteria to produce indophenol blue from the oxidation of dimethyl-p-phenylenediamine and a-naphthol. Gaby and Hadley (2) introduced a more sensitive method by using N, N-dimethyl-p-phenylenediamine oxalate where all staphylococci were oxidase negative. In a positive reaction the enzyme cytochrome oxidase combines with N, N-dimethyl-p-phenylenediamine oxalate and α-naphthol to form the dye, indophenol blue.



Positive Oxidase reaction using Oxidase Discs (DD018)

Pseudomonas aeruginosa ATCC 27853

Quality Control

Appearance

Filter paper discs of 10 mm diameter

Cultural Response

DD018: Typical oxidase reaction given by 18-48 hour culture observed within 5-10 seconds at 25-30°C.

Organism	Reaction Observed
Pseudomonas aeruginosa ATCC 27853	positive, deep purplish blue colouration of disc
Neisseria gonorrhoeae ATCC 19424	positive, deep purplish blue colouration of disc
Escherichia coli ATCC 25922	negative, no colour change
Staphylococcus aureus ATCC 25923	negative, no colour change

Reference

1. Gordon J. and Mcleod J.W., 1928, J. Path. Bact., 31:185
2. Gaby W.L and Hadley C., 1957. J. Bact., 74:356
3. Steel. K.J. 1962. J. Appl. Bact. 25:445

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

HIMEDIA®

Spore Strips (Steam Sterilization Monitor Strips) DD032

Application

Steam Sterilization Monitor Strips are used for evaluating sterilization process. These indicators which are specified by the U.S. military specification MIL-S- 36586 are GMP requirements of U.S. FDA.

Directions

Place indicators in the areas of the pack or load least accessible to steam. Places such as the geometrical center, and the upper and lower regions of both front and rear of the load to be sterilized are considered suitable areas for placement of these indicators. A standard procedure should be established for the routine evaluation of each sterilizer. On completion of the sterilization cycle, remove the indicators from the test loads and deliver them to the laboratory for testing. All sterility tests should be performed in a clean dust free transfer area, preferably under positive air pressure, using rigid aseptic technique throughout the test procedure.

Using sterile scissors, cut open one end of the envelope. Thereafter remove the indicator strip with sterile tweezers and aseptically transfer it to a tube of sterile Soyabean Casein Digest Medium w/ Yeast Extract and Ferric pyrophosphate (M207) or Soyabean Casein Digest Medium (M011). Incubate the tubes for seven days at 55-60°C. Observe the tubes daily. If turbidity develops, failure of the sterilization process is indicated.

Precautions

The spore strips or broth cultures of *Bacillus stearothermophilus* must be autoclaved at 121°C for at least 30 minutes prior to discarding.

Each spore strip is individually packaged in a steam-permeable envelope.

Principle and Interpretation

Bacillus stearothermophilus is a thermophilic bacteria which can grow at 55°C and above. The spores are highly heat resistant and are used to monitor autoclave performance (1).

Sterilization is the freeing of an article from all living organisms including viable spores (1). Sterilization quality control can only be achieved through the use of calibrated biological indicators (endospores). These indicators consist of *Bacillus stearothermophilus* spores impregnated on chromatography paper strips, individually placed into envelopes. Number of spores present per stri: 10^6. These organisms are difficult to destroy because they

are more resistant to heat than other vegetative bacteria and viruses. Therefore, if they are destroyed during sterilization, it is assumed that all other life forms are also destroyed. This test is considered the most sensitive check of the autoclave's efficiency.

Quality Control

Appearance

Filter paper strip impregnated with spores of standard culture of *B.stearothermophilus*.

Number of spores

10^6 spores/strip

Cultural Response

Sterility checking of the autoclave was carried out using Spore strip. After autoclaving, strip was inoculated in 100ml of sterile Soyabean Casein Digest Medium (M011) and incubated at 55°C for upto 7 days. An unexposed spore strip was also inoculated separately in 100ml of Soyabean Casein Digest Medium (M011)

	Unexposed Spore Strip	Exposed Spore Strip	Positive control	Negative control
Growth of Spore strips in M011	luxuriant	no growth	luxuriant	no growth

Reference

1. Mackie and McCartney, 1996, Practical Medical Microbiology, 14th ed., Vol. 2, Collee J. G., Fraser A. G., Marmion B, P., Simmons A (Eds.), Churchill Livingstone, Edinburgh.

Storage and Shelf-Life

Store between 15-27°C. Use before expiry date on the label.

Spore Strips (Radiation Sterilization Monitor Strips) DD039

Application

Radiation Sterilization Monitor Strips are used for evaluating radiation sterilization process. These indicators which are specified by the U.S. military specification MIL-S-36586 are GMP requirements of U.S. FDA.

Directions

Place indicators in the areas of the pack or load least accessible to radiation. Places such as the geometrical center, and the upper and lower regions of both front and rear of the load to be sterilized are considered suitable areas for placement of these indicators. A standard procedure should be established for the routine evaluation of each sterilizer. On completion of the sterilization cycle, remove the indicators from the test loads and deliver them to the laboratory for testing. All sterility tests should be performed in a clean dust free transfer area, preferably under positive air pressure, using rigid aseptic technique throughout the test procedure.

Using sterile scissors, cut open one end of the envelope. Thereafter remove the indicator with sterile tweezers and aseptically transfer it to a tube of sterile Soyabean Casein Digest Medium w/Yeast Extract & Ferric Pyrophosphate (M207) or Soyabean Casein Digest Medium (M011). Incubate the tubes for seven days at 35-37°C. Observe the tubes daily. If turbidity develops, failure of the radiation sterilization process is indicated.

Precautions

The spore strips or broth cultures of *Bacillus pumilus* must be autoclaved at 121°C for at least 30 minutes prior to discarding.

Principle and Interpretation

Bacillus pumilus is a radiation resistant species. The spores are highly radiation resistant and are used to monitor radiation sterilization (1).

Sterilization is the freeing of an article from all living organisms including viable spores (1). Radiation sterilization quality control can only be achieved through the use of calibrated biological indicators (endospores). These indicators consist of *Bacillus pumilus* spores impregnated on chromatography paper strips, individually placed into envelopes. Number of spores present per strip : 10^6. These organisms are difficult to destroy since they are more resistant to radiation than other vegetative bacteria and viruses. Therefore, if they are destroyed during sterilization, it is assumed that all other life forms are also destroyed. This test is considered the most sensitive check of efficiency of radiation sterilization.

The spore strips or broth cultures of *Bacillus pumilis* must be autoclaved at 15 lbs pressure (121°C) for atleast 30 minutes prior to discarding.

Quality Control

Appearance

Filter paper strip impregnated with spores of standard culture of *B. pumilus*

Number of spores

10^6 spores/strip

Cultural Response

Spore strip exposed to 2.5 Mrad of radiation was inoculated in 100ml of sterile Soyabean Casein Digest Medium (M011) & incubated at 35-37°C upto 7 days. Simultaneously unexposed spore strip was inoculated in another 100ml of Soyabean Casein Digest Medium M011

	Unexposed Spore Strip	Exposed Spore Strip	Positive control	Negative control
Growth of Spore strips in M011	luxuriant	no growth	luxuriant	no growth

Reference

1. Mackie and McCartney, 1996, Practical Medical Microbiology, 14th ed., Vol. 2, Collee J. G., Fraser A. G., Marmion B. P., Simmons A (Eds.), Churchill Livingstone, Edinburgh.

Storage and Shelf-Life

Store at 15-27°C. Use before expiry date on the label.

HIMEDIA®

X Factor/ V Factor/ X+V Factor DD020/DD021/DD022

Application

X Factor, V Factor, X+V Factor discs are used for the presumptive identification of *Haemophilus* species on the basis of their requirements for X or V factors or both.

Directions

Inoculate the surface of a Blood Agar Base (M073) plate or Brain Heart Infusion Agar (M211) plate with the test organisms by either streaking or surface spreading.

Aseptically place the X, V and X+V factor discs on the plate, in the following positions:

Disc position on the Agar plate	
X factor disc	12 O' clock
V factor disc	4 O' clock
X+V factor disc	8 O' clock

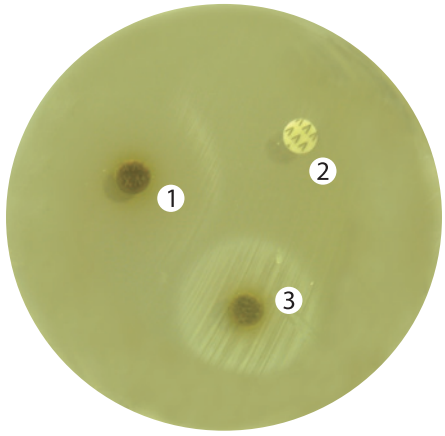
Incubate the plates at 35-37°C for 24-48 hours. Observe for growth in the neighbourhood of the disc.

Precautions

Use known strains of *Haemophilus influenzae* to monitor the performance of the differentiation discs and the medium. Do not use heavy suspension of the test organisms as X or V factor carryover from the primary growth medium may take place.

Principle and Interpretation

Both X and V factor are growth factors required by certain organisms eg. *Haemophilus* species and for enhanced growth of *Neisseria* species. The X factor (hemin) and V factor (Coenzyme-diphosphopyridine nucleotide) are impregnated on sterile filter paper discs of 6 mm diameter. The test organism requiring X factor alone, grow only in the vicinities of X and X+V factor discs. Those which require V factor alone grow in the vicinities of V and X+V factor discs. If both X and V factors are required, then the organism will grow only in the vicinity of the X+V factor discs. Thus satellite growth is seen around the disc promoting growth (1).



1) X Factor (DD020)
2) V Factor (DD021)
3) X+V Factor (DD022)

Haemophilus influenzae ATCC 35056

X, V and X+V factor discs are sterile filter paper discs impregnated with growth factors which are used for differentiating *Haemophilus* species. *Bordetella* and *Haemophilus* species can be identified on the basis of their requirement for X and V growth factors in the basal medium.

Members of the genus *Haemophilus* require hemin (X factor) and/ or nicotinamide-adenine-dinucleotide (V factor). Together with the X factor and the V factor, the need for either one or both factors provides the main means of differentiation of these organisms. *Haemophilus* species requiring both X and V factors exhibit growth only in the vicinity of the X + V factor discs.

Quality Control

Appearance

DD020: Filter paper discs of 6 mm diameter bearing letters "X" in continuous printing style.
DD021: Filter paper discs of 6 mm diameter bearing letters "V" in continuous printing style.
DD022: Filter paper discs of 6 mm diameter bearing letters "X+V" in continuous printing style.

Cultural Response

DD020: Cultural characteristics observed on Brain Heart Infusion Agar (M211) or Blood Agar Base (M073) after an incubation at 35-37°C for 24-48 hours.
DD021: Cultural characteristics observed on Brain Heart Infusion Agar (M211) or Blood Agar Base (M073) after an incubation at 35-37°C for 24-48 hours.
DD022: Cultural characteristics observed on Brain Heart Infusion Agar (M211) or Blood Agar Base (M073) after an incubation at 35-37°C for 24-48 hours.

Organism	Growth without growth factor	Growth with X+V factor (DD022)	Growth with V factor (DD021)	Growth with X factor (DD020)
DD020/ DD021/ DD022				
Bordetella pertussis ATCC 8467	positive (initial isolation on Bordet Gengou Agar (M175)	positive (initial isolation on Bordet Gengou Agar (M175)	positive (initial isolation on Bordet Gengou Agar (M175)	positive (initial isolation on Bordet Gengou Agar (M175)
Haemophilus influenzae ATCC 35056	negative (no growth)	positive (growth)	negative (no growth)	negative (no growth)
Haemophilus parainfluenzae ATCC 7901	negative (no growth)	positive (growth)	positive (growth)	negative (no growth)
Haemophilus haemoglobinophilus ATCC 19416	negative (no growth)	positive (growth)	negative (no growth)	positive (growth)

Reference

1. Murray PR, Baron EJ, Jorgensen J H, Pfaller M A, Yolken R H (Eds.),8th ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.

Storage and Shelf-Life

Store X factor at 2-8°C. For prolonged use store at -20°C and V & X+V factor discs below -10°C. Use before expiry date on the label.

Vibrio 0129 Differential Disc (10mcg / 150mcg) DD047/ DD048

Application

For differentiation of *Vibrio* species based on sensitivity to Vibriostatic agent O129.

Directions

With a sterile swab, streak the pure, fresh culture of the test organism from sample on a non-selective Blood Agar Plate (containing 0.5% NaCl). Aseptically place both Vibrio O129 differential disc [10 mcg, (DD047) and 150 mcg (DD048)] on the swabbed plates. Incubate at 35 – 37°C for 24 hours. Observe for zones of inhibition.

Principle and Interpretation

Shewan and Hodgkiss recognized the sensitivity of *Vibrio* to the vibrio-static agent O129 (2,4-diamino-6,7-di-isopropylpteridine phosphate) (1). O129 was found to be useful in the differentiation of *Vibrio* from other gram-negative bacteria especially *Aeromonas*, which are characteristically resistant to O129(2). Even among the genus *Vibrio*, different species show different sensitivities to O129(3); hence two different concentration discs are to be simultaneously tested to determine the degree of sensitivity of the species. O129 discs of two concentrations are available: 10-µg and 150-µg. Methods for standardized disc antimicrobial susceptibility testing are employed, with any zone of inhibition around O129 disks being regarded as sensitive. Medium to be used should be supplemented with 0.5% Sodium Chloride, as sodium ions stimulate the growth of all *Vibrio* species and are required by most.

Interpret the results as follows

Sensitive - Zone of inhibition around both 10µg (DD047) and 150µg (DD048) disc.

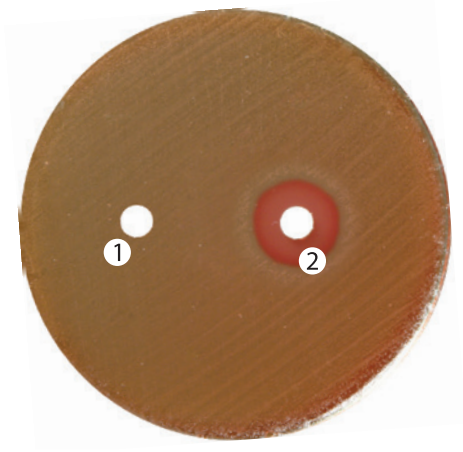
Resistant - No zone of inhibition around both the disc (DD047) and (DD048)
Partially sensitive - Zone of inhibition around
150µg (DD048) disc and no zone around 10µg (DD047) disc.

Quality Control

Appearance

DD047 : Vibrio O129 differential filter paper discs of 6 mm diameter containing 10mcg concentration.

DD048 : Vibrio O129 differential filter paper discs of 6 mm diameter containing 150mcg concentration.



1) Vibrio O129 Differential Disc - 10mcg (DD047)
2) Vibrio O129 Differential Disc - 150mcg (DD048)

Vibrio parahaemolyticus ATCC 17802 (Partially sensitive : Showing zone against DD048 & No zone with DD047)

Cultural Response

Cultural response observed on seeded non-selective Blood Agar Plates, with placed Vibrio O129 discs of 10 mcg (DD047) & 150 mcg (DD048), after incubation at 35-37°C for 24 hours.

Organism	Results
Vibrio parahaemolyticus ATCC 17802	Partially sensitive
Aeromonas hydrophila ATCC 7966	Resistant

Reference

1. Shewan JM, Hodgkiss W. Nature 1954; 63:208-9.
2. Isenberg HD, Ed. Clinical microbiology procedures handbook, Vol 1. Washington, DC: ASM, 1994.
3. Murray PR, Baron EJ, Pfaller MA, Tenover FC, Yolken RH, Eds. Manual of clinical microbiology. 7th ed. ASM, 1999.
4. MacFaddin JF. Biochemical tests for the identification of medical bacteria, 3rd ed. Williams & Wilkins, 2000.

Storage and Shelf-Life

Store at 2-8°C. For prolonged use store at -20°C. Use before expiry date on the label.

HiBio-ID Reader (LA686)

- For accurate reading of results obtained on following KB and KBM strips after inoculation
- Culture will be Identified till species level
- Saves time, material and minimises manual error

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Biochemical Identification Test Kits	
HiIMViC™ Biochemical Test Kit a combination of 12 tests for differentiation of *Enterobacteriaceae* species.	KB001
HiAssorted™ Biochemical Test Kit a combination of 12 tests for identification of Gram-negative rods.	KB002
Hi25™ Enterobacteriaceae Identification Kit a combination of 25 tests for identification of *Enterobacteriaceae* species.	KB003
HiStaph™ Identification Kit a combination of 12 tests for identification of *Staphylococcus* species.	KB004
HiStrep™ Identification Kit a combination of 12 tests for identification of *Streptococcus* species.	KB005A
HiCandida™ Identification Kit a combination of 12 tests for identification of *Candida* species.	KB006
HiVibrio™ Identification Kit a combination of 12 tests for identification of *Vibrio* species.	KB007
HiNeisseria™ Identification Kit a combination of 12 tests for identification of *Neisseria* species.	KB008
HiCarbo™ Kit a combination of 36 tests for utilization of carbohydrate tests. Kit contains Part A, Part B & Part C	KB009
HiCarbo™ Kit- Part A	KB009A
HiCarbo™ Kit- Part B	KB009B1
HiCarbo™ Kit- Part C Note : KB009 is available as a total kit and also individually as Part A, Part B & Part C	KB009C
HiE. coli™ Identification Kit a combination of 12 tests for identification of *E. coli*.	KB010
HiSalmonella™ Identification Kit a combination of 12 tests for identification of *Salmonella* species.	KB011
HiListeria™ Identification Kit a combination of 12 tests for identification of *Listeria* species.	KB012A
HiBacillus™ Identification Kit a combination of 12 tests for identification of *Bacillus* species.	KB013
HiAcinetobacter™ Identification Kit a combination of 12 tests for identification of *Acinetobacter* species.	KB014
Motility Biochemical Test Kits	
HiMotility™ Biochemical Kit for E.coli a combination of 12 tests for confirmation of *E. coli* based on motility and other biochemical tests.	KBM001
HiMotility™ Biochemical Kit for Salmonella a combination of 12 tests for confirmation of *Salmonella* based on motility and other biochemical tests.	KBM002
HiMotility™ Biochemical Kit for Listeria a combination of 12 tests for confirmation of *Listeria* based on motility and other biochemical tests.	KBM003A

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- Phenylalanine deamination
- Urea utilization
- Malonate utilization
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- Nitrate reduction
- Pyrrolidonyl-b-Naphthylamide hydrolysis (PYR)
- Esculin hydrolysis
- H_2S production
- Motility test
- Catalase test
- Oxidase test
- Salt tolerance test
- ONPG test
- Alkaline phosphatase

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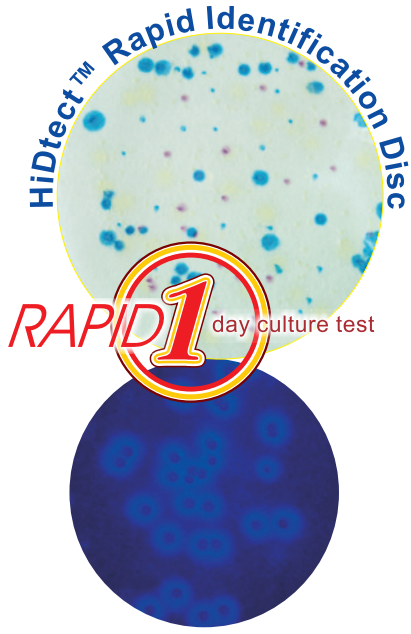

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