Instructions For Use
ENGLISH

Nutrient Agar ISO 16266

Medium for cultivating non-fastidious organisms and confirming *Pseudomonas aeruginosa*, according to ISO 16266.

DESCRIPTION

Nutrient Agar ISO 16266 is a medium used for the cultivation of non-fastidious organisms from water samples.

This medium is formulated according to ISO 16266 for the detection and enumeration of *Pseudomonas aeruginosa* in water by the membrane filtration technique.

TYPICAL FORMULA*	(g/litre)
Peptone	5.0
Meat Extract	1.0
Yeast Extract	2.0
Sodium Chloride	5.0
Agar	15.0
Final pH 7.4 ± 0.2 at 25°C	

*Adjusted and/or supplemented as required to meet performance specifications.

METHOD PRINCIPLE

Peptone and meat extract provide amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Yeast extract is a source of vitamins, particularly of B-group. Sodium chloride maintains the osmotic balance of the medium. Agar is the solidifying agent.

PREPARATION

Dehydrated medium	Suspend 28 g of the powder in 1 liter of distilled or deionized water. Mix well. Heat to boil shaking frequently until completely dissolved. Sterilize in autoclave at 121°C for 15 minutes.
Medium in bottles	Melt the content of the bottle in a water bath at 100°C (loosing the cap partially removed) until completely dissolved. Then screw the cap and check the homogeneity of the dissolved medium, if it is the case turning the bottle upside down. Cool at 45-50°C, mix well avoiding foam formation and aseptically distribute into Petri dishes.

TEST PROCEDURE

According to ISO 16266, transfer the membrane and presumptive *Pseudomonas aeruginosa* to the plate medium.

Incubate aerobically at 36 ± 2°C for 20-24 hours.

Alternatively, the medium can be inoculated by spread plating or direct streaking of the sample over the agar surface.

INTERPRETING RESULTS

Observe for colony growth. Confirm *P. aeruginosa* by performing the oxidase test (ref. 88029).

STORAGE

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed. Store bottles, tubes and prepared plates at 10-25°C away from light. Do not use the product beyond its expiry date on the label or if product shows any evidence of contamination or any sign of deterioration.

Avoid quick temperature shifts of plated medium to prevent condensation.

SHELF LIFE

Dehydrated medium: 4 years.
Medium in bottles: 2 years.
Medium in slant tubes: 1 year.
Ready-to-use plates: 6 months.

QUALITY CONTROL

Appearance of Dehydrated Medium: Free-flowing, homogeneous, beige.

Appearance of Prepared Medium: Slightly opalescent, light amber.

Expected Cultural Response:

Control strain		Inoculum	Incubation	Specification
Pseudomonas aeruginosa	WDCM 00025 (ATCC 27853, NCTC 12903)	50-100 CFU	20-24 h 36 ± 1°C	Good growth
Escherichia coli	WDCM 00013 (ATCC 25922, NCTC 12241)			

Please refer to the actual batch related Certificate of Analysis (CoA).

WARNING AND PRECAUTIONS

For professional use only. Operators must be trained and have certain experience in the laboratory methods. Please read the instructions carefully before using this product. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this document.

Consult the Safety Data Sheet (SDS) for information regarding hazards and safe handling practices.

DISPOSAL OF WASTE

Disposal of waste must be carried out according to national and local regulations in force.

BIBLIOGRAPHY

See the references at the end of this document.

TABLE OF SYMBOLS

See the table of symbols at the end of this document.

The product is available in the various configurations listed below. There may be additional product ref. numbers as well. For an updated listing of available products, visit **liofilchem.com**

Product	Format	Packaging	Ref.
Nutrient Agar ISO 16266	Plate 90 mm	20 plates	10044
Nutrient Agar ISO 16266	Slant tube	10 x 7 ml	30083
Nutrient Agar ISO 16266	Bottle	6 x 100 ml	402190
Nutrient Agar ISO 16266	Bottle	6 x 200 ml	412190
Nutrient Agar ISO 16266	Bottle	6 x 500 ml	470060
Nutrient Agar ISO 16266	Dehydrated media	100 g	620036
Nutrient Agar ISO 16266	Dehydrated media	500 g	610036
Nutrient Agar ISO 16266	Dehydrated media	5 kg	6100365

This IFU document and the SDS are available from the online Support Center:

liofilchem.com/ifu-sds

Chromatic™ Vibrio

Chromogenic medium for detection of enteropathogenic *Vibrio* spp, from clinical and nonclinical samples.

DESCRIPTION

Chromatic™ Vibrio is a chromogenic medium used for the selective isolation and cultivation of vibrios, including *V. cholerae*, *V. parahaemolyticus V. vulnificus* and *V. alginolyticus* from stool specimens, food, water and environmental samples.

TYPICAL FORMULA	(g/l)
Peptone	15.0
Yeast Extract	3.0
Salts	59.1
Chromogenic Mix	0.3
Agar	15.0
Final pH 8.4 ± 0.2 at 25°C	

METHOD PRINCIPLE

Peptone and yeast extract provide amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Salts included in the medium have the dual effect of stimulating vibrios growth and suppressing Gram-positive bacteria and coliforms. The alkaline pH is also inhibitory for most contaminant microorganisms while enhances the recovery of *V. cholerae*. Chromogenic mix allows to identify the *Vibrio* genus on the basis of the color and morphology of the colonies. Agar is the solidifying agent.

PREPARATION

Dehydrated medium: Suspend 92.4 g of the powder in 1 liter of distilled or deionized water. Mix well. Heat to boil shaking frequently until completely dissolved. DO NOT AUTOCLAVE.

TEST PROCEDURE

Inoculate the medium by direct streaking or spread plating.
Incubate aerobically at 35 ± 2°C for 18-24 hours.

NB. Heavy inoculation is recommended. Swabs containing specimen material should be transported to the laboratory in Cary Blair Transport Medium (ref. 470290) if a delay in reaching the laboratory is anticipated. Specimens for cultivation of vibrios should not be frozen.

INTERPRETING RESULTS

After incubation observe the color of the colonies and interpret the results as indicated in the ID table. Confirm typical colonies with proper biochemical tests.

ID Table.

Microorganism	Typical colony color
V. parahaemolyticus	Mauve
V. vulnificus / V. cholerae	Green blue to turquoise blue
V. alginolyticus	Colorless

APPEARANCE

Dehydrated medium: free-flowing, homogeneous, light beige to green beige.
Prepared medium: clear to slightly opalescent, green.

STORAGE

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed. Store prepared plates at 2-8°C away from light. Do not use the product beyond its expiry date on the label or if product shows any evidence of contamination or any sign of deterioration.

SHELF LIFE

Dehydrated medium: 2 years.
Ready-to-use plates: 4 months.

QUALITY CONTROL

Plates are inoculated with the microbial strains indicated in the QC table.
Inoculum for productivity: 50-100 CFU.
Inoculum for selectivity: 10^4-10^6 CFU.
Incubation conditions: aerobically at 35 ± 2°C for 18-24 hours.

QC Table.

Microorganism		Growth	Specification
Vibrio vulnificus	ATCC® 27562	Good	Green colonies
Vibrio parahaemolyticus	ATCC® 17802	Good	Mauve colonies
Vibrio alginolyticus	ATCC® 17749	Good	Creamy colonies
Escherichia coli	ATCC® 25922	Inhibited	---
Staphylococcus aureus	ATCC® 25923	Inhibited	---

WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. It is nevertheless recommended to consult the safety data sheet for its correct use. The product is intended for *in vitro* diagnostic use and must be used only by properly trained operators.

DISPOSAL OF WASTE

Disposal of waste must be carried out according to national and local regulations in force.

BIBLIOGRAPHY

1. American Public Health Association (1992): compendium of methods for the microbiological examination of foods, 3rd edition.
2. Dewitt, W.E., E.J. Gangarosa, I. Huq, and A. Zarifi (1971) Holding media for the transport of *Vibrio cholerae* from field to laboratory. Am. J. Trop. Med. Hyg. 20:685-688.
3. Kobayashi, T., S. Enomoto, R. Sakazaki, and S. Kuwahara (1963) A new selective medium for pathogenic vibrios: T.C.B.S. Agar (Modified Nakanishiís Agar). Jap. J. Bacteriol. 18:387-391.

PRESENTATION		Contents	Ref.
Chromatic™ Vibrio	90 mm ready-to-use plates	20 plates	11633
Chromatic™ Vibrio	Dehydrated medium	500 g of powder	610633

TABLE OF SYMBOLS

LOT Batch code	**IVD** *In Vitro* Diagnostic Medical Device	Manufacturer	Use by	Fragile, handle with care
REF Catalogue number	Temperature limitation	Contains sufficient for <n> tests	Caution, consult Instruction For Use	Do not reuse

LIOFILCHEM® s.r.l.
Via Scozia zona ind.le, 64026 Roseto degli Abruzzi (Te) Italy
Tel. +39 0858930745 Fax +39 0858930330 www.liofilchem.net liofilchem@liofilchem.net

CE IVD

CLOSTRIDIUM DIFFICILE AGAR

Medium for the isolation of C*lostridium difficile*.

TYPICAL FORMULA	(g/l)
Proteose Peptone	40.0
Fructose	6.0
Disodium Hydrogen Phosphate	5.0
Potassium Dihydrogen Phospahate	1.0
Magnesium Sulphate	0.1
Sodium Chloride	2.0
Agar	15.0
Final pH 7.4 ± 0.2 at 25°C	

DESCRIPTION

CIOSTRIDIUM DIFFICILE AGAR is a medium used for the isolation of *Clostridium difficile*.

PRINCIPLE

Proteose peptone provides nitrogen, vitamins, minerals and amino acid essential for growth. Fructose is the fermentable carbohydrate used to facilitate the recovery and growth of *C. difficile*. Potassium dihydrogen phospahate and disodium hydrogen phosphate act as buffering system. Magnesium sulphate is source of magnesium ions essential for several enzymatic reactions and DNA duplication. Sodium chloride maintains the osmotic balance of the medium. This basal medium needs to be added with horse blood that supplies essential growth factors and selective agents to inhibit the growth of most of the microorganisms present in fecal sample other than *C. difficile*.

PREPARATION

Suspend 34.6 g of powder in 500 ml of distilled water. Heat until completely dissolved. Autoclave at 121°C for 15 minutes. Cool to 45-50°C. Aseptically add 1 vial of CLOSTRIDIUM difficile *Supplement* (ref. 81007) previously reconstituted with 2 ml of sterile distilled water and 35 ml of HORSE BLOOD DEFIBRINATED (ref. 83395). Mix gently and dispense in Petri dishes.

TECHNIQUE

Direct inoculum of fecal samples

Lightly inoculate the medium with fecal sample spreading part of the original inoculum in order to obtain well separated colonies. Incubate the plates at 36+/-1°C for 18-24 hours anaerobically.

Treatment for alcohol shock

Mix equal part of absolute ethyl alcohol and the fecal specimen. Homogenize using a vortex mixer. Leave at room temperature of 1 hour. Inoculate the medium and incubate the plates at 36+/-1°C for 18-24 hours anaerobically.

INTERPRETATION OF RESULTS

C. difficile grows with whitish opaque colonies of 4-6mm diameter irregular.

STORAGE

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed and use it before the expiry date on the label or until sings of deterioration or contamination are evident. Store prepared plates at 2-8°C away from light.

WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. it is nevertheless recommended to consult the safety data sheet for its correct use. The product must be used only by properly trained operators.

DISPOSAL OF WASTE

Disposal of waste must be carried out according to national and local regulations in force.

REFERENCES

1. Levett (1985) J. Clin. Pathol. 38: 233-234.
2. Barlett,. J.G. et al. (1978) N. Eng. J. Med., 298 , 531.
3. Boriello, S.P. et al (1981) J. Antimicrob. Chemother. 7 Supp. A. 53-62.
4. George, R.H. et al (1976) J. Clin. Microbiol. 6, 214-219.

LIOFILCHEM® S.r.l.
Via Scozia, Zona Ind.le - 64026, Roseto degli Abruzzi (TE) - ITALY
Tel +39 0858930745 Fax +39 0858930330 Website: www.liofilchem.net E-mail: liofilchem@liofilchem.net

CE IVD

PRODUCT SPECIFICATIONS

NAME
CLOSTRIDIUM DIFFICILE AGAR

PRESENTATION
Dehydrated medium

STORAGE
10-30°C

PACKAGE

Ref.	Content	Packaging
610115	500 g	500 g of powder in plastic bottle
620115	100 g	100 g of powder in plastic bottle

pH OF THE MEDIUM
7.4 ± 0.2

USE
CIOSTRIDIUM DIFFICILE AGAR is a medium used for the isolation of *Clostridium difficile*

TECHNIQUE
Refer to technical sheet of the product

APPEARANCE OF THE MEDIUM
Dehydrated medium
Appearance: free-flowing, homogeneous
Colour: beige

Prepared medium
Appearance: opalescent
Colour: cherry red

SHELFLIFE
4 years

QUALITY CONTROL
1. Control of general characteristics, label and print

2. Microbiological control
   Inoculum for productivity: 10-100 CFU/ml
   Inoculum for selectivity: 10^4-10^5 CFU/ml
   Inoculum for specificity: ≤10^4 CFU/ml
   Incubation conditions:18-24 h at 36 ± 1°C anaerobically

Microorganism		Growth
Escherichia coli	ATCC® 25922	Inhibited
Clostridium difficile	ATCC® 9869	Good

TABLE OF SYMBOLS

LOT Batch code	IVD *In vitro* diagnostic medical device	Manufacturer	Use by	Fragile, handle with care
REF Catalogue number	Temperature limitation	Contains sufficient for <n> tests	Consult instruction for use	Do not reuse
Keep away from heat sources				

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Via Scozia, Zona Ind.le - 64026, Roseto degli Abruzzi (TE) - ITALY
Tel +39 0858930745 Fax +39 0858930330 Website: www.liofilchem.net E-mail: liofilchem@liofilchem.net

CE IVD

ENGLISH

CLOSTRIDIUM difficile *Supplement*

Selective supplement for the enrichment of the medium CLOSTRIDIUM difficile AGAR BASE
for the isolation of *Clostridium difficile*

DESCRIPTION

CLOSTRIDIUM difficile *Supplement* is a selective supplement for the isolation of *Clostridium difficile,* made of a una freeze-dried D-Cicloserin and Cefoxitin mixture. CLOSTRIDIUM difficile *Supplement* is used for the selective enrichment of CLOSTRIDIUM difficile AGAR BASE medium code 610115 or 620115.

PACKAGE CONTENTS

Each package contains:

- 10 bottles of freeze-dried CLOSTRIDIUM difficile *Supplement*
- 1 instruction sheet

PRINCIPLE OF THE METHOD

The selectivity of CLOSTRIDIUM difficile *Supplement,* set by Levett, is due to the activity of D-Cicloserin and Cefoxitin which inhibit the growth of most of Enterobacteriaceae, such as *Enterococcus faecalis*, stafilococci, Gram-negative anaerobic non sporigens bacilli and some strains of Clostridia except for *C. difficile*.

COMPOSITION

CLOSTRIDIUM difficile *Supplement*		
	Content / bottle	*Content / l of medium*
Cicloserin	125.0 mg	250.0 mg
Cefoxitin	4.0 mg	8.0 mg

TEST PROCEDURE

1. Reconstitute aseptically the content of one bottle of CLOSTRIDIUM difficile *Supplement* with 2 ml of sterile distilled water. Shake until completely dissolved, avoiding foam formation.
2. Add aseptically the entire content of one bottle (2 ml) to 500 ml of CLOSTRIDIUM difficile Agar Base medium code 610115-620115 autoclaved, cooled at 45-50 °C and with the addition of 7% of defibrinated horse blood (in this case preferred to ram's blood).
3. Mix with care.
4. Distribute into Petri dishes.

TECHNIQUE AND INTERPRETATION OF RESULTS

Refer to the technical documentation for CLOSTRIDIUM difficile AGAR BASE code 610115 or 620115.

QUALITY CONTROL

1. Control of the appearance: freeze-dried product, white colour.
2. Microbiological control .

Prepare the plates using as base CLOSTRIDIUM difficile AGAR BASE code 610115 or 620115 supplemented with CLOSTRIDIUM difficile *Supplement* (1 bottle in 500 ml of medium) and with 7% of defibrinated horse blood. Plates are inoculated with the strains indicated in the microbiological control table.
Incubation conditions: 24 h at 36 ± 1 °C, in anaerobiosis.
Microbiological control:

Control strains		**Growth**
Clostridium difficile	ATCC 11204	Good
Escherichia coli	ATCC 25922	Inhibited

PRECAUTIONS

The product CLOSTRIDIUM difficile *Supplement* is classifiable as hazardous under current legislation; it is recommended that the Safety Data Sheet be consulted on its use.
CLOSTRIDIUM difficile *Supplement* is a selective supplement to be used only for *in vitro* diagnostic use. It is intended for use in a professional environment and must be used in the laboratory by properly trained personnel, using approved asepsis and safety methods for handling pathogenic agents.

STORAGE

Store CLOSTRIDIUM difficile *Supplement* at 2-8 °C in its original packaging. In such conditions CLOSTRIDIUM difficile *Supplement* will remain valid until the expiry date indicated on the label. Do not use beyond that date. Eliminate without using if there are signs of deterioration.

REFERENCES

- Levett (1985) J. Clin. Pathol. 38. 233-234.
- Hall, I. and O'Toole, E. (1935) Am. J. Dis. Child. 49. 390.

PRESENTATION

Product	REF	Σ
CLOSTRIDIUM difficile *Supplement*	81007	10 bottles

One bottle is sufficient to prepare 500 ml of medium.

TABLE OF SYMBOLS

IVD *In Vitro* Diagnostic Medical Device	Do not reuse	Manufacturer	Contains sufficient for <n> tests	Temperature limitation
REF Catalogue number	Fragile, handle with care	Use by	Caution, consult accompanying documents	LOT Batch code

LIOFILCHEM Bacteriology Products
Via Scozia Zona Ind.le - 64026 Roseto D.A. (TE) - Italy
Tel. +390858930745 Fax +390858930330 Website: *www.liofilchem.net* E-Mail: *liofilchem@liofilchem.net*

CE IVD

Rev.0 / 06.04.2005

ENGLISH

POTASSIUM TELLURITE 1% *Supplement*

Selective supplement for the isolation of Staphylococci

DESCRIPTION

POTASSIUM TELLURITE 1% *Supplement* is a selective supplement consisting of a 1% potassium tellurite aqueous solution for microbiological use, to be used in preparation of VOGEL JOHNSON AGAR culture medium (REF. 610186 or 620186) for isolation of Staphylococci and in other culture media the composition of which provides for the inclusion of potassium tellurite.

KIT CONTENTS

Each kit contains:

- bottles containing 10 ml of POTASSIUM TELLURITE 1% *Supplement*
- 1 Instruction sheet

PRINCIPLE OF THE METHOD

POTASSIUM TELLURITE 1% *Supplement* is a selective supplement used in preparation of the VOGEL JOHNSON AGAR medium (REF. 610186 or 620186) for isolation of Staphylococci. These micro-organisms, which reduce the tellurite to tellurium, grow with grey-black colonies. Potassium tellurite is also included in the composition of other culture media.

COMPOSITION

POTASSIUM TELLURITE 1% *Supplement*	
Contents / bottle	
Potassium tellurite	100.0 mg
Distilled water	10.0 ml

PROCEDURE FOR USE

1. Aseptically add the entire contents of a bottle of POTASSIUM TELLURITE 1% *Supplement* (10 ml) to 500 ml of VOGEL JOHNSON AGAR medium (REF. 610186 or 620186) autoclaved and cooled to 45-50°C. When potassium tellurite is included in the composition of other media, refer to the specific instructions for the medium concerned on the quantity of POTASSIUM TELLURITE 1 % *Supplement* that should be added to it.
2. Mix with care.
3. Distribute into Petri dishes.

TECHNIQUE AND INTERPRETATION OF THE RESULTS

Refer to the technical documentation for VOGEL JOHNSON AGAR medium (REF. 610186 or 620186), or for the specific medium being prepared.

QUALITY CONTROL

1. Visual inspection: clear, colourless solution.
2. Microbiological control.

Prepare the plates using as base VOGEL JOHNSON AGAR medium (REF. 610186 or 620186) supplemented with POTASSIUM TELLURITE 1% *Supplement* (1 bottle in 500 ml of medium). The plates are seeded with the strains indicated in the microbiological control table.

Incubation conditions: 24-48 h at 36±1°C.

Microbiological control

Control strains		**Growth**	**Colonies**
Staphylococcus aureus	ATCC 25923	Good	Black
Escherichia coli	ATCC 25922	Inhibited	-------

PRECAUTIONS

The product POTASSIUM TELLURITE 1 % *Supplement* is not classified as dangerous under current legislation; it is nevertheless recommended that the Safety Data Sheet be consulted on its correct use.
POTASSIUM TELLURITE 1 % *Supplement* is a supplement to be used only for *in vitro* diagnostic use. It is intended for use in a professional environment and must be used in the laboratory by properly trained personnel, using approved asepsis and safety methods for handling pathogenic agents.

STORAGE

Store POTASSIUM TELLURITE 1 % *Supplement* at 2-8°C in its original packaging. In such conditions POTASSIUM TELLURITE 1% *Supplement* will remain valid until the expiry date indicated on the label. Do not use beyond that date. Eliminate without using if there are signs of deterioration.

REFERENCES

- United States Pharmacopoeia XXI (1985) Microbial Limit Tests. Rockville. Md.
- Vogel, R.A., and Johnson, M.J. (1961). Pub. Hlth. Lab. 18: 131.

PRESENTATION

product	REF	Σ
POTASSIUM TELLURITE 1% *Supplement*	80022	5 bottles x 10 ml
POTASSIUM TELLURITE 1% *Supplement*	80422	10 bottles x 10 ml

TABLE OF SYMBOLS

IVD *In Vitro* Diagnostic Medical Device	Do not reuse	Manufacturer	Contains sufficient for <n> tests	Temperature limitation
REF Catalogue number	Fragile, handle with care	Use by	Caution, consult accompanying documents	LOT Batch code

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Via Scozia, Zona Ind.le - 64026, Roseto degli Abruzzi (TE) - ITALY
Tel +39 0858930745 Fax +39 0858930330 Website: www.liofilchem.net E-mail: liofilchem@liofilchem.net

CE IVD

73831 Tinsdale Supplement (Diphtheria Virulence Supplement)

Tinsdale Supplement is used together with Tinsdale Agar Base for the primary isolation and identification of *Corynebacterium diptheriae*.

Composition
(1 vial of each is sufficient for 1000 ml medium)

Tinsdale Supplement part A (Prod No 74933):
horse serum 100ml

Tinsdale Supplement part B (Prod No 66784):
potassium tellurite (1% after diluting in 30 ml water) 1ml

Directions
Warm up the rfrigerated content of 1 vial part B and aseptically add 29 ml sterile distilled water. Mix thoroughly. Warm up part A & B to 50°C and add aseptically the contents of part A & B of Tinsdale Supplement to 1000 ml sterile molten (45-50°C) Tinsdale Agar Base (Prod No 89747). Mix throughly and pour into sterile dishes.

Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

89747 Tinsdale Agar Base

Tinsdale Agar is used for the primary isolation and identification of *Corynebacterium diptheriae*. The medium differentiates between *C. diphtheriae* and the diphtheroids found in the upper respiratory tract and sometimes on the skin. This differentiation was based on the ability of *C. diphtheriae* to produce greyish-black colonies, surrounded by a brown/black halo. Diphtheroids do not have this ability.

Composition:

Ingredients	Grams/Litre
Peptic digest of animal tissue	20.0
Sodium chloride 5.0g/l	5.0
L-Cystine	0.24
Sodium thiosulfate	0.43
Agar	15.0

Store prepared media below 8 °C, protected from direct light. Store dehydrated powder in a dry place in tightly-sealed containers at 2-25 °C.

Directions:
Suspend 40.7g of agar base in 1000ml of distilled water. Bring to the boil and dissolve completely. DO NOT AUTOCLAVE. Allow to cool to 50°C and add aseptically the contents of 1 unit (part A+B) of Tinsdale Supplement (Prod No 73831). Mix throughly and pour into sterile dishes.

Principle and Interpretation:
Tinsdale original formulated an agar containing cystine, serum, tellurite and formolised blood.[1] Later it was modified by Billings to a medium with improved differential qualities.[2,3]
Peptic digest of animal tissue serves as nitrogen, carbon, vitamin and amino acid source. The serum, present in the supplement, contains essential growth factors and sodium chloride maintains the osmotic balance of the medium. Cystine and sodium thiosulfate are substrates for the production of H_2S. Due to the production of H_2S the potassium tellurite (present in the supplement) is reduced to a metallic tellurite and forms a dark (black to brown) halo surrounding the colony. Additionally, potassium tellurite inhibits the gram negative bacteria and most of the upper respiratory tract normal flora. Do not incubate the plates in 5-10% CO_2 as it retards the development of characteristic halos.[4]
Diphtheroids (*C. pseudodiphtheriticum*), *Haemophilus*, *Klebsiella*, *Neisseria*, *Staphylococcus* and *Streptococcus* species build dark, brown colonies without halos. The colonies are tiny and show no discoloration of the medium.

Cultural characteristics after 40-48 hours at 35°C.

Organisms (ATCC)	Growth	Colony characteristics
C. diphtheriae type gravis (19409)	+++	small, shiny black, brown to black with halo
C. diphtheriae type mitis (11051)	+++	convex colonies, brown to black with halo
C. diphtheriae type intermedius (51279)	+++	brown to black with halo
C. ulcerans (51799)	+++	convex colonies, brown to black with halo
Proteus mirabilis (12453)	+++	brown to black colonies, characteristic odour and morphology.
Streptococcus pyrogenes (19615)	++	Black pin point, without halo
Klebsiella pneumoniae (13883)	-	-

References:

1. G.F. Tinsdale, J. Path. and Bacteriol., 59, 461 (1955)
2. E. Billings, An Investigation of Tinsdale's Tellurite Medium, its Usefullness and Mechanism of Halo Formation, Thesis Univ. Michigan (1956)
3. M.S. Moore, E.I. Parsons, J. Infect. Dis., 102, 88 (1958)
4. Lennette and others (Eds.), Manualof Clinical Microbiology, 4th ed. ASM (1985)
5. H.D. Isenberg, Clinical microbiology procedures handbook, American Society for Microbiology, Washington D.C. (1992)

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

ENGLISH

VP (NaOH) Reagent

Kit of reagents for the execution of Voges-Proskauer test

DESCRIPTION

VP (NaOH) Reagent is a kit containing the reagents for the execution of Voges-Proskauer test used for identification of microorganisms performing 2-3 butilneglicole fermentation.

CONTENT OF THE KIT

Each package contains 2 DROPPERS and the follow reagents:

- ALPHA NAPHTHOL 5 Vials x 10 ml
- NaOH 40% 5 Vials x 10 ml

METHOD PRINCIPLE

Voges-Proskauer reaction distinguishes organisms that ferment glucose to acids from those that ferment to acetoin (2-3 butilenglicole fermentation). After 48 hours incubation ALPHA NAPHTHOL and NaOH 40% reagents are added to sample. Positive reaction to Voges-Proskauer test is highlighted by the formation of a red coloration obtained from acetyl-methyl-carbinol oxidation in the presence of alkali and oxygen.

REAGENTS TABLE

Reagents	Composition
ALPHA NAPHTHOL	Alpha Naphthol 6 g, Ethyl Alcohol 100 ml
NaOH 40%	Sodium Hydroxide 40 g, Distilled Water 100 ml

TEST PROCEDURE AND RESULTS INTERPRETATION

The VP test is performed on cultures of 48 hours of incubation prepared dissolving a pure culture in a tube of MR-VP BROTH (ref. 610032 or 620032) properly prepared, or in a tube of MR-VP MEDIUM (ref. 20149). Add 3 drops of ALPHA NAPHTHOL and 2 drops of NaOH 40% solution to 3 mL of broth culture. The positive test is given by the development of a bright red colour 15 minutes after addition of reagents.

QUALITY CONTROL FOR THE USER

Visual inspection

NaOH 40% is a colourless clear solution;
ALPHA NAPHTHOL is a beige colour clear solution.

Microbiology control

Each batch of V.P. TEST Reagent is subjected to quality control using a bacterial culture of *Escherichia coli* ATCC 25922 as negative control and one of *Enterobacter cloacae* ATCC 13047 as positive control.

PRECAUTIONS

VP (NaOH) Reagent is classified as hazardous under current legislation; for its use it is recommend to consult the MSDS. The product is a disposable device to be used only for in vitro diagnostic use, is intended for professional use and should be used in the laboratory by properly trained operator, with approved methods of aseptic and safety against pathogens.

STORAGE

Store at 2-8°C away from light in its original package, until the expiry date shown on the label. Eliminate if signs of deterioration or contamination are evident.

ELIMINATION OF THE USED MATERIAL

After use, VP (NaOH) Reagent and the material that comes in contact with the sample must be decontaminated and disposed in accordance with the techniques used in the laboratory for decontamination and disposal of potentially infectious materials.

BIBLIOGRAPHY

1. Edwin H. Lenette*: Manual of Clinical Microbiology (1995).*
2. Blazevic, D.J., and Ederer, G.M.: Principles of biochemical tests in diagnostic microbiology. 63-67. New York, John Wiley & Sons,1975
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PRESENTATION

Product	REF	Σ
VP (NaOH) Reagent	80280	100 tests

TABLE OF SYMBOLS

LOT	Batch code	IVD	*In vitro* Diagnostic Medical Device		Manufacturer		Use by		Fragile, handle with care
REF	Catalogue number		Temperature limitation	Σ	Contains Sufficient for <n> tests		Consult instructions for use		Do not reuse

LIOFILCHEM® S.r.l.
Via Scozia, 64026 Roseto degli Abruzzi (TE) - ITALY
Tel +39 0858930745 Fax +39 0858930330 www.liofilchem.com liofilchem@liofilchem.com

CE IVD

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