ENGLISH

Optochine Test

Diagnostic discs for pneumococci identification.

DESCRIPTION
Optochine Test is constituted by paper discs, each one containing 5 µg of Optochin (Ethylhydrocupreine hydrochloride), used for differentiating *Streptococcus pneumoniae* from the other alpha-haemolytic streptococci.

CONTENT OF THE PACKAGES
Each package contains:
- 2 cartridges with 50 discs each, packaged in a heat-sealed container.
- Dryer.

PRINCIPLE OF THE METHOD
Optochin is an agent specifically active against *Streptococcus pneumoniae*, the other alpha-haemolytic streptococci are resistant. The disc is placed onto the surface of a culture medium that is suitable for the growth of streptococci, inoculated with a pure liquid culture of the microorganism under examination. After the incubation all the plates are examined for the presence or absence of an inhibition halo around the disc of Optochin.

COMPOSITION
Each disc contains 5 µg of Optochin.

PREPARATION OF THE SPECIMEN
1. Mixed cultures or clinical specimens must not be used to determine susceptibility to Optochin.
2. Inoculate a tube of Brain Heart Infusion Broth (ref. 20104) with pure colonies of the microorganism under examination.
3. Incubate at 36 ± 1°C overnight.

TEST PROCEDURE
1. Take the cartriges container from the refrigerator and leave it on the test bench until it reaches room temperature (about 30 minutes). This will prevent humidity being deposited on the discs when the package is opened, which could prejudice their long-term stability.
2. Using a sterile swab, evenly inoculate the surface of a plate of blood agar such as Tryptic Soy Blood Agar (ref. 11037), Columbia Blood Agar (ref. 11025) or other blood medium, with the suspension of the streptococcus under examination.
3. Using sterile tools, gently press one disc of Optochin on the inoculated surface.
4. Turn the plate upside down and incubate at 36 ± 1°C for 18-24 hours in atmosphere containing 5% of CO_2.
5. Check for presence or absence of an inhibition halo around the disc of Optochin.

INTERPRETATION OF THE RESULTS
The test organism is considered sensitive to Optochin and presumptively *Streptococcus pneumoniae* if the inhibition zone is ≥ 14 mm diameter. The presumptive identification must be confirmed by serological tests.

QUALITY CONTROL
Each batch of Optochine Test is tested for susceptibility to Optochin by using *Streptococcus pneumoniae* ATCC® 6305 for positive control, and *Streptococcus pyogenes* ATCC® 19615 for negative control, inoculated on Columbia Blood Agar with 5% of defibrinated sheep blood.

PRECAUTIONS
Optochine Test cannot be classified as being hazardous according to the current legislation. Optochine Test is a disposable device to be used only for diagnostic use *in vitro*. It must be used in the laboratory by properly trained personnel, using approved aseptic and safety methods for handling pathogenic agents.

STORAGE
Store Optochine Test at -20°C/+8°C in the original packaging. Keep away from sources of heat and avoid excessive changes in temperature. In such conditions, the product 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.

DISPOSAL OF USED MATERIAL
After use, Optochine Test and material that has come into contact with the sample must be decontaminated and disposed of in accordance with the techniques used in the laboratory for decontamination and disposal of potentially infected material.

BIBLIOGRAPHY

- https://www.gov.uk/government/uploads/system/uploads/attachment_data/file/394193/TP_25i3.pdf
- J. Lab. Clin. Med., 49: 641, 1957.
- J. Clin. Path., 8: 58, 1955.
- Serological Studies on Pneumococci, Munksgaard, Copenhagen, Oxford University Press, London, 1943.
- J. Exp. Med. 22: 269, 1915.

PRESENTATION

Product	Ref.	Test
Optochine Test	9501	100

TABLE OF SYMBOLS

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

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

McFARLAND BARIUM SULPHATE STANDARD

Standard di torbidità per la preparazione di sospensioni di microrganismi.
Turbidity standard for preparing suspensions of microorganisms.

DESCRIZIONE

Gli standard McFarland vengono utilizzati come standard di torbidità nella preparazione delle sospensioni di microrganismi ed in particolar modo nella preparazione degli inoculi batterici per l'esecuzione dell'antibiogramma.

PRINCIPIO

Gli standard di torbidità sono composti da sostanze chimiche che miscelate precipitano formando una soluzione di riproducibile torbidità.
Gli standard McFarland vengono preparati aggiungendo acido solforico ad una soluzione acquosa di cloruro di bario.
La miscela porta alla formazione di precipitato di solfato di bario.
Per ciascun standard McFarland in tabella 1 è riportata la densità corrispondente espressa in cellule/ml. La concentrazione batterica dipende dalla dimensione dei microrganismi. I valori riportati nella tabella 1 rappresentano valori medi di concentrazione validi per i batteri. Per i lieviti, che hanno dimensioni maggiori, bisogna dividere gli stessi numeri per 30.

PROCEDURA

Prima dell'uso, agitare vigorosamente lo standard di torbidità, utilizzando un vortex meccanico.
Comparare la torbidità di una sospensione batterica preparata alla torbidità dello standard, in presenza di una luce adeguata.
Alternativamente, utilizzare lo standard di torbidità per calibrare un turbidimetro elettrometrico.

INTERPRETAZIONE DEI RISULTATI

L'utilizzo degli standard McFarland consente la preparazione di inoculi standardizzati da utilizzare nelle procedure per l'esecuzione dell'antibiogramma.

DESCRIPTION

McFarland standards are used as turbidity standards in the preparation of suspensions of microorganisms and has particular application in the preparation of bacterial inocula for performing antimicrobial susceptibility testing.

PRINCIPLE

Turbidity standards are prepared by mixing chemicals that precipitate to form a solution of reproducible turbidity.
McFarland standards are prepared by adding sulphuric acid to an aqueous solution of barium chloride, which results in the formation of a suspended barium sulphate precipitate.
For each McFarland standard in table 1 is reported the correspondent density expressed in cells/ml. Bacterial concentration depends on microorganisms size. The mentioned values in table 1 represent average values of concentration valid for bacteria. For yeast, which are larger in size, these numbers should be divided by about 30.

PROCEDURE

Vigorously agitate the turbidity standard on a mechanical vortex mixer just before use.
Using adequate light, compare the turbidity of a bacterial suspension to the turbidity standard.
Alternatively, use the turbidity standard to calibrate a electrometric turbidimeter.

RESULTS INTERPRETATION

McFarland standards will enable the preparation of standardized inocula for use in the performance of standardized antimicrobial susceptibility testing procedures.

Tabella / Table 1.

McFarland Standard	Densità (cellule/ml) / Density (cells/ml)
0.5	1.5×10^8
1.0	3.0×10^8
2.0	6.0×10^8
3.0	9.0×10^8
4.0	12.0×10^8

BIBLIOGRAFIA / BIBLIOGRAPHY

1. Mc Farland, 1907. J.Am.Med.Assoc.49:1176.
2. Patricia M. Tille. 2014. Bailey & Scott's Diagnostic Microbiology, 13th edition by Mosby, Inc., an affiliate of Elsevier Inc.
3. CLSI M7-A9, 2012. Methods for dilution antimicrobial susceptibility test for bacteria that grow aerobically.
4. CLSI M11-A7, 2007. Methods for dilution antimicrobial susceptibility testing of anaerobic bacteria.

PRESENTAZIONE / PRESENTATION

Prodotto / Product	REF	Σ
McFARLAND 0.5 BARIUM SULPHATE STANDARD	80400	1
McFARLAND 1.0 BARIUM SULPHATE STANDARD	80401	1
McFARLAND 2.0 BARIUM SULPHATE STANDARD	80402	1
McFARLAND 3.0 BARIUM SULPHATE STANDARD	80403	1
McFARLAND 4.0 BARIUM SULPHATE STANDARD	80404	1
McFARLAND STANDARD SET (McFARLAND 0.5, 1.0, 2.0, 3.0, 4.0)	80405	5

TABELLA DEI SIMBOLI / TABLE OF SYMBOLS

LOT Codice del lotto / Batch Code	Σ Contenuto sufficiente per <n> saggi / Content sufficient for <n> tests	Fabbricante / Manufacturer	Non riutilizzare / Do not reuse
REF Numero di catalogo / Catalogue Number	Attenzione, vedere le istruzioni per l'uso / Attention, see instructions for use	Fragile, maneggiare con cura / Fragile, handle with care	

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

Rev.3 / 10.01.2019

S.S. AGAR (MODIFIED)

Selective medium for the isolation of *Salmonella* spp. and *Shigella* spp.

TYPICAL FORMULA	(g/l)
Peptone	5,5
Meat Extract	5,0
Lactose	10,0
Sodium Tiosulfate	8,5
Yeast Extract	5,0
Sodium Citrate	1,0
Bile Salts N.3	1,5
Ferric Ammonium Citrate	1,5
Brilliant Green	0,33 mg
Neutral Red	0,025
Agar	14,0
Final pH 7,0 ± 0,2	

DESCRIPTION

S.S. AGAR (MODIFIED) is a highly selective medium for the isolation of *Salmonella* spp. and some species of *Shigella* from clinical specimens and food.

PRINCIPLE

Gram-positive microorganisms and coliforms are inhibited by selective components: brilliant green, bile salts n.3, sodium tiosulfate and citrate. The differentiations of microorganisms is obtained through the introduction of lactose in the medium. Lactose fermented bacteria cause acidification, thus formation of red colonies for the presence of neutral red. Not-fermented microorganisms form instead colourless colonies. Sodium tiosulfate in combination with iron acts as indicator for sulphur production causing the blackening of the colony center.

PREPARATION

Suspend 52.0 g of the powder in 1 litre of distilled or deionized water. Mix well. Heat to boil shacking frequently until dissolved completely. DO NOT AUTOCLAVE. Cool to 45-50°C. In aseptic conditions dispense in Petri dishes and let solidify the medium with the lids of the plates partially removed.

TECHNIQUE

Inoculate the plate streaking the sample onto the agar surface to isolate pure colonies from samples containing a mixed flora. Incubate at 36+/-1°C for 18-24 hours.

INTERPRETATION OF RESULTS

Salmonella spp. and other lactose not-fermented microorganisms can produce opaque, translucent or transparent colonies, with or without black center. Shigella colonies are colourless. The few lactose fermented microorganisms, that are able to growth on the medium, show reddish mucoid colonies.

STORAGE

10-30°C away from light, until the expiry date on the label. Eliminate if signs of deterioration or contamination are evident.

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 designed for *in vitro* diagnostic use only and must be used by properly trained operators.

DISPOSAL OF WASTE

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

REFERENCES

1. Gray L.D. (1995). Escherichia, Salmonella, Shigella and Yersinia, p. 450-456. In Manual of clinical microbiology, 6th ed. American society of microbiology.
2. Leifson E. (1935). J. Pathol. Bacteriol. 40: 581.
3. Rose, H.M., and M.H. Kolodny (1942). J. Lab. Clin. Med. 27: 1081-1083.

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
S.S. AGAR (MODIFIED)

PRESENTATION
Dehydrated medium

STORAGE
10-30°C

PACKAGING

Ref.	Content	Packaging
610042	500 g	500 g of powder in plastic bottle
620042	100 g	100 g of powder in plastic bottle
6100425	5 kg	5 kg of powder in plastic container

pH OF THE MEDIUM
7,0 ± 0,2

USE
S.S. AGAR (MODIFIED) is a highly selective medium for the isolation of *Salmonella* spp. and some species of *Shigella* from clinical specimens and foods

TECHNIQUE
Refer to technical sheet of the product

APPEARANCE OF THE MEDIUM
Dehydrated medium
Appearance: free-flowing, homogeneous
Colour: light-pink
Prepared medium
Appearance: opalescent
Colour: purple

SHELFLIFE
4 years

QUALITY CONTROL
1. Control of general characteristics, label and print
2. Microbiological control
   Inoculum for productivity: 10-100 UFC/ml
   Inoculum for selectivity: 10^4-10^5 UFC/ml
   Inoculum for specificity: ≤10^4 UFC/ml
   Incubation Conditions: 18-24 h at 35 ± 2°C, in aerobiosis

Microorganism		Growth	Features
Shigella flexneri	ATCC® 12022	Good	Colourless colonies
Salmonella typhimurium	ATCC® 14028	Good	Colourless colonies with or without black center
Enterococcus faecalis	ATCC® 29212	Inhibited	—
Staphylococcus aureus	ATCC® 25923	Inhibited	—
Escherichia coli	ATCC® 25922	Partially Inhibited	Pink or red colonies

TABLE OF SYMBOLS

LOT	Batch code	IVD	*In vitro* Diagnostic Medical Device		Manufacturer		Use by
REF	Catalogue number		Temperature limitation	Σ	Contains sufficient for <n> tests		Caution, consult instructions for use

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

INDOLO TEST *STICK*

Test rapido su stick per la determinazione dell'enzima triptofanasi.
Rapid test on stick for determination of tryptophanase enzyme.

IMPIEGO

Organismi che producono l'enzima triptofanasi degradano l'aminoacido triptofano in acido piruvico, ammonio ed indolo. L'indolo viene evidenziato aggiungendo un indicatore aldeidico che sviluppa un'intesa colorazione verde. Il test può essere usato per differenziare le specie di *Proteus* e per l'identificazione presuntiva di *Escherichia coli.*

N.B.

E' necessario eseguire il test dell'indolo su colonie isolate da terreni di coltura contenenti triptofane, quali Plate count agar, Tryptic soy agar ed altri.

USE

Organisms that produce enzyme tryptophanase degrades the amino acid tryptophan into pyruvic acid, ammonia and indole. Indole is underlined by adding an aldehydic indicator that develops a strong green coloration. The test can be used to differentiate Proteus species and for presumptive identification of Escherichia coli.

N.B.

It is necessary to perform indolo test on colonies isolated from culture media containing tryptofane, like Plate count agar, Tryptic soy agar and others.

MODALITA' D'USO

- Prelevare uno stick dal contenitore.
- Toccare con un'ansa appuntita la colonia da sottoporre al test e strisciare sull'estremità dello stick.
- Aggiungere 2 gocce di Indole Test Reagent .
- Lo sviluppo di un colore verde immediato indica una reazione positiva.

INSTRUCTION FOR USE

- *Pick up one stick from the container.*
- *With a pointed loop, touch the colony to subject to test and slide on the stick's edge.*
- *Add 2 drops of Indole Test Reagent .*
- *Development of an immediate green coloration indicates positive reaction.*

CONTROLLO QUALITA'

Ogni lotto di INDOLE TEST STICK viene sottoposto al test per la produzione dell' Indolo utilizzando ceppi batterici di *Escherichia coli ATCC 25922* per il controllo positivo e *Proteus mirabilis ATCC 25933* per il controllo negativo.

QUALITY CONTROL

Each batch of INDOLE TEST STICK is subjected to test for indole production, using bacterial strains of Escherichia coli ATCC 25922 for positive control and Proteus mirabilis ATCC 25933 for negative control.

CONTENUTO

Indolo Test.....30 Stick
Indolo Test Reagent.....1 x 3ml

CONTENT

Indole Test.....30 Stick
Indole Test Reagent.....1 x 3ml

CONSERVAZIONE

5-12°C

STORE AT

5-12C

VALIDITA'

1 anno

SHELF LIFE

1 year

PRODOTTO / *PRODUCT*	CODICE / *CODE*	CONFEZIONE / *PACKAGING*
INDOLO TEST *STICK*	88032	30 TESTS

BIBLIOGRAPHY

1. *Blazevic, D.J., and Ederer, G.M.: Principles of Biochemical Tests in Diagnostic Microbiology. pp. 63-67. New York, John Wiley & Sons, 1975.*
2. *Vracko R. and Sherris J.C.: Indole-Spot Test in Bacteriology. Am. J. Clin. Pathol., 39: 429-432, 1963.*

COLUMBIA AGAR BASE

Medium for fastidious microorganisms isolation from clinical samples.

TYPICAL FORMULA (g/l)

Peptospecial	23.0
Starch	1.0
Sodium Chloride	5.0
Agar	14.0

Final pH = 7.3 ± 0.2 at 25 °C.

DIRECTIONS

Suspend 43.0 g of powder in 1 liter of distilled or deionized water. Heat to boiling until completely dissolved. Sterilize in autoclave at 121 °C for 15 minutes. Cool to 45-50 °C and aseptically add 5% defibrinated sterile sheep blood. Mix well. Dispense in petri dishes.

Columbia Agar Base can be also enriched in various way:

- with 2 vials of CNA (Staf / Strep) supplement (colistin sulphate 5 mg/vial, nalidixic acid 8 mg/vial, code 81048), each one reconstituted with 5 ml of sterile distilled water; final medium will contain colistin sulphate 10 mg/l and nalidixic acid 16 mg/l.
- with 2 vials of *Gardnerella vaginalis* supplement (gentamicin 3 mg/vial, amphotericin B 1mg/vial, nalidixic acid 15 mg/vial, code 81040), each one reconstituted with 5 ml of a 1:1 solution of ethyl alcohol and sterile distilled water; final medium will contain gentamicin 6 mg/l, amphotericin B 2 mg/l and nalidixic acid 30 mg/l.

DESCRIPTION

COLUMBIA AGAR BASE, enriched with sterile sheep blood (5%), is suitable for isolation and growth of fastidious microorganisms such as streptococci, staphylococci, pneumococci and listeriae from clinical samples.

TECHNIQUE

Inoculate the medium with the specimen streaking by a sterile loop and incubate at 36 ± 1 °C for 18-48 hours aerobically, anaerobically or under conditions of increased CO_2 (5-10%), in accordance with established laboratory procedures.

Examine plates for growth and hemolytic reactions. Four types of hemolysis on blood agar media can be described:

1. α-hemolysis is the reduction of hemoglobin to methemoglobin in the medium surrounding the colony, causing a greenish discolorization of the medium.
2. β-hemolysis is the lysis of red blood cells, producing a clear zone surrounding the colony.
3. γ-hemolysis indicates no destruction of red blood cells and no change in the color of the medium.
4. á-hemolysis indicates a partial lysis.

QUALITY CONTROL

Dehydrated medium

Appearance: free-flowing, homogeneous.

Color: beige.

Prepared medium

Appearance: opaque.

Color: cherry red.

Incubation conditions: 36 ± 1°C for 18-48 hours at 5-10% CO_2.

Microorganism	ATCC	Growth	Characteristics
Streptococcus pyogenes	19615	good	β-hemolysis
Streptococcus pneumoniae	6303	good	α -hemolysis
Staphylococcus aureus	25923	good	β-hemolysis
Gardnerella vaginalis	14018	good	β-hemolysis

LIOFILCHEM s.r.l.
Via Scozia, Zona Ind.le - 64026, Roseto D.A. (TE) - ITALY
Phone +390858930745 Fax +390858930330
Website: www.liofilchem.net E-mail: liofilchem@liofilchem.net

CE IVD

PERFORMANCE AND LIMITATIONS

When this medium is enriched with 10% sterile sheep blood, heated at 80 °C for 10 minutes until a chocolate color is obtained, and an antibiotic mixture is added (vancomycin, colimycin, trimethoprim, amphoterycin B) it is suitable for the selective isolation of the pathogens neisseria. If used without the addition of blood, the medium is suitable for growing of *Brucella abortus, Yersinia pestis, Clostridium perfrigens and enterobacteria.* Hemolytic reactions of some strains of Group D streptococci have been shown to be affected by differences in animal blood. Such strains are beta –hemolytic on horse and rabbit blood agar and alpha-hemolytic on sheep blood agar.

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 signs of deterioration or contamination are evident.

Store prepared plates at 2-8 °C.

REFERENCES

1. Ellner, P.D., C.J. Stoessel., E. Drakeford, and F. Vasi (1966). A new culture medium for medical bacteriology. Am. J.Clin. Path. 45, 502-504.
2. Isenberg, H.D. (ed.) (1992). Clinical microbiology procedures handbook, vol. 1 American Society for Microbiolgy, Washington, DC.

PRESENTATION

Product	REF	Σ
COLUMBIA AGAR BASE (11.6 l)	610013	500 g
COLUMBIA AGAR BASE (2.3 l)	620013	100 g
COLUMBIA AGAR BASE (116.2 l)	6100135	5 Kg
SHEEP BLOOD DEFIBRINATED	83296	50 ml
CNA (Staf / Strep) supplement	81048	10 vials
Gardnerella vaginalis supplement	81040	10 vials

TABLE OF SYMBOLS

LOT Batch code	Caution, consult accompanying documents	Manufacturer	Contains sufficient for <n> tests	IVD *In Vitro* Diagnostic Medical Device
REF Catalogue number	Fragile, handle with care	Use by	Temperature limitation	Keep away from heat source

LIOFILCHEM s.r.l.
Via Scozia, Zona ind.le - 64026, Roseto D.A. (TE) - ITALY
Phone +390858930745 Fax +390858930330
Website: www.liofilchem.net E-mail: liofilchem@liofilchem.net

CE IVD

Slanetz Bartley Agar Base

Selective medium for detection and enumeration of enterococci in water and other materials, according to ISO 7899-2.

TYPICAL FORMULA	(g/l)
Tryptose	20.0
Yeast Extract	5.0
Glucose	2.0
Dipotassium Hydrogen Phosphate	4.0
Sodium Azide	0.4
Agar	13.0

Final pH 7.2 ± 0.2 at 25°C

DESCRIPTION

Slanetz Bartley Agar Base is a selective medium used with supplement for isolating and enumerating enterococci from environmental samples of sanitary importance and clinical specimens.

This medium complies with ISO 7899-2 for the detection of intestinal enterococci in water by the membrane filtration technique.

PRINCIPLE

Tryptose provides amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Yeast extract is a source of vitamins, particularly of B-group. Glucose is the fermentable carbohydrate. Sodium phosphate acts as buffer. Sodium azide is the selective agent suppressing the growth of Gram-negative bacteria. Agar is the solidifying agent.

Supplementation with TTC 1% Supplement serves to add triphenyl tetrazolium chloride (TTC) as indicator of bacterial growth.

PREPARATION

Suspend 44.4 g of powder in 1 liter of deionized or distilled water. Bring to boil and shake until completely dissolved. Sterilize at 121°C for 15 minutes. Cool up to 45-50°C. Aseptically, add 10 ml of TTC 1% Supplement (ref. 80300). Mix well. Pour in Petri dishes.

TECHNIQUE

ISO 7899-2 recommends to filter the water sample trough a filter membrane (0.45 µm pore diameter), transfer the membrane onto a Slanetz Bartley Agar plate and incubate aerobically at 36 ± 2°C for 40-48 hours.

Alternatively, sample can be inoculated by spread plating, pour plating or by direct streaking on the medium surface.

INTERPRETATION OF RESULTS

Count all raised colonies which show a red, maroon or pink color as enterococci.

Confirm by subculturing to Bile Aesculin Azide Agar (ref. 163572).

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 contains hazardous substances and is classified as dangerous. It is recommended to consult the safety data sheet for its correct use. The product is designed for *in vitro* diagnostic use only and must be used by properly trained operators.

DISPOSAL OF WASTE

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

REFERENCES

- ISO 7899-2:2000. Water quality – Detection and enumeration of intestinal enterococci – Part 2: Membrane filtration method.
- Slanetz L.W. and C.H. Bartley (1957) Numbers of enterococci in water, sewage and faeces determined by the membrane filtration technique with an improved medium. J. Bact. 74:591-595.

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
Slanetz Bartley Agar Base

PRESENTATION
Dehydrated medium

STORAGE
10-30°C

PACKAGING

Ref.	Content	Packaging
610134	500 g	500 g of powder in plastic bottle
620134	100 g	100 g of powder in plastic bottle

pH OF THE MEDIUM
7.2 ± 0.2

USE
Slanetz Bartley Agar Base is a selective medium used with supplement for isolating and enumerating enterococci from water and other samples according to ISO 7899-2

TECHNIQUE
Refer to technical sheet of the product

APPEARANCE OF THE MEDIUM
Powder medium
Appearance: free-flowing, homogeneous
Colour: light beige
Ready-to-use medium
Appearance: slightly opalescent
Colour: light amber

SHELFLIFE
4 years

QUALITY CONTROL
1. Control of general characteristics, label and print
2. Microbiological control
   Inoculum for productivity: 50-100 CFU
   Inoculum for selectivity: 10^4-10^6 CFU
   Incubation Conditions: 44-48 h at 36 ± 2°C, in aerobiosis

Microorganism		Growth	Colony color
Enterococcus faecalis	WDCM 00009	Good	Red-maroon-pink
Enterococcus faecium	WDCM 00177	Good	Red-maroon-pink
Escherichia coli	WDCM 00013	Inhibited	–
Staphylococcus aureus	WDCM 00034	Inhibited	–

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 instructions 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 Website: www.liofilchem.net E-mail: liofilchem@liofilchem.net

CE IVD

Instructions For Use
ENGLISH

Hektoen Enteric Agar

Selective and differential medium for detection of pathogenic intestinal bacteria from food and clinical specimens, according to ISO 21567.

DESCRIPTION

Hektoen Enteric Agar is a moderately selective medium used for the isolation and cultivation of Gram-negative enteric microorganisms, especially *Shigella* spp, from faeces, foodstuffs and other materials of sanitary importance.

This medium meets the requirements of the APHA and ISO 21567 for the isolation and differentiation of *Salmonella* and *Shigella* spp.

TYPICAL FORMULA

	(g/l)
Enzymatic Digest of Meat	12.0
Yeast Extract	3.0
Lactose	12.0
Saccharose	12.0
Salicin	2.0
Bile Salts No. 3	9.0
Sodium Chloride	5.0
Sodium Thiosulfate	5.0
Ammonium Ferric Citrate	1.5
Acid Fuchsin	0.1
Bromothymol Blue	0.065
Agar	15.0

Final pH 7.5 ± 0.2 at 25°C

METHOD PRINCIPLE

Enzymatic digest of meat provides amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Yeast extract is a source of vitamins, particularly of B-group. Lactose, saccharose and salicin are fermentable carbohydrates. Bile salts and acid fuchsin inhibit Gram-positive organisms. Sodium chloride maintains the osmotic balance of the medium. Ammonium ferric citrate and sodium thiosulfate enable the detection of hydrogen sulfide production. Bromothymol blue together with acid fuchsin act are the pH indicator system. Agar is the solidifying agent.

PREPARATION

Dehydrated medium	Suspend 76 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.
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

Inoculate the plates by directly streaking the specimen on the agar surface or spread the sample from an enrichment culture. Incubate aerobically at 35 ± 2°C for 18-24 h

INTERPRETING RESULTS

Shigella and *Providencia* spp, form green, moist colonies.

Salmonella and *Proteus* spp, grow as blue-green colonies, with or without black center due to H_2S production.

Coliforms, which are mostly rapid lactose-saccharose-salicin fermenters, develop red-salmon colonies surrounded by a zone of bile precipitate.

Enterococci, Staphylococci and other Gram-positive bacteria are partially or completely inhibited.

Notice that further testing should be conducted to confirm the presumptive identification of organisms isolated on this medium.

APPEARANCE OF THE MEDIUM

Dehydrated medium: free-flowing, homogeneous, light beige.
Prepared medium: 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 bottles 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.

SHELF LIFE

Dehydrated medium: 4 years.
Medium in bottles: 2 years.
Ready-to-use plates: 6 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
Salmonella Typhimurium	ATCC® 14028	Good	Blue-green colonies with black centre
Shigella flexneri	ATCC® 12022	Good	Green colonies
Proteus mirabilis	ATCC® 12453	Good	Blue-green colonies with black centre
Klebsiella pneumoniae	ATCC® 13883	Good	Red-salmon colonies with zone of bile precipitate
Escherichia coli	ATCC® 8739	Partially to completely inhibited	Red-salmon colonies with or without zone of bile precipitate
Enterococcus faecalis	ATCC® 29212	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 only and must be used by properly trained operators.

DISPOSAL OF WASTE

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

BIBLIOGRAPHY

- ISO 21567:2004. Microbiology of food and animal feeding stuffs -- Horizontal method for the detection of *Shigella* spp.
- Perez J.M., P. Cavalli, C. Roure, R. Renac, Y. Gille, and A. M. Freydiere (2003) Comparison of Four Chromogenic Media and Hektoen Agar for Detection and Presumptive Identification of Salmonella Strains in Human Stools. J Clin Microbiol; 41(3):1130–1134.
- American Public Health Association (1992) Compendium of Methods for the Microbiological Examination of Foods 3rd Edition. APHA Inc. Washington DC.
- Bisciello N.B. jr. and Schrade J.(1974) Evaluation of Hektoen Enteric Agar for the detection of Salmonella in foods and feeds. - Journ of AOAC; 57: 992-996.

PRESENTATION

		Contents	Ref.
Hektoen Enteric Agar	90 mm ready-to-use plates	20 plates	10043
Hektoen Enteric Agar	90 mm ready-to-use plates	100 plates	10043*
Hektoen Enteric Agar	Bottles	6 x 100 ml bottles	402230
Hektoen Enteric Agar	Bottles	6 x 200 ml bottles	412230
Hektoen Enteric Agar	Dehydrated medium	500 g of powder	610021
Hektoen Enteric Agar	Dehydrated medium	100 g of powder	620021
Hektoen Enteric Agar	Dehydrated medium	5 kg of powder	6100215

TABLE OF SYMBOLS

LOT	Batch code	IVD	*In vitro* Medical Diagnostic Device		Manufacturer		Use by		Fragile, handle with care
REF	Catalogue number		Temperature limitation	Σ	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

Instructions For Use
ENGLISH

Oxidase Test Disc

Rapid test for detection of cytochrome oxidase enzymatic activity.

DESCRIPTION

Oxidase Test Disc is a diagnostic test used for differentiation and microbial identification, particularly of Gram-negative bacteria, on the basis of the presence of enzyme cytochrome oxidase.

The product matches with recommendations of EN ISO 16266 and ISO 9308-1 for detection of *Pseudomonas aeruginosa* and for confirmation of *Escherichia coli* and coliform bacteria, respectively.

CONTENTS OF THE PACKAGES

Each package contains 1 cartridge of 30 discs.

METHOD PRINCIPLE

Oxidase-positive bacteria produces the enzyme cytochrome oxidase (indophenol oxidase) that catalyzes the transport of electrons from donor compounds (NADH) to electron acceptors (usually oxygen).

Tetramethyl-p-phenylenediamine dihydrochloride contained in Oxidase Test Disc acts as an artificial electron donor and is oxidized by oxidase-positive bacteria forming the coloured compound indophenol blue.

COMPOSITION

Each disc of Oxidase Test Disc is impregnated with a solution of N,N,N´,N´-tetramethyl-p-phenylenediamine dihydrochloride.

TEST PROCEDURE

1. Allow container to come to room temperature before opening, for minimizing condensation on the disc.
2. Pick up one or more than one well isolated colony and smear on the disc. Alternatively, deposit one disc into a suspension of test organism.
3. Observe for the development of a color within 60 seconds (NB. The usage of very dilute microbial suspensions may result in longer reactions time).

INTERPRETING RESULTS

The development of a blue-purple color indicates a positive reaction. No color change corresponds to a negative test, i.e. the organism under investigation does not produce the enzyme cytochrome oxidase.

LIMITATIONS

The most suitable cultures for the oxidase test are those from culture media without dyes, indicators or inhibitors. Bacterial colonies taken from media with pH values below 5.5 (e.g. after the metabolism of carbohydrates with subsequent acidification of the culture medium) can give a false negative oxidase reaction. Colonies taken from media containing nitrate may give unreliable results. Do not use steel, nichrome or iron containing loops to pick the colony. A platinum or plastic loop, or wooden applicator stick is recommended.

STORAGE

Store 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

1 year.

QUALITY CONTROL

Control strains are indicated in the QC table.

QC Table.

Microorganism		Oxidase reaction
Escherichia coli	WDCM 00013	Negative, no color change
Pseudomonas aeruginosa	WDCM 00025	Positive, deep blue-purple coloration

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

- ISO 9308-1:2014. Water quality – Enumeration of *Escherichia coli* and coliform bacteria – Part 1: Membrane filtration method for waters with low bacterial background flora.
- EN ISO 16266:2008. Water quality – Detection and Enumeration of *Pseudomonas aeruginosa* – Method by membrane filtration (ISO 16266:2006).
- Steel K. J. (1962) J. Appl. Bact. 25:445-447.

PRESENTATION	Contents	Ref.
Oxidase Test Disc	30 discs	88004

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		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

ENGLISH

GRAM COLOR KIT

DESCRIPTION
GRAM COLOR KIT is a kit for staining micro-organisms that allows them to be differentiated into two categories: Gram-positives (Gram+), which are coloured blue, and Gram-negatives (Gram-), which are coloured red. Combined with direct observation of the cell morphology, this staining constitutes the first level in the taxonomic classification of prokaryotes.

CONTENT OF THE PACKAGES
The reagents are contained in plastic bottles, sealed by thermo-induction and provided with a dropper lid. Each pack contains:
- 1 bottle containing 250 ml of Crystal Violet Solution
- 1 bottle containing 250 ml of Lugol-PVP Solution
- 1 bottle containing 250 ml of Decolourant Solution
- 1 bottle containing 250 ml of Safranine Solution

PRINCIPLE OF THE METHOD
Gram staining is based on the property of Crystal Violet of combining with iodine to form compounds that cannot be decoloured with alcohol or with an alcohol-acetone mixture. Some bacteria have a special affinity for this reaction and, once stained with crystal violet, do not lose the colour if treated with alcohol or alcohol-acetone mixture, thus retaining the blue colouring (Gram-positive bacteria). Others lose the blue colour and are stained by Safranine, taking a red colour (Gram-negative bacteria).

COLLECTION OF SAMPLES
Samples to be subjected to Gram staining are usually clinical material and microbial cultures. The colonies to be subjected to Gram staining must be taken from young cultures (18-24 hours) preferably on an agar medium.

TEST PROCEDURE
Preparation and fixing

On clean slides, make a smear of the culture or pathological material. Leave to dry in the air and fix by heat, passing rapidly over the flame. Do not overheat the sample when fixing. Other fixing methods may be used.

Staining

1. Cover the slide with the Crystal Violet Solution. Wait 1 minute, then rinse gently with water.
2. Cover the slide with the Lugol-PVP Solution. Wait 1 minute, then rinse delicately with water.
3. Decolour with the Decolourant Solution for as long as the preparation releases colour (about 30-60 seconds), then rinse delicately with water.
4. Cover the slide with the Safranine Solution. Wait 30-60 seconds, then rinse delicately with water.
5. Dry.
6. Examine the preparation under the microscope with the objective for immersion.

INTERPRETATION OF THE RESULTS
The Gram-negative micro-organisms appear as red in colour.
The Gram-positive micro-organisms appear as blue in colour.
The Gram staining makes it possible to distinguish between:
- Gram-negative bacilli from Gram-positive ones;
- Gram-negative cocci from Gram-positive ones;
- Gram-negative coccobacilli from Gram-positive ones;
- Gram-negative diplococci from Gram-positive ones.

QUALITY CONTROL
Each lot of GRAM COLOR KIT is subjected to quality control using a culture of *Escherichia coli* ATCC 25922 for the test for Gram-negative bacteria (red colour) and a culture of *Staphylococcus aureus* ATCC 25923 for the test for Gram-positive bacteria (blue colour).

LIMITS
- Gram staining provides a preliminary identification but does not replace normal cultural studies of the sample.
- Antibiotic therapy may make Gram-positive bacteria more
- sensitive to decolouration, so that they appear pinkish-red instead of blue.
- Cells taken from young, 18-24 hour cultures have a greater affinity for the stains than cells taken from older cultures.
- Gram staining is altered by the physical destruction of the cell wall or protoplasm. In fact the cell wall of Gram-positive bacteria constitutes a barrier which impedes release of the Crystal Violet-iodine complex from the cytoplasm, and the cell wall of Gram-negative bacteria contains lipids soluble in organic solvents that permit decolouration of the cytoplasm. Hence, micro-organisms physically destroyed by an excess of heat do not react as expected to the Gram stain test.

PRECAUTIONS
The GRAM COLOR KIT package contains substances classified as hazardous by current legislation. It recommended that the Safety Data Sheets be consulted on their use. GRAM COLOR KIT is a kit for bacteria staining, only for diagnostic use *in vitro*. It is intended for use in a professional environment and must be used in a laboratory by adequately trained personnel using approved asepsis and safety methods for dealing with pathogenic agents.

CONSERVATION
Store GRAM COLOR KIT at 10-25°C in the original packaging. Keep away from sources of heat and avoid excessive changes of temperature. In such conditions the product GRAM COLOR KIT will be valid until the expiry date shown on the label. Do not use beyond that date. Eliminate without using if there are signs of deterioration (changes in the colour of the solutions or presence of substantial precipitates).

DISPOSAL OF USED MATERIAL
After use, the slides stained with the GRAM COLOR KIT and any material that has come into contact with the sample must be decontaminated and disposed of in accordance with the techniques used in the laboratory for decontamination and disposal of potentially infected material.

BIBILIOGRAPHY

- Kruczak-Filipov, P., and R.G. Shively.1992. Gram stain procedure, p.1.5.1-1.5.18. In H.D. Isenberg (ed.) Clinical Microbiology Procedures Handbook, vol. 1. American Society for Microbiology, Washington, D.C.
- Murray, P.R. (ed.) 1999. Manual of Clinical Microbiology, 7th ed. American Society of Microbiology, Washington, D.C.

PRESENTATION

Product	Ref	Content
GRAM COLOR KIT	80293	4 x 250 ml

TABLE OF SYMBOLS

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

Liofilchem®
Via Scozia-Zona industriale - 64026 Roseto degli Abruzzi Tel. +39.085.8930745 - Fax +39.085.8930330
Web site: http://www.liofilchem.net E-mail: liofilchem@liofilchem.net

F00411
Rev.3 / 09.05.2017

Translation from German

IFTA AG
Natürlich. Für Zukunft.

CERTIFICATE

The certification body confirms to

sifin diagnostics gmbh
Berliner Allee 317-321
13088 Berlin
Germany

for the development, manufacturing and sale of products for human and veterinary medical in-vitro-diagnostics as well as for the microbiological examination of water and food and other diagnostic applications the conformity of the introduced quality management system with the standard

DIN EN ISO 9001:2015

Start of validity:	07.07.2017
End of validity:	06.07.2020
Report and certificate number:	IC00016 038 17
The certificate consists of	1 page

This certificate includes an annual examination of the QMS by IFTA AG, according to the specified standard.

Berlin, 06.07.2017

Prof. Dr. Jörn Karge
CEO

DAkkS
Deutsche
Akkreditierungsstelle
D-ZM-16072-01-00

Certificate

mdc medical device certification GmbH
certifies that

sifin

sifin diagnostics gmbh
Berliner Allee 317-321
13088 Berlin
Germany

for the scope

development, manufacturing and distribution of in vitro diagnostic medical devices for the product groups: blood grouping, bacteriological test reagents and culture media as well as manufacturing of raw materials for manufacturing of in vitro diagnostic medical devices

has introduced and applies a

Quality Management System

The mdc audit has proven that this quality management system meets all requirements of the following standard

EN ISO 13485

Medical devices – Quality management systems – Requirements for regulatory purposes

EN ISO 13485:2016 + AC:2016 - ISO 13485:2016

Valid from	2018-10-23
Valid until	2021-10-22
Registration no.	D1058700042
Report no.	P18-00745-121758
Stuttgart	2018-07-16

Head of Certification Body

medical device certification mdc

DAkkS Deutsche Akkreditierungsstelle D-ZM-16002-05-00

mdc medical device certification GmbH
Kriegerstraße 6
D-70191 Stuttgart, Germany
Phone: +49-(0)711-253597-0
Fax: +49-(0)711-253597-10
Internet: http://www.mdc-ce.de