Maltose

Component in microbiological culture media

DESCRIPTION

Maltose is a fermentable carbohydrate and is used when preparing microbiological culture media in a laboratory setting. Maltose is a fermentable carbohydrate which acts as an energy source in culture media.

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 media at 2-8 °C.

DISPOSAL OF WASTE

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

PACKAGE

Code	Content	Packaging
611602	500 g	500 g of product in plastic bottle

SHELF LIFE

4 years

QUALITY CONTROL

Dehydrated powder
Appearance: free-flowing, homogeneous.
Colour: White crystalline.

TABLE OF SYMBOLS

LOT	Batch code		Consult instructions for use		Manufacturer		Contains sufficient for <n> tests
REF	Catalogue number		Temperature limitation		Use by		Keep away from heat sources

LIOFILCHEM® S.r.l.
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Mannitol

Component in microbiological culture media

PHYSIC-CHEMICAL CHARACTERISTIC

Solubility in water	Complete
Loss on drying	< 5.0%
Melting point	165-170 °C
Heavy metals	< 5 ppm
Ash	< 0.1%

DESCRIPTION

Mannitol is a fermentable carbohydrate and is used when preparing microbiological culture media in a laboratory setting. Mannitol is a fermentable carbohydrate which acts as an energy source in culture media.

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 media at 2-8 °C.

DISPOSAL OF WASTE

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

PACKAGE

Code	Content	Packaging
611017	500 g	500 g of product in plastic bottle

SHELF LIFE

4 years

QUALITY CONTROL

Dehydrated powder
Appearance: free-flowing, homogeneous.
Colour: White crystalline.

TABLE OF SYMBOLS

LOT	Batch code		Consult instructions for use		Manufacturer		Contains sufficient for <n> tests
REF	Catalogue number		Temperature limitation		Use by		Keep away from heat sources

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Rice Starch

Raw Material for the Cosmetic, Nutraceutical and Pharmaceutical Industry.

PHYSICO-CHEMICAL CHARACTERISTICS

Solubility	practically insoluble in cold water and alcohol
Specific Weight (g/l)	about 400
Granulometry	<= 75 microns
Proteins (N x 6.25)	<= 1.0 %
Humidity	<= 14.0 %
Calorific Value	about 400 Kjoule/100 g
Ashes	<= 0.60 %
Lipids	<= 0.10 %
Viscosity at 95 ^C (Brabender, 8 % M.S.)	450 - 600 BU
Whiteness	>= 97 %
pH (10 % solution)	5.5 – 7.0
Sulphur Dioxide	absent
Cellulose	absent
Sodium, Potassium & Iron	conforms
Residual Solvents	absent (not used)
Heavy Metals	<= 10 ppm
Lead	<= 3 ppm
Arsenic	<= 1 ppm
Cadmium	<= 1 ppm
Mercury	<= 0.1 ppm
Total Bacterial Count	<= 1000 cfu/g
Molds & Yeasts	<= 500 cfu/g
Pathogens	absent

DESCRIPTION

Very fine white powder, odourless and tasteless; it is a native starch from pure rice obtained by wet grinding. The rice starch, dispersed in cold water and subjected to subsequent boiling, swells with formation of a suspension which, on cooling, gels by retrogradation. It does not contain allergens, Gluten or GMO. It’s main use is in all those thickening applications where a binding agent is required to assure more strength to the formulation.

STORAGE CONDITIONS

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.

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 the national and local regulations in force.

SHELFLIFE

4 years

PACKAGING

Ref. 610500	Dehydrated raw material	500 g pf powder in plastic bottle

TABLE OF SYMBOLS

LOT Batch code	Keep away from Sunlight	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.
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Instructions For Use

ENGLISH

Sabouraud Dextrose Agar

Medium for cultivation and enumeration of yeasts and moulds from clinical and nonclinical specimens

INTENDED PURPOSE

Medium for the cultivation and enumeration of yeasts and moulds from clinical and non-clinical specimens. This medium is intended as an aid in the diagnosis, requiring further tests to complete the diagnostic results.

DESCRIPTION

Sabouraud Dextrose Agar (SDA) is a non-selective isolation medium used for the growth and maintenance of pathogenic and non-pathogenic fungi from clinical and nonclinical specimens. It is also used for recovery and total counting of yeasts and moulds in environmental monitoring.
This medium complies with EN ISO 11133 for microbiological examination of food, animal feed and water, where it is described as the main reference medium to carry out quantitative testing on culture media intended for fungi.
Its formula conforms to the recommendations of the harmonized method in the United States Pharmacopoeia (USP), European Pharmacopoeia (EP) and Japanese Pharmacopoeia (JP) for the microbiological examination of non-sterile products. The medium is also available as gamma-irradiated triple bagged plates, particularly suitable for use in restricted areas like isolators and clean rooms.

TYPICAL FORMULA*	(g/litre)
Pancreatic Digest of Casein	5.0
Peptic Digest of Animal Tissue	5.0
Dextrose	40.0
Agar	15.0
Final pH 5.6 ± 0.2 at 25°C	

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

METHOD PRINCIPLE

Pancreatic digest of casein and peptic digest of animal tissue provide amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Dextrose is an energy source. Agar is the solidifying agent. The high concentration of dextrose and the acidic pH of the medium permit selectivity of fungi.
The medium can be supplemented with chloramphenicol to increase bacterial inhibition and recovery of dermatophytes.

PREPARATION

Dehydrated medium

Suspend 65 g of the powder in 1 liter of distilled or deionized water. Mix well. Heat to boil for 1 minute 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 (losing 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.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as: Autoclave, water bath, sterile Petri plates, test tubes, inoculating loops, swabs, incubator, quality control organisms.

SPECIMENS

Clinical specimens should be sampled at the acute stage, before antimicrobial therapy (where possible) and examined as soon as possible after collection.
SDA is not suitable for direct inoculation of blood samples.
Good laboratory practices for collection, transport and storage of the clinical specimens should be applied.
Refer to specific guidelines for more information about specimen collection and preparation.

TEST PROCEDURE

Ensure there is no visible moisture on the plates before use.

For use in medical microbiology

Streak the specimen as soon as possible after it is received in the laboratory to obtain isolated colonies. Prepared tubed slants primarily are intended for use with pure cultures for maintenance or other purposes. Incubation conditions may vary according to the type of specimen and the microorganisms being tested for.

For use in food, animal feed and water testing.

Refer to EN ISO 11133 for specific instructions.

For use in industrial microbiology

Control of non-sterile products

Refer to the procedure described in the harmonized chapters of the Pharmacopoeia.

Passive Air Monitoring

Take the lid off the settle plate and leave the medium exposed to the air for a period of time no longer than 4 hours (settling plates filled with 30 ml of medium may compensate for water loss during extended incubation periods). Plates can be placed according to the 1/1/1 scheme (for 1 h, about 1 above the floor, at least 1 m from the walls or any obstacle).

Surfaces and Personnel Hygiene Monitoring

Take a swab sample for irregular surfaces or use the sampling template 10x10 (ref. 96762) to sample a well-defined area of the test surface. Inoculate a 90 mm plate by streaking the swab over the agar surface. Furthermore, the medium is suitable for personnel hygiene monitoring to detect microbial contamination of gloves or hands e.g. in a 5-finger-print.
Incubate the plates at 20-25°C for 5-7 days or at 30-35°C for 24-48 hours.

INTERPRETING RESULTS

Transfer of growth from slants to plated media may be required in order to obtain pure cultures of fungi. Examine for fungal colonies exhibiting typical microscopic and colonial morphology. Biochemical tests may be required for final identification.
The total combined yeasts/moulds count (TYMC) is considered to be equal to the number of CFU found per each plate.
When an acceptable criterion for microbiological quality is prescribed it is interpreted as follows:

- 10^1 CFU: maximum acceptable count = 20;
- 10^2 CFU: maximum acceptable count = 200;
- 10^3 CFU: maximum acceptable count = 2000, and so forth.

In procedures intended for environmental and personnel hygiene monitoring, observe daily for the formation of colonies.

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.
Medium in tubes: 1 year
Ready-to-use plates: 6 months.

QUALITY CONTROL

Appearance of Dehydrated Medium: free-flowing, homogeneous, light-beige.

Appearance of Prepared Medium: slightly opalescent, light amber.

Expected Cultural Response:

Control strain		Inoculum	Incubation	Criteria
Saccharomyces cerevisiae	WDCM 00058; (ATCC® 9763; NCTC 10716)	50-100 CFU	3-5 days / 22.5 ± 2.5°C	Good Growth ($P_R \geq 0.7$)
Aspergillus brasiliensis	WDCM 00053; (ATCC® 16404)		72 ± 2 h / 22.5 ± 2.5°C	
Candida albicans	WDCM 00054; (ATCC® 10231)		46 ± 2 h / 22.5 ± 2.5°C	
Candida albicans	WDCM 00054; ATCC® 10231		24 - 48 h / 32.5 ± 2.5°C	

A productivity ratio (P_R) of 0.7 is equivalent to a recovery rate of 70%.

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

PERFORMANCE CHARACTERISTICS

Performance testing of Sabouraud Dextrose Agar was carried out using the QC strains listed above. The results obtained met the established criteria.

LIMITATIONS

Invalid results can be caused by poor specimen quality, improper sample collection, improper transportation, improper laboratory processing, or a limitation of the testing technology. The operator should understand the principles of the procedures, including its performance limitations, in advance of operation to avoid potential mistakes.

The medium may support the growth of some bacteria. Incubation at 30-37°C, is suitable for yeasts, but dermatophytes may be inhibited above 30°C.

Some fungi (e.g., *Blastomyces dermatitidis*) may not be recovered on this medium due to the high carbohydrate content. For identification, organisms must be in pure culture.

SDA is intended as an aid in the diagnosis of infectious diseases, requiring further tests to complete the diagnostic results.

WARNING AND PRECAUTIONS

1) **For *in vitro* diagnostic use (IVD).**
2) **For laboratory professional use only.**
3) Operators must be trained and have certain experience. Please read the instructions carefully before using the product. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this document.
4) Consult the Safety Data Sheet (SDS) for information regarding hazards and safe handling practices.
5) Do not use if the product or packaging appears to be damaged.
6) Follow standard precautions. All patient specimens should be considered potentially infectious and handled accordingly.
7) Handle all specimens as if infectious using safe laboratory procedures. Dispose of hazardous or biologically contaminated materials according to the practices of your institution.
8) Avoid cross-contamination of samples by using disposable tips and changing them after each sample.
9) Do not mix reagents of different batches. Please use the product within the validity period.
10) Do not eat, drink, smoke, apply cosmetics or handle contact lenses in areas where reagents and human specimens are handled.
11) Results should be interpreted by a trained professional in conjunction with the patient's history and clinical signs and symptoms, and epidemiological risk factors.
12) Ensure laboratory equipment is calibrated and maintained in accordance with the laboratory's procedure.
13) When test results are transmitted from the laboratory to an informatics centre, attention has to be done to avoid erroneous data transfer.

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.
Sabouraud Dextrose Agar (SDA)	Plate 90 mm	20 plates	10035
	Slant tube	10 x 7 ml	30093
	Bottle	6 x 100 ml	402280
		6 x 200 ml	412280
		6 x 500 ml	470040
		25 x 200 ml	452280
	Dehydrated media	500 g	610103
		100 g	620103
		5 kg	6101035

Revision History

Revision	Release Date	Change Summary
0	2024-01-29	Updated layout and content in compliance with IVDR 2017/746, version reset to revision 0

In case of malfunctions or defects, contact immediately Liofilchem (*) or the local representative.

In case of incident associated with the device, notify immediately Liofilchem (*) or its local representative and the National Competent Authority.

*Please login to https://www.liofilchemstore.it/login.php (user ID and password required) and click on Complaint.

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

liofilchem.com/ifu-sds

Istruzioni per l'uso

ITALIANO

Sabouraud Dextrose Agar

Terreno per la coltivazione ed il conteggio di lieviti e muffe da campioni clinici e non clinici.

USO PREVISTO

Terreno per la coltivazione e l'enumerazione di lieviti e funghi da campioni clinici e non clinici. Il terreno è inteso come ausilio alla diagnosi, e richiede ulteriori test per completare i risultati diagnostici.

DESCRIZIONE

Sabouraud Dextrose Agar (SDA) è un terreno non selettivo utilizzato per la crescita ed il mantenimento di funghi patogeni e non patogeni da campioni clinici e non clinici. È anche utilizzato per il recupero ed il conteggio totale di lieviti e muffe nel monitoraggio ambientale.
Questo terreno è conforme con EN ISO 11133 per l'esame microbiologico degli alimenti, mangimi ed acqua, dove viene descritto come principale terreno di riferimento per effettuare test quantitativi su terreni di coltura specifici per funghi.
Il terreno è formulato secondo le raccomandazioni del metodo armonizzato nelle Farmacopee Statunitense (USP), Europea (EP) e Giapponese (JP) per l'esame microbiologico dei prodotti non sterili. Disponibile anche come piastre confezionate in triplo involucro sottovuoto, sterilizzate a raggi gamma, idonee per l'impiego nelle aree microbiologicamente controllate, come isolatori e camere bianche.

FORMULA TIPICA*	(g/litro)
Digerito Pancreatico di Caseina	5.0
Digerito Peptico di Tessuti Animali	5.0
Destrosio	40.0
Agar	15.0
Final pH 5.6 ± 0.2 at 25°C	

*Adattata e/o integrata per soddisfare le specifiche di performance richieste.

PRINCIPIO DEL METODO

Digerito pancreatico di caseina e digerito peptico di tessuti animali forniscono aminoacidi, azoto, carbonio, vitamine e minerali che supportano la crescita dei microrganismi. Il destrosio è una fonte di energia. L'agar è l'agente solidificante. L'alta concentrazione di destrosio ed il pH acido del terreno determinano la selettività per i funghi. Al terreno può essere aggiunto il cloramfenicolo per incrementare l'inibizione batterica ed il recupero dei dermatofiti.

PREPARAZIONE

Terreno disidratato
Sospendere 65 g di polvere in 1 litro di acqua distillata o deionizzata sterile. Mescolare bene. Riscaldare e bollire fino a completa dissoluzione. Sterilizzare in autoclave a 121 °C per 15 minuti.

Terreno in flaconi
Sciogliere il contenuto di un flacone a bagnomaria a 100°C (con il tappo leggermente svitato) fino a completa dissoluzione del terreno. Verificare, una volta fuso, la buona omogeneità del terreno capovolgendo il flacone dopo averne avvitato il tappo. Raffreddare a 45-50°C, mescolare bene evitando la formazione di bolle. Versare in piastre Petri in condizioni di asepsi.

MATERIALI RICHIESTI MA NON FORNITI

Forniture e apparecchiature microbiologiche standard come: autoclave, bagnomaria, piastre Petri sterili, provette, anse da inoculo, tamponi, incubatore, microrganismi per il controllo qualità.

CAMPIONI CLINICI

I campioni clinici dovrebbero essere prelevati nella fase acuta, prima della terapia antimicrobica (ove possibile) ed esaminati il prima possibile dopo la raccolta.
L'SDA non è adatto per l'inoculazione diretta di campioni di sangue.
Dovrebbero essere applicate le buone pratiche di laboratorio per la raccolta, il trasporto e la conservazione dei campioni clinici.
Fare riferimento alle linee guida specifiche per ulteriori informazioni sulla raccolta e la preparazione dei campioni.

PROCEDURA DEL TEST

Assicurarsi che non vi sia umidità visibile sulle piastre prima dell'uso.

Per l'uso in microbiologia medica

Strisciare il campione clinico il prima possibile dopo il suo arrivo in laboratorio per ottenere colonie isolate. Le provette pronte con terreno solidificato a becco di clarino sono principalmente destinate all'uso con colture pure per il mantenimento o per altri scopi. Le condizioni di incubazione possono variare in funzione della tipologia del campione e del microrganismo testato.

Per l'uso nell'esame di alimenti, mangimi, ed acque

Fare riferimento ad EN ISO 11133 per istruzioni specifiche.

Per l'uso nella microbiologia industriale

Controllo di prodotti non sterili

Fare riferimento alle procedure descritte nei capitoli armonizzati della Farmacopea.

Monitoraggio Passivo dell'Aria

Rimuovere il coperchio dalla piastra e lasciare il terreno esposto all'aria per un periodo di temo non superiore alle 4 ore (le piastre per sedimentazione - settle plate - riempite con 30 ml di terreno possono compensare la disidratazione del terreno durante lunghi periodi di incubazione). Le piastre possono essere posizionate secondo lo schema 1/1/1 (per 1 ora, circa 1 m dal pavimento, almeno 1 m dalle pareti o da altri ostacoli).

Monitoraggio dell'Igiene delle Superfici e del Personale

Utilizzare un tampone per il campionamento di superfici irregolari o servirsi del sampling template 10x10 (ref. 96762) per campionare una area ben definita della superficie da esaminare. Inoculare una piastra da 90 mm strisciando il tampone sulla superficie dell'agar. Inoltre, il terreno è adatto per il monitoraggio dell'igiene del personale e la determinazione della contaminazione microbica di guanti o mani (5-finger-print). Incubare le piastre a 20-25°C per 5-7 giorni o a 30-35°C per 24-48 ore.

INTERPRETAZIONE DEI RISULTATI

Può essere necessario trasferire la crescita dalle provette con terreno a becco di clarino a terreni in piastra per ottenere colture fungine pure. Esaminare le colonie che mostrano morfologia tipica. L'identificazione finale può richiedere test biochimici. La conta totale combinata lieviti/muffe (TYMC) viene considerata equivalente al numero di UFC trovate per ciascuna piastra.

Quando è prescritto un criterio per stabilire la qualità microbiologica, i risultai sono interpretati come di seguito indicato:

- 10^1 CFU: conta massima accettabile = 20;
- 10^2 CFU: conta massima accettabile = 200;
- 10^3 CFU: conta massima accettabile = 2000, e così via.

Nelle procedure destinate al monitoraggio dell'igiene ambientale e del personale, osservare giornalmente la formazione di colonie.

CONSERVAZIONE

La polvere è fortemente igroscopica, conservare la polvere a 10-30°C, in un ambiente asciutto, nel suo contenitore originale ben chiuso. Conservare i flaconi e le piastre preparate a 10-25°C al riparo dalla luce. Non utilizzare il prodotto oltre la data di scadenza indicata in etichetta o se il prodotto presenta segni di contaminazione o deterioramento.

VALIDITÀ

Terreno disidratato: 4 anni.
Terreno in flaconi: 2 anni.
Terreno in provette: 1 anno
Piastre pronte all'uso: 6 mesi.

CONTROLLO QUALITÀ

Aspetto del Terreno Disidratato: omogeneo, fine granulometria, beige chiaro.

Aspetto del Terreno Preparato: ambra chiaro, leggermente opalescente.

Risultati Attesi dei Test Colturali:

Ceppi di controllo		Inoculo	Incubazione	Criteri
Saccharomyces cerevisiae	WDCM 00058; (ATCC® 9763; NCTC 10716)	50-100 CFU	3-5 days / 22.5 ± 2.5°C	Buona Crescita ($P_R \geq 0.7$)
Aspergillus brasiliensis	WDCM 00053; (ATCC® 16404)		72 ± 2 h / 22.5 ± 2.5°C	
Candida albicans	WDCM 00054; (ATCC® 10231)		46 ± 2 h / 22.5 ± 2.5°C	
Candida albicans	WDCM 00054; ATCC® 10231		24 - 48 h / 32.5 ± 2.5°C	

Un rapporto di produttività (P_R) di 0.7 è equivalente ad un tasso di recupero del 70%.

Fare riferimento al certificato di analisi (CoA) relativo al lotto effettivo.

CARATTERISTICHE PRESTAZIONALI

I test di performance per Sabouraud Dextrose Agar sono stati effettuati utilizzando i ceppi CQ sopra elencati. I risultai ottenuti hanno soddisfatto i criteri stabiliti.

LIMITAZIONI

Risultati non validi possono essere causati da una scarsa qualità del campione, da una raccolta inadeguata del campione, da un trasporto inadeguato, da un'elaborazione inadeguata da parte del laboratorio o da una limitazione della tecnologia di analisi. L'operatore deve comprendere i principi delle procedure, compresi i limiti prestazionali, prima dell'operazione per evitare potenziali errori.

Il terreno può supportare la crescita di alcuni batteri. L'incubazione a 30-37°C è adatta per i lieviti, ma sopra i 30°C i dermatofiti possono essere inibiti.

È possibile che alcuni funghi (es. *Blastomyces dermatitidis*) non vengano recuperati in questo terreno a causa dell'elevato contenuto di carboidrati.

SDA è inteso come ausilio nella diagnosi delle malattie infettive, richiedendo ulteriori test per completare i risultati diagnostici.

AVVERTENZE E PRECAUZIONI

1) **Per uso diagnostico in vitro (IVD).**
2) **Solo per uso professionale di laboratorio.**
3) Gli operatori devono essere formati e avere una certa esperienza. Si prega di leggere attentamente le istruzioni prima di utilizzare il prodotto. L'affidabilità dei risultati del test non può essere garantita in caso di deviazioni dalle istruzioni contenute in questo documento.
4) Consultare la scheda di sicurezza (SDS) per informazioni sui pericoli e sulle pratiche di manipolazione sicure.
5) Non utilizzare se il prodotto o la confezione sembrano danneggiati.
6) Seguire le precauzioni standard. Tutti i campioni dei pazienti devono essere considerati potenzialmente infetti e maneggiati di conseguenza.
7) Maneggiare tutti i campioni come infetti utilizzando procedure di laboratorio sicure. Smaltire materiali pericolosi o biologicamente contaminati secondo le pratiche del proprio istituto.
8) Evitare la contaminazione incrociata dei campioni utilizzando puntali monouso e sostituendole dopo ogni campione.
9) Non mescolare reagenti di lotti diversi. Si prega di utilizzare il prodotto entro il periodo di validità.
10) Non mangiare, bere, fumare, applicare cosmetici o maneggiare lenti a contatto nelle aree in cui vengono manipolati reagenti e campioni umani.
11) I risultati devono essere interpretati da un professionista qualificato insieme alla storia del paziente, ai segni e sintomi clinici e ai fattori di rischio epidemiologici.
12) Assicurarsi che le apparecchiature di laboratorio siano calibrate e mantenute in conformità con la procedura del laboratorio.

13) Quando i risultati dei test vengono trasmessi dal laboratorio a un centro informatico, è necessario prestare attenzione per evitare trasferimenti di dati errati.

SMALTIMENTO DEI RIFIUTI

Lo smaltimento dei rifiuti deve essere effettuato in conformità alle normative nazionali e locali in vigore.

BIBLIOGRAFIA

Vedere i riferimenti alla fine di questo documento.

TABELLA DEI SIMBOLI

Vedere la tabella dei simboli alla fine di questo documento.

Il prodotto è disponibile in diverse configurazioni. Vedere l'elenco nella lingua inglese.

In caso di malfunzionamenti o difetti, contattare immediatamente Liofilchem (*) o il rappresentante locale.

In caso di incidente associato al dispositivo, avvisare immediatamente Liofilchem (*) o il suo rappresentante locale e l'Autorità Nazionale Competente.

*Si prega di effettuare il login su https://www.liofilchemstore.it/login.php (user ID e password richiesti) e cliccare su "Complaint".

Questo documento IFU e la SDS sono disponibili dal Support Center online: **liofilchem.com/ifu-sds**

References / Riferimenti

1. EN ISO 11133:2014+Amd1:2018. Microbiology of food, animal feed and water – Preparation, production, storage and performance testing of culture media.
2. European Pharmacopoeia 6.5 2009 2.6.13. Microbiological examination of non-sterile products: Test for specified microorganisms.
3. United States Pharmacopoeia 32 NF 27 2009 <62> Microbiological examination of non-sterile products: Test for specified microorganisms.
4. Japanese Pharmacopoeia 4.05 2008 Microbiological examination of non-sterile products: Test for specified microorganisms.
5. Sabouraud, R. 1892 Ann. Dermatol. Syphilol. 3:1061.

Table of Symbols / Tabella dei Simboli

LOT	Batch code / Codice lotto
REF	Catalogue number / Numero di catalogo
IVD	*In Vitro* Diagnostic Medical Device / Dispositivo Medico Diagnostico *in vitro*
	Manufacturer / Fabbricante
	Use by / Utilizzare entro
	Fragile, handle with care / Fragile, maneggiare con cura
	Temperature limitation / Limiti di temperatura
	Contains sufficient for <n> tests / Contenuto sufficiente per <n> saggi
	Consult instructions for use / Consultare le istruzioni per l'uso
	Do not reuse / Non riutilizzare
	Keep away from sunlight / Tenere al riparo dalla luce solare

Liofilchem® s.r.l.

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CE IVD

ENGLISH

ComASP® Colistin 0.25-16

System for colistin susceptibility testing with broth microdilution method, according to ISO 20776-1.

INTENDED PURPOSE

ComASP® Colistin 0.25-16 is an in vitro diagnostic product for antimicrobial susceptibility testing (AST) of clinical isolates based on growth of the test organism in the presence of various concentrations of an antimicrobial agent.

This ComASP® system is used for quantitative determination of the minimum inhibitory concentration (MIC) of colistin against the following non-fastidious organisms:

Gram-negative bacteria
Enterobacterales
Pseudomonas aeruginosa
Acinetobacter spp.

DESCRIPTION

Clinicians are re-thinking to colistin as therapeutic option for the treatment of severe infections caused by multidrug-resistant microorganisms, such as *Pseudomonas aeruginosa*, *Acinetobacter baumannii* and carbapenem-resistant *Enterobacterales*. However, colistin-resistant strains are worldwide disseminated leading to the need to determine the value of minimum inhibitory concentration (MIC) before the drug is prescribed. Although several commercial tests based on different techniques have been developed, broth microdilution (BMD) is considered the best method for performing colistin susceptibility testing so far.

ComASP® Colistin 0.25-16 is a 4-test panel containing the dried-up antibiotic in 7 two-fold dilutions:

Colistin concentration ranges 0.25 to 16 µg/ml.

The ComASP® system is a compact version of the broth microdilution (BMD) reference method allowing to perform the antimicrobial susceptibility testing of Colistin (polymyxin E) as recommended by international standards (i.e. CLSI, EUCAST, ISO) but in a simpler and less time-consuming way.

KIT CONTENT

- 4 Systems (panels) of ComASP® Colistin 0.25-16
  (panels individually packed in foil with silica gel desiccant)
- 16 Tubes of Mueller Hinton II Broth (3.6 ml)
- Sealing Film
- Instructions Sheet and Test Results Form

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as:

- loops - pipettes - physiological solutions - swabs - culture media - incubator
- test tubes - 0.5 McFarland turbidity standard - quality control organisms

CONFIGURATION

Test	Colistin Concentration (µg/ml)							
A	Growth	0.25	0.5	1	2	4	8	16
B	Growth	0.25	0.5	1	2	4	8	16
C	Growth	0.25	0.5	1	2	4	8	16
D	Growth	0.25	0.5	1	2	4	8	16

Growth indicates growth control: No antimicrobial agent in the well.

PRINCIPLE OF THE METHOD

Four bacterial isolates may be tested on a single panel.
All wells in a single row (test A, B, C or D) are rehydrated with a standardized microbial suspension.
After incubation for 16-20 hours the result is read and interpreted.

SPECIMEN COLLECTION AND PREPARATION

ComASP® Colistin 0.25-16 is not for use directly with clinical or other specimens. The microorganism to be tested must first be isolated on a nonselective culture medium, such as blood agar or tryptic soy agar (TSA). In case of mixed culture, selected colonies should be purified by subculturing. Differential media harboring chromogenic or fluorogenic substrates should not be used for the subculture. It is recommended that cultures be no more than 24 hours old unless additional incubation is required to achieve sufficient growth.

TEST PROCEDURE

1. Take a panel from its envelope and leave it at room temperature for 10 min,
   DO NOT DISCARD THE ENVELOPE until all 4 tests have been carried out.
2. Prepare a suspension of the test organism using either the direct colony suspension or growth method.
3. Standardize the suspension to the density of a McFarland 0.5 standard.
4. Optimally within 15 min of preparation, dilute the adjusted suspension 1:20 in saline; this will be the **Solution A**.
5. Add 0.4 ml of Solution A to a tube of MH II Broth* provided in the kit to obtain the **Solution B**.
6. Dispense 100 µl of Solution B into each well in a row.
7. Cover the panel with the lid provided and incubate at 36 ± 2°C for 16-20 hours in ambient air.

* Mueller Hinton II Broth (g/l): Beef Extract 3.0 g; Acid Hydrolysate of Casein 17.5 g; Starch 1.5 g; Distilled Water 1000 ml; pH 7.3 ± 0.1 (adjusted with appropriate salts to provide 20-25 mg/l of calcium and 10-12.5 mg/l of magnesium).

READING THE RESULTS

At the end of the incubation period observe the growth in the wells and establish the MIC, i.e. the lowest concentration of antibiotic that inhibits visible growth.

The result can be read visually or automatically. Reading by the naked eye can be improved by use of bright indirect lighting against a dark background.

Growth appears as turbidity or as a button at the bottom of the well (compare with the amount of growth in the growth-control well).

The growth-control well should be evaluated first. Make sure it is positive for growth. If not, check the viability of the colonies picked and repeat the test using a new row in the same panel or a new panel and a microbial culture of recent growth.

Note the results on the Test Results Form included in the kit (copy as many form as necessary).

INTERPRETATION OF THE RESULTS

The MIC obtained should be interpreted according to current EUCAST interpretative criteria (see below).

Antimicrobial agent	Organism	MIC Criteria (µg/ml)	
		S≤	R>
Colistin	*Enterobacterales*	2	2
	Pseudomonas spp.	4	4
	Acinetobacter spp.	2	2

Disclaimer: This breakpoint table might be out-of-date and does not replace EUCAST published guidelines, which always should be consulted before MIC categorization.

NOTE: Each individual panel is used for performing four tests. If less than 4 tests have been performed, use the film provided in the kit to seal the inoculated rows so to prevent any leakage of contaminated fluids. Then, return the panel into its own desiccant envelope and into the refrigerator (see STORAGE).

USER QUALITY CONTROL

Quality control of ComASP® Colistin 0.25-16 is performed using the following reference strains:

1. *Escherichia coli* ATCC® 25922; NCTC 12241
2. *Pseudomonas aeruginosa* ATCC® 27853; NCTC 12903
3. *Escherichia coli* NCTC 13846

Strain 1 and 2 are both susceptible to colistin and the acceptable **MIC QC range** is 0.25–2 µg/ml for *E. coli* and 0.5–4 µg/ml for *P. aeruginosa*. For *E. coli* NCTC 13846 (mcr-1 positive), which is instead resistant to colistin, the MIC target value is 4 µg/ml and should only on occasion be 2 or 8 µg/ml.

PERFORMANCE CHARACTERISTICS

Accuracy

Accuracy of ComASP® Colistin 0.25-16 was determined by evaluating the agreement of the AST system result with the result generated for the same isolate with the broth microdilution (BMD) reference method. To assess accuracy, Essential Agreement (EA) and bias of the method were calculated. EA occurs when the MIC of the ComASP® system and the reference method agree exactly or is within ± 1 dilution of each other. Bias of the method is the evaluation of test device results to determine whether the results that differ from the reference method are significantly skewed or predominantly in one direction. A total of 300 clinical isolates were tested by three operators. The following table summarizes performance data from these studies.

Antimicrobial agent	Organism	N	%EA	%Bias
Colistin	*Enterobacterales*	174	96.6	13.5
	Pseudomonas aeruginosa	69	100	
	Acinetobacter spp.	57	98.2	
	TOTAL	300	97.7	

N, Number of isolates **EA**, Essential Agreement

Reproducibility
99.3% of ComASP® Colistin 0.25-16 results (3 *E. coli*, 1 *K. pneumoniae*, 1 *E. aerogenes*, 2 *A. baumannii* and 3 *P. aeruginosa* tested in triplicate by 3 operators on 3 days) were within a doubling dilution of reference microdilution results.

Repeatability
100% of ComASP® Colistin 0.25-16 results (3 *E. coli*, 1 *K. pneumoniae*, 1 *E. aerogenes*, 2 *A. baumannii* and 3 *P. aeruginosa* tested in triplicate) were within a doubling dilution of reference microdilution results.

LIMITATIONS

Invalid results can be caused by poor specimen quality, improper sample collection, improper transportation, improper laboratory processing, or a limitation of the testing technology. The operator should understand the principles of the procedures, including its performance limitations, in advance of operation to avoid potential mistakes.

WARNINGS AND PRECAUTIONS

1) **For *in vitro* diagnostic use (IVD) only.**
2) **For laboratory professional use only.**
3) Operators must be trained and have certain experience. Please read the instructions carefully before using the kit. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this document.
4) Do not use if a panel, tube or packaging appears to be damaged.
5) Follow standard precautions. All patient specimens should be considered potentially infectious and handled accordingly.
6) Handle all specimens as if infectious using safe laboratory procedures. Dispose of hazardous or biologically contaminated materials according to the practices of your institution.
7) Avoid cross-contamination of samples by using disposable tips and changing them after each sample.
8) Do not mix reagents of different batches. Please use the kit within the validity period.
9) Do not eat, drink, smoke, apply cosmetics or handle contact lenses in areas where reagents and human specimens are handled.
10) Results should be interpreted by a trained professional in conjunction with the patient's history and clinical signs and symptoms, and epidemiological risk factors.
11) Ensure laboratory equipment is calibrated and maintained in accordance with the laboratory's procedure.
12) When test results are transmitted from the laboratory to an informatics centre, attention has to be done to avoid erroneous data transfer.

STORAGE

Store ComASP® Colistin 0.25-16 at 2-8°C in the original packaging. Once an envelope is opened the panel shall be used within 7 days and stored at 2-8°C. Do not use the panels beyond the expiry date indicated on the label. Eliminate without using if there are signs of deterioration.

DISPOSAL OF USED MATERIAL

After use, ComASP® Colistin 0.25-16 and material that has come into contact with the sample must be decontaminated and disposed of in accordance with guidelines used in the laboratory for decontamination and disposal of potentially infected material.

SUGGESTIONS FOR TROUBLESHOOTING

For out-of-range QC, first repeat the test with a pure culture or a freshly subcultured QC strain. If the issue is unresolved, follow this guidance for additional suggestions for troubleshooting out-of-range QC results and unusual clinical isolates results.

Observation	Probable Cause	Comments/Suggested Actions
MIC too low	Inoculum too light	Repeat using McFarland 0.5 turbidity standard or standardizing device. Check expiration date and proper storage if using barium sulfate or latex standards. Check steps in inoculum preparation and incubation procedure. Perform colony count check of growth control well immediately after inoculation and before incubation (*E. coli* ATCC® 25922 closely approximates 5 x 10^5 CFU/ml)
MIC too high	Inoculum too heavy	
MIC too high	Antimicrobial agent is degrading	Use alternative lot. Check STORAGE and package integrity
Skipped wells	Contamination. Inadequate mixing of inoculum or improper inoculation of panel	Repeat QC test

In case of other malfunctions or defects, contact immediately Liofilchem (*) or the local representative.

In case of incident associated with the device, notify immediately Liofilchem (*) or its local representative and the National Competent Authority.

*Please login to https://www.liofilchemstore.it/login.php (user ID and password required) and click on Complaint.

REFERENCES

1. Carretto E. *et al*. Clinical validation of the SensiTest™ Colistin, a broth microdilution based method to evaluate colistin MICs. J Clin Microbiol. 2018;17. pii: JCM.01523-17. http://jcm.asm.org/content/early/2018/01/11/JCM.01523-17
2. CLSI. Performance Standards for Antimicrobial Susceptibility Testing. 31st ed. CLSI Supplement M100S. Wayne, PA: Clinical and Laboratory Standards Institute; 2021.
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9. Sekyere JO *et al*. Comparative Evaluation of CHROMagar COL-APSE, MicroScan Walkaway, ComASP Colistin, and Colistin MAC Test in Detecting Colistin-resistant Gram-Negative Bacteria. Scientific Reports volume 10, Article number: 6221 (2020) https://www.nature.com/articles/s41598-020-63267-2
10. Galani I *et al*. Evaluation of ComASP™ Colistin (formerly SensiTest™ Colistin), a commercial broth microdilution-based method to evaluate the colistin minimum inhibitory concentration for carbapenem-resistant Klebsiella pneumoniae isolates. J Glob Antimicrobial Resistance. 2018; 15:123-126. DOI: 10.1016/j.jgar.2018.07.006

A Summary of Safety and Performance (SSP) will be available on Eudamed (subject to Eudamed availability). This summary is also available on request at liofilchem@liofilchem.com

Product	**Packaging**	**Ref.**
ComASP® Colistin 0.25-16	4x4 tests	75001

CE 0123 IVD

Table of Symbols

IVD	*In Vitro* Diagnostic Medical Device
REF	Catalogue number
LOT	Batch code
	Do not reuse
	Fragile, handle with care
CE 0123	Identification number of notified body
	Manufacturer
	Use by
Σ	Contains sufficient for <n> tests
	Consult instructions for use
	Temperature limits

Revision History

Revision	Release Date	Change Summary
3.1	04 Sep 2018	Added schematic representation of test procedure
4	29 Sep 2023	Updated layout and content in compliance with IVDR 2017/746

Note: Minor typographical, grammar, and formatting changes are not included in revision history; Revision numbers are incremented by decimals (e.g. version 1.1 to v 1.2) when there are minor modifications that do not affect requirements or procedures.

This document is also available from the online Support Center: **liofilchem.com/ifu-sds**

For other language translations, please contact your local Liofilchem representative or **liofilchem.com**

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Liofilchem®

Microbiology products since 1983

TEST RESULTS FORM

ComASP® Colistin 0.25-16

Patient data
Name
Surname
Age
Gender
Specimen
Notes

Indicate with the sign **+** the microbial growth.

Indicate with the sign **–** the absence of microbial growth.

Read the MIC and interpret the result according to the current EUCAST interpretive criteria.

TEST	COLISTIN CONCENTRATION (µg/ml)								MIC VALUE AND INTERPRETATION
A	Growth	0.25	0.5	1	2	4	8	16	.....
B	Growth	0.25	0.5	1	2	4	8	16	.....
C	Growth	0.25	0.5	1	2	4	8	16	.....
D	Growth	0.25	0.5	1	2	4	8	16	.....

Use a distinct test results form for each sample under investigation. A horizontal line should be drawn on the empty spaces related to an unused test, if any.

Date of Test	**Operator**

Peptone Bacteriological

Peptone obtained by enzymatic hydrolysis of animal tissue

PHYSIC-CHEMICAL CHARACTERISTIC

Solubility in water at 5%	Complete
Loss on drying	≤ 6.0%
Total nitrogen	≤ 12.5%
α-amino nitrogen AN	3-4.5%
Ash	5%

DESCRIPTION

Peptone Bacteriological is an enzymatic hydrolysate of meat that supplies a limpid, colorless and very stable watery solution. It is used in the preparation of culture media as a nitrogen source readily available for bacterial growth. It is a general use very nutritive peptone, with neutral pH. Peptone Bacteriological can be used as an ingredient of dehydrated culture media and need dissolution in distilled or deionized water and sterilization by autoclaving.

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.

DISPOSAL OF WASTE

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

PACKAGE

Code	Content	Packaging
611701	500 g	500 g of product in plastic bottle
621701	100 g	100 g of product in plastic bottle
6117015	5000 g	5000 g of product in plastic bottle

pH of THE MEDIUM

7.0 ± 0.5 (5% solution)

SHELF LIFE

4 years

QUALITY CONTROL

Dehydrated powder
Appearance: free-flowing, homogeneous.
Colour: white.

TABLE OF SYMBOLS

LOT	Batch code		Consult instructions for use		Manufacturer		Contains sufficient for <n> tests
REF	Catalogue number		Temperature limitation		Use by		Keep away from heat sources

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Instructions For Use

ENGLISH

Indole Test

Rapid test for detection of indole production.

INTENDED PURPOSE

Rapid test for evidence indole production in bacterial isolates. This test is intended as an aid in the diagnosis, requiring further tests to complete the diagnostic results.

DESCRIPTION

Indole test is used for the rapid qualitative determination of indole production as a first step in screening non- lactose fermenting colonies from agar plates.

KIT CONTENT

- 30 tubes of Indole Test
- 1 Kovac's reagent (3.0 ml)

METHOD PRINCIPLE

Bacteria that produce the enzyme tryptophanase can degrade the amino acid tryptophan into pyruvic acid, ammonia and indole. The indole produced reacts with p-dimethylaminobenzaldehyde of the Kovac's (indole) reagent to form a red-violet compound.
The production of indole from tryptophan is a characteristic absent in *Salmonella* but present in *E.coli*, *Morganella* and some species of *Klebsiella*, *Aerobacter* and *Citrobacter*.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as: incubator, inoculating loop, pipettes, culture media, physiological solution (0,85% saline), test tubes, 0.5 McFarland turbidity standard, quality control organisms.

REAGENTS

- A tube of Indole Test contains desiccated biochemical substrates and nutritive components such as phenol red and tryptone, which includes tryptophane.
- Kovac's Reagent consist of 5% p-dimethylaminobenzaldehyde dissolved in a solution of 25% hydrochloric acid and 75% isobutyl alcohol.

SPECIMEN

Collect specimens in sterile containers or with sterile swabs and transport to the laboratory. Process each specimen using procedures appropriate for that sample. This product is recommended for use only with pure cultures.

Refer to specific guidelines for more detailed information.

TEST PROCEDURE

1. Take the number of Indole Test tubes needed from the fridge and allow them to reach room temperature.
2. Add 0.3 ml of physiological solution to each tube.
3. Inoculate heavily with the test organism (*) from a fresh overnight pure culture.
4. Incubate at 35 ± 2 °C for up to 24 hours (**).
5. Examine tubes at intervals of 1,4 and 24 hours for broth colour change.
6. Add 2-3 drops of Kovac's Reagent to perform the Indole test.

*Microbial suspension equivalent to 0.5 McFarland turbidity standard (which corresponds to 1-2 x 10^8 CFU/ml for *E.coli*).

**Prolonged incubation may result in a false-positive test due to hydrolysis of proteins in the medium.

Note: Positive and negative controls should be run simultaneously with the organism to be tested (see QUALITY CONTROL)

INTERPRETING RESULTS

The appearance of a red ring at the surface of the broth within 30 seconds (after addition of Kovac's Reagent) indicates a positive indole reaction.

Further tests are required for confirmation.

STORAGE

2-8 °C in its original packaging. Keep away from sources of heat and avoid excessive changes of temperature. Use until the expiry date indicated on the label. Eliminate without using if there are signs of deterioration.

SHELF LIFE

1 year.

QUALITY CONTROL

Appearance of Indole Test: Yellowish powder, yellow solution once reconstituted.

Appearance of Kovac's Reagent: Yellow-green liquid, clear.

Control strain		Inoculum	Incubation	Characteristic reactions
Escherichia coli	WDCM 00013 ATCC® 25922 NCTC 12241	1-2 x 10^8 CFU/ml	Up to 24 h/ 35 ± 2°C	Positive reaction: Formation of a red ring
Proteus mirabilis	WDCM 00023 ATCC® 29906 NCTC 11938			Negative reaction: Yellow/ brown ring
Salmonella Tiphymurium	WDCM 00031 ATCC®14028 NCTC 12023			Negative reaction: Yellow ring

PERFORMANCE CHARACTERISTICS

Performance testing of Indole Test was carried out using the QC strains listed above. The results obtained met the established criteria.

LIMITATIONS

Due to nutritional variation, some strains may result in poor growth or fail to grow on this medium providing false negative results.
Allow at least 30 seconds for the color to develop before considering the indole test negative. Pigmented organisms may yield ambiguous results.
Indole production by certain anaerobes, such as *Clostridium* species, can be rapidly degraded resulting in false negative reactions.

WARNING AND PRECAUTIONS

1) **For *in vitro* diagnostic use (IVD).**
2) **For laboratory professional use only**.
3) Operators must be trained and have certain experience. Please read the instructions carefully before using the product. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this document.
4) Consult the Safety Data Sheet (SDS) for information regarding hazards and safe handling practices.
5) Do not use if the product or packaging appears to be damaged.
6) Follow standard precautions. All patient specimens should be considered potentially infectious and handled accordingly.
7) Handle all specimens as if infectious using safe laboratory procedures. Dispose of hazardous or biologically contaminated materials according to the practices of your institution.
8) Avoid cross-contamination of samples by using disposable tips and changing them after each sample.
9) Do not mix reagents of different batches. Please use the product within the validity period.
10) Do not eat, drink, smoke, apply cosmetics or handle contact lenses in areas where reagents and human specimens are handled.
11) Results should be interpreted by a trained professional in conjunction with the patient's history and clinical signs and symptoms, and epidemiological risk factors.
12) Ensure laboratory equipment is calibrated and maintained in accordance with the laboratory's procedure.

13) When test results are transmitted from the laboratory to an informatics centre, attention has to be done to avoid erroneous data transfer.

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.

ORDER INFORMATION

Product	Packaging	Ref.
Indole Test	30 tests	88017

Revision History

Revision	Release Date	Change Summary
1	2024-09-16	Added notice to report any malfunction, defect or incident.
0	2023-05-15	Updated layout and content in compliance with IVDR 2017/746, version reset to revision 0

In case of malfunctions or defects, contact immediately Liofilchem (*) or the local representative.

In case of incident associated with the device, notify immediately Liofilchem (*) or its local representative and the National Competent Authority.

*Please login to https://www.liofilchemstore.it/login.php (user ID and password required) and click on Complaint.

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

liofilchem.com/ifu-sds

Istruzioni per l'uso

ITALIANO

Indole Test

Test rapido per la rilevazione della produzione di indolo.

DESTINAZIONE D'USO

Test rapido per evidenziare la produzione di indolo su isolati batterici. Il test è inteso come ausilio alla diagnosi, e sono necessari ulteriori test per completare i risultati diagnostici.

DESCRIZIONE

Il test dell'indolo è usato per la determinazione qualitativa della produzione di indolo come primo passo nello screening di colonie non fermentanti il lattosio da piastre di agar.

CONTENUTO DEL KIT

- 30 provette di Indole Test
- 1 provetta di Kovac's Reagent (3.0 ml)

PRINCIPIO DEL METODO

I batteri che producono l'enzima triptofanasi sono in grado di degradare l'amminoacido triptofano in acido piruvico, ammoniaca e indolo. L'indolo prodotto reagisce con la p-dimetilamminobenzaldeide del Kovac's (Indole) Reagent formando un composto rosso-violetto.
La produzione di indolo dal triptofano è una caratteristica assente in *Salmonella* ma presente in *E.coli*, *Morganella* e alcune specie di *Klebsiella*, *Aerobacter* e *Citrobacter*.

MATERIALI RICHIESTI MA NON FORNITI

Forniture e apparecchiature microbiologiche standard come: incubatore, ansa da inoculo, pipette, terreni di coltura, soluzione fisiologica (0,85% salina), provette, standard di torbidità 0.5 McFarland, microrganismi per il controllo qualità.

REAGENTI

- Una provetta di Indole Test contiene substrati biochimici essiccati e componenti nutritive come rosso fenolo e triptone, che include il triptofano.
- Kovac's Reagent è costituito dal 5% di p-dimetilamminobenzaldeide disciolto in una soluzione di acido cloridrico al 25% e alcool isobutilico al 75%.

CAMPIONI

Raccogliere i campioni prelevati in contenitori sterili o con tamponi sterili e trasportarli in laboratorio. Trattare I campioni secondo la procedura appropriata per ognuno di loro. Questo prodotto è consigliato per l'uso esclusivamente con colture pure.
Fare riferimento alle linee guida specifiche per informazioni più dettagliate.

PROCEDURA DEL TEST

1. Prelevare dal frigorifero il numero necessario di provette di Indole Test ed aspettare che raggiungano la temperatura ambiente
2. Aggiungere 0.3 mL di soluzione fisiologica in ciascuna provetta.
3. Inoculare con il microrganismo da esaminare (*) proveniente da una coltura recente incubata over night.
4. Incubare a 35 ± 2°C fino a un massimo di 24 ore (**).
5. Osservare le provette a intervalli di 1, 4 e 24 ore per il cambio di colore del brodo.
6. Aggiungere 2-3 gocce di Kovac's Reagent per eseguire il Test Indolo

* Sospensione microbica equivalente allo standard di torbidità 0.5 McFarland (che corrisponde a 1-2 x 10^8 CFU/ml per *E. coli*).

** L'incubazione prolungata può dare luogo a un test falso positivo a causa dell'idrolisi delle proteine nel terreno.

Nota: I controlli positivi e negativi devono essere eseguiti simultaneamente con l'organismo da testare (vedere CONTROLLO DI QUALITÀ).

INTERPRETAZIONE DEI RISULTATI

Un risultato positivo per Indole Test è indicato dallo sviluppo di un anello rosso sulla superficie del brodo entro 30 secondi (dopo l'aggiunta del Kovac's Reagent).

Ulteriori test sono necessari per la conferma.

CONSERVAZIONE

Conservare a 2-8°C nella sua confezione originale. Tenere lontano da fonti di calore ed evitare eccessivi cambiamenti di temperatura. Utilizzare entro la data di scadenza indicata in etichetta. Eliminare se vi sono segni di deterioramento.

VALIDITÀ

1 anno.

CONTROLLO QUALITÀ

Aspetto dell'Indole Test: Polvere giallastra, soluzione gialla una volta ricostruita.
Aspetto Kovac's Reagent: Liquido giallo-verde, limpido.

Ceppi di controllo		Inoculo	Incubazione	Caratteristiche di reazione
Escherichia coli	WDCM 00013 ATCC® 25922 NCTC 12241	1-2 x 10^8 CFU/ml	Fino a 24 h/ 35 ± 2°C	Reazione positiva: Sviluppo di un anello rosso
Proteus mirabilis	WDCM 00023 ATCC® 29906 NCTC 11938			Reazione negativa: Sviluppo di un anello giallo / marrone
Salmonella Tiphymurium	WDCM 00031 ATCC®14028 NCTC 12023			Reazione negativa: Sviluppo di un anello giallo

CARATTERISTICHE PRESTAZIONALI

Il controllo delle prestazioni dell'Indole Test è stato eseguito utilizzando i ceppi sopra elencati. I risultati ottenuti hanno soddisfatto i criteri stabiliti.

LIMITAZIONI

A causa della variazione nutrizionale, alcuni ceppi possono causare una scarsa crescita o non crescere su questo terreno, fornendo risultati falsi negativi.

Attendere almeno 30 secondi affinché il colore si sviluppi prima di considerare negativo il test dell'indolo.

Gli organismi pigmentati possono produrre risultati ambigui.

La produzione di indolo da parte di alcuni anaerobi, come la specie *Clostridium*, può essere rapidamente degradata con conseguenti reazioni false negative.

AVVERTENZE E PRECAUZIONI

1) **Per uso diagnostico in vitro (IVD).**
2) **Solo per uso professionale di laboratorio.**
3) Gli operatori devono essere formati e avere una certa esperienza. Si prega di leggere attentamente le istruzioni prima di utilizzare il prodotto. L'affidabilità dei risultati del test non può essere garantita in caso di deviazioni dalle istruzioni contenute in questo documento.
4) Consultare la scheda di sicurezza (SDS) per informazioni sui pericoli e sulle pratiche di manipolazione sicure.
5) Non utilizzare se il prodotto o la confezione sembrano danneggiati.
6) Seguire le precauzioni standard. Tutti i campioni dei pazienti devono essere considerati potenzialmente infetti e maneggiati di conseguenza.
7) Maneggiare tutti i campioni come infetti utilizzando procedure di laboratorio sicure. Smaltire materiali pericolosi o biologicamente contaminati secondo le pratiche del proprio istituto.
8) Evitare la contaminazione incrociata dei campioni utilizzando puntali monouso e sostituendole dopo ogni campione.
9) Non mescolare reagenti di lotti diversi. Si prega di utilizzare il prodotto entro il periodo di validità.

10) Non mangiare, bere, fumare, applicare cosmetici o maneggiare lenti a contatto nelle aree in cui vengono manipolati reagenti e campioni umani.
11) I risultati devono essere interpretati da un professionista qualificato insieme alla storia del paziente, ai segni e sintomi clinici e ai fattori di rischio epidemiologici.
12) Assicurarsi che le apparecchiature di laboratorio siano calibrate e mantenute in conformità con la procedura del laboratorio.
13) Quando i risultati dei test vengono trasmessi dal laboratorio a un centro informatico, è necessario prestare attenzione per evitare trasferimenti di dati errati.

SMALTIMENTO DEI RIFIUTI

Lo smaltimento dei rifiuti deve essere effettuato in conformità alle normative nazionali e locali in vigore.

BIBLIOGRAFIA

Vedere i riferimenti bibliografici alla fine di questo documento

TABELLA DEI SIMBOLI

Vedere la tabella dei simboli alla fine di questo documento.

INFORMAZIONI PER L'ORDINE

Prodotto	**Confezionamento**	**Rif.**
Indole Test	30 tests	88017

In caso di malfunzionamenti o difetti, contattare immediatamente Liofilchem (*) o il rappresentante locale.

In caso di incidente associato al dispositivo, avvisare immediatamente Liofilchem (*) o il suo rappresentante locale e l'Autorità Nazionale Competente.

*Si prega di effettuare il login su https://www.liofilchemstore.it/login.php (user ID e password richiesti) e cliccare su "Complaint".

Questo documento IFU e la SDS sono disponibili dal Support Center online: **liofilchem.com/ifu-sds**

References / Riferimenti

1. EN ISO 11133:2014+Amd1:2018+Amd2:2020. Microbiology of food, animal feed and water − Preparation, production, storage and performance testing of culture media.
2. Public Health England. (2019). Urease test. UK Standards for Microbiology Investigations. TP 36 Issue 4. https://www.gov.uk/guidance/uk-standards-for-microbiology-investigations-smi-quality-and-consistency-in-clinical-laboratories
3. Public Health England. (2018). Indole test. UK Standards for Microbiology Investigations. TP 19 Issue 4
4. Isemberg H.D. and Sundheim L.H.(1980) Indole reactions in bacteria. J. Bacteriol. 75:682-690. Baltimore, Williams & Wilkins.
5. MacFaddin J.F. (1980) Biochemical tests for identification for medicalbacteria. 2nd ed. 173-183. Baltimore, Williams & Wilkins.
6. Blazevic D.J. and Ederer, G.M.(1975) Principles of biochemical tests in diagnostic microbiology. 63-67. New York, John Wiley & Sons.

Table of Symbols / Tabella dei Simboli

LOT	Batch code / Codice del lotto
REF	Catalogue number / Numero di catalogo
IVD	*In Vitro* Diagnostic Medical Device / Dispositivo Medico Diagnostico *in vitro*
	Manufacturer / Fabbricante
	Use by / Utilizzare entro
	Fragile, handle with care / Fragile, maneggiare con cura
	Temperature limitation / Limiti di temperatura
	Contains sufficient for <n> tests / Contenuto sufficiente per <n> saggi
	Consult instructions for use / Consultare le istruzioni per l'uso
	Do not reuse / Non riutilizzare
	Keep away from sunlight / Tenere al riparo dalla luce solare

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CE IVD

Glucose

Glucose for bacteriological use

PHYSIC-CHEMICAL CHARACTERISTIC

Solubility in water at 5%	Complete
Humidity	≤ 0.50 %
Molar weight	180
Acidity	≤ 0.15 ml
Ash	≤ 0.10 %
Granulometry	98% lower to 70 Mesh 45% lower to 1400 Mesh

DESCRIPTION

Glucose is used as a source of energy readily available for bacteria in fermentation tests (i.e. *Salmonella typhimurium* and *Escherichia coli* ferment glucose). It is free from other sugars and from starch, proteins and metals. In liquid media is used generally at 0.5% concentration; high concentrations are used for the preparation of solid media. Glucose can be used as an ingredient of dehydrated culture media and need dissolution in distilled or deionized water and sterilization preferably by filtration.

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.

DISPOSAL OF WASTE

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

PACKAGE

Code	Content	Packaging
611601	500 g	500 g of product in plastic bottle
621601	100 g	100 g of product in plastic bottle
6116015	5000 g	5000 g of product in plastic bottle

SHELF LIFE

4 years

QUALITY CONTROL

Dehydrated powder
Appearance: free-flowing, homogeneous.
Colour: white.

TABLE OF SYMBOLS

LOT	Batch code		Consult instructions for use		Manufacturer		Contains sufficient for <n> tests
REF	Catalogue number		Temperature limitation		Use by		Keep away from heat sources

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STRIP Control GST E6

Biological indicators for steam sterilization processes with *G. stearothermophilus* ATCC® 7953 spores inoculated on paper strips.

DESCRIPTION

STRIP Control GST E6 are used to monitor the effectiveness of steam sterilizing process. These biological indicators (BI's) are produced under strictly controlled conditions in order to satisfy the requirements in USP, ISO 11138 and EN 866 standards.

STRIP Control GST E6 contain bacterial spores on special filter paper strips and glass test tubes with screw cap. Each tube contains a validated growth medium with pH indicator. Strips are inoculated with spores of *Geobacillus stearothermophilus* (ATCC 7953) in predefined concentrations.

A Certificate of Analysis including population, strain, D-value (121°C), Z-Value (115°C, 121°C, 124°C), survival time, kill time, lot number and expiration date is inserted in the packaging.

COMPOSITION

Strips contain *G. stearothermophilus* (ATCC 7953) spores in concentrations 1-5 x10^6 CFU/strip. Each strip is enclosed in an envelope; lot number and expiration date are printed on each envelope.

Tubes contain 4 ± 0.5 mL of a sterile modified soybean casein digest broth with a pH indicator (Steri-Test medium); lot number and expiration date are printed on each tube.

PRINCIPLE

Spores are completely killed off after the sterilization at 121°C. If no spores survive, there is no growth during the subsequent incubation and the medium inside the ampoule remains violet. A failure in the sterilization process (lower temperature and/or shorter sterilization time), is detected by the colour change of the medium to yellow due to spores survival and proliferation of bacteria.

TECNIQUE

1. Place one or more strips of STRIP Control GST E6 (each in its envelope) in the most challenging location of the steam sterilizer such as on the bottom shelf, near the door, and over the drain. The number of strips to be used will depend on the size of the sterilize chamber and/or regional requirements or load in the sterilizer. Typically, for autoclaves having an internal volume lower than or equal to 250 litres, two strips are used for each selected point of the autoclave. For autoclaves with volume higher than 250 litres, six or more strips can be used per point.
2. Remove the strip (still in its envelope) after sterilization cycle, aseptically open the envelope with sterile scissors or by tearing the edges, transfer the strip to a tube of Steri-Test medium included in the package.
3. Incubate the tube containing the strip at 55-60°C (131-140°F) for 7 days or for a different time validated by the user.
4. Incubate, at the same conditions of time and temperature, a tube of Steri-Test medium with a strip not submitted to the sterilization cycle and belonging to the same batch, as spore growth control (positive control).
5. Examine the Steri-Test medium and interpret the result as per EVALUATION TABLE: a colour change of medium from violet/clear to yellow/turbid indicates microbial growth and therefore an unsuccessful sterilization. On the contrary, the persistence of the initial colour of the medium (violet/clear) indicates absence of microbial growth and therefore a successful sterilization.

INTERPRETATION OF RESULTS

Geobacillus stearothermophilus (ATCC 7953) spores are killed off if the sterilization cycle has been efficient: in this case the broth contained in STRIP Control GST E6 remains violet/clear even after incubation at 55-60°C (131-140°F) for 7 days or for the selected time. If the sterilization cycle has not been efficient, spores partially survive and the Steri-Test medium turns yellow/turbid after incubation at 55-60°C (131-140°F) for 7 days or for the selected time. The Steri-Test medium not submitted to the sterilization cycle and used as spore growth control has to turn yellow/turbid after incubation. On the contrary, the test must be repeated after having investigated the causes of the negative result.

EVALUATION TABLE		
MEDIUM COLOUR	**SPORE**	**STERILIZATION**
Violet / clear	Killed off	Successful
Yellow / turbid	Vital	Unsuccessful

Note: Any colour change with no turbidity (the medium remains clear after incubation) indicates a successful sterilization cycle (no growth).

STORAGE

Store at room temperature (10-25°C). In these conditions the product maintains its validity until the expiry date indicated on the label.

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 professional use only and must be used by properly trained operators.

DISPOSAL OF WASTE

After use, sterilize positive vials (yellow/turbid) in autoclave at 121°C for at least 30 minutes and eliminate them in accordance with the procedures in the laboratory.

REFERENCES

- United States Pharmacopoeia latest edition.
- Deutsches Arzneibuch latest edition.
- European Pharmacopoeia latest edition.
- ISO 11138 and EN 866 latest edition.

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PRODUCT SPECIFICATIONS

NAME
STRIP Control GST E6

PRESENTATION
Paper strips inoculated with *Geobacillus stearothermophilus* ATCC 7953 spores in predefined concentrations and glass tubes with a growth medium

STORAGE
10-25°C

PACKAGING

REF	CONTENT	PACKAGING
91055	20 unities 1 Instruction Sheet 1 Certificate of Performance	20 unities in thermally soldered envelope 20 envelopes + 20 tubes in cardboard boxes

TECHNICAL PROPERTIES

STRIP
Spore carrier type: filter paper
Spore carrier: approximately 38 x 6 mm paper strip
Species: *Geobacillus stearothermophilus* ATCC 7953
Mean Population Recovery: 1 x 10^6 - 5 x 10^6 spores/strip
Purity: Bacterial contaminates less than 1 percent of the labeled population
Resistance data: decimal reduction time (D-Value), survival time and kill time

Steri-Test Medium (growth medium)
pH: 7.4 ± 0.1
Fill volume: 4.0 ± 0.5 mL
Tube: glass tube; height approximately 15 ± 1 mm (screw cap)
Growth promotion: meets USP current edition
Colour: violet (colour change to yellow and/or turbidity indicates bacterial growth)

USE
Biological indicators STRIP Control GST E6 are used for regular control of steam sterilization cycles (i.e. 15 minutes at 121°C) and control of any steam autoclave functionality

TECHNIQUE
Refer to technical sheet of the product

APPEARANCE
Strips are white in colour. The medium is violet, clear

QUALITY CONTROL
1. Control of general characteristics, label and print
2. Purity: < 1% contamination. No moulds
3. Heat shocked population: 1-5 x 10^6 Spores/strip
4. D-Value (121°C): 1.5-4.5 minutes
5. Z-Value (115°C, 121°C, 124°C): ≥ 6°C
6. Growth: 55-60°C for 18-24 hours; colour change of the medium from violet/clear to yellow/turbid

SHELFLIFE
3 years

TABLE OF SYMBOLS

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

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Lactose

Lactose for bacteriological use.

PHYSIC-CHEMICAL CHARACTERISTIC

Solubility in water at 5%	Complete
Humidity	0.10 %
Ash	0.05%

DESCRIPTION

Lactose is used as a source of fermentable carbohydrates for bacteria in fermentation tests (i.e. Escherichia coli ferments lactose whilst Salmonella typhimurium gives a negative reaction). It is not hygroscopic and is free from othersugars and from starch, proteins and metals. Lactose can be used as an ingredient of dehydrated culture media and need dissolution in distilled or deionized water and sterilization preferably by filtration.

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.

DISPOSAL OF WASTE

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

PACKAGE

Code	Content	Packaging
610498	500 g	500 g of product in plastic bottle
620498	100 g	100 g of product in plastic bottle

pH of THE MEDIUM

4.3 (5% solution)

SHELF LIFE

4 years

QUALITY CONTROL

Dehydrated powder
Appearance: free-flowing, homogeneous.
Color: white.

TABLE OF SYMBOLS

LOT	Batch code		Consult instructions for use		Manufacturer		Contains sufficient for <n> tests
REF	Catalogue number		Temperature limitation		Use by		Keep away from heat sources

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