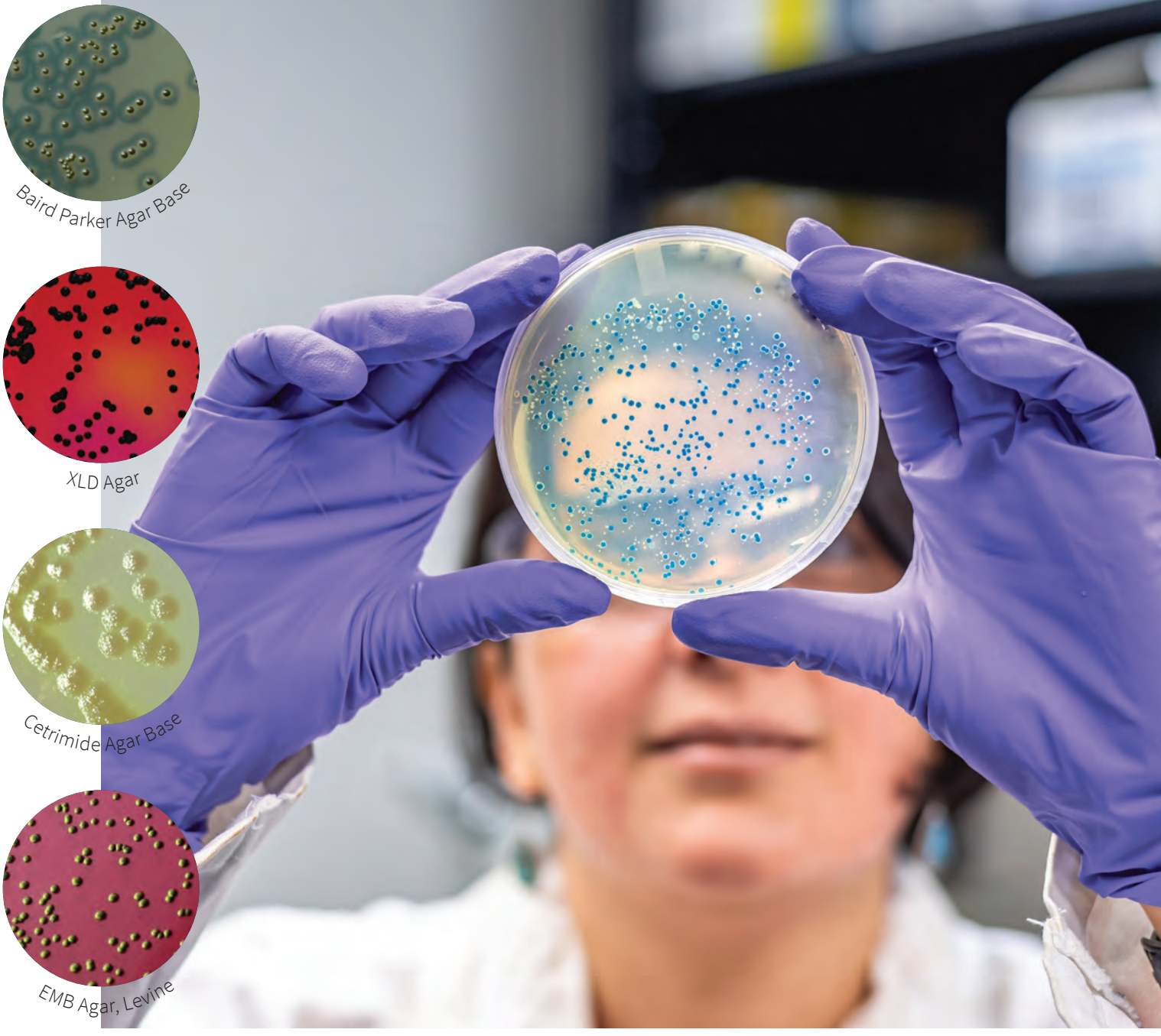
Culture Media Guide

For Pharmaceuticals



HiMediaLaboratories™
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HIMEDIA®

Striving for Excellence

PRIDE OF महाराष्ट्र AWARDS (FOR CORPORATE EXCELLENCE)

MIEDA & SME Chamber of Commerce

25th Foundation Day Function

21st February 2019

Dr. G. M. Warke - HiMedia Brand Ambassador of Maharashtra

Dr. G. M. Warke, Founder & CMD, HiMedia Laboratories Pvt. Ltd., honoured with the PRIDE OF MAHARASHTRA AWARD 2018 for the "BRAND AMBASSADOR OF MAHARASHTRA (Category: Research & Innovation)" by Dr. Raghunath Mashelkar, President of Global Research Alliance & Former Director General of Council of Scientific & Industrial Research (CSIR) on 21st February 2019 at Mumbai.

Awards initiated by Maharashtra Industrial and Economic Development Association (MIEDA). (L to R) Shri. Chandrakant Salunkhe, Founder & President, Maharashtra Industrial & Economic Development Association and SME Chamber of India, Dr. G. M. Warke , Dr. Raghunath Mashelkar.

HiMedia - The Best Company of the Year

Dr. Raghunath Mashelkar, President of Global Research Alliance & Former Director General of Council of Scientific & Industrial Research (CSIR) presenting the PRIDE OF MAHARASHTRA AWARD 2018 for the "BEST COMPANY OF THE YEAR (Category: Research & Innovation)" to Dr. G. M. Warke, Founder & CMD, HiMedia Laboratories Pvt. Ltd.,

Mr. V. M. Warke-Co-founder & Director, and Mrs. Saroj G. Warke-Cofounder & Director on 21st February 2019 at Mumbai.

Awards initiated by Maharashtra Industrial and Economic Development Association (MIEDA). Shri. Chandrakant Salunkhe, Founder & President, MIEDA and SME Chamber of India was present.

OUR QUALITY COMMITMENT

IQNet

THE INTERNATIONAL CERTIFICATION NETWORK

CERTIFICATE

Quality Austria
has issued an IQNet recognized certificate that the organization:

HiMedia Laboratories Private Limited
Unit-1 : B-4-5-6 / MIDC, Palkhed, Dindori,Nashik – 422 202 Maharashtra,India
Registered Office : 23, Vadhani Industrial Estate, LBS Marg, Ghatkopar (West) Maharashtra, India

for the following scope:

Design and Development, Manufacturing and Supply of Biosciences Products for application in Microbiology (including Dehydrated culture Media, Antimicrobial Susceptibility Systems, Culture Media Bases and Bacteriological Differentiation Aids), Animal Tissue Culture, Plant Tissue Culture and Molecular Biology

EAC: 19.2

has implemented and maintains a

QUALITY MANAGEMENT SYSTEM

which fulfils the requirements of the following standard

ISO 9001:2015

This attestation is directly linked to the IQNet Partner's original certificate and shall not be used as a stand-alone document

Issued on: 2019-04-08
Validity date: 2022-03-31
Quality Austria certified since: 2015-12-29

Registration Number: AT-17285/0

Alex Stoichitoiu
President of IQNet

Mag. Friedrich Khuen-Belasi
Authorised Representative of Quality Austria

qualityaustria

IQNet

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EAC: 19.2

has implemented and maintains a

QUALITY MANAGEMENT SYSTEM

which fulfils the requirements of the following standard

EN ISO 13485:2016

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Issued on: 2019-04-09
Validity date: 2022-03-31
Quality Austria certified since: 2017-11-21

Registration Number: AT-00275/0

Alex Stoichitoiu
President of IQNet

Mag. Friedrich Khuen-Belasi
Authorised Representative of Quality Austria

qualityaustria

National Accreditation Board for Testing and Calibration Laboratories
(A Constituent Board of Quality Council of India)

CERTIFICATE OF ACCREDITATION

HIMEDIA LABORATORIES PVT. LTD.
(QC MICROBIOLOGY LABORATORY)

has been assessed and accredited in accordance with the standard

ISO/IEC 17025:2005

"General Requirements for the Competence of Testing & Calibration Laboratories"

for its facilities at

19, 20, 21, Vadhani Industrial Estate, Dist.-Thane, Mumbai, Maharashtra

in the field of

TESTING

Certificate Number TC-6395
Issue Date 12/10/2017 Valid Until 11/10/2019

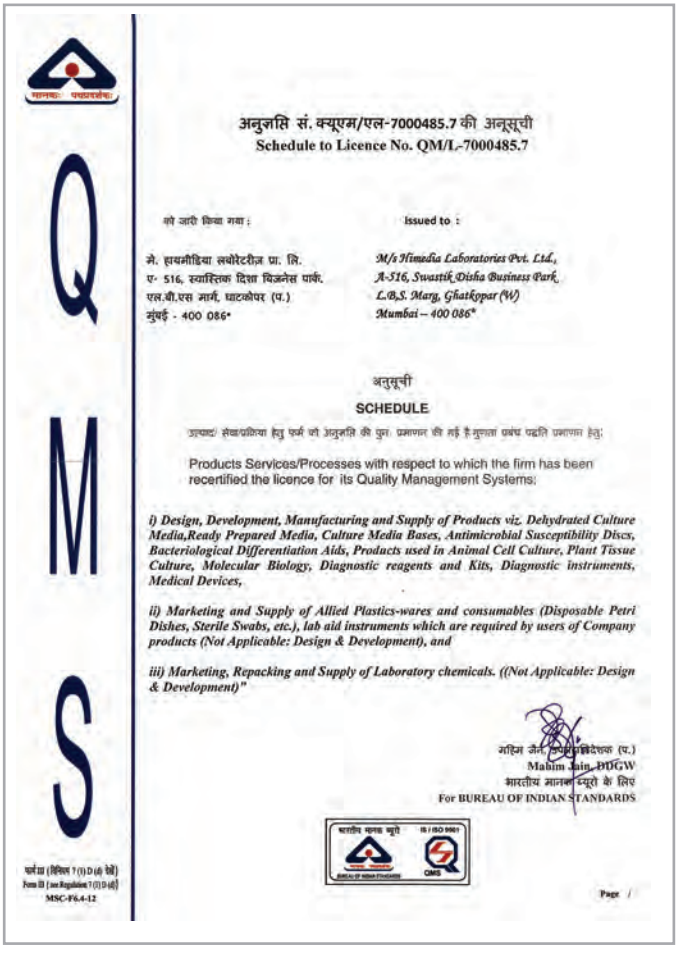
This certificate remains valid for the Scope of Accreditation as specified in the annexure subject to continued satisfactory compliance to the above standard & the relevant requirements of NABL.

(To see the scope of accreditation of this laboratory, you may also visit NABL website www.nabl-india.org)

Signed for and on behalf of NABL

N. Venkateswaran
Program Director

Anil Relia
Chief Executive Officer



अनुज्ञप्ति सं. क्यूएम/एल-7000485.7 की अनुसूची
Schedule to Licence No. QM/L-7000485.7

को जारी किया गया :
मे. हायमीडिया लबोरेटरीज़ प्रा. लि.
ए- 516, स्वास्तिक दिशा बिजनेस पार्क,
एल.बी.एस मार्ग, घाटकोपर (प.)
मुंबई - 400 086*

Issued to :
M/s Himedia Laboratories Pvt. Ltd.,
A-516, Swastik Disha Business Park,
L.B.S. Marg, Ghatkopar (W)
Mumbai – 400 086*

अनुसूची
SCHEDULE

उत्पाद/ सेवा/प्रक्रिया हेतु फर्म को अनुज्ञप्ति की पुन: प्रमाणन की गई है सुसंगत संबंध पद्धति प्रमाणन हेतु:

Products Services/Processes with respect to which the firm has been recertified the licence for its Quality Management Systems:

i) Design, Development, Manufacturing and Supply of Products viz. Dehydrated Culture Media,Ready Prepared Media, Culture Media Bases, Antimicrobial Susceptibility Discs, Bacteriological Differentiation Aids, Products used in Animal Cell Culture, Plant Tissue Culture, Molecular Biology, Diagnostic reagents and Kits, Diagnostic instruments, Medical Devices,

ii) Marketing and Supply of Allied Plastics-wares and consumables (Disposable Petri Dishes, Sterile Swabs, etc.), lab aid instruments which are required by users of Company products (Not Applicable: Design & Development), and

iii) Marketing, Repacking and Supply of Laboratory chemicals. ((Not Applicable: Design & Development)"

महिम जैन, उपमहानिदेशक (प.)
Mahim Jain, DDG(W)
भारतीय मानक ब्यूरो के लिए
For BUREAU OF INDIAN STANDARDS

MSC-F6.4-12

This NABL accreditation in accordance with ISO/IEC 17025:2005 standard in the discipline of Biological Testing of Microbiological Media used for food and water analysis.

OUR QUALITY COMMITMENT

CERTIFICATE

Quality Austria Central Asia Private Limited
(A Division of Peacock Global Company)
Awards this Certificate to

HiMedia Laboratories Pvt. Ltd.
Unit I : B/4-6, MIDC, Palkhed, Dindori, Nashik-422 202, Maharashtra, India
Unit II : W-239(B), MIDC Phase II, Shivaji Udyog Nagar, Dombivli, District Thane – 421 204, Maharashtra, India
Unit III : D-61 MIDC, Phase-II, Near Shani Mandir, Dombivli, District Thane – 421204, Maharashtra, India

Unit I : Manufacture & supply of Biosciences products for applications in Microbiology (includes Dehydrated Culture Media, Culture Media Bases, Antimicrobial Susceptibility Systems & Bacteriological Differentiation Aids), Animal Cell Culture, Plant Tissue Culture and Molecular Biology
Unit II : Manufacture and supply of Sterile Ready Prepared Media
Unit III : Manufacture and supply of Sterile Ready Prepared Media

This Certificate confirms the application and further development of an effective

WHO GMP Compliance System

Complying with the requirement of

WHO GMP Guidelines

Report No.:QACA/WHO/069
Issue Date: 21/12/2016
Expiry Date: 20/12/2019

qualityaustria central asia
Succeed with Quality

India: 20 Dec 2016
Quality Austria Central Asia Private Limited (A division of Peacock Global company)

Alok Kumar
Country Head

WHO GMP CERTIFIED

CE

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CERTIFICATE

Quality Austria - Trainings, Zertifizierungs und Begutachtungs GmbH awards this qualityaustria certificate to the following organisation:

This qualityaustria certificate confirms the application and further development of an effective

HiMedia Laboratories Private Limited
Unit-1 : B-4-5-6 / MIDC, Palkhed, Dindori, Nashik – 422 202 Maharashtra,India

Registered Office : 23, Vadhani Industrial Estate, LBS Marg, Ghatkopar (West) Maharashtra, India

Design and Development, Manufacturing and Supply of Biosciences Products for application in Microbiology (including Dehydrated culture Media, Antimicrobial Susceptibility Systems, Culture Media Bases and Bacteriological Differentiation Aids), Animal Tissue Culture, Plant Tissue Culture and Molecular Biology

The validity of the qualityaustria certificate will be maintained by annual surveillance audits and one renewal audit after three years.

QUALITY MANAGEMENT SYSTEM
complying with the requirements of standard
ISO 9001:2015

Registration No.: 17285/0
Date of initial issue: 29 December 2015
Valid until: 31 March 2022

Vienna, 08 April 2019

Quality Austria - Trainings, Zertifizierungs und Begutachtungs GmbH,
AT-1010 Vienna, Zelinkagasse 10/3

Konrad Scheiber
General Manager

Dr. Mag. Anni Koubek
Specialist representative

The current validity of the certificate is documented exclusively on the Internet under http://www.qualityaustria.com/en/cert EAC: 19.2

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CERTIFICATE

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The validity of the qualityaustria certificate will be maintained by annual surveillance audits and one renewal audit after three years.

QUALITY MANAGEMENT SYSTEM
complying with the requirements of standard
EN ISO 13485:2016
Medical devices - Quality management systems - Requirements for regulatory purposes

Registration No.: 00275/0
Date of initial issue: 21 November 2017
Valid until: 31 March 2022

Vienna, 08 April 2019

Quality Austria - Trainings, Zertifizierungs und Begutachtungs GmbH,
AT-1010 Vienna, Zelinkagasse 10/3

Konrad Scheiber
General Manager

Ing. Andreas Aichinger, MSc
Specialist representative

The current validity of the certificate is documented exclusively on the Internet under http://www.qualityaustria.com/en/cert EAC: 19.2

HIMEDIA

Introduction

The Pharmaceutical industry having the highest rate of innovation has always operated under stringent regulations. Quality is always at the center stage for pharmaceutical manufacturing products & processes. With the increasing stringency in GMP norms, foolproof systems are to be installed at every step of pharmaceutical process chain. Product & process quality control is an inseparable part of this process chain.

Reference standards, guidelines, manuals like EP/ USP/JP/BP/IP/GMP lay down the guiding principles for quality assessment. The implementation of quality systems requires lab chemicals of proven track record.

HiMedia's endeavor to implement latest regulatory norms has churned out its own quality management system. The system continuously upgrades, tests & only releases products complying with international norms & standards. The US FDA registered facility and WHO-GMP compliant with ISO 9001:2015/ ISO 13485:2016 quality certifications in place encompasses the operation of a quality management system. The documentation support provide with this system not only provides documented compliance during audits but also concentrates on meeting customer expectations and conformity with regulatory requirements.

As a continual effort to provide assistance to customer, HiMedia, is delighted to present a user friendly, informative and yet comprehensive guide, for selection of appropriate culture media. This revised edition covers the essentials in identifying the medium of choice and brings forth the composition, intended use and directions to guide the discretion of the user. This edition also highlight's the list of vegetable peptone based HiVeg™ Media and Chemically defined media (HiCynth™ Media) suitable for pharmaceutical application- the most innovative and imperative product of the era. This animal meat-free (non-animal base) culture medium fulfills the growing demand of pharmaceutical industry for non-animal based materials that eliminate the risk of agents that cause Bovine or transmissible spongiform encephalopathy's (BSE/TSEs), such as mad cow disease and variant Creutzfeldt-Jakob diseases in humans. These media are proven safe and provide total solution to Media fill trials in the industry.

This comprehensive guide would in fact be the frequent reference to microbiologist throughout the pharmaceutical industry. However if the user still needs further information, HiMedia has the solution in form of The HiMedia Manual and The HiVeg Manual. As a part of evolution and improvisation HiMedia requests from its valued customers suggestions and corrections if any.

With a desire to serve the scientific genre and industry, offering world class quality products, HiMedia extends its services to the satisfaction of customers.

The harmonization of USP/EP/BP/JP/IP currently demands usage of new testing protocols and subsequently newer dehydrated culture media compositions. We at HiMedia are ready with our DCM range of products required for pharmaceutical product validation and routine purpose which are in line with new harmonized specifications.

HIMEDIA®
For Life is Precious

ATCC®

Credible leads to incredible™

Introducing Zero passage original ATCC cultures from ATCC: The Global Bioresource centre

Are you looking for Microbial Cultures from ATCC , Cell Biology Research products....

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» ATCC Standard Development Organization
» Proficiency Standard Program
» ATCC Virus Reference Material
» Certified Reference Materials (CRMs)

Contents

Definitions:

- Purified Water : Water prepared from drinking water using unit operations that include deionization, distillation, ion exchange, reverse osmosis,filtration or other suitable procedures.
- Distilled Water : Water that has virtually all of its impurities removed through distillation (boiling the water and re-condensing the steam into a clean container, leaving contaminants behind).
- Water R : Reagent Grade Water is prepared by either distillation, mixed bed deionization and reverse osmosis (with high quality feed water) and is suitable for preparation of reagents and for use in sensitive and analytical procedures.
- pH for HiMedia formulations are measured at 25°C while pH for formulations from pharmacopoeia's are measured after sterilization, as recommended in respective pharmacopoeias. As a part of harmonization of different pharmacopoeias, pH can also be measured after sterilization at 25°C

For more technical details like sterilization and cultural response refer to "The HiMedia Manual" for HiMedia products and pharmacopoeia for respective products
The parameters mentioned for various culture media are subject to change, as per the revision, if any, in the updated pharmacopoeia.

AQUINO

REHYDROL - Ⓑ

"A Perfect isotonic solution to keep your bacteria viable, healthy and active for 3 weeks."

Ready™ PREPARED HIMEDIA

REHYDRATION FLUID FOR GPT

Product Code : LQ254IX

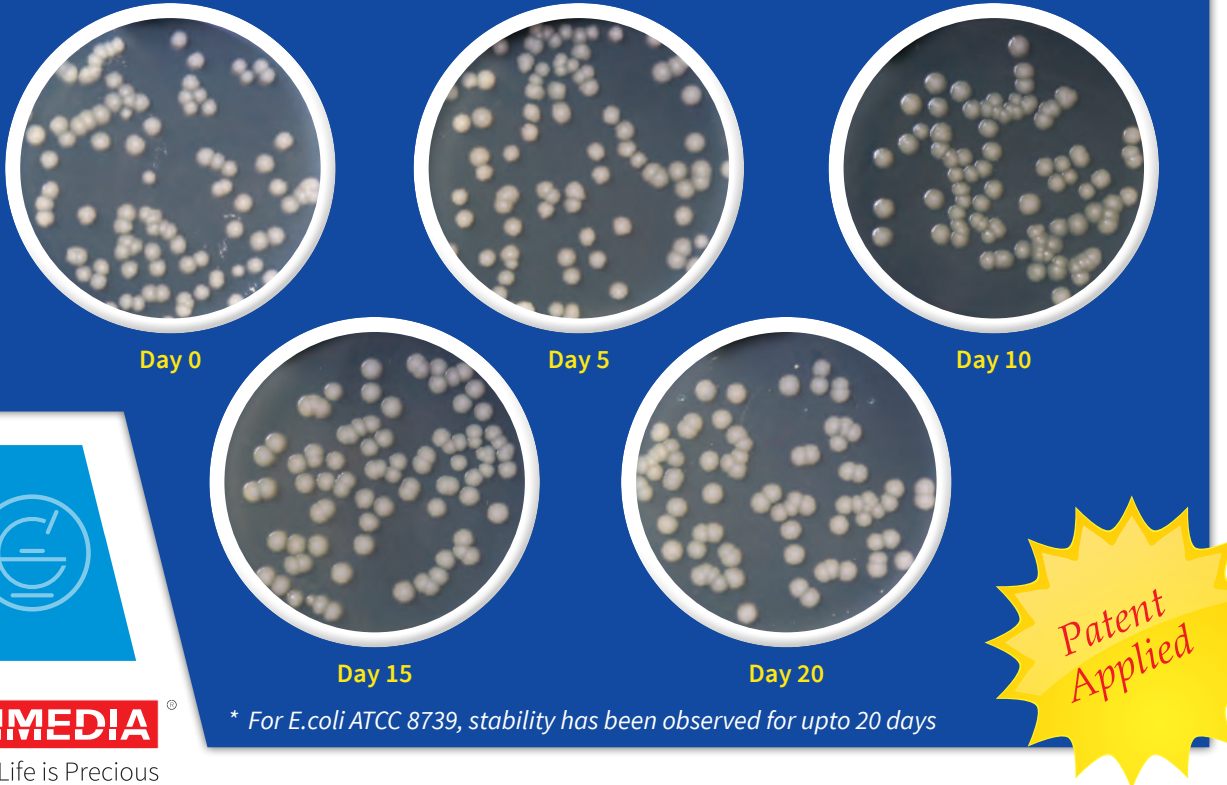
Rehydration fluid for GPT is recommended as a diluting fluid for performing Growth Promotion Test with enhanced stability

FREEDOM FROM TEDIUM

- Growth Promotion Tests (GPT), described in various pharmacopoeias (USP, BP, EP, JP, IP) is carried out to determine suitability of test medium for growth of specified microorganisms
- Dilution methods for GPT are tedious while commercial CFU preparations are expensive
- Culture preparations in this fluid can be stored upto 20 days
- Reduces overall time taken for inoculum preparation
- Overcomes tedious dilutions preparations
- Cost effective

The basic requirements for the GPT are as follows :

1. The new batch of medium must be inoculated with a small number of micro-organisms i.e. 10-100 CFU/0.1ml.
2. The laboratory should test the medium with the microorganisms required by the pharmacopoeias.
3. The microorganisms must not be more than five passages removed from reference culture.



** For E.coli ATCC 8739, stability has been observed for upto 20 days*

Patent Applied

HIMEDIA®

For Life is Precious

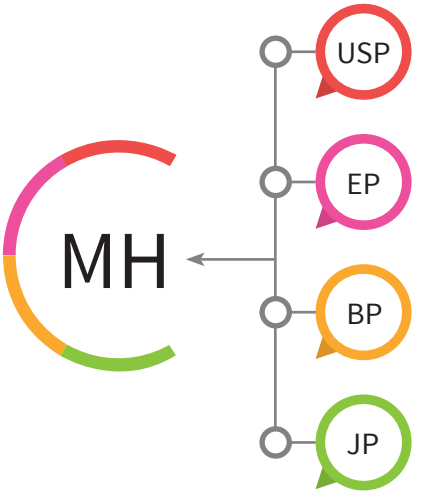
Harmonized Media

USP/EP/BP/JP Compliance

The goal of global pharmacopoeial harmonisation is to promote the acceptance of consistency and uniformity of microbiology methods used by companies throughout the world. HiMedia has incorporated quality control testing methods and specifications according to the new harmonized methods. With the Harmonization of USP/EP/BP/JP/IP newer testing protocols are to be implemented for testing of non sterile drug products. This also includes changes in composition of culture media used to test these products for specified organisms. The Indian Pharmacopoeia Commission (IPC) has initiated plans to bring in compliance with the Global Standards of Harmonization. The IPC has already formulated the composition of eleven media as per the harmonized methods. The eleven media are Buffered Sodium chloride Peptone Solution pH 7.0, Soybean Casein Digest Broth (Casein Soyabean Digest Broth), Soybean Casein Digest Agar (Casein Soyabean Digest Agar), Enterobacteria Enrichment Broth Mossel, Violet Red Bile Glucose Agar, Rappaport Vassiliadis Salmonella Enrichment Broth, Xylose Lysine Deoxycholate Agar, Reinforced Medium for Clostridia, Mannitol Salt Agar, Columbia Agar and Cetrimide Agar.

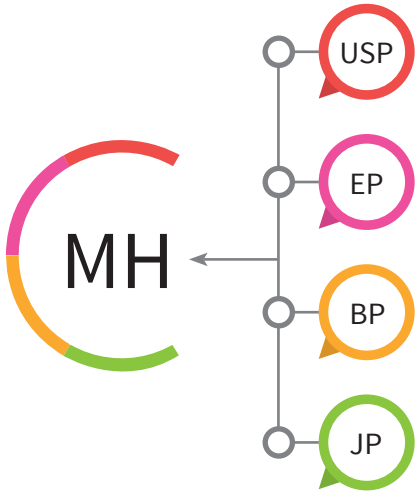
Culture Media as per Harmonized Methods

To meet the consistency in microbiological testing methods, Himedia has formulated the dehydrated culture media in accordance with the Harmonized methods and the same have been made available under 'MH' Codes. The following table contains the detailed description of the Harmonized Testing Methods:



Organism / Test	Recommended Culture Media as per Harmonized Methods	HiMedia's Equivalent Dehydrated Media (Codes)	Equivalent Ready Prepared Media Codes
Buffers and Solutions	Buffered Sodium chloride Peptone Solution pH 7.0	Buffered Sodium chloride Peptone Solution pH 7.0 **(MH1275)**	Buffered Sodium chloride Peptone Solution pH 7.0 **(LQ123)**
Test for *Pseudomonas aeruginosa*	Cetrimide Agar	Cetrimide Agar **(MH024)**	Cetrimide Agar Plate **(MPH024, SPH024G)**
Test for Clostridia	Columbia Agar	Columbia Agar **(MH144)**	Columbia Agar Plate **(MPH144, SPH144G)**
	Reinforced Medium for Clostridia	Reinforced Medium for Clostridia **(MH443)**	Reinforced Medium for Clostridia **(LQ130)**
Test for Bile Tolerant Gram- negative bacteria	Enterobacteria Enrichment Broth Mossel	Enterobacteria Enrichment Broth Mossel **(MH287)**	Enterobacteria Enrichment Broth Mossel **(LQ119)**
	Violet Red Bile Glucose Agar	Violet Red Bile Glucose Agar **(MH581)**	Violet Red Bile Glucose Agar Plate **(MPH581, SPH581)**
Test for *E. coli*	MacConkey Broth	MacConkey Broth **(MH083)**	MacConkey Broth **(LQ115)**
	MacConkey Agar	MacConkey Agar **(MH081)**	MacConkey Agar Plate **(MPH081, SPH081)**
Test for *Staphylococcus aureus*	Mannitol Salt Agar	Mannitol Salt Agar **(MH118)**	Mannitol Salt Agar Plate **(MPH118, SPH118)**
Test for *Candida albicans*	Sabouraud Dextrose Broth	Sabouraud Dextrose Broth **(MH033)**	Sabouraud Dextrose Broth **(LQ120)**
	Sabouraud Dextrose Agar	Sabouraud Dextrose Agar **(MH063)**	Sabouraud Dextrose Agar Plate **(MPH063, SPH063G)**

Culture Media as per Harmonized Methods



Organism / Test	Recommended Culture Media as per Harmonized Methods	HiMedia's Equivalent Dehydrated Media (Codes)	Equivalent Ready Prepared Media Codes
Test for *Salmonella* species	Rappaport Vassiliadis Salmonella Enrichment Broth	Rappaport Vassiliadis Salmonella Enrichment Broth **(MH1491)**	Rappaport Vassiliadis Salmonella Enrichment **(LQ104, LQ104I)**
	Xylose Lysine Deoxycholate Agar	Xylose Lysine Deoxycholate Agar **(MH031)**	Xylose Lysine Deoxycholate Agar Plate **(MPH031, SPH031)**
Microbial enumeration test	Soybean Casein Digest Broth	Soybean Casein Digest Broth (Casein Soyabean Digest Broth) **(MH011)**	Soybean Casein Digest Broth **(LQ027)**
	Soybean Casein Digest Agar	Soybean Casein Digest Agar (Casein Soyabean Digest Agar) **(MH290)**	Soybean Casein Digest Agar Plate **(MPH290, SPH290G)**
	Potato Dextrose Agar	Potato Dextrose Agar **(MH096)**	Potato Dextrose Agar Plate **(MPH096, SPH096)**
	Sabouraud Dextrose Agar	Sabouraud Dextrose Agar **(MH063)**	Sabouraud Dextrose Agar Plate **(MPH063, SPH063G)**
Sterility Medium	Soybean Casein Digest Broth	Soybean Casein Digest Broth (Casein Soyabean Digest Broth) **(MH011)**	Soybean Casein Digest Broth **(LQ027)**

From year 2012, MU, ME, MM, M....B codes of the above media will be available as Harmonized media under MH... code.

'MP' - Ready to use prepared medium in 90 mm plate
'SP' - Ready to use prepared medium in Scored plate
'LQ' - Ready to use Liquid medium in tubes / bottles

Buffered Sodium Chloride-Peptone Solution, pH 7.0 M1275

Intended Use:

Buffered Peptone Water (MH1275 / GMH1275) is recommended as a diluent for carrying out microbial limit testing by harmonized methodology of pharmaceutical products in accordance with USP/EP/BP/JP/IP.

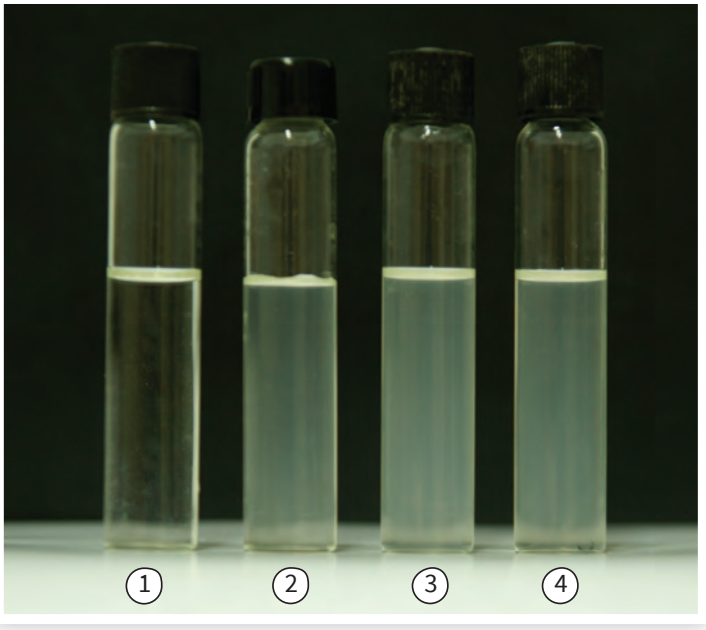
Also recommended as a diluent for carrying microbial limit test from clinical and non clinical specimens .

Directions:

Suspend dehydrated medium as per table in 1000ml of purified /distilled water. Heat if necessary to dissolve the medium completely. In case of M1275, GM1275, MV1275 and MCD1275 add 0.1 to 1% w/v polysorbate 20 or 80 if desired. In case of MH1275 and GMH1275, add 0.1% w/v Polysorbate 80 to assist the suspension, of poorly wettable substances for preparation of nonfatty products insoluble in water. Dispense in tubes or flasks and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M1275	GM1275	MH1275	GMH1275	MV1275	MCD1275
HMC Peptone#	–	–	1.00	1.00	–	–
Peptone	1.00	1.00	–	–	–	–
HiVeg™ peptone	–	–	–	–	1.00	–
HiCynth™ Peptone No.1##	–	–	–	–	–	1.00
Sodium chloride	4.30	4.30	4.30	4.30	4.30	4.30
Disodium hydrogen phosphate, dihydrate	–	–	7.20	7.20	–	–
Disodium hydrogen phosphate	7.23	7.23	–	–	7.23	7.23
Potassium dihydrogen phosphate	3.56	3.56	3.60	3.60	3.56	3.56
Grams/litre	16.09	16.09	14.64	14.64	16.09	16.09
Final pH (at 25°C)	7.0± 0.2	7.0± 0.2	7.00	7.00	7.0± 0.2	7.0± 0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

#Peptone (meat or casein)
##Chemically defined peptone



MH1275 Buffered Sodium chloride-Peptone solution pH 7.0

1. Control
2. *Bacillus subtilis* subsp. *spizizenii* ATCC 6633 (00003*)
3. *Escherichia coli* ATCC 8739 (00012*)
4. *Salmonella* Typhimurium ATCC 14028 (00031*)

*= corresponding WDCM nos.

Buffered Sodium Chloride-Peptone Solution, pH 7.0 M1275

Principle And Interpretation

The composition of MH1275 / GMH1275 medium is as per harmonized methodology of USP/EP/BP/JP/IP (1, 2, 3, 4, 5). This medium is recommended for preparation of stable test strain suspension employed for validating the microbiological testing procedures of non-sterile products. The standardized stable suspensions are used so that the suitability of this test to detect microorganism in presence of product can be established. Non-fatty products insoluble in water and water-soluble products are diluted/dissolved using this solution.

HMC Peptone, Peptone, HiVeg™ peptone, HiCynth™ peptone No. 1 serves as nutrient source and maintains the cell viability. Phosphates in the medium act as good buffering agents. Sodium chloride maintains the osmotic balance and cell integrity. Polysorbates reduce surface tension and also inactivate phenolic compound, if present in the test sample.

Edel and Kampelmacher (7) noted that sub lethal injury to *Salmonella*e might occur in many food preservation processes. Pre-enrichment in Buffered Sodium chloride Peptone solution pH 7.0 at 35°C for 18-24 hours results in repair of injured cells (6). This medium supports the repair of injured cells that have sensitivity to low pH. It is also recommended for pre-enrichment and repair of injured cells (6).

Type of specimen

Pharmaceutical samples;

Clinical samples: pathological specimens, Food and dairy samples; Water samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (10, 11, 12).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (6).

For pharmaceutical samples, follow appropriate techniques for sample collection processing as per pharmacopoeia (1,2, 3, 4, 5).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. The medium contains low nutrients and hence is not recommended for the growth of organisms.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

White to cream homogeneous free flowing powder

GM1275 / GMH1275 : White to cream granular media

Colour and Clarity of prepared medium

Colourless clear solution without any precipitate

pH

M1275 / GM1275 / MV1275 / MCD1275 : 7.00 ± 0.2

MH1275 / GMH1275 - 7.00

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP.

Cultural response

Cultural characteristics observed after recovery on Soybean Casein Digest Agar after an incubation at 30-35°C for the specified time for bacteria and Sabouraud Dextrose Agar at 30-35°C for fungi.

Cultural response

Organism (ATCC)	Inoculum (CFU)	Recovery within 2 hours of incubation	Recovery within 4 hours of incubation	Recovery within 24 hours of incubation
Preparation of test strain				
Escherichia coli 8739 (00012*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Escherichia coli 25922 (00013*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Escherichia coli NCTC 9002	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Pseudomonas aeruginosa 9027 (00026*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Pseudomonas aeruginosa 27853 (00025*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Salmonella Typhimurium 14028 (00031*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Salmonella Abony NCTC 6017 (00029*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Micrococcus luteus 9341	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Candida albicans 10231 (00054*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)
Candida albicans 2091 (00055*)	50 -100	no decrease in colony count	no decrease in colony count	no decrease in colony count (stored at 2-8°C)

Key: * Corresponding WDCM numbers

Buffered Sodium Chloride-Peptone Solution, pH 7.0 M1275

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. The United States Pharmacopoeia, 2019, The United States Pharmacopoeia Convention. Rockville, MD.
2. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
3. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
4. Japanese Pharmacopoeia, 2016.
5. Indian Pharmacopoeia, 2018, Govt. of India, Ministry of Health & Family Welfare, New Delhi, India.
6. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
7. Edel W. & Kampelmacher E.H.,1973, Bull, Wld. Hlth. Org., 48:167.
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
10. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
11. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
12. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Media			
LQ123 LQ123X LQ123XX LQ123C LQ123L LQ123D	Buffered Sodium Chloride Peptone Solution pH 7.0	for the preparation of test suspension in accordance with harmonized methods of USP, EP, BP, JP & IP.	25X9ml / 50X9ml 25X10ml / 50X10ml 25X20ml 10X100ml 5X300ml 5X500ml

Soyabean Casein Digest Medium (Casein Soyabean Digest Broth) M011

Intended Use:

Soybean Casein Digest Medium is a general-purpose medium used for cultivation of a wide variety of microorganisms and for sterility testing of moulds and lower bacteria in accordance with the harmonized method of USP/EP/BP/JP/IP (Medium 1).

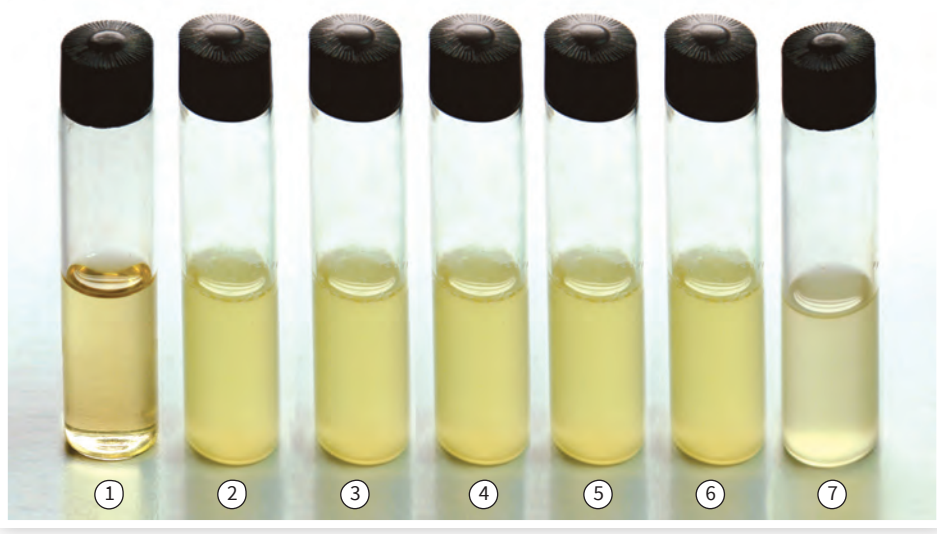
Directions:

Suspend dehydrated medium as per table in 1000 ml purified/ distilled water. Heat if necessary to dissolve the medium completely. Mix well and dispense in tubes or flasks as desired. Sterilize by autoclaving as specified below.

Note: If any fibres are observed in the solution, it is recommended to filter the solution by using a 0.22 micron filter to eliminate the possibilty of presence of fibres.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M011	GM011	MH011	GMH011	MV011	MCD011
Tryptone#	17.00	17.00	17.00	17.00	_	_
HiVeg™ hydrolysate	_	_	_	_	17.00	_
HiCynth™ Peptone No.3###	_	_	_	_	_	17.00
HiCynth™ Peptone No.5###	_	_	_	_	_	3.00
Soya peptone##	3.00	3.00	3.00	3.00	3.00	_
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00
Dextrose (Glucose)	2.50	2.50	_	_	2.50	2.50
Glucose monohydrate	_	_	2.50	2.50	_	_
Dipotassium hydrogen phosphate	2.50	2.50	2.50	2.50	2.50	2.50
Grams/litre	30.00	30.00	29.77	29.77	30.00	30.00
Final pH (at 25°C)	7.3± 0.2	7.3± 0.2	_	_	7.3± 0.2	7.3± 0.2
pH after sterilization (at 25°C)	_	_	7.3± 0.2	7.3± 0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer sterilization, at 25°C
#Pancreatic digest of casein
##Papaic digest of soyabean meal/soyabean
Chemically defined peptones



Soyabean Casein Digest Medium (MH011)

1. Control
2. *Staphylococcus aureus* subsp. *aureus* ATCC 6538 (00032*)
3. *Pseudomonas aeruginosa* ATCC 9027 (00026*)
4. *Candida albicans* ATCC 10231 (00054*)
5. *Bacillus subtilis* subsp. *spizizenii* ATCC 6633 (00003*)
6. *Escherichia coli* ATCC 8739 (00012*)
7. *Salmonella* Typhimurium ATCC 14028 (00031*)

*= corresponding WDCM nos.

Soyabean Casein Digest Medium (Casein Soyabean Digest Broth) M011

Principle And Interpretation

Soyabean Casein Digest Medium is recommended by various pharmacopeias as a sterility testing medium. The media formulation is in accordance with the harmonized methodology of USP/EP/BP/JP/IP (1, 2, 3, 4, 5). It is used for the sensitivity testing of antimicrobial agents by the tube dilution method (6). It is also employed in diagnostic research in microbiology. This medium is used as an diluent and suspending medium or preparation of samples or test strains. It is also employed in sample preparation for testing products, wherein incubation is carried out, only to serve sufficient resuscitation of the cell, while avoiding multiplication of the organism.

The combination of tryptone / HiVeg™ hydrolysate / HiCynth™ peptone No. 3, HiCynth™ peptone No. 5 and soya peptone makes this medium nutritious by providing nitrogenous and carbonaceous compounds, amino acids and long chain peptides for the growth of microorganisms. Natural sugars in soyabean promote growth of fastidious organism. Glucose/dextrose is the fermentable source of carbon and dipotassium hydrogen phosphate serves as the buffer in the medium. Sodium chloride maintains the osmotic balance of the medium.

This medium is recommended by various Pharmacopoeia for sterility checking and for studying total aerobic microbial count in verification of microbiological testing procedures employed for sterility checking.

Type of specimen

Pharmaceutical samples.

Environmental samples, Clinical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 2, 3, 4, 5).

For Clinical samples, follow appropriate techniques for handling specimens as per established guidelines (7, 8).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Biochemical characterisation is necessary to be performed on colonies from pure cultures for further identification.
2. This medium is general purpose and may not support the growth of fastidious organisms.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM011 / GMH011 : Cream to yellow granular media

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate.

pH

7.30 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP.

Stability test

Light yellow coloured clear solution without any precipitation or sedimentation at room temperature for 7 days.

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating not more than 100 CFU (at 30-35°C for 18-24 hours).

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Incubation temperature	Incubation period
Growth promoting				
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Escherichia coli 8739 (00012*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Escherichia coli 25922 (00013*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Escherichia coli NCTC 9002	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Micrococcus luteus 9341	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Salmonella Abony NCTC 6017 (00012*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Streptococcus pneumoniae 6305	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Sterility Testing- Growth promotion+Validation				
The medium is tested with suitable strains of micro-organisms inoculating ≤ 100 CFU and Incubating at 20-25°C for not more than 3 days in case of bacteria and not more than 5 days in case of fungi.				
Staphylococcus aureus subsp. aureus 6538 (00032*)	50 -100	luxuriant	20 -25 °C	≤3 d
Staphylococcus aureus subsp. aureus 25923 (00034*)	50 -100	luxuriant	20 -25 °C	≤3 d
Escherichia coli 8739 (00012*)	50 -100	luxuriant	20 -25 °C	≤3 d
Escherichia coli 25922 (00013*)	50 -100	luxuriant	20 -25 °C	≤3 d
Escherichia coli NCTC 9002	50 -100	luxuriant	20 -25 °C	≤3 d
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	20 -25 °C	≤3 d
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	20 -25 °C	≤3 d
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	20 -25 °C	≤3 d
Micrococcus luteus 9341	50 -100	luxuriant	20 -25 °C	≤3 d
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	20 -25 °C	≤3 d
Salmonella Abony NCTC 6017 (00012*)	50 -100	luxuriant	20 -25 °C	≤3 d
Streptococcus pneumoniae 6305	50 -100	luxuriant	20 -25 °C	≤3 d
Candida albicans 10231 (00054*)	50 -100	luxuriant	20 -25 °C	≤3 d
Candida albicans 2091 (00055*)	50 -100	luxuriant	20 -25 °C	≤3 d
#Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant	20 -25 °C	≤3 d

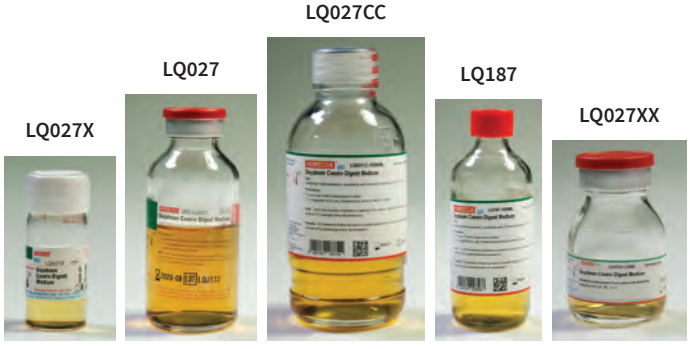
Key: # Formerly known as *Aspergillus niger*
* Corresponding WDCM numbers

Soyabean Casein Digest Medium (Casein Soyabean Digest Broth) M011

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.



Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7, 8).

Reference

1. The United States Pharmacopoeia, 2019, The United States Pharmacopoeial Convention. Rockville, MD.
2. European Pharmacopoeia, 2017 European Dept. for the quality of Medicines.
3. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
4. Japanese Pharmacopoeia, 2016.
5. Indian Pharmacopoeia, 2018, Govt. of India, the controller of Publication, Delhi, India.
6. Wright and Welch, 1959-60, Antibiotics Ann., 61.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook, 2nd Edition.
8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Media			
LQ027 LQ027CC LQ027D LQ027CV LQ027DW LQ187 LQ243 LQ027IX LQ027X LQ027XX	Soyabean Casein Digest Medium Soyabean Casein Digest Medium-Double Packed Soyabean Casein Digest Medium Soyabean Casein Digest Medium (100ml in 125 ml glass bottle) Soyabean Casein Digest Medium	sterility test medium prepared in accordance with harmonized methods of USP, EP, BP, JP, IP.	10x100ml 5x200ml 5x500ml 10x100ml 10X100ml 5X90ml 10x100ml 20X9ml/100X9ml 25X10ml/ 50X10ml 50X20ml
LQ024	Sterility Kit I	sterility test media prepared in accordance with IP/USP/EP/BP/ JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ026 - Sterile Fluid Thioglycollate Medium and LQ027 - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5kt / 20kt
LQ024A	Sterility Kit-I	sterility test media prepared in accordance with IP/USP/EP/BP/ JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ026A - Sterile Fluid Thioglycollate Medium and LQ027A - Sterile Soyabean Casein Digest Medium. Recommended for all purposes	5kt / 20kt
LQ024S	Sterility Kit I	sterility test media prepared in accordance with IP/USP/EP/BP/ JP. One Kit contains 2 glass bottles of 50 ml following media : LQ026S - Sterile Fluid Thioglycollate Medium and LQ027S - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5kt / 20kt
LQ025	Sterility Kit - II	sterility test media prepared in accordance with IP/USP/EP/BP/ JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ028 - Sterile Alternative Thioglycollate Medium and LQ027 - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5kt / 20kt
LQ025A	Sterility Kit - II	sterility test media prepared in accordance with IP/USP/EP/BP/ JP. One Kit contains 2 glass bottles each of 100 ml following media LQ028A - Sterile Alternative Thioglycollate Medium and LQ027A - Sterile Soyabean Casein Digest Medium . Recommended for all purposes	5kt / 20kt

Soybean-Casein Digest Agar (Casein-Soyabean Digest Agar) M290

Intended Use:

Soybean Casein Digest Agar (MH290 / GMH290) is used as a general purpose medium used for cultivation of a wide variety of microorganisms from pharmaceutical products in accordance with harmonized method of USP/EP/BP/JP/IP (Medium 2).

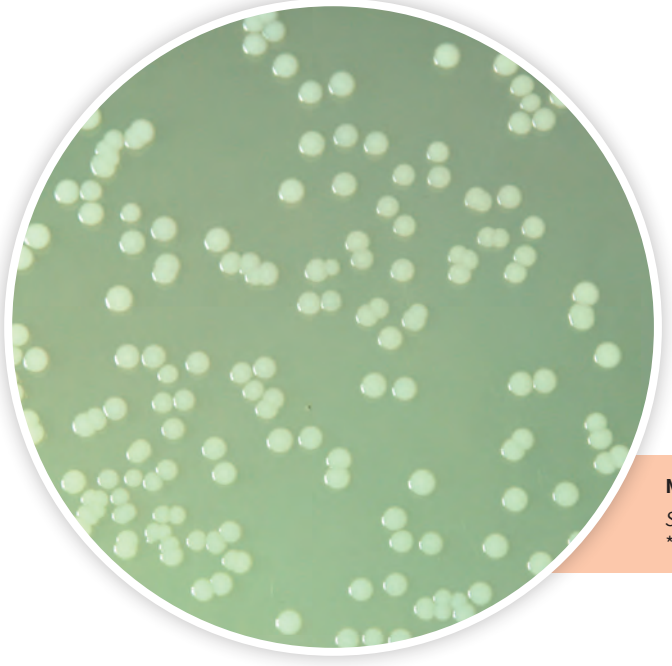
It is a general purpose medium used for cultivation of a wide variety of microorganisms and for sterility testing in pharmaceutical procedures.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. If desired, aseptically add 5% v/v defibrinated blood in previously cooled medium to 45-50°C for cultivation. Cool to 45-50°C.Mix well and pour into sterile Petri plates or as desired.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M290	GM290	MH290	GMH290	MV290	MCD290
Tryptone#	15.00	15.00	15.00	15.00	_	_
HiVeg™ hydrolysate	_	_	_	_	15.00	_
Soya peptone##	5.00	5.00	5.00	5.00	5.00	_
HiCynth™ Peptone No.1###	_	_	_	_	_	15.00
HiCynth™ Peptone No.6###	_	_	_	_	_	5.00
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00
Agar	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	40.00	40.00	40.00	40.00	40.00	40.00
Final pH (at 25°C)	7.3± 0.2	7.3± 0.2	_	_	7.3± 0.2	7.3± 0.2
pH after sterilization (at 25°C)	_	_	7.3±0.2	7.3±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer sterilization, at 25°C
#Equivalaent to Pancreatic digest of casein
##Equivalent to Papaic digest of soyabean meal/soyabean
Chemically defined peptones



MH290 Soyabean Casein Digest Agar

Salmonella Typhimurium ATCC 14028 (00031*)
*= corresponding WDCM no.

Soybean-Casein Digest Agar (Casein-Soyabean Digest Agar) M290

Principle And Interpretation

Various pharmacopoeias recommend Soybean Casein Digest Agar as sterility testing medium. It is also used in validation of sterility checking procedure in accordance with the microbial limit testing harmonized methodology of USP/EP/BP/JP/IP (1, 2, 3, 4, 6).

This medium is used in microbial limit test and antimicrobial preservative- effective test. Gunn et al (5) used this medium for the growth of fastidious organisms and study of haemolytic reaction after addition of 5% v/v blood.

The combination of Tryptone, HiVeg™ hydrolysate, HiCynth™ peptone and soya peptone makes these media nutritious by providing nitrogenous, carbonaceous compounds, amino acids and long chain peptides for the growth of microorganisms. Natural sugars of soy enhance growth of microorganism.

Sodium chloride maintains the osmotic balance in the medium. Agar is the solidifying agent.

The total aerobic count is considered to be equal to the number of colony forming units found on this medium, if colonies of fungi are detected on this medium they are counted along with total aerobic count.

Type of specimen

Pharmaceutical samples.

Food and dairy samples, water samples, Environmental samples, Clinical samples - Blood.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (1, 2, 3, 4, 6).

For Food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (9, 10, 11).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (12).

For Clinical samples follow appopriate techniques for handling specimens as per established guidelines (7, 8)

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. This medium is general purpose medium and may not support the growth of fastidious organisms.
2. Biochemical characterisation is necessary to be performed on colonies from pure cultures for further identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM290 / GMH290 : Cream to yellow granular media

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in Petri plates

pH

7.30 ± 0.2

Growth Promotion Test

Growth Promotion was carried out in accordance with the harmonized method of USP/EP/BP/JP/IP, and growth was observed after an incubation at 30-35°C for 18-24 hours. Recovery rate is considered 100% for bacteria growth on Blood Agar and fungus growth on Sabouraud Dextrose Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU (at 30-35°C for 18 hours).

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Incubation period
Growth promoting				
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	≥70%	18 -24 hrs
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	luxuriant	≥70%	18 -24 hrs
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	luxuriant	≥70%	18 -24 hrs
Escherichia coli 25922 (00013*)	50 -100	luxuriant	≥70%	18 -24 hrs
Escherichia coli 8739 (00012*)	50 -100	luxuriant	≥70%	18 -24 hrs
Escherichia coli NCTC 9002	50 -100	luxuriant	≥70%	18 -24 hrs
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	≥70%	18 -24 hrs
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	≥70%	18 -24 hrs
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant	≥70%	18 -24 hrs
Micrococcus luteus 9341	50 -100	luxuriant	≥70%	18 -24 hrs
Streptococcus pneumoniae 6305	50 -100	luxuriant	≥70%	18 -24 hrs
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	≥70%	18 -24 hr
Candida albicans 10231 (00054*)	50 -100	luxuriant	≥70%	≤5 d
Candida albicans 2091 (00055*)	50 -100	luxuriant	≥70%	≤5 d
#*Aspergillus brasiliensis* 16404 (00053*)	50 -100	luxuriant	50-70%	≤5 d

Key: # Formerly known as *Aspergillus niger*
* Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Soybean-Casein Digest Agar (Casein-Soyabean Digest Agar) M290

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7, 8).

Reference

1. The United States Pharmacopoeia, 2019, The United States Pharmacopeial Convention. Rockville, MD.
2. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
3. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
4. Japanese Pharmacopoeia, 2016.
5. Gunn. B. A. et al, 1977, J. Clin. Microbiol., 5(6) : 650
6. The Indian Pharmacopoeia 2018, Govt of India, Ministry of Health and Family Welfare, New Delhi.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
9. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
10. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
11. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
12. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Petri Plate			
MPH290 MPH290GT MPH290V	Soybean Casein Digest Agar Plate Soybean Casein Digest Agar Plate (γ irradiated) (Triple pack) Soybean Casein Digest Agar Plate (High Fill volume)	for the subculture of aerobic organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	20pt / 50pt 20 pt 50pt
MP290 MP290G MP290GT MP290AGT	Soyabean Casein Digest Agar Plate (Tryptone Soya Agar Plate) Soyabean Casein Digest Agar Plate (Tryptone Soya Agar Plate) (γ - irradiated) Soyabean Casein Digest Agar Plate (Tryptone Soya Agar Plate) (γ - irradiated) (Triple Pack) Soyabean Casein Digest Agar Plate w/ 1% Glycerol (γ irradiated) (Triple Pack)	a general purpose medium used for cultivation of a wide variety of microorganisms.	20pt / 50pt 20pt / 50pt 20pt / 50pt 20pt / 50pt
Category : Ready Prepared Media in 55 mm Scored Polystyrene Plates			
SPH290G SPH290GT SPH290GG	Soybean Casein Digest Agar Plate (γ irradiated) Soybean Casein Digest Agar Plate (γ irradiated) (Triple Pack) Soyabean Casein Digest Agar Plate w/ 1% Glycerol (γ-irradiated)	for the subculture of aerobic organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	100plts 100plts 100plts
SP290 SP290GT SP290G SP290A	Soyabean Casein Digest Agar Plate (Tryptone Soya Agar Plate) Soyabean Casein Digest Agar Plate (Tryptone Soya Agar Plate) (γ-irradiated) (Triple Pack) Soyabean Casein Digest Agar Plate (Tryptone Soya Agar Plate (γ-irradiated) Soyabean Casein Digest Agar Plate (Tryptone Soya Agar Plate) (75mm scored Petri plate)	a general purpose medium for cultivation of a wide variety of microorganisms.	100plts 100plts 100plts 100plts
Category : Ready Prepared Solid Media in Glass bottles			
SMH290	Soyabean Casein Digest Agar	for the subculture of aerobic organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	5X100ml
SMH290CCL	Soyabean Casein Digest Agar	for the subculture of aerobic organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	5X250ml
SMH290D	Soyabean Casein Digest Agar	for the subculture of aerobic organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	5X500ml
Category : Ready Prepared Media Strips for Air Sampler System			
PS290	Agar Strip - SD	(TSA-Agar for total count)	10st / 20st / 50st
PS290A	Agar Strip - SP	(TSA-Penase-Agar for total count in Penicillins and semisynthetic Penicillins production area).	20st
Category : Ready Prepared Slant in Glass Tubes			
SL290	Soyabean Casein Digest Agar Slant	a general purpose growth medium used for the cultivation of a wide variety of microorganisms	10sl / 25sl / 50sl

Enterobacteria Enrichment Broth Mossel M287

Intended Use:

Enterobacteria Enrichment Broth, Mossel is used for selective enrichment of *Enterobacteriaceae* from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 5).

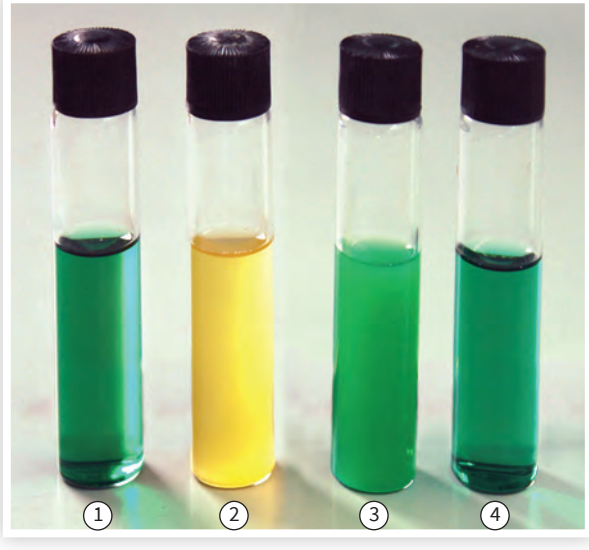
Also used for selective enrichment of *Enterobacteriaceae* in bacteriological examination of foods.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Dispense in tubes or flasks as desired. Stopper with cotton plugs or loose fitting caps. Heat in free flowing steam or boiling water (100°C) for 30 minutes and cool immediately. DO NOT AUTOCLAVE.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™
	M287	GM287	MH287	GMH287	MV287
Gelatin peptone #	_	_	10.00	10.00	_
Peptone	10.00	10.00	_	_	_
HiVeg™ peptone	_	_	_	_	25.00
Glucose monohydrate	_	_	5.00	5.00	
Dextrose (Glucose)	5.00	5.00	_	_	5.00
Dehydrated bile ##	_	_	20.00	20.00	_
Bile, purified###	20.00	20.00	_	_	_
Synthetic detergent No.II	_	_	_	_	5.00
Disodium hydrogen phosphate, dihydrate	_	_	8.00	8.00	_
Disodium hydrogen phosphate	6.45	6.45	_	_	6.45
Potassium dihydrogen phosphate	2.00	2.00	2.00	2.00	2.00
Brilliant green	0.0135	0.0135	0.015	0.015	0.0135
Grams/litre	43.46	43.46	42.93	42.93	43.46
Final pH (at 25°C)	7.2 ± 0.2	7.2 ± 0.2	_	_	7.2 ± 0.2
pH after heating (at 25°C)	_	_	*7.2±0.2	*7.2±0.2	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling	Boiling

*pH can also be measured afer heating at 25°C
#Pancreatic digest of gelatin
##dehydrated ox bile
Ox bile, purified



Enterobacteria Enrichment Broth Mossel (MH287)

1. Control
2. *Escherichia coli* ATCC 8739 (00012*)
3. *Pseudomonas aeruginosa* ATCC 9027 (00026*)
4. *Staphylococcus aureus* subsp. *aureus* ATCC 6538 (00032*)

*= corresponding WDCM nos.

Enterobacteria Enrichment Broth Mossel M287

Principle And Interpretation

The family *Enterobacteriaceae* consists of *Salmonella*, *Shigella* and other enteric pathogens. These organisms find entry into the food system through faecally contaminated water. Majority of these organisms may be eliminated under the stringent food processing parameters. But some of these organisms may become sub lethally injured during the changes in pH, exposure to steam or heat and other unfavourable conditions (1). Therefore the important aspect of food monitoring depends upon the identification and enumeration of these injured cells, after resuscitation. EE Broth, Mossel, formulated by Mossel et al (2) is recommended as an enrichment medium for bile tolerant gram-negative bacteria in the biological examination of foods (2), animal feed stuffs (3). This medium (MH287 / GMH287) is prepared in accordance with the harmonized method of USP/EP/BP/JP/IP (4, 5, 6, 7, 11).

Gelatin peptone, Peptone, HiVeg™ peptone and Glucose monohydrate allows the growth of most of the members of *Enterobacteriaceae*. Brilliant green, Bile purified, Dehydrated bile and Synthetic detergent No. II are the inhibitory agents for gram-positive bacteria. Phosphates act as a good buffering agent and neutralizes acids produced by lactose fermenters that otherwise would adversely affect the growth of the organism. Lactose negative, anaerogenic lactose-positive or late lactose fermenting *Enterobacteriaceae* are often missed by the standard Coli-aerogenes test. To overcome this problem, lactose is replaced by glucose (dextrose) in this medium. Phosphates form the buffering system of the medium. The cells damaged while drying or low pH are resuscitated in well-aerated Soybean Casein Digest Broth (MH011) for 2 hours at 25°C prior to enrichment in EE Broth. The resuscitation procedure is recommended for dried foods (8), animal feeds (9) and semi-preserved foods (10). EE Broth is an enrichment broth and should be used in conjunction with Violet Red Bile Glucose Agar (MH581). A loopful of the enriched sample from EE Broth. is subcultured onto Violet Red Bile Glucose Agar (MH581) after an initial incubation at 30-35°C for 24 hours. Typical pink colonies from MH581 are subcultured for biochemical confirmation by oxidase and fermentation reactions (4). Decimal dilutions of the food homogenate are used if the expected counts are high or else initial suspension is used.

Type of specimen

Pharmaceutical samples; Food samples, Clinical samples.

Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (14).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (4, 5, 6, 7, 11).

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (12, 13).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Some strains may show poor growth due to nutritional variation.
2. Further biochemical identification is recommended to be performed on pure colonies for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to greenish yellow homogeneous free flowing powder

GM287 / GMH287 : Light yellow to greenish yellow granular media

Colour and Clarity of prepared medium

Emerald green coloured, clear solution without any precipitate.

pH

7.20 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP. Cultural response was observed after an incubation at 30-35°C for specified time.

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU (at 30-35°C for ≤24 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temp for not less than longest period of time specified inoculating ≥ 100CFU (at 30-35°C for ≥ 48 hours).

Enterobacteria Enrichment Broth Mossel M287

Cultural Response

Cultural characteristics observed after incubation at 30-35 °C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Acid	Incubation temperature	Incubation period
Growth promoting					
Escherichia coli 8739 (00012*)	50 -100	luxuriant	positive reaction, yellow colour	30 -35 °C	≤24 hrs
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	—	30 -35 °C	≤24 hrs
Inhibitory					
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	≥10^3	inhibited	—	30 -35 °C	≥48 hrs
Additional Microbiological testing					
Escherichia coli 25922 (00013*)	50 -100	luxuriant	positive reaction, yellow colour	30 -35 °C	24 -48 hrs
Escherichia coli NCTC 9002	50 -100	luxuriant	positive reaction, yellow colour	30 -35 °C	24 -48 hrs
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	—	30 -35 °C	24 -48 hrs
#Klebsiella aerogenes 13048 (00175*)	50 -100	luxuriant	positive reaction, yellow colour	30 -35 °C	24 -48 hrs
Proteus mirabilis 25933	50 -100	luxuriant	positive reaction, yellow colour	30 -35 °C	24 -48 hrs
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	positive reaction, yellow colour	30 -35 °C	24 -48 hrs
Shigella boydii 12030	50 -100	luxuriant	negative reaction	30 -35 °C	24 -48 hrs
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	≥10^3	inhibited	—	30 -35 °C	≥48 hrs

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

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (12, 13).

Reference

1. Mossel D. A. A., and Harrewijn G. A., 1972, Alimenta II, 29-30
2. Mossel D. A. A., Vissar M. and Cornellisen A. M. R., 1963, J. Appl.Bacteriol., 26(3):444.
3. Van Schothurst M. et al, 1966, Vet Med., 13(3):273.
4. The United States Pharmacopoeia, 2019, The United States Pharmacopeial Convention. Rockville, MD.
5. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
6. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
7. Japanese Pharmacopoeia, 2016.
8. Mossel D.A.A. and Ratto M.A., 1970, Appl. Microbiol., 20:273.
9. Mossel D.A.A. and Shennan J.L. and Clare V., 1973, J. Sci. Fd. Agric., 24:499.
10. Mossel D.A.A., Ratto M.A., 1973, J. Fd. Technol., 8 : 97.
11. The Indian Pharmacopoeia 2018, Govt. of India,. The Controller of Publication, Delhi
12. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
13. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
14. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Medium in Tubes			
LQ119X	Enterobacteria Enrichment Broth, Mossel	for the enrichment of bile tolerant organisms in accordance with the harmonized methods of USP/BP/EP/JP/IP.	25X10ml / 50X10ml
Category : Ready Prepared Liquid Medium in Glass bottles (for Microbial Limit Test)			
LQ119 LQ119D	Enterobacteria Enrichment Broth Mossel	for the enrichment of bile tolerant organisms in accordance with harmonized methods of USP, EP, BP, JP & IP.	10X100ml 5X500ml

Violet Red Bile Glucose Agar M581

Intended Use:

Violet Red Bile Glucose Agar(MH581 / GMH581) is recommended for detection and enumeration of *Enterobacteriaceae* from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 6).

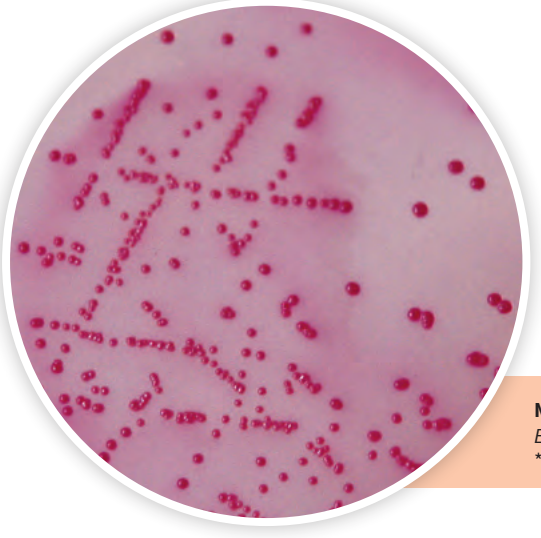
It is also used for detection and enumeration of *Enterobacteriaceae* in raw foods and clinical samples.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified /distilled water. Heat to boiling to dissolve the medium completely. Cool to 45-50°C. Mix well and pour into sterile Petri plates. DO NOT HEAT IN AN AUTOCLAVE.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M581	GM581	MH581	GMH581	MV581	MCD581
Peptone	7.00	7.00	_	_	_	_
Gelatin peptone#	_	_	7.00	7.00	_	_
HiVeg™ peptone	_	_	_	_	7.00	_
HiCynth™ Peptone No.2##	_	_	_	_	_	7.00
HiCynth™ Peptone No.5##	_	_	_	_	_	3.00
Yeast extract	3.00	3.00	3.00	3.00	3.00	_
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00
Bile salts mixture	1.50	1.50	_	_	_	_
Bile salts	_	_	1.50	1.50	_	_
Synthetic detergent No. I	_	_	_	_	1.50	1.50
Glucose (Dextrose)	10.00	10.00	_	_	10.00	10.00
Glucose monohydrate	_	_	10.00	10.00	_	_
Neutral red	0.03	0.03	0.03	0.03	0.03	0.03
Crystal violet	0.002	0.002	0.002	0.002	0.002	0.002
Agar	12.00	12.00	15.00	15.00	12.00	12.00
Grams/litre	38.53	38.53	40.62	40.62	38.53	38.53
Final pH (at 25°C)	7.4± 0.2	7.4± 0.2	_	_	7.4± 0.2	7.4± 0.2
pH after heating (at 25°C)	_	_	*7.4±0.2	*7.4±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling

*pH can also be measured afer heating, at 25°C
#Pancreatic digest of gelatin
##Chemically defined peptones



MH581 Violet Red Bile Glucose Agar
E. coli ATCC 8739 (00012*)
* corresponding WDCM no.

Violet Red Bile Glucose Agar M581

Principle And Interpretation

Violet Red Bile Glucose Agar is a selective medium recommended for detection and enumeration of *Enterobacteriaceae* especially the bile tolerant gram negative bacteria in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (1, 2, 3, 4, 5) from non-sterile products and pharmaceutical preparations.

Peptone, Gelatin peptone, HiVeg™ peptone, HiCynth™ peptones and yeast extract provide nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other nutrients essential for bacterial metabolism. This media is selective due to presence of the inhibitors; bile salts/Synthetic detergent No. 1 and crystal violet. Crystal violet inhibits gram-positive organisms especially Staphylococci. Neutral red indicator helps to detect glucose fermentation. Glucose fermenting strains produce red colonies with pink-red halos in the presence of neutral red. Sodium chloride maintains the osmotic equilibrium in the medium. The red colour is due to absorption of neutral red and a subsequent colour change of the dye when the pH of medium falls below 6.8.

Type of specimen

Pharmaceutical samples, Food samples, Clinical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (6, 7).

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

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (1, 2, 3, 4, 5).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Due to nutritional variations some strains may show poor growth.
2. Further biochemical test must be carried out for confirmation

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pinkish beige homogeneous free flowing powder

GM581 / GMH581 : Light yellow to pinkish beige granular media

Gelling

Firm, comparable with 1.2% Agar gel of M581/GM581/ MV581/ MCD581 and 1.5% Agar gel of MH581/GMH581.

Colour and Clarity of prepared medium

Reddish purple coloured clear to slightly opalescent gel forms in Petri plates.

pH

7.40 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP. Cultural response was observed after an incubation at 30-35°C for 18-24 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU (at 30-35°C for ≤18 hours).

Indicative properties

Colonies are comparable in appearance and indication reaction to those previously obtained with previously tested and approved lot of medium occurs for the specified temperature for a period of time within the range specified inoculating ≤100 CFU (at 30-35°C for 18-24 hours).

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Growth Promoting +\Indicative				
Escherichia coli 8739 (00012*)	50 -100	luxuriant	≥50%	pink-red with bile precipitate
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	≥50%	pink-red
Additional Microbiological Testing				
Escherichia coli NCTC 9002	50 -100	good-luxuriant	≥50%	pink-red with bile precipitate
Escherichia coli 25922 (00013*)	50 -100	good-luxuriant	≥50%	pink-red with bile precipitate
Salmonella Enteritidis 13076 (00030*)	50 -100	good-luxuriant	≥50%	light pink
#Klebsiella aerogenes 13048 (00175*)	50 -100	good-luxuriant	≥50%	light pink
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	$\geq 10^3$	inhibited	0%	
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	$\geq 10^3$	inhibited	0%	

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

Violet Red Bile Glucose Agar M581

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

Reference

1. The United States Pharmacopoeia, 2019, The United States Pharmacopoeial Convention. Rockville, MD.
2. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
3. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
4. Japanese Pharmacopoeia, 2016.
5. Indian Pharmacopoeia, 2018 Ministry of Health and Family Welfare, Govt. of India
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
7. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015), Manual of Clinical Microbiology, 11th Edition. Vol. 1.
8. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media in 90 mm Petri Plate			
MPH581	Violet Red Bile Glucose Agar Plate	for the selection and subculture of bile tolerant organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts
Category : Ready Prepared Media in 55 mm Petri Plate			
SPH581	Violet Red Bile Glucose Agar Plate	for the selection and subculture of bile tolerant organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	100plts
Category : Ready Prepared Media in Polystyrene BiPlates			
HB015	HiCombi™ VRBGA - VRBGA Agar Plate	for the selection and subculture of bile tolerant organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts
Category : Ready Prepared Solid Media in Glass bottles			
SMH581 SMH581D	Violet Red Bile Glucose Agar	for the selection and subculture of bile tolerant organisms in accordance with the harmonized method of USP/EP/BP/JP/IP.	5x100ml 5X500ml

MacConkey Broth M083

Intended Use:

MacConkey Broth (MH083 / GMH083) is used for the selective enrichment of *E.coli* from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 7).

Also is used for presumptive identification of coliforms from variety of specimens such as water, milk and food etc.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Dispense into test tubes with inverted Durham tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	IP	HiVeg™
	M083	GM083	MH083	GMH083	MM083	MV083
Gelatin peptone#	_	_	20.00	20.00	20.00	_
Peptone	20.00	20.00	_	_	_	_
HiVeg™ peptone	_	_	_	_	_	23.00
Lactose monohydrate	_	_	10.00	10.00	_	_
Lactose	10.00	10.00	_	_	10.00	10.00
Dehydrated bile##	_	_	5.00	5.00	5.00	_
Bromo cresol purple	0.01	0.01	0.01	0.01	0.01	0.01
Sodium taurocholate	5.00	5.00	_	_	_	_
Sodium chloride	5.00	5.00	_	_	_	5.00
Synthetic detergent No. V	_	_	_	_	_	2.00
Grams/litre	40.01	40.01	34.51	34.51	35.01	40.01
Final pH (at 25°C)	7.4± 0.2	7.4± 0.2	_	_	_	7.4± 0.2
pH after sterilization (at 25°C)	_	_	*7.3±0.2	*7.3±0.2	*7.3±0.2	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

*pH can also be measured afer heating, at 25°C
#Pancreatic digest of gelatin
##Dehydrated ox bile

MH083 MacConkey Broth
1. Control
2. *Escherichia coli* ATCC 8739 (00012*)
3. *Staphylococcus aureus* subsp. *aureus* ATCC 6538 (00032*)
*= corresponding WDCM nos.

MacConkey Broth M083

Principle And Interpretation

MacConkey Broth is a modification of MacConkey Medium (1). Childs and Allen (2) demonstrated the inhibitory effect of neutral red and therefore substituted it by the less inhibitory bromocresol purple dye. BCP is more sensitive in recording pH variation in the medium. This medium is prepared in accordance with the harmonized method of USP/EP/BP/JP/IP (3, 4, 5, 6, 7)

Gelatin peptone, Peptone and HiVeg™ peptone provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential growth nutrients. Lactose is the fermentable carbohydrate. Dehydrated bile/Sodium taurocholate/Synthetic detergent No. V inhibits gram positive organisms. Bromocresol purple is the pH indicator in the medium, which turns yellow under acidic condition.

Lactose fermenting organisms turn the medium yellow due to the acidity produced on lactose fermentation. The colour change of the dye is observed when the pH of the medium falls below 6.8. Lactose non-fermenting organisms like *Salmonella* and *Shigella* do not alter the appearance of the medium.

Transfer homogenate in Soyabean Casein Digest Medium (MH011) containing 1 gm or 1 ml of the preparation to be examined to 100 ml MacConkey Broth Incubation is carried at 43°-45°C for 24-48 hours. For further isolation, subculture on MacConkey Agar (MH081). Growth of red generally non-mucoid colonies, sometimes surrounded by a reddish precipitation zone, indicates pressure of coliforms.

Type of specimen

Pharmaceutical samples, Food and dairy samples, Water samples, Clinical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (10, 11, 12).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(13).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (3, 4, 5, 6, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further isolation has to be carried out on MacConkey Agar for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow with green tinge homogeneous free flowing powder

GM083 / GMH083 : Cream to yellow with green tinge granular media

Colour and Clarity of prepared medium

Purple coloured clear to slightly opalescent solution in tubes.

pH

MH083 / GMH083 / MM083 : 7.10-7.50

M083 / GM083 / MV083 : 7.20 - 7.60

Growth promotion Test

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

Growth promoting properties

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

Inhibitory properties

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

Cultural response

Cultural characteristics observed after an incubation at 42-44°C for 18-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Acid	Gas	Incubation Temperature	Incubation Period
Growth Promoting						
Escherichia coli 8739 (00012*)	50 -100	luxuriant	positive reaction, yellow colour	positive reaction	42-44°C	≤24 hrs
Inhibitory						
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	$\geq 10^3$	inhibited			42 -44 °C	≥48 hrs
Additional Microbiological testing						
Escherichia coli 25922 (00013*)	50-100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
Escherichia coli NCTC 9002	50-100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
#*Klebsiella aerogenes* 13048 (00175*)	50-100	luxuriant	positive reaction, yellow colour	positive reaction	30 -35 °C	18 -24 hrs
Salmonella Choleraesuis 12011	50-100	fair-good	negative reaction	negative reaction	30 -35 °C	18 -24 hrs
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	$\geq 10^3$	inhibited			30 -35 °C	≥48 hrs

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

MacConkey Broth M083

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. MacConkey A. T., 1900, The Lancet, ii: 20.
2. Childs E. and Allen, 1953, J. Hyg: Camb. 51:468-477.
3. The United States Pharmacopoeia, 2019, The United States Pharmacopeial Convention. Rockville, MD.
4. European Pharmacopoeia, 2017, European Dept. for the Quality of Medicines.
5. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
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7. Indian Pharmacopoeia 2018 Govt. of India, Ministry of Health & Family Welfare, New Delhi, India.
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
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11. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
12. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
13. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Medium in Tubes			
LQ115	MacConkey Broth	for the selective enrichment of *Escherichia coli* in accordance with the harmonized methods of USP, EP, BP & JP.	50X10ml
Category : Ready Prepared Liquid Medium in Bottles			
LQ115C LQ115D	MacConkey Broth	for the selective enrichment of *Escherichia coli* in accordance with the harmonized methods of USP, EP, BP & JP.	10X100ml 5X500ml

MacConkey Agar M081

Intended Use:

MacConkey medium (MH081 / GMH081) is recommended for selective isolation and differentiation of *E.coli* and other enteric bacteria from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 8).

Also recommended for the selective isolation and differentiation of coliform organisms and other enteric pathogens from clinical and non-clinical samples.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates. The surface of the medium should be dry when inoculated

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	IP	HiVeg™	Chemically defined
	M081	GM081	MH081	GMH081	MM081	MV081	MCD081
Gelatin peptone#	17.00	17.00	17.00	17.00	17.00	_	_
HMC peptones##	_	_	3.00	3.00	3.00	_	_
Peptone	1.50	1.50	_	_	_	_	_
HiVeg™ peptone	_	_	_	_	_	1.50	_
HiVeg™ peptone No. 2	_	_	_	_	_	17.00	_
HiCynth™ Peptone No.3###	_	_	_	_	_	_	17.00
HiCynth™ Peptone No.5###	_	_	_	_	_	_	3.00
Tryptone	1.50	1.50	_	_	_	_	_
HiVeg™ hydrolysate	_	_	_	_	_	1.50	_
Lactose monohydrate	_	_	10.00	10.00	_	_	_
Lactose	10.00	10.00	_	_	10.00	10.00	10.00
Bile salts	1.50	1.50	1.50	1.50	1.50	_	_
Synthetic detergent	_		_	_	_	1.50	1.50
Neutral red	0.03	0.03	0.03	0.03	0.03	0.03	0.03
Crystal violet	0.001	0.001	0.001	0.001	0.001	0.001	0.001
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Agar	15.00	15.00	13.50	13.50	13.50	15.00	15.00
Grams/litre	51.53	51.53	49.53	49.53	50.03	51.53	51.53
Final pH (at 25°C)	7.1± 0.2	7.1± 0.2	_	_	_	7.1± 0.2	7.1± 0.2
pH after sterilization (at 25°C)	_	_	*7.1±0.2	*7.1±0.2	*7.1±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer heating, at 25°C
#Pancreatic digest of gelatin
##Equivalent to Peptones (Meat & Casein)
###Chemically defined peptones

MH081 MacConkey Agar
E. coli ATCC 8739 (00012*)
*= corresponding WDCM no.

MacConkey Agar M081

Principle And Interpretation

MacConkey Agar is the earliest selective and differential medium for cultivation of coliform organisms (1, 2). Subsequently MacConkey Agar and Broth have been recommended for use in microbiological examination of foodstuffs (3) and for direct plating / inoculation of water samples for coliform counts (4). This medium is also accepted by the Standard Methods for the Examination of Milk and Dairy Products (5). It is recommended in pharmaceutical preparations and is in accordance with the harmonized method of USP/EP/BP/JP (6, 7, 8, 9).

Gelatin peptone, HMC peptone, Peptone, Tryptone, HiVeg™ peptone, HiVeg hydrolysate and HiCynth™ peptone provide the essential nutrients, vitamins and nitrogenous and carbonaceous factors required for growth of microorganisms. Lactose is the fermentable source of carbohydrate. The selective action of this medium is attributed to crystal violet, bile salts and Synthetic detergent which are inhibitory to most species of gram-positive bacteria.

Sodium chloride maintains the osmotic balance in the medium.

After enrichment of *Escherichia coli* in MacConkey Broth (MH083), it is then subcultured on MacConkey Agar. Gram negative bacteria usually grow well on the medium and are differentiated by their ability to ferment lactose. Lactose fermenting strains grow as red or pink and may be surrounded by a zone of acid precipitated bile. The red colour is due to production of acid from lactose, absorption of neutral red and a subsequent colour change of the dye when the pH of medium falls below 6.8. Lactose non-fermenting strains, such as *Shigella* and *Salmonella* are colourless and transparent and typically do not alter appearance of the medium. *Yersinia enterocolitica* may appear as small, non-lactose fermenting colonies after incubation at room temperature.

Type of specimen

Pharmaceutical samples, Clinical samples, Food & Dairy samples, Water samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (11, 12).

For Food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3, 5, 13).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (4).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (6, 7, 8, 9, 10).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. The surface of the medium must be dry before inoculation.
2. Though the medium is recommended for selective isolation, further biochemical identification is recommended of pure colonies for complete identification.
3. Over incubation may result in reversion of lactose fermentors, wherein colourless colonies is observed in inoculum zone.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

GM081 / GMH081 : Light yellow to pink granular media

Gelling

Firm comparable with 1.35% Agar gel (MH081, GMH081, MM081).

Firm comparable with 1.5% Agar gel (M081, GM081, MV081, MCD081).

Colour and Clarity of prepared medium

Red with purplish tinge coloured clear to slightly opalescent gel forms in Petri plates.

pH

7.10 ± 0.2

Cultural Response

Growth Promotion is carried out in accordance with the harmonized method of ICH (USP/EP/BP/JP). Cultural response was observed after an incubation at 30-35°C for 18-72 hours. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU (at 30-35°C for ≤18 hours).

Indicative properties

Colonies are comparable in appearance and indication reaction to those previously obtained with previously tested and approved lot of medium occurs for the specified temperature for a period of time within the range specified inoculating ≤100 CFU (at 30-35°C for 18-72 hours).

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Growth Promoting + Indicative				
Escherichia coli 8739 (00012*)	50 -100	luxuriant	≥50%	pink-red with bile precipitate
Additional Microbiological testing				
Escherichia coli 25922 (00013*)	50-100	luxuriant	≥50%	pink to red with bile precipitate
Escherichia coli NCTC 9002	50-100	luxuriant	≥50%	pink to red with bile precipitate
#*Klebsiella aerogenes* 13048 (00175*)	50-100	luxuriant	≥50%	pink to red
Enterococcus faecalis 29212 (00087*)	50-100	fair to good	30 -40%	colourless to pale pink
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	≥50%	colourless
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	$\geq 10^3$	inhibited	0%	
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	$\geq 10^3$	inhibited	0%	
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	≥50%	colourless
Salmonella Paratyphi A 9150	50 -100	luxuriant	≥50%	colourless
Salmonella Paratyphi B 8759	50 -100	luxuriant	≥50%	colourless
Salmonella Typhi 6539	50 -100	luxuriant	≥50%	colourless
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant	≥50%	colourless
Proteus vulgaris 13315	50 -100	luxuriant	≥50%	colourless
Shigella flexneri 12022 (00126*)	50 -100	fair to good	30 -40%	colourless
Staphylococcus epidermidis 12228 (00036*)	$\geq 10^3$	inhibited	0%	
Corynebacterium diphtheriae type gravis	$\geq 10^3$	inhibited	0%	

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

MacConkey Agar M081

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (11, 12).

Reference

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2. MacConkey, 1905, J. Hyg., 5:333.
3. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
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5. Wehr H M and Frank J H., 2004, Standard Methods for the Examination of Dairy Products, 17th ed., APHA Inc., Washington, D.C.
6. The United States Pharmacopoeia, 2019, The United States Pharmacopeial Convention.Rockville, MD.
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11. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
12. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
13. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Petri Plate			
MPH081 MPH081T	MacConkey Agar Plate MacConkey Agar Plate (Triple Pack)	for selective isolation and differentiation of *E. coli* and other enteric bacteria from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP.	20plts / 50plts 10plts
MP081	MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl Plate	for selective isolation and differentiation of coliform organisms and other enteric pathogens.	20plts / 50plts
Category : Ready Prepared Media in 55 mm Petri Plate			
SPH081	MacConkey Agar Plate	for the selection and subculture of *Escherichia coli* in accordance with the harmonized method of USP/EP/BP/JP.	100plts
Category : Ready Prepared Media in Polystyrene BiPlates			
HB001	HiCombi™ Nutrient - MacConkey Agar Plate	ccombination of Nutrient Agar + MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl recommended for selective isolation and differentiation of coliform and other enteric pathogens	20plts / 50plts
HB003	HiCombi™ CLED - MacConkey Agar Plate	combination of CLED Agar + MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl recommended for isolation and differentiation of urinary pathogens on the basis of lactose fermentation.	20plts / 50plts
HB004	HiCombi™ XLD - MacConkey Agar Plate	combination of XLD Agar + MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl recommended for selective isolation and enumeration of *Salmonella* species and differentiation of enteric pathogens	20plts / 50plts
HB005	HiCombi™ Cetrimide - MacConkey Agar Plate	combination of Cetrimide Agar + MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl recommended for selective isolation of *Pseudomonas* and differentiation of coliform and other enteric pathogens	20plts / 50plts
HB006	HiCombi™ Blood- MacConkey Agar Plate	combination of Blood Agar + MacConkey Agar recommended for isolation and cultivation of fastidious organisms and differentiation of coliforms and other enteric pathogens.	20plts / 50plts
HB007	HiCombi™ MacConkey-Mannitol Salt Agar Plate	ccombination of MacConkey + Mannitol Salt Agar recommended for cultivation and differentiation of enteric bacteria, restricting the swarming of Proteus species along with potentially pathogenic Gram positive organisms especially pathogenic Staphylococci.	20plts / 50plts
HB010	HiCombi™ Chocolate - MacConkey Agar Plate	combination of Chocolate + MacConkey Agar Plate recommended for the isolation and cultivation of fastidious organisms and differentiation of coliforms and other enteric pathogens	20plts / 50plts
HB012	HiCombi™ MacConkey - MacConkey Agar Plate	for selective isolation and differentiation of *E. coli* and other enteric bacteria from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP.	20plts / 50plts
Category : Ready Prepared Solid Media in Glass bottles			
SMH081D	MacConkey Agar	for the selection and subculture of *Escherichia coli* in accordance with the harmonized method of USP/EP/BP/JP.	5X500ml
SM081	MacConkey Agar	for selective isolation and differentiation of coliform organisms and other enteric pathogens.	5X100ml

Mannitol Salt Agar M118

Intended Use:

Mannitol Salt Agar is used for selective isolation of pathogenic Staphylococci from pharmaceutical products in accordance with Microbial Limit Test by harmonized method of USP/EP/BP/JP/IP (Medium 14).

It is used as a selective media for the isolation of pathogenic Staphylococci from clinical and non-clinical samples .

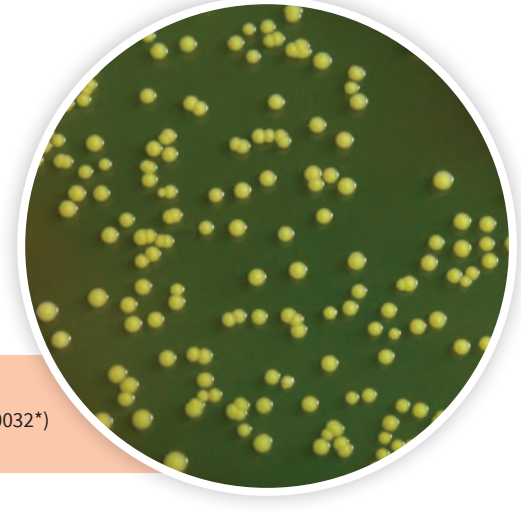
Directions:

Suspend dehydrated medium as per table in 1000ml of purified /distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates. If desired add 5% v/v Egg Yolk Emulsion (FD045) to M118 / MV118.

Note : This product contains 7.5% Sodium chloride as one of its ingredients. On repeated exposure to air and absorption of moisture, sodium chloride has tendency to form lumps, therefore we strongly recommend storage in tightly closed containers in dry place away from bright light.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M118	GM118	MH118	GMH118	MV118	MCD118
Tryptone#	_	_	5.00	5.00	_	_
Proteose peptone	10.00	10.00	_	_	_	_
HM Peptone B##	1.00	1.00	1.00	1.00	_	_
Peptone###	_	_	5.00	5.00	_	_
HiVeg™ peptone No. 3	_	_	_	_	10.00	_
HiCynth™ Peptone No.1####	_	_	_	_	_	10.00
HiCynth™ Peptone No.5####	_	_	_	_	_	1.00
HiVeg™ Extract	_	_	_	_	1.00	_
D-Mannitol	10.00	10.00	10.00	10.00	10.00	10.00
Phenol red	0.025	0.025	0.025	0.025	0.025	0.025
Sodium chloride	75.00	75.00	75.00	75.00	75.00	75.00
Agar	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	111.02	111.02	111.02	111.02	111.02	111.02
Final pH (at 25°C)	7.4± 0.2	7.4± 0.2	_	_	7.4± 0.2	7.4± 0.2
pH after sterilization (at 25°C)	_	_	*7.4±0.2	*7.4±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer heating, at 25°C
#Pancreatic digest of casein
##Equivalent to Beef extract
###Peptic digest of animal tissue
####Chemically defined peptones



MH118 Mannitol Agar
Staphylococcus aureus subsp. *aureus* ATCC 6538 (00032*)
*= corresponding WDCM nos.

Mannitol Salt Agar M118

Principle And Interpretation

Staphylococci are widespread in nature, although they are mainly found on the skin, skin glands and mucous membranes of mammals and birds. The coagulase-positive species i.e *Staphylococcus aureus* subsp. *aureus* is well documented as a human opportunistic pathogen. The ability to clot plasma continues to be the most widely used and accepted criterion for the identification of pathogenic staphylococci associated with acute infections (1). Staphylococci have the unique ability of growing on a high salt containing media (2). Isolation of coagulase-positive staphylococci on Phenol Red Mannitol Agar supplemented with 7.5% NaCl was studied by Chapman (3). The resulting Mannitol Salt Agar Base is recommended for the isolation of coagulase positive staphylococci from cosmetics, milk, food and other specimens (1, 4-7). The additional property of lipase activity of *Staphylococcus aureus* subsp. *aureus* can be detected by the addition of the Egg Yolk Emulsion (FD045). The lipase activity can be visualized as yellow opaque zones around the colonies (8).

It is also used in the performance of microbial limit tests for the selective isolation of *Staphylococcus*. The formulation is in accordance with the harmonization of USP/EP/BP/JP/IP (9, 10, 11, 13, 14).

The medium contains Proteose peptone, HM Peptone B, tryptone, peptone, HiVeg™ peptone, HiVeg™ extract and HiCynth™ peptones which makes it very nutritious as they provide essential growth factors, nitrogenous, carbonaceous compounds, long chain amino acids and trace nutrients. Many other bacteria except Staphylococci are inhibited by 7.5% sodium chloride. Mannitol is the fermentable carbohydrate fermentation of which leads to acid production, detected by phenol red indicator. *S.aureus* ferment mannitol and produce yellow coloured colonies surrounded by yellow zones. Coagulase-negative strains of *S.aureus* are usually mannitol non-fermenters and therefore produce pink to red colonies surrounded by red-purple zones. Presumptive coagulase-positive yellow colonies of *S.aureus* should be confirmed by performing the coagulase test [tube or slide (1)]. Lipase activity of *S.aureus* can be detected by supplementing the medium with egg yolk emulsion. A possible *S.aureus* must be confirmed by the coagulase test. Also the organism should be subcultured to a less inhibitory medium not containing excess salt to avoid the possible interference of salt with coagulase testing or other diagnostic tests (e.g. Nutrient Broth) (M002) (12). Few strains of *S.aureus* may exhibit delayed mannitol fermentation. Negative results should therefore be re-incubated for an additional 24 hours before being discarded (12).

Type of specimen

Pharmaceutical samples, Clinical samples, Food and dairy samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1, 15).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (6, 16, 17).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (9, 10, 11, 13, 14).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Several *Staphylococcus* species other than *S. aureus* are mannitol positive. Therefore, further biochemical tests are necessary for identification of species.
2. Incubation period of 48-72 hours is recommended to detect all *Staphylococcus* species.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder
GM118 / GMH118 : Light yellow to pink granular media

Gelling

Firm,comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

Red coloured clear to slightly opalescent gel forms in Petri plates.

pH

7.20 ± 0.2

Growth Promotion Test

Growth Promotion was carried out in accordance with the harmonized method of USP/EP/BP/JP/IP, after an incubation at 30-35°C for 18-72 hours. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU(at 30-35°C for ≤18 hours).

Indicative properties

Colonies are comparable in appearance and indication reaction to those previously obtained with previously tested and approved lot of medium occurs for the specified temperature for a period of time within the range specified inoculating ≤100CFU (at 30-35°C for 18-72 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temp for not less than longest period of time specified inoculating ≥100CFU (at 30-35°C for ≥ 72 hours).

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Growth Promoting + Indicative				
Staphylococcus aureus subsp. aureus 6538 (00032*)	50 -100	luxuriant	≥50%	yellow/white colonies surrounded by yellow zone
Inhibitory				
Escherichia coli 8739 (00012*)	$\geq 10^3$	inhibited	0%	
Additional Microbiological testing				
Staphylococcus aureus subsp. aureus 25923 (00034*)	50-100	luxuriant	≥50%	yellow/white colonies surrounded by yellow zone

Staphylococcus epidermidis 12228 (00036*)	50-100	fair - good	30 -40%	red
Staphylococcus epidermidis 14990 (00132*)	50-100	fair - good	30 -40%	red
Proteus mirabilis 12453	50-100	none-poor	0 -10%	yellow
Escherichia coli 25922 (00013*)	$\geq 10^3$	inhibited	0%	
Escherichia coli NCTC 9002	$\geq 10^3$	inhibited	0%	
#Klebsiella aerogenes 13048 (00175*)	$\geq 10^3$	inhibited	0%	

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

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (1, 15).

Reference

1. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
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4. Hitchins A. D., Tran T. and McCarron J. E., 1995, FDA Bacteriological Analytical Manual, 8th Ed., AOAC International, Gaithersburg, Md.
5. Davis J. G., 1959, Milk testing, 2nd Ed., Dairy Industries Ltd, London.
6. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
7. Silverton R. E. and Anderson M. J., 1961, Handbook of Medical Laboratory Formulae, Butterworths, London.
8. Gunn B. A., Dunkelberg W. E. and Creitz J. R., 1972, Am. J. Clin. Pathol., 57:236.
9. The United States Pharmacopoeia, 2019, The United States Pharmacopoeial Convention, Rockville, MD.
10. European Pharmacopoeia, 2017, The European Directorate for the Quality of Medicines & HealthCare (EDQM).
11. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia.
12. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore
13. Japanese Pharmacopoeia, 2016
14. Indian Pharmacopoeia, 2018, Govt. of India, Ministry of Health and Family Welfare, New Delhi.
15. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
16. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
17. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Petri Plate			
MPH118 MPH118T	Mannitol Salt Agar Plate Mannitol Salt Agar Plate (Triple Pack)	for selection and subculture of *Staphylococcus aureus* in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts 10plts
MP118	Mannitol Salt Agar Plate	for selective isolation of pathogenic Staphylococci.	20plts / 50plts
Category : Ready Prepared Media in 55 mm Petri Plate			
SPH118	Mannitol Salt Agar Plate	for the selection and subculture of *Staphylococcus aureus* in accordance with the harmonized method of USP/EP/BP/JP/IP.	100plts
Category : Ready Prepared Media in Polystyrene BiPlates			
HB007	HiCombi™ MacConkey-Mannitol Salt Agar Plate	ccombination of MacConkey + Mannitol Salt Agar recommended for cultivation and differentiation of enteric bacteria, restricting the swarming of Proteus species along with potentially pathogenic Gram positive organisms especially pathogenic Staphylococci.	20plts / 50plts
HB009	HiCombi™ Blood -Mannitol Salt Agar Plate	combination of Blood + Mannitol Salt Agar recommended for isolation of *Neiserria* and other fastidious microorganisms along with potentially pathogenic Gram positive organisms especially pathogenic Staphylococci	20plts / 50plts
HB011	HiCombi™ Mannitol Salt - Mannitol Salt Agar Plate	for selection and subculture of *Staphylococcus aureus* in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts
Category : Ready Prepared Solid Media in Glass bottles			
SMH118 SMH118C SMH118D	Mannitol Salt Agar	for the selection and subculture of *Staphylococcus aureus* in accordance with the harmonized method of USP/EP/BP/JP/IP.	5X100ml 10X100ml 5X500ml
Category : Ready Prepared Media Strips for Air Sampler System			
PS118	Agar Strip - MS	(Mannitol-Salt-Agar for Staphylococci)	20strips

Cetrimide Agar M024

Intended Use:

Cetrimide Agar (MH024 / GMH024) is used for the selective isolation of *Pseudomonas aeruginosa* from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 13).

It is also used for the selective isolation of *Pseudomonas aeruginosa* from clinical specimens.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water containing 10 ml glycerol. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M024	GM024	MH024	GMH024	MV024	MCD024
Gelatin peptone#	20.00	20.00	20.00	20.00	_	_
HiVeg™ peptone No. 2	_	_	_	_	20.00	_
HiCynth™ Peptone No.3##	_	_	_	_	_	20.00
Magnesium chloride	1.40	1.40	1.40	1.40	1.40	1.40
Dipotassium sulphate	_	_	10.00	10.00	_	_
Potassium sulphate	10.00	10.00	_	_	10.00	10.00
Cetrimide	0.30	0.30	0.30	0.30	0.30	0.30
Agar	15.00	15.00	13.60	13.60	15.00	15.00
Grams/litre	46.70	46.70	45.30	45.30	46.70	46.70
Final pH (at 25°C)	7.2± 0.2	7.2 ± 0.2	_	_	7.2 ± 0.2	7.2 ± 0.2
pH after sterilization (at 25°C)	_	_	7.2±0.2	7.2±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer heating, at 25°C
#Pancreatic digest of gelatin
##Chemically defined peptone

MH024 Cetrimide Agar
Pseudomonas aeruginosa ATCC 9027 (00026*)
*= corresponding WDCM nos.

Cetrimide Agar M024

Principle And Interpretation

Cetrimide Agar was described by King et al (1). This media formulation is in accordance with the harmonized method of USP/EP/BP/JP/IP (2,3,4,5,7). It is used as a selective medium for the isolation of *Pseudomonas aeruginosa* from pharmaceutical products. This medium is also used for microbial limit testing for non- sterile products. Lowburry first reported the use of cetrimide as an agent for selective isolation of *Pseudomonas* (6). This medium is also used for determining the ability of an organism to produce fluorescein and pyocyanin. Cetrimide (N-acetyl-N,N,Ntrimethylammonium bromide) is incorporated in the medium to inhibit bacteria other than *Pseudomonas aeruginosa*. This compound a cationic detergent acts as a quaternary ammonium compound, which causes nitrogen and phosphorus to be released from bacterial cells other than *Pseudomonas aeruginosa*. Magnesium chloride and potassium sulphate incorporated in the medium enhances the production of pigment pyocyanin, which is a blue-green pigment, diffusing into the medium. This improves detection of *Pseudomonas* on this medium. Presence of magnesium ions can also neutralize EDTA, if present in the sample.

Gelatin peptone, HiVeg™ peptone No. 2 and HiCynth™ peptone provides the essential nutrients for growth of *Pseudomonas*, while glycerin serves as slow and continuous carbon source for the growing cell. For the isolation of *Pseudomonas aeruginosa*, plates of Cetrimide Agar should be inoculated from non-selective medium such as Soybean Casein Digest Medium (MH011). If the count is high the test sample can be directly inoculated onto this medium. *Pseudomonas aeruginosa* colonies may appear pigmented greenish (under UV light also). Addition of nalidixic acid can aid in inhibiting the growth of accompanying flora.

Type of specimen

Pharmaceutical samples, Clinical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (2, 3, 4, 5, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. This medium is a selective medium, some strains may show poor growth as cetrimide is highly toxic.
2. Further biochemical tests must be carried out for further confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM024 / GMH024 : Cream to yellow granular media

Gelling

Firm, comparable with 1.36% Agar gel of MH024 / GMH024

Firm, comparable with 1.5% Agar gel of M024 / GM024 / MV024 / MCD024

Colour and Clarity of prepared medium

Light amber coloured opalescent gel with a slight precipitate forms in Petri plates

pH

7.20 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP. Cultural response was observed after an incubation at 30-35°C for specified time. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU (at 30-35°C for ≤18 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temp for not less than longest period of time specified inoculating ≥100 CFU (at least 100 CFU) (at 30-35°C for ≥ 72 hours).

Cultural Response

Cultural characteristics observed after incubation at 30-35 °C for 18-72 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar

Organism (ATCC)	Inoculum (CFU)	Growth	Incubation period
Growth promoting			
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	≤18 hrs
Inhibitory			
Escherichia coli 8739 (00012*)	≥10^3	inhibited	≥72 hrs
Additional Microbiological testing			
Pseudomonas aeruginosa 27853 (00025*)	50-100	luxuriant	18 -24 hrs
Pseudomonas aeruginosa 25668 (00114*)	50-100	luxuriant	18 -24 hrs
Stenotrophomonas maltophila 13637	≥10^3	inhibited	≥72 hrs
Escherichia coli 25922 (00013*)	≥10^3	inhibited	≥72 hrs
Escherichia coli NCTC 9002	≥10^3	inhibited	≥72 hrs
Staphylococcus aureus subsp. aureus 6538 (00032*)	≥10^3	inhibited	≥72 hrs
Staphylococcus aureus subsp. aureus 25923 (00034*)	≥10^3	inhibited	≥72 hrs
Salmonella Typhimurium 14028 (00031*)	≥10^3	inhibited	≥72 hrs
Proteus mirabilis 29906 (00023*)	≥10^3	inhibited	≥72 hrs

Key: * Corresponding WDCM numbers

Cetrimide Agar M024

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. King, Ward and Raney, 1954, J. Lab. Clin. Med., 44:301.
2. The United States Pharmacopoeia, 2019 The United States Pharmacopoeial Convention. Rockville, MD.
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9. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media in 90 mm Petri Plate			
MP024	Cetrimide Agar Plate	for selective isolation of *Pseudomonas aeruginosa* from clinical specimens.	20plts / 50plts
MPH024 MPH024T MPH024GT	Cetrimide Agar Plate Cetrimide Agar Plate (Triple Pack) Cetrimide Agar Plate (γ irradiated) (Triple pack)	for the selection and subculture of *Pseudomonas aeruginosa* in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts 10plts 20plts / 50plts
Category : Ready Prepared Media in 55 mm Petri Plate			
SPH024G	Cetrimide Agar Plate (γ irradiated)	for the selection and subculture of *Pseudomonas aeruginosa* in accordance with the harmonized method of USP/EP/BP/JP/IP.	100plts
Category : Ready Prepared Media in Polystyrene BiPlates			
HB005	HiCombi™ Cetrimide - MacConkey Agar Plate	combination of Cetrimide Agar + MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl recommended for selective isolation of *Pseudomonas* and differentiation of coliform and other enteric pathogens.	20plts / 50plts
HB013	HiCombi™ Cetrimide - Cetrimide Agar Plate	for the selection and subculture of *Pseudomonas aeruginosa* in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts
Category : Ready Prepared Media HiTouch™ FlexiPlate™			
FL014	HiTouch™ FlexiPlate™ - CT	for enumeration (count) of *Pseudomonas* aeruginosa.	50plts
Category : DriFilter™ Membrane Nutrient Pad Media			
MF007	Cetrimide Medium (without Membrane Filter)	for detection and enumeration of *Pseudomonas*	20plts / 50plts

Rappaport Vassiliadis Salmonella Enrichment Broth M1491

Intended Use:

Rappaport Vassiliadis Salmonella Enrichment Broth (MH1491 and GMH1491) is recommended for selective enrichment of *Salmonella* species from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 9).

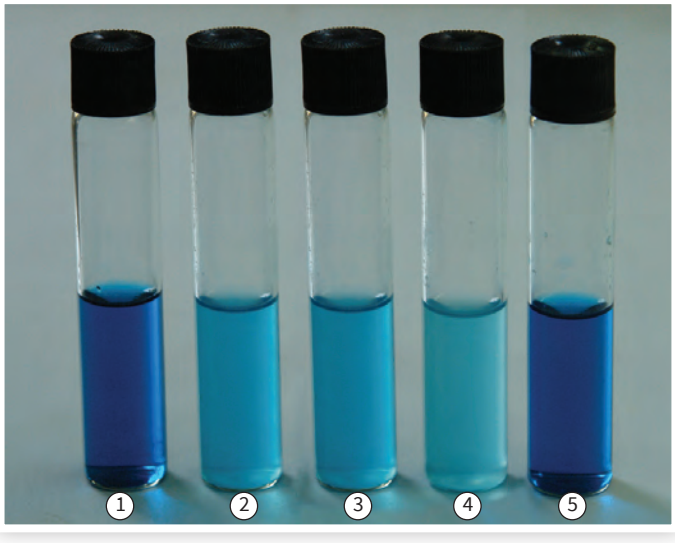
It is recommended as a selective enrichment medium for the *Salmonella*e species from the food and animal feeding stuffs.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified /distilled water. Heat if necessary to dissolve the medium completely. Dispense as desired into tubes and sterilize by autoclaving at 115°C as per validated cycle.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	IP	Chemically defined
	M1491	GM1491	MH1491	GMH1491	MM1491	MCD1491
Soya peptone#	4.50	4.50	4.50	4.50	4.50	_
HiCynth™ peptone No. 4##	_	_	_	_	_	4.50
Sodium chloride	8.00	8.00	8.00	8.00	8.00	8.00
Dipotassium hydrogen phosphate	0.40	0.40	0.40	0.40	0.40	0.40
Potassium dihydrogen phosphate	0.60	0.60	0.60	0.60	0.60	0.60
Magnesium chloride, hexahydrate	29.00	29.00	29.00	29.00	29.00	29.00
Malachite green	0.036	0.036	0.036	0.036	0.036	0.036
Grams/litre	27.11	27.11	27.11	27.11	27.11	27.11
Final pH (at 25°C)	5.2± 0.2	5.2± 0.2	_	_	_	5.2± 0.2
pH after sterilization (at 25°C)	_	_	*5.2±0.2	*5.2±0.2	*5.2±0.2	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 115°C-15 min	Autoclaving 115°C-15 min	Autoclaving 115°C or as per validated cycle	Autoclaving 115°C or as per validated cycle	Autoclaving 115°C-30 min or as per validated cycle	Autoclaving 115°C-15 min

*pH can also be measured afer heating, at 25°C
#Pancreatic digest of soyabean meal
##Chemically defined peptone



MH1491 Rappaport Vassiliadis Salmonella Enrichment Broth
1. Control
2. *Salmonella* Typhimurium ATCC 14028 (00031*)
3. *Salmonella* Abony NCTC 6017 (00029*)
4. *Salmonella* Enteritidis ATCC 13076 (00030*)
5. *Staphylococcus aureus* subsp. *aureus* ATCC 6538 (00032*)

*= corresponding WDCM nos.

Rappaport Vassiliadis Salmonella Enrichment Broth M1491

Principle And Interpretation

Rappaport Vassiliadis Salmonella Enrichment Medium is designed according to the revised formulation by Van Schothorst et al (1) and is recommended for the selective enrichment of *Salmonella*e from pharmaceutical products. This medium can also be used in direct enrichment of samples containing low inoculum. Present medium is a modification of the Rappaport Vassiliadis Enrichment Broth described by Van Schothorst and Renauld (2). It is prepared in accordance with the harmonized methodology of USP/EP/BP/JP (3,4,5,6) has been found to be superior to other *Salmonella* selective medias. This medium is also recommended by IP (7). Addition of magnesium chloride to the medium was reported by Peterz et al. *Salmonella* species can be isolated from human faeces without pre-enrichment by using this medium.

Salmonella generally survive at little high osmotic pressure, grow at slightly low pH and are resistant to malachite green compared to other bacteria. These characteristics are exploited in this medium for selective enrichment of *Salmonella*. Magnesium chloride present in the medium raises the osmotic pressure. Natural sugars of soya peptone provide essential growth nutrients and enhance the growth of *Salmonella* (8). Phosphate buffers the medium to maintain constant pH. Sodium chloride maintains the osmotic balance. Malachite green inhibits many gram-positive bacteria, while selectively enriches *Salmonella*.

The relatively lower concentration of nutrition, also aids selective enrichment of *Salmonella*. This medium was reported to be superior to *Salmonella* selective medium like Tetrathionate Broth and Selenite enrichment broth and to Tetrathionate Brilliant Green Broth for the detection of *Salmonella*e in milk samples. The enriched culture of Rappaport Vasiliadis Salmonella Enrichment Broth (MH1491) can be further subcultured and isolated on Xylose Lysine Deoxycholate Agar (MH031).

Type of specimen

Pharmaceutical samples, Clinical samples, Food samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (10).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (3, 4, 5, 6, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Overheating may destroy the selectivity of medium.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to light blue homogeneous free flowing powder

GM1491 / GMH1491 : Light yellow to light blue granular media

Colour and Clarity of prepared medium

Greenish blue coloured clear to slightly opalescent solution with a slight precipitate in tubes.

pH

5.20 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with harmonized method of USP/BP/EP/JP. Cultural response was observed after an incubation at 30-35°C for specified time. Recovery is carried out using Xylose Lysine Deoxycholate Agar (MH031), after enrichment in Rappaport Vassiliadis Salmonella Enrichment Broth.

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU (at 30-35°C for ≤18 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temperature for not less than longest period of time specified inoculating ≥100 CFU (at least 100 CFU) (at 30-35°C for ≥ 24 hours).

Cultural Response

Organism (ATCC)	Inoculum (CFU)	Growth	Colour of colony	Incubation period
Growth promoting				
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	red with black centers	≤18 hrs
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant	red with black centers	≤18 hrs
Inhibitory				
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	$\geq 10^3$	inhibited		≥24 hrs
Additional Microbiological testing				
Escherichia coli 25922 (00013*)	50-100	none-poor	yellow	18 -24 hrs
Escherichia coli 8739 (00012*)	50-100	none-poor	yellow	18 -24 hrs
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	red with black centre	18 -24 hrs
Salmonella Paratyphi B 8759	50 -100	luxuriant	red with black centre	18 -24 hrs
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	$\geq 10^3$	inhibited		≥24 hrs
Pseudomonas aeruginosa 9027 (00026*)	$\geq 10^3$	inhibited		≥24 hrs
Pseudomonas aeruginosa 27853 (00025*)	$\geq 10^3$	inhibited		≥24 hrs
Enterococcus faecalis 29212 (00087*)	$\geq 10^3$	inhibited		≥24 hrs
***E.coli* +S.Typhimurium (mixed culture)**				
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	red with black centre	18 -72 hrs

Key: * Corresponding WDCM numbers

Rappaport Vassiliadis Salmonella Enrichment Broth M1491

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

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

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Petri Plate			
LQ104 LQ104V LQ104XX LQ104C	Rappaport Vassiliadis Salmonella Enrichment Broth	for selective enrichment of *Salmonella* species from pharmaceutical & clinical sample in accordance with harmonized methods of USP, EP, BP & JP.	12X10ml / 25X10ml / 50X10ml 50X5ml 25X20ml 5X100ml
LQ104I	Rappaport Vassiliadis Salmonella Enrichment Broth (As per IP)	for the selective enrichment of *Salmonella* species in accordance with Indian pharmacopoeia.	25X10ml / 50X10ml

Xylose –Lysine Deoxycholate Agar M031

Intended Use:

Xylose-Lysine Deoxycholate Agar (MH031 / GMH031) is a selective medium recommended for the isolation and enumeration of *Salmonella* Typhi and other *Salmonella* species from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 12).

Also as a selective medium recommended for the isolation and enumeration of *Salmonella* Typhi and other *Salmonella* species from clinical and non clinical samples.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified /distilled water. Heat with frequent agitation until the medium boils. DO NOT HEAT IN AN AUTOCLAVE. Transfer immediately to a water bath at 50°C. After cooling, pour into sterile Petri plates. It is advisable not to prepare large volumes, which will require prolonged heating and may produce precipitate.

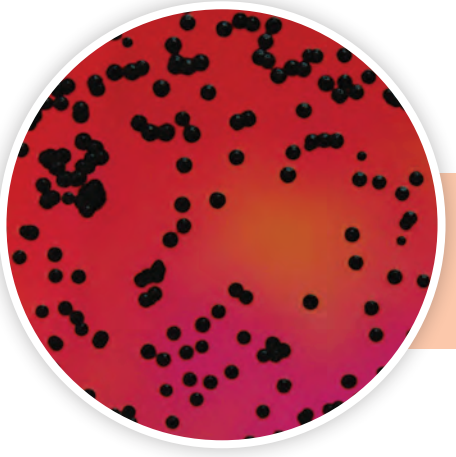
Note: Slight precipitation in the medium may occur, which is inheritant property of the medium, and does not affect the performance of the medium.

Principle And Interpretation

Enterobacteriaceae is a family of gram-negative, non-spore-forming bacilli that contains more than 100 species of bacteria that normally inhabit the intestines of humans and animals. Members forming part of the normal intestinal microflora are referred to as coliforms. The clinically significant genera of *Enterobacteriaceae include Cedecea, Citrobacter, Edwardsiella, Enterobacter, Escherichia, Ewingella, Hafnia, Klebsiella, Kluyvera, Proteus, Salmonella, Shigella* and *Yersinia* (1).

The *Salmonellae* are the most complex of all the *Enterobacteriaceae*. Human *Salmonella* infections are most commonly caused by ingestion of food, water or milk, contaminated by human or animal excreta (2). A large number of media have been developed for the selective isolation and identification of enteric bacilli including *Salmonella*.

Xylose Lysine Deoxycholate Agar is a selective as well as differential medium formulated by Taylor (3-7) for the isolation and identification of enteric pathogens especially *Shigellae* from stool samples. It is also used for pharmaceutical testing



MH031 Xylose-Lysine Deoxycholate Agar
Salmonella Typhimurium ATCC 14028 (00031*)
*Corresponding WDCM no.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M031	GM031	MH031	GMH031	MV031	MCD031
Xylose	3.50	3.50	3.50	3.50	3.50	3.50
HiCynth™ Peptone No.5#	_	_	_	_	_	4.00
Yeast extract	3.00	3.00	3.00	3.00	4.00	_
L-lysine	5.00	5.00	5.00	5.00	5.00	5.00
Lactose monohydrate	_	_	7.50	7.50	_	_
Lactose	7.50	7.50	_	_	7.50	7.50
Sucrose	7.50	7.50	7.50	7.50	7.50	7.50
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00
Sodium deoxycholate	2.50	2.50	2.50	2.50	_	_
Sodium thiosulphate	6.80	6.80	6.80	6.80	6.80	6.80
Ferric ammonium citrate	0.80	0.80	0.80	0.80	0.80	0.80
Phenol red	0.08	0.08	0.08	0.08	0.08	0.08
Synthetic detergent No. III	_	_	_	_	1.50	1.50
Agar	15.00	15.00	13.50	13.50	15.00	15.00
Grams/litre	56.68	56.68	54.80	54.80	56.68	56.68
Final pH (at 25°C)	7.4± 0.2	7.4± 0.2	_	_	7.4± 0.2	7.4± 0.2
pH after sterilization (at 25°C)	_	_	*7.4±0.2	*7.4±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling

*pH can also be measured afer heating, at 25°C
#Chemically defined peptone

and nonsterile product testing for the detection (or absence) of *Salmonella* after enrichment in Rappaport Vassiliadis Salmonella Enrichment Broth (MH1491) in accordance with the harmonized method of USP/EP/BP/JP/IP (8-12).

Deoxycholate, ferric ammonium citrate, sodium thiosulphate and Synthetic detergent No. III are selective agents that inhibit gram-positive microorganisms. Essential nutrients, growth factors for growth of microorganism are provided by yeast extract and HiCynth™ peptone No. 5. Xylose, sucrose and lactose are the fermentable sugars in this medium. Xylose is fermented by almost all the enteric bacteria except *Shigellae*, which enable the differentiation of *Shigellae* from *Salmonellae*. *Salmonellae* metabolize the xylose and decarboxylate lysine and thus change the pH to alkaline and mimic *Shigellae* reaction. However to prevent this reaction by lysine positive coliforms, lactose and sucrose are added in excess to produce acid and hence nonpathogenic H_2S producers do not decarboxylate lysine. Sodium thiosulphate helps in reactivation of sulphur containing compounds and prevents the desication of these compounds during storage. It also forms the substrate for enzyme thiosulphate reductase, which breaks it to form H_2S. Thiosulphate and ferric ammonium citrate are the H_2S indicators in the medium. Sodium thiosulphate is also inactivator of halogens, mercurial and aldehyde and can minimize its toxicity in the testing sample, if any during microbial limit tests. Sodium chloride maintains the osmotic equilibrium in this medium. Phenol red is the pH indicator.

Degradation of fermentable sugars proceed concurrently and generates acids, which cause pH indicator to give various shades of colour, causing a color change in the colonies and in the medium from red to yellow on prolonged incubation. Hydrogen sulfide production results in colonies with black centers under alkaline conditions, which can be inhibited by acid production by carbohydrate fermentation. Alkaline condition causes the color of the medium to change back to red. This medium is an ideal medium for screening samples containing mixed flora of enteric pathogens as recovery of *Salmonellae* and *Shigellae* is not conspicuous by even profuse growth of other species (13, 14).

Type of specimen

Pharmaceutical samples, Clinical samples, Food samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (14, 15).

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (16).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (8, 9, 10, 11, 12).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Though this medium is selective for *Salmonella* other species of *Enterobacteriaceae* may grow.
2. *Salmonella* Typhi and *Shigella* species may not grow on this medium.
3. Moreover *Proteus*, *Pseudomonas* and *Citrobacter* species may mimic enteric pathogens by producing small red colonies.
4. Further confirmation has to be carried out on presumptive *Salmonella* isolates.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder

GM031 / GMH031 : Light yellow to pink granular media

Gelling

Firm, comparable with 1.35% Agar gel of MH031 / GMH031

Firm, comparable with 1.5% Agar gel of M031 / GM031 / MV031 / MCD031

Colour and Clarity of prepared medium

Red coloured clear to slightly opalescent gel forms in Petri plates

pH

7.40 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP. Cultural response was observed after an incubation at 30-35°C for specified time. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU(at 30-35°C for ≤18 hours).

Indicative properties

Colonies are comparable in appearance and indication reaction to those previously obtained with previously tested and approved lot of medium occurs for the specified temperature for a period of time within the range specified inoculating ≤100CFU (at 30-35°C for 18-72 hours).

Inhibitory properties

No growth of the test microorganism occurs for the specified temp for not less than longest period of time specified inoculating ≥100CFU (at 30-35°C for ≥ 72 hours).

Cultural Response

Cultural characteristics observed after incubation at 30-35°C for 18-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony	Incubation period
Growth Promoting + Indicative					
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	≥50%	red with black centre	18 -72 hrs
Salmonella Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	≥50%	red with black centre	18 -72 hrs
Additional Microbiological testing					
Escherichia coli 8739 (00012*)	50 -100	fair	20 -30%	yellow	18 -72 hrs
Escherichia coli 25922 (00013*)	50-100	fair	20 -30%	yellow	18 -72 hrs

Xylose –Lysine Deoxycholate Agar M031

Escherichia coli NCTC 9002	50-100	fair	20 -30%	yellow	18 -72 hrs
Proteus vulgaris 13315	50-100	good-luxuriant	≥50%	grey with black centres	18 -72 hrs
Salmonella Paratyphi A 9150	50 -100	good-luxuriant	≥50%	red	18 -72 hrs
Salmonella Paratyphi B 8759	50-100	good-luxuriant	≥50%	red with black centres	18 -72 hrs
Salmonella Enteritidis 13076 (00030*)	50-100	good-luxuriant	≥50%	red with black centres	18 -72 hrs
Salmonella Typhi 6539	50-100	good-luxuriant	≥50%	red with black centres	18 -72 hrs
Shigella dysenteriae 13313	50 -100	good-luxuriant	≥50%	red	18 -72 hrs
Shigella flexneri 12022 (00126*)	50-100	fair-good	30-40%	red	18 -72 hrs
Shigella sonnei 25931	50-100	fair-good	30-40%	red	18 -72 hrs
#*Klebsiella aerogenes* 13048 (00175*)	50-100	fair	20-30%	yellow	18 -72 hrs
Enterobacter cloacae 13047 (00083*)	50 -100	fair	20-30%	yellow	18 -72 hrs
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	$\geq 10^3$	inhibited	0%		≥72 hrs
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	$\geq 10^3$	inhibited	0%		≥72 hrs
Enterococcus faecalis 29212 (00087*)	$\geq 10^3$	inhibited	0%		≥72 hrs

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

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (14, 15).

Reference

1. Pelczar M. J. Jr., Reid R. D., Chan E. C. S., 1977, Microbiology, 4th Ed., Tata McGraw-Hill Publishing Company Ltd, New Delhi.
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3. Taylor W. L., 1965, Am. J. Clin. Pathol., 44:471-475.
4. Taylor W. L. and Harris B., 1965, Am. J. Clin. Pathol., 44:476.
5. Taylor W. L. and Harris B., 1967, Am. J. Clin. Pathol., 48:350.
6. Taylor W. L. and Schelhart B., 1967, Am. J. Clin. Pathol., 48:356.
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9. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
10. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia.
11. Japanese Pharmacopoeia, 2016.
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13. McCarthy M.D., 1966, N.Z. J. Med. Lab. Technol., 20:127.
14. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
15. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1
16. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Petri Plate			
MP031	Xylose Lysine Deoxycholate Agar (XLD Agar) Plate	for selective isolation and enumeration of *Salmonella* Typhi and other *Salmonella* species.	20plts / 50plts
MPH031	Xylose Lysine Deoxycholate Agar Plate	for the selection and subculture of *Salmonella* in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts
Category : Ready Prepared Media in 55 mm Petri Plate			
SPH031	Xylose Lysine Deoxycholate Agar Plate	for the selection and subculture of *Salmonella* in accordance with the harmonized method of USP/EP/BP/JP/IP.	100plts
Category : Ready Prepared Media in Polystyrene BiPlates			
HB004	HiCombi™ XLD - MacConkey Agar Plate	combination of XLD Agar + MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl recommended for selective isolation and enumeration of *Salmonella* species and differentiation of enteric pathogens	
HB014	HiCombi™ XLD - XLD Agar Plate	for the selection and subculture of *Salmonella* in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts
Category : Ready Prepared Dual Media for B lood Specimens in Glass Bottles			
LQ030	HiCombi™ Dual Performance Salmonella Medium - XLD	combination of XLD Agar + MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl recommended for selective isolation and enumeration of *Salmonella* species and differentiation of enteric pathogens	10 bottles

Reinforced Medium for Clostridia M443

Intended Use:

Reinforced Medium for Clostridia (MH443) is used for the enrichment of *Clostridia* from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 15).

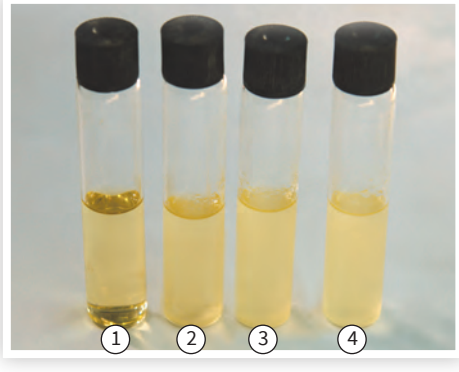
It is used for the cultivation and enumeration of *Clostridia* and other anaerobes.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Dispense into tubes or flask as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M443	MH443	GMH443	MV443	MCD443
Peptone#	_	10.00	10.00	_	_
Tryptone	10.00	_	_	_	_
HM Peptone B###	10.00	10.00	10.00	_	_
HiVeg™ hydrolysate	_	_	_	10.00	_
HiVeg™ extract	_	_	_	10.00	_
Yeast extract	3.00	3.00	3.00	3.00	_
HiCynth™ Peptone No.2##	_	_	_	_	15.00
HiCynth™ Peptone No.5##	_	_	_	_	8.00
Glucose monohydrate	_	5.00	5.00	_	_
Dextrose (Glucose)	5.00	_	_	5.00	5.00
Sodium chloride	5.00	5.00	5.00	5.00	5.00
Soluble starch	1.00	1.00	1.00	1.00	1.00
Cysteine hydrochloride	_	0.50	0.50	_	_
Sodium acetate	3.00	3.00	3.00	3.00	3.00
L-Cysteine hydrochloride	0.50	_	_	0.50	0.50
Agar	0.50	0.50	0.50	0.50	0.50
Grams/litre	38.00	37.54	37.54	38.00	38.00
Final pH (at 25°C)	6.8 ± 0.2	_	_	6.8 ± 0.2	6.8 ± 0.2
pH after sterilization (at 25°C)	_	*6.8±0.2	*6.8±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 115°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 115°C-15 min	Autoclaving 115°C-15 min

*pH can also be measured afer heating, at 25°C
#Peptic digest of animal tissue
Chemically defined peptones
###Equivalent to Beef extract



MH443 Reinforced Medium for Clostridia
1. Control
2. *Clostridium perfringens* ATCC 13124
3. *Clostridium sporogenes* ATCC 19404 (00008*)
4. *Bacteroides vulgatus* ATCC 8482

*Corresponding WDCM no.

Reinforced Medium for Clostridia M443

Principle And Interpretation

Reinforced Medium for Clostridia was formulated by Hirsch and Grinsted (1). This media is prepared in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (2, 3, 4, 5, 8). It is recommended for sterility checking of non-sterile products, nutritional and dietary supplements. It can be used to initiate growth from small inocula and to obtain the highest viable count of clostridia. Barnes and Ingram used the broth medium for diluting an inoculum of vegetative cells of *Clostridium perfringens* (6, 7). It can be used in studies of spore forming anaerobes, especially *Clostridium butyricum* in cheese, for enumeration of Clostridia in tube dilution counts or for preparation of plates for isolation (7). Other spore forming anaerobes, Streptococci and Lactobacilli also grow in these media. These are enriched but non-selective media.

Peptone, Yeast extract, HM Peptone B, Tryptone, HiVeg™ hydrolysate, HiVeg™ extract and HiCynth™ peptones provide nitrogenous and carbonaceous compounds, long chain amino acids, vitamins, minerals and all the necessary nutrients for the growth of clostridia. Glucose / dextrose monohydrate is a fermentable carbohydrate in the medium while sodium chloride maintains osmotic equilibrium. Cysteine hydrochloride acts as reducing agent. Small amount of soluble starch removes toxic metabolites from the medium. Sodium acetate also acts as a good buffering agent. Small quantity of agar keeps the medium semi solid and helps in maintaining anaerobic conditions.

Type of specimen

Pharmaceutical samples, Food samples, Clinical samples.

Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (11).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (2, 3, 4, 5, 8).

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (9, 10).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Due to nutritional variations some strains may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GMH443 : Cream to yellow granular media

Colour and Clarity of prepared medium

Light yellow coloured clear solution in tubes.

pH

6.80 ± 0.2

Growth Promotion Test

Growth promotion was carried out in accordance with the harmonized method of USP/EP/BP/JP/IP, and growth was observed under anaerobic conditions after an incubation at 30-35°C for ≤48 hours

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating not more than 100 CFU under anaerobic conditions (at 30-35°C for ≤48 hours).

Cultural Response

Cultural characteristics observed in an anaerobic atmosphere, after an incubation at 30-35°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Incubation temperature	Incubation period
Growth promoting				
Clostridium sporogenes 11437	50 -100	good - luxuriant	30 -35 °C	≤48 hrs
Clostridium sporogenes 19404 (00008*)	50 -100	good - luxuriant	30 -35 °C	≤48 hrs
Bacteroides vulgatus 8482	50 -100	good - luxuriant	30 -35 °C	≤48 hrs
Additional Microbiological testing				
Bacteroides fragilis 23745	50-100	good - luxuriant	30 -35°C	24 -48 hrs
Clostridium sporogenes 13124 (00007*)	50-100	good - luxuriant	30 -35°C	24 -48 hrs

Key: * Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Reinforced Medium for Clostridia M443

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (9, 10).

Reference

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Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Medium in Glass Bottles for Microbial Limit Test			
LQ130C LQ130D	Reinforced Medium for Clostridia	for the enrichment of Clostridia from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP, EP, BP, JP & IP.	5X100ml 5X500ml

Columbia Agar M144

Intended Use:

Columbia Agar (MH144 / GMH144) is used for detection of *Clostridium sporogenes* from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP (Medium 16).

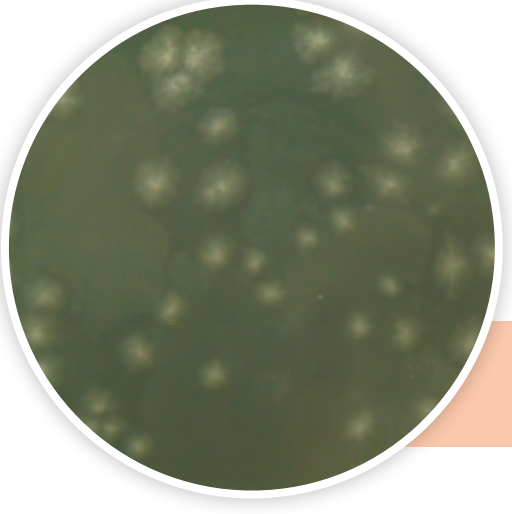
It is used as an efficient base for preparation of blood agar, chocolate agar and for preparation of various selective and identification media and isolation of organisms from clinical and non clinical samples.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified /distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C, if required add the rehydrated contents of 1 vial of Gentamicin Selective Supplement (FD252) in MH144 / GMH144. Mix well before pouring into sterile Petri plates.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg™	Chemically defined
	M144	GM144	MH144	GMH144	MV144	MCD144
Tryptone#	_	_	10.00	10.00	_	_
HM extract##	_	_	5.00	5.00	_	_
HM hydrolysate###	_	_	3.00	3.00	_	_
Peptone, special	23.00	23.00	_	_	_	_
HiVeg™ special peptone	_	_	_	_	23.00	_
HiCynth™ Peptone No.3####	_	_	_	_	_	23.00
Yeast extract	_	_	5.00	5.00	_	_
Maize starch	_	_	1.00	1.00	_	_
Corn starch	1.00	1.00	_	_	1.00	1.00
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00
Agar	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	44.00	44.00	44.00	44.00	44.00	44.00
Final pH (at 25°C)	7.3± 0.2	7.3± 0.2	_	_	7.3± 0.2	7.3± 0.2
pH after sterilization (at 25°C)	_	_	*7.3±0.2	*7.3±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min
Supplements	Blood and / or FD's as desired	Blood and / or FD's as desired	FD252	FD252	Blood and / or FD's as desired	Blood and / or FD's as desired

*pH can also be measured afer heating, at 25°C
#Equivalaent to Pancreatic digest of casein
##Equivalent to Meat peptic digest
###Equivalent to Heart pancreatic digest
####Chemically defined peptone



MH144 Columbia Agar
Clostridium sporogenes ATCC 19404 (00008*)
*Corresponding WDCM no.

Columbia Agar M144

Principle And Interpretation

Columbia Blood Agar Base used as a general-purpose nutritious medium was devised by Ellner et al from Columbia University, which was further enriched by the addition of sheep blood (1). It can also be used for the isolation of organisms by addition of various supplements. Columbia Agar is prepared in accordance with the microbial limit testing harmonized methodology of USP/EP/BP/JP/IP (2,3,4,5,6). This medium is recommended to check the presence of *Clostridium* in non-sterile products like food, dietary, nutritional supplements related products. The genus *Clostridium* belongs to the family *Clostridiaceae* in the class Clostridia.

The product to be examined is initially enriched in Reinforced medium for clostridia. This medium contains 0.05% Agar and cysteine, which creates anaerobic conditions, thereby allowing anaerobic organisms to grow. The enriched sample is then subcultured on Columbia Agar. Columbia Agar is used as a base for media containing blood and for selective media formulations in which different combinations of antimicrobial agents are used as additives.

This medium is highly nutritious as it contains Tryptone, peptone special, HM extract, HM hydrolysate, HiCynth™ peptone, HiVeg™ special peptone, Yeast extract which supports rapid and luxuriant growth of fastidious as well as non-fastidious organisms providing nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential growth factors. Sodium chloride maintains osmotic balance of medium. Maize starch acts as an energy source and also neutralizes toxic metabolites if produced. It is used in detection of Clostridia from pharmaceutical products. Gentamicin (FD252)(used in MH144 / GMH144) inhibits a number of contaminating gram-negative organisms and *Staphylococcus* species.

Clostridia grows under anaerobic conditions as gram positive rods giving a catalase negative test. Further confirmation is carried out by identification tests.

Type of specimen

Pharmaceutical samples, Clinical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (7, 8).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (2, 3, 4, 5, 6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Certain fastidious organisms like *Haemophilus influenzae* may not grow on the medium, blood supplementation may be required.
2. As this medium have a relatively high carbohydrate content, β-haemolytic streptococci may exhibit a greenish haemolytic reation which may be mistaken for the α-haemolysis.
3. Carry out confirmatory tests of all the colonies.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM144 / GMH144 : Cream to yellow granular media

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri plates

pH

7.10-7.50

Growth Promotion Test

Growth Promotion was carried out in accordance with the harmonized method of USP/EP/BP/JP, and growth was observed under anaerobic conditions after an incubation at 30-35°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Casein Soybean Digest Agar (Soybean Casein Digest Agar).

Growth promoting properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤100 CFU under anaerobic conditions (at 30-35°C for ≤48 hours).

Cultural Response (MH144)

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Incubation temperature	Incubation period
Growth promoting					
Clostridium sporogenes 19404 (00008*)	50 -100	luxuriant	≥50%	30 -35°C	≤48 hrs
Clostridium sporogenes 11437	50 -100	luxuriant	≥50%	30 -35°C	≤48 hrs
Bacteroides vulgatus 8482	50 -100	luxuriant	≥50%	30 -35°C	≤48 hrs
Additional Microbiological testing					
Clostridium perfringens 13124 (00007*)	50-100	luxuriant	≥50%	30 -35°C	≤48 hrs
Bacteroides fragilis 23745	50-100	luxuriant	≥50%	30 -35°C	≤48 hrs

Columbia Agar M144

Cultural Response (M144)

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Haemolysis
Neisseria meningitidis 13090	50 -100	luxuriant	≥70%	none
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	luxuriant	≥70%	beta / gamma
Staphylococcus epidermidis 12228 (00036*)	50 -100	luxuriant	≥70%	gamma
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	luxuriant	≥70%	beta / gamma
Staphylococcus aureus NCIMB 9518	50 -100	luxuriant	≥70%	beta / gamma
Streptococcus pneumoniae 6303	50 -100	luxuriant	≥70%	alpha
Streptococcus pyogenes 19615	50 -100	luxuriant	≥70%	beta
Clostridium sporogenes 19404 (00008*)	50 -100	luxuriant	≥50%	-
Clostridium sporogenes 11437	50 -100	good-luxuriant	≥50%	-
Clostridium perfringens 13124 (00007*)	50 -100	luxuriant	≥50%	-
Clostridium perfringens 12934	50 -100	luxuriant	≥50%	-

Key: * Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7, 8).

Reference

1. Ellner, Stoessel, Drakeford and Vasi, 1966, Am. J. Clin. Pathol., 45:502.
2. The United States Pharmacopoeia, 2019, The United States Pharmacopeial Convention. Rockville, MD.
3. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
4. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia.
5. Japanese Pharmacopoeia, 2008. Revision : 2 / 2015
6. Indian Pharmacopoeia, 2018, Govt.of India, the Controller of Publication, New Delhi
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Petri Plate			
MP144	Columbia 5% Sheep Blood Agar Plate	for isolation and cultivation of fastidious organisms.	20plts / 50plts
MPH144 MPH144GT	Columbia Agar Plate Columbia Agar Plate (γ irradiated) (Triple pack)	for the selection and subculture of *Clostridium sporogenes* in accordance with the harmonized method of USP/EP/BP/JP/IP.	20plts / 50plts 20plts / 50plts
Category : Ready Prepared Media in 55 mm Scored Petri Plate			
SPH144	Columbia Agar Plate	for the selection and subculture of *Clostridium sporogenes* in accordance with the harmonized method of USP/EP/BP/JP/IP.	100plts
Category : Ready Prepared Solid Media in Glass Bottles			
SMH144 SMH144D	Columbia Agar	for the selection and subculture of *Clostridium sporogenes* in accordance with the harmonized method of USP/ EP/ BP/ JP/ IP.	5X100ml 5X500ml

Sabouraud Dextrose Broth M033

Intended Use:

Sabouraud Dextrose Broth (MH033 / GMH033) is used for cultivation of yeasts, moulds and aciduric microorganisms from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP. (Medium 3)

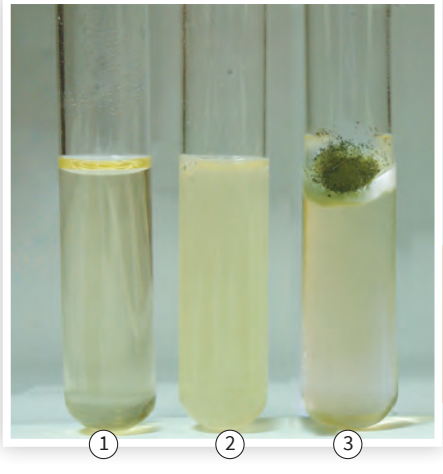
Also used for cultivation of yeasts, moulds and aciduric microorganisms from clinical and environmental samples.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	IP	HiVeg™	Chemically defined
	M033	GM033	MH033	GMH033	MM033	MV033	MCD033
Mixture of Peptone and Tryptone (1:1)#	_	_	10.00	10.00	_	_	_
Peptone, special	10.00	10.00	_	_	_	_	_
HMC peptones###	_	_	_	_	10.00	_	_
HiVeg™ special peptone	_	_	_	_	_	10.00	_
HiCynth™ Peptone No.2##	_	_	_	_	_	_	10.00
Dextrose (Glucose)	20.00	20.00	20.00	20.00	_	20.00	20.00
Dextrose monohydrate	_	_	_	_	20.00	_	_
Grams/litre	30.00	30.00	30.00	30.00	28.18	30.00	30.00
Final pH (at 25°C)	5.6 ± 0.2	5.6 ± 0.2	_	_	_	5.6 ± 0.2	5.6 ± 0.2
pH after sterilization (at 25°C)	_	_	*5.6±0.2	*5.6±0.2	5.6 ± 0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer heating, at 25°C
#Equivalent to Mixture of Peptic digest of animal tissue and Pancreatic digest of casein (1:1)
Chemically defined peptone
###Equivalent to peptones (meat and casein)



MH033 Sabouraud Dextrose Broth
1. Control
2. *Candida albicans* ATCC 10231 (00054*)
3. *Aspergillus brasiliensis* ATCC 16404 (00053*)

* Corresponding WDCM nos.

Sabouraud Dextrose Broth M033

Principle And Interpretation

Fungi were among the first microorganisms recognized because some of the fruiting structures, such as the mushrooms, are large enough to be seen without a microscope. Fungi can be grouped simply on the basis of morphology as either yeasts or moulds (1). Fungal diseases that occur on the skin, hair and mucous membrane are called superficial mycoses, and the organism that cause them, the dermatophytes (2). Where fungi are to be isolated, it is good practice to use a medium that favors their growth but is not optimal for the growth of bacteria.

Sabouraud Dextrose Broth is a modification of Dextrose Agar described by Sabouraud (3). It is useful for the cultivation of fungi. This medium is in accordance with the harmonized method of USP/EP/BP/JP (4, 5, 6, 7) and is recommended for microbiological examination of non-sterile products. This medium is also recommended by IP (8).

Peptone special, HMC peptone, Hiveg™ special peptone, HiCynth™ peptone and Mixture of peptone and Tryptone provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other nutrients essential for the growth of fungi. Dextrose (Glucose) acts as the energy source.

Type of specimen

Pharmaceutical samples, Clinical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1, 9).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (4, 5, 6, 7, 8).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. It is a general purpose medium, so bacterial cultures will also grow.
2. Further isolation and biocemical testing should be carried out for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM033 / GMH033 : Cream to yellow granular media

Colour and Clarity of prepared medium

Light amber coloured clear solution in tubes

pH

5.60 ± 0.2

Growth Promotion Test

Growth Promotion was observed in accordance with the harmonized method of USP/EP/BP/JP after an incubation at 30-35°C for 3-5 days.

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating 100 CFU (at 30-35°C for 3-5 days).

Cultural Response

Cultural characteristics observed after incubation at 20-25 °C for 3-5 days

Organism (ATCC)	Inoculum (CFU)	Growth	Incubation temperature	Incubation period
Growth promoting				
Candida albicans 10231 (00054*)	50 -100	luxuriant	30 -35°C	≤3 d
Growth Promotion + Total Yeast and Mould count				
Candida albicans 10231 (00054*)	50 -100	luxuriant	20 -25 °C	≤5 d
#Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant	20 -25 °C	≤5 d
Additional Microbiological testing				
Saccharomyces cerevisiae 9763 (00058*)	50-100	luxuriant	20 -25 °C	3 -5 d
Saccharomyces cerevisiae ATCC 2601	50-100	good-luxuriant	20 -25 °C	3 -5 d
Candida albicans 2091 (00055*)	50 -100	luxuriant	20 -25 °C	3 -5 d

Key: # Formerly known as *Aspergillus niger*
* Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (1, 9).

Sabouraud Dextrose Broth M033

Reference

1. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
2. Pelczar M. J., Jr., Reid R. D., Chan E. C. S., 1977, Microbiology, 4th Edi, Tata McGraw-Hill Publishing Company Ltd, New Delhi
3. Sabouraud, 1892, Ann. Dermatol. Syphilol, 3:1061.
4. The United States Pharmacopoeia, 2019, The United States Pharmacopeial Convention,Rockville, MD.
5. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
6. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
7. Japanese Pharmacopoeia, 2016.
8. Indian Pharmacopoeia, 2018, Govt.of India, the Controller of Publication, New Delhi.
9. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Medium in Glass Bottles for Microbial Limit Test			
LQ129V LQ129	Sabouraud Dextrose Broth	for cultivation of yeasts, moulds and aciduric bacteria.	25X5ml / 50X5ml 50X20ml
LQ120X LQ120C LQ120D	Sabouraud Dextrose Broth	for the enrichment of *Candida albicans* in accordance with harmonized methods of USP, EP, BP & JP.	25X10ml / 50X10ml 10X100ml 5X500ml

Sabouraud Dextrose Agar M063

Intended Use:

Sabouraud Dextrose Agar (MH063 / GMH063) is recommended for the cultivation of yeasts, moulds and aciduric bacteria from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP.

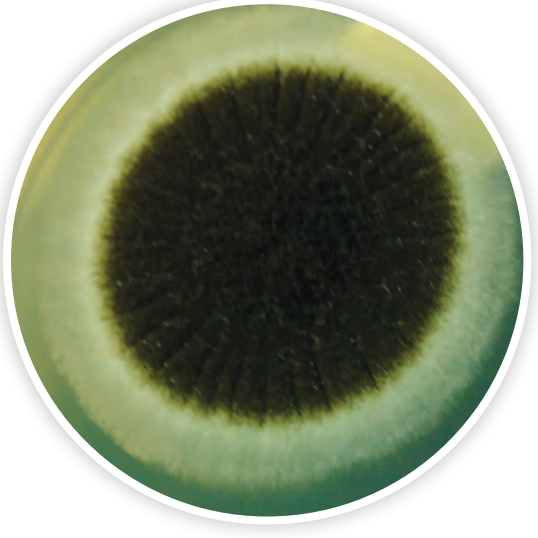
Also used for the cultivation of yeasts, moulds and aciduric bacteria from clinical and non clinical samples.

Directions:

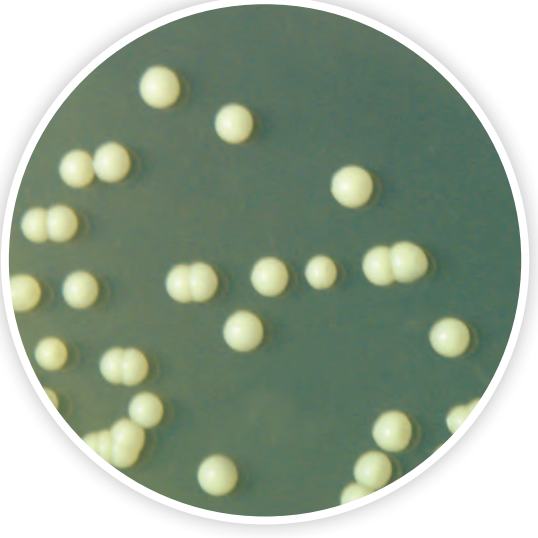
Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Ingredients	Granulated	Harmonized	Harmonized Granulated	IP (Granulated)	HiVeg™	Chemically defined
	GM063	MH063	GMH063	GMM063	MV063	MCD063
Mixture of Peptone and Tryptone (1:1)#	_	10.00	10.00	_	_	_
HMC peptones ##	_	_	_	10.00	_	_
Mycological, peptone	10.00	_	_	_	_	_
HiVeg™ peptone No. 1	_	_	_	_	10.00	_
HiCynth™ Peptone No.1###	_	_	_	_	_	10.00
Dextrose (Glucose)	40.00	40.00	40.00	_	40.00	40.00
Dextrose monohydrate	_	_	_	40.00	_	_
Agar	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	65.00	65.00	65.00	61.36	65.00	65.00
Final pH (at 25°C)	5.6 ± 0.2	_	_	5.6 ± 0.2	5.6 ± 0.2	5.6 ± 0.2
pH after sterilization (at 25°C)		*5.6±0.2	*5.6±0.2	_	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer heating, at 25°C
#Mixture of Peptic digest of animal tissue and Pancreatic digest of casein (1:1)
Peptones (meat and casein)
###Chemically defined peptone



MH063 Sabouraud Dextrose Agar
Aspergillus brasiliensis ATCC 16404 (00053*)



MH063 Sabouraud Dextrose Agar
Candida albicans ATCC 10231 (00054*)

*Corresponding WDCM nos.

Sabouraud Dextrose Agar M063

Principle And Interpretation

Fungi were among the first microorganisms recognized because some of the fruiting structures, such as the mushrooms, are large enough to be seen without a microscope. Fungi can be grouped simply on the basis of morphology as either yeasts or moulds (1). Fungal diseases that occur on the skin, hair and mucous membrane are called superficial mycoses, and the organism that cause them, the dermatophytes (2). Where fungi are to be isolated, it is good practice to use a medium that favors their growth but is not optimal for the growth of bacteria.

Sabouraud Dextrose Agar is Carliers modification (3) of the formulation described by Sabouraud (4) for the cultivation of fungi (yeasts, moulds), and aciduric microorganisms. Sabouraud Dextrose Agar is recommended for microbiological examination of non-sterile products in accordance with the harmonized method of USP/EP/BP/JP (5, 6, 7, 8). This medium is also employed in microbial limit tests in pharmaceutical testing, food, cosmetics, and clinical specimens (1). This medium is also recommended by IP (10).

Peptone, HMC peptone, Mycological peptone, HiVeg™ peptone, HiCynth™ peptone, Mixture of peptone and Tryptone provides nitrogenous, carbonaceous compounds, long chain amino acids, vitamins and other essential growth factors. Dextrose (glucose) provides an energy source. High dextrose (glucose) concentration and low pH favors fungal growth and inhibits contaminating bacteria from clinical specimens (11).

Some pathogenic fungi may produce infective spores, which are easily dispersed in air, so examination should be carried out in safety cabinet. For heavily contaminated samples, the plate must be supplemented with inhibitory agents for inhibiting bacterial growth. Growth of white colonies may be indicative of presence of *Candida albicans*. The total combined yeast and molds count is considered to be equal to the number of colony forming unit found using this medium, If bacterial colonies are detected they are counted as part of total yeast and mold count. In case the bacterial colonies exceeds the acceptance criterion, then antibiotics can be supplemented in this medium.

Type of specimen

Pharmaceutical samples, Clinical samples, food samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1, 11).

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (12).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (5, 6, 7, 8, 10).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. For heavily contaminated samples, the plate must be supplemented with inhibitory agents for inhibiting bacterial growth with lower pH.
2. Some pathogenic fungi may produce infective spores which are easily dispersed in air, so examination should be carried out in safety cabinet
3. Further biochemical tests should be carried out for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM063 / GMH063 / GMM063 : Cream to yellow granular media

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri plates

pH

5.60 ± 0.2

Growth Promotion Test

Growth Promotion was carried out in accordance with the harmonized method of ICH (USP/EP/BP/JP), after an incubation at 30-35 °C for 24-48 hours.Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar and fungus growth on Sabouraud Dextrose Agar

Growth Promoting Properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤ 100 CFU (at 30-35°C for ≤24 hours).

Indicative properties

Colonies are comparable in appearance and indication reaction to those previously obtained with previously tested and approved lot of medium occurs for the specified temperature for a period of time within the range specified inoculating ≤100CFU (at 30-35°C for 24-48 hours)

Cultural Response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Incubation temperature	Incubation period
Growth promotion + Indicative					
Candida albicans 10231 (00054*)	50 -100	luxuriant (white colonies)	≥70 %	30 -35°C	24 -48 hrs
Growth Promotion + Total Yeast and Mould count					
Candida albicans 10231 (00054*)	50 -100	luxuriant	≥70 %	20 -25 °C	≤5 d
#*Aspergillus brasiliensis* 16404 (00053*)	50 -100	luxuriant	≥70 %	20 -25 °C	≤5 d
Additional Microbiological testing					
Candida albicans 2091 (00055*)	50-100	luxuriant	≥70 %	30 -35 °C	24 -48 hrs
Saccharomyces cerevisiae 9763 (00058*)	50-100	good-luxuriant	≥70 %	30 -35 °C	24 -48 hrs

Sabouraud Dextrose Agar M063

Escherichia coli 25922 (00013*)	50 -100	good (inhibited on media with low pH)	≥70 %	30 -35 °C	24 -48 hrs
Escherichia coli 8739 (00012*)	50 -100	good (inhibited on media with low pH)	≥70 %	30 -35 °C	24 -48 hrs
Escherichia coli NCTC 9002	50 -100	good (inhibited on media with low pH)	≥70 %	30 -35 °C	24 -48 hrs
Trichophyton rubrum 28919	50 -100	good	≥70 %	20 -25 °C	≤5 d
Lactobacillus casei 334	50 -100	luxuriant	≥70 %	30 -35 °C	24 -48 hrs

Key: # Formerly known as *Aspergillus niger*
* Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (1, 11).

Reference

1. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
2. Pelczar M. J., Jr., Reid R. D., Chan E. C. S., 1977, Microbiology, 4th Edi, Tata McGraw-Hill Publishing Company Ltd, New Delhi
3. Carlier G. I. M., 1948, Brit. J. Derm. Syph., 60:61.
4. Sabouraud K., 1892, Ann. Dermatol. Syphilol, 3:1061
5. The United States Pharmacopoeia, 2019, The United States Pharmacopoeial Convention.,,Rockville, MD.
6. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
7. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
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9. Bacteriological Analytical Manual, 8th Edition, Revision A, 1998. AOAC, Washington D.C.
10. Indian Pharmacopoeia, 2018, Govt. of India, Ministry of Health & Family Welfare, New Delhi, India.
11. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
12. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Plates			
MPH063 MPH063GT	Sabouraud Dextrose Agar Plate Sabouraud Dextrose Agar Plate (γ- irradiated) (Triple Pack)	for the subculture of *Candida albicans* in accordance with the harmonized method of USP/EP/BP/JP.	20pt / 50pt 20pt / 50pt
MP063G MP063GT MP063AGT	Sabouraud Dextrose Agar Plate (γ- irradiated) Sabouraud Dextrose Agar Plate (γ- irradiated) (Triple Pack) Sabouraud Dextrose Agar Plate w/ 1% Glycerol (γ- irradiated) (Triple Pack)	for cultivation of yeasts, moulds and aciduric microorganisms.	20pt / 50pt 50pt 50pt
Category : Ready Prepared Media in 55 mm scored Plates			
SP063G SP063GT	Sabouraud Dextrose Agar Plate (γ- irradiated) Sabouraud Dextrose Agar Plate (γ- irradiated) (Triple Pack)	for cultivation of yeasts, moulds and aciduric microorganisms.	100pt 100pt
SPH063G	Sabouraud Dextrose Agar Plate (γ- irradiated)	for the subculture of *Candida albicans* in accordance with the harmonized method of USP/EP/BP/JP.	100pt
SP063	Sabouraud Dextrose Agar Plate	for the cultivation of yeast moulds and aciduric bacteria.	100pt
Category : Ready Prepared Solid Media in Glass Bottles			
SM063D	Sabouraud Dextrose Agar	for cultivation of yeasts, moulds and aciduric microorganisms.	5X500ml
SMH063 SMH063CCL SMH063D	Sabouraud Dextrose Agar	for cultivation of *C. albicans* in accordance with the harmonized method of USP/EP/BP/JP.	5X100ml 5X250ml 5X500ml
Category : Ready Prepared Solid Media in Glass Bottles			
PS063	Agar Strip - SB	Sabouraud-Dextrose-Agar for Yeasts and Moulds	10strips / 20strips / 50strips

Potato Dextrose Agar M096

Intended Use:

Potato Dextrose Agar (MH096 / GMH096) is recommended for the cultivation of yeasts and moulds from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of USP/EP/BP/JP.

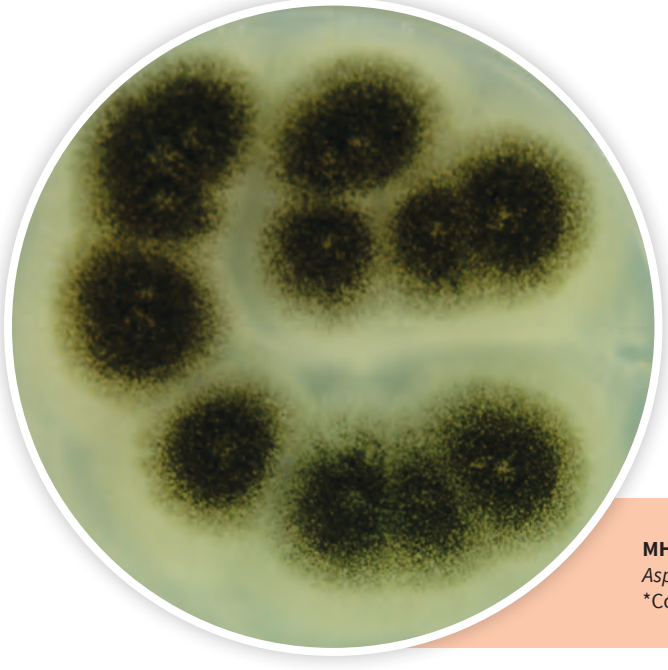
Also recommended for the isolation and enumeration of yeasts and moulds from dairy and other food products.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified /distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates. In specific work, when pH 3.5 is required, acidify the medium with sterile 10% tartaric acid. The amount of acid required for 100 ml. of sterile, cooled medium is approximately 1 ml. Do not heat the medium after addition of the acid.

Ingredients	Granulated	Harmonized	Harmonized Granulated	Chemically defined
	GM096	MH096	GMH096	MCD096
Infusion from potatoes	$200.00	$200.00	$200.00	_
Dextrose (Glucose)	20.00	20.00	20.00	20.00
HiCynth™ Peptone No.2#	_	_	_	4.00
Agar	15.00	15.00	15.00	15.00
Grams/litre	39.00	39.00	39.00	39.00
Final pH (at 25°C)	5.6 ± 0.2	_	_	5.6 ± 0.2
pH after sterilization (at 25°C)	_	*5.6±0.2	*5.6±0.2	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

*pH can also be measured afer heating, at 25°C
Chemically defined peptone
$ Equivalent to 4 grams



MH096 Potato Dextrose Agar
Aspergillus brasiliensis ATCC 16404 (00053*)
*Corresponding WDCM nos.

Potato Dextrose Agar M096

Principle And Interpretation

Yeast and moulds constitute a large and divergent group of microorganisms consisting of several thousands species. Yeast and moulds can cause various degrees of food decomposition. Invasion and growth may occur on virtually any type of food if environmental conditions are not limiting. Some foodborne yeasts and moulds are undesirable because of potential hazards to human and animal health (1).

Potato Dextrose Agar, prepared in accordance with the harmonized methodology of USP/EP/BP/JP (2,3,4,5) is recommended for microbial limit tests in pharmaceutical testing. It is also used for stimulating sporulation, for maintaining stock cultures of certain dermatophytes and for differentiation of typical varieties of dermatophytes on the basis of pigment production (6).

Potato infusion and dextrose (glucose) promote luxuriant fungal growth. Adjusting the pH of the medium by tartaric acid to 3.5 inhibits the bacterial growth. Heating the medium after acidification should be avoided as it may hydrolyse the agar, which can render the agar unable to solidify.

Type of specimen

Pharmaceutical samples, Food and dairy samples, Clinical samples.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1, 7, 8).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmacopoeias (2, 3, 4, 5).

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (9, 10).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Due to nutritional variations some strains may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM096 / GMH096 : Cream to yellow granular media

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri plates.

pH

5.60 ± 0.2

Growth Promotion Test

Growth Promotion was carried out in accordance with the harmonized method of USP/EP/BP/JP, and growth was observed at 20-25°C for specified time. Recovery rate is considered as 100% for fungus growth on Sabouraud Dextrose Agar

Growth Promoting Properties

Growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating ≤ 100 CFU

Cultural Response

Cultural characteristics observed after incubation at 20-25°C for 2-5 days. Recovery rate is considered as 100% for fungus growth on Sabouraud Dextrose Agar.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Incubation temperature	Incubation period
Test strain preparation					
#Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant	≥50 %	20 -25 °C	5 -7 Day
Additional Microbiological Testing					
Aspergillus fumigatus 9197	50 -100	luxuriant	≥50 %	20 -25 °C	5 -7 Day
Candida albicans 10231 (00054*)	50 -100	luxuriant	≥70 %	20 -25 °C	2 -3 Day
Saccharomyces cerevisiae 9763 (00058*)	50-100	luxuriant	≥70 %	20 -25 °C	2 -5 Day
Rhodotorula mucilaginosa DSM 70403		luxuriant		20 -25 °C	3 -5 Day
Geotrichum candidum DSM 1240		good-luxuriant		25 -30 °C	3 -5 Day
Penicillium communae 10248		fair -good		25 -30 °C	3 -5 Day
Trichophyton ajelloi 28454		fair -good		25 -30 °C	3 -7 Day

Key: # Formerly known as *Aspergillus niger*
* Corresponding WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Potato Dextrose Agar M096

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (9, 10).

Reference

1. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
2. The United States Pharmacopoeia, 2019, The United States Pharmacopoeial Convention. Rockville, MD.
3. European Pharmacopoeia, 2017, European Dept. for the quality of Medicines.
4. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
5. Japanese Pharmacopoeia, 2016.
6. MacFaddin J., 1985, Media for the Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol.1, Williams and Wilkins, Baltimore
7. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
8. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
9. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
10. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media in 90 mm Plates			
MPH096	Potato Dextrose Agar Plate	for the subculture of fungi in accordance with the harmonized method of USP/EP/BP/JP.	20pt / 50pt
MP096	Potato Dextrose Agar Plate	for isolation and enumeration of yeasts and moulds from dairy and other food products.	20pt / 50pt
Category : Ready Prepared Media in 55 mm scored Plates			
SPH096G	Potato Dextrose Agar Plate (γ- irradiated)	ffor the subculture of fungi in accordance with the harmonized method of USP/EP/BP/JP.	100pt
Category : Ready Prepared Solid Media in Glass Bottles			
SM096 SM096D	Potato Dextrose Agar	for isolation and enumeration of yeasts and moulds from dairy and other food products.	5X100ml 5X500ml
SMH096 SMH096D	Potato Dextrose Agar	for the subculture of fungi in accordance with the harmonized method of USP/EP/BP/JP.	5X100ml 5X500ml
Category : Ready Prepared Slant in Glass Tubes			
SL096	Potato Dextrose Agar Slant	for isolation and enumeration of yeast and moulds from dairy and other food products.	10SL / 25SL

Systematic Scheme for Test For Specified Micro Organisms*

As per Harmonized method of USP/EP/BP/JP

T E S T S A M P L E
Pharmaceutical raw materials or finished products

Phosphate Buffer pH 7.2 /Buffered Sodium Chloride Peptone Solution pH 7.0 (MH1275) OR Soybean Casein Digest Broth (MH011) OR Soybean Casein Digest Broth W/ Neutralizers

Test For Specified Micro Organisms

Test for *Escherichia coli*	Test for *Salmonella*	Test for *Pseudomonas aeruginosa*	Test for *Staphylococcus aureus*	Test for *Clostridia*	Test for Bile Tolerant Organisms	Test for *Candida albicans*
Sample Preparation Soybean Casein Digest Broth (MH011)	**Sample Preparation** Soybean Casein Digest Broth (MH011)	**Sample Preparation** Soybean Casein Digest Broth (MH011)	**Sample Preparation** Soybean Casein Digest Broth (MH011)	**Sample Preparation** Soybean Casein Digest Broth (MH011)	**Sample Preparation** Soybean Casein Digest Broth (MH011)	**Sample Preparation** Sabouraud Dextrose Broth (MH033)
Selection MacConkey Broth (MH083)	**Selection** RVS Broth (MH1491)	**Selection & Subculture** Cetrimide Agar (MH024)	**Selection & Subculture** Mannitol Salt Agar (MH118)	**Selection** Reinforced Medium For *Clostridia* (MH443)	**Selection** Enterobacteria Enrichment Broth Mossel (MH287)	**Selection & Subculture** Sabouraud Dextrose Agar (MH063)
Subculture MacConkey Agar (MH081)	**Subculture** Xylose Lysine Deoxycholate Agar(MH031)			**Subculture** Columbia Agar (MH144)	**Subculture** Violet Red Bile Glucose Agar (MH581)	

Total Aerobic Microbial Count

Plate Method	Multiple Tube Method
Soybean Casein Digest Agar (MH290) & **Yeast & Mold Count** Sabouraud Dextrose Broth (MH033) or Sabouraud Dextrose Agar (MH063) or Potato Dextrose Agar (MH096) for *Candida albicans*	Soybean Casein Digest Broth (MH011)

* Formerly known as Microbial Limit Test

Sterility Testing Media

There are set guidelines for sterility testing of biologics in various pharmacopoeias and also Section 21 of the Code of Federal Regulations (CFR) and Food and Drug Administration. This document comprehends test methods and sample requirements for the most common types of biological products.

Every biological product manufactured under GMP conditions require sterility testing performed under GMP guidelines. Two common types of sterility test methods widely described are:

Direct Inoculation

The direct inoculation or immersion method involves the test article be inoculated directly into specified media.

Membrane Filtration

While in the membrane filtration method the test article has to first pass through a size exclusion membrane capable of retaining microorganisms and that filter is rinsed and transferred to the specified test medium.

The pharmacopoeias recommend using media and rinse fluids for both the immersion and membrane as per their specifications. In both test methods the test article or membrane is incubated for 14 days in the test media. The majority of biological samples will be tested using the immersion method. But if it deals with larger volumes then membrane filtration method may be required.

Fluid Thioglycollate Medium and Soybean-Casein Digest Medium are the media generally used for sterility testing. Alternative media types may be appropriate where the nature of the product or method of manufacture can result in the presence of fastidious organisms (eg vaccines, blood products). Validation studies should indicate that alternative media are capable of supporting the growth of a wide range of micro-organisms in the presence of the product.

HiMedia provides complete set of sterility media and rinse solutions as per various pharmacopoeias. Each batch of our media is tested for pH, appearance, clarity, selective ratio, growth promotion and other parameters so as to meet the specifications in the standards.

Media fill studies, simulates the filling process during production and helps in detecting contamination in the production line, if any. Soyabean casein digest medium or Tryptic Soya Broth (non-sterile bulk powder) from a commercial source, is generally used. The media is prepared, steam sterilized or filter sterilized through a 0.2 micron filter and is used to investigate presence or absence of contamination. For this purpose HiMedia provides ready prepared sterility testing media kits and gamma irradiated dehydrated culture media powder for faster, efficient and safer testing.

Media Fill : Maximum Benefits & Minimizing Risks with HiVeg™ Gamma Irradiated TSB.

Sterile (Control) | Microbial Growth (Change of colour)

Media fills simulate the whole process in order to evaluate the sterility confidence of the process. Process simulation studies include formulation, filtration and filling with suitable media. In general, a microbiological growth medium such as Tryptic Soy Broth should be used. Use of anaerobic growth media (e.g. Fluid Thioglycollate medium) should be considered in special circumstances.

With the spurt in number of BSE symptoms across global bovine population & and its exhibit CJD in humans concerns were raised about bovine origin products.

Elimination of BSE/TSE Risk can be achieved by use of raw material from right origin & right parts of the animal. Definition of Risk Categories by EU:

Category A: High infectivity (e.g. brain, spinal cord)
Category B: Moderate infectivity (e.g. spleen, lung, liver)
Category C: No infectivity found (e.g. milk, bile, skeletal muscle, heart, skin)

HiMedia only sources from risk category 'C' for its products. Moreover as per the Definition of Geographical BSE Risk by EU, raw material sourced from India has no listings. In spite of such a proven track record of quality, a step further to provide more secure process HiVeg™ culture media was launched. Both USP & EP preferred or recommend that alternative, non-animal source ingredients be substituted for animal-source ingredients whenever possible.

The risk of Mycoplasma is always lurking in the raw material. Moreover Mycoplasma can move through 0.2 mm filters & Reach high titers (10^7 — 10^8 CFU/ml) without producing pH changes or media turbidity proving itself as invisible threat. In such cases a prudent step ahead to provide maximum quality assurance is to provide γ-irradiated TSB.

γ- Irradiation does not affect product performance, and results in a Contaminant-free material, this has been evaluated by comparative studies on growth performance of pharmacopoeia listed pathogens. Thus HiVeg™ γ-irradiated TSB is the choice of a prudent quality system.

Introduced gamma irradiated HiFill™ Test Medium recommended for the evaluation of sterility in manufacturing process for easy detection of contamination. The medium is designed with TSB containing an MFT indicator wherein the colour change is from yellow to pink red.

Reference:

- The USP Perspective to Minimize the Potential Risk of TSE-infectivity in Bovine-derived Articles Used in the Manufacture of Medical Products; with Ian DeVeau and Roger Dabbah. Pharmacopoeial Forum. 30(5):1911-1921. 2004
- European Pharmacopoeia (Supplement 6.3), 2008, European Department, for the Quality of Medicines

HiMedia No.	Product Range for Media Fill trials
M011G-500G M011G-2.5KG M011G-5KG	**Soyabean Casein Digest Medium, Sterile Powder** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process.
MV011G-500G MV011G-2.5KG MV011G-5KG	**Soyabean HiVeg® Medium, Sterile Powder** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process.
GMV011G-500G	**Soyabean HiVeg® Medium, Granulated, Sterile** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process.
MH011G-500G	**Soyabean Casein Digest Medium, Sterile powder** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process.
GMH011G-500G	**Soyabean Casein Digest Medium, Granulated, Sterile** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process.
M1856G-500G M1856G-2.5KG	**Soyabean Casein Digest Medium w/ Mannitol, Sterile Powder** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process. It can also be used for cultivation of a wide variety of microorganisms.
M1655G-500G M1655G-2.5KG M1655G-5KG	**Soyabean Casein Digest Medium w/ BCP, Sterile Powder** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process.
M010G-500G M010G-2.5KG M010G-5KG	**Alternative Thioglycollate Medium, Sterile Powder** γ-irradiated sterile powder recommended for evaluation of sterility in manufacturing process.
MV010G-500G MV010G-2.5KG MV010G-5KG	**Alternative Thioglycollate HiVeg® Medium, Sterile Powder** γ-irradiated sterile powder recommended for evaluation of sterility in manufacturing process.
MU010G-500G MU010G-2.5KG MU010G-5KG	**Alternative Thioglycollate Medium, Sterile Powder** γ-irradiated sterile powder recommended for evaluation of sterility in manufacturing process in accordance with USP.
M2018G-500G	**HiFill™ Test Medium** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process for easy detection of contamination by Media Fill Test.
MV2018G-500G	**HiFill™ Test HiVeg® Medium** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process for easy detection of contamination by Media Fill Test.
MCD2018G-500G	**HiFill™ Test HiCynth™ Medium** γ-irradiated sterile powder recommended for the evaluation of sterility in manufacturing process for easy detection of contamination by Media Fill Test.
RM565G-5KG RM565G-50KG	**Lactose monohydrate, Sterile** (γ irradiated sterile powder)
RM565GT-5KG	**Lactose monohydrate, Sterile Powder** (γ irradiated Triple Pack)
RM570G-5KG RM570G-50KG	**D-Mannitol, A. R. sterile** (γ irradiated)

Alternative Thioglycollate Medium (NIH Thioglycollate Broth) M010

Intended Use:

Alternative Thioglycollate Medium is recommended for sterility testing of turbid or viscous biological products in accordance with United States Pharmacopoeia.

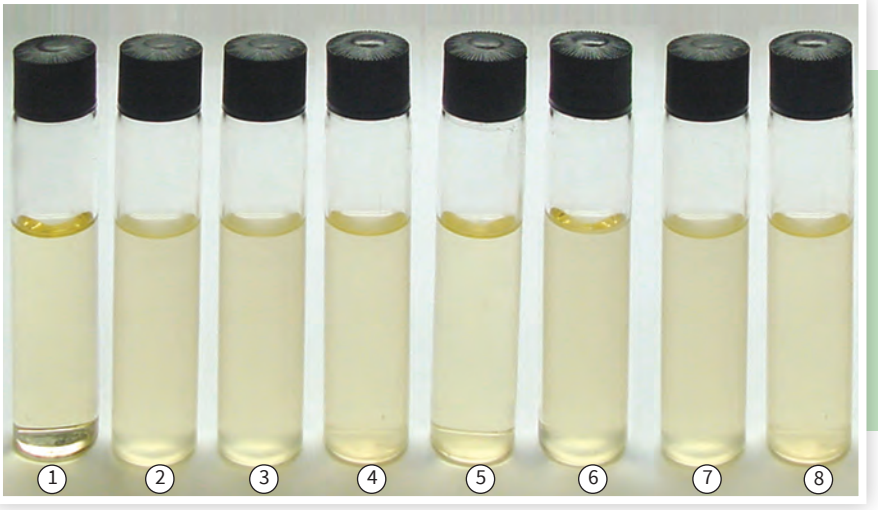
Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Distribute into flasks or tubes as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Note: It is preferable to use freshly prepared medium, alternatively it should be boiled and cooled just once prior to use or with reheating, toxic oxygen radicals are formed.

Ingredients	HiMedia	Granulated	USP	IP	HiVeg	Chemically defined
	M010	GM010	MU010	MM010	MV010	MCD010
Tryptone#	15.00	15.00	15.00	15.00	_	_
HiVeg™ hydrolysate	_	_	_	_	15.00	_
Yeast extract	5.00	5.00	5.00	5.00	5.00	_
HiCynth™ Peptone No.3##	_	_	_	_	_	15.00
HiCynth™ Peptone No.5##	_	_	_	_	_	5.00
Dextrose monohydrate	_	_	5.50	5.50	_	_
Dextrose (Glucose)	5.50	5.50	_	_	5.50	5.50
Sodium chloride	2.50	2.50	2.50	2.50	2.50	2.50
L-Cystine	0.50	0.50	0.50	0.50	0.50	0.50
Sodium thioglycollate	0.50	0.50	0.50	0.50	0.50	0.50
Grams/litre	29.00	29.00	28.50	28.50	29.00	29.00
Final pH (at 25°C)	7.1 ± 0.2	7.1 ± 0.2		7.1 ± 0.2	7.1± 0.2	7.1 ± 0.2
pH after sterilization (at 25°C)	_	_	7.1 ± 0.2	_	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

#Pancreatic digest of casein
##Chemically defined peptones



M010 Alternative Thioglycollate Medium (NIH Thioglycollate Broth)

1. Control
2. *Streptococcus pyogenes* ATCC 19615
3. *Staphylococcus aureus* subsp. *aureus* ATCC 25923 (00034*)
4. *Bacillus subtilis* subsp *spizizenii* ATCC 6633 (00003*)
5. *Bacteroides vulgatus* ATCC 8482
6. *Candida albicans* ATCC 10231 (00054*)
7. *Bacteroides fragilis* ATCC 25285
8. *Clostridium sporogenes* ATCC 19404 (00008*)

*Corresponding WDCM Nos.

Alternative Thioglycollate Medium (NIH Thioglycollate Broth) M010

Principle And Interpretation

Alternative Thioglycollate Medium is formulated as described in N.I.H. Memorandum (1), U.S. Pharmacopoeia (2). It is also recommended by IP (3), EP (6) and BP (7). This medium is recommended for sterility testing for detecting the presence of viable forms of microorganisms in or on pharmaceutical preparations. This medium is also used for sterility checking for devices having tubes with small lumina. Alternative thioglycollate Medium is generally used for products containing mercurial preservatives when the oxidation reduction indicator is not present or required. Lack of an indicator in the medium avoids possible toxicity to organisms.

Alternative Thioglycollate Medium contains sodium thioglycollate that can neutralize the bacteriostatic effect of mercurial preservatives. Absence of agar makes it suitable for testing viscous materials and devices having tubes with small lumina.

Tryptone, HiVeg™ hydrolysate, yeast extract, HiCynth™ peptone, provides nitrogenous and carbonaceous compounds, long chain amino acid, vitamin B complex, trace elements and other essential growth nutrients. Dextrose monohydrate and L-cystine serves as an energy source. Sodium Thioglycollate and L-cystine lower the oxidation-reduction potential of the medium by removing oxygen radicals and thus preventing the accumulation of peroxides that can be toxic to some organisms. The sulfhydryl groups of these compounds also neutralize the antibacterial effect of mercurial preservatives with heavy metals. Dextrose is the fermentable carbohydrate energy source, and Sodium chloride maintains the osmotic balance of the medium.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (2, 3, 6, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. The tubes should not be reheated as frequent boiling leads to development of toxic products.
2. Prior to use the medium should be boiled once to remove the absorbed oxygen.
3. Before inoculation, the tubes should be brought to room temperature
4. The medium should not be used in fermentation process as medium contains yeast extract which is high in carbohydrate content.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

GM010 - Cream to yellow granular media.

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate.

Reaction

pH : 7.1±0.2

Cultural Response

Growth Promotion observed in accordance with USP, under anaerobic condition after an incubation at 30-35°C for ≤3 days.

Organism (ATCC)	Inoculum (CFU)	Growth
Growth Promotion Test		
Clostridium sporogenes 19404 (00008*)	50 -100	luxuriant
Clostridium sporogenes 11437	50 -100	luxuriant
Bacteroides vulgatus 8482	50 -100	luxuriant
Additional Microbiological testing		
Staphylococcus aureus subsp. aureus 25923 (00034*)	50 -100	luxuriant
Staphylococcus aureus subsp. aureus 6538 (00032*)	50 -100	luxuriant
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant
Escherichia coli 25922 (00013*)	50 -100	luxuriant
Escherichia coli 8739 (00012*)	50 -100	luxuriant
Escherichia coli NCTC 9002	50 -100	luxuriant
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant
Clostridium perfringens 13124 (00007*)	50 -100	luxuriant
Bacteroides fragilis 23745	50 -100	luxuriant

Key : * corresponding WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Alternative Thioglycollate Medium (NIH Thioglycollate Broth) M010

Reference

1. N.I.H. Memorandum, 1955 : Culture Media for Sterility Tests, 4th Revision.
2. The United States Pharmacopoeia 2019, US Pharmacopoeial Convention Inc. ,Rockville, M.D
3. Indian Pharmacopoeia, 2018, Govt. of India, the controller of Publication, Delhi, India.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
6. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia.
7. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Medium in Glass Bottles for Sterility Test Media			
LQ028 LQ028DW	Alternative Thioglycollate Medium Alternative Thioglycollate Medium - Double Packed	sterility test medium prepared in accordance with USP	10x100ml 10x100ml
Category : Ready Prepared Steriity Test Medium Kit in Glass Bottles			
LQ025	Sterility Kit - II	sterility test media prepared in accordance with IP/USP/EP/BP/ JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ028 - Sterile Alternative Thioglycollate Medium and LQ027 - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5kit
LQ025A	Sterility Kit - II	sterility test media prepared in accordance with IP/USP/EP/BP/ JP. One Kit contains 2 glass bottles each of 100 ml following media LQ028A - Sterile Alternative Thioglycollate Medium and LQ027A - Sterile Soyabean Casein Digest Medium . Recommended for all purposes	20kit
Category : Ready Prepared Transport Medium with Swabs			
MS010	HiCulture™ Transport Swabs w/ Alternative Thioglycollate Medium	for transportation of aerobes, anaerobes and microaerophiles	10no / 50no
MS010S	HiCulture™ Transport Swabs w/ Alternative Thioglycollate Medium w/ Metal stick	for transportation of aerobes, anaerobes and microaerophiles	10no / 50no

Fluid Thioglycollate Medium M009

Intended Use:

Fluid Thioglycollate Medium is used for sterility testing of biologicals and for cultivation of aerobes, anaerobes and microaerophiles in accordance with Pharmacopoeia.

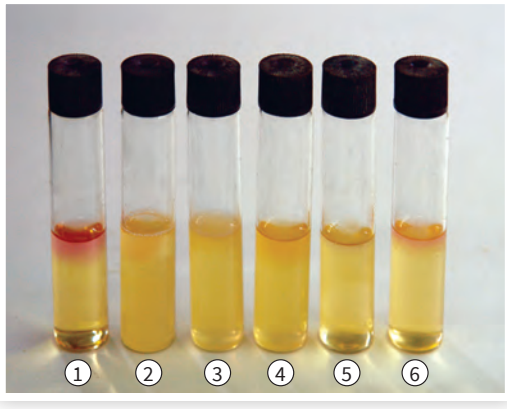
Directions:

Suspend dehydrated medium as per table in 1000 ml purified/ distilled water. Heat to boiling to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 25°C and store in a cool dark place preferably below 25°C.

Note: If more than the upper one-third of the medium has acquired a pink colour, the medium may be restored once by heating in a water bath or in free flowing steam until the pink colour disappears.

Ingredients	HiMedia	Granulated	USP	EP	BP	IP	HiVeg	Chemically defined
	M009	GM009	MU009	ME009	M009B	MM009	MV009	MCD009
Tryptone#	15.00	15.00	15.00	15.00	15.00	15.00	_	_
HiVeg™ hydrolysate	_	_	_	_	_	_	15.00	_
HiCynth™ Peptone No.3##	_	_	_	_	_	_	_	15.00
HiCynth™ Peptone No.5##	_	_	_	_	_	_	_	5.00
Yeast extract	5.00	5.00	5.00	5.00	5.00	5.00	5.00	_
Sodium chloride	2.50	2.50	2.50	2.50	2.50	2.50	2.50	2.50
Dextrose monohydrate (Glucose monohydrate)	_	_	5.50	5.50	5.50	5.50	_	_
Dextrose(Glucose)	5.50	5.50	_	_	_	_	5.50	5.50
L-Cystine	0.50	0.50	0.50	0.50	0.50	0.50	0.50	2.50
Sodium thioglycollate	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Resazurin sodium	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
Agar	0.750	0.750	0.750	0.750	0.750	0.750	0.750	0.750
Grams/litre	29.75	29.75	29.25	29.25	29.25	29.25	29.75	29.75
Final pH (at 25°C)	7.1± 0.2	7.1± 0.2	_	_	_	_	7.1± 0.2	7.1± 0.2
pH after sterilization (at 25°C)	_	_	7.1± 0.2	7.1± 0.2	7.1 ± 0.2	7.1± 0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer sterilization, at 25°C
#Pancreatic digest of casein
##Chemically defined peptones



M009 Fluid Thioglycollate Medium

1. Control
2. *Clostridium sporogenes* ATCC 19404 (00008*)
3. *Clostridium sporogenes* ATCC 11437
4. *Clostridium perfringens* ATCC 13124 (00007*)
5. *Salmonella* Typhimurium ATCC 14028 (00031*)
6. *Pseudomonas aeruginosa* ATCC 9027 (00026*)

*corresponding WDCM Nos.

Fluid Thioglycollate Medium M009

Principle And Interpretation

Brewer (1) formulated Fluid Thioglycollate Medium for rapid cultivation of aerobes as well as anaerobes by adding a reducing agent and small amount of agar. The USP (2) , BP (3) , EP (4) IP (10) and AOAC (5) have recommended the media for sterility testing of antibiotics, biologicals and food products and for determining the phenol coefficient and sporicidal effect of disinfectants.

However, it is intended for the examination of clear liquid or water-soluble materials.

Tryptone, HiVeg™ hydrolysate, HiCynth™ peptone and yeast extract provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential nutrients. Dextrose monohydrate (glucose monohydrate) is the energy sources and L-cystine provide the growth factors necessary for bacterial multiplication. Sodium thioglycollate and L-cystine act as a reducing agent lowering the oxidation-reduction potential by removal of oxygen This condition helps to prevent the accumulation of peroxides which is toxic in nature. The SH group also neutralizes the antibacterial effect of mercurial preservatives and other heavy metal compounds which exert a bacteriostatic effect in the materials under examination. Any increase in the oxygen content is indicated by a colour change of redox indicatorresazurin; to red (6,7,8). The small amount of agar helps in maintaining low redox potential and stabilizes the medium (9).

In sterility checking, it is recommended to dilute the sample containing preservatives, with this broth to reduce the toxicity and enhance the growth of contaminants, if any.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (2, 3, 4, 10).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. It is intended for the examination of clear liquid or water soluble materials.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM009 : Cream to yellow granular media

Colour and Clarity of prepared medium

Light straw coloured clear to slightly opalescent solution with upper 10% or less medium pink on standing.

Reaction

pH : 7.1±0.2

pH

6.90-7.30

Growth Promotion Test

As per United States Pharmacopoeia

Cultural response

Cultural characteristics observed after an incubation at 30-35°C for not more than 3 days.

Organism (ATCC)	Inoculum (CFU)	Growth
Growth promoting		
Clostridium sporogenes 19404 (00008*)	50 -100	luxuriant
Clostridium sporogenes 11437	50 -100	luxuriant
Clostridium sporogenes NBRC 14293	50 -100	luxuriant
Clostridium perfringens 13124 (00007*)	50 -100	luxuriant
Bacteroides fragilis 23745	50 -100	luxuriant
Bacteroides vulgatus 8482	50 -100	luxuriant
Staphylococcus aureus subsp. aureus 25923 (00034*)	50 -100	luxuriant
Staphylococcus aureus subsp. aureus 6538 (00032*)	50 -100	luxuriant
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant
Micrococcus luteus 9341	50 -100	luxuriant
Streptococcus pneumoniae 6305	50 -100	luxuriant
Escherichia coli 25922 (00013*)	50 -100	luxuriant
Escherichia coli 8739 (00012*)	50 -100	luxuriant
Escherichia coli NCTC 9002	50 -100	luxuriant
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant
Bacillus subtilis subsp. spizizenii 6633 (00003*)	50 -100	luxuriant

Key : * corresponding WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (11, 12).

Fluid Thioglycollate Medium M009

Reference

1. Brewer, 1940, J. Am. Med. Assoc., 115:598.
2. U.S. Pharmacopoeia, 2019, United States Pharmacopoeia Convention, Inc., Rockville, MD.
3. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia.
4. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines.
5. Williams. (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th ed., AOAC, Washington, D.C.
6. Marshall, Gunnison and Luxen, 1940, Proc. Soc. Exp. Biol. Med., 43:672.
7. Nungester, Hood and Warren, 1943, Proc. Soc. Exp. Biol. Med., 52:287.
8. Portwood, 1944, J. Bact., 48:255.
9. MacFaddin J.F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of ,,Medical Bacteria, Vol.1, Williams and Wilkins, Baltimore.
10. Indian Pharmacopoeia, 2018, Govt. of India, the controller of Publication, Delhi, India.
11. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
12. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Media in Tubes			
LQ026V LQ026IX LQ026X LQ026XX LQ026AX	Fluid Thioglycollate Medium	sterility test medium prepared in accordance with USP, EP, BP & JP.	25X5ml / 50X5ml 20X9ml / 100X9ml 25X10ml / 50X10ml 50X20ml 1X10ml / 5X10ml / 25X10ml
Category : Ready Prepared Liquid Media in Glass Bottles for Sterility Test Media			
LQ509	Fluid Thioglycollate Medium	sterility test medium also used as a general purpose medium for the growth of aerobes, anaerobes & microaerophiles	5X200ml
LQ026 LQ026A LQ026CV LQ026D LQ242 LQ242N LQ242DW	Fluid Thioglycollate Medium Sterile Fluid Thioglycollate Medium (Double packed) Fluid Thioglycollate Medium	recommended for sterility testing of biologics and for cultivation of aerobes, anaerobes and microaerophiles.	10X100ml 10X100ml 10X100ml 5X500ml 10X100ml 10X100ml 10X100ml
Category : Ready Prepared Sterility Test Medium Kits in Glass Bottles			
LQ024	Sterility Kit I	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ026 - Sterile Fluid Thioglycollate Medium and LQ027 - Sterile Soya-bean Casein Digest Medium. Recommended for injectables	5 kit / 20 kit
LQ024A	Sterility Kit I	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ026A - Sterile Fluid Thioglycollate Medium and LQ027A - Sterile Soyabean Casein Digest Medium. Recommended for all purposes	5 kit / 20 kit
LQ024S	Sterility Kit I	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles of 50 ml following media : LQ026S - Sterile Fluid Thioglycollate Medium and LQ027S - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5 kit / 20 kit

Soyabean Casein Digest Medium (Tryptone Soya Broth) M011

Intended Use:

Soyabean Casein Digest Medium is a general purpose medium used for cultivation of a wide variety of microorganisms and recommended for sterility testing of moulds and lower bacteria. This medium is also recommended for carrying out microbial limit tests of pharmaceutical raw materials as well as finished products and preparation.

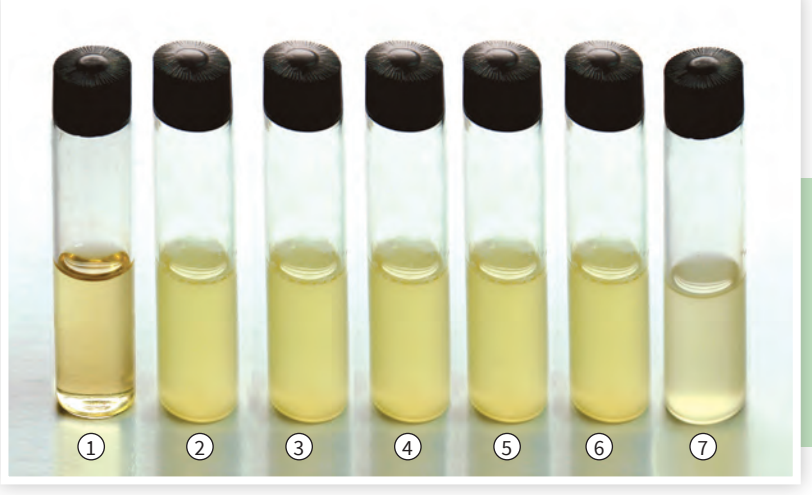
Directions:

Suspend dehydrated medium as per table in 1000 ml purified/ distilled water. Heat if necessary to dissolve the medium completely. Mix well and dispense in tubes or flasks as desired. Sterilize by autoclaving as specified below.

Note: If any fibres are observed in the solution, it is recommended to filter the solution by using a 0.22 micron filter to eliminate the possibilty of presence of fibres.

Ingredients	HiMedia	Granulated	Harmonized	Harmonized Granulated	HiVeg	Chemically defined
	M011	GM011	MH011	GMH011	MV011	MCD011
Tryptone#	17.00	17.00	17.00	17.00	_	_
HiVeg™ hydrolysate	_	_	_	_	17.00	_
HiCynth™ Peptone No.3###	_	_	_	_	_	17.00
HiCynth™ Peptone No.5###	_	_	_	_	_	3.00
Soya peptone##	3.00	3.00	3.00	3.00	3.00	_
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00
Dextrose(Glucose)	2.50	2.50	_	_	2.50	2.50
Dipotassium hydrogen phosphate	2.50	2.50	2.50	2.50	2.50	2.50
Glucose monohydrate	_	_	2.50	2.50	_	_
Grams/litre	30.00	30.00	29.77	29.77	30.00	30.00
Final pH (at 25°C)	7.3± 0.2	7.3± 0.2	_	_	7.3± 0.2	7.3± 0.2
pH after sterilization (at 25°C)	_	_	7.3± 0.2	7.3± 0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured afer sterilization, at 25°C
#Equivalent to Pancreatic digest of casein
##Equivalent to Papaic digest of soyabean meal/soyabean
Chemically defined peptones



M011 Soyabean Casein Digest Medium

1. Control
2. *Staphylococcus aureus* subsp. *aureus* ATCC 6538 (00032*)
3. *Staphylococcus aureus* subsp. *aureus* ATCC 25923 (00034*)
4. *Candida albicans* ATCC 10231 (00054*)
5. *Bacillus subtilis* subsp. *spizizenii* ATCC 6633 (00003*)
6. *Escherichia coli* ATCC 8739 (00065*)
7. *Salmonella* Typhimurium ATCC 14028 (00031*)

*corresponding WDCM Nos.

Soyabean Casein Digest Medium (Tryptone Soya Broth) M011

Principle And Interpretation

Soyabean Casein Digest Medium is recommended by various pharmacopoeias as a sterility testing medium. The media formulation is in accordance with the harmonized methodology of USP/EP/BP/JP/IP (1, 2, 3, 4, 6). It is used for the sensitivity testing of antimicrobial agents by the tube dilution method (6). It is also employed in diagnostic research in microbiology. This medium is used as an diluent and suspending medium or preparation of samples or test strains. It is also employed in sample preparation for testing products, wherein incubation is carried out, only to serve sufficient resuscitation of the cell, while avoiding multiplication of the organism.

The combination of tryptone, HiVeg™ hydrolysate, HiCynth™ peptones and soya peptone makes this medium nutritious by providing carbonaceous, nitrogenous, amino acids and other essential growth nutrients for the growth of microorganisms. Natural sugars in soyabean promote growth of fastidious organism. Glucose/dextrose is the fermentable source of carbon and dibasic potassium phosphate serves as the buffer in the medium. Sodium chloride maintains the osmotic balance of the medium.

This medium is recommended by various Pharmacopoeia for sterility checking and for studying total aerobic microbial count in verification of microbiological testing procedures employed for sterility checking.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1, 2, 3, 4, 6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Due to nutritional variations, some strains may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM011/GMH011 : Cream to yellow granular media

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate.

pH

7.10-7.50

Growth Promotion Test

Growth Promotion is carried out in accordance with the harmonized method of USP/EP/BP/JP/IP.

Stability test

Light yellow coloured clear solution without any precipitation or sedimentation at room temperature for 7 days.

Growth promoting properties

Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating not more than 100 CFU (at 30-35°C for 18-24 hours).

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Incubation temperature	Incubation period
Growth promoting				
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Staphylococcus aureus subsp. aureus 25923 (00034*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Escherichia coli 8739 (00012*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Escherichia coli 25922 (00013*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Escherichia coli NCTC 9002	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Micrococcus luteus 9341	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Salmonella Abony 6017 (00029*)	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Streptococcus pneumoniae 6305	50 -100	luxuriant	30 -35 °C	18 -24 hrs
Sterility Testing- Growth promotion+Validation				
The medium is tested with suitable strains of micro-organisms inoculating ≤ 100 CFU and Incubating at 20-25°C for not more than 3 days in case of bacteria and not more than 5 days in case of fungi.				
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	luxuriant	20 -25 °C	≤3 d
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	luxuriant	20 -25 °C	≤3 d
Escherichia coli 8739 (00012*)	50 -100	luxuriant	20 -25 °C	≤3 d
Escherichia coli 25922 (00013*)	50 -100	luxuriant	20 -25 °C	≤3 d
Escherichia coli NCTC 9002	50 -100	luxuriant	20 -25 °C	≤3 d
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	20 -25 °C	≤3 d
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	20 -25 °C	≤3 d
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	20 -25 °C	≤3 d
Micrococcus luteus 9341	50 -100	luxuriant	20 -25 °C	≤3 d
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	20 -25 °C	≤3 d
Salmonella Abony 6017 (00029*)	50 -100	luxuriant	20 -25 °C	≤3 d
Streptococcus pneumoniae 6305	50 -100	luxuriant	20 -25 °C	≤3 d
Candida albicans 10231 (00054*)	50 -100	luxuriant	20 -25 °C	≤3 d
Candida albicans 2091 (00055*)	50 -100	luxuriant	20 -25 °C	≤3 d
#Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant	20 -25 °C	≤3 d

Key : * corresponding WDCM number
Formerly known as *Aspergillus niger*

Soyabean Casein Digest Medium (Tryptone Soya Broth) M011

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7, 8).

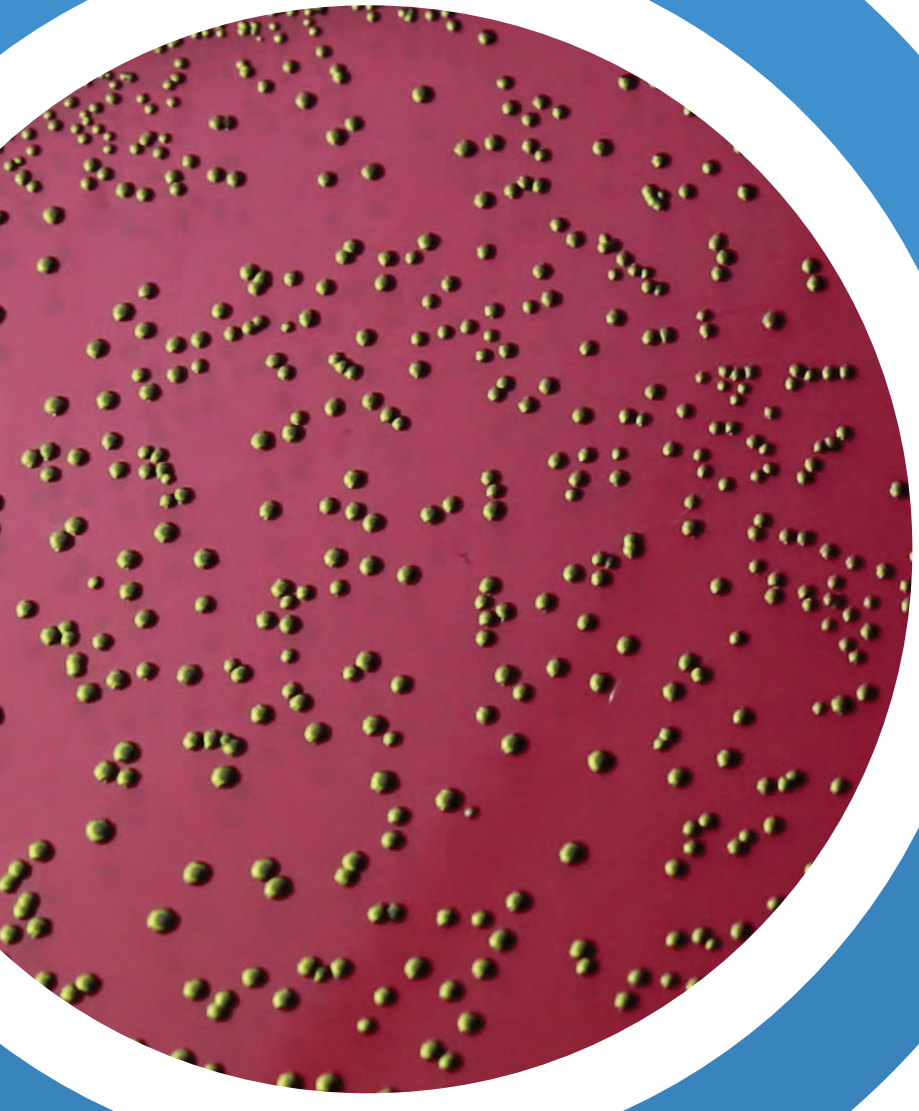
Reference

1. The United States Pharmacopoeia, 2019, The United States Pharmacopoeial Convention. Rockville, MD.
2. British Pharmacopoeia, 2016, The Stationery office British Pharmacopoeia
3. European Pharmacopoeia, 2017 European Dept. for the quality of Medicines.
4. Japanese Pharmacopoeia, 2018.
5. Wright and Welch, 1959-60, Antibiotics Ann., 61.
6. Indian Pharmacopoeia, 2018, Govt. of India, the controller of Publication, Delhi, India.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Media			
LQ027 LQ027CC LQ027D LQ027CV LQ027DW LQ027IX LQ027X LQ027XX LQ187 LQ243	Soyabean Casein Digest Medium Soyabean Casein Digest Medium-Double Packed Soyabean Casein Digest Medium Soyabean Casein Digest Medium (100ml in 125 ml glass bottle)	sterility test medium prepared in accordance with harmonized methods of USP, EP, BP, JP, IP.	10x100ml 5x200ml 5x500ml 10x100ml 10X100ml 100x9ml 50x10ml 50x20ml 5X90ml 10x100ml
LQ024	Sterility Kit I	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ026 - Sterile Fluid Thioglycollate Medium and LQ027 - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5kt / 20kt
LQ024A	Sterility Kit-I	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ026A - Sterile Fluid Thioglycollate Medium and LQ027A - Sterile Soyabean Casein Digest Medium. Recommended for all purposes	5kt / 20kt
LQ024S	Sterility Kit I	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles of 50 ml following media : LQ026S - Sterile Fluid Thioglycollate Medium and LQ027S - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5kt / 20kt
LQ025	Sterility Kit - II	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles each of 100 ml following media : LQ028 - Sterile Alternative Thioglycollate Medium and LQ027 - Sterile Soyabean Casein Digest Medium. Recommended for injectables	5kt / 20kt
LQ025A	Sterility Kit - II	sterility test media prepared in accordance with IP/USP/EP/BP/JP. One Kit contains 2 glass bottles each of 100 ml following media LQ028A - Sterile Alternative Thioglycollate Medium and LQ027A - Sterile Soyabean Casein Digest Medium . Recommended for all purposes	5kt / 20kt

Pharmacopoeial Media
Other than harmonized
For microbial examination

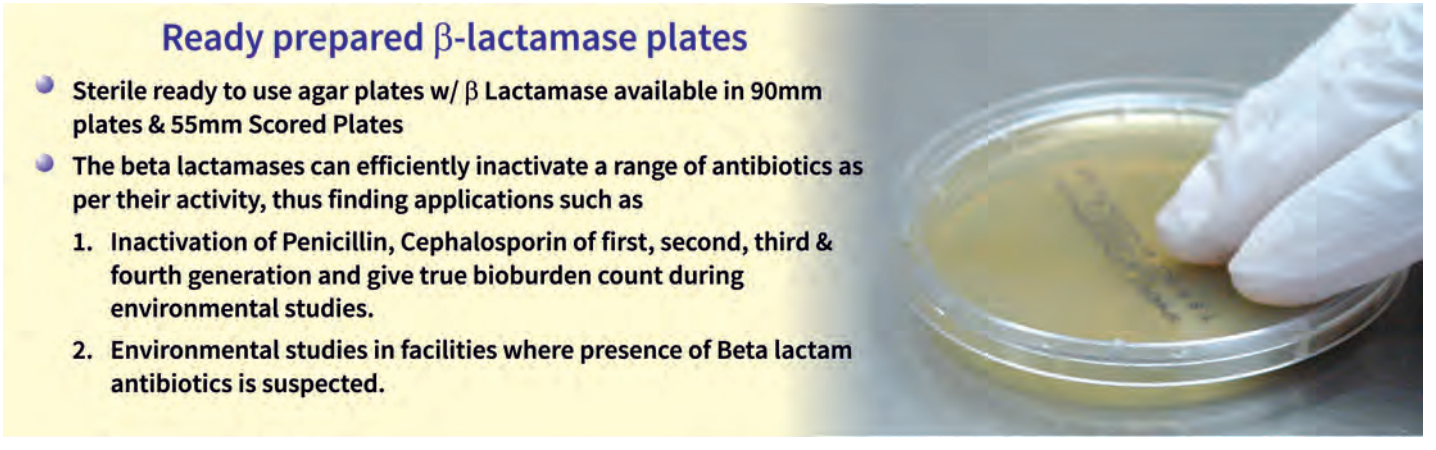


Quality control of microbiological culture media is vital to the success of the QC microbiology laboratory. Microbiological examination of nonsterile products are described in two chapters in the United States and European Pharmacopoeia; Microbial Enumeration Test and Test for Specified Microorganisms. As an effort to harmonize the various pharmacopoeias the harmonized Sterility Test incorporates requirements for regular sterility testing of media. The harmonized Test For Specified Micro Organisms (Microbial Limit Test) now incorporates the evaluation of nutritive, differential and selective properties of the media.

All media supplied by HiMedia is checked for physical, chemical parameters and growth promotion.

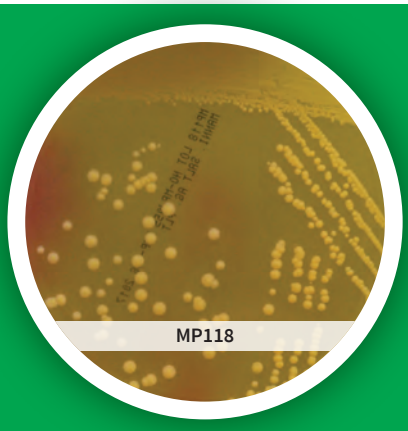
Microbiological examination of nonsterile products allows quantitative enumeration of mesophilic bacteria and fungi that may grow under aerobic conditions. These are primary tests and of simple design enabling counting the number of microorganism in terms of colony forming units (CFU) in representative nonsterile product or raw material. The usual procedure involves, immersing the sample into solution and then plate aliquots to determine the CFU/gram (or mL) of initial sample. In case the product cannot be put into solution, Most Probable Number method can be employed. The plating can be either pour plate, spread plate or the filtration of material and then placing the membrane filter on the surface of an agar plate. The membrane filtration method is generally used when there are few expected colony forming units in the sample to be tested as it is a good method to test a large volume of liquid, but can only count up to approximately 100 CFU/membrane.

Ready Prepared Media in Polystyrene Plates with ß Lactamase For Environmental Monitoring

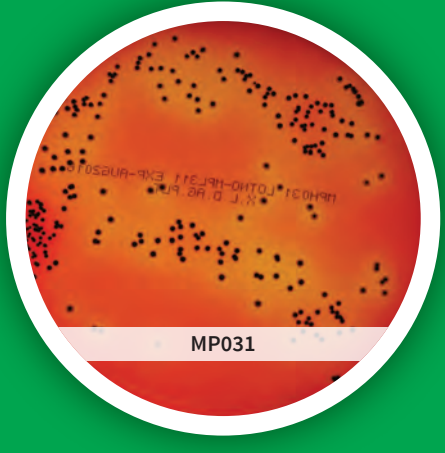


Ready™ PREPARED HIMEDIA

COMPREHENSIVE RANGE OF READY PREPARED MEDIA


MP118

- Products Manufactured under WHO GMP Norms
- Conforms Regulatory Needs
- Standard and Customized Media in Ready Prepared Form
- Complete Solutions for pharmaceutical Diagnostics


MP031

Baird Parker Agar Base M043

Intended Use:

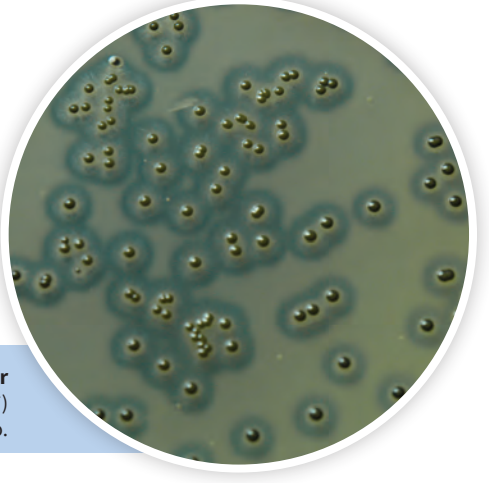
Baird Parker Agar Base with supplements is recommended for the isolation and enumeration of coagulase positive staphylococci from pharmaceutical ingredients and finished products as specified under microbial limit tests. It is also recommended for microbiological examination of food, nutritional and dietary supplements.

Directions:

Suspend dehydrated media as per table in 950 ml / 940 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize the media as specified below or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates. Add the supplement as specified below.

Ingredients	HiMedia	Granulated	USP	EP	BP	IP	HiVeg™	Chemically defined
	M043	GM043	MU043	ME043	M043B	MM043	MV043	MCD043
Tryptone#	_	_	10.00	10.00	10.00	10.00	_	_
Tryptone	10.00	10.00	_	_	_	_	_	_
HM Peptone B##	5.00	5.00	5.00	5.00	5.00	5.00	_	_
HiVeg™ hydrolysate	_	_	_	_	_	_	10.00	_
HiVeg™ extract	_	_	_	_	_	_	5.00	_
HiCynth™ Peptone No.2###	_	_	_	_	_	_	_	10.00
HiCynth™ Peptone No.7###	_	_	_	_	_	_	_	6.00
Yeast extract	1.00	1.00	1.00	1.00	1.00	1.00	1.00	_
Glycine	12.00	12.00	12.00	12.00	12.00	12.00	12.00	12.00
Sodium pyruvate	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00
Lithium chloride	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Agar	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00
Grams/litre	63.00	63.00	63.00	63.00	63.00	63.00	63.00	63.00
Final pH (at 25°C)	7.0±0.2	7.0±0.2	_	_	_	_	7.0±0.2	7.0±0.2
pH after sterilization	_	_	*6.8±0.2	*6.8±0.2	*6.8±0.2	6.8±0.2	_	-
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min
Supplements	I) FD045/ FD047 ii) FD046 iii) FD069 iv) FD195	I) FD045/ FD047 ii) FD046 iii) FD069 iv) FD195	i)FD045 ii) FD052	i)FD045 ii) FD052	i)FD045 ii) FD052	i)FD045 ii) FD052	I) FD045/ FD047 ii) FD046 iii) FD069 iv) FD195	I) FD045/ FD047 ii) FD046 iii) FD069 iv) FD195

* pH can also be measured after sterilization, at 25°C
Alternatively the medium can be sterilized by autoclaving at 115°C for 30 minutes.
Pancreatic digest of casein
Equivalent to Beef extract
Chemically defined peptones



MU043 Baird Parker Agar
Staphylococcus aureus subsp. *aureus* ATCC 6538 (00032*)
*Corresponding WDCM No.

Baird Parker Agar Base M043

Principle And Interpretation

This medium was first described in 1952. This medium was developed by Baird-Parker (1, 2) from the Tellurite-Glycine formulation of Zebovitz et al (3) for selective isolation of *Staphylococcus aureus* from foods. *Staphylococcus* species are common contaminants in food, dairy, pharmaceutical and cosmetics related products (9). This medium is recommended for microbial limit tests of non-sterile pharmaceutical products and to detect *S.aureus*. Baird Parker Agar Medium was reported to be the best medium for selective detection of coagulase positive and enterotoxigenic *Staphylococcus* (4). This medium was found to be less inhibitory to *S.aureus* than other media, at the same time medium is being more selective (5, 6, 8). Subsequently it was officially adapted by the AOAC (9) This medium is also recommended by various Pharmacopoeia for use in Microbial limit test (7, 10, 11, 12).

HM Peptone B, yeast extract, Tryptone, HiVeg™ extract, HiVeg™ hydrolysate and HiCynth™ peptones provides essential carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins, minerals and other essential nutrient substances and other growth requirements. Sodium pyruvate protects injured cells and helps recovery. Lithium chloride and potassium tellurite inhibit most of contaminating microflora except *S.aureus*. Glycine, pyruvate enhances growth of *Staphylococcus*. With the addition of egg yolk the medium becomes yellow and opaque.

Proteolytic bacteria produce a clear zone around colony in egg yolk containing media also known as Lecithinase reaction. A clear zone and grey-black colonies on this medium are diagnostic for coagulase positive Staphylococci.

Upon further incubation, an opaque zone is developed around colonies, which can be due to lipolytic activity. Identity of *Staphylococcus aureus* isolated on Baird-Parker Agar must be confirmed with a coagulase reaction.

Type of specimen

Clinical samples : Pus, blood; Food and dairy samples; Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples (M043 / GM043 / MV043 / MCD043) follow appropriate techniques for handling specimens as per established guidelines (16, 17).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (13, 14, 15).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (7, 10, 11, 12).

After use, contaminated materials must be sterilized by autoclaving before discarding

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Though the medium is recommended for detection of coagulase positive *Staphylococcus aureus*, other bacteria may grow.
2. Further biochemical test have to be performed for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM043 : Cream to yellow granular media

Gelling

Firm, comparable with 2.0% agar gel.

Colour and Clarity of prepared medium

Basal medium: Yellow coloured clear to slightly opalescent gel. After addition of Egg Yolk Emulsion and Tellurite Emulsion: Yellow coloured opaque gel forms in Petri plates.

pH

MU043/ME043/M043B/MM043-pH : 6.8±0.2

M043/GM043/MV043/MCD043 : 7.0+0.2

Growth Promotion Test

Growth Promotion is carried out in accordance USP. Cultural response was observed after an incubation at 30-35°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony	Lecithinase
Staphylococcus aureus subsp. aureus 6538 (00032*)	50 -100	good - luxuriant	≥50%	grey-black shiny	Positive, opaque zone around the colony
Additional Microbiological testing					
Staphylococcus aureus subsp. aureus 25923 (00034*)	50 -100	good - luxuriant	≥50	grey-black shiny	Positive, opaque zone around the colony
Proteus mirabilis 25933	50 -100	good - luxuriant	≥50%	brown - black	Negative
Micrococcus luteus 10240	50 -100	poor - good	30 -40%	shades of brown-black (very small)	Negative
Staphylococcus epidermidis 12228 (00036*)	50 -100	poor - good	30 -40%	black	Negative
Bacillus subtilis subsp. spizizenii 6633 (00003*)	50 -100	none - poor	0 -10%	dark brown matt	Negative
Escherichia coli 8739 (00012*)	50 -100	none - poor	0 -10%	large brown black	Negative
Escherichia coli 25922 (00013*)	50 -100	none - poor	0 -10%	large brown black	Negative
Escherichia coli NCTC 9002	50 -100	none - poor	0 -10%	large brown black	Negative

Key : * : Corresponds to WDCM number

Baird Parker Agar Base M043

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (16, 17).

Reference

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16. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
17. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media Plates			
MP043	Baird Parker Agar Plate (90 mm)	for the isolation and enumeration of coagulase positive Staphylococci from food and clinical sample.	20 PT / 50 PT
MP043L	Baird Parker Agar Plate (150 mm)	for the isolation and enumeration of coagulase positive Staphylococci from food and clinical sample.	20 PT

Bismuth Sulphite Agar M027

Intended Use:

Bismuth Sulphite Agar is recommended for the selective isolation and preliminary identification of *Salmonella* Typhi and other *Salmonellae* from pathological materials, sewage, water supplies, food etc. This medium is also recommended for carrying out microbial limit tests of pharmaceutical raw materials as well as finished preparations.

Directions:

Suspend dehydrated medium in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. DO NOT STERILIZE IN AUTOCLAVE or by fractional sterilization, since overheating may destroy the selectivity of the medium. The sensitivity of the medium depends largely upon uniform dispersion of precipitated bismuth sulphite in the final gel, which should be dispersed before pouring into sterile Petri plates.

For MM027, Sterilize Part A by autoclaving at (115°C) for 30 minutes or as per validated cycle. Part B: Heat to boiling. Cool to room temperature. Add 1 volume of Part B to 10 volume of Part A previously melted and cooled to a temperature of 55°C and pour into sterile Petri plates. The medium should be stored at 2-8°C for 5 days before use.

Ingredients	HiMedia	Granulated	USP	IP	HiVeg™	Chemically defined
	M027	GM027	MU027	MM027**	MV027	MCD027
				Part-A		
Tryptone	_	_	5.00	_	_	_
Peptone	10.00	10.00	5.00	10.00	_	_
HM Peptone B#	5.00	5.00	5.00	6.00	_	
HiVeg™ peptone	_	_	_	_	10.00	_
HiVeg™ extract	_	_	_	_	5.00	_
HiCynth™ Peptone No. 2##	_	_	_	_	_	10.00
HiCynth™ Peptone No. 6##	_	_	_	_	_	5.00
Dextrose (Glucose)	5.00	5.00	5.00	_	5.00	5.00
Sodium phosphate	_	_	4.00	_	_	_
Disodium hydrogen phosphate	4.00	4.00	_	_	4.00	4.00
Ferric citrate	_	_	_	0.40	_	_
Ferrous sulphate	0.30	0.30	0.30	_	0.30	0.30
Bismuth sulphite indicator	8.00	8.00	8.00	_	8.00	8.00
Brilliant green	0.025	0.025	0.025	0.01	0.025	0.025
Agar	20.00	20.00	20.00	24.00	20.00	20.00
Sterilization	_	_	_	Maintaining at 115°C-30 min	_	_
				Part-B		
Ammonium bismuth citrate	_	_	_	3.00	_	_
Sodium sulphite	_	_	_	10.00	_	_
Anhydrous, disodium hydrogen phosphate	_	_	_	5.00	_	_
Dextrose monohydrate	_	_	_	5.00	_	_
Grams/litre	52.33	52.33	52.32	62.95• Part A: 40.4 Part B: 22.54	52.33	52.33
Final pH (at 25°C)	7.7±0.2	7.7±0.2	*7.6±0.2	_	7.7±0.2	7.7±0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling

**Available in twin packs, lValue represent combination of Part A and Part B
*pH can also be measured after heating at 25°C
Equivalent to Beef extract
Chemically defined peptones

Bismuth Sulphite Agar M027

Principle And Interpretation

Bismuth Sulphite Agar Medium is prepared in accordance with USP (1). It is employed for the isolation and preliminary identification of *Salmonella* Typhi and other *Salmonellae* from pathological materials, sewage, water, food, pharmaceuticals and other products. Bismuth Sulphite Agar is recommended by various Associations (2, 3, 4, 5, 6). It is a modification of Wilson and Blair medium (7, 8, 9). This medium is also recommended by Indian pharmacopoeia (11).

Brilliant green and bismuth sulphite incorporated into the medium inhibit the intestinal gram-negative and gram-positive bacteria, Peptone, Tryptone, HM Peptone B, HiVeg™ peptone, HiVeg™ extract and HiCynth™ peptone are rich source of carbonaceous, nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances for growth of the organism. The fermentable source of carbohydrate in this medium is dextrose (Glucose), which provides energy for enhanced microbial growth. Phosphates incorporated in the medium act as a good buffering agent. The bismuth ions are reduced to metallic bismuth, which impart the metallic sheen around the colonies. Sulphite is reduced to black ferric sulphide giving the black colour with release of H_2S.

Salmonella Enteritidis and *Salmonella* Typhimurium typically grow as black colonies (rabbit eye colonies) with a surrounding metallic sheen. *Salmonella* ParaTyphi A grow as light green colonies. This medium also favors use of larger inoculum and heavily contaminated samples as compared to other selective media, as it has unique inhibitory action towards gram-positive and coliform organisms. The medium may be inhibitory to some strains of *Salmonella* species and therefore should not be used as the sole selective medium for these organisms. *Shigella* species are mostly inhibited on this medium and also some *Salmonellae* like *S.* Sendai, *S.* Berta, *S.* Gallinarum, *S.* Abortus-equally are inhibited. *Proteus* species are inhibited but few strains give dull green or brown colonies with metallic sheen.

Type of specimen

Clinical samples : faeces, urine, blood and other pathological material, Food and dairy samples, water samples, pharmaceutical samples.

Specimen Collection and Handling

For clinical samples (M027 / GM027 / MV027 / MCD027) follow appropriate techniques for handling specimens as per established guidelines (5, 12).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (4, 6, 10).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (3).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1, 11).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. DO NOT AUTOCLAVE OR OVERHEAT THE MEDIUM, as it destroys the selectivity of the medium.
2. *S.*Typhi and *S.*Arizonae exhibit typical brown colonies, with or without metallic sheen.
3. This medium is highly selective and must be used in parallel with less selective media for isolation.
4. With certain *Salmonella* species, typical black colonies with metallic sheen is observed near heavy inoculation and isolated colonies may show green colonies.
5. *Shigella* species are mostly inhibited on this medium; exceptions being *S. flexneri* and *S. sonnei* (14)
6. Some *Salmonella* like *S.* Sendai, *S.* Berta, *S.* Gallinarum, *S.* Abortus-equi are also inhibited (14).

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to greenish yellow homogeneous free flowing powder

GM027 : Light yellow to greenish yellow granular media

MM027: Part A - Light yellow to greenish yellow free flowing powder,

Part B - White to cream homogeneous free flowing powder

Gelling

Firm, comparable with 2.0% agar gel of M027/GM027/MV027/MU027/MCD027 and 2.4% Agar gel of MM027

Colour and Clarity of prepared medium

Yellow to greenish yellow opalescent gel with flocculant precipitate forms in Petri plates.

pH

MU027 - pH : 7.6 ± 0.2

M027/GM027/MV027/MCD027 - pH : 7.7 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with USP/IP. Cultural response was observed after an incubation at 30-35°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Bismuth Sulphite Agar M027

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Salmonella Abony NCTC 6017 (00029*)	50 -100	good - luxuriant	≥50%	black with metallic sheen
Salmonella Typhimurium 14028 (00031*)	50 -100	good - luxuriant	≥50	black with metallic sheen
Additional Microbiological testing				
#Klebsiella aerogenes 13048 (00175*)	50 -100	none-poor	≤10%	brown-green (depends on the inoculum density)
Enterococcus faecalis 29212 (00087*)	≥10^3	inhibited	0%	
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	≥50%	black with metallic sheen
Salmonella Typhi 6539	50 -100	luxuriant	≥50%	black with metallic sheen
Shigella flexneri 12022 (00126*)	50 -100	none - poor	≤10%	brown
Escherichia coli 8739 (00012*)	50 -100	none - poor	≤10%	Brown to green, depends on inoculum density

Key : * : Corresponds to WDCM number
: Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 12).

Reference

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MM027 Bismuth Sulphite Agar
Salmonella Typhimurium ATCC 14028 (00031*)
*Corresponding WDCM No.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : DriFilter™ Membrane Nutrient Pad Media			
MF005	Bismuth Sulphite Medium (without Membrane Filter)	for detection and enumeration of *Salmonella.*	50 PT
MF005E	Bismuth Sulphite Medium (Economy Pack) (without Membrane Filter)	for detection and enumeration of *Salmonella.*	50 PT

Brilliant Green Agar Base, Modified M016

Intended Use:

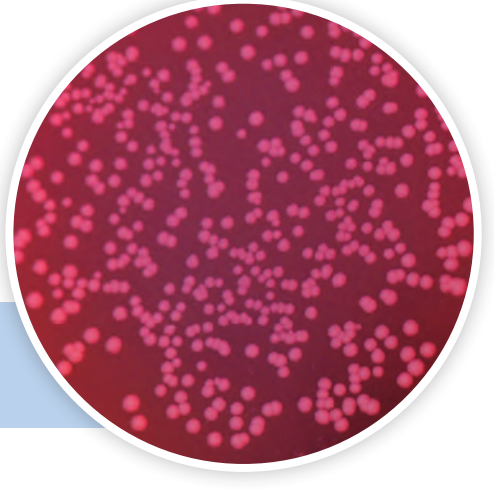
Brilliant Green Agar Medium is used for selective isolation of *Salmonellae* other than *Salmonella* Typhi from faeces, foods, dairy products and various excipients and finished products used in pharmaceutical preparations.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving 15 lbs (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. For M016, MV016 and MCD016 aseptically add rehydrated contents of 2 vials of Sulpha Supplement (FD068). Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	USP	EP	BP	IP	HiVeg™	Chemically defined
	M016	MU016	ME016	M016B	MM016	MV016	MCD016
HMC Peptone#	_	_	10.00	10.00	_	_	_
Peptone##	_	5.00	_	_	_	_	_
Tryptone###	_	5.00	_	_	_	_	_
Peptone	_	_	_	_	10.00	_	_
Proteose peptone	10.00	_	_	_	_	_	_
HiVeg™ peptone No. 3	_	_	_	_	_	10.00	_
Yeast extract	3.00	3.00	3.00	3.00	3.00	3.00	_
HiCynth™ Peptone No.5####	_	_	_	_	_	_	3.00
HiCynth™ Peptone No.4####	_	_	_	_	_	_	10.00
Lactose, monohydrate	_	_	10.00	10.00	_	_	_
Lactose	10.00	10.00	_	_	10.00	10.00	10.00
Sucrose	10.00	10.00	10.00	10.00	10.00	10.00	10.00
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Phenol red	0.08	0.08	0.08	0.08	0.08	0.08	0.08
Brilliant green	0.0125	0.0125	0.0125	0.0125	0.0125	0.0125	0.0125
Agar	20.00	20.00	20.00	20.00	12.00	20.00	20.00
Grams/litre	58.09	58.09	57.59	57.59	50.09	58.09	58.09
Final pH (at 25°C)	6.9±0.2	_	_	_	_	6.9±0.2	6.9±0.2
pH after sterilization	_	*6.9±0.2	6.9±0.2	6.9±0.2	6.9±0.2	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validaed cycle	Autoclaving 121°C-15 min or as per validaed cycle	Autoclaving 121°C-15 min or as per validaed cycle	Autoclaving 121°C-15 min or as per validaed cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min
Supplements	FD068	_	_	_	_	FD068	FD068

*pH can also be measured after sterilization at 25°C
Peptones (meat and casein)
Peptic digest of animal tissue
Pancreatic digest of casein
Chemically defined peptones.



M016 Brilliant Green Agar Base
Salmonella Typhimurium 14028 (00031*)
*Corresponding WDCM No.

Brilliant Green Agar Base, Modified M016

Principle And Interpretation

Brilliant Green Agar medium is recommended as a primary plating medium for isolation of *Salmonella* species was first described by Kristensen *et al* as medium for differentiation of paratyphoid B from other Gram negative enteric bacteria (1). Kauffmann further modified it for isolation of *Salmonella* from stool samples (2). Brilliant green agar is also recommended by APHA (3, 4, 10, 11, 12) FDA (5) this medium is recommended by various pharmacopoeia for isolation of *Salmonella* species (6, 7, 8, 9). This medium is employed in testing clinical specimens. Heavy inocula and heavily contaminated samples can be analyzed due to the outstanding selectivity of this medium. Brilliant Green Agar is used in the microbial limits test and with novobiocin for testing food and pharmaceutical products.

Combination of Peptone, Tryptone, Proteose peptone, HMC peptone, HiVeg™ peptone no 3, Yeast extract and HiCynth™ peptones makes the medium highly nutritious and supplies nitrogeneous and carbonaceous compounds, amino acids and long chains of peptides. Sodium chloride maintains the osmotic equilibrium.

Lactose and sucrose are the fermentable carbohydrate sources. Phenol red serves as an acid base indicator giving yellow colour to lactose and or sucrose fermenting bacteria. This medium also contains brilliant green, which inhibits growth of majority of Gram-negative and Gram-positive, bacteria. *Salmonella* Typhi, *Shigella* species, *Escherichia coli*, *Proteus* species, *Pseudomonas* species, *Staphylococcus aureus subsp. aureus* are mostly inhibited.

Clinical specimens can be directly plated on this medium. However, being highly selective, it is recommended that this medium should be used along with a less inhibitory medium to increase the chances of recovery. Often cultures enriched in Selenite or Tetrathionate Broth are plated on Brilliant Green Agar along with Bismuth Sulphite Agar, SS Agar, MacConkey Agar. Non-lactose fermenting bacteria develop white to pinkish red colonies within 18-24 hours of incubation. *Salmonella* Typhi and *Shigella* species may not grow on this medium, moreover *Proteus*, *Pseudomonas* and *Citrobacter* species may mimic enteric pathogens by producing small red colonies.

Type of specimen

Clinical : faeces; Food and dairy samples; Water samples; Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (13, 14).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3, 4, 10, 11).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (12).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (6, 7, 8, 9).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Though this medium is selective for *Salmonella* other species of *Enterobacteriaceae* may grow.
2. *Salmonella* Typhi and *Shigella* species may not grow on this medium.
3. Being highly selective, it is recommended that this medium should be used along with a less inhibitory medium to increase the chances of recovery.
4. Moreover *Proteus*, *Pseudomonas* and *Citrobacter* species may mimic enteric pathogens by producing small red colonies.
5. Further confirmation has to be carried out on presumptive *Salmonella* isolates.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to light pink homogeneous free flowing powder

Gelling

Firm, comparable with 2.0% agar gel of MU016/M016/ME016/M016B/MV016/MCD016 and 1.2% Agar gel of MM016

Colour and Clarity of prepared medium

Greenish brown clear to slightly opalescent gel forms in Petri plates

pH

6.90 ± 0.2

Growth Promotion Test

Growth Promotion was observed in accordance with USP/EP/BP/IP, after an incubation at 30-35°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Salmonella Typhimurium 14028 (00031*)	50 -100	good-luxuriant	≥50%	pinkish white
Salmonella Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	≥50%	pinkish white
Additional Microbiological testing				
Salmonella Enteritidis 13076 (00030*)	50 -100	good-luxuriant	≥50%	pinkish white
Salmonella Typhi 6539	50 -100	fair-good	30 -40%	reddish
Escherichia coli 25922 (00013*)	50 -100	none-poor	<10%	yellowish green
Escherichia coli 8739 (00012*)	50 -100	none-poor	<10%	yellowish green
Escherichia coli NCTC 9002	50 -100	none-poor	<10%	yellowish green
Staphylococcus aureus subsp. aureus 25923 (00034*)	$\geq 10^3$	inhibited	0%	
S. aureus subsp. aureus 6538 (00032)*	$\geq 10^3$	inhibited	0%	

Key : * : Corresponds to WDCM number

Brilliant Green Agar Base, Modified M016

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20-30°C. For M016 / MV016 / MCD016 preprared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (13, 14).

Reference

1. Kristensen M., Lester V, and Jurgens A., 1925, Brit.J.Exp.Pathol., 6:291.
2. Kauffman F., 1935, Seit F. Hyg. 177:26
3. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
4. Standard Methods for the Microbiological Examination of Dairy Products, 1995, 19th Ed, APHA, Washington, D.C.
5. Bacteriological Analytical Manual, 8th Ed, 1998, AOAC, Washington D.C.
6. The European Pharmacopoeia, 2008, Council or Europe, Strasbourg.
7. The British Pharmacopoeia, 2008, vol. II, London.
8. Indian Pharmacopoeia, 2010, Ministry of Health and Family Welfare, Govt., of India.
9. United States Pharmacopoeia, 2019, U. S. Pharmacopoeial Convention, Inc., Rockville, MD.
10. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
11. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
12. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
13. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
14. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media Plates			
MP016	Brilliant Green Agar Modified Plate	for selective isolation of *Salmonellae* other than *Salmonella* Typhi from faeces, food, dairy products.	10 PT / 50 PT
Category : Ready Prepared Solid Media in Glass Bottles			
SM016C	Brilliant Green Agar, Modified	for selective isolation of *Salmonellae* other than *Salmonella* Typhi from faeces, food & dairy products	10X100ML

Deoxycholate Citrate Agar M065

Intended Use:

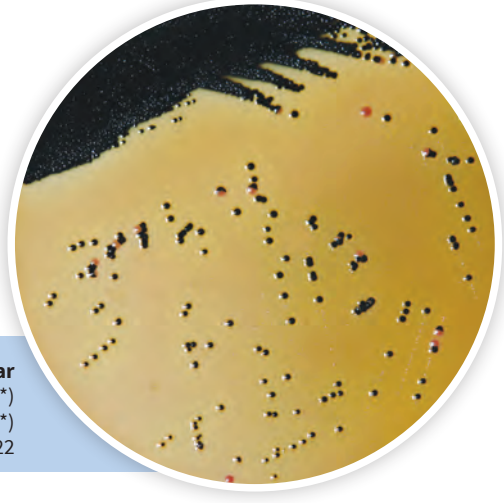
Deoxycholate Citrate Agar is a selective medium recommended for the isolation of enteric pathogens particularly *Salmonella* and *Shigella* species as per the guideline of microbial limit test.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified / distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Avoid excessive heating as it is detrimental to the medium. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	Granulated	EP	BP	IP	HiVeg™	Chemically defined
	M065	GM065	ME065	M065B	MM065	MV065	MCD065
HM Peptone#	—	—	10.00	10.00	—	—	—
Peptone	—	—	—	—	10.00	—	—
Proteose peptone	10.00	10.00	—	—	—	—	—
HM Peptone B###	—	—	10.00	10.00	10.00	—	—
HI, solids##	10.00	10.00	—	—	—	—	—
HiVeg™ peptone No. 3	—	—	—	—	—	13.00	—
HiVeg™ infusion	—	—	—	—	—	10.00	—
HiCynth™ Peptone No.2####	—	—	—	—	—	—	12.00
HiCynth™ Peptone No.6####	—	—	—	—	—	—	11.00
Lactose, monohydrate	—	—	10.00	10.00	10.00	—	—
Lactose	10.00	10.00	—	—	—	10.00	10.00
Trisodium citrate	—	—	—	—	20.00	—	—
Synthetic detergent No. III	—	—	—	—	—	2.00	—
Synthetic detergent	—	—	—	—	—	—	2.00
Sodium citrate	20.00	20.00	20.00	20.00	—	20.00	20.00
Ferric ammonium citrate	2.00	2.00	—	—	—	2.00	2.00
Ferric citrate	—	—	1.00	1.00	1.00	—	—
Sodium deoxycholate	5.00	5.00	5.00	5.00	5.00	—	—
Neutral red	0.02	0.02	0.02	0.02	0.02	0.02	0.02
Agar	13.50	13.50	13.50	13.50	13.50	13.50	13.50
Grams/litre	70.52	70.52	69.02	69.02	69.02	70.52	70.52
Final pH (at 25°C)	7.5±0.2	7.5±0.2	—	—	—	7.5±0.2	7.5±0.2
pH after heating	—	—	7.3±0.2	7.3±0.2	7.3±0.2	—	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling

Meat peptone
Heart infusion, solids
Equivalent to Beef extract
Chemically defined peptones



M065 Deoxycholate Citrate Agar
1. Salmonella Typhimurium 14028 (00031*)
2. Escherichia coli 25922 (00013*)
3. Shigella flexneri 12022

Deoxycholate Citrate Agar M065

Principle And Interpretation

Deoxycholate Citrate Agar is prepared as per the modified formula of Leifson (1). This medium is used for the isolation and maximum recovery of intestinal pathogens belonging to *Salmonella* and *Shigella* groups from foods products (2). However, it is recommended to use less inhibitory medium when *Shigella* have to be isolated (3). This medium is recommended by various Pharmacopoeia (4, 5, 6, 7). *Salmonella*, major causative agent of enteric disease especially food borne toxic infection and typhoid was first observed by Eberth in 1880. This medium is routinely used to check the presence of *Salmonella* contamination in food and pharmaceutical products. *Proteus* and other Gram positive organisms are inhibited due to higher concentration of both citrate and deoxycholate salts in this medium. Sodium deoxycholate at pH 7.3 to 7.5 is inhibitory for gram-positive bacteria. Sodium thiosulphate also helps in reactivation of sulphur containing compounds and prevents the desiccation of these compounds during storage. It also forms the substrate for enzyme thiosulphate reductase, which breaks it to form H_2S. H_2S then reacts with Fe ions in the medium and produces black FeS precipitate. This gives the indicative appearance of colonies with black center. Sodium thiosulphates are also inactivators of halogens and can minimize its toxicity in the testing sample, if any during microbial limit tests. Citrate salt, concentration included in the formulation, are inhibitory to gram positive bacteria and most other normal intestinal organisms.

Combination of HM Peptone B, Peptone, Proteose peptone, HiVeg™ peptone No 3, HM Peptone, HI solids, HiVeg™ infusion and HiCynth™ peptones supplies carbonaceous, nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances required for enhanced growth. Lactose, Lactose monohydrate supplies fermentable carbohydrate source in this medium. Neutral red acts as indicators, in presence of which lactose fermenters like coliform bacteria give pink colonies while lactose non-fermenters give colourless colonies.

Salmonella gives either colourless and opaque colonies with or without black center, while *Shigella* gives colourless colonies without black center indicating absence of H_2S production. Precipitation of deoxycholate by acid produced by lactose fermenters may give a zone of precipitation around the colony. This medium provides essential growth factors for growth of several auxotrophic strains of ParaTyphi and Typhi. The selectivity of this medium permits the use of fairly heavy inocula without danger of overgrowth of the *Shigella* and *Salmonella* by other microflora. For the routine examination of stool and urine specimens, it is suggested that other media such as MacConkey Agar, Bismuth Sulphite Agar etc. be used in conjunction with this medium.

Type of specimen

Clinical- faeces, Urine, Foods samples, Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (2).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (4, 5, 6, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further biochemical identification is required for confirmation of species.
2. Due to nutritional variations some organisms may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature

Quality Control

Appearance

Light yellow to pinkish beige homogeneous free flowing powder

GM065 : Light yellow to pinkish beige granular media

Gelling

Firm, comparable with 1.35% Agar gel.

Colour and Clarity of prepared medium

Reddish orange coloured clear to slightly opalescent gel forms in Petri plates.

pH

M065/GM065/MV065/MCD065 - 7.3-7.7

MM065/ME065/M065B - 7.1-7.5

Growth Promotion Test

Growth Promotion is carried out in accordance with (USP/EP/BP/IP). Cultural response was observed after an incubation at 36-38°C for 18-24 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Deoxycholate Citrate Agar M065

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Test for specified microorganism				
Salmonella Typhimurium 14028 (00031*)	50 -100	good-luxuriant	≥50%	Colourless and opaque with or without black centres
Salmonella Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	≥50	Colourless and opaque with or without black centres
Additional microbiological testing				
Salmonella Enteritidis 13076 (00030*)	50 -100	good-luxuriant	≥50%	Colourless and opaque with or without black centres
Salmonella Typhi 6539	50 -100	good-luxuriant	≥50%	Colourless and opaque with or without black centres
Escherichia coli NCTC 9002	50 -100	none-poor	0 -10%	Pink with bile precipitate
Escherichia coli 8739 (00012*)	50 -100	none-poor	0 -10%	Pink with bile precipitate
Shigella flexneri 12022 (00126*)	50 -100	none-poor	0 -10%	colourless
Enterococcus faecalis 29212 (00087*)	$\geq 10^3$	inhibited	0%	

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. Leifson, 1935, J. Path. Bact., 40:581.
2. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th ed., APHA, Washington, D.C.
3. Frieker C.R., 1987, J. Appl. Bact., 63:99.
4. Indian Pharmacopoeia, 2007, Govt. of India, Ministry of Health and Family Welfare, New Delhi, India.
5. European Pharmacopoeia, 2008, European Dept. for the quality of Medicines.
6. British Pharmacopoeia, 2008, The Stationery office British Pharmacopoeia
7. The United States Pharmacopoeia, 2019. USP Conv. Rockville, MD.
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media Plates			
MP065	Deoxycholate Citrate Agar Plate	selective medium for the isolation of enteric pathogens particularly *Salmonella* and *Shigella* species.	10 PT / 50 PT

EMB Agar, Levine M022

Intended Use:

EMB Media (Eosin Methylene Blue Agar) is recommended for the isolation and differentiation of gram negative enteric bacteria from clinical and non-clinical specimens. It is used for differentiation of *Escherichia coli* and *Klebsiella aerogenes*, as well as for rapid identification of *Candida albicans*. It is also recommended for bacteriological testing of dietary and nutritional supplements as well as for carrying out microbial limit test for various pharmaceutical raw material and finished products.

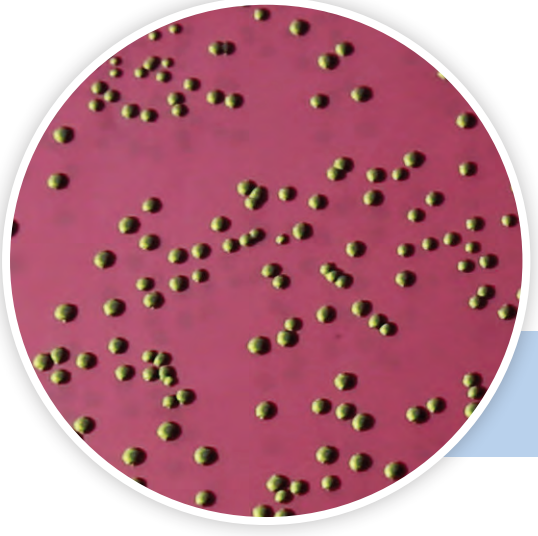
Directions:

Suspend dehydrated medium as per table in 1000 ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. AVOID OVERHEATING. Cool to 45-50°C and shake the medium in order to oxidize the methylene blue (i.e. restore its blue colour) and to suspend the flocculent precipitate.

***Note** : Store the medium away from light to prevent photo-oxidation.*

Ingredients	HiMedia	Granulated	USP	IP	HiVeg™
	M022	GM022	MU022	MM022	MV022
Gelatin peptone#	—	—	10.00	10.00	—
Peptone	10.00	10.00	—	—	—
HiVeg™ peptone	—	—	—	—	10.00
Dipotassium hydrogen phosphate	2.00	2.00	2.00	2.00	2.00
Lactose	10.00	10.00	10.00	10.00	10.00
Eosin - Y	0.40	0.40	0.40	0.40	0.40
Methylene blue	0.065	0.065	0.065	0.065	0.065
Agar	15.00	15.00	15.00	15.00	15.00
Grams/litre	37.46	37.46	37.46	37.46	37.46
Final pH (at 25°C)	7.1±0.2	7.1±0.2	—	—	7.1±0.2
pH after sterilization	—	—	*7.1±0.2	*7.1±0.2	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min or as per validaed cycle	Autoclaving 121°C- 15 min or as per validaed cycle	Autoclaving 121°C- 15 min

*pH can also be measured after sterilization at 25°C
Pancreatic digest of gelatin



M022 EMB Agar, Levine
Escherichia coli 25922 (00013*)
*Corresponding WDCM No.

EMB Agar, Levine M022

Principle And Interpretation

Levin Eosin Methylene Blue Agar was developed by Levine (1, 2) and is used for the differentiation of *Escherichia coli* and *Klebsiella aerogenes* and also for the rapid identification of *Candida albicans*. This medium is recommended for the detection, enumeration and differentiation of members of the coliform group by American Public Health Association (3, 4, 5). This medium is also recommended by IP & USP (6, 9).

Eosin-Y and methylene blue make the medium slightly selective and inhibit certain gram-positive bacteria. Both dyes act as indicator and inhibiting agent. These dyes differentiate between lactose fermenters and non-fermenters. Eosin Y and methylene blue forms a complex at acidic pH which acts as inhibiting agent. Some gram-positive bacteria such as faecal *Streptococci*, yeasts grow on this medium and form pinpoint colonies.

Gelatin peptone, Peptone, HiVeg™ peptone provide carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances for growth factors. Phosphates act as good buffering agent. *E.coli* forms colonies with green metallic sheen, indicating strong lactose fermentation.

Weld (7, 8) proposed the use of Levine EMB Agar, with added Chlortetracycline hydrochloride, for the rapid identification of *Candida albicans* in clinical specimens. A positive identification of *Candida albicans* can be made after 24-48 hours incubation at 35 - 37°C in 10% carbon dioxide atmosphere, from specimens such as faeces, oral and vaginal secretions and nail or skin scraping etc. However, the typical appearance is variable.

Type of specimen

Clinical samples - urine, faeces, oral and vaginal secretions, Food and dairy samples; Water samples, Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (10, 11).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (4, 5).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (3).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (6, 9).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. A non-selective medium should be inoculated in conjunction with EMB Agar.
2. Some strains of *Salmonella* and *Shigella* species do not grow in the presence of eosin and methylene blue.
3. Confirmatory tests should be further carried out for identification of isolated colonies.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature

Quality Control

Appearance

Light pink to purple homogeneous free flowing powder
GM022 : Light pink to purple granular media

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Reddish purple with greenish cast coloured opalescent gel with finely dispersed precipitate forms in Petri plates.

pH

7.10 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with (USP/IP) Cultural response was observed after an incubation at 36-38°C for 18-24 hours (IP) and 30-35°C for 24-48 hours (USP). Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar & Fungal growth on Sabouraud Dextrose Agar.

Cultural response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery Lot value (CFU)	Colour of colony
Test for specified microorganism				
Escherichia coli 8739 (00012*)	50 -100	good-luxuriant	≥50%	blue-black colonies with metallic sheen
Escherichia coli NCTC 9002	50 -100	good-luxuriant	≥50%	blue-black colonies with metallic sheen
Additional Microbiological testing				
Escherichia coli 25922 (00013*)	50 -100	good-luxuriant	≥50%	blue-black colonies with metallic sheen
#*Klebsiella aerogenes* 13048 (00175*)	50 -100	good-luxuriant	≥50%	pink to red
Salmonella Typhi*murium* 14028 (00031*)	50 -100	good-luxuriant	≥50%	colourless
Pseudomonas aeruginosa 9027 (00026*)	50 -100	good-luxuriant	≥50%	colourless
Enterococcus faecalis 29212 (00087*)	$\geq 10^3$	inhibited	0%	-
Staphylococcus aureus subsp. aureus 6538 (00032*)	$\geq 10^3$	inhibited	0%	-
Candida albicans 10231 (00054*)	50 -100	good-luxuriant	≥50%	colourless
Saccharomyces cerevisiae 9763 (00058*)	50 -100	none-poor	0 -10%	cream

Key : * : Corresponds to WDCM number

#Formerly known as *Enterobacter aerogenes*

EMB Agar, Levine M022

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (10, 11).

Reference

1. Levine M., 1918, J. Infect. Dis., 23:43.
2. Levine M., 1921, Bull. 62, Iowa State College Engr. Exp. Station.
3. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
4. Wehr H M and Frank J H., 2004, Standard Methods for the Examination of Dairy Products, 17th ed., APHA Inc., Washington, D.C.
5. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th ed., APHA, Washington, D.C.
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8. Weld J. T., 1953, Arch. Dermat. Syph., 67(5):433.
9. United States Pharmacopoeia, 2019, USP conv. rockville, MD.
10. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
11. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media Plates			
MP022	EMB Agar, Levine Plate	for isolation, enumeration and differentiation of members of *Enterobacteriaceae.*	20 pt / 50 pt

Fluid Selenite Cystine Medium (Twin pack) M025

Intended Use:

Fluid Selenite Cystine Medium / Broth is used as an enrichment medium for isolation of *Salmonellae* from faeces, urine or other pathological materials. Also recommended by various pharmacopoeias in carrying out microbial limit test as well as bacteriological testing of nutritional and dietary supplements.

Directions:

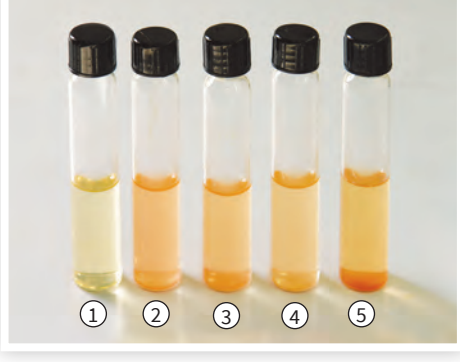
Suspend dehydrated medium as per table in 1000 ml of purified/ distilled water. Warm to dissolve the medium completely. Distribute in sterile test tubes. Sterilize in a boiling water bath or free flowing steam for 10 minutes or as per validated cycle. DO NOT AUTOCLAVE. Excessive heating is detrimental. Discard the prepared medium if large amount of selenite is reduced (indicated by red precipitate at the bottom of tube / bottle).

Instead of Part B: DD056 - Sodium Biselenite disc (1 disc per 10 ml of medium) or DB001 - Sodium Biselenite Bud (1 bud per 100 ml of medium) can be added to the medium after boiling.

[**Caution** : Sodium hydrogen selenite (Sodium bi-selenite) is very toxic, corrosive agent and causes teratogenicity. Handle with great care. Upon contact with skin, wash immediately with lot of water]

Ingredients	HiMedia	USP	IP	HiVeg™
	M025	MU025	MM025	MV025
	Part A	Part A	Part A	Part A
Tryptone#	—	5.00	5.00	—
Tryptone	5.00	—	—	—
HiVeg™ hydrolysate	—	—	—	5.00
Lactose	4.00	4.00	4.00	4.00
Sodium phosphate	10.00	10.00	10.00	10.00
L-Cystine	0.01	0.01	0.01	0.01
	Part B	Part B	Part B	Part B
Sodium hydrogen selenite	4.00	—	4.00	4.00
Sodium acid selenite	—	4.00	—	—
Sodium Biselenite Bud (DB001)	10 bud	10 bud	10 bud	10 bud
Sodium Biselenite Disc (DD056)	100 disc	100 disc	100 disc	100 disc
Grams/litre (Part A + Part B) (19.01 + 4.0)	23.01	23.01	23.01	23.01
Final pH (at 25°C)	7.0±0.2	—	*7.0±0.2	7.0±0.2
pH after heating	—	*7.0±0.2	—	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling

Though IP and USP has mentioned this medium as a single entity; due to corrosive nature of sodium acid /hydrogen selenite, HiMedia provides this medium as Twin pack only.
pH can also be measured after sterilization at 25°C
Pancreatic digest of casein



Fluid Selenite Cystine Medium (Selenite Cystine Broth) (Twin Pack)M025
1. Control
2. *Salmonella* Typhimurium ATCC 14028 (00031*)
3. *Salmonella* Enteritidis ATCC 13076 (00030*)
4. *Salmonella* Typhi ATCC 6539
5. *Escherichia coli* ATCC 25922 (00013*)
*Corresponding WDCM no.

Fluid Selenite Cystine Medium (Twin pack) M025

Principle And Interpretation

Selective inhibitory effects of selenite were first demonstrated by Klett (1). Guth (2) used it to isolate *Salmonella* Typhi. Leifson studied selenite and formulated a medium using selenite. Fluid Selenite Cystine Medium is a modification of Leifsons (3) formula with added cystine (4). The formulation corresponds to that recommended by AOAC (5) for the detection of *Salmonella* in foodstuff, particularly egg products. It is also recommended by APHA (6, 7, 14) USP & IP (8, 11). Selenite Cystine Broth is useful for detecting *Salmonella* in the non-acute stages of illness when organisms occur in the faeces in low numbers and for epidemiological studies to enhance the detection of low numbers of organisms from asymptomatic or convalescent patients (9). *Salmonella* are also injured during various food processing procedures, including exposure to low temperatures, sub-marginal heat, drying, radiation, preservatives or sanitizers, (10). Recovery of *Salmonella* involves pre-enrichment, selective enrichment and selective plating since *Salmonella* may be present in low numbers in food sample in a injured conditions. This medium is also recommended for ISO for detection of *Salmonella* from food and animal feeding stuff (15). Fluid Selenite Cystine Medium is used as selective enrichment medium for the cultivation of *Salmonella* species. This medium is formulated to allow the proliferation of *Salmonella* while inhibiting the growth of competing non-*Salmonella* organisms.

Tryptone , HiVeg™ hydrolysate provides carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances. Lactose is the fermentable carbohydrate and maintains the pH in medium as selenite is reduced by bacterial growth and alkali is produced. An increase in pH lessens the toxicity of the selenite and results in overgrowth of other bacteria. The acid produced by bacteria due to lactose fermentation serves to maintain a neutral pH. Phosphate maintains a stable pH and also lessens the toxicity of selenite. L-cystine is the reducing agent, improving the recovery of *Salmonella*.

Enriched broth is subcultured on solid medium. Do not incubate the broth longer than 24 hours as inhibitory effect of selenite reduces after 6 - 12 hours of incubation (10).

Inoculate the food sample into recommended pre-enrichment broth, and then transfer 1 ml of mixture to 10 ml of Fluid Selenite Cystine Medium and also to 10 ml Tetrathionate Broth. Incubate and subsequently subculture on to Bismuth Sulphite Agar, Xylose-Lysine-Deoxycholate Agar, Hektoen Enteric Agar.

Type of specimen

Clinical samples - urine, food and dairy samples, water samples, pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (12, 13).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (5, 6, 7).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (14).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (8, 11).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Being highly selective some organisms may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature

Quality Control

Appearance

Part A : Cream to light yellow homogeneous free flowing powder
Part B : White to cream homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured, clear to slightly opalescent solution of complete medium.

pH

7.0 ± 0.2

Fluid Selenite Cystine Medium (Twin pack) M025

Cultural response

Cultural characteristics observed after an incubation at 30-35°C for 18-48 hours when sub cultured on XLD Agar (MU031 / MM031) or Brilliant Green Agar Medium (MU016/MM016).

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Growth on Agar Medium L				
Salmonella Typhi*murium* 14028 (00031*)	50 -100	luxuriant	≥50%	pinkish white
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant	≥50	pinkish white
Additional Microbiological testing K				
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	≥50%	red with black centres
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	≥50%	red with black centres
Salmonella Abony NCTC 6017 (00029*)	50 -100	good - luxuriant	≥50%	red with black centres
Salmonella Typhi 6539	50 -100	good - luxuriant	≥50%	red with black centres
Escherichia coli 8739 (00012*)	50 -100	fair	20 -30%	yellow
Escherichia coli 25922 (00013*)	50 -100	fair	20 -30%	yellowish green
Escherichia coli NCTC 9002	50 -100	fair	20 -30%	yellowish green
Organism (ATCC)	**Inoculum (CFU)**	**Growth**	**Recovery**	**Colour of colony**
Growth on Agar Medium L				
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	≥50%	pinkish white
Salmonella Typhi 6539	50 -100	fair - good	30-40%	reddish
Escherichia coli 8739 (00012*)	50 -100	fair	20 -30%	yellowish green

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (12, 13).

Reference

1. Klett A., 1900, Zeitsch Fer Hyg. Und. Infekt., 33: 137.
2. Guth F., 1916, Zbl. Bakt. I. Orig., 77:487.
3. Leifson E., 1936, Am. J. Hyg., 24(2): 423.
4. North W. R. and Bartram M. T., 1953, Appl. Microbiol., 1:130.
5. FDA Bacteriological Analytical Manual, 2005, 18th Ed., AOAC, Washington, DC.
6. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., APHA, Washington, D.C.
7. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
8. The United States Pharmacopeia, 2019, USP, The United States Pharmacopeial Convention, Rockville, M.D.
9. Murray P. R., Baron E. J., Jorgensen J. H., Pfaller M. A., Yolken R. H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.
10. Chattopadhyay W. and Pilford J. N., 1976, Med. Lab. Sci., 33:191.11. Hartman P. A. and S. A., Munich, 1981, J. Food Pract., 44: 385-386
11. Indian Pharmacopeia, 2010, Govt. of India, The Controller of Publication, Delhi.
12. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
13. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
14. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
15. ISO 6579-1:2017, Microbiology of the food chain - Horizontal Method for the detection, enumeration and serotyping of *Salmonella* _ Part I Detection of *Salmonella* species.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Media in tubes			
LQ070 / LQ070V	Selenite Broth	an enrichment medium for isolation of *Salmonella* species from faeces, urine or other pathological materials.	LQ070-25X10ML/ LQ070-50X10ML/ LQ070V-25X5ML/ LQ070V-50X5ML

Fluid Casein Digest Soya Lecithin Medium (Twin pack) M117

Intended Use:

Fluid Casein Digest Soya Lecithin Medium is recommended for sanitary examination of surfaces. Also recommended as primary enrichment medium for various pharmaceutical raw materials and finished products as specified under microbial limit test. It is further recommended for microbiological examination of food products, nutritional and dietary supplements.

Directions:

Suspend dehydrated media as per table in 960ml purified/distilled water. Heat if necessary to dissolve the medium completely. Add 40ml of Part B. Mix well and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	USP	IP	HiVeg™
	M117	MU117	MM117	MV117
	Part A	Part A	Part A	Part A
Tryptone#	20.00	20.00	20.00	—
HiVeg™ hydrolysate	—	—	—	20.00
Soya lecithin / Soy lecithin*	5.00	5.00	5.00	5.00
	Part B	Part B	Part B	Part B
Polysorbate 20	40 ml	40 ml	40 ml	40 ml
Grams/litre (Part A + Part B)	65.00	65.00	65.00	65.00
Final pH (at 25°C)	7.3±0.2	—	—	7.3±0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min

* In case of USP
# Pancreatic digest of casein

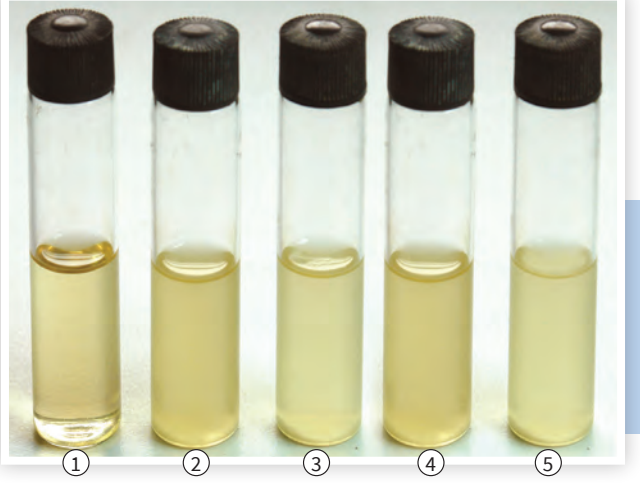
Principle And Interpretation

Fluid Casein Digest Soya Lecithin Medium is recommended by USP for use in Microbial Limit Tests (1) and by the Indian Pharmacopeia (2) for sanitary examination of surfaces. Weber and Black had described the importance of a highly nutritional goodmedium containing neutralizing agents for neutralizing quaternary ammonium compounds (3, 4). This medium is also recommended by NASA for the microbiological sampling of environmental surfaces sanitized with quaternary ammonium compounds (5). It is further recommended for microbiological examination of food products, nutritional and dietary supplements.

The medium contains Tryptone, HiVeg™ hydrolysate which provides nitrogenous, carbonaceous compounds, long chain amino acids vitamins and other essential nutrient substances for the growth of the organisms. Soya lecithin neutralizes the quaternary ammonium compounds while polysorbate 20 neutralizes phenolic disinfectants, hexachlorophene and formalin (6).

Type of specimen

Food and dairy samples; Water samples; Pharmaceutical samples.



Fluid Casein Digest Soya Lecithin Medium (Twin pack) M117
1. Control
2. *Escherichia coli* ATCC 25922 (00013*)
3. *Staphylococcus aureus* subsp. *aureus* ATCC 25923 (00034*)
4. *Bacillus subtilis* subsp. *spizizenii* ATCC 6633 (00003*)
5. *Candida albicans* ATCC 10231 (00054*)

*Corresponding WDCM no.

Fluid Casein Digest Soya Lecithin Medium (Twin pack) M117

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (9, 10, 11).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (12).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

Due to nutritional variations, some organisms may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Part A : Cream to yellow homogeneous free flowing powder
Part B : Colourless clear viscous liquid.

Colour and Clarity of prepared medium

Yellow coloured, clear solution without any precipitate.

pH

M117/ MV117 : 7.30 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours (for fungal species incubate at 25-30°C for 24-48 hrs).

Organism (ATCC)	Inoculum (CFU)	Recovery
Candida albicans 10231 (00054*)	50 -100	good-luxuriant
Bacillus subtilis subsp. spizizenii 6633 (00003*)	50 -100	good-luxuriant
Escherichia coli 25922 (00013*)	50 -100	good-luxuriant
Staphylococcus aureus subsp. aureus 25923 (00034*)	50 -100	good-luxuriant
Escherichia coli NCTC 9002	50 -100	good-luxuriant
Escherichia coli 8739 (00012*)	50 -100	good-luxuriant
Staphylococcus aureus subsp. aureus 6538 (00032*)	50 -100	good-luxuriant

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 15 - 25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7, 8).

Reference

1. The United States Pharmacopeia, 2019, The United States Pharmacopeial Convention. Rockville, MD.
2. Indian pharmacopoeia, 2010, Govt. of India, Ministry of Health and Family Welfare, Vol. II, Controller of Publications, New Delhi.
3. Weber and Black, 1948, Soap and Sanitary Chemicals, 24:134.
4. Weber and Black, 1948, Am. J. Public Health, 38:1405.
5. National Aeronautics and Space Administration, 1966, Standard Procedures for the Microbiological Examination of Space Hardware
6. Favero (chm.), 1967, Microbiological Sampling of Surfaces, Biological Contamination Control Committee, American Asso. for Contamination Control.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
9. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2018, Compendium of Methods for the Microbiological Examination of Foods, 5th ed., APHA, Washington, D.C.
10. Wehr H M and Frank J H., 2004, Standard Methods for the Examination of Dairy Products, 17th ed., APHA Inc., Washington, D.C.
11. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
12. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

GN Broth M242

Intended Use:

GN Broth is recommended for enrichment of *Shigella* from pharmaceutical products in accordance with the Indian Pharmacopoeia and for selective isolation of gram negative organisms from clinical specimen.

Directions:

Suspend dehydrated media as per table in 1000 ml purified / distilled water. Mix and allow to stand for 15 minutes. With continuous stirring, bring gently to boil and maintain at boiling point until completely dissolved. DO NOT AUTOCLAVE. AVOID EXCESSIVE HEATING. Dispense in sterile test tubes or flasks as desired.

Ingredients	HiMedia	Granulated	IP	HiVeg™
	M242	GM242	MM242	MV242
Tryptose	20.00	20.00	—	—
Polypeptone peptone	—	—	20.00	—
HiVeg™ hydrolysate No.1	—	—	—	20.00
Mannitol	2.00	2.00	—	2.00
Glucose (Dextrose)	1.00	1.00	1.00	1.00
Sodium citrate	5.00	5.00	2.00	5.00
Synthetic detergent No. III	—	—	—	0.50
Sodium deoxycholate	0.50	0.50	0.50	—
Di-potassium hydrogen phosphate	4.00	4.00	4.00	4.00
Potassium dihydrogen phosphate	1.50	1.50	1.50	1.50
Sodium chloride	5.00	5.00	5.00	5.00
Grams/litre	39.00	39.00	34.00	39.00
Final pH (at 25°C)	7.0 ± 0.2	7.0 ± 0.2	7.0 ± 0.2	7.0 ± 0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Boiling	Boiling	Boiling	Boiling

Principle And Interpretation

GN Broth is recommended by the Indian Pharmacopoeia (1) for the selective isolation of *Shigella* species with subsequent isolation on a selective medium, XLD Agar (MM031). Croft and Miller isolated more strains of *Shigella* from rectal swabs using this medium (2). Taylor and Schelhart showed the superiority of GN Broth to selenite enrichment media for isolation of *Shigella* (3). Hajna (4, 5) also suggested the enrichment of organisms from rectal swabs in this medium for 1-6 hours before plating on solid media.

The medium contains Polypeptone Peptone, Tryptose, HiVeg™ hydrolysate No. 1 which provides carbonaceous nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances to support bacterial growth. The combination of sodium citrate and sodium deoxycholate/ Synthetic detergent No. III inhibit gram-positive and some gram-negative bacteria such as coliforms. Phosphates serve as a buffering system. Sodium chloride maintains osmotic equilibrium. *Proteus*, *Pseudomonas* and coliforms do not over grow *Salmonella* and *Shigella* in GN Broth during the first 6 hours of incubation. This enrichment broth should be used in conjunction with selective and nonselective plating media to increase the probability of isolating pathogens (6, 7, 8).

Type of specimen

Clinical : faeces; Food samples; Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (9, 10).

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (6).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. It is an enrichment broth, so subculturing on selective or non selective media is required.
2. Overgrowth of *Proteus, Pseudomonas* and coliforms may occur, so subculturing within 8 hours of incubation is recommended.
3. Biochemical / serological confirmation is required for complete identification.

GN Broth M242

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder.
GM242 : Cream to yellow coloured granular media

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent solution in tubes.

pH

7.0 ± 0.2

Growth Promotion Test:

Growth Promotion is carried out in accordance with the Indian pharmacopoeia and cultural characteristics was observed after incubation at 30-35°C for 24-48 hours.

Growth Promoting properties:

Clearly visible growth of microorganism comparable to that obtained with previously tested and approved lot of the medium occurs at the specified temperature for the time specified inoculating ≤100 CFU (i.e. 30-35°C for ≤24 hours).

Inhibitory properties:

No growth of the test microorganism occurs at the specified temperature for not less than longest period of time specified inoculating ≥100 CFU (atleast 100 CFU) (at 30-35°C for ≥48 hours).

Cultural Response

Organism (ATCC)	Inoculum (CFU)	Recovery
Growth promoting		
Shigella boydii 9207	50 -100	good-luxuriant
Inhibitory		
Staphylococcus aureus subsp. *aureus 6538* (00032*)	$\geq 10^3$	inhibited

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 15 - 25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (9, 10).

Reference

1. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt. of India.
2. Croft C. C., Miller M. J., 1956, Am. J. Clin. Pathol., 26:411
3. Taylor W.I., Schelhart D., 1968, Appl. Environ. Microbiol., 16:1383.
4. Hajna A. A., 1955, Publ. Health Lab., 13:83.
5. Hajna A. A., 1956, Air. Univ. Sch. Ar. Med., USAF, 56:39
6. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
7. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
8. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
9. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
10. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

GN Broth (M242)
1. Control
2. *Shigella boydii* ATCC 9207
3. *Staphylococcus aureus subsp. aureus* ATCC 6538 (00032*)

*corresponding WDCM no.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Medium in Tubes			
LQ151	Medium 11. GN Broth	recommended for the enrichment of *Shigella* from pharmaceutical & clinical products in accordance with IP 2014.	50X10ml
Category : Ready Prepared Liquid Medium in Glass Bottles			
LQ157	GN Broth, Hajna	for the selective enrichment of Gram negative organisms of the enteric group from clinical & non clinical sample.	5X100ml
LQ151C	Medium 11.GN Broth	for the enrichment of *Shigella* from pharmaceutical products in accordance with IP 2014.	5X100ml

Hektoen Enteric Agar M467

Intended Use:

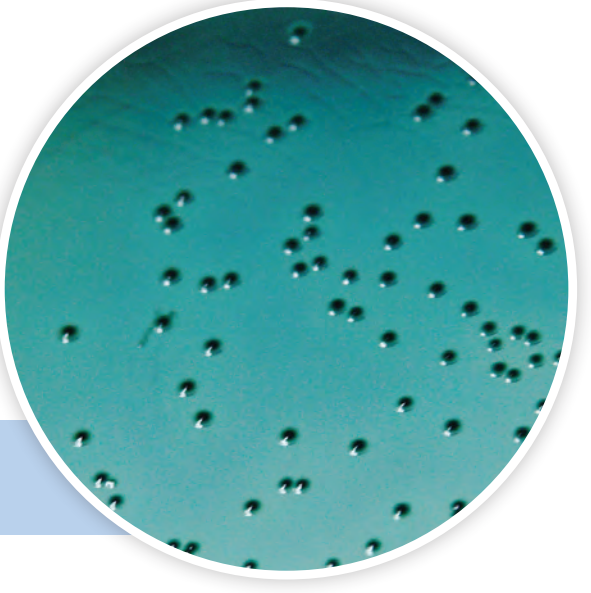
Hektoen Enteric Agar is recommended for differential and selective isolation of *Shigella* Hektoen Enteric Agar is recommended for differential and selective isolation of *Shigella* nutritional and dietary supplements, by USP.

Directions:

Suspend dehydrated media as per table, in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE.

Ingredients	HiMedia	Granulated	USP	HiVeg™	#Chemically defined
	M467	GM467	MU467	MV467	MCD467
Proteose peptone	12.00	12.00	—	—	—
Protease peptone	—	—	12.00	—	—
HiVeg™ peptone No. 3	—	—	—	19.00	—
HiCynth™ Peptone No.2#	—	—	—	—	19.00
HiCynth™ Peptone No.6#	—	—	—	—	3.00
Yeast extract	3.00	3.00	3.00	3.00	—
Lactose	12.00	12.00	12.00	12.00	12.00
Saccharose (Sucrose)	12.00	12.00	2.00	12.00	12.00
Salicin	2.00	2.00	9.00	2.00	2.00
Synthetic detergent No.1	—	—	—	2.00	2.00
Synthetic detergent	—	—	—	—	2.00
Bile salts mixture (equivalent to Bile salt No. 3)	9.00	9.00	9.00	—	—
Sodium chloride	5.00	5.00	5.00	5.00	5.00
Sodium thiosulphate	5.00	5.00	5.00	5.00	5.00
Ferric ammonium citrate	1.50	1.50	1.50	1.50	1.50
Acid fuchsin	0.10	0.10	0.10	0.10	0.10
Bromo thymol blue	0.065	0.065	0.065	0.065	0.065
Agar	15.00	15.00	14.00	15.00	15.00
Grams/litre	76.67	76.67	72.66	76.67	76.67
Final pH (at 25°C)	7.5±0.2	7.5±0.2	7.5±0.2	7.5±0.2	7.5±0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling	Boiling

Chemically defined peptones



M467 Hektoen Enteric Agar
Salmonella Typhimurium ATCC 14028 (00031*)
*Corresponding WDCM No.

Hektoen Enteric Agar M467

Principle And Interpretation

Hektoen Enteric Agar, a selective and differential medium designed to isolate and differentiate members of the species, *Salmonella* and *Shigella* from other *Enterobacteriaceae* and was developed by King and Metzger (1, 2). When compared with other selective medium, this medium inhibits the growth of *Salmonella* and *Shigella* very slightly; thus giving high yields of these microorganisms, but at the same time inhibits accompanying gram positive and other microorganisms. This medium is recommended by United States Pharmacopoeia, for testing the presence of *Salmonella* in dietary supplements (3). This medium is recommended in testing of *Salmonella* in food sample by various standards (4, 5, 6).

Compared to other differentiating media commonly used in clinical laboratories, Hektoen Enteric Agar is efficient in increasing the isolation rate of *Salmonella* sp. Bile salts, bromthymol blue and acid fuchsin inhibit the growth of most Gram positive organisms. Lactose, salicin and sucrose, serves as fermentable source of carbohydrates to encourage the growth and differentiation of enteric bacteria. In this medium by increasing the carbohydrate and peptone content of the medium, the inhibitory effect of bile salts and indicators are countered.

Proteose peptone, Protease peptone, HiVeg™ peptone No. 3, and HiCynth™ peptones provides carbonaceous, nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances required for organism growth. Yeast extract is a vitamin source. Sodium chloride maintains the osmotic balance of the medium. Sodium thiosulfate provides a source of sulfur. Hektoen Enteric Agar can also detect the production of hydrogen sulfide gas, which turns part of the medium black. Ferric ammonium citrate serves as iron source, which cause production of hydrogen sulfide from sodium thiosulphate and also aids in the visualization of hydrogen sulfide production by reacting with hydrogen sulfide gas to form a black precipitate.

Enterobacters that are capable of fermenting one or more of the carbohydrates produces yellow or salmon-orange coloured colonies like *Klebsiella pneumoniae*, that ferments lactose. Non-fermenters will produce blue-green colonies. Organisms that reduce sulfur to hydrogen sulfide will produce black colonies or blue-green colonies with a black center. *Salmonella* reduce sulfur to hydrogen sulfide, producing a black precipitate. *Micrococcus luteus* does not grow.

Type of specimen

Clinical : faeces, blood; Food and dairy samples; Water samples; Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (4, 5, 10).

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

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further incubation will improve differentiation between *Salmonella* and *Shigella*. *Proteus* species may resemble *Salmonella* or *Shigella*; hence further testing must be carried out for confirmation.
2. Since the medium is selective it must be used in conjunction with other media.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow with tancast homogeneous free flowing powder.

GM467 : Cream to yellow with tancast granular media

Gelling

Firm, comparable with 1.4% agar gel of MU467 or 1.5% Agar gel of M467/GM467/MV467

Colour and Clarity of prepared medium

Green coloured clear to slightly opalescent gel forms in Petri plates.

pH

7.50 ± 0.2

Cultural response

Cultural characteristics observed, after an incubation at 30-35°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Growth Promotion Test				
Salmonella Typhi*murium* 14028 (00031*)	50 -100	luxuriant	≥50%	blue-green with orwithout black centres
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant	≥50%	blue-green with or without black centres
Additional Microbiological testing				
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	≥50%	blue-green with or without black centres
Salmonella Typhi 6539	50 -100	fair-good	30 -40%	blue-green with or without black centres
Escherichia coli 25922 (00013*)	50 -100	none-poor	≤10%	orange (may have bile precipitate)
Escherichia coli 8739 (00012*)	50 -100	none-poor	≤10%	orange (may have bile precipitate)
Shigella flexneri 12022 (00126*)	50 -100	luxuriant	≥50%	greenish blue
Enterococcus faecalis 29212 (00087*)	$\geq 10^3$	inhibited	0%	-
Staphylococcus aureus subsp. aureus 6538 (00032*)	$\geq 10^3$	inhibited	0%	-

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Hektoen Enteric Agar M467

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20 - 30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the
hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. King, S., and W. I. Metzger. 1968. Appl. Microbiol. 16:577.
2. King, S., and W. I. Metzger. 1968. Appl Microbiol. 16:579.
3. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
4. Marshall, R. T. (ed.). 1993. Standard methods for the microbiological examination of dairy products, 16th ed. American Public Health Association, Washington, D.C.
5. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods For The Microbiological Examination of Foods, 5th ed., APHA, Washington, D.C.
6. AOAC, 2005, Bacteriological Analytical Manual, 18th ed., AOAC, Washington, DC.
7. Baird R.B., Eaton A. D., Rice E. W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 21st ed., APHA, Washington, D.C.
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
10. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Plates			
MP467	Hektoen Enteric Agar Plates	for differential and selective isolation of *Salmonella* and *Shigella* species from enteric pathological specimens.	10 pt / 50 pt
Category : Ready Prepared Dual Media for Blood Specimens in Glass Bottles			
LQ035	HiCombi™ Dual Performance Selective Medium - HEA	a qualitative test for rapid growth and confirmation of *Salmonella*. Combination of solid (7 ml) and liquid (20 ml) media in single bottle.	10bt
LQ035A	HiCombi™ Dual Performance Selective Medium - HEA	a qualitative test for rapid growth and confirmation of *Salmonella*. Combination of solid (20 ml) and liquid (40 ml) media in single bottle.	10bt

Lactose Broth M1003

Intended Use:

Lactose Broth is used for the detection of coliform bacteria in water, foods, dairy products as per Standard Methods. Also recommended by various pharmacopoeia to carry out microbial limit tests of pharmaceutical raw materials as well as finished products and preparations.

Directions:

Suspend dehydrated media as per table in 1000 ml purified/ distilled water. Heat if necessary to dissolve the medium completely. Dispense in tubes containing inverted fermentation vial (Durham's tube) as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	Granulated	USP	EP	BP	IP	HiVeg™
	M1003	GM1003	MU1003	ME1003	M1003B	MM1003	MV1003
Gelatin peptone#	—	—	5.00	5.00	5.00	5.00	—
Peptone	5.00	5.00	—	—	—	—	—
HM Peptone B##	3.00	3.00	3.00	3.00	3.00	3.00	—
HiVeg™ peptone	—	—	—	—	—	—	5.00
HiVeg™ extract	—	—	—	—	—	—	3.00
Lactose, monohydrate	—	—	—	5.00	5.00	5.00	—
Lactose	5.00	5.00	5.00	—	—	—	5.00
Grams/litre	13.00	13.00	13.00	12.75	12.75	12.75	13.00
Final pH (at 25°C)	6.9±0.2	6.9±0.2	—	—	—	—	6.9±0.2
pH after sterilization	—	—	*6.9±0.2	6.9±0.2	6.9±0.2	*6.9±0.2	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min

* pH can also be measured after sterilization at 25°C
# Pancreatic digest of gelatin
Equivalent to Beef extract

Principle And Interpretation

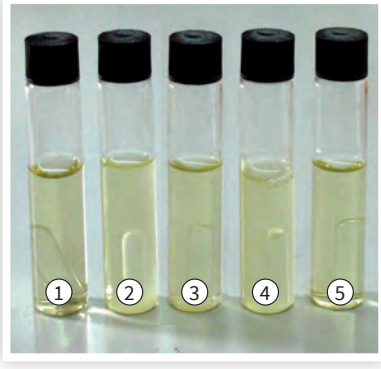
Fluid Lactose Medium is recommended by APHA and USP (1) in the performance and confirmation of the presumptive test for coliform bacteria in water (2), food (3) and milk (4, 10). This medium is also recommended by USP/IP/BP/EP (1, 5, 6, 7). This medium can be used as an alternate to Lauryl Sulphate Broth in the presumptive test of the MPN of standard coliforms. This medium is also used for pre-enrichment of *Salmonella* for its detection in pharmaceutical raw materials.

Gelatin peptone, Peptone, HM Peptone B, HiVeg™ peptone and HiVeg™ extract provide nitrogenous and carbonaceous compounds, long chain amino acids and supply essential nutrients to the organisms. Lactose is a fermentable carbohydrate for the coliforms. Tubes of Fluid Lactose Medium are inoculated with dilutions of water or milk, etc. under test, and incubated at 35-37°C and examined for gas formation after 24 and 48 hours.

Members of the coliform group are defined as aerobic and facultative anaerobic gram-negative and non-sporing bacilli which ferment lactose with gas formation within 48 hours at 35°C. In testing dairy products, Fluid Lactose Medium is used only in the completed test (3). Large water samples may require double strength Fluid Lactose Medium to minimize the final volume.

Type of specimen

Food and dairy samples; Water samples; Pharmaceutical samples.



Lactose Broth M1003
1. Control
2. *Escherichia coli* ATCC 8739 (00012*)
3. *Enterococcus faecalis* ATCC 29212 (00087*)
4. #*Klebsiella aerogenes* ATCC 13048 (00175*)
5. *Pseudomonas aeruginosa* ATCC 9027 (00026*)

\# Formerly known as *Enterobacter aerogenes*
*Corresponding WDCM no.

Lactose Broth M1003

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3, 4, 10).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (2).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1, 5, 6, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further subculture biochemical tests must be carried out for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

Colour and Clarity of prepared medium

Light amber coloured clear solution in tubes.

pH

6.90 ± 0.2

Cultural Response

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

Organism (ATCC)	Inoculum (CFU)	Growth	Gas
#*Klebsiella aerogenes* 13048 (00175*)	50 -100	luxuriant	Positive reaction
Enterococcus faecalis 29212 (00087*)	50 -100	luxuriant	Negative reaction
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	Negative reaction
Escherichia coli 8739 (00012*)	50 -100	luxuriant	Positive reaction

Key : * : Corresponds to WDCM number
Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 15 - 25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
3. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods For The Microbiological Examination of Foods, 5th ed., APHA, Washington, D.C
4. Wehr H M and Frank J H., 2004, Standard Methods for the Examination of Dairy Products, 17th ed., APHA Inc., Washington, D.C.
5. The Indian Pharmacopoeia 2014, Govt. of India, The Controller of Publication, Delhi
6. British Pharmacopoeia 2008, The Stationery Office, British Pharmacopoeia
7. European Pharmacopoeia 2008, European Department, for the Quality of Medicines.
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
10. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Medium in Glass Bottles			
LQ212C	Lactose Broth	for the detection of coliform bacteria in water foods, dairy products as per standard methods.	5X100ml
LQ212D	Lactose Broth	for the detection of coliform bacteria in water foods, dairy products as per standard methods.	5X500ml

Lactose Sulphite Broth Base M1287

Intended Use:

Lactose Sulphite Broth Base is recommended for the detection and enumeration of *Clostridium perfringes* in pharmaceutical products.

Directions:

Suspend dehydrated media as per table in 1000 ml purified/ distilled water. Heat if necessary to dissolve the medium completely. Dispense in tubes containing inverted fermentation vial (Durham's tube) as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C and add filter sterilized solution of 1.2% sodium metabisulphite R (0.5 ml) and 1.0% ferric ammonium citrate (0.5 ml) to each tube.

Ingredients	HiMedia	EP	BP
	M1287	ME1287	M1287B
Tryptone	5.00	—	—
Tryptone#	—	5.00	5.00
Yeast extract	2.50	2.50	2.50
Sodium chloride	2.50	2.50	2.50
Lactose monohydrate	—	10.00	10.00
Lactose	10.00	—	—
Cysteine hydrochloride	0.30	0.30	0.30
Grams/litre	20.30	19.80	19.80
Final pH (at 25°C)	7.1±0.2	—	—
pH after sterilization	—	7.1±0.1	7.1 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min or as per validated cycle

Pancreatic digest of casein

Principle And Interpretation

Lactose Monohydrate Sulphite Medium is prepared as per the British Pharmacopoeia and European Pharmacopoeia (1, 2) and is cited as Medium R. This medium is useful in semi-quantitative test for presence of *Clostridium perfringes* in pharmaceutical products where the level of this species is a criterion of quality.

The medium contains Tryptone and Yeast extract, which provides essential nitrogenous and carbonaceous substance, long chain amino acids, vitamin B complex and other essential growth factor compounds for *Clostridium* species. Lactose monohydrate serves as carbon or fermentable carbohydrate source. Cystine hydrochloride rich in sulphur content provides reduced conditions. Sodium metabisulphate and ferric ammonium citrate act as indicators of sulphate reduction, blackening of indicated by blackening of the medium.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Due to nutritional variations some organisms may show poor growth.
2. Recovery to be done further on selective media for isolation.
3. Biochemical / serological testing to be carried out for complete identification.

Lactose Sulphite Broth Base M1287

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

Colour and Clarity of prepared medium

Light amber coloured clear solution without any precipitate.

pH

ME1287/M1287B - pH : 7.10 ± 0.1

M1287 - pH : 7.10 ± 0.2

Cultural Response:

Cultural characteristics observed after an incubation at 46±0.5°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	H_2S	Gas
Clostridium perfringens 13124 (00007*)	50 -100	luxuriant	Positive reaction, blackening of medium	Positive reaction
Clostridium sporogenes 19404	50 -100	luxuriant	negative reaction, no blackening of medium	Positive reaction

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 15 - 25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. British Pharmacopoeia, 2008, The Stationery office British Pharmacopoeia.
2. European Pharmacopoeia 2008, European Department, for the Quality of Medicines.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Nutrient Agar w/ 1% Peptone M012

Intended Use:

Nutrient Agar is used as general purpose culture media for the cultivation of microorganisms.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified /distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving as specified in table or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	IP	HiVeg™
	M012	MM012	MV012
Peptone	10.00	10.00	—
HM Peptone B##	5.00	10.00	—
HiVeg™ peptone	—	—	10.00
HiVeg™ extract	—	—	5.00
Sodium chloride	5.00	5.00	5.00
Agar	15.00	12.00	15.00
Grams/litre	35.00	37.00	35.00
Final pH (at 25°C)	7.4±0.2	—	7.4±0.2
pH after sterilization	—	7.3±0.1	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min

#Equivalent to Beef extract

Principle And Interpretation

Nutrient Agar is a basic culture medium used for maintaining microorganisms (1), for purity checking prior to biochemical or serological testing. It is used for the cultivation and enumeration of bacteria, which are not particularly fastidious. In semisolid form it is used for maintenance of control or standard organisms. Indian Pharmacopoeia has recommended it for microbial limit tests of viable aerobic microorganism present in pharmaceutical substances (2).

Peptone, HM Peptone B, HiVeg™ peptone and HiVeg™ extract provide the necessary nitrogen compounds, carbon, long chain amino acids, vitamins and also some trace ingredients. Sodium chloride maintains osmotic equilibrium. Nutrient media may be used as enriched media by the addition of 10% v/v blood or other biological fluids like ascitic fluid, serum etc.

Type of specimen

Clinical : faeces; Food and dairy samples; Water samples; Pharmaceutical samples.

Specimen Collection and Handling

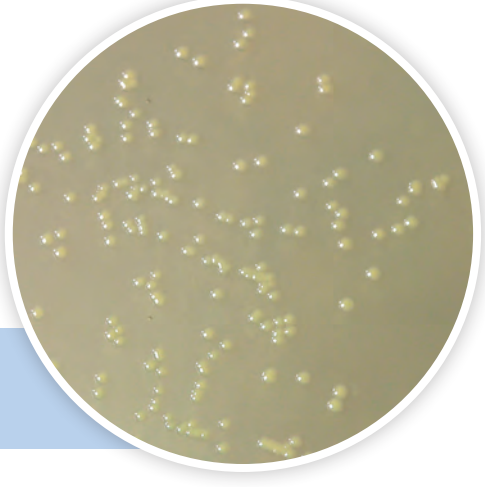
For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3, 4).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (5, 6, 7).

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

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (2).

After use, contaminated materials must be sterilized by autoclaving before discarding.



Nutrient Agar w/ 1% Peptone (M012)
Staphylococcus aureus subsp. *aureus* ATCC 6538 (00032*)
*corresponding WDCM no.

Nutrient Agar w/ 1% Peptone M012

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Due to nutritional variations some strains may show poor growth.
2. The medium is general purpose so further biochemical tests must be carried for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

Gelling

Firm, comparable with 1.2% Agar gel of MM012 and 1.5% Agar gel of M012/MV012.

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

MM012 - 7.3 ± 0.1
M012/MV012 - 7.2 ± 0.2

Growth Promotion Test

Growth Promotion is carried out as per Indian Pharmacopoeia.

Cultural Response

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

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery
Escherichia coli 8739 (00012*)	50 -100	good-luxuriant	≥70%
Staphylococcus aureus subsp. aureus 6538 (00032*)	50 -100	good-luxuriant	≥70%
Salmonella Typhi*murium* 14028 (00031*)	50 -100	good-luxuriant	≥70%
Salmonella Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	≥70%
Pseudomonas aeruginosa 9027 (00026*)	50 -100	good-luxuriant	≥70%

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20 - 30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. Lapage S., Shelton J. and Mitchell T., 1970, 'Methods in Microbiology', Norris J. and Ribbons D. (ed.), Vol. 3A., MM012 - 7.20-7.40 Academic Press, London.
2. Indian Pharmacopoeia, 2018, Govt. of India, The Controller of Publications, Delhi.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1
5. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
6. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
7. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
8. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Nutrient Broth w/ 1% Peptone M244

Intended Use:

Nutrient Broth w/ 1% Peptone is used as a sterility testing and general purpose medium.

Directions:

Suspend dehydrated media as per the table in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure(121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	IP	HiVeg™
	M244	MM244	MV244
Peptone	10.00	10.00	—
HM Peptone B##	10.00	10.00	—
HiVeg™ peptone	—	—	10.00
HiVeg™ extract	—	—	10.00
Sodium chloride	5.00	5.00	5.00
Grams/litre	25.00	25.00	25.00
Final pH (at 25°C)	7.4±0.2	—	7.4±0.2
pH after sterilization	—	7.3±0.1	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min

Peptic digest of animal tissue
##Equivalent to Beef extract

Principle And Interpretation

Nutrient Broth Medium is a general purpose medium used for the examination of water and dairy products according to Standard Methods for the Examination of Water and Wastewater (1) and Dairy Products (2) in accordance with IP (4). It can also be used for cultivating several less fastidious microorganisms.

Peptone, HM Peptone B, HiVeg™ extract and HiVeg™ peptone provide the necessary carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances and also some trace ingredients to the non-fastidious organisms. Sodium chloride maintains osmotic equilibrium of the medium.

Type of specimen

Food & dairy samples, Water samples, Pharmaceutical samples.

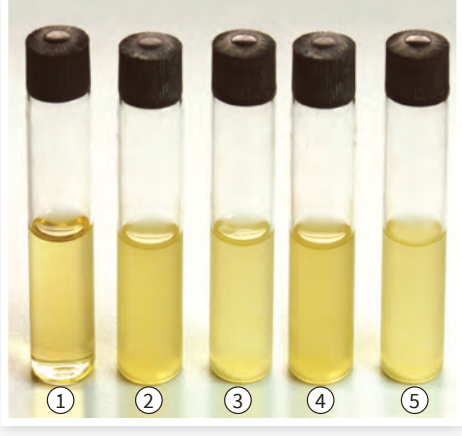
Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (2, 3, 7).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(1).

For pharmaceutical samples, follow appropriate technique for sample collection and processing as per guidelines (4)

After use, contaminated materials must be sterilized by autoclaving before discarding.



Nutrient Broth w/ 1% Peptone (M244)
1. Control
2. *Escherichia coli* 8739 (00012*)
3. #*Klebsiella aerogenes* ATCC 13048 (00175*)
4. *Klebsiella pneumoniae* ATCC 13883 (00097*)
5. *Salmonella Typhimurium* ATCC 14028 (00031*)

Formerly known as *Enterobacter aerogenes*
*corresponding WDCM no.

Nutrient Broth w/ 1% Peptone M244

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Due to nutritional variation, some strains may show poor growth.
2. Further biochemical test have to be performed for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

Colour and Clarity of prepared medium

Light yellow coloured clear solution.

pH

MM244 - 7.3 ± 0.1,

M244/MV244 - 7.4 ± 0.2

Growth Promotion Test

Growth promotion is carried out as per Indian Pharmacopoeia.

Cultural Response:

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth
Escherichia coli 8739 (00012*)	50 -100	luxuriant
Escherichia coli NCTC 9002	50 -100	luxuriant
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant
Escherichia coli 25922 (00013*)	50 -100	luxuriant
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant
#*Klebsiella aerogenes* 13048 (00175*)	50 -100	luxuriant
Klebsiella pneumoniae 13883 (00097*)	50 -100	luxuriant

Key : * : Corresponds to WDCM number
Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 15 - 25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
2. American Public Health Association, 1978, Standard Methods for the Examination of Dairy Products, 14th ed., APHA, Inc., Washington, D.C.
3. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
4. Indian Pharmacopoeia, 2018, Third Edition, Government of India Ministry of Health of family Welfare.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1
7. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Pseudomonas Agar (For Pyocyanin) M119

Intended Use:

Pseudomonas Agar (For Pyocyanin) is recommended for the detection of pyocyanin production by *Pseudomonas* species. This medium is also recommended for microbial limit testing of pharmaceutical raw materials and finished products as well as of other biological preparations.

Directions:

Suspend dehydrated medium as per table in 1000 ml purified / distilled water, containing 10ml glycerol. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	Granulated	USP	IP	HiVeg™
	M119	GM119	MU119	MM119	MV119
Gelatin peptone#	—	—	20.00	20.00	—
Peptone	20.00	20.00	—	—	—
HiVeg™ peptone	—	—	—	—	20.00
Anhydrous potassium sulphate	—	—	10.00	10.00	—
Potassium sulphate	10.00	10.00	—	—	10.00
Anhydrous magnesium chloride	—	—	1.40	1.40	—
Magnesium chloride	1.40	1.40	—	—	1.40
Agar	15.00	15.00	15.00	15.00	15.00
Grams/litre	46.40	46.40	46.40	46.40	46.40
Final pH (at 25°C)	7.0±0.2	7.0±0.2	—	—	7.0±0.2
pH after sterilization	—	—	*7.2±0.2	*7.2±0.2	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Glycerol / Glycerin	10ml	10ml	10ml	10ml	10ml
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

*pH can also be measured after sterilization, at 25°C
Pancreatic digest of gelatin

Principle And Interpretation

Pseudomonas Agar is based on the formulation described by King et al (1) and as recommended by U.S. Pharmacopoeia (2) and Indian Pharmacopoeia (6) for detecting pyocyanin, a water soluble pigment by *Pseudomonas* species (3). It is also recommended for microbial limit tests for pharmaceutical and other biological preparations by USP. *Pseudomonas* strains are reported to produce phenazine pigments like Pyocyanin- blue green redox-active secondary metabolite pigment, pyorubin- rust brown pigment, -oxyphenzinea breakdown product of Pyocyanin, pyoverdin-a water soluble yellow green pigments also known as fluorescein. Pyocyanin is readily recovered in large quantities in sputum from patients with cystic fibrosis, an infection caused by *Pseudomonas* (4, 5). This medium enhances the formation of Pyocyanin and/or pyorubin and reduces that of fluorescein.

Gelatin peptone, Peptone, HiVeg™ peptone provides carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances for growth of *Pseudomonas*, while glycerol, glycerin provides carbon and energy to the cell. The pyocyanin pigment diffuses from the colonies of *Pseudomonas* into the agar and shows blue colouration. Potassium sulphate and magnesium chloride enhances the pyocyanin production and suppresses the fluorescein production. Low content of phosphorous in the medium also aids in inhibiting the production of fluorescein.

MM119 Psuedomonas Agar for detection of Pyocyanin
Pseudomonas aeruginosa ATCC 9027 (00026*)
*Corresponding WDCM No.

Pseudomonas Agar (For Pyocyanin) M119

Some *Pseudomonas* strains produce small amounts of fluorescein resulting in a blue-green colouration. Strains of *Pseudomonas aeruginosa* that may fail to produce pyocyanin are not detected in this medium. Production of other pigments may mask the presence of pyocyanin.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (2, 6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Salt concentration exceeding 2% affects pigment production.
2. Due to nutritional variations, some strains may show variation in growth and pigmentation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

GM119 : Cream to yellow granular media

Gelling

Firm comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

M119 / GM119 / MV119 - 7.0 ± 0.2

MU119 / MM119 - 7.2 ± 0.2

Cultural response

Growth Promotion is carried out in accordance with the harmonized method of USP. Cultural response was observed after an incubation at 33-37°C for not less than 3 days. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Characteristic colonial morphology	Fluorescence in UV light	Oxidase
Test for *Pseudomonas aeruginosa*						
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	≥70%	Generally greenish	Blue	positive
Additional Microbiological testing						
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	≥70%	Generally greenish	Blue	positive

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20 - 30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 7).

Reference

1. King, Ward and Raney, 1954, J.Lab. and Clin. Med., 44:301.
2. United States Pharmacopoeia, 2019, United States Pharmacopoeia Convention, Inc., Rockville, MD.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Daly J A, Boshard R, and Matsen J M, 1984, J Clin Microbiol. 19: 742.
5. Lau GW, Hassett DJ, Ran H, Kong F., 2004. Trends Mol Med. 10:599.
6. The Indian Pharmacopoeia 2007, Govt. of India, The Controller of Publication, Delhi.
7. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media in 90 mm plates			
MP119	Pseudomonas Pyocyanin Agar Plate	for detection of pyocyanin production by *Pseudomonas* species.	20pt / 50pt

Pseudomonas Agar (For Fluorescein) M120

Intended Use:

Pseudomonas Agar (For Fluorescein) is recommended for the detection of fluorescein production by *Pseudomonas* species. This medium is recommended by various pharmacopoeias for carrying out microbial limit tests of pharmaceutical raw material and ingredients as well as pharmaceutical preparations and finished products.

Directions:

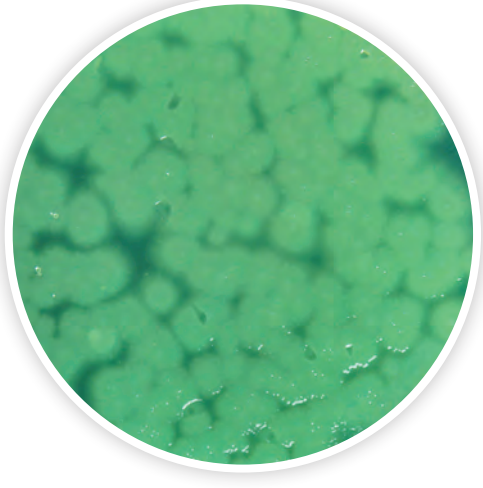
Suspend dehydrated medium as per table in 1000 ml purified / distilled water, containing 10ml glycerol. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	Granulated	USP	IP	HiVeg™	Chemically defined
	M120	GM120	MU120	MM120	MV120	MCD120
Tryptone#	10.00	10.00	10.00	10.00	—	—
Peptone##	—	—	10.00	10.00	—	—
Proteose peptone	10.00	10.00	—	—	—	—
HiVeg™ hydrolysate	—	—	—	—	10.00	—
HiVeg™ peptone No. 3	—	—	—	—	10.00	—
HiCynth™ Peptone No. 3	—	—	—	—	—	15.00
HiCynth™ peptone No. 5	—	—	—	—	—	5.00
Dipotassium hydrogen phosphate	1.50	1.50	—	—	1.50	1.50
Anhydrous dibasic potassium phosphate	—	—	1.50	1.50	—	—
Magnesium sulphate	1.50	1.50	—	—	1.50	1.50
Magnesium sulphate, 7H O	—	—	1.50	1.50	—	—
Agar	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	38.00	38.00	37.23	37.23	38.00	38.00
Final pH (at 25°C)	7.0±0.2	7.0±0.2	—	—	7.0±0.2	7.0±0.2
pH after sterilization	—	—	*7.2±0.2	*7.2±0.2	—	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Glycerol / Glycerin	10ml	10ml	10ml	10ml	10ml	10ml
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

*pH can also be measured after sterilization, at 25°C
Pancreatic digest of casein
##Peptic digest of animal tissue

Principle And Interpretation

Pseudomonas Agar (For Fluorescein) is based on the formula described by King et al (1) and as modified in the U.S. Pharmacopoeia (2) and Indian Pharmacopoeia (4) for the detection of fluorescein production a water soluble, chloroform insoluble fluorescent pigment by *Pseudomonas* species (3). *Pseudomonas* is ubiquitous in environment and is a common causative agent of burn, skin and nosocomial infections. They are also common contaminant of pharmaceutical and cosmetics related preparations. *Pseudomonas* strains are reported to produce phenazine pigments like Pyocyanin- blue green redoxactive secondary metabolite pigment, pyorubin-rust brown pigment, -oxyphenzine- a breakdown product of Pyocyanin, pyoverdin-a water soluble yellow green pigments also known as fluorescein. This medium enhances the elaboration of fluorescein by *Pseudomonas* and inhibits the yellowish pyocyanin formation. The fluorescein pigment diffuses from the colonies of *Pseudomonas* into the agar and shows yellow fluorescent colouration. Some *Pseudomonas* strains produce small amounts of pyocyanin resulting in a yellow- green colouration.



MV120 Psuedomonas HiVeg Agar for detection of Fluorescein
Pseudomonas aeruginosa ATCC 9027 (00026*)
(*Corresponding WDCM No.)

Pseudomonas Agar (For Fluorescein) M120

Peptone, Tryptone, Proteose peptone, HiVeg™ hydrolysate, HiVeg™ peptone No. 3, HiCynth™ peptone No. 3 and HiCynth™ peptone No. 5 provides the essential nitrogenous and carbonaceous nutrients, long chain amino acids, vitamins, sulfur and trace elements for the growth of *Pseudomonas*. These nutrients are also conducive to the production of fluroescein. Peptone and phosphorous in the medium enhance the production of pyoverdin/fluorescein pigment. Dipotassium hydrogen phosphate buffers the medium while magnesium sulphate provides necessary cations for the activation of fluorescein production. Salt concentration exceeding 2% affects pigment production. UV illumination may be bactericidal, so make sure that there is good growth before placing culture under UV light (3).

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (2, 4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Salt concentration exceeding 2% affects pigment production.
2. UV illumination may be bactericidal, so make sure that there is good growth before placing culture under UV light .

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

GM120 : Cream to yellow granular media

Gelling

Firm, comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

MU120/MM120 - 7.20 ± 0.2

M120/GM120 /MV120 / MCD120 - 7.00 ± 0.2

Cultural response

Growth Promotion is carried out in accordance with the harmonized method of USP/IP. Cultural response was observed after an incubation at 33-37°C for not less than 3 days. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Characteristic colonial morphology	Fluorescence in UV light	Oxidase
Test for *Pseudomonas aeruginosa*						
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant	≥70%	Generally colourless to yellowish	positive	positive
Additional Microbiological testing						
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant	≥70%	Generally colourless to yellowish	positive	positive

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20 - 30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. King, Ward and Raney, 1954, J.Lab. Clin. Med., 44 : 301.
2. United States Pharmacopoeia, 2019 United States Pharmacopoeia Convention, Inc., Rockville, MD.
3. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification and Maintenance of Medical Bacteria, Vol. I, Quality Control Williams and Wilkins, Baltimore.
4. The Indian Pharmacopoeia 2007, Govt. of India, The Controller of Publication, Delhi.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media in 90 mm plates			
MP120	Pseudomonas Fluorescein Agar Plate	for detection of fluorescein production by *Pseudomonas* species.	20pt / 50pt

R-2A Agar M962

Intended Use:

R-2A Agar is used for heterotrophic plate count of water samples, which provides an estimate of aerobic and facultatively anaerobic heterotrophs in water samples.

Directions:

Suspend dehydrated medium as per the table in 1000 ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates. DO NOT OVERHEAT.

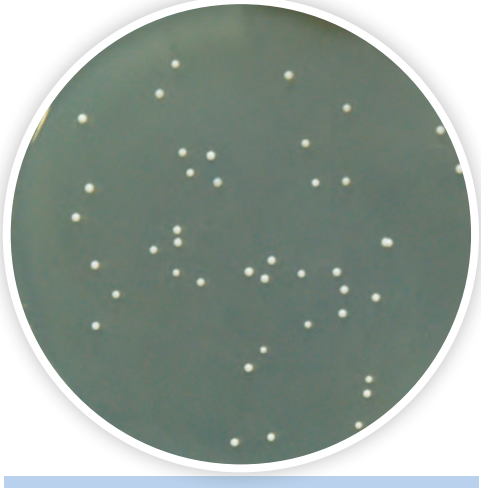
Ingredients	HiMedia	Granulated	EP	BP	HiVeg™	Chemically defined
	M962	GM962	ME962	M962B	MV962	MCD962
Acicase	0.50	0.50	—	—	—	—
Casitose#	—	—	0.50	0.50	—	—
Proteose peptone	0.50	0.50	0.50	0.50	—	—
HiVeg™ peptone No. 3	—	—	—	—	0.50	—
HiCynth™ Peptone No. 3##	—	—	—	—	—	0.50
HiCynth™ Peptone No. 5##	—	—	—	—	—	1.00
HiVeg™ acid hydrolysate	—	—	—	—	0.50	—
Yeast extract	0.50	0.50	0.50	0.50	0.50	—
Glucose (Dextrose)	0.50	0.50	0.50	0.50	0.50	0.50
Starch	—	—	0.50	0.50	—	—
Starch soluble	0.50	0.50	—	—	0.50	0.50
Dipotassium hydrogen phosphate	0.30	0.30	0.30	0.30	0.30	0.30
Magnesium sulphate	0.024	0.024	0.024	0.024	0.024	0.024
Sodium pyruvate	0.30	0.30	0.30	0.30	0.30	0.30
Agar	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	18.12	18.12	18.12	18.12	18.12	18.12
Final pH (at 25°C)	7.2±0.2	7.2±0.2	—	—	7.2±0.2	7.2±0.2
pH after sterilization	—	—	7.2±0.2	7.2±0.2	—	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

Casein hydrolysate
Chemically defined peptones

Principle And Interpretation

R-2 A Agar is used for the heterotrophic plate counts and for sub culturing isolates from potable waters using longer incubation periods as per European Pharmacopoeia (1, 2). The media is also recommended by British Pharmacopoeia (4). It is recommended for pour plate, spread plate and membrane filter techniques.

Plate count recommended for the bacterial examination of potable waters, gives an estimate of the aerobic and facultatively anaerobic bacteria, which grow best at 35°C on a rich medium (3). However these organisms may represent a small number of total bacteria as other bacteria are either unable to grow under these conditions, or grow very slowly which cannot be detected in 48 hours. R-2 A Agar is modified for better recovery of these bacteria from treated waters under different incubation conditions (3). Many bacteria from natural waters, which contain limited nutrients at ambient temperature, grow best on the media with less nutrient levels. Moreover, they grow better at the temperatures below the routine laboratory incubation temperatures of 35° to 37°C (3).



M962 R-2A Agar
Salmonella Enteritidis ATCC 13076 (00030*)
* Corresponding WDCM No.

R-2A Agar M962

R-2 A Agar, Modified is a low nutrient medium consisting of less Acicase, Casitose, Proteose peptone, HiCynth™ peptone No. 3, HiCynth™ peptone No. 5, HiVeg™ Peptone No. 3, Yeast extract and glucose as compared to Standard Methods Agar. This medium allows the growth of stressed, injured and chlorine tolerant bacteria present in treated waters due to the presence of pyruvate and starch (2). The number of colonies on a plate is reported as CFU (Colony Forming Units) per volume of sample.

Type of specimen

Water samples, Pharmaceutical samples.

Specimen Collection and Handling

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

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1, 4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Longer incubation time other than specified is required for slow growing organisms.
2. The media is intended for water samples for recovery of stressed or injured organisms.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

GM962 : Cream to yellow granular media

Gelling

Firm, comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

7.00-7.40

Cultural response

Cultural characteristics observed by using standard cultures after an incubation at 30-35°C for 24-72 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery
Candida albicans 10231 (00054*)	50 -100	good-luxuriant	≥50%
Enterococcus faecalis 29212 (00087*)	50 -100	good-luxuriant	≥50%
Salmonella Enteritidis 13076 (00030*)	50 -100	good-luxuriant	≥50%
Salmonella Typhi 6539	50 -100	good-luxuriant	≥50%
Escherichia coli 8739 (00012*)	50 -100	good-luxuriant	≥50%

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20 - 30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. European pharmacopoeia, 2017, European Dept. for the Quality of Medicines.
2. Reasoner and Geldreich, 1985, Appl. Environ. Microbiol., 49:1.
3. Collins and Willoughby, 1962, Arch. Microbiol., 43:294.
4. British Pharmacopoeia 2016, The Stationery Office, British Pharmacopoeia.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
7. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media in 90 mm plates			
MP962 MP962G MP962GT	R-2A Agar Plate R-2A Agar Plate (γ - irradiated) R-2A Agar Plate (γ - irradiated) (Triple pack)	for heterotrophic plate count of treated potable water, using longer incubation period.	20pt / 50pt 20pt / 50pt 20pt / 50pt
Category : Ready Prepared Solid Media in Glass Bottles			
SMEB962 SMEB962CCL SMEB962D	R2A Agar	for heterotrophic plate count of treated potable water using longer incubation periods	5X100ml 5X250ML 5X500ml

Sabouraud Glucose Agar w/Antibiotics M1472

Intended Use:

Sabouraud Glucose Agar with Antibiotics is used for selective cultivation of Yeasts and moulds. It is recommended by EP/BP & IP.

Directions:

Suspend dehydrated medium as per table, in 995 ml Water R/ purified/distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Add the rehydrated contents of 1 vial of FD196 (Tetracycline Selective Supplement). Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	EP	BP	IP
	M1472	ME1472	M1472B	MM1472
HMC Peptone#	—	10.00	10.00	10.00
Tryptone	5.00	—	—	—
Peptone	5.00	—	—	—
Glucose (Dextrose)	40.00	—	—	—
Dextrose monohydrate (Glucose monohydrate)	—	40.00	40.00	40.00
Agar	15.00	15.00	15.00	15.00
Grams/litre	65.00	61.36	61.36	61.36
Final pH (at 25°C)	5.6± 0.2	—	—	—
pH after sterilization	—	5.6± 0.2	5.6± 0.2	*5.6± 0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C - 15 min	Autoclaving 121°C - 15 min or as per validated cycle	Autoclaving 121°C - 15 min or as per validated cycle	Autoclaving 121°C - 15 min
Supplements (FD196)	5 ml	5 ml	5 ml	5 ml

*pH may also be measured after sterilization, at 25°C
#Equivalent to Peptones (meat and casein)

Principle And Interpretation

Sabouraud Dextrose Agar Medium w/ antibiotics is recommended for cultivation of yeasts and moulds by IP/EP/BP (1, 6, 7). This medium was described originally by Sabouraud (2) for the cultivation of fungi, particularly useful for the fungi associated with skin infections. The medium is used with antibiotics such as tetracycline and benzylpenicillin (3) for the isolation of pathogenic fungi from materials containing large numbers of fungi or bacteria.

Peptone, Tryptone, HMC Peptone provide nitrogenous and carbonaceous compounds longer chain amino acids, vitamins and other essential nutrients. Dextrose, Glucose provides an energy source. Tetracycline and benzyl penicillin inhibits a wide range of Gram-positive and Gram-negative bacteria which makes the medium selective for fungi (4). The low pH favours fungal growth and inhibits contaminating bacteria from clinical specimens (5).

Some pathogenic fungi may produce infective spores which are easily dispersed in air, so examination should be carried out in safety cabinet.

Sabouraud Glucose Agar w/Antibiotics M1472
Candida albicans ATCC 10231 (00054*)
* Corresponding WDCM No.

Sabouraud Glucose Agar w/Antibiotics M1472

Type of specimen

Clinical, Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1, 6, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Some pathogenic fungi may produce infective spores which are easily dispersed in air, so examination should be carried out in safety cabinet.
2. For heavily contaminated samples, the plate must be supplemented with inhibitory agents for inhibiting bacterial growth with lower pH.
3. Further biochemical tests should be carried out for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

Gelling

Firm, comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri plates.

pH

5.60 ± 0.2

Cultural Response

Cultural response was carried out in accordance with IP/EP/BP with added Tetracycline Selective Supplement (FD196), after an incubation at 20-25 C for ≤5 days. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar and fungus growth on Sabouraud Dextrose Agar.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery
Candida albicans 10231 (00054*)	50 -100	Luxuriant (white colonies)	≥50%
#Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant	≥50%
Candida albicans 2091 (00055*)	50 -100	luxuriant	≥50%
Saccharomyces cerevisiae 9763 (00058*)	50 -100	luxuriant	≥50%
Escherichia coli 25922 (00013*)	$\geq 10^3$	inhibited	0%
Escherichia coli 8739 (00012*)	$\geq 10^3$	inhibited	0%
Escherichia coli NCTC 9002	$\geq 10^3$	inhibited	0%
Trichophyton rubrum 28191	50 -100	good	-
Lactobacillus casei 334	$\geq 10^3$	inhibited	0%

Key : * : Corresponds to WDCM number
: Formerly known as *Aspergillus niger*

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. Indian Pharmacopoeia 2010, Ministry of Health and Family welfare, Government of India, New Delhi.
2. Sabouraud K., 1892, Ann. Dermatol. Syphilol, 3:1061.
3. Ajello L., 1957, J. Chron. Dis., 5:545.
4. Lorian (Ed.), 1980, Antibiotics In Laboratory Medicine, Williams and Wilkins, Baltimore.
5. Murray, P. R 2005, In Manual of Clinical Microbiology, 7th ed., ASM, Washington, D.C.
6. European Pharmacopoeia 2017, European Dept. for the quality of Medicines.
7. British Pharmacopoeia 2016, The Stationery Office, British Pharmacopoeia.
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Sabouraud Chloramphenicol Agar M1067

Intended Use:

Sabouraud Chloramphenicol Agar is recommended for selective cultivation of yeasts and moulds. It is also recommended by EP / BP & IP.

Directions:

Suspend dehydrated medium as per table in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Ingredients	HiMedia	EP	BP	IP	HiVeg™
	M1067	ME1067	M1067B	MM1067	MV1067
HMC Peptone#	—	10.00	10.00	10.00	—
Tryptone	5.00	—	—	—	—
Peptone	5.00	—	—	—	—
HiVeg™ peptone	—	—	—	—	5.00
HiVeg™ hydrolysate	—	—	—	—	5.00
Dextrose monohydrate (Glucose monohydrate)	—	40.00	40.00	40.00	—
Dextrose (Glucose)	40.00	—	—	—	40.00
Chloramphenicol	0.05	0.05	0.05	0.05	0.05
Agar	15.00	15.00	15.00	15.00	15.00
Grams/litre	65.05	61.41	61.41	61.41	65.05
Final pH (at 25°C)	5.6 ± 0.2	—	—	—	5.6 ± 0.2
pH after sterilization	—	5.6 ± 0.2	5.6 ± 0.2	*5.6 ± 0.2	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min or as per validated cycle	Autoclaving 121°C- 15 min

*pH may also be measured after sterilization, at 25°C
Peptone (meat and casein)

Principle And Interpretation

Sabouraud Chloramphenicol Agar is recommended for cultivation of yeasts and moulds by IP/EP/BP (1, 6, 7). This medium was described originally by Sabouraud (2) for the cultivation of fungi, particularly useful for the fungi associated with skin infections. The medium is often used with antibiotics such as Chloramphenicol (3) for the isolation of pathogenic fungi from materials containing large numbers of fungi or bacteria.

HMC Peptone, Tryptone, Peptone, HiVeg™ hydrolysate and HiVeg™ peptone provide nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential growth nutrients. Dextrose, Glucose provides an energy source. Chloramphenicol inhibits a wide range of gram-positive and gram-negative bacteria making the medium selective for fungi (4). The low pH favours fungal growth and inhibits contaminating bacteria from clinical specimens (5).

Some pathogenic fungi may produce infective spores which are easily dispersed in air, so examination should be carried out in safety cabinet.

Type of specimen

Clinical : skin, Food samples, Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (8, 9).

Sabouraud Chloramphenicol Agar (M1067)
Candida albicans ATCC 10231 (00054*)
* Corresponding WDCM No.

Sabouraud Chloramphenicol Agar M1067

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (10).
For pharmaceutical samples, follow appropriate technique for sample collection and processing as per guidelines (1, 6, 7)
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Some pathogenic fungi may produce infective spores which are easily dispersed in air, so examination should be carried out in safety cabinet.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder.

Gelling
Firm, comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium
Light amber coloured clear to slightly opalescent gel forms in Petri plates.

pH
5.4-5.8

Growth Promotion Test
Cultural response was observed in accordance with IP/EP/BP, after an incubation at 20-25°C for ≤5 days. Recovery rate is considered as 100% for bacteria growth on Soybean Casein Digest Agar and fungus growth on Sabouraud Dextrose Agar.

Cultural Response:

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery
Candida albicans 10231 (00054*)	50 -100	luxuriant (white colonies)	≥50%
#Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant	≥50%
Candida albicans 2091 (00055*)	50 -100	luxuriant	≥50%
Saccharomyces cerevisiae 9763 (00058*)	50 -100	luxuriant	≥50%
Escherichia coli 25922 (00013*)	≥10^3	inhibited	0%
Escherichia coli 8739 (00012*)	≥10^3	inhibited	0%
Escherichia coli NCTC 9002	≥10^3	inhibited	0%
Trichophyton rubrum 28191	50 -100	good	-
Lactobacillus casei 334	≥10^3	inhibited	0%

Formerly Known as *Aspergillus niger*
Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store the dehydrated and prepared media between 15 - 25°C in a tightly closed container. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (8, 9).

Reference

1. Indian Pharmocopoeia, 2014, Ministry of Health and Family Welfare, Govt. of India.
2. Sabouraud K., 1892, Ann. Dermatol. Syphilol, 3:1061.
3. Ajello L., 1957, J. Chron. Dis., 5:545.
4. Lorian (Ed.), 1980, Antibiotics In Laboratory Medicine, Williams and Wilkins, Baltimore.
5. Murray, P. R 2005, In Manual of Clinical Microbiology, 7th ed., ASM, Washington, D.C.
6. European Pharmacopoeia 2017, European Department, for the Quality of Medicines.
7. British Pharmacopoeia 2016, The Stationery Office, British Pharmacopoeia
8. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
9. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
10. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Media in 90 mm Polystyrene Plates			
MP1067 MP1067G MP1067GT	Sabouraud Chloramphenicol Agar Plate Sabouraud Chloramphenicol Agar Plate (γ - irradiated) Sabouraud Chloramphenicol Agar Plate (γ - irradiated) (Triple Pack)	for selective cultivation of yeasts and moulds.	20pt / 50pt 50pt 20pt / 50pt
Category : Ready Prepared Solid Media in Glass Bottles			
SM1067C SM1067CCL	Sabouraud Chloramphenicol Agar	for selective cultivation of Yeast and moulds.	5x100ml 5x250ml
Category : Ready Prepared Slant in Glass Tubes			
SL1067L	Sabouraud Chloramphenicol Agar Slant (long tube)	for selective cultivation of yeast and moulds.	10slants / 25slants

Selenite Broth (Twin Pack) M052

Intended Use:

Selenite media are recommended as enrichment media for the isolation of *Salmonella* from faeces, urine or other pathological materials.

Directions:

Suspend 4.0 grams of Part B in 1000 ml purified / distilled water. Add 19.0 grams of Part A. Mix well. Warm to dissolve the medium completely. Distribute in sterile test tubes. Sterilize in a boiling water bath or a free flowing steam for 10 minutes or as per validated cycle. DO NOT AUTOCLAVE. Excessive heating is detrimental. Discard the prepared medium if large amount of selenite is reduced (indicated by red precipitate at the bottom of tube/bottle).

Note : Recommended to adjust the pH if slight drift is occurring after addition of selenite.

Instead of Part B: DD056 - Sodium Biselenite disc (1 disc per 10 ml of medium) or DB001 - Sodium Biselenite Bud (1 bud per 100 ml of medium) can be added to the medium after boiling.

Caution : Sodium hydrogen selenite (Sodium biselenite) is very toxic, corrosive agent and causes teratogenicity. Handle with great care. If there is contact with skin, wash immediately with lot of water.

Ingredients	HiMedia	Granulated	IP
	M052	GM052	MM052
	Part A	Part A	Part A
Peptone	—	—	5.00
Tryptone	5.00	5.00	—
Lactose	4.00	4.00	4.00
Sodium phosphate	10.00	10.00	—
Disodium hydrogen phospate	—	—	10.00
	Part B	Part B	Part B
Sodium hydrogen selenite	4.00	4.00	4.00
Sodium biselenite disc (DD056)	100 disc	100 disc	100 disc
Sodium biselenite bud (DB001)	10 bud	10 bud	10 bud
Grams/litre (Part A + Part B)	23.00	23.00	23.00
Final pH (at 25°C)	7.0±0.2	7.0±0.2	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling
Temperature and Time	—	—	By maintaining at 100°C for 30 min

Though IP has mentioned Selenite F Broth medium as a single entity, due to corrosive nature of sodium hydrogen selenite, HiMedia provide this medium as Twin pack

Principle And Interpretation

Klett (1) first demonstrated the selective inhibitory effects of selenite and Guth (2) used it to isolate *Salmonella* Typhi. Leifson fully investigated selenite and formulated the media. The formulation corresponds to that of recommended by the Indian Pharmacopoeia (3) for detection of *Salmonella* in foodstuffs, pharmaceuticals and pathological materials. Enrichment media are routinely employed for detection of pathogens in faecal specimens as the pathogens are present in a very small number in the intestinal flora. Selenite F Broth is useful for detecting *Salmonella* in the nonacute stages of illness when organisms occur in the test sample in low numbers and for epidemiological studies to enhance the detection of low number of organisms from asymptomatic or convalescent patients (4).

Peptone and Tryptone provides nitrogenous, carbonaceous subsances, long chain amino acids, vitamins and other essential nutrients. Lactose is the fermentable carbohydrate. Selenite is reduced by bacterial growth producing alkalinity. This causes increase in pH which can reduce the toxicity of the selenite and results in overgrowth of other bacteria. The acid produced by bacteria by lactose fermentation counters the high pH and neutralizes the medium. Sodium phosphate maintains a stable pH and also minimizes the toxicity of selenite.

Enriched broth is subcultured on differential plating media such as Bismuth Sulphite Agar, Brilliant Green Agar, XLD Agar etc. Do not incubate the broth longer than 24 hours as inhibitory effect of selenite decreases after 6 - 12 hours of incubation (5).

Type of specimen

Clinical : faeces, Food samples, Pharmaceutical samples.

Selenite Broth (Twin Pack) M052

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (6, 7).

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

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. As this medium is highly selective it may not support the growth of all *Salmonella* species.
2. Recovery on selective medium is required for isolation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

M052, MM052: Part A : White to light yellow homogeneous free flowing powder.

GM052 : Part A : White to light yellow granular media

Part B : Cream to white homogeneous free flowing powder.

DD056 : White to cream coloured disc.

DB001 : White to cream coloured bud.

Colour and Clarity of prepared medium

Cream to yellow clear to slightly opalescent solution.

Cultural response

Cultural characteristics observed when subcultured on XLD Agar(M031) after an incubation at 35-37°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Recovery	Colour of colony
Salmonella Choleraesuis 12011	50 -100	good-luxuriant	red with black centre
Salmonella Typhi 6539	50 -100	good-luxuriant	red with black centre
Salmonella Typhimurium 14028 (00031*)	50 -100	good-luxuriant	colourless
Escherichia coli 8739 (00012*)	50 -100	none to poor (no increase in numbers)	Yellow
Escherichia coli NCTC 9002	50 -100	none to poor (no increase in numbers)	Yellow

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

Reference

1. Klett A., 1900, Zeitsch Fer Hyg. Und. Infekt., 33: 137.
2. Guth F., 1916, Zbl. Bakt. I. Orig., 77:487.
3. Indian Pharmacopeia, 2007, Govt. of India, The Controller of Publication, Delhi.
4. Murray P. R., Baron E. J., Jorgensen J. H., Pfaller M. A., Yolken R. H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.
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6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
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Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Media in tubes			
LQ070 / LQ070V	Selenite Broth	an enrichment medium for isolation of *Salmonella* species from faeces, urine or other pathological materials.	LQ070-25X10ML/ LQ070-50X10ML/ LQ070V-25X5ML/ LQ070V-50X5ML
Category : Liquid Transport Medium with Swabs			
MS052A	HiCulture™ Transport swabs w/Selenite Medium (A)	with 2.0 ml Medium recommended for enrichment of enteric organisms from fecal specimens.	50 NO

Tetrathionate Broth Base (w/o Iodine and BG) M032

Intended Use:

Tetrathionate Broth Base (w/o Iodine and BG) is used as an enrichment broth for selective isolation of *Salmonella* Typhi and other *Salmonella* from food products, other materials of sanitary importance and clinical specimens such as urine, stools etc. This medium is also recommended by various pharmacopoeia to carry out microbial limit testing of pharmaceutical raw materials as well as finished products.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified / distilled water and heat just to boiling. Cool to below 45-50°C and add 20ml of iodine solution (iodine - 6 grams and potassium iodide - 5 grams in 20 ml distilled water) and 10ml of 0.1% brilliant green solution. Mix well. This complete medium should be used on the day of preparation otherwise sterilized broth base may be stored for some time. Do not heat after the addition of iodine solution. Use the medium immediately after addition of iodine.

For MM032, on the day of use, add 20ml of iodine solution.

NOTE : Due to the presence of calcium carbonate, the prepared medium forms opalescent solution with white precipitate.

Ingredients	HiMedia	Granulated	USP	IP	HiVeg™	Chemically defined
	M032	GM032	MU032	MM032	MV032	MCD032
Tryptone###	—	—	2.50	—	—	—
Tryptone	2.50	2.50	—	—	—	—
Peptone	—	—	—	4.50	—	—
Peptone##	2.50	2.50	2.50	—	—	—
HM Peptone B#	—	—	—	0.90	—	—
HiVeg™ hydrolysate	—	—	—	—	2.50	—
HiVeg™ peptone	—	—	—	—	2.50	—
HiCynth™ Peptone No.1####	—	—	—	—	—	5.00
Yeast extract	—	—	—	1.80	—	—
Bile salts	1.00	1.00	1.00	—	—	—
Synthetic detergent	—	—	—	—	1.00	1.00
Sodium chloride	—	—	—	4.50	—	—
Calcium carbonate	10.00	10.00	10.00	25.00	10.00	10.00
Sodium thiosulphate	30.00	30.00	30.00	40.70	30.00	30.00
Grams/litre	46.00	46.00	46.00	77.40	46.00	46.00
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling
Supplements	20ml iodine solution and 10ml 0.1% Brilliant green solution	20ml iodine solution and 10ml 0.1% Brilliant green solution	20ml iodine solution and 10ml 0.1% Brilliant green solution	20ml iodine solution	20ml iodine solution and 10ml 0.1% Brilliant green solution	20ml iodine solution and 10ml 0.1% Brilliant green solution

#Equivalent to Beef extract
Peptic digest of animal tissue
Pancreatic digest of casein

Principle And Interpretation

Tetrathionate Broth Medium was originally described by Mueller (1) and found that the medium selectively inhibit coliforms and permit unrestricted growth of enteric pathogens. The medium is also recommended by Indian Pharmacopoeia, United States Pharmacopoeia & FDA (2, 7, 8).Compendium of Microbiological Examination of Foods (3) and Standard Methods for the Examination of Water and Wastewater (4) specify this medium as enrichment medium for *Salmonella* species. *Salmonella* is the common causative agent of mild gastroenteritis to typhoid. It is common contaminant in food and other biological products. This medium supports the rejuvenation of *Salmonella* cells injured by food processing which are incapable of forming colonies on plate, but on injection can cause infection.

The selectivity depends on the ability of thiosulphate and tetrathionate (formed by addition of Iodine and Potassium iodide) in combination to suppress commensal coliform organisms (5, 6). The microorganism harboring tetrathionate reductase flourish in this broth. Sodium thiosulphates are inactivators of halogens and can minimize its toxicity in the testing sample, if any during microbial limit tests.

Tetrathionate Broth Base (w/o Iodine and BG) M032

HM Peptone B, Peptone, Tryptone, HiVeg™ peptone, HiVeg™ hydrolysate and HiCynth™ peptone provides essential carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances in this medium. Calcium carbonate neutralizes the acidic tetrathionate decomposition products. Sodium chloride maintains osmotic balance. For further confirmation, streak the enriched cultures after incubation, on the plates of Brilliant Green Agar (MM016/MU016/ME016/M016B/M016/MV016), Xylose Lysine Deoxycholate Agar (MM031/MU031/M031B/ME031/M031/MV031), Bismuth Sulphite Agar (MM027/MU027/M027/MV027), MacConkey Agar (M081/MU081/MM081/ME081/M081B/MV081).

Type of specimen

Clinical : faeces; Food samples; Water samples; Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (11, 12).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3, 9, 10).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(4).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (2, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Futher subculture must be carried out on selective medium.
2. Being highly selective, some strains may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder.

GM032 : Cream to yellow granular media.

Colour and Clarity of prepared medium

Complete medium with added brilliant green and iodine solution - Light green opalescent solution with white precipitate, on standing the precipitate settles down.

Cultural response

Cultural characteristics observed with added brilliant green and iodine solution when sub cultured on Xylose Lysine Deoxycholate Agar after enrichment in Tetrathionate medium, after an incubation at 35-37°C for 18-72 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery on XLD Agar	Colour of colony
Growth Promotion Test				
Salmonella Typhi*murium* 14028 (00031*)	50 -100	luxuriant	≥50%	red with black centres
Salmonella Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	≥50%	red with black centres

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (11, 12).

Reference

1. Mueller, 1923, Compt. Rend. Sco. Biol., 89:434.
2. The Indian Pharmacopoeia 1996.
3. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods For The Microbiological Examination of Foods, 5th ed., APHA, Washington, D.C.
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8. Bacteriological Analytical Manual.
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10. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
11. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
12. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Tetrathionate Brilliant Green Bile Broth M1255

Intended Use:

Tetrathionate Brilliant Green Bile Broth is used for isolation and identification of *Salmonellae*.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified / distilled water. Heat just to boiling. DO NOT AUTOCLAVE OR REHEAT. Dispense as desired aseptically.

Note : Due to the presence of calcium carbonate the prepared medium forms opalescent solution with white precipitate.

Ingredients	HiMedia	Granulated	EP	BP	IP	HiVeg™	Chemically defined
	M1255	GM1255	ME1255	M1255B	MM1255	MV1255	MCD1255
Peptone	8.60	8.60	8.60	8.60	8.60	—	—
HiVeg™ peptone	—	—	—	—	—	11.6	—
HiCynth™ Peptone No. 2	—	—	—	—	—	—	16.50
Bile#	8.00	8.00	—	8.00	—	—	—
Bile dried##	—	—	8.00	—	—	—	—
Dehydrated bile###	—	—	—	—	8.00	—	—
Synthetic detergent No. I	—	—	—	—	—	—	0.10
Synthetic detergent No. II	—	—	—	—	—	5.00	—
Sodium chloride	6.40	6.40	6.40	6.40	6.40	6.40	6.40
Calcium carbonate	20.00	20.00	20.00	20.00	20.00	20.00	20.00
Potassium tetrathionate	20.00	20.00	20.00	20.00	20.00	20.00	20.00
Brilliant green	0.07	0.07	0.07	0.07	0.07	0.07	0.07
Grams/litre	63.07	63.07	63.07	63.07	63.07	63.07	63.07
Final pH (at 25°C)	7.0±0.2	7.0±0.2	—	—	—	7.0±0.2	7.0±0.2
pH after heating	—	—	7.2±0.2	7.2±0.2	*7.0±0.2	—	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling	Boiling

*pH may also be measured after heating at 25°C
#Ox bile
##Ox bile, dried
Dehydrated ox bile

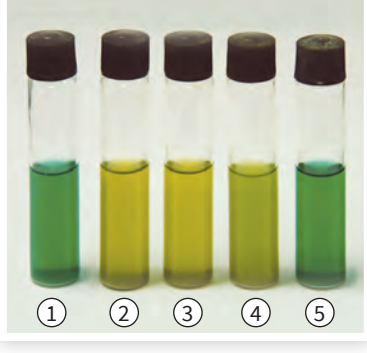
Principle And Interpretation

Salmonella are gram-negative, facultatively anaerobic, non-sporulating, non-motile rods in the family *Enterobacteriaceae*. They are widely distributed in animals affecting mainly the stomach and the intestines. These organisms are difficult to differentiate biochemically from *Escherichia coli*. Tetrathionate Broth was originally described by Mueller (1) and later modified by Kauffman (2, 3). Tetrathionate Brilliant Green Bile Broth is used as an enrichment medium for *Salmonella*. Enrichment broth is usually recommended to facilitate the recovery of small numbers of *Salmonella* species (4). Tetrathionate Brilliant Green Bile Broth is also mentioned in I.P. (5) for isolation and identification of *Salmonella* species from foods, water and other materials of sanitary importance.

Peptone, HiVeg™ peptone and HiCynth™ peptones in the medium provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential nutrient substances for growth of *Salmonellae*. Brilliant green and bile inhibit both gram-positive as well as some selected gram-negative organisms. Potassium tetrathionate inhibits normal flora of faecal specimens. Sodium chloride helps in maintaining osmotic equilibrium. After incubation, streak the culture from Tetrathionate Brilliant Green Bile Broth (M1255) onto differential medium for isolation and identification. Tetrathionate Brilliant Green Bile Broth is not suitable for growth of *Salmonella* Typhi and *Salmonella* ParaTyphi (6).

Type of specimen

Clinical : faeces; Food and dairy samples; Water samples; Pharmaceutical samples.



Tetrathionate Brilliant Green Bile Broth (M1255)
1. Control
2. *Salmonella* Abony NCTC 6017 (00029*)
3. *Salmonella* Typhimurium ATCC 14028 (00031*)
4. *Salmonella* Enteritidis ATCC 13076 (00030*)
5. *Staphylococcus aureus* subsp. *aureus* ATCC 6538 (00032*)

*corresponding WDCM no.

Tetrathionate Brilliant Green Bile Broth M1255

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (11, 12).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (7, 8, 9).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(10).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (5, 13, 14).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Futher subculture must be carried out on selective medium.
2. Being highly selective, some strains may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pale greenish yellow homogeneous free flowing powder.

GM1255 : Light yellow to pale greenish yellow granular media

Colour and Clarity of prepared medium

Bluish green coloured opalescent solution with white precipitate.

pH

7.00 ± 0.2

Cultural response

Cultural characteristics observed after enrichment in Broth Medium I at 41-43°C for 18-24 hours, and then subcultured on Xylose Lysine Deoxycholate Agar, and Brilliant Green, Phenol red, lactose monohydrate Sucrose Agar and incubated at 35-37°C for 18-72 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Growth on Agar Medium K				
Salmonella Typhi*murium* 14028 (00031*)	50 -100	luxuriant	≥50%	red with black centres
Salmonella Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	≥50%	red with black centres
Additional Microbiological Testing Growth on Agar Medium K				
Salmonella Enteritidis *13076 (00030*)*	50 -100	luxuriant	≥50%	red with black centres
Staphylococcus aureus subsp. aureus 6538 (00032*)	$\geq 10^3$	inhibited	0%	
Escherichia coli 8739 (00012)*	50 -100	fair	20 - 30%	yellow
Growth on Agar Medium L				
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant	≥50%	pinkish white
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant	≥50%	pinkish white
Additional Microbiological Testing Growth on Agar Medium L				
Salmonella Enteritidis 13076 (00030*)	50 -100	luxuriant	≥50%	pinkish white
Staphylococcus aureus subsp. aureus 6538 (00032*)	$\geq 10^3$	inhibited	0%	
Escherichia coli 8739 (00012*)	50 -100	fair	20 - 30%	yellow

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (11, 12).

Reference

1. Mueller L., 1923, C. R. Soc. Biol., (Paris), 89, 434.
2. Kauffman F., 1930, Hyg. Abt. I. Orig., 113, 148.
3. Kauffman F., 1935, Z. Hyg. Infektionskr., 117, 26.
4. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.) 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
5. Indian Pharmacopoeia, 2010, Ministry of Health and Family Welfare, Govt. of India.
6. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
7. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
8. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
9. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
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13. British Pharmacopoeia 2016, The Stationery Office, British Pharmacopoeia
14. European Pharmacopoeia 2017, European Department, for the Quality of Medicines.

Triple Sugar Iron Agar M021

Intended Use:

Triple Sugar Iron Agar is used in the differentiation of enteric pathogens to determine their ability to ferment carbohydrates and produce hydrogen sulphide. This medium is recommended by various pharmacopoeias to carry out microbial tests of pharmaceutical raw materials as well as preparations for identification of gram negative bacilli. It is also recommended for bacteriological testing of nutritional and dietary supplements.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Dispense in tubes and sterilize by autoclaving as specified in table or as per validated cycle. Cool the tubed medium in slanting position.

Ingredients	HiMedia	Granulated	USP	EP	BP	IP	HiVeg™	Chemically defined
	M021	GM021	MU021	ME021	M021B	MM021	MV021	MCD021
HMC Peptone##	—	—	—	20.00	20.00	—	—	—
Peptone	10.00	10.00	10.00	—	—	20.00	—	—
Tryptone#	10.00	10.00	10.00	—	—		—	—
HM Peptone B###	3.00	3.00	—	3.00	3.00	3.00	—	—
HiVeg™ peptone	—	—	—	—	—	—	10.00	—
HiVeg™ extract	—	—	—	—	—	—	3.00	—
HiVeg™ hydrolysate	—	—	—	—	—	—	10.00	—
Yeast extract	3.00	3.00	—	3.00	3.00	3.00	3.00	—
HiCynth™ Peptone No. 1####	—	—	—	—	—	—	—	23.00
HiCynth™ Peptone No. 6####	—	—	—	—	—	—	—	3.00
Lactose, monohydrate	—	—	—	10.00	10.00	—	—	—
Lactose	10.00	10.00	10.00	—	—	10.00	10.00	10.00
Saccharose (Sucrose)	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00
Dextrose, monohydrate (Glucose, monohydrate)	—	—	—	1.00	1.00	1.00	—	—
Dextrose (Glucose)	1.00	1.00	1.00	—	—	—	1.00	1.00
Sodium chloride	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Ferric ammonium citrate	—	—	—	0.3-	0.30	—	—	—
Ferrous ammonium sulphate	—	—	0.20	—	—	—	—	—
Ferrous sulphate	0.20	0.20	—	—	—	0.20	0.20	0.20
Sodium thiosulphate	0.30	0.30	0.20	0.30	0.30	0.30	0.30	0.30
Phenol red	0.024	0.024	0.025	0.025	0.025	0.024	0.024	0.024
Agar	12.00	12.00	13.00	12.00	12.00	12.00	12.00	12.00
Grams/litre	64.52	64.52	59.42	64.02	64.02	64.42	64.52	64.52
Final pH (at 25°C)	7.4±0.2	7.4±0.2	—	—	—	—	7.4±0.2	7.4±0.2
pH after sterilization	—	—	7.3 ± 0.2	7.4±0.2	7.4±0.2	—	—	
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Autoclaving 121°C/15 min	Autoclaving 121°C/15 min	Autoclaving 121°C/15 min or as per validated cycle	Autoclaving 121°C/15 min or as per validated cycle	Autoclaving 121°C/15 min or as per validated cycle	Autoclaving 121°C/15 min or as per validated cycle	Autoclaving	Autoclaving 121°C/15 min

*pH may also be measured after heating at 25°C
#Pancreatic digest of casein
Peptones (Casein & Beef)
Equivalent to Beef extract
####Chemically defined peptones

Triple Sugar Iron Agar M021

Principle And Interpretation

Triple Sugar Iron Agar Medium was originally proposed by Sulkin and Willett (1) and modified by (CFU) Hajna (2) for identifying *Enterobacteriaceae* . This medium is in accordance with USP/EP/BP/IP (3, 4, 5, 6) and is recommended in pharmaceutical testing for identification of Gram-negative bacilli. This medium complies with recommendation of APHA, for the examination of meat and food products (7), for the examination of milk and dairy products (8) and for microbial limit test for confirming the presence of *Salmonellae* (9) and in the identification of gram-negative bacilli (9, 10).

Tryptone, Peptone, HMC Peptone, HM Peptone B, HiVeg™ peptone and HiVeg™ hydrolysate, HiVeg™ extract, HiCynth™ Peptone No. 1 and HiCynth™ Peptone No. 6 provide nitrogenous and carbonaceous compounds, long chain amino acids, sulphur, trace elements and vitamin B complex and other essential growth nutrients. Sodium chloride maintains osmotic equilibrium. Lactose, sucrose and dextrose (Glucose) are the fermentable carbohydrates. Sodium thiosulphate helps in reactivation of sulphur containing compounds and prevents the desiccation of these compounds during storage. It also forms the substrate for enzyme thiosulphate reductase, which breaks it to form H_2S. Sodium thiosulphate and ferric or ferrous ions make H_2S indicator system. Sodium thiosulphates are also inactivators of halogens and can minimize its toxicity in the testing sample, if any during microbial limit tests. Phenol red is the pH indicator. Organisms that ferment dextrose (Glucose) produce a variety of acids, varying the colour of the medium from red to yellow. More amounts of acids are liberated in butt region (fermentation) than in the slant (respiration).

Growing bacteria also form alkaline products from the oxidative decarboxylation of peptone and these alkaline products neutralize the large amounts of acid present in the butt. Thus the appearance of an alkaline (red) slant and an acid (yellow) butt after incubation indicates that the organism is a dextrose (Glucose) fermenter but is unable to ferment lactose and/or sucrose. Bacteria that ferment lactose or sucrose (or both), in addition to dextrose (Glucose), produce large amounts of acid enables no reversion of pH in that region and thus bacteria exhibit an acid slant and acid butt. Gas production (CO_2) is detected by the presence of cracks or bubbles in the medium, when the accumulated gas escapes. Thiosulphate is reduced to hydrogen sulphide by several species of bacteria and H_2S combines with ferric ions of ferric salts to produce the insoluble black precipitate of ferrous sulphide. Reduction of thiosulphate proceeds only in an acid environment and blackening usually occurs in the butt of the tube.

Triple Sugar Iron Agar should be used in parallel with Urea Agar / Broth to distinguish between *Salmonella* and *Proteus* species. The reactions can be summarized as follows: Alkaline slant / acid butt- only dextrose (Glucose) fermented. Acid slant / acid butt- dextrose (Glucose) and sucrose fermented or dextrose (Glucose) and lactose fermented or all the three sugars, dextrose (Glucose), lactose and sucrose fermented.

Bubbles or cracks present - gas production. Black precipitate present - H_2S gas production Some members of the *Enterobacteriaceae* and H_2S producing *Salmonella* may not be H_2S positive on TSI Agar. Some bacteria may show H_2S production on Kligler Iron Agar but not on TSI Agar. This can happen because utilization of sucrose in TSI Agar suppresses the enzymic pathway that result in H_2S production.

Type of specimen

Pure isolate

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (12, 13).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (3, 9, 11).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(10).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (3, 4, 5, 6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Some bacteria may show H_2S production on Kligler Iron Agar but not on TSI Agar. This can happen because utilization of sucrose in TSI Agar suppresses the enzymic pathway that result in H_2S production.
2. Urea Agar Base should be used in parallel with TSI to distinguish *Salmonella* from *Proteus* species.

Performance and Evaluation

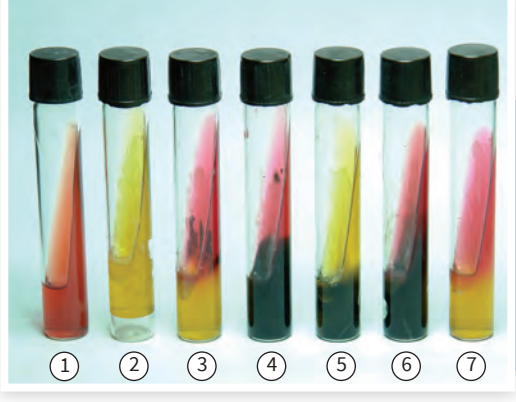
Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder.

GM021 : Light yellow to pinkish granular media



M021 Triple Sugar Iron Agar

1. Control
2. *Escherichia coli* ATCC 25922 (00013*)
3. *Salmonella* Typhi ATCC 6539
4. *Proteus vulgaris* ATCC 13315
5. *Citrobacter freundii* ATCC 8090
6. *Salmonella* Typhimurium ATCC 14028 (00031*)
7. *Shigella flexneri* ATCC 12022 (00126*)

*Corresponding WDCM Nos.

Triple Sugar Iron Agar M021

Gelling
Firm, comparable with 1.2% Agar gel of M021 / GM021/MM021/ ME021/M021B/MV021/MCD021.
Firm, comparable with 1.3% Agar gel of MU021.

Colour and Clarity of prepared medium
Pinkish red coloured clear to slightly opalescent gel forms in tubes as slants.

pH
MU021 - 7.3 ± 0.2
M021/ME021/M021B/MV021 - 7.40 ± 0.2

Cultural response
Cultural characteristics observed after an incubation at 30-35 C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Slant	Butt	Gas	H_2S
Salmonella Abony NCTC 6017 (00029*)	50 -100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Blackening of medium
Salmonella Typhimurium 14028 (00031*)	50 -100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Blackening of medium
Citrobacter freundii 8090	50 -100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	Blackening of medium
#Klebsiella aerogenes 13048 (00175)*	50 -100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	No blackening of medium
Klebsiella pneumoniae 13883 (00097)*	50 -100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	No blackening of medium
Proteus vulgaris 13315	50 -100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Negative reaction	Blackening of medium
Salmonella ParaTyphi A 9150	50 -100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Positive reaction	No blackening of medium
Salmonella Typhi 6539	50 -100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Negative reaction	Blackening of medium
Shigella flexneri 12022 (00126*)	50 -100	Luxuriant	Alkaline reaction, red colour of the medium	Acidic reaction, yellowing of the medium	Negative reaction	No blackening of medium
Escherichia coli 8739 (00012*)	50 -100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	No blackening of medium
Klebsiella pneumoniae 10031	50 -100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	No blackening of medium
Escherichia coli NCTC 9002	50 -100	Luxuriant	Acidic reaction, yellowing of the medium	Acidic reaction, yellowing of the medium	Positive reaction	No blackening of medium

Key : * : Corresponds to WDCM number
: Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life
Store between 10 - 30°C in a tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal
User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (12, 13).

Reference

1. Sulkin, E.S. and Willet J.C., 1940, J. Lab. Clin. Med., 25:649.
2. Hajna A.A., 1945, J. Bacteriol 49:516.
3. The United States Pharmacopoeia, 2019 United States Pharmacopeial Convention, Rockville, Md.
4. European Pharmacopoeia 2008, European Department, for the Quality of Medicines.
5. British Pharmacopoeia 2008, The Stationery Office, British Pharmacopoeia
6. Indian Pharmacopoeia, 1996, Ministry of Health and Family Welfare, Govt. of India.
7. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
8. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
9. Finegold S. M. and Baron E. J., 1986, Bailey and Scotts Diagnostic Microbiology, 7th Ed., The C.V. Mosby Co., St. Louis.
10. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
11. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
12. Isenberg, H.D. Clinical Microbiology Procedures Handb0ook. 2nd Edition.
13. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Solid Media in Glass Bottles			
SM021	Triple Sugar Iron Agar	for identification of Gram negative enteric bacilli on the basis of dextrose, lactose and sucrose fermentation and hydrogen sulphide production.	5X100ML
Category : Ready Prepared Slant in Glass Tubes			
SL045 SL045T	Triple Sugar Iron Agar Slant Triple Sugar Iron Agar Slant in tubes	for identification of Gram-negative enteric bacilli on the basis of dextrose, lactose and sucrose fermentation and hydrogen sulphide production.	10slants / 25slants 10slants / 25slants

Urea Broth Base M111

Intended Use:

Urea Broth base is recommended for the identification of bacteria on the basis of urea utilization, specifically for the differentiation of *Proteus* species from *Salmonella* and *Shigella* species.

Directions:

Suspend dehydrated medium in purified / distilled water as per table. For M111, sterilize by autoclaving at 15 lbs (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Add 50 ml of sterile 40% Urea solution (FD048). Mix well and distribute 10 ml amounts in sterile test tubes. For MM111, mix well and sterilize by filtration. DO NOT AUTOCLAVE OR HEAT the medium. Dispense in sterile tubes.

Ingredients	HiMedia	IP
	M111	MM111
Yeast extract	0.10	0.10
Potassium dihydrogen orthophophate	—	9.10
Potassium dihydrogen phosphate	9.10	—
Dipotassium hydrogen phosphate	9.50	—
Anhydrous disodium hydrogen phosphate	—	9.50
Phenol red	0.01	0.01
Urea	—	20.00
Grams/litre	18.71	38.71
Final pH (at 25°C)	6.8±0.2	—
Water	950ml Purified/ Distilled	Purified/ Distilled
Sterilization	Autoclaving	Filtration
Temperature and Time	121°C / 15 min	—
Supplements	FD048	—

Principle And Interpretation

Urea Broth Medium was developed by Rustigian and Stuart (1). This medium is especially recommended by Indian Pharmacopoeia (5) for the differentiation of *Proteus* species from *Salmonella* and *Shigella* species in the enteric infection diagnosis (2), based on urea utilization (3, 4). It is also recommended for microbial limit tests. Other Gram negative enteric bacilli are unable to utilize urea and fails to grow because of reduced availability of other nutrients.

Urea Broth Medium becomes alkaline as the utilization of urea by the organisms liberate ammonia during the incubation, indicated by pink red colour. All urea test media rely on the alkalinity formation and so they are not specific for urease testing.

Yeast extract provides carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins and other essential nutrient substances and other growth factors. Phosphates aids as good buffering agent. The utilization of proteins may raise the pH to alkalinity due to protein hydrolysis and excess of amino acids results in false-positive reaction. This medium shows positive reaction with Genus *Proteus*, few *Providencia* and *Morganella* species.

Urea Broth Base (M111)
1. Control
2. *Salmonella* Abony NCTC 6017 (00029*)
3. *Salmonella* Typhimurium ATCC 14028 (00031*)
4. *Klebsiella pneumoniae* ATCC 13883 (00097*)
5. *Escherichia coli* ATCC 8739 (00012*)

*corresponding WDCM no.

Urea Broth Base M111

Type of specimen

Pure isolate.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (6, 7).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Prolonged incubation may cause alkaline reaction in the medium.
2. Also, all urea test media rely on the alkalinity formation and so they are not specific for determining the absolute rate of urease activity (4).

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to light pink homogeneous free flowing powder.

Colour and Clarity of prepared medium

Yellow orange coloured clear solution.

Cultural response

MM111: Cultural characteristics observed after an incubation at 36-38 C for 18-24 hours.

M111: Cultural characteristics observed with added sterile 40% Urea solution (FD048) after an incubation at 35-37°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Urease
Salmonella Abony NCTC 6017 (00029*)	50 -100	Negative reaction, no change
Klebsiella pneumoniae 13883 (00097)*	50 -100	Positive reaction, cerise colour
Proteus vulgaris 13315	50 -100	Positive reaction, cerise colour
Salmonella Typhimurium 14028 (00031*)	50 -100	Negative reaction, no change
Escherichia coli 8739 (00012)*	50 -100	Negative reaction, no change
Klebsiella pneumoniae 10031	50 -100	Positive reaction, cerise colour
Escherichia coli NCTC *9002*	50 -100	Negative reaction, no change
Escherichia coli 25922 (00013)*	50 -100	Negative reaction, no change
#Klebsiella aerogenes 13048 (00175*)	50 -100	Negative reaction, no change

Key : * : Corresponds to WDCM number

Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

M111 - Store between 10 - 30°C and MM111 - Store between 2 - 8°C in a tightly closed container and the prepared medium at 15 - 25°C.

Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

Reference

1. Rustigian and Stuart, 1941, Proc. Soc. Exp. Biol. Med., 47:108.
2. Forbes, B.A.; Sahm, D.F. and Weissfelf, A.S., 2002, Bailey and Scott s Diagnostic Microbiology, 11th ed., The C.V. Mosby Co., St. Louis.
3. Christensen, 1946, J. Bact., 52:461.
4. MacFaddin J., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd ed., Williams and Wilkins, Baltimore.
5. Indian Pharmacopoeia, 2007 Govt. of India, Ministry of Health and Family Welfare, New Delhi, India.
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
7. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015),Manual of Clinical Microbiology, 11th Edition. Vol. 1

Violet Red Bile Agar w/ Glucose and Lactose M1684

Intended Use:

Violet Red Bile Agar w/ Glucose and Lactose is used as a selective medium for detection and enumeration of gram negative bile-tolerant bacteria from food and dietary supplement preparations. It is also used for microbiological examination of non-sterile products.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified / distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45-50°C. Mix well and pour in to sterile Petri plates containing the inoculum.

Ingredients	HiMedia	USP	EP	BP	IP
	M1684	MU1684	ME1684	M1684B	MM1684
Gelatin peptone#	—	7.00	7.00	7.00	7.00
Peptone##	7.00	—	—	—	—
Yeast extract	3.00	3.00	3.00	3.00	3.00
Lactose	10.00	10.00	—	—	—
Lactose monohydrate	—	—	10.00	10.00	10.00
Bile salts	—	1.50	1.50	1.50	1.50
Bile salts mixture	1.50	—	—	—	—
Dextrose (Glucose)	10.00	—	—	—	—
Dextrose monohydrate (Glucose monohydrate)	—	—	10.00	10.00	10.00
D-Glucose monohydrate	—	10.00	—	—	—
Sodium chloride	5.00	5.00	5.00	5.00	5.00
Neutral red	0.03	0.03	0.03	0.03	0.03
Crystal violet	0.002	0.002	0.002	0.002	0.002
Agar	12.00	15.00	15.00	15.00	15.00
Grams/litre	48.53	50.62	50.12	50.12	50.12
Final pH (at 25°C)	7.4±0.2	—	—	—	*7.3±0.2
pH after heating	—	7.4±0.2	7.4±0.2	7.4±0.2	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization	Boiling	Boiling	Boiling	Boiling	Boiling

*pH may also be measured after heating at 25°C
#Pancreatic digest of gelatin
##Peptic digest of animal tissue

Principle And Interpretation

This medium is a selective medium, recommended for detection and enumeration of gram negative bile-tolerant bacteria in accordance with USP(1) from food and dietary supplement preparations It is also recommended by EP/IP/BP (3, 4, 5).

Gelatin peptone, Peptone and Yeast extract provide carbonaceous and other nitrogenous compounds, long chain amino acids, vitamins and other nutrient substances essential for bacterial metabolism. This media is selective due to presence of the inhibitors; bile salts and crystal violet. Crystal violet inhibits gram positive organisms especially Staphylococci. Neutral red indicator helps to detect lactose and glucose monohydrate fermentation. Lactose and glucose monohydrate fermenting strains grow as red or pink and may be surrounded by a zone of acid precipitated bile. Sodium chloride maintains the osmotic equilibrium in the medium. The red colour is due to absorption of neutral red and a subsequent colour change of the dye when the pH of medium falls below 6.8.

Type of specimen

Food and Dairy samples; Water samples; Pharmaceutical samples.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (6, 7, 8).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(9).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1, 3, 4, 5).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Violet Red Bile Agar w/ Glucose and Lactose M1684

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Some strains may show poor growth due to nutritional variation.
2. Further biochemical identification is recommended to be performed on pure colonies for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pinkish beige homogeneous free flowing powder.

Gelling

Firm, comparable with 1.5% Agar gel of MU1684/ME1684/ M1684B/MM1684 and 1.2% Agar gel of M1684.

Colour and Clarity of prepared medium

Reddish purple coloured clear to slightly opalescent gel forms in Petri plates.

pH

M1684/MU1684/ME1684/M1684B : 7.4 ± 0.2

MM1684 : 7.3 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with USP/EP/BP/ IP Cultural response was observed after an incubation at 30-35°C for 18-24 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Cultural Response

Organism (ATCC)	Inoculum (CFU)	Recovery	Colour of colony
Test for *Enterobacteriaceae*			
Escherichia coli 8739 (00012*)	50 -100	≥50%	pink-red with bile precipitate
Pseudomonas aeruginosa 9027 (00026*)	50 -100	≥50%	pink to purple
Additional Microbiological Testing			
Escherichia coli NCTC 9002	50 -100	≥50%	pink-red with bile precipitate
Escherichia coli 25922 (00013*)	50 -100	≥50%	pink-red
Salmonella Enteritidis 13076 (00030*)	50 -100	≥50%	light pink
#*Klebsiella aerogenes* 13048 (00175*)	50 -100	≥50%	pink-red
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	$\geq 10^3$	0%	
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	$\geq 10^3$	0%	

Key : * : Corresponds to WDCM number

: Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20 - 30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the

hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (10, 11).

Reference

1. The United States Pharmacopoeia, 2019 Convention. Rockville, MD.
2. Davis J.G., 1951, Milk Testing, dairy Industries Limited, London; pg.131.
3. European Pharmacopoeia 2017, European Department, for the Quality of Medicines
4. British Pharmacopoeia 2008, The Stationery Office, British Pharmacopoeia.
5. The Indian Pharmacopoeia 2008, Govt. of India, The Controller of Publication, Delhi.
6. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
7. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
8. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
9. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
10. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
11. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015),Manual of Clinical Microbiology, 11th Edition. Vol. 1

Violet Red Bile Agar w/ Glucose and Lactose M1684
Escherichia coli ATCC 8739 (00012*)
* Corresponding WDCM No.

Vogel Johnson Agar Base w/o Tellurite (V. J. Agar) M023

Intended Use:

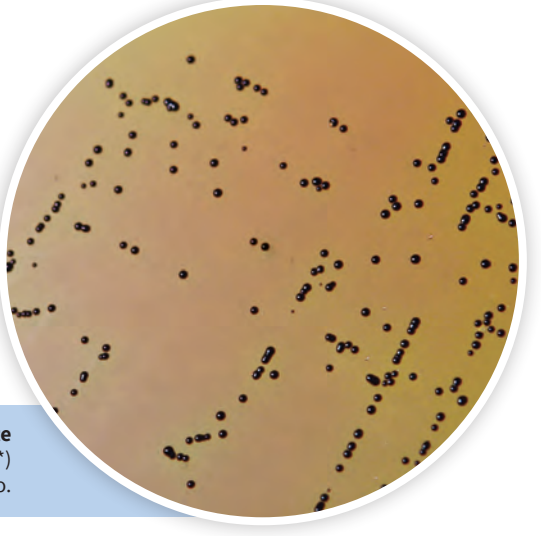
Vogel-Johnson Agar Base w/o Tellurite (V.J. Agar) with addition of potassium tellurite permits early detection of coagulase positive and mannitol positive colonies of *Staphylococcus aureus subsp. aureus* . This medium is recommended for carrying out microbial limit tests of pharmaceutical raw materials and ingredients as well as preparation and finished products.

Directions:

Suspend dehydrated medium as per table in 1000 ml of purified / distilled water. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C and add 20 ml of sterile 1% Potassium tellurite solution (FD052). Mix gently and pour in to sterile Petri plates.

Ingredients	HiMedia	USP	IP	HiVeg™	Chemically defined
	M023	MU023	MM023	MV023	MCD023
Tryptone #	10.00	10.00	10.00	—	—
Yeast extract	5.00	5.00	5.00	5.00	—
HiVeg™ hydrolysate	—	—	—	10.00	—
HiCynth™ Peptone No. 2##	—	—	—	—	15.00
Mannitol	10.00	10.00	10.00	10.00	10.00
Dipotassium hydrogen phosphate	5.00	5.00	5.00	5.00	5.00
Lithium chloride	5.00	5.00	5.00	5.00	5.00
Glycine	10.00	10.00	10.00	10.00	10.00
Phenol red	0.025	0.025	0.025	0.025	0.025
Agar	16.00	16.00	16.00	16.00	16.00
Grams/litre	61.02	61.02	61.02	61.02	61.02
Final pH (at 25°C)	7.2±0.2	—	—	7.2±0.2	7.2±0.2
pH after heating	—	*7.2±0.2	*7.2±0.2	—	—
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min
Supplements	FD052	FD052	FD052	FD052	FD052

*pH may also be measured after heating at 25°C
#Pancreatic digest of casein
Chemically defined peptone



M023 Vogel Johnson Agar Base w/o Tellurite
Staphylococcus aureus ATCC 25923 (00034*)
*Corresponding WDCM No.

Vogel Johnson Agar Base w/o Tellurite (V. J. Agar) M023

Principle And Interpretation

Vogel-Johnson Agar Medium is prepared according to the formula of Vogel and Johnson (1) and is recommended for the microbial limit test (pharmaceutical testing) in USP (2). It is also recommended by IP. Originally it was developed by Zebovitz (3) as a Tellurite Glycine Agar, a selective medium for the detection of coagulase positive Staphylococci. This medium is used to detect *Staphylococcus* in pharmaceutical and cosmetics products (4). *Staphylococcus* is prevalent pathogen in food borne poisoning due to its enterotoxin production. It is commensal found on skin and scalp of human body. Vogel-Johnson modified the medium by adding phenol red as a pH indicator and increased the mannitol quantity.

Tryptone, Yeast extract, HiCynth™ peptone and HiVeg™ hydrolysate provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamin B complex and other growth nutrients. Dibasic potassium phosphate gives buffering capacity to the medium. During first 24 hours of incubation, contaminating organisms are almost inhibited by tellurite, lithium chloride and high glycine content. *Staphylococcus aureus* may be inhibited by these inhibitors but get compensated by mannitol and glycine. Coagulase-positive Staphylococci reduce potassium tellurite to metallic free tellurium and thus produce black colonies surrounded by yellow zones. This yellow colour is due to phenol red indicator, which turns yellow in acidic condition by the fermentation of mannitol. Prolonged incubation may result in the growth of black coagulase negative colonies.

Type of specimen

Clinical : faeces, pus , blood; Food and Dairy samples; Water samples; Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (6.7).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (8, 9, 10).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(11).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (2, 5).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Though this medium is recommended for selective isolation of coagulase positive staphylococci, other strains may grow.
2. Prolonged incubation may result in the growth of black coagulase negative colonies
3. Due to nutritional variation, certain strains may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pink homogeneous free flowing powder.

Gelling

Firm, comparable with 1.6% Agar gel.

Colour and Clarity of prepared medium

Red coloured clear to slightly opalescent gel forms in Petri plates.

pH

7.00-7.40

Cultural response

Cultural characteristics observed with added 1% Potassium Tellurite solution (FD052), after an incubation at 30-35°C for 18-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony
Test for specified microorganisms				
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	good- to luxuriant	≥50%	black colony surrounded by yellow zone
Additional Microbiological testing				
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	good- to luxuriant	≥50%	black colony surrounded by yellow zone
Staphylococcus epidermidis 12228 (00036)*	50 -100	fair to good	30-40%	translucent to blackish
Proteus mirabilis 25933	50 -100	none - poor	≤10%	yellow
Escherichia coli 8739 (00012)*	$\geq 10^3$	inhibited	0%	
Escherichia coli 25922 (00013*)	$\geq 10^3$	inhibited	0%	

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and the prepared medium at 20 - 30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Vogel Johnson Agar Base w/o Tellurite (V. J. Agar) M023

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

Reference

1. Vogel and Johnson, 1960, Public Health Lab., 18:131.
2. The United States Pharmacopoeia, 2019. United States Pharmacopoeial Convention, Inc. Rockville, MD.
3. Zebovitz, Evans and Niven, 1955, J. Bacteriol., 70:686.
4. Bacteriological Analytical Manual, 8th Edition, Revision A, 1998. AOAC, Washington D.C.
5. The Indian Pharmacopoeia 2007, Govt. of India, The Controller of Publication, Delhi.
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
7. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015), Manual of Clinical Microbiology, 11th Edition. Vol. 1.
8. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
9. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
10. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington, D.C.
11. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Media in 90 mm Polystyrene Plates			
MP023	Vogel Johnson Agar Plate (V.J. Agar Plate)	for selective isolation of coagulase positive, mannitol fermenting *Staphylococcus aureus* from heavily contaminated food and clinical specimens.	20plts / 50plts

Wilson and Blair's BBS Agar M331

Intended Use:

This medium is recommended for the selective subculture of *Salmonella* species.

Directions:

Suspend 60.0 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. To sterile melted base, add 4 ml of 1% Brilliant green solution and 70 ml of Selective reagent.

Solution 1: 40 gm Sodium sulphite in 100 ml distilled water.

Solution 2: 21 gm Dibasic sodium phosphate in 100 ml distilled water.

Solution 3: 12.5 gm Bismuth ammonium citrate in 100 ml distilled water,

Solution 4: 0.96 gm Ferrous sulphate in 20 ml distilled water with 2 drops of Hydrochloric acid. Prepare each solution separately and boil the combined solution until a slate grey colour develops.

MM331 Suspend 4.5 grams in 100 ml purified/ distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure 121°C for 15 minutes or as per validated cycle. Suspend 5.6 grams of Part B in 20 ml sterile purified / distilled water, boil the solution till all the ingredients are dissolved properly. Suspend 0.045 grams of Part C in 4.5 ml sterile purified/ distilled water. Mix aseptically 20 ml of solution (i) and 4.5 ml of solution (ii) with 100 ml of previously melted Part A and cool to a temperature 60°C and pour into sterile Petri plates.

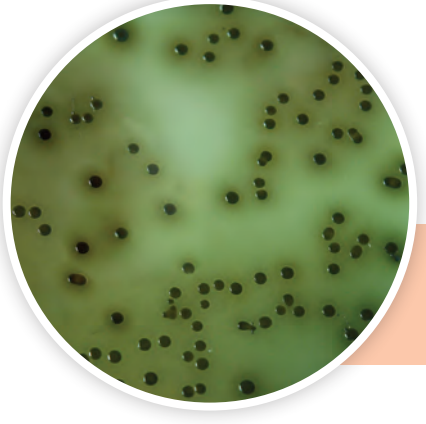
Principle And Interpretation

Wilson and Blair Agar, formulated by Wilson and Blair (2) is recommended for isolating *Salmonella* species. This medium is particularly valuable for the isolation of *S.*Typhi. The medium is highly selective for *Salmonella*e, being inhibitory to coliforms, *Proteus*; occasional strains of coliforms grow to form dull green or brown colonies, but without a surrounding metallic sheen. This medium is recommended by Indian Pharmacopoeia for the selective subculture of *Salmonella* after enrichment in Rappaport Vassliadis *Salmonella* Enrichment Broth (1).

Peptone, Special peptone, HiVeg™ special peptone and HM Peptone B and HiVeg™ extracts provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other growth nutrients. Sodium chloride maintains the osmotic balance. Brilliant green dye inhibits all gram positive bacteria. Glucose is the fermentable carbohydrate. Bismuth is a heavy metal, which is inhibitory to most gram-negative enteric bacilli other than *Salmonella*. Ferric citrate is reduced by *Salmonella* species in presence of bismuth amonium citrate and glucose to form iron sulphide, indicated by black coloured colonies. Disodium hydrogen phosphate buffers the medium well.

Ingredients	HiMedia	IP	HiVeg™
	M331	MM331	MV331
		Part A	
Nutrient Agar**			
Peptone	10.00	–	–
Special peptone	–	10.00	–
HiVeg™ special peptone	–	–	10.00
HM Peptone B#	5.00	10.00	–
HiVeg™ extract	–	–	5.00
Dextrose (Glucose)	10.00	–	10.00
Sodium chloride	5.00	5.00	5.00
Agar	30.00	20.00	30.00
		Part B	
Solution (i) Bismuth Sulphite Glucose Phosphate mixture			
Bismuth ammonio- citrate scales	–	6.00	–
Sodium sulphite	–	20.00	–
Disodium hydrogen phosphate	–	20.00	–
Glucose (Dextrose)	–	10.00	–
		Part C	
Solution (ii) Iron citrate brilliant green mixture			
Ferric citrate, brown scales	–	0.40	–
Brilliant green	–	0.05	–
Grams/litre	60.00	As per direction	60.00
Final pH (at 25°C)	7.3± 0.2	7.4± 0.2	7.3± 0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C- 15 min	Part A Autoclaving 121°C for 15 min or as per validaed cycle Part B Boiling Part C Sterile purified / distilled water	Autoclaving 121°C- 15 min

#Equivalent to Beef extract



M331 Wilson and Blair's BBS Agar
Salmonella Typhimurium 14028 (00031*)
*Corresponding WDCM nos.

Wilson and Blair's BBS Agar M331

Type of specimen

Clinical : faeces; Food samples; Water samples; Pharmaceutical samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3, 4).

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (5).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards(6).

For pharmaceutical samples follow appropriate techniques for sample collection, processing as per Pharmaceutical standards (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Though this medium is selective for *Salmonella* other species of *Enterobacteriaceae* may grow.
2. *Salmonella* Typhi and *Shigella* species may not grow on this medium.
3. Moreover *Proteus*, *Pseudomonas* and *Citrobacter* species may mimic enteric pathogens by producing small red colonies.
4. Further confirmation has to be carried out on presumptive *Salmonella* isolates.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

MM331:

Part A : Cream to yellow homogeneous free flowing powder.
Part B : White to cream homogeneous free flowing powder
Part C : Green crystalline granules

M331 / MV331

Cream to yellow homogeneous free flowing powder.

Gelling

MM331 : Firm comparable with 2.0% Agar gel.

M331 / MV331 : Firm comparable with 3.0% Agar gel.

Colour and Clarity of prepared medium

Greenish yellow coloured opaque gel forms in Petri plates.

pH

M331 / MV31 : 7.30 ± 0.2

MM331 : 7.40 ± 0.2

Growth Promotion Test

Growth Promotion is carried out in accordance with IP. Cultural response was observed after an incubation at 30-35°C for 24-48 hours. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

Cultural Response

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Growth promoting+ Indicative				
Salmonella Typhimurium 14028 (00031*)	50 -100	good-luxuriant	≥50%	Green colonies with black centres (uniformly black in 48 hours)
Salmonella Abony NCTC 6017 (00029*)	50 -100	good-luxuriant	≥50%	Green colonies with black centres (uniformly black in 48 hours)
Inhibitory				
Escherichia coli 8739 (00012)*	$\geq 10^3$	inhibited	0%	-
Shigella boydii 9207	$\geq 10^3$	inhibited	0%	-

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10 - 30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. The Indian Pharmacopoeia 2018, Goverment of India 2010, The Controller of Publication.
2. Wilson W. J. and Blair E. M., 1926, J. Pathol. Bacteriol., 29 : 310.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
5. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
6. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.

Antibiotic Assay Media

Antibiotic Assay Media are used for determining the potency of antibiotic using microbiological assay technique The activity (potency) of an antibiotic can be demonstrated under suitable conditions by its inhibitory effect on microorganisms. Reduction in antimicrobial activity may reveal changes not demonstrated by chemical methods. Antibiotic assays are performed by the cylinder plate method and the turbidimetric "tube" assay.

The cylinder plate method, first described by Abraham et al (1) for the assay of penicillin, was later modified by Foster and Woodruff (2) and by Schmidt and Moyer (3). Antibiotic assay media are prepared according to the specifications of the USP, European Pharmacopoeia, British Pharmacopoeia and Indian Pharmacopoeia. The antibiotic media are identified numerically with names assigned by Grove and Randall in Assay Methods of Antibiotics (4).

1. Cylinder plate method:

This method was first devised by Abraham et al (1) and later modified by Schmidt and Moyer (3) and it depends upon diffusion of the antibiotic from vertical steel cylinders placed on the surface of inoculated agar medium. This produces zones of inhibition around the cylinder containing antibiotic solution depending upon the concentration of the antibiotic. This method is commonly employed in the assay of pharmaceutical preparations of Penicillin and other antibiotics. For assay, use Petri plates with 20 X 100 mm dimension and stainless steel or porcelain cylinders with the outside diameter 8 mm, inside diameter 6 mm and length 10 mm. All dimensions should have a tolerance of 0.1 mm. The cylinders should be carefully cleaned to remove all the impurities. For assays requiring base and seed layer, the base layer is allowed to solidify first and then overlaid with the seed agar containing the proper concentration of the test organism. Most assays require base layer of 21 ml and seed layer of 4 ml. Generally 6 Cylinders are used per plate. The cylinders are placed on inoculated plates at equal distance.

2. Punched-hole method:

Holes are punched out of the inoculated culture medium and the antibiotic solutions are then pipetted into them. Rest of the procedure is similar to the cylinder plate method.

3. Paper-disc method:

Paper discs with a diameter of 9 mm are impregnated with the antibiotic solution and placed on the culture medium. Antibiotic can also be applied to the disc after it has been placed on the medium. Plates containing a single layer of medium with 2 mm thickness may be used for these tests. All other steps are similar to the cylinder plate method. Antibiotic Agar No. 2 or 5 can be employed depending on the antibiotic assayed.

4. Serial dilution method:

Minimum inhibitory concentration (MIC) of an antibiotic can be expressed by determining the antibiotic activity quantitatively. It can be done by using the known sensitivity of a test organism towards a particular antibiotic. Serial dilutions of an antibiotic to be tested are pipetted into the antibiotic broth which is then inoculated with a defined quantity of the relevant test organism. The last tube which does not show any turbidity due to suppression of microbial growth indicates the presence of active antibiotic at a concentration corresponding to MIC.

5. Turbidimetric Assay:

The turbidimetric method depends upon the inhibition of test organism in a medium containing uniform solution of an antibiotic. This method has an advantage over the "Cylinder plate method" in that it requires shorter incubation period of 3-4 hours. Use 18x150 mm test tubes that are free from impurities. In this method, working dilutions of the antibiotic reference standards are prepared in specific concentrations. Add 9 ml of inoculated broth to one ml quantities of these solutions in test tubes. Similarly solutions of sample under test containing approximately the same antibiotic activity are simultaneously tested. The tubes are then incubated for 3-4 hours at the specified temperature in a water bath. After the incubation period, the growth is stopped by addition of one drop of formalin and the amount of growth is determined by measuring the light transmittance with a suitable Spectrophotometer. The concentration of the antibiotic is determined by comparing amounts of growth obtained with that given by the reference standard solutions.

Preparation of Inoculum

The test organisms are maintained on agar slants and transferred at 2 weeks interval. On the day of the assay, prepare the stock suspension of the test organism by suspending the growth in sterile saline. This stock suspension is then diluted to contain the desired concentration of the test organism.

Note

Antibiotic assay medium is prepared in distilled water as per Indian Pharmacopoeia, using purified water as per United States Pharmacopoeia and using R-water as per European Pharmacopoeia and British Pharmacopoeia.

Unless otherwise indicated the media should be sterilized by heating in an autoclave at 121°C for 15 minutes

References

1. Abraham, Chain, Fletcher, Florey, Gardner, Heatley and Jennings. 1941. Lancett ii:177.
2. Foster and Woodruff. 1943. J. Bacteriol. 46:187.
3. Schmidt and Moyer. 1944. J. Bacteriol. 47:199.
4. Grove and Randall. 1955. Assay methods of antibiotics. Medical Encyclopedia, Inc. New York, N.Y.

For users convenience, HiMedia has categorized media as :

Code starting with only M - HiMedia codes for general use Media
Code starting with MU - For Media in accordance to United States Pharmacopoeia
Code starting with MM - For Media in accordance to Indian Pharmacopoeia
Code starting with ME - For Media in accordance to European Pharmacopoeia
Code ending with a suffix B (i.e MxxxB) - For Media in accordance to British Pharmacopoeia
Codes starting with GM - Granulated Media
Code starting with only MV - For general use Media (HiVeg™)
Code starting with only MCD - For Chemically Defined Media

For detailed direction, principle and interpretation, and other relevant details, please refer to HiMedia Manual or contact at info@himedialabs.com for technical data for individual media.

Antibiotic Assay Medium No.1 M003

Intended Use:

Antibiotic Assay Medium No.1 is used for microbiological assay of ß-lactam and other antibiotics.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice : Recommended for the microbiological assay as per specified.

Ingredients	HiMedia	USP	EP	BP	IP	HiVeg™
	M003	MU003	ME003	M003B	MM003	MV003
Peptone	6.00	6.00	6.00	6.00	6.00	_
Tryptone#	4.00	4.00	4.00	4.00	4.00	_
HM peptone B##	1.50	1.50	1.50	1.50	1.50	_
HiVeg™ peptone	_	_	_	_	_	6.00
HiVeg™ hydrolysate	_	_	_	_	_	4.00
HiVeg™ extract	_	_	_	_	_	1.50
Yeast extract	3.00	3.00	3.00	3.00	3.00	3.00
Dextrose (Glucose)	1.00	1.00	_	_	1.00	1.00
Glucose monohydrate	_	_	1.00	1.00	_	_
Agar	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	30.50	30.50	30.40	30.40	30.50	30.50
Final pH (at 25°C)	6.6 ± 0.2	_	7.0 ± 0.1	_	_	6.6 ± 0.2
pH after sterilization (at 25°C)	_	6.6 ± 0.1	_	6.6 ± 0.1	6.6 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

Pancreatic digest of casein
Equivalent to Beef extract

Principle And Interpretation

This medium is also used as inoculum and maintenance medium for different test organisms for antibiotic assays. Composition of this medium is in accordance with US Pharmocopoeia (1) and is recommended by FDA(2) and identified numerically with the name assigned by Grove and Randall (3). This medium is also recommended by EP (6), BP (7) and IP (8).

Essential nutrients, vitamins, mineral, trace elements and growth factors are supplied by peptone, tryptone, yeast extract, HM peptone B, HiVeg™ peptone, HiVeg™ hydrolysate and HiVeg™ extract. Dextrose (Glucose) in the medium serves as the carbon source for stimulating the growth of the test microorganism. Agar provides excellent medium for antibiotic diffusion and gives well defined zones of inhibition. Freshly prepared plates should be preferably used for assaying antibiotics. Test organisms is inoculated in sterile seed agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar. All conditions in the microbiological assay must be controlled carefully. One of the critical and important step for obtaining good results is use of appropriate standard culture media.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 6, 7, 8).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Antibiotic Assay Medium No.1 M003

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Yellow coloured slightly opalescent gel forms in Petri plates.

pH

MU003 / M003B / MM003 - 6.60 ± 0.1

ME003 - 7.0 ± 0.1

M003 / MV003 - 6.6 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at specified temperature and period.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Inoculum medium	Incubation temperature / period	Anti-biotics assayed
Bordetella bronchiseptica 4617	50 -100	good-luxuriant	≥50%	Colistimethate sodium, Colistin, Polymyxin B	32-35°C/ 24 hours	
Escherichia coli 10536	50 -100	luxuriant	≥70%	Chloramphenicol	32-35°C/ 24 hours	
Klebsiella pneumoniae 10031	50 -100	good-luxuriant	≥50%	Capreomycin, Dihydro-streptomycin, Neomycin, Streptomycin, Troleandomycin	36-37.5°C/ 16-24 hours	
Micrococcus luteus 9341	50 -100	luxuriant	≥70%	Erythromycin	32-35°C/ 24 hours	
Micrococcus luteus 10240	50 -100	good-luxuriant	≥70%	Bacitracin	32-35°C/ 24 hours	Bacitracin
Pseudomonas aeruginosa 25619	50 -100	luxuriant	≥70%	Carbenicillin	36-37.5°C/ 24 hours	
Staphylococcus epidermidis 12228 (00036*)	50 -100	good-luxuriant	≥70%	Gentamicin, Netilmicin, Neomycin, Novobiocin, Paromomycin, Sisomycin	32-35°C/ 24 hours	Novobiocin
Staphylococcus aureus 29737	50 -100	luxuriant	≥70%	Amikacin, Cephalothin, Cephaperin, Chlortetracycline, Cloxacillin, Cycloserine, Demeclocycline, Kanamycin, Methacycline, Nafcillin, Penicillin-G, Rolitetracycline, Tetracycline, Tobramycin, Tylosin	32-35°C/ 24 hours	Cephalothin, Cephaperin, Cloxacillin, Nafcillin, Penicillin-G,

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. United States Pharmacopoeia USP 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436,Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc. New York.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
6. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines.
7. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
8. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.

Antibiotic Assay Medium No.2 M005

Intended Use:

Antibiotic Medium No.2 is used as basal medium for microbiological assay of antibiotics.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Ingredients	HiMedia	USP	IP	HiVeg™
	M005	MU005	MM005	MV005
Peptone	6.00	6.00	6.00	_
Yeast extract	3.00	3.00	3.00	3.00
HiVeg™ Peptone	_	_	_	6.00
HiVeg™ extract	_	_	_	1.50
HM peptone B#	1.50	1.50	1.50	_
Agar	15.00	15.00	15.00	15.00
Grams/litre	25.50	25.50	25.50	25.50
Final pH (at 25°C)	6.6 ± 0.2	_	_	6.6 ± 0.2
pH after sterilization (at 25°C)	_	6.6 ± 0.1	6.55 ± 0.05	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

This medium is commonly used as base agar for microbiological agar diffusion assays for wide variety of antibiotics. Agar diffusion assays can be performed by cylinders, punched-hole or paper disc tests. This medium is identical numerically with the name assigned by Grove and Randall (1). This medium is prepared according to the specifications detailed in the USP and CFR (2, 3). It is also recommended by IP (4).

Peptone, yeast extract, HM peptone B, HiVeg™ peptone and HiVeg™ extract provides nitrogeneous and carbonaceous compounds, vitamins and minerals required for the growth of test organisms. This medium provides solidified substratum for growth of organisms and supports the overlayering of soft agar.

To perform an antibiotic assay the Antibiotic assay medium No.2 is used as Base Agar. This medium should be prepared on the same day as the test. For the cylinder method, a base layer of 21 ml is required. Once the base medium has solidified, Antibiotic assay medium No.1 as seed agar, inoculated with the standardized culture can be overlaid. Even distribution of the layer is important.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2, 4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Antibiotic Assay Medium No.2 M005

Quality Control

Appearance
Cream to yellow coloured homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Amber coloured slightly opalescent gel forms in Petri plates.

pH
M005 / MV005 - 6.60 ± 0.2

MU005 - 6.60 ± 0.1

MM005 - 6.55 ± 0.05

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

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Basal layer
Micrococcus luteus 10240	50 -100	luxuriant	≥70%	Bacitracin
Staphylococcus aureus 9144 (00035*)	50 -100	luxuriant	≥70%	Tylosin
Staphylococcus aureus 29737	50 -100	luxuriant	≥70%	Amikacin, Cephalothin, Cephapirin, Cloxacillin, Cycloserine, Chlortetracycline, Demeclocycline, Doxycycline, Kanamycin, Methacycline, Nafcillin, Oxytetracycline, Rolitetracycline, Tetracycline
Staphylococcus epidermidis 12228 (00036*)	50 -100	good-luxuriant	≥70%	Novobiocin
Klebsiella pneumoniae 10031	50 -100	luxuriant	≥70%	Capreomycin, Streptomycin, Troleandomycin
Enterococcus hirae 10541 (00011*)	50 -100	luxuriant	≥70%	Gramicidin, Thiostrepton, Tobramycin
Escherichia coli 10536	50 -100	luxuriant	≥70%	Chloramphenicol, Spectinomycin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc. New York.1.
2. United States Pharmacopoeia / National Formulary 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
3. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436,Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
4. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.3 M042

Intended Use:

Antibiotic Assay Medium No.3 is used as the broth medium in turbidimetric or serial dilution assay of a wide variety of antibiotics in accordance with United States Pharmacopoeia and Indian Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Advice : Recommended for the microbiological assay as per specified.

Ingredients	HiMedia	USP	IP	HiVeg™
	M042	MU042	MM042	MV042
Peptone	5.00	5.00	5.00	_
HM peptone B#	1.50	1.50	1.50	_
Yeast extract	1.50	1.50	1.50	1.50
HiVeg™ peptone	_	_	_	5.00
HiVeg™ extract	_	_	_	1.50
Dextrose (Glucose)	1.00	1.00	1.00	1.00
Sodium chloride	3.50	3.50	3.50	3.50
Dipotassium hydrogen phosphate	3.68	3.68	3.68	3.68
Potassium dihydrogen phosphate	1.32	1.32	1.32	1.32
Grams/litre	17.50	17.50	17.50	17.50
Final pH (at 25°C)	7.0 ± 0.2	_	_	7.0 ± 0.2
pH after sterilization (at 25°C)	_	7.0 ± 0.05	7.0 ± 0.05	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

Grove and Randall have elucidated the antibiotic assays and medias in their comprehensive treatise on antibiotic assays (1). Antibiotic assay Medium No. 3 is used as the broth medium in turbidimetric or serial dilution assay of a wide variety of antibiotics. This medium is formulated in accordance with The United States Pharmacopoeia (2). It is also recommended by Indian Pharmacopoeia (3).

Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganims in a liquid medium containing a uniform concentration of an antibiotic. After incubation of the test organism in the working dilutions of the antibiotics, the amount of growth is determined by measuring the light transmittance using spectrophotometer. The concentration of antibiotic is determined by comparing amounts of growth obtained with that given by the reference standard solutions. Use of this method is appropriate only when test samples are clear.

Peptone, HM peptone B, HiVeg™ peptone, HiVeg™ extract and yeast extract provide nitrogenous carbonaceous compounds, nutrients, vitamins and growth factors for enhanced microbial growth. Sodium chloride maintains the osmotic equilibrium and retains the cell viability and cell intergrity. Phosphates in the medium provide good buffering action. Dextrose (Glucose) serves as the carbon and energy source for luxuriant growth.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2, 3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Antibiotic Assay Medium No.3 M042

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate.

pH

M042 / MV042 - 7.0 ± 0.2

MU042 / MM042 - 7.0 ± 0.05

Cultural Response

Cultural characteristics observed after incubation at specified temperature.

Organism (ATCC)	Inoculum (CFU)	Growth	Serial dilution with	Incubation temperature / period
Escherichia coli 10536	50 -100	luxuriant	Chloramphenicol	32-35°C / 24 hours
Klebsiella pneumoniae 10031	50 -100	luxuriant	Capreomycin, Dihydrostreptomycin, Streptomycin, Troleandomycin	36-37.5°C / 16-24 hours
Staphylococcus aureus 29737	50 -100	luxuriant	Amikacin, Chlortetracycline, Cycloserine, Demeclocycline, Doxycycline, Kanamycin, Lincomycin, Methacycline, Oxytetracycline, Rolitetracycline, Tetracyclin, Tobramycin	32-35°C/ 24 hours
Enterococcus hirae 10541 (00011*)	50 -100	luxuriant	Gramicidin	36-37.5°C / 16-18 hours
Staphylococcus aureus 9144 (00035*)	50 -100	luxuriant	Tylosin	35-39°C/16-18 hours

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics, Medical Encyclopedia, Inc. New York
2. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
3. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.4 M140

Intended Use:

Antibiotic Assay Medium No. 4 is used for detection of Penicillin in milk samples and in microbiological assay of different antibiotics in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Ingredients	HiMedia	USP	HiVeg™
	M140	MU140	MV140
Peptone	6.00	6.00	_
HiVeg™ peptone	_	_	6.00
Yeast extract	3.00	3.00	3.00
HM peptone B#	1.50	1.50	_
HiVeg™ extract	_	_	1.50
Dextrose (Glucose)	1.00	1.00	1.00
Agar	15.00	15.00	15.00
Grams/litre	26.50	26.50	26.50
Final pH (at 25°C)	6.6 ± 0.2	_	6.6 ± 0.2
pH after sterilization (at 25°C)	_	6.6 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

This dehydrated culture medium is suitable for plate counts in pharmaceutical and related products and for the microbial assay and detection of antibiotics like penicillin in milk. This medium is formulated in accordance to the specifications and procedures listed by the Food and Drug Administration and USP (1, 2).This medium is identical numerically with name assigned by Grove and Randall (3).

Peptone, yeast extract, HM peptone B, HiVeg™ peptone and HiVeg™ extract provides nitrogenous, carbonaceous compounds, long chain amino acids, vitamins and other nutritional requirement for growth of the indicator organims like *Bacillus stearothermophilus, Micrococcus luteus*. This medium is similar to Antibiotic assay medium no. 2 except for the additional ingredient Dextrose (Glucose) which serves as an easily available source of carbon stimulating luxuriant growth of the test organisms.

Generally presence of penicillin in milk is detected by the cylinder plate method, using *Micrococcus luteus* as the test organism, and by paper disk method, using *Bacillus stearothermophilus*. The cylinder plate method is recommended as the standard for quantification of ß-lactam residues. A description of the cylinder plate method for detecting penicillin in dry powdered milk is given by Kramer et al. (4). The same basic procedure is also recommended to the assay of penicillin in fluid milk.

Freshly prepared plates should be used for antibiotic assays. The use of this medium assures well defined zones of the test organism. All conditions in the microbiological assay must be controlled carefully. The use of standard culture medium in the test is one of the important steps for obtaining good results.

Type of specimen

Pharmaceutical samples.

Antibiotic Assay Medium No.4 M140

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder.

Gelling

Firm, comparable with 1.5% Agar gel.

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

M140 / MV140 - 6.60 ± 0.2

MU140 - 6.6 ± 0.1

Cultural Response

Growth Promotion is carried out in accordance with USP. Cultural characteristics observed after an incubation at 32-35°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Incubation temperature	Incubation period
Micrococcus luteus 10240	50 -100	good-luxuriant	≥50%	32-35°C	18-24 hours
Bacillus stearothermophilus 7953	50 -100	good-luxuriant	≥50%	55°C	18-24 hours

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
2. United States Pharmacopoeia/National Formulary 2019, US Pharmacopoeial Convention, Inc.,Rockville, MD.
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc. New York.
4. Kramer, J., G.G. Carter, B. Arret, J. Wilner, W.W. Wright, and A. Kirshbaum. 1968. Antibiotic residues in milk, dairy products and animal tissues: methods, reports and protocols. Food and Drug Administration, Washington, DC.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.5 M006

Intended Use:

Antibiotic Assay Medium No. 5 is used for microbiological assay of Dihydrostreptomycin using *Bacillus subtilis* in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice : Recommended for the Microbiological assay of Dihydrostreptomycin .

Ingredients	HiMedia	USP	IP	HiVeg™
	M006	MU006	MM006	MV006
Peptone	6.00	6.00	6.00	_
HiVeg™ peptone	_	_	_	6.00
Yeast extract	3.00	3.00	3.00	3.00
HM peptone B#	1.50	1.50	1.50	_
HiVeg™ extract	_	_	_	1.50
Agar	15.00	15.00	15.00	15.00
Grams/litre	25.50	25.50	25.50	25.50
Final pH (at 25°C)	7.9 ± 0.2	_	_	7.9 ± 0.2
pH after sterilization (at 25°C)	_	7.9 ± 0.1	7.9 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

This medium is used in the assay of commercial preparations of antibiotics, as well as for antibiotics in body fluids, feeds etc. Medium composition is in accordance to the specifications detailed in the USP, FDA and IP (1, 2, 5) and numerically identical to the name assigned by Grove and Randall (3).

Peptone, yeast extract, HM peptone B, HiVeg™ peptone and HiVeg™ extract provides nitrogenous, carbonaceous compounds, long chain amino acids and other necessary growth nutrients for the test organisms like *Bacillus subtilis*. This medium provides solidified substratum for growth of organims. The pH 7.9 maintained in this medium provides optimum growth conditions for *Bacillus subtilis* (4). This medium is used to prepare the base as well as seed layer in the microbiological assay of Dihydrostreptomycin.

To perform the antibiotic assay, the Base Agar should be prepared on the same, inoculated with the standardized test culture can be overlaid. Even distribution of the layer is important.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 5).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Antibiotic Assay Medium No.5 M006

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Medium amber coloured slightly opalescent gel forms in Petri plates.

pH

M006 / MV006 - 7.90 ± 0.2

MU006 / MM006 - 7.9 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 32-35°C for 5 days.

Organism (ATCC)	Inoculum (CFU)	Growth	Antibiotics assayed
MU006			
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	good-luxuriant	Dihydrostreptomycin
MM006			
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	good-luxuriant	Framycetin, Kanamycin B, Teicoplanin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

Reference

1. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259.
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc. New York.
4. Stearn and Stearn, 1933, J Bacteriol. 26(1): 37-55.
5. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
7. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.6 M223

Intended Use:

Antibiotic Assay Medium No. 6 is used for induction of spore production in *Bacillus subtilis* strains used in antibiotic assays.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Distribute into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	HiVeg™
	M223	MV223
Tryptone	17.00	_
HiVeg™ hydrolysate	_	17.00
Soya peptone	3.00	3.00
Sodium chloride	5.00	5.00
Dextrose (Glucose)	2.50	2.50
Dipotassium hydrogen phosphate	2.50	2.50
Manganese sulphate	0.030	0.030
Grams/litre	30.03	30.03
Final pH (at 25°C)	7.0 ± 0.2	7.0 ± 0.2
Water	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

Principle And Interpretation

Antibiotic Assay media are used in the performance of antibiotic assays. Grove and Randall have elucidated those antibiotic assays and media in their comprehensive treatise on antibiotic assays (1). Schmidt and Moyer have reported the use of antibiotic assay medium for the liquid formulation used in the performance of antibiotic assay (2). These media are prepared as per FDA (3).

Tryptone, HiVeg™ hydrolysate and soya peptone provides nitrogenous, carbonaceous compounds, long chain amino acids, vitamins and other nutrients and growth factors. Dextrose (Glucose) provides as energy source. Dipotassium phosphate provides the buffering system. Manganese sulphate helps in the early initiation of spore *Bacillus* species.

Type of specimen

Bacillus subtilis culture.

Specimen Collection and Handling

Follow appropriate techniques for sample collection, processing as per guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Antibiotic Assay Medium No.6 M223

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Light amber coloured clear solution may contain slight precipitate.

pH
7.00 ± 0.2

Cultural Response
Cultural characteristics observed after an incubation at different temperatures for 6 days.

Organism (ATCC)	Inoculum (CFU)	Growth	Incubated at	Spores
Bacillus cereus 10876	50 -100	luxuriant	30°C	positive
Bacillus stearothermophilus 7953	50 -100	luxuriant	55°C	positive
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	30°C	positive
Bacillus pumilus 14884	50 -100	luxuriant	30°C	positive

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc, New York.
2. Schmidt and Moyer, 1944; J. Bact, 47:199.
3. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983. Title 21, part 436, Subpart D, Washington, D.C. U.S Government printing office, paragraphs 436, 100-436, 106 pg 242-259 (April 1).
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.8 M041

Intended Use:

Antibiotic Assay Medium No. 8 is used for microbiological assay of Vancomycin in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice: Recommended for the Microbiological assay of Vancomycin, Oxytetracycline and Tetracycline.

Ingredients	HiMedia	USP	IP	HiVeg™
	M041	MU041	MM041	MV041
Peptone	6.00	6.00	6.00	_
HiVeg™ peptone	_	_	_	6.00
Yeast extract	3.00	3.00	3.00	3.00
HM peptone B#	1.50	1.50	1.50	_
HiVeg™ extract	_	_	_	1.50
Agar	15.00	15.00	15.00	15.00
Grams/litre	25.50	25.50	25.50	25.50
Final pH (at 25°C)	5.9 ± 0.2	_	_	5.9 ± 0.2
pH after sterilization (at 25°C)	_	5.9 ± 0.1	5.9 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

The composition of this medium is in accordance to USP and CFR (1, 2) and identical numerically with the name assigned by Grove and Randall (3). This medium also recommended by IP (5).

Peptone, yeast extract, HM peptone B, HiVeg™ peptone and HiVeg™ extract provides nitrogenous and carbonaceous compounds, long chain amino acids and other essential growth nutrients for the test organims. This medium provides solidified substratum for growth of organisms. This medium provides the optimal pH 5.9 for assay of tetracycline as these antibiotics are stable at slightly lower pH (4). This pH condition also supports the growth of test organisms. This medium is also used as base and seed agar medium for agar diffusion assay for mitomycin, mithramycin, plicamycin and Vancomycin (5).

To perform the antibiotic assay the Base Agar should be prepared on the same day as the test. The potency of an antibiotic can be demonstrated by its inhibitory effect on microorganisms under suitable conditions. For the cylinder method, a base layer of 21 ml is required. Once the base medium has solidified, seed layer inoculated with the standardized test culture can be overlaid. Even distribution of the layer is important.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 5).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Antibiotic Assay Medium No.8 M041

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm,comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light amber coloured slightly opalescent gel forms in Petri plates.

pH

M041 / MV041 - 5.90 ± 0.2

MM041 / MU041 - 5.90 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 32 - 35°C for 18 24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
MU041				
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	≥70%	Vancomycin
MM041				
Bacillus cereus var mycoides 11778 (00001*) 32-35°C / 5 days	50 -100	luxuriant	≥70%	Oxytetracycline, Tetracycline

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

Reference

1. United States Pharmacopoeia/National Formulary 2019 US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April1).
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc. New York.
4. Chapin-Robertson and Edberg, 1991, Measurement of Antibiotics in Human Body fluids: Techniques and significance. Antibiotics in Laboratory medicine, New York 311.
5. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
7. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.9 M147

Intended Use:

Antibiotic Assay Medium No. 9 is used as Base layer for plate assay of Carbenicillin, Colistimethate sodium and Polymyxin B in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice: Recommended for the microbiological assay of Carbenicillin, Colistimethate sodium and Polymyxin B .

Ingredients	HiMedia	USP	HiVeg™
	M147	MU147	MV147
Tryptone#	17.00	17.00	_
HiVeg™ hydrolysate	_	_	17.00
Soya peptone##	3.00	3.00	3.00
Dextrose (Glucose)	2.50	2.50	2.50
Sodium chloride	5.00	5.00	5.00
Dipotassium hydrogen phosphate	2.50	2.50	2.50
Agar	20.00	20.00	20.00
Grams/litre	50.00	50.00	50.00
Final pH (at 25°C)	7.2 ± 0.2	_	7.2 ± 0.2
pH after sterilization (at 25°C)	_	7.2 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Pancreatic digest of casein
##Papaic digest of soybean

Principle And Interpretation

The composition of this medium is in accordance to USP and CFR. This medium is widely recommended for assay of Polymyxin B, Colistimethate sodium and Colistin using *Bordetella bronchiseptica* as test organims. Carbenicillin assay is also performed using this medium with *Pseudomonas aeruginosa*. The medium is formulated in accordance with USP and CFR (1,2) and numerically identical with the name assigned by Groove and Randall (3).

Tryptone, Soya peptone and HiVeg™ hydrolysate provides carbonaceous and nitrogenous compounds, long chain amino acids, vitamins and other essential nutrients for growth of organisms. Dextrose (Glucose) stimulates the growth by providing carbon and energy. Phosphates in the medium enhance buffering action and sodium chloride maintains osmotic equilibrium in the medium. Agar concentration provides control over the diffusion activity of Polymyxin B antibiotics and provides solid substratum to support the seed agar layer.

To perform the antibiotic assay the Base Agar should be prepared on the same day as the test. For the cylinder method, a base layer of 21 ml is required. Once the base medium has solidified, seed layer inoculated with the standardized culture can be overlaid. Even distribution of the layer is important.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Antibiotic Assay Medium No.9 M147

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm, comparable with 2.0% agar gel.

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri plates.

pH

M147 / MV147 - 7.2 ± 0.2

MU147 - 7.2 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 36-37.5°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Bordetella bronchiseptica 4617	50 -100	luxuriant	≥50%	Polymyxin B, Colistimethate sodium, Colistin
Pseudomonas aeruginosa 25619	50 -100	luxuriant	≥70%	Carbenicillin

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. United States Pharmacopoeia, 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April1).
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc. New York.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.10 M225

Intended Use:

Antibiotic Assay Medium No. 10 is used as seed layer for antibiotic plate assay of Carbenicillin, Colistimethate sodium, Colistin sulphate and Polymyxin B.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water containing 10 ml of Polysorbate 80. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Coolt to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Ingredients	HiMedia	USP	IP	HiVeg™
	M225	MU225	MM225	MV225
Tryptone #	17.00	17.00	17.00	_
HiVeg™ hydrolysate	_	_	_	17.00
Soya peptone ##	3.00	3.00	3.00	3.00
Dextrose (Glucose)	2.50	2.50	2.50	2.50
Sodium chloride	5.00	5.00	5.00	5.00
Dipotassium hydrogen phosphate	2.50	2.50	2.50	2.50
Agar	12.00	12.00	12.00	12.00
Grams/litre	42.00	42.00	42.00	42.00
Final pH (at 25°C)	7.2 ± 0.2	_	_	7.2 ± 0.2
pH after sterilization (at 25°C)	_	7.2 ± 0.1	7.2 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Pancreatic digest of casein
##Papaic digest of soyabean

Principle And Interpretation

This medium is used as seed agar for assay of Polymyxin B, Colistimethate sodium, Colistin and Carbenicillin. The medium composition is in accordance to USP, CFR and IP (1, 2, 6) and numerically identical with the name assigned by Groove and Randall (3).

Combination of tryptone, soya peptone and HiVeg™ hydrolysate provides carbonaceous and nitrogenous compounds, long chain amino acids, vitamins and other essential nutrients for the growth of test organisms. Dextrose (Glucose) provides the carbon source, enhances the growth of test organim. Phosphates in the medium enhances buffering action and sodium chloride maintains osmotic equilibrium. Polymyxins are reported to have slow diffusion in agar giving smaller zone of inhibition (4). Hence the reduced agar concentration (1.2%) in this medium improves the diffusion of polymyxin in the medium. Polysorbate 80 is reported to function synergistically with Polymyxins on spheroplasts of *Pseudomonas aeruginosa*. Polysorbate 80 enhances the penetration of Polymyxin to the cytoplasmic membrane and hence is an appropriate ingredient in the medium used for assay of Polymyxin (5).

Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar. All conditions in the microbiological assay must be controlled carefully. The use of standard culture media in the test is one of the important steps for good results.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Antibiotic Assay Medium No.10 M225

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm,comparable with 1.2% Agar gel.

Colour and Clarity of prepared medium

Medium amber coloured clear to very slightly opalescent gel forms in Petri plates.

pH

M225 / MV225 - 7.2 ± 0.2

MU225 / MM225 - 7.2 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 32-37°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
MU225 / MM225				
Bordetella bronchiseptica 4617	50 -100	luxuriant	≥70%	Colistimethate sodium, Colistin, Polymyxin B
Pseudomonas aeruginosa 25619	50 -100	luxuriant	≥70%	Carbenicillin
MM225				
Escherichia coli 10536	50 -100	luxuriant	≥70%	Colistimethate sodium, Colistin sulphate

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (7, 8).

Reference

1. United States Pharmacopoeia / National Formulary 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc. New York.
4. Barry, 1991, Procedure and theoretical considerations for testing antimicrobial agents in agar media. Antibiotics in Laboratory medicine, New York pp 3
5. Brown & Winsley, 1968. J Gen Microbiol. 1968 50(3) Suppl:ix.
6. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.11 M004

Intended Use:

Antibiotic Assay Medium No.11 is used for microbiological assay of antibiotics in accordance with various pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice: Recommended for the microbiological assay of antibiotics.

Ingredients	HiMedia	Granulated	USP	EP	BP	IP	HiVeg™
	M004	GM004	MU004	ME004	M004B	MM004	MV004
Peptone	6.00	6.00	6.00	6.00	6.00	6.00	_
Tryptone#	4.00	4.00	4.00	4.00	4.00	4.00	_
Yeast extract	3.00	3.00	3.00	3.00	3.00	3.00	3.00
HiVeg™ Peptone	_	_	_	_	_	_	6.00
HiVeg™ hydrolysate	_	_	_	_	_	_	4.00
HM peptone B##	1.50	1.50	1.50	1.50	1.50	1.50	_
HiVeg™ extract	_	_	_	_	_	_	1.50
Dextrose (Glucose)	1.00	1.00	1.00	_	_	1.00	1.00
Glucose monohydrate	_	_	_	1.00	1.00	_	_
Agar	15.00	15.00	15.00	15.00	15.00	15.00	15.00
Grams/litre	30.50	30.50	30.50	30.40	30.40	30.50	30.50
Final pH (at 25°C)	8.3 ± 0.2	8.3 ± 0.2	_	_	_	_	8.3 ± 0.2
pH after sterilization (at 25°C)	_	_	8.3 ± 0.1	7.9 ± 0.1	7.9 ± 0.1	7.9 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Pancreatic digest of casein
##Equivalent to Beef extract

Principle And Interpretation

This medium is formulated in accordance to USP and CFR; and is employed to analyze the neomycin content as per FDA and the USP (1, 2). It is indentical numerically with the name assigned by Grove and Randall (3). This medium is also recommended by EP (7), BP (8) and IP (4). This medium provides a pH range of 8.3 while Antibiotic assay medium no.1 provides pH range of 6.5-6.7.

Peptone, tryptone, yeast extract, HM peptone B, HiVeg™ hydrolysate and HiVeg™ extract provides carbonaceous and nitrogenous compounds, long chain amino acids and other essential nutrients, vitamins, mineral, trace elements and growth factors. Dextrose (Glucose) in the medium serves as the carbon source for stimulating the growth of the test microorganism. Agar provides excellent medium for antibiotic diffusion and gives well defined zones of inhibition. Higher pH provides the optimal conditions for activity of antibiotic and also supports the growth of test organims.

Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar. All conditions in the microbiological assay must be controlled carefully.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 4, 7, 8).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Antibiotic Assay Medium No.11 M004

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

GM004 : Cream to yellow granular media

Gelling

Firm,comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

M004 /GM004 / MV004 - 8.3 ± 0.2

MU004 - 8.3 ± 0.1

ME004 / MM004 /M004B - 7.9 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 32-35°C for 24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Anti-biotics assayed
Micrococcus luteus 9341	50 -100	luxuriant	≥70%	Erythromycin
Staphylococcus epidermidis 12228 (00036*)	50 -100	luxuriant	≥70%	Gentamicin, Netilmicin, Neomycin, Sisomicin, Paromomycin
MM004				
Bacillus pumilus 14884 (32-35°C) for 5 days	50 -100	luxuriant	≥70%	Chlortetracycline, Framycetin, Kanamycin sulphate, Erythromycin
Kocuria rhizophila 9341 (00036*) (32-35°C) for 24 hours	50 -100	luxuriant	≥70%	Chlortetracycline, Framycetin, Kanamycin sulphate, Erythromycin
Staphylococcus epidermidis 12228 (00036*) (32-35°C) for 24 hours	50 -100	luxuriant	≥70%	Gentamycin, Neomycin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436,Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April1).
3. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc. New York.
4. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
7. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines.
8. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.

Antibiotic Assay Medium No.12 M280

Intended Use:

Antibiotic Assay Medium No.12 (Nystatin Assay Agar) is used for microbiological assay of Amphotericin B and, Nystatin using *Saccharomyces cerevisiae*.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice: Recommended for the Microbiological assay of Amphotericin B and Nystatin.

Ingredients	HiMedia	HiVeg™
	M280	MV280
Peptone	10.00	_
HiVeg™ peptone	_	10.00
HM peptone B#	2.50	_
HiVeg™ extract	_	2.50
Yeast extract	5.00	5.00
Sodium chloride	10.00	10.00
Dextrose (Glucose)	10.00	10.00
Agar	25.00	25.00
Grams/litre	62.50	62.50
Final pH (at 25°C)	6.1 ± 0.2	6.1 ± 0.2
Water	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

This medium is prepared from the Groove and Randall formula (1) Antifungal antibiotics like Amphotericin B and Nystatin can be assayed using this medium.

Peptone, Yeast extract, HM peptone B, HiVeg™ peptone and HiVeg™ extract provides carbonaceous and nitrogenous compounds, long chain amino acids, vitamins, minerals and other essential nutrients for the growth of test organism. Dextrose (Glucose) in the medium provides enhanced source of carbon and energy. Osmotic equilibrium in the medium is by sodium chloride which maintain the cell intergrity and viability.

Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterilised agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar. Prediffusion of antibiotics for 10-20 mins in the agar by incubating at temperature below the optimal growth temperature for microorganism would facilitate better diffusion of antibiotics followed by incubation of plates for microbial growth.

Type of specimen

Saccharomyces cerevisiae culture.

Specimen Collection and Handling

Follow appropriate techniques for sample collection, processing as per guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Antibiotic Assay Medium No.12 M280

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
Cream to yellow coloured homogeneous free flowing powder

Gelling
Firm,comparable with 2.5% Agar gel

Colour and Clarity of prepared medium
Light amber coloured slightly opalescent gel forms in Petri plates.

pH
6.1 ± 0.2

Cultural Response
Cultural characteristics observed after an incubation at 25-30°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Saccharomyces cerevisiae 2601	50 -100	luxuriant	≥70%	Amphotericin B, Nystatin

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2, 3).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc. New York.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.13 M254

Intended Use:

Antibiotic Assay Medium No. 13 is used for the turbidimetric microbiological assay of Candicidin using *Saccharomyces cerevisiae* ATCC 9763 as the test organism and for studying the effectiveness of antibiotics on yeast and molds in accordance with United States Pharmacopeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flask as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	USP	HiVeg™
	M254	MU254	MV254
Peptone	10.00	10.00	_
HiVeg™ peptone	_	_	10.00
Dextrose (Glucose)	20.00	20.00	20.00
Grams/litre	30.00	30.00	30.00
Final pH (at 25°C)	5.6 ± 0.2	_	5.6 ± 0.2
pH after sterilization (at 25°C)	_	5.6 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

Principle And Interpretation

This medium is formulated in accordance to USP and CFR (1,2) and is numerically identical with the name assigned by Groove and Rundall (3). Schmidt & Moyer has reported the use of antibiotic assay medium for liquid formulation in performance of antibiotic assay (4). This medium is widely used in turbidometric assay of antifungals like candicidin using test organism like *Saccharomyces cerevisiae*. This medium is also termed as Sabouraud Liquid Broth Modified or Fluid Sabouraud Medium.

This medium facilitates enhanced growth of test organism *Saccharomyces cerevisiae* employed in assay of candicidin, a polyene antibiotic with antifungal activity. Assay is performed by enumerating the blastospores or by analysing the turbidity of the medium. Dextrose (Glucose) serves as carbon source and peptone provides essential nutrients and growth promoting factors. Optimal pH for growth of *Saccharomyces cerevisiae* is maintained in this medium. Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganism in a liquid medium containing a uniform concentration of an antibiotic. After incubation of the test organism in the working dilutions of the antibiotics, the amount of growth is determined by measuring the light transmittance using spectrophotometer. The concentration of antibiotic is determined by comparing amounts of growth obtained with that given by the reference standard solutions. Use of this method is appropriate only when test samples are clear.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Antibiotic Assay Medium No.13 M254

Quality Control

Appearance
Cream to yellow coloured homogeneous free flowing powder

Colour and Clarity of prepared medium
Light amber coloured clear solution without any precipitate

pH
M254 / MV254 - 5.6 ± 0.2

MU254 - 5.6 ± 0.1

Cultural Response
Cultural characteristics observed after an incubation at 29-31°C for 18-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Serial dilution with
Saccharomyces cerevisiae 9763 (00058*)	50 -100	luxuriant	Candicidin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. United States Pharmacopoeia / National Formulary 2019, US Pharmacopoeial Convention, Inc.,Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436,Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
3. Grove and Randall, 1955, Assay Methods of Antibiotics, Medical Encyclopaedia, Inc. New York
4. Schmidt and Moyer, 1944. J.Bact., 47:199.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.19 M101

Intended Use:

Antibiotic Assay Medium No.19 is used for the microbiological assay of Amphotericin B, Natamyin, Candicin and Nystatin using *Saccharomyces cerevisiae* as the test organisms.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well pour into sterile Petri plates or as desired.

Advice : Recommended in the microbiological assay of Amphotericin B, Natamycin, Candicidin and Nystatin

Ingredients	HiMedia	USP	IP	HiVeg™
	M101	MU101	MM101	MV101
Peptone	9.40	9.40	9.40	_
HiVeg™ peptone	_	_	_	9.40
Yeast extract	4.70	4.70	4.70	4.70
HM peptone B#	2.40	2.40	2.40	_
HiVeg™ extract	_	_	_	2.40
Dextrose (Glucose)	10.00	10.00	10.00	10.00
Sodium chloride	10.00	10.00	10.00	10.00
Agar	23.50	23.50	23.50	23.50
Grams/litre	60.00	60.00	60.00	60.00
Final pH (at 25°C)	6.1 ± 0.2	_	_	6.1 ± 0.2
pH after sterilization (at 25°C)	_	6.1 ± 0.1	6.1 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

The medium composition is in accordance to USP and CFR (1, 2). This medium is used as seed agar for assay of antifungal agents like Amphotericin B and Nystatin. This medium is used for maintenance and inoculum development of *Saccharomyces cerevisiae*. This medium is also used for assaying mycostatic activity in pharmaceutical formulations. This medium is forumulated as reported by Kirshbam and Arret (3). This medium is also recommeded by IP (4).

Ingredients like peptone, yeast extract, HM peptone B, HiVeg™ peptone and HiVeg™ extract provides carbonaceous and nitrogenous compounds, long chain amino acids and other essential nutrients and growth factors for the growth of test organism. Dextrose (Glucose) in the medium provides enhanced source of carbon and energy. Osmotic equilibrium in the medium is maintained by sodium chloride which retains the cell intergrity and viability. Antibiotic assay medium No.19, is used as both base and seed medium for agar diffusion assay for antibiotics like Amphotericin B and Nystatin.

Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar precooled to 40-45°C and spread evenly over the surface of solidified base agar. Prediffusion of antibiotics for 20 minutes in the agar by incubating at temperature below the optimal growth temperature for microorganism would facilitate better diffusion of antibiotic, followed by incubation of the plates for microbial growth.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Antibiotic Assay Medium No.19 M101

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm, comparable with 2.35% Agar gel.

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates

pH

M101 / MV101 - 6.1 ± 0.2

MU101/ MM101 - 6.1 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 29-31°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
MU101				
Saccharomyces cerevisiae 2601	50 -100	luxuriant	≥70%	Nystatin
Saccharomyces cerevisiae 9763 (00058*)	50 -100	luxuriant	≥70%	Amphotericin B, Candicidin
MM101				
Saccharomyces cerevisiae 9763 (00058*)	50 -100	luxuriant	≥70%	Candicidin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
3. Krishbam A and Arret B, 1967, J.Pharma. Sci. 56:512.
4. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.20 M167

Intended Use:

Antibiotic Assay Medium No. 20 is used for the microbiological assay of Amphotericin B using *Candida tropicalis*.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	HiVeg™
	M167	MV167
Tryptone	10.00	_
HiVeg™ hydrolysate	_	10.00
Peptone	5.00	_
HiVeg™ Peptone	_	5.00
Yeast extract	6.50	6.50
HM extract B#	1.50	_
HiVeg™ extract	_	1.50
Dextrose (Glucose)	11.00	11.00
Sodium chloride	3.50	3.50
Dipotassium hydrogen phosphate	3.68	3.68
Potassium dihydrogen phosphate	1.32	1.32
Grams/litre	42.50	42.50
Final pH (at 25°C)	6.6 ± 0.2	6.6 ± 0.2
Water	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

Equivalent to Beef extract

Principle And Interpretation

Antibiotic Assay media are used in the performance of antibiotic assays. Grove and Randall have elucidated those antibiotic assays and media in their comprehensive treatise on antibiotic assays (1). Schmidt and Moyer have reported the use of antibiotic assay medium for the liquid formulation used in the performance of antibiotic assay (2). These media are prepared according by the FDA (3). Antibiotic Assay Medium No. 20 is used for turbidometric assay of Amphotericin B using *Candida tropicalis* ATCC 13803 as test organism. This medium is also known as Yeast MB Broth. This medium is also used in assaying mycostatic activity in pharmaceutical related preparations.

High nutritional content like peptone, tryptone, yeast extract, HM peptone B, HiVeg™ extract and HiVeg™ peptone provides carbonaceous and nitrogenous compounds, long chain amino acids and other essential growth nutrients for growth of *Candida tropicalis*. Dextrose (Glucose) provides carbon and energy for growth of the organism. Osmotic equilibrium to maintain cell intergrity and viability is provided by sodium chloride, while phosphate functions to provide proper buffering action. Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganims in a liquid medium containing a uniform concentration of an antibiotic. After incubation of the test orgainism in the working dilutions of the antibiotics, the amount of growth is determined by measuring the light transmittance using spectophotometer. The concentration of antibiotic is determined by comparing amounts of growth obtained with that given by the reference standard solutions. Use of this method is appropriate only when test samples are clear.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For Pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1,3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Antibiotic Assay Medium No.20 M167

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Medium amber coloured clear solutiion

pH

6.6 ± 0.2

Cultural Response

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

Organism (ATCC)	Growth	Serial dilution wit
Candida tropicalis 13803	luxuriant	Amphotericin B

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopedia, Inc, New York.
2. Schmidt and Moyer, 1944; J. Bact, 47:199.
3. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983. Title 21, part 436, Subpart D, Washington, D.C. U.S Government printing office, paragraphs 436, 100-436, 106 pg 242-259 (April 1).
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
6. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Antibiotic Assay Medium No.32 M1141

Intended Use:

Antibiotic Assay Medium No. 32 is recommended for preparing inoculum of *Bacillus subtilis* to be used as test organism for assaying Dihydrostreptomycin and Vancomycin by plate assay method in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to disslove the medium completely. Sterilise by autoclaving at 15lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice : Recommended for the microbiological assay of Dihydrostreptomycin and Vancomycin.

Ingredients	HiMedia	USP	HiVeg™
	M1141	MU1141	MV1141
Peptone	6.00	6.00	_
HiVeg™ peptone	_	_	6.00
Tryptone#	4.00	4.00	_
HiVeg™ hydrolysate	_	_	4.00
Yeast extract	3.00	3.00	3.00
HM peptone B##	1.50	1.50	_
HiVeg™ extract	_	_	1.50
Dextrose (Glucose)	1.00	1.00	1.00
Manganese sulphate	0.30	0.30	0.3
Agar	15.00	15.00	15.00
Grams/litre	30.80	30.80	30.80
Final pH (at 25°C)	6.6 ± 0.2	_	6.6 ± 0.2
pH after sterilization (at 25°C)	_	6.6 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Pancreatic digest of casein
##Beef extract

Principle And Interpretation

This medium is formulated in accordance to USP and FDA (1,2) and is a modification of Antibiotic assay medium No. 1. This medium is used to develop inoculum of *Bacillus subtilis* for antibiotic assay.

Essential nutrients, vitamins, mineral, trace elements and growth factors are supplied by peptone, tryptone, yeast extract, HM peptone B, HiVeg™ peptone, HiVeg™ hydrolysate and HiVeg™ extract provides nitrogenous and carbonaceous compounds, long chain amino acids, vitamins and other essential nutrients. Dextrose (Glucose) in the medium serves as the carbon source for stimulating the growth of the test microorganism. Manganese sulphate in this medium facilitates the sporulation and growth of *Bacillus subtilis* (3,4,5), which is generally used as test organisms for plate assay of Dihydrostreptomycin and Vancomycin.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium No.32 M1141

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

MU1141 - 6.6 ± 0.1

M1141 / MV1141 - 6.6 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at 32-35°C for 5 days.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	good-luxuriant	≥70%	Dihydrostreptomycin, Vancomycin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

Reference

1. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
3. Vasantha & Freese, 1979, J.Gen.Microbiol. 112:329-336
4. Charney, J., Fisher, W.P. and Hegarty, C.P. 1951. J. Bacteriol. 62:145.
5. Curran, H.R. and Evans, F.R. 1954. J. Bacteriol. 67: 489
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
7. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.34 M797

Intended Use:

Antibiotic Assay Medium No. 34 is used for preparation of suspension of *Mycobacterium smegmatis* used as a test organism for the assay of Bleomycin.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water containing 10 grams of glycerol. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Advice : Recommended for the preparation of suspension in microbiological assay of Bleomycin.

Ingredients	HiMedia	USP
	M797	MU797
Peptone	10.00	10.00
HM peptone B#	10.00	10.00
Sodium chloride	3.00	3.00
Grams/litre	23.00	23.00
Final pH (at 25°C)	7.0 ± 0.2	_
pH after sterilization (at 25°C)	_	7.0 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle

#Equivalent to Beef extract
Peptic digest of animal tissue (Peptone)

Principle And Interpretation

This medium is formulated in accordance with USP and CFR (1,2). This medium is generally employed to prepare *Mycobacterium smegmatis* suspension required for assaying antineoplastic agent like Bleomycin. This medium provides optimal conditions to maintain the viability of the test organism *Mycobacterium smegmatis*.

Peptone and HM peptone B in the medium provides carbonaceous and nitrogenous compounds, long chain amino acids, vitamins and other nutrients essential for growth, while addition of glycerol provides slow and continous supply of carbon and energy source. The osmotic equilibrium for integrity of cell and its viability is maintained in presence of sodium chloride present in this medium.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may result in erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Antibiotic Assay Medium No.34 M797

Quality Control

Appearance
Cream to yellow coloured homogeneous free flowing powder

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

pH
MU797 - 7.00 ± 0.1

M797 - 7.00 ± 0.2

Cultural Response
Cultural characteristics observed after an incubation at 35-37.5°C for 18-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Serial dilution with
Mycobacterium smegmatis 607	50 -100	luxuriant	Bleomycin

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436,Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1).
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.35 M798

Intended Use:

Antibiotic Assay Medium No.35 is used for the microbiological assay of Bleomycin using *Mycobacterium smegmatis* as a test organisms.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water containing 10 gms glycerol. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice : Recommended for the microbiological assay of Bleomycin.

Ingredients	HiMedia	USP	IP	HiVeg™
	M798	MU798	MM798	MV798
Peptone	10.00	10.00	10.00	_
HiVeg™ peptone	_	_	_	10.00
HM peptone B#	10.00	10.00	10.00	_
HiVeg™ extract	_	_	_	10.00
Sodium chloride	3.00	3.00	3.00	3.00
Agar	17.00	17.00	17.00	17.00
Grams/litre	40.00	40.00	40.00	40.00
Final pH (at 25°C)	7.0 ± 0.2	7.0 ± 0.1	_	7.0 ± 0.2
pH after sterilization (at 25°C)	_	_	7.0 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

This medium is formulated in accordance with USP, CFR and IP (1, 2, 3). This medium is employed widely as base agar for agar diffusion assay of Bleomycin using *Mycobacterium smegmatis* .

Peptone, HM peptone B, HiVeg™ peptone and HiVeg™ extract in the medium provides carbonaceous and nitrogenous compounds, long chain amino acids, vitamins and other nutrients essential for growth. Agar provides excellent solid substratum for support and overlayering of seed agar, for the assay of Bleomycin. Addition of glycerol is important for slow and continuous provision of carbon to the test organism.

To perform the antibiotic assay the Base Agar should be prepared on the same day as the test. For the cylinder method, a base layer of 21 ml is required. Once the base medium has solidified, seed layer inoculated with the standardized culture can be overlaid. Even distribution of the layer is important.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium No.35 M798

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm,comparable with 1.7% agar gel.

Colour and Clarity of prepared medium

Medium amber coloured clear to slightly opalescent gel forms in Petri plates.

pH

M798 / MV798 - 7.0 ± 0.2

MU798 / MM798 - 7.0 ± 0.1

Cultural Response

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

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Mycobacterium smegmatis 607	50 -100	luxuriant	≥50%	Bleomycin

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1)
3. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.36 M1666

Intended Use:

Antibiotic assay medium No. 36 is used for cultivation of a wide variety of microorganisms and sterility testing in pharmaceutical procedures in accordance to USP and IP.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Ingredients	HiMedia	USP	IP	HiVeg™
	M1666	MU1666	MM1666	MV1666
Tryptone#	15.00	15.00	15.00	_
HiVeg™ hydrolysate	_	_	_	15.00
Soya peptone##	5.00	5.00	5.00	5.00
Sodium chloride	5.00	5.00	5.00	5.00
Agar	15.00	15.00	15.00	15.00
Grams/litre	40.00	40.00	40.00	40.00
Final pH (at 25°C)	7.3 ± 0.2	_	_	7.3 ± 0.2
pH after sterilization (at 25°C)	_	7.3 ± 0.1	7.3 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Pancreatic digest of casein
Papaic digest of soybean

Principle And Interpretation

Grove and Randall have elucidated the antibiotic assays and medias in their comprehensive treatise on antibiotic assays (1). Antibiotic assay Medium No. 36 is recommended for preparation of inoculum of *Mycobacterium smegmatis* for the assay of Bleomycin. This medium is also used for the cultivation of a wide variety of microorganisms and sterility testing of pharmaceutical preparations. (3). This medium is recommended by The United States Pharmacopoeia (2) and IP (4).

The combination of tryptone, HiVeg™ hydrolysate and soya peptone makes this medium nutritious by providing carbonaceous and nitrogenous compounds, long chain amino acids and vitamins for the growth of microorganisms. Sodium chloride maintains the osmotic balance of the medium.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2, 4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Antibiotic Assay Medium No.36 M1666

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Light yellow coloured clear to slightly opalescent gel forms in Petri plates

pH
M1666 / MV1666 - 7.3 ± 0.2

MM1666 / MU1666 - 7.3 ± 0.1

Cultural Response
Cultural characteristics observed after an incubation at 36 -37.5°C for 18-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Mycobacterium smegmatis 607	50 -100	luxuriant	≥50%	Bleomycin

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics, Medical Encyclopedia, Inc. New York.
2. United States Pharmacopoeia 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
3. Wright and Welch, 1959-60, Antibiotics Ann., 61.
4. Indian Pharmacopoeia, 2018, Ministry of Health and Family Welfare, Govt., of India.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.37 M1667

Intended Use:

Antibiotic assay medium No. 37 is used for cultivation of a wide variety of microorganisms and sterility testing of moulds in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	USP	HiVeg™
	M1667	MU1667	MV1667
Tryptone#	17.00	17.00	_
HiVeg™ hydrolysate	_	_	17.00
Soya peptone##	3.00	3.00	3.00
Dextrose (Glucose)	2.50	2.50	2.50
Sodium chloride	5.00	5.00	5.00
Dipotassium hydrogen phosphate	2.50	2.50	2.50
Grams/litre	30.00	30.00	30.00
Final pH (at 25°C)	7.3 ± 0.2	_	7.3 ± 0.2
pH after sterilization (at 25°C)	_	7.3 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Pancreatic digest of casein,
##Papaic digest of soybean meal

Principle And Interpretation

Grove and Randall have elucidated the antibiotic assays and medias in their comprehensive treatise on antibiotic assays (1). Antibiotic assay Medium No. 37 is recommended for the cultivation of a wide variety of microorganisms and sterility testing of pharmaceutical preparations. This medium is also used for the sensitivity testing by the tube dilution method for antimicrobial agents (3). This medium is formulated in accordance with The United States Pharmacopoeia (2).

Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganisms in a liquid medium containing a uniform concentration of an antibiotic. After incubation of the test organism in the working dilutions of the antibiotics, the amount of growth is determined by measuring the light transmittance using spectrophotometer. The concentration of antibiotic is determined by comparing amounts of growth obtained with that given by the reference standard solutions. Use of this method is appropriate only when test samples are clear.

The combination of Tryptone, HiVeg™ hydrolysate and Soya peptone makes this medium nutritious by providing carbonaceous and nitrogenous compounds, amino acids and long chain peptides for the growth of microorganisms. Dextrose (Glucose) serves as the carbohydrate source and dipotassium phosphate facilitates buffering in the medium. Sodium chloride maintains the osmotic balance of the medium.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium No.37 M1667

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate

pH

M1666 / MV1666 - 7.3 ± 0.2

MU1666 - 7.3 ± 0.1

Cultural Response

Cultural characteristics observed (i) for Bacteria at 30-35°C after 18-48 hours (ii) for Fungi at 20-25°C after 2-5 days.

Organism (ATCC)	Inoculum (CFU)	Growth
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	luxuriant
Escherichia coli 8739 (00012*)	50 -100	luxuriant
Pseudomonas aeruginosa 9027 (00026*)	50 -100	luxuriant (when incubated anaerobically)
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant
Streptococcus pyogenes 19615	50 -100	good-luxuriant
Candida albicans 10231 (00054*)	50 -100	luxuriant
Candida albicans 2091	50 -100	luxuriant
Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics, Medical Encyclopedia, Inc. New York
2. United States Pharmacopoeia 1985 US Pharmacopoeial Convention, Inc.,Rockville, MD.
3. Wright and Welch, 1959-60, Antibiotics Ann., 61.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.38 M799

Intended Use:

Antibiotic Assay Medium No.38 is used for the microbiological assay of Ticarcillin,using *Pseudomonas aeruginosa* as the test organism as per United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Ingredients	HiMedia	USP	HiVeg™
	M799	MU799	MV799
Peptone	15.00	15.00	_
HiVeg™ peptone	_	_	15.00
Soya peptone#	5.00	5.00	5.00
Dextrose (Glucose)	5.50	5.50	5.50
Sodium chloride	4.00	4.00	4.00
L-Cystine	0.70	0.70	0.70
Sodium sulphite	0.20	0.20	0.20
Agar	15.00	15.00	15.00
Grams/litre	45.40	45.40	45.40
Final pH (at 25°C)	7.0 ± 0.2	_	7.0 ± 0.2
pH after sterilization (at 25°C)	_	7.0 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Papaic digest of soybean

Principle And Interpretation

This medium follows the specification of USP and CFR (1, 2) and is routinely employed for agar diffusion assay of Ticarcillin using Gram negative test organisms specially *Psuedomonas aeruginosa*. This medium is used as both base agar and seed agar for assay of Ticarcillin.

Peptone, HiVeg™ peptone and soya peptone provides carbon, nitrogen compounds, long chain amino acids, vitamins and essential nutrients and growth factors for the growth of test organisms. Dextrose (Glucose) serves as carbon source. Sodium chloride maintains the osmotic equilibrium. L-Cystine and sodium sulphite are suphur providers that aids assimilation of sulphur during microbial growth. L-Cystine also acts as growth stimulator and enrich the medium with amino acid source for promoting the growth. The high nutritional content along with high sulfur (cystine and sodium sulphite) content improves growth with chromogenicity of test organism *Pseudomonas.* Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium No.38 M799

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

M799 / MV799 - 7.0 ± 0.2

MU799 - 7.0 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 35-37.5°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Pseudomonas aeruginosa 29336	50 -100	luxuriant	≥70%	Ticarcillin

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. United States Pharmacopoeia / National Formulary (USP21/NF16) 1985, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983 Title 21, Part 436, Subpart D, Washington, D.C.: U.S. Government Printing Office, paragraphs 436, 100-436, 106, p. 242-259, (April 1)
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.39 M1142

Intended Use:

Antibiotic Assay Medium No. 39 is used for the microbiological assay of Neomycin using *Klebsiella pneumoniae* and Tylosin using *Staphylococcus aureus* as the test organism in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	USP	HiVeg™
	M1142	MU1142	MV1142
Peptone	5.00	5.00	_
HiVeg™ peptone	_	_	5.00
Yeast extract	1.50	1.50	1.50
HM peptone B#	1.50	1.50	_
HiVeg™ extract	_	_	1.50
Dextrose (Glucose)	1.00	1.00	1.00
Sodium chloride	3.50	3.50	3.50
Dipotassium hydrogen phosphate	3.68	3.68	3.68
Potassium dihydrogen phosphate	1.32	1.32	1.32
Grams/litre	17.50	17.50	17.50
Final pH (at 25°C)	7.9 ± 0.2	_	7.9 ± 0.2
pH after sterilization (at 25°C)	_	7.9 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Equivalent to Beef extract

Principle And Interpretation

Antibiotic Assay media are used in the performance of antibiotic assays. Grove and Randall have elucidated those antibiotic assays and media in their comprehensive treatise on antibiotic assays (1). Schmidt and Moyer have reported the use of antibiotic assay medium for the liquid formulation used in the performance of antibiotic assay (2). This medium is also recommended by USP (3) and the FDA (4).

Nutrients and growth factors like carbonaceous, nitrogenous compounds and amino acids are provided by ingredients like Peptone, HiVeg™ peptone, HM peptone B and Yeast extract. Dextrose (Glucose) is the source of energy. Sodium chloride maintains the osmotic equilibrium whereas the phosphates act as the buffering system.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium No.39 M1142

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate

pH

M1142 / MV1142 - 7.9 ± 0.2

MU1142 - 7.9 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at specified temperature for specified time.

Organism (ATCC)	Inoculum (CFU)	Growth	Serial dilution with	Incubation temp/ period
Klebsiella pneumoniae 10031	50 -100	luxuriant	Neomycin	36-37.5°C/ 16-24 hours
Staphylococcus aureus 9144 (00035*)	50 -100	luxuriant	Tylosin	36-37.5°C/ 16-24 hours

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics Medical Encyclopaedia, Inc, New York.
2. Schmidt and Moyer, 1944; J. Bact, 47:199.
3. United States Pharmacopoeia/ National Formulary (USP 42), 2019. US Pharmacopoeial Convention Inc, Rockville, MD.
4. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs, FDA, CFR, 1983. Title 21, part 436, Subpart D, Washington, D.C. U.S Government printing office, paragraphs 436, 100-436, 106 pg 242-259 (April 1).
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.40 M1143

Intended Use:

Antibiotic assay medium No. 40 is used in microbiological assay of Thiostrepton using *Enterococcus hirae* as test organism in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Ingredients	HiMedia	USP	HiVeg™
	M1143	MU1143	MV1143
Polypeptone	_	5.00	_
Peptone	2.50	_	_
Tryptone	2.50	_	_
HiVeg™ peptone	_	_	2.50
HiVeg™ hydrolysate	_	_	2.50
Yeast extract	20.00	20.00	20.00
Dextrose (Glucose)	10.00	10.00	10.00
Polysorbate 80 (Tween 80)	0.10	0.10	0.10
Potassium dihydrogen phosphate	2.00	2.00	2.00
Agar	10.00	10.00	10.00
Grams/litre	47.10	47.10	47.10
Final pH (at 25°C)	6.7 ± 0.2	_	6.7 ± 0.2
pH after sterilization (at 25°C)	_	6.7 ± 0.2	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

Principle And Interpretation

This medium is formulated in accordance to USP (1). This medium is used as maintenance medium for test organism *Enterococcus hirae* ATCC 10541 for assay of Thiostrepton.

Polypeptone peptone, yeast extract, tryptone, HiVeg™ peptone and HiVeg™ hydrolysate provides carbonaceous, nitrogeneous compounds, amino acids, minerals and other essential growth factors. Dextrose (Glucose) functions as carbon and energy source for enhancing the growth of test organism.

During maintenance of the test organims, good buffering action is provided by phosphates in the medium. Incorporation of polysorbates reduces the surface tension, maintaining uniform suspension of cells and can also neutralize phenolic compounds in the test sample, if any.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium No.40 M1143

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Gelling

Firm, comparable with 1.0% Agar gel

Colour and Clarity of prepared medium

Light amber coloured slightly opalescent gel forms in Petri plates.

pH

M1143 / MV1143 - 6.7 ± 0.2

MU1143 - 6.7±0.1

Cultural Response

Cultural characteristics observed after an incubation at 36-37.5°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Enterococcus hirae 10541 (00011*)	50 -100	luxuriant	≥70%	Thiostrepton

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2, 3).

Reference

1. United States Pharmacopoeia 2019 (USP42/NF37), US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium No.41 M1144

Intended Use:

Antibiotic Assay Medium No. 41 is used for the microbiological assay of Thiostrepton using *Enterococcus hirae* as the test organism in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	USP	HiVeg™
	M1144	MU1144	MV1144
Tryptone #	9.00	9.00	_
HiVeg™ hydrolysate	_	_	9.00
Yeast extract	5.00	5.00	5.00
Dextrose (Glucose)	20.00	20.00	20.00
Potassium hydrogen phosphate	1.00	1.00	1.00
Dipotassium hydrogen phosphate	1.00	1.00	1.00
Sodium citrate	10.00	10.00	10.00
Grams/litre	46.00	46.00	46.00
Final pH (at 25°C)	6.8 ± 0.2	_	6.8 ± 0.2
pH after sterilization (at 25°C)	_	6.8 ± 0.1	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#Pancreatic digest of casein

Principle And Interpretation

This medium is formulated in accordance with USP (1). This medium is used for turbidimetric microbiological assay of Thiostrepton, a polypeptide antibiotic.

Tryptone, HiVeg™ hydrolysate and yeast extract provides carbonaceous and nitrogeneous compounds, long chain amino acids, minerals and other essential growth factors. Dextrose (Glucose) provides carbon and energy source for enhancing the growth of test organism. Good buffering action is maintained by phosphates in the medium. Sodium citrate provides additional source of carbon and energy and promote enhanced growth of the test organism.

Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganism in a liquid medium containing a uniform concentration of an antibiotic. After incubation of the test organism in the working dilutions of the antibiotics, the amount of growth is determined by measuring the light transmittance using spectrophotometer. The concentration of antibiotic is determined by comparing amounts of growth obtained with that given by the reference standard solutions. Use of this method is appropriate only when test samples are clear.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium No.41 M1144

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate

pH

M1144 / MV114 - 6.8 ± 0.2

MU1144 - 6.8 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 36-37.5°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Serial dilution with
Enterococcus hirae 10541 (00011*)	50 -100	luxuriant	Thiostrepton

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2, 3).

Reference

1. United States Pharmacopoeia / National Formulary 2019, US Pharmacopoeial Convention, Inc., Rockville, MD.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium B M1346

Intended Use:

Antibiotic Assay Medium B is used for the microbiological assay of Colistimethate sodium using *Bordetella bronchiseptica* ATCC 4617 and *Escherichia coli* ATCC 10536 in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water with 10 ml polysorbate 80. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Adjust the pH of the medium, using freshly prepared buffer solution as recommended by the European pharmacopoeia and British Pharmacopoeia for the antibiotic assayed.

Ingredients	HiMedia	EP	BP
	M1346	ME1346	M1346B
Tryptone#	17.00	17.00	17.00
Soya peptone##	3.00	3.00	3.00
Sodium chloride	5.00	5.00	5.00
Dextrose (Glucose)	2.50	_	_
Glucose monohydrate	_	2.50	2.50
Dipotassium hydrogen phosphate	2.50	2.50	2.50
Agar	15.00	15.00	15.00
Grams/litre	45.00	44.77	44.77
Final pH (at 25°C)	7.3 ± 0.2	_	_
pH after sterilization (at 25°C)	_	7.3 ± 0.1	7.3 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

#Equivalent to Pancreatic digest of casein
Equivalent to Papaic digest of soyabean

Principle And Interpretation

Antibiotic Assay Medium B is prepared according to European Pharmacopoeia (1) and British Pharmacopoeia (2). It is recommended for the assay of Colistimethate sodium and colistin sulphate using *Bordetella bronchiseptica* and *Escherichia coli* as the test organism.

Combination of Tryptone and soya peptone provides nitrogeneous and carbonaceous compounds, long chain amino acids and other essential nutrients for the growth of test organisms. Dextrose and Glucose monohydrate provides fermentable source of carbon and enhances the growth of test organims. Phosphates in the medium enhance buffering action and sodium chloride maintains osmotic equilibrium.

Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar cooled to 40-45°C and spread evenly over the surface of solidified base agar. All conditions in the microbiological assay must be controlled carefully. The use of standard culture media in the test is one of the important steps for good results.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Antibiotic Assay Medium B M1346

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri plates

pH

M1346 - 7.3 ± 0.2

ME1346 / M1346B - 7.3 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 35-39°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Bordetella bronchiseptica 4617	50 -100	luxuriant	≥70%	Colistimethate sodium, Colistin sulphate
Escherichia coli 10536	50 -100	luxuriant	≥70%	Colistimethate sodium, Colistin sulphate

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines.
2. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium C M555

Intended Use:

Antibiotic Assay Medium C is used as the broth medium in turbidimetric assay of a wide variety of antibiotics in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Adjust the pH of the medium, using freshly prepared buffer solution as recommended by the European / British pharmacopoeia for the antibiotic assayed.

Advice : Recommended for the microbiological assay of Colistimethate sodium, Dihydrostreptomycin sulphate, Erythromycin estolate, Erythromycin ethylsuccinate, Framycetin sulphate, Gentamicin sulphate, Gramicidin, Kanamycin acid sulphate, Kanamycin monosulphate, Neomycin sulphate, Rifamycin sodium, Spiramycin, Streptomycin sulphate, Tylosin, Tylosin tartarate, Tyrothricin and Vancomycin hydrochloride.

Ingredients	HiMedia	EP	BP
	M555	ME555	M555B
Peptone	6.00	6.00	6.00
HM peptone B#	1.50	1.50	1.50
Yeast extract	3.00	3.00	3.00
Glucose monohydrate	_	1.00	1.00
Dextrose (Glucose)	1.00	_	_
Sodium chloride	3.50	3.50	3.50
Potassium dihydrogen phosphate	1.32	1.32	1.32
Dipotassium hydrogen phosphate	3.68	3.68	3.68
Grams/litre	20.00	19.90	19.90
Final pH (at 25°C)	7.0 ± 0.2	_	_
pH after sterilization (at 25°C)	_	*7.0 ± 0.1	*7.0 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

#Beef extract

*While assaying Josamycin & Josamycin sulphate adjust the pH to 8.0 ±0.1

Principle And Interpretation

This medium is used in turbidimetic assay of several antibiotics. The composition of the medium is in accordance to the specifications detailed in the European Pharmacopoeia (1) and British Pharmacopoeia (2). Turbidimetric methods for determining the potency of antibiotics are inherently more accurate and more precise than comparable agar diffusion procedures (3).

Peptone, HM peptone B and yeast extract provides nitrogenous and carbonaceous compounds, long chain amino acids and other essential nutrients and growth factors for enhanced microbial growth. Sodium chloride maintains the osmotic equilibrium while phosphates are incorporated in the medium to provide good buffering action. Glucose monohydrate serves as the carbon and energy source for faster growth.

Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganims in a liquid medium containing a uniform concentration of an antibiotic (4). Use of this method is appropriate only when test samples are clear.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Antibiotic Assay Medium C M555

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate

pH

M555 - 7.0 ± 0.2

ME555 / M555B - 7.0 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Serial dilution with
Escherichia coli 9637	50 -100	luxuriant	Colistimethate sodium, Colistin sulphate
Escherichia coli 10536	50 -100	luxuriant	Rifamycin sodium
Enterococcus hirae 10541 (00011*)	50 -100	luxuriant	Gramicidin, Tyrothricin
Klebsiella pneumoniae 10031	50 -100	luxuriant	Dihydrostreptomycin sulphate, Streptomycin sulphate
Staphylococcus aureus subsp. *aureus* 6538P (00033*)	50 -100	luxuriant	Erythromycin estolate, Erythromycin ethylsuccinate, Erythromycin stearate, Framycetin sulphate, Gentamicin sulphate, Gramicidin, Kanamycin monosulphate, Kanamycin acid sulphate, Neomycin sulphate, Spiramycin, Tobramycin; For Josamycin & Josamycin propionate adjust the pH of the medium to 8.0 ± 0.1, For Vancomycin hydrochloride incubate at 37-39°C.
Staphylococcus aureus 9144 (00035*)	50 -100	luxuriant	Tylosin, Tylosin tartarat

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5, 6).

Reference

1. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines.
2. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
3. Rippere RA. Some principles of microbiological turbidimetric assays of antibiotics. J Assoc Off Anal Chem. 1979 62 (4):951-6.
4. Chapin-Robertson and Edberg, 1991, Measurement of Antibiotics in Human Body fluids: Techniques and significance. Antibiotics in Laboratory medicine, New York pp 305.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium D M556

Intended Use:

Antibiotic Assay Medium D is used for the microbiological assay of Erythromycin estolate using *Klebsiella pneumoniae* ATCC 10031 as a test organism in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Adjust the pH of the medium using freshly prepared buffer solution as recommended by the European pharmacopoeia and British Pharmacopoeia for the antibiotic assayed.

Ingredients	HiMedia	EP	BP
	M556	ME556	M556B
HMH extract#	1.50	1.50	1.50
Yeast extract	1.50	1.50	1.50
Casitose##	5.00	5.00	5.00
Glucose monohydrate	1.00	1.00	1.00
Sodium chloride	3.50	3.50	3.50
Dipotassium hydrogen phosphate	3.68	3.68	3.68
Potassium dihydrogen phosphate	1.32	1.32	1.32
Potassium nitrate	2.00	2.00	2.00
Grams/litre	19.40	19.40	19.40
Final pH (at 25°C)	7.0 ± 0.2	_	_
pH after sterilization (at 25°C)	_	7.0 ± 0.1	7.0 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

Heart extract
Peptone casein

Principle And Interpretation

This medium is widely used for turbidimetric assay of erthromycin estolate using *Klebsiella pneumoniae* as test organism in accordance with European Pharmacopoeia (1) and British Pharmacopoeia (2). Turbidimetric methods for determining the potency of antibiotics are inherently more accurate and more precise than comparable agar diffusion procedures.

Combination of Casitose, HMH extract and yeast extract supply nitrogen and carbon compounds, amino acids, nutrients and essential mineral and growth factors for enhanced microbial growth. Potassium nitrate serves as inorganic source of nitrogen for the growth of test organism. Sodium chloride maintains the osmotic equilibrium while phosphates are incorporated in the medium to provide good buffering action. Glucose monohydrate serves as the carbon and energy source for faster growth.

Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganism in a liquid medium containing a uniform concentration of an antibiotic. Use of this method is appropriate only when test samples are clear.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Antibiotic Assay Medium D M556

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent solution

pH

M556 - 7.0 ± 0.2

ME556 / M556B - 7.0 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Serial dilution with
Klebsiella pneumoniae 10031	50 -100	luxuriant	Erythromycin estolate

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. European Pharmacopoeia 2017, European Department, for the Quality of Medicines.
2. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium E M1347

Intended Use:

For microbiological assay of Framycetin Sulphate and Neomycin sulphate using *Bacillus subtilis* and/or *Bacillus pumilus* in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired. Adjust the pH of the medium, using freshly prepared buffer solution as recommended by the European / British pharmacopoeia for the antibiotic assayed.

Advice : Recommended for the microbiological assay of Framycetin sulphate and Neomycin sulphate.

Ingredients	HiMedia	EP	BP
	M1347	ME1347	M1347B
Peptone	5.00	5.00	5.00
Disodium hydrogen phosphate,12H_2O	26.90	26.90	26.90
HM extract#	3.00	3.00	3.00
Agar	10.00	10.00	10.00
Grams/litre	28.67	28.67	28.67
Final pH (at 25°C)	7.9 ± 0.2	_	_
pH after sterilization (at 25°C)	_	7.9 ± 0.1	7.9 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

#Equivalent to Meat extract

Principle And Interpretation

This medium is formulated in accordance with European Pharmacopoeia (1) and British Pharmacopoeia (2). This medium is widely used for as seed agar in plate assay of Framycetin sulphate and Neomycin sulphate using *Bacillus subtilis* and/or *Bacillus pumilus* as test organism.

Peptone and HM extract supply nitrogenous and carbonaceous compounds, long chain amino acids and other nutrients essential for microbial growth. Phosphates are incorporated in the medium to provide good buffering action. The low concentration of agar facilitates proper diffusion of antibiotic in the seed agar.

Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar cooled to 40-45°C and spread evenly over the surface of solidified base agar. Zones of inhibition around the antibiotic are then measured. All conditions in the microbiological assay must be controlled carefully. The use of standard culture media in the test is one of the important steps for good results.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium E M1347

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.0% Agar gel.

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

ME1347 / M1347B - 7.90 ± 0.1

M1347 - 7.9 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Bacillus pumilus NCTC 8241	50 -100	luxuriant	≥70%	Framycetin sulphate, Neomycin sulphate
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	≥70%	Framycetin sulphate and Neomycin sulphate

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines.
2. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium F M923

Intended Use:

Antibiotic Medium F is used for microbiological assay of Amphotericin B and Nystatin using *Saccharomyces cerevisiae* ATCC 9763 and *Candida tropicalis* CIP 1433-83 in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired. Adjust the pH of the medium, using freshly prepared buffer solution as recommended by the European / British pharmacopoeia for the antibiotic assayed.

Advice : Recommended for the microbiological assay of Amphotericin B and Nystatin.

Ingredients	HiMedia	EP	BP
	M923	ME923	M923B
Peptone	9.40	9.40	9.40
Yeast extract	4.70	4.70	4.70
HM peptone B#	2.40	2.40	2.40
Sodium chloride	10.00	10.00	10.00
Glucose monohydrate	_	10.00	10.00
Dextrose (Glucose)	10.00	_	_
Agar	23.50	23.50	23.50
Grams/litre	60.00	59.09	59.09
Final pH (at 25°C)	6.0 ± 0.2	_	_
pH after sterilization (at 25°C)	_	*6.0±0.1	*6.0±0.1
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

*While assaying Amphotericin B adjust the pH to 6.1±0.1
#Equivalent to Beef extract

Principle And Interpretation

Grove and Randall have elucidated the antibiotic assays and medias in their comprehensive treatise on antibiotic assays (1). Antibiotic assay Medium F is recommended for the microbiological assay of Nystatin and Amphotericin B using *Saccharomyces cerevisiae* and *Candida tropicalis*. This medium is formulated in accordance with the European Pharmacopoeia (2) and British Pharmacopoeia (3). Freshly prepared plates should be used for antibiotic assays. Test organisms are inoculated in sterile seed agar cooled to 40-45°C and spread evenly over the surface of solidified base agar. After incubation the concentration of the antibiotic being assayed is determined by measuring the zone of inhibition obtained, with that of reference standard antibiotic. All conditions in the microbiological assay must be carefully controlled. The use of standard culture media in the test is one of the important steps for good results.

Peptone, yeast extract and HM peptone B supply nitrogen, carbon compounds, amino acids and other essential nutrients, minerals and growth factors for the growth of the test organisms. Glucose monohydrate in the medium provides enhanced source of carbon and energy. Osmotic equilibrium in the medium is provided by sodium chloride thereby maintaining the cell viability and integrity. Higher agar concentration provides solid substratum for growth of colonies and controls the diffusion of antibiotics.

Type of specimen

Pharmaceutical samples.

Antibiotic Assay Medium F M923

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2, 3).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm,comparable with 2.35% Agar gel.

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

M923 - 6.0 ± 0.2

ME923 / M923B - 6.0 ± 0.1

Cultural Response

Cultural characteristics observed after an incubation at specified temperature for 18 - 24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed	Incubation Temperature
Saccharomyces cerevisiae 9763 (00058*)	50 -100	luxuriant	≥70%	Amphotericin B, Nystatin	35-37°C 30-32°C
Candida ropocalis CIP 1433-83	50 -100	luxuriant	≥70%	Nystatin	30-37°C

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. Grove and Randall, 1955, Assay Methods of Antibiotics, Medical Encyclopedia, Inc. New York
2. European Pharmacopoeia 2017, European Department, for the Quality of Medicines.
3. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium G M553

Intended Use:

Antibiotic Assay Medium G is used for the microbiological assay of Bleomycin sulphate using *Mycobacterium smegmatis*, as a test organism in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water containing 10 grams glycerol. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Advice : Recommended for the microbiological assay of Bleomycin sulphate.

Ingredients	HiMedia	EP	BP
	M553	ME553	M553B
Peptone	10.00	10.00	10.00
HM extract#	10.00	10.00	10.00
Sodium chloride	3.00	3.00	3.00
Agar	15.00	15.00	15.00
Grams/litre	38.00	38.00	38.00
Final pH (at 25°C)	7.0 ± 0.2	_	_
pH after sterilization (at 25°C)	_	7.0 ± 0.1	7.0 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

#Equivalent to Meat extract

Principle And Interpretation

This medium is formulated in accordance to European Pharmacopoeia (1) and British Pharmacopoeia (2). This medium is employed widely as base agar for agar diffusion assay of Bleomycin using *Mycobacterium smegmatis*. It is also used for preparing the inoculum of *Mycobacterium smegmatis* for assay.

Peptone and HM extract in this medium provides nitrogen, carbon compounds, amino acids and other essential growth nutrients for the test organism. Agar provides excellent solid substratum for support and over layering of seed agar, for the assay of Bleomycin. Addition of glycerol is important for provision of carbon to the test organism.

To perform the antibiotic assay the Base Agar should be prepared on the same day as the test. For the cylinder method, a base layer of 21 ml is required. Once the base medium has solidified, seed layer inoculated with the standardized culture can be overlaid. Even distribution of the layer is important.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium G M553

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm,comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH

M553 : 7.0 ± 0.2

ME553/ M553B : 7.0 ± 0.1

Cultural Response

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

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotic assayed
Mycobacterium smegmatis 607	50 -100	good-luxuriant	≥70%	Bleomycin sulphate

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. European Pharmacopoeia, 2017, European Department, for the Quality of Medicines
2. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen,J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium H M1665

Intended Use:

Antibiotic Assay Medium H is used for the microbiological assay of Teicoplanin using *Bacillus subtilis* as a test organism in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Cool to 45-50°C. Mix well and pour into sterile Petri plates or as desired.

Ingredients	EP	BP
	ME1665	M1863B
Peptone	5.00	5.00
HM peptone B#	3.00	3.00
Agar	15.00	15.00
Grams/litre	23.00	23.00
Final pH (at 25°C)	7.9 ± 0.1	_
pH after sterilization (at 25°C)	_	7.9 ± 0.1
Water	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

#Equivalent to Beef extract

Principle And Interpretation

This medium is formulated in accordance with European Pharmacopoeia (1). It is also recommended by British Pharmacopoeia (2) This medium is employed in the microbiological assay of Teicoplanin using *Bacillus subtilis* .

Essential nutrients for growth of test organism are provided by Peptone, Tryptone and HM peptone B powder in this medium. Agar provides excellent medium for antibiotic diffusion and gives well defined zones of inhibition.

Freshly prepared plates should be preferably used for assaying antibiotics. Test organisms is inoculated in sterile seed agar pre-cooled to 40-45°C and spread evenly over the surface of solidified base agar. All conditions in the microbiological assay must be controlled carefully. One of the critical and important step for obtaining good results is use of appropriate standard culture media.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Antibiotic Assay Medium H M1665

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Light yellow coloured clear to slightly opalescent gel forms in Petri plates

pH
ME1665 / M1863B - 7.9 ± 0.1

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

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant	≥70%	Teicoplanin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. European Pharmacopoeia, 2017, European Department for the Quality of Medicines
2. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Antibiotic Assay Medium I M1847B

Intended Use:

Antibiotic Assay Medium I is used for the microbiological turbidimetric assay of Apramycin using *Salmonella* Cholerasuis as a test organism in accordance with British Pharmacopoeia.

Directions:

Suspend 18.0 grams in 1000 ml R-water/distilled/purified water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle. Dispense as desired.

Ingredients	HiMedia	BP
	M1665	M1847B
Tryptone	6.00	6.00
Yeast extract	2.00	2.00
D-Glucose (Dextrose)	10.00	10.00
Grams/litre	18.00	18.00
Final pH (at 25°C)	8.0 ± 0.2	–
pH after sterilization (at 25°C)	–	8.0
Water	Purified/ Distilled	R-water / Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle

Principle And Interpretation

This medium is formulated in accordance to British Pharmacopoeia (1). This medium is employed for turbidimetric assay of Apramycin, an antibiotic of the aminocyclitol group, using *Salmonella* Cholerasuis. Turbidimetric methods for determining the potency of antibiotics are inherently more accurate and more precise than comparable agar diffusion procedures. Essential nutrients for growth of test organism is provided by Tryptone and yeast extract in this medium. D-Glucose serves as source of carbon to the test organism. Turbidimetric antibiotic assay is based on the change or inhibition of growth of a test microorganism in a liquid medium containing a uniform concentration of an antibiotic. Use of this method is appropriate only when test samples are clear.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Freshly prepared plates must be used or it may give erroneous results.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution in tubes

pH

M1665 / M1847B - 8.0 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 12 - 24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth	Recovery	Antibiotics assayed
Salmonella Choleraesuis ATCC 12011	50 -100	luxuriant	≥70%	Apramycin

Key : * : Corresponds to WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and use freshly prepared medium. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

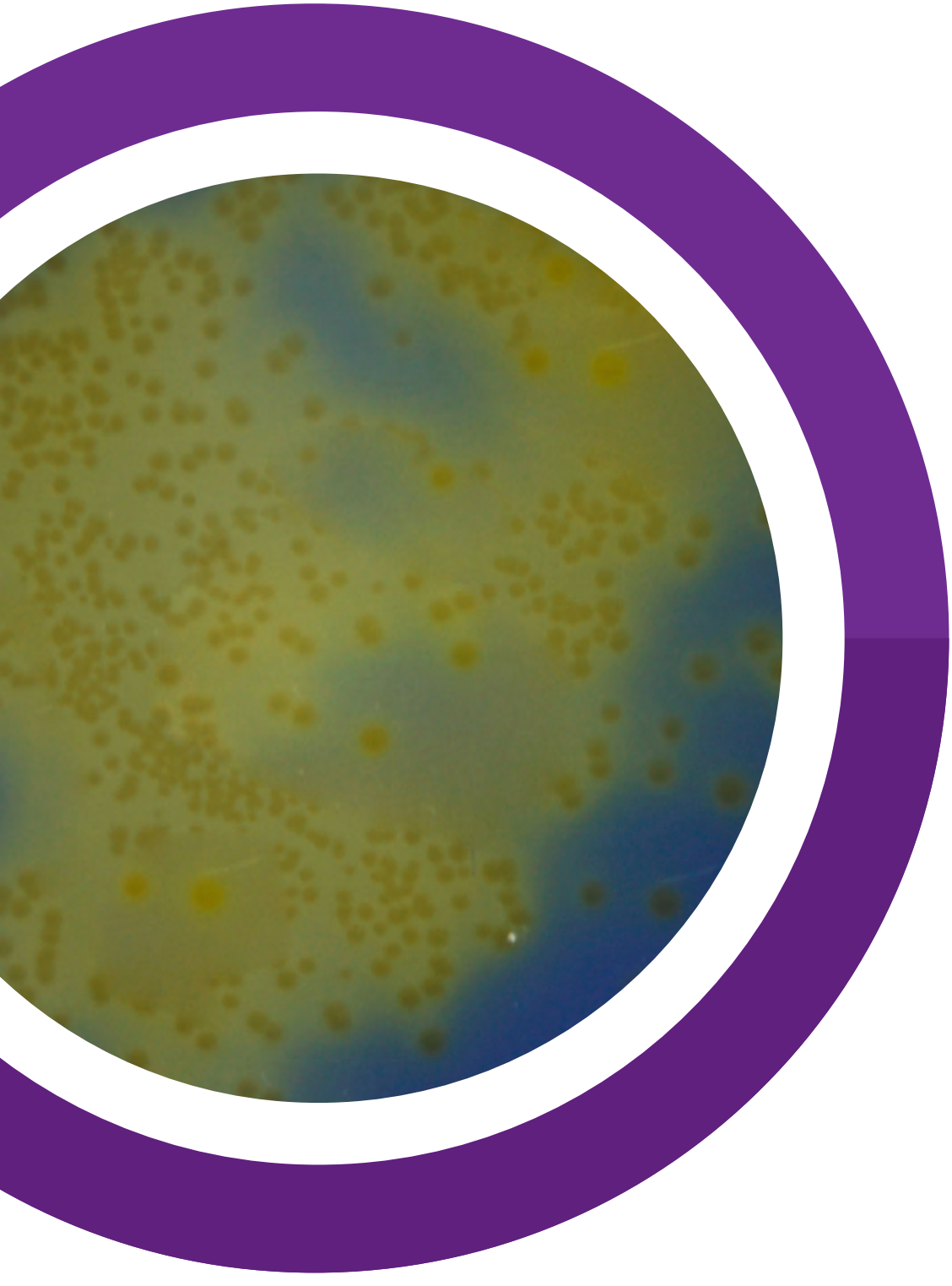
Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2, 3).

Reference

1. British Pharmacopoeia, 2016, British Pharmacopoeia Commission
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Neutralizing Media



Dey-Engley (D/E) Neutralizing Broth M1062

Intended Use:

Recommended in disinfectant testing and neutralization of antiseptics and disinfectants for determining its bactericidal activity. The composition is in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Ingredients	HiMedia	USP	HiVeg™	HiCynth
	M1062	MU1062	MV1062	MCD1062
Tryptone#	5.00	5.00	_	_
HiVeg™ hydrolysate	_	_	5.00	_
Yeast extract	2.50	2.50	2.50	_
HiCynth™ Peptone No.1##	_	_	_	5.00
HiCynth™ Peptone No.5##	_	_	_	2.50
Dextrose (Glucose)	10.00	10.00	10.00	10.00
Sodium thioglycollate	1.00	1.00	1.00	1.00
Sodium thiosulphate	6.00	6.00	6.00	6.00
Sodium bisulphite	2.50	2.50	2.50	2.50
Lecithin	7.00	7.00	7.00	7.00
Polysorbate 80 (Tween 80)	5.00	5.00	5.00	5.00
Bromocresol purple	0.02	_	0.02	0.02
Grams/litre	39.02	39.00	39.02	39.02
Final pH (at 25°C)	7.6 ± 0.2	_	7.6 ± 0.2	7.6 ± 0.2
pH after sterilization (at 25°C)	_	_	_	_
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min

#Pancreatic digest of casein
##Chemically defined peptones

Principle And Interpretation

Dey-Engley (D/E) Neutralizing Broth (without Bromo cresol purple) is formulated as per United States Pharmacopoeia (1). It neutralizes a broad spectrum of antiseptics and disinfectants including quaternary ammonium compounds, phenolics, iodine and chlorine preparations, mercurials, formaldehyde and glutaraldehyde. Sodium thioglycollate, sodium thiosulphate, sodium bisulphite, soya lecithin and Polysorbate 80 (Tween 80) act as neutralizing components.

For testing disinfectants, prepare two sets of test tubes, one containing 9 ml Dey-Engley Neutralizing Broth (MU1062) and other with 9 ml Dey-Engley Neutralizing Broth Base. Add 1 ml of disinfectant under test. Mix well and allow it to stand for 15 minutes. Inoculate 0.1 ml of 1:100,000 dilution of overnight broth cultures and incubate at 30-35°C for 48 hours Growth in Neutralizing Broth and no growth in Neutralizing Broth Base indicates neutralization of disinfectant. To check bactericidal activity, both broth tubes are inoculated on D/E Neutralizing Agar (M186). Positive growth from negative tubes of Neutralizing Broth Base indicates bacteriostatic substance while negative growth indicates a bactericidal disinfectant. All positive tubes should show growth on Dey-Engley Neutralizing Agar. The control disinfectants used in test procedure are 2% chlorine, 2% formaldehyde, 1% glutaraldehyde, 2% iodine, 2% phenol, 1/750 quaternary ammonium compounds, 1/1000 mercurials etc. Dey Engley Neutralizing Broth (M1062 / MV1062 / MCD1062) contains Bromocresol purple indicator which turns yellow in presence of dextrose fermenting organisms.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Dey-Engley (D/E) Neutralizing Broth M1062

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Turbidity cannot be used as a measure to detect growth. Hence suspected tubes must be subcultured
2. Further biochemical tests must be carried out for confirmation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

MU1062 : Light yellow coloured opalescent solution

M1062 / MV1062 / MCD1062 : Purple coloured opalescent solution

pH

M1062 / MV1062 / MCD1062 - 7.6 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at i)For bacteria at 30-35°C for ≤3 days ii) For fungi at 20-25°C for ≤5days.

Organism (ATCC)	Inoculum (CFU)	Growth
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	luxuriant
Pseudomonas aeruginosa 27853 (00025*)	50 -100	luxuriant
Salmonella Typhimurium 14028 (00031*)	50 -100	luxuriant
Escherichia coli 8739 (00012*)	50 -100	luxuriant
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	luxuriant
#Aspergillus brasiliensis 16404 (00053*)	50 -100	luxuriant
Candida albicans 10231 (00054*)	50 -100	luxuriant

Key : # : Formerly known as *Aspergillus niger*

* : corresponding to WDCM numbers

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. Engley and Dey, 1970. Chem. Spec. Manuf. Assoc. Proc., Mid-Year Meet., p. 100.
2. The United States Pharmacopoeia 2019, The US Pharmacopoeial Convention Inc., Rockville, MD.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Medium in tubes			
LQ162C LQ162CC	Dey Engley Neutralizing Broth	for neutralizing and determining bactericidal activity of quaternary ammonium compounds.	10X100ML 5X200ML
LQ162X	Dey Engley Neutralising Broth	for neutralising and testing antiseptics and disinfectants.	25X10ML/ 50X10ML
Category : Ready Prepared Transport medium with swabs			
MS1062 MS1062S	HiCulture™ Transport Swabs w/ Dey-Engley Neutralizing Broth HiCulture™ Transport Swabs w/ Dey-Engley Neutralizing Broth with metal stick	for transporting microbial specimens in presence of antiseptics and disinfectants.	10 NO / 50 NO 10 NO / 50 NO
MQ1062	HiCulture™ Transport Swab w/ Dey Engley Neutralizing Broth	Recommended for transporting microbial specimens in presence of antiseptics and disinfectants. Note : on γ-irradiation the the colour of the tubes / media may vary with no effect on the performance of the medium	20 NO / 50 NO

T.A.T. Broth Base M562

Intended Use:

T.A.T. (Tryptone Azolectin Tween) Broth with addition of Tween 20 is used for sterility testing of highly viscous or gelatinous substances like salves, ointments and other cosmetic products in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 960ml of purified / distilled water and add 40 ml of Polysorbate 20 (Tween 20). Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Ingredients	HiMedia	Granulated	USP	HiVeg™
	M562	GM562	MU562	MV562
Tryptone	20.00	20.00	20.00	_
Lecithin	_	_	5.00	_
HiVeg™ hydrolysate	_	_	_	20.00
Azolectin	5.00	5.00	_	5.00
Grams/litre	25.00	25.00	25.00	25.00
Final pH (at 25°C)	7.2 ± 0.2	7.2 ± 0.2	_	7.2 ± 0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

Principle And Interpretation

T.A.T. Broth is prepared according to the formula recommended by United States Food and Drug Administration and United States Pharmacopoeia (1, 2) for enrichment and further isolation and cultivation of gram-negative bacteria in cosmetics, tropical drugs and sterility testing of viscous or gelatinous substances.

Prepare decimal dilutions of the sample to be tested from 10^{-1} to 10^{-6}. Inoculate 1 gram (1 ml) sample and 1 ml of each dilution into 40 ml of T.A.T. Broth (3). After incubation subculture the growth on MacConkey Agar (MH081) and TSI Agar (MU021).

Type of specimen

Cosmetics, Pharmaceutical samples.

Specimen Collection and Handling

For cosmetic samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. The tubes must be further subcultured for identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

T.A.T. Broth Base M562

Quality Control

Appearance
Off-white to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Light yellow coloured clear to slightly opalescent solution

GM562 : Light yellow coloured granular media

Cultural Response
Cultural characteristics observed with added Polysorbate 20 after an incubation at 35-37°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	good-luxuriant
Candida albicans 10231 (00054*)	50 -100	fair-good
Pseudomonas aeruginosa 9027 (00026*)	50 -100	good-luxuriant
Salmonella Typhi 6539	50 -100	good-luxuriant
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	good-luxuriant
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	good-luxuriant

Key : * : corresponding to WDCM numbers

Storage and Shelf Life
Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal
User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (4, 5).

Reference

1. Food and Drug Administration, 1969, Procedure for Examination of Tropical Drugs and Cosmetics.
2. The United States Pharmacopoeia, 2019. The United States Pharmacopeial Convention. Rockville, MD.
3. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
5. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready Prepared Liquid Medium in tubes			
LQ525IX	TAT Broth	for sterility testing of highly viscous or gelatinous substances such as salves, ointments and other cosmetic products, in accordance with USP.	25X9ML / 50X9ML
LQ525IXB	TAT Broth w/ beads		25X9ML / 50X9ML
LQ525XC	TAT Broth w/o beads		5X90ML
LQ525XCB	TAT Broth w/ beads		5X90ML
LQ525XCC	TAT Broth w/o beads		5X190ML
LQ525XCCB	TAT Broth w/ beads		5X190ML
LQ525XD	TAT Broth w/o beads		5X490ML
LQ525XDB	TAT Broth w/ beads		5X490ML
LQ525XMB	TAT Broth w/ beads		2X990ML
LQ525XM	TAT Broth w/o beads		2X990ML

Letheen Broth M165

Intended Use:

Recommended for determination of bactericidal activity of quaternary ammonium compounds using *Escherichia coli* or *Staphylococcus aureus* subsp. *aureus* in accordance with United States Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Ingredients	HiMedia	USP	HiVeg™
	M165	MU165	MV165
Peptone #	_	10.00	_
Peptone	10.00	_	_
HiVeg™ peptone	_	_	10.00
HM peptone B##	5.00	5.00	_
HiVeg™ extract	_	_	5.00
Polysorbate 80 (Tween 80)	5.00	5.00	5.00
Sodium chloride	5.00	5.00	5.00
Lecithin	0.70	0.70	0.70
Grams/litre	25.70	25.70	25.70
Final pH (at 25°C)	7.0 ± 0.2	_	7.0 ± 0.2
Water	Purified/ Distilled	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min

#equivalent to Peptamin
##equivalent to Beef extract

Principle And Interpretation

Letheen Broth was developed by Quisno, Gibby & Foter (3) by the addition of lecithin & polysorbate 80 to FDA Broth. Letheen Broth is recommended by AOAC to determine the Phenol coefficient of cationic surfactants (2). Letheen media are recommended by USP in disinfectant challenge testing (4).

Peptone, HM peptone B, HiVeg™ peptone and HiVeg™ extract supply nitrogenous compounds, carbon, sulphur and other trace elements to the organisms. Lecithin and Polysorbate 80 (Tween 80) enables the recovery of bacteria from solutions containing residues of disinfectant used in sanitization of utensils and equipments. Lecithin neutralizes quaternary ammonium compounds and Polysorbate 80 (Tween 80) neutralizes phenolic disinfectants, hexachlorophene and formalin (5, 6).

Dehydrated medium may appear moist with brown sugar appearance, does not indicate deterioration.

Type of specimen

Water samples, Pharmaceutical samples.

Specimen Collection and Handling

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1).

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical guidelines (4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further biochemical tests must be carried out for confirmation.

Letheen Broth M165

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear to slightly opalescent solution

pH

M165/MV165 - 7.0 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	good-luxuriant
Escherichia coli 8739 (00012*)	50 -100	good-luxuriant

Key : * corresponding WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (7, 8).

Reference

1. APHA, 1960, Standard Methods for the Examination of Water and Wastewater, 11th ed., APHA, N.Y.
2. Horwitz, (Ed.), 2000, Official Methods of Analysis of AOAC International, 17th Ed., vol. I, AOAC International, Gaithersburg, Mb.
3. Weber and Black, 1948, Am. J. Public Health, 38:1405.
4. The United States Pharmacopoeia/National Formulary 2019, US Pharmacopeial Convention Inc. Rockville, M.D.
5. Favero (Chm.), 1967, A State of the Art Report, Biological Contamination Control Committee, American Association for Contamination Control.
6. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Ready Prepared Media

Code	Product Name	Usage	Packing
Category : Ready prepared Liquid Medium in glass bottles			
LQ108	Letheen Broth	for determination of bacterial activity of quarternary ammonium compounds using *Escherichia coli* or *Staphylococcus aureus*	5X100ML
Category : Ready prepared Transport Medium with Swabs			
MS5397	HiCulture™ Sterile Swabs w/ Letheen Broth	for determination of bacterial activity of quaternary ammonium compounds using *Escherichia coli* or *Staphylococcus aureus*	25no / 50no

Diluting Fluid A M1415

Intended Use:

Diluting Fluid A is recommended for sterility testing of pharmaceuticals in accordance with USP.

Directions:

Suspend 1.0 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Ingredients	HiMedia
Peptone	1.00
Grams/litre	1.00
Final pH (at 25°C)	7.1 ± 0.2

Principle And Interpretation

Diluting Fluid A is recommended as rinsing fluid for membrane filter method used in validation tests for bacteriostasis and fungistasis activity of pharmaceutical articles before carrying out sterility test procedures as per USP (1). After filtering the specified quantity of the test specimen, the membrane is rinsed with measured portions of rinsing or diluting fluid. This rinse is inoculated with known number of test bacteria and fungi as specified in pharmacopoeia. The resultant growth is compared with positive control to determine presence of fungistasis or bacteriostasis activity in test specimen.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution

pH

7.10 ± 0.2

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth
Candida albicans 10231 (00054*)	50 -100	good
Escherichia coli 25922 (00013*)	50 -100	good
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	good
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	good

Key : * corresponding WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Diluting Fluid A M1415

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (2, 3).

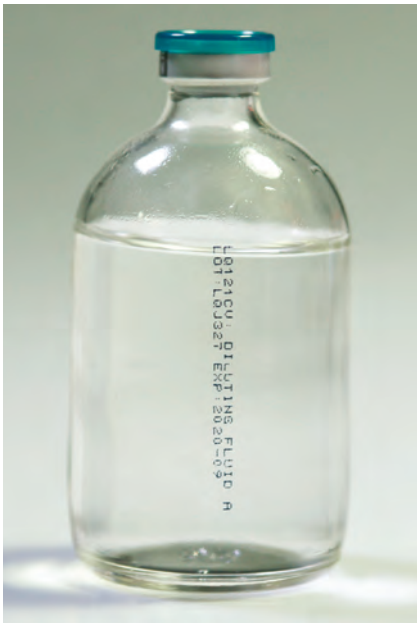
Reference

1. The United States Pharmacopoeia / National Formulary, 2019, Asian Edition, US Pharmacopeial convention Inc., Rockville, MD.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

LQ121CA

100 ml

LQ121CV



100 ml

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Medium in glass bottles			
LQ121C LQ121CC LQ121L LQ121D LQ121CV LQ121CV LQ121DW LQ121XC	Diluting Fluid A Diluting Fluid A-Double Packed	diluent in testing of pharmaceuticals in accordance with USP	5X100ML 5X200ML 5X300ML 5X500ML 10X100ML 5X100ML 10X100ML 5X90ML

Diluting Fluid K M1416

Intended Use:

Diluting Fluid K is recommended for sterility testing of pharmaceuticals in accordance with USP.

Ingredients	HiMedia
Peptone	5.00
HM peptone B#	3.00
Polysorbate 80 (Tween 80)	10.00
Grams/litre	18.00
Final pH (at 25°C)	6.9 ± 0.2

#Equivalent to Beef extract

Directions:

Suspend 18.0 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Diluting Fluid K is recommended as rinsing fluid for membrane filter method used in validation tests for bacteriostasis and fungistasis activity of pharmaceutical articles before carrying out sterility test procedures as per USP (1). After filtering the specified quantity of the test specimen, the membrane is rinsed with measured portions of rinsing or diluting fluid. This rinse is inoculated with known number of test bacteria and fungi as specified in pharmacopoeia. The resultant growth is compared with positive control to determine presence of fungistasis or bacteriostasis activity in test specimen.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution

pH

6.70-7.10

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth
Candida albicans 10231 (00054*)	50 -100	good
Escherichia coli 25922 (00013*)	50 -100	good
Escherichia coli 8739 (00012*)	50 -100	good
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	good
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	good

Key : * corresponding WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Diluting Fluid K M1416

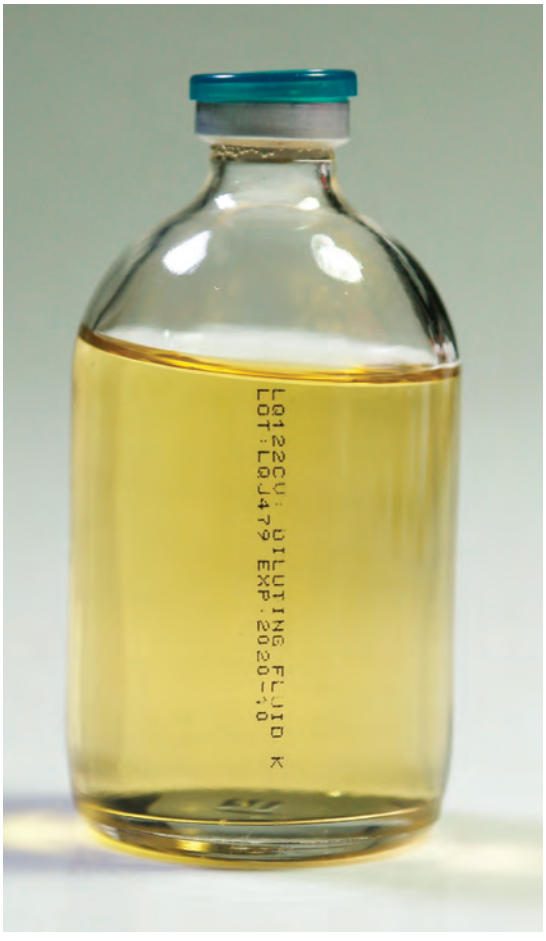
Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (2, 3).

Reference

1. The United States Pharmacopoeia 2019, US Pharmacopeial convention Inc., Rockville, MD.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

LQ122CV



100 ml

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Medium in glass bottles			
LQ122C LQ122L LQ122CV	Diluting Fluid K	diluent in testing of pharmaceuticals in accordance with USP	10X100ML 5X300ML 10X100ML

Diluting Fluid D M1686

Intended Use:

Diluting Fluid D is used for sterility testing of pharmaceuticals in accordance with USP.

Directions:

Suspend 2.0 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes i.e. validated cycle.

Ingredients	HiMedia
Peptone	1.00
Polysorbate 80	1.00
Grams/litre	2.00
Final pH (at 25°C)	7.1 ± 0.2

Principle And Interpretation

Diluting Fluid D is recommended as rinsing fluid for membrane filter method used in validation tests for bacteriostasis and fungistasis activity of pharmaceutical articles before carrying out sterility test procedures as per USP (1). After filtering the specified quantity of the test specimen, the membrane is rinsed with measured portions of rinsing or diluting fluid. This rinse is inoculated with known number of test bacteria and fungi as specified in pharmacopoeia. The resultant growth is compared with positive control to determine presence of fungistasis or bacteriostasis activity in test specimen. This medium is recommended for articles containing lecithin or oil or for devices labeled as sterile pathway (1).

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature

Quality Control

Appearance

Cream to yellow coloured homogeneous free flowing powder

Colour and Clarity of prepared medium

Light amber coloured clear solution without any precipitate

Reaction

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

pH

6.90-7.30

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth
Candida albicans 10231 (00054*)	50 -100	good
Escherichia coli 25922 (00013*)	50 -100	good
Staphylococcus aureus subsp. *aureus* 25923 (00034*)	50 -100	good
Escherichia coli 8739 (00012*)	50 -100	good
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	good

Key : * corresponding WDCM number

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Diluting Fluid D M1686

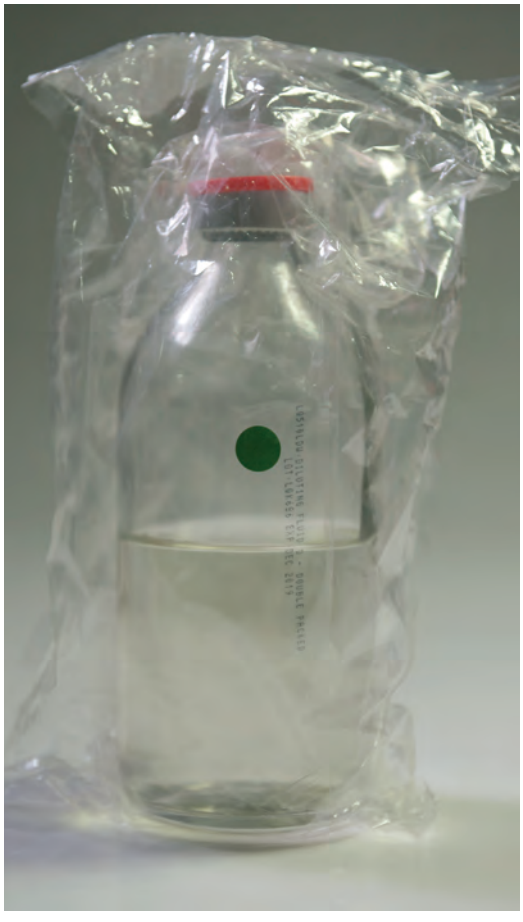
Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (2, 3).

Reference

1. The United States Pharmacopoeia USP37 2019, The US Pharmacopeial convention Inc., Rockville, MD.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

LQ510LDW



300 ml

Ready Prepared Media			
Code	**Product Name**	**Usage**	**Packing**
Category : Ready Prepared Liquid Medium in glass bottles			
LQ510CV LQ510L LQ510DC3 LQ510LDW	Diluting Fluid D Diluting Fluid D - Double packed	diluent in testing of pharmaceuticals in accordance with USP	10X100ML 5X300ML 2X800ML 5X300ML

Neutralizing Fluid ME1420 / M1420B

Intended Use:

Neutralizing Fluid is recommended for neutralizing the activity of antimicrobial agents in accordance with European Pharmacopoeia and British Pharmacopoeia.

Directions:

Suspend dehydrated medium as per table in 1000ml of purified / distilled water containing 30 gm of polysorbate 80. Heat if necessary to dissolve the medium completely. Distribute into tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Ingredients	EP	BP
	ME1420	M1420B
HMC peptone#	1.00	1.00
Lecithin (egg)	3.00	3.00
Histidine hydrochloride	1.00	1.00
Sodium chloride	4.30	4.30
Potassium dihydrogen phosphate	3.60	3.60
Disodium hydrogen phosphate dihydrate	7.20	7.20
Grams/litre	18.64	18.64
Water	Purified/ Distilled	Purified/ Distilled
Sterilization Temperature and Time	Autoclaving 121°C-15 min or as per validated cycle	Autoclaving 121°C-15 min or as per validated cycle

#Peptone (meat and casein)

Principle And Interpretation

Neutralising fluid is used to neutralize the activity of antimicrobial agents generally present in pharmaceutical materials(1).

This is required to neutralize the effect of antimicrobials while testing the sterility of such materials. This medium may be added to Buffered Sodium Chloride Peptone Solution, pH 7.0 before sterilization. If utilized their efficacy and non-toxicity towards microorganisms are demonstrated (1).

The neutralising agents present in the medium neutralises the activity of antimicrobial agents present in various pharmaceutical products which may interfere with microbial limit tests or sterility testing analysis. Egg lecithin and Polysorbate 80 (Tween 80) act as neutralising agents. Sodium chloride maintains osmotic equilibrium and phosphates serve as buffering agents.

Type of specimen

Pharmaceutical samples.

Specimen Collection and Handling

For pharmaceutical samples, follow appropriate techniques for sample collection, processing as per pharmaceutical standards (1, 2).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. The broth must be subcultured for further identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature

Neutralizing Fluid ME1420 / M1420B

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Light yellow coloured opalescent solution in tubes

Cultural Response
Cultural characteristics observed when subcultured on Tryptone Soya Agar (M290), after an incubation at 35-37°C for 40-48 hours.

Organism (ATCC)	Inoculum (CFU)	Growth
Bacillus subtilis subsp. *spizizenii* 6633 (00003*)	50 -100	good
Escherichia coli 8739 (00012*)	50 -100	good
Pseudomonas aeruginosa 9027 (00026*)	50 -100	good
Staphylococcus aureus subsp. *aureus* 6538 (00032*)	50 -100	good
Salmonella Typhimurium 14028 (00031*)	50 -100	good

Key : * corresponding WDCM number

Storage and Shelf Life
Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

Disposal
User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3, 4).

Reference

1. European Pharmacopoeia, 2017, European Department, Directorate for the Quality of Medicines of the Council of Europe.
2. British Pharmacopoeia, 2016, British Pharmacopoeia Commission.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Culture Media Cross Reference

Equivalent Media from various Pharmacopoeia

Harmonized Media

HIMEDIA	GRANULATED	HARMONIZED	HARMONIZED -GRANULATED	IP	HIVEG	HIVEG - GRANULATED	HICYNTH
Buffered Peptone Water w/ NaCl (M1275)	Buffered Peptone Water w/ NaCl, Granulated (GM1275)	Buffered Sodium Chloride-Peptone Solution, pH 7.0 (MH1275)	Buffered Sodium Chloride-Peptone Solution, pH 7.0, Granulated (GMH1275)	X	Buffered HiVeg™ Peptone Water w/ NaCl (MV1275)	X	Buffered HiCynth™ Peptone Water w/ NaCl (MCD1275)
Cetrimide Agar Base (M024)	Cetrimide Agar Base, Granulated (GM024)	Cetrimide Agar (MH024)	Cetrimide Agar, Granulated (GMH024)	X	Cetrimide HiVeg™ Agar Base (MV024)	X	Cetrimide HiCynth™ Agar Base (MCD024)
Columbia Blood Agar Base (M144)	Columbia Blood Agar Base, Granulated (GM144)	Columbia Agar (MH144)	Columbia Agar, Granulated (MH144)	X	Columbia Blood Agar Base, HiVeg™(MV144)	X	Columbia Blood HiCynth™ Agar Base (MCD144)
EE Broth, Mossel (M287)	EE Broth, Mossel , Granulated (GM287)	Enterobacteria Enrichment Broth Mossel (MH287)	Enterobacteria Enrichment Broth Mossel, Granulated (GMH287)	X	EE HiVeg™ Broth, Mossel	X	X
MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl (M081)	MacConkey Agar w/ 0.15% Bile Salts, CV and NaCl, Granulated (GM081)	MacConkey Agar (MH081)	MacConkey Agar, Granulated (GMH081)	Medium 8. MacConkey Agar (MM081)	MacConkey HiVeg™ Agar w/ CV, NaCl, 0.003% NR and 1.5% Agar (MV081)	X	MacConkey HiCynth™ Agar w/, CV and NaCl (MCD081)
MacConkey Broth Purple w/ BCP (M083)	MacConkey Broth Purple w/ BCP, Granulated (GM083)	MacConkey Broth (MH083)	MacConkey Broth, Granulated (GMH083)	Medium 7. MacConkey Broth (MM083)	MacConkey HiVeg™ Broth Purple w/ BCP (MV083)	X	X
Mannitol Salt Agar Base (M118)	Mannitol Salt Agar Base, Granulated (GM118)	Mannitol Salt Agar (MH118)	Mannitol Salt Agar , Granulated (GMH118)	X	Mannitol Salt HiVeg™ Agar Base (MV118)	X	Mannitol Salt HiCynth™ Agar Base (MCD118)
X	Potato Dextrose Agar, Granulated (GM096)	Potato Dextrose Agar, (MH096)	Potato Dextrose Agar, Granulated (GMH096)	X	X	X	Potato Dextrose HiCynth™ Agar (MCD096)
Rappaport Vassiliadis Soya Broth (RVS Broth) (M1491)	Rappaport Vassiliadis Soya Broth (RVS Broth), Granulated (GM1491)	Rappaport Vassiliadis Salmonella Enrichment Broth (MH1491)	Rappaport Vassiliadis Salmonella Enrichment Broth (GMH1491)	Medium 9. Rappaport Vassiliadis Salmonella Enrichment Broth (MM1491)	X	X	Rappaport Vassiliadis Soya HiCynth™ Broth (RVS HiCynth™ Broth) (MCD1491)
Reinforced Clostridial Broth (M443)		Reinforced Medium for Clostridia (MH443)	Reinforced Medium for Clostridia, Granulated (GMH443)	X	Reinforced Clostridial HiVeg™ Broth (MV443)	X	Reinforced Clostridial HiCynth™ Broth (MCD443)
X	Sabouraud Dextrose Agar, Granulated (GM063)	Sabouraud Dextrose Agar (MH063)	Sabouraud Dextrose Agar, Granulated (GMH063)	Sabouraud Dextrose Agar Medium 4, Granulated (GMM063)	Sabouraud Dextrose HiVeg™ Agar (MV063)	X	Sabouraud Dextrose HiCynth ™ Agar (MCD063)
X	Sabouraud Dextrose Broth (Sabouraud Liquid Medium), Granulated (GM033)	Sabouraud Dextrose Broth (MH033)	Sabouraud Dextrose Broth Granulated (GMH033)	Sabouraud Dextrose Broth Medium 3 (MM033)	Sabouraud Dextrose HiVeg™ Broth (Sabouraud Liquid HiVeg™ Medium) (MV033)	Sabouraud Dextrose HiVeg™ Broth (Sabouraud Liquid HiVeg™ Medium), Granulated (GMV033)	Sabouraud Dextrose HiCynth™ Broth (Sabouraud Liquid HiCynth™ Medium) (MCD033)

Culture Media Cross Reference

Equivalent Media from various Pharmacopoeia

Harmonized Media

HIMEDIA	GRANULATED	HARMONIZED	HARMONIZED -GRANULATED	IP	HIVEG	HIVEG - GRANULATED	HICYNTH
Soyabean Casein Digest Medium (Tryptone Soya Broth) (M011)	Soyabean Casein Digest Medium (Tryptone Soya Broth, Granulated (GM011)	Soyabean Casein Digest Medium (Casein Soyabean Digest Broth) (MH011)	Soyabean Casein Digest Medium (Casein Soyabean Digest Broth), Granulated (GMH011)	X	Soyabean HiVeg™ Medium (Tryptone Soya HiVeg™ Broth) (MV011)	Soyabean HiVeg™ Medium (Tryptone Soya HiVeg™ Broth), Granulated (MV011)	Soyabean Casein Digest HiCynth™ Medium (Tryptone Soya HiCynth™ Broth) (MCD011)
Soyabean Casein Digest Agar (Casein Soyabean Digest Agar) (Tryptone Soya Agar) (M290)	Soyabean Casein Digest Agar (Casein Soyabean Digest Agar) (Tryptone Soya Agar), Granulated (GM290)	Soybean-Casein Digest Agar (Casein-Soyabean Digest Agar) (MH290)	Soybean-Casein Digest Agar (Casein-Soyabean Digest Agar)Granulated (GMH290)	X	Soyabean Casein Digest HiVeg™ Agar (Casein Soyabean Digest HiVeg™ Agar) (Tryptone Soya HiVeg™ Agar) (MV290)	X	Soyabean Casein Digest HiCynth™ Agar (Casein Soyabean Digest HiCynth™ Agar) (Tryptone Soya HiCynth™ Agar) (MCD290)
Violet Red Bile Glucose Agar w/o Lactose (M581)	Violet Red Bile Glucose Agar w/o Lactose, Granulated (GM581	Violet Red Bile Glucose Agar (MH581)	Violet Red Bile Glucose Agar, Granulated (GMH581)	X	Violet Red Glucose HiVeg™ Agar w/o Lactose (MV581)	X	Violet Red Glucose HiCynth™ Agar w/o Lactose (MCD581)
Xylose Lysine Deoxycholate Agar (XLD Agar) (M031)	Xylose Lysine Deoxycholate Agar (XLD Agar), Granulated (GM031)	Xylose –Lysine Deoxycholate Agar (MH031)	Xylose –Lysine Deoxycholate Agar, Granulated (GMH031)	X	XLD HiVeg™ Agar (MV031	X	Xylose Lysine Deoxycholate HiCynth™ Agar (XLD HiCynth™ Agar) (MCD031)

Sterility Testing Media

HIMEDIA	GRANULATED	HARMONIZED	HARMONIZED -GRANULATED	USP	IP	EP	BP	HIVEG	HIVEG - GRANULATED	HICYNTH
Alternative Thioglycollate Medium (NIH Thioglycollate Broth) (Thioglycollate Broth, Alternative) (M010)	Alternative Thioglycollate Medium (Thioglycollate Broth, Alternative), Granulated (GM010)	X	X	Alternative Thiogly-collate Medium (MU010)	Alternative Thiogly-collate Medium (MM010)	X	X	Alternative Thioglycol-late HiVeg™ Medium (Thioglycol-late HiVeg™ Broth, Alternative) (MV010	X	Alternative Thioglycollate HiCynth™ Medium (Thioglycollate HiCynth™ Broth Alternative) (MCD010)
Fluid Thioglycollate Medium (Thioglycollate Medium, Fluid) (M009)	Fluid Thioglycollate Medium (Thioglycollate Medium, Fluid), Granulated (GM009)	X	X	Fluid Thio-glycollate Medium (MU009)	Fluid Thio-glycollate Medium (MM009)	Fluid Thiogly-collate Medium (ME009)	Fluid Thio-glycollate Medium (M009B)	Fluid Thioglycol-late HiVeg™ Medium (Thioglycol-late HiVeg™ Medium Flu-id) (MV009)	X	Fluid Thioglycollate HiCynth™ Medium (Thioglycollate HiCynth™ Medium, Fluid) (MCD009)
Soyabean Casein Digest Medium (Tryptone Soya Broth) (M011)	Soyabean Casein Digest Medium (Tryptone Soya Broth, Granulated (GM011)	Soyabean Casein Digest Medium (Casein Soyabean Digest Broth) (MH011)	Soyabean Casein Digest Medium (Casein Soyabean Digest Broth), Granulated (GMH011)	X	X	X	X	Soyabean HiVeg™ Medium (Tryptone Soya HiVeg™ Broth) (MV011)	Soyabean HiVeg™ Medium (Tryptone Soya HiVeg™ Broth), Granulated (MV011)	Soyabean Casein Digest HiCynth™ Medium (Tryptone Soya HiCynth™ Broth) (MCD011)

Culture Media Cross Reference

Equivalent Media from various Pharmacopoeia

Microbial Limit Test

HIMEDIA	GRANULATED	USP	EP	BP	IP	HIVEG	HICYNTH
Baird Parker Agar Base (M043)	Baird Parker Agar Base, Granulated (GM043)	Baird Parker Agar Medium (MU043)	Baird Parker Agar (Agar Medium O) (ME043	Baird Parker Agar (Agar Medium O) (M043B)	Baird Parker Agar Medium (MM043)	Baird Parker HiVeg™ Agar Base (MV043)	Baird Parker HiCynth™ Agar Base (MCD043)
Bismuth Sulphite Agar (M027)	Bismuth Sulphite Agar, Granulated (GM027)	Bismuth Sulphite Agar Medium (MU027)	X	X	Bismuth Sulphite Agar Medium (Twin Pack) (MM027)	Bismuth Sulphite HiVeg™ Agar (MV027)	Bismuth Sulphite HiCynth™ Agar (MCD027)
Brilliant Green Agar Base, Modified (M016)	X	Brilliant Green Agar Medium (MU016)	Brilliant Green, Phenol Red, Lactose Monohydrate, Sucrose Agar (Agar Medium L) (ME016)	Brilliant Green, Phenol Red, Lactose Monohydrate, Sucrose Agar (Agar Medium L) (M016B)	Brilliant Green Agar Medium 16 (In accordance with IP 2007) (MM016)	Brilliant Green HiVeg™ Agar Base, Modified (MV016)	Brilliant Green HiCynth™ Agar Base, Modified (MCD016)
Deoxycholate Citrate Agar (M065)	Deoxycholate Citrate Agar, Granulated (GM065)	X	Deoxycholate-Citrate Agar (Agar Medium J) (ME065)	Deoxycholate-Citrate Agar (Agar Medium J) (M065B)	Deoxycholate-Citrate Agar Medium 14 (In accordance with IP 2007) (MM065)	Deoxycholate Citrate Agar, HiVeg™ (MV065)	Deoxycholate Citrate HiCynth™ Agar (MCD065)
EMB Agar, Levine (M022)	EMB Agar, Levine, Granulated (GM022)	Levine Eosin - Methylene Blue Agar Medium (MU022)	X	X	EMB Agar, Levine (Levine - Eosin Methylene Blue Agar Medium) (In accordance with IP 2007) (MM022)	EMB HiVeg™ Agar, Levine (MV022)	X
Fluid Selenite Cystine Medium (Selenite Cystine Broth) (Twin Pack) (M025)	Fluid Selenite Cystine Medium (Selenite Cystine Broth) (Twin Pack), Granulated (GM025)	Fluid Selenite Cystine Medium (Twin Pack) (MU025)	X	X	Fluid Selenite Cystine Medium (Twin Pack) (MM025)	Fluid Selenite Cystine HiVeg™ Medium (Selenite Cystine HiVeg™ Broth) (Twin Pack) (MV025)	X
Fluid Casein Digest Soya Lecithin Medium (Twin Pack) (M117)	X	Fluid Casein Digest-Soy-Lecithin Polysorbate 20 Medium (Twin Pack) (MU117)	X	X	Fluid Casein Digest-Soya Lecithin-Polysorbate 20 Medium (Twin Pack) (MM117)	Fluid Casein Digest Soya Lecithin HiVeg™ Medium (Twin Pack) (MV117)	X
GN Broth Hajna (M242)	X	X	X	X	GN Broth (MM242)	GN HiVeg™ Broth Hajna (MV242)	X
Hektoen Enteric Agar (M467)	Hektoen Enteric Agar, Granulated (GM467)	Hektoen Enteric Agar Medium (MU467)	X	X	X	Hektoen Enteric HiVeg™ Agar (MV467)	Hektoen Enteric HiCynth Agar (MCD467)
Lactose Broth (M1003)	Lactose Broth, Granulated (GM1003)	Fluid Lactose Medium (MU1003)	Lactose Monohydrate Broth (Broth Medium D) (ME1003)	Lactose Monohydrate Broth (Broth Medium D) (M1003B)	Lactose Broth (Fluid Lactose Medium) (MM1003)	Lactose HiVeg™ Broth (MV1003)	X
Lactose Sulphite Broth Base (M1287)	X	X	Lactose Monohydrate Sulphite Medium (Medium R) (ME1287)	Lactose Monohydrate Sulphite Medium (Medium R) (M1287B)	X	X	X
Nutrient Agar w/ 1% Peptone (M012)	X	X	X	X	Nutrient Agar Medium (MM012)	Nutrient HiVeg™ Agar w/ 1% HiVeg™ Peptone (MV012)	X
Nutrient Broth w/ 1% Peptone (M244)	X	X	X	X	Nutrient Broth (MM244)	Nutrient HiVeg™ Broth w/ 1% HiVeg™ Peptone (MV244)	X
Pseudomonas Agar (For Pyocyanin) (M119)	Pseudomonas Agar, Granulated (For Pyocyanin) (GM119)	Pseudomonas Agar For Detection of Pyocyanin (MU119)	X	X	Pseudomonas Agar Medium For Detection of Pyocyanin Medium 21 (MM119)	Pseudomonas HiVeg™ Agar (For Pyocyanin) (MV119)	X

Culture Media Cross Reference

Equivalent Media from various Pharmacopoeia

Microbial Limit Test

HIMEDIA	GRANULATED	USP	EP	BP	IP	HIVEG	HICYNTH
Pseudomonas Agar (For Fluorescein) (M120)	Pseudomonas Agar, Granulated (For Fluorescein) (GM120)	Pseudomonas Agar Medium For Detection of Fluorescein (MU120)	X	X	Pseudomonas Agar Medium For Detection of Fluorescein Medium 20 (In accordance with IP 2007) (MM120)	Pseudomonas HiVeg™ Agar (For Fluorescein) (MV120)	Pseudomonas HiCynth™ Agar (For Fluorescein) (MCD120)
R-2A Agar (M962)	R-2A Agar, Granulated (GM962)	X	R2A Agar (Agar Medium S) (ME962)	R2A Agar (Agar Medium S) (M962B)	X	R-2A HiVeg™ Agar (MV962)	R-2A HiCynth™ Agar (MCD962)
Sabouraud Glucose Agar w/ Antibiotics (M1472)	X	X	Sabouraud-Glucose Agar with Antibiotics (Agar Medium C) (ME1472)	Sabouraud-Glucose Agar with Antibiotics (Agar Medium C) (M1472B)	Sabouraud Dextrose Agar Medium w/ Tetracycline (In accordance with IP 2010)	X	X
Sabouraud Chloramphenicol Agar (M1067)	X	X	Sabouraud-Glucose Agar with Chloramphenicol (Agar Medium C) (ME1067)	Sabouraud-Glucose Agar with Chloramphenicol (Agar Medium C) (M1067B)	Sabouraud Dextrose Agar with Chloramphenicol Medium 4 (In accordance with IP 2014) (MM1067)	Sabouraud Chloramphenicol HiVeg™ Agar (MV1067)	X
Selenite Broth (Selenite F Broth) (Twin Pack) (M052)	Selenite Broth (Selenite F Broth) Granulated (Twin Pack), (GM052)	X	X	X	Selenite F Broth (Twin Pack) Medium 11 (In accordance with IP 2007) (MM052)	X	X
Tetrathionate Broth Base (w/o Iodine and BG) (M032)	Tetrathionate Broth Base w/o Iodine and BG, Granulated (GM032)	Fluid Tetrathionate Medium (MU032)	X	X	Tetrathionate Broth Medium (MM032)	Tetrathionate HiVeg™ Broth Base (w/o Iodine and BG) (MV032)	Tetrathionate HiCynth™ broth Base w/o Iodine and BG (MCD032)
Tetrathionate Brilliant Green Bile Broth (M1255)	Tetrathionate Brilliant Green Bile Broth, Granulated (GM1255)	X	Tetrathionate Bile-Brilliant Green Broth (Broth Medium I) (ME1255)	Tetrathionate Bile-Brilliant Green Broth (Broth Medium I) (M1255B)	Tetrathionate Bile-Brilliant Green Broth Medium (MM1255)	Tetrathionate Brilliant Green HiVeg™ Broth (MV1255)	Tetrathionate Brilliant Green HiCynth™ Broth (MCD1255)
Triple Sugar Iron Agar (M021)	Triple Sugar Iron Agar, Granulated (GM021)	Triple Sugar-Iron-Agar Medium (MU021)	Triple Sugar, Iron Agar (Agar Medium M) (ME021)	Triple Sugar, Iron Agar (Agar Medium M) (M021B)	Triple Sugar Iron Agar (In accordance with IP 1996) (MM021)	Triple Sugar Iron HiVeg™ Agar (MV021)	Triple Sugar Iron HiCynth™ Agar (MCD021)
Urea Broth Base (Diagnostic Stuarts Urea Broth Base) (M111)	X	X	X	X	Urea Broth Medium 18 (In accordance with IP 2007) (MM111)	X	X
Violet Red Bile Agar w/ Glucose and Lactose (M1684)	X	Violet-Red Bile Agar with Glucose and Lactose (MU1684)	Agar Medium F (Crystal Violet, Neutral Red, Bile Agar with Glucose) (ME1684)	Agar Medium F (Crystal Violet, Neutral Red, Bile Agar with Glucose) (M1684B)	Crystal Violet, Neutral Red, Bile Agar with Dextrose (MM1684)	X	X
Vogel-Johnson Agar Base w/o Tellurite (V.J. Agar) (M023)	X	Vogel-Johnson Agar Medium (MU023)	X	X	Vogel Johnson Agar Medium 22.(In accordance with IP 2007) (MM023)	Vogel-Johnson HiVeg™ Agar Base w/o Tellurite (V. J. HiVeg™ Agar) (MV023)	Vogel Johnson HiCynth™ Agar Base w/o Tellurite (V.J. HiCynth™ Agar) (MCD023)

Culture Media Cross Reference

Equivalent Media from various Pharmacopoeia

Antibiotic Assay Media

HIMEDIA	GRANULATED	USP	EP	BP	IP	HIVEG	HICYNTH
Antibiotic Assay Medium No.1 (Seed Agar) (M003)	X	Antibiotic Assay Medium No.1 (MU003)	Antibiotic Assay Medium A (ME003)	Antibiotic Assay Medium A (M003B)	Antibiotic Assay Medium A (MM003)	Antibiotic HiVeg™ Assay Medium No.1 (Seed HiVeg™ Agar) (MV003	X
Antibiotic Assay Medium No. 2 (Base Agar) (M005)	X	Antibiotic Assay Medium No. 2 (MU005)	X	X	Antibiotic Assay Medium B (MM005)	Antibiotic HiVeg™ Assay Medium No. 2 (Base HiVeg™ Agar) (MV005)	X
Antibiotic Assay Medium No. 3 (Assay Broth) (M042)	X	Antibiotic Assay Medium No. 3 (MU042)	X	X	Antibiotic Assay Medium C (MM042)	Antibiotic HiVeg™ Assay Medium No. 3 (Assay HiVeg™ Broth) (MV042)	X
Antibiotic Assay Medium No. 4 (Yeast MB Agar) (M140)	X	Antibiotic Assay Medium No. 4 (MU140)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 4 (Yeast MB HiVeg™ Agar) (MV140)	X
Antibiotic Assay Medium No. 5 (Streptomycin Assay Agar w/ Yeast extract) (M006)	X	Antibiotic Assay Medium No. 5 (MU006)	X	X	Antibiotic Assay Medium E (MM006)	Antibiotic HiVeg™ Assay Medium No. 5 (Streptomycin HiVeg™ Assay Agar w/ Yeast extract) (MV006)	X
Antibiotic Assay Medium No. 6 (M223)	X	X	X	X	X	Antibiotic HiVeg™ Assay Medium No. 6 (MV223)	X
Antibiotic Assay Medium No. 8 (Base Agar w/ low pH) (M041)	X	Antibiotic Assay Medium No. 8 (MU041)	X	X	Antibiotic Assay Medium F (MM041)	Antibiotic HiVeg™ Assay Medium No. 8 (Base HiVeg™ Agar w/ low pH) (MV041)	X
Antibiotic Assay Medium No. 9 (Polymyxin Base Agar) (M147)	X	Antibiotic Assay Medium No. 9 (MU147)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 9 (Polymyxin HiVeg™ Base Agar) (MV147)	X
Antibiotic Assay Medium No. 10 (Polymyxin Seed Agar) (M225)	X	Antibiotic Assay Medium No. 10 (MU225)	X	X	Antibiotic Assay Medium H (MM225)	Antibiotic HiVeg™ Assay Medium No. 10 (Polymyxin Seed HiVeg™ Agar) (MV225)	X
"Antibiotic Assay Medium No.11 (Neomycin, Erythromycin Assay Agar) (Erythromycin Seed Agar) (M004)"	"Antibiotic Assay Medium No.11 (Neomycin, Erythromycin Assay Agar) (Erythromycin Seed Agar), Granulated (GM004)"	Antibiotic Assay Medium No. 11 (MU004)	Antibiotic Assay Medium A with pH 7.9 (ME004)	Antibiotic Assay Medium A with pH 7.9 (M004B)	Antibiotic Assay Medium D (MM004)	Antibiotic HiVeg™ Assay Medium No.11 (Neomycin, Erythromycin HiVeg™ Assay Agar) (Erythromycin Seed HiVeg™ Agar) (MV004)	X
Antibiotic Assay Medium No. 12 (Nystatin Assay Agar) (M280)	X	X	X	X	X	Antibiotic HiVeg™ Assay Medium No. 12 (Nystatin HiVeg™ Assay Agar) (MV280)	X
Antibiotic Assay Medium No. 13 (Nystatin Assay Broth) (M254)	X	Antibiotic Assay Medium No. 13 (MU254)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 13 (Nystatin HiVeg™ Assay Broth) (MV254)	X

Culture Media Cross Reference

Equivalent Media from various Pharmacopoeia

Antibiotic Assay Media

HIMEDIA	GRANULATED	USP	EP	BP	IP	HIVEG	HICYNTH
Antibiotic Assay Medium No. 19 (M101)	X	Antibiotic Assay Medium No. 19 (MU101)	X	X	Antibiotic Assay Medium G (MM101)	Antibiotic HiVeg™ Assay Medium No. 19 (MV101)	X
Antibiotic Assay Medium No. 20 (Yeast MB Broth) (M167)	X	X	X	X	X	Antibiotic HiVeg™ Assay Medium No. 20 (Yeast HiVeg™ Broth) (MV167)	X
Antibiotic Assay Medium No. 32 (M1141)	X	Antibiotic Assay Medium No. 32 (MU1141)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 32 (MV1141)	X
Antibiotic Assay Medium No. 34 (M797)	X	Antibiotic Assay Medium No. 34 (MU797)	X	X	X	X	X
Antibiotic Assay Medium No. 35 (M798)	X	Antibiotic Assay Medium No. 35 (MU798)	X	X	Antibiotic Assay Medium I (MM798)	Antibiotic HiVeg™ Assay Medium No. 35 (MV798)	X
Antibiotic Assay Medium No. 36 (M1666)	X	Antibiotic Assay Medium No. 36 (MU1666)	X	X	Antibiotic Assay Medium J (MM1666)	Antibiotic HiVeg™ Assay Medium No. 36 (MV1666)	X
Antibiotic Assay Medium No. 37 (M1667)	X	Antibiotic Assay Medium No. 37 (MU1667)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 37 (MV1667)	X
Antibiotic Assay Medium No. 38 (M799)	X	Antibiotic Assay Medium No. 38 (MU799)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 38(MV799)	X
Antibiotic Assay Medium No. 39 (M1142)	X	Antibiotic Assay Medium No. 39 (MU1142)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 39 (MV1142)	X
Antibiotic Assay Medium No. 40 (M1143)	X	Antibiotic Assay Medium No. 40 (MU1143)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 40 (MV1143)	X
Antibiotic Assay Medium No. 41 (M1144)	X	Antibiotic Assay Medium No. 41 (MU1144)	X	X	X	Antibiotic HiVeg™ Assay Medium No. 41 (MV1144)	X
Antibiotic Assay Medium B (M1346)	X	X	Antibiotic Assay Medium B (ME1346)	Antibiotic Assay Medium B (M1346B)	X	X	X
Antibiotic Assay Medium C (M555)	X	X	Antibiotic Assay Medium C (ME555)	Antibiotic Assay Medium C (M555B)	X	X	X
Antibiotic Assay Medium D (M556)	X	X	Antibiotic Assay Medium D (ME556)	Antibiotic Assay Medium D (M556B)	X	X	X
Antibiotic Assay Medium E (M1347)	X	X	Antibiotic Assay Medium E (ME1347)	Antibiotic Assay Medium E (M1347B)	X	X	X
Antibiotic Assay Medium F (M923)	X	X	Antibiotic Assay Medium F (ME923)	Antibiotic Assay Medium F (M923B)	X	X	X
Antibiotic Assay Medium G (M553)	X	X	Antibiotic Assay Medium G (ME553)	Antibiotic Assay Medium G (M553B)	X	X	X
X	X	X	Antibiotic Assay Medium H (ME1665)	X	X	X	X
Antibiotic Assay Medium H (M1665)	X	X	X	Antibiotic Assay Medium I (M1847B)	X	X	X

Culture Media Cross Reference

Equivalent Media from various Pharmacopoeia

Neutralizing Media

HIMEDIA	GRANULATED	USP	EP	BP	HIVEG	HICYNTH
Dey-Engley Neutralizing Broth (M1062)	X	Dey-Engley (D/E) Neutralizing Broth (without Bromo cresol purple) (MU1062)	X	X	Dey-Engley Neutralizing HiVeg™ Broth (MV186)	Dey Engley Neutralizing HiCynth™ Broth (MCD186)
T.A.T. Broth Base (M562)	T.A.T. Broth Base, Granulated (GM562)	T.A.T. Broth with Tween 20 (MU562)	X	X	T.A.T. HiVeg™ Broth Base (MV562)	X
Letheen Broth, AOAC (M165)	X	Letheen Broth (MU165)	X	X	Letheen HiVeg™ Broth, AOAC (MV165)	X
Diluting Fluid A (M1415)	X	X	X	X	X	X
Diluting Fluid K (M1416)	X	X	X	X	X	X
Diluting Fluid D (M1686)	X	X	X	X	X	X
X	X	X	Neutralizing fluid (ME1420)	Neutralizing fluid (M1420B)	X	X

Pharmaceutical Cultures-Cross Reference

Organisms	IP 2010	BP 2011	USP 2011	EP 2011
Staphylococcus aureus	6538	6538	6538	6538 / NCIMB 9518
Pseudomonas aeruginosa	9027	9027	9027	9027
Escherichia coli	8739 / NCTC 9002	8739	8739	8739
Salmonella Typhimurium	X	X	14028	14028
Bacillus subtilis	6633	6633	6633	X
Candida albicans	10231	10231	10231	10231
**Aspergillus brasiliensis*	16404	16404	16404	X
Salmonella Abony	NCTC 6017	NCTC 6017	X	NCTC 6017
Clostridium sporogenes	X	19404	11437/19404	19404
Bacteroides vulgatus	8482	X	X	X
Clostridium perfringens	X	13124	X	13124
Antibiotic Assay				
Staphylococcus aureus	29737/9144	X	29737/9144	6538P
Saccharomyces cerevisiae	9763/2601	X	9763/2601	9763
Micrococcus luteus	10240	X	10240/9341	NCTC 7743, ATCC 10240
Mycobacterium smegmatis	607	X	607	607
Pseudomonas aeruginosa	25619	X	25619	X
Bacillus pumilis	14884	X	X	NCTC 8241
Micrococcus luteus	9341	X	X	X
Bacillus subtilis	6633	X	6633	NCTC 8236, ATCC 6633
Staphylococcus epidermidis	12228	X	12228	X
Bacillus cereus var mycoides	11778	X	X	X
Bordetella bronchiseptica	4617	X	4617	NCTC 8344, ATCC 4617
Klebsiella pneumoniae	10031	X	10031	10031
Escherichia coli	X	X	10536	10536/9637
Enterococcus hirae	X	X	10541	10541
Candida tropicalis	X	X	X	NCYC 1393
Clostridium sporogenes	19404	X	X	X
**Aspergillus brasiliensis*	16404	X	X	X
Candida albicans	10231/2091 NCYC 854	X	X	X

Key * - Formerly known as *Aspergillus niger*.

Culture Media Alphabetical Reference

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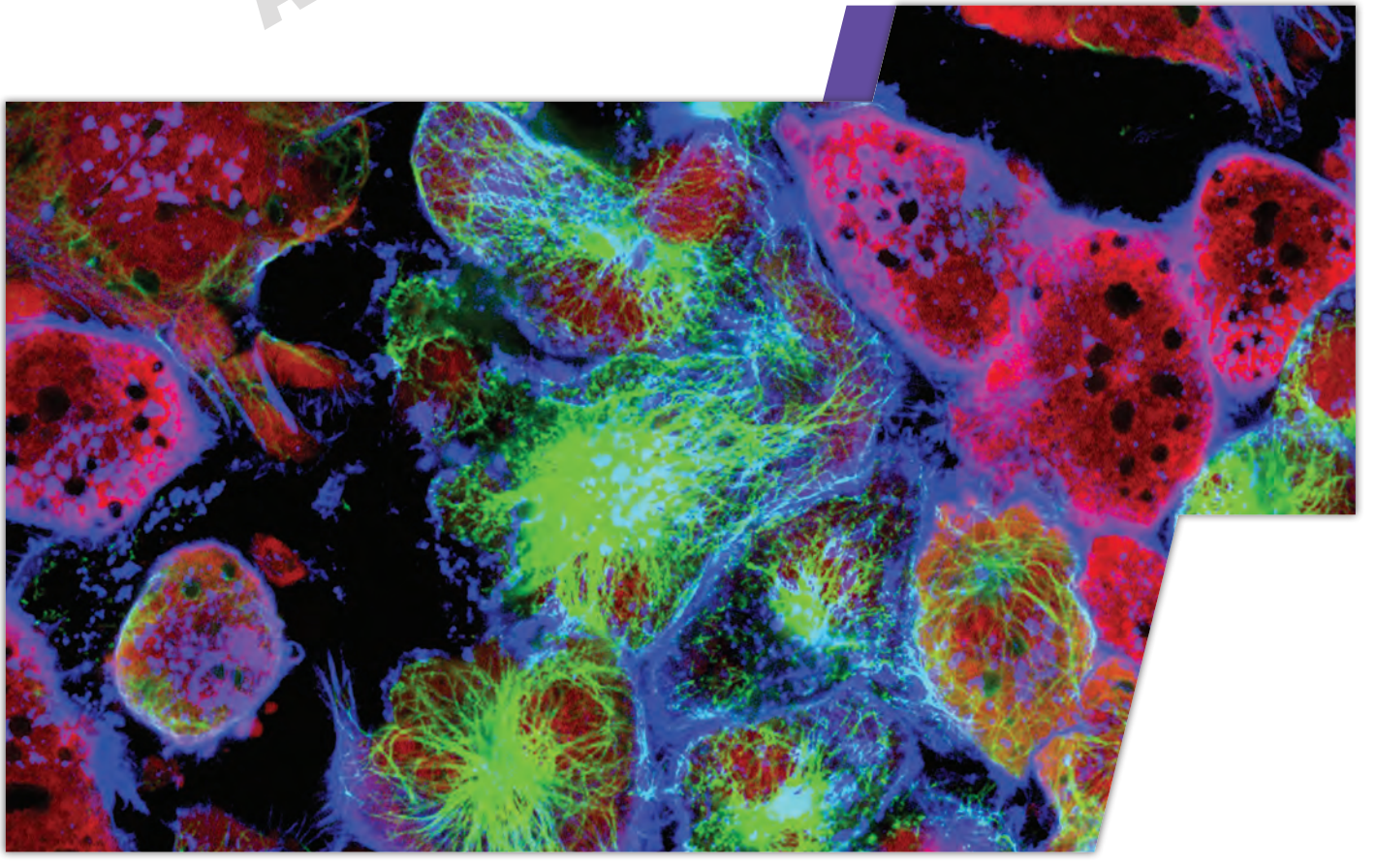

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- CORPORATE OFFICE -

A-516, Swastik Disha Business Park, Via Vadhani Indl Est, LBS Marg,
Mumbai - 400 086, India.
Tel : +91-22-6147 1919 / 2500 3747 | Fax : +91-22-6147 1920 / 2500 5764
Email : info@himedialabs.com

- OVERSEAS OFFICES -

USA & Canada

HiMedia Laboratories LLC, 107 W Dorothys Way, Lincoln University, West Chester,
Pennsylvania 19352, USA.
Tel : +1-484-734-4401 | Fax : +1-484-734-4402
Email : infous@himedialabs.com

Europe

HiMedia Laboratories GmbH, Marie-Curie-Str. 3, 64683,
Einhausen, Germany.
Tel : +49 6251 989 24 26 | Fax : +49 6251 989 24 27
Email : infoeu@himedialabs.com

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