Pseudomonas CN Agar Base ISO

Cat. 1153

For the identification and enumeration of Pseudomonas aeruginosa by membrane filtration

Practical information

Aplications	Categories
Selective enumeration	Pseudomonas aeruginosa
Detection	Pseudomonas aeruginosa

Industry: Water

Regulations: ISO 11133 / ISO 16266

Principles and uses

Pseudomonas CN Agar Base is used for the identification of Pseudomonas aeruginosa by membrane filtration technique, based on the detection of pyocyanin production. It is a modification of Pseudomonas P Agar (King A Medium - Cat. 1531). This medium is recommended by ISO 16266.

Pseudomonas aeruginosa is an opportunist pathogen for humans, capable of growing in water with a low concentration of nutrients. This is why natural mineral water and spring water should be checked to be free of Pseudomonas aeruginosa at the time of commercialization. This microorganism can also be found in swimming pool water.

Peptone and casein provide nitrogen, vitamins, minerals and amino acids essential for growth. Cetrimide is added as a selective agent, and nalidixic acid to suppress contaminants of cetrimide media such as Klebsiella, Proteus and Providencia spp. Potassium sulfate and magnesium chloride provide cations to activate pyocyanin production and enhance pigment production. Bacteriological agar is the solidifying agent.

Formula in g/L

Bacteriological agar	13	Cetrimide	0,2
Gelatin peptone	16	Magnesium chloride anhydrous	1,4
Nalidixic acid	0,015	Casein hydrolysate	10
Anhydrous potassium sulfate	10		

Typical formula g/L * Adjusted and/or supplemented as required to meet performance criteria.

Preparation

Suspend 50,6 grams of the medium in one liter of distilled water. Add 10 ml of glycerol. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 118 ºC for 15 minutes. Cool to 45-50 ºC, mix well and dispense into plates to obtain an agar layer of at least 5 mm thick. Do not remelt the medium.

Instructions for use

According to ISO 16266 for the detection and enumeration of Pseudomonas aeruginosa:
- Filter a certain volume of water sample through a filter membrane and place the membrane on a Pseudomonas CN Agar Base plate (Cat. 1153).
- Incubate at a temperature of 36±2 ºC for 44±4 h.
- Count the colonies that have a green/blue pigmentation (pyocyanin) as confirmed P. aeruginosa.
- Examine the membrane under UV light.
- All colonies that are fluorescence (+) and reddish-brown colonies should be confirmed.
- Spread all the colonies that should be confirmed on Nutrient Agar plates (Cat. 1156) to obtain pure cultures. Incubate at 36±2 ºC for 22±2 h
- Perform oxidase assay to the reddish-brown colonies.
- Streak the oxidase (+) colonies on King B Medium (Cat. 1532) to check the fluorescence production. Incubate at 36±2 ºC for up to 5 days. Normally 24 hours are enough.
- Inoculate all the fluorescence (+) colonies, both in CN agar and in King B Medium, in the Acetamide Broth (Cat. 1155 o Cat.2017) medium and add one or two drops of Nessler reagent to check the ammonia production. Incubate at 36±2 ºC for 22±2 h.
- The colonies that produce pyocyanin in CN agar, the colonies fluorescence (+) in CN agar and ammonia (+) in Acetamide broth, and the reddish brown colonies in CN agar, oxidase (+), fluorescence (+) in King B Agar and ammonia (+) in Acetamide Broth, are counted as confirmed P. aeruginosa.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Beige	Amber slightly opalescent	7,1±0,2

Microbiological test

According to ISO 11133:
Incubation conditions: (36±2 ºC / 22±2 h)
Inoculation conditions: Productivity quantitative (100±20.Min.50 CFU), Selectivity (10^4-10^6 CFU).
Reference media: TSA.

Microorganisms	Specification	Characteristic reaction
Enterococcus faecalis ATCC 19433	Total inhibition (0)	
Escherichia coli ATCC 25922	Total inhibition (0)	
Pseudomonas aeruginosa ATCC 27853	Good growth >50%	Blue-green colonies with fluorescence under UV light (360 ± 20 nm)
Pseudomonas aeruginosa ATCC 9027	Good growth >50%	Blue-green colonies with fluorescence under UV light (360 ± 20 nm)

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

UNE-EN 12780: 2002, Quality of water. Identification and enumeration of Pseudomonas aeruginosa by membrane filtration.
EN ISO 16266 Water quality -- Detection and enumeration of Pseudomonas aeruginosa -- Method by membrane filtration

Todd Hewitt Broth

Cat. 1236

For the cultivation of ß-hemolytic streptococci for serologic typing from clinical samples

Practical information

Aplications	Categories
Enrichment	Streptococcus

CE
IVD

Principles and uses

Todd Hewitt Broth is recommended for the cultivation of streptococci and other fastidious microorganisms. It was originally developed for the production of streptococcal hemolysin. The broth was modified by Updyke and Nickle and is used preferentially to cultivate beta-hemolytic strains, especially for serological typing, from clinical specimens and for epidemiological studies.

The medium is also recommended as an enrichment medium for the growth of streptococcal cells in the identification of Groups A and B. This medium was used as an enrichment broth for Group A streptococci in a comparison study of a rapid antigen test.

Bacteriological Peptone and Beef Heart infusion provide nitrogen, vitamins, minerals and amino acids essential for growth. Disodium phosphate and Sodium carbonate act as a buffer to prevent the destruction of the hemolysin by the acid produced through fermentation of the carbohydrate Dextrose, source of carbon and energy. Sodium chloride maintains the osmotic balance of medium.

Formula in g/L

Bacteriological peptone	20	Dextrose	2
Disodium phosphate	0,4	Sodium carbonate	2,5
Sodium chloride	2	Heart infusion	3,1

Preparation

Suspend 30 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 121°C for 15 minutes.

Instructions for use

Inoculate and incubate tubes at 35 ± 2°C for 18 - 48 hours.

To prepare Todd Hewitt Agar, add 13 - 1 5 g/l of Bacteriological Agar (Cat. 1800/1802) to the broth and sterilize as above.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Clear beige	Amber	7,8 ± 0,2

Microbiological test

Incubation conditions: (35±2 ºC / 18-48 h)

Microorganisms	Specification
Neisseria meningitidis ATCC 13090	Good growth

Streptococcus pyogenes ATCC 19615	Good growth
Streptococcus pneumoniae ATCC 6305	Good growth

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

Todd and Hewitt J. Path l Bact. 35:973. 1932 Updyke and Nickle. Applied. Microbiol 2: 117. 1954
Diagnostic Procedures and Reagents. 4th Ed. APHA Inc. New York 1963.
Isenberg H.D. (ed) 1992. Clinical Microbiology procedures handbook, American Society for Microbiology, Washington, D.C. Murray, P.R., E. J. Baron, M.A. Pfaller, F,C, Tencver and R.H. Yolken (ed) 1995 Manual of clinical Microbiology, 6th ed. American Society for Microbiology, Washington, D.C

Chromogenic Cronobacter Isolation Agar (CCI) ISO

Cat. 1446

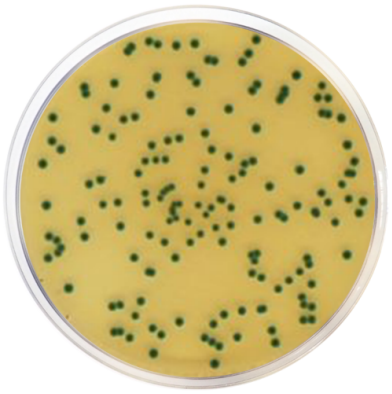
For the isolation of presumptive Cronobacter spp. in food products and environmental samples.

Practical information

Aplications	Categories
Selective isolation	Cronobacter

Industry: Food

Regulations: ISO 22964



Principles and uses

Chromogenic Cronobacter Isolation Agar (CCI) is a selective medium for the detection of Cronobacter spp. in food products and ingredients intended for human consumption and the feeding of animals, and environmental samples in the area of food production and food handling.

ISO 22964:2016 describes a horizontal method for the detection of Cronobacter spp. and recommend this medium for the isolation of Cronobacter spp.

Triptone provide nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is source of vitamins, particularly the B-group, essential for bacterial growth. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Sodium deoxycholate inhibits the accompanying gram positive flora. Sodium thiosulfate increase the selectivity and the recovery of Cronobacter and Enterobacter species. 5-Bromo-4-chloro-3-indolyl a-D-glucopyranoside is the chromogenic substrate.
Cronobacter (formerly Enterobacter sakazakii) is currently considered and emerging pathogen responsible for severe meningitis and necrotic enterocolitis in un-weaned babies that can be the cause of mortality rate between 40-80%.

The pathogenicity of Cronobacter for un-weaned babies' makes it necessary to review the manufacturing process of the milk-based products specialized for babies, guaranteeing the absence of the bacteria in the final product
Additional prevention measures at a hospital include the sanitary hygiene of the prepared food; reducing the time between the preparation and its administration, to impede the multiplication of microorganisms.

Formula in g/L

Bacteriological agar	15	Ferric ammonium citrate	1
Sodium chloride	5	Sodium deoxycholate	0,25
Sodium thiosulfate	1	Yeast extract	3
Tryptic digest of casein	7	5-bromo-4-chloro-3-indolyl-alpha-D-glucopyranoside	0,15

Preparation

Suspend 32,4 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121 ºC for 15 minutes. Cool to 50 ºC, homogenize gently and dispense into Petri dishes in amounts of 15 ml.

Instructions for use

According to ISO 22964:
- Pre-enrich the test portion in a non-selective medium such as Buffered Peptone Water BPW (Cat. 1402).
- Incubate at a temperature of 34-38 °C for 18±2 h.
- Inoculate the culture obtained in BPW in a selective medium of enrichment: Selective Broth for Cronobacter (CSB) (Cat. 2143).
- Incubate at a temperature of 41,5±1 °C for 24±2 h.

- Sow and identify the colonies in the Chromogenic Cronobacter Isolation Agar (CCI) (Cat. 1446).
- Incubate at a temperature of 41,5±1 °C for 24±2 h.
- For confirmation, typical colonies are selected from chromogenic agar, purified on a non-selective agar such as TSA (Cat. 1068) and characterized biochemically.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Beige	Slightly amber	7,3±0,2

Microbiological test

Incubation conditions: (41,5±1 ºC / 24±2 h).
Inoculation conditions: Productivity quantitative (100±20. Min. 50 CFU) / Selectivity (10^4-10^6 CFU) / Specificity (10^3-10^4 CFU).

Microorganisms	Specification	Characteristic reaction
Enterobacter cloacae ATCC 13047	Growth (1-2)	The colonies do not have green or greenish-blue color.
Staphylococcus aureus ATCC 25923	Total inhibition (0)	
Cronobacter sakazakii ATCC 29544	Good growth (2)	Blue-green colonies of small to medium size (1-3 mm)
Cronobacter muytjensii ATCC 51329	Good growth (2)	Blue-green colonies of small to medium size (1-3 mm)

Storage

Temp. Min.:2 ºC
Temp. Max.:8 ºC

Bibliography

ISO normative 22964:2016 Microbiology of the food chain — Horizontal method for the detection of Cronobacter spp.
GUILLAUME-Gentil, O., Sonnard, V. Kandahai, M.C., Mauragg, J.D. and Jootsen, H. A simple and Rapad Cultural Method for Detection of Enterobacter Sakazakii in environmental samples. Journal of Food. Protection, 68 (1), 2005, pp. 64-69.

Condalab

Reference: 6004 **Technical Data Sheet**

Product: **PALCAM LISTERIA SELECTIVE SUPPLEMENT ISO**

CE IVD

Specification

A sterile selective supplement used for the isolation of *Listerias spp.*

Presentation

	Packaging Details	Shelf Life	Storage
10 Freeze dried vials Vial with: 3 ± 0.1 g	23x60 mm glass vials, tag labelled, White plastic cap - 10 vials per box.	49 months	2-25 ºC

Composition

Compositon (g/vial)

Polymyxin B.....0.0050
Acriflavine.....0.0025
Ceftazidime.....0.0100

NOTE : Each vial is sufficient to supplement 500 ml of PALCAM medium Base.

Reconstitute the original freeze-dried vial
by adding
Sterile Distilled Water.....6 ml

Description /Technique

Description:

Listeria PALCAM selective supplement is added to PALCAM Medium base in order to obtain a complete selective medium used for the detection and the isolation of *Listeria monocytogenes* from foods.

Palcam Agar is based on the formulation described initially by van Netten *et al.* which has a high selectivity and produces good colonial differentiation. Selectivity is achieved by the inclusion of lithium chloride, acriflavine, polymyxin B and ceftazidime, since they inhibit the growth of almost all the Gram negative and most of the Gram positive companion bacteria.

Listeria hydrolyze esculin to esculetin, which reacts with ferric ammonium citrate producing a dark precipitate and green-grey colonies with beige halos. If colonies of enterococci or staphylococci do grow on this medium they can be easily recognized, since they utilise mannitol and produce yellow colonies and haloes, contrasting with the cherry-red colour of medium. However, when there are many *Listeria* colonies, the entire medium darkens, which can cause interference in differentiation. In these cases it is advisable to perform the inoculation with a more diluted sample.

Technique:

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.
Reconstitute the vial with the 6 ml sterile distilled water in aseptic conditions and add it to 500 ml of sterilized PALCAM agar base cooled to 50ºC. Do not overheat once suplemented.
Pour the complete medium into Petri dishes and, once solidified on a flat surface, spread the plates by streaking methodology or by spiral method.
Incubate the plates in aerobic atmosphere at 37 ± 1ºC for 44 ± 4h.
(Incubation times longer than those mentioned above or different incubation temperatures may be requied depending on the sample, on the specifications,...)
After incubation, enumerate all the colonies that have appeared onto the surface of the agar, observing any blackening of the medium due to esculin hydrolysis, typical for Listeria strains.

Presumptive isolation of *Listeria sp.* must be confirmed by further microbiological and biochemical tests.

Quality control

Physical/Chemical control

Color : Orange pH: at 25ºC

Microbiological control

Reconstitute 1 vial as indicated in COMPOSITION; shake and dissolve completely
Add 1 vial to 500 ml of medium base. DO NOT HEAT once supplemented.
Isolation by loop spreading
Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020

Aerobiosis. Incubation at 37 ºC ± 1, reading after 44 ± 4h

Microbiological control according to the current version of the ISO 11133:2014/A1:2018.

Microorganism	Growth
L. monocytogenes ATCC® 13932, WDCM 00021	Good - Esculin Positive reaction
Escherichia coli ATCC® 25922, WDCM 00013	Inhibited
Enterococcus faecalis ATCC® 29212, WDCM 00087	Inhibited
L. monocytogenes ATCC® 7644	Good - Esculin Positive reaction

Sterility Control

Incubation 48 h at 30-35 ºC and 48 h at 20-25 ºC: NO GROWTH.
Check at 7 days after incubation in same conditions.
Add 5mL of the sample to 100 mL of TSB and to 100 mL Thioglycollate.

Bibliography

· ATLAS, R.M. (1993) Handbook of Microbiological Media. CRC Press Boca Raton Florida.
· ISO 11290 standard (1996) Microbiology of food and animal feeding stuff. Horizontal method for the detection and enumeration of Listeria monocytogenes. Part 1 - Detection method. Part 2 - Enumeration method.
· ISO 11290-1:2017 Standard. Microbiology of the food chain. Horizontal method for the detection and enumeration of Listeria monocytogenes and for Listeria spp.- Part 1: Detection Method
· ISO 11290-2:2017 Standard. Microbiology of the food chain. Horizontal method for the detection and enumeration of Listeria monocytogenes and for Listeria spp.- Part 2: Enumeration Method
. ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
· VANDERZANT, C. & D.F. SPLITTSTOESSER (1992) Compendium of methods for the microbiological examination of foods. APHA. Washington DC.
· Van NETTEN, P., J. PERALES, A.van deMOOSDUCK, G.D.W. CURTIS & D.A.A. MOSSEL (1989) Liquid and solid selective differential media for the detection and enumeration of Listeria monocytogenes. Int. J. Food Microbiol. 8:299-316.

Condalab

Reference: 6021 **Technical Data Sheet**

Product: **BACILLUS CEREUS SUPPLEMENT ISO**

Specification

Sterile selective supplement used for *Bacillus cereus* isolation and enumeration in food samples.

Presentation

	Packaging Details	Shelf Life	Storage
10 Freeze-dried vials Vial with: 3 ± 0.1 g	22±0.25 x 55±0.5 mm glass vials, tag labelled, White plastic cap - 10 vials per box.	49 months	2-25 ºC

Composition

Composition (IU/vial)

Polymyxin B sulphate.....50.000 IU
Excipient (sufficient amount)

Reconstitute the original freeze-dried vial
by adding
Sterile Distilled Water.....6 ml

NOTE : Each vial is sufficient to supplement 500 ml of *Bacillus cereus* agar base.

Description /Technique

Description:

This supplement is recomended for Bacillus Cereus Selective Agar, following PEMBA formulation and/or MYP one.
These media permit an easily and readly detectation of a small number of *Bacillus Cereus* in a presence of a large number of food contaminants *: Bacillus cereus* grows in very typical colonies and it allows a rapid macroscopic identification.
PEMBA= blue colonies, surrounded by a clear zone of egg yolk
MYP= brilliant pink opaque colonies, with clear lecithinase halo

Technique:

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.
Reconstitute the vial with the sterile diluent in aseptic conditions and add it to 450 ml of melted Agar base cooled to 50ºC, previously supplemented also with 50 ml of sterile Egg Emulsion.
Do not overheat once suplemented.
Pour the complete medium into Petri dishes and, once solidified on a flat surface, spread the plates either by streaking or by spiral method.
Incubate the plates in aerobic atmosphere at 30 ± 1ºC for 24-48h.
Incubation times longer than those mentioned above or different incubation temperatures may be requied depending on the sample or the specifications.
After incubation, count all the colonies that have appeared onto the surface of the agar.
Presumptive isolation of *Bacillus cereus* must be confirmed by further microbiological and biochemical tests.

Quality control

Physical/Chemical control

Color : White-Gray pH: at 25ºC

Microbiological control

Reconstitute 1 vial as indicated in COMPOSITION; shake and dissolve completely

Distribute the complete medium, cooled at 50ºC, in plates

Inoculate: Practical range 100 ± 20 CFU. Min. 50 CFU (Productivity)/ 10^4-10^6 (Selectivity).

Aerobiosis. Incubation at 35ºC ± 2 ºC, reading at 24-48 hours

Microorganism	Growth
Bacillus cereus ATCC® 11778, WDCM 00001	Good
Escherichia coli ATCC® 25922, WDCM 00013	Inhibited

Sterility Control

Add 5 ml of the sample to:
100 ml TSB and 100 ml Thioglycollate.
Incubation 48 hours at 30-35 ºC and 48 hours at 20-25 ºC: NO GROWTH.

Bibliography

· ATLAS, R.M. & L.C. PARKS (1993) Handbook of Microbiological Media. CRC Press. London.

· CORRY, J.E.L., G.D.W. CURTIS & R.M. BAIRD. (2003) Handbook of Culture Media for Food Microbiology. Elsevier Sci. B.V. Amsterdam. The Netherlands.

· DOWNES, F.P. & K. ITO (2001) Compendium of methods for the microbiological examination of foods. 4th ed. APHA. Washington DC. USA.

· FIL-IDF 181:1998 Provisional Int. Standard. Dried Milk Products. Enumeration of *Bacillus cereus*.- Most probable number technique.

· ISO 7932 Standard (2004) 3rd ed. Microbiology of food and animal feeding stuffs. Horizontal method for the enumeration of presumptive *Bacillus cereus*. Colony count technique at 30ºC.

. ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.

· ISO 21871 Standard (2006) Microbiology of food and animal feeding stuffs.- Horizontal method for the determination of low numbers of presumptive *Bacillus cereus*.- Most probable number technique and detection method.

· MOSSEL, D.A.A., KOOPMAN. M.J. & JONGERIUS, E. (1967) Enumeration of *Bacillus cereus* in foods. Appl. Microbiol. 15:650-653.

· PASCUAL ANDERSON, Mª.Rª (1992) Microbiología Alimentaria. Díaz de Santos, S.A. Madrid.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

BUFFERED PEPTONE WATER (ISO) CM1049

BUFFERED PEPTONE WATER (ISO) **CM1049**

Typical Formula*

Peptone	grams per litre	10.0
Sodium chloride		5.0
Disodium hydrogen phosphate (anhydrous)		3.5
Potassium dihydrogen phosphate		1.5

* adjusted as required to meet performance standards

Directions

Dissolve 20g in 1 litre of distilled water. Mix well and distribute into final containers. Sterilize by autoclaving at 121°C for 15 minutes.

Physical Characteristics

Light straw to straw, free-flowing powder
Colour on reconstitution - straw 1 to straw 3
Moisture level - less than or equal to 7%
pH 7.0 ± 0.2 at 25°C
Clarity - clear
Buffering capacity test - passes test

Microbiological Tests Using Optimum Inoculum Dilution

Control Media: Tryptone Soya Agar or Columbia Blood Agar Base enriched with 5% v/v horse blood, where appropriate

Inoculate 9ml of the medium with 1ml of the test organism containing more than or equal to 5E+04 cfu/ml. At time zero (0 minutes) and after holding at 20-25°C for 45minutes to 1 hour (for *Escherichia coli and Staphylococcus aureus*) or 18-22°C for 1 hour ± 5 minutes (for *Listeria monocytogenes*), subculture onto control medium.

Reactions after incubation at 37 ± 2°C for 18 ± 2 hours

Tested as a non-selective pre-enrichment broth

Medium is challenged with 10-100 colony forming units

Salmonella nottingham	NCTC7832	Turbid growth
Salmonella poona	NCTC4840	Turbid growth
Escherichia coli	ATCC®11775	Turbid growth

A satisfactory result is represented by visible growth.

Testing performed in accordance with ISO11133:2014

Reactions after incubation at 37 ± 2°C for 18 ± 2 hours

Tested as a non-selective pre-enrichment broth

Medium is challenged with 10-100 colony forming units

Salmonella typhimurium	ATCC®14028	WDCM00031	Turbid growth
Salmonella enteritidis	ATCC®13076	WDCM00030	Turbid growth
Escherichia coli	ATCC®25922	WDCM00013	Turbid growth
Escherichia coli	ATCC®8739	WDCM00012	Turbid growth

A satisfactory result is represented by visible growth.

Reactions after incubation at 37 ± 2°C for 18 ± 2 hours

Tested as a diluent

Medium is challenged with 50-150 colony forming units

Escherichia coli	ATCC®8739	WDCM00012	1-2mm white/grey colonies
Escherichia coli	ATCC®25922	WDCM00013	1-2mm white/grey colonies
Staphylococcus aureus	ATCC®25923	WDCM00034	0.5-1mm white/grey colonies
Listeria monocytogenes	ATCC®35152	WDCM00109	1-2mm white/grey colonies
Listeria monocytogenes	ATCC®13932	WDCM00021	1-2mm white/grey colonies

A satisfactory result is represented by recovery of ± 30% of the Control cfu (0 minutes) after holding at 20-25°C for 45 minutes (*Escherichia coli and Staphylococcus aureus*) or 18-22°C for 1 hour (*Listeria monocytogenes*).

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

BUFFERED PEPTONE WATER (ISO) CM1049

Testing performed in accordance with ISO22964:2017

Reactions after incubation at 36 ± 2°C for 18 ± 2 hours

Medium is challenged with 10-100 colony forming units

Cronobacter sakazakii	ATCC®29544	WDCM00214	Turbid growth
Cronobacter muytjensii	ATCC®51329	WDCM00213	Turbid growth

A satisfactory result is represented by visible growth from an inoculum of 10-100 colony forming units.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

BUFFERED PEPTONE WATER (ISO) CM1049

Revision History

Section / Step	Description of Change	Reason for Change	Reference
Physical characteristics	Change of colour	Change control	MOC-2024-1290

Distribution: Central File

Date: 11/12/12
Supersedes: 14/10/09

OXOID QUALITY ASSURANCE
PRODUCT SPECIFICATION

NUTRIENT BROTH **CM0001**

Typical Formula*

'Lab-Lemco' powder	grams per litre	1.0
Yeast extract		2.0
Peptone		5.0
Sodium chloride		5.0

* adjusted as required to meet performance standards

Directions

Dissolve 13g in 1 litre of distilled water. Mix well and distribute into final containers. Sterilize by autoclaving at 121°C for 15 minutes.

Physical Characteristics

Straw, free-flowing powder
Colour on reconstitution - straw 1-2
Moisture level - less than 7%
pH 7.4 ± 0.2 at 25°C
Clarity - clear

The medium is tested for compatibility using 10% v/v oxalated horse blood, defibrinated horse blood or defibrinated sheep blood. There shall be no evidence of lysis, after incubation at 37°C and 4°C for 18 hours.

Microbiological Tests Using Optimum Inoculum Dilution

Control Media: Tryptone Soya Agar or Columbia Blood Agar Base enriched with 5% v/v horse blood, where appropriate.

Reactions after incubation at 37°C for 18 hours

Medium is challenged with less than 100 colony-forming units

Enterococcus faecalis	ATCC® 19433	Turbid growth
Streptococcus pyogenes	ATCC® 19615	Turbid growth
Staphylococcus aureus	ATCC® 25923	Turbid growth
Escherichia coli	ATCC® 25922	Turbid growth
Bacillus subtilis	ATCC® 6633	Turbid growth
Pseudomonas aeruginosa	ATCC® 27853	Turbid growth

A satisfactory result is represented by visible growth.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

SABOURAUD DEXTROSE LIQUID MEDIUM (CM0147)

SABOURAUD DEXTROSE LIQUID MEDIUM **CM0147**

Typical Formula*

Glucose	grams per litre	20.0
Pancreatic digest of casein		5.0
Peptic digest of animal tissue		5.0

* adjusted as required to meet performance standards

Directions

Dissolve 30g in 1 litre of water (purified, as required). Mix well and distribute into final containers. Sterilize by autoclaving at 121°C for 15 minutes.

Physical Characteristics

Light straw, free-flowing powder
Colour on reconstitution - straw 2-3
Moisture level - less than 7%
pH 5.6 ± 0.2 at 25ºC
Clarity - clear

Microbiological Tests Using Optimum Inoculum Dilution

Control Medium: Sabouraud Dextrose Agar

Reactions after incubation at 20-25°C for 2-5 days

Medium is challenged with 10-100 colony-forming units

Saccharomyces carlsbergensis	ATCC® 2700	Turbid growth
Penicillium chrysogenum	ATCC® 9179	White mycelia, green spores/no spores

A satisfactory result is represented by visible growth.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

SABOURAUD DEXTROSE LIQUID MEDIUM (CM0147)

Tested in accordance with current USP/EP/BP/JP

Reactions after incubation at 30-35°C for 3-5 days

Medium is challenged with 10-100 colony-forming units

Candida albicans	ATCC® 10231	Turbid growth

Reactions after incubation at 20-25°C for 2-3 days

Medium is challenged with 10-100 colony-forming units

Candida albicans	ATCC® 10231	Turbid growth

Reactions after incubation at 20-25°C for 2-5 days

Medium is challenged with 10-100 colony-forming units

Aspergillus brasiliensis	ATCC® 16404	White mycelia, black spores/no spores

A satisfactory result is represented by visible growth.

The Microbiological Quality Control of this product complies with the following pharmacopoeia;

1. European Pharmacopoeia: Current version.
   2.6.12 Microbiological Examination of Non-Sterile Products: Harmonised Method: Microbial Enumeration tests
   2.6.13 Microbiological Examination of Non-Sterile Products: Tests for Specified Microorganisms. B. Harmonised Method
2. United States Pharmacopoeia: Current version.
   61 Microbiological Examination of Non-Sterile Products: Microbial Enumeration tests.
   62 Microbiological Examination of Non-Sterile Products: Tests for Specified Microorganisms
3. Japanese Pharmacopoeia: Current version.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

SABOURAUD DEXTROSE LIQUID MEDIUM (CM0147)

Revision History

Section / Step	Description of Change	Reason for Change	Reference
Entire Document	Reformatting to new template Addition of 'Tested in accordance with' statement	Change control	BT-CC-1477

Distribution: Central File

Date: 07/03/17
Supersedes: 05/10/16

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

BRILLIANT GREEN AGAR CM0263

Typical Formula*

Proteose peptone	grams per litre	10.0
Yeast extract		3.0
Lactose		10.0
Sucrose		10.0
Sodium chloride		5.0
Phenol red		0.08
Brilliant green		0.0125
Agar		12.0

* adjusted as required to meet performance standards

Directions

Suspend 50g in 1 litre of distilled water. Bring to the boil to dissolve completely. Sterilize by autoclaving at 121°C for 15 minutes. Cool to 50°C. Mix well and pour into sterile Petri dishes.

Physical Characteristics

Straw/green, free-flowing powder
Colour on reconstitution - green/brown or red/brown
Moisture level - less than 7%
pH 6.9 ± 0.2 at 25°C
Clarity - clear
Gel strength - firm, comparable to 12.0g/litre of agar

Microbiological Tests Using Optimum Inoculum Dilution

Control Medium: Tryptone Soya Agar

Reactions after incubation at 37°C for 18 hours

Medium is challenged with 10-100 colony-forming units

Salmonella enteritidis	ATCC® 13076	0.5-3mm red colonies and medium
Salmonella typhimurium	ATCC® 14028	0.5-3mm red colonies and medium
Salmonella virchow	NCTC 5742	1-2mm red colonies and medium

A satisfactory result is represented by recovery of positive strains equal to or greater than 70% of the control medium.

Reactions after incubation at 37°C for 18 hours

Medium is challenged with 10-100 colony-forming units

Pseudomonas aeruginosa	ATCC® 9027	No growth or 0.5-2.5mm red colonies and medium
Escherichia coli	ATCC® 25922	No growth or pinpoint-2mm yellow/green colonies
Escherichia coli	ATCC® 11775	No growth or pinpoint-2mm yellow/green colonies
Enterobacter cloacae	ATCC® 13047	No growth or 0.5-2mm yellow/green colonies
Proteus mirabilis	ATCC® 12453	No growth or pinpoint colourless colonies with no swarming or slight swarming

For negative strains, a satisfactory result is represented by recovery equal to or less than 100% of the control medium.

Equivalent results obtained after incubation at 30-35°C for 18-24 hours.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

BUFFERED PEPTONE WATER (CM0509)

BUFFERED PEPTONE WATER **CM0509**

Formula

Peptone	grams per litre	10.0
Sodium chloride		5.0
Di-sodium phosphate		3.5
Potassium dihydrogen phosphate		1.5

Directions

Add 20g to 1 litre of distilled water. Mix well and distribute into final containers. Sterilize by autoclaving at 121°C for 15 minutes.

Physical Characteristics

Straw, free flowing powder
Colour on reconstitution - straw 2-3
Moisture level - less than 7%
pH - 7.2 ± 0.2 at 25ºC
Clarity - clear (single and double strength broth)
Buffering Capacity Test - passes test

Microbiological Tests Using Optimum Inoculum Dilution

Control Medium: Tryptone Soya Agar

Reactions after incubation at 37 ± 2°C for 18 ± 2 hours

Medium is challenged with 10-100 colony forming units

Salmonella nottingham	NCTC 7832	Turbid growth
Escherichia coli	ATCC® 11775	Turbid growth

A satisfactory result is represented by visible growth.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

BUFFERED PEPTONE WATER (CM0509)

Testing performed in accordance with ISO11133:2014

Reactions after incubation at 37 ± 2°C for 18 ± 2 hours

Medium is challenged with 10-100 colony forming units

Salmonella typhimurium	ATCC® 14028	WDCM 00031	Turbid growth
Salmonella enteritidis	ATCC® 13076	WDCM 00030	Turbid growth
Escherichia coli	ATCC® 8739	WDCM 00012	Turbid growth
Escherichia coli	ATCC® 25922	WDCM 00013	Turbid growth

A satisfactory result is represented by visible growth from an inoculum of 10-100 colony forming units.

Testing performed in accordance with ISO22964:2017

Reactions after incubation at 36 ± 2°C for 18 ± 2 hours

Medium is challenged with 10-100 colony forming units

Cronobacter sakazakii	ATCC® 29544	WDCM 00214	Turbid growth
Cronobacter muytjensii	ATCC® 51329	WDCM 00213	Turbid growth

A satisfactory result is represented by visible growth from an inoculum of 10-100 colony forming units.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

BUFFERED PEPTONE WATER (CM0509)

Revision History

Section / Step	Description of Change	Reason for Change	Reference
Entire document	Update to new template and addition of ISO22964:2017 section	Change control	BT-CC-1531

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

E.C. BROTH CM0853

E.C. BROTH **CM0853**

Typical Formula*

Tryptone	grams per litre	20.0
Lactose		5.0
Bile Salts No. 3		1.5
Di-potassium phosphate		4.0
Mono-potassium phosphate		1.5
Sodium chloride		5.0

* adjusted as required to meet performance standards

Directions

Dissolve 37g in 1 litre of distilled water. Dispense into final containers and sterilize by autoclaving at 121°C for 15 minutes.

Physical Characteristics
White, free-flowing powder
Colour on reconstitution - straw 2-3
Moisture Level - less than or equal to 7%
pH - 6.9 ± 0.2 at 25°C
Clarity - clear

Microbiological Tests using Optimum Inoculum Dilution

Control Medium: Tryptone Soya Agar

Reactions after incubation at 44 ± 2°C for 24 ± 2 hours

Medium is challenged with 10-100 colony-forming units

Enterobacter aerogenes	ATCC®13048	Turbid growth, no gas

A satisfactory result is represented by visible growth and no gas.

Reactions after incubation at 44 ± 2°C for 48 ± 2 hours

Medium is challenged with 1E+04 to 1E+06 colony-forming units

Bacillus subtilis	ATCC®6633	No growth
Staphylococcus aureus	ATCC®25923	No growth

Negative strains shall be inhibited.

Testing performed in accordance with ISO11133:2014

Reactions after incubation at 44 ± 2°C for 24 ± 2 hours

Medium is challenged with 10-100 colony-forming units

Escherichia coli	ATCC®25922	WDCM00013	Turbid growth & gas
Escherichia coli	ATCC®8739	WDCM00012	Turbid growth & gas

A satisfactory result is represented by visible growth and gas.

Reactions after incubation at 44 ± 2°C for 48 ± 2 hours

Medium is challenged with 1E+04 to 1E+05 colony-forming units

Pseudomonas aeruginosa	ATCC®27853	WDCM00025	No growth

Negative strains shall be inhibited.

OXOID QUALITY ASSURANCE PRODUCT SPECIFICATION

E.C. BROTH CM0853

Revision History

Section / Step	Description of Change	Reason for Change	Reference
ISO11133 section	Update to test specification	Change control	BT-CC-1506
Entire Document	Update to new document format and correction of typographical/minor errors.	Change control	BT-CC-2263