

PRODUCT INFORMATION

Product Type: Bottle product (100ml)

Cat No. BP461/100S - SP-4 BROTH, 100 ML

Intended Use:

SP-4 Broth is a specialized liquid culture medium designed for the isolation, cultivation, and qualitative detection of Mycoplasma species, including Mycoplasma pneumoniae and other human and animal mycoplasmas, widely used in clinical, veterinary, and research laboratories.

Principles and uses:

Nutrient-rich: Contains beef heart infusion, peptone, CMRL 1066 medium, yeast extract, and fetal bovine serum, which together supply essential nutrients, growth factors, cholesterol, and proteins required for the fastidious growth of mycoplasmas.

Selective agents: Penicillin is included to inhibit the growth of most bacteria other than mycoplasmas.

pH indicator: Phenol red is added as a pH indicator. A color change (red to yellow) indicates acid production from glucose metabolism by fermentative mycoplasmas such as *M. pneumoniae*.

Variants: SP-4 Broth can be supplemented with glucose, arginine, or urea to support or differentiate specific mycoplasma or urea plasma species.

Typical Uses

Cultivation and detection of *Mycoplasma spp.*:

Used for primary isolation and qualitative detection from clinical specimens (e.g., respiratory secretions, blood, tissue, or CSF).

Antimicrobial susceptibility testing:

Recommended by CLSI for broth microdilution testing of mycoplasmas, such as *M. bovis* and *M. pneumoniae*.

Differentiation of species:

By supplementing with glucose, arginine, or urea, SP-4 Broth can help distinguish between fermenting and non-fermenting mycoplasma species.

Interpretation

Growth Indication:

A change in color from red to yellow (with glucose) indicates acid production and growth of fermentative mycoplasmas.

Colony morphology on agar:

When plated, mycoplasmas form tiny "fried-egg" or granular colonies.

Composition:

Tryptone Peptone (Bacto Tryptone is an animal origin (AO) pancreatic digest of casein) - 10 g/L
Heart Infusion - 3.14 g/L
Yeast Enriched Peptone - 5.23 g/L
Sodium Chloride - 2.61 g/L
Peptone (Enzymatic digest of animal origin) - 5.3 g/L
Penicillin G 10MU - 627 mg/L
CMRL* Medium, no glutamine - 50 ml/L
FBS South American - 170 ml/L
Dextrose - 5 g/L
Phenol Red - 20 mg/L
Yeast Dialyzate - 100 ml/L
Fresh Yeast Extract - 8.75 g/L

***CMRL Medium, no glutamine 50 ml/L (components final concentration mg/L in SP-4 BROTH:**

Amino Acids

Glycine - 2.5
Hydroxy L-proline - 0.5
L-Alanine - 1.25
L-Arginine hydrochloride - 3.5
L-Aspartic acid - 1.5
L-Cysteine - 10
L-Cystine - 1
L-Glutamic Acid - 3.75
L-Histidine hydrochloride-H₂O - 1
L-Isoleucine - 1
L-Leucine 3
L-Lysine hydrochloride - 3.5
L-Methionine - 0.75
L-Phenylalanine - 1.25
L-Proline - 2
L-Serine - 1.25
L-Threonine - 1.5
L-Tryptophan - 0.5
L-Tyrosine - 2
L-Valine - 1.25

Vitamins

Ascorbic Acid - 2.5
Biotin - 0.0005
Cholesterol - 0.01
Choline chloride - 0.025
D-Calcium pantothenate - 0.0005
Folic Acid - 0.0005
Niacinamide - 0.00125
Nicotinic acid (Niacin) - 0.00125
Para-Aminobenzoic Acid - 0.0025
Pyridoxal hydrochloride - 0.00125
Pyridoxine hydrochloride - 0.00125
Riboflavin - 0.0005
Thiamine hydrochloride - 0.0005
i-Inositol - 0.0025

Inorganic Salts

Calcium Chloride ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$) - 13.2
Magnesium Sulfate ($\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$) - 10
Potassium Chloride (KCl) - 20
Sodium Bicarbonate (NaHCO_3) - 110
Sodium Chloride (NaCl) - 340
Sodium Phosphate monobasic ($\text{NaH}_2\text{PO}_4 \cdot 2\text{H}_2\text{O}$) - 7.9

Other Components

2'-Deoxyadenosine - 0.5
2'-Deoxycytidine - 0.5
2'-Deoxyguanosine - 0.5
5-Methyl-deoxycytidine - 0.005
Co-carboxylase - 0.05
Coenzyme A - 0.125
D-Glucose (Dextrose) - 50
Diphosphopyridine nucleotide (NAD) - 0.35
FAD (flavin adenine dinucleotide) - 0.05
Glutathione (reduced) - 0.5
Phenol Red - 1
Sodium acetate- $3\text{H}_2\text{O}$ - 4.15
Sodium glucuronate- H_2O - 0.21
Thymidine - 0.5
Triphosphopyridine Nucleotide (NADP) - 0.05
Tween 80® - 0.25
Uridine 5'-triphosphate - 0.05

Storage: 2-8°C

pH at RT: 7.2 - 7.6

Appearance: Light red to orange-red, Clear and transparent

Exp. Date: Printed on label and on the item.

Required materials not supplied: Laboratory equipment as required.

Warning and Precautions:

Warning and Precautions - For professional use only. Follow good microbiological lab practices while handling specimens and culture. Do not use Bottles if they show evidence of microbial contamination, discoloration, drying, cracking, or other signs of deterioration. Avoid freezing and overheating. The Bottles may be used / inoculated up to the expiration date and incubated for the recommended incubation times. After use and prior to discarding, specimen containers and all contaminated material, including the used culture media and contaminated culture containers, must be sterilized or incinerated by validated procedures. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium.

Waste Disposal

After interpretation all items should be destroyed by standard incineration methods.

Performance Testing Results

GPT: Inoculum 10-100 cfu.

TEST	ATCC	Incubation Temp. (°C)	Incubation Cond.	Reaction 1
<i>Mycoplasma orale</i>	23714	35-37 °C	5-10% CO2 or 90-99% N2 + 5-10% CO2, up to 7 days	Growth
<i>Mycoplasma pneumoniae</i>	15293	35-37 °C	5-10% CO2 or 90-99% N2 + 5-10% CO2, up to 7 days	Growth
<i>Acholeplasma laidlawii</i>	23206	35-37 °C	5-10% CO2 or 90-99% N2 + 5-10% CO2, up to 7 days	Growth
<i>Mycoplasma hyorhinis</i>	23839	35-37 °C	5-10% CO2 or 90-99% N2 + 5-10% CO2, up to 7 days	Growth
<i>Mycoplasma gallisepticum</i>	19610	35-37 °C	5-10% CO2 or 90-99% N2 + 5-10% CO2, up to 7 days	Growth
<i>Mycoplasma synoviae</i>	25204	35-37 °C	5-10% CO2 or 90-99% N2 + 5-10% CO2, up to 7 days	Growth

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Hy Laboratories Ltd.

6 Menachem Plaut St., Park Tamar, Rehovot 7670606, Israel
Tel. +972.8.9366475 Email. hylabs@hylabs.co.il

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