

PSEUDOMONAS CEPACIA AGAR (7458)

Intended Use

Pseudomonas Cepacia Agar is used for the selective isolation and detection of *Pseudomonas cepacia* (*Burkholderia cepacia*) from clinical and non-clinical specimens.

Product Summary and Explanation

Pseudomonas cepacia was first described as a phytopathogen, and isolated from rotting onions and various soil and animal sources.¹ In recent years *Pseudomonas cepacia* has become an opportunist, causing nosocomial infections including endocarditis, pneumonia, wound, and urinary tract infections.² This organism has been isolated from disinfectants, nebulizers, incubators, and intravenous fluids in hospitals worldwide.¹ *P. cepacia* causes chronic lung infections in cystic fibrosis patients, and can lead to life-threatening systemic complications.³ *P. cepacia* is highly resistant to aminoglycosides and other antibiotics, making it difficult to treat.²

In 1985 Gilligan et al.³ developed Pseudomonas Cepacia Agar. This medium has shown good recovery of *P. cepacia*, even among mixed populations. Some *P. cepacia* strains produce a nonfluorescent phenazine pigment, while others nonpigmented.¹ A sweet odor similar to that associated with *P. aeruginosa* is produced by some strains.¹

Principles of the Procedure

Enzymatic Digest of Animal Tissue provides nitrogen, vitamins, and carbon in Pseudomonas Cepacia Agar. Bile Extract and Crystal Violet are used to inhibit Gram-positive organisms, and Polymyxin B and Ticarcillin are used to inhibit Gram-negative organisms. Magnesium Sulfate, Ammonium Sulfate, and Ferrous Ammonium Sulfate are used to promote the growth of *Pseudomonas cepacia*. Dipotassium Phosphate and Monopotassium Phosphate are the buffering agents in this formulation. Sodium Pyruvate is utilized by *P. cepacia*, producing alkaline by-products and causing the medium to turn pink to hot-pink in color. Phenol Red is the indicator. Agar is the solidifying agent.

Formula / Liter

Enzymatic Digest of Animal Tissue.....	1 g
Sodium Pyruvate.....	5 g
Bile Extract.....	1.5 g
Ammonium Sulfate.....	1 g
Ferrous Ammonium Sulfate.....	0.01 g
Magnesium Sulfate.....	0.2 g
Dipotassium Phosphate.....	4.3 g
Monopotassium Phosphate.....	2.1 g
Phenol Red.....	0.02 g
Crystal Violet.....	0.001 g
Agar.....	15 g

Final pH: 7.1 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Supplement (10mL) / Liter

Ticarcillin (100 mg)
Polymyxin B (300,000 units)

Precautions

1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

1. Suspend 30 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.
4. Cool to 45 - 50°C and aseptically add a filter sterilized solution containing Ticarcillin (100 mg) and Polymyxin B (300,000 units) dissolved in 10 mL of sterile water.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and light beige.

Prepared Appearance: Prepared medium is trace to slight hazy and peach.

Expected Cultural Response: Cultural response on Pseudomonas Cepacia Agar at 35°C after 18 - 48 hours incubation.

Microorganism	Response	Reactions
<i>Burkholderia cepacia</i> ATCC® 25416	fair to good growth	lime green colonies with a pink to hot-pink color change in the medium
<i>Escherichia coli</i> ATCC® 25922	inhibited	---
<i>Pseudomonas aeruginosa</i> ATCC® 27853	inhibited	---
<i>Staphylococcus aureus</i> ATCC® 25923	inhibited	---

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

1. Inoculate medium using the streak plate method to obtain isolated colonies.
2. Incubate for 18 – 72 hours at 35°C.

Results

Examine for presence of growth. *Pseudomonas cepacia* colonies will be lime green, and the medium surrounding the colonies will be pink to hot-pink in color.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.
2. Colonies growing on Pseudomonas Cepacia Agar require further identification through biochemical testing. Other microorganisms can utilize Sodium Pyruvate, produce alkaline by-products, and cause the medium to turn pink to hot-pink.

Packaging

Pseudomonas Cepacia Agar	Code No.	7458A	500 g
		7458B	2 kg
		7458C	10 kg

References

1. **Gilardi, G. L.** 1991. In A. Balows, W. J. Hausler, Jr., K. L. Herrmann, H. D. Isenberg, and H. J. Shadomy (eds.) Manual of clinical microbiology, 5th ed. American Society of Microbiology, Washington, D.C.
2. **Iglewski, B. H.** 2001. *Pseudomonas*. In S. Baron (ed). Medical Microbiology. University of Texas Medical Branch, Galveston, TX.
3. **Chaparro, C., J. Maurer, C. Gutierrez, M. Krajden, C. Chan, T. Winton, S. Keshavjee, M. Scavuzzo, E. Tullis, M. Hutcheon, and S. Kesten.** 2001. Infection with *Burkholderia cepacia* in Cystic Fibrosis. Am. J. Respir. Crit. Care Med. **163**:43-48.
4. **Gilligan, P. H., et al.** 1985. In P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.). Manual of clinical microbiology, 4th ed. American Society of Microbiology, Washington, D.C.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.