



Pseudomonas Selective Agar

Medium for detection and enumeration
of Pseudomonas in water and food

Granulated Culture Media – Safer from Merck.



Pseudomonas Selective Agar Base

Medium for the detection and enumeration of Pseudomonas. Cat. No. 1.07620.0500 (500 g)

Pseudomonas CFC Selective Agar

When supplemented with Pseudomonas CFC Selective Supplement (1.07627.0001) the medium complies with the recommendations of ISO 13720 for the detection and enumeration of Pseudomonas spp. in foods and animal feed.

Pseudomonas CN Selective Agar

When supplemented with Pseudomonas CN Selective Supplement (1.07624.0001) the medium complies with the recommendations of DIN/EN 12780 for the detection and enumeration of Pseudomonas aeruginosa in water using the membrane filtration technique.

Mode of action

The peptone mixture in Pseudomonas Selective Agar Base allows growth of a broad spectrum of Pseudomonades.

The amount of potassium sulfate and magnesium chloride supports forming of pigments.

By use of the appropriate selective supplement and the incubation temperature the medium becomes selective for Pseudomonas spp. including Burkholderia cepacia, formerly known as Pseudomonas cepacia (CFC Agar), or Pseudomonas aeruginosa (CN Agar).

Typical composition (g/Liter)

Peptone from gelatine 16.0; Casein hydrolysate 10.0; potassium sulfate 10.0; magnesium chloride 1.4; agar-agar 11.0.

Preparation

Suspend 24.2 g in 500 ml of purified water, add 5 ml glycerol and heat to boiling until dissolved completely.

Autoclave (15 min. at 121 °C)

Cool the medium to 45 - 50°C and aseptically add the contents of one vial of Pseudomonas CFC Selective Supplement (1.07627.0001) or Pseudomonas CN Selective Supplement (1.07624.0001). Mix thoroughly and pour plates.

pH: 7.1 ± 0.2 at 25 °C

The prepared plates are clear and colourless and can be stored for up to 4 weeks at 2 - 8°C in the refrigerator.

Protect from light and drying. Do not keep the liquid medium (45 - 50°C) longer than 4 hours. Do not remelt the medium several times.

Experimental procedure and evaluation

Pseudomonas CFC Selective Agar

Inoculate the medium using the surface spread method.

Incubation: 44 ± 4 hours at 25 ± 1 °C

All grown colonies are suspect Pseudomonas spp. and counted as such. The suspect colonies must be confirmed. Colonies which show a positive oxidase reaction but no glucose fermentation are confirmed Pseudomonas spp. colonies.

Pseudomonas CN Selective Agar

Inoculate the medium using the membrane filtration technique. The filter material impacts results. Good results were achieved using Cellulose-Mixed Ester membranes (e.g. Pall GN-6).

Incubation: 44 ± 4 hours at 36 ± 2 °C

Check the membrane filters for growth after 22 ± 2 h and 44 ± 4 h.

All grown colonies with a blue-green pigmentation are considered confirmed Pseudomonas aeruginosa colonies and counted as such.

Check the membrane filters under UV-light. All colonies not showing the blue-green pigmentation but fluoresce are suspect P. aeruginosa colonies and confirmed by use of acetamide solution. All other reddish-brown pigmenting colonies, which do not fluoresce are considered suspect P. aeruginosa colonies and confirmed by the oxidase test (1.13300.0001), acetamide solution and King's B Medium.

Pseudomonas, a threatening danger

Pseudomonas are Gram-negative bacteria widely distributed in soil, water, milk and animal food. They require oxygen for growth and they are clinically relevant because the majority of the species is resistant to antibiotics. Although some species only occur in Asia, Africa and the Middle East, they nevertheless find their way to all parts of the world by means of the food chain. And, most importantly, 80 % of the infections caused by the most dangerous pseudomona (*P.aeruginosa*) end fatally.

Precisely because they occur in such inconspicuous surroundings – like water, milk or animal food – it is very important to take adequate precautions. Merck offers you culture media for the detection of pseudomonas in food and water. So you won't be taking risks in the future.



Literature

MOSSEL, D.A.A., VISSER, M. and MENGERINK, W.H.J.:
A comparison of media for the enumeration of moulds and yeasts in foods and beverages.

• *Lab. Pract.* 11: 109-112 (1962).

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Nalidixic Acid Cetrimide Agar. A New Selective Plating Medium for the Selective Isolation of Pseudomonas aeruginosa.

• *Japan. J. Microbiol.* 14: 65 – 72.

MEAD, G.C. and B.W. ADAMS. 1977.

A selective medium for the rapid isolation of Pseudomonas associated with poultry meat spoilage.

• *Br. Poult. Sci.* 18: 661 – 670.

ISO INTERNATIONAL STANDARDISATION ORGANISATION.

Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of Pseudomonas spp.

• ISO/WD 13720:2000.

EN EUROPEAN STANDARD.

Water Quality – Detection and enumeration of Pseudomonas aeruginosa by membrane filtration.

• DIN/EN 12780:2002.

Pseudomonas CFC Selective Supplement

Cat. No. 1.07627.0001 (Pack contains 16 vials)

Additive for the preparation of Pseudomonas CFC Selective Agar for the detection and enumeration of Pseudomonas spp. from food and animal feed

Mode of action

The selective supplement is a mixture of 3 different inhibitors in lyophilized form.

Cetrimide, Fucidin and Cephalotin inhibit the Gram-positive and Gram-negative accompanying flora.

Composition (per vial)

Cetrimide 5 mg; Fucidin 5 mg; Cephalotin 25 mg

Preparation

Aseptically add 2 ml of a 50/50 mixture of purified water and ethanol to the contents of one vial and mix gently to avoid foaming.


Aseptically add the contents of one vial to 500 ml of Pseudomonas Selective Agar Base (with 5 ml glycerol) cooled to 45 - 50°C. Mix to suspend evenly.

Quality control


Pseudomonas CFC Selective Agar

44 h ± 4 h at 25 °C ± 1 °C


Test strains		Recovery rate
<i>Pseudomonas aeruginosa</i>	ATCC 27853	> 70%
<i>Pseudomonas putida</i>	ATCC 12633	> 70%
<i>Pseudomonas fluorescens</i>	ATCC 13525	> 70%
<i>Pseudomonas fragi</i>	ATCC 27362	> 70%
<i>Burkholderia cepacia</i>	ATCC 17759	> 70%
<i>Proteus mirabilis</i>	ATCC 14153	< 0.01%
<i>Staphylococcus aureus</i>	ATCC 25923	< 0.01%



Pseudomonas aeruginosa
ATCC 27853



Pseudomonas fluorescens
ATCC 13525



Pseudomonas putida
ATCC 12633

Pseudomonas CN Selective Supplement

Cat. No. 1.07624.0001 (Pack contains 16 vials)

Additive for the preparation of Pseudomonas CN Selective Agar for the detection and enumeration of *Pseudomonas aeruginosa* in water by the membrane filter technique

Mode of action

The selective supplement is a mixture of 2 different inhibitors in lyophilized form. Cefrimide and Nalidixic acid inhibit the Gram-positive and Gram-negative accompanying flora.

Composition (per vial)

Cefrimide 100 mg; Nalidixic acid 7.5 mg

Preparation

Aseptically add 2 ml of a 50/50 mixture of purified water and ethanol to the contents of one vial and mix gently to avoid foaming.

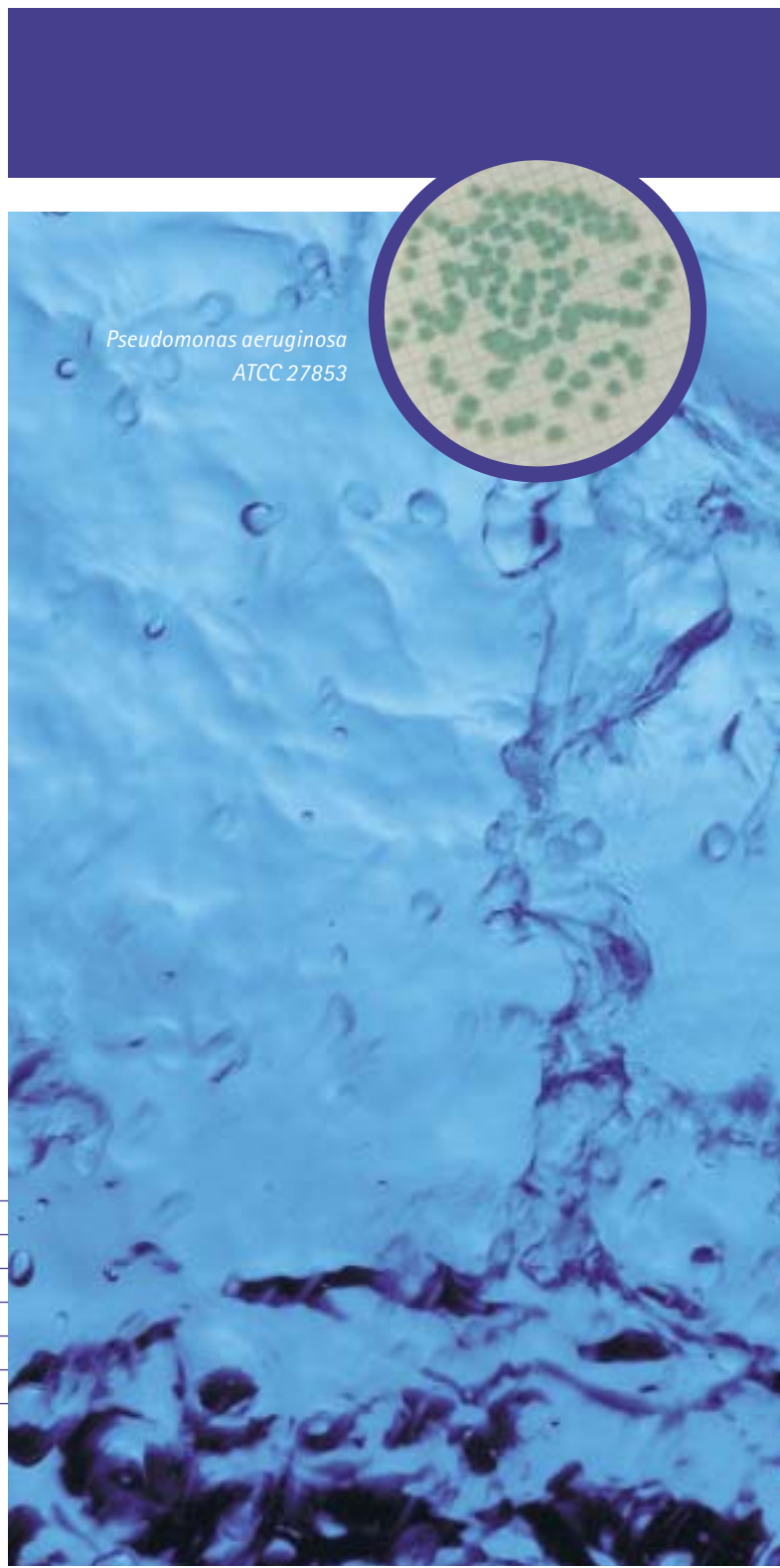
Aseptically add the contents of one vial to 500 ml of Pseudomonas Selective Agar Base (with 5 ml glycerol) cooled to 45 - 50°C. Mix to suspend evenly.

Quality control

Pseudomonas CN Selective Agar

44 h ± 4 h at 36°C ± 2°C

Test strains		Recovery rate
<i>Pseudomonas aeruginosa</i>	ATCC 27853	> 70%
<i>Pseudomonas fluorescens</i>	ATCC 13525	< 0.01%
<i>Aeromonas hydrophila</i>	ATCC 7966	< 0.01%
<i>Klebsiella pneumoniae</i>	ATCC 13883	< 0.01%
<i>Proteus mirabilis</i>	ATCC 14153	< 0.01%
<i>Providencia rustigianii</i>	ATCC 13159	< 0.01%





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Merck KGaA
64271 Darmstadt, Germany
Fax: +49 (0) 61 51/72 60 80
E-mail: mibio@merck.de
Internet: microbiology.merck.de

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