Pseudomonas Selective Agar Base

Medium for the detection and enumeration of *Pseudomonas*.

Pseudomonas CFC Selective Agar

When supplemented with Pseudomonas CFC Selective Supplement (Cat.No.1.07627.) 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 (Cat.No. 1.07624) 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 for 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 (Cat.No.1.07627) or Pseudomonas CN Selective Supplement (Cat.No.1.07624). Mix thoroughly and pour plates.

pH: 7.1 ± 0.2 at 25 °C.

The prepared plates are clear and colorless 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, acetamide solution and King's B Medium.

Literature

Goto, S., and S. Enomoto. 1970. 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**.

Ordering Information

Product	Merck Cat. No.	Pack size
Pseudomonas Selective Agar Base	1.07620.0500	500 g
Pseudomonas CFC Selective Supplement	1.07627.0001	1 x 16 vials
Pseudomonas CN Selective Supplement	1.07624.0001	1 x 16 vials

Pseudomonas Selective Agar Base

Quality Control

Pseudomonas CFC Selective Agar 44 h ± 4 h at 25 ± 1°C

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

Pseudomonas CN Selective Agar 44 h ± 4 h at 36 ± 2°C

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

Pseudomonas aeruginosa ATCC 27853 C-N-Supplement

Pseudomonas aeruginosa ATCC 27853 C-F-C-Supplement