Reference: DSHB3092 **A.B.E. - Technical Data Sheet**

Product: **Selective Supplement for CN**

Specification

Sterile selective supplement for the isolation of *Pseudomonas spp.* according to ISO.

Presentation

	Packaging Details	Shelf Life	Storage
10 Freeze dried vials 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)

Cetrimide..... 0.1000
Nalidixic acid, sodium salt..... 0.0075

Reconstitute the original freeze-dried vial by adding :

Sterile distilled water.....6 ml

Note: Each vial is sufficient to supplement 500 ml of Pseudomonas Agar Base (ISO).

Description /Technique

Description:
CN selective supplement is added to Pseudomonas Agar Base in order to obtain a complete medium suitable for the isolation of *Pseudomonas spp.*
Pseudomonas CN Agar is a selective medium recommended by ISO for the enumeration of *Pseudomonas spp* in water.
Gelatin peptone and enzymatic digest of casein provide nitrogen, vitamins, minerals and amino acids essential for growth and allows the growth of a great number of *Pseudomonas spp*. The potassium sulfate and magnesium chloride help the formation of pigmentation (pyocyanin). The addition of cetrimide, make the medium more selective for *Pseudomonas spp.*
Cetrimide, inhibits the Gram-positive bacteria and supports the growth of *Pseudomonas spp,* (including *P. aeruginosa*), with nalidixic acid inhibiting most other Gram negative bacteria.

Technique:
Reconstitute the vial with 6 ml sterile diluent in aseptic conditions and add it to 500 ml of Pseudomonas Agar Base (ISO) cooled to 50 ºC. Pour into MF plates.
Do not overheat once supplemented.
Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.
Incubate the plates right side up aerobically at 25 - 30 ºC for 48 h.
(Incubation times longer than those mentioned above or different incubation temperatures may be required depending on the sample, on the specifications,..)
After incubation, enumerate all the colonies that have appeared onto the surface of the agar.
Each laboratory must evaluate the results according to their specifications.
Presumptive isolation of *Pseudomonas spp.* must be confirmed by further microbiological or biochemical tests.
Colonies which show a positive oxidase reaction are *Pseudomonas spp.*

Quality control

Physical/Chemical control

Color : White

Microbiological control

Add 1 vial to 500 ml of medium base. DO NOT HEAT once supplemented.

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

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Incubate according instructions for complete medium indicated in COMPOSITION.

Microbiological control according to ISO 11133:2014/A1:2018.

Microorganism	Growth
Ps. aeruginosa ATCC® 9027, WDCM 00026	Good
Escherichia coli ATCC® 8739, WDCM 00012	Inhibited

Sterility control

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

Bibliography

· BROWN, V.L. & E.J.L. LOWBURY (1965) Use of an improved Cetrimide Agar Medium and of culture methods for *P. aeruginosa*. J., Clin. Pathol. 18:752.
· EN 12780 Standard (2002) Water Quality. Detection and enumeration of *P. aeruginosa* by membrane filtration.
· GOTO S. & S. ENOMOTO (1970) Nalidixic acid cetrimide agar. A new selective plating medium for the selective isolation of *P. aeruginosa*. Jpn. J. Microbiol. 14:65.
· ISO 16266 Standard (2006) Water Quality. - Detection and enumeration of *Pseudomonas aeruginosa*. - Method by membrane filtration.
· KING, E.O., M.K. WARD & E.E. RANEY (1954) Two simple media for the demonstration of pyocianin and fluorescein. J. Lab. Clin. Med. 44:301.
· ROBIN, T. & J.M. JANDA (1984) Enhanced recovery of *P. aeruginosa* from diverse clinical specimens on a new selective agar. Diag. Microbiol. Infect Dis. 2:207.
· SCHWEIZERISCHE LEBENMITTELSBUCH (2005) Kap. 56 Mikrobiologie. Bundesamt für Gesundheit. Direktionsbereich Verbraucherschutz. Bern.10/01/202010/01/202010/01/202010/01/2020