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INTENDED USE

Difco™ Yersinia Antimicrobial Supplement CN is used in the preparation of Yersinia Selective Agar (CIN Agar). The complete medium, based on the Cefsulodin-Irgasan™-Novobiocin (CIN) Agar formulation of Schiemann, is recommended for use in the selective isolation and cultivation of *Yersinia enterocolitica* from clinical and nonclinical sources.

SUMMARY AND EXPLANATION

CIN Agar, also known as Yersinia Selective Agar, was first described by Schiemann as an alternative to MacConkey Agar and other commonly used media for isolation of *Y. enterocolitica*, a causative agent of gastroenteritis.¹ CIN Agar has been found to be far superior to MacConkey, SS, CAL or Y agars for the recovery of *Y. enterocolitica*.²

PRINCIPLES OF THE PROCEDURE

Fermentation of mannitol in the presence of neutral red results in a characteristic "bull's-eye" colony, colorless with red center. Selective inhibition of gram-negative and gram-positive organisms is obtained by means of crystal violet, sodium desoxycholate and Irgasan (triclosan). Supplementation with Yersinia Antimicrobial Supplement CN (cefsulodin and novobiocin) improves inhibition of normal enteric organisms. Organisms that do not metabolize mannitol to acid end products will form colorless, translucent colonies.

REAGENTS

Difco Yersinia Antimicrobial Supplement CN

Approximate Formula* per 10 mL vial

Cefsulodin 4.0 mg

Novobiocin 2.5 mg

*Adjusted and/or supplemented as required to meet performance criteria.

Warnings and Precautions:

For Laboratory Use

Use aseptic technique in rehydrating Yersinia Antimicrobial Supplement CN and adding the supplement to the base. Follow established laboratory procedures in handling and disposing of infectious materials.

Danger



H315 Causes skin irritation. **H317** May cause an allergic skin reaction. **H319** Causes serious eye irritation. **H334** May cause allergy or asthma symptoms or breathing difficulties if inhaled. **H335** May cause respiratory irritation.

P264 Wash thoroughly after handling. **P280** Wear protective gloves/protective clothing/eye protection/face protection.

P305+P351+P338 "IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing." **P312** Call a POISON CENTER or doctor/physician if you feel unwell. **P403+P233** Store in a well-ventilated place. Keep container tightly closed. **P501** Dispose of contents/container in accordance with local/regional/national/international regulations.

Storage Instructions:

Store at 2–8 °C. Use immediately after rehydration or within 24 h. Store rehydrated vials at 2–8 °C. Do not open or rehydrate vials until ready to use.

Product Deterioration

Do not use the rehydrated supplement if it is contaminated, partially or completely evaporated, or shows other signs of deterioration.

PROCEDURE

Materials Provided: Difco Yersinia Antimicrobial Supplement CN.

Materials Required But Not Provided: Ancillary culture media, reagents, quality control organisms and laboratory equipment as required for this procedure.

Rehydration:

1. Aseptically add 10 mL of purified water to one vial of **Difco** Yersinia Antimicrobial Supplement CN.
2. Invert the vial several times to dissolve the powder.
3. For the preparation of CIN Agar, aseptically add 10 mL of rehydrated Yersinia Antimicrobial Supplement CN to the molten base cooled to 45–50 °C. Mix well.
4. Test samples of the finished product for performance using stable, typical control cultures.

Inoculation and Incubation:

Use standard procedures to obtain isolated colonies from specimens. Incubate plates at 25° C for 24–48 h.

If a cold enrichment procedure is desired, inoculate the specimen into phosphate buffered saline and hold at 4 °C for up to 21 days.^{3,4} Periodically subculture onto plates of CIN Agar, streaking for isolation. Incubate plates as stated above.

RESULTS

Typical *Y. enterocolitica* colonies will have deep-red centers surrounded by a transparent border giving the appearance of a “bull’s eye.”

Growth of non-*Yersinia* organisms is markedly to completely inhibited.

USER QUALITY CONTROL

1. Examine the lyophilized and rehydrated antimicrobial supplement for evidence of deterioration as described under “Product Deterioration.”
2. Check the performance of the base and antimicrobial supplement by testing in the complete medium. Prepare the medium per label directions. Inoculate and incubate at 25 ± 2 °C for 18–24 and 42–48 h.

ORGANISM	ATCC®	INOCULUM CFU	RECOVERY	COLONY COLOR
<i>Escherichia coli</i>	25922	10 ⁴ –10 ⁵	Partial to complete inhibition	—
<i>Yersinia enterocolitica</i>	9610	10 ⁷	Good	“Bull’s eye” reaction (translucent colonies with dark pink centers)

LIMITATIONS OF THE PROCEDURE

Although certain strains of *Yersinia* can be recovered by direct plating, others may require cold enrichment (4° C) in phosphate-buffered saline.³ However, cold enrichment may not be practical because of the long incubation time and because it selects for nonpathogenic *Yersinia* species.⁴

AVAILABILITY

Cat. No. Description

231961 **BD Difco™** Yersinia Antimicrobial Supplement CN, 6 x 10 mL.

REFERENCES

1. Schiemann, D.A. 1979. Synthesis of a selective agar medium for *Yersinia enterocolitica*. Can. J. Microbiol. 25:1298-1304.
2. Head, C.B., D.A. Whitty, and S. Ratnam. 1982. Comparative study of selective media for recovery of *Yersinia enterocolitica*. J. Clin. Microbiol. 16:615-621.
3. Weissfeld, A.S. and A.C. Sonnenwirth. 1982. Rapid isolation of *Yersinia* spp. from feces. J. Clin. Microbiol. 15:508-510.
4. Brockmühl, J., and J.D. Wong, 2003. *Yersinia*, p. 672-683. In P.R. Murray, E.J. Baron, J.H. Jorgensen, M.A. Pfaller, and R. H. Tenenbaum (ed.), Manual of clinical microbiology, 8th ed., vol. 1. American Society for Microbiology, Washington, D.C.

Technical Information: In the United States, contact BD Technical Service and Support at 800-638-8663 or www.bd.com/ds.



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AAAA-MM-DD / AAAA-MM (MM = slutning af måned)
JJJJ-MM-TT / JJJJ-MM (MM = Monatsende)
EEEE-MM-HH / EEEE-MM (MM = τέλος του μήνα)
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AAAA-KK-PP / AAAA-KK (KK = kuu lõpp)
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ÉÉÉÉ-HH-NN / ÉÉÉÉ-HH (HH = hónap utolsó napja)
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YYYY-MM-DD/YYYY-MM(MM = 월말)
MMMM-MM-DD / MMMM-MM (MM = mēnesio pabaiga)
GGGG-MM-DD/GGGG-MM (MM = mėneša beigas)
JJJJ-MM-DD / JJJJ-MM (MM = einde maand)
AAAA-MM-DD / AAAA-MM (MM = sluten av månaden)
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GGGG-MM-DD / GGGG-MM (MM = kraj meseca)
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