Yersinia Selective Agar Base (CIN) ISO

Cat. 1126

Selective medium for Yersinia enterocolitica used with supplement

Practical information

Aplications	Categories
Selective isolation	Yersinia enterocolitica
Detection	Yersinia enterocolitica

Industry: Clinical / Food

Regulations: ISO 10273 / ISO 11133

CE IVD

Principles and uses

Yersinia Select Agar Base (ISO 10273) is a selective and differential medium when used with supplements. The formula is based on the CIN Agar described by Schiemann, and is recommended by ISO 10273 for the isolation and detection of presumptive pathogenic Yersinia enterocolitica from a variety of clinical and food samples.

Antibiotics are added as a supplement in order to inhibit the accompanying flora. The growth of Yersinia is promoted by pyruvate as well as by the nutrients content in the base. Yersinia degrades the mannitol of the medium to an acid form; the colonies are turning to red color due to the neutral red indicator. Mannitol fermentation in the presence of neutral red produces a characteristic “bull’s-eye” colony, colorless with a red center.

Mannitol is the fermentable carbohydrate, source of carbon and energy. Enzymatic digest of gelatin and the enzymatic digest of casein and animal tissues provide nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group. Sodium pyruvate is added as a source of energy and as a protective substance in order to overcome oxygen toxicity biologically produced by the organisms. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Magnesium sulphate is an ion required in a large variation of enzymatic reactions, including DNA replication. Neutral red is the pH indicator. Selective inhibition of Gram-negative and Gram-positive organisms is obtained through crystal violet, sodium desoxycholate and Irgasan (triclosan). Cefsulodin and novobiocin improve the inhibition of normal enteric organisms.

Formula in g/L

Bacteriological agar	12,5	Crystal violet	0,001
Magnesium sulfate heptahydrated	0,01	Mannitol	20
Neutral red	0,03	Sodium chloride	1
Sodium deoxycholate	0,5	Sodium pyruvate	2
Yeast extract	2	Enzymatic digest of gelatin	17
Enzymatic digest of animal tissues and casein	3		

Preparation

Suspend 29 grams of the medium in 500 ml of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121 ºC for 15 minutes. Cool to 45 ºC and aseptically add one vial of Yersinia Selective Supplement (Cat. 6033), previously reconstituted in 5 ml of sterile distilled water. Homogenize gently and dispense into Petri dishes.

Instructions for use

» For clinical diagnosis, the type of sample is fecal samples and human rectal swabs.

- Inoculate on the surface making parallel striae with the handle or swab.
- Incubate at 30 ºC for 24-48 hours.
- Reading and interpretation of the results.

» For other uses not covered by the CE marking:

Detection of Yersinia enterocolitica according to ISO 10273:

- Obtain the initial suspension. Add a test portion of 25 g or 25 ml to 225 g or 225 ml of Sorbitol Peptone Broth and Bile Salt (PSB) (Cat. 1298) to obtain a tenfold dilution, and homogenize.
- Inoculate the suspension (direct plating) onto 2 to 4 selective agar plates of Yersinia Selective Agar (CIN).
- Transfer the initial suspension in the liquid enrichment medium PSB to the selective enrichment medium ITC (Irgasan Ticarcillin and Potassium Chlorate Broth) (Cat. 1361), and incubate the two enrichment liquid media at a temperature of 25 ºC for 44 ºC.
- Plate out the enrichment with KOH treatment (mixing 0,5 ml of enrichment in 4,5 ml of 0,5 % KOH solution for 20 s) on plates of Yersinia Selective Agar (CIN).
- Incubate the plates of Yersinia Selctive Agar (CIN) at 30 ºC for 24 hours.
- Verify the colony morphology as presumptive pathogenic Y. enterocolitica by successive culturing on selective plates.Typical colonies of Y. enterocolitica, will appear colorless, with dark red centers, like bull's eye, surrounded by a transparent border.
- Confirm the presence of pathogenic Y. enterocolitica species by biochemical or molecular confirmation test.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Beige	Purple red	7,4±0,2

Microbiological test

According to ISO 11133:
Incubation conditions: (30±1 ºC / 21±3 h).
Inoculation conditions: Productivity qualitative (<100 CFU) / Selectivity (10^4-10^6).

Microrganisms	Specification	Characteristic reaction
Yersinia enterocolitica ATCC 23715	Good growth (2)	Colonies with dark red centers, surrounded by a transparent border.
Escherichia coli ATCC 25922	Total or partial inhibition (0-1)	
Staphylococcus aureus ATCC 25923	Total inhibition (0)	
Yersinia enterocolitica CECT 9144	Good growth (2)	Colonies with dark red centers, surrounded by a transparent border.

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

American Public Health Association: Compendium of Methods for the microbiological Examination of Foods.
Schiemann, D:A: Synthesis of a selective agar medium for Yersinia enterocolitica.- Canad.J.Microbiol,25 1298-1304
ISO 10273 Microbiology of food and animal stuffs. Horizontal method for the detection of presumptive pathogenic Yersinia enterocolitica