

Irgasan Ticarcillin and Potassium Chlorate Broth (ITC) ISO

Cat. 1361

 For the selective enrichment of *Yersinia enterocolitica*

Practical information

Applications	Categories
Selective enrichment	<i>Yersinia enterocolitica</i>

Industry: Food

Regulations: ISO 10273 / ISO 11133

Principles and uses

Irgasan Ticarcillin and Potassium Chlorate Broth (ITC) is recommended by ISO 10273 as a selective enrichment broth for the detection of the human pathogenic strain of *Yersinia enterocolitica* in food and water samples.

Enzymatic casein digest provides nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group essential for bacterial growth. Magnesium chloride and malachite green, make the broth highly selective. Irgasan inhibits Gram-positive bacteria, Ticarcillin has bactericide on Gram-negative and Gram-positive bacteria and potassium chlorate has a disinfecting property.

Formula in g/L

Enzymatic digest of casein	10	Magnesium chloride anhydrous	28,1
Malaquite green	0,01	Sodium chloride	5
Yeast extract	1		

Preparation

Suspend 44,0 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121 °C for 15 minutes. Cool to 47 °C and aseptically add two vials of ITC Supplement (Cat. 6051), previously reconstituted in 8 ml of sterile distilled water each. Homogenize gently and dispense into sterile containers.

Instructions for use

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) (Cat. 1126).
- Transfer the initial suspension in the liquid enrichment medium PSB to the selective enrichment medium Irgasan Ticarcillin and Potassium Chlorate Broth (ITC) (Cat. 1361), and incubate the two enrichment liquid media at a temperature of 25 °C for 44 °C.
- 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 Selective Agar* (CIN) at 30 °C 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	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Light beige	Blue green	6,9±0,2

Microbiological test

According to ISO 10273:

Incubation conditions: (25±1 °C /44±4 h).

Inoculation conditions: Target microorganisms (<100 CFU) / Non-target microorganism (>1000 CFU) / Selectivity (10⁴-10⁶ CFU).

Microrganisms	Specification	Characteristic reaction
Yersinia enterocolitica ATCC 23715 + Escherichia coli ATCC 8739 + Pseudomonas aeruginosa ATCC 27853	>10 characteristic colonies on CIN Agar	Characteristic colonies according to each medium
Proteus mirabilis ATCC 29906	Total inhibition (0) or partial inhibition (<10 colonies) on TSA	
Yersinia enterocolitica CECT 9144 + Escherichia coli ATCC 8739 + Pseudomonas aeruginosa ATCC 27853	>10 characteristic colonies on CIN Agar	Characteristic colonies according to each medium

Storage

Temp. Min.:2 °C

Temp. Max.:25 °C

Bibliography

ISO 10273 Microbiology of food and animal feeding stuffs - Horizontal method for the detection of presumptive pathogenic Yersinia enterocolitica