

Specification

A selective supplement for pre-enrichment of *Campylobacter* species in food samples.

Presentation

	Packaging Details	Shelf Life	Storage
10 Freeze dried vials Vial with: 3 ± 0.1 g	23x60 mm glass vials, tag labelled, White plastic cap - 10 vials per box.	49 months	2-25 °C

Composition

Compositon (g/vial)	NOTE : Each vial is sufficient to supplemented 500 ml of Bolton Selective Enrichment Broth.
Vancomycin..... 0.010	
Trimethoprim..... 0.010	
Cefoperazone..... 0.010	
Cycloheximide..... 0.025	

Reconstitute the original freeze-dried vial
by adding :
Sterile Distilled Water/ Ethanol(50:50)..... 6 ml

Description /Technique

Description:

Bolton Selective Enrichment Broth is intended for the pre-enrichment of *Campylobacter* in food samples. *Campylobacter* are Gram-negative, spirally shaped microaerophilic organisms which may be present in raw milk, untreated water, improperly handled food and undercooked meats, poultry and shellfish.

Bolton Selective Enrichment Broth contains nutrients to aid resuscitation of sublethally injured cells, and the inclusion of sodium metabisulphite and sodium pyruvate quenches toxic compounds that may form in the culture medium. These additions also increase the aero-tolerance of the culture.

The antibiotics contained in Bolton Broth Selective Supplement optimise the selectivity for *Campylobacter spp*:

Vancomycin : Inhibits Gram+

Cefoperazone: Inhibits Gram-

Trimethoprim: Inhibits a wide variety of Gram- and Gram+

Cycloheximide: Inhibits pathogenic fungi (yeast and mold)

Note: ISO recommends amphotericin B, but has been replaced by cycloheximide for greater availability, solubility and efficacy.

Technique:

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.

Reconstitute the vial with 6 ml of the sterile diluent in aseptic conditions and add it to 500 ml of the medium base cooled to 50°C previously supplemented with lysed defibrinated horse.

Do not overheat once supplemented.

Pour the complete medium into tubes and inoculate it.

Incubate the tubes in microaerophilic conditions at 37°C for 4-6 hours, then at 41,5°C for 44 ± 4 hours.

Campylobacter spp. best grown at 42°C (except *Campylobacter fetus* subsp. *fetus*).

Incubation times longer than those mentioned above or different incubation temperatures may be required depending on the sample, on the specifications.

Each laboratory must evaluate the results according to their specifications.

Presumptive isolation of *Campylobacter spp.* must be confirmed by further microbiological and biochemical tests.

Quality control**Physical/Chemical control**

Color : White-Gray

Microbiological control

Reconstitute 1 vial as indicated in COMPOSITION; shake and dissolve completely

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

Microaerophilia. 37°C ± 1 during 5h±1; After 41,5°C±1 during ± 44h ±4

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

Subculture after incubation onto appropriate media

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

Microorganism

Campylobacter jejuni ATCC® 29428, WDCM 00156

Escherichia coli ATCC® 25922, WDCM 00013

Proteus mirabilis ATCC® 29906, WDCM 00023

Growth

Good to excelent - Typical colonial appearance

Inhibited

Inhibited

Sterility control

Add 5 ml of the sample to:

100 ml TSB and 100 ml Thioglycollate.

Incubation 48 h at 30-35 °C and 48 h at 20-25 °C: NO GROWTH.

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