

Specification

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

Presentation

	Packaging Details	Shelf Life	Storage
10 Freeze dried vials			
Vial	22±0,25 x 55±0,5 mm glass vials, tag labelled, White	49 months	2-25°C
with: 3 ± 0,1 g	plastic cap - 10 vials per box.		

Composition

Composition (g/vial)

Vancomycin.....	0.010
Trimethoprim.....	0.010
Cefoperazone.....	0.010
Cycloheximide.....	0.025

NOTE : Each vial is sufficient to supplemented
500 ml of Bolton Selective Enrichment Broth.

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Note: ISO recommends amphotericin B, but has been replaced by cycloheximide for greater availability, solubility and efficacy.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

pH: at 25°C

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

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 5ml of the sample to

100ml TSB and 100ml Thioglycollate

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

Bibliography

- BAYLIS, C.L., (editor) (2007) Manual of Microbiological Methods for the Food and Drinks Industry. 5th ed. Method 3.3.1:2007. CCFRA. Chipping Campden. UK.
- BOLTON, F.J. (2000) Methods for isolation of campylobacters from humans, animals, food and water. In "The increasing incidence of human campylobacteriosis" Report and Proceedings of a WHO Consultation of Experts. Copenhagen Denmark 21-25 November 2000, WHO/CDS/ CSRAPH 2001. p. 87-93.
- BOLTON, F.J., D. COATES, P.M. HINCHCLIFFE & L. ROBERTSON (1983) Comparison of selective media for isolation of *Campylobacter jejuni/coli*. J. Clin. Pathol. 36:78-83.
- BOLTON, F.J., D. COATES & D.N. HUTCHINSON (1984) The ability of *Campylobacter* media supplements to neutralize photochemically induced toxicity and hydrogen peroxide. J. Appl. Bacteriol. 56:151-157.
- CORRY, J.E.L., H. IBRAHIM ATABAY, S.J. FORSYTHE & L.P. MANSFIELD (2003) Culture Media for the isolation of campylobacters, helicobacters and arcobacters. In "Handbook of Culture Media for Food Microbiologists". J.E.L. Corry et al. (Eds.) Elsevier Science B.V. Amsterdam.
- DOYLE, M.P. & D.J. ROMAN (1982) Recovery of *Campylobacter jejuni* and *C. coli* from inoculated foods by selective enrichment. Appl. Environm. Microbiol. 43:1343-1353.
- FDA (Food and Drug Administrations) (1998) Bacteriological Analytical Manual. 8th ed. Revision A. AOAC International. Gaithersburg. Maryland. USA.
- HUNT, J.M., C. ABEYTA & T. TRAN (1998) *Campylobacter*. In: FDA BAM 8th ed. (revision A) 7.01-7.027 AOAC International. Gaithersburg. MD. USA.
- ISO 10272-1 Standard (2017) Microbiology of the food chain - Horizontal Method for detection and enumeration of *Campylobacter* spp. - Part 1: Detection method.
- ISO 10272-2 Standard (2017) Microbiology of the food chain - Horizontal Method for detection and enumeration of *Campylobacter* spp. - Part 2: Colony count-technique.
- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- STERN, N.J., J.E. LINE & H.C. CHEN (2001) *Campylobacter* in "Compendium of methods for the Microbiological Examination of Foods" 4th ed. F.P. Downes & K. Ito (Eds.) APHA, Washington. DC. USA.