

CAMPY THIOGLYCOLLATE MEDIUM

INTENDED USE

Remel Campy Thioglycollate Medium is a liquid medium recommended for use as a holding medium for specimens suspected of containing *Campylobacter* species prior to inoculation of solid media.

SUMMARY AND EXPLANATION

Campylobacter is a major cause of diarrheal disease in adults and children.¹ Dekeyser et al. isolated *Campylobacter jejuni* from the feces of patients with diarrhea using a filtration technique and a medium containing antibiotics to suppress normal flora.² In 1977, Skirrow used three antibiotics in a selective medium for recovery of *C. jejuni*.³ Blaser et al. reported successful recovery of *C. jejuni* from specimens which had been refrigerated overnight in a holding medium, consisting of thioglycollate broth with 0.16% agar and four antibiotics.⁴ After overnight incubation at 4°C the holding medium was subcultured to Campy Blood Agar plates. In further testing, Campy Thioglycollate was found to be especially useful for recovery of *C. jejuni* when small numbers are present in fecal specimens.⁵ Rubin et al. reported an increase in the number of isolates (from 35 to 54) recovered from specimens which had been placed in Campy Thioglycollate Medium and refrigerated prior to inoculation of agar plates.⁶

PRINCIPLE

Casein and soy peptones supply nitrogenous compounds and amino acids necessary for the growth of *Campylobacter*. Dextrose is an energy source. Sodium chloride supplies essential electrolytes and maintains osmotic equilibrium. Sodium thioglycollate is a reducing agent that removes molecular oxygen from the medium and prevents the accumulation of peroxides. Agar impedes the diffusion of oxygen, providing an environment conducive to the cultivation of *Campylobacter*. Trimethoprim, vancomycin, polymyxin B, cephalothin, and amphotericin B are antibiotics that inhibit the commensal microbial flora in fecal specimens and allow improved recovery of *Campylobacter* spp. Refrigeration temperatures also help to slow the growth of *Enterobacteriaceae* in the specimens.

REAGENTS (CLASSICAL FORMULA)*

Casein Peptone	17.0 g	Cephalothin	15.0 mg
Dextrose.....	6.0 g	Vancomycin	10.0 mg
Soy Peptone	3.0 g	Trimethoprim	5.0 mg
Sodium Chloride	2.5 g	Amphotericin B	2.0 mg
Sodium Thioglycollate	0.5 g	Polymyxin B.....	2500 U
Sodium Sulfite.....	0.1 g	Agar	1.6 g
L-Cystine.....	0.25 g	Demineralized Water	1000.0 ml

pH 7.0 ± 0.2 @ 25°C

*Adjusted as required to meet performance standards.

PROCEDURE

Note: Specimens for the isolation of *Campylobacter* spp. should be placed in a transport medium when a delay or more than 2 hours is anticipated or when a rectal swab is collected.¹

1. Campy Thioglycollate Medium should be inoculated by placing the specimen about 1 cm below the surface. Liquid stools (5 drops) and swabs are inoculated directly into the upper part of the medium; swabs may be left in the tube after breaking off the shaft. Solid stools should be emulsified in sterile saline (0.85%), before adding 5 drops to the medium.
2. Refrigerate specimen in Campy Thioglycollate Medium overnight, with cap tightened.
3. Withdraw an aliquot of the medium using a sterile Pasteur pipette positioned about 2 cm below the surface. Inoculate 2-3 drops onto Campy Blood Agar (R01280) or a suitable alternative. Streak for isolation. To achieve optimal recovery of *Campylobacter* spp. use a combination of media, such as Campy Blood Agar and Charcoal Selective Medium (R01294).¹
4. Incubate at 40-42°C for 48 hours in a microaerophilic environment (5% O₂, 10% CO₂, and 85% N₂). Plates may be set in duplicate and incubated at 33-37°C, as well as 40-42°C, to allow for the growth of certain *Campylobacter* spp.
5. Observe for characteristic colonies which can be flat, irregular, or spreading on fresh medium. Some strains appear as a thin film on the agar or form colonies that tail along the line of streaking. On less fresh medium, colonies are 1 to 2 mm in diameter, round, convex, and glistening. Colonies can be yellowish to gray or pinkish in color and are nonhemolytic.

QUALITY CONTROL

All lot numbers of Campy Thioglycollate Medium have been tested using the following quality control organism and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

CONTROL

Campylobacter jejuni ATCC® 33291

INCUBATION

Microaerophilic, up to 48 h @ 40-42°C

RESULTS

Growth recovered on subculture

LIMITATIONS

1. Some *Campylobacter* spp. are inhibited by cephalothin, including *C. fetus*, *C. upsaliensis*, *C. hyointestinalis*, and *C. lari*.^{1,7}

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Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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