Campylobacter Selective Supplement (Karmali)

PRODUCT INFORMATION

C010-1g - Cefoperazone Sodium, Powder, 1g C010-5g - Cefoperazone Sodium, Powder, 5g

V001-250mg - Vancomycin HCl, Powder, 250mg

V001-1g - Vancomycin HCl, Powder, 1g

V001-5g - Vancomycin HCl, Powder, 5g

C001-1g - Cycloheximide, Powder, 1g

C001-5g - Cycloheximide, Powder, 5g

DESCRIPTION

Campylobacter Agar Base (Karmali) with Campylobacter Selective Supplement (Karmali) is a blood free selective medium for the isolation of *Campylobacter jejuni* and *Campylobacter coli* when incubated at 42°C.

BACKGROUND

Sodium pyruvate is commonly added to cell culture media as an additional source of energy, but may also have protective effects against hydrogen peroxide.

Cefoperazone is a third generation cephalosporin antibiotic. It is one of few cephalosporin antibiotics effective in treating Pseudomonas bacterial infections which are otherwise resistant to these antibiotics.

Vancomycin is a glycopeptide antibiotic used in the prophylaxis and treatment of infections caused by Gram-positive bacteria.

Cycloheximide is widely used in biomedical research to inhibit protein synthesis in eukaryotic cells studied in vitro (i.e. outside of organisms). Its effects are rapidly reversed by simply removing it from the culture medium.

Mechanism of action

Vancomycin acts by inhibiting proper cell wall synthesis in Gram-positive bacteria. Due to the different mechanism by which Gram-negative bacteria produce their cell walls and the various factors related to entering the outer membrane of Gram-negative organisms, vancomycin is not active against Gram-negative bacteria (except some non-gonococcal species of *Neisseria*).

Cycloheximide is an inhibitor of protein biosynthesis in eukaryotic organisms, produced by the bacterium Streptomyces griseus. Cycloheximide exerts its effect by interfering with the translocation step in protein synthesis thus blocking translational elongation.

APPLICATION IN CAMPYLO-BACTER AGAR BASE (KAR-MALI)

Campylobacter Medium (Karmali) is based on the formulation described by Karmali et al. and is recommended for the isolation of *Campylobacter jejuni* and *Campylobacter coli* from clinical specimens.

The original Campylobacter Blood Free medium contains sodium pyruvate in the agar base. Campylobacter Medium (Karmali) incorporates this ingredient into the selective supplement. The original medium also contains sodium desoxycholate for the inhibition of Gram positive organisms, whereas with Campylobacter Medium (Karmali) suppression of Gram positives is achieved by the inclusion of vancomycin.

Campylobacter jejuni strains produce grey, moist, flat spreading colonies after 42 hour incubation at 42°C.

If plates are first examined after 24 hours incubation, read them immediately and quickly return them to a reduced oxygen atmosphere to ensure continued viability of the more oxygen-sensitive strains.

At 42°C selectivity is increased and growth is faster but non-thermophilic strains will not grow e.g. *Campylobacter fetus* subsp. *fetus*.

Colonies tend to swarm when initially isolated from clinical specimens.

Content concentrations

Typical Formula*	mg/litre	
Campylobacter Agar Base (Karmali)		
Columbia Agar Base	39	
Activated charcoal	4	
Haemin	0.032	
Final pH 7.4 ± 0.2 @ 25°C		
Campylobacter Selective Supplement (Karmali)		

Sodium pyruvate	100
Cefoperazone	32
<u>Vancomycin</u>	20
Cycloheximide	100
* Adjusted as required to meet performance standards	

 Table 1 typical formula for Campylobacter Agar Base (Karmali) and Campylobacter Selective Supplement (Karmali)

METHOD

Preparation

Add appropriate amount of Campylobacter Agar Base (Karmali) to distilled water and bring to the boil to dissolve. Sterilise by autoclaving at 121°C for 15 minutes. Cool to 50°C. Aseptically add Campylobacter Selective Supplement (Karmali) reconstituted as directed. Mix well and pour into sterile Petri dishes

Protocol

1. Prepare Campylobacter Selective Medium (Karmali) plates as described in the preparation for use.

2. Emulsify approximately 0.5 g of the specimen in 5 ml of sterile 0.1% peptone water to form a 1:10 dilution.

3. Inoculate on to selective medium with cotton tipped swabs so that single isolated colonies are formed.

4. Incubate the plates in an atmosphere consisting of approximately 5-6% oxygen, 10% carbon dioxide and 84-85% nitrogen for 48 hours at 42°C.

5. Examine the plates and confirm the typical colonies as *Campylobacter jejuni* or *Campylobacter coli*. A simple schema for differentiating *Campylobacter* species is described by Skirrow and Benjamin.

Quality control

Positive control:

Campylobacter jejuni ATCC[®] 33291: Good growth; grey coloured colonies

Negative control:

Escherichia coli ATCC® 25922: Inhibited

REFERENCES

1. Karmali M.A., Simor A.E., Roscoe M., Fleming P.C, Smith S.S. and Lane J. (1986) J.Clin.Micro. 23. 456-459.

2. Skirrow M.B. and Benjamin J. (1980) J.Clin.Path. 33.1122.

3. Data on file at Oxoid.