

CCDA Selective Supplement

PRODUCT INFORMATION

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

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

A007-100mg - Amphotericin B, Powder, 100mg

A007-250mg - Amphotericin B, Powder, 250mg

A007-1g - Amphotericin B, Powder, 1g

A007-5g - Amphotericin B, Powder, 5g

DESCRIPTION

Campylobacter Blood-Free Selective Agar Base with CCDA Selective Supplement can be used for the isolation of *Campylobacter jejuni*, *Campylobacter coli* and *Campylobacter laridis*.

BACKGROUND

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.

Amphotericin B is a polyene antifungal drug, often used intravenously for systemic fungal infections. It was originally extracted from *Streptomyces nodosus*. Its name originates from the chemical's amphoteric properties. Two amphotericins, amphotericin A and amphotericin B are known, but only B is used clinically, because it is significantly more active in vivo.

Mechanism of action

As with other polyene antifungals, amphotericin B associates with ergosterol, the main component of fungal cell membranes, forming a transmembrane channel that leads to monovalent ion (K⁺, Na⁺, H⁺ and Cl⁻) leakage, which is the primary effect leading to fungal cell death.

APPLICATION IN CAMPYLOBACTER BLOOD-FREE SELECTIVE AGAR BASE

Campylobacter blood-free selective agar is based on the

original formulation described by Bolton et al. which was developed to replace blood with charcoal, ferrous sulphate and sodium pyruvate. Improved selectivity was achieved when cephalosporin in the original formulation was replaced by cefoperazone as the selective agent. More recent work has shown an increased isolation rate can be achieved if the plates are incubated at 37°C rather than 42°C.

Amphotericin B has been added to the formula to suppress the growth of yeast and fungal contaminants that may occur at 37°C.

Campylobacter blood-free selective agar and Campylobacter BAP medium were equal in performance in a rapid colony-lift procedure for detection of *thermophilic campylobacters* in which membranes are blotted on agar cultures and then subjected to immunoassay.

In a study of healthy puppies and kittens for carriage of *Campylobacter* species, campylobacter blood-free selective agar was found to be a suitable medium and more productive for *Campylobacter upsaliensis* in this application than CAT medium. Campylobacter blood-free selective agar has been confirmed as suitable for isolation of *Campylobacter* spp. from non-clinical samples following enrichment in exeter broth.

Content concentrations

Typical Formula*	mg/litre
Campylobacter Blood-Free Selective Agar Base	
'Lab-Lemco' powder	10
Peptone	10
Sodium chloride	5
Bacteriological charcoal	4
Casein hydrolysate	3
Sodium desoxycholate	1
Ferrous sulphate	0.25
Sodium pyruvate	0.25
Agar	12
Final pH 7.4 ± 0.2 @ 25°C	
CCDA Selective Supplement	
Cefoperazone	32
Amphotericin B	10
* Adjusted as required to meet performance standards	

Table 1 typical formula for Campylobacter Blood-Free Selective Agar Base and CCDA Selective Supplement

METHOD

Preparation

Suspend appropriate amount of campylobacter blood-free selective agar base in distilled water and bring to the boil to dissolve. Sterilise by autoclaving at 121°C for 15 minutes. Cool to 50°C. Aseptically add supplements reconstituted as directed. Mix well and pour into sterile Petri dishes.

Protocol

1. Prepare campylobacter blood-free selective agar as described in the preparation.
2. Emulsify approximately 0.5 g of the specimen in 5 ml of sterile 0.1% peptone water to form an approximate 1:10 dilution.
3. Inoculate onto the 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 37°C.

The colonial morphology of campylobacters can be used as a guideline for identification to species level. *Campylobacter jejuni* strains produce grey, moist flat spreading colonies. Some strains may have a green hue or a dry appearance, with or without a metallic sheen. *Campylobacter coli* strains tend to be creamy-grey in colour, moist, slightly raised and often produce discrete colonies.

Colonies tend to swarm when initially isolated from clinical specimens.

Quality control

Incubation at 37°C for 48 hours.

Positive control:

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

Negative control:

Escherichia coli ATCC® 25922: No growth

REFERENCES

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4. MAFF Validated Methods for the Analysis of Foodstuffs: Method for the detection of thermotolerant *Campylobacter* in Foods (v30) J. Assoc. Publ. Analysts (1993) 29, 253-262.

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