Blood Free Campylobacter Selectivity Agar Base

M887

Intended use

Recommended for selective isolation and differentiation of *Campylobacter* species from food and animal feeding stuffs. The composition and performance criteria of this medium are as per the specifications laid down in ISO 10272-1&2:2017.

Composition**

Ingredients	Gms / Litre
HM Peptone B #	10.000
Peptone	10.000
Tryptone	3.000
Sodium chloride	5.000
Sodium deoxycholate	1.000
Ferrous sulphate	0.250
Sodium pyruvate	0.250
Charcoal, bacteriological	4.000
Agar	12.000
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Equivalent to Beef extract

Directions

Suspend 22.75 grams in 500 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add rehydrated contents of 1 vial of Campylobacter Supplement V (FD067). Alternatively to increase the selectivity of the medium, rehydrated content of one vial of CAT Selective Supplement (FD145) may be added to 500 ml sterile molten base. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Campylobacters are carried in the intestinal tract of animal and therefore contaminate foods of animal origin (11). *Campylobacter* causes intestinal upset or abortion in animals. It is also one of the most important causes of human gastroenteritis, particularly in children. Initially blood was used in the isolation of *Campylobacter*. But, later it was reported by Bolton et al (3) that charcoal can be effectively used in place of blood. This rules out the variability obtained due to the use of blood.

Blood Free Campylobacter Selectivity Agar Base (2) formulated as per APHA (11) and recommended by the ISO Committee (5) is used for selective isolation of *Campylobacter* species. Cephalothin in the original formulation was replaced by Cefoperazone as the selective agent since the latter gave better selectivity (4). *Campylobacter* species are highly resistant to cefoperazone, an antibiotic which effectively suppresses growth of *Pseudomonas* and *Enterobacteriaceae* (1,7, 9). Addition of cefoperazone increases the selectivity of the medium. Due to this addition, the medium is also known as Campylobacter Charcoal Differential Agar (CCDA). Charcoal, sodium pyruvate and ferrous sulphate reduces the aero-tolerance of medium by quenching photo-chemically generated toxic oxygen derivatives (9). Peptone, tryptone and HM peptone B serve as sources of carbon, nitrogen, amino acids, vitamins and other essential nutrients. Casein is added to help grow certain strains of nalidixic acid resistant thermophilic *Campylobacter* that are environmental organisms (10).

Additional Amphotericin B in Blood Free Campylobacter Broth Base suppresses the growth of yeast and mold contaminants. Colonies tend to swarm when initially isolated from clinical specimens.

Type of specimen

Food and animal feeding samples.

Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (11).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. Some species may show poor growth due to nutritional variations.
2. Further biochemical testing is required for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
Grey to black homogeneous free flowing powder

Gelling
Firm, comparable with 1.2% Agar gel

Colour and Clarity of prepared medium
Black coloured, opaque gel forms in Petri plates

Reaction
Reaction of 4.55% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH
7.20-7.60

Cultural Response
Cultural characteristics observed with added Campylobacter Supplement V(FD067),after an incubation at 42°C for 24-48 hours.

Organism	Growth	Inoculum (CFU)	Recovery	Colour of colony
Campylobacter coli ATCC 33559	good-luxuriant	50-100	>=50%	creamy-grey
Campylobacter jejuni ATCC 29428 (00156*)	good-luxuriant	50-100	>=50%	grey
Escherichia coli ATCC 25922 (00013*)	inhibited	>=10^4	0%	
Campylobacter laridis ATCC 35222	good-luxuriant	50-100	>=50%	varying type

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use.
Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (6,8).

Reference

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