PRODUCT SPECIFICATION SHEET

Campylobacter Agar Base (DM054)

Intended Use
Campylobacter Agar Base (DM054) is used for selective isolation of Campylobacter species from faecal, food and environmental specimens.

Product Summary and Explanation
In 1972, Dekeyser et al. reported that *C. jejuni* was isolated from the feces of patients with diarrhea and acute gastroenteritis using a filtration technique and a blood-containing selective medium with antimicrobics to suppress the normal enteric flora.[1] Subsequently, Skirrow and other investigators reported similar blood-based selective media that differed in the numbers and types of antimicrobics.[2-6] Bolton et al. reported that charcoal can effectively replace the blood in selective media for Campylobacters.[7] The selective enrichment technique is recommended for specimens and food samples that are expected to be heavily contaminated and/or carry small numbers of viable colony forming units. Infection with a *Campylobacter* species is one of the most common causes of human bacterial gastroenteritis. Most species are found in animals (cattle, swine) and cause infertility and abortion. Campylobacter Agar Base is recommended by APHA for selective isolation of *Campylobacter* species (6).

Principles of the Procedure
Campylobacter Agar supports the growth of *Campylobacter* species due to the content of peptones, yeast extract and liver digests, extracts and components specific for the individual formulations provided. Osmotic equilibrium of the medium is maintained by sodium chloride. *Campylobacter* isolation relies, in addition, on a medium's selectivity, which depends on the antimicrobial agents in the medium, a microaerophilic environment and the incubation temperature of 42°C, which suppresses the growth of most normal bacteria. Blood serves as an additional source of nutrients including X-factor. The antibiotic supplements namely Blaser-Wang (MS004) and Skirrow (MS007) markedly reduce the growth of normal enteric bacteria while enhancing the growth and recovery of *C. jejuni* from faecal specimens. Amphotericin B in Blaser- Wang supplement greatly or completely inhibits growth of fungi. *C jejuni* colonies appear non-haemolytic, flat and gray with an irregular edge or raised and round with a mucoid appearance. Some strains may appear tan or slightly pink. Swarming may be observed on moist surfaces. Incubation at 35-37°C may show a delayed growth of *C. jejuni* cultures. Incubating the plates at 42°C can fasten this.

Formula / Liter

Ingredients	Gms / Liter
Proteose peptone	15.00
Liver digest	2.50
Yeast extract	5.00
Sodium chloride	5.00
Agar	12.00
Final pH: 7.4 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

Precautions
1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions
1. Suspend 19.75 grams of the medium in 500ml of distilled water.
2. Heat to boiling, to dissolve the medium completely.

3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Cool to 45-50°C and aseptically add 5-7 %v/v sterile lysed horse blood or 10% sterile defibrinated sheep blood and rehydrated contents of 1 vial of Campylobacter Supplement-I (Blaser-Wang) (MS004) or Campylobacter Supplement-III (Skirrow) (MS007).
5. Mix well and pour into sterile Petri plates.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow colored, homogeneous, free flowing powder
Prepared Medium	Basal medium: Yellow coloured clear gel After addition of 5-7% v/v lysed blood: Reddish brown coloured opaque gel forms in Petri plates
Reaction of 3.95% Solution	pH : 7.4 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.2% Agar gel

Expected Cultural Response: Cultural characteristics observed under reduced oxygen atmosphere after an incubation at 35-37°C for 24-48 hours. (MS004Campylobacter supplement I, Blaser-Wang/ MS007-Campylobacter supplement III, Skirrow)

Sr. no	Organism	Growth w/ added MS004	Growth w/ added MS007
1.	*Candida albicans ATCC10231*	None - poor	Moderate
2.	*Campylobacter jejuni ATCC29428*	Good-luxuriant	Good-luxuriant
3.	*Escherichia coli ATCC25922*	None - poor	None - poor
4.	*Enterococcus faecalis ATCC29212*	None - poor	None - poor

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

1. Use standard procedures to obtain isolated colonies from specimens.
2. If immediate inoculation of a *Campylobacter* agar cannot be performed, the use of a suitable holding medium (e.g. Campylobacter Thioglycollate Medium with five Antimicrobics) is recommended.
3. Incubate inoculated plates at 42°C in an atmosphere conducive to the primary isolation and cultivation of microaerophilic organisms.
4. This atmosphere can be achieved using Anaerobic Jar and Anaerobic Gaspak for Campylobacter.
5. Alternatively, the atmosphere can be achieved in the Anaerobic Jar using evacuation and replacement with cylinder gases.

Results

Campylobacter jejuni produces two types of colonies. One is small, raised, grayish-brown, smooth and glistening with an entire translucent edge. The other colony type is flat, mucoid, translucent, grayish and has an irregular edge. A small percentage of strains may appear tan or slightly pinkish.[7] Colonies tend to spread and swarming may be observed, especially when initially isolated from fresh clinical specimens.
Incubation at 35-37°C may show a delayed growth of *C. jejuni* cultures. Incubating the plates at 42°C can fasten this.

Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

PRODUCT SPECIFICATION SHEET

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. Due to the presence of 15 mg/L of cephalothin, growth of *C. fetus* subsp. *fetus* will be inhibited on Campylobacter Agar with five Antimicrobics and 10% Sheep Blood; therefore, this medium is not recommended for the isolation or culture of this subspecies.
2. Since *C. jejuni* is thermophilic, it is important to incubate the plates at 42°C; otherwise, growth will be delayed. Also, the higher temperature improves selectivity by inhibiting the normal flora.

Packaging

Product Name : Campylobacter Agar Base
Product Code : DM054
Available Pack sizes : 100gm / 500gm

References

1. Dekeyser, Gossuin-Detrain, Butzler and Sternon. 1972. J. Infect. Dis. *125*:390.
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4. Blaser, Berkowitz, LaForce, Cravens, Reller and Wang. 1979. Ann. Intern. Med. *91*:179.
5. Wilson and Wang. October 13, 1979. Background and culture techniques for *Campylobacter fetus* subsp. *jejuni*. Information flier, Campylobacter Laboratory, Veterans Administration Hospital, Denver, Co.
6. Reller, Mirrett and Reimer. 1983. Abstr. C274. Abstr. Annu. Meet. Am. Soc. Microbiol. 1983.
7. Kaplan. 1980. *In* Lennette, Balows, Hausler and Truant (ed.). 1980. Manual of clinical microbiology, 3rd ed. American Society for Microbiology, Washington, D.C.
8. Karmali and Fleming. 1979. J. Clin. Microbiol. *10*:245.

Further Information

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DM054PSS, QAD/FR/024,Rev.00/01.01.2018

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PRODUCT SPECIFICATION SHEET

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