



## CAMPY-BLOOD FREE SELECTIVE MEDIUM (CCDA) (7527)

### Intended Use

**Campy Blood-Free Selective Medium (CCDA)** is used with Cefoperazone for the selective isolation of *Campylobacter* spp.

### Product Summary and Explanation

Many experts consider *Campylobacter* to be the leading cause of enteric illness in the US.<sup>1</sup> *Campylobacter* spp. can cause mild to severe diarrhea, with loose, watery stools often followed by bloody diarrhea.<sup>1</sup> These pathogens are highly infective, and transmitted by contaminated food or water.

Campy Blood-Free Selective Medium (CCDA) was described by Bolton et al.<sup>2</sup> This medium was formulated to replace blood with charcoal, ferrous sulfate, and sodium pyruvate. To improve selectivity, Cefoperazone replaced Cephazolin in the original formulation.<sup>3</sup> Bolton<sup>4</sup> recommended incubating inoculated plates at 37°C to improve isolation rates. Yeast and fungal contaminants are inhibited with the addition of amphotericin B.

Campy Blood-Free Selective Medium (CCDA) is recommended for food testing.<sup>1,5</sup> Campy Blood-Free Selective Medium with the addition of Yeast Extract and Cefoperazone is used in the Isolation of *Campylobacter* species from Foods and Swabs in the FDA/BAM Method.<sup>1</sup>

### Principles of the Procedure

Nutrient Broth No. 2 and Casein Acid Hydrolysate provides nitrogen and vitamin sources in this medium. Charcoal absorbs toxic compounds and metabolites. Sodium Desoxycholate and Cefoperazone are selective agents to inhibit enteric flora. Ferrous Sulfate and Sodium Pyruvate are present as oxygen scavengers. Agar is the solidifying agent.

### Formula / Liter

Nutrient Broth No. 2 .....	25 g
Charcoal.....	4 g
Casein Acid Hydrolysate.....	3 g
Sodium Desoxycholate .....	1 g
Ferrous Sulfate .....	0.25 g
Sodium Pyruvate .....	0.25 g
Agar .....	12 g

Final pH: 7.4 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

### Supplement (7981)

Campylobacter Supplement (CFP)  
(Cefoperazone, 16 mg)

### Precautions

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

### Directions

1. Suspend 45.5 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.
4. Cool medium to 45 - 50°C and aseptically add 4 mL of a filtered sterilized aqueous solution containing 32 mg of Cefoperazone (7981). (Each Campylobacter Supplement vial supplements 500 mL of medium.)
5. Mix well and pour into petri dishes.

### Quality Control Specifications

**Dehydrated Appearance:** Powder is homogeneous, free flowing, and grey-black.

**Prepared Appearance:** Prepared medium is opaque and black.

**Expected Cultural Response:** Cultural response on Campy Blood-Free Selective Medium (CCDA) supplemented with Cefoperzone incubated at 35 ± 2°C and examined for growth after 18 - 48 hours in an appropriate atmosphere.

Microorganism	Approx. Inoculum (CFU)	Response
<i>Campylobacter jejuni</i> ATCC® 33291	10 - 300	Good growth
<i>Escherichia coli</i> ATCC® 25922	~ 1000	Inhibited

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

### Test Procedure

1. Inoculate the specimen directly onto the surface of the prepared Campy Blood-Free Selective Medium (CCDA). If an enrichment broth is required, refer to the appropriate references.<sup>1,5,6</sup> Streak for isolation.
2. Incubate inoculated plates at 37°C or 42°C in an atmosphere composed of 5 - 6% oxygen, 3 - 10% carbon dioxide and 84 - 85% nitrogen for 24 - 48 hours. Selective temperatures are required for certain *Campylobacter* spp, refer to appropriate references on the proper temperature.<sup>1</sup>

### Results<sup>1</sup>

*Campylobacter* colonies are round to irregular with smooth edges. They may have translucent, white colonies to spreading, flat, transparent growth. Some strains appear tan or slightly pink. Typically, *Campylobacter* spp. are oxidase positive and catalase positive. Normal enteric flora is completely to markedly inhibited. For complete identification of species and biotype, refer to the appropriate procedures for biochemical reactions.<sup>1,4</sup>

### Storage

Store dehydrated medium at 2 - 30°C. Once opened and recapped, place the container in a low humidity environment at the same storage temperature. Protect from moisture and light.

### Expiration

Refer to 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.

### Limitation of the Procedure

Due to nutritional variation, some strains may grow poorly or fail to grow on this medium.

### Packaging

<b>Campy Blood Free Selective Medium (CCDA)</b>	<b>Code No.</b>	<b>7527A</b>	<b>500 g</b>
		<b>7527B</b>	<b>2 kg</b>
		<b>7527C</b>	<b>10 kg</b>
<b>Campylobacter Supplement (CFP) (Cefoperazone 16 mg)</b>		<b>7981</b>	<b>10 vials/pkg</b>
	<b>Yeast Extract</b>	<b>7184A</b>	<b>500 g</b>

### References

1. **U.S. Food and Drug Administration.** 1995. Bacteriological analytical manual, 8<sup>th</sup> ed., AOAC International, Gaithersburg, MD.
2. **Bolton, F. J., D. N. Hutchinson, and D. Coates.** 1984. J. Clin. Microbiol. **19**:169-171.
3. **Bolton, F. J., and D. N. Hutchinson.** 1984. J. Clin. Pathol. **34**:956-957.
4. **Bolton, F. J., D. N. Hutchinson, and G. Parker.** 1988. Eur. J. Clin. Microbiol. Infect Dis. **7**:155-160.
5. **Vanderzant, C., and D. F. Splittstoesser (eds.).** 1992. Compendium of methods for the microbiological examination of food, 3<sup>rd</sup> ed. American Public Health Association, Washington, D.C.
6. **Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.).** 1995. Manual of clinical microbiology, 6<sup>th</sup> ed. American Society for Microbiology, Washington, D.C.

### Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.