# Campylobacter Nitrate Broth

**M1240**

Campylobacter Nitrate Broth is used for the identification of *Campylobacter* species on the basis of nitrate reduction

## Composition**

| Ingredients | Gms / Litre |
|---|---|
| Beef heart, infusion from | 500.000 |
| Tryptose | 10.000 |
| Sodium chloride | 5.000 |
| Potassium nitrate | 2.000 |
| Final pH ( at 25°C) | 7.0±0.2 |

**Formula adjusted, standardized to suit performance parameters

## Directions

Suspend 27 grams in 1000 ml distilled water. Heat, if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and dispense as desired.

## Principle And Interpretation

*Campylobacter* species are ubiquitous in the environment inhabiting a wide variety of ecological niches (3). Infection with *Campylobacter* species is one of the most common causes of human bacterial gastroenteritis (3). Most species are found in animals (cattle, swine) and cause infertility and abortion. Campylobacter species are non-fermentative and non-oxidative in their metabolism, deceiving energy from the use of amino acids (1). Also, they do not ferment or oxidize the usual carbohydrate substrates. Campylobacter Nitrate Broth is formulated as per APHA and is used for identification of *Campylobacter* species on the basis of nitrate reduction (2). *Campylobacter jejuni* is oxidase positive and reduces nitrates.

Beef heart infusion and tryptose in the medium provide the essential nutrients including mainly the nitrogenous and a few carbon compounds to *Campylobacter species* . Sodium chloride maintains the osmotic balance of the medium. Potassium nitrate serves as the nitrate source. Biochemical reactions by which species may be differentiated are relatively few because of their inability to ferment or oxidize the usual carbohydrate substrates.

Preparation of Nitrate Test Reagents and Technique:

1. Sulphanilic acid: Dissolve 8 grams of sulphanilic acid in 1 litre 5 N acetic acid.

2. Alpha-naphthylamine reagent: Dissolve 5 grams of alpha-naphthylamine in 1 litre 5 N acetic acid.

For the test:

Put 2 - 3 drops of each reagent into the tube containing culture to be tested. A distinct red or pink colour indicates nitrate reduction. A control (uninoculated) tube should also be tested.

## Quality Control

**Appearance**
Cream to yellow homogeneous free flowing powder

**Colour and Clarity of prepared medium**
Amber coloured, clear solution without any precipitate

**Reaction**
Reaction of 2.7% w/v aqueous solution at 25°C. pH : 7.0±0.2

**pH**
6.80-7.20

**Cultural Response**
M1240: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

| Organism | Inoculum (CFU) | Growth | Nitrate reduction |
|---|---|---|---|
| **Cultural Response** | | | |
| *Acinetobacter calcoaceticus ATCC 23055* | 50-100 | good-luxuriant | negative, no colour development |
| *Campylobacter jejuni ATCC 29428* | 50-100 | good-luxuriant | positive, red colour developed within 1-2 minutes |
| *Escherichia coli ATCC 25922* | 50-100 | good-luxuriant | positive, red colour developed within 1-2 minutes |
| *Enterobacter aerogenes ATCC 13048* | 50-100 | good-luxuriant | positive, red colour developed within 1-2 minutes |
| *Salmonella Typhimurium ATCC 14028* | 50-100 | good-luxuriant | positive, red colour developed within 1-2 minutes |

## Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on label.

## Reference

1. Koneman E. W., Allen S. D., Janda W. M, Schrenckenberger P. C., Winn W. C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed, J. B. Lippincott Company.
2. Vanderzant C. and Splittstoesser D. F., (Eds.), 1992, Compendium of Methods for the Microbiological Examination of Foods, 3rd Ed., APHA, Washington, D.C.
3. Manning H., Duim B., Wassenaar T., Wagenaar A., Ridley A., Newell D. G., 2001, Appl. Environ. Microbiol., 67:1185

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