# **Nitrate Broth**



Medium used for the identification and determination of microorganisms on the basis of nitrate reduction.

## CONTENTS (Liter)

Peptone	5.0 g
Meat Extract	3.0 g
Potassium Nitrate	1.0 g
Final pH = 7.0 $\pm$ 0.2 at 25°C	-

# • PROCEDURE

Suspend 9.0 G of powder in 1 L of distilled or deionized water. Heat until completely dissolved. Sterilize by autoclave at 121°C for 15 minutes. Cool to 45 - 50°C in water bath. Mix well. Pour into tubes.

### INTERPRETATION

Nitrate Broth is a medium used for the identification and determination of microorganisms on the basis of nitrate reduction. Peptone and meat extract provide amino acids, minerals and other nitrogenous substances to support bacterial growth. Potassium nitrate for nitrate reduction is detected by the addition of alpha-naphthylamine reagent and sulphanillic acid reagent which detect the presence of nitrite through the formation of a red-orange color in the test tube.

#### TECHNIC

Inoculate fresh cultured colonies using a sterile loop to the medium. Incubate at  $35 \pm 2^{\circ}C$  for 24 hours. After incubation, add 2 - 3 drops of Sulphanilic Acid Reagent and 2 - 3 drops of  $\alpha$ -Naphthylamine Reagent. Shake gently and await some seconds. Observe the development of red-orange color (Positive reaction). In case of negative reaction add a small quantity of zinc powder and observe the eventual development of the red color. Refer appropriate references for recommended test procedure.

### • QUALITY CONTROL FOR USE

<u>Dehydrated medium</u> Appearance: free-flowing, homogeneous Color: light beige <u>Prepared medium</u> Appearance: clear Color: light amber Incubation conditions:  $35 \pm 2^{\circ}C / 24$  hours

Microorganism	ATCC	Growth	Nitrate Reduction
Enterobacter aerogenes	13048	good	+
Escherichia coli	25922	good	+
Listeria monocytogenes	15313	good	-

# STORE

The powder is very hygroscopic. Store the powder at room temperature, in a dry environment, in its original container tightly closed and use it before the expiry date on the label. Store prepared medium at 2 - 8°C.

## REFERENCES

- 1. Ewing 1986. Edwards & Ewing's. Identification of Enterobacteriaceae, 4th ed. Elsevier Science Publishing Co. Inc. New York.
- 2. MacFaddin 1980. Biochemical Tests for the identification of medical bacteria, 2nd ed. Williams & Wilkins, Baltimore.
- 3. Finegold and Baron. 1986 Bailey and Scott's. Diagnostic microbiology, 7th ed. The C.V. Mosby Company, St. Louis.
- 4. Kelly, Brenner and Farmer. 1985. In Lennette, Balows, Hausler and Shadomy (ed.), Manual of clinical microbiology, 4th ed. ASM, Washington, D.C.

### PACKAGE

Cat. No : MB-N1322 Nitrate Broth	500 G
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