



## Ellners Broth

M466

Ellners Broth is used to induce spore formation in *Clostridium perfringens*.

### Composition\*\*

Ingredients	Gms / Litre
Proteose peptone	10.000
Yeast extract	3.000
Starch	3.000
Magnesium sulphate	0.100
Monopotassium phosphate	1.500
Disodium phosphate	50.000
Final pH ( at 25°C)	7.8±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 67.6 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Dispense in tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

### Principle And Interpretation

Ellners Broth is recommended (1, 2) for inducing sporulation in *Clostridium perfringens*. Spores are rarely seen in culture (a diagnostic feature) but can be obtained on Ellners Medium (1). In practice, the routine characterization of clostridia to species level involves morphological examinations, biochemical tests and identification of specific toxins. All clostridia produce spores but they vary markedly in their readiness to do so. Some of which may require prolonged incubation.

*C. perfringens* are gram-positive rods, often capsulated. In sugar-containing media, the *Clostridium* rods are shorter whereas in protein-containing media, they may become filamentous. Spores formed are usually in small numbers and are not formed in the presence of fermentable carbohydrates. Typically oval, sub-terminal or central spores are formed and are not bulging. Special media like Ellners Broth are used to produce spores.

Medium is composed of proteose peptone and yeast extract, which supply the necessary nutrients for the growth of the Clostridia. Generally sporulation is stimulated by a carbohydrate source and hence starch is included in the medium. Sulphate and phosphate not only buffer the medium but also help in sporulation. Clostridia are anaerobic organisms and hence anaerobiosis may be ensured by heating the medium at 100°C for 10 minutes and cooling just before inoculation. It is important that the inoculum should be adequate. 0.5 ml of an actively growing 4-12 hours Meat Broth culture should be introduced with a pipette into the bottom of the tubed medium and incubated anaerobically.

### Quality Control

#### Appearance

Cream to yellow homogeneous free flowing powder

#### Colour and Clarity of prepared medium

Amber coloured, clear to slightly opalescent solution

#### Reaction

Reaction of 6.76% w/v aqueous solution at 25°C. pH : 7.8±0.2

#### pH

7.60-8.00

#### Cultural Response

M466: Cultural characteristics observed under anaerobic condition, after an incubation at 35-37°C for 24-76 hours.

Organism	Inoculum (CFU)	Growth	Sporulation
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#### Cultural Response

*Clostridium perfringens* 50-100 luxuriant positive  
ATCC 12924

### 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 the label.

### Reference

1. Collee J. G., Duguid J. P., Fraser A. G., Marmion B. P., (Eds.), Mackie and McCartney, Practical Medical Microbiology, 1989, 13th Edition, Churchill Livingstone.
2. Trevor W. A., 1977, Anaerobic Bacteriology, 3rd Ed., Butterworths and Co. Ltd.

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