

## **NUTRIENT BROTH No. 2 B.P.**

A general purpose liquid medium for the cultivation of microorganisms

**Code: KM1075**

<b>Typical formula</b>	<b>(g/l)</b>
Beef Extract	10.0
Peptone	10.0
Sodium Chloride	5.0

pH 7.3 +/- 0.2

### **Directions**

Suspend 25g in 1000ml of cold distilled water. Mix well, distribute and sterilise by autoclaving at 121°C for 15 minutes.

### **Description**

Nutrient Broth No.2 containing meat peptone and sodium chloride is a general purpose liquid medium richer than Nutrient Broth E. It gives good growth from small inocula. Recommended as basal medium by ISO 10272 for the preparation of Campylobacter Enrichment Broth.

**Quality assurance** (24 hrs - 37°C)

### Productivity control

*S.aureus* ATCC 25923: growth

*E.coli* ATCC 25922: growth

### **Storage**

Dehydrated media: 15-30°C

User prepared plates: 1 month at 2-8°C

User prepared tubes and flasks: 3 months at 2-8°C

### **Reference**

ISO 10272:1995-Microbiology of food and animal feed -Horizontal method for the detection of thermotolerant *Campylobacter*.

## NUTRIENT GELATIN

For determination of gelatin liquefaction and for the 20°C plate count.

**Code: KM1076**

### Typical formula (g/l)

Beef Extract	3.0
Peptone	5.0
Gelatin	120.0

pH 6.8 +/- 0.2

### Directions

Suspend 128g in 1000ml of cold distilled water, leave for 10 minutes then bring to the boil to dissolve completely, distribute and sterilise by autoclaving at 121°C for 15 minutes.

### Description

Nutrient Gelatin is used for the enumeration of microorganisms in water specimens at an incubation temperature of 20°C, and to determine the ability of bacteria to liquefy gelatin. Gelatin was one of the first solidifying agents in the preparation of culture media for microbiology. However, because of its low gel point (25°C), a temperature, which is definitely not optimal for microbial growth. It was later that Gelatin was replaced with agar. The medium is now mainly used to determine the ability to liquefy gelatin, a standard method in taxonomic studies.

### Method

To perform a gelatin liquefaction test, inoculate a test tube of Nutrient Gelatin solidified at 4°C with a drop of heavy microbial suspension and incubate at 22-25°C for 24 hours to 14 days. Every 24 hours put the tubes at 2-8°C for 10 minutes and observe whether the gelatin is able to solidify the medium. If the microorganisms being examined contain proteolytic enzymes, which hydrolyse gelatin, the medium remains liquid. For the standard plate count on water recommended by APHA (1946), dilute the sample with sterile water and place 0.5 or 1ml of the dilutions in each dish of at least two duplicate sets of sterile petri dishes. Cool the Nutrient Gelatin to around 42°C and aseptically add 10ml to each dish. Mix the medium with the inoculum, solidify as soon as possible after pouring, and immediately place in an incubator at 19-21°C. Incubate for 48 hour and count at least two plates made from the dilution giving between 30 and 300 colonies per plate.

**Quality assurance** (48 hrs -20°C)

### Productivity control

*S.aureus* ATCC 25923: growth, gelatin liquefaction

*E.coli* ATCC 25922: growth

### Storage

Dehydrated media: 15-30°C

User prepared tubes and flasks: 3 months at 2-8°C

### References

APHA (1946) Standard Methods for the Examination of Water and Sewage, 5<sup>th</sup> ed.

## NUTRIENT AGAR

A general purposes medium

**Code: KM1073**

<b>Typical formula</b>	<b>(g/l)</b>
Beef Extract	3.0
Peptone	5.0
Sodium Chloride	8.0
Agar	12.0

Final pH 7.3 +/- 0.2

### **Directions**

Suspend 28g of Nutrient Agar in 1000ml of cold distilled water. Heat to dissolve and sterilise by autoclaving at 121°C for 15 minutes.

### **Description**

Nutrient Agar is used for the cultivation of non-fastidious microorganisms. The Beef Extract and Peptone supply a quantity of carbon, nitrogen and vitamins sufficient for the growth of most non-fastidious microorganisms (enteric bacteria, staphylococci). The APHA recommends the use of Nutrient Agar in the microbiological examination of water and dairy products. These media can be used as bases to which different materials can be added such as; carbohydrates, salts, dyes, etc., in order to obtain a selective, differential and enriched media.

Nutrient Agar was the first media utilised in microbiology and can still be routinely used in the examination of water and foods for preparing stock cultures; for the preliminary cultivation of a sample undergoing successive bacteriological examinations; and for the isolation of microorganisms from a pure culture.

**Quality assurance** (24 h -37°C)

### Productivity control

*S.aureus* ATCC 25923: growth

*E.coli* ATCC 25922: growth

### **Storage**

Dehydrated media: 15-30°C

Prepared plates: 1 month at 2-8°C

Prepared tubes and flasks: 3 months at 2-8°C

### **References**

APHA (1960) - Standard Methods for Examination of Water and Wastewater.

APHA (1976) - Compendium of Methods for Microbiological Examination of Foods.

## **NUTRIENT BROTH “Economy”**

A general purpose liquid medium for the cultivation of non-demanding microorganisms.

**Code: KM1075E**

<b>Typical formula</b>	<b>(g/l)</b>
Beef Extract	1.0
Yeast Extract	2.0
Peptone	5.0
Sodium Chloride	5.0

pH 7.4 +/- 0.2

### **Directions**

Suspend 13g in 1000ml of cold distilled water. Mix well, distribute and sterilise by autoclaving at 121°C for 15 minutes.

### **Description**

Nutrient Broth E, containing peptones, sodium chloride and yeast extract, is a general-purpose liquid medium for the cultivation of microorganisms not exacting in their nutritional requirements. Nutrient Broth E, can be enriched with other ingredients such as carbohydrates, blood etc. for special purposes.

**User quality assurance** (24 h -37°C)

### Productivity control

*S.aureus* ATCC 25923: growth

*E.coli* ATCC 25922: growth

### **Storage**

Dehydrated media: 15-30°C

User prepared tubes and flasks: 3 months at 2-8°C