Nutrient Gelatin

M060

Intended Use:

Recommended for detection of gelatin liquefaction by proteolytic microorganisms from clinical and nonclinical samples.

Composition**

Ingredients	g / L
Peptone	5.000
HM peptone B #	3.000
Gelatin	120.000
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters

- Equivalent to Beef extract

Directions

Suspend 128.0 grams in 1000 ml of warm (50°C) purified / distilled water. Heat to boiling to dissolve the medium completely. Dispense into test tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Allow the tubed medium to cool in an upright position.

Principle And Interpretation

Nutrient Gelatin is prepared as per the formulation formerly used in the examination of water, sewage and other materials of sanitary importance (1). Gelatin liquefaction is one of the essential test for the differentiation of enteric bacilli (2). This medium can also be used for the microbial plate counts of water.

Peptone and HM peptone B supplies nitrogen and carbon source, long chain amino acids and other growth nutrients for the growth of non-fastidious organisms. Gelatin is the substrate for the determination of the ability of an organism to produce gelatinase, a proteolytic enzyme active in the liquefaction of gelatin.

An 18-24 hours old pure culture from Triple Sugar Iron Agar (M021) or Kligler Iron Agar (M078) is stab-inoculated in Nutrient Gelatin with an inoculating needle directly down the centre of the medium to a depth of approximately one half an inches from the bottom of the tube. Incubate the tubes including an un-inoculated control at 35±2°C for 24-48 hours. Many species require prolonged incubation (3,4) for gelatin liquefaction. Gelatin is solid at 20°C or less temperature and liquid at 35°C or higher temperature. Gelatin liquefies at about 28°C, so incubation is carried out at 35°C but kept in a refrigerator for about 2 hours before interpretation of the results (3). Liquefaction of gelatin occurs on the surface layer, so care should be taken not to shake the tubes (5). Control is run along with every testing as gelling ability of gelatin varies (3) and also the gelatin concentration should not exceed 12% as it may inhibit growth (6). For plate counts of water, the incubation is carried out at 20-22°C for upto 30 days.

Nutrient Gelatin Medium is not recommended for determination of gelatin liquefaction by fastidious species and obligate anaerobes. At various intervals during the incubation process, examine the tubes for growth and liquefaction. At each interval, tighten the caps and transfer the tubes to refrigerator for sufficient time period to determine whether liquefaction has occurred or not.

Type of specimen

Isolated Microorganisms from clinical samples; Water samples

Specimen Collection and Handling:

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1). For clinical samples follow appropriate techniques for handling specimens as per established guidelines (7,8). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic Use. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. It is not recommended for determination of gelatin liquefaction by fastidious species and obligate anaerobes.
2. If the tubes are incubated at temperatures greater than 20ºC the tubes must be chilled below 20ºC before reactions can be carried out (9).
3. Do not shake the tubes after incubation, as some positive liquefaction reactions will be missed (9) .

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing slightly coarse powder

Gelling

Semisolid, comparable with 12.0% Gelatin gel

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in tubes as butts

Reaction

Reaction of 12.8% w/v aqueous solution at 25°C. pH : 6.8±0.2

pH

6.60-7.00

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 1 to 7 days, (Incubated anaerobically for *Cl.perfringens*), (For gelatinase test, cool below 20°C).

Organism	Growth	Gelatinase
Clostridium perfringens ATCC 12924	good-luxuriant	positive reaction
** *Bacillus spizizenii* ATCC 6633 (00003*)	good-luxuriant	positive reaction
Escherichia coli ATCC 25922 (00013*)	good-luxuriant	negative reaction
## *Proteus hauseri* ATCC 13315	good-luxuriant	positive reaction
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	good-luxuriant	positive reaction

Key : (*) Corresponding WDCM numbers.

**Formerly known as *Bacillus subtilis* subsp. *spizizenii* ## Formerly known as *Proteus vulgaris*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (7,8).

Reference

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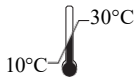
IVD ***In vitro* diagnostic medical device**



Storage temperature

EC REP CEpartner4U, Esdoornlaan 13,
3951DB Maarn, NL
www.cepartner4u.eu

CE **CE Marking**

Do not use if package is damaged

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