

# **70151 Nutrient Gelatin NutriSelect® Plus**

Nutrient Gelatin is recommended for the determination of gelatin-liquefying microorganisms as well as for the enumeration of proteolytic organisms in water by the plate count test.

## **Composition:**

Ingredients	Grams/Litre	
Meat extract	3.0	
Peptone	5.0	
Gelatin	120.0	

Final pH 6.8 +/- 0.2 at 20°C

Store granulated media between 10-30°C in tightly closed container and the prepared medium at 20-30°C. Avoid freezing and overheating. Once opened keep powdered medium closed to avoid hydration. Use before expiry date on the label.

Appearance(color): Faint yellow and Faint beige and Faint brown, free flowing powder

Gel strength: Semisolid, comparable with 12.0% Gelatin gel

Color and Clarity: Light amber coloured clear to slightly opalescent gel forms in tubes as butts

#### **Directions:**

Suspend 128 g in 1 litre of distilled water. Bring to the boil to dissolve completely. Sterilize by autoclaving at 121°C for 15 minutes. Mix well before pouring and cool to below 20°C or leave to set in a refrigerator.

## **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 tests for the differentiation of enteric bacilli (4). This medium can also be used for the microbial plate counts of water.

Peptone 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. Gelatinases are proteases secreted extracellularly by some bacteria which hydrolyze or digest gelatin. The production of gelatinases is used as a presumptive test for the identification of various organisms, including Staphylococcus sp., Enterobacteriaceae, and some grampositive bacilli.

An 18–24-hour old pure culture from Triple Sugar Iron Agar or Kligler Iron Agar is stab-inoculated in Nutrient Gelatin with an inoculating needle directly down the center of the medium to a depth of approximately one half an inch from the bottom of the tube. Incubate the tubes including an uninoculated control at 35±2°C for 24-48 hours. Many species require prolonged incubation (2, 5) 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 (2). Liquefaction of gelatin occurs on the surface layer, so care should be taken not to shake the tubes (3). Control is run along with every testing as gelling ability of gelatin varies (2) and the gelatin concentration should not exceed 12% as it may inhibit growth (2). For plate counts of water, the incubation is carried out at 20-22°C for up to 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 to determine whether liquefaction has occurred or not.

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

Organisms (ATCC)	Inoculum (CFU)	Growth	Gelatinase
Clostridium perfringens (12924/-) Bacillus subtilis (6633/00003)	50-100 50-100	++/+++ ++/+++	positive reaction positive reaction
Escherichia coli (25922/ 00013) Staphylococcus aureus	50-100 50-100	+++/+++ ++/+++	negative reaction positive reaction
subsp. aureus (25923/ 00034) Proteus vulgaris (13315/-)	50-100	++/+++	Positive reaction

#### References:

- 1. American Public Health Association, 1975, Standard Methods for the Examination of Water and Wastewater, 14th Ed., APHA, Washington, D.C.
- 2. Cawan S. and Steel K., 1966, Manual for the Identification of Medical Bacteria, Cambridge University Press, Pg. 19, 27-28, 116 and 156.
- 3. Frobisher M., 1957, Fundamentals of Microbiology, 6th Ed., W.B. Saunders Co., Philadelphia, p. 239.
- 4. Ewing, 1986, Edwards and Ewings Identification of Enterobacteriaceae, 4th Ed., Elsevier Science Publishing Co., Inc. New York.
- 5. Lautrop H., 1956, Acta. Pathol. Microbiol. Scand., 39:357

### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

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