Technical Information

Glucose Starch Agar

Product Code: DM 1989

Application: - Glucose Starch Agar is used as a basal medium with the addition of salicin, raffinose and phenol red for detection of *Clostridium perfringens.*

Composition**

Ingredients	Gms / Litre
Proteose peptone	15.000
Dextrose	10.000
Starch, soluble	5.000
Sodium chloride	5.000
Disodium hydrogen phosphate	3.000
Gelatin	20.000
Agar	10.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Clostridial species are one of the major causative agent of food poisoning/ gastro-intestinal illnesses. They are gram-positive, spore-forming rods that occur naturally in the soil [(1).] Among the family are: *Clostridium botulinum,* produces the most potent toxins; *Clostridium tetani,* causative agent of tetanus; and *Clostridium perfringens,* commonly found in wound infections and diarrhoea cases. The use of toxins to damage the host is a method adopted by many bacterial pathogens. The major virulence factor of *C.perfringens* is the CPE enterotoxin, which is secreted upon invasion of the host gut, and contributes to food poisoning and other gastrointestinal illnesses [(1).] Glucose Starch Agar is used as a basal medium, which with the addition of raffinose, salicin and phenol red indicator is used for detecting *C. perfringens* [(2).] This medium is also recommended by APHA [(3).]

The medium contains proteose peptone, which supplies the nitrogenous nutrients for *C.perfringens.* Dextrose is the fermentable carbohydrate source and is fermented by most Clostridia. However, raffinose and salicin are fermented with acid and gas production by only some strains of *C.perfringens.* Dispense the medium in different tubes and add a few drops of phenol red, the pH indicator, which turns yellow at acidic pH. Gas production is indicated by bubble formation. Gelatin is liquefied by *C.perfringens* within 48 hours. Sodium chloride maintains the osmotic balance of the medium.

Methodology

Suspend 68 grams of powder media in 1000 ml distilled water. Shake well & heat to dissolve the medium completely. Dispense in tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 30 minutes. Allow the tubed medium to cool in an upright positio n.

Quality Control

Physical Appearance
Cream to beige homogeneous free flowing powder

Gelling
Firm, comparable with 1.0% Agar gel and 2.0% Gelatin.

Colour and Clarity of prepared medium
Light amber coloured, clear to slightly opalescent gel forms in tubes as butts

Reaction
Reaction of 6.8% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH Range 7.00-7.40

Cultural Response/ characteristices
DM1989**:** Cultural characteristics observed after an incubation at 35-37°C for 24-72 hours. Dextrose fermentation is detected using phenol red indicator

Organism	Inoculum (CFU)	Growth	Raffinose (72 hours)	Salicin (24 hours)
Clostridium perfringens ATCC 12924	50-100	luxuriant	acid production, yellow colour	negative reaction, no colour change or red
Clostridium paraperfringens	50-100	luxuriant	negative reaction, no colour change or red	prodution,yellow colour and bubble formation
Escherichia coli ATCC 25922	50-100	luxuriant	negative reaction, no colour change or red	negative reaction, no colour change or red

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8^{o} in sealable plastic bags for 2-5 days.

Further Reading

1. Czeczulin J.R., Hanna P.C., Mcclane B.A., Cloning, nucleotide sequencing, and expression of the Clostridium perfringens enterotoxin gene in! Escherichia coli @. Infect. Immun. 61: 3429-3439 (1993).
2. Hauschild A. H. W. and Hilsheimer R., 1974, Appl. Microbiol., 27:78.
3. Speck M. L., (Eds.), 1984, Compendium of Methods for the Microbiological Examination of Foods, 2nd Ed., APHA, Washington, D.C.

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