**TECHNICAL SHEET**

| **B544** | **GLUCOSE STARCH AGAR** |
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**Formula**

| **Ingredients :** | **gms/lit.** |
|---|---|
| Proteose peptone | 15.00 |
| Dextrose | 10.00 |
| Starch, soluble | 5.00 |
| Sodium chloride | 5.00 |
| Disodium hydrogen phosphate | 3.00 |
| Gelatin | 20.00 |
| Agar | 10.00 |

**Final pH (at 25°C) : 7.2 ± 0.2**

**Directions :**

Suspend 68.0 grams in warm 1000 ml purified/distilled water. Heat to boiling 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 position.

**Principle :**

Clostridial species are one of the major causes of food poisoning/ gastro-intestinal illnesses. They are gram-positive, spore-forming rods that occur naturally in the soil. Among the family are: *Clostridium botulinum*, which produces one of the most potent toxins in existence; *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 deployed 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. 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*. This medium is also recommended by APHA.

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.

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| **QC Tests – (I)Dehydrated Medium** | |
| Colour : | Cream to Beige |
| Appearance : | Homogeneous Free Flowing powder |
| **(II)Rehydrated medium** | |
| pH (post autoclaving/heating) : | 7.2 ± 0.2 |
| Colour (post autoclaving/heating) : | Light amber |
| Clarity (post autoclaving/heating) : | Clear to slightly opalescentgel forms in tubes as butts |

**(III)Q.C. Test Microbiological**

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

| MICROORGANISM (ATCC ) | GROWTH | SALICIN (24 HOURS) | RAFFINOSE (72 HOURS) |
|---|---|---|---|
| Clostridium perfringens (12924) | Luxuriant | - | A |
| Clostridium paraperfringens | Luxuriant | AG | - |
| Escherichia coli (25922) | Luxuriant | - | - |
| Key : A = Acid production AG = Acid and Gas production | | | |

**Precautions :**

1. For Laboratory Use.
2. Follow proper, established laboratory procedures in handling and disposing of infectious materials.

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| **Limitations :** | 1. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium. | | | | |
| **Use :** | Used as a basal medium with the addition of salicin,raffinose and phenol red for detection of Clostridium perfringens. | | | | |
| **Storage :** | Dehydrated medium- below 30°C Prepared medium– Between 2 to 8°C. | | | | |
| **Packing :** | 500 gm bottle | | | | |
| **Product profile:** | Reconstitution | Quantity on Preparation (500g) | pH (25°C) | Supplement | Sterilization |
| **B544** | 68g/l | 7.35 L | 7.2 ± 0.2 | nil | $121^0$C / 15 minutes |