# MP-5 Medium

M596

MP-5 Medium is used for the detection of pectinolytic microorganisms especially those producing polygalacturonase.

## Composition**

| Ingredients | Gms / Litre |
|---|---|
| Pectin | 5.000 |
| Monopotassium phosphate | 4.000 |
| Disodium phosphate | 6.000 |
| Ammonium sulphate | 2.000 |
| Yeast extract | 1.000 |
| Ferrous sulphate | 0.001 |
| Magnesium sulphate | 0.200 |
| Calcium chloride | 0.001 |
| Boric acid | 0.00001 |
| Manganese sulphate | 0.00001 |
| Zinc sulphate | 0.00007 |
| Copper sulphate | 0.00005 |
| Molybdenum trioxide | 0.00001 |
| Agar | 15.000 |
| Final pH ( at 25°C) | 5.5±0.5 |

**Formula adjusted, standardized to suit performance parameters

## Directions

Suspend 33.2 grams in 1000 ml. distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121oC) for 15 minutes. Mix well and pour into sterile Petri plates.

Note : Due to presence of various inorganic salts, slight precipitate may develop upon heating. Shake well before pouring into sterile Petri plates.

Polysaccharide precipitant solution : Dissolve 1.0 gm of hexadecyltrimethyl ammonium bromide in 100 ml of water and the solution is sterilized by autoclaving if desired.

## Principle And Interpretation

Pectin is an important cell wall component of higher plants that helps in cementing plant cells together. Most pectin-degrading organisms are associated with raw agricultural products and with soil. Detection of pectinolytic activity of an organism is carried out either by observing depression in the gel around the colony where the substrate has been degraded or by flooding the plate with a precipitant solution.

MP-5 Medium is used for the detection of pectinolytic organisms especially those producing polygalactouronase. MP-5 medium is recommended by APHA for detecting pectinolytic organisms (1).

Detection of polygalactouronase by plate assay is generally done by lowering the pH of the medium, designed for detection of pectate lyase, (i.e. MP-7 Medium) to 6 or below so that polygalactouronase will be active and pectate lyases will be inactive (2, 3). Thus the acidic pH of MP-5 Medium is the main parameter used to distinguish polygalactouronase producers from pectate lyase producers. A 1.0% aqueous solution of hexadecyltrimethyl ammonium bromide (4) is used to detect pectinolytic activity. After incubating the plates for 2-3 days at 30-35°C, the polysaccharide precipitant is poured over the surface of the plate taking care not to dislodge the colonies. Zones of pectin hydrolysis will be visible usually within few minutes and can be best viewed against dark background. The reagent precipitates the intact pectin in the medium whereas pectinolytic growth is surrounded by a clear halo in an opaque medium. High phosphate level in the medium is required to observe pectinolytic activity.

## Quality Control

**Appearance**
Cream to yellow homogeneous free flowing powder

**Gelling**
Firm, comparable with 1.5% Agar gel.

**Colour and Clarity of prepared medium**
Yellow coloured clear to slightly opalescent gel forms in Petri plates

**Reaction**
Reaction of 3.32% w/v aqueous solution at 25°C. pH : 5.5±0.5

**pH**
5.00-6.00

**Cultural Response**
Cultural characteristics observed after an incubation at 35-37°C for 2-3 days.

**Cultural Response**

| **Organism** | **Polygalacturonase production** |
|---|---|
| **Cultural Response** | |
| **Aspergillus brasiliensis ATCC 16404* | positive, clear halo around the colony when flooded with 1% polysaccharride precipitant. |
| *Fusarium moniliforme* | positive, clear halo around the colony when flooded with 1% polysaccharride precipitant |

## Storage and Shelf Life
Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

## Reference
1. Downes F. P. and Ito K. (Eds.), 2001, Compendium for the Microbiological Examination of Foods, 4th Ed. APHA, Washington, D.C.
2. Hankin L. and Anagnostakis S. L., 1975, Mycologia 67:597.
3. Vaughn R. H., Balatsouras G. D., York G. K. II and Nagel C. W., 1957, Food Res. 22:597.
4. Jayasankar N. P. and Graham P. H., 1970, Can J. Microbiol., 16:1023.

Revision : 2 / 2015