



# **Rose Establishment Medium**

With Vitamins, Sucrose, 6-BAP, IAA and Agar

## **Product Code: PT144**

## **Product Description :**

Rose Establishment Medium has been formulated for the *in vitro* culture of *Rosa*, family *Rosaceae*. It is based on the Murashige and Skoog media composition with certain alterations improving its suitability for the Rose species. The formulation is a nutrient blend of inorganic salts, vitamins, amino acid, carbohydrate and gelling agent.

The medium provides all the essential macroelements and microelements. Potassium nitrate and ammonium nitrate serve as sources of nitrogen. Potassium dihydrogen phosphate provides phosphate. Microelements like Manganese, Molybdenum, Copper, Iron and Zinc enhance metabolism in plants. Boron plays a key role in carbohydrate metabolism. Thiamine, pyridoxine, nicotinic acid act as enzymatic cofactors in universal pathways including glycolysis and TCA cycle along with primary and secondary metabolism in the plants. Glycine serves as a source of amino acid. Ascorbic acid stimulates cell division and cell elongation. It also acts as antioxidant along with citric acid and helps to reduce browning of the medium. 6-BAP aids in stem elongation and proliferation while IAA induces callusing and rooting.

The product is plant tissue culture tested but it is the sole responsibility of the user to ensure the suitability of the medium for individual species.

### **Composition :**

| Ingredients                   | mg/L     |
|-------------------------------|----------|
| MACROELEMENTS                 |          |
| Ammonium nitrate              | 1650.000 |
| Calcium chloride              | 332.200  |
| Magnesium sulphate            | 180.690  |
| Potassium nitrate             | 1900.000 |
| Potassium phosphate monobasic | 170.000  |
| MICROELEMENTS                 |          |
| Boric acid                    | 6.200    |
| Cobalt chloride hexahydrate   | 0.025    |

| Copper sulphate pentahydrate   | 0.025     |
|--------------------------------|-----------|
| EDTA disodium salt dihydrate   | 37.300    |
| Ferrous sulphate heptahydrate  | 27.800    |
| Manganese sulphate monohydrate | 16.900    |
| Molybdic acid (sodium salt)    | 0.213     |
| Potassium Iodide               | 0.830     |
| Zinc sulphate heptahydrate     | 8.600     |
| VITAMINS                       |           |
| L-Ascorbic acid                | 50.000    |
| myo-Inositol                   | 100.000   |
| Nicotinic acid (free acid)     | 0.500     |
| Pyridoxine HCl                 | 0.500     |
| Thiamine hydrochloride         | 0.400     |
| AMINO ACID                     |           |
| Glycine                        | 2.000     |
| CARBOHYDRATE                   |           |
| Sucrose                        | 30000.000 |
| GELLING AGENT                  |           |
| Agar                           | 7000.000  |
| OTHERS                         |           |
| 6-Benzylaminopurine            | 2.000     |
| Citric acid                    | 50.000    |
| Indole-3-acetic acid           | 0.300     |
| Total(gms/litre)               | 41.5      |

## Material required but not provided :

• Autoclaved distilled water

• 1N NaOH/HCl

## **Precautions :**

• Ensure appropriate pH of the medium before addition of gelling agent as acidic pH will lead to decreased gelation resulting in semi solid flowing gel while alkaline pH will lead to formation of hardened gel.

• Use of Distilled water/Tissue culture grade water is recommended for media preparation as tap water or lower grade water may lead to salt precipitation and improper gelation.

• Avoid preparation of concentrated solutions, as it will lead to precipitation of salts.

## **Directions :**

• Reconstitute medium by adding required quantity of powder in two-third of total volume with constant, gentle stirring till the medium gets completely dissolved.

• Add heat stable supplements prior to autoclaving.

• Make up the final volume with distilled water.

 $\bullet$  Adjust the pH of the medium to  $5.75\pm0.5$  using 1N NaOH/ HCl.

• Heat the medium to boiling till complete dissolution of gelling agent.

• Sterilize the medium by autoclaving at 15 lbs and 121°C for 15 min.

• Cool the autoclaved medium to about 45°C before adding heat labile supplements.

• Aseptically dispense the desired amount of medium under a laminar airflow unit in sterile culture vessels.

## **Quality Control:**

#### Appearance

White to off-white, homogenous, free flowing powder

#### Solubility

41.5 gms/litre soluble after boiling in distilled water

#### **Colour and Clarity**

Colourless to light yellow solution, hazy gel is formed on cooling

#### Gelling

Firm gel formed at pH:  $5.75 \pm 0.5$ 

#### pH at 25°C

3.30 - 4.30

#### **Plant Tissue Culture Test**

The growth promoting properties of medium is assessed by providing plant cultures with relative humidity of about  $60\% \pm 2\%$ , temperature  $22^{\circ}C \pm 2^{\circ}C$  and photoperiod of about 16:8. The plant species showed actively growing callus and shoots with no structural, necrotic and toxic deformity.

## **Storage and Shelf Life:**

• The plant tissue culture medium powder is extremely hygroscopic and must be stored at 2-8°C in air tight containers.

• Preferably, entire content of each package should be used immediately after opening.

• Use before the expiry date.

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#### Disclaimer :

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