

Technical Information

Rose Establishment Medium

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

Product Code: PT1144

Application: Rose Establishment Medium has been formulated for their *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/Litre
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	41.5ms/litre

Material required but not provided

- Autoclaved distilled water
- 1N NaOH/HCl

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

pH at 25°C

3.30-4.30

Gelling

Firm gel formed at pH: 5.75 ± 0.5

Plant Tissue Culture Test

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

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.
- Adjust the pH of the medium to 5.75 ± 0.5 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

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.

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.

Disclaimer

- User must ensure suitability of the product(s) in their application prior to use.
- The product conforms solely to the technical information provided in this booklet and to the best of knowledge research and development work carried at **CDH** is true and accurate.
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