

Gamborg B5 Medium

With Calcium Chloride, Vitamins, Sucrose and Agar

Product Code: PT017

Product Description :

Gamborg B5 medium has been established by Gamborg O.L (1968) for the callus and cell suspension culture of *Glycine max*, family *Fabaceae*. This medium is widely used for *in vitro* plant cell, tissue and organ culture.

Gamborg B5 medium is a nutrient blend of inorganic salts, vitamins, carbohydrate and gelling agent. Increased potassium nitrate serves as a sole source of nitrate as nitrate content is beneficial for soyabean root callus and ammonium sulphate enhances the cell growth. Sodium dihydrogen phosphate serves as a phosphate source and the microelements like Boron, Manganese, Molybdenum, Copper, Iron and Zinc play vital role in plant metabolism. Boron plays a key role in carbohydrate metabolism. Vitamins like thiamine, pyridoxine, nicotinic acid act as enzymatic cofactors in universal pathways including glycolysis and TCA cycle along with the primary and secondary metabolism in the plants.

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 sulphate	134.000
Calcium chloride	113.250
Magnesium sulphate	122.090
Potassium nitrate	2500.000
Sodium phosphate monobasic	130.420
MICROELEMENTS	
Boric acid	3.000
Cobalt chloride hexahydrate	0.025
Copper sulphate pentahydrate	0.025
EDTA disodium salt dihydrate	37.300
Ferrous sulphate heptahydrate	27.800
Manganese sulphate monohydrate	10.000
Molybdic acid (sodium salt)	0.213

Potassium Iodide	0.750
Zinc sulphate heptahydrate	2.000
VITAMINS	
myo-Inositol	100.000
Nicotinic acid (free acid)	1.000
Pyridoxine HCl	1.000
Thiamine hydrochloride	10.000
CARBOHYDRATE	
Sucrose	20000.000
GELLING AGENT	
Agar	8000.000
Total(gms/litre)	31.2

Material required but not provided :

- Autoclaved distilled water
- Plant growth regulators
- 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.
- 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.

Quality Control:

Appearance

White to off-white, homogenous, free flowing powder

Solubility

31.2 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 ± 0.5

pH at 25°C

5.00 - 6.00

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 $22^\circ\text{C} \pm 2^\circ\text{C}$ 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.

Revision : 01 / 2017

Disclaimer :

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