



Technical Data Sheet

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CHU (N6) Medium (With Vitamins and Sucrose Without Agar)

RDM-CN-01

Principle

CHU (N6) Medium has been developed for in vitro anther culture of *Oryza sativa*, family *Graminae*. The formulation is composed of macronutrients, micronutrients, carbon sources, amino acid and vitamins. The macronutrients contain essential elements like nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulphur (S) for satisfactory growth and morphogenesis. The micronutrients include iron (Fe), manganese (Mn), zinc (Zn), boron (B), copper (Cu) and molybdenum (Mo) required for the growth of plant tissue and cells. The carbohydrates are used as carbon sources and energy source. Sucrose was reported to act as morphogenetic trigger in the formation of auxiliary buds and branching of adventitious root. Amino acid is used for optimal growth, they are important for establishing cultures of cells and protoplasts. Amino acids provide plant cells with a source of nitrogen that is easily assimilated by tissues and cells faster than inorganic nitrogen sources. Vitamins are required for normal growth and development of plant cells; they are required by plants as catalysts in various metabolic processes. The commonly used vitamins are: thiamin (B1), nicotinic acid and pyridoxine (B6). Thiamin is necessarily required by all cells for growth. Nicotinic acid and pyridoxine, however not essential for cell growth of many species, they are often added to culture media.

Use: Recommendation for the in vitro anther culture of *Oryza sativa*, family *Graminae*.

Contents*

Ingredients

Ingredients	mg/l	Ingredients	mg/l
MACROELEMENTS		VITAMINS	
Nitrate source#	463.000	Nicotinic acid	0.500
Calcium chloride	125.330	Pyridoxine HCl	0.500
Magnesium sulphate	90.370	Thiamine HCl	1.000
Potassium nitrate	2830.000	CARBOHYDRATE	
Potassium phosphate monobasic	400.000	Sucrose	20000.000
MICROELEMENTS		AMINO ACID	
Boric acid	1.600	Glycine	2.000
EDTA disodium salt	37.300		
Ferrous sulphate	27.800	Total	23.98 g/l
Manganese sulphate	3.330		
Potassium iodide	0.800		
Zinc sulphate	1.500		

* Formula adjusted for optimum performance and parameters

Equivalent to ammonium nitrate

Directions: Dissolve 24.00 gram in 750 ml distilled water mix well to dissolve all the ingredients. Add Heat stable ingredients & make up the volume to 1000 ml. Adjust the pH 5.75 $\pm$ 0.2 using 1N HCl/NaOH. Boil to dissolve the gelling agar completely. Sterilize by autoclaving at 15 lbs pressure (121 °C) for 15 min. Cool to 40°C for addition of heat labile ingredient. Aseptically dispense the medium in desired.

Precautions to be taken

These plant tissue culture media are intended for the in-vitro use only. All the handling, experiments, storage, and discarding should be performed with the help of skilled and knowledgeable technicians and as per the established guidelines. The material should be disposed only after proper sterilization by autoclaving. Please go through the MSDS of the media to avoid any accidents or in emergency.

Performance and Evaluation

The expected performance of the medium is liable to use as per the direction on the label when stored at optimum conditions and within expiry date.

Quality Control

Appearance	Off-white homogenous, free flowing powder.
Solubility	24.00 g/l soluble in distilled water
pH at 25 °C	5.75 ±0.2
Color and clarity of ready medium	Colorless clear solution
Growth promoting properties	Assessed by providing plant cultures with relative humidity of about 60%±2%, temperature 22°C±2°C and photoperiod of about 16:8.
Cultural Response	Plant species showed actively growing callus and shoots with no structural, necrotic and toxic deformity

Storage and Shelf Life

Hygroscopic; keep container tightly closed. Store in cool dry place. Preferably, entire content of each package should be used immediately after opening. Use before the expiry date.

Disposal: To avoid the contamination or propagation of any hazardous microbes the used, unusable or modified preparation of this product must be disposed after autoclaving after completion of task.

Reference

1. Kalyan Kumar De (2008), *An introduction to Plant Tissue Culture*, 7th Ed., Darjeeling Govt. College, Darjeeling, India. New Central Book Agency(P) Ltd. Kolkata, India.
2. Narayanaswamy S (2005). *Plant cell and tissue Culture*, Tata Mc-Graw Hill publishing Co. Ltd. 6th reprint.
3. Roberta H.Smith .(2005), *Plant Tissue Culture-Techniques and Experiments*, 2nd Ed., Texas A&M University, College Station, Texas.Academic Press. An Imprint of Elsevier, San Diego California,U.S.A

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CHAITANYA AGRO BIOTECH PVT. LTD.

An ISO 11133:2014, ISO 13485:2016, ISO 9001:2015 CE, CIN NO.: U24210MH1995PTC095220S,
S. No. 120/2, Laxmi Nagar, Umbarnala Road, Malkapur-443101, Dist.: Buldana (M.S.) India. Customer Care +91-8669083859
rdmsales@chaitanyagroupindia.com, mkt.cabt@chaitanyagroupindia.com, www.chaitanyagroupindia.com