MICROPROPAGATION OF AN ENDANGERED MEDICINAL HERB *OCIMUM CITRIODORUM* VIS.

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Abstract: An efficient protocol has been developed for rapid micropropagation of *Ocimum citriodorum* Vis., an endangered medicinal herb. The cotyledons were excised from the *in vitro* germinating seedlings and used as explants for the present study. The explants yielded the highest frequency of 87.49% shoot regeneration with an average shoot length of 4.98 cm on Murashige and Skoog (MS) medium supplemented with 1 mg l^{-1} 6- benzylamino purine (BAP) + 0.1 mg l^{-1} napthalene acetic acid (NAA) + 500 mg l^{-1} casein hydrolysate (CH) + 25 mg l^{-1} adenine sulphate (AS). Alteration from the optimal concentration of BAP resulted in the formation of callus. Regenerated microshoots were separated and rooted on MS medium containing NAA (0.5 mg l^{-1}). Well-developed complete plantlets were transferred onto plastic cups containing sterile soil and humus (1:1). Subsequently the acclimatized plantlets were successfully grown in garden. The regenerated plants were morphologically identical and exhibited similar growth characteristics as compared to the donor plants. Cytological studies of the regenerants revealed no change in chromosome numbers. Thus, regeneration protocol demonstrated in the present study provides a basis for the germplasm conservation and investigation of its medicinally active constituents.

Keywords: Cotyledonary explant, cytology, histological observations, *ocimum citriodorum*

REFERENCES

Asghari, F.; Hossieni, B.; Hossani, A. and Habib S. (2012). Effect of explants source and different hormonal combinations on direct regeneration of basil plants (*O. basilicum L.*). *Australian Journal of Agricultural Engineering*, 3 (1): 12-17.

Begum, F.; Amin, N. and Azad A.K. (2000). *In vitro* clonal propagation of holy basil- *Ocimum sanctum* L. *Plant Tissue Culture* ,10: 31-37.

Burch, L.R. and Stuchbury, T. (1987). Activity and distribution of enzymes that interconvert purine bases, ribosides and ribotides in the tomato plant and possible implications in cytokinin metabolism. *Physiologia Plantarum*, 69: 283- 288.

Chakravarty, B. and Sen, S. (1992). Chromosome and nuclear DNA in regenerants of *Scillaindica* (Roxb.) Baker derived from two explants sources. *Cytologia*, 57: 41-46.

Dibax, R.; Quisen, R.C.; Bona, C. and Quoirin, M. (2010). Plant regeneration from cotyledonary explants of *Eucalyptus camaldulensis* Dehn and histological study of organogenesis *in vitro*. *Brazilian Archives of Biology and Technology*, 53 (2) 311-318.

Epriliati, I. and Ginjom, I.R. (2012) Bioavailability of phytochemicals, *Phytochemicals - A Global Perspective of Their Role in Nutrition and Health*, Dr Venketeshwer Rao (Ed.). InTech.

Francis, D. and Sorell, D.A. (2001). The interphase between the cell cycle and plant growth regulators- a mini review. *Plant Growth Regulators*, 33: 1-12.

Gopi, C.; Sekhar, Y.N. and Ponmurugan, P. (2006). *In vitro* multiplication of *Ocimum gratissimum L.* through direct regeneration. *African Journal of Biotechnology*, 5(9):723-726.

Janarthanam, B. and Sumathi, E. (2012). Plantlet regeneration from nodal explants of *Ocimum citriodorum Vis. Bangladesh Journal of Scientific and Industrial Research*, 47(4): 433-436.

Kusmapudi, S.; Selvakkumar, C.; Senthil, A.K. and Beddireddi, S.L. (2010). *In vitro* root culture of *O. sanctum L.* and evaluation of its free radical scavenging activity. *Plant Cell, Tissue and Organ Culture*, 101: 105-109.

Mendoza, M.G. and Kaeppler, H.F. (2002). Auxin and sugar effects on callus induction and plant regeneration frequencies from mature embryos of Wheat (*Triticum aestivum L.*) *In Vitro Cellular and Developmental Biology- Plant*, 38: 39- 45.

Miel, F.B.; Guignard, J.L.; Bury, M. and Agier, C. (1985). Glutamine as an active component of casein hydrolysate: Its balancing effect on plant cells cultured in phosphorus deficient medium. *Plant Cell Report*, 4: 161- 163.

Mohammad, A.; Moieni, A. and Badi, H.N. (2013). Morphological, physiological, cytological and phytochemical studies in diploid and colchicines-induced tetraploid plants of *Echinacea purpurea* (L.). *Acta Physiologiae Plantarum*, 35: 2075-2083.

Murashige, T. (1974). Plant propagation through tissue culture. *Annual Review of Plant Physiology*, 25:135–166.

Murashige, T. and Skoog F. (1962). A revised for rapid growth and bioassays with tobacco tissue cultures. *Physiol. Plant*, 15: 473- 497.

Nandgopal, S. and Kumari, B.D.R. (2007). Effectiveness of auxin induced *in vitro* culture in chicory. *Journal of Central European Agriculture*, 8 (1): 73-80.

Narayanswamy, S. (1977). Regeneration of plants from tissue cultures. In: Applied and Fundamental Aspects of Plant Cell Tissue and Organ Culture, *Ornamental Species*, Volume 5, McGraw-hill Publishing Company, USA, pp 3-9.

Paton, A. and Putievsky, (1996). Taxonomic problems and cytotaxonomic relationships between and within varieties of *Ocimum basilicum* and related species (Labiatae). *Kew Bulletin,* 51: 509-524.

Pattnaik, S. and Chand, P.K., (1996). In vitro propagation of the medicinal herbs O. americanum L. syn. O. canum Sims. (hoary basil) and O. sanctum L. (holy basil). *Plant Cell Reports,* 15: 846- 850.

Radic, S.; Prolic, M.; Pavlica, M. and Pevalek, K.B. (2005). Cytogenetic stability of *Centaurea ragusina* long term culture. *Plant Cell Tissue and Organ Culture,* 82: 343-348.

Saha, S.; Sengupta, C. and Ghosh P. (2014). Molecular and phytochemical analyses to assess genetic stability in alginate- encapsulated microshoots of *O. gratissimum* L. following *in vitro* storage, *Nucleus*, DOI 10.1007/s13237-014-0107-y

Sahoo, Y.; Pattnaik, S.K. and Chand, P.K. (1997). *In Vitro* clonal propagation of aromatic medicinal herb *Ocimum basilicum* (Sweet Basil) by axillary shoot proliferation. *In Vitro Cellular Developmental Biology- Plant,* 33: 293-296.

Sahu, A.R.; Rath, S.C. and Panigrahi, J. (2013). *In vitro* propagation of *Aerva lanata* (L.) Juss. ex Schult. through organogenesis, *Indian Journal of Biotechnology,* 12: 260-264.

Shahzad, A.; Faisal, M.; Ahmad, N.; Anis, M.; Alatar, A. and Hend, A.A. (2012). An efficient system for *in vitro* multiplication of *Ocimum basilicum* through node culture. *African Journal of Biotechnology*, 11 (22): 6055- 6059.

Shibli, R.A.; Ajlouni, M. M.; Jaradat, A.; Aljanabi, S. and Shatnawi, M. (1997). Micropropagation in wild pear (*Pyrussyrica*), *Scientia Horticulturae,* 68: 237-242.

Siddiqui, I.; and Anis, M. (2009). Morphogenic response of the alginate encapsulated nodal segment and antioxidative enzymes analysis during acclimatization of *O. basilicm L. Jouranl of Crop Science and Biotechnology*, 12 (4): 233-238.

Singh, N.K. and Sehgal, C.B. (1999). Micropropagation of Holy Basil (*Ocimum sanctum* Linn.) from young inflorences of mature plants. Plant Growth Regulation, 29: 161- 166

Stanko, K.C.; Orli, S.; Politeo, O.; Strikić, F.; Kolak, I. and Satovic, Z. (2010a). Composition and antibacterial activities of essential oils of seven *Ocimum* taxa. *Food Chemistry*, 119: 196–201.

Stanko, K.C.; Liber, Z.; Besendorfer, V.; Javornik, B.; Bohanec, B.; Kolka, I. and Satovic, Z. (2010b) Genetic relations among basil taxa (*Ocimum* L.) based on molecular markers, nuclear DNA content and chromosome number. *Plant Systematics and Evolution*, 285: 13-22.

Sudha, C.G. and Seeni, S. (2001). Establishment and analysis of fast growing normal root culture of *Decalepis arayalpathra*, a rare endemic medicinal plant. *Current Science*, 81: 371-374.

Sulistiarini, D. (1999). *Ocimum gratissimum L.* In: Oyen, LPA, NaguyenXuan Dung (eds.) Plant Resource of South East Asia, No. 19: *Essential Oils Plants.* Prosea Foundation, Bogor, Indonesia, pp. 140-142.

Thom, M.A.; Maretzki, K.E. and Sakai, W.S. (1981). Nutrient uptake and accumulation by sugarcane cell culture in relation to growth cycle. *Plant Cell Tissue and Organ Culture,* 1:3–14.

Tripathi, A. (2011). Anticarcinogenic potential of some medicinal plants, *The Botanica*, 59 (61): 169-175.

Venugopal, L.G. and Rao, A.P. (2011). *In vitro* propagation and callus induction of endangered medicinal herb *Ocimum citriodorus*. World Congress on Biotechnology, *Biotechnology*-2011.

Wang, Q.M.; Gao, F.Z.; Gao, X.; Zou, F.U.; Sui, X.; Wang, M.; Hui, Y.J. and Wang, L. (2012). Regeneration of *Clivia miniata* and assessment of clonal fidelity of plantlets. *Plant Cell Tissue and Organ Culture*, 109: 191-200.

Washida, D.; Shimomura, K.; Takido, M. and Kitanaka, S. (2004). Auxins affected ginsenoside production and growth of hairy roots in Panax hybrid. *Biological and Pharmaceutical Bulletin*, 27: 657-660.