

PLANT REGENERATION FROM SEED CULTURE OF *CUCUMIS DIPSACEUS* Ehrenb.

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Abstract: The present paper focuses on *in vitro* regeneration method for the tendril climber and economic important medicinal plant *Cucumis dipsaceus* Ehrenb. Was established. The seeds were treated with GA₃ produced *in vitro* grown seedlings within ten days intervals. Among the different treatments water as well as GA₃ induced the highest percentage 86% maturation and germination of embryos. The *in vitro* generated shoots turned brown at base and gradually turned black resulting in reduced plantlets rooting. Addition pinch of charcoal to the rooting medium resulted in increased rooting percentage. The rooted plants, transferred to a soil mix had a survival rate of 91% in the green house. All the plants that were transferred to wetlands survived.

Key Words: *In vitro*, medicinal plant, hardening

INTRODUCTION

Cucumis dipsaceus Ehrenb. Ex Spach is a species of flowering plant belonging to the family Cucurbitaceae. It has its origin in Ethiopia and is known by several common names like teasel gourd, Arabian cucumber, hedgehog, pepino-diablito, concombres porc-epic etc. Usually *Cucumis dipsaceus* are consumed as a leafy vegetable (Verdcourt and Trump, 1969), its fruit juice is topically applied to prevent hair loss (Rainer and Ashley, 2010). However, being a fruit growing in wild no attempt has been taken to evaluate the parameters which could support its use as vegetable. In India, herbal origin is used for various diseases and Indian folk medicine is used as prescriptions for therapeutic purposes such as wounds, inflammation, skin infection, leprosy, diarrhea, scabies, venereal disease, ulcers, snake bites (Hemamalini *et al.*, 2013). The country ranks sixth for harboring the largest number of threatened plant species. The IUCN Red list indicates that 91% plant species are threatened due to habitat loss and degradation (Hilton, 2001). Approximately 70% of India's medicinal plants are found in the tropical regions (Negi *et al.*, 2007). Conservation of bio- wealth and genetic resources is essential for ensuring future food of the Nation. In order to utilize plant species in a judicious way, a thorough knowledge of ecological needs of endangered and endemic plants is a prerequisite (Hawkes, 1976). This paper reports the protocol for improvement of micro propagation of this plant using seeds as the explants.

MATERIALS AND METHODS

The fruits were collected during the month of November 2014. The collected plant material was identified and their authenticity was confirmed by comparing the voucher specimen at the herbarium of Botanical survey of India (BSI), Southern circle Coimbatore, Tamil Nadu (No.BSI/SRC/5/23/2011-12/Tech-1466). Freshly collected fruits were cleaned to remove adhering dust and then dried under shade. The dried sample was powdered and used for further studies. The seeds were washed thoroughly in running tap water followed by glass distilled water. Surface sterilization was done inside laminar air flow chamber with mercuric chloride (0.1%) for 5 minutes.

The followed by washing with sterile distilled water 3 - 4 times. The various treatments including GA₃ treatment used for the seed germination studies. The cultures were incubated at $26 \pm 2^{\circ}\text{C}$ and photoperiod of 15 h at light intensity of 3000 lux provided by cool white fluorescent tubes.



Fig 1. Seeds development on the medicinal plant *Cucumis dipsaceus*

RESULT AND DISCUSSION

The present studies on *C. dipsaceus* were carried out to identify the factors for in vitro studies of this medicinally important plant which is over exploited for medicinal purposes. The studied explants show encouraging results under the influence of GA₃ and growth regulators. The explants sterilized with 0.1% HgCl₂ and different treatments shows variation in plantlet production. An initial dark period for 48 hours was found to be limiting factor for the establishment of cultures during present study. These cultures when exposed to fluorescent light of various photo period during the first 48 hours of incubation turn albino lost the green color. Some observations have been made Rukhsana, 2004. This suggests the inhibitory role that light has on the in vitro culture of many other angiosperm species (Razdan, 2001).

The plantlet production from seed explants on GA₃ medium within ten days of inoculation. However, in case of different growth regulators the response for production of plantlets. Mature showed maximum germination and they grew well on GA₃ medium while immature seeds showed abnormal growth with subsequent seedling death. However, the present survival of control plantlets was good 90% and when they were transferred to field they grew well. Similar result was observed in *Gnetum ula* (Siva Subramanian *et al.*, 2005) *Aeschynomene indica* (Indira *et al.*, 2012) and *Ginkgo biloba* (Aseesh *et al.*, 2011). The resurgence of public interest in plant based medicine tied with the rapid expansion of pharmaceutical industries dictated an increased demand of medicinal plants, leading to exploitation that threatened the survival of many medicinal plants. Plant tissue culture provides propagation of plants which are rare or economically important, so that industrial demands could be met without disturbing the natural population of plants. The present investigation showed that a sufficient number of healthy plantlets of *C. dipsaceus* with 90% survival may be produced by applying GA₃ treatment within short period of time which could be an easy cost effective, rapid and promising method of macro propagation.

Table 1: Effect of pre-treatments on the germination behavior of seeds

Treatments	Duration	Germination Percentage	Days for Germination
IBA	2 minutes	55	20
	5 minutes	67	19
	10 minutes	59	22
NAA	2 minutes	69	17
	5 minutes	62	22
	10 minutes	64	19
IAA	2 minutes	59	25
	5 minutes	62	28
	10 minutes	57	32
Boiling Water	2 minutes	46	21
	5 minutes	57	27
	10 minutes	43	30
GA₃	2 minutes	77	16
	5 minutes	86	10
	10 minutes	82	21
Water medium	2 minutes	81	18
	5 minutes	86	14
	10 minutes	79	24
Acid treatment	2 minutes	23	21
	5 minutes	16	32
	10 minutes	11	27

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REFERENCES

1. **Aseesh Pandey., Sushma T. and Dinesh G.** (2011). Role of auxin on adventitious root formation and subsequent growth of cutting raised plantlets of *Ginkgo biloba* L. *Inter. Jour. Biodiver. Conser.* 3 (4): 142 – 146.
2. **Flora of Tamil Nadu, India 1996.** Edited by Henry AN and Nair N C Vol. I p. 91. .
3. **Indira P. L. Siva Subramanian S., Dyaku M., Anitha M. Karthika Devi R. Pothammal P., Suresh D., renuka A.**
4. **Balamani R. Arulmathi V. and Kanimozhi R.** 2012. Plant regeneration from seed culture of *Aeschynomene indica* L. *Jour. Bio. Sci. Resear.* 3 (1): 34 – 36.

5. **Rainer W. Bussmann and Ashley G. 2010.** *Medicinal plants used in Northern Peru for reproductive problems and female health. Jour. Ethnobiology and Ethnomedicine 6: 30 – 37.*
6. **Rukhsana A. 2004.** *In vitro studies on Viola odorata l. Dissertation submitted to the University of Kashmir.*
7. **Murashige T. and Skoog F.(1962).** *A revised medium for rapid growth and bioassays of tobacco tissue cultures. Physiol.Pl.15: 473-497.*
8. **Negi V.M., Dutt N and Chauhan N.S. (2007).** *Threatened medicinal and aromatic plants of sangla valley in Himachal Himalaya causes and remedies, International Journ.Ecology and Environmental Sciences, 33(2-3) : 219- 233.*
9. **Siva Subramanian S., Louis Jesudass L., Manickam V., Seeni S. and Harikrishnan S. 2005.** *Abnormal seedlings in Gnetum ula. Asian Jour. Microbio. Biotech. Envir. Sci. 9 (3): 715 – 716.*
10. **Verdcourt, B. and Trump, E. C. 1969.** *Common poisonous plants of East Africa. Collins, London..*