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Research paper

Successful Hardening of Anther-derived Plants to field and Morphological Variations in Snapdragon

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Abstract

Plants produced through anther cultures were transferred from culture room to glass house and ultimately in the field in Snapdragon (Antirrhinum majus, family Scrophulariaceae). Different methods of hardening were studied to have maximum survival of plants. After successful transfer to the field, plants produced flower and pollen grains. There were significant variations in the morphology , shape and size of leaves and flowers, suggesting them as somaclonal variants.

Key words: Snapdragon, somaclonal variations, anther, *Antirrhinum majus*

Introduction

Antirrhinum majus L. (Snapdragon) is an ornamental plant belonging to the Family Scrophulariaceae. It is valued due to its multi-coloured flowers. It has been widely studied in Genetics to study the phenomenon of Partial Dominance as well as Transposons (Jumping Genes). In this, we have produced plants from anther-derived callus masses. Plants produced in culture tubes are very delicate and highly susceptible to harsh environmental conditions of *ex vitro*. Their subsequent transfer to the field requires special attention and they are made hard in a gradual manner. Various methods of hardening known till date involve transfer of plants in the soil in small earthenware pots containing soil, covered with polythene bags to stop dehydration and kept in culture room under controlled environmental conditions for a few days before transferring them to glass house in the Botanical Garden. In several cases plants did not survive till flowering stage due to lack of their proper hardening . The transfer of plants from culture room to glass house requires gradual transfer so that they are well acclimated with the outside new environment. Deb and Imchem (2010) have discussed an efficient hardening technique for tissue culture raised plants. In another report a detailed review has been given regarding acclimatization of tissue-cultured plants (Hazarika *et al.*, 2003).

Material and Methods

Plants of *Antirrhinum* were obtained from anther cultures as per experimental details already given (Gautam et al., 1993). For hardening of plants following steps were followed.

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(a) Plants in culture tubes - Plants in flasks - In vermiculite (Culture room) - Garden soil (In culture room) - Glass house

(b) Plants in flask - plants in pots (garden soil) - Glass house

(c) Plants in flask - in pots (garden soil) - in culture room- in Glass house

(d) Plants in flask - in pots in Garden soils - in laboratory- in Glass house

Culture room conditions were the same as described earlier in our reports (Gautam *et al*., 1993).The temperature in the culture room, Laboratory and Glass house was maintained between 25-26^{0}C. The morphological characters of anther-derived plants were compared with plants developed from seeds in the Botanical Garden of the Department.

Results and Discussion

Plants from anther cultures have been produced in several species with the objective of haploid production. Most of the plants responding to anther cultures belonged to the Family Solanaceae. However androgenic haploids within the family Scrophulariaceae have been produced in few species such as *Digitalis purpurea* (Gorduan and Spix, 1975) and *Digitalis obscura* (Perez-Bermuda *et al*.,1985) that too at low frequencies. We obtained a high frequency of multiple shoots and roots from anther cultures. The transfer of plants from the culture room to the glass house was also successful, irrespective of the method employed(Fig.1). All plants grew well, were healthy and also flowered (Fig.2). In all fifteen plants flowered successfully. The morphology of anther-derived plants was significantly different from field grown (control) plants. In general, experimental plants were shorter in height, bushy in appearance and with weak and trailing branches. The size of leaves, flowers and anthers were also smaller (Fig.3). Anther squashes of *in vitro* raised plants showed that 89% of the pollen were smaller in size, empty and non-viable as compared to the control pollen which showed 85% viability as tested by FDA or Tetrazolium chloride test. Our observations showed these plants were somaclones and can be used for further basic and applied studies.

Summary and Conclusion

Anther derived plants can be successfully transferred to soil and subsequently to glass houses. Various complicated steps of hardening, involving sequential transfer from peat to vermiculite, to silica sand and then finally to soil are not necessarily required for *Antirrhinum majus* shoots. The rate of survival of anther -derived plants in glasshouse conditions is satisfactory. Most of the anther-derived plants grew normally, produced flowers but were sterile and did not set seeds. External morphology of anther-derived plants was different from that of the control. Their height, size of leaves, flower and anthers were less than those of control.

References

1. Corduan, G. and Spix, C. 1975. Haploid callus and regeneration of plants from anthers of *Digitalis purpurea* L. Planta 124: 1-11.
2. Deb, C.R. and Imchem. 2010. An efficient *in vitro* hardening technique of tissue culture raised plants. Biotechnology 9: 79-83.
3. Gautam, V.K. ; Nanda, K. and Gupta, S.C. 1993. Morphological variants from anther- derived

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callus of *Antirrhinum majus* L. Int. Bot. Cong., Tokyo, pp.350.

4. Hazarika, B. N. 2003. Acclimatization of tissue cultured plants. Curr.Sci. 85 : 1704-1712.

5. Perez- Bermudez,P.; Cornejo, M. J. and Segura J. 1985. Pollen plant formation from anther cultures of *Digitalis obscura* L. Pl. Cell Tissue Organ Cult. 5: 63-68.

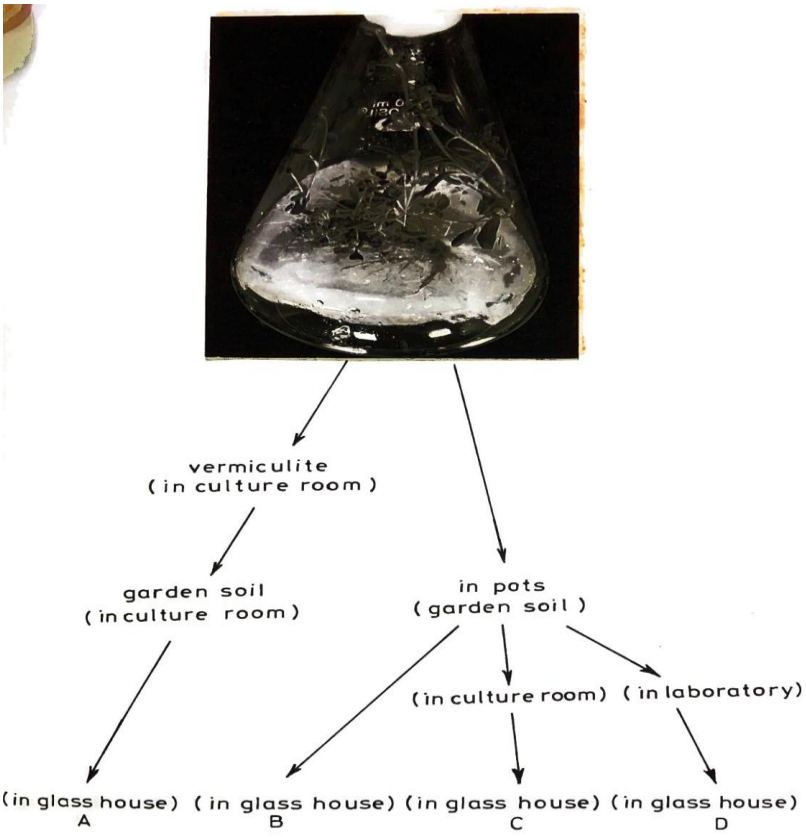

Fig.1.Transfer of anther-derived plants of *Antirrhinum majus* to field through different steps.

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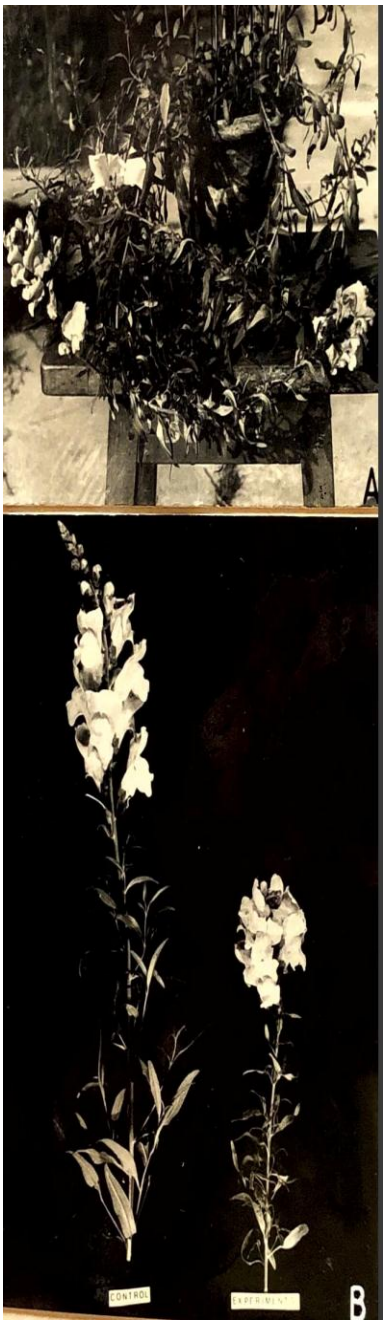

Fig.2.Morphology of *in vivo* grown anther derived plants of *Antirrhinum majus*. A. Flowering anther- derived plants. B. Comparison of twigs of *in vivo* grown control (taller) and anther-derived plants(shorter).

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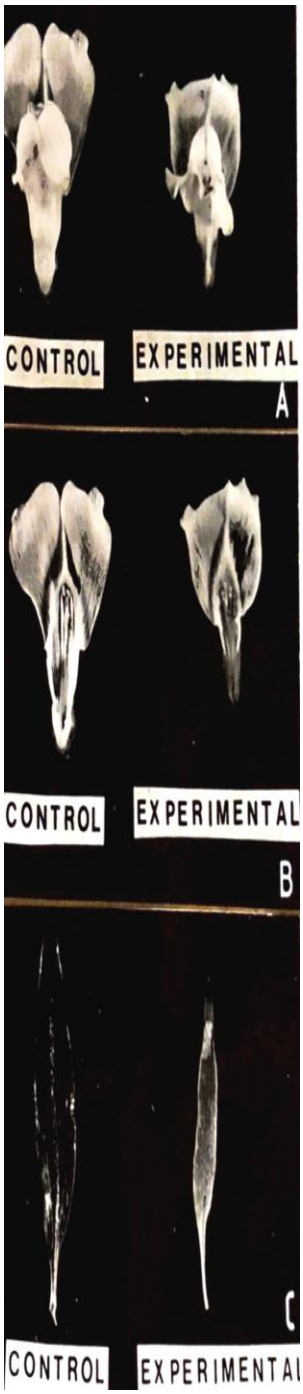

Fig.3.Comparative morphology of *in vivo* grown anther-derived and field grown (control) plants. A-C. Comparative size differences in the flower, anthers and leaves,respectively.