INTERNATIONAL JOURNAL OF PLANT, ANIMAL AND ENVIRONMENTAL SCIENCES
IJPAES
Volume-4, Issue-3, July-Sept-2014
ISSN 2231-4490

Coden : IJPAES www.ijpaes.com
Received: 20th June-2014 *Revised: 16th July-2014* *Accepted: 17th July-2014*
Research article

AN EFFICIENT MICROPROPAGATION PROTOCOL FOR *GERBERA JAMESONII* BOLUS FROM FLOWER BUDS

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ABSTRACT: An highly efficient micropropagation protocol for commercial multiplication of gerbera was developed with unopened flower buds as explants. Media for culture establishment, shoot regeneration, propagule multiplication, elongation, rooting and hardening were standardized. For culture establishment and propagule multiplication MS medium supplemented with BAP 3mg/l. + NAA 0.1 mg/l was found good. Shoots were elongated in MS medium supplemented with NAA 0.1mg/l. For root morphogenesis, MS medium supplemented with IBA 1mg/l was effective with more number of roots and longer roots. Plantlets were successfully hardened in sand soil coir pith mixture. Response was genotype dependent in all stages of *in vitro* propagation.
Key words: Gerbera, Micropropagation, flower bud explants

INTRODUCTION

Gerbera jamesonii Bolus commonly known as African daisy is grown commercially mainly for cut flowers. The long vase life, ability to rehydrate after long transportation make it extremely popular as a cut flower. Gerbera flowers are now- a -days indispensable items for floral arrangements and decorations for marriages and other ceremonial functions.

Gerbera is generally propagated by division of suckers or clumps, but multiplication rate is very slow and production of large number uniform propagules still remains as a major problem in commercial cultivation. Moreover, new varieties/ hybrids introduced every year are to be multiplied in large numbers for commercial cultivation.

Hence studies on micropropagation was carried out at Centre for Plant Biotechnology and Molecular Biology, College of Horticulture, Kerala Agricultural University during 2011-13 to develop an efficient micropropagation protocol for mass multiplication of Gerbera.

MATERIALS AND METHODS

Un opened flower buds were selected as explants from two gerbera selections procured from AVT, Kochi, of which the selection Dubai is white flowered and Shania is red flowered.

The unopened flower buds with basal diameter 1.0-1.2cm were collected from source plants maintained in net house of Centre for plant Biotechnology and Molecular Biology, College of Horticulture, Kerala Agricultural University, Thrissur. The excised flower buds were washed in running tap water and pretreated with 0.1 percent Bavistin and one drop of detergent for ten minutes followed by washing in sterile distilled water. Further sterilisation with 0.1percent $HgCl_2$ for seven minutes was carried out in Laminar Air Flow Chamber followed by washing thrice in sterile distilled water. The outer bracts and sepals were removed from flower bud and it was made into six to eight longitudinal segments and each segment was inoculated to MS medium supplemented with BAP - NAA and BAP - IAA combinations.

In the culture establishment medium, shoot regeneration was also observed and cultures were sub cultured for propagule multiplication in the same medium at one month interval. For elongation of shoots, MS medium supplemented with a low concentration of NAA was found good. MS medium supplemented with a very low level of IBA produced very good *in vitro* roots. Rooted plantlets were acclimatized on sand soil and peat mixture.

RESULTS AND DISCUSSION

The age of flower buds (judged by size) was found to be a crucial factor for culture establishment and shoot regeneration, the optimum being unopened flower buds with basal diameter of 1.0-1.2 cm, the bigger and smaller buds were not giving response with respect to culture establishment and shoot regeneration. Genotypic difference was also observed in all stages of *in vitro* propagation (Table 1).

Table-1. Effectiveness of the micropropagation protocol in different Gerbera genotypes

Genotypes	Culture establishment & shoot regeneration (%)	No. of shoots regenerated / bud segment in culture establishment medium	No. of shoots proliferated /culture in second sub culture	Days taken for rooting	No. of roots initiated	Root length (cm)	Rooting (%)	Hardening survival (%)
Dubai (white flowered)	95.83	3.41	42.33	20.0	3.33	2.5	94.44	90.0
Shania (red flowered)	44.44	1.16	36.86	20.33	4.0	1.96	83.32	66.66
Mean	70.14	2.29	39.59	20.16	3.67	2.23	88.88	78.83
t value	7.39**	7.73**	2.02	0.25	1.01	4.44*	2.45	2.65*

** Significant at 1% level * Significant at 5% level

Of the various media tried for culture establishment, MS medium supplemented with BAP 3mg/L. + NAA 0.1 mg/L. gave establishment and shoot regeneration to the extent of 44.44 – 95.83 per cent in the two genotypes studied, white flowered type exhibited a higher establishment and shoot regeneration. The number of shoots regenerated in initial culture varied from 1.16 – 3.41 but when sub cultured in same media, the propagule multiplication was found very high in the two genotypes studied. However, white flowered genotype exhibited higher shoot proliferation (42.33) as compared to red (36.86) in second subculture cycle even though statistically they were on par. Shoos were elongated in MS medium supplemented with NAA 0.1 mg/l. For root morphogenesis, of the various auxins tried, MS medium supplemented with IBA 1mg/l produced good rooting percentage (88.88) with more number of roots and longer roots. Plantlets with well developed roots were planted out to potting medium consisting of soil, sand and coir pith. Hardening survival was also found genotype dependent and white flowered genotype recorded higher survival percentage (90) as compared to red flowered type.

Even though several explants like leaves [2,5], petioles [4] , capitulum [6], shoot tips [3,1] and flower buds [7] were tried for *in vitro* propagation of gerbera, successful protocols were reported only from shoot tip and flower buds.

As compared to shoot tips, the flower bud explants are very good option as they are non destructive with respect to further propagation of plants. In the protocol developed by Son et al (2011) [7] with flower bud explants also, high genotypic difference was observed for *in vitro* response. When present protocol is compared with the protocol developed by Son et al (2011) [7], the size of the flower bud and hormonal combinations are different in the culture establishment phase and in rooting phase. Also, a separate elongation phase with a lower level of auxin and without cytokinin standardised in the present protocol was also found beneficial for getting better rooting. Such variations in hormonal combinations at various stages of *in vitro* propagation might be due to the difference in genotypes investigated in the present study.

CONCLUSION

An highly efficient protocol for commercial micropropagation of gerbera was thus developed with unopened flower buds as explants. Genotypic differences were observed in all stages of *in vitro* propagation. The performance at all stages of micropropagation was found good for the white flowered genotype which is a highlight of the present protocol as white is the predominant gerbera flower traded and used in all auspicious occasions and decorations.

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