

## PLANTLET REGENERATION FROM COTYLEDONARY CULTURES OF A FOREST TREE *PRUNUS CERASOIDES* D. DON

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In recent years, cotyledons have formed a convenient explant for the initiation of cultures (Lakshmi Sita, 1979; Knittel *et al.* 1991) and they have been found to possess high potential for plant regeneration without the intervening callus phase. Whole plants are easily regenerated from these shoots. Cotyledons have the advantage of being easily and quickly available and do not demand much effort for their excision. We describe here an efficient method for the fast regeneration of Himalayan cherry from cotyledons of aseptically germinated seeds of *Prunus cerasoides* D. Don, a multipurpose tree species of Temperate Himalayas.

Nuts of *P. cerasoides* D. Don were collected around March-April, washed with teepol and water thoroughly. The hard nut covering was removed by gently hammering to expose the seeds. These seeds were sterilised in 70% ethanol for 1 min. followed by 4% sodium hypochloride for 2 min. The sterile seeds were imbibed in water for 6 hours in darkness and then germinated on solidified, half strength Murashige and Skoog (1962) (MS) medium containing 6% sucrose. Seeds were kept in darkness at  $25 \pm 2^\circ\text{C}$  for three days before transfer to light (2000 lux). Cotyledons were excised from the 7 day old plantlets and divided into two halves basal and distal and three each of basal and distal parts of the cotyledons were placed in each

flask containing MS media with hormonal combination of 6-benzyl-amino-purine (BAP) and  $\alpha$ -naphthalene acetic acid (NAA), and indole-3-acetic acid (IAA) and Kinetin (Kn) in the range of 0.5-10mg/l. The cultures were incubated at  $25 \pm 2^\circ\text{C}$  at 14 hr. photoperiod duration using cool white fluorescent light at 2000 lux intensity. The whole experiment was repeated thrice and the observations recorded.

The cut surface of the basal (proximal to the embryo) part of the cotyledon showed slight swellings in various concentrations of NAA and BAP combinations (Table-I), the

**Table-1:** Mean number of shoots from the basal and distal (in brackets) surfaces of the cotyledons of *P. cerasoides* D. Don in response to various concentrations of BAP and NAA (mg/l)

NAA	BAP			
	0.5	2.5	5	10
0.5	Nil	2(2)	6(3)	5(2)
2.5	Nil	Nil	3(1)	3
5	Nil	Nil	Nil	1
10	Nil	Nil	Nil	Nil

best being 0.5 mg/1 NAA and 5 mg/1 BAP and at the end of around 4 weeks a cluster of small shoots (6-8) originated from these surfaces. The distal part too showed similar responses from the cut surface but the frequency of shoots produced were very less. In IAA and Kn combination profuse nodular red-pigmented callus was observed (best in medium with 2.5 mg/1 IAA and 0.5 mg/1 Kn) but no regeneration occurred. The 8-10 weeks old shoots were excised and rooted in agar-solidified half-strength MS medium containing 6% sucrose. Plantlets with well developed roots were transferred and kept in the green house, care being taken to spray water daily. This potentiality of

the basal part of the cotyledons to regenerate a large number of plantlets could be useful for the large-scale micropropagation of this species to ensure year round supply of this material.

## References

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NAA (mg/l)	BAP	
	5	10
0.5	10 (5)	15 (10)
2.5	15 (10)	20 (15)
5	20 (15)	25 (20)
10	25 (20)	30 (25)