Table 1: Effect of BAP and IAA for multiplication of different explant

Explant	Conc. of growth regulator (mg/L)		Shoot length (Mean± SE)	% of shoot formation
	BAP	IAA		
apical shoot tip	0.5	0.2	1.88± 0.073	30
	1.0	0.2	2.64± 0.129	32
	1.5	0.2	5.52± 0.122	37
	2.0	0.2	7.90± 0.149	51
	2.5	0.2	9.16± 0.160	49
	3.0	0.2	8.24± 0.172	47
Axillary bud	0.5	0.2	6.70± 0.130	35
	1.0	0.2	8.04± 0.143	37
	1.5	0.2	8.14± 0.140	44
	2.0	0.2	9.62± 0.139	52
	2.5	0.2	8.76± 0.214	50
	3.0	0.2	7.82± 0.149	49

*After 25 days mean ± SE of 5 replicate

Effect of BAP shoots multiplication

The present investigation it was confined that explants viz. apical shoot, axillary bud and nodal explants was found effective for the development and multiple shoots formation. The two Cytokininsviz. BAP and KIN were tested. BAP was more effective than KIN for multiplication. MS media containing 3% sucrose, 3 mg/L Clerigel and different concentration of BAP 10.5, 1.0, 1.5, 2.0, 2.5, 3.0 mg/l alone and with IBA 0.2, 0.4, 0.6, 0.8, 1.0 mg/L, were tested. Concentration of BAP along with NAA 0.2, 0.4, 0.6, 0.8, 1.0 mg/L gives average percentage of multiple shoots in *Ceropegia*.(Fig.1) Maximum number of multiple shoot wererecorded in case BAP 2.0 mg/L, with combination IBA 0.2 mg/L.(Fig.2, table 1)

Explant such as apical shoot and axillary bud were prominent to show results when inoculated on MS medium supplement with 3% sucrose, 2.5% Clerigel. Maximum shoot length and multiple shoot formation was recorded at 1.6 mg/L BAP in combination with 0.2 mg/L IBA. Various concentrations of IAA were added into the MS medium to achieve rhizogenesis. Better *in vitro* rhizogenesis was achieved with 0.5 mg/lit IAA. Plants were hardened and planted in soil for field trails. In vitro regenerated plants had shown 75 % viability.

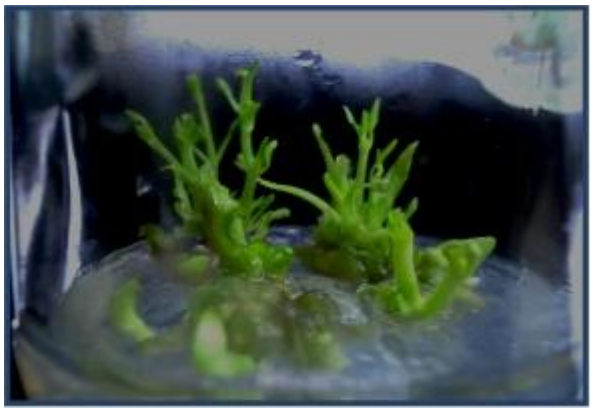

Figure 1: Multiple shoots formation along with callus

Figure 2: Multiple shoot formation along with tubers

Formation of tubers was recorded during growth of the cultures. The tubers are hard, light green in color and after subculture give rise to new plantlets. Similar results were recorded for callus and shoot multiplication using various explants in different plants *like Tylophora indica (*Gupta et al. 2010, Shah and Kapoor,1976*)*. Proliferating shoot cultures was established by repeatedly sub culturing the mother explants on the hormone free medium. Repeated sub-culturing was said to be one of the methods of maintaining juvenility (Choudhary and Jha. 2004). In the present work highest number of shoot percentage was recorded in third sub culturing. Somatic embryos were developed into plantlets and subsequently grown to maturity (Kirtikar and Basu, 2001). These results indicate that nodal explants have high competence for somatic embryogenesis in *Plumbago indica*(Das and Rout, 2002). In the present study nodal explants have shown direct multiple shoot formation.

4. Conclusion

Conservation of plants in the changing scenario like draught and other calamities is a difficult task. Species of Ceropegia only appears in the nature after rainfall till end of September.

Voracious collection of tubers of Ceropegias by the tribals is serious matter for the existence of these plants. Propagation is easier but further maintenance is difficult. Awareness for conservation of these species in nature is expected from local peoples. This protocol will really help for large scale propagation of these novel plants.

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