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Effect of seasoning on sprouting of stem cutting and their survival in threatened medicinal important plant, *Commiphora wightii* (Arn.) Bhan.

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ABSTRACT : *Commiphora wightii* (Arn.) Bhan. known as Indian bdellium is a large shrub of family Burseraceae. Its oleo-gum resin is known to be anti-inflammatory, antirheumatic, hypocholesterolmic, hypolipidemic, and antifertility agent. In order to extract more oleo-gum resin it has been overtapped in past, killing its natural populations thus prompting IUCN to keep it in 'Data Deficient category'. Its *ex-situ* conservation through seed as well as vegetative propagation has met with variable success. Seasoning of stem cuttings for a duration that ensures more sprouting and consequent survival was, therefore, aim of this study. Cut stem cuttings kept for 0, 1, 2, 3, 4 and 5 days and treated with IBA (5000 ppm) revealed that maximum root biomass (0.56 g), maximum number of shoots (6) and length of shoots (49.50 cm) was found in five days seasoned stem cuttings. After 27 months of plantation maximum survival was found in four and five days seasoned stem cuttings (50% each) cuttings. Collar diameter (1.75 cm) was maximum in five days seasoned stem cuttings. Maximum plant height (119.75 cm) was in four days seasoned cuttings followed by five days seasoned cuttings and their subsequent survival in field plantation.

KEY WORDS : Commiphora wightii, Collar diameter, IUCN, Survival, Seasoning

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INTRODUCTION

Existing population of C. wightii are continuously

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 Kenter Contral Contrentent Contral Contral Contral Contral Contrent Contre declining in abundance and extent of occurrence due to its overexploitation for getting more and more oleo-gum resin which has high medicinal value. Deeper and more cuts in its stem have killed this plant. Its regeneration from seeds is poor in nature (Kumar and Shankar, 1982). It has been successfully propagated through stem cuttings of 25 cm length and 1 cm thickness (Mertia and Nagarajan, 2000; Chandra *et al.*, 2001 and Kumar *et al.*, 2002). Earlier, one-meter long and 10 mm thick woody stem cuttings were reported suitable for raising of *Commiphora wightii* (Dalal and Patel, 1995), thus, requiring a large number of plant material for propagation. Indole-3-butyric acid (IBA) is most widely used auxin for rooting in stem cuttings and to increase the success percentage of cuttings (Al-Saqri and Alderson, 1996). However, effect of seasoning on rooting is poorly understood. Less is also known about the effect of ambient conditions on stem cutting when these will give maximum sprouting as also best survival in the field.

In view of this, an experiment was carried out to see effect of different duration of seasoning on sprouting and survival of rooted stem cuttings of *C. wightii* in field.

EXPERIMENTAL METHODS

An experiment was conducted to study effect of seasoning on rooting, root traits, biomass production and success in the field establishment. 100 Stem cuttings from healthy C. wightii plants of same genetic stock were collected. Each cutting was 20 cm long and 1.0-1.5 cm thick. Cuttings were taken in first week of months of August-2009 and spread out under shade of green house with diffused light to callus for different durations from 0 to 5 days. After seasoning treatment lower tip of cuttings were dipped in 5000 ppm IBA solution prepared in 70 per cent alcohol. These treated cuttings were put for rooting in polybags containing rooting mixture of a clay, sandy soil and FYM (1:2:1) and kept under diffused light in nursery for rooting. At regular interval, watering was done and rooting data recorded. In first week of October-2010; 10 rooted cuttings were planted in field to study survival and data of growth parameters like height, collar diameter, mortality were recorded at regular intervals. At the same time from above treatments, four each rooted cuttings were taken for biomass analysis. Observations of mean values of all parameters like number of shoots, length of shoots, diameter of shoots, number of primary roots, length of primary roots, number of secondary roots, length of secondary roots, diameter of secondary roots, root biomass and shoot biomass were recorded. For biomass roots and shoots were dried in oven at 80°C till constant weight achieved.

EXPERIMENTAL RESULTS AND ANALYSIS

Evidently, longer the duration of seasoning, more was the swelling of tissues at the cut end of the stem cuttings. Maximum root biomass (0.56 g) was recorded in five days seasoned stem cuttings followed by four days

Table 1 : Different 1	root and shoot traits of	rooted cuttings (avg. of four sa	plings) of C. wightii seasone	ed for different days		
Days of seasoning	Nc. of shoots	Avg length of shoot (cm)	Avg dia. of shoots (cm)	No. of prim. roots	Length of prim. root (cm)	Avg dia. of prim. roots (cm)
	5.25±2.06	17.14±6.95	0.26 ± 0.02	11.75±3.50	12.45±13.62	0.11 ± 0.030
0	3-7	7.73-23.83	0.24-0.28	8-16	2.9-32.5	.09-0.14
-	6.00±1.41	16.58±4.44	0.32 ± 0.03	8.50±2.65	4.98±2.79	0.13±0.05
->	5-8	13.50-23.13	0.30-0.35	6-12	2.20-8.50	0.07-0.17
c	1.75±1.71	14.53 ± 4.36	0.28 ± 0.03	11.00±9.63	8.38±8.53	0.10 ± 0.01
N	3.7	8.83 19.37	0.25 0.32	3 26	2.50 20.70	0.06 0.14
,	4.25±0.96	26.78±9.27	0.33 ± 0.05	8.25±5.32	16.48±12.24	0.17±0.04
0	3-5	18-39.80	0.27-0.38	4-15	7.40-34.50	0.12-0.22
	3.75±1.71	28.96±15.94	0.36 ± 0.14	6.00±4.76	18.13±6.06	0.21±0.06
4	2-6	12.50-46.83	0.23-0.55	3-13	12-26.50	0.16-0.30
	6.00±2.45	49.50±8.01	0.41 ± 0.11	4.25±2.22	35.73±9.27	0.28 ± 0.04
0	3-8	39-58.50	0.28-0.56	2-7	23.50-46	0.22-6.31
DAV	S	25.58	0.33	8.79	16.02	0.17
C.D. (P=0.05)	2.64	13.14	0.12	7.9	14.1	0.07
						Table 1 : Contd

					Table 1 : Cont
Days of seasoning	No. of sec. roots	Length of sec. root (cm)	Avg dia. Sec. root (cm)	Root biomass (g)	Shoot biomass (g)
¢	8.00±3.61	2.60 ± 0.80	0.03 ± 0.02	0.11 ± 0.05	0.91 ± 0.46
0	5-12	1.8-3.4	0.02-0.06	0.06-0.17	0.27-1.27
-	4.50⊥0.71	916.92	0.04±0.01	0.0610.05	1.15⊥0.46
Т	4-5	0.03-0.05	0.03-0.05	0.02-0.12	0.86-1.83
	00.00	000	000	0.08 ± 0.04	0.93 ± 0.55
v	0000	0.0	00:0	0.04-0.13	0.45-1.71
ŕ	2.67±1.15	12.67±7.65	0.07 ± 0.04	0.18±0.12	1.98±1.08
0	2-4	8-21.50	0.04-0.13	0.06-0.35	1.24-3.57
-	7.67±8.08	16.40±3.76	0.13±0.01	0.39±0.35	4.03±3.6
-	3-17	12.50-20	0.11-0.15	0.13-0.92	0.84-8.97
	5.00+2.58	20.48±10.37	0.10 ± 0.04	0.56±0.21	8.56±3.97
0	28	5.50 28.90	0.08 0.16	0.33 0.83	1.31-12.47
AVG	3.50	8.12	0.05	0.23	2.93
C.D. (P=0.05)	6.09	10.65	0.06	3.37	0.26

EFFECT OF SEASONING ON SPROUTING OF STEM CUTTING & THEIR SURVIVAL IN THREATENED MEDICINAL IMPORTANT PLANT, Commiphora wightii (Arn.) Bhan.

seasoned (Fig 1). While, it was minimum in one day seasoned (Table 1). In case of shoot biomass of rooted stem cuttings in the month of August 2009, as number of days of seasoning increases shoot biomass increases. Maximum shoot biomass (8.56 g) was produced (Fig 2) in five days seasoned stem cuttings (Table 1).







Fig. 2: Effect of callusing treatment on shoot biomass production

Values for all traits of rooted stem cuttings seasoned for four to five days were higher than the other rooted stem cuttings (Table 1). Maximum number of shoots (6) and length of shoots (49.50 cm) were in five days seasoned stem cuttings while minimum shoot length (14.53 cm) in two days seasoned stem cuttings in which secondary roots were absent (Table 1). Though numbers of primary roots were maximum in case of zero day seasoned cuttings (11.75), length of primary roots were less as compared to four and five day seasoned cuttings. Similar trend was found in case of secondary roots.

After 27 months of plantation of these rooted stem cuttings in field, maximum height was attained in four days seasoned cuttings (119.75 cm) followed by five days seasoned cuttings (96 cm, Table 2). Two days seasoned cuttings showed minimum height (53.50 cm). Similar trend was observed in case of two days seasoned stem cuttings for collar diameter (1.35 cm) and survival (20%). Collar diameter was maximum in five days seasoned cuttings (Table 3). But survival in field was recorded maximum in four and five days seasoned cuttings (50% each) followed by zero and three day's seasoned stem cuttings (30% each), while it was absent in two days seasoned cuttings (Table 4).

These results suggest that cuttings rooted in the month of August and seasoned for four and five days showed more survival in the field and these plants had more height and collar diameter. Again cuttings rooted in the month of August and callused for four to five days showed more shoot and root biomass. May be this could be the reason for more survival of cuttings rooted in the month of August and seasoned for four to five days, because biomass was also produced more which helps plants to grow vigorously giving more chances of survival in field conditions. Similar results in yellow cedar were reported by Karlson (1974 and 1981); Raimondi and Kermode (2004); Bonner and Karrfalt (2008) led to the development of yellow-cedar planting seedlings produced by rooted cuttings.

Season, timing or period of the year in which cuttings are taken can play an important role in rooting (Bassuk and Howard, 1981). In propagating deciduous species, hardwood and semi-hardwood cuttings can be taken during the dormant season when buds are not active and before buds start to force out in the rainy season. There

Table 2 : Average height of C. wightii plants raised from cuttings seasoned for different days						
No. of seasoning days	After 1 month	After 6 months	After 17 months	After 21 months	After 27 months	
0	19.80	23.75	40.63	47.90	60.75	
0	3.5-33.5	9-34	9-74	17-75	30-107	
1	16.78	22.75	46.06	50.50	60.40	
1	3.5-39	4.5-34	13-77	25-78	40-80	
2	22.28	20.83	45.50	44.00	53.50	
2	12-29.2	18-22.5	21-84	40-51	37-70	
2	24.99	21.36	70.20	75.70	71.20	
5	11.6-36.3	6.5-47	45-93	40-94	44-120	
4	54.25	58.95	85.10	86.30	119.75	
4	30-87	34-88.5	84-110.5	84-1132	113-168	
5	57.60	60.33	80.58	82.20	96.00	
3	33-68	43-102	45.5-117.5	47-115	45-130	
3 4 5	24.99 11.6-36.3 54.25 30-87 57.60 33-68	21.36 6.5-47 58.95 34-88.5 60.33 43-102	70.20 45-93 85.10 84-110.5 80.58 45.5-117.5	75.70 40-94 86.30 84-1132 82.20 47-115	44-120 119.75 113-168 96.00 45-130	

Table 3 : Average collar diameters of C. wightii plants raised from cuttings seasoned for different days No. of seasoning days After 1 month After 6 months After 17 months After 21 months After 27 months 0.34 0.28 1.02 1.11 1.36 0 0.18-0.56 0.11-0.46 0.75-1.30 0.77-1.35 1.02-1.89 0.27 0.28 1.10 1.52 1.12 1 0.14-0.41 0.21-0.33 0.90-1.37 0.87-1.29 1.35-1.72 0.35 0.29 0.94 1.07 1.35 2 0.27-0.43 0.25-0.31 0.59-1.40 0.76-1.32 1.14-1.55 0.37 0.32 1.28 1.20 1.44 3 0.31-0.47 0.21-0.43 1.01-1.62 0.92-1.69 1.08-1.81 1.35 0.52 0.54 1.35 1.71 4 0.34-0.70 0.35-0.94 0.91-1.67 1.01-1.62 1.66-1.78 0.55 0.61 1.19 1.24 1.75 5 0.40-0.81 0.32-0.93 0.72-1.58 0.79-1.53 0.87-3.13

is an optimal period for rooting many species, which is necessary to maximize the rooting process (Arnand and Herbalein 1975 and Hartmann et al., 1997). Davis (1984) observed a vivid seasonal change in shoot production and rooting in Ficus pumila. Heavy flushing was also observed during the rainy periods, a time of intense vegetative growth, which may tend to increase rooting percentage. Cuttings do not root normally in the dry season, however, rejuvenated shoots may still have superior root development probably because of higher food reserves and other rooting co-factors. Evans (1992), contended that probably the best time to take cuttings from the field is at the beginning of the rainy season. Stem cutting method for plant production in C. wightii has been studied by several workers (Shah et al., 1983; Dalal et al., 1989; Singh et al., 1989; Mertia and Nagarajan, 2000; Kasera and Chawan, 2001 and Yadava, 2011) as a means to conservation. Their study reports various growth regulators, season and method of collection and planting of cuttings, for improved establishment and growth. As many as nine published studies (Table 5) report use of IBA in sprouting stem cuttings of *C. wightii*. These researchers reported IBA concentrations of 10-5000 ppm in which stem cuttings were given either pulse treatment as quick dips or soaked lower portion of cuttings in IBA solution for durations ranging from 5-10 minutes or even a lower IBA dose (50-100 ppm) through injection (Table 5). In view of such divergent treatments, when there is no single agreed concentration of IBA, prevailing IBA concentration of 5000 ppm as quick dip in its nursery raising was followed in this work.

Rathore (1984) had also observed the enhanced rooting in Kiwifruit with IBA application. Maximum number of primary, secondary and average root number in *Pyrus pyrifolia* L. were recorded at 5000

Table 4 : Survival per cent of C. wightii plants raised from rooted cuttings seasoned for different days							
No. of seasoning days	After 1 month	After 6 months	After 17 months	After 21 months	After 27 months		
0	100	90	40	30	30		
1	100	90	10	10	10		
2	100	90	0	0	0		
3	100	70	30	30	30		
4	100	100	80	60	50		
5	100	90	50	50	50		

Table 5 :	IBA treatments of stem root	ting of C. wightii repo	orted by different worker

Sr. No.	Hormone	Duration	Results	Authors
1.	50 and 100 ppm IBA	Injected with syringe	Maximum number of roots produced	Mishra, 2011
2.	1000, 2000, 3000, 4000 and 5000 ppm IBA	-	2000 ppm IBA concentration most effective for rooting of cuttings of 5-6 mm diameter and 6-10 inch long	Rai et al., 2011
3.	100, 200, 500 and 1000 ppm IBA	10 min dipping	200 ppm IBA best for rooting in cuttings of 0.25- 0.50 cm diameter and 20-25 cm long	Tripathi et al., 2014
4.	10, 50, 200, 300 and 500 ppm IBA	10 min dipping	500 ppm IBA best for rooting in cuttings of diameter < 0.25 cm and 6-10 cm long (micro cuttings)	Tripathi et al., 2014
5.	100, 200, 500 and 1000 ppm IBA	10 min dipping	200 ppm IBA concentration is optimum for rooting of stem cutting of 0.25-0.50 cm diameter and 15-20 cm long	Mishra and Kumar, 2014
6.	500, 1000, 1500, 2000, 2500 and 3000 ppm IBA	Quick dip method (5 seconds)	1500 and 2000 ppm IBA effective in softwood cuttings 25 cm long	Kumar et al., 2006
7.	2000, 4000 and 6000 ppm IBA	5 min dipping	4000 ppm IBA effective rooting in hardwood cuttings 20 cm long	Bandi et al., 2012
8.	200, 300, 400 and 500 ppm IBA	-	$300~{\rm ppm}$ IBA for cuttings of 6-10 mm thick and 15-20 cm long	Dhar, 2011
9.	5000 ppm IBA	Quick dip method	20-30 cm long and 1.5-2.0 cm diameter stem cuttings dipped and taken out	Jindal et al., 2005

ppm IBA up to certain concentration (Khalon and Singh, 1981). Hitchcock and Zimmerman (1940) also observed that, auxin treatment results in high percentage rooting and the development of more roots per cutting. Apparently, the movements of boron (B), nitrogen (N), zinc (Zn) and potassium (K) to the rooting zone as observed by Blazich et al. (1983) is enhanced by auxin treatment. In most of the naturally vegetatively propagated species, adventitious root formation is there without any need for hormone treatment, while others require different growth regulators usually auxin (Syros et al., 2004). Auxins (IAA and IBA) are widely used as it induces root formation by breaking root apical dominance induced by cytokinin (Cline, 2004). Pretreatment with IBA and NAA increased both rooting and sprouting in Jatropha (Kochhar et al., 2008).

Thus, it can be concluded that for large scale multiplication of *C. wightii*, its stem cuttings should be treated with hormone and should be kept for rooting in the month of August with four to five days callusing treatment.

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