3
Results and Discussion

I) Sprouting potential study:

It is well known that mangroves need fresh water conditions for their establishment (Bhosale, 1978). It is believed by some that mangroves do not grow under fresh water conditions. However, it is observed that all species grow well under fresh water conditions and may need salinity at a later stage of growth (Bhosale, 1993). The stem cuttings of E. agallocha L. from the natural population, were collected to observe the response of them to fresh water condition using nursery techniques. Every plant has its own sprouting potential but the response to changed habitat is different. Ding Hou (1958) reported that Excoecaria have abundant reserve buds in the stem. So attempt has been made to study it by observing the sprouting potential of them. For this purpose the present study has been categorised into following groups :

A) Effect of length variation:

The stem cuttings of various lengths i.e. 10 cm. to 20 cm. (Set I) were considered for the present study. During the sprouting potential study it was observed from the Fig. 1a that, as the length increases from 10 to 20 cm. there is increase in per cent of sprouting, along with the increase in number of sprouts. As L_{20} i.e. 20 cm. length of cuttings shows highest i.e. 60 % sprouting potential.

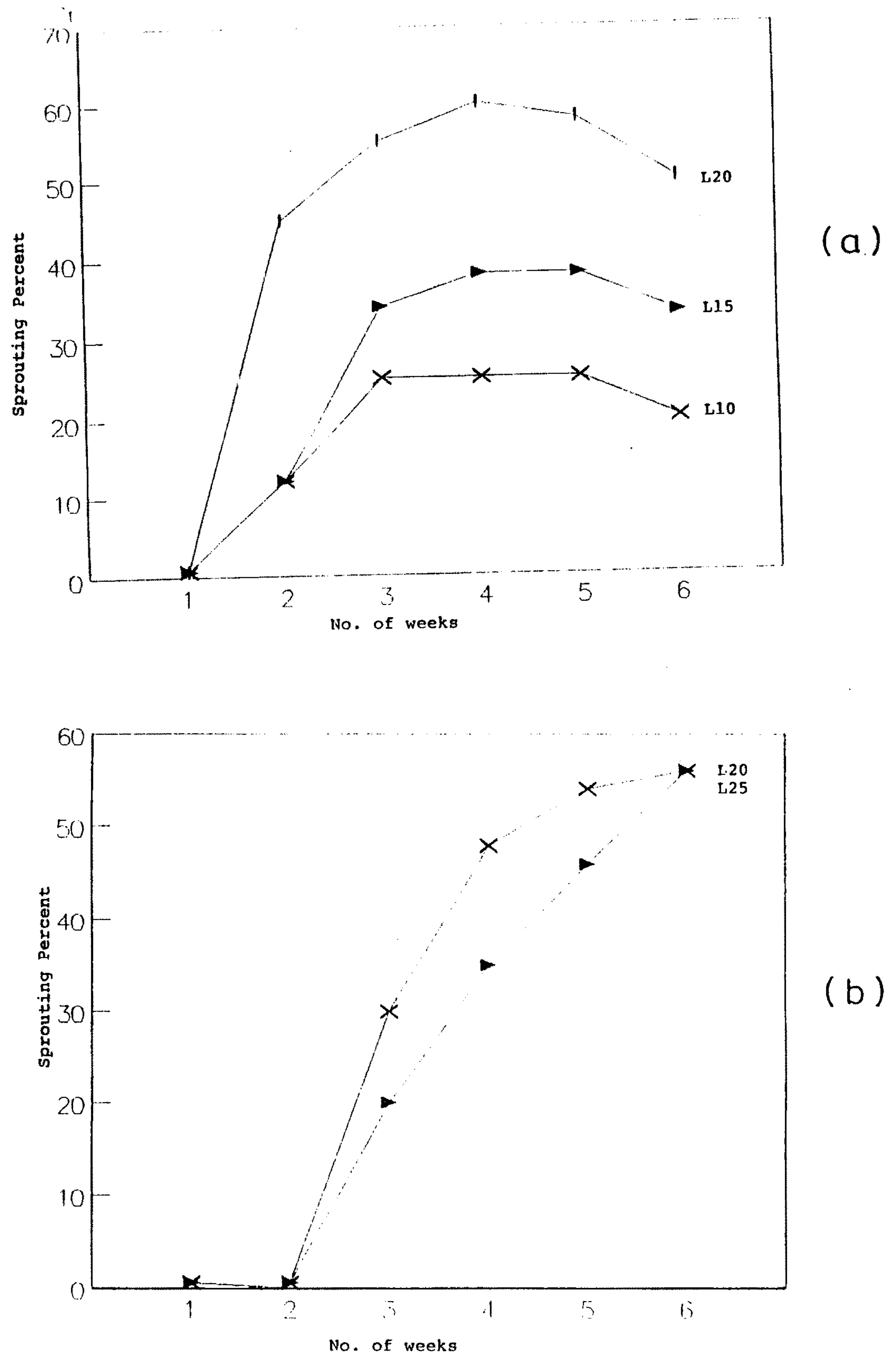


Fig.1 - Effect of length variation on sprouting potential.

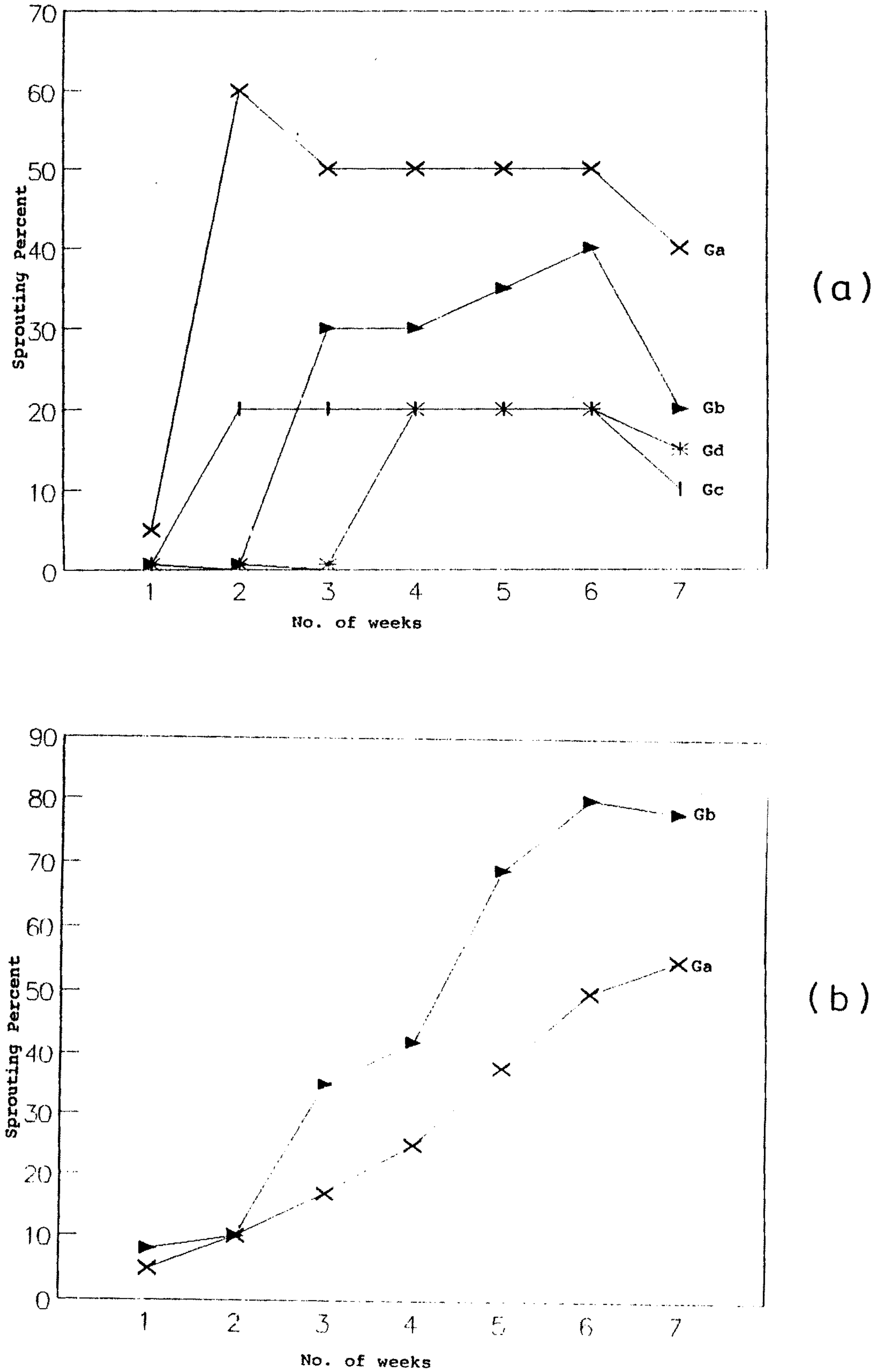


Fig. 2 - Effect of Girth-size variation on sprouting potential.

In the set II the stem cuttings of length 20 cm. and 25 cm. were considered. It is oobserved from the Fig. 1b that the highest sprouting potential in both the cases is equal i.e. 56% (Plate 3a). But the survival per cent of L_{20} cuttings is more as compared to other lengths. Hence L_{20} has been considered as suitable length for further studies (Plate 3b). Divekar (1984) reported that in case of Carissa carandas L. also influenced on the rooting phenomenon. Same case is observed in case of E. agallocha in which length is influenced by the sprouting potential.

B) Effect of Girth-size Variation:

The stem cuttings of various girth-size i.e. from 0.5 mm to 1.6mm were used for the present study (Plate 4). It is observed from Fig. 2a (Set I) that, there are more number of sprouts along with the increasing sprouting potential percentage i.e. 50 % and 40 % in Ga and Gb groups respectively. While in Gc and Gd groups the sprouting potential is less i.e. 20 % . So in Set II, Ga and Gb groups were studied. From Fig. 2b it is observed that,Gb group shows highest sprouting potential i.e. 80% than Ga group i.e. 50 %. From these observations it is cleared that Gb group i.e. 0.9 to 1.1 mm range of cuttings is suitable for further studies.

C) Effect of Plant Growth Regulators (PGR):

Cooper (1935) was the first to investigate the practical

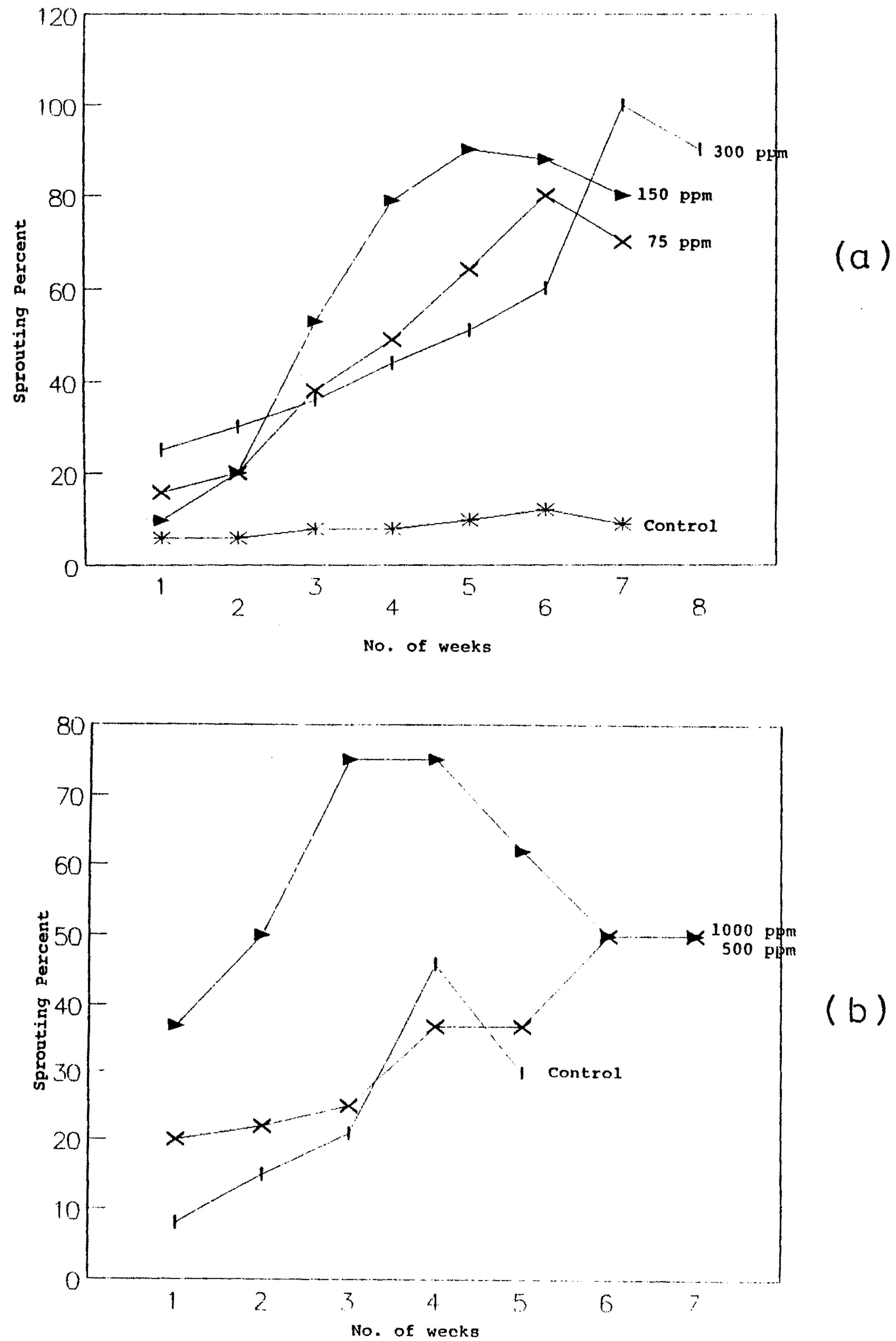


Fig.3 - Effect of plant growth regulators (PGR) on sprouting potential.

I) IAA treatment

application for growth regulators in plant propagation. Plant growth regulators such as, Indole acetic acid (IAA) and Indole butyric acid (IBA) have been used to treat the cuttings.

Sprouting - From Fig. 3a it is observed that, at 75 ppm IAA the sproutting potential is 80 % in case of 150 ppm IAA it is 90 %.While in 300 ppm IAA it is 100 % (Fig. 3a). Control shows very less sprouting potential i.e. 12% as compared to treated one (Plate 5). Cooper (1944) found the concentrated solution method of treatment quite effective and more convenient than the dilute solution method. By taking into consideration this view the highest concentration of IAA i.e. 500 ppm and 1000 ppm IAA have been tried for sprouting as well as rooting. IAA brings about its mainfold effects and in particular how IAA functions in intact plants. For this reason the preferred auxin is IAA when it can be used (Narasimhan et al., 1970). From Fig.3b, it is observed that sprouting potential is highest i.e. 75 % at 1000 ppm IAA than that of 500 ppm IAA i.e. 50 %. While control shows 30 % sprouting potential (Plate 6).

Fig.3c and Fig.3d shows same observations in case of IBA as that of IAA, as discussed in Fig. 3a and 3b. From all above observations we can say that at lowest concentration of IAA as well as IBA the sprouting potential is lower than that of highest concentrations.

Rooting : Along with the environmental conditions, like light and

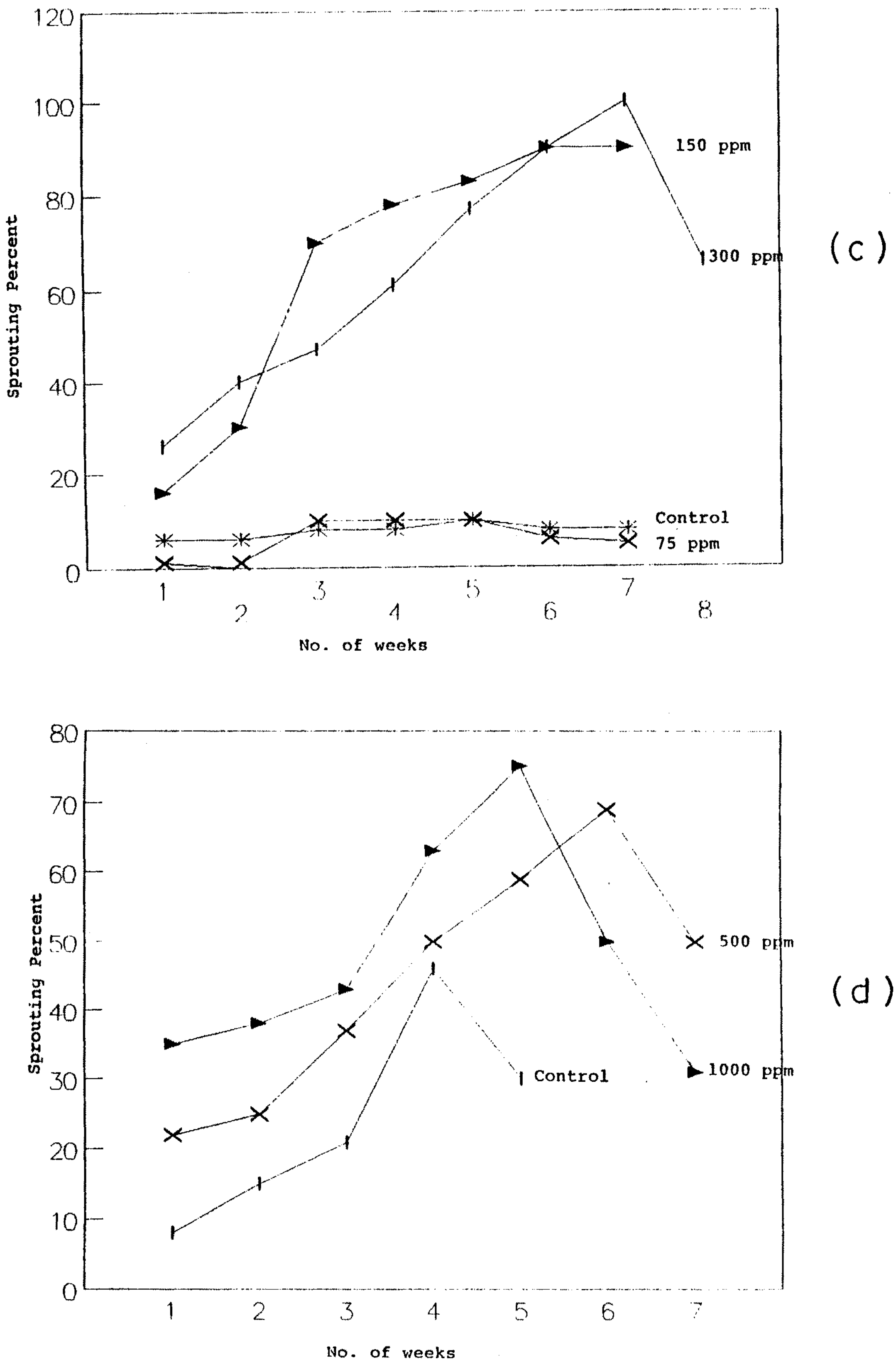


Fig. 3 - Effect of plant growth regulators (PGR) on sprouting potential.
II) IBA treatment

temperature, PGR also plays an important role in the root formation on stem cuttings (Nanda, 1976). In case of E. agallocha stem cuttings showed vigorous rooting at highest concentration i.e. 1000 ppm IAA and 1000 ppm IBA which are depicted in Plate 7.

According to Divekar (1984) application of 1000 ppm IBA by quick deep method shows maximum rooting per cent as compared to other concentrations which has been also observed in case of E. agallocha. Growth regulators like auxins are well known to be successful for inducing rooting and several workers have reported a very high percentage of rooting in a large number of plant species (Kale and Bhujabal,1972; Singh, 1980). Paul and John (1991) recorded the maximum percentage of rooting i.e. 86.66% in 1500 ppm IBA treated cuttings. While the control cuttings produced the minimum percentage of rooting i.e. 13.33 %. It is also reported that IBA and NAA significantly promoted root formation. The root promoting effects of IBA and NAA in various plant species has been reported by several workers (Gandotra et al.,1975; Singh,1980).

D) Effect of Keradix treatment :

Sprouting : Keradix powder is very much popular and used as rooting medium in nursery practices (Bhosale Personal Communication). Plate 8a shows the effect of Keradix powder on sprouting potential of stem cuttings. From Fig.4 it is clear that

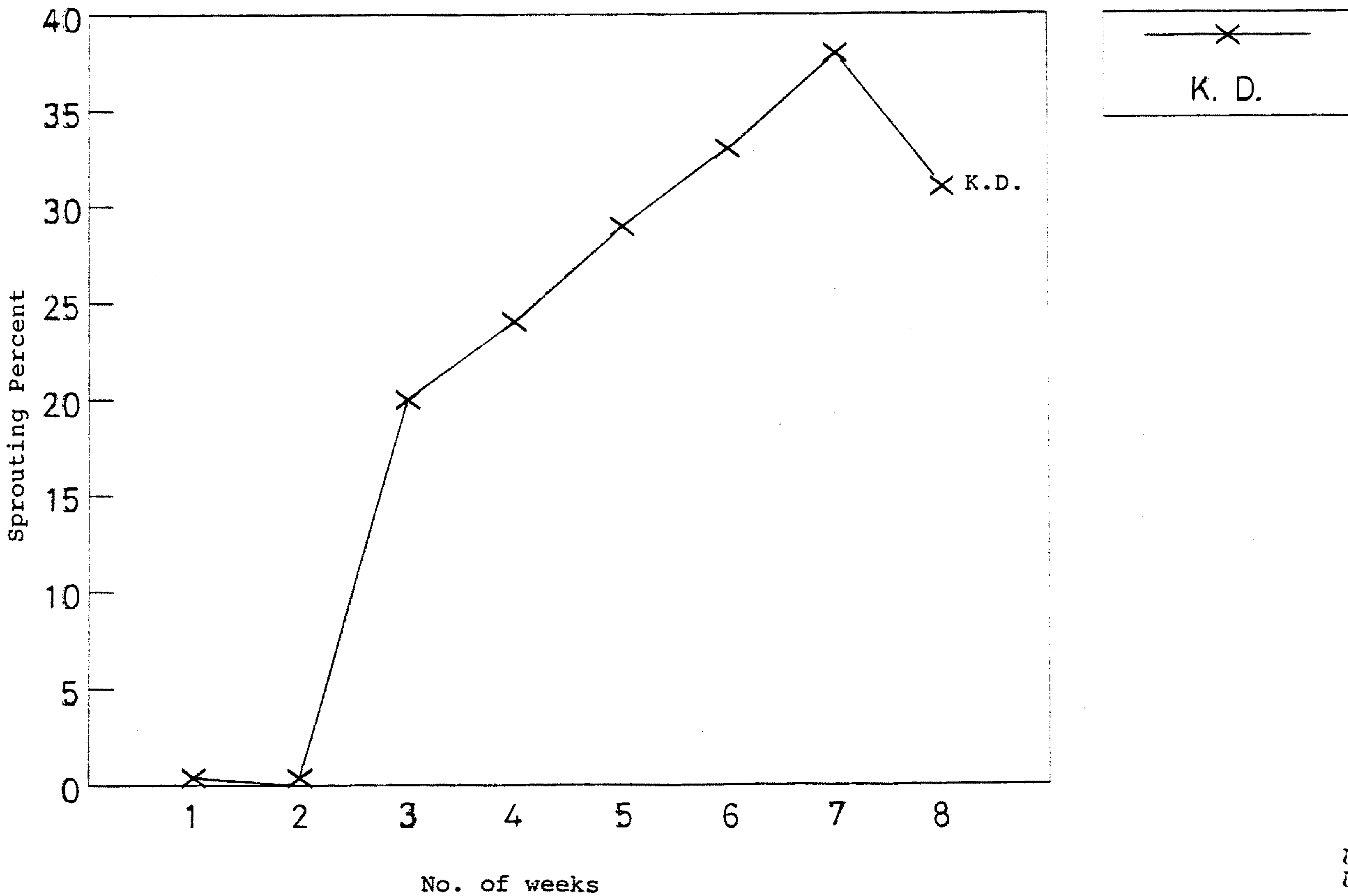


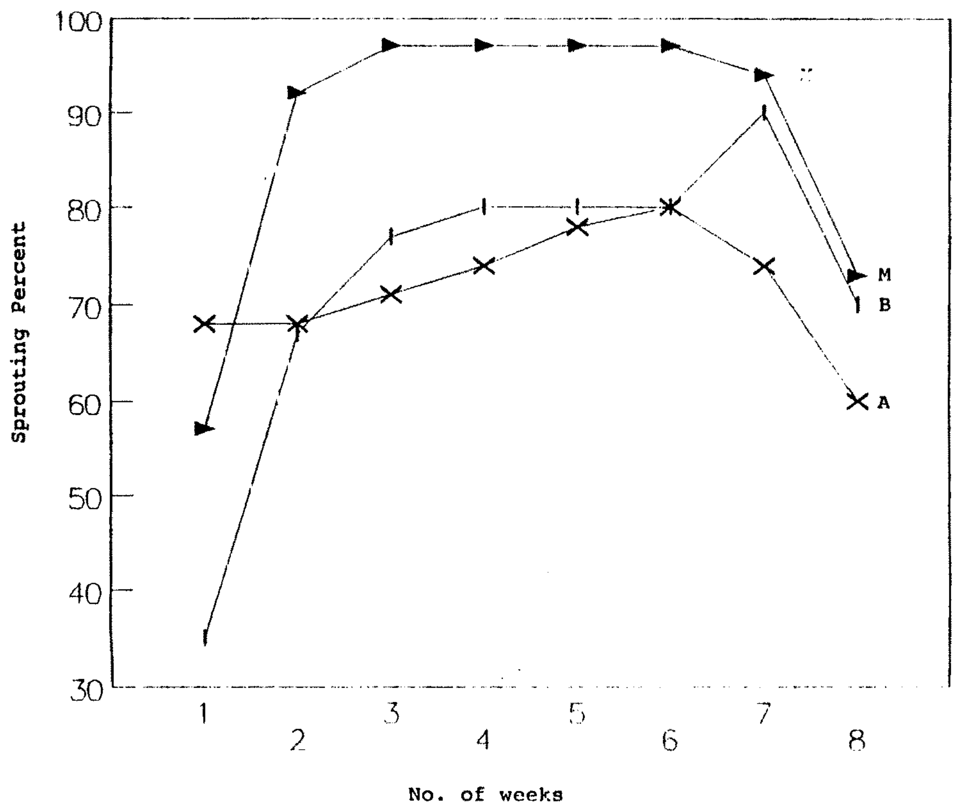
Fig. 4 - Effect of Keradix treatment on sprouting potential.

there is increase in the sprouting potential i.e. upto 38 %.

Rooting : Powder-dip method was first suggested by Grace (1937) in which cuttings are treated with keradix powder. Plate 8b shows, the rooting in the cuttings treated by Keradix powder. Number of roots are induced. The cuttings which are having less girthsize i.e. Ga and which is having more girth size i.e. Gd produced roots i.e. Keradix powder induces rooting of E. agallocha.

E) Effect of Morphological position:

In the present work Fig. 5a, shows that the highest per cent sprouting i.e. 97 % are observed in case of 'M' position i.e. middle position. While less number of sprouts and per cent sprouting are observed in the 'A' region, i.e. apical region. But the apical region cuttings failed to survive. Only the middle and basal region cuttings could survive upto final stage. The sprouting of middle and basal region cuttings can be observed in Plate 9a and 9b. The failure of survival of apical region may be possibly due to ill developed conducting strand. Hansen, Jurgen (1986) reported that the cuttings of Schefflera arboricola from sub-apical positions rooted more slowly produced fewer roots, and had a lower rooting percentage than cuttings from the more basal region. Fig.5b also shows the higher response of sprouting potential 'M' region cutting and could survive upto final stage.



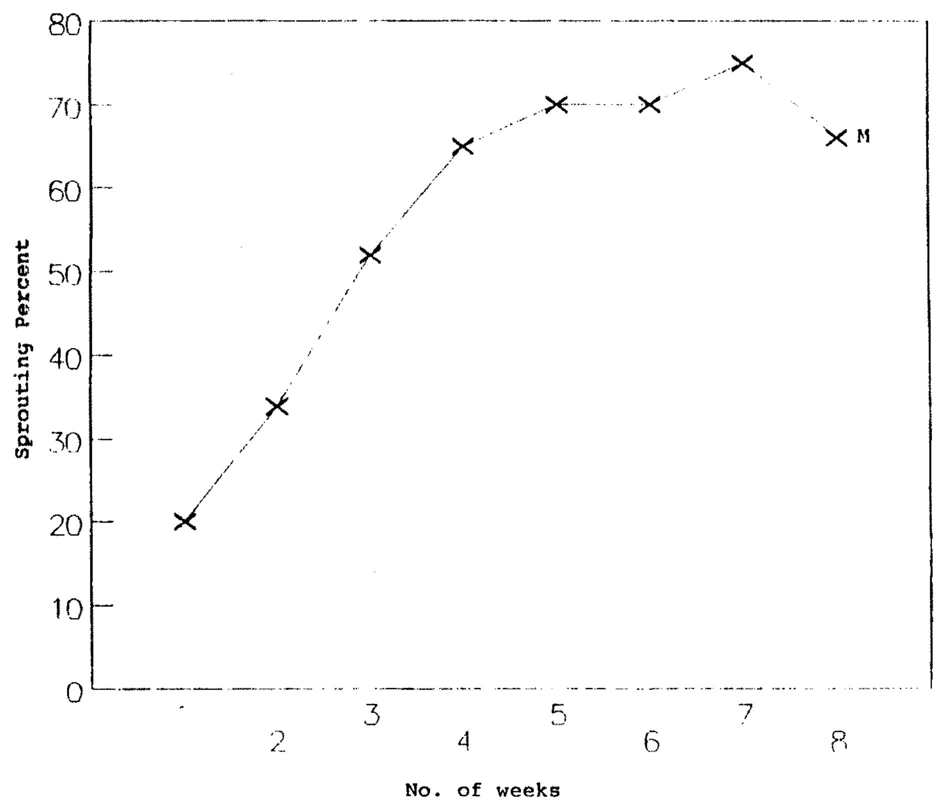


Fig. 5 – Effect of Morphological positions on sprouting potential.

Rooting: Hansen, Jurgen (1986) reported that the number of roots and rooting percentage increased with the length of the stem below the node. Plate 10a shows the rooting after 5 months and Plate 10b shows the vigorous rooting after 13 months As the middle region stem showed highest sprouting potential resulted into vigorous rooting which has been survived for ever and considered as suitable position for culture technique.

II) Response of cuttings to fresh water conditions :

The study of culture of mangroves have received less attention little or no work is available to distinguish species of mangrove genera at their seedling stage. Same case is observed with *E. agallocha*. So present attempt is made to study the growth performance of *E. agallocha*. Various types of cuttings were planted under laboratory condition and studied for various growth parameters, whenever they reached to stable condition.

The statistical data of the results of the experiment, 7 months after plantation, are presented in Table 1. Table 2, shows the average response of stem cuttings to fresh water condition. It is observed that the survival percentage is highest in 500 ppm and 1000 ppm IAA and lowest is 0.2 % in the middle position. From these observations it is clear that the plants of middle position although they are 8 months old showed less survival percentage but the 4 months old cuttings of 500 IAA and

Table 2 : Average response of E. agallocha cuttings to fresh water conditions.

Cuttings	Survival percentage	No. of branches	Length of branch (cm)	No. of leaves	Leaf thickness (mm)
Middle position					
I	0.2	5.4	41.5	82	28.17
II	3.5	4	21.7	35	28
Basal position					
I	6	4.2	30.9	91	27.4
Length 20 cm					
II	3.5	4	25.1	35	28
Keradix treated					
II	12	1.2	20.2	9	31.1
75 ppm IAA treated					
II	10	1.7	16.4	15	25.3
150 ppm IAA treated					
II	24.2	3	17	32	27.4
150 ppm IBA treated					
II	18.5	2	17.3	24	27
500 ppm IAA treated					
III	57.1	2	3.2	11	33.6
500 ppm IBA treated					
III	50	3.1	1.9	15	28.3
1000 ppm IAA treated					
III	51.7	2.1	2	10	32.6
IV	34.4	3.2	14.3	35	32.3
1000 ppm IBA treated					
III	33.9	3.7	3.6	11	32.4
IV	41.9	3.2	12.1	35	32.6

1000 IAA showed the best response. It is also significant that the cuttings of 1000 ppm IBA although are of 2 months old showed very good response.

If number of branches are considered it can be easily seen that highest number is found in middle position (I), while lowest has been found in Keradix treatment. It is also significant that the 4 months old 1000 ppm IBA showed the best response. Highest length of branches has been achieved in middle position (I). While lowest is observed in 500 ppm IBA (III). One can not neglect that the 2 months old 1000 ppm IAA and 1000 ppm IBA showed the increase in length within very short duration. Kulkarni (1990) has studied growth performance of Rhizophora mucronata and Rhizophora apiculata propagules collected at different time under laboratory conditions as well as under natural conditions at Shirgaon, Ratnagiri.

Number of leaves are found to be highest in the middle position (I) while lowest has been found in Keradix treatment. It can not be denied that, 1000 ppm IAA and 1000 ppm IBA cuttings of 2 months old also showed remarkable response to this parameter. Regarding the thickness which is found to be highest in 500 ppm IAA (III) while lowest has been observed in 75 ppm IAA (II). It is also a fact that, 1000 ppm IAA and 1000 ppm IBA of 4 months and 2 months old cuttings showed a significant increase in thickness within limited time. Deshmukh and

Karmarkar (1991) have recorded average height, leaves/plant, Number of lateral branches/plant for *R. mucronata* propogules under natural condition at Vikroli mangroves, Bombay.

From all above observations, it is clear that response of cuttings to fresh water condition is best for 1000 ppm IAA and 1000 ppm IBA. While better response is observed for the middle position which can be confirmed by their extensive rootings shown in Plate 7 and Plate 10 respectively.

III Response of seedlings to fresh water condition :

In India work on seed germination in mangroves has been reported by Bharucha and Shirke (1947); Joshi et al. (1972), Jamale (1975) and Bhosale (1978). The need to raise mangrove nurseries was based on the following consideration. In nature, propagules of different mangrove species are available only for some part of the year (Deshmukh and Karmarkar 1991). In case of *E. agallocha* the germination percentage of seeds is very low (Bhosale 1990). In our laboratory, it has been already observed by Mulik (1987) that, in the month of September different seed developing stages are found seeds start maturing from September. During September to October the number of seedlings of *E. agallocha* are very less, out of which only few could survive in the nature. Multiplication through seed will take longer time as seed is produced only at maturity and seed planting can not be isogenic (Khoshoo, 1988). So it was thought worthwhile to

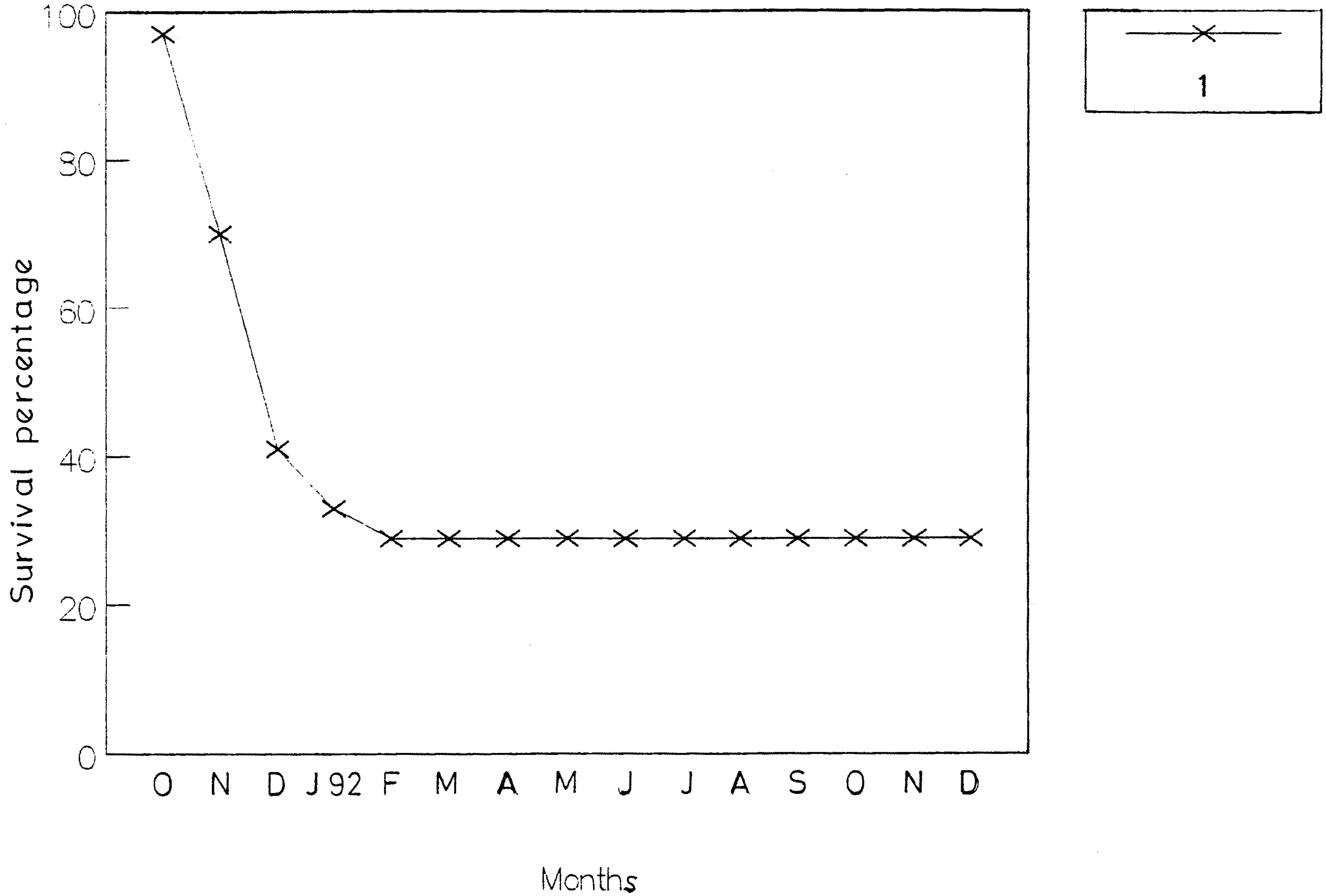


Fig. 6 – Survival percentage of seedlings.

bring available seedlings to the laboratory and grow them in the nursery. 80 seedlings were planted in polybags under fresh water conditions and following parameters have been studied. Plate 11a shows planted young seedlings at the initial stage.

A) Survival percentage :

After plantation the study of survival percentage has been carried out. From Fig.6 it is clear that the survival percentage in the month of October is 97 %, while it is decreased upto 29% in the month of February. There might be some biological factors which affected in the growth and survival of E. agallocha seedlings under fresh water condition. Further, its survival percentage is found to be constant throughout the year. Benecke and Arnold (1931) studied development of mangrove seedlings under culture conditions. Field (1985) has shown that, higher salinities decline the growth rate of seedlings. According to Bhosale (1976) Bruguiera and Acanthus plants can successfully grow under culture condition. Teas (1975) has been able to grow Avicennia seedlings. From all above observations it is clear that E. agallocha can be successfully grown under fresh water condition, this can be observed in Plate 11.b which shows healthy survived seedlings after 5 months.

B) Growth Performance :

Whenever the seedlings have shown the healthy growth

Table 3 : Monthly Variation in growth parameters of _E. agallocha_ seedlings :

Growth parameters	June	July	August	September	October	November	December
1) Average height of seedlings (cm.)	40.7 ± 1.37	44.5 ± 0.82	49.00 ± 2.64	57.2 ± 2.57	63.4 ± 2.37	70.4 ± 0.55	76.5 ± 2.27
2) Average girth-size of seedling (mm)	0.451 ± 0.005	0.520 ± 0.028	0.560 ± 0.001	0.591 ± 0.013	0.610 ± 0.003	0.639 ± 0.020	0.681 ± 0.01
3) Number of branches per seedling	1	2	3	3	3	4	4
4) Number of leaves per seedling	44 ± 1	51 ± 5	59 ± 1	63 ± 2	69 ± 1	78 ± 2	74 ± 3
5) Leaf thickness (mm)	27.2 ±1.75	25.2 ±7.7	28.00 ±9.00	28.2 ±0.25	29.00 ±3.0	30.00 ±3.0	30.00 ±1.75

Table 4 : Average annual growth performance of E. agallocha seedlings.

	Growth parameters	Initial stage	Final stage
1)	Height of seedling/plant (cm)	12.90 ±1.15	76.5 ±2.27
2)	Girth-size of seedling/plant (mm)	0.243 ±0.030	0.681 ±0.01
3)	Number of branches/plant	Nil	3.85 ±0.15
4)	Length of branch/plant	Nil	11.1 ±14.4
5)	Frequency of branching	Nil	40%
6)	Number of leaves/plant	6	74 ±3
7)	Leaf thickness (mm)	25.25 ±3.9	30.25 ±1.75
8)	Leaf length (cm)	1.5	9.87 ±4.13
9)	Leaf breadth (cm)	1	4.19 ±2.31
10)	Leaf area $(cm)^2$	1	28.484 ±32.98

* Sample size - 80 seedlings planted.

and constant survival per cent (Fig.6) it has been decided to transfer the seedlings in the big pots after 6 months as shown in Plate 12 a. Transplanted seedlings have been allowed to stabilize for a month under same condition. The data on their growth performance was collected from June to December. Growth parameters include, Height of seedling, girth size of seedlings, number of branches/seedling, number of leaves/seedling and leaf thickness which are given in Table 3. It shows that as there is increase in the age of the seedling, there is also increase in the height, girth size, number of branches, number of leaves and leaf thickness.

The growth performance at initial and final stage has been given in Table 4. Plate 12 a shows the extensive growth of the seedlings at the final stage. The net growth in height from 12.9 cms. to 76.5 cms. has been attained by the seedlings. Girth-size has also been increased considerably which ranges from 0.24 to 0.68 mm. Number, length and frequency of the branches/seedling has been increased.There is considerable increase in the number of leaves i.e. 12 times.There is also increased in leaf thickness from 25.25 mm to 30.25 mm after which defoliation has been observed. Leaf length, breadth and area has been also remarkably increased. From these observations it is clear that, the seedlings show better growth performance upto final stage without any infestation, which is also observed by Kulkarni (1990), Deshmukh and Karmarkar (1991) in case of

<u>Rhizophora</u> propagules. Greenway (1968) has got best results from cultured <u>Atriplex nummularia</u> in culture solution at low concentration of NaCl. Bhosale (1978) has reported that majority of propagules of mangroves established themselves in rainy season indicating the requirement of fresh water condition or less salinity condition for propagules or seedlings growth.

Better growth performance of the seedlings can also be confirmed by the extensive growth of the roots which can be observed in Plate 12 b.

C) <u>Distribution of organic constituents</u>:

Earlier workers have tried to understand the problem of mangroves by studying some physiological aspects (Bhosale, 1974; Jamale, 1975). In continuation of earlier work in our laboratory, the present investigating attempts are carried out to study physiological parameters represented by some organic constituents. It is thought worthwhile to know the distribution of various constituents like chlorophylls and Polyphenols from the seedlings grown under fresh water condition as well as from the leaves of <u>E. agallocha</u> growing under natural conditions. Table 5 shows the values of chlorophylls and Polyphenols during winter, summer and monsoon season.

i) <u>Chlorophylls</u> :

Bhosale et al. (1983) reported that Chlorophyll is the important plant pigment, depends upon the environmental conditions. In the present work, seasonal variation occurs in the content of Chlorophyll. Salinity in general reduces the Chlorophyll contents of the leaves in mangroves (Bhosale, 1974; Bhosale et al., 1983; Joshi and Bhosale, 1982; Shinde and Bhosale, 1986; Mishra 1967). Table 5 shows that the salinity affects the Chlorophyll content. Amount of chlorophyll content is more in the leaves which are planted in nursery i.e. under fresh water condition and Chlorophyll content is less in case of the leaves collected from the natural population i.e. Chlorophyll content is directly related with salinity. It is also clear from the Table 5 that highest chlorophyll value is seen in monsoon. This is generally seen in many saline plants investigated in our laboratory (Bhosale, 1974). It also shows that, E. agallocha leaves contains more amount of chlorophyll 'b' than chlorophyll 'a'. Jamale (1975)has seen that chlorophyll 'b' is always more than chlorophyll 'a' in many mangrove species like Excoecaria and Sonneratia. Mishra (1967) reported that in Clerodendron inerme amount of chlorophylls is reduced to half under saline conditions as compared to garden plants. Chlorophyll a/b ratio automatically increases in case of fresh water seedlings as compared to natural population. Chlorophyll a/b ratio obtained here is lower than that obtained for C_3 plants viz. from 2.5 to 3.7 (Holden, 1973). Bhosale (1974) has reported more chlorophyll 'b' than chlorophyll 'a' from mangrove species A. majus while in A. ilicifolius and A. officinalis a/b ratio is more than 1. According

Table 5 - Seasonal Variation in Chlorophylls and Polyphenols in the leaves of *E. agallocha*

Season	Locality	Chloro.a	Chloro.b	Chloro. a+b	Chloro. a/b	Poly-phenol
Winter	Natural	39.44	71.56	111.01	0.55	3.75
	Fresh water	62.35	62.90	125.25	0.99	2.79
Summer	Natural	19.10	52.19	71.30	0.36	4.13
	Fresh water	48.42	51.16	99.59	0.94	3.58
Monsoon	Natural	61.32	111.20	172.53	0.55	5.30
	Fresh water	95.13	116.40	211.53	0.81	4.46

* Chlorophyll values expressed as mg/100 g. fresh w't tissues

* Polyphenol values expressed as g/100 g. fresh w't tissues.

to Karkar (1984) the ratio of chlorophyll a/b in Rhizophora apiculata is 1.74 and Kandelia candel 1.64. In case of E. agallocha. Chlorophyll a/b ratio is less than 1 or near to 1 during all the seasons.

According to Lessage (1891) and Arnold (1955), many halophytes show pale colour which suggests reduction ;in chlorophyll content. The amount of chlorophyll present in the leaves depends on the status of plant nutrition. Excess or deficiency of various mineral elements are known to disturb the development of chlorophyll. The low chlorophyll contents under saline conditions may be due to the deficiency of some ions or the effect of high NaCl which may disturb the mechanism of development of chlorophyll. The role of Mg in the synthesis of chlorophyll is well known (Mishra, 1967). Possibly Mg is responsible for this in case of E. agallocha seedlings. From all above these observations it is seen that chlorophyll synthesis is efficient in the seedlings although grown under fresh water condition.

ii) Polyphenols:

Polyphenols are the important constituents of all the parts of mangrove trees. Monsoon is favourable condition for polyphenol synthesis in mangroves (Bhosale, 1974). It is evident from the table 5 that, in case of E. agallocha monsoon is favourable season for polyphenol synthesis in mangroves. The polyphenol contents of Acrostichum aureum were low in very young leaves, increased as the fern leaflets opened and became mature and decreased in old leaves (Shetty, 1971). Jamale and Joshi (1978) have suggested that the amount of polyphenol present in the leaves of mangroves is dependant upon the age of leaf and polyphenol degradating enzymes. In general mangroves are known to synthesize more polyphenols which are not formed in fresh water plants in such large amounts (Chirputkar, 1969). Table 5 shows that, fresh water seedlings synthesis less polyphenols as compared to natural one. The function of polyphenols in mangroves is unknown (Kulkarni, 1990) Humpries (1967) has reported the polyphenol content upto 35% and stated that the mangroves, in spite of more salt concentration contain large amounts of water soluble tannins. Tannis have strong protein binding capacity and therefore are able to inhibit enzymes. In the living plants it is possible that tannins aid in resistance to fungi (Walsh, 1974).

E. agallocha seedlings under fresh water conditions receives sufficient light and no salinity, hence the values obtained are towards the response to enough light. The role played by light in the synthesis of phenolic compounds is well understood today. The excellent work of Mohr (1963) and Seigelman (1963) clearly demonstrated this. The extensive work of Zucker (1965) indicates the activity of phenylalanine ammonia lyase is induced by light.

VI) Response of male and female cuttings to fresh water conditions:

E. agallocha is commonly found at Ratnagiri and Ganapatipule. It is small glabrous bushy tree. The tree has spreading surface roots which are richly furnished with lenticels. The leaves are simple alternate, obovate to elliptic, glabrous, obtuse. The leaves are green in colour and turns red before falling. During field observation it has been observed that, after the month of December defoliation of leaves starts. According to Bhosale (1990) male and female plants of E. agallocha are separate.There is no major morphological difference in the male and female plants to distinguish between them unless flowered. So it has become necessary to identify male and female plants at the flowering stage only.

Flowering period in the mangroves differs with the species. Most of the species shows flowering in summer (Jones, 1971; Graham et al., 1975; Byrnes et al. 1977). Joshi and Bhosale (1982) reported that E. agallocha is a monsoon flowering estuarine plant i.e. flowering period is from June to August. The flowering is recorded in Excoecaria in a 2 year old plant (Mulik, 1987).The flowering of the mangroves in the Sunderbans can be arranged into several 15 days 'Phase' or 'term'. And the flowering of E. agallocha occurs in 4th phase i.e. from 20th May to 5th June (Chakrabarti and Choudhuri, 1972).

While along the west coast of Maharashtra i.e. at Ratnagiri and Ganapatipule, during field survey, it has been observed that male plants developed inflorescence from April and in female plants the inflorescence develop in the month of June. Real blooming of male and female inflorescence occurred from June to July.At the initial stage of the formation of inflorescence, male and female plants could not be differentiated. So attempt has been made to differentiate between male and female plants in the month of July, whenever real blooming has been observed. During our field study, on the inflorescence anther like and style like structures are observed on different plants. So to confirm it, these have been brought to the laboratory and observed under microscope.

Plate 13a shows mounted pollen grains. Some workers have done the work on palynology of *E. agallocha*. It is dioecious and bears flowers in catkins and possess 2-celled pollen grains (Venkateswarlu and Rao, 1975) and it can be presumed that it is wind pollinated (Clifford and Specht, 1979; Saenger, 1979). The sexine of pollen is as thick as nexine (Chaubal and Kotmire, 1985). Pollen type of *Excoecaria* is autochthonous pollens hence it includes under mangrove group (Blasco, 1984). After confirmation it was observed that male flowers are crowded into slender green catkin, having dark green bracts. 2-3 stamnens are supported on short stipe. Length of filaments are 1-2 mm. long and anthers are yellow in colour.

Female flowers are short having pedicel. Calyx is small consisting 3 acute thick lobes, 3 styles shortly connate at base. Fruit is green to brown in colour, smooth and three lobes. Plate 13a shows, T.S. of ovary like structure having trilocular ovary with single ovule, which confirms it as a female plant. The flowers of Excoecaria are small, certain genera have honey glands, they possess unisexual or polygamous flowers (Van Steenis, 1958).Fruiting stage starts from July and it continues upto August end. The period of seed germination in *E. agallocha* is from May to September (Bhosale and Mulik, 1991). Plate 14a shows the female twig of *E. agallocha* with seeds and Plate 14b shows the twig with male inflorescence.

After confirmation of male and female plants at blooming stage, these have been labelled to observe the response of male and female cuttings under fresh water condition. For this purpose male and female cuttings have been brought to the laboratory separately and it has been decided to study the response of male and female cuttings. It is also observed by Carr, (1970) that auxin induced cell elongation depending upon the availability of some unidentified growth limiting proteins.

Response of female cuttings :

Whenever the male and female cuttings were treated with

75 ppm, 300 ppm, 500 ppm and 1000 ppm IAA respectively, female cuttings showed better response than male cuttings. In all concentrations except at 1000 ppm IAA the male plants show lethal response. However, the response of female cuttings at this concentration is better than that of male. Plate 15 shows the better response of female cuttings at all concentrations than male. Laibach and Kribben, (1950) applied 0.1 % solution in lanolin of IAA which affected the relative number of male and female flowers in the axillary clusters at the first seven nodes of the plant. Addink and Meijer, (1972) found that IAA always induces a temporary increase in growth rate whether in darkness or after irradiation. Laibach, (1952) also concluded that female flowers tend to differentiate under higher concentrations of growth substances than male flowers. Same type of response is found in case of E. agallocha female cuttings, which showed better response.

Plate 16, shows the response of rooting at various concentrations. From this it can be observed that at lowest concentration i.e. at 75 ppm IAA shows good response while at highest concentration i.e. at 1000 ppm IAA the female cuttings showed better response than all the concentrations. However, at 300 ppm and 500 ppm of IAA, the rooting response is poor than lowest and highest concentrations. If the response of female cuttings observed it is better than male.Thimann and Koepfli, (1952) showed synthetic IAA have high activity for root formation on pea stems.

using IAA. Thimann and Lane, (1952) showed that the inhibition of root growth which first appears after auxin treatment is later followed by an acceleration both of elongation and of branching i.e. formation of secondary roots, which would lead to increased total root system. From these observations it can be concluded that the female cuttings with IAA treatment showed better rooting response.

Response of male cuttings :

The response of male and female cuttings at various concentrations of IBA i.e. 300 and 1000 ppm. can be seen in Plate 17a and b. From this it can be observed that both the concentrations of IBA showed better response to male cuttings than the female cuttings. While the response of female cuttings is found to be lethal to both the conc. of IBA. Although the male cuttings showed better response than female, no rooting has been observed even at 1000 ppm IBA.

Brase (1938) has got favourable results of treatment with IAA and IBA on pear and cherry, while this treatment was not successful with several varieties of apple. In case of E. agallocha the IBA treatment is found to be unsuccessful as there is no rooting at all (Plate 17c). Hitchcock and Zimmerman, (1940) have reported that a mixture of IAA and IBA produced better rooting response than when either of them were used separately. So possibly

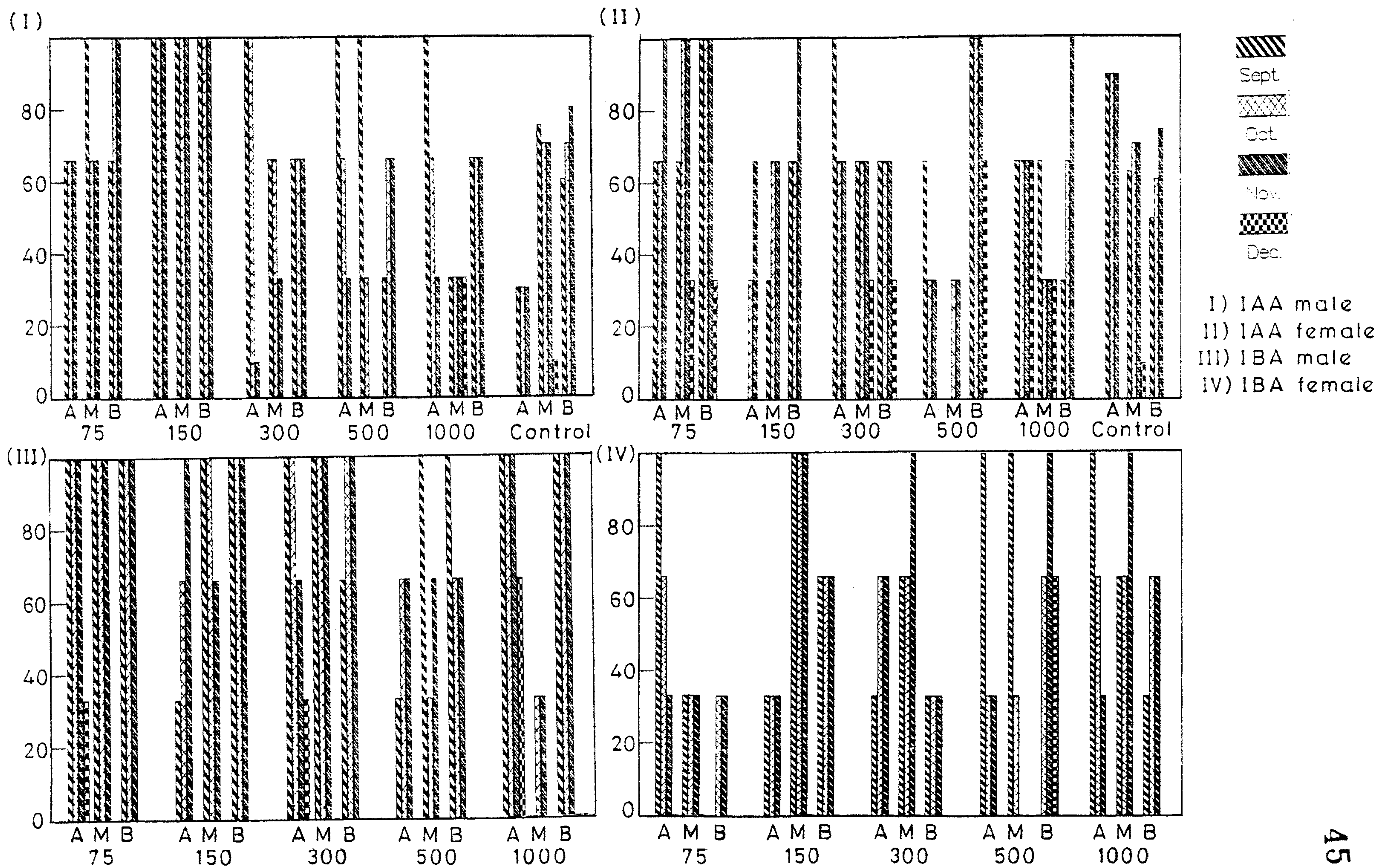


Fig. 7 – Effect of male and female cuttings on sprouting potential.

in case of E. agallocha a mixture of IAA and IBA will give better response to rooting.

Response of male and female cuttings have been also studied in terms of their sprouting potential and survival percentage. From Fig.7 it can be observed that in case of IAA treatment rate of survival and sprouting per cent is more in female cuttings than male. Possibly this may be the reason due to which female cuttings showed better response. In case of IBA treatment, male cuttings showed better response to rate of survival and sprouting per cent than female cuttings.

V) Biomsss Study:

Fire/fuel wood is the most important source of energy in the rurral sector not only in India but also throughout the developing world. The scope and limitation of the terrestrial biomss as an additionl source of energy. The chief objective of energy plantations is to produce biomass from selected tree/shrub species in the shortest possible time at the minimal cost so as to satisfy local energy needs. Bioconversion, as the name implies, is the conversion of biomass and organic wastes into energy (fuels) as also into fertilizer, food and chemicals. These processes/techniques have a vast potential to meet the increasing demand for energy (Khoshoo, 1988).

It was found that conifers, (slow growers) species have good coppicing ability and can be vegetatively multiplied. Multiplication through seed will take longer time and also unisogenic. High regeneration potential or coppicing ability at least 5-6 times after harvest, easy vegetative propagation multiple use like fuel, fodder, fiber etc. (e.g. Leucaena leucocephala) ability to tolerate competition are required. Recent efforts by Calvin (1974, 1978, 1979 and 1980) have been done and found that Euphorbia appear to be a rather promising material. According to Nemethy et al. (1980 a),the annual species E. tirucalli, can produce a biomass of 25 tonnes/hectare/year without any special agronomy. As it is a herbaceous species, the plant as a whole is oven dried and powdered.

As E. agallocha is a euphorbiaceae member, so it was thought worthwhile to study the biomass of it. The cuttings which showed better response to various parameters studied uptill now i.e. L_{20}, 'M' region, keradix and various concentrations of IAA and IBA, i.e. 75, 150, 500, 1000 ppm IAA, 150, 500, 1000 ppm IBA and seedlings. It is decided to remove all above mentioned cuttings and study their biomass by the method of Cintron and Novelli (1984). The length of roots and number of roots have been measured. In this investigation response has been studied in terms of their fresh weight, dry weight and moisture content.

Table 6 : Biomass study of different parts of the cuttings and seedlings.

Type of cuttings/ Seedlings		Fresh weight (g)	Dry weight (g)	Moisture content	No.of roots	Length of roots (cm)
		A) Successfully grown cuttings:				
L_{20}	Roots	5.0	2.9	2.1		
	Shoot	19.2	9.1	10.1	4	17.2
	Leaves	8.6	3.0	5.6		
	Total	32.8	15.0	17.8		
'M' region	Roots	3.3	1.3	2.0		
	Shoot	16.2	8.2	8.0	5	16.7
	Leaves	12.5	4.4	7.8		
	Total	32.0	13.9	17.8		
Keradix	Roots	5.6	2.8	2.8		
	Shoot	26.25	11.3	14.9	18	14.7
	Leaves	5.9	2.0	3.9		
	Total	37.75	16.1	21.6		
		B) IAA Treatment: (ppm)				
75	Roots	2.6	2.1	0.5		
	Shoot	6.1	4.0	2.1	15	12.5
	Leaves	2.4	1.5	0.9		
	Total	11.1	7.6	3.5		
150	Roots	2.5	1.8	0.7		
	Shoot	9.5	4.6	4.9	16	15.4
	Leaves	2.4	1.5	0.9		
	Total	14.4	7.9	6.5		
500	Roots	9.2	8.1	1.1		
	Shoot	12.6	8.6	4.0	23	36.4
	Leaves	2.4	1.2	1.2		
	Total	24.2	17.9	6.3		
1000	Roots	3.6	2.3	1.0		
	Shoot	9.0	5.4	3.6	9	23.8
	Leaves	3.1	1.6	1.5		
	Total	15.7	9.3	6.1		
		C) IBA Treatment (ppm)				
150	Roots	2.1	1.1	1.0		
	Shoot	8.5	5.2	3.3	21	40.5
	Leaves	2.8	1.3	1.5		
	Total	13.4	7.6	5.8		
500	Roots	3.5	1.4	2.1		
	Shoot	12.2	5.1	7.1	4	10.9
	Leaves	3.9	1.8	2.1		
	Total	19.6	8.3	11.3		
1000	Roots	16.5	6.6	9.9		
	Shoot	21.3	10.2	11.1	26	31.2
	Leaves	11.8	5.6	6.2		
	Total	49.6	22.4	27.2		
		D) Seedlings:				
Planted in polythene bag	Toots	12.1	5.4	6.7		
	Shoot	17.6	7.6	10.0	6	43.0
	Leaves	10.0	5.0	5.0		
	Total	39.7	18.0	21.7		
Planted in big pot	Roots	19.4	10.3	9.1		
	Shoot	33.3	16.1	17.2	10	76.5
	Leaves	20.7	12.9	7.8		
	Total	73.4	39.3	34.1		

L_{20}, 'M' region and keradix treated cuttings showed better response in moisture content i.e. 17.8, 17.8 and 21.6. Number of roots are less in 'M' region and L_{20} cuttings, while keradix treated cuttings showed better number of roots i.e. 18. By the comparison between L_{20},'M' region and keradix treated cuttings (Table 6A), Keradix is found to be superior in all respects except length of root which is somewhat less than others.

The biomass, number of roots and length of root of cuttings have been considered at different concentrations of IAA i.e. 75, 150, 500 and 1000 ppm. It is found from the Table 6B that, as the concentration of IAA increases the performance in the parameters such as, fresh weight, dry weight, moisture content, number of roots and length of root increases. While at 1000 ppm IAA it showed less performance.

If the cuttings of 150, 500 and 1000 ppm IBA have been studied it is clear from the Table 6C that, as the concentration increases, there is increase in fresh weight, dry weight and moisture content. While number of roots and length of root is maximum at lower and highest concentration i.e. 150 and 1000 ppm IBA. At 500 IBA it is found to be very less.According to Paul and John, (1991)the length and number of roots were also significantly more in cuttings treated with IBA 1500 ppm i.e. length is 2.2 cm and number of roots are 37.All the untreated cuttings planted in field failed to survive. Bose and Mondal, (1973) has reported better response of percentage rooting with NAA than IBA in some species like Erythrina cristagalli.

Along with the cuttings, if the biomass studied, the number of roots and length of root in case of seedlings have been considered. The results of which are shown in Table 6D. It can be observed that less biomass along with the number of roots and length of roots is found in the seedlings planted in polythene bags. While the seedlings planted in big pots showed very good response in terms of fresh weight, dry weight, moisture content,number of roots and length of root.According to Ustad,(1993) it can be said that, biomass production by the species at seedling stage depends much upon what type of species it is. Further, the contribution of different organs is also specific to the mode of reproduction i.e. in viviparous seedlings original hypocotyl has highest mass which continues further.

From all above observations it can be concluded that in case of cuttings, keradix treatment showed best response than others. The cuttings treated with lower concentrations of IAA showed better response than at highest concentrations i.e. at 1000 ppm IAA.While theree is best response at highest concentration of IBA i.e. at 1000 ppm. The seedlings in big pot showed highest results than the polythene bags.This may be due to the availability of the space to the seedlings. i.e. If the sufficient space is given to the seedlings for its growth it can grow upto full extent giving good biomass.

From all above observations we can conclude that seedlings which showed less percentage of germination under natural conditions could grow successfully with their maximum extent under fresh water condition. There is need to establish test plantations involving different tree/shrub species on marginal lands at present lying fallow in different agroclimates and to attain on their performance. Data are also needed on the rates of production of plant matter per year for different sites and for different species with reference to age and harvest time.

In the recent years, Calvin (1978-80) has advocated study of the petrocrops as a possible feedstock for petroleum like materials. Calvin group has found that the family euphorbiaceae in general and the genus Euphorbia (2000 species) in particular is one that reduces CO_2 beyond carbohydrates. Sachs et al. (1981) reported that the leaves contain the maximum hydrocarbons which is also higher in non-irrigated than in irrigated plants. E. tirucalli, a perennial has also cultivated in California as also in Okinawa, Japan. Nearly 7 cm. long cuttings in an year yield 2 Kg of biomass the yields in Okinawa are higher. The oil from this sp. resembles that of E. lathyris and the yields are 10 barrels of crude plus 25 tonnes of lignocellulose. E. lathyris, hydrocarbon content ranges from 2.91 to 4.4 % which may have an upper limit (Nemethy et al., 1980 b). Recently, Buchanan et al., (1978) have also screened a number of prospective plants and found E. lathyris as one of the euphorbias that is potential.

E. agallocha is the member of family euphorbiaceae having latex, possibly which may be the source of hydrocarbons which will be useful as a petrocrops. However according to Calvin, (1980) 90 % of the co-carcinogens are oxidised during drying and 10 % is neutralised during extraction. The hydrocarbons, from these plants can be grown for the purposes of energy either for direct use or as feedstock for more convenient liquid fuel or other energy chemicals. Plants may be either used to get diesel fuel or after their conversion for high quality liquid fuel.