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PROPAGATION OF SOME ENDANGERED INDIGENOUS TREES FROM THE SOUTH NANDI DISTRICT OF KENYA USING CHEAP, NON-MIST TECHNOLOGY

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ABSTRACT

Vegetative propagation studies were carried out at Maseno University, Kenya in the year 2004 and 2005 using stem cuttings on three endangered indigenous tree species namely Asystasia schimperi, carissa edulis and Toddalia asiatica to test the effect of IBA on rooting of the stem cuttings of these plant species. Juvenile stem cuttings of these plant species were dipped in different concentrations of auxin (indole Butyric Acid (IBA) of 0, 100 ppm, 200ppm, 400ppm and 500ppm. Completely, randomized design (C.R.D) was used and the treatments replicated three times in a non mist polypropagator. The treated cuttings were planted in polythene pots. The duration of the experiment was four months. Data taken were plant height, number of leaves and number of rooted cuttings every 2 weeks. Data was subjected to analysis of variance (ANOVA) and means separated by L.S.D at 5% significance level. The results showed that hormone concentration, species and date of sampling affected the number of rooted plants, plant height and number of leaves Asystasia schimperi had the best rooting and subsequent plant growth followed by Carissa edulis and lastly Toddalia asiatica.

It can be concluded that Asystasia schimperi and Toddalia asiatica can be propagated by stem cuttings easily hence farmers can cultivate them.

Keywords: Non mist polypropagator, propagation, stem cuttings, auxin, concentration, species, endangered.

INTRODUCTION

Wild plants supply food, shelter, clothing, fuel, medicine, crafts and cosmetics to rural and urban areas. In addition, they are sources of income and employment to the rural people who collect and sell natural herbal products to urban and rural area (Olembo, 1995, Kokwaro, 1976 and 1993), important herbal teas, medicinal raw materials, aromatic plants and other flavouring and dietary supplements (Kokwaro, 1976).

Due to high human population growth in South Nandi District of Kenya demand for indigenous herbal products is escalating and some of the important plants have been over harvested which have severely reduced the inventory of these wild resources. Deforestation caused by the need for human settlement and allied infrastructure development; agricultural expansion, charcoal production, timber and overgrazing have further caused the shortage of herbal plants. Deforestation directly reduces the biodiversity of wild plant resources and indirectly so through the loss of habitat creating unfavourable conditions for these plants as well as for other organisms important for ecosystem function (Repetto, 1989).

Demand for herbal products is however, on the increase exerting a lot of pressure on the remaining indigenous medicinal plants. The alarming rate of habitat alteration coupled with unsustainable mode of harvesting medicinal plants, where roots and stem bark are most preferred is a threat to their survival. Strategies must therefore be devised to increase their supply and one of the strategies is vegetative propagation. Thus the objectives of the present study is to investigate the effect of different auxin hormone concentrations on the rooting of stem cuttings of some medicinal plants in S. Nandi District using non-misted low input enclosure (polypropagator)

The hypothesis of the study is that different auxin concentrations promote or increase the rooting of stem cuttings of the endangered indigenous trees which are mainly used as sources of herbal products.

MATERIALS AND METHODS

The experiment of vegetative propagation of the medicinal plants namely *Carissa Edulis* (M1), *Asystasia Schimperi* (M2) and *Toddalia Asiatica* (M3) was done at the botanical gardens of Maseno University, Kenya in a non mist polypropagator. Juvenile stem cuttings were harvested from different medicinal plant species from the wild in Aldai division of South Nandi District and transported to Maseno University where they were kept in a refrigerator. 1 to 4 node cuttings were used depending on species (about 50-60mm) long to facilitate handling and with a leaf area of about 50cm^2. In large leaves species, leaf areas were reduced by trimming prior to severance to reduce water loss and to allow photosynthesis to take place. The basal end of the cuttings were cut right angles and treated with different hormone concentration before being planted into the media. The cuttings were dipped for 12 hours in Roothormone S solutions (0.3 % IBA) to a depth of about 2.5mm before they were planted in a non-mist propagator. The IBA formulation was prepared in liquid form using one litre of distilled water and standard chemical methods used to calculate the different hormone concentrations to be used for the study. To minimize stress, the cuttings were inserted into the non-mist propagator as soon as they were dry. The plant cuttings

were then planted in polythene tubes (17cm by 16cm) filled with compost and sandy soil at a ratio 50:50. An American electric pressure steam sterilizer (model No. 25) was used at 250F. The media was put into polythene bags and placed in the sterilizer for two hours. The media was subsequently removed and spread to cool for two hours and then transported to the experimental site where the cuttings were planted. After planting the cuttings were watered using a watering can and subsequently twice a day (morning and evening). The watering can carried five liters of water. A biweekly assessment was carried out on the cuttings starting two weeks after planting. At each assessment the number of leaves plant heights and the number rooted cuttings were recorded on a tagged plant until the end of the experiment four months later.

Temperature and humidity values were also taken twice a day (morning and evening) using a wet and dry thermometer. In all instances, however, the propagator temperature was 22-27^{0}C. Three plant species (M1, M2 and M3) were grown at different concentration levels (0ppm, 100ppm, 200ppm, 300ppm, 400ppm, 500ppm) were used in the completely randomized design (CRD) with factorial arrangement. The treatments were replicated three times. The non-mist polypropagator used in the study is based on that of Howland (1975), modified by Leakey and Longman (1988) and modified further so that it does not require daily watering. It comprises a wooden frame enclosed in clear polythene so that the base is water tight (Leakey, 1989). The frame also provides support for the enclosed volume of water. The polythene base of propagator is covered in a thin layer of sand to prevent the polythene from being punctured by the large stones (6-10cm), which are placed to a depth of 10-15cm. These stones are then covered by successive layers of small stones (3-6cm) and gravel (0.5-1.0cm) to a total depth of 20cm. The gravel provides support for the rooting medium, which is the upper most layers, while the spaces between the stones are filled with water (Leakey, 1990). The rest of the frame is covered tightly with a single piece of clear polythene and a closely fitting lid as attached. It is a low-cost technology which can be used in the tropical countries compared to the more expensive intermittent mist.

RESULTS

Results on effects on rooting

The date of sampling significantly ($p<0.05$) affected the rooting of cuttings and the subsequent growth of the plantlets (Table-1). This depended on species type. The interaction between species type and weeks after planting was significant. At week 8 the data was unaffected by the date of taking the measurements in M1 and M3 but M2 was significantly affected. The number rooted decreased for the first two weeks and then from week 10 it increased and was maximum at week 12, then decreased up to week 14 then increased sharply up to week 14 then increased sharply up to week 16 (Table-1) Conversely, M3 started rooting at week 10, then increased slightly, maximized at week 14 then increased sharply again (Table-1). The rooting of M1 was unaffected by date of sampling (Table-1). At week 10 and 12 M2 and M3 were significantly affected. M2 had significantly higher rooting percent than M3 at week 10. The same trend continued up to the end of the sampling date (Table-1). Hormone concentration affected rooting of cuttings and this did not depend on weeks after planting (Table-2) but depended on species for M2, there was an increase in rooting from 0-200ppm, then there was a decrease up to 400ppm then an increase up to 500ppm. For M1, there was an increase up to 200ppm, and then more or less constant number of rooted plants then decreases up to 500ppm. For M3, there was an increase.

Table-1. Means for weeks after planting by species interaction on root cuttings Least Squares Means.

wap	Species	Rootcut LSMEAN	Standard Error	Pr > |t|
8	m1	0.44444444	0.26871214	0.0992
8	m2	4.77777778	0.26871214	<.0001
8	m3	0.44444444	0.26871214	0.0992
10	m1	0.44444444	0.26871214	0.0992
10	m2	4.33333333	0.26871214	<.0001
10	m3	1.16666667	0.26871214	<.0001
12	m1	0.27777778	0.26871214	0.3021
12	m2	4.55555556	0.26871214	<.0001
12	m3	1.33333333	0.26871214	<.0001
14	m1	0.16666667	0.26871214	0.5356
14	m2	4.50000000	0.26871214	<.0001
14	m3	1.38888889	0.26871214	<.0001
16	m1	0.38888889	0.26871214	0.1489
16	m2	4.50000000	0.26871214	<.0001
16	m3	1.33333333	0.26871214	<.0001

Table-2. Means for species by hormone concentration interaction on root cuttings

Species	hormone	Rootcut LSMEAN	Standard Error	Pr >|t|
m1	v1	1.04166667	0.23271154	<.0001
m1	v2	1.33333333	0.23271154	<.0001
m1	v3	1.25000000	0.23271154	<.0001
m1	v4	1.16666667	0.23271154	<.0001
m1	v5	0.83333333	0.23271154	0.0004
m1	v6	0.45833333	0.23271154	0.0499
m2	v1	3.62500000	0.23271154	<.0001
m2	v2	3.91666667	0.23271154	<.0001

m2	v3	3.37500000	0.23271154	<.0001
m2	v4	3.20833333	0.23271154	<.0001
m2	v5	4.37500000	0.23271154	<.0001
m2	v6	3.62500000	0.23271154	<.0001
m3	v1	0.29166667	0.23271154	0.2111
m3	v2	1.58333333	0.23271154	<.0001
m3	v3	0.20833333	0.23271154	0.3714
m3	v4	1.20833333	0.23271154	<.0001
m3	v5	1.66666667	0.23271154	<.0001
m3	v6	0.25000000	0.23271154	0.2836

Small increase from 0-100 ppm then a sharp increase to 200pm then a sharp decrease to 300 ppm and then subsequently a steady increase upto 500 ppm (Table-3).

Table-3. Means for weeks after planting by species interaction on number of leaves.

WAP	Species	Leaf no LSMEAN	Standard Error	Pr >|t|
2	m1	1.2777778	2.8829495	0.6579
2	m2	3.2333333	2.8829495	0.2630
2	m3	0.2777778	2.8829495	0.9233
4	m1	4.0000000	2.8829495	0.1664
4	m2	4.1555556	2.8829495	0.1506
4	m3	0.3888889	2.8829495	0.8928
6	m1	3.3333333	2.8829495	0.2486
6	m2	5.3381702	3.0014759	0.0764
6	m3	0.2833333	2.8829495	0.9218
8	m1	1.2333333	2.8829495	0.6691
8	m2	9.2055556	2.8829495	0.0016
8	m3	0.5388889	2.8829495	0.8519
10	m1	2.0722222	2.8829495	0.4729
10	m2	19.5833333	2.8829495	<.0001
10	m3	1.8444444	2.8829495	0.5228
12	m1	1.6722222	2.8829495	0.5623
12	m2	34.8333333	2.8829495	<.0001
12	m3	2.8944444	2.8829495	0.3162
14	m1	1.5555556	2.8829495	0.5899
14	m2	34.3166667	2.8829495	<.0001
14	m3	3.7777778	2.8829495	0.1911
16	m1	1.3888889	2.8829495	0.6303
16	m2	42.5555556	2.8829495	<.0001
16	m3	1.9277778	2.8829495	0.5042

As the hormone increased from 100 ppm to 200ppm there was an increase in rooting percent by about 150% and then it steadily decreased up to hormone concentration of 400 ppm then decreased again and finally decreased at the final level of 500 ppm.

There was no interaction between weeks after planting species and hormone concentration.

Number of leaves

The weeks after planting significantly ($p < 0.05$) affected the number of leaves. This depended on species type, and hormone concentration (Table-4). There was no interaction between weeks after planting and species from week 2 to week 6. Conversely, at week 8, 10, 12, 14, 16 there was an interaction. M2 had the highest number of leaves from week 8 upto 16th week after planting than M1 and M3 (Table-4).

Table-4. Means for species by hormone concentration interaction on number of leaves.

Species	Hormone	Leaf no LSMEAN	Standard Error	Pr >|t|
m1	v1	1.2500000	2.4967075	0.6170
m1	v2	1.7500000	2.4967075	0.4839
m1	v3	5.9000000	2.4967075	0.0188
m1	v4	1.9166667	2.4967075	0.4433
m1	v5	1.0416667	2.4967075	0.6768
m1	v6	0.5416667	2.4967075	0.8284
m2	v1	13.2161276	2.5740764	<.0001
m2	v2	22.1666667	2.4967075	<.0001
m2	v3	17.9583333	2.4967075	<.0001
m2	v4	7.4708333	2.4967075	0.0030
m2	v5	40.8958333	2.4967075	<.0001
m2	v6	13.2083333	2.4967075	<.0001
m3	v1	0.1666667	2.4967075	0.9468
m3	v2	1.8791667	2.4967075	0.4523
m3	v3	0.1666667	2.4967075	0.9468
m3	v4	2.5083333	2.4967075	0.3159
m3	v5	4.0208333	2.4967075	0.1084
m3	v6	0.2083333	2.4967075	0.9336

There was no effect of the hormone concentration on the number of leaves for M1 except at V3 (300 ppm). (Table-5) For M2 there was an increase in number of leaves from V1 to v2 from v3 (300 ppm) to V4, there was decrease and then increase at V5 then increase. For M3, there was no effect of the hormone on the number of leaves. There was no 3-way interaction between weeks after planting species and hormone concentration between weeks after planting, species and hormone concentration (Table-4).

Plant height

Weeks after planting significantly ($p < 0.05$) affected plant height and this depended on species type and hormone concentration. As the weeks after planting increased M2 increased slightly up to week 14-16. M3 had slight increase in height from week 10 upto week 16 (Table-5). M1 decreased upto week 8, then death of the plants.

Table-5. Means for weeks after planting by species interaction on plant height.

Wap	Species	Plantht LSMEAN	Standard Error	Pr >|t|
2	m1	0.9000000	1.1036795	0.4155
2	m2	1.8833333	1.1036795	0.0890
2	m3	0.3055556	1.1036795	0.7821
4	m1	4.2222222	1.1036795	0.0002
4	m2	7.7444444	1.1036795	<.0001
4	m3	0.4777778	1.1036795	0.6654
6	m1	2.2500000	1.1036795	0.0424
6	m2	9.6500000	1.1036795	<.0001
6	m3	0.8333333	1.1036795	0.4508
8	m1	0.3362859	1.1490550	0.7700
8	m2	10.9333333	1.1036795	<.0001
8	m3	1.1388889	1.1036795	0.3030
10	m1	1.3333333	1.1036795	0.2280
10	m2	11.8777778	1.1036795	<.0001
10	m3	2.3222222	1.1036795	0.0362
12	m1	0.9666667	1.1036795	0.3818
12	m2	12.5055556	1.1036795	<.0001
12	m3	2.1333333	1.1036795	0.0542
14	m1	1.0388889	1.1036795	0.3474
14	m2	13.6500000	1.1036795	<.0001
14	m3	2.3722222	1.1036795	0.0324
16	m1	0.8722222	1.1036795	0.4300
16	m2	26.8222222	1.1036795	<.0001
16	m3	2.7722222	1.1036795	0.0126

For M1 hormone concentration affected plant height at 300 ppm. However, for other rates there was no effect M2 was affected significantly up to the level from v1 to v4 but was decreasing then increased at v4 to v3 sharply. For M3, V4 and V5 increased plant height significantly up to V5 then decreased (Table-6).

Table-6. Means for species by hormone concentration interaction on plant height.

Species	Hormone	Plantht LSMEAN	Standard Error	Pr >|t|
m1	v1	0.8000000	0.9558145	0.4033
m1	v2	1.1375000	0.9558145	0.2350
m1	v3	4.3125000	0.9558145	<.0001
m1	v4	1.4730478	0.9854336	0.1361
m1	v5	0.4583333	0.9558145	0.6319
m1	v6	0.7583333	0.9558145	0.4282
m2	v1	11.5291667	0.9558145	<.0001
m2	v2	10.7083333	0.9558145	<.0001
m2	v3	9.7458333	0.9558145	<.0001
m2	v4	4.7500000	0.9558145	<.0001
m2	v5	23.9291667	0.9558145	<.0001
m2	v6	10.6375000	0.9558145	<.0001
m3	v1	0.1250000	0.9558145	0.8960
m3	v2	2.0375000	0.9558145	0.0339
m3	v3	0.4000000	0.9558145	0.6759
m3	v4	3.2041667	0.9558145	0.0009
m3	v5	3.3750000	0.9558145	0.0005
m3	v6	0.1250000	0.9558145	0.8960

DISCUSSIONS

The results of the present study showed that auxin promote the rooting of stem cuttings of three l plants species tested. Several workers have reported promotion of rooting by auxins in other plant species (Hartmann *et al.* 1995; Tchoundjeu and Leakey, 1996; Haissig and Davies, 1994; Cope and Mandel, 2000). The results also indicate that the species M1, M2 and M3 have different ranges of effective auxin concentration with the latter two having broader range than M1. This agrees with Leakey, 1990 who reported that increase in auxin concentration increases rooting as in this study and that auxin or IBA has abroad range of root enhancing activity. Other works have also reported root formation enhancing ability of auxin (Lundquist and Torrey, 1984; Husen and Mohinder, 2006; Leakey, 1990).

For species M1, there was a range of increased rooting from 0 to 200 ppm, then decrease up to 400 ppm then increase. This implies the rates from 200ppm to 400ppm were too high and killed the cells. When auxins are too high they are injurious to the cells (Tchoundjeu, 2002). High levels of IBA (300mg) were supraoptional in the rooting of peaches but 200mg promoted rooting (Tchoundjeu, 2002). In the present study 0-200 ppm promoted rooting in M1 while 300-400 ppm was supraoptional for rooting. This increase in rooting at 500 ppm is isolated case and therefore difficult to explain. For M2, the range of root promotion was from 0-200 ppm and

at the rest of the concentration rooting decreased slowly. So it can be reasoned that the optional levels of the auxins for root promotion was very small compared to the supraoptional levels where rooting was reduced. This contrasts sharply with M3 where the ranges of auxin for root promotion were more than in M1 and M2. Therefore M3 appears to be easy-to-root species. M1 and M2 which are difficult to root in this study may have endogenous rooting inhibitors (Cuir *et al.,* 1997; Vietz *et al.,* 1987, Biran and Halev, 1973). Such inhibitors have been reported to be phenolic compounds (Biran *et al.,* 1973) and manganese (Jarvis, 1986). M3, because it was easy-to- root in the present study did not have these inhibitors but had essential root promoting substances called morphogens and auxins which may have been lacking in M1 and M2 (Fadl and Hartmann, 1967). For M2 and M3, there was an increase in rooting with date of planting upto week 14 then increase. This shows that the effect of the auxin concentration was decreasing with time. This could be due the breakdown of the auxins by microorganism or effect of continuous watering of the cuttings, which may have leached it. In species M2, increase in concentration of auxin generally increased the number of leaves and so was plant height, with few exceptions for M2 and M3 but not M1. This can be attributed to the fact that auxins may have mobilized carbohydrates and borons from the leaves, which promote growth activities (Patrick and Warping, 1973, Altaman and Wareing, 1975; Booth *et al.,* 1962). With increased date of sampling there was reduction in plant height. In M1 which eventually died. It appears that the auxins applied may have added to the endogenous auxins which killed the cells with time because they were too much.

For M2 and M3, it appears that the mobilization of carbohydrates and Boron increased with time and this promoted growth more. For the species which had difficulty in rooting in the present study it may have been due to seasonal effects where rooting could be high in some months but lower in others (Roberts and Fuchigami, 1973).

CONCLUSIONS

It can be concluded that Indole Butyric acid (IBA) can be used to root cuttings of species M1, M2 and M3. The concentrations to be used optimally for all of them should be from 100-400 ppm. It can also be concluded that species M2 and M3 are easier to root than M1 with M3 being the best. Finally, it can also be concluded that the non mist poly propagator be used to propagate M1, M2 and M3 because it promotes good rooting and it can be constructed from easily available and cheap materials. It is therefore recommended that IBA be used for rooting of Juvenile cuttings in a non-mist polypropagator at the rates of 100 ppm to 400 ppm.

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