

Vegetative propagation of *Inga feuillei* from shoot cuttings and air layering

ERIC B. BRENNAN* and KENNETH W. MUDGE

Department of Floriculture and Ornamental Horticulture, Cornell University, Ithaca, NY, USA (* Present address: Department of Pomology, University of California, Davis, CA, 95616)

Accepted 17 February 1997

Key words: cuttage, auxin, marcottage, mist, polyethylene

Application. The stem cutting and air layering techniques described here, may be useful for increasing the supply and year round availability of *Inga feuillei* D.C. which is limited due to viviparous germination and recalcitrant seeds. These techniques can be applied on the farm and/or nursery-level, and may also be useful in genetic improvement and conservation efforts.

Abstract. Cuttage and air layering were evaluated as means of vegetative propagation of the tropical woody tree, *Inga feuillei*. Effects of moisture management systems, leafiness, auxin application, and stem diameter on rooting of semihardwood cuttings were investigated. Cuttings were taken from 2-year-old seed-propagated, greenhouse-grown stock plants. Compound leaves were either reduced in area or removed entirely before auxin pretreatment with 0, 0.3, or 0.8% indolebutyric acid (IBA) followed by sticking under mist or in a polyethylene enclosure. Leafless cuttings did not root regardless of moisture management system or auxin pretreatment, whereas 55% rooting of leafy cuttings was observed. Leafy cuttings rooted significantly better under mist than in a polyethylene enclosure. Auxin treatment at the higher level increased rooting percentage approximately two fold for larger diameter cuttings (8.1 to 20 mm), but had no effect on the smaller cuttings (2 to 8 mm), and resulted in an approximately three fold increase in the number of roots/rooted cutting for both stem diameter classes. Auxin treatment did not affect rooting percentage of leafy softwood cuttings under fog, however it did increase the number of roots per rooted cutting. One hundred percent of air layered shoots rooted within 5 weeks with or without auxin pretreatment, and all rooted layers survived transplanting to soil. Possible implications of this research on agroforestry, selection, genetic improvement, and conservation are discussed.

Introduction

Inga Mill. is a large neotropical genus in the Leguminosae which includes about 350 species of woody plants (Lewis 1988) that are indigenous from the Plata River delta in Argentina (34° S) to central Mexico (25° N) (Leon 1966). Species of *Inga* range in size from 3 m tall (*I. cookii* Pittier.) to 40 m (*I. altissima*) (Leon 1966).

Inga is known by various common names including ice cream bean, inga (Brazil), pacay (Peru), guamo (Columbia, Venezuela) and vanilla (Mexico)

(Leon 1966). Use of *Inga* in agroforestry systems in Latin America dates back to pre-Colombian times when the trees were used as shade for cacao (*Theobroma cacao* L.) (Leon 1966). *Inga feuillei* spread from its center of domestication (Peru) during the Inca Empire, extending from Chile and Bolivia to Ecuador (Yacovleff and Herrera 1934 cited in Leon 1966). Extensive cultivation of *Inga* and other nitrogen fixing tree species as shade for coffee (*Coffea* sp. L.) in Latin America is well documented (Leon 1966; Palm and Sanchez 1990; Pursglove 1974; Roskoski 1982; Van Kessel and Roskoski 1981, 1983). Studies in coffee plantations report that *I. jinicuil* Schlecht. can fix 35 kg N/ha/year (Roskoski 1981). *Inga* spp. provide fuelwood (Leon 1966), timber, honey bee forage, and human food (National Academy of Science 1980). Species within the genus are adapted naturally to a wide range of soil moisture conditions (including flooded soils) (Leon 1966) and acidity, from pH 4.5 to limestone soils (Denslow et al. 1991; National Academy of Sciences 1980). The immense diversity within the genus could undoubtedly benefit agroforestry systems worldwide.

Inga feuillei and other species such as *I. edulis* Mart., *I. vera* Willd., *I. jinicuil* and *I. paterno* Harms. are prized for the sweet edible aril that surrounds the naked, fleshy seeds. In addition, the seeds of some species are cooked and eaten as vegetables (Leon 1966). Although the use of ingas are largely restricted to areas where they are grown, their fruits (pods) and those of other relatively unknown exotic species may soon be exported to the United States (Anonymous 1989).

Inga trees are commonly propagated by seed (National Academy of Sciences 1980; Nichols 1990). Like many tropical trees, the seeds of this genus are recalcitrant, i.e. seed cannot withstand chilling or drying, and hence cannot be stored for more than a few weeks. Also, in many cases viviparous germination occurs (Leon 1966). These characteristics result in unavailability of seed during much of the year and this presents obstacles to distribution and domestic use of the genus. Because of these limitations to seed propagation, vegetative propagation of *Inga* could increase its utility. There are no known reports in the literature of clonal propagation for any species of *Inga*. Therefore, the overall objective of this study was to determine if vegetative propagation by shoot cuttings and air layering could be developed as a practical alternatives to seed propagation. With respect to cutting propagation, experiments were conducted to test the effects of moisture management, auxin pretreatment, cutting diameter, leafiness, and degree of woodiness. The effect of auxin pretreatment was evaluated for air layering.

Materials and methods

In August 1993, seeds were collected from an isolated tree of *I. feuillei* growing on the north shore of O'ahu, Hawai'i. The seeds were wrapped in moist sphagnum moss and transported to Cornell University (Ithaca, New York) where they were sown in a greenhouse in 30-cm diameter pots in a potting medium of 1 soil:2 peat:1 perlite (by vol.). Eight seedlings were grown as stock plants for cuttings. After 8 months, the 2 m tall seedlings were coppiced 3 cm above the soil line and prunings were used to prepare 20 cuttings per seedling for use in a preliminary rooting experiment. In this preliminary experiment, leafy cuttings were treated or not with 0.3 or 0.8% indole-3-butyric acid (IBA) in talc (Hormex 2 and Hormex 3 respectively, Brooker Chem. Corp., North Hollywood, CA). Approximately 30% of untreated cuttings and those treated with 0.3% IBA (Hormex 2) rooted, while 73% of those treated with 0.8% IBA (Hormex 3) rooted. Rooted cuttings (approx. 150) from this experiment were potted and grown, along with the original coppiced seedlings, for 6 months (until October 1994) to generate approximately 1300 single-node semihardwood leafy cuttings which were used in experiments described below. Stock plants from which cuttings were obtained for these experiments appeared to be in the juvenile growth phase since flowering was not observed at any time during greenhouse culture.

Experiment 1: Effects of moisture management, auxin treatment and shoot leafiness on rooting of semihardwood cuttings. The experiment, conducted in October, 1994, was a randomized complete block design with a 3 way factorial arrangement of treatments: 2 moisture management systems (MMS) (either intermittent mist or enclosure of cuttings within a polyethylene tent), 3 levels of auxin (A) (0, 0.3 and 0.8% IBA); and 2 foliation levels (leafy or leafless). The 3 blocks included sections along the length of a propagation bench. The experimental unit (EU) was a single propagation flat, filled with perlite rooting medium, containing 33 cuttings varying in stem diameter (SD) from 2 to 20 mm. Cuttings were completely randomized within a flat with respect to stem diameter and care was taken to assure that cuttings of the entire range of stem diameters were equally represented in all flats. Individual flats held cuttings of only one treatment, i.e., a single combination of auxin, leafiness, and moisture management system. Within each block there were 12 propagation flats, one for each of the 12 treatments.

Single node cuttings were taken and the single pinnate leaf at each node was trimmed to leave the rachis and the basal (proximal) pair of leaflets. Cuttings ranged in stem diameter from 2 to 20 mm and were 12 to 15 cm long with a single dormant axillary bud at the distal end. Semihardwood cuttings

included those with both green and brown stems, with the latter indicating greater lignification.

Immediately prior to sticking, cuttings assigned to the leafless treatment groups had their single pair of leaflets removed. Auxin was applied by dipping 1.5 cm of the premoistened basal end of each cutting in the powder formulation of either 0, 0.3%, 0.8% IBA. Following auxin treatment each cutting was tapped gently to remove excess powder. Cuttings were stuck with the aid of a dibble, 2 to 3 cm deep in the 10 deep × 36 wide × 51 cm long plastic Dynaflats (Hummert International, St. Louis, MO) filled with moist perlite. For the polyethylene tent moisture management treatment, a 25 high × 36 wide × 51 cm long wire frame was attached to the propagation flat. After thoroughly misting the cuttings, the entire flat plus frame was enclosed in a clear 1.1 mil polyethylene bag, which was sealed with a twist tie. The flats in both the polyethylene and mist treatments were placed under intermittent mist, with the polyethylene bag isolating the enclosed cuttings from the mist. The interval between 6 second mist cycles ranged from 2 to 20 minutes and was controlled by an electric timer adjusted as appropriate for daily solar radiation. Day/night greenhouse temperatures were approximately 21 to 24 °C during the day and 18 to 20 °C at night. Temperatures within the polyethylene enclosure (under mist) and under the open mist system under full sun at mid day were 25° and 23 °C respectively. The polyethylene enclosed flats remained closed during the entire 8 week rooting period. All cuttings were shaded using a single layer of 40% shade cloth. The natural day length at the beginning of the experiment was 12 hours; 8 weeks later, at the end of the experiment, it was 9 hours.

Experiment 2: Effect of alternative media on rooting of semihardwood cuttings. The objective of this experiment was to compare the effects of soil and sand media on the rooting of leafy, semihardwood cuttings, since these media might be more readily available than perlite in most on-farm and many nursery applications of this propagation technology. Leafy cuttings with stem diameters between 2 and 8 mm were treated with 0.8% IBA and stuck in separate misted propagation benches filled 20 cm deep with either soil or sand. The soil was a sandy loam consisting of 26.3% gravel, 35.7% sand, 19.9% silt, and 18.1% clay. The particle size distribution of the sand was 28.7% very coarse, 48.1% coarse, 15.9% medium, 6.6% fine, and 0.7% very fine. In the soil medium, 45 cuttings were randomized among 5 rows (4 rows of 10 and one row of 5). In the sand medium, 30 cuttings were randomized among 3 rows of 10 cuttings each. For this experiment the EU was the row of cuttings.

Experiment 3: Effect of auxin treatments on rooting of softwood cuttings in a fog propagation system. Three weeks after initiating experiments 1 and 2, single-node softwood terminal cuttings were taken from the regrowth of the stock plants from which cuttings had earlier been harvested. The single compound leaf of each softwood cutting was trimmed to a single pair of leaflets. Softwood cuttings stem diameters ranged from 1.9 to 3.4 mm (2.6 mm average) and were 10 to 12 cm long. Cuttings were randomized among three auxin levels (0, 0.3% and 0.8% IBA in talc) and stuck 2–3 cm deep in a propagation bench filled with perlite to a depth of 20 cm. The EU was a row of 17 cuttings and there were 3 rows per treatment. In contrast to the previous experiments, all cuttings were placed in a fogging chamber consisting of 4 mil polyethylene stretched over a 45 high × 90 wide × 115 cm long wood frame. This chamber was attached to an automatic electric ultrasonic (cool mist) humidifier (Goldstar Ultrasonic Humidifier, GH-552UA, Goldstar Inc., Long Island City, NY) through a 20 cm long section of 6 cm diameter plastic tubing. The entire experiment was shaded as previously described.

Experiment 4: Air layering. This experiment was conducted to determine if air layering (marcottage) of semihardwood stems could be used as an alternative clonal propagation method for *Inga*. The experiment was a completely randomized design with two treatments: air layering with and without IBA (0.3%). There were 30 air layers (replications) for each of the two treatments. Air layering was done in May, 1994, in the greenhouse on 1 to 2.5 cm diameter leafy branches, of 1 to 3 m tall stock plants grown from rooted cuttings from previous experiments. Most branches had only one air layer of a given treatment, however, in a few cases (i.e. with branches longer than 2 m), more than one air layer of either treatment occurred.

An air layering technique commonly used by farmers in Thailand (Brennan, unpublished observation) was used in this experiment because it is easier and more quickly performed than the conventional method (Hartmann et al. 1990). In this case a clear plastic bag (9×20 cm) was filled with premoistened sphagnum moss and tied shut in advance. At the time of layering, the moss-filled bag was slit open lengthwise on one side, to expose the moss, and the bag and moss were molded around the 1.5 cm of girdled stem. Adhesive tape was then wrapped around the moss-filled bag to secure it to the stem.

Cutting evaluation. Eight weeks after the initiation of experiments 1 and 2, the semihardwood cuttings were carefully lifted from the rooting media and scored either as rooted (at least 1 root 2 mm long), alive but not rooted, or dead. Stem diameter for each cutting was measured with a dial caliper (Mitutoyo, Japan). The number of roots per rooted cutting (RRC) was counted for cuttings

with less than 5 roots. For cuttings with more than 5 roots, RRC was estimated to the nearest five as 5, 10, 15 or 20. All rooted cuttings were transplanted into a 1 soil:2 peat:1perlite (v/v) potting medium in 10 cm plastic pots. After 3 days under mist and 70% shade, the transplanted cuttings were moved from the mist into a green house with full sun. Supplemental lighting on a 13 hour photoperiod was provided by 400-watt high pressure sodium lamps placed 3 m above the transplanted cuttings. The softwood cuttings under fog (experiment 3) were evaluated at 5.5 weeks instead of 8 weeks, but otherwise as described for experiments 1 and 2.

Air layer evaluation. Nondestructive evaluation of rooting (presence or absence), as observed through the moss-filled clear polyethylene bags was made at 3, 5, and 8 weeks after air layering. After 8 weeks, rooted layers were excised below the moss filled bag and transplanted into pots filled with the 1:2:1 potting medium.

Statistical analysis

For experiment 1, the percentage of rooted cuttings (%R) and mean RRC were determined for each flat. Percent rooting refers to the cuttings that rooted and survived transplanting (98–100% of the rooted cuttings survived transplanting). Prior to analysis, %R data were transformed by the Arcsine $\sqrt{\quad}$ to homogenize the variances, since the range exceeded 40% (Little and Hills 1978). Analysis of variance (ANOVA) was performed to determine differences in %R (Arcsin $\sqrt{\quad}$) and RRC. The initial ANOVA (Table 1) compared leafy and leafless cuttings based on the response of all 33 cuttings within a flat without regard of stem diameter. Because a very low percentage of leafless cuttings rooted in this experiment ($\leq 2\%$), and because there appeared to be a difference in rooting between small diameter and large diameter leafy cuttings, the data from leafy cuttings only were reanalyzed after assigning each cutting to a small (0–8 mm) or large (8.1–20 mm) stem diameter class. Within each flat of 33 cuttings, approximately 10 were in the large stem diameter class and the rest were in the small stem diameter class. Orthogonal contrasts of interest were constructed to partition the degrees of freedom (df) for auxin level into single df contrasts to compare %R and RRC between the various auxin treatments.

Stem diameter was not considered in experiments 2 and 3 since cuttings were all within the small diameter stem diameter class. Percent rooting (Arcsin $\sqrt{\quad}$) and RRC data were analyzed as for experiment 1. The GLM procedure in Minitab statistical software (Minitab 1994) was used for the analysis of each experiment.

Table 1. Effect of moisture management system (MMS), leafiness (L), and auxin treatment (A) on %R of semi-hardwood cuttings of *I. feuillei* (experiment 1), with accompanying ANOVA.

MMS/auxin (A)	Leafy		Defoliated	
	Mist	Poly.	Mist	Poly.
0%	52.5 ± 7.9 ^a	48.5 ± 6.3	1 ± 1	0
0.3%	62.2 ± 7.4	46.5 ± 2.7	0	1 ± 1
0.8%	77.8 ± 2.7	39.4 ± 9.1	1 ± 1	2 ± 1
Mean	65.4 ± 4.9	44.7 ± 3.6	0.6 ± 0.4	1.0 ± 0.5

ANOVA of %R of semihardwood leafy and leafless cuttings			
Source	df	F ^b	p > F
Block	2	1.7	0.210
MMS	1	9.0	0.007
L	1	572.4	0.000
A	2	1.6	0.223
MMS × L	1	12.9	0.002
MMS × A	2	1.4	0.280
L × A	2	0.3	0.751
MMS × L × A	2	4.5	0.023
MSE	22	0.0099	
Total	35		

^a Mean ± 1 standard error, averaged across all stem diameter classes;

^b Mean squares, used to calculate F-values, based on Arcsine $\sqrt{\%R}$.

Results

Cuttage experiments

Rooting. Leafiness of cuttings was highly significant, with almost no rooting ($\leq 2\%$) of leafless cuttings compared with 45–65% R for leafy cuttings across all auxin levels (Table 1). Moisture management system was also significant across all auxin levels, with 65% of the leafy cuttings rooting under mist compared with only 45% under polyethylene.

Due to the poor rooting response of leafless cuttings, and an apparent differential response between smaller and larger diameter cuttings, in experiment 1, the %R and RRC data from leafy cuttings were reanalyzed after assigning each cutting to a small (2–8 mm) or large (8.1–20 mm) stem diameter class. From Table 2 (ANOVA) and Table 3 (treatment means) it is apparent that MMS continued to have a significant effects on %R, but

not RRC, with somewhat higher %R under mist than under polyethylene. Auxin application significantly increased RRC of semihardwood (Tables 2 and 3, experiment 1) and softwood cuttings (Tables 3 and 4, experiment 3). Although %R was higher in auxin treated semihardwood cuttings under mist, regardless of stem diameter, auxin stimulation of %R was significant only in larger diameter cuttings under polyethylene, accounting for the significant $A \times SD$ interaction in experiment 1 (Table 2). This indicates that, overall, the larger (more difficult to root) but not the smaller stem diameter semihardwood cuttings responded positively to auxin application, as illustrated in Figure 1A. On the other hand, RRC of semihardwood cuttings was increased by auxin application in both stem diameter classes (Figure 1B) regardless of moisture management system (Table 2, Table 3). From orthogonal contrasts (Table 2) it is evident that %R was significantly higher in cuttings treated with either concentration of IBA compared to the untreated cuttings, but %R did not differ significantly between the 0.3% and 0.8% IBA levels. In contrast, for softwood leafy cuttings rooted under fog (Experiment 3), only RRC, but not %R, was significantly increased with auxin application (Tables 3 and 4). Although not compared directly to other MMS (mist, poly) %R and RRC were satisfactory in the fog system (Experiment 3, Tables 3 and 4). It is apparent from Experiment 2 that rooting in soil and sand was comparable (Table 3), and, although not compared directly in the same experiment, rooting in perlite appears similar (Experiments 1 and 3, Table 3).

Root system morphology. Rooting medium affected the morphology of the adventitious root system of misted cuttings treated with 0.8% IBA (data not shown). The roots of cuttings in perlite and sand (Table 3, experiments 1 and 2) were uniformly distributed along the 1.5 cm basal end of the cutting where the auxin was applied. In contrast, about half of the cuttings rooted in soil, had roots forming only near the soil surface; the remaining basal portion of the cutting was rotted, probably due to water logging.

Leaf retention and shoot growth. Upon transfer from the propagation bench, approximately half of the rooted semihardwood cuttings had an actively growing lateral shoot from the bud present at the time of cutting (shoots ranged from 1–5 cm long). Nearly all of the rooted leafy cuttings retained the trimmed single pair of leaflets up to 3 months after transplanting. All rooted cuttings grew vigorously after transplanting.

Air layering

Three weeks after air layering, 97% of the auxin treated layers, and 76% of the control layers had visible roots. By week 5, 100% of both treatments

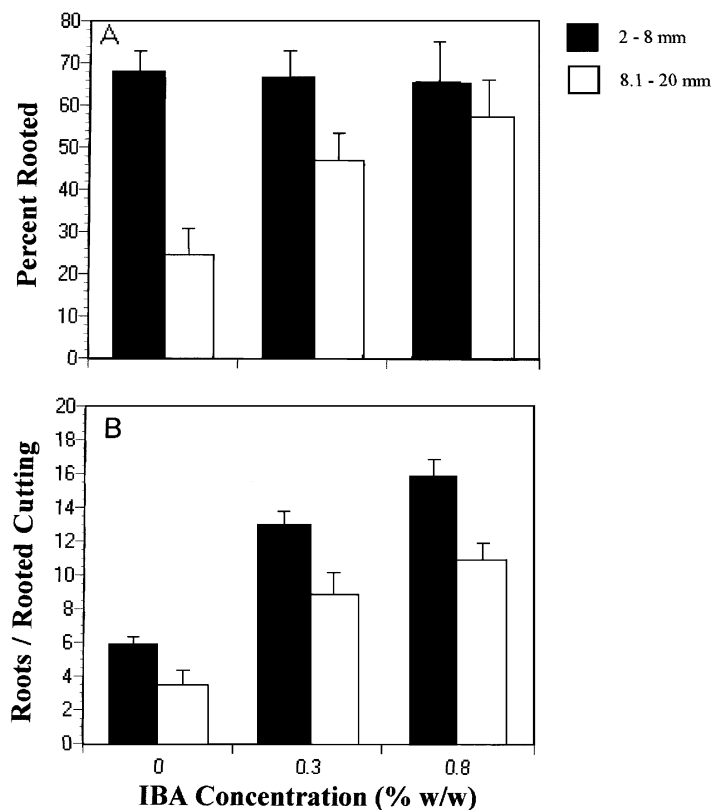


Figure 1. The effect of auxin (IBA) concentration and stem diameter on %R (A) and RRC (B) of leafy cuttings of *I. feuillei* for both mist and polyethylene moisture management systems (combined) from experiment 1.

had roots. When the layers were harvested (after 8 weeks) there were no differences in the number of roots per layer between the two treatments, and all air layers transplanted successfully.

Discussion

Results of this study suggest the presence of at least a single pair of leaflets on semihardwood cuttings is a prerequisite for adventitious rooting of *I. feuillei* in both the mist or polyethylene moisture management systems. The fact that leafless cuttings failed to root indicates that some leaf-associated factor is necessary for adventitious rooting. Davis (1988) concluded that photosynthesis promotes adventitious root formation with most types of leafy cuttings. None the less, he also cited cases where leafy cuttings rooted better

Table 2. ANOVA for %R and RRC for leafy cuttings of *Inga feuillei* in the two stem diameter classes (experiment 1).

Source	df	% Rooting		RRC	
		F ^a	p > F	F	p > F
Block	2	0.0	0.971	0.4	0.709
Auxin (A)	2	3.4	0.051	39.0	0.000
<i>Contrasts</i>					
Control vs Auxin	1	6.3	0.020	71.5	0.000
0.3% vs 0.8% IBA	1	0.5	0.478	6.6	0.018
MMS (Mist vs Poly.)	1	4.4	0.049	0.2	0.629
SD (2–8 vs 8.1–20 mm)	1	18.6	0.000	21.8	0.000
A × MMS	2	1.9	0.175	0.3	0.758
A × SD	2	3.8	0.038	0.8	0.449
MMS × SD	1	0.4	0.534	0.2	0.667
A × MMS × SD	2	2.0	0.157	0.9	0.417
MSE	22 (21) ^b	0.0332 (5.8) ^c			
Total	35 (34) ^d				

^a Mean square terms, used to calculate F-values, based on arcsine $\sqrt{\%R}$

^b Error degrees of freedom for %R (for RRC)

^c Error Mean Square for %R (for RRC)

^d Total degrees of freedom for %R (for RRC)

than leafless cuttings even in the dark, so other factors besides photosynthesis could be involved.

Auxins such as IBA enhance rooting of cuttings of a wide range of species which have at least some natural tendency to root (Blazich 1988). That *I. feuillei* has this tendency is evidenced by 100% success with air layering regardless of auxin treatment, and greater than 50% rooting of leafy, smaller diameter control cuttings (Table 3). The stimulation of %R of semihardwood cuttings by auxin in all treatment combinations except small diameter cuttings under polyethylene clearly indicates that auxin application, if available, would be justified in practical applications of this technology. The positive effects of auxin on RRC had no influence on post propagation survival since nearly 100% of rooted cuttings survived transplanting to the greenhouse regardless of treatment.

Differences in the root system morphology of cuttings rooted in perlite and sand compared with soil was observed, with those in soil frequently having rotted bases, which may be due to water logging and reduced aeration. Nonetheless, cuttings rooted well (82%) in soil and survived transplanting as well as those rooted in better-aerated media (perlite and sand).

Table 3. Effect of rooting medium, stem diameter (SD), moisture management system (MMS), and auxin on %R and RRC of leafy cuttings of *I. feuillei*. Semi-hardwood cuttings were used in experiments 1 and 2 and softwood cuttings were used in experiment 3.

Expt	Rooting Medium	SD (mm)	MMS	Auxin level (%IBA)		
				None	0.3	0.8
1	Perlite	2–8	Mist	62.1 ± 6.7 ^a	74.5 ± 7.7	86.2 ± 3.2
				11.2 ± 2.0 ^b	13.4 ± 1.2	15.8 ± 2.0
1	Perlite	8.1–20	Mist	23.3 ± 3.3	55.0 ± 7.9	60.0 ± 5.0
				3.8 ± 1.5	7.2 ± 0.9	11.2 ± 2.0
1	Perlite	2–8	Poly.	73.5 ± 6.8	59.3 ± 8.9	45.5 ± 5.6
				6.1 ± 0.4	12.7 ± 1.0	16.1 ± 1.8
1	Perlite	8.1–20	Poly.	25.9 ± 13.4	39.4 ± 8.9	55.5 ± 17.8
				3.0 ± 0.4	10.4 ± 2.3	10.8 ± 1.1
2	Soil	2–8	Mist	na	na	82.3 ± 5.4
						13.3 ± 0.7
2	Sand	2–8	Mist	na	na	76.7 ± 6.6
						18.2 ± 0.6
3	Perlite	2–3	Fog	52.9 ± 1.7	78.4 ± 5.2	66.7 ± 10.4
				5.8 ± 0.5	18.0 ± 0.6	17.5 ± 1.0

^a %R ± 1 standard error;

^b RRC ± 1 standard error.

na is not applicable.

Table 4. ANOVA for %R and RRC of softwood leafy cuttings of *Inga feuillei* in perlite under fog (experiment 3).

Source	df	% Rooting ^a		RRC	
		F	p > F	F	p > F
Block	2	1.2	0.398	0.1	0.954
Auxin (IBA)	2	1.2	0.310	67.3	0.001
<i>Contrasts</i>					
Control vs Auxin	1	1.9	0.245	134.2	0.000
0.3 vs 0.8% IBA	1	0.6	0.503	0.3	0.621
MSE	4	0.0473			
Total	8				

^a Mean square terms, used to calculate F-values, based on arcsine $\sqrt{\%R}$.

Moisture management system, via its influence on cutting water relations, has a major effect on adventitious root formation on cuttings of many species (Leakey et al. 1990; Loach 1988; Mudge et al. 1995). We found that moisture management system significantly affected %R but not RRC of *Inga* cuttings regardless of stem diameter (Table 2). The temperature in most (unmist) polyethylene systems is often several degrees higher than in a misted environment (Hartmann et al. 1990; Leakey et al. 1990; Mudge et al. 1995), and the day temperature of enclosed non-mist polyethylene propagators has been reported to vary by about 10 °C with a peak of 34 °C at mid day (Leakey et al. 1990). The relatively small air temperature differences (2 °C) in our mist and polyethylene treatments were no doubt minimized by placing the polyethylene-enclosed flats under mist and shade.

Wide spread fungal growth on the dead cuttings in the poly tents suggests that cuttings may have benefited by fungicide treatment at the time of sticking, but it is possible that observed fungal growth was a secondary infection on already dead or dying tissue. The mist propagated cuttings, in contrast, had relatively few rotted cuttings. Reduced air circulation in our poly tents may also have increased microbial populations in the rooting zone of cuttings. Leakey et al. (1990) reported higher basal rotting of cuttings in misted propagators enclosed in polyethylene than those without polyethylene.

The effect of stem diameter on %R and RRC was dramatic, particularly when cuttings were not treated with auxin (Figure 1A and B). Smaller diameter (≤ 8 mm) softwood (Table 3) and semihardwood cuttings (Table 2) rooted readily without auxin application.

Based on extrapolation from results of experiment 1, a single seedling could produce 15 stock plants after 6 months, using mist propagation of leafy cuttings (2 to 20 mm in diameter, treated with 0.3 or 0.8% IBA). After an additional six months a total of at least 100 cuttings could be produced from a single seedling-derived stock plant.

Conclusions

The results of this research are applicable to both large and small-scale production systems involving *Inga*. The cutting techniques seem particularly appropriate for systems which require large numbers of plants and have access to mist and/or polyethylene, and auxin. Although nonmist polyethylene methods (Leakey et al. 1990; Mudge et al. 1995) may be useful in nurseries and on some small farms, *Inga* is clearly more difficult to root than other popular leguminous tree species commonly used in tropical agroforestry, like *Gliricidia sepium* (Jacquin) Kunth ex Walp. and *Erythrina* spp. L., which are typically rooted from leafless hardwood cuttings (Mudge et al. 1992). Because air layer-

ing is easily accomplished without special facilities (mist or polyethylene enclosures) it seems more applicable than cuttage for use on small farms. Air layering of other species of fruit trees is a popular technique among farmers in tropical locations such as Thailand because it is simple, requires few materials, and results in relatively large (up to approx. 1 m tall) plants that are easily transported, and established in the field (Brennan, pers. observation, 1988 to 1992). Interestingly, small-scale Thai farmers often generate more income from selling air layers of fruit trees like longan (*Dimocarpus longan* Lour.) and lychee (*Litchi chinensis* Sonn.), than they do selling fruit. The ability to clonally propagate tropical fruit and multipurpose trees like *Inga*, can directly benefit small-scale farmers who often do not have access to, or the time to collect seed. This may be particularly true for species such as *Inga* with recalcitrant, viviparous seeds, whose maturation often coincides with crop planting.

Another reason to consider vegetative propagation of *Inga* is to exploit the self-incompatibility (SI) mechanism thought to be associated with its sexual reproduction. The few studies (Koptur 1984; Leon 1966) thus far indicate a high degree of SI within the genus, although one of us (Brennan) has observed good fruiting of isolated *I. feuillei* trees. If most *Inga* are self incompatible, as are many woody mimosoids like *Calliandra* Benth. and *Albizia* Durazz. (Koptur 1984), vegetative propagation could be a useful tool in selection and genetic improvement programs. For example, vegetative propagation of numerous unrelated individuals of a SI *Inga* species, and the subsequent interplanting of these in open pollinated seed orchards, could enhance efforts to produce and conserve genetic diversity within a species. The SI system could also be used to control pollination in inter- and intraspecific seed orchards. In addition, selection for improved *Inga* fruit characteristics (Leon 1966) could be improved through phenotypic selection followed by clonal propagation. Lastly, vegetatively propagating *Inga* cultivars from mature trees might induce earlier flowering and fruit set than seed propagated trees, as is the case with most fruit and nut cultivars (Hartmann et al. 1990)

Our observation that *I. feuillei* is moderately easy-to-root, hopefully will encourage propagation studies with other *Inga* species, especially in the tropical area where they are grown.

Acknowledgments

The authors would like to thank Dr. Gary Churchill, Department of Plant Breeding and Biometry, Cornell University, for advice on statistical analysis, and Mr. John Mood for *Inga* seeds.

References

- Anonymous. 1989. More varieties of tropical fruit are expected to be sold in U.S. Rural Enterprise 3: 26.
- Blazich, F. A. 1988. Chemicals and formulations used to promote adventitious rooting, pp. 132–149. In: Davis, T. D., Haissig, B. E. and Sankhla, N. (Eds) Adventitious Root Formation in Cuttings. Dioscordes Press, Portland, OR.
- Davis, T. D. 1988. Photosynthesis during adventitious rooting, pp. 78–87. In: Davis, T. D., Haissig, B. E. and Sankhla, N. (Eds) Adventitious Root Formation in Cuttings. Dioscordes Press, Portland, OR.
- Denslow, J. S., Newell, E. and Ellison, A. M. 1991. The effect of understory palms and cyclanths on the growth and survival of *Inga* seedlings. Biotropica 23: 225–234.
- Hartmann, H. T., Kester, D. E. and Davies, F. T., Jr. 1990. Plant Propagation: Principles and Practices, 5th ed. Prentice Hall, Englewood Cliffs, NJ.
- Koptur, S. 1984. Outcrossing and pollinator limitation of fruit set: Breeding systems of neotropical *Inga* trees (Fabaceae: Mimosoideae). Evolution 38: 1130–1143.
- Leakey, R. R. B., Mensen, J. F., Tchoundjeu, Z., Longman, K. A., McP. Dick, J., Newton, A., Matin, A., Grace, J., Munro, R. C. and Muthoka, P. N. 1990. Low-technology techniques for the vegetative propagation of tropical trees. Commonwealth Forestry Review 69: 246–257.
- Leon, J. 1966. Central American and West Indian species of *Inga* (Leguminosae). Annals of the Missouri Botanical Garden 53: 265–359.
- Lewis, G. P. 1988. A new species of *Inga* (Leguminosae-Mimosoideae) from Ecuador. Kew Bulletin 43: 707–709.
- Little, T. M. and Hills, F. J. 1978. Agricultural Experimentation: Design and Analysis. Wiley and Sons, New York.
- Loach, K. 1988. Water relations and adventitious rooting, pp. 102–116. In: Davis T. D., Haissig, B. E. and Sankhla, N. (Eds) Adventitious Root Formation in Cuttings. Dioscordes Press, Portland, OR.
- Minitab Reference Manual. Release 10 for Windows. 1994. Minitab Incorporated, State College, PA.
- Mudge, K. W., Mwaka, A., Isutsa, D., Musoke, R., Foster, D., Ngoda, B. 1992. Plant Propagation, a Teaching Resource Packet with Emphasis on Agroforestry and Fruit Trees in East Africa. Published by Department of Floriculture and Ornamental Horticulture, Cornell University, Ithaca, NY.
- Mudge, K. W., Mwaja, V. N., Itulya, F. M. and Ocheing, J. 1995. Comparison of four moisture management systems for cutting propagation of bougainvillea, hibiscus and kei apple. Journal of the American Society of Horticultural Science 120: 366–373.
- National Academy of Sciences, ad hoc panel of the Advisory Committee on Technology Innovation, Board on Science and Technology for International Development, Commission on International Relations. 1980. Firewood Crops: Shrub and Tree Species for Energy Production. National Academy of Sciences. Washington D.C.
- Nichols, D. 1990. *Inga edulis* with coffee in Perez Zeledon, Costa Rica. Nitrogen Fixing Tree Research Reports 8: 145.
- Palm, C. A. and Sanchez, P. A. 1990. Decomposition and nutrient release patterns of the leaves of three tropical legumes. Biotropica 22: 330–338.
- Pursglove, J. W. 1974. Tropical Crops: Dicotyledons. Longman Singapore Publishers, Singapore.
- Roskoski, J. P. 1981. Nodulation and N₂ fixation by *Inga jinicuil*, a woody legume in coffee plantations. I. Measurements of nodule biomass and field C₂H₂ reduction rates. Plant and Soil 59: 201–206.
- Roskoski, J. P. 1982. Nitrogen fixation in a Mexican coffee plantation. Plant and Soil 67: 283–291.

- Van Kessel, C. and Roskoski, J. P. 1981. Nodulation and N₂ fixation by *Inga jinicuil*, a woody legume in coffee plantations. II. Effect of soil nutrients on nodulation and N₂ fixation. *Plant and Soil* 59: 207–215.
- Van Kessel, C. and Roskoski, J. P. 1983. Nodulation and N₂ fixation by *Inga jinicuil*, a woody legume in coffee plantations. III. Effect of fertilizers and soil shading on nodulation and nitrogen fixation (acetylene reduction) of *I. jinicuil* seedlings. *Plant and Soil* 72: 95–105.