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Open-pollinated, interspecific F1 hybrid seed orchards of *Leucaena*

I. *Rootstock effects*

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Abstract. Seed orchards of Leucaena were established at two experimental sites in Hawaii to produce either triploid or tetraploid interspecific hybrid seed by interplanting clonal self incompatible diploid (*L. diversifolia*, *L. esculenta*) or tetraploid (*L. pallida*) species with self-compatible *L. leucocephala* lines and the F1 hybrid of *L. diversifolia* ($2n = 4x$) × *L. leucocephala*. The seed parents were grafted onto three seedling rootstocks (*L. diversifolia* ($2x = 4n$), shrubby *L. leucocephala*, and giant *L. leucocephala*) to compare rootstock effects on scion growth. This paper reports on the effects of rootstock on scion growth during the first year based on total truck cross sectional area, height growth, and crown habit. In the triploid seed orchard, rootstock significantly affected the growth of four of the six clones, while in the tetraploid orchard the growth of two of the five clones was significantly affected by rootstock. Crown habit was independent of rootstock. Our study suggests that rootstock effects depend on the specific scion and rootstock combination, and site.

Introduction

The genus Leucaena has received considerable attention in research and practice over the past two decades relative to other tropical woody legumes. *Leucaena leucocephala* is one of the best known and widely used multipurpose tree species in tropical agroforestry systems (National Research Council, 1984). Important products from the leucaenas include fuelwood, fodder, green manure, and in some areas timber, and human food. In addition, leucaenas are extensively used for soil conservation and improvement, shade, and reforestation.

Leucanena research has concentrated largely on the value of the various products and the integration of leucaenas (especially *L. leucocephala*) into agroforestry systems. Additional research has focused on interspecific hybridization, and evaluating species and hybrids for biomass production, fodder and wood quality, and leucaena psyllid (*Heteropsylla cubana*) resistance (Brewbaker and Sorensson, 1990).

Although leucaenas are easily propagated from seed, there is a need for reliable clonal propagation techniques for the large-scale propagation of superior hybrids, and to take advantage of self-incompatibility in hybrid leucaena seed orchards (Brewbaker, 1988). Considerable effort has revolved around developing clonal propagation techniques including tissue culture, cuttage, and graftage (Brennan and Mudge, 1998). Brewbaker (1988) reported that in Indonesia, seedless interspecific leucaena hybrids are grafted for use as shade over coffee and tea. Seedless and sterile hybrids are desirable in agroforestry systems because they are not prone to weediness, as is *L. leucocophala* (Hughes and Styles, 1987; Brennan, 1990), and may allocate more photosynthate to vegetative biomass (Brewbaker, 1988).

All diploid species of *Leucaena* and the tetraploid *L. pallida* are gametophytically self-incompatible (SI) within any given clone, while other tetraploids are self-compatible (SC) (Sorensson and Brewbaker, 1994). Self-incompatibility is a mechanism that usually ensures obligate outcrossing (Richards, 1986). Vegetative propagation is necessary to exploit the intraclonal SI mechanism by allowing interspecific F1 hybrid seed production on the SI parent, under open-pollination conditions (Brewbaker and Sorensson, 1990). Wheeler (1991) suggested grafting to produce seed trees of SI species for use in open-pollinated interspecific hybrid seed orchards. In such orchards a single clone of a SI species such as *L. pallida* or diploid *L. diversifolia* would presumably set seed only from cross-pollination with interplanted cross compatible species like *L. leucocephala*. Several interspecific F1 hybrids produced by hand pollination have exhibited excellent field performance (Sorensson et al., 1994; Sorensson and Brewbaker, 1994) and psyllid resistance (Austin et al., 1997). However, due to the difficulty of clonal propagation of the genus, there are no known seed orchards that currently produce F1 interspecific leucaena hybrids through open-pollination.

Grafting is a low-technology clonal method of propagation widely utilized in the tropics to propagate fruit and other trees. Previous studies on grafting leucaenas (Brennan and Mudge, 1998; and citations therein) did not investigate rootstock effects on scion performance. With apple and other horticultural fruit trees, rootstock genotype is known to influence tree size, growth habit, fruiting, precocity, disease resistance and cold hardiness (Hartmann et al., 1990). Similarly in seed orchards of temperate forest species, grafting is extensively used to maximize cross-pollination between ‘elite’ seed parents (Jagawickrama et al., 1991).

The main objectives of this two part study were to (1) evaluate three rootstocks in interspecific F1 hybrid seed orchards (present paper), and (2) to analyze the parentage of seed produced in such orchards (accompanying paper, Brennan et al., 1998). We were specifically interested in identifying dwarfing rootstocks that might increase seed production/unit land area and simplify seed orchard management. We hypothesized that the scion growth on a particular rootstock could be predicted by the growth of an ungrafted tree of the same rootstock genotype. This paper reports on the effects of three rootstocks on the growth of eight scion clones during the first year based on total trunk cross sectional area, height growth and crown habit.

Materials and methods

Rootstock seed collection

Rootstocks included the giant type *L. leucocephala* ssp. *glabrata* K636, the smaller shrubby type *L. leucocephala* spp. *leucocephala* K997 and tetraploid *L. diversifolia* ssp. *diversifolia* K156 (Table 1). These lines were chosen because they are among the most widely available leucaenas and differ in size, and adaptability to the climates at the experimental sites in the study.

Open-pollinated seed of each rootstock line was collected in Hawaii during July and August 1993. Seed of *L. leucocephala* K636 and *L. diversifolia* K156 were from specimens at the University of Hawaii, Waimanalo Research Station, Oahu. Seed of *L. leucocephala* K997 came from a wild tree along the roadside in Kailua, Oahu. Seed for each rootstock line was from a single tree. Because *L. leucocephala* and tetraploid *L. diversifolia* are highly self-pollinating (Sorensson and Brewbaker, 1994) and thus presumably highly homozygous, we assumed genetic uniformity within each rootstock line.

Scion collection

Scion ramets of *L. diversifolia* K907, *L. leucocephala* (K420, K481, K636), the F1 hybrid KX3 (*L. diversifolia* K156 × *L. leucocephala* K636), and *L. pallida* K804 were obtained from adult phase (reproductive) branches of selected ortets at the Waimanalo Research Station. Scions were grafted onto seedling rootstocks (K156, K636) from June to August of 1993 using the grafting methods described in Brennan and Mudge (1998). Grafted plants were transported to Cornell University, Ithaca, New York, in August 1993, to serve as scion donor plants for subsequent grafting from December 1993 to March 1994. Attempts to graft scions from an adult phase ortet of *L. esculenta* K138 failed, therefore a seedling of *L. esculenta* K948 obtained from the University of Hawaii germplasm collection, was used to produce scions for this species. Scions of *L. pallida* K953 were similarly obtained from a two-year-old juvenile phase seedling grown in the greenhouse at Cornell University from University of Hawaii germplasm. Although the SC trees to be used as pollen donors (K420, K481, K636 and KX3) in the seed orchards could have been grown from seed, we grafted them to investigate rootstock effects and hasten flowering since these scions were in the adult phase of growth.

Selection of the original ortets at the Waimanalo Research Station was based on their flowering, growth form, and potential as parents of interspecific hybrids. The latter was based on earlier evaluation of hybrid seedling progeny (from hand pollinations of the same trees) for seedling vigor, psyllid

Table 1. Scion and rootstock taxa used in Leucaena hybrid seed orchards near Kona, Hawaii, USA.

Scion taxon	UH K #[a]	Ploidy[b]	Orchard	# of trees per rootstock			Growth phase[c]
				K156	K997	K636	
L. diversifolia × *L. leucocephala*	K156 × K636 (KX3)	$2n = 4x = 104$	1	15	7	15	A
			2	11	9	10	A
L. leucocephala	K420	$2n = 4x = 104$	1	10	12	11	A
			2	11	7	11	A
L. leucocephala	K481	$2n = 4x = 104$	2	9	7	9	A
L. leucocephala	K636	$2n = 4x = 104$	1	11	13	15	A
			2	11	9	13	A
L. diversifolia	K907	$2n = 2x = 52$	1	20	21	25	A
			2	na	na	na	A
L. esculenta	K948	$2n = 2x = 52$	1	7	5	6	A
			2	na	na	na	A
L. pallida	K804	$2n = 4x = 104$	1	na	na	na	A
			2	4	4	0	A
L. pallida	K953	$2n = 4x = 104$	1	na	na	na	J
			2	3	12	12	J

Rootstock taxon	UH K #	Ploidy	Orchard	Ungrafted[d]	Growth phase
L. diversifolia	K156	$2n = 4x = 104$	1	12	J
			2	10	J
L. leucocephala	K997[e]	$2n = 4x = 104$	1	10	J
			2	10	J
L. leucocephala	K636	$2n = 4x = 104$	1	11	J
			2	10	J

[a] University of Hawaii K accession #; [b] Ploidy level and somatic chromosome #; [c] J = juvenile, A = adult; [d] The # of ungrafted rootstocks controls included in the analysis in orchards 1 and 2; na, not applicable.

resistance, and cold tolerance (Sorensson et al., 1994; Sorensson and Brewbaker, 1994; W. Sun and CT Sorensson, pers. comm.).

Greenhouse grafting

In September 1993, the rootstock seeds were scarified with boiling water and soaked for 12 hours. They were germinated on moist paper towels in the dark and inoculated with the *Rhizobium* peat mixture TAL 1145 (Agroforester Tropical Seeds, Holualoa, Hawaii). Seedlings were transferred into 3.8 cm diameter × 14 cm deep Super Stubby Cells (Stuewe and Sons, Inc., Corvallis, Oregon) filled with Metro-Mix 360 rooting medium (Grace Sierra Horticultural Products Company, Milpitas, California). The rootstocks were grown in greenhouses under 400 watt, high pressure sodium lamps with a 13 hour photo period, and day and night temperatures of 23 to 27 °C and 21 to 23 °C, respectively. Scions from the donor plants were grafted onto the seedling rootstocks in the greenhouse. With exception of *L. pallida* K953, all clones were grafted onto four to seven month old seedling rootstocks using the graft guide tool and single bud splice grafting technique (Brennan and Mudge, 1998). The K953 scions were grafted onto one-year-old seedling rootstocks using modified crown veneer grafting (Brennan and Mudge, 1998) due to a shortage of younger rootstocks.

In May 1994, the grafted trees and ungrafted (control) rootstock trees were transported by air to Kona, Hawaii. The grafted trees were reacclimated in an open nursery for one to two weeks prior to transplanting.

Experiment 1: (Orchard 1)

This orchard was designed to produce open-pollinated F1 seed of up to six triploid interspecific hybrids. These included the complete factorial combinations of two SI diploids (*L. diversifolia* K907, and *L. esculenta* K948) and three SC tetraploids including *L. leucocephala* (K420, K636), and the F1 hybrid KX3 (*L. diversifolia* K156 × *L. leucocephala* K636). Crossing of the two SI diploids was considered unlikely because of their very low crossability index (Sorensson and Brewbaker, 1994).

Field description. The orchard is located 400 meters above sea level (msl). The mean annual rainfall is 1650 mm, and mean annual soil temperature ranges between 18 and 20 °C (USDA, 1973). The soil is classified as a medial-skeletal isohyperthermic lithic haplustand in the Waiaha series. This soil series is an extremely stony, well-drained, silty clay loam derived from volcanic ash and a 10,000-year-old lava flow. The pH of the soil is 5.6. A few months prior to planting, a bulldozer was used to clear the existing vegetation (weedy strawberry guava (*Psidium cattleianum*), Christmas berry (*Schinus terbithifolius*) and silk oak (*Grevillea robusta*)), crush the lava rock and smooth the site as is the typical site preparation for macadamia, coffee and citrus orchards in the area.

Experimental design. The experiment was a partially balanced incomplete block (PBIB) design with $r = 6$ complete blocks, $v = 40$ trees per complete block that was further divided into $k = 4$ trees per incomplete block; r, v, and k are standard design parameters for the PBIB design (Lentner and Bishop, 1993). The PBIB design was used to account for soil variation within the large complete blocks. There were 18 treatments including the five scion clones (noted above) grafted onto all three seedling rootstocks (15 scion/rootstock combinations), and the three ungrafted rootstock controls. The experiment contained 240 trees (experimental units), including 204 that were grafted (hybrid seed parents) and 12 each of the ungrafted rootstocks. The number of individuals of each scion/rootstock combination is noted in Table 1. With the randomization procedure, we aimed to maximize the efficiency of the PBIB design by taking the largest balanced subset of experimental plants (six trees for all 18 treatments) and then assigning remaining trees to 22 additional 'pseudotreatments'. The GENDEX computer software (CSIRO-IAPP Biometrics Unit, Clayton, Victoria, Australia) was used to optimize placement of the 18 treatments and 22 'pseudotreatments' throughout the field, and to facilitate cross pollination between grafted SI maternal clones and the SC pollen donors.

The spacing between trees was 2 × 2 m, with complete blocks 1 and 2 end to end, producing four rows of 20 trees each. Complete blocks 3 and 4, and blocks 5 and 6 were arranged similarly, with a 6 m alley between each of the three pairs of blocks.

Experiment 2: (Orchard 2)

This orchard was designed to produce open-pollinated F1 hybrid seed of up to eight tetraploid interspecific hybrids. These included the complete factorial combinations of two SI lines of *L. pallida* (K804, K953) and four SC tetraploids including three of *L. leucocephala* (K420, K481, K636), and the F1 hybrid KX3 (*L. diversifolia* K156 × *L. leucocephala* K636). Although cross pollination of the two *L. pallida* clones was a possibility, it did not occur since *L. pallida* K953 did not flower during the study. If necessary we had planned to manage the flowering of the two *L. pallida* clones, by irrigation and pruning, to prevent them from cross pollinating each other.

Field description. This seed orchard is located at 200 msl, 200 m below orchard 1. The mean annual rainfall is 1000 mm, and the mean annual soil temperature is 22 to 23 °C (USDA, 1973). The soil is a hydrous-skeletal isothermic lithic hydrand in the Napoopoo series. This soil series is an extremely stony well-drained silty loam derived from volcanic ash and a 10,000-year-old lava flow. The pH of the soil is 6.7. The natural vegetation is dominated by shrubby *L. leucocephala*. The seed orchard at this site was interplanted in the alleys of an existing five-year-old citrus orchard. The 1 to 2 m tall citrus were in contour rows 7.5 m apart, with an in-row spacing of 5 m. Various nitrogen-fixing trees (*Calliandra calothyrsus*, *Gliricidia sepium*, *L. diversifolia* K749 ($2n = 2x$), *and Sesbania sesban*) were grown to 2 m tall in-row, between the citrus, for green manure.

Experimental design. This experiment was PBIB design ($v = 40$, $k = 4$, $r = 5$), similar to that of Site 1. There were 21 treatments including the five scion clones grafted onto all three rootstocks (18 scion/rootstock combinations), and the grafted rootstock controls. This experiment contained 200 trees, including 170 grafted trees (hybrid seed parents) and 10 trees each of the ungrafted rootstocks. For randomization procedures with the GENDEX software, there were 21 treatments and 19 'pseudotreatments'. The 200 trees were planted at a 2 × 2 m spacing.

Tree planting procedures. The seed orchard trees at both sites were planted in the field in June 1994, when they were approximately 50 cm tall. In orchard 1, the soil surface at each planting hole was amended with one cup (222 ml) of Basalt and Andesite Rock Flour (10% Ca, 3% Fe, 1% Mg; Remineralization Products Inc., Eugine, OR). The soil in orchard 2 was similarly amended with Colloidal Clay (2% available phosphoric acid; Longala, Inc., Florida). These amendments were applied to mark each tree site and improve soil fertility. Due to the extremely rocky nature of the soil, a large steel crowbar was used to make a hole just large enough for the root ball of each tree. The grafted trees were planted with the graft union approximately 1 cm above the soil level. One month after planting (July 1994), all flowers were pinched off of the trees to encourage initial vegetative growth.

Each tree was drip-irrigated by an automated polyethylene system. The amount of irrigation at a given site and day was adjusted between 0 to 4 liter/tree depending on rainfall. For weed control, 1 m^2 around each tree was mulched with recycled cardboard held down with rocks. Additional weeding was done with a brush cutter and hand sickle.

Measurements. Tree height was measured immediately after field planting. Flowering status (flowering or not) was recorded at one, two, and seven months after planting. In January 1995 (seven months after planting and approximately one year after grafting) scion stem diameter was measured 10 cm above the soil level. An electronic digital caliper (Mitutoyo, Japan) was used to measure two diameters perpendicular to each other for each main scion stem. Total trunk(s) cross sectional area (TTCSA) was later calculated from the average stem diameters for each tree. At the same time tree height was measured and height growth (HG) was calculated. TTCSA and HG are common measurements in rootstocks studies (Schechter et al., 1991; Guerriero et al., 1988).

In January 1995, each graft union was visually assessed for incompatibility symptoms including breakage and scion overgrowth. At the same time the crown habit of each tree was scored as either arborescent (a single dominant stem) or multistemmed (several main stems). In January, all dry pods were collected from SI trees for analysis of parentage (Brennan et al., 1998).

Statistical analysis

Analysis of variance (ANOVA) on TTCSA and HG data was conducted using the Proc GLM procedure in SAS (SAS Instituted Inc., Cary, NC). Based on residual analysis, the data met the assumptions of ANOVA and thus did not require transformation. Each experiment was analyzed separately due to differences in the experimental design in each orchard. To determine the level of significance of the main effects (rootstock and scion) and interaction (rootstock × scion) only the grafted treatments were included in the analysis. Trees with the *L. pallida* K953 scion were excluded from the ANOVA since they were grafted onto older rootstocks than the other scion clones. Grafted trees that died or failed to grow due to a poor graft union were also excluded. Preplanned comparisons (contrasts) of rootstock effects on a given scion, and between ungrafted rootstocks were made using both single degree of freedom contrasts to control comparisonwise error, and Tukey's HSD to control experimentwise error. All preplanned contrasts were conducted using the data for both grafted and ungrafted control trees. In the ANOVA and contrasts, significance is ascertained at the $P \leq 0.05$ level, however the actual P-value is included where $0.05 \leq P \leq 0.10$ to indicate nearly significant results. Categorical data of crown habit were analyzed in SAS using Fisher's exact test.

Results

The rate of grafting success was between 67 and 74% across all scions and rootstocks (Brennan and Mudge, 1998).

Rootstock effects on growth

The ANOVA for TTCSA and HG after seven months of field growth are shown in Table 2. In orchard 1 for TTCSA, rootstock ($P < 0.01$), scion ($P < 0.001$) and their interaction ($P < 0.01$) were all significant, while in orchard 2 only scion ($P < 0.01$) was significant. In general the effects of rootstock on TTCSA and HG were similar. Significant main effects are not a prerequisite for conducting single degree of freedom preplanned contrasts since the overall F test is an average across all contrasts and may not accurately detect significant differences (Chew, 1976). The preplanned contrasts shown in Table 3 indicate significant effects of rootstocks on TTCSA and HG for several scions. The contrasts and plotted means of TTCSA (Figure 1) indicate that the effect of rootstock depends on a given scion/rootstock combination, and although not statistically tested also appears to be site dependent.

In both orchards, the TTCSA and HG of the shrubby *L. leucocephala* K997 ungrafted rootstock control was significantly less than that of either *L. diversifolia* K156 or giant *L. leucocephala* K636 rootstock controls. However, there

Table 2. ANOVA for total trunk(s) cross sectional area (TTCSA) and height growth (HG) in orchards 1 and 2, near Kona, Hawaii, USA.

Source	df	Mean squares and level of significance[a]			
		Experiment 1 (Orchard 1)		Experiment 2 (Orchard 2)	
		TTCSA	HG	TTCSA	HG
IC Block	59 (49)	91027 ns (0.07)	6190 ***	41240 *	5823 *
Treatment	14 (14)	256653 ***	6575 ***	52453 ns (0.10)	5416 ns
Rootstock (R)	2 (2)	391981 **	7282 *	45361 ns	7225 ns (0.10)
Scion (S)	4 (4)	336513 ***	12478 ***	87445 *	3455 ns
R × S	8 (8)	182891 **	3466 ns (0.7)	36729 ns	5944 ns (0.09)
Error	119 (62)	65681	1839	26555	3256
Total	192 (125)				
C.V.		62.7	34.7	51.2	34.1
R^2		0.53	0.69	0.63	0.64

[a] ns is not significant at the $P > 0.05$ level; *, ** and *** are significant at the 0.05, 0.01 and 0.001 levels, respectively; the actual P value is shown where $0.05 > P < 0.10$.

were significant differences between the TTCSA and HG of ungrafted *L. diversifolia* K156 and *L. leucocephala* K636 only at the lower elevation (orchard 2) where K636 had significantly more growth.

In orchard 1, rootstock significantly affected the growth of three (KX3, K907, K948) of the six scions tested, while in orchard 2 only two (KX3, K636) of the five scions were affected by rootstock (Table 3). At the higher elevation site (orchard 1), the shrubby *L. leucocephala* K997 rootstock significantly reduced the growth of both hybrid KX3 and *L. esculenta* K948 relative to the other rootstocks; a similar rootstock effect was seen in orchard 2 for the scions KX3 and *L. leucocephala* K636. The dwarfing and invigorating effect of the giant *L. leucocephala* K636 rootstock on diploid *L. diversifolia* K907 and *L. leucocephala* K636 scions respectively, is evidence of the significant rootstock × scion interaction in orchard 1. Interestingly, in orchard 2, rootstock had significantly different effects on the two giant *L. leucocephala* scions (K420 and K636), where K420 growth was independent of rootstock, in contrast to K636 which had maximum growth grafted onto the K636 rootstock.

Table 3. Significance levels for preplanned comparisons of total trunk cross sectional area (TTSCA) and height (HG) of the ungrafted rootstocks and various scion/rootstock combinations, near Kona, Hawaii, USA.

Contrasts[a]	Experiment 1 (Orchard 1)		Experiment 2 (Orchard 2)	
	TTCSA	HG	TTCSA	HG
Ungrafted rootstocks				
K156 vs K997	** (*)	*** (*)	* (*)	** (*)
K156 vs K636	ns (ns)	ns (ns)	** (*)	** (*)
K997 vs K636	** (*)	*** (*)	*** (*)	*** (*)
Grafted trees				
KX3/K156 vs KX3/K997	** (*)	** (*)	* (*)	0.06 (*)
KX3/K156 vs KX3/K636	ns (ns)	ns (ns)	ns (ns)	ns (ns)
KX3/K997 vs KX3/K636	* (*)	* (*)	* (*)	** (*)
K420/K156 vs K420/K997	ns (ns)	ns (ns)	ns (ns)	ns (ns)
K420/K156 vs K420/K636	ns (ns)	ns (ns)	ns (ns)	ns (ns)
K420/K997 vs K420/K636	ns (ns)	ns (ns)	ns (ns)	ns (ns)
K481/K156 vs K481/K997	–	–	ns (ns)	ns (ns)
K481/K156 vs K481/K636	–	–	ns (ns)	ns (ns)
K481/K997 vs K481/K636	–	–	ns (ns)	ns (ns)
K636/K156 vs K636/K997	ns (ns)	ns (ns)	ns (ns)	ns (ns)
K636/K156 vs K636/K636	ns (ns)	*** (*)	** (*)	*** (*)
K636/K997 vs K636/K636	ns (ns)	** (*)	** (*)	** (*)
K636 vs K636/K156	ns (ns)	ns (ns)	** (*)	* (*)
K636 vs K636/K997	ns (ns)	** (*)	** (*)	** (*)
K636 vs K636/K636	ns (ns)	ns (ns)	ns (ns)	ns (ns)
Grafted SI trees				
K907/K156 vs K907/K997	ns (ns)	ns (ns)	–	–
K907/K156 vs K907/K636	*** (*)	** (*)	–	–
K907/K997 vs K907/K636	* (*)	* (*)	–	–
K948/K156 vs K948/K997	* (*)	ns (ns)	–	–
K948/K156 vs K948/K636	* (*)	0.1 (*)	–	–
K948/K997 vs K948/K636	*** (*)	ns (ns)	–	–
K804/K156 vs K804/K997	–	–	ns (ns)	ns (ns)
K953/K997 vs K953/K636	–	–	0.06 (ns)	ns (ns)

[a] The significance levels of comparisons of interest are shown for both single df contrasts (not in parenthesis) and Tukey's HSD procedure (in parenthesis). – is not applicable; ns is not significant at the $P > 0.1$ level; *, ** and *** are significant at the 0.05, 0.01 and 0.001 levels, respectively; the actual P value is shown where $0.05 > P \leq 0.1$. Single df contrasts are based on a comprisonwise error rate, while Tukey's HSD is based on an experimentwise error rate.

Crown habit

There were obvious differences in crown habit among the various scions and the ungrafted rootstock controls. In both orchards the majority of the *L. leucocephala* K636 and *L. diversifolia* K156 rootstock control trees were arborescent (not shrubby) compared with *L. leucocephala* K997 which were predominantly shrubby ($P \leq 0.05$). However, despite the significant differ-

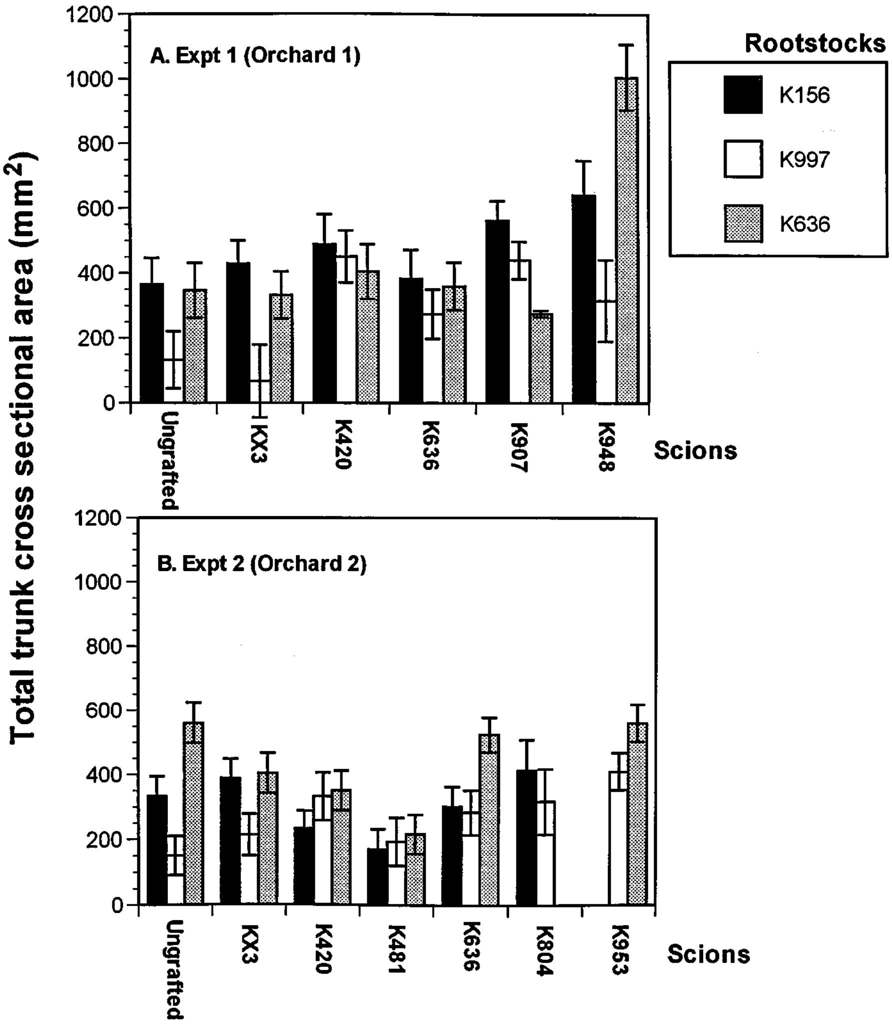

Figure 1. Total trunk cross sectional area of ungrafted rootstocks and various scion/rootstock combinations in Experiment 1 (Orchard 1) (A), and Experiment 2 (Orchard 2) (B) after six months of growth at field sites near Kona, Hawaii, USA.

ences between the crown habits of the grafted rootstocks, rootstock did not significantly affect scion crown habit.

Flowering and pod production

The clones collected from adult phase trees in Hawaii all retained their ontogenetic age following grafting as evidenced by flowering of scion donor plant in the greenhouse. However, less than 50% of the grafted adult phase trees flowered and set pods during the first year in the field. We were thus unable to determine the effect of rootstock on flowering or seed production. In orchard 1 and 2, respectively, 18 and 15% of grafted *L. leucocephala* K636 trees flowered during the first seven months of field growth compared to ungrafted *L. leucocephala* K636 rootstock controls which did not flower. In orchard 1, 18% (12/66) of the *L. diversifolia* K907 trees set pods but the majority of these aborted before maturity and thus we failed to obtain a sample of seed. In orchard 2, 38% (3/8) *L. pallida* K804 trees set pods which developed to maturity and were collected for analysis of parentage as described in the accompanying paper (Brennan et al., 1998).

Graft compatibility

Inspection of the graft unions at the time of planting and seven months later indicated early signs of graft incompatibility with one of the scion varieties, *L. esculenta* K948. Although there were relatively few (six to eight) trees of *L. esculenta* K948 on each rootstock, scion overgrowth was observed on all three rootstocks. Scion overgrowth of this scion was most common on *L. diversifolia* K156 (71%) followed by *L. leucocephala* K997 (60%) and *L. leucocephala* K636 (33%) rootstocks. Furthermore the foliage of most grafted K948 trees was chlorotic and sparse. Trees with scion overgrowth had stem diameters, immediately above the graft union, approximately twice the diameter of that below the union. Scion overgrowth was noticeable within six weeks of grafting.

Discussion

We hypothesized that scion growth (TTCSA and HG) could be predicted from ungrafted rootstock performance. Based on this hypothesis and considering the growth of the ungrafted rootstock controls, in orchard 1 we expected scions grafted onto the shrubby *L. leucocephala* K997 to be dwarfed relative to those grafted onto the giant *L. leucocephala* K636 and *L. diversifolia* K156. Similarly, in orchard 2 we expected maximum growth of scions on the K636 rootstock, intermediate growth on K156 and minimal growth on K997. However, based on the growth of the scions tested on all three rootstocks, we accept the hypothesis for only one of the five scions (KX3) in orchard 1, and none of the scions at orchard 2. The predictive ability of ungrafted rootstock growth is however more reliable when considering only the two *L. leucocephala* rootstocks (K636, K997). In orchard 1, two (KX3 and K948) of the five scions were significantly dwarfed by the K997 rootstock, while in the other orchard, three (KX3, K636 and K953) of the five scions were significantly dwarfed by K997 relative to the K636 rootstock. Breeders selecting for size controlling rootstocks and interstocks have found little or no correlation between the growth of ungrafted rootstocks and the growth of scions grafted onto those rootstocks (Schmidtling, 1983; Schmidtling and Gruppe, 1988). In fact, some studies found small bushy trees to be invigorating rootstocks (Jones, 1986) and vigorous trees to be dwarfing rootstocks (Renaud et al., 1988).

Interactions between rootstock, scion, and environment present major challenges in evaluating and selecting rootstocks (Hartman et al., 1990). Scion × rootstock interactions were significant ($P < 0.01$) for TTCSA only in orchard 1. With *L. leucocephala* K636 as a scion, environment (site) also appears to influence rootstock effect. At the higher elevation (orchard 1), *L. leucocephala* K636 scions grafted onto *L. diversifolia* K156 and *L. leucocephala* K636 rootstocks, had the same TTCSA, while at the lower elevation (orchard 2) K636 scions had significantly greater TTCSA on K636 than on the K156 rootstock. This apparent (but not statistically tested) site × rootstock interaction may be explained by the fact that *L. diversifolia* K156 is better adapted to the cooler climate at orchard 1, relative to *L. leucocephala* K636, which is better adapted to warmer climates like orchard 2 (Figure 1; Brewbaker and Sorensson, 1987).

The primary reason to clone SI species in seed orchards is to facilitate open-pollinated hybrid seed production. Our data show that rootstock can significantly affect the vegetative growth of some scions such as *L. diversifolia* K907 and *L. esculenta* K948. Due to low flowering and seed production in this study, it is not clear if a dwarfing or invigorating rootstock would be most appropriate in hybrid leucaena seed orchards. This most likely depends on the design and management of an orchard and economic factors such as labor costs for grafting, pruning and seed collection. Due to the graft-incompatibility symptoms of *L. esculenta* K948, it appears that all of the rootstocks tested are inappropriate and that it may be preferable to graft clones of this species onto *L. esculenta* seedling rootstocks, or clone it from cuttings.

The SC pollen donors (with the exception of the F1 hybrid KX3) are highly homozygous and can be grown with acceptable uniformity from seed. Leucaenas planted from seed normally flower within one to two years (Wheeler, 1991). Our data showed that adult phase scions retained their ontogenetic age since they flowered a few months after grafting; thus grafting the pollen donors may provide some advantage in seed orchards. Another reason to graft SC clones is to control their growth and size with a dwarfing rootstock. Our data (Figure 1, Table 3) show that at the lower elevation (orchard 2), shrubby *L. leucocephala* K997 and *L. diversifolia* K156 rootstocks significantly reduced the TTCSA of the grafted giant *L. leucocephala* K636 by approximately 50% compared to the ungrafted K636 rootstock control or K636 grafted onto itself. Similarly, the shrubby *L. leucocephala* K997 rootstock significantly reduced the growth of the KX3 hybrid relative to the other rootstocks at both sites. In high-density apple orchards, dwarfing rootstocks have been used to increase the fruit production/unit land area (Cummins and Aldwinckle, 1974). Such rootstocks might be especially useful in multipurpose agroforestry systems that aim to produce hybrid seed and other crops.

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