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Genetic resources of *Cedrela odorata* L. and their efficient use in Mesoamerica

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ABSTRACT

The general objective of this work is to assess the genetic resources of Spanish cedar (*Cedrela odorata* L.) and study possibilities for their efficient use. It is a highly valued forest species, chiefly because of its high quality wood. It has been severely extracted in natural forests and is considered endangered in Mesoamerica.

This work examines genetic diversity and population differentiation in genetic resources of Spanish cedar from several countries of Mesoamerica, and surveys the plantation of field experiments and conservation gardens.

In the present study within-population variability in molecular markers and in quantitative traits were not correlated over populations. The amount of interpopulation differentiation was higher for molecular markers ($F_{ST} = 0.67$) than for quantitative traits ($Q_{ST} \approx 0.30$), suggesting that the deviation in the quantitative traits was less than could have been achieved by genetic drift alone. However, pair-wise population comparisons of marker genes and quantitative differentiation exposed a high positive correlation ($r = 0.66$), signifying that the degree of divergence in the molecular markers can be used to predict the degree of population differentiation in quantitative traits.

The progeny-provenance tests and agroforestry experiments indicated that all variables studied showed significant differences between provenances. The best performing provenances showed outstanding ratings for diameter and height growth, insect resistance and single stem regrowth after insect attack.

The coffee mixtures that provided the best environment for the growth of *C. odorata* consisted of mature coffee trees with *C. odorata* trees planted between the coffee rows. The

attack of the shoot borer *Hypsipyla grandella* was also more inhibited in mixtures containing mature coffee bushes than in recently planted or pruned (to 30 cm from the base) bushes. The number of shoots that re-sprouted following attack by the shoot borer was significantly lower in the blocks where *C. odorata* was planted within the coffee rows because of the strong lateral competition between the *C. odorata* trees and the coffee branches as well as the lateral shade they provided. Agroforestry systems using mixed plantings of *C. odorata* and coffee can provide a good economical option for conserving populations of Spanish cedar.

Two of the challenges to overcome in the management of the broad-leaved forests with Spanish Cedar and other valuable trees are increasing the harvest and commercialisation of several species, in order to decrease excessive pressure on the utilisation of the traditional valuable timber species and to establish plantations of such species both in agroforestry and mixed plantations.

Conservation work will require a coordinated effort among all the Mesoamerican countries, where farmers will participate in conserving, planting and managing forests that contain *C. odorata*.

Policies to develop community forestry projects for conservation on-farm (*circa situ*) should be promoted. Such projects could be developed within the framework of the Mesoamerican Biological Corridor. Given its socio-economic importance, our results highlight the need for future studies encompassing the whole natural distribution of the species including the yet unstudied populations in South America.

1. INTRODUCTION

1.1. GENERAL ASPECTS

Cedrela odorata L. (Cedro amargo (S: Spanish, Mesoamerica), Cigar-Box Wood (E: English), Red Cedar (E), Spanish Cedar (E), Acajou Rouge (F: French), Acajou-Bois (F), Cedrat (F), Cedro Rojo (S.), is one of the most important mahogany species of the neotropics, chiefly because of its high quality wood. It has been planted in different countries in pure plantation trials (Burley and Lamb 1971). However, results of plantation projects with this species have not been satisfactory because of shootborer attack. Farmers with small and medium sized properties have sometimes had good results in growing scattered trees associated with several annual and perennial crops (Guevara 1988, Ford 1979).

C. odorata is a semi-deciduous tree up to 40 m tall and 2 m in diameter producing a lightweight timber. Its natural distribution range is confined to the Neotropics, extending from northern Mexico (26 °N) to Argentina (28°S), including the Caribbean (Styles 1981, Navarro 1999, CAB 2000).

Although Spanish cedar is widespread geographically, it is not common throughout moist tropical American forests, and its numbers are continuing to be reduced by exploitation (Cintron 1990). Individual trees are typically scattered in mixed semi-evergreen or semi-deciduous forests dominated by other trees. It is a rare species with less than one individual per ha over most of its range (Patiño 1997).

Plantation forestry is one option for sustainable production of Spanish cedar. The ease of management in the nursery, fast growth, adaptability to different soils and climatic conditions and the possibility of growing it in agroforestry systems, have made *C. odorata* one of the

most popular species to be planted by small farmers. The wood is appreciated in the local markets; its wood is aromatic and resistant to termites and rotting.

However, *C. odorata* is highly susceptible to the attack of the shootborer (*Hypsipyla grandella* Zeller), which is considered to be one of the most severe forest pests in Latin America and the Caribbean. This pest reduces growth, increases the costs of maintenance and weeding, and induces bifurcation with consequent loss in value of the timber (Hilje and Cornelius 2001, Taveras 2002). The problem is more acute in pure plantations, while less damage and better survival have been observed in mixtures, at low densities or in agroforestry systems.

Spanish cedar is of great interest to Mesoamerican governments and the FAO has been establishing a network to facilitate its genetic conservation, together with that of other species of the family (Patiño 1997). Exploitation has continued on a large scale over the past 200 years and the species is now widely threatened at the provenance level (see summaries by Cintron 1990, Patiño 1997, Hilton-Taylor 2000). The species was assessed in 1997 for the Red List Category and Criteria and was classified as "vulnerable by selective logging" (Hilton-Taylor 2000).

The genomic size of *C. odorata* (1C=90 Mb) is smaller than that of *Arabidopsis* (1C=120 Mb) or tobacco (1C=4200Mb) (Wilson *et al*. 2001). Chromosome numbers lie within the range of 2n = 50 to 2n = 56 for different chromosomic races of *C. odorata* (Styles and Koshla 1976).

1.2. THEORETICAL FRAMEWORK

1.2.1. Conservation genetics

> "The understanding of phenotypic evolution at the species level requires information on the evolutionary forces operating on populations and on the relative consequences of such forces for phenotypic divergence within and among populations" (Lynch *et al*. 1999).

Conservation genetics and research into evolutionary biology presuppose a basic understanding of the causes and extent of local adaptation, as well as distribution of genetic variability among and within different populations. Substantial research has been done to characterise variability and population differentiation of many species using neutral molecular markers (e.g. Ward *et al*. 1992, Avise 1994, Smith and Wayne 1996). Much less effort has been done in this respect with genes coding for quantitative traits (Lynch 1996, Frankham 1999, Reed and Frankham 2001, Merilä and Crnokrak 2001). Estimates of genetic variances and heritabilities are restricted to single or a few populations (but see: Cheverud *et al*. 1994, Waldmann and Andersson 1998, Pfrender *et al*. 2000). Studies comparing neutral markers and quantitative genetic data are rare (reviews in: Reed and Frankham 2001, Merilä and Crnokrak 2001, McKay and Latta 2002).

In recent times, interest has grown in evaluating the usefulness of neutral marker genes for drawing inferences on quantitative genetic variability (Cheverud *et al*. 1994, Butlin and Tregenza 1998, Waldmann and Andersson 1998, Pfrender *et al*. 2000, McKay *et al.* 2001) and differentiation (reviews in: Reed and Frankham 2001, Merilä and Crnokrak 2001, McKay and Latta 2002). This interest has been motivated by two closely related objectives. First, in conservation genetics, there is a need to establish whether variability in molecular markers

reflects variability in quantitative traits. Molecular markers are sometimes used as a basis for management recommendations under the assumption that maximizing marker variability will provide the remnant populations with the greatest evolutionary potential, and at the same time, minimise the negative consequences of inbreeding (e.g. Vrijenhoek 1994, Avise and Hamrick 1996, Haig 1998, Knapp and Rice 1998). Similarly, analysis of differentiation has been recommended for designing strategies for conservation (e.g. Moritz *et al*., 1995) and for selecting the best populations as translocation or restoration sources (Templeton 1986, Haig 1998, Knapp and Rice 1998).

Secondly, for basic evolutionary biological research, both quantitative and molecular data are valuable in evaluating the relative importance of genetic drift and natural selection as causes of population differentiation (Merilä and Crnokrak 2001). Since the differentiation of neutral marker genes is expected to be directed primarily by forces of genetic drift and migration (Hartl and Clark 1989) while that of genes coding quantitative traits is very likely affected by natural selection as well, the difference in standard coefficients of population differentiation can be used to deduce effects of selection on quantitative traits (Wright 1951, Rogers 1986, Spitze 1993, Merilä and Crnokrak 2001). The empirical work so far done on the basis of theoretical deliberations (Lande and Barroclough 1987; Lynch 1996) suggests that the correspondence between levels of genetic variability in neutral marker loci and loci coding for quantitative traits is poor (Cheverud *et al*. 1994, Butlin and Tregenza 1998; Waldmann and Andersson 1998; Pfrender *et al*. 2000; but see Briscoe *et al*. 1992).

In contrast, in a recent comparative study of the degree of population differentiation in marker genes (as measured by F_{ST}) and quantitative traits (as measured by Q_{ST}), Merilä and Crnokrak (2001) found that although the differentiation in quantitative traits typically exceeded that in neutral marker genes, the two measures of differentiation were positively correlated across different studies. However, in their review of empirical data, Reed and

Frankham (2001) failed to find any correlation between levels of differentiation between marker gene and quantitative traits. The difference in conclusions between these two studies could be explained by differences in the type of data included.

Any conclusions in respect to positive correlation between marker and quantitative trait divergence are also subject to some degree of uncertainty due to the limited number of empirical studies available. In fact, the across-species comparisons of differentiation in marker genes and genes coding for quantitative traits may give an overly optimistic picture of the correlation between the two measures (Merilä and Crnokrak 2001). In surveys of plant species, long-lived tree species with large dispersal capacities are often compared to short-lived herbaceous species with limited dispersal abilities. The effects of gene flow on Q_{ST} and F_{ST} will generally be different in the two groups, making comparisons highly dubious. Consequently, across-population comparisons within a given species may actually be more informative about the correspondence between marker and quantitative trait differentiation than comparisons made across different species.

To my knowledge, all intraspecific comparisons of genetic differentiation in marker genes and quantitative traits have so far focused on degree of differentiation (i.e. have compared mean F_{ST} and Q_{ST}). None has examined whether the pair-wise estimates among different populations are positively correlated. Hardy *et al*. (2000) made a comparison between analogues of pairwise Q_{ST} and $F_{ST,}$ but among individuals within a single population. Their results show that most quantitative traits have a significant spatial structure for their genetic component. Allozyme markers and the genetic component of quantitative traits generally show similar patterns of spatial autocorrelation.

1.2.2. Agroforestry and provenance studies

Trees and crops have been associated for many reasons, and with great benefits. For instance, cash crops can give returns while the farmer is waiting for the wood production. The association of timber tree species with tree crops such as coffee must be designed for optimal economic returns. Trees are planted to provide good shade and facilitate the cultivation of coffee. However, the growth of the tree component may be affected by the shade and by root competition of the main perennial crop, coffee. "Analogue forestry" has been proposed to imitate the diversity of species and strata in the natural forest. Such farming systems aim at maintaining the balance of nutrients, light, water, etc. and avoiding pests and diseases. Beer and Heuveldop (1989) and Beer *et al*. (1997) considered the management of natural regeneration of *C. odorata* in coffee (*Coffea arabica* L.) plantations. They consider *C. odorata* as one of best tree species for providing coffee shade.

Agroforestry has been proposed as a low external input system to alleviate the attack by the shootborer that precludes the establishment of Spanish cedar plantations on a commercial scale. Efforts have been devoted to studying the shootborer, particularly its biological and chemical control (Gripjma 1973, Newton et al. 1993, 1999, Mayhew and Newton 1998). Pruning methods to improve tree form after attack and to minimise the degree of damage are presented by Cornelius (2001). The use of antifeedant plant extracts of *Quassia amara* L., *Ruta chalepensis* L. and *Azadirachta indica* A. Juss have given promising results (Mancebo *et al*. 2000, 2001, 2002).

Provenance-progeny trials of *C. odorata* show considerable variation in growth and insect resistance. Some fast growing provenances were capable of producing one main shoot after *H. grandella* attack and retained a good form (Chaplin 1980, McCarter 1986). Quantitative trait variation has been reported in several studies (e.g. Burley and Lamb, 1971, Navarro and

Vasquez 1986, Navarro *et al*. 2002). Provenances from dry and wet areas in Costa Rica and Nicaragua differed significantly for seed and seedling traits (Navarro and Vasquez 1987). Studies of resistance of *C. odorata* to the attack by the shootborer indicate that provenances and families from dry areas were more resistant. However, those from wet areas grew faster (López *et al*. 1997).

1.2.3. Studies on reproductive isolation

The forest cover of Mesoamerica has been reduced drastically for agricultural production, cattle farming, fuelwood and human settlements. Fragmentation of the forest has caused reproductive isolation (Saunders *et al*. 1991), subsequent loss of genetic variability, reduced gene flow, and inbreeding depression (Templeton *et al*. 1990, Young *et al*. 1996, Young *et al*. 2000).

Studies of the impacts of logging and fragmentation point out the negative effects on reproduction due to isolation of the mature trees and destruction of the natural environment that would favour insect pollinators (Jennersten 1988, Aizen and Feinsinger 1994, Rocha and Aguilar 2001). Jennersten (1988) showed that habitat fragmentation resulted in a lower flower visitation rate and seed set in *Dianthus deltoides* when compared to non-fragmented habitats. The relevance of data on this small temperate dry-land herb to large humid tropic trees is certainly questionable, but no better comparison is presently available. Similarly, Aizen and Feinsinger (1994a) showed that pollination level and seed output decreased nearly 20% from forest to fragments in the Chaco region of the Republic of Argentina. These findings indicate that the reduction of continuous habitat can have a negative effect on the reproductive biology of plants. Rocha and Aguilar (2001) showed that seeds from pastures

are a poor source for establishing commercial plantations, as the resulting progeny is likely to be less vigorous than that from trees in continuous forests.

Inbreeding, in particular selfing, may lead to reduced fertility and slower growth rates of progenies (Hodgson 1976, Park and Fowler 1982, Sim 1984, Griffin and Lindgren 1985, Griffin 1991) because forest trees often carry a heavy genetic load of deleterious recessive alleles (e.g. Williams and Savolainen 1996, Eldridge and Griffin 1983). The risk of inbreeding must be seriously considered in activities dealing with genetic resources, use of germplasm in practical forestry and tree improvement.

2. OBJECTIVES

This work aimed at producing information that will help in the conservation and plantation of Spanish cedar under different forestry and agroforestry systems.

The objectives of this study were

1. To assess the genetic resources of *C. odorata* in the Mesoamerican region comprising the area between the Isthmus of Tehuantepec in Mexico and the River Atrato in Panama.

2. To evaluate the genetic variability for quantitative markers of *C. odorata* and the correspondence between (1) genetic variability in molecular markers and ecologically important traits (as reflected in additive genetic variance and heritability) and (2) degree of population differentiation in quantitative traits and molecular markers.

3. To evaluate the impact of fragmentation and mother tree isolation on the performance of *C. odorata* progenies.

4. To explore the use of agroforestry systems involving mixtures of *C. odorata* and coffee as an alternative farming system that helps control insect attack.

5. To discuss strategies for the efficient use and conservation of the genetic resources of this important species for the Mesoamerican region.

3. MATERIALS AND METHODS

3.1. COLLECTION OF SEEDS AND EVALUATION OF GENETIC RESOURCES

We collected *C. odorata* seeds from different types of sites chosen according to geographical climatic criteria, including topography, geology, soil type, vegetation, and land use. Socio-economic considerations included human population density, type of agriculture and availability of transportation and infrastructure. This information was used to define sampling areas and estimate the likely extent of within-species variation, based on their heterogeneity. We also determined the best time for collecting seeds with local informers and visits to the field.

To reduce the possibility of collecting seed from related or inbred trees, I took pollination biology and seed dispersal into account when determining the minimum distance between trees and populations. Bees, small wasps, moths, and thrips (Bawa et al. 1985, Patiño 1997, and Navarro 1999) pollinate the unisexual flowers of *C. odorata*.

No information was available about the movement of pollen and seeds in *C. odorata*, but I used information from other tropical tree species, e.g. in a disturbed area of tropical dry forest in Guanacaste Province, Costa Rica. Frankie *et al*. (1976) found that individuals of eight species of bees moved between trees 0.8 km apart. Long-distance pollen dispersal of up to 10 km by wasps has also been recorded (Nason *et al*. 1996). Considering these factors, the minimum collecting distance between trees in a population was set at 100 m, the distance of maximum flight recorded for seeds (Navarro *et al.* 2002a).

3.2. POPULATIONS STUDIED FOR MOLECULAR MARKERS; QUANTITATIVE TRAITS, AGROFORESTRY AND REPRODUCTIVE ISOLATION.

The 34 Mesoamerican populations collected in 1998-99 originated from the area covering the Tehuantepec Isthmus in Mexico to the Atrato River in Panama including the Yucatan Peninsula, corresponding to a latitudinal distribution that extends from ca 21°N in Mexico to 8°N in Panama (Table 1 and Fig.1). Consequently, the study populations cover an area of about 41 000 km^2 including a variety of environmental conditions. For instance, mean annual rainfall among the study populations ranges from 912 to 4818 mm and the number of dry months from zero to six (Table 1, Fig. 1). Table 1 contains the acronyms used in the subsequent tables to identify the populations.

Table 1. Characterisation data determined on the study populations, their coordinates with associated climatic data. NDM = number of dry months.

Country	Population	Acronyms	Latitude (°N)	Longitude (°W)	Altitude (m)	Rainfall (mm/year)	Rain (start-end)	NDM
Costa Rica	Cañas	CA	10.32	-85.04	100	2273.6[c]	May – Nov	5
Costa Rica	Carmona	CAR	10.01	-85.25	100	1779.9[c]	May – Nov	5
Costa Rica	Cóbano	CO	9.65	-85.12	20	2896.8[c]	May – Nov	5
Costa Rica	Hojancha	HO	10.07	-85.40	250	2232.3[c]	May – Nov	5
Costa Rica	Jiménez	GU	10.19	-83.79	240	4465.8[c]	May – Apr	0
Costa Rica	La Suiza	SUI	9.85	-83.61	670	2657.3[c]	Apr – Feb	1
Costa Rica	Liberia	LI	10.63	-85.45	150	1652.7[c]	May – Nov	5
Costa Rica	Pacífico Sur	PS	8.62	-82.88	40	4817.7[c]	May – Apr	0
Costa Rica	Pérez Zeledón	PZ	9.34	-83.65	700	2934.5[c]	Apr – Nov	4
Costa Rica	Quepos	QUE	9.42	-84.16	50	3851[c]	Apr – Dec	3
Costa Rica	San Carlos	SC	10.47	-84.58	90	4574.1[c]	Apr – Feb	1
Costa Rica	Talamanca	TA	9.65	-82.79	75	2812[c]	Apr – Nov	4
Costa Rica	Upala	UPA	10.86	-85.02	75	2558.3[c]	May – Jan	3
Guatemala	Los Esclavos	LE	14.25	-90.28	737	1929[a]	May – Oct	6
Guatemala	Tikal	TI	17.22	-89.61	250	1366.7[b]	May – Nov	5
Honduras	Cedros	CE	14.66	-87.30	555	1272[a]	May – Oct	6
Honduras	Comayagua	COM	14.41	-87.05	579	912[a]	May – Oct	6
Honduras	La Paz	PAZ	14.15	-87.61	726	1976[a]	May – Oct	6
Honduras	Meambar	MEA	14.83	-88.10	600	2425[a]	May – Oct	6
Honduras	Taulabe	TAU	14.83	-88.10	633	2425[a]	May – Oct	6
Mexico	Nachi-Cocoon	NA	18.48	-89.24	100	1094.0	Jun – Jan	4
Mexico	Bacalar	BA	18.85	-88.30	15	1400	Jun – Jan	4
Mexico	Blanca Flor	B	18.92	-88.49	100	1400	Jun – Jan	4
Mexico	Escárcega	ES	18.62	-90.78	100	1400	Jun – Jan	4
Mexico	Limones-Felipe	LFC	19.01	-88.00	50	1400	Jun – Jan	4
Mexico	Reforma-Bacalar	RB	18.85	-88.67	100	1400	Jun – Jan	4
Mexico	Tres Garantías	TG	18.12	-89.14	300	1600	Jun – Jan	4
Mexico	Tulum-FCP	TFC	19.35	-88.01	30	1400	Jun – Jan	4
Mexico	Xpujil	XPU	18.54	-90.14	150	1094[a]	Jun – Jan	4
Mexico	Yucatán	YU	20.59	-89.39	50	936[a]	Jun – Nov	6
Panama	Almirante	AL	9.28	-82.41	50	3319.0	Apr – Dec	3
Panama	Charagre	CHA	9.40	-82.56	50	3319[a]	Apr – Dec	3
Panama	Gualaca	GUA	8.59	-82.23	150	2620[a]	Apr – Nov	4
Panama	Las Lajas	LA	8.22	-81.86	20	2620[a]	Apr – Nov	4

[a] Data from: FAO 1985. Agroclimatological Data of Latin America and the Caribbean. FAO Plant Production and Protection Series. Roma. 19 p.
[b] Data from: Aguilar, M. and M. C. Aguilar. 1992. Arboles de la Biosfera Maya Petén. Universidad de San Carlos de Guatemala. 272 p.
[c] Data from: Ministerio de Recursos Naturales, Energia y Minas. Instituto Metereológico Nacional. 1988. Catastro de las series de precipitaciones medidas en Costa Rica. San José, Costa Rica. 361 p.

During collection the following information for each tree was recorded: population name, collector's name, date of collection, country, department or province, address, owner, climatic data (precipitation, temperature, number of dry months), slope, position (valley, slope, etc.), altitude, latitude and longitude (GPS), Holdridge life zone, land uses (primary forest, secondary forest, pasture, and agricultural field), associated species and characteristics of the tree: height, diameter at breast height, height of main stem, tree form, and phenological aspects. All the information was filed in a database and maps of distribution were made using GIS MapMaker software.

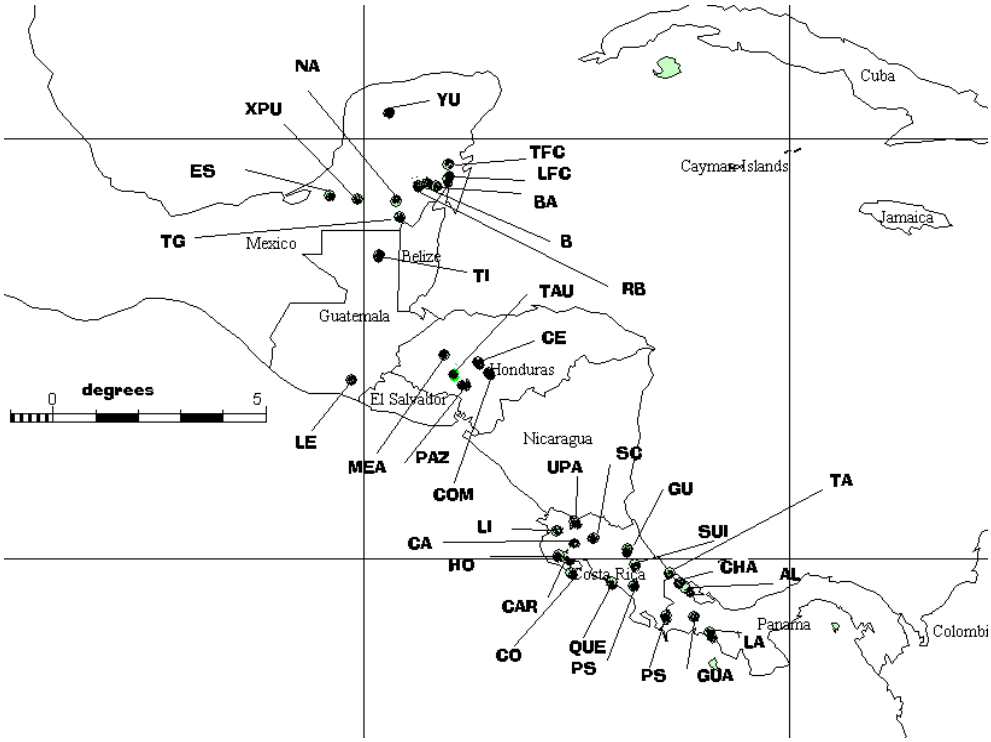


Figure 1. Map of the seed collections made of *Cedrela odorata* in Mesoamerica. C.Navarro.

3.3 DROUGHT ADAPTATION STUDY

The experiment for drought adaptation was made with 63 families selected at random from 14 provenances on the Atlantic and Pacific slopes of Costa Rica (Table 2 and Fig. 2). The provenances used are not the same as populations mentioned in Table 1 and were especially collected for the contrasts between mesic (m) and xeric (x) habitats. A habitat is considered as xeric when the dry season exceeds three months. *C. odorata* grows well on a wide range of soil types but it is intolerant of waterlogging on some clay soils. In addition, a provenance is a wider concept than population. Thus, the single trees collected within a provenance may in fact be far apart, virtually belonging to different populations.

The climatic data given for each provenance are taken from the nearest observation stations representing the collected provenance conditions. Provenances from the Atlantic and the Southern Pacific regions experience a shorter dry season than populations in the North Pacific region, and usually receive higher rainfall (Table 2). Each provenance was assigned accordingly to either the mesic or xeric climatic group. The dry areas are not necessarily very arid, and have a period of at least 1000 mm of precipitation, but the dry period may last up to six months in the North Pacific region of Costa Rica.

The field experiment was located in Turrialba, Costa Rica (Lat. 9.86 ^{o}N, Long. 83.62 ^{o}W rainfall 2657mm/year, one dry month, mean annual temperature 21 ^{o}C). Seeds were germinated in a bed filled to approximately five cm with sand previously washed and sterilised with formalin. They were positioned vertically with the embryo closest to the substrate, but were not covered with sand. This position had resulted in superior germination in previous trials. Humidity of the seedbed was kept constant to avoid desiccation and inhibit fungal growth. Seeds germinated in 7 to 12 days.

Seedlings were removed from the germination bed about a week after germination and carefully transplanted avoiding desiccation. The seedlings were planted in plastic bags 10.2 cm wide by 20.3 cm long, in a soil mix of one part fine sand, three parts compost, and with 50 g of a complete fertiliser (10-30-10) added. Plants were kept under a shade cloth (50 % light penetration) for two weeks after removal from the germination bed to permit recovery from transplanting.

A randomised complete block design was used in the field with families as the treatments, and using one tree plots in the three replications.

The model for analysis of variance was:

$$Y_{ijkl} = \mu + B_i + C_l + P_k(C_l) + F_j(P_k*C_l) + e_{ijkl},$$

where Y_{ijkl} is the phenotypic value of an individual tree, μ is the experimental mean, B_i is the block effect, $F_j(P_k*C_l)$ is the family effect within provenance and climate, $P_k(C_l)$ is the effect of the provenance within climate, C_l is the effect of the climatic group to which the provenance belongs and e_{ijkl} is the experimental error.

A sample of 100 seeds from each of the 63 families was weighed and the length and width was measured on five seeds per family. The ratio of seed width to seed length was calculated.

At 73 days after planting, seedlings were measured for total plant height in cm (ht), root collar diameter (rcd) in mm, length (ll) and width (lw) of the largest leaflet in mm**.** Leaflet width was divided by leaflet length to obtain an index of leaflet shape (lw/ll).

Table 2. Meteorological data of the provenances and families evaluated from xeric and mesic origins (closest met. station in brackets)

Provenance	Number of Families	Rainfall(mm)	Number of dry months	Altitude (m)	Life zone[a]	Climatic Group[b]
Cañas (Las Juntas)	5	2 273	5	140	TMF	x
Hojancha (Nicoya)	5	2 232	5	120	TMF	x
Carmona (Colonia Carmona)	5	1 779	5	100	TMF	x
Cobano (Cobano)	3	2 897	5	160	TMF	x
Cobano (Cabuya)	7	2 873	4	3	TMF	x
Liberia (Llano Grande)	5	1 652	5	85	TDF	x
Talamanca (Vesta, Penshur)	2	3 981	0	50	TMF	m
Talamanca (Chase, Bri – Bri)	2	2 662	0	40	TMF	m
Guapiles (Los Diamantes)	4	4 465	0	250	PWF	m
Upala (Upala)	4	2 558	3	50	TWF	m
San Carlos (La Fortuna)	7	3 608	0	250	PWF	m
San Carlos (Santa Clara)	10	4 317	0	160	TWF	m
Zona Sur (Palmar Sur)	1	3 706	3	16	PWF	m
Zona Sur (Golfito)	3	4 817	0	15	TWF	m

[a] TDF tropical dry forest, TMF tropical moist forest, PWF premontaine wet forest, TWF tropical wet forest. (After Holdridge 1967)

[b] x= xeric, m = mesic

Least square means were estimated for all seedling traits for provenance and drought groupings using the LSMEANS statement of PROC GLM (SAS Institute 1999). The Scheffe adjustment for least square means comparisons was used, as these were *post hoc* comparisons (SAS Institute 1999). Cluster analysis was based on family level means for seedling traits, and on family and provenance level means for seeds. The unweighted group-pair method (UPGMA) of obtaining linkages was applied on the basis of unsquared arithmetic means standardised to an overall mean of zero and standard deviation of unity. (PROC CLUSTER, SAS Institute 1999). Analysis of variance was performed using PROC GLM (SAS Institute 1999), with the climatic grouping as a fixed effect; all other effects were considered to be random.

In the analysis of variance (ANOVA) the individual effects may be significant, but contribute only marginally to the explanation of the overall variance. Thus, variance component analysis has been developed to determine the relative contributions of different effects to total variance (Fleiss 1969, Underwood and Petraitis 1993), including models with both fixed and random effects (Vaughan and Corballis 1969, Dodd and Schultz 1973). To determine the components of variance, the expected mean sums of squares were calculated for each effect with Proc VARCOMP (SAS Institute 1999), according to the formulas provided by Winer *et al*. (1991). Variance components were calculated using restricted maximum likelihood of PROC VARCOMP of SAS/STAT software ® (SAS Institute 1999). This method is relatively robust for both unbalanced designs (Huber et al. 1994) and departures from normality (Westfall 1987).

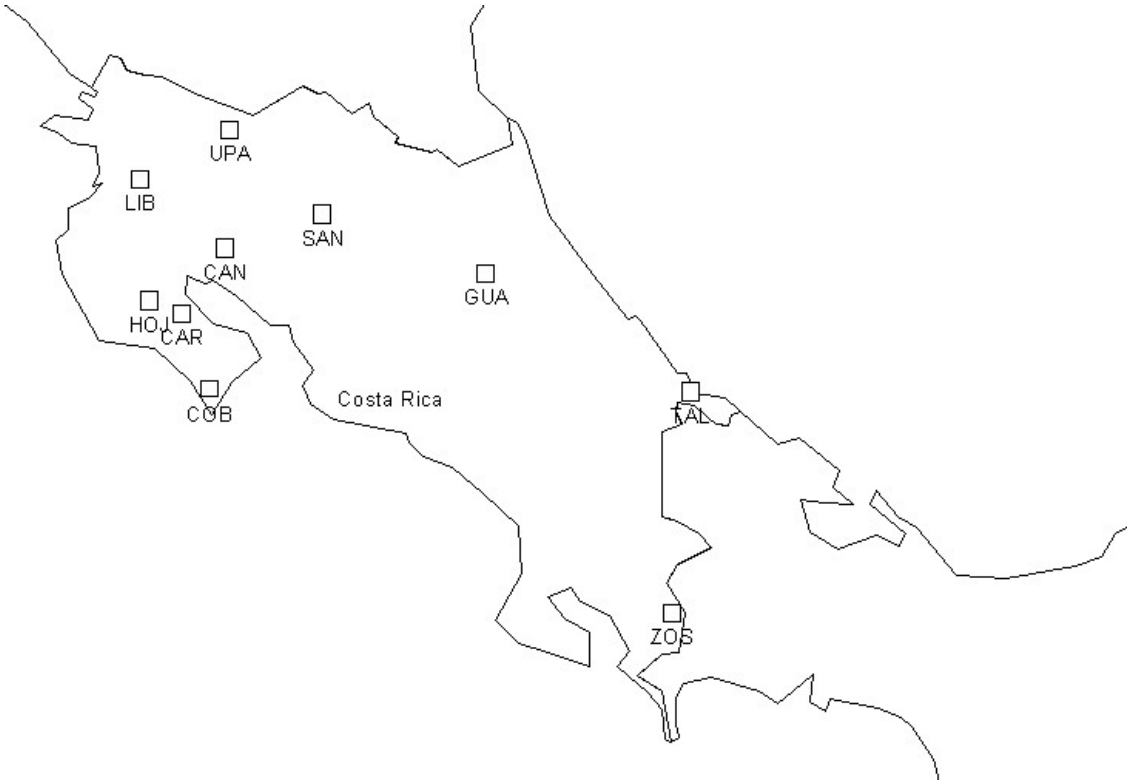


Figure 2. Distribution of *C. odorata* provenances used in the drought study in Costa Rica.

3.4. MOLECULAR MARKER STUDY

To characterise molecular genetic variability using RAPD-markers, I was confined to genotypic material from only 14 populations as indicated in Table 12. All populations came from the collection indicated on Figure 1 and Table 1.

DNA samples were extracted from germinated seeds of single mother trees with the CTAB method as described in Wilson *et al.* (2001). The PCR protocol included amplification of the DNA in a 25 µl volume using 10 pairs of Operon Technologies Ltd. standard primers (OPC1–10). A MJR thermal cycler followed a programme consisting of 45 PCR cycles each comprising 1 minute to 94 ºC (denaturing), 1 minute to 36 ºC (first-DNA union), 2 minutes at 72 ºC (extension) and a 72 ºC final cycle of 7 minutes (final extension). Each PCR reaction included 1/10 buffer, 2 mM dNTP, 1 unit of dynazyme taq polymerase (Finnzymes) and 0.4 µm primers made up to 25 µl with distilled water. The products were visualised under UV transillumination (in 0.1% TBE buffer containing few drops of ethidium bromide) after separation in 1.8% SIGMA agarose gels. The RAPD phenotypes were classified into groups giving similar banding patterns (Pappinen *et al.*, 1996). On average, 13 (range: 8 – 19) individuals were genotyped from each of the populations.

Shannon's Diversity Index (SDI) was used as a measure of intra-population genetic diversity. The index was calculated using POPGENE v1.31 (Yeh and Boyle 1997). It is well suited to the analysis of RAPD data as it is relatively insensitive to the bias produced by failures to detect heterozygous individuals (Dawson *et al*. 1995). For the calculation of $F_{ST,}$ seven populations were genotyped at Helsinki University and data from seven additional populations were obtained from Gillies *et al*. (1997). For this reason the analysis was made on two sub-sets of data: Subset 1, populations genotyped in the laboratories of the University

of Helsinki; Subset 2, populations genotyped by Gillies *et al*. 1997. In addition I made a pooled data analysis.

The coefficient of population differentiation, F_{ST}, was obtained by partitioning the variability in the data into within-V_w and between-V_b population components:

$$F_{ST} = \frac{v_b}{v_b + v_w} \qquad (1)$$

Standard errors were obtained using a Bayesian approach (Holsinger and Lewis 2002 and Holsinger *et al.* 2002). Random amplified polymorphic DNA markers (RAPDs), allow analysis of species for which previous DNA sequence information is lacking, but dominance makes it impossible to apply standard techniques to calculate *F*-statistics. The method is constructed in terms of the classical *F*-statistics of Wright (1951) and Malécot (1948). The Bayesian method allows direct estimates of F_{ST} from dominant markers. In contrast to existing alternatives, it does not assume previous knowledge of the degree of within-population inbreeding. In particular, it does not assume that genotypes within populations are in Hardy–Weinberg proportions. The estimate of F_{ST} incorporates uncertainty about the magnitude of within-population inbreeding. Simulations show that samples from even a relatively small number of loci and populations produce reliable estimates of F_{ST}.

3.5. QUANTITATIVE GENETIC ANALYSES

The seedlings for the quantitative study were grown in a nursery experiment in a greenhouse of the University of Helsinki. The seedlings were raised in a mix containing 10 % sand, 40 % vermiculite and 50 % peat. Temperature (25 °C), humidity (90 %) and day-length (12:12 dark/light) in the greenhouse were kept constant, and the seedlings were watered daily.

Estimates of within-population genetic variability and the coefficient of population genetic differentiation were based on a nursery experiment using a randomised complete block design. An average of 13.5 (4 – 22) seeds was sampled from each open-pollinated mother tree. One seedling per family was sown in each of the six blocks. Because of some mortality (1.2 %) during the experiment, only an average of 5.6 individuals per family (1080 in total) were measured for the traits described below. The wide range of variation in the number of families utilised per population has a simple explanation: *C. odorata* is a scarce and endangered species, and the number of families per population utilised in the experiment reflects the local population sizes (Table 12).

At 62 days after sowing the following measurements were taken: (1) height (H62 in mm), (2) leaflet length (LL62 in mm), (3) width of the third leaflet from the tip of the leaf LW62), and (4) leaflet shape index obtained by dividing leaf length by leaflet width (LL/LW62). At 252 days after sowing four measurements were taken: (5) height (H252 in cm), (6) internodal distance (ID252; the length of the stem from the tip to the fourth branch in cm), (7) stem base diameter (D252 in cm 2 cm above the soil) and (8) the number of leaflets per leaf (NL252). The weights of fresh leaves (FLWE), branches (FBWE) dry leaves (DLWE) and branches (DBWE) were taken. The mean values (± Standard errors) of these traits are given in Appendix 1.

The averages of the minimum squares were determined for all the variables for each lineage and climatic grouping using LSMEANS of PROC GLM of SAS/STAT software (SAS Institute 1999). The Scheffe adjustment for LSMEANS was used (SAS Institute 1999). The variance analysis was made using PROC GLM (SAS Institute 1999), with the groups purple (mesic) or green (xeric) as a fixed effect, and all the other effects as random variables. The variance components were determined using the restricted maximum likelihood procedure (PROC VARCOMP, SAS Institute 1999). An analysis of conglomerates was made using family means for all the quantitative and qualitative characteristics using Euclidean distances and the method PROC CLUS AVERAGE (SAS Institute 1999) applied to examine the similarities between families and populations.

To obtain a standardised estimate of among-population differentiation comparable to F_{ST} for molecular markers, we estimated Q_{ST} values as:

$$Q_{ST} = \frac{\sigma^2_{GB}}{2\sigma^2_{GW} + \sigma^2_{GB}} \qquad (2)$$

where σ^2_{GB} is the among-population component of genetic variance, and σ^2_{GW} is its within-population genetic component (Wright 1951, Merilä and Crnokrak 2001).

3.6. STATISTICAL METHODS FOR COMPARING QUANTITATIVE AND MOLECULAR MARKERS

Molecular variability for RAPD markers was estimated with Shannon's Diversity Index while quantitative variation was assessed by means of heritability estimates. The comparisons between the two parameters thus obtained were made using Spearman pairwise product

moment correlations (1) for each of the traits separately and (2) for the mean of the heritability estimates for different traits. In view of the fact that the different traits vary largely both in terms of their size and dimensionality, I carried out these analyses also using coefficients of additive genetic variability (CV_A, Houle 1992). Because the genetic variability measures based on RAPD markers may not be comparable between the two sub-sets of data (cf. our data and data from Gillies *et al*. 1997), I performed the tests also for data involving (1) only populations scored for RAPDs by me, (2) only populations scored for RAPDs by Gillies *et al*. (1997) and (3) on combined data.

To compare the levels of molecular genetic differentiation F_{ST} and quantitative genetic differentiation Q_{ST}, I first compared the overall estimates of F_{ST} and Q_{ST} for the two sub-sets of data using two-sample *t*-tests. In these tests, each locus and trait was considered as an independent observation. To see whether estimates of F_{ST} and Q_{ST} calculated pair-wise among all possible pairs of populations are correlated, I performed a Mantel's test (5000 permutations) on F_{ST} and Q_{ST} (averaged over traits) estimates.

All tests were performed with SAS statistical software, Version (8) of the SAS System for Windows, except for Mantel's tests, which were conducted with an Excel add-in 'PopTools' (version 2.3 available at www.cse.csiro.au/CDG/poptools).

3.7. PERFORMANCE OF *C. ODORATA* IN ASSOCIATION WITH COFFEE

3.7.1. Provenances and coffee components

The 21 provenances included in this study (Table 3) cover an area from the Tehuantepec Isthmus in Mexico, including the Yucatan Peninsula, to the Atrato River in Panama.

Latitudes range from 21°N in Mexico to 8°N in Panama. The provenances represent different soils and a variety of climatic conditions, from the very dry seasonal climate of North Yucatan and the Pacific Coast of Costa Rica, to the annually wet Atlantic region of Costa Rica and Panama. Annual rainfall varies from 1094 to 4818 mm, and the number of dry months from zero to six (Table 1).

Seeds are from single tree collections (Navarro *et al*. 2002) kept in cold storage at the seed bank of the Tropical Agricultural Research and Higher Education Centre (CATIE) in Costa Rica. Parent trees were not selected for traits of any kind. The nursery conditions and the plantation techniques are described in Navarro and Hernández (2001).

The seeds were sown during the second week of June 1999 in the nursery of CATIE and the seedlings were planted in November, 1999 in a 12 ha coffee plantation belonging to CATIE (Lat. 9.86, Long. 83.62, Alt. 625 m, annual rainfall 2657 mm, 1 dry month (Salas 2002)).

Table 3. Description of the populations and single tree families evaluated in the agroforestry trial of *C. odorata* mixed with coffee in Turrialba, Costa Rica.

Country	Populations	Families
Costa Rica	CA	662, 663, 699, 6270
Costa Rica	CO	6110, 6112, 6114
Costa Rica	GU	6141, 6145
Costa Rica	HO	6105, 6108, 6166, 6176, 6101, 6103
Costa Rica	PS	6207, 6213
Costa Rica	PZ	6232, 6240, 6274
Costa Rica	SC	683
Costa Rica	TA	6121, 6123, 6125
Costa Rica	UPA	6177, 6189
Guatemala	LE	32, 33, 35, 36, 38, 39, 312, 314, 317, 319
Guatemala	TI	341, 343 – 346, 349, 351 – 355
Honduras	CE	446 – 449, 451, 454, 456, 457, 459
Honduras	PAZ	44 – 46, 48, 410, 412 – 414
Honduras	MEA	467, 469, 470, 472
Honduras	TAU	422, 423, 426 – 430
Mexico	BA	134, 139,187, 192, 194
Mexico	TG	144, 146, 147
Mexico	TFC	168, 170, 171, 175 – 177, 180, 182, 185
Mexico	XPU	11, 112, 115
Panama	AL	711–714, 71–710, 715
Panama	GUA	745, 747, 752, 766, 768

Cedar-coffee mixtures were designed as follows: 1) coffee just planted (1 month old), 2) coffee with total pruning (10 years old) and 3) coffee in production (5 years old). Two systems of cultivation were used: 1) cedar planting between coffee rows (BCR) and 2) cedar within row planting (WCR). Details are set out in Table 4.

Table 4. The cedar-coffee trial at CATIE. Cedar planting between coffee rows (BCR) and cedar within row planting (WCR)

Block number	**Coffee conditions, height (m)**	**Cedar spacing (m)**	**Cedar plants per plot**	**Topography**	**Cedar plantation**
1	Young coffee, 0.50	3x6	1	Flat terrain	WCR
2	Young coffee, 0.50	3x6	2	Smooth slope	WCR
3	Old coffee pruned, 1	6x6.5	2	Flat terrain	WCR
4	Production coffee, 1.8	6x7	1	Flat terrain	WCR
5	Production coffee, 1.8	6x6	2	Flat terrain	WCR
6	Production coffee, 1.8	6x6	2	Flat terrain	BCR
7	Production coffee, 1.8	3x6	2	Flat terrain	BCR
8	Production coffee, 1.8	3x6	2	Flat terrain	BCR
9	Production coffee, 2.0	6x7	2	Smooth slope	BCR
10	Production coffee, 2.0	6x7	1	Smooth slope	WCR
11	Production coffee, 1.8	6x7	2	Smooth slope	WCR
12	Production coffee, 1.8	6x7	2	Smooth slope	WCR
13	Production coffee, 1.8	6x7	2	Smooth slope	WCR
14	Production coffee, 1.8	6x6	2	Smooth slope	WCR
15	Production coffee, 1.8	6x6	2	Smooth slope	WCR
16	Production coffee, 1.8	6x6	2	Smooth slope	WCR
17	Production coffee, 1.8	6x6	1	Smooth slope	WCR

3.7.2. Experimental design and measurements

The experiment was made with 115 selected families from the original collection (progenies from single trees) from 21 provenances. The families were distributed randomly in 17 blocks, in plots of one or two trees.

The statistical analysis was based on the following linear model:

$$Y_{jkl} = \mu + B_j + f_k + fB_{jk} + w_{kjl}$$

where: Y_{jkl} = the l^{th} tree in the j^{th} block and k^{th} family, μ = general mean; B_j = effect of the j^{th} block, f_k = random effect of the k^{th} family, fB_{jk} = random plot error due to interaction between j^{th} block and k^{th} family; w_{jkl} random tree error of l^{th} tree in $j_k{}^{th}$ plot.

The variables measured on seedlings were: (1) root collar diameter (mm), (2) height (cm), (3) number of attacks of the shootborer (susceptibility) and (4) the number of shoots that re-sprouted after attacks of *H. grandella* (recovery). The last two variables are the cumulative summation of 18 measurements at regular one-month intervals. Only the presence or absence of attack was noted, no attempt was made to evaluate the intensity of attack. When there was a shootborer attack, the tree was pruned back to the end of the larval tunnel and the larvae were thereby eliminated.

The first evaluation after planting was done in June 2000 and the last in December 2001, 25 months after planting. Susceptibility and recovery were measured once a month. Diameter and height were measured quarterly. Exploratory analyses of the data using Proc Univariate of SAS software were done for the population means for all traits to test for normal distribution and equal variances. The variables were evaluated using the General Linear Model of SAS.

A multivariate analysis of variance (MANOVA) was done with the mean values of the quantitative traits.

3.7.3. Calculation of heritabilities and coefficients of additive variance

The PROC MIXED and PROC VARCOMP with the algorithm REML procedure of SAS software were used to estimate heritabilities. Heritabilities have to take into consideration the population effects of all the progenies included in the trial (115). In this study the calculation for heritabilities was done in two ways: first only for the best local provenances from the wet areas and second for all the progenies using only the within populations component. For more details on the procedure, see Hodge *et al*. (2002).

The heritabilities were calculated using the formula $h^2 = 3 \times \sigma^2_F / V_P$, where σ^2_F is the variance component due to family and V_P is the total phenotypic variance of the trait (i.e. $\sigma^2_F + \sigma^2_{B*F} + \sigma^2_E$), where σ^2_{B*F} and σ^2_E are the interaction block by families, and error respectively. The coefficient of 3 instead of 4 was based on the following deliberation: 1) *C. odorata* is a rare species with very small effective population sizes due to excessive exploitation. 2) Short distance pollinators, mainly small bees (Navarro 1999) and small moths (Bawa *et al*. 1985) probably pollinate *C. odorata*, and the effective number of male tree pollinators is sometimes less than 20 in a population. Thus a small effective population size, short distance pollinators and fragmentation of the forest could lead to the presence of some full-sibs and inbreeding in the open pollinated mother tree, and subsequent overestimation of the heritability (Squillace 1974). The heritability standard errors were calculated according to Dieters *et al*. (1995). Coefficients of additive genetic variance (CV_A; Houle 1992), were calculated as $CV_A = 100 \sqrt{V_A} / x$, where V_A is the additive genetic variance, and x is the mean trait value.

3.8. REPRODUCTIVE ISOLATION AND FRAGMENTATION INFLUENCE ON GROWTH TRAITS

This study involved gathering information about seed trees in natural conditions and ecological aspects of the sites where the trees were sampled. The isolation conditions of the mother trees were defined using the following key: 1) Isolated mother tree (no other trees of the same species closer than 500 m), 2) Semisolated (other trees no closer than 100 m) and 3) Mother tree in clusters or associated with more than two trees within a radius of less than 100 m. This variable was analysed with orthogonal contrasts (*a priori* statistical test), analysis of variance and Tukey means comparisons (*a posteriori* statistical test).

Seeds from 115 families were selected from the original collection based on the variability of climatic conditions and latitudinal distribution and sown in a nursery in Turrialba, Costa Rica in June, 1999. Seedlings were subsequently planted in October-November in the experimental farm of CATIE. The number of seedlings for each level of isolation and population is presented in Table 5.

Experimental design: The experiment had a randomised block design, 17 blocks and 2 plants per plot.

The following linear model was used for the analysis of isolation:

$$Y_{jkl} = \mu + B_j + I_k + w_{kjl}$$

where: Y_{jkl} = height of the l^{th} tree in j^{th} block and k^{th} isolate, μ = general mean; bj effect of the j^{th} block, I_k = random effect of the k^{th} isolate, w_{jkl} random tree error of l^{th} tree in $j_k{}^{th}$ plot.

The seedlings were measured for: 1) height (cm) and 2) number of attacks of the shootborer (susceptibility). Based on previous studies of the species, the mother trees were divided into

xeric (populations of seasonal areas with more than 3 months of dry period) and mesic (non seasonal areas with more than 100 mm of rain per month) (Navarro *et al*. 2002ab)

Table 5. Description of isolated mother trees, number of seedlings evaluated per mother tree and corresponding population. Isolated mother trees experiment, Turrialba, Costa Rica.

Population	**Number of seedlings (isolation class)**[a]
CA	30(1),30(2)
CO	60(2), 30(3)
HO	30(1),150(3)
LI	30(1), 30(2)
GU	30(2), 30(3)
PS	30(2), 30(3)
PZ	55(2), 30(3)
SC	27(2)
TA	30(2), 60(3)
UPA	30(2), 30(3)
LE	300(1)
TI	326(3)
CE	269(3)
PAZ	237(1)
MEA	120(1)
TAU	206(1)
BA	90(1), 30(2), 30(3)
TG	90(1)
TFC	210(1),60(2)
XPU	90(1)
AL	91(1), 360(3)
GUA	30(1), 122(3)

[a] See page 34 for isolation classes.

4. RESULTS

4.1. STATUS OF DISTRIBUTION OF THE SPECIES ACROSS MESOAMERICA

The survey and collection field trips indicate that the species is highly endangered. On the basis of this survey, I consider that over 90% of the distribution has been overexploited and many areas where trees occur are no longer in natural forest. Many of these areas are used for agriculture, urban settlements and cattle farming. The lack of samples from Nicaragua and the Olancho area of Honduras and logistic limitations in carrying out the work in these regions are recognised, but during surveys of these areas, I found no populations suitable for collection.

In most cases, the trees were found in man-made grasslands earlier covered by tropical mixed forests. Interestingly, many people had protected them in their home gardens and patios as they recognized their value. The status of the tree populations collected is indicated in Table 6.

C. odorata was present in non-flooded areas in the surveyed countries; most trees grew in zones with good conditions for human living and/or farming. The most deforested *C. odorata* areas lie on the Pacific coast of Mesoamerica. Janzen (1988) indicates that 98 % of the Central American dry forests have been destroyed. Around 70–90% of the natural distribution of important species like *C. odorata* and *Swietenia macrophylla* K. show signs of exploitation that range from selective logging to total extinction of some populations (Navarro 1999, Navarro 2001, Navarro *et al*. 2002c). In Mesoamerica, natural forests containing important populations of *C. odorata* have been reduced to less than 10 % of the area described by Pennington (1981). Not only has the area declined but it has also become

increasingly fragmented by agriculture, cattle farming, and selective cutting. *C. odorata* is now more abundant in grasslands and agroforestry systems than in natural forests.

Table 6. Status of *C. odorata* populations across Mesoamerica.

Population	**Population characteristics**	**Associated species**
Cañas	Grasslands and scattered trees	*Cassia grandis*
Cóbano	Grasslands and scattered trees	*Gliricidia sepium, Spondias, Bombacopsis quinata, Cassia grandis, Anacardium excelsum*
Guápiles	Grasslands, trees in home orchards and scattered in the grasslands	Grasses
Hojancha	Grasslands and scattered trees	*Hyparrhenia (grass), Cordia, Bombacopsis, Enterolobium, Guazuma, Schizolobium parahybum, Pithecellobium, Cassia grandis*
Liberia	Grasslands	Grasses
Pacífico Sur	Grasslands, scattered trees, secondary forests	*Hyeronima, C. odorata, Zanthoxylum, Spondias, Cordia, Carapa, Inga, Tabebuia, Ceiba*
Pérez Zeledón	Grass, coffee, scattered trees, secondary forests	*Vochysia, Didymopanax, Anacardium excelsum, Gliricidia*
San Carlos	Grasslands	Grasses
Talamanca	Grasslands	Grasses
Upala	Grasslands	*Cordia alliodora, Cecropia, Inga, Bursera simarouba, Gliricidia sepium, Zanthoxylum.*
Los Esclavos	Grasslands, crop fields	Grasses
Tikal	Natural forest	*Swietenia macrophylla, C. odorata, Trichilia, Achras, Spondias mombin, Ceiba pentandra*
Cedros	Home orchards, grasslands and river borders	Grasses
La Paz	Grasslands	Grasses
Meambar	Grasslands	Grasses
Taulabé	Grasslands	Grasses
Bacalar	Grasslands, close to village	Grasses, fruit trees, *Achras, Brosimum utile, C. odorata, Pimienta dioica*
Tres Garantías	Grasslands and crop fields	Maize, grasses
Tulum	Fruit trees close to village	*Mangifera indica*
Xpujil	Grasslands, deforested	Grass, maize.
Almirante	Grasslands and fragments of secondary forests.	*Cordia alliodora, Ficus maxima, Terminalia oblonga,T.cattapa, Cecropia peltata, Ochroma lagopus, Spondias mombin, Guazuma ulmifolia, Luehea seemannii, Hyeronima alchorneoides, Erithryna poeppigiana, Byrsonima crassifolia*
Gualaca	Fragmented secondary forests and cattle farms,	*Cordia, Ficus, Terminala, Inga, Gliricidia sepium, Guazuma, Spondias, Ochroma, Bursera simarouba, Vochysia ferruginea, Enterolobium cyclocarpum, Byrsonima crassifolia, Miconia, Tabebuia, Albizzia, Cecropia*

4.2. DROUGHT ADAPTATION

Xeric (X) and mesic (M) populations of *C. odorata* differ in morphology and adaptation (Tables 7 and 8). Seedlings from Cañas had the greatest dimensions for all the traits and those from Zona Sur the smallest dimensions (Table 7).

Table7. Least square means and standard errors (SE) by provenance for selected seed and seedling traits.

Group	Provenance	Seed weight			Seedling height			Seedling rcd			Leaf length			Leaf width			Ratio lw/ll		
		lsm	SE	sep[a]	lsm	SE	sep	lsm	SE	sep	lsm	SE	sep	lsm	SE	sep	lsm	SE	sep
X	Cañas	2.31	0.14	1	16.4	0.47	1	6.47	0.22	1	7.61	0.19	1	3.13	0.11	1	0.41	0.02	1
X	Carmona	2.05	0.13	-	14.73	0.47	1	6.07	0.22	1	7.11	0.19	1	2.73	0.11	1	0.39	0.02	-
X	Cobano	2.20	0.09	1	14.67	0.33	1	6.33	0.16	1	7.95	0.14	1	3.05	0.08	1	0.38	0.01	1
X	Hojancha	2.35	0.13	-	14.47	0.47	1	6.13	0.22	1	6.94	0.19	1	2.75	0.11	1	0.40	0.02	1
X	Liberia	2.17	0.11	1	15.67	0.47	1	6.27	0.22	1	7.93	0.19	1	3.04	0.11	1	0.38	0.02	-
M	Guápiles	1.32	0.14	-	11.17	0.52	-	3.58	0.25	2	5.87	0.22	-	1.83	0.12	2	0.31	0.02	-
M	San Carlos	1.25	0.07	2	9.65	0.26	2	2.81	0.12	2	5.44	0.11	2	1.61	0.06	2	0.30	0.01	2
M	Talamanca	1.32	0.14	-	8.58	0.52	2	2.75	0.25	2	6.02	0.22	-	1.65	0.12	2	0.27	0.02	-
M	Upala	1.21	0.13	-	10.4	0.47	2	3.07	0.22	2	5.57	0.19	2	1.63	0.11	2	0.29	0.02	-
M	Zona Sur	1.26	0.14	2	6.67	0.52	2	2.08	0.25	2	4.3	0.22	2	1.45	0.12	2	0.33	0.02	-

[a] In the sep (separation) column, provenances with the same value do not differ significantly from one another. Provenances with different values differ at the 0.01 significance level (Scheffe). Provenances marked with hyphen (-) do not differ from either group.

Seeds from dry regions were 43 % heavier and seedlings 61 % taller and 117 % greater in diameter than those from the moist regions (p=0.0001 for all traits, Table 8). Leaflets of seedlings from the dry areas were 39 % longer, 81 % wider, and 25 % more ovoid (p=0.0001 for all traits, Table 7).

TABLE 8. Least square means and standard errors by regional grouping for selected seed and seedling traits. Seedling root collar diameter (rcd) and ratio leaf width/leaf length (lw/ll).

Climatic group	**Seed weight**			**Seed length**			**Seed width**		
	lsm	SE	sep[a]	lsm	SE	sep	Lsm	SE	sep
X	2.22	0.05	1	0.98	0.03	1	0.41	0.01	1
M	1.27	0.06	2	0.74	0.03	2	0.34	0.01	2
	Seedling height			**Seedling rcd**					
	lsm	SE	sep	lsm	SE	sep			
X	15	0.2	1	6.3	0.09	1			
M	9.3	0.21	2	2.9	0.1	2			
	Leaf length			**Leaf width**			**Ratio lw/ll**		
	lsm	SE	Sep	lsm	SE	sep	Lsm	SE	sep
X	7.5	0.08	1	2.9	0.05	1	0.4	0.01	1
M	5.4	0.09	2	1.6	0.05	2	0.3	0.01	2

[a] Separation by the Scheffe criteria at the 0.0001 significance level.

Xeric and mesic regional groups differed at the 0.0001 level of significance for all traits. Statistical differences between the climatic groups were always highly significant, and explained on average 66 % of the total variance (Table 9). For seedling traits climatic group, too, was always very significant, and contained most of the explained variance (mean of 52 % of total variance or 80 % of the genetic variance, Table 9). Provenance level effects within climatic regions were never significant, and explained very little of the total variance. For seedling traits, family level effects within provenances were usually highly significant, and explained a mean of 31 % of the total variance or 20 % of the genetic variation (Table 9). Results were confirmed with cluster analysis on seedling traits (Fig. 3) and seed (Fig. 4) traits, which revealed two natural groupings of families: 1) an Atlantic and Southern Pacific group and 2) a dry Northern Pacific group.

Table 9. Significant effects (p) from analysis of variance and percentage of total variance explained by each variance component (vc %) for seed and seedling traits[b].

	Seed							Seedling										
	Weight		Length		Width		Mean[b]	Height		rcd		Leaf Length		Leaf Width		Ratio lw/ll		Mean
EFFECT	p[a]	vc%	p	vc%	p	vc%		p	vc%	p	vc%	p	vc%	p	vc%	p	vc%	vc %
Var(block)								ns	0	**	1	ns	0	ns	0	ns	0	0
Var(clim)	***	86	***	62	***	51	66	***	52	***	71	***	44	***	63	***	31	52
Var[pop(clim)]	ns	0	ns[d]	0	ns	0	0	ns	3	ns	0	ns	4	ns	0	ns	0	1
Var[fam(clim*pop)]								***	25	***	11	***	30	***	11	ns	0	15
Var(error)		14		38		49	34		20		17		22		26		69	31
% Vb [c]		nd		nd		nd	nd[e]		68		87		61		85		100	80
% Vw[c]		nd		nd		nd	nd		32		13		39		15		0	20

[a] ns p>.05, **p<.01, ***.0001>p

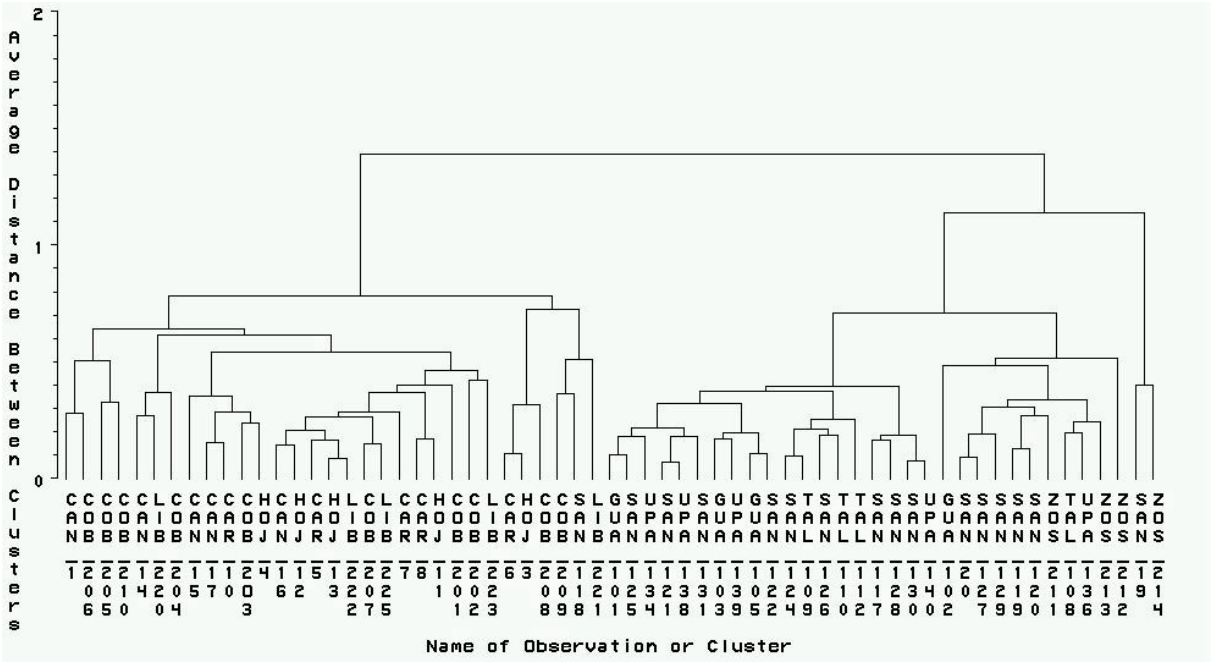
[b] Mean variance components.

[c] For each trait the total genetic variation is divided into between-provenance (clim + pop) and within-provenance (fam) variation.

Vb % refers to the genetic variance between provenances. Vw % refers to the genetic variance within provenances.

[d] non-significant.

[e] not determined.



Can = Cañas, Cob= Cobano, San= San Carlos, Car=Carmona, UPA= Upala, Tal= Talamanca, Zos=Zona Sur, Lib = Liberia, Hoj=Hojancha, Gua = Guápiles.

Fig. 3. Phanerogram from cluster analysis on seedling traits with a clear separation between Atlantic and Pacific groups in *C. odorata*. For population acronyms see Table 1.

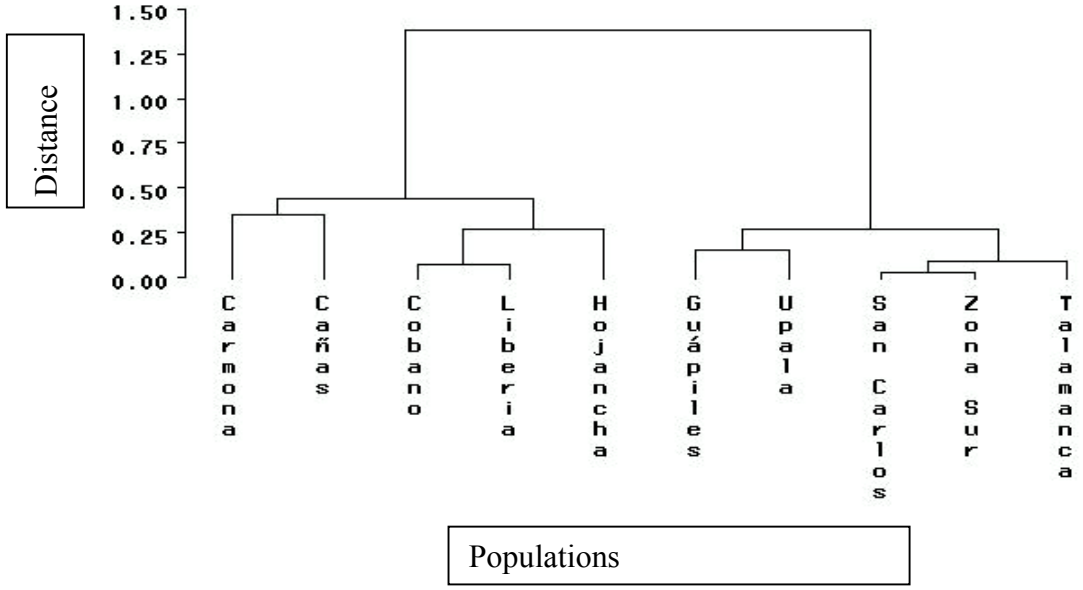


Fig 4. Phanerogram from cluster analysis on seed traits showing the separation of the populations into two main groups.

4.3. GEOGRAPHICAL VARIATION, GENETIC DIVERSITY AND POPULATION DIFFERENTIATION ACROSS MESOAMERICA

The least square means indicate larger values for all the characters in the populations of the Northern Central America and Mexico in contrast to the Atlantic populations of Costa Rica (Table 10). All the quantitative traits examined were significantly and positively correlated with latitude (Table 11).

Table 10. Least square means of xeric and mesic groups for seedlings traits of *C. odorata* in Mesoamerica. All between group means differ at $p < .0001$ (Scheffe).

Trait	**Acronyms**	**Xeric**	**Mesic**
Height in mm at 62 days	H62	121	99.2
Leaf Width in mm at 62 days	LW62	14.4	10.5
Leaf Length in mm at 62 days	LL62	36.5	30.9
Leaf Length /Leaf Width at 62 days	LL/LW62	2.59	3.04
Height in cm at 252 days	H252	66	13.5
Root collar diameter in cm at 252 days	D252	0.8	0.3
Internodal distance in cm at 252 days	ID252	37.7	13.1
Number of leaflets per leaf at 252 days	NL252	16.9	7.79
Fresh leaves weight in grams	FLWE	103	22.3
Fresh branches weight in grams	FBWE	77	11
Dry leafs weight in grams	DLWE	25.4	5.2
Dry branches weight in grams	DBWE	15.7	1.86

Table 11. Adjusted regression coefficients, models and significances of selected quantitative characters with latitude. Acronyms as in Table 10.

Dependent variable	**Intercept**		**Latitude**		**Adj R-Sq**	**Pr > F (model)**
62 days	Estimate	Pr > |t|	Estimate	Pr > |t|		
H62	75.77707	<.0001	2.92355	<.0001	0.1513	<.0001
LW62	8.01869	<.0001	0.36440	<.0001	0.1730	<.0001
LL62	28.56619	<.0001	0.43355	<.0001	0.0650	<.0001
LL/LW62	3.44218	<.0001	-0.05020	<.0001	0.1315	<.0001
252 days						
H252	-27.1630	<.0001	5.41590	<.0001	0.3067	<.0001
D252	-0.0175	0.7087	0.04782	<.0001	0.2521	<.0001
ID252	-11.9983	<.0001	2.86753	<.0001	0.3435	<.0001
NL252	-4.09159	<.0001	1.20821	<.0001	0.3283	<.0001
FLWE	-23.6055	0.2329	7.04814	<.0001	0.1384	<.0001
DLWE	-5.5791	0.2783	1.70704	<.0001	0.1215	<.0001
FBWE	-34.1309	0.0259	6.27573	<.0001	0.1783	<.0001
DBWE	-7.1251	0.0455	1.27771	<.0001	0.1412	<.0001

4.3.1. Comparison of genetic variability

The correlation between genetic variability in molecular markers and heritability was not significant (r_s = 0.03, n = 14, P = 0.50). The same concerns CV_As and molecular markers. When the test was done on a single trait basis for different groupings of data, none of the 24 (i.e. 8 traits × 3 groupings) possible correlations were significant. Mean heritabilities and CV_As as well as their ranges are given in Table 12.

Table 12. Summary of mean genetic variability parameters for different quantitative traits (h^2 = heritability; CV_A = coefficient of additive genetic variance) and RAPD markers (SDI = Shannon-Weaver diversity estimate) in 14 populations of the Spanish cedar. n_Q = number families/individuals in quantitative genetic analyses. n_M = number of individuals in molecular genetic analyses.

		Mean		**Range**			
Population	n_Q	$h^2 \pm SE$	CV_A	h^2	CV_A	SDI ± SE	n_M
CHA	15/90	0.488 ± 0.63	14.1	0.196 – 1.109	11.0 – 24.1	0.166 ± 0.292	11
CO	7/40	0.373 ± 0.97	14.2	0.069 – 0.716	3.0 – 26.8	0.337 ± 0.279†	8
ES	22/130	0.742 ± 0.52	25.6	0.315 – 1.499	9.4 – 74.5	0.042 ± 0.115	8
LE	20/110	0.731 ± 0.30	28.7	0.181 – 1.431	7.0 – 59.1	0.027 ± 0.077	19
HO	9/54	0.792 ± 0.67	18.8	0.475 – 1.428	13.4 – 35.7	0.313 ± 0.290†	6
GU	5/25	0.578 ± 0.98	17.1	0.349 – 1.878	11.2 – 58.8	0.240 ± 0.292†	5
PAZ	13/61	0.153 ± 0.22	6.3	0.056 – 0.460	10.8 – 15.3	0.011 ± 0.038	15
CA	6/30	1.386 ± 0.81	68.6	0.694 – 1.994	15.7 – 148.6	0.369 ± 0.276†	6
PS	19/114	0.867 ± 0.14	33.8	0.509 – 1.524	13.1 – 84.4	0.175 ± 0.263	12
SC	15/85	0.507 ± 0.22	19.6	0.060 – 1.299	8.5 – 38.5	0.361 ± 0.281†	14
TA	4/24	0.266 ± 0.72	10.6	0.177 – 0.277	8.8 – 58.0	0.296 ± 0.307†	4
UPA	19/111	1.066 ± 0.36	52.7	0.602 – 1.479	15.8 – 108.9	0.277 ± 0.307†	7
XPU	22/132	0.765 ± 0.34	22.4	0.043 – 1.365	2.9 – 60.6	0.071 ± 0.196	13
YU	13/74	0.820 ± 0.59	28.5	0.330 – 1.361	8.3 – 46.8	0.107 ± 0.225	13

†Estimates based on data from Gillies *et al*., 1997.

4.3.2. Comparison of genetic differentiation

The estimates of genetic differentiation (F_{ST}) for molecular markers showed a high degree of population differentiation in both sub-sets of data (sub-set 1: $F_{ST} = 0.670 \pm 0.060$; $P < 0.001$; sub-set 2: $F_{ST} = 0.329 \pm 0.002$; $P < 0.001$). The results are similar if Nei's (Nei 1987) G_{ST} estimator of F_{ST} is used (sub-set 1: $G_{ST} = 0.60$; sub-set 2: $G_{ST} = 0.36$). The wider geographic range of sub-set 1 could account for its higher degree of differentiation. The geographic distance between populations was statistically different for both subsets of data using the Mann-Whitney test ($z = 4.67$, $n = 42$, $P < 0.001$).

Approximately 68 % of the variation for markers is among populations indicating a high subdivision within the species. The degree of quantitative trait differentiation, even though considerable, was much lower than that observed in molecular markers (Table 13).

Table 13. Nested analyses of variance of quantitative traits and RAPD markers, together with associated Q_{ST} and F_{ST} estimates for different subdivisions of data. Q_{ST} estimates are given under assumption of half-sib (HS) and full-sib (FS) structure of the data. POP = population, FAM = family (nested within population), ERR = error variance components, respectively. Greenhouse and laboratory studies, Helsinki, Finland. For trait acronyms see Table 10.

Trait	**POP**	**FAM**	**ERR**	**$Q_{ST(HS)}$ ± SE**	**$Q_{ST(FS)}$ ± SE**
All populations					
H62	410.24**	130.06***	429.63	0.283 ± 0.014	0.441 ± 0.034
LW62	7.182**	1.95***	5.88	0.315 ± 0.017	0.479 ± 0.039
LL62	22.549**	5.56***	26.37	0.336 ± 0.019	0.503 ± 0.043
LL/WL62	0.090**	0.05***	0.23	0.172 ± 0.006	0.293 ± 0.017
HC252	711.66**	205.29***	319.62	0.302 ± 0.015	0.464 ± 0.037
D252	0.08**	0.02***	0.05	0.324 ± 0.014	0.489 ± 0.033
ID252	246.14**	26.47**	115.13	0.538 ± 0.048	0.699 ± 0.081
NL252	44.46**	6.48**	32.55	0.462 ± 0.037	0.632 ± 0.069
Mean Q_{ST}				0.341 ± 0.021	0.500 ± 0.044
Subset 1					
H62	100.06*	119.40***	485.50	0.230 ± 0.004	0.173 ± 0.012
LW62	2.71*	1.83***	5.962	0.260 ± 0.009	0.270 ± 0.027
LL62	5.78*	5.39***	24.75	0.260 ± 0.006	0.211 ± 0.018
LL/WL62	0.04*	0.02***	0.158	0.150 ± 0.000	0.282 ± 0.001
HC252	737.04*	219.01***	463.720	0.230 ± 0.013	0.457 ± 0.045
D252	0.07*	0.02**	0.060	0.270 ± 0.008	0.490 ± 0.030
ID252	226.69*	15.25	141.230	0.430 ± 0.014	0.788 ± 0.053
NL252	43.24*	1.27	33.837	0.460 ± 0.009	0.895 ± 0.035
Mean Q_{ST}				0.286 ± 0.008	0.446 ± 0.028
F_{ST}				0.667 ± 0.064	
Subset 2					
H62	422.09*	152.33***	321.080	0.257 ± 0.026	0.409 ± 0.065
LW62	9.34*	2.18***	5.729	0.348 ± 0.044	0.517 ± 0.097
LL62	36.37*	5.89**	29.521	0.436 ± 0.063	0.607 ± 0.123
LL/WL62	0.08*	0.12***	0.376	0.083 ± 0.004	0.153 ± 0.013
HC252	635.71*	162.38***	88.442	0.329 ± 0.039	0.495 ± 0.089
D252	0.08*	0.02***	0.024	0.320 ± 0.035	0.485 ± 0.080
ID252	252.55*	43.95**	72.621	0.418 ± 0.063	0.590 ± 0.126
NL252	43.99	15.60**	30.453	0.261 ± 0.028	0.413 ± 0.069
Mean Q_{ST}				0.306 ± 0.038	0.459 ± 0.083
F_{ST}				0.325 ± 0.093	

*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$

The average Q_{ST} for all populations was 0.34 ± 0.02, ranging from 0.17 to 0.54 for individual traits (Table 13). The Q_{ST} value for sub-set 1 was 0.29 ± 0.01 and for sub-set 2 was 0.31 ± 0.04. These estimates are significantly lower than (subset 1) or similar to (subset 2) the corresponding F_{ST} estimates for molecular markers (t-tests; Sub-set 1: $t_{21} = 19.34$, $P < 0.001$; sub-set 2: $t_{16} = 0.63$, $P = 0.53$). If a full-sib family structure is assumed (Table 13), the Q_{ST} estimate is lower than for subset 1 ($t_{21} = 6.66$, $P < 0.0001$), but Q_{ST} and F_{ST} estimates for subset 2 are not statistically different ($t_{16} = 1.61$, $P = 0.15$).

The pair-wise Q_{ST} estimates were strongly and positively correlated with equivalent F_{ST} estimates in both subsets of data (Mantel's tests; sub-set 1: $r = 0.69$, $P < 0.001$; sub-set 2: $r = 0.55$, $P = 0.020$; Fig. 5 a and b). This is true also when data from the two sub-sets are pooled (Mantel's test: $r = 0.93$, $P < 0.001$; Fig. 5c).

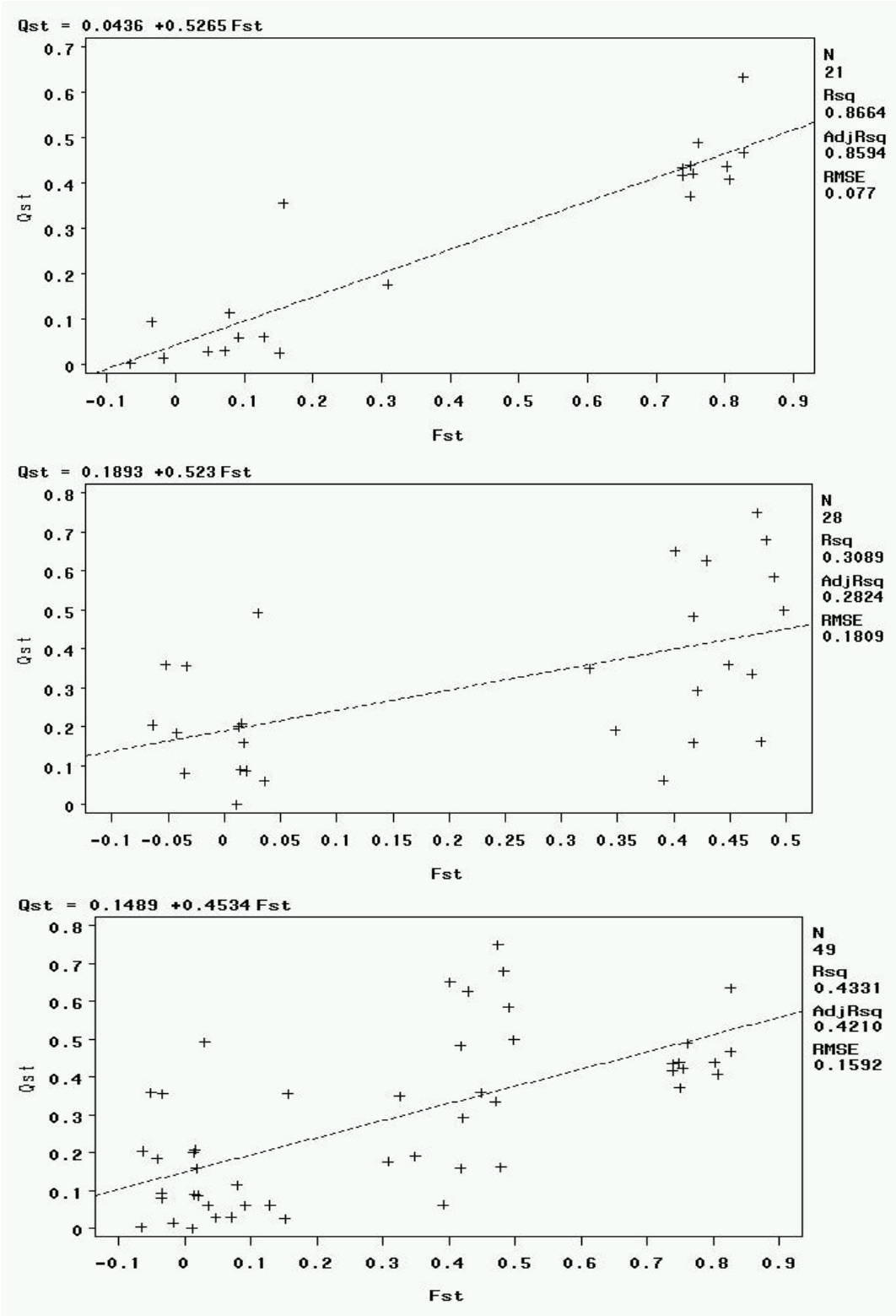


Fig. 5. Comparison of pair-wise Q_{ST} and F_{ST} estimates across the study populations for (a) sub-set 1, (b) sub-set 2 and (c) and combined data. The solid line marks the 1:1 expectation for the correspondence between Q_{ST} and F_{ST} estimates, and the dotted lines are least square regression lines given for ease of interpretation. See text for statistical tests.

4.4. AGROFORESTRY RESULTS FROM *C. ODORATA*-COFFEE MIXTURES.

4.4.1. Performance of the provenances

There was a significant provenance effect for all variables (Pr > F<.0001) (Table 14). The best performing provenances for diameter were PZ (3.6 cm/year), PS (3.5 cm/year), TAL, SC and GU, all from the wettest region of Costa Rica. PZ (193 cm/year) and GU (166 cm/year) were significantly superior to the others in rate of growth.

Table 14. Summary of analysis of variance (GLM) of diameter, height, number of attacks of *H. grandella* and number of shoots of *C. odorata* at 25 months after planting at Turrialba, Costa Rica.

Variables	**DF**	**F Value**	**p > F**	**DF**	**F Value**	**p > F**
	Diameter			**Height**		
Block	16	29.08	<.0001	16	14.47	<.0001
Population	20	31.74	<.0001	20	42.63	<.0001
Progeny(population)	95	2.38	<.0001	95	3.26	<.0001
Block*population	316	1.50	<.0001	316	1.53	<.0001
Error	2627			2627		
	Susceptibility			**Recovery**		
Block	16	7.25	<.0001	16	9.61	<.0001
Population	20	3.44	<.0001	20	7.76	<.0001
Progeny(population)	93	1.30	0.0295	93	1.58	0.0004
Block*population	316	1.02	0.4063	316	1.06	0.2235
Error	2501			2501		

Provenances from dry areas: CA, CO, GUA and TFC were most resistant to the shootborer (Table 15), suffering only one attack on average, but did differ significantly from the best progenies in diameter and height.

Table 15. Population means and standard errors (SE) of *C. odorata* for diameter (mm), height (cm), number of attacks by *H. grandella* (susceptibility) and number of new shoots re-sprouted after attack (recovery), (ranking in parenthesis).

Populations	**Diameter**	**SE**	**Height**	**SE**	**Susceptibility**	**SE**	**Recovery**	**SE**
CA	42.27 (21)	2.79	184.00 (17)	18.91	1.07 (1)	0.13	16.23 (9)	0.35
CO	47.78 (18)	1.95	176.74 (18)	13.21	1.14 (3)	0.14	16.49 (12)	0.37
GU	67.79 (5)	2.27	348.74 (2)	15.36	1.63 (15)	0.17	16.29 (10)	0.44
HO	47.05 (20)	1.34	175.30 (20)	9.12	1.40 (8)	0.10	16.59 (13)	0.26
PS	73.66 (2)	2.27	341.38 (3)	15.40	1.37 (6)	0.23	15.50 (2)	0.59
PZ	74.67 (1)	1.85	398.58 (1)	12.52	1.69 (17)	0.14	16.22 (8)	0.36
SC	69.23 (4)	3.90	336.48 (4)	26.39	1.64 (16)	0.29	17.27 (18)	0.75
TAL	70.08 (3)	1.86	326.02 (6)	12.63	1.79 (20)	0.14	16.32 (11)	0.37
UPA	64.39 (7)	2.25	328.08 (5)	15.22	1.47 (10)	0.17	15.84 (4)	0.43
LE	53.14 (13)	1.02	174.34 (21)	6.91	1.54 (12)	0.08	16.65 (14)	0.20
TI	59.41 (10)	0.97	285.57 (9)	6.60	1.60 (13)	0.07	17.46 (19)	0.19
CE	52.21 (14)	1.08	241.91 (12)	7.36	1.72 (18)	0.08	18.27 (21)	0.21
PAZ	51.44 (15)	1.18	199.27 (16)	8.01	1.60 (14)	0.09	16.67 (15)	0.23
MEA	64.28 (8)	1.65	277.52 (10)	11.19	1.80 (21)	0.13	18.09 (20)	0.33
TAU	54.39 (12)	1.28	240.23 (13)	8.68	1.72 (19)	0.10	16.83 (16)	0.25
BA	48.33 (17)	1.58	218.05 (14)	10.70	1.22 (5)	0.12	15.92 (5)	0.31
TG	49.63 (16)	1.97	217.16 (15)	13.33	1.38 (7)	0.15	15.95 (6)	0.39
TFC	47.09 (19)	1.15	175.39 (19)	7.78	1.20 (4)	0.09	15.55 (3)	0.23
XPU	56.01 (11)	1.87	252.47 (11)	12.71	1.49 (11)	0.15	17.08 (17)	0.38
AL	65.62 (6)	0.83	318.93 (7)	5.61	1.45 (9)	0.06	16.03 (7)	0.16
GUA	63.71 (9)	1.44	295.36 (8)	9.79	1.13 (2)	0.11	15.39 (1)	0.28

4.4.2. Effects of coffee on growth and resistance of C. odorata

The size of accompanying coffee trees and cultivation method had highly significant effects on the variables evaluated. The coffee in production and trees planted between coffee rows (BCR) (Blocks 6, 7 and 8, Tables 4 and 16) provided the best conditions for the growth of *C. odorata.*

Table 16. Block least square means for diameter (mm), height (cm), number of attacks by *Hypsipyla grandella* (susceptibility) and number of shoots re-sprouting after attack (inverse of recovery); (ranking in parenthesis).

Block	**Diameter**	**SE**	**Height**	**SE**	**Attacks**	**SE**	**Shoots**	**SE**
1	63.55 (5)	1.66	304.87(3)	11.22	1.52 (11)	0.12	18.47 (17)	0.32
2	65.18 (3)	1.29	248.46 (11)	8.71	2.44 (17)	0.10	18.34 (16)	0.26
3	60.63 (8)	1.30	261.46 (7)	8.79	1.67 (16)	0.10	18.14 (15)	0.27
4	63.52 (6)	1.64	251.46 (8)	11.12	1.77 (16)	0.12	16.33 (9)	0.32
5	62.29 (7)	1.21	240.25 (14)	8.19	1.54 (13)	0.09	16.06 (8)	0.23
6	70.78 (1)	1.23	300.01 (4)	8.32	1.50 (10)	0.09	17.90 (14)	0.24
7	70.05 (2)	1.22	338.52 (1)	8.26	1.05 (1)	0.09	16.62 (13)	0.24
8	65.08 (4)	1.23	307.22 (2)	8.34	1.16 (2)	0.09	16.33 (10)	0.24
9	55.90 (10)	1.20	292.17 (5)	8.10	1.52 (12)	0.09	16.47 (12)	0.24
10	51.74 (14)	1.67	238.28 (15)	11.28	1.29 (5)	0.13	15.18 (1)	0.33
11	53.53 (11)	1.20	250.83 (9)	8.10	1.50 (9)	0.09	16.01 (7)	0.23
12	50.38 (15)	1.18	244.20 (12)	7.97	1.32 (6)	0.09	15.50 (4)	0.23
13	42.62 (16)	1.25	186.43 (17)	8.42	1.35 (7)	0.09	15.45 (2)	0.24
14	51.69 (15)	1.20	250.12 (10)	8.09	1.54 (14)	0.09	16.43 (11)	0.24
15	53.33 (12)	1.22	241.23 (13)	8.24	1.27 (4)	0.09	15.93 (5)	0.23
16	52.15 (13)	1.21	227.96 (16)	8.19	1.24 (3)	0.09	15.47 (3)	0.24
17	57.04 (9)	1.67	278.25 (6)	11.29	1.47 (8)	0.13	16.00 (6)	0.33

C. odorata interplanted with coffee in production was less susceptible (1 and 1.2) which means fewer attacks by the shootborer. Young coffee associated with *C. odorata* resulted in more shoots regenerating which is considered harmful for the rearing of monocormic trees. *C. odorata* planted within the coffee rows and coffee in production showed the lowest production of new shoots, i.e., the highest degree of recovery. Trees cultivated with young coffee were most frequently attacked by the shootborer (2.4 attacks).

4.4.3. Progeny analysis, heritabilities and additive genetic variance

The families studied showed highly significant differences for all the variables studied (Table 17, Fig. 6). For example, the top progenies 6232 (PZ), 6240(PZ), 6177(UPA) and 745 (GUA) grew 207, 205, 188 and 186 cm/year in height, respectively; the two slowest (168 and 171, both from TFC) grew 72 and 73 cm/year in height, respectively.

Table 17. Summary of analysis of variance (GLM) of diameter, height, and number of shoots re-sprouting (recovery) after shootborer attack in progenies of *C. odorata* and susceptibility to *H. grandella,* 25 months after planting in Turrialba, Costa Rica.

Source	**DF**	**F Value**	**Pr > F**
Diameter			
Block	16	48.68	<.0001
Progeny	115	9.94	<.0001
Block*progeny	1744	1.59	<.0001
Error	1199		
Height			
Block	16	23.65	<.0001
Progeny	115	12.90	<.0001
Block*progeny	1744	1.58	<.0001
Error	1199		
Recovery			
Block	16	17.77	<.0001
Progeny	113	3.18	<.0001
Block*progeny	1694	1.31	<.0001
Error	1123		
Susceptibility			
Block	16	12.65	<.0001
Progeny	113	2.16	<.0001
block*progeny	1694	1.40	<.0001
Error	1123		

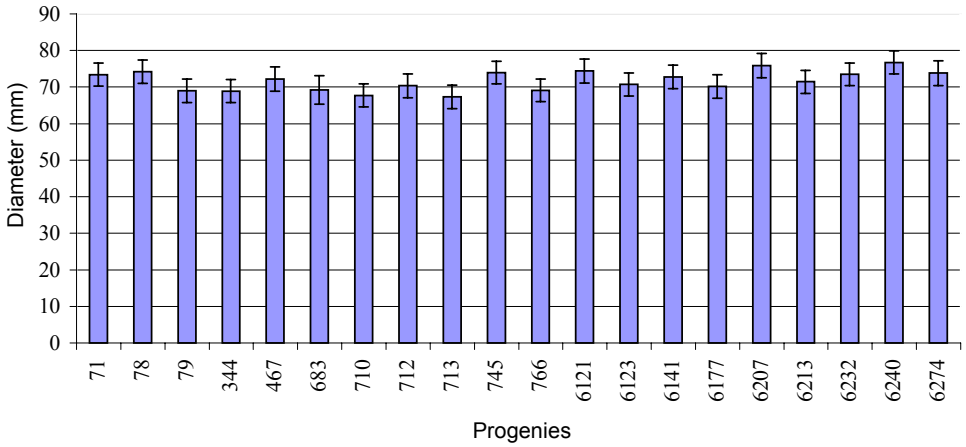
Remarkable distinctions in resistance between progenies during all the evaluation period were observed. Figure 6 shows the less susceptible progenies.

The additive genetic variance for all progenies within provenances (excluding the variance component of provenances) for diameter and height are 12 and 20 % of the total variance.

The heritabilities for growth for all progenies within provenances were 0.122 ± 0.001 and 0.202 ± 0.002 for diameter and height, respectively. The coefficients of additive genetic variance (CV_A; Houle 1992) were 10.7 and 21.4 for diameter and height.

I also made the analysis for only the local best performing provenances for which additive genetic variance represented 15 and 21% of the total variance. Estimates of heritabilities were 0.154 ± 0.006 for diameter and 0.212 ± 0.010 for height. The CV_A values were 12.4 and 21.9.

In this study the heritability estimates are based on a single location field trial and the genetic kinship of the families may include both half and full sibs and indeed selfings. Thus, heritability estimates may be less accurate than the coefficients of additive genetic variance for deciding on how to use genetic variation for efficient conservation and breeding.


Diameter (mm)
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80
70
60
50
40
30
20
10
0
71
78
79
344
467
683
710
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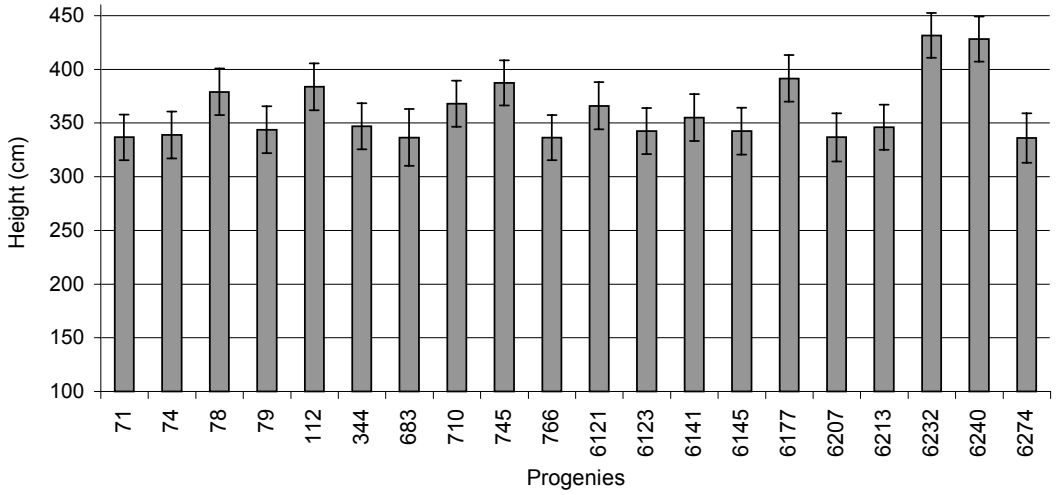
6121
6123
6141
6177
6207
6213
6232
6240
6274
Progenies


Height (cm)
450
400
350
300
250
200
150
100
71
74
78
79
112
344
683
710
745
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6123
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Progenies

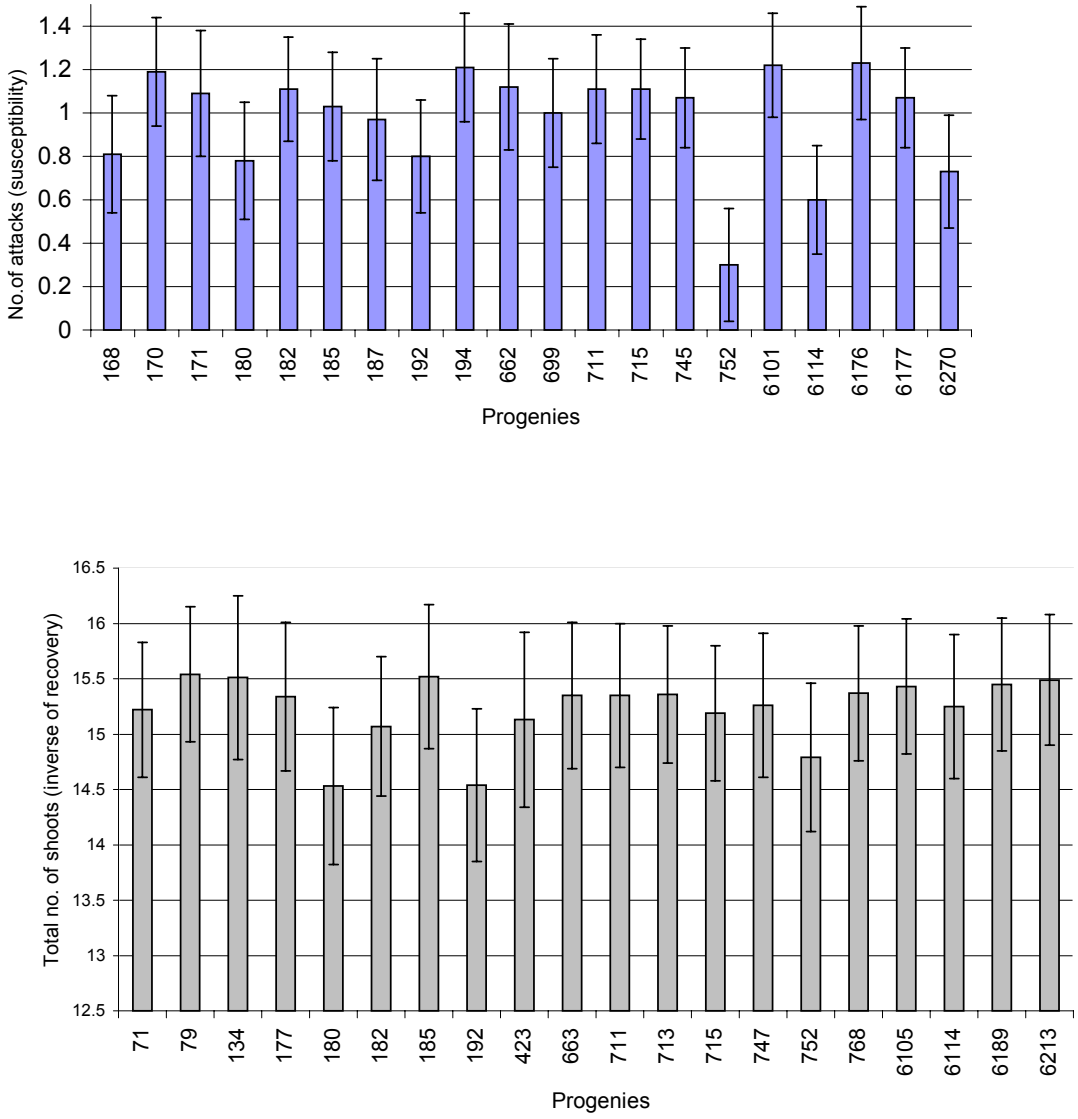


Fig. 6. The best 20 progenies of *C. odorata* for four traits: Diameter (mm), height (cm), susceptibility and recovery.

4.5. REPRODUCTIVE ISOLATION

Results gathered from the genetic resources collection of *C. odorata* indicate that the trees were present mostly in fragmented areas throughout the Mesoamerican range (Table 6). In most cases, the trees were found on man-made grasslands previously covered by tropical mixed forests. The collected specimens were always located naturally on well-drained soils, contrasting with other species of the same family e.g. *Carapa guianensis* or *Swietenia macrophylla* (mahogany) which grow on sites with heavy or waterlogged soils (Navarro *et al.* 2002b). The remaining trees associated with *C. odorata* are listed with the population characteristics (Table 6).

4.5.1. Analysis of variable isolation

Height showed highly significant differences depending on isolation levels (see page 34 for definitions) for both xeric (for all measurements F ≥59.09, $P \leq .0001$) and mesic progenies (for all measurements F ≥7.25, $P \leq 0.0008$)(Table 18). The progenies of non-isolated mother trees showed superior growth compared with those of isolated mother trees.

The effect of the degree of isolation upon susceptibility was highly significant for xeric progenies (F ≥13.65 $P < .0001$) and significant for mesic progenies (F ≥5.39 P≤ 0.0047).

Table 18. Analysis of variance for height and resistance of *C. odorata* progenies from three levels of isolation of mother trees in xeric and mesic habitats.

Source of variance	**DF**	**Mean square**	**F Value**	**Pr > F**
Height xeric habitat				
Block	16	147413.64	13.63	<.0001
Mother-tree	2	638840.25	59.09	.0001
Error	2138			
Height mesic habitat				
Block	16	184214.58	7.77	<.0001
Mother-tree	2	171873.52	7.25	.0008
Error	899			
Resistance xeric habitat				
Block	16	12.20	7.33	<.0001
Mother-tree	2	22.71	13.65	<.0001
Error	2058			
Resistance Mesic habitat				
Block	16	6.00	4.24	<.0001
Mother-tree	2	7.63	5.39	0.0047
Error	869			

The orthogonal contrasts of mother trees revealed highly significant differences (Table 19). The difference between xeric mother trees in isolation 1 and 2 was not great but both were significantly different from isolation 3. For mesic trees, isolation 1 was significantly different from isolations 2 or 3.

Table 19. Isolation of mother trees. Orthogonal contrasts for height and susceptibility to *H. grandella*.

Contrast	DF	Contrast SS	Mean Square	F Value	Pr > F
Height	Xeric				
1 vs 2 and 3	1	63910.140	63910.140	5.91	0.0151
2 vs 3	1	605585.542	605585.542	56.01	<.0001
3 vs 1and 2	1	1083320.507	1083320.507	100.20	<.0001
Susceptibility	Xeric				
1 vs 2 and 3	1	16.043	16.043	9.64	0.0019
2 vs 3	1	45.341	45.341	27.25	<.0001
3 vs 1 and 2	1	36.595	36.595	21.99	<.0001
Height	Mesic				
Contrast					
1 vs 2 and 3	1	332477.745	332477.745	14.02	0.0002
2 vs 3	1	51199.400	51199.400	2.16	0.1421
3 vs 1 and 2	1	49397.410	49397.410	2.08	0.1493
Susceptibility	Mesic				
1 vs 2 and 3	1	9.904	9.904	7.00	0.0083
2 vs 3	1	8.402	8.402	5.93	0.0150
3 vs 1 and 2	1	0.039	0.039	0.03	0.8675

The mother tree progenies in isolation 1 grew less in height than the progenies in isolations 2 or 3 (Pr > F <0.0001) (Table 20).

The difference in height of the progeny of the isolated mother trees and those of non-isolated mother trees tends to increase with age of the progenies. Differences in the first measurement in the nursery were minimal, but after planting in the field the differences became appreciable for both xeric and mesic progenies (Figure 7 and 8, respectively).

Table 20. Effect of seed tree isolation on height and resistance of *C. odorata* progenies 25 months after planting at CATIE, Turrialba, Costa Rica. Tukey G (Tukey grouping) and N, number of plants measured.

Xeric	**Height**			**Mesic**	**Height**		
Mother	Mean (SE)	Tukey G	N	Mother	Mean (SE)	Tukey G	N
3	247.98 (3.64)	A	842	2	348.29 (11.35)	A	188
1	201.84 (3.12)	B	1150	3	330.62 (6.35)	A	622
2	179.89 (8.12)	C	165	1	277.96 (14.88)	B	108
	Susceptibility				**Susceptibility**		
3	1.58 (0.04)	A	822	2	1.69 (0.08)	A	188
1	1.48 (0.04)	A	1097	3	1.44 (0.05)	B A	578
2	1.01 (0.10)	B	158	1	1.23 (0.11)	B	104

A comparison of means also showed significant differences in rates of growth of the isolated mother trees (Table 20). Differences in height were 37 % for xeric progenies and 25 % for mesic progenies at 25 months. The mother tree progenies in isolation 3 showed superior height growth but were more heavily attacked (more susceptible). Due to their good growth, they showed a high recovery rate, however.

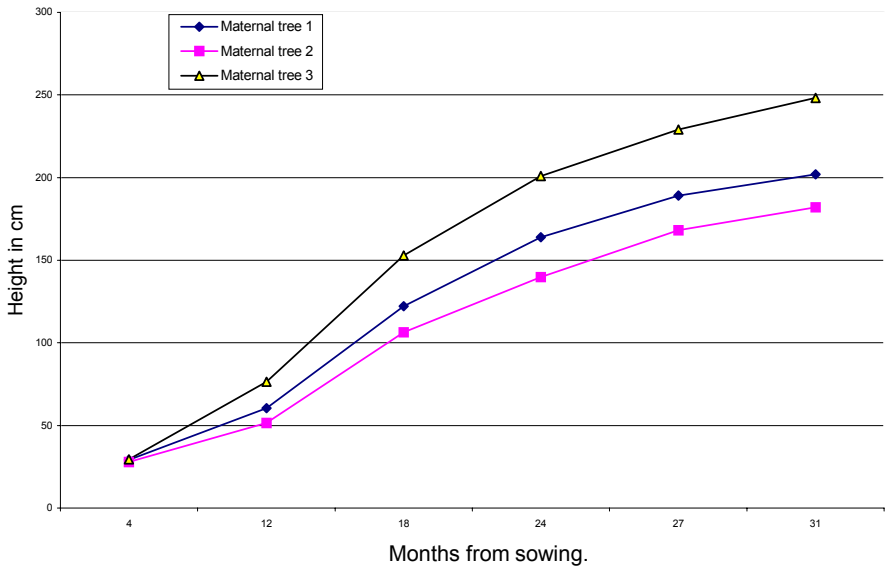


Figure 7. Development of height growth of *C. odorata* in variously isolated trees in xeric conditions.

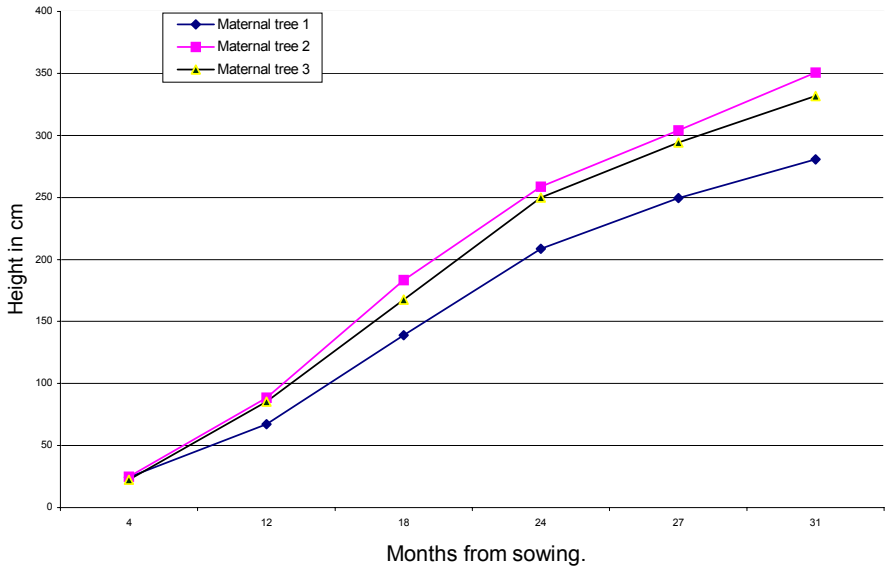


Figure 8. Development of height growth with age of *C. odorata* between variously isolated trees in mesic conditions.

5. DISCUSSION

5.1. STATUS OF CONSERVATION

The survey in the six countries visited indicates that remaining populations are very small in terms of number of individuals and even smaller in effective population size (Table 1 and Figure 1). Since mature individuals that will never produce new recruits should not be counted (e.g. densities are too low for fertilisation or no possibilities for the seeds to become new trees), the conservation status of *C. odorata* can be considered as endangered (EN A1cd) in cases where populations have been eroded as follows:

> An observed, estimated, inferred or suspected reduction of at least 70% over the last 10 years or three generations, whichever is the longer, caused by any of the following: a decline in area of occupancy, extent or occurrence and/or quality of habitat and actual or potential levels of exploitation. This classification is based on IUCN Red List Categories (IUCN 2001).

This classification considers all individuals present in home orchards or farms as natural. If we exclude these individuals, the Red List Category and Criteria will be Critically Endangered (CR A1cd). For more details about categorisation, see Red List Categories and Criteria (IUCN 2001).

C. odorata presents different characteristics related to conservation in comparison with *S. macrophylla* (mahogany). For example, mahogany is naturally present in some protected areas but *C. odorata* - because of its long exploitation - has been exterminated in most of the areas. Loss of the natural habitat of *S. macrophylla* is around 80 % (Navarro *et al*. 2002) but

natural *C. odorata* has been exterminated across its range and in most cases survives mainly as a "domesticated" species in home orchards, grasslands and other man-made habitats. The most important area for conservation of *C. odorata* in Mesoamerica is the Tikal National Park in Guatemala (personal observation). We must look for other areas for conservation within the range of distribution. In order to conserve the genetic resources of this species, local communities can help conserve natural stands or on their farms (*circa-situ*). Protected areas as national parks or *ex-situ* gene banks or seed orchards are necessary but not sufficient. The small numbers of lasting areas of uninterrupted forests are highly dispersed and correspond to only a minute proportion of the total area.

The situation in South America is similar to the one I am presenting, i.e. some conservation measures have been taken. For instance, Colombia' s populations of *C. odorata* have been added to CITES Appendix III. Trade in Appendix III species and their parts and derivatives is permitted, but requires CITES documents. In the case of Mesoamerican *C. odorata* and Bigleaf mahogany (*S. macrophylla* K.), regulations apply only to logs, sawn wood and veneer sheets. Foreign exporters must present either a CITES export permit or a CITES certificate of origin. Peru's populations of *C. odorata* were also recently added to CITES Appendix III. These listings are already in effect. (Canadian Government Publishing 2001).

5.2. DROUGHT ADAPTATION

The climatic groupings of xeric and mesic populations in the analysis of variance were highly significant and the cluster analysis showing a remarkably clear bifurcation into two distinct groups (Fig. 3 and 4). Studies on other species have demonstrated a relationship between characteristics of the seedlings and environmental parameters but have not shown such clear

phenotypic segregation into a few groups (Ladiges *et al.*1981, Sorenson 1983, Toval and Puerto 1985, Loopstra and Adams 1989, Sorenson *et al.* 1990, Kundu and Tigerstedt 1997). Evidently dry (xeric) and wet (mesic) environments have caused considerable selection pressures and diversification in *C. odorata*.

In a study (Navarro and Vásquez 1987) based on characteristics of seeds and seedlings characteristics from the Pacific and Atlantic coast of Costa Rica and Nicaragua, *C. odorata* also displayed strong differentiation between populations from dry and wet zones. Significant differences between dry and moist provenances in the length, width and surface area of the seeds, and in the height, root collar diameter, and root length of seedlings were obtained. Alvarez (1999) compared seeds and fruits of *C. odorata* trees from Las Juntas (Pacific) and Jimenez (Atlantic). The trees from Jimenez had smaller fruits and more seeds per fruit than trees from Las Juntas.

Larger seed weights in tree populations from dry areas have been observed elsewhere (Sorenson 1983, Toval and Puerto 1985, Loopstra and Adams 1989, Sorenson *et al.* 1990, Wright *et al.* 1992). It appears that high seed weights can affect early seedling growth due to larger seed storage resources. However, results point at diametrically opposite seedling growth effects, evidently due to poorly available water resources (Roman 1996, Li 1998). In the *Abies procera/A. magnifica* species complex, Sorenson *et al*. (1990) found that seedlings from southern, drier areas were larger in the first year, but that their growth rate slowed down considerably in subsequent growing seasons. Initial faster growth was associated with larger seed size and longer growing seasons. Wright *et al.* (1992) also found that fast early seedling growth in populations of *Pinus banksiana* from dry areas was mostly attributable to their larger seed size in contrast to smaller seeds from adjacent wet areas.

In the present study, differences in growth rates between provenances may be attributable to adaptation for survival in dry and wet environments. The lower rainfall and long dry season in the North Pacific part of Costa Rica could have selected the individuals with rapid growth at the seedling stage. Larger leaves allow for more photosynthesis, potentially faster root and stem growth during a short wet period, which would facilitate plant survival during the dry season. However, these patterns may change at a later phase of the life cycle. In *C. odorata* provenance/species trials in St. Croix (USVI), Puerto Rico, Uganda and Tanzania, from the nursery stage up to 14 months, the provenance from Guanacaste, Costa Rica (dry zone) was usually superior in growth to other sources (Whitmore 1971, Karani 1973, Rauno 1973). After two years, other sources overtook the Guanacaste provenance for height (Karani 1973, Rauno 1973, Whitmore 1978).

The genetic variability in this study is ecotypic rather than clinal. This indicates limited gene flow between the two groups (xeric and mesic) of populations that have become differentiated (Ridley 1990).

The climatic differences could have produced separation in flowering time that would reduce gene flow between climatic regions. There are several possible explanations for the observed patterns of variation. Differentiation may have occurred when the Cordilleras Volcanica Central and Guanacaste raised a barrier within the range of the species in Costa Rica, thereby reducing gene flow, whereupon the populations in the two zones began to evolve independently. Alternatively, these two groups could have spread from different refuges after the Pleistocene glaciations.

The Cobano population is similar in seedling traits to other dry Pacific provenances, but is located at the tip of the Nicoya Peninsula in an area of higher rainfall. The area may have been colonized by, or maintained in a common gene pool with trees from the dry Pacific regions, because seed movement or gene flow from other mesic areas was prevented by the central mountains (Volcanica Central and Guanacaste). The Upala population is similar to mesic Atlantic populations but is in an area intermediate in rainfall between the Pacific and the east Atlantic. It may have been colonised by, or formed a common gene pool with Atlantic populations if movement from the Pacific were blocked by the Cordillera. Although it is found on the south Pacific coast of Costa Rica, the Zona Sur population has affinities with the Atlantic mesic populations. A RAPDs study of *C. odorata* indicated similarity between Zona Sur and a population from northern Panama (C. Navarro, unpublished). Central Panama may serve as a pathway for gene flow between the Atlantic and southern Pacific populations, or have been a refuge for these populations during glacial periods (Colinvaux 1996, p. 397).

The differentiation could also be the result of adaptations to the contrasting moisture regimes encountered by the two sets of populations. These climatic differences could have resulted in separation in flowering time that would reduce gene flow between climatic regions. In that case, the RAPDs markers identified by Gillies *et al*. (1997) must be either linked to loci subjected to selection, or are under selection themselves, rather than neutral markers.

The amount of differentiation among populations within a species can range from almost none to levels of distinctness usually attributed to different species (Mayr 1963 cited by Ridley 1990). The differentiation seen in these populations in RAPDs markers is greater than that seen in some closely related species (Gillies *et al.* 1997), and suggests that *C. odorata* may be in the process of forming new species in Costa Rica. The distinction between the two groups of populations investigated is presumably maintained by a pre- or post-reproductive

isolating mechanism (Ridley 1990), which deserves further investigation. The potential cline between the Upala and Liberia populations should also be investigated as evidence for incipient speciation.

The distribution of genetic variation between (80 %) and within populations (20 %) in the present study parallels earlier findings using RAPDs molecular markers for these same populations (Gillies *et al*. 1997), even though the adaptive traits are different from gene markers. RAPDs are presumably neutral markers resulting from dominant alleles; while quantitative traits are multigenic and more influenced by selection. With RAPDs, 35 % of the genetic variance occurred between the wet and dry zones, none among populations within zones, and 65 % within populations. Based on the Shannon Diversity Index, 55 % of the variation occurred among populations (including between the two zones) and 45 % of the genetic variation occurred within populations. The current studies are in contrast with the general observation that the vast majority of genetic variation is within populations (80 – 90%) in woody and perennial outbreeding species (Hamrick and Godt 1990). *C. odorata* is a wide-ranging species and overall genetic variability is high compared to species with narrower geographic or ecological distributions (Hamrick and Godt 1990).

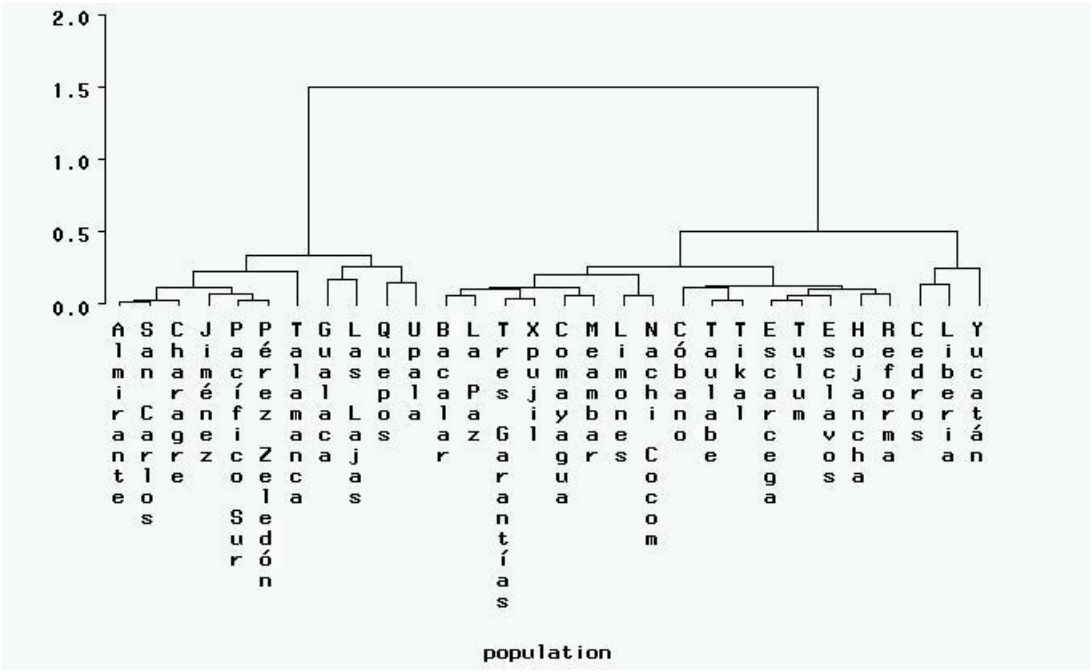
5.3. GEOGRAPHICAL VARIATION, GENETIC DIVERSITY AND POPULATION DIFFERENTIATION ACROSS MESOAMERICA

There is a clear differentiation into two main groups or ecotypes; the northern group (Mexico, Guatemala, Honduras, Nicaragua and Pacific coast of Costa Rica) and the Atlantic coast and South Pacific group of Costa Rica and Panama (Fig. 9). As to quantitative traits, these results are concordant with the earlier reports of a relatively high degree of differentiation within and among Costa Rican and Nicaraguan populations of this species (Navarro and Vasquez 1987, Navarro *et al.* 2002).

The much wider geographical range covered by the current study shows that these earlier studies capture only a limited proportion of the diversity in this widespread species. Likewise, our analyses of RAPD differentiation concurred with results of Gillies *et al*. (1997), with the difference that increased coverage of the geographical sampling revealed a higher degree of intraspecific differentiation. In fact, the results presented here suggest the existence of two distinct forms of *C. odorata* that are well separated both in terms of neutral marker genes and genes coding for quantitative traits.

This dichotomy is made more apparent if we subject the quantitative and molecular genetic data to cluster analyses: two clear clusters of populations corresponding to populations inhabiting xeric and mesic environments are revealed. Hence, as to the quantitative traits, there may exist at least two well-differentiated forms of *C. odorata*, each of which may be locally adapted to live in contrasting environmental conditions (see also Graham 1999). The results also hint at possible incipient speciation and/or subspecies status of different *C. odorata* populations living in contrasting environmental conditions.

a



b

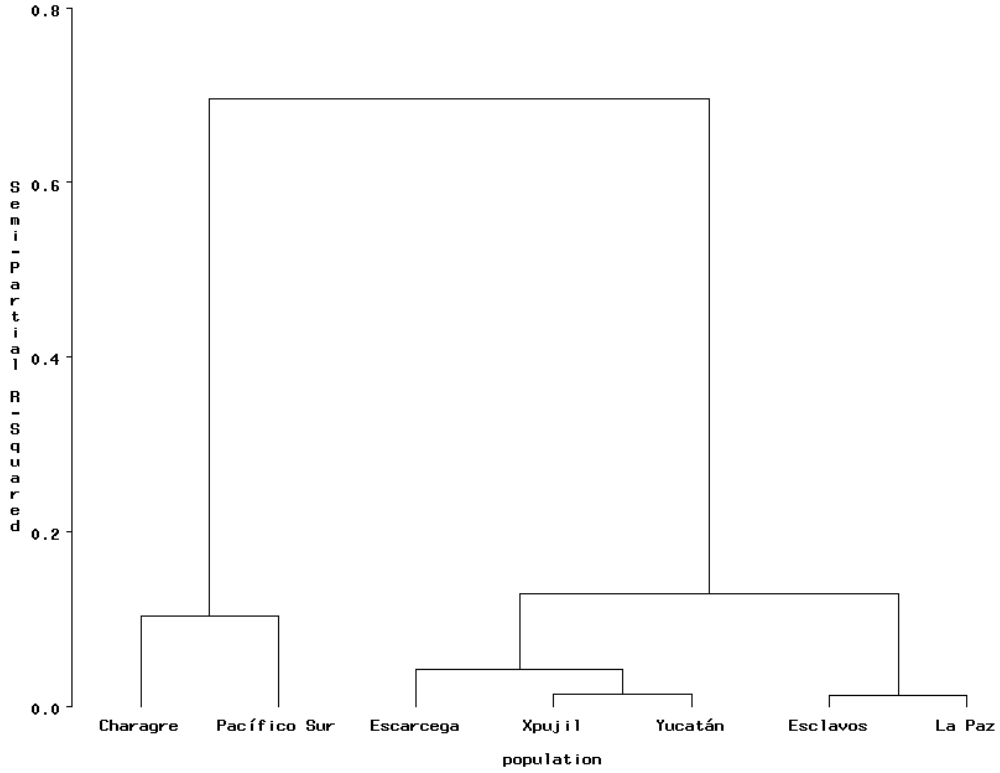


Fig. 9. Cluster analyses of (a) quantitative traits and (b) RAPD data for the seven *C. odorata* populations in the sub-set 1 (see methods). The two clusters in both data sets correspond to mesic and xeric environments. The morphological distances are based on Euclidean distances and the genetic distances on Nei's (1987) genetic distance.

The most outstanding findings of the present study were that while the degree of differentiation in the genes coding quantitative traits was much less than that observed in neutral markers, these two measures of genetic differentiation were strongly positively correlated across pairs of populations. Conversely, the levels of intra-population diversity in molecular markers and quantitative traits were uncorrelated across the populations. The implication of these results is that there may be a need to maintain *in-situ* conservation areas, as well as *ex-situ* and *circa-situ* gene banks and plantations, not only of one, but of several forms of the endangered *C. odorata*. Naturally, priority should be given to the areas where the species is most endangered.

5.3.1. Selection, drift or stabilising selection

A frequent pattern in studies which have compared F_{ST} and Q_{ST} values, is that the degree of differentiation for quantitative traits typically surpasses that for molecular markers, i.e. $Q_{ST} > F_{ST}$ (reviewed in: Merilä and Crnokrak 2001, McKay and Latta 2002). This implies that quantitative traits are typically under directional selection imposed by environmental factor(s) that favours different mean trait values in different populations. Under the assumption of neutrality, F_{ST} and Q_{ST} values are expected to be equal (e.g. Whitlock 1999). If $Q_{ST} < F_{ST}$, this suggests that the quantitative trait divergence among populations is less than expected if a balance was determined between genetic drift and migration only. However, none of the studies published so far have reported a situation where this would have been the case (Merilä and Crnokrak 2001). Hence, this study is the first one to report a situation where the quantitative trait differentiation is less than that observed for presumably neutral molecular markers (i.e. $Q_{ST} < F_{ST}$), and suggests that the quantitative traits in different populations of *C. odorata* have been under some form of stabilising selection. This is somewhat surprising given the fact that the populations were shown to be strongly differentiated in mean values of all traits, and that the magnitude of differentiation in quantitative traits (mean $Q_{ST} = 0.34$) was

comparable to that observed in other studies (mean Q_{ST} of 18 published studies = 0.37, Merilä and Crnokrak 2001). However, one possible explanation for this interesting result is that the opportunity for differentiation due to drift in this species is very high. Hence, isolation into small local populations is a likely explanation for the high degree of differentiation for neutral loci, but in the case of quantitative traits, this differentiation can be counteracted by selection.

Another explanation for the observation that $Q_{ST} < F_{ST}$ for all traits observed is that we have underestimated the degree of differentiation for quantitative traits. There are two obvious reasons why such could be the case. First, our estimates of additive genetic variance are likely to include maternal and dominance effects, which will lead to underestimates of Q_{ST} (see equation 2, page 26). Likewise, since we used open pollinated mother plants, it is not entirely, certain whether the offspring were full- or half-sibs, or even selfings. However, in the absence of more detailed information, it is perhaps safest to assume that the offspring in a given family was half- rather than full-sibs (Squillace 1974). Nevertheless, even if we had assumed all the offspring in the given family to be full-sibs, the mean Q_{ST} would have been 0.44 or 0.45 for subsets 1 and 2, respectively. Hence, even though the degree of differentiation might actually lie within the range explained by genetic drift alone, it is fairly clear that the quantitative trait differentiation among the Spanish cedar populations I studied does not exceed the value to be expected due to drift alone.

5.3.2. Marker vs. quantitative trait divergence across different populations

In their review of earlier studies, Merilä and Crnokrak (2001) showed that the degree of among-population differentiation in quantitative traits could be predicted from knowledge of the degree of differentiation for neutral markers across the various studies conducted so far. Although this finding indicates that molecular markers could be used as surrogate estimates of the degree of adaptive differentiation among populations when quantitative genetic data are

not available, this may be a premature conclusion as the data on which this result is based comprised studies where both F_{ST} and Q_{ST} values ranged from zero to unity. Such a variation is hardly ever observed in intraspecific studies and, in fact, only one study to date has attempted to test for this relationship with intraspecific data (Long and Singh 1995). Their study showed a strong positive correlation between pair-wise F_{ST} and Q_{ST} estimates, corroborating the interspecific level comparisons of Lynch *et al*. (1999) and Merilä and Crnokrak (2001). This is noteworthy as it suggests that knowledge of the degree of population differentiation for molecular markers in *C. odorata* reveals the degree of genetic differentiation in ecologically important traits.

One explanation for this good correspondence is that the differentiation in both sets of markers is driven at least partly by the same causal factor(s). For instance, although quantitative trait divergence among the populations was less than expected on the basis of genetic drift, it is quite likely that part of differentiation in these traits does arise from genetic drift. An alternative, and mutually compatible explanation is that the genes coding for the expression of quantitative traits in question exhibit some degree of linkage with the RAPD markers used, which would tend to restrict the divergence between Q_{ST} and F_{ST} estimates.

However, although I cannot rule out any of these explanations, the most prudent conclusion regarding this correlation between divergence in Q_{ST} and F_{ST} estimates may come about through their sharing a common relationship to geographic distance (Merilä and Crnokrak 2001). For neutral marker genes, the differentiation among populations typically (but not always) increases with geographic distance, as is the case also in our data for sub-set 1 (correlation between geographic distance and F_{ST}: $r = 0.64$, $P < .0001$). As to quantitative traits, for which patterns of differentiation are thought to be governed mainly by variation in local selection pressures, one would expect a similar relationship if the heterogeneity in selection pressures were also a function of geographic distance. While this currently remains

an untested hypothesis, it seems plausible that the correlation between F_{ST} and Q_{ST} estimates could be driven by different processes which, however, bear a similar relationship to geographic distance separating pairs of populations.

5.3.3. Marker vs. quantitative genetic diversity

The levels of intrapopulational genetic variability for neutral markers were unrelated to intrapopulational variability for quantitative traits as measured by heritability and the coefficient of additive genetic variance. This finding accords with the few similar tests conducted so far (Cheverud *et al*. 1994, Waldmann and Andersson 1998, Lynch *et al*. 1999, Hurme *et al*. 2000, Pfrender *et al.* 2000, but see: Briscoe *et al*. 1992), and perhaps should not surprise us given the multitude of factors that might influence levels of variability in quantitative traits (reviewed in Pfrender *et al*., 2000). Furthermore, despite the fact that *C. odorata* seems to have a very small genome (Wilson *et al*. 2001), the relatively few RAPD loci included in the present study may not give a representative picture of the genome-wide genetic variability in these populations. This paucity of loci, together with limited number of populations I studied may render my conclusion quite conservative. However, given the fact that sample sizes were not smaller than those typically used in genetic conservation studies of wild populations, my results are in line with the conjecture that neutral molecular markers may not be very useful for purposes of inferring levels of variability in quantitative traits (Lynch 1996, Pearman 2001, Pfrender *et al.* 2000).

Since heritability was estimated on a greenhouse trial in which very young progenies were used, strong maternal effects could be the most important factors influencing the estimates. Mothers have the ability to profoundly affect the quality of their offspring. In many instances, these maternal effects may be the single most important contributors to variation in offspring fitness (Mousseau and Fox 1998).

5.4. GENETIC IMPROVEMENT OF *C. ODORATA* ASSOCIATED WITH COFFEE

5.4.1. Effects of C. odorata provenance and coffee cultivation conditions on growth of C. odorata

The growth of *C. odorata* associated with coffee was remarkable compared with other published reviews and reports (Cintron 1990, Guevara 1988, Newton *et al*. 1998,). For example, in Apartado, Colombia, Guevara (1988) reported a mean annual increment in diameter of 2.1 cm on the best sites at 24 months. The height growth was in the range of 2.5 m/year to 0.5 and 0.3 m/year at 2 years old. In Costa Rica the height growth for pure *C. odorata* plantations has been reported as 1 m/year (Newton *et al*. 1988 cited by Mesén 1999).

In the analysis of provenances of *C. odorata* in the international tests coordinated by Oxford Forestry Institute in 1967, fast-growing provenances such as San Carlos (SC) were able to grow a new leader with strong apical control and to recover from the attack by the shootborer (Chaplin 1980, McCarter 1986, Newton *et al*. 1993). Similarly in the present study, SC was in the top five provenances followed by PZ, PS and TAL.

Although the Tikal provenance is from xeric northern Guatemala, it ranked ninth for height and tenth for diameter becoming the best xeric provenance, even better than some of the provenances from the mesic regions. Genotype x environment interactions for growth traits are important in *C. odorata*, and appear to be related to climatic effects. Therefore, provenances will also have to be compared in Mexican trials to give information on which to base successful development of forest plantations.

Provenances from dry areas were more resistant to *H. grandella*, representing a lower frequency of attack than for provenances from wet areas. These results accord with the results reported by Lopez *et al*. (1997) for monocultures in Costa Rica.

The young coffee plots presented the highest mortality for *C. odorata* (21 %), presumably because the trees were less protected against sunshine and less hidden from the moth of *H. grandella*.

Mortality in blocks where the trees were planted within the coffee rows was only 6 – 12%. This low mortality is probably due to shading, which favours tree growth and protects them from insect attacks.

The results of the agroforestry trial involving *C. odorata*-coffee mixtures are encouraging, and could motivate new studies under different climatic conditions using the methodology and the technological package presented in the agroforestry study. Prior to the present work, not many reports concerning *C. odorata* in agroforestry systems have been published. Guevara (1988) presented results for this species in Colombia. *C. odorata* in association with plantain (*Musa paradisiaca*) presented the best increments on alluvial soils (mean annual increment of 2.2 and 3.3 m/year in height at 4.25 and 1.8 years) while *C. odorata* associated with annual crops in rotation with fruit trees grew 2.9 m/year in height and 3.2 cm/year in diameter which compares well with the present study. A mean annual increment of 0.8 m/year in height and 1.5 cm/year in diameter was obtained in San Carlos, Costa Rica in coffee agroforestry systems (Ford 1979).

5.4.2. Heritability, progeny effects and genetic gain

The results of the agroforestry trial with *C. odorata*-coffee mixtures showed the presence of important family differences for all the traits analysed. The difference between the best (6232) and the worst (168) progenies was 186 % for height and 100 % for diameter (6240 and 699). Lopez (1997) reported a range of growth between progenies from 0.95 to 2.0 m for height at nine months in a pure plantation. The results in the agroforestry systems *C. odorata*-coffee are comparable (2.07 m/year for height in the best progeny and 0.72 m/year in the progeny with lowest growth, at 25 months).

James *et al*. (1998) estimated that the multilocus outcrossing rate for *C. odorata* was 0.969, suggesting that this species may be self-incompatible. Inbreeding can still nonetheless occur, since it can result from any kind of mating between individuals that are related to each other by ancestry; also the presence of full sibs is possible because of the reduced number of trees in some deforested populations. Diminution of pollinators in the natural forest can lead to inbreeding, as was demonstrated for *Shorea* by Ghazoul *et al*. (1998). The coefficient of relationship among open pollinated offspring depends on the frequency of selfing, frequency of related matings, and number of pollen parents (Squillace 1974). A study to estimate this coefficient for *C. odorata* could lead to better estimates of the heritability values.

The tropical dry forest species *Bombacopsis quinata* showed single-site heritabilities for height of 0.23 at three years and 0.24 at eight years and for diameter at breast height 0.18 at 3 years and 0.27 at eight years (Hodge *et al*. 2002). Their values are comparable to those found in the present study.

Genetic gains at 5 % selection intensity were 10 % for diameter and 21 % for height. The additive coefficient of variance is a convenient way of expressing the size of the additive

variance that controls a trait or the potential gain through selection for a specific trait. The genetic gain, expressed as a percentage (Hodge *et al*. 2002, p.286) when the top 5 % is selected (i = 2.063) is calculated with the next example for height:

$\Delta G_5 = 100\ \% \times (2.063 \times (5157.4)^{0.5}\ ) / 328\ cm = 45\%$; where 5157.4 and 328 are the additive variance and the mean of the families within the selected local provenances, respectively.

The values of 25 % and 45 % for diameter and height are the potential gains from selecting the top 5 % of the population, and represent the maximum genetic gain without error (heritability equals one) for an elite population (Hodge *et al*. 2002). However, to achieve a maximal heritability of 1.0 is only possible by using clonal propagation that could be easily done with *C. odorata*. My estimates for height (0.2) and diameter (0.12) suggest a smaller genetic gain for open pollinated trees.

The results of this study demonstrate that there are excellent possibilities for selecting progenies and provenances with better performance in growth and resistance to the shootborer under agroforestry systems. Progenies should be selected for resistance to *H. grandella* and for producing one main shoot after attack. Progenies that were more resistant such as 752, 6114, 6270, 180 and 192 should be analysed chemically, to determine the presence of possible chemicals that prevent the attack of the moth.

The association of *C. odorata* with coffee is not only convenient for production purposes but also for conservation of endangered *C. odorata* populations.

5.5. REPRODUCTIVE ISOLATION

Unisolated mother trees (isolation 3) produced progenies superior to those from isolated mother trees. I interpret this observation as indicating inbreeding in the progenies of isolated trees (isolations 1 and 2). Kärkkäinen *et al.* (1996) and Koski and Muona (1986) in studies with *Pinus sylvestris* have shown the same possible effect of inbreeding. Significant differences have been obtained in plant vigour between seedlings from continuous forests and pastures, suggesting that habitat fragmentation may, by increasing the rates of self-pollination, disrupt the ability of the trees to regulate the quality of their progeny and thus cause the pasture trees to produce fewer viable fruits or fruits with depressed vigour (Rocha and Aguilar 2001). Griffin (1991) also mentions that outcrossing rates from natural populations are not necessarily good indicators of the breeding systems under non-natural conditions, and that facultative selfing or other types of inbreeding leading to inbreeding depression could occur in very fragmented populations.

Mother trees in isolation 3 in xeric conditions (Table 19), were significantly superior to mother trees in isolations 1 and 2. In mother trees of mesic areas, the progenies of isolations 2 and 3 were significantly superior to that of isolation 1. *C. odorata* is probably pollinated by short distance pollinators, mainly small bees (Navarro 1999) and small moths (Bawa *et al*. 1985) that could have difficulty flying long distances in grasslands or agricultural fields. I speculate that the presence of secondary forest, more associated tree species and also the presence of natural forests in the vicinity provide corridors for pollinators that would enable isolates (2) in mesic areas to become cross-pollinated. On the other hand, xeric isolates (2) are in pastures, highly susceptible to fires and other human interventions like agriculture, this could explain a low presence of pollinators. The presence of associated species could be important in the survival of the pollinators for alternative foraging while *C. odorata* is not

flowering. This could explain why isolation 2 was not significantly different from isolation 3 in the mesic progenies.

Xeric regions have the longest history of human intervention in the neotropics. In contrast, mesic regions of the Atlantic coasts of Panama and Costa Rica lack a clear dry season, and farmers have not used fires to burn the forest to change the land use to pastures or agriculture.

Janzen (1986) notes that isolated trees are "living fossils", with no conservation value. In the light of the present study, the isolation levels differ in their ability to produce progenies of good quality i.e. trees isolated from both their conspecifics and other associated trees are less likely to produce good progenies. However, isolated trees could play an important role in the pollination process if populations were improved by planting conspecifics and improving the environment with other associated species. Genetic resource management should consider fragmentation and reproductive isolation (Aldrich and Hamrick 1998).

Forest conservation genetics and tree improvement programmes must consider the risk of inbreeding depression when seed is collected from single trees far apart. The mating system, including estimates of selfing, should be assessed using relevant molecular and quantitative markers in the highly deforested dry areas of Mesoamerica. Alternatively, propagating such trees vegetatively in special genetic archives where they could belong to artificially constructed populations could save the genetic resources of trees isolated by long distance.

C. odorata is a rare species (Condit *et al*. 1996) presenting even a lower proportion of individuals per hectare than *S. macrophylla*. For instance, in Quintana Roo, Mexico, conservation forests carry 0.65 trees per ha for *S. macrophylla*, (>15 cm diameter) and 0.175 trees per ha (45 – 65 cm diameter) for *C. odorata* (Patiño 1997). This low density could be due to the greater susceptibility of *C. odorata* to *H. grandella* attack in comparison to *S. macrophylla.* The effective number of pollinators was sometimes less than 20 per population,

so the effective population sizes could be very small. Deforested areas have fewer pollinators. In my analysis, I found that the trees in exploited areas were less vigorous. Pollinator decline could be affecting the mating of the species, leading to inbreeding and reduce the vigour of the seeds, even lethality.

The criteria used for logging of the adult trees in the field are minimum diameter, height, good form, and absence of a hollow trunk. Genetic diversity studies on mahogany were correlated with the level of exploitation or destruction of the forest (Gillies *et al.* 1999). Their results support the present in that the progeny of solitary trees expressed depressed growth. This indicates that negative selection by human exploitation of the best trees could have restricted genetic variability, mainly of the best performing mother trees, reducing the overall genetic diversity.

The collection of seeds from individual trees to grow families has provided evidence to indicate the value of careful studies on population structures and pollination vectors. This is especially important when assessing inbreeding risks in sparse populations that may be the result of long-term forest exploitation. I recommend combining quantitative studies with molecular markers in future studies in order to save valuable genetic resources. Only after such studies will it be possible to plan optimal tree breeding programmes for species such as *C. odorata*.

The coefficient of inbreeding of *C. odorata* (F_{IS}) was estimated at 0.67± 0.12. The estimate of the multilocus outcrossing rate for *C. odorata* is 0.969 and suggests this species may be self-incompatible (James *et al.* 1998). Inbreeding, will, however, result from any kind of mating between individuals that are related to each other by ancestry. Thus, inbreeding and the presence of full sibs is possible in *C. odorata* in some deforested populations and where natural pollinators have declined (Ghazoul *et al.* 1998, Aizen and Feinsinger 1994a, 1994b).

The study of isolation brings about important implications for the efficient use and conservation of genetic resources of tropical forest trees. It shows that seed from isolated trees is of inferior quality for establishing plantations; the progenies will perform more poorly than the trees from clusters or natural forests. Analogous results with other species have been explained as a consequence of a disruption of the mating systems (Rocha and Aguilar 2001, Stephenson 1992, Cascante *et al*. 2002). Nevertheless, isolated genotypes/trees can still be extremely valuable if introduced into breeding programs after testing.

5.6. FUTURE USE OF *C. ODORATA* GENETIC RESOURCES

5.6.1. Implications for management of endangered populations

Results of this study of molecular markers and quantitative traits have revealed strong differentiation among the Mesoamerican populations of *C. odorata*. The much wider geographical range covered by the present study shows that earlier studies captured only a limited proportion of the diversity of *C. odorata* (Gillies *et al*. 1997). It is clearly necessary to preserve *in-situ* conservation areas, as well as *ex-situ* and *circa-situ* gene banks and plantations, not only of one, but also of several provenances of *C. odorata*.

Reduced effective population size may cause inbreeding. Conservation can be integrated with other land uses, such as wood production, cattle farming, recreation, agriculture, and be practised even in villages or cities. Much of the resources of *C. odorata* lie outside the reserves, mainly in farmers' agroforestry systems, which form a mosaic in the agroecosystem with the forest fragments. Similar situation prevails for other important species like *Pinus*, *Leucaena*, *Prosopis*, *Cordia, Gliricidia* (See Ledig et al. 1988, Hughes 1988, Barrance 2000). So *in situ* conservation on-farm, also called *circa situ* conservation by Hughes (1988), includes all conservation in managed farming landscapes, e.g. trees in pastures, fodder trees, agroforestry systems, living fences, border lines, shade for crops e.g. coffee shade, home gardens, windbreaks and other systems long used in traditional farming in Mesoamerica.

5.6.2. Strategies for conservation and population improvement

The strategies for *ex-situ* conservation and population improvement include:

1. Vegetative propagation of mother trees. For this approach to be successful, it is important that the mother trees are not inbred and are growing under natural conditions. In other words, the mother trees must arise from relatively non-related parents and be naturally selected from hundreds of seedlings (see Griffin 1991). The policy of vegetatively propagated mother trees can be taken a step further by composing "clonal archives" consisting of all mother tree ramets representing a well-defined climatic zone. In this way, one may compose new artificial populations where pollination takes place between ramets representing ortets from different single trees otherwise located far apart. The problem that might arise from using inbred ortets for ramet production is the fact that inbreeding may cause reduced flowering and weak seed set.

2. Importation of seeds from other populations outside the one to be improved. This should be done when there are too few mother trees in the population, or the population is extinct.

3. Establishment of conservation gardens that include at least twenty non-related progenies. In the case of seed orchards, an option would be to use single-tree plots to avoid mating between individuals that are related to each other by ancestry (see Boshier and Lamb 1997, Lindgren 2000). The use of 20 unrelated parents is a lower limit to save a valuable population. Higher numbers, up to 100 would be more suitable to avoid risks of inbreeding in later generations.

4. Development of low intensity breeding programs for the tropical species. In the tropics, there are hundreds of important species, and it is not feasible to develop expensive breeding programmes for each one. Since tropical countries are generally poor, it is more appropriate to develop low intensity breeding programmes that could involve many species with a reduced amount of resources (see Lindgren 2000, Boshier and Lamb 1997, Hughes 1988).

The results of the present studies suggest possible incipient speciation and/or subspecies status of different *C. odorata* populations living in contrasting (mesic vs. xeric) environmental conditions. Given the socio-economic importance and endangered status of *C. odorata* populations, our results highlight the need for future studies encompassing the whole natural distribution range of the species, including the still unstudied populations in South America.

The importance of *C. odorata* in Central America warrants a coordinated effort between all the Mesoamerican countries. The activities must include farmer participation in the conservation, plantation and management of this important species. One of the challenges to overcome in the management of the broad-leaved forests with *C. odorata* is that of increasing the harvesting and commercialisation of other species, in order to decrease excessive pressure on the utilization of precious woods. To this end, plantations of the species could be established both in agroforestry and in mixed plantations.

Further studies on the attacks of *H. grandella* on *C. odorata* throughout the range of distribution are needed. The comparison of molecular markers studies and quantitative markers in the populations of South America will give a better picture of the genetic variation of the species and adaptation to clines, climate and soil.

Genetic modification for inducing resistance to this endangered species is of great interest for the many companies that want to plant the species. Presently companies are reluctant to make heavy investments due to shootborer attack. Access to strains modified for resistance would render investors more willing to plan large-scale plantings. This would be feasible in areas where the species is already extinct or outside the natural distribution to prevent risks from contamination of genetically modified (GM) trees into the forest through pollen flow. The plantation of GM trees of *C. odorata* in the natural distribution of the species would be

dependent on the development of methods and procedures for evaluating potential ecological and environmental risks associated with the release of GM trees.

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REFERENCES

Aguilar, M. and Aguilar, M.C. 1992. Arboles de la Biosfera Maya Petén. Universidad de San Carlos de Guatemala, Guatemala. 272 p.

Aizen, M.A. and Feinsinger, P. 1994a. Forest fragmentation, pollination, and plant reproduction in a chaco dry forest, Argentina. Ecology 75: 330–351.

Aizen, M.A. and Feinsinger, P. 1994b. Habitat fragmentation, native pollinators and feral honey bees in Argentine "chaco serrano". Ecological applications 4: 378–392.

Aldrich, P.R. and Hamrick, J.L. 1998. Reproductive dominance of pasture trees in a fragmented tropical forest mosaic. Science 281: 103–105.

Alvarez, M. 1999. Caracterización de frutos y semillas de *Cedrela odorata* L., *Tabebuia rosea, Alnus acuminata* y *Cupressus lusitanica*. *In*: Avances en la Producción de Semillas Forestales en América Latina R. Salazar (ed). CATIE, Turrialba, Costa Rica. pp. 145–150

Avise, J.C. 1994. Molecular markers, natural history, and evolution. Chapman and Hall, New York. 511 p.

Avise, J.C and Hamrick, J.L. 1996. Conservation genetics. Case histories from nature. Chapman and Hall, New York. 512 p.

Barrance A. 2002. Circa situm Conservation of Multi-Purpose Tree Species Diversity in Honduran Dry Forest Agroecosystems. http://www.condesan.org/infoandina/Foros/insitu99/1412.htm.

Barres, H. 1963. A provenance study of *Swietenia*. USDA Forest Service Study Plan 2421 (unpublished). Institute of Tropical Forestry, Rio Piedras, Puerto Rico.

Bawa, K.S., Bullock, S.J., Perry, D.R., Beach, J.H. 1985. Reproductive biology of tropical lowland rain forest trees. II Pollination systems. Am. J. Bot. 72: 346–356.

Beer, J., Muschler, R.; Somarriba, E.; and Kass, D.L. 1997. Maderables como sombra para café. Serie Boletín PROMECAFE (IICA). 76–77: p. 5–7.

Beer, J.W.; Heuveldop, J. (eds.). 1989. A critical analysis of an agroforestry project in Acosta and Puriscal, Costa Rica. CATIE, Turrialba (Costa Rica). Serie Técnica. no. 147. p. 90–104.

Benencia F, 1995 Immunomodulatory activities of *Cedrela tubiflora* leaf aqueous extracts. J Ethnopharmacol 49(3): 133–139.

Boshier, D.H. and Lamb, A.T. 1997. *Cordia alliodora*: Genética y Mejoramiento de Arboles. Tropical Forestry Paper 36, Oxford Forestry Institute. 70 p.

Briscoe, D.A., Malpica, J.M., Robertson, A., Smith G.J., Frankhamn, R., Banks, R.G. and Barker, J.S.F. 1992. Rapid loss of genetic variation in a large captive population of *Drosophila* flies: implications for the genetic management of captive populations. Conserv. Biol. 6: 416–425.

Burley J, Lamb A.F.A. 1971. Status of the CFI international provenance trial of *Cedrela odorata* (including *C. mexicana* and *C. tubiflora*). Comm. For. Rev. 50(3): 234–237.

Butlin, R.K. and Tregenza, T. 1998. Levels of genetic polymorphism: marker loci versus quantitative traits. Proc. R. Soc. Lond. B. 358: 187–198.

CAB International. 2000. Forestry Compendium Global Module. CAB International, Wallingford, UK. 247 p.

Canadian Government Publishing. 2001. Canada Gazette Part II. Vol 135. No. 26. SOR/DORS/2001-522. Ottawa. Canada. Pp 1912–1917 and 2850–2858.

Carswell, F.E., Grace, J., Lucas, M.E. and Jarvis, P.G. 2000. Interaction of nutrient limitation and elevated CO_2 concentration on carbon assimilation of a tropical tree seedling (*Cedrela odorata*). Tree Physiology 20: 977–986.

Cascante, A., Quesada, M., Lobo J. and Fuchs E. 2002. Effects of dry tropical forest fragmentation on the reproductive success and genetic structure of the tree *Samanea saman.* Cons. Biol. 16: 137–147.

Castro O, 1996 Chemical and biological evaluation of the effect of plant extracts against *Plasmodium berghei.* Rev. Biol. Trop. 44(2A): 361–367.

Chaplin G.E. 1980. Progress with provenance exploration and seed collection of *Cedrela* spp. Proc. 11th Commonwealth For. Conf., Commonwealth Forestry Institute, Oxford, UK. pp. 1–17.

Cheverud J., Routman, E., Jaquish, C., Tardif, S., Peterson, G., Belfiore, N. and Forman, L. 1994. Quantitative and molecular genetic variability of captive cotton-top *tamarins (Saguinus oedipus*). Conserv. Biol. 8: 95–105.

Cintron, B.B., 1990. *Cedrela odorata* L. Cedro Hembra, Spanish Cedar, Meliaceae. Mahogany family. In: Silvics of North America. Hardwoods. Agric. Handbook, 654. Washington, DC. USDA. Vol. 2. Pp 250–257.

Clausen, J., Keck, D.D., and Hiesey, W.M. 1948. Experimental studies on the nature of species. III. Environmental responses of climatic races of *Achillea*. Carnegie Inst. Wash. Publ. 581. Washington, D.C. 128 p.

Clausen, J., D. D. Keck, and W. M. Hiesey. 1940. Experimental studies on the nature of species. I. The effect of varied environments on western North American plants. Carnegie Inst. Wash. Publ. 520. Washington, D. C. 120 p.

Colinvaux, P. A. 1996. Quaternary environmental history and forest diversity. *In:* Jackson, J.B.C., Budd A.F. and Coates A.G. (eds.). Evolution and Environment in Tropical America. University of Chicago, Illinois. Pp. 359–405.

Condit R., Hubbell, S.P, Foster R.B.. 1996. Changes in tree species abundance in a neotropical forest impact of climate change. J. Trop. Ecol. 12: 231–256.

Cornelius, J. 2001. The effectiveness of pruning in mitigating *Hypsipyla grandella* attack on young mahogany (*Swietenia macrophylla* King) trees. For. Ecol.Manag. 148: 287–289.

Cotterill, P.P. 1987. Short note: on estimating heritability according to practical applications. Silvae Genet. 36: 46–48.

Dawson, I., Simons, A., Waugh, R. and Powell, W. 1995. Diversity and genetic differentiation among subpopulations of *Gliricida sepium* revealed by PCR-based assays. Heredity 74: 10–18.

Dieters, M.J., White, T.L., Littell, R.C. and Hodge, G.R. 1995. Application of approximate variances of variances components and their ratios in genetic tests. Theor. Appl. Genet. 91: 15–24.

Dodd, D. H. and Schultz, Jr., R.F. 1973. Computational procedures for estimating magnitude of effects for some analysis of variance designs. Psycho. Bull. 79: 391–395.

Eldridge, K.G. and Griffin, A.R. 1983. Selfing effects in *Eucalyptus regnans*. Silvae Genetica. 32: 216–221.

Excoffier, L., Smouse, P.E. and Quattro, J.M. 1992. Analysis of molecular variance inferred from metric distances among DNA haplotypes: application to human mitochondrial DNA restriction data. Genetics 131: 479–491.

Fleiss, J. L. 1969. Estimating the magnitude of experimental effects. Psycho. Bull. 72: 273–276.

Ford, L.B. 1979. An estimate of the yield of *Cedrela odorata* L. (Syn. *C. mexicana* Roem.) grown in association with coffee. CATIE. Turrialba Costa Rica. Centro Agronómico Tropical

de Investigación y Enseñanza, Turrialba (Costa Rica); United Nations Univ., Tokyo (Japón). Workshop Agro-Forestry Systems in Tropical Latin America. Turrialba (Costa Rica). 1979. 11 p.

Frankham, R. 1999. Quantitative genetics in conservation biology. Genet. Res. 74: 237–244

Frankie, G.W., Opler, P.A., and Bawa, K. 1976. Foraging behaviour of solitary bees: Implications for outcrossing of a Neotropical forest tree species. J. Ecol. 64: 1049-57

Geary, T. F., Barres, H. and Ybarra-Coronado, R.. 1973. Seed source variation in Puerto Rico and Virgin Islands grown mahoganies. USDA Forest Service Research Paper ITF-17.

Ghazoul, J., Liston, K.H., Boyle, T.J.B. 1998. Disturbance-Induced Density-Dependent Seed Set in *Shorea siamensis* (Dipterocarpaceae), a Tropical Forest Tree. J. Ecol. 86 (3): 462-473

Gillies, A.C.M., Cornelius, J.P., Newton, A., Navarro, C., Hernández M. and Wilson. J. 1997. Genetic variation in Costa Rican populations of the tropical timber species *Cedrela odorata* L. assessed using RAPDs. Mol. Ecol. 6: 1133–1146.

Gillies, A., Navarro, C., Lowe, A.J, Newton, A.C., Hernandez, M., Wilson J. and Cornelius, J.P. 1999. Genetic diversity in Mesoamerican populations of mahogany (*Swietenia macrophylla*), assessed using RAPDs. Heredity 83: 722–732.

Graham, A. 1999. Studies in Neotropical paleobotany. XIII. An Oligo-Miocene palynoflora from Simojovel (Chiapas, Mexico). Am. J. Bot., 86(1): 17–31.

Griffin, A.R. 1991. Effects of inbreeding on growth of forest trees and implications for management of seed supplies for plantation programmes. *In*: Bawa, K.S. and Hadley, M., (eds). Reproductive Ecology of Tropical Forest Plants. Man and the Biosphere Series, vol 7. Unesco/Parthenon Publishing, Paris and Carnforth. Pp: 355–374

Griffin, A.R., and Lindgren, D. 1985. Effect of inbreeding on production of filled seed in *Pinus radiata* experimental results and a model of gene action. Theor. Appl. Genet. 71: 334–43.

Gripjma, P. Ed. 1973. Studies on the shootborer *Hypsipyla grandella* (Zeller) Lep., Pyralidae. San José, Costa Rica. IICA Miscellaneous Publ. No. 101. V. 1. 91 p.

Guevara, G. 1988. Experiencias Colombianas con Cedro. Corporación Nacional de Investigación y Fomento Forestal. Serie Documentación No.12. Bogotá, Colombia. 85 p.

Haig, S.M. 1998. Molecular contributions to conservation. Ecology 79: 413–425.

Hammond, D.S., Dolman, P. and Watkinson, A.R. 1995. Modern Ticuna swidden-fallow management in the Colombian Amazon: ecologically integrating market strategies and subsistence-driven economies? Human Ecology 23**:** 335-356.

Hamrick, J.L. and Godt, J.M. 1990. Allozyme diversity in plant species. *In* Brown, A.D.H., Clegg, M. T., Kahler, A. L. and Weir, B. S. (eds.). Plant Population Genetics Breeding and Genetic Resources, Sinauer, New Jersey. Pp. 43–63

Hardy, O. J., Vanderhoeven, S., Meerts P. and Vekemans, X. 2000. Spatial autocorrelation of allozyme and quantitative markers within a natural population of *Centaurea jacea* (Asteraceae). J. Evol. Biol. 13: 656–667.

Hartl, D.L. and Clark, A.G. 1989. Principles of population genetics. Sinauer, Sunderland, Massachussets. 481 p.

Hilje, L., Cornelius, J. 2001. Es inmanejable *Hypsipyla grandella* como plaga forestal? Manejo Integrado de Plagas. CATIE (Costa Rica). No. 61. 1–4

Hilton-Taylor, C. (compiler) 2000. IUCN Red List of Threatened Species. IUCN, Gland, Switzerland and Cambridge, UK. xviii + 61p.

Hodge, G.R., Dvorak, W.S., Urueña, H. and Rosales, L. 2002. Growth, provenance effects and genetic variation of *Bombacopsis quinata* in field tests in Venezuela and Colombia. For. Ecol. Manage. 158: 273–289.

Hodgson, L.M. 1976. Some aspects of flowering and reproductive biology in *Eucalyptus grandis* at J.D.M. Keet Forest Research Station. 2. The fruit, seed, seedlings, self-fertility, selfing and inbreeding effects. South African Forestry Journal 98: 32–43.

Holdridge, L.R. 1967. Life zone ecology. Tropical Science Center, San José, Costa Rica. 206p.

Holsinger, K.E. and Lewis, P. 2002. Hickory. http://darwin.eeb.uconn.edu/hickory/hickory.html

Holsinger, K.E., Lewis, P.O. and Dey, D.K. 2002. A Bayesian approach to inferring population structure from dominant markers. Molec. Ecol. 11: 1157–1164.

Houle, D. 1992. Comparing evolvability and variability of quantitative traits. Genetics 130: 195–204.

Huber, D.A., White, T.L. and Hodge, G.R. 1994. Variance component estimation techniques with forest genetic architecture through computer simulation. Theor. Appl. Genet. 88: 236–242.

Hughes, C.E. 1998. Leucaena - A Genetic Resources Handbook. Tropical Forestry Papers No. 37. Oxford Forestry Institute. Oxford.

Hurme, P., Sillanpää, M.J., Arjas, E., Repo, T. and Savolainen, O. 2000. Genetic Basis of Climatic Adaptation in Scots Pine by Bayesian Quantitative Trait Locus Analysis. Genetics 156: 1309–1322.

IUCN. 2001. IUCN Red list categories and criteria: Version 3.1. IUCN Species Survival Commission. IUCN, Gland, Switzerland and Cambridge, UK. Ii + 30 p.

James, T., Vege, S., Aldrich, P. and Hamrick, J.L. 1988. Mating systems of three tropical dry forest tree species. Biotropica 30 (4): 587–594.

Janzen, D.H. 1983. Costa Rican natural history. The University of Chicago Press, Chicago, Illinois, USA. 789 p.

Janzen, D.H. 1986. Blurry catastrophes. Oikos. 47:1–2.

Janzen, D.H. 1988. Tropical Dry Forests: the Most Endangered Major Tropical Ecosystem., *In*: Wilson, E.O. (ed.) Biodiversity. National Academy Press. Washington, D.C. Pp.130–137.

Jarvis, D.I., Myer, L., Klemick, H., Guarino, L., Smale, M., Brown, A.D.H., Sadiki, M., Sthapit, B. and Hodgkin, T. 2000. A Training Guide for *In Situ* Conservation On-farm. Version 1. International Plant Genetic Resources Institute, Rome, Italy. 189 p.

Jennersten, O. 1988. Pollination in *Dianthus deltoides* (Caryophyllaceae): effects of habitat fragmentation on visitation and seed set. Conserv. Biol. 2: 359–366.

Karani, P.K. 1973. International provenance trials in Uganda, progress report on *C. odorata*. *In*: Burley, J. and Nikles, D.G. (eds). Tropical Provenance and Progeny Trials Research and International Cooperation. Oxford University, Oxford. Pp. 241–249.

Kärkkäinen, K., Koski, V. and Savolainen, O. 1996. Geographical variation in the inbreeding depression of Scots Pine. Evolution 50(1): 111–119.

Knapp, E.E. and Rice, K.J. 1998. Comparison of isozymes and quantitative traits for evaluating patterns of genetic variation in purple needlegrass (*Nasella pulchra*). Conserv. Biol. 12: 1031–1041.

Koski, V. and Muona, O. 1986. Probability of inbreeding in relation to clonal differences in male flowering and embryonic lethals. IUFRO Conference Proceedings. Oct. 13–17. 1986. Williamsburg, Virginia. Pp. 391–400.

Kundu, S. and Tigerstedt, P.M.A. 1997. Geographical variation in seed and seedling traits of Neem (*Azadirachta indica* A. Juss) among ten populations studied in growth chamber. Silvae Genet. 46: 129–137.

Ladiges, P.Y., Gray, A.M. and Lassak, E.V. 1981. Pattern of geographic variation, based on seedling morphology, in *Eucalyptus ovata* Labill. and *E. brookerana* A. M. Gray and comparisons with some other Eucalyptus species. Aust. J. Bot. 29: 593–604.

Lande, R and Barroclough, G.F. 1987. Effective population size, genetic variation, and their use in population management. *In*: Soulé M. (ed.) Viable populations for conservation. Cambridge Univ. Press, Cambridge. Pp. 87–123,

Lande, R. 1992. Neutral theory of quantitative genetic variance in an island model with local extinction and colonisation. Evolution 46: 381–389.

Ledig, F.T. 1988. The conservation of genetic diversity in forest trees. BioScience 38: 471–479.

Li, C. 1998. Variation of seedling traits of *Eucalyptus microtheca* origins in different watering regimes. Silvae Genet. 47: 132–136.

Lindgren, D. 2000. Low-intensity tree breeding. *In*: Lundkvist K. (ed.). Rapid generation turnover in the breeding population and low-intensity breeding. Department of Forest Genetics, Uppsala, SLU, Sweden. Research Notes 55: 37–48.

Long, A.D. and Singh, R.S. 1995. Molecules versus morphology: the detection of selection acting on morphological characters along a cline in *Drosophila melanogaster*. Heredity 74: 569–589.

Loopstra, C. A. and Adams, W.T. 1989. Patterns of variation in first-year seedling traits within and among Douglas-fir breeding zones in Southwest Oregon. Silvae Genet. 38: 235–242.

López, J. 1997. Variación en resistencia de cedro (*Cedrela odorata* L.) al ataque de *Hypsipla grandella* Zeller en Costa Rica. Tesis M.Sc., CATIE, Turrialba, Costa Rica. 111 p.

López, J., Jara, L.F. and Mesén, F. 1997. Variación en resistencia de *Cedrela odorata* al ataque de *Hypsipyla grandella*. Variation in *Cedrela odorata* resistance to *Hypsipyla grandella* attack. Revista Forestal Centroamericana (CATIE). 19: 20–25.

Lynch, M. 1996. A quantitative genetic perspective on conservation issues. *In*: Avise, J.C. and Hamrick, J.L, (eds). Conservation Genetics: Case Histories from Nature. Chapman and Hall, New York. Pp. 471–501.

Lynch, M., Pfrender, M., Spitze, K. Lehman, N., Hicks, J., Allen, D., Latta, L., Ottene, M. Bogue, F. and Colbourne, J. 1999. The quantitative and molecular genetic architecture of a subdivided species. Evolution 53: 100–110.

MacKinnon S. 1997. Antimalarial activity of tropical Meliaceae extracts and gedunin derivatives. J. Nat. Prod. 60(4): 336–341.

Malécot, G. 1948. Les Mathématiques de l'Hérédité. Masson et Cie, Paris.

Mancebo, F., Hilje, L., Mora, G.A., and Salazar, R. (2000) Antifeedant activity of *Quassia amara* (Simaroubaceae) extracts on *Hypsipyla grandella* (Lepidoptera: Pyralidae) larvae. Crop Prot. 19: 301–305.

Mancebo, F., Hilje, L., Mora, G.A., Castro, V.H. and Salazar, R. 2001. Biological activity of *Ruta chalepensis* (Rutaceae) and *Sechium pittieri* (Cucurbitaceae) extracts on *Hypsipyla grandella* (Lepidoptera: Pyralidae) larvae. Revista de Biología Tropical 49: 501–508.

Mancebo, F., Hilje, L., Mora, G.A., and Salazar, R. 2002. Biological activity of two neem (*Azadirachta indica* A. Juss., Meliaceae) products on *Hypsipyla grandella* (Lepidoptera: Pyralidae) larvae. Crop Prot. 21: 107–112.

McCarter, P.S. 1986. The evaluation of the international provenance trials of *Cordia alliodora* and *Cedrela* spp. Annual report to the UK Overseas Development Administration. Oxford Forestry Institute, Oxford.

Mayhew, J.E., Newton, A.C. 1998. The silviculture of mahogany. Wallingford, Oxford. CABI Publishing. 226 p.

McKay, J.K. and Latta, R.G. 2002. Adaptive population divergence: markers, QTL and traits. Trends Ecol. Evol. 17: 285–290.

McKay, J.K., Bishop, J.G., Lin, J.-Z., Richards, J.H., Sala, A. and Mitchell-Olds, T. 2001. Local adaptation across a climatic gradient despite small effective population size in the rare sapphire rockress. Proc. R. Soc. Lond. 268: 1–7.

Merilä, J. and Crnokrak, P. 2001. Comparison of genetic differentiation at marker loci and quantitative traits. J. Evol. Biol. 14: 892–903.

Mesén, F. 1999. Avances en el mejoramiento genético de meliáceas con referencia especial a América Central. Workshop "Consulta Mesoamericana sobre Conservación de Recursos Genéticos Forestales con Enfasis Especial en Meliáceas", (REDCA), San Salvador, El Salvador, 25 de marzo de 1999. 19 p.

Moritz, C., Lavery, S. and Slade, R. 1995. Using allele frequency and phylogeny to define units of conservation and management. Amer. Fish Soc. Symp. 17: 249–262.

Muona, O. 1989. Population genetics in forest tree improvement. *In*: Kahler, A.L. and Weir, B.S. (eds). Plant Population Genetics, Breeding and Genetic Resources. Sinauer Associates, Sunderland, MA, USA. Pp. 282–298.

Mousseau T. and Fox C. (eds). 1998. Maternal effects as adaptations. Oxford University Press. 400 p.

Nason, J. D., Herre, E. A., and Hamrick, J. L. 1996. Paternity analysis of the breeding structure of strangler fig populations: Evidence for substantial long-distance wasp dispersal. J. Biogeogr. 23: 501–512.

Navarro, C. 1999. Annual year report of the Mesoamerican Domestication of *Cedrela* and *Swietenia* project. USDA. Grant No. FG-CR-109. Project No. CS-FS-2 USDA CATIE. Costa Rica, Turrialba. 20 p.

Navarro, C. 2001. Diagnóstico de la caoba en Mesoamérica. Silvicultura-Genética CCT/ PROARCA/CAPAS. San José. Costa Rica. 53 p.

Navarro, C. 2002. Population fragmentation, reproductive isolation and progeny performance of the endangered species *Cedrela odorata*. Strategies for conservation and population improvement. Conserv. Ecol. (submitted).

Navarro, C. and Cornelius, J. 1996. El muestreo de poblaciones y los estudios de diversidad como base para la conservación y el mejoramiento genético forestal. *In*: Salazar, R (ed.). Avances en la producción de semillas forestales en América Latina (Proceedings of a symposium held in Managua, Nicaragua, 16–20 October 1995). Turrialba, Costa Rica, CATIE. Pp. 51–57

Navarro C. and Hernández G. 2001. Cómo introducir cedro (*Cedrela odorata*) y caoba (*Swietenia macrophylla*) dentro de cafetales: consejos prácticos para promover sistemas agroforestales. Revista Agroforestería de las Americas. Vol. 8 N° 30. http://www.catie.ac.cr/informacion/RAFA/.

Navarro C., Hernández, M., Wilson J. and. Gillies A. 2002a. Germplasm collection of *Swietenia macrophylla* King in Central America and Mexico. Presented at the International Conference on Big Leaf Mahogany: Ecology, Genetic Resources and Management, IITF, San Juan, Puerto Rico, October 22–24, 1996. Springer-Verlag (in press).

Navarro C., Montagnini, F. and Hernandez G. 2002b. Genetic variability of *Cedrela odorata* Linnaeus: results of early performance of provenances and progenies from Mesoamerica grown in association with coffee. For. Ecol. Manag. (submitted).

Navarro, C., Pappinen A., Tigerstedt P. and Merilä J. 2002. Comparison of quantitative and molecular genetic variability and differentiation of Spanish cedar (*Cedrela odorata)* populations. J. Evol. Biol (submitted).

Navarro C. and Vázquez, W. 1987. Genetic variability in seeds and seedlings of *Cedrela odorata*. Paper presented at Congreso Forestal Nacional de Costa Rica, San Jose, Costa Rica, Nov. 1986; CAB Abstracts 1990–1991. Centro Agronómico Tropical de Investigación y Enseñanza (CATIE). 12 p.

Navarro C., Ward, S. and Hernandez M. 2002. The tree *Cedrela odorata* (Meliaceae): a morphologically subdivided species in Costa Rica. Rev. Biol. Trop. 50(1): (in press).

Navarro, C., Wilson, J., Gillies, A. and Hernández, M. 2002. A New Mesoamerican Collection of Big-Leaf Mahogany. Presented at the International Conference on Big Leaf Mahogany: Ecology, Genetic Resources and Management, IITF, San Juan Puerto Rico, October 22-24, 1996. Springer-Verlag (in press).

Negreros C., P. and Mize, C. 1994. El efecto de la abertura del dosel y eliminación del sotobosque sobre la regeneración natural de una selva de Quintana Roo. *In*: Snook, L. and A. Barrera de Jorgenson, A. (eds). Madera, Chicle, Caza y Milpa: Contribuciones al Manejo Integral de las Selvas de Quintana Roo. INIFAP/PROAFT/AID/WWF-US, Mérida, México. Pp. 107–126.

Nei, M. 1987. Molecular Evolutionary Genetics. Columbia University Press. 512 p.

Newton, A.C., Baker, P., Ramnarine, S., Mesen, F., Leakey, R.R.B. 1993. The mahogany shoot borer: Prospects for control. For. Ecol. Manage. 57: 301–328.

Newton, A.C., Cornelius J.P., Mesén J.F., Corea E.A. and Watt A.D. 1998. Variation in attack by the mahogany shoot borer, *Hypsipylla grandella* (Lepidoptera: Pyralidae), in relation to host growth and phenology. Bull. Entomol. Res. 88: 319–326.

Newton, A.C., Watt, A.D., López, F., Cornelius, J.P., Mesén, J.F. and Corea, E.A. 1999. Genetic variation in host susceptibility to attack by the mahogany shootborer, *Hypsipyla grandella* (Zeller). Agric. For. Entomol. 1:11–18.

Nores, M.M. 1997. Immunomodulatory activities of *Cedrela lilloi* and *Trichilia elegans* aqueous leaf extracts. J. Ethnopharmacol. 55(2): 99–106.

Pappinen, A., Kasanen, R. and von Weissenberg, K. 1996. Identification of multiple *Endocronartium pini* infections in Scots pine with PCR. Eur. J. For. Path. 26: 183–191.

Park, Y.S. and Fowler, D.P. 1982. Effects of inbreeding and genetic variances in a natural population of tamarack [*Larix laricina* (Du Roi) K. Koch] in eastern Canada. Silvae Genet. 31(1): 21–26.

Patiño, F. 1997. Genetic resources of *Swietenia* and *Cedrela* in the Neotropics. United Nations Food and Agriculture Organization, Rome, Italy. 58 p.

Pearman, P.B. 2001. Conservation value of independently evolving units: Sacred cow or testable hypothesis. Cons. Biol. 15(3): 780–783.

Pennington, T.D., Styles B.T. and Taylor D.A.H. 1981. Meliaceae. Fl. Neotrop. Monogr. 28: 1–470.

Pfrender, M.E, Spitze K., Hicks J., Morgan K., Latta L. and Lynch, M. 2000. Lack of concordance between genetic diversity estimates at the molecular and quantitative-trait levels. Cons. Genet. 1: 263–269.

Poddar G. 1971. A note on the chemical investigation on the bark of *Cedrela toona* Roxb. Bull. Calcutta Sch. Trop. Med. 19(4): 89.

Ramírez L.G., Alfaro R., Fonseca C., Rodríguez O. and Rodríguez G. 1998. Avances de investigación sobre el asocio de árboles maderables con café. http//:www.infoagro.go.cr/ tecnologia/cafe98/ doc2.html.

Rauno, A. 1973. *Cedrela* spp. International provenance trial planted in 1971 at Longuza, Tanga Region, Tanzania. *In*: Burley, J. and Nikles, D.G. (eds). Tropical Provenance and Progeny Trials Research and International Cooperation. Oxford University, Oxford. Pp. 262–266.

Reed, D.H and Frankham, R. 2001. How closely correlated are molecular and quantitative measures of genetic variation? A meta-analysis. Evolution 55: 1095–1103.

Ridley, M. 1990. Evolution. Blackwell, London. 370 p.

Rocha, O.J., Aguilar, G. 2001. Reproductive biology of the dry forest tree *Enterolobium cyclocarpum* (Guanacaste) in Costa Rica: a comparison between trees left in pastures and trees in continuous forest. Amer. J. Bot. 88(9): 1607–1614.

Rogers, A.R. 1986. Population differences in quantitative characters as opposed to gene frequencies. Amer. Nat. 127: 729–730.

Román, A. R., Vargas Hernández, J.J., Veláquez Martinez, A., Jasso Mata, J. and Ortega Jimenez, E. 1996. Geographic and genetic variation of growth and biomass production in cocoite (*Gliricidia sepium*) under two soil moisture conditions. Agrociencia 30: 549–558.

Salas, A. 2002. Centro Agronómico Tropical de Investigación y Enseñanza. Resumen acumulado de datos meteorológicos. Estación meteorológica CATIE, Turrialba, Costa Rica. http://www.catie.ac.cr/meteorologia/acumulados.htm.

SAS Institute Inc. 1999. SAS/STAT® User's Guide, Version 8, Cary, NC: SAS Institute Inc. ® 3884 p.

Saunders, D.A., Hobbs, R.J. and Margules, C.R. 1990. Biological consequences of ecosystem fragmentation: a review. Conservation Biology 5: 18–32.

Sim, B.L. 1984. The genetic base of *Acacia mangium* Willd in Sabah. *In*: Barnes, R.D. and Gibson, G.L. (eds). Provenance and Genetic Improvement Strategies in Tropical Forest Trees, Oxford University, Department of Forestry, Oxford. Pp. 597–603.

Smith, T.B. and Wayne R.K. 1996 (eds). Molecular Genetic Approaches in Conservation. Oxford University Press. 504 p.

Sorenson, F.C. 1983. Geographic variation in seedling Douglas-fir (*Pseudotsuga menziesii*) from the western Siskiyou Mountains of Oregon. Ecology 64: 696–702.

Sorenson, F.C., Campbell, R.K. and Franklin, J.F. 1990. Geographic variation in growth and phenology of seedlings of the *Abies procera/A. magnifica* complex. For. Ecol. Manag. 36: 205–232.

Spitze, K. 1993. Population structure in *Daphnia obtusa*: quantitative genetic and allozyme variation. Genetics 135: 367–374.

Squillace, A.E. 1974. Average genetic correlations among offspring from open pollinated forest trees. Silvae Genet. 23: 149–156.

Stephenson, A.G. 1992. The regulation of maternal investment in plants. *In*: Marshall, C. and Grace, J. (eds). Fruit and seed production: aspects of development, environmental physiology and ecology. Cambridge University Press, Cambridge. Pp. 151–171.

Styles, B.T. 1981. Swietenioideae. *In*: Pennington, T.D. Styles, B.T. and Taylor, D.A.H. (eds). Meliaceae. Flora Neotropica Monographs, no. 28. New York Botanical Garden, New York. Pp. 359–418.

Styles, B.T. and Khosla, P.K. 1976. Cytology and reproductive biology of Meliaceae. *In*: Burley J. and Styles B. T. (eds). Tropical trees: variation, breeding and conservation. Linnean Society Symposium. Series Number 2: 61–68.

Taveras, R., Hilje, L., Hanson, P., Mexzon, R., Carballo, M. and Navarro, C. 2002. Population trends and damage patterns of *Hypsipyla grandella* (Lepidoptera: Pyralidae) in a mahogany stand, in Turrialba, Costa Rica. Agr. For. Entomol. (submitted).

Templeton, A. 1986. Coadaptation and outbreeding depression. *In*: Soulé, M. (ed.). Conservation Biology: the science of scarcity and diversity. Sinauer, Sunderland, MA. Pp. 105–116.

Templeton, A.R., Shaw, K., Routman, E. and Davis, S. 1990. The genetic consequences of habitat fragmentation. Ann. Miss. Bot. Garden 77: 13–27.

Toval, G. and Puerto, G. 1985. Variación geográfica de procedencias de *Pinus coulteri* D. Don and *P. sabiana* Dougl. en vivero. Ann. Inst. Nac. Inv. Agr. 9: 29–46.

Underwood, A.J. and Petraitis, P.S. 1993. Structure of intertidal processes at different locations: how can local processes be compared? *In*: Ricklefs, R.E. and Schluter, D. (eds). Species Diversity in Ecological Communities. University of Chicago, Chicago. Pp. 39–51.

Vaughan, G.M. and Corballis, M.C. 1969. Beyond tests of significance: estimating strength of effects in selected anova designs. Psycho. Bull. 72: 204–213.

Veitch N.C., Wright G.A. and Stevenson P. 1999. Four new tetranortriterpenoids from *Cedrela odorata* associated with leaf rejection by *Exopthalmus jekelianus*. J. Nat. Prod. 62: 1260–1263.

Vrijenhoek, R.C. 1994. Genetic diversity and fitness in small populations. *In*: Loeschcke, V. and Jain, S. (eds). Conservation Genetics. Birkhauser, Basel. Pp. 37–53

Waldmann, P. and Andersson, S. 1998. Comparison of quantitative genetic variation and allozyme diversity within and between populations of *Scabiosa canescens* and *S. columbaria*. Heredity 81: 79–86.

Ward, R.D., Skibinsky, D.O.F. and Woodwark, M. 1992. Protein heterozygosity, protein structure and taxonomic differentiation. Evol. Biol. 26: 3–131.

Westfall, P.H. 1987. A comparison of variance component estimates of arbitrary underlying distributions. J. Amer. Statist. Assoc. 82: 866–873.

Whitlock, M.C. 1999. Neutral additive genetic variance in a metapopulation. Genet. Res. 74: 215–221.

Whitmore, J.L.1971. *Cedrela* provenance trial in Puerto Rico on St. Croix: nursery phase assessment. Turrialba 21: 343–349.

Whitmore, J.L.1978. *Cedrela* provenance trial in Puerto Rico and St. Croix: establishment phase. Research Note ITF 16. Rio Piedras, Puerto Rico. 11p.

Williams, C.G., and Savolainen, O. 1996. Inbreeding depression in conifers: implications for breeding strategy. For. Sci. 42: 102–117.

Wilson J., Lowe A., Cavers S., Navarro C., Hernandez M., Kremer A., Caron H., Labbe P., Margis R., Margis, M., Breyne, P., Gribel, R. and Lemes, M. 2001. Assessment of levels and dynamics of intra-specific genetic diversity of tropical trees. Final Scientific Report 1997–2001. European Commission. 76 p.

Winer, B.J., Brown, D.R. and Michels, K.M. 1991. Statistical Principles in Experimental Design. McGraw-Hill, San Francisco, California. 1057 p.

Wright, R.A., Wein, R.W. and Dancik, B.P. 1992. Population differentiation in seedling root size between adjacent stands of jack pine. For. Sci. 38: 777–785.

Wright, S. 1951. The genetic structure of populations. Annu. Eugenics 15: 323–420.

Yeh, F.C. and Boyle, T.J.B. 1997. Population genetic analysis of co-dominant and dominant markers and quantitative traits. Belg. J. Bot. 129: 157.

Young, A., Boyle T. and Brown, T. 1996. The population genetics of habitat fragmentation for plants. Trends Ecol. Evol. 11: 413–418.

Young, A., Boshier, D. and Boyle, T. (eds). 2000. Forest Conservation Genetics. Principles and Practice. CABI. 360 p.

Appendix 1. Least square means and standard errors for quantitative traits in *Cedrela odorata* greenhouse study. See methods for trait abbreviations.

n = number of families/individuals.

Population	H62 (1)	LW62 (2)	LL62 (3)	LL/LW62 (4)	H252 (5)	D252 (6)	ID252 (7)	NL252 (8)	n
Charagre	99.7 ± 1.5	11.2 ± 0.2	32.0 ± 0.3	2.93 ± 0.05	8.3 ± 0.5	0.30 ± 0.01	5.7 ± 1.9	4.3 ± 1.7	15/90
Cobano	99.2 ± 3.3	12.8 ± 0.6	31.8 ± 1.3	2.56 ± 0.09	60.9 ± 5.3	0.92 ± 0.08	36.1 ± 1.7	15.6 ± 0.7	7/40
Escarcega	133.8 ± 1.7	15.2 ± 0.2	36.6 ± 0.4	2.46 ± 0.03	64.3 ± 3.9	0.80 ± 0.04	37.7 ± 1.8	17.1 ± 0.7	22/130
Esclavos	126.8 ± 2.7	15.3 ± 0.3	37.2 ± 0.6	2.48 ± 0.04	65.9 ± 3.3	0.80 ± 0.03	36.9 ± 1.5	17.0 ± 0.6	20/110
Hojancha	123.0 ± 2.3	16.6 ± 0.3	43.0 ± 0.7	2.63 ± 0.05	64.1 ± 2.7	0.89 ± 0.03	42.5 ± 1.5	18.2 ± 1.1	9/54
Jiménez	86.5 ± 5.0	9.4 ± 0.4	27.8 ± 1.1	3.04± 0.13	10.1 ± 1.0	0.31 ± 0.02	8.1 ± 2.6	5.1 ± 2.0	5/25
Cañas	97.5 ± 2.9	12.7 ± 0.4	32.4 ± 1.1	2.63 ± 0.13	50.9 ± 0.8	0.68 ± 0.02	24.4 ± 1.7	14.5 ± 1.2	6/30
La Paz	120.4 ± 2.9	14.5 ± 0.4	38.4 ± 0.8	2.71 ± 0.07	70.3 ± 7.3	0.89 ± 0.08	38.1 ± 2.9	16.2 ± 0.8	13/61
Pacífico Sur	116.6 ± 2.6	11.7 ± 0.2	32.2 ± 0.3	2.82 ± 0.03	12.9 ± 0.7	0.32 ± 0.02	8.1 ± 2.2	3.2 ± 1.4	19/114
San Carlos	77.0 ± 1.5	7.6 ± 0.2	24.2 ± 0.4	3.25 ± 0.07	9.0 ± 0.9	0.23 ± 0.01	8.9 ± 5.9	7.1 ± 1.9	15/85
Talamanca	58.8 ± 3.3	8.7 ± 0.5	28.1 ± 1.5	3.41 ± 0.13	8.4 ± 0.5	0.25 ± 0.02	7.3 ± 5.5	7.3 ± 2.8	4/24
Upala	111.8 ± 1.9	11.6 ± 0.2	34.2 ± 0.4	3.11 ± 0.07	17.8 ± 13	0.40 ± 0.02	14.6 ± 1.7	9.4 ± 4.6	19/111
Xpujil	119.0 ± 1.7	14.7 ± 0.2	36.9 ± 0.4	2.55 ± 0.03	73.7 ± 4.1	1.04 ± 0.06	39.9 ± 2.1	19.3 ± 1.0	22/132
Yucatán	124.4 ± 1.9	14.6 ± 0.2	34.0 ± 0.5	2.36 ± 0.03	52.7 ± 5.4	0.80 ± 0.04	35.0 ± 2.8	15.8 ± 0.6	13/74