

# GENETIC STRUCTURE AND GENETIC DIVERSITY OF *SWIETENIA MACROPHYLLA* (MELIACEAE): IMPLICATIONS FOR SUSTAINABLE FOREST MANAGEMENT IN MEXICO

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Received April 2013

**ALCALÁ RE, SALAZAR H, GUTIÉRREZ-GRANADOS G & SNOOK LK. 2014. Genetic structure and genetic diversity of *Swietenia macrophylla* (Meliaceae): implications for sustainable forest management in Mexico.** The genetic structure and genetic diversity of Mexican populations of *Swietenia macrophylla* were evaluated. In Mexico, this neotropical tree reaches its northernmost distribution limit. The relationship between genetic diversity and geographic position (longitude, latitude) of four populations was described. The mean genetic and pairwise genetic differentiations were estimated to study the geographic pattern in the genetic structure. The mean overall values for observed heterozygosity ( $H_O$ ), expected heterozygosity ( $H_E$ ) and fixation coefficient ( $F_{IS}$ ) were 0.41, 0.71 and 0.449 respectively. The mean values per population of  $H_O$ ,  $H_E$  and  $F_{IS}$  correlated with latitude only. The genetic differentiation between populations revealed by the coefficient of differentiation ( $F_{ST} = 0.047$ ) was statistically significant ( $p = 0.0001$ ). Unweighted pair group method analysis showed that the extent of pairwise genetic differentiation increased with latitudinal position of populations, although no isolation by distance was observed ( $r = 0.76$ ,  $p = 0.15$ ). Results were discussed in the context of the marginal distribution of Mexican populations. Implications of the results on the conservation of *S. macrophylla*, particularly for populations located in the Mayan zone were discussed.

Keywords: Big-leaf mahogany, microsatellite loci, peripheral populations, marginal populations, tropical timber

**ALCALÁ RE, SALAZAR H, GUTIÉRREZ-GRANADOS G & SNOOK LK. 2014. Struktur genetik dan kepelbagaian genetik *Swietenia macrophylla* (Meliaceae): implikasi terhadap pengurusan hutan secara mampan di Mexico.** Struktur genetik dan kepelbagaian genetik populasi *Swietenia macrophylla* di Mexico dinilai. Di Mexico, pokok neotropika ini tumbuh sehingga ke had taburan yang paling utara. Hubungan antara kepelbagaian genetik dengan kedudukan geografi (longitud, latitud) empat populasi diuraikan. Purata pembezaan genetik dan pembezaan genetik berpasangan dianggar bagi mengkaji corak geografi dalam struktur genetik. Purata nilai keseluruhan bagi heterozigos yang dicerap ( $H_O$ ), heterozigos yang dijangka ( $H_E$ ) dan pekali penetapan ( $F_{IS}$ ) ialah masing-masing 0.41, 0.71 dan 0.449. Purata  $H_O$ ,  $H_E$  dan  $F_{IS}$  setiap populasi berkorelasi dengan latitud sahaja. Pembezaan genetik antara populasi seperti yang ditunjukkan oleh pekali pembezaan ( $F_{ST} = 0.0047$ ) adalah signifikan ( $p = 0.0001$ ). Analisis kaedah kumpulan berpasangan tidak berpemberat menunjukkan yang pembezaan genetik berpasangan meningkat dengan kedudukan latitud populasi walaupun tidak terdapat pengasingan dari segi jarak ( $r = 0.76$ ,  $p = 0.15$ ). Keputusan dibincangkan dari konteks taburan pinggiran populasi di Mexico. Implikasi kajian terhadap pemuliharaan *S. macrophylla* dibincangkan terutamanya untuk populasi yang terletak di zon Maya.

## INTRODUCTION

The genetic structure of a species is the result of ecological or historical factors that have shaped the relative effects of natural selection, gene flow, genetic drift and mutation. The roles that these evolutionary processes play on populations

determine the extent of genetic variation maintained within populations and genetic differentiation among them (Avice 2000, Nybom 2004). Molecular markers represent a powerful tool to infer, through the analysis of nuclear

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and cytoplasmic DNA variation, the genetic structure and different aspects of the history and evolutionary ecology of species. This knowledge is relevant within the framework of conservation genetics because understanding the evolutionary component of populations is recognised as a key factor to identify potential threats to their genetic diversity or to propose actions that can contribute to species conservation (Allendorf & Luikart 2007, Frankham et al. 2010).

In comparison with temperate woody plants, tropical trees are expected to be particularly susceptible to changes in habitat characteristics because of their low population density, auto-incompatible breeding systems and dependence on animal interactions for pollination and seed dispersal (Lowe et al. 2005, Dick et al. 2008). In addition, because some tropical species are valuable, they are subjected to selective extraction of individuals. This consequently diminishes the population density and increases the distance between reproductive individuals (Obayashi et al. 2002, Sebbenn et al. 2008). Populations under these conditions are expected to be more exposed to processes that promote genetic differentiation and reduce genetic diversity (Young et al. 1996). Therefore, knowledge about factors that have shaped the pattern of gene flow and that consequently explain the genetic differentiation and intra-population genetic diversity is useful to design or modify strategies to conserve harvested species (Lowe et al. 2005, Navarro et al. 2010).

Big-leaf mahogany (*Swietenia macrophylla*, Meliaceae) extends from southern Mexico to

the Amazon basin in Brazil (Pennington 1981). It is a monoecious, protogynous, emergent light-demanding tree (ca. 40 m in height) with low population density of about one adult reproductive tree per ha. *Swietenia macrophylla* has small flowers that seem to be visited by generalist insects (Styles 1972). The seeds are winged and dispersed by wind. Median dispersal distance is around 30 m (Grogan & Galvao 2006). In the context of conservation of genetic diversity of tropical trees, big-leaf mahogany is an interesting case. It has been the most important neotropical timber species traded for centuries (Snook 1998). As a consequence of that trade, it was listed in 2002 on Appendix II of the Convention on International Trade of Endangered Species of Fauna and Flora (CITES). Mahogany is probably the most studied timber species with regard to genetics with studies covering populations from most of its geographic range (Lemes et al. 2003, Novick et al. 2003, Navarro et al. 2010). As a result, estimates of the extent of genetic differentiation and genetic diversity are available for different ecological conditions and at contrasting geographic scales (Table 1). However, information for Mexican populations is scarce, considering that in Mexico big-leaf mahogany reaches the northernmost limit of its geographic distribution and may reflect different dynamics with regard to central populations (Lesica & Allendorf 1995).

The isolation by distance model of genetic structuring of populations predicts a tendency in which genetic similarity between populations decreases with increased geographic separation

**Table 1** Sampling conditions and microsatellite-derived genetic information in *Swietenia* spp.

Reference	Geographic range (km)	Population sampled	Sample size	Loci	$F_{ST}/R_{ST}$	$H_O$	$H_E$	$F_{IS}$	A
Céspedes et al. (2003)	100	1 (5 plots)	20	5	$F_{ST} = 0.063$	0.45–0.58	0.47–0.54	-0.070–0.050	2.8–4.0
Lemes et al. (2003)	2100	7	24–30	8	$F_{ST} = 0.091$ $R_{ST} = 0.144$	0.68–0.81	0.75–0.81	-0.004–0.100	7.6–10.7
Lowe et al. (2003)	5	3	27–53	3	$F_{ST} = 0.240$	0.14–0.52	0.30–0.58	0.100–0.530	2.6–5.0
Novick et al. (2003)	1600	8	16–55	7	$F_{ST} = 0.109$ $R_{ST} = 0.177$	0.47–0.67	0.59–0.68	0.030–0.247	6.4–8.8
White et al. (1999)	5	1 (3 sites)	22–97	10	$R_{ST} = 0.032$	0.47–0.49	0.50–0.54	0.190–0.210	4.2–9.5

$F_{ST}$  or  $R_{ST}$  = measures of the overall genetic differentiation,  $H_O$  = mean observed heterozygosity,  $H_E$  = mean expected heterozygosity,  $F_{IS}$  = mean fixation coefficient, A = mean number of alleles

as the homogenising influence of gene flow diminishes (Wright 1943). In this study, from the analysis of possible correlations of the level of within-population genetic diversity with geographic variables and the pattern of pairwise genetic differentiation, we concluded that factors other than isolation by distance could have played a role in the genetic structure and levels of genetic diversity of Mexican populations of *S. macrophylla*.

We also related the information from our study to the conservation of populations of *S. macrophylla* in Mexico, particularly those located in the Mayan zone. That region harbours extensive tropical forests that are logged under a management plan (Bray et al. 2003). Ecological information on factors that affect natural regeneration and the potential for sustainable logging has increased recently (Cámara-Cabral & Kelty 2009, Gutiérrez-Granados et al. 2011, Negreros-Castillo & Mize 2011). However, the issue of maintaining genetic diversity has not yet been considered in local management plans.

## MATERIALS AND METHODS

### Study sites

From a database provided by the Comisión Nacional para el uso y manejo de la Biodiversidad (CONABIO), which contains records of collections done between 1934 and 2004 from at least 10 different herbaria, we selected seven

sampling locations of *S. macrophylla* that we thought would be informative for characterisation of the genetic structure of Mexican populations. However, some of them were no longer extant, for example in Yucatan and Tabasco. We used four populations, adhering to the original sampling criteria. Populations of *S. macrophylla* covering the maximum geographic separation (ca. 1000 km) were located in different regions of the Yucatan Peninsula and included the northernmost location in Mexico (Figure 1).

In Campeche, trees were sampled 13 km north-east of Nuevo Becal, a zone containing medium-height semi-evergreen tropical forest in the municipality of Calakmul (Martínez & Galindo-Leal 2002). In Quintana Roo, trees were sampled in Naranjal Poniente (hereafter Naranjal), a town located to the west of Felipe Carrillo Puerto. The trees were sampled in a zone set aside as a local reserve for seed collection, also within a medium-height semi-evergreen tropical forest. In this stand, normal diameter (1.3 m) of mahogany trees exceeds the minimum cutting diameter of 55 cm. In Chiapas, trees were sampled within the influence zone of the Montes Azules Biosphere Reserve at 1 km south-east of the Lacandon community of Bonampak, near the archaeological monuments of Bonampak. The vegetation is characterised as tropical rain forest (Meave del Castillo 1990). In this area, some individuals with normal diameter of > 70 cm could be found. The fourth population was located in Zozocolco (44 km south-east of



**Figure 1** Location of the four populations of *Swietenia macrophylla* sampled in the states of Campeche (Nuevo Becal), Chiapas (Bonampak), Quintana Roo (Naranjal) and Veracruz (Zozocolco), Mexico

the city of Papantla), in the northern region of the state of Veracruz. In this zone, forests had been transformed into pastures for cattle raising. Forested areas were located on inaccessible sites such as hill tops or ravines. Here, large-diameter individuals of *S. macrophylla* were located exclusively in private lands.

On each stand, most trees in the study were separated by a minimum distance of 100 m to reduce the likelihood of collecting closely-related individuals. Leaf samples were collected from 18 to 33 trees per population using a slingshot and an extendable pruner to collect materials from the lower and the higher parts of the crown. After inspection, only leaves free of apparent damage by pathogens and fungi were selected. For each *S. macrophylla* tree, material taken from about 10 different sampled leaves were mixed and kept individually in sealed plastic bags containing silica gel to dry the tissue and stabilise DNA.

### DNA procedures

In the laboratory, DNA was isolated using a standard cetyl trimethyl ammonium bromide-based procedure (Doyle & Doyle 1987). The quality of samples was improved using GeneClean®. Genetic structure was tested using eight microsatellite primers developed previously for *S. macrophylla* (Lemes et al. 2002). For all primers, a series of factorial trials was run in which the variation of the concentration of primers (0.5–2.0 mM), MgCl<sub>2</sub> (1–4 mM), template DNA (0.25–5.0 ng) and TAQ polymerase (0.5–1.5 U) was tested. Bright and reproducible bands were obtained with only four primers: sm01, sm31, sm47 and sm51. The final reaction consisted of 15 mL total volume containing 1 × polymerase chain reaction buffer, 1 mM MgCl<sub>2</sub>, 0.2 mM dNTP's, 2 μM of primer, 1 u Taq DNA polymerase and 5 ng DNA. Amplification conditions for primer sm47 were an initial denaturing step of 96 °C for 1 min followed by 30 cycles of 96 °C for 1 min, 56 °C for 1 min, 72 °C for 1 min and a final extension step of 72 °C for 7 min. For the other three primers, the conditions were an initial step of 94 °C for 3 min followed by the same cycles. Polymerase chain reaction products were subjected to electrophoresis in 6% polyacrilamide gels in tris-borate ethylenediaminetetraacetic buffer and stained with ethidium bromide. In each gel, a 20-pair base DNA ladder was included. The different alleles for each locus were scored

according to their molecular mass (White et al. 2002, Céspedes et al. 2003). The most anodal allele was arbitrarily identified as 1 and the remaining alleles were identified sequentially (Céspedes et al. 2003).

### Genetic analysis

The genetic diversity was described using mean number of alleles (A) as well as observed (H<sub>O</sub>) and expected heterozygosity (H<sub>E</sub>) values. Departures from Hardy–Weinberg equilibrium were tested by the fixation coefficient (F<sub>IS</sub>) using Fisher's exact test. The significance of F<sub>IS</sub> was estimated using Monte Carlo chain with 1000 iterations (Excoffier et al. 2005). To evaluate whether genetic diversity revealed a geographic pattern, independent Spearman rank correlation analyses were performed. The mean values per population of A, H<sub>O</sub>, H<sub>E</sub> and F<sub>IS</sub> were used against latitude and longitude.

The genetic structure of *S. macrophylla* was estimated using analysis of molecular variance (AMOVA) with the software Arlequin version 3 (Excoffier et al. 2005) that was used to extract the variance components corresponding to the variation retained within and among populations. In the model, the AMOVA estimates the percentage from the total genetic variance that is explained by difference among populations while the rest represents the percentage of genetic variance attributable to the inter-individual differences within populations. From the variance component corresponding to the among population level, F<sub>ST</sub> (an analogue of F<sub>ST</sub> that measures the correlation between two gametes drawn at random from each subpopulation as a measurement of the degree of gene differentiation) was estimated. The genetic structure was calculated under the assumption of the infinite allele model. The significance of F<sub>ST</sub> was obtained through Monte Carlo approach in which individuals were randomly reassigned among populations with 1000 replicates. Within the genetic structure tools implemented in AMOVA, we calculated the Nei's D, which estimated the average number of substitutions that had taken place since two populations shared their last common ancestor. Therefore, Nei's D was used to explore the extent of pairwise genetic differentiation and genetic relationships among populations, as recommended when divergences were recent

(Takezaki & Nei 1996). The magnitude and significance of the correlation coefficient ( $r$ ) between geographic and genetic distances were evaluated by Mantel test. This test evaluates whether genetic differentiation is explained by the geographic separation of populations. In this case, only the overland distance between populations was considered.

## RESULTS

### Within population genetic diversity

The allele richness in Naranjal, Bonampak and Nuevo Becal ranged from 7.5 to 8.0 (Table 2). In contrast, the allele richness in Zozocolco was 6.25. The mean observed heterozygosity ( $H_O$ ) in Bonampak and Zozocolco showed the highest (0.52) and lowest (0.27) values respectively. The mean value of  $H_O$  was around half that of  $H_E$ .

Mean values of  $H_O$  and  $H_E$  were negatively correlated with geographic latitude (Figure 2). In contrast,  $F_{IS}$  values were positively correlated with latitude. There was no association between the four genetic variables with longitude (results not shown).

### Genetic differentiation

The AMOVA revealed significant overall genetic differentiation between populations of *S. macrophylla*. Of the total molecular variance, 4.68% ( $p = 0.0001$ ) was due to differences between populations (Table 3). This value corresponded to  $F_{ST}$  of 0.047. In the dendrogram, populations of Bonampak and Nuevo Becal showed the highest genetic similitude (Figure 3). Naranjal was added to this group whereas Zozocolco showed the highest genetic

differentiation. Although the amount of pairwise genetic differentiation varied by one order of magnitude, there was no correlation between geographic distance and population ( $r = 0.76$ ,  $p = 0.15$ ; results not shown).

## DISCUSSION

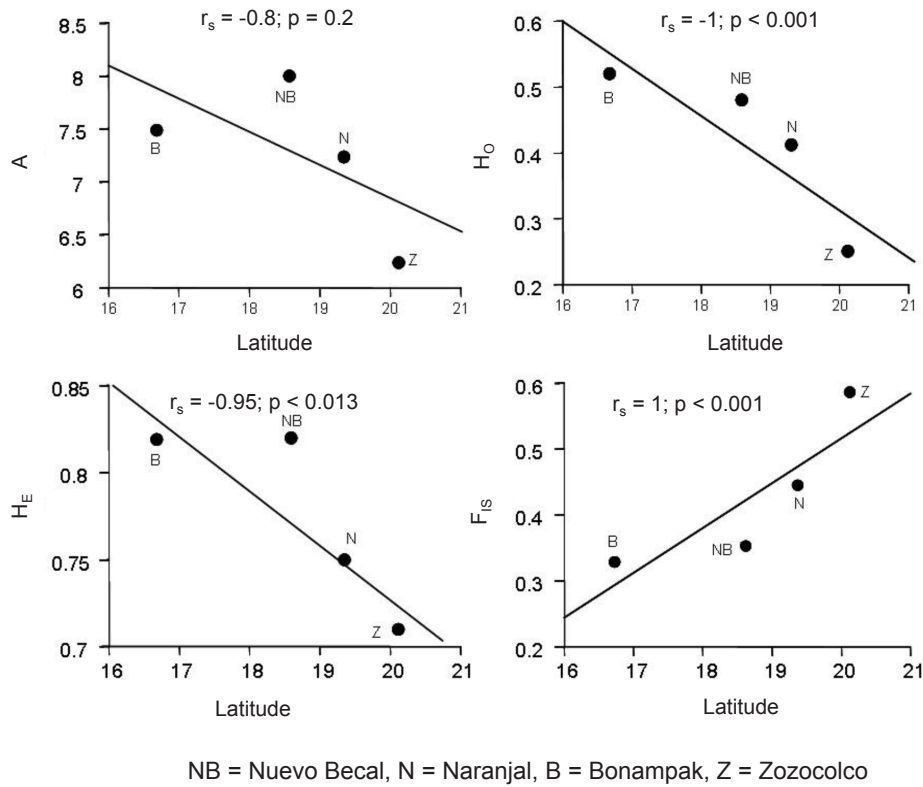
The microsatellite-derived level of within-population genetic diversity of *S. macrophylla* is highly variable along its geographic distribution. For Mesoamerican populations, for example, the observed heterozygosity ( $H_O$ ) ranged from 0.45 to 0.67 (Table 1). Only one Costa Rican population deviated from this range, exhibiting a very low value ( $H_O = 0.14$ ). Comparatively, Mexican populations maintained low levels of genetic diversity as the mean observed heterozygosity per population was  $H_O = 0.41$ . Mexican populations also presented a high deficiency of heterozygotes ( $F_{IS} = 0.449$ ) that exceeded the scores observed for most tropical trees (Ward et al. 2005) and those published and reported in Table 1 for *S. macrophylla* in Amazonian populations (-0.004–0.100) and Mesoamerica (0.030–0.247) (Table 1). A possible loss of information due to the genotyping procedure and the reduced number of primers used in this study could partially account for the low mean genetic diversity measured in Mexican populations. However, these low mean values and other genetic results derived from our work are better explained considering the location of populations of *S. macrophylla* at the northern portion of the species distribution.

We first explored the effect of anthropogenic disturbance as it has been reported as a main factor affecting the mean values of the within-population genetic diversity. At a local scale and

**Table 2** Geographic position and genetic diversity averaged over loci in four Mexican populations of *Swietenia macrophylla*

Population	Sample size	Latitude (N)	Longitude (W)	Altitude (m)	A	$H_O$	$H_E$	$F_{IS}$
Bonampak	18	16° 45' 07"	91° 07' 19"	223	7.75	0.52	0.83	0.333*
Naranjal	23	19° 21' 25"	88° 27' 28"	30	7.50	0.39	0.75	0.443*
Nuevo Becal	20	18° 36' 36"	89° 18' 07"	220	8.00	0.47	0.82	0.356*
Zozocolco	33	20° 08' 01"	97° 34' 45"	250	6.25	0.27	0.71	0.587*
Mean	–	–	–	–	7.38	0.41	0.78	0.449*

A = mean number of alleles,  $H_O$  = mean observed heterozygosity,  $H_E$  = mean expected heterozygosity,  $F_{IS}$  = mean genotypic deviation; \* $p < 0.05$



**Figure 2** Spearman rank correlation analyses between geographic latitude and the number of alleles (A), observed heterozygosity ( $H_o$ ), expected heterozygosity ( $H_E$ ) and fixation coefficients ( $F_{IS}$ )

**Table 3** Analysis of molecular variance (AMOVA) performed on the four populations of *Swietenia macrophylla*

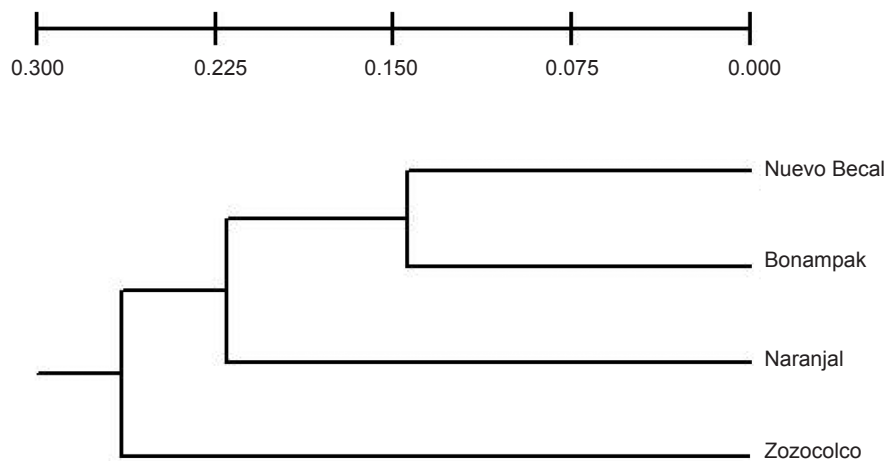
Source of variation	Df	Sum of square	Variance component	% of variance	$F_{ST}$	p
Between population	3	12.91	0.06	4.68	0.0468	0.0001
Within population	184	242.86	1.31	95.32		
Total	187	255.78	1.38			

Df = degree of freedom,  $F_{ST}$  = coefficient of genetic differentiation, probability

specifically for *S. macrophylla*, selective logging (André et al. 2008) and habitat fragmentation (White et al. 1999) have been reported to reduce the within-population genetic diversity. At a regional level, the genetic diversity found among 20 Mesoamerican populations of *S. macrophylla* was negatively correlated with the extent of human disturbance (Gillies et al. 1999). This pattern could explain our results, as in this study the highest heterozygosis was found in the pristine conditions of Bonampak, whereas the lowest occurred at Zozocolco, a heavily deforested area. However, high levels of

genetic diversity derived from random amplified polymorphic DNA (Gillies et al. 1999) and microsatellite loci (Novick et al. 2003) have been reported in extremely disturbed populations. These inconsistencies suggest that factors other than anthropogenic disturbance can better explain the variation in genetic diversity.

On the other hand, the relationships between genetic diversity or the fixation coefficient with latitude (Figure 2) could reflect the history of colonisation of *S. macrophylla* in Mexico. Firstly, for some long-lived perennial species, it has been shown that the within-population genetic



**Figure 3** Dendrogram constructed with Nei's unbiased genetic distances through the unweighted pair group method with arithmetic mean algorithm for four Mexican populations of *Swietenia macrophylla*

diversity frequently correlates with geographic position of populations (latitude or longitude). This correlation often reveals differences between central and peripheral populations that are explained mainly by biogeographical factors (Maizura et al. 2006, Mimura & Aitken 2007).

Secondly, in this study, the mean values of  $H_O$  and  $H_E$  were negatively correlated with latitude but were not correlated with longitude. This is interesting considering that geographic separation along the longitudinal axis is twice as great as that registered on the latitudinal axis, which spans only  $4^\circ$  (Figure 2, Table 2). This indicates that the geographic distance itself (which is more apparent from the longitudinal variation) does not account for the geographic variation in genetic diversity in Mexican populations. In contrast, the observed effect of genetic diversity being affected only by the increase of distance towards the north seems compatible with the peripheral condition of these populations. We based this argument on the evidence available at a regional level, namely, the negative relationship between the within-population genetic diversity with latitude in *S. macrophylla*. For example, Gillies et al. (1999) reported a negative slope for the relationship between both variables among the 20 Mesoamerican populations. The lower microsatellite-based mean genetic diversity in Mesoamerican populations with regard to those located on the Amazon Basin has been interpreted as a result of the colonisation of *S. macrophylla* towards the northern latitudes from South America (Lemes et al. 2003, Novick et al. 2003, Lemes et al. 2010).

Thirdly, the mean values for the excess of homozygotes per population ( $F_{IS}$ ) were positively correlated with the latitudinal position of the four populations sampled in Mexico. Null alleles are an unlikely explanation because there is no theoretical expectation that the occurrence of null alleles would reflect a latitudinal pattern. Higher levels of selfing could explain high positive  $F_{IS}$  scores (Busch 2005, Mimura & Aitken 2007). However, high outcrossing rates have been reported in a remnant logged population of *S. macrophylla*, suggesting a high tolerance to disturbance conditions (Lemes et al. 2007). We suggest that the excess of homozygotes in the Mexican populations has resulted from a high rate of mating between close relatives, as inbreeding has been reported in some other populations (Table 1). We considered that the positive correlation between  $F_{IS}$  and latitude reflected, at a shorter geographic scale, that proposed for Mesoamerican populations of *S. macrophylla*, as they were expected to evolve under high levels of biparental inbreeding due to their expansion from South America (Lemes et al. 2010).

Our results on the genetic differentiation between the four populations seem to be compatible with the history of colonisation of *S. macrophylla* (Gillies et al. 1999, Novick et al. 2003). For example, with regard to other Mesoamerican populations and according to some unweighted pair group method analyses (UPGMA), Mexican populations of *S. macrophylla* are genetically closer to populations of northern Central America than to populations located in Honduras, Nicaragua,

Costa Rica and Panama. In fact, the low genetic distances within the northern group (that included a Mexican population) suggest a recent evolution. With less time to diverge, a low genetic differentiation could be expected (Novick et al. 2003). In this context, the genetic differentiation of Mexican populations found in this study ( $F_{ST} = 0.047$ ) was low considering their geographic separation (ca. 1000 km) compared with the genetic differentiation found between populations separated by shorter distances (Table 1).

In addition, Mexican populations appeared genetically differentiated even from closer populations located in Guatemala and Belize although more clearly for RAPD markers (Gillies et al. 1999) than for microsatellites (Novick et al. 2003). This genetic distinction suggests that evolution in the northernmost limit of the species geographic distribution has been somewhat independent. Interestingly, the analysis of the genetic relationships among Mexican populations (UPGMA) echoes this pattern at a smaller geographic scale. Populations situated at northern locations (Zozocolco and Naranja) showed the highest genetic differentiation, whereas southern populations located in Campeche and Chiapas maintained a higher genetic similitude. It should be noted that this pattern of pairwise genetic differentiation was not explained by the geographical separation between populations, as the test for isolation by distance was not statistically significant. Although a low number of populations sampled could explain the absence of relationship between genetic and geographic distances, geographic distance itself seemed not to be the cause of genetic differentiation as we found that the amount of genetic differentiation increased towards higher latitudes.

Overall, the low mean genetic differentiation, pairwise genetic differentiation (higher genetic differentiation towards higher latitudes in the absence of isolation by distance) and correlation of genetic diversity and fixation coefficient with latitude but not with longitude were compatible with available information. These indicated that populations of *S. macrophylla* located at higher latitudes could have been more recently founded. Our results indicated that only the increase in distance in a northern direction affected the genetic diversity and pairwise genetic differentiation in Mexican populations of *S. macrophylla*.

## Conservation considerations

The current management programme for *S. macrophylla* in Mexico was implemented in the 1980s in the states of Campeche and Quintana Roo where extensive forests still maintain populations of commercial sizes. The programme was thought to guarantee sustainability of this species because selective logging was based on a minimum cutting diameter (Bray et al. 2003). In particular, with regard to the Mayan zone located in central Quintana Roo, different ecological studies have focused on understanding factors that affect natural regeneration in harvested populations. There is evidence of negative effects of the current management programme on population dynamics. Therefore, recommendations such as increase of minimum cutting diameter, clearing treatments, use of plantations and inclusion of old large trees with no commercial value as seed sources have been made (Cámara-Cabrales & Kely 2009, Gutiérrez-Granados et al. 2011, Negreros-Castillo & Mize 2011).

In contrast, there is no direct evaluation of the effects of selective logging on genetic parameters. However, it can be expected that Mexican populations present negative effects as a result of harvesting. For example, populations of tropical trees subjected to selective logging frequently present lower outcrossing rates and allelic richness and higher inbreeding coefficients in comparison with unmanaged populations (Degen et al. 2006, Silva et al. 2008). In this context, our results are important as selective logging can have higher negative impact than expected. This is because historically Mexican populations of *S. macrophylla* seem to maintain lower genetic diversity and higher positive fixation coefficients compared with other Mesoamerican populations. Under this more limited scenario, we propose several recommendations that can be considered in the current management programme in order to increase the sustainability of harvests of *S. macrophylla*.

First, theoretically the effective population size is inversely related to the per generation rate of loss of genetic diversity produced by random fixation of alleles (Frankham et al. 2010). Effective population size in populations within the Mayan zone can be lower. This is because although the reproductive potential of big-leaf mahogany is positively correlated



with tree size (Snook et al. 2005), most of reproductive individuals are removed by selective logging. In addition, the high variance in the number of propagules produced per parent across generations also diminishes effective size (Hedrick 2000). Mahogany trees reveal a high inter-annual variation in seed production (Snook et al. 2005). Consequently, in future, the current diameter cutting limit could accelerate the loss of genetic diversity. Management of *S. macrophylla* should promote practices that allow more individuals of *S. macrophylla* to contribute to the gene pool. The genetic information derived from this study supports the recommendation by Snook et al. (2005) to leave standing some mahogany trees of 75 cm diameter or larger as seed trees in managed forests, rather than harvesting all individuals greater than 55 cm.

Second, if seed collection and planting is considered as a strategy to retain higher genetic diversity, efforts should be made to balance the number of seeds collected per tree and the number of seed trees. The common practice of collecting hundreds of seeds only from a few individuals (Santos Jimenez et al. 2005) should be avoided and more adult trees should be included. As a complement, separation between trees should be widely extended to reduce sampling within genetic families as we found high  $F_{IS}$  even when sampled individuals were separated by at least 100 m.

Third, provenance tests to guide seed collection based on the local adaptation can be particularly useful under a scenario of climatic change (Navarro et al. 2010). In the case of Mexico, the available information indicates no evidence of local adaptive differences across populations of *S. macrophylla* within the Yucatan Peninsula (i.e. Campeche and Quintana Roo). This is probably because of the homogeneous climate at the geographic scale evaluated (Wightman et al. 2008). However, differences could appear if individuals of other populations were tested such as those located in Chiapas inhabiting shadier and wetter conditions or those in north Veracruz that were exposed to the highest isolation. In the absence of this information, although dealing with neutral variation (which is theoretically little or not affected by natural selection), our study could guide programmes of seed exchange. This is because neutral variation frequently reveals the

history of gene flow. Therefore, pairwise genetic differentiation suggests that seed exchange should follow a geographic pattern. Northern populations of *S. macrophylla* along the Gulf of Mexico (i.e. Veracruz) are more likely to have been derived from southern populations in Campeche and Chiapas than from those of Quintana Roo. Conversely, restoration of stands located in the state of Yucatan should be based on seeds from the northern Mayan region.

Finally, peripheral or marginal populations of widely distributed species tend to inhabit more stressful habitats and be more exposed to higher inbreeding conditions (Hampe & Petit 2005). In spite of their lower genetic diversity and higher fixation coefficients, northern populations of *S. macrophylla* located in Veracruz or within the Yucatan peninsula could be important for conservation. This is because of their possible adaptation to drier and warmer conditions that could change the future pattern of distribution of Mexican tropical forest.

## ACKNOWLEDGEMENTS

The authors thank LY Bustos, S de la Cruz and V Vargas for help in collecting tissues. The Laboratory of Molecular Systematics and Evolution provided facilities for the isolation and amplification of DNA samples. The authors thank the Organización de Ejidos Productores Forestales de la Zona Maya, SC for support and the local government for facilities to access the sites. The study was funded by the Comisión Nacional para el Uso y Manejo de la Biodiversidad (CONABIO).

## REFERENCES

- ALLENDORF FW & LUIKART G. 2007. *Conservation and the Genetics of Populations*. Blackwell Publishing, Malden.
- ANDRÉ T, LEMES, MR, GROGRAN J & GRIBEL R. 2008. Post-logging loss of genetic diversity in a mahogany (Meliaceae) population in Brazilian Amazonia. *Forest Ecology and Management* 255: 340–345.
- AVISE JC. 2000. *Phylogeography: the History and Formation of Species*. Harvard University Press, Cambridge.
- BRAY DB, MERINO-PÉREZ L, NEGREROS-CASTILLO P, SEGURA-WARNHOLTZ G, TORRES-ROJO JM & VESTER HFM. 2003. Mexico's community-managed forests as a global model for sustainable landscapes. *Conservation Biology* 17: 672–677.
- BUSCH JW. 2005. The evolution of self-compatibility in geographically peripheral population of *Leavenworthia alabamica* (Brassicaceae). *American Journal of Botany* 92: 1503–1512.

- CÁMARA-CABRALES L & KELTY MJ. 2009. Seed dispersal of big-leaf mahogany (*Swietenia macrophylla*) and its role in natural forest management in the Yucatán Peninsula, Mexico. *Journal of Tropical Forest Science* 21: 235–245.
- CÉSPEDES M, GUTIÉRREZ MV, HOLBROOK NM & ROCHA OJ. 2003. Restoration of genetic diversity in the dry forest tree *Swietenia macrophylla* (Meliaceae) after pasture abandonment in Costa Rica. *Molecular Ecology* 12: 3201–3112.
- DEGEN B, BLANC L, CARON H, MAGGIE L, KREMER A & GOURLET-FLEURY S. 2006. Impact of selective logging on genetic composition and demographic structure of four tropical tree species. *Biological Conservation* 131: 386–401.
- DICK CW, HARDY OJ, JONES FA & PETIT RJ. 2008. Spatial scales of pollen and seed-mediated gene flow in tropical rain forest trees. *Tropical Plant Biology* 1: 20–33.
- DOYLE JJ & DOYLE JL. 1987. Isolation of plant DNA from fresh tissue. *Focus* 12: 13–15.
- EXCOFFIER L, LAVAL G & SCHNEIDER S. 2005. Arlequin ver. 3.0: An integrated software package for population genetics data analysis. *Evolutionary Bioinformatics Online* 1: 47–50.
- FRANKHAM R, BALLOU JD & BRISCOE DA. 2010. *Introduction to Conservation Genetics*. Second edition. Cambridge University Press, Cambridge.
- GILLIES ACM, NAVARRO C, LOWE AJ, NEWTON AC, HERNÁNDEZ M, WILSON J & CORNELIUS JP. 1999. Genetic diversity in Mesoamerican populations of mahogany (*Swietenia macrophylla*), assessed using RAPDs. *Heredity* 83: 722–732.
- GROGAN J & GALVAO J. 2006. Factors limiting post-logging seedling regeneration by big-leaf mahogany (*Swietenia macrophylla*) in southeastern Amazonia, Brazil, and implications for sustainable management. *Biotropica* 38: 219–228.
- GUTIÉRREZ-GRANADOS G, JUÁREZ V & ALCALÁ RE. 2011. Natural and human disturbances affect natural regeneration of *Swietenia macrophylla*: implications for rainforest management. *Forest Ecology and Management* 262: 161–169.
- HAMPE A & PETIT RJ. 2005. Conserving biodiversity under climate change: the rear edge matters. *Ecology Letters* 8: 461–467.
- HEDRICK PW. 2000. *Genetics of Populations*. Second edition. Jones and Barlett Publishers, Sudbury.
- LEMES MR, BRONDANI RPV & GRATTAPAGLIA D. 2002. Multiplexed systems of microsatellite markers for genetic analysis in mahogany, *Swietenia macrophylla* King (Meliaceae), a threatened neotropical timber species. *Journal of Heredity* 93: 287–290.
- LEMES MR, DICK CW, NAVARRO C, LOWE AJ, CAVERS SA & GRIBEL R. 2010. Chloroplast DNA microsatellites reveal contrasting phylogeographic structure in mahogany (*Swietenia macrophylla* King, Meliaceae) from Amazonia and Central America. *Tropical Plant Biology* 3: 40–49.
- LEMES MR, GRATTAPAGLIA D, GROGAN J, PROCTOR J & GRIBEL R. 2007. Flexible mating system in a logged population of *Swietenia macrophylla* King (Meliaceae), implications for the management of a threatened neotropical tree species. *Plant Ecology* 192: 169–179.
- LEMES MR, GRIBEL R, PROCTOR J & GRATTAPAGLIA D. 2003. Population genetic structure of mahogany (*Swietenia macrophylla* King, Meliaceae) across the Brazilian Amazon, based on variation at microsatellite loci, implications for conservation. *Molecular Ecology* 12: 2875–2883.
- LESICA P & ALLENDORF FW. 1995. When are peripheral populations valuable for conservation? *Conservation Biology* 9: 753–760.
- LOWE AJ, BOSHIER D, WARD M, BACLES CFE & NAVARRO C. 2005. Genetic resource impacts of habitat loss and degradation: reconciling empirical evidence and predicted theory for neotropical trees. *Heredity* 95: 255–273.
- LOWE AJ, JOURDE B, BREYNE P, COLPAERT N, NAVARRO C, WILSON J & CAVERS S. 2003. Fine-scale structure and gene flow within Costa Rican populations of mahogany (*Swietenia macrophylla*). *Heredity* 90: 268–275.
- MAIZURA I, RAJANAIDU N, ZAKRI AH & CHEAH SC. 2006. Assessment of genetic diversity in oil palm (*Elaeis guineensis* Jacq.) using restriction fragment length polymorphism (RFLP). *Genetic Resources and Crop Evolution* 53: 187–195.
- MARTÍNEZ E & GALINDO-LEAL C. 2002. La vegetación de Calakmul, Campeche: clasificación, descripción y distribución. *Boletín de la Sociedad Botánica de México* 71: 7–32.
- MEAVE DEL CASTILLO J. 1990. *Estructura y Composición de la Selva Alta Perennifolia de los Alrededores de Bonampak*. Colección Científica, Serie Arqueología. Instituto Nacional de Antropología e Historia, México.
- MIMURA M & AITKEN SN. 2007. Adaptive gradients and isolation by distance with postglacial migration in *Picea sitchensis*. *Heredity* 99: 224–232.
- NAVARRO C, BOSHIER D, CAVERS S & LOWE A. 2010. Genetic resources and conservation of mahogany in Mesoamerica. Pp 369–383 in Mery G et al. (eds) *Forests and Society—Responding to Global Drivers of Change*. IUFRO, Vienna.
- NEGREROS-CASTILLO P & MIZE CW. 2011. Soil–site preferences for mahogany (*Swietenia macrophylla* King) in the Yucatan Peninsula. *New Forests*. doi 10.1007/s11056-011-9303-7.
- NOVICK RR, DICK CW, LEMES MR, NAVARRO C, CACCONI A & BIRMINGHAM E. 2003. Genetic structure of Mesoamerican populations of big-leaf mahogany (*Swietenia macrophylla*) inferred from microsatellite analysis. *Molecular Ecology* 12: 2885–2893.
- NYBOM H. 2004. Comparison of different nuclear DNA markers for estimating intraspecific genetic diversity in plants. *Molecular Ecology* 13: 1143–1155.
- OBAYASHI K, TSUMURA Y, IHARA-UJINO T, NIYAMA K, TANOUCHI H, SUYAMA Y, WASHITANI I, LEE CT, LEE SL & MUHAMMAD N. 2002. Genetic diversity and outcrossing rate between undisturbed and selectively logged forests of *Shorea curtisii* (Dipterocarpaceae) using microsatellite DNA analysis. *International Journal of Plant Sciences* 163: 151–158.
- PENNINGTON TD. 1981. *Flora Neotropica: Meliaceae*. Monograph 28. New York Botanical Garden, New York.
- SANTOS JIMÉNEZ V, MAS KANTÚN P, LÓPEZ C, SNOOK L & ESCOBAR RUIZ C. 2005. El manejo forestal y la caoba en los ejidos de la zona maya, México. Desarrollo histórico, condiciones actuales y perspectivas. *Recursos Naturales y Ambiente* 44: 27–36.

- SEBBENN AM, DEGEN B, AZEVEDO VCR, SILVA M, LACERDA AB, CIAMPI AY, KANASHIRO M, CARNEIRO FS, THOMPSON I & LOVELESS MD. 2008. Modelling the long-term impact of selective logging on genetic diversity and demographic structure of four tropical tree species in the Amazon forest. *Forest Ecology and Management* 254: 335–349.
- SILVA MB, KANASHIRO K, CIAMPI AY, THOMPSON I & SEBBENN AM. 2008. Genetic effects of selective logging and pollen gene flow in a low-density population of the dioecious tropical tree *Bagassa guianensis* in the Brazilian Amazon. *Forest Ecology and Management* 255: 1548–1558.
- SNOOK LK. 1998. Sustaining harvest of mahogany (*Swietenia macrophylla* King) from Mexico's Yucatán forests: past, present and future. Pp 61–80 in Primack et al. (eds) *Timber, Tourists and Temples: Conservation and Development of Maya Forests of Belize, Guatemala and Mexico*. Island Press, Washington DC.
- SNOOK LK, CAMARA-CABRALES L & KELTY MJ. 2005. Six years of fruit production by mahogany trees (*Swietenia macrophylla* King): patterns of variation and implications for sustainability. *Forest Ecology and Management* 206: 221–235.
- STYLES BT. 1972. The flower biology of the Meliaceae and its bearing on tree breeding. *Silvae Genetica* 21: 175–182.
- TAKEZAKI N & NEI M. 1996. Genetic distances and reconstruction of phylogenetic trees from microsatellite DNA. *Genetics* 144: 389–399.
- WARD M, DICK CW, GRIBEL R, LEMES M, CARON H & LOWE AJ. 2005. To self, or not to self... a review of outcrossing and pollen-mediated gene flow in neotropical trees. *Heredity* 95: 246–254.
- WHITE G, BOSHIER DH & POWELL W. 1999. Genetic variation within a fragmented population of *Swietenia humilis* Zucc. *Molecular Ecology* 8: 899–1909.
- WHITE GM, BOSHIER DH & POWELL W. 2002. Increased pollen flow counteracts fragmentation in a tropical dry forest: an example from *Swietenia humilis* Zuccarini. *Proceedings of the National Academy of Sciences* 99: 2038–2042.
- WIGHTMAN KE, WARD SE, HAGGAR JP, RODRIGUEZ SB & CORNELIUS JP. 2008. Performance and genetic variation of big-leaf mahogany (*Swietenia macrophylla* King) in provenance and progeny trials in the Yucatan Peninsula of Mexico. *Forest Ecology and Management* 255: 346–355.
- WRIGHT S. 1943. Isolation by distance. *Genetics* 28: 114–138.
- YOUNG A, BOYLE T & BROWN AHD. 1996. The population genetic consequences of habitat fragmentation for plants. *Trends in Ecology and Evolution* 11: 413–418.