

## NOTES

### MATING SYSTEM IN A PLANTED POPULATION OF *SHOREA MACROPHYLLA* (DIPTEROCARPACEAE)

K. K. S. Ng, K. C. Ang & S. L. Lee

Forest Research Institute Malaysia, Kepong, 52109 Kuala Lumpur, Malaysia

The “Engkabang” tree, *Shorea macrophylla* (Dipterocarpaceae), is an endemic species on the island of Borneo (Newman *et al.* 1996). It is commonly abundant on periodically flooded alluvium and river banks but uncommon on hillsides, below 600 m. This species is a strong light demander and a very fast grower, reaching 50 to 60 cm diameter at breast height (dbh) in 20 to 30 years and regenerating profusely after 15 to 16 years in plantation (Appanah 1995).

The tree is easily distinguished by streams; leaves large, elliptic-oblong, base obtuse or subcordate, with 13 to 18 pairs of prominent, well-spaced nerves and hairy midrib above; petiole stout; bark smooth or shallowly scaly; bole short (Newman *et al.* 1996). The timber is categorised under the light red meranti group. The woods can be used for light construction work and the seeds, which contain oil, are used by the local people in Sarawak as medicine. Due to the medicinal value it has been commercialised and exported.

To date, no quantitative studies on the mating system of *S. macrophylla* has been reported. No information is available on the actual outcrossing rate of this tree under natural or planted condition and also on the extent of inbreeding occurring either through selfing or biparental mating.

Open-pollinated seeds (20 to 40 each tree) were collected from 14 single-progeny families from a planted *S. macrophylla* stand in Forest Research Institute Malaysia (FRIM), which was developed using seed sources from Borneo. The population area is about 5 ha and consists of approximately 150 adult trees. However, the detailed information of the seed sources being used to establish the stand was not available. The electrophoretic analysis was conducted in FRIM. Preparation of the material for electrophoresis and the buffer systems employed were as in Lee *et al.* (2000a). The four polymorphic loci that expressed well are Glucosphosphate isomerase (*Gpi*), Phosphoglucumutase (*Pgm*), Shikimate dehydrogenase (*Sdh*), and Uridine diphosphogluconate pyrophosphatase (*Ugph*).

The mixed-mating model of Ritland (1994) was used to determine the multilocus ( $t_m$ ) and single locus ( $t_s$ ) outcrossing rates, maternal genotypes as well as pollen and ovule allele frequencies. Further explanations of the model are in Ritland & Jain (1981) and Ritland (1983).

There was significant difference ( $p < 0.05$ ) in pollen and ovule allele frequencies for three loci (Table 1). The differences between the pollen and the ovule allelic frequencies can arise through differences in male and female representation in the progeny or through immigrant pollen from outside the area (Murawski & Hamrick 1992) or may be due to the existence of mating among relatives (Ritland & Jain 1981). These suggested that inbreeding can be attributed to biparental mating. Homogeneity of pollen pool gene frequencies over female parents (Table 2) indicated that maternal trees did not receive pollen at random from all synchronously flowering trees, but rather received a significant proportion of it from relatively few, possibly nearby individuals.

**Table 1** Maximum likelihood estimates of pollen and ovule allele frequencies and chi-square statistic to test the differences between pollen and ovule frequencies in the progeny of *Shorea macrophylla*

Locus	N	a	Pollen	Ovule	G <sub>st</sub> <sup>(a)</sup>	DF	χ <sup>2</sup> <sup>(b)</sup>
<i>Gpi</i>	411	1	0.592	0.759	0.0368	5	151.31*
		2	0.038	0.034			
		3	0.281	0.069			
		4	0.008	0.034			
		5	0.076	0.069			
		6	0.004	0.034			
<i>Pgm</i>	332	1	0.569	0.607	0.0025	2	3.35
		2	0.380	0.321			
		3	0.051	0.071			
<i>Sdh</i>	322	1	0.4	0.643	0.0383	2	49.42*
		2	0.432	0.286			
		3	0.168	0.071			
<i>Ugp</i>	374	1	0.091	0.129	0.0029	5	11.15*
		2	0.363	0.387			
		3	0.44	0.387			
		4	0.08	0.032			
		5	0.022	0.032			
		6	0.004	0.032			

<sup>(a)</sup> G<sub>st</sub> = genetic differences between pollen and ovule pool,

<sup>(b)</sup> χ<sup>2</sup> = 2 N G<sub>st</sub> (a-1), where N = number of seeds examined, a = number of alleles and (a-1) = degree of freedom (DF).

\* Significant at p < 0.05.

The multilocus outcrossing rate (Table 3) is much lower ( $t_m = 0.605$ ) compared to the existing report on outcrossing rate for some of the Dipterocarpaceae such as *S. congestiflora* ( $t_m = 0.87$ , Murawski *et al.* 1994a), *Dryobalanops aromatica* (mean  $t_m = 0.82$ , Kitamura *et al.* 1994), *S. megistophylla* ( $t_m = 0.81$  for undisturbed forest and 0.71 for selectively logged population, Murawski *et al.* 1994b), *Stemonoporus oblongifolius* ( $t_m = 0.84$ , Murawski & Bawa 1994), *S. leprosula* ( $t_m = 0.84$ , Lee *et al.* 2000b) and *D. aromatica* ( $t_m = 0.92$  for primary forest; 0.77 for logged forest; 0.55 for seed orchard and 0.66 for artificial forest, Lee 2000).

Generally, the results of this study may indicate that this species exhibited mixed-mating system. On the other hand, the results may be due to the unsuitable environment in promoting wide-scale development of pollinator populations in forest plantation compared to natural forest environment. The multilocus outcrossing estimates were different from the average single-locus estimates ( $t_m - t_s = 0.105 \pm 0.025$ ,  $p < 0.05$ ) indicating that genetic structure or biparental inbreeding do play a significant role in the breeding structure of these populations and it was further supported by the inbreeding coefficients based on the inbreeding equilibrium (Table 3).

Our results showed that planted *S. macrophylla* possessed moderate level of outcrossing rate and a significant level of inbreeding. These indicated that there was mating among relatives and strongly proved that the plantation was established using closely related seed sources. However, detailed knowledge of the mating system for natural population is necessary to understand the distribution of genetic variation within and among individuals,

**Table 2** Test of homogeneity of the pollen pool over maternal trees

Locus	N	a	Allele frequency														$G_{it}^{(a)}$	DF	$\chi^2_{(b)}$
			fam 1	fam 2	fam 3	fam 4	fam 5	fam 6	fam 7	fam 8	fam 9	fam 10	fam 11	fam 12	fam 13	fam 14			
<i>Gpi</i>	420	1	0.85	0.74	0.03	0.81	0.79	0.56	0.62	0.80	0.49	0.77	0.83	0.72	0.72	0.53			
		2	0.03	0.03	0.80	0.03	0.03	0.05	0.03	0.03	0.03	0.03	0.03	0.04	0.04	0.05			
		3	0.03	0.16	0.03	0.06	0.09	0.05	0.20	0.08	0.31	0.09	0.03	0.12	0.04	0.19			
		4	0.03	0.03	0.07	0.03	0.03	0.05	0.03	0.03	0.03	0.03	0.03	0.04	0.04	0.05			
		5	0.03	0.03	0.03	0.03	0.03	0.24	0.07	0.03	0.10	0.06	0.03	0.04	0.12	0.14			
		6	0.03	0.03	0.03	0.03	0.03	0.05	0.03	0.03	0.03	0.03	0.03	0.04	0.04	0.05	0.172	5	360.91*
<i>Pgm</i>	346	1	0.32	0.25	0.04	0.18	0.31	0.58	0.49	0.07	0.67	0.47	0.46	0.45	0.57	0.89			
		2	0.64	0.71	0.63	0.78	0.07	0.35	0.46	0.89	0.27	0.50	0.49	0.48	0.39	0.05			
		3	0.04	0.03	0.33	0.04	0.62	0.07	0.04	0.03	0.07	0.03	0.05	0.07	0.05	0.05	0.225	2	155.52*
<i>Sdh</i>	334	1	0.56	0.87	0.64	0.80	0.59	0.17	0.73	0.04	0.05	0.64	0.66	0.51	0.43	0.06			
		2	0.40	0.10	0.05	0.15	0.37	0.50	0.23	0.74	0.66	0.33	0.27	0.43	0.53	0.72			
		3	0.04	0.03	0.31	0.05	0.04	0.33	0.04	0.22	0.28	0.03	0.07	0.06	0.05	0.22	0.225	2	150.63*
<i>Ugp</i>	382	1	0.17	0.37	0.04	0.08	0.10	0.06	0.14	0.03	0.28	0.03	0.04	0.05	0.04	0.11			
		2	0.70	0.45	0.11	0.75	0.67	0.59	0.62	0.03	0.03	0.12	0.08	0.25	0.56	0.70			
		3	0.03	0.03	0.74	0.04	0.13	0.17	0.07	0.84	0.58	0.75	0.76	0.50	0.23	0.05			
		4	0.03	0.09	0.04	0.04	0.03	0.06	0.11	0.03	0.03	0.03	0.04	0.05	0.09	0.05			
		5	0.03	0.03	0.04	0.04	0.03	0.06	0.04	0.03	0.10	0.05	0.04	0.10	0.04	0.05			
		6	0.03	0.03	0.04	0.04	0.03	0.06	0.04	0.03	0.03	0.03	0.04	0.05	0.04	0.05	0.263	5	503.35*

<sup>(a)</sup>  $G_{it}$  = genetic differences between family (fam) gene pool.<sup>(b)</sup>  $\chi^2 = NG_{it} (a-1)$ , where  $N$  = number of seeds examined,  $a$  = number of alleles and  $(a-1)$  = degree of freedom (DF).\* Significant at  $p < 0.05$ .

**Table 3** Outcrossing rates and inbreeding coefficients for *Shorea macrophylla*

No. of seeds examined	Mean $t_s$ (SD)		$t_m$ (SD)	$t_m - t_s$	$F_c^*$
	Locus	$t_s$			
411	<i>Gpi</i>	0.511 (0.717)	0.605 (0.087)	0.105	0.2461
332	<i>Pgm</i>	0.334 (0.132)			
322	<i>Sdh</i>	0.490 (0.085)			
374	<i>Ugp</i>	0.414 (0.143)			
Unweighted mean		0.500 (0.090)			

\* Inbreeding coefficients of seedlings if FRIM population is in inbreeding equilibrium.

$F_c = (1 - t_m) / (1 + t_m)$  where  $t_m$  is the multilocus outcrossing rates. Standard error is not calculated.

gene flow within and among populations as well as resultant sub-structuring of populations which are crucial in the design of sampling strategies for genetic conservation and improvement. To obtain reliable estimates of genetic parameters for such sampling strategies, parental trees must be highly outcrossed, unrelated and produced progenies derived from many pollen parents (Namkoong 1966, Lee *et al.* 2000b). Future studies need to compare them with those from a natural population.

### Acknowledgements

IRPA Grant 070102 supported this study. We would like to thank Mariam D., Sharifah T. and Ghazally J. for their technical assistance.

### References

- APPANAH, S. 1995. Plantation forestry: its prospects in Malaysia. *Wallaceana* 76: 7–12.
- KITAMURA, K., ABDUL RAHMAN, M. Y., OCHIAI, Y. & YOSHIMARU, H. 1994. Estimation of the outcrossing rate on *Dryobalanops aromatica* Gaertn. f. in primary and secondary forest in Brunei, Borneo, Southeast Asia. *Plant Species Biology* 9: 37–41.
- LEE, S. L. 2000. Mating system parameters of *Dryobalanops aromatica* Gaertn. f. (Dipterocarpaceae) in three different forest types and a seed orchard. *Heredity* 85: 338–345.
- LEE, S. L., WICKNESWARI, R., MAHANI, M. C. & ZAKRI, A. H. 2000a. Inheritance of allozyme variants in *Shorea leprosula* Miq. (Dipterocarpaceae). *Journal of Tropical Forest Science* 12: 124–138.
- LEE, S. L., WICKNESWARI, R., MAHANI, M. C. & ZAKRI, A. H. 2000b. Mating system parameters in a tropical tree species, *Shorea leprosula* Miq. (Dipterocarpaceae) from Malaysian Lowland Dipterocarp Forest. *Biotropica* 32: 693–702.
- MORAN, G. F., BELL, J. C. & GRIFFIN, A. R. 1989. Reduction in levels of inbreeding in a seed orchard of *Eucalyptus regnans* F. Muell. compared with natural populations. *Silvae Genetica* 38: 32–36.
- MURAWSKI, D. A. & HAMRICK, J. L. 1992. The mating system of *Cavanillesia platanifolia* under extremes of flowering-tree density: a test of predictions. *Biotropica* 24: 99–101.
- MURAWSKI, D. A. & BAWA, K. S. 1994. Genetic structure and mating system of *Stemonoporus oblongifolius* (Dipterocarpaceae) in Sri Lanka. *American Journal of Botany* 81: 155–160.
- MURAWSKI, D. A., DAYANANDAN, B. & BAWA, K. S. 1994a. Outcrossing rates of two endemic *Shorea* species from Sri Lanka tropical rain forests. *Biotropica* 26: 23–29.
- MURAWSKI, D. A., GUNATILEKE, I. A. U. N. & BAWA, K. S. 1994b. The effect of selective logging on inbreeding in *Shorea megistophylla* (Dipterocarpaceae) from Sri Lanka. *Conservation Biology* 8: 997–1002.

- NAMKOONG, G. 1966. Inbreeding effect on estimation of genetics additive variance. *Forest Science* 12: 8–13.
- NEWMAN, M. F., BURGESS, P. F. & WHITMORE, T. C. 1996. Pp. 163–165 in *Manuals of Dipterocarps for Forester. Borneo Island Light Hardwoods: Anisoptera, Parashorea, Shorea (Red, White and Yellow Meranti)*. Royal Botanic Garden Edinburgh. Huddersfield, United Kingdom.
- RITLAND, K. & JAIN, S. K. 1981. A model for the estimation of outcrossing rate and gene frequencies based on independent loci. *Heredity* 47: 37–54.
- RITLAND, K. 1983. Estimation of mating systems. Pp. 289–302 in Tanksley, S. D. & Orton, T. J. (Eds.) *Isozymes in Plant Genetics and Breeding, Part A*. Elsevier Science Publishers, Amsterdam.
- RITLAND, K. 1994. Multilocus Mating System Program MLTR Version 0.9. Department of Botany, University of Toronto.