

GENOTYPIC VARIATIONS OF OPEN-POLLINATED FAMILIES OF *CINNAMOMUM CAMPHORA* SEEDLINGS FROM SOUTH CHINA

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To get better understanding of genotypic variations of camphor tree (*Cinnamomum camphora*), a total of 179 open-pollinated families of seedlings from six provinces of south China were evaluated using leaf and growth characteristics. Families differed significantly in leaf features (leaf area, leaf margin circumference, leaf maximum length, leaf maximum width and leaf length to width ratio), and in growth traits (seedling height, seedling basal diameter and seedling height to basal diameter ratio). High heritability values were found for entire eight traits, ranging from 0.623 to 0.974. Genetic and phenotypic correlation analysis revealed significant and positive associations between leaf features, growth traits as well as both types of traits. Principle component analyses showed considerable diversity in the studied families and 90.65% of total variations were explained by the first three components. Cluster analysis classified these progenies into 16 clusters and each was distinguished by specific characteristics in terms of leaf and growth traits. Linear regression analysis established equations with significant coefficients and correlations for the linking of leaf area to both height and basal diameter. These data could be considered as valuable genetic resources for selecting breeding programmes in future.

Keywords: Leaf features, growth traits, principle component analysis, cluster analysis, regression analysis

INTRODUCTION

Plants have evolved sophisticated responses to inhabiting environments that are displayed at different levels such as morphology, anatomy, physiology and growth (Pyakurel & Wang 2013). Leaf is the most important organ for tree growth and survival and is sensitive to the inhabiting environments. Leaf morphology is controlled by the competing demands of maximising photosynthetic gain and minimising transpirational loss of water (Marron et al. 2007). Remarkable variations have been discovered in leaf morphology of numerous tree species, such as *Eucalyptus* spp. (Warren et al. 2005, Rance et al. 2014), *Azadirachta indica* (Kundu & Tigerstedt 1997), *Quercus rugosa* (Uribe-Salas et al. 2008) and *Betula papyrifera* (Pyakurel & Wang 2013).

In addition to leaf features, growth traits also have been proven to be very diverse in nature and their variations have been broadly used to develop programmes for selecting the most productive genotypes. However, due to the

extreme complexity in genetic basis of growth traits, screening for improved genotypes is time-consuming, costly and laborious (Marron et al. 2007). Many scientists have proposed new approaches to achieve this goal by indirectly selecting morphophysiological traits that are cheap and easy to measure (Harrington et al. 1997, Marron & Ceulemans 2006, Marron et al. 2007). However, the successful application of this strategy requires that the morphophysiological traits are genetically correlated with biomass production and/or growth rates, and that both types of traits are moderately or highly inheritable (Tuberosa et al. 2002). The functional and structural components of leaves of the tree, such as total and individual leaf area, internal leaf structure, stomatal morphology, leaf growth physiology and photosynthetic efficiency, have been reported to be associated with growth rates and productivity (Wu & Stettler 1998, Marron & Ceulemans 2006, Marron et al. 2007).

Camphor tree (*Cinnamomum camphora*) is a large evergreen tree that could grow up to over 20 m tall (Zeng et al. 2012). As a native pioneer tree species, it is broadly distributed across south China and has a record of cultivation for more than 2000 years with the purpose of producing valuable timber and camphor oil (Ren et al. 2000, Zeng et al. 2012). The interaction of its genetic diversity and wide environmental range have led to vast diversity in growth and form traits, such as height, diameter at breast height (DBH) and individual stem volume, stem straightness and fork (Ren et al. 2000, Zhang et al. 2014). Furthermore, camphor tree is considered very diverse in terms of the level of leaf features (e.g. leaf area, margin circumference, leaf length and width), but little has been reported about its leaf morphological variability and its relationship with growth traits so far. In this study, we examined the leaf morphological variability of 179 open-pollinated families of camphor tree progenies that were collected from six provinces within its main distribution area, and the relatedness of leaf morphological variations to growth traits. The achieved results would be positively applied in directly and/or indirectly selecting favourable genotypes of camphor trees for artificial afforestation and for breeding programmes in future.

MATERIALS AND METHODS

Plant material

In year 2011 an intensive survey and inspection of natural stands of *C. camphora* were conducted in six provinces of south China, namely, Guangdong, Guangxi, Fujian, Jiangxi, Hunan and Yunnan. A total of 179 plus trees were screened out by scoring traits of interest in relation to five selected check trees (Ledig 1974). The scored traits included height, DBH, stem volume of individual tree, stem form and crown size (Zeng et al. 2013). The origin of these 179 plus trees was located in 31 prefectures within above six provinces, in the range of 22° 21'–29° 31' N, longitude 102° 32'–117° 48' E and at elevation from 20–2200 m (Figure 1, Appendix 1).

Evaluation method

Seeds from the 179 plus trees were collected for germination. Thirty seedlings of each family were transplanted for an open-pollinated progeny trails carried out in a flat seedling nursery at Guangzhou. The trial used randomised complete block design with six-seedling row plots and five replicates. Seedlings were planted with 30 cm × 30 cm spacing. Seven months later, we measured



Figure 1 Locations of collection sites for the plus trees of camphor tree responsible for generating open-pollinated families of progenies in south China

four leaf features of each individual tree—leaf area, leaf margin circumference, leaf maximum length and leaf maximum width, and two growth traits—seedling height and basal diameter. By using these data, leaf length to width ratio and seedling height to basal diameter ratio were calculated. To measure the leaf features, four well-developed leaves were harvested from each seedling in the same crown position within the middle crown and their leaf features measured using a leaf image acquisition and processing system (Wu & He 2014). To reveal the genetic component of variance responsible for parent–offspring resemblance, both half-sib individual and family heritability which represented additive genetic portion of the phenotypic variance were estimated for the open-pollinated progenies (Zeng et al. 2013).

Statistical analysis

Subsequent analysis was performed on the mean values of four leaves of each individual tree (Pyakurel & Wang 2013). Variance components were estimated using the SAS programs PROC GLM and VARCOMP (1999). The model used for the one-way analysis of variance (ANOVA) for each trait was described as previously (Zeng et al. 2013). Genotypic and phenotypic correlations between traits were determined as described by Zeng et al. (2013). Phenotypic correlations were obtained from individual seedling values and genetic correlations were calculated on best linear unbiased prediction estimates of genetic effects (Zhang et al. 2014).

Principle component analysis and cluster analysis were carried out with five leaf morphological variables and three growth traits. The principle component solution was determined based on the Scree plot. To avoid effects of scaling differences, mean of each trait was normalised prior to cluster analysis using Z scores (Khadivi-Khub et al. 2015). All statistical analysis was conducted using SAS version 9.1.

RESULTS AND DISCUSSION

Description of leaf and growth traits

Mean, median, individual minimum and maximum observed values for leaf and growth traits, along with their coefficients of variation, individual and family heritability are shown in Table 1. ANOVA analysis revealed significant differences in leaf and growth traits between various open-pollinated families at $p = 0.05$ level (data not shown), suggesting strong family effect on eight traits. Among leaf features, leaf area displayed the largest coefficients of variation ($CV = 31.5\%$) compared with margin circumference ($CV = 16.35\%$), length ($CV = 16.85\%$), width ($CV = 17.99\%$) and length to width ratio ($CV = 13.87\%$). These leaf morphological variations were comparable with those observed on birch (Pyakurel & Wang 2013), red ironbark (Warren et al., 2005) and southern beech (Hovenden & Schoor 2004). Environmental difference at the origin genetic resources had likely contributed to leaf variations in birch (Pyakurel & Wang 2013). This might imply that the geographic variations

Table 1 Leaf and growth features of camphor trees in this study

Trait	Unit	Min	Max	Mean	Median	SE	CV (%)	h_f^2	h_i^2
LA	mm ²	454.354	17727.424	1487.000	1455.000	468.442	31.504	0.854(0.016)	0.619(0.067)
LMC	mm	108.541	283.851	190.661	192.000	31.168	16.351	0.856(0.016)	0.626(0.068)
LL	mm	43.204	114.543	78.258	79.000	13.186	16.849	0.855(0.016)	0.623(0.067)
LW	mm	15.548	41.339	28.913	29.000	5.202	17.988	0.858(0.016)	0.636(0.068)
LWR	-	1.871	5.668	2.740	2.714	0.379	13.872	0.877(0.013)	0.728(0.074)
HT	m	0.230	1.300	0.538	0.547	0.188	35.187	0.974(0.003)	0.855(0.111)
BD	cm	0.100	2.350	0.524	0.503	0.187	36.542	0.891(0.013)	0.812(0.079)
HDR	-	15.903	466.672	107.387	104.174	34.786	32.396	0.946(0.006)	0.821(0.105)

LA = leaf area, LMC = leaf margin circumference, LL = leaf length, LW = leaf width, LWR = leaf length to width ratio, HT = seedling height, BD = seedling basal diameter, HDR = seedling height to basal diameter ratio, Min = minimum, Max = maximum, SE = standard error, CV = coefficient of variation, h_f^2 = half-sib family heritability, h_i^2 = half-sib individual heritability

of the studied 179 families of camphor trees from six provinces across south China play a role in determining leaf morphological variations.

For growth traits, basal diameter was the most variable trait (CV = 36.54%) with strong differences observed between minimum and maximum values (0.10 and 2.35 cm respectively). Such strong differences between individual seedling values was also observed for other growth traits, namely, height and height to diameter ratio, suggesting that their CVs were stronger than those of leaf features. The CVs of growth traits in this study were much higher than those in our previous study on Guangdong-originated camphor trees, in which the CVs of 9-year-old progenies were about 25% for DBH and about 15% for height (Zhang et al. 2014). This might be reasonable considering that the origin of camphor trees in this study (six provinces) was much broader than that in our previous study (one province). The seedlings were growing in a nursery and their young age was likely to be another reason for the high CVs. Many trees have been reported to show decreased CVs with increased age, i.e. higher CVs at early ages and lower CVs at later ages (Zeng et al. 2013).

Half-sib individual heritability values were generally high for all eight traits, ranging from

0.619 to 0.728 for leaf features and from 0.812 to 0.855 for growth traits, with generally small standard errors (Table 1). Due to the presence of non-additive genetic variance, family mean heritability values were much higher than individual heritability values, varying from 0.854 to 0.877 for leaf features and from 0.891 to 0.974 for growth traits. These results were comparable with high values of broad-sense heritability reported for growth traits of our previous 9-year-old camphor tree progenies (Zhang et al. 2014) and for main stem leaves of poplar hybrids: 0.52–0.90 for individual leaf area (Wu & Stettler 1998), 0.74–0.95 for leaf number increment (Monclus et al. 2005) and 0.64–0.67 for specific leaf area (Wu & Stettler 1998), suggesting that genetic control of leaf and growth traits were relatively stable and strong for camphor trees.

Phenotypic and genotypic correlation analysis

Correlation values ($p < 0.05$) are shown in Table 2. Four leaf features (except length to width ratio) were significantly and positively correlated between each other, ranging from 0.625 to 0.977 genetically and from 0.681 to 0.978 phenotypically. Length to width ratio showed significantly positive correlations with length

Table 2 Genotypic (above diagonal line) and phenotypic (below diagonal line) correlations and associated standard errors (in parentheses) of eight traits for camphor trees

Trait	LA	LMC	LL	LW	LWR	HT	BD	HDR
LA	-	0.914** (0.015)	0.858** (0.023)	0.923** (0.013)	-0.154 (0.086)	0.303* (0.049)	0.421** (0.024)	0.086 (0.084)
LMC	0.926** (0.003)	-	0.977** (0.004)	0.732** (0.041)	0.205 (0.084)	0.191 (0.080)	0.323* (0.078)	0.006 (0.085)
LL	0.873** (0.004)	0.978** (0.001)	-	0.625** (0.054)	0.359* (0.077)	0.117 (0.082)	0.242 (0.082)	-0.035 (0.085)
LW	0.929** (0.003)	0.779** (0.007)	0.681** (0.010)	-	-0.502** (0.066)	0.388** (0.071)	0.471** (0.069)	0.175 (0.083)
LWR	-0.131 (0.018)	0.181 (0.018)	0.327* (0.017)	-0.446** (0.015)	-	-0.331** (0.074)	-0.295* (0.079)	-0.238* (0.080)
HT	0.278* (0.032)	0.249* (0.024)	0.216* (0.024)	0.295* (0.022)	-0.113 (0.026)	-	0.733** (0.039)	0.834** (0.026)
BD	0.340** (0.030)	0.301* (0.017)	0.259* (0.018)	0.354* (0.017)	-0.141 (0.019)	0.642** (0.015)	-	0.050 (0.035)
HDR	-0.057 (0.022)	-0.049 (0.022)	-0.040 (0.022)	-0.058 (0.022)	0.037 (0.023)	0.459** (0.024)	-0.076 (0.006)	-

*significant at $p < 0.05$, **significant at $p < 0.01$; LA = leaf area, LMC = leaf margin circumference, LL = leaf length, LW = leaf width, LWR = leaf length to width ratio, HT = seedling height, BD = seedling basal diameter, HDR = seedling height to basal diameter ratio

(genetically 0.359 and phenotypically 0.327), strongly negative associations with width (-0.502 and -0.446 respectively), but low correlations with leaf area and margin circumference. For the growth traits, phenotypic and genetic correlations were found to be strong and positive between two pairs (height vs basal diameter and height vs height to diameter ratio), whereas both types of correlations were very low for height to diameter ratio vs basal diameter.

To identify leaf traits that are important for generating high yield of camphor trees, the relationship among leaf features and growth traits were examined. Four leaf features (besides length to width ratio) showed significant and positive associations with both height and basal diameter except for two genetic correlations (margin circumference vs height and leaf length vs height). Length to width ratio was negatively correlated with three growth traits and the correlations were genetically but not phenotypically significant. Height to diameter ratio was not significantly correlated with five leaf features except for its genetic correlation with length to width ratio (-0.238). Leaf has prime effects on light interception, carbon assimilation and consequently on biomass accumulation (Harrington et al. 1997). Leaf features, such as single leaf area, leaf increment rate and petiole length have already been considered reliable productivity determinants (Harrington et al.

1997, Monclus et al. 2005, Marron & Ceulemans 2006). The close link of leaf features to growth traits in this study (Table 2) might imply that the leaf features are potential determinants of camphor tree in terms of growth.

Principle component analysis

Principle component analysis is a technique that reduces the dimensionality of data set (Iezzoni & Pritts 1991). It is able to identify the most significant traits in the data set and establish genetic relationship between the evaluated genotypes (Khadivi-Khub et al. 2015). In this study, principle component analysis showed that the first three principal components provided reasonable summary of the data and explained 90.65% of the total variations (Table 3). The fact that variance in the data set was distributed in the first three variables was likely due to high correlations between variables and genotype data (Table 2). The first principle component explained 49.16% of the total variance, which was mainly contributed by leaf area, margin circumference, length and width. The second principal component accounted for 27.2% of variations chiefly through seedling height, height to diameter ratio and leaf length to width ratio. The third principal component explained 14.3% of variations principally via length to width ratio and height to diameter ratio.

Table 3 Eigenvalues of the principal component axes from principle component analysis of leaf and growth variables in the studied camphor trees

Trait	Prin1	Prin2	Prin3	Prin4	Prin5	Prin6	Prin7	Prin8
LA	0.481	-0.141	-0.140	-0.126	0.674	-0.099	-0.499	-0.007
LMC	0.448	-0.289	0.111	-0.049	-0.701	-0.034	-0.445	0.105
LL	0.414	-0.352	0.210	-0.041	0.033	0.037	0.497	-0.640
LW	0.456	0.034	-0.366	-0.185	-0.018	0.116	0.523	0.580
LWR	-0.085	-0.435	0.685	0.174	0.229	0.035	0.100	0.490
HT	0.271	0.513	0.333	0.097	-0.022	-0.727	0.113	0.040
BD	0.302	0.301	0.024	0.792	0.025	0.432	-0.051	-0.027
HDR	0.126	0.479	0.457	-0.529	0.018	0.508	-0.085	-0.032
Total	3.933	2.176	1.144	0.702	0.021	0.011	0.009	0.003
% of variance	49.16	27.20	14.30	8.78	0.27	0.14	0.11	0.04
Cumulative %	49.16	76.36	90.65	99.43	99.70	99.84	99.96	100

Prin1–8 = principle components 1 to 8, LA = leaf area, LMC = leaf margin circumference, LL = leaf length, LW = leaf width, LWR = leaf length to width ratio, HT = seedling height, BD = seedling basal diameter, HDR = seedling height to basal diameter ratio

The loading bi-plot drawn for the first and the second principal components (Figure 2) indicated the importance of leaf features and growth traits for explaining the variance among the genotypes. Leaf margin circumference, length, area and width showed nearly the same direction, which was in agreement with positive correlations between them (Table 2). The same trend with positive correlations was also observed for seeding height, basal diameter and height to diameter ratio. Leaf length to width ratio had opposite direction from growth traits, further confirming their negative correlation. In the bi-plot for the first and the third principal components, leaf length to width ratio was loosely correlated with other traits, whereas the four leaf features (width, area, length and margin circumference) and the two growth traits (height and diameter) were nearly identically positioned showing positive correlation between them.

Cluster analysis

Cluster analysis generated a dendrogram as illustrated in Figure 2. The analysis based on mean values of the measured traits classified these 179 families of camphor trees into 16 clusters on the basis of similarity (distance = 0.6). Each cluster was markedly different from the other clusters and consisted of various numbers of families (Figure 3, Appendix 2).

Cluster 1 was the largest group and included 56 families (31.3% of subjects) from provinces Fujian, Guangdong, Hunan and Jiangxi. This cluster was characterised by small to middle size leaf features and inconsistent characteristics of growth traits. Cluster 2 was the third largest and consisted of 29 families (16.2% of subjects) from provinces Hunan, Guangdong, Jiangxi and Yunnan. This group was distinguished by middle to large margin circumference and length to width ratio as well as unclear characteristics of growth traits. Cluster 3 was the second largest group ($n = 39$; 21.8% of subjects) with middle to large leaf area, middle to large leaf width, low to middle length to width ratio and instinct growth traits. These families were predominantly from provinces Fujian, Hunan, Jiangxi and Guangdong. Cluster 4 was composed of 10 families from provinces Hunan, Guangdong and Jiangxi. These families showed low to middle margin circumference and length to width ratio as well as high or very high height and height

to diameter ratio. Cluster 10 had 18 families mainly from provinces Fujian, Hunan, Yunnan and Guangxi. This group was characterised by small to middle size leaf area, short height and low height to diameter ratio. Cluster 12 consisted of 8 families from provinces Hunan, Fujian and Jiangxi, and displayed clear characteristic of very large leaf area, middle height and low to middle height to diameter ratio. The remaining 10 clusters were small groups consisting of one to four families, among which only one family was discovered in clusters 5, 6, 11, 14 and 16, two families in clusters 7, 13 and 15, and four families in clusters 8 and 9. Camphor trees in these clusters showed explicit characteristics in terms of leaf features and/or growth traits.

Most neighboring families within a cluster were geographically close to each other (Figures 1 and 3), which was similar with the observation in walnut (Khadivi-Khub et al. 2015). These camphor trees shared high similarity in leaf features and growth traits. Short geographical distance may have played significant role in determining the similarity in phenotypes since camphor trees are more likely to achieve similarity in climate situation and gene flow within a short distance (Sefc et al. 2000).

Regression analysis

To establish a model for bridging the gap between leaf and growth traits is one of our ultimate goals. Many studies have reported that the level of genetic control of a trait and its interrelationship with other traits determine the feasibility of indirect selection in breeding programme (Marron et al. 2007). In this study, significant linear relationships were observed between leaf and growth traits and both types of traits were under strong genetic control (Table 2). This strongly suggested that leaf features are ideal candidate traits for indirect selection of growth traits of the seedlings. Since leaf area is the composite of leaf features and more convenient to score, leaf area was used to build linear regression equations with height and diameter respectively (Table 4).

Linear regression equations were constructed on the basis of values of each cluster (clusters 1 to 16) for the individual seedling or of the entire 179 families (the total). For the total equation, removal of tree to tree variation considerably increased correlation (r^2) between leaf area and height from 0.278 to 0.861 and that between

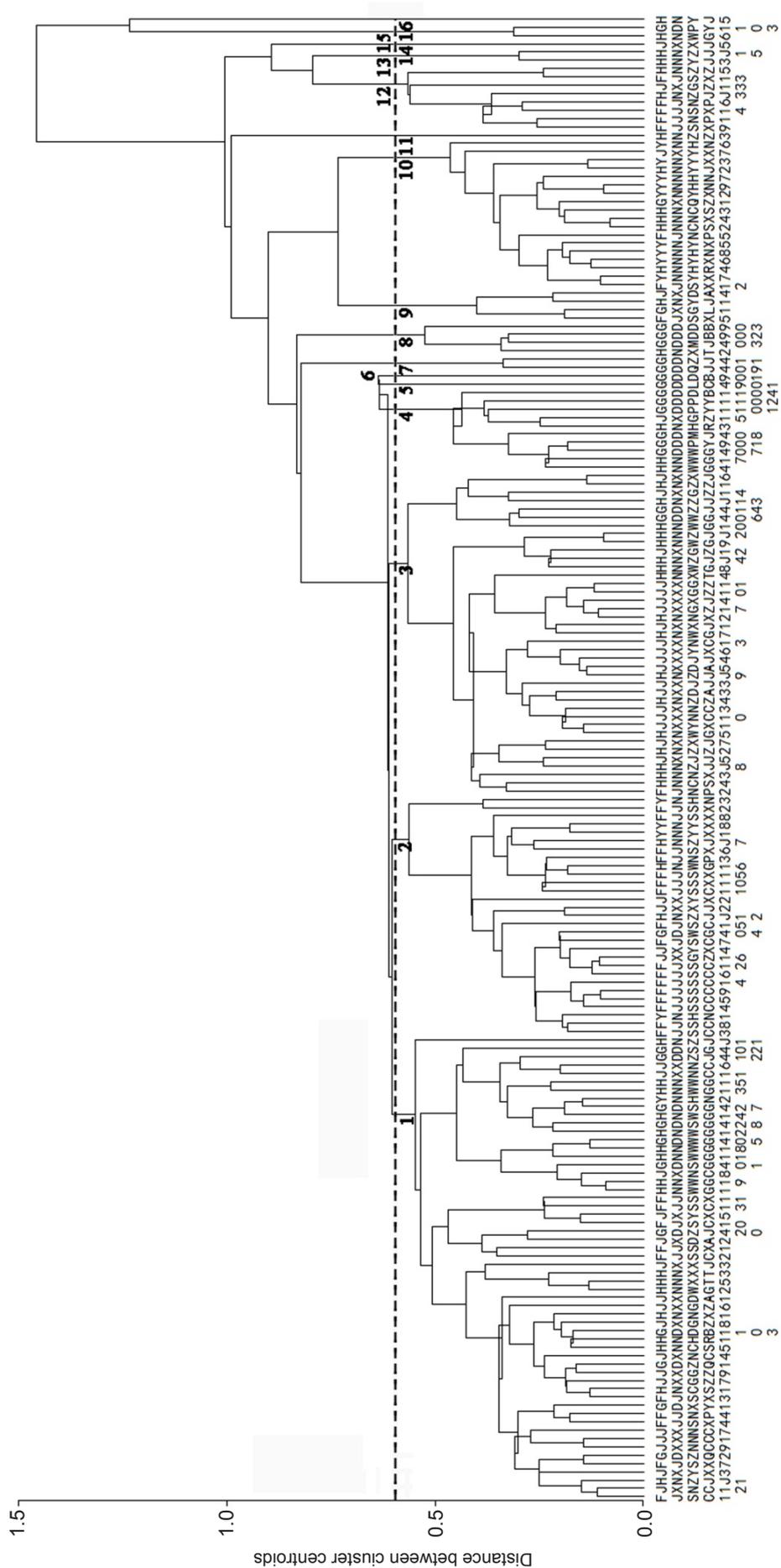


Figure 3 Cluster analysis of 179 open-pollinated families of camphor tree seedlings based on leaf and growth traits; codes of genotypes are given in Appendix 1

leaf area and basal diameter from 0.340 to 0.873 (Tables 2 and 4). Similarly, high correlations (> 0.745) of leaf area vs height and leaf area vs diameter were also observed on seedlings from each cluster. The regression coefficients were highly significant ($p < 0.0001$) with values ranging from 1.595×10^{-4} to 5.269×10^{-4} for leaf area vs height and from 2.965×10^{-4} to 4.878×10^{-4} for leaf area vs diameter (Appendix 3).

Leaf area has already been considered one of most reliable productivity determinants of poplar hybrids (Harrington et al. 1997). It is comparatively easy and cheap to score and its relationships with growth traits of poplar are valid irrespective of growing conditions, which has made it a robust and useful early indicator of tree vigour (Marron et al. 2007). Significant and stable equations linking leaf area to growth traits and high regression coefficients in this study (Table 4) strongly suggested that leaf area was a reliable indirect indicator of seedling growth for camphor tree progenies.

CONCLUSIONS

The 179 open-pollinated families of camphor tree seedlings were highly variable in terms of leaf features (leaf area, margin circumference,

maximum length, maximum width and length to width ratio) and growth traits (seedling height, basal diameter and the height to basal diameter ratio). The variability observed in these characters is under high genetic control and likely to be caused by geographic variations in the origins of camphor trees that are located in six provinces of south China. These progenies were composed of 16 clusters and each cluster had distinct characteristics with regard to their leaf and growth traits. Most of the leaf features were positively and highly correlated with seedling growth traits. As composite of leaf features, leaf area was used to establish linear regression equations with seedling height and basal diameter respectively. Leaf area was a reliable indirect indicator of seedling growth for camphor tree progenies.

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Table 4 Relationship between leaf area (LA) and two growth traits (HT = seedling height, BD = seedling basal diameter) for camphor tree seedlings

Cluster	Regression equation	r ²	p value	Regression equation	r ²	p value
1	HT = 3.663×10^{-4} LA	0.892	< 0.0001	BD = 3.616×10^{-4} LA	0.878	< 0.0001
2	HT = 3.263×10^{-4} LA	0.907	< 0.0001	BD = 3.085×10^{-4} LA	0.916	< 0.0001
3	HT = 3.294×10^{-4} LA	0.880	< 0.0001	BD = 3.172×10^{-4} LA	0.870	< 0.0001
4	HT = 3.385×10^{-4} LA	0.861	< 0.0001	BD = 3.286×10^{-4} LA	0.873	< 0.0001
5	HT = 5.199×10^{-4} LA	0.916	< 0.0001	BD = 3.915×10^{-4} LA	0.822	< 0.0001
6	HT = 4.510×10^{-4} LA	0.951	< 0.0001	BD = 3.050×10^{-4} LA	0.948	< 0.0001
7	HT = 4.851×10^{-4} LA	0.921	< 0.0001	BD = 4.878×10^{-4} LA	0.893	< 0.0001
8	HT = 5.269×10^{-4} LA	0.907	< 0.0001	BD = 3.446×10^{-4} LA	0.882	< 0.0001
9	HT = 3.497×10^{-4} LA	0.892	< 0.0001	BD = 3.497×10^{-4} LA	0.887	< 0.0001
10	HT = 2.196×10^{-4} LA	0.903	< 0.0001	BD = 2.896×10^{-4} LA	0.913	< 0.0001
11	HT = 1.845×10^{-4} LA	0.827	< 0.0001	BD = 2.957×10^{-4} LA	0.843	< 0.0001
12	HT = 2.464×10^{-4} LA	0.895	< 0.0001	BD = 2.666×10^{-4} LA	0.890	< 0.0001
13	HT = 3.865×10^{-4} LA	0.923	< 0.0001	BD = 3.171×10^{-4} LA	0.907	< 0.0001
14	HT = 2.742×10^{-4} LA	0.890	< 0.0001	BD = 4.116×10^{-4} LA	0.843	< 0.0001
15	HT = 5.386×10^{-4} LA	0.888	< 0.0001	BD = 4.370×10^{-4} LA	0.870	< 0.0001
16	HT = 1.595×10^{-4} LA	0.749	< 0.0001	BD = 2.965×10^{-4} LA	0.746	< 0.0001
Total	HT = 3.388×10^{-4} LA	0.861	< 0.0001	BD = 3.287×10^{-4} LA	0.873	< 0.0001

REFERENCES

- HARRINGTON CA, RADWAN MA & DEAN DS. 1997. Leaf characteristics reflect growth rates of 2-year-old *Populus* trees. *Canadian Journal of Forest Research* 27: 1321–1325.
- HOVENDEN MJ & SCHOOR J. 2004. Nature vs nurture in the leaf morphology of southern beech, *Nothofagus cunninghamii* (Nothofagaceae). *New Phytologist* 161: 585–594.
- IEZZONI AF & PRITTS MP. 1991. Applications of principal components analysis to horticultural research. *HortScience* 26: 334–338.
- KHADIVI-KHUB A, EBRAHIMI A, MOHAMMADI A & KARI A. 2015. Characterization and selection of walnut (*Juglans regia* L.) genotypes from seedling origin trees. *Tree Genetics and Genomes* 11: 54. doi 10.1007/s11295-015-0882-x.
- KUNDU SK & TIGERSTEDT PMA. 1997. Geographical variation in seed and seedling traits of neem (*Azadirachta indica* A. Juss.) among ten populations studied in growth chamber. *Silvae Genetica* 46: 2–3.
- LEDIG FT. 1974. Analysis of methods for the selection of trees from wild stands. *Forest Science* 20: 2–16.
- MARRON N & CEULEMANS R. 2006. Genetic variation of leaf traits related to productivity in a *Populus deltoides* × *Populus nigra* family. *Canadian Journal of Forest Research* 36: 390–400.
- MARRON N, DILLEN S Y & CEULEMANS R. 2007. Evaluation of leaf traits for indirect selection of high yielding poplar hybrids. *Environmental and Experimental Botany* 61: 103–116.
- MONCLUS R, DREYER E, DELMOTTE FM ET AL. 2005. Productivity, leaf traits and carbon isotope discrimination in 29 *Populus deltoides* × *P. nigra* clones. *New Phytologist* 167: 53–62.
- PYAKUREL A & WANG JR. 2013. Leaf morphological variation among paper birch (*Betula papyrifera* Marsh.) genotypes across Canada. *Open Journal of Ecology* 3: 284–295.
- RANCE SJ, MENDHAM DS & CAMERON DM. 2014. Assessment of leaf mass and leaf area of tree crowns in young *Eucalyptus grandis* and *E. globulus* plantations from measurements made on the stems. *New Forests* 45: 523–543.
- REN HD, YAO XH, SUN YX, ZHANG JZ & CHAO JS. 2000. Study on the biomass variance and comprehensive evaluation at the seedling stage of *Cinnamomum camphora* provenances. *Forest Research* 13: 80–85.
- SEFC KM, LOPEZ MS, LEFORT F & BOTTA R. 2000. Microsatellites variability in grapevine cultivars from different European regions and evaluation of assignment testing to assess the geographic origin of cultivars. *Theoretical and Applied Genetics* 100: 498–505.
- TUBEROSA R, SALVI S, SANGUINETI MC, LANDI P, MACCAFERRI M & CONTI S. 2002. Mapping QTLs regulating morphophysiological traits and yield: case studies, shortcomings and perspectives in drought-stressed maize. *Annals of Botany* 89: 941–963.
- URIBE-SALAS D, SÁENZ, RC, GONZÁLEZ RA, TÉLLEZ VO & OYAMA K. 2008. Foliar morphological variation in the white oak *Quercus rugosa* Née (Fagaceae) along a latitudinal gradient in Mexico: potential implications for management and conservation. *Forest Ecology and Management* 256: 2121–2126.
- WARREN CR, TAUSZ M & ADAMS MA. 2005. Does rainfall explain variation in leaf morphology and physiology among populations of red ironbark (*Eucalyptus sideroxylon* subsp. *tricarpa*) grown in a common garden? *Tree Physiology* 25: 1369–1378.
- WU Q & HE BX. 2014. Phenotypic diversity of leaf morphology for *Castanopsis fissa*. *Guangdong Forest Science and Technology* 30: 8–13.
- WU R & STETTLER RF. 1998. Quantitative genetics of growth and development in *Populus*. III. Phenotypic plasticity of crown structure and function. *Heredity* 81: 299–310.
- ZENG LH, HE BX, LIAN HM, CAI YL, WANG YS & LUO M. 2013. Age trends in genetic parameters for growth and resin-yielding capacity in masson pine. *Silvae Genetica* 62: 7–18.
- ZENG LH, LIAN HM, ZHANG Q, CAI YL & HE BX. 2012. Camphor tree resources and utilization. *IUFRO World Series, Asia and the Pacific Workshop* 30: 59–61.
- ZHANG Q, ZENG LH, CAI YL, HE BX, LIAN HM & ZHOU LH. 2014. Genetic analyses on growth and form traits of open-pollinated families of *Cinnamomum camphora*. *Journal of Central South University of Forest Technology* 34: 1–6.

Appendix 1 Geographical and ecological data of the localities and sampling sizes of the camphor trees study

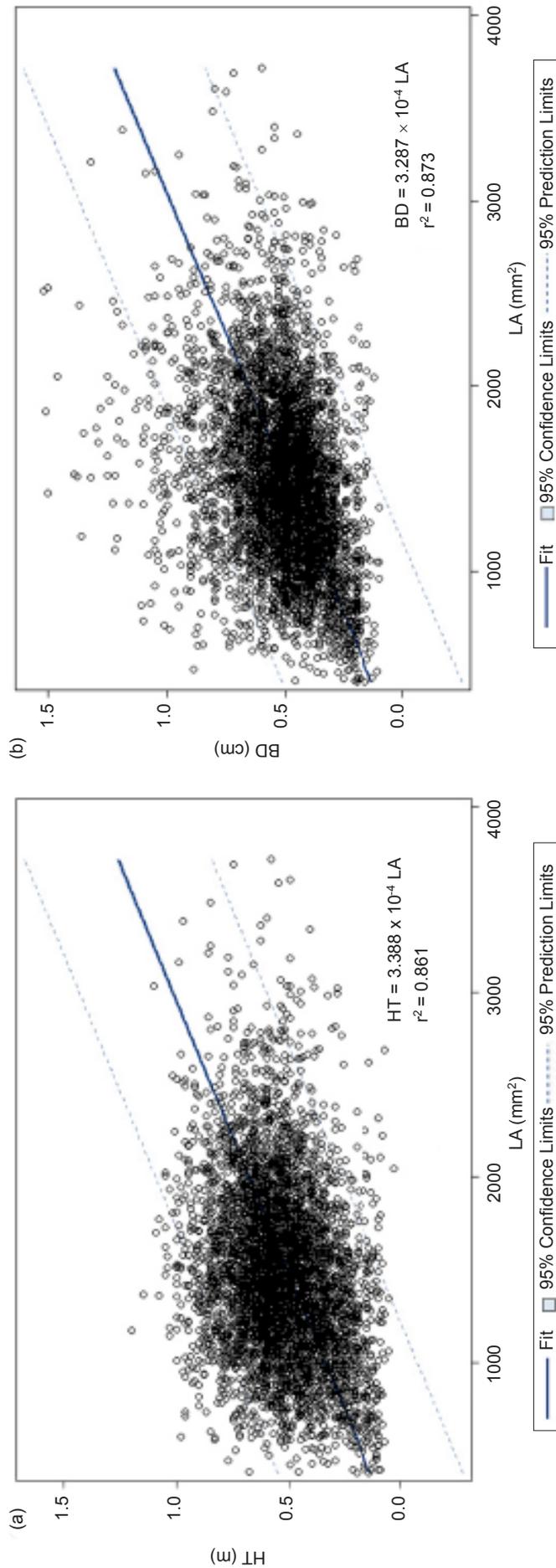
Location	Sample size	Code of genotype	Longitude (E)	Latitude (N)	Altitude (m)	Average annual temperature (°C)	Annual rainfall (mm year ⁻¹)
Shunchang, FJ	15	FJSC1-15	117° 48'	26° 47'	200–600	18.9	1621
Shaxian, FJ	15	FJSX1-15	117° 48'	26° 47'	250–500	19.6	1840
Nanping, FJ	6	FJNP1-6	118° 10'	26° 38'	300–800	19.3	1660
Dapu, GD	5	GDDDB1103-1104, 901-903	116° 41'	24° 21'	100–500	21.2	1659
Longchuan, GD	1	GDLC4901	115° 15'	24° 06'	100–550	21.8	1501
Meijiang, GD	2	GDMJ403, 408	116° 06'	24° 18'	80–400	21.0	1690
Pingyuan, GD	4	GDPY901, 1101-1103	115° 53'	24° 34'	160–350	21.7	1637
Qujiang, GD	1	GDQJ409	113° 35'	24° 41'	70–450	20.1	1640
Shaoguan, GD	4	GDSG401-402, 427-428	113° 35'	24° 48'	60–500	20.2	1572
Wujiang, GD	4	GDWG404-407	113° 34'	24° 47'	85–450	20.3	1537
Xinyi, GD	1	GDXY1	110° 56'	22° 21'	90–500	22.3	1709
Zhenjiang, GD	4	GDZJ411-412, 414, 420	113° 36'	24° 48'	120–500	19.0	1592
Zhaoqing, GD	2	GDZQ1-2	112° 27'	23° 02'	50–400	21.2	1650
Guiling, GX	1	GXGL1	110° 17'	25° 16'	150–550	18.9	1949
Quanzhou, GX	1	GXQZ1	111° 04'	25° 55'	150–450	17.7	1303
Changsha, HN	5	HNCS1-5	112° 56'	28° 13'	70–300	17.0	1422
Huarong, HN	3	HNHR1, 3-4	112° 32'	29° 31'	30–300	16.6	1114
Wugang, HN	18	HNWG1-2, 4-19	110° 37'	26° 43'	300–600	16.8	1218
Nanxian, HN	4	HNNX1-4	112° 23'	29° 21'	30–300	16.6	1238
Xiangtan, HN	4	HNXT2-5	112° 56'	27° 49'	70–400	17.1	1350
Yuanjiang, HN	4	HNYY1-3, 5	112° 20'	28° 50'	40–350	16.9	1322
Zhangjiajie, HN	11	HNZJJ1-4, 7-9, 11-13, 15	110° 28'	29° 07'	160–450	17.0	1420
Zhuzhou, HN	5	HNZZ1-5	113° 07'	27° 49'	50–320	17.2	1471
De'an, JX	5	JXDA1-5	115° 45'	29° 18'	30–200	16.8	1354
Ganzhou, JX	12	JXGZ6-17	114° 55'	25° 50'	100–400	18.8	1605
Jiujiang, JX	3	JXJJ3-5	115° 59'	29° 42'	40–350	16.5	1450
Nanchang, JX	11	JXNC1, 3-4, 6-13	115° 51'	28° 41'	20–280	17.7	1550
Xinjian, JX	7	JXXJ1-7	115° 48'	28° 41'	40–300	17.5	1640
Yongxiu, JX	7	JXYX1-7	115° 48'	29° 01'	40–300	17.4	1486
Huaning, YN	7	YNHN1-3, 6-9	102° 55'	24° 11'	1600–2100	16.6	1123
Yuxi, YN	7	YNYX1-3, 5-8	102° 32'	24° 21'	1650–2200	20.5	837
Total	179						

FJ = Fujian, GD = Guangdong, GX = Guangxi, HN = Hunan, JX = Jiangxi and YN = Yunnan

Appendix 2 Z scores of leaf and growth traits of camphor trees within each cluster

Subject	No. of families	LA	LMC	LL	LW	LWR	HT	BD	HDR
Cluster 1	56	-1.734-0.410	-1.516-0.455	-1.400-0.621	-2.024-0.480	-1.698-1.856	-1.199-1.267	-1.078-2.447	-1.596-1.888
Cluster 2	29	-0.450-1.035	0.168-1.367	0.296-1.643	-0.843-0.453	0.334-2.815	-1.180-1.107	-1.251-0.965	-1.571-1.203
Cluster 3	39	-0.014-1.848	-0.294-1.408	-0.436-1.148	0.440-2.294	-1.672-0.396	-1.016-2.050	-0.719-2.866	-1.552-1.353
Cluster 4	10	-0.877-0.665	-1.376-0.035	-1.554-0.008	-0.429-1.311	-2.246-0.500	0.950-2.101	0.083-1.599	0.608-2.107
Cluster 5	1	0.250	0.616	0.696	0.095	0.708	2.159	1.223	1.943
Cluster 6	1	0.994	0.745	0.528	1.404	-0.914	1.827	0.071	2.310
Cluster 7	2	-0.922-0.833	-0.463-0.380	-0.147-0.056	-1.666-1.377	1.820-2.101	0.839-1.085	1.076-2.061	-0.382-0.398
Cluster 8	4	-0.961-0.183	-0.885-0.329	-1.047-0.047	-1.104-0.178	-0.520-1.497	1.194-2.032	-0.606-0.101	2.572-2.999
Cluster 9	4	-2.153-1.279	-2.426-1.400	-2.188-1.377	-2.030-0.999	-0.508-0.055	-1.139-0.579	-1.185-0.849	-0.513-0.040
Cluster 10	18	-1.357-0.288	-1.298-0.459	-1.063-0.564	-1.709-0.429	0.064-2.027	-2.274-1.214	-2.080-0.268	-2.112-0.765
Cluster 11	1	0.522	-0.471	-0.924	1.247	-2.235	-1.624	-0.055	-2.107
Cluster 12	8	1.440-3.247	1.696-3.313	1.759-3.017	0.623-2.291	0.002-1.744	-0.729-0.825	-0.213-1.838	-1.125-0.050
Cluster 13	2	1.945-2.312	1.631-1.695	1.658-1.831	1.734-2.219	-0.583-0.045	1.529-2.114	1.003-1.537	0.965-1.286
Cluster 14	1	0.430	1.334	1.340	0.189	1.362	-0.611	2.035	-1.887
Cluster 15	2	-2.601-2.694	-2.965-3.198	-3.151-3.367	-2.284-2.309	-0.835-1.059	-0.198-0.456	-0.830-0.886	0.841-1.821
Cluster 16	1	-2.251	-2.605	-2.495	-2.097	-0.414	-2.563	-1.852	-2.543

LA = Leaf area, LMC = leaf margin circumference, LL = leaf length, LW = leaf width, LWR = leaf length to width ratio, HT = seedling height, BD = seedling basal diameter, HDR = seedling height to basal diameter ratio



Appendix 3 Regression plots for (a) leaf area with seedling height and (b) for leaf area with seedling basal diameter for the entire 179 open-pollinated families of camphor tree seedlings; LA = leaf area, HT = seedling height, BD = seedling basal diameter