

GENETIC VARIATION IN PINUS BRUTIA TEN.: WOOD DENSITY TRAITS

Bilgin Guller,^{a,*} Kani Isik,^b and Senay Cetinay^c

Trees in two provenance-progeny experimental sites of *Pinus brutia* Ten. were sampled to study variation in wood density and its components among and within six populations and to estimate heritability of wood density and ring components. Wood increment cores (12 mm thick) were collected from 29-year-old trees at breast height. A total of 1023 wood strips were scanned with the X-ray densitometry technique. There were significant differences among populations and among families (within populations) in all the characters studied. Murtbeli (M) population from the middle altitude (486 m asl) had the highest values in both ring-area-weighted density and late wood proportion (500 ± 2.7 kg/ m³ and 49 \pm 0.3 %, respectively), whereas Hacibekar (H) population from the highest altitude (1032 m asl) exhibited the lowest values (468 ± 3.0 kg/ m³, 42 \pm 0.3 %). Relatively high heritability values (h^2 between 0.45 and 0.74; h_{hs}^2 between 0.68 and 0.80) for investigated characters (except latewood density) suggest that these characteristics are under moderate to strong genetic control, and thus, by selecting populations (and families within populations) having high wood density, genetic improvement can be achieved in *P. brutia*. Correlations between wood density and other growth traits have indicated that simultaneous selection of desired genotypes for most of the wood density components is possible for the species. Latewood percentage and earlywood density present an optimal target for selection to improve wood density.

Keywords: *Pinus brutia*; Tree breeding; Heritability; Wood density

Contact information: a: Suleyman Demirel Univ., Faculty of Forestry, Dept. of Forest Product Engineering, Isparta, Turkey; b: Akdeniz Univ., Faculty of Sciences, Biology Dept., Antalya, Turkey; c: Southwest Anatolia Forest Research Institute, Antalya, Turkey;

* Corresponding author: bilginguller@sdu.edu.tr

INTRODUCTION

Pinus brutia is naturally distributed mainly in the eastern Mediterranean region: Turkey, Greece, Cyprus, Syria, Palestine, Jordan, Iraq, and northeastern neighbors of Turkey (Crimea, Azerbaijan, Ukraine and Georgia). This species has a large ecological adaptability, growing performance and can be found in a variety of different locations from sea level up to 1700 m (Anonymus 1993, 2002).

The most efficient way to meet the increasing global wood demand is to establish plantations with genetically improved seeds. Due to its relative fast growth and wide geographic distribution (Arbez 1974; Atalay et al. 1998; Boydak 2004), the major emphasis in plantations has been devoted to Turkish red pine (*Pinus brutia* Ten.) in Turkey (Guller 2007). Its wood is used in variety of ways in the forest products industry,

including timber, furniture, pulp and paper, board products, fuel wood, window and door framing, flooring, structural material in home construction, package, etc. The rotation age for pulp and paper production is about 25 years. It may go up to 80 years for timber production, depending on site quality and genetic stock (generally 60 years for good sites).

Genetic improvement of *Pinus brutia* in Turkey was started in the mid-1970s, with extensive work with tree selections and establishment of seed orchards (Isik 1986; Ozturk and Siklar 2000). The Turkish Tree Improvement Institute started progeny testing of selections in the late 1990s (Koski and Antola 1993). Most of the genetic variation studies on Turkish red pine have been focused on growth rate, stem form, and crown characteristics (Isik and Isik 1999; Isik et al. 1999; Gulcu and Celik 2009). These studies showed that there is a considerable genetic variation both within and between-populations for growth and stem quality traits, with relatively high heritability, indicating that selection on these traits will provide significant genetic gains in the plantations. Isik et al. (1999) found that variation in stem form among populations was far much greater than variation within populations for stem straightness. Populations from higher elevations had genetically straighter stems compared to those from low elevations.

Selections of trees for fast growth and deployment in intensively managed plantations have raised concerns about the quality of wood produced (Zobel and van Buijtenen 1989). The rotation ages of pine plantations have been reduced in many species, and the wood produced from such plantations is generally juvenile wood (Zobel and Sprague 1998). Juvenile wood has several undesirable characteristics, such as less strength, higher lignin content, and low specific gravity (Zobel and van Buijtenen 1989; Blair et al. 1976). Wood density is considered to be a reliable indicator for strength properties of wood and pulp and paper quality (Zobel and van Buijtenen 1989; Hashemi and Kord 2011). Therefore, improved wood density trait is expected to meet a wide range of demands and potential for future utilization. Recent studies have indicated that wood density, with high heritability and large economic gain from selection, is an ideal selection criterion for tree breeding programs because of its strong effect on yield and quality of wood (Jett et al. 1977; Hodge and Purnell 1993; Hannrup et al. 2000; Ivkovich et al. 2002; Apiolaza 2009, 2011). Heritability of wood density is generally found to be higher than those for stem form or growth traits in forest trees. Published heritability for density in pines varies from 0.40 to 0.85, compared to the usual range of 0.15 to 0.25 for many growth traits (Zobel and Jett 1995). This indicates that genetic manipulation of wood density can result in good gains. In radiata pine (*Pinus radiata*), Cown and Ball (2001) determined that heritability of wood density located at the juvenile wood sections ranges between 0.62 and 0.68. In a study conducted in slash pine (*Pinus elliottii*), Hodge and Purnell (1993) also found that the heritability of density for rings near the pith was slightly higher than outward. In contrast, Zobel and Jett (1995) stated that for loblolly pine, heritability has a clear tendency to increase with cambial age. Louzada and Fonseca (2002) reported that the highest heritability values were obtained on the earlywood components in *Pinus pinaster*.

An International symposium was organized on Turkish red pine in 1993 (Anonymus 1993), and subsequently various studies have been published on general wood properties of this species (Bektas et al. 2003; Gundogan et al. 2005; Guller 2007).

However, we have very limited knowledge on genetic variation and heritability of wood density and other ring characteristics in the species. The density range of 7-year-old *Pinus brutia* trees has been reported as 0.351 to 0.571 gcm⁻³, and both individual-tree and family heritability estimates for wood density were intermediate (0.39 and 0.55, respectively) (Yildirim et al. 2006; Ozturk et al.2008).

The objectives of this study were to partition phenotypic variance for wood density into genetics (populations, families) and environmental components, estimate genetic parameters, examine the magnitude and nature of genotype by environment interaction for wood density, find correlations among density components, and predict genetic gain from direct and indirect selection of a single trait.

EXPERIMENTAL SETTING

Materials and Methods

The wood samples for this study were collected from 30-year-old *Pinus brutia* trees at the Kepez (90 m above sea level) and Duzlercami (350 m asl) test sites near Antalya city, Turkey. The two sites are geographically close (about 10 km in bird's-eye view) to each other. The Duzlercami (Dc) site (site index:19.8 at age 25 years) is more fertile than the Kepez (K) site (site index:13.7) . The test sites are a part of a provenance+progeny trial established in 1979 (Isik 1986; Isik et al. 1999). The trials include six natural populations from two altitudinal transects (eastern transect referred to as No 1, and western transect No 2) extending from the Mediterranean coast through the Taurus Mountains. Two populations were from low [Sarilar (S, 92 m asl) and Doyran (D, 61 m); referred to as L1 and L2, respectively]; two from middle [Murtbeli (M, 486 m) and Buk (B , 481 m); referred to as M1 and M2,]; and two were from high altitudes [Kapan (K, 932 m) and Hacibekar (H, 1032 m); referred to as H1 and H2]. The field experimental design applied on the test sites was single-tree-plots randomized complete block design with three interlocked replications (Libby and Cockerham 1980; Isik 1988).

Wood samples collection

Wood cores were collected at breast height (1.3 m) and obtained by extraction of one increment core (12 mm in diameter) per tree, from pith to bark. Initially a total of 1080 wood cores were collected from the two sites, at least six cores per family per site. Immediately after removal of the wood core from a tree, the increment cores were stored in a cooler bag and subsequently vacuumed in plastic bags and stored in green condition in the refrigerator (+2 °C). The cores were divided into two radii, and then dried at room conditions. One part of the each core was glued to poplar strips (core holders) and 2 mm thick radial strips were cut. Following the cutting process, 1023 clear samples were evaluated. Diameters at breast height (dbh) of the sampled trees were also measured following the completion of core samples.

Wood properties assessed

The radial strips were conditioned to 8% equilibrium moisture content and then these wood strips were scanned using the x-ray densitometer (Quintek Measurement

Systems, Model QTRS-01X). The first annual rings from bark of each sample were not evaluated, because they were usually incomplete or damaged while removing bark. The x-ray attenuation measured by the densitometer was related to density by $\mu l = \mu m \times \rho$, where μl is the measured attenuation of the x-ray beam passed through the sample, μm is the sample mass attenuation coefficient, and ρ is density. Therefore, calculating density required the mass attenuation coefficient (cm^2/g) of the wood to be known. Calibration to the appropriate mass attenuation coefficient was conducted using a set of 35 radial strips from cores with densities previously determined by maximum moisture content method (Smith 1954). The 35 mass attenuation coefficients were averaged to provide the final value to be used for calculating wood density. The x-ray densitometer produced average ring density, minimum density, maximum density, earlywood density (EWD), latewood density (LWD), ring width (RW), and latewood percentage (LWP) from each wood strip. The fixed threshold value of 0.450 gcm^{-3} density was taken as the limit between earlywood and latewood within a ring (Guller 2010). The ring area-weighted wood density (RWD) for each tree was calculated based on average density of each ring and ring area using the following formula (Hannrup et al. 2000),

$$\text{RWD} = \sum(a_i \times W_{di}) / \sum a_i \quad (1)$$

where RWD is the ring area-weighted density, a_i is the cross-sectional area of the i th growth ring assuming that each ring is perfectly circular, and W_{di} is the wood density of the i th growth ring.

Statistical Analyses

Estimation of genetic parameters

The following linear mixed model was fit to wood increment core density values to partition observed variation into its environmental and genetic components.

$$y_{ijkl} = \mu + S_i + P_j + SP_{ij} + F(P)_{k(j)} + SF(P)_{ik(j)} + e_{ijkl} \quad (2)$$

where y_{ijkl} is the l^{th} observation of the k^{th} family of the j^{th} population in the i^{th} site; μ is the overall mean; S_i is the fixed i^{th} site effect ($i = 1, 2$); P_j is the random j^{th} population effect with $\sim \text{NID}(0, \sigma_p^2)$ ($j = 1, \dots, 6$); SP_{ij} is the random i th site by j^{th} population interaction effect with $\sim \text{NID}(0, \sigma_{sp}^2)$; $F_{k(j)}$ is the k^{th} family effect nested within j^{th} population with $\sim \text{NID}(0, \sigma_{f(p)}^2)$ ($k = 1, \dots, f$); $SF(P)_{ik(j)}$ is k^{th} family by i th site interaction nested within j^{th} population $\sim \text{NID}(0, \sigma_{sf(p)}^2)$; and e_{ijkl} is the random error associated with each observation with $\sim \text{NID}(0, \sigma_e^2)$.

In the expression $y = X\beta + Z\gamma + \varepsilon$, y is the vector of observations, X is the incidence matrix of fixed effects, β is the vector of fixed effects, Z is the incidence matrix of random effects, γ is the vector of random effects, and ε is the vector of residuals. The variance of response variables (vector y) is $\text{Var}(y) = ZGZT + R$. The $G = (\sigma_p^2 I, \sigma_{sp}^2 I, \sigma_{f(p)}^2 I, \sigma_{sf(p)}^2 I)$ is the diagonal variance-covariance matrix of random effects with the variances of random effects in the diagonal and zero covariances in the off diagonal. The I is the identity matrix. The R is the variance-covariance matrix of random errors

($R=\sigma^2In$). Variance components were estimated using the SAS Mixed procedure (SAS Institute 1996).

Individual-tree (h_i^2) and half-sib family mean (h_{hs}^2) heritabilities were estimated as follows:

$$h_i^2 = \frac{4\sigma_{f(p)}^2}{\sigma_{f(p)}^2 + \sigma_{sf(p)}^2 + \sigma_e^2} \quad (3)$$

$$h_{hs}^2 = \frac{\sigma_{f(p)}^2}{\sigma_{f(p)}^2 + \frac{\sigma_{sf(p)}^2}{t} + \frac{\sigma_e^2}{tn}} \quad (4)$$

where, $\sigma^2f(p)$ is the general combining ability variance (GCA), $\sigma^2sf(p)$ is the variance of GCA by site interaction, σ^2e is the random error variance, t is the number of sites ($t=2$), and n is the harmonic mean number of trees per site ($n \sim 8.5$). Standard errors of heritabilities were estimated by the Delta Method (Lynch and Walsh 1998) and calculated by a SAS IML code (Isik et al. 2008).

We also estimated type B genetic correlation for wood density as $r_B = \sigma^2f(p) / [\sigma^2f(p) + \sigma^2sf(p)]$ to examine the extent of genotype by environment interaction for the species. In order to estimate genetic parameters for each site, a reduced model was fit to data of each site. In such a model, population by site and family by site interactions terms were dropped from Eq. 2. Similarly, heritability estimates and repeatability of family means for individual sites did not include the site by family interaction term and were estimated as $h_i^2 = 4\sigma^2f(p) / [\sigma^2f(p) + \sigma^2e]$ and $h_{hs}^2 = \sigma^2f(p) / [\sigma^2f(p) + \sigma^2e/n]$, respectively.

Correlations among the variables

We also estimated phenotypic and genetic correlations among wood density-wood density components. Such correlations could tell us about the relationships between growth rates of trees and wood density of the species. The phenotypic correlations estimated using the CORR procedure in SAS. Genetic correlations ($r_{g(x,y)}$) between traits x and y and standard errors were estimated using following formulas derived from Falconer and Mackay (1996). (Some of abbreviations we used are different from the original formulas)

$$r_{g(x,y)} = \frac{Covf(x,y)}{\sqrt{\sigma^2f(x) * \sigma^2f(y)}} \quad (6)$$

$Covf(x,y)$: cross-covariance, $\sigma^2f(x)$, $\sigma^2f(y)$, are the family variance for traits.

$$SE(r_g) = \frac{1 - r_g^2}{\sqrt{2}} \sqrt{\frac{SE_{(h^2x)} SE_{(h^2y)}}{(h^2x)(h^2y)}} \quad (7)$$

where $SE(r_g)$ is the standard error of genetic correlation, r_g is the genetic correlation between traits (x and y), $SE(h^2_x)$ and $SE(h^2_y)$ are standard errors of heritability estimates for trait x and y, and (h^2_x) , (h^2_y) are heritability estimates for trait x and y.

The predicted genetic gain (ΔG) from direct selection of a single trait and the predicted correlated genetic responses of indirect selection were calculated according to Falconer and Mackay (1996). The heritability (h^2) is the ratio of additive genetic variance (σ^2_A) to phenotypic variance (σ^2_P), it is of key importance in estimating genetic gains which determined by the heritability of the trait and selection differential is dependent on two factors selection intensity (i) and phenotypic standard deviation (σ_P), therefore selection differential, $S=i\sigma_P$, and predicted genetic gain calculated as $\Delta G=ih^2\sigma_P$. The correlated response to selection is calculated as $CR_y=ih_x h_y r_g \sigma_{P_y}$, where CR_y is the correlated response to selection of trait y given selection for trait x; i is the selection intensity; h_x and h_y are the heritabilities of traits x and y, respectively; r_g is the genetic correlation between traits x and y; and σ_{P_y} is the phenotypic standard deviation of the sample population for trait y.

RESULTS AND DISCUSSION

Variation Across Sites and Among Populations

Diameter (dbh) variation

One of mid-altitude population (M1) and a high-elevation populations H2 were significantly different from each other at Duzlercami (Fig. 1, Table 1). The rank order was M1, L1, H1, L2, M2, and H2). However, there were no statistically significant differences among the populations L1, L2, and H1 at the Duzlercami site.

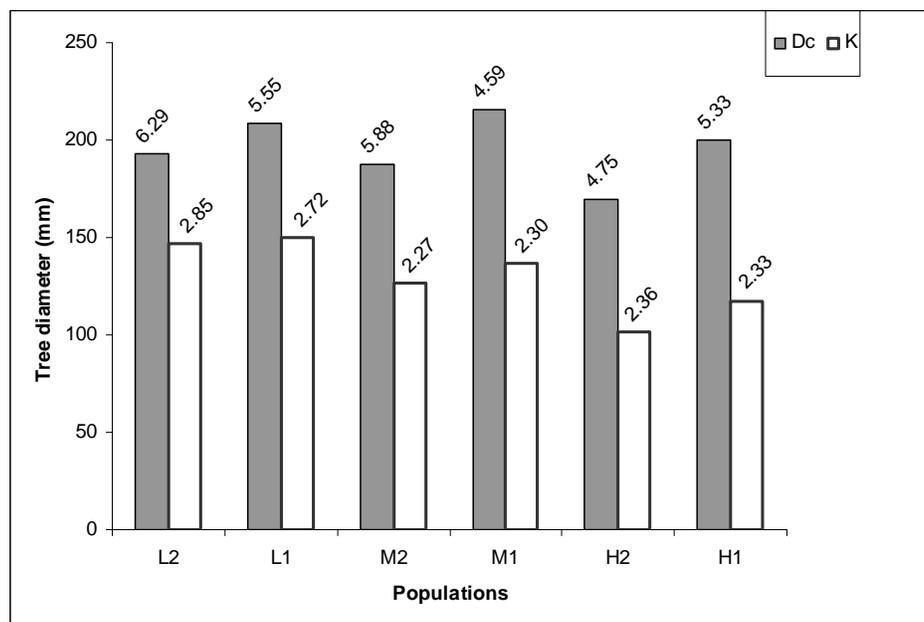


Fig. 1. Variation of diameter (dbh) growth among six populations of *Pinus brutia* at two common garden test sites (standard errors were given at the top of each bars)

The site ranks based on total radial growth were not similar at the two sites. The rank order at Kepez was L1, L2, M1, M2, H1, and H2. The relationship between radial diameter and altitude of populations' origins showed different characteristics for the two sites (Fig. 2). H2 population coming from high altitude, western transect had the smallest radial increment in both sites. M2 population coming from mid altitude of western transect, also had relatively small radial increment in Duzlercami. The relationship between radial diameter and altitude of populations' origins shows a polynomial curve characteristic in Duzlercami because of the fact that low and high altitude populations show lower radial increment than mid-altitude populations (Fig. 2). Population M1 responded to the greatest degree in radial growth rate as the site quality improved.

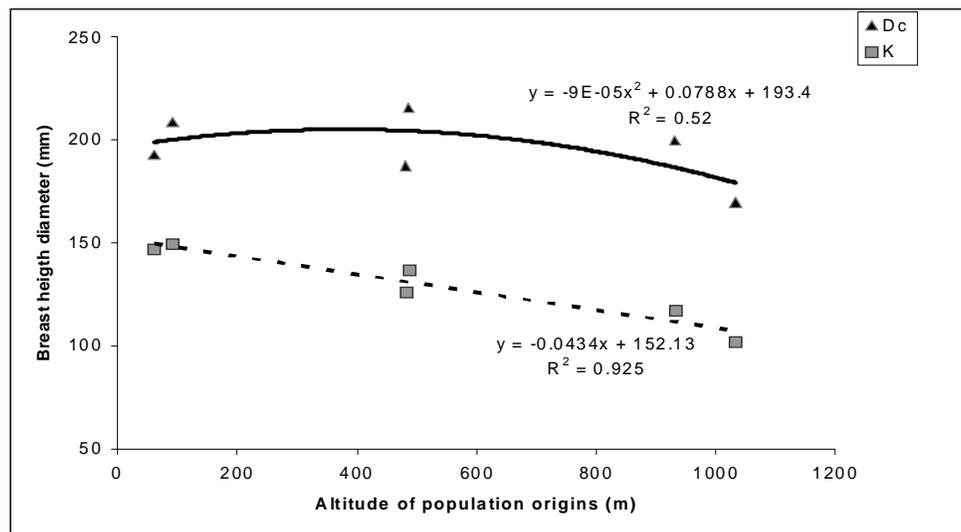


Fig. 2. Relationships between breast height diameter and altitude of populations' origins for *Pinus brutia* at two common garden test sites (in the graph one data point represents the mean of at least 76 data points)

Density variation among and within populations

Middle and low elevation populations (M2, M1, L1, and L2) had higher wood density compared to populations H1 and H2 (coming from higher elevations). Despite considerable growth differences between the two sites, the populations generally had similar frequency distribution and ranking for wood density (Figs. 3 and 4). The high latewood group consisted of M1, L2, M2, and L1, both in the Duzlercami and in the Kepez sites. Latewood densities for all populations in the Duzlercami site (where site quality is better) were higher than the Kepez site. There were no significant differences between the two sites for early wood densities. Ring-area-weighted density values were higher for populations from mid- and low-elevations than high-elevation populations (Table 1). The relationship between RWD and altitude of populations' origins showed similar characteristic for two sites (Fig. 5). Populations coming from high altitude had the lowest RWD in both sites. Three of six populations (L1, M1, and M2) showed no significant change in RWD between the test sites. In particular the M1 population was very unique among all the populations because of the fact that it exhibited the highest response (80%) of radial increment at better site without any decline in density.

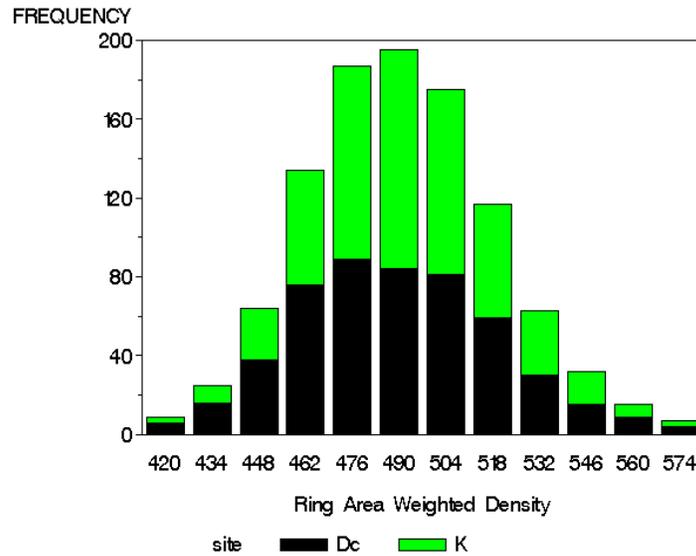


Fig. 3. Frequency distribution of ring-area-weighted wood density (RWD) for *Pinus brutia* at two common garden test sites

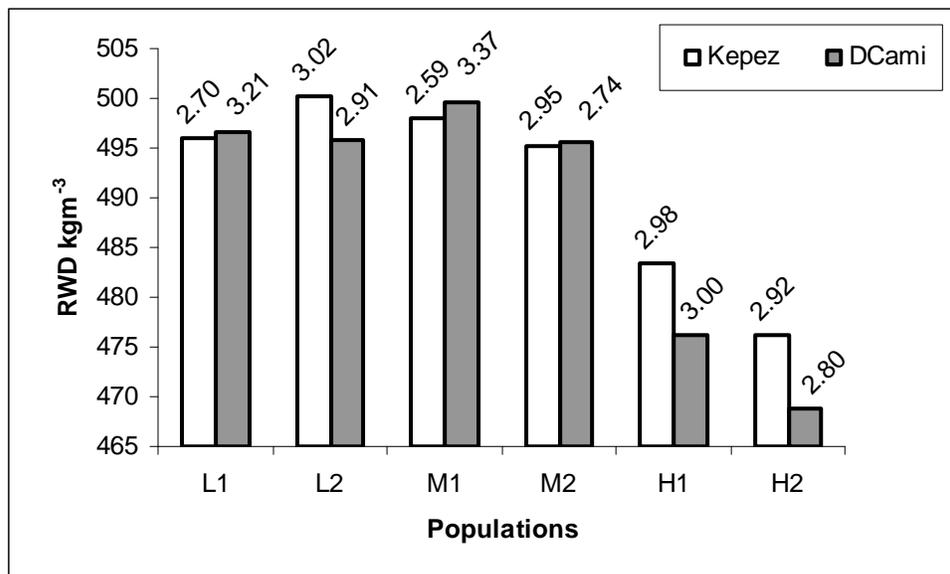


Fig. 4. Comparisons of ring-area-weighted density (RWD) among six populations of *Pinus brutia* at two common garden test sites (standard errors were given at the top of each bars)

Partitioning of variance for wood density showed that population (12%) and family (11%), were important sources of variations (Fam*Pop 1%). ANOVA results showed that differences among and within populations were significant for each site. The contribution of population variance to total variance was 16% at Duzlercami and 8% at Kepez. The corresponding values for family variance were 11% and 12% at Duzlercami and Kepez, respectively. The results showed that density variations due to family were higher than those due to populations at the Kepez site.

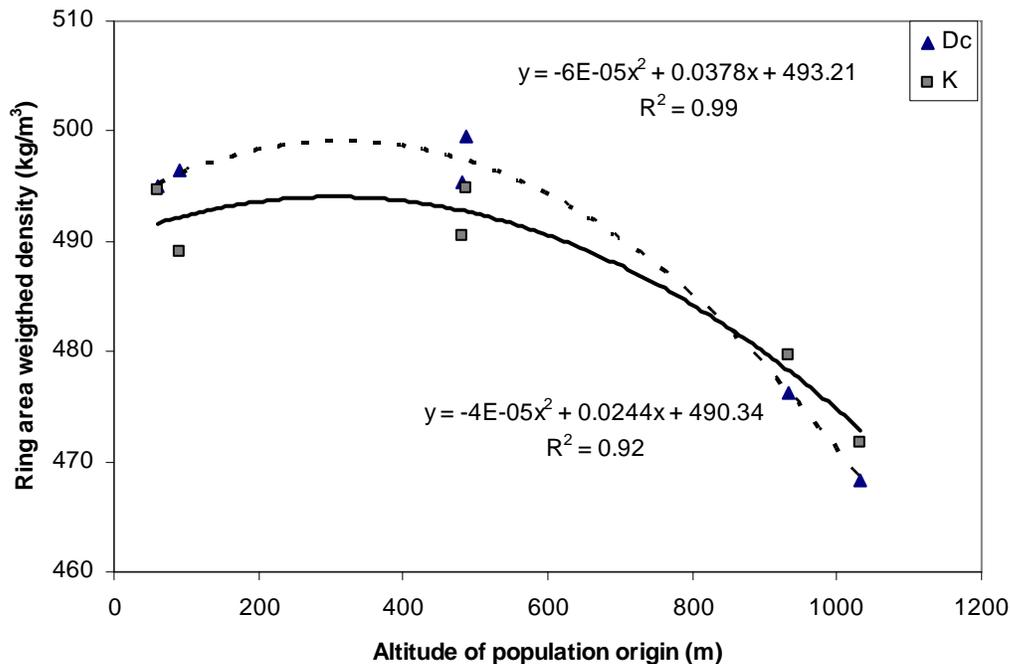


Fig. 5. Relationship between ring-area-weighted density and altitude of populations' origins for *Pinus brutia* at two common garden test sites (in the graph one data point represents the mean of at least 76 observations)

We determined no significant changes in EWD and RWD, slight increase in LWD for most of populations in two sites (Table 1). On the other hand Gundogan et al. (2005) found that air-dry and oven dry densities of *P. brutia*, based on limited tree samples of 80-year-old trees at natural stands, decreased with increasing site index.

Correlations among radial growth and density components

RWD was significantly (and positively) correlated with LWP, LWD and EWD traits at the both test sites (Table 2). On the other hand, correlations between RW and LWP, and also between RW and EWD were negative and significant. However, the relationship between RW and LWD was positive and significant (Table 2.). EWD was also highly correlated (positive) with LWP. Similar results were reported for *Pinus taeda* (Megraw 1985).

Moderate to high genetic and phenotypic correlation coefficients of RWD, LWP, EWD, and LWD indicate that the selection of one of these components will result in a substantial correlated response in the others. Among all pairs, high correlation coefficients were found between RWD and LWP, and RWD and EWD. In this way, it can be expected that the LWP increase selection would a correlated response for RWD and EWD. Ring density is more dependent on earlywood density than latewood ones. The importance of earlywood density was also confirmed for *Pinus pinaster* (Louzada 2002; Gaspar et al. 2008).

Table 1. Means*, Standard Errors (SE, in parenthesis), and Coefficient of Variations (% CV, *italics*) for Wood Density Traits by Populations in *Pinus brutia* at two Test Sites

Populations	Traits					
	Dbh mm	RWD kg/m ³	RW mm	LWP %	LWD kg/m ³	EWD kg/m ³
Site: Duzlercami (350 m asl, site index =19.8 at age 25 years)						
L1	208.54 b (5.55) <i>28.81</i>	496.50 a (2.91) <i>5.44</i>	3.78 b (0.13) <i>30.43</i>	46.78 b (0.36) <i>30.22</i>	630.53 a (1.39) <i>8.75</i>	390.17 b (0.60) <i>6.11</i>
L2	193.02 bc (6.29) <i>35.25</i>	495.00 a (3.21) <i>5.74</i>	3.68 bc (0.13) <i>29.47</i>	47.51 ba (0.40) <i>31.36</i>	625.25 b (1.49) <i>8.96</i>	388.59 b (0.68) <i>6.53</i>
M1	215.50 a (4.59) <i>25.31</i>	499.58 a (2.74) <i>5.27</i>	4.02 a (0.10) <i>23.95</i>	48.47 a (0.33) <i>28.05</i>	630.26 a (1.35) <i>8.93</i>	392.69 a (0.59) <i>6.24</i>
M2	187.40 c (5.88) <i>34.65</i>	495.35 a (3.37) <i>6.19</i>	3.58 c (0.12) <i>31.38</i>	48.43 a (0.36) <i>28.43</i>	629 ba (1.53) <i>9.54</i>	389.50 b (0.64) <i>6.37</i>
H1	199.87 b (5.33) <i>29.60</i>	476.21 b (2.80) <i>5.54</i>	3.73 b (0.11) <i>26.45</i>	43.60 c (0.33) <i>31.12</i>	616.82 c (1.36) <i>9.03</i>	381.14 c (0.59) <i>6.35</i>
H2	169.58 d (4.75) <i>31.55</i>	468.38 b (3.00) <i>5.70</i>	3.30 d (0.09) <i>23.58</i>	42.12 d (0.33) <i>30.24</i>	618.37 c (1.52) <i>9.48</i>	378.33 d (0.64) <i>6.51</i>
Site: Kepez (90 m asl, site index =13.7 at age 25 years)						
L1	149.72 a (2.72) <i>23.48</i>	489.03 a (3.02) <i>5.87</i>	2.34 a (0.05) <i>20.70</i>	45.60 d (0.36) <i>33.11</i>	616.70 b (1.26) <i>8.72</i>	390.31 b (0.54) <i>5.84</i>
L2	146.79 a (2.85) <i>26.29</i>	494.72 a (2.70) <i>5.08</i>	2.32 a (0.06) <i>23.50</i>	48.51 b (0.37) <i>31.68</i>	613.69 b (1.24) <i>8.40</i>	391.85 ba (0.56) <i>5.90</i>
M1	137.05 b (2.96) <i>25.35</i>	494.91 a (2.95) <i>5.77</i>	2.26 b (0.05) <i>19.71</i>	48.36 b (0.35) <i>30.99</i>	620.48 a (1.23) <i>8.48</i>	392.21 a (0.51) <i>5.58</i>
M2	126.32 c (2.27) <i>24.52</i>	490.52 a (2.59) <i>4.86</i>	2.11 c (0.05) <i>20.21</i>	49.66 a (0.37) <i>29.86</i>	616.59 b (1.31) <i>8.56</i>	389.80 c (0.54) <i>5.52</i>
H1	117.62 c (2.33) <i>26.74</i>	479.69 b (2.92) <i>5.51</i>	2.11 c (0.05) <i>19.82</i>	47.15 c (0.41) <i>33.08</i>	607.02 c (1.40) <i>8.94</i>	388.81 c (0.63) <i>6.12</i>
H2	101.92 d (2.36) <i>26.75</i>	471.79 b (2.98) <i>5.62</i>	1.99 d (0.05) <i>20.02</i>	45.84 d (0.40) <i>33.37</i>	605.51 c (1.49) <i>9.51</i>	380.47 d (0.67) <i>6.74</i>
* Means with the same letter in a given trait within a given test site are not significantly different from each other at the 5% level according to Duncan Test.						

Table 2. Phenotypic (above diagonal) and Genetic (below diagonal) Correlations among Wood Density Components for *Pinus brutia* at Two Test Sites (Standard errors are given in parentheses)

Duzlercami	RWD	RW	LWP	LWD	EWD
RWD	1	0.006	0.824**	0.416**	0.716**
RW	0.006 (0.003)	1	-0.219**	0.285**	-0.297**
LWP	0.913 (0.050)	-0.229 (0.04)	1	0.034	0.747**
LWD	0.454 (0.060)	0.300 (0.07)	0.036 (0.006)	1	-0.045
EWD	0.845 (0.090)	-0.332 (0.03)	0.850 (0.080)	-0.050 (0.020)	1
Kepez	RWD	RW	LWP	LWD	EWD
RWD	1	-0.053	0.684**	0.374**	0.594**
RW	-0.029 (0.003)	1	-0.314**	0.258**	-0.274**
LWP	0.711 (0.170)	-0.165 (0.03)	1	-0.142**	0.782**
LWD	0.394 (0.070)	0.138 (0.08)	-0.144 (0.08)	1	-0.282**
EWD	0.625 (0.190)	-0.146 (0.03)	0.790 (0.12)	-0.289 (0.07)	1

** Correlation is significant at the 0.01 level

Ring width influences economic return of plantation programs. Although a lot of research has been done on the correlation between wood density and radial growth rate over the years, a general relationship is still ambiguous. Despite the very widespread interest and research, there still are controversial reports in the literature. Zobel and van Buijtenen (1989) summarized these correlations for hard pines, showing 59 references. Among these references 35 of them showed no relationship between radial growth rate and specific gravity, 9 exhibited a small correlation, while 11 showed a significant reduction in specific gravity with faster growth rate, and only 4 of them showed a higher specific gravity for the faster growing trees. Chaperon et al. (1992) suggested a positive relationship between density and ring width in *Pinus pinaster*. Even though the correlation coefficients are low, the result (ring width is positively correlated at genetic and phenotypic level with average ring density for *Pinus pinaster*) was confirmed later by Louzada (2003). However, Bouffier et al. (2009) found significantly lower (-2.0% and -4.5%) wood density for the two improved (+24.0% and +20.6% for volume) varieties of maritime pine. In some cases conflicting results were reported for the same species. For example, Wilcox et al. (1975) found a weak negative correlation between density and diameter in *Pinus radiata*. On the other hand, Cown et al. (2002) did not indicate any obvious relationship between growth rate and density at the same species.

Heritability Estimates

Individual-tree (h_i^2) and half-sib family mean (h_{hs}^2) heritabilities and standard errors (given in parentheses) for wood density on pooled data were estimated as 0.51 (0.133) and 0.70 (0.081). We also estimated a type B genetic correlation for wood density as 0.95 (0.022). The high estimate of type B genetic correlation implied limited operational importance of genotype x environment interaction. In order to estimate genetic parameters for each site, a reduced model was fit to data of each site.

Moderate to high heritability estimates were obtained for wood density, earlywood density, latewood percentage, and ring width. However, latewood density heritability estimates were quite low in each of the two sites (Table 3).

Table 3. Heritability Estimates (and Standard Errors) for *Pinus brutia* at 2 Sites

Site	Trait	h_i^2 (S.E)	h_{hs}^2 (S.E)
Duzlercami	RWD	0.53 (0.15)	0.72 (0.21)
	LWD	0.14 (0.10)	0.38 (0.28)
	EWD	0.74 (0.29)	0.80 (0.31)
	LWP	0.62 (0.28)	0.76 (0.34)
	RW	0.49 (0.20)	0.70 (0.29)
Kepez	RWD	0.55 (0.16)	0.73 (0.21)
	LWD	0.09 (0.08)	0.27 (0.25)
	EWD	0.65 (0.25)	0.77 (0.30)
	LWP	0.57 (0.27)	0.74 (0.36)
	RW	0.45 (0.21)	0.68 (0.31)

The highest predicted genetic gain (ΔG) from direct selection of single trait for RWD was found as 8% for Duzlercami site and 6% for Kepez site. The second highest ΔG was EWD (5%). Predicted genetic gain of direct selection of RW was found to be quite low (1%). We also calculated the predicted correlated genetic responses of indirect selection among traits. They were quite similar for both site as RWD-EWD (6.9%), RWD- LWP (7.2%) and RWD-RW (0.8%). RWD was under strong genetic control (Table 3), which is consistent with results on some other *Pinus* species with moderate to high heritability estimates ranging from 0.38 to 0.85 (Talbert et al. 1983; Zobel and Jett 1995; Louzada and Fonseca 2002; Kumar 2002; Gaspar et al.2008; Gapare et al. 2009). Heritability estimates for EWD (Table 3) were higher than those for LWD. This is a parallel and a confirming result that previously published for *Pinus pinaster*, earlywood characteristics are subject to stronger genetic control and that latewood components are more subject to environmental factors (Gaspar et al. 2008). According to Lebourgeois (2000) LWD appears to be more sensitive to climatic fluctuations. In temperate zones earlywood tracheids, which have thin cell walls with a large lumen diameter, are formed earlier in the growing season when conditions (water availability, temperature and photoperiod) are optimal for tree growth. During these optimal conditions genetic effects become prevalent. On the other hand latewood, which has narrower lumens and thicker cell walls, formed later in vegetation period (in summer or fall) when cambial cell division and expansion rates decline. Because of high temperatures and water stress, growth conditions are less stable in summer, which may increase the phenotypic variance and lead to reduced heritability (Plomion et al. 2001).

In this study, density components varied significantly among populations and families and displayed high genetic control (except LWD); EWD density heritability found considerably higher than that of LWD. These results are consistent with studies on *Pinus pinaster* (Louzada and Fonseca 2002), *Pinus radiata* (Kumar 2002), and all of which indicate that EWD is the component most dependent on genetic effects. Second highest heritability estimates was for LWP, whereas RWD -LWP correlation was positive and strong. Since latewood has higher density than earlywood, a higher LWP would contribute to higher ring density. This means that LWP, which is relatively easy to measure, would be a good target for wood density selection in the species. However, the proportion of earlywood is considerably greater than that of latewood for larger rings, and considerably higher EWD contribute more to ring density. Therefore, selection

including LWP and EWD may present an optimal target to improve mean density and density homogeneity within ring.

When the trees were 13 and 17 years old at the Duzlercami test site, Isik and Isik (1999) and Isik et al (1999) evaluated several growth, biomass, stem quality, and branching traits of *P. brutia*. Their results showed that “populations originating from mid altitudes showed better growth, exhibited more desirable bole straightness, and allocated a higher proportion of biomass to the stem”. The present study showed that mid-elevation populations also exhibited more desirable wood density characteristics, which means that mid-elevation populations are preferable for further breeding and plantating of *P. brutia*.

CONCLUSIONS

The study results showed that wood density and wood density components displayed moderate to high genetic control (except for latewood density). Wood density components (ring density, earlywood density, late wood density and percent latewood) were strongly correlated. No statistically significant and/or strong relations were found between ring density and ring width. Earlywood density showed higher genetic and phenotypic correlations with ring density than latewood density. Although differences among and within populations were statistically significant for each site, coefficients of variation for density traits were markedly smaller than those of radial growth traits. Thus, wood density components were less variable (<10%) than latewood percentage and ring width traits (>20%) in and among populations. In operational forestry this means that while there may be greater variation among trees in terms of their growth rates, trees exhibit lesser variation in terms of their wood density traits. Wood density of *P. brutia* can be improved by increasing either latewood portion, earlywood density, or both. Latewood percentage, which is relatively easy to measure and highly variable, would be a good target for wood density selection in the species. However, the proportion of earlywood is considerably greater than that of latewood for larger rings, and higher earlywood contributes more to ring density. Therefore, combining selection of latewood percentage and earlywood density may present an optimal target to improve mean density and density homogeneity within ring.

ACKNOWLEDGMENTS

The study was supported by The Scientific and Technological Research Council of Turkey (TUBITAK) under a project (106O442) and a post-doctoral research grant to Dr. Bilgin Guller at North Carolina State University, Raleigh, N.C., USA. We wish to thank Dr. Fikret Isik, Dr. Steve McKeand, and staff of NCSU Tree Improvement Program. We also thank Yusuf Kurt, Adnan Guller, and Asli Gocmen for their help during sampling of the wood cores. The test site was established by Dr Kani Isik in 1979, and maintained and cared through the years by the staff of Forest Service and Southwest Anatolia Forest Research Institute in Antalya. We thank Akdeniz University Scientific Research Fund and Suleyman Demirel University for the use of laboratory facilities.

REFERENCES CITED

- Anonymus, (1993). "International *Pinus brutia* symposium proceedings," Ministry of Forestry, Marmaris, Turkey.
- Anonymus, (2002). *Pines of Silvicultural Importance*, CAB International publishing, New York, USA.
- Apiolaza, L. A. (2009). "Very early selection for solid wood quality: Screening for early winners," *Annals of Forest Science* 66(6), 601, 1-10.
- Apiolaza, L.A., Shakti S. C., and Walker, J. C. F. (2011). "Genetic control of very early compression and opposite wood in *Pinus radiata* and its implications for selection," *Tree Genetics & Genomes* 7, 563-571.
- Arbez, M. (1974). "Distribution, ecology and variation of *Pinus brutia* in Turkey," *FAO Forest Genetics Resources Information* 3, 21-23.
- Atalay, I., Sezer, I., and Cukur, H. (1998). "The ecologic properties of *P. brutia* Ten. forests and their regioning in terms of seed transfer," Turkish Ministry of Forestry Tree Breeding Institute Publishing No: 6 (Turkish and English)
- Bektas, I., Alma, M. H., Goker, Y., Yuksel, A., and Gundogan, R. (2003). "Influence of site on sapwood and heartwood ratios of Turkish calabrian pine," *Forest Products Journal* 53(4), 48-50.
- Blair, R. B., Zobel, B., Hitchings, R. C., and Jett, J. B. (1976). "Pulp yield and physical properties of young loblolly pine with high-density juvenile wood," *Applied Polymer Symp.* 28, 435-444.
- Bouffier, L., Raffin, A., Rozenberg, P., Meredieu, C. and Kremer, A. (2009). "What are the consequences of growth selection on wood density in the French maritime pine breeding programme?" *Tree Gen. Genom.* 5, 11–25.
- Boydak, M. (2004). "Silvicultural characteristics and natural regeneration of *Pinus brutia* Ten.- A review," *Plant Ecology* 171, 153-163.
- Chaperon, H., Castera, P., and El Ouadrani, A. (1992). "Qualité du bois de pin maritime: Comment l'améliorer ?" *AFOCEL-ARMEF, Informations-Forêt* 433(2), 113-130.
- Cown, D. J., and Ball, R. D. (2001). "Wood densitometry of 10 *Pinus radiata* families at seven contrasting sites: Influence of tree age, site, and genotype," *N. Z. J. For. Sci.* 31, 88-100.
- Cown, D. J., McKinley, R. B., and Ball, R. D. (2002). "Wood density variation in 10 mature *Pinus radiata* clones," *New Zealand Journal of Forestry Science* 32 (1), 48-69.
- Falconer, D. S., and Mackay, T. F. C. (1996). *Introduction to Quantitative Genetics*, Fourth Edition, Longman Group Ltd., Essex, England, 464 p.
- Gapare, W. J., Ivkovich, M., Baltunis, B. S., Matheson, A. C., and Wu, H. X. (2009). "Genetic stability of wood density and diameter in *Pinus radiata* D. Don plantation estate across Australia," *Tree Genetics & Genomes* 6, 113-125.
- Gaspar, M. J., Louzada, J. L., Silva, M. E., Aguiar, A., and Almeida, M. H. (2008). "Age trends in genetic parameters of wood density components in 46 half-sibling families of *Pinus pinaster*," *Can. J. For. Res.* 38, 1470-1477.

- Gulcu, S., and Celik, S. (2009). "Genetic variation in *Pinus brutia* Ten seed stands and seed orchards for growth, stem form and crown characteristics," *African J. of Biotechnology* 8(18), 4387-4394.
- Guller, B. (2007). "The effects of thinning treatments on density, MOE, MOR and maximum crushing strength of *Pinus brutia* Ten. wood," *Ann. For. Sci.* 64, 467-475
- Guller, B. (2010). "Determining wood density of Turkish red pine (*Pinus brutia* Ten.) by using x-ray densitometer," *SDU Forestry Faculty Journal* 2, 97-109.
- Gundogan, R., Bektas, I., Alma, M. A., and Yuksel, A. (2005). "Relationship between site index and some physical properties of calabrian pine," *Forest Products Journal* 55(1), 45-48.
- Hannrup, B., Ekberg, I., and Persson, A. (2000). "Genetic correlations among wood, growth capacity and stem traits in *Pinus sylvestris*," *Scan. J. For. Res.* 15, 161-170.
- Hashemi, H. K. S., and Kord, B. (2011). "Variation of within-stem biometrical and physical property indices of wood from *Cupressus sempervirens* L.," *BioResources* 6(2), 1843-1857.
- Hodge, G. R., and Purnell, R. C. (1993). "Genetic parameter estimates for wood density, transition age, and radial growth in slash pine," *Can. J. For. Res.* 23, 1881-1891.
- Isik, F., Gumpertz, M., Li, B., Goldfarb, B., and Sun, X. (2008). "Analysis of cellulose microfibril angle (MFA) using a linear mixed model in *Pinus taeda* clones," *Canadian J. Forest Research* 38, 1676-1689.
- Isik, F., Isik, K., and Lee, J. S. (1999). "Genetic variation in *Pinus brutia* Ten in Turkey: I. Growth, biomass and stem quality traits," *Forest Genetics* 6(2), 89-99.
- Isik, K. (1986). "Altitudinal variation in *Pinus brutia* Ten.: Seed and seedling characteristics," *Silvae Genetica* 35 (2-3), 58-66.
- Isik, K. (1988). "Interlocked honeycomb design with three replications and its application on a population genetics analysis of Turkish red pine (*Pinus brutia* Ten.)," *Doga Turk Tarim ve Ormancilik Dergisi* 12(3), 377-386 (in Turkish with English summary)
- Isik, K., and Isik, F. (1999). "Genetic variation in *Pinus brutia* Ten in Turkey: II. Branching and crown traits," *Silvae Genetica* 48(6), 293-302.
- Ivkovich, M., Namkoong, G., and Koshy, M. (2002). "Genetic variation in wood properties of interior spruce. I. Growth, latewood percentage, and wood density," *Can. J. For. Res.* 32, 2116-2127.
- Jett, J. B., Weir, R. J., and Barker, J. A. (1977). "The inheritance of cellulose in loblolly pine," TAPPI For. Biol. Comm. Meeting, Madison, Wisconsin, 4 pp.
- Koski, V., and Antola, J. (1993). "National Tree Breeding and Seed Production Program for Turkey (1994–2003)," Forest Seeds and Trees Improvement Research Directorate, Ankara.
- Kumar, S. (2002). "Earlywood–latewood demarcation criteria and their effect on genetic parameters of growth ring density components and efficiency of selection for end-of-rotation density of radiata pine," *Silvae Genet.* 51, 241-246.
- Lebourgeois, F. (2000). "Climatic signals in earlywood, latewood and total ring width of Corsican pine from western France," *Ann. For. Sci.* 57, 155-164, [doi:10.1051/forest:2000166](https://doi.org/10.1051/forest:2000166).
- Li, L., and Wu, H. X. (2005). "Efficiency of early selection for rotation-aged growth and wood density traits in *Pinus radiata*," *Can. J. For. Res.* 35, 2019-2029.

- Libby, W. J., and Cockerham, C. C. (1980). "Random non-contiguous plots in interlocking field layouts," *Silvae Genetica* 29(5/6), 183-190.
- Louzada, J. L. P. C., and Fonseca, F. M. A. (2002). "The heritability of wood density components in *Pinus pinaster* Ait. and the implications for tree breeding," *Ann. For. Sci.* 59, 867-873.
- Louzada, J. L. P. C. (2003). "Genetic correlations between wood density components in *Pinus pinaster* Ait.," *Ann. For. Sci.* 60, 285-294.
- Lynch, M., and Walsh, B. (1998). *Genetics and Analysis of Quantitative Traits*, Sinauer Associates, Inc., Sunderland, MA, 980 p.
- Ozturk, H., and Siklar, S. (2000). "Turkiye milli ağaç islahi ve tohum uretimi programi: Ozellikleri ve gerceklestirilen calismalar," *Orman Agacları ve Tohumları Islah Arastirma Mdl Dergisi* 1, 1-41.
- Ozturk, H., Yıldırım, K., Siklar, S., Ezeni, T., Alan, M., Ilter, E., Balkız, O. D., and Kaya, Z. (2008). "Kizilcam (*Pinus brutia* Ten.) Akdeniz Bolgesi alcak rakim (0-400 m) ıslah zonu dol denemesinde gecenç odun yogunluguna iliskin genetik parametreler," Çevre ve Orman Bakanlığı, OATIAM Yayın no: 34, 48 ss.
- Plomion, C., Leprovost, G., and Stokes, A. (2001). "Wood formation in trees. Plant Physiol," 127, 1513-1523, doi:10.1104/pp.127.4. 1513. PMID:11743096.
- SAS Institute Inc. (1996). "SAS/STAT Software: Changes and enhancements (through release 6.11)," SAS Institute Inc., Cary, N.C.
- Smith, D. M. (1954). "Maximum moisture content method for determining specific gravity of small wood samples," USDA For. Serv. For. Prod. Lab. Bull. 2014.
- Talbert, J. T., Jett, J. B., and Bryant, R. L. (1983). "Inheritance of wood specific-gravity in an unimproved loblolly pine population: 20 years of results," *Silvae Genet.* 32, 33-37.
- Vargas-Hernandez, J., and Adams, W. T. (1992). "Age-age correlations and early selection for wood density in young coastal Douglas-fir," *Forest Sci.* 38, 467-478.
- Wilcox, M. D., Shelbourne, C. J. A., and Firth, A. (1975). "General and specific combining ability in eighth selected clones of radiata pine," *New Zealand Journal of Forestry Science* 5(2), 219-225.
- Yildirim, K., Ozturk, H., Siklar, S., and Kaya, Z. (2006). "Inheritance of wood specific gravity and its genetic correlation with growth traits in young *Pinus brutia* progenies," IUFRO-Division 2 Joint Conference: Low Input Breeding and Genetic Conservation of Forest Tree Species. 9-13 Oct 2006, Belek, Antalya-Turkey, in Abstracts, p: 61
- Zobel, B. J., and Van Buijtenen, J. P. (1989). *Wood Variation: Its Causes and Control*, Springer-Verlag, Berlin, Springer Series in wood science, 363 p., ISBN: 038750298X.
- Zobel, B. J., and Jett, J. B. (1995). *Genetics of Wood Formation*, Springer-Verlag, 337 p.
- Zobel, B. J., and Sprague, J. R. (1998). *Juvenile Wood in Forest Trees*, Springer-Verlag, Berlin, Springer Series in wood science, 300 p., ISBN: 3-540-64032-0

Article submitted: July 6, 2011; Peer review completed: August 7, 2011; Revised version received and accepted: August 20, 2011; Published: August 23, 2011.