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Within-stem variations in mechanical properties of *Melia azedarach* planted in northern Vietnam

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Abstract

Within-stem variations in the mechanical properties of 17–19-year-old *Melia azedarach* planted in two sites in northern Vietnam were examined by destructive and nondestructive methods. Wood samples were collected from 10, 50, and 90% of the radial length from pith on both sides (North and South) at 0.3, 1.3, 3.3, 5.3, and 7.3 m heights above the ground. The mean values in whole trees of wood density (WD), modulus of rupture (MOR), modulus of elasticity (MOE), and dynamic modulus of elasticity (E_d) at 12% moisture content were 0.51 g/cm³, 78.58 MPa, 9.26 GPa, and 10.93 GPa, respectively. Within the stem, the radial position was a highly (p < 0.001) significant source of variation in mechanical properties. MOR, MOE, and E_d increased from pith to bark. WD had a strong positive linear relationship with both MOR (r = 0.85, p < 0.001) and MOE (r = 0.73, p < 0.001). This suggests that it is potentially possible to improve mechanical properties through controlling WD. MOR had also a strong linear relationship with E_d (r = 0.84, p < 0.001). This indicates that E_d is a good indicator to predicting the strength of wood if the density of measured element is known. Besides, the stress wave method used in this study provides relatively accurate information for determining the stiffness of *Melia azedarach* planted in northern Vietnam.

Keywords Melia azedarach · Mechanical properties · Nondestructive evaluation · Radial position

Introduction

Melia azedarach is an important fast-growing plantation species in Vietnam. It belongs to the Meliaceae family [1]. The wood has been used for manufacture agricultural implements, plywood, boxes, poles, tool handles, furniture, and light construction materials [2–4]. However, the published information is insufficient for a detailed quantification in mechanical properties and for the potential variation to be found within tree of Melia azedarach. A better understanding of the wood variability within tree is of value to both wood quality improvement and efficient wood processing

and utilization. In a previous study [5], we reported that the *Melia azedarach* trees planted in site 2 (Son La provenance) were higher in wood specific gravity and fiber length and smaller in microfibril angle of the S₂ layer of cell wall than those planted in site 1 (Tuyen Quang provenance). This implies that mechanical properties on wood of *Melia azedarach* planted in site 2 may also be higher than those planted in site 1. Therefore, this finding motivated us to examine within-stem variations and compare mechanical properties of *Melia azedarach* planted in two sites in northern Vietnam.

It is well known that wood quality assessment involves the consideration of wood density (WD) and mechanical properties [6]. WD is one of the most important properties of lignocellulosic materials due to its effect on strength, performance, and the general quality of the final products [7]. Besides, modulus of elasticity (MOE) and modulus of rupture (MOR) are also important properties for the use of wood as structural material. MOE is an indication of stiffness of board or structural member, while MOR is an indication of strength [8]. Therefore, the determination of mechanical properties (MOR and MOE) together with WD is important to understand their relationships.



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Currently, rapid and efficient methods for the determination of material properties are desired. Bucur [9] and Keunecke et al. [10] highlighted the benefits of the ultrasonic technique for the determination of the properties of wood, among which are the capability to test small specimens and the possibility of testing the same specimen several times due to the nondestructive nature of these measurements. Thus, if the structural timber can be classified by means of a nondestructive method, it is expected that the optimal utilization would be applied to each wood group.

Therefore, the main objective of this study was to quantify and estimate variations within stem and compare the mechanical properties of *Melia azedarach* planted in two sites in northern Vietnam. This study was also intended to determine the relationship between WD and mechanical properties; between the mechanical properties measured by destructive and nondestructive methods. Besides, the quality of timber produced from *Melia azedarach* in Vietnam was assessed based on their mechanical properties for grading purpose.

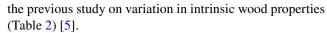
Materials and methods

Sampling

Six trees around 17–19 years old (ring count at 15 cm above the ground) were chosen basing on straight trunks, normal branching, and no signs of any diseases or pest symptoms and felled in August 2016 from two state-owned plantations in Vietnam. The trees were planted at a stand density of 830 trees per hectare at spacing of 4 m × 3 m from seedlings produced by seeds from natural forests located near each site. As described in a previous paper [5], the location and detail information of the two sites are given in Table 1. The trees were felled through cutting their stems at 15 cm above the soil level. 50 cm long logs were taken at different height levels (0.3, 1.3, 3.3, 5.3, and 7.3 m heights from above the ground). The north and south sides of each tree were marked before felling. Sample trees were the same as those used in

Table 1 General characteristics of the study sites [5]

Description	Site 1: Northeast	Site 2: Northwest		
Province	Tuyen Quang	Son La		
Latitude	22°17′01″N	20°56′18″N		
Longitude	105°19′22″E	104°26′25″E		
Altitude (m)	112	434		
Mean rainfall (mm year ⁻¹)	2000	1300		
Mean temperature (°C)	23.4	24		
Soil origin	Calcisols	Ferralsols		



From each log, specimens $(20 \times 20 \times 320 \text{ mm}, \text{radial} \times \text{tangential} \times \text{longitudinal})$ were cut at three distances from pith (10, 50, and 90% of the radial length) on both sides (North and South). MOR and MOE were assessed in accordance with Japanese Industrial Standards (JIS Z2101) [11]. The total number of small clear wood specimens was 180. The specimens were conditioned in a room at a constant temperature (20 °C) and relative humidity (60%) to constant weight. The average moisture content of the test pieces after this state was approximately 12%.

WD and dynamic modulus of elasticity (E_d)

Before measuring the MOR and MOE by destructive method, WD and $E_{\rm d}$ were measured for each specimen. WD was obtained as a percentage of the wood weight per wood volume. $E_{\rm d}$ was measured by a stress wave method (Fakopp machine). The Fakopp device measures the time of acoustic wave propagation through timber element, including the length of coaxial connections of probes. The acoustic signal is created by a hammer strike on the acoustic probe. The velocity of acoustic wave propagation is a ratio of length of specimen per propagation time. $E_{\rm d}$ in the direction parallel to grain was then determined by a simple relation:

$$E_{\rm d} = \rho \cdot v^2$$

where $E_{\rm d}$ is dynamic modulus of elasticity (GPa); ρ is wood density (kg/cm³); and ν is velocity of acousitic wave propagation (m/s).

MOR and MOE

Specimens were subjected to bending test using Instron Tester over a span length of 280 mm. Load was applied to the center of the specimen at a constant speed of 5 mm per minute. MOR and MOE were calculated as

Table 2 Age, diameter at breast height, and total stem height of sampled *Melia azedarach* trees [5]

Site	Tree no.	Age ^a (years)	DBH (cm)	H(m)
Site 1	1	18	32.5	19.6
	2	19	32.2	21.1
	3	17	32.5	21.4
Site 2	4	18	33.8	20.1
	5	18	32.2	19.1
	6	17	29.9	21.4

DBH diameter at breast height (at 1.3 m above the ground), H tree height



^aMeasured by ring counting at the 15 cm above the ground

$$MOR = \frac{3PL}{2bh^2}$$

$$MOE = \frac{P_1 L^3}{4d_1 bh^3}$$

where MOR is modulus of rupture; MOE is modulus of elasticity; P is maximum load (N); P_1 is load at the limit of proportionality (N); L is span length (mm); b is width of the specimen (mm); h is thickness of the specimen (mm); and d_1 is the deflection at the limit of proportionality (mm).

Data analysis

Analysis of variance (ANOVA) for all wood mechanical properties (MOR, MOE, and $E_{\rm d}$) was performed according to the model shown in Table 3 to test the significance of site, tree, height level, and radial position effects. Trees were considered as random effects, and the other sources of variation as fixed effects. Variance components for the sources of variation were also estimated. Average WD, MOR, MOE, and $E_{\rm d}$ were computed for each radial position, stem height, site and for total sites. Besides, the differences among radial and height positions within stem were examined by Tukey–Kramer HSD test. Statistical analysis was performed using R software version 3.2.3.

Grade yield

Grade yield for the specimens was checked using the grading standard of mechanical properties of timbers from Southeast Asia and Pacific regions by Forestry and Forest Products Research Institute (FFPRI) in Japan (Table 4) [12].

Table 3 Model used in the analysis of variance

No.	Source of variation
1	Site (S)
2	Tree/site (T/S)
3	Height level (L)
4	$L \times S$
5	$L \times T/S$
6	Radial position (P)
7	$P \times S$
8	$P \times T/S$
9	$P \times L$
10	$P \times L \times S$
11	$P \times L \times T/S$
12	Residuals

Table 4 Grading standard of mechanical properties of timber from Southeast Asia and Pacific regions by Forestry and Forest Products Research Institute (FFPRI) (1975)

Grade	MOR (MPa)	MOE (GPa)
I	≤ 58.8	≤ 7.4
II	58.9-82.4	7.5–10.3
III	82.5-106.9	10.4-13.2
IV	107.0-130.4	13.3-16.2
V	≥ 130.5	≥ 16.3

Units for MOR (kg/cm²) and MOE (10³ kg/cm²) in original material were, respectively, converted to MPa and GPa for convenience in allocation the values in this study

MOR modulus of rupture, MOE modulus of elasticity

Results and discussion

WD and mechanical properties

Table 5 shows the mean WD and mechanical properties each radial position, stem height, site and for total sites of *Melia azedarach* planted in the northern Vietnam. The overall WD, MOR, MOE, and $E_{\rm d}$ were 0.51 g/cm³, 78.58 MPa, 9.26 GPa, and 10.93 GPa, respectively. The results are consistent with those published for *Melia azedarach* planted in southeastern Vietnam [13].

Table 6 shows the results obtained for the analysis of variance made for each mechanical property, regarding the statistical significance and proportion of explained variation for the different sources of variation. The residual effect was responsible for 7.53–15.4% of the total variation, and it is due to other factors that were not taken into account in this experimental design.

Within tree, the variation of mechanical properties with height was very small and without statistical significant (Table 6). In radial direction, MOR, MOE, and E_d increased gradually from 10 to 90% of the radial length from pith and this trend was consistent in both sites (Fig. 1). Radial position is the most important and highly significant source of variation in mechanical properties, explaining 55.31, 30.93, and 37.53% of the total variation of the MOR, MOE, and $E_{\rm d}$, respectively (Table 6). The present results are in line with those of Machado et al. [14] who reported that height level was not a significant source, while radial position was highly significant source of variation for the mechanical properties on Acacia melanoxylon R. Br. Variation along the radial direction is the best known and most studied withintree variability in wood, which is generally reflected as radial pattern of change in wood characteristics. The radial change in wood properties varies in magnitude and type in different species [15–17]. The radial variation in mechanical properties of other hardwood species such as Swietenia macrophylla Roxb [6], Tectona grandis [18], and Nauclea



Table 5 Variations of wood density (WD), modulus of rupture (MOR), modulus of elasticity (MOE), and dynamic modulus of elasticity (E_d) within stem and between sites of *Melia azedarach*

Variable	Description	n	WD (g/cm ³)	MOR (MPa)	MOE (GPa)	E _d (GPa)
Radial direction (%)	10	60	0.44 ± 0.01^{c}	$54.64 \pm 2.55^{\circ}$	7.81 ± 0.22^{c}	9.15 ± 0.24^{c}
	50	60	$0.52 \pm 0.01^{\rm b}$	82.18 ± 2.23^{b}	9.55 ± 0.20^{b}	11.20 ± 0.22^{b}
	90	60	0.57 ± 0.01^{a}	98.94 ± 1.48^{a}	10.41 ± 0.21^{a}	12.45 ± 0.23^{a}
Stem height (m)	7.3	36	0.51 ± 0.01^{a}	77.58 ± 4.20^{a}	9.71 ± 0.32^{a}	11.25 ± 0.38^{a}
above the ground	5.3	36	0.49 ± 0.01^{a}	74.70 ± 4.01^{a}	8.87 ± 0.35^{a}	10.76 ± 0.40^{a}
	3.3	36	0.51 ± 0.01^{a}	78.78 ± 4.31^{a}	9.52 ± 0.31^{a}	11.25 ± 0.36^{a}
	1.3	36	0.50 ± 0.01^{a}	81.28 ± 4.05^{a}	9.36 ± 0.29^{a}	10.86 ± 0.34^{a}
	0.3	36	0.52 ± 0.01^{a}	80.57 ± 4.07^{a}	8.84 ± 0.34^{a}	10.55 ± 0.38^{a}
Site	1	90	0.50 ± 0.01^{b}	$66.20 \pm 2.55^{\text{b}}$	8.25 ± 0.19^{b}	9.73 ± 0.21^{b}
	2	90	0.52 ± 0.01^{a}	90.97 ± 1.90^{a}	10.27 ± 0.16^{a}	12.13 ± 0.18^{a}
Mean		180	0.51 ± 0.01	78.58 ± 1.84	9.26 ± 0.15	10.93 ± 0.17

Mean values are followed by standard errors

n number of wood specimen

Table 6 Variance components for mechanical properties of *Melia azedarach*

Source of variation	df	MOR		MOE		$E_{\rm d}$	
		p value	Var (%)	p value	Var (%)	p value	Var (%)
Site (S)	1	0.001	25.43	0.001	26.78	0.001	29.15
Tree/site (T/S)	4	0.862	0.55	0.013	5.12	0.001	6.78
Height level (L)	4	0.717	0.91	0.103	3.18	0.432	1.55
$L \times S$	4	0.974	0.21	0.618	1.08	0.001	0.57
$L \times T/S$	16	0.996	2.24	0.298	7.08	0.843	3.90
Radial position (P)	2	0.001	55.31	0.001	30.93	0.028	37.53
$P \times S$	2	0.001	2.09	0.209	0.76	0.199	0.61
$P \times T/S$	8	0.031	1.62	0.400	1.80	0.001	4.39
$P \times L$	8	0.150	1.19	0.869	1.50	0.923	0.61
$P \times L \times S$	8	0.933	0.29	0.864	0.90	0.766	0.95
$P \times L \times T/S$	32	0.511	2.62	0.483	5.47	0.927	2.57
Residuals	90		7.53		15.4		11.40

MOR modulus of rupture, MOE modulus of elasticity, $E_{\rm d}$ dynamic modulus of elasticity, df degrees of freedom, Var variance (%)

diderrichii [19] supports the findings of the present study. In addition, the present study also confirms that the magnitude of mechanical properties varied from pith to bark (Fig. 1).

Besides, site was also a significant source of variation in MOR, MOE, and $E_{\rm d}$. Tree-to-tree within site was a significant source of variation only in MOE and $E_{\rm d}$. Site explained from 25.43 to 29.15% of the total variation in mechanical properties (Table 6). The average MOR, MOE, and $E_{\rm d}$ on wood of *Melia azedarach* planted in site 2 (Son La provenance) are totally higher than those planted in site 1 (Tuyen Quang provenance) (Table 5). The different results of MOR and MOE of *Melia azedarach* planted in different locations were also reported such as in Mexico (112.0 MPa–10.3 GPa), Argentina (111.0 MPa–9.7 GPa), Indonesia (101.0 MPa–9.9 GPa), and Paraguay (93.0 MPa–9.8 GPa) [1]. The variations in

the wood properties of the same species are due to different genotypes and ecological conditions of sites such as altitude, precipitation, temperature, soil, water, and nutrients. These two factors affect both the growth and development of trees [20]. Genetic structure is the main source of change of wood's properties, while ecological conditions of site directly or indirectly affect on the development and fertility, body form, and height of tree. Thus, in the present study, the significant difference in all mechanical properties between site 1 and site 2 may be contributed by the differences in altitude, mean annual rainfall, and soil types between two sites (Table 1). However, further experiments will be needed to determine genetic effect on variation in mechanical properties for *Melia azedarach* planted in northern Vietnam.



^{a,b,c}Means with different superscript within a column significantly differ (p < 0.05)

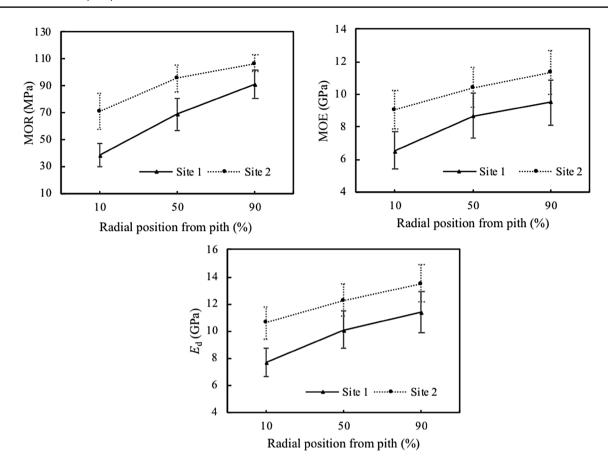


Fig. 1 Radial variation of modulus of rupture (MOR), modulus of elasticity (MOE), and dynamic modulus of elasticity (E_d) by sites for *Melia azedarach* (bars: mean standard deviation)

Correlation of WD with mechanical properties

Relationships between WD and mechanical properties are presented in Fig. 2 and Table 7. The results showed that WD had significant positive linear relationships at the 0.001 confidence level with MOR (r=0.93 for site 1, r=0.84 for

site 2) and MOE (r=0.79 for site 1, r=0.72 for site 2). This implies that WD can be used as a parameter for predicting mechanical properties. Therefore, improving WD which can be obtained by tree breeding programs, growth conditions, or section for high wood density parts, etc. would have a positive impact on mechanical properties of *Melia*

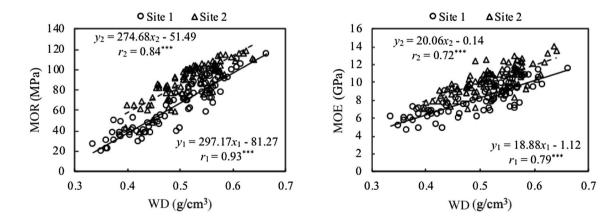


Fig. 2 Relationship between wood density (WD) and modulus of rupture (MOR); and WD and modulus of elasticity (MOE) for *Melia azedarach* in two sites (solid and dash lines are linear relationships for site 1 and site 2, respectively. Triple asterisks: p < 0.001)



Table 7 Relationship between wood density (WD) and mechanical properties; dynamic (E_d) and static (MOR, MOE) properties for each site and combined sites of *Melia azedarach*

Site	Linear regression model $y = ax + b$							
	Mechanical properties	WD			$E_{ m d}$			
		a	b	r	\overline{a}	b	r	
Site 1	MOR	297.17	-81.27	0.93***	9.80	-29.19	0.82***	
	MOE	18.88	-1.12	0.79***	0.83	0.22	0.93***	
	$E_{ m d}$	23.08	-1.72	0.86***	_	_	_	
Site 2	MOR	274.68	-51.49	0.84***	7.46	0.49	0.72***	
	MOE	20.06	-0.14	0.72***	0.76	1.06	0.85***	
	$E_{ m d}$	25.72	-1.20	0.82***	_	_	-	
N	MOR	312.25	-79.86	0.85***	9.24	-22.45	0.84***	
	MOE	21.29	-1.55	0.73***	0.81	0.41	0.92***	
	$E_{\rm d}$	26.35	-2.44	0.79***	-	_	-	

MOR modulus of rupture, MOE modulus of elasticity, $E_{\rm d}$ dynamic modulus of elasticity, r correlation coefficient

^{***}p < 0.001

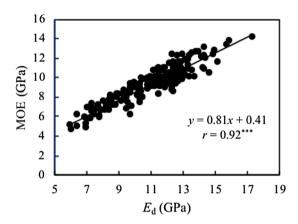


Fig. 3 Correlation between moduli of elasticity measured by acoustic method and destructive test for combined sites of *Melia azedarach* (triple asterisk: p < 0.001)

azedarach. Positive linear relationships between WD and mechanical properties were also found on other hardwood species such as *Tectona grandis* [18], *Eucalyptus tereticornis* [21], and *Acacia melanoxylon* [14, 22]. The *r* values of WD and mechanical properties for site 1 are slightly higher than those for site 2. This could be caused by the differences in growth conditions such as altitude, mean annual rainfall, and soil types between two sites.

Correlation between moduli of elasticity

The correlation coefficient between the moduli of elasticity of the acoustic method and the destructive test for the combined sites was very high (r=0.92, p<0.001) (Fig. 3). Similar results were also reported by different researchers for both structural timber and small specimen [23–27]. Widmann [23] and Johansson et al. [24] found high coefficient

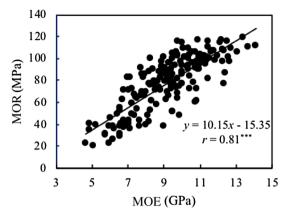
of correlation between $E_{\rm d}$ and MOE (r=0.94 and 0.92) for beech and spruce, respectively. Sales et al. [25] and Vazquez et al. [26] also reported a high correlation between $E_{\rm d}$ measured by ultrasonic technique and static modulus of destructive test on structural lumber of *Goupia glabra* and *Castanea sativa* Mill., respectively. The strong correlation coefficient between MOE and $E_{\rm d}$ (r=0.91) for the combined species of small clear specimens [27] also supports the findings of the present study. Therefore, the present result indicated that the stress wave method used in this study provided relatively accurate information for MOE of *Melia azedarach* planted in northern Vietnam, if the density of measured element is known.

However, the value of MOE was measured by destructive test which best defines class of timber, is usually lower than the value of $E_{\rm d}$ measured by acoustic method. In present study, the average value of MOE is lower than that of $E_{\rm d}$ about 15.28% or 1.67 GPa (Table 5). This is confirmed by other reports in literature. Wang et al. [27] showed the difference between dynamic and static modulus of elasticity observed for small clear specimens of Western hemlock 20.74% (1.93 GPa) and Sitka spruce 28.78% (2.76 GPa). Posta et al. [28] confirmed that the value of $E_{\rm d}$ measured by Fakopp is higher than the value of MOE about 27% for Norway spruce.

Prediction of bending strength

MOE is closely related to MOR. It could be said that it is the best indicator of MOR, and thus classes of timber [28]. The results of relationship between moduli of elasticity and strength are presented in Fig. 4 and Table 7. The results revealed that there were high correlation coefficients not only for each site, but also for combined sites between bending strength and moduli of elasticity detected by both





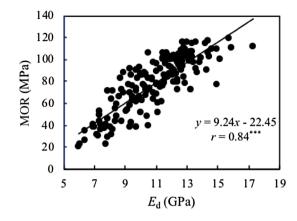


Fig. 4 Relationship between modulus of rupture (MOR) and modulus of elasticity (MOE); and MOR and dynamic modulus of elasticity (E_d) for combined sites of *Melia azedarach* (triple asterisk: p < 0.001)

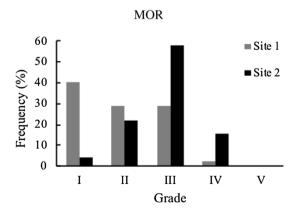
destructive test and acoustic method. The r values of MOR with MOE and $E_{\rm d}$ at the 0.001 confidence level for the combined sites are 0.81 and 0.84, respectively (Fig. 4). This showed that $E_{\rm d}$ is a good indicator to predicting the strength of *Melia azedarach* wood. The results confirm the findings from the literature. Posta et al. [28] reported the relationship between MOR and MOE (r=0.92) and between MOR and $E_{\rm d}$ (r=0.81) on Norway spruce laths. Wang et al. [27] presented good correlation coefficients between MOR and $E_{\rm d}$ (r=0.68 for Western hemlock, r=0.69 for Sitka spruce) of small clear specimens. The positive relationship between MOR and $E_{\rm d}$ on Brazilian hardwood [29] also supports the findings of the present study.

Grade yield of mechanical properties

Figure 5 shows the grade yield for both MOR and MOE using grading standard of mechanical properties of timbers from Southeast Asia and Pacific regions (FFPRI, 1975). The grade yield for MOR in site 1 was the highest for grade I,

followed by grade II and III. The grade IV was the lowest. The highest frequency of specimens for MOR in site 2 belonged to grade III, followed by grades II and IV. Grade I was the lowest. There was no grade yield of MOR in grade V for both sites. MOE for *Melia azedarach* grown in site 1 can be allocated mainly in to grades I and II while that in site 2 can be allocated mainly in to grades II and III. Based on results in Table 5 and Fig. 5, wood for *Melia azedarach* grown in site 1 (Tuyen Quang provenance) can be allocated into grade II while that in site 2 (Son La provenance) can be allocated in grade III using grading standard of mechanical properties of timber from Southeast Asia and Pacific regions.

It is well known that wood property is closely related to its structure. Relationship between structure and property has been of interest to wood scientists [30]. The previous researchers [15, 30, 31] reported that wood with longer fiber length and lower microfibril angle has higher specific gravity and mechanical properties for some hardwood species. In the present study, together with higher



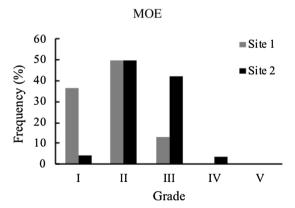


Fig. 5 Specimen grade allocation in terms of modulus of rupture (MOR) and modulus of elasticity (MOE) for both sites according to grading standard of mechanical properties of timbers from Southeast Asia and Pacific regions by FFPRI (1975)



specific gravity, longer fiber length, and lower microfibril angle [5], *Melia azedarach* trees planted in site 2 also had higher mechanical properties than trees planted in site1. This implies that site 2 or any other location with similar environmental conditions (soil, rainfall, temperature, and altitude) to site 2 should be preferred for establishment of *Melia azedarach* plantations in northern Vietnam. In the future, we will investigate and clarify the effect of fiber length and microfibril angle on the within-tree variation in mechanical properties of *Melia azedarach* plantations in northern Vietnam.

The common steps in establishing grades for lumber are: testing of small clear specimens according to guidelines, establishing strength values and allowable properties, establishing visual grading rules, and lastly verifying grades using in-grade testing [32]. This research has established the first step in assigning allowable mechanical properties for *Melia azedarach* grown in northern Vietnam. This will help in assignment of standard grades that will ensure the efficient utilization of *Melia azedarach* timber in Vietnam.

Conclusions

This research has clarified within-stem variations in mechanical properties (MOR, MOE, and E_d) of Melia azedarach grown in northern Vietnam. Within tree, the mechanical variation with height was very small and without statistical significant. In radial direction, MOR, MOE, and E_d increased from pith to bark and radial position is the most important and highly significant source of variation in mechanical properties. The average MOR, MOE, and E_d on wood of Melia azedarach planted in site 2 are higher than those planted in site 1. WD had a strong positive linear relationship with mechanical properties. This suggests that controlling WD would have a positive impact on mechanical properties. The average value of MOE is lower than that of $E_{\rm d}$ about 15.28% or 1.67 GPa. High coefficient of correlation was found between $E_{\rm d}$ measured by stress wave method and MOE measured by destructive test. This suggests that the stress wave method used in this study provides relatively accurate information for MOE. Besides, MOR of Melia azedarach can be predicted by nondestructive technique, if the density of measured element is known. Furthermore, the present results are a foundation that will provide a technical basis for the machine grading of Melia azedarach timber in northern Vietnam.

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Compliance with ethical standards

Conflict of interest The authors declare no conflict of interest.

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