Effects of localized environment on eucalyptus clone chemical composition

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ABSTRACT: Eucalyptus wood is becoming the most important feedstock for bleached kraft pulp production, particularly in South America. However, pulp manufacturers have observed that some eucalyptus clones are resistant to wind action and others are not. Those lacking wind resistance are prone to breaking, which results in productivity losses. This study aims to investigate a correlation between the chemical composition of eucalyptus clones and their resistance to wind action. We found differences among the wind-resistant and non-wind-resistant clones, but that the fragility of trees studied for wind resistance does not correlate to the chemical composition of the wood. Four notable differences were observed. First, clones grown at lower altitudes differed from clones grown at higher altitudes in their acetyl, insoluble lignin, and extractives contents, and in their syringyl/guaiacyl (S/G) ratios. However, we found no relationship among these components and wind susceptibility. Second, soluble lignin content increased with the increase of wind resistance in the high altitude clones. Third, the low altitude clones showed slightly lower xylan content (12.4%) and higher lignin content (29.6%) than the others. Finally, the insoluble lignin content, total acetyl groups, extractives, ash contents, and S/G ratios of the clones were influenced by the region where they were grown.

Application: The results of this paper provide important information for helping pulp mills increase wood productivity and decrease pulp production costs.

Lignocellulosic biomass sources such as hardwoods and softwoods are the main raw materials for pulp production. They deliver the suitable fiber characteristics to final paper products that are cost competitive. In this context, eucalyptus is becoming the most important raw material for pulp mills worldwide. The major interest in eucalyptus wood comes from its relatively low production cost in certain regions, such as South America, mainly because of its high forest productivity and high pulping yield. Much research has been done to improve eucalyptus productivity. According to the Brazilian Tree Industry association (IBA) [1], the average productivity of Brazilian eucalyptus forests increased from 24 m^3/ha/year in 1980 to 39 m^3/ha/year in 2015, which represents a 61% increase.

Low wood cost can be affected by production losses in the field. One of the most important sources of wood loss in Brazil is breakage of eucalyptus trees in the field by wind action. Wind causes disturbances in natural and planted forests, with damage recorded since 1940 [2] and reports worldwide [3-6]. Pulp manufacturers have noted that some eucalyptus clones have higher wind resistance than others. Trees with low wind resistance are more easily broken in the field, and even when not broken, the resulting wood might have a chemical composition modified by the formation of reaction wood.

Studies have been conducted to investigate tree resistance to wind action in South America. Melo [7] studied the properties of different woods, with emphasis on wood with associated wind damage for clones grown at different altitudes. The mechanical strength of eucalyptus clones was measured in works by Ferreira et al. [8] and Martino [9]. They did not, however, explain why some clones are wind resistant and others are not. No published studies have examined the chemical composition of the woods to explain this behavior.

Wood quality is an extremely important factor in pulp production when the goals are to obtain high yields, low cost, and high quality. The chemical composition of wood is an important factor for pulp manufacturing. The eucalyptus genus includes different species growing in different countries, according to their adaptability to the weather and soil conditions. In this way, the wood composition can vary as a result of many interactions between the planted trees and area edaphoclimatic conditions [10]. Thus, research about the quality of the wood harvested from forest plantations should be considered extremely important to manufacturers because this factor is closely related to process performance and the characteristics of the final pulp and paper products. In this context, this study aims to investigate if there is a correlation between the wood chemical composition of eucalyptus clones and their resistance to wind action.

MATERIALS AND METHODS

Six eucalyptus clones from *Eucalyptus urophylla* × *Eucalyptus grandis* hybrids that were 24 months of age and had different degrees of resistance to wind action (susceptible, intermediate, and tolerant) were examined. Two regions with different altitudes (high altitude = approximately 780 m, and low altitude = approximately 410 m) were also investigated (**Table I**). The chemical composition of the woods from these clones were examined, which included carbohydrates, acetyl groups, uronic acids, acid soluble and insoluble lignin, syringyl/guaiacyl (S/G) ratio, total extractives content, and ash.

Four trees of each clone from both regions were collected. The eucalyptus clones sampled were chipped in a laboratory chipper (Chugoku-Kikai; Japan) equipped with three knives and two screens (40 mm and 13 mm). The chips were well mixed in a 260 m^3 rotary mixer and the samples were screened according to the SCAN-CN 40:94 procedure. The chips retained in the 3 mm and 7 mm screens were collected and mixed again, air dried to about 15% moisture, and stored in large plastic bags. For the chemical analyses, about 1 kg of each clone was sampled and ground in a Wiley type mill to produce sawdust of variable size. This sawdust was screened in accordance with TAPPI Standard Test Method T 257-cm85 "Sampling and preparing wood for analysis." The sawdust that passed through the 40 mesh screen and was retained on the 60 mesh screen was selected for the chemical analyses. The sawdust was air dried and conditioned in a temperature and humidity controlled room (23±1°C, 50±2% RH) until an equilibrium moisture was achieved (~10%). This sawdust was used for the chemical analyses. All chemical analyses were performed in duplicate, according to laboratory standards listed in **Table II** [11-15].

Clone	Situation	Region
A	Susceptible	Low altitude
B	Susceptible	Low and high altitude
C	Intermediate	Low and high altitude
D	Intermediate	Low altitude
E	Tolerant	Low and high altitude
F	Tolerant	Low altitude

I. Eucalyptus clones and their wind resistance and planting region.

Statistical analysis

The evaluation of the chemical composition in each region (completely randomized design) was statistically examined by analysis of variance (ANOVA). Significant differences among the treatments were determined by paired comparisons by the Tukey test using a significance level of 5%. Statistical analyses were performed using STATISTICA 7.0 software (StatSoft/Dell; Tulsa, OK, USA).

To evaluate the interaction effect between the growing regions, the results obtained for each parameter were statistically analyzed, considering the completely randomized design with six treatments and four replications. The treatments were considered in a 3×2 factorial design, three clones (B, C, and E) and two regions (high altitude and low altitude). When there was a significant interaction observed, a study of the region effect in the clones was performed. The level of significance used was 5%. The concentration of each wood chemical component by region was also evaluated.

RESULTS AND DISCUSSION

Eucalyptus wood chemistry composition

<u>Extractives and ash content</u>

Table III shows the results for total extractives and ash content. The extractives content of the six clones of *E. urophylla* × *E. grandis* ranged from 2.7% to 5.0% in low altitude regions and from 2.4% to 2.8% in high altitude regions. The results showed that for the low altitude region, the lowest extractives content was found in intermediate (D) and tolerant (E) clones (2.7% to 2.9%). On the other hand, for the high altitude region,

Analysis	Methodology
Soluble lignin content	Goldschimid (1971) [11]
Insoluble lignin content	TAPPI Standard Test Method T 222 om-11 "Acid-insoluble lignin in wood and pulp"
Carbohydrates content	Wallis et al. (1996) [12]
Total uronics content	Scott (1979) [13]
S/G ratio	Lin and Dence (1992) [14]
Total extractives content	TAPPI Standard Test Method T 264 cm-97 "Preparation of wood for chemical analysis"
Acetyl group content	Solar et al. (1987) [15]
Ash content	TAPPI Standard Test Method T 211 om-12 "Ash in wood, pulp, paper, and paperboard: combustion at 525°C"

II. Laboratory standards employed to analyze chemical composition of eucalyptus wood (S/G ratio = syringyl/guaiacyl ratio).

Region	Clone		Extractives	Ash
Low altitude region	Susceptible	A	4.7 [a]	0.41 [a]
	Susceptible	B	4.1 [a]	0.31 [a]
	Intermediate	C	5.0 [a]	0.39 [a]
	Intermediate	D	2.9 [b]	0.31 [a]
	Tolerant	E	2.7 [b]	0.43 [a]
	Tolerant	F	4.3 [a]	0.35 [a]
p value			0.000	0.107
High altitude region	Susceptible	B	2.4 [a]	0.50 [a]
	Intermediate	C	2.8 [a]	0.30 [b]
	Tolerant	E	2.4 [a]	0.56 [a]
p value			0.214	0.000

[a, b] Mean values in the same column followed by the same letter are not significantly different by Tukey test at 5% probability level.

III. Percentage of extractives and ash in clones from low and high altitude regions.

no significant differences were seen in the total extractive contents among the clones. For wood extractives content in the eucalyptus clones, the values were in the same range as those found in the literature for commercial eucalyptus clones used for pulp production [16-19].

Ash in wood chips is detrimental to the pulping process. It causes corrosion and deposits on equipment, reduces biomass heating value, and decreases mill throughput [19]. The ash content of the six *E. urophylla* × *E. grandis* clones ranged from 0.31% to 0.41% in those from low altitude regions and from 0.30% to 0.56% in those from high altitude regions. No significant differences were seen in the inorganic ash content among the evaluated samples.

Table IV shows the extractives and ash content from the analysis of clones common to both regions examined in this study. The results showed that, in the low region clones, the extractives content (3.9%) is higher than in high altitude clones (2.5%), and the ash content (0.37%) is smaller than in the low altitude ones (0.45%).

Region	Extractives	Ash
Low altitude region	3.9 [a]	0.37 [a]
High altitude region	2.5 [b]	0.45 [b]
p value	0.000	0.007

[a, b] Mean values in the same column followed by the same letter are not significantly different by Tukey test at 5% probability level.

IV. Percentage of extractives and ash content in clones by region.

Carbohydrates

The carbohydrate content was investigated by examining the levels of glucans, xylans, galactans, mannans, arabinans, and uronic acids. **Table V** shows the results. The contents ranged from 44.5% to 46.7% for glucans, 12.4% to 13.0% for xylans, 1.7% to 1.8% for galactans, 1.3% to 1.4% for mannans, 3.9% to 4.1% for uronic acids, and 2.7% to 3.5% for acetyl groups. The contents were similar to those reported in the literature for eucalyptus wood glucans (39.6% to 53.0%) and xylans (11.6% to 18.6%) [21,22]. In general, the results showed no statistical difference among these parameters in the clones. Although differences were seen in the acetyl content among the clones of the low altitude region, these differences did not follow a trend. No significant differences were observed for clones A (susceptible) and C (intermediate), which had the lowest acetyl group content (2.7% to 3.0%), and clones D (intermediate) and E (tolerant), which had the highest acetyl group content (3.5% to 3.6%).

Table VI shows the averaged results of carbohydrate, uronic acids, and acetyl group content of each clone by region. The results indicated that clones from the lower altitude region, where most wind damage action occurs, had lower xylans (12.4%) and galactans (1.7%) content when compared with the clones from the high altitude region. However, these results did not show an interaction between the carbohydrate levels, the side uronic acids and acetyl group content, and the wind resistance of the clones.

Lignin

Lignin is a major wood constituent responsible for support and rigidity of the tree. **Table VII** shows the average percentages of soluble and acid insoluble lignin, total lignin (sum of

Region Clone			Glucans	Xylans	Galactans	Mannans	Arabinans	Uronic acids	Acetyl group
Low altitude region	Susceptible	A	45.1 ^{a}	12.5 ^{a}	1.7 ^{a}	1.3 ^{a}	0.3 ^{a}	4.0 ^{a}	3.0 ^{a,d}
	Susceptible	B	46.7 ^{a}	12.5 ^{a}	1.7 ^{a}	1.4 ^{a}	0.3 ^{a}	4.0 ^{a}	3.1 ^{a,b}
	Intermediate	C	46.7 ^{a}	12.3 ^{a}	1.8 ^{a}	1.3 ^{a}	0.3 ^{a}	4.1 ^{a}	2.7 ^{d}
	Intermediate	D	44.5 ^{a}	12.7 ^{a}	1.8 ^{a}	1.3 ^{a}	0.3 ^{a}	4.0 ^{a}	3.5 ^{b,c}
	Tolerant	E	45.1 a	12.4 ^{a}	1.7 ^{a}	1.4 ^{a}	0.3 ^{a}	3.8 ^{a}	3.6 ^{c}
	Tolerant	F	44.9 ^{a}	12.5 ^{a}	1.8 ^{a}	1.3 ^{a}	0.3 ^{a}	3.9 ^{a}	3.3 ^{a,b,c}
p value		0.123	0.904	0.734	0.831	1.000	0.557	0.000	
High altitude region	Susceptible	B	46.0 ^{a}	12.7 ^{a}	1.8 ^{a}	1.3 a	0.3 ^{a}	4.0 ^{a}	2.9 ^{a}
	Intermediate	C	45.6 ^{a}	13.0 ^{a}	1.8 ^{a}	1.3 a	0.3 ^{a}	4.1 ^{a}	3.2 ^{a}
	Tolerant	E	46.6 ^{a}	12.4 ^{a}	1.9 ^{a}	1.4 ^{a}	0.3 ^{a}	4.0 ^{a}	3.0 ^{a}
p value		0.174	0.147	0.932	0.454	0.405	0.902	0.194	

$^{a, b, c, d}$ Mean values in the same column followed by the same letter are not significantly different by Tukey test at 5% probability level.

V. Percentages of glucans, hemicelluloses, acetyl, and uronic acid groups in clones from low and high altitude regions.

Region	Glucans	Xylans	Galactans	Mannans	Arabinans	Uronic acids	Acetyl group
Low altitude region	46.2 ^{a}	12.4 ^{a}	1.7 ^{a}	1.3 ^{a}	0.3 ^{a}	4.0 ^{a}	3.1 ^{a}
High altitude region	46.1 ^{a}	12.7 ^{b}	1.8 ^{b}	1.3 ^{a}	0.3 ^{a}	4.0 ^{a}	3.0 ^{a}
p value	0.833	0.048	0.032	1.000	0.331	0.711	0.239

$^{a, b}$ Mean values in the same column followed by the same letter are not significantly different by Tukey test at 5% probability level.

VI. Percentage of glucans, hemicelluloses, acetyl, and uronic acid groups in clones by region.

Region	Clone		Soluble Lignin	Acid Insoluble Lignin	Total Lignin	S/G Ratio
Low altitude region	Susceptible	A	4.9 ^{a}	26.1 ^{ab}	31.0 ^{a}	3.8 ^{ab}
	Susceptible	B	4.0 ^{a}	24.9 ^{b}	28.9 ^{a}	3.4 ^{ab}
	Intermediate	C	3.4 ^{a}	25.9 ^{ab}	29.2 ^{a}	3.2 ^{a}
	Intermediate	D	4.5 ^{a}	26.6 ^{a}	31.0 ^{a}	3.5 ^{ab}
	Tolerant	E	5.5 ^{a}	25.4 ^{ab}	30.9 ^{a}	3.8 ^{b}
	Tolerant	F	3.9 ^{a}	26.7 ^{a}	30.6 ^{a}	3.3 ^{ab}
p value			0.054705	0.011615	0.028970	0.014999
High altitude region	Susceptible	B	3.9 ^{a}	25.5 ^{a}	29.4 ^{a}	3.7 ^{b}
	Intermediate	C	2.9 ^{b}	27.1 ^{b}	29.9 ^{a}	2.6 ^{a}
	Tolerant	E	4.6 ^{c}	24.1 ^{a}	28.7 ^{a}	3.9 ^{b}
p value			0.000096	0.001383	0.150477	0.000001

$^{a, b, c}$ Mean values in the same column followed by the same letter are not significantly different by Tukey test at 5% probability level.

VII. Percentage of soluble lignin, acid insoluble lignin, total lignin, and S/G ratio in clones from low and high altitude regions.

Region	Soluble Lignin	Acid insoluble Lignin	Total Lignin	S/G ratio
Low altitude region	4.3 a	25.4 a	29.6 a	3.5 a
High altitude region	3.8 a	25.6 a	29.3 b	3.4 a
p value	0.112927	0.577982	0.482000	0.393197
a, b Mean values in the same column followed by the same letter are not significantly different by Tukey test at 5% probability level.				

VIII. Percentage of soluble lignin, insoluble lignin, total lignin content, and S/G ratio in clones by region.

Source of Variation	Mean Square						
	Glucans	Acid Insoluble Lignin	Total Lignin	Acetyl Group	S/G Ratio	Extractives	Ash
Place/Clone B	0.9800 ns	0.7813 ns	0.6050 ns	0.0800 ns	0.1250 ns	5.6112 *	0.0741 *
Place/Clone C	2.4200 ns	3.0013 *	1.2013 ns	0.5513 *	0.6613 *	9.6800 *	0.0153 ns
Place/Clone E	4.6512 ns	3.5113 *	9.6800 *	0.7200 *	0.0113 ns	0.1013 ns	0.0351 *
* = significant at 5% probability level. ns = not significant.							

IX. Results of the interaction unfolding and evaluation of the location effect within the clone for the wood chemical analysis.

soluble and acid insoluble lignin), and S/G ratio of the lignin present in the eucalyptus trees we evaluated. The total lignin content found in all clones ranged between 28.7% and 31.0%. These results are commonly found in eucalyptus species, varying between 26% and 35% [14].

In the low altitude region, the soluble and total lignin content showed no statistical difference in the clones. Although the percentage of insoluble lignin differed with a susceptible clone (B) to an intermediate clone (D) and a tolerant clone (F), these values (26.6% and 26.7%) are ideal for pulping. For the other clones from the low altitude region and all clones from the high altitude region, the acid insoluble lignin content is the same among a susceptible clone (B) and tolerant clone (E).

In the high altitude region, the soluble lignin content differed among all clones, where the highest value was observed with the tolerant clone E (4.6%), which did not occur in the low altitude region. The results of S/G ratio were different for clones C (intermediate) and E (tolerant) in both regions. The tolerant clone had the highest S/G ratio (3.8 in the low, and 3.9 in the high region). The S/G ratio results were higher than commonly found in hybrids of *E. grandis* × *E. urophylla*. This could explain the fragility of the tree if these results were found only in susceptible clones. However, this was not observed. In a study of this hybrid eucalyptus, Morais [24] concluded that young clones tend to have high S/G values. The high values found in this work can be explained by the fact that the samples analyzed were not older than 24 months.

Table VIII shows the results of the S/G ratio by region. The low altitude region showed a total lignin content slightly greater than the high altitude region. The content of soluble and acid insoluble lignin and the S/G ratios were not statistically different between the regions.

Regarding the lignin analysis, it is possible to conclude that the lignin content (soluble, insoluble, or total lignin), and S/G ratio do not correlate to clone wind resistance when considering the same region.

Effect of interaction between regions

The effect of interactions between clone and region was statistically evaluated for all chemical constituents using a 3×2 factorial analysis at 5% probability level by analysis of variance (ANOVA). **Table IX** shows the result of the interaction unfolding for the interactions that were significant in the ANOVA analysis.

Although the interaction region versus clone in the analysis of variance for glucans content was significant, this parameter was not significant for any of the clones (Table IX), indicating that the significance found in the analysis of variance for interaction of clone versus region is related to other glucans contrasts, and not with the effect of the region on the clone. According to the results of the interactions shown in Table IX, clone B (susceptible) was affected by region only by the extractives and ash content. For clone C (intermediate), the results showed that this clone was affected by the region regarding its acid insoluble lignin, S/G ratio, acetyl group, and extractives contents. The acid insoluble lignin, total lignin, acetyl group, and ash contents of clone E (tolerant) were affected by region.

CONCLUSIONS

We observed that planted area altitude affected the chemical wood composition. On the other hand, the fragility of tree clones to wind action does not seem to correlate with the wood chemical composition, although differences among clones were found, such as the following:

1. The low altitude clones differed in their acetyl, insoluble lignin, S/G ratio and extractives contents; however, there was not an observed relationship among these components and wind susceptibility.
2. The soluble lignin content increased with the increase of wind resistance in the high altitude clones.
3. The low altitude clones showed slightly lower xylan content (12.4%) and higher lignin content (29.6%) than the others.
4. The insoluble lignin content, total acetyl groups, S/G ratio, extractives, and ash contents of the clones are influenced by the region where they are grown. **TJ**

ACKNOWLEDGMENTS

Financial support from EU-project LIGNODECO (KBBE-2009-3-244362), the Minas Gerais State Research Foundation (Fapemig), and the Brazilian National Council for Scientific Research and Technological Development (CNPq) is greatly appreciated.

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ABOUT THE AUTHORS

We work in areas of pulp and paper technology that focus on new opportunities for pulp mills. Avoiding wood losses in the field is important for improving pulp mill feasibility.

Our previous research projects have aimed at improving mill throughput. The results of this study might help save wood and avoid wood cost increases for pulp mills.

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The most difficult job in this study was the identification of the tress with different degrees of resistance to wind action (susceptible, intermediate, and tolerant). The support of the pulp mill, which provided the materials, was very important.

The main finding of this work was that the fragility of tree clones to wind action does not seem to correlate with the wood chemical composition. The results of this paper are an important tool for pulp mill research and development areas, because wood productivity is very important for decreasing pulp production costs.

Our next step will be to investigate other parameters that might be responsible for the resistance of the trees to wind action.

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