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Effect of Altitude on Polysaccharide and Lignin Contents of Brutian pine (*Pinus brutia* Ten.) Wood and Kraft Pulp

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Abstract: This study investigated the chemical properties of Brutian pine (*Pinus brutia* Ten.) wood samples collected at altitudes of 800 m, 900 m, and 1000 m and the behaviors of the chemical components of the same materials during kraft pulping. Chemical component analysis indicated that the wood and pulp sample from 1000 m contained higher holocellulose and α-cellulose contents, and a lower lignin content. For both wood and pulp samples, a continuous increase in holocellulose and α-cellulose contents and a continuous decrease in lignin content were observed with increasing altitude. Gas chromatography analysis of monosaccharides determined that the highest cellulose and hemicellulose contents were obtained from the wood and pulp sample from 1000 m. Quantity of these components increased with increasing altitude. Lastly, kraft pulping degraded 14.10-15.70% of cellulose, 56.49-61.35% of galactoglucomannan, 65.75-69.61% of arabinoglucoronoxylan, and 92.42-93.48% of lignin in the samples.

Keywords: Brutian pine, chemical composition, kraft pulping, altitude.

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1. INTRODUCTION

The substantial reliance on wood, wood products, and paper in spite of a shortage of cellulosic and forestry resources has prompted wood and paper researchers to study in more detail the appropriate use of resources (Nemati and Samariha, 2011).

Cellulose, hemicelluloses, and lignin are the main chemical components of wood. Cellulose and hemicelluloses are the polysaccharides of the plant world; cellulose can comprise between 40% and 50% of wood, with hemicellulose making up another 15%-35%. Cellulose is constituted of glucose units that offer a linear and homopolymeric structure. In contrast, hemicelluloses present a branched heteropolymeric structure; in wood, they arise from pentoses, hexoses, and uronic acids. In softwood species, O-acetylgalactoglucomannan is the dominant hemicellulose polymer, comprising between 12% and 18%, depending on species. The main molecular chain of O-acetylgalactoglucomannan consists of β–D-glucopyranosyl and β–D-mannopyranosyl units that are linked linearly with one another by β-(1,4) bonds, with acetyl groups bonded to the -OH groups of the C2 and C3 atoms of β–D-mannopyranosyl. This main chain is then branched with α–D-galactopyranose units linked by α-(1,6) bonds. Another common softwood hemicellulose is arabino-4-O-methylglucoronoxylan, which makes up between 8% and 10%, depending on species. The main molecular chain of arabino-4-O-methylglucoronoxylan is constituted of β–D-xylopyranose units that are linked linearly with one another by β-(1,4) bonds; to this chain, α–L-arabinofuranose and 4-O-methyl-α-D-glucopyranosyluronic acid units are bonded by α-(1,3) and α-(1,2) bonds. The third major chemical component of wood, lignin, is a natural biopolymer complex constituted of phenylpropane units. Softwood lignins appear to consist primarily of guaiacyl units with small quantities of syringyl and p-hydroxyphenylpropane, while hardwood lignins appear to consist of guaiacyl and syringyl units with a small portion of p-hydroxyphenylpropane. After cellulose, lignin is the second most abundant biopolymer in the plant world, comprising between 20% and 32% of wood content (Fengel and Wegener, 1984; Krassig, 1993; Sjoström, 1993; Teleman et al., 2009; Kapu and Trajano, 2014; Persson and Jönsson, 2017).

In addition to being the main component in the production of paper, rayon, or cellulose derivatives, cellulose is an important raw material when converted into glucose units

(Fengel and Wegener, 1984; Sjoström, 1993). Like cellulose, hemicelluloses have attracted interest in recent years as raw materials for the production of derivatives, with hydrolysis of different hemicelluloses yielding different monosaccharide units (Sun et al., 1999, 2000, 2004; Yaşar, 2018; Yaşar and Kılınç, 2018). Most recently, a partial pre-extraction of hemicelluloses has been proposed that can be carried out without affecting the quality or yield of pulp produced for use in the pulp and paper industry (Van Heiningen, 2006). This provides a source of hemicelluloses for diverse industrial processes. For wood monosaccharides, including glucose, mannose, xylose, galactose, arabinose, rhamnose, and uronic acids, the main utilization is to produce other industrial chemicals. The third main component of wood, lignin, can be utilized in a wide range of applications, from fuels to advanced chemicals and materials. However, about 85% of the world's total lignin production consists of kraft lignin, which is a kind of industrial lignin recovered from kraft pulp. About 630,000 tons of kraft lignin is produced annually, and is primarily used for heat recovery in the form of combustion, which is a low-value use (Wegener, 1982a, 1982b; Fengel and Wegener, 1984; Sjoström, 1993; Yoon and Van Heiningen, 2008; Vila et al., 2011; Hongzhang, 2015).

Brutian pine (*Pinus brutia* Ten.) is distributed extensively throughout the Mediterranean, Aegean, and Marmara regions, and also in some localities of the western Black Sea region. In Turkey, the total growing area of Brutian pine is approximately 5.8 million hectares, which is 25.11% of the nation's forests (OGM, 2015). Brutian pine is the leading species for Turkish wood production, with 5140007 m^3 being used as industrial wood and 1055335 m^3 as fuelwood (OGM, 2016). The wood is used in the production of construction materials, packing cases, agricultural tools, telegraph poles, mine poles, palings, and watercraft (Bozkurt, 1971, 1982). In addition, it is a useful source for paper production using the kraft (sulfate) process (Erten and Taşkın, 1985; Öktem, 1987). Moreover, Brutian pine wood is one of the most important sources of raw material for use in the production of particle board and fiberboard (Özdemir and Uçar, 2016). Finally, Brutian pine bark can be used in tannin production (Erten and Taşkın, 1985; Öktem and Sözen, 1995).

Recently, an integrated bio-refinery that can extract a portion of hemicellulose content while retaining cellulose for the production of pulp has come into prominence (Van Heiningen, 2006). As kraft lignin obtained from kraft pulp comprises 85% of the total lignin production of the world (Hongzhang, 2015), this study investigated the chemical composition of Brutian pine woods harvested from different altitudes and of kraft pulps produced from the same woods. Moreover, the effect of harvesting altitude on the chemical composition of wood and pulp was evaluated.

2. MATERIAL AND METHODS

2.1. Material

Wood samples were obtained from Ağlasun-Burdur in Turkey. Three trees were harvested, one each from 800 m, 900 m, and 1000 m above sea level. A disk with a thickness of 50 cm was taken from a height of 1.30 m on the trunk of each tree. The disks were separately debarked, chipped, and dried naturally in an air-conditioned room for a month.

2.2. Method

Kraft pulping

Kraft pulping was performed to cook the Brutian pine wood chips. Each trial used 500 g Brutian pine chips (on an oven-dry basis). Pulping was carried out automatically in a controlled way in the laboratory with a 15 L stainless steel reactor rotating at four rotations per minute and heated by electricity. The conditions of kraft pulping were as follows (Gülsoy et al., 2015): The active alkali concentration of the pulping liquor was 20%, with equal sulfidity of 25%. Cooking was carried out for 75 minutes at a maximum temperature of 170 ℃. The ratio of liquor to wood was 4:1. After cooking, the treated chips were washed with water to remove the black liquor, and then disintegrated. To determine the effect of kraft cooking on the entire sample, the pulp was used directly for chemical analysis, without any separation of rejected or screened pulp. For this reason, the total yield of each sample was used in the study, expressed as a percentage of oven-dried material weight.

Chemical analyses

For chemical analyses, wood chips were milled in a Retsch SK-1 mill so as to pass through 40 to 100 mesh. To remove extractives, the milled wood and pulp samples were extracted with cyclohexane:ethanol (2:1 v/v) and then ethanol in a Buchi Extraction System B-811. To determine holocellulose and α-cellulose contents in extract-free wood and pulp samples, the acid chlorite method developed by Browning (1967) and the ASTM D 1103 method (1980) were applied. Holocellulose and α-cellulose contents were calculated as percentages of oven-dried material weight. The lignin contents of the wood and pulp samples were determined on a Perkin Elmer Lambda 20 UV/visible Spectrometer using the acetyl bromide procedure introduced by Liyama and Wallis (1990). Lignin content was reported as a percentage of oven-dried material weight. Extract-free wood and pulp samples were hydrolyzed using the acid hydrolysis method developed by Dill et al. (1984) with a slight modification (Yaşar et al., 2010). The monosaccharides in the acid hydrolysates of the extract-free wood and pulp samples were analyzed using a Perkin Elmer Autosystem XL gas chromatograph according to the method developed by Cao et al. (1997). Monosaccharide contents were calculated as percentages of oven-dried material weight. Finally, uronic acid content was analyzed on a Perkin Elmer Lambda 20 UV/visible Spectrometer using the method described by Filisetti-Cozzi and Carpita (1991) and expressed as a percentage of oven-dried material weight.

Statistical analysis

Statistical analyses were done using Minitab 16 software. If a significant difference between the mean values of the

samples was found by ANOVA, then Duncan's test was performed to determine the different groups.

3. RESULTS AND DISCUSSION

The main chemical components of Brutian pine wood and pulp samples are listed in Table 1. The holocellulose contents of wood samples were in agreement with the values of 70.8%, 72.57%, 72.07%, 72.64%, and 72.09% reported for Brutian pine wood by Kırcı (1991), Kılıç et al. (2010), Güler and Yaşar (2018), Beram and Yasar (2020), and Güler and Yaşar (2020). Similarly, the α-cellulose contents of wood samples were consistent with the values of 48.56%, 46.5%, 48.16%, 48.23%, and 48.24% observed by Tutuş et al. (2012), Taş (2017), Güler and Yaşar (2018), Beram and Yasar (2020), and Güler and Yaşar (2020). Lignin contents of wood samples in this study were higher than the previously reported klason lignin contents of 27.4%, 27.34%, 27.60%, 27.16%, 27.98%, and 27.33% demonstrated by Kırcı (1991), Kılıç et al. (2010), Tutuş et al. (2012), Güler and Yaşar (2018), Beram and Yasar (2020), and Güler and Yaşar (2020). However, klason lignin content reflects only the acid-insoluble part of the total lignin content of wood (Rodrigues et al., 1999). For Brutian pine pulp, Taş (2017) reported holocellulose and klason lignin contents of 94.86% and 5.77%, respectively. The holocellulose contents of pulp samples in this study were consistent with the result of Taş (2017), while the lignin contents were higher; as with the wood samples, this is because klason lignin in the pulp represents the acid-insoluble part of total lignin content (Koljonen et al., 2004; Shin et al., 2004).

Table 1. Main chemical components of Brutian pine wood and pulp samples.

Sample	Holocellulose (%)	α-cellulose (%)	Lignin (%)
Wood-800 m	71.82 (0.09)[1] a[2]	47.36 (0.08) a	30.83 (0.08) a
Wood-900 m	72.04 (0.16) a	47.91 (0.09) b	30.02 (0.07) b
Wood-1000 m	72.82 (0.18) b	48.33 (0.11) c	29.75 (0.09) b
Pulp-800 m	94.41 (0.02) a	84.01 (0.03) a	5.34 (0.04) a
Pulp-900 m	94.42 (0.02) a	84.07 (0.01) a	5.13 (0.08) b
Pulp-1000 m	94.67 (0.10) b	84.34 (0.23) b	4.27 (0.03) c

[1] Standard deviation, [2] Homogenous groups by Duncan test: $p < 0.001$ for wood holocellulose, wood α-cellulose, and wood lignin; $p < 0.01$ for pulp holocellulose; $p < 0.05$ for pulp α-cellulose; and $p < 0.001$ for pulp lignin.

The holocellulose and α-cellulose contents of pulp samples produced from Brutian pine wood samples using kraft pulping increased slightly with increasing altitude, whereas lignin contents were reduced (Table 1). The same trends were observed in wood samples. Generally, the wood sample findings here are consistent with observations by Dönmez et al. (2013) in Scots pine wood samples, for which a continuous increase in altitude from 822 m to 1031 m led to continuous increase in holocellulose and α-cellulose contents and decrease in klason lignin contents. However, Musule et al. (2016) reported continuous increase in cellulose and klason lignin contents of oyamel fir wood samples with increasing altitude from 3000 m to 3500 m.

After kraft pulping, the total yield of pulp obtained was 43.77%, 43.99%, and 44.01% for samples of Brutian pine harvested from 800 m, 900 m, and 1000 m altitudes, respectively. This corresponded to 218.85 g, 219.95 g, and 220.05 g of pulp (on an oven-dry basis) being obtained from 500 g of dry chips (the starting material for cooking).

In the starting materials, holocellulose contents were 359.10 g, 360.20 g, and 364.10 g for samples from 800 m, 900 m, and 1000 m, respectively. After kraft pulping, 206.62 g, 207.68 g, and 208.32 g holocellulose was recovered in the pulp samples (Figure 1). Thus, kraft cooking reduced the holocellulose content by 42.46%, 42.34%, and 42.79% respectively for samples harvested from 800 m, 900 m, and 1000 m. The decrease in holocellulose content is explained by polysaccharide degradation during the cooking process due to peeling reactions and alkaline hydrolysis (Fengel and Wegener, 1984; Krassig, 1993; Sjoström, 1993).

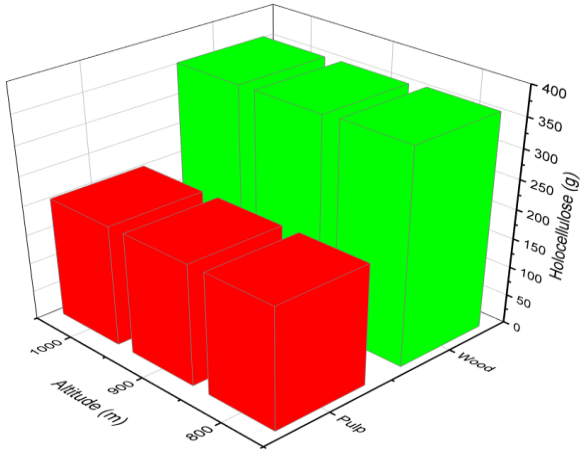

Figure 1. Change in Brutian pine holocellulose content during kraft pulping

Brutian pine wood (500 g dry chips) had α-cellulose contents of 236.80 g, 239.55 g, and 241.65 g for samples collected at 800 m, 900 m, and 1000 m, respectively. In the pulps produced by cooking, the corresponding contents were 183.86 g, 184.91 g, and 185.59 g (Figure 2), representing decreases of 22.36%, 22.81%, and 23.20%, respectively.

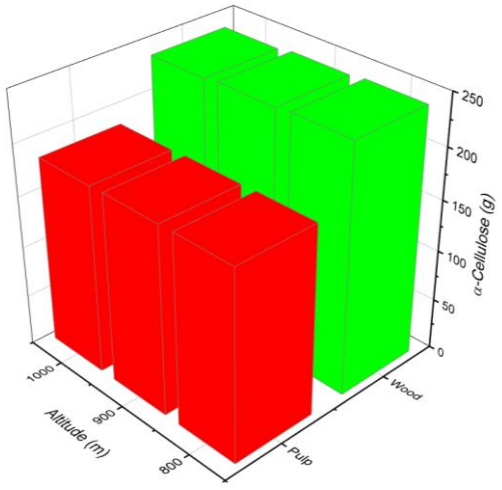

Figure 2. Change in Brutian pine α-cellulose content during kraft pulping

Meanwhile, lignin contents of the starting materials were 154.15 g, 150.10 g, and 148.75 g respectively for samples collected at 800 m, 900 m, and 1000 m. In pulp samples, the cooking process decreased lignin content to 11.69 g, 11.28 g, and 9.37 g (Figure 3), corresponding to losses of 92.42%, 92.48%, and 93.68% during cooking. The findings for lignin content are in agreement with Huang et al. (2015), who reported that 93.9%-95.5% of lignin could be removed from wood with the use of 22% effective alkali and 20% sulfidity and cooking at a temperature of 160 ℃ for 70 minutes. Similarly, Yuan et al. (2017) removed 97.54% of lignin from wood chips during kraft pulping (170 °C, effective alkali 22%, sulfidity 25%).

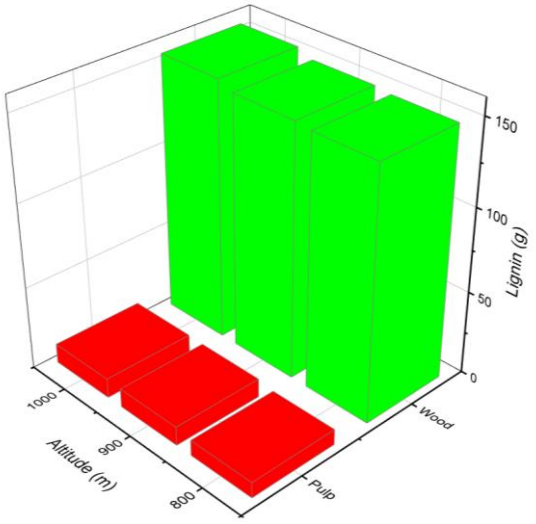

Figure 3. Change in Brutian pine lignin content during kraft pulping

The monosaccharide units that constituted Brutian pine polysaccharides are presented in Figure 4, and consisted of glucose, galactose, mannose, xylose, and arabinose in both wood and pulp.

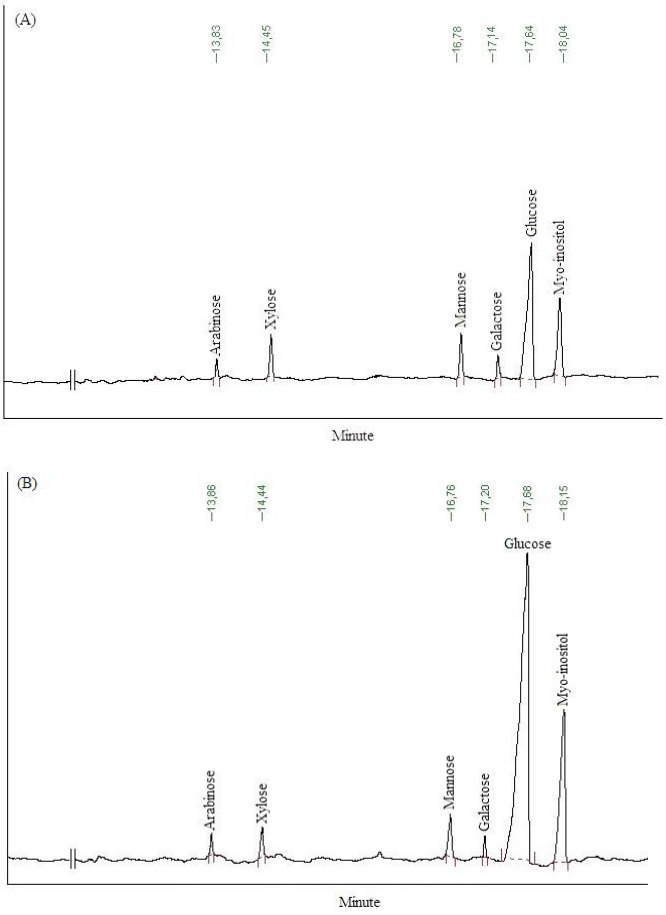

Figure 4. GC chromatograms of Brutian pine wood (A) and pulp (B) samples

Monosaccharide and uronic acid contents of Brutian pine wood and pulp samples are indicated in Table 2. The monosaccharide composition of Brutian pine wood samples was in agreement with observations by Yaşar (2014), Yaşar and Güler (2018), and Güler and Yaşar (2020). Glucuronic acid was not found in the pulp samples, as that this unit is more prone to decomposition in kraft pulping than other monosaccharide units (Fengel and Wegener, 1984).
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Table 2. Monosaccharide and uronic acid composition of Brutian wood and pulp samples.

Sample	Glu (%)	Man (%)	Xyl (%)	Gal (%)	Ara (%)	UA (%)
Wood-800 m	47.61 (0.03)[1]	10.78 (0.01)	7.82 (0.02)	2.13 (0.01)	1.07 (0.01)	1.56 (0.01)
Wood-900 m	48.16 (0.02)	11.02 (0.02)	7.99 (0.01)	2.18 (0.02)	1.09 (0.01)	1.60 (0.01)
Wood-1000 m	48.89 (0.04)	11.24 (0.02)	8.15 (0.02)	2.22 (0.01)	1.12 (0.01)	1.63 (0.01)
Pulp-800 m	87.85 (0.01)	4.40 (0.01)	2.70 (0.01)	0.51 (0.01)	0.48 (0.01)	-
Pulp-900 m	87.91 (0.03)	4.53 (0.01)	2.90 (0.01)	0.55 (0.01)	0.52 (0.01)	-
Pulp-1000 m	88.20 (0.02)	5.17 (0.01)	3.17 (0.01)	0.60 (0.01)	0.56 (0.01)	-

Glu: Glucose, Man: Mannose, Xyl: Xylose, Gal: Galactose, Ara: Arabinose, UA: Uronic acid, 1: Standard deviation.

The determined glucose contents were related to the cellulose and O-acetylgalactoglucomannan contents of Brutian pine wood. For softwoods, the reported mannose:glucose ratio in O-acetylgalactoglucomannan is 3:1 (Timell and Mian, 1960; Fengel and Wegener, 1984). Using this ratio, the glucose content of each wood and pulp sample was calculated and then combined with mannose and galactose contents to indicate the galactoglucomannan content. The remainder of the determined glucose content was attributed to cellulose and used to calculate the cellulose content of each sample. Pekgözlü et al. (2020) similarly determined the rhamnose and galacturonic acid

contents of Brutian pine samples, which compose the pectin in wood, but rhamnose was not found in the samples in this study.

In addition, the xylose:methylglucuronic acid:arabinose ratio of arabino-4-O-methylglucoronoxylan in softwoods was reported to be 8:1.6:1 (Fengel and Wegener, 1984). Here, the determined ratios for Brutian wood samples were 7.82:1.56:1.07, 7.99:1.60:1,09, and 8.15:1.63:1.12, which are consistent with observations by Fengel and Wegener (1984). This is meaningful as the uronic acids determined in the present study can be ascribed to glucuronic acids. Xylose, uronic acid, and arabinose contents were therefore used to calculate the arabinoglucoronoxylan content of each sample. However, when composing the polymeric structure of polysaccharides in wood, the monosaccharide and uronic acid units lose 1 mol H_2O (Fengel and Wegener, 1984; Krassig, 1993; Sjoström, 1993). Therefore, we used a conversion factor of 0.9 when determining the cellulose and hemicellulose (galactoglucomannan + arabinoglucoronoxylan) contents of each sample (Table 3).

Table 3. Polysaccharide composition of Brutian wood and pulp samples.

Sample	Cellulose (%)	Galactoglucomannan (%)	Arabinoglucoronoxylan (%)	Hemicellulose (%)
Wood-800 m	39.62	14.85	9.41	24.26
Wood-900 m	40.04	15.19	9.61	24.80
Wood-1000 m	40.63	15.49	9.81	25.30
Pulp-800 m	77.75	5.74	2.86	8.60
Pulp-900 m	77.76	5.93	3.08	9.01
Pulp-1000 m	77.83	6.74	3.36	10.10

Brutian pine wood (500 g of chips) had cellulose contents of 198.1 g, 200.20 g, and 203.15 g for samples collected at 800 m, 900 m, and 1000 m, respectively. In the produced pulp samples, the corresponding contents were 170.16 g, 171.03 g, and 171.26 g (Figure 5), representing decreases of 14.10%, 14.57%, and 15.70% due to the kraft cooking. The findings here are supported by Huang et al. (2015) and Yuan et al. (2017). Huang et al. (2015) indicated that over 80% of glucan could be retained in pulped wood with milder sulfidity conditions (i.e. 14 to 24%). Similarly, Yuan et al. (2017) reported the extraction of 20.80% of glucan from original wood chips during kraft pulping (170 °C, effective alkali 22%, sulfidity 25%).

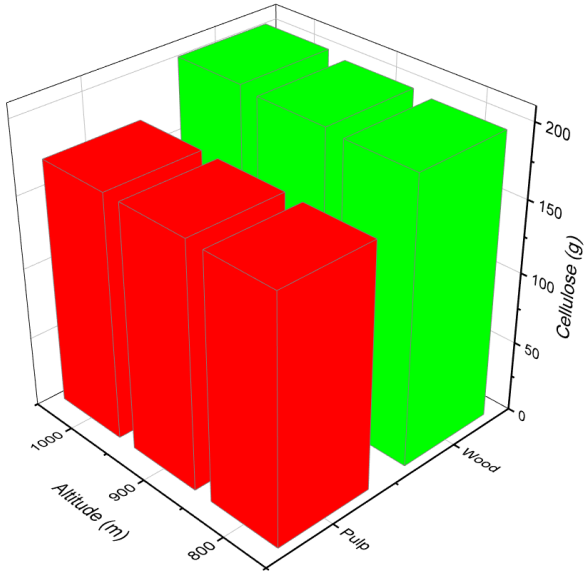

Figure 5. Change in Brutian pine cellulose content during kraft pulping

The hemicellulose contents of the wood samples were 121.30 g, 124.00 g, and 126.50 g respectively when harvested at 800 m, 900 m, and 100 m altitude. Meanwhile, the corresponding pulp contents were 18.82 g, 19.82 g, and 22.23 g (Figure 6), indicating respective degradation of 84.48%, 84.02%, and 82.43% of hemicelluloses during kraft pulping. The findings here are compatible with observations by Yuan et al. (2017), who reported that over 75% of hemicelluloses were extracted from original wood chips during kraft pulping (170 °C, effective alkali 22%, sulfidity 25%).

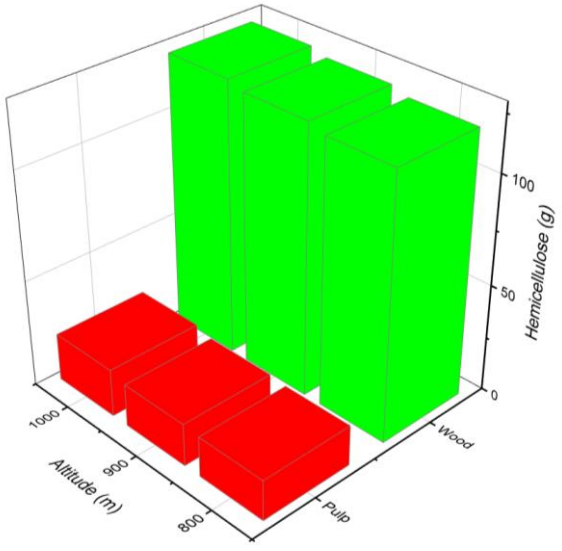

Figure 6. Change in Brutian pine hemicellulose content during kraft pulping

Taken together, the wood sample collected at 1000 m offered more cellulose and hemicellulose contents and less lignin content than did the wood samples collected at 800 m and 900 m. Similarly, the pulp produced from the wood

sample collected at 1000 m yielded the highest cellulose and hemicellulose contents, but the lowest lignin content.

4. CONLUSION

In this study, the chemical properties of Brutian pine wood samples, which are used for pulp production by kraft pulping, were investigated in the context of harvesting altitude (800 m, 900 m, and 1000 m). Moreover, the chemical composition of produced pulps was determined. Proximate analysis showed that increasing the harvest altitude of Brutian pine wood could raise the holocellulose and α-cellulose contents of wood and pulp, while decreasing the lignin content. GC analysis indicated a continuous increase in cellulose and hemicellulose contents with increasing altitude. Kraft pulping was determined to degrade 14.10-15.70% of cellulose, 56.49-61.35% of galactoglucomannan, 65.75-69.61% of arabinoglucoronoxylan, and 92.42-93.48% of lignin. After pulping, a considerable quantity of extracted polysaccharide was obtained that could be used in the food, pharmacology, and bio-refinery industries. The lignin content, which is generally considered an energy resource for heat recovery in the form of combustion, was remarkably degraded during kraft pulping.

Ethics Committee Approval
N/A

Peer-review
Externally peer-reviewed.

Author Contributions
All authors have read and agreed to the published version of manuscript.

Conflict of Interest
The authors have no conflicts of interest to declare.

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