

Hemicellulose content in rice straws of several high-quality rice grains

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Abstract:

A by-product of rice farming, rice straw is made up of lignin, cellulose, and hemicellulose and comprises lignocellulosic biomass. Particularly rich in this biomass are rice-producing nations like Vietnam, which ranks sixth in the world for rice exports. Because 30-40% of this bioresource is burned to make way for new rice fields for cultivation, its use has not been optimum. Air pollution has been linked to this, particularly in Vietnam. Indeed, burning half of the world's rice straw produces almost 100 million tons of carbon dioxide emissions. Therefore, to increase the benefits of rice and reduce environmental pollution, lignocellulosic biomass reuse is crucial. This paper reports the hemicellulose content in three rice straw types (OM5451, IR50404, and 6976 commons from An Giang province, Vietnam). Alkaline extraction assisted with ultrasound was employed. In this process, samples were mixed with 2 M sodium hydroxide and ultrasonicated for 30 min at 90°C. Then, the mixture was continuously heated at 90°C and stirred at 40 rpm for 1.5 h. Ethanol was used to precipitate hemicellulose. The highest yields obtained of crude hemicellulose were 23.17% in OM5451, 23.1% in IR50404, and 22.94% in 6976 at pH 4.0, however, there was no significant difference at a 95% confidence level as determined by a two-way ANOVA with p-values >0.05. The extracted hemicellulose was confirmed using Fourier-transform infrared spectroscopy (FTIR), X-ray diffraction (XRD), and thermo-gravimetric analysis (TGA).

Keywords: alkaline extraction, biomass, circular agriculture engineering (CAE), gravimetry, hemicellulose, rice straw.

Classification number: 2.2

1. Introduction

Rice straw, a by-product of rice cultivation, contains lignocellulosic biomass and is composed of lignin (5-24%), cellulose (32-47%), and hemicellulose (19-27%) [1, 2]. This biomass is particularly abundant in rice countries such as Vietnam (the fifth in global rice exportation) [3]. However, utilizing this bioresource has not been optimized as 30-40% is burned to clear rice fields for the next cultivation seasons. This has been known to cause air pollution, especially in Vietnam [4]. Indeed, approximately 100 million tons of carbon oxide is emitted from burning 50% of global rice straw [5]. Therefore, the reuse of lignocellulosic biomass is much needed to improve rice benefits and mitigate environmental pollution [6].

Hemicellulose is the second-most abundant class of short-chain polysaccharides, which is different from cellulose and is branched in nature [7, 8]. As a non-crystalline heteropolysaccharide, hemicellulose is made of pyranoses and furanoses sugar including xylans (predominant materials in plant cell walls), xyloglucans, manans, and other compounds of linkage β -glucans.

The main acid groups of hemicellulose make them very hydrophilic and soluble in alkaline. Hemicellulose is easily hydrolysed by dilute acid, i.e., HCl and H₂SO₄, or bases [9]. Hydrolysis of hemicellulose can produce xylans that are widely used in commercial products such as various pharmaceuticals, food, and biofuels [10].

Hemicellulose from plant cells can be isolated by ionic liquid extraction, organic solvent solution, alkaline treatment, and liquid hot water extraction. Of these methods, the alkaline method is most commonly used in industries and labs due to its efficiency. While sodium hydroxide solution is applied for the hydrolysis of the soft plant cell wall, potassium hydroxide solution is usually used for hardwood [11-13]. After hydrolysis, ester linkages will be cut and this process produces hemicelluloses. Then, ethanol can be used to precipitate hemicelluloses from the alkaline extraction [14]. The efficiency of hemicellulose can be increased by using ultrasound, which can help reduce the extraction time down to 1.5-2.5 h. The ultrasound waves easily break rice straw cells by disturbing the cells of the biomass and thus promote hemicellulose extraction [15-17].

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An Giang is a province of Vietnam with the largest rice production in the Mekong delta with approximately 4 million tons in 2017 [18, 19]. Common rice varieties in An Giang with high yield and good grain quality are OM5451, IR50404, and 6976 [20]. The amount of rice straw generated was about 3891 thousand tons, of which 62% was used for composting, cattle feed, and selling while 36.36% was open-burned. As reported, the highest emission of open-burning is CO₂ with 5.7 million tons while other emissions are CO, SO₂, NO₂, PM_{2.5} and PM₁₀ with 135.1, 7.78, 0.28, 54.4, and 14.4 thousand tons, respectively [21]. Since rice straw is a bioresource rich in carbon, nitrogen, and potassium, such open-burning contributes to global air pollution. Therefore, recycling rice straw is necessary to reduce negative environmental impacts.

The temperature and concentration of the alkaline solution in the hemicellulose extraction process significantly affects hemicellulose yield [22]. For example, high yields of lignin, hemicellulose, and nanocellulose fibres separated from rice straw were collected with a 2 M NaOH solution at 90°C. Another work indicated a higher extraction yield versus pH conditions [23]. On the other hand, hemicellulose is a group of polysaccharides in biomass and they possess different properties depending on the variety of biomass [24]. Therefore, the effect of pH values on hemicellulose precipitation in ethanol and the comparison of hemicellulose characteristics generated from sources were two factors investigated in this study. Knowing the hemicellulose concentration in rice straws can aid in optimizing their benefits and recycling. Rice straw from the rice cultivars in An Giang may contain various hemicellulose concentrations, which have not been well studied. Therefore, this research focused on revealing the hemicellulose contents in rice straws generated from those cultivars to provide background data for rice straw hemicellulose studies.

2. Materials and methods

2.1. Sample preparation and materials

Rice straws of OM5451, IR50404, and 6976 were collected from paddy fields in Cho Moi district, An Giang province. All samples (10 kg) were firstly washed by distilled water (room temperature) to remove fine sand particles, then sun-dried for seven days to have the average sample moisture of 4-5.5%. The samples were milled to a size of 1 mm to obtain dried rice straw (DRS)

for all the experiments as suggested by Kim, et al. (2020) [25]. After that, the samples were cut into small forms, finely ground (sieve screen: $\phi=0.08$ mm), kept in airtight containers, and stored at room temperature [26].

The chemicals of acid hydrochloric, ethanol 99,5%, sodium hydroxide, acid perchloric, and acetone were purchased from Merck, Germany.

2.2. Extraction of hemicellulose

First, 360 ml of acetone 5% was added to 15 g DRS in a Soxhlet system controlled at 70°C for 4 h, which then became the extracted rice straw (ERS). After 4 h, the ERS was dried in an oven at 105-110°C to constant mass [27].

Each ERS sample (10 g) was first mixed with NaOH 2 M by the ratio of 1 g straw ratio: 20 ml NaOH 2 M and ultrasonicated for 30 min at 90°C. An S100-Elmasonic was used to create the ultrasound waves. After that, the mixture was heated at 90°C and continuously stirred at 40 rpm for 1.5 h. At the end of the 1.5 h period, vacuum filtration was used to collect the filtrate containing hemicellulose. Then, hydrochloric acid 6 M was added to adjust the filtrate pH to the values of 3.5; 4.0; 4.5; and 5.0. The mixture was maintained to stand at 4°C for 24 h. Then, three volumes (500 ml) of ethanol 95% were added to the liquid fraction and this mixture was kept at 4°C for 6 h to precipitate hemicelluloses at the bottom. Vacuum suction was employed to remove the clear solution above the hemicellulose precipitate. The precipitate was washed 3 times with 70% ethanol to remove the others. The extracted hemicellulose was dried under sunlight to constant mass. The crude hemicellulose (CH) yield was the difference between the dried CH and ERS. All samples are presented in Table 1.

Table 1. Hemicellulose extraction samples.

pH values	3.5	4.0	4.5	5.0
Extracted rice straw types	OM5451	OM5451	OM5451	OM5451
	IR50404	IR50404	IR50404	IR50404
	6976	6976	6976	6976
The fixed factors in the extraction experiments:				
Sodium hydroxide concentration (mol/l):	2 M			
Ultrasonication time (min):	30			
Reaction temperature (°C):	90			
Hydrochloric acid concentration (mol/l):	6 M			
Replicates:	3			

2.3. Hemicellulose characterization

Hemicellulose yields:

The yield of pH was determined regarding the reported study and calculated by using Eq. (1) [28]:

$$\text{Yield} = \frac{m_1}{m_0} \times 100\% \quad (1)$$

where m_1 : the mass of crude hemicellulose; m_0 : the mass of extracted rice straw (ERS) employed in above section.

Hemicellulose characteristics:

This study used XRD and TGA to assess hemicellulose characteristics [29, 30]. Hemicelluloses were characterized using FTIR with an Alpha Bruker spectrophotometer with a resolution of 4 cm^{-1} in the range of $400\text{--}4500 \text{ cm}^{-1}$.

XRD was performed with an Aeris Benchtop X-ray Diffractometer Malvern PANalytical to investigate the phase and crystallinity of the hemicelluloses, of which the XRD patterns were recorded in the region of 2θ from $5\text{--}40^\circ$ [31].

Thermogravimetric analyses were applied to investigate hemicellulose thermal degradation [32]. In this paper, the thermal decompositions of samples were measured on a TGA instrument Q5000 with temperature ranging from ambient temperature (28°C) to 600°C with nitrogen as the purge gas at a rate of 40 ml/min .

Statistical analysis:

All experiments were performed in triplicate. Data were analysed using a two-way ANOVA to determine the significant differences of variance.

3. Results and discussion

3.1. Hemicellulose extraction

Results of raw hemicellulose yields were showed in Table 2. The total crude hemicellulose obtained varied in the range of $15.12\text{--}23.09\%$ for all samples. A two-way ANOVA was used to analyse the hemicellulose yield variance with one dependent variable (yield) and two factors (pH and grains). ANOVA test results showed a statistically significant difference in average hemicellulose yield according to pH values at the 95% significance level ($p < 0.05$), whereas there was no significant difference at 95% confidence level between experiments of grains in the same treatment of pH level ($p = 0.330$).

Table 2. Hemicellulose yields from three rice straw grains

Classes	pH value			
	3.5	4.0	4.5	5.0
OM5451	18.05 ± 0.07^a	23.17 ± 0.48^b	20.07 ± 0.29^c	15.21 ± 0.32^d
IR50404	17.97 ± 0.07^a	23.11 ± 0.13^b	19.98 ± 0.11^c	15.06 ± 0.06^d
6976	18.10 ± 0.14^a	22.94 ± 0.08^b	20.02 ± 0.11^c	15.08 ± 0.11^d
Average yields	18.04 ± 0.09	23.09 ± 0.23	20.09 ± 0.17	15.12 ± 0.17

Note: F-value: 455.762, CV: 1.3, Means $\pm$ SE^{a,b,c,d} (i.e. 18.05 ± 0.07^a) with difference letters are significantly different at 95% confidence level.

Hemicelluloses are soluble in the dilute alkali because of the deprotonated hydroxyl groups on hemicelluloses. The yield of hemicellulose started to increase from 15.12 and 20.09% at pH 5 and pH 4.5, respectively. This could be explained when the pH of the liquid started to decrease, the pronation dominated and hemicellulose precipitation occurred. At pH 4.0, hydroxyl groups on hemicelluloses were neutralized, so the yield of raw hemicellulose was 23.09% . However, at pH 3.5, the average yield of three rice straw grains was 18.04% because the glycosidic bonds of hemicellulose can be broken in an acidic medium and promote hemicellulose degradation with lower pH [33]. The hemicellulose yields can increase from pH 5–4.5 because of the high lignin contents released due to lower pH (Fig. 1) [34].

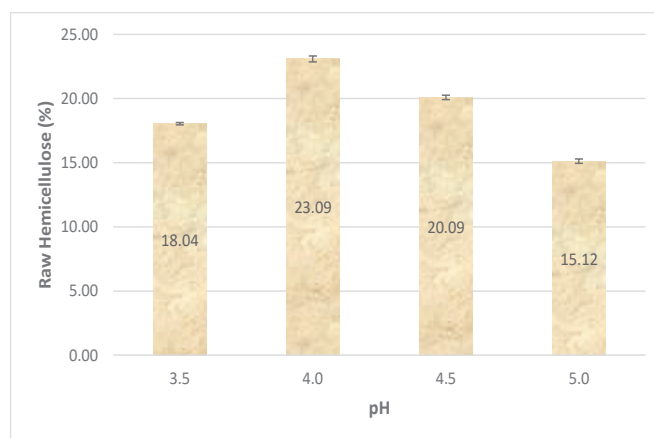


Fig. 1. Effect of pH on the yield of hemicellulose.

3.2. Hemicellulose characteristics

Figure 2 obtained from FTIR shows that the structures of the three hemicelluloses (OM5451, IR50404, 6976) within the spectra of $500\text{--}4000 \text{ cm}^{-1}$ were similar, but the contents of functional groups were different. The broadbands were between 3453.37 and 3451.76 cm^{-1} , at which signals of hydroxyl groups were present in the hemicellulose components [35]. The presence of

an absorption band at 2362.13 cm^{-1} was understood as the stretching vibrations of $\text{O}=\text{C}=\text{O}$. The carbonyl stretching region was at 1644.13 cm^{-1} from 1326.54 to 1476.54 cm^{-1} , which was the appearance of OH or CH_2 linkage [36]. As shown in Fig. 2, the typical spectrum region of hemicellulose ranged from 850 to 1200 cm^{-1} encompassing the absorption band at about 1157.71 – 1159.13 cm^{-1} , which indicated the presence of C–O vibration, while the absorption band at 1093 and 1095 cm^{-1} was that of the C–O–C stretching glycosidic bonds in the xylan groups [37, 38]. The absorption at 471 to nearly 800 cm^{-1} was low because the samples contained lignin components. In general, the three rice straw samples' FTIR spectra had similar functional groups and bands as the hemicelluloses extracted from the different varieties of the rice straw carried out by previous studies mentioned.

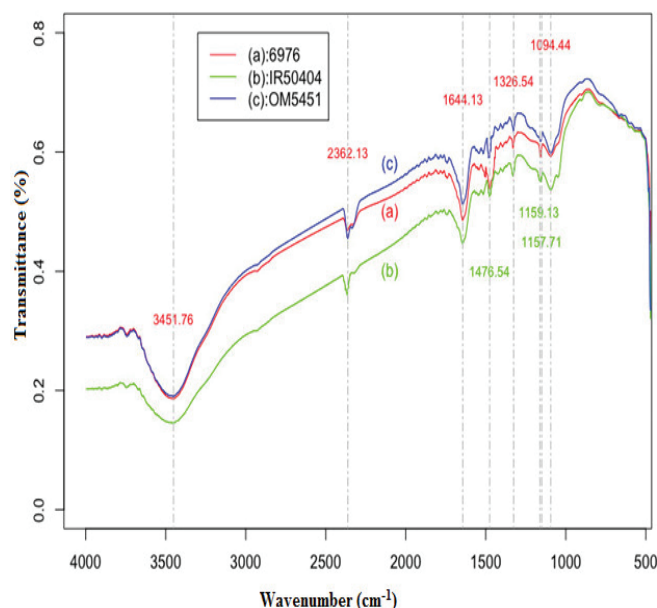


Fig. 2. FTIR spectra of hemicellulose extracted from rice straw.

The XRD patterns (Fig. 3) of the hemicellulose samples had a wide peak at a 2θ angle close to 22° , which indicates the amorphous nature of hemicellulose [39]. The height of the peaks at $2\theta=22^\circ$ was clearly. They had a non-crystalline structure related to their heterogeneous chemical structure [40]. These structures were similar among the samples. Compared to the XRD analysis of hemicelluloses from untreated rice straws investigated by [41], the crystallinity regions of this study were broader.

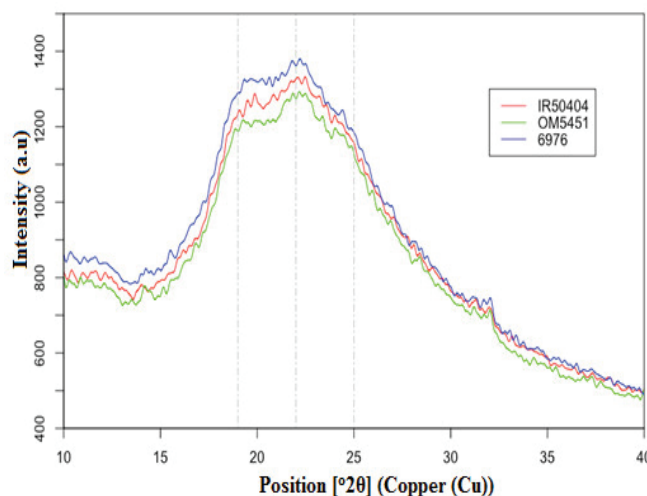


Fig. 3. XRD analysis of rice straw hemicelluloses.

Figure 4 shows the TGA curves of OM5451, IR50404, and 6976, respectively. The initial weight loss at about 50°C was related to the release of moisture in the samples. The decomposition of hemicellulose started easily, with weight loss commonly occurring between 270 and 300°C . The weights of the three samples suddenly decreased from 250 to 300°C . At 280°C , the weight loss of the three samples was approximately 15 to 42% . When the temperature was raised to 400°C , the weight loss reached nearly 53% . The TGA characteristics of the three hemicellulose samples were the same, which were not stable at higher temperatures ($>250^\circ\text{C}$). The experiment outcome indicated that the thermal degradation of rice straw hemicellulose occurred at about 250°C , which agrees with previous studies [42].

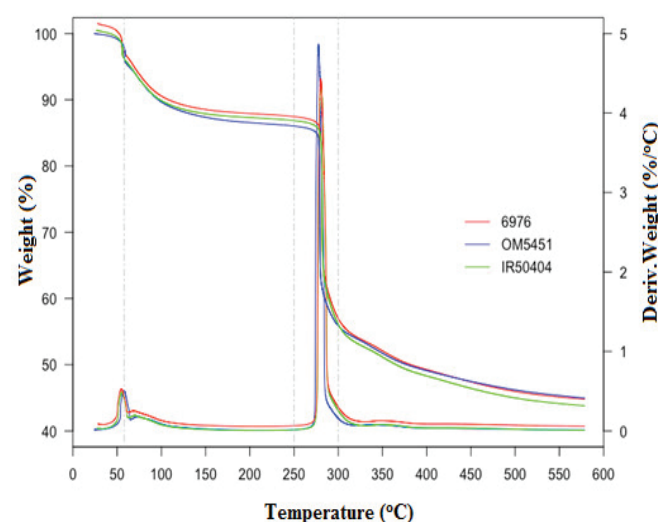


Fig. 4. TGA of rice straw hemicelluloses.

Conclusions

The hemicellulose contents of three rice straws did not significantly differ at each pH. We were able to obtain the highest yield of hemicellulose of 23.09% at pH 4.0. The FTIR results showed lignin in the samples, which could affect the purity of the extracted hemicellulose. XRD revealed the amorphous region was at $2\theta=22^\circ$. TGA indicated the weight loss of hemicellulose samples was nearly 53% at 400°C. This study is a comprehensive demonstration of hemicellulose in rice straws of common rice grains from the An Giang Province. We highlight that the application of the simple techniques used in our study efficiently extract hemicellulose.

CRedit author statement

Thuy-An Ngo: Conceptualization, Methodology, Software, Writing and Editing; Dao-Chi Vo Thi: Data curation, Writing - Original draft preparation; Nhan-Tanh Nguyen Tran: Conceptualisation, Methodology and Writing.

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COMPETING INTERESTS

The authors declare that there is no conflict of interest regarding the publication of this article.

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