Effect of Chemical Composition on Thermal Decomposition Behavior of Herbaceous Plants for Production of Plant-based Biochar for Storing Carbon in Soils

Keishi Tanifuji,[a,b,*] Kensuke Sakurai,[a] Toyohisa Miyamoto,[a,*] and Yuji Okayasu [a]

In order to use plant biomass as biochar for storing carbon in soils, the relationship between the chemical composition of plants and biochar yield obtained by thermogravimetric (TG) analysis was studied. The extractive content of herbaceous plants such as dokudami (*Houttuynia cordata*), halcyon (*Erigeron philadelphicus*), and mugwort (*Artemisia* Spp.) was higher than that of Japanese cypress (*Chamaecyparis obtusa*), and the yields at 250 and 300 °C tended to decrease with increasing extractive content of plants. This indicated the possibility that thermal decomposition of herbaceous plants can be conducted at low temperatures (below 350 °C). In addition, the content of crystalline carbohydrates (remaining upon treatment with 5% sulfuric acid) of herbaceous plants was lower than that of Japanese cypress, and the yield at 400 °C tended to decrease with increasing crystalline carbohydrate content of plants. This indicated the possibility that herbaceous plants can be used to obtain biochar with higher yield at 400 °C than woody plants. Therefore, herbaceous plants are considered to be feasible resources as raw materials for biochar production for storing carbon in soils.

DOI: 10.15376/biores.18.3.5057-5072

Keywords: Biochar; Chemical composition of plants; Thermogravimetric analysis; Herbaceous plant

*Contact information: a: Innovative Materials and Resources Research Center, Public Works Research Institute, Tsukuba, Ibaraki, 305-8516, Japan; b (present affiliation): Forestry and Forest Products Research Institute, Tsukuba, Ibaraki, 305-8687, Japan; *Corresponding authors: tanifujik4@affrc.go.jp (present), recycle-imarrc21@pwri.go.jp*

INTRODUCTION

Large amounts of plant biomass are obtained *via* public works such as mowing and pruning along rivers, roads, and parks. The authors' laboratory has focused on utilizing this biomass in wastewater treatment plants (WWTPs). We have studied the biomass utilization for methane generation by mixed anaerobic digestion with sewage sludge (Hidaka *et al.* 2013; Wang *et al.* 2014a,b), as a dehydration aid for the sludge (Tanifuji *et al.* 2022; Yamasaki and Shigemura 2017, 2022), and as an alternative auxiliary fuel during the incineration process of the sludge (Miyamoto *et al.* 2021, 2022). However, in some cases, this plant biomass may not be suitable for use at WWTPs due to transportation costs and restrictions on the sites and facilities of sewage treatment plants. Some types of plant biomass, such as grass clippings, and pruned branches produced by the maintenance of rivers, roads, and parks, have been used as compost feedstock. Others have been disposed of by incineration or natural decomposition instead of being used effectively (NILIM 2015). In this case, the carbon dioxide (CO_2) absorbed and fixed by the plant biomass during its growth is considered to be released into the atmosphere (Prentice *et al.* 2001).

Regarding efforts to reduce CO_2 in the atmosphere, the storage of carbon in soils in agricultural land by applying biochar obtained by the carbonization of plant biomass is being considered. A method to estimate the impact of biochar on soil carbon stocks in mineral soils for croplands and grasslands has been refined in the "2019 Refinements to the 2006 IPCC National Greenhouse Gas Inventory Guidelines," which were adopted and accepted during the 49th Session of the Intergovernmental Panel on Climate Change (IPCC) in May 2019. Biochar is defined as a solid material generated by heating biomass to a temperature in excess of 350 °C under conditions of controlled and limited oxidant concentrations to prevent combustion (IPCC 2019). However, carbonization would require an input of energy from the outside, so there is a need to clarify appropriate carbonization conditions for various plants in order to produce biochar to store carbon efficiently.

During the thermal decomposition of wood, Kuriyama (1979) reported that hemicellulose was mainly decomposed at temperatures below 260 °C, cellulose was mainly decomposed at 260 to 310 °C, and lignin was decomposed at 310 to 400 °C. In addition, Kijima (2012) described that the carbonization yield of most wood is 40% or less at temperatures below 400 °C. Many studies have gathered knowledge about the thermal decomposition behavior of wood. However, there are few reports on the thermal decomposition behavior of herbaceous plants compared with that of wood. In addition, herbaceous plants with low lignin content should have lower carbon content than wood, so there is a concern that the carbonization yield will decrease when herbaceous plants are used as biochar for carbon storage.

When the Klason method is applied to estimate the lignin content of herbaceous plants and tree leaves, the quantitative value of lignin is overestimated due to other components such as proteins (Jin *et al.* 2003). This is one of the factors impeding elucidation of the thermal decomposition behavior of herbaceous plants. Toda *et al.* (2015) studied pre-extraction treatments of tree leaf samples using a variety of solvents to determine the lignin content of tree leaves; however, the potential overestimation of lignin content in leaves by the Klason method cannot be prevented by applying pre-extraction treatment of leaves. To determine lignin in forage samples, the acid detergent (AD) treatment is often applied before 72% sulfuric acid hydrolysis, and the obtained residue (AD lignin) is considered to reflect the amount of lignin (Van Soest 1963). The hemicellulose in the biomass is liberated during AD treatment, so the amount of carbohydrates in the filtrate obtained by 72% sulfuric acid hydrolysis after AD treatment may not reflect the total carbohydrate in the samples. Iwasaki (1960) studied the pre-treatment of oat (*Avena sativa*) for lignin estimation using 72% sulfuric acid; the lignin content of oat pretreated with 5% sulfuric acid was 3.7 to 8.0%, which was similar to that treated by the method of Ellis *et al.* (1946) and was lower than that of untreated samples (16.8 to 18.8%). This study focused on the treatment of herbaceous plants with 5% sulfuric acid as a pre-treatment for lignin determination by the Klason method, with the hypothesis that it would be possible to measure the lignin content accurately while measuring the carbohydrate content in the sample by analyzing the filtrate obtained by treatment with 5% sulfuric acid.

The goal of this study was to use plant biomass as biochar for storing carbon in soils. To clarify the chemical composition of herbaceous plants, pre-treatment for estimating lignin content by the Klason method was studied. Thermogravimetric (TG) analysis of plants was performed, and the relationship between the yield at each temperature and the chemical composition of plants was studied. Based on the obtained results, the possibility of using herbaceous plants as raw materials for producing biochar for storing carbon was studied.

EXPERIMENTAL

Materials

Japanese cypress (*Chamaecyparis obtusa*) wood chips, moso-bamboo (*Phyllostachys pubescens*), rice straw (*Oryza sativa*), dokudami (*Houttuynia cordata*), halcyon (*Erigeron philadelphicus*), mugwort (*Artemisia* Spp.), and a grass mixture were used as samples. Japanese cypress wood chips and rice straw were obtained from markets in Japan. Moso-bamboo was collected from a park in Japan, and its nodes were removed before use in the experiments. The wild plants of dokudami, mugwort, halcyon, and grass mixture, naturally occurring in Tsukuba, the Public Work Research Institute, Japan, were collected. The petal parts of dokudami and halcyon were also removed before use in the experiments. Mugwort was divided into leaf parts and stem parts, and its whole, and its leaf, and stem parts were used for the experiments. The samples were crushed in a blender mill. Japanese cypress, moso-bamboo, rice straw, and grass mixtures were passed through a 40-mesh sieve used for the experiments. Dokudami, halcyon, and mugwort were used in the experiments without passing through a sieve because they became flocculent upon crushing. The moisture content and ash content of samples were determined from the weights upon drying at 110 °C and ashing at 600 °C to a constant weight, respectively. Filter paper (Whatman No. 1), starch (Fujifilm Wako Pure Chemical), and Klason lignin residue (prepared in accordance with the procedure from a non-extracted Japanese cypress) were subjected to TG analysis as model samples for cellulose, other carbohydrates, and lignin, respectively.

Hot Water Extraction

The sample (1 g oven-dried weight) was added to a 50 mL centrifuge tube with a stopper, 30 mL of distilled water was added to the same tube, and the mixture was heated in a boiling water bath for 1.5 h with occasional stirring. After that, centrifugation (15,000 × *g*, 20 min) was carried out to separate the residue and the supernatant, and the supernatant was collected in another bottle. Thirty milliliters of distilled water was added again to the remaining sample (residue), and the same operation was performed. The hot water extraction was repeated three times in total. The all of supernatants were recovered and dried in a porcelain dish, and the ratio of the obtained weight on the porcelain dish per oven-dried sample weight was defined as the content of hot water extractives.

Organic Solvent Extraction

The extraction with organic solvent was performed using the lipid determination method of sewage sludge and micro algae, which has been reported from the authors' laboratory (Tanifuji *et al.* 2021). After hot water extraction, the sample (0.5 g oven-dried weight) and 10 mL of ethyl acetate/ethanol mixture (1:1 v/v) were added to a 50 mL test tube with a stopper and heated to 60 °C for 2.5 h in a water bath with occasional stirring using a vortex mixer. After heating, the test tube was air-cooled to room temperature; then, 10 mL of n-hexane was added and stirred thoroughly. Furthermore, 10 mL of distilled water was added and the mixture was stirred well. Next, the mixture was allowed to stand until it separated into two layers. The upper organic solvent layer was collected from it and transferred to an eggplant-shaped flask. The lower aqueous layer with the remaining sample was further washed twice with 10 mL of n-hexane, and this organic solvent layer was also collected in the same eggplant-shaped flask. The collected organic solvent was dried under reduced pressure at 60 °C in a rotary evaporator, and the content of ethyl acetate/ethanol extractives was determined as a

percentage of the obtained weight per oven-dried untreated sample weight. In this study, the total amount of hot water extractives and the ethyl acetate/ethanol extractives was evaluated as the extractive content. For Japanese cypress, the sample before hot water extraction was also extracted with ethyl acetate/ethanol mixture in the same way. Japanese cypress (0.5 g oven-dried weight) was also subjected to Soxhlet extraction using acetone and n-hexane for 8 h (refluxing for 15 to 20 min each time, with a solvent volume of approximately 50 mL each time), and the amounts of extractives obtained by each method were compared.

Pre-treatment of Herbaceous Plants for Klason Lignin Analysis

Sulfuric acid (5%) treatment and AD treatment were conducted as pre-treatments for lignin analysis by the Klason method. In this study, 2 g of cetyltrimethylammonium bromide dissolved in 100 mL of 5% sulfuric acid was used as an AD solution. Five milliliters of sulfuric acid (5%) or AD solution was added to the sample (0.15 g oven-dried weight) after successive extraction with hot water and ethyl acetate/ethanol mixture and treated in an autoclave at 105 °C for 1 h. In the case of treatment with 5% sulfuric acid, after autoclaving, the mixture was filtered through a glass filter paper (Whatman 1 GF/B), and then washed with hot water. The obtained filtrate was adjusted to 100 mL. One milliliter of the diluted filtrate was mixed with 1 mL of 7.5% sulfuric acid (the concentration of sulfuric acid in the solution was about 4%), and treated at 121 °C for 1 h. Carbohydrates liberated by the treatment with 5% sulfuric acid should be hydrolyzed to monosaccharides by this treatment. The carbohydrate content was measured by the phenol-sulfuric acid method in accordance with the following procedure. In the case of the AD treatment, after filtering, and washing in the same manner, the residue was also washed with cold acetone and the weight of the collected residue was measured.

Measurements of Lignin and Carbohydrates

Primary hydrolysis of 50 to 100 mg oven dried sample was performed using 3 mL of 72% sulfuric acid for 2.5 h at room temperature. Then, secondary hydrolysis of the sample was conducted with 4% sulfuric acid by adding distilled water to the mixture for 1 h at 121 °C (Ohi *et al.* 1997). This mixture was separated by filtration using a glass filter (Whatman 1 GF/B), and the acid-insoluble residue was washed with hot distilled water. The obtained acid-insoluble residue was dried at 110 °C and weighed. Furthermore, the acid-insoluble residue was ashed at 600 °C for 1 h, and the ash content in the residue was determined. The lignin content was defined as the percentage of the weight of the residue after removing the ash weight per the oven-dried untreated sample weight.

The volume of the filtrate obtained by secondary hydrolysis was adjusted to 500 mL, and the carbohydrate content was measured by the phenol-sulfuric acid method (Nielsen 2010). Here, 0.5 mL of 5% (w/v) phenol aqueous solution was added to 0.5 mL of the appropriately diluted filtrate, and the mixture was well stirred. Next, 2.5 mL of concentrated sulfuric acid was added to this mixture to cause a vigorous reaction, and the mixture was allowed to stand for 20 min or longer to develop a color. After that, the absorbance at 490 nm was measured. Aqueous glucose solutions were used as the standards for calibration, and the carbohydrate content was determined in the form of glucan, which is a polymer of glucose.

TG Analysis

TG/DTA 6300 (Seiko Instruments Inc., Chiba, Japan) was used for the experiments. Approximately 10 mg of the sample was placed in a platinum pan and

subjected to thermogravimetric (TG) analysis at a heating rate of 10 °C/min under a nitrogen stream (flow rate of 50 mL/min). For a blank, air was used. The yield obtained by TG analysis was defined as the percentage of the value subtracting ash weight from the weight reading at each temperature per the value subtracting ash weight from the weight reading at the first point at which a temperature of 150 °C was exceeded. It was assumed that ash weight did not change during TG analysis and subtracting ash weight from the remaining weight was considered as the volatile matter-based yield.

The change in the rate of weight loss from the differential thermogravimetric (DTG) curve was examined. In the DTG curve, the value obtained by subtracting the ash weight from the weight at the first point at which a temperature of 150 °C was exceeded was used as the standard. Each point was divided by this value and expressed as the percentage change per minute (%/min) against the standard weight. The maximum weight loss rate and the temperature were read from the obtained DTG curve.

RESULTS AND DISCUSSION

Comparison of Ethyl Acetate/Ethanol Mixture Extraction with Acetone and n-Hexane Soxhlet Extraction

Table 1 shows a comparison of the extractive amounts between the ethyl acetate/ethanol mixture extraction and Soxhlet extraction with acetone and n-hexane. The results of the previously reported ethyl acetate/ethanol mixture extraction of sewage sludge and micro algae (Tanifuji *et al.* 2021) are also presented.

Ethyl acetate/ethanol mixture extraction has been reported by Lu *et al*. (2015) as a method to recover lipids from micro algae. Based on this report, the temperature and time for ethyl acetate/ethanol mixture extraction were changed to suitable conditions (60 °C and 2.5 h) for the analysis of primary sewage sludge and micro algae that were cultured using nutrients in the influent of municipal WWTP. The amount of extractives from the primary sewage sludge with the ethyl acetate/ethanol mixture was similar to that obtained by the Brigh-Dyer method (chloroform/methanol 1:1 v/v) and n-hexane Soxhlet extraction (Table 1 and Tanifuji *et al*. 2021).

In this study, ethyl acetate/ethanol mixture extraction method was applied to Japanese cypress. However, the amount of extractives from Japanese cypress obtained using the ethyl acetate/ethanol mixture was 1.2%, which was lower than that by acetone Soxhlet extraction (3.4%) but higher than that by n-hexane Soxhlet extraction (0.3%). The result of acetone extraction was similar to that of ethanol/benzene extraction (2.7%), as reported in the literature (Fukushima 2010). Normally, ethanol/benzene mixture is used to measure the extractive content of wood. However, since benzene is carcinogenic, extraction using alternative solvents that are less harmful to the human body and the environment has been considered (TAPPI Proposed Revision of T204 cm-97 2007). Yokoyama *et al.* (2002) reported that the amount of extractives from loblolly pine obtained by acetone was similar to that by ethanol/benzene. In TAPPI Proposed Revision of T204 cm-97 (2007), a method of acetone extraction is also described. In this study, ethyl acetate/ethanol mixture extraction was conducted by recovering the organic layer by adding n-hexane and water to induce separation into two layers after extraction. The amount of extractives from Japanese cypress by n-hexane Soxhlet extraction was lower than that by other solvents. One of the possible reasons for the low amount of extractives by ethyl acetate/ethanol mixture extraction is that, at this stage, many of the extracted components remained in the aqueous layer instead of the organic layer.

To solve this problem, hot water extraction was applied before ethyl acetate/ethanol mixture extraction. The amount of hot water extractives was 2.7%, and the total amount of extractives obtained using the hot water-ethyl acetate/ethanol mixture was 3.0%, which was similar to that by acetone Soxhlet extraction. The acetone Soxhlet extraction requires more solvent than hot water-ethyl acetate/ethanol mixture extraction and has a large environmental impact. Therefore, in this study, the total amount of hot water-ethyl acetate/ethanol mixture extraction was evaluated as the extractives that showed a value similar to that with the conventional extraction method with acetone.

Table 1. Comparison of Amounts of Extractives Using Various Solvents

	EtAC/EtOH [c]	n-hexane	Acetone	Hot water	Hot water-EtAc/EtOH [c]	Brigh-Dyer [d]
	(% of oven-dried sample weight)					
Japanese cypress	1.2	0.3	3.4	2.7	3.0	- [e]
Primary sludge [a]	17.0	14.2	- [e]	- [e]	- [e]	14.6
Micro algae [b]	11.8	- [e]	- [e]	- [e]	- [e]	13.1

[a] Primary sludge obtained from the WWTP of a city; these data were reported by Tanifuji *et al.* (2021). [b] Micro algae cultivated using the influent of municipal WWPT; these data were reported by Tanifuji *et al*. (2021). [c] Mixture of ethyl acetate/ethanol (1:1 v/v). [d] Solvent using a mixture of chloroform/methanol (2:1 v/v). [e] Unmeasured.

Effects of Sulfuric Acid and AD Treatments as Pre-treatments for the Klason Lignin Analysis

Figure 1 shows the lignin content estimated for the samples subjected to the Klason method after extraction with a mixture of hot water-ethyl acetate/ethanol. Japanese cypress showed the highest Klason lignin content (32.5%) among the plant samples, which was similar to the value reported in the literature (29.0%, Fukushima 2010). Meanwhile, among the samples used in this study, it was considered that for herbaceous plants (moso-bamboo, rice straw, dokudami, halcyon, mugwort, and grass mixture) only lignin was not properly evaluated, even after successive extraction with hot water and ethyl acetate/ethanol mixture. This may have been because it has been suggested that the lignin content as determined by the Klason method is affected by other components in plant leaves and herbaceous plants (Jin *et al.* 2003). Toda *et al.* (2015) evaluated the effect of pre-extraction treatments on lignin determination in ginko and zelkova leaves via several different pre-extraction treatments. However, they concluded that the potential overestimation of lignin content in leaves by the Klason method cannot be prevented by subjecting leaves to pre-extraction treatments.

Iwasaki (1960) conducted pre-treatment with 5% sulfuric acid for estimating the lignin level in oat. He reported that the lignin content estimated upon 5% sulfuric acid pre-treatment was similar to that estimated by the method of Ellis *et al.* (1946). Therefore, in this study, treatment with 5% sulfuric acid was applied before the estimation of lignin content by the Klason method for herbaceous plant samples. However, the estimated values of all samples tended to increase upon treatment with 5% sulfuric acid compared with Klason lignin content without pre-treatment (Fig. 1). Iwasaki (1960) washed the residues with hot water and cold acetone after 5% sulfuric acid treatment. However, in this study, washing with cold acetone was omitted for the purpose of measuring the amount of carbohydrates in the filtrate. The increase in the lignin content estimated upon pre-treatment with 5% sulfuric acid might have been caused by this difference; however, it was also indicated that the removal of carbohydrates by treatment with 5% sulfuric acid had less of an effect on the analysis

of lignin content by the Klason method.

Therefore, AD treatment was studied as a pre-treatment for the Klason method by using a surfactant (2% w/v cetyltrimethylammonium bromide) added to 5% sulfuric acid and washing of the obtained residue with hot water and cold acetone. Among the herbaceous plants used in this study, AD treatment reduced the estimated lignin content of moso-bamboo, rice straw, halcyon, mugwort (whole and stem parts), and grass mixtures, which included numerous stem parts, compared with the Klason method without pre-treatment. In addition, AD treatment had only a slight effect on the estimated lignin content of Japanese cypress (33.7%) compared with the Klason method. The lignin content estimated by AD treatment (AD lignin content) can be applied to wood and its content appeared to be reasonable. However, the lignin contents estimated by the Klason method without pre-treatment and by AD treatment were almost the same for dokudami and mugwort leaf parts. There still remains a question of whether these estimated lignin contents were reasonable and reflected the true amount of lignin in plant leaves. AD treatment is often applied for the determination of lignin in forage samples (Jin *et al.* 2003); however, there is no definitive method to accurately determine the quantity of lignin in leaves and herbaceous plants (Toda *et al.* 2015). In this study, applying lignin estimation by AD treatment to wood did not differ from that by the Klason method without pre-treatment, and at least for herbaceous plants with a lot of stem parts, the lignin content estimated by AD treatment was lower than that by the Klason method. In this study, the AD lignin content was used as an indicator of lignin.

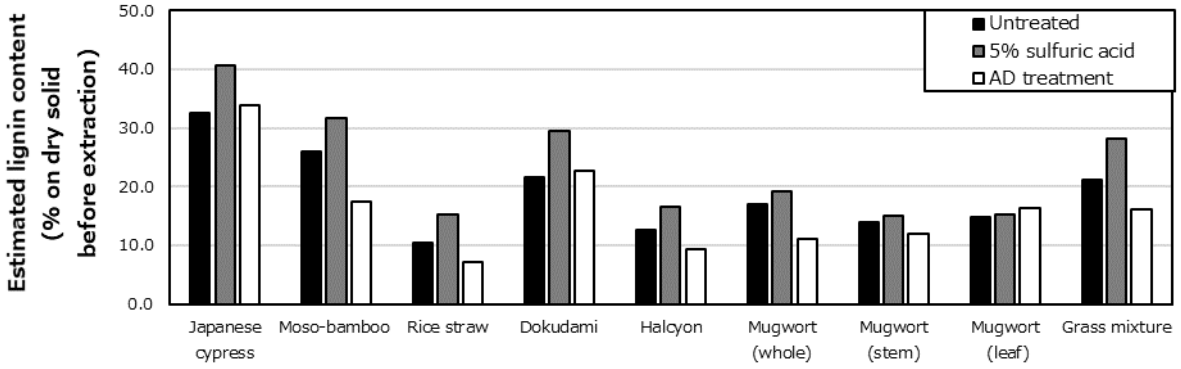

Fig. 1. Comparison of lignin content estimated by the Klason method between untreated and pretreated plant samples

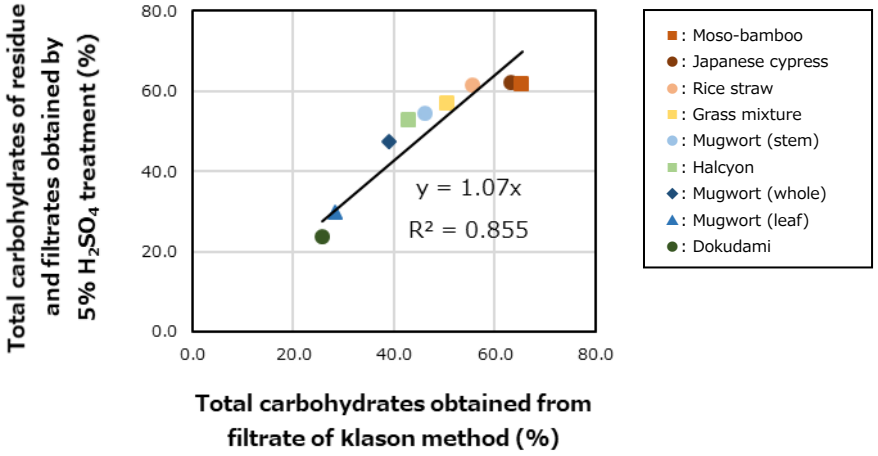

Fig. 2. Comparison of carbohydrates estimated by the phenol-sulfuric acid method between filtrate obtained by the Klason method, and residue and filtrate obtained by 5% sulfuric acid treatment

Table 2. Chemical Compositions and Carbonization Yields of Plant Samples

	Content (% relative to oven-dried sample weight)							Yield (% as volatile matter)			
	Extractives	AD lignin	Carbohydrates			Ash	Total	250 °C	300 °C	350 °C	400 °C
			Liberated after 5% H_2SO_4 treatment	Remaining after 5% H_2SO_4 treatment	Total carbohydrates						
Japanese cypress	3.0	33.7	10.9	52.1	63.0	0.3	100.0	97.2	86.2	56.5	34.2
Moso-bamboo	6.6	17.5	19.1	46.3	65.4	1.7	91.2	94.8	77.6	42.7	38.0
Rice straw	17.9	7.1	22.7	32.9	55.6	8.9	89.4	92.9	76.5	44.2	35.5
Dokudami	37.2	22.2	5.4	19.8	25.2	15.4	100.0	85.3	64.3	49.9	43.4
Halcyon	27.2	9.4	14.0	29.0	43.0	7.9	87.5	89.2	69.4	41.8	36.3
Mugwort (whole)	26.5	11.2	14.6	24.6	39.2	7.4	84.2	89.0	67.1	43.6	38.0
Mugwort (stem)	24.9	12.1	11.9	34.5	46.3	6.3	89.6	88.4	66.8	42.1	37.2
Mugwort (leaf)	34.4	16.5	7.8	20.7	28.5	10.1	89.5	88.3	68.8	49.4	42.7
Grass mixture	14.9	16.1	23.6	27.1	50.7	3.6	85.2	95.2	79.7	54.1	41.3

The amounts of carbohydrates were estimated by the phenol-sulfuric acid method and compared between the filtrate obtained by the Klason method and the total residue and filtrate obtained by 5% sulfuric acid treatment. The results are shown in Fig. 2. They produced approximately the same values. Based on these results, the amount of carbohydrates liberated by 5% sulfuric acid could be evaluated as the carbohydrates in an amorphous state and easily decomposed by acid, such as hemicellulose, and the carbohydrates that remaining upon treatment with 5% sulfuric acid could be evaluated as the amount of carbohydrates with high crystallinity and resistance to acid hydrolysis such as cellulose.

Chemical Composition of Plants

Table 2 shows the chemical composition of the wood and herbaceous plant samples. The extractive content of Japanese cypress was 3.0%, which was lower than for the other herbaceous plants. The extractive contents of dokudami and mugwort leaf parts were 37.2 and 34.4%, respectively, and the extractive content tended to be high when the sample included a lot of leaf parts. The content of carbohydrates of Japanese cypress liberated by 5% sulfuric acid was 10.9%, which was lower than those of moso-bamboo (19.1%) and rice straw (22.7%). However, the contents of carbohydrates of dokudami (5.4%) and mugwort leaf parts (7.8%) liberated by 5% sulfuric acid were even lower than that of these plants. In addition, the content of carbohydrates remaining upon treatment of Japanese cypress (52.1%) with 5% sulfuric acid was the highest among samples, and those of moso-bamboo (46.3%), rice straw (32.9%), and mugwort stem (34.5%) were relatively high (exceeding 30%) among herbaceous plants. It was indicated that samples including a lot of stem parts should contain a large amount of cellulose.

Effect of the Chemical Composition of Plants on the Yield from TG Analysis

Figure 3 shows the TG curve and Table 2 also shows the carbonization yield at each temperature. The TG curve of filter paper as a model sample of cellulose showed rapid weight loss at temperatures around 350 °C. Crystalline cellulose is known to undergo rapid weight loss at temperatures around 350 °C during TG analysis (Kawamoto 2015), and a similar tendency was observed in this study. Starch started to decompose at a lower temperature than that of filter paper (around 250 °C), and the yield at 400 °C (26.9%) was higher than that of filter paper (8.5%). Furthermore, the Klason lignin residue obtained from Japanese cypress did not show rapid weight loss compared with filter paper and starch, and the yield at 400 °C was the highest (61.9%) among samples used in this study.

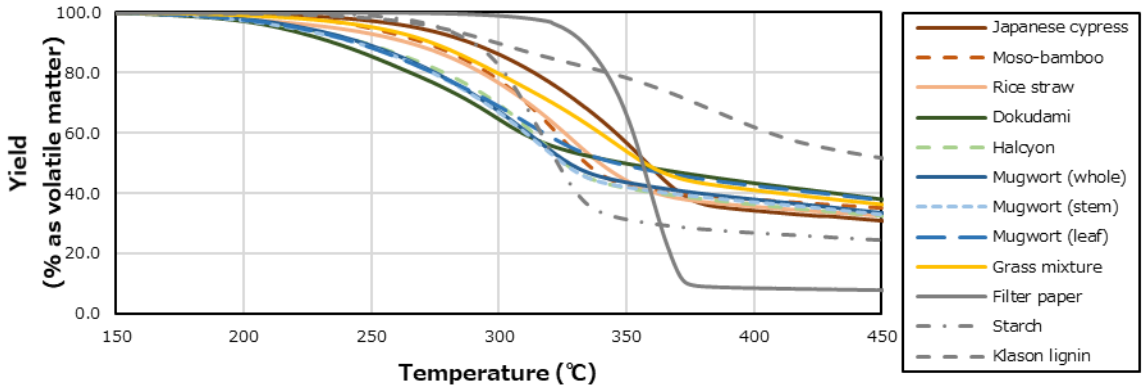

Fig. 3. TG curves of plants, carbohydrates, and lignin model samples

The yield at 400 °C of all plant samples was 34.2 to 43.4%, which was similar to that in a previous report that mentioned that carbonization yields at temperatures below 400 °C are around 40% or less for wood (Kijima 2012). In addition, it was expected that Japanese cypress with high AD lignin content would show a high yield because lignin has a higher carbon ratio in the molecule than carbohydrates. However, the yield of Japanese cypress at 400 °C was 34.2%, which was lower than that of other plant samples. Under these conditions, no increment in the yield of Japanese cypress with high AD lignin content was observed.

Linear regression analysis was performed on the nine plant samples, and the relationship between the chemical composition of the samples (horizontal axis) and the yield obtained by TG analysis (vertical axis) was investigated. The results are shown in Fig. 4, and the slope, intercept, and coefficient of determination (R^2) are shown in Table 3. The highest R^2 of the yields at 250 and 300 °C were obtained when regression analysis was performed with extractives (0.91 and 0.84, respectively), and the yields at 250 and 300 °C tended to decrease with increasing extractive content of plants. The highest R^2 of the yield at 350 °C was obtained with AD lignin (0.60), and the highest R^2 of the yield at 400 °C was obtained with carbohydrates remaining upon treatment with 5% sulfuric acid (0.54), when linear regression was performed. The yield at 350 °C tended to increase with increasing AD lignin content, and that at 400 °C tended to decrease with increasing content of carbohydrates remaining upon treatment of plants with 5% sulfuric acid.

Kuriyama (1979) has reported that hemicellulose was mainly decomposed at less than 260 °C, cellulose was mainly decomposed at 260 to 310 °C, and lignin was decomposed at 310 to 400 °C during carbonization. It was thought that the decomposition of the extractives, which are unstable components, started at 250 to 300 °C, and lignin should contribute to maintaining the high yield at 350 °C by containing a large amount of carbon. Furthermore, the yields at 400 °C tended to decrease with increasing content of carbohydrates in plants remaining upon treatment with 5% sulfuric acid because the crystalline carbohydrates such as cellulose in plants should decompose significantly during TG analysis.

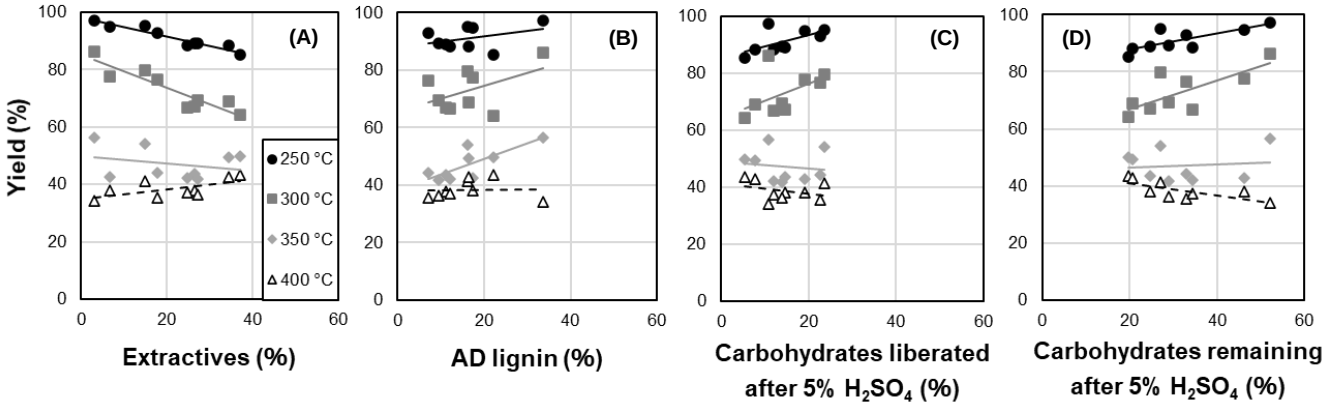

Fig. 4. Relationships between chemical composition of samples and the carbonization yield obtained by TG analysis. Notes: A, extractives; B, AD lignin; C, carbohydrates liberated by 5% sulfuric acid treatment; and D, carbohydrates remaining upon treatment with 5% sulfuric acid.

Table 3. Slopes, Intercepts, and Coefficients of Determination Obtained from Regression Curves for Yields and Chemical Compositions

TG (°C)	Extractives			AD lignin			Carbohydrates liberated after 5% H_2SO_4 treatment			Carbohydrates remaining after 5% H_2SO_4 treatment		
	S	I	R^2	S	I	R^2	S	I	R^2	S	I	R^2
250	-0.33	98	0.91	0.18	88	0.13	0.40	85	0.39	0.28	82	0.60
300	-0.57	85	0.84	0.45	66	0.24	0.59	64	0.26	0.50	57	0.58
350	-0.12	50	0.07	0.53	39	0.60	-0.12	49	0.02	0.06	45	0.01
400	0.17	35	0.40	0.01	38	0.00	-0.17	41	0.11	-0.21	45	0.54
S, slope; I, intercept; and R^2, coefficients of determination.												

Effect of the Chemical Composition of Plants on the Maximum Weight Loss Rate

Figure 5 shows the DTG curves of each sample. DTG analysis is useful for obtaining information on the temperature at which the thermogravimetric weight loss rate is maximal. Table 4 shows the maximum weight loss rate and the temperature at that rate as per Fig. 5. The maximum weight loss rate (31.4%/min) of the filter paper was higher than for the other samples, and the temperature at which that rate occurred was 353 °C. Starch showed a peak at a lower temperature (319 °C) than filter paper, and the maximum weight loss rate was 16.4%/min. Kim *et al.* (2010) conducted a TG analysis of three types of cellulose with different crystallite sizes (*Halocynthia* > cotton > microcrystalline cellulose) and found that the thermal decomposition of cellulose shifted to higher temperatures with increasing crystallite size (389, 366, and 341°C, respectively). The filter paper used in this study showed a temperature at the maximum weight loss rate similar to that of cotton reported by Kim *et al.* (2010), and the starch showed a lower temperature than filter paper and celluloses, as reported by Kim *et al.* (2010). Klason lignin residue showed two peaks at 303 and 385 °C, but the maximum weight loss rate was 3.7%/min (at 385 °C), which was lower than for other samples. This suggests that lignin did not cause rapid thermal decomposition compared with carbohydrates.

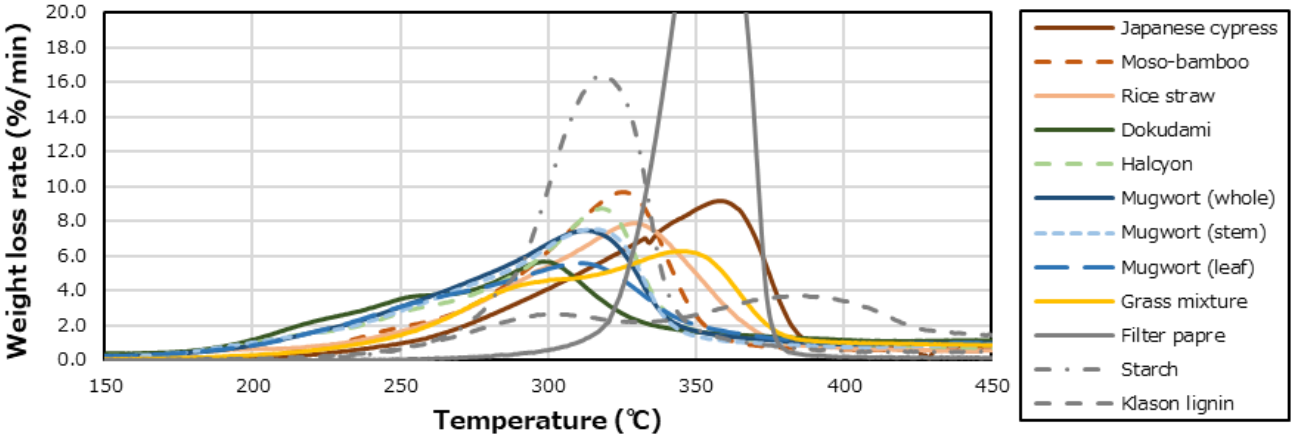

Fig. 5. DTG curves of plants and carbohydrates and lignin model samples

For plant samples, Japanese cypress, and grass mixture showed the maximum weight loss rates at 359 and 345 °C, respectively, which were similar to the temperature of the filter paper. The other herbaceous samples showed the maximum weight loss rate at 298 to 329 °C, which was similar to the temperature of starch. Among the plant samples, the maximum weight loss rates of Japanese cypress and moso-bamboo were 9.1 and 9.7%/min, respectively, which were higher than those of other herbaceous plants, but lower than those of filter paper and starch.

Table 4. Maximum Weight Loss Rate and Temperature at which that Rate Occurred

	Japanese cypress	Moso-bamboo	Rice straw	Dokudami	Halcyon	Mugwort (whole)	Mugwort (stem)	Mugwort (leaf)	Grass mixture
M [a] (%/min)	9.1	9.7	8.0	5.6	8.7	7.4	7.6	5.6	6.3
T [b] (°C)	359	325	329	298	318	314	315	312	345

[a] maximum weight loss rate. [b] temperature at maximum weight loss rate.

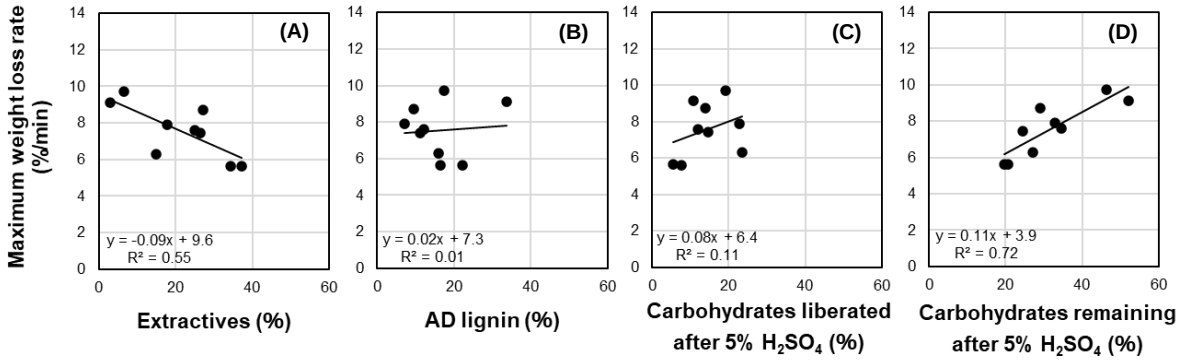

Fig. 6. Relationships between c hemical composition of samples and the maximum weight loss rate. Notes: A, extractives; B, AD lignin; C, carbohydrates liberated by 5% sulfuric acid treatment; and D, carbohydrates remaining upon treatment with 5% sulfuric acid

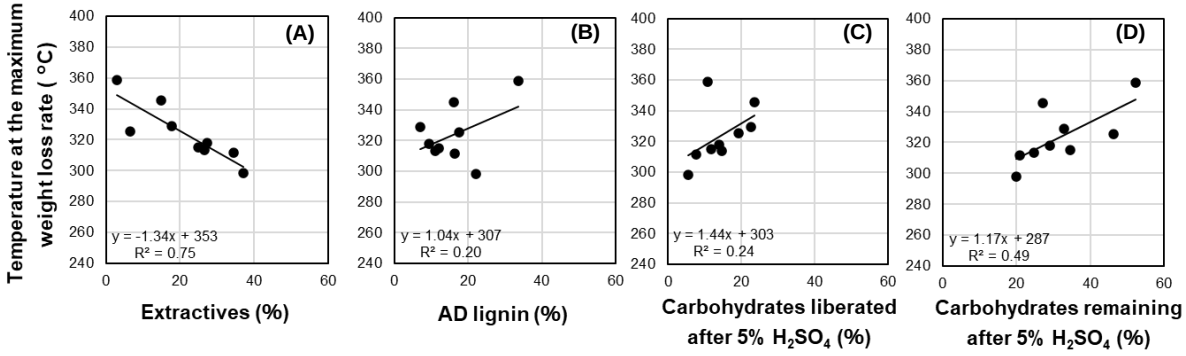

Fig. 7. Relationships between chemical composition of samples and temperature at which the maximum weight loss rate occurred. Notes: A, extractives; B, AD lignin; C, carbohydrates liberated by 5% sulfuric acid treatment; and D, carbohydrates remaining upon treatment with 5% sulfuric acid

To investigate the effects of the chemical composition of plants on the maximum weight loss rate and the temperature at which that rate occurred, linear regression analysis was performed. The results are shown in Fig. 6 (maximum weight loss rate) and Fig. 7 (temperature). The highest R^2 of the maximum weight loss rate was obtained when regression analysis was performed with carbohydrates remaining upon treatment with 5% sulfuric acid (0.72). The highest R^2 of the temperature at which the maximum weight loss rate occurred was obtained when regression analysis was performed with extractives (0.75). A high content of carbohydrates remaining upon treatment with 5% sulfuric acid showed more rapid thermal decomposition because crystalline cellulose would decompose rapidly during TG analysis, as shown in the results of the filter paper (Fig. 5). The temperature at that rate was shifted to a lower level because the decomposition of the extractives, which are unstable components, would begin at a low temperature and the temperature at which the maximum weight loss rate occurred would also be affected by unstable components.

Potential of Herbaceous Plants as a Feedstock of Biochar

As shown in Table 2, the extractive content of herbaceous plant samples such as dokudami, halcyon, and mugwort was higher than that of Japanese cypress, moso-bamboo, and rice straw. In addition, the carbohydrate content of these herbaceous plant samples remaining upon treatment with 5% sulfuric acid was lower than those of Japanese cypress, moso-bamboo, and rice straw. From the results of TG analysis, the yield at 250 to 300 °C of these herbaceous plant samples was lower than that of Japanese cypress, moso-bamboo, and rice straw (Table 2 and Fig. 3). The yield of these herbaceous samples at 400 °C was higher than those of Japanese cypress and rice straw (Table 2 and Fig. 3). At present, the carbonization temperature should be in excess of 350 °C in the production of biochar for carbon storage in the soil derived from wood and herbaceous plants, as described in the 2019 Refinement to the 2006 IPCC Guidelines (IPCC 2019). However, for herbaceous plants such as dokudami, halcyon, and mugwort that have high extractive contents, it is recommended to lower the thermal decomposition temperature to below 350 °C. In addition, these herbaceous plants had a low crystalline carbohydrate content, such as cellulose (carbohydrates remaining upon treatment with 5% sulfuric acid). Therefore, relatively high yields can be achieved even if the carbonization temperature is above 350 °C. Herbaceous plants are considered to be promising resources as biochar raw materials for producing biochar for storing carbon in soils.

CONCLUSIONS

1. Acid detergent (AD) treatment appears to be a suitable pre-treatment prior to estimating the lignin content of herbaceous plants. However, the question remained of whether these estimated lignin contents reasonably reflect the true amount of lignin in plant leaves.
2. The amount of carbohydrates liberated by 5% sulfuric acid could be evaluated as those with an amorphous state, such as hemicellulose. The amout of carbohydrates remaining upon treatment with 5% sulfuric acid could be evaluated as those with high crystallinity, such as cellulose. However, the removal of carbohydrates by 5% sulfuric acid treatment had a smaller effect on the analysis of lignin content by the Klason method.

3. The yields at 250 and 300 °C obtained by TG analysis tended to decrease with increasing extractives content. The yield at 350 °C tended to increase with increasing AD lignin content and that at 400 °C tended to decrease with increasing crystalline carbohydrate content of plants, such as cellulose.
4. Herbaceous plants such as dokudami, halcyon, and mugwort that have high extractives contents. In such cases, it is recommended to lower the thermal decomposition temperature to below 350 °C. In addition, these herbaceous plants had a low crystalline carbohydrate content, such as cellulose. Therefore, relatively high yields can be achieved even if the carbonization temperature is above 350 °C. Herbaceous plants are considered to be feasible resources as raw materials for biochar production for storing carbon in soils.

REFERENCES CITED

Ellis, G. H., Matrone, G. and Maynard, L. A. (1946). "A 72 percent H_2SO_4 method for the determination of lignin and its use in animal nutrition studies," *Journal of Animal Science* 5 (3), 285-297. DOI: 10.2527/jas1946.53285x

Fukushima, K. (2010). "Mokuzai no soshiki (chapter 1, in Japanese)." in: *Mokushitsu no kagaku*, The Japan wood research society (ed.), Buneido, Tokyo, Japan, pp. 3.

Hidaka, T., Arai, S., Okamoto, S. and Uchida, T. (2013). "Anaerobic co-digestion of sewage sludge with shredded grass from public green spaces," *Bioresource Technology* 130, 667-672. DOI: 10.1016/j.biortech.2012.12.068

Intergovernmental Panel on Climate Change (IPCC) (2019). Method for estimating the change in mineral soil organic carbon stocks from biochar amendments: Basis for future methodological development (Appendix4), 2019, *Refinement to the 2006 IPCC guidelines for national greenhouse gas inventories*, Volume 4, (https://www.ipcc-nggip.iges.or.jp/public/2019rf/pdf/4_Volume4/19R_V4_Ch02_Ap4_Biochar.pdf), accessed 18 Jan 2022.

Iwasaki, T. (1960). "A study on the method for the determination of lignin in herbage (in Japanese)," *Journal of Japanese Society of Nutrition and Food Science* 13 (2), 95-99

Jin, Z., Akiyama, T., Chung, B. Y., Matsumoto, Y., Iiyama, K. and Watanabe, S. (2003). "Changes in lignin content of leaf litters during mulching," *Phytochemistry* 64 (5), 1023-1031. DOI: 10.1016/s0031-9422(03)00423-0

Kawamoto, H. (2015). "Reactions and molecular mechanisms of cellulose pyrolysis (in Japanese)," *Journal of the Japan Wood Research Society* 61(1), 1-24.

Kijima, M. (2012). "Wood biorefinery (in Japanese)," *Journal of the Society of Materials Science, Japan* 61(9), 803-809

Kim, U. J., Eom, S. H., and Wada, M. (2010). "Thermal decomposition of native cellulose: Influence on crystallite size," *Polymer Degradation and Stability* 95(5), 778-781. DOI: 10.1016/j.polymdegradstab.2010.02.009

Kuriyama, A. (1979). A study on the carbonization process of wood (Bulletin of the Forestry and Forest Products Research Institute No. 304, in Japanese), (https://www.ffpri.affrc.go.jp/labs/kanko/304-2.pdf), accessed 18 Jan 2022, FFPRI, Tsukuba, Japan

Lu, W., Wang, Z., and Yuan, Z. (2015). "Characteristics of lipid extraction from Chlorella sp. cultivated in outdoor raceway ponds with mixture of ethyl acetate and ethanol for

biodiesel production," *Bioresource Technology* 191, 433-437. DOI: 10.1016/j.biortech.2015.02.069

Miyamoto, T., Tanifuji, K., Omoto, T., and Shigemura, H. (2021). "Possibility of supplying vegetation biomass auxiliary fuel to a sewage sludge incinerator using a screenings co-firing line (in Japanese)," *Journal of EICA (The Society of Environmental Instrumentation, Control and Automation)* 26 (2·3), 60-67

Miyamoto, T., Tanifuji, K., Sakurai, K., Shigemura, H., Takaoka, M. and Okayasu, Y. (2022). "Basic study on the usage of combustible general waste as auxiliary fuel in sewage sludge incinerators (in Japanese)," *Journal of EICA (The Society of Environmental Instrumentation, Control and Automation)* 27 (2·3), 110-118

National Institute for Land and Infrastructure Management (NILIM) (2015). Technical note on the energy application method for plant waste materials derived from urban areas (Technical note of NILIM No. 845, in Japanese), (http://www.nilim.go.jp/lab/bcg/siryou/tnn/tnn0845pdf/ks0845.pdf), accessed 18 Jan 2022, NILIM, Tsukuba, Japan

Nielsen, S. S. (2010). "Phenol-sulfuric acid method for total carbohydrates," in: *Food analysis laboratory manual*, Nielsen, S. S. (ed.), Springer, Berlin, pp. 47-53. DOI: 10.1007/978-1-4419-1463-7_6

Ohi, H., Ju, Y., and Kuroda, K. (1997). "Structural analysis of lignin by pyrolysis-gas chromatography (VII) conditions for acid hydrolysis of wood pulps and characteristics of acid-insoluble residues," *Japan Tappi Journal* 5(10), 1578-1586

Prentice, I. C., Farquhar, G. D., Fasham, M. J. R., Goulden, M. L., Heimann, M., Jaramillo, V. J., Kheshgi, H. S., Le Quéré, C., Scholes, R. J., and Wallace, D. W. R. (2001). "The carbon cycle and atmospheric carbon dioxide," in: *Climate Change 2001: The Scientific Basis*, Houghton, J. T., Ding, Y., Griggs, D .J., Noguer, M., van der Linden, P. J., Dai, X., Maskell, K., and Johnson, C. A. (eds.), Cambridge University Press, New York, NY, pp. 188-189. ISBN: 0521807670

TAPPI proposed revision of T204 cm-97 (2007). "Solvent extractives of wood and pulp,", *Tappi*, Atlanta, GA, (https://www.tappi.org/content/sarg/t204.pdf), accessed 18 Jan 2022

Tanifuji, K., Miyamoto, T. and Shigemura, H. (2021). "Chloroform wo mochiinai gesuiodeichu no shishitsu no bunsekihouhou (in Japanese)" in: Proceedings of 58th Japan sewage works association annual technical conference, Osaka, Japan, pp. 386-388

Tanifuji, K., Yamasaki, Y., Miyamoto, T. and Shigemura, H. (2022). "Effect of structural features of plant biomass as a dewatering aid for digested sludge from a wastewater treatment plant," *BioResources* 17 (2), 2043-2052. DOI: 10.15376/biores.17.2.2043-2052

Toda, M., Akiyama, T., Yokoyama, T., and Matsumoto, Y. (2015). "Quantitative examination of pre-extraction treatment on the determination of lignin content in leaves," *BioResources* 10 (2), 2328-2337. DOI: 10.15376/biores.10.2.2328-2337

Van Soest, P. J. (1963). "Use of detergents in the analysis of fibrous feeds. I. Preparation of fiber residues of low nitrogen content," *Journal of the Association of Official Analytical Chemists* 73 (4), 487-491. DOI: 10.1093/jaoac/73.4.487

Wang, F., Hidaka, T., Sakurai, K. and Tsumori, J. (2014a). "Anaerobic co-digestion of steam-treated *Quercus serrata* chips and sewage sludge under mesophilic and thermophilic conditions," *Bioresource Technology* 166, 318-325. DOI: 10.1016/j.biortech.2014.05.057

Wang, F., Hidaka, T., and Tsumori, J. (2014b). "Enhancement of anaerobic digestion of shredded grass by co-digestion with sewage sludge and hyperthermophilic pretreatment," *Bioresource Technology* 169, 299-306. DOI: 10.1016/j.biortech.2014.06.053

Yamasaki, Y., and Shigemura, H. (2017). "Development of sludge reduction system with grass clippings from public spaces (in Japanese)," *Transaction of the Japan Society of Civil Engineers G (Environmental)* 73, III_365-III_373. DOI: 10.2208/jscejer.73.III_365

Yamasaki, Y., and Shigemura, H. (2022). "Environmental impact assessment of the nationwide introduction of plant biomass-mixed dehydration systems at wastewater treatment plants (in Japanese)," *Journal of the Japan Society of Material Cycles and Waste Management* 33, 54-68. DOI: 10.3985/jjsmcwm.33.54

Yokoyama, T., Kadla, J. F., and Chang, H. M. (2002). "Microanalytical method for the characterization of fiber components and morphology of woody plants," *Journal of Agricultural and Food Chemistry* 50 (5), 1040-1044. DOI: 10.1021/jf011173q

Article submitted: March 13, 2023; Peer review completed: April 29, 2023; Revised version received: May 24, 2023; Published: June 2, 2023.
DOI: 10.15376/biores.18.3.5057-5072