Supporting information to

Chemical Insight into the Base-Tuned Hydrothermal Treatment of Side Stream Biomasses

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Content

1. Material and methods

1.1. Hydrothermal synthesis

Then the mixture of 3.6 ml water and 1.2g biomass and different amounts of KOH (mass of KOH was calculated based on carbohydrate content (see fiber analysis section in supplementary material) in each biomass (Chestnut foliage: 27.5 wt%, Bark: 28.8 wt%, Bamboo: 80.0 wt% and Sugar beet: 40.2% carbohydrates + 24.8% sugars) and corresponding programmed KOH equivalent, *e.i.* VT150 means 1.50 eq. of KOH to carbohydrate content. All data are summarized in Table S1. Reaction mixture was transferred into 10 ml pressure vessels with removable PTFE cup from Parr (Moline, IL, USA). The hydrothermal process was carried out for 16h in the oven after reaching 220˚C. At the end of the reaction period, the autoclaves were taken out from the oven and remained in room temperature to be cooled down to below 100°C, and then were subsequently cooled down in water bath to around 26℃. After opening the pressure vessels, the slurry was transferred into centrifuge tubes and centrifuged (at 4500 rpm for 10min). Resulted solid and liquid fractions were analyzed after decantation.

1.2. Characterizations

1.2.1. *Carbon distribution. Carbon content in solid:* solid fraction was dried at 105 °C and then elemental analysis was performed. The total solid percentage (TS%) in solid samples was determined by treating samples at 105 °C for 24 h and organic total solid (oTS%) was measured by subtracting the TS105 from its ash content, which was achieved after treating the samples at 550°C for 5h (see Fig. 1). *Carbon content in liquid:* total carbon (TC_L) and total inorganic carbon (TIC_L) in liquid fraction measured by Shimadzu TOC-5050A Analyzer. The TOC_L (total organic carbon), was obtained by subtracting the TIC_L (total inorganic carbon) concentration from the TC_L.

1.2.2. *FTIR spectroscopy* was measured on the Nicolet iS5 with an iD5 ATR crystal, Thermo Fisher Scientific Inc. (Waltham, Massachusettes, USA), with a resolution of 0.5 cm^{-1}.

The FTIR spectra of the products of hydrothermal treatment at different base ratio are depicted in Fig. S2a. The broad absorption band at 3330–3400 cm^{-1} is caused by the stretching vibrations of the -OH groups, partially derived from adsorbed water. Hygroscopicity is higher when the number of oxygen-containing groups in the products increases, and correspondingly, water is contributing to the peak more. Here, excessive KOH suppresses acid-catalyzed condensation processes, resulting in higher abundance of oxygenated functional groups. This observation is in a good agreement with elemental analysis data that shows gradual increase of oxygen content with increase of KOH concentration. The peaks between 2800–2950 cm^{-1} are attributed to the aliphatic stretching vibrations of $-CH_3$ and $-CH_2-$ and more explicit when higher concentrations of KOH are used. In hydrothermal process, higher amount of base allows more aliphatic chains to be deliberated from the biomass structure. This is in a good agreement with NMR observations, which

also reveals considerably increased singlets at 1.23 and 1.05 ppm (see Fig. 2a) assigned to -CH_3 and -CH_2- from aliphatic chains. Another two peaks of particular interest are at 1311 and 1345 cm^{-1} and correspond to hydroxyphenolic groups. These peaks are not observed on FTIR spectra of products from hydrothermal processes with VT025 up to VT100. Normally, these groups are derived from hydrolyzed lignin surface compartments. When operating at such low KOH concentration, acid-catalyzed condensation consumes all these preformed groups. While at higher amounts of KOH (from VT150 to VT500) the reaction mixture avoids acidic conditions and these groups are not the part of acid-catalyzed condensation.

1.2.3. *^1H NMR.* ^{1}H NMR spectra were recorded on Agilent 400 MHz. Lyophilized mixture of liquid fractions for each run was systematically taken for ^{1}H NMR analyses. Then dissolved in d-DMSO at 1 wt%.

1.2.4. *HSQC.* HSQC NMR spectra were recorded on Agilent 600 MHz. To improve the structural knowledge of the humic acids synthesized through hydrothermal process, two-dimensional NMR was employed (see Fig. 5). The heteronuclear single quantum coherence (HSQC) experiment is a classical two-dimensional NMR technique that correlates the chemical shifts of protons (^{1}H nuclei) with their directly bonded ^{13}C nuclei, which produces peaks in the ^{1}H-^{13}C spectral space. Various methyl (-CH_3), methylene (-CH_2-) and methine (=CH-) groups are observed in the aliphatic region (δC/δH: 10 - 50 / 0.5 - 2.5 ppm). The general trend is similar to ^{1}H-NMR technique: more alkali deliberates more aliphatic chains presented in biomass. VT500 (Fig. 5b) shows higher intensities than VT150 (Fig. 5a) at this region. In addition, a phase sensitive mode employed for VT500 can even reveal -CH_3 and -CH against -CH_2- functional groups. Resonances from functionalized carbon chains (substituted with heteroelement-containing groups), as well as carbohydrate are found in the oxygenated aliphatic region (δC/δH: 35 - 110 / 2.5 - 5.5 ppm). This region shows abundance of intermediate carbohydrates and cyclic carbohydrates with α,β-C to O-Ester function (-CH_2-OOR) which can be found as D-Allose or 3,4-Altrosan (see Fig. 2b) in VT150. On the contrary, intensities in this region for VT500 much decreased because of alkali excess meaning better efficiency of carbohydrate retro-aldol splitting towards final compound without accumulation of intermediates. Furanes resonating downfield (δC/δH: 110 - 150 / 6.0 - 8.0 ppm) and are found for VT150. This confirms the conclusion that some leftovers of the furan derivatives cannot react due to insufficient acidity and remain in the HSQC NMR spectra after the reaction. On the contrary, VT500 illustrates complete conversion of these units resulting in a clean aromatic region.

1.2.5. *CPMAS NMR*. 13C{1H} cross polarization magic angle spinning (CPMAS) measurements were carried out using a Bruker range Avance 400 MHz Solid State spectrometer operating at 100.6 MHz and a Bruker 4 mm double resonance probe-head operating at a spinning rate of 8 kHz.

1.2.6. *SEM and EDX.* The surface morphology and structure of the prepared samples were analyzed by field emission scanning electron microscopy (FESEM, Regulus SU8100).

1.2.7. *Detergent fiber analysis.* The fiber fractions of the feedstocks (e.g., neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL)) were determined with an ANKOM A2000 Automated Fiber Analyzer (ANKOM Technology, Macedon NY, USA) according to VDLUFA (2012; methods 6.5.1, 6.5.2 and 6.5.3). The hemicellulose, cellulose, lignin were calculated from the fiber fractions as: hemicellulose = NDF – ADF, cellulose = ADF – ADL, and lignin = ADL.

1.2.8. *HPLC*. The aromatics (phenols and furans), were measured quantitatively using a HPLC (ICS-3000) with the UV-detector (Dionex ICS 3000; Thermo Fisher Scientific Inc., USA). Prior to the measurement, liquid samples were diluted 1:4 with a 10% acetonitrile solution, and filtered with PTFE-syringe filters (Neolab PTFE 0.2µm-green). To quantitatively measure the organic acids (acetic, lactic and formic acids) and, sugars (fructose, glucose), the samples were diluted 1:1 with deionized water, and analyzed with the Ultimate 3000 UPLC system from DIONEX (column: Eurokat H (300mm; 8 mm, 10 µm), company KNAUER) and 0.01 N sulfuric acid were used as mobile phase. The aromatics, sugars and acids were extracted from solid products by putting 1 g of wet solid in 40 ml of distilled water at 60°C and mixed for 40 (min) with a stirring speed of 150 rpm. The resulting mixture was filtered by PTFE-syringe filter and the liquid was submitted to the HPLC and UPLC systems, respectively.

1.2.9. *Elemental analysis* was performed with a vario-MICRO cube CHNOS Elemental Analyzer (Elementar Analysensysteme GmbH, Langenselbold) The Elements have been detected with an Thermal conductivity detector (TCD) for C, H, N and O.

2. Figures

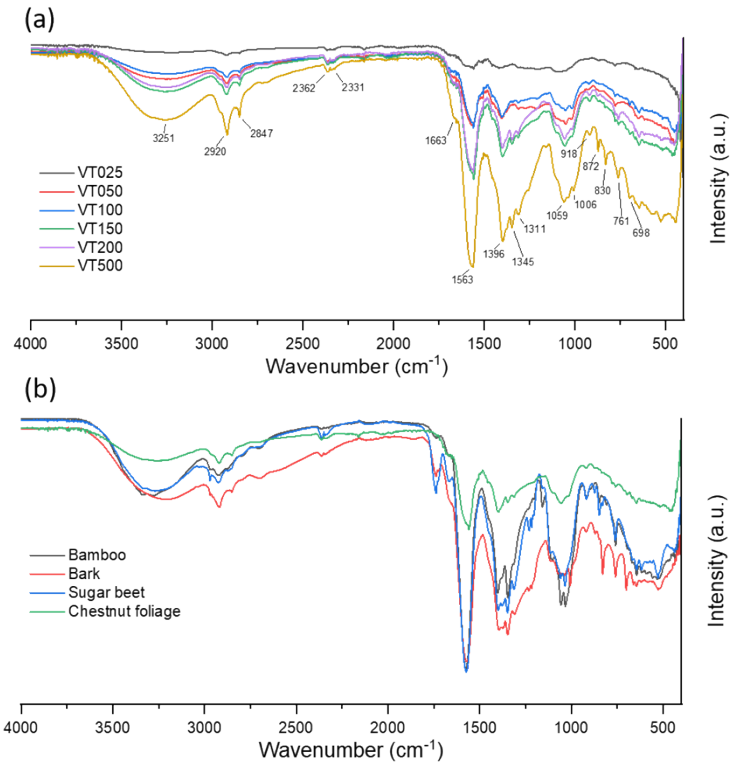

Figure S1. (a) FTIR spectra of the hydrothermal treatment products derived from chestnut foliage in the presence of different amounts of base. (b) FTIR spectra of the hydrothermal treatment products derived from bamboo, bark and sugar beet treated at ratio 1.50 eq. of KOH to carbohydrates. Summary of assigned peaks represented in Table S3.

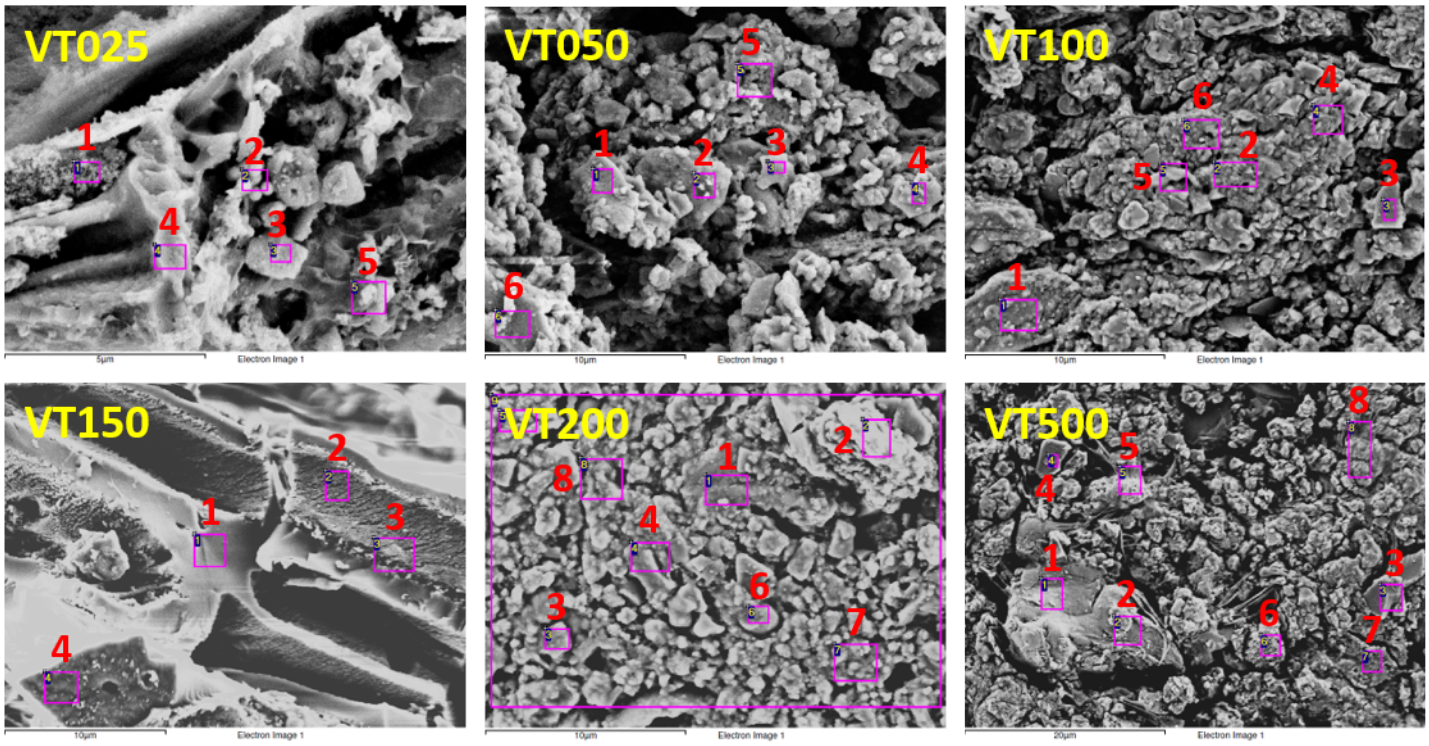

Figure S2. SEM images and corresponding selected EDX zones of chestnut foliage solid fractions after hydrothermal treatment at different KOH concentration. Elemental analyses of selected EDX zones are represented in the Table S4.

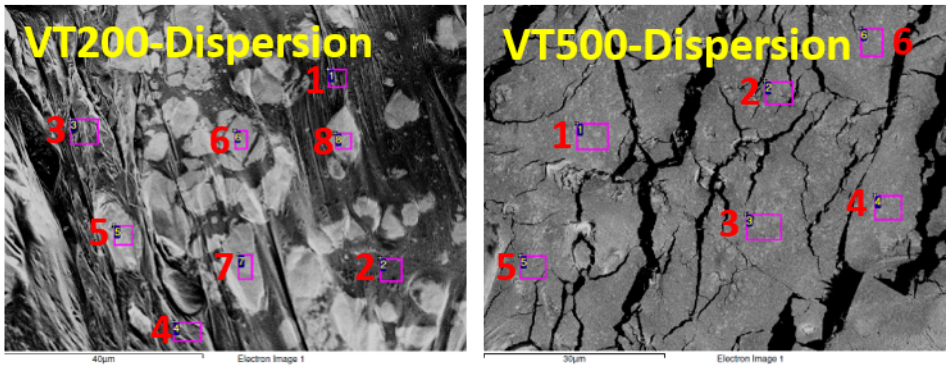

Figure S3. SEM images and corresponding selected EDX zones of chestnut foliage dispersion fractions after hydrothermal treatment at different KOH concentration. Elemental analyses of selected EDX zones are represented in the Table S5.

3. Tables

Table S1. Summarized experimental data for hydrothermal treatment of biomasses.

Sample	VT025	VT050	VT100	VT150	VT200	VT500	VT150	VT150	VT150
Biomass	Chestnut foliage						Bark	Bamboo	Sugar beet
eq. KOH	0.25	0.50	1.00	1.50	2.00	5.00	1.50	1.50	1.50
$m_{Biomass}$, g	1.2	1.19	1.22	1.20	1.22	1.22	1.20	1.20	1.20
$m_{KOH,practic}$, g	0.024	0.056	0.113	0.163	0.240	0.557	0.170	0.495	0.402
$m(H_2O)$, g	3.50	3.54	3.60	3.60	3.60	3.60	3.60	3.60	3.60
$pH_{before\ reaction}$	12.09	12.41	12.67	12.81	12.95	13.27	12.83	13.22	13.14
$pH_{after\ reaction}$	4.13	5.36	5.65	6.80	7.26	8.56	6.48	6.66	6.71

Table S2. Summarized data on detergent fiber analysis.

Biomass	NDF*, %	ADF*, %	ADL*, %	w(Carbohydrates), %
Chestnut leaves	51.11	42.19	23.65	27.5
Bark	47.34	35.14	18.54	28.8
Bamboo	91.95	65.52	11.99	80.0
Sugar beet	44.14	22.53	3.92	40.2

* NDF – neutral detergent fiber, ADF – acid detergent fiber and ADL – acid detergent lignin.

Table S3. Summarized FTIR data for chestnut foliage.

Peak, cm-1	Absorption, cm-1	Appearance	Group	Compound Class
Measured	Reference data			
3251	3550-3200	strong, broad	O-H stretching	water
	3300-2500	strong, broad	O-H stretching	carboxylic acid
2920, 2847	3000-2840	medium	C-H stretching	alkane
2362, 2331	2349	strong	O=C=O stretching	carbon dioxide
1663	1685-1666	strong	C=O stretching	conjugated ketone
	2000-1650	weak	C-H bending	aromatic compound
1563	1550-1500	strong	N-O stretching	amino group
	1650-1566	medium	C=C stretching	cyclic alkene
1396	1440-1395	medium	O-H bending	carboxylic acid
1345, 1311	1390-1310	medium	O-H bending	phenol
1311	1390-1310	medium	O-H bending	phenol
1059	1085-1050	strong	C-O stretching	primary alcohol
	1250-1020	medium	C-N stretching	amine
872	880 ± 20	strong	C-H bending	1,3-disubstituted
	880 ± 20	strong	C-H bending	1,2,4-trisubstituted
830	840-790	medium	C=C bending	alkene
761	750 ± 20	strong	C-H bending	monosubstituted
	780 ± 20	strong	C-H bending	1,2,3-trisubstitu

Table S4. Summarized EDX data based on Figure S3 spectra for solid phase fraction.

Zone	C, %	N, %	O, %	Mg, %	Al, %	Si, %	K, %
			VT025				
1	58.10	1.95	29.99			9.95	
2	43.79		38.15			18.06	
3			59.94		6.45	22.00	11.60
4	84.24		15.04			0.72	
5	76.79	4.73	15.89	0.74		1.85	
			VT050				
1	83.18		16.82				
2	82.68		16.40			0.92	
3	84.33		15.67				
4	78.97		19.08			1.95	
5	77.59		22.41				
6	81.75		17.07			1.18	
			VT100				
1	74.08		25.06			0.86	
2	74.46		23.12			2.42	
3	81.08		17.75			1.17	
4	74.28		22.91			2.81	
5	73.69		22.06			4.25	
6	74.00		24.32			1.68	
			VT150				
1	76.24		20.00			0.58	3.18
2	68.65		20.70			1.15	9.51
3	66.05		24.77			0.93	8.24
4	67.93		22.97			1.05	8.04
5	71.14		20.02			0.98	7.85
			VT200				
1	69.18		21.80			1.80	7.22
2	61.19		25.59			0.80	12.42
3	66.18		22.34			0.65	10.83
4	66.33		21.63			2.97	9.07
5	50.47		31.73			5.33	12.47
6	74.69		17.89				7.42
7	64.37		23.05			0.64	11.94
8	66.15		23.03			0.59	10.24
9	63.56		22.80			1.13	12.51
			VT500				
1	74.71		25.29				
2	24.01		69.65				6.35
3	77.69		22.31				
4	85.73		14.27				
5	81.76		18.24				
6	59.52		28.68			0.92	9.75
7	70.92		25.09				4.00
8	69.85		29.48				

Table S5. Summarized EDX data based on Figure S3 spectra for liquid/colloidal phase fraction.

Zone	C, %	O, %
	VT200	
1	64.15	35.85
2	62.73	37.27
3	64.39	35.61
4	60.90	39.10
5	61.64	38.36
6	64.79	35.21
7	66.91	33.09
8	65.97	34.03
	VT500	
1	65.01	34.99
2	65.73	34.27
3	66.83	33.17
4	37.30	32.70
5	65.68	34.32
6	66.97	33.03