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CHANGES OF CHEMICAL STRUCTURE AND COMPOSITION OF CHARCOAL BY RADIOCARBON PRETREATMENTS: DECONTAMINATION BY ABA AND ABOx TREATMENTS

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ABSTRACT. Charcoal is widely used for radiocarbon dating in archaeological and paleoenvironmental studies. Reliable ^{14}C dating requires appropriate chemical treatment to remove postdeposition contamination from the charcoal samples. This study assesses two pretreatments: acid-base-acid (ABA) and acid-base-oxidation with stepped combustion (ABOx-SC). In addition to ^{14}C, the effects of the treatments on the chemical structure and composition of charcoal were studied using Fourier transform infrared spectroscopy (FTIR) and C/H/O elemental analysis. Samples of pine wood charred in the laboratory at 270, 300, 400, 500, and 600°C, and environmental samples of charred pine wood from pyroclastic flow deposits in southern Kyushu, Japan, were tested. The laboratory-charred samples showed that NaOH treatment removed highly hydrophilic organic components derived from endogenous and exogenous organic materials in the samples and that oxidation treatment caused the oxidative degradation of molecules in samples starting from its edges. The ABA-treated environmental charcoal yielded younger ^{14}C dates than the ABOx-treated samples, probably owing to the effects of remaining organic contaminants bound to the edges of the aromatic molecular structures produced by the original pyrolysis. Meanwhile, it was found that ABA-SC treatment can reduce contaminants as effectively as ABOx-SC treatment. This implies that the stepped combustion (SC), not the chemical oxidation, is the key to reduce contaminant residue left after ABA and ABOx treatments. The results in this study indicate that the investigation of the structural and compositional changes of charcoal during its pretreatment is useful for assessment of the reliability of the ^{14}C ages.

KEYWORDS: charcoal, radiocarbon dating, chemical pretreatment, ABA, ABOx-SC.

INTRODUCTION

Plant-derived charcoal is formed by exposure of lignocellulosic biomass to high temperatures under restricted oxygen conditions. The lignocellulosic biomass is composed of carbohydrate polymers (cellulose, hemicellulose) and an aromatic polymer (lignin). The high temperatures and restricted oxygen cause the structure of lignocellulosic biomass to degrade, and chemically stable aromatic rings are formed by pyrolysis. As the pyrolysis temperature increases, aromatic rings are polymerized and chemically stabilized to become resistant to postdepositional chemical alternation and diagenesis (Preston and Schmidt 2006). Therefore, charcoal can remain well preserved for long periods in archaeological and geological deposits, making it useful for radiocarbon dating.

However, the aromaticity and reactivity of charcoal depend on the combustion temperature: low temperatures lead to charcoal with low aromaticity and high reactivity, while high temperatures give high aromaticity and low reactivity (Bird and Ascough 2012). Naturally charred material can be produced over a wide temperature range from 300 to 600°C (Swift et al. 1993), and its aromaticity varies with its production temperature. Especially at lower temperatures, insufficient pyrolysis of lignocellulosic biomass produces charcoal with various components, including aromatic components, reaction-intermediate components with low molecular weight, and aliphatic components such as degraded cellulose and lignin. Therefore, naturally produced charcoal is not homogeneous (Franklin 1951).

Furthermore, charcoal suffers alteration and degradation once buried, a process called self-humification (Cohen-Ofri et al. 2006; Ascough et al. 2011). This process alters the chemical structure of the charred material, increasing its oxygen content (wt.%) due to the presence of carboxylic groups. Among the various components of environmental charcoal, the highly aromatic

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components are the most chemically stable endogenous components of the original material due to pyrolysis. These components have high molecular stability and low hydrophilicity, and are generally influenced little by exogenous carbon, leading them to generate reliable ^{14}C data. Therefore, removing all components other than the chemically stable aromatic components is very important in the pretreatment of charcoal for ^{14}C analysis.

Buried charcoal often alters and degrades and can become contaminated by exogenous materials containing carbon. Such exogenous materials are mostly carbonates and humic substances (fulvic acid and humic acid) from the surrounding soil. Humic substances are produced by the degradation of organic material, and contain many hydrophilic substituents such as carboxylic groups (–COOH and –COO^{-}), hydroxyl groups (–OH), and amorphous polymers. Humic acid contaminants move up and down the deposition with the groundwater. The contaminants have a different ^{14}C age from that of the charred material, and this can adversely affect ^{14}C dating.

Organic contaminants are generally removed from charcoal samples by acid-base-acid (ABA) treatment, an established technique used from the early days of ^{14}C measurement (Olson and Broecker 1958). Initial acid treatment with HCl can remove carbonate contaminants from charcoal samples, and subsequent base treatment with NaOH can remove humic acid contaminants derived from soil during burial. A further treatment with HCl can remove atmospheric CO_2 absorbed during the NaOH treatment. This treatment sequence produces reliable ^{14}C age measurement of charcoal samples, although some studies have reported that for "old" charcoal samples it does not remove contaminants sufficiently (Wood et al. 2012). An alternative chemical method proposed by Bird et al. (1999) involves acid-base-oxidation with stepped combustion (ABOx-SC). Higham et al. (2009) reported that this method removed contaminants from charcoal samples more efficiently than ABA treatment. Indeed, after ABOx-SC some charcoal samples older than 20 ka were found to be thousands of years older than after ABA treatment, which cannot remove newer organic contaminants (Turney et al. 2001; Bird et al. 2002; Brock and Higham 2009; Douka et al. 2010; Wood et al. 2012). Therefore, ABOx-SC is regarded as the best pretreatment method for "old" charcoal samples, especially samples with ages close to the ^{14}C dating limit (Bird et al. 2014). However, the effectiveness of the method has been proved by the ^{14}C results, not by a rigorous chemical assessment of the processes during decontamination. Therefore, it is necessary to elucidate the effectiveness of decontamination by ABOx-SC by monitoring the changes of the chemical structure and composition of charcoal samples. Recent studies have chemically analyzed archaeological charcoal to investigate the diagenetic alteration and degradation of charcoal during burial and the effect of decontamination by chemical pretreatment. For example, Alon et al. (2002) used Raman spectroscopy to monitor the removal of humic substances from charcoal during ABA treatment. Rebollo et al. (2008) characterized the structure of charcoal during ABA treatment using Fourier transform infrared spectroscopy (FTIR) to assess sample contamination and charcoal preservation. Rebollo et al. (2011) reported that charred samples with C contents lower than 50% after pretreatment should be not used for ^{14}C dating because they cannot produce reliable ^{14}C dates, even if treated by ABOx-SC. Styring et al. (2013) used elemental composition and FTIR data to analyze the changes in chemical structure and composition of charred crop samples (not charcoal) during charring, postdeposition, and ABA treatment.

The changes in chemical structure and composition that ABA and ABOx treatments produce in charcoal as they remove humic substances have not been compared alongside the differences in ^{14}C dating after the treatments. Therefore, in this work, we examined the changes in chemical structure and elemental composition of the charred material during chemical treatment. We also elucidated the differences between ABA and ABOx decontamination processes and their mechanisms. To understand the two decontamination processes and the diagenesis, we investigated the chemical characteristics of two types of charcoal samples during their chemical treatment: laboratory-produced charcoal and samples from a geological site exposed to environmental conditions for a long time. The chemical changes in the charcoal were measured using FTIR and elemental analysis (C/H/O) at each stage of the chemical treatments.

SAMPLES AND EXPERIMENTAL METHODS

Samples

Laboratory Charcoal

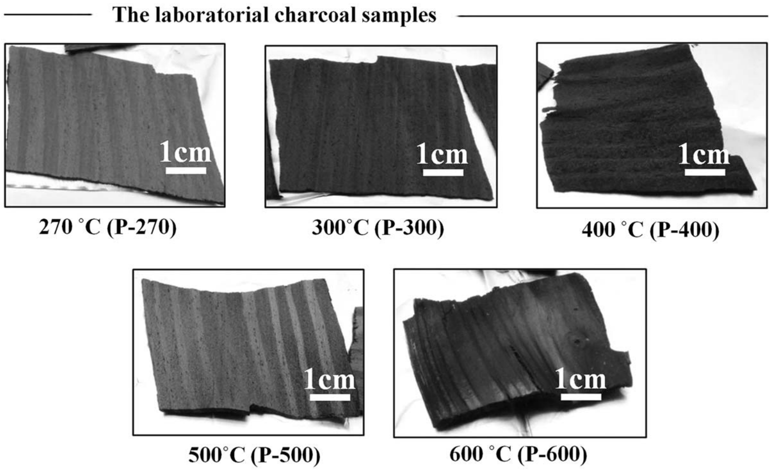
Charcoal samples were produced in the laboratory from pine wood (*Pinaceae densiflora*; sample code: P-; Figure 1) grown in Nagoya University, Japan, and felled in 2001. Samples were charred for 1 hr at 270, 300, 400, 500, and 600°C in a muffle furnace. The loss on ignition (LOI, wt.%) was calculated as the difference in sample mass before and after heating.

Environmental Charcoal

Environmental samples of charred pine (*Pinaceae larix*; sample code: Env-; Figure 2) were obtained from deposits of a pyroclastic flow in southern Kyushu, Japan. In this study, three samples were collected from the central and outer parts of a charred branch and also a peeled fragment considered to be the outermost part of the branch, which had soil deposits on its surface. The samples are considered to have various levels of postdepositional addition of exogenous materials, with such addition more likely in the samples taken from the outer parts of the branch. After the physical removal of soil using a stainless steel spatula, the samples were subjected to the following analysis.

Chemical Treatment

Each sample was roughly crushed to less than about 5 mm. The environmental samples were ultrasonicated in Milli-Q™ water to remove contaminants such as roots and sediment from the sample surface, and then dried at 80°C. Five different pretreatments were conducted on samples of both types: no treatment, only acid (A) treatment, acid-base-acid (ABA) treatment, acid-base-oxidation (ABOx) treatment, and acid-oxidation (AOx) treatment. The A treatment



270 °C (P-270)
300°C (P-300)
400 °C (P-400)
500°C (P-500)
600 °C (P-600)

Figure 1 Photos of laboratory charcoal samples

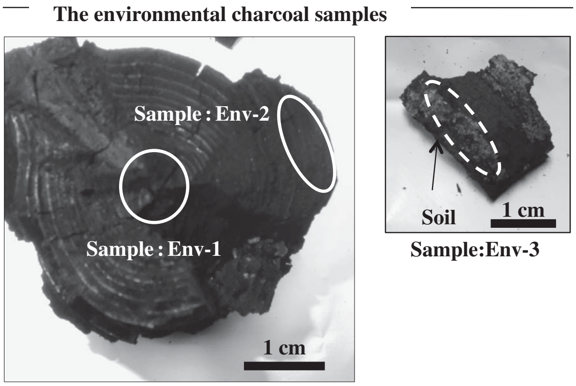


Figure 2 Photos of environmental charcoal samples. Samples Env-1 and -2 were collected from the inner and outer parts of the same branch. Sample Env-3 showed soil deposits on its outer surface.

was with 1.0M HCl overnight at 60°C. The ABA treatment involved an A treatment step with 1.0M HCl for 12 hr at 60°C, a subsequent base treatment with 1.0M NaOH for 3 hr at 60°C, and a repeat A treatment with 1.0M HCl overnight at 60°C. The ABOx treatment had three steps: an A treatment and then a base treatment, followed by an oxidation treatment using 0.1M $K_2Cr_2O_7$/2.0M H_2SO_4 for 20 hr at 60°C (Brock et al. 2010). The AOx treatment consists of an A treatment step with 1.0M HCl overnight at 60°C followed by an oxidation treatment.

Fourier Transform Infrared Spectroscopy (FTIR)

Absorption spectra of the laboratory and environmental samples were obtained to study the structural changes in their chemical compounds during treatment. A KBr pellet was prepared from a powdered sample and infrared (IR) spectra were obtained with an FTIR spectrometer (Spectrum 2000, Perkin Elmer) in the range 350–7800 cm^{-1} with a spectral resolution of 4 cm^{-1}, and data were averaged from 50 scans.

Spectra assignments were made according to previous reports by Ishimaru et al. (2007), Ascough et al. (2011), and Yizhaq et al. (2005). The main information obtained from the IR spectra in this study was in the spectral region below 2000 cm^{-1} because it contains the fingerprint region.

Elemental Analysis

The abundances of carbon, hydrogen, and oxygen in the charred samples were measured with an elemental analyzer (vario EL cube, Elementar Inc.). We used sulfanilamide ($NH_2C_6H_4SO_2NH_2$) as a standard for the measurement of C and H contents (wt.%), and benzoic acid (C_6H_5COOH) for the measurement of O content (wt.%). We calculated the H/C and O/C atomic ratios from the C, H, and O contents of the samples and plotted them in van Krevelen diagrams.

^{14}C Dating

Each of the dried samples (about 3–7 mg) was placed in a quartz tube (350 mm long, 9 mm diameter) together with CuO, Ag thread, and Cu. The quartz tube and CuO were reused during the experiments and were preheated before each use. The tube was evacuated and sealed, and then heated by either fixed combustion or stepped combustion (SC). During fixed combustion, the sealed tube was heated at 900°C for 4 hr to oxidize the sample carbon into CO_2 gas. The SC program was as follows: the sample in the sealed tube was precombusted at 630°C for 2 hr to remove the least stable organic components and any air or CO_2 adsorbed during the other procedures; the tube with the sample residue and CuO after precombustion was evacuated and resealed; and the sealed tube was heated at 900°C for 4 hr to oxidize sample carbon into CO_2. The CO_2 obtained by the two methods was purified in a vacuum line using refrigerants of ethanol/liquid N_2, n-pentane/liquid N_2, and liquid N_2. The purified CO_2 gas (about 1.5 mg C) was introduced into a quartz tube with an Fe catalyst (about 3 mg) and H_2 gas, and the tube was sealed. The sealed tube was heated at 620°C for 6 hr to convert the CO_2 gas into graphite. The graphite was packed in an aluminum holder and ^{14}C dated with a Tandetron AMS (4130-AMS by HVEE) at the Institute for Space-Earth Environmental Research, Nagoya University. For ^{14}C dating, NIST HOx-II served as a ^{14}C standard, and charred wood fragments in pyroclastic flow deposits derived from Aso-3, which is a 120-ka caldera in Kyushu, Japan (Matsumoto et al. 1991), and Kishida® oxalic acid containing ^{14}C-free carbon served as ^{14}C background samples. The Aso-3 standard was pretreated by the ABA method, same as the samples. The ABA-treated Aso-3 and the Kishida oxalic acid were heated to produce CO_2 gas, and converted the CO_2 gas into graphite.

RESULTS

Mass Loss of Samples by Chemical Treatments

Laboratory charcoal samples produced at ≥400°C (P-400, -500, -600) showed lower mass loss after all pretreatments than those produced at ≤300°C (P-270 and -300), and their LOI was much higher than that of the charcoal samples at ≤300°C (Table 1). This suggests that the chemical resistance of the charred pine depends on the charring temperature and increases greatly when the charring temperature is ≥400°C.

The environmental samples (Env-1, -2, -3) collected from different positions in a charred wood branch showed no significant differences in mass loss after pretreatment (Table 2). The first A treatment led to a sample loss of 30–40%. The ABA and ABOx treatments led to losses of 50–70%.

FTIR Analysis

Laboratory Samples (P-270, -300, -400, -500, -600)

Infrared absorption spectra of the laboratory samples are shown in Figure 3. IR spectral assignments are listed in Table 3. The spectra of samples P-270 and -300 are dominated by signals at 1030–1160 and 1450 cm^{-1} corresponding to aliphatic material with C–O–C and alcohol OH groups, 1420–1430 cm^{-1} corresponding to cellulose, and 1510 cm^{-1} corresponding to lignin (Kuriyama 1979; Ascough et al. 2011). They also contain bands with low absorbance at 1600 and 1700 cm^{-1} for aromatic carboxyl groups, aromatic $C = C$ ring structures, and aromatic carbonyl groups with $C = O$ stretching (Ascough et al. 2011). The spectra of these samples showed significant changes after A and ABA treatments. After ABOx and AOx treatments, which include an oxidation procedure, the absorption bands at 1200–1220 cm^{-1} for aromatic C–O, and at 1270 and 1700 cm^{-1} for carboxylic acids (Rebollo et al. 2008; Ascough et al. 2011) increased.

Samples charred at ≥400°C showed significant spectral changes. The spectra of P-400 are dominated by signals at 1200–1220 cm^{-1} for aromatic C–O, 1270 and 1700 cm^{-1} for carboxylic acids, and bands for lignin and cellulosic materials disappeared. Aromatic absorption bands

Table 1 Elemental composition of laboratory charcoal samples before and after chemical treatments.

			Elemental composition				
Sample	Chemical treatment	Weight loss (%)	C (%)	H (%)	O (%)	H/C	O/C
P-0							
	Nil	—	49.7	7.0	43.6	1.67	0.66
P-270							
(LOI 13%)	Nil	—	52.2	6.2	41.3	1.42	0.59
	A	11	54.1	6.5	40.6	1.43	0.56
	ABA	28	51.5	6.6	42.1	1.53	0.61
	ABOx	24	47.7	5.9	44.3	1.46	0.70
	AOx	12	51.2	6.1	41.5	1.41	0.61
NaOH extract		—	21.9	2.8	19.3	1.55	0.66
P-300							
(LOI 23%)	Nil	—	55.3	6.2	40.7	1.32	0.55
	A	3	56.9	6.5	37.9	1.35	0.50
	ABA	16	56.0	6.4	38.6	1.36	0.52
	ABOx	9	49.3	5.6	43.6	1.35	0.66
	AOx	9	51.0	5.7	43.8	1.34	0.65
NaOH extract		—	16.2	2.1	12.3	1.55	0.57
P-400							
(LOI 62%)	Nil	—	71.9	4.6	24.2	0.75	0.25
	A	2	71.6	4.9	24.9	0.82	0.26
	ABA	8	68.4	5.1	28.1	0.88	0.31
	ABOx	2	63.7	4.5	35.0	0.83	0.41
	AOx	7	69.7	4.6	27.0	0.79	0.29
NaOH extract		—	N/A	N/A	N/A	N/A	N/A
P-500							
(LOI 71%)	Nil	—	78.1	4.3	19.7	0.65	0.19
	A	0	74.4	4.2	19.1	0.67	0.19
	ABA	11	74.8	4.2	21.1	0.66	0.21
	ABOx	5	73.1	3.7	26.1	0.60	0.27
	AOx	4	72.6	3.9	22.5	0.64	0.23
NaOH extract		—	N/A	N/A	N/A	N/A	N/A
P-600							
(LOI 80%)	Nil	—	85.0	2.9	15.2	0.41	0.13
	A	1	82.7	3.9	15.0	0.56	0.14
	ABA	−1	83.1	3.0	14.1	0.43	0.13
	ABOx	−12	71.5	3.1	23.1	0.51	0.24
	AOx	−31	65.7	3.4	30.9	0.62	0.35
NaOH extract		—	N/A	N/A	N/A	N/A	N/A

Nil = untreated sample; N/A = NaOH extract was not obtained for analysis.

were observed at 700–900 cm^{-1} for aromatic C–H deformation (Ishimaru et al. 2007; Ascough et al. 2011) and at 1600 and 1700 cm^{-1} for aromatic carboxyl groups. After A and ABA treatments, the spectra of these samples were not changed significantly, whereas ABOx and AOx treatments caused an increase in the intensity of the absorption bands

Table 2 Elemental composition of environmental charcoal samples before and after chemical treatments.

			Elemental composition				
Sample	Chemical treatment	Weight loss (%)	C(%)	H(%)	O(%)	H/C	O/C
Env-1							
Series 1	Nil	—	54.0	3.5	41.7	0.78	0.58
	A	30	65.5	2.8	36.0	0.50	0.41
	ABA	70	67.1	2.9	34.9	0.51	0.39
	ABOx	73	58.4	3.0	41.7	0.60	0.54
	AOx	43	57.8	2.8	40.9	0.58	0.53
	NaOH extract	—	51.8	2.6	32.6	0.59	0.47
Series 2	Nil	—	54.3	3.7	41.4	0.81	0.57
	A	32	65.7	3.1	36.2	0.56	0.41
	ABA	50	65.7	3.1	33.8	0.56	0.39
	ABOx	69	56.5	2.9	41.6	0.62	0.55
	AOx	37	57.3	3.1	41.8	0.64	0.55
	NaOH extract	—	39.8	2.0	27.5	0.60	0.52
Env-2							
Series 1	Nil	—	47.3	3.4	39.1	0.86	0.62
	A	33	65.2	3.0	37.2	0.54	0.43
	ABA	69	66.4	3.0	35.9	0.53	0.41
	ABOx	67	58.0	2.8	41.3	0.58	0.53
	AOx	46	58.5	3.0	40.7	0.61	0.52
	NaOH extract	—	51.6	2.6	33.3	0.60	0.48
Series 2	Nil	—	48.3	3.5	40.2	0.86	0.62
	A	36	64.7	3.0	36.2	0.54	0.42
	ABA	51	64.8	3.2	35.9	0.59	0.42
	ABOx	64	57.2	3.1	41.6	0.64	0.55
	AOx	38	61.7	3.2	41.8	0.61	0.51
	NaOH extract	—	36.2	1.8	24.3	0.60	0.50
Env-3							
Series 1	Nil	—	47.7	3.4	40.0	0.85	0.63
	A	33	63.5	2.9	37.8	0.54	0.45
	ABA	54	65.5	3.1	37.5	0.57	0.43
	ABOx	68	58.9	3.1	41.0	0.62	0.52
	AOx	36	59.3	3.1	41.7	0.62	0.53
	NaOH extract	—	43.3	2.2	32.5	0.59	0.56
Series 2	Nil	—	47.3	3.4	40.5	0.85	0.64
	A	34	63.5	3.0	39.0	0.56	0.46
	ABA	49	63.5	2.9	37.7	0.54	0.45
	ABOx	51	57.7	3.1	41.1	0.64	0.53
	AOx	44	51.8	3.0	43.1	0.68	0.62
	NaOH extract	—	38.7	2.0	27.8	0.61	0.54

Nil = untreated sample.

at 1200–1220 cm^{-1} for aromatic C–O. The spectra of P-500 and -600 are dominated by the aromatic bands at 700–900, 1200–1220, and 1600 cm^{-1}. These spectra were not significantly changed by A and ABA treatments, whereas ABOx and AOx treatments increased the intensity

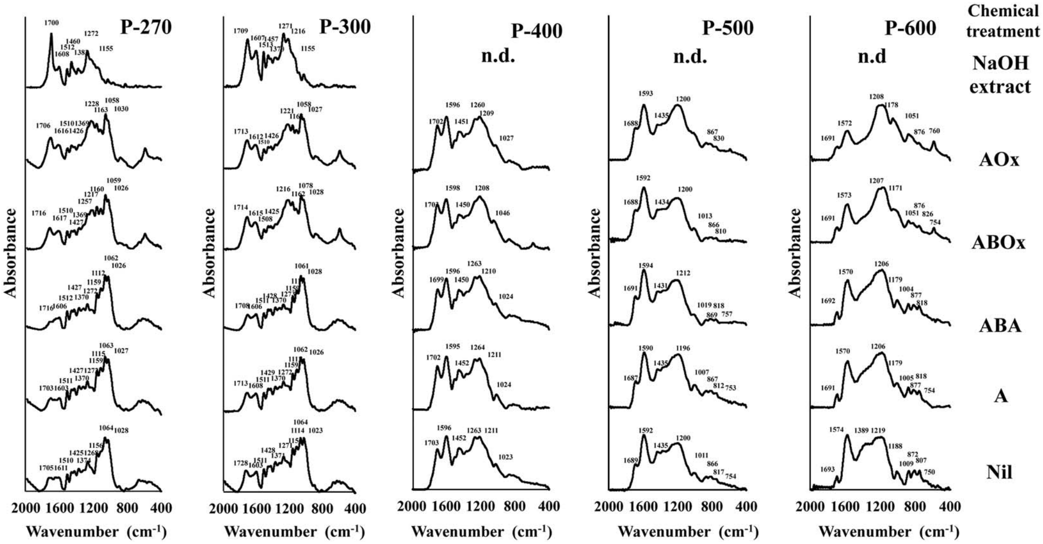


Figure 3 IR absorption spectra of laboratory charcoal samples. Nil = untreated sample; n.d. = not analyzed because NaOH extract was not obtained.

Table 3 Assignment of IR absorption spectra, referred to Ascough et al. (2011), Ishimaru et al. (2007), Yizhaq et al. (2005), and Kuriyama (1979).

Wavenumber (cm^{-1})	Assignment
1700	C = O stretching in COOH
1600	C = C vibrations of aromatic structures
1510	Lignin
1450	C–H deformation of aliphatic material
1420–1430	Cellulosic CH_2
1380	Ionized carboxylate group (COO^-)
1270–1250	O–H deformation in COOH
1220–1200	Aromatic C–O
1190	C–O of esters, phenols and ethers
1050	C–OH vibration
1030–1160	Aliphatic ether C–O and alcohol C–O
700–900	Aromatic C–H deformation

of the bands at 1200–1220 cm^{-1} and 1050 cm^{-1} attributed to C–OH vibration (Ishimaru et al. 2007) and weakened the absorption of the bands for aromatic C–H at 700–900 cm^{-1}. Samples P-270 and -300 charred at ≤300°C yielded NaOH-extracted materials, but the samples charred at ≥400°C (P-400, -500, -600) did not.

Environmental Samples (Env-1, -2, -3)

The spectra of the environmental samples (Env-1, -2, -3) are shown in Figure 4. The different samples showed similar characteristic spectral changes according to the pretreatment.

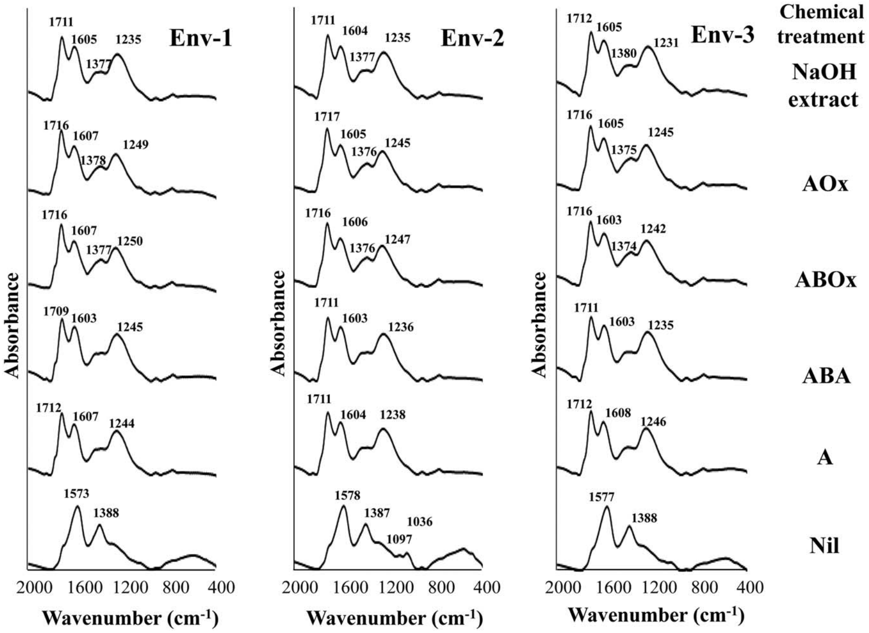


Figure 4 IR absorption spectra of environmental charcoal samples; Nil = untreated sample.

The spectra of the untreated samples are dominated by signals at 1380 and 1570 cm^{-1} for ionized carboxylate groups (COO^{-}) and aromatic C = C stretching. These bands disappeared after the first A treatment, and bands at 1250, 1600, and 1700 cm^{-1} appeared. This change indicates that carboxylate groups (COO^{-}) with metal ions lost their counter ions during the first acid treatment to form carboxyl groups (COOH) (Rebollo et al. 2008; Styring et al. 2013). Spectra recorded after the ABA, ABOx, and AOx treatments are similarly dominated by signals at 1250, 1600, and 1700 cm^{-1} for the aromatic carboxyl groups. The two treatment schemes that included oxidation led the absorption bands at 1380 and 1700 cm^{-1} for carboxylate groups and carboxyl groups (COOH) to increase slightly. The spectra of all of the materials extracted in NaOH are dominated by bands at 1220, 1380, 1600, and 1700 cm^{-1}, which are attributable to aromatic carboxylic groups.

Elemental Analysis

Laboratory Samples (P-270, -300, -400, -500, -600)

The results of elemental analysis for the laboratory charred wood samples are shown in Table 1 and Figure 5. The gray areas in Figure 5 represent the ranges of H/C and O/C ratios of carbohydrate, lignin, and two pyrolysis products of char and black carbon (data from Hammes et al. 2006). Both ratios for the laboratory charcoal samples decreased with increasing charring temperature (dotted regression line, Figure 5). This is similar to the results of previous work (e.g. Kuriyama 1979; Ascough et al. 2010). The decrease might be due to the dehydration of the organic components and the formation of polyaromatic components. The charcoal samples showed H/C and O/C ratios that increased slightly along the regression line according to the treatment. However, the O/C ratio after the ABOx and AOx treatments increased strongly, deviating from the line. The O/C ratio increments after ABOx and AOx treatments were 0.08–0.16 and 0.02–0.22, respectively.

Environmental Samples (Env-1, -2, -3)

Elemental analysis results for the environmental wood samples are shown in Table 2 and Figure 6. The untreated sample Env-1 showed C content of 54%, while the untreated samples Env-2 and -3 showed C content of 47–48%. The result suggests that sample Env-1, which was collected from the central part of a charred branch, was well preserved, compared with samples

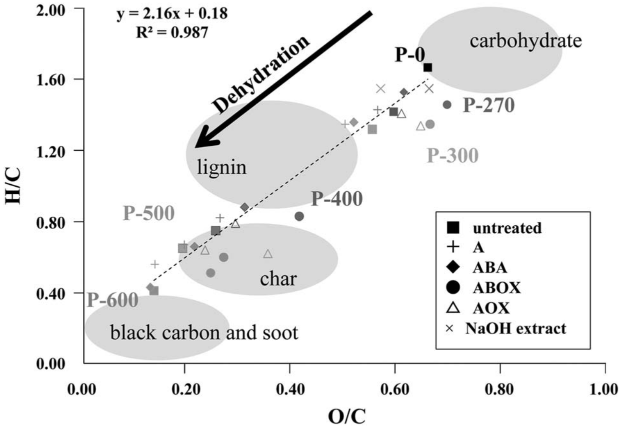


Figure 5 van Krevelen diagram of laboratory charcoal samples. The dotted line shows the liner regression for all untreated samples, representing the process of dehydration due to pyrolysis. The gray area represents ranges of H/C and O/C ratios for various biomolecular components (Hammes et al. 2006).

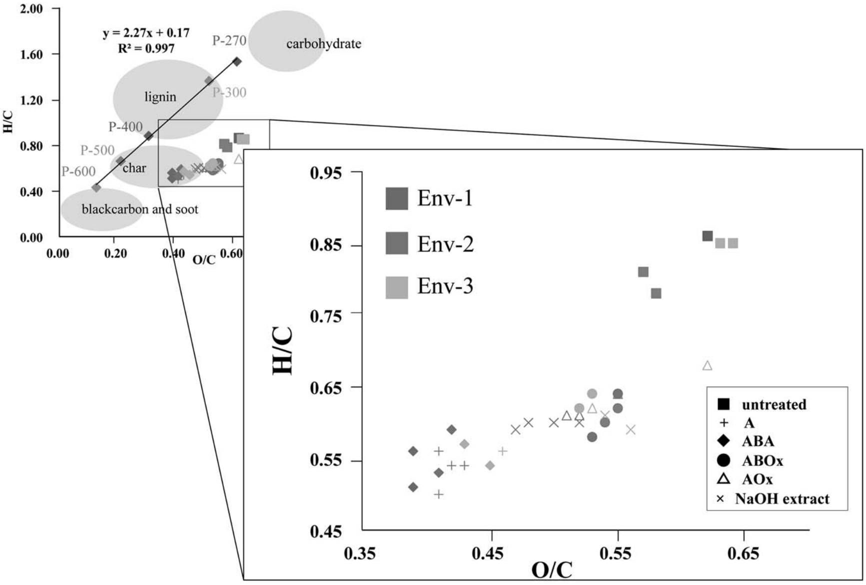


Figure 6 van Krevelen diagram of environmental charcoal samples. In the upper figure, the gray area represents ranges of H/C and O/C ratios for biomolecular components (Hammes et al. 2006), and the line shows the linear regression for the laboratory-produced sample after ABA treatment.

Env-2 and -3. However, all the chemically treated charcoal samples of Env-1, -2, and -3 showed C content more than 50%, and they could produce reliable ^{14}C dates (Rebollo et al. 2011).

The H/C ratios for the untreated samples lie within the range 0.78–0.86, and their O/C ratios within the range 0.57–0.64. All the environmental samples showed higher O/C ratios than the laboratory samples (Figures 5 and 6). The O/C ratio of the untreated sample Env-3 was higher than the O/C ratios of the other samples, indicating that this sample had more contaminants and components produced by oxidative alteration. The H/C and O/C ratios of the environmental samples were decreased after the first A treatment owing to increases of carbon content and reductions of H and O contents. After ABA treatment, the O/C ratio decreased slightly, whereas the H/C ratio was unchanged. This indicates that the elemental composition was not altered during the NaOH and HCl treatments following the first A treatment. The O/C ratio increased by about 1.0 after ABOx and AOx treatments, whereas it decreased after A and ABA treatments. The H/C ratio of the material extracted with NaOH was 0.59–0.61, which was lower than the H/C ratios for the untreated samples.

^{14}C Dating of the Environmental Samples (Env-1, -2, -3)

Results for the ^{14}C dating of the environmental charcoal samples are shown in Table 4 and Figure 7. The ^{14}C blank values were 47,750 ± 266 BP (NUTA2-22508) and 48,058 ± 258 BP (NUTA2-22536) for the Aso-3, and 48,003 ± 295 BP (NUTA2-22499) for the Kishida oxalic acid. The results show that contamination introduced into samples during the ABA treatment is small and can be ignored. In this study, the value of 48,058 ± 258 BP for the Aso-3 standard was used for blank correction of sample ^{14}C ages. The corrected ^{14}C ages of samples were calibrated with OxCal v 4.2 (Bronk Ramsey 2009) based on the calibration curve data of IntCal13 (Reimer et al. 2013).

The untreated samples showed ^{14}C ages of 42,540 ± 280 BP for Env-1, 42,000 ± 260 BP for Env-2, and 42,360 ± 360 BP for Env-3. These dates are the youngest recorded for each sample. The first A treatment did not significantly alter these ^{14}C ages, but older ^{14}C ages were observed after the ABA treatment, and the oldest ^{14}C age for each sample was found after the ABOx treatment (Env-1: 43,850 ± 300 BP; Env-2: 43,230 ± 300 BP; and Env-3: 43,670 ± 310 BP). The ^{14}C age of the material extracted by NaOH from sample Env-3 was younger than the

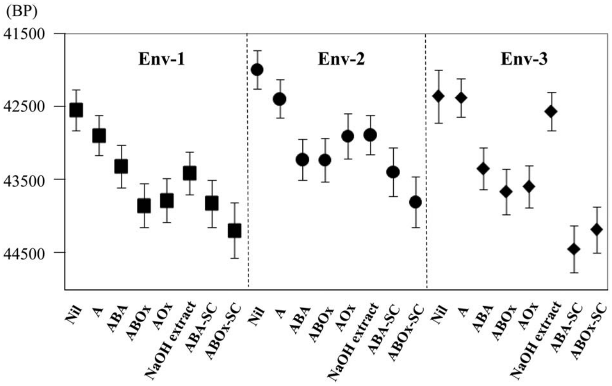


Figure 7 ^{14}C ages of environmental charcoal samples. Square: sample Env-1; Circle: sample Env-2; Rhombus: sample Env-3; Nil = untreated sample; SC = stepped combustion after ABA or ABOx treatment. Error bars are ±1σ.

Table 4 Radiocarbon dates of environmental charcoal samples.

Sample	Chemical treatment	Sample size (mg)	Carbon yield (%)	$\delta^{13}C_{VPDB}$ (‰)	^{14}C age ±1σ (BP)	Calibrated age ±2σ (cal BP)	Lab nr NUTA2-
Env-1							
	Nil	4.16	46	−26	42,543 ± 280	46,300–45,150	22502
	A	3.51	59	−23	42,895 ± 277	46,740–45,490	22503
	ABA	3.31	60	−25	43,317 ± 296	47,260–45,790	22504
	ABOx	3.29	56	−23	43,850 ± 302	47,910–46,240	22505
	AOx	3.91	52	−23	43,780 ± 299	47,820–46,180	22506
	NaOH extract	5.35	45	−24	43,412 ± 293	47,370–45,870	22510
	ABA-SC	5.43	34	−25	43,822 ± 322	47,920–46,190	22529
	ABOx-SC	5.17	32	−24	44,188 ± 376	48,480–46,450	22530
Env-2							
	Nil	4.65	42	−24	41,994 ± 260	45,890–44,820	22511
	A	3.24	62	−23	42,395 ± 263	46,230–45,120	22512
	ABA	4.15	60	−23	43,224 ± 282	47,120–45,730	22513
	ABOx	3.50	54	−24	43,229 ± 296	47,150–45,720	22514
	AOx	4.10	47	−26	42,905 ± 305	46,800–45,460	22515
	NaOH extract	4.30	56	−23	42,887 ± 273	46,720–45,490	22519
	ABA-SC	5.93	53	−22	43,392 ± 336	47,440–45,800	22531
	ABOx-SC	6.24	35	−20	43,802 ± 344	47,940–46,150	22532
Env-3							
	Nil	3.62	34	−27	42,357 ± 361	46,360–44,970	22520
	A	4.41	46	−22	42,377 ± 260	46,210–45,110	22521
	ABA	4.82	56	−23	43,350 ± 289	47,280–45,830	22522
	ABOx	4.70	56	−24	43,665 ± 306	47,700–46,070	22523
	AOx	4.60	53	−21	43,594 ± 286	47,575–46,040	22527
	NaOH extract	7.37	42	−22	42,568 ± 264	46,390–45,260	22528
	ABA-SC	5.86	50	−23	44,442 ± 321	48,680–46,780	22537
	ABOx-SC	6.92	41	−23	44,181 ± 316	48,340–46,530	22538

Nil = untreated sample; SC = sample with stepped combustion after ABA or ABOx treatment; The $\delta^{13}C_{VPDB}$ values, which were measured with AMS, have errors of ±1‰.

^{14}C ages of sample Env-2, and largely younger than those of Env-1. All the ABA-SC and ABOx-SC samples with additional SC, which is also a decontamination process, after ABA and ABOx showed older ^{14}C ages. Note that the ^{14}C ages of all samples treated with the ABOx-SC procedure are in agreement within 1 standard deviation.

DISCUSSION

Changes of Chemical Structure and Elemental Composition during Pretreatment

The chemical structures of the laboratory charcoal samples suggest that aromatic compounds were mainly formed at ≥400°C. Sample P-400 mainly consists of aromatic carboxyl groups, whereas samples P-500 and -600 consist of fewer carboxyl groups and more aromatic C–H at the edges of their molecular structures. Charcoal is chemically more stable when produced at a high temperature (Preston and Schmidt 2006). The changes in chemical structure and elemental composition were not observed in all the laboratory samples during A and ABA treatments, but the oxidation step in ABOx and AOx treatments caused chemical changes in all the samples. After the oxidation, aromatic C–O bonds between molecules increased, resulting in an increase of O/C ratio. This suggests that oxidative degradation occurred even in the charcoal samples produced at 500 and 600°C. At the same time, aromatic C–H, which is often bound at edge sites, decreased. Therefore, oxidative degradation occurred preferentially from the edge sites of the organic molecules.

The IR spectra of the NaOH-extracted material from the environmental samples appear similar to those of the charcoal samples after ABA and ABOx treatments. In contrast, the NaOH-extracted materials showed younger ^{14}C ages than the charcoal samples after ABA or ABOx treatment, especially in the sample Env-3, which is supposed to be the most contaminated of the three environmental samples. The NaOH-extracted material and ABOx-treated sample appears larger for Env-3 than for Env-1 and for Env-2. It is noted that the ^{14}C differences are likely to correspond with the levels of postdepositional addition of exogenous materials. These chemical structure and composition results of charcoal samples suggest that the NaOH-extracted material contains both the younger exogenous material and the charcoal sample dissolved by the NaOH.

The O/C and H/C ratios of the NaOH-extracted material were about 0.1 and 0.05, respectively, which were higher than those of the samples after ABA treatment. Therefore, NaOH treatment can dissolve the more hydrophilic molecules preferentially in the charcoal samples that might contain exogenous material and leave the less hydrophilic and more aromatic components. The high O/C and H/C ratios of the untreated samples relative to the treated samples probably arise from oxidative diagenesis (Cohen-Ofri et al. 2006) as well as from exogenous materials such as humic substances. Consequently, the NaOH-extracted material appears to be mainly composed of the charcoal itself, as Ascough et al. (2011) point out.

The ^{14}C ages of samples Env-1 and Env-3 after ABOx treatment were older than the ^{14}C ages measured after ABA treatment, whereas the age of sample Env-2 was not. Furthermore, the H/C and O/C ratios of the environmental samples were higher after ABOx treatment than after ABA treatment. These results show that the oxidation procedure can effectively remove exogenous material, while also degrading aromatic compounds in the charcoal sample. They also show that charcoal samples after ABA treatment could still retain contaminants and that the additional oxidation procedure reduced contaminant residue left after NaOH treatment.

The SC led to older ^{14}C ages than fixed combustion, suggesting that it removed exogenous carbon contaminants younger than the sample (Figure 7). Sources of the contamination are thought to be copper oxide (Vandeputte et al. 1998), organic remnants after chemical treatment, and atmospheric CO_2 adsorbed during sample loading (Bird et al. 1999) or Milli-Q cleaning. Figure 7 shows ABA-SC treatment could reduce contaminants as effectively as ABOx-SC treatment. This implies that the SC is key to reduce contaminant residue left after NaOH treatment and even after the oxidation procedure.

Interpretation of Decontamination by ABA and ABOx Treatments

Humic substances consist of exogenous organic material present in the environmental charcoal samples, and constitute a source of error in their measured ^{14}C age. They are highly hydrophilic due to the presence of many carboxyl and hydroxyl groups. In comparison, freshly charred material produced by pyrolysis contains a high proportion of aromatic components, but during burial the proportion of carboxyl groups increases through oxidative attack of the aromatic centers (Cohen-Ofri et al. 2006). Therefore, environmental charred material is more hydrophilic than freshly charred material. This diagenetic process increases the probability of the charred material being contaminated by exogenous humic acid. When exogenous organic matter (mainly humic acid) contaminates charred material, it is chemically bound to the edges of the molecular structure of the charred material. As described above, NaOH treatment can remove highly hydrophilic components from charred material, and dissolve together both exogenous materials and the degraded components of the charcoal sample. That is, components of relatively low intermolecular stability, which include more organic contaminant (humic acid), can be removed preferentially by the NaOH treatment, whereas the more stable components remain. However, the charcoal residues after NaOH treatment still be contaminated by humic acid bound tightly to the edges of the aromatic molecular structure of charcoal. The oxidation treatment of the laboratory charcoal samples appears to have degraded the compounds from their edge sites, suggesting that it can preferentially remove exogenous materials bound to the edges of an aromatic molecular structure. The oxidative degradation by $K_2Cr_2O_7$ can break the molecular structure of the charcoal into smaller components, isolating the inner parts of the aromatic components. These inner parts are less contaminated than the outer parts, and so could provide a more reliable ^{14}C age.

Assessment of Diagenesis and decontamination in Charcoal Samples

The assessment of diagenesis and decontamination in charred material is important for discussing the reliability of ^{14}C dates. Cohen-Ofri et al. (2006) suggested that oxidative processes cause charcoal to degrade. In this case, the carboxylation of the charcoal components is a primary factor, and leads to a higher O/C ratio relative to that of the fresh charcoal (Ascough et al. 2010, 2011). The charcoal residue after ABA treatment contained highly aromatic components that probably resulted from pyrolysis. These components were more resistant to diagenesis compared with degradation components, although they were not more resistant than the residue after ABOx treatment because of the chemical changes caused by the oxidation procedure. Therefore, it is possible that the elemental composition of the residue reflects that of the freshly charred material. The H/C and O/C ratios of the environmental charcoal samples (Env-1, -2, and -3) did not fall on the regression line of data for the laboratory charcoal treated by ABA (Figure 6). This means that the environmental wood samples after ABA treatment still contain contaminants acquired during burial that decrease the reliability of ^{14}C ages and/or consist of degraded components by oxidative alteration (Table 4 and Figure 7). If the H/C and O/C ratios of the charcoal samples after ABA treatment fell close to the regression line of Figure 6, it would imply their effective decontamination and thus that their ^{14}C ages are reliable.

Furthermore, the C content of a charcoal sample is a potential parameter for indicating the reliability of ^{14}C dating. A high C content after chemical treatment would suggest a sample high in aromatic components, which are supposed to be less contaminated. Rebollo et al. (2011) noted that charcoal with a C content lower than 50% after ABA or ABOx-SC treatment should not be used for ^{14}C dating because a charred wood sample should have 50–70% C content (Braadbaart and Poole 2008; Braadbaart et al. 2009). They also reported that charcoal samples lose 10–20% more C content after ABOx-SC treatment than after ABA treatment due to the increased mass proportion of other materials such as silica aggregates. However, we found that oxidation of the charcoal samples increased the O content and decreased the C content (Tables 1 and 2). The environmental samples showed C mass proportions after ABOx treatment that were about 9% lower than those after ABA treatment (Table 2). Lower C proportions after ABOx treatment than after ABA treatment were also observed in the laboratory samples

(Table 1). The ABOx treatment could reduce the C mass proportion to below the 50% reference value for the suitability of a sample for ^{14}C dating. Therefore, this reduction of C content should not lead to erroneous ^{14}C data because it is not caused by an increase in exogenous C material but rather by the degradation of the original C components (particularly contaminants). Consequently, attention should be paid to whether a sample has a low relative C content (around 50–60%) before its chemical treatment because the C content after ABOx treatment might be below 50%. The low C content might be attributed to the presence of other materials, such as silica aggregates, which have no carbon. Silica aggregates are not dissolved during chemical treatment, whereas the organic components are dissolved. Then, the content of silica aggregates in the charcoal sample after chemical treatment would increase and the C content would decrease.

From the results, O/C and H/C ratios together with C content of chemically treated charcoal samples can become potential indicators for assessment of diagenesis and decontamination in the samples to obtain reliable ^{14}C dates. Further study is needed to examine the effectiveness of the indicators by more analysis of charcoals of various ^{14}C ages and with various levels of contamination.

CONCLUSION

The chemical changes in laboratory-made and environmental charcoal during ABA and ABOx treatments were studied to compare decontamination processes. Laboratory samples produced by charring at 270–600°C, which represents the temperature of natural fires (Swift et al. 1993), were degraded via oxidization with $K_2Cr_2O_7$ solution after either acid or acid-base pretreatments. The IR spectra and measured O/C ratios of samples P-500 and P-600 suggest that the oxidative degradation commenced at the edge sites of their aromatic molecular structures and reduced the structure's original molecular weight. Therefore, the oxidation could preferentially degrade exogenous organic material bound to the edges of the original molecular structure, leaving the inner parts of the aromatic compounds, which are more likely to be components produced by the initial pyrolysis. This study shows that ABOx treatment can remove organic contaminants more efficiently than ABA treatment, as supported by previous work (Bird et al. 1999, 2014; Douka et al. 2010; Wood et al. 2012), and that SC is key to reduce contaminant residue left after ABA and ABOx treatments.

The assessment of diagenesis and decontamination in charcoal is important for discussing the reliability of ^{14}C dating. Elemental analysis (C/H/O) of charcoal samples is a potential method for identifying the presence of diagenetic alteration and contaminants. The results of this study suggest that contamination may remain in the environmental charcoal samples after ABA treatment. We also propose that the H/C and O/C ratios (plotted as a van Krevelen diagram) of charcoal samples after ABA treatment can be used to assess the possibility of residual contamination or diagenetic alternation. However, it is not clear how the burial diagenesis (carboxylation) of charcoal affects its ^{14}C date. Understanding this is necessary for assessing the decontamination and reliability of charcoal ^{14}C dates.

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