Accelerated aging of biochars; impact on anion exchange capacity

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Abstract

Little is known about the stability of biochar anion exchange capacity (AEC) and by what mechanisms AEC changes as biochar ages and weathers in soil environments. The goal of this study was to investigate chemical changes that may occur during ageing of biochar in neutral or alkaline soils and to assess the impact of ageing on AEC. To simulate and accelerate ageing, biochars were oxidized in alkaline hydrogen peroxide for 4 months. Spectroscopic evidence (*FTIR*, *XPS* and ¹³*C-NMR*) revealed that ageing increased carbonyl and alcoholic character in biochars produced at 500 °C and effected endoperoxide formation in biochars produced at 700 °C; the latter exhibited greater arene carbon character. Ageing caused biochar AEC to decline on average by 54% with greater decreases in biochars produced at 500 °C in contrast to biochars produced at 700 °C. The AEC of biochar derived from alfalfa meal and cellulose produced at 700 °C did not change significantly (p = 0.20 and p = 0.50, respectively) with ageing. Stability of AEC in the high temperature biochars is attributed to the presence of oxonium groups in bridging positions of arene carbon, which are sterically resistant to nucleophilic attack.

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1. Introduction

Biochar is the solid residue obtained from the pyrolysis of biomaterials. Anion exchange capacity (AEC) is a little studied but critically important property of biochar that will influence its value. Soils amended with high AEC biochar may exhibit reduced nutrient leaching. Furthermore, biochars with high AEC have the potential to be used in water treatment for the removal of anionic contaminants; hence high AEC is a potentially useful property that may contribute to biochar value. In previous work, we demonstrated that biochars can be produced with considerable AEC. Measured AEC values ranged from 0.602 to 27.76 cmol Kg⁻¹ for biochars produced from alfalfa meal, cellulose, and maize stover. We identified oxonium groups, pyridinium groups, and the adsorption of protons by the basal planes of π ring systems in condensed aromatic C as likely sources of the positive charge responsible for AEC. [1] Understanding the stability of biochar AEC in various environments is important as AEC stability will have a large influence on potential product use and hence value.

The surface chemistry and various physical and chemical properties of biochar are known to change with time in soil environments. However, there has been only one prior study that has addressed the stability of AEC in biochar. Cheng et al. [2] measured moderate AECs of 9.92 and 6.96 cmol Kg⁻¹ for biochars produced from oak using traditional kiln methods. After incubation for 12 months at 30 °C, AEC declined to 1.80 cmol Kg⁻¹, and biochar incubated for 12 months at 70 °C exhibited negligible AEC. Biochar coated with humic acid exhibited an AEC of 5.8 cmol Kg⁻¹, a slight decrease from the fresh biochar AEC of 8.4 cmol Kg⁻¹. This decline in AEC was attributed to surface conditioning with humic acid, not the degradation of AEC functional sites. The evidence presented by Cheng et al. [2] indicates that biochar AEC decreases with exposure to soil environments, yet the mechanisms responsible for change in AEC are not understood.

Anion exchange sites in biochar may change due to oxidation. Oxidation of biochar results in an increase in O containing functional groups; specifically hydroxyl, carboxylate, and aldehyde moieties [2,3]. Cheng et al. [2] reported Fourier transform infra-red (FTIR) spectroscopic evidence of significant increase in hydroxyl and carbonyl character of biochars recovered from 130 year old kiln sites and biochars incubated for 12 months relative to freshly made biochars prepared by similar methods. Elemental analysis confirmed an increase in O content in the kiln site and incubated biochars. Though not discussed in their publication, an increase in absorption in the 1100 to 1300 cm^{-1} range in Fig. 2 is consistent with alkyl C-O stretching, suggesting that these bonding structures are also generated during oxidation of biochar. They reported an increase in negative surface charge and a decrease in isoelectric point of the aged biochars; due to carboxylation of biochar surfaces with ageing. Formation of carboxylate groups on biochar surfaces increases negative surface charge and cation exchange capacity (CEC) but does not contribute to biochar AEC.

Oxidation of biochar C by various oxidizing agents yields surface changes unique to the oxidant [4,5]. For example, Moreno-Castilla et al. [5] found that oxidation of activated carbon with nitric acid yielded nitro groups on the surface of the activated carbon while oxidation with hydrogen peroxide and ammonium persulfate treatments yielded more carbonyl and carboxylate moieties. Oxidation with hydrogen peroxide, in particular, generated more ether groups on the activated carbon surfaces. Biochars produced by traditional kiln methods and incubated biotically developed carboxylate, lactone, and phenolic surface groups, with an increase in carboxylate character being the most prominent change [6]. Furthermore, different phase-dependent oxidation mechanisms may occur with carbon blacks, with formation of anhydride,

carboxylate, and hydroxyl groups occurring in liquid phase treatments and carbonyl groups forming on the surfaces during vapour phase treatments [7].

Ageing of biochar in soil environments may be caused by a variety of mechanisms, including radical addition [8,9], reaction with superoxide anions [10], and reaction with singlet oxygen [11]. Biotic oxidation may involve hydrogen peroxide released by soil biota [12,13]. Oxidation of polyaromatic hydrocarbons and other organic compounds in aquatic environments by reaction with singlet oxygen ($^1\text{O}_2$) has been demonstrated [14,15]. The fact that $^1\text{O}_2$ oxidizes dissolved organic compounds demonstrates that it is sufficiently stable in water to react in solution. $^1\text{O}_2$ is generated by photoexcitation of ground state oxygen in the air and may then diffuse into solution. Likewise, biochar at the soil surface is prone to reaction with $^1\text{O}_2$, with increased oxidation brought about by tillage events. In the soil, reactions with peroxides, $^1\text{O}_2$, and free radicals can occur depending on the chemistry of the biochar surfaces and the reductive-oxidative and pH conditions prevailing in the soil. Temperature, solar intensity, and soil moisture content all may influence reaction kinetics and mechanisms. Condensed aromatic carbon oxidizes slowly when exposed to reactive forms of oxygen; by contrast plant materials and microbial biomass in various stages of decomposition are much more rapidly oxidized. Ultimately, biochar C does change with ageing [1,3] and we seek to understand the mechanisms by which ageing occurs and the impact of these changes on biochar AEC.

The purpose of this study was to characterize biochar reactions with $^1\text{O}_2$ under alkaline conditions in an effort to understand oxidation and other aging mechanisms that may occur in neutral or alkaline soil environments. To accelerate the ageing process, biochars were exposed to $^1\text{O}_2$ generated by Fenton transformed H_2O_2 in an alkaline environment. The impact of this

ageing treatment on surface chemistry and AEC is assessed and reaction mechanisms are discussed.

2. Experimental

2.1 Biochar preparation

Biochars were produced from alfalfa meal, maize stover, and cellulose by slow pyrolysis using a muffle furnace at highest treatment temperatures (HTTs) of 500 °C and 700 °C. Details of biochar preparation are described in Lawrinenko and Laird [1]. Here *fresh biochars* refers to samples that were dialyzed against deionized (DI) water until the electrical conductivity of the bath solution was stabilized using Spectra / POR[®] MW 6-8000, 32 mm tubing. *Oxidized biochars* are subsamples (20 g) of the fresh biochars that were transferred to 125 mL high density polyethylene bottles and combined with 50 mL 1 M NaOH. Weekly, 1 mL of 30% H₂O₂ was added to this mixture, allowed to stand for 10 minutes, capped, and then shaken on a reciprocating shaker. This treatment was continued for a total of 4 months. pH of biochar slurries was checked using pH paper to ensure a high level of alkalinity throughout the oxidation treatment. On termination of this alkaline-oxidation treatment, biochars were rinsed with deionized water on 0.45 µm Teflon filter paper until conductivity of the rinsate was below 5 µS_{cm}⁻¹. The biochars were subsequently dried in a convection oven at 105 °C for 3 three weeks.

2.2 Elemental analysis

Fresh and oxidized biochar samples were analyzed in duplicate to determine C, N, H, and S content using an Elementar Vario Micro Cube combustion analyzer with argon as a carrier gas. Oxygen content was computed as the difference between the sum of C, N, H,

and S mass fractions and unity for biochars produced from cellulose. The other biochars contained significant levels of ash which precludes accurate determination of structural O by difference.

A Philips PW 2404 X-ray fluorescence (XRF) spectrometer was used for bulk chemical analysis. The spectrometer was equipped with a rhodium X-ray tube that was operated at 3600 Watts for this analysis. The spectrometer was flushed with helium (He) gas during all measurements. All measurements were corrected for tube drift by monitoring a reference sample (AUSMON-silicate minerals reference monitor). For the analysis, 5 to 10 g biochar samples were ground in a SPEX shatterbox puck mill for two minutes and two gram subsamples were placed into disposable sample cups and sealed with polypropylene film (6- μ m thick). Only single-sample measurements were made by XRF. No standard reference materials (SRMs) for the elements of interest were available with a comparable matrix. Therefore calibration standards were prepared by mixing the 700 °C cellulose biochar (0.92% ash) and the 700 °C alfalfa meal biochar (30.89% ash) with known quantities of reference standards, specifically NIST 1633a, NIST 2691, ACS Grade potassium chloride, USGS Nod-A-1, NIST 2910, and AWP Std I.

2.3 FTIR Analysis

FTIR spectra of biochar samples were obtained using a Nicolet 560 Magna - IR spectrophotometer using the diffuse reflectance accessory. Absorbance spectra were recorded from 4000 to 400 cm^{-1} at a resolution of 4 cm^{-1} . A total of 200 scans were averaged to produce the final spectra. Biochars were co-ground with spectroscopy grade KBr in a SPEX 5100 mixer mill at 1% wt/wt in KBr. Spectra were interpreted using published tables [16].

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139 *2.4 ¹³C solid state NMR spectroscopy analysis*

140 ¹³C nuclear magnetic resonance (NMR) spectroscopic analysis was performed on
141 biochars derived from cellulose. Two methods were employed due to the lack of ¹H
142 present in the 700 °C material, and the short T1C. Spectra of 500 °C HTT biochars were
143 acquired using Multi-CP, a recently developed quantitative cross polarization (CP)
144 technique [17] at 100 MHz using a Bruker DSX400 spectrometer and a Bruker 4-mm
145 ¹H–¹³C double-resonance magic angle spinning (MAS) probe head at 14 KHz. Biochar
146 samples were ground in a SPEX shatterbox puck mill for two minutes and were packed
147 into 4-mm diameter zirconia rotors with 5-mm long glass inserts at the bottom to
148 constrain the samples to the space within the radio frequency coil. The Multi-CP
149 experiments used a ¹H repolarization delay of 0.5 s with a ten-step ¹H ramp of 90-100%
150 of optimized CP conditions. Each CP period was looped six times totaling 50 ms of
151 cross-polarization followed by a 1 s recycle delay. A 4μs ¹H and 4.5μs ¹³C 90° pulse
152 lengths were used with 60 kHz ¹H decoupling during acquisition and during the Hahn
153 echo.

154 Carbon moieties were assayed relative to total carbon based on cumulative NMR
155 response in the following regions: carbonyl C 164 – 200 ppm, aromatic C 90 – 164 ppm,
156 ether or ester (o-alkyl) C 61 – 90 ppm, and alkyl C 0 – 61 ppm. Aromatic C not bound to
157 O (90 – 143 ppm) was differentiated from aromatic C bound to O (143 – 164 ppm) with
158 assignment of aromatic C two bond lengths from O in the range of 90 – 120 ppm and
159 aromatic C greater than two bond lengths from O in the range of 120 – 143 ppm.

Individual regions in the spectra were integrated from boundary to boundary. Region total area fractions were determined and are reported as fraction of C type.

The 700 °C HTT biochars were analyzed by solid state direct polarization (DP) NMR using an Avance II 600 NMR spectrometer, featuring a wide bore 14.1 T superconducting magnet. Biochar samples were ground in a SPEX shatterbox puck mill for two minutes then packed into 2.5 mm solid-state NMR rotors, with approximately 7.5 mg of biochar packed in each rotor. The ^{13}C spectra were obtained using a ^{13}C single pulse sequence with high powered ^1H decoupling, under 24 kHz magic angle spinning. The sequence used a 5 μs ^{13}C 90° pulse followed by an acquisition time of 13.5 ms under 100 kHz ^1H TPPM decoupling. Each spectrum was averaged over 16384 scans with a 5 second recycle delay, calibrated to ensure full signal relaxation time in between each scan. The spectral width was 75 kHz, or 500 ppm, centered at 115 ppm.

2.4 XPS analysis

Spectral measurements were performed using a Kratos Amicus/ESCA 3400 instrument. using unmonochromated Mg $K\alpha$ x-rays generated at 240 W. Photoelectrons emitted at 0° from the surface normal were energy analyzed using a DuPont type detector. The pass energy was set at 75 eV and a Shirley baseline was removed from all reported spectra. Deconvolution of C1s spectra was performed using Casa XPS.

2.5 AEC analysis

Anion exchange capacity was measured in triplicate using bromide as the index anion and chloride as the replacing anion. Biochar samples (1g, accurately weighed) in Milli-Q water were equilibrated to pH 6 by adding HBr, saturated with Br by adding 1M KBr, rinsed to remove excess salt, and the retained Br^- was displaced by washing with 2.5 M

CaCl₂. The solution containing the extracted Br⁻ was diluted to a known volume and a subsample was analyzed for Br⁻ using a Dionex[®] 1100 ion chromatograph equipped with an ASRS 300 4 mm conductivity detector. The mobile phase was 8 mM Na₂CO₃ / 1 mM NaHCO₃ and the method was run isocratically at 0.7 mL min⁻¹ using an IonPac[®] AG14A 5 µm 3 x 30 mm guard column and an IonPac[®] AS14A 5 µm 3 x 150 mm analytical column. A complete description of the AEC method is given in Lawrinenko and Laird [1].

2.6 Statistical analysis

Statistical analysis of AEC measurements was performed using SAS 9.3 (GLIMMIX, SAS 9.3, SAS Institute Inc., Cary, NC, USA). Data was treated using a 2-way ANOVA in a randomized complete block design in which individual biochars were treated as blocks. The fixed effect of age, fresh versus oxidized biochars, was assessed as well as the block effect and the 2-way interaction. Unpaired t-tests for differences between AEC means of fresh versus oxidized biochars were also performed and reported at the 0.05 significance level.

3. Results

3.1 Compositional changes

Oxygen is a component of many polar functional groups that accrue with oxidation and therefore the structural O content of biochars is an important indicator of the extent of biochar oxidation. Quantifying structural O in biochars, however, is difficult because O is associated with both organic and inorganic phases. Inorganic phases such as oxides, hydroxides, and carbonates are formed both during pyrolysis when biochar is produced and during the combustion of biochars. The thermal lability of carbonates, hydroxides, and hydrates alludes that the stoichiometry of

inorganic mineral phases can change during combustion, which is central to analysis of C, N, and H by combustion analysis techniques. Hence, determining structural O by subtracting ash, C and H from unity is not accurate for biochars that contain significant amounts of ash. Acid washing biochars will remove accessible salts, carbonates and hydroxides but will not remove all mineral phases and can effect changes in surface chemistry and structural O composition. Thus the structural O content of acid washed biochars determined by difference may not accurately reflect the structural O content of the original biochar. For these reasons, we determined structural O content by difference from unity after subtracting the C and H fractions only for the cellulose biochars as the cellulose used in this study had negligible ash content and therefore was not subject to the above discussed errors.

Carbon content decreased among all biochars following the ageing treatments (Table 1), as would be expected assuming an increase in O content with oxidation. The 500 °C and 700 °C cellulose biochars accrued 2.7% and 8.4% O, respectively, during ageing. The greater increase in O content of the 700 °C cellulose biochar suggests a different reaction pathway for the 700 °C biochar in contrast to the 500 °C biochar. Nitrogen content of the alfalfa meal biochars did not change appreciably following the ageing treatments, while a large decrease in N after these treatments was observed for the maize stover biochars. Nitrogen content of the original cellulose and the cellulose biochars was negligible. Ageing decreased H content in all but the 700 °C HTT cellulose and maize stover biochars. The slight increase in H content of the 700 °C cellulose and maize stover biochars following ageing may be due to abstraction of H from H₂O by radicals formed during pyrolysis and by basal planes of condensed aromatic C, which act as strong Lewis bases [9,18,19]. The S content of the biochars decreased following ageing (Table 1), indicating that S functional groups are labile under alkaline and oxidative conditions. Changes in

composition of the 700 °C HTT alfalfa meal biochar due to ageing were minor and the H content of this biochar was lowest among biochars in this study suggesting that the 700 °C alfalfa biochar may have had a greater degree of condensation of aromatic C in contrast to the other biochars which promoted stability.

Table 1. C, N, H, S, and O contents of fresh and aged biochars.

Feedstock	HTT (°C)	Age	% C	% N	% H	% S	% O
Alfalfa	500	Fresh	66.03	3.40	2.43	0.18	*
Alfalfa	700	Fresh	68.80	3.23	1.45	0.25	*
Cellulose	500	Fresh	84.80	0.00	2.98	0.08	10.82
Cellulose	700	Fresh	90.30	0.01	1.72	0.12	6.12
Maize Stover	500	Fresh	75.45	1.48	2.67	0.08	*
Maize Stover	700	Fresh	77.54	1.23	1.48	0.13	*
Alfalfa	500	Aged	61.46	3.57	2.18	0.11	*
Alfalfa	700	Aged	68.01	3.39	1.22	0.15	*
Cellulose	500	Aged	83.68	0.00	2.76	0.02	13.54
Cellulose	700	Aged	81.83	0.01	3.56	0.07	14.53
Maize Stover	500	Aged	74.85	0.74	1.97	0.05	*
Maize Stover	700	Aged	71.25	0.00	1.62	0.10	*

Results are averages for duplicate analysis of samples. RSDs for C, N, H, and S in standards range from 1.3 to 3.2%. RSDs for C in biochar samples were <3%, while RSDs for N, H, and S in biochar samples were as high as 30% as concentrations of these elements approached the instrument detection limit. * not measurable.

The alfalfa meal and maize stover biochars contained higher levels of inorganic elements than the cellulose biochar (Table 2). Nutrients, including P, Na, K, Mg, Ca, Si, Fe, Mn, and Zn, are present in plant tissue and are conservatively partitioned into biochars during pyrolysis due to their low volatility [20,21,22]. Other elements including Cr, Ti, and much of the Fe detected in the biochars probably leached from the stainless steel containment vessel during pyrolysis. The observed Cr (180 ppm) in the 700 °C cellulose biochar indicates the necessity of using the right materials in the construction of pyrolyzers to avoid heavy metal contamination of biochars intended for environmental application and water filtration. The presence of transition metals in biochar indicates that they may participate in ageing processes of biochar C in the soil.

Concentrations of Ca, Al, Fe, Ti, and Mn increased, while Si, Mg, Cl, and K decreased or were unchanged after the oxidation treatments. Most alkaline earth and transition metals tend to be very insoluble at high pHs and therefore remained as solid phases adsorbed to or embedded in the biochar surface. Silicon, Cl, and K are soluble in alkaline aqueous solutions and thus simply solubilized and were washed out of the biochar during the oxidation treatments. Other elements were unchanged or present at very low concentrations.

Table 2. Element composition of biochars measured by XRF. Values are reported as mass percent content.

	Feedstock	HTT (°C)	Na	Mg	Al	Si	P	Cl	K	Ca	Ti	Cr	Mn	Fe	Ni	Cu	Zn	Sr
Fresh	Alfalfa	500	0.064	1.4	0.10	2.1	0.60	1.92	5.9	3.6	0.017	0	0.013	0.060	0	0	0.0038	0.015
		700	0.064	1.6	0.12	2.1	0.64	2.12	6.2	3.8	0.014	0	0.011	0.065	0	0	0.0035	0.017
	Cellulose	500	0.12	0.17	0.0034	0.043	0.00089	0.34	0.0080	0.50	0	0	0	0.031	0	0	0	0.0034
		700	0.13	0.22	0.0036	0.062	0.00075	0.51	0.038	0.60	0	0.018	0	0.056	0	0	0	0.0054
	Maize Stover	500	0.021	1.3	0.12	7.5	0.25	0.33	2.9	1.5	0.024	0	0.019	0.12	0	0.011	0.0020	0.0036
		700	0.028	1.6	0.14	8.6	0.29	0.41	3.0	1.6	0.026	0	0.020	0.12	0	0.012	0.0028	0.0043
Aged	Alfalfa	500	0.058	1.3	0.14	1.7	0.58	0.018	0.26	7.8	0.022	0	0.016	0.082	0	0.0069	0.0055	0.018
		700	0.053	1.8	0.19	1.9	0.72	0.079	0.26	7.9	0.028	0	0.019	0.12	0	0.012	0.0072	0.019
	Cellulose	500	0.060	0.17	0.0036	0.042	0	0	0.023	1.6	0	0	0	0.041	0	0	0	0
		700	0.036	0.19	0.0040	0.052	0.0012	0.041	0.011	0.47	0	0.025	0	0.086	0.013	0	0	0.0036
	Maize Stover	500	0.046	1.1	0.16	5.9	0.19	0	0.19	1.8	0.040	0	0.028	0.19	0	0.010	0.0041	0.0032
		700	0.054	1.6	0.14	5.5	0.30	0.024	0.23	3.4	0.028	0	0.030	0.13	0	0.014	0.0032	0.0043

3.2 FTIR interpretations

Infra-red spectroscopy revealed evidence of surface chemical changes of biochars following the ageing treatments. Spectra of the 500 °C HTT biochars (Fig. 1) reveal increased intensity of the broad and relatively weak OH stretching bands (3200-3500 cm⁻¹) following the oxidation treatments indicating the development of alcohol functionality. The carbonyl peak at 1700 cm⁻¹ increased in relative intensity for all 500 °C HTT biochars, suggesting the formation of new carbonyl groups with ageing. The 1590 cm⁻¹ peak associated with oxonium heterocycles

increased in prominence after ageing [1]. Aromatic C-C stretching, however, also contributes to this peak and would contribute more with breakdown of large molecular structures due to greater dipole change associated with less restricted systems.

No evidence of hydroxyl character was observed in the FTIR spectra (Fig. 2) of the 700 °C HTT biochars either before or after the ageing treatments. Ageing increased absorbance in the functional group region between 1000 – 1800 cm⁻¹ and the fingerprint region between 560 – 960 cm⁻¹. Ageing of the 700 °C HTT alfalfa meal biochar also produced a peak at 1074 cm⁻¹ which is assigned as ether C-O stretching. No evidence of increased intensity for the 1700 cm⁻¹ carbonyl band was observed following ageing. These changes are consistent with a decrease in the degree of aromatic character and development of ether and peroxy-ether moieties manifested as C-O stretching between 1000-1300 cm⁻¹ and out of plane C-O and aromatic C-H deformation bands in the fingerprint region [23]. These data suggest that biochars produced at the different temperatures aged differently.

278
279 Fig. 1. FTIR spectra of 500 °C HTT biochars. Spectra are labeled as fresh and aged.

Fig. 2. FTIR spectra of 700 °C HTT biochars. Spectra are labeled as fresh and aged.

3.3 NMR

¹³C NMR spectroscopy of the 500 °C cellulose biochar was performed using a novel multi cross polarization technique (Multi-CP NMR) [17]. Table 3 presents the type of C moiety and amounts in the sample for both total and non-protonated C. Aromatic C constitutes the largest fraction of C in biochar. Almost half (48%) of total C in the fresh biochar was non-protonated indicating a high degree of condensation of aromatic C in this biochar. There is some evidence of methyl and other sp³ hybridized C as manifested by the 5% alkyl C character in the spectrum of the fresh biochar, which decreased to 4% following the ageing treatment. Carbonyl C and aromatic C bonded to O both increased by 1% with ageing. These changes observed by ¹³C-NMR spectroscopy agree with the 2.7% increase in O content determined by elemental

analysis (Table 1) and are indicative of development of carbonyl, phenol, and pyran-like moieties which is further supported by FTIR evidence (Fig. 1). Aromatic C bound to O increased from 8 to 9% on oxidation while non-protonated aromatic C two bond lengths away from O decreased from 4 to 2%. The decrease in aromatic C two bond lengths from O is also consistent with increased ageing of biochar. Aromatic non-protonated C greater than two bond lengths from O remained constant throughout ageing at 32% of total C indicating the recalcitrance of this form of C to ageing. Much of this C may have been deep within biochar and not present at the surface where oxidation and other weathering processes occur.

The Multi-CP technique was not able to yield ^{13}C NMR spectra of the 700 °C HTT cellulose biochars. The low H content of these biochars make cross polarization inefficient and the conductive nature of the material results in radio frequency induced heating. Despite dilution in a non-conductive matrix (laponite), these biochars still showed ^1H (wobble curve) of >2 MHz. The DP NMR spectra of these biochars were acquired using an alternate method and are presented in Fig. S2. These spectra exhibit broad peaks centered at 125 ppm, indicating a dominant presence of aromatic C. Additionally, homogeneous peak broadening resulted from rapid (<2 ms) T2 relaxation of ^{13}C nuclei leading to a broad halo effect making resolution between aromatic and other types of C in these samples impossible. This short T2 is likely a consequence of the electrical conductivity of arene and aromatic C and thus reflect high aromatic C content in biochars produced at 700 °C HTT.

Table 3. ^{13}C Multi-CP NMR results for the 500 °C HTT cellulose biochar. SSB are spinning side bands. bl is bond length.

	Carbon Moiety	Chemical shift (ppm)	Fresh (%)	Aged (%)
Total C	Alkyl sp³	0 - 61	5	4
	sp³ bound to O (ether, ester,	61 - 90	2	1

	alcohol)			
	Ar C with SSB	90 - 164	92	91
	carbonyl C	164 - 200	2	3
	Total C		101	99
Ar C bond distance to O	Ar C 2 bl from O	90 - 120	16	14
	Ar C > bl from O	120 - 143	60	61
	Ar C bound to O	143 - 164	8	9
	Non-protonated Ar C 2 bl from O	90 - 120	4	2
	Non-protonated Ar C > bl from O	120 - 143	32	32
	Non-protonated Ar C bound to O	143 - 164	8	9

314

315 3.4 XPS

316 X-ray photoelectron spectroscopy previously revealed the presence of oxonium heterocycles in
317 biochar [1]. The C1s spectra of 700 °C HTT biochars derived from maize stover and cellulose,
318 in addition to that of two model compounds, 1,3,5-triphenylbenzene and 2,4,6-triphenylpyrylium
319 bisulphate, are depicted in Fig. 4. Carbon bonded to oxonium in the spectrum of 2,4,6-
320 triphenylpyrylium bisulphate manifests as increased intensity ranging from 286.4 to 288.1 eV in
321 binding energy. This phenomenon is absent in the C1s spectrum of 1,3,5-triphenylbenzene
322 which is wholly comprised of aromatic C. Increased intensity in the 286.4 to 288.1 eV region of
323 the C1s spectra of the biochars is consistent with C bonded to oxonium in condensed aromatic
324 structures. Deconvolution of the C1s XPS spectra (Table 4) revealed that C represented by the
325 peak maximum at 285 eV decreased following ageing of biochar produced from maize stover
326 which is consistent with a statistically significant decline ($p = 0.049$) in AEC of this biochar with

327 ageing (Table 5). By contrast, 286.4 to 288.1 eV region of the C1s spectra exhibited only subtle
328 change following ageing in biochar produced from cellulose at the HTT of 700 °C, and the AEC
329 of this biochar did not significantly change with ageing ($p = 0.50$). Precise quantification of the
330 high binding energy component having a maximum at 287.3 eV in the pyrylium standard
331 spectrum and ranging from 287.9 to 288.6 eV in the spectra of the biochars is not possible
332 because the results of the integration are influenced by subtraction of the Shirley background.
333 However, these data show that highly oxidized C, oxonium heterocycles, formed in the pyrolysis
334 of cellulose at 700 °C are stable against ageing and that similar structures formed from other
335 components of lignocellulosic biomass; hemicellulose and lignin, are more subject to reduction.
336
337

338 Table 4: Deconvolution analysis of C1s spectra for the 700 °C HTT biochars and 2,4,6-
339 triphenylpyrylium bisulphate. FWHM = full width at half maximum. The goodness of fit test
340 statistic is X-square based.

Sample		Position (eV)	FWHM (eV)	Integrated Area	Composition %	Goodness of Fit
C₂₃H₁₈O₅S		284.6	1.4	15730.3	67.72	1146.53
		285.0	2.7	6653.79	28.64	1146.53
		287.3	1.1	845.994	3.64	1146.53
Maize Stover	Fresh	284.5	1.3	13809.1	50.18	1304.3
		285.2	2.7	12342.4	44.85	1304.3
		288.0	3.8	1369.17	4.98	1304.3
	Aged	284.5	1.3	15297	51.98	1384.08
		285.4	2.6	8081.35	27.46	1384.08
		287.9	5.5	6052.09	20.56	1384.08
Cellulose	Fresh	284.6	1.2	19509.5	59.77	1432.36
		285.3	2.7	9382.13	28.74	1432.36
		288.4	6.1	3747.53	11.48	1432.36
	Aged	284.5	1.3	15385.7	58.12	1565.34
		285.5	2.8	6868.4	25.94	1565.34
		288.6	6.6	4218.86	15.94	1565.34

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Fig. 4: C1s spectra of select 700 °C HTT biochars and model compounds.

3.5 AEC

The AEC of 5 of the 6 studied biochars decreased significantly ($p < 0.0001$) by an average of 54% following the ageing treatments (Table 4). By contrast, the AEC of the 700 °C HTT alfalfa meal biochar was 16.4 % higher following ageing, however based on an unpaired t-test, this difference was not significant ($p = 0.20$). The decrease in AEC following ageing for the 700 °C HTT cellulose biochar was also not significant, but the decline in AEC for the maize

stover biochar produced at 700 °C was significant. The ageing treatments caused significant decline in AEC in all biochars produced at the HTT of 500 °C.

Table 5. AEC (cmol Kg⁻¹ biochar) values for fresh and oxidized biochars measured at pH 6. Data presented as means (standard deviation).

Feedstock	HTT (°C)	Fresh AEC	Aged AEC	% Change	2-tailed p value
Alfalfa	500	3.01 (0.279)	1.52 (0.164)	-50.8	0.001
Alfalfa	700	9.64 (1.08)	11.2 (1.39)	+16.4	0.200
Cellulose	500	2.63 (0.211)	0.726 (0.359)	-72.4	0.001
Cellulose	700	18.1 (8.66)	10.5 (1.54)	-42.1	0.209
Maize Stover	500	3.77 (0.658)	1.72 (0.901)	-54.5	0.033
Maize Stover	700	13.8 (4.23)	6.88 (0.733)	-50.2	0.049

4. Discussion

4.1 Generation of ¹O₂

Conditions during the ageing treatment were alkaline, as the initial aqueous medium was 1 M NaOH; hence added H₂O₂ rapidly deprotonated to HO₂⁻. The peroxide anion reacts by one electron transfer with transition metals [24-27], defect sites in aromatic C [9,19], and electron rich basal planes of aromatic C [18,29] to form the peroxide radical. Equation 1 illustrates this reaction using iron (Fe) as an example of an electron acceptor.

Peroxide radical may further react with transition metals or aromatic C to form $^1\text{O}_2$ (O-O:) (Equation 2). Upon addition of H_2O_2 to biochar slurries, significant effervescence was observed which indicated formation of O_2 gas. Bottles were left open for 10 minutes to allow most of the gas to escape before the bottles were resealed and returned to the shaker to avoid rupture. The strong alkaline conditions drove formation of $^1\text{O}_2$ as acid generated (Equation 2) was consumed by ^-OH . Generation of $\text{H-O}^\bullet$ would be favored in neutral or acidic solutions, however the Haber-Weiss reaction (Equation 3) was discouraged by the strong alkaline conditions used in this study. Therefore, we assume that the predominant oxidative specie available in this system was $^1\text{O}_2$.

Fig. 5. Resonance structures of $^1\text{O}_2$.

4.2 Ageing pathways

Three resonance structures exist for $^1\text{O}_2$ as illustrated in Fig. 5. The zwitterion structures of $^1\text{O}_2$ are highly reactive and are a strong nucleophile which reacts with olephenic [29] and aromatic C [11,19] as depicted by the scheme of oxidation for naphthalene in Fig. 6.

383

384 Fig. 6. Oxidation of naphthalene by $^1\text{O}_2$.

385 We use naphthalene as an example to demonstrate the reaction of polyaromatic structures
 386 with $^1\text{O}_2$. In structure A, the π -electrons in the C-C sp^2 bond attract the electropositive oxygen
 387 resulting in the intermediate (B) in which the α -C attached to the peroxy group becomes sp^3
 388 hybridized and the β -C becomes a carbocation. Coulombic forces quickly end this intermediate
 389 by formation of the peroxy ether depicted (C). This structure rearranges with ring opening to
 390 form a dialdehyde (D). The aldehyde may be further oxidized to various carboxylates or reduced
 391 to alcohols, which are present in aged biochar [1,2]. The structure depicted in C, however, can
 392 be stable in graphite and graphene structures when the β -C is bridging between two aromatic
 393 rings; thus it is reasonable to assume that similar peroxy ether structures are also stable in
 394 biochars, which are dominated by condensed aromatic C.

Defect sites on graphitic surfaces are known to chemisorb both $^1\text{O}_2$ and triplet O forming stable five and six member ring configurations [30]. Condensed aromatic C also reacts with $^1\text{O}_2$ in a four plus two π electron scheme as depicted in Fig. 7. Formation of the six membered ring results in a stable endoperoxide structure [29,31]. This mechanism accounts for the increased O content of the 700 °C cellulose biochar (Table 1) and increased C-O stretching character observed in the FTIR spectra for all 700 °C biochars (Fig. 2).

The data suggest that more condensed aromatic C changes dominantly through a 4 plus 2 π electron, cycloaddition scheme (Fig. 7) [29,31] while less condensed aromatic C oxidizes by a 2 plus 2 π electron scheme (Fig. 6). The 2 plus 2 π electron mechanism involves formation of a strained 4 membered intermediate which is inherently more thermodynamically demanding than the concerted reaction. Due to high activation energy requirements, this reaction proceeds with little yield as evidenced by the 2.7% O content increase, yet accounts for the increase in carbonyl character of biochars produced at a HTT of 500 °C. The concerted, 4 plus 2 π electron scheme, which dominated for the more condensed 700 °C biochars, occurred to a greater extent as manifested by the 8.4% increase in O content of the aged 700 °C cellulose biochar and resulted in the formation of stable endoperoxides.

Fig. 7. Ageing of aromatic C with endoperoxide formation.

We suggest that the selectivity for ageing pathways is strongly governed by the nucleophilicity of aromatic π electrons and the presentation of bonding orbitals in the reactions between aromatic C and $^1\text{O}_2$. More condensed aromatic C may be better able to donate electrons to facilitate 4 plus 2 π cycloadditions. This mechanism implies bonding between the unoccupied molecular orbitals of singlet oxygen and π orbitals of aromatic C, which are perpendicular to the plane of the basal structure, thus steric factors will influence bonding. Greater condensed aromatic C has more planar sheet character and therefore more sites for this reaction pathway. By contrast, the 2 plus 2 π electron scheme is more likely to occur at edge sites and is promoted by the presence of electron donating heteroatoms which are of greater abundance in lower HTT biochars. The decrease in aromatic C two bond lengths away from O presented in the NMR data (Fig. 3, Table 3) is consistent with the 2 plus 2 π oxidation mechanism.

Superoxide (Fig. 8) may also have contributed to the ageing reactions.

Superoxide may form by one electron transfer from radicals found at defect sites in aromatic carbon [9,19] or through the oxidation of transition metals [24-27]. Superoxide reacts rapidly with water to form peroxide anion and hydroxide, however alkalinity in the reaction solution would inhibit this transformation. Itkis and co-workers [32] demonstrated that carbon black is readily oxidized by superoxide resulting in increased carboxylate surface character. Their study was performed in a dry, aprotic, environment. In soils this reaction is anticipated only when biochars are close to the soil surface under low humidity conditions. The strongly alkaline aqueous environment used in our study likely inhibited reaction between biochar and superoxide

and the FTIR and ^{13}C -NMR data do not indicate increased carboxylation of biochar in the ageing treatments.

4.3 Impact of oxidation on AEC

Anion exchange capacity in biochar is multicausal but is at least partly attributed to oxonium heterocycles [1]. The decrease in AEC after the ageing treatments (Table 5) may be caused by reduction of oxonium to ether. Nucleophilic addition of hydroxide to the α -C adjacent to oxonium results in formation of a hydroxyl cyclic ether (Fig. 9). Bond hybridization between C-O $^+$ changes from sp^2 to sp^3 with oxidation of the α -C and reduction of the O $^+$ resulting in a hydroxy cyclic ether structure (B) or a pyrone, depending on aromaticity of the ring. The reaction (Fig. 9) explains the decrease in AEC (Table 5) as a loss of formal charge on the hetero O $^+$ and is consistent with the observed as increased O content (Table 1), the decrease in high binding energy C identified by deconvolution of the C1s XPS of the 700 °C HTT maize stover biochar (Table 4), and the increase in alcohol character evident in the FTIR spectra of the 500 °C HTT biochars (Fig. 1) all of which occurred during the ageing treatments.

Fig. 9. Nucleophilic attack resulting in simultaneous reduction of oxonium heterocycle and oxidation of an exposed α -C.

The charge on the oxonium heterocycle makes it susceptible to nucleophilic attack when the α -C is exposed as described above. When both C atoms directly bonded to O^+ are bridging C, however, nucleophilic attack is sterically restricted and reduction of the O^+ heterocycle to ether is less likely. Hence presence of O^+ surrounded by bridging C in highly condensed aromatic structures may explain the stability of AEC for the 700 °C HTT alfalfa meal biochars (Table 4). The high aromaticity of C in the 700 °C alfalfa meal biochar is demonstrated by negligible changes in C, N, and H content after ageing and the lowest H content among the studied biochars (Table 1). The study results also suggest that the stability of AEC sites in biochar is related to the biomass feedstock. Cellulose employed in this study was a neat material, consisting of almost pure α -cellulose. Maize stover contains more hemicellulose than alfalfa meal and both materials contain cellulose, hemicellulose, and lignin [33,34]. Results indicate that the chemistry and composition of biomaterials may influence the chemistry of biochar. Further work is needed to explore the transformations of hemicellulose and lignin in pyrolysis to determine if biochar resulting from these materials is different from that derived from cellulose and if such biochar oxidizes by other pathways.

Pyridinium moieties may contribute some pH dependent AEC in biochar [1]. The ageing treatments had negligible effect on N content of biochar derived from alfalfa meal, but substantially reduced N content in biochar derived from maize stover (Table 1). This finding suggests that N in biochar derived from the maize stover and alfalfa meal may have been in different functional forms. Unfortunately, the FTIR and NMR spectra provided no clear evidence to either support or reject this hypothesis. From the literature, however, we know that most N in

biochars is present as 5 and 6 member heterocycles; specifically pyridine, pyrrole, and pyridone structures [1,35-37]. Of these N heterocycles, only pyridine is a weak base capable of being protonated and contributing to biochar AEC at relevant soil pHs. The pKa of pyridine, however, is 5.2 and we assume that pyridine functional groups in biochar have similar pKa values. If so, then most biochar pyridine groups would have been unprotonated and electrically neutral at the pH (6.0) at which AEC was measured. Furthermore, there is no apparent relationship between N content (Table 1) and AEC (Table 5) of the studied biochars ($r^2 = 0.01$) or between the change in N content and the change in AEC due to ageing ($r^2 = 0.24$). Had all of the N in the alfalfa meal, maize stover and cellulose biochars (average N is 3.40, 0.86, and 0.005%, respectively; Table 1) been present as pyridinium groups; N would have contributed on average of 243, 62, and 0.4 cmol kg⁻¹ to the AEC of the biochars, which averaged 6.3, 6.5, and 8.0 cmol kg⁻¹ (Table 5), respectively. The evidence suggests that pyridinium groups had negligible effect on the AEC of the studied biochars or the change in AEC following ageing. A different result might have been obtained had the study been conducted at a lower pH; as pyridinium groups are anticipated to contribute AEC to biochar under low pH conditions.

Lastly, protonation of the basal planes of aromatic C may also contribute pH dependent AEC to biochar [1,18,38]. Any decrease in aromaticity of biochar during ageing due to endoperoxide formation on polynuclear aromatic surfaces would have reduced the number of sites where protons can be stabilized in the aromatic π ring system. Consistent with this mechanism, we note that there was little change in aromaticity and no loss of AEC for the 700 °C alfalfa meal biochar during the ageing treatments and larger decreases in both aromaticity and AEC among the other biochars during the treatments. Although we were not able to measure the contribution of

protonation of aromatic C to biochar AEC in this study; the contribution is believed to be small at pH 6 due to the pH dependence of this mechanism [18,38].

5. Conclusions

Biochars produced at a HTT of 700 °C exhibit greater condensed aromatic character than biochars produced at 500 °C HTT; and changed by a concerted 4 plus 2 π electron mechanism yielding endoperoxide surface structures. By contrast, the 500 °C biochars dominantly oxidized by a 2 plus 2 π electron pathway, which led to the formation of more carbonyl and hydroxyl groups on surfaces of these biochars.

Anion exchange capacity of most biochars declined with ageing; exhibiting a mean decrease of 54%. Biochars produced at a HTT of 700 °C had higher AEC values and exhibited greater resistance to loss of AEC during the ageing treatments in contrast to 500 °C HTT biochars. The observed decline in AEC during ageing is partly attributed to the reduction of oxonium heterocycles to pyran structures due to nucleophilic attack on, and oxidation of, non-bridging α -C by OH⁻ groups. Hetero O⁺ atoms that are adjacent to two bridging aromatic C atoms, which are resistant to nucleophilic attack, are recalcitrant. Thus the 700 °C pyrolysis temperature yielded biochars with more condensed aromatic C that were more likely to have O⁺ groups with two adjacent bridging aromatic C atoms and hence be recalcitrant to nucleophilic attack and loss of AEC. Other possible mechanisms for the observed loss of biochar AEC during the ageing treatments include the loss of heterocyclic N in pyridinium structures and a decrease in the number of aromatic sites which may abstract protons. Both of these processes, however, are highly pH dependent and believed to contribute little to biochar AEC at pH 6. Further research

is needed to accurately quantify the contribution of all three potential sources of AEC in biochars as a function of pH, feedstock biochemistry, and pyrolysis conditions.

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Appendix: Supplementary Data

Electronic Supplementary Information (ESI) available: [details of any supplementary information available should be included here]. See DOI: 10.1039/b000000x/

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