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Radiocarbon Dating of Individual Lignin Phenols: A New Approach for Establishing Chronology of Late Quaternary Lake Sediments

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The reliability of chronology is a prerequisite for meaningful paleoclimate reconstructions from sedimentary archives. The conventional approach of radiocarbon dating bulk organic carbon in lake sediments is often hampered by the old carbon effect, i.e., the assimilation of ancient dissolved inorganic carbon (DIC) derived from carbonate bedrocks or other sources. Therefore, radiocarbon dating is ideally performed on organic compounds derived from land plants that use atmospheric CO_2 and rapidly delivered to sediments. We demonstrate that lignin phenols isolated from lake sediments using reversed phase high performance liquid chromatography (HPLC) can serve as effective ^{14}C dating materials for establishing chronology during the late Quaternary. We developed a procedure to purify lignin phenols, building upon a published method. By isolating lignin from standard wood reference substances, we show that our method yields pure lignin phenols and consistent ages as the consensus ages and that our procedure does not introduce radiocarbon contamination. We further demonstrate that lignin phenol ages are compatible with varve counted and macrofossil dated sediment horizons in Steel Lake and Fayetteville Green Lake. Applying the new method to lake sediment cores from Lake Qinghai demonstrates that lignin phenol ages in Lake Qinghai are consistently younger than bulk total organic carbon (TOC) ages which are contaminated by old carbon effect. We also show that the age offset between lignin and bulk organic carbon differs at different Lake Qinghai sedimentary horizons, suggesting a variable hard water effect at different times and that a uniform age correction throughout the core is inappropriate.

Obtaining an accurate chronology is one of the most important requirements in paleoclimate and paleoenvironment reconstruction. Radiocarbon dating is widely used for dating late Quaternary lake sediments.[1] Conventionally, radiocarbon dates are measured on biogenic materials (either bulk organic or inorganic matter) to obtain ages for sediments. However, in some lakes, ages obtained from radiocarbon measurements on sedimentary biogenic matter do not reflect the actual age of the sediment due to the lake reservoir effect,[2] i.e., the input of old and/or radiocarbon-dead dissolved inorganic carbon (DIC) from lake catchments or hydrothermal activity. Organic matter synthesized by aquatic organisms from this DIC pool will show abnormally old ages, confounding the ages of the sediment cores and interpretation of

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(1) Walker, M. *Quaternary Dating Methods*; John Wiley & Sons Ltd: Hoboken, NJ , 2005.

(2) Deevey, E. S., Jr.; Gross, M. S.; Hutchison, G. E.; Kraybill, H. L. *Proc. Natl. Acad. Sci. U.S.A.* **1954**, *40*, 285–288.

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climate records. A preferred technique to obtain reliable chronology and avoid the reservoir effect is to date preserved terrestrial plant remains (e.g., twigs, leaves, seeds, charcoal) that were rapidly transported to and preserved in lake sediment. Terrestrial plant material is synthesized using atmospheric CO_2 with contemporary ^{14}C, and its radiocarbon ages do not suffer from reservoir effects. However, many lake sediment cores are devoid of terrestrial fragments (especially if dating is required at specific sediment horizons), which prevents the use of this method for accurate age determination.

Dating purified organic compounds produced by terrestrial plants and preserved in lake sediments offers an alternative approach to constructing reliable chronology in lake sediments. Preparative capillary gas chromatography (PCGC) has been employed to isolate individual lipid compounds, such as hydrocarbons, fatty acids, and sterols for ^{14}C measurement.[3] For example, Uchikawa et al.[4] dated five samples of long chain leaf wax *n*-alkanes in a sediment core from Ordy Pond, Hawaii, after isolation by PCGC. The authors found that two *n*-alkane samples have similar ages as plant macrofossils, but three samples have either abnormally old or stratigraphically mixed ages. Uchikawa et al.[4] argue the inconsistencies of *n*-alkane ages in three out of the five samples are derived from contamination during sample preparation. Although dating leaf wax *n*-alkanes might provide supporting information for sediment chronology, a key challenge of dating *n*-alkanes is the possibility that *n*-alkanes derived from ancient soils and sediments may be delivered to the lakes by aeolian or fluvial transport.[5−8] Studies have shown leaf wax *n*-alkanes in sediments can have considerably older ages than bulk sediment.[9,10] Additionally, aeolian transport of leaf wax *n*-alkanes can deliver material from relatively wide source regions that are far beyond the immediate lake catchment area. Thus, the sources of *n*-alkanes to lake sediments can be complex and may complicate the interpretation of radiocarbon ages.

Lignin phenols may represent a better choice for radiocarbon measurements because lignin is unique to vascular plants[11] and represents up to 30% of vascular plant woody biomass (in comparison, leaf waxes constitute <1% of terrestrial plant biomass). Lignin is also abundant in lake and marine sediments and has been widely used for assessing terrestrial plant inputs to aquatic environments.[12,13] Another advantage of lignin relative to leaf waxes is that lignin is generally transported into sediments by water (rivers, streams, runoff, groundwater) as part of dissolved humic substances or plant debris and suspended particles.[14,15] As lignin is rapidly recycled in soils,[10,16,17] it is unlikely that significantly old lignin will be transported to lake sediments. McNichol et al. successfully isolated and dated individual lignin phenols from standard wood samples using PCGC.[18] The method requires derivatization of lignin phenols prior to the PCGC isolation, which increases the potential for contamination and the possibility of isotopic fractionation through incomplete derivatization reactions. This method also dilutes the sample carbon with large amounts of derivative carbon (~30−40% of the total carbon) and requires mass balance corrections. All these factors could potentially increase the error in the final radiocarbon measurement. Additionally, the relatively small loading capacity of capillary GC columns used in PCGC can make the isolation process time-consuming.

Here, we present a new method to purify the lignin phenols from natural samples for ^{14}C measurements. We use a microwave digestion procedure to extract lignin phenols from the samples[19] and combine solid phase extraction with high performance liquid chromatography (HPLC) coupled with diode array detector (DAD)[20] and fraction collector (FC) to purify and collect individual phenolic compounds for radiocarbon measurements. This HPLC-based method avoids derivatization and has a relatively high sample capacity (up to 30 μg per injection) and a short cycle time (each injection is ~25 min). The objectives of this work are (1) to demonstrate the feasibility of the technique using known age wood standards; (2) to compare the lignin phenol ^{14}C age with the varve counting age in lake sediments; (3) to investigate the temporal changes in reservoir ages from a well-studied lake, Lake Qinghai; and (4) to perform systematic dating in a laminated sediment core, Punderson Lake, to evaluated the consistency of the lignin radiocarbon ages.

EXPERIMENTAL SECTION

Chemicals and Standards. Lignin phenols listed in Table 1 obtained from Adrich (Milwaukeee, WI) were used as identification phenol standards. Copper(II) oxide powder, sodium hydroxide pellets, ferrous ammonium sulfate, anhydrous sodium sulfate, hydrochloric acid, phosphoric acid, ethyl acetate, methanol, dichloromethane, and water were obtained from EMD chemicals. All chemicals and standards acquired were of the highest purity available commercially, and the solvents were of HPLC or GC grades. All glassware was cleaned and precombusted for 12 h at 550 °C. Glass pipettes were precombusted for 12 h at 500 °C. All polytetrafluoroethylene (PTFE) tubes and vessels were cleaned and sonicated for 30 min using dichloromethane/methanol (v/v = 2:1) three times prior to use.

(3) Eglington, T. I.; Aluwihare, L. I.; Bauer, J. E.; Druffel, E. R. M.; McNichol, A. P. *Anal. Chem.* **1996**, *68*, 904–912.
(4) Uchikawa, J.; Popp, B.; Schoonmaker, J.; Xu, L. *J. Paleolimnol.* **2008**, *39*, 43–60.
(5) Bendle, J.; Kawamura, K.; Yamazaki, K.; Niwai, T. *Geochim. Cosmochim. Acta* **2007**, *71*, 5934–5955.
(6) Bendle, J.; Kawamura, K.; Yamazaki, K. *Geochim. Cosmochim. Acta* **2006**, *70*, 13–26.
(7) Harji, R. R.; Yvenat, A.; Bhosle, N. B. *Environ. Int.* **2008**, *34*, 959–965.
(8) Zhao, M.; Dupont, L.; Eglinton, G.; Teece, M. *Org. Geochem.* **2003**, *34*, 131–143.
(9) Eglinton, T. I.; Benitez-Nelson, B. C.; Pearson, A.; McNichol, A. P.; Bauer, J. E.; Druffel, E. R. *Science* **1997**, *277*, 796–799.
(10) Mollenhauer, G.; Eglinton, G. *Limnol. Oceanogr.* **2007**, *52*, 558–576.
(11) Sarkanten, K. V.; Ludwig, C. H. *Lignins: occurrence, formation, structure and reactions*; Wiley-Interscience: New York, 1971.
(12) Meyers, P. A. *Org. Geochem.* **2003**, *34*, 261–289.
(13) Meyers, P. A.; Ishiwatari, R. *Org. Geochem.* **1993**, *20*, 867–900.
(14) Hedges, J. I.; Clark, W. A.; Quay, P. D.; Rickey, J. E.; Devol, A. H.; Santos, U. *Limnol. Oceanogr.* **1986**, *31*, 717–738.
(15) Gordon, E. S.; Goni, M. A. *Mar. Chem.* **2004**, *92*, 331–352.
(16) Heim, A.; Schmidt, W. I. *Eur. J. Soil Sci.* **2006**, *58*, 599–608.
(17) Hofmann, A.; Heim, A.; Christensen, B. T.; Miltner, A.; Gehre, M.; Schmidt, W. I. *Eur. J. Soil Sci.* **2009**, *60*, 250–257.
(18) McNichol, A. P.; Ertel, J. R.; Eglinton, T. I. *Radiocarbon* **2000**, *42*, 219–227.
(19) Goni, M. A.; Montgomery, S. *Anal. Chem.* **2000**, *72*, 3116–3121.
(20) Lobbes, J. M.; Fitznar, H. P.; Kattner, G. *Anal. Chem.* **1999**, *71*, 3008–3012.

Table 1. Standard Phenols Isolated by the HPLC in This Study

peak no.	lignin phenols	abbreviation	retention time (min)	peak duration (min)	peak resolution
1	4-hydroxybenozoic acid	PAD	7.40	0.32	5.2
2	4-hydroxybenzaldehyde	PAL	9.16	0.36	2.8
3	4-hydroxy-3-methoxy-benzoic acid	VAD	10.13	0.34	2.4
4	3,5-dimethoxy-4-hydroxy-benzoic acid	SAD	11.01	0.38	1.0
5	4-hydroxyacetophenone	PON	11.45	0.46	2.0
6	4-hydroxy-3-methoxy-benzaldehyde	VAL	12.22	0.32	2.2
7	4-hydroxy-cinnamic acid	CAD	12.90	0.31	0.8
8	3,5-dimethoxy-4-hydroxy-benzaldehyde	SAL	13.18	0.35	3.3
9	4-hydroxy-3-methoxy acetophenone	VON	14.35	0.36	12.0
10	4-hydroxy-3-methoxy zimt acid (ferulic acid)	FAD	17.95	0.24	

Table 2. Radiocarbon Data for Standard Wood Samples and Lake Sediments

samples	sample i.d.	phenols	CO_2 yield (μmol)	PB[a] CO_2 (μmol)	fm[b]	fm error	fm/age exp[c]	fm corr[d]	error (corr)	phenol age (yr BP)	phenol age error
IEAE-C5	IAEA-C5−2	PAL	7.99	0.21	0.2313	0.0031	0.2305	0.2295	0.0088	11800	300
(Two Creeks wood)	IAEA-C5−4	SAD	68.70	0.21	0.2320	0.0017	0.2305	0.2319	0.0018	11700	60
	IAEA-C5−7	CAD	14.00	0.21	0.2306	0.0031	0.2305	0.2295	0.0059	11800	200
FIRI-A	FIRI-A-2	PAL	2.60	0.21	0.0598	0.0030	0.0033	0.0385	0.0268	26200	5600
(Kauri wood)	FIRI-A-4	SAD	12.10	0.21	0.0180	0.0014	0.0033	0.0130	0.0055	34900	3400
	FIRI-A-7	CAD	3.10	0.21	0.0518	0.0033	0.0033	0.0337	0.0221	27200	5300
FIRI-D	FIRI-D-2	PAL	5.65	0.21	0.5532	0.0056	0.5705	0.5633	0.0132	4610	190
(Dendro-dated wood)	FIRI-D-4	SAD	77.60	0.21	0.5667	0.0029	0.5705	0.5674	0.0031	4550	40
	FIRI-D-7	CAD	10.30	0.21	0.5609	0.0057	0.5705	0.5664	0.0086	4570	120
FIRI-H	FIRI-H-3	VAD	3.80	0.21	0.7485	0.0071	0.7574	0.7745	0.0187	2050	200
(Dendro-dated wood)	FIRI-H-6	VAL	46.30	0.21	0.7515	0.0026	0.7574	0.7535	0.0030	2270	30
Steel Lake (laminated)	STL-121	COMB	7.59	0.21	0.9176	0.0099	680 v[e]	0.9350	0.0131	557 c[f]	107
F. Green Lk. (laminated)	FG-80	COMB	3.45	0.21	0.8169	0.0138	1330 v	0.8479	0.0230	1238 c[f]	310
	FG-120	COMB	2.70	0.21	0.6936	0.0149	2462 v	0.7378	0.0372	2502 c[f]	683
Braya Sø (Peaty)	BS-28	COMB	3.84	0.21	0.6867	0.0104	1359 c	0.7071	0.0191	2937 c[f]	383
	BS-71	COMB	3.24	0.21	0.5582	0.0100	4911 c	0.5754	0.0225	5135 c[f]	479
Qinghai (hard water)	QH-183	COMB	2.41	0.21	0.5842	0.0142	5250	0.6061	0.0277	4020	370
	QH-388	COMB	3.69	0.21	0.3395	0.0077	10201	0.3420	0.0206	8620	480
	QH-439	COMB	5.63	0.21	0.2717	0.0052	11200	0.2706	0.0131	10500	390
Punderson (laminated)	PUN-135	COMB	4.88	0.21	0.8719	0.0103		0.8942	0.0157	900	140
	PUN-250	COMB	5.05	0.21	0.8132	0.0100		0.8361	0.0168	1440	160
	PUN-410	COMB	6.76	0.21	0.7030	0.0075		0.7159	0.0122	2690	140
	PUN-510	COMB	5.96	0.21	0.6483	0.0084		0.6609	0.0139	3330	170
	PUN-745	COMB	7.90	0.21	0.5548	0.0042		0.5619	0.0093	4630	130
	PUN-855	COMB	3.72	0.21	0.4794	0.0085		0.4898	0.0195	5730	320

[a] PB: procedure blank. [b] fm: fraction modern. [c] fm/age exp: expected fm or ages from varve counting or bulk organic matter. [d] fm corr and err corr: fm and error of the sample after correcting for the contribution of the process blank. The process blank contains 0.21 μmol C and is assumed to have an fm value of 0.30 ± 0.05 based on laboratory experience at NOSAMS (McNichol, unpublished data). [e] v: varve counting age. [f] c: calibrated age (yr BP), using Calib 6 online (http://intcal.qub.ac.uk/calib/calib.html).

Standard reference materials used for method validation are listed in Table 2. Three wood samples from the Fourth International Radiocarbon Intercomparison project (FIRI A, FIRI D, FIRI H)[21] and one wood sample from International Atomic Energy Agency (IAEA),[22] IAEA C5, were selected for comparison between the known age and phenol ^{14}C measurements. The radiocarbon ages of these wood samples have been determined and ascertained through multilaboratory analyses (Table 2).

Samples. Lake sediment samples with varve-counting age were selected from Fayetteville Green Lake (New York State, USA) and Steel Lake (Minnesota,USA), for comparison with the phenol ^{14}C ages. Three sediment samples from Lake Qinghai were selected to compare bulk organic carbon ^{14}C age and phenol ^{14}C ages in a lake with significant reservoir age.[23] Two sediment samples from a Greenland lake, Braya Sø, were selected to test the method in a lake surrounded by tundra peat land and arctic permafrost soils. Finally, a sediment core from Punderson Lake, Ohio, was dated in a series of down core samples to test the consistency of the lignin ages through time.

Prior to oxidation, the wood samples were pulverized. The sediment samples were reacted with diluted hydrochloric acid (0.5 N HCl) to remove the carbonate and rinsed thoroughly with deionized water and freeze-dried. All samples were extracted using dichloromethane/methanol (v/v = 9:1) by accelerated solvent extraction (ASE) to remove extractable lipid compounds before microwave digestion.

Oxidation and Extraction. We adopted the procedure of alkaline CuO oxidation with the microwave digestion described in Goñi and Montgomery[19] with minor modifications. Briefly, the

(21) Scott, E. M. *Radiocarbon* **2003**, *45*, 135–150.

(22) Rozanski, K.; Stichler, W.; Gonfiantini, R.; Scott, E. M.; Beukens, R. P.; Kromer, B.; van der Plicht, J. *Radiocarbon* **1992**, *34*, 506–519.

(23) Shen, J.; Liu, X.; Wang, S. M.; Matsumoto, R. *Quat. Int.* **2005**, *136*, 131–140.

oxidations were performed using a Milestone Ethos 1600 Advanced Microwave Lab Station fitted with up to 12 Teflon PTFE reaction vessels. NaOH solution was bubbled overnight with helium to remove dissolved oxygen in the water. A known amount of sample, 1 g of CuO powder and 100 mg of ferrous ammonium sulfate, was loaded into each of 11 vessels. The last vessel was loaded only with chemicals without samples, to serve as a procedural blank. After adding 20 mL of the degassed NaOH bubbled solution, the vessels were capped and placed in the rotating tray in the microwave. The reaction temperature (150 °C) was reached within 10 min and maintained for 90 min. The reaction vessels were allowed to cool after the reactions were completed. The contents of each vessel were transferred to PTFE centrifuge tubes, which were centrifuged to separate the solids from the hydrolysate. The supernates were transferred to new 60 mL Pyrex tubes. The solid in the PTFE centrifuge tubes was washed with clean 1N NaOH solution and centrifuged two more times to ensure efficient transfer. The alkaline solution was acidified to pH = 1 by addition of 6 N HCl. The organic components in the solution were extracted using ethyl acetate (6−8 mL) three or four times and were transferred to a new 40 mL Pyrex tube. Any excess water that may have been present in the samples was removed using cleaned Na_2SO_4, and the samples were transferred into clean 40 mL vials. The ethyl acetate was dried in Turbo-Vap under high-purity N_2 flow.

Purification and Verification. We modified the HPLC method of Lobbes et al.[20] to purify samples. Prior to HPLC separation, the samples were redissolved in methanol and passed through a Bakerbond spe Octadecyl (C18) Disposable Extraction Column to remove some of the neutral components. The methanol was dried under a stream of high-purity N_2. The dried samples were redissolved in HPLC-grade water. The water solution was passed through an Alltech True PTFE Syringe Filter using Norm-Ject 1 mL Single Use Syringes into 2 mL vials.

Further purification and separation of lignin phenols were carried out on an Agilent 1200 HPLC system consisting of a degasser (G1379B), a binary pump (G1312A), an isocratic pump (G1310A), an injection autosampler (G1329A), a diode array detector (G1315D), a thermostat column compartment (G1316A), a fraction collector (G1364C), a fraction collector-auto sampler (G1330B), and a 6310 quadrupole LC-MS. A ZORBAX Eclipse XDB-C18 column of dimensions 5 μm × 4.6 × 150 mm was used along with a ZORBAX Eclipse AAA 4.6 × 12.5 mm × 5 μm guard column for separation. The column temperature was maintained at 55 °C. For elution, a binary gradient program was used. Mobile phase A consisted of 7.4 mM phosphoric acid solution, and phase B was composed of 7.4 mM phosphoric acid/methanol/acetonitrile (4:4:3 by volume). This method was modified from Lobbes et al. The flow rate was set to 1 mL/min. The multistep gradient is shown in Table 3. Multiple injections were made automatically by the autosampler to ensure a sufficient quantity of phenols for radiocarbon measurements. Individual phenols were collected in 2 mL HPLC vials using the fraction collector autosampler.

In order to verify the composition and test compound purity, the individual phenols isolated from one sediment sample (FG80) were analyzed by gas chromatography/mass spectrometry (GC/MS). The solution in the 2 mL HPLC vial was transferred into an 8 mL vial, and 1 mL of 5% NaCl solution was added. One milliliter of dichloromethane was added to each vial. Phenols were extracted from the aqueous phase by shaking the 8 mL vials (vial 1) and allowing the two phases to separate. The water phase was transferred into a clean 8 mL vial (vial 2) using a pipet, and the dicholoromethane was transferred to a 4 mL vial. The residual water phase in vial 1 was combined with the water phase in vial 2 in order to repeat the extraction three times to maximize recovery of the organic extract. Water in the extracts was removed by addition of clean Na_2SO_4. The DCM was transferred into another 4 mL vial and was blown down by N_2. Thirty microliters of bis (trimethylsilyl) trifluoroacetamide (BSTFA) and 40 μL of pyridine were added to each vial. The vials were blown into nitrogen gas for 10 s to remove oxygen gas. The vials were capped tightly and were heated at 60 °C for 8 h. After the reaction, the solution was transferred into a new 2 mL GC vial for GC/MS analysis.

Table 3. Gradient of Mobile Phase during the HPLC Separation

time (minutes)	% mobile phase B
0	0
8	30
12	40
14	50
15	100
20	100
23	0

Radiocarbon Measurements. Following separation by HPLC, the phenols were extracted three times with DCM. Clean Na_2SO_4 was added to each vial to remove water. The DCM with the phenols was transferred into combustion tubes and blown to dryness using high-purity N_2. Then, the samples were converted to graphite and prepared for radiocarbon measurements at National Ocean Science Accelerator Mass Spectrometry Facility (NOSAMS). The adoption of AMS has successfully reduced the size of the sample required from grams to milligrams. Currently, the AMS community has been able to measure samples of micrograms. This is critical to this study, as the size of phenols separated from the sediments is small.

RESULTS AND DISCUSSION

Compound Identification. The chromatograms of phenol standards and lignin phenols derived from NIST wood samples and lake sediments are shown in Figure 1 The 10 phenol standards were clearly separated in about 20 min, and all peaks were highly symmetrical. The peak resolution factors of two peaks varied between 0.8 and 12.0 (Table 1). The separation took 20 min with the first standard phenol, 4-hydroxybenzoic acid, eluting after about 7 min, and the last compound eluting around 18 min.

In wood samples of FIRI A, FIRI D, and IAEA-C5, three major phenol compounds (PAL, SAD, and CAD) were separated using HPLC (Figure 1). Seven phenol compounds (PAL, VAD, SAD, VAL, CAD, SAL, and VON) were separated from FIRI H (Figure 1). The compositions of the phenols are consistent with those expected from the types of wood represented by the standards. The woody tissues of the gymnosperms FIRI A, FIRI D, and IAEA C-5 contain mainly guaicyl units and produce fewer types of lignin phenols than the angiosperm FIRI H.[12] The sediment samples

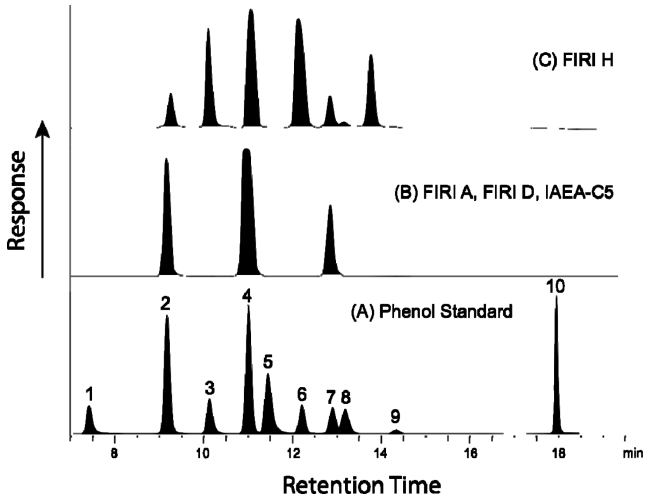

Figure 1. HPLC chromatogram of phenol standards (A) and wood standards, (B) FIRI A, FIRI D, and IAEA-C5 and (C) FIRI H. The phenol standards are numbered as listed in Table 1.

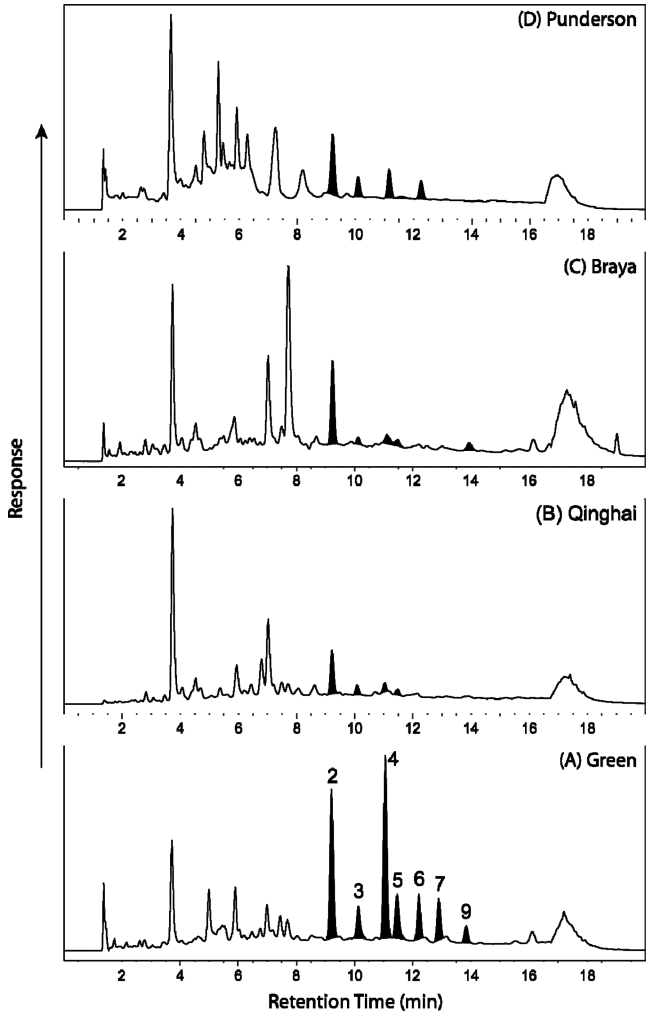

Figure 2. HPLC chromatogram of the lake sediments after microwave digestion: (A) Fayetteville Green Lake, New York, USA; (B) Lake Qinghai, China; (C) Braya Sø, Greenland; (D) Punderson Lake, Ohio, USA. The phenols are highlighted and numbered as the phenol standards in Figure 1.

show various numbers of phenol compounds (Figure 2). The lignin phenols from wood and sediment samples were identified

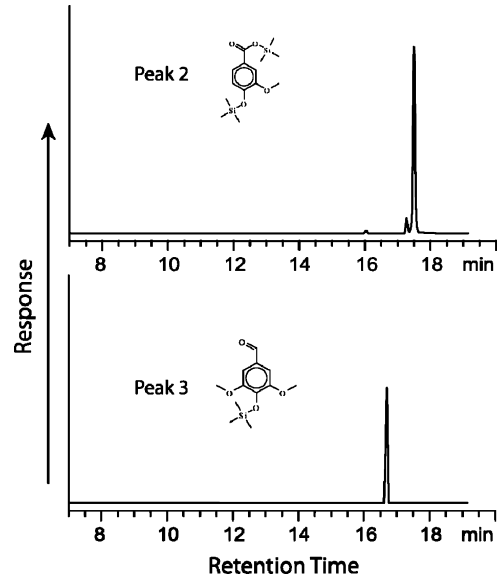

Figure 3. GC/MS chromatogram of peaks 2 and 3 (see Figure 3) from Fayetteville Green Lake. The small peak beside the major peak in peak 2 is an isomer of the PAL.

Table 4. CO_2 Yield (μmol) from Procedural Blanks (PB)

PB1	PB2	PB3	PB4	PB5	PB6	PB7	average	stdev
0.2	0.243	0.196	0.21	0.23	0.198	0.189	0.21	0.02

by comparing with the retention time of the standard phenols. In addition, the phenols from one of the sediment samples (FG-80) were further confirmed by gas chromatography/mass spectrometry (GC/MS) analysis. Figure 3 shows the mass spectra of two phenol compounds derived from the sediment sample from Fayetteville Green Lake, USA. The GC/MS chromatogram indicates the high purity of individual phenols isolated from the HPLC.

Procedural Blanks. A series of procedural blanks (PB) were measured in order to quantify the amount of CO_2 produced during the entire sample preparation process. The amount of CO_2 present in each blank represents the carbon that may have entered each sample and would affect the actual ^{14}C measurements. The CO_2 yields of the procedural blanks were listed in Table 4. The average yield of the CO_2 of the procedure blanks is 0.209 ± 0.02 μmol. The blank sample CO_2 is less than 10% of the CO_2 derived from the wood standards and sediment samples. (See Table 2 for the CO_2 yield from the samples.)

The precision of our analyses is ultimately limited by the process blank associated with sample handling relative to the size of the sample we isolate and measure.[24] For this study, we determined the amount of carbon in the process blank by performing the entire sample isolation procedure seven times with no sample included (0.21 ± 0.02; Table 4). We were not able to measure the fraction modern (fm) of the blank and have conservatively chosen a value of 0.30 ± 0.05 based on laboratory experience at NOSAMS (McNichol, unpublished data). Fraction modern is a measurement of the deviation of the $^{14}C/^{12}C$ ratio of a sample from "modern", where "modern" is defined as 95% of the radiocarbon concentration (in AD 1950) of NBS Oxalic Acid I normalized to $\delta^{13}C_{VPDB} = -19$ ‰.[25,26] It is certain that the fm

(24) Shah, S. R.; Pearson, A. *Radiocarbon* **2007**, *49*, 2175–2188.

value of the process blank will be between 0 and 1, and experience has shown that it is usually significantly below modern (i.e., fm = 1). We use a mass balance to calculate the fm of the sample from the measured value.

$$m_m = m_s + m_{pb} \quad (1)$$

$$m_m fm_m = m_s fm_s + m_{pb} fm_{pb} \quad (2)$$

The equations can be rewritten as

$$fm_s = r_m fm_m - (r_m - 1) fm_{pb} \quad (3)$$

where $r_m = m_m/m_s$. Error propagation leads to the following equation for the standard deviation, σ, in fm_s:

$$\sigma_{fm_s}^2 = r_m^2 \sigma_{fm_m}^2 + (1 - r_m)^2 \sigma_{fm_{pb}}^2 + (fm_m - fm_{pb})^2 \times \left[\sigma_{m_m}^2 \left(\frac{m_{pb}}{(m_m - m_{pb})^2} \right)^2 + \sigma_{m_{pb}}^2 \left(\frac{r_m}{(m_m - m_{pb})} \right)^2 \right] \quad (4)$$

NIST Wood Standards. We use the measurements on standard wood samples to evaluate our assumptions above. Corrected fraction modern (fm corr) using the equations (1−4) is adopted to express the ^{14}C contents of the samples (Table 2). Figure 4 shows the fm values we calculated for the individual lignin phenols isolated from the standard materials: FIRI H, FIRI D, FIRI A, and IAEA C-5.

The calculated fm values agree with the expected ones within the error we have calculated (Figure 4), although more data will be needed for a more robust statistical analysis. Different individual phenols isolated from FIRI D, FIRI H, and IEAE C5 have very similar fm values.For example, the average fm corr of phenols for FIRI H is 0.7540 ± 0.0029, which is very close to the known fm (0.7574). The oldest wood sample, FIRI A, had the largest offset from the expected ^{14}C value (expected fm = 0.0033, measured fm = 0.0152) with standard deviation of 0.0052 (Figure 4). This probably results from the fact that the FIRI-A is nearly radiocarbon free, even a small fraction of contaminants during the sample preparation would result in a relatively large offset.

Our results from wood standards suggest that the individual phenols can provide sufficiently accurate ^{14}C measurements for natural samples. Moreover, if the amount of individual phenols in natural samples is not in sufficient quantity for ^{14}C measurements, different phenols could be combined to provide a larger sample for ^{14}C measurements in order to minimize the measurement uncertainties.

Sediment Samples. The CO_2 yields of individual phenols from the sediment samples were relatively small for radiocarbon measurement. Therefore, the CO_2 from individual phenols were combined for ^{14}C measurements to minimize measurement errors (Table 2).

Steel Lake. Steel Lake is a small glacial lake located in north-central Minnesota. The chronology for the sediment core was

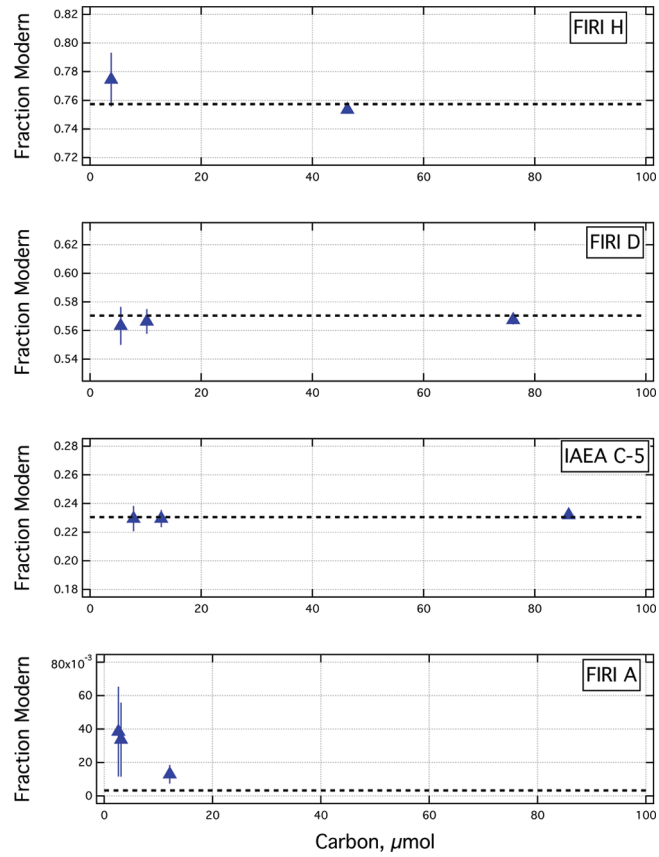

Figure 4. Comparison between measured fm (fraction modern, after correction for the procedure blank) for the individual lignin phenols isolated from the standard materials, FIRI H, FIRI D, FIRI A and IAEA C-5, and the known fm values. Note there are three separate phenol compounds (PAL, SAD, and CAD) in FIRI A, IAEA C5, and FIRI D and two phenol compounds (VAD and VAL) in FIRI H (Table 2). The dashed line in the figure represents the consensus values reported for the standards.

constructed on the basis of radiocarbon measurements on plant macrofossils and varve counting.[27] One ^{14}C measurement of phenol from Steel Lake is in principle consistent with the varve counting (Table 2). The calibrated age of the phenols at 121 cm is 557 ± 107 (between 502 and 654 yrBP). The calibrated age of plant macrofossil from the same depth is 703 ± 52 year, where the varve counting is 680 ± 29 year. The relatively large range associated with the calibrated phenol age results from the radiocarbon plateau effect, which is inherited in the radiocarbon date calibration.[28] The age error caused by radiocarbon plateau is unavoidable even though the measurement error of AMS ^{14}C dating is relatively small (Table 2).

Fayetteville Green Lake. Fayetteville Green Lake is a small, deep (maximum 52 m), meromictic lake in the state of New York, USA. In sediments, a dark lamina consisting of bacterial production in autumn and white lamina consisting of calcite deposition in spring are thought to form regularly each year. The sediment is, thus, considered varved, and a chronology based on varve counting has been developed (Dr. John King, unpublished data). The calibrated ages of the two sediment samples are consistent with varve

(25) Olsson, I. U. In *Radiocarbon Variations and Absolute Chronology*; Olsson, I. U., Ed.; John Wiley & Sons: New York, 1970; pp 17.
(26) Stuiver, M.; Polach, H. A. *Radiocarbon* **1977**, *19*, 355–363.
(27) Tian, J.; Brown, T. A.; Hu, F. S. *The Holocene* **2005**, *15*, 510–517.
(28) Niklaus, T. R.; Bonani, G.; Suter, M.; Wolfli, W. *Nucl. Instrum. Methods Phys. Res. B* **1994**, *92*, 194–200.

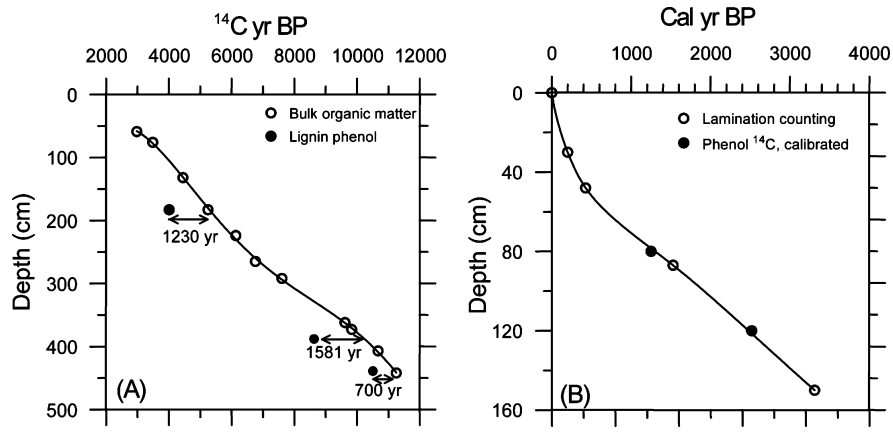

Figure 5. (A) Comparison between phenol ages and bulk organic matter ages for sediment samples from Lake Qinghai, China. Open circles represent the ^{14}C ages measured on bulk organic matter from sediment; solid dots represent mean ^{14}C ages measured on lignin phenols in this work. (B) Calibrated ^{14}C ages of lignin phenols (solid dots) are broadly consistent with varve counting (open circles) at the Fayetteville Green Lake, USA. Data for Lake Qinghai and Fayetteville Green Lake are given in Table 2 with errors.

counting ages (Table 2, Figure 5), despite of the relatively large errors.

Braya Sø. Braya Sø is a small lake in southwestern Greenland,[29] which is surrounded by permafrost peat land soil. The calibrated phenol ^{14}C ages for the two samples at 28 and 71 cm are, surprisingly, older than the calibrated bulk organic carbon ^{14}C age (Table 2). The data appear to contradict our hypothesis that the terrestrial-derived phenol ^{14}C should be younger than the bulk organic age due to the absence of aquatic old carbon effect. However, the temperature records derived from the alkenones in the sediment cores could help reconcile the problem.[30] Coincidentally, the two sediment samples are from relatively warm intervals in the past few thousand years. It is likely that warm conditions lead to increased recycling of the ancient peat carbon (preserved in permafrost horizons in soils) and transportation of old lignin phenol into the lake sediments.[29] Therefore, caution should be taken when attempting to date lake sediments in high arctic regions and other peat-rich settings using lignin phenols.

Lake Qinghai. Lake Qinghai is known to have significant reservoir age.[23] The ^{14}C age of dissolved inorganic carbon (DIC) in modern lake water is 1040 year.[23] However, it is not clear how the reservoir age may have changed in the past. The offset between bulk organic matter ^{14}C and lignin phenol ^{14}C measurements provides very important information on the past changes in reservoir ages. The ^{14}C ages of the phenols are consistently younger than the bulk organic carbon ^{14}C ages (Figure 5). Most importantly, the offset of the ^{14}C ages at the different sedimentary horizons varied significantly, from 700 to 1581 year in the late glacial and the Holocene. Therefore, it is difficult to achieve accurate sediment ages by subtracting a single assigned age offset obtained by measuring the DIC in modern lake water.[23]

Punderson Lake. Punderson Lake is a small lake in Ohio, USA. The phenols from six horizons at the sediment core have been radiocarbon dated in order to test the consistency of the lignin radiocarbon dates and obtain a reliable chronology (Table 2,

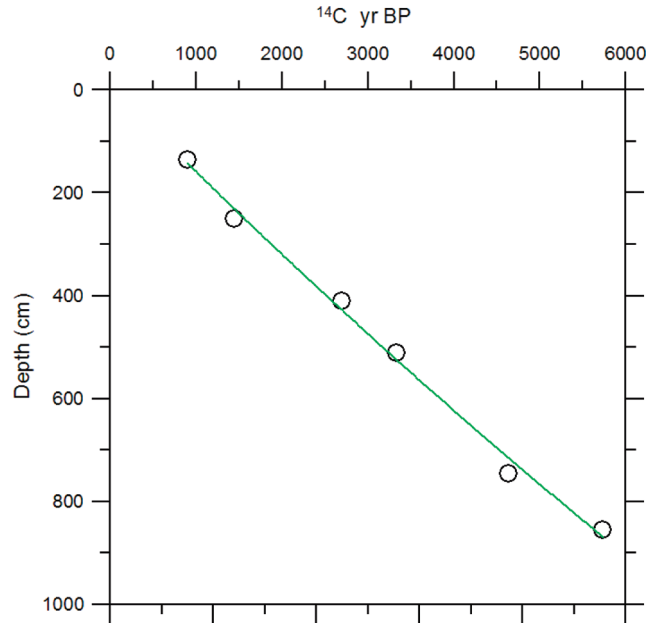

Figure 6. Chronology construction based on phenol ^{14}C ages for Lake Punderson, Ohio, USA.

Figure 6). The nearly linear depth-age relationship demonstrates the consistency of our lignin radiocarbon dating methods. The estimated sedimentation rate for this lake is 800 year/m, which is consistent with the average sedimentation rate of the regional lakes.[31] This demonstrates that the phenol ^{14}C measurements can provide a reliable chronology for lake sediment cores. Unfortunately, we do not, at present time, have varve counting ages in this sediment core for comparison with lignin ages.

CONCLUSIONS

Radiocarbon measurement of individual lignin phenol is an effective approach for dating late Quaternary lacustrine sediments. The HPLC separation of individual lignin phenols following CuO oxidation of sediment samples provides an efficient way to purify

(29) D'Andrea, W. J.; Huang, Y. *Org. Geochem.* **2005**, *36*, 1234–1241.
(30) D'Andrea, W. Ph.D. Dissertation, Brown University, Providence, RI, 2008.
(31) Shuman, B.; Finney, B. In *Encyclopedia of Quaternary Science*; Elias, S. A., Ed.; Elsevier: Burlington, MA, 2007, pp 1374−1383.

lignin components derived from terrestrial plants. In contrast to preparative gas chromatography, the HPLC separation does not require derivitization of the phenols as does PCGC, hence minimizing dating errors from isolation and purification procedures. The relatively large loading capacity (up to 30 μg of pure carbon) of HPLC also allows rapid isolation of compounds.

Ages of lignin phenols in several temperate lakes are consistent with varve counting or ages of terrestrial plant fragments, suggesting minimal residence time of lignin phenols in catchment soils. However, in an arctic lake in southwestern Greenland surrounded by permafrost soils, sediment lignin phenols are significantly older than bulk sediment ages, suggesting remobilization of preserved ancient lignin in soils. The long residence time of lignin in such systems could, therefore, compromise the use of lignin phenols for establishing sediment chronology.

Our results from Lake Qinghai suggest that reservoir age in the sediment core is not always constant with time. The conventional approach of subtracting a single reservoir age from bulk sediment ages in a lake with significant hard water effect may, thus, not provide the most robust age control for late Quaternary sediments.

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