ADVANCING EARTH AND SPACE SCIENCE

Global Biogeochemical Cycles

RESEARCH ARTICLE

10.1029/2017GB005854

Aging and Molecular Changes of Dissolved Organic Matter Between Two Deep Oceanic End-Members

S. K. Bercovici[1,2], **B. P. Koch**[3], **O. J. Lechtenfeld**[3,4], **S. L. McCallister**[5], **P. Schmitt-Kopplin**[6,7], **and D. A. Hansell**[1]

[1]Department of Ocean Sciences, RSMAS, University of Miami, Miami, FL, USA, [2]Polar Science Center, University of Washington, Seattle, WA, USA, [3]Alfred Wegener Institute, Helmholtz Center for Polar and Marine Research, Bremerhaven, Germany, [4]Department of Analytical Chemistry, UFZ-Helmholtz Centre for Environmental Research, Leipzig, Germany, [5]Department of Biology, Center for Environmental Studies, Virginia Commonwealth University, Richmond, VA, USA, [6]Helmholtz Zentrum München, Analytical BioGeoChemistry, Neuherberg, Germany, [7]Analytical Food Chemistry, Technische Universität Muenchen, Freising/Weihenstephan, Germany

Key Points:

- Solid-phase extracted DOM (SPE-DOM) from deep waters in the far North Pacific has a ^{14}C age of ~6,400 years
- Deep Pacific SPE-DOM has an intrinsically stable component
- Specific molecular formulae are largely preserved in SPE-DOM as it undergoes meridional overturning circulation

Supporting Information:

- Supporting Information S1

Correspondence to:
S. K. Bercovici,
sberco@uw.edu

Citation:
Bercovici, S. K., Koch, B. P., Lechtenfeld, O. J., McCallister, S. L., Schmitt-Kopplin, P., & Hansell, D. A. (2018). Ageing and molecular changes of dissolved organic matter between two deep oceanic end-members. *Global Biogeochemical Cycles*, *32*, 1449–1456. https://doi.org/10.1029/2017GB005854

Received 1 DEC 2017
Accepted 13 SEP 2018
Accepted article online 15 SEP 2018
Published online 3 OCT 2018

Abstract The global ocean contains a massive reservoir of dissolved organic carbon (DOC), rivaling the atmosphere's pool of CO_2. The most recalcitrant fractions have mean radiocarbon ages of ~4,000 years in the Atlantic to ~6,000 years in the Pacific. Knowing the radiocarbon signatures of DOC and the molecular composition of dissolved organic matter (DOM) is crucial to develop understanding of the persistence and lifetime of the DOC pool. In this research, we collected samples from the deep North Pacific in August 2013 (aboard the RV *Melville*) to couple the Δ^{14}C content of solid-phase-extracted DOM (Δ^{14}C-SPE-DOM) with its molecular composition in the ocean's oldest deep waters. We find that deep waters in this region held a mean Δ^{14}C-SPE-DOM value of -554 ± 9‰ (~6,400 ^{14}C years), substantially more depleted than that in the deep Atlantic, which held a mean Δ^{14}C-SPE-DOM value of -445 ± 5‰. While we find a more degraded molecular composition of DOM in the deep Pacific than the deep Atlantic, the molecular formulae within the Island of Stability (Lechtenfeld et al., 2014, https://doi.org/10.1016/j.gca.2013.11.009), are largely retained. These results imply that a fraction of deep DOM is resistant to removal and present in both the deep Atlantic and Pacific Oceans.

1. Introduction

A major fraction of dissolved organic matter (DOM) is produced in the surface ocean via primary production where it is either directly released into the water column or solubilized via heterotrophic consumption of particulate organic material (Carlson & Hansell, 2014). Although algal-derived DOM is produced in the euphotic zone, >95% of DOM is contained in the dark ocean, where it survives multiple meridional overturning circulations (Druffel et al., 1992). However, the turnover time and overall biogeochemical fate of this large, deep pool are largely unknown. In this analysis, we investigate the cycling of the deep pool of marine DOM using isotopic and compositional signatures.

The concentration of dissolved organic carbon (DOC; the carbon component of DOM) varies across the deep ocean basins. DOC concentrations are highest at depth (48 μmol/kg) in the North Atlantic near the formation of North Atlantic Deep Water, due to the surface subtropical ocean source of those waters (Hansell et al., 2009; Hansell & Carlson, 1998). Deep waters reach ~40 μmol/kg in the Southern and Indian Oceans (Bercovici & Hansell, 2016; Ogawa et al., 1999) and ~34–37 μmol/kg in the deep Pacific (Hansell, 2013; Hansell et al., 2009).

While early studies suggest that the deep DOC concentration gradient resulted from slow removal over time (Hansell & Carlson, 1998), conserved mixing and localized sinks are now considered to play a role in establishing this gradient as well (Bercovici & Hansell, 2016; Hansell et al., 2009; Hansell, 2013; Hawkes et al., 2015; Lang et al., 2006). The difference in ^{14}C age of DOM between the deep North Atlantic and the deep North Pacific is ~2,000 years (Druffel et al., 1992). Apart from transit time and related radiocarbon decay along the deep ocean conveyer belt (Bercovici et al., 2018), an additional aspect that would contribute to this difference in ^{14}C age is the preferential removal of modern components (Beaupré & Aluwihare, 2010; Druffel et al., 1992; Hansell & Carlson, 2012, Lechtenfeld et al., 2014; Williams & Druffel, 1987). The DOM pool is dynamic, consisting of multiple molecular components cycling on different timescales (Follett et al., 2014; Loh et al., 2004;

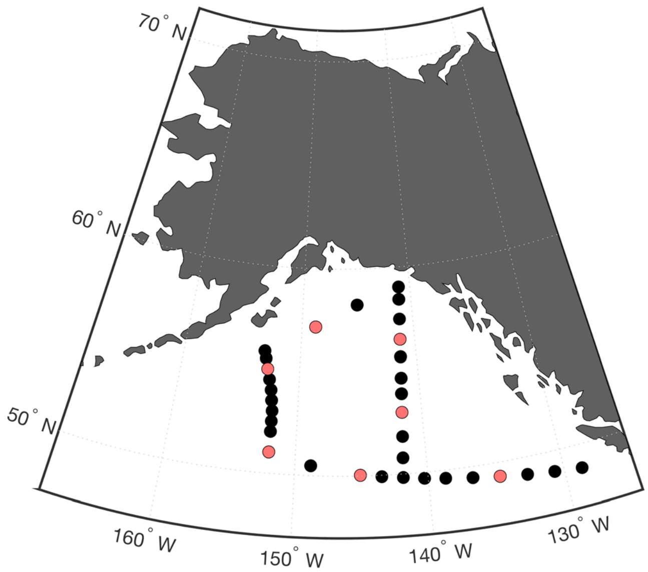

Figure 1. Map of station occupations for the *MV-1310* cruise. The pink dots represent where samples were collected for this analysis.

Repeta & Aluwihare, 2006; Walker et al., 2011; Zigah et al., 2017). Observations suggest that small, dissolved molecules are the most highly aged components of DOM and have much older radiocarbon ages than the larger components of the pool (Broek et al., 2017; Loh et al., 2004; Walker et al., 2011; Walker, Primeau, et al., 2016; Walker, Beaupré, et al., 2016). A recent study by Zigah et al. (2017) shows that components of DOM separated by size and polarity have varied isotopic signatures, ranging from modern to ancient ($>$10,000 years).

Recently, the analytical capabilities of Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR-MS) allow for advances in our understanding of the relationship between molecular composition of solid-phase extracted DOM (SPE-DOM) and its Δ^{14}C content (Δ^{14}C-SPE-DOM). Two prior studies spanning the Atlantic Ocean provided a comprehensive data set that coupled radiocarbon and molecular composition (Flerus et al., 2012; Lechtenfeld et al., 2014). These studies isolated the specific formulae present in each FT-ICR-MS spectrum that had a significant positive or negative correlation with Δ^{14}C-SPE-DOC ($p < 0.05$, $r > 0.65$); these formulae represent ~61% of the summed magnitudes of all peaks in the spectrum (Flerus et al., 2012). Lechtenfeld et al. (2014) extended this analysis via a first-order degradation between radiocarbon age and individual molecular formulae. They suggested that a portion of DOM molecules have a much longer residence time in the water column than the age of the bulk DOC pool would otherwise indicate. Using those molecular formulae that hold longer residence times within the whole SPE-DOM pool, Lechtenfeld et al. identified ~361 distinct molecular formulae that were persistent in virtually all samples (island of stability [IOS]), representing the most stable fraction in DOM.

In this study, we couple molecular composition of SPE-DOM with its Δ^{14}C values to determine the intrinsic stability of SPE-DOM in the far North Pacific, where deep bulk DOC concentrations and SPE-Δ^{14}C-DOM values are at their lowest. In addition, we compare the molecular composition of deep SPE-DOM from two distinctly different ocean basins: in the North Atlantic, deep waters have recently been formed and ventilated; in the North Pacific, deep waters have been out of contact with the atmosphere for centuries. This comparison of the DOM between these two end-members provides a deeper understanding of the intrinsic stability of deep DOM on a global scale.

2. Methods

Samples were collected on a cruise aboard the *R.V. Melville* (MV-1310) in August 2013 (Figure 1). In the following sections, we describe the methods for each variable discussed in this manuscript.

2.1. Total Organic Carbon Measurements

Total organic carbon (TOC) is composed of suspended particulate organic carbon (POC) and DOC. DOC is operationally defined as passing through a pore size of 0.2 to 0.7 μm. Here TOC samples were collected directly from Niskin bottles into acid-cleaned (10% HCl) 60-ml polycarbonate bottles. TOC was measured by high-temperature combustion using a Shimadzu TOC-L with auto-injection (precision of ±1.5 μmol/kg; Dickson et al., 2007). Measurements were quality controlled using Consensus Reference Material (CRM), distributed to the international oceanographic community by the Hansell laboratory. Low C and N reference water was employed to determine system blanks. Hereafter, TOC samples are referred to as DOC, as POC accounts for a minimal portion of TOC (Hansell & Carlson, 2001; further information is described in section 3.2).

2.2. Sampling for Solid Phase Extraction (SPE)

Water samples for SPE were collected with a conductivity-temperature-depth water rosette sampler at 5, 50, 1,000, 2,000, 3,000, and a few meters above the seafloor. First, 8 L of each sample were filtered through a 0.2 μm Supor filter and acidified to pH 2 with high purity hydrochloric acid. Mega Bond Elut PPL (1 g, Varian) cartridges were used for SPE following the protocol of Dittmar et al. (2008). Following the

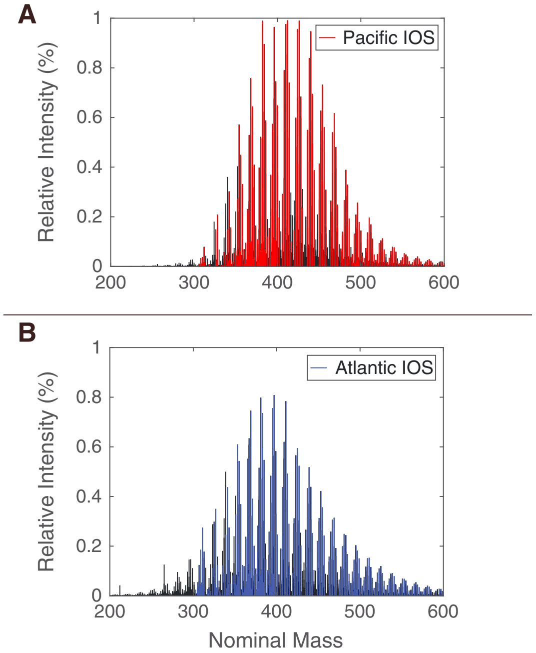

Figure 2. Fourier transform ion cyclotron resonance mass spectrometry mass spectra of (a) a deep Pacific and (b) a deep Atlantic sample. The red and blue highlighted peaks represent those in the island of stability (IOS).

extraction, the cartridges were rinsed with two cartridge-full volumes (~8 ml) of acidified milliQ (pH 2) and subsequently dried with ultra-pure nitrogen gas. The SPE samples (SPE-DOM) were eluted in 8 ml of methanol and stored at −20 °C until analysis.

To measure the amount of DOC recovered, a 150 μl aliquote of the SPE eluate (V_{EL}) was added to a combusted glass vial, dried completely with ultra-pure nitrogen gas, left to dry overnight, and resuspended in 15 ml of milliQ. The resultant sample was then measured for DOC following the method described in section 2.1. Extraction efficiencies (EEs) for DOC were calculated using following formula:

$$\mathrm{EE} = \left(\frac{\mathrm{DOC}_{\mathrm{Ex}}(\mu\mathrm{M})}{\mathrm{DOC}_{\mathrm{orig}}(\mu\mathrm{M})}\right) * \frac{V_{\mathrm{mQ}}(\mathrm{mL})}{V_{\mathrm{EL}}(\mathrm{mL})} * \frac{V_{\mathrm{E}}(\mathrm{mL})}{V_{\mathrm{S}}(\mathrm{mL})} * 100\% \tag{1}$$

where $\mathrm{DOC}_{\mathrm{Ex}}$ is the DOC concentration of the extract, $\mathrm{DOC}_{\mathrm{orig}}$ is the DOC concentration of the original water sample, V_{mQ} is the volume of milliQ water added to the dried eluate, V_{E} is the volume of the total eluate, V_{EL} is the volume of the eluate used for analysis, and V_{S} is the total sample volume that was enriched using solid-phase extraction.

2.3. FT-ICR MS

SPE-DOM samples were analyzed in the Analytical Biogeochemistry Laboratory at the Helmholtz Zentrum München (Munich, Germany) using a FT-ICR-MS (SolariX, Bruker Daltonics, Billerica, MA), equipped with a 12 T superconducting magnet (Magnex Scientific Inc., Yarnton, GB). An Apollo II dual electrospray source (ESI, Bruker) was used in negative ion mode (capillary voltage: −4200 V, infusion flow rate: 2 μl/min). Prior to measurement on the FT-ICR MS, extracts were diluted with methanol to obtain the same SPE-DOM concentration for all aliquots. High-resolution mass spectra were acquired for the mass range of 147 to 2,000 *m/z* with 512 scans per spectrum. Spectra were externally calibrated with arginine clusters, which are charged solute ions with established *m/z* values (Meng & Fenn, 1991).

Ions were singly charged and spectra were evaluated in the range of 200 to 600 *m/z* according to prior studies (Flerus et al., 2012; Lechtenfeld et al., 2014). Molecular formulae were calculated from the *m/z* values with the same algorithm as described in Lechtenfeld et al. (2014). Relative peak magnitudes were normalized by dividing the mass peak magnitude by the summed magnitude of all mass peaks in that sample. Only mass peaks having a normalized magnitude $>$0.005% were considered in this analysis. An initial quality control was conducted to remove surfactants. The source and quantity of these surfactants are largely unknown, but they are easily ionized and thus have been previously identified in marine FT-ICR-MS samples (Lechtenfeld et al., 2013). While the analytical methods in this study were similar to previous studies (Flerus et al., 2012; Lechtenfeld et al., 2014), a direct comparison between the two data sets might be affected by instrument conditions, as evidenced by a slight shift in the mass distribution of the spectra (Figure 2). To compare the spectra from the Pacific and the Atlantic, we normalized the FT-ICR-MS data; the sum of the magnitudes of all molecular formulae in a given sample is set to equal 1, and the relative magnitude of each individual formula is described as a percentage of the total magnitude.

2.4. ^{14}C Measurements

Aliquots of the methanol extracts (~0.15 ml) were transferred to precombusted Pyrex tubes (6 mm diameter). The contents of each tube were transferred to 1 cm diameter quartz tubes, from which the solvent was removed by vacuum. Each sample was washed with 1 ml of ultrapure water (milliQ), dried and flame sealed under vacuum and combusted to CO_2 at 750 °C with a CuO/Ag catalyst (Sofer, 1980). The CO_2 was subsequently reduced with H_2 to graphite over a Co catalyst at the National Ocean Sciences Accelerator Mass Spectrometer (NOSAMS) facility at Woods Hole Oceanographic Institution (Woods Hole, MA). SPE DOC blanks did not yield any detectable amount of carbon ($<$1 nM C). Radiocarbon ages of SPE-DOM were derived from their respective Δ^{14}C-DOM values following Stuiver and Polach (1977).

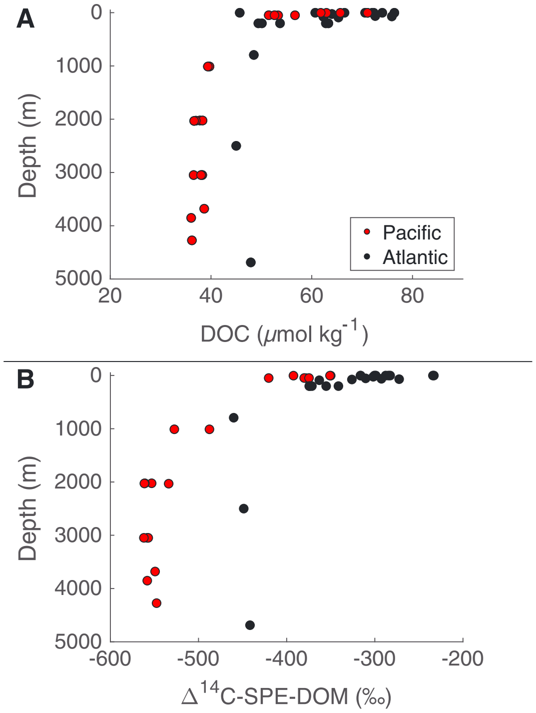

Figure 3. (a) Dissolved organic carbon (DOC) concentration and (b) Δ^{14}C-SPE-DOM versus depth in the Pacific (red; data from this cruise) and the Atlantic (black; data from Flerus et al., 2012).

3. Results and Discussion

3.1. EE of SPE-DOM

The EE of the SPE-DOM (equation (1)) was 41 ± 10% and independent of depth and DOC concentration. The SPE extraction method for these Pacific samples was slightly different than that used in the Atlantic data employed here. In this study, we extracted 8 L of seawater through 1 g PPL cartridges, whereas previous studies extracted 5 L of seawater (Flerus et al., 2012; Lechtenfeld et al., 2014). However, the EE in the previous studies (42 ± 7%) is effectively the same as here and within the same range (~40%) of other published SPE EEs of marine DOM (Coppola et al., 2015; Dittmar et al., 2008).

3.2. Bulk DOC Concentrations and Δ^{14}C-SPE-DOM Values

Bulk DOC concentrations for samples in the far North Pacific ranged from 36 to 71 μmol/kg (Figure 3a). Concentrations were highest in the surface layer, varying from 51 to 71 μmol/kg (59 ± 7 μmol/kg; depths ≤50 m, *n* = 8; Figure 3a), as compared to subsurface concentrations (depths ≥1,000 m, *n* = 10; Figure 3a), with a much narrower range of 36 to 39 μmol/kg (37 ± 1 μmol/kg; Figure 3a). Although DOC data here are unfiltered (see section 2.1), there is typically a minor contribution of POC to the total pool (1 to 4%; Hansell & Carlson, 2001). Given that the concentrations range from 51 to 71 μM C in our surface samples, a 1–4% contribution of POC would imply an addition of ~0.5–3 μM C, similar to the precision of the TOC measurement. Furthermore, at depths >1,000 m, where this study focuses, there is no resolvable difference between filtered and unfiltered DOC measurements (Hansell & Carlson, 2001).

In the Pacific dataset, the mean Δ^{14}C-SPE-DOM value at the surface (defined here as depths <100 m) was −370 ± 30‰ (~3,700 ^{14}C years). These data are more depleted in ^{14}C than those in the Atlantic (−293 ± 34‰; ~2,800 ^{14}C years; Flerus et al., 2012). Furthermore, the surface values reported here are more depleted in ^{14}C than bulk Δ^{14}C-DOC surface values, which range from −180 to −260‰ in the Atlantic, Pacific, and Indian Ocean basins (Bercovici et al., 2018; Druffel et al., 1992, 2016; Druffel & Griffin, 2015; Follett et al., 2014; Walker et al., 2011, 2016; Zigah et al., 2017). However, our results are similar to those Δ^{14}C-SPE-DOM values reported by Coppola et al (2015) collected in the East Pacific (−318 ± 11‰ and −335 ± 7‰).

The offset between the surface values of the Δ^{14}C-SPE-DOM data from this Pacific data set and others could be due to several factors. Published Δ^{14}C-SPE-DOM data is scarce, and there is variability between experimental and instrumental procedures in between studies. In addition, it is impossible to entirely rule out contamination and methylation in the extraction procedure; however, the impact of methylation was kept to a minimum by cold storage of the extracts (Flerus et al., 2011). In addition, hydrography of the contrasting sampling regions is important to consider. The mean DOC concentration in the surface Atlantic data is 67 ± 8 μmol/kg (Flerus et al., 2012), whereas that in the surface Pacific was generally lower at 59 ± 7 μmol/kg. According to National Aeronautics and Space Administration (NASA) Ocean Color, the surface chlorophyll in November 2008 (when the Flerus et al. Atlantic samples were collected) reached up to 10 mg/L near the African coast, whereas the surface chlorophyll in August 2013 (when the Pacific samples here were collected) reached ~0.5 mg/L. The lower chlorophyll and the lower DOC concentrations between the two ocean basins indicate that there was likely lower primary productivity in our Pacific sites than in the Atlantic (Flerus et al., 2012).

At depth, the Δ^{14}C-SPE-DOM values reported in the Pacific are −554 ± 9‰ (~6,400 ^{14}C years), whereas those from the Atlantic data set were 445 ± 5‰ (~4,700 ^{14}C years; Table 1 and Figure 3b; Flerus et al., 2012). This offset between the deep Atlantic and Pacific of ~1,700 years is similar to the ~2,000-year offset depicted in the first bulk radiocarbon profiles comparing the Atlantic and Pacific Oceans (Druffel et al., 1992). Furthermore, the Δ^{14}C-SPE-DOM data from the deep Pacific (Table S1) are similar to those Δ^{14}C data of

Table 1

Mean (±Standard Deviation) Nominal Mass, O/C Ratio, H/C Ratio, Δ^{14}C-SPE-DOM Value (With Corresponding ^{14}C Age in Parentheses), and Relative Proportion of the IOS$_A$ in the Deep Atlantic and Pacific SPE-DOM Samples

	Nominal mass (Da)	O/C	H/C	Δ^{14}C-SPE-DOM (‰)	Relative proportion of IOS$_A$
Deep Atlantic	418 ± 99	0.48 ± 0.14	1.24 ± 0.22	−445 ± 5‰ (~4,700 years)	49 ± 2%
Deep Pacific	421 ± 88	0.49 ± 0.13	1.25 ± 0.21	−554 ± 9‰ (~6,400 years)	61 ± 2%

DOC taken from whole water in the deep Pacific (−501 to −564‰; Druffel et al., 1992, Druffel & Griffin, 2015, Walker et al., 2011; Walker, Beaupré, et al., 2016; Walker, Primeau, et al., 2016; Zigah et al., 2017).

While our Δ^{14}C-SPE-DOM data at depth are similar to the whole water in the Pacific, it is known that there is some variability between bulk Δ^{14}C-DOC and Δ^{14}C-SPE-DOM values. Coppola et al. (2015) reported that Δ^{14}C-SPE-DOM data from marine samples were equal to or lower than that of the Δ^{14}C of the total DOC pool. A fraction of the most polar (and presumably labile) portion of the DOM is not recovered with extraction (e.g., molecules such as acetate). Furthermore, in the Bauer et al. (1992) comparison of bulk DOC versus SPE-DOM, DOM extracted on the XAD resin was older than the ^{14}C age of the bulk DOM.

3.3. FT-ICR-MS Analysis

From the FT-ICR-MS spectra (Figure 2a), a total of 2,726 molecular formulae were identified, with 2,154 of these formulae present in all samples. The magnitude average nominal mass between each sample was 421 ± 3 Da, and the average O/C and H/C ratios were 0.49 ± 0.13 and 1.25 ± 0.21, respectively (Table 1). These values are similar to the deep Atlantic, where the magnitude average nominal mass was 418 ± 1 Da (Flerus et al., 2012), and the average O/C and H/C ratios were 0.48 ± 0.14 and 1.24 ± 0.22, respectively (Table 1). It is important to note that the assigned molecular formulae do not provide information on structural diversity, as different structural isomers can share the same molecular formula.

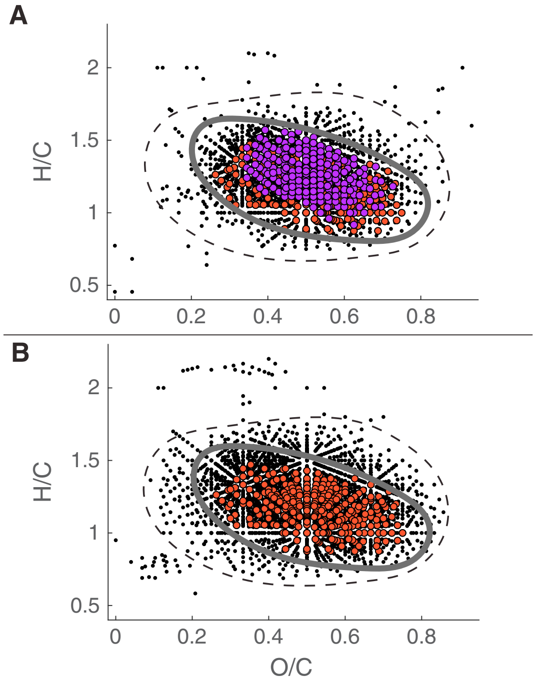

Figure 4. Van Krevelen plots of (a) the deep Pacific and (b) the deep Atlantic, where the *x* axis is the oxygen to carbon ratio (O/C) and the *y* axis is the hydrogen to carbon ratio (H/C). The orange and purple colored dots represent the IOS$_A$ and IOS$_P$, respectively. The grey circle represents a general outline of the carboxyl-rich alicyclic molecule regions in van Krevelen space, and the dashed black line is a reference around the majority of the molecular formulae present in the deep Atlantic van Krevelen plot to show the decrease in molecular formulae from the deep Atlantic to the deep Pacific. The relative magnitude of island of stability (IOS) compounds increases 10% from the deep Atlantic (IOS$_A$ = ~49% of total; Lechtenfeld et al., 2014) to the deep Pacific (IOS$_A$ = 60% of the total).

3.4. Extension of the IOS

The IOS is a subset of molecular formulae found in all samples in the deep Atlantic (IOS$_A$) and deemed intrinsically stable due to their positive correlation with ^{14}C age (Lechtenfeld et al., 2014). We found that virtually all of the formulae present in the IOS$_A$ in the deep Atlantic were present in the deep Pacific (Figures 4a and 4b, blue circles). In the deep Pacific spectra, the mean molecular weight of the IOS$_A$ was 439 ± 79 Da, similar to the average molecular weight of all molecular formulae (421 ± 88 Da; Tables 1 and 2 and Figure 2). In addition, the O/C and H/C ratios for the IOS$_A$ were 0.5 ± 0.1 and 1.2 ± 0.2, respectively, thus occupying a narrower O/C and H/C range than the total distribution of peaks (Table 2 and Figure 4a).

To assess whether the intrinsically stable molecular formulae in the deep Pacific are similar in character to those in the Atlantic, we used the same approach as in Lechtenfeld et al. (2014) to calculate a distinct set of formulae for an IOS in the Pacific (termed here as IOS$_P$). The IOS$_P$ formulae were isolated as those molecular formulae with high correlations ($r > 0.65$) with observed gradients in Δ^{14}C-SPE-DOM (termed here as IOS$_P$). Examples of molecular formulae with high and low correlations with observed gradients in Δ^{14}C-SPE-DOM are in Figure S1. There were 217 molecular formulae in the IOS$_P$, with a mean molecular weight of 438 ± 53 Da, an O/C ratio of 0.48 ± 0.06, and an H/C ratio of 1.23 ± 0.12 (Table 2 and Figure 4a, red circles).

It is important to address our limitations in this analysis: each molecular formula within SPE-DOM represents a multitude of different compounds that can vary in both Δ^{14}C content and ionization efficiency in the FT-

ICR-MS analysis. As a result, correlating Δ^{14}C-SPE-DOM with mass peak magnitudes of molecular formulae within an FT-ICR mass spectrum introduces an analytical uncertainty. However, we observe a strong correlation between the peak magnitude of specific IOS formulas with Δ^{14}C-SPE-DOM values in different ocean basins. Therefore, it is likely that the observed mass peak magnitude of IOS formulae is predominated by the chemical structures that contribute to their persistence. Moreover, normalizing the data is a way of looking at the relative intensity of a molecular formulae within a given sample, and how that relative intensity would change between samples.

Table 2
Mean Nominal Mass, O/C Ratio, and H/C Ratio for the IOS Constructed in the Atlantic (IOS$_A$) and Pacific (IOS$_P$)

	Nominal mass (Da)	O/C	H/C
IOS$_A$	439 ± 79	0.46 ± 0.10	1.24 ± 0.17
IOS$_P$	438 ± 53	0.48 ± 0.06	1.23 ± 0.12

A key to assessing the degradation of DOM in its transit along the deep ocean's conveyer is to compare the relative proportion of IOS$_A$ in both data sets. The similar molecular character between IOS$_A$ and IOS$_P$ is evident in the visual representation in the van Krevelen diagram (Figure 4a; IOS$_A$ is in orange dots; IOS$_P$ is in purple dots). For one, both IOS$_A$ and IOS$_P$ formulae have a clear distribution within the formulae associated with carboxyl-rich alicyclic molecules (Hertkorn et al., 2006; Figure 4, black outline). These alicyclic groups have been found ubiquitously in marine (Arakawa et al., 2017; Hertkorn et al., 2006, 2013) and freshwater systems (Arakawa & Aluwihare, 2015; Lam et al., 2007).

Additionally, while the molecular formulae in the IOS$_A$ and IOS$_P$ have the same average molecular weights, O/C ratios, and H/C ratios, the IOS$_P$ consists of an even more narrow range of formulae than the IOS$_A$ (Figure 4a and Table 2). We find that the proportion of IOS$_A$ in the deep Pacific spectra comprises a larger portion (61 ± 2%) of the SPE-DOM than in the deep Atlantic spectra (~49 ± 3%; Lechtenfeld et al., 2014; Figure 4). This distinct difference in the relative proportion of IOS$_A$ in each deep ocean basin shows that there are more diverse molecular formulae in the Atlantic (and thus, the IOS$_A$ makes up a smaller proportion of the total DOM). In the Pacific, diversity is reduced due to removal of molecular formulae beyond detectable levels (and thus the IOS$_A$ makes up a larger proportion of the total DOM). Contrasting the molecular composition between these two basins indicates that as DOC is transported through the deep ocean conveyer belt, compounds that are not intrinsically stable are removed relative to molecular formulae in the IOS.

DOM in the deep Atlantic carries with it a surface signature reflecting Atlantic Meridional Overturning, in which surface-derived DOC is sequestered into newly formed deep waters from the North Atlantic (Hansell et al., 2009). This condition is indicated by the higher DOC concentrations (Hansell & Carlson, 1998; Hansell et al., 2009), younger radiocarbon ages (Druffel et al., 1992, 2016; Druffel & Griffin, 2015), and a greater diversity of molecular formulae (Flerus et al., 2012; Lechtenfeld et al., 2014). In the far North Pacific, deep waters have been out of contact with the atmosphere for centuries (Matsumoto, 2007; Stuiver et al., 1983). As such, less stable compounds in the DOC pool have been removed, and what remains is an intrinsically recalcitrant core of molecular formulae (i.e., those similar in elemental ratios as the IOS), which has residence times greater than the timescales of meridional overturning circulation, explaining why they are in larger proportion to the whole pool.

A recent report suggests that dilution is what primarily controls DOM dynamics in the deep ocean (Arrieta et al., 2015). While dilution may limit microbial response to DOM (i.e., there is not a high enough concentration of a specific substrate), our results demonstrate that the long-term stability of DOM is related to its elemental composition. These results are similar to those concluded by Walker, Beaupré, et al., (2016) and Walker, Primeau, et al., (2016), who used ^{14}C dating of ultra-filtered DOM to suggest that molecular size and composition are important factors in determining the long term storage of DOC in the deep. It is possible that dilution acts to limit the removal of compounds in DOM that are structurally more biologically available. We show that there are molecular formulae in DOM with a certain compositional character that are resistant to removal over long timescales. This concept, where refractory DOC is controlled by both the concentrations and molecular character of individual molecules (Jiao et al. 2014) is illustrated here through using molecular composition and Δ^{14}C values of SPE-DOM.

4. Summary

Here we find that the Δ^{14}C content of SPE-DOM in the Pacific is more deplete than in the deep Atlantic by −109 ± 10‰ (or ~1,700 ^{14}C years older) and that its molecular composition is more degraded.

Consequently, while intrinsically stable molecular formulae (the IOS) are present ubiquitously throughout the ocean, they make up a higher relative proportion of DOM in the deep waters in the Pacific than in the Atlantic. These results show that a portion of deep DOM is intrinsically stable and is present in both Atlantic and Pacific deep waters.

Acknowledgments
This work was supported by U.S. NSF OCE-1153930. We thank the science party and crew of MV1310 aboard the *R. V. Melville* in August 2013. We thank the staff of the National Ocean Sciences Accelerator Mass Spectrometer at Woods Hole Oceanographic Institute for their analyses of these radiocarbon samples, Meredith Jennings for the DOC data, Ivan Dubinenkov for his assistance in preparing and running these samples, and the Analytical BioGeoChemistry Lab at Helmholtz Zentrum München for their assistance in the FT-ICR-MS analysis. The hydrographic and DOC data used here are publicly available at https://www.bco-dmo.org/project/523281. The Δ^{14}C-SPE-DOM values are available in the supporting information, and the FT-ICR-MS data are publicly available on PANGAEA Data Publisher for Earth and Environmental Science (https://doi.pangaea.de/10.1594/PANGAEA.893426).

References

Arakawa, N., & Aluwihare, L. (2015). Direct identification of diverse alicyclic terpenoids in Suwannee River Fulvic Acid. *Environmental Science & Technology*, *49*(7), 4097–4105. https://doi.org/10.1021/es5055176

Arakawa, N., Aluwihare, L. I., Simpson, A. J., Soong, R., Stephens, B. M., & Lane-Coplen, D. (2017). Carotenoids are the likely precursor of a significant fraction of marine dissolved organic matter. *Science Advances*, *3*(9), e1602976. https://doi.org/10.1126/sciadv.1602976

Arrieta, J. M., Mayol, E., Hansman, R. L., Herndl, G. J., Dittmar, T., & Duarte, C. M. (2015). Dilution limits dissolved organic carbon utilization in the deep ocean. *Science*, *348*(6232), 331–333. https://doi.org/10.1126/science.1258955

Bauer, J. E., Williams, P. M., & Druffel, E. R. M. (1992). C-14 activity of dissolved organic carbon fractions in the north- central Pacific and Sargasso Sea. *Nature*, *357*(6380), 667–670. https://doi.org/10.1038/357667a0

Beaupré, S. R., & Aluwihare, L. (2010). Constraining the 2- component model of marine dissolved organic radiocarbon. *Deep Sea Research, Part II*, *57*(16), 1494–1503. https://doi.org/10.1016/j.dsr2.2010.02.017

Bercovici, S. K., & Hansell, D. A. (2016). Dissolved organic carbon in the deep Southern Ocean: Local versus distant controls. *Global Biogeochemical Cycles*, *30*, 350–360. https://doi.org/10.1002/2015GB005252

Bercovici, S. K., McNichol, A. P., Xu, L., & Hansell, D. A. (2018). Radiocarbon content of dissolved organic carbon in the South Indian Ocean. *Geophysical Research Letters*, *45*, 872–879. https://doi.org/10.1002/2017GL076295

Broek, T. A. B., Walker, B. D., Guilderson, T. P., & McCarthy, M. D. (2017). Coupled ultrafiltration and sold phase extraction approach for the targeted study of semi-labile high molecular weight and refractory low molecular weight dissolved organic matter. *Marine Chemistry*, *194*(20), 146–157. https://doi.org/10.1016/j.marchem.2017.06.007

Carlson, C. A., & Hansell, D. A. (Eds.) (2014). *Biogeochemistry of marine dissolved organic matter* (2nd ed., 712 pp.). Academic Press. https://doi.org/10.1016/C2012-0-02714-7

Coppola, A. I., Walker, B. D., & Druffel, E. R. M. (2015). Solid phase extraction method for the study of black carbon cycling in dissolved organic carbon using radiocarbon. *Marine Chemistry*, *177*, 697–705. https://doi.org/10.1016/j.marchem.2015.10.010

Dickson, A. G., Sabine, C. L., & Christian, J. R. (Eds.) (2007). *Guide to best practices for ocean CO$_2$ measurements*, *PICES Special Publication* (Vol. 3, p. 191). Sidney, British Columbia: North Pacific Marine Science Organization.

Dittmar, T., Koch, B. P., Hertkorn, N., & Kattner, G. (2008). A simple and efficient method for the solid-phase extraction of dissolved organic matter (SPE-DOM) from seawater. *Limnology and Oceanography: Methods*, *6*, 230–235.

Druffel, E. R. M., & Griffin, S. (2015). Radiocarbon in dissolved organic carbon of the South Pacific Ocean. *Geophysical Research Letters*, *42*, 4096–4101. https://doi.org/10.1002/2015GL063764

Druffel, E. R. M., Griffin, S., Coppola, A. I., & Walker, B. D. (2016). Radiocarbon in dissolved organic carbon of the Atlantic Ocean. *Geophysical Research Letters*, *43*, 5279–5286. https://doi.org/10.1002/2016GL068746

Druffel, E. R. M., Williams, P. M., Bauer, J. E., & Ertel, J. R. (1992). Cycling of dissolved and particulate organic matter in the open ocean. *Journal of Geophysical Research*, *97*(C10), 15,639–15,659. https://doi.org/10.1029/92JC01511

Flerus, R., Koch, B. P., Schmitt-Kopplin, P., Witt, M., & Kattner, G. (2011). Molecular level investigation of reactions between dissolved organic matter and extraction solvents using FT-ICR MS. *Marine Chemistry*, *124*(1-4), 100–107. https://doi.org/10.1016/j.marchem.2010.12.006

Flerus, R., Lechtenfeld, O. J., Koch, B. P., McCallister, S. L., Schmitt-Kopplin, P., Benner, R., et al. (2012). A molecular perspective on the ageing of marine dissolved organic matter. *Biogeosciences*, *9*, 935–1955.

Follett, C. L., Repeta, D. J., Rothman, D. H., Xu, L., & Santinelli, C. (2014). Hidden cycle of dissolved organic carbon in the deep ocean. *Proceedings of the National Academy of Sciences of the United States of America*, *111*(47), 16,706–16,711. https://doi.org/10.1073/pnas.1407445111

Hansell, D. A. (2013). Recalcitrant dissolved organic carbon fractions. *Annual Review of Marine Science*, *5*(1), 421–445. https://doi.org/10.1146/annurev-marine-120710-100757

Hansell, D. A., & Carlson, C. A. (1998). Deep ocean gradients in dissolved organic carbon concentrations. *Nature*, 263–266.

Hansell, D. A., & Carlson, C. A. (2001). Biogeochemistry of total organic carbon and nitrogen in the Sargasso Sea: Control by convective overturn. *Deep Sea Research, Part II*, *48*(8–9), 1649–1667. https://doi.org/10.1016/S0967-0645(00)00153-3

Hansell, D. A., & Carlson, C. A. (2012). Net removal of major marine dissolved organic carbon fractions in the subsurface ocean. *Global Biogeochemical Cycles*, *26*, GB1016. https://doi.org/10.1029/2011GB004069

Hansell, D. A., Carlson, C. A., Repeta, D. J., & Schlitzer, R. (2009). Dissolved organic matter in the ocean: New insights stimulated by a controversy. *Oceanography*, *22*, 52–61.

Hawkes, J. A., Rossel, P. E., Stubbins, A., Butterfield, D., Connelly, D. P., Achterberg, E. P., et al. (2015). Efficient removal of recalcitrant deep-ocean dissolved organic matter during hydrothermal circulation. *Nature Geoscience*, *8*(11), 856–860. https://doi.org/10.1038/ngeo2543

Hertkorn, N., Benner, R., Frommberger, M., Schmitt-Kopplin, P., Witt, M., Kaiser, K., et al. (2006). Characterization of a major refractory component of marine dissolved organic matter. *Geochimica et Cosmochimica Acta*, *70*(12), 2990–3010. https://doi.org/10.1016/j.gca.2006.03.021

Hertkorn, N., Harir, M., Koch, B. P., Michalke, B., & Schmitt-Kopplin, P. (2013). High-field NMR spectroscopy and FTICR mass spectrometry: Powerful discovery tools for the molecular level characterization of marine dissolved organic matter. *Biogeosciences*, *10*(3), 1583–1624. https://doi.org/10.5194/bg-10-1583-2013

Jiao, N., Robinson, C., Azam, F., Thomas, H., Baltar, F., Dang, H., et al. (2014). Mechanisms of microbial carbon sequestration in the ocean—Future research directions. *Biogeosciences*, *11*(19), 5285–5306. https://doi.org/10.5194/bg-11-5285-2014

Lam, B., Baer, A., Alaee, M., Lefebre, B., Moser, A., Williams, A., et al. (2007). Major structural components in freshwater dissolved organic matter. *Environmental Science & Technology*, *41*(24), 8240–8247. https://doi.org/10.1021/es0713072

Lang, S. Q., Butterfield, D. A., Lilley, M. D., Paul Johnson, H., & Hedges, J. I. (2006). Dissolved organic carbon in ridge-axis and ridge-flank hydrothermal systems. *Geochimica et Cosmochimica Acta*, *70*(15), 3830–3842. https://doi.org/10.1016/j.gca.2006.04.031

Lechtenfeld, O. J., Kattner, G., Flerus, R., McCallister, S. L., Schmitt-Kopplin, P., & Koch, B. P. (2014). Molecular transformation and degradation of refractory dissolved organic matter in the Atlantic and Southern Ocean. *Geochimica et Cosmochimica Acta*, *126*, 321–337. https://doi.org/10.1016/j.gca.2013.11.009

Lechtenfeld, O. J., Koch, B. P., Gašparović, B., Frka, S., Witt, M., & Kattner, G. (2013). The influence of salinity on the molecular and optical properties of surface microlayers in a karstic estuary. *Marine Chemistry*, *150*, 25–38. https://doi.org/10.1016/j.marchem.2013.01.006

Loh, A. N., Bauer, J. E., & Druffel, E. R. M. (2004). Variable aging and storage of dissolved organic components in the open ocean. *Nature*, *430*(7002), 877–881. https://doi.org/10.1038/nature02780

Matsumoto, K. (2007). Radiocarbon-based circulation age of the world oceans. *Journal of Geophysical Research*, *112*, C09004. https://doi.org/10.1029/2007JC004095

Meng, C. K., & Fenn, J. B. (1991). Formation of charged clusters during electrospray ionization of organic solute species. *Journal of Mass Spectrometry*, *26*(6), 542–549. https://doi.org/10.1002/oms.1210260604

Ogawa, H., Fukuda, R., & Koike, I. (1999). Vertical distributions of dissolved organic carbon and nitrogen in the Southern Ocean. *Deep Sea Research, Part I*, *46*(10), 1809–1826. https://doi.org/10.1016/S0967-0637(99)00027-8

Repeta, D. J., & Aluwihare, L. I. (2006). Radiocarbon analysis of neutral sugars in high- molecular-weight dissolved organic carbon: Implications for organic carbon cycling. *Limnology and Oceanography*, *51*(2), 1045–1053. https://doi.org/10.4319/lo.2006.51.2.1045

Sofer, Z. (1980). Preparation of carbon dioxide for stable carbon isotope analysis of petroleum fractions. *Analytical Chemistry*, *52*(8), 1389–1391. https://doi.org/10.1021/ac50058a063

Stuiver, M., & Polach, H. (1977). Discussion of reporting of ^{14}C data. *Radiocarbon*, *19*(03), 355–363. https://doi.org/10.1017/S0033822200003672

Stuiver, M., Quay, P., & Oslund, H. G. (1983). Abyssal water carbon-14 distribution and the age of the world oceans. *Science*, *219*(4586), 849–851. https://doi.org/10.1126/science.219.4586.849

Walker, B., Beaupré, S., Guilderson, T., Druffel, E., & McCarthy, M. (2011). Large-volume ultrafiltration for the study of radiocarbon signatures and size vs. age relationships in marine dissolved organic matter. *Geochimica et Cosmochimica Acta*, *75*(18), 5187–5202. https://doi.org/10.1016/j.gca.2011.06.015

Walker, B. D., Beaupré, S. R., Guilderson, T. P., McCarthy, M. D., & Druffel, E. R. M. (2016). Pacific carbon cycling constrained by organic matter size, age and composition relationships. *Nature Geoscience*, *9*(12), 888–891. https://doi.org/10.1038/ngeo2830

Walker, B. D., Primeau, F. W., Beaupré, S. R., Guilderson, T. P., Druffel, E. R. M., & McCarthy, M. D. (2016). Linked changes in marine dissolved organic carbon molecular size and radiocarbon age. *Geophysical Research Letters*, *43*, 10,385–10,393. https://doi.org/10.1002/2016GL070359

Williams, P. M., & Druffel, E. R. M. (1987). Radiocarbon in dissolved organic matter in the Central North Pacific Ocean. *Nature*, *330*(6145), 246–248. https://doi.org/10.1038/330246a0

Zigah, P. K., McNichol, A. P., Xu, L., Johnson, C., Santinelli, C., Karl, D. M., et al. (2017). Allochthonous sources and dynamic cycling of ocean dissolved organic carbon revealed by carbon isotopes. *Geophysical Research Letters*, *44*, 2407–2415. https://doi.org/10.1002/2016GL071348