UNIVERSITY OF CALIFORNIA, SAN DIEGO

Natural Abundance Radiocarbon Studies of Dissolved Organic Carbon (DOC) in the Marine Environment

A Dissertation submitted in partial satisfaction of the

Requirements for the degree Doctor of Philosophy

in

Oceanography

by

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2008

The Dissertation of Roman Paul de Jesus is approved, and it is acceptable in quality and form for publication on microfilm.

Chair

University of California, San Diego

2008

EPIGRAPH

It is clear, therefore, that the information content of organic molecules, which also carry imbedded stable isotopic signatures and radiocehmical clocks, the unsurpassed by any other seawater component. The 10^{12} diverse organic molecules dissolved in every milliliter of seater are the only constitutnets whose stared information approaches the richeness needed to understand where that water has been and what has happened within it over time. The future of oceanographic research belongs in large part to those who can learn to read these molecular messages.

-John I. Hedges, 2002

Mark it, Dude.

-The Big Lebowski

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LIST OF ABBREVIATIONS

DOC	dissolved organic carbon
DOM	dissolved organic mattter
HPLC	high performance liquid chromatography
NMR	nuclear magnetic resonance
POC	particulate organic carbon
POM	particulate organic matter
SPE	solid phase extraction (or extract)
TOC	total organic carbon
UDOC	ultrafiltered dissolved organic carbon

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ACKNOWLEDGEMENTS

First and foremost, I thank my parents (Valerie and Rene) and brother (Matthew) and extended family for their unconditional support for this thesis and all of my endeavors. Dr. Victoria Cordy has always been a great friend and very supportive of me throughout my graduate career - I thank you and you will always have a special place in my heart.

I am grateful to my advisor, Lihini, for all her guidance, advice and support. I was so fortunate to have arrived in your lab; it has truly been an honor to have you as a thesis advisor. I will always treasure the friendships that grew from the lab with Travis and Roberta; I can't imagine two better people to share the lab with. I will miss the two of you. I thank the rest of the Aluwihare Lab (past and present) for their help: Dr. Susan Lang, Jordan Watson, and Laurie Chong.

Thanks to Bruce Deck for his technical assistance with isotope measurements, the gas purification line, and his sense of humor. I am gracious to Michaele Kashgarian and the girls in the Graphite Lab, Paula and Dot, at the Center for Accelerator Mass Spectrometry at Lawrence Livermore National Lab. I thank John Sharp for his general guidance with the TOC analyzer. Thanks to the CalCOFI techs, Dave, Jim, Jen, and Fernando, for their savvy on the cruises.

Finally, I thank all my friends with whom I have shared some great times and who have offered great advice over the past few years. Thank you: Natalie Rodriguez, Drs. Evan Solomon, Katherine Maloney, Wendy Strangman, Brian Hopkinson, Julie

Robidart, Jenna Hill, Chris Reddy, soon to be Drs. Ryan Rykaczewski, Heather Graven, Melissa Headly, Mike Vardaro, Andy King. It was a fun ride while it lasted, ;-).

Chapter 3, in part, will be submitted for publication with Dr. Lihini I. Aluwihare and Dr. Michaele Kashgarian. The dissertation author was the primary investigator and author of this paper. We thank the CalCOFI technicians and crews of the R/V Roger Revelle and R/V New Horizon for their technical assistance.

Chapter 4, in part, will be submitted for publication with Dr. Lihini Aluwihare to Nature. The dissertation author was the primary investigator and author of this paper. We thank the crews of the R/V Roger Revelle and R/V Knorr for their technical assistance.

Chapter 5, in part, will be submitted for publication with Dr. Lihini I. Aluwihare. The dissertation author was the primary investigator and author of this paper. We thank the crews of the R/V Atlantis and R/V Hugh R. Sharp for their technical assistance.

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de Jesus, Roman P. and Faulkner, D. John (2003). Chlorinated Acetylenes from the San Diego sponge *Haliclona lunisimilis*. *J. Natural Products*, 66(5), 671-674.

FIELDS OF STUDY

Major fields of study

Studies in Organic Geochemistry
Professor Lihini I. Aluwihare

Studies in Marine Natural Products
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ABSTRACT OF THE DISSERTATION

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Professor Lihini I. Aluwihare, Chair

Marine dissolved organic carbon (DOC), an active reservoir in the global carbon cycle, has an average age of 6000 years and is comprised of biochemicals which may or may not cycle on different time scales. The primary objective of this thesis is to investigate the relationship between radiocarbon (^{14}C) signature and chemical composition of various fractions within DOC. Specifically, this thesis was designed to provide an analytical framework to explicitly explore the relationship between DOC

molecular weight, chemical composition, and reactivity. Chapter 2 describes spatial and temporal total organic carbon (TOC) concentration gradients in 2005 from the California Cooperative Fisheries Oceanic Cooperative (CalCOFI) region, which is the main study area for this thesis. Chapter 3 describes a novel solid phase extraction (SPE) method and reports the chemical and isotopic characteristics of fractionated DOC components based on their solubility. These fractions are compared to DOC isolated by the widely applied UF techniques and the results indicate that chemical composition and ^{14}C content were related. Using compound specific isotope analysis (CSIA) of sugars and lipids, Chapter 4 shows that both SPE DOC and UDOC contain specific compounds with similar chemical and isotopic characteristics and residence times. In Chapter 5, CSIA was applied to SPE samples collected from the Delaware River and Estuary and the terrestrially influenced Eel River Margin to examine whether riverine DOC inputs contribute specific compounds or common components to the coastal ocean. Finally, the final chapter discusses the implications and significance of these results for marine DOC cycling.

1 INTRODUCTION

1.1 SIGNIFICANCE OF MARINE DISSOLVED ORGANIC CARBON

Marine dissolved organic carbon (DOC) is operationally defined as the fraction of marine organic carbon which passes through a 0.2 μm filter. This reservoir plays an active role in the global carbon cycle and at 650 Gt C (1 Gt = 10^{15} g), is approximately equivalent to the amount of CO_2 in the atmosphere (Hedges, 1992; Hansell, 2002). Marine DOC interacts with atmospheric CO_2 through the biological fixation and remineralization of dissolved inorganic carbon (DIC), and its accumulation is controlled by numerous biological, physical, and chemical processes. Radiocarbon ($\Delta^{14}C$) measurements have shown that DOC may persist for thousands of years (Williams and Druffel, 1987; Druffel et al., 1992), but some components part of the larger pool turnover more rapidly (Santschi et al., 1996, Aluwihare et al., 2002). Together these studies show that DOC essentially acts as a 'capacitor' for carbon due to its capability to store and release CO_2 over various timescales. Therefore, understanding the cycling and role of marine DOC is important to any study of the global carbon cycle and by extension, global climate.

1.2 DISTRIBUTIONS, SOURCES, AND SINKS OF MARINE DOC

Surface DOC concentrations are typically 60-70 μM C in the surface open ocean but can exceed 100 μM C in highly productive areas such as upwelling and coastal

regions. In the deep North Pacific ocean, at the end of the ocean's conveyor belt circulation, DOC concentrations decrease to 35-37 μM C in the deep ocean (Williams and Druffel, 1987; Druffel et al., 1992; Hansell and Carlson, 2001). In addition to spatial variation, DOC concentrations can vary temporally. Regular sampling and measurements at the Bermuda Atlantic Time Series station revealed that both physical processes, such as overturning and advection of surface waters, and biological processes affect DOC distributions (Carlson and Ducklow, 1995; Hansell and Carlson, 2001).

New DOC can be introduced into the water column in several ways. The biological pump converts dissolved CO_2 into organic carbon in the surface ocean where it may be stored in living or detrital particulate organic carbon (POC) and DOC. Marine organisms can exude DOC in many forms including polysaccharides, proteins and metal binding ligands (Bruland, 1989; Rue and Bruland, 1997; Carlson, 2002) and sloppy feeding by larger organisms can also introduce DOC into the water column (Strom et al., 1997). In addition, rivers contribute terrestrial DOC to the ocean (Meyers-Schulte et al., 1986; Hedges et al., 1997 and references therein; Opsahl and Benner, 1997; Benner and Opsahl, 2001) The annual flux of terrestrial DOC (0.25 Gt C) is enough to sustain the steady state turnover in the ocean however, studies have shown that <10% of marine DOC is composed of terrestrial material (Meyers-Schulte et al., 1984). Other processes, such as chemosynthesis in the deep ocean (e.g., Ingalls et al., 2006), hydrothermal processes (Lang et al., 2006), and diffusion from sediments (Bauer et al., 1995) also add DOC into the marine reservoir. Over time, DOC can be transformed through a number of processes including abiotic degradation (e.g. photochemical; Mopper et al., 1996), humification (Meyers-Schulte and Hedges, 1986), biotic transformation (Ogawa et al.,

2001), adsorption onto particles (Hwang et al., 2003), and aggregation into particles or gels (Chin et al., 1998; Alldredge et al., 1993). These processes are likely to have an important effect on the composition, bioavailability and residence time of DOC in the water column. In addition, deep water formation can sequester recently produced DOC while upwelling can resurface aged DOC from depth (Hansell, 2002). The sequestration of potentially labile DOC in the deep ocean is an important factor since it isolates this material from the more biologically active surface ocean. While slow removal processes in the deep ocean may be important for deep ocean budgets where most of the DOC pool resides (Hansell and Carlson, 1998).

1.3 RADIOCARBON DISTRIBUTIONS AND USE AS A TRACER OF DOC CYCLING

Radiocarbon (^{14}C) measurements have shown that the average '^{14}C-age' of the DOC reservoir is ~6000 years with respect to atmospheric CO_2 (Williams and Druffel, 1987; Druffel et al., 1992). Together with its total inventory, this average residence time yields a relatively small steady state flux of material through the DOC reservoir each year (0.1 Gt C/yr; Williams and Druffel, 1987; Hedges and Oades, 1997). In contrast, both the higher concentration and greater ^{14}C content of DOC in surface waters (<500 m) indicates the production and removal of as much as 50% of the surface inventory on annual to decadal timescales (Williams and Druffel, 1987). Although there is sufficient evidence to support the production and removal of DOC, despite several decades' worth

of research relatively little is still known about its chemical structure and the distribution of ^{14}C within this reservoir.

This thesis uses ^{14}C measurements to estimate the residence time of various DOC fractions and enables a comparison of sources and ages of carbon through the use of $\Delta^{14}C$ (‰) notation. The definition and conventions for ^{14}C usage can be found in Section 3.2.5.

A radiocarbon review of processes relevant in marine systems and a general description of conventions and definitions are described in detail by McNichol and Aluwihare (2007). The diverse distribution of ^{14}C in various environmental carbon reservoirs makes it a unique tracer for carbon cycle studies (Figure 1.1). During the 1950s and 1960s above ground nuclear weapons testing created a 'bomb' spike in the abundance of ^{14}C in the atmosphere. The bomb spike peaked in the early 1960s to approximately 800‰ in the atmosphere. Since that time, the abundance of atmospheric ^{14}C has been steadily decreasing due to anthropogenic inputs of ^{14}C-dead (-1000‰) CO_2 from fossil fuel burning (also known as the Seuss Effect; Levin and Hassheimer, 2000) and loss to the biosphere and mainly the surface ocean. The bomb spike was also transferred to marine surface DIC approximately 10 years later although it was not as significant relative to the atmosphere (Mahadevan, 2001 and references therein).

Radiocarbon in marine surface DIC, once equilibrated through air-sea gas exchange, is incorporated into organic matter during surface ocean biological processes. Two factors control $\Delta^{14}C$ value: the source of carbon (e.g. atmospheric CO_2, DIC, fossil fuels) and radioactive decay. Once incorporated into organic matter, ^{14}C decays predictably and acts as an atomic clock for DOC in the ocean. Therefore we can

evaluate the residence time of a DOC sample in the ocean to relative to the surrounding water mass (e.g. surface waters) using its $\Delta^{14}C$ value and the $\Delta^{14}C$-DIC value. This is also the case for terrestrial organic matter, except that it retains the atmospheric ^{14}C signature as opposed to marine DIC signature. Until recently the $\Delta^{14}C$ signature of atmospheric C was distinct from that of marine DIC, and so DOC derived from marine production was distinguishable from allochthonous DOC. Other sources of carbon with unique $\Delta^{14}C$ signatures, such as sedimentary carbon, methane seeps, DIC in the deep ocean, and mantle CO_2 can also be traced through their unique isotopic values.

1.4 REFRACTORY MARINE DOC

The ^{14}C-age of DOC in the deep Pacific was found to be over 6000 years old, indicating that a large fraction of the DOC reservoir has cycled through the entire ocean several times without being respired as CO_2 (Figure 1.2; Williams and Druffel, 1987; Druffel et al., 1992). For comparison, whole ocean circulation occurs on timescales of 1500 years. The persistence of organic compounds over long time scales suggests that marine microorganisms do not or cannot readily utilize certain parts of the DOC reservoir. By definition, the refractory fraction of DOC is not expected to be bioavailable. However, radiocarbon based studies examining microbial uptake of natural DOC suggest that bacteria are able to utilize ^{14}C-depleted DOC to support their growth (Cherrier et al., 1999; McCallister et al., 2004). However, it is not known whether organisms directly access refractory DOC, or whether these compounds are first transformed into more labile material by abiotic processes, such as photochemistry in

surface waters (Mopper et al., 1991). The transformation of fresh DOC into refractory material is an important step towards carbon sequestration in the ocean. Therefore, elucidating processes that act on organic carbon during its lifetime in the ocean is essential for understanding the role of DOC in the global carbon cycle.

The apparently old ^{14}C-age of DOC has been attributed to two different and conflicting scenarios. The first scenario is the slow accumulation of fresh DOC which survives remineralization as it circulates through the ocean (Druffel et al., 1992; Hansell and Carlson, 1998). This scenario is difficult to test because the annual accumulation may be too msall to be observed by both available analytical methods and the interval over which current observations exist. On the other hand, mounting evidence has supported a 2-component model of DOC cycling throughout the ocean. This hypothesis assumes that an old, refractory component is persistent at all depths and that fresh DOC is added in the surface. DOC and radiocarbon profiles have supported this model of DOC residence time. In fact, a Keeling plot (Keeling, 1958) analysis using two components, one modern and one refractory be too small to be observed by both available analytical methods and the interval can accurately reproduce the Δ^{14}C distribution in a variety of ocean sites as a function of the DOC concentration profile (Figure 1.3; Mortazavi and Chanton, 2004).

If DOC is indeed composed primarily of refractory and modern components then the source of the refractory component remains elusive. The weathering of sub-surface soils can introduce a pre-aged (i.e., ^{14}C-depleted) terrestrial DOC component into rivers and coastal waters (Raymond and Bauer, 2001). However, the dearth of terrestrial chemical and isotopic data precludes a definitive assessment of the contribution of ^{14}C-

depleted terrestrial organic matter to the DOC pool accumulating in marine environments (Meyers-Schulte and Hedges, 1986). Regardless, terrestrial inputs to coastal regions do impact coastal biological and chemical processes thereby representing an indirect but important source of DOC as well (Hedges et al., 1994; Vlahos et al., 2002; Benner et al., 2004). Alternatively, hydrothermal vents may introduce ^{14}C depleted DOC into the water column (Lang et al., 2006). However, DOC distributions in the deep ocean do not support significant contributions of these point sources in the ocean. In addition, stable isotope data indicate that marine refractory DOC has an autochthonous source (Williams and Gordon, 1970; Williams and Druffel, 1987). Therefore, other factors must control the formation, bioavailability and residence time of refractory DOC.

1.5 ANALYTICAL APPROACHES TO ELUCIDATING THE CHEMICAL STRUCTURE OF MARINE DOC

One of the major obstacles to studying DOC composition and cycling is the extraction of sufficient amounts of organic material from seawater. DOC concentrations are approximately 0.5-1 mg C L^{-1} of seawater, which are insignificant in comparison to the salt content (approximately 35 g L^{-1} seawater, 5,000 times that of organic carbon). Due to the high salt content, traditional organic chemistry analytical techniques are practically useless for whole seawater analysis. Therefore researchers have been compelled to develop methods that isolate and concentrate DOC from seawater while excluding sea salts and minimizing alterations to DOC chemical properties.

Two primary methods have been used to isolate marine DOC, solid phase extraction (SPE) and ultrafiltration (UF). These two methods isolate DOC based on chemical structure (hydrophobic compounds), and molecular size (>1000 Da), for SPE and UF respectively. Differences between these isolation techniques have led to the characterization of different fractions of DOC.

SPE was used in preliminary studies to isolate DOC from acidified seawater and this fraction was operationally defined as marine humic substances (Stuermer and Harvey, 1974; Gagosian and Stuermer, 1977; Druffel et al., 1992). Based on initial studies of elemental and isotopic data, Gagosian and Stuermer (1977) hypothesized that DOC was a condensed structure composed of known biochemicals such as sugars, amino acids, and lipids (Figure 1.4). They noted that marine DOC underwent a humification process similar to terrestrial DOC and was probably resistant to biological degradation. $\Delta^{14}C$ measurements established these fractions as refractory components of DOC because they were depleted in ^{14}C relative to bulk DOC (Druffel et al. 1992).

More recently, researchers have focused on ultrafiltered DOC (UDOC) which is retained by a 1 nm filter (nominally >1000 Da). This technique separates by size and not composition, so isolated compounds are expected to be more representative of total DOC (Benner, 2002). From NMR and compositional data, UDOC is mainly composed of compounds with polar functional groups. The nearly constant ratio of neutral sugars in DOC throughout the ocean has suggested the existence of a common biopolymer which may be resistant to microbial degradation (McCarthy et al., 1996; Aluwihare et al., 1997).

Compositional studies have shown that marine DOC is comprised of several chemical moieties (reviewed in Benner, 2002) and may be derived from a variety of sources (Hedges and Oades, 1997). Therefore, individual components could have very different rates of biological and chemical reactivity. For example, compound-specific measurements have shown that the $\Delta^{14}C$ value of carbohydrates dissolved in the surface ocean is similar to $\Delta^{14}C$-DIC in these same waters (Repeta and Aluwihare, 2006). This finding is consistent with recent production of carbohydrates during photosynthesis and subsequent removal on short timescales. Alternatively, studies have indicated the presence of dissolved components with ^{14}C-ages that exceed 6000 years (Wang et al., 2001, Loh et al., 2004). Together, these available data confirm the presence of both modern (< 50 years) and old (>6000 radiocarbon years) organic compounds in the DOC reservoir. While many studies have focused on the elemental and isotopic characteristics of the bulk DOC pool, it is becoming increasingly clear that both short- and long-term variations in DOC concentration and cycling will be better understood if we examine specific components within these reservoirs.

Hwang and Druffel (2003) showed that the acid-insoluble fraction of suspended POC was remarkably old and had a stable carbon isotopic signature similar to lipids. They further extended this observation to hypothesize that these old lipids may have originally been part of the refractory DOC pool and were absorbed onto suspended particles in the water column (Hwang et al., 2006). Chemical characterization (Benner et al., 1992; McCarthy et al., 1996; Aluwihare et al., 1997), bacterial uptake experiments (Amon and Benner, 1994), and $\Delta^{14}C$ measurements (Aluwihare et al., 2002; Loh et al., 2004; Repeta and Aluwihare, 2006) support the hypothesis that UDOC is rapidly

recycled in the surface ocean. However, ^{14}C-depleted compounds have also been isolated from UDOC (Loh et al., 2004; Repeta and Aluwihare, 2006) and a recent study comparing surface and deep UDOC deduced a hypothetical structure for refractory DOC that was dominated by carboxy-rich aliphatic macromolecules (CRAM; Hertkorn et al., 2006). This latter result is consistent with the observed enrichment of carbon in DOC with depth in the ocean (Hopkinson and Vallino, 2005). As previous studies provided some consistent and some conflicting information regarding the relationship between DOC molecular weight, chemical composition, and reactivity, this thesis was designed to provide an analytical framework to explicitly explore this relationship.

1.6 GOALS OF THIS THESIS

The primary goal of this thesis is to chemically and isotopically characterize marine DOC and identify modern and refractory components. The data presented here will provide evidence for the 2-component age model for marine DOC through coupled radiocarbon and chemical characterization. In order to understand processes that control DOC cycling within the Southern California Bight, seasonal TOC measurements were performed throughout the California Cooperative Ocean and Fisheries Investigation (CalCOFI) study area. This study site was easily accessible through quarterly CalCOFI cruises and was a suitable site to carry out marine studies because terrestrial influences were minimal in this region.

The development of a solid phase extraction (SPE) technique provided a novel scheme for DOC fractionation. Fractions obtained were representative of marine DOC

and were suitable for further chemical and isotopic analyses. The residence time of chemically diverse fractions were examined using stable and radioisotopes combined with chemical characterization (^{1}H NMR). Finally, compound specific isotope analysis (CSIA) was employed to evaluate whether SPE and UF isolate compounds with similar residence times in the ocean and rivers and to investigate mechanisms that contribute to the formation of refractory DOC.

1.7 ORGANIZATION OF THESIS

Chapter II of this thesis describes the seasonal distribution of TOC in the Southern California Bight using samples collected during routine, quarterly CalCOFI cruises. The observed TOC distribution was interpreted within the context of the complex physical and biological regimes in the region and was aided by the ancillary data available as part of this extensively studied 50+ year time-series.

In Chapter III, the development of a novel SPE method is described and initial findings from the application of this method to isolate and characterize DOC in the CalCOFI region are discussed. Briefly, the chemical and isotopic characteristics of DOC components fractionated based on their solubility are reported and compared to DOC isolated by the widely applied UF techniques.

Chapter IV presents data from CSIA of sugars and lipids isolated from SPE and UDOC samples collected in the CalCOFI region. Goals of this chapter were to determine whether these two methods access similar pools of specific compounds and examine factors controlling DOC residence time in the surface ocean.

In chapter V, CSIA was applied to SPE samples collected from the Delaware River and Estuary and the terrestrially influenced Eel River Margin to examine whether riverine DOC inputs contribute specific compounds or common components to the coastal ocean.

Chapter VI presents general conclusions from this thesis.

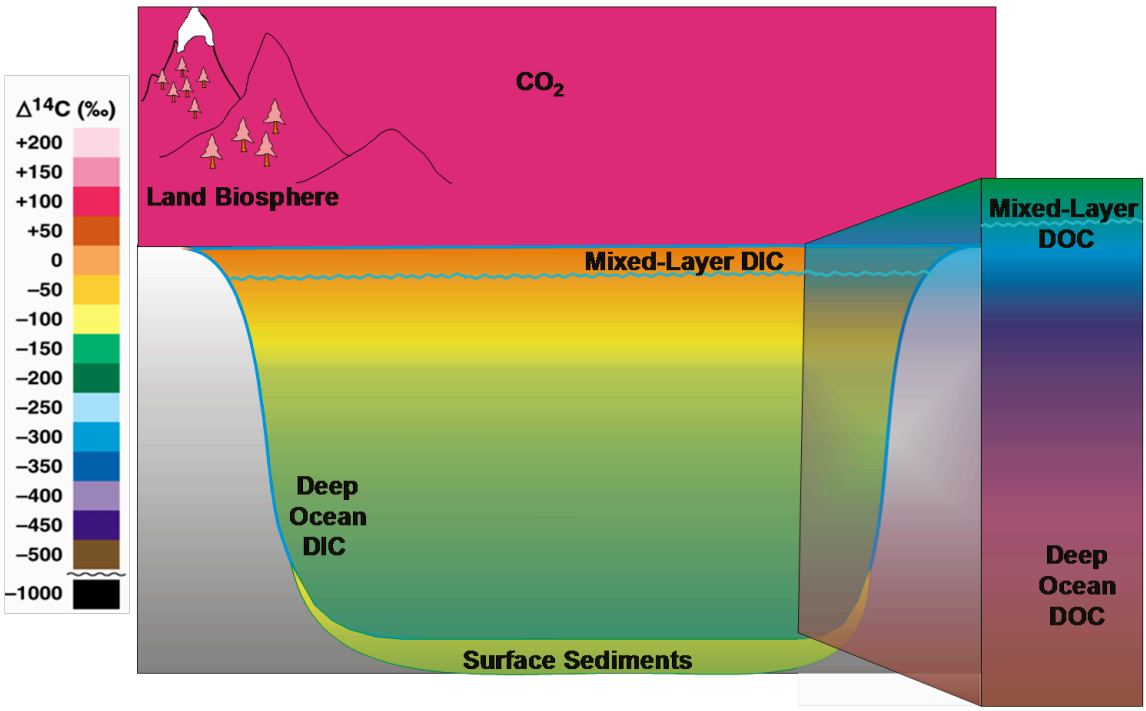

Figure 1.1 Distribution of radiocarbon (^{14}C) in the environment (from McNichol and Aluwihare, 2007).

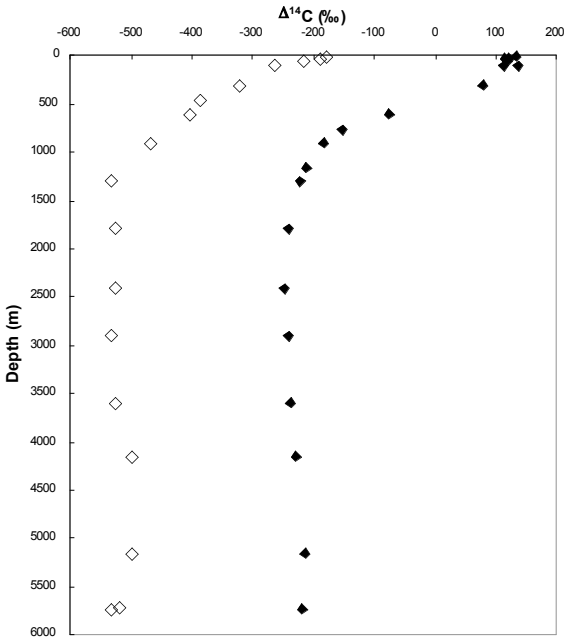

Figure 1.2 Radiocarbon distribution of DIC (closed symbols) and DOC (open symbols) with depth in the North Pacific Ocean (data from Druffel et al., 1992 in 1987).

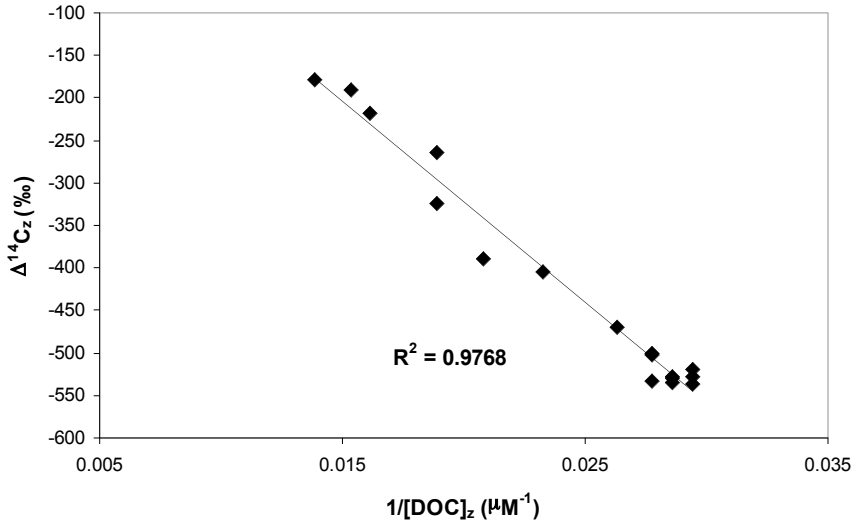

Figure 1.3 Keeling plot of $\Delta^{14}C$ values vs. 1/[DOC] from various sites in the North Pacific Ocean. The relationship supports a 2-component age model of DOC (from Mortazavi and Chanton, 2004).

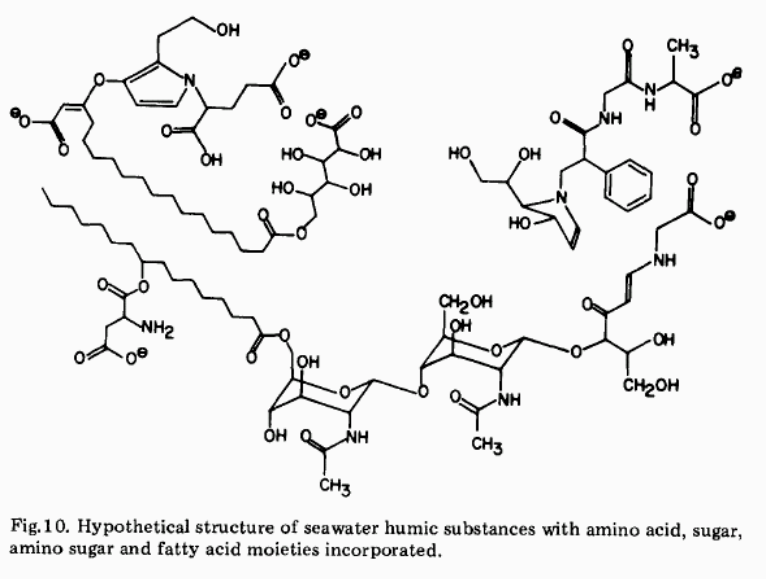

Fig. 10. Hypothetical structure of seawater humic substances with amino acid, sugar, amino sugar and fatty acid moieties incorporated.

Figure 1.4 Hypothetical structure of marine humics (from Gagosian and Stuermer, 1977).

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2 SPATIAL AND TEMPORAL OBSERVATIONS OF ORGANIC MATTER IN THE SOUTHERN CALIFORNIA BIGHT

2.1 INTRODUCTION

The marine dissolved organic matter (DOM) reservoir represents one of the largest active reservoirs of carbon on earth, comprising some 650 Gt C; despite its size and potential importance, the dynamics of DOM have not been resolved fully, and, therefore the ultimate role of this reservoir for the global and marine carbon cycle remains to be established. The challenges we face involve both (1) identifying the chemical, biological and physical factors that control the size of this reservoir; and (2) identifying the range of timescales on which DOM cycles. Historically, studies on DOM cycling focused on variations in bulk properties, whether through monitoring spatial arrays or at fixed points such as Station ALOHA and BATS (Hansell and Carlson, 2001; Church et al., 2002; Abell et al., 2000 and 2005; Knapp et al., 2005). Observations at BATS have highlighted the importance of both biological and physical processes in controlling the upper ocean inventory of dissolved organic carbon (DOC). Seasonal variations have been observed at this site with accumulations of DOC observed immediately following the spring bloom. The range of surface values observed at BATS over a 5-year period was 63 to ~68 μM C (Hansell and Carlson, 2001), and the greatest seasonal variability occurred between the depths of 100 m and 300 m, when convective mixing of surface accumulated stocks introduced DOC into subsurface waters. In contrast, both Hansell and Carlson (2001) and Knapp et al. (2005) observed little

temporal variability in dissolved organic nitrogen (DON) concentration at BATS. This observation argues for a decoupling of DON and DOC production and degradation in the upper ocean at this site. At Station ALOHA, Church et al. (2002) also observed a decoupling of DOC and DON dynamics in the upper ocean; and Abell et al. (2000, 2005) reached a similar conclusion based on studies of DOM remineralization within thermocline waters of the North Pacific Subtropical Gyre. In addition, Church et al. (2002) reported a secular increase in DOC concentration at Station ALOHA over the time period of observation between 1989 and 1999, which they attributed to ecosystem transitions in response to variations in physical forcing on decadal timescales.

An extensive study examining the temporal and spatial variability in DOC was conducted by Vlahos et al. (2002) in the Mid-Atlantic Bight during the Ocean Margins Program. Samples were collected in spring 1994 and 1996, and summer 1996. High spring accumulations of DOC were reported for coastal waters in this region and were attributed to recent primary production. The region sampled in this study was significantly impacted by terrestrial inputs but the study concluded that terrestrial DOC did not contribute significantly to general enrichments observed along the coast.

These previous studies have documented the influence of physical and biological processes on DOC dynamics in the upper ocean. In particular, DOC accumulations were associated with biological production and upper ocean stratification; and convective overturning of the water column led to significant decreases in the concentration of DOC in surface waters. In addition, these previous studies indicated that seasonal variations in DON were relatively subdued. Finally, one study at Station ALOHA implied that long-term variations in DOC dynamics may be linked to climate change. To expand these

observations and monitor DOM dynamics in an upwelling driven system, that is also strongly influenced by climate variability on a variety of timescales, a time series of total organic carbon (TOC), total nitrogen (TN = nitrate + nitrite + ammonium + DON) and particulate organic matter (POM) measurements was begun offshore of central and southern California. This region is particularly attractive for such an effort because coastal ocean processes can be studied exclusive of major terrestrial inputs. The region also served as the main study site for much of the DOC fraction-specific research that was conducted as part of this thesis, and so, it was important to constrain the spatial and temporal variability in TOC and TN concentration.

Sampling for this study began in 2004 through participation in California Cooperative Oceanic Fisheries Investigation (CalCOFI) survey cruises. The CalCOFI program has operated quarterly (winter, spring, summer, fall) survey cruises since 1949 collecting hydrographic and biological data at several stations (66 at present) off the coast of central and southern California (Figure 2.1; Hayward and Venrick, 1998), and it represents the longest, ongoing, ocean time-series. The major hydrographic features of the region are the near-surface California Current (CC) that transports relatively cold, nutrient rich subarctic waters equatorward at a distance of 200-300 km offshore, and two poleward flowing, relatively warmer and nutrient poor currents in the coastal zone. The poleward flowing currents are the California Undercurrent (centered on the slope at a depth of 100-400 m; Bodgrad *et al.,* 2001 and reference therein) which carries waters from the tropics, and a variable surface flow (the Inshore Countercurrent) which is driven equatorward by wind forcing and coastal upwelling in the spring and summer but poleward in the winter and fall. In addition, seasonal variations in both the California

Countercurrent and Undercurrent are found to occur in relation to the Southern California Eddy (Lynn and Simpson, 1990).

In addition to representing one of the longest time series on "the physics, chemistry and biology of a dynamic oceanic regime" (Bograd and Lynn 2001), the spatially resolved CalCOFI time series has demonstrated a strong coupling between physical, chemical and biological processes in this region. Here, the biological production is driven primarily by nutrients delivered to the surface ocean as a result of wind driven coastal upwelling (the primary site of upwelling for the CalCOFI region occurs off Point Conception) as is typical of eastern boundary currents and offshore upwelling associated with wind stress curl. Because of the unique properties of each surface and sub-surface water mass and the location of nearshore and offshore upwelling, the nutrient regime of the system is structured according to the hydrography and divides the region into unique biogeographic provinces (Hayward and Venrick, 1998). In addition, the system also responds strongly to local and remote physical forcing on seasonal, interannual and lower frequency timescales (Di Lorenzo et al., 2005).

This chapter discusses data from the first full-year (winter, spring, summer and fall 2005) of organic matter (OM) observations and presents a detailed description of OM distributions within surface waters of the CalCOFI grid (Figure 2.1). A few previous studies have characterized TOC cycling in the California Current region, and they have documented concentration variations associated with physical and biological processes (Bauer et al., 1998; Loh and Bauer, 2000; Hill and Wheeler, 2002). The current data set represents the most spatially and temporally expansive study and refines conclusions

drawn from earlier observations.

2.2 METHODS

2.2.1 Sample collection

All OM samples described here were collected by CalCOFI personnel in 2005 based on sampling protocols developed for this thesis from samples collected during spring and fall 2004. Seawater samples were collected from a rosette of 10L Niskin bottles. TOC/TN samples were collected in Jan (0501), April (0504), July (0507) and November (0511) 2005 aboard each CalCOFI cruise. 40 mL of seawater was collected at 4-6 depths in pre-combusted glass vials fitted with acid rinsed PTFE caps, after rinsing with 3 volumes of sample water. Depths included surface (1-3 m), chlorophyll max (based on CTD data), 115 m, 320 m and 515 m. Note that water samples were not filtered and measurements would yield *total* organic carbon and *total* nitrogen concentrations. Based on previous studies we opted to omit the filtration step to reduce potential sources of contamination. To examine the vertical distribution of TOC within the CalCOFI grid at higher depth resolution, four 'cardinal' stations (90.53, 90.100, 80.55, 80.100) were chosen where a complete vertical profile was sampled down to 500 m. Immediately following sample collection 1 μL HCl (trace metal grade; ACS) per 1 mL of seawater was added to preserve TOC samples and adjust the sample to pH 2 in order to convert all dissolved carbonate species to CO_2.

POM samples were filtered from 1-2 L of seawater through combusted 25mm glass fiber filters (GF/F) using a vacuum filtration rig. Seawater was collected into

polycarbonate bottles directly from the CTD, filtered immediately, and filters were wrapped in combusted foil and stored at -20^{o}C until analysis.

2.2.2 TOC and TN measurements

TOC and TN measurements were performed on a Shimadzu TOC-V analyzer fitted with a TN detector and autosampler. Samples were purged with CO_2 free air for 1 minute to remove CO_2 (derived from inorganic carbon species in the sample) from the sample and then injected onto a quartz column filled with platinum covered aluminum catalyst heated at 720^{o}C.

At the beginning of each run Milli-Q was repeatedly injected into the system until the baseline became stable. Typically 15-30 Milli-Q injections were necessary to stabilize the system's baseline. Platinum catalyst was regenerated every 100-150 samples (not including blanks) by soaking in dilute HCl, and re-generated catalyst was dried in an oven overnight before re-packing the column. Before re-use, the quartz combustion tube was also soaked in dilute HCl to remove salt deposits. The combustion tube was packed with combusted quartz wool retained by a platinum mesh both above and below the catalyst.

Measurements were conducted according to Sharp et al. (2002). Five point calibrations curves were constructed with a range from 0-200 μmol C and 0-50 μmol N. Potassium hydrogen phthalate was used for carbon calibration curves and potassium nitrate was used for nitrogen calibration curves. Organic nitrogen standards and mixed inorganic and organic nitrogen standards were also examined, and inorganic nitrogen calibration curves were found to yield the most reproducible results. Milli-Q water

(used to make the standards) was injected after every 4-5 samples to insure that the baseline remained at its original response. In addition, certified reference materials (CRMs; courtesy of Dennis Hansell, RSMAS) in the form of low carbon (LCW; <2 μM C) and deep Sargasso Sea water (>2000 m, 44-47 μM C and 21 μM N) were included prior to and during each sample run to monitor drift in machine response. Concentrations were calculated from an average of 4 or 5 injections and integrating the area under the curve. Acceptable measurements had coefficients of variation (CV) less than 5%.

2.2.3 Particulate Organic Carbon and Nitrogen (POC and PON)

Samples were frozen and stored for analysis on shore. POM samples were dried in an oven at 50^{o}C then acidified in a desiccator under an atmosphere of HCl. The acidified filters were again dried and weighed. A portion of the dried filter was cut out then folded in a tin cup for analysis. Elemental (C and N) measurements were performed using an elemental analyzer at the SIO Unified Laboratory Facility.

2.3 RESULTS

2.3.1 General California Current System (CCS) conditions in 2005

During 2005, no major climatic events affected the CCS region thus an 'average' year could be sampled. However, sea surface temperature (SST) anomalies were up to 4^{o}C above normal throughout the CCS during spring and early summer. Anomalies were attributed to local effects of relatively weak winds and diminished upwelling throughout

the year and not an apparent larger, gyre-wide shift. Strong upwelling returned to the CCS later in the summer and fall, especially within the southern California region (Goericke et al., 2005; Peterson et al., 2006).

At the beginning of 2005, the CC flowed strongly at the western edge of the station grid. During the spring the CC shifted close to the Channel Islands while moderate poleward currents were observed close to the coast. The CC moved further offshore in the summer and branched into two arms near station 87.70. The Southern California Eddy (SCE) formed strongly around station 87.45. Upwelling was present near Point Conception and extended into the SCE. In the fall, the CC moved even further offshore and remained split into two branches, and the SCE remained strong through the fall. These observations and the following discussion of hydrographic properties, and nutrient and chlorophyll distributions are taken from the State of the California Current report (Goericke et al., 2005; Peterson et al., 2006).

Although conditions in 2005 were typical for the CalCOFI grid, it is unknown what effect, if any, these hydrographic, chemical and biological conditions have on TOC and total organic nitrogen (TON) distributions in the region. TON is defined here as TN minus DIN, where DIN is dissolved inorganic nitrogen as measured by the CalCOFI program. Based on previous studies we anticipated significant variation in TOC near upwelling regions and coastal environments; and did not anticipate any major seasonal variation in TON.

2.3.2 0501 (Winter)

2.3.2.1 Hydrographic and nutrient data

During 0501 (Figure 2.2), SST ranged from 12-16^{o}C throughout the grid and was near overall long-term average conditions (Table 2.1). Warmest SSTs were found at the southwestern edge of the grid, which overlaps the edge of the North Pacific Subtropical Gyre (NPSG); coolest SSTs were found around the central stations on Lines 80 and 77. Surface nitrate was also low during the winter and potential density at the surface indicated low vertical mixing from subsurface waters. Coastal stations were slightly less saline than normal, which was attributed to increased precipitation and runoff during the days leading up to the cruise (Goericke et al., 2005). Chlorophyll *a* concentrations ([chl a]) ranged from 0-2 μg/L with slightly elevated concentrations near the coast and to the north, near Point Conception.

2.3.2.2 Total and particulate organic matter

Lowest TOC concentrations of the year were present in 0501 with values ranging from 55-75 μM C (Table 2.1). TOC was unusually high (151 μM C) at station 90.28 and may reflect addition of OC from urban, freshwater runoff since the salinity was also very low (28.4) and nitrate was relatively high at this location at the time of sampling (Goericke et al., 2005). Stations on the edge of the California Current, 87.70 and 83.80, had the highest TOC values which were ~80 μM C. Lower values were generally found further offshore of those stations and inshore values were nearly constant throughout the coastal region. Lowest TOC concentrations were present around Pt. Conception. This area is known to have frequent upwelling events which can bring TOC depleted waters to the surface and decrease surface TOC concentrations.

TON values were low throughout the winter, ranging from 2-10 μM N. Values

were highest along lines 87 and 83 and along the entire coast. The elevated values coincide with the southern part of an eddy centered on station 80.70 (Goericke et al., 2005).

Lowest POC concentrations were also measured during 0501 and ranged from 5-10 μM C with highest values in the northeastern edge of the grid. Two scenarios could explain the higher POC concentration at this location: 1) Recent upwelling has brought nutrients to surface waters thus increasing primary production; 2) upwelled waters near the shelf can physically bring sedimentary POC to the surface (Bauer et al., 1998). The observation that [chl a] was elevated supports the former scenario. PON was very low in the winter and no value above 2 μM N was observed throughout the region.

TOC and density sections along line 90 showed that TOC concentrations were >60 μM C throughout the mixed layer (~60-70m; Figure 2.3). Surface TOC concentrations appeared to be higher in the filament of water immediately to the east of the shoaling isopycnals near stations 80 and 90. Gyre waters also had elevated surface TOC concentrations. Below the mixed layer, TOC values decreased with depth predictably. Surface TOC concentrations were lower along Line 80 (Fig. 2.4) than along Line 90, and showed a more uniform distribution. Depleted TOC values were also present at shallower depths in the water column as the mixed layer was shallower along Line 80 during 0501.

2.3.3 0504 (Spring)

2.3.3.1 Hydrographic data and nutrients

Spring upwelling occurred near Point Conception extending southward through

the northern Channel Islands into line 87 (Fig. 2.5). The resulting plume was observed with moderate [chl a] and elevated nitrate values. The outer part of the grid remained similar to 0501 with relatively cool SSTs (14-16^{o}C) and low [chl a]. The southern coastal region, inshore of the islands, was not influenced by upwelling processes and had slightly warmer temperatures than those found immediately offshore. The CC effectively isolated coastal and offshore waters from each other and brought relatively cooler water through the middle of the grid.

2.3.3.2 Total and particulate organic matter

During 0504, surface TOC values began to increase with a range between 55 and 99 μM C (Table 2.1). Elevated TOC values are clearly associated with upwelled waters near Pt. Conception and follow the southwest flowing, cooler, denser surface water mass Fig. 2.5). In addition, higher TOC values are still detectable further offshore along Line 83 even though physical, nutrient and biological signatures associated with the upwelling plume are markedly diluted. This observation suggests that TOC accumulations associated with recent biological production may persist to be exported offshore. Further offshore, TOC values remained lower but were generally higher than 0501 values (Table 2.1).

TON concentrations ranged from 1-10 μM N across the grid and were slightly higher along northern lines than in the southern and offshore stations. These concentrations were also generally higher in regions affected by recent upwelling however, the spatial correlation with temperature and density was not as clear for TON as it was for TOC. The persistence of TOC accumulations identified above did not

affect TON to the same degree; and may indicate either the enhanced production of TOC relative to TON or the relatively faster removal of TON by bacterial uptake. However, spatial correlations between TON and TOC were broadly consistent.

POC and PON concentrations also showed some jets/plumes and more closely followed the nitrate distribution and [chl a] in surface waters (Fig. 2.5). Values increased markedly from 0501 values; POC ranged from 5-30 μM C and PON ranged 0-4 μM N (Table 2.1). In nearshore stations POC and TOC distributions are fairly well correlated confirming that recent primary production is likely to be the source of both organic carbon pools. However, POC deviates from TOC particularly in central and offshore waters lending further support to the relative persistence of TOC accumulations. One coastal station 90.28, had 60+ μM C, however it is unclear whether or not this sample was contaminated.

Across line 90, higher TOC values are found where isopycnals shoal toward the coast. In addition, at a few locations subsurface accumulations of TOC were visible at depths as great as 150 m. Along Line 80, surface TOC values were lower at offshore stations and gradually increased in the CC and coastal stations. Again, surface TOC concentrations along this line were more uniform and offshore values were elevated compared to Line 90. High TOC concentrations penetrated to depths of 75 m over most of the line; between stations 50 and 70, high TOC concentrations were observed down to a depth of 150 m. The relatively high TOC concentration in the upper 75 m along this line (inshore of station 80) is likely associated with the southward flowing water mass that was upwelled near Pt. Conception.

2.3.4 0507 (Summer)

2.3.4.1 Hydrographic data and nutrients

During 0507 the upwelling signal was persistent around Point Conception down through the Channel Islands and into Line 87 (Figure 2.6). The Southern California Eddy (SCE) had formed around 87.45 (based on dynamic height anomalies; Peterson et al., 2006) and as expected, the area surrounding this station had colder, denser waters. Increased [chl a] was associated with upwelling off Pt. Conception, and nitrate concentrations near Pt. Conception and within the eddy clearly indicate upwelling at these locations. Enriched nitrate at the surface, up to 8 μM, was recorded near upwelling areas and extended further off Pt. Conception to stations 80.70 and 77.60. During the summer SSTs ranged from 13-19^{o}C and coldest SSTs corresponded to upwelled waters. Warmest SSTs were found in the southern corners of the grid, near the coast and offshore as part of the NPSG. The CC again flowed between nearshore and offshore regions and formed two branches around station 90.70 (Peterson et al., 2006).

2.3.4.2 Total and particulate organic matter

Highest TOC concentrations during the year were observed during the summer (Table 2.1) and ranged from 60-155 μM C. High surface TOC concentrations were generally found outside the CC (in nearshore waters and offshore waters), and highest TOC values were found around San Clemente and Catalina Islands. In the northeastern part of the grid TOC was well correlated with [chl a] distributions, but the extremely high values at inshore stations along line 87 and 90 were not well correlated with [chl a]. It is suggested here that elevated TOC at the southern sites are related to the presence of

the SCE. For example, a locus of high nitrate is also observed at inshore stations along Line 87. In general, around the islands, TOC was elevated relative to other seasons.

TON values were not significantly different from concentrations measured during 0504, but coastal waters appeared to have slightly higher concentrations. As with 0504 the general spatial pattern of higher TON values was broadly consistent with the pattern of TOC accumulation. TON concentrations were best correlated with the inshore spatial distribution of TOC but were more restricted.

POC and PON concentrations were highest in the summer (PON concentrations reached values as high as 8 μM N) and as with TOC, highest concentrations were also found around the Channel Islands. Spatial distributions of POC and PON were well correlated but were not identical to the spatial distribution of [chl a] or TOC. This finding again confirms that different processes act to determine both the accumulation and the timescale of the accumulation of these organic matter reservoirs in the surface ocean. In contrast to TOC, offshore levels of POC and PON remained low.

Along Line 90 TOC was very variable, with highest concentrations observed at inshore stations (between 78 and 122 μM C; Figure 2.3; Table 2.1). Elevated TOC concentrations extended to deeper depths during this month with notably high concentrations near 80 m depth and 320 m depth at station 53. The low TOC concentration at 250 m is not an artifact associated with our vertical sampling resolution because 90.53 is one of the cardinal stations and therefore, all CalCOFI depths were sampled (see later discussion). The maximum at 320 m may be associated with the northward flowing Undercurrent or a downwelling feature, perhaps coupled to the SCE. Along Line 80 TOC was less variable and showed none of the subsurface features

associated with Line 90. Coastal stations did have elevated TOC concentrations, and relatively high subsurface concentrations were only observed for stations offshore of station 80.

During 0507, POC was as high as 60 μM and TOC concentrations were only 90 μM C; it is extremely unlikely that DOC concentrations were as low as 30 μM C in surface waters at this time. Therefore, based on the general results presented here, a simple subtraction of POC from TOC does not necessarily yield DOC concentrations. Several processes may contribute to this observed discrepancy including the different volumes filtered for each analysis (1-2 L for POC versus 40 mL for TOC), the different analytical methods employed (e.g., sample processing, combustion temperature, detection etc.) and the patchy distribution of suspended POM. In addition, studies have suggested that at high POC concentrations, DOC may be absorbed to the GF/F filter and is therefore, included in POC concentration measurements (Hwang et al., 2006).

2.3.5 0511 (Fall)

2.3.5.1 Hydrographic data and nutrients

During 0511, the two branches of the CC moved further offshore, nearer to the edge of the grid with an eddy located at station 90.90. The SCE was still present with the same intensity at the 0507 location. Based on SSTs and density (Fig. 2.7) upwelling appeared to be occurring near Pt. Conceptions but nitrate levels had dropped from 0507 to near 0501 levels. Elevated [chl a] was located just offshore of the northern Channel Islands and Pt. Conception consistent with the spatial distribution of SST. Everywhere else in the region [chl a] was very low. Enhanced [chl a] and diminished nitrate near Pt.

Conceptions suggests that the phytoplankton bloom was well-developed and had significantly depleted the nitrate concentration.

2.3.5.2 Total and particulate organic matter

In fall 2005, TOC decreased from high summer concentrations to values reminiscent of spring (Table 2.1); values ranged from 58 μM C at station 83.110 to 110 μM C at station 77.49. Highest TON concentrations were measured during this season, with relatively high accumulations detected near the coast and Pt. Conception. Highest TOC values were also detected in the northern region around Pt. Conception consistent with [chl a]. High POC (10 to 30 μM C) and PON concentrations were observed along the coast and in regions that exhibited high [chl a]. Interestingly, peak TOC and TON concentrations appeared to spatially "lag" peak [chl a] and POC. As observed throughout the rest of the year, offshore POC and PON levels were quite low.

TOC values along Line 90 decreased from summer values indicating biological processes had decreased during the fall. TOC concentrations in the mixed layer were 60-80 μM C, with higher concentrations near the coast. There was a tongue of more depleted values at 70 m centered between stations 70 and 60, which were near the CC location during this time (Peterson et al., 2006). Outer stations along Line 80 had similar mixed layer TOC concentrations to Line 90, but TOC values were much higher (98 μM C) at station 80.60 and coincided with an area of increased density and upwelling. It appears that values were beginning to decrease before winter.

2.4 DISCUSSION

2.4.1 TOC seasonal and spatial distributions in surface waters

In the preceding section surface distributions of organic matter were shown to exhibit significant spatial and temporal variations. For example, during 2005 surface TOC concentrations generally ranged from 60 μM to 90 μM C, but at coastal stations concentrations were as high as 110 μM C. The average TOC concentration in surface waters during the summer was 81±15 μM C, and measured 0507 concentrations were more variable and elevated relative to winter concentrations (65±3 μM C). In general, an inshore-offshore gradient was observed and appeared to be related to increased primary productivity near the coast; offshore waters also exhibited a smaller range of surface TOC values (60-75 μM) than inshore stations (65-100 μM C; Table 2.1). TOC variability on the edge of the CC just outside the islands was, at times, comparable to coastal stations.

Spatial correlations between hydrographic properties, nitrate distribution, [chl a] and organic matter indicated the importance of upwelling driven primary production in driving both seasonal and spatial variations in OM within the CalCOFI grid. Elevated [chl a] was usually associated with nitrate rich, cold waters, and data presented in this chapter showed for the first time that TOC and POC within the CCS were also elevated in regions where upwelling had recently elevated primary production. However, the relationship between upwelling and TOC concentration is not simple. For example, results showed that newly upwelled water is low in TOC (e.g., near Pt. Conception in 0501), but if the upwelled water mass resided at the surface long enough to accumulate [chl a] it accumulated POC and some TOC as well (e.g., TOC distributions during

0504).

Surface POC and TOC concentrations were plotted versus density to identify production of organic matter within recently upwelled waters (Fig. 2.8). In all cases, correlations with density were low. However, the positive correlation between density and POC increased over the course of the year and during the summer, highest POC concentrations were found in the densest surface waters. TOC concentrations were not well correlated with density however, during 0507, two different surface TOC-density relationships were apparent. In particular, the relatively fresh, less dense CC waters had lower TOC concentrations than the denser surrounding surface waters (Fig. 2.6). However, the TOC rich, lower density water mass near the coast at Line 93 and Line 90 affected the TOC-density relationship in 0507 (Fig. 2.8) as well. Recent upwelling can also dilute the surface TOC inventory (Bauer et al., 1998; Chapter 3) and so a negative relationship between surface TOC and density may be anticipated. However, even in the spring, low TOC values were confined to low density waters. This observation suggests that at the time sampling in 0504, recently upwelled waters had been at the surface long enough to accumulate fresh organic matter.

Figure 2.9 explores the relationship between organic matter concentration and [chl a]. As the year progressed the TOC and POC concentration in surface waters became better correlated with [chl a]. In fact, during fall [chl a] is a relatively good predictor of POC concentrations. However, [chl a] is not necessarily expected to be an accurate predictor of TOC accumulations. For example, Norrman et al. (1995) showed in a mesocosm experiment using a natural diatom assemblage, that although DOC production was occurring throughout the bloom, accumulations of DOC were not

apparent until POC and [chl a] concentrations had begun to decrease. This decoupling of POC and DOC accumulation was attributed to variations in the balance between the rate of DOC production by senescing diatoms and DOC consumption by bacteria. In the current field data set, strongest correlations between TOC and POC were observed for the spring ($R^2 = 0.5$) and the fall ($R^2 = 0.4$) (Fig. 2.10).

2.4.2 Vertical Distributions

Shown in Fig. 2.11 are the depth profiles for TOC collected at the cardinal stations. The surface water build-up of TOC was apparent for all stations and in most cases, highest surface ocean concentrations were observed during the summer (0507; triangles) and lowest concentrations were observed during the winter (0501; diamonds). These observations are consistent with surface distributions discussed above. In general, TOC began to decrease below about 100 m (Fig. 2.11). Lowest TOC values previously observed in the North Pacific Ocean were ~34 μM C (Bauer et al., 1998); lowest TOC concentrations measured at cardinal stations approached this value of 34 μM C given that all values shown in Fig. 2.11 included a measurement error of ~ ±3 μM C (based on consecutive measurements of CRM). At stations 80.100 and 90.100 subsurface TOC concentrations were highest in July, and appeared to be consistent with the entrainment of DOC into deep waters. Subsurface concentrations of TOC also increased at stations 80.55 and 90.53 during July but the vertical distribution of TOC at these stations hinted at the presence of a TOC-rich subsurface water mass. Data from following years should be used to examine the seasonal persistence of such a subsurface feature.

In contrast to the surface TOC-density relationship discussed above and shown in

Fig. 2.8, TOC at depth showed a relatively strong correlation with density. Along Line 90 TOC was well correlated with density at all depths during winter and fall (Fig. 2.12), but showed significant variability in July. This observation is consistent with the subsurface variability in TOC observed at cardinal stations during this cruise. If simple mixing processes led to the subsurface increase in TOC observed during 0507, a better relationship with density might have been predicted. At all depths and during all seasons TOC along Line 80 showed a relatively strong correlation with density (Figure 2.13). These data support the conclusion that mixing processes were important for re-distributing TOC along Line 80 during this time period.

The relationship between TN and density along Lines 80 and 90 are also shown on these figures. TN is expected to show a strong correlation with density except at surface sites where DON may be added. An interesting feature was the lack of a good linear correlation between TN and density along Line 90 during 0507 (and to some extent, 0511). These data suggest that a two endmember mixing model does not accurately predict the density-TN distribution observed at this site during 0507 and at least one other endmember must be considered. This observation is consistent with the previous suggestion that high, summer TOC concentrations observed at a depth of 350 m near station 90.53 were derived from a source external to the CCS. One possible candidate is a northward flowing TOC-rich Undercurrent that had not significantly influenced Line 80 at the time of sampling.

In summary, vertical distributions of TOC in the CalCOFI grid are consistent with observations from other regions (e.g., Hansell and Carlson, 2001). High subsurface values were noted at some stations and coincided with the subsurface chlorophyll

maximum which was found between 20-90 m during 2005. However, maxima at the surface did occur and were frequently found at coastal and inshore stations, especially near regions characterized by upwelling.

In general, data from cardinal stations (Fig. 2.11) and surface TOC distribution shown in Figs. 2.2, 2.5, 2.6 and 2.7 supported previous observations of TOC accumulation during primary production in the upper ocean. In addition, data from cardinal stations and TOC-density sections shown in Figures 2.3 and 2.5 indicated seasonal entrainment of TOC into subsurface waters, and identified a potential allochthonous DOC source to the CCS. On average, TOC values typically decreased below the chlorophyll max as expected and appeared to be largely controlled by physical processes below 100 m.

2.4.3 Santa Barbara Basin

In addition to general observations discussed above, special attention was also paid to OM dynamics in the Santa Barbara Basin (SBB). This basin is known for its high productivity and hypoxic waters near the bottom. During 2004 and 2005, bottom water nitrate concentrations were the lowest (<15 μM) recorded since observations began in 1984 (CalCOFI data reports; Goericke et al., 2005; Peterson et al., 2006; Figure 2.14 0501 and 0511). These low concentrations were indicative of strong denitrification within the basin's sediments. Denitrification occurs when nitrate is used as the terminal electron acceptor for OM degradation where oxygen concentrations are low.

During most cruises to the basin in 2005, bottom water oxygen concentrations were found to be near or below detection limits. Bottom waters of the SBB are generally

retained in the basin by a sill at 440 m below the surface. Therefore, waters below the shelf do not freely exchange with outside sources unless a flushing event replenishes bottom waters in the basin. In the absence of flushing events, remineralization of sinking particles leads to hypoxic, and sometimes anoxic conditions near the bottom.

N* has been used as a proxy for assessing nitrogen balance in various marine environments (Gruber and Sarmiento, 1997). N* is defined as the deviation from the Redfield Ratio of nitrogen and phosphorous where N* in its simplified form = (NO_3 + NO_2) – 16P. Strongly negative values for N* identify regions where denitrification influences the N-content of the water column. Positive N* values are indicative of DOM remineralization or N_2-fixation. Here N* was calculated for each of the quarterly cruises in 2005 to provide an index of the extent of denitrification in bottom waters of the SBB (Fig. 2.14).

During 0501 surface TOC was 81-82 μM C with a subsurface maximum of 107 μM C at 60m. Subsurface TOC values were extremely variable and showed little relationship to nutrient distributions. Outside the SBB, TOC profiles collected within the CCS during this year did not show this type of unusual vertical structure. However, other investigators have reported a similarly unusual structure for TOC profiles in Santa Monica Basin (Berelson, unpublished). TON values at the surface were 7 μM N, decreased with depth, and then increased to 5-7 μM N below 530 m. Strong denitrification was indicated by N* (-80) and hypoxic conditions below 470 m (Fig. 2.14). N* began to decrease dramatically around 430 m, which was coincident with a very high C:N (>100) for DOM. C:N in surface DOM is between 10 and 14, which is typical of open ocean values (Hopkinson and Vallino, 2005), and the large deviation in

C:N at 430 m in SBB suggests that N may have been removed from all pools.

During 0504 TOC was 103-126 μM C, TON was 7-11 μM N, and high [chl *a*] were indicative of vigorous primary production at the surface. Mid-depth (100-375 m) TOC concentrations were 45-60 μM C and were slightly lower than winter values. At these depths N* ranged from -8 to -14 and was slightly lower than in winter (N* = -5 to -12). At this time, nitrate and oxygen were replenished to the bottom 20-30 m of the basin. Either this indicated the beginning of the flushing event or the event did not completely replenish the basin. Both TOC and TON decreased in bottom waters relative to winter values.

By 0507, primary production had increased further however, TOC values were only 80-82 μM C and [chl a] was 1-3 μg/L. N* values were also close to 0 (-2 to -4). Basin bottom waters appeared to have mixed with overlying waters since nitrate at 540 m had increased from 0.6 μM to 22 μM. However, oxygen at the bottom remained low and N* decreased from spring values (-33 to -35) to -41 to -53 in the summer, indicating that denitrification was beginning to affect the basin.

During 0511 TOC and N* values throughout the water column were similar to 0507 values, except at the bottom. The TOC profile was also beginning to show greater variability in the subsurface region. Signs of strong denitrification were present just below the sill and it appeared that the basin was approaching conditions encountered in 0501. Denitrification was in full effect and very active as nitrite values were 3-7 μM N. During 0501 nitrite was only 2 μM indicating that denitrification, and potentially annamox, had already consumed much of the inorganic N. During 0511, active denitrification had begun to strip N, but nitrate levels were still non-zero (2-5 μM) and

N* was -61 to -67 at this time.

TOC and TON values reported here are comparable to DOC values reported by Hansell et al. (1993) in the Santa Monica Basin (SMB). However, C:N values in 2005 were slightly lower than those from SMB.

2.4.4 Comparison to 0411 and 0404

During 2004, TOC samples were taken aboard the spring (0404) and fall (0411) cruises to supplement studies presented in the following chapters. TOC data are plotted in Figure 2.15 are similar to 2005, although fall has slightly higher TOC values than in the spring. A more detailed description of this data is presented in Chapter 3.

2.5 CONCLUSION

This represents the most comprehensive study of organic matter fractions within the CalCOFI grid to date. In 2005, the CCS contained by the CalCOFI grid exhibited seasonal biogeochemical characteristics consistent with the previous 50+ years of CalCOFI data. The behavior of TOC, TON, POC, and PON in surface waters appears to be related to the extent of seasonal production, physical structure of the water column and position of the CC. For example, highest TOC accumulations were observed during 0507 when [chl a] and nitrate data supported increases in upwelling induced primary production. In addition, during 0507 TOC data also identified a relative depletion of TOC within the CC. In contrast to other studies, this study documented spatial and seasonal variations in TON, but accumulations were not as extensive as those observed

for TOC. In addition, during certain seasons (spring and summer, in particular) TON accumulated in regions where TOC was also accumulating; but the reverse was not always true and so, there was some spatial decoupling of TOC and TON. As expected POC and PON exhibited similar patterns of variability. Finally, observations summarized here suggest that DOC and POC dynamics are often de-coupled. Entrainment of TOC into subsurface waters was most apparent during summer 2005. However, increased deep water TOC along Line 90 could not be explained by mixing alone and supported the intriguing possibility that a TOC-rich subsurface water mass entered the southeastern region of the CCS along the slope at this time. The depth of the TOC maximum is consistent with the depth of the Undercurrent.

In addition, TOC and TN distributions in the SBB were also explored. TOC and TON exhibited some unusual subsurface dynamics, but the cause of these variations is not well known. Patchy TOC distributions may result from the presence of hydrocarbon seeps in this region. Alternatively, the low oxygen concentration and associated microbial processes may be responsible for the unusual behavior of total organic matter pools.

These data summarize the first year of observations. Data collected during 2006 and 2007 should enable a more comprehensive assessment of the initial observations presented here.

Table 2.1 The computed averages for coastal, offshore, and mid-CC regions. Coastal stations are inside station 55, offshore are outside station 90, and mid-CC are stations 60-80.

	TOC	+/-	TON	+/-	Chl-a	+/-	PON	+/-	POC	+/-
0501										
Average	65.5	3.1	5.5	2.5	0.7	0.5	0.8	0.4	5.8	2.7
Coastal	66.2	3.7	6.2	2.4	0.9	0.5	0.9	0.4	6.8	2.9
Offshore	63.5	2.1	4.8	2.0	0.3	0.2	0.5	0.3	3.5	1.6
Mid-CC	66.5	2.7	5.2	2.9	0.7	0.4	1.0	0.4	6.2	2.0
0504										
Average	76.5	13.4	5.2	2.1	1.1	1.5	1.6	1.5	10.9	10.7
Coastal	86.4	9.7	6.2	1.6	2.0	1.7	2.4	1.4	17.3	9.4
Offshore	67.2	5.1	4.1	1.8	0.1	0.1	0.4	0.2	2.2	0.6
Mid-CC	72.7	14.7	4.9	2.5	0.8	1.0	1.3	1.5	7.8	9.6
0507										
Average	81.1	15.5	5.6	1.6	1.9	3.8	2.4	2.3	12.9	13.5
Coastal	92.4	15.1	6.8	1.7	4.1	5.0	3.5	2.4	22.5	15.3
Offshore	73.1	8.8	4.5	0.7	0.1	0.0	1.1	1.5	3.3	1.0
Mid-CC	72.6	10.6	4.8	0.8	0.4	0.5	1.8	2.2	6.4	3.7
0511										
Average	70.6	11.2	6.9	2.9	1.6	3.0	1.5	1.6	11.0	12.4
Coastal	76.0	12.4	8.5	3.6	2.4	3.7	2.2	2.0	16.5	15.0
Offshore	64.5	4.2	5.5	1.0	0.2	0.1	0.5	0.1	2.9	1.1
Mid-CC	66.8	9.5	5.8	1.4	1.5	2.8	1.3	1.3	8.2	8.6

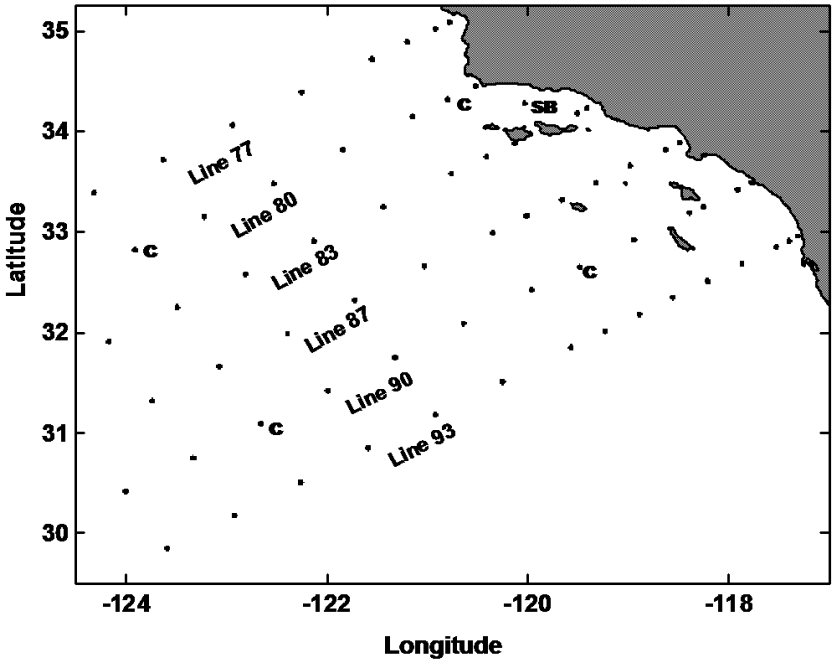

Figure 2.1 CalCOFI station grid with line designations. Cardinal stations (C) and Santa Barbara Basin (SB) are also denoted.

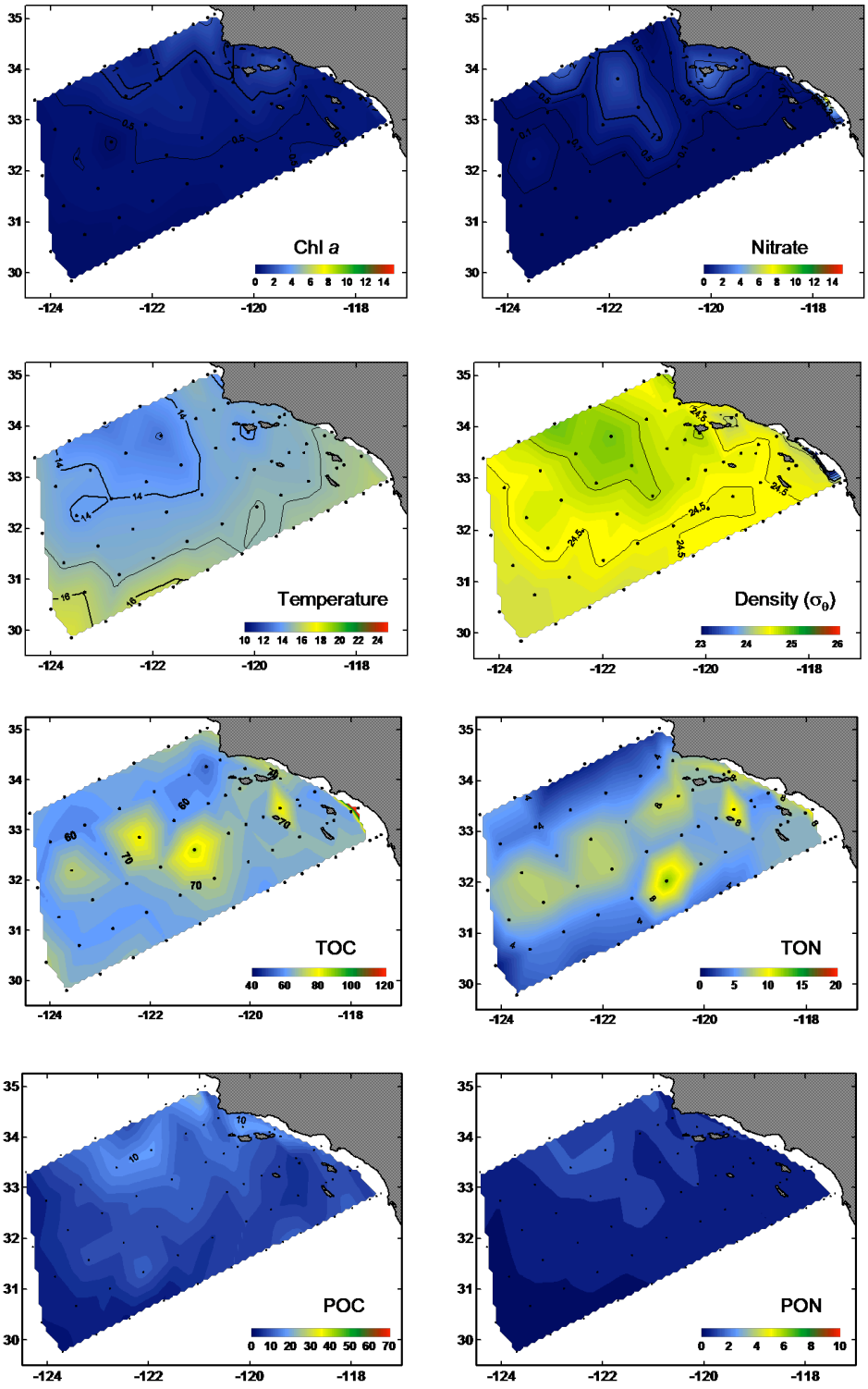

Figure 2.2 0501 surface plots of chl *a*, nitrate, temperature, density (σ_θ), TOC, TON, POC, and PON.

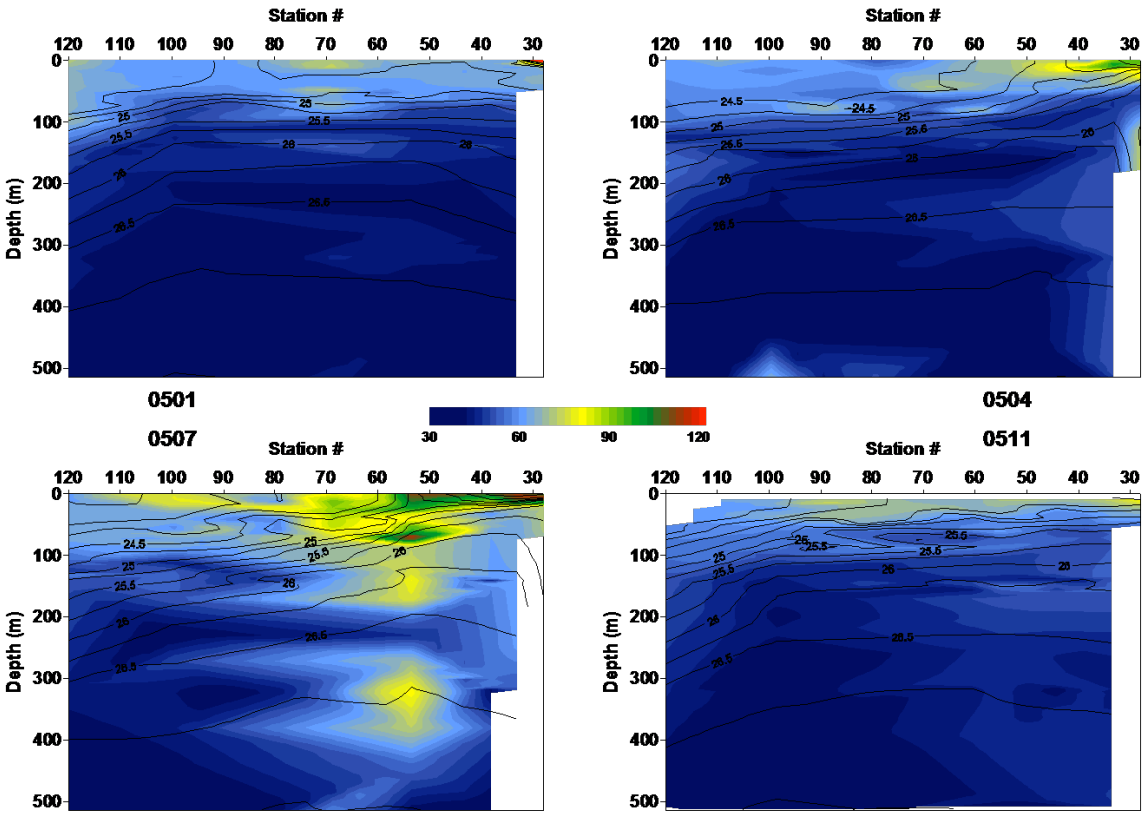

Figure 2.3 Seasonal contour profiles of line 90 with density lines overlaid.

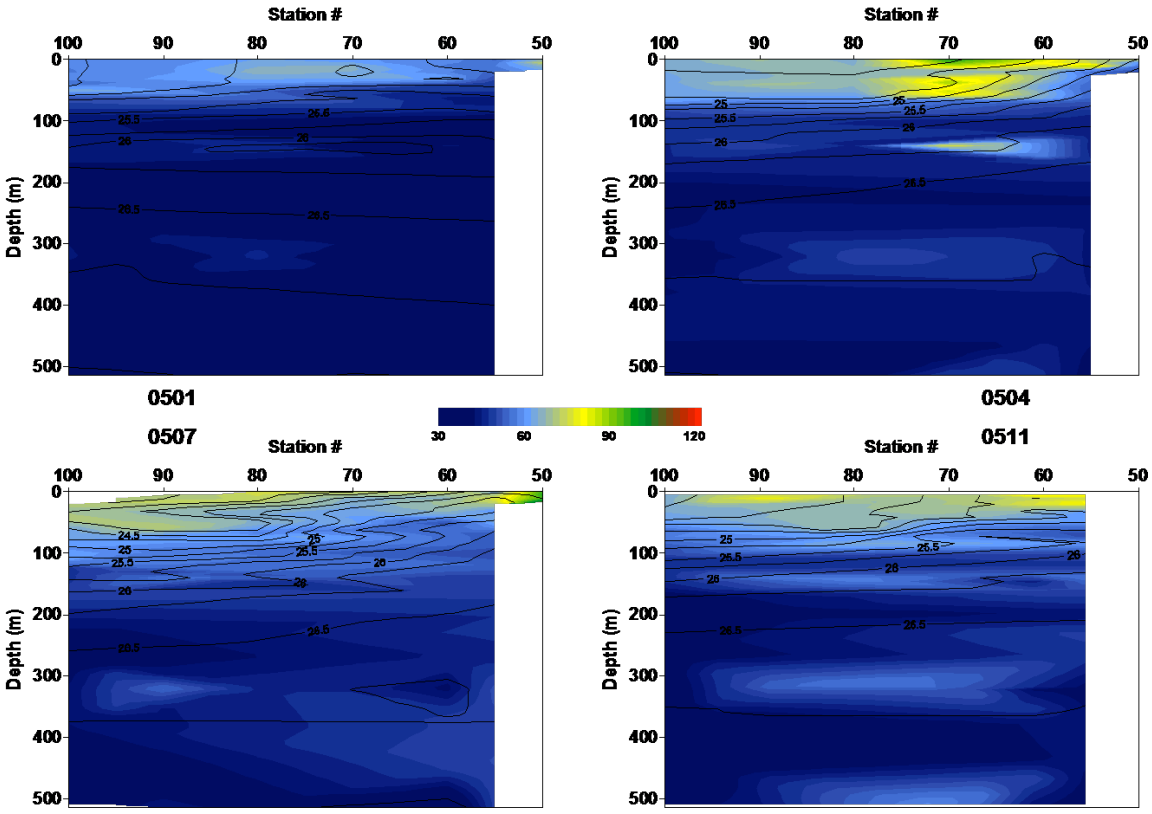

Figure 2.4 Seasonal contour profiles of line 80 with density lines overlaid.

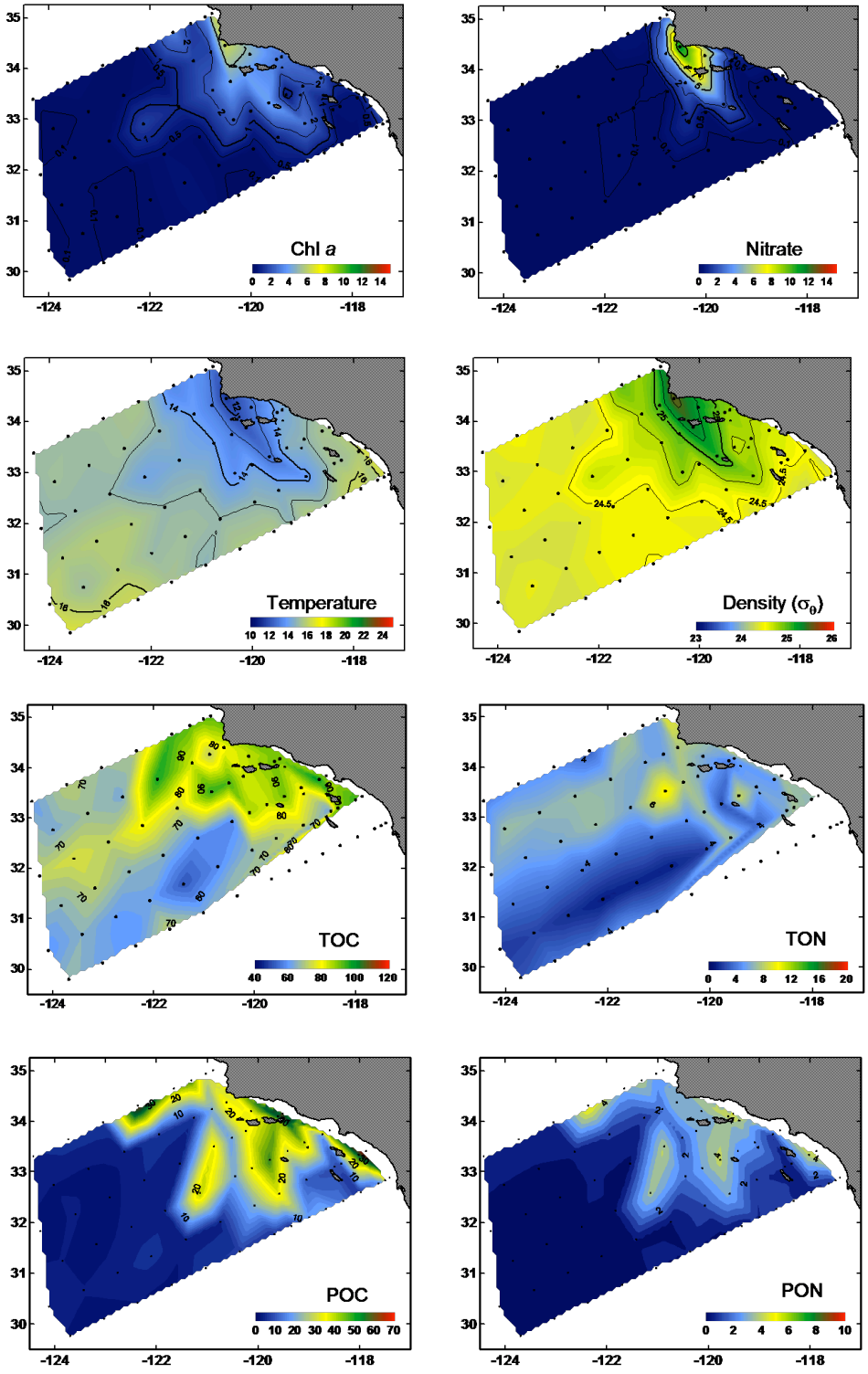

Figure 2.5 0504 surface plots of chl *a*, nitrate, temperature, density (σ_θ), TOC, TON, POC, and PON.

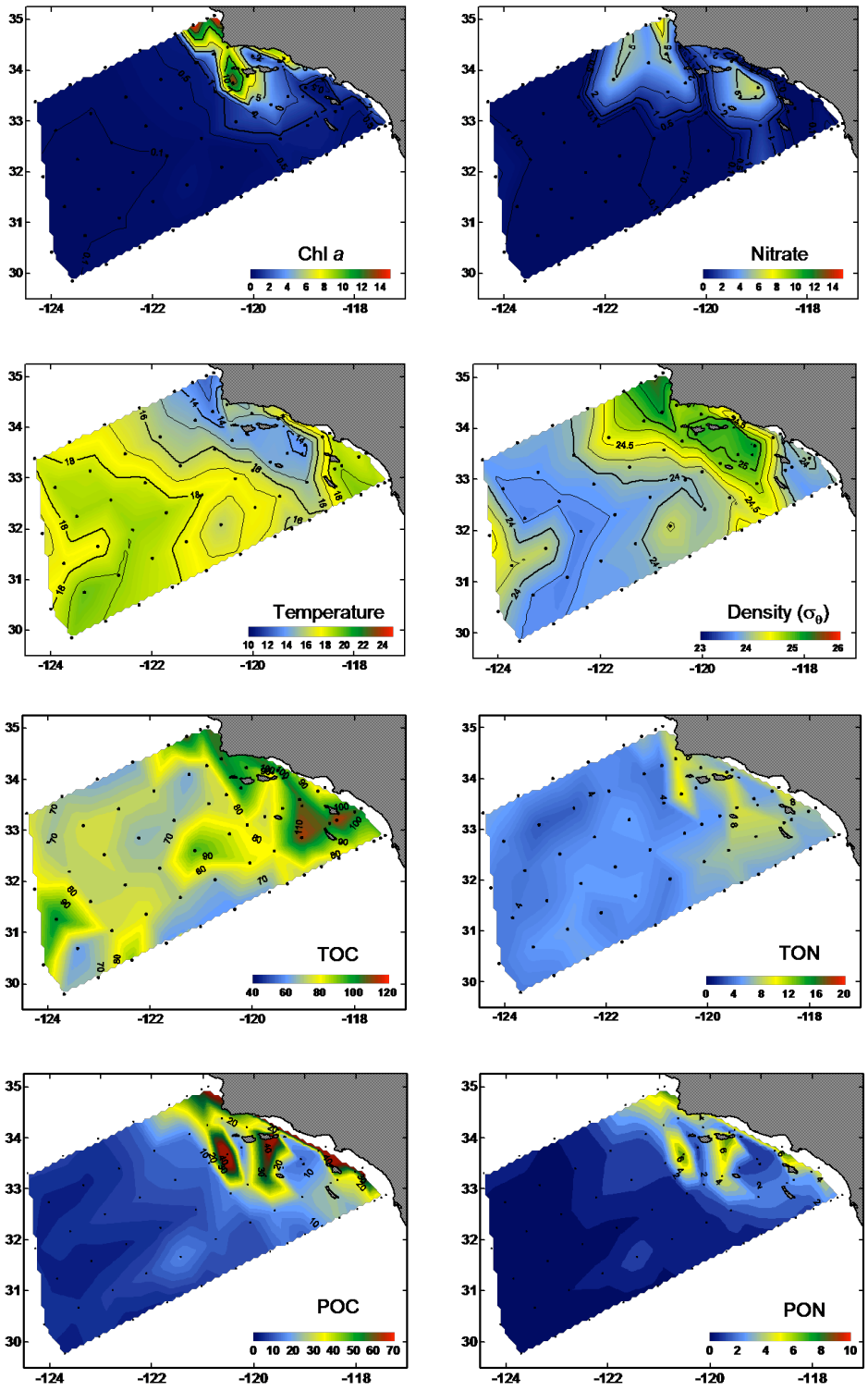

Figure 2.6 0507 surface plots of chl *a*, nitrate, temperature, density (σ_θ), TOC, TON, POC, and PON.

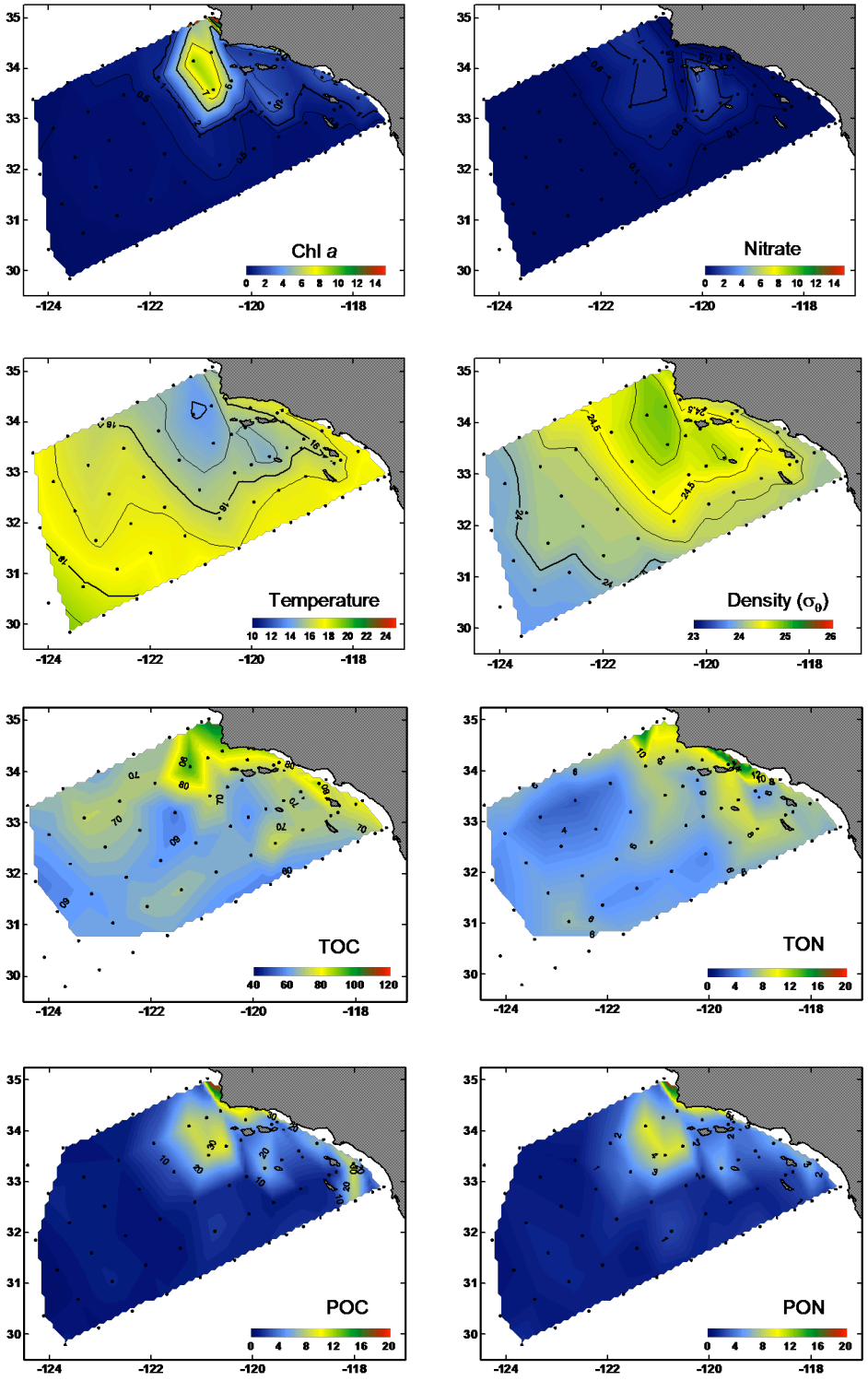

Figure 2.7 0511 surface plots of chl *a*, nitrate, temperature, density (σ_θ), TOC, TON, POC, and PON.

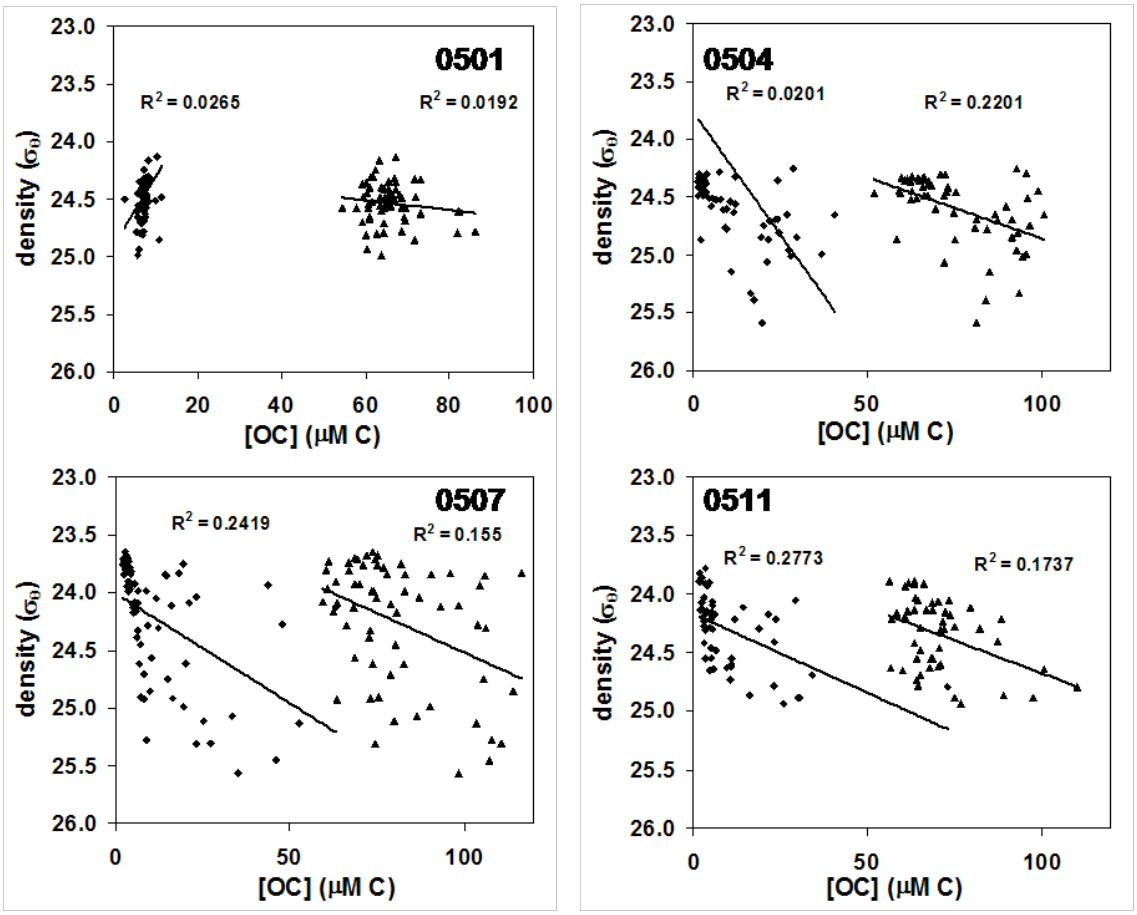

Figure 2.8 Seasonal surface values of POC (diamonds) and TOC (triangles) against density.

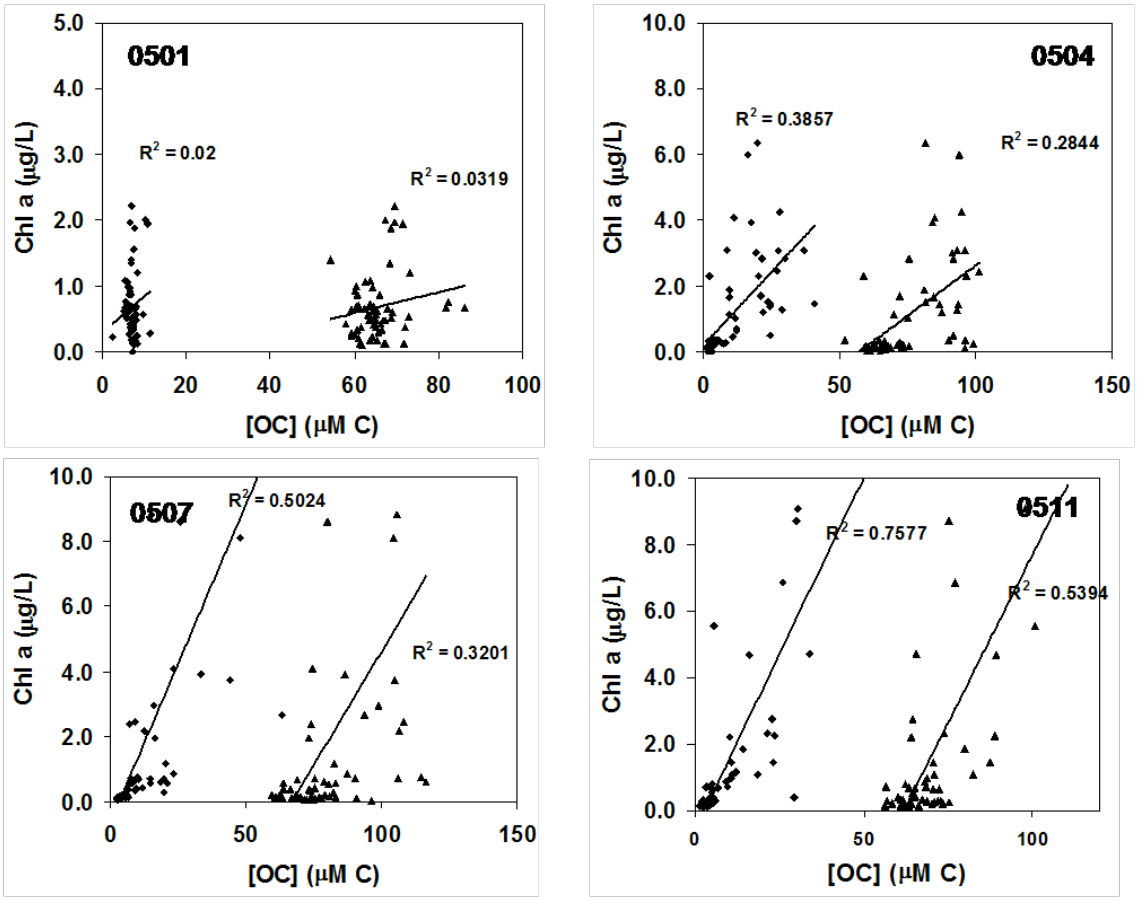

Figure 2.9 Seasonal surface values of POC (diamonds) and TOC (triangles) against Chl a concentration (µg L^{-1}).

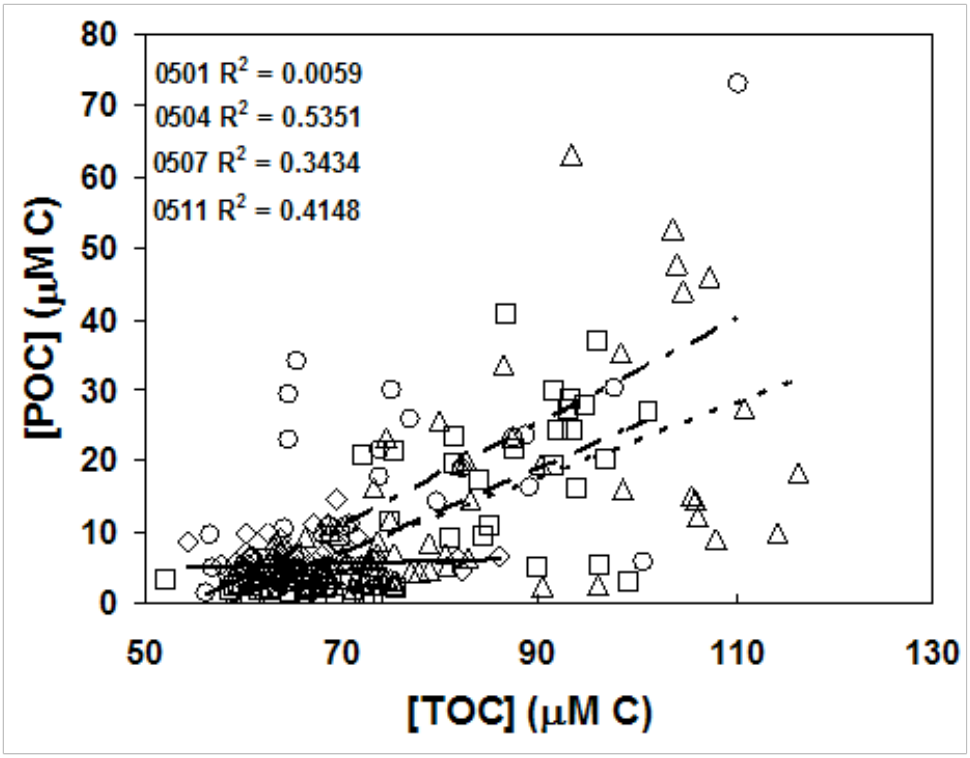

Figure 2.10 Seasonal concentration plots of POC (diamonds) vs. TOC (triangles) for 0501 (diamonds), 0504 (squares), 0507 (triangles), and 0511 (circles).

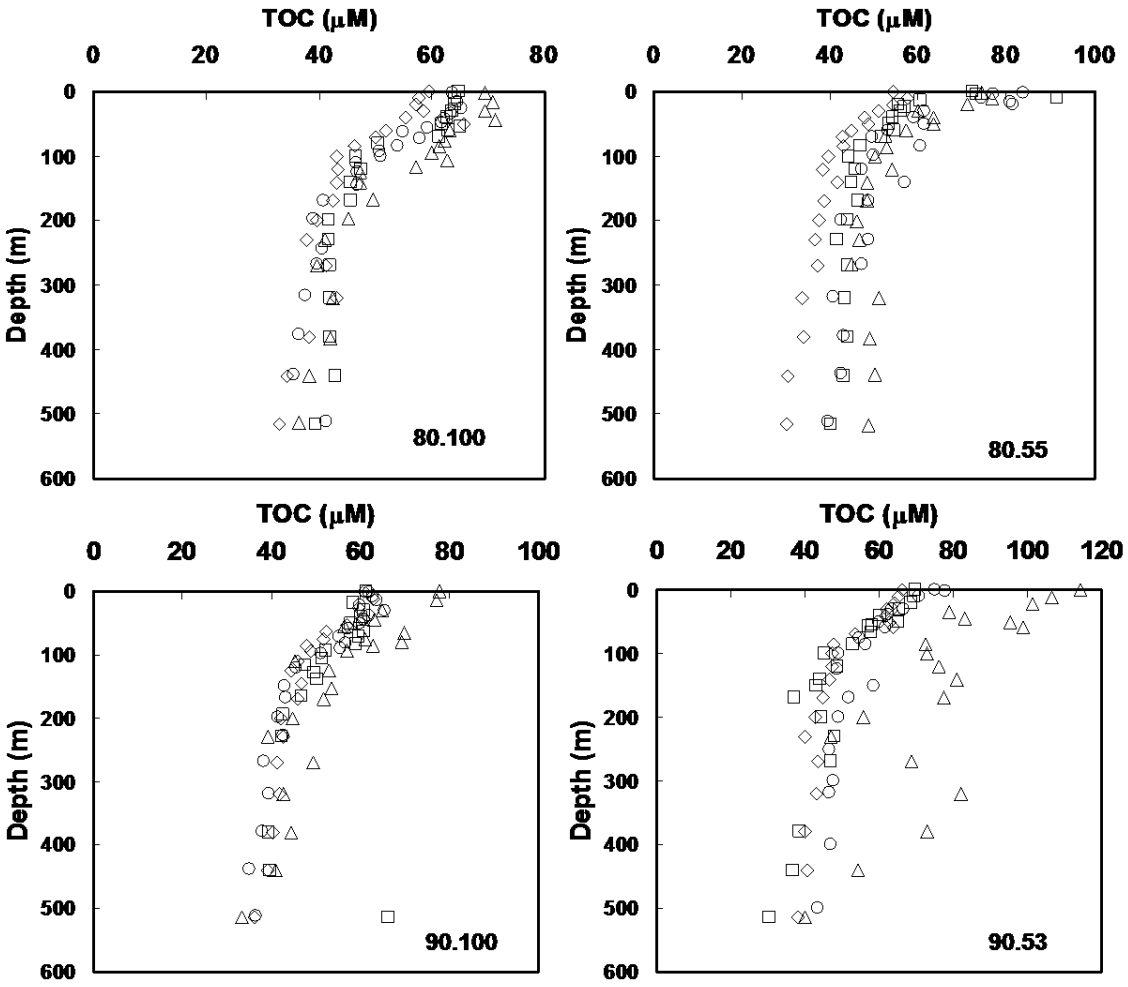

Figure 2.11 Cardinal stations with seasonal profiles 0501 (diamonds), 0504 (squares), 0507 (triangles), and 0511 (circles).

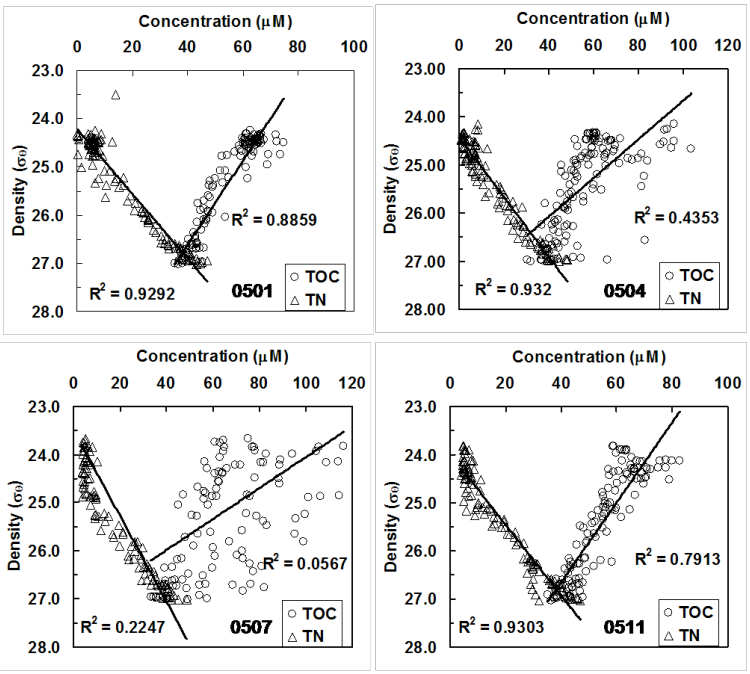

Figure 2.12 Density plot vs. TOC and TN along line 90.

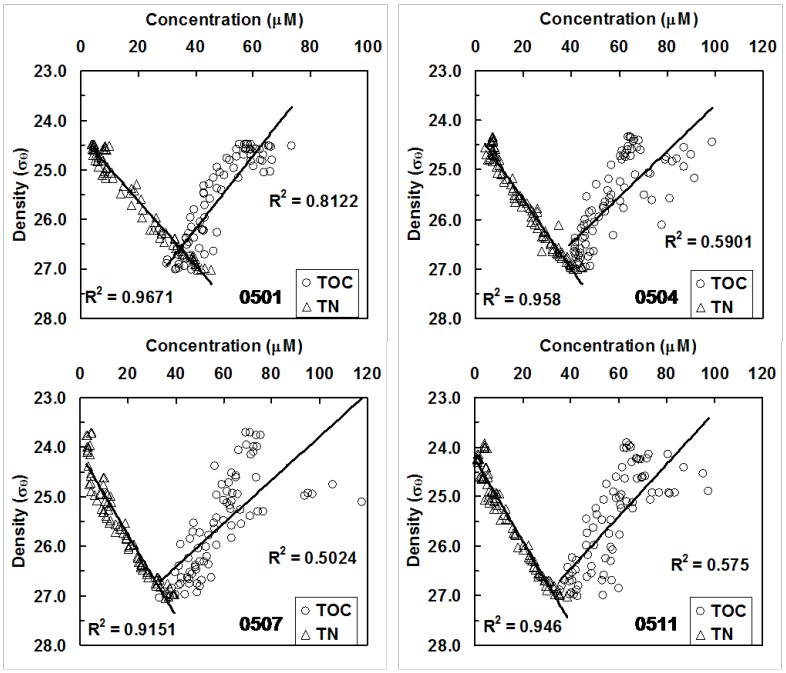

Figure 2.13 Density plot vs. TOC and TN along line 80.

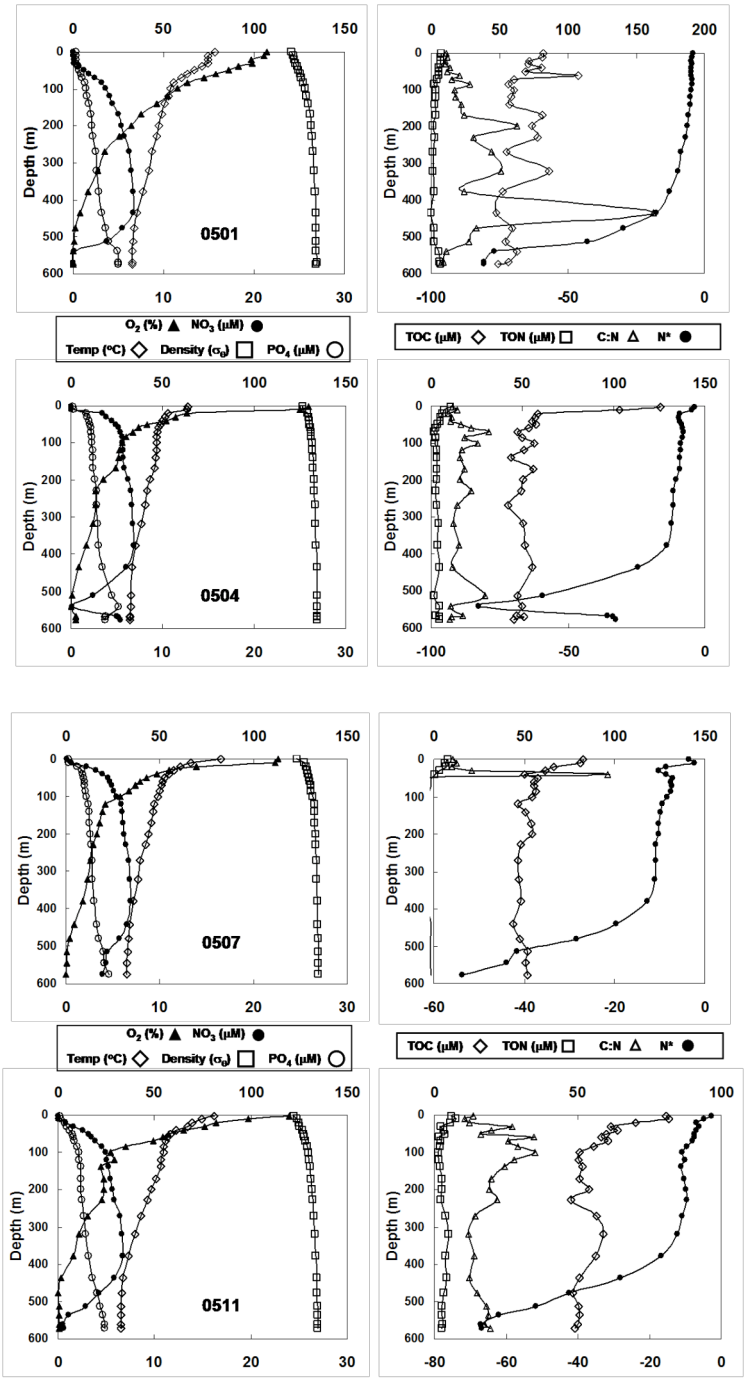

Figure 2.14 Santa Barbara Basin profiles for 0501, 0504, 0507, 0511. On the left, dark symbols are plotted on the primary (top axis) and open symbols plotted on the secondary (bottom) axis. On the right, all values are plotted on the primary (top) axis except N*.

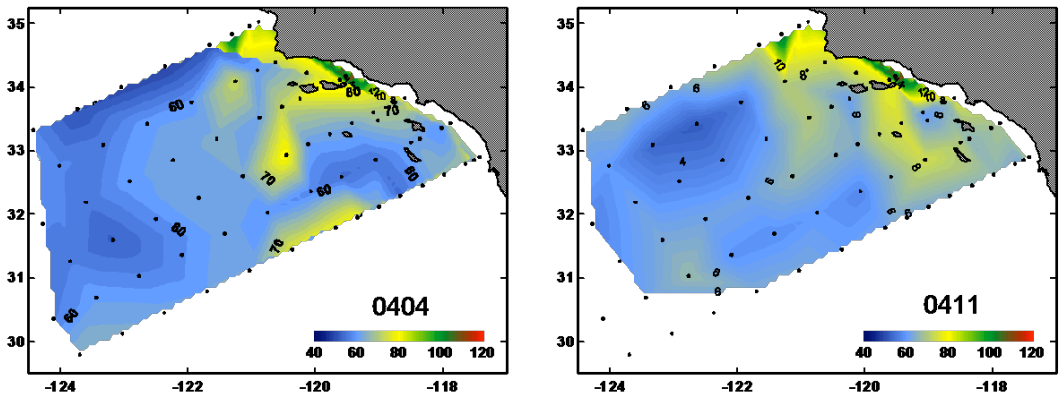

Figure 2.15 TOC surface plots for 0404 and 0411.

2.6 APPENDIX

This section reports the data which was used to construct plots, contours, and charts depicted in Chapter 2. Blanks or gaps in the data represent no sample taken or unreliable data. Ancillary hydrographic data can be found on the web at calcofi.org.

2.6.1 Cruise 0404 (April 2004)

Station	TOC (μM C)	TON (μM N)	Station	TOC (μM C)	TON (μM N)
77.49			87.70	64.0	4.8
77.51			87.80		
77.55			87.90		
77.60	51.9	5.4	87.100	53.5	2.9
77.70	49.1	3.4	87.110	57.8	3.3
77.80			90.27	70.7	5.1
77.90	51.3	3.2	90.30	59.1	3.9
77.100	58.4	4.8	90.35	63.0	3.3
80.51			90.37	63.2	3.7
80.55	62.1	5.2	90.45	56.2	3.8
80.60	74.0	7.2	.90.53	55.3	5.6
80.70	60.7	5.8	90.60	58.4	6.1
80.80	61.5	6.0	90.70	60.1	4.7
80.90	57.4	3.8	90.80	63.1	5.6
80.100	58.4	3.1	90.90		
82.47	70.1	6.0	90.100		
83.41			90.110	61.6	4.9
83.42	87.0	7.8	90.120	57.6	5.1
83.51			93.27		
83.55	73.5	5.2	93.28	75.5	
83.60	63.1	2.6	93.30	69.0	
83.70	64.6	5.2	93.35	66.7	
83.80			93.40		
83.90	61.5	4.9	93.45	55.1	
83.100	56.4	3.4	93.50	65.7	
83.110		0.0	93.55	80.1	4.1
87.33			93.60		
87.35	72.0	3.5	93.70	77.1	4.3
87.40	70.0	8.5	93.80	62.8	2.6
87.45	64.7	4.9	93.90	62.0	3.1
87.50			93.100	63.1	4.2
87.55	62.0	5.0	93.110		
87.60	82.6	5.0	93.120	66.5	2.6

2.6.2 Cruise 0411 (November 2004)

Station	TOC (µM C)	TON (µM N)	Station	TOC (µM C)	TON (µM N)
77.49			87.70	68.2	3.7
77.51	77.3	5.9	87.80	68.8	3.6
77.55	64.1	3.0	87.90	63.7	4.4
77.60	63.3	3.4	87.100	64.9	3.2
77.70	64.9	3.1	87.110	68.2	2.8
77.80	67.0	3.0	90.27	77.5	4.3
77.90	71.4	3.1	90.30	69.6	3.6
77.100	63.5	2.7	90.35	72.4	3.6
80.51	64.4	4.1	90.37	69.0	3.6
80.55			90.45	89.9	4.8
80.60	63.9	3.4	.90.53	69.5	3.6
80.70	63.5	3.4	90.60	69.4	3.4
80.80	68.6	3.8	90.70	60.0	3.8
80.90	63.6	3.1	90.80	57.9	3.0
80.100	63.7	2.8	90.90	64.0	2.9
82.47	65.3		90.100	70.6	3.5
83.41	68.7	3.1	90.110	68.4	4.0
83.42			90.120	68.7	3.3
83.51	69.9	3.5	93.27	73.8	3.2
83.55	64.8	3.3	93.28	74.4	2.9
83.60	72.9	4.6	93.30	76.4	3.7
83.70	69.5	4.8	93.35	73.6	2.9
83.80	62.6	3.0	93.40	69.6	3.1
83.90	62.8	3.2	93.45	68.9	3.1
83.100	67.2	3.2	93.50		
83.110	66.3	3.4	93.55	71.0	3.5
87.33	93.8	4.1	93.60	77.8	3.3
87.35	78.7	4.3	93.70	79.8	2.8
87.40	66.5	4.3	93.80	84.7	3.5
87.45		4.4	93.90	71.1	2.4
87.50			93.100	76.0	2.2
87.55			93.110	63.8	2.8
87.60	66.3	4.1	93.120	61.9	2.7

2.6.3 Cruise 0501 (January 2005)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
77.49	1%	71.6	2%	2.3	9.2	0.9	13.54	24.859	1.2	1.95
77.51	5%	69.5	6%	1.6	14.5	2.1	14.28	24.687	0.7	2.22
77.55	2%	68.7	6%	2.0	11.1	1.5	14.87	24.494	0	1.88
77.60	2%	69.4	3%	1.7	8.5	1.3	14.32	24.581	0.4	1.96
77.70	1%	64.4	5%	1.1	5.1	1.0	13.42	24.794	0.9	0.62
77.80	1%	63.7	4%	0.9	8.9	1.6	12.91	24.987	2.7	1.09
77.90	2%	60.9	4%	5.5	6.4	1.0	13.43	24.685	0.5	0.71
77.100	3%	64.3	3%	5.6	2.6	1.0	14.88	24.504	0	0.23
80.51	1%	60.7	2%	7.3	6.3	0.9	14.39	24.578	1	0.86
80.55	3%	54.4	5%	4.6	8.8	1.3	14.52	24.582	0.5	1.4
80.60	4%	59.2	5%	3.8	5.8	0.9	14.34	24.699	0.3	0.63
80.70	2%	60.3	4%	4.3	10.0	1.7	12.92	24.933	1.9	1
80.80	2%	62.7	6%	4.4	9.9	1.6	13.4	24.799	0.4	1.06
80.90	3%	57.8	8%	3.9	5.4	0.8	14.15	24.58	0	0.43
80.100	1%	59.4	2%	4.0	4.3	0.4	14.29	24.482	0	0.28
82.47					11.5	1.5				
83.41	4%	73.2	4%	9.3	6.0	0.7	14.63	24.332	1.3	1.2
83.42	3%	67.2	9%	6.7	11.1	1.1	14.69	24.139	1.7	2
83.51	2%	68.5	3%	6.5	7.2	1.0	13.8	24.787	3.1	1.34
83.55	5%	64.3	4%	8.7	7.7	1.2	14.18	24.712	1.1	0.98
83.60	2%	60.9	9%	8.4	6.6	0.9	14.47	24.661	0.4	0.66
83.70	2%	60.2	4%	6.3	7.2	1.1	13.42	24.816	1.2	0.92
83.80	5%	81.8	4%	8.0	6.4	1.0	13.84	24.796	0.9	0.67
83.90					4.6	0.6	13.996	24.609	0	0
83.100	4%	73.0	3%	8.0	4.6	0.6	13.89	24.624	0.4	0.53
83.110	3%	59.2	3%	6.3	2.6	0.3	14.73	24.368	0	0.24
87.33	2%	63.2	2%	8.6	7.6	0.9	15.12	24.16	1.2	0.56
87.35	1%	60.5	4%	4.4	5.4	0.9	15.19	24.456	0.1	0.7
87.40	0%	63.3	5%	4.7	3.3	0.6	14.77	24.554	0.3	0.55
87.45	4%	82.2	1%	10.8	4.6	0.8	14.58	24.608	0.4	0.76
87.50	4%	63.6	5%	6.0	5.0	0.9	14.66	24.594	0.4	0.72
87.55	1%	65.9	4%	5.5	6.6	1.0	14.77	24.565	0	0.87
87.60	10%	69.1	4%	5.9	4.9	0.8	14.5	24.655	0.2	0.6
87.70	5%	86.0	10%	5.7	6.6	1.2	14.1	24.779	1.1	0.68
87.80	6%	68.6	4%	8.0	5.1	0.9	14.25	24.576	0	0.51
87.90	3%	63.5	1%	7.3	5.3	0.7	14.37	24.489	0	0.47
87.100	3%	64.3	5%	7.8	5.3	0.7	14.48	24.576	0	0.48
87.110	3%	60.2	7%	6.8	2.7	0.4	14.79	24.364	0	0.26
90.27	2%		3%	11.5	13.8	1.8	14.91	20.934	9	1.56
90.30	2%	62.5	8%	6.5	5.0	0.7	15.51	24.25	0	0.65
90.35	4%	65.4	2%	6.3	5.3	0.8	15.28	24.411	0	0.62
90.37	3%	64.4	5%	6.5	4.5	0.7	15.28	24.417	0	0.69
90.45	3%	66.6	3%	5.8	3.5	0.5	14.78	24.516	0.1	0.34
.90.53	3%	66.3	13%	5.8	4.3	0.6	14.89	24.463	0	0.43

2.6.3 Cruise 0501 (continued)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
90.60	0%	64.7	2%	6.6	5.0	0.7	15.05	24.481	0	0.41
90.70	4%	72.1	3%	12.7	3.9	0.6	14.79	24.482	0	0.37
90.80	1%	65.6	4%	4.8	4.5	0.7	14.78	24.486	0	0.31
90.90	0%	61.4	6%	4.4	4.8	0.6	14.72	24.528	0	0.37
90.100	2%	60.8	10%	4.3	3.3	0.4	14.72	24.403	0	0.22
90.110	3%	61.3	4%	4.1	2.5	0.3	16	24.313	0	0.13
90.120	4%	71.7	5%	0.7	1.7	0.2	16.29	24.332	0	0.12
93.27					7.3	1.2	15.28	24.355	0.1	0.61
93.28					5.1	0.7	15.58	24.458	0	0.44
93.30	1%	67.2	5%	6.4	4.8	0.7	16.03	24.424	0	0.47
93.35	1%	67.9	2%	6.3	6.8	0.9	15.6	24.465	0	0.65
93.40	5%	66.7	2%	6.4	5.4	0.7	15.42	24.51	0	0.48
93.45	1%	65.0	4%	6.3	4.7	0.5	15.4	24.518	0	0.57
93.50					6.6	0.8	15.31	24.513	0	0.69
93.55	5%	65.2	6%	4.1	6.3	0.8	15.33	24.523	0	0.47
93.60	1%	60.6	3%	3.4	4.6	0.6	14.94	24.544	0	0.33
93.70	3%	65.0	2%	4.1	5.0	0.7	15	24.545	0	0.38
93.80	1%	63.7	3%	3.7	3.8	0.5	15.51	24.405	0	0.17
93.90	2%	61.9	4%	3.3	1.6	0.2	16.27	24.307	0	0.11
93.100	3%	67.2	11%	2.5	1.6	0.2	16.4	24.346	0	0.13
93.110	3%	65.4	7%	3.4	2.8	0.4	15.6	24.351	0	0.17
93.120	3%	67.0	1%	2.6	1.5	0.2	16.7	24.319	0	0.13

2.6.4 Cruise 0504 (April 2005)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
93.27					8.7	0.6	15.54	24.613	0	3.08
93.28					24.3	2.6	16.51	24.356	0.1	1.37
93.30					12.1	1.0	15.44	24.559	0	0.72
93.35					7.4	0.8	16.34	24.285	0.1	0.26
93.40					3.5	0.4	15.9	24.375	0.1	0.21
93.45					8.0	1.0	15.24	24.519	0	0.28
93.50					11.8	1.3	15.77	24.321	0	0.64
93.55					10.7	1.0	14.81	24.542	0	0.46
93.60					2.8	0.3	15.61	24.419	0	0.15
93.70					3.1	0.3	15.62	24.407	0	0
93.80	3%	64.2	6%	4.2	4.4	0.5	14.84	24.515	0	0.34
93.90	6%	72.7	3%	4.9	3.8	0.6	15.37	24.458	0.1	0.21
93.100	3%	60.7	5%	3.6	1.6	0.2	16.3	24.352	0.1	0.08
93.110	1%	62.8	5%	3.8	1.9	0.2	16.16	24.328	0	0.1
93.120	4%	67.6	7%	2.7	1.4	0.1	16.71	24.365	0	0.13
90.28	15%	93.1	3%	7.2	28.8	4.0	16.78	24.254	0	1.29
90.30	34%	96.0	7%	6.5	5.6	0.8	15.69	24.506	0	0.35
90.35	22%	89.9	10%	7.2	5.3	1.0	15.22	24.579	0	0.36
90.37	3%	62.5	5%	6.1	6.3	1.0	15.39	24.522	0	0.3
90.45	2%	75.3	7%	3.2	21.4	3.2	13.76	24.865	0.2	2.84
.90.53	4%	69.8	6%	7.3	9.5	1.4	14.51	24.601	0.1	1.12
90.60	14%	67.5	25%	2.1	2.6	0.5	14.96	24.485	0	0.18
90.70	2%	59.1	17%	1.2	2.6	0.4	15.04	24.449	0	0.14
90.80	8%	52.0	8%	1.3	3.4	0.4	15.25	24.467	0	0.36
90.90	6%	62.8	13%	1.1	2.4	0.5	15.08	24.448	0.1	0.16
90.100	2%	61.4	1%	1.6	2.1	0.2	15.74	24.344	0	0
90.110	4%	66.0	13%	2.3	2.6	0.3	15.18	24.417	0	0.15
90.120	1%	59.8	3%	2.0	2.7	0.2	16.08	24.333	0.1	0.02
87.33	6%	86.8	5%	7.1	40.8	4.8	15.55	24.659	0	1.44
87.35	8%	101.0	5%	5.1	26.9	3.0	15.43	24.653	0	2.43
87.40	4%	81.0	4%	5.8	9.2	1.0	14.91	24.771	0	1.87
87.45	1%	91.8	1%	8.5	24.4	2.7	14.7	24.7	0.2	0.49
87.50	6%	85.0	5%	2.8	11.0	1.7	12.7	25.15	2.3	4.08
87.55	5%	87.6	3%	7.0	21.9	4.2	14.1	24.708	0.2	1.2
87.60	3%	58.6	5%	4.5	2.0	0.3	13.58	24.868	1.1	2.3
87.70	3%	59.5	4%	3.7	2.9	0.6	15	24.471	0	0.16
87.80	6%	66.2	5%	3.3	1.5	0.2	15.04	24.491	0.2	0.31
87.90	3%	66.5	5%	3.5	2.1	0.3	15.73	24.349	0	0.08
87.100	2%	72.5	4%	3.9	1.6	0.2	15.81	24.302	0	0.09
87.110	8%	66.2	4%	3.7	2.0	0.3	15.64	24.327	0	0.13
82.47	2%	95.8	3%	8.1	35.4	4.6	15.76	24.296	0	0.09
83.41	4%	91.5	4%	5.0	29.8	4.6	14.318	24.85	0.9	2.84
83.42	8%	91.5	10%	4.2	19.4	2.9	14.27	24.849	0.3	3.02
83.51	7%	84.0	5%	6.4	17.4	2.7	12.19	25.396	7.8	3.95

2.6.4 Cruise 0504 (continued)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
83.55	13%	94.7	3%	7.6	27.9	4.2	13.18	25.009	2.6	4.25
83.60	3%	96.6	2%	10.5	20.2	3.0	14.04	24.749	0.1	2.32
83.70	4%	74.8	3%	6.1	11.5	2.0	14.45	24.638	0.1	1.02
83.80	3%	81.4	2%	7.0	23.5	3.8	14.15	24.693	0.1	1.53
83.90	3%	73.1	4%	6.1	2.7	0.4	15	24.423	0	0.2
83.100	8%	75.6	6%	5.5	2.5	0.4	15.01	24.454	0	0.16
83.110	5%	73.2	8%	5.7	1.4	0.2	14.9	24.414	0	0.12
80.51	2%	81.4	1%	7.6	19.7	3.2	11.48	25.587	11.7	6.35
80.55	3%	72.2	4%	5.6	21.1	3.6	13.13	25.065	3.6	1.71
80.60	5%	84.3	4%	5.8	9.6	2.9	13.94	24.781	0.1	1.65
80.70	7%	99.0	7%	6.6	3.2	1.0	14.87	24.448	0	0.24
80.80	2%	65.0	5%	6.9	3.1	0.7	15.26	24.36	0	0.19
80.90	2%	68.5	6%	6.6	2.2	0.5	15.1	24.399	0	0.14
80.100	2%	64.7	3%	7.0	1.7	0.4	15.24	24.335	0	0
77.49	4%	93.8	4%	9.3	16.3	2.8	12.64	25.335	2.5	5.99
77.51	4%	92.9	1%	5.8	27.4	4.0	13.35	24.959	1.5	3.08
77.55	6%	93.4	15%	5.6	24.5	3.1	13.76	24.804	0.4	1.47
77.60	4%	95.9	2%	7.5	36.8	5.3	13.27	24.992	0.5	3.1
77.70	3%	70.9	14%	3.3	3.6	0.9	15.54	24.308	0	0.2
77.80	15%	67.8	5%	4.8	3.5	0.8	15.24	24.388	0	0.19
77.90	4%	72.1	4%	4.9	2.8	0.8	15.18	24.486	0	0.29
77.100	1%	62.7	5%	4.3	2.0	0.5	15.13	24.369	0	0.16
93.27					8.7	0.6	15.54	24.613	0	3.08
93.28					24.3	2.6	16.51	24.356	0.1	1.37
93.30					12.1	1.0	15.44	24.559	0	0.72
93.35					7.4	0.8	16.34	24.285	0.1	0.26

2.6.5 Cruise 0507 (July 2005)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
93.27	4%	98.6	5%	8.0	15.9	2.2	17.91	24.114	0	2.97
93.28	2%	87.3	4%	6.4	23.2	2.9	18.23	24.037	0	0.85
93.30	4%	83.1	10%	5.8	14.5	2.2	18.94	23.847	0.7	0.58
93.35	2%	82.1	8%	5.6	19.5	2.9	19.48	23.749	0.1	0.28
93.40	1%	75.0	8%	5.9	11.7	1.2	18.3	24.047	0	0.42
93.45	2%	68.8	6%	5.9	10.1	1.1	16.04	24.568	0.8	0.69
93.50	6%	72.9	4%	5.5	6.0	0.6	16.18	24.393	1.9	0.4
93.55	3%	78.8	4%	6.7	8.3	0.8	15.43	24.708	0.8	0.59
93.60	19%	62.5	5%	5.6	5.9	0.6	16.83	24.167	0	0.21
93.70	13%	59.6	3%	5.4	5.5	0.6	17.13	24.085	0	0.21
93.80	8%	61.0	7%	4.2	4.0	0.4	17.38	23.969	0.1	0.14
93.90	12%	60.6	6%	4.5	2.7	0.3	18.24	23.808	0	0.15
93.100	20%	90.4	7%	5.6	2.3	0.2	18.6	23.843	0	0.12
93.110	8%	68.6	6%	4.4	3.4	0.3	18.94	23.704	0	0.13
93.120	7%	66.9	6%	4.1	2.3	0.2	19.3	23.747	0	0.09
90.27	4%	104.7	5%	7.0	43.9	5.1	18.66	23.939	0	3.74
90.30					21.1	3.1	18.07	24.094	0.1	0.58
90.35	6%	116.4	2%	6.2	18.2	2.7	18.97	23.832	0	0.62
90.37	5%	105.8	6%	7.9	14.6	2.4	18.89	23.856	0	0.71
90.45					12.2	2.5				
.90.53	4%	114.3	5%	7.3	9.8	8.5	14.99	24.86	1.4	0.75
90.60	6%	80.8	5%	6.6	5.3	4.6	16.94	24.174	0	0.19
90.70	9%	83.0	8%	4.1	6.5	5.6	17.47	23.992	0	0.14
90.80	6%	66.2	6%	5.1	9.4	8.0	15.96	24.286	0.2	0.38
90.90	5%	70.2	3%	4.3	5.5	4.7	17.67	23.926	0.1	0.14
90.100	6%	77.8	9%	5.5	4.4	3.8	18.48	23.845	0	0.09
90.110	4%	75.3	1%	5.0	3.2	2.8	19.09	23.678	0.1	0.09
90.120	5%	61.1	10%	4.0	3.8	3.3	19.74	23.736	0	0.09
87.33	5%	93.4	0%	9.6	63.2	7.8	17.94	24.119	0.1	2.66
87.35	15%	73.7	7%	5.5	8.8	1.3	18.48	23.986	0	0.37
87.40	4%	80.5	5%	5.7	7.0	1.0	16.82	24.455	0.2	0.54
87.45	7%	107.9	7%	8.9	8.9	1.2	13.54	25.279	6.5	2.45
87.50	1%	86.5	4%	5.1	33.5	4.9	14.22	25.076	5.4	3.93
87.55	1%	73.3	5%	5.0	16.3	1.8	14.39	24.92	3	1.96
87.60					9.1	0.7				
87.70	5%	78.9	4%	4.9	4.7	0.6	16.55	24.102	0	0.22
87.80					3.5	0.3	17.51	23.946	0.1	0.09
87.90	3%	96.1	5%	5.2	2.7	0.3	18.08	23.839	0.1	0.05
87.100	2%	71.2	10%	4.2	2.1	0.2	18.88	23.764	0	0.07

2.6.5 Cruise 0507 (continued)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
87.110	4%	73.7	2%	4.0	2.7	0.2	19.02	23.648	0	0.08
83.41	3%	104.1	5%	5.8	47.7	7.6	17.33	24.282	0	8.12
83.42	3%	106.2	4%	9.2	12.2	1.5	17.22	24.312	0	2.18
83.51	6%	79.9	3%	5.7	25.5	4.3	14.21	25.116	4.6	8.61
83.55	1%	103.6	3%	10.7	52.7	7.4	13.96	25.133	0.3	14.23
83.60	4%	75.4	2%	3.9	7.1	1.2	14.37	24.91	4.2	0.71
83.70	3%	73.3	4%	5.3	6.5	1.2	16.15	24.33	2.3	0.37
83.80	5%	68.3	3%	4.8	5.2	1.0	16.05	24.135	0	0.16
83.90	4%	66.8	11%	4.6	4.4	0.3	18.24	23.815	0	0.1
83.100	2%	74.6	5%	4.0	3.5	0.2	18.85	23.712	0	0.06
83.110	3%	72.1	11%	4.1	2.9	0.3	18.85	23.682	0.1	0.08
82.47	2%	82.6	5%	7.7	20.0	3.2	16.53	24.623	0.6	1.19
80.51	2%	105.6	3%	9.5	15.0	2.3	15.79	24.756	0.2	8.82
80.55	2%	74.5	3%	3.9	23.3	3.6	13.13	25.31	5.5	4.08
80.60	4%	63.5	3%	4.0	8.2	1.5	14.7	24.93	4	0.58
80.70	4%	73.8	2%	4.7	6.9	1.0	15.68	24.618	5.1	2.38
80.80	2%	74.1	9%	3.1	3.6	0.5	17.1	23.992	0	0.18
80.90	18%	75.3	3%	2.9	2.7	0.4	18.46	23.765	0	0.09
80.100	7%	69.3	5%	4.4	2.5	0.4	18.74	23.715	0.1	0.1
77.49	5%	107.4	4%	7.8	46.1	7.9	13	25.452	5.5	15.92
77.51	11%	98.5	1%	5.6	35.3	6.1	12.32	25.566	8.8	11.33
77.55	2%	110.9	3%	6.8	27.3	4.9	13.39	25.308	3.5	15.72
77.60	4%	90.3	4%	4.4	19.5	3.6	14.25	24.99	5.5	0.7
77.70	3%	63.5	15%	4.6	6.5	1.1	16.06	24.089	0	0.38
77.80	3%	63.3	6%	5.0	3.7	0.4	17.1	23.906	0.1	0.14
77.90	9%	68.6	5%	5.1	4.4	0.6	17.34	23.924	0	0.2
77.100	6%	77.0	4%	5.8	4.2	0.6	18.17	23.785	0	0.15

2.6.6 Cruise 0511 (November 2005)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
77.49	3%	110.3	4%	11.8	73.1	10.2	14.72	24.79	17.51	0
77.51	3%	100.7	4%	10.1	5.8	1.3	15.31	24.64	5.55	0
77.55	4%	89.1	2%	17.4	16.2	2.7	14.12	24.86	4.70	0.8
77.60	3%	68.1	5%	8.2	9.5	1.5	14.82	24.63	0.89	1.4
77.70	1%	68.3	25%	7.8	3.5	0.7	15.20	24.56	0.74	0.6
77.80	4%	63.5	9%	5.9	3.2	0.6	15.91	24.42	0.70	0
77.90	3%	68.8	10%	6.5	2.3	0.4	16.57	24.08	0.27	0
77.100	2%	66.3	10%	6.1	3.5	0.6	17.73	23.91	0.16	0
80.51	4%	87.4	8%		23.1	2.8	16.22	24.41	1.46	0.1
80.55	3%	77.1	3%		26.0	3.3	14.01	24.93	6.88	0.9
80.60	6%	97.8	6%	7.8	30.4	4.7	13.94	24.89	9.09	1.1
80.70	5%	71.0	5%	4.0	11.0	1.4	15.22	24.60	1.08	0.5
80.80	3%	71.9	5%	3.0	3.0	0.4	16.63	24.24	0.28	0
80.90	4%	72.6	10%	3.5	5.2	0.7	16.32	24.15	0.28	0
80.100	4%	63.6	4%	5.0	4.3	0.5	17.14	23.92	0.22	0
82.47					28.6	2.8				
83.41	1%		4%	18.9	12.1	1.7	17.09	24.22	1.18	0.1
83.42	5%	73.8	3%	9.7	17.8	2.0	16.19			
83.51	4%	64.2	5%	5.4	10.4	1.7	14.90	24.74	2.20	2.2
83.55	4%	65.5	5%	6.7	34.1	4.5	15.04	24.69	4.74	0.1
83.60	4%	75.2	3%	7.7	30.0	4.5	14.24	24.89	8.74	1.1
83.70	3%	56.7	3%	5.3	9.5	2.1	15.13	24.63	0.74	1
83.80										
83.90										
83.100										
83.110	2%	56.3	4%	4.4	1.6	0.3	17.80	23.89	0.15	0.1
87.33	1%	73.9	6%	8.8	21.4	3.1	17.29	24.17	2.31	0
87.35	6%	88.7	4%	6.2	23.7	2.8	17.07	24.21	2.24	0
87.40	3%	68.9	6%	5.1	10.8	1.8	15.70	24.55	1.00	0.2
87.45	1%	70.6	7%	8.7	10.9	1.6	15.54	24.62	1.44	1
87.50	2%	64.6	7%	8.1	23.1	3.1	14.53	24.78	2.77	1.1
87.55	4%	60.1	3%	6.2	4.7	0.8	15.19	24.66	0.40	1.4
87.60	2%	64.4	2%	6.3	5.1	0.8	15.68	24.55	0.54	0.5
87.70	2%	62.5	2%	6.7	5.0	0.8	15.42		0.79	
87.80	9%	60.8	3%	5.6	2.7	0.8	16.91	24.20	0.31	0
87.90	4%	63.6	3%	5.7	2.8	0.5	17.15	24.14	0.22	0.1
87.100	8%	61.5	9%	5.9	3.8	0.7	16.86	24.15	0.32	0
87.110					1.7	0.4				
90.27	3%	79.8	3%	7.4	14.3	2.2	17.53	24.12	1.86	0.1
90.30					31.9	3.2				
90.35	4%	72.3	6%	6.4	5.3	1.0	16.82	24.30	0.66	0
90.37	3%	70.7	3%	7.7	5.1	1.1	16.02	24.46	0.64	0.1
90.45	4%	71.0	2%	8.6	3.6	0.6	17.05	24.30	0.27	0
.90.53	4%	75.1	10%	6.4	3.2	0.6	17.03	24.27	0.24	0

2.6.6 Cruise 0511 (continued)

Station	CV	TOC (μM C)	CV	TON (μM N)	POC (μM C)	PON (μM N)	Temp (^{O}C)	Density (σ_θ)	NO_3 (μM)	Chl-A (μg/L)
90.60	0%	65.3	11%	4.5	4.9	0.7	16.85	24.29	0.45	0
90.70	2%	65.3	7%	5.0	6.7	1.2	16.08	24.48	0.68	0.4
90.80	2%	68.7	6%	4.8	2.2	0.4	17.15	24.14	0.28	0
90.90	1%	67.2	4%	4.9			17.51	24.13	0.32	0
90.100	3%	62.0	5%	6.8	4.3	0.6	17.46	23.90	0.23	0
90.110	5%	63.6	2%	5.5	2.1	0.5	18.13	23.89	0.15	0
90.120					1.7	0.5				
93.27	7%	82.3	7%	8.0	18.8	2.6	16.70	24.30	1.10	0
93.28	9%	73.5	7%	6.1			17.71	24.06	0.21	0
93.30	8%	70.3	1%	6.0	5.5	0.7	17.66	24.07	0.20	0
93.35	3%	64.7	5%	6.6	29.5	1.2	17.79	24.06	0.39	0
93.40	4%		14%		5.5	0.8	17.75	24.09	0.40	0
93.45	2%		2%		5.2	0.6	17.23	24.18	0.28	0
93.50										
93.55	3%		7%		6.2	0.9	17.21	24.18	0.28	0.1
93.60	3%	56.9	0%	5.5	5.0	0.8	17.41	24.21	0.30	0
93.70	5%	58.5	5%	5.3	3.9	0.6	17.00	24.16	0.22	0
93.80	4%	60.9	5%	5.9	4.2	0.5	17.73	23.93	0.16	0
93.90	5%	64.0	4%	6.0	3.1	0.5	17.34	24.03	0.21	0
93.100	4%		8%		2.3	0.6	18.06	23.86	0.15	0
93.110	2%		12%		1.6	0.4	18.39	23.82	0.15	0
93.120	3%		6%		3.4	0.8	19.40	23.78	0.12	0.1

2.6.7 Line 90 (by cruise)

0501 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0504 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
28	0	2%	158.9	20.93	27	0	5%	95.9	24.16
28	5	4%	93.5	23.50	27	4	4%	91.8	24.28
28	10	1%	66.6	24.32	27	9	2%	94.8	24.66
28	20	3%	64.2	24.44	27	15	3%	82.6	25.23
28	30	3%	67.5	24.45	28	0	15%	93.1	24.25
28	40	2%	64.4	24.45	28	10	4%	81.4	24.77
28	50	1%	61.8	24.48	28	28	6%	81.6	25.45
30	0	2%	62.5	24.25	28	40	6%	55.5	25.77
30	10	1%	60.7	24.47	28	60	5%	47.6	25.90
30	23	1%	63.8	24.49	30	0	34%	96.0	24.51
30	55	5%	60.9	24.56	30	12	8%	84.1	24.92
30	140	3%	45.7	25.87	30	40	5%	63.7	25.52
30	320	4%	41.5	26.62	30	55	6%	51.2	25.68
30	515	3%	37.0	26.95	30	140	3%	51.2	26.28
35	0	4%	65.4	24.41	30	320	5%	52.8	26.69
35	10	0%	62.4	24.48	30	515	7%	54.6	26.94
35	30	3%	63.6	24.50	35	0	22%	89.9	24.58
35	70	0%	49.9	25.25	35	10	4%	103.7	24.67
35	140	1%	47.3	25.94	35	23	4%	65.1	24.84
35	320	2%	41.1	26.59	35	50	3%	59.7	25.50
37	0	3%	64.4	24.42	35	140	5%	48.3	26.25
37	10	5%	63.2	24.44	37	0	3%	62.5	24.52
37	30	1%	65.2	24.50	37	10	5%	91.4	24.52
37	40	1%	65.0	24.50	37	40	4%	65.4	25.03
37	50	2%	59.4	24.67	37	45	6%	71.4	
37	70	2%	54.7	25.23	37	50	3%	60.3	25.41
37	140	3%	48.9	25.96	37	60	13%	49.0	25.61
37	320	3%	42.9	26.61	37	140	5%	47.2	26.28
37	515	4%	37.8	26.94	37	320	7%	44.5	26.72
45	0	3%	66.6	24.52	37	515	6%	41.9	26.99
45	10	2%	62.5	24.54	45	0	2%	75.3	24.87
45	20	1%	62.2	24.60	45	10	4%	79.8	24.87
45	50	1%	59.3	24.78	45	20	5%	77.4	24.89
45	140	2%	46.8	26.11	45	40	3%	61.9	25.20
45	320	5%	43.0	26.70	45	160	8%	41.5	26.25
45	515	4%	40.8	26.92	45	320	4%	39.8	26.73
53	0	3%	66.3	24.46	45	515	7%	32.2	27.01
53	10	3%	65.1	24.53	53	0	4%	69.8	24.60
53	20	2%	64.3	24.55	53	10	5%	69.4	24.68
53	30	2%	62.3	24.62	53	20	4%	68.8	24.72
53	40	2%	61.9	24.63	53	30	4%	65.7	24.75
53	50	1%	62.7	24.65	53	40	4%	60.5	24.77
53	60	1%	63.7	24.68	53	50	4%	65.2	24.79
53	70	3%	53.5	25.08	53	55	3%	58.3	24.81

2.6.7 Line 90 (continued)

0501 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0504 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
53	85	1%	47.8	25.38	53	58	6%	57.3	24.85
53	100	2%	47.5	25.59	53	68	5%	57.8	25.00
53	120	1%	47.4	25.92	53	85	4%	52.9	25.25
53	140	3%	46.8	26.08	53	100	7%	45.3	25.59
53	170	1%	44.8	26.30	53	120	7%	48.7	25.94
53	200	3%	42.6	26.43	53	140	4%	44.1	26.14
53	230	2%	40.2	26.51	53	150	7%	43.0	26.19
53	270	1%	43.4	26.59	53	170	4%	37.0	26.25
53	320	2%	43.1	26.68	53	200	2%	44.4	26.41
53	380	2%	39.9	26.79	53	230	2%	48.1	26.48
53	440	1%	40.7	26.88	53	270	4%	47.1	26.60
53	515	5%	38.2	26.96	53	380	4%	38.6	26.79
60	0	0%	64.7	24.48	53	440	5%	37.0	26.87
60	15	3%	64.0	24.49	53	515	3%	30.4	26.98
60	42	2%	61.2	24.53	60	0	14%	67.5	24.49
60	52	1%	54.2	24.76	60	16	5%	67.8	24.50
60	320	1%	44.3	26.68	60	45	5%	66.5	24.50
60	515	2%	39.8	26.96	60	82	4%	71.6	24.96
70	0	4%	72.1	24.48	60	140	4%	48.5	25.83
70	10	5%	74.6	24.49	60	320	4%	41.5	26.67
70	47	4%	73.4	24.74	60	515	7%	41.0	26.96
70	60	3%	63.2	24.78	70	0	2%	59.1	24.45
70	70	5%	68.7	24.96	70	15	3%	62.8	24.45
70	85	2%	61.7	25.24	70	45	4%	72.0	24.45
70	140	3%	53.6	26.04	70	65	17%	53.1	24.61
70	320	5%	45.7	26.68	70	75	4%	54.4	24.86
70	515	4%	45.2	26.97	70	85	30%	45.9	25.00
80	0	1%	65.6	24.49	70	95	4%	52.2	25.17
80	15	2%	62.7	24.49	70	145	5%	43.7	25.85
80	45	3%	62.1	24.51	70	320	6%	43.7	26.63
80	55	4%	58.9	24.64	70	515	11%	42.4	26.96
80	145	2%	45.7	26.10	80	0	8%	52.0	24.47
80	320	2%	39.7	26.70	80	15	1%	59.6	24.47
80	515	3%	38.2	27.00	80	45	2%	56.8	24.47
90	0	0%	61.4	24.53	80	80	2%	60.8	24.48
90	10	1%	61.5	24.53	80	150	3%	47.0	25.86
90	20	1%	60.0	24.53	80	320	28%	83.0	26.56
90	30	4%	60.8	24.54	80	515	10%	51.1	26.92
90	50	3%	58.4	24.57	90	0	6%	62.8	24.45
90	70	2%	49.3	25.38	90	15	4%	60.7	24.45
90	140	1%	43.3	26.15	90	60	5%	57.0	24.47
90	320	1%	40.5	26.73	90	75	8%	68.1	24.49
90	515	2%	35.2	27.00	90	85	3%	64.5	24.66
100	0	2%	60.8	24.40	90	95	2%	51.5	24.91

2.6.7 Line 90 (continued)

0501 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0504 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
100	13	5%	63.0	24.40	90	100	4%	53.0	25.00
100	27	2%	59.8	24.41	90	105	3%	50.3	25.12
100	42	1%	60.8	24.49	90	150	1%	38.3	25.80
100	53	3%	57.1	24.69	90	320	5%	34.7	26.63
100	64	1%	52.3	24.78	90	515	2%	44.3	26.95
100	76	4%	51.6	25.08	100	0	2%	61.4	24.34
100	85	2%	47.9	25.30	100	18	2%	58.5	24.34
100	95	2%	48.9	25.41	100	30	2%	60.7	24.34
100	110	2%	45.8	25.70	100	41	3%	60.3	24.34
100	125	1%	44.6	25.91	100	52	3%	57.8	24.34
100	145	5%	46.8	26.07	100	63	0%	60.9	24.49
100	170	2%	46.0	26.19	100	72	1%	59.5	24.66
100	200	1%	42.1	26.36	100	83	4%	59.1	24.77
100	230	2%	42.7	26.49	100	94	3%	52.2	24.97
100	270	2%	41.4	26.59	100	106	5%	51.4	25.13
100	320	2%	41.9	26.69	100	117	3%	47.7	25.24
100	380	4%	40.4	26.81	100	128	5%	49.7	25.41
100	440	2%	39.1	26.89	100	140	3%	50.4	25.57
100	515	1%	36.4	27.01	100	165	3%	46.8	25.89
110	0	3%	61.3	24.31	100	195	3%	42.5	26.16
110	15	0%	61.0	24.31	100	230	4%	42.3	26.34
110	45	4%	64.0	24.33	100	380	3%	39.3	26.76
110	75	0%	60.2	24.45	100	440	1%	39.7	26.85
110	145	3%	43.6	25.76	100	515	2%	66.4	26.97
110	320	4%	39.7	26.63	110	0	4%	66.0	24.42
110	515	2%	38.5	26.95	110	15	4%	60.8	24.42
120	0	4%	71.7	24.33	110	45	1%	60.6	24.42
120	15	4%	65.7	24.33	110	75	4%	59.5	24.64
120	45	1%	66.0	24.35	110	145	6%	48.2	25.77
120	75	2%	66.3	24.37	110	270	1%	40.6	26.53
120	85	2%	65.0	24.43	110	320	4%	38.6	26.65
120	95	7%	69.0	24.74	110	320	2%	42.5	
120	115	3%	61.0	25.01	110	515	3%	36.7	26.98
120	140	3%	52.6	25.33	120	0	1%	59.8	24.33
120	165	4%	48.0	25.63	120	15	6%	61.2	24.33
120	320	5%	43.3	26.59	120	60	1%	57.5	24.34
120	515	6%	40.5	26.95	120	100	4%	56.3	24.75
					120	105	1%	54.9	24.82
					120	110	4%	52.3	24.90
					120	125	5%	45.7	25.15
					120	140	3%	51.8	25.35
					120	165	5%	55.5	25.66
					120	320	1%	42.9	26.60
					120	515	4%	37.1	26.98

2.6.7 Line 90 (continued)

0507 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0511 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
27	0	4%	104.7	23.94	28	2	3%	79.8	24.12
27	5	7%	114.2	24.15	28	5	4%	82.6	24.12
27	10	13%	99.9	25.25	28	20	4%	69.9	24.61
27	15	3%	95.7	25.33	28	50	3%	55.3	25.34
27	20	5%	72.7	25.57	30	10	3%	77.8	24.11
30	0			24.09	30	19	4%	79.0	24.53
30	10			24.90	30	40	4%	64.7	25.14
30	20			25.44	30	138	4%	54.2	26.19
30	40			25.86	30	317	3%	44.1	26.68
30	140			26.30	30	511	6%	47.9	26.97
30	320			26.66	35	2	4%	72.3	24.30
30	515			26.89	35	13	3%	66.5	24.36
35	0	6%	116.4	23.83	35	21	4%	63.9	24.42
35	10	5%	108.9	24.17	35	64	7%	51.8	25.64
35	20	2%	81.7	25.38	35	140	1%	44.5	26.28
35	50	6%	60.3	25.94	35	282	2%	43.0	26.62
35	140	3%	59.7	26.36	37	2	3%	70.7	24.46
35	320	11%	57.0	26.69	37	10	3%	70.4	24.48
35	380	6%	48.6	26.79	37	30	4%	59.7	24.75
37	0	5%	105.8	23.86	37	40	2%	55.9	25.01
37	10	5%	88.7	24.23	37	50	3%	53.5	25.16
37	22	5%	74.6	25.23	37	60	4%	52.1	25.40
37	30	4%	58.9	25.62	37	140	4%	46.1	26.27
37	35	4%	58.2		37	318	3%	46.0	26.68
37	40	8%	67.1	25.81	37	512	2%	43.2	26.95
37	140	3%	42.6	26.34	45	2	4%	71.0	24.30
37	320	2%	39.4	26.69	45	10	4%	66.4	24.30
37	515	4%	38.4	26.96	45	30	2%	57.7	25.00
53	0	4%	114.3	24.86	45	60	3%	49.8	25.64
53	12	7%	106.5	24.87	45	139	3%	43.4	26.24
53	23	8%	101.4	24.89	45	318	2%	39.7	26.70
53	34	7%	78.9	25.28	45	512	4%	37.4	26.97
53	44	4%	83.2	25.47	53	1	4%	75.1	24.27
53	52	4%	95.5	25.65	53	2	2%	77.7	24.27
53	60	7%	99.1	25.81	53	10	2%	70.7	24.27
53	70	6%	122.7	25.89	53	30	2%	66.8	24.36
53	85	6%	72.6	26.01	53	31	5%	63.5	24.37
53	100	4%	72.9	26.16	53	39	6%	62.3	24.52
53	120	19%	76.0	26.27	53	50	4%	60.0	25.11
53	140	17%	81.0	26.32	53	60	4%	61.9	25.31
53	170	6%	77.5	26.44	53	75	7%	54.9	25.51
53	200	5%	55.7	26.52	53	85	2%	56.5	25.60
53	230	5%	46.9	26.60	53	100	3%	49.0	25.84
53	270	7%	68.6	26.70	53	125	4%	48.7	26.14
53	320	6%	82.2	26.76	53	150	4%	58.4	26.24
53	380	4%	72.9	26.83	53	170	5%	51.8	26.32

2.6.7 Line 90 (continued)

0507 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0511 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
53	440	5%	54.5	26.93	53	200	3%	49.1	26.42
53	515	4%	39.9	27.01	53	250	5%	46.5	26.57
60	0	6%	80.8	24.17	53	300	5%	47.7	26.67
60	10	6%	81.8	24.17	53	319	5%	46.7	26.70
60	45	2%	78.1	24.49	53	400	3%	46.9	26.84
60	60	2%	75.0	24.84	53	500	5%	43.7	26.95
60	140	9%	70.5	26.10	60	3	0%	65.3	24.29
60	320	4%	76.2	26.70	60	10	4%	66.3	24.29
60	515	4%	49.3	27.01	60	40	1%	55.3	25.09
70	0	9%	83.0	23.99	60	69	1%	48.0	25.52
70	10	6%	94.1	23.99	60	151	1%	43.2	26.32
70	30	14%	89.1	24.11	60	319	4%	44.2	26.76
70	40	10%	88.3	24.50	60	513	3%	38.2	26.97
70	50	5%	88.9	24.61	70	2	2%	65.3	24.48
70	70	3%	76.0	24.88	70	12	4%	67.4	24.49
70	140	4%	61.0	26.12	70	20	3%	68.8	24.50
70	320	3%	54.2	26.71	70	49	3%	56.3	25.09
70	515	7%	57.6	27.00	70	75	1%	49.5	25.80
80	0	6%	66.2	24.29	70	140	2%	46.7	26.27
80	10	10%	57.5	24.29	70	321	2%	42.8	26.77
80	40	5%	56.2	24.77	70	514	3%	43.0	27.03
80	55	5%	56.3	24.92	80	2	2%	68.7	24.14
80	140	4%	46.6	26.16	80	15	2%	69.3	24.14
80	320	5%	43.0	26.73	80	45	4%	65.1	24.21
80	515	6%	40.5	27.02	80	84	3%	59.6	25.17
90	0	5%	70.2	23.93	80	145	3%	48.4	26.16
90	15	3%	78.0	23.92	80	318	4%	44.1	26.75
90	57	3%	72.1	24.22	80	510	2%	41.5	26.99
90	68	2%	64.1	24.39	90	2	1%	67.2	24.13
90	80	4%	62.4	24.56	90	10	2%	75.5	24.13
90	88	2%	61.7	24.72	90	35	1%	69.3	24.38
90	95	13%	61.5	24.86	90	40	4%	66.6	24.85
90	145	3%	47.9	25.71	90	51	3%	58.3	25.15
90	320	5%	39.8	26.62	90	60	3%	54.9	25.29
90	515	4%	34.4	26.96	90	86	3%	52.5	25.71
100	0	6%	77.8	23.85	90	168	3%	43.6	26.37
100	15	6%	77.2	23.85	90	318	3%	41.8	26.71
100	30	4%	65.0	23.84	90	514	4%	37.6	27.00
100	45	4%	63.0	24.06	100	2	3%	62.0	23.90
100	55	3%	56.5	24.14	100	8	1%	62.9	23.90
100	65	7%	69.8	24.21	100	15	1%	63.7	23.92
100	75	5%	61.0	24.40	100	23	2%	60.0	23.96
100	80	8%	69.2		100	31	3%	65.5	24.05
100	85	10%	62.8	24.58	100	38	1%	62.1	24.13
100	95	2%	56.9	24.80	100	46	2%	60.8	24.35
100	110	2%	45.2	25.05	100	52	5%	60.0	24.58

2.6.7 Line 90 (continued)

0507 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0511 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
100	125	4%	53.1	25.28	100	59	4%	57.2	24.70
100	154	5%	53.6	25.65	100	71	2%	55.2	24.85
100	170	7%	51.7	25.90	100	82	3%	56.7	25.04
100	200	6%	44.6	26.17	100	91	2%	55.6	25.23
100	230	4%	39.3	26.36	100	98	4%	51.3	25.43
100	270	3%	49.5	26.48	100	120	2%	45.7	25.87
100	320	2%	42.6	26.62	100	150	4%	42.9	26.16
100	380	4%	44.5	26.73	100	168	1%	43.4	26.24
100	440	6%	41.1	26.82	100	199	4%	41.4	26.37
100	515	4%	33.4	26.96	100	228	4%	42.7	26.46
110	0	4%	75.3	23.68	100	269	6%	38.2	26.59
110	15	7%	64.5	23.70	100	319	5%	39.6	26.70
110	60	6%	64.4	24.37	100	378	1%	38.1	26.80
110	100	2%	59.9	24.87	100	438	3%	35.1	26.91
110	515	3%	35.7	26.98	100	512	2%	36.5	27.03
120	0	5%	61.1	23.74	110	2	5%	63.6	23.89
120	10	3%	61.0		110	15	5%	61.5	23.89
120	15	3%	63.1	23.75	110	60	2%	59.1	24.36
120	45	5%	64.6	23.93	110	90	2%	55.0	24.78
120	75	3%	62.9	24.37	110	104	4%	56.0	24.97
120	110	4%	53.6	24.67	110	140	4%	47.0	25.49
120	125	1%	47.4	24.85	110	318	1%	40.5	26.61
120	130	4%	58.0	25.06	110	522	1%	38.5	26.98
120	140	6%	50.9	25.27	120	45	4%	59.0	23.81
120	320	6%	43.8	26.61	120	60	2%	58.5	23.81
120	515	5%	35.4	26.96	120	75	5%	57.2	24.30
					120	83	5%	57.4	24.43
					120	139	4%	51.2	25.26
					120	323	7%	45.3	26.60
					120	512	3%	38.6	26.92

2.6.8 Line 80 (by cruise)

0501 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0504 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
50	0	2%	65.6	24.51	50	0	3%	70.7	25.51
50	5	12%	73.4	24.52	50	5	3%	60.7	25.63
50	10	6%	66.2	24.54	50	10	3%	56.4	25.67
50	20	5%	61.0	24.58	50	15	4%	53.6	25.78
51	0	1%	60.7	24.58	50	22	3%	48.7	25.84
51	10	1%	60.8	24.58	51	0	2%	81.4	25.59
51	20	3%	62.2	24.59	51	10	4%	73.7	25.62
51	80	4%	49.0	25.41	51	20	8%	61.9	25.78
55	0	3%	54.4	24.58	51	65	7%	49.1	26.23
55	10	2%	57.5	24.58	55	0	3%	72.2	25.07
55	20	0%	54.4	24.74	55	5	3%	73.1	25.08
55	30	2%	51.0	24.77	55	10	2%	91.5	25.18
55	40	1%	47.7	24.95	55	15	4%	60.5	25.24
55	50	4%	48.6	25.05	55	20	2%	55.5	25.43
55	60	3%	44.9	25.28	55	25	5%	56.9	25.51
55	70	5%	42.9	25.33	55	30	4%	56.1	25.58
55	85	4%	43.0	25.46	55	40	4%	54.4	25.64
55	100	4%	39.4	25.81	55	50	3%	53.5	25.75
55	120	2%	38.3	25.96	55	60	1%	54.4	25.85
55	140	5%	41.7	26.05	55	70	2%	51.5	25.95
55	170	3%	38.7	26.16	55	85	1%	46.8	26.09
55	200	3%	37.6	26.29	55	100	1%	44.3	26.18
55	230	2%	36.7	26.39	55	120	1%	45.7	26.35
55	270	5%	37.1	26.52	55	140	4%	45.0	26.38
55	320	2%	33.5	26.60	55	170	5%	46.2	26.51
55	380	2%	33.9	26.71	55	200	2%	44.0	26.59
55	440	11%	30.3	26.83	55	230	4%	41.5	26.64
55	515	4%	30.1	26.89	55	270	4%	44.0	26.69
60	0	4%	59.2	24.70	55	320	5%	43.3	26.73
60	10	5%	56.9	24.70	55	380	5%	44.1	26.79
60	30	2%	63.5	24.70	55	440	5%	43.1	26.86
60	85	2%	45.0	25.60	55	515	3%	40.0	26.96
60	140	3%	42.9	26.23	60	0	5%	84.3	24.78
60	320	4%	38.2	26.74	60	7	2%	79.4	24.81
60	515	3%	34.2	26.99	60	18	2%	66.9	25.11
70	0	2%	60.3	24.93	60	43	2%	62.3	25.37
70	12	2%	66.0	25.03	60	140	5%	57.5	26.32
70	25	2%	64.0	25.05	60	320	4%	47.3	26.76
70	53	3%	47.4	25.37	60	515	4%	46.3	26.97
70	140	3%	36.6	26.37	70	0	7%	99.0	24.45
70	140	2%	54.6		70	10	10%	87.1	24.55
70	320	5%	35.4	26.74	70	35	7%	89.9	24.70
70	515	5%	38.6	27.02	70	45	2%	79.7	24.74
80	0	2%	62.7	24.80	70	50	4%	82.5	24.84
80	20	4%	66.7	24.80	70	55	4%	87.0	24.93
80	30	3%	63.4	24.81	70	140	9%	78.0	26.12

2.6.8 Line 80 (continued)

0501 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0504 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
80	40	5%	59.3	24.82	70	320	2%	48.1	26.67
80	50	3%	53.3	24.97	70	515	7%	48.1	26.94
80	60	3%	55.0	25.15	80	0	2%	65.0	24.36
80	140	2%	47.5	26.27	80	15	3%	66.6	24.40
80	320	3%	46.3	26.65	80	30	3%	67.4	24.55
80	515	2%	36.1	26.95	80	50	4%	65.3	24.59
90	0	3%	57.8	24.58	80	60	2%	61.1	24.62
90	10	1%	59.2	24.58	80	65	3%	60.6	24.73
90	20	2%	59.0	24.59	80	75	5%	55.8	25.19
90	40	3%	57.3	24.59	80	140	5%	45.6	26.12
90	140	6%	42.4	26.21	80	320	3%	49.5	26.65
90	320	1%	37.6	26.75	80	515	1%	39.1	26.95
90	515	2%	33.4	27.01	90	0	2%	68.5	24.40
100	0	1%	59.4	24.48	90	15	4%	66.2	24.43
100	10	4%	57.6	24.48	90	42	2%	69.2	24.60
100	20	1%	57.2	24.48	90	70	2%	56.9	24.90
100	30	2%	58.5	24.49	90	140	3%	50.0	26.06
100	40	2%	55.3	24.49	90	320	3%	41.8	26.70
100	50	2%	65.7	24.53	90	515	13%	41.1	27.00
100	60	3%	52.0	24.80	100	0	2%	64.7	24.34
100	70	4%	50.1	25.10	100	10	4%	64.0	24.34
100	85	1%	46.3	25.16	100	20	5%	64.0	24.53
100	100	2%	43.0	25.49	100	30	4%	63.6	24.61
100	120	2%	43.3	25.71	100	40	4%	62.7	24.67
100	140	1%	43.1	25.97	100	50	1%	61.9	24.71
100	170	4%	42.5	26.22	100	55	6%	65.0	24.73
100	200	4%	39.6	26.38	100	60	5%	62.9	24.75
100	230	3%	37.8	26.47	100	70	2%	61.2	24.80
100	270	2%	41.3	26.59	100	80	2%	50.6	25.29
100	320	3%	43.2	26.71	100	100	2%	46.6	25.54
100	380	3%	38.2	26.80	100	120	2%	47.5	25.87
100	440	2%	34.3	26.91	100	140	4%	45.7	26.02
100	515	5%	32.9	27.02	100	170	3%	45.6	26.25
					100	200	3%	41.8	26.37
					100	230	3%	41.8	26.46
					100	270	2%	41.8	26.58
					100	320	4%	42.0	26.67
					100	380	3%	41.9	26.80
					100	440	2%	42.9	26.90
					100	515	3%	39.5	27.01

2.6.8 Line 80 (continued)

0507 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0511 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
50	0	5%	95.3	24.94	51	2	4%	87.4	24.41
50	5	5%	97.3	24.95	51	10	3%	87.2	24.42
50	10	2%	93.8	24.99	51	25	3%	95.3	24.53
50	20	5%	117.8	25.10	51	40	3%	63.4	25.10
51	0	2%	105.6	24.76	51	70	3%	56.9	25.47
51	10	6%	67.4	25.56	55	2	3%	77.1	24.93
51	20	4%	53.9	25.78	55	5	0%	83.8	24.93
51	35	3%	42.1	25.96	55	11	2%	74.3	24.93
55	2	2%	74.5	25.31	55	16	2%	80.9	24.93
55	10	1%	76.7	25.31	55	20	7%	81.3	24.95
55	20	7%	71.0	25.39	55	30	1%	61.3	24.97
55	31	2%	59.9	25.48	55	40	4%	59.1	25.03
55	40	3%	63.5	25.60	55	50	1%	61.3	25.24
55	50	2%	63.4	25.83	55	60	7%	53.2	25.48
55	61	5%	57.1	25.98	55	71	3%	49.6	25.63
55	68	4%	52.6	26.02	55	85	4%	60.6	25.78
55	87	2%	52.8	26.21	55	99	3%	50.0	25.99
55	101	4%	50.2	26.31	55	120	3%	47.2	26.18
55	120	9%	53.9	26.38	55	140	4%	56.9	26.30
55	141	5%	48.3	26.42	55	169	8%	48.6	26.41
55	170	4%	48.4	26.47	55	200	2%	42.4	26.47
55	201	4%	46.0	26.54	55	230	3%	48.7	26.56
55	229	4%	46.7	26.57	55	268	4%	47.1	26.62
55	268	3%	44.8	26.63	55	318	4%	40.6	26.69
55	321	4%	51.0	26.69	55	379	4%	43.1	26.80
55	383	3%	49.1	26.76	55	436	3%	42.6	26.89
55	439	3%	50.2	26.81	55	512	2%	39.6	26.98
55	517	4%	48.8	26.94	60	2	6%	97.8	24.89
60	0	4%	63.5	24.93	60	9	3%	73.2	25.06
60	10	4%	65.8	24.94	60	32	3%	66.2	25.11
60	30	6%	67.0	25.26	60	142	2%	42.6	26.28
60	50	3%	49.9	25.73	60	319	3%	39.6	26.72
60	140	7%	48.1	26.39	60	512	2%	40.3	26.95
60	320	4%	41.1	26.80	70	3	5%	71.0	24.60
60	515	4%	33.1	27.05	70	10	1%	70.1	24.60
70	0	4%	73.8	24.62	70	20	5%	69.9	24.63
70	10	2%	65.3	24.62	70	40	2%	68.0	24.65
70	30	2%	63.2	25.09	70	50	3%	60.9	25.18
70	40	5%	61.6	25.13	70	69	7%	56.2	25.75
70	52	2%	62.7	25.17	70	139	5%	54.7	26.25
70	61	8%	55.8	25.53	70	318	3%	53.8	26.59
70	140	5%	51.9	26.31	70	512	32%	60.1	26.85
70	320	9%	45.6	26.76	80	2	3%	71.9	24.24
70	515	3%	37.4	27.02	80	15	6%	67.4	24.24
80	0	2%	74.1	23.99	80	45	4%	67.8	24.25
80	15	4%	72.7	23.99	80	55	5%	69.5	24.25

2.6.8 Line 80 (continued)

0507 Station	Depth (m)	CV	TOC (μM C)	Density (σ_θ)	0511 Station	Depth	CV	TOC (μM C)	Density (σ_θ)
80	50	5%	66.1	24.06	80	60	3%	68.5	24.26
80	70	1%	56.4	24.39	80	75	13%	60.7	25.02
80	75	4%	65.1	24.58	80	85	4%	66.1	25.24
80	80	4%	59.4	24.76	80	144	5%	57.7	26.08
80	145	7%	46.2	25.85	80	316	0%	57.2	26.70
80	320	7%	45.7	26.67	80	514	12%	53.5	27.00
80	515	3%	39.8	26.98	90	2	4%	72.6	24.15
90	0	18%	75.3	23.77	90	11	3%	80.8	24.15
90	15	3%	73.5	23.76	90	40	7%	65.8	24.30
90	60	5%	72.5	24.10	90	60	2%	62.6	24.64
90	95	1%	56.9	24.97	90	139	6%	56.8	26.02
90	140	0%	56.4	25.66	90	318	7%	53.1	26.70
90	320	5%	54.9	26.64	90	514	3%	42.9	27.00
90	515	3%	44.0	26.97	100	2	4%	63.6	23.92
100	3	7%	69.3	23.72	100	16	4%	64.5	23.97
100	16	4%	70.9	23.72	100	26	2%	65.3	24.01
100	30	5%	69.5	23.96	100	35	4%	63.5	24.02
100	45	2%	71.3	24.16	100	42	2%	62.3	24.03
100	60	4%	63.2	24.52	100	49	2%	61.7	24.41
100	76	2%	62.3	24.72	100	56	2%	59.2	24.61
100	85	3%	61.4	24.90	100	63	4%	54.9	24.75
100	94	3%	59.9	25.07	100	73	4%	57.8	24.91
100	106	6%	62.8	25.25	100	84	4%	53.9	25.07
100	116	5%	57.1	25.43	100	92	2%	50.6	25.25
100	125	3%	47.4	25.53	100	100	4%	50.9	25.37
100	141	3%	47.2	25.72	100	110	1%	46.6	25.46
100	167	4%	49.5	26.04	100	124	1%	46.9	25.74
100	197	5%	45.1	26.25	100	144	6%	46.7	25.98
100	230	2%	41.0	26.41	100	169	1%	40.7	26.24
100	270	4%	39.6	26.53	100	198	3%	39.0	26.42
100	321	3%	42.3	26.64	100	244	4%	40.7	26.54
100	383	6%	42.0	26.76	100	268	6%	39.7	26.61
100	440	3%	38.2	26.85	100	317	3%	37.5	26.69
100	514	7%	36.5	26.97	100	377	6%	36.5	26.80
					100	438	4%	35.6	26.88
					100	512	6%	41.3	26.98

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3 CHEMICAL AND ISOTOPIC CHARACTERISTICS OF ORGANIC MATTER FRACTIONS IN THE EASTERN NORTH PACIFIC OCEAN

3.1 INTRODUCTION

The biological and chemical reactivity of dissolved organic compounds in seawater has been a focus of study for several decades (Williams and Gordon, 1970; Stuermer and Harvey, 1977; Bada and Lee 1977; Willams and Druffel, 1987; Benner et al., 1992; Amon and Benner, 1994; Carlson and Duckow, 1995; Aluwihare et al., 1997; Aluwihare et al., 2005; Repeta and Aluwihare, 2006). This dissolved organic carbon (DOC) reservoir contains approximately 650 Gt C (Hansell, 2002) that is available to exchange on relatively short timescales with other reservoirs of the global carbon cycle (e.g., dissolved inorganic carbon (DIC) and atmospheric CO_2). However, radiocarbon measurements ($\Delta^{14}C$) have shown that the average ^{14}C-age of the DOC reservoir is ~6000 years with respect to atmospheric CO_2 (Willams and Druffel, 1987). Together with its total inventory, this average residence time yields a relatively small steady state flux of material through the DOC reservoir each year (0.1 Gt C/yr; Willams and Druffel, 1987, Hedges and Oades, 1997). In contrast, both the higher concentration and greater ^{14}C content of DOC in surface waters (<500 m) indicates the production and removal of as much as 50% of the surface inventory on annual to decadal timescales (Willams and Druffel, 1987).

Compositional studies have shown that marine dissolved organic matter (DOM) is comprised of several chemical moieties (reviewed in Benner, 2002) and may be

derived from a variety of sources (Hedges and Oades, 1997). Therefore, individual components could have very different rates of biological and chemical reactivity. For example, compound-specific measurements have shown that the $\Delta^{14}C$ value of carbohydrates dissolved in the surface ocean is similar to $\Delta^{14}C$-DIC in these same waters (Repeta and Aluwihare, 2006). This finding is consistent with recent production of carbohydrates during photosynthesis and subsequent removal on short timescales. Alternatively, studies have indicated the presence of dissolved components with ^{14}C-ages that exceed 6000 years (Wang et al., 2001, Loh et al., 2004). Together, these available data confirm the presence of both modern (<50 years) and old (>6000 radiocarbon years) organic compounds in the DOC reservoir.

It has been estimated that up to 50% of the organic carbon fixed in surface waters, or ~25 Gt C, is channeled through the DOC pool each year (Fuhrman and Azam, 1982). The old ^{14}C-age of marine DOC could result from the aging of a component of this autochthonous DOC, in which case a continuum of radiocarbon ages between modern and >6000 radiocarbon years should be present for DOC in the ocean. Alternatively, DOC may be either pre-aged in an external reservoir prior to entering the marine water column or recently biosynthesized using a radiocarbon depleted carbon source (e.g., ^{14}C-depleted methane). These latter pathways would not give rise to a broad continuum of radiocarbon ages.

In this study we investigated two hypotheses: (1) chemically-distinct compounds express the ^{14}C diversity of marine DOC and (2) marine $\Delta^{14}C$-DOC results from a mixture of a few discrete ^{14}C-age groups rather than a broad continuum of ^{14}C-ages. To test these hypotheses we designed a solid phase extraction (SPE) method to separate a

subset of marine DOC into chemically-distinct fractions. This is the first study to directly examine the relationship between chemical composition and ^{14}C-content of dissolved organic compounds in the marine environment.

3.2 EXPERIMENTAL METHODS

3.2.1 Sample Locations and Collection

All surface SPE samples were collected in April 2004 (cruise 0404) and October 2004 (cruise 0411) during routine, quarterly California Cooperative Oceanic Fisheries Investigation (CalCOFI) survey cruises (see Chapter 2; Figure 3.1). Surface samples were collected using a diaphram pump and filtered through a Pall SuporLife 200, polyethersulfone cartridge filter (Cat. No. SCS92DP71S; pore size 0.2 μm; 0.5 m^2 surface area) fitted with Bev-A-Line (Cole Parmer) tubing and stored in acid cleaned 200 L HDPE barrels.

Two sub-surface samples were also isolated for SPE. Briefly, during cruise 0411 seawater from 1000 m was collected in twenty-four 10 L Niskin bottles mounted on a rosette and filtered as above by pumping directly out of each bottle. In addition, ~600 L of seawater were collected from 670 m depth at the Natural Energy Laboratory of Hawaii Authority (NELHA; Kailua-Kona, Hawaii; 19.70ºN, 156.05ºW) in November 2003.

Samples for ultrafiltration (UF) were collected in October 2003 (cruise 0310). 200 L of pre-filtered seawater were collected from 3 sites within the CalCOFI region by direct pumping from surface waters; one 200 L sample was also collected from 1000 m

using Niskin bottles.

During cruise 0404 CalCOFI stations 77.60, 82.47, 80.80, 83.60, 87.55, 90.53, and 93.50 were sampled; and on cruise 0411 stations 77.60, 82.47, 87.80, 90.100, 90.30, 90.70, 93.80 and 93.120 were sampled for POM.

3.2.2 Extraction and Fractionation of Organic Matter

In the case of SPE samples, a peristaltic pump was used to pump seawater through acid-cleaned silicone tubing at a rate of ~5 L hr^{-1} into a combusted glass column (450mm by 15mm, Ace Glass) containing ~50 mL of cleaned HP-20 resin (Mitsubishi) retained with a Teflon mesh screen. Prior to sample processing the resin was washed extensively with water, methanol, acetone, and ethyl acetate (500 mL each per 100 mL of resin), and the column was packed in water. Once seawater had been passed through the column the resin was eluted sequentially with 2 column volumes (120 mL) of water (Fraction A), 1:1 (v/v) methanol:water (Fraction B), 100% methanol (Fraction C), and finally, 100% acetone (Fraction D). The latter two organic fractions were further fractionated in a separatory funnel by partitioning between equal volumes of water (Fraction C and D; see Figure 3.2) and ethyl acetate (Fractions CL and DL). All fractions were colored so they were stored in the dark at room temperature until they were dried by rotary evaporation and/or lyophilization. Altogether, this method afforded 6 fractions per sample (Figure 3.2).

Seawater at NELHA was pumped directly through an HDPE pipe into shore-based taps at a rate of ~800 L s^{-1}. In this study, seawater exiting the taps (~ 5 L min^{-1}) was filtered as above and stored in 200 L HDPE barrels during SPE. In each case, SPE

and chemical fractionation was performed exactly as above for surface samples.

Finally, for comparison, ultrafiltered dissolved organic matter (UDOM) was also isolated by UF following pre-filtration through 0.2 μm, according to Meador et al. (2007). The UF system was fitted with a 1,000 Dalton molecular weight cut-off (MWCO) polysulfone, spiral-wound filter membrane. In the laboratory frozen, concentrated UDOM samples were lyophilized under vacuum (using an oil-free vacuum pump) and the dry powder was subjected to a suite of chemical analyses.

For POM filtration, seawater was pumped directly onboard with a diaphragm pump into 20 L, acid-rinsed, stainless steel, ball lock style vessels. Vessels were sealed and pressurized using ship's air filtered through an inline charcoal filter. Seawater under pressure was passed through an in-line pre-combusted (450ºC) GF/F filter (Whatman) housed in an acid-rinsed polysulfone filter holder. Several blank GF/F filters were also processed to establish the carbon content and isotopic composition of the filter material.

All hardware, plastic tubing and fittings were acid-rinsed before each cruise and all glassware was pre-combusted overnight at 450ºC; additionally, all components used in UF were acid rinsed between samples.

3.2.3 TOC Analysis

Vertical profiles of total organic carbon (TOC) concentration throughout the CalCOFI grid were collected in combusted 40 mL glass vials fitted with acid-rinsed Teflon-lined caps, and immediately acidified with trace metal grade HCl (20 μL) to pH 2 then stored at 4^{o}C until analysis (3 months). Total organic carbon measurements were performed on a Shimadzu TOC-V instrument fitted with an autosampler. The

concentration of each sample was calculated from an average of four 100 μL injections using a five point calibration curve. Prior to and during each sample run, the instrument was calibrated using a five-point potassium hydrogen phthalate standard curve and certified reference materials (courtesy of Dennis Hansell, RSMAS) in the form of low carbon (2μM C; LCW) and deep Sargasso Sea water (>2000 m, 44-47 μM C) . Milli-Q water (used to make the standards) was injected after every 4-5 samples to insure that the baseline remained at its original response.

3.2.4 Elemental and Stable Isotope (^{13}C and ^{15}N) Analysis

The elemental and stable isotope composition of solid, dry DOM fractions was determined at the Scripps Institution of Oceanography Unified Laboratory Facility using standard elemental analyzer isotope ratio mass spectrometry (EA-IRMS) techniques (Owens and Rees, 1989; Meador et al., 2007). In the case of suspended POM, subsamples were randomly selected from acidified, dried 47 mm GF/F filters and analyzed as above. Elemental (carbon and nitrogen) results are reported as %C and the molar ratio of carbon to nitrogen (C:N); stable isotopic composition is reported in the standard δ notation (‰) relative to Pee Dee Belemnite (marine carbonate; PDB) or atmospheric nitrogen (N_2); for example, $\delta^{13}C = [((^{13}C/^{12}C)_{sample}/(^{13}C/^{12}C)_{PDB}) - 1] \times 1000$

3.2.5 Radiocarbon (^{14}C) Analysis

Organic matter fractions were prepared for ^{14}C measurements according to the closed tube combustion method of Vogel et al. (1997). Samples (DOM or subsamples of

pre-acidified GF/F filters for POM) were freeze dried or dried under N_2 in pre-combusted (850°C) quartz tubes. Pre-combusted CuO and Ag were added to tubes containing each sample and tubes were then evacuated and sealed under vacuum; sealed samples were combusted at 850°C for 4 hours. The CO_2 resulting from the combustion was purified and quantified in a vacuum gas extraction line and graphitized under an atmosphere of H_2 over an Fe catalyst. Graphitized samples were pressed into targets and analyzed at the Center for Accelerator Mass Spectrometry, Lawrence Livernmore National Laboratory via accelerator mass spectrometry. Data are reported according to Stuiver and Polach (1987) in $\Delta^{14}C$ (‰).

$\Delta^{14}C$ (‰) is reported relative to an oxalic acid standard (Oxalic Acid II, HOxII) and are calculated from "Fraction Modern" (f_m) using the sample collection date. Fraction Modern, is the deviation of the $^{14}C/^{12}C$ ratio of a sample from a "modern" standard (e.g. McNichol and Aluwihare, 2007). All data are corrected for isotopic fractionation by normalizing to a $\delta^{13}C$ value of -25‰ using the measured $\delta^{13}C$ (versus PDB) of the sample. The error associated with the $\Delta^{14}C$ measurement did not exceed ~9‰.

3.2.6 Isotope blank corrections

All isotope data were corrected for contributions from procedural blanks. DOM samples were corrected using the isotope values listed in Table 3.1. For POM blanks, several batches of blank Whatman GF/F filters were vacuum-sealed in quartz tubes and combusted at 850 ^{o}C for 4 hours. The resulting CO_2 was measured manometrically and submitted for isotope (^{13}C and ^{14}C) analysis. The measured values from the process

blanks were used to calculate the true isotope values using a simple mass balance equation:

$$\Delta_{s+b} = (1-x_b)\Delta_s + x_b\Delta_b$$

where Δ_{s+b} is the measured isotopic value; Δ_s, Δ_b are the sample and blank isotope values, respectively; and x_b is the fractional mass contribution of the process blank to the sample.

3.2.7 ^{1}H NMR spectroscopy

Water soluble samples were first freeze dried in D_2O to reduce exchangeable protons. After freeze drying samples were either re-dissolved in D_2O or in the case of organic fractions, dissolved directly into CD_3OD, and transferred into a dry NMR tube using a pre-combusted Pasteur pipette. Spectra were collected on a Varian 300 MHz Inova spectrometer using standard pulse sequences. Spectra were averaged over 128 scans and referenced to 4.79 ppm for D_2O or 3.31 ppm for CD_3OD.

3.2.8 Avoiding ^{14}C tracer level contamination

Tracer level primary productivity ^{14}C experiments were performed on cruises when sampling took place. Avoiding contamination for natural abundance ^{14}C analysis was of paramount importance. Initially, swab tests aboard the ship labs were obtained before and after the cruises to assess any potential cross contamination. Aboard the ship, analysis and storage of all tracer experiments were kept in separate containers, freezers, and refrigerators from natural abundance samples. A separate radiation van was used to conduct radioisotope experiments and measurements. Any personnel working with tracers wore booties inside and only inside the radio lab. In addition, personnel avoided

areas in the lab where natural abundance samples were collected and processed. Post cruise swabs were negative for contamination in the labs after all our cruises.

3.3 RESULTS AND DISCUSSION

The SPE method developed here isolated 12-18 mg C from 200 L of filtered, non-acidified seawater; this represents approximately 10 to 15% of the TOC as calculated from dry weight, volume of seawater extracted and initial TOC concentration at each site. The first fraction, Fraction A, was eluted with Milli-Q water and primarily contained residual sea salts. As its carbon content was very low (<2%) we did not consider this fraction for subsequent analyses. Fraction B which eluted with 1:1 (v/v) methanol:water, represented approximately two-thirds of the total carbon isolated by SPE in each sample. Organic solvent fractions were eluted with either 100% methanol or 100% acetone and then partitioned between water (Fraction C and D) and ethyl acetate (Fraction CL and DL) to separate lipid material from water soluble material (Figure 3.2). As expected, compounds soluble only in ethyl acetate (e.g., neutral lipids; CL and DL) were present in very low abundance. Material isolated from fraction D did contain DOM but also contained contaminants from the experimental setup and is therefore omitted from further discussion.

Each UDOM sample collected by ultrafiltration in fall 2003 (Cruise 0310) represented approximately 20-30% of the TOC based on dry weight, volume of seawater filtered and average TOC concentrations in the region.

3.3.1 Procedural Blanks

Blanks from the resin after cleaning resulted in <35 μg C (Table 3.1) for each fraction which is less than 1% of sample C collected from 200 L seawater. The largest amount of blank-carbon was present in non-polar fractions. The organic washes were collected, combined, and analyzed to determine the chemical composition and isotopic signature of resin contaminants. The combined wash material had a $\delta^{13}C$ value -25.5‰, a $\delta^{15}N$ value of -4.5‰ and a $\Delta^{14}C$ value of -975‰. The highly depleted $\Delta^{14}C$ value is indicative of ^{14}C-dead carbon (-1000‰) consistent with a petroleum or fossil source. Any contaminating N material could be easily identified by its very light isotope signature. However, carbon contaminants were more difficult to discern through isotope data alone and so, ^{1}H NMR was employed to assess potential organic carbon contamination. Figure 3.3 shows that most of the organic contaminants possessed very distinct aromatic structures as expected from this polyaromatic resin. Some samples also showed the presence of phthalate (plasticizer), the source of which was isolated to some HDPE barrels. As with the other contaminants, phthalate has an easily identifiable ^{1}H NMR spectrum (Fig. 3.4). As a result of these analyses the washing protocol outlined in section 3.2.2 was designed and it was found that washing and conditioning of the resin using this sequence of organic solvents produces a clean medium for DOM extraction In summary, organic contaminants from the resin could be easily identified through isotope values or NMR, and were rarely seen in samples. When contaminants were present in samples, we excluded them from the data set.

3.3.2 Elemental and Isotope Data

Table 3.2 shows the average carbon content (%C), carbon to nitrogen elemental

ratio (C:N), and isotopic signature of individual fractions collected during each cruise. Fraction C had a higher C:N than Fraction B in both surface and deep waters. For example, in surface waters during 0411 the average C:N (±S.D.) was 27 (±3) and 42 (±6) for Fraction B and C, respectively. At the NELHA site C:N (±S.D. of replicates) was 21(±1) for Fraction B and 32(±4) for C at 670 m. In all cases, the C:N of SPE fractions was elevated relative to the C:N of UDOM which was 16 (±2) in surface samples and 17 in the deep sample (Table 3.2). These C:N values for UDOM are consistent with data from other marine regions (Benner, 2002; Meador et al., 2007). Relative to DOM, suspended POM in the surface ocean was enriched in N with C:N of 9±1 during the spring and 8±1 in the fall.

Suspended POM samples collected throughout the region had an average $\delta^{13}C$ (±S.D.) value of -22.0‰ (±1.5‰) during both seasons. In surface waters similar values were also observed for UDOM (-22.1‰±0.3‰) and SPE fraction B (-22.6‰±0.5‰ and -22.8‰±0.3‰ during 0404 and 0411, respectively). More depleted $\delta^{13}C$ values were measured from Fraction C: -24.9‰ (±0.9‰) during 0404 and -24.0‰ (±0.6‰) during 0411. In general, $\delta^{13}C$ values reported in Table 3.2 are consistent with previous studies of marine POM, SPE DOM, and UDOM. For example, suspended POM in surface waters of the upper slope, rise, and offshore areas of the eastern North Pacific (just north of the CalCOFI region) had $\delta^{13}C$ signatures between -22.9‰ and -24.7‰ (Druffel et al., 1998). Druffel et al. (1992) reported $\delta^{13}C$ values in the range -22‰ to -23.5‰ for marine humic substances isolated onto XAD resins from pre-acidified seawater (~15-20% of the total DOC). In addition, values reported for UDOM are typical of marine samples (Benner, 2002).

The overall trend for UDOM and SPE fractions ranges from polar/semi-polar chemical fractions (UDOM and Fraction B) with lower C:N and marine-like $\delta^{13}C$ values to non-polar fractions (e.g., Fraction C) with higher C:N and relatively depleted $\delta^{13}C$ values. The fraction-specific $\delta^{13}C$ patterns are reminiscent of the carbon isotope fractionation observed for the biosynthesis of lipids where cellular, polar biochemicals are enriched in ^{13}C relative to cellular lipids (Deniro and Epstein, 1977). The total lipid fraction isolated from marine suspended and sinking particles for example, has also been shown to be depleted in ^{13}C relative to polar biochemical fractions such as carbohydrates and proteins on these same particles (Hwang et al., 2003).

To observe differences in sources and cycling of nitrogen the $\delta^{15}N$ signature within each organic matter fraction was also examined. Values for suspended POM samples averaged 5.8‰ (±1.1‰) during the spring and 7.1‰ (±1.2‰) during the fall. Overall, $\delta^{15}N$ values of suspended POM were more variable than $\delta^{15}N$ values of surface SPE samples (Table 3.2; measurement error is ±0.5‰). Previous studies have observed that $\delta^{15}N$ values of POM and DOM vary independently in the surface ocean (Knapp et al., 2005; Meador et al., 2007,). Similar $\delta^{15}N$ values were observed for all surface DOM fractions examined in this study (SPE and UDOM) – in the range of 4‰ to 5‰. A recent study reported $\delta^{15}N$ values in the same range for several UDOM samples isolated from surface ocean sites throughout the North Pacific (5.4.±0.7‰, n = 16; Meador et al., 2007). Based on these data we conclude that similar processes must act to determine the $\delta^{15}N$ signature of surface SPE fractions and UDOM despite significant differences in the C:N of these DOM fractions. Two data points from 1000 m in the CalCOFI domain indicated that deep UDOM and SPE samples are enriched in ^{15}N compared to their

surface counterparts. However, the $\delta^{15}N$ signature of each SPE fraction isolated from 670 m at NELHA is indistinguishable from surface Pacific Ocean values. At present our data set is not large enough to distinguish reproducible differences in the $\delta^{15}N$ signature of surface and deep DOM.

3.3.3 ^{1}H NMR data

We used ^{1}H NMR spectroscopy to analyze and compare the chemical functional groups present within SPE and UDOC fraction. Figure 3.5 shows these basic functional groups and their respective integrated areas. As shown in Figure 3.6 and 3.7, several chemical moieties were distinguished in the ^{1}H NMR spectrum of each fraction. Beginning with downfield resonances the quantified regions of the spectrum included aromatic protons (6.0-8.0 ppm), anomeric protons of carbohydrates (4.9-6.0 ppm), protons geminal to hydroxyl or amino functionalities (3.0-4.7 ppm), protons attached to carbons alpha (α) to a heteroatom (1.5-3.0 ppm), and lipid/alkyl protons (0.0-1.5 ppm). Table 3.3 reports these integrations as a percentage of the total integrated area across the entire spectrum for several representative samples. We assume here that the integration accurately reflects the relative abundance of each functional group within samples as ^{1}H NMR spectroscopy is expected to detect all protons without bias.

Spectra of particular SPE fractions (e.g., Fraction B) are similar across samples and show little seasonal variation. However, differences in composition do exist among fractions and are consistent with the solvent partitioning scheme applied during sample isolation (Figure 3.2). Fraction B contained the least amount of aliphatic resonances and the greatest amount of polar resonances detected in SPE samples. For example, proton

resonances in the region between 0 and 1.5 ppm (aliphatic region) accounted for ~32±2% of the total area in Fraction B versus 40-45% of the total area in Fraction C (Table 3.3). In contrast, the region between 3.0 and 4.5 ppm (protons geminal to hydroxyl or amino groups) contained 30±3% of proton resonances in Fraction B but only 19±1% of resonances in Fraction C. In the case of Fraction CL, most proton resonances were present in the aliphatic region of the spectrum (Figure 3.6).

Surface UDOM (Figure 3.7) was enriched in polar material with only 23% of protons present in the aliphatic region and 41% of protons present geminal to hydroxyl or amino functional groups (Table 3.3). This distribution of functional groups in UDOM is consistent with previous reports based on ^{1}H NMR spectroscopy (Benner et al., 1992; Aluwihare et al., 2002). The abundance of aromatic protons varied across samples, but was usually low (<5% of the total area). Based on these NMR data, UDOM contained the highest percentage of polar functional groups, followed by Fraction B, Fraction C and finally, Fraction CL. These compositional trends are consistent with fraction-specific C:N discussed in the previous section and the δ^{13}C pattern expected for biochemicals rich in polar versus aliphatic functional groups.

Effects of the water-ethyl acetate partition on sample composition are clearly noticeable in Figure 3.4. Fractions C and CL were first eluted as one combined sample with 100% methanol and then partitioned between water (C) and ethyl acetate (CL). Nuclear magnetic resonance spectra of C and CL were acquired in different deuterated solvents (D_2O versus CD_3OD) and therefore, cannot be compared quantitatively; however, it is clear from the spectrum of CL that compounds dominated by aliphatic functional groups can be separated from semi-polar DOM by simple solvent extraction.

Deep SPE samples from 0411 and NELHA had fewer polar ^{1}H NMR resonances than their surface counterparts (Figure 3.8 and Table 3.3). In the case of deep Fraction B, for example, only 19% to 21% of proton resonances were present in the 3.5 to 4.5 ppm region of the spectrum. This trend between surface and deep samples was also apparent for UDOM (Figure 3.7). Fraction B collected from 1000 m was enriched in aromatic resonances relative to surface SPE fractions; lower amounts of aromatic resonances in surface samples may be due to photooxidation of these compounds at the surface (Mopper et al., 1991).

NMR data confirmed that Fractions C and CL were enriched in lipids and hydrophobic compounds relative to UDOM and SPE fraction B. Furthermore, deep UDOM and surface Fraction B shared a similar distribution of functional groups as identified by ^{1}H NMR spectroscopy. NMR data, stable isotope signatures and elemental ratios together showed that the fractionation scheme developed for this study effectively separated DOM into fractions with some unique chemical properties. In this regard, the SPE method described in the current paper appears to isolate a more diverse mixture of compounds than C-18 and XAD resins.

3.3.4 Radiocarbon data

The average Δ^{14}C value for suspended POC (±S.D.) isolated from stations 77.60, 93.50, 87.55, 80.80, and 82.47 during 0404 was 22‰±15‰. During 0411, the average Δ^{14}C value of POC isolated from surface waters was 21‰±19‰. Highest values observed for POC (± measurement error) were 49‰±4‰ during 0404 and 50‰±9‰ during 0411. In May 2006 the Δ^{14}C signature of dissolved inorganic carbon (DIC; ±

measurement error) ranged from 16‰±3‰ to 49‰±4‰ in the upper 200 m at 3 sites within the CalCOFI region. These values are in good agreement with average $\Delta^{14}C$ values reported above for suspended POM. Three suspended POM counterparts to SPE samples had depleted$\Delta^{14}C$ signatures and these data are discussed in section 3.3.5.

Most SPE fractions were depleted in ^{14}C compared to POM samples isolated at the same site (the only exception was station 90.53 sampled during 0404). Average $\Delta^{14}C$ values for SPE fractions B, C and CL isolated during 0404 were -124‰±17‰, -158‰±23‰, and -166‰±57‰, respectively (n=9; Table 3.2). During 0411, $\Delta^{14}C$ values were -100‰±7‰, -100‰±17‰ and -120‰±47‰ for fractions B, C, and CL, respectively (Table 3.2).

Figure 3.9 (a and b) shows the $\Delta^{14}C$ signature of individual SPE fractions collected during 0404 (3.9a) and 0411 (3.9b). The lipid-rich fraction, CL, had the most variable $\Delta^{14}C$ signature and in most cases, this fraction also had the most ^{14}C-depleted signature. Notable exceptions are CL fractions isolated from nearshore stations 90.30 and 93.30 during 0404 and station 90.30 during 0411. During 0404 Fraction B was typically enriched in ^{14}C relative to other fractions (only two stations, 77.60 and 77.90, departed from this trend). In contrast, during the fall Fraction C was either more enriched in ^{14}C than Fraction B or both fractions had similar $\Delta^{14}C$ signatures. If 0404 samples from station 77.60 and 77.90 (see below; Figure 3.9a) are omitted from the comparison then the $\Delta^{14}C$ signature of Fraction B during both seasons is comparable: -111‰±13‰ and -100‰±7‰, during 0404 and 0411, respectively. However, 0411

Fractions C and CL were consistently enriched in ^{14}C-compared to 0404 samples and in general, 0411 $\Delta^{14}C$ signatures showed less spatial variability (see Figures 3.9a and b).

The average surface ocean TOC concentration in the general CalCOFI region was 64±8 μM C in the spring, not significantly different from the average fall concentration of 69±6 μM C. However, the average TOC concentration at stations where SPE samples were collected was 62±6 μM C during 0404 as compared with 72±8 μM C during 0411. During 0404 stations 77.60 and 77.90 had the lowest TOC concentrations measured at a surface station (51±3 μM C; no comparable concentrations were observed in the CalCOFI region during 0411). Station 77.60, which had the lowest $\Delta^{14}C$ signature of any Fraction B or C (Figure 3.9a), also had a surface water temperature of 12.7ºC and a surface ocean nitrate concentration of 3.1 μM. These data suggest that an upwelling event had recently occurred at or near station 77.60 during 0404; such an event would mix the water column and entrain ^{14}C-depleted DOM with similar chemical properties from subsurface waters into the surface ocean. For example, Fraction C from 1000 m is depleted in ^{14}C relative to the same fraction in surface waters (Table 3.2). Although data are only available from deep SPE samples in 0411, we expect no seasonal differences in $\Delta^{14}C$ of SPE at 1000 m. Therefore, a greater contribution of subsurface DOC would significantly deplete the $\Delta^{14}C$ value of Fraction C in surface waters. In summary we attribute the observed ^{14}C-enrichment (particularly for Fraction C) and higher TOC concentration in fall samples to both a more stable water column during 0411 (Figure 3.10) and the addition and accumulation of some fresh

DOC between spring and fall. A more stable water column in 0411 would allow for the enrichment of $\Delta^{14}C$ in DIC.

Three surface UDOM samples isolated during 0310 had an average $\Delta^{14}C$ value of -50‰±46‰ (individual values were -2‰ (station 93.30), -75‰ (77.80) and -85‰ (83.60)). Two of the UDOM samples had values that were only slightly enriched in ^{14}C relative to the average $\Delta^{14}C$ signature of Fraction B and C isolated during 0411. The nearshore UDOM sample (93.30) was relatively enriched in ^{14}C suggesting that a higher percentage of modern DOC was present at this location.

3.3.5 Isotopically depleted POM samples

During 0404, suspended POC samples collected at station 90.53 and 83.60 had much lower $\Delta^{14}C$ values: -116‰ and -42‰, respectively. During 0411 POC isolated from surface waters at station 83.110 also had a depleted $\Delta^{14}C$ signature (-29‰). At station 83.60 density profiles and surface nitrate concentrations (2.2 μM) indicated POC from deeper waters was entrained into the surface ocean. The water that recent upwelling had mixed the water column, and it is likely that some suspended column at 90.53 had also been recently mixed and this station had the highest surface ocean nitrate concentration (4.5 μM) measured along Line 90 during this particular cruise. Figure 3.10 shows the relationship between depth and potential density (σ_θ) during 0404 and 0411 along Line 83 (station 83.55 to station 83.110). These profiles confirm a greater degree of mixing between surface and subsurface waters during 0404. Although the $\delta^{13}C$ signature of these POM samples was marine (-23.2‰, -21.2‰ and -23.2‰ for 0404-

90.53, 0404-83.60, and 0411-83.110, respectively), the observed $\Delta^{14}C$ values are more depleted than previous reports in the literature (e.g. Druffel et al., 1996; 2000). Therefore, we cannot rule out the possibility that these POM samples were contaminated by a ^{14}C-depleted component during the filtration procedure.

3.3.6 Vertical distribution of $\Delta^{14}C$

Using DOC concentration measurements and total DOC radiocarbon mass balances it has been inferred that refractory DOC with a uniform $\Delta^{14}C$ signature is present throughout the water column (Williams and Druffel, 1987; Mortazavi and Chanton, 2004). The fact that surface DOM fractions are depleted in ^{14}C relative to surface suspended POM and DIC in the CalCOFI region confirms that all DOM fractions contain some refractory DOC. However, as our surface SPE fractions and UDOM samples were enriched in ^{14}C relative to their deep counterparts, modern DOC must be added to every fraction in surface waters. Radiocarbon values are not available for total DOC at the sites sampled during this study; however, a sample collected during October/November 2004 at a station about 1 degree to the north of CalCOFI station 77.80 yielded a $\Delta^{14}C$ value of -303‰±2‰ for total DOC (concentration ~70 μM C; Beaupre et al., 2007). This value is more depleted than any $\Delta^{14}C$ value measured for surface SPE fractions. Previous studies have suggested that UDOM is enriched in a modern component relative to the total DOC pool; for example, highly enriched $\Delta^{14}C$ values were observed for UDOM isolated from surface waters near NELHA (10‰ and 46‰ for two samples; Repeta and Aluwihare, 2006). Our surface data from the eastern North Pacific are consistent with this observation, but values here are more depleted than

those observed for DOM fractions at the more stratified open ocean locations like NELHA. In the case of deep samples, the $\Delta^{14}C$ signature of Fractions C and CL from NELHA (670m) and the CalCOFI region (1000 m) are similar to or more depleted than the $\Delta^{14}C$ signature of total DOC at comparable depths in the eastern and central North Pacific. This is in contrast to deep UDOM which has been shown to be enriched in ^{14}C relative to total DOC at comparable depths (this study; Aluwihare et al., 2002; Loh et al., 2004; Repeta and Aluwihare, 2006). As part of the chemical separation we isolated relatively non-polar samples (Fraction D and DL; Figure 3.2) that were depleted in ^{14}C relative to total DOC in both surface and deep waters; however, contamination of this fraction from the resins and sampling equipment could not be ruled out as the source of ^{14}C-depleted material (see above).

The narrow range of $\Delta^{14}C$ values observed for SPE fractions B and C during 0411 (when surface waters were more stratified than in the spring) suggests that the distribution of ^{14}C within these fractions is similar. In comparison, some surface UDOM was slightly enriched in ^{14}C and Fraction CL was depleted in ^{14}C showing that these fractions contained either more or less modern DOC as compared with fractions B and C. Fractions isolated from 670 m at the NELHA site showed the greatest range in $\Delta^{14}C$ values suggesting that individual fractions have distinct residence times at these depths in the ocean. However, Fractions C and CL isolated from 1000 m in the CalCOFI domain have relatively similar $\Delta^{14}C$ values. The progression in $\Delta^{14}C$ values with depth (surface 0404 and 0411, NELHA, and 1000 m) could be interpreted as the progressive removal of a modern component as water masses are advected and age, until background $\Delta^{14}C$ values for each fraction are approached. Data from NELHA also suggest that the

modern DOC in individual fractions does not have the same reactivity and/or different amounts of modern DOC are added to each fraction at the surface (Table 3.2). However, more samples from the deep ocean are required to confirm these preliminary interpretations.

On the basis of the data shown here we conclude that ^{14}C-depleted, refractory DOM accumulating in the deep ocean has a more reduced chemical structure. This conclusion is supported by the fraction-specific $\Delta^{14}C$ data and the surface to deep gradients in chemical composition identified by NMR of UDOM and SPE (e.g. this study; Benner et al., 1992 (^{13}C NMR); Aluwihare et al., 2002 (^{1}H NMR)). In addition, vertical gradients in C:N of total DOM also support this conclusion (Loh et al., 2000; Hopkinson and Vallino, 2005).

3.3.7 Relationships between Chemical Composition and Radiocarbon Signature

As discussed previously, each surface SPE fraction is enriched in ^{14}C relative to subsurface samples. We interpret this pattern as the addition of modern DOC ($\Delta^{14}C > -50‰$) to each fraction in surface waters. The major compositional difference we observe between UDOM and Fraction CL, for example, and surface and deep fractions, is the loss of NMR resonances corresponding to polar functional groups (compare Figure 3.6 to Figure 3.7; Figure 3.7,). In addition, these compositional differences are accompanied by variations in $\Delta^{14}C$ signature (see Table 3.2 and Figure 3.9a and b). Finally, differences between the spring and fall, and surface and deep samples require that at least two $\Delta^{14}C$ groups are present within each DOC fraction. Based on these observations and assumptions and the simplest case of a 2-component mixing model, we

now use the NMR data to estimate the relative contribution of refractory DOC to individual fractions isolated in this study. For this exercise we estimate lipid (reduced chemical component) abundance using the NMR region between 0 and 1.5 ppm and assume that this component represents refractory DOM. We then include radiocarbon data in the mass balance to constrain the accuracy of this assumption. The mass balance is as follows:

$$\Delta^{14}C = (-550‰ \times (\% \text{ Lipid})) + (\Delta^{14}C\text{-DIC} \times (\% \text{ Oxidized}))$$

where -550‰ is the $\Delta^{14}C$ signature of background refractory DOC according to Bauer et al. (1998). The % Lipid as carbon is calculated according to the ^{1}H NMR integration between 0-1.5 ppm and then weighted for C:H ratios based on chemical shifts (e.g., lipid resonances in the range 0 to 1.5 ppm are expected to have an average C:H ratio of 1:2); % Oxidized is the remainder of the integrated NMR spectrum (1.5-8.0 ppm) and the C:H ratio in these functional groups is expected to be 1:1. (See Table 3.4 for calculated fraction-specific values). The $\Delta^{14}C$-DIC is the $\Delta^{14}C$ value of the local DIC approximated from previous measurements to be 25‰ in surface waters and -190‰ at 1000 m in the CalCOFI domain. At NELHA $\Delta^{14}C$-DIC was found to be -155‰ at 670 m (Repeta and Aluwihare, 2006). We use local $\Delta^{14}C$-DIC at 670 m and 1000 m because we assume that "modern" DOC at these depths (mesopelagic) was advected from the surface ocean and has "aged" to the same extent as DIC at the same depth.

From the values estimated using this mass balance (Table 3.5) it can be seen that the simple model of DOC being comprised of both a refractory lipid-rich component and a modern component enriched in polar functional groups, effectively approximates the observed $\Delta^{14}C$ values. Other studies have also suggested that lipids within DOC are

depleted in ^{14}C (Wang et al., 2001; Loh et al., 2004) and represent the refractory background DOC component (Hertkorn et al., 2006). The mass balance works well for to the simple assumption in our model, this finding requires the region between 1.5 and 8 ppm in each deep NMR spectrum to also contain a contribution from relatively refractory compounds.

3.4 CONCLUSION

In summary, based on compositional data, stable isotope and elemental data and patterns observed for $\Delta^{14}C$ of individual fractions, we constructed a composition-based radiocarbon mass balance for isolated DOC fractions. This mass balance showed that a lipid-rich refractory component and a predominantly oxidized modern component can adequately reproduce the measured ^{14}C distribution in SPE and UDOM fractions. This mass balance uses a unique approach to confirm that only two major radiocarbon components are needed to satisfy the $\Delta^{14}C$ distribution in DOM (consistent with previous studies, e.g., Williams and Druffel, 1987). Further, elemental and isotopic data of fractions examined in this study support a relationship between chemical composition and DOC residence time in the ocean. To confirm these initial results future studies will need to purify each SPE fraction further and make compound-specific $\Delta^{14}C$ measurements.

3.5 ACKNOWLEDGMENTS

Chapter 3, in part, will be submitted for publication with Dr. Lihini I. Aluwihare and Dr. Michaele Kashgarian. The dissertation author was the primary investigator and author of this paper. We thank the CalCOFI technicians and crews of the R/V New Horizon and R/V Roger Revelle for their technical assistance.

Table 3.1 Quantity and isotopic compositions of blank fractions.

Blank	**B**	**C**	**CL**	**D**	**DL**
Amount Carbon	10 ug	18 ug	31 ug	17 ug	34 ug
$\delta^{15}N$	-4.55‰				
$\delta^{13}C$	-25.5‰				
$\Delta^{14}C$	-971‰				

Table 3.2 Elemental ratios (C:N), stable isotope signatures ($\delta^{13}C$ and $\delta^{15}N$) and radiocarbon content ($\Delta^{14}C$) of organic matter fractions in the North Pacific. In the case of surface samples 0404, 0411 and 0310, the average for each fraction is reported with standard deviations (± 1σ), all other data are for single samples. Error for $\Delta^{14}C$ values were generally less than 9‰.

Cruise	Fraction	% C	C:N ratio	$\delta^{13}C$ (‰)	$\delta^{15}N$ (‰)	$\Delta^{14}C$ (‰)
0404	B (n = 11)	37% (±6)	25 (±3)	-22.6 (±0.5)	4.6 (±0.5)	-124 (±17)
	C (n = 9)	42% (±14)	35 (±8)	-24.9 (±0.9)	4.2 (±0.7)	-158 (±23)
	CL (n = 11)	44% (±18)	-	-	-	-166(±57)
	POM (n = 5)	5% (±3)	9 (±1)	-22.0 (±1.6)	5.8 (±1.1)	3 (±35)
0411	B (n = 10)	39% (±3)	27 (±3)	-22.8 (±0.3)	5.0 (±0.4)	-100 (±7)
	C (n = 10)	46% (±2)	42 (±6.)	-24.0 (±0.6)	4.5 (±0.4)	-100(±17)
	CL (n = 10)	52% (±15)	-	-	-	-121(±47)
	POM (n = 8)	3% (±1)	8 (±1)	-22.0 (±1.9)	7.1 (±1.2)	21 (±19)
0411 (1000 m)	B	1%	-	-23.3	-	n.d.
	C	41%	25	-23.8	6.9	-427
	CL	n.d	-	-	-	-454
NELHA	B	51%	22	-22.5	5.4	-297
	C	41%	35	-23.1	4.1	-391
	CL	n.d	-	-	-	-478
0310	UDOM (n = 3)	18% (±9)	16 (±2)	-22.1 (±0.3)	4.9 (±0.1)	-50 (±46)
0310 (1000 m)	UDOM	6%	17	-21.1	6.0	-335

Table 3.3 ^{1}H NMR integration values for individual DOM samples.

ppm	0.0-1.5	1.5-3.0	3.5-4.7	4.9-6.0	6.0-8.0		0.0-1.5	1.5-3.0	3.5-4.7	4.9-6.0	6.0-8.0
Cruise-stn											
0404-B						0404-C					
93.30	31%	29%	30%	6%	3%		44%	30%	19%	4%	4%
93.50	32%	29%	30%	6%	3%		46%	27%	16%	4%	7%
93.120	34%	30%	27%	6%	3%		46%	29%	18%	4%	2%
90.53	34%	31%	26%	6%	3%		45%	30%	16%	5%	4%
90.30	33%	30%	29%	6%	3%		47%	29%	18%	4%	2%
87.55	30%	30%	30%	6%	4%		44%	30%	18%	5%	4%
83.60	30%	27%	35%	6%	3%		43%	30%	19%	5%	3%
82.47	28%	27%	35%	7%	4%		44%	30%	19%	4%	3%
80.80	33%	30%	26%	7%	4%		43%	31%	17%	5%	3%
average	***32%***	***29%***	***30%***	***6%***	***3%***		***45%***	***29%***	***18%***	***4%***	***4%***
0411-B						0411-C					
93.80	34%	29%	28%	6%	3%		41%	29%	19%	6%	4%
90.100	32%	29%	29%	6%	3%		39%	30%	20%	6%	4%
90.70	32%	30%	29%	6%	3%		39%	31%	20%	6%	4%
90.30	32%	29%	30%	6%	3%		37%	32%	20%	7%	4%
87.35	30%	29%	31%	7%	4%		41%	31%	19%	6%	4%
87.80	34%	30%	28%	6%	3%		41%	32%	18%	6%	4%
83.110	33%	29%	29%	6%	3%		40%	31%	20%	6%	3%
83.60	32%	31%	28%	7%	4%		41%	31%	19%	6%	3%
77.60	33%	31%	27%	6%	3%		40%	31%	19%	6%	4%
average	***32%***	***30%***	***29%***	***6%***	***3%***		***40%***	***31%***	***19%***	***6%***	***4%***
Deep-B (1000 m)	34%	34%	19%	6%	7%	Deep-B (1000 m)	48%	29%	16%	3%	4%
NEHLA-B (670 m)	36%	35%	21%	5%	3%	NEHLA-C (670 m)	31%	30%	22%	5%	6%
0310 UDOC	23%	28%	41%	6%	4%						
0310 UDOC (1000 m)	28%	32%	29%	6%	6%						

Table 3.4 Values calculated from 2-component model for individual samples.

	Lipid C	**Polar C**	**model $\Delta^{14}C$**	**Meas. $\Delta^{14}C$**	**[TOC] µM**		**Lipid C**	**Polar C**	**Model $\Delta^{14}C$**	**Meas. $\Delta^{14}C$**
Cruise-stn										
0404-B						0404-C				
93.30	19%	81%	-82	-85	69.0		28%	72%	-135	-197
93.50	19%	81%	-84	-102	65.7		30%	70%	-147	-
93.120	20%	80%	-92	-120	57.6		30%	70%	-147	-172
90.53	21%	79%	-93	-130	55.3		29%	71%	-141	-150
90.30	20%	80%	-88	-104	59.1		31%	69%	-153	-179
87.55	18%	82%	-77	-109	62.0		28%	72%	-135	-154
83.60	18%	82%	-76	-117	63.1		28%	72%	-134	-126
82.47	16%	84%	-68	-111	70.1		28%	72%	-138	-146
80.80	20%	80%	-89	-120	61.5		28%	72%	-134	-
average	19%	81%	-83	-111	62		29%	71%	-140	-160
stdev	1%	1%	8	13	6				7	24
0411-B						0411-C				
93.80	20%	80%	-91	-108	84.7		26%	74%	-124	-133
90.100	19%	81%	-85	-90	70.6		24%	76%	-115	-101
90.70	19%	81%	-84	-105	60.0		25%	75%	-116	-119
90.30	19%	81%	-86	-103	69.6		23%	77%	-106	-105
87.35	18%	82%	-77	-91	78.7		25%	75%	-121	-97
87.80	20%	80%	-91	-102	68.8		25%	75%	-121	-93
83.110	20%	80%	-89	-106	66.3		25%	75%	-121	-98
83.60	19%	81%	-83	-92	72.9		26%	74%	-124	-74
77.60	20%	80%	-88	-105	63.3		25%	75%	-119	-81
average	19%	81%	-86	-100	72		25%	75%	-118	-100
stdev	1%	1%	5	7	8				6	18
Deep-B	20%	80%	-271	n.d	-	Deep-C	31%	69%	-310	-427
NEHLA-B	22%	78%	-241	-297	-	NEHLA-C	20%	80%	-234	-391
0310 UDOM	13%	87%	-48	-50.1	-					
Deep UDOM	16%	84%	-256	-335	-					

Table 3.5 Calculated average $\Delta^{14}C$ values resulting from the 2-component mass and radiocarbon balance (data for individual stations is provided in Table 3.4). Note: NMR results from Line 77 were not used for 0404-B, which is why the average measured $\Delta^{14}C$ value for 0404-B in Table 1 and 2 does not match.

	% Lipid carbon	% Polar carbon	Estimated $\Delta^{14}C$ (‰)	Measured $\Delta^{14}C$ (‰)	[TOC] (μM)
0404-B	19%	81%	-83 (±8)	-111 (±13)	62 (±6)
0411-B	19%	81%	-86 (±7)	-101 (±7)	72 (±8)
Deep-B	20%	80%	-271	n.d	40
NELHA-B	22%	78%	-241	-297	40
0310 UDOC	13%	87%	-48	-50	65
Deep UDOC	16%	84%	-256	-335	40
0404-C	29%	71%	-140 (±12)	-161 (±24)	62 (±6)
0411-C	25%	75%	-118 (±5)	-100 (±17)	72 (±8)
Deep-C	31%	69%	-310	-427	40
NELHA-C	20%	80%	-234	-391	40

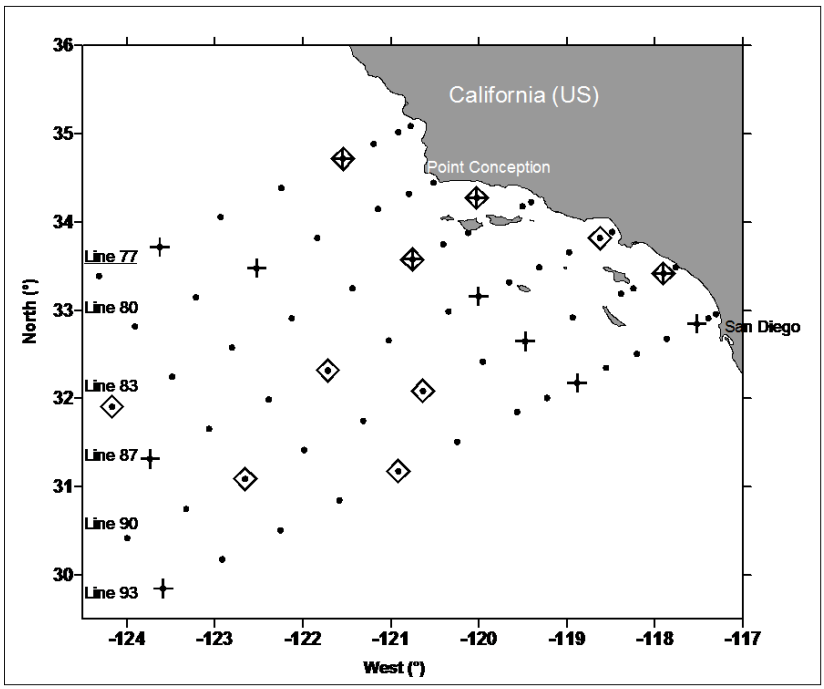

Figure 3.1 Sample locations: Crosses (+) identify sampling sites during April 2004 aboard R/V New Horizon (0404), and diamonds (◊) identify sites sampled during November 2004 aboard R/V Roger Revelle (0411). Station numbers and geographic coordinates can be obtained at www.calcofi.org.

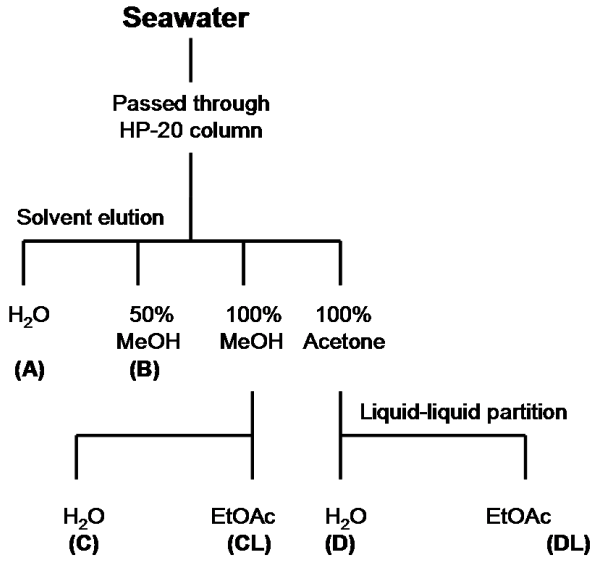

Figure 3.2 DOM fractionation scheme

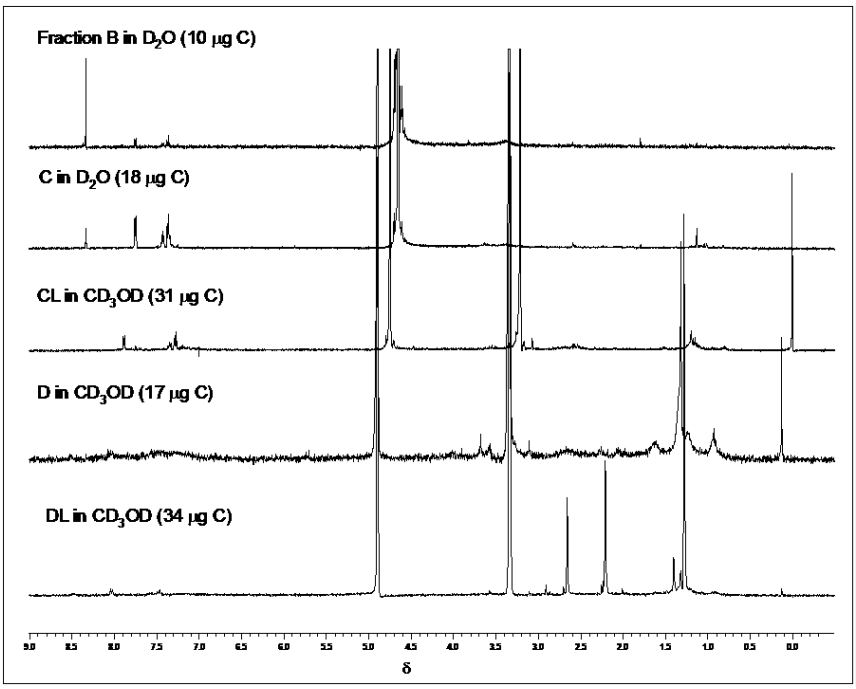

Figure 3.3 ^{1}H NMR spectra (0-9 ppm) of fraction-specific column blanks. Shown on the figure is the solvent in which NMR spectra were acquired as well as the typical fraction-specific yield of blank carbon from new resin prior to washing with solvents.

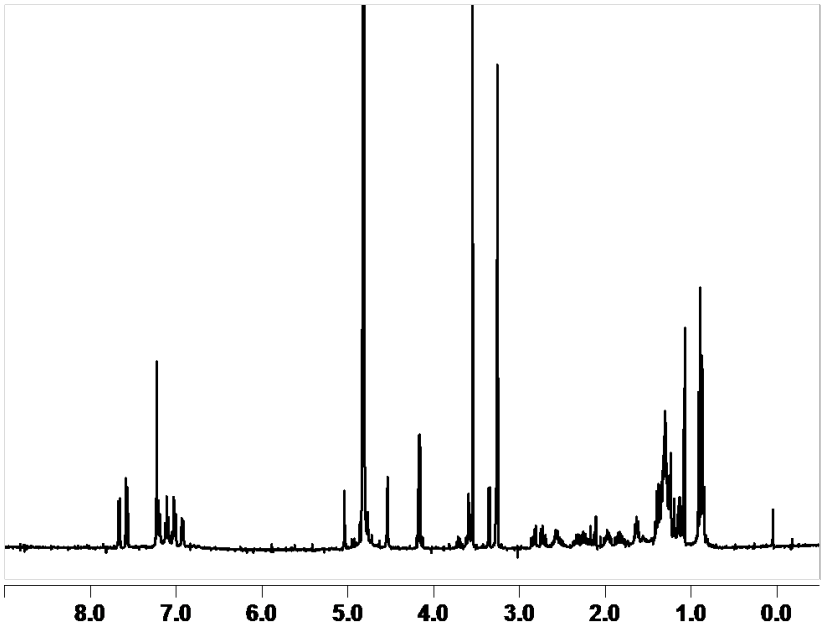

Figure 3.4. ^{1}H NMR spectrum of diisooctyl phthalate and resin contaminant in $(CD_3)_2CO$ (0-9 ppm).

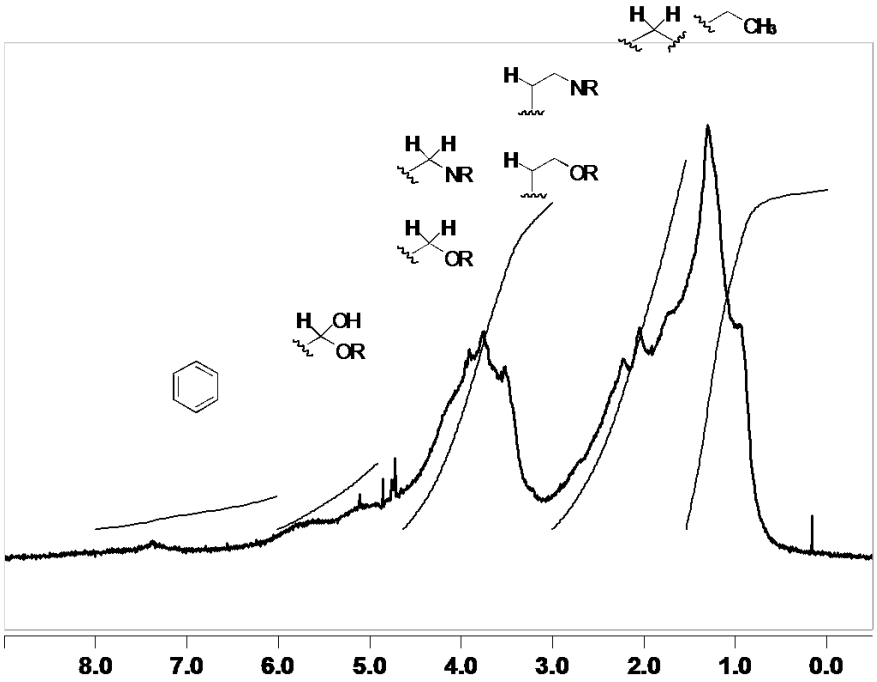

Figure 3.5 ^{1}H NMR integration of a representative Fraction B sample: 0-1.5, 1.5-3.0, 3.0-4.7, 4.9-6.0, 6.0-8.0 ppm.

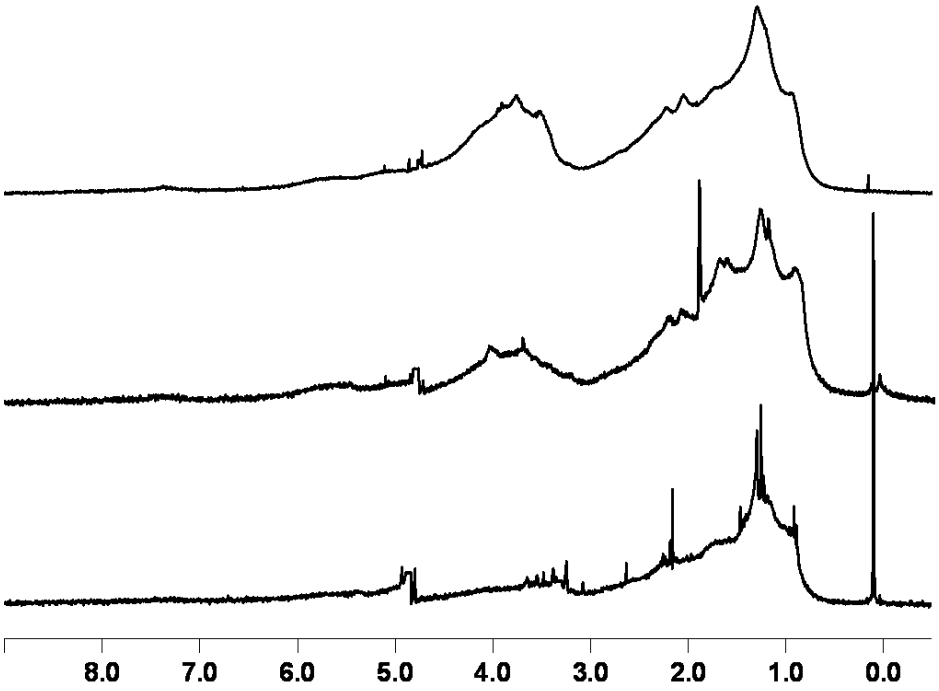

Figure 3.6 ^{1}H NMR of surface fractions (Top to bottom) 0411-87.70 B, C, CL.

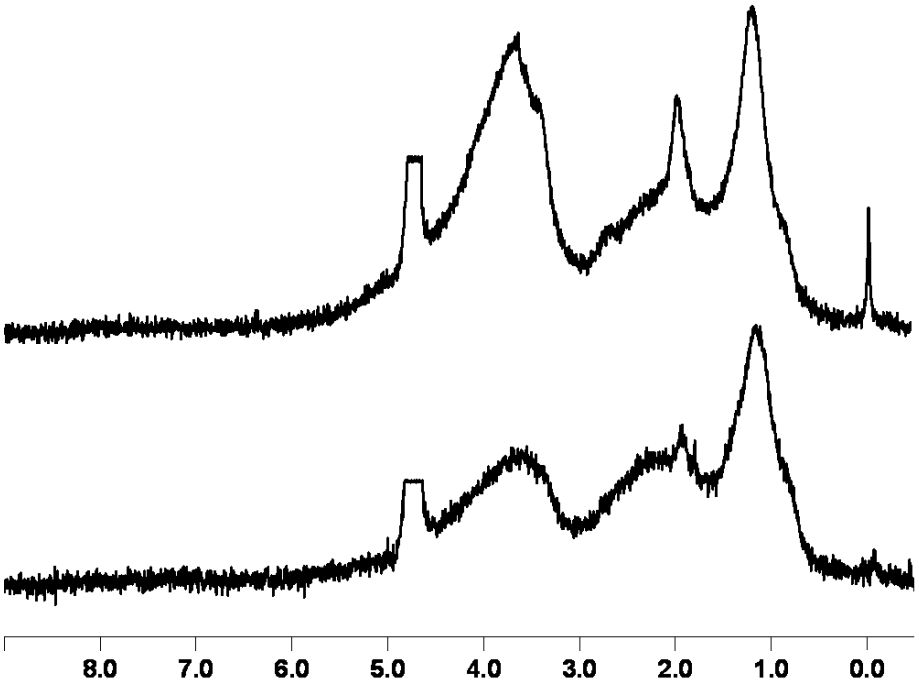

Figure 3.7 ^{1}H NMR of 0310 UDOM samples top: surface bottom: deep (1000m).

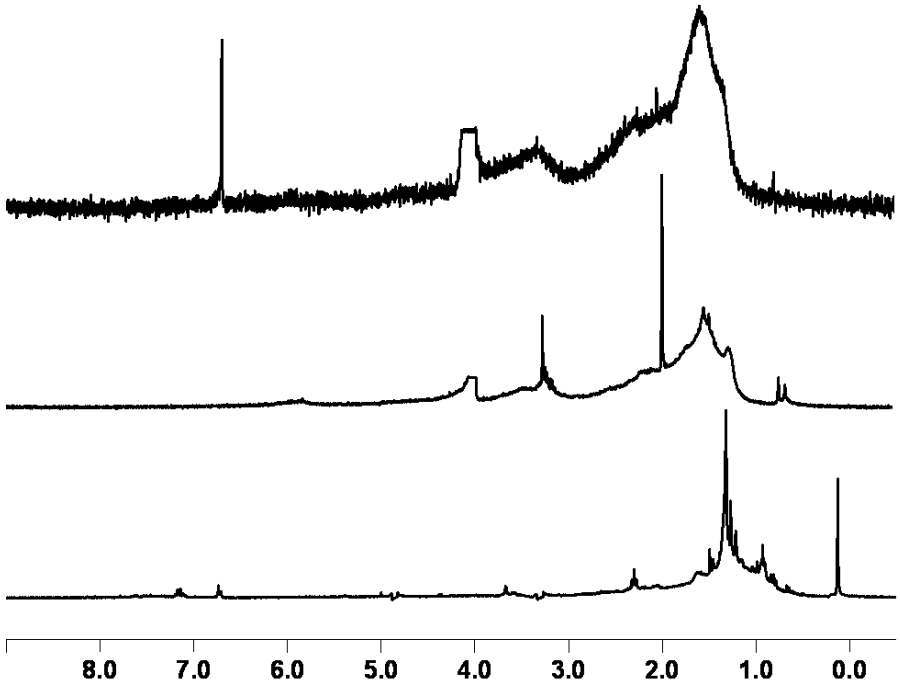

Figure 3.8 ^{1}H NMR of 0411 Deep (Top to bottom) B, C, CL.

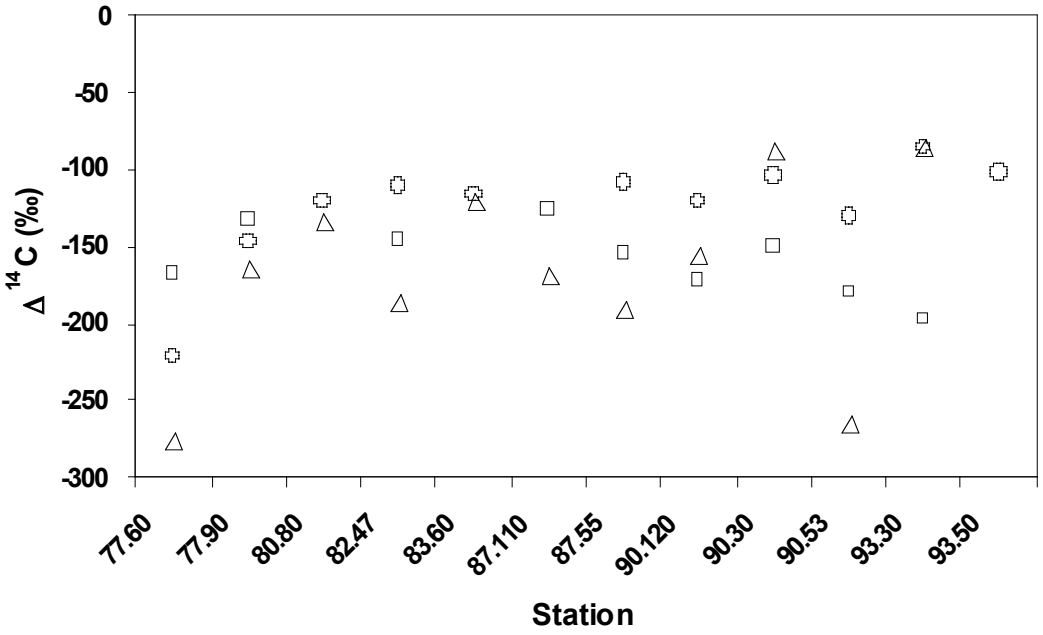

Figure 3.9a Cruise 0404 station data for fraction B (open circle), C (open square) and CL (open triangle).

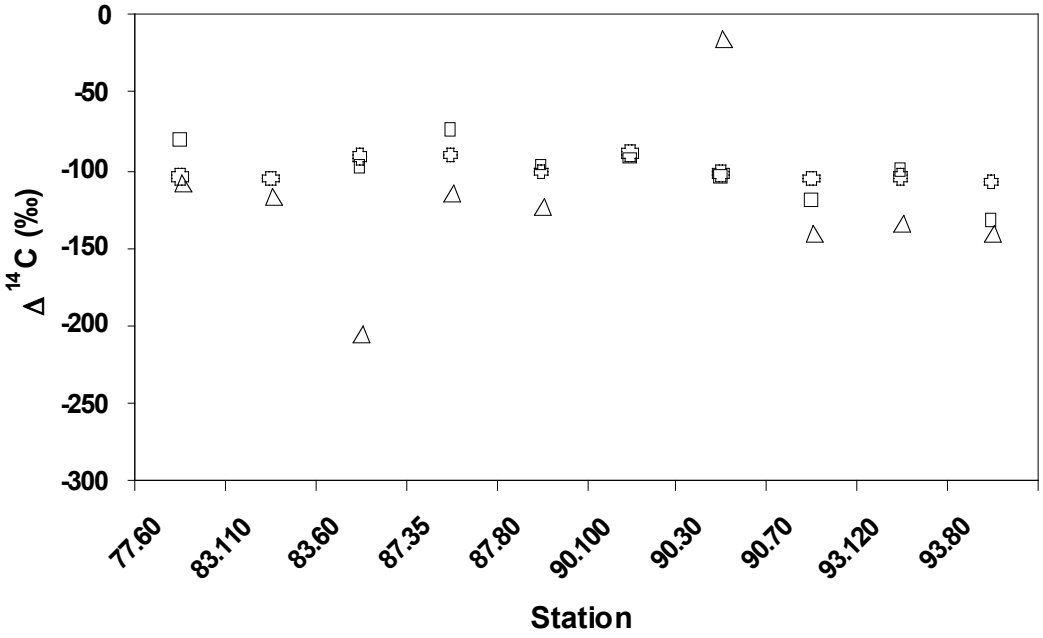

Figure 3.9b Cruise 0411 station data for fraction B (open circle), C (open square) and CL (open triangle).

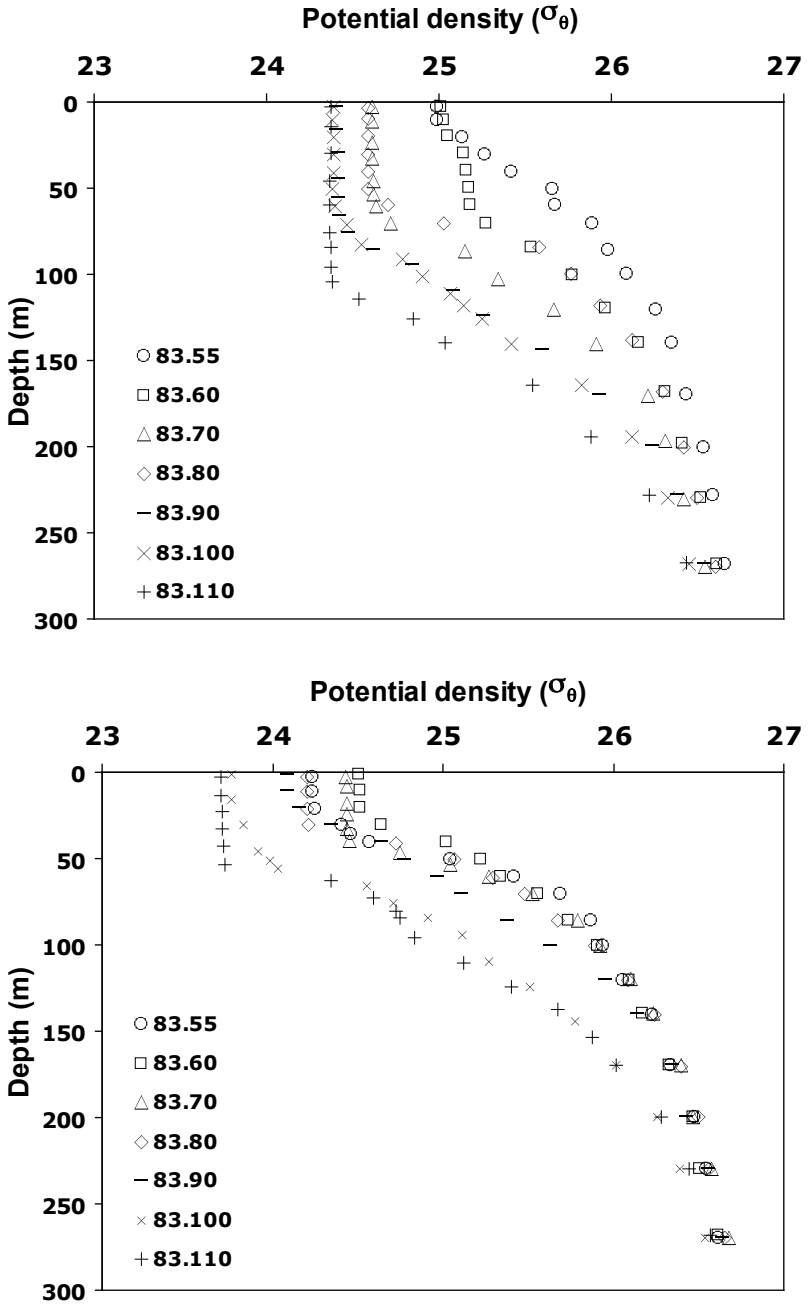

Figure 3.10 Potential density (σ_θ) with depth for cruises 0404 (top) and 0411 (bottom).

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4 CHEMICAL COMPOSITION AND RESIDENCE TIME RELATIONSHIPS IN MARINE DISSOLVED ORGANIC MATTER FRACTIONS

4.1 INTRODUCTION

The marine dissolved organic carbon (DOC) reservoir represents ~650 Gt of carbon (Hedges, 1992) and has an average radiocarbon age of ~6000 years in the deep ocean (Williams and Druffel, 1987; Druffel et al., 1992). If a small fraction of carbon that is fixed during photosynthesis each year escapes degradation and slowly accumulates within the DOC reservoir, an age continuum of between modern ($\Delta^{14}C$ >-50‰; Williams and Druffel, 1987) and >6000 years (refractory; $\Delta^{14}C$ < -550‰; Bauer et al., 1998) would be expected. Yet, there is no evidence to support a continuum of $\Delta^{14}C$ values in DOC and mounting evidence now supports the existence of only two major DOC ^{14}C-age components (Williams and Druffel, 1987; Mortazavi and Chanton, 2004; Repeta and Aluwihare, 2006; Chapter 3). Therefore, processes that contribute to the presence of refractory DOC in marine environments remain to be determined. To identify mechanisms by which refractory marine DOC is formed, the refractory component must first be isolated from bulk DOC for radiocarbon ($\Delta^{14}C$) measurements.

Two primary methods have been used to isolate marine DOC. Solid phase extraction (SPE) was used in preliminary studies to isolate DOC through hydrophobic interactions from acidified seawater; this fraction was operationally defined as marine humic substances (Stuermer and Harvey, 1974; Gagosian and Stuermer, 1977). More recently, researchers have focused on ultrafiltered DOC (UDOC) which is retained by a

1 nm filter (nominally >1000 Da). This technique separates by size and not composition; so isolated compounds are expected to be more representative of total DOC (Benner, 2002).

$\Delta^{14}C$ measurements established SPE fractions as refractory components of marine DOC because they were depleted in ^{14}C relative to bulk DOC (Druffel et al., 1992). Spectroscopic and molecular-level characterization of this SPE DOC identified the presence of amino acids, carbohydrates, and lipids, and stable carbon isotope ($\delta^{13}C$) measurements revealed its primarily marine origin (Gagosian and Stuermer, 1977; Meyers-Schulte and Hedges, 1986; Engbrodt and Kattner, 2005). In addition, low C:N values distinguished this marine fraction from DOM isolated in terrestrial and riverine environments using similar SPE techniques (Gagosian and Stuermer, 1977; Hedges et al., 1992). The overall composition of marine humic substances has been likened to humic substances isolated from terrestrial environments, which are thought to exist as a complex, condensed structure. For example, Engbrodt and Kattner (2005) suggested that carbohydrates in neutral SPE DOC were recalcitrant and associated with refractory non-polar compounds.

In contrast, chemical characterization (Benner et al., 1992; McCarthy et al., 1996; Aluwihare et al., 1997), bacterial uptake experiments (Amon and Benner, 1994), and $\Delta^{14}C$ measurements (Aluwihare et al., 2002; Loh et al., 2004; Repeta and Aluwihare, 2006) support the hypothesis that UDOC is rapidly recycled in the surface ocean. In fact, these results have led several investigators to conclude that a size-reactivity continuum model adequately explains the $\Delta^{14}C$ distribution in DOC (Benner, 2002; Loh et al., 2004). $\Delta^{14}C$ measurements have suggested that carbohydrates (Repeta and

Aluwihare, 2006), amino-acid-like fractions (Loh et al., 2004) and proteins (Meador et al., 2007) in particular, constitute a labile component of UDOC in the surface ocean. In some cases, $\Delta^{14}C$ signatures of these biochemical classes resembled that of dissolved inorganic carbon (DIC) in the surface ocean indicating their recent biosynthesis (Repeta and Aluwihare, 2006; Meador et al., 2007). However, ^{14}C-depleted compounds have also been isolated from UDOC (Loh et al., 2004; Repeta and Aluwihare, 2006). For example, ^{14}C- depleted free lipids were extracted from surface UDOC (Loh et al., 2004), but considering that free lipids are generally not large enough to be retained in UDOC and the absence of further chemical characterization has questioned the existence of refractory free lipids within UDOC (McNichol and Aluwihare, 2007). A recent study comparing surface and deep UDOC deduced a hypothetical structure for refractory DOC that was dominated by carboxy-rich aliphatic macromolecules (CRAM; Hertkorn et al., 2006). Refractory DOM dominates the deep ocean DOC reservoir, and therefore, this potential dominance of lipids (as CRAM) in refractory DOC is consistent with the observed enrichment of carbon (relative to N and P) in DOC with depth in the ocean (Hopkinson and Vallino, 2005). However, CRAM substances have yet to be isolated from UDOC, and so their existence and residence time remains in question.

As discussed previously, similar chemical components have been identified in both SPE DOC and UDOC. For example, SPE DOC and UDOC were shown to contain a similar suite and relative distribution of neutral monosaccharides (e.g., McCarthy et al., 1996; Engbrodt and Kattner, 2005). However, $\Delta^{14}C$ measurements have shown bulk SPE fractions to be depleted in ^{14}C (e.g. Druffel et al., 1992) while carbohydrates isolated from UDOC are of contemporary origin (e.g., Repeta and Aluwihare, 2006).

There is no *a priori* reason to believe that sugars present in both fractions share the same source or residence time in marine environments, yet the refractory nature of carbohydrates present in SPE DOC had not been definitively established.

Rarely has SPE-DOC and UDOC been isolated from the same environment. Previous work in surface waters of the eastern North Pacific showed that SPE-DOC and UDOC in this environment shared some similar chemical characteristics including functional groups characteristic of carbohydrates and lipids (Chapter 3). These studies also indicated a positive relationship between DOC ^{14}C-age and the relative abundance of functional groups corresponding to lipids (Chapter 3) consistent with the hypothesis that refractory DOC is comprised of lipids (e.g., Hertkorn et al., 2006). In fact, these early results suggested that the presence of polar functional groups in SPE DOC, such as those corresponding to carbohydrates, caused DOC to be enriched in ^{14}C, and that carbohydrates contained in SPE DOC may also be of contemporary origin (similar to carbohydrates in UDOC). As a result of these studies it was suggested that the 2-component ^{14}C-age model proposed by other investigators was able to adequately reproduce the age distribution within DOC because DOC was dominated by two chemically distinct fractions that had distinct residence times in the marine environment (Chapter 3). Based on this model ^{14}C-depleted SPE DOC isolated by Druffel et al. (1992) must have been enriched in lipids relative to more modern fractions of DOC. In the current study Δ^{14}C signatures of carbohydrates and lipids isolated from SPE-DOC and UDOC were examined the to test the hypothesis that the ^{14}C distribution in DOC fractions of the eastern North Pacific is controlled by the relative abundance of modern carbohydrates and refractory lipids.

4.2 METHODS

4.2.1 DOC isolation

Seawater SPE and UDOC samples were collected at multiple locations within the region surveyed by quarterly California Cooperative Oceanic Fisheries Investigation (CalCOFI) cruises aboard the R/V *Roger Revelle* in fall 2004 and 2006 (Figure 4.1). Surface seawater was collected using a diaphragm pump fitted with Bev-A-Line (Cole Parmer) tubing. Seawater was pre-filtered using both a 0.5 μm and 0.2 μm polyethersulfone cartridge filter (Millipore) and stored in 200 L HDPE barrels lined with an acid cleaned PTFE bag. To isolate SPE DOC (November 2004) a peristaltic pump fitted with acid cleaned silicon tubing was used to pass the pre-filtered seawater (2000 L total) at a rate of ~5 L hr^{-1} through a combusted glass column (300mm by 25mm, Ace Glass) containing approximately 200 mL of HP-20 (Mitsubishi) resin retained with a Teflon mesh. After each 400-600 L batch of seawater was extracted, the column was washed with 2 column volumes (500 mL) of water before the DOC fraction of interest was eluted in 50% aqueous methanol. The isolated DOC fraction is referred to as SPE DOC throughout the text, and is similar to the hydrophobic neutral fraction isolated by Engbrodt and Kattner (2005), which is discussed in more detail below.

During October 2006, approximately 1600 L of surface seawater, pre-filtered as above, was concentrated through ultrafiltration (UF) using a membrane with a nominal pore size of 1 nm to isolate ultrafiltered dissolved organic carbon (UDOC). This sample represented of composite of several samples (200 L each) collected from a variety of

sites within the CalCOFI grid. The UF system, designed in collaboration with Separation Engineering Inc. (Escondido, CA), was fitted with a polysulfone, spiral-wound filter; and the 1 nm cutoff is expected to retain compounds with a nominal molecular weight > 1,000 Daltons. After concentration UDOC was desalted by rinsing with Milli-Q water to a final volume of ~4 L (Meador et al. 2007; Chapter 3) and stored frozen at -20 ºC until further processing. In the laboratory, frozen, concentrated UDOC samples were lyophilized under vacuum (using an oil-free vacuum pump) to a dry powder for subsequent analyses

During May 2006, dissolved inorganic carbon (DIC) samples were collected aboard the *R/V Knorr* during the California Coastal Ecosystem (CCE) Long Term Ecological Research (LTER) program's process cruise. DIC samples were collected in 500 mL ground glass stoppered quartz flasks, poisoned with 100 μL $HgCl_2$, sealed, and sent to the National Ocean Sciences Accelerator Mass Spectrometry (NOSAMS) facility for AMS analysis.

4.2.2 Sugar quantification

Sugars were quantified through their alditol acetates on an Agilent 6890 Gas Chromatograph (GC; Aluwihare et al., 2002). SPE DOC or UDOC samples (2-10 mg) were spiked with 25 μg of myo-inositol as an internal standard and hydrolyzed in 4 M trifluoroacetic acid (TFA; 1-2 mL) at 121^{o}C for 2 hours. Hydrolyzed monosaccharides were then reduced in a solution of 1M ammonium hydroxide containing 10 mg/mL sodium borohydride for 2 hours. This reaction was quenched with acetic acid and dried under nitrogen (N_2). The dry alditols were then acetylated with acetic anhydride (150

uL) and 1-Methylimidazole (30 μL) for 30 min. Samples were quenched with water and the aqueous solution was extracted with methylene chloride (2 x 500 μL), dried under a stream of nitrogen and resuspended in methanol for injection onto a SP2330 (Supelco) GC column. Monosaccharides were identified based on comparison to retention times of known standards as detected by FID. The temperature program began at 180^{o}C (1 min) then increased at 4^{o}C min^{-1} to 240^{o}C and held at that temperature for 5 min, increased at 10^{o}C min^{-1} and held at the final temperature of 250^{o}C for 5 min.

4.2.3 Compound specific isotope analysis

Sugar isolation for radiocarbon measurements was conducted via high performance liquid chromatography (HPLC). DOC (15-50 mg) samples were hydrolyzed as above but not spiked with a standard. The hydrosylate was dried under a stream of N_2, resuspended in 1 mL of Milli-Q water and then extracted with ethyl acetate (2x 2mL) to isolate the hydrolyzed lipid extract (HLE) fraction. These HLE fractions were dried under N_2 and characterized by ^{1}H NMR before preparing them for radiocarbon analysis. The aqueous solution was freeze dried, resuspended at 50 mg/mL of Mill-Q water, and filtered before HPLC injections. Typical HPLC injections were 25-50 μL onto a Supelco Pb^{2+} column (78mm x 300mm) fitted with a guard column (50mm x 46 mm). The column was eluted at a flow rate of 1.5 mL/min at 80^{o}C and peaks of interest were detected using a refractive index detector. Initially, the seven neutral sugars were collected as one fraction then freeze dried and re-injected to yield three fractions. The fractions consisted of – S1: glucose, S2: xylose, rhamnose and galactose,

S3: fucose, arabinose, and mannose (Figure 4.1). The presence and purity of sugars was confirmed by ^{1}H-NMR spectroscopy before preparing the fractions for ^{14}C analysis. The amount and ^{14}C-content of the process blank was established using 1 L of Milli-Q water that was passed through the HPLC system.

Due to the salt content of samples, the HPLC column was gradually degraded. Therefore, the column was regenerated between samples by passing 100 mL of 0.1% $PbNO_3$ at 0.1 mL/min through the column overnight. Following regeneration the column was immediately flushed with Milli-Q water and a standard sugar mixture was injected to confirm the retention times of the standard monosaccharides.

4.3 RESULTS

The spatial area in the eastern North Pacific Ocean over which DOC samples were collected encompasses diverse physical and biological regimes. Several biogeographic provinces have been delineated in the region based on the diverse hydrography (Hayward and Venrick, 1998), and the area is not strongly affected by terrestrial inputs. Therefore, the spatially-integrated samples examined in this study are expected to be representative of marine DOC.

4.3.1 HPLC process blank and isolations

Approximately 1 L of Milli-Q water was run through the Pb^{2+} column and used to establish the HPLC process blank. After freeze drying, the blank was sealed in a pre-combusted (850ºC) quartz tube containing pre-combusted CuO, evacuated under

vacuum, sealed and combusted at 850ºC. The CO_2 produced during the combustion was quantified manometrically and found to yield 74 μg C. The $\Delta^{14}C$ value of the blank carbon was -164‰, which is depleted relative to the collected sugar fractions. Based on the total volume of Milli Q water collected during each HPLC sugar isolation (<20 mL) the contribution of carbon from the process blank represented <2% of the total sample. In addition, no consistent relationship between sugar fractions and isotope signatures was observed, indicating that sample impurities (i.e., compounds other than carbohydrates) did not alter carbohydrate isotope values.

Individual sugars have been previously isolated from UDOC using HPLC, and the extensive isolation scheme results in very low individual monosaccharide recoveries (Aluwihare et al., 2002; Repeta and Aluwihare, 2006). The isolation scheme implemented here was similar to that used in these previous studies but to avoid poor sample recoveries bulk sugars fractions were not re-injected onto a second HPLC column (amino) for the isolation of individual monosaccharides.

In the current method standard sugars were first injected onto the Pb^{2+} column to determine the order and quality of chromatographic separation (Figure 4.2). The first noticeable feature is the co-elution of galactose and rhamnose as well as arabinose and mannose. Although it was not possible to achieve absolute baseline separation, three major fractions could be collected with little contamination from surrounding peaks. In addition, the use of ^1H NMR proved beneficial in determining the purity of individual sugar fractions.

The chromatograms of UDOC and SPE DOC hydrosylates are depicted in Figures 4.3 and 4.4, respectively. During the first round of injections, a bulk sugar

fraction was collected to minimize any carryover from undesired material that initially elutes from the column (Figure 4.3a and 4.4a). Subsequent re-injections improved the chromatographic separation (Figure 4.3b and 4.4b) to allow the isolation of three, clean sugar fractions. ^{1}H NMR spectra confirmed that the three fractions contained the expected sugars.

4.3.2 Sugar quantification

In order to evaluate the mole fraction contributions of sugars and calculate HPLC recoveries, sugars were quantified through GC analysis of the alditol acetates. The relative carbon recoveries of total hydrolyzed neutral sugars (THNS) and HLE are reported in Table 4.1. THNS comprised 14% of UDOC consistent with other studies in marine environments (Benner et al., 1992; McCarthy et al., 1996; Aluwihare et al., 1997). The SPE sample contained slightly lower concentrations (9%) of THNS, similar to yields from Engbrodt and Kattner (2005). As expected SPE DOC samples contained relatively more hydrophobic DOC compared to UDOC, which was confirmed by NMR data. SPE DOC and UDOC yielded 29% and 6% of HL, respectively. Since UDOC is primarily composed of polar material, it is expected to have low quantities of lipids; thus the difference in HLE abundances between SPE DOC and UDOC are consistent. ^{1}H NMR spectra of HLE fractions confirmed the dominance of lipid–related NMR resonances and identified small amounts of other polar material.

4.3.3 Isotope data

Three DIC profiles were collected from the Southern California Bight (SCB) in

April 2006 (Figure 4.5). The average Δ^{14}C-DIC signature of surface waters at this time was 25‰ (±9‰; n=3). DIC data agree well with suspended POC (Chapter 3) which represents living cells and should bear local, surface ocean Δ^{14}C-DIC signatures. At sites just to the north of our sampling region Bauer et al. (1998) measured surface Δ^{14}C-DIC with a similar range of values (52±13‰).

Isotope measurements were performed on bulk SPE DOC and UDOC prior to acid hydrolysis. SPE DOC and UDOC possessed marine δ^{13}C values and were depleted in ^{14}C relative to DIC (Figure 4.5; Chapter 3). Both bulk fractions were enriched in Δ^{14}C relative to total DOC isolated at a nearby site, Station M, in 2004 (-303‰, Figure 4.4; Beaupre et al., 2007). The δ^{13}C and Δ^{14}C value of SPE DOC was indistinguishable from an average of 10 individual samples collected at the same time (-23.1‰ and -105‰ for δ^{13}C and Δ^{14}C, respectively, Chapter 3). UDOC was slightly enriched in both isotopes (δ^{13}C and Δ^{14}C values of -22.2‰ and -96‰, respectively) compared with SPE DOC. UDOC data were comparable to data collected in 2003 for UDOC in the same region and in agreement with previous reports (see Chapter 3).

Isotope data for sugars and HLE are reported in Table 4.1. HLE from UDOC was markedly depleted in ^{13}C and ^{14}C with values of -24.5‰ and -314‰, respectively. Very depleted isotope values were previously reported for free lipids extracted from marine UDOC (Loh et al., 2004). These authors found that Δ^{14}C values of free lipids in surface samples were -637‰ and -551‰ in the Atlantic and Pacific, respectively. δ^{13}C values for free lipids observed in these samples were ~-28‰. However, free lipids only accounted for 0.1-0.3% of UDOC extracted using a modified Bligh-Dyer method (Loh et

al., 2004); in the current study HLE comprised 6% of the bulk UDOC. For comparison, saponification and subsequent extraction recovered <2% of UDOC as lipids (Aluwihare et al., 2002). Therefore, the acid hydrolyzed UDOC sample analyzed here was enriched in solvent extractable lipids compared to unhydrolyzed UDOC. Although both HLE fractions had similar $\delta^{13}C$ values (-24.5‰ and -24.9‰) the $\Delta^{14}C$ value of SPE HLE (-189‰) did not have as depleted a $\Delta^{14}C$ value as UDOC HLE.

SPE sugar fractions originated from a marine source ($\delta^{13}C$ = -21.6‰ to -22.1‰) and were freshly produced (i.e. since ~1960; Druffel et al., 1992) with $\Delta^{14}C$ values ranging from -16‰ (S2) to 48‰ (S1). Sugars from UDOC had similar isotope values ranging from -7‰ to 42‰ and -21.0‰ to -21.7‰ for $\Delta^{14}C$ and $\delta^{13}C$, respectively.

4.4 DISCUSSION

4.4.1 Isotope values

Following isolation, each sample was processed according to the scheme shown in either Figure 4.6 (UDOC) or Figure 4.7 (SPE); a series of solvent extraction, hydrolysis and purification steps aimed at examining the composition and $\Delta^{14}C$ signature of hydrophilic and hydrophobic compounds were included in each scheme.

Consistent with the expectation that SPE isolates more hydrophobic material than UDOC a greater percentage of ^{1}H NMR resonances were observed in the 0-3 ppm range for the SPE sample (Figures 4.6 and 4.7) and the $\delta^{13}C$ signature of this fraction was slightly depleted (Deniro and Epstein, 1977; Table 4.1). The bulk UDOC sample, which was dominated by oxygen-rich ^{1}H NMR resonances, had a $\delta^{13}C$ isotope signature that is

typical of bulk marine particulate organic matter in this region (-22±1.9‰; Chapter 3).

The composition of free lipids in each sample was examined by dissolving the dry sample in methanol prior to hydrolysis. Free lipids in the SPE sample, where enough C was recovered for isotope measurements, had a $\Delta^{14}C$ signature that was indistinguishable from the HLE fraction (-204‰ versus -190‰). Water soluble fractions of both UDOC and SPE were then hydrolyzed with acid and partitioned again between organic and aqueous phases (Figure 4.6 and 4.7). Although not compositionally identical, both HLE fractions represent aliphatic compounds that were previously insoluble in an organic solvent due to acid labile associations with hydrophilic compounds. Both HLE fractions were depleted in ^{13}C relative to the $\delta^{13}C$ signature of bulk UDOC, which is consistent with the observed methyl and methylene-rich NMR spectra. However, HLE isolated from UDOC is more depleted in ^{14}C than SPE HLE; $\Delta^{14}C$ values were -313‰ and -190‰ for UDOC HLE and SPE HLE, respectively. Note that the $\Delta^{14}C$ signature of the UDOC-HLE fraction is similar to, or even more depleted than the $\Delta^{14}C$ signature of total DOC (Figure 4.5).

Sugars were present in the hydrolyzed, water soluble fraction of both UDOC and SPE samples and were always in enriched in ^{14}C relative to all other fractions examined in this study. $\Delta^{14}C$ values suggested that sugars in both fractions were reactive on timescales <40 years and spanned the expected range of values for $\Delta^{14}C$-DIC in surface waters of the eastern North Pacific Ocean (Figure 4.5; Table 4.1). The observed range in $\Delta^{14}C$ values (65‰) of sugar fractions isolated from SPE DOC and UDOC is likely a result of the diverse biological and physical processes characteristic of the sampling region (Bauer et al., 1998). In particular, upwelling of sub-surface waters can deplete

the $\Delta^{14}C$ value of DIC which serves as the carbon source for contemporary sugars. Figure 4.5 shows the combined profile of $\Delta^{14}C$-DIC at 3 different stations in the region and the ^{14}C-depletion of this reservoir at depths below 200 m is apparent. In addition, upwelling also introduces DOC that is depleted in ^{14}C into surface waters. For example, studies have shown that sugars isolated from subsurface waters are depleted in ^{14}C (Aluwihare et al., 2002; Loh et al., 2004; Repeta and Aluwihare, 2006). Furthermore, enhanced primary production can elevate the local $\Delta^{14}C$ of certain carbohydrates in regions, such as the coastal ocean, that recently experienced an injection of nutrients. The range in $\Delta^{14}C$ values observed here contrasts with previous ^{14}C measurements of UDOC-sugars accumulating in the stable water column of the oligotrophic North Pacific Ocean (Repeta and Aluwihare, 2006; the range of $\Delta^{14}C$ values at these sites was only 20‰). Consistent with this observation, bulk $\Delta^{14}C$-UDOC values at sites examined by Repeta and Aluwihare were also significantly enriched relative to values reported in Table 4.1 for bulk SPE and UDOC.

4.4.2 Humics and the 2-component model

Sugars in UDOC were enriched in ^{14}C as expected from previous studies (Santschi et al., 1995; Aluwihare et al., 2002; Loh et al., 2004; Repeta and Aluwihare, 2006). On the other hand, the enriched $\Delta^{14}C$ signature of SPE sugar fractions was unexpected as they were isolated from the hydrophobic, "humic" DOC fraction that had previously been identified as refractory DOC. For example, Engbrodt and Kattner (2005) suggested that sugars in SPE DOC were refractory. There has been a long standing-debate in the humic substances literature regarding the genesis of these

operationally defined compounds (E.g., Hedges, 1988). Humic substances may be produced as intact, heterogeneous biochemicals that undergo post-production modification and selective preservation to yield structures that cannot be easily recognized or traced to a particular source compound (e.g., Stuermer and Harvey, 1974; Gagosian and Stuermer, 1977; Hatcher et al., 1983). In this case, the $\Delta^{14}C$ signature of individual components that make up each humic molecule should be similar. Alternatively, biochemicals from different sources, formed at different times and modified by distinct processes, may interact to render their total structure unrecognizable. In this case, individual components would not be expected to have similar $\Delta^{14}C$ signatures. Isolated carbohydrates and HLE have distinct $\Delta^{14}C$ signatures, and so, data presented in Table 4.1 favor this second model, where a core lipid structure appears to be associated with and modified by relatively polar biochemicals. Though no direct evidence can be provided here, it is inferred from the isolation method that carbohydrates and lipids are associated. Engbrodt and Kattner (2005) made a similar observation for their neutral SPE DOC fraction isolated from the Greenland Sea. Based on their analyses, the authors concluded that the adsorption of DOC to their XAD resins was determined by the overall chemical composition of the associated molecules rather than the composition of the sugars themselves. If sugars isolated from SPE DOC in the current study were not associated with or bonded to lipids isolated as HLE, then they should not have been retained by the SPE method. Similarly, lipids present as UDOC HLE are unlikely to have been isolated without some association with larger, polar molecules.

Previous studies have inferred the presence of aliphatic compounds within

refractory marine organic matter. In fact, the SPE HLE (in particular) and UDOC HLE ^{1}H-NMR spectra share some chemical characteristics with the ^{1}H-NMR spectrum predicted for CRAM structures that are thought to accumulate as refractory DOC (Hertkorn et al., 2006). This is the first study to combine chemical characterization and Δ^{14}C measurements to demonstrate that refractory compounds in DOC are rich in aliphatic functionalities. In general, refractory DOC is expected to have a Δ^{14}C signature that is, at the very least, -550‰ (Bauer et al., 1998) and even the UDOC HLE fraction is relatively enriched in ^{14}C relative to this value. However, surface waters, especially in this region of relatively high productivity, should contain lipids from contemporary organisms. Therefore, it is likely that the SPE-HLE fraction contains both the refractory and some labile lipids; such a mixture would give rise to the relatively enriched Δ^{14}C signature of -190‰ (Table 4.1). Along with a relatively complex mixture of lipids, the presence of simple lipids in related SPE HLE fractions, including a homologous series of even-numbered, short-chain fatty acids, is consistent with the presence of labile lipids in this fraction (Chapter 5). Despite the relatively enriched Δ^{14}C values reported in Table 4.1, the current study concludes that the HLE fraction is enriched in a refractory DOC component.

Based on an interpretation of the data presented both here and in Hertkorn et al. (2006) a schematic of the possible relationship between refractory and labile DOC in the ocean is depicted in Figure 4.8. This figure suggests that despite the association between some refractory and some labile components, these two components do cycle independently, implying that microorganisms are able to eventually extract carbohydrates from the complex matrix. This “extraction” could result from either

extracellular enzymatic hydrolysis or abiotic release of the carbohydrate fraction in acidic or basic micro-environments. In combination, ^{1}H-NMR spectroscopy, molecular-level radiocarbon analyses and chemical characterization indicated that refractory and labile DOC components are best identified as lipid-rich and hydrophilic biochemicals (likely sugars), respectively.

4.5 CONCLUSION

This study has shown that sugars in surface ocean DOC, isolated by two diverse isolation methods, are turning over relatively rapidly, and this residence time (<50 years) must be similar for most carbohydrates accumulating in DOC. This finding also demonstrates that all components isolated as humic substances cannot be assumed to be refractory and associations between labile and refractory compounds within the humic substances fraction must be considered. The study further confirms that refractory compounds are concentrated in the relatively hydrophobic fraction of DOC that may, in part, stay dissolved in seawater through associations with hydrophilic compounds. These associations may also help to explain why some carbohydrates are not rapidly degraded (timescale <days) in surface waters and accumulate in surface ocean DOC. In fact, Engbrodt and Kattner (2005) used a monosaccharide-based degradation index to infer that carbohydrates isolated from SPE DOC were less “degraded” than their non-extractable counterparts and suggested that carbohydrates may be preserved due to associations with the humic substance matrix. The refractory component of DOC has not been definitively identified and characterized here but a fractionation scheme that

isolates it has been devised. A natural extension to this study is one that focuses on identifying the structure and $\Delta^{14}C$ signature of compounds within HLE fractions. The mechanism by which refractory and labile components associate is not known (although an ester bond is shown in Figure 4.8); and until refractory lipids are identified, mechanisms by which these lipids form will remain elusive.

4.6 ACKNOWLEDGEMENTS

Chapter 4, in part, will be submitted for publication with Dr. Lihini Aluwihare. The dissertation author was the primary investigator and author of this paper. We thank the CalCOFI technicians and crew of the R/V Roger Revelle for their technical assistance.

Table 4.1. Elemental and isotope data for the various DOC fractions isolated as in Figure 4.6 and 4.7. Bulk refers to a split from either the total, freeze dried solid phase extract eluted in methanol:water (as above) or total, freeze dried ultrafiltered (>1 kDa) DOC; HLE (hydrolyzed lipid extract) represents the dried ethyl acetate extract of the acid hydrosylate (4 M trifluoroacetic acid (TFA)). Sugar fractions are defined in Figure 4.1. %HLE C is the carbon recovered as HLE. % Sugars C is the carbon recovered as the monosaccharides rhamnose, fucose, arabinose, xylose, mannose, galactose and glucose by gas chromatography (GC). LC Sugars/Sugars is the % of sugars recovered as S1+ S2 +S3 relative to total sugar yields from GC. % Recovered C refers to the total carbon present in bulk SPE or UF recovered as S1+S2+S3+HLE.

	SPE DOC	UDOC
%C	42%	15%
C:N	26.5	16.4
%HLE C	29%	6%
%Sugars C	9%	14%
LC Sugars/Sugars	57%	62%
%Recovered C	34%	15%
δ^{13}C bulk	-23.1‰	-22.2‰
Δ^{14}C bulk	-105‰	-96‰
δ^{13}C HLE	-24.9‰	-24.5‰
Δ^{14}C HLE	-190‰	-313‰
δ^{13}C S1	-22.1‰	-21.7‰
Δ^{14}C S1	48‰	-7‰
δ^{13}C S2	-21.8‰	-21.0‰
Δ^{14}C S2	-16‰	23‰
δ^{13}C S3	-21.6‰	nd
Δ^{14}C S3	22‰	42‰

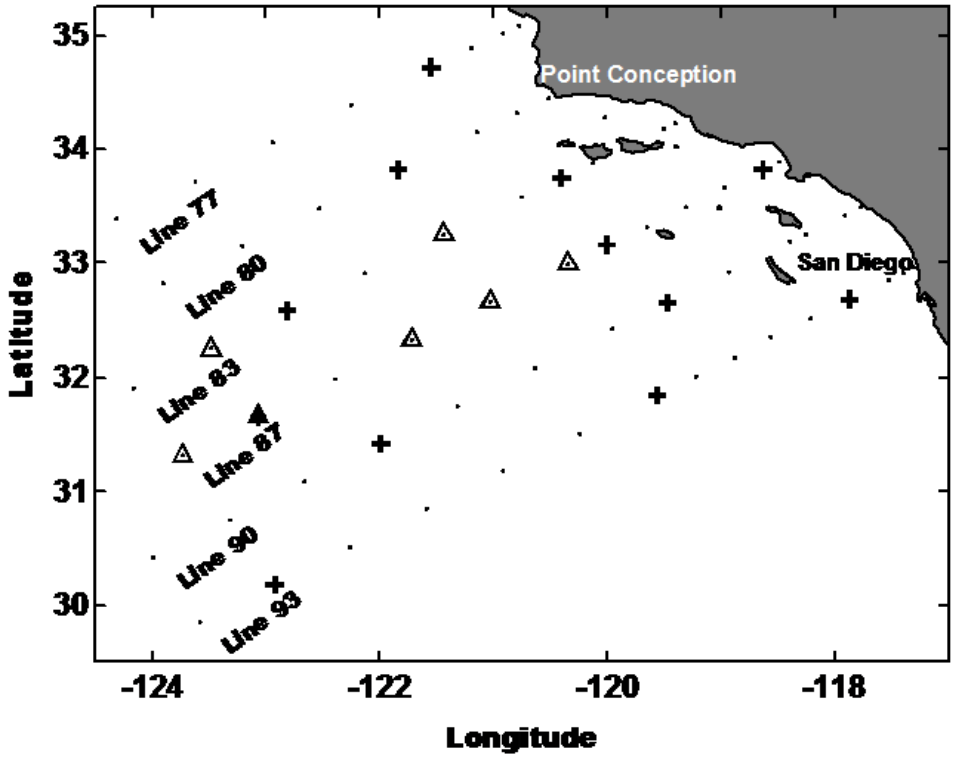

Figure 4.1 Locations of integrated sample for SPE DOC (crosses) in November 2004 and UDOC (open triangles) in October 2006.

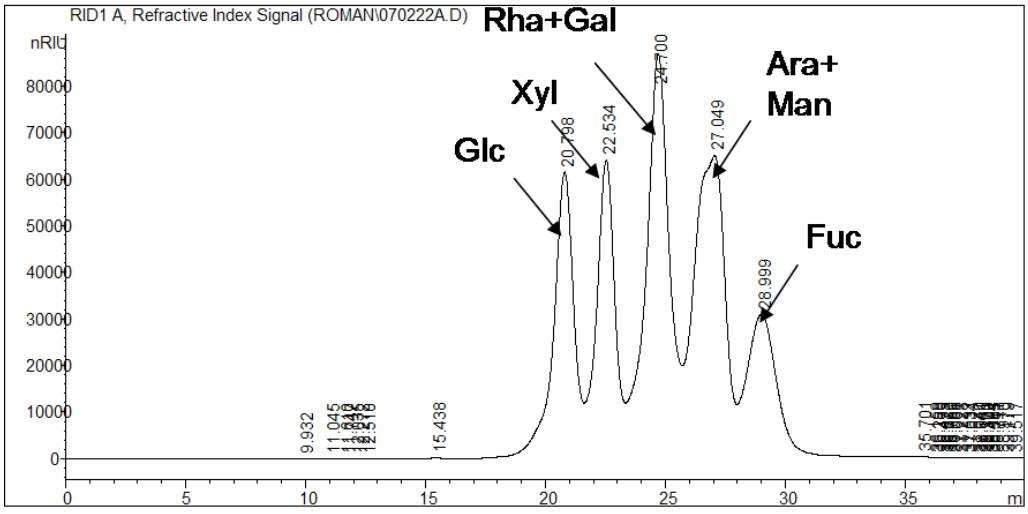

Figure 4.2 HPLC chromatogram of seven neutral sugars and order of elution on a Pb^{2+} column (Supelco). Between bulk samples, the HPLC column was regenerated with $PbNO_3$ and the retention times of the sugars were comparable to previous runs although not exactly the same.

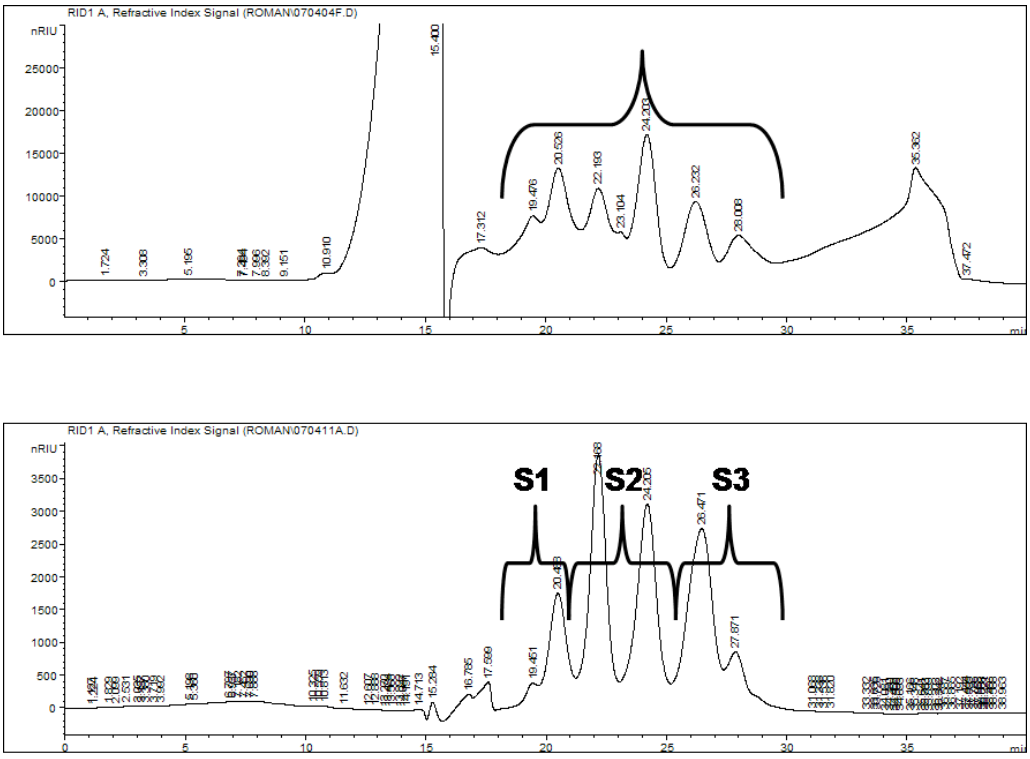

Figure 4.3 HPLC chromatogram of hydrolyzed UDOC sample. Top (a): Initially, a bulk sugar fraction was collected. Bottom (b): The fraction was subsequently reinjected then three fractions were collected separately - S1 (glucose), S2 (xylose, rhamnose and galactose) and S3 (arabinose, fucose and mannose).

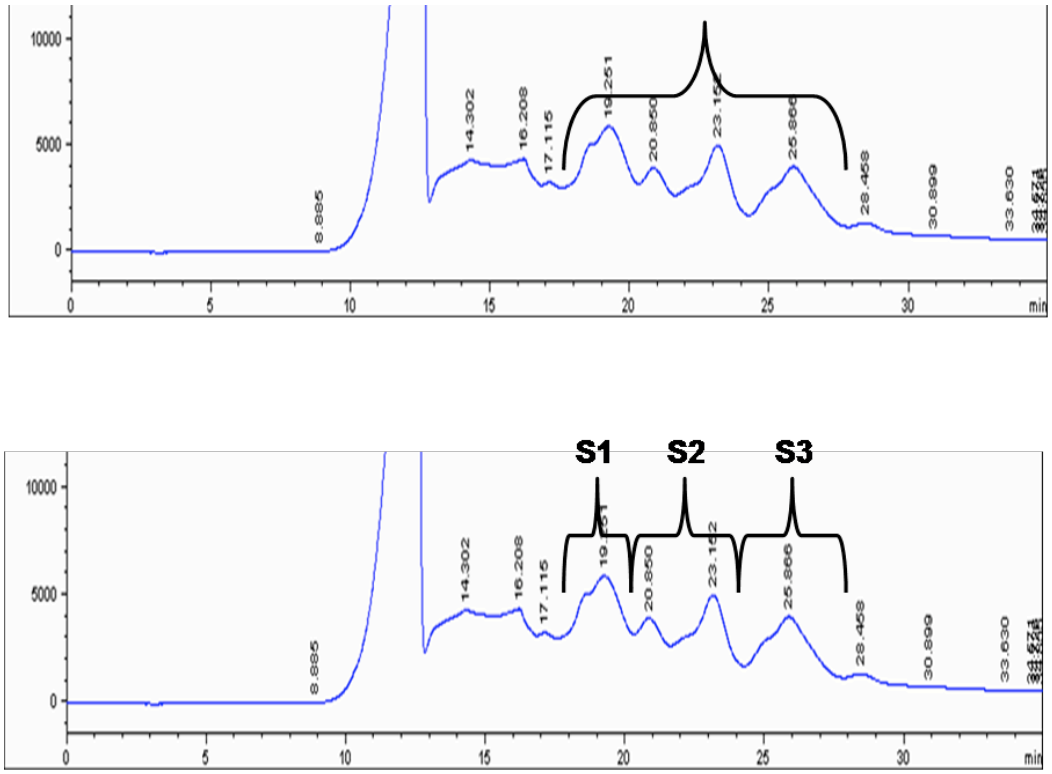

Figure 4.4 HPLC chromatogram of hydrolyzed SPE DOC sample. Top (a): Initially, a bulk sugar fraction was collected. Bottom (b): The fraction was subsequently reinjected then three fractions were collected separately - S1 (glucose + unknown), S2 (xylose, rhamnose and galactose) and S3 (arabinose, fucose and mannose).

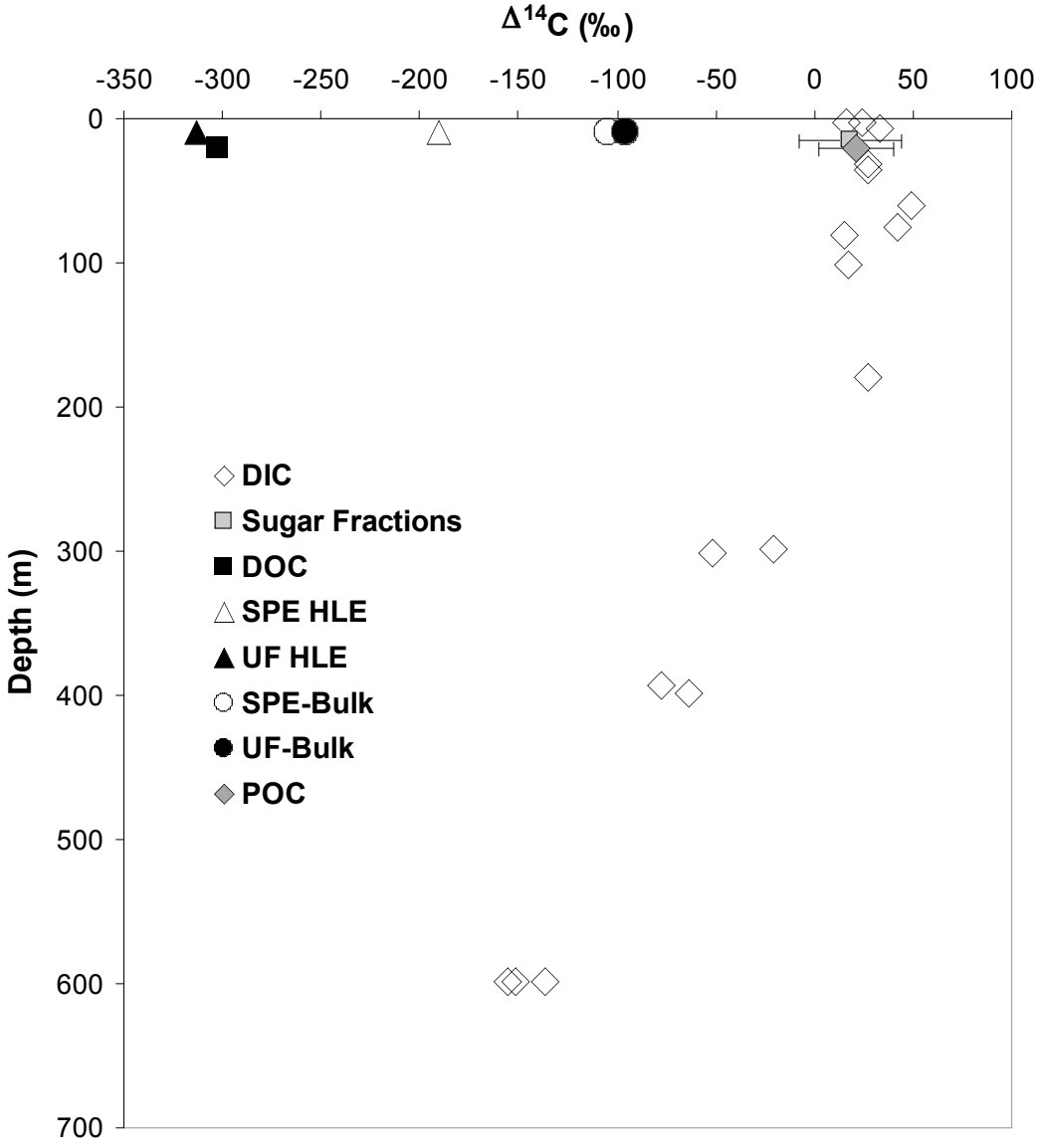

Figure 4.5 $\Delta^{14}C$ values for dissolved inorganic carbon (DIC; open diamonds) collected from three locations within the CalCOFI region in May/June 2006. A $\Delta^{14}C$ value for total DOC isolated in October/November 2004 from a nearby site (34°50'N, 123°00' W) is shown for comparison (filled square; Beaupre et al. 2007). Also shown is the $\Delta^{14}C$ range observed for combined sugars fractions in SPE and UDOC samples (stars joined by a dotted line), and individual $\Delta^{14}C$ values for SPE-HLE and UF-HLE (open and filled triangles, respectively) and Bulk SPE and UF fractions (open or filled circles, respectively). Data for fractions are taken from Table 4.1.

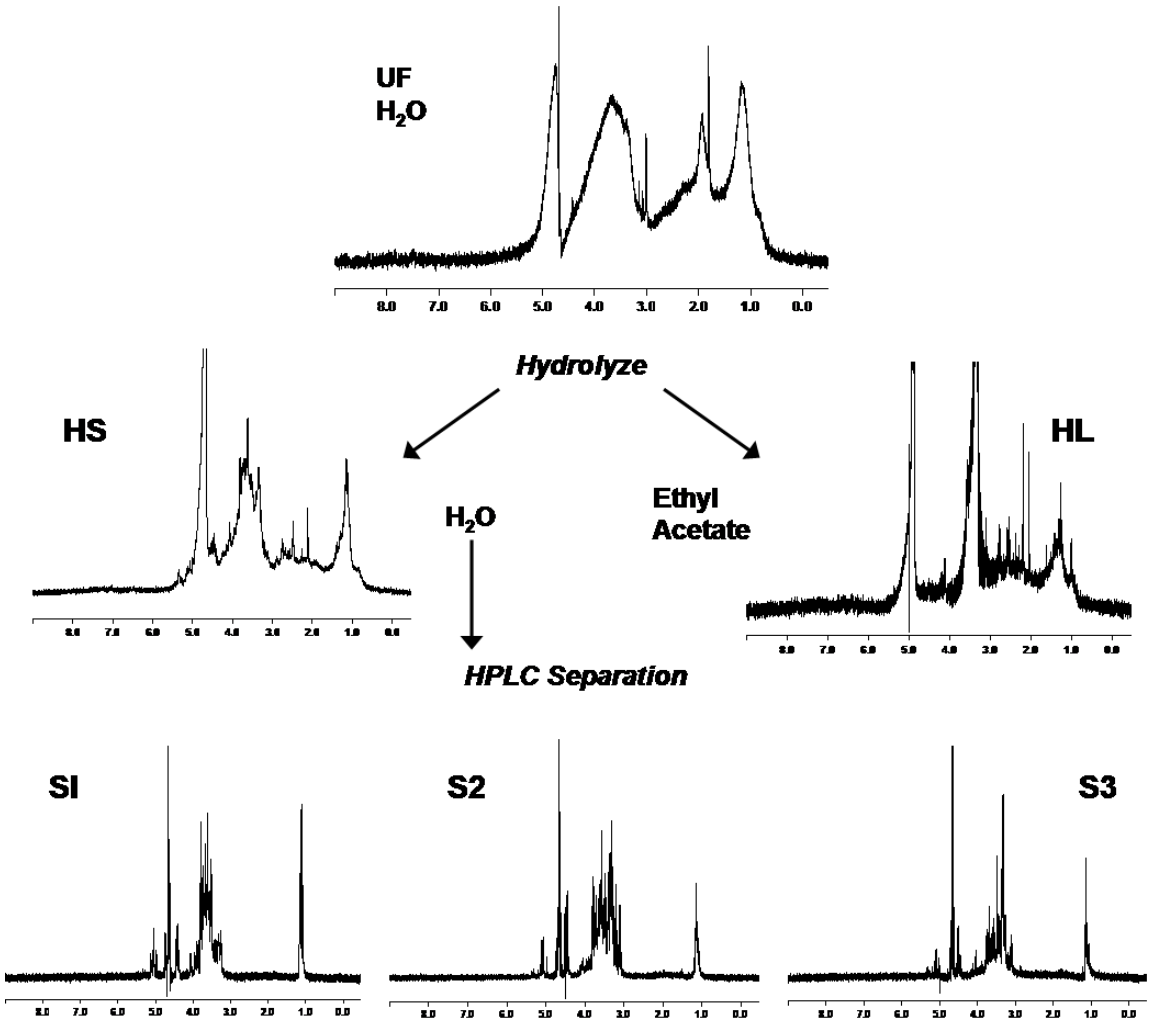

Figure 4.6 Chemical fractionation scheme for the UDOC sample collected in October 2006 (refer to Table 4.1 for abbreviations and sample details). The general composition of each fraction is determined by ^{1}H NMR spectra. S1 (glucose + unknown), S2 (xylose, rhamnose and galactose) and S3 (arabinose, fucose and mannose). All NMR scales span 0.0 to 8.0 ppm.

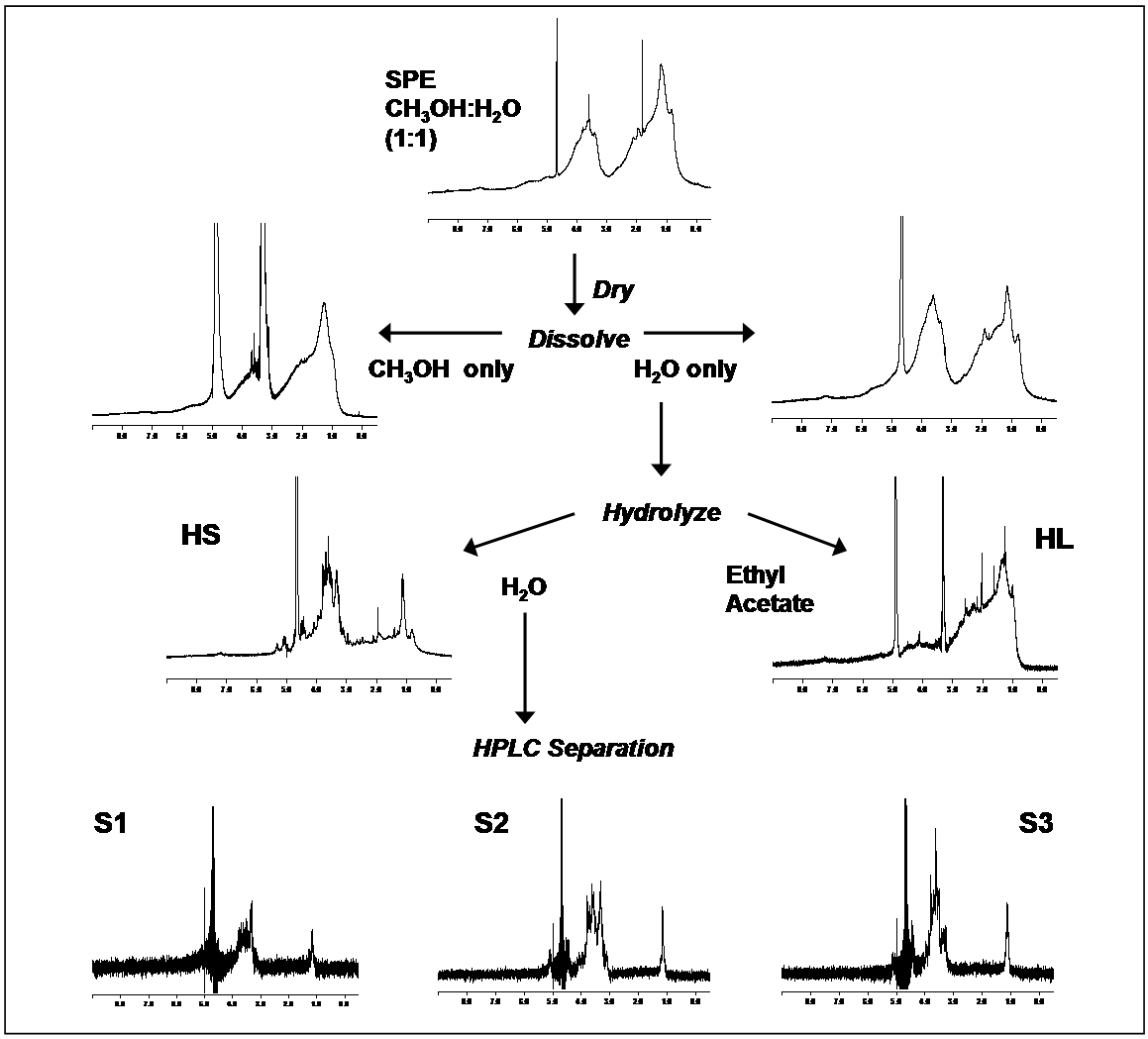

Figure 4.7 As in Figure 4.6 but for the SPE DOC sample collected in November 2004.

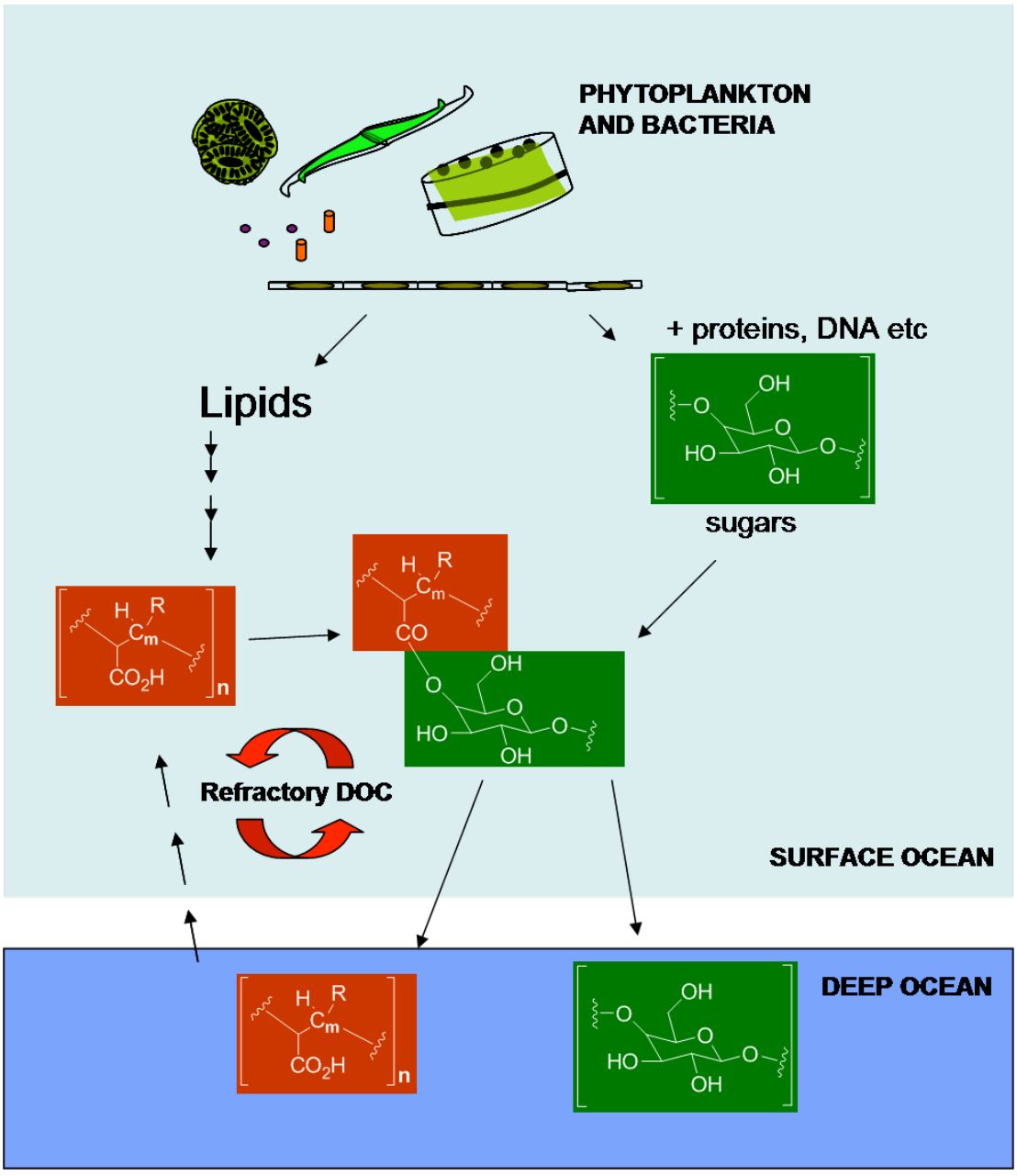

Figure 4.8 A schematic of the cycling of labile and refractory DOC that is consistent with our data. The refractory lipid structure is taken from Hertkorn et al., (2006) and is drawn to be consistent with the NMR spectrum of HLE fractions. Multiple arrows identify processes that occur on long timescales. The refractory lipid in marine environments may be autochthonous or allochthonous, and might, eventually, be removed from the ocean if the reservoir is at steady state. The association between refractory lipids and labile carbohydrates is shown here as an ester bond that may be hydrolyzed during transit through the water column.

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5 ISOTOPIC ANALYSES OF SUGARS AND LIPID MATERIAL FROM MARINE AND RIVERINE DISSOLVED ORGANIC MATTER

5.1 INTRODUCTION

The size of the marine dissolved organic carbon (DOC) reservoir (~650 Gt; Hansell, 2002) and its 6000 year residence time in the ocean requires a steady state input flux of ~0.1 Gt C/yr to maintain the current inventory. The annual terrestrial DOC flux into the ocean is 0.25 Gt C yr^{-1} (Hedges et al., 1997), and so, provides a potential source for DOC accumulating in marine environments. In fact, studies have identified the presence of terrestrial DOC in coastal environments (Meyers-Schulte and Hedges, 1986; Moran et al., 1991; Guo et al., 1997; Opsahl and Benner, 1997; Opsahl et al., 1999; Mannino and Harvey, 2000b; Benner and Opsahl, 2001; Benner et al., 2004) and lignin phenols – an unambiguous marker of terrestrial organic matter – have been identified in DOC isolated from surface waters of the open ocean. (e.g., Meyers-Schulte and Hedges, 1986). Furthermore, several studies have suggested that aged terrestrial material is exported into the ocean and may contribute to the relatively old age of marine DOC (Opsahl and Benner, 1997; Raymond and Bauer, 2001a).

Although evidence does exist to support some input of terrestrial DOC into the ocean, quantifying this flux and assessing its residence time in the marine environment has proven challenging. Existing data suggests that most terrestrial material is remineralized through bacterial degradation or photo-oxidation and only a small percentage survives in the ocean (Hedges et al., 1997; Opsahl and Benner, 1997; Benner,

2002). Based on lignin phenol concentrations, terrestrial DOC may only comprise a small percentage (<2%) of marine DOC, and most isotopic and molecular evidence indicates that the DOC accumulating in the ocean is of marine origin (e.g., Druffel et al., 1992).

Estuaries represent highly bioactive environments where terrestrial DOC transformation, removal, and addition may occur (Hedges et al., 1997, Benner, 2002). Recently, Wang et al. (2006) and Loh et al. (2006), measured $\Delta^{14}C$ and $\delta^{13}C$ values of operationally defined fractions of DOC isolated by ultrafiltration using a pore size of 1 nm (nominal molecular weight >1000 Daltons; UDOC) at sites in the eastern US that were similar and in some cases, identical, to sites sampled in the current study. In addition, several studies have also performed extensive chemical characterization of this same size fraction of DOC at similar locations (e.g. Mannino and Harvey 1999, 2000a, 2000b; Aluwihare et al., 2002; Loh et al., 2006). In combination, these studies have suggested that much of the DOC exported by rivers is replaced in regions of the estuary where salinity exceeds ~20, however, particularly during peak discharge, some terrestrial organic matter, including lipids, may be exported into the surrounding marine environment.

Previous studies in marine environments have suggested that lipid-rich hydrophobic components isolated by both ultrafiltration and solid phase extraction represent the refractory component of marine DOC (e.g., Druffel et al., 1992; Loh et al., 2004; Hertkorn et al., 2006). Studies in riverine and estuarine environments have identified ^{14}C depleted lipids in DOC isolated by ultrafiltration (e.g. Wang et al., 2006; Loh et al., 2006), but bulk riverine UDOC and XAD (a resin which preferentially

adsorbs hydrophobic, lipid-rich DOC) isolates have been shown to be relatively enriched in ^{14}C (Hedges et al., 1986; Benner et al., 2004; Mayorga et al., 2005). Raymond and Bauer (2001a) note that although ^{14}C enriched DOC is present throughout rivers, this material is labile and degraded on short timescales, therefore, DOC that is ultimately exported into the marine environment represents the fraction that escapes degradation in estuaries and the coastal ocean, and likely represents refractory terrestrial DOC.

Although previous studies engaged in either molecular level chemical characterization or fraction-specific isotope measurements, none of them combined this approach to demonstrate the chemical composition of refractory and labile terrestrial organic matter that enters the marine environment. In order to understand specific contributions that terrestrial DOC makes to marine DOC, it is important to constrain the isotopic content of specific compounds. These data would be particularly useful in determining whether terrestrial material contributes to refractory marine DOC. Terrestrial DOC is distinguishable from marine DOC by depleted $\delta^{13}C$ values, and so any terrestrial signature present in the Delaware Estuary should be identifiable by isotope analysis.

This study uses compound specific isotope analysis to determine the changes in sugar composition and residence time in the Delaware River (DR) and Estuary (DE) and Eel River Margin (ERM). The isotope data is complemented by information from the relative abundances of neutral sugars in each sample which may be used to determine the relative diagenetic state of DOC. Finally, the stable and radiocarbon signature of hydrolyzed lipid extracts are compared across samples to determine if terrestrial lipids (more specifically, hydrophobic compounds) represent a source of refractory DOC to the

coastal environment.

5.2 METHODS AND MATERIALS

5.2.1 Sample Collection Sites

Dissolved organic matter (DOM) samples were collected along the Delaware River and at a site 20 miles offshore of the Eel River mouth near Eureka, CA (USA). Exact locations are described in Table 5.1. Delaware River (DR) and Estuary (DE) samples were collected aboard the *R/V Hugh R. Sharp* in June 2006; samples were collected along the Delaware River (DR), near Trenton NJ (USA) and in the Delaware Estuary (DE). DOC at DR and DE locations has been studied extensively and the stations sampled here are comparable to sampling locations from Mannino and Harvey (1999, 2000, 2001) and Wang et al. (2004 and 2006). Eel River Margin (ERM) samples were collected aboard the *R/V Atlantis* over several days in July 2006. ERM is contained within the California Current system and shares some similarities to coastal stations in the California Cooperative Oceanic and Fisheries Investigations (CalCOFI) region sampled in Chapters 2 and 3. However, more intense wind driven upwelling is characteristic of this region (Hickey, 1998), and unlike the southern California Current, this region is influenced by terrestrial inputs from the discharge of the nearby Eel River (e.g., Blair et al. 2002). The flow of the river was low during our sampling period, but the maximum discharge rates in 2006 approached the maximum flow rate measured since 2000 (www.cdec.water.ca.gov), suggesting that significant amounts of terrestrial organic matter may have been delivered to our site during the high flow period (winter)

of 2006. In addition, the coastal upwelling index computed by NOAA PMEL at two nearby sites (39ºN, 125ºW and 42ºN 125ºW) showed that upwelling was greatest during July and August, 2006 (las.pfeg.noaa.gov). In addition, biological and chemical processes associated with seafloor methane seeps along the margin further complicate this highly dynamic region.

5.2.2 Bulk carbon sample isolation

Water was pumped using a diaphragm pump through 0.5 μm and 0.2 μm polyethersulfone cartridge pre-filters (Millipore), fitted in series, and stored in 200 L HDPE barrels lined with an acid cleaned PTFE bag. Acid cleaned Bev-A-Line (Cole Parmer) tubing was used throughout for each transfer. The DR and ERM samples were acidified to pH 2 in the barrel using HCl and then pumped with a peristaltic pump fitted with acid cleaned silicon tubing. Seawater was introduced into a combusted glass column (300mm by 25mm, Ace Glass) containing approximately 100 mL of HP-20 (Mitsubishi) resin retained with a Teflon mesh screen. The flow rate for the extraction was approximately 5 L hr^{-1}. The column was washed with 2 column volumes (400 mL) of water before the DOC fraction of interest was eluted in 50% aqueous methanol. The isolated DOC fraction is referred to as SPE DOC throughout the text.

Suspended particulate organic matter (POM) samples were vacuum filtered in triplicate through pre-combusted (450ºC) GF/F filters (Whatman) housed in acid-rinsed polysulfone filter holders. Following filtration POM samples were frozen until further processing for isotope analysis. All tubing used during the filtration was acid-washed silicone tubing.

Samples for total organic carbon (TOC) concentration determination were collected during the Delaware River cruise in combusted 40 mL glass vials fitted with acid washed, Teflon lined septa. Water samples were taken directly from Niskin bottles and acidified to pH 2 by adding trace metal-grade HCl.

Dissolved inorganic carbon (DIC) samples were collected in 500 mL ground glass stoppered quartz flasks from the National Ocean Sciences Accelerator Mass Spectrometry (NOSAMS) Facility. DIC samples were poisoned with 100 μL $HgCl_2$, sealed and sent to NOSAMS for analysis.

5.2.3 Stable isotope and radiocarbon analysis

The elemental and stable isotopic compositions of solid, dried SPE DOC, sugar fractions and hydrolyzed lipid extracts (HLE) as well as acidified POM filters were determined at the Scripps Institution of Oceanography Unified Laboratory Facility using standard elemental analyzer isotope ratio mass spectrometry (EA-IRMS) techniques (Owens and Rees, 1989). POM samples were dried overnight in an oven, placed in an acidic atmosphere for 24 hours and dried again before placing in tin cups. Elemental (carbon and nitrogen) results are reported as C:N and stable isotope composition is reported in the standard δ notation (‰) relative to a Pee Dee Belemnite sample (carbonate).

Radiocarbon samples were prepared at SIO according to the closed tube combustion method of Vogel et al., (1987). DOC samples were freeze dried or dried under N_2 in pre-combusted (@ 850°C) quartz tubes. Pre-combusted CuO and Ag were added to tubes containing each sample, evacuated and sealed under vacuum and

combusted at 850°C for 4 hours. The resulting CO_2 was purified and measured in a gas extraction line under vacuum then sealed in an evacuated glass tube. ^{14}C analyses of the resulting CO_2 were performed at NOSAMS. Data are reported according to Stuiver and Polach (1977) in $\Delta^{14}C$ (‰) relative to an oxalic acid standard (Oxalic Acid II, HoxII).

5.2.4 Sugars quantification and isolation

Sugars were quantified through their alditol acetates on an Agilent 6890 GC. SPE DOC samples (2-10 mg) were spiked with 25 μg of myo-inositol as an internal standard and hydrolyzed in 4 M trifluoroacetic acid (TFA; 1-2 mL) at 121^{o}C for 2 hours. Hydrolyzed monosaccharides were then reduced in a solution of 1M ammonium hydroxide containing 10 mg/mL sodium borohydride for 2 hours. This reaction was quenched with acetic acid and dried under nitrogen. The dry alditols were then acetylated with acetic anhydride (150 uL) in 1-Methylimidazole (30 μL) for 30 min. Samples were quenched with water and the aqueous solution was extracted with methylene chloride (2 x 500 μL), dried under a stream of nitrogen and resuspended in methanol for injection onto a SP2330 (Supelco) gas chromatography (GC) column. Monosaccharides were identified based on comparison to retention times of known standards as detected by FID. The temperature program began at 180^{o}C for 1 min then increased at 2^{o}C per min to 240^{o}C and held for 5 min.

Sugar isolation for radiocarbon measurements was conducted via high performance liquid chromatography (HPLC). DOC (15-50 mg) samples were hydrolyzed as above. The hydrosylate was dried under a stream of N_2 before

resuspending in 1 mL of Milli-Q water. The hydrosylate was then extracted with ethyl acetate (2x 2 mL) to isolate the hydrolyzed lipid fraction (HLE). These HLE fractions were dried under N_2 and characterized by ^{1}H NMR before preparing them for radiocarbon analysis as above. The aqueous solution was freeze dried, resuspended at 50 μg/μL, and filtered before HPLC injections. Typical HPLC injections were 25-50 μL onto a Supelco Pb^{2+} column (78mm x 300mm) fitted with a guard column (50mm x 46 mm) with a flow rate of 1.5 mL/min at 80^{o}C. Initially, seven neutral sugars were collected as one fraction then freeze dried and re-injected to yield three fractions. The fractions consisted of – S1: glucose, S2: xylose, rhamnose and galactose, S3: fucose, arabinose, and mannose (see Chapter 4; Figure 4.1). The presence and purity of the sugars was confirmed by ^{1}H-NMR then prepared for ^{14}C analysis.

5.2.5 DOC characterization

Fractions eluted as SPE DOC were each analyzed by ^{1}H NMR spectroscopy for crude chemical characterization. Organic fractions including bulk SPE DOC and HLE fractions were dissolved in CD_3OH, and water soluble sugar fractions were first freeze dried in D_2O to reduce exchangeable protons. After freeze drying, samples were redissolved in D_2O and transferred into a dry NMR tube using a pre-combusted Pasteur pipette. Spectra were collected on a Varian 300 MHz Inova spectrometer using standard pulse sequences. Spectra averaged over 128 scans and referenced to 4.79 ppm for D_2O or 3.31 ppm for CD_3OD.

5.3 RESULTS

Relevant hydrographic and chemical data are provided in Table 5.1. The sampling sites span freshwater (salinity = 0) to completely marine (salinity = 34) and traverse a variety of biological regimes (Table 5.1 and Mannino and Harvey, 2001). Table 5.2 reports the $\delta^{13}C$ and $\Delta^{14}C$ values for DIC, POC, bulk DOC samples, neutral sugars, and hydrolyzed lipids extracts (HLE).

5.3.1 Bulk carbon reservoirs

5.3.1.1 Delaware River (DR) and Estuary (DE)

In general $\Delta^{14}C$-DIC values of surface water samples exhibited post-bomb $\Delta^{14}C$ values (>-50‰), the one exception was in the Delaware River (DR). DR had a very depleted $\Delta^{14}C$-DIC value (-116‰) indicating that processes other than gas exchange with the overlying atmosphere was responsible for delivering DIC to this site. Contemporary atmospheric CO_2 in 2006 has a value ~40‰ (McNichol and Aluwihare, 2007) and riverine DIC should approach this value if gas exchange dominates the source of DIC. For example, Raymond et al., (2004) reported a $\Delta^{14}C$-DIC value of 43‰ at an upstream station north of Port Jervis, NY (41°22'14"N, 74°41'52"W) on the Delaware River during spring 2000, and we observed a $\Delta^{14}C$-DIC value of 46‰ at another downstream site. The remineralization of organic matter with a depleted $\Delta^{14}C$ signature could have contributed to the low $\Delta^{14}C$ value of the upstream DIC sample. Consistent with this explanation, the $\delta^{13}C$-DIC value at DR was -11‰ as compared with -7‰ at sites with more modern $\Delta^{14}C$ values (e.g. Raymond et al., 2004). A stable carbon isotope

mass balance using ~0‰ as the atmospheric CO_2 endmember and -25‰ as the $\delta^{13}C$ signature of terrestrial organic matter that may be remineralized to produce DIC at this site, requires that approximately 56% of the DIC at this site is derived from atmospheric carbon. Based on these contributions and the measured $\Delta^{14}C$-DIC values (45‰ for other Delaware River sites and -116‰ for DIC at this site) the estimated $\Delta^{14}C$ signature of remineralized POC would have to be ~-320‰. For comparison, in the Hudson River, the measured $\Delta^{14}C$-DIC value was depleted at -65‰ ($\delta^{13}C$-DIC = -11.2‰) and suspended POC at the same site had a $\Delta^{14}C$ value of -425‰ (Raymond et al., 2004). $\Delta^{14}C$ values for organic matter fractions in the Delaware River did not approach such depleted values as in the Hudson.

We were unable to determine a $\Delta^{14}C$ value for suspended POC at the DR site, but Raymond et al., (2004) report a value of 6‰ for their Port Jervis site. The $\delta^{13}C$-POC value measured during their study was -25.6‰; our DR $\delta^{13}C$-POC value was more depleted, -30.3‰, but both values are consistent with a predominantly terrestrial source for suspended POC accumulating at this site. Although the measured $\Delta^{14}C$-POC signature was more depleted than the value expected for modern terrestrial organic matter, it was much more enriched than the DR $\Delta^{14}C$-DIC value. Raymond et al. (2004) also measured the carbon isotopic content of bulk DOC at the Port Jervis site on the Delaware River. The $\Delta^{14}C$-DOC value was -114‰ and the $\delta^{13}C$ value (-27.1‰) indicated that this material had a predominantly terrestrial origin. Although this value is depleted in ^{14}C it does not approach the estimated $\Delta^{14}C$ value of ~-320‰ needed to reconcile the depleted $\delta^{13}C$ and $\Delta^{14}C$ values of DR DIC. SPE DOC at the DR site

appeared to be composed of fresh terrestrial material with $\delta^{13}C$ and $\Delta^{14}C$ values of -27.7‰ and 38‰ respectively. From the same site, Wang et al. (2006) isolated UDOC with values of -24.8‰ and -21‰ for $\delta^{13}C$ and $\Delta^{14}C$, respectively. These data are consistent with UDOC isolating predominantly polar compounds that are relatively enriched in $\delta^{13}C$ compared to hydrophobic compounds concentrated in SPE DOC (de Jesus et al. submitted). However, our SPE DOC fraction is much more enriched in ^{14}C relative to both the DOC (Raymond et al., 2004) and UDOC (Wang et al., 2006) isolated from DR, suggesting that the SPE fraction is enriched in a modern, terrestrial component. The $\Delta^{14}C$ and $\delta^{13}C$ signature of our SPE DOC sample is similar to Wang et al. (2006) operationally defined hydrolysable amino acid fraction (-27.1‰ and 34‰, respectively); however, our ^{1}H-NMR data (Figure 5.2) confirm the predominantly lipid-rich composition of our SPE DOC fraction.

It is notable that neither shales nor ancient marine carbonates are present along the Delaware River (J. Sharp., pers. comm.) but Wang et al., (2006) suggested that anthropogenic inputs from oil refineries could alter the $\Delta^{14}C$ content of carbon reservoirs in the Delaware River.

Samples for TOC concentration measurements were collected throughout the Delaware River and averages are shown in Table 5.1 (data provided by Jonathan Sharp, University of Delaware). The DR site had the lowest TOC concentration (193 μM C) measured for freshwater stations suggesting that riverine production was relatively low at this site. These data are consistent with the predominantly terrestrial $\delta^{13}C$ signature of POC and SPE DOC at DR. Addition of TOC at stations immediately downstream of DR is clearly visible in a plot of TOC concentration versus salinity (Figure 5.1). In fact, the

downstream station with the Δ^{14}C-DIC value of 45‰ had a TOC concentration of 272 μM C. If the additional TOC is from primary production in the river, then we could expect an influx of atmospheric CO_2 into the river at sites where production is greatest. DIC at these sites would be expected to retain carbon isotopic signatures associated with atmospheric CO_2. On the other hand, DIC at sites where primary production is low could be more strongly influenced by the addition of DIC from the remineralization of organic matter. For comparison, the TOC concentration at the Port Jervis site sampled by Raymond et al., (2004) was 242 μM C.

Although the salinity was 22 PSU at the DE site (approximately 67% marine water), carbon isotopic values of DIC were predominantly marine and modern (0.51‰ and 35‰ for δ^{13}C-DIC and Δ^{14}C-DIC, respectively); the similar Δ^{14}C-values for atmospheric CO_2 and surface ocean DIC makes it difficult to assign terrestrial versus marine sources to contemporary organic matter based on Δ^{14}C signatures alone, so we relied on δ^{13}C values to identify any terrestrial contributions at the DE site. Suspended POC at the DE site had δ^{13}C and Δ^{14}C values of -20.5‰ and 10.8‰, respectively; again indicating a primarily marine and contemporary source for POC. SPE DOC isolated at DE represents approximately 20% of the total DOC (TOC concentrations at this site were 161 μM C; Table 5.1).

The measured δ^{13}C value of SPE DOC at DE was -23.3‰, which is similar to SPE DOC isolated from marine sites in the eastern North Pacific (Chapter 3; no other data are available for Atlantic Ocean sites). The slight depletion in ^{13}C relative to a truly marine signature (e.g. Hedges et al., 1997) is consistent with the predominance of hydrophobic compounds in this SPE fraction (Figure 5.3). At a nearby site in the

Delaware Bay (salinity = 26), Wang et al. (2004) reported a value of -23.1‰ for the $\delta^{13}C$ signature of UDOC (at this site DOC concentrations were approximately 140 μM C). This value is more depleted than would be expected for marine UDOC (Benner, 2002) and suggests some terrestrial influence on the composition of bulk UDOC at this site. The $\delta^{13}C$ signature of Wang et al.'s operationally defined hydrolysable amino acid fraction, for example, was -20‰ at this site, and is representative of values expected for polar compounds isolated as *marine* DOC. Consistent with the marine stable carbon isotopic signature, the $\Delta^{14}C$ value for DE was depleted (-45‰) relative to DR and its source $\Delta^{14}C$-DIC (35‰). Marine SPE DOC isolated from other sites (e.g., see ERM discussion below) exhibit relatively depleted $\Delta^{14}C$ values consistent with the ubiquitous presence of a very refractory marine DOC fraction (e.g. Druffel et al., 1992; Chapter 3). These data confirm that much of the suspended POC and SPE DOC isolated at DE have an origin that is distinct from the bulk SPE DOC and POC present at DR. Comparison with Wang et al., (2004 and 2006) indicates that our SPE DOC has a cycle that is distinct from UDOC at both the DE and DR sites.

5.3.1.2 Eel River Margin (ERM)

ERM $\Delta^{14}C$-DIC value was 6‰ which is the most depleted marine surface water value that we have recorded in the contemporary eastern North Pacific. For comparison, in the southern California Current surface water $\Delta^{14}C$-DIC values ranged from 16‰ to 49‰ during May/June 2006 (Chapter 4). However, as previously mentioned, wind-driven upwelling is particularly strong near this site, and strong winds and highest upwelling rates were computed for July and August of 2006. $\Delta^{14}C$-DIC values are low

below 250 m in the southern California Current and approach values as low as -20‰ to -50‰ at 300 m (Chapter 4). Therefore, upwelling of subsurface waters is the likely source of this ^{14}C-depleted DIC. We expect that any contemporary organic matter formed at this site from marine production would record the relatively depleted ^{14}C value of DIC.

ERM particulate organic carbon (POC) isotope signatures were -26.4‰ and -34.9‰ for $\delta^{13}C$ and $\Delta^{14}C$ respectively. Although the ERM site was primarily a marine site (salinity = 33 PSU), the stable carbon isotope value indicates a terrestrial source for POC (e.g., Hedges et al. 1997). For the Eel River, Blair et al. (2003) computed a $\delta^{13}C$ signature of -26.5±1.1‰ for the terrestrial vascular plant endmember, and similar values have been measured for suspended organic matter in the river and in sediments up to 40 km offshore of the river mouth (Blair et al. 2002). However, organic matter derived from terrestrial plants are expected to have modern $\Delta^{14}C$ signatures (~40‰) as demonstrated by Blair et al. (2003); but this same study noted relatively depleted $\Delta^{14}C$ values for riverine suspended organic carbon (containing only 40% of modern C), and these depleted values may have contributed to the ^{14}C-depletion observed for suspended POC in the current study. From the depleted $\delta^{13}C$ values, it appears that terrestrial POC may also be transported offshore in the water column. Alternatively, the depleted $\delta^{13}C$ and $\Delta^{14}C$ values of POC may result from the isolation of particles derived from the incorporation of methane. Hinrichs et al., (1999) reported $\delta^{13}C$ values of -49.5‰ for methane bubbling into the water column at one of the sites sampled in the current study, and values between -57.6 to -69.1‰ have been reported for near surface hydrates in the region (Brooks et al., 1991). Methane can be expected to be depleted in ^{14}C at these sites

but no data are available for this parameter. An isotope mass balance using the Hinrichs et al. value for methane and assuming a marine, primary production endmember $\delta^{13}C$ signature of -20‰ provides an estimate of the maximum amount of C contributed to suspended POC from methane incorporation (~30%). Any fractionation associated with the incorporation of methane (as has been detected for the methanotrophic community inhabiting sediments at this site; e.g., Hinrichs et al. 1999) would lower this estimated contribution. Although we are unable to rule out a contribution from methane consumption to suspended POC accumulating in surface waters at this site, we assume throughout the rest of the chapter that the depleted carbon isotope values of POC derive primarily from the influence of terrestrial and riverine processes.

Carbon isotope values for SPE DOC isolated from the ERM were -21.8‰ and -87‰ for $\delta^{13}C$ and $\Delta^{14}C$, respectively. These values are consistent with data from other sites along the California coast (Chapter 3; although slightly enriched in $\delta^{13}C$ for an SPE sample) and identify a marine source for SPE DOC at this site. The chemical composition of this fraction as determined by ^{1}H-NMR was also typical of other marine samples (Figure 5.4; Chapter 3). The $\Delta^{14}C$ value for SPE DOC at this site is enriched relative to bulk DOC accumulating in surface waters (e.g., -302‰ at a site just south of our sampling region in 2004; Beaupre et al., 2007) and is therefore, enriched in a modern, labile DOC component.

5.3.2 Monosaccharide quantification and isotope signatures

In order to evaluate the mole fraction contributions of sugars and calculate HPLC recoveries, we quantified sugars through GC analysis of the alditol acetates. The relative

carbon recoveries of sugars are reported in Table 5.2. Isotopic compositions of the isolated compounds (sugar fractions and lipids) are reported in Table 4. Based on GC analysis, recoveries of sugars from HPLC isolations ranged from 47%-64%. The LC chromatogram was split into 3 regions based on retention times which yielded 3 fractions (S1-S3; Chapter 4, Figure 4.1) of neutral sugars: S1-Glucose (Glc); S2-Xylose (Xyl), Rhamnose (Rha), Galactose (Gal); S3-Fucose (Fuc), Arabinose (Ara), Mannose (Man). The absence of any impurities and presence of only sugars in each fraction was confirmed by NMR spectroscopy.

5.3.2.1 Delaware River and Estuary.

The abundance of sugar carbon (C) in SPE DOC was lowest in DR and DE samples, 3% and 2%, respectively. These recoveries are significantly less than that for the operationally defined total carbohydrate fraction (TCHO) recovered from UDOC at similar sites by Wang et al., (37%-38%; 2004). SPE preferentially isolates hydrophobic compounds relative to ultrafiltration therefore; the higher sugar recoveries reported by Wang et al. (2004) are not unexpected. In addition, low sugar yields (1-4%) from riverine DOC have been reported in the Amazon, Mississippi, and Arctic rivers (Hedges et al., 1994; Benner and Opsahl, 2001; Amon and Benner, 2003).

Relative sugar distributions in DR reveal that rhamnose and glucose are elevated relative to other neutral sugars at 20% and 18% respectively, followed by approximately equal contributions from the remaining neutral sugars (Table 5.3). Elevated glucose abundances in both DR and DE may reflect the presence of cellulose, which is composed of glucose monomers and is expected to be derived from terrestrial environments.

DE had higher relative abundances of fucose, glucose and mannose (19%, 19%, and 18%, respectively) while arabinose accounted for only 9% of total neutral monosaccharides. Rhamnose was more abundant than fucose in the DE sample, which reflected an inversion of the deoxy sugar distribution in the estuarine sample.

DR had the highest HPLC sugar recoveries (64% based on GC analysis) even though sugars comprised only 3% of the SPE DOC. $\delta^{13}C$ values of the recovered sugars were -24.1‰ and -25.3‰, for S2 and S3, respectively, which are enriched relative to bulk SPE DOC at this site. This enrichment in ^{13}C is expected as sugars represent polar biochemicals, while SPE DOC is dominated by hydrophobic compounds (Deniro and Epstein, 1977; van Dongen et al., 2002). In general, these stable carbon isotope data show that carbohydrates at DR are terrestrially derived. $\Delta^{14}C$ values for S2 (-15‰) and S3 (-29‰) were depleted relative to values expected for fresh, terrestrial organic matter (e.g., bulk SPE DOC had a $\Delta^{14}C$ value of 38‰ at this site). However, these values were enriched in ^{14}C relative to $\Delta^{14}C$-DIC (-116‰) at this site. An isotope mass balance, assuming a value of 45‰ for atmospheric $\Delta^{14}C$-CO_2, yields a result that is consistent with up to 60% of carbohydrates at DR being of terrestrial origin; in this calculation, the remaining carbohydrates are assumed to be derived from in-situ riverine production using DIC with a $\Delta^{14}C$ signature of -116‰. On the other hand, carbohydrates may be weathered from old soils and delivered to the DR site. With the current data set we are unable to definitively identify processes contributing to the ^{14}C-depleted signature to carbohydrates. Given that carbohydrate comprise such a small fraction of SPE DOC at this site, it is not surprising that these sugars have a unique source and/or residence time when compared with bulk SPE DOC.

At DE, $\delta^{13}C$ values for sugars ranged from -23.3‰ (S1) to -20.4 ‰ (S3). Two sugar fractions were relatively enriched in ^{13}C, S2 (21.3‰) and S3 (-20.4 ‰) also had relatively enriched $\Delta^{14}C$ values (48.1‰ and 21‰, respectively). These data are consistent with S2 and S3 fractions having a predominantly marine source at DE. However, S1 (containing glucose) had a low δ13C signature (-23.3‰), comparable to the lipid-rich bulk SPE DOC fraction, and had a depleted $\Delta^{14}C$ signature (-36.4‰) that was comparable to sugars present at the DR site. The intermediate $\delta^{13}C$ value suggests that S1 (composed of glucose) had a mixed marine and riverine origin at DE, while the other sugars fractions are completely replaced by marine carbohydrates at the DE site.

5.3.2.2 Eel River margin

SPE DOC isolated at the ERM site was composed of 20% neutral sugar C with a very high relative abundance of glucose (73%). Enriched levels of glucose in DOC have been observed at several other marine and riverine locations (Borch and Kirchman, 1997; (Amon et al., 2001; Benner and Opsahl, 2001; Ittekkot, 1982).

Since the relative abundance of glucose was so high, it was also useful to perform glucose-free calculations (Table 5.4; Cowie and Hedges, 1984). From these calculations, arabinose and xylose appeared to be depleted compared to the other sugars, 8% each. Deoxy sugars, rhamnose and fucose, were 19% and 22% respectively. Mannose was the most abundant sugar (besides glucose) comprising 26% of glucose-free sugars. The marine samples (SPE DOC and UDOC; Chapter 4) have similar distributions of neutral sugars and were depleted in rhamnose but slightly elevated in mannose (glucose-free). The results agree with previous reports of neutral sugar

compositions for marine HMW DOC sample (McCarthy et al., 1996; Aluwihare et al., 1997) which observed approximately equal contributions from seven neutral sugars. SPE DOC isolated from the Greenland Sea (Engbrodt and Kattner, 2005) was also observed to have similar distributions of neutral sugars even though geographically distant from these sample sites (ERM and SCB). The similar compositions of SCB and UDOC sugars further support the notion that the component isolated by both SPE and UDOC is similar and must sample a similar pool of total DOC.

Individual sugar fractions at ERM had $\Delta^{14}C$ signatures ranging from -42‰ (for S1, which was predominantly glucose in this case) to 6‰ (for S2), and were, on average, more depleted in ^{14}C than sugar fractions isolated from the southern California Current region (-18‰ versus 18‰ for the average $\Delta^{14}C$ signature of sugar fractions). Sugars recovered from ERM had $\delta^{13}C$ signatures ranging from -18.8‰ (for S1) to -21.3‰ (for S2). The range of isotope values observed for different sugar fractions suggest that monosaccharides may have more than a single source at this site, and in the case of $\Delta^{14}C$ values, monosaccharides may be present in polymers that cycle on different timescales. Glucose, which dominated S1, had the most unusual combination of carbon isotope signatures – relatively enriched in ^{13}C and very depleted in ^{14}C. Note that the $\Delta^{14}C$ signature of suspended POC and S1 are very similar at this site. However, POC is more depleted in ^{13}C and carries a value similar to that observed for vascular plants in this region (Blair et al., 2003).

In general, the enriched $\delta^{13}C$ signature of sugar fractions is consistent with a marine source for carbohydrates, which is similar to observations at other sites in the North Pacific Ocean (e.g. Chapter 3; Repeta and Aluwihare, 2006).

5.3.3 Hydrolyzed lipid extracts (HLE) quantification and isotope composition

In preparation for neutral sugar isolation, hydrolyzed lipid extracts (HLE) were partitioned into ethyl acetate following the 4 M TFA hydrolysis. NMR spectra of HLE fractions (Figures 5.2, 5.3 and 5.4) confirmed the lipid structure and identified minor amounts of other polar material. Preliminary mass spectrometry data (not shown) from DE confirm a complex lipid mixture including a homologous series of even numbered, short-chain fatty acids.

HLE were most abundant in DR and DE after hydrolysis, accounting for 35% and 36% of the SPE DOC (Table 5.1). For comparison, Mannino and Harvey (1999) extracted 1-2% of free lipid material from UDOC in the Delaware Estuary. These yields are not unexpected given that SPE preferentially isolates hydrophobic compounds. ERM yielded the least amount of lipids - approximately 13% of SPE DOC. The ERM yield was lower than that observed for HLE isolated from SPE DOC collected in the Southern California Current (SCC) region (~29%), but higher than HLE isolated from UDOC in the SCC (~6%).

As expected, at DR HLE was depleted in $\delta^{13}C$ (-28.0‰) relative to sugar fractions which had $\delta^{13}C$ values of -24.1‰ and -25.3‰. These differences are consistent with stable carbon isotope fraction effects associated with carbohydrate and lipid biosynthesis (Deniro and Epstein, 1977; van Dongen et al., 2002). HLE had a slightly enriched $\Delta^{14}C$ value compared to the average $\Delta^{14}C$ value of sugar fractions (-14‰ versus -22‰); however, S2 and HLE had $\Delta^{14}C$ values that were indistinguishable from each other.

At DE, HLE had a $\delta^{13}C$ signature that was consistent with lipids from a marine source (-24.8‰), and its $\Delta^{14}C$ signature was lower than that of its counterpart at DR. However, this signature is comparable to $\Delta^{14}C$ values of SPE DOC isolated from the SCC region. The distinct isotope signature of HLE at DR versus DE indicates that HLE has unique sources or compositions at each site. Consistent with this conclusion, the chemical composition of HLE fractions from DR and DE are also different (Figures 5.2 and 5.3); HLE at the DE site appear to be more enriched in methylene groups (1-1.5 ppm).

HLE isolated from ERM had a $\delta^{13}C$ signature that was consistent with a marine origin (-24‰) and more enriched than suspended POC at this site. The $\Delta^{14}C$ signature of HLE at ERM was the most depleted of any site, with a value of -323‰, showing either that lipids at this site are much more refractory than carbohydrates isolated from SPE DOC or lipids and carbohydrates at this site were derived from distinct sources.

5.4 DISCUSSION

5.4.1 Sugar distributions

5.4.1.1 Delaware River and Estuary

Carbohydrates in SPE DOC isolated from DR and DE contain relatively high amounts of glucose. Glucose is the most common monosaccharide in terrestrial environments due to the abundance of cellulose in vascular plants, therefore, the presence of glucose in riverine DOM is expected (Cowie and Hedges, 1984). Glucose from DE was depleted in ^{13}C compared to other sugar fractions at the same site, but

approached values measured for carbohydrate fractions at DR. These data suggest that unlike monosaccharides in S2 and S3, the glucose fraction at DE contained a major contribution from terrestrial sources. Consistent with this observation is the low $\Delta^{14}C$ signature of glucose at the DE site as compared with monosaccharides in fractions S2 and S3; this value at DE was similar to the $\Delta^{14}C$ signature of S2 and, in particular, S3, isolated from DR. These isotopic data show, for the first time, that glucose containing polymers of presumably terrestrial origin have a long enough residence time in riverine environments to persist into estuaries and likely, coastal surface waters. Glucose abundance does not vary significantly between DE and DR and supports the absence of selective degradation or preservation over the salinity gradient. In addition, the riverine and estuarine cycle of glucose is clearly distinguished from that of the monosaccharides which dominate HPLC fractions S2 and S3. $\Delta^{14}C$ data from DE indicate that monosaccharides in S2 and S3 are of contemporary, predominantly marine origin and were replaced on relatively short timescales. This in turn requires that riverine and terrestrial monosaccharides contained in fraction S2 and S3 do not accumulate in higher salinity environments.

The sugar distributions in DE are very similar to those found in HMW DOC from marine samples (Table 5.4; this study; McCarthy et al., 1996; Aluwihare et al., 1997) and $\delta^{13}C$ values suggest sugars are produced within the estuary. In all marine samples (Table 5.4), fucose is more abundance than rhamnose, yet the reverse is true for DR.

The constant abundances of sugars across marine HMW DOC samples suggest that a common biopolymer may be resistant to degradation and persist throughout the

ocean (McCarthy et al., 1996; Aluwihare et al., 1997). Considering that $\Delta^{14}C$ values of DE sugars reflect local $\Delta^{14}C$-DIC (35‰), it is apparent that the sugar composition may be controlled with in the estuary and probably do not represent a terrestrial recalcitrant biopolymer. The exception to this as evident by the intermediate isotope values is DE S1 which likely contains terrestrial glucose (see above).

Cowie and Hedges (1984) identified the potential of deoxy sugars to serve as a tool for distinguishing DOM derived from bacteria, which preferentially produce these monosaccharides in culture, versus phytoplankton derived sugars. Therefore deoxy sugars are expected to indicate the presence of bacterial processes or modification, especially in the presence of fresh DOC (Cowie and Hedges, 1984; Aluwihare et al., 1999). Along the salinity gradient of the Delaware River, rhamnose and fucose constitute 32-33% of total monosaccharides at both Delaware sites. Deoxy sugars may also be selectively preserved and enriched abundances may indicate a higher degree of degradation (Cowie and Hedges, 1984; Benner and Opsahl, 2001). However, $\Delta^{14}C$ values of sugars argue against the notion that this biopolymer survives degradation over decadal and millennial timescales (this study; Repeta and Aluwihare, 2006). If this indeed were the case, depleted $\Delta^{14}C$ values relative to those already observed would be expected.

5.4.1.2 Eel River Margin

Glucose comprised 74% of total neutral sugars in ERM. Highly elevated levels of glucose have been reported in DOC near the Oregon coast (48%; Borch and Kirchman, 1997), in sea-ice floes (75%; Amon et al., 2001; 55%, Engbrodt and Kattner,

2005), the Mississippi River plume (48%; Benner and Opsahl, 2001), and a phytoplankton bloom in the North Sea (Ittekkot, 1982). Although, chlorophyll data were not recorded during sample collection, it is possible that a phytoplankton bloom contributed to the enriched glucose levels. Alternatively, glucose in Eel River DOM might be preferentially preserved as observed above for the Delaware River system

However, the $\delta^{13}C$ signature of glucose isolated from SPE DOC at ERM was even more enriched than that of monosaccharides isolated in S2 and S3, indicating that a vascular plant source for this material was unlikely. In fact, all other sugar fractions examined in this study and isolated from the SCC region had more enriched $\delta^{13}C$ signatures than glucose isolated at ERM. Glucose is not typically enriched in ^{13}C relative to other monosaccharides isolated from the same organism (e.g. van Dongen et al., 2002; Teece and Fogel, 2007), though glucose and other monosaccharides are typically enriched relative to the bulk organismal carbon. Therefore, the $\delta^{13}C$ signature of glucose at ERM suggests that this monosaccharide has an origin that is distinct from that of other neutral monosaccharides. One possibility is that glucose is produced in relatively abundant quantities during marine primary production at this site. For example, the Eel River Margin is part of the California Current System which is a relatively productive location due to seasonal wind-driven upwelling along the northern California coast (Hickey, 1998). During bloom conditions in the southern California Current region, the stable carbon isotope composition of POC can reach relatively enriched values, consistent with that observed for glucose at ERM. (Bloom conditions are identified based on increases in POC and chlorophyll concentration as well as marked decreased in nitrate concentration). However, the $\Delta^{14}C$ value of S1 (glucose) in

ERM was -42‰, which is ^{14}C-depleted relative to S2, S3, and DIC at this location suggesting that contemporary marine production is not the source of this glucose. Other sources of glucose, with enriched $\delta^{13}C$ values include C4 plants (between -9 and -17‰; Boutton et al., 1991) in the surrounding Eel River estuary. For example, marsh grasses found in California wetlands such as *Spartina alterniflora*, *Spartina foliosa* and *Distichlis spicata* were all shown to have relatively enriched $\delta^{13}C$ signatures (between -13‰ and -15‰, Cloern et al., 2002; Savidge and Blair, 2004). However, Blair et al. (2003) collected several grass samples throughout the Eel River watershed and showed that these grasses had a $\delta^{13}C$ signature of -27.3 ±1.9‰. Blair et al. (2003) also showed that grass samples examined in their study had modern $\Delta^{14}C$ values. Therefore, if glucose at ERM originated from C4 production then this glucose must have a relatively long residence time in the coastal ocean to possess the observed $\Delta^{14}C$ signature. Another source of isotopically enriched glucose is the benthic algae that control riverine primary production, and which dominate during low flow periods. Blair et al., (2003) reported a $\delta^{13}C$ range between -12.9‰ and -27.5‰ for benthic algae at this site; no radiocarbon data are available for benthic riverine algae at this site. Based on stable carbon isotope data, benthic algal production could supply the unusually large amounts of glucose identified in the ERM sample. Suspended POC at ERM had a similar $\Delta^{14}C$ signature to glucose isolated from SPE DOC, but its depleted $\delta^{13}C$ signature was more compatible with a strictly terrestrial source. Unlike suspended POC, total SPE DOC had marine-like $\delta^{13}C$ signatures.

Other monosaccharides isolated from ERM had relatively depleted radiocarbon

values as well (between -17‰ and 6‰), but possessed typically marine $\delta^{13}C$ signatures. We conclude that the relatively depleted $\Delta^{14}C$ values for S2 and S3 monosaccharides, compared with sugars isolated from other sites in the eastern North Pacific (e.g., Chapter 3), are primarily a result of enhanced upwelling at the ERM site at the time of sampling. This physical process could affect the $\Delta^{14}C$ signature of organic matter in two ways. First, the upwelling of ^{14}C-depleted DIC, and subsequent primary production utilizing this DIC could result in the production of organic matter that is isotopically depleted. For example, at ERM, $\Delta^{14}C$-DIC is similar to the $\Delta^{14}C$ signature of S2. Second, upwelling along the continental slope will inject ^{14}C-depleted organic matter into surface waters (Bauer et al., 1998), thereby decreasing the average $\Delta^{14}C$ signature of DOC isolated from the surface ocean. These processes should influence the observed $\Delta^{14}C$ value of glucose and suspended POC as well. However, the observed depletion is too great, and the $\delta^{13}C$ signature is too unique to be attributed to these processes alone.

Another process that is unique to ERM and which could affect the isotope signature of organic reservoirs is the presence of methane seeps in sediments underlying the surface ocean sites sampled in this study (e.g., Hinrichs et al., 1999). If we assume that the methane released from these seeps and subsequently incorporated into marine DOM through biological consumptions is ^{14}C-dead, then only 4% of methane derived carbon needs to be incorporated to deplete the $\Delta^{14}C$ signature of glucose to -42‰.

5.4.2 Contribution of organic matter from rivers and estuaries to the coastal ocean

5.4.2.1 Sugars

The depleted $\delta^{13}C$ values for SPE DOC, sugar fractions and HLE at DR (-24.1‰ to -28.0‰) are consistent with a terrestrial source for these fractions, In addition, suspended POC had a $\delta^{13}C$ value of -30.3‰, again suggesting a predominantly terrestrial source for organic matter at DR. Despite the depleted $\Delta^{14}C$ -DIC value, bulk SPE DOC had a contemporary $\Delta^{14}C$ signature of 38‰ at DR, which is more consistent with production in terrestrial environments and transport to DR than with in situ production at DR. However, $\Delta^{14}C$-DIC values upstream (Raymond et al., 2004) and downstream of DR were 43‰ and 46‰, respectively, so, SPE DOC may have been produced at a different location along the river. Interestingly, UDOC isolated from a nearby site in the Delaware River (Wang et al., 2006) was more depleted in ^{14}C than SPE DOC isolated in the current study, suggesting that SPE DOC is either more reactive than UDOC or has a different source in the Delaware River.

Sugar fractions and HLE fractions isolated at DR were all depleted in $\delta^{13}C$ and $\Delta^{14}C$ relative to bulk SPE DOC. In fact, these DOC fractions had $\Delta^{14}C$ signatures that were similar to UDOC isolated near this site by Wang et al. (2006). In addition, measured values were enriched in ^{14}C relative to DIC at this same site indicating that in-situ production at DR could have contributed ≤40% of the C contained in the carbohydrate or HLE fractions.

Isotopic compositions of SPE DOC, POC and sugar fractions S2 and S3 at DE are all different from their counterparts at DR, suggesting that these organic carbon fractions are replaced by either riverine production or similar fractions of marine origin (e.g., stable carbon isotopic data are more consistent with a marine source). TOC concentrations in the Delaware River peak between our DR and DE sites, (Figure 5.1)

and Harvey and Mannino (2001) indicate that the turbidity maximum and the chlorophyll maximum also occur between our DR and DE sites. Figure 5.3 shows a non-conservative mixing along the salinity gradient indicating that DOC is lost as salinity increases. Non-conservative mixing has been observed previously in the Delaware Estuary and York River system as biomarkers for terrestrial DOC are selectively removed (Mannino and Harvey, 2000a; Raymond and Bauer, 2001b). Selective sorption of amino acids on to particles in the turbidity maximum was observed in the Delaware Estuary and resulted in a relative enrichment of polysaccharides (Mannino and Harvey, 2000a). Therefore it is not unexpected that DE S2, S3, and HLE have isotope signatures similar to marine samples even though bulk SPE DOC suggests the presence of terrestrial material.

Sugar fraction S1 at DE did not conform to the trends discussed above. Unfortunately, no data are available, but we assume that S1 had similar carbon isotopic characteristics to S2 and S3 at DR. What is clear is that S1 at DE had isotopic characteristics that were more similar to organic matter fractions isolated at DR than at DE (with the exception of bulk SPE DOC). Therefore, we conclude that some terrestrially derived glucose (S1) persists in the Delaware estuary and may be available for export into the coastal environments. No other organic matter fraction examined in this study shares this characteristic. Evidence of the persistence of terrestrial material in coastal and open ocean waters has certainly been demonstrated in previous studies (e.g., Meyers-Schulte and Hedges, 1986; Opsahl and Benner, 1997; Benner et al., 2005), but glucose has not been previously identified as a component that is preserved in riverine and estuarine environments. These novel results suggest that glucose may be a useful

molecular tracer of terrestrial and riverine inputs into the coastal ocean.

The isotopic data for glucose at ERM do not support a strictly terrestrial source; however, they do suggest that the cycling of glucose at this terrestrially impacted site is unique. Stable carbon isotope data for glucose at ERM may support a riverine, benthic algal source for the glucose accumulating in surface waters. However, $\Delta^{14}C$ data cannot be used to support or refute this conclusion as the $\Delta^{14}C$ distribution in various carbon reservoirs at this site is not well constrained. In addition, our current data set is too small to differentiate contributions from the various, complex processes involved in the carbon cycle at this site.

5.4.2.2 Hydrolyzed lipid extracts

HLE at DR had the most enriched ^{14}C signature of all organic fractions isolated from SPE DOC. This finding is unique to DR as HLE was consistently more depleted in ^{14}C than sugar fractions isolated from the same sample at all other sites examined in this study and in the SCC region. SPE DOC itself was more enriched in ^{14}C than HLE, and based on the isotope composition of HLE, much of the non-HLE SPE DOC (65% of the SPE DOC) must also be relatively enriched in ^{14}C (~50‰).

Isotopic compositions of HLE at DE and DR were different, showing that terrestrial/riverine lipids were relatively labile and replaced by a primarily marine endmember at the DE site. Previous studies have considered a terrestrial source for refractory DOC accumulating in marine environments (Meyers-Schulte and Hedges, 1986; Opsahl and Benner, 1997; Raymond and Bauer, 2001a), and several recent studies have suggested that hydrophobic compounds represent the refractory component of

marine organic matter (e.g., Hwang et al., 2004; Hertkorn et al., 2005; Chapter 3). If hydrophobic compounds do indeed represent refractory marine DOC, our preliminary results from the Delaware River indicate refractory, marine lipids are not derived from terrestrial sources. However, HLE represent a complex mixture of compounds, and it is possible that small quantities of a lipid component, with a uniquely depleted isotope signature and a terrestrial source, are preferentially preserved. Further compound specific lipid isotope measurements are needed to unambiguously exclude terrestrial sources of refractory marine DOC.

Also notable is the fact that DR HLE was more enriched in ^{14}C than free lipids isolated from UDOC ($\Delta^{14}C$ values were -14‰ and -946‰ (Wang et al., 2006), respectively). Wang et al. (2006) suggest that petroleum hydrocarbons from oil refineries may be the source of these isotopically-depleted lipids and/or that samples were contaminated during collection. Our DR HLE carbon isotopic data indicate that lipids were recently biosynthesized at the DR site. It is also interesting to note a small, undefined hump in the aromatic region which suggests the presence of lignin-type material in HLE (Figure 5.2).

In all cases, lipids extracted after hydrolysis of marine SPE DOC were depleted in $\Delta^{14}C$ relative to bulk DOC and sugar fractions, indicating the refractory nature of lipids in marine environments. The most depleted ^{14}C signature observed for HLE was detected at ERM (-322‰). This data is consistent with the previously cited conclusion that lipids/hydrophobic material represent the refractory component of DOC. However, HLE isolated from SCB (Chapter 3; -190‰) or DE is not as depleted in ^{14}C as HLE at ERM. At ERM, both the strong upwelling of sub-surface, isotopically depleted DOC

and/or the presence of ^{14}C-depleted lipids released from bottom hydrocarbon seeps could influence the $\Delta^{14}C$ signature of HLE. However, a similarly depleted value was observed for HLE extracted from hydrolyzed UDOC (-313‰) isolated in the SCC region (Chapter 3). Sites where UDOC was isolated are remote from hydrocarbon seeps, and so, our data favor the influence of upwelling on the $\Delta^{14}C$ signature of surface HLE at ERM.

NMR data of HLE for marine samples show small differences. However, it appears that HLE from SCB contains slightly more polar material (3-5 ppm) than ERM or UDOC. This could contribute to the enriched ^{14}C value for SCB which is not surprising from results found in Chapter 3. It is also interesting to note that, although a bit ambiguous, NMR data for HLE from UDOC resembles lipid material isolated from SPE samples. These implications have already been discussed in Chapter 4.

5.4.3 Sugars composition as indicators for diagenesis

Despite different locations and extraction techniques, the relative abundance of sugars was consistent with previous studies of marine and riverine DOC (Cowie and Hedges, 1984; Amon and Benner, 2003; Benner and Opsahl, 2001; Engbrodt and Kattner, 2005). The low yield of sugars from DR and DE suggest that the DOC accumulating at these sites was not derived from recent phytoplankton production; high yields of neutral sugar carbon have been shown to be correlated with freshly produced DOM (Benner and Opsahl, 2001; Amon and Benner, 2003). However, the validity of such an index for DOM derived from terrestrial sources has not been carefully examined, and our isotopic data suggest instead that carbohydrates our DE site were derived from recent production.

The glucose free relative abundance (Table 5.4) of rhamnose decreased from 25% to 17% from DR to DE, while fucose increased from 16 % to 23%. A similar trend was reported for a comparison between the Mississippi River and its plume in the Gulf of Mexico during a phytoplankton bloom where rhamnose decreased from 18% to 14% and fucose increased from 14% to 19% (Benner and Opsahl, 2001). These changes could result from either the production of fucose rich carbohydrates and/or the preferential degradation of rhamnose bearing carbohydrates, and these two processes cannot be distinguished on the basis of our current data set. However, these variations could be tied to shifts in the biological community that occur over the salinity gradient sampled in this study.

The change in relative sugar abundances between DR and DE corroborates the isotopic data that indicate the removal and addition of carbohydrates present in S2 and S3. Microbial transformation and diagenesis of carbohydrates in riverine and estuarine environments has been reported in several previous studies (Cowie and Hedges, 1984; Benner et al., 2001) but this study is the first to provide complementary isotopic evidence in support of these transformations. Although carbohydrates in S2 and S3 at DE are derived primarily from marine sources, we conclude that terrestrial glucose is still conserved at this sites as indicated by the depleted $\delta^{13}C$ and $\Delta^{14}C$ values.

Recently, one other study determined sugar distributions from SPE DOC from the Greenland Sea (Engbrodt and Kattner, 2005). Sugar distributions were not dissimilar from those found in HMW DOC. The previous study also used the ratio of deoxy sugars (Rha + Fuc) to pentose sugars (Ara + Xyl) as a proxy for the degree of DOC degradation. A lower ratio inferred a higher degree of microbial reworking. SCB

sample had a ratio of 0.9 and UDOC a ratio of 1.0, demonstrating that as expected, UDOC possesses a relatively fresher characteristic than SPE. ERM had the highest pentose:deoxy ratio of 2.6 which would suggest freshly produced DOC. However, as mentioned before, the utility of these degradation indices is still questionable in regions where multiple sources of carbohydrates are present.

5.5 CONCLUSION

The carbohydrate specific and HLE-specific isotope analyses clearly add a dimension to DOC studies not previously considered. In general neutral sugars retained a post-bomb ^{14}C signature. Although individual sugars were not isolated for isotope analysis, it appears that sugar fractions provide similar information to that reported by Repeta and Aluwihare (2006). In addition, SPE DOC samples provided enough material for radiocarbon measurements of sugar fractions. The relative abundance of sugars corroborates the isotope data for DR and DE samples, which has not been reported before. These data provide clear isotopic evidence that glucose is preferentially preserved along the salinity gradient in the Delaware River. This result suggests that glucose may be a useful tracer for terrestrial derived DOC.

Finally, the use of HLE isotope analysis proved to be useful in determining the origin of lipids in DE. The isotope data suggests that HLE in DE are derived from estuarine or marine production and have a depleted ^{14}C age. This suggests that the Delaware River may not be a source of pre-aged DOC to the coastal ocean.

5.6 ACKNOWLEDGEMENTS

Chapter 5, in part, will be submitted for publication with Dr. Lihini I. Aluwihare. The dissertation author was the primary investigator and author of this paper. We thank the crews of the R/V Atlantis and R/V Hugh R. Sharp for their technical assistance.

Table 5.1 Sample locations with hydrographic data and general SPE DOC elemental properties.

	DR	DE	ERM
Longitude	74.88°W	75.18°W	124.60°W
Latitude	40.08°N	39.07°N	40.79°N
Salinity (PSU)	0.02	22	35
chl a ($\mu g\ L^{-1}$)	3.2	10.2	n.d.
[DOC] (μM)	193	161	61
%C isolated	19%	20%	13%
DOC %C	46%	50%	42%
C:N	16.1	19.4	15.6

Table 5.2 $\delta^{13}C$ and $\Delta^{14}C$ values for bulk and fraction specific DOC.

	DR	DE	ERM
Bulk			
$\delta^{13}C$ DIC	-11.1	0.5	1.2
$\Delta^{14}C$ DIC	-116	35	6
$\delta^{13}C$ Bulk	-27.7	-23.3	-21.8
$\Delta^{14}C$ Bulk	38	-45	-87
$\delta^{13}C$ POC	-30.3	-20.5	-26.4
$\Delta^{14}C$ POC	nd	11	-35
Specific			
$\delta^{13}C$ HLE	-28.0	-24.8	-24.0
$\Delta^{14}C$ HLE	-14	-98	-323
$\delta^{13}C$ S1	Lost	-23.3	-18.6
$\Delta^{14}C$ S1	Lost	-36	-42
$\delta^{13}C$ S2	-24.1	-21.3	-21.3
$\Delta^{14}C$ S2	-15	48	6
$\delta^{13}C$ S3	-25.3	-20.4	-20.9
$\Delta^{14}C$ S3	-29	21	-17

Table 5.3 Relative sugar distributions in SPE DOC samples. SCB and HMW are the same samples as SPE DOC and UDOC, respectively, from Chapter 4.

	DR	DE	ERM	SCB	HMW
Rhamnose	20%	14%	5%	11%	9%
Fucose	13%	19%	6%	16%	17%
Arabinose	16%	9%	2%	16%	13%
Xylose	12%	11%	2%	15%	13%
Mannose	10%	18%	7%	15%	15%
Galactose	11%	10%	5%	17%	19%
Glucose	18%	19%	73%	12%	13%

Table 5.4 Glucose-free sugar distributions and deoxy to pentose ratios.

	DR	DE	ERM	SCB	HMW
Rhamnose	25%	17%	19%	12%	10%
Fucose	16%	23%	22%	18%	20%
Arabinose	19%	11%	8%	18%	15%
Xylose	15%	14%	8%	17%	15%
Mannose	12%	22%	26%	17%	18%
Galactose	13%	13%	18%	19%	22%
Rha + Fuc to Ara + Xyl	1.2	1.6	2.6	0.9	1.0

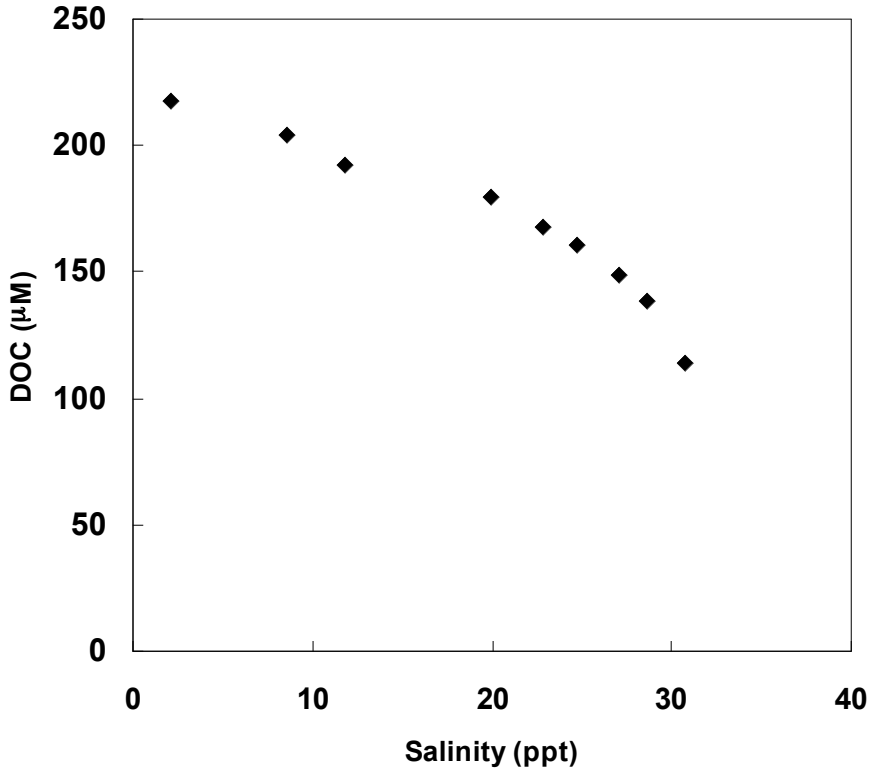

Figure 5.1 Delaware River DOC (μM) vs. salinity (PSU) displays non-conservative mixing.

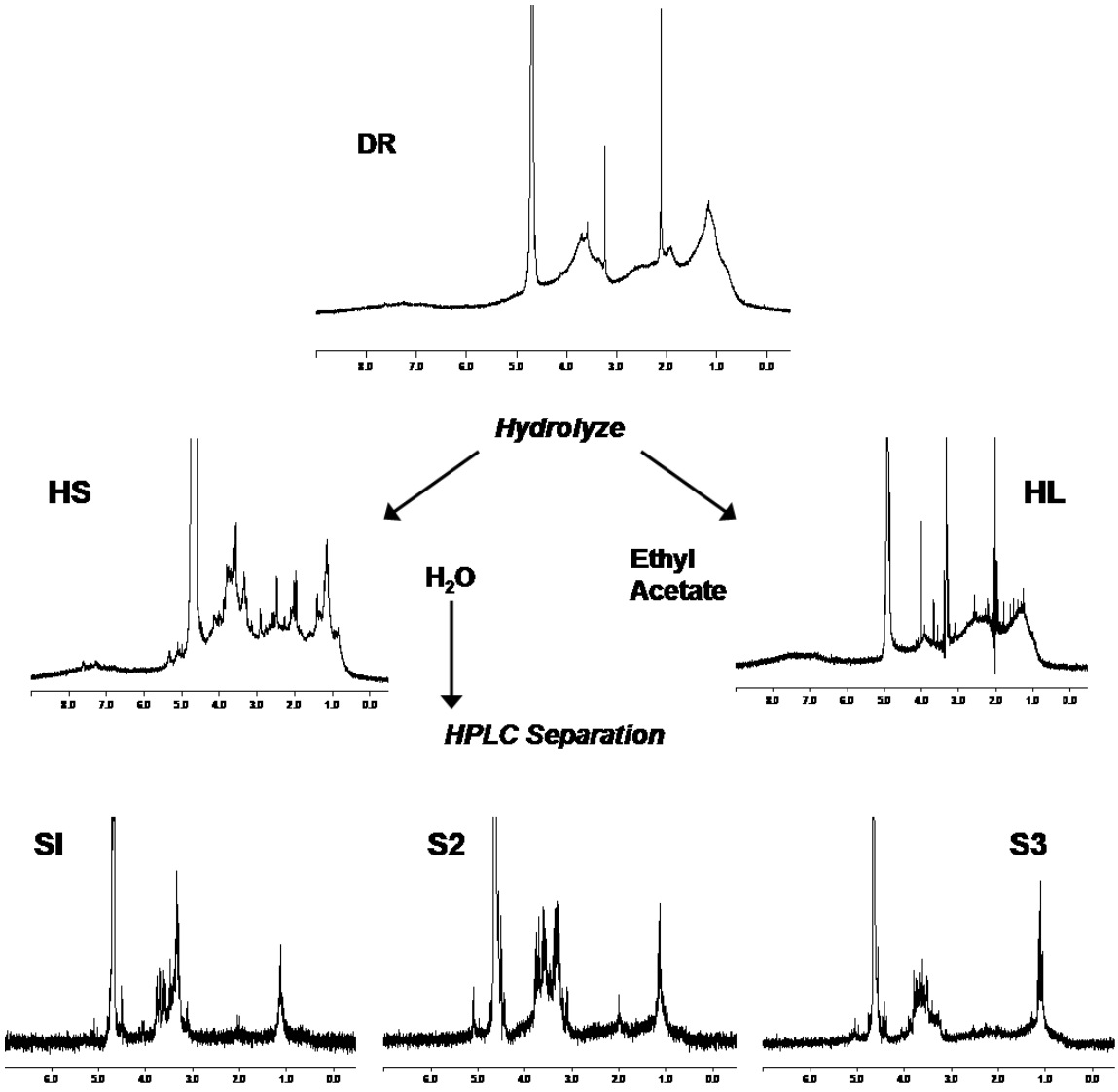

Figure 5.2. Chemical fractionation scheme for the DR. The general composition of each fraction is determined by 1H NMR spectra. S1 (glucose + unknown), S2 (xylose, rhamnose and galactose) and S3 (arabinose, fucose and mannose). All NMR scales span 0.0 to 8.0 ppm.

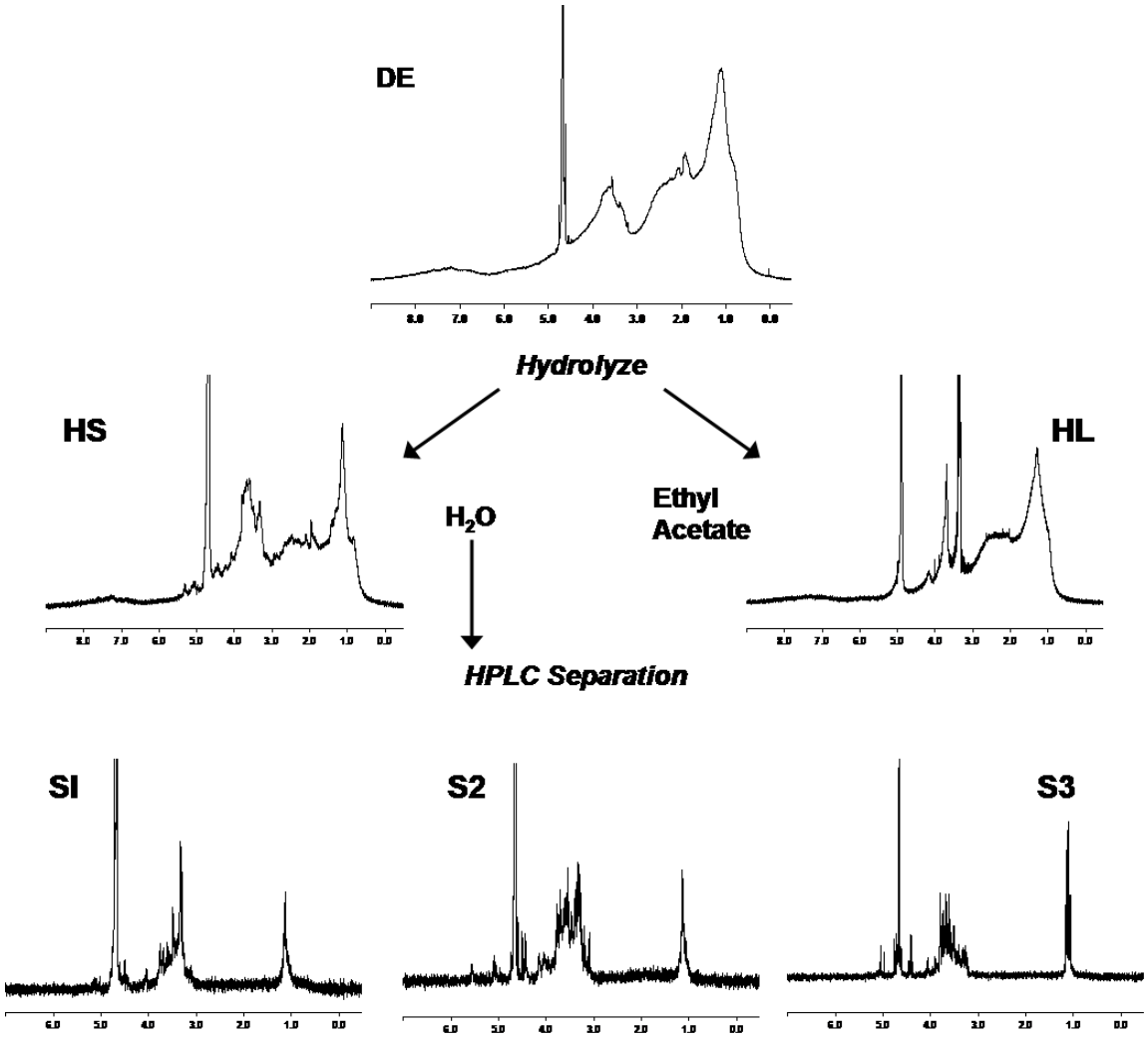

Figure 5.3. Chemical fractionation scheme for the DE. The general composition of each fraction is determined by ^{1}H NMR spectra. S1 (glucose + unknown), S2 (xylose, rhamnose and galactose) and S3 (arabinose, fucose and mannose). All NMR scales span 0.0 to 8.0 ppm.

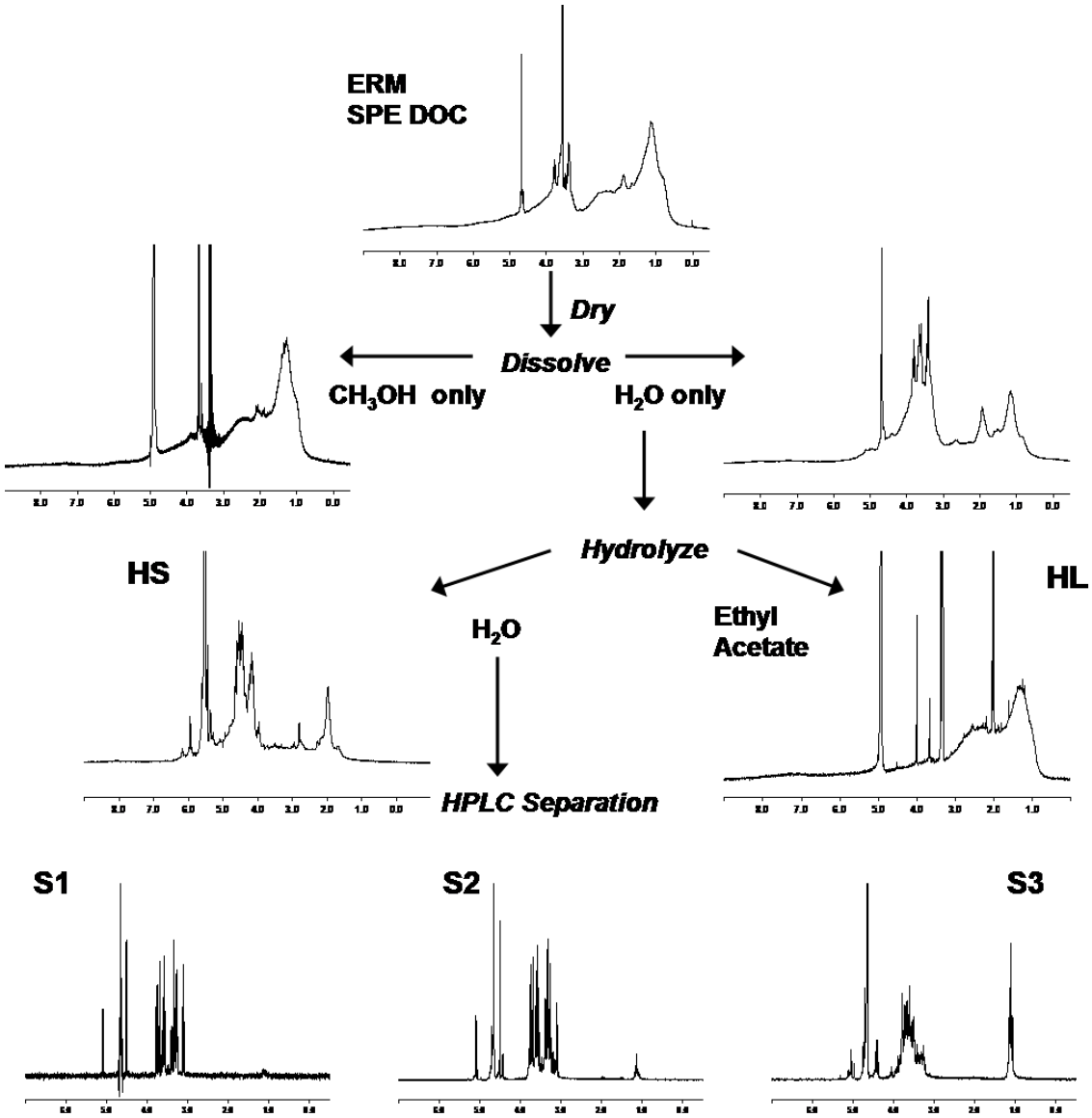

Figure 5.4 Chemical fractionation scheme for ERM. The general composition of each fraction is determined by ^{1}H NMR spectra. S1 (glucose), S2 (xylose, rhamnose and galactose) and S3 (arabinose, fucose and mannose). All NMR scales span 0.0 to 8.0 ppm.

5.7 **APPENDIX**

The amount of CO_2 purified from sugar and lipid samples from Chapter 4 and this chapter are tabulated with the respective Fraction modern (F_m; corrected for $\delta^{13}C$) and $\Delta^{14}C$ values. $\delta^{13}C$ values were obtained from splits at NOSAMS.

Sample	μg C (as CO_2)	Fraction modern	$\delta^{13}C$ (‰)	$\Delta^{14}C$ (‰)	$\Delta^{14}C$ error
DF2-3HL	813.2	0.9932	-27.98	-13.5	4.2
DF2-3S1	35.9	Lost	-	-	-
DF2-3S2	86.7	0.9917	-24.07	-15.0	7.4
DF2-3S3	100.9	0.9777	-25.25	-28.9	5.9
DF2-24HL	825.1	0.9086	-24.82	-97.5	3
DF2-24S1	38.6	0.9702	-23.29	-36.4	8.7
DF2-24S2	49.4	1.0552	-21.3	48.1	8
DF2-24S3	60.9	1.0279	-20.4	21.0	9.2
Eel-BHL	765.7	0.6819	-23.97	-322.7	2.5
Eel-S1	167.2	0.9647	-18.56	-41.8	5.3
Eel-S2	169.3	1.0128	-21.26	6.0	6.3
Eel-S3	152.3	0.9899	-20.88	-16.8	6.5
HMW-HL	484.1	0.6913	-24.51	-313.4	3
HMW-S1	79.2	0.9997	-21.73	-7.0	7.4
HMW-S2	185.5	1.0299	-20.98	22.9	5.8
HMW-S3	270.8	1.0492	-21	42.1	3.5
0411-BHL	381.2	0.8161	-23.77	-189.2	4.2
0411-S1	42.7	1.0548	-22.05	47.9	13.4
0411-S2	67.7	0.9908	-21.77	-15.7	14.5
0411-S3	167.9	1.0273	-21.55	20.6	9.1

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6 CONCLUSION

6.1 INTRODUCTION

The amount of dissolved organic carbon (DOC) in the deep ocean is approximately equal to that of atmospheric CO_2 making it an important reservoir in the global carbon cycle (Hansell and Carlson, 1998). The average ^{14}C-age of marine DOC in the deep North Pacific Ocean is approximately 6000 years relative to atmospheric CO_2, which is substantially longer than the mixing time of the oceans (~1000-1500 years; Williams and Druffel, 1987; Druffel et al., 1992). The ocean's capability to sequester organic carbon over long timescales and several mixing cycles must be understood to describe a more complete global carbon cycle and by extension, provide better constraints on processes controlling global climate.

The transformation of fresh DOC into refractory material is an important step toward carbon sequestration in the ocean. While many studies have focused on the elemental and isotopic characteristics of the bulk DOC pool (e.g., Bauer 2002; Benner 2002), it is becoming increasingly clear that both short- and long-term variations in DOC concentration and cycling will be better understood if we examine specific components within this reservoir. As a result focus has shifted toward characterizing certain fractions of DOC. One of the most powerful tools available to researchers who examine the reactivity and residence time of DOC and its constituents is natural abundance radiocarbon measurements ($\Delta^{14}C$). In combination with chemical characterization, radiocarbon analysis has proven useful in identifying the labile compounds in DOC. Yet

the fraction of DOC that is responsible for contributing ^{14}C-depleted components is poorly studied, which has seriously hampered efforts aimed at fully describing the DOC cycle in marine environments. This thesis attempted to advance studies of DOC cycling by characterizing specific fractions of dissolved organic matter (DOM) isolated from the eastern North Pacific Ocean to identify both refractory and labile DOC components.

6.2 GENERAL CONCLUSIONS AND IMPLICATIONS

Chapter 2 provided a description of total organic carbon (TOC) and total organic nitrogen (TON) distributions in the California Current System during 2005 as a first step toward identifying the chemical, biological and physical factors that control the size of the DOM reservoir. Clear seasonal variations in TOC and TON were related to the extent of seasonal production, physical structure of the water column, and position of the California Current. For example, highest TOC accumulations were observed during July 2005 when chlorophyll a and nitrate data recorded increases in upwelling induced primary production. In the fall TOC concentrations began to approach low winter values, and entrainment into subsurface waters, offshore transport and bacterial degradation may have all contributed to the removal of TOC and TON.

The Santa Barbara Basin represents a complex and dynamic environment where denitrification and a flushing event were observed in 2005. TOC and TON values responded to these events, which were identified by hydrographic and nutrient data, specifically N*. Both TOC and TON increased in near bottom waters where denitrification was active (0501 and 0511), consistent with the release of C and N from

bottom waters.

Overall, seasonal and spatial variations were consistent with the production and removal of labile DOC on annual timescales making the California Current System an ideal location to study refractory and labile DOC components. As such, dissolved fractions were isolated from this region to identify the range of timescales over which DOC cycles.

The major goal of Chapters 3, 4 and 5 was to identify modern and refractory DOC components using a combination of isotope and chemical analyses. In Chapter 3, a novel solid phase extraction (SPE) method was developed to isolate and chemically fractionate DOC. The method, when applied correctly with careful consideration of resin associated contaminants, is capable of isolating a representative fraction of DOC. DOC isolated by this method was shown to include both labile and refractory components through radiocarbon analysis.

Proton nuclear magnetic resonance (^{1}H NMR) spectroscopy was used in this study to show that SPE DOC contained both polar material with chemical characteristics resembling carbohydrates and non-polar, aliphatic compounds. Chemical fractionation of SPE DOC indicated that fractions enriched in aliphatic compounds were also more depleted in ^{14}C, and hinted at a relationship between chemical composition and DOC residence time. Based on these observations, a two component model was constructed to examine whether knowledge of the chemical composition of each fraction enabled an estimation of its Δ^{14}C signature. The non-polar region of the ^{1}H NMR spectrum where functional groups from aliphatic compounds exhibited resonances (0-1.5 ppm) was quantified to estimate the contribution of refractory DOC. This fraction of aliphatic

carbon was assigned a $\Delta^{14}C$ signature of -550‰ (Bauer et al., 1998). The polar or oxygen-rich region of the spectrum was quantified and assigned to a modern carbon component (with a $\Delta^{14}C$ signature of 20‰ - the $\Delta^{14}C$ value of inorganic carbon in surface waters). These two components together were sufficient to reproduce the measured $\Delta^{14}C$ signature of each SPE fraction in surface waters. This study provided the first conclusive evidence in support of a unique chemical composition for refractory DOM.

In Chapter 4 neutral sugars and lipids were isolated from hydrolyzed SPE DOC and ultrafiltered DOC samples for radiocarbon analysis. This study was conducted to further investigate the ^{14}C-age and chemical structure relationship, and to validate the two component model presented above. Results revealed that both SPE and ultrafiltration methods isolated sugars with a modern ^{14}C signature. Furthermore, lipids released from the acid hydrolysis of both samples were ^{14}C-depleted relative to bulk DOC fractions. These results confirmed the presence of both labile and refractory compounds in DOM. The presence of these compounds in fractions that were isolated by two very different techniques suggested that current isolation methods are studying representative DOC fractions. Furthermore, the chemical degradation techniques used to isolate lipids and sugars indicated that refractory and labile components were associated together in each sample. This association between hydrophobic (lipids) and hydrophilic (sugars) compounds may explain how lipids stay dissolved in seawater, and why some carbohydrates accumulate in the surface ocean.

Results from this investigation have important implications for DOC cycling in the marine environment. If fresh DOC is in fact chemically or physically associated

with humic material, it could be rendered unavailable to microorganisms. The associated materials could be subducted into the deep ocean where further biotic or abiotic reactions could take place to alter the chemical structure of DOC. Alternatively, if fresh DOC is selectively preserved and subducted into the deep ocean then this association represents a mechanism for natural CO_2 sequestration. The subduction and subsequent preservation of polar material in the deep ocean could also explain the observed deviation of subsurface samples from the NMR model presented in Chapter 3.

Previous studies have suggested that DOC could be encapsulated and protected from degradation by large macromolecular structures (Nguyen and Harvey, 2001). Results from this thesis support this potential mechanism for DOM preservation and identifies hydrophobic compounds as the main constituent of these protective matrices. Data presented here further support the recent assertion that DOC accumulating in the deep ocean resembles highly carboxylated terpenoids (i.e., aliphatic, relatively hydrophobic compounds; Hertkorn et al., 2006). Furthermore, the separation and isolation of two classes of compounds with distinct $\Delta^{14}C$ values further supports the two component age model for marine DOC.

The final study of this thesis (Chapter 5) used the methods employed in Chapter 4 to reveal information about the cycling of riverine and terrestrial DOC. This study represents the first to apply compound specific isotopic analyses (CSIA) to study terrestrial DOC cycling in rivers. Results revealed that DOC derived from terrestrial production was quickly removed in the Delaware Estuary and replaced with chemically similar components that have isotopic signatures unique to the marine environment. This finding is consistent with earlier studies that detected relatively low concentrations

of terrestrial DOM in marine environments (e.g., Meyers-Schulte and Hedges, 1986). However, glucose appeared to be selectively preserved in the estuary as revealed by $\delta^{13}C$ and $\Delta^{14}C$ analyses, and could represent a previously unrecognized source of terrestrial DOC to marine environments. Lipids were also replaced in the estuary indicating that refractory lipids in marine DOC are not derived from terrestrial sources.

At the Eel River Margin site, CSIA were able to effectively demonstrate that glucose accumulating in this terrestrially influenced system, had a unique biological source and/or residence time compared to other sugars in the same sample. Isotope data also revealed that terrestrial/riverine inputs, methane oxidation, and upper ocean primary production may all contribute DOC to this site.

A two component radiocarbon and mass balance model has been widely used to describe the DOC cycle in marine environments, yet no study has focused on isolating dissolved refractory compounds from the ocean to validate this model. Through the use of ^{14}C analysis and chemical characterization this thesis has provided sufficient evidence in support of a refractory component accumulating within marine DOC, and directly validates the two component ^{14}C-age model.

6.3 EVALUATION OF THESIS

TOC and TON measurements in 2005 represent the largest spatial survey over an entire year. Although strict statistical analysis was not performed in this study, patterns from hydrographic data could be superficially extracted. A more detailed statistical analysis could reveal clear information about the temporal and spatial cycling of TOC

and TON throughout the California Current System.

The novel method of this thesis presents a unique perspective on the chemical structure and residence time of DOC in the ocean. Although only capable of extracting 15% of total organic carbon in the surface ocean CSIA showed that SPE possessed similar compounds to ultrafiltered DOC (UDOC). Furthermore, lipid material is more concentrated in the SPE fractions relative to UDOC thus representing a broader suite of compounds.

As with any technique involving marine DOC and natural abundance ^{14}C analyses, the potential for contamination exists in many of the experimental set ups and sampling procedures. These pitfalls were largely avoided after initial studies and a more careful inspection of the sources of organic contaminants. Process blanks allowed for isotope corrections.

The CSIA and subsequent use of ^{1}H NMR was an attempt to show that operational techniques to isolate additional compounds to those which are targeted, that is, after acid hydrolysis, neutral sugars only represent a small amount of material from the hydrosylate as calculated by GC analysis. Therefore, it should be taken into consideration that compounds be positively identified through chemical analyses after prescribed operational isolations.

Overall, this thesis represents a major step in studying the cycling and residence time of DOC in marine environments including bulk DOC and specific compounds. This thesis provides ample evidence that, as indicated by $\Delta^{14}C$ values, lipids and sugars have different biological and chemical reactivity in the surface ocean. This finding is significant in understanding DOC cycling as it relates to the potential of DOC as a

natural pathway for CO_2 sequestration in the ocean.

6.4 FUTURE STUDIES

An exciting follow-on to this study is to embark on the characterization of lipid fractions obtained after hydrolysis. Such a study, when combined with CSIA, would identify the composition and source of refractory, hydrophobic compounds accumulating in the marine environment. The DOC isolation scheme described in this thesis will prove instrumental in this endeavor. In addition, further isolation and characterization of individual compounds should be aimed at determining the nature of the association between carbohydrates and lipids in both SPE and ultrafiltered DOM. Such an effort to describe mechanisms that produce refractory organic matter is a necessary part of any investigation striving to understand carbon sequestration in marine environments. The ^{14}C content of compounds and fractions studied here were all equal to or enriched relative to bulk DOM, and these results require the existence of other compounds within DOM that are even more depleted in ^{14}C. Data from this study points to their existence in the hydrophobic DOM fraction, and so, compounds should be isolated for CSIA from this fraction so that the ^{14}C signature of refractory DOC can be definitively established.

In addition, future studies should focus on obtaining data from deep samples where refractory DOM is predominant. For example, in Chapter 3, the two component model based on NMR data and ^{14}C measurements was unable to accurately reproduce the $\Delta^{14}C$ signature of subsurface SPE DOC. One explanation may be due to either an underestimation of ^{1}H NMR resonances belonging to refractory DOC or the presence of

^{14}C-depleted, O-rich DOM in the deep ocean. To distinguish the underestimation of refractory C from an overestimation of the ^{14}C-signature of carbohydrates, CSIA could be applied to lipids and sugars isolated from the deep ocean. This underestimation also warrants further investigation to confirm or refute the two component model proposed in this thesis.

Although studies that comprised this thesis isolated sugars and lipids from DOC, a large fraction of marine DOC still remains uncharacterized. The uncharacterized fraction includes low molecular weight compounds (<1000 MW; LMW), acidic and acetylated sugars, and amino acids, all of which could be isolated for CSIA. The size continuum model suggests that LMW compounds are refractory (Amon and Benner, 1994); therefore efforts concentrating on LMW DOC could harbor exciting results. CSIA of these compounds would provide further insight into the composition and residence time of refractory DOC.

Finally, two major technological advances will aid in the^{14}C analysis of specific compounds. The first is the capability to perform ^{14}C analyses on <10 μg of carbon (e.g., Ingalls et al., 2006, Santos et al., 2007), and the second is the in-line AMS system being currently developed at the National Ocean Sciences Accelerator Mass Spectrometry facility which eliminates the need to convert CO_2 to graphite.

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