



## Rapid nitrification of wastewater ammonium near coastal ocean outfalls, Southern California, USA



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### ARTICLE INFO

#### Article history:

Received 3 November 2014

Received in revised form

14 April 2016

Accepted 12 May 2016

Available online 13 May 2016

#### Keywords:

Southern California bight

Anthropogenic nutrients

Wastewater

Nitrification

Nitrogen cycling

Stable isotopes

### ABSTRACT

In the southern California Bight (SCB), there has been a longstanding hypothesis that anthropogenic nutrient loading is insignificant compared to the nutrient loading from upwelling. However, recent studies have demonstrated that, in the nearshore environment, nitrogen (N) flux from wastewater effluent is equivalent to the N flux from upwelling. The composition of the N pool and N:P ratios of wastewater and upwelled water are very different and the environmental effects of wastewater discharges on coastal systems are not well characterized. Capitalizing on routine maintenance of the Orange County Sanitation District's ocean outfall, wherein a wastewater point source was "turned off" in one area and "turned on" in another for 23 days, we were able to document changes in coastal N cycling, specifically nitrification, related to wastewater effluent. A "hotspot" of ammonium ( $\text{NH}_4^+$ ) and nitrite ( $\text{NO}_2^-$ ) occurred over the ocean outfall under normal operations and nitrification rates were significantly higher offshore when the deeper outfall pipe was operating. These rates were sufficiently high to transform all effluent  $\text{NH}_4^+$  to nitrate ( $\text{NO}_3^-$ ). The dual isotopic composition of dissolved  $\text{NO}_3^-$  ( $\delta^{15}\text{N}_{\text{NO}_3}$  and  $\delta^{18}\text{O}_{\text{NO}_3}$ ) indicated that N-assimilation and denitrification were low relative to nitrification, consistent with the relatively low chlorophyll and high dissolved oxygen levels in the region during the study. The isotopic composition of suspended particulate organic matter (POM) recorded low  $\delta^{15}\text{N}_{\text{PN}}$  and  $\delta^{13}\text{C}_{\text{PN}}$  values around the outfall under normal operations suggesting the incorporation of "nitrified"  $\text{NO}_3^-$  and wastewater dissolved organic carbon into POM. Our results demonstrate the critical role of nitrification in nitrogen cycling in the nearshore environment of urban oceans.

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### 1. Introduction

Nitrogen (N) pollution is considered to be one of the greatest consequences of human-accelerated global change on coastal oceans (Howarth and Marino, 2006). However, in upwelling dominated systems such as the Southern California Bight (SCB), there has been a general perception that the flux of anthropogenic nutrient inputs is insignificant relative to upwelling flux, and therefore anthropogenic inputs have relatively little effect on the productivity of coastal waters (Chavez and Messié, 2009; Capone and Hutchins, 2013). Recent studies in the SCB have shown that, in nearshore regions associated with high urbanization, N loads

from wastewater effluent are roughly equivalent to nutrient loads from upwelling (Howard et al., 2014). This has effectively doubled the N loading to the shelf, potentially altering the composition of the N pool (effluent N is primarily ammonium, upwelled N is primarily nitrate) as well as the nitrogen: phosphorus (N:P) ratio (P discharges are low relative to N; effluent N:P ~115). The effect of these discharges on nearshore biological community composition and nutrient cycling is largely unknown.

There is an emerging body of circumstantial evidence suggesting that terrestrial, anthropogenic nutrients, dominated by wastewater effluent, are having an effect on primary productivity and respiration in Southern California coastal waters. In particular, increased nearshore primary productivity and the spatial extent and duration of algal blooms have increased over the past decade (Nezlin et al., 2012). Furthermore, respiration of high levels of organic matter from algal blooms may be contributing to

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observations of reduced coastal dissolved oxygen concentrations relative to offshore trends (Booth et al., 2014). However, the extent and magnitude of increased productivity and respiration, particularly that which is supported by wastewater nutrients, relative to baseline production in the SCB is still uncertain.

The San Pedro Shelf in the SCB is an ideal place to study the fate of wastewater ammonium in urban coastal waters. Two of the four major Publicly Owned Treatment Works (POTW) in the SCB discharge wastewater on the San Pedro Shelf and these outfalls make up to 50% of all of the POTW discharges in the SCB (Howard et al., 2012). In addition, between 1971 and 2000, the effluent volume discharged to the SCB increased by 31% and N emissions increased 91% (Lyon and Stein, 2009). For this study, we took advantage of planned maintenance of the Orange County Sanitation District's (OCS)D effluent outfall pipes. For three weeks in the fall of 2012, OCS)D diverted its effluent stream (138 million gallons of wastewater per day) from its deeper outfall to its shallower outfall while the latter was cleaned and repaired. This diversion provided a unique opportunity to conduct an *in situ* experiment (normally prohibited by regulatory requirements) to directly document the effects of turning off a wastewater point source in one area and turning it on in another. We focus on the effect of wastewater N because the SCB is largely N limited (Thomas et al., 1974; Cullen and Eppley, 1981) and the wastewater contains high levels of nitrogenous species. Because effluent N is primarily ammonium and previous observations in the SCB have indicated the ammonium is a minor N species despite the large ammonium-dominated wastewater discharges (Howard et al., 2012), we hypothesized that wastewater ammonium is rapidly nitrified near POTW outfalls.

To understand the fate of effluent N, we employed stable isotope techniques to trace dissolved nitrate, ammonium, and particulate organic matter; we also determined rates of nitrification of ammonium, the dominant N form in effluent. The stable isotopic compositions of dissolved nitrate ( $\delta^{15}\text{N}_{\text{NO}_3}$  and  $\delta^{18}\text{O}_{\text{NO}_3}$ ) and suspended particulate matter ( $\delta^{15}\text{N}_{\text{PN}}$  and  $\delta^{13}\text{C}_{\text{PN}}$ ) are natural tracers of N sources and cycling in the ocean. Variation in the isotopic composition is attributable to distinct source signatures and the mass dependent isotopic discriminations associated with various biogeochemical transformations that constitute the marine N cycle. Because each pathway causes a characteristic shift in isotope composition of the products and reactants, the isotopic composition of the dissolved and particulate pools can provide useful information on the mechanism of these transformations (Sigman et al., 2005; Wankel et al., 2007; Sugimoto et al., 2009).

## 2. Materials and methods

### 2.1. Field collection

The OCS)D discharges wastewater from its treatment facility in Huntington Beach, California offshore via one of two outfall pipes located on the San Pedro Shelf in Southern California. Normal operations require OCS)D to discharge effluent from its deeper outfall located 7 km off shore and in 56 m of water depth (33°34.5'N; 118°00.5'W), hereafter referred to as “deep outfall”; however, during maintenance of the deep outfall, wastewater was diverted to OCS)D's secondary outfall pipe located 1.6 km offshore in 16.7 m water depth (33°36.8'N; 117°58.5'W), hereafter referred to as “shallow outfall”. Wastewater was diverted from 11 September 2012 until 3 October 2012. Sampling was conducted pre-diversion (6 September 2012), two weeks into the diversion (20 September 2012), shortly after the diversion ended (less than 24 h), hereafter called transition (3 October 2012), and two weeks post-diversion (17 October 2012). Sampling cruises on 6 Sep and 20 Sep were conducted aboard the *R/V Yellowfin* and cruises on 3 Oct and 17 Oct

were conducted on the *M/V Nerissa*.

Sampling was conducted at stations along an across shelf transect from the shallow outfall pipe past the deep outfall to the offshore stations (Fig. 1). Vertical profiles were collected at each station using a package containing a SBE911plus (Seabird Electronics, Inc.) conductivity-temperature-depth (CTD) profiler with a chlorophyll sensor and a colored dissolved organic matter (CDOM) sensor; however the CDOM sensor aboard the *R/V Yellowfin* failed quality assurance tests and, thus, data from that sensor was rejected for cruises on 6 Sep and 20 Sep.

Discrete samples were collected from Niskin bottles (1.5 and 3 L) on a rosette deployed with the sensor package. Samples were collected at three depths at each station: surface, deep chlorophyll maximum (12–30 m), and below the mixed layer (“subeuphotic”, 30–75 m). Sampling was adaptive at each site, with sample depths determined by downcast observations of chlorophyll (to determine the layer of maximum chlorophyll fluorescence) and temperature, salinity and CDOM (to determine the below mixed layer depth that fell within the plume track). The “subeuphotic” samples (the deepest samples collected) from the periods when effluent was discharged from the deep outfall were collected within the center of the plume track directly over the outfall (at station 2205). However, because the plume-track rose just below the euphotic zone as it moved offshore, subeuphotic samples at the offshore stations were collected below the center of the plume track, in more effluent-diluted waters, to minimize photoinhibition of nitrification. All samples were transferred from the Niskins using acid-washed Tygon tubing into acid-washed 2 L high density polyethylene (HDPE) bottles that were triple rinsed with sample water before filling. Sub-samples for nutrient and nitrate stable isotope analysis were vacuum filtered through a 0.45  $\mu\text{M}$  polycarbonate filter (Millipore) and collected in triple-rinsed, 60 mL HDPE amber bottles, stored on ice for transport to the laboratory, and frozen until analysis (Wankel et al., 2006, 2007; Santoro et al., 2010a). Samples for ammonium stable isotope analysis were vacuum filtered through a 0.45  $\mu\text{M}$  polycarbonate filter (Millipore), collected into triple-rinsed, 2 L HDPE bottle, acidified with several drops of concentrated ammonia-free sulfuric acid to a pH less than 2 and stored on ice for transport, and kept at 4 °C until analysis (Holmes et al., 1998). Samples for nitrification rate incubations were collected directly into acid washed, triple-rinsed, 2 L HDPE bottles and stored on ice in the dark until the incubations could begin in the laboratory within six hours of collection.

Suspended particulate and net tow samples were collected by vacuum filtration onto pre-combusted (450 °C for 4 h) glass fiber filters (Whatman GF/F). Suspended particulate samples were subsampled from the same whole water sample collected for nutrient and stable isotope analyses. Net tow samples were collected by lowering both a 20  $\mu\text{m}$  and 200  $\mu\text{m}$  net through the water column 3 times to 10 m to generate a composite sample of the plankton communities. Tow samples were collected into a 1L HDPE bottle prior to filtration. Filters were collected into snap-close petri dishes and stored in Ziploc bags on ice in the dark for transport. Filters in the petri dishes were dried at 50 °C in the dark until analysis.

OCS)D supplied samples of effluent for analysis prior to each sampling event. These samples were collected as a composite over 24 h in an acid washed 1 L HDPE bottle. Effluent was stored at 4 °C while the composite was generated. The composite was subsampled into HDPE bottles and hand-filtered through 0.45  $\mu\text{M}$  polycarbonate filter (Millipore) for nutrient concentrations, nitrate dual isotope analysis, and was acidified for ammonium stable isotope analysis.

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