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Seasonal and annual dynamics of harmful algae and algal toxins revealed through weekly monitoring at two coastal ocean sites off southern California, USA

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Abstract Reports of toxic harmful algal blooms (HABs) attributed to the diatom *Pseudo-nitzschia* spp. have been increasing in California during the last several decades. Whether this increase can be attributed to enhanced awareness and monitoring or to a dramatic upswing in the development of HAB events remains unresolved. Given these uncertainties, the ability to accurately and rapidly identify an emerging HAB event is of high importance. Monitoring of HAB species and other pertinent chemical/physical parameters at two piers in southern California, Newport and Redondo Beach, was used to investigate the development of a site-specific bloom definition for identifying emerging domoic acid (DA) events. Emphasis was given to abundances of the *Pseudo-nitzschia seriata* size category of *Pseudo-nitzschia* due to the prevalence of this size class in the region. *P. seriata* bloom thresholds were established for each location based on deviations from their respective long-term mean abundances, allowing the identification of major and minor blooms. Sixty-five percent of blooms identified at Newport Beach coincided with measurable DA concentrations, while 36 % of blooms at Redondo Beach coincided with measurable DA. Bloom definitions allowed for increased specificity in multiple regression analysis of environmental forcing factors significant to the presence of DA and *P. seriata*. The strongest relationship identified was between *P. seriata* abundances 2 weeks following upwelling events at Newport Beach.

Keywords Harmful algal bloom · *Pseudo-nitzschia* · Domoic acid · Bloom definition · Time series · Microalgae

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Introduction

Substantial increases in microalgal biomass in planktonic ecosystems, generally observed as increases in chlorophyll *a* concentrations or cell abundances, serve as the foundation of highly productive oceanic food webs, spawning productive fisheries and foraging areas for marine mammals, birds, and other large predators (Legendre 1990). However, toxic or harmful algal blooms (HABs) produced by a few species of microalgae can have negative impacts on local food webs as well as threaten human health. Nearly 300 of the >4,000 currently described species of marine microalgae are considered capable of forming HABs and approximately 80 of those species are known to be capable of producing compounds that are toxic to co-occurring marine species and/or humans (Sournia 1995). HAB events are highly diverse in their taxonomic composition, spatiotemporal distributions, and detrimental effects, complicating the understanding of their ecology, reducing the accuracy of predicting outbreaks, and impeding the development of successful management strategies (Smayda 1997; Zingone and Enevoldsen 2000; Anderson et al. 2012). Anthropogenically influenced

changes in climate, and nutrient loading is, in part, responsible for the global increase in the incidence, magnitude, and duration of HAB events (Paerl 1997; Van Dolah 2000; Anderson et al. 2002; Glibert et al. 2005; Heisler et al. 2008; Kudela et al. 2008; Paerl and Paul 2012). The impact anthropogenic driven change will have on a given region will be determined by the HAB organisms present and the magnitude of the change experienced in environmental conditions responsible for local HAB initiation, maintenance, and demise.

HABs along the coastline of California have been common for many years, although poorly characterized until relatively recently. The first dependable records most likely began with the documentation of massive red tides attributed to the dinoflagellate *Lingulodinium polyedrum* that have occurred sporadically along the coast since the beginning of the twentieth century (Torrey 1902; Allen 1938, 1946; Gregorio and Pieper 2000; Shipe et al. 2008; Omand et al. 2011). Yessotoxin production by *L. polyedrum* in California strains has been confirmed (Howard et al. 2008), but this toxin has failed to be accredited as the cause of any instance of marine animal or human illness in the area. Numerous other potentially toxic species of microalgae have also been documented in California waters, including several raphidophytes and dinoflagellate species within the genera *Akashiwo*, *Alexandrium*, *Cochlodinium*, and *Dinophysis* (Jessup et al. 2009; Jester et al. 2009; Caron et al. 2010; Garneau et al. 2011; Howard et al. 2012; Lewitus et al. 2012).

Members of the diatom genus *Pseudo-nitzschia* are one of the few non-flagellated microalgae currently known to be capable of toxin production. The toxin they produce, domoic acid (DA), is also produced by a closely related diatom, *Nitzschia navis-varingica* (Lundholm and Moestrup 2000; Kotaki et al. 2004; Lundholm et al. 2004; Lelong et al. 2012), and potentially by the diatom *Amphora coffeaeformis* (Maranda et al. 1990; Sala et al. 1998; Bates 2000). The capability of *Pseudo-nitzschia* to produce the powerful neurotoxin was originally discovered on Prince Edward Island, Canada, in 1987 when more than 150 people were sickened and three perished after consuming blue mussels (*Mytilus edulis*) contaminated with DA during a bloom of *P. multiseries* (Bates et al. 1989; Wright et al. 1989). Human consumption of shellfish contaminated with the neurotoxin DA causes amnesic shellfish poisoning (ASP), the main symptoms include disorientation, gastrointestinal illness, memory loss and even death. *Pseudo-nitzschia* spp. has been reported as a frequent contributor to the microalgal community in coastal waters of California since the early 1900s (Allen 1934, 1936; Fryxell et al. 1997), however toxin production by these species was not documented until 1991 following the poisoning and deaths of brown pelicans (*Pelecanus occidentalis*) and Brandt's cormorants (*Phalacrocorax penicillatus*) in Santa Cruz, CA, corresponding with a bloom of *Pseudo-nitzscia australis* (Fritz et al. 1992; Work et al. 1993). DA continues to be the cause of mortality events of marine mammals and birds along the US west coast and elsewhere (Lefebvre et al. 1999; Scholin et al. 2000; de la Riva et al. 2009; Fire et al. 2009; Fire et al. 2010; Hall and Frame 2010; Lefebvre et al. 2010) although no human deaths have been reported since the initial Canadian ASP outbreak in 1987. The threat of DA poisoning in humans from seafood other than shellfish has been highlighted in studies of exposure via recreational fishing activities (Busse et al. 2006; Mazzillo et al. 2010), as well as the potential for DA to be transferred to trophic levels not directly consuming toxic *Pseudo-nitzschia* cells (Busse et al. 2006; Vigilant and Silver 2007; Kvitek et al. 2008; Mazzillo et al. 2011).

DA has been detected in strains of *Pseudo-nitzschia* from California, other US Pacific coast states and in multiple other locations around the globe (Anderson et al. 2006; Schnetzer et al. 2007; Lelong et al. 2012; Seubert et al. 2012; Stauffer et al. 2012; Trainer et al. 2012, Schnetzer et al. 2012). The locations in which DA has been detected are highly variable with respect to chemical and physical oceanography, meteorology and nutrient inputs. The ability of multiple species of *Pseudo-nitzschia* to produce DA and the inconsistent nature of toxin production imply that more than one "trigger" may be involved in stimulating DA production, a possibility that is supported by several laboratory studies. Numerous hypotheses exist regarding the conditions that can give rise to DA production in *Pseudo-nitzschia* including biological, chemical and physical aspects of the ecosystem (reviewed in Lelong et al. 2012; Lewitus et al. 2012). A definitive cause for blooms of *Pseudo-nitzschia* spp. and DA production has not been determined, hindering attempts to predict these events, in spite of the wealth of research on the subject. Anthropogenic eutrophication and climate change does not currently appear to be the reason behind *Pseudo-nitzschia* bloom events and DA production (Lewitus et al. 2012) although those factors have been found to be responsible for the increased incidence of other HAB events globally. A general lack of predictability of DA outbreaks remains the primary motivation for implementing HAB monitoring programs that include *Pseudo-nitzschia* and DA. Models are now emerging that provide some basic predictive power, but they are still relatively early in their development, generally requiring direct measurements of microalgal abundances and toxin concentrations for parameterization and validation (Anderson et al. 2009; Lane et al. 2009; Anderson et al. 2010). Long-term datasets of microalgal abundance and environmental parameters are essential for documenting blooms and to extend the understanding of potential environmental conditions leading to them (Kim et al. 2009; Glibert et al. 2010; Frolov et al. 2012).

The Southern California Coastal Ocean Observing System (SCCOOS) HAB Monitoring program was initiated in 2008. The program entails weekly samples collected for monitoring

the abundances of potentially harmful microalgal species, particulate DA and nutrient concentrations at pier locations along the coast of southern California. Sampling is carried out by investigators at California Polytechnic University, University of California Santa Barbara, University of California Los Angeles, University of Southern California, and the Scripps Institution of Oceanography, University of California San Diego. Automated sensor packages maintained by SCCOOS provide basic chemical/physical parameters on a continuous basis and daily discrete samples for temperature and salinity collected as a part of the Scripps Institution of Oceanography Shore Station Program occurs at several of the SCCOOS HAB monitoring locations. In this report, information collected for 3.5 years at the pier in the City of Newport Beach, Orange County (33° 36′ N, 117° 55′ W) and for 2 years at the pier in the City of Redondo Beach, Los Angeles County (33° 50′ N, 118° 23′ W) were analyzed in order to establish a baseline for identifying algal blooms in the region based on easily-collected parameters and to determine whether this information could be used to specifically identify blooms of *Pseudo-nitzschia* and DA events. A statistical analysis of these datasets was conducted with the objective of improving our understanding of the conditions that give rise to toxic *Pseudo-nitzschia* blooms in southern California and to investigate algal bloom definitions. Historically, algal blooms have been arbitrarily defined, with large variations in definitions between locations, laboratories and researchers. Most investigations into identifying bloom events have centered around unusually high chlorophyll *a* concentrations, focusing on deviations from a mean chlorophyll concentration specific to the location being studied (Carstensen et al. 2007; Henson and Thomas 2007; Allen et al. 2008; Kim et al. 2009). These studies have differed in the time span over which the mean chlorophyll concentration has been determined and the method of collection of the chlorophyll concentrations (i.e., satellite imagery, in vivo fluorescence, fluorometric, or spectrophotometric analysis of chlorophyll concentration from discrete water samples filtered onto and extracted from membrane filters).

Materials and methods

Sample collection and processing

Weekly sampling at Newport Pier in Orange County, California, began on June 30, 2008, as a part of the SCCOOS HAB Monitoring Program (Fig. 1). Surface seawater (2 l) was collected in acid-washed (5 % HCl) polycarbonate bottles every Monday, kept cool and out of direct sunlight until processing in the laboratory approximately 1–2 h after collection. Net tow samples were collected in conjunction with the whole seawater samples with a 20-μM net (Sea-gear, Melbourne, FL) for relative abundance determinations of microalgae present. In January 2010, the HAB monitoring effort was expanded to include the Redondo Beach pier in Los Angeles County, using similar methods (Fig. 1). Samples for dissolved inorganic nutrients were collected at Newport pier by filtering approximately 30 mL of water through a 0.2-μm syringe filter using syringes that were pre-washed with 5 % HCl and rinsed three times with sample water. Samples were dispensed into 50 mL polypropylene conical tubes and frozen at −20 °C upon arrival at the laboratory until analysis. Nutrient samples were not collected at the Redondo Beach pier location during the 2-year monitoring period. Concentrations of nitrate plus nitrite (0.2 μM limit of detection), nitrite (0.1 μM limit of detection), ammonium (0.1 μM limit of detection), phosphate (0.1 μM limit of detection), and silicate (1.0 μM limit of detection) were measured with±5 % precision on a QuikChem 8000 flow injection analyzer (Lachat Instruments; Loveland, CO, USA) by the Analytical Lab at the Marine Sciences Institute at University of California Santa Barbara. Samples for chlorophyll and particulate DA (pDA) concentrations were collected in duplicate by vacuum filtration of 100 mL and 200 mL samples, respectively, onto GF/F Whatman filters. Samples for determining the concentration of chlorophyll *a* were extracted in 100 % acetone for 24 h at −20 °C and analyzed on a calibrated laboratory fluorometer (TD-700; Turner Designs Inc, Sunnyvale, CA, USA) using the 5 % HCl acidification method for the correction for phaeopigment (Parsons et al. 1984). The use of a 100 % acetone extraction with 24 h storage at −20 °C has been proven to be equally as robust as the traditional 90 % acetone extraction with 24 h storage in a refrigerator (Caron 2001). Prior to analysis, pDA sample filters were extracted in 3 mL of 10 % methanol, sonicated for 30 s, and centrifuged for 10 min at 4,000 rpm. The resulting supernatant was analyzed using the Mercury Science Inc. DA Enzyme-Linked ImmunoSorbent Assay (ELISA; Durham, NC) following the methods described in Seubert et al. (2012). The detection limit for the ELISA assay used was 0.02 μg/L.

Subsamples for characterizing the microalgal community composition were preserved with 4 % formaldehyde and examined by inverted light microscopy at 400× after settling 25 mL in Utermöhl chambers for 24 h (Utermohl 1958). Forty fields of view were counted, giving a limit of detection of 3,000 cells/L. Samples were stored at room temperature in glass bottles, out of direct sunlight, until enumeration within 1 day to 1 week of collection. As a part of the SCCOOS HAB Monitoring Program, microalgal community analysis is focused on the identification of potential HAB formers known to be present in southern California waters. Organisms specifically identified are the dinoflagellates *Akashiwo sanguinea*, *Alexandrium* spp.,

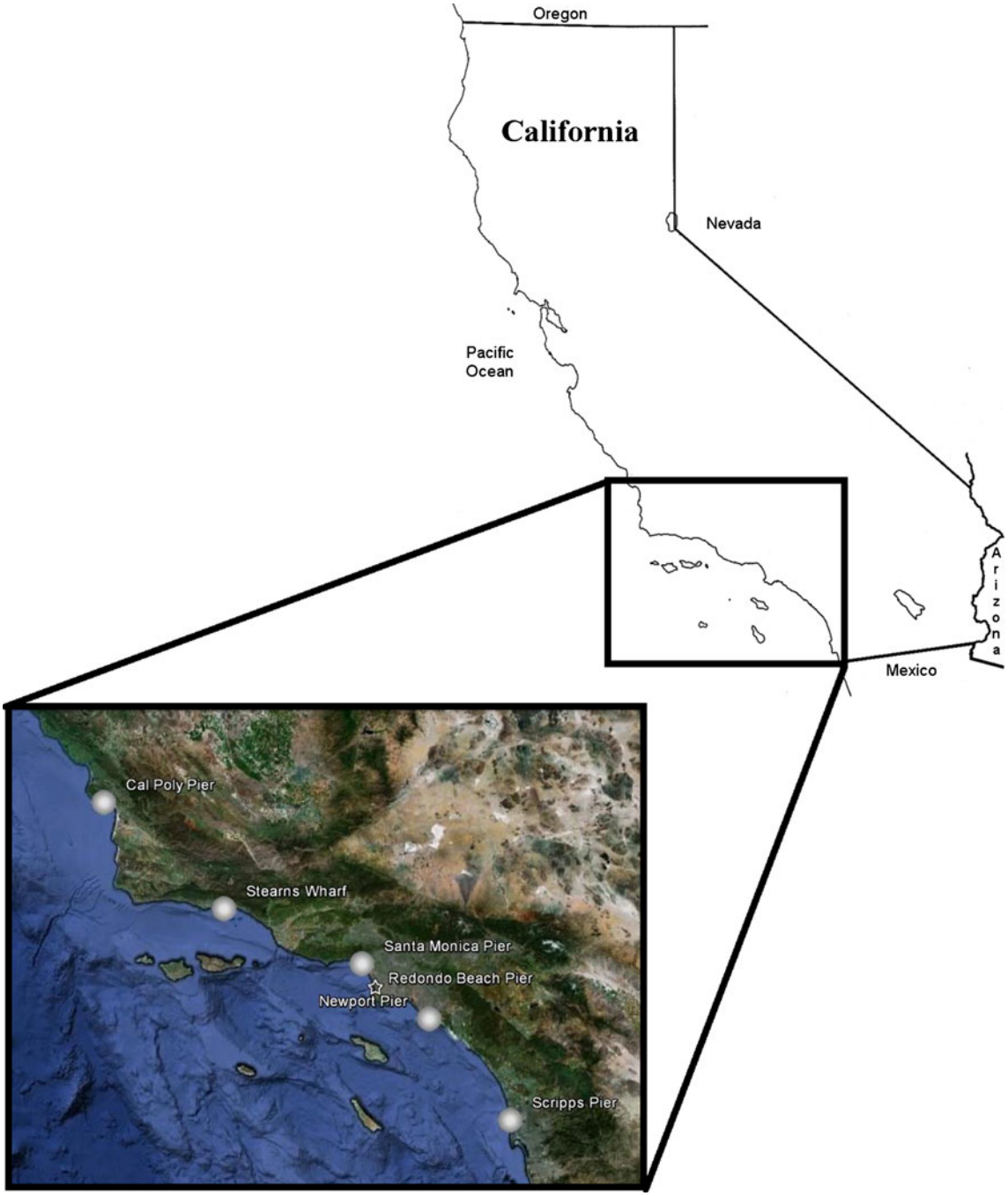


Fig. 1 Location of the SCCOOS weekly HAB monitoring piers in southern California, maintained through a collaboration of five university laboratories, beginning in June 2008. USC maintains the Newport pier sampling location in Orange County, CA. Redondo Beach pier (noted on the map with a *star*), located in Los Angeles County, CA, was added to the USC HAB monitoring effort in 2010

Dinophysis spp., *L. polyedrum*, and *Prorocentrum* spp. and the diatom genus *Pseudo-nitzschia*, all other cells in these classes counted were grouped into the categories "other dinoflagellate" or "other diatom." Other HAB organisms known to not preserve well that can be present, such as *Cochlodinium*, *Heterosigma*, and *Phaeocystis*, were recorded in the relative abundance determinations of live net tows from each location. Conclusive identification of *Pseudo-nitzschia* to species is not possible without using molecular methods (Scholin et al. 1996; Miller and Scholin 1998; Lundholm et al. 2002; Hubbard et al. 2008) or electron microscopy (Hasle et al. 1996; Hasle and Syvertsen 1997); consequently *Pseudo-nitzschia* cells were divided into two size classes, the *P. seriata* size class with frustule widths greater than 3 μm, and the *Pseudo-nitzschia delicatissima* size class with frustule widths smaller than 3 μm

(Hasle and Syvertsen 1997), which is easily accomplished by light microscopy.

Ancillary physical data

Rainfall data were obtained from the University of California Davis, Integrated Pest Management Program weather station at the Santa Ana Fire Station (NCDC # 7888, 33° 45′ N, 117° 52′ W) in Orange County, CA, for use with the Newport pier dataset and the Santa Monica weather station (CIMIS # 99, 34° 3′ N, 118° 29′ W) in Los Angeles County, CA, for use with the Redondo Beach pier dataset (www.ipm.ucdavis.edu). Rainfall data were collected as daily totals measured by an 8-in. diameter gauge and the information were binned into weekly totals. Daily river discharge data for the Santa Ana river was obtained from the US Geological Survey station #11078000 (33° 39′ N, 117° 54′ W) and binned into weekly totals for inclusion in the Newport pier dataset (http://waterdata.usgs.gov/nwis/). The weekly binning was performed by totaling information from the last sample date until the next sample collected, the majority of which were 7-day totals, with 8-day totals needing to be made when samples were collected on days other than Monday and 6-day totals when sampling returned to Monday collection. There is no major river that discharges in close proximity to the Redondo Beach pier location and therefore river discharge was not included in the Redondo dataset. Information on water temperature, salinity and chlorophyll fluorescence was collected at Newport pier by SCCOOS using automated sensors. The Newport pier automated station contains a Sea-Bird Electronics (Bellevue, WA, USA) 16plus SeaCAT conductivity and a temperature and pressure meter with measurements collected every 4 min. Temperature data collected by the Newport pier SCCOOS sensor package was obtained from the SCCOOS website (www.sccoos.org) for each day of sampling at the Newport pier. The temperature information from the automated sensor was verified by comparing sensor data to discrete temperature information manually collected in conjunction with discrete sample collection. Salinity data from the SCCOOS sensor package was discovered to be impacted by bio-fouling on the sensor when compared to salinity data collected by the SCCOOS manual shore station program at Newport pier. The manual shore station data were used in replacement of the flawed salinity sensor data in the regression analyses. The manual shore station program collects salinity data with the Guildline, model 8410, inductive salinometer and is available on the University of California San Diego Shore Station website (ftp://ftp.iod.ucsd.edu/shore/). Sensor data were not available for the Redondo Beach pier but temperature data were collected weekly simultaneous to discrete sample collection. Salinity information was obtained from a SCCOOS automated station located within Santa Monica Bay at the Santa Monica Pier, approximately 20 km north of the Redondo Beach pier.

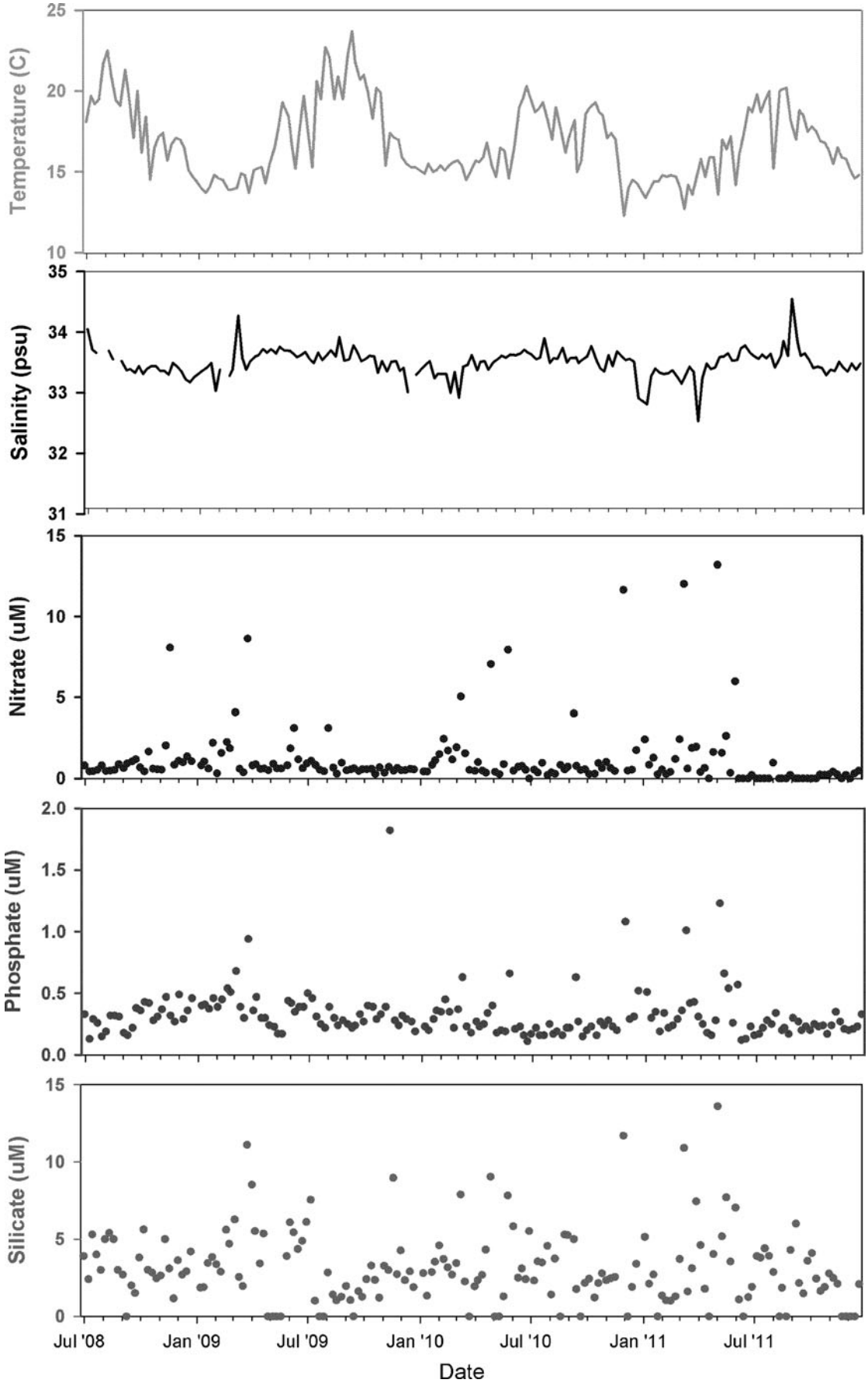
Statistical analyses

Simple linear and multiple linear regressions were performed in SigmaPlot (v. 11.0.0, Systat Software, Inc.) on both the Newport and Redondo Beach pier datasets. Multiple linear regression on $\log_{10}$-tranformed pDA+1 and *P. seriata*+1 concentrations was carried out using a backward step-wise approach in which independent variables were iteratively removed from the analysis based upon multi-collinearity (VIF >4), non-significant contributions ($p>0.05$) and low F values (<2). Multiple regressions of the Newport pier dataset were computed using the independent variables temperature, salinity, Santa Ana river discharge and rainfall, chlorophyll, nitrate, ammonium, nitrite, phosphate and silicate concentrations, and ratios of nutrient concentrations, silicate to nitrate, silicate to phosphate, and phosphate to nitrate. All independent variables failed tests for normality; temperature, salinity, chlorophyll, ammonium, and phosphate were $\log_{10}$-transformed while the remaining variables Santa Ana river discharge, rainfall, nitrate, nitrite, silicate, silicate to nitrate ratio, silicate to phosphate ratio, and the phosphate to nitrate ratio were $\log_{10}$-transformed after an addition transform of 1. Multiple regressions of the Redondo Beach pier dataset were computed using the independent variables temperature, salinity, rainfall and chlorophyll. Temperature, salinity, and chlorophyll were $\log_{10}$-transformed and rainfall was $\log_{10}$-transformed following an addition transform of 1. Regressions were computed with no time lag of environmental variables as well as with 1 and 2-week time lagging of environmental variables, to investigate a possible delay in the biological response to environmental forcing factors. Chlorophyll concentrations were included within the regression analysis to investigate a potential relationship between overall microalgal biomass and DA events. The adjusted R^2 values for each regression were used to compare the non-lagged, as well as 1 and 2-week lagged multiple regressions. The adjusted R^2 takes into account sample size and variable number, weakening the likelihood of R^2 being artificially maximized by the inclusion of non-significant factors.

Results

Environmental conditions at Newport and Redondo beach piers

Temperatures recorded by the SCCOOS automated sensor at Newport pier ranged from 12.3 to 23.7 °C, with an average temperature of 16.9±2.33 °C , during the 3.5 year monitoring period (Fig. 2). A smooth seasonal trend of warming during the summer months and cooling during the winter was interrupted


Temperature (C)
25
20
15
10
Salinity (psu)
35
34
33
32
31
Nitrate (uM)
15
10
5
0
Phosphate (uM)
2.0
1.5
1.0
0.5
0.0
Silicate (uM)
15
10
5
0
Jul '08
Jan '09
Jul '09
Jan '10
Jul '10
Jan '11
Jul '11
Date

◀ **Fig. 2** Temperature and salinity measured at Newport pier with the SCCOOS automated sensor is plotted from June 30, 2008 to December 19, 2011. Nitrate, phosphate, and silicate measured in the discrete samples from the same time period are also plotted

by sharp decreases in temperature due most likely to storm events and upwelling in the region. Salinity ranged from 32.5 to 34.6 psu, with an average salinity of 33.5±0.23 psu (Fig. 2). Concentrations of nitrate, phosphate and silicate demonstrated numerous, sporadic increases, with the highest concentrations often coinciding with decreases in temperature due to the presence of upwelled water (Fig. 2). Detectable nitrate concentrations ranged from 0.21 to 13 μM, with an overall average concentration of 1.2±2.0 μM. Detectable nitrite concentrations ranged from 0.10 to 0.96 μM, with an overall average concentration of 0.13±0.16 μM. Ammonium concentrations were always above the detection limit of the method (see Materials and Methods) and ranged from 0.21 to 31.6 μM, with an overall average concentration of 3.19±4.31 μM. Phosphate concentrations were always above the detection limit of the method used (see Materials and Methods) and ranged from 0.11 to 1.8 μM, with an overall average concentration of 0.32± 0.20 μM. Detectable silicate concentrations ranged from 1.0 to 14 μM, with an average concentration of 3.1±2.4 μM. Temperatures recorded in the weekly discrete sampling at Redondo Beach pier ranged from 13.2 to 22.0 °C, with an average temperature of 17.2±2.33 °C, during the two year monitoring period (Fig. 3). Salinity measurements taken from the SCCOOS automated sensor on Santa Monica pier, approximately 20 km north of Redondo Beach pier, ranged from 31.3 to 33.4 psu, with an average concentration of 32.8±0.47 psu (Fig. 3).

Establishing bloom conditions based on chlorophyll concentrations

Bloom definitions (major and minor) were investigated in this study for the Newport and Redondo Beach piers, applying constant chlorophyll *a* bloom thresholds established from chlorophyll data specific to each location. A long-term mean of chlorophyll *a* concentration from weekly discrete samples was calculated from the entire dataset for each location in this study. Newport pier exhibited an overall average chlorophyll *a* concentration of 4.31±4.31 μg/L during the 3.5-year monitoring period, while Redondo Beach pier had an average concentration of 2.84±3.14 μg/L during the two year monitoring period. A major bloom threshold specific to Newport pier was defined from the discrete chlorophyll *a* data as two standard deviations over the long term mean, or 12.9 μg/L (Table 1A). A major bloom threshold for Redondo Beach pier was defined as 9.13 μg/L based on a similar definition for the chlorophyll data at that site (Table 2A).

Minor bloom thresholds for each dataset were established by first removing all chlorophyll values above the major bloom threshold (to remove the influence of those extreme values on the mean) and then the new average chlorophyll *a* concentrations were determined. The average chlorophyll concentration for Newport pier after the removal of the major bloom values was 3.88±2.93 μg/L, and a minor bloom

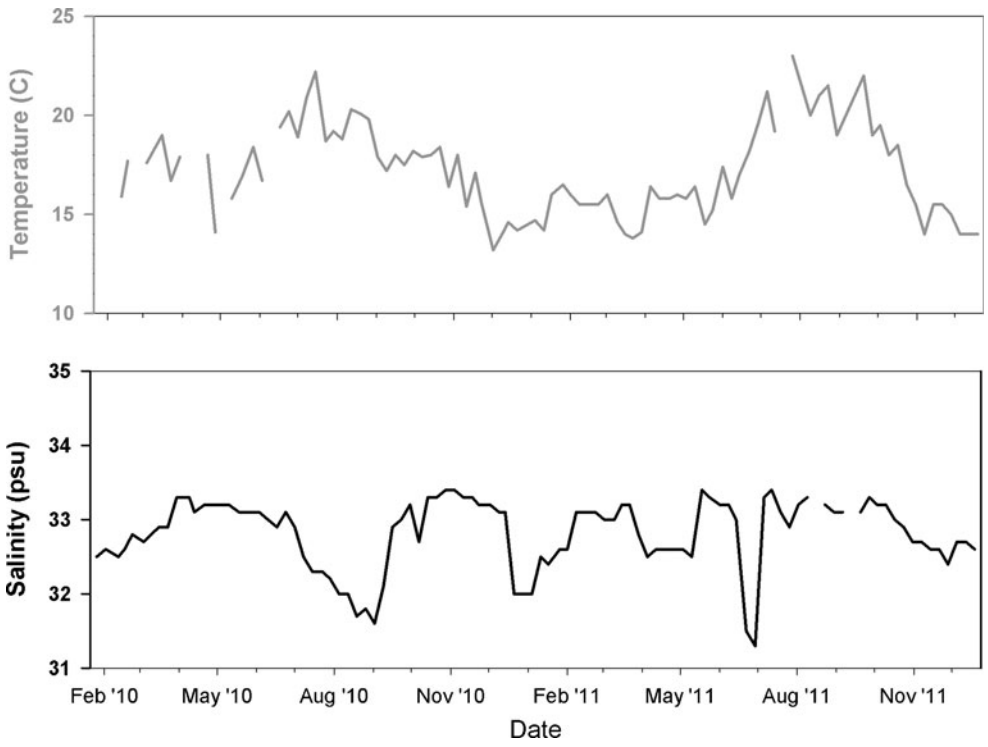


Fig. 3 Temperature measured during the discrete sampling at Redondo Beach Pier from January 26, 2010 to December 19, 2011 is plotted in conjunction with the salinity measurements taken with the SCCOOS automated sensor at Santa Monica Pier during the same period

Table 1 Information on major and minor bloom events identified at Newport pier (A) using chlorophyll *a* concentrations to define blooms and (B) defined by abundances of cells in the *P. seriata* size class from *Pseudo-nitzschia* cell counts

	Major blooms	Minor blooms
(A) Using chlorophyll *a* concentrations to define blooms		
Threshold	12.9 μg/L	9.74 μg/L
No. identified	4	12
% Occurrence	2 %	7 %
No. co-occurring with [pDA]	1	3
% with [pDA]	25 %	25 %
Total bloom samples	16	
Total % with [pDA]	25 %	
(B) Defined by abundances of cells in the *P. seriata* size class from *Pseudo-nitzschia* cell counts		
Threshold	88,000 cells/L	40,000 cells/L
No. identified	8	12
% Occurrence	4 %	7 %
No. co-occurring with [pDA]	5	8
% with [pDA]	63 %	67 %
Total Bloom Samples	20	
Total % with [pDA]	65 %	

threshold of 9.74 μg/L was defined as two standard deviations above that average (Table 1A). The average chlorophyll *a* concentration for the Redondo Beach pier dataset after removal of the major bloom values was 2.32±1.67 μg/L, yielding a minor bloom threshold of 5.66 μg/L (Table 2A). These treatments of the datasets enabled identification of four major and twelve minor blooms at the Newport pier site for a total of 16 bloom events during the 3.5 year dataset (Table 1A; Fig. 4a). Four major and four minor blooms events were identified at Redondo Beach pier for a total of 8 bloom samples in the 2 year dataset (Table 2A; Fig. 5a).

Major and minor bloom events defined by anomalously high concentrations of chlorophyll (Figs. 4a and 5a) were compared to detectable concentrations of pDA in the same samples to examine the potential for identifying toxic *Pseudo-nitzschia* spp. blooms based on total microalgal biomass (i.e. chlorophyll concentration). Detectable concentrations of pDA in the Newport pier dataset occurred during only one major bloom and three minor bloom events, corresponding to 25 % of the samples identified as blooms (Table 1A; Fig. 4b). Particulate DA was not detected in any of the eight samples identified as blooms in the Redondo Beach pier dataset based on chlorophyll concentrations (Table 2A; Fig. 5b).

Table 2 Information on major and minor bloom events identified at Redondo Beach pier (A) using chlorophyll *a* concentrations to define blooms and (B) based upon the abundance of *P. seriata* size class cells from *Pseudo-nitzschia* cell counts

	Major blooms	Minor blooms
(A) Using chlorophyll *a* concentrations to define blooms		
Threshold	9.13 μg/L	5.66 μg/L
No. identified	4	4
% Occurrence	4 %	4 %
No. co-occurring with [pDA]	0	0
% with [pDA]	0 %	0 %
Total bloom samples	8	
Total % with [pDA]	0 %	
(B) Based upon the abundance of *P. seriata* size class cells from *Pseudo-nitzschia* cell counts		
Threshold	110,000 cells/L	56,000 cells/L
No. identified	4	7
% Occurrence	4 %	7 %
No. co-occurring with [pDA]	2	2
% with [pDA]	50 %	29 %
Total bloom samples	11	
Total % with [pDA]	36 %	

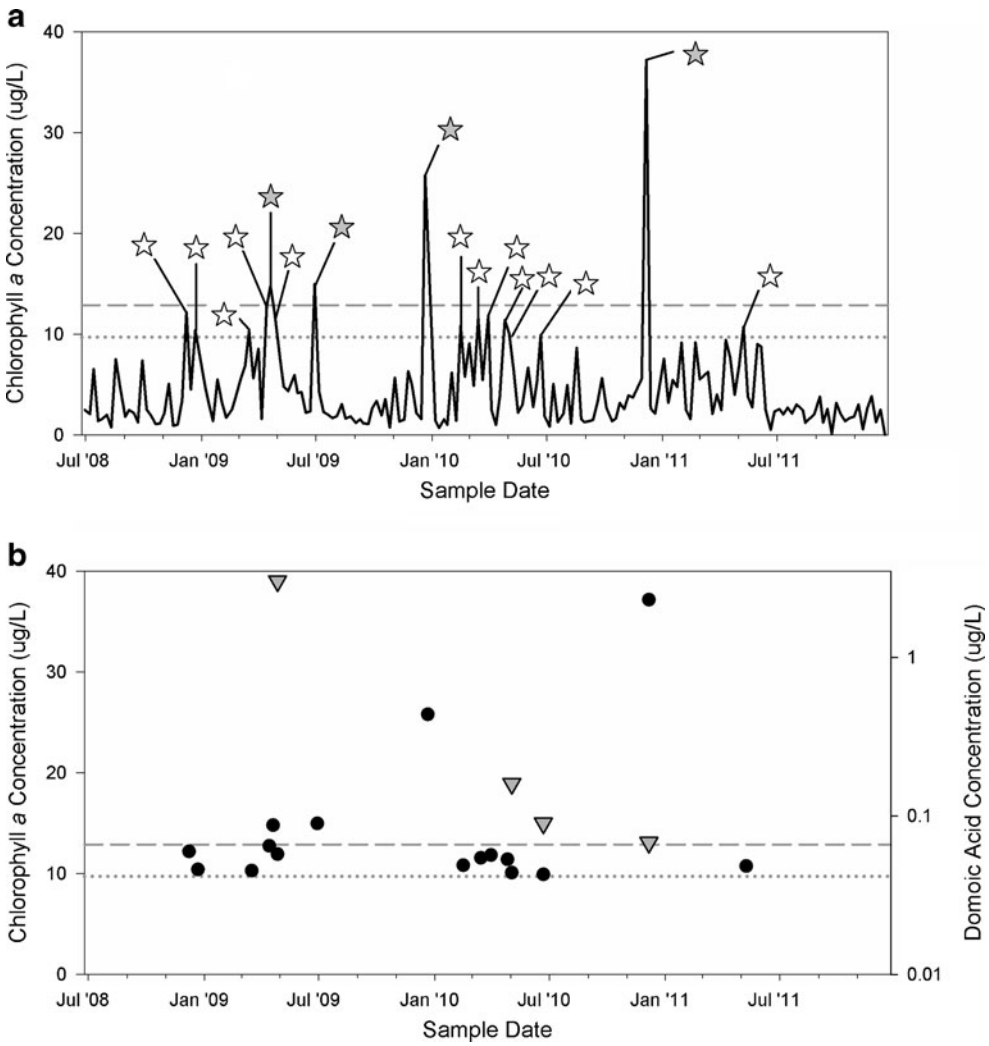


Fig. 4 **a** Chlorophyll *a* concentrations measured in the weekly samples collected at Newport pier from June 30, 2008 to December 19, 2011. The major bloom threshold chlorophyll concentration of 12.9 μg/L and minor bloom threshold of 9.74 μg/L are shown by the *dashed* and *dotted lines*, respectively. The 4 major blooms are marked by the *gray stars* and the 12 minor blooms are marked by the *white stars*. **b** All samples that were not identified as major or minor blooms by the chlorophyll concentration definition were removed, and only bloom sample chlorophyll concentrations are plotted with the *black circles*. The major and minor bloom thresholds are shown by the *dashed* and *dotted lines*, respectively. pDA concentrations measured in any of the chlorophyll defined major and minor are plotted with the *light gray triangles*; pDA samples below the detection limit were not included

Establishing bloom conditions based on *Pseudo-nitzschia* cell abundances

Many HAB species do not need to attain exceedingly high cell abundances or dominate total microalgal biomass in order to cause ecosystem damage and present a human health risk. Thus, it was not surprising to find that a chlorophyll-based definition for blooms at our coastal sites poorly identified situations in which pDA production by *Pseudo-nitzschia* spp. was detectable. Correlations with cell abundances of *Pseudo-nitzschia* were investigated as an alternative means of identifying potential emerging DA events.

Pseudo-nitzschia spp. abundances were divided into two size classes in the cell count data collected for the SCCOOS HAB Monitoring Program; a *P. seriata* size class and a *P. delicatissima* size class (see Methods and Materials). The *P. seriata* size class contains species of *Pseudo-nitzschia* that are known to be capable of DA production. Documented toxic events in southern California have been most often attributed to *P. australis* and *P. multiseries*, both members of the *P. seriata* size class (Anderson et al. 2006; Busse et al. 2006; Schnetzer et al. 2007, Schnetzer et al. 2012). The *P. delicatissima* size class contains some species capable of toxin production; however, blooms of this size class are rarely associated with toxic events (Smith et al. 1991; Adams et al. 2000; Stehr et al. 2002; Orsini et al. 2004).

Cellular abundance data collected for the *P. seriata* size class at the Newport and Redondo Beach piers were used to establish major and minor *Pseudo-nitzschia* bloom events in a manner analogous to that used with the chlorophyll *a* concentrations, described above. Long-term means of *P. seriata* size class cell abundance data were calculated for each location and major bloom thresholds were established based on two standard deviations from the overall mean values for each sampling site. A long-term mean for the Newport pier samples was calculated as 14,000± 37,000 cells/L, yielding a major bloom threshold of 88,000 cells/L of the *P. seriata* group (Table 1B). After removal of the major blooms values, a new long term mean

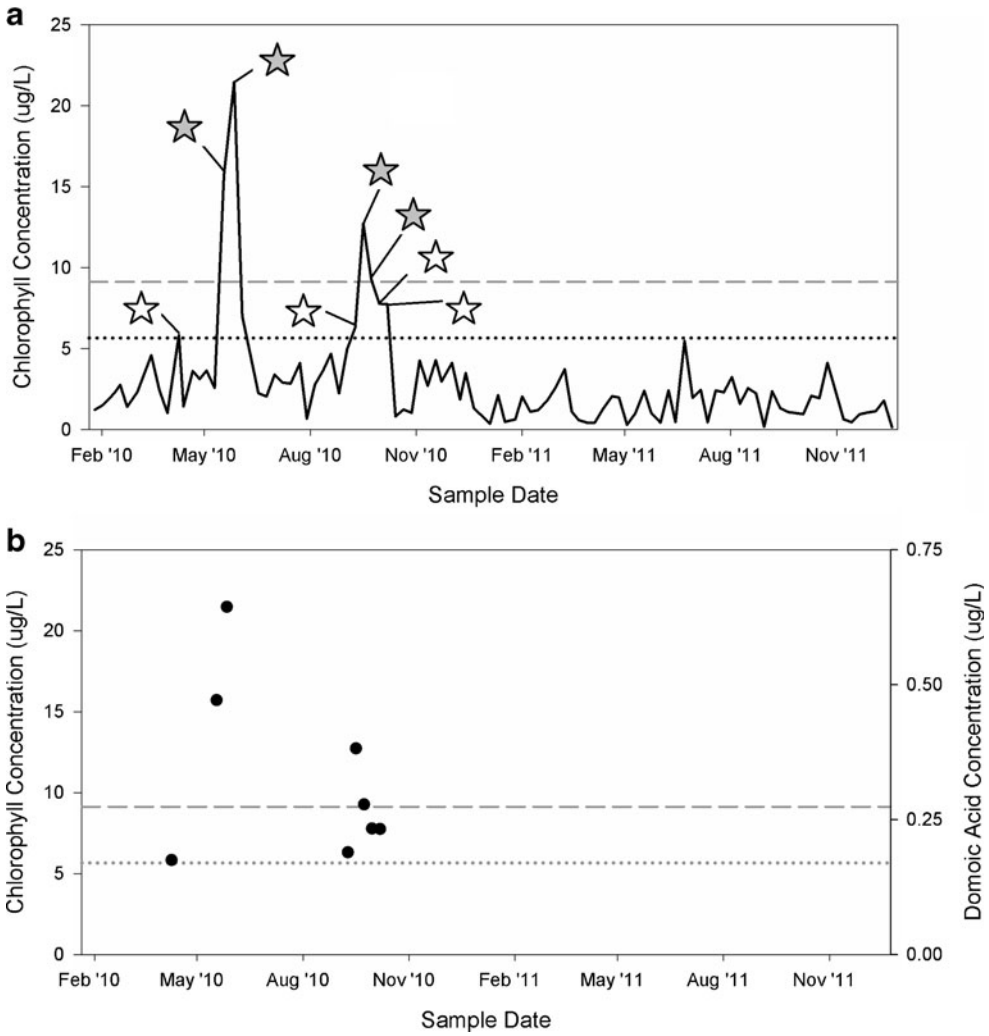


Fig. 5 **a** Chlorophyll *a* concentrations measured in the weekly samples collected at Redondo Beach pier from January 26, 2010 to December 19, 2011. The major bloom threshold chlorophyll concentration of 9.13 μg/L and minor bloom threshold of 5.66 μg/L are shown by the *dashed* and *dotted lines*, respectively. The four major blooms are marked by the grey stars and the four minor blooms are marked by the *white stars*. **b** All samples that were not identified as major or minor blooms by the chlorophyll concentrations definition were removed and only bloom sample chlorophyll concentrations are plotted. The major and minor bloom thresholds are shown by the *dashed* and *dotted line*, respectively. Domoic acid was not measured in any of the chlorophyll bloom samples from Redondo Beach Pier

was determined to be 7,100±16,000 cells/L, creating a minor bloom threshold of 40,000 cells/L of the *P. seriata* group. The long-term mean for the Redondo Beach pier samples was determined to be 19,000±46,000 cells/L, yielding a major bloom threshold of 110,000 cells/L of the *P. seriata* group (Table 2B). The long term mean for these samples following removal of the major bloom samples was 11,000±22,500 cells/L, yielding a minor bloom threshold of 56,000 cells/L.

These treatments of the cell abundance data identified eight major and twelve minor blooms of the *P. seriata* size class at the Newport pier sampling site for a total of twenty bloom samples (Table 1B; Fig. 6a), and four major and seven minor blooms at the Redondo Beach pier sampling site for a total of eleven bloom samples (Table 2B; Fig. 7a). The bloom events identified using the abundances of the *P. seriata* size class were compared to pDA concentrations at each location (Figs. 6b and 7b). Thirteen of the Newport pier samples identified as blooms corresponded to samples with detectable pDA concentrations (Table 1B), while 4 of the 11 Redondo Beach pier samples identified as blooms had detectable pDA concentrations (Table 2B). Bloom events at the Newport pier site, defined using the abundances of the *P. seriata* size class, corresponded to detectable pDA concentrations in 65 % of the samples (Table 1B) compared to 25 % correspondence for a bloom definition based on anomalously high chlorophyll concentrations (Table 1A). Measureable pDA concentrations occurred in 29 of the 178 total samples (16 %) collected at Newport pier and 13 of the 29 (45 %) were detected during blooms of the *P. seriatia* size class. The samples with detectable pDA that were not identified as blooms ranged from 0.060 to 0.34 μg/L, with an average concentration of 0.075±0.073 μg/L, while the samples corresponding to identified blooms had pDA concentrations that ranged from 0.046 to 3.0 μg/L, with an average concentration of 0.35±0.82 μg/L.

Bloom events at the Redondo Beach pier site, defined using the abundances of the *P. seriata* size class, corresponded to detectable pDA concentrations in 36 % of the bloom samples (Table 2B) compared to 0 % correspondence for a bloom definition based upon anomalously high chlorophyll concentrations (Table 2A). Measurable pDA concentrations

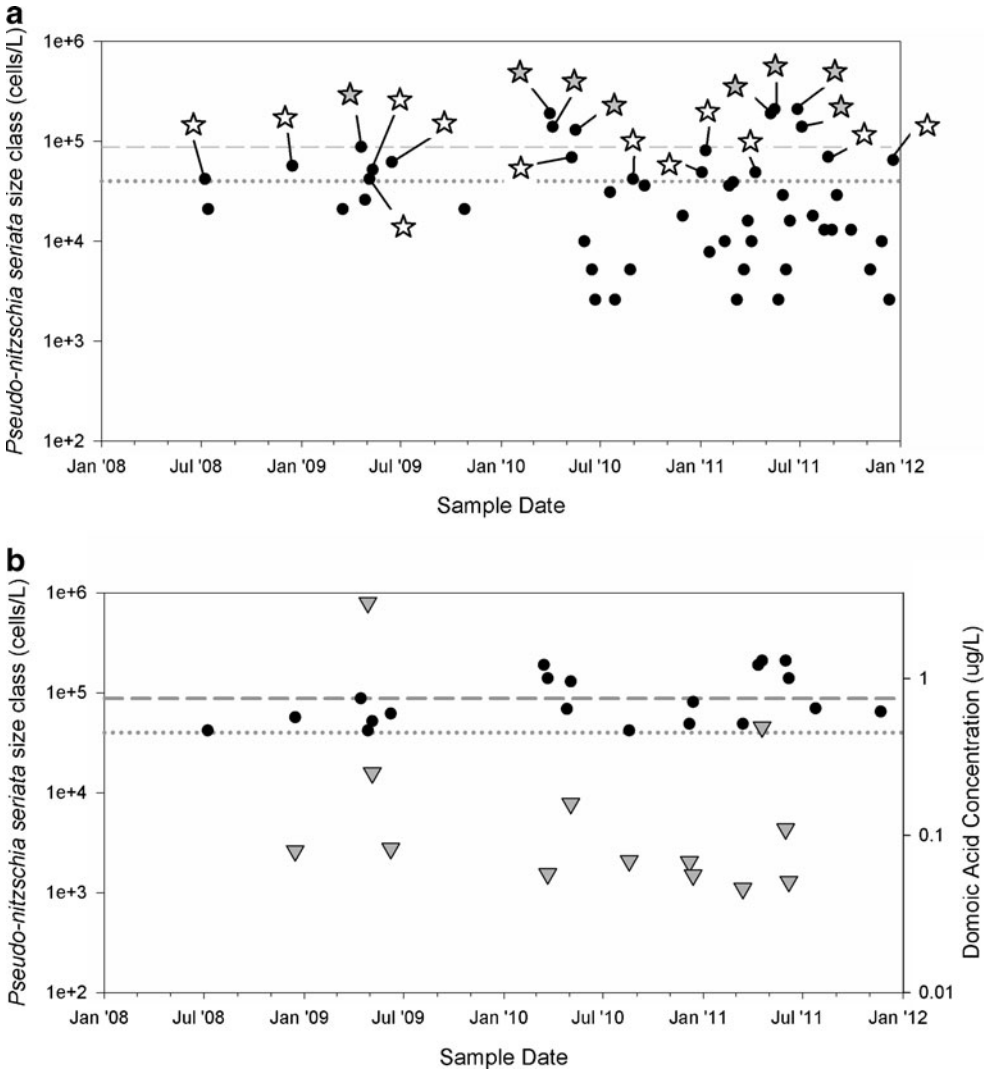


Fig. 6 **a** The concentration of *Pseudo-nitzschia seriata* size class cells measured in the Newport pier weekly samples from June 30, 2008 to December 19, 2011 are plotted in the *top panel figure*. The major bloom threshold concentration of 88,000 cells/L and minor bloom threshold of 40,000 cells/L are plotted as the *dashed* and *dotted lines*, respectively. The 8 major blooms are marked by the *gray stars* and the 12 minor blooms are marked by the *white stars*. **b** Only the identified blooms by the *P. seriata* size class definition, major and minor, are plotted in the *bottom panel with black circles* and the associated pDA concentrations measured in the same sample are plotted as the *gray triangles*; pDA samples below the detection limit were not included. The major and minor bloom thresholds are plotted as the *dashed* and *dotted lines*, respectively

occurred in 23 of the total 98 samples (23 %) collected at the Redondo Beach sampling site, and 4 of the 23 (17 %) were detected during blooms of the *P. seriata* size class. The samples with detectable pDA that were not identified as blooms ranged in concentration from 0.048 to 0.30 μg/L, with an average concentration of 0.082±0.069 μg/L, and the bloom associated pDA concentrations range from 0.041 to 0.57 μg/L, with an average concentration of 0.21±0.25 μg/L.

Correlating *Pseudo-nitzschia* blooms and DA events to environmental variables

Multiple and simple linear regression analyses were carried out on each of the complete datasets from Newport and Redondo Beach piers and then compared to regression analysis on a subset of samples defined as blooms of the *P. seriata* size class at each location. The comparison of the regression results was performed to discern if the identification of bloom events using the abundances of *Pseudo-nitzschia* in the *P. seriata* size class improved the specificity of statistical analysis by eliminating samples that were unrelated to *Pseudo-nitzschia* or toxic events.

The first set of regression analyses on the entire 3.5 year Newport pier dataset were undertaken to identify significant relationships between ammonium, chlorophyll *a*, nitrate, nitrite, phosphate, and silicate concentrations, ratios of nutrient concentrations for silicate to nitrate, silicate to phosphate, and phosphate to nitrate, Santa Ana river discharge, rainfall, temperature and salinity to pDA and *P. seriata* size class concentrations. The most significant relationships identified on the dataset without time lagging were obtained with multiple linear regressions. Salinity, chlorophyll *a* concentrations and the ratio of silicate to phosphate concentrations were significantly correlated, albeit weakly, with pDA concentrations (R^2 adj=0.0844, $p<0.001$). The salinity and chlorophyll values were positively correlated with pDA and silicate to phosphate ratios were negatively correlated with pDA. A significant and slightly stronger relationship was identified between temperature, chlorophyll and nitrite concentrations, and ratios of silicate to nitrate with *P. seriata*

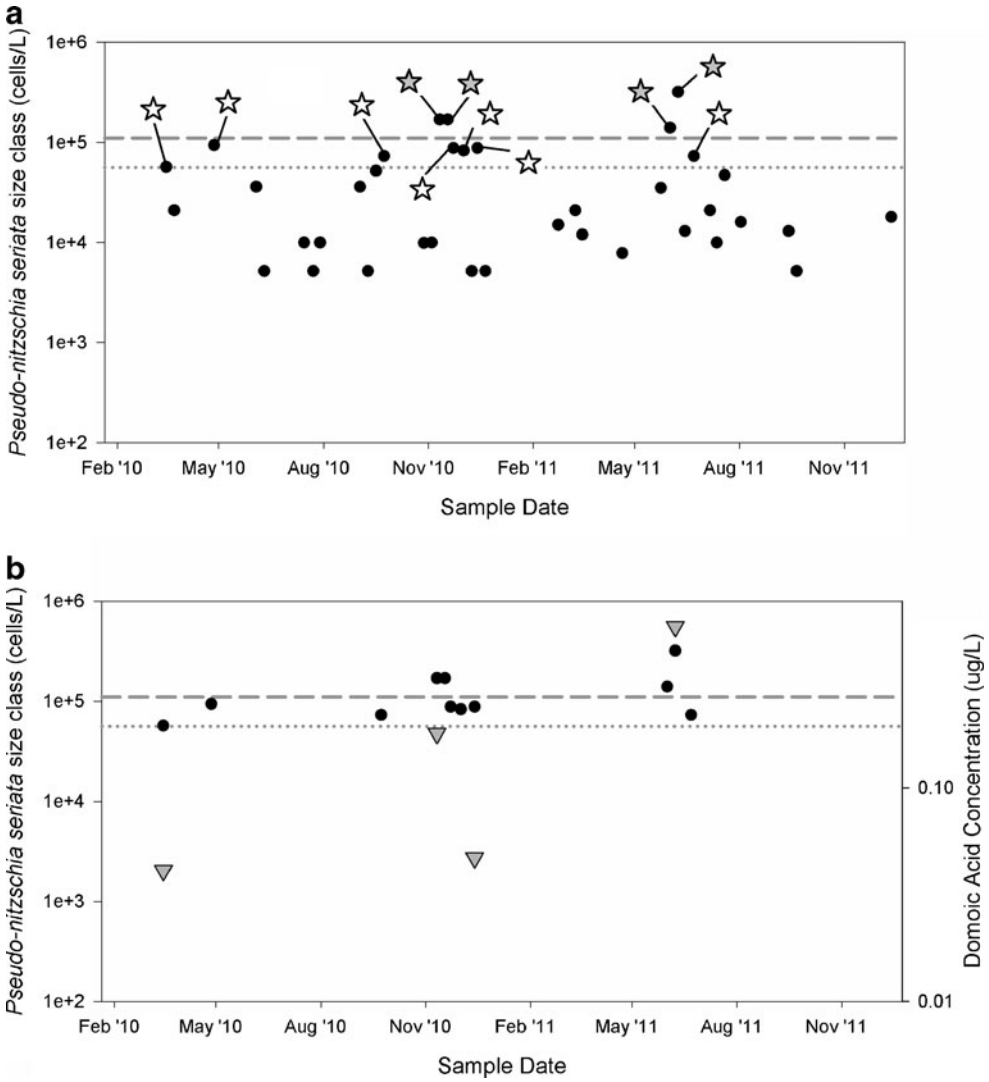


Fig. 7 **a** The concentration of *Pseudo-nitzschia seriata* size class cells measured in the Redondo Beach pier weekly samples from January 26, 1010 to December 19, 2011, are plotted. The major bloom threshold concentration of 110,000 cells/L and minor bloom threshold of 56,000 cells/L are plotted as the *dashed* and *dotted line*, respectively. The four major blooms are marked by the *gray stars* and the minor blooms are marked by the *white stars*. **b** Only the identified blooms by the *P. seriata* size class definition, major and minor, are plotted as the *black circles* with the associated domoic acid concentrations measured in the same sample plotted as the *gray triangles*; pDA samples below the detection limit were not included. The major and minor bloom thresholds are plotted as the *dashed* and *dotted lines*, respectively

size class concentrations (R^2 adj=0.193, $p<0.001$). Temperature, nitrite concentration and the ratio of silicate to nitrate concentrations were negatively correlated with *P. seriata* whereas chlorophyll was positively correlated. A 1-week time lag of environmental data revealed a significant relationship between temperature, salinity, chlorophyll concentration and the ratio of silicate to nitrate with pDA concentrations (R^2 adj=0.0676, $p=0.007$). Temperature was negatively correlated to pDA while salinity, chlorophyll and the ratio of silicate to nitrate were positively correlated with pDA. The same parameters were significantly correlated with *P. seriata* (R^2 adj=0.171, $p<0.001$), temperature was

Table 3 Results of multiple regression analysis of the Newport pier dataset

Variable	Time lag	R^2 adj	F	P	Significant variables
pDA concentration	None	0.125	3.707	0.07	− Silicate/Phosphate
	1 week	0.537	6.228	0.004	Chlorophyll, Salinity, Silicate/Nitrate, − Temperature
	2 weeks	0.113	2.078	0.16	Salinity, Chlorophyll
P. seriata Abundance	None	0.613	8.904	.002	− Ammonium, − Phosphate/Nitrate, −Temperature
	1 week	0.242	2.913	0.069	− Ammonium, − Phosphate/Nitrate, − Salinity
	2 weeks	0.653	8.997	0.001	Rainfall, − Salinity, Silicate, Temperature

Prior to analysis, samples were identified as blooms based upon the *P. seriata* size class abundance definition and all non-bloom values were removed. Negative correlations are identified with a minus sign (−)

negatively correlated and salinity, chlorophyll and the ratio of silicate to nitrate were positively correlated with *P. seriata*. A 2-week time lag of environmental data revealed a significant positive correlation between chlorophyll and the silicate to nitrate ratio with pDA concentrations (R^2 adj=0.0559, p=0.005). Temperature was negatively correlated with *P. seriata* and the silicate to nitrate ratio was positively correlated with *P. seriata* with a 2-week time lag (R^2 adj=0.615, p=0.001).

Bloom events identified by anomalously high abundances of the *P. seriata* size class were then used to select a subset of data for analysis. Samples not identified as blooms were removed in an effort to improve specificity of the statistical analysis. All identified major (8) and minor (12) blooms of the *P. seriata* size class were used in regression analyses investigating relationships between ammonium, chlorophyll, nitrate, phosphate and silicate concentrations, ratios of nutrient concentrations silicate to nitrate, silicate to phosphate and phosphate to nitrate, Santa Ana river discharge, rainfall, temperature and salinity to pDA concentrations and *P. seriata* size class abundances (Table 3). The most significant relationship identified for pDA concentrations was with 1-week-lagged temperature, salinity, chlorophyll, and silicate to nitrate ratio (R^2 adj=0.537, p=0.004). Temperature was negatively correlated while salinity, chlorophyll, and silicate to nitrate ratio were positively correlated with pDA. Salinity and chlorophyll concentrations were also positively correlated with pDA concentrations with a 2-week lag (R^2 adj=0.113, p=0.16), although the relationship was less significant than with a 1-week time lag. Without time lagging of the data, the most significant relationship identified was with a simple linear regression and a negative correlation between the silicate to phosphate ratio and pDA concentrations (R^2 adj=0.125, p=0.07). The most significant relationship identified in linear regressions with *P. seriata* size class abundances was with 2-week time lagged temperature, salinity, rainfall and silicate concentration information (R^2 adj=0.653, p=0.001). Salinity was negatively correlated while temperature, rainfall and silicate concentrations were positively correlated with *P. seriata* abundances.

The initial regression analyses performed with the entire 2-year Redondo Beach pier dataset investigated relationships between chlorophyll concentrations, rainfall, temperature and salinity to pDA concentrations. Regressions failed to produce statistical significance between any variable (without time lagging) and pDA concentrations, as demonstrated by high p values (>0.05), low F values (<2) and low power values. F values improved using a 1 and 2-week time lag of variables, but p values remained insignificant (>0.05). Regressions with the same variables to abundance of the *P. seriata* size class revealed a significant positive relationship with chlorophyll concentration (R^2 adj=0.0386, p=0.032), although the relationship had very weak predictive power.

Samples from the Redondo Beach pier dataset meeting the criteria of a bloom event defined by the abundances of *Pseudo-nitzschia* in the *P. seriata* size class were also examined for relationships to environmental variables. Multiple regressions were performed using the subset including major (4) and minor (7) blooms to investigate relationships between pDA concentrations and chlorophyll concentrations, rainfall, temperature, and salinity (Table 4). Negative correlations between chlorophyll concentration, lagged 1 and 2 weeks prior to pDA concentrations, were identified but lacked significance (p=0.155 and 0.050, respectively). Regressions were repeated with the same environmental variables to investigate relationships with abundances of *Pseudo-nitzschia* in the *P. seriata* size class. The only significant relationship was identified from a simple linear regression between the *Pseudo-nitzschia* abundances and salinity values lagged 1 week (R^2 adj=0.358, p=0.03).

Discussion

The goals of the present study were: (a) to employ a long-term, weekly dataset of microalgal biomass, community composition and pDA concentrations to establish site-specific, quantitative metrics to identify emerging blooms of potentially toxic species of the diatom genus *Pseudo-nitzschia* and DA events resulting

Table 4 Results of multiple regression analysis of the Redondo Beach pier dataset

Variable	Time lag	R^2 adj	F	P	Significant variables
pDA Concentration	None	**	**	**	**
	1 week	0.124	2.416	0.155	− Chlorophyll
	2 weeks	0.293	5.144	0.050	− Chlorophyll
P. seriata Abundance	None	**	**	**	**
	1 week	0.358	6.581	0.03	Salinity
	2 weeks	**	**	**	**

Prior to analysis, samples were identified as blooms based upon the *P. seriata* size class abundance definition and all non-bloom values were removed. Negative correlations are identified with a minus sign (−). Asterisks denote failed regressions without statistical significance, low F values (<2) and low power values

from these species; (b) to establish the relationship between blooms and DA events with the intent of identifying easily-acquired measurements that might indicate an emerging HAB event; and (c) to provide an investigative approach for relating *Pseudo-nitzschia* blooms and DA events to chemical and physical environmental parameters co-occurring or preceding these events.

Defining blooms and their relation to DA events

The ability to rapidly identify an emerging HAB event is a prerequisite for preventing human exposure and for taking steps that might minimize or mitigate potential ecological impacts. Unfortunately, microalgal blooms have been rather arbitrarily defined, or have been defined based on the particular harmful algal species involved. Microalgal blooms have generally been defined based on total algal biomass, often using chlorophyll concentration (either extracted chlorophyll analyses or *in vivo* chlorophyll fluorescence) as a proxy for algal biomass. Only a few studies have taken a rigorous, quantitative approach to defining blooms using this parameter (Carstensen et al. 2007; Henson and Thomas 2007; Allen et al. 2008; Kim et al. 2009). Moreover, only some HAB species dominate the microalgal assemblage so strongly that total microalgal biomass is a reasonable proxy for their abundance. Species in this latter category include massive blooms of the nuisance brown tide algae, *Aureococcus anophagefferens* and *Aureoumbra lagunensis*, the dinoflagellates *L. polyedrum*, or the toxic flagellate *Prymnesium parvum* that can constitute most of the algal pigment present in water samples collected during blooms of these species (Allen 1938; Omand et al. 2011; Roelke et al. 2011; Gobler and Sunda 2012). Blooms of *A. anophagefferans*, for example, have been defined based on cell abundances, indicating abundances at which this species dominates the pelagic food web and poses the potential for ecological damage (Gastrich and Wazniak 2002). In contrast, species of the dinoflagellates genus *Alexandrium* that produce paralytic shellfish toxins can constitute a significant human health risk even at relatively low abundances (i.e. a small percentage of the total biomass of the microalgal community in a sample).

Defining microalgae blooms based on anomalously high chlorophyll concentrations (two standard deviations above a long-term mean) indicated the presence of numerous major or minor bloom events over the 3.5 or 2-year period of observation at the Newport pier or Redondo Beach pier, respectively (Tables 1A and 2A; Figs. 4a and 5a). While these events indicate significant increases in microalgal biomass at these sites and times, blooms defined in this manner were very poor predictors of detectable concentrations of pDA (Tables 1B and 2B; Figs. 4b and 5b). Blooms defined on anomalously high abundances of the *P. seriata* size class of *Pseudo-nitzschia* spp. also allowed for the identification of numerous bloom events, with blooms defined in this manner yielding greater correspondence to samples that also exhibited detectable concentrations of pDA (Tables 1B and 2B; Figs. 6b and 7b). Documented DA events during blooms of *Pseudo-nitzschia* spp. in the southern California region have been attributed to members of the *P. seriata* size class, most commonly to *P. multiseries* and *P. australis* (Anderson et al. 2006; Schnetzer et al. 2007; Lewitus et al. 2012, Schnetzer et al. 2012). The history of the *P. seriata* size class involvement in DA events in our region was the motivation for focusing on this size class for the basis of an emerging event definition.

Developing a simple *P. seriata* abundance-based metric to establish the presence of a potentially emerging *Pseudo-nitzschia* bloom and DA event would reduce the time and cost that is inherent in more sophisticated, albeit more informative, approaches. Technology is only now becoming available for *in situ* measurements of DA. The Environmental Sample Processor (ESP) is capable of quantifying pDA concentrations in situ and is also capable of identifying a suite of *Pseudo-nitzschia* species that might be the source of the toxin (Greenfield et al. 2006; Greenfield et al. 2008; Doucette et al. 2009). However, the cost and maintenance logistics of the instrument are presently prohibitive for most scientific studies, routine monitoring and regulatory programs. Alternatively, direct toxin measurements performed on microalgal samples returned to a laboratory can also provide direct confirmation of the presence and quantities of pDA in samples, but toxin analysis by ELISAs, high performance liquid chromatography or liquid chromatography/mass spectrometry require fairly extensive sample preparation and thus significant time between sample collection and the availability of the data. Emerging molecular approaches, such as microarray-based technologies, for quantifying HAB species will provide speed and power in time and our approach will provide some bounding on where and when to sample intensively for their application (Ahn et al. 2006; Gescher et al. 2008; Kegel et al. 2012).

Until these more-insightful methods and instruments become easily accessible and less costly, the ability to infer an emerging DA event based upon unusual increases in the abundance of the *P. seriata* size class of *Pseudo-nitzschia* cells provides a crude, yet useful and relatively rapid assessment of developing events that can applied to large sample sets. Moreover, continued observations and documentation of blooms of *Pseudo-nitzschia* at Newport and Redondo Beach piers and other coastal sites along the California coast will augment the datasets that are presently available in this region, allowing for more accurate differentiation of toxic and non-toxic events. Defining a site-specific *Pseudo-nitzschia* bloom metric was also motivated by a desire to improve our understanding of the environmental forcing factors that give rise to these events in our region. Past studies of *Pseudo-nitzschia* blooms in California have typically used a definition of 10,000 cells/L of either size class as a bloom threshold for species in this genus (Fehling et al. 2006;

Howard et al. 2007; Jester et al. 2009; Lane et al. 2009). Based on this definition, *Pseudo-nitzschia* attained bloom abundances in 65 of the 178 total samples collected at Newport pier. *Pseudo-nitzschia* cells were observed in a total of 87 of the 178 samples, and therefore 75 % of the samples with *Pseudo-nitzschia* collected at Newport pier during the 3.5-year monitoring period would be considered bloom samples using the non-site specific bloom metric. This number was far greater than the number of samples exhibiting detectable pDA concentrations. Applying the same 10,000 cells/L bloom definition to *P. seriata* size class abundances only instead of the abundance of all *Pseudo-nitzschia* cells would still identify 38 of the 53 samples in which *P. seriata* was observed as bloom samples (72 %). Basing a bloom definition on a long-term mean of the abundances of the *P. seriata* size class of *Pseudo-nitzschia* reduced the number of samples that potentially contained detectable pDA concentrations, and thereby improved our ability to investigate relationships between emerging toxic events and environmental forcing factors.

Correlating bloom or toxic events to environmental forcing factors

The most significant relationship observed in the regression analysis of the datasets from either pier and pDA concentrations from identified bloom samples occurred at Newport pier with a 1-week time lag of chlorophyll, salinity, silicate to nitrate, and temperature (R^2 adj=0.537, p=0.004, Table 3). Chlorophyll, salinity and the silicate to nitrate ratio were positively correlated while temperature was negatively correlated with pDA concentrations. The positive correlation with chlorophyll concentrations observed in the preceding week may indicate that the detection of pDA during bloom events follows periods that are conducive to general microalgal growth. This result is consistent with experimental studies that have observed a link between DA production by *Pseudo-nitzschia* species and stationary growth (Bates 1998). A positive correlation with salinity and negative correlation with temperature suggests a correlation between pDA production and upwelling events as water brought to the surface in an upwelling event will be colder, slightly more saline and rich in nutrients. Recent research in the nearshore environment of southern California has shown that upwelling can stimulate *Pseudo-nitzschia* population growth specifically, eventually stimulating pDA production (Schnetzer et al. 2012). Silicate to nitrogen ratios for marine diatoms have been shown to be 1 Si:1 N but may fluctuate according to species due to differences in the silica content of their frustule. The positive correlation to high silicate to nitrate ratios the week prior to detectable pDA concentrations may signify an excess of silicate present for the stimulation of diatom growth, although *Pseudo-nitzschia* has been shown to be lightly silicified and capable of prospering in low silicate conditions (Fehling et al. 2006).

In the regression analysis with *P. seriata* abundances as the dependent variable there was a positive correlation between silicate concentrations 2 weeks prior to identified *P. seriata* blooms in conjunction with positive correlations to rainfall and temperature and a negative correlation to salinity at Newport pier (R^2 adj=0.653, p=0.001, Table 3). The switch between a negative correlation with temperature for pDA concentrations to a positive correlation with temperature for *P. seriata* abundances may demonstrate the difference in environmental parameters that stimulate *Pseudo-nitzschia* growth versus parameters that stimulate pDA production directly. Application of the *P. seriata* size class bloom identification metric to larger datasets collected in the region will allow researchers to separate events that have occurred in which *Pseudo-nitzschia* growth has been stimulated without inciting pDA production for comparison to events in which both growth and pDA production has been promoted. The information gleaned from the statistical relationships identified here can direct future laboratory experimentation and field sampling efforts as we try to further elucidate the conditions that stimulate pDA production in southern California.

Conclusion

The identification, and eventually prediction, of emerging toxic bloom events of *Pseudo-nitzschia* along the coast of southern California is imperative given the recent documentation of increases in frequency and severity of the occurrence of DA in these waters and poisoning events resulting from this neurotoxin (Lewitus et al. 2012). An empirically defined, site-specific bloom definition using *Pseudo-nitzschia* cell abundance information was investigated in this study to provide researchers with a relatively rapid and inexpensive tool for identifying the onset of potentially toxic events. This information constitutes an initial screening tool with which to guide more intensive or expensive observations and measurements in the future as well as be applied to larger datasets collected in the region allowing for deeper analysis into the causation of historical *Pseudo-nitzschia* events. The differences in results obtained in the regression analysis for Newport and Redondo Beach piers demonstrated that while bloom events could still be identified with the *P. seriata* abundance based metric, the specificity of statistical relationships identified were weak at the Redondo Beach location at which only basic microalgal community information was collected. We suggest for monitoring programs that include objectives for the understanding of *Pseudo-nitzschia* events nutrient and reliable salinity information should be collected.

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