Harmful Algae 67 (2017) 36–43

Contents lists available at ScienceDirect

Harmful Algae

journal homepage: www.elsevier.com/locate/hal

Understanding the blob bloom: Warming increases toxicity and abundance of the harmful bloom diatom *Pseudo-nitzschia* in California coastal waters

Zhi Zhu, Pingping Qu, Feixue Fu, Nancy Tennenbaum, Avery O. Tatters, David A. Hutchins*

Marine and Environmental Biology, University of Southern California, Los Angeles, CA 90089, USA

ARTICLE INFO

Article history:
Received 7 February 2017
Received in revised form 12 May 2017
Accepted 11 June 2017
Available online xxx

Keywords:
Domoic acid
Pseudo-nitzschia
Warming
Temperature
HAB
The blob

ABSTRACT

The toxic diatom genus *Pseudo-nitzschia* produces environmentally damaging harmful algal blooms (HABs) along the U.S. west coast and elsewhere, and a recent ocean warming event coincided with toxic blooms of record extent. This study examined the effects of temperature on growth, domoic acid toxin production, and competitive dominance of two *Pseudo-nitzschia* species from Southern California. Growth rates of cultured *P. australis* were maximal at 23 °C (~0.8 d^{-1}), similar to the maximum temperature recorded during the 2014–2015 warming anomaly, and decreased to ~0.1 d^{-1} by 30 °C. In contrast, cellular domoic acid concentrations only became detectable at 23 °C, and increased to maximum levels at 30 °C. In two incubation experiments using natural Southern California phytoplankton communities, warming also increased the relative abundance of another potentially toxic local species, *P. delicatissima*. These results suggest that both the toxicity and the competitive success of particular *Pseudo-nitzschia* spp. can be positively correlated with temperature, and therefore there is a need to determine whether harmful blooms of this diatom genus may be increasingly prevalent in a warmer future coastal ocean.

1. Introduction

With increasing anthropogenic CO_2 emissions, global temperatures are predicted to increase 2.6–4.8 °C by 2100 (IPCC, 2014). Increases in the frequency and severity of harmful algal blooms (HABs) may be among the many impacts of this warming (Hallegraeff, 2010; Fu et al., 2012). For instance, growth of the toxic dinoflagellates *Alexandrium* and *Dinophysis* appears to be benefiting from ongoing sea surface temperature increases in both the Atlantic and Pacific Oceans (Gobler et al., 2017). Another potentially toxic algal group that could be favored by current global warming trends is the diatom genus *Pseudo-nitzschia* spp.

Many *Pseudo-nitzschia* species produce the neurotoxin domoic acid (DA), the causal agent of amnesic shellfish poisoning. *Pseudo-nitzschia* blooms occur frequently around the world and have been reported in North America, South America, Australia, Asia, and Europe (Bates et al., 1998; Hasle, 2002; Hernández-Becerril et al., 2007; Takahashi et al., 2007; Trainer et al., 2012). In the United States, *Pseudo-nitzschia* blooms recur nearly every year along the California coast, where they cause mass mortalities of sea lions and seabirds (Trainer et al., 2012). DA is capable of dispersing widely in coastal food webs, where it has been detected in organisms ranging from mussels and sardines up to blue whales (Lefebvre et al., 2002; Schnetzer et al., 2013). The occurrence of toxic harmful algal blooms dominated by *Pseudo-nitzschia* spp. is often difficult to predict, because the same species can be toxic or non-toxic (Lundholm et al., 1994; Lelong et al., 2012). Various factors including temperature, irradiance, nutrient availability, and bacterial interactions can also affect DA production (Bates et al., 1998; Pan et al., 1998; Sun et al., 2011; Tatters et al., 2012; Lelong et al., 2012; Sison-Mangus et al., 2014).

In 2014–2015, an extraordinarily widespread and sustained harmful *Pseudo-nitzschia* bloom was observed along nearly the entire west coast of the United States, coinciding with a persistent warm-water anomaly that was nicknamed 'the Blob' (Bond et al., 2015). The toxic bloom formed during the course of this unprecedented regional warming event from early spring 2014 until the summer of 2015, a period during which sea surface temperatures were up to 4 °C above long-term averages from

* Corresponding author.
E-mail address: dahutch@usc.edu (D.A. Hutchins).

http://dx.doi.org/10.1016/j.hal.2017.06.004
1568-9883/

Alaska to Southern California (Bond et al., 2015; McCabe et al., 2016). The bloom was promoted by upwelled nutrients after the spring transition (McCabe et al., 2016). Historically high concentrations of domoic acid were present throughout the marine food web in this area, creating widespread mortality of marine mammals and birds (McCabe et al., 2016). Economic losses from this massive bloom due to closure of the Dungeness crab fishery were estimated at >$50 million (McCabe et al., 2016; Leising et al., 2015; Cavole et al., 2016).

Research on the combined effects of upwelling and short-term warming events on the occurrence of harmful algal bloom is still scarce. A recent study found correlations between historical outbreaks of elevated domoic acid levels, and warm water transitions due to regional Pacific Decadal Oscillation trends and El Niño occurrences (McKibben et al., 2017). The linkage of the most devastating harmful *Pseudo-nitzschia* bloom ever observed along the west coast to the 2014/2015 regional warming event underscores the need for research on the impacts of warming on DA production, and on the composition of natural phytoplankton communities containing *Pseudo-nitzschia* spp. A number of previous studies have examined the influence of changing temperature on various *Pseudo-nitzschia* species (Amato et al., 2010; Lewis et al., 1993; Lundholm et al., 1994; Santiago-Morales and García-Mendoza, 2011; Thorel et al., 2014). These thermal response studies, however, found contradictory results. Lundholm et al. (1994) and Amato et al. (2010) demonstrating that warming decreased cellular DA concentrations in *Pseudo-nitzschia* spp., while Lewis et al. (1993) and Thorel et al. (2010) observed increased DA at warmer temperatures.

The study presented here addresses this still unresolved issue by examining the effects of temperature on domoic acid production in a *Pseudo-nitzschia australis* strain isolated from the Southern California Bight near the beginning of the Blob event. Additional experiments tested the effects of temperature increases on the species composition of two *Pseudo-nitzschia delicatissima*-containing natural phytoplankton communities from this region. The objective of this study was to gain a better understanding of how anomalous warming events such as the Blob, or future sustained climate warming, may affect the occurrence and toxicity of harmful *Pseudo-nitzschia* blooms in the California Current region.

2. Materials and methods

2.1. Culture thermal response curves

P. australis S7 was micropipette isolated from public nearshore water collected at 33.73 N, 118.35 W in Los Angeles County, California in March, 2014. This strain was maintained at 16 °C on a 12 h light: 12 h dark cycle in Aquil* media (Sunda et al., 2005) under 150 μmol photons $m^{-2} s^{-1}$ of cool white fluorescent light.

For thermal response curve experiments, the *P. australis* S7 culture was transferred to temperatures of 12 °C, 15 °C, 18 °C, 20 °C, 23 °C, 26 °C, 28 °C, and 30 °C step by step in triplicate 500 ml acid washed polycarbonate bottles, under the same light conditions as stock cultures. Cultures were diluted every 2 days to keep the culture at exponential growth stage with Aquil* medium pre-acclimated to the appropriate temperature. After steady-state growth was attained, as indicated by stable growth rates for 3–5 consecutive transfers, the cultures were sampled 48 h after dilution.

2.2. Natural community temperature manipulations

Two experiments were conducted with *Pseudo-nitzschia*-containing assemblages to examined change in community structure over a range of temperatures after adding nutrients to simulate an upwelling event. Whole seawater was collected at Stearns Wharf (34.41N, 119.69W), Santa Barbara, California in February and March 2016 at ambient temperatures of 13 °C and 15 °C, respectively. The seawater was filtered with 80 μm mesh to remove large zooplankton. The February experiment (SB1) used 800 ml of seawater in triplicate 1L polycarbonate bottles with additions of 20 μM nitrate ($NaNO_3$), 20 μM silicate (Na_2SiO_3) and 2 μM phosphate (NaH_2PO_4) and vitamin and trace metal additions following the Aquil* recipe to simulate nutrient supplementation from upwelling (Sunda et al., 2005). Bottles were incubated at 13 °C, 18 °C, and 23 °C. For seawater collected in March (SB2), 800 ml aliquots of seawater were added to triplicate 1L polycarbonate bottles with 100 μM nitrate ($NaNO_3$), 100 μM silicate (Na_2SiO_3) and 10 μM phosphate (NaH_2PO_4) and vitamins and trace metals following the Aquil* recipe. In SB2, the bottles were incubated at 15 °C, 20 °C, 25 °C, and 28 °C. Both incubation experiments were incubated on a 12 h light: 12 h dark cycle under 150 photons $m^{-2} s^{-1}$ of cool white fluorescent light in laboratory incubators (Percival, IA), and all the bottles were gently mixed manually at least once per day. Samples were collected at the beginning of the experiments (T0) and at the final timepoints for both SB1 and SB2.

2.3. Growth rates

In vivo fluorescence of each culture was measured using a 10-AU™ fluorometer (Turner Designs, CA) to calculate growth rates. Specific growth rates, expressed as d^{-1}, were calculated as: $\mu = (\ln N_1 - \ln N_0)/t$, where N_0 and N_1 are *in vivo* fluorescence at the beginning and end of a dilution period, respectively, and t is the duration of the dilution period. Q_{10} of growth rates of *P. australis* was calculated as: $Q_{10} = (\mu_2/\mu_1)^{10/(T2-T1)}$ (Chaui-Berlinck et al., 2002), where μ_1 and μ_2 are the specific growth rates of the phytoplankton at temperature T_1 (Celsius) and T_2, respectively.

2.4. Elemental analysis

In addition to potential temperature effects, nutrient limitation has also been shown to affect the toxicity of *Pseudo-nitzschia* spp. (Bates et al., 1998; Sun et al., 2011; Tatters et al., 2012; Lelong et al., 2012). As an indicator of potential thermal effects on nutrient stress, C: N: P ratios were examined in the *P. australis* culture experiment and the natural community experiment SB1 only, due to logistical constraints during SB2. 20–100 ml culture samples of each treatment were filtered onto pre-combusted GF/F filters (500 °C for 2 h) and dried in a 60 °C oven overnight for particulate organic carbon/nitrogen (POC/PON) and particulate organic phosphorus (POP) analyses, respectively. POC/PON samples were analyzed using a 440 Elemental Analyzer (Costech Inc, CA) following Fu et al. (2007). POP was analyzed using a molybdate colorimetric method according to Fu et al. (2007). 20 to 100 ml of culture samples of each treatment was filtered onto 2 μm polycarbonate filters (GE Healthcare, CA) and dried in a 60 °C oven overnight for biogenic silica (BSi) analysis following Paasche (1980).

2.5. Chlorophyll an analysis

20 to 100 ml culture samples were filtered onto GF/F filters and 10 μm polycarbonate filters (Whatman) at low vacuum, frozen overnight, and extracted with 90% aqueous acetone for 24 h at −20 °C, followed by measurements using the non-acidification method on a 10-AU™ fluorometer (Turner Designs, CA), as in Fu et al. (2007).