Positive feedbacks between bottom-up and top-down controls promote the formation and toxicity of ecosystem disruptive algal blooms: A modeling study

William G. Sunda [a],[*], Kyle W. Shertzer [b]

[a] Beaufort Laboratory, National Centers for Coastal Ocean Science, NOS, NOAA, 101 Pivers Island Road, Beaufort, NC 28516, USA
[b] Beaufort Laboratory, Southeast Fisheries Science Center, NMFS, 101 Pivers Island Road, Beaufort, NC 28516, USA

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ABSTRACT

Harmful algal blooms that disrupt and degrade ecosystems (ecosystem disruptive algal blooms, EDABs) are occurring with greater frequency and severity with eutrophication and other adverse anthropogenic alterations of coastal systems. EDAB events have been hypothesized to be caused by positive feedback interactions involving differential growth of competing algal species, low grazing mortality rates on EDAB species, and resulting decreases in nutrient inputs from grazer-mediated nutrient cycling as the EDAB event progresses. Here we develop a stoichiometric nutrient–phytoplankton–zooplankton (NPZ) model to test a conceptual positive feedback mechanism linked to increased cell toxicity and resultant decreases in grazing mortality rates in EDAB species under nutrient limitation of growth rate. As our model EDAB alga, we chose the slow-growing, toxic dinoflagellate *Karenia brevis*, whose toxin levels have been shown to increase with nutrient (nitrogen) limitation of specific growth rate. This species was competed with two high-nutrient adapted, faster-growing diatoms (*Thalassiosira pseudonana* and *Thalassiosira weissflogii*) using recently published data for relationships among nutrient (ammonium) concentration, carbon normalized ammonium uptake rates, cellular nitrogen:carbon (N:C) ratios, and specific growth rate. The model results support the proposed positive feedback mechanism for EDAB formation and toxicity. In all cases the toxic bloom was preceded by one or more pre-blooms of fast-growing diatoms, which drew dissolved nutrients to low growth rate-limiting levels, and stimulated the population growth of zooplankton grazers. Low specific grazing rates on the toxic, nutrient-limited EDAB species then promoted the population growth of this species, which further decreased grazing rates, grazing-linked nutrient recycling, nutrient concentrations, and algal specific growth rates. The nutrient limitation of growth rate further increased toxin concentrations in the EDAB algae, which further decreased grazing-linked nutrient recycling rates and nutrient concentrations, and caused an even greater nutrient limitation of growth rate and even higher toxin levels in the EDAB algae. This chain of interactions represented a positive feedback that resulted in the formation of a high-biomass toxic bloom, with low, nutrient-limited specific growth rates and associated high cellular C:N and toxin:C ratios. Together the elevated C:N and toxin:C ratios in the EDAB algae resulted in very high bloom toxicity. The positive feedbacks and resulting bloom formation and toxicity were increased by long water residence times, which increased the relative importance of grazing-linked nutrient recycling to the overall supply of limiting nutrient (N).

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1. Introduction

Harmful algal blooms (HABs) have occurred with increasing frequency in recent years with eutrophication and other anthropogenic alterations of coastal ecosystems (Glibert et al., 2005; Heisler et al., 2008). Many of these harmful blooms severely alter or degrade ecosystem function, and have been referred to as ecosystem disruptive algal blooms (EDABs) (Sunda et al., 2006). These blooms are often caused by toxic or unpalatable species that decrease grazing rates by herbivores, and thereby disrupt the transfer of nutrients and energy to higher trophic levels and decrease nutrient cycling. EDAB species include brown tide pelagophytes, and toxic haptophytes, cyanobacteria, and dinoflagellates among others (Sunda et al., 2006).

* Corresponding author. Tel.: +1 252 728 8754.
E-mail address: bill.sunda@noaa.gov (W.G. Sunda).

Both algal nutrient availability and herbivore grazing are known to influence the dynamics of EDAB events and algal blooms in general (Calbet et al., 2003; Irigoien et al., 2005; Buskey, 2008; Strom et al., 2013). Potential interactions between these two factors have also been noted because nutrient limitation of algal prey can increase the grazing defenses in phytoplankton (Graneli and Flynn, 2006; Sunda et al., 2006) and thereby decrease the population growth and nutrient recycling rates of zooplankton which are linked to grazing rates (Sterner and Elser, 2002; Mitra and Flynn, 2005).

Sunda et al. (2006) presented a conceptual model for the development of EDABs based on positive feedback interactions among algal growth competition for limiting nutrients, low herbivore grazing rates on EDAB species, and grazing-linked nutrient regeneration. This model was based on the hypothesis of fundamental tradeoffs between functional traits that promote high rates of nutrient acquisition, growth and reproduction in an algal species and traits that promote low grazing mortality rates (large cell size, toxicity, and unpalitability) (Yoshida et al., 2003; Litchman et al., 2007; Sunda and Hardison, 2010). Due to these tradeoffs species with high maximum specific growth rates (high-nutrient adapted, r-selected species) were also susceptible to high rates of grazing mortality, while low-nutrient-adapted, *K*-selected species were well defended against grazing, but had much lower maximum growth rates. Thus, each set of species is adapted for optimal net population growth under different sets of nutrient and grazing conditions.

In their conceptual model Sunda et al. (2006) proposed that nutrient inputs first stimulated blooms of fast-growing, readily-grazed, r-selected species (e.g., diatoms) which drove dissolved nutrients to low concentrations and promoted the growth of zooplankton. This set the stage for population growth of *K*-selected EDAB species, which were better adapted for growth at low nutrient levels and were well defended from zooplankton grazing due to their toxicity or low palatability. This combination of traits allowed the EDAB species to proliferate at the expense of the r-selected species, whose growth rate was greatly reduced at low nutrient concentrations and were less well-defended against grazing. Sunda et al. (2006) noted that the increased biomass of the poorly-grazed EDAB species would cause zooplankton grazing rates to decline, which would decrease grazing linked nutrient recycling and further decrease limiting nutrient concentrations, and thereby further promote population growth of the EDAB species at the expense of competing algae. The authors proposed that these effects constituted an inherent positive feedback that should promote the development of harmful blooms (Sunda et al., 2006). They also noted that there would be an additional EDAB-promoting positive feedback in cases where the toxicity and related grazing defenses of the EDAB species increased with nutrient limitation of growth rate, which commonly occurs in toxic algae (Sunda et al., 2006; Graneli and Flynn, 2006).

Previously, Sunda and Shertzer (2012) developed a stoichiometric nutrient–phytoplankton–zooplankton (NPZ) model to test the positive feedback hypothesis of Sunda et al. (2006) for the development of EDAB events where the EDAB species had high levels of the grazing defenses, which did not vary with nutrient limitation of growth rate. In the present model we examined the effect of an additional positive feedback associated with increases cellular toxins with nutrient limitation of growth rate of the EDAB species (Sunda et al., 2006). Such increases in cellular toxicity with nutrient limitation were proposed to substantially promote both the occurrence and toxicity of EDAB events (Mitra and Flynn, 2006; Sunda et al., 2006).

In the present model the slow growing, toxic EDAB species *Karenia brevis* (K.b.) was competed with two faster growing, less well defended diatom species (*Thalassiosira pseudonana* [T.p.] and

T. weissflogii [T.w.]). The selection of K.b. as our model EDAB species is supported by its nearly annual formation of toxic red tide blooms in the Gulf of Mexico, which adversely affect human health, marine ecosystems, and coastal economies (Flewelling et al., 2005; Kirkpatrick et al., 2006; Watkins et al., 2008). K.b. was also chosen because of the recent publication of quantitative relationships between nutrient (N and phosphorus [P]) limitation of specific growth rate and increased ratios of cellular toxins (brevetoxins) to cell carbon (C) in this species (Hardison et al., 2012, 2013). These relationships provided the data needed to construct an NPZ model that directly examines the effect of increased cellular toxins under N-limitation of growth rate on the occurrence and toxicity of EDAB events.

Another major reason for the choice of the above three algal species in our stoichiometric NPZ model was the recent availability of data describing relationships among nutrient (NH_4^+) concentration, NH_4^+ uptake rates, cellular N:C ratios, and specific growth rate in these and other algal species (Sunda and Hardison, 2007, 2010; Sunda et al., 2009; Hardison et al., 2012). These data were used to compute growth competition for NH_4^+ among the three species. Due to analytical difficulties, no equivalent growth versus nutrient data are available for any other N-substrates or for phosphate, which precluded the inclusion of other N-substrates such as nitrate or dissolved organic N (DON) in our model.

In addition, NH_4^+ was chosen as the limiting nutrient for several other reasons. Nitrogen is the primary limiting nutrient in coastal waters and stratified ocean waters (Ryther and Dunstan, 1971; Sanders et al., 1987; Litchman et al., 2007; Moore et al., 2013) and NH_4^+ is frequently the primary limiting inorganic N species in N-limited marine waters (Harrison et al., 1996). Also NH_4^+ is a major form of regenerated N (Dugdale and Goering, 1967; Verity, 1985), and thus, is an essential component of NPZ models (like this one) that include grazer-mediated N-regeneration.

A central process in our NPZ model is differential grazing by zooplankton, which removes individual algal species at varying rates, and thus, along with differing growth rates, controls the abundance and species composition of the algal community. Algal grazing supports the growth and reproduction rate of the grazing predator and has another critical function: it facilitates the recycling of nutrients through excretion. Therefore, it is essential for resupplying the nutrient (NH_4^+) pool, especially during periods of low external nutrient supply.

In the present model, the toxin content and associated grazing defenses in our model EDAB alga (K.b.) increased with N-limitation of specific growth rate. K.b. produces a suite of structurally related neurotoxins (brevetoxins), which activate voltage-gated sodium channels in cell membranes and thereby disrupt normal activity of excitable membranes needed, for example, for nerve transmission and muscle activity. These channels are widely distributed in biological systems, including unicellular protozoan grazers (Liebeskind et al., 2011). By interfering with these gated sodium channels in protozoans, these toxins may disrupt critical cellular functions such as cell signaling, flagellar motility, prey ingestion, or the coordinated beating of cilia used in motility and feeding. Recently, cellular brevetoxins were shown to deter grazing by zooplankton, specifically copepods (Cohen et al., 2007; Hong et al., 2012; Waggett et al., 2012), suggesting that toxic blooms of K.b. could be promoted by low grazing rates directly linked to cellular brevetoxin concentrations.

The predator in the NPZ model was a generic protozoan grazer, whose grazing rates and metabolic growth efficiency were designed to mimic those of a large ciliate (Hansen et al., 1997). Ciliates are a dominant group of microzooplankton (Pierce and Turner, 1992), and microzooplankton are responsible for most algal grazing in coastal waters (Calbet and Landry, 2004). In addition, ciliates have been identified as dominant grazers during a

toxic dinoflagellate bloom (Calbet et al., 2003). Protozoan grazers reproduce through binary fission, which greatly simplifies the linkage between grazing rates and zooplankton growth and reproduction.

2. Methods

2.1. Model structure

Our ecosystem model (Fig. 1) assumed a single well-mixed system in which water entered and exited at the same rate, such as in a chemostat or an idealized coastal system. For simplicity, the model intentionally ignored hydrodynamic factors such as advection (Stumpf et al., 2008) and vertical migration of K.b. (Kamykowski and Yamazaki, 1997), which may influence bloom formation. This allowed us to focus strictly on the influence of several core ecosystem processes (algal growth competition for nutrients, differential grazing linked to cell size and toxicity, and recycling of nutrients linked to grazing) on bloom development and toxicity (Sunda et al., 2006). The inflowing water in the system contained nutrients but no algae or grazers. Nitrogen in the form of ammonium (NH_4^+) was assumed to be the sole limiting nutrient. Its concentration (μmol N l^{-1}) was denoted by the symbol N. A simple algal community contained three competing species: a small fast-growing diatom *Thalassiosira pseudonana* (T.p., ~4.5 μm diameter; maximum specific growth rate (μ_{max}) = 1.45 d^{-1} at 20 °C) a larger slower-growing diatom *T. weissflogii* (T.w., ~10–11 μm diameter; μ_{max} = 0.89 d^{-1} at 20 °C), and an even slower growing toxic EDAB species *Karenia brevis* (K.b., 9–10 μm diameter; μ_{max} = 0.40 d^{-1} at 20 °C) (Sunda and Hardison, 2010; Hardison et al., 2012). Thus, K.b. should be able to out-compete the diatoms only if its grazing mortality rate is much lower than those of the diatoms.

The carbon biomass concentration of each algal species (μmol C l^{-1}) was denoted by P_i, where i = 1, 2 and 3 represents
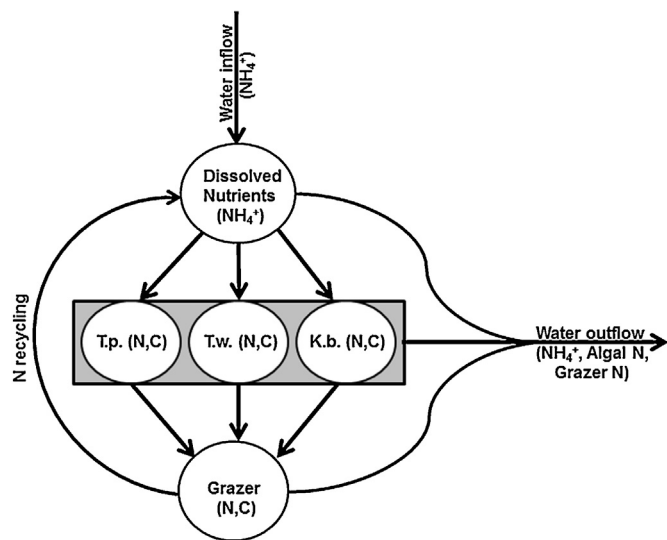

Fig. 1. Diagamatic model of the nutrient–phytoplankton–zooplankton system. Arrows indicate nitrogen flux into, within, and out of the system. Inflowing water into the system contains a set concentration of N in the form of NH_4^+. The NH_4^+ is taken up by the three competing algal species (T.p, T.w., and K.b.) and used to synthesize algal N and C biomass. The algae are consumed by the grazer at differing rates and their N and C content are used to fuel grazer growth and reproduction. The algal N not assimilated into grazer biomass (N, C) is recycled back into the NH_4^+ pool. N is lost from the system from water outflow in the form of dissolved NH_4^+, algal N, and grazer N and this N loss rate equals the rate of NH_4^+ inflow. The total N concentration in the system (that present as dissolved NH_4^+, algal N, and grazer N) equals the NH_4^+ concentration in the inflowing water.

K.b., T.p., and T.w., respectively. The system contained a zooplankton grazer present at a C-biomass concentration Z (μmol C l^{-1}) that consumed each algal species at different C-specific rates. We quantified this conceptual system using a stoichiometric NPZ model, similar to that described by Sunda and Shertzer (2012). Here we highlight several of the model's key features and assumptions. A full model description and parameter units and values are given in the Supplementary Material (S.M.), Appendix 1.

The NPZ model was governed by a system of differential equations:

$$\frac{dN}{dt} = \delta(N_{in} - N) - \sum_i V_i P_i + (R_D + R_E + R_m)Z \tag{1}$$

$$\frac{dQ_i}{dt} = V_i - \mu_i Q_i \tag{2}$$

$$\frac{dP_i}{dt} = (\mu_i - \delta)P_i - I_i Z \tag{3}$$

$$\frac{dZ}{dt} = (\mu_z - m - \delta)Z \tag{4}$$

where Q_i is the N:C ratio of the algal species i; V_i is the NH_4^+ uptake rate of algal species i normalized to cell C; R_D, R_E, and R_m are the N-recycling rates associated with the grazer's fecal defecation, excretion of excess assimilated N, and mortality, respectively; μ_i is the specific growth rate of algal species i; μ_z is the specific growth rate of the grazer; m is the specific mortality rate of the grazer; and I_i is the C-specific ingestion rate of algal species i by the grazer. The NH_4^+ input rate equals the concentration in the inflowing water (N_{in}) times the dilution rate (δ) of the system (equals 1/residence time), and the NH_4^+ washout rate equals the NH_4^+ concentration in the system (N) times the dilution rate. The total NH_4^+ uptake rate by the algae equals the sum of the uptake rates of the three algal species, while the net growth rate of each species (dP_i/dt; mol algal C liter^{-1} d^{-1}) equals the specific growth rate (μ_i, d^{-1}) times the C-biomass (mol C l^{-1}), minus the rates of loss from washout and grazing. The remainder of the *Methods* section provides additional detail on terms in the NPZ model (Eqs. (1)–(4)).

2.2. Nutrient uptake and growth of phytoplankton

Ammonium uptake rates of T.w. and T.p. (V_i, Eq. (2)) were modeled using a modified Michaelis–Menten equation (S.M., Appendix 1; Sunda et al., 2009). Uptake rates of both species increased approximately linearly with the NH_4^+ concentration until saturating at a maximum rate. For that reason and because of data limitations, we assumed a piecewise linear model for the NH_4^+ uptake rate of K.b. (S.M., Appendix 2). For all three algal species, specific growth rates (μ_i) were modeled using a modified Droop equation (S.M., Appendix 1; Sunda et al., 2009), in which the growth rate is a function of the cellular N:C ratio. Parameter values for T.w. and T.p. were determined previously (Sunda et al., 2009), and those for K.b. were estimated for this analysis using similar procedures (S.M., Appendix 2).

2.3. Grazing and growth of zooplankton

The grazer was modeled after a generic ciliate whose maximum grazing rate (3.6 d^{-1}), half saturation grazing value (12 μmol C l^{-1}), and gross growth efficiency (0.39) were based on average data for large ciliates (n = 5; cell volume 6.9–21 × 10^4 μm^3) (Hansen et al., 1997). Grazing rates were assumed to follow a multispecies type-II functional response similar to that

of Fussmann and Heber (2002), but modified to allow for a threshold below which grazing ceases (Gentleman et al., 2003; Tian, 2006). Carbon-specific ingestion rates (I_i) of algal species i were modeled as:

$$I_i = \begin{cases} g_{\max,i}(P_T - P_\tau)(P_i/P_T)/(K_z + P_T - P_\tau) & \text{if } P_T > P_\tau \\ 0 & \text{otherwise} \end{cases} \quad (5)$$

where $g_{\max,i}$ is the maximum grazing rate on algal species i, P_T is the total algal C-biomass (mol C l^{-1}; $P_T = \sum_j P_j$), P_τ is the grazing threshold (detailed below), and K_z is the half-saturation constant for grazing, which for simplicity, is assumed to be the same for all algal species (Sunda and Shertzer, 2012).

The $g_{\max,i}$ value for T.p., the fastest growing and most readily grazed species, was set at 3.6 d^{-1}, based on published data (Hansen et al., 1997). The $g_{\max,i}$ value for the large diatom T.w. was set at 40% of that for T.p. (1.44 d^{-1}) to reflect the observed lower grazing rates on larger algal species (Thingstad and Sakshaug, 1990; Kiørboe, 1993). The $g_{\max,i}$ value for K.b. was varied in our model, and was scaled downward from a maximum value ($g'_{\max,1} = 1.08$ d^{-1}; 30% of that of T.p.) by a negatively sloped linear function,

$$g_{\max,1} = g'_{\max,1}(\max(0.0, 1.0 - \beta_3 B)) \quad (6)$$

where B is the cellular brevetoxin:C ratio (g brevetoxin [mol C]$^{-1}$), and β_3 (mol C g^{-1}) is the slope parameter that scales the relationship between grazing deterrence in K.b. and the cell brevetoxin:C ratio. Henceforth β_3 is referred to as the brevetoxin grazing deterrence factor.

Based on Eq. (6), the maximum grazing rates varied from 1.08 d^{-1} to zero, depending on the value of β_3 and the cell brevetoxin:C ratio (B). The parameter β_3 was assigned a standard value of 1.5 mol C (g brevetoxin)$^{-1}$, but this value was also modified to simulate various levels of grazing deterrence for K.b., as described below in the *Model Scenarios* section. The model implicitly assumed that deterrence of grazing on K.b. was based on the zooplankton's ability to discriminate against the toxic cells, rather than to an inhibition of metabolic processes in the grazer, as has been shown previously for zooplankton (copepod) grazing on mixed populations of the toxic dinoflagellate *Alexandrium* sp. and non-toxic dinoflagellates (Teegarden, 1999). In those experiments, low levels of ingestion of the toxic dinoflagellate did not inhibit grazing rates on co-occurring non-toxic species, which we also implicitly assume in our model.

In our model, the cellular brevetoxin:C ratio (B) was inversely related to the specific growth rate (μ_1) based on data for the behavior of K.b. (strain CCMP 2228) with decreasing specific growth rate caused by N- or P-limitation (Hardison et al., 2012, 2013). This relationship was assumed to be linear:

$$B = \beta_1 - \beta_2 \mu_1 \quad (7)$$

where β_1 is the cellular brevetoxin at zero growth rate (1.17 g [mol C]$^{-1}$) and the slope parameter β_2 has a value of 2.15 g [mol C]$^{-1}$ d.

We utilized a grazing threshold in our grazing model (Eq. (5); $P_\tau = 2.0$ μmol C l^{-1}). We note that algal grazing data are often modeled as a saturation function of algal concentration with the curve passing through the origin; i.e., zero grazing rate at an algal concentration of zero (Hansen et al., 1997; Tian, 2006). However, grazing data are often noisy with few or no observations at algal concentrations near zero, so whether the curve passes through the origin cannot usually be determined. Grazing curves derived from less noisy data at very low algal C biomass often have a positive x-intercept, with no measurable grazing until some critical algal threshold is reached (Frost, 1975; Strom, 1991; Gismervik, 2005; Chen et al., 2010). Nutrient–phytoplankton–zooplankton (NPZ) models often require the inclusion of grazing thresholds to prevent large oscillations in phytoplankton populations, which do not

conform to field data for natural plankton (Frost, 1993; Strom et al., 2000; Leising et al., 2003).

The model tracked both C- and N-limited growth rates of the grazer (μ_C and μ_N) to determine which nutrient element would be most limiting. C-limited growth was modeled as,

$$\mu_C = \epsilon_g \left(\epsilon_a \sum_i I_i - r \right) \quad (8)$$

where I_i is the C ingestion rate of species i (Eq. (5)), r is specific basal respiration rate (0.1 d^{-1}), ε_a is the gut assimilation efficiency of grazing (0.8), and ε_g is the growth efficiency (0.5), equal to the increase in grazer C biomass divided by the net assimilated C available for growth (S.M, Appendix 1). Similarly, N-limited growth was modeled as,

$$\mu_N = \epsilon_a \sum_i \frac{I_i Q_i}{Q_z} \quad (9)$$

assuming a fixed N:C ratio in the zooplankton (Q_z) of 1:7 (Redfield et al., 1963; Sterner and Elser, 2002). The specific growth rate of the grazer (μ_z) was taken to be whichever was most limiting, $\mu_z = \min(\mu_C, \mu_N)$. In the present model, the zooplankton growth rate was always limited by C, and the excess assimilated N was excreted.

2.4. Nutrient recycling

Grazer-mediated recycling of N (per mol grazer C) stemmed from three sources: defecation of feces (R_D), excretion of excess assimilated N (R_E), and grazer mortality (R_m). Defecated N equaled the N ingested but not assimilated, defined by the complement of grazing assimilation efficiency (i.e., $1 - \varepsilon_a$),

$$R_D = (1 - \epsilon_a) \sum_i I_i Q_i \quad (10)$$

Nitrogen excretion equaled the difference between the N assimilation (A_N) and the assimilated N utilized for growth (G_N), $R_E = A_N - G_N$. In addition, the N content of dead zooplankton was recycled, $R_m = mQ_z$, where m is the specific mortality rate. For simplicity, we assumed that the N content of feces and dead zooplankton was rapidly converted to ammonium. We also assumed all excreted N was in the form of NH$_4^+$, quantitatively the most important N excretion product in microzooplankton (Verity, 1985; Caron and Goldman, 1990).

2.5. Model scenarios

We used the stoichiometric NPZ model to examine top-down and bottom-up effects in the system. For top-down effects, we first considered time-course simulations (0–200 or 0–500 d) with or without a grazer in the system. When present, the grazer preyed on all algal species according to differences in their maximum grazing rates, which for K.b. varied with the cell brevetoxin:C ratio (Fig. 2A, Eq. (7)) and the grazing deterrent effect of the brevetoxins (Fig. 2B, Eq. (6)). We further considered asymptotic dynamics of the system across a range of brevetoxin grazing deterrence factors (β_3 in Eq. (6)).

In the model, bottom-up effects were driven by the dilution rate (δ) and the NH$_4^+$ concentration in the incoming water (N_{in}). In addition to our standard dilution rate (0.1 d^{-1}), we examined time-course simulations for lower and higher dilution rates (0.05 and 0.3 d^{-1}). We also examined asymptotic dynamics across a range of rates ($\delta \in [0.01, 0.6]$ d^{-1}). And in addition to our standard NH$_4^+$ concentration in the inflowing water (12 μM) we conducted time-course simulations with lower and higher N_{in} values (4 and 24 μM). In all cases, parameters not used to define a particular scenario were fixed at their base values (S.M., Appendix 1).

3. Results

3.1. Algal growth models

Modeled relationships among specific growth rates of the three algal species and NH_4^+ concentration are shown in Fig. 3A. In these modeled curves, the small diatom *Thalassiosira pseudonana* (T.p.) outgrows the other two species at all ammonium (NH_4^+) levels except at very low concentrations (<0.008 μM), where it grows at rates similar to those for *Karenia brevis* (K.b.). Likewise, *T. weissflogii* (T.w.) has growth rates intermediate to T.p. and K.b. except at very low NH_4^+ (<0.01 μM) where rates for all species converge. Thus, NH_4^+ concentration is an important factor regulating the relative population growth of the three algal species. In the presence of the grazer, the high cellular brevetoxin:C ratios and associated low grazing rates under N-limitation of growth rate should help K.b. to compete with the two diatoms at low NH_4^+ levels where the growth rates of the three species converge.

The algal specific growth rate is ultimately determined by the cellular concentration of the limiting nutrient N (Droop, 1968; Sterner and Elser, 2002). Modeled Droop relationships between specific growth rate and cellular N:C ratio show only an ~2-fold range in this ratio with changes in growth rate in K.b. and T.w. and a slightly narrower range for T.p. (Fig. 3B).

3.2. Competition models

3.2.1. Top-down effects

Model runs were conducted to examine the effect of a grazer and of varying toxin-linked grazing rates on K.b. on the population dynamics of the three algal species. In the absence of the grazer, the fast growing diatom T.p. rapidly outgrew the other two species and
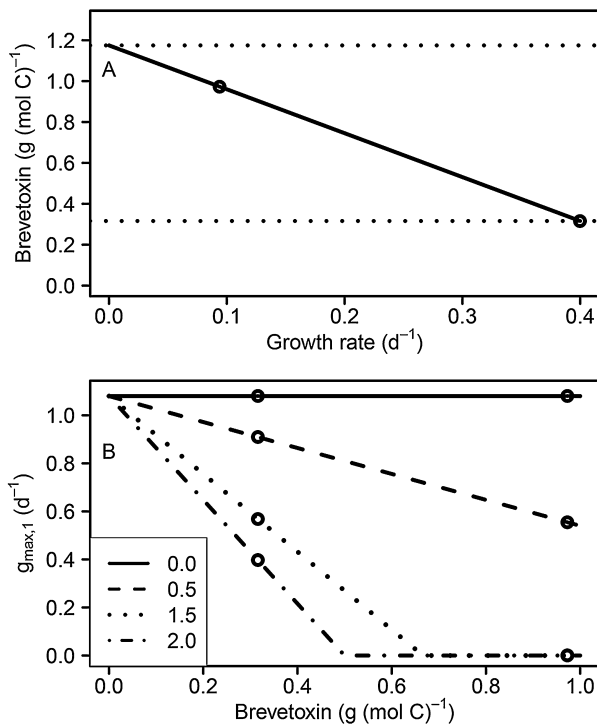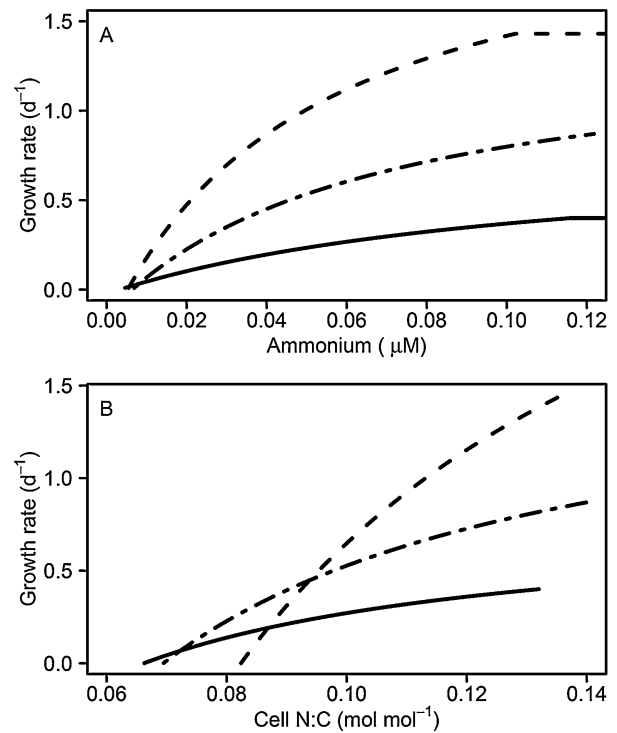
Fig. 3. Steady-state growth rates of *K. brevis* (solid line), *T. pseudonana* (dashed line), and *T. weissflogii* (short-long dashed line) as a function of NH_4^+ concentration (A), and cellular N:C molar ratio (B). Lines shown are modeled relationships based on laboratory culture data.

Fig. 2. A. Relationship between the cellular brevetoxin:C ratio and N-limited specific growth rate of *K. brevis* (K.b.). Dotted lines represent the minimum and maximum brevetoxin concentrations, and open circles represent measured values for K.b. strain CCMP 2228 (Hardison et al., 2012). B. Relationship between $g_{max,1}$ (the maximum grazing rate on K.b.) and the cellular brevetoxin:C ratio. The four lines are for different toxin grazing deterrence factors (β_3, lower left insert), which control how strongly brevetoxins decrease grazing rates (Eq. (6)).

became the sole dominant species after several days (Fig. 4C). By contrast in the presence of the grazer, the system showed quite complex behavior that was dependent on the toxin-linked maximum grazing rate on K.b. relative to the maximum grazing rates on the two diatoms (3.6 d^{-1} for T.p. and 1.44 d^{-1} for T.w.). When the brevetoxin grazing deterrence factor (β_3; Eq. (6), Fig. 2B) was zero (i.e., there was no toxin effect on grazing), no K.b. bloom occurred. Instead, there was an initial bloom of T.p. followed by stable oscillations among concentrations of NH_4^+, grazer, and T.w. (Fig. 4E–G). The NH_4^+ concentrations oscillated between growth rate limiting levels (0.03 μM) at high plankton biomass and high concentrations (~7 μM) at low biomass that supported maximum growth rates of all algal species (Fig. 4E). However, when β_3 was 1.5 (high grazing deterrence by brevetoxins), an entirely different dynamic occurred. Here there was again an initial bloom of T.p. which drew NH_4^+ concentrations to growth rate-limiting levels, followed by one or more successive blooms of T.w. and offset oscillating concentrations of NH_4^+ and grazer biomass (Fig. 4I–K). However, these dynamics were then followed by a sharp rise in K.b. biomass and brevetoxins which then persisted (Fig. 4K and L). During this time, there were initially offset blooms of K.b. and T.w. that underwent damped oscillations (Fig. 4K). This was followed by a replacement of T.w. by a lower steady state carbon biomass of the faster-growing T.p., high steady state K.b. biomass and brevetoxin concentration, and a constant but moderate grazer biomass (Fig. 4J–L). During this period the NH_4^+ was maintained at a constant, low concentration (~0.02 μM) due to its rapid uptake by the bloom algae (Fig. 4I), which led to low, N-limited specific growth rates of K.b. (~0.1 d^{-1}) and associated high brevetoxin:C ratios (see Fig. 2A). The high toxin:C ratio, in combination with the high K.b. carbon biomass (Fig. 4K), led to high overall brevetoxin concentrations (Fig. 4L). The exact time-dependent patterns for dissolved NH_4^+, grazer biomass, and biomass of the three algal species was quite complex and varied with the value of β_3.
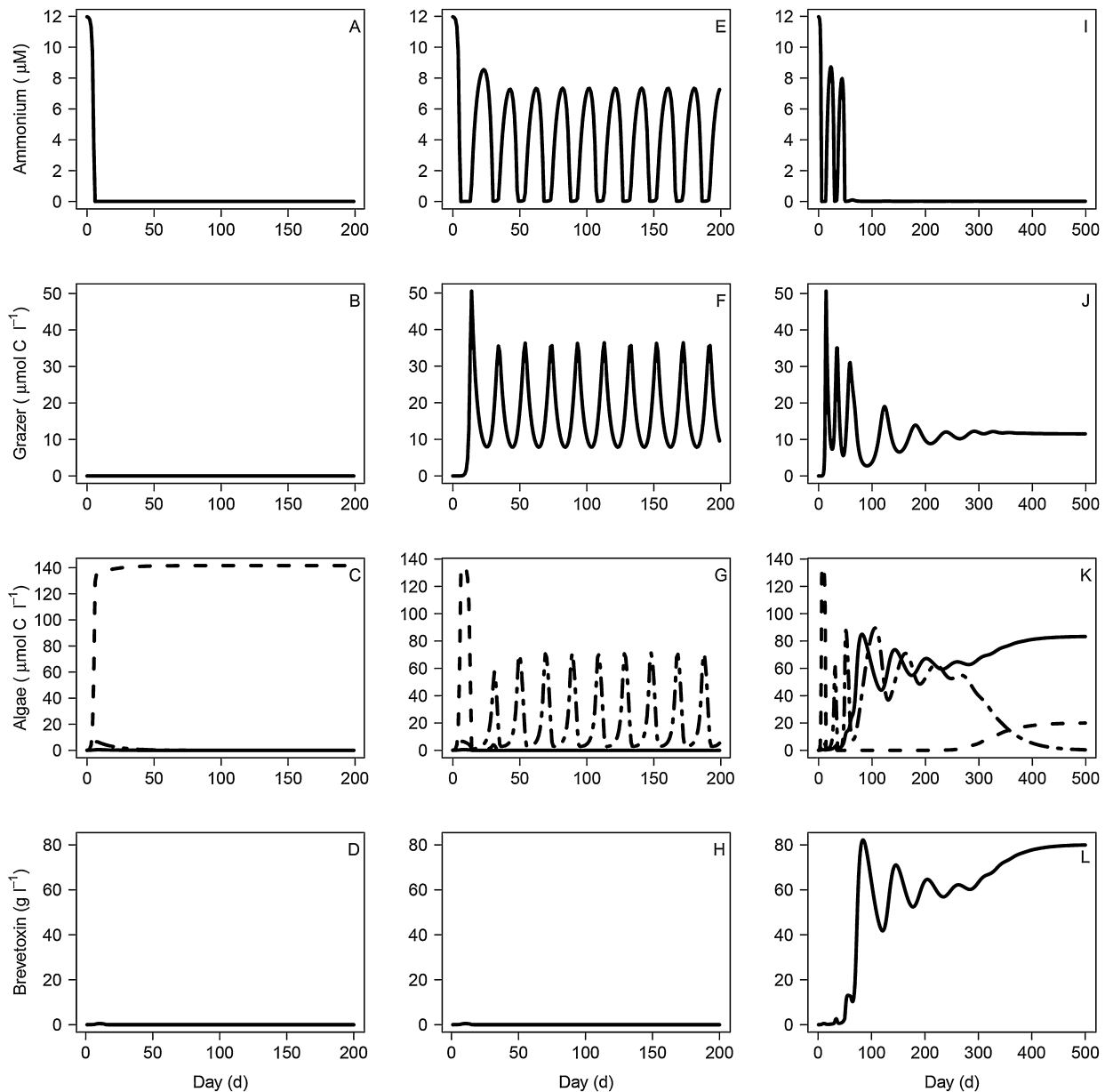
Fig. 4. Three time-course simulations: (A–D) without a grazer; (E–H) with a grazer but no deterrent effect of brevetoxins on grazing ($\beta_3 = 0$; Eq. (6)), and (I–L) with grazing deterred by brevetoxins ($\beta_3 = 1.5$, Eq. (6)). All other model parameters were set at their base values (S.M., Appendix 1), including the dilution rate ($\delta = 0.1$ d^{-1}) and the inflowing NH$_4^+$ ($N_{in} = 12$ μM). In panels C, G, and K, the algal species are *K. brevis* (solid line), *T. pseudonana* (dashed), and *T. weissflogii* (short-long dashed). Note that the time scale is shorter in the first two columns than in the third.

The transition between stable diatom oscillations and the onset of persistent, highly toxic K.b. blooms was quite sensitive to small variations in the toxin grazing deterrence factor β_3 (Fig. 5A–F). Plots of the long term (5500–6000 d) K.b. bloom response as a function of β_3 showed that K.b. blooms did not occur at $\beta_3 \leq 1.12$ but did occur at $\beta_3 \geq 1.13$, and the K.b. biomass and brevetoxin concentration was unchanged with further increases in β_3 (Fig. 5C and D). Thus, the transition from no K.b. bloom to an intense, persistent toxic bloom occurred over a change of β_3 of < 1%. However, increases in β_3 above 1.13 decreased the time needed for K.b. bloom formation, and decreased the number of pre-blooms of diatoms (data not shown).

3.2.2. Bottom-up effects

The model bloom dynamics were also highly sensitive to the dilution rate (Figs. 6 and 7), which along with the inflowing NH$_4^+$ concentration, controls the NH$_4^+$ input rate to the system (Fig. 1).

Model runs were made at our standard dilution rate (0.1 d^{-1}; Fig. 4I–L), and also at lower (0.05 d^{-1}; Fig. 6A–D) and higher dilutions rates (0.30 d^{-1}; Fig. 6E–H). Decreasing the dilution rate increased the likelihood of a K.b. bloom, and decreased the time needed for bloom formation and the number of pre-blooms of diatoms (Figs. 4I–L and 6). For example, at a dilution rate of 0.05 d^{-1}, there were single pre-blooms each of T.p. and T.w. followed by a persistent K.b. bloom after ~50 d (Fig. 6C), while at a dilution rate of 0.3 d^{-1}, there were only successive diatom blooms and no K.b. bloom formation (Fig. 6G).

As with variations in the grazing deterrence factor, K.b. bloom formation was highly sensitive to small changes in dilution rate. In our long term model runs (5500–6000 d) under the standard β_3 value (1.5 mol C [g brevetoxin]$^{-1}$), there was an abrupt occurrence of K.b. blooms at dilution rates <0.17 d^{-1} and only recurring diatom blooms at higher dilution rates ($\geq$0.18 d^{-1}), whose species composition varied with changes in dilution rate (Fig. 7D–F).
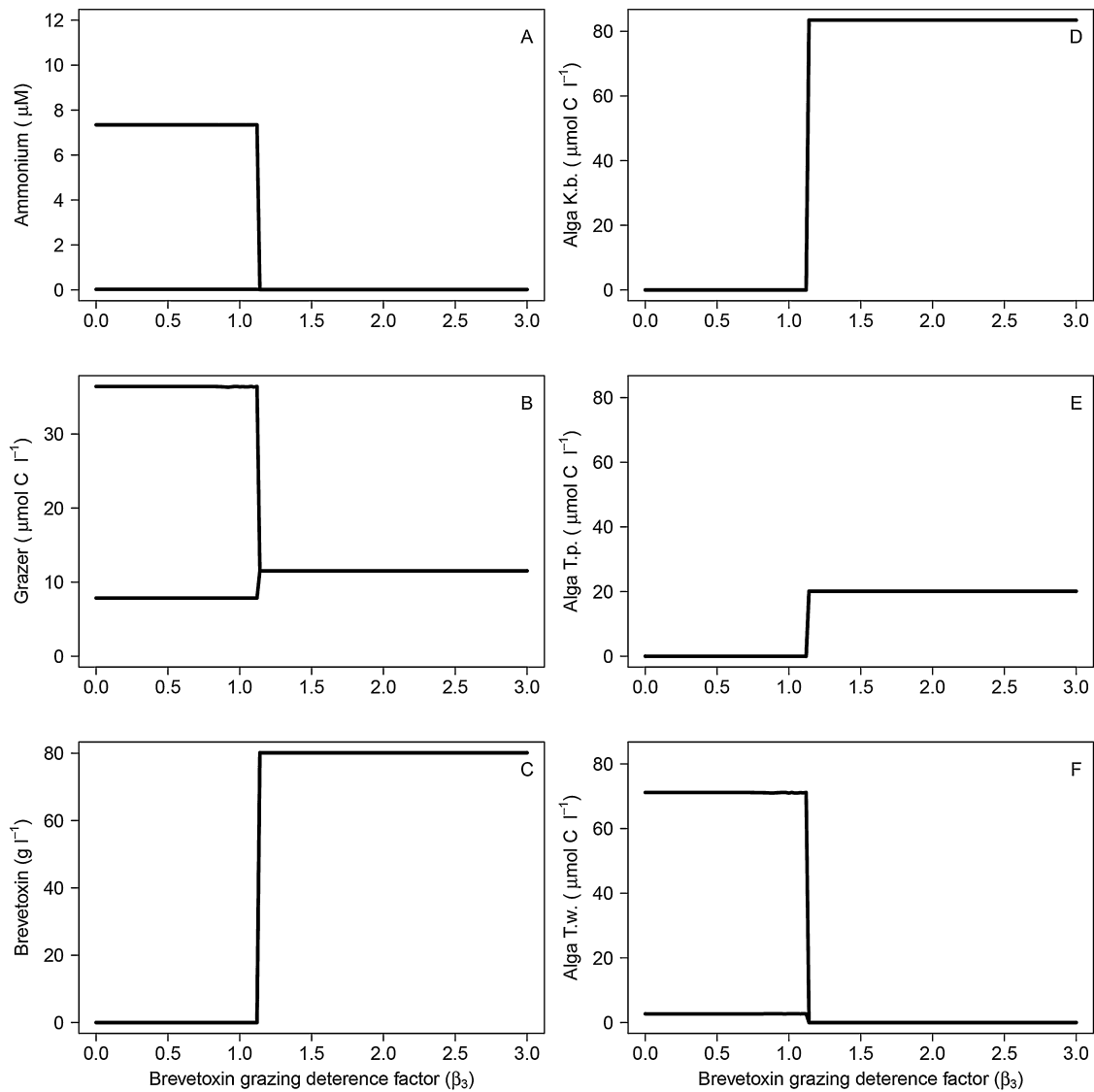
Fig. 5. Bifurcation diagram showing the long-term response of the model and its various model parameters (NH$_4^+$ concentration, grazer biomass, total brevetoxin concentration, and the carbon biomass of the three algal species) to changes in the brevetoxin grazing deterrence factor β_3. The results were computed from extended model runs (5500–6000 d) and show stable long-term dynamics. The grazing deterrence factor (β_3) controls the relationship between the maximum grazing rate on *K. brevis* and the cellular brevetoxin:C ratio (Eq. (6)), with higher values corresponding to stronger inhibition of grazing rates. Within that long-term 500 d window, model dynamics either achieved a steady state (single response line) or stable oscillations (two lines showing the upper and lower bounds). All other model parameters are the same as in Fig. 4. Algal species are *K. brevis* (K.b.), *T. pseudonana* (T.p.), and *T. weissflogii* (T.w.).

Within the region of stable K.b. bloom formation, decreases in dilution rates from 0.16 to 0.01 d^{-1}, were accompanied by a decrease in NH$_4^+$ concentration from 0.031 to 0.005 μM and a decrease in specific growth rate of K.b. from 0.16 to 0.01 d^{-1}, or from 40% to 3% of the maximum growth rate (see Fig. 3A). This increase in N-limitation of growth rate was accompanied by a 2.8-fold increase in K.b. C biomass (Fig. 7D) and a 4-fold increase in bloom toxicity (g brevetoxins l^{-1} water) (Fig. 7C). Thus, our model showed that decreasing dilution rate favors K.b. bloom formation and can greatly increase bloom biomass and toxicity.

As expected, increasing the nutrient concentration in the inflowing water substantially affected bloom biomass and dynamics. A six-fold increase in NH$_4^+$ concentration increased the final K.b. bloom biomass and brevetoxin levels by six-fold, but it also increased the time required for bloom initiation by about three-fold (Fig. 8C, F, D and H). Furthermore, at the lower NH$_4^+$ level, there was an initial bloom of T.p. (which always occurs), followed by a single bloom of T.w., which rapidly subsided and was replaced with simultaneous increases in K.b. and a three-fold lower

biomass of T.p. (Fig. 8C). The biomasses of these species rapidly reached steady state conditions. By contrast, at the higher NH$_4^+$ concentration, there were several pre-blooms of T.w. after the initial T.p. bloom, followed by co-occurring blooms of K.b and T.w. and then an even higher steady state bloom of K.b. and lesser amounts of T.p. (Fig. 8F). Thus, while the relationship between inflowing nutrient concentration and final bloom density may be straight forward, that between nutrient concentration and bloom dynamics can be quite complex.

4. Discussion

4.1. A quantitative test of the positive feedback hypothesis

Our modeling effort was undertaken to test the positive feedback hypothesis of Sunda et al. (2006), which proposed that ecosystem disruptive algal blooms (EDABs) resulted from complex interactions among bottom-up control by nutrients, top-down control by grazers, and positive feedbacks involving grazer-mediated nutrient
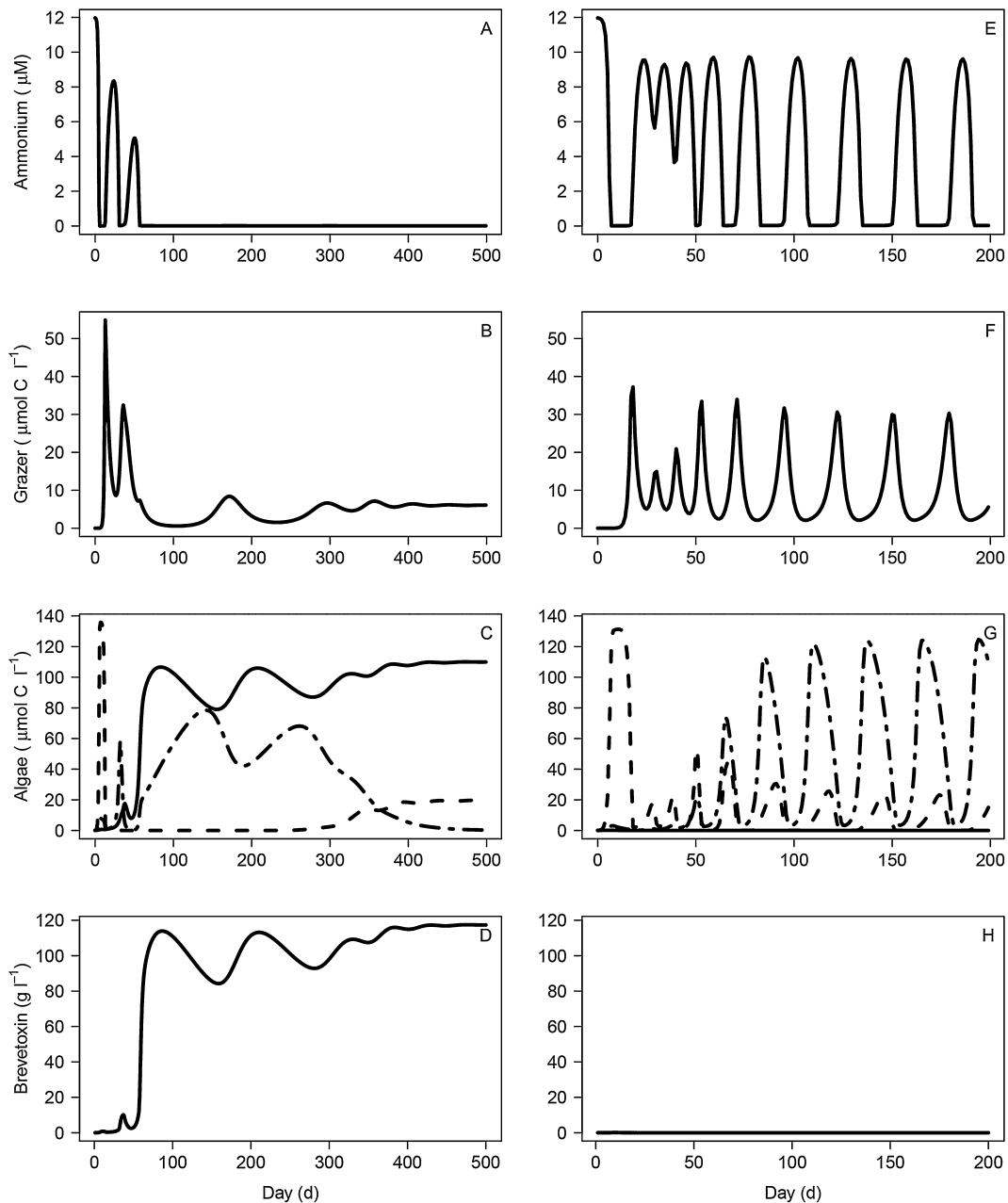
Fig. 6. Time course simulations at low and high dilution rates: (A, B, C, D) with dilution rate $\delta = 0.05$ d^{-1}, and (E, F, G, H) with $\delta = 0.30$ d^{-1}. All other model parameters were set their base values (S.M., Appendix 1), including the brevetoxin grazing deterrence factor ($\beta_3 = 1.5$) and inflowing NH$_4^+$ concentration ($N_{in} = 12$ μM). In panels C and G, the algal species are *K. brevis* (solid line), *T. pseudonana* (dashed line), and *T. weissflogii* (short-long dashed line). Note that the time scale is shorter in the right hand column.

recycling. Sunda et al. (2006) noted that EDAB algae are usually low-nutrient adapted (*K*-selected) species that are poorly grazed by herbivores due to their robust grazing defenses such as toxins. They further noted that toxin levels and other grazing defenses often increase with nutrient limitation of growth rate, and that this would further promote positive feedback interactions during bloom formation and would increase bloom toxicity. In a previous paper (Sunda and Shertzer, 2012), we used an NPZ model to examine Sunda et al.'s (2006) first positive feedback hypothesis in which the EDAB species has robust grazing defenses, which do not vary with nutrient limitation of growth rate. In the present model we examined Sunda et al.'s (2006) second feedback hypothesis, in which EDAB formation was promoted by grazing defenses related to cellular toxins where the cellular toxin levels (and grazing defenses) increased with nutrient limitation of growth rate.

Sunda et al. (2006) proposed that EDAB events required high inputs of utilizable nutrients, which would initially stimulate the growth of diatoms or other high-nutrient adapted (r-selected) species. They hypothesized that the pre-bloom of such r-selected species would draw down nutrients to low concentrations, setting the stage for a competitive switch to the EDAB species. In our current simulations, one or more pre-blooms of high-nutrient (r-selected) diatoms preceded every K.b. bloom, and were a prerequisite for toxic bloom formation (Figs. 4K, 6C, 8C and 8F). The pre-bloom(s) had three important effects: (1) their high nutrient demand drew nutrients (NH$_4^+$) to low levels (0.008–0.022 μM), which limited the growth rate of all algal species, but had a larger relative adverse effect on the growth rate of the diatoms (see Fig. 3A); (2) the resulting N-limitation of growth rate of K.b. increased cellular toxins, thereby further decreasing grazing rates on the EDAB species (Fig. 2A and B); and (3) the diatom
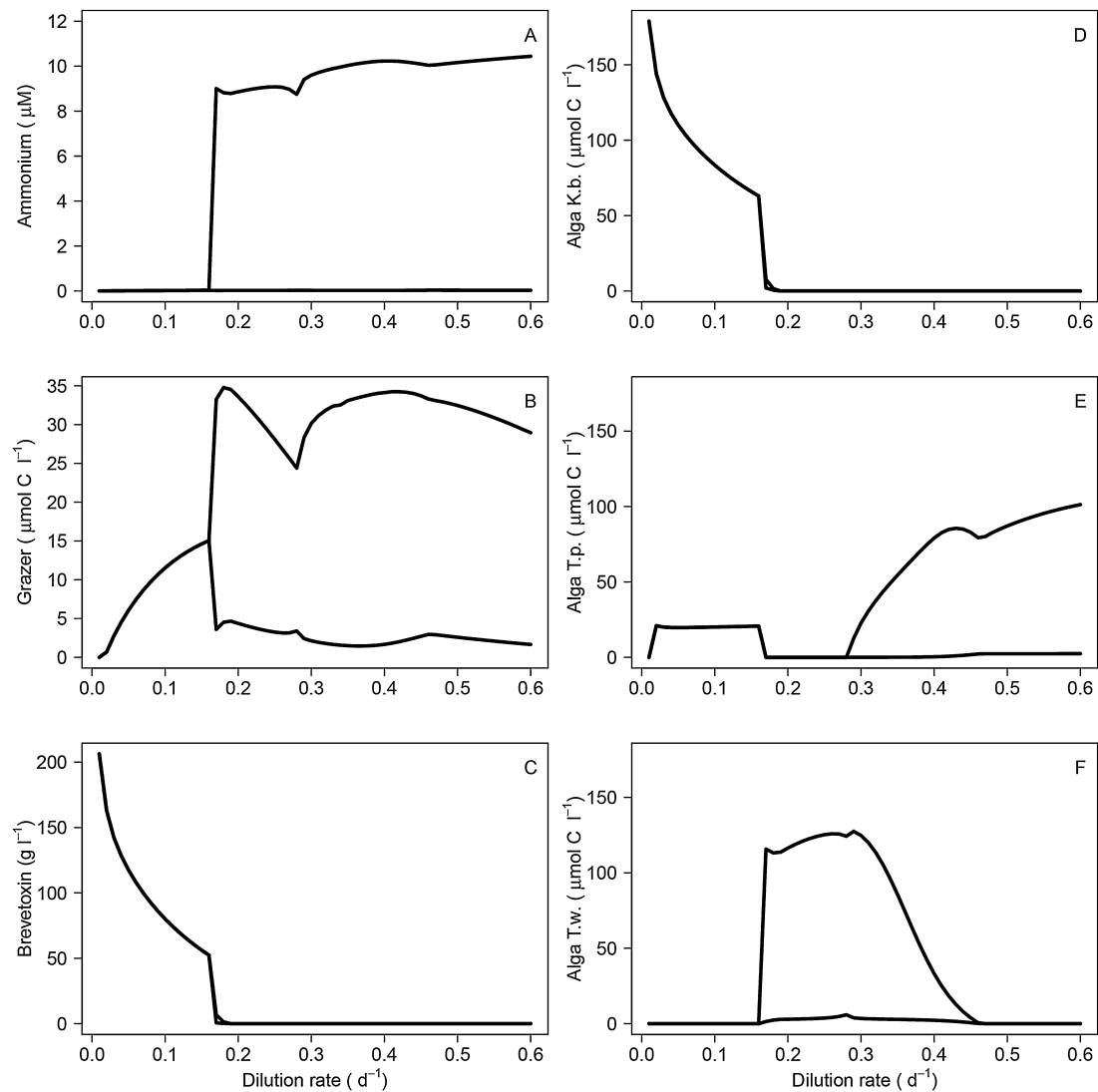
Fig. 7. Bifurcation diagram showing the effect of changing dilution rate on stable long-term bloom dynamics (computed from extended model runs, days 5500–6000). Within that long-term 500 d window, model dynamics either achieved a steady state (single response line) or stable oscillations (two lines showing the upper and lower bounds). All other model parameters were the same as in Fig. 4I–L. Algal species in D–F are *K. brevis* (K.b.), *T. pseudonana* (T.p.), and *T. weissflogii* (T.w.).

blooms stimulated the growth rate of the grazer, ultimately leading to high grazer populations and high grazing mortality rates for the diatoms. Because K.b. was poorly grazed, the high grazing rates on the diatoms, in conjunction with their larger relative decrease in specific growth rate at low nutrient concentrations, allowed K.b. to out-compete these algae, and thereby form a toxic bloom. The high diatom grazing rates also promoted high rates of grazing-linked nutrient recycling, which directly fed the emerging toxic dinoflagellate bloom.

Pre-blooms of diatoms or other high-nutrient adapted species that drew dissolved nutrients to low concentrations have been observed in most EDAB events for which such data are available, including brown tide blooms of *Aureococcus anophagefferens* in coastal bays of Long Island, New York (Gobler and Sanudo-Wilhelmy, 2001; Sunda et al., 2006), the massive toxic bloom of *Chrysochromulina polylepis* that occurred in coastal waters of southern Scandinavia in 1988 (Maestrini and Graneli, 1991), and a bloom of the dinoflagellate *Gymnodinium instriatum* in the Lafaette River, Virginia, a tributary of the Chesapeake Bay (Egerton et al., 2014). In a brown tide bloom of *A. anophagefferens* in Great South Bay, Long Island, New York, USA during 1999, the addition of inorganic nutrients (N and P) increased the specific growth of the

algal community by 36% on average in six experiments, indicating that the growth of the algae was nutrient-limited (Gobler et al., 2002). Also during the nearly eight year brown tide bloom of *Aureoumbra lagunensis* in the Laguna Madre, Texas, the mean particulate C:N molar ratio (28 ± 8) and cellular chlorophyll *a* (0.019 ± 0.002 pg cell) were both indicative of highly N-limited cells based on data for cellular C:N and chlorophyll *a* cell^{-1} vs specific growth rate from NH$_4^+$-limited chemostats (DeYoe and Suttle, 1994; Liu et al., 2001). In addition, low dissolved inorganic and organic P levels were observed during a bloom of the toxic dinoflagellate *Karenia mikimotoi* in the East China Sea (Li et al., 2009). Phosphate additions significantly stimulated algal N uptake rates, suggesting the bloom growth was P-limited. Finally in a four year survey on the West Florida Shelf, *Karenia brevis* cell densities were inversely correlated with concentrations of dissolved inorganic nitrogen and urea (a highly available DON molecule) and were directly correlated with the abundance of haptophytes and cyanobacteria, two groups of low-nutrient adapted phytoplankton (Dixon et al., 2014). By contrast, the abundance of *K. brevis* was inversely correlated with that of diatoms (a high nutrient adapted algal group) as also seen in our present model simulations. Thus, there is considerable evidence for depletion of dissolved nutrients to growth rate limiting levels
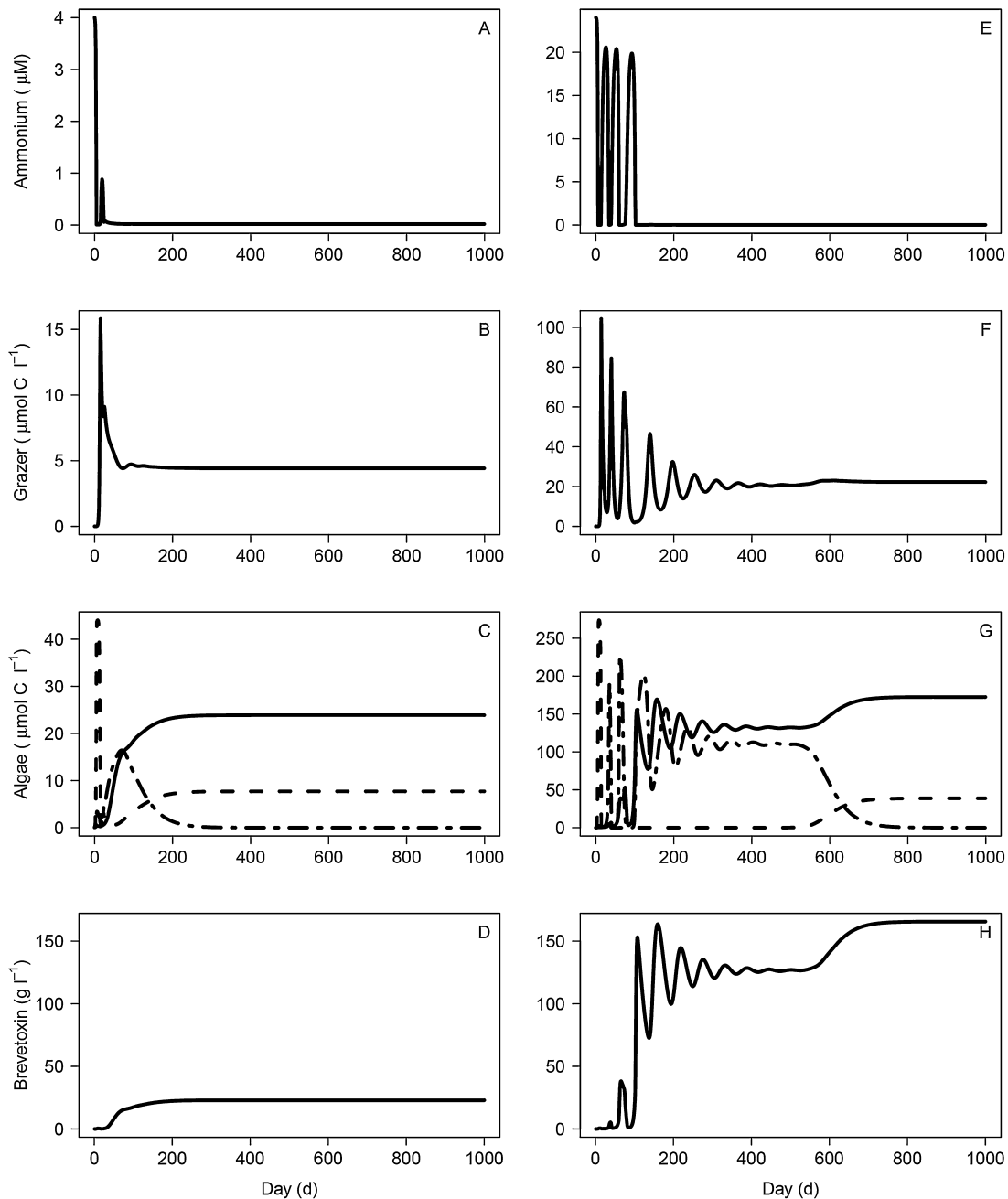
Fig. 8. Two time-course simulations, one with an NH_4^+ input concentration N_{in} = 4 μM (A–D), and one with N_{in} = 24 μM (E–H). All other model parameters were set at their base values (S.M, Appendix 1), including the brevetoxin grazing deterrence factor (β_3 = 1.5) and dilution rate (0.1 d^{-1}). The y-axis scales are six-fold greater for simulations with N_{in} = 24 μM than for N_{in} = 4 μM. In panels C and G, the algal species are *K. brevis* (solid line), *T. pseudonana* (dashed), and *T. weissflogii* (short-long dashed).

during both pre-blooms and EDAB events. As in our current models, the total concentration of the limiting nutrient element (N or P) remained high, but most became tied up in the bloom algae.

Sunda et al. (2006) proposed that a combination of an ability to grow more competitively at low levels of limiting nutrients and low grazing mortality rates would allow EDAB species to proliferate relative to populations of competing phytoplankton at the low dissolved nutrient concentrations brought on by pre-blooms of other species. The increased abundance of poorly-grazed EDAB species would further decrease grazing rates and thereby decrease grazer populations and grazer-mediated nutrient recycling. This decreased inputs of recycled nutrients, which in turn would decrease available nutrient concentrations, and increase nutrient limitation of growth rate in all species and cellular toxins

in the EDAB species. The increased toxicity would further decrease grazing rates on the EDAB species, which would further increase its net population growth rate. Sunda et al. (2006) hypothesized that these interactions constituted a positive feedback that would promote the formation, persistence, and toxicity of EDAB events, with co-occurring low levels of dissolved nutrients, algal grazers, and competing phytoplankton. These predictions are verified in our current model simulations (Figs. 4I–L, 6A–D and 8A–H). However, for the K.b. bloom to persist in our simulations, there had to be co-occurring populations of diatoms to support the grazer population. Without the grazer (Fig. 4A–D) or preferential grazing on the diatoms (Fig. 4E–H), K.b. blooms were not possible and the model system became dominated by diatoms.

The model simulations presented here and previously (Sunda and Shertzer, 2012) reveal an additional positive feedback that promotes the formation of EDABs, linked to decreases in cellular N:C ratios and N recycling rates with N-limitation of algal growth rates. The N:C ratios in diatoms and K.b. cells whose growth rates are severely N-limited are up to two-fold lower than those in optimally growing N-sufficient cells (Fig. 3B). Consequently, as algal growth rates become N-limited during the formation of the K.b. bloom, the N:C ratio in all of the algae consumed by the grazer decreases, which decreases the excess assimilated N available for excretion by the zooplankton. Hence, the low algal N:C ratios in the N-limited algae reduces N excretion rates and grazing-linked N-recycling rates, thereby further decreasing NH_4^+ concentrations and cellular N:C ratios in the consumed algae. Such positive feedbacks linked to algal nutrient:C stoichiometry have been previously described, at least in terms of their effect on the nutritional quality of algal prey and adverse effects on the growth rate of grazer species (Irigoien et al., 2005; Mitra and Flynn, 2006). Similar stoichiometric feedback mechanisms would be expected with other limiting nutrients such as phosphorous.

Another feature of the current stoichiometric NPZ model that was not predicted in the conceptual feedback model of Sunda et al. (2006) (and was not dealt with in our former EDAB model simulations; Sunda and Shertzer, 2012) was the impact of nutrient limitation of specific growth rate of the EDAB algae on bloom toxicity. Our current model shows that toxic red tide blooms are accompanied in every case by an increase in the fraction of total limiting nutrient (N) residing in the biomass of the toxic EDAB species at the expense of other nutrient reservoirs, including dissolved nutrients (NH_4^+), and N present in competing algae and grazers. This by itself increased EDAB biomass and toxicity. In addition, as predicted (Sunda et al., 2006), every K.b. bloom was accompanied by low dissolved nutrient (NH_4^+) levels and associated N-limitation of algal specific growth rates. This N-limitation had two effects that substantially promoted bloom toxicity. First, it increased C:N ratios in the toxic species, which increased C-biomass of these algae by up to two-fold for a given total N concentration in the system (see Fig. 3B). Second, it increased cellular toxin:C ratios in K.b. by up to three-fold relative to those in N-sufficient algal cells (Fig. 2A). These two factors are multiplicative and together have the potential to increase brevetoxins per liter of seawater by up to six-fold relative to what would have occurred if the specific growth rate of the toxic algae was not N-limited.

In our present simulations at the standard brevetoxin grazing deterrence factor (β_3 = 1.5 mol C [g brevetoxin]$^{-1}$), as the dilution rate increased above ~0.18 d^{-1}, K.b. blooms disappeared and were replaced by repeating diatom blooms, first of T.w. by itself, then T.w. and T.p. together, and finally T.p. alone at the highest dilution rates (>0.45 d^{-1}) (Fig. 7D–F). Thus, the dominant bloom species was highly dependent on the dilution rate, with low dilution rates (long water residence times) favoring K.b. blooms and high dilution rates favoring blooms of the fast growing diatom T.p. The reason for this effect is that decreasing dilution rates decrease inputs of "new" nutrients, which both increases nutrient limitation of growth rate (via decreases in NH_4^+ concentrations) and increases the relative importance of grazer-linked nutrient recycling to the overall supply of nutrients. Both effects promote the positive feedbacks described above that give rise to EDAB events. Furthermore, low dilution rates also increase bloom intensity in terms of K.b. C-biomass (g l^{-1}) (Fig. 7D) and bloom toxin concentrations (g toxin l^{-1}) (Fig. 7C) through decreases in NH_4^+ concentrations and increases in N-limitation of specific growth rate. This model prediction is consistent with K.b. bloom patterns on West Florida Shelf during a four year survey (2007–2010), where the most intense K.b. bloom occurred during the driest year (2007) when inputs of "new" nutrients from coastal runoff were at their lowest (Dixon et al., 2014).

An interesting feature of the current model was the sensitivity of the simulated red-tide bloom to small variations in the toxin grazing deterrence factor (β_3) (Fig. 5D) and the dilution rate of the system (Fig. 7D). Although, the conceptual model of Sunda et al. (2006) had predicted that decreasing grazing rates on EDAB species and decreasing system dilution rates would both favor EDAB events, our current NPZ model sheds light on just how sharp the transitions can be between recurring diatom blooms and persistent EDAB events with changes in these parameters. Similar sharp transitions between blooms of diatoms and those of EDAB species with decreasing grazing rates on the EDAB species and decreasing system dilution rates were also observed in our previous simulations of brown tide blooms (Sunda and Shertzer, 2012). These sharp transitions were attributed to the positive feedbacks that occur during initiation of EDAB events. However, the transitions in the current model were more abrupt than observed previously (Figs. 5D and 7D), which we attribute to the added positive feedback effect of increased cellular toxins and grazing deterrence in the EDAB species with nutrient limitation of specific growth rate.

The nutrient concentration in the incoming water also affected bloom biomass and dynamics as expected. Increasing the incoming NH_4^+ concentration increased the intensity of the toxic bloom in terms of both biomass and total toxin concentration, but it also delayed the occurrence of the toxic bloom, and increased the number of pre-blooms of diatoms (Fig. 8C–H). Thus, increasing the nutrient inputs delayed the K.b. bloom and made it somewhat less likely to occur; however, when it did occur, its biomass and toxicity were much greater. Such higher toxicity blooms fueled by high nutrient inputs would be accompanied by more severe negative effects, such as disruption of food webs, increased fish kills, and adverse impacts on human health and coastal economies.

4.2. Model generalization and assumptions

Based on the arguments in Sunda et al. (2006), the positive feedback dynamics that promoted toxic blooms of K.b. in our current NPZ model should also occur in many other EDAB species whose toxicity and/or grazing defenses increase with nutrient limitation of growth rate. Such increases in toxins and defenses with nutrient limitation are widespread in marine algae and also occur widely in terrestrial plants (Lambers et al., 2006; Hardison et al., 2013). For example, hemolytic activity in the toxic, bloom-forming prymnesiophytes *Prymnesium parvum* and *Chrysochromulina polylepis* increases by up to 10-fold under N- or P-limitation of growth rate (Johansson and Graneli, 1999a, 1999b). Similarly, silica- and P-limitation increases cell quotas of the neurotoxin domoic acid in the diatom *Pseudo-nitzschia multiseries* (Pan and Rao, 1996; Pan et al., 1998; Sun et al., 2011). In addition, N- and P-limitation increases cellular karlotoxins by 2- to 15-fold in the dinoflagellate *Karlodinium veneficum*, which has caused fish-killing blooms (Adolf et al., 2009; Fu et al., 2010) and P-limitation causes over 10-fold increases in cell quotas of the toxin nodularin in the N-fixing cynaobacterium *Nodularia spumigena*, which has formed massive blooms in the Baltic Sea (Sunda et al., 2006). Thus, while our current model was constructed for *Karenia brevis*, similar positive feedback models should also apply to many other bloom-forming, toxic species. Indeed, we chose K.b. for the present model, not because we thought the positive feedback model of Sunda et al. (2006) and Mitra and Flynn (2006) would be particularly applicable, but rather because of the availability of recent data for brevetoxin:C vs specific growth rate in N-sufficient and N-limited algae (Hardison et al., 2012) and the availability of data demonstrating the grazing deterrent effects of brevetoxins (Hong et al., 2012). In this regard, K.b. represents a generic toxic, EDAB

species whose cellular toxin levels increase with nutrient limitation of cellular growth rate.

Our NPZ model was deliberately simplified to capture the often complex interactions among several basic processes and factors that influence the formation and persistence of virtually all harmful algal blooms. It shows that nutrient-linked differences in the growth of algal species, depletion of dissolved nutrients by algal blooms, increased cellular toxicity with nutrient limitation, decreased grazing rates linked to cell toxicity, and decreases in nutrient regeneration rates caused by decreasing grazing rates and lower cell N:C ratios lead to positive feedbacks that promote the occurrence, persistence, and toxicity of EDAB events. Thus, our model represents a useful tool for examining the ecosystem dynamics of toxic blooms of dinoflagellates and other toxic EDAB species, and for identifying key mechanisms and factors that influence bloom dynamics and toxicity. It also demonstrates that even a deliberately simplified model system can exhibit quite complex behavior.

However, we also recognize that our simplified model ignores many factors and processes that can influence blooms of K.b. or other toxic EDAB species. One simplification was the use of a single biologically available form of N (NH_4^+), when indeed many other forms of N – nitrate, labile N-containing organic molecules (e.g., urea and amino acids) and complex mixtures of more biologically refractory organics (e.g., proteins and humic materials) – are important N-sources to marine phytoplankton (Bronk et al., 2007; Wawrik et al., 2009). A sole N source (NH_4^+) was used not only to simplify the model, but also because quantitative data for relationships between N uptake and assimilation rates and concentrations of other N-containing substrates are currently not available for any algal species. However, the positive feedback mechanisms described by Sunda et al. (2006) and Mitra and Flynn (2006) should be independent of the type or chemical form of the limiting nutrient, so it seems likely that our NPZ model would have provided qualitatively similar results if the form of inflowing N was different or if a mixture of N substrates had been used.

One could argue that our model would be more realistic if the N-substrate introduced into the system was nitrate (NO_3^-) rather than NH_4^+ since NO_3^- is the predominant form of utilizable inorganic N introduced into surface waters via upwelling, vertical mixing, and river inputs (Falkowski and Raven, 2007). The initial bloom species following external NO_3^- inputs are typically diatoms, which utilize the high NO_3^- levels and their high maximum specific growth rates to support bloom formation (Margalef, 1978; Bronk et al., 2007). In culture studies, the maximum specific growth rate (μ_{max}) of K.b. was similar for growth on NH_4^+ ($0.37\ d^{-1}$) and NO_3^- (0.35^{-1}) (Glibert et al., 2009) and the μ_{max} values for our coastal diatoms (T.p. and T.w.) grown on the two substrates were also similar to one another (Sunda and Huntsman, 1995; Maldonado and Price, 1996; Sunda and Hardison, 2007). So replacing NO_3^- with NH_4^+ as our inflowing limiting nutrient would not have substantially affected our results. We still would have seen pre-blooms of diatoms that drew nutrients down to low concentrations, whether the inflowing limiting nutrient was NH_4^+ or NO_3^-.

There are several other factors not included in our simplified model that would promote the growth of K.b. and other toxic dinoflagellates, but would not affect that of diatoms and other non-motile, r-selected species. One such factor is the vertical migration of K.b. and other dinoflagellates driven by the diel light/dark cycle and varying nutrient concentrations with depth (Kamykowski and Yamazaki, 1997; Sinclair and Kamykowski, 2008). In this diel cycle, dinoflagellates vertically migrate downward at night to access higher nutrient concentrations at depth, and migrate back to near the surface during the day to access light needed for photosynthesis. This diel migration pattern increases the nutrient uptake and

growth of dinoflagellates in stratified coastal systems where surface nutrient concentrations have been depleted by algal growth, and thus, it tends to promote dinoflagellate blooms (Kamykowski and Yamazaki, 1997). However, it would not allow toxic dinoflagellates to out-compete diatoms during pre-bloom conditions when surface nutrient concentrations are sufficiently high to support maximum growth rates of all competing algal species.

A second widespread growth enhancing factor for many HAB/EDAB species is mixotrophy (Burkholder et al., 2008), including phagotrophy, the ingestion of other microorganisms (Li et al., 2000; Jeong et al., 2005a; Stoecker et al., 2006) and osmotrophy, the utilization of dissolved organic molecules (Glibert and Legrand, 2006). Both processes can supply fixed carbon and nutrient elements (e.g., N and P) to supplement that supplied from photosynthetic C-fixation and from cellular uptake of inorganic nutrients (Adolf et al., 2006). Both osmotrophy and phagotrophy often increase substantially under nutrient- or light-limitation of growth rate (Smalley et al., 2003; Stoecker et al., 2006; Burkholder et al., 2008).

Osmotrophic utilization of DON as an N source is widespread in marine algae and is particularly prevalent in many low-nutrient, K-adapted species, including *Karenia brevis* (Sipler et al., 2013) and many other HAB/EDAB species (Bronk et al., 2007; Glibert and Legrand, 2006; Burkholder et al., 2008). Biologically labile DON molecules, such as urea and amino acids, are often an important component of grazer-mediated regenerated N so biological utilization of DON can be important source of recycled N to phytoplankton. Furthermore, there is evidence that the release of DON by *Trichodesmium* or other diazotrophic cyanobacteria can promote the growth of *K. brevis* blooms (Sipler et al., 2013). However, such increase in fixed-N supply from N_2-fixation can drive these systems into P-limitation of algal growth rate, which also increases cellular brevetoxin:C ratios (Hardison et al., 2013).

Phagotrophy occurs widely in many groups of phytoplankton, and is particularly common in low-nutrient, K-selected algae, including many toxic HAB/EDAB species (Stoecker et al., 2006; Burkholder et al., 2008; Flynn et al., 2013). It is widespread in dinoflagellates, prymnesiophytes, and chrysophytes, but is not known to occur in diatoms (Stoecker et al., 2006; Jeong et al., 2010; Flynn et al., 2013). Phagotrophic consumption of the cyanobacterium *Synechococcus* sp. by *Karenia brevis* has been observed in cultures (Jeong et al., 2005b; Glibert et al., 2009), and virtually all dinoflagellates examined to date have been shown to be phagotrophic, including many formerly thought to be pure phototrophs (Jeong et al., 2005a,b).

The inclusion of DON utilization or of phagotrophic consumption of competing algae in the present model or in similar models involving other phagotrophic EDAB species (Jeong et al., 2010) would almost certainly affect predicted bloom dynamics. The inclusion of regenerated or diazotrophically produced DON should favor the formation of harmful blooms of *Karenia brevis* and other EDAB/HAB species which are known to effectively utilize various organic forms of N (Burkholder et al., 2008; Sipler et al., 2013). Phagotrophy would also promote toxic blooms because it both increases the supply of nutrients and carbon to support population growth of the EDAB species, and also removes competing algal species (Glibert et al., 2009; Jeong et al., 2010). These effects in turn would further reduce zooplankton grazing rates and populations, and grazer-mediated recycling of nutrients. These dynamics represent an additional potential positive feedback that could help promote toxic blooms, which would operate in conjunction with the ones in our current model. At least initially, the added nutrient assimilation from phagatrophy could moderate toxin increases linked to nutrient limitation of specific growth rate. However, eventually the increasing population growth of EDAB species and decline in more readily grazed competing algae would

decrease grazing linked nutrient recycling and the supply of nutrients to all utilizable pools (DIN, DON, and consumable phytoplankton) needed to support further toxic bloom growth. As in our current model, the specific growth rate of the EDAB algae would then become increasingly nutrient-limited, leading to increases in their toxin:C and C:N cell ratios and a high biomass, highly toxic bloom. Evidence for such a scenario comes from a *K. brevis* bloom on the west Florida shelf, where *Synechococcus* was abundant outside of the bloom, but was reduced to nearly undetectable levels within the bloom proper, presumably because of phagotropic consumption by the toxic dinoflagellate (Heil et al., 2007; Glibert et al., 2009).

5. Conclusions

Our NPZ model indicates that the formation of ecosystem disruptive algal blooms (EDABs) depends critically on both bottom-up and top-down trophic effects, and feedback inter-actions between the two. It shows that positive feedbacks linked to increased toxicity of EDAB species under nutrient limitation of growth rate can promote the occurrence and toxicity of ecosystem disruptive algal blooms, such as those of *Karenia brevis*. In the model, every toxic dinofalgellate bloom was preceded by one or more pre-blooms of diatoms, whose specific growth rates were much higher than those of the toxic species at high nutrient levels, but were also more readily grazed by zooplankton. The diatom pre-blooms drew down available nutrients to low, growth rate-limiting levels and promoted the population growth of zooplankton. The EDAB species was then able to proliferate at the expense of the diatoms owing to its low grazing mortality rate and to a much smaller decrease in its specific growth rate relative to diatom growth rates as dissolved nutrient levels declined. A positive feedback occurred because the resultant higher abundance of N-limited, more highly toxic and poorly-grazed EDAB species decreased grazing rates and grazing-linked nutrient recycling, which further decreased dissolved nutrient levels, and thereby further increased toxin levels in the EDAB species and decreased diatom growth rates relative to growth rates of the EDAB algae. This positive feedback led to a high biomass bloom, in which the specific growth rate of the EDAB algae was increasingly restricted by low nutrient levels. This low nutrient stress increased both C:N and toxin:C ratios in the EDAB algae, and thus it also greatly increased bloom carbon biomass and toxicity.

Decreasing dilution rates (increasing water residence times) in our model system promoted harmful bloom formation and toxicity by decreasing the input of "new" nutrients to the system, which both increased nutrient limitation of algal specific growth rates and increased the importance of recycling to the overall supply of limiting nutrients. Thus, our model predicts that increases in water residence times in coastal systems (e.g., from climate change related droughts or river water diversion for agriculture) could increase the occurrence and toxicity of EDABs.

The model also indicates that bloom biomass and toxicity is the fundamentally limited by the concentration of inputted nutrients. Thus, to minimize bloom toxicity and harmful impacts, environmental managers and regulators need to reduce nutrient inputs into coastal systems, especially those from anthropogenic sources such as agricultural fertilizers, municipal wastes, and NO_x from fossil fuel burning. The positive feedback dynamics described here should not only apply to blooms of our model EDAB species *Karenia brevis*, but also to other EDAB algae, whose toxins and grazing deterrence increase with nutrient limitation of growth rate.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.hal.2014.09.005.

References

Adolf, J.E., Bachvaroff, T.R., Place, A.R., 2009. Environmental modulation of karlo-toxin levels in strains of the cosmopolitan dinoflagellate, *Karlodinium veneficum* (Dinophyceae). J. Phycol. 45, 176–192.

Adolf, J.E., Stoecker, D.K., Harding, L.W., 2006. The balance of autotrophy and heterotrophy during mixotrophic growth of *Karlodinium micrum* (Dinophy-ceae). Pankton Res. 28, 1–15.

Bronk, D.A., See, J.H., Bradley, P., Killberg, L., 2007. DON as a source of bioavailable nitrogen for phytoplankton. Biogeosciences 4, 283–296.

Burkholder, J.M., Glibert, P.M., Skelton, H.M., 2008. Mixotrophy, a major mode of nutrition for harmful algal species in eutrophic waters. Harmful Algae 8, 77–93.

Buskey, E.J., 2008. How does eutrophication affect the role of grazers in harmful algal bloom dynamics? Harmful Algae 8, 152–157.

Calbet, A., Landry, M.R., 2004. Phytoplankton growth, microzooplankton grazing, and carbon cycling in marine systems. Limnol. Oceanogr. 49, 51–57.

Calbet, A., Vaqué, D., Felipe, J., Vila, M., Sala, M.M., Alcaraz, M., Estrada, M., 2003. Relative grazing impact of microzooplankton and mesozooplankton on a bloom of the toxic dinoflagellate *Alexandrium minutum*. Mar. Ecol. Prog. Ser. 259, 303–309.

Caron, D.A., Goldman, J.C., 1990. Protozoan nutrient regeneration. In: Capriulo, G.M. (Ed.), Ecology of Marine Protozoa. Oxford University Press, New York, pp. 283–306.

Chen, B., Liu, H., Lau, M.T.S., 2010. Grazing and growth responses of a marine oligotrichous ciliate fed with two nonoplankton: does food quality matter for micrograzers? Aquat. Ecol. 44, 113–119.

Cohen, J.H., Tester, P.A., Forward, R.B., 2007. Sublethal effects of the toxic dinofla-gellate *Karenia brevis* on marine copepod behavior. J. Plankton Res. 29, 301–315.

DeYoe, H.R., Suttle, C.A., 1994. The inability of the Texas "brown tide" alga to use nitrate and the role of nitrogen in the initiation of a persistent bloom of this organism. J. Phycol. 30, 800–806.

Dixon, L.K., Kirkpatrick, G.J., Hall, E.R., Nissanka, A., 2014. Nitrogen, phosphorus and silica on the West Florida Shelf: patterns and relationships with *Karenia* spp. occurrence. Harmful Algae, http://dx.doi.org/10.1016/j.hal.2014.07.001.

Droop, M.R., 1968. Vitamin B_{12} and marine ecology. IV. The kinetics of growth and inhibition in *Monochrysis lutheri*. J. Mar. Biol. Assoc. U.K. 48, 689–733.

Dugdale, R.C., Goering, J.J., 1967. Uptake of new and regenerated forms of nitrogen in primary productivity. Limnol. Oceanogr. 12, 196–206.

Egerton, T.A., Morse, R.E., Marshall, H.G., Mulholland, M.R., 2014. Emergence of algal blooms: the effects of short-term variability in water quality on phytoplankton abundance, diversity, and community composition in a tidal estuary. Micro-organisms 2, 33–57.

Falkowski, P.G., Raven, J.A., 2007. Aquatic Photosynthesis. Blackwell Science, Oxford 484 pp.

Flewelling, L.J., Naar, J.P., Abbott, J.P., Baden, D.G., Barros, N.B., Bossart, G.D., Bottein, M.Y.D., Hammond, D.G., Haubold, E.M., Heil, C.A., Henry, M.S., Jacocks, H.M., Leighfield, T.A., Pierce, R.H., Pitchford, T.D., Rommel, S.A., Scott, P.S., Steidinger, K.A., Truby, E.W., Van Dolah, F.M., Landsberg, J.H., 2005. Red tides and marine mammal mortalities. Nature 435, 755–756.

Flynn, K.J., Stoecker, D.K., Mitra, A., Raven, J.A., Glibert, P.M., Hansen, P.J., Granéli, E., Burkholder, J.M., 2013. Misuse of the phytoplankton–zooplankton dichotomy: the need to assign organisms as mixotrophs within plankton functional types. J. Plank. Res. 35, 3–11.

Frost, B.W., 1975. A threshhold feeding behavior in *Calanus pacificus*. Limnol. Oceanogr. 20, 263–266.

Frost, B.W., 1993. A modeling study of processes regulating plankton standing stock and production in the open subarctic Pacific Ocean. Prog. Oceanogr. 32, 17–56.

Fu, F.X., Place, A.R., Garcia, N.S., Hutchins, D.A., 2010. CO_2 and phosphate availability control the toxicity of the harmful bloom dinoflagellate *Karlodinium veneficum*. Aquat. Microb. Ecol. 59, 55–65.

Fussmann, G.F., Heber, G., 2002. Food web complexity and chaotic dynamics. Ecol. Lett. 5, 394–401.

Gentleman, W., Leising, A., Frost, B., Strom, S., Murray, J., 2003. Functional responses for zooplankton feeding on multiple resources: a review of assumptions and biological dynamics. Deep-Sea Res. II 50, 2847–2875.

Gismervik, I., 2005. Numerical and functional responses of choreo- and oligotrich planktonic ciliates. Aquat. Microb. Ecol. 40, 163–173.

Glibert, P.M., Anderson, D.M., Gentien, P., Graneli, E., Sellner, K.G., 2005. The global, complex phenomena of harmful algal blooms. Oceanography 18, 136–147.

Glibert, P.M., Burkholder, J.M., Dana, T.M., Alexander, J., Skelton, H., Shilling, C., 2009. Grazing by *Karenia brevis* on *Synechococcus* enhances its growth rate and may help to sustain blooms. Aquat. Microb. Ecol. 55, 17–30.

Glibert, P.M., Legrand, C., 2006. The diverse nutrient strategies of harmful algae: focus on osmotrophy. In: Graneli, E., Turner, J.T. (Eds.), Ecology of Harmful Algae. Springer, Berlin, pp. 163–175.

Gobler, C.J., Sanudo-Wilhelmy, S.A., 2001. Temporal variability of groundwater seepage and brown tide blooms in a Long Island embayment. Mar. Ecol. Prog. Ser. 217, 299–309.

Gobler, C.J., Renaghan, M.J., Buck, N.J., 2002. Impacts of nutrients and grazing mortality on the abundance of *Aureococcus anophageferrens* during a New York brown tide bloom. Limnol. Oceanogr. 47, 129–141.

Graneli, E., Flynn, K., 2006. Chemical and physical factors influencing toxin content. In: Graneli, E., Turner, J.T. (Eds.), Ecology of Harmful Algae. Springer, Berlin, pp. 229–241.

Jeong, H.J., Yoo, Y.D., Park, J.Y., Song, J.Y., Kim, S.T., Lee, S.H., Kim, K.Y., Yih, W.H., 2005a. Feeding by phototrophic red-tide dinoflagellates: five species newly revealed and six species previously known to be mixotrophic. Aquat. Microb. Ecol. 40, 133–150.

Jeong, H.J., Park, J.Y., Nho, J.H., Park, M.O., Ha, J.H., Seong, K.A., Jeng, C., Seong, C.N., Lee, K.Y., Yih, W.H., 2005b. Feeding by red-tide dinoflagellates on the cyanobacterium *Synechococcus*. Aquat. Microb. Ecol. 41, 131–143.

Jeong, H.J., Yoo, Y.D., Kim, J.S., Seong, K.A., Kang, N.S., Kim, T.H., 2010. Growth, feeding and ecological roles of the mixotrophic and heterotrophic dinoflagellates in marine planktonic food webs. Ocean Sci. J. 45, 65–91.

Hansen, P.J., Bjornsen, P.K., Hansen, B.W., 1997. Zooplankton grazing and growth: scaling within 2–2,000-μm body size range. Limnol. Oceanogr. 42, 687–704.

Hardison, D.R., Sunda, W.G., Litaker, R.W., Shea, D., Tester, P.A., 2012. Nitrogen limitation increases brevetoxins in *Karenia brevis* (dinophyceae): implications for bloom toxicity. J. Phycol. 48, 844–858.

Hardison, D.R., Sunda, W.G., Shea, D., Litaker, R.W., 2013. Increased toxicity of *Karenia brevis* during phosphate limited growth: ecological and evolutionary implications. PLOS ONE 7, e36845, http://dx.doi.org/10.1371/journal.pone.0036845.

Harrison, W.G., Harris, L.R., Irwin, B.D., 1996. The kinetics of nitrogen utilization in the oceanic mixed layer: nitrate and ammonium interactions at nannomolar concentrations. Limnol. Oceanogr. 41, 16–32.

Heil, C.A., Revilla, M., Glibert, P.M., Murasko, S., 2007. Nutrient quality drives phytoplankton community composition on the West Florida Shelf. Limnol. Oceanogr. 52, 1067–1078.

Heisler, J., Glibert, P.M., Burkholder, J.M., Anderson, D.M., Cochlan, W., Dennison, W.C., Dortch, Q., Gobler, C.J., Heil, C.A., Humphries, E., Lewitus, A., Magnien, R., Marshall, H.G., Sellner, K., Stockwell, D.A., Stoecker, D.K., Suddleson, M., 2008. Eutrophication and harmful algal blooms: a scientific consensus. Harmful Algae 8, 3–13.

Hong, J., Talapatra, S., Katz, J., Tester, P.A., Waggett, R.J., Place, A.R., 2012. Algal toxins alter copepod feeding behavior. PLoS ONE 7, e36845, http://dx.doi.org/10.1371/journal.pone.0036845.

Irigoien, X., Flynn, K.J., Harris, R.P., 2005. Phytoplankton blooms: a 'loophole' in microzooplankton grazing impact? J. Plank. Res. 27, 313–321.

Johansson, N., Graneli, E., 1999a. Cell density, chemical composition and toxicity of *Chrysochromulina polylepis* (Haptophyta) in relation to different N:P supply ratios. Mar. Biol. 135, 209–217.

Johansson, N., Graneli, E., 1999b. Influence of different nutrient conditions on cell density, chemical composition and toxicity of *Prymnesium parvum* (Haptophyta) in semi-continuous cultures. J. Exp. Mar. Biol. Ecol. 239, 243–258.

Kamykowski, D., Yamazaki, H.A., 1997. A study of metabolism-influence orientation in the diel vertical migration of marine dinoflagellates. Limnol. Oceanogr. 42, 1189–1202.

Kirkpatrick, B., Fleming, L.E., Backer, L.C., Bean, J.A., Tamer, R., Kirkpatrick, G., Kane, T., Wanner, A., Dalpra, D., Reich, A., Baden, D.G., 2006. Environmental exposures to Florida red tides: effects on emergency room respiratory diagnoses admissions. Harmful Algae 5, 526–533.

Kiørboe, T., 1993. Turbulence, phytoplankton cell size, and the structure of pelagic food webs. Adv. Mar. Biol. 29, 1–72.

Lambers, H., Chapin, F.S., Pons, T.L., 2006. Plant Physiological Ecology. Springer, New York, pp. 413–436.

Leising, A.W., Gentleman, W.C., Frost, B.W., 2003. The threshold feeding response of microzooplankton within Pacific high-nitrate low-chlorophyll ecosystem models under stady and variable iron input. Deep-Sea Res. II 50, 2877–2894.

Li, A., Stoecker, D.K., Coats, D.W., 2000. Mixotrophy in *Gyrodinium galatheanum* (Dinophyceae): grazing responses to light intensity and inorganic nutrients. J. Phycol. 36, 33–45.

Li, J., Glibert, P.M., Zhou, M., Lu, S., Lu, D., 2009. Relationships between nitrogen and phosphorus forms and ratios and the development of dinoflagellate blooms in the East China Sea. Mar. Ecol. Prog. Ser. 383, 11–26.

Litchman, E., Klausmeier, C.A., Schofield, O.M., Falkowski, P.G., 2007. The role of functional traits and trade-offs in structuring phytoplankton communities: scaling from cellular to ecosystem level. Ecol. Lett. 10, 1170–11812.

Liebeskind, B.J., Hillis, D.M., Harold, H., Zakon, H.H., 2011. Evolution of sodium channels predates the origin of nervous systems in animals. Proc. Nat. Acad. Sci. 108, 9154–9159.

Liu, H., Laws, E.A., Villareal, T.A., Buskey, E.J., 2001. Nutrient-limited growth of *Aureoumbra lagunensis* (Pelagophyceae), with implications for its capability to outgrow other phytoplankton species in phosphate-limited environments. J. Phycol. 37, 500–508.

Maestrini, S.Y., Graneli, E., 1991. Environmental conditions and ecophysiological mechanisms which led to the 1988 *Chrysochromulina polylepis* bloom: an hypothesis. Oceanol. Acta 14, 397–413.

Maldonado, M.T., Price, N.M., 1996. Influence of N substrate on Fe requirements of marine centric diatoms. Mar. Ecol. Prog. Ser. 141, 161–172.

Margalef, R., 1978. Life-forms of phytoplankton as survival alternatives in an unstable environment. Oceanol. Acta 1, 493–509.

Mitra, A., Flynn, K., 2005. Preditor-prey interactions: is 'ecological stoichiometry' sufficient when good food goes bad? J. Plank. Res. 27, 393–399.

Mitra, A., Flynn, K., 2006. Promotion of harmful algal blooms by zooplankton predatory activity. Biol. Lett. 2, 194–197.

Moore, C.M., Mills, M.M., Arrigo, K.R., Berman-Frank, I., Bopp, L., Boyd, P.W., Galbraith, E.D., Geider, R.J., Guieu, C., Jaccard, S.L., Jickells, T.D., La Roche, J., Lenton, T.M., Mahowald, N.M., Marañón, E., Marinov, I., Moore, J.K., Nakatsuka, T., Oschlies, A., Saito, M.A., Thingstad, T.F., Tsuda, A., Ulloa, O., 2013. Processes and patterns of oceanic nutrient limitation. Nat. Geosci. 6, 701–710.

Pan, Y., Rao, S., 1996. Changes in domoic acid production and cellular chemical composition of the toxigenic diatom *Pseudo-nitzschia multiseries* under phosphate limitation. J. Phycol. 32, 371–381.

Pan, Y., Bates, S., Cembella, A., 1998. Environmental stress and domoic acid production by *Pseudo-nitzschia*: a physiological perspective. Nat. Toxins 6, 127–135.

Pierce, R.W., Turner, J.T., 1992. Ecology of planktonic ciliates in marine food webs. Rev. Aquat. Sci. 6, 139–181.

Redfield, A.C., Ketchum, B.H., Richards, F.A., 1963. The influence of organisms on the composition of seawater. In: Hill, M.N. (Ed.), The Sea, vol. 2. Wiley, New York, pp. 26–77.

Ryther, J.H., Dunstan, W.M., 1971. Nitrogen, phosphorus, and eutrophication in the coastal marine environment. Science 171, 1008–1013.

Sanders, J., Dibik, S.J., D'Elia, C.F., Boynton, W.R., 1987. Nutrient enrichment studies in a coastal plain estuary: changes in phytoplankton species composition. Can. J. Fish. Aquat. Sci. 44, 83–90.

Sinclair, G.A., Kamykowski, D., 2008. Benthic-pelagic coupling in sediment-associated populations of *Karenia brevis*. J. Plank. Res. 30, 829–883.

Sipler, R.E., Bronk, D.A., Seitzinger, S.P., Lauck, R.J., McGuinness, L.R., Kirkpatrick, R.J., Heil, C.A., Kerkhof, L.J., Schofield, O.M., 2013. Trichodesmium-derived dissolved organic matter is a source of nitrogen capable of supporting the growth of toxic red tide, *Karenia brevis*. Mar. Ecol. Prog. Ser. 483, 31–45.

Smalley, G.W., Coats, D.W., Stoecker, D.K., 2003. Feeding in the mixotrophic dinoflagellate *Ceratium furca* is influenced by intracellular nutrient concentrations. Mar. Ecol. Prog. Ser. 262, 137–151.

Sterner, R.W., Elser, J.J., 2002. Ecological Stoichiometry. Princeton University Press, Princeton.

Stoecker, D., Tillmann, U., Graneli, E., 2006. Phagotrophy in harmful algae. In: Graneli, E., Turner, J.T. (Eds.), Ecology of Harmful Algae. Springer, Berlin, pp. 177–187.

Strom, S.L., 1991. Growth and grazing rates of the herbivorous dinoflagellate *Gymnodinium* sp from the open subarctic Pacific Ocean. Mar. Ecol. Prog. Ser. 78, 103–113.

Strom, S.L., Harvey, E.L., Fredrickson, K.A., Menden-Deuer, S., 2013. Broad salinity tolerance as a refuge from predation in the harmful raphidophyte alga *Heterosigma akashiwo* (Raphidophyceae). J. Phycol. 49, 20–31.

Strom, S.L., Miller, C.B., Frost, B.W., 2000. What sets the lower limit to phytoplankton stocks in high-nitrate, low-chlorophyll regions of the open ocean? Mar. Ecol. Prog. Ser. 193, 19–31.

Stumpf, R.P., Litaker, R.W., Lanerolle, L., Tester, P.A., 2008. Hydrodynamic accumulation of *Karenia* off the west coast of Florida. Cont. Shelf Res. 28, 189–213.

Sun, J., Hutchins, D.A., Feng, Y., Seubert, E.L., Caron, D.A., Fu, F., 2011. Effects of changing P_{CO2} and phosphate availability on domoic acid production and physiology of the marine harmful bloom diatom *Pseudo-nitzschia multiseries*. Limnol. Oceanogr. 56, 829–840.

Sunda, W.G., Graneli, E., Gobler, C.J., 2006. Positive feedback and the development and persistence of ecosystem disruptive algal blooms. J. Phycol. 42, 963–974.

Sunda, W.G., Hardison, D.R., 2007. Ammonium uptake and growth limitation in marine phytoplankton. Limnol. Oceanogr. 52, 2496–2506.

Sunda, W.G., Hardison, D.R., 2010. Evolutionary tradeoffs among nutrient acquisition, cell size, and grazing defense in marine phytoplankton promote ecosystem stability. Mar. Ecol. Prog. Ser. 401, 63–76.

Sunda, W.G., Huntsman, S.A., 1995. Iron uptake and growth limitation in oceanic and coastal phytoplankton. Mar. Chem. 50, 189–206.

Sunda, W.G., Shertzer, K., 2012. Modeling ecosystem disruptive algal blooms: positive feedback mechanisms. Mar. Ecol. Prog. Ser. 447, 31–47.

Sunda, W.G., Shertzer, K.W., Hardison, D.R., 2009. Ammonium uptake and growth models in marine diatoms: Monod and Droop revisited. Mar. Ecol. Prog. Ser. 386, 29–41.

Teegarden, G.J., 1999. Copepod grazing selection and particle discrimination on the bassi of PSP toxin content. Mar. Ecol. Prog. Ser. 181, 163–176.

Thingstad, T.F., Sakshaug, E., 1990. Control of phytoplankton growth in nutrient recycling ecosystems. Theory and terminology. Mar. Ecol. Prog. Ser. 63, 261–272.

Tian, R.C., 2006. Toward standard parameterizations in marine biological modeling. Ecol. Model. 193, 363–386.

Verity, P.G., 1985. Grazing, respiration, excretion, and growth rates of tintinnids. Limnol. Oceanogr. 30, 1268–1282.

Waggett, R.J., Hardison, D.R., Tester, P.A., 2012. Toxicity and nutritional inadequacy of *Karenia brevis*: synergistic mechanisms disrupt top-down grazer control. Mar. Ecol. Prog. Ser. 444, 15–30.

Watkins, S.M., Reich, A., Fleming, L.E., Hammond, R., 2008. Neurotoxic shellfish poisoning. Mar. Drugs 6, 431–455.

Wawrik, B., Callaghan, A.V., Bronk, D.A., 2009. Use of inorganic and organic nitrogen by *Synechococcus* spp. and diatoms on the west Florida shelf as measured using stable isotope probing. Appl. Environ. Microbiol. 75, 6662–6670.

Yoshida, T., Jones, L.E., Ellner, S.P., Fussmann, G.F., Hairston, N.G., 2003. Rapid evolution drives ecological dynamics in a predator-prey system. Nature 424, 303–306.