A simplified dynamic bioenergetic model for coral-*Symbiodinium* symbioses

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Abstract

This is the abstract.

Introduction

The nutritional exchange between corals and *Symbiodinium* directly underlies the capacity of corals to build coral reef ecosystems, worth trillions of US Dollars annually (Costanza, Groot, and Sutton 2014). However, the complex symbiotic metabolism of corals is vulnerable to disruption by numerous anthropogenic environmental perturbations, jeopardizing their future persistence. In order to understand and predict coral responses to complex changes in the environment, a mechanistic understanding of how multiple interacting factors drive the individual and emergent physiology of both symbiotic partners is necessary. Such a task is well suited for theoretical modeling frameworks such as Dynamic Energy Budget (DEB) theory (Kooijman 2010), although the complexity of such theory makes these efforts inaccessible to many biologists (Jager, Martin, and Zimmer 2013). In order to bridge this gap, we present here a simplified dynamic bioenergetic model for coral-*Symbiodinium* symbioses that aims to mechanistically integrate the impacts of complex environmental change on the physiological ecology of reef corals.

In reef coral symbioses, intracellular *Symbiodinium* translocate photosynthetically-fixed carbon to support coral metabolism, and utilize the animal's metabolic waste products, including nitrogenous compounds and carbon dioxide, in return (Muscatine and Porter 1977). Previous application of DEB theory to this syntrophic system (Muller et al. 2009) demonstrated a stable symbiotic relationship and qualitatively realistic growth and biomass ratios across gradients of ambient irradiance, nutrients, and food. This model assumed that 1) *Symbiodinium* has priority access to carbon through photosynthesis, 2) the coral animal has priority access to dissolved nitrogen through contact with seawater, and 3) each partner shares with the other only what it cannot use itself. In its simplest form, this principle of sharing the surplus sufficiently describes diverse syntrophic interactions among organs and organisms (e.g., trees, duckweeds, corals), suggesting the mechanism is mathematically and evolutionarily robust (Nisbet et al., submitted).

While the work of Muller et al. (2009) applying DEB formalism to this system represents the most significant theoretical contribution in coral symbiosis research to date, we aim to strengthen the role of theory and broaden its potential application to corals in three primary ways:

1. *Develop a detailed module of environmental stress.* Of primary interest to coral biologists and ecologists is symbiosis dysfunction under environmental stress, resulting in coral "bleaching"–the loss of algal symbionts from the association (Jokiel and Coles 1977). Photooxidative stress in *Symbiodinium* is considered a primary trigger of bleaching in response to high temperature and/or light (Weis 2008), and prolonged or severe bleaching can result in mortality, though corals sometimes recover their symbionts. Bleaching susceptibility, severity, and recovery may by influenced by interacting factors such as heterotrophy and

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nutrient availability (Wooldridge 2014b), and the genetic identity of *Symbiodinium* (Glynn et al. 2001). To simulate these bleaching-related phenomena, we develop a generalized framework linking overreduction of the photosynthetic light reactions to downstream impacts of photoinhibition and photodamage.

2. *Reduce theoretical and mathematical complexity.* Following the logic of Jager, Martin, and Zimmer (2013), we exclude certain features of formal DEB models in order to capture behaviors of interest with the simplest possible formulation. Here, we present a model without reserves, maturity, or reproduction (see Kooijman 2010). This formulation restricts the model's scope to the bioenergetics of growth and symbiosis dynamics in adult corals, but greatly reduces theoretical complexity and parameter numbers, which is advantageous given the relative paucity of data for corals. However, our primary motivation for reducing complexity was to increase accessibility and applicability for biologists and ecologists without requiring significant expertise in DEB theory.
3. *Provide well-documented, open-access code.* In order to facilitate the continued development and application of theoretical modeling tools for coral symbioses, we provide open access to the model in the form of detailed, commented code written in the R language (R Core Team 2014). With an accessible and modular framework, we envision this as a resource for futher development by the scientific community to include additional complexity and problem-specific components. The R language was chosen because it is freely available and in common use by biologists and ecologists, to widen the audience for this work.

With these as our primary motivations, we describe a simplified approach to dynamic bioenergetic modeling of coral-algal symbioses that tracks carbon and nitrogen acquisition and sharing between partners. This theoretical framework dynamically integrates the influence of external irradiance, nutrients, and prey availability on coral growth and symbiosis dynamics (i.e., symbiont:host biomass ratios), allowing for the possibility of coral bleaching in the event of photooxidative stress. In the following sections, we describe the formulation of this model and justify its structure and parameter values based on relevant literature. We then demonstrate the model's behavior and discuss some of its major implications and outcomes, and the wide range of potential applications for this model in the study of cnidarian-algal symbioses.

Model description

In this model of coral-algal symbiosis, carbon and nitrogen are acquired by each partner and used to construct biomass. A graphical representation of the model is presented in Fig. 1. We use C-moles as the unit of biomass for consistency with the rigorous mass balance of DEB theory: 1 C-mole is equivalent to the amount of biomass containing 1 mole of Carbon atoms. Host biomass (H), symbiont biomass (S), and prey biomass (X) have fixed, but different, molar N:C ratios (Table 1). Carbon and nitrogen are combined to produce biomass by synthesizing units (SU), which are mathematical specifications of the formation of a product from two substrates; we use the "parallel complementary" formulation of Kooijman (2010) to specify these fluxes. The two state variables in this dynamical system are symbiont biomass and coral biomass; because resources are acquired proportionally to surface area (and surface area is assumed proportional to volume for corals (i.e., they are "V1-morphs" in DEB terminology (Kooijman 2010))), biomass increases exponentially during growth. The rate of increase in coral biomass (i.e. growth) and the ratio of symbiont to host biomass are the (i.e. symbiosis dynamics) are the responses of interest of the system. Below we describe the formulation of each flux involved in producing these responses.

Table 1

Symbol	Description	Value	Units
n_{NH}	N:C molar ratio in host biomass	0.19	–
n_{NS}	N:C molar ratio in symbiont biomass	0.2	–
n_{NX}	N:C molar ratio in prey biomass	0.13	–
j^0_{HT}	Specific turnover rate of host biomass	0.03	d^{-1}
j^0_{ST}	Specific turnover rate of symbiont biomass	0.03	d^{-1}
σ_{NH}	Proportion host nitrogen turnover recycled	0.9	–
σ_{CH}	Proportion host carbon turnover recycled	0.9	–

Symbol	Description	Value	Units
σ_{NS}	Proportion symbiont nitrogen turnover recycled	0.9	–
σ_{CS}	Proportion symbiont carbon turnover recycled	0.9	–
j_{Xm}	Maximum specific feeding rate of host	0.1292	$molX \cdot CmolH^{-1} \cdot d^{-1}$
K_X	Half-saturation constant for prey uptake by host	20e-6	$molX \cdot L^{-1}$
j_{Nm}	Maximum specific DIN uptake rate by host	0.048	$molN \cdot CmolH^{-1} \cdot d^{-1}$
K_N	Half-saturation constant for DIN uptake by host	0.46e-6	$molN \cdot L^{-1}$
$j^p_{CO_2}$	Passive CO_2 delivery to symbiont	4.04e-3	$molC \cdot CmolH^{-1} \cdot d^{-1}$
$j^a_{CO_2}$	Active CO_2 delivery to symbiont	0.32–18.0	$molC \cdot CmolH^{-1} \cdot d^{-1}$
j_{HGm}	Maximum specific growth rate of host	1	d^{-1}
n_{LC}	Quantum yield of photosynthesis	0.1	$molC \cdot molph^{-1}$
$\bar{a}^*$	Effective light-absorbing cross-section of symbiont	1.34	$m^2 \cdot CmolS^{-1}$
j_{NPQ}	Non-photochemical quenching capacity of symbiont	40	$molph \cdot CmolS^{-1} \cdot d^{-1}$
k_{ROS}	Excess photon energy that doubles ROS prod.	40-80	$molph \cdot CmolS^{-1} \cdot d^{-1}$
k	Exponent on ROS production rate	1	–
j_{CPm}	Maximum specific photosynthesis rate of symbiont	2.8	$molC \cdot CmolS^{-1} \cdot d^{-1}$
j_{SGm}	Maximum specific growth rate of symbiont	0.25	d^{-1}

Also include tables of fluxes and environmental inputs?

Coral animal fluxes

The coral animal acquires both carbon and nitrogen from feeding on prey from the environment. Prey acquisition is specified by Michaelis-Menten kinetics using a maximum area-specific feeding rate and half-saturation constant:

$$j_X = \frac{j_{Xm} \cdot X}{X + K_X} \tag{1}$$

Additionally, the coral animal can acquire nitrogen from the surrounding seawater. This nitrogen source is assumed to represent ammonium, the primary form utilized by corals (Wang and Douglas 1998; Yellowlees, Rees, and Leggat 2008). The uptake of nitrogen from the environment is specified by Michaelis-Menten kinetics using a maximum area-specific uptake rate and half-saturation constant:

$$j_N = \frac{j_{Nm} \cdot N}{N + K_N} \tag{2}$$

Coral biomass formation is specified by a parallel complementary SU that combines carbon and nitrogen to form biomass. In addition to the carbon and nitrogen acquired by direct uptake (Eqs. 1 and 2), the coral recycles a portion of the nitrogen liberated by biomass turnover ($r_{NH} = \sigma_{NH} j^0_{HT}$), and receives surplus fixed carbon shared by the symbiont (ρ_C), such that the total biomass formation is specified as:

$$j_{HG} = \left(\frac{1}{j_{HGm}} + \frac{1}{\rho_C \frac{S}{H} + j_X} + \frac{1}{(j_N + n_{NX} j_X + r_{NH})/n_{NH}} - \frac{1}{\rho_C \frac{S}{H} + j_X + (j_N + n_{NX} j_X + r_{NH})/n_{NH}} \right)^{-1} \tag{3}$$

The amount of nitrogen input to the coral biomass SU in excess of what is actually consumed in biomass formation (i.e., surplus nitrogen, or the "rejection flux" in SU terminology) is then made available to the symbiont, and is specified as:

$$\rho_N = (j_N + n_{NX} j_X + r_{NH} - n_{NH} j_{HG})_+ \tag{4}$$

Due to the inherent inefficiency of the parallel complementary SU formulation, there will always be some nitrogen shared with the symbiont even when coral biomass formation is strongly nitrogen-limited. Likewise, there is always a non-zero rejection flux of carbon from the coral biomass SU, which is assumed to be lost to the environment.

Symbiodinium *fluxes*

The symbiont produces fixed carbon through photosynthesis, a process represented here by a single SU with two substrates: light (photons) and inorganic carbon (CO_2). The amount of light absorbed by the symbiont depends on the scalar irradiance at the site of light absorption, which is modified substantially relative to external downwelling irradiance owing to multiple scattering by the coral skeleton and self-shading by surrounding symbionts (Enríquez, Méndez, and Iglesias-Prieto 2005; Marcelino et al. 2013). We used data from Marcelino et al. (2013) to empirically derive the ratio of internal scalar irradiance to external downwelling irradiance as a function of symbiont density (expressed as symbiont to host biomass ratio), and subsequently multiply this quantity by the external downwelling irradiance L and the effective light-absorbing surface area of symbiont biomass ds to specify the amount of light absorbed:

$$j_L = \left[1.26 + 1.39 \cdot \exp(-6.48 \cdot \frac{S}{H})\right] \cdot L \cdot \bar{a}^* \tag{5}$$

We then specify two pathways for input of inorganic carbon to the photosynthesis SU: 1) passive diffusion of CO_2 from the external environment, and 2) active delivery of CO_2 to the symbiont by the host. The passive flux ensures that some CO_2 is always available to photosynthesis, and the active flux encompasses the potentially diverse mechanisms by which the host may enhance CO_2 availability for the symbiont, including active transport of bicarbonate, carbonic anhydrase-catalyzed conversion of bicarbonate to CO_2 to promote diffusion toward the symbiont, and acidification of the symbiosome to increase localized CO_2 concentrations. Since the host physically separates the symbiont from the external environment, both the passive and active flux rates are proportional to host surface area. A more rigorous, mechanistic model of inorganic carbon processing would require spatially explicit internal pools accounting for pH and carbon speciation, which is beyond the current scope of this work. Instead, the specification of CO_2 delivery rates offers the user the opportunity to compare different rates of CO_2 delivery that may characterize different coral species (Wooldridge 2014a). The input of CO_2 to the photosynthesis SU is therefore specified as:

$$j_{CO_2} = j^p_{CO_2} + j^a_{CO_2} \tag{6}$$

In addition to the inputs CO_2 specified in Eqs. 7, additional CO_2 representing the metabolic production of CO_2 from host and symbiont biomass turnover ($r_{CH} = \sigma_{CH} j^0_{HT}; r_{CS} = \sigma_{CS} j^0_{ST}$) is made available to the photosynthesis SU. fixed carbon is produced by the photosynthesis SU according to:

$$j_{CP} = \left(\frac{1}{j_{CPm}} + \frac{1}{n_{LC} j_L} + \frac{1}{(j_{CO_2} + r_{CH})\frac{H}{S} + r_{CS}} - \frac{1}{n_{LC} j_L + (j_{CO_2} + r_{CH})\frac{H}{S} + r_{CS}}\right)^{-1} \cdot c^{-1}_{ROS} \tag{7}$$

where j_{CPm} is the maximum specific rate of photosynthesis, and c_{ROS} is the photooxidative stress multiplier (see below). Dividing by c_{ROS} causes the rate of photosynthesis to decline in response to photo-oxidative stress, a phenomenon known as photoinhibition.

Light energy absorbed in excess of what is used to fix carbon is specified by the SU "rejection flux", according to:

$$j_{eL} = (j_L - j_{CP}/n_{LC})_+ \tag{8}$$

This excess light energy must be quenched by alternative pathways in order to prevent photo-oxidative damage. *Symbiodinium* may utilize a variety of pathways for non-photochemical quenching (NPQ; Roth 2014), which we collect in a total capacity for NPQ as a parameter of the symbiont (j_{NPQ}). If light energy exceeds the total capacity of both carbon fixation and NPQ, then damaging reactive oxygen species (ROS) may be produced. We represent this as a scaled flux of ROS c_{ROS}, which takes a value of 1 when all light absorbed is quenched by photochemistry and NPQ, and increases as the amount of excess excitation energy increases.

$$c_{ROS} = 1 + \left[\left(\frac{j_{eL} - j_{NPQ}}{k_{ROS}}\right)^k\right]_+ \tag{9}$$

where j_{NPQ}, k_{ROS}, and k are parameters of the symbiont that determine the onset and rate of ROS production. Importantly, c_{ROS} as specified here is not a function of absolute light absorption, but rather the amount of excess light energy j_{eL} after accounting for carbon fixation and NPQ. A direct consequence of this formulation is that carbon-limitation of photosynthesis can lead to photo-oxidative stress, a mechanism of biological importance (Wooldridge 2009) that was not captured by previous representations of photo-oxidative stress (Eynaud, Nisbet, and Muller 2011). Moreover, this formulation allows functional diversity among symbiont types to be explored by changing the parameters j_{NPQ}, k_{ROS}, and k.

Carbon fixed by photosynthesis (j_{CP}) is then used in conjunction with nitrogen shared by the host (ρ_N) and a proportion of nitrogen recycled from symbiont biomass turnover ($r_{NS} = \sigma_{NS} j^0_{ST}$) to build new symbiont biomass, following the SU equation:

$$j_{SG} = \left(\frac{1}{j_{SGm}} + \frac{1}{j_{CP}} + \frac{1}{(\rho_N \frac{H}{S} + r_{NS})/n_{NH}} - \frac{1}{j_{CP} + (\rho_N \frac{H}{S} + r_{NS})/n_{NH}} \right)^{-1} \tag{10}$$

The rejection flux of carbon from this SU represents the amount of fixed carbon produced by photosynthesis in excess of what can be used to produce symbiont biomass; this surplus ρ_C is translocated to the coral host:

$$\rho_C = (j_{CP} - j_{SG})_+ \tag{11}$$

The rejection flux of nitrogen from the symbiont biomass SU is lost to the environment.

Symbiont biomass turnover includes a component of constant turnover specified by the parameter j^0_{ST}, representing fixed maintenance costs, plus a component that scales with the magnitude of ROS production.

$$j_{ST} = j^0_{ST}(1 + 5 \cdot (c_{ROS} - 1)) \tag{12}$$

This second component of symbiont biomass loss can represent both photodamage and/or symbiont expulsion (i.e., bleaching), both of which happen in response to higher levels or ROS production. The constant 5 is included to increase biomass loss in response to ROS. (Note that recycling of symbiont biomass turnover (rNS and rCS) only occurs based on the basal maintenance related turnover (i.e., j^0_{ST}), and not the bleaching-related biomass loss, as this loss represents biomass being damaged or being expelled from the holobiont).

Model state equations

Finally, the balance equations representing the specific growth rates of symbiont and host biomass over time can be expressed as:

$$\frac{dS}{Sdt} = j_{SG} - j_{ST} \tag{13}$$

$$\frac{dH}{Hdt} = j_{HG} - j^0_{HT} \tag{14}$$

Numerical analysis

A time-stepping Euler method was used to solve the state equations since the production and rejection fluxes of the SUs are imiplicitly defined. Specifically, the rejection fluxes of carbon and nitrogen from the symbiont and host biomass SUs act as reciprocal input fluxes to the other SU. In addition, the rejection flux of excitation energy from the photosynthesis SU acts to reduce its own production flux (i.e., photoinhibition), and hence a discretized time-stepping procedure was necessary. A vector of time values was created for each simulation run, along which dynamic environmental forcing functions (irradiance, DIN, and prey abundance) can be designed. These vectors, along with initial values of symbiont and host biomass, then serve as input to the time-stepping function, which solves for the current system state using values of the previous system state where necessary. A default time step of 0.1 days was used for all simulations, which were performed using R code that is available in the data repository accompanying this article: github.com/jrcunning/Rcoral.

Model behavior evaluation

To analyze qualitative behavior, we ran the model to steady state across gradients of external irradiance and DIN, and plotted the specific growth rate (Fig. 2A) and symbiont to host biomass ratio (Fig. 2B). Results are consistent with observations in corals: specific growth rates

Discussion - Potential applications/utility

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List of Figures

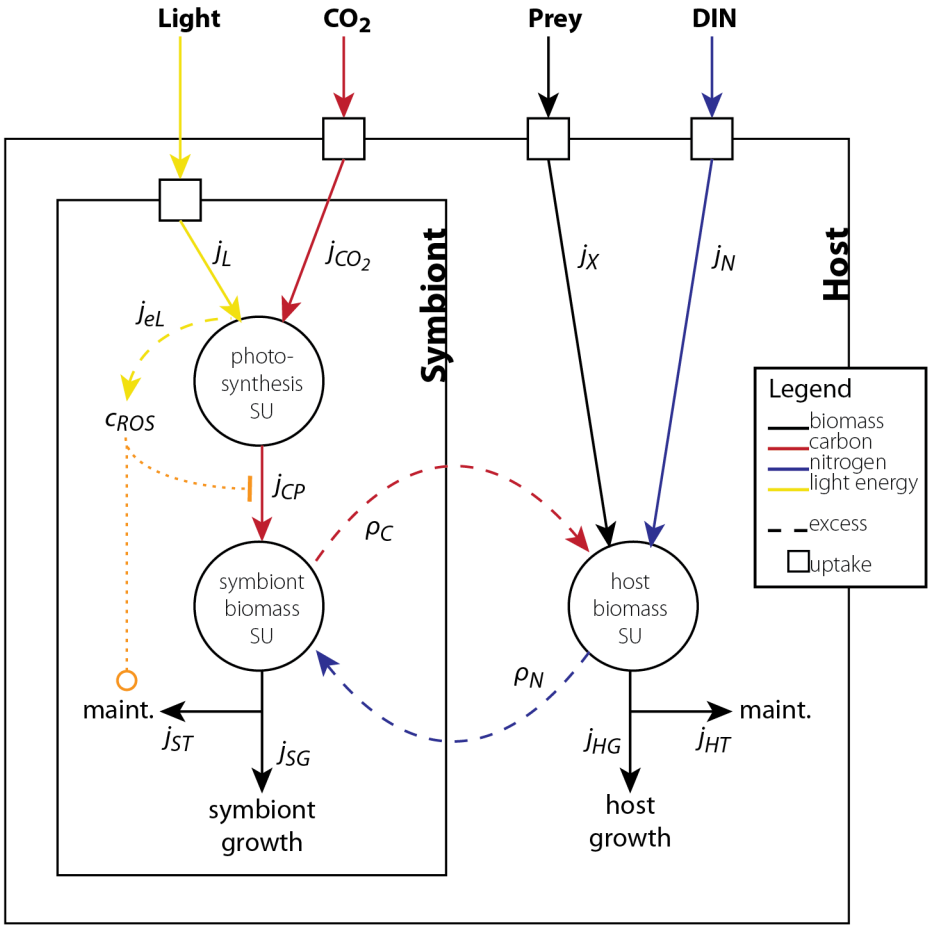

Figure 1: Graphical representation of coral-algal symbiosis model.

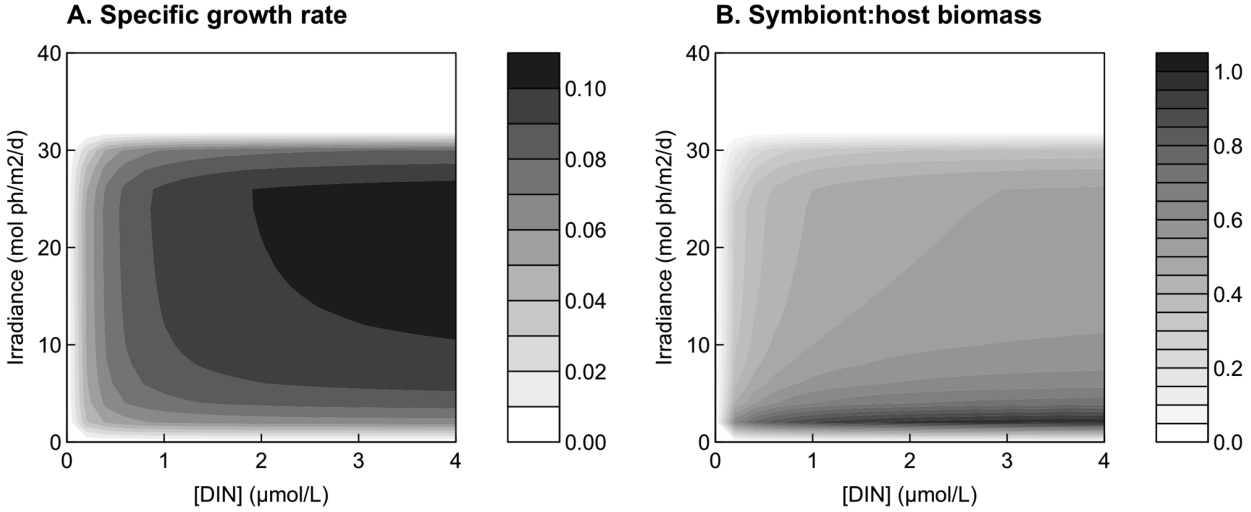

Figure 2: Steady state values of (A) specific growth ($Cmol \cdot Cmol^{-1} \cdot d^{-1}$) and (B) the symbiont to host biomass ratio ($CmolS \cdot CmolH^{-1}$) across gradients of external irradiance and dissolved inorganic nitrogen. Simulations for each combination of light and nutrients (21 points along each axis) were run for 100 days with a time step of 1 day. Negative steady state growth rates, and corresponding S:H ratios, were set to zero.