Fishers' Perceptions of Ciguatoxin Fish Poisoning and Modeling Biomagnification of Ciguatoxin in the Trophic Dynamics of Caribbean Coral Reef Ecosystems

by

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Ciguatoxin fish poisoning (CFP) is caused by the consumption of tropical and subtropical fishes and other marine species with high levels of ciguatoxin (CTX) in their tissues. CTX is a polycyclic neurotoxin produced by single-celled, photosynthetic dinoflagellates in the *Gambierdiscus* and *Fukuyoa* genera which are found in close association with benthic autotrophs. CTX enters the food web when these dinoflagellates are inadvertently consumed by herbivores grazing on their preferred substrates. The toxin biomagnifies up the food chain to the top predators and if humans consume seafood with high levels of CTX it can cause a variety of harsh symptoms. The best way to avoid CFP is to avoid toxic fishes. However, CTX is undetectable by physical inspection. To help prevent CFP cases, I, along with my colleagues, created two predictive models in Ecopath with Ecosim with data we collected and data from the literature, for areas in Puerto Rico, identified as CTX hotspots and coldspots by fishers. I confirmed the fishers' information by sampling fishes and estimating CTX levels in their tissues using a bioassay. We also collected toxic dinoflagellates and show here that the hotspot had much higher densities of these cells than the coldspot. We recommend managers in Puerto Rico establish a routine monitoring program for the toxic dinoflagellates to be used in conjunction with the models presented here to help predict when certain fishes may be toxic along specific reefs. This would mean a safer, more efficient, and more robust fishing economy in Puerto Rico.

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DEDICATION

For Lucy, whose dedicated companionship and loving support I could not repay even with a lifetime of ear scratches and dog bones.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

CFP	Ciguatoxin Fish Poisoning
CTX	Ciguatoxin
EE	Ecotrophic Efficiency
EwE	Ecopath with Ecosim
FDA	United States Food and Drug Administration
IK	Indigenous Knowledge
LEK	Local Ecological Knowledge
Pbtx	Brevetoxins
ppb	Parts Per Billion
PR	Puerto Rico
qPCR	Quantitative Polymerase Chain Reaction
STT	St. Thomas, USVI
STX	Saxitoxins
TEK	Traditional Ecological Knowledge
TTX	Tetrodotoxins
vBGF	von Bertalanffy Growth Function
ww	Wet Weight

Chapter 1. INTRODUCTION

Purpose of Study

Ciguatera fish poisoning (CFP) is a human health concern caused by consuming seafood with high levels of ciguatera toxin (CTX). CFP is endemic to tropical and subtropical regions globally. However, rising sea temperatures and the demand for imported seafood from these regions means cases are spreading beyond the tropics (Mattei *et al.*, 2014; Rodríguez *et al.*, 2017; Friedemann, 2019; de Haro *et al.*, 2020). CTX enters fishes and other marine species when it biomagnifies up the food chain, starting with its production in single-celled dinoflagellates in the *Gambierdiscus* and *Fukuyoa* genera. Herbivores consume these dinoflagellates when feeding on their preferred substrates, which introduces the toxin to the food web. Subsequently, predators consume these herbivores and acquire the toxin. Humans then target these predators and herbivores for consumption, where if consumed in high enough levels, causes severe illness. CTX is odorless, tasteless, and is heat-stable; it is impossible to identify in tissues by physical inspection, and there are no rapid tests available. The best way to determine a marine organism's toxicity is to run complicated and lengthy bioassays that are not feasible to fishers, consumers, or the general public. There are no good predictive models or routine monitoring sampling protocols to attempt to prevent CFP outbreaks. Therefore, in this project, we developed predictive models that fisheries managers in Puerto Rico can use to identify when fishes may be toxic at certain coral reefs. Using interviews with fishers we[1] identified one CTX hotspot (an area with high levels of CTX), and one CTX coldspot (low levels of CTX) to sample. Then, we[2] parameterized two food web models, one for the hotspot and one for the coldspot. I

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[1] The interview team in Puerto Rico was Dr. Miguel Del Pozo, Dr. Joseph Luczkovich, and Henry Raab

[2] The model parameterization and validation team was Dr. Joseph Luczkovich, Dr. Stuart Borrett, and Henry Raab

believe that, along with a routine monitoring sampling program for these toxin-producing dinoflagellates, these models can estimate the length of time it takes CTX to reach FDA recommended consumption advisory levels in marine species commonly caught in Puerto Rico. Fisheries managers could use these data to guide fishing habits during riskier CTX months and help prevent widespread CFP events.

This dissertation was a funded Puerto Rico SeaGrant project intended to investigate the socio-ecological role of CTX in Puerto Rico fisheries. The goals were to: 1) identify one hotspot (high levels of CTX) and one coldspot (low levels of CTX) site off the coast of Puerto Rico identified by commercial fishers from a series of personal interviews; 2) sample fishes at those sites and test them for CTX concentrations to confirm the information provided by fishers; 3) sample ciguatoxin-producing dinoflagellates to identify quantity and species at each site; 4) use fish data collected at each site along with biomass estimates, and previously published diet, production, and consumption data from the literature to parameterize a food web model for each the hotspot and coldspot; and 5) simulate the movement of CTX through the food web networks to estimate the time it takes fishes of varying trophic levels to acquire >0.1 ppb in their tissues, the FDA identified consumer advisory level for consumption for Caribbean chemical strains of CTX. We will provide these simulation models, the output results, and our recommendations for the use of these models to the Puerto Rico fisheries managers in the Departmento de Recursos Naturales y Ambientales (Department of Natural and Environmental Resources, DRNA) and the Caribbean Fisheries Management Council (CFMC). Fisheries managers can use these models as an additional resource for identifying and predicting future CTX outbreaks to potentially limit ciguatoxic fish poisoning (CFP) events from Puerto Rico fishes. It could also increase the efficiency of Puerto Rico fisheries by allowing the catch of more fishes with minimal risk of having high levels of CTX. We propose a routine monitoring program to sample toxic dinoflagellates along the reefs in Puerto Rico. Combined with the simulation models developed in this project, these data could help managers make more informed decisions about potentially toxic or safe fishes to eat and how long after a dinoflagellate bloom that marine species would reach the FDA recommended consumption advisory level of Caribbean CTX's of 0.1 ppb.

I would like to clarify the terminology I will use to refer to ciguatoxin in fish tissues at the outset of this dissertation. I used the mouse neuroblastoma cell-culture bioassay (N2a-cba) to determine if ciguatoxin was present in fish tissues(Pawlowiez *et al.*, 2013; Reverté *et al.*, 2014; Litaker *et al.*, 2017). Cells in the bioassay will die if ciguatoxin is present; according to Pawlowiez *et al*. (2013), Reverté *et al.*(2014), and Litaker *et al.* (2017), the more cell death present in a fish tissue sample, the greater the concentration of ciguatoxin (referenced to the Pacific ciguatoxin, P-CTX-3C). Thus, when I mention CTX in the scope of the laboratory testing with the N2a neuroblastoma cell-based assay, I will express it in P-CTX-3C equivalents, or CTX3C equiv. or CTX-3C equiv. However, when I mention ciguatoxin generally, I will use "CTX" because there are no readily available Caribbean ciguatoxin standards (for C-CTX-1C) to use for liquid chromatography/mass spectrometry (LC/MS); without the standards it is impossible to confirm that ciguatoxin is present in my samples when using the N2a-cba method alone. However, because the N2a-cba test is sensitive to Pacific P-CTX-3C, I assumed that a similar sensitivity exists for the Caribbean C-CTX-1C; this is a common assumption used by investigators of Caribbean CTX and well known in the ciguatoxin literature (Hardison *et al.*, 2016; Suzuki *et al.*, 2017; Estevez *et al.*, 2019). The C-CTX-1C found in Caribbean fish tissues is a chemical congener of Pacific ciguatoxin P-CTX-3C, has similar binding kinetics to P-CTX-3C, and can be converted to P-CTX-3C equiv. for Caribbean studies with a conversion constant of 1.3 (Hardison *et al.*, 2016). The Pacific ciguatoxin P-CTX-3C is readily available in a concentrated form for use in standard curves (Fujifilm Wako Chemicals). Therefore, I can compare toxin concentration profiles from the N2a-cba for C-CTX-1C in my samples to P-CTX-3C equiv. and estimate the Caribbean strains' toxicity using standard toxicity curves (Litaker *et al.*, 2017). Thus, I will report C-CTX-1C levels of toxicity in Caribbean fishes in P-CTX-3C equivalents. For further explanation of the methods that I used to assay for CTX, see the section below and Methods in Chapter 4.

Hypotheses

By collecting TEK from local fishers (Johnson and Griffith, 1996, 2010; Bernard, 2011) regarding ciguatoxin to identify locations with high and low levels of CTX in fish, invertebrates, and algae, sampling those species to parameterize two Ecopath with Ecosim (EwE) food web models, and running simulations using the Ecotracer module within EwE, I wanted to identify the length of time it would take ciguatoxin to biomagnify in coral reef food web networks from the low trophic levels (dinoflagellates to herbivores) to the higher trophic level predators (great barracuda, hogfish, etc.). The three three main hypotheses tested are:

H1: Fish through from TEK-identified hotspot will have higher concentrations of CTX equivalents in their tissue samples than fish collected in coldspots.

H2: Higher dinoflagellate cell counts (of the same species and strains) at the hotspot area will cause the top trophic levels to reach 0.1ppb faster than lower dinoflagellate cell counts with the same toxin concentration.

H3: The levels of CTX3C equivalents in top trophic predators will be higher than the levels of CTX3C equivalents in herbivores and lower trophic levels with the same initial composition (cell quantity and toxin concentration) of dinoflagellate species or strains.

I tested the first hypothesis using the neuroblastoma cell-based assay to estimate CTX concentrations in fishes and the second and third hypotheses using model simulations in EwE.

Ciguatoxin Fish Poisoning: A Global Health Issue

People living in tropical and subtropical regions worldwide rely on fish and other marine organisms for sustenance, tourism, and recreation. However, fishes in these regions, specifically in the Pacific and Indian Oceans and the Caribbean Sea, can harbor ciguatera toxin (ciguatoxin or CTX), a potent neurotoxin produced by several different species of dinoflagellates, most notably in the *Gambierdiscus* and *Fukuyoa* genera (Lewis *et al.*, 1991; Pottier *et al.*, 2002). If humans ingest tissues of marine coral reef species that accumulate this toxin in a high concentration then it can cause a variety of severe symptoms, i.e., vomiting, diarrhea, abdominal pain, paresthesia (burning of the skin), the reversal of hot and cold sensations, and occasionally, death (Lehane and Lewis, 2000). The muscle tissues (the fish filets most people consume) have the potential to be toxic. Also, the roe, gonads, liver, and other organs in the fishes carry higher levels of CTX than muscle tissues, and these organs may be more dangerous to consume than muscles (de Fouw *et al.*, 2001). Different structures and chemical congeners of ciguatoxins in the Indian Ocean, Pacific Ocean, and the Caribbean Sea cause variations in symptoms from those regions (Murata *et al.*, 1990; Lewis, 1998, 2000). The sickness from consuming ciguatoxic fish is known as ciguatoxin fish poisoning (CFP).

CTX-producing dinoflagellates are endemic mostly to Caribbean and Pacific island reefs (i.e., the Caribbean; the U.S. Virgin Islands, Antigua and Barbuda, the Cayman Islands, Puerto Rico, Jamaica, etc. and in the Pacific; American Samoa, the Cook Islands, Fiji, Hawaii, French Polynesia, etc.) (Lewis, 2001; Litaker *et al.*, 2010; Friedman *et al.*, 2017). Pacific ciguatoxins (P-CTX) are 10-fold more toxic than Caribbean ciguatoxins (C-CTX), therefore, the FDA recommended advisory consumption levels for the primary Caribbean CTX chemical strain is less than 0.1 ppb C-CTX-1 equiv. toxicity and 0.01 ppb P-CTX-1 equiv. toxicity for Pacific ciguatoxins (Vernoux and Lewis, 1997; Lewis *et al.*, 1999; Lehane and Lewis, 2000; Pearn, 2001; Dickey and Plakas, 2010).

Despite the impact CTX has on fisheries and consumers, it is challenging to pinpoint global CFP incidence rates with a high confidence level due to poor CTX detection techniques (Friedman *et al.*, 2017). CTX is colorless, odorless, and tasteless (Copeland *et al.*, 2014) and is heat-stable, meaning cooking the fish does not affect the toxin (Lewis, 2000). Local folk methods for identifying toxic fish (such as feeding a small piece of fish to a pet animal and monitoring its reaction, rubbing the flesh with a coin, or leaving a portion of the fish near insects to see if they avoid it) are unreliable (Darius *et al.*, 2013). Also, Oceanit® discontinued CTX dockside test strips (Cigua-Check®) due to the tests' inaccuracy. They were confusing to administer and inaccurate, with a high chance of false-negatives (Bienfang *et al*., 2011). Some of the other reasons that the number of global CFP cases are hard to estimate are misdiagnosis (some symptoms are similar to the flu or other gastrointestinal issues) (Swift and Swift, 1993; Ruff and Lewis, 1994); the prevalence in underdeveloped tropical areas with poor access to medical treatment (Banner, 1976), underdiagnosis due to a lack of knowledge by doctors of ciguatera fish poisoning (only 47% of Florida doctors knew that ciguatera was a reportable condition) (McKee *et al.*, 2000); the avoidance of hospitals when people become sick (less than 0.1% of intoxicated people visit a physician for a consultation (Tosteson, 1995); and, the lack of testing of the suspected fish meal for CTX that is needed to confirm the diagnosis of CFP after sickness occurs (CDC, 2009). Yearly estimates can range anywhere from 25,000 globally (Lewis and Sellin 1992; Lewis 2001) to 20,000 to 40,000 in Puerto Rico and the U.S. Virgin Islands alone (Tosteson, 1995). On the small island of Culebra in Puerto Rico, the incidence rates have been estimated as high as 75 cases/10,000 people (Azziz-Baumgartner *et al.*, 2012). More recently, an analysis of data from the United States National Poison Center on CFP (including Guam, Puerto Rico, and the U.S. Virgin Islands) done by the National Institutes of Health showed a yearly call rate incidence rate (calls per year to poison control) to report CFP between 2001 and 2011 of 0.003 cases/10,000 residents (Gingold *et al.*, 2014). To compare, in some of the more toxic areas like Raivavae (Australes) in French Polynesia, incidence rates are estimated to be 140 cases/10,000 people (Chinain *et al.*, 2010b), 250 cases/ 10,000 people in Southern Kiribati (Skinner *et al.*, 2011), and up to 1,436 cases/10,000 people in the Cook Islands (Skinner *et al.*, 2011). Although incidence rates are relatively low in the United States, CFP has been estimated to cause economic losses between $15 to $22 million annually due to increased hospitalizations, lost work, and lost fisheries resources (Anderson *et al.*, 2000; Hoagland *et al.*, 2002). These numbers likely significantly underestimate actual losses due to under-reporting. There are no proven treatments for CFP besides fluids and rest although some anecdotal evidence shows that a single-dose of mannitol can help subdue symptoms. A randomized double-blind showed that normal saline had a similar effectiveness for treating CFP with more symptoms and did not support single-dose mannitol for treatment for CFP (Schnorf *et al.*, 2002).

Biomagnification in Food Webs

It is essential to distinguish among biosynthesis, depuration, bioaccumulation, bioconcentration, and biomagnification of toxins in animals and food webs (following the terminology described by Bienfang *et al.* (2013). Biosynthesis occurs when an organism produces a toxin internally within its cells. Alternatively, animals may absorb toxins from their abiotic environment or their biotic environment (their prey or food) and store it in their tissues. They may also metabolize absorbed or consumed toxins and excrete them; this is depuration. Bioaccumulation is the increase in the concentration of a toxin from the abiotic environment within one trophic level. Bioconcentration is the increase in the concentration of a toxin from the abiotic and biotic environment within one trophic level. The storage of a toxin in a predator's tissues is usually due to the retention of lipophilic toxins in fatty tissues. When predators consume prey in a food chain, biomagnification increases toxin in these predators; greater toxin levels occur at higher trophic levels.

Ciguatoxins are polyether neurotoxins produced by epibenthic dinoflagellate microalgae in the genera *Gambierdiscus* and *Fukuyoa;* this first step is referred to as the biosynthesis of CTX at the base of the food web. These dinoflagellates are ubiquitously distributed in shallow, tropical, hard bottom communities throughout the Atlantic and Pacific, with varying cell densities (Pottier *et al.*, 2002; Litaker *et al.*, 2010). They occupy reef ecosystems and are closely associated with macroalgae, algal turfs, seagrasses, coral rubble, and other similar substrates (Parsons and Preskitt, 2007; Rains and Parsons, 2015; Pisapia *et al.,* 2017). In regions where cell concentrations remain between 100-1,000 cells g^{-1} wet weight of macroalgae, people consuming fish do not experience significant CFP issues, however, when dinoflagellate blooms exceed 1,000 cells g^{-1} wet weight macroalgae, the probability of a CFP event increases (Litaker *et al.*, 2010). Data from the Pacific indicates that although a significant rise in dinoflagellate density is required to cause a CFP event, some blooms are dominated by low-toxicity species that otherwise pose a little risk even at densities > 1,000 cells g^{-1} wet weight of macroalgae (Chinain *et al*. 2010a, 2010b). Thus, dinoflagellate species identification is essential to assess when a CFP event occurs.

The currently accepted theory that explains how the biosynthesized CTX enters the food web is called the “ciguatoxin food chain model” (Randall, 1958). This theory states that various herbivores, mainly herbivorous fishes, feed on the macroalgal substrates harboring dinoflagellates (Lewis, 2001; Ledreux *et al.*, 2014). The herbivorous fish consume these ciguatoxins along with macroalgae, then metabolize and partially excrete the toxins (depuration), while some toxin remains in the tissues (bioaccumulation; See Mechanism of Action and Biometabolism section). This bioaccumulation of toxins in tissues of the herbivorous fishes occurs because CTX is a lipophilic compound. Large carnivores consume the smaller herbivorous fish, which leads to the toxin biomagnifying in the food web; it increases in concentration at the higher trophic levels. The consumption of fishes with accumulated toxin could account for the highest toxin concentrations in top trophic-level species like great barracuda (*Sphyraena barracuda*), various species of jacks (Carangidae), and groupers (Serranidae). However, this bioaccumulation and biomagnification process may not be efficient, as significant depuration or a reduction in the toxin's assimilation may occur at the initial herbivore consumption stage (Ledreux *et al*., 2014). In summarization, Randall’s food chain theory postulates that top trophic-level species, such as a great barracuda, are dependent upon food originating from primary producers spread over a wide area of a reef; top predators also live a long time and thus indirectly consume the production from biosynthesizing producers and bioaccumulating herbivores spread over a wide area and a long time. This theory appears to be occurring on coral reefs where *Gambierdicsus* occurs: biosynthesis of CTX by *Gambierdiscus*, bioaccumulation by herbivorous fishes and intermediate consumers in fatty tissues, and biomagnification of CTX at higher trophic levels.

Effect of CTX on Fish Behavior

Fishes experience hyper- and hypoactive behaviors after consuming ciguatoxin-producing dinoflagellates (Ledreux *et al.*, 2014). Researchers investigated these behaviors and reported that fish consuming prey with CTX displayed some hypoactive actions that included; relaxation of the jaw, resting at the bottom of the tank, fin paralysis, and loss of equilibrium (Ledreux *et al.*, 2014). Some fishes had hyperactive behaviors like erratic swimming, jerky feeding habits, and convulsions (Ledreux *et al.*, 2014). These behaviors could increase the fish's vulnerability, which would make them more susceptible to predation. A fish’s inability to flee from predators could either reduce the amount of time to accumulate CTX in their tissues or increase the amount of toxin in its predators’ tissues due to consuming more toxic fishes.

Coral Reef Ecosystem Changes, Dinoflagellates and CTX

Reef composition could be an essential factor in the number of toxin-producing dinoflagellates on reefs. A coral-algal phase shift is when a reef in a primarily coral state transition to an algal state due to a variety of factors like increased nutrient runoff, coral diseases, pollution from sewage, sedimentation, as well as increasing global water temperatures and pH changes (Alcolado, 1990; Hughes, 1994; Linton *et al.*, 2002). Algal cover in a reef that exceeds 75% for a prolonged time constitutes a full coral-algal shift (Hughes, 1994). This shift may result in a preferable habitat for CTX-producing dinoflagellates, causing an increase in CFP-related incidents (Morrison *et al.*, 2008). A study done in Cuba showed that communities with higher rates of CFP had local reefs that were heavily dominated by algae compared to communities with lower rates of CFP that had a below-average algal cover (Morrison *et al.*, 2008). The transition to algal reefs may lead to higher cell densities of the toxin-producing dinoflagellate stains and, subsequently, a more toxic reef.

Mechanism of Action and Biometabolism

Pacific strains of ciguatoxins (P-CTX) are far more toxic than the Caribbean strains (C-CTX), which are less polar (Lehane and Lewis, 2000). Both congeners are heat-stable and lipid-soluble (Lewis, 2000). They are voltage-gated sodium channel selective toxins, and the congeners all have a similar structure, with cyclic ether rings and differences in oxidation at the ends of the molecule (Dechraoui *et al.*, 2011). They work by binding to site 5 of the voltage-gated sodium ion channel and stimulate repetitive Na^+ influx, which causes the repetitive firing of neurons (Lombet *et al.*, 1987; Caillaud *et al.*, 2012). Voltage-gated sodium ion channels are transmembrane structures responsible for action potentials and, therefore, electronic transmission along a neuron (excitable cell and the main component of nervous tissue) in the body (Hodgkin and Huxley, 1952). This repetitive firing of neurons drives neurological symptoms in people with CFP.

Each species or strain of CTX-producing dinoflagellate creates different precursors to toxic chemical congeners. The types of chemical congeners found in fishes share a similar cyclic ether backbone with varying oxygenation degrees at the ends (Bottein *et al.*, 2011). For example, *Gambierdiscus toxicus* produces over 20 precursor chemical congeners. Fishes livers metabolize chemical congeners, altering the toxins' chemical structure as they bioaccumulate and biomagnify in the food chain (Lehane and Lewis, 2000). Ledreux *et al*. (2014) did a study that looked at the trophic transfer dynamics of CTX from *Gambierdiscus polynesiensis* cells (a

Pacific species producing P-CTX) to the second trophic level (an Atlantic herbivore, *Mugil cephalus,* striped mullet). The sampled striped mullet blood after being fed a known concentration of CTX in a gel pellet meal. Once ingested by the mullet, ciguatoxin was rapidly absorbed into the intestine and was detectable in the bloodstream within three hours; however, the fish eliminated a large majority (95%) via metabolic depuration and eventual excretion within 24 h (Ledreux *et al.*, 2014). The CTX metabolism may have happened in the fish's liver or gall bladder, similar to humans' metabolism. Ciguatoxins induce multiple cytochrome P450 enzymes in mice (Morey *et al.*, 2008). Scientists have discovered various isomers of these enzymes in fish (Yogi *et al.*, 2011), which means that the same type of hepatic metabolism could be present in fish. The ciguatoxin chemical congeners created by metabolism in the liver are either oxopene ciguatoxins (highly oxygenated) or oxocene ciguatoxins (Ledreux *et al.*, 2014). The oxopene congeners are more lipophilic and less toxic than oxocene congeners due to the butadiene side-chain on the A-ring; they are selectively retained in the striped mullet because they are deposited in fatty tissues used for energy storage. *Gambierdiscus* spp. and herbivorous fish contain both oxocenes and oxopene ciguatoxins, whereas carnivorous fish have an abundance of oxopenes (Yogi *et al.*, 2011). However, oxocene congeners are less likely to be retained, about 5%, which has implications for the trophic dynamics of CTX levels in fishes (Ledreux *et al.*, 2014). Ledreux *et al*. (2014) showed that striped mullet excreted 95% of the CTX's as oxocenes after consuming toxic dinoflagellates; thus, herbivorous fishes like mullet should have a low concentration of oxopene CTX in their tissues (Ledreux *et al.*, 2014). The excretion of oxocenes may minimize the ciguatoxic effects in striped mullet, causing short-term paralysis, not long-term behavioral impact.

For this reason, no apparent or wide-spread fish kills of herbivorous fishes are observed on reefs with CTX. However, short-term paralysis from oxocene CTX may make herbivores suspectable to predators and facilitate the transfer of the lipophilic oxopene CTX congeners to higher trophic levels. Similarly, gastropods and other benthic invertebrates that do not have livers that metabolize compounds via these P450 pathways could retain both forms of CTX (oxocenes and oxopenes) more than herbivorous fishes do; this difference in toxicity and transfer of CTX congeners via herbivorous fish and invertebrates has implications for the routes that CTX takes through the coral reef food web and my simulation modeling. Consequently, based on these reported findings, I have chosen to model the transfer of CTX in herbivorous fishes differently than in herbivorous invertebrates, assuming a 95% loss of CTX from trophic level 1 to 2 for herbivorous fishes.

Neuroblastoma Cell-Based Assay (N2a-cba) for CTX Estimation

Identifying fishes that have CTX compounds in their tissues is challenging. Accurate, rapid testing isn’t feasible for fishers and the general public consuming potentially toxic fishes. The only dockside test strips developed were riddled with issues (false positives, confusing instructions), and detection by physical examination or folk methods is highly unreliable (Bienfang *et al.*, 2011; Darius *et al.*, 2013). Two popular folk methods of detecting CTX in fishes, the rigor mortis test (RMT) and the bleeding test (BT), had the best results, with locals in Raivavae (a small island in French Polynesia) detecting 55% and 69.2% of CTX-positive samples, respectively (Darius *et al.*, 2013). These tests' unreliability and the subsequent need to detect CTX in marine species samples to confirm cases of CFP lead scientists to develop more accurate protocols for estimating CTX levels in fish tissues.

One of the most reliable methods for estimating CTX in marine species tissues is the N2a-neuroblastoma cell-based assay, which has been a robust and highly-sensitive screening tool for CTX estimation (Pawlowiez *et al.*, 2013; Reverté *et al.*, 2014). The N2a-cba can distinguish between voltage-gated sodium channel-specific toxins and other modes of action toxic compounds and is effective at discrimination between CTX's and other sodium-channel binding toxic compounds, such as saxitoxins (STX) and tetrodotoxins (TTX) (Dickey and Plakas, 2010), which allows scientists to utilize standard curves of other CTX chemical strains to estimate toxicity in species without confirming the species with LC/MS. It is more challenging to distinguish CTX's from brevetoxins (Pbtx). Since Caribbean CTX standards are not currently commercially available, we can expose the N2a cells to ouabain (O) and veratridine (V) to allow for an enhancement (CTX) or blockage (STX, TTX) of the toxic effect of the intracellular influx of Na^+ (Caillaud *et al.*, 2012). This distinction discriminates between the ciguatoxins and the saxitoxins, and tetrodotoxins. It is much harder to distinguish whether samples have CTX or Pbtx. CTX-1C has between a 440 and 2300-fold higher potency and was more sensitive (12-fold) in the N2a-cba than Pbtx's. In contrast, a different detection method, a receptor binding assay (RBA) (Hardison *et al.*, 2016), shows an 8-fold higher potency and higher sensitivity for Pbtx's, which allows scientists to distinguish between CTX's and Pbtx's in the samples (Dechraoui *et al.*, 2005). Because I did not use the receptor binding assay, I cannot eliminate the possibility of Pbtx's causing the N2a-cba cell responses I observed; however, the probability is far greater that CTX chemical congeners caused the responses due to the > 400 times higher potency of CTX congeners in the N2a cell culture bioassay.

Thus, the neuroblastoma cell-based assay allows for estimating CTX compounds in fish and phytoplankton extracts (Pawlowiez *et al.*, 2013; Pisapia *et al.*, 2017). The neuroblastoma cell line, Neuro-2a (N2a), was incubated in a mixture of Eagle's Minimum Essential Medium (EMEM) (ATCC® 30-2003™) with fetal bovine serum (FBS) and penicillin-streptomycin to prevent contamination, in a 37C environment with 5% CO_2 in a 96-well microtiter plate. The cells are natively resistant to CTX, so controls without treatment quantify cell mortality by non-sodium binding compounds in the sample (Hardison *et al.*, 2016). The experimental treatment of some of the N2a cell cultures with Ouabain (O) and veratridine (V) undoes the native resistance, so when added to the cells, O and V allows me to observe the toxic effects of CTX. Ouabain inhibits the NA^+/K^+ -ATPase pump that transports NA^+ out of the cell, and veratridine keeps the channels open in a modified-open position (Manger *et al.*, 1993; Caillaud *et al.*, 2012). In cell cultures with O and V treatments, cell death rates can be observed and compared against the control cultures. Two rows of cells were treated with a decreasing concentration of P-CTX-3C standard, from 0.001 – 2,000 pg $mL^{-1,}$ to achieve a standard dilution curve; unknown fish tissue extract samples could then be compared to the standard P-CTX-3C curve to obtain a toxicity level in P-CTX-3C equiv. Then, extracted fish samples are added to wells with and without O and V, and any samples containing CTX congeners will bind to the cells in the O and V treatment and kill them; the untreated cells are controls for other causes of death. A methylthiazolyldiphenyl-tetrazolium bromide (MTT) colorimetric assay aids in the visualization of the cells that have lived or died in each well. The MTT was added to each well, where living cells reduced the yellow dimethyl thiazol diphenyltetrazolium to purple formazan by the mitochondrial dehydrogenase. The plate was read at 540nm absorbance to quantify the remaining live cells. The cell death percentage is converted to CTX in ppb for each sample using a conversion of P-CTX-3C to C-CTX-1 in CTX3C equiv. (Hardison *et al.*, 2016). The full protocol for the N2a-cba toxicity test is provided in Chapter 4.

ECOPATH with ECOSIM

The Ecopath with Ecosim (EwE) software is used to create food web network models with data commonly collected by marine ecologists and fisheries scientists, such as biomass, production, and consumption data. An analyst using EwE takes node-specific data and inserts them into a spreadsheet-like interface that parameterizes a series of mass-balance equations; that is, every node in a network of food-web interactions must balance incoming and outgoing biomass, energy, or carbon. Ecopath then uses these parameters and the programmed mass-balance equations and linear algebra to simulate energy flows between nodes within the specified ecosystem. The network model consists of “compartments” (network nodes, which can be species or aggregated groups of species and some non-living compartments like detritus) representing all the species' biomass pools in an ecosystem that share similar diets (consumption profiles) and similar predators (consumer profiles). Each compartment must have the following data (in a standard unit of currency, normally g C m^{-2}): biomass in habitat area, production/biomass per year, and consumption/biomass per year to parameterize a model. A diet composition matrix representing the proportional amount of food consumption by consumers (columns of the matrix) from each of the other compartments in the network (rows of the matrix) is needed to complete the model. The resulting food web network model shows the flow of carbon between compartments (consumption matrix) and into and out of the ecosystem (import and export vectors). Leontif (1986) derived the network modeling approach from input-output modeling of an industrial economy, for which he won a Nobel Prize in Economics in 1973.

Network models have been applied previously in ecological studies of coral reefs. Ecopath I was the first iteration of the network model I used, which inspired the current computerized Ecopath model (Polovina, 1984). It utilizes compartments (groups of species) with a top-down predator control approach to estimate mean annual biomass given a set of parameter estimates (Polovina, 1984). Ecopath II took this principle of solving biomass budget equations and added EE (ecotrophic efficiency) coefficient and loss through exports to the model (Christensen and Pauly, 1992). The Ecopath II (also known as Ecopath with Ecosim, EwE) version is a computerized program that allows users to input these variables to simulate their food web systems. The model assumes equilibrium conditions year-to-year, which result in a series of biomass budget equations given for each compartment as:

Production of compartment biomass for *i* – all predation on *i* – other mortality of *i* = 0 (1)

(Polovina, 1984). The current model of EwE was developed by Pauly and Christensen (1992), which takes the original mass balance equation stated in (1) and adds total export and total import for each compartment:

Production of compartment biomass for *i* – all predation on *i* – other mortality of *i* – export of *i*= 0 (2)

(Christensen and Pauly, 1992). This mass balance equation can be stated as:

$$\boldsymbol{P_i - M2_i - P_i(1 - EE_i) - EX_i = 0} \quad (3)$$

where P_i is the production of i, $M2_i$ is the predation mortality of i, EE_i is the Ecotrophic Efficiency of i, (1-EE_i) is the “other mortality” of i and EX_i is the total export of i (Christensen and Pauly, 1992). The mass balance equation can be re-expressed in terms of Ecopath input:

$$\boldsymbol{B_i PB_i EE_i - \sum_{j=1}^{n} B_j QB_j DC_{ji} - EX_i = 0} \quad (4)$$

where:

B_i = Biomass of i

PB_i = Production/biomass ratio of i

QB_j = Consumption/biomass ratio of i

DC_{ji} = proportion of prey (i) of the diet of predator (j), and

EX_i = Total export from the system

From (4), a series of linear equations can be created and solved using standard matrix algebra (Christensen and Pauly, 1992).

Other related models contained within this Ecopath II software can take these initial basic input data and model flow estimates and project them over time (Ecosim), distribute them through space (Ecospace), and trace any contaminants through the food web network (Ecotracer).

Ecotracer traces a contaminant or toxic molecule's flow through the biomass pools, depending on the trophic energy flow among compartments (Ecopath Developer Site). The Ecotracer module allows for several different toxin transfer modes in the system (Figure 1-1). For compartment *i*, the contaminant can enter through direct uptake from the environment, be absorbed from consumption by compartment i, and begin in compartment *i*, as initial concentration. Thus, Ecotracer models bioaccumulation, bioconcentration, and biomagnification. Ecotracer assumes an external contaminant exists (e.g., from industrial pollution) in the environment and does not allow for toxins' biosynthesis within a compartment. So Ecotracer routines had to be modified for this study. Ecotracer then uses the original Ecopath model created with the time-steps of Ecosim (months being the lowest time step) to simulate the flow of carbon transfer between different pools and over time. Because EwE uses mass-balance equations, any contaminant that enters compartment *i* is accounted for and either transfer to the next compartment via consumption or is metabolized and excreted, after which it enters detritus through natural mortality or is removed from the system by fishing pressure or full metabolization. If compartment *i* feeds on detritus, then any contaminant in detritus enters compartment *i*. If this is the case, the contaminant re-enters the food chain, and biomagnification can begin again. The toxin enters the environment box (C0) in the model if compartment *i* excretes it.

The Ecotracer module in EwE models bioconcentration (Bienfang *et al.*, 2013) for toxicants in the environment, such as radionuclides (Walters and Christensen, 2018), the spread of pollutants (Larsen *et al.*, 2016), and transfer of PCB's (McGill *et al.*, 2017) in fishes. However, in our study, the contaminant is produced in a biosynthetic (Bienfang *et al.*, 2013) organism (photosynthetic algae). Since EwE does not allow biosynthesis but only bioconcentration, I modified the model to account for ciguatoxin's biosynthesis in dinoflagellates. To do this, I calculated the growth and production of the toxin in the dinoflagellate and entered that into the 'direct uptake' node (Walters and Christensen, personal communication, September 2020, wjw24@psu.edu, v.christensen@oceans.ubc.ca). Direct uptake is a rate of uptake into compartment *i* from the environment can be expressed as:

$$\textit{Direct uptake (from the environment)} = u_i B_i C_0 \tag{5}$$

where C_0 [$g\ m^2$] is the concentration in the environment, Bi [*g*] is the biomass of compartment *i*, and u_i [$m^2\ g^{-1}\ yr^{-1}$] is the environmental uptake rate for compartment *i* (mass of contaminant uptake/biomass/environmental concentration/year) (Walters and Christensen, 2018). Using the direct uptake from the environment (g yr^{-1}) as a proxy for CTX production by *Gambierdiscus* spp. allowed us to simulate biosynthesis in the model missing from the standard Ecotracer input parameters.

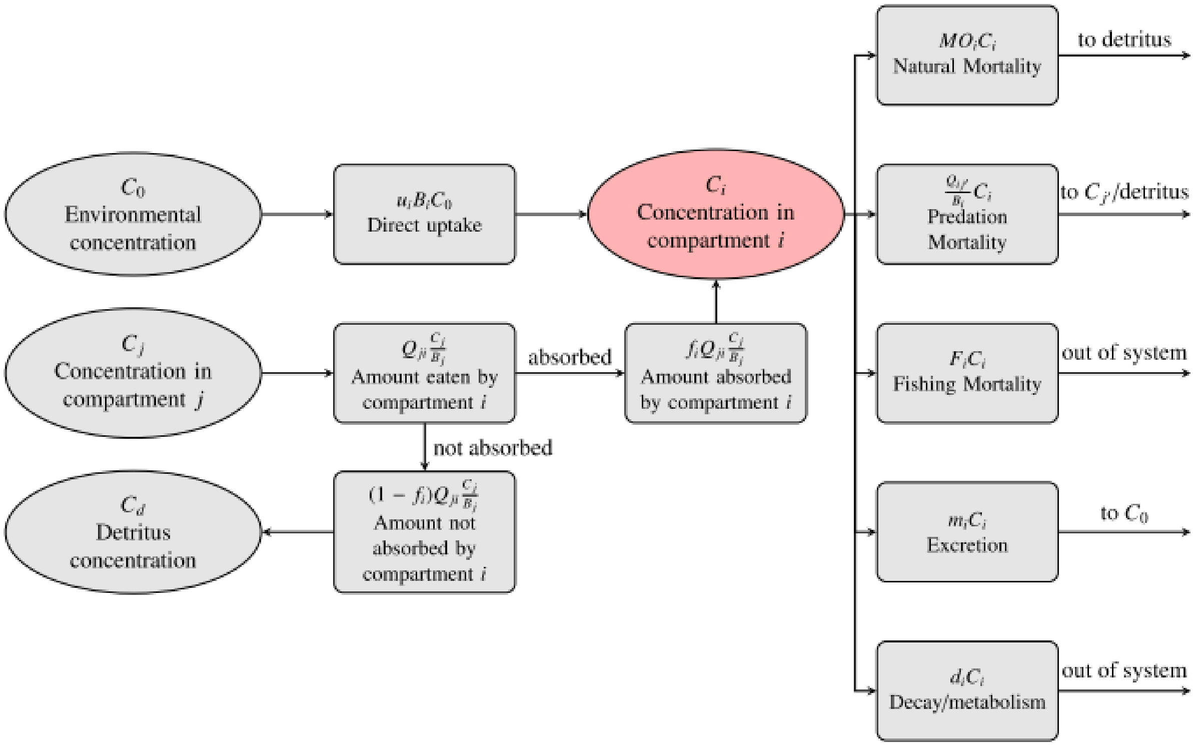

Figure 1-1 Ecotracer flow accounting for compartment i in a system (Walters and Christensen, 2018)

Traditional Ecological Knowledge

The term traditional ecological knowledge (TEK) is used to describe indigenous people’s ongoing accumulation of knowledge that is acquired from direct contact with the environment. There is no universally accepted definition of TEK and the topic is subject to interpretation. According to F. Berkes, traditional ecological knowledge (TEK) is, “a cumulative body of knowledge, practice, and belief, evolving by adaptive processes and handed down through generations by cultural transmission, about the relationship of living beings (including humans) with one another and with their environment” (Berkes 2018, p. 8) (I will be using this definition when mentioning TEK throughout the dissertation). Some researchers also use local ecological knowledge (LEK) or indigenous knowledge (IK) and it is important to note the differences in the three. TEK is ongoing accumulation of knowledge and beliefs about an ecological relationship that changes based on new observations that take place over time, while LEK is the knowledge and beliefs about ecological relationships gained from interaction with a resource which can be shared among other resource users (Charnley *et al.*, 2007). Indigenous knowledge can be interpreted as knowledge of indigenous peoples with a few tenets: *biodiversity and peoples' knowledge are inherent concepts in the idea of indigenous territoriality* (the idea of a social and physically bound space), *integral indigenous territoriality, its recognition, and reconstitution are prerequisites for enabling the creative and inventive genius of indigenous people to flourish…,* and *knowledge and determination of the use of resources are collective and intergenerational* (Viergever, 1999, p. 335-336). For this dissertation I will focus on TEK and LEK.

Merriam-Webster defines tradition as *an inherited, established, or customary pattern of thought, action, or behavior,* and can also be extended to material objects and institutions (Shils, 1981) which gives the impression of inflexibility and rigidity (Merriam-Webster, Accessed December 2020). However, this may not be the case. Traditions might undergo drastic changes, but in small, sequential steps, that over generations, are seen as small changes while the overall tradition is seemingly preserved (Shils, 1981). The next words, 'ecological knowledge,' are generally used to describe the relationships among organisms and their environments. If the term 'ecological' is used strictly in the western science sense, then there can be no traditional ecological knowledge because indigenous peoples are generally not trained ecologists (Berkes, 1993). However, if ecological knowledge describes an organism's relationship with other organisms and their abiotic environment, TEK is identical to scientific ecological knowledge

(SEK) and makes more practical sense (Berkes, 1993). SEK refers to western science based on academic, literate transmission, while TEK is mostly transmitted orally (Mazzocchi, 2006).

It is essential to understand that traditional ecological knowledge isn't just an anecdote to western science. Aboriginal people don't believe that TEK is simply a knowledge base, but a way of living life, which is different than the view of non-Aboriginal TEK scholars such as Berkes (McGregor, 2004). Aborigines view TEK as an action rather than a knowledge base and it is more about the relationship with knowledge than the actual knowledge itself (McGregor, 2004). Another difference between Aboriginal and non-Aboriginal TEK, as described by McGregor (2004), is that Native TEK is holistic and individual pieces cannot be separated from the overall body of TEK while non-Native scholars believe knowledge can be separated from the holistic view and studied independently. TEK can be understood from the indigenous people using specialized anthropological techniques such as open-ended interviews and card-sorting techniques. Understanding TEK can be useful when historical, SEK data are absent and when ecological research is constrained due to limited resources, poor replication, and short-time frames (Don, 2010).

There are similarities between TEK and SEK, such as the idea that the processes can create order out of chaos (Berkes, 1993). There are also many differences between TEK and western ecological science. Some of these include TEK being more qualitative, intuitive, and holistic than SEK; a collection of TEK data comes from hunting, catching, and observing the resources themselves, rather than researchers generating the data (Berkes, 1993).

Traditional knowledge guides society and regional customs and beliefs. Children learn from elders and use that information to navigate society as they get older. However, culture and society can also shape knowledge (Ruddle and Chesterfield, 1977). Developing a body of knowledge over generations is partly due to the information taking on its linguistic form. Local ecological vocabularies define terms like species, habitats, etc. The younger generation needs to learn the ecological vocabulary to replicate it or repeat it (Ruddle, 1991). Knowing and understanding basic ecological terms become a building block for the younger generations to understand more complex ecological relationships. It is easiest for elders to explain these concepts to the children when there is a standard set of terms. The transmission of knowledge can either be informal or formal. Previous theories show that knowledge is transmitted in an informal and disorganized way (Ruddle, 1991). However, Ruddle and Chesterfield's (1977) study of the mixed peasant economy in the Orinoco Delta in Venezuela showed that traditional knowledge transmission could be structured and systematic.

The experience of direct human contact with nature for hundreds, if not thousands of years, is essential and cannot be overstated. However, TEK may become lost during the urbanization of the world. Ecological knowledge is lost in wealthier communities and countries (Pilgrim *et al.*, 2008). Populations spend less time in nature as people become more urban, which causes dissociation between peoples and their environment and reduces local ecological knowledge. It may become more difficult for communities to manage their resources without this information (Pilgrim *et al.*, 2008). However, TEK may be increasingly useful in artisanal fisheries and data-poor regions. Interviews with fishers provide helpful information about fishing techniques (Grant and Berkes, 2007), stock assessments, spatial dynamics of fish (Mackinson, 2001; Moreno-báez *et al.*, 2010), fish behavior (Pizzini and Garcia-Quijano, 2009), and other fishing practices. It seems unlikely that TEK in itself is sufficient to make policy-decisions alone, but combining TEK with scientific studies could mean better data and better management decisions.

Combining TEK with Scientific Ecological Knowledge

Western scientists trained in the scientific method may dismiss the idea of qualitative research and how useful it can be. This can sometimes hold science and practical management back. Combining TEK with SEK may be a better way to manage resources; however, it is difficult. It is challenging to integrate both quantitative and qualitative data while giving them equal merit and consideration. This is especially true with current resource management strategies that don't allow for multiple disciplines. For example, fisheries are generally managed strictly by population and stock assessments. There has been a recent push to incorporate other factors in the decision-making process despite the natural resistance. Studies show there is an agreement between TEK and scientific data (Beaudreau and Levin, 2014) and integrating traditional ecological knowledge with scientific data is mutually beneficial to both the fisheries and fishing communities. This can be done by creating new strategies developed for management that are not hindered by a single-discipline approach (Correia *et al.*, 2018).

Although it might be in its infancy, combining TEK and SEK has been shown to be useful for management. In 2009, Gagnon & Berteaux collected information from Inuit on arctic fox winter feeding habits which was previously unidentified (Roth, 2002). Information collected from the Inuit described winter feeding habits that included animals with previously unidentified prey, such as birds (Gagnon and Berteaux, 2009). Researchers and managers could use the fox population's diet information to protect the foxes' game and allowing the population to recover naturally. Another area that utilizing TEK with SEK can be helpful is population monitoring. It is costly and time-consuming to monitor populations, especially wildlife, that migrates or is difficult to see. TEK can assist because it does not require the researchers to monitor and count populations. It has been shown that not only does TEK match the known SEK in this area of study, but additional information about species' population dynamics can be uncovered (Huntington, 2000; Gilchrist *et al.*, 2005). This information is essential, especially if managers are unaware of a drastic population decline and are making decisions based on inflated population numbers.

Another demonstrated use for TEK-SEK coupling is the identification of wildlife habitat and spawning areas. An investigation by a group looking to predict woodland caribou habitat selection compared ecological resource selection function models (RSF) with TEK-based habitat suitability index models from interviews with local members of the Taku River Tlingit territory of northern British Columbia. They found that both the TEK and RSF models were highly accurate in predicting caribou locations. The models showed agreement during the summer months and less, but still significant agreement in winter months (Polfus *et al.*, 2014), which demonstrates TEK and science-based models' ability to be used together to help predict suitable habitat for wildlife.

TEK is also effective at interpreting environmental change due to climate change or other factors. Studies have shown that indigenous populations can identify the decrease in species abundance due to climate change (Ambrose *et al.*, 2014). The bridge between SEK and TEK to understand the effects of climate change on species' populations can be attributed to five aspects of TEK which are *local-scale expertise, a source of climate history and baseline data, formulating research questions, insights into Arctic communities,* and *long-term community-based monitoring* (Riedlinger and Berkes, 2001). These five areas can be a building block to bridge the gap between scientific research and local communities' knowledge on broad topics (Riedlinger and Berkes, 2001). Local-scale expertise is the idea that since climate change will be first noticeable in the northern climates through a biophysical change in sea ice, wildlife composition, and permafrost, the Inuit communities will notice the difference. TEK can provide information about climate history and variability to compare the current changing climate and can contribute to formulating research hypotheses as an alternate way of understanding the environment. Insights into Arctic communities can describe how the local peoples respond to climate change and how their culture is altered. Lastly, much of TEK is community-based monitoring by local peoples which, are used in conjunction with western science for long term monitoring studies TEK can infer data about local species, abundance, habitat, regional change, and that TEK and scientific data can be combined to manage fisheries (Riedlinger and Berkes, 2001).

Although the evidence is clear that gathering TEK and LEK can be useful for resource managers, it has its downfalls. For one there are power struggles and dynamics that occur between SEK and LEK/TEK (Agrawal, 1995). This makes giving the two equal merit and consideration challenging when deciding the best course for managing resources and is exacerbated by the fact that modern management strategies haven’t fully grasped a multidisciplinary approach. Another critique of TEK and LEK is the lack of formal procedures for selecting “local knowledge experts” (Davis and Wagner, 2003). Davis and Wagner (2003) studied the literature and determined that the methods for selecting important informants was lacking and future studies should give more attention to the methods for identifying these informants. Also, finding similar language and translating TEK and LEK into data that western science can work with is difficult (Huntington, 2000).

Overall, TEK or SEK are not as effective to use for management strategies alone. There is great value from combining the two to achieve a complete picture of the target for management. The more refined the technique of combining multiple disciplines becomes, the more efficiently resources can be managed.

TEK and Fisheries

Fisheries are notoriously challenging to manage. There are many examples of mismanaged fisheries, which caused significant, rapid declines in stocks (Hannesson, 1996; Gjøsæter *et al.*, 2009; Froese and Quaas, 2012). Fish stocks can collapse for different reasons (Myers *et al.*, 2007), making it harder for managers to identify the issue and make recommendations to resolve it. Generally, more data is better, especially when working with selective populations. Studies have shown that using TEK from fishers can improve fisheries' management (Silvano and Valbo-Jørgensen, 2008). Also, TEK can be used in data-poor artisanal fisheries to create tools to adjust management strategies and sustainability policies (Pita *et al.*, 2016). A study in the Patos Lagoon estuary in Brazil revealed that the artisanal fishermen knew about 124 more fishing areas than the 25 regions previously known and marked on a nautical chart (Schafer and Reis, 2008). Since the information was useful, can it be recorded and used systematically to make sense for management? According to a study done in Brazil, it can (Silvano and Valbo-Jørgensen, 2008). They compared TEK and scientific data of 29 hypotheses about fisheries in Brazil and Southeast Asia that were formulated from the TEK they acquired from fishermen. The researchers showed that fishermen did have detailed knowledge about the fish behavior and ecology and the data were able to be recorded and tested against a hypothesis (Silvano and Valbo-Jørgensen, 2008). Not only were they able to test the hypotheses with these data, but in some cases the data collected via TEK correlated strongly with the scientific literature (Silvano and Valbo-Jørgensen, 2008).

Fishers understand fishes' migration, habitat connectivity, and population dynamics and anthropologists are suited to tease this information from them (Garcia-Quijano, 2007). Garcia-Quijano (2007) shows that southeastern Puerto Rico fishers are not only adept at identifying habitats for a large number of fishes, but also determining sentinel species for reef health (by identifying which fishes there are fewer numbers of despite the lowered fishing pressure), and remembering ecological patterns in spite of varying ecological diversity and complexity due to their fishing success depending on it. Fishers' success also depends on how well their catch sells at the market. If fishers are bringing substandard catch to the market or fish houses then their reputation may decline and the likelihood of them continuing to sell their catch decreases. We believe this is related to the catch of ciguatoxic fish. Fishers may avoid bringing fishes at high risk to cause CFP to the market to avoid the stigma and negative reputation for selling toxic fish. If they are knowledgeable of fish habitats and habitat connectivity, they may be aware of the dinoflagellates with those habitats that create CTX or of which fishes in certain habitats may be more or less likely to be ciguatoxic.

Using TEK to Locate Hotspots

To better manage resources, the managers should understand the ecology and the people of the area they are responsible for. It is not always efficient to perform lengthy studies on these areas due to time and cost constraints. However, they can incorporate TEK and LEK of the local peoples into what is already known. Managing fisheries is difficult due to the challenging nature of knowing true population numbers of fishes. This is an area that can benefit from experienced users' ecological knowledge

The identification of essential fish habitat is vital for fishery managers to make informed decisions. Studies have shown that fishers are able to identify essential fish habitats and presence or absence of species (Bergmann *et al.*, 2004; Garcia-Quijano, 2007; Rasalato and Maginnity, 2019). This is useful to fisheries managers when understanding the range and habitat of the species they are responsible for. A better understanding of species' habitat will allow managers to better protect spawning and nursery areas. TEK and LEK can also be used to identify important fishing sites. The knowledge fishers possess about local fishing grounds could be more comprehensive than solely relying on LIDAR data or other mapping techniques by scientists. Fishers are essentially a form of a continuous monitoring program for fishing locations. Price & Rulifson (2004) used this to their advantage when looking to reduce bycatch in the white perch (*Morone americana)* gill-net fishery. The white perch fishery is regulated by gill-net mesh size, fishing season, and net tending restrictions for the sole reason to reduce striped bass (*Morone saxatilis)* bycatch (Price and Rulifson, 2004). They acquired TEK and LEK from local commercial fishers who expressed that they could reduce the bycatch by merely placing their nets at specific sites. After fishers identified areas, the researchers set nets in those locations along with random sites for comparison. The informant-associated nets reduced the amount of bycatch of striped bass (Price and Rulifson, 2004), which shows fishers' ability to pinpoint specific sites even though fish are not stationary.

For this project, we asked fishers and other knowledgeable individuals about specific ciguatoxic hotspots and which fishes are most likely to be toxic. Ciguatoxic hotspots are challenging to identify and the test for CTX levels in fishes is not trivial to administer. Therefore, we used TEK and LEK from these knowledgeable people to identify ciguatoxic areas and fishes. We show that fishers are able to identify CTX hotspots and coldspots (regular fishing sites). Their knowledge comes both from previous generations (uncles, parents, grandparents) and peers (other fishers, friends) that have experience fishing these areas.

Puerto Rico Fisheries

Puerto Rico has a rich history of fishing with the exploitation of fisheries resources dating back to prehistoric times (Wing and Wing, 2001). Currently, fisheries in Puerto Rico are managed by the Caribbean Fisheries Management Council and more locally, the Departmento de Recursos Naturales Y Ambientales (Department of Natural and Environmental Resources DRNA). The commercial fishery is considered small-scale and artisanal with few vessels larger than 40’ (Griffith *et al.*, 2007). Nearly half of the fishers in Puerto Rico (46.5%) have an additional job outside of fishing to help supplement their income (Griffith *et al.*, 2007). Puerto Rico is divided into 78 second-order administrative divisions called municipalities which are all lead by a mayor. Most coastal municipalities have some sort of fishing activity which means they also have fish markets called *villas pesqueras*, or fish houses where fishes are bought from fishers to be sold to the public. Officially, there are between 88-100 landing centers in Puerto Rico(Griffith *et al.*, 2007). Some of the markets are privately owned, but others were built by the agriculture department in the 1970’s (Del Pozo, personal communication, December 2020, Miguel.delpozo@upr.edu). All of the publicly owned fish houses are specifically called *villas pesqueras* and have the same physical structure and arrangement, while the privately-owned fish houses could have their own name and structure. The types of fishing in our two study locations, Guayama and Fajardo, differ in many ways. In the southern municipality, Guayama, it is a more traditional artisanal fishery. Fishing traps are the dominant gear type used as well as lobster pots, however, in Fajardo it is more of a hybrid fishery with rod and reels, electric reels for deep snapper, and diving for conch being the predominant means of fishing (Del Pozo, personal communication, December 2020, Miguel.delpozo@upr.edu). Lobster pots and fish traps are not as common in Fajardo. There is also more charter fishing in Fajardo due to its geographical closeness to San Juan and Culebra[3]. This information could be used to identify risk of CFP in Guayaman and Fajardo. It is possible that the artisanal fishers in Guayama fish more with traps because traps select for smaller and possibly lower trophic level fishes with less CTX in their tissues.

There has been little work in identifying CTX hotspots and coldspots in Puerto Rico. This research adds to the previous literature that fishers in Puerto Rico are able to correctly identify CTX hotspots and coldspots. We determined that these locations are hotspots and coldspots due to the differences in overall CTX levels in the fishes (particularly the high trophic level species tested) and the 35-fold increase in toxin-producing dinoflagellates at the hotspot compared to the coldspot. Fishers identifying CTX hotspots and coldspots could be highly valuable for fishers and fisheries managers in Puerto Rico.

We are also adding to the CTX literature the number of fishes that we tested for CTX, a total of 92, of varying trophic levels. This information could be valuable to future researchers investigating the levels of CTX in a variety of coral reef fishes. This research will also add to the body of literature for dinoflagellate cell density and species at different Puerto Rico locations. Litaker *et al*. (2010) put together a graph showing the cell densities around the Caribbean, none of which were the sites that we sampled. Lastly, this study estimated the length of time it takes CTX to get from the lowest trophic levels to the top predators and provides two parameterized Ecopath models for Puerto Rico fisheries managers. The goal is for toxic dinoflagellates to be routinely sampled and use the models to help predict when fishes may be toxic or safe in a

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[3] This was before the SARS-CoV-2 global pandemic. The charters may not be operating at this time. This was a statement about the fisheries in Fajardo in normal circumstances and not under a pandemic with local shutdowns.

known time frame. We hope to increase the already-productive fisheries in Puerto Rico and help prevent future CFP outbreaks.

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Chapter 2. PRELIMINARY MODELING WITH ECOPATH

Abstract

There are a variety of vectors that cause food-related illnesses worldwide. These include bacteria, parasites, and viruses from either improper handling of food or by consuming already contaminated food. However, properly-prepared seafood can also induce toxin-related illnesses. These illnesses include ciguatera fish poisoning or CFP. The consumption of fishes containing high levels of ciguatera toxins (ciguatoxins or CTX's) causes Ciguatera fish poisoning (CFP) in humans. Single-celled dinoflagellates in the *Gambierdiscus* and *Fukuyoa* genera produce precursors to the toxins. CTX enters the food chain when herbivores consume the dinoflagellates while grazing on their preferred substrates. CTX is then biomagnified up the food chain, and eventually enters top trophic predators. If humans consume fishes with high enough toxin concentrations, it can cause various severe illnesses and, occasionally, death. There is no reliable method to detect CTX in marine species besides a rigorous bioassay that is unavailable to the public. This makes avoiding CFP difficult. We hypothesize that the quantity and species of these dinoflagellates on the reef drives the CTX levels in marine species in the same food web. The preliminary models developed in this chapter use Ecopath with Ecosim software to estimate the length of time it would take fishes to acquire >0.1 ppb (FDA recommended consumption advisory levels) of CTX in their tissues after an algal bloom of toxin-producing dinoflagellates. We show that low densities of highly toxic dinoflagellate species cause fishes to pass the 0.1 ppb consumer advisory level in less than six months. Lower toxic species at low densities cause higher trophic level fishes to cross the 0.1 ppb mark in 16 months. Knowing how long it takes for fishes to become toxic may be a robust tool for

managers attempting to reduce the number of cases of CFP and to ensure a robust and productive fisheries economy, which is an integral part of the economic structure of Puerto Rico.

Introduction

CFP is a human health concern caused by consuming seafood with high levels of CTX. CFP is endemic to tropical and subtropical regions globally. However, rising sea temperatures and the demand for imported seafood from these regions means cases are spreading beyond the tropics (Mattei *et al.*, 2014; Rodríguez *et al.*, 2017; Friedemann, 2019; de Haro *et al.*, 2020). If humans consume marine species with high levels of CTX then ciguatoxin fish poisoning can occur (Pearn, 2001). The toxin, produced by photosynthetic dinoflagellates in the *Gambierdiscus* and *Fukuyoa* genera, enters the food web when benthic invertebrates or herbivorous fishes feed on their preferred substrates and indirectly consume the dinoflagellates (Randall, 1958; Lewis, 2001; Ledreux *et al.*, 2014). The dinoflagellates migrate up and down in the water column to respond to light conditions, salinity, temperature, and nutrient availability, eventually settling on benthic autotrophs (Kamykowski, 1981; MacIntyre *et al.*, 1997). These dinoflagellates are closely associated with a wide variety of macroalgae. *Gambierdiscus toxicus* (a species of dinoflagellate that produces the precursor to CTX) is an epiphyte of macroalgae (Holmes *et al.*, 1991). The number of cells g^{-1} wet weight (ww) algae can range from just a few cells to over 100,000 cells g^{-1} ww algae (Litaker *et al.*, 2010). The most frequently observed abundances of cells on benthic algae are from 0-1000 cells g^{-1} ww algae (>85%) with less than 10% of observations in the 1000-100,000 cells g^{-1} ww algae range (Litaker *et al.*, 2010).

Ciguatoxin is metabolized in fishes after they consume the benthic dinoflagellates. Although the metabolism of ciguatoxins by marine consumers is not well understood, Ledreux *et al.* (2014) investigated the trophic transfer dynamics of CTX from *Gambierdiscus polynesiensis* cells to the second trophic level (*Mugil cephalus,* striped mullet) and showed that once ingested, ciguatoxin is rapidly absorbed in the bloodstream but fish excrete a large majority (95%) due to liver metabolism (Ledreux *et al.*, 2014). A series of cytochrome P450's metabolizes toxins when they enter human or mouse livers. This same response could be happening in fish (Guengerich, 2008). Ciguatoxins induce multiple cytochrome P450 enzymes in mice (Morey *et al.*, 2008), and scientists discovered multiple isomers of these in fish flesh (Yogi *et al.*, 2011). The same type of hepatic metabolism could be present in fish. When metabolized, the congeners created are either highly oxygenated oxopenes or oxocenes (Ledreux *et al.*, 2014). Herbivorous fish are more likely to contain oxocenes, whereas carnivorous fish have an abundance of oxopenes (Yogi *et al.*, 2011). However, oxocene congeners are less likely to be retained, which may be one reason for the low retention rate of ciguatoxins ~5% (Ledreux *et al.*, 2014). There are different metabolic rates among species of marine organisms, particularly between organisms with and without a liver. Fishes, for example, metabolize the toxin differently than gastropods, which could affect the biomagnification of CTX to different organisms.

Ecopath with Ecosim is a software designed to simulate food web models. It is useful because the data needed to parameterize the models are data often collected fisheries data such as, biomass (g m^{-2}), production/biomass ratio (P/B), consumption/biomass ratio (Q/B). A predator/prey diet composition matrix is also needed to parameterize the food webs. The software uses a mass-balance approach using two master equations, describing the production long term and one for each group's energy balance (ECOPATH developer site). The first Ecopath equation,

Production = catches + predation mortality + biomass accumulation + net migration + other mortality (1) or more formally,

$$P_i = Y_i + B_i M2_i + E_i + BA_i + P_i(1 - EE_i) \tag{2}$$

where P_i is the total production rate of group (i), Y_i is the total fishery catch rate of (i), $M2_i$ is the total predation rate for group (i), E_i is the net migration rate (emigration – immigration), BA_i is the biomass accumulation rate for (i), while P_i * (1-EE$_i$) is the 'other mortality' rate for group i, describes how the production for each group is split into components (ECOPATH developer site). The second Ecopath equation defines the energy balance of a compartment,

Consumption = production + respiration + unassimilated food (3)

A compartment may be a group of ecologically-related species, a single species, or a single size/age group of a given species (ECOPATH developer site).

Production is the elaboration of tissue by group i over time (ECOPATH developer site). When applied to mass-balance models, total mortality is equal to production over biomass (Allen, 1971). Therefore, total mortality (*Z*) of a group can be used as the P/B ratio in EwE (ECOPATH with ECOSIM developer site). Total mortality (*Z)* is the sum of the natural mortality (*M*) of group i plus fishing mortality (*F*) of group i. If catch-at-age data is unavailable or group i is not fished, natural mortality can be used for P/B. Natural mortality is better suited for groups with no fishing pressure, and *Z* is used when fishing occurs. Opitz assumed no fishing pressure in her model, so *M* is used for all groups. *M* can be estimated by using an empirical relationship among *M*, two variables from the von Bertalanffy Growth Function (vBGF) ($K$and L_∞), and the mean habitat temperature for group i (Pauly, 1980)

$$M = K^{0.65} * L_\infty^{-0.279} * T_C^{0.463} \tag{4}$$

where M is the natural mortality, K is the growth parameter from the vBGF, and T_c is the mean habitat temperature for group i.

Opitz (1996) published a 50-compartment coral reef food web from the Caribbean that focused on Puerto Rico using previous literature, including Randall's (1967) diet composition data. Opitz performed a cluster analysis on variables that reflected the species' food consumption, the species' size, the species' activity level, and the type of food the species consumed (Opitz 1996). The non-fish compartmentalization was more complicated since their taxonomic range was much more extensive. Opitz (1996) follows a different series of criteria: availability of data for P/B and Q/B ratios, size, diet similarity, lifestyle similarity, and taxonomic closeness (Opitz 1996).

The preliminary models displayed in this chapter estimate ciguatoxins' movement through coral reef food webs. The models also show the length of time after a bloom, marine species may become toxic, and which species are more likely to accumulate high toxin levels. The models used Opitz's (1996) data from her 50-compartment model with compartments for *Gambierdiscus* spp. These models will direct future sampling and food web model estimation in Puerto Rico.

Methods

To estimate the length of time it takes CTX to biomagnify in coral reef food webs, several marine food web networks were parameterized with varying cell densities and species of toxin-producing dinoflagellates by adding a dinoflagellate compartment to a previously balanced, 50-compartment coral reef food web (Opitz, 1996). Previously published literature on the growth rate, abundance, and toxicity of chemical strains was used to provide the data for the new dinoflagellate compartments. The CTX pathways were traced through the newly-parameterized food web models using the Ecotracer module within EwE. All calculated values for this chapter are in Appendix E.

The module Ecotracer was designed to simulate toxicants' bioconcentration from the environment rather than biosynthesized toxins' biomagnification. To adjust the model for biosynthesis, I used the direct absorption rate input parameter. This is a rate of absorption of toxin from the environment (g m^{-2}) into the specific compartment. Using the direct absorption rate allowed me to use the absorption as a proxy for the production of toxins in the compartment. This was done based on the recommendation of the creators of the Ecopath with Ecosim software (Walters and Christensen, personal communication, September 2020, wjw24@psu.edu, v.christensen@oceans.ubc.ca). The cells were assumed to be in a steady-state. Any production over the original cell quantity was assumed to be consumed by predators. This value was entered into the direct absorption rate box in the Ecotracer module. Since the direct absorption rate is a percentage of the toxin in the environment, the initial concentration in the environment was set to 1 g m^{-2}.

I recreated the 50 compartment Opitz (1996) model in Ecopath with Ecosim. A new compartment was added to the model to represent toxin-producing dinoflagellate species with varying toxicity levels and growth rates that are common to the Caribbean. *Gambierdiscus excentricus*, has a slower growth rate but is highly toxic, and *Gambierdiscus carolinianus*, has a faster growth rate but is less toxic, were both species used in separate models. We chose those species due to the difference in toxicity and growth rates and accessibility to weight data for the cells from the NOAA Southeast Fisheries Science Center Beaufort Lab, which was essential for calculating each species' basic input parameters. We parameterized two models using these data, one for each species and 1000 and 100,000 cells g^{-1} wet weight algae based on concentrations established by. Litaker *et al*. (2010). Cell counts of 1000 cells g^{-1} wet weight algae are relatively common, while 100,000 cells g^{-1} wet weight algae are less common but still observable. These two simulations represent a more common occurrence (1000 cells g^{-1} wet weight algae) and a worst-case scenario (100,000 cells g^{-1} wet weight algae).

We calculated basic input data for the new compartments (biomass and production per biomass or P/B) and assumed the dinoflagellates were adhered to the benthic autotrophs to simulate the dinoflagellates in close association with a wide variety of macroalgae. Herbivores and herbivorous fish consuming these toxic dinoflagellates while grazing on their preferred substrates are part of Randall's food chain hypothesis on the entry of the CTX from *Gambierdiscus spp.* to the food web. This assumption was useful for two reasons; it allowed us to convert the species' biomass in the system using the Litaker *et al*. (2010) estimates of 100 and 100,000 cells g^{-1} wet weight of algae to g m^{-2}. This assumption allowed us to parameterize the diet composition matrix to automatically force the species to feed on benthic autotrophs to consume the toxic dinoflagellates. The number of cells g^{-1} wet weight algae was multiplied by the benthic autotroph biomass (g m^{-2}) from the model to get the number of cells m^{-2} to find the total biomass (g m^{-2}) of the dinoflagellates needed for the basic input. The total biomass was then multiplied by the individual cell mass (Holland C., personal communication, July 2020, chris.holland@noaa.gov) to calculate the compartment's biomass for *Gambierdiscus excentricus* in g m^{-2}. A detrital import value of 15,000 g m^{-2} $year^{-1}$ was added to the basic input parameter table to balance the model. The output from the basic input parameters showed an ecotrophic efficiency of detritus >1; detritus was consumed more than it was being generated.

I used cell division rates to calculate the production/biomass/year ratio (P/B). The only available growth rates were from a lab under optimal growth conditions. Therefore, growth rates might be higher than expected in the environment. However, this is the best estimate available for the cells. We multiplied the division rate d^{-1} for each species by the cell weight and the total number of cells on 1300 g m^{-2} benthic algae (Opitz, 1996) to get biomass g m^{-2} per day. This value was converted to biomass $year^{-1}$ and divided by the dinoflagellates' weight on the total algal biomass to get production/biomass/year.

I reduced the proportion of the predators' diet on the benthic autotroph prey to consider the different dinoflagellate cell densities and species added to the models to parameterize the diet composition matrix. The diet matrix was adjusted to reflect the assumption that the dinoflagellates adhere to the benthic autotrophs. If predator *j* consumed the benthic autotrophs, they also consumed the dinoflagellates in the new diet matrix. We altered each predators' diet to include the new compartment. As a proportion of the predator diet, the benthic algae prey compartments were reduced by the biomass of *Gambierdiscus* sp. included in the predator diet. To find the new ratio of benthic autotrophs in the predators' diet, we subtracted the proportion of dinoflagellates from the original proportion of benthic autotrophs in the diet done for all prey compartments in the predators' diet for each model.

The contaminant tracing in Ecotracer was the next step after the basic input, and the diet composition matrix was parameterized. We used an assumption of steady-state in the model; predators consumed growth calculated over the model's base number of cells. We entered this into the direct absorption rate box for the *Gambierdiscus* spp. compartment. The number of cells were multiplied by the growth rate in divisions day^{-1} to find new growth. Each model used a growth period of 30 days because Ecosim works with monthly time-steps. The number of cells

g^{-1} ww algae was multiplied by the algal biomass in g m^{-2} (1300) to get the total number of cells m^{-2}. The total number of cells was multiplied by the growth rate in $divisions^{-1}$ for the specific species, then by the amount of CTX3C equiv. per cell (See Appendix D Table D-6). Growth and production of CTX after 30 days were subtracted by the original value to get excess growth for one month. I entered this value into the direct absorption rate box in g m^{-2} $year^{-1}$ and set the initial concentration in the environment to 1 t km^{-2}; the direct absorption rate box is a proportion of the environmental concentration. The initial concentrations for each species were then calculated and added to the initial concentration box in the Ecotracer module. The toxicity of the species in fg $cell^{-1}$ was divided by the cell's weight to get the initial concentration in g toxin g^{-1} cell.

A 5% retention rate, or 0.95 proportion of contaminant excreted, was added to compartments in the Ecotracer module with fishes with a trophic level lower than 3.0 that consumed benthic autotrophs (compartments 9, 10, 11, 16, 19-26) to simulate the metabolism and excretion of CTX-oxocene congeners by fishes. Compartments of fishes > 3.0 ETL retained 100% of the toxin since the amount of oxopene congeners retained is unknown.

All input data is listed in Appendix D.

Results

The contaminant tracing results with the Ecosim/Ecotracer module indicated large differences in toxin accumulation between 100 and 100,000 cells g^{-1} wet weight algae for both species (Figure 2-1). After 25 months, the simulation with the low toxicity species and small bloom (C) achieved levels of 0.2 ppb in the large jacks compartment, compared to the simulation with the high toxicity species (*G. excentricus*) with a small bloom (A) where the large jacks compartment reaches nearly 9 ppb. There were also considerable differences in CTX concentrations between species with a large bloom scenario of 100,000 cells g^{-1} ww algae. The large jacks compartment in the higher toxicity species model (B) acquired very high concentrations of toxin at nearly 2000 ppm, or 2,000,000 ppb after 25 months, while the large jack compartment in the model with the lower toxicity species (D) saw CTX levels of 20,000 ppb. These values are not realistic biologically.

Consistent with expectations, the models' overall toxicity estimates using the higher toxicity *G. excentricus* species were higher than those with the lower *G. carolinanus* species. The models starting with 100 cells g^{-1} wet weight algae most likely represent actual CTX-like activity in real-life biological ecosystems.

The contaminant tracing results also showed a difference when it takes compartments of varying ETL's to reach the FDA recommended consumer advisory level for Caribbean CTX of 0.1 ppb. Figure 2-2 presents the results that show it takes fewer months to reach 0.1 ppb in a simulation with the higher toxicity species for all compartments over ETL 2.0. The hemiramphidae, gastropods, and large scarids compartments never reach 0.1 ppb in the model with the lower toxicity species. These results display that the more toxic species drives the concentration of CTX in species to over 0.1 ppb more rapidly than the lower toxic species.

Inconsistent with previous expectations, the increase in ETL does not always correlate with a decrease in the length of time it takes these compartments to reach 0.1 ppb.

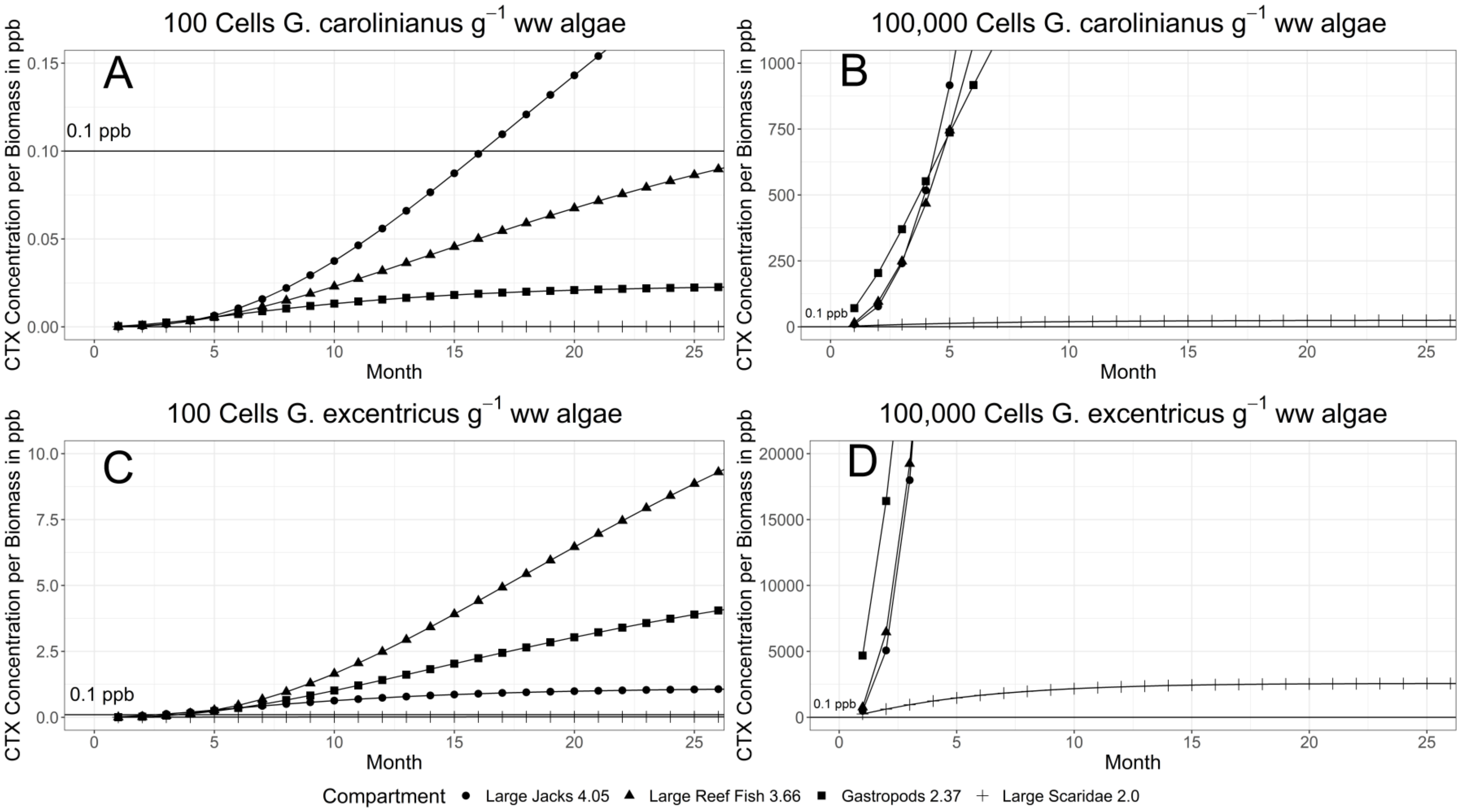

Figure 2-1 CTX concentrations per biomass in ppb of four compartments from the Opitz 1996 model with compartments added for 100 and 100,000 cells g^{-1} wet weight algae for both the highly toxic *Gambierdiscus excentricus* and low toxicity species, *Gambierdiscus carolinianus* of CTX-producing dinoflagellates for one month of growth of the cells. The most important species in each fish group (by biomass) are Large jacks- *Seriola dumerili* and *Trachinotus falcatus*, Large Reef Fish- *Diodon holocanthus and Sphyraena barracuda*, and Large Scarids- *Scarus guacamaia* and *Scarus vetula*. The numbers in parentheses are calculated trophic levels for the corresponding group.

For the model with the less toxic species (*G. carolinianus*), it took the intermediate reef fish (ETL 3.44) to pass the 0.1 ppb mark at 20 months, while it took higher ETL compartments, large reef fish (3.66), and large groupers (3.89), 30 and 44 months respectively. For the model with the more toxic species (*G. excentricus*), the hemiramphidae (2.52) group was quicker to pass the 0.1 ppb threshold (5 months) than the large groupers (6 months). These results indicate that food chain pathways may have a large effect on CTX levels.

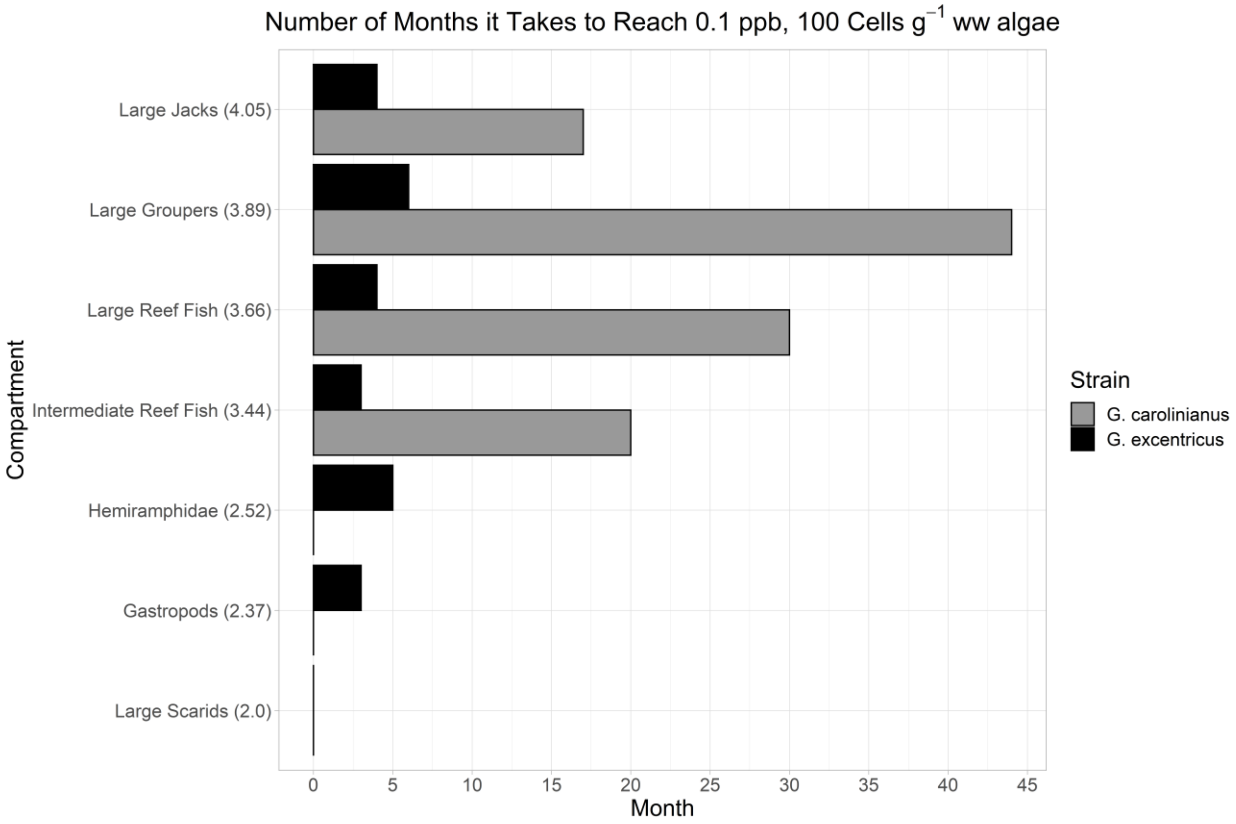

Figure 2-2 Figure displaying the number of months it takes different compartments of varying trophic levels to achieve 0.1 ppb when starting with 100 cells g^{-1} ww algae for both low and high toxicity *Gambierdiscus* spp. The numbers in parentheses are ETL (effective trophic level).

Discussion

One of the most interesting findings from this study is how toxic some species can become with a low density of highly toxic cells (Figure 2-1 A). According to Litaker *et al*.

(2010), 100 cells g^{-1} ww algae is an expected density of dinoflagellates in coral reef systems. This small concentration of highly toxic cells can cause high trophic level species (large jacks) to reach nearly 10 ppb in 25 months. This level of CTX would be highly toxic to humans if consumed.

Less likely, although biologically possible, is a large bloom of a highly toxic species (Figure 2-1 B). *Gambierdiscus excentricus* is one of the slower-growing toxin-producing dinoflagellates but is highly toxic (469 fg $cell^{-1}$) (Litaker *et al.*, 2017). Since these cells divide slowly, it may be challenging to divide fast enough to create a large bloom scenario. However, this concentration of highly toxic cells would represent a theoretical worst-case scenario. The compartments in this simulation were above the 0.1 ppb threshold in the first month, and the large jacks reached CTX levels of 2,000 ppm in 25 months (Figure 2-1 D). These models significantly overestimate the CTX levels in fishes.

Similarly, a large bloom with the less toxic species also causes the compartments to be above the 0.1ppb threshold in the first month. However, the CTX levels do not get as high as the simulation's compartments with the highly toxic species, although the large jacks exceed 20,000 ppb after 25 months. It is improbable that these toxin levels can accumulate in fishes due to metabolism and excretion and the fish wouldn’t be without debilitating effects or even death. Fishes experience hyper- and hypoactive behaviors after consuming ciguatoxin-producing dinoflagellates (Ledreux *et al.*, 2014). Researchers investigated these behaviors and saw some hypoactive actions, which included; relaxation of the jaw, resting at the bottom of the tank, fin paralysis, and loss of equilibrium (Ledreux *et al.*, 2014). The fishes' hyperactive actions were erratic swimming, jerky feeding habits, and convulsions (Ledreux *et al.*, 2014). These behaviors could increase the fish's vulnerability, which would make them more susceptible to predation, therefore reducing the time that fish could accumulate ciguatoxin. Scientists have identified CTX levels of 2.0 ppb C-CTX-1 (Lewis *et al.*, 1999), and C-CTX-1 levels could get as high as 10–20 ppb (Holland C. and Litaker W., personal communication, March 2016, chris.holland@noaa.gov, wayne.litaker@noaa.gov). We believe the reason for the high levels of toxicity is the unlikely extremely high number of cells calculated for the model (100,000 cell g ww^{-1} algae). This may occur in some reefs in short time frames and it is unlikely that it would stay that high for months. Future models that we parameterize should use dinoflagellate data that we collect.

The most likely scenario is a low density of cells with a less toxic species, represented by 100 cells of *G. carolinianus* g^{-1} ww algae (Figure 2-1 C). Even with few cells of a relatively low toxic species, large jacks still cross the 0.1 ppb threshold, although it takes 16 months. The large reef fish compartment is close to the 0.1 ppb threshold after 25 months and crosses above 0.1 ppb in 30 months (not shown). The gastropods and scarids reached a plateau at relatively low levels of CTX, which represents a scenario where a species starts to colonize an otherwise "clean reef" (with no previous issues of CTX) and causes the fishes to accumulate CTX in their tissues. The top trophic level predators may become toxic while the lower trophic levels stay under the 0.1 ppb threshold.

The data shows that low concentrations of highly toxic cells cause the compartments listed, besides the scarids, to be above the 0.1 ppb threshold in 6 months or less (Figure 2-2). If small numbers of cells can cause toxicity in fishes, that is a significant concern to managers and fishers where a reef could become highly toxic in a few months with a small bloom of a highly toxic species. Fishes that were typically safe to catch would become toxic with little warning or time to change fishing locations. It would only be apparent that the site is contaminated when consumers start to report illnesses. Due to the underreporting of CFP, the information might not get back to the fishers for months.

The preliminary models show that estimating CTX biomagnification in fishes is possible, but our models significantly overestimate CTX over time. The low-density *G carolinianus* simulation gave CTX measurements between 0 and 0.2 ppb after 25 months (depending on the trophic group), which is the range observed in species tested for CTX in the Caribbean (Dechraoui *et al.*, 2005; Martin *et al.*, 2015; Loeffler *et al.*, 2018). However, higher trophic level species are above levels seen in the literature. In the scenarios with 100,000 cells of *G. excentricus*, CTX is overestimated. This is due to the high concentration of CTX in the cells and the high density of cells assumed in the model. This is an unlikely scenario and future models will take into account more realistic dinoflagellate counts.

The literature and research on Caribbean CTX's are not as extensive as the literature and research on Pacific species. Therefore, to better parameterize the models, I made assumptions based on data from Pacific CTX's. According to a study by Yogi *et al*. (2011), there are different structures of CTX's and each have their properties and congeners that form as a result of metabolic activity. Several different oxopene and oxocene congeners are produced when the precursor compounds P-CTX-1B, and P-CTX-3C are metabolized, respectively (Yogi *et al.*, 2011). Although dinoflagellates can produce both oxopene and oxocene precursors, oxocene congeners are more common in algae, while highly oxygenated oxopene congeners are common in piscivores (Chinain *et al.*, 2010; Yogi *et al.*, 2011; Ledreux *et al.*, 2014). Ledreux et al. (2014) suggest this is due to piscine detoxification pathways and led them to suggest that oxopene congeners are poorly retained. However, the more fat-soluble oxopene congeners are retained and increase in potency over time due to further metabolism. (Ledreux *et al.*, 2014). In all simulations, I assumed 95% excretion of CTX's for compartments with an ETL < 3.0 and set the proportion of contaminant excreted box in the Ecotracer module to 95% for these compartments (compartments 9, 10, 11, 16, 19-26).

When modeling biotoxins in marine systems, it is vital to understand all of the contaminants' pathways, mostly in the detrital pool. As previously mentioned, oxopene congeners are in piscivores. When fishes die from natural mortality, their tissues become organic detritus, and any CTX still in their tissues would subsequently enter the detrital pool. Fishes that feed on detritus uptake this CTX in the form of oxopene congeners, not the oxocene congeners from feeding on algae, which are quickly metabolized and excreted. In Ecotracer this is modeled by the concentration in compartment i (C_i) entering the detrital pool via natural mortality (mortality rate MO_i) as C_iMO_i (Walters and Christensen, 2018).

I used Opitz's 1996 model because it was a finished Caribbean coral reef food web model, the data were in the same format that the Ecopath software accepted, and it used well-respected diet data from Randall (1967). Future studies should better parameterize models for CTX estimation. Opitz's aggregating species' methods using Q/B values and diet data caused some species grouping that were less ideal for modeling CTX biomagnification. For example, Opitz's compartment 12, Large reef fish (carnivorous) (seen in Figures 2-1 and 2-2), contains *Sphyraena barracuda* (great barracuda) and *Lachnolaimus maximus* (hogfish). Barracuda are primarily piscivores, while hogfish are molluscivorous. This distinction is essential to make when modeling a biotoxin through food web models where the pathway that the toxin takes could be a driving factor in how toxic each compartment can become and how quickly they reach highly toxic levels. It was difficult to make biological inferences from the preliminary models due to the widely varying diets in some compartments. For modeling toxin contamination l believe that grouping species solely on diet data alone is acceptable. Also, Opitz’s model was parameterized for a general Caribbean coral reef. The models needed to be further parameterized for specific reefs to investigate why individual reefs or fishing areas become toxic, and others don’t, and how quickly they become toxic. Therefore, specific reefs should be targeted for sampling to create new models, one for high levels of CTX (hotspot) and one for low levels of CTX (coldspot).

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Chapter 3. USING TRADITIONAL ECOLOGICAL KNOWLEDGE AND LOCAL ECOLOGICAL KNOWLEDGE TO IDENTIFY CTX HOTSPOTS AND COLDSPOTS

Abstract

Traditional ecological knowledge (TEK) and local ecological knowledge (LEK) have been shown to be effective at supplementing data in data-poor regions as well as area where long-term ecological studies are ineffective or inefficient. We believe that TEK and LEK from fishers in Puerto Rico can be used to identify CTX hotspots and coldspots to help prevent ciguatera fish poisoning (CFP). CFP is associated with consuming high ciguatoxins (CTX) in fishes in tropical and sub-tropical regions. To understand the fishers' knowledge of CTX around Puerto Rico, we interviewed 21 fishers in Puerto Rico. We identified *Villas pesqueras,* or fish houses, in different Puerto Rican municipalities to sample for data collection and interviewed fishers who were identified with a modified snowball sampling approach. We asked them a series of questions in an open-ended interview format, which included a pile sort to determine which fishes they avoided due to the potential for CTX. They were not asked about specific locations but asked to circle areas they believed had high levels of CTX on a map or verbally identified the reef according to what municipality was closest (i.e., Guayama hotspot, Fajardo coldspot). The data show that the fishermen identified the reefs off the coast of Guayama and Salinas as CTX hotspots. The informants agreed that the hogfish (*Lachnolaimus maximus*), great barracuda (*Sphyraena barracuda*), black jack (*Caranx lugubris*), cero (*Scomberomorus regalis*), king mackerel (*Scomberomorus cavalla*), amberjack (*Seriola dumerili*), and horse-eye jack (*Caranx latus*) were most likely to have high levels of CTX. These results suggest that fisheries

managers could use TEK and LEK to identify problematic ecological hotspots, such as those that are likely to have toxic fish from CTX in specific reefs.

Introduction

The term traditional ecological knowledge (TEK) is used to describe indigenous people's ongoing accumulation of knowledge that is acquired from direct contact with the environment. There is no universally accepted definition of TEK and the topic is subject to interpretation. According to F. Berkes, traditional ecological knowledge (TEK) is, "a cumulative body of knowledge, practice, and belief, evolving by adaptive processes and handed down through generations by cultural transmission, about the relationship of living beings (including humans) with one another and with their environment" (Berkes, 2018, page 8). Some researchers also use local ecological knowledge (LEK) which is the knowledge and beliefs about ecological relationships gained from interaction with a resource which can be shared among other resource users.

It is essential to understand that traditional ecological knowledge isn't just an anecdote to scientific ecological knowledge (SEK), or "western science". Aboriginal people don't believe that TEK is simply a knowledge base, but it is a way of living life, which is different than the view of non-Aboriginal TEK scholars such as Berkes (McGregor, 2004). Aborigines view TEK as an action rather than a knowledge base and it is more about the relationship with knowledge than the actual knowledge itself (McGregor, 2004). Another difference between Aboriginal and non-Aboriginal TEK, as described by McGregor (2004), is that Native TEK is holistic and individual pieces cannot be separated from the overall body of TEK while non-Native scholars believe knowledge can be separated from the holistic view and studied independently. TEK can be understood from the indigenous people using specialized anthropological techniques such as open-ended interviews and card-sorting techniques. Understanding TEK can be useful when historical, SEK data are absent and when ecological research is constrained due to limited resources, poor replication, and short-time frames (Don, 2010).

Fishers understand fishes' migration, habitat connectivity, and population dynamics and anthropologists are suited to tease this information from them (Garcia-Quijano, 2007). Garcia-Quijano (2007) shows that southeastern Puerto Rico fishers are not only adept at identifying habitats for a large number of fishes, but also determining sentinel species for reef health (by identifying which fishes there are fewer numbers of despite the lowered fishing pressure), and remembering ecological patterns in spite of varying ecological diversity and complexity due to their fishing success depending on it. Fishers' success also depends on how well their catch sells at the market. If fishers are bringing substandard catch to the market or fish houses then their reputation may decline and the likelihood of them continuing to sell their catch decreases. We believe this is related to the catch of ciguatoxic fish. Fishers may avoid bringing fishes at high risk to cause CFP to the market to avoid the stigma and negative reputation for selling toxic fish. If they are knowledgeable of fish habitats and habitat connectivity, they may be aware of the dinoflagellates with those habitats that create CTX or of which fishes in certain habitats may be more or less likely to be ciguatoxic.

This study aims to identify and use the TEK and LEK of fishers in Puerto Rico to identify ciguatoxic hotspots and coldspots in Puerto Rico and investigate which fishes they avoid due to the potential for CFP. We conducted open-ended interviews and pile sorts to determine locations with higher ciguatoxin levels in the fishes' tissues and which fishes are likely to be avoided due to the potential for CFP. These sites and fishes will be sampled for CTX estimation for confirmation.

Methods

We conducted 21 interviews with commercial fishers in Puerto Rico to, identify hotspot and coldspot locations to sample fishes for CTX estimation and determine which fishes would likely have higher levels of CTX in those areas. These data will guide the protocol for both the fish and toxic dinoflagellate sampling.

We chose informants by geographically targeting *Villas pesqueras* (fish houses) in different municipalities on the southeast, east, and northeast coasts of Puerto Rico. These key informants were chosen based on their knowledge regarding Puerto Rico's fisheries in general and ciguatera in particular. The interviews took place in different municipalities on the southeast, east, and northeast coasts of Puerto Rico. At the beginning of the fieldwork, we were unsure of where to start. As mentioned before, literature about CTX hotspots and coldspots is poor everywhere, including Puerto Rico. The first interview took place in the west with a highly respected and knowledgeable fisher, Adán[4]. This first interview extended to almost three hours, providing a great deal of data that lead us to move toward the East.

Using a snowball sampling technique, we located other knowledgeable individuals (Figure 3-1). Along with the informants mentioned being on the southeast and east coast, the east coast of Puerto Rico was easier to sample the east coast of Puerto Rico for fishes and toxic dinoflagellates due to shorter travel times. San Juan is a location to acquire diving equipment. Also, the University of Puerto Rico at Humacao permitted us to use the biology laboratories for sample workups, which is on the east coast. We interviewed the newly found informants with a semi-structured format which allowed for specific questions essential for analysis and allowed

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[4] Adán is a pseudonym to protect the informant's identity.

the interviewees to expand upon subjects they saw fit. The preformed seven questions are listed in Appendix E.

Interviews were voice recorded and coded into data to identify where the perceived hotspots and coldspots were. Informants were allowed to describe where they believed the hotspots were in a free listing exercise. They identified locations using the municipality along the ocean in those areas (i.e., Guayama hotspot), but we also provided nautical charts too so they could pinpoint the sites. The goal was to identify hotspots and coldspots along the coast.

A pile sort was administered to all interviewees to investigate which fishes and other marine species they believed were most toxic. These data were used to identify which fishes would be best for sampling. We created a set of laminated cards with a different species of fish on each one. The fishes on the cards consisted of commonly caught species of commercial value in Puerto Rico except for *Sphyraena barracuda*. Barracuda were included because they are known to have high levels of CTX in their tissues, and the Puerto Rican government has a moratorium on the commercial catch and sale of this species due to CTX concerns. The informants put the cards into two piles according to whether they avoid catching that species due to CTX or not. Results were analyzed using a consensus analysis in UCINET.

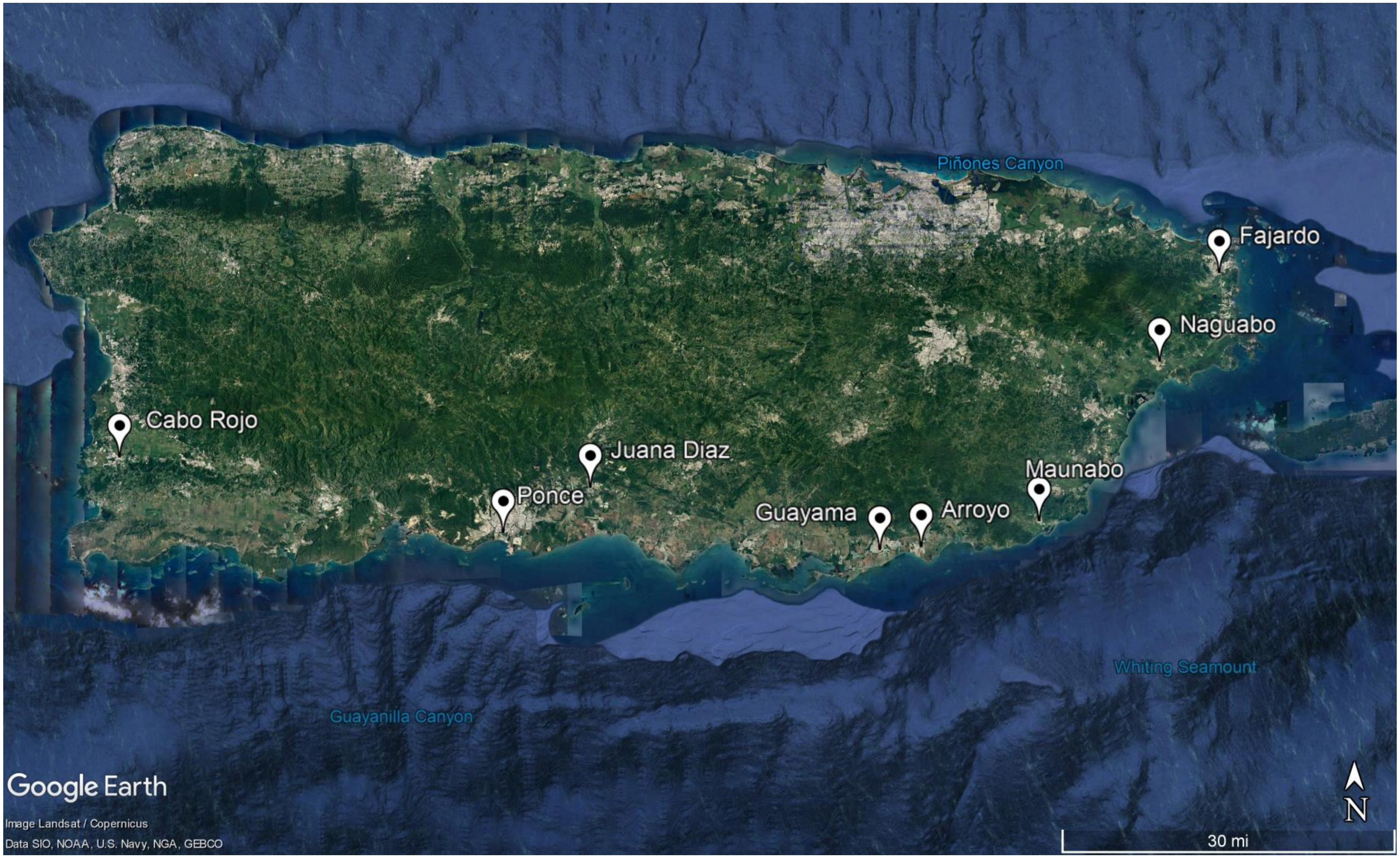

Figure 3-1 Locations of interviews conducted in Puerto Rico. Twenty-one interviews were conducted in total: one interview in Cabo Rojo, five interviews in Fajardo, five interviews in Guayama, three interviews in Arroyo, three interviews in Juana Diaz, two interviews in Ponce, one interview in Maunabo, and one interview in Naguabo.

Results

The results of the free-listing exercise for hotspot identification show that 12 out of 21informants believed that Guayama was a hotspot for CTX. Figure 3-2 is a visual network representation of these data. The circular nodes represent informants. The square nodes represent the locations they listed as a hotspot (any area with higher levels of CTX, or more informally for the fishermen, had more toxic fish on the reef). The data shows that most fishers chose Guayama and Salinas as having high levels of toxin. Out of the 21 fishers interviewed, 12 of them identified Guayama as a hotspot area. Guayama and Salinas are both on the Southeastern coast of Puerto Rico (Salinas is 15 miles west of Guayama) and fishers from those municipalities share common fishing grounds. Few fishermen chose cities on the eastern and northeastern coast of Puerto Rico. No fishermen selected Fajardo, which is an area in the Northeast known for its commercial fisheries.

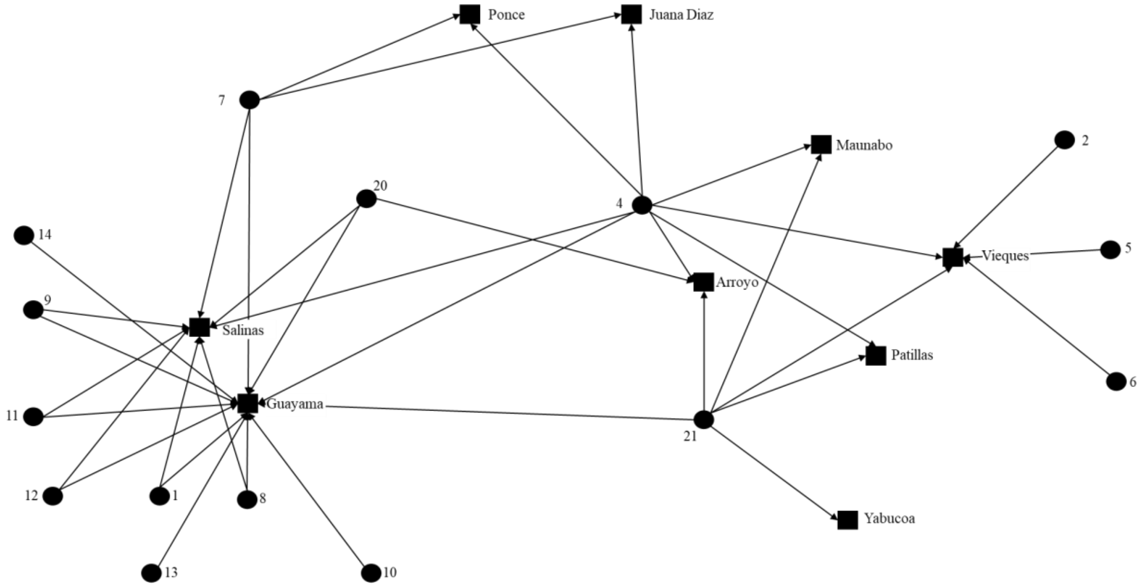

Figure 3-2 Visual social network of free-listing exercise to determine which locations were most likely to be toxic. The circular nodes represent the informant, and the square nodes represent the locations identified as hotspots.

The consensus analysis results from UCINET on the pile sort data indicated a strong agreement among the fishers on the fish most likely to be ciguatoxic. The large eigenratio (25.544) and the lack of negative competence scores indicate a good fit for the consensus model. The answer key (Table 3-1) shows fishers avoided hogfish (*Lachnolaimus maximus*), barracuda (*Sphyraena barracuda*), king mackerel (*Scomberomorus cavalla*), black jack (*Caranx lugubris*), greater amberjack (*Seriola dumerili*), and horse-eye jack (*Caranx latus*) due to CTX.

Table 3-1 Table of results from the pile sort. Fishers place marine species into two categories, avoid due to CTX or don't avoid due to CTX.

Sample ID	Fish	Avoid due to CTX?	Sample ID	Fish	Avoid due to CTX?
1	Hogfish	Yes	22	Coney	No
2	Barracuda	Yes	23	Yellowfin Grouper	No
3	King Mackeral	Yes	24	Queen Parrotfish	No
4	Cero	No	25	Rainbow Parrotfish	No
5	Black Jack	Yes	26	Stoplight Parrotfish	No
6	Amberjack	Yes	27	Striped Mojarra	No
7	Blue Runner	No	28	Yellowfin Mojarra	No
8	Horse-eye Jack	Yes	29	Sand Tilefish	No
9	Jack Crevalle	No	30	Spadefish	No
10	Cubera Snapper	No	31	Trunkfish	No
11	Queen Snapper	No	32	Redear Sardine	No
12	Silk Snapper	No	33	White Mullet	No
13	Blackfin Snapper	No	34	Ballyhoo	No
14	Lane Snapper	No	35	Blue Crab	No
15	Mutton Snapper	No	36	Queen Conch	No
16	Mangrove Snapper	No	37	West Indian Top Shell	No
17	Yellowtail Snapper	No	38	Escolar	No
18	Schoolmaster	No	39	African Pompano	No
19	Dog Snapper	No	40	Longfin Yellowtail	No
20	Tiger Grouper	No	41	Cobia	No
21	Red Hind	No			

Figure 3-3 is a visual social network analysis of the pile sort data. In this graph, the square nodes are the fishes asked about in the pile sort, and the circular nodes are the informants.

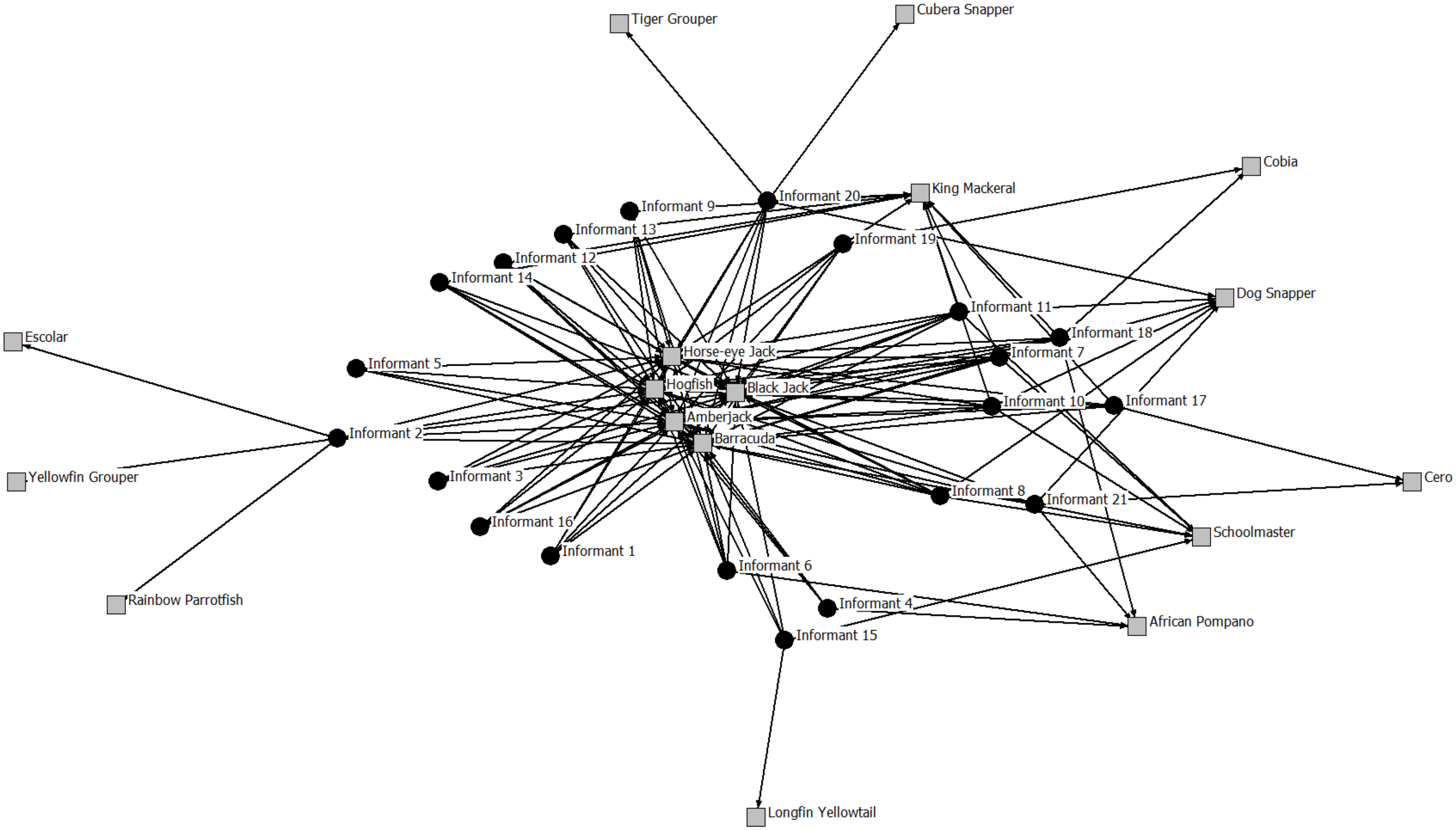

Figure 3-3 Visual social network of the pile sort data from fishers in Puerto Rico. Lines are connecting informant nodes to fish nodes to show which fish that informants avoid due to CTX.

Most informants chose the horse-eye jack, the hogfish, the black jack, the amberjack, and the barracuda as being most likely to have ciguatoxin. The fishes around the outside of the network are less chosen as problematic fish.

Discussion

Studies show that TEK is passed through formal teachings (Ruddle, 1994) and informal demonstrations and discussions (Hauzer *et al.*, 2013). The fishing information gathered was passed down to commercial fishers in Puerto Rico from their relatives and from peers, including sites and fishes, to avoid CTX (interviews with informants). Fishers along the east coast of Puerto Rico identified Guayama as a CTX hotspot (Figure 3-2), including fishers from Cabo Rojo to Guayama and the northeast coast of Fajardo. Commercial fishers use the information passed down to from older generations or by peers to alter their fishing trips and gear type used to try and avoid catching toxic fish. In most cases, when a potentially toxic fish is caught, it is discarded.

There haven't been formal ciguatera hotspot identification studies done in Puerto Rico, only in the Pacific in Hawaii. More research is done on ciguatoxin in the Pacific than in the Caribbean due to more funding allocation, and P-CTX's are 10-fold more toxic than C-CTX's (Lewis *et al.*, 1999). There are also readily available Pacific ciguatoxin standards available for purchase to run assays with (P-CTX-3C, Fujifilm Wako Chemicals), but none is available for Caribbean chemical strains. The lack of data emphasizes the importance of fishers' TEK. TEK can improve fisheries' management and can be used in data-poor artisanal fisheries to understand fishing grounds (Silvano and Valbo-Jørgensen, 2008; Pita *et al.*, 2016). Fisheries managers should utilize TEK to understand local knowledge of CTX and ecological differences between hotspots and coldspots.

Fishers across Puerto Rico generally agreed on which locations were more likely to be toxic and which fishes to avoid, which means there is some form of data transmission. Information could be passed through fish houses to fishers at other fish houses, rumors of locals getting sick and tracing that illness back to which fish house the specimen was purchased from, or passed down from elders in the communities. The spread of this information could lead to reduced cases in CFP, although insufficient data on CFP cases make this challenging to investigate. However, if fishers avoid catching certain species more likely to be toxic (higher trophic level) and fishes at higher risk areas, this could reduce the incidence rates of CFP. Casual consumers most likely do not know as much about CTX as commercial fishers. Researchers should investigate this knowledge gap. Non-commercial fishers with less knowledge may keep riskier fishes leading to CFP outbreaks.

It is difficult to determine which fish are toxic when there are no dockside tests available, potentially leading to a deterministic view of ciguatera fish poisoning (Nellis and Barnard, 1986). Nellis and Bernard (1986) show that in the USVI, when it comes to CFP, people believed they would eventually get it, and there wasn't much they could do about it. There was a similar sentiment from fishers in Puerto Rico. Their methods of avoiding toxic fish could only go so far; catching a contaminated fish was inevitable. One informant in Guayama mentioned that he had CFP multiple times. His fishing style didn't change. Partially because he couldn't change it. Those who live in the more impoverished areas cannot change their fishing locations due to economic restraints. Their smaller boats can only travel safely so far, and they don't have access to a truck and trailer to move their boat overland to fish in other areas. Besides, the fishers had their fishing grounds, and from the interviews, it was clear they did not appreciate other people fishing in their areas. Fishers may catch less risky fish, but they can still cause CFP. One fisherman mentioned that they got sick from white grunts and another, ballyhoo, which are lower trophic level fishes that we do not expect to have high toxicity levels.

During the interviews, we came across a few spearfishers that were not part of the formal talks that worked at one of the fish houses and asked them about CTX. They were adamant that certain "mushroom-shaped" algae were closely associated with high levels of CTX and would avoid spearfishing if they saw that algae on the bottom. There may be morphological differences in some algae that cause them to harbor more dinoflagellates than other algae on the reefs. Future studies should utilize a pile sort, like the one done with fishes in this study, to evaluate the fishers' knowledge of the algae on the reef and identify which algae are more likely associated with toxin-producing dinoflagellates. The algae should be sampled and compared for dinoflagellate species and cell counts.

In conclusion, we believe that fishers' TEK can predict CTX hotspots and coldspots and should be utilized by fisheries managers and collected scientific data to improve Puerto Rico's fisheries and protect the population from CFP.

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Chapter 4. CTX CONCENTRATION ESTIMATION OF CTX IN FISHES IN HOTSPOTS AND COLDSPOTS

Abstract

The consumption of fishes and other marine organisms containing high concentrations of ciguatera toxins (CTX or ciguatoxins) can cause ciguatera fish poisoning (CFP). Single-celled dinoflagellates in the *Gambierdiscus* and *Fukuyoa* genera produce precursors to the toxins. These are photosynthetic algal cells with several different species in each genus. Each species has a different growth model and different concentrations of the toxin. The toxin enters the food chain when the toxic dinoflagellates are consumed by herbivores grazing on their preferred substrates, are biomagnified up the food chain, and eventually enter top trophic predators. They can also enter the food chain through gastropods and other marine benthic invertebrates, inadvertently consuming the dinoflagellates on benthic algal substrates. If humans consume fishes with toxin concentrations of CTX, it can cause various severe illnesses and occasionally death. Hospitals have a difficult time administering treatment to patients with CFP since there is no current accepted treatment, except for fluids for dehydration. Due to rising sea temperatures that expand the algae's habitat that produce the toxin and seafood globalization, CFP cases are on the rise. The best way to prevent CFP is not to consume toxic fish. However, there is no quick or reliable dockside test for fishers to determine if the fish will cause toxicity, and cooking and adequately storing the fish does not reduce the risk of becoming ill from ciguatera.

To reduce toxic fish consumption, local fishers in regions endemic to CTX use local knowledge to avoid consuming potentially toxic species. For example, they may avoid larger fishes and species higher in trophic levels and avoid these fishes in certain areas and specific reefs. The premise of this study is to investigate the ecological knowledge that fishers can identify CTX hotspots and coldspots, places where there are high levels and low levels of toxins in fishes, respectively. Fishes of varying trophic levels were sampled from each site and tested with the N2a neuroblastoma cell-based assay, a highly sensitive assay for estimating CTX concentration in fishes, benthic invertebrates, and algae. Our data show no significant difference between the identified hotspot and coldspot among all fishes ($p = 0.09$, $n = 76$); however, the sites differ in CTX concentration at the higher trophic levels. The higher trophic level fishes had a higher concentration of CTX in the hotspot than in the coldspot, which points to biomagnification occurring at the top predator level rather than in the herbivorous fishes.

Introduction

Communities that live on island ecosystems in the Pacific and the Caribbean rely on fish for sustenance, tourism, and recreation. However, fish in tropical and subtropical regions (Lewis *et al.*, 1991) can carry ciguatera toxin (ciguatoxins, or CTX), a potent neurotoxin produced by several different species of dinoflagellates, most notably in the *Gambierdiscus* and *Fukuyoa* genera (Pottier *et al.*, 2002). Suppose humans consume fishes with high concentrations of CTX. In that case, it can cause a variety of severe symptoms, i.e., vomiting, diarrhea, abdominal pain, paresthesia (burning of the skin), the reversal of hot and cold sensations, and occasionally, death (Lehane and Lewis, 2000). The sickness from consuming ciguatoxic fish is known as ciguatoxin fish poisoning (CFP).

CTX is undetectable in fish tissues with a visual inspection. CTX is colorless, odorless, and tasteless (Copeland *et al.*, 2014) and is heat-stable; cooking the fish has no effect on the toxin (Lewis, 2000). Local folk methods for identifying toxic fish (such as feeding a small piece of fish to a pet animal and monitoring its reaction, rubbing the flesh with a coin, or leaving a portion of the fish near insects to see if they avoid it aren’t useful (Darius *et al.*, 2013). CTX dockside test strips (Cigua-Check®) were available from Oceanit but discontinued due to the inaccuracy of the tests (Bienfang *et al*., 2011). An accurate way to identify the concentration of CTX in fish tissues is the neuroblastoma cell-based assay, or N2a-cba (N2a is short for Neuro-2a, the cell line used). The N2a-cba is a proven method for estimating and screening for levels of CTX in fishes and their tissues (Pawlowiez *et al.*, 2013). The technique used was initially developed by Manger *et al*. (1993) and modified by Dickey *et al*. (2000) and Hardison *et al*. (2016).

Fishers may have adopted strategies to reduce the chance of getting sick from CFP or selling contaminated fishes to fish houses. This information could have been learned by previous generations and passed down as traditional ecological knowledge. Traditional ecological knowledge (TEK) has been increasingly useful in artisanal fisheries and data-poor regions. Fishers can provide helpful information in regards to fishing techniques (Grant and Berkes, 2007), stock assessments, spatial dynamics of fish (Mackinson, 2001; Moreno-báez *et al.*, 2010), fish behavior (Pizzini and Garcia-Quijano, 2009), and other fishing practices. In Hawaii, CTX hotspots (areas with high levels of CTX) were identified by catching and testing grouper around the island of O'ahu (Copeland *et al.*, 2014). In this study, we asked fishers which areas they believed have high and low ciguatoxin levels, designated as "hotspots" and "coldspots." The identified hotspots and coldspots were sampled for fish tissues from various trophic levels to test for CTX.

Methods

First, hotspots and coldspots were identified by commercial fishers using open-ended interviews and participatory mapping techniques. Local fish houses, or *villas pesqueras,* were visited in Puerto Rico from *Salinas* in the south to *Fajardo* in the northeast. This stretch along the coast was chosen due to several informants already working in Puerto Rico on CTX (president and members of local fishing associations in the east) who believed there were indeed hotspots and coldspots here. Targeting these areas were more efficient than randomly sampling the whole island for toxic fishes. Fishers were asked nine questions related to CFP and were allowed to elaborate on any of the topics they saw fit (See Chapter 3).

Next, we asked fishers to circle areas they identified as hotspots and coldspots on nautical charts (NOAA booklet charts 25650, 25977, and 25668). The closest municipality to the circled area's location was designated as the name of that hotspot or coldspot. For example, a circled area off the coast of Guayama was simply “Guayama.” Each fisher had a new booklet chart to draw on to discourage biased results from previous fishers. The location that most fishers agree on was the hotspot. We chose Fajardo as the coldspot as this is where a substantial amount of commercial fishing on the island takes place, and most fishers identified this area as safe from CTX. Fishers were also given a set of laminated cards with pictures of fishes to put into piles: fish they avoid catching due to ciguatoxin and fish they don’t avoid catching due to CTX. The pile sort results aided in identifying species that we targeted for CTX testing (See Chapter 3).

We sampled fishes at two reefs at two different depths in October 2019 for two consecutive days (Fajardo coldspot (Figure 4-1), and the Guayama hotspot (Figure 4-2)). Fishes of all trophic levels were targets for the study; however, hogfish (*Lachnolaimus maximus)* and barracuda (*Sphyraena barracuda)* were a high priority.

Figure 4-1 Coldspot sites sampled off the northeast coast of Puerto Rico. CTX-1 (green pin) was 25 m deep, and CTX-2 (yellow pin) was 22 m deep. Fishes were caught by locals spearfishing and regular rod and reel fishing.

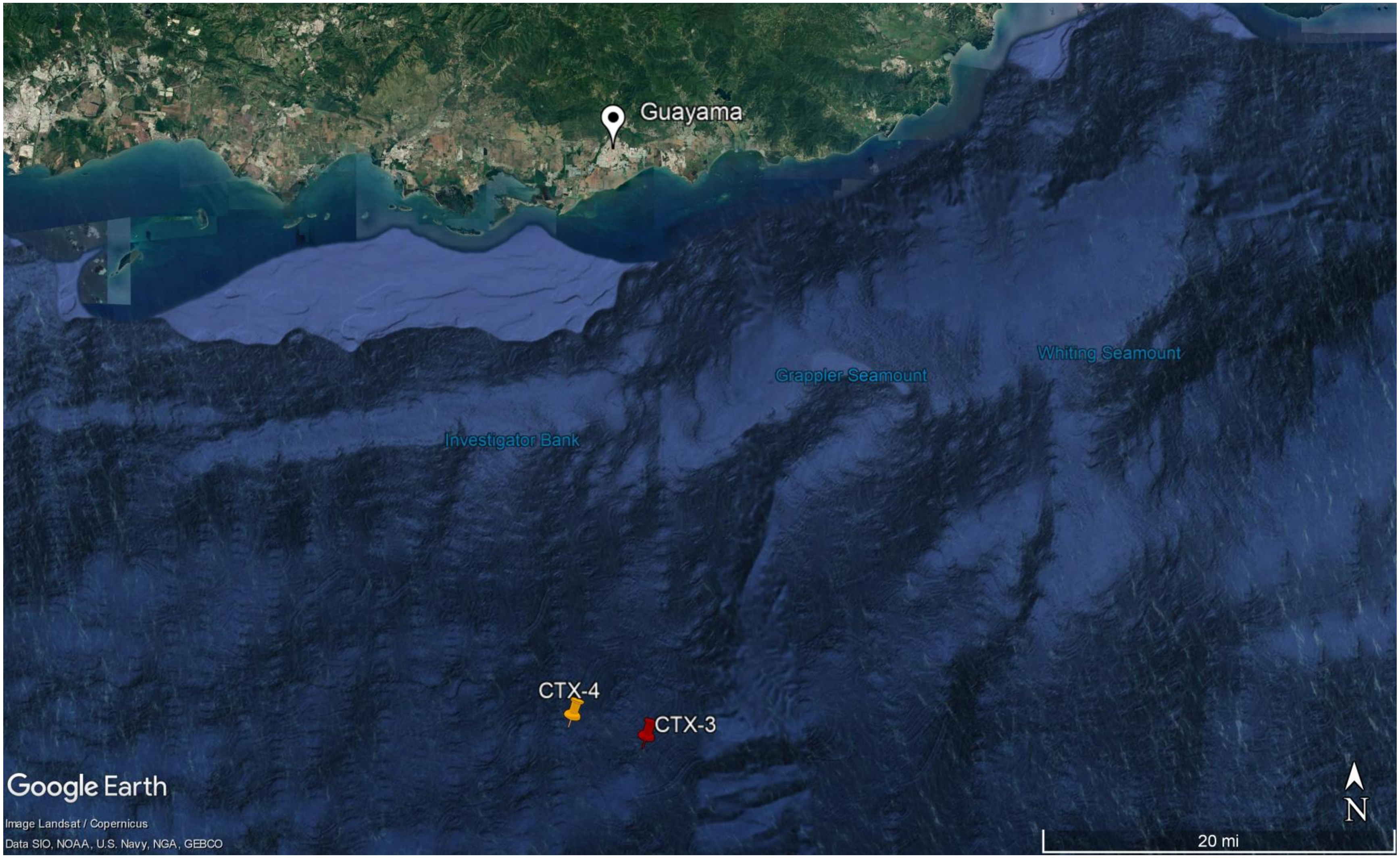

Figure 4-2 Hotspot sites located off the southeastern coast of Puerto Rico. These sites are the "Guayama hotspot." CTX-3 (red pin) was 27.4 m deep, and CTX-4 (orange pin) was 18.3 m deep. Fishes were sampled by locals using spearfishing and regular rod and reel techniques.

We targeted barracuda due to their high trophic position (~4.0) in the food web and the commercial harvesting ban of these fishes. We also targeted hogfish because of their trophic position (~3.66). They are a commercially important species to the fishers of Puerto Rico. Some informants mentioned them as highly toxic in some areas and not harmful in other areas. The fishes were captured by locals diving and by catching them with rod and reel. Once captured, the fishes were sacrificed with appropriate concentrations of MS222, weighed, and measured. Then tissue, liver, and gonads (if possible) were taken and frozen in Whirl-Pak® bags. The N2a-cba was run with these samples at East Carolina University's Brody School of Medicine Department of Toxicology and Pharmacology to estimate CTX levels in the tissues.

We prepared the samples for the N2a-cba. First, CTX was isolated from muscle tissues and suspended in 100% methanol. Five grams of fish tissue was homogenized twice in 10ml 100% methanol in a 50ml Falcon centrifuge tube using an electric tissue homogenizer. After each homogenization step, we transferred the methanol from the 50ml Falcon tube to a glass HPLC scintillation vial. It was essential to use glass vials because CTX can stick to plastics. The methanol layer was allowed to dry under an N^2 stream until only the precipitate remained. Then, 5ml dichloromethane (DCM) and 5ml 60% methanol were added to the glass scintillation vial twice.

After each substance's addition, the vial was swirled, then its contents were added to a 250ml glass separatory funnel. The layers were separated after shaking lightly, and the DCM layer was added in a new glass scintillation vial. The N^2 stream dried the sample until the precipitate remained. Next, 5ml cyclohexane and 5ml 80% methanol were added to the new glass scintillation vial, twice. After each addition, the liquid was swirled around in the vial then added to a clean 250ml separatory funnel. After being shaken lightly, the layers were allowed to separate. We collected the 80% methanol layer in a new glass scintillation vial. Finally, the methanol layer was allowed to dry under an N^2 stream completely. After reconstituting the sample in 200µl 100% methanol, the vial was fastened with a lid, secured with Parafilm, labeled, and placed in a -20ºc freezer until it was ready for the assay.

Mouse neuroblastoma cells (N2a) (ATCC, CCL131) were cultured and maintained in Eagle's Minimum Essential Media (EMEM, ATCC) with 10% fetal bovine serum (ATCC) and 5ml penicillin-streptomycin (10,000U/mL) (ThermoFisher Scientific) in a 37ºC incubator at 5% CO$_2$:95% air atmosphere. We plated the cells at 30,000 cells per well in a 96-well tissue culture plate (Fisher Scientific, 07-200-90). The cells were allowed to incubate overnight in the previously described growth medium. After 18-22 hours of incubation, the cells were treated with either plain medium or medium with Ouabain (31.3µM) and Veratradine (3.13 µM) (O/V), enough to achieve 20% cell death in positive control. Two rows of wells with O/V had the P-CTX3C serial dilution standard added, and four rows of wells (two with O/V and two without O/V) had the extracted samples added. The samples were allowed to incubate overnight.

After 18-22 hours of incubation, the medium was removed from the wells using an electric pump and suction pipette. The MTT bromide (3-[4,5-dimethylthiazole-2-yl]-2,5-diphenyltetrazolium bromide) colorimetric assay was performed, followed by an absorbance reading at 544nm for each well. First, we added 1ml MTT to the 5ml growth medium and then the MTT mixture to each well in 50µl aliquots. The full 96-well plate setup can be found in Appendix F. The cells were left to incubate for 30-60mins until a purplish color appear. MTT is catalyzed to MTT-formazan by mitochondrial succinate dehydrogenase, which creates a dark purple color. The more metabolically-active cells in a well, the darker the color, and therefore the higher the absorbance when measured by a spectrophotometer. After reaching the time limit, we removed the MTT via the “flick” method and added 100µl of dimethyl sulfoxide (DMSO) to each well. DMSO acts as a lysing agent to the cells that release the color from the cells' inside. The plate was put on an orbital shaker to distribute the coloring for 15 minutes evenly and read at an absorbance at 544nm.

After reading the wells' absorbance, we imported the data into a spreadsheet created by the NOAA Center for Coastal Fisheries and Habitat Research Lab, Beaufort, NC. The spreadsheet allows for the visual representation of the P-CTX-3C standard curve and computing variables within the data to achieve a concentration from absorbance numbers. To complete the worksheet and get final ppb estimates, we plotted the standard curve from the plate with the x-axis as the Log of the agonist concentration (P-CTX-3C) and the y-axis as the response (the absorbances of the standard curve from the plate). The data were fit as a nonlinear regression curve using the sigmoidal, 4PL, X is log(concentration) equation in GraphPad Prism®. The mean absorbances of the fish sample wells interpolated the ppb in CTX-3C equiv. from the standard curve using a 95% confidence interval. The standard curve determined the EC50 of the agonist, which is the halfway response between the bottom and top of the curve. After running this analysis, GraphPad Prism® calculated the parameters: top, bottom, EC50, R^2, and HillSlope, the steepness of the family of curves. These values, along with the interpolated X values, were added to the worksheet and final ppb; in P-CTX-3C, equivalents were given for samples.

Results

We estimated the concentration of CTX (in CTX3C equiv.) in fishes captured in identified hotspots and coldspots in Puerto Rico. Overall, the fishes in the hotspot had a higher concentration of CTX3C equiv. than the fishes in the coldspot (Welch’s two-sample t-test, $p = 0.0331$, $n = 44$).

Figure 4-3 shows the differences in CTX3C equiv. between different fish species at varying trophic levels. At the hotspot, *Sphyraena barracuda* had a much higher median CTX3C equiv. concentration in its tissues than the *Sphyraena barracuda* in the coldspot. The median value of CTX3C equiv. in the hotspot is close to the 0.1 ppb recommended advisory consumption levels set by the FDA. There is little difference between the herbivorous fish's median values. *Sparisoma viride* had a slightly higher median CTX3C equiv. concentration in the coldspot fishes than the hotspot. The molluscivore, *Lachnolaimus maximus,* had higher median concentrations of toxin concentration than those in the coldspot.

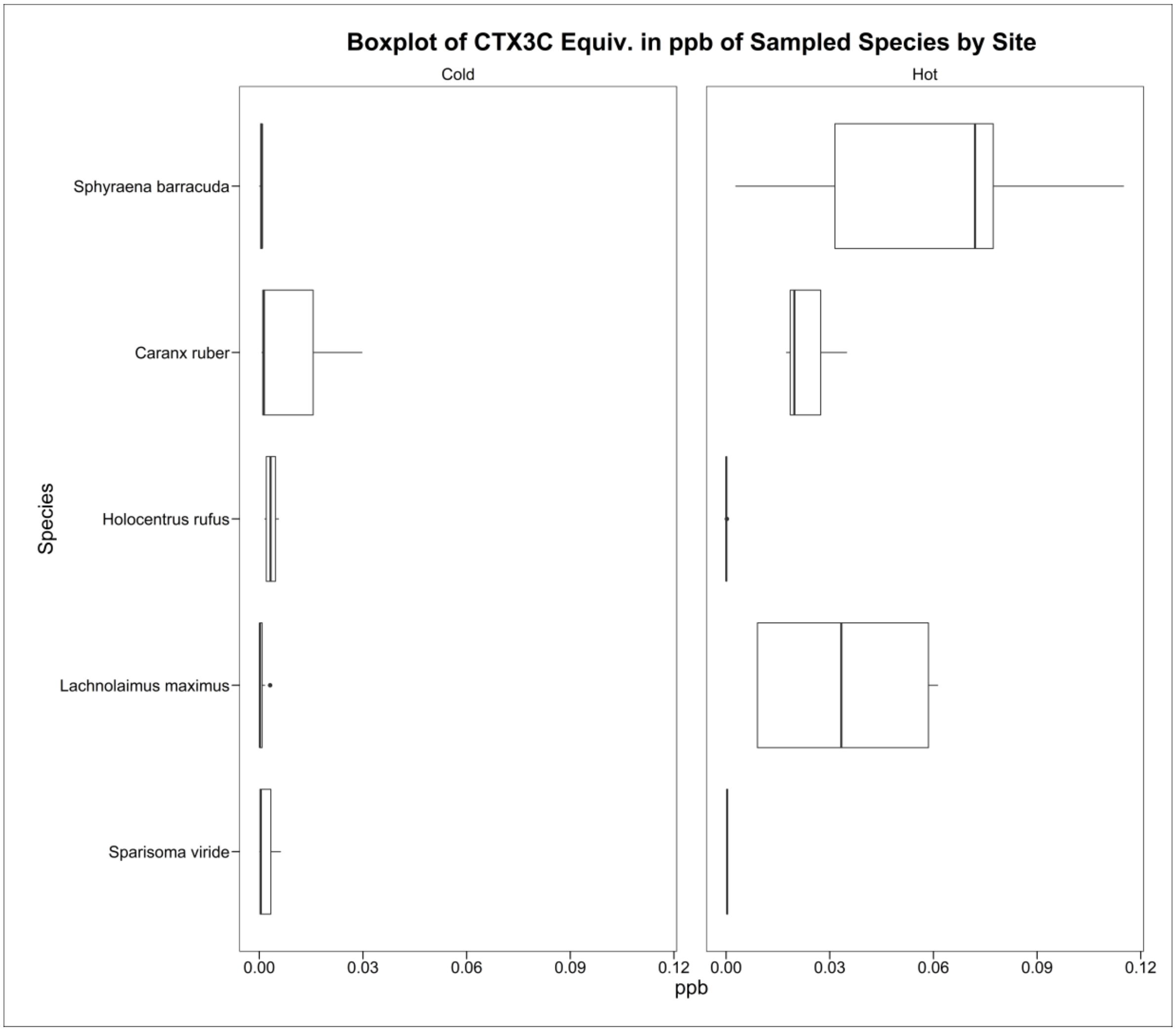

Figure 4-3 Boxplot of median CTX3C equiv. concentrations in ppb by species in the hotspot and coldspot. The top trophic predators had a higher median CTX3C equiv. concentration in the hotspot compared to the coldspot. Species are listed from highest ETL to lowest.

Like the *Sparisoma* viride, *Holocentrus rufus* had similar CTX3C equiv. concentration in with the coldspot fish being slightly higher. Lastly, the median CTX3C equiv. of *Caranx ruber* was higher in the hotspot than it was in the coldspot.

A two-way interaction ANOVA was conducted to compare the effects of spot, trophic group, and the spot-trophic group interaction on toxin concentration in fishes. There was a significant effect of hotspot/coldspot site on toxin concentration in fishes (F = 6.359, df = 1, p = 0.016) as well as an effect of trophic group on toxin concentration in fishes, (F = 5.078, df = 2, p = 0.0111). The hotspot/coldspot site interaction with trophic group did not show a significant effect on toxin concentration in fishes (F = 2.54, df = 2, p = 0.0922).

I used a Tukey HSD post-hoc comparison to identify which trophic groups differed in toxin concentration. The low trophic group had 5.01 x 10^{-5} ppb less toxin than the high trophic group (p = 0.029), while the medium trophic group was 1.24 x 10^{-4} ppb less toxic than the high

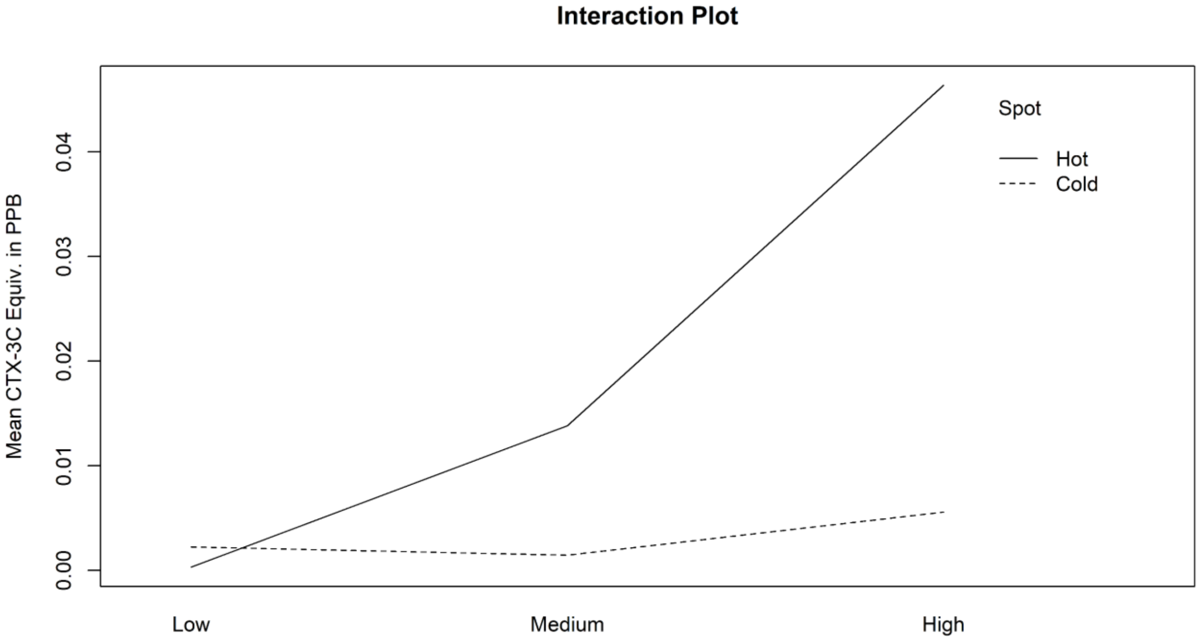

Figure 4-4 Interaction plot of CTX3C equiv. concentrations in three trophic level groups (low, medium, and high) between the hotspot and coldspot.

trophic group (p = 0.027). There was no significant difference between low and medium trophic groups.

Although the interaction between spot and group was not significant, the p-value was close enough to 0.05 to warrant examining the site by group comparisons using the Tukey HSD post-hoc comparison (p = .092). The high trophic level fishes at the hotspot were .04 ppb higher than the high trophic level fishes at the coldspot (p = 0.053). The low and medium trophic groups were not significantly different in the same sites or between hotspot and coldspot. However, looking at the interaction plot, the CTX3C equiv. concentration rises with trophic level and is higher in the hotspot than in the coldspot (Figure 4-4). The hotspot and coldspot have similar CTX3C equiv. levels at the low trophic level groups (near 0ppb) and keep diverging through the medium trophic groups to the high trophic level predators.

Discussion

Overall, fishes in the hotspot had higher levels of CTX in their tissues than the fishes in the coldspot, which supports our hypothesis that fishers can identify CTX hotspots and coldspots. It is difficult to pinpoint what factors drive this difference, but some could be attributed to the higher cell densities in the hotspot than the coldspot (See Chapter 5). The coldspot in Puerto Rico is on the north side of the island, and a study by Loeffler *et al*. (2018) shows lower toxicity in fishes collected on the north side of the U.S. Virgin Islands. The scientists in this study show greater wave energy on the north side of the USVI, leading to a more deficient growing environment for CTX-producing dinoflagellates.

The fishes that differed from the hotspot and coldspot were the *Sphyraena barracuda* (barracuda), *Lachnolaimus maximus* (hogfish), and *Caranx ruber* (bar jack). These species are all higher trophic level organisms compared to the other species compared. The barracuda consumes mostly fishes with some octopuses and crustaceans, similar to the bar jack, while the hogfish primarily consume mollusks (Randall, 1967). Interestingly, hogfish have higher levels of CTX3C equiv. in their tissues than the bar jack when the bar jack is at a higher trophic level. The metabolism of CTX by fishes may explain this phenomenon (See Chapter 2 Introduction). When secondary consumers feed on the CTX-producing dinoflagellates, they metabolize the toxin and excrete 95% in the form of oxocenes, which drastically reduces the amount of CTX that gets transferred to the next trophic levels (Ledreux *et al.*, 2014). However, the same metabolism is most likely not present in gastropods, the hogfishes' preferred prey. Suppose gastropods consume toxin-producing dinoflagellates while grazing on their preferred substrates and are not metabolizing it like fishes. In that case, they could be transferring more CTX to higher trophic levels than if it had gone through herbivorous fishes. This CTX transfer could explain the higher levels of CTX3C equiv. in hogfish compared to the bar jack. Future studies should test the CTX3C equiv. concentration in gastropods and secondary consumers in the same locations and compare that to the dinoflagellate density and species composition on the same reef. This study may begin to explain how the pathways that CTX takes through the food web play a role in the toxicity of some species.

The interaction plot (Figure 4-4) shows an increase in the hotspot's toxicity as the trophic level increases. In the coldspot, the toxicity is similar at the low and medium trophic levels but increases in the high trophic level; biomagnification happens with CTX in the food web. Top trophic levels consume fishes that have stored CTX in their tissues, while the low and medium trophic level fishes are consuming CTX from the dinoflagellates directly. Along with metabolism and excretion, the low and medium trophic level fish consume the dinoflagellates that produce the toxin. The toxin load in these cells is much smaller than a whole fish accumulating toxin as it grazes.

Future studies should sample fishes, perform gut-content analyses on these fishes, and test the prey in their stomachs for CTX3C equiv. The guy content analysis would allow scientists to understand better the pathways that CTX takes in the food web. If a fish's prey has a high level of CTX, then that is likely a pathway that CTX travels to increase toxicity in species.

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Chapter 5. DINOFLAGELLATES AT HOTSPOTS AND COLDSPOTS

Abstract

Dinoflagellates in the *Gambierdiscus* and *Fukuyoa* genera produce ciguatoxins (CTXs), potent neurotoxins that cause ciguatera fish poisoning (CFP) if consumed in high enough concentrations by humans. Herbivores and herbivorous fish inadvertently consumed these dinoflagellates feeding on their preferred substrates, which leads to biomagnification to the toxin in coral reef food webs. It is not clear why some reefs or fishing grounds become toxic and why others do not. One hypothesis is that large blooms of these toxin-producing dinoflagellates and higher numbers of these cells lead to higher CTX uptake rates via the food web. We show here that CTX hotspots identified by fishers had higher cell counts of *Gambierdiscus* spp. than the CTX coldspots identified by fishers. The CTX hotspot had 11,666 cells $L^{-1,}$ while the coldspot had only 333 cells L^{-1} of *Gambierdiscus* spp. This 35-fold increase in cell density at the hotspot could explain the higher toxin concentrations seen in higher trophic level fishes in Chapter 4.

Introduction

Communities that live on island ecosystems in the Pacific and the Caribbean rely on fish for sustenance, tourism, and recreation. However, fish in tropical and subtropical regions (Lewis *et al.*, 1991) can carry ciguatera toxin (ciguatoxins, or CTX), a potent neurotoxin produced by several different species of dinoflagellates, most notably in the *Gambierdiscus* and *Fukuyoa* genera (Pottier *et al.*, 2002). Suppose humans consume fishes with high concentrations of CTX. In that case, it can cause a variety of severe symptoms, i.e., vomiting, diarrhea, abdominal pain, paresthesia (burning of the skin), the reversal of hot and cold sensations, and occasionally, death (Lehane and Lewis, 2000). The sickness from consuming ciguatoxic fish is known as ciguatoxin fish poisoning (CFP).

Ciguatoxin enters the food web when gastropods or herbivorous fishes feed on their preferred substrates and indirectly consume the dinoflagellates (Randall, 1958b; Lewis, 2001; Ledreux *et al.*, 2014). The dinoflagellates migrate up and down in the water column to respond to light conditions, salinity, temperature, and nutrient availability, eventually settling on benthic autotrophs (Kamykowski, 1981; MacIntyre *et al.*, 1997). These dinoflagellates are closely associated with a wide variety of macroalgae. *Gambierdiscus toxicus* (a species of dinoflagellate that produces the precursor to CTX) is an epiphyte of macroalgae (Holmes *et al.*, 1991). The number of cells g^{-1} wet weight (ww) algae can range from just a few cells to over 100,000 cells g^{-1} ww algae (Litaker *et al.*, 2010). The most frequently observed abundances of cells on benthic algae are from 0-1000 cells g^{-1} ww algae (>85%) with less than 10% of observations in the 1000-100,000 cells g^{-1} ww algae range (Litaker *et al.*, 2010).

Each alga in the *Gambierdiscus* spp. genera have different growth rates and toxin concentrations, with the slower-growing species generally producing more toxin than the faster-growing species (Kibler *et al.*, 2012). Caribbean species' toxicity can range from 0.27 ± 0.43 fg CTX3C equiv. $cell^{-1}$ in *Gambierdiscus carolinianus* to 19.6 fg CTX3C equiv. $cell^{-1}$ in *Gambierdiscus silvae* and can reach as high as 469 fg CTX3C equiv. $cell^{-1}$ in *Gambierdiscus excentricus* (Kibler *et al.*, 2012). The extensive range in the species' toxicities is an issue when trying to understand optimal conditions for large and small-scale CFP events. Small blooms of highly toxic cells might be enough to cause fishes to cross the 0.1 ppb, and 0.01 ppb recommended advisory consumption levels for Caribbean and Pacific chemical congeners, respectively, set by the FDA (Vernoux and Lewis, 1997; Lehane and Lewis, 2000; Dickey and Plakas, 2010).

This study added *Gambierdiscus* species for several areas in Puerto Rico and the overall abundance of these dinoflagellates at these sites to the current knowledge base. Local fishers identified regions of likely high and low ciguatoxicity (hotspot and coldspot), which we sampled for dinoflagellates. We identified four species at three different reefs with cell abundances ranging from 333.33 cells L^{-1} in the coldspot to 11,666.67 cells L^{-1} in the hotspot.

Methods

We sampled dinoflagellates at sites CTX-1 (25m), CTX-2 (22m), CTX-3 (27.4m), and CTX-4 (18.2m) in October 2019 over two consecutive sampling days. During the first day, the screen sampling rigs were set (Figure 5-1). We deployed five repeat rigs on the bottom in a line 10m apart for each site. The rigs were a simple weight attached to a fishing bobber with a barrel swivel attached 1m from the weight and a mesh screen attached to the swivel. After 24 hours, divers collected the rigs by placing a jar over the screens and unhooking the swivels; the lids were tightened on the jars and brought to the surface. The samples were taken to the University of Puerto Rico at Humacao and preserved with Lugol's solution. The water samples and screen filters were transferred to brown plastic bottles, with 20% of the water volume poured off to create headspace for shaking. The bottles were shaken vigorously for 15-20 seconds to free the dinoflagellates from the screen. The sample was immediately poured through a 200µm mesh sieve to remove larger particles

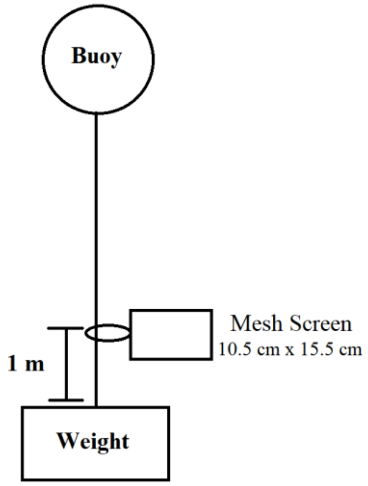

Figure 5-1 **Screen sampler rig for toxic dinoflagellates. Five screens were deployed at each site and were allowed to soak for 24 hours. After 24 hours the screen was collected. A glass jar was placed over the screen, and the swivel was detached from the line.**

into another brown bottle. Four drops of Lugol's solution were added to the water and gently mixed to preserve the specimens. The water samples were stored in brown plastic bottles and brought back to ECU, then transferred to the NOAA Southeast Fisheries Science Center. They were counted for the number of *Gambierdiscus* spp. cells and to identify which species were present using qPCR.

Results

Figure 5-2 ***Gambierdisus*** **spp. identified from the CTX-4 hotspot site.** ***Photo courtesy of Sylvia M. Velez-Villamil, M.S., Universidad de Puerto Rico en Humacao, 2019.***

The samples show that there were *Gambierdiscus* spp. present (Figure 5-2). The repeats were combined by site (CTX-1, CTX-2, CTX-3, CTX-4) and analyzed for differences. The number of *Gambierdiscus spp.* cells L^{-1}in the hotspots were higher than in the coldspots (Figure 5-3). The median values in coldspot CTX-1 and coldspot CTX-2 were 333.33 cells L^{-1} and 1000 cells L^{-1}, respectively. The hotspots' median values were higher at 2333.33 cells L^{-1} at CTX-3 and 11,666.67 cells L^{-1} at CTX-4. The short boxes in sites CTX-1, CTX-2, and CTX-3 show a high agreement among the replicate samples, while CTX-4 suggests more considerable differences in the repeats. The lower whisker in the CTX-3 plot site overlaps the first quartile in the CTX-2 site plot. These data show that there are some similar cell counts in CTX-3 and CTX-2. The CTX-4 site had many more cells L^{-1} than any other sites as no parts of the boxplot overlap any of the other sites' plots.

I then combined the samples from CTX-1 and CTX-2 into one "coldspot" and the samples from CTX-3 and CTX-4 into one "hotspot" for a Welch's two-sample independent t-test due to the two sites having unequal variances and is robust for skewed distributions. The sites differed significantly ($t = -3.8705$, $p=0.003714$) with a mean of 633 *Gambierdiscus* spp. cells L^{-1} in the coldspot and a mean 7500 *Gambierdiscus* spp. cells L^{-1} in the hotspot.

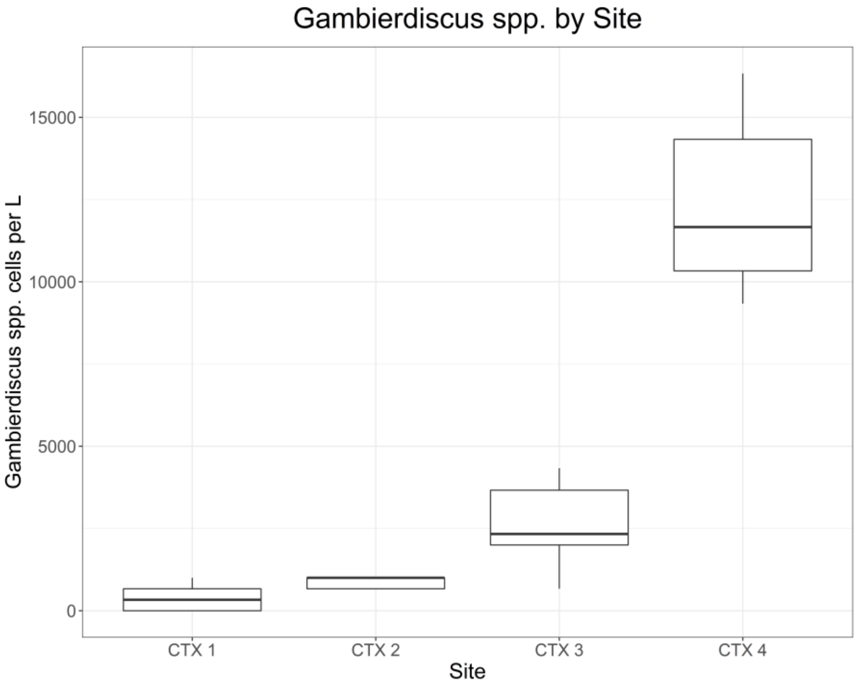

Figure 5-3 ***Gambierdiscus*** **spp. cells $L^{-1}$for the coldspots (CTX-1 and CTX-2) and the hotspots (CTX-3 and CTX-4). The experts at the NOAA Southeast Fisheries Laboratory (Beaufort, NC) counted the cells and confirmed the cells are in the** ***Gambierdiscus*** **genera.**

The NOAA Southeast Fisheries Laboratory ran a qPCR on the water samples to identify the species present at each site. We identified several species across all sites. *Gambierdiscus caribaeus* was present at CTX-1, CTX-3, and CTX-4, *Gambierdiscus carpenteri* was present at CTX-3, *Gambierdiscus belizeanus* was present at CTX-1, and *Gambierdiscus carolinianus* was present at CTX-3 and CTX-4 (Figure 5-3). Unfortunately, the extended time between the manual cell counts and the qPCR caused some DNA degradation, which caused low cell equiv. counts with the qPCR, and also no species were identified in the CTX-2 coldspot samples.

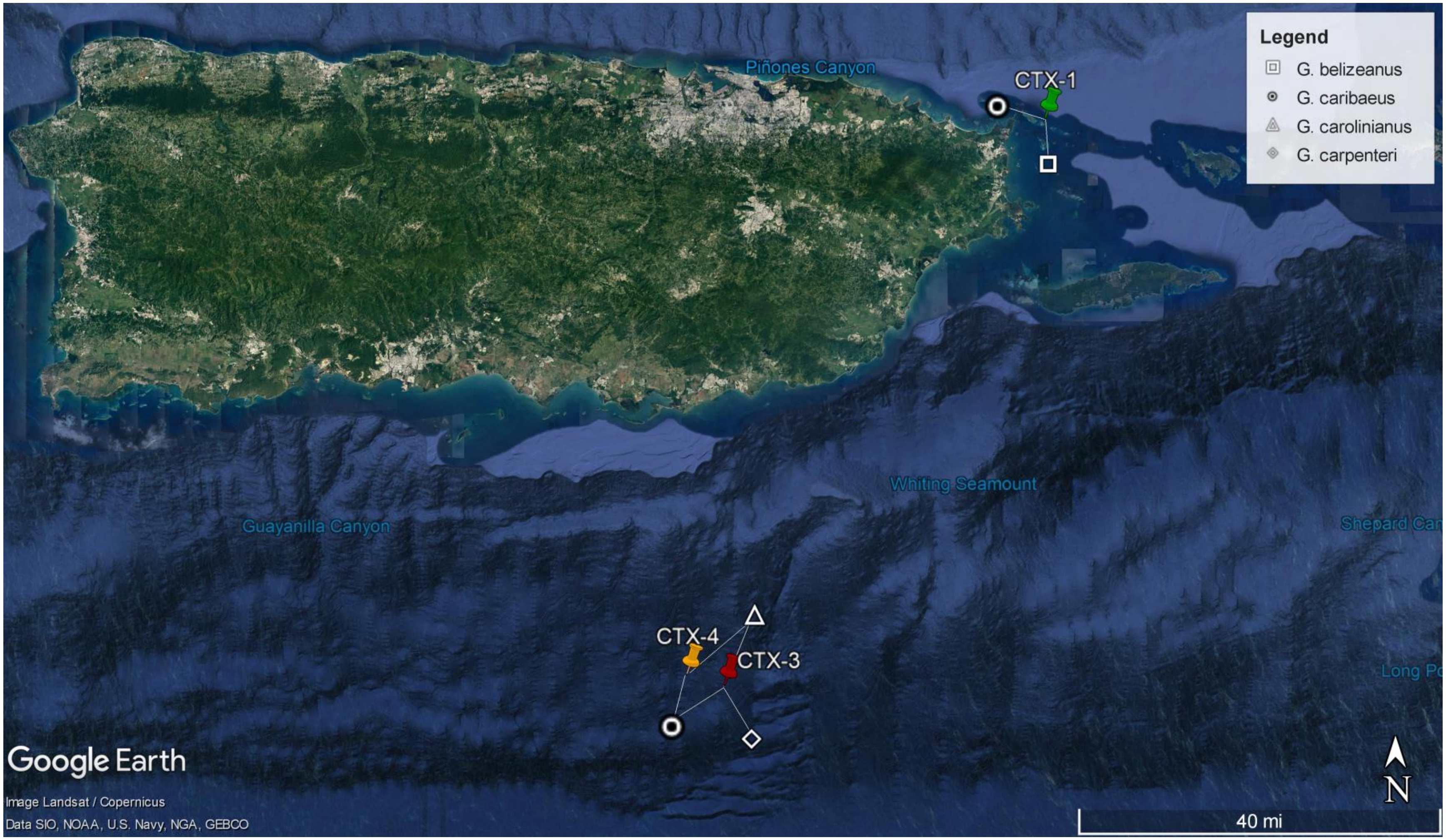

Figure 5-4 Map of the sampled sites and the *Gambierdiscus* spp. identified at those sites. CTX-1 (green pin) was 25m deep, CTX-3 (red pin) was 27.4m deep, and CTX-4 (orange pin) was 18.2m deep. CTX-2 is not listed because there were no species identified in that site due to the samples' degraded DNA.

Discussion

Overall, the data shows higher cell counts in the hotspot samples than in the coldspot samples. The higher number of cells L^{-1} could be causing toxicity in higher trophic level fishes at those sites (see Chapter 4). Since herbivores and herbivorous fish consume these dinoflagellates when feeding on their preferred substrates, any increase in the number of cells resting on the algae would increase the amount of toxin entering the system (Randall, 1958; Lewis, 2001; Ledreux *et al.*, 2014).

The suite of species found was different at each site, with *G. caribaeus* being the only species found at both hotspot and coldspot. Litaker *et al*. 2017 describe each species' toxin concentration that we found at the hotspot and coldspot: the toxin concentration of *Gambierdiscus caribaeus* is 0.66 ± 0.34 fg CTX3C equiv. cell^{-1}, *Gambierdiscus carpenteri* is 0.89 ± 0.41 fg CTX3C equiv. cell^{-1} *Gambierdiscus belizeanus* is 0.85 ± 0.81 fg CTX3C equiv. cell^{-1} and *Gambierdiscus carolinianus* is 0.27 ± 0.43 fg CTX3C equiv. cell^{-1}. Assuming an equal distribution of cells, although unlikely, the average toxicity of the cells at the coldspot (0.8 fg CTX3C equiv. cell^{-1}) is almost twice as high as the cell toxicity at the hotspot (0.465 fg CTX3C equiv. cell^{-1}). Since this is counterintuitive to what we predicted, there may be more toxic species cells than low toxic species in the hotspot. Future studies should include more in-depth dinoflagellate sampling protocols, including doing the qPCR right after the cells are captured instead of waiting (the qPCR was delayed due to the global SARS-CoV-2 pandemic, and therefore, some DNA was degraded).

Interestingly, the species composition at CTX-3 and CTX-4 was similar; both consisted of *Gambierdiscus caribaeus* and *Gambierdiscus carolinianus*. However, CTX-3 had

Gambierdiscus carpenteri, and CTX-4 did not, which could be due to the wide range of the optimum light tolerance of *Gambierdiscus carpenteri* (55-388 µmol photons m^{-2} s^{-1}) (Kibler *et al.*, 2012). The depth of these two sites may contribute to this species being present in one over the other. The CTX-3 site is roughly 9m deeper than the CTX-4 site.

If the number of cells of these toxin-producing dinoflagellates drives the toxicity in high trophic level fishes, scientists will benefit from a routine monitoring program of the algae. Divers should collect *Gambierdiscus* spp. using the screen-sampler method, count the number of cells, and identify the species present using PCR, which would also help fill the large data gap in these cells' global distribution (Litaker *et al.*, 2010). We generally know which species habituate the Pacific and Indian Oceans and the Caribbean Sea. However, scientists know little about the specific reefs and coasts to which these dinoflagellates thrive. There is some evidence that increased wave and wind action reduces the toxicity of reefs (Loeffler *et al.*, 2018); northern coasts of the Caribbean Islands experience harsher conditions, disturbing the growth of these algae. Studies should sample along the north and south coasts of Puerto Rico and compare the dinoflagellate profiles to the wind and wave energy exerted on these areas.

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Chapter 6. MODEL PARAMETERIZATION

We used the following methods to parameterize the CTX hotspot and coldspot food web models. ECOPATH with ECOSIM requires several parameters for simulation, including several on the “basic input” tab and a diet composition predator/prey matrix for each compartment. The three parameters within the basic input page are biomass per habitat area in g m^{-2}, the production/biomass/year (*P/B*) of the compartment, which can be expressed by $Z = M + F$, where Z is the total mortality, M is the natural mortality, and F is the fishing mortality and the consumption/biomass/year (*Q/B*) which can be calculated holistically by the sum of several different characteristics of the species in the compartment (ECOPATH developer site).

We calculated biomass in habitat per area in g m^{-2} by multiplying the total number of fishes identified or sampled in each hotspot and coldspot by the species' mean mass. The total biomass per species was divided by the area sampled.

For each compartment, natural mortality can be used for the production/biomass ratio when fishing mortality data is absent in the form:

$$M = K^{0.65} * L_{\infty}^{-0.279} * T_c^{0.463} \qquad \text{Eq. 1}$$

where *M* is the natural mortality (/year), *K* is the curvature parameter of the von Bertalanffy growth function (/year), L_∞ is the asymptotic length (total length, cm), and T_c is the mean habitat (water) temperature, in C°, or total mortality, if fishing mortality data is present, for the compartment as shown:

$$Z = \frac{P}{B} = \frac{K*(L_\infty - \overline{L})}{\bar{L} - L'} \qquad \text{Eq. 2}$$

where Z is the total mortality, L_∞ is the asymptotic length, K is the von Bertalanffy growth function curvature parameter, $\bar{L}$ is the mean length in the population, and L’ is the mean length at entry into the fishery assuming knife-edge selection.

The data for consumption to biomass ratio for each species came from the Opitz (1996) Caribbean coral reef food web. Opitz (1996) calculated food consumption per unit biomass (Q/B) using the empirical model of the form:

$$\log_{10}\frac{\mathrm{Q}}{\mathrm{B}} = -0.0771 - 0.2018\log_{10}W_\infty + 0.612\log_{10}T + 0.515\log_{10}A + 0.5471F \qquad \text{Eq. 3}$$

(Palomares and Pauly 1989) where Q/B is the food consumption of an age-structured population over its mean biomass, W_∞ is the mean asymptotic (or maximum) weight (as defined by the vBGF) of the fish of a given population, T is the mean environmental temperature in C °, A is an index of the mean activity level of the fish of a given species derived from the aspect ratio of its caudal fin and F is the food type, with carnivorous = 0 and herbivorous = 1 (Opitz, 1996). The aspect ratio of the caudal fin (A) is:

$$A = \frac{h^2}{s} \qquad \text{Eq. 4}$$

where h is the height of the caudal fin and s is the surface area.

We enlisted locals' help to dive, count, and sample fishes along the reef in Puerto Rico to parameterize a food web model for the hotspot and coldspot areas. The sampling method consisted of swimming in an “L” shape and identifying the species and quantity of fishes of those species seen on the reef. Each fish species had its parameters calculated for the basic input and diet matrix, with some species being grouped into compartments using hierarchical cluster analysis. The number of fishes counted was used to calculate the biomass in habitat area in g m^{-2}

for ECOPATH with ECOSIM's basic input. If there were more than a few species, then the habitat area's biomass was calculated for the model. If fewer than five fishes were caught, then the average biomass in habitat area in g/m^2 was used from the Opitz (1996) model. Some fishes were not identified while diving or sampling but are most likely present along the reef. Opitz's 1996 food web model was used to supplement the species missing in our models. For the fish compartments created from our sampling, the biomass and *P/B* values were removed from its respective Opitz compartment to avoid repeating the same species' measurements. We added the new Opitz compartment to our model with sampled fishes' data removed. For example, the Opitz compartment "Small Scaridae" contains *Scarus iseri*, *Sparisoma Aurofrenatum*, and *Sparisoma radians*. We sampled or observed both *Scarus iseri* and *Sparisoma aurofrenatum* in the hotspot in Puerto Rico. The biomass in g/m^2 was calculated for each species, and that value was removed from the biomass of the Opitz compartment 25 "Small Scaridae." A new compartment for "Small Scaridae" from the original Opitz model was added to our model with the updated biomass estimate, which allowed us to include all of the critical species of a coral reef, parameterizing it for each site monitored.

We used the *P/B* values for individual species from the Opitz (1996) model. Opitz used natural mortality (*M*) for all species. If individual species values were not available for the *P/B*, then the value from the compartment in which that species was grouped from the Opitz model was added. All *Q/B* values were used from the individual species from the Opitz (1996) model. If we sampled fishes and removed them from the Opitz (1996) compartments, their P/B and Q/B were removed from their respective Opitz compartments. We calculated a new median value for the *P/B* and *Q/B* for the new Opitz's compartments with the remaining species in the compartments.

Calculating Sampled Species Basic Input Values for Compartments (Hotspot)

These values are represented in Table 6-1.

HOTSPOT

1. Sergeant Major (*Abudefduf saxatilis*)

Fishes counted while diving	8
Fishes sampled (fishing/diving)	0
Total	8

Biomass g/m^2 Over 4 replicate site sampling days, 8 fish were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 2. The average mass of the sergeant major was used from the Opitz 1996 model (125g).

Then, 2 was multiplied by the mean mass 125g for the biomass, 250g. 250g divided by the area of the hotspot (3600m^2) = **0.069444 g/m^2**

Production/biomass/year 1.265 P/B (compartment 8 value) (Opitz, 1996)

Consumption/biomass/year 13.28 Q/B (individual species value) (Opitz, 1996)

2. Ocean Surgeonfish (*Acanthurus bahianus*)

Fishes counted while diving	11
Fishes sampled (fishing/diving)	0
Total	11

Biomass g/m^2 Over 4 replicate site sampling days, 11 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 2.75. The mean mass of the ocean surgeonfish was used from the Opitz 1996 model (74 g).

Then, 2.75 was multiplied by 74g for the biomass, 203.5g. 203.5g divided by the area of the hotspot (3600m^2) = **0.0565277778 g/m^2**

Production/biomass/year 1.0 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 34.38 Q/B (individual species value) (Opitz, 1996)

3. Doctorfish (*Acanthurus chirurgus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the doctorfish was used from the Opitz 1996 model (212g).

Then, 0.25 was multiplied by 212g for the biomass, 53g. 53g divided by the area of the hotspot (3600m^2) = **0.014722222g/m^2**

Production/biomass/year 0.71 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 24.7 Q/B (individual species value) (Opitz, 1996)

4. Blue Tang (*Acanthurus coeruleus*)

Fishes counted while diving	10
Fishes sampled (fishing/diving)	0
Total	10

Biomass g/m^2 Over 4 replicate site sampling days, 10 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 2.5. The average mass of the blue tang was used from the Opitz 1996 model (254g).

Then, 2.5 was multiplied by 254g for the biomass, 635g. 635g divided by the area of the hotspot (3600m^2) = **0.17388889 g/m^2**

Production/biomass/year 0.7 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 24.4 Q/B (individual species value) (Opitz, 1996)

5. Scrawled Filefish (*Aluterus scriptus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The scrawled filefish' average mass was used from the Opitz 1996 model (759g) since we only sampled one fish.

Then, 0.25 was multiplied by 759g for the biomass, 189.75g. 189.75g divided by the area of the hotspot (3600m^2) = **0.0527083333 g/m^2**

Production/biomass/year 0.55 P/B (compartment 19) (S. Opitz, 1996)

Consumption/biomass/year 6.8 Q/B (individual species value) (S. Opitz, 1996)

6. Porkfish (*Anisotremus virginicus*)

Fishes counted while diving	6
Fishes sampled (fishing/diving)	0
Total	0

Biomass g/m^2 Over 4 replicate site sampling days, 6 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.5. The average mass of the porkfish was used from the Opitz 1996 model (219g).

Then, 1.5 was multiplied by 219g for the biomass, 328.5g. 328.5g divided by the area of the hotspot (3600m^2) = **0.09125 g/m^2**

Production/biomass/year 1.265 P/B (compartment 8) (Opitz, 1996)

Consumption/biomass/year 10.3 Q/B (individual species value) (Opitz, 1996)

7. Queen Triggerfish (*Balistes vetula*)

Fishes counted while diving	7
Fishes sampled (fishing/diving)	2
Total	9

Biomass g/m^2 Over 4 replicate site sampling days, 9 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 2.25. The average mass of the queen triggerfish was calculated from sampled fishes (886.8g).

Then, 2.25 was multiplied by 886.8g for the biomass, 1995.3g. 1995.3g divided by the area of the hotspot (3600m^2) = **0.55425 g/m^2**

Production/biomass/year 0.56 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 6.9 Q/B (individual species value) (Opitz, 1996)

8. Spanish Hogfish (*Bodianus rufus*)

Fishes counted while diving	7
Fishes sampled (fishing/diving)	0
Total	0

Biomass g/m^2 Over 4 replicate site sampling days, 7 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.75. The average mass of the spanish hogfish was used from the Opitz 1996 model (402g).

Then, 1.75 was multiplied by 402g for the biomass, 703.5g. g divided by the area of the hotspot (3600m^2) = **0.195416667g/m^2**

Production/biomass/year 0.64 P/B (compartment 13) (Opitz, 1996)

Consumption/biomass/year 5.9 Q/B (individual species value) (Opitz, 1996)

9. Pluma Porgy (*Calamus pennatula*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the pluma porgy was used from the Opitz 1996 model (439g).

Then, 0.25 was multiplied by 439g for the biomass, 109.75g. 109.75g divided by the area of the hotspot (3600m^2) = **0.030486111 g/m^2**

Production/biomass/year 1.265 P/B (compartment 11) (S. Opitz, 1996)

Consumption/biomass/year 9.3 Q/B (individual species value) (S. Opitz, 1996)

10. Black Jack (*Caranx lugubris*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	1
Total	2

Biomass g/m^2 Over 4 replicate site sampling days, 2 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.5. The black jack's average mass was used from the Opitz 1996 model (1691g) since we only sampled one fish.

Then, 0.5 was multiplied by 1691g for the biomass, 845.5g. 845.5g divided by the area of the hotspot (3600m^2) = **0.234861111g/m^2**

Production/biomass/year 1.17 P/B (compartment 4 data) (Opitz, 1996)

Consumption/biomass/year 9.6 Q/B (individual species value) (Opitz, 1996)

11. Bar Jack (*Caranx ruber*)

Fishes counted while diving	5
Fishes sampled (fishing/diving)	1
Total	6

Biomass g/m^2 Over 4 replicate site sampling days, 6 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.5. The bar jack's average mass was used from the Opitz 1996 model (815g) since only one fish was sampled in the hotspot.

Then, 1.5 was multiplied by 815g for the biomass, 1222.5g. 1222.5g divided by the area of the hotspot (3600m^2) = **0.339583333 g/m^2**

Production/biomass/year 1.17 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 10.1 Q/B (individual species value) (Opitz, 1996)

12. Coney (*Cephalopholis fulva*)

Fishes counted while diving	4
Fishes sampled (fishing/diving)	0
Total	4

Biomass g/m^2 Over 4 replicate site sampling days, 4 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1. The average mass of the coney was used from the Opitz 1996 model (165g).

Then, 1 was multiplied by 165g for the biomass, 165g. 165g divided by the area of the hotspot ($3600m^2$) = **0.04583333 g/m^2**

Production/biomass/year 0.78 P/B (individual species) (Opitz, 1996)

Consumption/biomass/year 7.8 Q/B (individual species value) (Opitz, 1996)

13. Foureye Butterflyfish (*Chaetodon capistratus*)

Fishes counted while diving	7
Fishes sampled (fishing/diving)	0
Total	0

Biomass g/m^2 Over 4 replicate site sampling days, 7 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.75. The average mass of the foureye butterflyfish was used from the Opitz 1996 model (31g).

Then, 1.75 was multiplied by 31g for the biomass, 54.25g. 54.25g divided by the area of the hotspot ($3600m^2$) = **0.015068444 g/m^2**

Production/biomass/year 2.02 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 14.4 Q/B (individual species value) (Opitz, 1996)

14. Spotfin Butterflyfish (*Chaetodon ocellatus*)

Fishes counted while diving	5
Fishes sampled (fishing/diving)	0
Total	0

Biomass g/m^2 Over 4 replicate site sampling days, 5 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.25. The average mass of the spotfin butterflyfish was used from the Opitz 1996 model (52g).

Then, 1.25 was multiplied by 52g for the biomass, 65g. 65g divided by the area of the hotspot (3600m^2) = **0.018055556 g/m^2**

Production/biomass/year 1.55 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 12.9 Q/B (individual species value) (Opitz, 1996)

15. Banded Butterflyfish (*Chaetodon striatus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the banded butterflyfish was used from the Opitz 1996 model (42g).

Then, 0.25 was multiplied by 42g for the biomass, 10.5g. 10.5g divided by the area of the hotspot (3600m^2) = **0.002916667 g/m^2**

Production/biomass/year 1.7 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 13.1 Q/B (individual species value) (Opitz, 1996)

16. Blue Chromis (*Chromis cyaena*)

Fishes counted while diving	7
Fishes sampled (fishing/diving)	0
Total	7

Biomass g/m² Over 4 replicate site sampling days, 7 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.75. The average mass of the blue chromis was used from the Opitz 1996 model (24g).

Then, 1.75 was multiplied by 24g for the biomass, 42g. 42g divided by the area of the hotspot (3600m²) = **0.011666667 g/m²**

Production/biomass/year 1.6 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 12.7 Q/B (individual species value) (Opitz, 1996)

17. Creole Wrasse (*Clepticus parrae*)

Fishes counted while diving	5
Fishes sampled (fishing/diving)	0
Total	5

Biomass g/m² Over 4 replicate site sampling days, 5 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.25. The average mass of the creole wrasse was used from the Opitz 1996 model (115g).

Then, 1.25 was multiplied by 115g for the biomass, 143.75g. 143.75g divided by the area of the hotspot (3600m²) = **0.039930556 g/m²**

Production/biomass/year 0.064 P/B (compartment 13 value) (Opitz, 1996)

Consumption/biomass/year 9.4 Q/B (individual species value) (Opitz, 1996)

18. Round Scad (*Decapterus punctatus*)

Fishes counted while diving	“Lots”
Fishes sampled (fishing/diving)	0
Total	“Lots”

Biomass g/m^2 Over 4 replicate site sampling days, many of these fish were seen. No proper estimate was given. The value of the biomass in g/m^2 was used from the Opitz model as a result.

0.0185 g/m^2 (Opitz 1996)

Production/biomass/year 0.83 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 12.7 Q/B (individual species value) (Opitz, 1996)

19. Yellowline goby (*Elacatinus horsti*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the yellowline goby was used from the Opitz 1996 model (0.8g).

Then, 0.25 was multiplied by 0.8g for the biomass, 0.2g. 0.2 g divided by the area of the hotspot ($3600m^2$) = **0.000055556 g/m^2**

Production/biomass/year 3.14 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 17.2 Q/B (individual species value) (Opitz, 1996)

20. Yellowprow goby (*Elacatinus xanthiprora*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0

Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the yellowprow goby was used from the Opitz 1996 model for yellowline goby since data is scarce, and fishes are similar in size (0.8g).

Then, 0.25 was multiplied by 0.8g for the biomass, 0.2g. 0.2 g divided by the area of the hotspot ($3600m^2$) = **0.000055556 g/m^2**

Production/biomass/year 3.14 P/B (individual species yellowline goby value) (Opitz, 1996)

Consumption/biomass/year 17.2 Q/B (individual species value yellowline goby) (Opitz, 1996)

21. Rock hind (*Epinephelus adscensionis*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the rock hind was used from the Opitz 1996 model (511g).

Then, 0.25 was multiplied by 511g for the biomass, 127.75g. 127.75g divided by the area of the hotspot ($3600m^2$) = **0.035486111 g/m^2**

Production/biomass/year 0.64 P/B (compartment 13) (Opitz, 1996)

Consumption/biomass/year 6.3 Q/B (individual species value) (Opitz, 1996)

22. Trumpetfish (*Fistularia tabacaria*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the trumpetfish was used from the Opitz 1996 model (2786g).

Then, 0.25 was multiplied by 2786g for the biomass, 696.5g. 696.5g divided by the area of the hotspot (3600m^2) = **0.193472222 g/m^2**

Production/biomass/year 0.38 P/B (compartment 12) (Opitz, 1996)

Consumption/biomass/year 4.7 Q/B (individual species value) (Opitz, 1996)

23. Nurse Shark (*Ginglymostoma cirratum*)

Fishes counted while diving	2
Fishes sampled (fishing/diving)	0
Total	2

Biomass g/m^2 Over 4 replicate site sampling days, 2 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.5. The average mass of the nurse shark was used from the Opitz 1996 model (129000g).

Then, 0.5 was multiplied by 129000g for the biomass, 64500g. 64500g divided by the area of the hotspot (3600m^2) = 17.91666667 g/m^2. This value was reduced by 99.7% (same as compartment 1 in Opitz 1996 model) to make sure the new “large sharks and rays” compartment wasn’t a negative biomass number = **0.05375 g/m^2**

Production/biomass/year 0.24 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 4.5 Q/B (individual species value) (Opitz, 1996)

24. Spotted moray (*Gymnothorax moringa*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the spotted moray was used from the Opitz 1996 model (245g).

Then, 0.25 was multiplied by 245g for the biomass, 61.25g. 61.25g divided by the area of the hotspot (3600m^2) = **0.017013889 g/m^2**

Production/biomass/year 0.64 P/B (compartment 13) (Opitz, 1996)

Consumption/biomass/year 4.9 Q/B (individual species value) (Opitz, 1996)

25. French Grunt (*Haemulon flavolineatum*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the French grunt was used from the Opitz 1996 model (124g).

Then, 0.25 was multiplied by 124g for the biomass, 31g. 31g divided by the area of the hotspot (3600m^2) = **0.008611111 g/m^2**

Production/biomass/year 0.9 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 10.7 Q/B (individual species value) (Opitz, 1996)

26. White Grunt (*Haemulon plumieri*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the white grunt was used from the Opitz 1996 model (351g).

Then, 0.25 was multiplied by 351g for the biomass, 87.75g. 87.75g divided by the area of the hotspot (3600m^2) = **0.024375 g/m^2**

Production/biomass/year 0.67 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 9.4 Q/B (individual species value) (Opitz, 1996)

27. Yellowhead wrasse (*Halichoeres garnoti*)

Fishes counted while diving	14
Fishes sampled (fishing/diving)	0
Total	14

Biomass g/m^2 Over 4 replicate site sampling days, 14 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 3.5. The average mass of the yellowhead wrasse was used from the Opitz 1996 model (34g).

Then, 3.5 was multiplied by 34g for the biomass, 119g. 119g divided by the area of the hotspot (3600m^2) = **0.033055556 g/m^2**

Production/biomass/year 1.665 P/B (compartment 14) (Opitz, 1996)

Consumption/biomass/year 10.6 Q/B (individual species value) (Opitz, 1996)

28. Rainbow wrasse (*Halichoeres pictus*)

Fishes counted while diving	4
Fishes sampled (fishing/diving)	0
Total	4

Biomass g/m^2 Over 4 replicate site sampling days, 4 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1. The average mass of the rainbow wrasse was used from the Opitz 1996 model (6g).

Then, 1 was multiplied by 6g for the biomass, 6g. 6g divided by the area of the hotspot (3600m^2) = **0.001666667 g/m^2**

Production/biomass/year 3.82 P/B (compartment 17) (Opitz, 1996)

Consumption/biomass/year 14.65 Q/B (individual species value) (Opitz, 1996)

29. Blackear wrasse (*Halichoeres poeyi*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the blackear wrasse was used from the Opitz 1996 model (39g).

Then, 0.25 was multiplied by 39g for the biomass, 9.75g. 9.75g divided by the area of the hotspot (3600m^2) = **0.002708333 g/m^2**

Production/biomass/year 1.665 P/B (compartment 14) (Opitz, 1996)

Consumption/biomass/year 9.4 Q/B (individual species value) (Opitz, 1996)

30. Queen Angelfish (*Holacanthurus ciliaris*)

Fishes counted while diving	2
Fishes sampled (fishing/diving)	0
Total	2

Biomass g/m^2 Over 4 replicate site sampling days, 2 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.5. The average mass of the queen angelfish was used from the Opitz 1996 model (513g).

Then, 0.5 was multiplied by 513g for the biomass, 256.5g. 256.5g divided by the area of the hotspot (3600m^2) = **0.07125 g/m^2**

Production/biomass/year 0.43 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 5.9 Q/B (individual species value) (Opitz, 1996)

31. Rock Beauty (*Holacanthus tricolor*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the rock beauty was used from the Opitz 1996 model (337g).

Then, 0.25 was multiplied by 337g for the biomass, 84.25g. 84.25g divided by the area of the hotspot (3600m^2) = **0.23402778 g/m^2**

Production/biomass/year 0.55 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 7.2 Q/B (individual species value) (Opitz, 1996)

32. Longspine squirrelfish (*Holocentrus rufus*)

Fishes counted while diving	10
Fishes sampled (fishing/diving)	0
Total	10

Biomass g/m^2 Over 4 replicate site sampling days, 10 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 2.5. The average mass of the squirrelfish was used from the Opitz 1996 model (92.2g).

Then, 2.5 was multiplied by 92.2g for the biomass, 230.5g. 230.5g divided by the area of the hotspot ($3600m^2$) = **0.064027778 g/m^2**

Production/biomass/year 2.71 P/B (individual species value) (S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)(S. Opitz, 1996)

Consumption/biomass/year 9.8 Q/B (individual species value) (Opitz, 1996)

33. Barred Hamlet (*Hypoplectrus puella*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the barred hamlet was used from the Opitz 1996 model (17g).

Then, 0.25 was multiplied by 17g for the biomass, 4.25g. 4.25g divided by the area of the hotspot ($3600m^2$) = **0.001180556 g/m^2**

Production/biomass/year 1.665 P/B (compartment 14) (Opitz, 1996)

Consumption/biomass/year 12.9 Q/B (individual species value) (Opitz, 1996)

34. Hogfish (*Lachnolaimus maximus*)

Fishes counted while diving	3
Fishes sampled (fishing/diving)	2
Total	5

Biomass g/m^2 Over 4 replicate site sampling days, 5 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.25. The hogfish masses sampled at CTX3, CTX4, and GY were used to calculate an average mass (564.85). Then, 1.25 was multiplied by 564.85 for the biomass, 706.0625g. 706.0625g divided by the area of the hotspot ($3600m^2$) = **0.196 g/m^2**

Production/biomass/year Z= total mortality

The equation $Z = \frac{P}{B} = \frac{K*(L_\infty - \bar{L})}{\bar{L} - L'}$ was used to calculate production/biomass/year.

K = curvature parameter of vBGF (/year) = 0.08 (Ault *et al.*, 2008)

L_∞ = asymptotic length (total length, cm) = 178cm (Ault *et al.*, 2008)

$\bar{L}$ = mean length in the population = 34.1cm (Ault *et al.*, 2008)

L' = mean length at entry into the fishery = 21cm (Smallest fish in our samples collected)

$Z = \frac{P}{B} = \frac{K*(L_\infty - \bar{L})}{\bar{L} - L'}$ = **0.349 P/B**

Consumption/biomass/year 4.8 Q/B (individual species) (S. Opitz, 1996)

35. Mutton Snapper (*Lutjanus analis*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the mutton snapper was used from the Opitz 1996 model (1422g).

Then, 0.25 was multiplied by 1422g for the biomass, 355.5g. 355.5g divided by the area of the hotspot (3600m^2) = **0.09875 g/m^2**

Production/biomass/year 0.33 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 6.0 Q/B (individual species value) (Opitz, 1996)

36. Schoolmaster Snapper (*Lutjanus apodus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the schoolmaster snapper was used from the Opitz 1996 model (904g).

Then, 0.25 was multiplied by 904g for the biomass, 226g. 226g divided by the area of the hotspot (3600m^2) = **0.062777778 g/m^2**

Production/biomass/year 0.54 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 6.5 Q/B (individual species value) (Opitz, 1996)

37. Dog snapper (*Lutjanus jocu*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the dog snapper was used from the Opitz 1996 model (3452g).

Then, 0.25 was multiplied by 3452g for the biomass, 863g. 863g divided by the area of the hotspot (3600m^2) = **0.239722222 g/m^2**

Production/biomass/year Z= total mortality

The equation $Z = \frac{P}{B} = \frac{K*(L_\infty - \bar{L})}{\bar{L} - L'}$ was used to calculate production/biomass/year.

K = curvature parameter of vBGF (/year) = 0.15 (Potts and Burton 2017)

L_∞ = asymptotic length (total length, cm) = 78.3cm (Potts and Burton 2017)

$\bar{L}$ = mean length in the population = 55.4cm (Potts and Burton 2017)

L' = mean length at entry into the fishery = 20 cm (Potts and Burton 2017)

$Z = \frac{P}{B} = \frac{K*(L_\infty - \bar{L})}{\bar{L} - L'}$ = **0.646893 P/B**

Consumption/biomass/year 5.0 Q/B (individual species value) (Opitz, 1996)

38. Sand Tilefish (*Malacanthus plumieri*)

Fishes counted while diving	4

Fishes sampled (fishing/diving)	5
Total	9

Biomass g/m^2 Over 4 replicate site sampling days, 9 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 2.25. The average mass of the sand tilefish was used from the Opitz 1996 model (127g).

Then, 2.25 was multiplied by 127g for the biomass, 285.75g. 285.75g divided by the area of the hotspot (3600m^2) = **0.079375 g/m^2**

Production/biomass/year 0.42 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 6.8 Q/B (individual species value) (Opitz, 1996)

39. Black durgon (*Melichthys niger*)

Fishes counted while diving	51
Fishes sampled (fishing/diving)	0
Total	51

Biomass g/m^2 Over 4 replicate sampling days, 51 fish in total were counted from diving. The total number of fish (51) was divided by the replicate sampling trips (4) = 12.75. The mass of the triggerfish was used from Opitz (1996) 248g. Then, 12.75 was multiplied by 248g = 3162g. 3162g divided by 3600m^2 = **0.87833 g/m^2**.

Production/biomass/year 0.71 P/B (compartment 11) (Opitz, 1996)

Consumption/biomass/year 23.21 Q/B (individual species value) (Opitz 1996)

40. Yellowtail Damselfish (*Microspathodon chrysurus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0

Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the yellowtail damselfish was used from the Opitz 1996 model (106g).

Then, 0.25 was multiplied by 106g for the biomass, 26.5g. 26.5g divided by the area of the hotspot (3600m^2) = **0.007361111 g/m^2**

Production/biomass/year 0.71 P/B (compartment 11) (Opitz, 1996)

Consumption/biomass/year 25.8 Q/B (individual species value) (Opitz, 1996)

41. Yellow Goatfish (*Mulloidichthys martinicus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the yellow goatfish was used from the Opitz 1996 model (114g).

Then, 0.25 was multiplied by 114g for the biomass, 28.5g. 28.5g divided by the area of the hotspot (3600m^2) = **0.007916667 g/m^2**

Production/biomass/year 0.98 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 10.5 Q/B (individual species value) (Opitz, 1996)

42. Blackbar soldierfish (*Myripristis jacobus*)

Fishes counted while diving	2
Fishes sampled (fishing/diving)	0

Total	2

Biomass g/m² Over 4 replicate site sampling days, 2 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.5. The average mass of the blackbar soldierfish was used from the Opitz 1996 model (124g).

Then, 0.5 was multiplied by 124g for the biomass, 51g. g divided by the area of the hotspot (3600m^2) = **0.014166667 g/m²**

Production/biomass/year 1.265 P/B (compartment 8) (Opitz, 1996)

Consumption/biomass/year 11.2 Q/B (individual species value) (Opitz, 1996)

43. Yellowtail Snapper (*Ocyurus chrysurus*)

Fishes counted while diving	5
Fishes sampled (fishing/diving)	3
Total	8

Biomass g/m² Over 4 replicate site sampling days, 8 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 2. The yellowtail snapper's average mass was found from the fishes sampled in the hotspot (155.67g).

Then, 2 was multiplied by 155.67g for the biomass, 311.34g. 311.34g divided by the area of the hotspot (3600m^2) = **0.086483333 g/m²**

Production/biomass/year Z= total mortality

The equation $Z = \frac{P}{B} = \frac{K*(L_\infty - \bar{L})}{\bar{L} - L'}$ was used to calculate production/biomass/year.

K = curvature parameter of vBGF (/year) = 0.139 (Manooch and Drennon 1987)

L_∞ = asymptotic length (total length, cm) = 502.525cm (Manooch and Drennon 1987)

$\bar{L}$ = mean length in the population = 355cm (Manooch and Drennon 1987)

L' = mean length at entry into the fishery = 117cm (Manooch and Drennon 1987)

$Z = \frac{P}{B} = \frac{K*(L_\infty - \bar{L})}{\bar{L} - L'}$ = **0.619853 P/B**

Consumption/biomass/year 7.9 Q/B (individual species value) (Opitz, 1996)

44. Grey Angelfish (*Pomacanthus arcuatus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m² Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the grey angelfish was used from the Opitz 1996 model (3201g).

Then, 0.25 was multiplied by 3201g for the biomass, 800.25g. 800.25g divided by the area of the hotspot (3600m^2) = **0.222291667 g/m^2**

Production/biomass/year 0.63 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 6.7 Q/B (individual species value) (Opitz, 1996)

45. French angelfish (*Pomacanthus paru*)

Fishes counted while diving	2
Fishes sampled (fishing/diving)	0
Total	2

Biomass g/m² Over 4 replicate site sampling days, 2 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.5. The average mass of the grey angelfish was used from the Opitz 1996 model (714g).

Then, 0.5 was multiplied by 714g for the biomass, 357g. 357g divided by the area of the hotspot (3600m^2) = **0.099166667 g/m^2**

Production/biomass/year 0.62 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 7.6 Q/B (individual species value) (Opitz, 1996)

46. Spotted Goatfish (*Pseudupeneus maculatus*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the spotted goatfish was used from the Opitz 1996 model (101g).

Then, 0.25 was multiplied by 101g for the biomass, 25.25g. 25.25g divided by the area of the hotspot (3600m^2) = **0.007013889 g/m^2**

Production/biomass/year 0.95 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 10.8 Q/B (individual species value) (Opitz, 1996)

47. Lionfish (*Pterois volitans*)

Fishes counted while diving	4
Fishes sampled (fishing/diving)	0
Total	4

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1. The average mass of 194.5 was used from Darling *et al*. (2011).

Then, 1 was multiplied by 194.5g for the biomass, 194.5g. 194.5g divided by the area of the hotspot ($3600m^2$) = **0.054027778 g/m^2**

Production/biomass/year M= natural mortality

K = curvature parameter of vBGF (/year) = 0.42 (Edwards *et al* 2014)

L_∞ = asymptotic length (total length, cm) = 34.9 (Edwards *et al* 2014)

T_c = average temperature in C^o = 29.8 (google search, Ponce)

$M = P/B = K^{0.65} * L_\infty^{-0.279} * T_c^{0.463}$ = **1.016777 P/B**

Consumption/biomass/year 26.35 Q/B (Chargaris *et al* 2017)

48. Striped Parrotfish (*Scarus iseri*)

Fishes counted while diving	85
Fishes sampled (fishing/diving)	0
Total	85

Biomass g/m^2 Over 4 replicate site sampling days, 86 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 21.25. The average mass of the striped parrotfish was used from the Opitz 1996 model (36g).

Then, 29.25 was multiplied by 36 for the biomass, 765g. 765g divided by the area of the hotspot ($3600m^2$) = **.2125 g/m^2**

Production/biomass/year 0.94 P/B (compartment 25) (Opitz, 1996)

Consumption/biomass/year 36.8 Q/B (individual species value) (Opitz, 1996)

49. Princess Parrotfish (*Scarus taeniopterus*)

Fishes counted while diving	15
Fishes sampled (fishing/diving)	1
Total	16

Biomass g/m^2 Over 4 replicate site sampling days, 16 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 4. The average mass of the princess parrotfish was used from the Opitz 1996 model (284g).

Then, 4 was multiplied by 284g for the biomass, 1136g. 1136g divided by the area of the hotspot (3600m^2) = **0.315555556 g/m^2**

Production/biomass/year 1.2 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 20.8 Q/B (individual species value) (Opitz, 1996)

50. Queen Parrotfish (*Scarus vetula*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the queen parrotfish was used from the Opitz 1996 model (1434g).

Then, 0.25 was multiplied by 1434g for the biomass, 358.5g. 358.5g divided by the area of the hotspot (3600m^2) = **0.099583333 g/m^2**

Production/biomass/year 1.09 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 14.4 Q/B (individual species value) (Opitz, 1996)

51. King Mackerel (*Scomberomorus cavalla*)

Fishes counted while diving	1
Fishes sampled (trolling)	2
Total	3

Biomass g/m^2 Over 4 replicate site sampling days, 3 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.75. The king mackerel's average mass was calculated from 5 fishes sampled (2 from Guayama 3 from Maunabo) (3172.26g).

Then, 0.75 was multiplied by 3172.26g for the biomass, 2379.195g. 2379.195g divided by the area of the hotspot (100000m^2 trolling area) = **0.02379195 g/m^2**

Production/biomass/year 0.37 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 7.4 Q/B (individual species value) (Opitz, 1996)

52. Cero (*Scomberomorus regalis*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the cero was calculated from the fishes sampled in Maunabo (464.4g).

Then, 0.25 was multiplied by 464.4g for the biomass, 116.1g. 116.1g divided by the area of the hotspot (3600m^2) = **0.3225 g/m^2**

Production/biomass/year 0.47 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 10.8 Q/B (individual species value) (Opitz, 1996)

53. Redband Parrotfish (*Sparisoma aurofrenatum*)

Fishes counted while diving	2
Fishes sampled (fishing/diving)	0
Total	2

Biomass g/m^2 Over 4 replicate site sampling days, 2 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.5. The average mass of the redband parrotfish was used from the Opitz 1996 model (84g).

Then, 0.5 was multiplied by 84g for the biomass, 42g. 42g divided by the area of the hotspot (3600m^2) = **0.011666667 g/m^2**

Production/biomass/year 0.94 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 29.5 Q/B (individual species value) (Opitz, 1996)

54. Stoplight Parrotfish (*Sparisoma viride*)

Fishes counted while diving	9
Fishes sampled (fishing/diving)	3
Total	12

Biomass g/m^2 Over 4 replicate site sampling days, 12 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 3. The average mass of the stoplight parrotfish was calculated from the fishes sampled (513.33g).

Then, 3 was multiplied by 513.33g for the biomass, 1539.99g. 1539.99g divided by the area of the hotspot (3600m^2) = **0.427775 g/m^2**

Production/biomass/year 1.155 P/B (compartment 24) (Opitz, 1996)

Consumption/biomass/year 20.7 Q/B (individual species value) (Opitz, 1996)

55. Great Barracuda (*Sphyraena barracuda*)

Fishes counted while diving	5
Fishes sampled (fishing/diving)	2
Total	7

Biomass g/m^2 Over 4 replicate site sampling days, 7 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 1.75. The barracuda masses sampled at CTX3, CTX4, and GY were used to calculate an average mass (2920.03g). Then, 1.75 was multiplied by 2920.03g for the biomass, 5110.04g. 5110.04g divided by the area of the hotspot (3600m^2) = **1.42g/m^2**

Production/biomass/year 0.2319 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 3.3 Q/B (individual species value) (Opitz 1996)

56. Bicolor damselfish (*Stegastes partitus*)

Fishes counted while diving	27
Fishes sampled (fishing/diving)	0
Total	27

Biomass g/m^2 Over 4 replicate site sampling days, 27 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 6.75. The average mass of the bicolor damselfish was used from the Opitz 1996 model for *Stegastes leucostictus* (similar length) (13g).

Then, 6.75 was multiplied by 13g for the biomass, 87.75g. 87.75g divided by the area of the hotspot (3600m^2) = **0.024375 g/m^2**

Production/biomass/year 1.6 P/B (individual species value for *Stegastes leucostictus*, no data for *Stegastes partitus* and *S. leucostictus* was the closest species with available data) (Opitz, 1996)

Consumption/biomass/year 16 Q/B (compartment 21, similar to other damselfish in compartment) (Opitz, 1996)

57. Cocoa Damselfish (*Stegastes variabilis*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	1

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the cocoa damselfish was used from the Opitz 1996 model (14g).

Then, 0.25 was multiplied by 14g for the biomass, 3.5g. 3.5g divided by the area of the hotspot (3600m^2) = **0.000972222 g/m^2**

Production/biomass/year 1.6 P/B (compartment 20) (Opitz, 1996)

Consumption/biomass/year 23.1 Q/B (individual species value) (Opitz, 1996)

58. Bluehead wrasse (*Thalassoma bifasciatum*)

Fishes counted while diving	48
Fishes sampled (fishing/diving)	0
Total	48

Biomass g/m^2 Over 4 replicate site sampling days, 48 fish in total were counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 12. The average mass of the bluehead wrasse was used from the Opitz 1996 model (25g).

Then, 12 was multiplied by 25g for the biomass, 300g. g divided by the area of the hotspot (3600m^2) = **0.08333333 g/m^2**

Production/biomass/year 1.73 P/B (individual species value) (Opitz, 1996)

Consumption/biomass/year 9.7 Q/B (individual species value) (S. Opitz, 1996)

59. Redtail Triggerfish (*Xanthichthys ringens*)

Fishes counted while diving	1
Fishes sampled (fishing/diving)	0
Total	0

Biomass g/m^2 Over 4 replicate site sampling days, 1 fish in total was counted between diving and sampling. The total number of fish was divided by the replicate sampling trips (4) = 0.25. The average mass of the redtail triggerfish was used from the Opitz 1996 model (83g).

Then, 0.25 was multiplied by 83g for the biomass, 20.75g. 20.75g divided by the area of the hotspot (3600m^2) = **0.005763889 g/m^2**

Production/biomass/year 0.64 P/B (compartment 13) (Opitz, 1996)

Consumption/biomass/year 10.3 Q/B (individual species value) (S. Opitz, 1996)

Calculating New Opitz Compartments (Hotspot)

60. Large sharks/rays C | Opitz Compartment 1

Biomass Only one shark was identified from the sampling sites. The value of that compartment (nurse shark) was 17.9g/m^2. The nurse shark biomass was reduced by 99.7% (17.9g/m^2*.003= .05375g/m^2) to match the 99.7% biomass g/m^2 reduction Opitz did in her 1996 model and subtracted from the 0.3g/m^2 in the Opitz model for compartment 1 (0.3g/m^2). 0.3g/m^2 – 0.05375g/m^2 = **0.24625g/m^2**

P/B The P/B value was kept the same **0.24**

Q/B The Q/B value was kept the same **4.9**

61. Sharks/scombrids | Opitz Compartment 2

Biomass Opitz compartment 2 (0.414 g/m^2) – cero (0.03225g/m^2) – king mackeral 0.02379195g/m^2 = **0.35795805 g/m^2**

P/B The P/B value was calculated by removing the P/B value for king mackerel and cero from compartment 2 and finding the new median value = **0.29**

Q/B The Q/B value was calculated by removing the Q/B values for cero and king mackerel from compartment 2 and finding the new median value with the other species = **9.15**

62. Large Jacks C | Opitz Compartment 3

Biomass The biomass value was the same. No jacks from this compartment were observed or sampled in the hotspot = **0.181 g/m^2**

P/B The P/B value is the same as Opitz 1996 = **0.525**

Q/B The Q/B value is the same as Opitz 1996 = **5.7**

63. Intermediate jacks C (4)

Opitz Compartment 4

Biomass The biomass value of Opitz compartment 4 (1.63 g/m^2) – biomass for observed *Caranx lugubris* (0.234861111 g/m^2) – biomass for observed *Caranx ruber* (0.339583333 g/m^2) = **1.05555 g/m^2**

P/B The P/B value was kept the same as the original Opitz 1996 model since one value for P/B for the compartment was used, which is the single species value for *Caranx ruber* = **1.7**

Q/B The Q/B value was calculated by removing the values for *Caranx lugubris* and *Caranx ruber* from compartment 4 in the Opitz model and finding the new median value = **8.4**

64. Small jacks C | Opitz Compartment 5

Biomass The biomass value of Opitz compartment 5 (1.0g/m^2) – biomass for *Decapterus punctatus* (0.0185) = **0.9815 g/m^2**

P/B The P/B value was calculated by removing the value for *Decapterus punctatus* (0.83) from compartment 5 and calculating a new median value = **0.775**

Q/B The Q/B value was calculated by removing the value for *Decapterus punctatus* (12.7) from compartment 5 and calculating a new median value = **12.3**

65. Intermediate reef fish C1 | Opitz Compartment 6

Biomass The biomass value of Opitz compartment 6 (3.024 g/m^2) – biomass calculated for the sampled *Balistes vetula* (0.55 g/m^2) – *Lutjanus analis* (0.9875 g/m^2) -*Lutjanus apodus* (0.06277778 g/m^2) – *Ocyurus chrysurus* (0.086483333 g/m^2) – *Pomacanthus arcuatus* (0.222291667 g/m^2) = **1.999447222 g/m^2**

P/B The P/B value was calculated by removing the P/B values for the previously mentioned species and calculating the new median value from the remaining groups = **0.5055**

Q/B The Q/B value was calculated by removing the Q/B values for the previously mentioned species from group 6 then calculating the median of the remaining values = **7.0**

66. Large to Intermediate Schooling Fish P | Opitz Compartment 7

Biomass The biomass value was used from the Opitz 1996 model since no species from this compartment were sampled = **11.172 g/m^2**

P/B The P/B value was used from the Opitz 1996 model = **0.68**

Q/B The Q/B value was used from the Opitz 1996 model = **12.4**

67. Intermediate reef fish C2 | Opitz Compartment 8

Biomass The biomass of Opitz compartment 8 (6.493 g/m^2) – the new biomass values from the observed *Abudefduf saxatilis* (0.069444444 g/m^2) – *Anisotremus virginicus* (0.09125 g/m^2) – *Calamus pennatula* (0.030486111 g/m^2) – *Chaetodon capistratus* (0.015068444 g/m^2) – *Chaetodon ocellatus* (0.018055556 g/m^2) – *Chaetodon striatus* (0.002916667 g/m^2) – *Haemulon flavolineatum* (0.0.008611111 g/m^2) – *Haemulon plumierii* (0.024375 g/m^2) – *Holocentrus rufus* (0.64027778 g/m^2) – *Mulloidichthys martinicus* (0.007916667 g/m^2) – *Myripristis Jacobus* (0.014166667 g/m^2) – *Pseudupeneus maculatus* (0.007013889 g/m^2) = **6.139666667 g/m^2**

P/B The P/B value was calculated by removing the P/B values from the species listed above and finding the new median from the remaining groups = **1.325**

Q/B The Q/B value was calculated by removing the Q/B values from the species listed above and finding the new median from the remaining groups = **2.7**

68. Hemiramphidae H | Opitz Compartment 9

Biomass The biomass from Opitz 1996 compartment 9 since no fishes from this group were sampled or identified in the hotspot = **1.125 g/m^2**

P/B The P/B value was used from the Opitz 1996 model = **1.23**

Q/B The Q/B value was used from the Opitz 1996 model = **39.10**

69. Kyphosidae H | Opitz Compartment 10

Biomass The biomass from Opitz 1996 compartment 10 was used since no fishes from this group were sampled or identified in the hotspot = **2.42g/m^2**

P/B The P/B value was used from the Opitz 1996 model = **0.6**

Q/B The Q/B value was used from the Opitz 1996 model = **23.6**

70. Intermediate Reef Fish H | Opitz Compartment 11

Biomass The biomass of Opitz compartment 11 (9.651 g/m^2) – *Acanthurus bahianus* (0.056527778 g/m^2) – *Acanthurus chirurgus* (0.014722222g/m^2) – *Acanthurus coeruleus* (0.176388889g/m^2) – *Melichthys niger* (0.878333333g/m^2) – *Microspathodon chrysurus* (0.007361111g/m^2) = **8.517666667 g/m^2**

P/B The P/B value was calculated by removing the P/B values for the species listed above from compartment 11 and determining the median of the remaining values = **0.765**

Q/B The Q/B value was calculated by removing the Q/B values for the species listed above from compartment 12 and determining the median of the remaining values = **26.85**

71. Large Reef Fish | Opitz Compartment 12

Biomass Opitz compartment 12 (2.525g/m^2) – *Fistularia tabacaria* (0.193472222g/m^2) – *Lachnolaimus maximus* (0.196128472 g/m^2) – *Lutjanus jocu* (0.239722222 g/m^2) – *Sphyraena barracuda* (1.419456597 g/m^2) = **0.476220486 g/m^2**

P/B The P/B value was calculated by removing the P/B values for the species above from compartment 12 then recalculating the P/B for the group by finding the median from the remaining species = **0.38**

Q/B The Q/B value was calculated by removing the Q/B values for the species above from compartment 12 then recalculating the Q/B for the group by finding the median from the remaining species = **3.7**

72. Intermediate Reef Fish C3 | Opitz Compartment 13

Biomass Opitz compartment 13 (7.25 g/m^2) – *Bodians rufus* (0.195416667g/m^2) – *Clepticus parrae* (0.039930556 g/m^2) – *Epinephalus adscensionis* (0.035486111 g/m^2) – *Gymnothorax moringa* (0.017013889 g/m^2) – *Malacanthus plumieri* (0.079375 g/m^2) – *Xanthichthys ringens* (0.005763889 g/m^2) – *Cephalopholis fulva* (0.045833333g/m^2) = **6.139666667 g/m^2**

P/B The P/B value was calculated by removing the P/B values of the species listed above from the Opitz compartment and calculating the median of the remaining values = **0.64**

Q/B The Q/B value was calculated by removing the Q/B values of the species listed above from the Opitz compartment and calculating the median of the remaining values = **6.3**

73. Small Reef Fish C1 | Opitz Compartment 14

Biomass Opitz compartment 14 (4.419 g/m^2) – *Chromis cyaena* (.03990556 g/m^2) – *Hypoplectrus puella* (0.001180556 g/m^2) – *Halichoeres garnoti* (0.003055556 g/m^2) –

Halichoeres poeyi (0.002708333 g/m^2) – *Thalassoma bifasciatum* (0.8333333333 g/m^2) = **4.287055555 g/m^2**

P/B The P/B value was calculated by removing the P/B values of the above fishes from Opitz compartment 14 and finding the new median value = **1.285**

Q/B The Q/B value was calculated by removing the Q/B values of the above fishes from Opitz compartment 14 and finding the new median value = **9.75**

74. Small Schooling Fish P | Opitz Compartment 15

Biomass Opitz compartment 15 = **10.146 g/m^2**

P/B The P/B value of Opitz compartment 15 = **3.54**

Q/B The Q/B value of Opitz compartment 15 = **18.85**

75. Engraulidae H | Opitz Compartment 16

Biomass Opitz compartment 16 = **3.325 g/m^2**

P/B The P/B value of Opitz compartment 16 = **2.835**

Q/B The Q/B value of Opitz compartment 16 = **43.4**

76. Small Reef Fish H | Opitz Compartment 17

Biomass Opitz Compartment 17 = **1.298333333 g/m^2**

P/B The P/B value of Opitz compartment 17 = **3.82**

Q/B The Q/B value of Opitz compartment 17 = **14.65**

77. Large Groupers | Opitz Compartment 18

Biomass Opitz Compartment 18 = **.0725 g/m^2**

P/B The P/B value of Opitz compartment 18 = **0.37**

Q/B The Q/B value of Opitz compartment 18 = **2.3**

78. Intermediate Reef Fish C4 | Opitz Compartment 19

Biomass Opitz compartment 19 (1.431g/m^2) – *Aluterus scriptus* (0.052708333 g/m^2) – *Holoacanthus tricolor* (0.023402778 g/m^2) – *Holacanthus ciliaris* (0.07125 g/m^2) – *Pomacanthus paru* (0.099166667 g/m^2) = **1.184472222 g/m^2**

P/B The P/B value was calculated by removing the P/B values of the above fishes from Opitz compartment 19 and finding the new median value = **0.55**

Q/B The Q/B value was calculated by removing the Q/B values of the above fishes from Opitz compartment 19 and finding the new median value = **5.7**

79. Small Reef Fish O1 | Opitz Compartment 20

Biomass Opitz compartment 20 (3.534 g/m^2) – *Stegastes variabilis* (0.000972222 g/m^2) = **3.533027778 g/m^2**

P/B The P/B value was calculated by removing the P/B values of the above fishes from Opitz compartment 20 and finding the new median value = **1.6**

Q/B The Q/B value was calculated by removing the Q/B values of the above fishes from Opitz compartment 20 and finding the new median value = **18.2**

80. Small Reef Fish O2 | Opitz Compartment 21

Biomass Opitz compartment 21 = **0.99 g/m^2**

P/B The P/B value of Opitz compartment 21 = **1.505**

Q/B The Q/B value of Opitz compartment 21 = **16**

81. Small Reef Fish O3 | Opitz Compartment 22

Biomass Opitz compartment 22 = **0.96 g/m^2**

P/B The P/B value of Opitz compartment 22 = **2.53**

Q/B The Q/B value of Opitz compartment 22 = **39.7**

82. Large Scaridae H | Opitz Compartment 23

Biomass Opitz compartment 23 (18.982 g/m^2) – *Scarus vetula* (0.99583333 g/m^2) = **18.88241667 g/m^2**

P/B The P/B value was calculated by removing the P/B values of the above fishes from Opitz compartment 23 and finding the new median value = **0.85**

Q/B The Q/B value was calculated by removing the Q/B values of the above fishes from Opitz compartment 23 and finding the new median value = **13.5**

83. Intermediate Scaridae | Opitz Compartment 24

Biomass Opitz compartment 24 (5.51 g/m^2) – *Sparisoma viride* (0.427775 g/m^2) – *Scarus taeniopterus* (0.315555556 g/m^2) = **4.766669444 g/m^2**

P/B The P/B value was calculated by removing the P/B values of the above fishes from Opitz compartment 24 and finding the new median value = **1.11**

Q/B The Q/B value was calculated by removing the Q/B values of the above fishes from Opitz compartment 24 and finding the new median value = **20.05**

84. Small Scaridae | Opitz Compartment 25

Biomass Opitz compartment 25 (5.075 g/m^2) – *Scarus iseri* (0.2125 g/m^2) - – *Sparisoma aurofrenatum* (.011666667 g/m^2) = **4.850833333 g/m^2**

P/B The P/B value was calculated by removing the P/B values of the above fishes from Opitz compartment 25 and finding the new median value = **0.94**

Q/B The Q/B value was calculated by removing the Q/B values of the above fishes from Opitz compartment 25 and finding the new median value = **33.9**

85. Blenniidae H | Opitz Compartment 26

Biomass Opitz compartment 26 = **0.6 g/m^2**

P/B The P/B value of Opitz compartment 26 = **2.84**

Q/B The Q/B value of Opitz compartment 26 = **36.1**

86. Small Gobiidae C | Opitz Compartment 27

Biomass Opitz compartment 27 = **0.174988889 g/m^2**

P/B The P/B value of Opitz compartment 27 = **3.14**

Q/B The Q/B value of Opitz compartment 27 = **17.45**

Opitz's (1996) compartments 28-50 stayed the same.

Table 6-1 Table of basic input parameters calculated from identified and sampled species in the Guayama hotspot in Puerto Rico. Newly calculated Opitz compartments start with code #60.

Code	Compartment	Biomass g/m^2	P/B	Q/B	Code	Compartment	Biomass g/m^2	P/B	Q/B
1	Abudefduf saxatilis	0.069444444	1.265	13.28	**56**	Stegastes partitus	0.024375	1.6	16
2	Acanthurus bahianus	0.056527778	1	34.38	**57**	Stegastes variabilis	0.000972222	1.6	23.1
3	Acanthurus chirurgus	0.014722222	0.71	24.7	**58**	Thalassoma bifasciatum	0.083333333	1.73	9.7
4	Acanthurus coeruleus	0.176388889	0.7	24.4	**59**	Xanthichthys ringens	0.005763889	0.64	10.3
5	Aluterus scriptus	0.052708333	0.55	6.8	**60**	Large sharks/rays C	0.24625	0.24	4.9
6	Anisotremus virginicus	0.09125	1.265	10.3	**61**	Sharks/scombrids C	0.35795805	0.29	9.15
7	Balistes vetula	0.55425	0.56	6.9	**62**	Large jacks C	0.181	0.525	5.7
8	Bodianus rufus	0.195416667	0.64	5.9	**63**	Intermediate Jacks C	1.055555557	1.7	8.4
9	Calamus pennatula	0.030486111	1.265	9.3	**64**	Small jacks C	0.9815	0.83	12.5
10	Caranx lugubris	0.234861111	1.17	9.6	**65**	Intermediate Reef Fish C1	2.085930555	0.5055	7
11	Caranx ruber	0.339583333	1.17	10.1	**66**	Large to Intermediate Schooling Fish P	11.172	0.68	12.4
12	Cephalopholis fulva	0.045833333	0.78	7.8	**67**	Intermediate reef fish C2	6.139666667	1.325	2.7
13	Chaetodon capistratus	0.015069444	2.02	14.4	**68**	Hemiramphidae H	1.125	1.23	39.1
14	Chaetodon ocellatus	0.018055556	1.55	12.9	**69**	Kyphosidae H	2.42	0.6	23.6
15	Chaetodon striatus	0.002916667	1.7	13.1	**70**	Intermediate Reef Fish H	8.517666667	0.765	26.85
16	Chromis cyanea	0.011666667	1.6	12.7	**71**	Large Reef Fish	0.476220487	0.38	3.7
17	Clepticus parrae	0.039930556	0.64	9.4	**72**	Intermediate reef fish C3	6.831060555	0.64	6.4
18	Decapterus punctatus	0.0185	0.83	12.7	**73**	Small reef fish C1	4.287055555	1.285	9.75
19	Elacatinus horsti	5.55556E-05	3.14	17.2	**74**	Small schooling fish P	10.146	3.54	18.85
20	Elacatinus xanthiprora	5.55556E-05	3.14	17.2	**75**	Engraulidae H	3.325	2.835	43.4
21	Epinephelus adscensionis	0.035486111	0.64	6.3	**76**	Small reef fish H	1.298333333	3.82	14.65
22	Fistularia tabacaria	0.193472222	0.38	4.7	**77**	Large groupers C	0.725	0.37	2.3
23	Ginglymostoma cirratum	0.05375	0.24	4.5	**78**	Intermediate reef fish C4	1.184472222	0.55	5.7
24	Gymnothorax moringa	0.017013889	0.64	4.9	**79**	Small reef fish O1	3.533027778	1.6	18.2
25	Haemulon flavolineatum	0.008611111	0.9	10.7	**80**	Small reef fish O2	0.99	1.505	16
26	Haemulon plumierii	0.024375	0.67	9.4	**81**	Small reef fish O3	0.96	2.53	39.7
27	Halichoeres garnoti	0.033055556	1.665	10.6	**82**	Large Scaridae H	18.88241667	0.85	13.5
28	Halichoeres pictus	0.001666667	3.82	14.65	**83**	Intermediate scaridae H	4.766669444	1.11	20.05
29	Halichoeres poeyi	0.002708333	1.665	9.4	**84**	Small scaridae H	4.850833333	0.94	33.9
30	Holacanthus ciliaris	0.07125	0.43	5.9	**85**	Blenniidae H	0.6	2.84	36.1
31	Holacanthus tricolor	0.023402778	0.55	7.2	**86**	Small Gobiidae C	0.174988889	3.14	17.45
32	Holocentrus rufus	0.064027778	2.71	9.8	**87**	Sea birds	0.017	5.4	80
33	Hypoplectrus puella	0.001180556	1.665	12.9	**88**	Squids	1.5	1.3	17.5
34	Lachnolaimus maximus	0.196128472	0.3782	4.8	**89**	Sea turtles	0.5	0.15	3.5
35	Lutjanus analis	0.09875	0.33	6	**90**	Octopuses	8.4	1.9	6.76
36	Lutjanus apodus	0.062777778	0.54	6.5	**91**	Lobsters	3.27	1	7.4
37	Lutjanus jocu	0.239722222	0.64689	5	**92**	Crabs	19	1.6	14
38	Malacanthus plumieri	0.079375	0.42	6.8	**93**	Shrimps/hermit crabs/stomatopods	10	2.8	26.9
39	Melichthys niger	0.878333333	0.71	23.21	**94**	Amphipods/isopods/tanaids/pycnogonids	13.25	5	125.5
40	Microspathodon chrysurus	0.007361111	0.71	25.8	**95**	Asteroids	25	0.49	3.24
41	Mulloidichthys martinicus	0.007916667	0.98	10.5	**96**	Echinoids	100	1.1	3.7
42	Myripristis jacobus	0.014166667	1.265	11.2	**97**	Gastropods	46.8	2.8	14
43	Ocyurus chrysurus	0.086483333	0.61985	7.9	**98**	Chitons/scaphopods	62	0.36	11.7
44	Pomacanthus arcuatus	0.222291667	0.63	6.7	**99**	Polychaetes/priapuloids/ophiuroids	33	5.2	61.6
45	Pomacanthus paru	0.099166667	0.62	7.6	**100**	Holothurids/sipunculids/echiuroids/hemic	66.24	0.31	3.36
46	Pseudupeneus maculatus	0.007013889	0.95	10.8	**101**	Bivalves	109.25	2.23	9.5
47	Pterois volitans	0.054027778	1.01678	26.35	**102**	Ascidians/barnacles/bryozoans	137.4	2.3	20
48	Scarus iseri	0.2125	0.94	36.8	**103**	Sponges	800	1.5	5
49	Scarus taeniopterus	0.315555556	1.2	20.8	**104**	Corals/sea anemones	121	1.09	9
50	Scarus vetula	0.099583333	1.09	14.4	**105**	Zooplankton	32	40	165
51	Scomberomorus cavalla	0.02379195	0.37	7.4	**106**	Decomposers/microfauna	15	280	1900
52	Scomberomorus regalis	0.03225	0.47	10.8	**107**	Phytoplankton	40	70	N/A
53	Sparisoma aurofrenatum	0.011666667	0.94	29.5	**108**	Benthic autotrophs	1300	13.25	N/A
54	Sparisoma viride	0.427775	1.155	20.7	**109**	Detritus	2000	N/A	N/A
55	Sphyraena barracuda	1.419456597	0.25	3.3					

Diet Composition Matrix

I calculated a new diet composition matrix for both the hotspot and the coldspot models. Since both models' compartments are the same, and we could not sample enough fishes to complete a gut-content analysis due to unforeseen circumstances, both models have a similar diet matrix. This approach is not optimal because we believe that each reef species could be consuming different prey items or different amounts of each prey due to availability. However, Randall (1967) was the base for all diet matrix data, aside from a few species that were not in his report, and is a reliable source for Caribbean reef species diet data.

Randall listed prey values with volume. However, fishes were given a total volume in the diet and were not split among each species, family, or group within the prey group. If Randall listed a family, group, or genus, then we split the group again for the number of species within those groups in our models. The methods were as follows:

1. Prey values for each predator were identified from Randall’s (1967) report and were split evenly, and assigned to their respective compartments in our model. (Prey data for *Pterois volitans* was unavailable in Randall (1967), but was found in Morris and Akins (2009)
2. If the species/group/genus/family of fish was in the Randall predator diet list and was not identified while diving or sampling, then the prey item was added to its respective Opitz (1996) prey compartment in the model
3. For “fish” prey, the proportion of the diet listed was split evenly among the species/groups/genus/families listed underneath the “fish” category in Randall’s paper.

4. When groups/genus/families were listed, they were broken up evenly among all species in our model (i.e., if "scarids" made up 12% of the diet and there were three scarids that the predator ate in our model, then each scarid species was given a 4% diet contribution)
5. If prey items listed a group/genus/family AND an individual species of that group/genus/family in Randall's report, they were treated as two separate items. The unique species listed received the same diet proportion as the family. Then, the family was split evenly among all other species of that family in our model. Example: Species A is in the same family as Species B, and Species C. Randall (1967) listed both Species A and the family of Species A, B, and C. Species A was given the same proportion of the diet as the family with Species B and C.
6. If a prey item was listed as "unidentified crustaceans," it was split evenly among the crustaceans already listed in that predator's diet (stomatopods, isopods, crabs, shrimp, tanaids, amphipods, mysids, copepods, ostracods).
7. If "unidentified animal material" was listed, it was split evenly among all other compartments that fell under the "Animalia" kingdom.
8. If "fishes" was listed as a prey item, then the fishes' volume was divided evenly among all fishes in the model smaller than the predator fish.
9. The following were put into the "zooplankton" compartment as per Opitz 1996 paper; insects, polychaete larvae, scyphozoans (medusae), siphonophores (medusae), barnacle larvae, copepods, crab larvae, euphasids, hermit crab larvae, isopod larvae, mysids, ostracods, scyllarid larvae, shrimp larvae, stomatopod larvae, ctenophores (medusae), bivalve larvae, cephalopod larvae, heteropods, mollusk eggs, pteropods, appendicularians, salps, unidentified fish eggs, and unidentified fish larvae

10. If the prey items were in the same compartment in the model, they were added together

The hotspot predator/prey diet matrix is listed below, with the predators listed in the rows and the prey listed in the columns (Table 6-2). This diet matrix was then reduced into fewer compartments using hierarchical cluster analysis (Figure 6-1). The predator groups were the species observed at the hotspot and coldspot in Puerto Rico. At the same time, prey items were species observed in the hotspot and coldspot in addition to the 50 compartments from the Opitz (1996) model. An n x n matrix was created to perform the cluster analysis. Any Opitz compartment species observed in the hotspot or coldspot were removed from the Opitz compartment. The proportion of the diet of that species in the diet of a predator was listed in its prey group. For example, if Species A consumed Species B and Species C at 5% each, then those values were added to the prey compartment for Species B and C. If species B and C were not observed in Puerto Rico, but were part of the diet of Species A, then those values were added to their respective compartment in Opitz's 1996 model.

After the hierarchical cluster analysis, compartments for the models were chosen. Any species that were sampled for CTX estimation were put into their compartment. We wanted to avoid grouping species if we tested them for CTX estimates. Second, species with ≥90% selective interference confidence interval were put into a compartment. Some species, i.e., *Pomacanthus paru* and *Pomacanthus arcuatus*, had a confidence interval over 90 but weren't made into a separate group. They were grouped at the next highest node with other species with a similar diet. This aggregation allowed the model to be condensed further while allowing species to be still grouped by diet. Third, if species were removed from clustered groups due to them being for CTX and only one species left in a group, that species was added back into its original Opitz (1996) compartment.

Predator groups from the original diet matrix (Table 6-2) were combined into groups chosen from the hierarchical cluster analysis (Figure 6-1) using a weighted mean. For example, if the cluster analysis grouped species A and species B, and species A had a biomass of 1.0 g m^{-2} and species B had a biomass of 0.5 g m^{-2}, then that predator group would have 66% of its diet from species A and 33% of its diet from species B. The prey groups were simply added together. These data are reflected in the new diet composition matrix for the hotspot (Table 6-3).

To complete the diet composition matrix with *Gambierdiscus* spp. the weight of the total number of cells g ww^{-1} algae was divided by the biomass (g m^{-2}) of the benthic autotrophs in the model for a proportion of dinoflagellates to benthic autotrophs. We followed the assumption that the *Gambierdiscus* spp. are being consumed with the benthic autotrophs. I did not add the prey data for *Gambierdiscus* spp. in the final matrices due to the data changing for each model depending on the species and number of cells g ww^{-1} algae used. The different prey values for *Gambierdiscus* spp. for both hotspot and coldspot final models are in Table 6-11.

Table 6-2 Diet composition matrix of the species sampled in Puerto Rico using Randall (1967) diet data.

Predator	Code	Predator	Code	Predator	Code
Abudefduf saxatilis	F1	*Microspathodon chrysurus*	F40	Small Reef Fish O1	F79
Acanthurus bahianus	F2	*Mulloidichthys martinicus*	F41	Small Reef Fish O2	F80
Acanthurus chirurgus	F3	*Myripristis jacobus*	F42	Small Reef Fish O3	F81
Acanthurus coeruleus	F4	*Ocyurus chrysurus*	F43	Large Scaridae H	F82
Aluterus scriptus	F5	*Pomacanthus arcuatus*	F44	Intermediate Scaridae H	F83
Anisotremus virginicus	F6	*Pomacanthus paru*	F45	Small Scaridae H	F84
Balistes vetula	F7	*Pseudupeneus maculatus*	F46	Blenniidae H	F85
Bodianus rufus	F8	*Pterois volitans*	F47	Small Gobiidae C	F86
Calamus pennatula	F9	*Scarus iseri*	F48	Sea birds	B1
Caranx lugubris	F10	*Scarus taeniopterus*	F49	Squids	I1
Caranx ruber	F11	*Scarus vetula*	F50	Sea turtles	R1
Cephalopholis fulva	F12	*Scomberomorus cavalla*	F51	Octopuses	I2
Chaetodon capistratus	F13	*Scomberomorus regalis*	F52	Lobsters	I3
Chaetodon ocellatus	F14	*Sparisoma aurofrenatum*	F53	Crabs	I4
Chaetodon striatus	F15	*Sparisoma viride*	F54	Shrimps/hermit crabs/stomatopods	I5
Chromis cyanea	F16	*Sphyraena barracuda*	F55	Amphipods/isopods/tanaids/pycnogonids	I6
Clepticus parrae	F17	*Stegastes partitus*	F56	Asteroids	I7
Decapterus punctatus	F18	*Stegastes variabilis*	F57	Echinoids	I8
Elacatinus horsti	F19	*Thalassoma bifasciatum*	F58	Gastropods	I9
Elacatinus xanthiprora	F20	*Xanthichthys ringens*	F59	Chitons/scaphopods	I10
Epinephelus adscensionis	F21	Large Sharks/Rays	F60	Polychaetes/priapuloids/ophiuroids	I11
Fistularia tabacaria	F22	Sharks/Scombrids	F61	Holothurids/sipunculids/echiuroids/hemichordates	I12
Ginglymostoma cirratum	F23	Large Jacks	F62	Bivalves	I13
Gymnothorax moringa	F24	Intermediate Jacks C	F63	Ascidians/barnacles/bryozoans	I14
Haemulon flavolineatum	F25	Small Jacks C	F64	Sponges	I15
Haemulon plumieri	F26	Intermediate Reef Fish C1	F65	Corals/sea anemones	I16
Halichoeres garnoti	F27	Large Intermediate Schooling Fish	F66	Zooplankton	I17
Halichoeres pictus	F28	Intermediate Reef Fish C2	F67	Decomposers/microfauna	I18
Halichoeres poeyi	F29	Hemiramphidae	F68	Phytoplankton	A1
Holacanthus ciliaris	F30	Kyphosidae	F69	Benthic autotrophs	A2
Holacanthus tricolor	F31	Intermeduate Reef Fish H	F70	Detritus	D1
Holocentrus rufus	F32	Large Reef Fish C	F71		
Hypoplectrus puella	F33	Intermediate Reef Fish C3	F72		
Lachnolaimus maximus	F34	Small Reef Fish C1	F73		
Lutjanus analis	F35	Small Schooling Fish	F74		
Lutjanus apodus	F36	Engraulidae	F75		
Lutjanus jocu	F37	Small Reef Fish C2	F76		
Malacanthus plumieri	F38	Large Groupers	F77		
Melichthys niger	F39	Intermediate Reef Fish O	F78		

Predator/Prey Matrix	Code	F1	F2	F3	F4	F5	F6
Abudefduf saxatilis	**F1**	0	0	0	0	0	0
Acanthurus bahianus	**F2**	0	0	0	0	0	0
Acanthurus chirurgus	**F3**	0	0	0	0	0	0
Acanthurus coeruleus	**F4**	0	0	0	0	0	0
Aluterus scriptus	**F5**	0	0	0	0	0	0
Anisotremus virginicus	**F6**	0	0	0	0	0	0
Balistes vetula	**F7**	0.000376	0.000376	0.000376	0.000376	0.000376	0.000376
Bodianus rufus	**F8**	0	0	0	0	0	0
Calamus pennatula	**F9**	0	0	0	0	0	0
Caranx lugubris	**F10**	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158
Caranx ruber	**F11**	0	0	0	0.053509	0.009937	0
Cephalopholis fulva	**F12**	0	0.021905	0.021905	0.021905	0	0
Chaetodon capistratus	**F13**	0	0	0	0	0	0
Chaetodon ocellatus	**F14**	0	0	0	0	0	0
Chaetodon striatus	**F15**	0	0	0	0	0	0
Chromis cyanea	**F16**	0	0	0	0	0	0
Clepticus parrae	**F17**	0	0	0	0	0	0
Decapterus punctatus	**F18**	0	0	0	0	0	0
Elacatinus horsti	**F19**	0	0	0	0	0	0
Elacatinus xanthiprora	**F20**	0	0	0	0	0	0
Epinephelus adscensionis	**F21**	0	0	0	0	0	0
Fistularia tabacaria	**F22**	0	0	0	0	0	0
Ginglymostoma cirratum	**F23**	0	0.098889	0.098889	0.098889	0	0
Gymnothorax moringa	**F24**	0	0	0	0	0	0
Haemulon flavolineatum	**F25**	0	0	0	0	0	0
Haemulon plumieri	**F26**	0.001087	0.001087	0.001087	0.001087	0	0.001087
Halichoeres garnoti	**F27**	0	0	0	0	0	0
Halichoeres pictus	**F28**	0	0	0	0	0	0
Halichoeres poeyi	**F29**	0	0	0	0	0	0
Holacanthus ciliaris	**F30**	0	0	0	0	0	0
Holacanthus tricolor	**F31**	0	0	0	0	0	0
Holocentrus rufus	**F32**	0	0	0	0	0	0
Hypoplectrus puella	**F33**	0	0	0	0	0	0
Lachnolaimus maximus	**F34**	0	0	0	0	0	0
Lutjanus analis	**F35**	0	0.020479	0	0	0.003924	0.002032
Lutjanus apodus	**F36**	0	0	0	0	0	0
Lutjanus jocu	**F37**	0	0	0	0	0	0
Malacanthus plumieri	**F38**	0	0	0	0	0	0
Melichthys niger	**F39**	0.000646	0.000646	0.000646	0	0	0.000646

Code	**F7**	**F8**	**F9**	**F10**	**F11**	**F12**	**F13**	**F14**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0	0.000376	0.000376	0	0	0	0.000376	0.000376
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158
F11	0	0	0	0	0	0	0	0
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0	0	0	0	0	0.001087	0.001087	0.001087
F27	0	0	0	0	0	0	0.004357	0
F28	0	0	0	0	0	0	0.003531	0
F29	0	0	0	0	0	0	0.003531	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0	0	0	0	0	0.000387	0.000387
F33	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0.020479	0	0
F36	0	0.040467	0	0	0	0.002698	0	0
F37	0	0	0	0	0	0.014452	0	0
F38	0	0	0	0	0	0	0	0
F39	0	0	0	0	0	0.000646	0.000646	0.000646

Code	**F15**	**F16**	**F17**	**F18**	**F19**	**F20**	**F21**	**F22**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0.000376	0.000376	0.000376	0.000376	0.000376	0.000376	0.000376	0
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158	0
F11	0	0	0	0	0	0	0	0
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0.001087	0.001087	0.001087	0.001087	0.001087	0.001087	0	0
F27	0	0.004357	0	0	0.004357	0.004357	0	0
F28	0	0.003531	0	0	0.003531	0.003531	0	0
F29	0	0.003531	0	0	0.003531	0.003531	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0.000387	0.000387	0	0	0.000387	0.000387	0	0
F33	0	0	0	0	0.006667	0.006667	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0	0	0	0.004586	0	0	0.020479
F36	0	0	0	0	0	0	0.002698	0
F37	0	0	0.028905	0	0	0	0	0
F38	0	0	0	0	0	0	0	0
F39	0.000646	0.000646	0.000646	0.000646	0.000646	0.000646	0	0

Code	**F23**	**F24**	**F25**	**F26**	**F27**	**F28**	**F29**	**F30**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0	0.000376	0.000376	0.000376	0.000376	0.000376	0.000376	0.000376
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.013158	0	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158
F11	0	0	0	0	0	0	0	0
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0	0.001087	0.001087	0	0.001087	0.001087	0.001087	0
F27	0	0	0	0	0	0.004357	0	0
F28	0	0	0	0	0.003531	0.003531	0	0
F29	0	0	0	0	0.003531	0.003531	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0	0	0	0.000387	0.000387	0.000387	0
F33	0	0	0	0	0	0.006667	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0	0.002032	0.002032	0.02028	0	0	0
F36	0	0.040467	0.004047	0.004047	0	0	0	0
F37	0	0.028905	0.003212	0.028905	0	0	0	0
F38	0	0	0	0	0.0124	0.0124	0.0124	0
F39	0	0.000646	0.000646	0	0.000646	0.000646	0.000646	0

Code	**F31**	**F32**	**F33**	**F34**	**F35**	**F36**	**F37**	**F38**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0.000376	0.000376	0.000376	0	0	0	0	0.000376
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.013158	0.013158	0.013158	0.013158	0.013158	0.013158	0	0.013158
F11	0	0	0	0	0	0	0	0
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0.001087	0.001087	0.001087	0	0	0	0	0.001087
F27	0	0	0.004357	0	0	0	0	0
F28	0	0	0.003531	0	0	0	0	0
F29	0	0	0.003531	0	0	0	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0	0.000387	0	0	0	0	0
F33	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0.020479
F36	0	0	0.002698	0	0	0	0	0
F37	0.028905	0.028905	0	0	0	0	0	0
F38	0	0	0	0	0	0	0	0
F39	0	0.000646	0.000646	0	0	0	0	0.000646

Code	**F39**	**F40**	**F41**	**F42**	**F43**	**F44**	**F45**	**F46**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0.000376	0.000376	0.000376	0.000376	0.000376	0	0.000376	0.000376
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.013158	0.013158	0.013158	0.013158	0.013158	0	0.013158	0.013158
F11	0	0	0.053509	0	0	0	0	0.053509
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0.5
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0.001087	0.001087	0.001087	0.001087	0.001087	0	0	0.001087
F27	0	0	0	0	0	0	0	0
F28	0	0	0	0	0	0	0	0
F29	0	0	0	0	0	0	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0	0	0	0	0	0	0
F33	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0.020479
F36	0	0	0	0	0	0	0	0
F37	0	0	0	0	0	0	0	0.028905
F38	0	0	0	0	0	0	0	0
F39	0	0.000646	0.000646	0.000646	0.000646	0	0	0.000646

Code	**F47**	**F48**	**F49**	**F50**	**F51**	**F52**	**F53**	**F54**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0.0003758	0.0003758	0.0003758	0	0	0	0.0003758	0.0003758
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.0131579	0.0131579	0.0131579	0.0131579	0	0.0131579	0.0131579	0.0131579
F11	0	0.0535088	0.0111477	0.0111477	0	0	0.0535088	0.0535088
F12	0	0.0219048	0	0	0	0	0.0219048	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0.0201	0.0201
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0	0.0010873	0.0010873	0	0	0	0.0010873	0
F27	0	0	0	0	0	0	0	0
F28	0	0.0035308	0	0	0	0	0	0
F29	0	0.0035308	0	0	0	0	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0.0003871	0	0	0	0	0.0003871	0
F33	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0.0030961	0.0030961	0	0	0	0.0030961	0.0030961
F36	0.0080933	0.0202333	0.0202333	0	0	0	0.0404667	0.0101167
F37	0	0.0086714	0.0086714	0.0086714	0	0	0.0086714	0.0375762
F38	0	0	0	0	0	0	0	0
F39	0	0.0006458	0	0	0	0	0.0006458	0

Code	**F55**	**F56**	**F57**	**F58**	**F59**	**F60**	**F61**	**F62**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0	0.0003758	0.0003758	0.0003758	0.0003758	0	0	0
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0	0.0131579	0.0131579	0.0131579	0.0131579	0	0.0131579	0
F11	0	0	0	0	0	0	0	0
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0	0.0010873	0.0010873	0.0010873	0.0010873	0	0	0
F27	0	0.0043567	0.0043567	0.0043567	0	0	0	0
F28	0	0.0035308	0.0035308	0.0035308	0	0	0	0
F29	0	0.0035308	0.0035308	0.0035308	0	0	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0.0003871	0.0003871	0.0003871	0.0003871	0	0	0
F33	0	0.0066667	0.0066667	0	0	0	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0
F36	0	0	0	0	0	0	0	0
F37	0	0	0	0	0.0289048	0	0	0
F38	0	0	0	0	0	0	0	0
F39	0	0.0006458	0.0006458	0.0006458	0.0006458	0	0	0

Code	**F63**	**F64**	**F65**	**F66**	**F67**	**F68**	**F69**	**F70**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0	0	0	0.0003758	0	0.0003758	0.0003758	0.0003758
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579
F11	0	0	0	0.0535088	0	0	0	0
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0.0988889	0	0	0	0
F24	0	0	0	0	1	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0	0.0010873	0	0.0010873	0.0010873	0.0010873	0	0.0010873
F27	0	0	0	0	0.0043567	0	0	0
F28	0	0	0	0	0.0035308	0	0	0
F29	0	0	0	0	0.0035308	0	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0	0	0.0003871	0.0003871	0.0003871	0	0.0003871
F33	0	0	0	0	0.0066667	0	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0	0.0034509	0	0.0516985	0	0	0
F36	0	0	0	0	0.0283267	0	0	0
F37	0	0	0.0032116	0.0289048	0.0770794	0	0	0
F38	0	0	0	0	0	0	0	0
F39	0	0.0006458	0	0.0006458	0.0006458	0.0006458	0	0.0006458

Code	**F71**	**F72**	**F73**	**F74**	**F75**	**F76**	**F77**	**F78**
F1	0	0	0	0.0511667	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0	0.0003758	0.0003758	0	0.0003758	0	0	0.0003758
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579	0	0.0131579
F11	0	0	0	0.1043421	0.1043421	0	0	0.0099373
F12	0	0.0657143	0.1314286	0	0	0	0	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0.0988889	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0	0.0010873	0.0010873	0.0010873	0.0010873	0.0010873	0	0.0010873
F27	0	0	0.0043567	0.0043567	0.0043567	0.0043567	0	0
F28	0	0	0.0035308	0.0035308	0.0035308	0.0035308	0	0
F29	0	0	0.0035308	0.0035308	0.0035308	0.0035308	0	0
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0.0003871	0.0003871	0.0003871	0.0003871	0.0003871	0	0
F33	0	0	0.0066667	0.0066667	0.0066667	0.0066667	0	0
F34	0	0	0	0	0	0	0	0
F35	0	0.0218985	0.0244528	0	0	0	0	0.0039239
F36	0	0.1470289	0.0269778	0.0809333	0	0.0161867	0	0
F37	0	0.1043783	0	0.0578095	0	0	0	0
F38	0	0.0372	0.0372	0	0	0.0124	0	0
F39	0	0.0006458	0.0006458	0.0006458	0.0006458	0.0006458	0	0

Code	**F79**	**F80**	**F81**	**F82**	**F83**	**F84**	**F85**	**F86**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0
F6	0	0	0	0	0	0	0	0
F7	0.0003758	0.0003758	0.0003758	0.0003758	0.0003758	0.0003758	0.0003758	0.0003758
F8	0	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0	0
F10	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579	0.0131579
F11	0.0244612	0.0607707	0.0535088	0	0.035354	0.0111477	0.0535088	0
F12	0.0657143	0.0657143	0	0	0	0	0.0219048	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0	0
F15	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0
F21	0.1005	0	0	0	0.0402	0.0201	0	0
F22	0.5	0	0	0	0	0	0	0
F23	0.0988889	0	0	0.0988889	0.0988889	0.0988889	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0.0010873	0.0010873	0.0010873	0	0.0010873	0.0010873	0.0010873	0.0010873
F27	0.0043567	0.0043567	0.0043567	0	0	0	0.0043567	0.0043567
F28	0.0035308	0.0035308	0.0035308	0	0	0.0035308	0.0035308	0.0035308
F29	0.0035308	0.0035308	0.0035308	0	0	0.0035308	0.0035308	0.0035308
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0.0003871	0.0003871	0.0003871	0	0	0.0003871	0.0003871	0.0003871
F33	0.0066667	0.0066667	0.0066667	0	0	0	0.0066667	0.0066667
F34	0	0	0	0	0	0	0	0
F35	0.0271017	0.0039239	0.0085595	0	0.0113739	0.0030961	0	0.0045861
F36	0.0809333	0	0	0	0.0202333	0.0101167	0	0
F37	0	0	0	0.0086714	0.0260143	0.0086714	0	0
F38	0	0	0	0	0	0	0	0
F39	0.0006458	0.0006458	0.0006458	0	0	0.0006458	0.0006458	0.0006458

Code	**B1**	**I1**	**R1**	**I2**	**I3**	**I4**	**I5**	**I6**
F1	0	0	0	0	0	0	0	0
F2	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0.003
F5	0	0	0	0	0	0	0.003	0
F6	0	0	0	0	0	0.1692857	0.2145714	0.1498571
F7	0	0	0	0	0.0017425	0.0547425	0.0286425	0.0010425
F8	0	0	0	0	0	0.33025	0.12075	0.01325
F9	0	0	0	0	0	0.215	0.168	0
F10	0	0	0	0	0	0	0	0
F11	0	0.014	0	0	0	0.0043333	0.0406667	0
F12	0	0	0	0	0	0.1843333	0.3556667	0
F13	0	0	0	0	0	0	0	0
F14	0	0	0	0	0	0	0.03275	0
F15	0	0	0	0	0	0	0	0.0315
F16	0	0	0	0	0	0	0	0
F17	0	0	0	0	0	0	0	0
F18	0	0	0	0	0	0	0	0
F19	0	0	0	0	0	0	0	0.5
F20	0	0	0	0	0	0	0	0.5
F21	0	0	0	0	0	0.687	0.064	0
F22	0	0	0	0	0	0	0	0
F23	0	0.055	0	0.055	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0.0332	0	0.1596	0.057	0.0522
F26	0	0	0	0	0	0.2737262	0.0717262	0.0349762
F27	0	0	0	0	0	0.2777778	0.1527778	0
F28	0	0	0	0	0	0.2709247	0.0789247	0.0289247
F29	0	0	0	0	0	0.2709247	0.0789247	0.0289247
F30	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0
F32	0	0	0	0	0	0.573	0.162	0.019
F33	0	0	0	0	0	0.211	0.563	0.126
F34	0	0	0	0	0	0.061	0.049	0.01
F35	0	0	0	0.0316128	0.0196128	0.4446128	0.0596128	0
F36	0	0	0	0.0345	0	0.242	0.1115	0
F37	0	0.022	0	0.07	0.0885	0.1585	0	0
F38	0	0	0	0	0	0.185	0.164	0.016
F39	0	0	0	0	0	0.0293333	0.0198333	0

Code	**I7**	**I8**	**I9**	**I10**	**I11**	**I12**	**I13**	**I14**
F1	0	0	0.0591667	0	0.0151667	0	0	0.1071667
F2	0	0	0	0	0	0	0	0
F3	0	0	0.002	0	0.002	0	0	0
F4	0	0	0	0	0	0	0	0
F5	0	0	0.006	0	0	0	0	0.011
F6	0	0	0.038	0.01	0.305	0.004	0.055	0.015
F7	0.0143425	0.7283425	0.0163425	0.0013425	0.0543425	0.0093425	0.0463425	0.0063425
F8	0	0.14475	0.10475	0.00775	0.19775	0	0.08075	0
F9	0	0.04	0.082	0	0.282	0.09	0.123	0
F10	0	0	0	0	0	0	0	0
F11	0	0	0.004	0	0	0	0	0
F12	0	0	0	0	0	0	0	0
F13	0	0	0	0	0.356	0	0	0.063
F14	0	0	0	0	0.38975	0	0	0
F15	0	0	0	0	0.587	0	0	0
F16	0	0	0	0	0	0	0	0.339
F17	0	0	0.193	0	0	0	0	0.048
F18	0	0	0.09	0	0	0	0	0
F19	0	0	0	0	0.5	0	0	0
F20	0	0	0	0	0.5	0	0	0
F21	0	0	0.032	0.016	0	0	0	0
F22	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0.0152	0.0052	0.0652	0.4272	0.1522	0.0322	0
F26	0	0.1244762	0.0694762	0.0074762	0.2024762	0.1434762	0.0134762	0
F27	0	0.0327778	0.1377778	0.0227778	0.2127778	0.0077778	0.0727778	0
F28	0	0.0692581	0.2142581	0.0452581	0.1222581	0.0692581	0.0232581	0
F29	0	0.0692581	0.2142581	0.0452581	0.1222581	0.0692581	0.0232581	0
F30	0	0	0	0	0	0	0	0.013
F31	0	0	0	0	0	0	0	0
F32	0	0	0.078	0.006	0.116	0	0	0
F33	0	0	0	0	0	0	0	0
F34	0	0.046	0.397	0.006	0	0	0.426	0.005
F35	0	0	0.130613	0	0	0	0	0
F36	0	0	0.005	0	0	0	0	0
F37	0	0	0.036	0	0	0	0	0
F38	0	0.027	0	0.057	0.323	0.104	0	0
F39	0	0	0.055	0	0	0	0	0.018

Code	**I15**	**I16**	**I17**	**I18**	**A1**	**A2**	**D1**
F1	0	0.4381667	0.2411667	0	0	0.088	0
F2	0	0	0	0	0	0.541	0.459
F3	0	0	0	0	0	0.5265	0.4695
F4	0	0.001	0	0	0	0.532	0.464
F5	0.004	0.544	0	0	0	0.432	0
F6	0	0	0.0342857	0.005	0	0	0
F7	0	0.0025425	0	0	0	0.012	0
F8	0	0	0	0	0	0	0
F9	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0
F11	0	0	0.0728333	0	0	0	0
F12	0	0	0	0	0	0	0
F13	0	0.475	0.106	0	0	0	0
F14	0	0.51375	0.06375	0	0	0	0
F15	0	0.325	0.0565	0	0	0	0
F16	0	0	0.661	0	0	0	0
F17	0	0	0.759	0	0	0	0
F18	0	0	0.91	0	0	0	0
F19	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0
F26	0	0	0	0	0	0	0
F27	0	0	0	0	0	0	0
F28	0	0	0	0	0	0	0
F29	0	0	0	0	0	0	0
F30	0.968	0.005	0	0	0	0.014	0
F31	0.971	0.021	0	0	0	0.008	0
F32	0	0	0.034	0	0	0	0
F33	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0
F36	0	0	0	0	0	0	0
F37	0	0	0.018	0	0	0	0
F38	0	0	0	0	0	0	0
F39	0	0.006	0.0878333	0.001	0	0.752	0

Predator/Prey Matrix	**Code**	**F1**	**F2**	**F3**	**F4**	**F5**	**F6**
Microspathodon chrysurus	**F40**	0	0	0	0	0	0
Mulloidichthys martinicus	**F41**	0	0	0	0	0	0
Myripristis jacobus	**F42**	0	0	0	0	0	0
Ocyurus chrysurus	**F43**	0	0	0	0	0	0
Pomacanthus arcuatus	**F44**	0	0	0	0	0	0
Pomacanthus paru	**F45**	0	0	0	0	0	0
Pseudupeneus maculatus	**F46**	0	0	0	0	0	0
Pterois volitans	**F47**	0	0	0	0	0	0
Scarus iseri	**F48**	0	0	0	0	0	0
Scarus taeniopterus	**F49**	0	0	0	0	0	0
Scarus vetula	**F50**	0	0	0	0	0	0
Scomberomorus cavalla	**F51**	0	0	0	0	0	0
Scomberomorus regalis	**F52**	0	0	0	0	0	0.1281333
Sparisoma aurofrenatum	**F53**	0	0	0	0	0	0
Sparisoma viride	**F54**	0	0	0	0	0	0
Sphyraena barracuda	**F55**	0	0.0502632	0	0	0	0
Stegastes partitus	**F56**	0	0	0	0	0	0
Stegastes variabilis	**F57**	0	0	0	0	0	0
Thalassoma bifasciatum	**F58**	0	0	0	0	0	0
Xanthichthys ringens	**F59**	0	0	0	0	0	0
Large Sharks/Rays	**F60**	0.0002564	0.0052	0.0052	0.0052	0.0028571	0.0002564
Sharks/Scombrids	**F61**	0.0010256	0.002	0.002	0.002	0.0008571	0.0010256
Large jacks	**F62**	0.0041026	0.0044	0.0044	0.0044	0	0.0041026
Intermediate jacks C	**F63**	0.0028205	0.0107	0.0107	0.0107	0.0007143	0.0028205
Small jacks C	**F64**	0	0.002	0.002	0.002	0	0
Intermediate reef fish C1	**F65**	0	0	0	0	0	0
Large intermediate schooling fish	**F66**	0	0.001	0.001	0.001	0	0
Intermediate reef fish C2	**F67**	0.0005128	0.0004	0.0004	0.0004	0.0004286	0.0005128
Hemiramphidae	**F68**	0	0	0	0	0	0
Kyphosidae	**F69**	0	0	0	0	0	0
Intermediate reef fish H	**F70**	0	0	0	0	0	0
Large reef fish C	**F71**	0.0010256	0.0023	0.0023	0.0023	0.0004286	0.0010256
Intermediate reef fish C3	**F72**	0.0002564	0.0009	0.0009	0.0009	0.0004286	0.0002564
Small reef fish C1	**F73**	5.128E-05	0	0	0	0	5.128E-05
Small schooling fish	**F74**	0	0	0	0	0	0
Engraulidae	**F75**	0	0	0	0	0	0
Small reef fish C2	**F76**	5.128E-05	0.0001	0.0001	0.0001	0	5.128E-05
Large groupers	**F77**	0.0023077	0.002	0.002	0.002	0.0014286	0.0023077
Intermediate reef fish O	**F78**	0	0	0	0	0	0
Small reef fish O1	**F79**	7.692E-05	0	0	0	0	7.692E-05

Code	**F7**	**F8**	**F9**	**F10**	**F11**	**F12**	**F13**	**F14**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0.0139849	0.1538333	0	0	0
F52	0	0.0080083	0	0	0.0640667	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0	0.0045694	0.0502632	0.0045694	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0	0	0	0	0	0	0
F59	0	0	0	0	0	0	0	0
F60	0.0083333	0.0006944	0.0002564	0.00625	0.00625	0.0006944	0.0002564	0.0002564
F61	0.00125	0.0023333	0.0010256	0.00425	0.00425	0.0023333	0.0010256	0.0010256
F62	0.0166667	0.0034444	0.0041026	0.01875	0.01875	0.0034444	0.0041026	0.0041026
F63	8.333E-05	0	0.0028205	0	0	0	0.0028205	0.0028205
F64	0	0	0	0.00025	0.00025	0	0	0
F65	0	0	0	0	0	0	0	0
F66	0	0	0	0	0	0	0	0
F67	0	0.0004167	0.0005128	0.000125	0.000125	0.0004167	0.0005128	0.0005128
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0.0025	0.0011111	0.0010256	0.003	0.003	0.0011111	0.0010256	0.0010256
F72	0.0008333	0.0003333	0.0002564	0	0	0.0003333	0.0002564	0.0002564
F73	0	0	5.128E-05	0	0	0	5.128E-05	5.128E-05
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0	0	5.128E-05	0	0	0	5.128E-05	5.128E-05
F77	0.0070833	0	0.0023077	0.00125	0.00125	0	0.0023077	0.0023077
F78	0	0	0	0	0	0	0	0
F79	0	0	7.692E-05	0	0	0	7.692E-05	7.692E-05

Code	**F15**	**F16**	**F17**	**F18**	**F19**	**F20**	**F21**	**F22**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0	0.032148	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0.013985	0	0	0	0
F52	0	0.064067	0.064067	0	0	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0	0.029701	0	0	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0.008198	0	0	0.008198	0.008198	0	0
F59	0	0	0	0	0	0	0	0
F60	0.000256	0.000690	0.000694	0.001	0	0	0.000694	0.002308
F61	0.001026	0.000690	0.002333	0.0125	0	0	0.002333	0.001077
F62	0.004103	0	0.003444	0	0	0	0.003444	0
F63	0.002821	0	0	0	0	0	0	0
F64	0	0	0	0.00025	0	0	0	0
F65	0	0	0	0	0	0	0	0
F66	0	0	0	0	0	0	0	0
F67	0.000513	0	0.000417	0	0	0	0.000417	0.000077
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0.001026	0.000034	0.001111	0.005000	0	0	0.001111	0.000385
F72	0.000256	0.000345	0.000333	0.000750	0	0	0.000333	0
F73	0.000051	0.000103	0	0	0	0	0	0
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0.000051	0.000276	0	0	0	0	0	0
F77	0.002308	0	0	0.0025	0	0	0	0.017846
F78	0	0	0	0	0	0	0	0
F79	0.000077	0.000138	0	0	0	0	0	0

Code	**F23**	**F24**	**F25**	**F26**	**F27**	**F28**	**F29**	**F30**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0	0.0857263	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0	0	0	0
F52	0	0	0	0	0.0080083	0	0.0080083	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0.0045694	0.0045694	0	0	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0	0	0	0	0.0081977	0	0
F59	0	0	0	0	0	0	0	0
F60	0.0007692	0.0006944	0.0002564	0.0002564	0.0006897	0	0.0006897	0.0028571
F61	0	0.0023333	0.0010256	0.0010256	0.0006897	0	0.0006897	0.0008571
F62	0	0.0034444	0.0041026	0.0041026	0	0	0	0
F63	0	0	0.0028205	0.0028205	0	0	0	0.0007143
F64	0	0	0	0	0	0	0	0
F65	0	0	0	0	0	0	0	0
F66	0	0	0	0	0	0	0	0
F67	0	0.0004167	0.0005128	0.0005128	0	0	0	0.0004286
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0	0.0011111	0.0010256	0.0010256	3.448E-05	0	3.448E-05	0.0004286
F72	0	0.0003333	0.0002564	0.0002564	0.0003448	0	0.0003448	0.0004286
F73	0	0	5.128E-05	5.128E-05	0.0001035	0	0.0001035	0
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0	0	5.128E-05	5.128E-05	0.0002759	0	0.0002759	0
F77	0.0003846	0	0.0023077	0.0023077	0	0	0	0.0014286
F78	0	0	0	0	0	0	0	0
F79	0	0	7.692E-05	7.692E-05	0.0001379	0	0.0001379	0

Code	F31	F32	F33	F34	F35	F36	F37	F38
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0	0	0	0
F52	0	0	0	0	0	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0	0	0	0	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0	0.008198	0	0	0	0	0
F59	0	0	0	0	0	0	0	0
F60	0.002857	0.000256	0.000690	0.002308	0.008333	0.008333	0.002308	0.000694
F61	0.000857	0.001026	0.000690	0.001077	0.001250	0.001250	0.001077	0.002333
F62	0	0.004103	0	0	0.016667	0.016667	0	0.003444
F63	0.000714	0.002821	0	0	0.000083	0.000083	0	0
F64	0	0	0	0	0	0	0	0
F65	0	0	0	0	0	0	0	0
F66	0	0	0	0	0	0	0	0
F67	0.000429	0.000513	0	0.000077	0	0	0.000077	0.000417
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0.000429	0.001026	0.000034	0.000385	0.0025	0.0025	0.000385	0.001111
F72	0.000429	0.000256	0.000345	0	0.000833	0.000833	0	0.000333
F73	0	0.000051	0.000103	0	0	0	0	0
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0	0.000051	0.000276	0	0	0	0	0
F77	0.001429	0.002308	0	0.017846	0.007083	0.007083	0.017846	0
F78	0	0	0	0	0	0	0	0
F79	0	0.000077	0.000138	0	0	0	0	0

Code	F39	F40	F41	F42	F43	F44	F45	F46
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0.153833	0	0	0
F52	0	0	0	0	0	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0	0	0.050263	0	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0	0	0	0	0	0	0
F59	0	0	0	0	0	0	0	0
F60	0.0052	0.0052	0.000256	0.000256	0.008333	0.008333	0.002857	0.000256
F61	0.002	0.002	0.001026	0.001026	0.001250	0.001250	0.000857	0.001026
F62	0.0044	0.0044	0.004103	0.004103	0.016667	0.016667	0	0.004103
F63	0.0107	0.0107	0.002821	0.002821	0.000083	0.000083	0.000714	0.002821
F64	0.002	0.002	0	0	0	0	0	0
F65	0	0	0	0	0	0	0	0
F66	0.001	0.001	0	0	0	0	0	0
F67	0.0004	0.0004	0.0005128	0.0005128	0	0	0.0004286	0.0005128
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0.0023	0.0023	0.001026	0.001026	0.0025	0.0025	0.000429	0.001026
F72	0.0009	0.0009	0.000256	0.000256	0.000833	0.000833	0.000429	0.000256
F73	0	0	0.000051	0.000051	0	0	0	0.000051
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0.0001	0.0001	0.000051	0.000051	0	0	0	0.000051
F77	0.002	0.002	0.002308	0.002308	0.007083	0.007083	0.001429	0.002308
F78	0	0	0	0	0	0	0	0
F79	0	0	0.000077	0.000077	0	0	0	0.000077

Code	**F47**	**F48**	**F49**	**F50**	**F51**	**F52**	**F53**	**F54**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0	0	0	0
F52	0	0	0	0	0	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0.0050263	0.0050263	0.0050263	0	0	0.0050263	0.0050263
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0	0	0	0	0	0	0
F59	0	0	0	0	0	0	0	0
F60	0	0.0003333	0.0003333	0.0083333	0.0016667	0.0016667	0.0003333	0.0003333
F61	0	0.0006667	0.0001667	0.01	0.0016667	0.0016667	0.0006667	0.0001667
F62	0	0	0	0	0	0	0	0
F63	0	0.0406667	0.0121667	0.0053333	0	0	0.0406667	0.0121667
F64	0	0	0	0	0	0	0	0
F65	0	0	0	0	0	0	0	0
F66	0	0	0	0	0	0	0	0
F67	0	0.0033333	0.005	0.0066667	0	0	0.0033333	0.005
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0.0006667	0	0	0	0	0.0006667
F71	0	0.0003333	0.0013333	0.0033333	0	0	0.0003333	0.0013333
F72	0	0.002	0.0011667	0.0183333	0	0	0.002	0.0011667
F73	0	0.0006667	0	0	0	0	0.0006667	0
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0	0	0	0	0	0	0	0
F77	0	0	0	0.0826667	0.0008333	0.0008333	0	0
F78	0	0	0	0	0	0	0	0
F79	0	0.0013333	0	0	0	0	0.0013333	0

Code	**F55**	**F56**	**F57**	**F58**	**F59**	**F60**	**F61**	**F62**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0.0857263	0	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0	0	0	0
F52	0	0	0	0.0080083	0	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0	0	0	0	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0.0081977	0.0081977	0	0	0	0	0
F59	0	0	0	0	0	0	0	0
F60	0.0023077	0	0.005	0.0006897	0.0006944	0.0092308	0.0066667	0.003
F61	0.0010769	0	0.0028	0.0006897	0.0023333	0	0.0066667	0
F62	0	0	0	0	0.0034444	0	0	0
F63	0	0	0.0106	0	0	0	0	0
F64	0	0	0	0	0	0	0	0
F65	0	0	0	0	0	0	0	0
F66	0	0	0	0	0	0	0	0
F67	7.692E-05	0	0	0	0.0004167	0	0	0
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0.0003846	0	0.0034	3.448E-05	0.0011111	0	0	0.004
F72	0	0	0.008	0.0003448	0.0003333	0	0	0
F73	0	0	0.001	0.0001035	0	0	0	0
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0	0	0.0002	0.0002759	0	0	0	0
F77	0.0178462	0	0	0	0	0.0046154	0.0033333	0
F78	0	0	0	0	0	0	0	0
F79	0	0	0.0012	0.0001379	0	0	0	0

Code	**F63**	**F64**	**F65**	**F66**	**F67**	**F68**	**F69**	**F70**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0.0839091	0.0279697	0	0.3216515	0	0	0	0
F52	0	0.0640667	0	0.11532	0	0.0640667	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0.0731101	0.0342703	0.0045694	0.1553589	0.132512	0.0502632	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0	0	0	0.0081977	0	0	0
F59	0	0	0	0	0	0	0	0
F60	0.0375	0.003	0.0583333	0.01	0.0069231	0.002	0.06	0.026
F61	0.0255	0.0375	0.00875	0.176	0.0276923	0.115	0.02	0.01
F62	0.1125	0	0.1166667	0	0.1107692	0	0	0.022
F63	0	0	0.0005833	0.056	0.0761538	0	0.015	0.0535
F64	0.0015	0.00075	0	0.014	0	0	0	0.01
F65	0	0	0.001	0	0.009	0	0.003	0.004
F66	0	0	0	0.003	0	0	0	0.005
F67	0.00075	0	0	0.001	0.0138462	0	0.003	0.002
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0.018	0.015	0.0175	0.04	0.0276923	0	0.003	0.0115
F72	0	0.00225	0.0058333	0.002	0.0069231	0	0.003	0.0045
F73	0	0	0	0.002	0.0013846	0.002	0	0
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0	0	0	0	0.0013846	0	0	0.0005
F77	0.0075	0.0075	0.0495833	0	0.0623077	0	0.02	0.01
F78	0	0	0	0	0	0	0	0
F79	0	0	0	0.003	0.0020769	0.003	0	0

Code	**F71**	**F72**	**F73**	**F74**	**F75**	**F76**	**F77**	**F78**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0.0782778	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0
F47	0	0	0.0321483	0	0	0.1714746	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0.1538333	0	0	0
F52	0	0.0640667	0.024025	0.2050133	0.0640667	0.0080083	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0.0548325	0	0.1507895	0	0	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0	0	0.0081977	0.0081977	0.0081977	0.0081977	0	0
F59	0	0	0	0	0	0	0	0
F60	0.0207692	0.0201389	0.0165517	0.049	0.028	0.006	0.093	0.0085714
F61	0.0096923	0.0676667	0.0165517	0.136	0.04	0.02	0	0.0025714
F62	0	0.0998889	0	0	0	0	0	0
F63	0	0	0	0.11	0.107	0.001	0	0.0021429
F64	0	0	0	0.006	0.019	0	0	0
F65	0	0.057	0.005	0.062	0	0.003	0	0.003
F66	0	0	0	0.047	0.006	0	0	0
F67	0.0006923	0.0120833	0	0.067	0.002	0.001	0	0.0012857
F68	0	0	0	0.2	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0	0	0	0
F71	0.0034615	0.0322222	0.0008276	0.059	0.001	0	0	0.0012857
F72	0	0.0096667	0.0082759	0.002	0.002	0.01	0	0.0012857
F73	0	0	0.0024828	0.002	0.002	0.002	0	0
F74	0	0	0	0.002	0.002	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0	0	0.0066207	0	0	0.003	0	0
F77	0.1606154	0	0	0	0	0	0	0.0042857
F78	0	0	0	0	0	0	0	0
F79	0	0	0.0033103	0.004	0.004	0.004	0	0

Code	**F79**	**F80**	**F81**	**F82**	**F83**	**F84**	**F85**	**F86**
F40	0	0	0	0	0	0	0	0
F41	0	0	0	0	0	0	0	0
F42	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0.046	0	0	0	0	0
F47	0	0	0.2036002	0	0	0	0	0.0321483
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0	0	0	0
F52	0	0	0	0	0	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0.0502632	0	0.0050263	0.015079	0.0050263	0	0
F56	0	0	0	0	0	0	0	0
F57	0	0	0	0	0	0	0	0
F58	0.0081977	0.0081977	0.0081977	0	0	0	0.0081977	0.0081977
F59	0	0	0	0	0	0	0	0
F60	0.02	0.004	0.001	0.0166667	0.0013333	0.0003333	0	0
F61	0.0112	0.001	0	0.02	0.0006667	0.0006667	0.001	0
F62	0	0	0	0	0	0	0	0
F63	0.0424	0.053	0.053	0.0106667	0.0486667	0.0406667	0.053	0
F64	0	0	0	0	0	0	0	0
F65	0.02	0	0	0.03	0.016	0.016	0	0
F66	0	0	0	0	0	0	0	0
F67	0	0	0	0.0133333	0.02	0.0033333	0	0
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0	0	0	0	0.0026667	0	0	0
F71	0.0136	0.006	0.01	0.0066667	0.0053333	0.0003333	0	0
F72	0.032	0.001	0.015	0.0366667	0.0046667	0.002	0.002	0.004
F73	0.004	0.002	0.006	0	0	0.0006667	0.006	0.002
F74	0	0	0	0	0	0	0	0
F75	0	0	0	0	0	0	0	0
F76	0.0008	0	0.004	0	0	0	0.001	0.002
F77	0	0	0	0.1653333	0	0	0	0
F78	0	0	0	0	0	0	0	0
F79	0.0048	0.004	0.004	0	0	0.0013333	0.004	0.003

Code	**B1**	**I1**	**R1**	**I2**	**I3**	**I4**	**I5**	**I6**
F40	0	0	0	0	0	0	0.02575	0.01475
F41	0	0	0	0	0	0.0814286	0.0698571	0.1117857
F42	0	0	0	0	0	0.0771546	0.1541546	0.0551091
F43	0	0.0217778	0	0	0	0.2008278	0.0915778	0.0077778
F44	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0.3154286	0.2738571	0.0732857
F47	0	0	0	0	0	0.0714	0.214	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0.077	0	0	0	0	0	0
F52	0	0.023	0	0	0	0	0.016	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0	0.026	0	0	0	0
F56	0	0	0	0	0	0	0.026625	0.144125
F57	0	0	0	0	0	0	0.026625	0.144125
F58	0	0	0	0	0	0.077955	0.06333	0.091955
F59	0	0	0	0	0	0	0	0.5
F60	0.02	0.005	0.005	0.02	0.005	0.05	0.05	0
F61	0	0.01	0	0.045	0.005	0.02	0.05	0
F62	0	0	0	0	0	0.01	0.015	0
F63	0	0.004	0	0	0	0.004	0.031	0
F64	0	0	0	0	0	0	0	0
F65	0	0.001	0	0.014	0	0.09	0.03	0
F66	0	0	0	0	0	0	0.004	0
F67	0	0	0	0.05	0.001	0.1	0.02	0.031
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0.001	0	0.002	0	0	0	0	0
F71	0	0.001	0	0.009	0.002	0.032	0.03	0
F72	0	0	0	0.03	0	0.15	0.1	0.042
F73	0	0	0	0	0	0.1	0.04	0.042
F74	0	0	0	0	0	0	0	0.01
F75	0	0	0	0	0	0	0	0
F76	0	0	0	0.02	0	0.063	0.15	0.1
F77	0	0	0.035	0	0.16	0.07	0	0
F78	0	0	0	0	0	0.001	0.002	0
F79	0	0	0	0	0	0.01	0.01	0.056

Code	**I7**	**I8**	**I9**	**I10**	**I11**	**I12**	**I13**	**I14**
F40	0	0	0.00575	0	0.01475	0	0	0
F41	0	0.0055	0.0085	0.0845	0.2765	0.0695	0.1325	0
F42	0	0	0	0	0.0702	0	0	0
F43	0	0	0.0877778	0	0	0	0.0147778	0.0297778
F44	0	0	0	0	0	0	0	0.11025
F45	0	0	0	0	0	0	0	0.05
F46	0	0	0.011	0	0.144	0.047	0.07	0
F47	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0	0	0
F49	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0
F51	0	0	0	0	0	0	0	0
F52	0	0	0	0	0	0	0	0
F53	0	0	0	0	0	0	0	0
F54	0	0	0	0	0	0	0	0
F55	0	0	0	0	0	0	0	0
F56	0	0	0.022625	0	0.177125	0	0.017625	0
F57	0	0	0.022625	0	0.177125	0	0.017625	0
F58	0	0.01808	0.06008	0	0.12908	0	0	0.031955
F59	0	0	0	0	0	0	0	0
F60	0	0.008	0.01	0	0.05	0.05	0.067	0
F61	0	0	0.008	0	0.006	0	0.007	0
F62	0	0.082	0.158	0	0	0	0.057	0
F63	0	0	0.004	0	0	0	0	0
F64	0	0	0	0	0	0	0	0
F65	0.001	0.05	0.017	0	0.006	0.003	0.01	0.06
F66	0	0	0	0	0	0.12	0	0
F67	0	0.1	0.035	0.02	0.18	0.06	0.073	0.05
F68	0	0	0	0	0	0	0	0
F69	0	0	0	0	0	0	0	0
F70	0.002	0	0	0	0	0	0	0
F71	0	0.1	0.4	0	0	0	0.075	0
F72	0	0.06	0.004	0.01	0.05	0.01	0.023	0.04
F73	0	0.11	0.144	0.04	0.1	0.008	0.06	0
F74	0	0	0	0	0	0	0	0.03
F75	0	0	0	0	0	0	0	0
F76	0	0	0.005	0.002	0.079	0.001	0.03	0
F77	0	0	0	0	0	0	0	0
F78	0.002	0.003	0	0	0.006	0.006	0	0.023
F79	0	0	0.038	0	0.05	0	0.02	0.02

Code	**I15**	**I16**	**I17**	**I18**	**A1**	**A2**	**D1**
F40	0.00675	0.02675	0	0	0.00675	0.4465	0.45225
F41	0	0	0.1599286	0	0	0	0
F42	0	0	0.6433819	0	0	0	0
F43	0	0	0.4674278	0	0	0	0
F44	0.71025	0.07225	0.02325	0	0	0.084	0
F45	0.748	0.066	0.001	0	0	0.135	0
F46	0	0	0.0194286	0	0	0	0
F47	0	0	0.0714	0	0	0	0
F48	0	0.009	0	0	0	0.991	0
F49	0.015	0	0	0	0	0.985	0
F50	0.01	0.018	0	0	0	0.972	0
F51	0	0	0	0	0	0	0
F52	0	0	0	0	0	0	0
F53	0	0.009	0	0	0	0.991	0
F54	0.001	0.001	0	0	0	0.998	0
F55	0	0	0.019	0	0	0	0
F56	0.028625	0.056625	0	0	0	0.2575	0.269125
F57	0.028625	0.056625	0	0	0	0.2575	0.269125
F58	0	0	0.388205	0	0	0	0
F59	0	0	0.5	0	0	0	0
F60	0	0	0	0	0	0	0.02
F61	0	0	0	0	0	0	0
F62	0	0	0	0	0	0	0
F63	0	0	0.022	0	0	0	0
F64	0	0	0.938	0	0	0	0
F65	0.2	0.01	0.25	0	0	0.029	0
F66	0	0	0.722	0	0	0.088	0
F67	0.07	0.004	0.018	0	0	0.01	0
F68	0	0	0	0	0	0.8	0
F69	0	0	0	0	0	1	0
F70	0	0	0	0	0	0.533	0.458
F71	0	0	0.006	0	0	0	0
F72	0.07	0.003	0.15	0	0	0.052	0
F73	0	0	0.26	0	0	0.058	0
F74	0	0	0.95	0.006	0	0	0
F75	0	0	0.2	0	0.7	0	0.1
F76	0	0	0.5	0	0	0.028	0
F77	0	0	0	0	0	0	0
F78	0.921	0.007	0.001	0	0	0.028	0
F79	0.02	0.02	0.1	0	0	0.326	0.28

Predator/Prey Matrix	**Code**	**F1**	**F2**	**F3**	**F4**	**F5**	**F6**
Small reef fish O2	**F80**	0	0	0	0	0	0
Small reef fish O3	**F81**	0	0	0	0	0	0
Large scaridae H	**F82**	0	0	0	0	0	0
Intermediate scaridae H	**F83**	0	0	0	0	0	0
Small scaridae H	**F84**	0	0	0	0	0	0
Blenniidae H	**F85**	0	0	0	0	0	0
Small gobiidae C	**F86**	0	0	0	0	0	0
Sea birds	**B1**	0.0010256	0.001	0.001	0.001	0.0007143	0.0010256
Squids	**I1**	0	0	0	0	0	0
Sea turtles	**R1**	0	0	0	0	0	0
Octopuses	**I2**	0	0.0002	0.0002	0.0002	0	0.0000513
Lobsters	**I3**	0	0	0	0	0	0
Crabs	**I4**	0	0	0	0	0	0
Shrimps/hermit crabs/stomatopods	**I5**	0.0002564	0	0	0	0	0.0002564
Amphipods/isopods/tanaids/ pycnogonids	**I6**	0	0	0	0	0	0
Asteroids	**I7**	0	0	0	0	0	0
Echinoids	**I8**	0	0	0	0	0	0
Gastropods	**I9**	0	0	0	0	0	0
Chitons/scaphopods	**I10**	0	0	0	0	0	0
Polychaetes/priapuloids/ ophiuroids	**I11**	0	0	0	0	0	0
Holothurids/sipunculids/ echiuroids/hemichordates	**I12**	0	0	0	0	0	0
Bivalves	**I13**	0	0	0	0	0	0
Ascidians/barnacles/bryozoans	**I14**	0	0	0	0	0	0
Sponges	**I15**	0	0	0	0	0	0
Corals/sea anemones	**I16**	0	0	0	0	0	0
Zooplankton	**I17**	0	0	0	0	0	0
Decomposers/microfauna	**I18**	0	0	0	0	0	0
Phytoplankton	**A1**	0	0	0	0	0	0
Benthic autotrophs	**A2**	0	0	0	0	0	0
Detritus	**D1**	0	0	0	0	0	0

Code	F7	F8	F9	F10	F11	F12	F13	F14
F80	0	0	0	0	0	0	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.0008333	0.0008333	0.0010256	0.0075	0.0075	0.0008333	0.0010256	0.0010256
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	0	8.333E-05	5.128E-05	0	0	8.333E-05	5.128E-05	5.128E-05
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0
I5	0	0	0.0002564	0	0	0	0.0002564	0.0002564
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**F15**	**F16**	**F17**	**F18**	**F19**	**F20**	**F21**	**F22**
F80	0	0.0001035	0	0	0	0	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.0010256	0.0008621	0.0008333	0.0075	0	0	0.0008333	0.0003846
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	5.128E-05	0.0002414	8.333E-05	0	0	0	8.333E-05	7.692E-05
I3	0	0	0	0	0	0	0	0
I4	0	0.0003448	0	0	0	0	0	0
I5	0.0002564	0.0003448	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	F23	F24	F25	F26	F27	F28	F29	F30
F80	0	0	0	0	0.0001035	0	0.0001035	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0	0.0008333	0.0010256	0.0010256	0.0008621	0	0.0008621	0.0007143
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	0	8.333E-05	5.128E-05	5.128E-05	0.0002414	0	0.0002414	0
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0	0.0003448	0	0.0003448	0
I5	0	0	0.0002564	0.0002564	0.0003448	0	0.0003448	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**F31**	**F32**	**F33**	**F34**	**F35**	**F36**	**F37**	**F38**
F80	0	0	0.0001035	0	0	0	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.0007143	0.0010256	0.0008621	0.0003846	0.0008333	0.0008333	0.0003846	0.0008333
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	0	5.128E-05	0.0002414	7.692E-05	0	0	7.692E-05	8.333E-05
I3	0	0	0	0	0	0	0	0
I4	0	0	0.0003448	0	0	0	0	0
I5	0	0.0002564	0.0003448	0	0	0	0	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**F39**	**F40**	**F41**	**F42**	**F43**	**F44**	**F45**	**F46**
F80	0	0	0	0	0	0	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.001	0.001	0.0010256	0.0010256	0.0008333	0.0008333	0.0007143	0.0010256
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	0.0002	0.0002	5.128E-05	5.128E-05	0	0	0	5.128E-05
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0
I5	0	0	0.0002564	0.0002564	0	0	0	0.0002564
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	F47	F48	F49	F50	F51	F52	F53	F54
F80	0	0	0	0	0	0	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0	0.0006667	0.0008333	0.0006667	0.0033333	0.0033333	0.0006667	0.0008333
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**F55**	**F56**	**F57**	**F58**	**F59**	**F60**	**F61**	**F62**
F80	0	0	0	0.0001035	0	0	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.0003846	0	0.001	0.0008621	0.0008333	0	0.0133333	0.01
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	7.692E-05	0	0.0004	0.0002414	8.333E-05	0	0	0
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0.0003448	0	0	0	0
I5	0	0	0	0.0003448	0	0	0	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**F63**	**F64**	**F65**	**F66**	**F67**	**F68**	**F69**	**F70**
F80	0	0	0	0	0	0	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.045	0.0225	0.0058333	0.28	0.0276923	0.1	0	0.005
I1	0	0	0	0.15	0	0.015	0	0
R1	0	0	0	0	0	0	0	0
I2	0	0	0	0	0.0013846	0	0	0.001
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0
I5	0	0	0	0	0.0069231	0	0	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**F71**	**F72**	**F73**	**F74**	**F75**	**F76**	**F77**	**F78**
F80	0	0	0.0024828	0.003	0.003	0.003	0	0
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.0034615	0.0241667	0.0206897	0.2	0.1	0.008	0	0.0021429
I1	0	0	0	0.13	0.03	0	0	0
R1	0	0	0	0	0	0	0	0
I2	0.0006923	0.0024167	0.0057931	0	0	0.001	0	0
I3	0	0	0	0	0	0	0	0
I4	0	0	0.0082759	0	0	0.007	0	0
I5	0	0	0.0082759	0	0.01	0.005	0	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**F79**	**F80**	**F81**	**F82**	**F83**	**F84**	**F85**	**F86**
F80	0	0.003	0.003	0	0	0	0.003	0.002
F81	0	0	0	0	0	0	0	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0.004	0.003	0	0.0013333	0.0033333	0.0006667	0	0
I1	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0
I2	0.0016	0.002	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**B1**	**I1**	**R1**	**I2**	**I3**	**I4**	**I5**	**I6**
F80	0	0	0	0	0	0.05	0.04	0.035
F81	0	0	0	0	0	0	0	0.008
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0	0	0	0	0	0
F86	0	0	0	0	0	0	0	1
B1	0	0	0	0	0	0	0	0
I1	0	0.01	0	0	0	0	0.1	0
R1	0	0	0	0	0.06	0.025	0.04	0
I2	0	0	0	0.02	0	0.025	0.02	0
I3	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0.005	0.01	0.005
I5	0	0.005	0	0.025	0.005	0.002	0.002	0.04
I6	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0.01	0	0
I8	0	0	0	0	0	0	0	0
I9	0	0	0	0	0.002	0.002	0.004	0.006
I10	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0.015
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**I7**	**I8**	**I9**	**I10**	**I11**	**I12**	**I13**	**I14**
F80	0.021	0.05	0.035	0	0.1	0.005	0.027	0.02
F81	0	0	0	0	0.05	0	0.071	0
F82	0	0	0	0	0	0	0	0
F83	0	0	0	0	0	0	0	0
F84	0	0	0	0	0	0	0	0
F85	0	0	0.001	0	0	0	0	0
F86	0	0	0	0	0	0	0	0
B1	0	0	0	0	0	0	0	0
I1	0	0	0	0	0	0	0	0
R1	0.02	0.02	0.025	0	0	0	0.02	0.02
I2	0	0	0.5	0.01	0.005	0	0.4	0
I3	0	0	0.15	0	0.05	0	0.5	0
I4	0	0.05	0.05	0	0.004	0.001	0.05	0.02
I5	0.01	0.11	0.08	0.01	0.05	0.005	0.212	0
I6	0	0	0	0	0	0	0	0.05
I7	0.08	0.07	0.135	0.005	0.03	0.005	0.145	0.02
I8	0.006	0.015	0	0	0.004	0.005	0.01	0
I9	0	0	0.02	0.02	0.05	0.015	0.04	0.058
I10	0	0	0	0	0	0	0	0.04
I11	0	0	0.01	0	0.03	0	0.03	0.04
I12	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0

Code	**I15**	**I16**	**I17**	**I18**	**A1**	**A2**	**D1**
F80	0.08	0.1	0.109	0.005	0	0.2	0.1
F81	0	0	0.2	0	0	0.335	0.336
F82	0.002	0.001	0	0	0	0.9970001	0
F83	0.001	0	0	0	0	0.999	0
F84	0	0	0	0	0	1	0
F85	0	0	0.005	0	0	0.497	0.497
F86	0	0	0	0	0	0	0
B1	0	0	0.05	0	0	0	0
I1	0	0	0.565	0	0	0	0
R1	0.34	0.016	0	0	0	0.414	0
I2	0	0	0	0	0	0	0
I3	0	0	0	0	0	0.18	0.12
I4	0.05	0.01	0.03	0.03	0.02	0.6	0.048
I5	0	0	0.15	0.055	0	0.084	0.12
I6	0.06	0.04	0.04	0.04	0	0.6	0.17
I7	0.02	0.012	0	0.02	0	0.088	0.36
I8	0.05	0.05	0	0	0	0.86	0
I9	0.04	0.015	0	0.03	0	0.4	0.298
I10	0.02	0.008	0	0.3	0	0.632	0
I11	0.05	0.01	0.05	0.05	0.1	0.245	0.37
I12	0	0	0	0.1	0.01	0.13	0.76
I13	0	0	0	0.1	0.15	0	0.75
I14	0	0	0.15	0.1	0.2	0	0.55
I15	0	0	0	0.001	0	0	0.999
I16	0	0	0.15	0.1	0	0.65	0.1
I17	0	0	0	0.6	0.2	0	0.2
I18	0	0	0	0	0	0.05	0.95
A1	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0

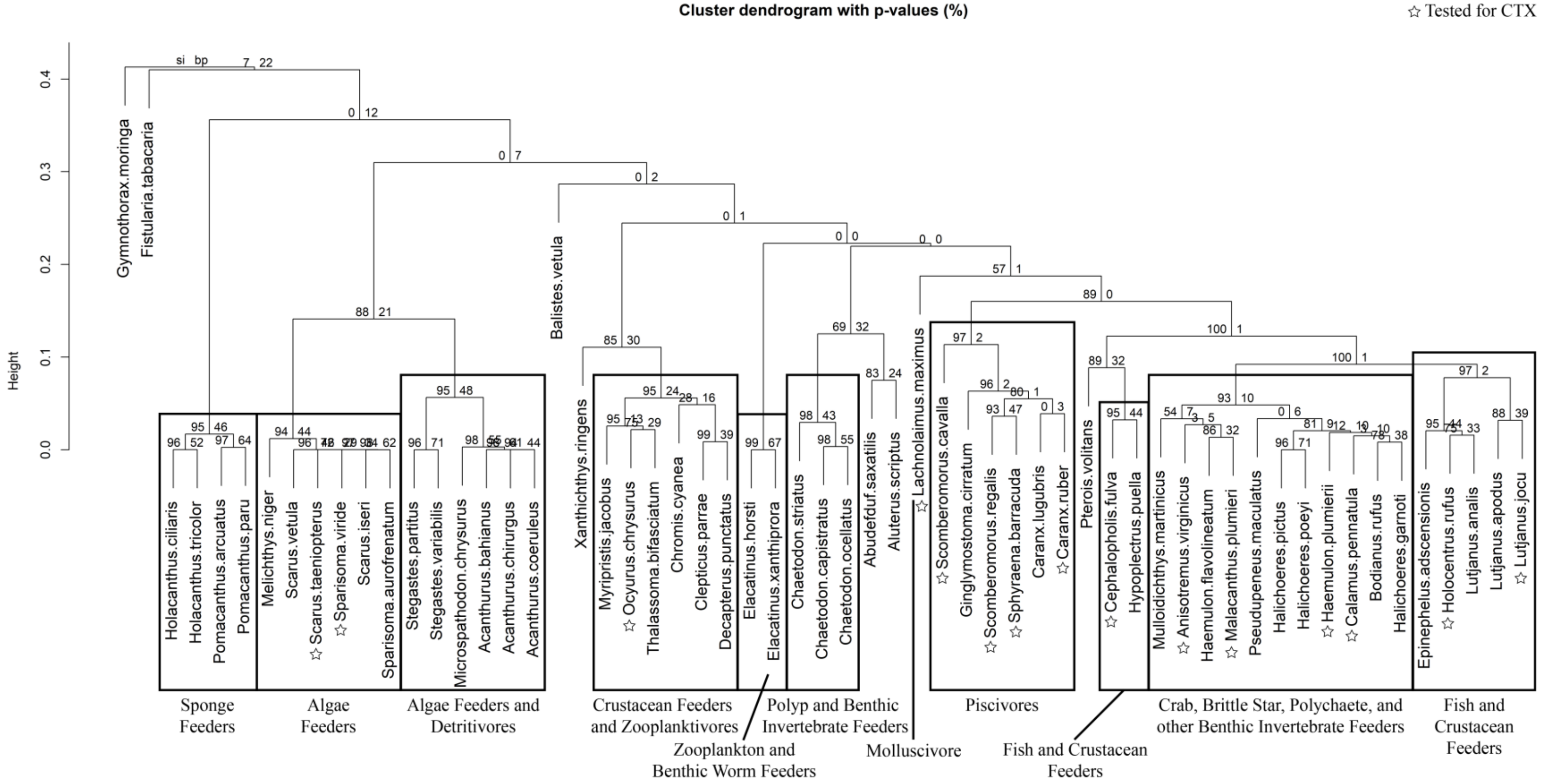

Figure 6-1 Result of the hierarchical cluster analysis from the species' diet matrix observed in the hotspot and coldspot. The predator groups were species we identified in those sites, and prey items were all species in addition to the 50 compartments from the Opitz (1996) model. Starred species were sampled for CTX estimation.

Final Hotspot Diet Composition Matrix

Table 6-3 The new, condensed diet composition matrix for the hotspot.

Compartment	**Code**	**Compartment**	**Code**
Algae feeders	F1	Large scaridae H	F27
Algae feeders and detritivores	F2	Large sharks/rays C	F28
Amphipods/isopods/tanaids/pycnogonids	I1	Large to intermediate schooling fish P	F29
Anisotremus virginicus	F3	Lobsters	I12
Ascidians/barnacles/bryozoans	I2	*Lutjanus jocu*	F30
Asteroids	I3	*Malacanthus plumieri*	F31
Bivalves	I4	Octopuses	I13
Blenniidae H	F4	*Ocyurus chrysurus*	F32
Calamus pennatula	F5	Polychaetes/priapuloids/ophiuroids	I14
Caranx ruber	F6	Polyp and benthic invertebrate feeders	F33
Cephalopholis fulva	F7	*Scarus taeniopterus*	F34
Chitons/scaphopods	I5	*Scomberomorus cavalla*	F35
Corals/sea anemones	I6	*Scomberomorus regalis*	F36
Crab, brittle star, and polychaete feeders	F8	Sea birds	B1
Crabs	I7	Sea turtles	R1
Crustacean feeders and zooplanktivores	F9	Sharks/scombrids C	F37
Decomposers/microfauna	I8	Shrimps/hermit crabs/stomatopods	I15
Echinoids	I9	Small gobiidae C	F38
Engraulidae H	F10	Small jacks C	F39
Fish and crustacean feeders	F11	Small reef fish C1	F40
Gastropods	I10	Small reef fish C2	F41
Haemulon plumieri	F12	Small reef fish O1	F42
Hemiramphidae H	F13	Small reef fish O2	F43
Holocentrus rufus	F14	Small reef fish O3	F44
Holothurids/sipunculids/echiuroids/ hemichordates	I11	Small scaridae H	F45
Intermediate jacks C	F15	Small schooling fish P	F46
Intermediate reef fish C1	F16	*Sparisoma viride*	F47
Intermediate reef fish C2	F17	*Sphyraena barracuda*	F48
Intermediate reef fish C3	F18	Sponge feeders	F49
Intermediate reef fish C4	F19	Sponges	I16
Intermediate reef fish H	F20	Squids	I17
Intermediate scaridae H	F21	Zooplanktivores and benthic worm feeders	F50
Kyphosidae H	F22	Zooplankton	I18
Lachnolaimus maximus	F23	Phytoplankton	A1
Large groupers C	F24	*Gambierdiscus* spp.	A2
Large jacks C	F25	Benthic autotrophs	A3
Large reef fish	F26	Detritus	D1

Predator/Prey Matrix	**Code**	**F1**	**F2**	**I1**	**F3**	**I2**	**I3**
Algae feeders	**F1**	0.00094	0.00236	0	0.00047	0.01315	0
Algae feeders and detritivores	**F2**	0	0	0.01531	0	0	0
Amphipods/isopods/tanaids/pycnogonids	**I1**	0	0	0	0	0.05	0
Anisotremus virginicus	**F3**	0	0	0.14986	0	0.015	0
Ascidians/barnacles/bryozoans	**I2**	0	0	0	0	0	0
Asteroids	**I3**	0	0	0	0	0.02	0.08
Bivalves	**I4**	0	0	0	0	0	0
Blenniidae H	**F4**	0	0	0	0	0	0
Calamus pennatula	**F5**	0	0	0	0	0	0
Caranx ruber	**F6**	0.11817	0.05351	0	0	0	0
Cephalopholis fulva	**F7**	0.04381	0.06571	0	0	0	0
Chitons/scaphopods	**I5**	0	0	0	0	0.04	0
Corals/sea anemones	**I6**	0	0	0	0	0	0
Crab, brittle star, and polychaete feeders	**F8**	6E-05	0.00124	0.0178	0	0	0
Crabs	**I7**	0	0	0.005	0	0.02	0
Crustacean feeders and zooplanktivores	**F9**	0	0.00815	0.05038	0	0.05092	0
Decomposers/microfauna	**I8**	0	0	0	0	0	0
Echinoids	**I9**	0	0	0	0	0	0.006
Engraulidae H	**F10**	0	0	0	0	0	0
Fish and crustacean feeders	**F11**	0.02046	0.00806	0	0.0008	0	0
Gastropods	**I10**	0	0	0.006	0	0.058	0
Haemulon plumierii	**F12**	0.00326	0.00652	0.03498	0.00109	0	0
Hemiramphidae H	**F13**	0	0	0	0	0	0
Holocentrus rufus	**F14**	0.00077	0.00077	0.019	0	0	0
Holothurids/sipunculids/ echiuroids/hemichordates	**I11**	0	0	0	0	0	0
Intermediate jacks C	**F15**	0.08922	0.05805	0	0.0047	0	0
Intermediate reef fish C1	**F16**	0.00024	0.00047	0.00022	7.9E-05	0.04874	0.0038
Intermediate reef fish C2	**F17**	0.01358	0.00158	0.03065	0.00051	0.05064	0
Intermediate reef fish C3	**F18**	0.02316	0.01156	0.04228	0.00026	0.03987	0
Intermediate reef fish C4	**F19**	0	0	0	0	0.02249	0.00191
Intermediate reef fish H	**F20**	0	0	0	0	0	0.002
Intermediate scaridae H	**F21**	0	0	0	0	0	0
Kyphosidae H	**F22**	0	0	0	0	0	0
Lachnolaimus maximus	**F23**	0	0	0.01	0	0.005	0
Large groupers C	**F24**	0.08467	0.008	0	0.00231	0	0
Large jacks C	**F25**	0.0044	0.0176	0	0.0041	0	0
Large reef fish	**F26**	0.00448	0.00896	0	0.00073	0	0
Large scaridae H	**F27**	0	0	0	0	0	0
Large sharks/rays C	**F28**	0.01166	0.07433	0	0.00021	0	0
Large to intermediate schooling fish P	**F29**	0.001	0.004	0	0	0	0
Lobsters	**I12**	0	0	0	0	0	0
Lutjanus jocu	**F30**	0.02601	0	0	0	0	0
Malacanthus plumieri	**F31**	0	0	0.016	0	0	0
Octopuses	**I13**	0.0002	0.0012	0	5.1E-05	0	0
Ocyurus chrysurus	**F32**	0	0	0.00778	0	0.02978	0
Polychaetes/priapuloids/ophiuroids	**I14**	0	0	0.015	0	0.04	0
Polyp and benthic invertebrate feeders	**F33**	0	0	0.00255	0	0.02634	0
Scarus taeniopterus	**F34**	0	0	0	0	0	0
Scomberomorus cavalla	**F35**	0	0	0	0	0	0
Scomberomorus regalis	**F36**	0	0	0	0.12813	0	0

Code	I4	F4	F5	F6	F7	I5	I6	F8	I7	F9
F1	0	0.00047	0	0	0.00047	0	0.00755	0.00283	0.02143	0.00236
F2	0.00159	0	0	0	0	0	0.00645	0	0	0
I1	0	0	0	0	0	0	0.04	0	0	0
F3	0.055	0	0	0	0	0.01	0	0	0.16929	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0.145	0	0	0	0	0.005	0.012	0	0.01	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0.123	0	0	0	0	0	0	0	0.215	0
F6	0	0.05351	0	0	0	0	0	0.10702	0.00433	0
F7	0	0.0219	0	0	0	0	0	0	0.18433	0
I5	0	0	0	0	0	0	0.008	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0.07841	0.00062	0	0	0	0.01441	0	0.00068	0.30865	0.00124
I7	0.05	0	0	0	0	0	0.01	0.00069	0.005	0.00069
F9	0	0.00408	0	0	0	0	0	0.00408	0.04528	0.00408
I8	0	0	0	0	0	0	0	0	0	0
I9	0.01	0	0	0	0	0	0.05	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0	0	0	0.00873	0.00226	0	0.04641	0.34789	0.01845
I10	0.04	0	0	0	0	0.02	0.015	0	0.002	0
F12	0.01348	0.00109	0	0	0.00109	0.00748	0	0.00652	0.27373	0.00544
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0.00039	0	0	0	0.006	0	0.00116	0.573	0.00077
I11	0	0	0	0	0	0	0	0	0	0
F15	0	0.04575	0.0047	0.00239	0.00239	0	0	0.02369	0.00327	0.01428
F16	0.01763	7.9E-05	7.9E-05	0	0	0.00028	0.00843	0.00055	0.0826	0.00039
F17	0.07218	0	0.00051	0.00012	0.00041	0.01978	0.00886	0.00193	0.09888	0.00092
F18	0.02292	0.00199	0.00026	0	0.00033	0.00997	0.00299	0.00179	0.1495	0.00202
F19	0	0	0	0	0	0	0.02988	0	0.00096	0
F20	0	0	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0.426	0	0	0	0	0.006	0	0	0.061	0
F24	0	0	0.00231	0.00125	0	0	0	0.00692	0.07	0.00481
F25	0.057	0	0.0041	0.01875	0.00344	0	0	0.01575	0.01	0.00755
F26	0.05333	0	0.00073	0.00213	0.00079	0	0	0.14748	0.02276	0.00512
F27	0	0	0	0	0	0	0.001	0	0	0
F28	0.055	0	0.00021	0.00513	0.00057	0	0	0.00233	0.04104	0.00273
F29	0	0	0	0	0	0	0	0	0	0
I12	0.5	0	0	0	0	0	0	0	0	0
F30	0	0	0	0	0.01445	0	0	0.03212	0.1585	0.0289
F31	0	0	0	0	0	0.057	0	0.0372	0.185	0
I13	0.4	0	5.1E-05	0	8.3E-05	0.01	0	0.00072	0.025	0.00062
F32	0.01478	0	0	0	0	0	0	0	0.20083	0
I14	0.03	0	0	0	0	0	0.01	0	0	0
F33	0	0	0	0	0	0	0.48227	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0.15383	0	0	0	0	0	0.01398
F36	0	0	0	0.06407	0	0	0	0.02402	0	0.13614

Code	**I8**	**I9**	**F10**	**F11**	**I10**	**F12**	**F13**	**F14**	**I11**	**F15**
F1	0.00073	0	0.00047	0	0.04019	0	0.00047	0.00047	0	0
F2	0	0	0	0	0.0023	0	0	0	0	0
I1	0.04	0	0	0	0	0	0	0	0	0
F3	0.005	0	0	0	0.038	0	0	0	0.004	0
I2	0.1	0	0	0	0	0	0	0	0	0
I3	0.02	0.07	0	0	0.135	0	0	0	0.005	0
I4	0.1	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0.001	0	0	0	0	0
F5	0	0.04	0	0	0.082	0	0	0	0.09	0
F6	0	0	0.10434	0	0.004	0	0	0	0	0
F7	0	0	0	0	0	0	0	0	0	0
I5	0.3	0	0	0	0	0	0	0	0	0
I6	0.1	0	0	0	0	0	0	0	0	0
F8	0	0.11641	0.00062	0	0.102	0	0	0	0.01073	0
I7	0.03	0.05	0	0	0.05	0	0	0	0.001	0
F9	0	0.00899	0.00408	0	0.08579	0	0	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0.015	0	0	0	0	0	0	0.005	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0	0	0.0027	0.05715	0.00181	0	0	0	0
I10	0.03	0	0	0	0.02	0	0	0	0.015	0
F12	0	0.12448	0.00109	0	0.06948	0	0.00109	0.00109	0.14348	0
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0	0.00039	0	0.078	0	0.00039	0	0	0
I11	0.1	0	0	0	0	0	0	0	0	0
F15	0	0	0.08992	0.00972	0.00327	0.0047	0.00239	0.0047	0	0.00479
F16	0	0.1924	7.9E-05	0.00016	0.01686	7.9E-05	7.9E-05	7.9E-05	0.00433	0
F17	0	0.09888	0.00198	0.00041	0.03527	0.00051	0	0.00051	0.05933	0.00087
F18	0	0.0598	0.00199	0.00199	0.00399	0.00026	0	0.00026	0.00997	0
F19	0	0.00287	0	0	0.00026	0	0	0	0.00574	0
F20	0	0	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0.046	0	0	0.397	0	0	0	0	0
F24	0	0	0	0.01417	0	0.00231	0	0.00231	0	0.00875
F25	0	0.082	0	0.03678	0.158	0.0041	0	0.0041	0	0.13125
F26	0	0.07111	0.00071	0.00435	0.28444	0.00073	0	0.00073	0	0.01493
F27	0	0	0	0	0	0	0	0	0	0
F28	0	0.00657	0.02298	0.01425	0.00821	0.00021	0.00164	0.00021	0.04104	0.03591
F29	0	0	0.006	0	0	0	0	0	0.12	0
I12	0	0	0	0	0.15	0	0	0	0	0
F30	0	0	0	0	0.036	0.0289	0	0.0289	0	0
F31	0	0.027	0	0	0	0	0	0	0.104	0
I13	0	0	0	8.3E-05	0.5	5.1E-05	0	5.1E-05	0	0
F32	0	0	0	0	0.08778	0	0	0	0	0
I14	0.05	0	0	0	0.01	0	0	0	0	0
F33	0	0	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0.15383	0	0	0	0	0	0	0.09789
F36	0	0	0.06407	0	0	0	0.06407	0	0	0

Code	**F16**	**F17**	**F18**	**F19**	**F20**	**F21**	**F22**	**F23**	**F24**	**F25**
F1	0	0.00094	0.00142	0	0.00047	0	0	0	0	0
F2	0	0	0	0	0	0	0	0	0	0
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0	0	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0	0	0
F6	0	0	0	0.01987	0	0.03535	0	0	0	0
F7	0	0	0.06571	0	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0	0.00062	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0	0	0
F9	0	0.00408	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0.00136	0.02742	0.0555	0.00309	0	0.01522	0	0	0	0
I10	0	0	0	0	0	0	0	0	0	0
F12	0	0.00217	0.00326	0.00109	0.00109	0.00109	0	0	0	0
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0.00039	0.00077	0	0.00039	0	0	0	0	0
I11	0	0	0	0	0	0	0	0	0	0
F15	0.00533	0.06939	0.00479	0.00713	0.04616	0.0422	0.01466	0.00239	0	0
F16	0.00079	0.00719	0.04527	0.00253	0.00324	0.01272	0.00245	0	0	0
F17	0	0.0142	0.01277	0.0017	0.00198	0.01978	0.00297	7.6E-05	0	0
F18	0.00664	0.00964	0.0103	0.00171	0.00449	0.00465	0.00299	0	0	0
F19	0	0	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0.00267	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0.05667	0.06462	0	0.00571	0.01	0	0.02	0.01785	0	0
F25	0.13333	0.11487	0.10678	0	0.022	0	0	0	0	0
F26	0.01422	0.02042	0.02449	0.00122	0.00818	0.00379	0.00213	0.00027	0	0.00284
F27	0	0	0	0	0	0	0	0	0	0
F28	0.05472	0.00589	0.01767	0.00938	0.02134	0.01881	0.04925	0.00189	0.07634	0.00246
F29	0	0	0	0	0.005	0	0	0	0	0
I12	0	0	0	0	0	0	0	0	0	0
F30	0.00321	0.07708	0.16219	0	0	0.02601	0	0	0	0
F31	0	0	0.0372	0	0	0	0	0	0	0
I13	0	0.00138	0.00258	0	0.001	0	0	7.7E-05	0	0
F32	0	0	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0	0	0
F33	0	0	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0	0	0
F36	0	0	0.06407	0	0	0	0	0	0	0

Code	**F26**	**F27**	**F28**	**F29**	**I12**	**F30**	**F31**	**I13**	**F32**	**I14**
F1	0	0	0	0.00047	0	0	0.00047	0	0.00047	0
F2	0	0	0	0	0	0	0	0	0	0.01651
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0	0	0.305
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0.03
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0	0	0.282
F6	0	0	0	0.05351	0	0	0	0	0	0
F7	0	0	0	0	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0	0	0	0	0	0	0	0.00112	0	0.20707
I7	0	0	0	0	0	0	0	0	0	0.004
F9	0	0	0	0	0	0	0	0	0	0.07012
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0.004
F10	0	0	0	0	0	0	0	0	0	0
F11	0.00806	0	0	0	0.00771	0	0.00806	0.02106	0	0
I10	0	0	0	0	0.002	0	0	0	0	0.05
F12	0	0	0	0.00109	0	0	0.00109	0	0.00109	0.20248
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0	0	0.00039	0	0	0	0	0	0.116
I11	0	0	0	0	0	0	0	0	0	0
F15	0	0.01112	0.00239	0.0482	0	0	0.00239	0	0.00246	0
F16	0	0.02378	0	7.9E-05	0.00037	0	7.9E-05	0.01106	7.9E-05	0.01615
F17	0.00076	0.01318	0	0.00099	0.00099	7.6E-05	0.00041	0.04944	0	0.17816
F18	0	0.03654	0	0.00199	0	0	0.00033	0.0299	0.00083	0.04983
F19	0	0	0	0	0	0	0	0	0	0.00574
F20	0	0	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0.17846	0.16533	0.005	0	0.16	0.01785	0	0	0.00708	0
F25	0	0	0	0	0	0	0.00344	0	0.01667	0
F26	0.00274	0.00474	0	0.02844	0.00142	0.00027	0.00079	0.0064	0.00178	0
F27	0	0	0	0	0	0	0	0	0	0
F28	0.01894	0.0314	0.00821	0.02593	0.0041	0.00189	0.00057	0.02627	0.00684	0.04104
F29	0	0	0	0.003	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0	0	0.05
F30	0	0.00867	0	0.0289	0.0885	0	0	0.07	0	0
F31	0	0	0	0	0	0	0	0	0	0.323
I13	0.00077	0	0	0	0	7.7E-05	8.3E-05	0.02	0	0.005
F32	0	0	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0	0	0.03
F33	0	0	0	0	0	0	0	0	0	0.3916
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0.32165	0	0	0	0	0.15383	0
F36	0	0	0	0.11532	0	0	0	0	0	0

Code	F33	F34	F35	F36	B1	R1	F37	I15	F38	F39
F1	0.00142	0	0	0	0	0	0	0.01449	0.00047	0.00047
F2	0	0	0	0	0	0	0	0.00308	0	0
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0.21457	0	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0.168	0	0
F6	0	0.01115	0	0	0	0	0	0.04067	0	0
F7	0	0	0	0	0	0	0	0.35567	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0.00062	0	0	0	0	0	0	0.12464	0.00062	0
I7	0	0	0	0	0	0	0	0.01	0	0
F9	0	0	0	0	0	0	0	0.04452	0.00408	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0.00628	0	0	0	0	0	0.10643	0.00872	0
I10	0	0	0	0	0	0	0	0.004	0	0
F12	0.00326	0.00109	0	0	0	0	0	0.07173	0.00109	0.00109
F13	0	0	0	0	0	0	0	0	0	0
F14	0.00116	0	0	0	0	0	0	0.162	0.00039	0
I11	0	0	0	0	0	0	0	0	0	0
F15	0.01411	0.01235	0	0.00239	0	0	0.00239	0.02536	0.00239	0.00239
F16	0.00024	7.9E-05	0	0	0	0	0	0.02972	7.9E-05	0
F17	0.00152	0.00494	0	0	0	0	0	0.01978	0	0
F18	0.00077	0.00116	0	0	0	0	0	0.09967	0.00399	0.00224
F19	0	0	0	0	0	0	0	0.00204	0	0
F20	0	0.00067	0	0	0.001	0.002	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0.049	0	0
F24	0.00692	0	0.00083	0.00083	0	0.035	0.00333	0	0	0.0075
F25	0.01231	0	0	0	0	0	0	0.015	0	0
F26	0.00219	0.00095	0	0	0	0	0	0.02133	0	0.01067
F27	0	0	0	0	0	0	0	0	0	0
F28	0.00063	0.00027	0.00137	0.00137	0.01642	0.0041	0.00547	0.04104	0	0.00246
F29	0	0	0	0	0	0	0	0.004	0	0
I12	0	0	0	0	0	0	0	0	0	0
F30	0	0.00867	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0.164	0	0
I13	0.00015	0	0	0	0	0	0	0.02	0	0
F32	0	0	0	0	0	0	0	0.09158	0	0
I14	0	0	0	0	0	0	0	0	0	0
F33	0	0	0	0	0	0	0	0.01641	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0	0	0.02797
F36	0	0	0	0	0	0	0	0.016	0	0.06407

Code	**F40**	**F41**	**F42**	**F43**	**F44**	**F45**	**F46**	**F47**	**F48**	**F49**
F1	0.00094	0.00047	0.00047	0.00047	0.00047	0.00047	0.00047	0	0	0
F2	0	0	0	0	0	0	0	0	0	0
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0	0	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0	0	0
F6	0	0	0.02446	0.06077	0.05351	0.01115	0.10434	0.05351	0	0
F7	0.13143	0	0.06571	0.06571	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0.00124	0.00062	0.00062	0.00062	0.00188	6E-05	0.00062	0	0	0
I7	0.00862	0.007	0	0	0	0	0	0	0	0
F9	0.00815	0.00408	0.00408	0.00408	0.00408	0	0.00408	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0.02396	0.04095	0.04511	0.00154	0.04718	0.00659	0.02024	0.00659	0	0
I10	0	0	0	0	0	0	0	0	0	0
F12	0.00217	0.00109	0.00109	0.00109	0.00109	0.00109	0.00109	0	0	0.00109
F13	0	0	0	0	0	0	0.2	0	0	0
F14	0.00077	0.00039	0.00039	0.00039	0.00039	0.00039	0.00039	0	0	0
I11	0	0	0	0	0	0	0	0	0	0
F15	0.00479	0.00321	0.03708	0.04575	0.04575	0.03566	0.09237	0.01235	0	0.00901
F16	0.00411	0.00237	0.01588	7.9E-05	7.9E-05	0.01272	0.04898	7.9E-05	0	0.00024
F17	0	0.00099	0	0	0	0.0033	0.06682	0.00494	7.6E-05	0.00127
F18	0.00859	0.00997	0.03189	0.001	0.01495	0.00199	0.00199	0.00116	0	0.00211
F19	0	0	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0.00067	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0	0.01785	0.01137
F25	0	0	0	0	0	0	0	0	0	0.01667
F26	0.00061	0	0.15412	0.00427	0.00711	0.00024	0.04196	0.00095	0.00027	0.00269
F27	0	0	0	0	0	0	0	0	0	0
F28	0.01415	0.00493	0.03413	0.00328	0.00082	0.01799	0.05794	0.00027	0.00189	0.01388
F29	0	0	0	0	0	0	0.047	0	0	0
I12	0	0	0	0	0	0	0	0	0	0
F30	0	0	0	0	0	0.00867	0.05781	0.03758	0	0.0289
F31	0.0372	0.0124	0	0	0	0	0	0	0	0
I13	0.00603	0.001	0.0016	0.002	0	0	0	0	7.7E-05	0
F32	0	0	0	0	0	0	0.07828	0	0	0
I14	0	0	0	0	0	0	0	0	0	0
F33	0	0	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0	0	0
F36	0.02403	0.00801	0	0	0	0	0.20501	0	0	0

Code	I16	I17	F50	I18	A1	A2	A3	D1
F1	0.00083	0	0.00094	0.06418	0		0.81479	0
F2	0.00277	0	0	0	0.00018		0.50646	0.44535
I1	0.06	0	0	0.04	0		0.6	0.17
F3	0	0	0	0.03429	0		0	0
I2	0	0	0	0.15	0.2		0	0.55
I3	0.02	0	0	0	0		0.088	0.36
I4	0	0	0	0	0.15		0	0.75
F4	0	0	0	0.005	0		0.497	0.497
F5	0	0	0	0	0		0	0
F6	0	0.014	0	0.07283	0		0	0
F7	0	0	0	0	0		0	0
I5	0.02	0	0	0	0		0.632	0
I6	0	0	0	0.15	0		0.65	0.1
F8	0	0	0.00124	0.00547	0		0	0
I7	0.05	0	0	0.03	0.02		0.6	0.048
F9	0	0	0.00815	0.5747	0		0	0
I8	0	0	0	0	0		0.05	0.95
I9	0.05	0	0	0	0		0.86	0
F10	0	0	0	0.2	0.7		0	0.1
F11	0	0	0.00872	0.01537	0		0	0
I10	0.04	0	0	0	0		0.4	0.298
F12	0	0	0.00217	0	0		0	0
F13	0	0	0	0	0		0.8	0
F14	0	0	0.00077	0.034	0		0	0
I11	0	0	0	0	0.01		0.13	0.76
F15	0	0.00327	0.00479	0.018	0		0	0
F16	0.15801	0.00079	0.00016	0.19752	0		0.02543	0
F17	0.06922	0	0	0.0205	0		0.01087	0
F18	0.06977	0	0	0.14992	0		0.05183	0
F19	0.88193	0	0	0.00096	0		0.04521	0
F20	0	0	0	0	0		0.533	0.458
F21	0.001	0	0	0	0		0.999	0
F22	0	0	0	0	0		1	0
F23	0	0	0	0	0		0	0
F24	0	0	0	0	0		0	0
F25	0	0	0	0	0		0	0
F26	0	0.00071	0	0.00427	0		0	0
F27	0.002	0	0	0	0		0.997	0
F28	0	0.01396	0	0	0		0	0.01642
F29	0	0	0	0.722	0		0.088	0
I12	0	0	0	0	0		0.18	0.12
F30	0	0.022	0	0.018	0		0	0
F31	0	0	0	0	0		0	0
I13	0	0	0	0	0		0	0
F32	0	0.02178	0	0.46743	0		0	0
I14	0.05	0	0	0.05	0.1		0.245	0.37
F33	0	0	0	0.08083	0		0	0
F34	0.015	0	0	0	0		0.985	0
F35	0	0.077	0	0	0		0	0
F36	0	0.023	0	0	0		0	0

Predator/Prey Matrix	**Code**	**F1**	**F2**	**I1**	**F3**	**I2**	**I3**
Sea birds	**B1**	0.003	0.005	0	0.00103	0	0
Sea turtles	**R1**	0	0	0	0	0.02	0.02
Sharks/scombrids C	**F37**	0.01333	0.0108	0	0.00103	0	0
Shrimps/hermit crabs/stomatopods	**I15**	0	0	0.04	0.00026	0	0.01
Small gobiidae C	**F38**	0	0	1	0	0	0
Small jacks C	**F39**	0.002	0.008	0	0	0	0
Small reef fish C1	**F40**	0.00133	0.001	0.04202	5.1E-05	0	0
Small reef fish C2	**F41**	0.0001	0.0006	0.1	5.1E-05	0	0
Small reef fish O1	**F42**	0.00267	0.0012	0.056	7.7E-05	0.02	0
Small reef fish O2	**F43**	0	0	0.035	0	0.02	0.021
Small reef fish O3	**F44**	0	0	0.008	0	0	0
Small scaridae H	**F45**	0	0	0	0	0	0
Small schooling fish P	**F46**	0	0	0.01	0	0.03	0
Sparisoma viride	**F47**	0	0	0	0	0	0
Sphyraena barracuda	**F48**	0.01508	0.05026	0	0	0	0
Sponge feeders	**F49**	0	0	0	0	0.07304	0
Sponges	**I16**	0	0	0	0	0	0
Squids	**I17**	0	0	0	0	0	0
Zooplanktivores and benthic worm feeders	**F50**	0	0	0.5	0	0	0
Zooplankton	**I18**	0	0	0	0	0	0

Code	**I4**	**F4**	**F5**	**F6**	**F7**	**I5**	**I6**	**F8**	**I7**	**F9**
B1	0	0	0.00103	0.0075	0.00083	0	0	0.00563	0	0.01108
R1	0.02	0	0	0	0	0	0.016	0	0.025	0
F37	0.007	0.001	0.00103	0.00425	0.00233	0	0	0.00679	0.02	0.01724
I15	0.212	0	0.00026	0	0	0.01	0	0.00146	0.002	0.00095
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0.00025	0	0	0	0	0	0.00025
F40	0.05998	0.006	5.1E-05	0	0	0.03999	0	0.00036	0.10003	0.00026
F41	0.03	0.001	5.1E-05	0	0	0.002	0	0.00071	0.063	0.0006
F42	0.02	0.004	7.7E-05	0	0	0	0.02	0.00051	0.01	0.00035
F43	0.027	0.003	0	0	0	0	0.1	0.00021	0.05	0.00021
F44	0.071	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0.001	0	0	0
F48	0	0	0	0.05026	0.00457	0	0	0.00457	0	0.0297
F49	0	0	0	0	0	0	0.05636	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	**I8**	**I9**	**F10**	**F11**	**I10**	**F12**	**F13**	**F14**	**I11**	**F15**
B1	0	0	0.1	0.0025	0	0.00103	0.1	0.00103	0	0.0525
R1	0	0.02	0	0	0.025	0	0	0	0	0
F37	0	0	0.04	0.00483	0.008	0.00103	0.115	0.00103	0	0.02975
I15	0.055	0.11	0.01	0	0.08	0.00026	0	0.00026	0.005	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0.019	0	0	0	0	0	0	0.00175
F40	0	0.10997	0.002	0	0.14396	5.1E-05	0.002	5.1E-05	0.008	0
F41	0	0	0	0	0.005	5.1E-05	0	5.1E-05	0.001	0
F42	0	0	0.004	0	0.038	7.7E-05	0.003	7.7E-05	0	0
F43	0.005	0.05	0.003	0	0.035	0	0	0	0.005	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0.006	0	0.002	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0.00457	0.05026	0	0	0.07768
F49	0	0	0	0	0	0	0	0	0	0
I16	0.001	0	0	0	0	0	0	0	0	0
I17	0	0	0.03	0	0	0	0.015	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0.6	0	0	0	0	0	0	0	0	0

Code	**F16**	**F17**	**F18**	**F19**	**F20**	**F21**	**F22**	**F23**	**F24**	**F25**
B1	0.00667	0.02872	0.02583	0.00286	0.005	0.00333	0	0.00038	0	0.01
R1	0	0	0	0	0	0	0	0	0	0
F37	0.01	0.02872	0.07233	0.00343	0.01	0.00067	0.02	0.00108	0	0
I15	0	0.00718	0	0	0	0	0	0	0	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0	0.01	0	0	0	0	0
F40	0	0.00144	0	0	0	0	0	0	0	0
F41	0	0.00144	0	0	0.0005	0	0	0	0	0
F42	0	0.00215	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0.00457	0.13251	0.05483	0	0	0.01508	0	0	0	0
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	F26	F27	F28	F29	I12	F30	F31	I13	F32	I14
B1	0.00385	0.00133	0	0.28	0	0.00038	0.00083	0	0.00083	0
R1	0	0	0	0	0.06	0	0	0	0	0
F37	0.01077	0.02	0	0.176	0.005	0.00108	0.00233	0.045	0.00125	0.006
I15	0	0	0	0	0.005	0	0	0.025	0	0.05
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0.014	0	0	0	0	0	0
F40	0	0	0	0.002	0	0	0	0	0	0.09997
F41	0	0	0	0	0	0	0	0.02	0	0.079
F42	0	0	0	0.003	0	0	0	0	0	0.05
F43	0	0	0	0	0	0	0	0	0	0.1
F44	0	0	0	0	0	0	0	0	0	0.05
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0.00503	0	0.15536	0	0	0	0.026	0.05026	0
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0.15	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0	0	0.5
I18	0	0	0	0	0	0	0	0	0	0

Code	F33	F34	F35	F36	B1	R1	F37	I15	F38	F39
B1	0.00308	0.00083	0.00333	0.00333	0	0	0.01333	0	0	0.0225
R1	0	0	0	0	0	0	0	0.04	0	0
F37	0.00308	0.00017	0.00167	0.00167	0	0	0.00667	0.05	0	0.0375
I15	0.00077	0	0	0	0	0	0	0.002	0	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0	0	0	0	0	0	0.00075
F40	0.00015	0	0	0	0	0	0	0.04014	0.002	0
F41	0.00015	0	0	0	0	0	0	0.15	0.002	0
F42	0.00023	0	0	0	0	0	0	0.01	0.003	0
F43	0	0	0	0	0	0	0	0.04	0.002	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0.00503	0	0	0	0	0	0	0	0.03427
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0.1	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	F40	F41	F42	F43	F44	F45	F46	F47	F48	F49
B1	0.02155	0.008	0.004	0.003	0	0.00067	0.2	0.00083	0.00038	0.00298
R1	0	0	0	0	0	0	0	0	0	0
F37	0.01724	0.02	0.0112	0.001	0	0.00067	0.136	0.00017	0.00108	0.00382
I15	0.00862	0.005	0	0	0	0	0	0	0	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0	0	0	0.006	0	0	0
F40	0.00259	0.002	0.004	0.002	0.006	0.00067	0.002	0	0	0
F41	0.0069	0.003	0.0008	0	0.004	0	0	0	0	0
F42	0.00345	0.004	0.0048	0.004	0.004	0.00133	0.004	0	0	0
F43	0.00259	0.003	0	0.003	0.003	0	0.003	0	0	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0.002	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0	0	0.05026	0	0.00503	0.15079	0.00503	0	0
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0.13	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	I16	I17	F50	I18	A1	A2	A3	D1
B1	0	0	0	0.05	0		0	0
R1	0.34	0	0	0	0		0.414	0
F37	0	0.01	0	0	0		0	0
I15	0	0.005	0	0.15	0		0.084	0.12
F38	0	0	0	0	0		0	0
F39	0	0	0	0.938	0		0	0
F40	0	0	3.7E-06	0.25993	0		0.05798	0
F41	0	0	0	0.5	0		0.028	0
F42	0.02	0	0	0.1	0		0.326	0.28
F43	0.08	0	0	0.109	0		0.2	0.1
F44	0	0	0	0.2	0		0.335	0.336
F45	0	0	0	0	0		1	0
F46	0	0	0	0.95	0		0	0
F47	0.001	0	0	0	0		0.998	0
F48	0	0	0	0.019	0		0	0
F49	0.77805	0	0	0.01266	0		0.07989	0
I16	0	0	0	0	0		0	0.999
I17	0	0.01	0	0.565	0		0	0
F50	0	0	0	0	0		0	0
I18	0	0	0	0	0.2		0	0.2

Final Hotspot Basic Input Data

I calculated the new biomass, P/B, and Q/B values for the hotspot model by combining the values from the species grouped with hierarchical cluster analysis of the fishes' diets (Figure 6-1). Species’ biomass combined into new compartments were summed for new biomass (g m^{-2}) values. New P/B and Q/B values for species were calculated by taking the median value. The unbalanced Ecopath basic input parameters for the hotspot are found in Table 6-4.

Part of parameterizing Ecopath models is balancing the compartments’ ecotrophic efficiency. In Ecopath, ecotrophic efficiency (*EE*) is a parameter that estimates the exploitation of a compartment in the model (Fetahi and Mengistou, 2007). If a compartment has an $EE > 1$, then the compartment is consumed more than it is producing, which causes an imbalance in the model. To correct this, we need to balance the model. There are several ways to balance an *EE*. You can increase the biomass (g m^{-2}) of the compartment overexploited, reduce the amount of predation on that compartment in the diet matrix, or decrease the biomass (g m^{-2}) of the predator of the prey compartment that is out of balance (there are others but these are the main ways to balance models). Each method is acceptable, but we chose to increase the biomass (g m^{-2}) of the overexploited compartments since the biomass (g m^{-2}) of the compartments is more uncertain than the diet data from Randall (1967). The balanced Ecopath model for the hotspot is found in Table 6-5.

Table 6-4 Basic input data for the hotspot model (unbalanced) where P/B is the production to biomass ratio per year, and Q/B is the consumption to biomass ratio per year. These data need to be balanced in Ecopath to be a complete model.

Compartment	**Code**	**Biomass**	**P/B**	**Q/B**
Algae feeders	F1	1.20208	0.81266	24.9436
Algae feeders and detritivores	F2	0.28035	1.0054	25.73
Amphipods/isopods/tanaids/pycnogonids	I1	13.25	5	125.5
Anisotremus virginicus	F3	0.09125	4.79412	10.3
Ascidians/barnacles/bryozoans	I2	137.4	2.3	20
Asteroids	I3	25	0.49	3.24
Bivalves	I4	109.25	2.23	9.5
Blenniidae H	F4	0.6	2.84	36.1
Calamus pennatula	F50	0.03049	1.265	9.3
Caranx ruber	F6	0.33958	1.17	10.1
Cephalopholis fulva	F7	0.04583	2.1	7.8
Chitons/scaphopods	I5	62	0.36	11.7
Corals/sea anemones	I6	121	1.09	9
Crab, brittle star, and polychaete feeders	F8	0.25639	0.83136	7.03711
Crabs	I7	19	1.6	14
Crustacean feeders and zooplanktivores	F9	0.1676	1.3226	10.2953
Decomposers/microfauna	I8	15	280	1900
Echinoids	I9	100	1.1	3.7
Engraulidae H	F10	3.325	2.835	43.4
Fish and crustacean feeders	F11	0.25104	0.57414	10.5471
Gastropods	I10	46.8	2.8	14
Haemulon plumieri	F12	0.02438	0.67	9.4
Hemiramphidae H	F13	1.125	1.23	39.1
Holocentrus rufus	F14	0.06403	1.38	9.8
Holothurids/sipunculids/ echiuroids/hemichordates	I11	66.24	0.31	3.36
Intermediate jacks C	F15	1.29042	1.60354	8.6184
Intermediate reef fish C1	F16	2.64018	0.51694	6.97901
Intermediate reef fish C2	F17	6.20911	1.31838	2.81833
Intermediate reef fish C3	F18	6.85384	0.64	6.39956
Intermediate reef fish C4	F19	1.23718	0.55	5.74686
Intermediate reef fish H	F20	8.51767	0.765	26.85
Intermediate scaridae H	F21	4.76667	1.11	20.05
Kyphosidae H	F22	2.42	0.6	23.6
Lachnolaimus maximus	F23	0.19613	0.34931	4.8
Large groupers C	F24	0.725	0.37	2.3
Large jacks C	F25	0.181	0.525	5.7
Large reef fish	F26	0.66969	0.38	3.9889
Large scaridae H	F27	18.8824	0.85	13.5

Compartment	**Code**	**Biomass**	**P/B**	**Q/B**
Large sharks/rays C	F28	0.3	0.24	4.82833
Large to intermediate schooling fish P	F29	11.172	0.68	12.4
Lobsters	I12	3.27	1	7.4
Lutjanus jocu	F30	0.23972	0.64689	5
Malacanthus plumieri	F31	0.07938	0.42	6.8
Octopuses	I13	8.4	1.9	6.76
Ocyurus chrysurus	F32	0.08648	0.61985	7.9
Polychaetes/priapuloids/ophiuroids	I14	33	5.2	61.6
Polyp and benthic invertebrate feeders	F33	0.03604	1.75865	13.5434
Scarus taeniopterus	F34	0.31556	1.2	20.8
Scomberomorus cavalla	F35	0.02379	0.63	7.4
Scomberomorus regalis	F36	0.03225	0.47	10.8
Sea birds	B1	0.017	5.4	80
Sea turtles	R1	0.5	0.15	3.5
Sharks/scombrids C	F37	0.35796	0.29	9.15
Shrimps/hermit crabs/stomatopods	I15	10	2.8	26.9
Small gobiidae C	F38	0.17499	3.14	17.45
Small jacks C	F39	0.9815	0.83	12.5
Small reef fish C1	F40	4.28824	1.2851	9.75087
Small reef fish C2	F41	1.29833	3.82	14.65
Small reef fish O1	F42	3.53303	1.6	18.2
Small reef fish O2	F43	0.99	1.505	16
Small reef fish O3	F44	0.96	2.53	39.7
Small scaridae H	F45	4.85083	0.94	33.9
Small schooling fish P	F46	10.146	3.54	18.85
Sparisoma viride	F47	0.42778	1.155	20.7
Sphyraena barracuda	F48	1.41946	0.23194	3.3
Sponge feeders	F49	0.41611	0.58887	6.80562
Sponges	I16	800	1.5	5
Squids	I17	1.5	1.3	17.5
Zooplanktivores and benthic worm feeders	F50	0.00011	3.14	17.2
Zooplankton	I18	32	40	165
Phytoplankton	A1	40	70	
Gambierdiscus spp.	A2			
Benthic autotrophs	A3	1300	13.25	
Detritus	D1	2000		

Table 6-5 Basic input data for the hotspot model (balanced) where P/B is the production to biomass ratio per year, Q/B is the consumption to biomass ratio per year, EE is ecotrophic efficiency which indicates the exploitation of a compartment, and P/Q/year is the production to consumption ratio per year. *Gambierdiscus* spp. values changed based on the model and are found in Table 6-10.

Compartment	**Code**	**Trophic level**	**Biomass (g/m^{-2})**	**P/B**	**Q/B**	**EE**	**P/Q (/year)**
Algae feeders	F1	2.3	7.5	0.81266	24.9436	0.77671	0.03257976
Algae feeders and detritivores	F2	2.06	6	1.0054	25.73	0.78353	0.039075
Amphipods/isopods/tanaids/pycnogonids	I1	2.28	35	5	125.5	0.80279	0.03984064
Anisotremus virginicus	F3	3.51	0.15	4.79412	10.3	0.72457	0.4654483
Ascidians/barnacles/bryozoans	I2	2.34	275	2.3	20	0.93822	0.115
Asteroids	I3	2.71	65	0.49	3.24	0.86622	0.1512346
Bivalves	I4	2.1	225	2.23	9.5	0.94668	0.2347368
Blenniidae H	F4	2.01	1	2.84	36.1	0.85358	0.07867036
Calamus pennatula	F5	3.43	0.3	1.265	9.3	0.69567	0.1360215
Caranx ruber	F6	3.71	0.85	1.17	10.1	0.83576	0.1158416
Cephalopholis fulva	F7	3.86	0.15	2.1	7.8	0.90003	0.2692308
Chitons/scaphopods	I5	2.38	150	0.36	11.7	0.78793	0.03076923
Corals/sea anemones	I6	2.34	300	1.09	9	0.94551	0.1211111
Crab, brittle star, and polychaete feeders	F8	3.44	7.5	0.83136	7.03711	0.83097	0.1181396
Crabs	I7	2.43	50	1.6	14	0.9162	0.1142857
Crustacean feeders and zooplanktivores	F9	3.57	2.5	1.3226	10.2953	0.84777	0.1284667
Decomposers/microfauna	I8	2	45	280	1900	0.76045	0.1473684
Echinoids	I9	2.17	150	1.1	3.7	0.93458	0.2972973
Engraulidae H	F10	2.32	5.5	2.835	43.4	0.92036	0.06532258
Fish and crustacean feeders	F11	3.81	0.75	0.57414	10.5471	0.86347	0.05443596
Gastropods	I10	2.37	100	2.8	14	0.98825	0.2
Haemulon plumieri	F12	3.4	1	0.67	9.4	0.49908	0.0712766
Hemiramphidae H	F13	2.52	2.5	1.23	39.1	0.86554	0.0314578
Holocentrus rufus	F14	3.53	0.5	1.38	9.8	0.57558	0.1408163
Holothurids/sipunculids/ echiuroids/ hemichordates	I11	2.1	250	0.31	3.36	0.91897	0.09226191
Intermediate jacks C	F15	3.73	1.290417	1.60354	8.61841	0.64398	0.1860597
Intermediate reef fish C1	F16	3.43	2.640181	0.51694	6.97901	0.6342	0.07407087
Intermediate reef fish C2	F17	3.44	6.209111	1.31838	2.81833	0.9972	0.4677882
Intermediate reef fish C3	F18	3.45	6.853838	0.64	6.39956	0.91204	0.1000069
Intermediate reef fish C4	F19	2.98	1.237181	0.55	5.74686	0.72256	0.09570438
Intermediate reef fish H	F20	2.01	8.517667	0.765	26.85	0.44028	0.02849162
Intermediate scaridae H	F21	2	4.766669	1.11	20.05	0.466	0.0553616
Kyphosidae H	F22	2	2.42	0.6	23.6	0.40708	0.02542373
Lachnolaimus maximus	F23	3.29	0.225	0.34931	4.8	0.94554	0.07277354
Large groupers C	F24	3.95	0.725	0.37	2.3	0.41221	0.1608696
Large jacks C	F25	4.07	0.181	0.525	5.7	0.63758	0.09210526
Large reef fish	F26	3.77	1.5	0.38	3.9889	0.94833	0.09526443
Large scaridae H	F27	2	18.88242	0.85	13.5	0.17861	0.06296296
Large sharks/rays C	F28	3.87	0.3	0.24	4.82833	0.65084	0.04970659
Large to intermediate schooling fish P	F29	3.44	25	0.68	12.4	0.92436	0.05483871
Lobsters	I12	2.82	7.5	1	7.4	0.9456	0.1351351
Lutjanus jocu	F30	4.02	0.2397222	0.64689	5	0.30748	0.1293785
Malacanthus plumieri	F31	3.59	0.8	0.42	6.8	0.71697	0.0617647
Octopuses	I13	3.31	12	1.9	6.76	0.89341	0.2810651
Ocyurus chrysurus	F32	3.68	1.5	0.61985	7.9	0.9203	0.0784624
Polychaetes/priapuloids/ophiuroids	I14	2.35	75	5.2	61.6	0.80351	0.08441558
Polyp and benthic invertebrate feeders	F33	3.38	1	1.75865	13.5434	0.64028	0.1298535
Scarus taeniopterus	F34	2.02	0.75	1.2	20.8	0.69818	0.05769231

Compartment	**Code**	**Trophic level**	**Biomass (g/m^{-2})**	**P/B**	**Q/B**	**EE**	**P/Q (/year)**
Scomberomorus cavalla	F35	4.42	0.35	0.63	7.4	0.10688	0.08513513
Scomberomorus regalis	F36	4.4	0.11	0.47	10.8	0.97099	0.04351852
Sea birds	B1	4.24	0.05	5.4	80	0.93511	0.0675
Sea turtles	R1	2.73	4	0.15	3.5	0.86951	0.04285714
Sharks/scombrids C	F37	4.16	0.45	0.29	9.15	0.92645	0.03169399
Shrimps/hermit crabs/stomatopods	I15	3.08	22	2.8	26.9	0.96708	0.1040892
Small gobiidae C	F38	3.28	0.35	3.14	17.45	0.99095	0.1799427
Small jacks C	F39	3.6	1.045	0.83	12.5	0.99922	0.0664
Small reef fish C1	F40	3.35	12	1.28511	9.75087	0.92559	0.1317939
Small reef fish C2	F41	3.57	3	3.82	14.65	0.86856	0.2607509
Small reef fish O1	F42	2.58	3.533028	1.6	18.2	0.87763	0.08791208
Small reef fish O2	F43	2.98	2	1.505	16	0.80876	0.0940625
Small reef fish O3	F44	2.48	1.5	2.53	39.7	0.94281	0.06372796
Small scaridae H	F45	2	4.850833	0.94	33.9	0.2759	0.02772861
Small schooling fish P	F46	3.59	15	3.54	18.85	0.96867	0.1877984
Sparisoma viride	F47	2	1	1.155	20.7	0.88218	0.0557971
Sphyraena barracuda	F48	4.3	1.419457	0.23194	3.3	0.14483	0.07028595
Sponge feeders	F49	2.97	1	0.58887	6.80562	0.61857	0.08652724
Sponges	I16	2	800	1.5	5	0.56784	0.3
Squids	I17	3.91	3.5	1.3	17.5	0.94934	0.07428571
Zooplanktivores and benthic worm feeders	F50	3.32	0.5	3.14	17.2	0.38336	0.1825581
Zooplankton	I18	2.6	75	40	165	0.81793	0.2424242
Phytoplankton	A1	1	65	70		0.99937	
Gambierdiscus spp.	A2						
Benthic autotrophs	A3	1	1300	13.25		0.79532	
Detritus	D1	1	10000			0.88619	

Final Coldspot Basic Input Parameters and Diet Matrix

Since species seen at the hotspot and coldspot were different, we wanted the models to reflect that. After the hotspot was parameterized, I used and altered those data for the coldspot model. First, any species' biomass (g m^{-2}) that we observed or sampled in the hotspot but not in the coldspot had its species compartment in the models was reduced to 0.001 g m^{-2} in the coldspot model. The compartments' biomasses reduced were *Calamus pennatula*, *Malacanthus plumieri*, *Scomberomorus cavalla*, *Scomberomorus regalis*, and the zooplanktivores and benthic worm feeders. The 'zooplankton and benthic worm feeders' compartment was reduced because both species in that compartment were sampled in the hotspot and not the coldspot. I did not reduce the biomass of species that were in Opitz's compartments. This reduction in biomass assumed that species were absent or had very few individuals in the coldspot since we did not observe or sample them. To account for the decrease in these compartments' biomass, I reduced the prey values for each predator in the diet matrix to 1.0×10^{-8}. The remaining diet percentages were distributed evenly among all other prey groups. After I ran the model, there were several groups with an EE > 1.0. To balance the model, I increased these compartments' biomass until all EE values were < 1.0. Since several compartments were reduced to represent missing species in the coldspot, I assumed that other compartments would have more biomass to make up for it. The initial basic input values for the final coldspot model can be seen in Table 6-6, the final balanced basic estimates are shown in Table 6-7, and the final diet composition matrix for the coldspot is in Table 6-8.

Table 6-6 Basic input parameters for the coldspot model (unbalanced) where P/B is the production to biomass ratio per year, Q/B is the consumption to biomass ratio per year, EE is ecotrophic efficiency which indicates the exploitation of a compartment, and P/Q/year is the production to consumption ratio per year. *Gambierdiscus* spp. values changed based on the model. Balanced values are in Table 6-10.

Compartment	Code	Trophic level	Biomass (g m^{-2})	P/B	Q/B	EE	P/Q (/year)
Algae feeders	F1	2.34	1.378676	0.81266	24.9436	3.72155	0.03257976
Algae feeders and detritivores	F2	2.14	11.61246	1.0054	25.73	0.391	0.039075
Amphipods/isopods/tanaids/pycnogonids	I1	2.45	35	5	125.5	0.80489	0.03984064
Anisotremus virginicus	F3	3.76	0.125	4.79412	10.3	0.56876	0.4654483
Ascidians/barnacles/bryozoans	I2	2.81	275	2.3	20	0.93911	0.115
Asteroids	I3	3.24	65	0.49	3.24	0.86639	0.1512346
Bivalves	I4	2.4	225	2.23	9.5	0.95532	0.2347368
Blenniidae H	F4	2.02	1	2.84	36.1	0.76566	0.07867036
Calamus pennatula	F5	3.7	0.001	7.25	9.3	1.39E-05	0.7795699
Caranx ruber	F6	3.86	0.036242	1.17	10.1	6.58057	0.1158416
Cephalopholis fulva	F7	4.09	0.578386	2.1	7.8	0.32831	0.2692308
Chitons/scaphopods	I5	2.42	150	0.36	11.7	0.79492	0.03076923
Corals/sea anemones	I6	2.4	300	1.09	9	0.9396	0.1211111
Crab, brittle star, and polychaete feeders	F8	3.66	13.11909	0.83136	7.03711	0.43215	0.1181396
Crabs	I7	2.57	50	1.6	14	1.19619	0.1142857
Crustacean feeders and zooplanktivores	F9	3.77	5.564583	1.3226	10.2953	0.37999	0.1284667
Decomposers/microfauna	I8	2	45	280	1900	0.76044	0.1473684
Echinoids	I9	2.23	150	1.1	3.7	0.96358	0.2972973
Engraulidae H	F10	2.39	5.5	2.835	43.4	0.84296	0.06532258
Fish and crustacean feeders	F11	4.01	2.4375	0.57414	10.5471	0.3146	0.05443596
Gastropods	I10	2.67	100	2.8	14	1.00467	0.2
Haemulon plumieri	F12	3.62	1	0.67	9.4	0.6266	0.0712766
Hemiramphidae H	F13	2.55	2.5	1.23	39.1	0.79334	0.0314578
Holocentrus rufus	F14	3.73	1.142082	1.38	9.8	0.23757	0.1408163
Holothurids/sipunculids/ echiuroids/ hemichordates	I11	2.42	250	0.31	3.36	0.91406	0.09226191
Intermediate jacks C	F15	3.88	1.290417	1.60354	8.61841	0.46112	0.1860597
Intermediate reef fish C1	F16	3.72	2.640181	0.51694	6.97901	0.66352	0.07407087
Intermediate reef fish C2	F17	3.72	6.209111	1.31838	2.81833	1.04883	0.4677882
Intermediate reef fish C3	F18	3.69	6.853838	0.64	6.39956	1.10425	0.1000069
Intermediate reef fish C4	F19	3.88	1.237181	0.55	5.74686	0.59336	0.09570438
Intermediate reef fish H	F20	2.03	8.517667	0.765	26.85	0.43101	0.02849162
Intermediate scaridae H	F21	2	4.766669	1.11	20.05	0.46719	0.0553616
Kyphosidae H	F22	2	2.42	0.6	23.6	0.41076	0.02542373
Lachnolaimus maximus	F23	3.56	0.7808145	0.34931	4.8	0.29336	0.07277354
Large groupers C	F24	4.15	0.725	0.37	2.3	0.41241	0.1608696
Large jacks C	F25	4.3	0.181	0.525	5.7	0.64645	0.09210526
Large reef fish	F26	4.01	1.5	0.38	3.9889	1.22803	0.09526443
Large scaridae H	F27	2.01	18.88242	0.85	13.5	0.1791	0.06296296
Large sharks/rays C	F28	4.09	0.3	0.24	4.82833	0.70163	0.04970659
Large to intermediate schooling fish P	F29	3.6	25	0.68	12.4	0.82465	0.05483871
Lobsters	I12	3.17	7.5	1	7.4	0.9808	0.1351351
Lutjanus jocu	F30	4.24	0.4794444	0.64689	5	0.16106	0.1293785
Malacanthus plumieri	F31	3.84	0.001	2.5	6.8	0.91291	0.3676471
Octopuses	I13	3.6	12	1.9	6.76	0.91465	0.2810651
Ocyurus chrysurus	F32	3.87	3.327953	0.61985	7.9	0.15007	0.0784624
Polychaetes/priapuloids/ophiuroids	I14	2.67	75	5.2	61.6	0.816	0.08441558
Polyp and benthic invertebrate feeders	F33	3.56	0.6666667	1.75865	13.5434	0.81955	0.1298535
Scarus taeniopterus	F34	2.03	0.140625	1.2	20.8	3.96524	0.05769231

Compartment	**Code**	**Trophic level**	**Biomass (g m^{-2})**	**P/B**	**Q/B**	**EE**	**P/Q (/year)**
Scomberomorus cavalla	F35	4.57	0.001	0.63	7.4	0.5801	0.08513513
Scomberomorus regalis	F36	4.57	0.001	0.47	10.8	0.47907	0.04351852
Sea birds	B1	4.37	0.05	5.4	80	0.9353	0.0675
Sea turtles	R1	3.14	4	0.15	3.5	0.86989	0.04285714
Sharks/scombrids C	F37	4.32	0.45	0.29	9.15	0.96332	0.03169399
Shrimps/hermit crabs/stomatopods	I15	3.42	22	2.8	26.9	1.10238	0.1040892
Small gobiidae C	F38	3.45	0.35	3.14	17.45	1.25063	0.1799427
Small jacks C	F39	3.75	1.045	0.83	12.5	0.68804	0.0664
Small reef fish C1	F40	3.54	12	1.28511	9.75087	0.9811	0.1317939
Small reef fish C2	F41	3.77	3	3.82	14.65	0.93746	0.2607509
Small reef fish O1	F42	2.94	3.533028	1.6	18.2	1.04626	0.08791208
Small reef fish O2	F43	3.32	2	1.505	16	0.74048	0.0940625
Small reef fish O3	F44	2.82	1.5	2.53	39.7	1.09568	0.06372796
Small scaridae H	F45	2	4.850833	0.94	33.9	0.27301	0.02772861
Small schooling fish P	F46	3.75	15	3.54	18.85	0.97503	0.1877984
Sparisoma viride	F47	2	0.2405697	1.155	20.7	2.70825	0.0557971
Sphyraena barracuda	F48	4.47	0.9076044	0.23194	3.3	0.23728	0.07028595
Sponge feeders	F49	3.79	2.25	0.58887	6.80562	0.30639	0.08652724
Sponges	I16	3	800	1.5	5	0.57341	0.3
Squids	I17	4.07	3.5	1.3	17.5	0.95145	0.07428571
Zooplanktivores and benthic worm feeders	F50	3.56	0.001	6.5	17.2	0.96253	0.3779069
Zooplankton	I18	2.75	75	40	165	0.82292	0.2424242
Phytoplankton	A1	1	65	70		0.99938	
Gambierdiscus spp.	A2						
Benthic autotrophs	A3	1	1300	13.25		0	
Detritus	D1	1	10000			0.1111	

Table 6-7 Basic input parameters for the coldspot model (balanced) where P/B is the production to biomass ratio per year, Q/B is the consumption to biomass ratio per year, EE is ecotrophic efficiency which indicates the exploitation of a compartment, and P/Q/year is the production to consumption ratio per year. *Gambierdiscus* spp. values changed based on the model and can be seen in Table 6-10

Compartment	Code	Trophic level	Biomass (g m^{-2})	P/B	Q/B	EE	P/Q (/year)
Algae feeders	F1	2.34	8	0.81266	24.9436	0.93529	0.0325797
Algae feeders and detritivores	F2	2.14	11.61246	1.0054	25.73	0.51099	0.039075
Amphipods/isopods/tanaids/pycnogonids	I1	2.45	45	5	125.5	0.83823	0.0398406
Anisotremus virginicus	F3	3.76	0.125	4.79412	10.3	0.95751	0.4654482
Ascidians/barnacles/bryozoans	I2	2.81	335	2.3	20	0.97845	0.115
Asteroids	I3	3.24	75	0.49	3.24	0.96195	0.1512346
Bivalves	I4	2.4	310	2.23	9.5	0.97436	0.2347368
Blenniidae H	F4	2.02	1.5	2.84	36.1	0.7289	0.0786704
Calamus pennatula	F5	3.7	0.001	7.25	9.3	2.19E-05	0.7795699
Caranx ruber	F6	3.86	0.25	1.17	10.1	0.98831	0.1158416
Cephalopholis fulva	F7	4.09	0.578386	2.1	7.8	0.57588	0.2692308
Chitons/scaphopods	I5	2.42	185	0.36	11.7	0.97158	0.0307692
Corals/sea anemones	I6	2.4	375	1.09	9	0.97949	0.1211111
Crab, brittle star, and polychaete feeders	F8	3.66	13.11909	0.83136	7.03711	0.73078	0.1181396
Crabs	I7	2.57	80	1.6	14	0.99496	0.1142857
Crustacean feeders and zooplanktivores	F9	3.77	5.564583	1.3226	10.2953	0.59525	0.1284668
Decomposers/microfauna	I8	2	45	280	1900	0.84281	0.1473684
Echinoids	I9	2.23	225	1.1	3.7	0.95514	0.2972973
Engraulidae H	F10	2.39	7	2.835	43.4	0.93179	0.0653226
Fish and crustacean feeders	F11	4.01	4.4375	0.57414	10.5471	0.23053	0.054436
Gastropods	I10	2.67	150	2.8	14	0.95877	0.2
Haemulon plumieri	F12	3.62	1	0.67	9.4	0.87979	0.0712766
Hemiramphidae H	F13	2.55	3	1.23	39.1	0.86966	0.0314578
Holocentrus rufus	F14	3.73	1.142082	1.38	9.8	0.36751	0.1408163
Holothurids/sipunculids/ echiuroids/ hemichordates	I11	2.42	300	0.31	3.36	0.97932	0.0922619
Intermediate jacks C	F15	3.88	1.290417	1.60354	8.61841	0.49454	0.1860597
Intermediate reef fish C1	F16	3.72	2.640181	0.51694	6.97901	0.86385	0.0740709
Intermediate reef fish C2	F17	3.72	10	1.31838	2.81833	0.95943	0.4677884
Intermediate reef fish C3	F18	3.69	11	0.64	6.39956	0.97718	0.1000069
Intermediate reef fish C4	F19	3.88	1.237181	0.55	5.74686	0.87005	0.0957044
Intermediate reef fish H	F20	2.03	8.517667	0.765	26.85	0.50767	0.0284916
Intermediate scaridae H	F21	2	4.766669	1.11	20.05	0.61065	0.0553616
Kyphosidae H	F22	2	2.42	0.6	23.6	0.49352	0.0254237
Lachnolaimus maximus	F23	3.56	0.7808145	0.34931	4.8	0.31365	0.0727735
Large groupers C	F24	4.15	0.725	0.37	2.3	0.41241	0.1608696
Large jacks C	F25	4.3	0.181	0.525	5.7	0.76717	0.0921053
Large reef fish	F26	4.01	2.5	0.38	3.9889	0.98101	0.0952644
Large scaridae H	F27	2.01	18.88242	0.85	13.5	0.2495	0.062963
Large sharks/rays C	F28	4.09	0.3	0.24	4.82833	0.70163	0.0497066
Large to intermediate schooling fish P	F29	3.6	28	0.68	12.4	0.98408	0.0548387
Lobsters	I12	3.17	12	1	7.4	0.89152	0.1351351
Lutjanus jocu	F30	4.24	0.4794444	0.64689	5	0.1789	0.1293785
Malacanthus plumieri	F31	3.84	0.001	2.5	6.8	2.10E-05	0.3676471
Octopuses	I13	3.6	18	1.9	6.76	0.95908	0.2810651
Ocyurus chrysurus	F32	3.87	3.327953	0.61985	7.9	0.20545	0.0784624
Polychaetes/priapuloids/ophiuroids	I14	2.67	85	5.2	61.6	0.93932	0.0844156
Polyp and benthic invertebrate feeders	F33	3.56	1	1.75865	13.5434	0.88315	0.1298535
Scarus taeniopterus	F34	2.03	0.85	1.2	20.8	0.90688	0.0576923

Compartment	**Code**	**Trophic level**	**Biomass (g m^{-2})**	**P/B**	**Q/B**	**EE**	**P/Q (/year)**
Scomberomorus cavalla	F35	4.57	0.001	0.63	7.4	0.5801	0.0851351
Scomberomorus regalis	F36	4.57	0.001	0.47	10.8	0.47907	0.0435185
Sea birds	B1	4.37	0.05	5.4	80	0.9353	0.0675
Sea turtles	R1	3.14	4	0.15	3.5	0.86989	0.0428571
Sharks/scombrids C	F37	4.32	0.45	0.29	9.15	0.96332	0.031694
Shrimps/hermit crabs/stomatopods	I15	3.42	35	2.8	26.9	0.96867	0.1040892
Small gobiidae C	F38	3.45	0.75	3.14	17.45	0.89605	0.1799427
Small jacks C	F39	3.75	1.045	0.83	12.5	0.90382	0.0664
Small reef fish C1	F40	3.54	20	1.28511	9.75087	0.91364	0.1317939
Small reef fish C2	F41	3.77	5	3.82	14.65	0.89712	0.2607509
Small reef fish O1	F42	2.94	6	1.6	18.2	0.94713	0.0879121
Small reef fish O2	F43	3.32	2	1.505	16	0.97998	0.0940625
Small reef fish O3	F44	2.82	3	2.53	39.7	0.86371	0.063728
Small scaridae H	F45	2	4.850833	0.94	33.9	0.37407	0.0277286
Small schooling fish P	F46	3.75	18	3.54	18.85	0.98834	0.1877984
Sparisoma viride	F47	2	1	1.155	20.7	0.95793	0.0557971
Sphyraena barracuda	F48	4.47	0.9076044	0.23194	3.3	0.26357	0.070286
Sponge feeders	F49	3.79	2.25	0.58887	6.80562	0.36752	0.0865272
Sponges	I16	3	800	1.5	5	0.72482	0.3
Squids	I17	4.07	5	1.3	17.5	0.981	0.0742857
Zooplanktivores and benthic worm feeders	F50	3.56	0.002	6.5	17.2	0.84113	0.3779069
Zooplankton	I18	2.75	80	40	165	0.95988	0.2424242
Phytoplankton	A1	1	80	70		0.92688	
Gambierdiscus spp.	A2						
Benthic autotrophs	A3	1	1300	13.25		0	
Detritus	D1	1	10000			0.1372	

Table 6-8 The new, condensed diet composition matrix for the coldspot.

Compartment	Code	Compartment	Code
Algae feeders	F1	Large scaridae H	F27
Algae feeders and detritivores	F2	Large sharks/rays C	F28
Amphipods/isopods/tanaids/pycnogonids	I1	Large to intermediate schooling fish P	F29
Anisotremus virginicus	F3	Lobsters	I12
Ascidians/barnacles/bryozoans	I2	*Lutjanus jocu*	F30
Asteroids	I3	*Malacanthus plumieri*	F31
Bivalves	I4	Octopuses	I13
Blenniidae H	F4	*Ocyurus chrysurus*	F32
Calamus pennatula	F5	Polychaetes/priapuloids/ophiuroids	I14
Caranx ruber	F6	Polyp and benthic invertebrate feeders	F33
Cephalopholis fulva	F7	*Scarus taeniopterus*	F34
Chitons/scaphopods	I5	*Scomberomorus cavalla*	F35
Corals/sea anemones	I6	*Scomberomorus regalis*	F36
Crab, brittle star, and polychaete feeders	F8	Sea birds	B1
Crabs	I7	Sea turtles	R1
Crustacean feeders and zooplanktivores	F9	Sharks/scombrids C	F37
Decomposers/microfauna	I8	Shrimps/hermit crabs/stomatopods	I15
Echinoids	I9	Small gobiidae C	F38
Engraulidae H	F10	Small jacks C	F39
Fish and crustacean feeders	F11	Small reef fish C1	F40
Gastropods	I10	Small reef fish C2	F41
Haemulon plumieri	F12	Small reef fish O1	F42
Hemiramphidae H	F13	Small reef fish O2	F43
Holocentrus rufus	F14	Small reef fish O3	F44
Holothurids/sipunculids/echiuroids/ hemichordates	I11	Small scaridae H	F45
Intermediate jacks C	F15	Small schooling fish P	F46
Intermediate reef fish C1	F16	*Sparisoma viride*	F47
Intermediate reef fish C2	F17	*Sphyraena barracuda*	F48
Intermediate reef fish C3	F18	Sponge feeders	F49
Intermediate reef fish C4	F19	Sponges	I16
Intermediate reef fish H	F20	Squids	I17
Intermediate scaridae H	F21	Zooplanktivores and benthic worm feeders	F50
Kyphosidae H	F22	Zooplankton	I18
Lachnolaimus maximus	F23	Phytoplankton	A1
Large groupers C	F24	*Gambierdiscus* spp.	A2
Large jacks C	F25	Benthic autotrophs	A3
Large reef fish	F26	Detritus	D1

Predator/Prey Matrix	**Code**	**F1**	**F2**	**I1**	**F3**	**I2**	**I3**
Algae feeders	**F1**	0.00098	0.0024	0	0.00051	0.01319	0
Algae feeders and detritivores	**F2**	0	0	0.01531	0	0	0
Amphipods/isopods/tanaids/pycnogonids	**I1**	0	0	0	0	0.05	0
Anisotremus virginicus	**F3**	0	0	0.14986	0	0.015	0
Ascidians/barnacles/bryozoans	**I2**	0	0	0	0	0	0
Asteroids	**I3**	0	0	0	0	0.02	0.08
Bivalves	**I4**	0	0	0	0	0	0
Blenniidae H	**F4**	0	0	0	0	0	0
Calamus pennatula	**F5**	0	0	0	0	0	0
Caranx ruber	**F6**	0.11817	0.05351	0	0	0	0
Cephalopholis fulva	**F7**	0.04381	0.06571	0	0	0	0
Chitons/scaphopods	**I5**	0	0	0	0	0.04	0
Corals/sea anemones	**I6**	0	0	0	0	0	0
Crab, brittle star, and polychaete feeders	**F8**	0.00011	0.00129	0.01785	0	0	0
Crabs	**I7**	0	0	0.005	0	0.02	0
Crustacean feeders and zooplanktivores	**F9**	0	0.00853	0.05076	0	0.0513	0
Decomposers/microfauna	**I8**	0	0	0	0	0	0
Echinoids	**I9**	0	0	0	0	0	0.006
Engraulidae H	**F10**	0	0	0	0	0	0
Fish and crustacean feeders	**F11**	0.02099	0.00859	0	0.00134	0	0
Gastropods	**I10**	0	0	0.006	0	0.058	0
Haemulon plumierii	**F12**	0.00334	0.00661	0.03506	0.00117	0	0
Hemiramphidae H	**F13**	0	0	0	0	0	0
Holocentrus rufus	**F14**	0.0008	0.0008	0.01903	0	0	0
Holothurids/sipunculids/ echiuroids/ hemichordates	**I11**	0	0	0	0	0	0
Intermediate jacks C	**F15**	0.08953	0.05836	0	0.00501	0	0
Intermediate reef fish C1	**F16**	0.00024	0.00048	0.00022	8.5E-05	0.04874	0.00381
Intermediate reef fish C2	**F17**	0.0136	0.0016	0.03067	0.00053	0.05066	0
Intermediate reef fish C3	**F18**	0.02317	0.01157	0.04229	0.00027	0.03988	0
Intermediate reef fish C4	**F19**	0	0	0	0	0.02249	0.00191
Intermediate reef fish H	**F20**	0	0	0	0	0	0.002
Intermediate scaridae H	**F21**	0	0	0	0	0	0
Kyphosidae H	**F22**	0	0	0	0	0	0
Lachnolaimus maximus	**F23**	0	0	0.01	0	0.005	0
Large groupers C	**F24**	0.08477	0.0081	0	0.00241	0	0
Large jacks C	**F25**	0.00472	0.01792	0	0.00442	0	0
Large reef fish	**F26**	0.00451	0.00899	0	0.00076	0	0
Large scaridae H	**F27**	0	0	0	0	0	0
Large sharks/rays C	**F28**	0.01169	0.07437	0	0.00025	0	0
Large to intermediate schooling fish P	**F29**	0.001	0.004	0	0	0	0
Lobsters	**I12**	0	0	0	0	0	0
Lutjanus jocu	**F30**	0.02601	0	0	0	0	0
Malacanthus plumieri	**F31**	0	0	0.016	0	0	0
Octopuses	**I13**	0.0002	0.0012	0	5.6E-05	0	0
Ocyurus chrysurus	**F32**	0	0	0.00778	0	0.02978	0
Polychaetes/priapuloids/ophiuroids	**I14**	0	0	0.015	0	0.04	0
Polyp and benthic invertebrate feeders	**F33**	0	0	0.00255	0	0.02634	0
Scarus taeniopterus	**F34**	0	0	0	0	0	0
Scomberomorus cavalla	**F35**	0	0	0	0	0	0
Scomberomorus regalis	**F36**	0	0	0	0.12813	0	0

Code	**I4**	**F4**	**F5**	**F6**	**F7**	**I5**	**I6**	**F8**	**I7**	**F9**
F1	0	0.00051	0	0	0.00051	0	0.00759	0.00287	0.02147	0.0024
F2	0.00159	0	0	0	0	0	0.00645	0	0	0
I1	0	0	0	0	0	0	0.04	0	0	0
F3	0.055	0	0	0	0	0.01	0	0	0.16929	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0.145	0	0	0	0	0.005	0.012	0	0.01	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0.123	0	0	0	0	0	0	0	0.215	0
F6	0	0.05351	0	0	0	0	0	0.10702	0.00433	0
F7	0	0.0219	0	0	0	0	0	0	0.18433	0
I5	0	0	0	0	0	0	0.008	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0.07846	0.00067	0	0	0	0.01446	0	0.00073	0.3087	0.00129
I7	0.05	0	0	0	0	0	0.01	0.00069	0.005	0.00069
F9	0	0.00446	0	0	0	0	0	0.00446	0.04566	0.00446
I8	0	0	0	0	0	0	0	0	0	0
I9	0.01	0	0	0	0	0	0.05	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0	0	0	0.00927	0.0028	0	0.04695	0.34842	0.01899
I10	0.04	0	0	0	0	0.02	0.015	0	0.002	0
F12	0.01356	0.00117	0	0	0.00117	0.00756	0	0.00661	0.27381	0.00552
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0.00042	0	0	0	0.00603	0	0.00119	0.57303	0.0008
I11	0	0	0	0	0	0	0	0	0	0
F15	0	0.04606	1.00E-10	0.0027	0.0027	0	0	0.02399	0.00358	0.01459
F16	0.01764	8.5E-05	1.00E-10	0	0	0.00029	0.00844	0.00056	0.0826	0.0004
F17	0.0722	0	1.00E-10	0.00013	0.00043	0.0198	0.00887	0.00195	0.0989	0.00094
F18	0.02294	0.002	1.00E-10	0	0.00034	0.00998	0.003	0.0018	0.14951	0.00203
F19	0	0	0	0	0	0	0.02988	0	0.00096	0
F20	0	0	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0.426	0	0	0	0	0.006	0	0	0.061	0
F24	0	0	1.00E-10	0.00135	0	0	0	0.00703	0.0701	0.00491
F25	0.05732	0	1.00E-10	0.01907	0.00377	0	0	0.01607	0.01032	0.00787
F26	0.05336	0	1.00E-10	0.00216	0.00082	0	0	0.14751	0.02279	0.00516
F27	0	0	0	0	0	0	0.001	0	0	0
F28	0.05503	0	1.00E-10	0.00517	0.00061	0	0	0.00237	0.04108	0.00277
F29	0	0	0	0	0	0	0	0	0	0
I12	0.5	0	0	0	0	0	0	0	0	0
F30	0	0	0	0	0.01445	0	0	0.03212	0.1585	0.0289
F31	0	0	0	0	0	0.057	0	0.0372	0.185	0
I13	0.4	0	1.00E-10	0	8.8E-05	0.01	0	0.00072	0.025	0.00062
F32	0.01478	0	0	0	0	0	0	0	0.20083	0
I14	0.03	0	0	0	0	0	0.01	0	0	0
F33	0	0	0	0	0	0	0.48227	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0.15383	0	0	0	0	0	0.01398
F36	0	0	0	0.06407	0	0	0	0.02402	0	0.13614

Code	**I8**	**I9**	**F10**	**F11**	**I10**	**F12**	**F13**	**F14**	**I11**	**F15**
F1	0.00077	0	0.00051	0	0.04023	0	0.00051	0.00051	0	0
F2	0	0	0	0	0.0023	0	0	0	0	0
I1	0.04	0	0	0	0	0	0	0	0	0
F3	0.005	0	0	0	0.038	0	0	0	0.004	0
I2	0.1	0	0	0	0	0	0	0	0	0
I3	0.02	0.07	0	0	0.135	0	0	0	0.005	0
I4	0.1	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0.001	0	0	0	0	0
F5	0	0.04	0	0	0.082	0	0	0	0.09	0
F6	0	0	0.10434	0	0.004	0	0	0	0	0
F7	0	0	0	0	0	0	0	0	0	0
I5	0.3	0	0	0	0	0	0	0	0	0
I6	0.1	0	0	0	0	0	0	0	0	0
F8	0	0.11646	0.00067	0	0.10204	0	0	0	0.01077	0
I7	0.03	0.05	0	0	0.05	0	0	0	0.001	0
F9	0	0.00937	0.00446	0	0.08617	0	0	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0.015	0	0	0	0	0	0	0.005	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0	0	0.00323	0.05769	0.00235	0	0	0	0
I10	0.03	0	0	0	0.02	0	0	0	0.015	0
F12	0	0.12456	0.00117	0	0.06956	0	0.00117	0.00117	0.14356	0
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0	0.00042	0	0.07803	0	0.00042	0	0	0
I11	0.1	0	0	0	0	0	0	0	0	0
F15	0	0	0.09023	0.01002	0.00358	0.00501	0.0027	0.00501	0	0.0051
F16	0	0.19241	8.5E-05	0.00016	0.01687	8.5E-05	8.5E-05	8.5E-05	0.00434	0
F17	0	0.0989	0.002	0.00043	0.03529	0.00053	1.9E-05	0.00053	0.05935	0.00088
F18	0	0.05981	0.002	0.002	0.004	0.00027	0	0.00027	0.00998	0
F19	0	0.00287	0	0	0.00026	0	0	0	0.00574	0
F20	0	0	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0.046	0	0	0.397	0	0	0	0	0
F24	0	0	0	0.01427	0	0.00241	0	0.00241	0	0.00885
F25	0	0.08232	0	0.0371	0.15832	0.00442	0	0.00442	0	0.13157
F26	0	0.07114	0.00074	0.00438	0.28447	0.00076	0	0.00076	0	0.01496
F27	0	0	0	0	0	0	0	0	0	0
F28	0	0.0066	0.02302	0.01429	0.00824	0.00025	0.00168	0.00025	0.04108	0.03595
F29	0	0	0.006	0	0	0	0	0	0.12	0
I12	0	0	0	0	0.15	0	0	0	0	0
F30	0	0	0	0	0.036	0.0289	0	0.0289	0	0
F31	0	0.027	0	0	0	0	0	0	0.104	0
I13	0	0	0	8.8E-05	0.5	5.6E-05	0	5.6E-05	0	0
F32	0	0	0	0	0.08778	0	0	0	0	0
I14	0.05	0	0	0	0.01	0	0	0	0	0
F33	0	0	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0.15383	0	0	0	0	0	0	0.09789
F36	0	0	0.06407	0	0	0	0.06407	0	0	0

Code	F16	F17	F18	F19	F20	F21	F22	F23	F24	F25
F1	0	0.00098	0.00146	0	0.00051	0	0	0	0	0
F2	0	0	0	0	0	0	0	0	0	0
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0	0	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0	0	0
F6	0	0	0	0.01987	0	0.03535	0	0	0	0
F7	0	0	0.06571	0	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0	0.00067	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0	0	0
F9	0	0.00446	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0.00189	0.02796	0.05604	0.00362	0	0.01575	0	0	0	0
I10	0	0	0	0	0	0	0	0	0	0
F12	0	0.00226	0.00334	0.00117	0.00117	0.00117	0	0	0	0
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0.00042	0.0008	0	0.00042	0	0	0	0	0
I11	0	0	0	0	0	0	0	0	0	0
F15	0.00564	0.0697	0.0051	0.00744	0.04647	0.04251	0.01497	0.0027	0	0
F16	0.0008	0.0072	0.04528	0.00253	0.00325	0.01273	0.00246	0	0	0
F17	0	0.01422	0.01279	0.00171	0.002	0.0198	0.00299	9.5E-05	0	0
F18	0.00666	0.00965	0.01031	0.00172	0.0045	0.00466	0.003	0	0	0
F19	0	0	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0.00267	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0.05677	0.06472	0	0.00582	0.0101	0	0.0201	0.01795	0	0
F25	0.13365	0.11519	0.1071	0	0.02232	0	0	0	0	0
F26	0.01425	0.02045	0.02452	0.00125	0.00821	0.00382	0.00216	0.0003	0	0.00288
F27	0	0	0	0	0	0	0	0	0	0
F28	0.05476	0.00593	0.01771	0.00942	0.02138	0.01885	0.04929	0.00193	0.07637	0.0025
F29	0	0	0	0	0.005	0	0	0	0	0
I12	0	0	0	0	0	0	0	0	0	0
F30	0.00321	0.07708	0.16219	0	0	0.02601	0	0	0	0
F31	0	0	0.0372	0	0	0	0	0	0	0
I13	0	0.00139	0.00259	0	0.001	0	0	8.2E-05	0	0
F32	0	0	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0	0	0
F33	0	0	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0	0	0
F36	0	0	0.06407	0	0	0	0	0	0	0

Code	**F26**	**F27**	**F28**	**F29**	**I12**	**F30**	**F31**	**I13**	**F32**	**I14**
F1	0	0	0	0.00051	0	0	1.00E-10	0	0.00051	0
F2	0	0	0	0	0	0	0	0	0	0.01651
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0	0	0.305
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0.03
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0	0	0.282
F6	0	0	0	0.05351	0	0	0	0	0	0
F7	0	0	0	0	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0	0	0	0	0	0	0	0.00116	0	0.20711
I7	0	0	0	0	0	0	0	0	0	0.004
F9	0	0	0	0	0	0	0	0	0	0.07049
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0.004
F10	0	0	0	0	0	0	0	0	0	0
F11	0.00859	0	0	0	0.00825	0	1.00E-10	0.0216	0	0
I10	0	0	0	0	0.002	0	0	0	0	0.05
F12	0	0	0	0.00117	0	0	1.00E-10	0	0.00117	0.20256
F13	0	0	0	0	0	0	0	0	0	0
F14	0	0	0	0.00042	0	0	0	0	0	0.11603
I11	0	0	0	0	0	0	0	0	0	0
F15	0	0.01143	0.0027	0.04851	0	0	1.00E-10	0	0.00277	0
F16	0	0.02379	0	8.5E-05	0.00037	0	1.00E-10	0.01107	8.5E-05	0.01615
F17	0.00078	0.0132	0	0.00101	0.00101	9.5E-05	1.00E-10	0.04946	0	0.17818
F18	0	0.03656	0	0.002	0	0	1.00E-10	0.02991	0.00084	0.04985
F19	0	0	0	0	0	0	0	0	0	0.00574
F20	0	0	0	0	0	0	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0.17857	0.16544	0.0051	0	0.1601	0.01795	0	0	0.00719	0
F25	0	0	0	0	0	0	1.00E-10	0	0.01699	0
F26	0.00277	0.00477	0	0.02848	0.00145	0.0003	1.00E-10	0.00643	0.00181	0
F27	0	0	0	0	0	0	0	0	0	0
F28	0.01898	0.03143	0.00824	0.02596	0.00414	0.00193	1.00E-10	0.02631	0.00688	0.04108
F29	0	0	0	0.003	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0	0	0.05
F30	0	0.00867	0	0.0289	0.0885	0	0	0.07	0	0
F31	0	0	0	0	0	0	0	0	0	0.323
I13	0.00077	0	0	0	0	8.2E-05	1.00E-10	0.02	0	0.005
F32	0	0	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0	0	0.03
F33	0	0	0	0	0	0	0	0	0	0.3916
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0.32165	0	0	0	0	0.15383	0
F36	0	0	0	0.11532	0	0	0	0	0	0

Code	F33	F34	F35	F36	B1	R1	F37	I15	F38	F39
F1	0.00146	0	0	0	0	0	0	0.01453	0.00051	0.00051
F2	0	0	0	0	0	0	0	0.00308	0	0
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0.21457	0	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0.168	0	0
F6	0	0.01115	0	0	0	0	0	0.04067	0	0
F7	0	0	0	0	0	0	0	0.35567	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0.00067	0	0	0	0	0	0	0.12469	0.00067	0
I7	0	0	0	0	0	0	0	0.01	0	0
F9	0	0	0	0	0	0	0	0.0449	0.00446	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0.00681	0	0	0	0	0	0.10697	0.00926	0
I10	0	0	0	0	0	0	0	0.004	0	0
F12	0.00334	0.00117	0	0	0	0	0	0.07181	0.00117	0.00117
F13	0	0	0	0	0	0	0	0	0	0
F14	0.00119	0	0	0	0	0	0	0.16203	0.00042	0
I11	0	0	0	0	0	0	0	0	0	0
F15	0.01441	0.01266	0	5.8E-06	0	0	0.0027	0.02567	0.0027	0.0027
F16	0.00024	0	0	0	0	0	0	0.02972	8.5E-05	0
F17	0.00154	0.00496	0	0	0	0	0	0.0198	0	0
F18	0.00078	0.00117	0	0	0	0	0	0.09968	0.004	0.00225
F19	0	0	0	0	0	0	0	0.00204	0	0
F20	0	0.00067	0	0	0.001	0.002	0	0	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0.049	0	0
F24	0.00703	0	3.2E-06	4.1E-06	0	0.0351	0.00344	0	0	0.0076
F25	0.01263	0	0	0	0	0	0	0.01532	0	0
F26	0.00222	0.00098	0	0	0	0	0	0.02136	0	0.0107
F27	0	0	0	0	0	0	0	0	0	0
F28	0.00067	0.00031	3.6E-05	3.8E-07	0.01645	0.00414	0.00551	0.04108	0	0.0025
F29	0	0	0	0	0	0	0	0.004	0	0
I12	0	0	0	0	0	0	0	0	0	0
F30	0	0.00867	0	0	0	0	0	0	0	0
F31	0	0	0	0	0	0	0	0.164	0	0
I13	0.00016	0	0	0	0	0	0	0.02	0	0
F32	0	0	0	0	0	0	0	0.09158	0	0
I14	0	0	0	0	0	0	0	0	0	0
F33	0	0	0	0	0	0	0	0.01641	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0	0	0.02797
F36	0	0	0	0	0	0	0	0.016	0	0.06407

Code	**F40**	**F41**	**F42**	**F43**	**F44**	**F45**	**F46**	**F47**	**F48**	**F49**
F1	0.00098	0.00051	0.00051	0.00051	0.00051	0.00051	0.00051	0	0	0
F2	0	0	0	0	0	0	0	0	0	0
I1	0	0	0	0	0	0	0	0	0	0
F3	0	0	0	0	0	0	0	0	0	0
I2	0	0	0	0	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0	0	0
F4	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0	0	0
F6	0	0	0.02446	0.06077	0.05351	0.01115	0.10434	0.05351	0	0
F7	0.13143	0	0.06571	0.06571	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
F8	0.00129	0.00067	0.00067	0.00067	0.00192	0.00011	0.00067	0	0	0
I7	0.00862	0.007	0	0	0	0	0	0	0	0
F9	0.00853	0.00446	0.00446	0.00446	0.00446	0	0.00446	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0.02449	0.04149	0.04564	0.00208	0.04772	0.00712	0.02077	0.00712	0	0
I10	0	0	0	0	0	0	0	0	0	0
F12	0.00226	0.00117	0.00117	0.00117	0.00117	0.00117	0.00117	0	0	0.00117
F13	0	0	0	0	0	0	0.2	0	0	0
F14	0.0008	0.00042	0.00042	0.00042	0.00042	0.00042	0.00042	0	0	0
I11	0	0	0	0	0	0	0	0	0	0
F15	0.0051	0.00352	0.03739	0.04606	0.04606	0.03597	0.09268	0.01266	0	0.00931
F16	0.00411	0.00238	0.01589	8.5E-05	8.5E-05	0.01273	0.04899	8.5E-05	0	0.00024
F17	0	0.00101	0	0	0	0.00331	0.06684	0.00496	9.5E-05	0.00129
F18	0.0086	0.00998	0.03191	0.00101	0.01496	0.002	0.002	0.00117	0	0.00212
F19	0	0	0	0	0	0	0	0	0	0
F20	0	0	0	0	0	0	0	0.00067	0	0
F21	0	0	0	0	0	0	0	0	0	0
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0	0.01795	0.01147
F25	0	0	0	0	0	0	0	0	0	0.01699
F26	0.00064	0	0.15415	0.0043	0.00714	0.00027	0.04199	0.00098	0.0003	0.00272
F27	0	0	0	0	0	0	0	0	0	0
F28	0.01419	0.00496	0.03417	0.00332	0.00086	0.01803	0.05797	0.00031	0.00193	0.01391
F29	0	0	0	0	0	0	0.047	0	0	0
I12	0	0	0	0	0	0	0	0	0	0
F30	0	0	0	0	0	0.00867	0.05781	0.03758	0	0.0289
F31	0.0372	0.0124	0	0	0	0	0	0	0	0
I13	0.00604	0.001	0.0016	0.002	0	0	0	0	8.2E-05	0
F32	0	0	0	0	0	0	0.07828	0	0	0
I14	0	0	0	0	0	0	0	0	0	0
F33	0	0	0	0	0	0	0	0	0	0
F34	0	0	0	0	0	0	0	0	0	0
F35	0	0	0	0	0	0	0	0	0	0
F36	0.02403	0.00801	0	0	0	0	0.20501	0	0	0

Code	I16	I17	F50	I18	A1	A2	A3	D1
F1	0.00087	0	2.77E-05	0.06422	0	0	0.81484	
F2	0.00277	0	0	0	0.00018	0	0.50646	0.44535
I1	0.06	0	0	0.04	0	0	0.6	0.17
F3	0	0	0	0.03429	0	0	0	0
I2	0	0	0	0.15	0.2	0	0	0.55
I3	0.02	0	0	0	0	0	0.088	0.36
I4	0	0	0	0	0.15	0	0	0.75
F4	0	0	0	0.005	0	0	0.497	0.497
F5	0	0	0	0	0	0	0	0
F6	0	0.014	0	0.07283	0	0	0	0
F7	0	0	0	0	0	0	0	0
I5	0.02	0	0	0	0	0	0.632	0
I6	0	0	0	0.15	0	0	0.65	0.1
F8	0	0	4.44E-05	0.00551	0	0	0	0
I7	0.05	0	0	0.03	0.02	0	0.6	0.048
F9	0	0	8.4E-06	0.57508	0	0	0	0
I8	0	0	0	0	0	0	0.05	0.95
I9	0.05	0	0	0	0	0	0.86	0
F10	0	0	0	0.2	0.7	0	0	0.1
F11	0	0	4.4E-06	0.0159	0	0	0	0
I10	0.04	0	0	0	0	0	0.4	0.298
F12	0	0	5.50E-05	0	0	0	0	0
F13	0	0	0	0	0	0	0.8	0
F14	0	0	4.94E-07	0.03403	0	0	0	0
I11	0	0	0	0	0.01	0	0.13	0.76
F15	0	0.00358	1.0763043	0.0183	0	0	0	0
F16	0.15802	0.0008	3E-06	0.19752	0	0	0.02544	0
F17	0.06924	0	0	0.02051	0	0	0.01089	0
F18	0.06978	0	0	0.14993	0	0	0.05184	0
F19	0.88193	0	0	0.00096	0	0	0.04521	0
F20	0	0	0	0	0	0	0.533	0.458
F21	0.001	0	0	0	0	0	0.999	0
F22	0	0	0	0	0	0	1	0
F23	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0
F26	0	0.00074	0	0.0043	0	0	0	0
F27	0.002	0	0	0	0	0	0.997	0
F28	0	0.01399	0	0	0	0	0	0.01845
F29	0	0	0	0.722	0	0	0.088	0
I12	0	0	0	0	0	0	0.18	0.12
F30	0	0.022	0	0.018	0	0	0	0
F31	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0
F32	0	0.02178	0	0.46743	0	0	0	0
I14	0.05	0	0	0.05	0.1	0	0.245	0.37
F33	0	0	0	0.08083	0	0	0	0
F34	0.015	0	0	0	0	0	0.985	0
F35	0	0.077	0	0	0	0	0	0
F36	0	0.023	0	0	0	0	0	0

Predator/Prey Matrix	**Code**	**F1**	**F2**	**I1**	**F3**	**I2**	**I3**
Sea birds	**B1**	0.00315	0.00515	0	0.00118	0	0
Sea turtles	**R1**	0	0	0	0	0.02	0.02
Sharks/scombrids C	**F37**	0.01347	0.01093	0	0.00116	0	0
Shrimps/hermit crabs/stomatopods	**I15**	0	0	0.04001	0.00027	0	0.01001
Small gobiidae C	**F38**	0	0	1	0	0	0
Small jacks C	**F39**	0.002	0.008	0	0	0	0
Small reef fish C1	**F40**	0.00133	0.00101	0.04202	5.30E-05	0	0
Small reef fish C2	**F41**	0.0001	0.0006	0.1	5.31E-05	0	0
Small reef fish O1	**F42**	0.00267	0.0012	0.056	7.92E-05	0.02	0
Small reef fish O2	**F43**	0	0	0.035	0	0.02	0.021
Small reef fish O3	**F44**	0	0	0.008	0	0	0
Small scaridae H	**F45**	0	0	0	0	0	0
Small schooling fish P	**F46**	0	0	0.01	0	0.03	0
Sparisoma viride	**F47**	0	0	0	0	0	0
Sphyraena barracuda	**F48**	0.01508	0.05026	0	0	0	0
Sponge feeders	**F49**	0	0	0	0	0.07304	0
Sponges	**I16**	0	0	0	0	0	0
Squids	**I17**	0	0	0	0	0	0
Zooplanktivores and benthic worm feeders	**F50**	0	0	0.5	0	0	0
Zooplankton	**I18**	0	0	0	0	0	0

Code	**I4**	**F4**	**F5**	**F6**	**F7**	**I5**	**I6**	**F8**	**I7**	**F9**
B1	0	0	1.00E-10	0.00765	0.00098	0	0	0.00579	0	0.01123
R1	0.02	0	0	0	0	0	0.016	0	0.025	0
F37	0.00713	0.00113	1.00E-10	0.00438	0.00247	0	0	0.00692	0.02013	0.01737
I15	0.21201	0	1.00E-10	0	0	0.01001	0	0.00147	0.00201	0.00096
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0.00025	0	0	0	0	0	0.00025
F40	0.05999	0.006	1.00E-10	0	0	0.03999	0	0.00036	0.10003	0.00026
F41	0.03	0.001	1.00E-10	0	0	0.002	0	0.00071	0.063	0.0006
F42	0.02	0.004	1.00E-10	0	0	0	0.02	0.00051	0.01	0.00036
F43	0.027	0.003	0	0	0	0	0.1	0.00021	0.05	0.00021
F44	0.071	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0.001	0	0	0
F48	0	0	0	0.05026	0.00457	0	0	0.00457	0	0.0297
F49	0	0	0	0	0	0	0.05636	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	**I8**	**I9**	**F10**	**F11**	**I10**	**F12**	**F13**	**F14**	**I11**	**F15**
B1	0	0	0.10015	0.00265	0	0.00118	0.10015	0.00118	0	0.05265
R1	0	0.02	0	0	0.025	0	0	0	0	0
F37	0	0	0.04013	0.00497	0.00813	0.00116	0.11513	0.00116	0	0.02988
I15	0.05501	0.11001	0.01001	0	0.08001	0.00027	0	0.00027	0.00501	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0.019	0	0	0	0	0	0	0.00175
F40	0	0.10997	0.002	0	0.14396	5.3E-05	0.002	5.3E-05	0.008	0
F41	0	0	0	0	0.005	5.3E-05	0	5.3E-05	0.001	0
F42	0	0	0.004	0	0.038	7.9E-05	0.003	7.9E-05	0	0
F43	0.005	0.05	0.003	0	0.035	0	0	0	0.005	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0.006	0	0.002	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0	0	0	0	0.00457	0.05026	0	0	0.07768
F49	0	0	0	0	0	0	0	0	0	0
I16	0.001	0	0	0	0	0	0	0	0	0
I17	0	0	0.03	0	0	0	0.015	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0.6	0	0	0	0	0	0	0	0	0

Code	**F16**	**F17**	**F18**	**F19**	**F20**	**F21**	**F22**	**F23**	**F24**	**F25**
B1	0.00682	0.02887	0.02598	0.00301	0.00515	0.00348	0	0.00054	0	0.01015
R1	0	0	0	0	0	0	0	0	0	0
F37	0.01013	0.02885	0.07247	0.00356	0.01013	0.0008	0.02013	0.00121	0	0
I15	0	0.00719	0	0	0	0	0	0	0	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0	0.01	0	0	0	0	0
F40	0	0.00144	0	0	0	0	0	0	0	0
F41	0	0.00144	0	0	0.0005	0	0	0	0	0
F42	0	0.00216	0	0	0	0	0	0	0	0
F43	0	0	0	0	0	0	0	0	0	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0.00457	0.13251	0.05483	0	0	0.01508	0	0	0	0
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	**F26**	**F27**	**F28**	**F29**	**I12**	**F30**	**F31**	**I13**	**F32**	**I14**
B1	0.004	0.00148	0	0.28015	0	0.00054	1.00E-10	0	0.00098	0
R1	0	0	0	0	0.06	0	0	0	0	0
F37	0.0109	0.02013	0	0.17613	0.00513	0.00121	1.00E-10	0.04513	0.00138	0.00613
I15	0	0	0	0	0.00501	0	0	0.02501	0	0.05001
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0.014	0	0	0	0	0	0
F40	0	0	0	0.002	0	0	0	0	0	0.09997
F41	0	0	0	0	0	0	0	0.02	0	0.079
F42	0	0	0	0.003	0	0	0	0	0	0.05
F43	0	0	0	0	0	0	0	0	0	0.1
F44	0	0	0	0	0	0	0	0	0	0.05
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0.00503	0	0.15536	0	0	0	0.026	0.05026	0
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0.15	0	0	0	0	0	0
F50	0	0	0	0	0	0	0	0	0	0.5
I18	0	0	0	0	0	0	0	0	0	0

Code	**F33**	**F34**	**F35**	**F36**	**B1**	**R1**	**F37**	**I15**	**F38**	**F39**
B1	0.00323	0.00098	4.06E-05	1.4E-06	0	0	0.01348	0	0	0.02265
R1	0	0	0	0	0	0	0	0.04	0	0
F37	0.00321	0.0003	3.52E-05	3.6E-05	0	0	0.0068	0.05013	0	0.03763
I15	0.00078	0	0	0	0	0	0	0.00201	0	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0	0	0	0	0	0	0.00075
F40	0.00016	0	0	0	0	0	0	0.04015	0.002	0
F41	0.00016	0	0	0	0	0	0	0.15	0.002	0
F42	0.00023	0	0	0	0	0	0	0.01	0.003	0
F43	0	0	0	0	0	0	0	0.04	0.002	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0.00503	0	0	0	0	0	0	0	0.03427
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0.1	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	**F40**	**F41**	**F42**	**F43**	**F44**	**F45**	**F46**	**F47**	**F48**	**F49**
B1	0.0217	0.00815	0.00415	0.00315	0	0.00082	0.20015	0.00098	0.00054	0.00313
R1	0	0	0	0	0	0	0	0	0	0
F37	0.01738	0.02013	0.01133	0.00113	0	0.0008	0.13613	0.0003	0.00121	0.00396
I15	0.00863	0.00501	0	0	0	0	0	0	0	0
F38	0	0	0	0	0	0	0	0	0	0
F39	0	0	0	0	0	0	0.006	0	0	0
F40	0.00259	0.002	0.004	0.002	0.006	0.00067	0.002	0	0	0
F41	0.0069	0.003	0.0008	0	0.004	0	0	0	0	0
F42	0.00345	0.004	0.0048	0.004	0.004	0.00134	0.004	0	0	0
F43	0.00259	0.003	0	0.003	0.003	0	0.003	0	0	0
F44	0	0	0	0	0	0	0	0	0	0
F45	0	0	0	0	0	0	0	0	0	0
F46	0	0	0	0	0	0	0.002	0	0	0
F47	0	0	0	0	0	0	0	0	0	0
F48	0	0	0	0.05026	0	0.00503	0.15079	0.00503	0	0
F49	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0.13	0	0	0
F50	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0

Code	**I16**	**I17**	**F50**	**I18**	**A1**	**A2**	**A3**	**D1**
B1	0	0	0	0.05315	0	0	0	0
R1	0.34	0	0	0	0	0	0.414	0
F37	0	0.01013	0	0	0	0	0	0
I15	0	0.00501	0	0.15001	0	0	0.08401	0.12001
F38	0	0	0	0	0	0	0	0
F39	0	0	0	0.938	0	0	0	0
F40	0	0	1.5E-07	0.25993	0	0	0.05799	0
F41	0	0	0	0.5	0	0	0.028	0
F42	0.02	0	0	0.1	0	0	0.326	0.28
F43	0.08	0	0	0.109	0	0	0.2	0.1
F44	0	0	0	0.2	0	0	0.335	0.336
F45	0	0	0	0	0	0	1	0
F46	0	0	0	0.95	0	0	0	0
F47	0.001	0	0	0	0	0	0.998	0
F48	0	0	0	0.019	0	0	0	0
F49	0.77805	0	0	0.01266	0	0	0.07989	0
I16	0	0	0	0	0	0	0	0.999
I17	0	0.01	0	0.565	0	0	0	0
F50	0	0	0	0	0	0	0	0
I18	0	0	0	0	0.2	0	0	0.2

Table 6-9 Diet composition matrix for two species of *Gambierdiscus* for the coldspot and hotspot models for 100 cells g ww^{-1} algae.

Coldspot			Coldspot			Hotspot			Hotspot		
100 Cells g ww-1 algae			100 Cells g ww-1 algae			100 Cells g ww-1 algae			100 Cells g ww-1 algae		
Gambierdiscus carolinianus			*Gambierdiscus excentricus*			*Gambierdiscus carolinianus*			*Gambierdiscus excentricus*		
Code	**A2**	**A3**	**Code**	**A2**	**A3**	**Code**	**A2**	**A3**	**Code**	**A2**	**A3**
F1	0.00341	0.81142	**F1**	0.00352	0.81131	**F1**	0.00341	0.81138	**F1**	0.00352	0.81127
F2	0.00212	0.50434	**F2**	0.00219	0.50427	**F2**	0.00212	0.50434	**F2**	0.00219	0.50427
I1	0.00251	0.59749	**I1**	0.00259	0.59741	**I1**	0.00251	0.59749	**I1**	0.00259	0.59741
F3	0	0	**F3**	0	0	**F3**	0	0	**F3**	0	0
I2	0	0	**I2**	0	0	**I2**	0	0	**I2**	0	0
I3	0.00037	0.08763	**I3**	0.00038	0.08762	**I3**	0.00037	0.08763	**I3**	0.00038	0.08762
I4	0	0	**I4**	0	0	**I4**	0	0	**I4**	0	0
F4	0.00208	0.49492	**F4**	0.00215	0.49485	**F4**	0.00208	0.49492	**F4**	0.00215	0.49485
F5	0	0	**F5**	0	0	**F5**	0	0	**F5**	0	0
F6	0	0	**F6**	0	0	**F6**	0	0	**F6**	0	0
F7	0	0	**F7**	0	0	**F7**	0	0	**F7**	0	0
I5	0.00265	0.62935	**I5**	0.00273	0.62927	**I5**	0.00265	0.62935	**I5**	0.00273	0.62927
I6	0.00272	0.64728	**I6**	0.00281	0.64719	**I6**	0.00272	0.64728	**I6**	0.00281	0.64719
F8	0	0	**F8**	0	0	**F8**	0	0	**F8**	0	0
I7	0.00251	0.59749	**I7**	0.00259	0.59741	**I7**	0.00251	0.59749	**I7**	0.00259	0.59741
F9	0	0	**F9**	0	0	**F9**	0	0	**F9**	0	0
I8	0.00021	0.04979	**I8**	0.00022	0.04978	**I8**	0.00021	0.04979	**I8**	0.00022	0.04978
I9	0.0036	0.8564	**I9**	0.00372	0.85628	**I9**	0.0036	0.8564	**I9**	0.00372	0.85628
F10	0	0	**F10**	0	0	**F10**	0	0	**F10**	0	0
F11	0	0	**F11**	0	0	**F11**	0	0	**F11**	0	0
I10	0.00168	0.39833	**I10**	0.00173	0.39827	**I10**	0.00168	0.39833	**I10**	0.00173	0.39827
F12	0	0	**F12**	0	0	**F12**	0	0	**F12**	0	0
F13	0.00335	0.79665	**F13**	0.00346	0.79654	**F13**	0.00335	0.79665	**F13**	0.00346	0.79654
F14	0	0	**F14**	0	0	**F14**	0	0	**F14**	0	0
I11	0.00054	0.12946	**I11**	0.00056	0.12944	**I11**	0.00054	0.12946	**I11**	0.00056	0.12944
F15	0	0	**F15**	0	0	**F15**	0	0	**F15**	0	0
F16	0.00011	0.02533	**F16**	0.00011	0.02533	**F16**	0.00011	0.02532	**F16**	0.00011	0.02532
F17	4.56E-05	0.01085	**F17**	4.71E-05	0.01084	**F17**	4.55E-05	0.01083	**F17**	4.70E-05	0.01083
F18	0.00022	0.05162	**F18**	0.00022	0.05161	**F18**	0.00022	0.05161	**F18**	0.00022	0.0516
F19	0.00019	0.04502	**F19**	0.0002	0.04502	**F19**	0.00019	0.04502	**F19**	0.0002	0.04502
F20	0.00223	0.53077	**F20**	0.0023	0.5307	**F20**	0.00223	0.53077	**F20**	0.0023	0.5307
F21	0.00418	0.99482	**F21**	0.00432	0.99468	**F21**	0.00418	0.99482	**F21**	0.00432	0.99468
F22	0.00419	0.99581	**F22**	0.00432	0.99568	**F22**	0.00419	0.99581	**F22**	0.00432	0.99568
F23	0	0	**F23**	0	0	**F23**	0	0	**F23**	0	0
F24	0	0	**F24**	0	0	**F24**	0	0	**F24**	0	0
F25	0	0	**F25**	0	0	**F25**	0	0	**F25**	0	0
F26	0	0	**F26**	0	0	**F26**	0	0	**F26**	0	0
F27	0.00418	0.99283	**F27**	0.00431	0.99269	**F27**	0.00418	0.99283	**F27**	0.00431	0.99269
F28	0	0	**F28**	0	0	**F28**	0	0	**F28**	0	0
F29	0.00037	0.08763	**F29**	0.00038	0.08762	**F29**	0.00037	0.08763	**F29**	0.00038	0.08762
I12	0.00075	0.17925	**I12**	0.00078	0.17922	**I12**	0.00075	0.17925	**I12**	0.00078	0.17922
F30	0	0	**F30**	0	0	**F30**	0	0	**F30**	0	0
F31	0	0	**F31**	0	0	**F31**	0	0	**F31**	0	0
I13	0	0	**I13**	0	0	**I13**	0	0	**I13**	0	0
F32	0	0	**F32**	0	0	**F32**	0	0	**F32**	0	0
I14	0.00103	0.24397	**I14**	0.00106	0.24394	**I14**	0.00103	0.24397	**I14**	0.00106	0.24394
F33	0	0	**F33**	0	0	**F33**	0	0	**F33**	0	0
F34	0.00412	0.98088	**F34**	0.00426	0.98074	**F34**	0.00412	0.98088	**F34**	0.00426	0.98074
F35	0	0	**F35**	0	0	**F35**	0	0	**F35**	0	0
F36	0	0	**F36**	0	0	**F36**	0	0	**F36**	0	0
B1	0	0	**B1**	0	0	**B1**	0	0	**B1**	0	0
R1	0.00173	0.41227	**R1**	0.00179	0.41221	**R1**	0.00173	0.41227	**R1**	0.00179	0.41221
F37	0	0	**F37**	0	0	**F37**	0	0	**F37**	0	0
I15	0.00035	0.08366	**I15**	0.00036	0.08365	**I15**	0.00035	0.08365	**I15**	0.00036	0.08364
F38	0	0	**F38**	0	0	**F38**	0	0	**F38**	0	0
F39	0	0	**F39**	0	0	**F39**	0	0	**F39**	0	0
F40	0.00024	0.05774	**F40**	0.00025	0.05773	**F40**	0.00024	0.05774	**F40**	0.00025	0.05773
F41	0.00012	0.02788	**F41**	0.00012	0.02788	**F41**	0.00012	0.02788	**F41**	0.00012	0.02788
F42	0.00137	0.32464	**F42**	0.00141	0.32459	**F42**	0.00137	0.32463	**F42**	0.00141	0.32459
F43	0.00084	0.19916	**F43**	0.00086	0.19914	**F43**	0.00084	0.19916	**F43**	0.00086	0.19914
F44	0.0014	0.3336	**F44**	0.00145	0.33355	**F44**	0.0014	0.3336	**F44**	0.00145	0.33355
F45	0.00419	0.99581	**F45**	0.00432	0.99568	**F45**	0.00419	0.99581	**F45**	0.00432	0.99568
F46	0	0	**F46**	0	0	**F46**	0	0	**F46**	0	0
F47	0.00418	0.99382	**F47**	0.00432	0.99368	**F47**	0.00418	0.99382	**F47**	0.00432	0.99368
F48	0	0	**F48**	0	0	**F48**	0	0	**F48**	0	0
F49	0.00033	0.07956	**F49**	0.00035	0.07955	**F49**	0.00033	0.07956	**F49**	0.00035	0.07955
I16	0	0	**I16**	0	0	**I16**	0	0	**I16**	0	0
I17	0	0	**I17**	0	0	**I17**	0	0	**I17**	0	0
F50	0	0	**F50**	0	0	**F50**	0	0	**F50**	0	0
I18	0	0	**I18**	0	0	**I18**	0	0	**I18**	0	0

Table 6-10 Basic input parameters for the final hotspot and coldspot models.

Site	**Compartment**	**Cell Density**	**Biomass (g m^{-2})**	**P/B (/year)**
Coldspot	*Gambierdiscus carolinianus*	100 g ww-1 algae	5.44	63.51
Coldspot	*Gambierdiscus excentricus*	100 g ww-1 algae	5.62	20.81
Hotspot	*Gambierdiscus carolinianus*	100 g ww-1 algae	5.44	63.51
Hotspot	*Gambierdiscus excentricus*	100 g ww-1 algae	5.62	20.81
Coldspot (Final Model)	*Gambierdiscus* spp. (multi-strain)	38208 cells m^{-2}	1.06	63.88
Hotspot (Final Model)	*Gambierdiscus* spp. (multi-strain)	412254 cells m^{-2}	14.0	62.05

Table 6-11 Diet composition data for final models for benthic autotrophs (BA) and *Gambierdiscus* species (Gamb. spp.) compartments.

	Coldspot			**Hotspot**				**Coldspot**			**Hotspot**	
Code	**BA**	**Gamb. spp.**	**Code**	**BA**	**Gamb. spp.**		**Code**	**BA**	**Gamb. spp.**	**Code**	**BA**	**Gamb. spp.**
F1	0.814169	0.000666	**F1**	0.806039	0.008756		**F25**	0.000000	0.000000	**F25**	0.000000	0.000000
F2	0.506048	0.000414	**F2**	0.501020	0.005442		**F26**	0.000000	0.000000	**F26**	0.000000	0.000000
I1	0.599509	0.000491	**I1**	0.593552	0.006448		**F27**	0.996185	0.000815	**F27**	0.986286	0.010714
F3	0.000000	0.000000	**F3**	0.000000	0.000000		**F28**	0.000000	0.000000	**F28**	0.000000	0.000000
I2	0.000000	0.000000	**I2**	0.000000	0.000000		**F29**	0.087928	0.000072	**F29**	0.087054	0.000946
I3	0.087928	0.000072	**I3**	0.087054	0.000946		**I12**	0.179853	0.000147	**I12**	0.178066	0.001934
I4	0.000000	0.000000	**I4**	0.000000	0.000000		**F30**	0.000000	0.000000	**F30**	0.000000	0.000000
F4	0.496593	0.000406	**F4**	0.491659	0.005341		**F31**	0.000000	0.000000	**F31**	0.000000	0.000000
F5	0.000000	0.000000	**F5**	0.000000	0.000000		**I13**	0.000000	0.000000	**I13**	0.000000	0.000000
F6	0.000000	0.000000	**F6**	0.000000	0.000000		**F32**	0.000000	0.000000	**F32**	0.000000	0.000000
F7	0.000000	0.000000	**F7**	0.000000	0.000000		**I14**	0.244800	0.000200	**I14**	0.242367	0.002633
I5	0.631483	0.000517	**I5**	0.625209	0.006792		**F33**	0.000000	0.000000	**F33**	0.000000	0.000000
I6	0.649468	0.000532	**I6**	0.643015	0.006985		**F34**	0.984195	0.000805	**F34**	0.974415	0.010585
F8	0.000000	0.000000	**F8**	0.000000	0.000000		**F35**	0.000000	0.000000	**F35**	0.000000	0.000000
I7	0.599509	0.000491	**I7**	0.593552	0.006448		**F36**	0.000000	0.000000	**F36**	0.000000	0.000000
F9	0.000000	0.000000	**F9**	0.000000	0.000000		**B1**	0.000000	0.000000	**B1**	0.000000	0.000000
I8	0.049959	0.000041	**I8**	0.049463	0.000537		**R1**	0.413661	0.000339	**R1**	0.409551	0.004449
I9	0.859297	0.000703	**I9**	0.850758	0.009242		**F37**	0.000000	0.000000	**F37**	0.000000	0.000000
F10	0.000000	0.000000	**F10**	0.000000	0.000000		**I15**	0.083940	0.000069	**I15**	0.083097	0.000903
F11	0.000000	0.000000	**F11**	0.000000	0.000000		**F38**	0.000000	0.000000	**F38**	0.000000	0.000000
I10	0.399673	0.000327	**I10**	0.395702	0.004298		**F39**	0.000000	0.000000	**F39**	0.000000	0.000000
F12	0.000000	0.000000	**F12**	0.000000	0.000000		**F40**	0.057938	0.000047	**F40**	0.057361	0.000623
F13	0.799346	0.000654	**F13**	0.791403	0.008597		**F41**	0.027979	0.000023	**F41**	0.027699	0.000301
F14	0.000000	0.000000	**F14**	0.000000	0.000000		**F42**	0.325736	0.000267	**F42**	0.322497	0.003503
I11	0.129894	0.000106	**I11**	0.128603	0.001397		**F43**	0.199836	0.000164	**F43**	0.197851	0.002149
F15	0.000000	0.000000	**F15**	0.000000	0.000000		**F44**	0.334726	0.000274	**F44**	0.331400	0.003600
F16	0.025416	0.000021	**F16**	0.025158	0.000273		**F45**	0.999182	0.000818	**F45**	0.989254	0.010746
F17	0.010882	0.000009	**F17**	0.010756	0.000117		**F46**	0.000000	0.000000	**F46**	0.000000	0.000000
F18	0.051796	0.000042	**F18**	0.051270	0.000557		**F47**	0.997184	0.000816	**F47**	0.987275	0.010725
F19	0.045175	0.000037	**F19**	0.044726	0.000486		**F48**	0.000000	0.000000	**F48**	0.000000	0.000000
F20	0.532564	0.000436	**F20**	0.527272	0.005728		**F49**	0.079829	0.000065	**F49**	0.079035	0.000859
F21	0.998183	0.000817	**F21**	0.988265	0.010735		**I16**	0.000000	0.000000	**I16**	0.000000	0.000000
F22	0.999182	0.000818	**F22**	0.989254	0.010746		**I17**	0.000000	0.000000	**I17**	0.000000	0.000000
F23	0.000000	0.000000	**F23**	0.000000	0.000000		**F50**	0.000000	0.000000	**F50**	0.000000	0.000000
F24	0.000000	0.000000	**F24**	0.000000	0.000000		**I18**	0.000000	0.000000	**I18**	0.000000	0.000000

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Chapter 7. FINAL ECOPATH WITH ECOSIM MODELS AND SIMULATIONS

Abstract

Human consumption of fishes and other marine organisms that contain high levels of ciguatoxin (CTX) can lead to ciguatoxin fish poisoning (CFP). CFP causes a wide variety of symptoms, including vomiting, diarrhea, a reversal of hot and cold sensations, and occasionally death. Single-celled dinoflagellates in the *Gambierdiscus* and *Fukuyoa* genera synthesize CTX. The toxin enters the food web when herbivores inadvertently consume these dinoflagellates when feeding on their preferred substrates. The toxin is metabolized and biomagnifies up the food web. Ciguatoxin is odorless and colorless and is heat-stable. There are no reliable dockside rapid tests to determine if a species has high levels of CTX. The best way to prevent CFP outbreaks is to avoid eating contaminated fishes. Here we present two food web models that estimate CTX levels in species in a CTX "hotspot" (high levels of CTX) and CTX coldspot (low levels of CTX). Fishers identified these areas from open-ended interviews. The highest trophic level species, *Sphyraena barracuda*, does not reach the FDA recommended consumption advisory levels of 0.1 ppb in the coldspot after 25 months. However, the three highest trophic level compartments reach the 0.1 ppb mark in less than nine months in the hotspot. The rapid CTX concentration gain in the hotspot could be due to the 35-fold increase in toxin-producing dinoflagellate cell density. We suggest that Puerto Rico fisheries managers develop a routine dinoflagellate sampling protocol. The results can be entered into our models depending on where the cells were sampled (hotspot or coldspot) to estimate when fishes will become toxic for management decisions.

Introduction

People living in tropical and subtropical regions worldwide rely on fish and other marine organisms for sustenance, tourism, and recreation. However, fishes in these regions, specifically in the Pacific and Indian Oceans and the Caribbean Sea, can harbor ciguatera toxin (ciguatoxin or CTX), a potent neurotoxin produced by several different species of dinoflagellates, most notably in the *Gambierdiscus* and *Fukuyoa* genera (Lewis *et al.*, 1991; Pottier *et al.*, 2002). Suppose humans ingest tissues of species that accumulate this toxin in a high enough concentration. In that case, it can cause a variety of severe symptoms, i.e., vomiting, diarrhea, abdominal pain, paresthesia (burning of the skin), the reversal of hot and cold sensations, and occasionally, death (Lehane and Lewis, 2000). Not only does the muscle tissue have the potential to be toxic, the roe, liver, and other organs in the fishes carry higher levels of CTX than muscle tissues and may be more dangerous to consume (de Fouw *et al.*, 2001). Different classes and congeners of ciguatoxins in the Indian Ocean, Pacific Ocean, and the Caribbean Sea cause variations in symptoms from those regions (Murata *et al.*, 1990; Lewis, 1998, 2000). The sickness from consuming ciguatoxic fish is known as ciguatoxin fish poisoning (CFP).

CTX-producing dinoflagellates are endemic mostly to Caribbean and Pacific island reefs (i.e., the Caribbean; the U.S. Virgin Islands, Antigua and Barbuda, the Cayman Islands, Puerto Rico, Jamaica, etc. and in the Pacific; American Samoa, the Cook Islands, Fiji, Hawaii, French Polynesia, etc.) (Lewis, 2001; Litaker *et al.*, 2010; Friedman *et al.*, 2017). Pacific ciguatoxins (P-CTX) are 10-fold more toxic than Caribbean ciguatoxins (C-CTX). The FDA recommended advisory consumption levels for the primary Caribbean CTX chemical strain less than 0.1 ppb C-

CTX-1 equiv. toxicity and 0.01 ppb P-CTX-1 equiv. toxicity for Pacific ciguatoxins (Vernoux and Lewis, 1997; Lehane and Lewis, 2000; Pearn, 2001; Dickey and Plakas, 2010).

Despite the impact CTX has on fisheries and consumers, it is challenging to pinpoint global CFP incidence rates with a high confidence level due to poor CTX detection techniques (Friedman *et al.*, 2017). CTX is colorless, odorless, and tasteless (Copeland *et al.*, 2014) and is heat-stable, meaning cooking the fish does not affect the toxin (Lewis, 2000). Local folk methods for identifying toxic fish (such as feeding a small piece of fish to a pet animal and monitoring its reaction, rubbing the flesh with a coin, or leaving a portion of the fish near insects to see if they avoid it) are unreliable (Darius *et al.*, 2013). CTX dockside test strips (Cigua-Check®) were available from Oceanit® but discontinued due to inaccuracy tests. They were confusing to administer and inaccurate, with a high chance of false-negatives (Bienfang *et al.*, 2011).

This study aimed to parameterize two Ecopath models, one for a CTX hotspot and one for a CTX coldspot, and to estimate the time it takes CTX to biomagnify in the food web to the top trophic levels and highly targeted species.

Methods

The biomagnification of CTX in coral reef food web networks was assessed by sampling fishes, filling data gaps with previously published models, sampling CTX-producing dinoflagellates, and using the Ecotracer module within Ecopath with Ecosim for two reefs off the coast of Puerto Rico. Fishers identified one reef as having high levels of CTX (CTX hotspot), and the other was a common fishing area for commercial fishers with low levels of CTX (CTX coldspot). Both sites are identified in Chapters 3-5. The CTX-1 and CTX-2 coldspot sites were combined into one coldspot model, while the CTX-3 and CTX-4 hotspot sites were combined into one hotspot model.

Once the sites were identified, divers identified the fishes present at each reef. Over two consecutive sampling days, divers swam in an “L” shape with an area of 3600 m^2 and identified fish species and the quantity of those species. Each species’ number of individuals was used to find the biomass (g m^{-2}) at both the hotspot and coldspot. Production to biomass (P/B) and consumption to biomass (Q/B) ratios for species identified in the hotspot and coldspot were in the Opitz (1996) model. Q/B ratios for all species were available. If P/B ratios were missing for a species, then the P/B ratio from its respective Opitz (1996) compartment was used instead.

Next, a predator/prey consumption matrix was created using prey data from Randall (1967). All prey items in the predators’ diet were assigned a proportion that added up to 1.0 (100% of the diet). A hierarchical cluster analysis was performed on the diet data to condense the species list into fewer compartments (Figure 6-1). A weighted mean was used to condense predators into each compartment; the proportion of the biomass of an individual species was used to calculate its relative impact on that group's diet. For example, if species A and species B were grouped from the cluster analysis, and if species A had a biomass of 20 g m^{-2} and species B had a biomass of 10 g m^{-2}, and they both consumed species C with a proportion of their diets as 0.50 and 0.35 respectively, the proportion of the diet for the prey species C in the combined compartment would be 0.33 from species A and 0.116 from species B. After all predator groups were combined using the weighted mean, the prey groups were incorporated into the same compartments by summing the proportions. The *Gambierdiscus* spp. prey group was calculated as a proportion of the benthic autotroph group like in Chapter 2. The biomass (g m^{-2}) of the *Gambierdiscus* spp. compartment was divided by the biomass of the benthic autotroph group.

That value was multiplied by the benthic autotroph prey compartments for the prey data for *Gambierdiscus* spp. The new benthic autotroph prey data was the difference between the original prey data minus the proportion of the *Gambierdiscus* spp. removed from the original benthic autotroph compartment.

Biomass (g m^{-2}) of species were added together to get a new value for each compartment from hierarchical cluster analysis. The P/B and Q/B ratios' median values were found for the new compartments using a weighted value with the biomass proportion of each species. The basic input parameters and the diet composition matrix for both hotspot and coldspot were completed. The complete methods are available in Chapter 7.

Before divers started performing fish counts, they placed screen sampling rigs (Figure 5-1) to collect *Gambierdiscus* spp (Figure 5-2). The repeat rigs were set 10m apart along a line at all four sites (CTX-1, CTX-2, CTX-3, CTX-4). The samples were allowed to soak for 24 hours and were collected the next day. The divers put a jar over the screens and unhooked the fishing swivels to collect the screen. Then, the samples were preserved and stored using the methods described in Chapter 5. The dinoflagellate cells were counted in each sample and were also run with a qPCR for species identification. Repeat samples from CTX-1 and CTX-2 were combined into one "coldspot," while repeat samples from CTX-3 and CTX-4 were combined into one "hotspot." Both were named *Gambierdiscus* spp. The number of cells L^{-1} was then converted to g m^{-2} using the area of the screen rigs, 10.5 cm x 15.5 cm, and the mass of each identified dinoflagellate species (Holland C., personal communication, July 2020, chris.holland@noaa.gov). The cell number (on the 150 cm^2 screen rig), which was also the number of cells L^{-1}, was multiplied by 66 to represent cells m^{-2}. The total cell number m^{-2} in the hotspot and coldspot was divided evenly among the species identified from the qPCR at each site. Each subset of the total cells was multiplied by the species' mass for a total mass (g m^{-2}). These were added together to get a biomass input parameter for the Ecopath model for the new *Gambierdiscus* spp. compartment. The P/B values were calculated using the growth rate (d^{-1}) for each species and finding the new compartment's median value. The biomass of each species (g) was multiplied by the growth rate (d^{-1}) and the total cell number to get additional mass per day for production. The production was multiplied by 365 to get biomass/year then divided by the original biomass of the total number of cells for production/biomass/year. The calculated P/B values were close to the phytoplankton P/B value from the original Opitz (1996) model. The growth rates (d^{-1}) were found in (Litaker *et al.*, 2017).

Final input parameters were entered into the Ecopath with Ecosim model for both the hotspot and coldspot, and basic estimates were calculated. Species absent in the coldspot but present at the hotspot (in their compartment in the model) had their biomass (g m^{-2}) reduced to 0.001. This represented the species missing from the food web but wasn't completely removed to ensure the models' compartments matched. The diet matrices of the coldspot model were adjusted to reflect this. Any predator feeding on one of the compartments with the reduced biomass had that prey reduced in their diet to 1.0×10^{-10}. The proportion of the prey that was reduced was split evenly among all other prey groups. A *Gambierdiscus* spp. compartment was added to each model to represent the multiple species and cell counts at each site previously identified. The biomass (g m^{-2}) and P/B were added to the basic input parameters for the new group. The models were balanced to keep all ecotrophic efficiencies (EE) under 1.0. An EE > 1.0 means that a compartment is being over consumed, corrected by increasing each compartment's biomass until EE < 1.0 to get a balanced model. The final basic input parameters are shown in Table 6-5.

Then, input parameters for the Ecotracer module were calculated. The initial concentration of the *Gambierdiscus* spp. compartments were calculated by dividing the toxicity of each species in fg CTX3C equiv. cell^{-1} by the mass of the species in g cell^{-1} to get fg g^{-1}. The toxicity in fg g^{-1} was converted (multiplied by10^{-15}) to g g^{-1}. The overall initial concentration for the *Gambierdiscus* spp. compartment was found by taking an average of the toxicity (g g^{-1}) for the species in the compartment. Ecotracer measures the bioaccumulation and biomagnification of substances obtained from the abiotic environment. However, CTX is biosynthesized in algae. Therefore, the *Gambierdiscus* spp. compartment was manipulated in the Ecotracer module to account for the growth of the algae, and subsequently, more toxin production. The *direct absorption rate* column in Ecotracer simulates contaminates taken up into compartments from the environment as a proportion of the initial environmental concentration. I set the initial concentration of CTX in the environment to 1 g m^{-2} (equal to 1 t km^{-2} in Ecotracer), which is a high value, however, it is only incorporated into the tissues of *Gambierdiscus* spp. as a function of toxin production by the algae. The direct uptake rate is the same as the algal production rate of CTX. For both the hotspot and the coldspot, direct absorption rate of their respective *Gambierdiscus* spp. compartments were calculated by multiplying the number of cells at day *n* by the growth rate (d^{-1}) to obtain the number of cells at day *n* + 1. Day *n* + 1 cell number was then multiplied by the growth rate (d-1) and added to the number of cells at day *n+1*. This was repeated until day *n* + 29 to simulate growth for 1 month. The final value was the total number of cells at day *n* + 29, multiplied by the toxicity in fg g^{-1} CTX3C equiv. to get production of CTX g m^{-2} for one month. The number of cells at day *n* was multiplied by the toxicity in fg g^{-1} CTX3C equiv. and was subtracted from the toxin produced at day *n* + 29. This was done to assume the algae is steady-state, and predators consumed any additional toxin production. One month of growth was used because Ecosim uses a one-month time step (Walters and Christensen, personal communication, September 2020, wjw24@psu.edu, v.christensen@oceans.ubc.ca). Data from Bomber *et al*. (1988) were then used to apply a forcing function to the *Gambierdiscus* spp. compartments in both the hotspot and the coldspot. A forcing function is a way to force primary production based on a monthly growth pattern directly. The forcing function pattern values can be seen in Appendix J, and below is the associated graph (Figure 7-1). Lastly, for the Ecotracer module, the proportion of contaminant excreted was added for all species under ETL 3.0 that consumed benthic autotrophs to simulate the 95% excretion by herbivorous fishes shown by Ledreux *et al*. (2014). This scenario with 30 days of CTX production is called the Ecosim 30-day Growth Scenario.

After the forcing function was applied, and all required input parameters were entered, models were run with the Ecotracer module. Species from 4 different trophic levels were chosen to show in the models. *Sphyraena barracuda* was selected because it is a high trophic level and is a species under strict non-commercial harvest rules in Puerto Rico due to CTX. *Lachnolaimus maximus* was selected because of its commercial importance and fishers in interviews believed they were either very toxic or completely safe to eat based on the location they were caught.

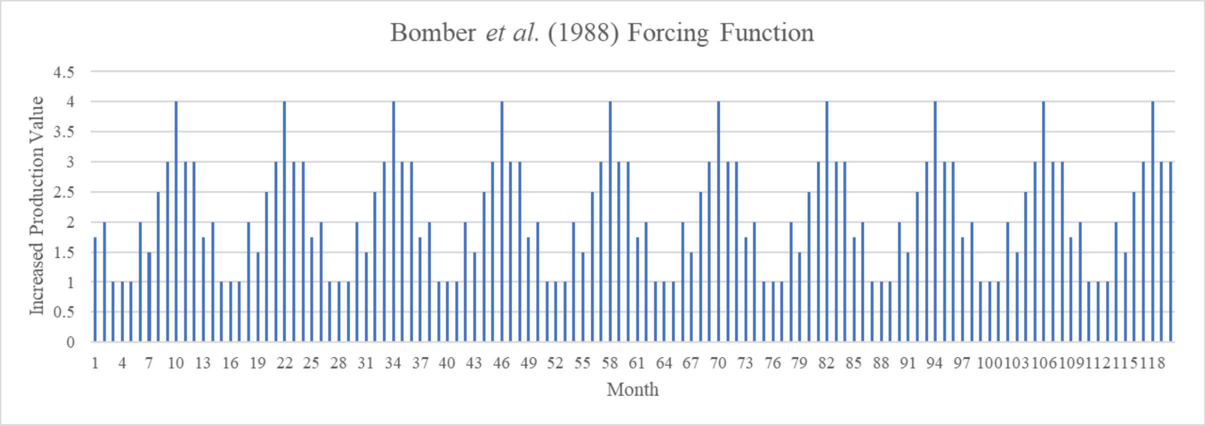

Figure 7-1 Forcing function used in both the final hotspot and coldspot models to force primary growth of the *Gambierdiscus* spp. compartments. The x-axis is years and the vertical bars are monthly values. The y-axis is the value multiplied by the maximum production/biomass ratio that can be realized of that compartment (Ecopath Developer Site)

Gastropods were chosen because they are a staple of the *Lachnolaimus maximus'* diet. They also feed on benthic autotrophs and inadvertently consume toxic dinoflagellates while doing this. Lastly, the large scaridae compartment was chosen because of their lower trophic level (2.0) and consume mostly benthic autotrophs, and therefore toxic dinoflagellates.

Model Validation

To be useful for their intended purpose, the Ecopath models and Ecotracer projections need to be validated with independent data. I used the measured CTX3C equiv. tissue concentrations from the N2A-cba assays performed in Chapter 4 for validation of the Ecotracer model runs (called validation scenarios). For these Ecotracer validation scenarios, all of the model parameters were kept the same except for the amount of growth added to the direct uptake rate compartment in Ecotracer. The direct uptake rate was changed because this is the most uncertain parameter in Ecotracer and it had a large impact on the predicted CTX concentrations from the model scenarios after the initial runs with the 30-day bloom direct uptake rate. The maximum CTX values of species in each scenario of the hotspot model was then compared to the CTX3C equiv. values that I measured in tissues of *Sphyraena barracuda, Lachnolaimus maximus,* and *Sparisoma viride* caught at the hotspot locations. If the measured CTX in tissues and the intial Ecopath models with the Ecotracer scenario projections for CTX for each of these species agreed, the scenarios was considered validated. If they were different, and new Ecotracer scenario was parameterized as described below and re-tested against the CTX tissue measurements.

For the first Ecotracer validation scenario, I ran the hotspot model with one day of growth in the direct absorption rate compartment calculated from the *Gambierdiscus* species and quantity of cells present. This hotspot model with one day of growth and no forcing function is referred to as Ecosim Scenario Validation 1. Then, different forcing functions were added to the one-day bloom model. A random-bloom forcing function was created by generating ten random numbers (7,17, 28, 37, 39, 43, 68, 73, 87, 98), each of which corresponded to a month out of 120 months (ten blooms occurring at random months over ten years). Blooms occurring in those months were given a relative intensity value in the forcing function of 3, which is 3x the production of the *Gambierdiscus* spp. compartment. This random-month-for-blooms model is referred to as Ecosim Scenario Validation 2. Finally, a forcing function of 12 straight months of growth was used as a worst-case scenario to see how quickly it takes the compartments to reach 0.1 ppb. This is Ecosim Scenario Validation 3. All forcing functions are shown in Appendix I.

Results

The initial Ecosim 30-day Growth Scenario shows the compartments in the hotspot model reach 0.1 ppb faster than the compartments in the coldspot model (Figure 7-2). The Ecosim scenario was based on the final Ecopath hotspot and coldspot models with the species

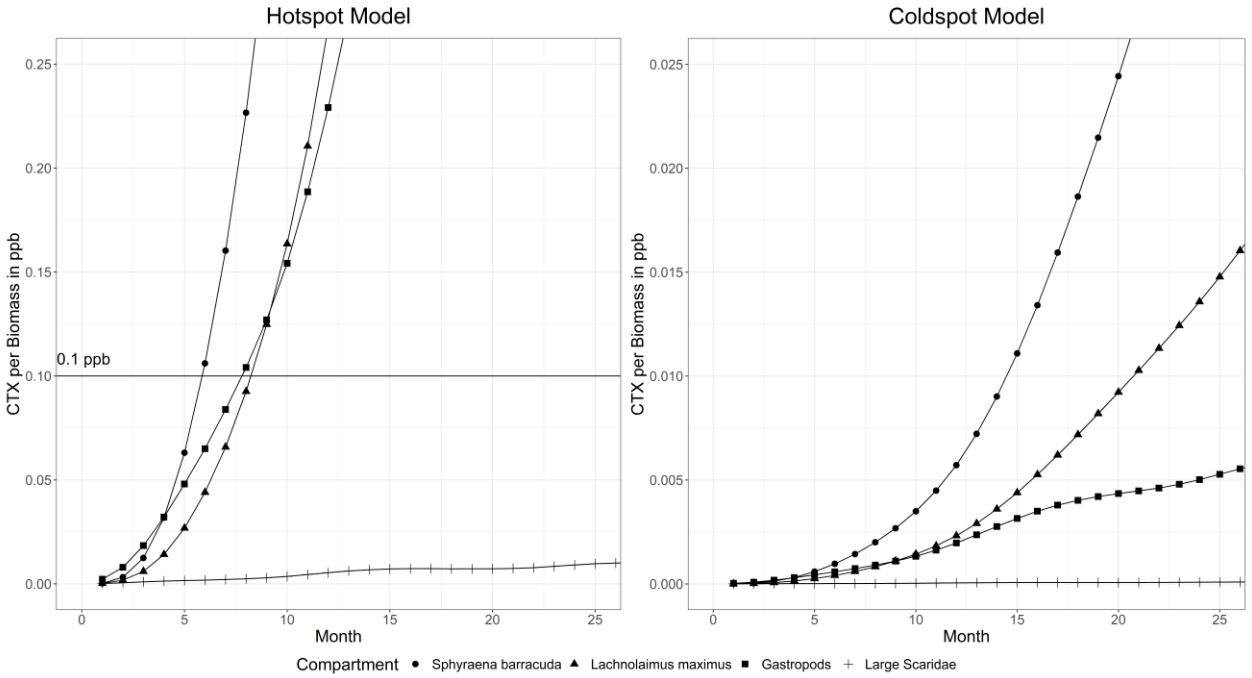

Figure 7-2 CTX biomagnification of 4 species in the hotspot and coldspot models with 30 days of CTX production in the direct uptake compartment and the forcing function from Bomber *et al.* (1988).

and quantity of cells sampled from those sites used to determine the biomass of the *Gambeirdiscus* compartment, 30 days of CTX production in the direct uptake compartment in Ecotracer, and a forcing function from Bomber *et al.* (1988). In the hotspot, *Sphyraena barracuda* passes 0.1 ppb in less than six months, *Lachnolaimus maximus* passes 0.1 ppb in less than nine months, and gastropods cross the 0.1 ppb threshold in less than eight months. None of the compartments in the coldspot model pass 0.1 ppb after 25 months. In the hotspot, after ten years the *Sphyraena barracuda* achieves CTX values of over 102 ppb as well as 15 ppb in *Lachnolaimus maximus* (not shown). These high values have not been seen in tested fishes, either in this study or elsewhere, which suggests the model overestimates the CTX values in these fish. This initial Ecosim 30-day Growth Scenario overestimates CTX biomagnification in the food web.

Since the initial Ecosim 30-day Growth Scenario overestimated the CTX in *S. barracuda* and *L. maximus,* I simulated Ecosim Scenario Validation 1. Figure 7-3 shows these results.

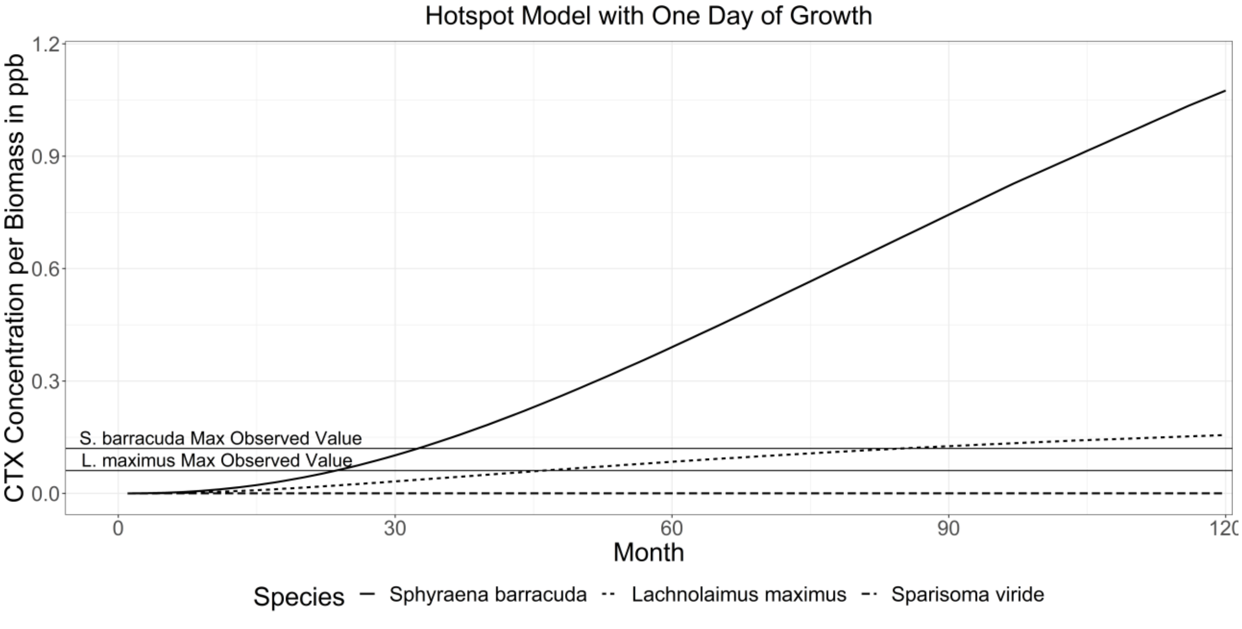

Figure 7-3 Ecosim Scenario Validation 1. The hotspot model with one day of growth input in the direct uptake parameter in the Ecotracer module. This scenario had no forcing function.

The *S. barracuda* and the *L. maximus* cross the 0.1 ppb FDA recommended advisory consumption level in 30 months and 71 months, respectively. The horizontal lines display the maximum estimated values for those fishes from the N2a assay. The *S. barracuda* and *L. maximus* both exceed the maximum estimated values, but the overestimate of CTX for *L. maximus* is not as large as the overestimate for *S. barracuda.* The maximum estimated value for *S. viride* in the hotspot using the N2a-cba was 0.0006 compared to the 0.00014 ppb seen after 120 months in the model. This model scenario slightly underestimates the CTX in *S. viride,* but it never exceeded the FDA level in either the model runs or the measured tissue samples.

After I simulated the hotspot model with one day of growth, I added a random bloom forcing function, this is Ecosim Scenario Validation 2 (Figure 7-4). Ten random months (out of 120) were picked to have a bloom forcing function value of three times the baseline level where all the other months had a baseline of one. The results show an overall higher CTX value after ten years. The *S. barracuda* and *L. maximus* cross the 0.1 ppb threshold after 27 months

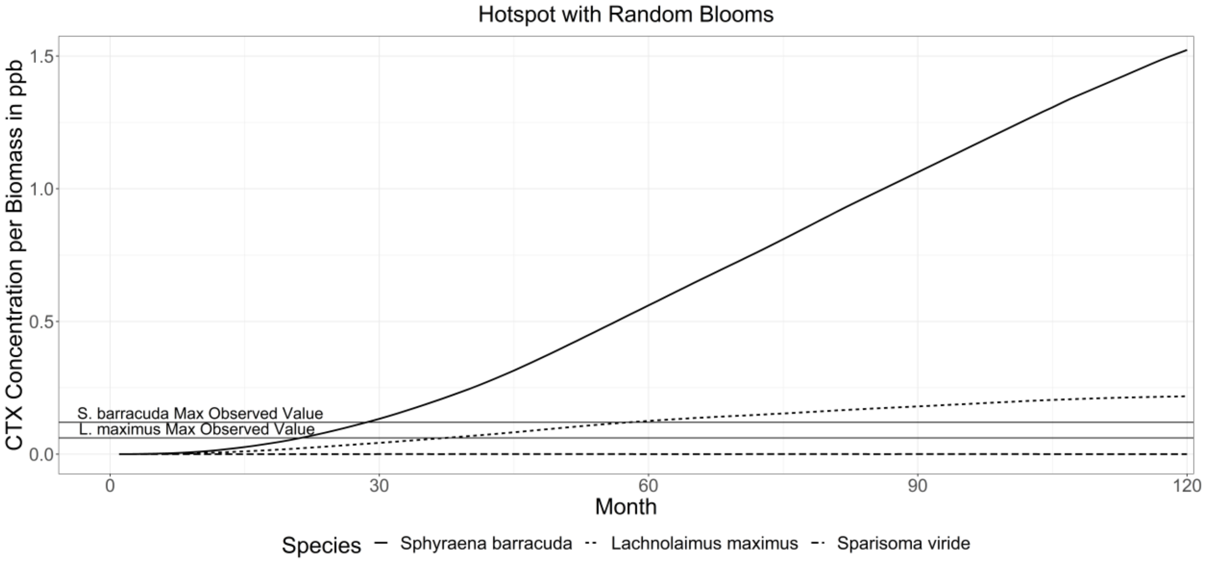

Figure 7-4 Ecosim Scenario Validation 2. Hotspot with one day of growth in the direct uptake rate compartment and a series of random blooms as a forcing function. Ten random months from 1-120 (ten years) were picked to have a 3x production function as a forcing function.

and 51 months, respectively, while the *S. viride* never does (not shown). Again, maximum CTX levels after 10 years in both *S. barracuda* and *L. maximus* are overestimated by the model when compared to the CTX3C equiv. levels measured from Chapter 4.

The last simulation, Ecosim Scenario Validation 3, represented a worst-case scenario with a bloom every month for the first 12 months of the simulation. The maximum CTX concentration in *S. barracuda* and *L. maximus* after 120 months were 1.64 ppb and 0.2 ppb, respectively (Figure 7-5). The *S. barracuda* reached 0.1 ppb in 16 months while the *L. maximus* reached 0.1 ppb in 29 months . These results show how quickly these species cross the 0.1 ppb threshold and how toxic they become over 10 years in the absence of any other dinoflagellate blooms.

The 120-month Ecopath/Ecotracer model run predictions for CTX levels in three fishes in each of the three scenarios used for validation were compared with the fish tissues measured at the hotspot and coldspot for CTX3C equiv. using the N2a bioassay (Table 7-1). The measured values of the *S. barracuda* and *L. maximus* tissues were lower than the predicted CTX values for all the validation scenarios after 120 months using the Ecopath hotspot model. The CTX3C equiv. measurements for *S. viride* tissue samples were higher than those in the Ecotracer scenario predictions for both Ecopath models. In the Ecopath coldspot model, both the *S. viride* and the *S. barracuda* measured CTX3C equiv. values are lower than the Ecotracer predicted values after 120 months. The *L. maximus* measured CTX3C equiv. value is almost twice as high as the predicted values at the Ecopath coldspot model. Although some measured CTX3C equiv. values are lower or higher than predicted values, they are closer than what was observed from the Ecosim 30-day Growth Scenario. Ecosim Validation Scenario 1 predicted CTX values closest to the sampled CTX3C values after 10 years and therefore, should be considered the best scenario for model validation.

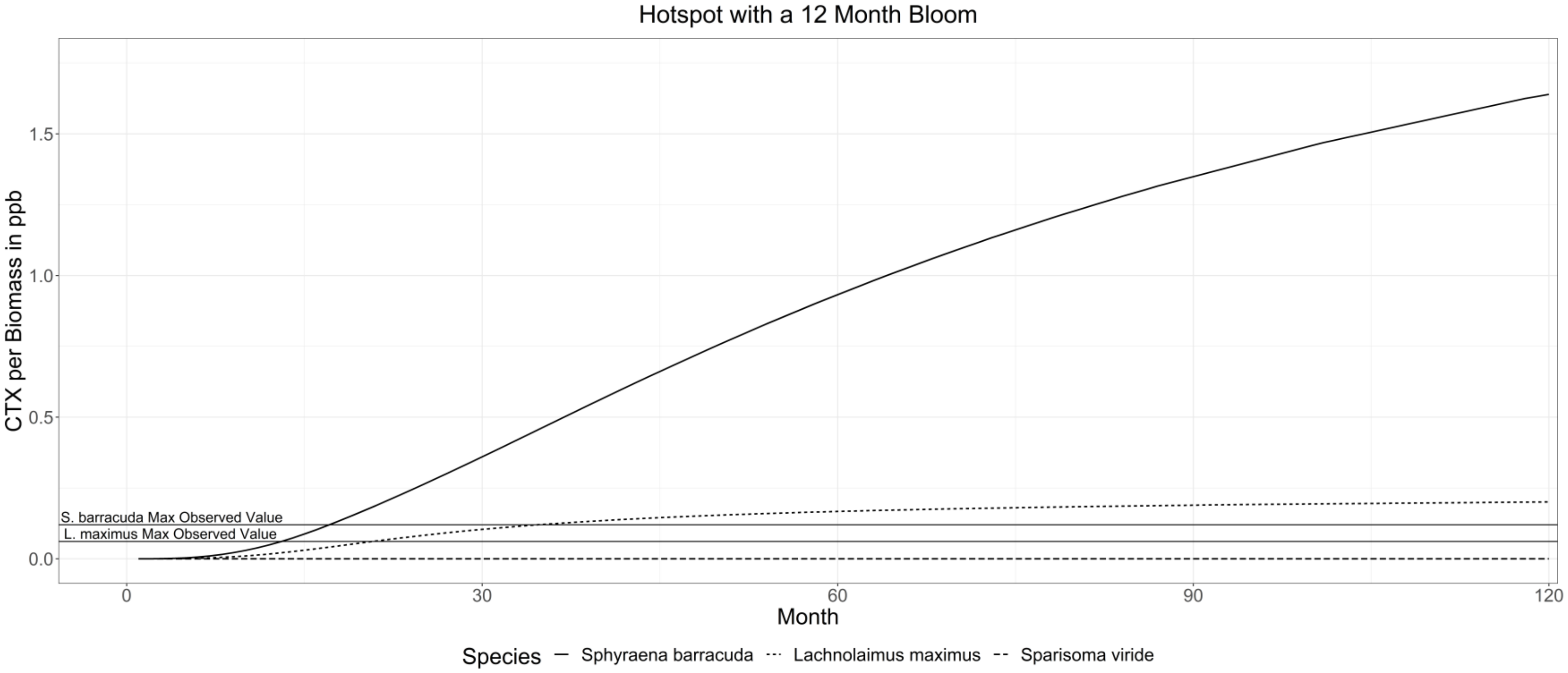

Figure 7-5 Ecosim Scenario Validation 3. Hotspot with one day of growth in the direct uptake rate compartment and 12 straight months of a dinoflagellate bloom as a forcing function. The first 12 months had a 3x production growth function as the forcing function in the *Gambierdiscus* spp. compartment.

Table 7-1 Maximum observed N2a-cba sampled values from Chapter 4 compared to predicted values from Ecosim Scenario Validation 1, 2, and 3

				120 Month CTX Values for Ecosim Scenario Validation 1, 2, and 3		
Site	**Species**	**Number of Fishes Sampled**	**Sampled CTX3C Values (N2a-cba)**	**1**	**2**	**3**
Hotspot	*Sphyraena barracuda*	5	0.1151 ppb	1.076 ppb	1.516 ppb	1.6043 ppb
Hotspot	*Lachnolaimus maximus*	4	0.0614 ppb	0.1559 ppb	0.2174 ppb	0.2012 ppb
Hotspot	*Sparisoma viride*	3	0.00031 ppb	0.00014 ppb	0.00015 ppb	0.00014 ppb
Coldspot	*Sphyraena barracuda*	3	0.0008 ppb	0.0093 ppb	0.0126 ppb	0.0138 ppb
Coldspot	*Lachnolaimus maximus*	8	0.0031 ppb	0.0014 ppb	0.0018 ppb	0.00178 ppb
Coldspot	*Sparisoma viride*	3	0.0062 ppb	0.00000122 ppb	0.00000136 ppb	0.00000124 ppb

Discussion

The main finding from the Ecopath and Ecotracer models developed in this project (Luczkovich & Raab, 2021) is that the hotspot model predicted high-trophic level fishes to become toxic more quickly than in the coldspot model. The 30-day Bloom Scenario with the Bomber *et al*. (1988) forcing function in Ecotracer was used initially to compare CTX concentrations predicted after 10 years in the hotspot Ecopath model and coldspot Ecopath model using the dinoflagellate species identified at each site (via qPCR) and cell densities measured at each area (using screen rig samplers). The equivalent hotspot compartments were more toxic than the coldspot compartments (specifically discussed in this chapter *S. barracuda*, *L. maximus* and gastropods), except for the parrotfish (Scaridae), which do not show any significant CTX accumulation in either model. The species composition and quantity of cells identified in the hotspot and coldspot areas apparently drove the compartments' toxicity differences in these two models.

The stoplight parrotfish (*S. viride*, Scaridae) in either model never accumulated significant concentrations of the toxin. I believe the excretion data from Ledreux *et al*. (2014), which was included in all the models I tested, produced this result. Ledreux *et al.* (2014) found that oxocene CTX congeners were not retained in the tissues of herbivorous fishes fed *Gambierdiscus polynesiensis* experimentally. The striped mullet (*Mugil cephalus)* in their study eliminated 95% of ciguatoxins in 24 hours, and repeated feedings did not promote toxin accumulation. At first, this seems counterintuitive to Randall's 1958 food chain hypothesis, where the concentration of the toxin at the herbivorous fish level is necessary for its bioaccumulation in the food web. However, this may not be the full story, ecologically. Striped mullet in the Ledreux *et al*. (2014) study had a myriad of hypo- and hyperactive behaviors.

These erratic behaviors may increase the striped mullet's or another herbivorous fish's chances of being consumed by predators in the wild. Predators could be feeding on prey that had just consumed a large dose of CTX via toxic dinoflagellates, causing neurotoxic effects on behavior, before the toxin had time to work its way through the bloodstream, with 5% retained in tissues as oxopenes congeners of CTX and the remaining oxocene CTX excreted. Predators consuming herbivorous fish in the first 24 hours after feeding on *Gambeirdiscus* would be receiving a large amount of the toxin from the recent herbivorous fish feeding and any toxin stored in the tissues or organs. One consideration for future modeling efforts is to determine if the oxcene excretion and selective retention of the oxopene congeners of CTX occurs at higher trophic levels. The rates of excretion of oxocenes should be included at each trophic transfer but are unknown so were not include in the Ecopath models reported here.

The path of the toxin through the food web can influence toxin concentration of a given species. The presence or absence of given prey species may influence the biomagnification in other species. For example, I believe one of the more common ways CTX enters the food web is through the consumption of benthic autotrophs by gastropods and their consumption by hogfish (*Lachnolaimus maximus*). If one site has fewer gastropods than another, then less CTX may get through the food web through that route, because of the reduced grazing by gastropods, thus causing hogfish to be less toxic. Hogfish at sites with few gastropods may switch to alternative prey that do not feed on *Gambierdiscus* or bioaccumulate ciguatoxins. For this reason, the diets of fishes in both the hotspot and coldspot areas should be examined directly in future studies; I was unable to gather such data for this project. In this study, I used Randall's (1958) data for all of the fishes' diets and Opitz's (1996) data for the benthic invertebrate diets, which may not be representative of the diets occurring at the hotspot and coldspots we modelled. Doing a more in-depth diet analysis of each site might show discrepancies from these dietary data, which could influence the movement of CTX through the food webs.

The initial model with the 30-day growth function as the direct uptake rate in Ecotracer and the Bomber *et al*. (1988) forcing function drastically overestimated CTX levels when projected over 10 years. The 30-day growth function used as amount of direct uptake pushes the *S. barracuda* over 102 ppb and the *L. maximus* to 15 ppb after 120 months. Such high levels of CTX have never been measured previously in these fish species. For example, in studies by O'Toole *et al.* (2012) in the Bahamas and Dechraoui *et al*. (2005) in the Florida Keys, maximum levels for great barracuda *S. barracuda* were 0.167 ppb and 2.1 ppb respectively. For hogfish *L. maximus* the maximum reported concentration was 0.044 ppb in the Florida Keys (Catasus 2019). In my study, maximum for *S. barracuda* was 0.1 ppb and the maximum for hogfish *L. maximus* was 0.06 ppb (Chapter 4). Therefore, this 30-day growth model was not considered validated; it needed to be adjusted before it could be useful to scientists and fisheries managers.

Once I reduced the direct uptake rate to account for one day of CTX production rather than 30-days, the 10-year CTX levels were aligned more closely to what is seen in the literature and what I estimated in the lab using the bioassay (Table 7-1). The values were biologically possible and in the general range of what we sampled in Chapter 4 unlike values seen from the preliminary models (Chapter 2) and the initial 30-day bloom models in this Chapter. However, in the *S. barracuda* and *L. maximus* CTX levels are still overestimated after 10 years in the predictions from simulations with 1-day growth parameters. Predictions after ten years was used as a general metric to compare with measured CTX3C levels using the N2a bioassay, but the fishes sampled at the hotspot and cold spot sites may not have had 10 years of exposure to CTX and *Gambierdiscus* at the time of sampling. It is probable that the fishes we sampled were less than 10 years old and there is no time series of the blooms occurring at the sites over the past ten years. The CTX3C equiv. levels observed in the N2a-cba assay more closely match the levels seen in the validation scenarios. Ecotracer Validation Scenario 1 (1 day of *Gambierdiscus* growth used as the direct uptake rate with no forcing function) was closest to the measured values. In the future, the validated models may need to be changed and revalidated using a similar approach. One high priority research need is that the time series of blooms be determined at these sites for developing an accurate forcing function parameter to improve the accuracy of the Ecotracer CTX projections. It would be ideal for future studies to sample more fishes and look at otolith samples to estimate fishes' age. I believe that with accurate monthly dinoflagellate samples to better parameterize the forcing function, and fish age estimates, the Ecotracer Validation Scenario 1 using the Ecopath hotspot model can be used to accurately predict CTX levels in fish tissues of great barracuda, hogfish, parrotfish and other species in this model in the months following a bloom.

Unfortunately, we were unable to sample monthly dinoflagellate counts to better parameterize the forcing function for the model. The random bloom (Validation Scenario 2) and the 12-month bloom scenario (Validation Scenario 3) are just educated guesses about some of the growth patterns that might happen with these algae. The Bomber *et al*., (1988) forcing function model is the closest one to actual observed values. However, the species of *Gambierdiscus* are not the same as at our sites and this study was reported over two decades ago. I am confident that the species' CTX levels after 10 years in the Ecopath hotspot model with the one-day growth period as the direct uptake rate (Ecotracer Validation Scenario 1) is the most similar to what I measured for fish tissues in the N2a-cba results and that this Ecopath model can be used to simulate CTX levels over time. It is critical that monthly sampling occurs at hotspots for dinoflagellate counts and N2a-cba tests on key species be done around Puerto Rico to better understand the bloom dynamics of these algae, to make accurate predictions for management purposes, and to issue consumption advisories to the public at the appropriate time after a bloom. Monthly cell counts should be obtained at the hotspot and coldspot areas studied here and thus can be used to better parameterize a forcing function that would be more representative to the locations sampled. This *Gambierdiscus* spp. cell count time series obtained from screen rigs deployed monthly, along with the Ecopath/Ecotracer models presented here using a one-day growth direct uptake rate parameter in Ecotracer (Validation Scenario 1), can be used to estimate CTX levels more accurately in fishes in Puerto Rico over time.

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Chapter 8. CONCLUSIONS AND RECOMMENDATIONS

Overall, the results of this study suggest that ciguatera is an ongoing human health problem in Puerto Rico and the Caribbean that can be addressed by coastal and fishery managers using the coral reef monitoring and modeling approach I have outlined here. The high cell counts and fish toxicity levels agree with the traditional ecological knowledge of fishers that the reef areas around Guayama, Puerto Rico comprise a ciguatera “hotspot” – there are frequent reports of fishes being caught there causing symptoms associated with ciguatera fish poisoning (CFP). Most fishers chose that location during open-ended interviews when given an opportunity to indicate on a map where they thought CFP occurred commonly. When we sampled the reefs using the standard screen-rig samplers, we found 35-fold higher cell counts of G*ambierdiscus* species on rigs deployed at stations on the reefs off Guayama than at the reefs near the coldspot Fajardo, Puerto Rico. There was a statistically significant large difference in median cells densities that, if persistent, could result in the higher CTX toxicity in the fish tissues that we measured at the hotspot at Guayama than the coldspot at Fajardo. The high trophic levels fishes that we sampled for CTX at the hotspot locations were significantly more toxic than the same species we tested at the coldspot locations (especially great barracuda *Sphyraena barracuda*, hogfish *Lachlonaimus maximus*, and bar jack *Caranx ruber*). All of these data strongly point to the ability of the fishers to identify CTX hotspots and coldspots.

The preliminary models that we created were a good start to begin to estimate CTX biomagnification in coral reef food webs. However, they were not based on the data collected in the later chapters, simply the data available from the literature, and needed work to be applicable to the locations we studied in Puerto Rico. We used those preliminary models to understand how Ecopath could be adapted to model CTX in the coral reef ecosystems in the Caribbean, the *Gambierdiscus* toxin production, the Ecotracer module could be used in CTX contaminant tracing, and as a proof-of-concept to submit with our grant to Puerto Rico Sea Grant. As it turned out, we discovered that Ecopath/Ecotracer modeling does not work well with biosynthesized molecules; all contaminants in Ecotracer were conceptualized as arising in the external environment outside of the biotic compartments of the model ecosystem (i.e, from an industrial source). After conferring with Ecopath users and communicating about this particular problem with the Ecopath developers (Walters and Christensen, personal communication, September 2020, wjw24@psu.edu, v.christensen@oceans.ubc.ca) about adapting the use of Ecotracer for our *Gambierdiscus* CTX biosynthesis problem, I am confident that setting the environmental concentration to 1.0 t km^{-2} (an unlimited external supply of CTX), but then calculating toxin production by *Gambierdiscus* over a fixed time period and using that CTX production for the *direct absorption rate* is the best current method to simulate production of a toxin in a biotic organism until this is addressed in the Ecopath. Indeed, varying the direct absorption rate made Ecotracer model runs quite different, so this parameter is very sensitive and the exact value that should be entered here needs to be further measured and explored. The Ecopath/Ecotracer developers suggested using the 30-day CTX production rate (as Ecopath with Ecosim has a 1-month time step), but I found that this rate over-estimated the CTX that biomagnified to the top trophic levels. As a result, I lowered the CTX production rate to one day of *Gambierdiscus* production. Additional parameters may need to be measured and specified with the current version of the Eopath and Ecotracer software, specifically the excretion rate by organisms that are not herbivorous fishes (trophic level 2). Excretion rates for CTX was set at 95% for herbivorous fishes, based on an experimental study by Ledreux *et al*. (2014), but other herbivores and consumers should have this measured and specified in Ecotracer. Further modifications of Ecotracer with an explicit biosynthesis parameter is recommended for the next version of Ecopath with Ecosim software.

Ciguatera could be contributing to economic disparity in Puerto Rico. Fishers in coldspots like Fajardo and Naguabo benefit from capturing larger, more expensive fishes than communities that live near a hotspot like Guayama where they are restricted to smaller, less valuable fishes. When traveling around Puerto Rico interviewing fishers, we asked to look in the freezers and coolers. The fish house freezers in Guayama at the suspected hotspots mostly consisted of grunts, goatfish, and smaller fishes, while the fishes in the freezers of the coldspot fish houses in Nagaubo had hogfish, grouper, and jacks. If more desirable seafood (groupers, hogfish, jacks, etc.) is available in Fajardo and not in Guayama, that might affect fishers' economic income and tourism. Tourists want to sample local, fresh cuisines and most likely want to eat prized fishes like the grouper or hogfish. Therefore, they may be more inclined to eat in restaurants that most likely carry those fish near the coldspots. This hypothesis about the economic impact of ciguatoxic fish avoidance needs to be tested by future researchers; however, we believe the disparity in fishes targeted at hotspots and coldspots could be contributing to economic differences in some municipalities and might have even a more significant impact on commercial fishers and locals in each region.

We believe that we obtained valuable information from the twenty-one open-ended interviews around Puerto Rico. However, we had plans to return and perform one hundred formal surveys in the form of yes/no questions, which would have further guided our sampling protocols and allow us to better quantify fishers' beliefs about CTX. Unfortunately, due to the SARS-CoV-2 global pandemic, traveling back to Puerto Rico was impossible. Asking informants in Puerto Rico to help complete these surveys would have put their lives and health in danger, so the surveys never materialized. We propose that future researchers take the information we found in Chapter 3 and generate a formal survey from it, adding questions as needed. The information should include questions about hotspots' locations and avoidance of more species due to CTX (especially the benthic invertebrates like gastropods and crustaceans such as crabs and lobster). Also, in the open-ended interviews, we learned that fishers in some fish houses believed there was a correlation between certain “mushroom-shaped” benthic algal species and toxic fishes. They said if they saw the specific algae, they would avoid spearfishing in that location. These algae were not further identified. We did not consider including macroalgae in our planning for pile-sort cards of organisms that might be toxic, nor were we able to go with the fishers while diving to sample these algae. We propose that future studies include a pile-sort procedure by fishers given cards showing different macroalgal species and benthic invertebrates to see if fishers have knowledge of other species associated with high CTX levels in the food web. This pile-sort procedure should be in conjunction with biological sampling and comparing benthic algae species for dinoflagellate cell counts and species identification composition using qPCR at suspected hotspot sites, as we did here for common fish species.

Ciguatoxin (CTX3C equiv.) in fishes that we sampled show that the high-trophic level species from the hotspot reefs have significantly higher toxicity levels than those same species in the coldspot reefs. In the hotspot, the measured CTX concentration in fish muscle tissue increased with trophic level, which supports Randall’s (Randall 1958) ciguatera biomagnification hypothesis and our initial hypothesis about toxicity levels increasing with effective trophic level in Ecopath and at areas identified by fishers as CTX hotspots. However, the low-trophic level species did not differ in toxicity between hotspot and coldspot areas. Low-trophic level species most likely must consume large quantities of toxic dinoflagellates to cross the 0.1 ppb threshold, and the levels we measured for herbivorous fishes in the Scaridae were much lower than this. We would need a greater sample size than we have now to have sufficient statistical power to detect a significant difference in CTX tissue concentration between parrotfishes collected at the hotspot and cold spot areas at such low tissue concentrations. In comparison, a high-trophic level species may only need to eat some small number of prey fishes to become toxic due to biomagnification. Statistical differences between areas in mean tissue CTX concentration for great barracuda and hogfish that were observed might be due to targeting high trophic level species, which resulted in larger sample sizes and high statistical power. The difference in means between areas is more likely to be observed when concentrations are high in fish tissues of the high trophic level species than when concentrations are near the CTX detection limit in the N2a assay as they are for low-trophic level species. The low-trophic level fishes can excrete CTX rapidly (Ledreux *et al.*, 2014) and had relatively low levels of CTX in our study at both the hotspot and coldspot; these results may appear to be counter to Randall’s 1958 food-chain hypothesis, where at least some elevated concentration of the toxin is necessary in herbivores for its bioaccumulation and biomagnification in the food web. However, suppose herbivorous fishes are experiencing hypo- and hyperactive behaviors due to consuming large amounts of ciguatoxin, as observed by Ledreux *et al.* (2014). Herbivorous fishes may be more vulnerable to predation at the hotspot areas, with short-term (< 24 h) elevated levels of CTX in their bodies, which could increase CTX bioaccumulation in the predators consuming them.

I tested 92 fishes for CTX3C equiv., which is one of the largest studies for a small region for CTX estimation in fishes. However, fewer fish than in this sample were used to compare the CTX in fish muscle tissues collected at the hotspot and coldspot areas; this is because the species of fishes differed at the hotspot and cold spots and we were not able to capture and sample the same species fish at each site. In part this is due to inherent differences between these sites, but also could be due to fishing pressures differing between the sites and CTX avoidance by fishers. We wanted to go back to Puerto Rico to fill those data gaps as much as possible, adding additional samples to increase statistical power to detect difference in mean CTX concentrations, but it just wasn't feasible. There were some species, like the coney *Cephalopholis fulva*, for which we had adequate CTX measurements, but only from one site (coldspot); we had obtained a few samples of a related grouper in the same family taxonomically (Epinephelidae), the red hind *Epinephelus guttatus,* from the hotspot area that could be used for comparison. Future research should include revisiting the hotspot and coldspot areas identified in this study, increasing sampling sizes for the species in all compartments of this Ecopath model, and sampling additional species at each site for comparison, while also comparing samples at the level of genera or families of fishes.

We sampled dinoflagellates at each hotspot and coldspot area, and there were 35-fold more cells at the hotspot than the coldspot. We believe the discrepancy in cell density is a primary driver for the biomagnification of CTX in the food web. Herbivorous fishes are consuming more toxic dinoflagellates in the hotspot from feeding on their preferred substrates. Unfortunately, we did not get to sample benthic algae to identify the *Gambierdiscus* species composition or measure the cell density on different species of algae. Future studies should aim to collect benthic autotrophs and identify which dinoflagellate species are present and how many cells are on each type of algae. Future studies should also look at the gut-content of herbivorous fishes and measure these fishes' consumption rates on the algae to better estimate how much ciguatoxin is entering the food web and herbivorous fishes' diets. The diets could be divided by benthic algae species and the Ecopath models could include them as separate compartments. The diet composition matrix would be more precise about which species consume macroalgae that have *Gambierdiscus* and how many cells these fishes are consuming.

The Ecosim 30-day Growth Scenario supports what our initial thoughts were about the hotspot and the coldspot. The higher cell numbers in the hotspot cause the top trophic levels to reach 0.1 ppb more quickly and reach higher CTX concentrations, although it overestimated CTX over 10 years. In the model validation procedures, I ran other scenarios with Ecotracer that produced lower CTX tissue levels consistent with what I measured in Chapter 4 and the maximum known literature values for great barracuda and hogfish. We need to sample the *Gambierdiscus* cell counts over the course of a year in the future to get realistic variation estimates for “bloom” levels on reefs at the hotspot and coldspot to further refine the Ecopath and Ecotracer models with realistic forcing functions. For some of the Ecotracer scenarios, I assumed a forcing function from a reef the Florida Keys (Bomber *et al.*, 1988), but it is unknown how well this pattern of production represented our Puerto Rican blooms. I recommend that regular surveillance of *Gambierdiscus* cells be conducted at the hotspot and coldspot areas identified in this study to better understand the variation in bloom levels. Only then can we really know if the models presented here is well validated for use in management of Puerto Rican reef fisheries.

I believe there are carbon-flow pathways in the hotspot model that are causing upper trophic level fishes to become toxic more quickly than at the coldspot. The pathway of *Gambierdiscus* with CTX through the macroalgae to the herbivorous fishes may cause top trophic level predators to become toxic less quickly than the pathway through the mollusks to the molluscivores due to the rapid excretion of CTX from herbivorous fish tissues (Ledreux et al

2014), which was included in the Ecotracer parameters for my model runs. We designed the models in this study to be a building block for future researchers to improve on. Future studies should do a more in-depth species assessment of both the hotspot and the coldspot to see how these sites differ in species diversity and gut-content analyses of species at the hotspot and coldspot for more targeted diet composition matrices at each location. The Randall data was adequate for an overall model. However, the data are dated and there are likely to be different feeding patterns in these species. An in-depth analysis of feeding behaviors would greatly improve both hotspot and coldspot models. We wanted to sample guts on the return trip to Puerto Rico, but it never happened due to the COVID-19 pandemic.

A study that could increase the quality of the models and add to the CTX literature is an analysis of CTX in sediment. Brevetoxin has already been identified in marine sediments and seagrass epiphytes and CTX is only 100 atomic mass units (AMU) lighter (Mendoza *et al.*, 2008; Hitchcock *et al.*, 2012). If CTX is found in the sediment these hotspots and coldspots then a time series of past CTX concentrations at these sites can be reconstructed. Also, fishes could be inadvertently consuming CTX's from the sediment which is another mechanism for CTX entering the food web.

The Ecosim 30-day Growth Scenario overestimated CTX over time which was evident from the models. It is highly unlikely that *S. barracuda* and *L. maximus* would ever reach the levels that are given in the Ecotracer output data. However, we believe the subsequent validated Ecosim scenarios do a much better job at estimating CTX in fishes. The estimated CTX3C equiv. from the N2a-cba assay show similar maximum values to the levels given by the validated scenarios after 120 months of biomagnification. However, the forcing functions I used were guesses based on different possible growth scenarios of toxic dinoflagellates. They may or may not be representative of what happens on the reefs. Therefore, going forward, we encourage fisheries managers in Puerto Rico to sample toxic dinoflagellates along reefs, monthly. These data would not only allow managers to understand the shift in species along reefs to understand which habitat different species like best, but also allow managers to come up with cell quantities to understand growth to create realistic forcing functions for the models. With this information, managers could use our validated Ecopath models along with their forcing function data to predict CTX levels in fishes over time.

Overall, I subscribe to statistician George Box’s aphorism

all models are wrong, but some are useful. However, the approximate nature of the model must always be borne in mind…

With the validated models presented in Chapter 7 and monthly dinoflagellates samples, I believe my models can be useful for their intended purpose; to help predict CTX levels in fishes over time.

Works Cited

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Appendix A IRB APPROVAL

EAST CAROLINA UNIVERSITY
University & Medical Center Institutional Review Board
4N-64 Brody Medical Sciences Building· Mail Stop 682
600 Moye Boulevard · Greenville, NC 27834
Office **252-744-2914** · Fax **252-744-2284** ·
www.ecu.edu/ORIC/irb

Notification of Initial Approval: Expedited

From: Social/Behavioral IRB
To: Henry Raab
CC:
Cynthia Grace-McCaskey
Date: 5/14/2018
Re: UMCIRB 18-000710
Modeling Ciguatoxin (CTX) Trophic Dynamics in a Coral Reef Ecosystem

I am pleased to inform you that your Expedited Application was approved. Approval of the study and any consent form(s) is for the period of 5/11/2018 to 5/10/2019. The research study is eligible for review under expedited category #6, 7. The Chairperson (or designee) deemed this study no more than minimal risk.

Changes to this approved research may not be initiated without UMCIRB review except when necessary to eliminate an apparent immediate hazard to the participant. All unanticipated problems involving risks to participants and others must be promptly reported to the UMCIRB. The investigator must submit a continuing review/closure application to the UMCIRB prior to the date of study expiration. The Investigator must adhere to all reporting requirements for this study.

Approved consent documents with the IRB approval date stamped on the document should be used to consent participants (consent documents with the IRB approval date stamp are found under the Documents tab in the study workspace).

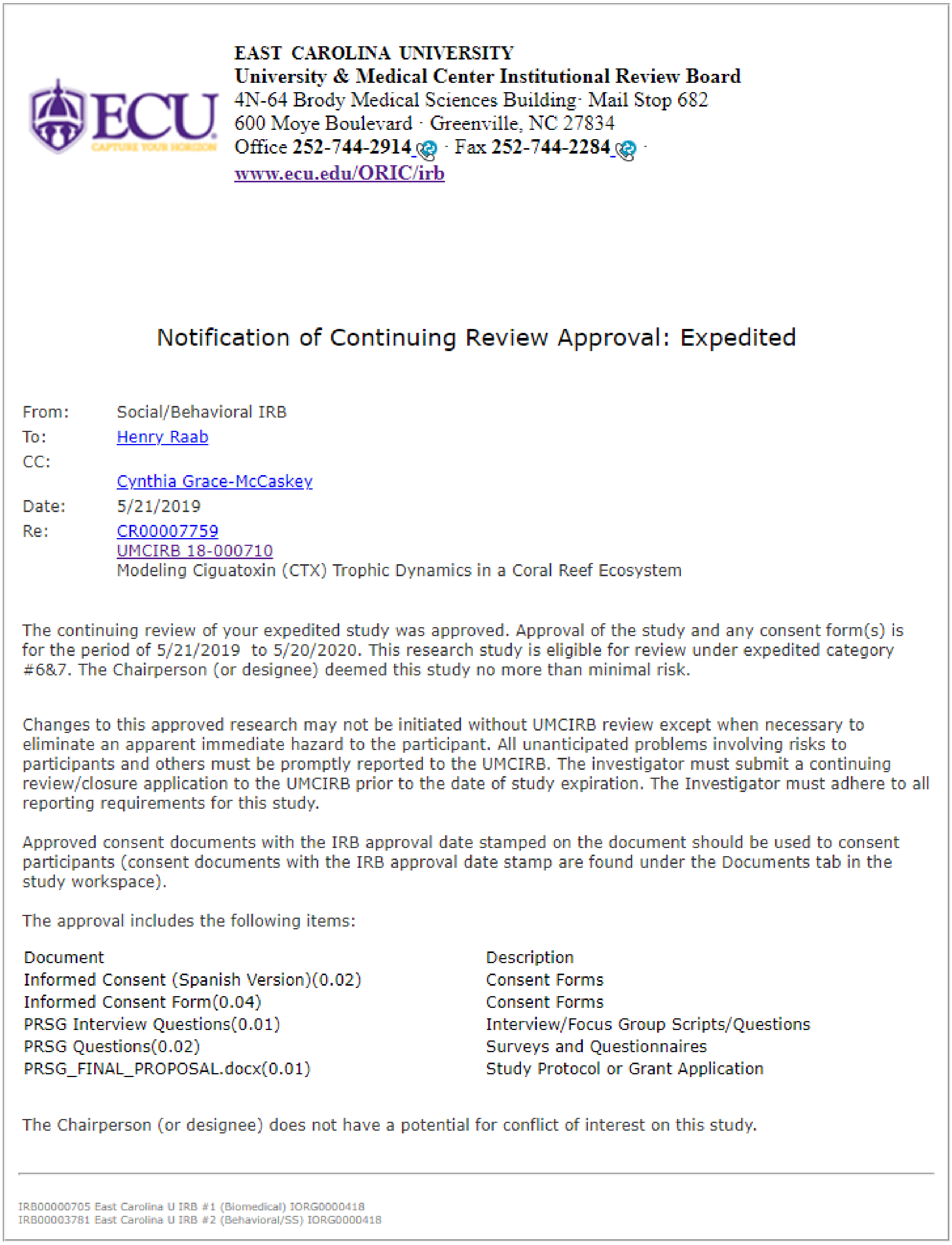
The approval includes the following items:

Name	Description
Informed Consent (Spanish Version)	Consent Forms
Informed Consent Form	Consent Forms
PRSG Interview Questions	Interview/Focus Group Scripts/Questions
PRSG Questions	Surveys and Questionnaires
PRSG_FINAL_PROPOSAL.docx	Study Protocol or Grant Application

The Chairperson (or designee) does not have a potential for conflict of interest on this study.

IRB00000705 East Carolina U IRB #1 (Biomedical) IORG0000418
IRB00003781 East Carolina U IRB #2 (Behavioral/SS) IORG0000418

Figure A-1 IRB approval for interviewing Spanish-speaking fishers in Puerto Rico and St. Thomas, USVI



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www.ecu.edu/ORIC/irb

Notification of Continuing Review Approval: Expedited

From: Social/Behavioral IRB
To: Henry Raab
CC:
Cynthia Grace-McCaskey
Date: 5/21/2019
Re: CR00007759
UMCIRB 18-000710
Modeling Ciguatoxin (CTX) Trophic Dynamics in a Coral Reef Ecosystem

The continuing review of your expedited study was approved. Approval of the study and any consent form(s) is for the period of 5/21/2019 to 5/20/2020. This research study is eligible for review under expedited category #6&7. The Chairperson (or designee) deemed this study no more than minimal risk.

Changes to this approved research may not be initiated without UMCIRB review except when necessary to eliminate an apparent immediate hazard to the participant. All unanticipated problems involving risks to participants and others must be promptly reported to the UMCIRB. The investigator must submit a continuing review/closure application to the UMCIRB prior to the date of study expiration. The Investigator must adhere to all reporting requirements for this study.

Approved consent documents with the IRB approval date stamped on the document should be used to consent participants (consent documents with the IRB approval date stamp are found under the Documents tab in the study workspace).

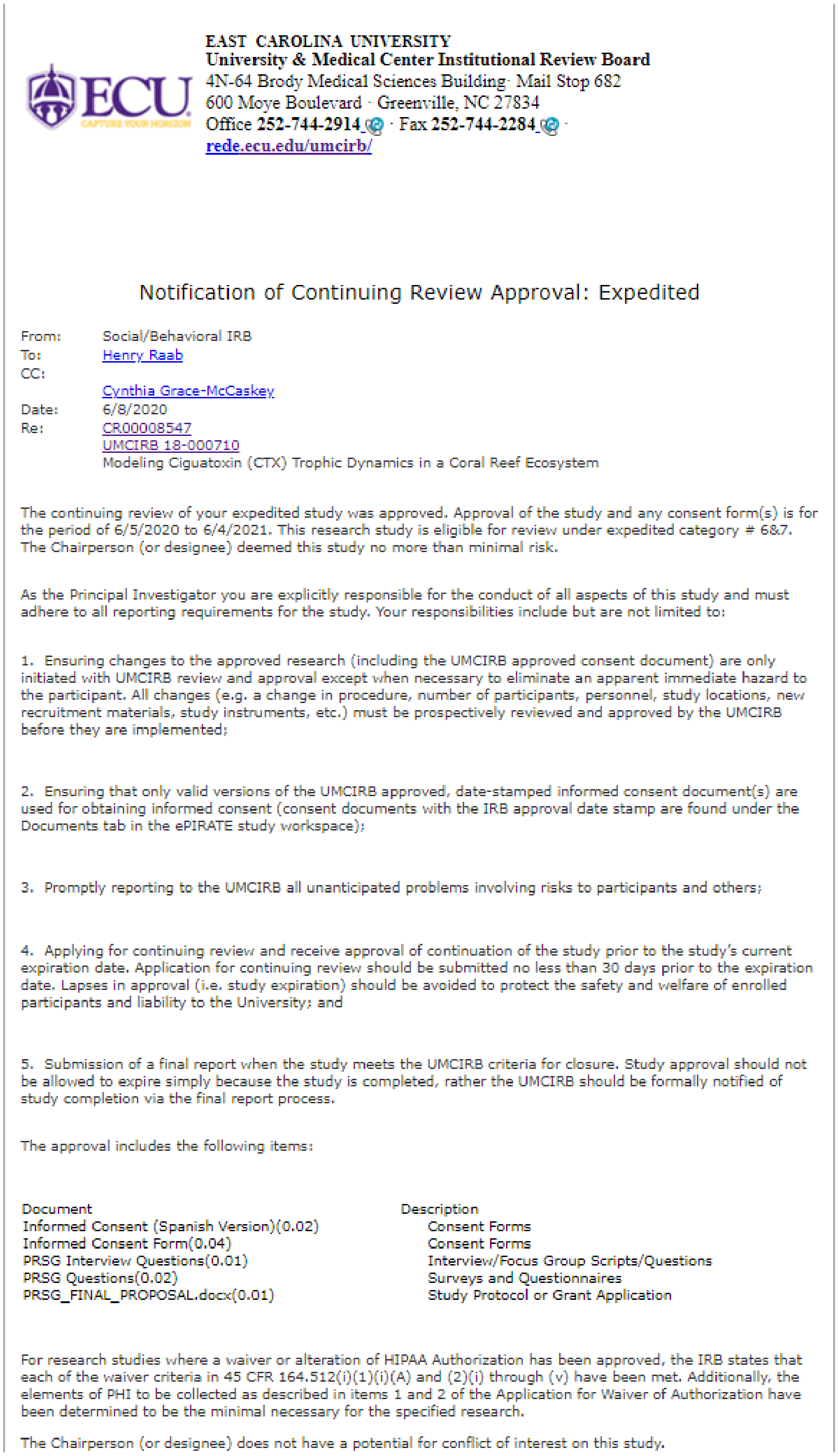
The approval includes the following items:

Document	Description
Informed Consent (Spanish Version)(0.02)	Consent Forms
Informed Consent Form(0.04)	Consent Forms
PRSG Interview Questions(0.01)	Interview/Focus Group Scripts/Questions
PRSG Questions(0.02)	Surveys and Questionnaires
PRSG_FINAL_PROPOSAL.docx(0.01)	Study Protocol or Grant Application

The Chairperson (or designee) does not have a potential for conflict of interest on this study.

IRB00000705 East Carolina U IRB #1 (Biomedical) IORG0000418
IRB00003781 East Carolina U IRB #2 (Behavioral/SS) IORG0000418

Figure A-2 2019 IRB Continuing Resolution



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Notification of Continuing Review Approval: Expedited

From: Social/Behavioral IRB
To: Henry Raab
CC:
Cynthia Grace-McCaskey
Date: 6/8/2020
Re: CR00008547
UMCIRB 18-000710
Modeling Ciguatoxin (CTX) Trophic Dynamics in a Coral Reef Ecosystem

The continuing review of your expedited study was approved. Approval of the study and any consent form(s) is for the period of 6/5/2020 to 6/4/2021. This research study is eligible for review under expedited category # 6&7. The Chairperson (or designee) deemed this study no more than minimal risk.

As the Principal Investigator you are explicitly responsible for the conduct of all aspects of this study and must adhere to all reporting requirements for the study. Your responsibilities include but are not limited to:

1. Ensuring changes to the approved research (including the UMCIRB approved consent document) are only initiated with UMCIRB review and approval except when necessary to eliminate an apparent immediate hazard to the participant. All changes (e.g. a change in procedure, number of participants, personnel, study locations, new recruitment materials, study instruments, etc.) must be prospectively reviewed and approved by the UMCIRB before they are implemented;

2. Ensuring that only valid versions of the UMCIRB approved, date-stamped informed consent document(s) are used for obtaining informed consent (consent documents with the IRB approval date stamp are found under the Documents tab in the ePIRATE study workspace);

3. Promptly reporting to the UMCIRB all unanticipated problems involving risks to participants and others;

4. Applying for continuing review and receive approval of continuation of the study prior to the study's current expiration date. Application for continuing review should be submitted no less than 30 days prior to the expiration date. Lapses in approval (i.e. study expiration) should be avoided to protect the safety and welfare of enrolled participants and liability to the University; and

5. Submission of a final report when the study meets the UMCIRB criteria for closure. Study approval should not be allowed to expire simply because the study is completed, rather the UMCIRB should be formally notified of study completion via the final report process.

The approval includes the following items:

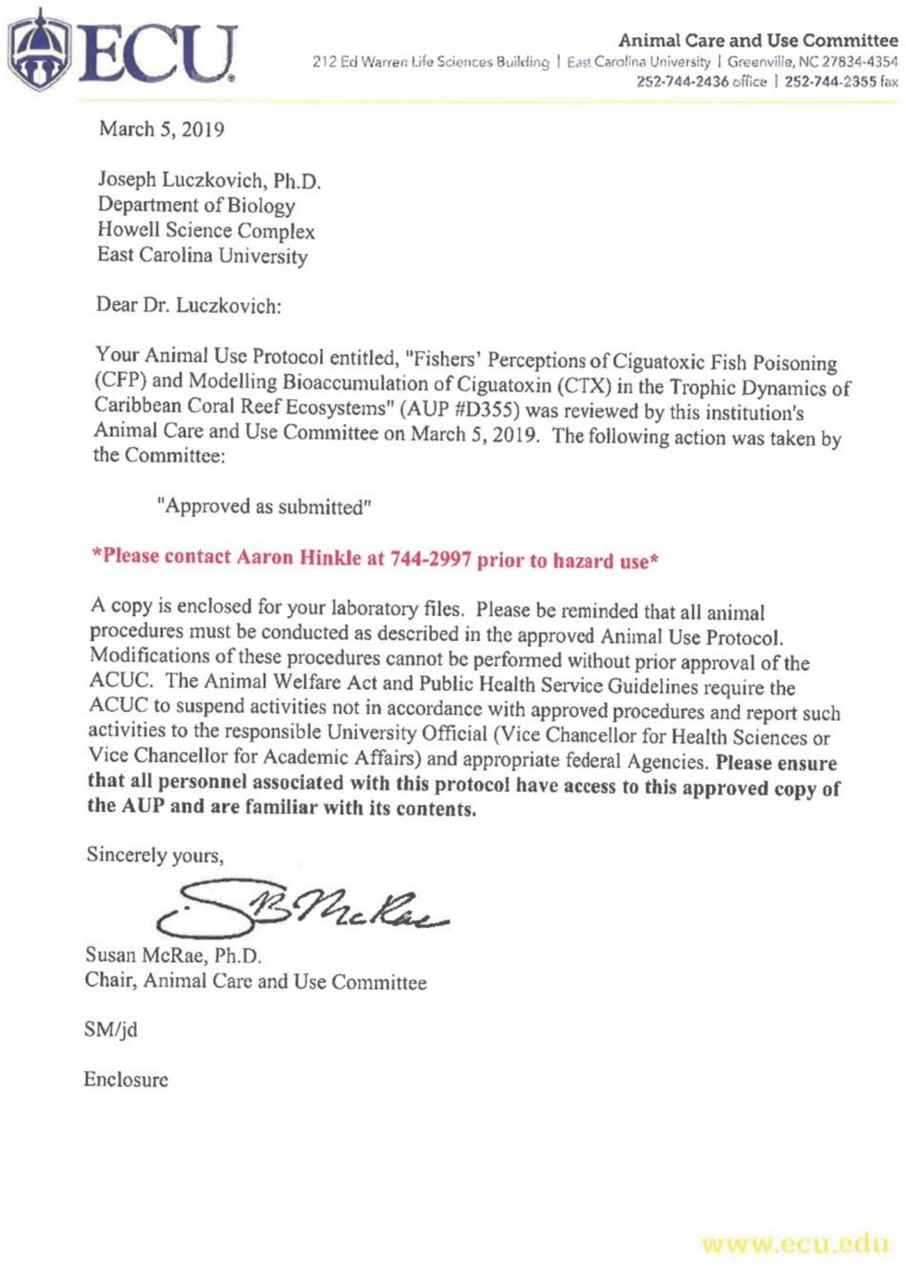
Document	Description
Informed Consent (Spanish Version)(0.02)	Consent Forms
Informed Consent Form(0.04)	Consent Forms
PRSG Interview Questions(0.01)	Interview/Focus Group Scripts/Questions
PRSG Questions(0.02)	Surveys and Questionnaires
PRSG_FINAL_PROPOSAL.docx(0.01)	Study Protocol or Grant Application

For research studies where a waiver or alteration of HIPAA Authorization has been approved, the IRB states that each of the waiver criteria in 45 CFR 164.512(i)(1)(i)(A) and (2)(i) through (v) have been met. Additionally, the elements of PHI to be collected as described in items 1 and 2 of the Application for Waiver of Authorization have been determined to be the minimal necessary for the specified research.

The Chairperson (or designee) does not have a potential for conflict of interest on this study.

Figure A-3 2020 IRB Continuing Resolution

Appendix B Animal Care and Use Protocol



ECU

Animal Care and Use Committee
212 Ed Warren Life Sciences Building | East Carolina University | Greenville, NC 27834-4354
252-744-2436 office | **252-744-2355** fax

March 5, 2019

Joseph Luczkovich, Ph.D.
Department of Biology
Howell Science Complex
East Carolina University

Dear Dr. Luczkovich:

Your Animal Use Protocol entitled, "Fishers' Perceptions of Ciguatoxic Fish Poisoning (CFP) and Modelling Bioaccumulation of Ciguatoxin (CTX) in the Trophic Dynamics of Caribbean Coral Reef Ecosystems" (AUP #D355) was reviewed by this institution's Animal Care and Use Committee on March 5, 2019. The following action was taken by the Committee:

"Approved as submitted"

Please contact Aaron Hinkle at 744-2997 prior to hazard use

A copy is enclosed for your laboratory files. Please be reminded that all animal procedures must be conducted as described in the approved Animal Use Protocol. Modifications of these procedures cannot be performed without prior approval of the ACUC. The Animal Welfare Act and Public Health Service Guidelines require the ACUC to suspend activities not in accordance with approved procedures and report such activities to the responsible University Official (Vice Chancellor for Health Sciences or Vice Chancellor for Academic Affairs) and appropriate federal Agencies. **Please ensure that all personnel associated with this protocol have access to this approved copy of the AUP and are familiar with its contents.**

Sincerely yours,

SBMcRae

Susan McRae, Ph.D.
Chair, Animal Care and Use Committee

SM/jd

Enclosure

www.ecu.edu

Figure B-1 Animal Care and Use Protocol approval for sampling fishes

Appendix C SARS-CoV-2 (COVID-19) Liability Waiver

RESEARCH ASSUMPTION OF RISK AND LIABILITY RELEASE

This is a legally-binding Release made by me, **Henry Raab** to East Carolina University (ECU) for the period of time between **12 June 2020** and 10 August 2020, during which time access to the ECU campus is restricted as part of North Carolina's effort to limit the spread of COVID-19.

I fully recognize that there are dangers and risks to which I may be exposed by participating in-person in research during the COVID-19 pandemic. I acknowledge that my in-person presence on campus or at a remote field site places me at **LOW** risk of exposure to COVID-19 because it may cause me to come into contact with individuals who unknowingly have or carry COVID-19. The following is a description and examples of specific, significant, non-obvious dangers and risks associated with this activity: personal illness that may require hospitalization and significant bodily injury, including but not limited to pneumonia, stroke or death and may also cause similar illness in others I come into contact with. I understand that the Institution does not require me to participate in this activity at this time, but I request to do so, despite the possible dangers and risks and despite this Release.

I agree to assume and take on myself all of the risks and responsibilities in any way associated with increased risk of exposure to COVID-19 due to participation in this activity. I agree that it is my responsibility to adhere to all infection prevention protocols at the research site. In consideration of and return for the services, facilities, and other assistance provided to me by the Institution in this activity, I release the Institution (and its governing board, employees, and agents) from any and all liability, claims and actions that may arise from COVID-19-induced injury or harm to me, from COVID-19-induced death or damage to my property in connection with participation in the research activities. I understand that this Release covers liability, claims and actions caused entirely or in part by any acts or failures to act of the Institution (or its governing board, employees, or agents), including but not limited to negligence, mistake, or failure to supervise by the Institution.

I recognize that this Release means I am giving up, among other things, rights to sue the Institution, its governing board, employees, and agents for injuries, damages, or losses I may incur. I also understand that this Release binds my heirs, executors, administrators, and assigns, as well as myself.

I acknowledge that my participation in the research is voluntary and not required by the Institution at this time. I acknowledge that I have been informed of my option to delay my research or otherwise not participate in the research activities due to concern of exposure to COVID-19. I understand that in some cases, not participating in research due to COVID-19 can delay completion of my degree, but the Institution will waive any requirements to complete my degree within a specific timeframe.

I have read this entire Release, I fully understand it and I agree to be legally bound by it.

THIS IS A RELEASE OF RIGHTS. READ CAREFULLY BEFORE SIGNING.

__________________________ _12 June 2020_______________

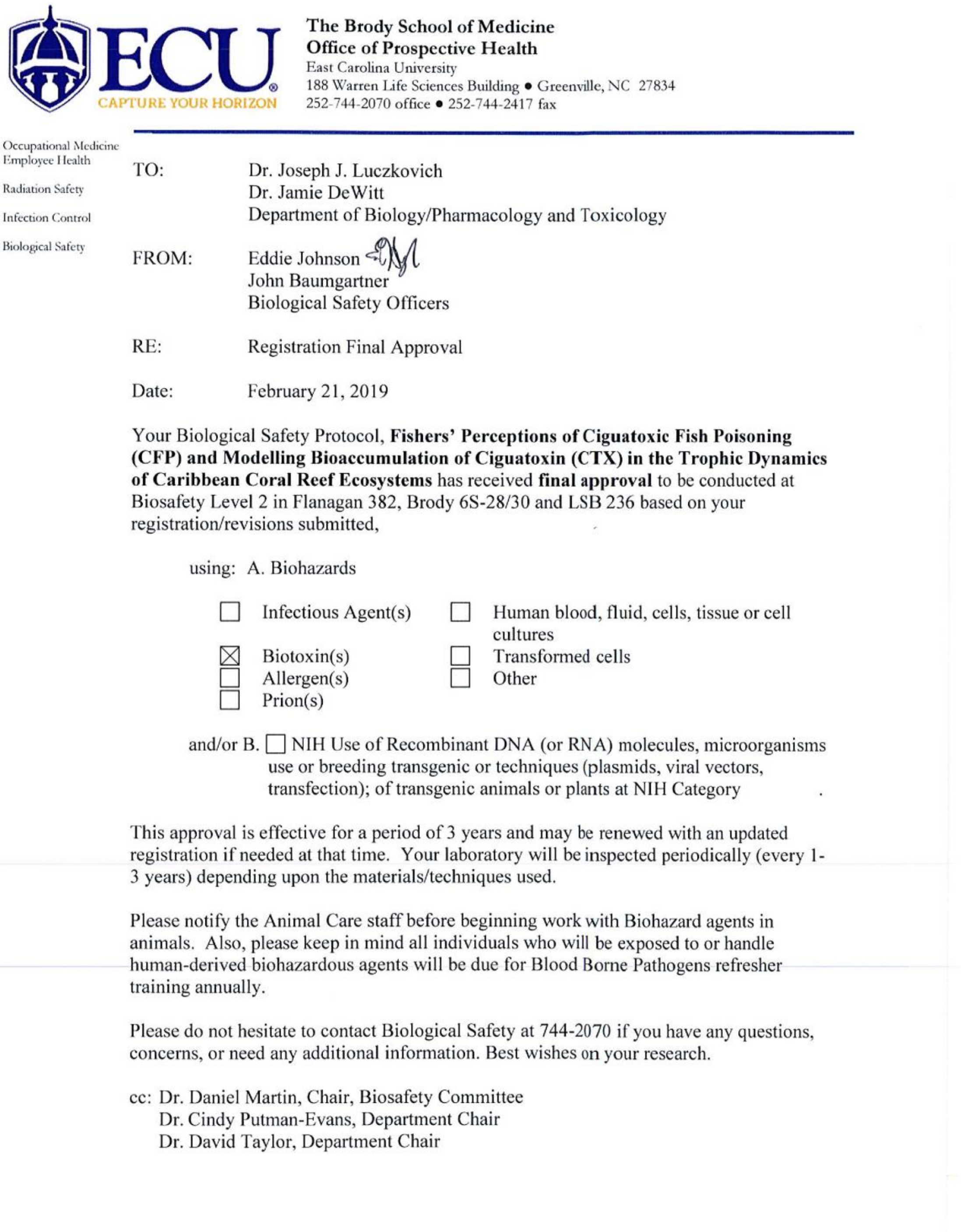
Student Signature Date

As the faculty mentor or supervisor, I have read and understood this agreement and my role in protecting the rights afforded to the student herein.

James D. Wit ____________ _6/12/20_____________________

Faculty Mentor/Supervisor Signature Date

Appendix D Institutional Biosafety Committee Approval



ECU
CAPTURE YOUR HORIZON

The Brody School of Medicine
Office of Prospective Health
East Carolina University
188 Warren Life Sciences Building ● Greenville, NC 27834
252-744-2070 office ● 252-744-2417 fax

Occupational Medicine
Employee Health
Radiation Safety
Infection Control
Biological Safety

TO: Dr. Joseph J. Luczkovich
Dr. Jamie DeWitt
Department of Biology/Pharmacology and Toxicology

FROM: Eddie Johnson
John Baumgartner
Biological Safety Officers

RE: Registration Final Approval

Date: February 21, 2019

Your Biological Safety Protocol, **Fishers' Perceptions of Ciguatoxic Fish Poisoning (CFP) and Modelling Bioaccumulation of Ciguatoxin (CTX) in the Trophic Dynamics of Caribbean Coral Reef Ecosystems** has received **final approval** to be conducted at Biosafety Level 2 in Flanagan 382, Brody 6S-28/30 and LSB 236 based on your registration/revisions submitted,

using: A. Biohazards

- ☐ Infectious Agent(s)
- ☒ Biotoxin(s)
- ☐ Allergen(s)
- ☐ Prion(s)
- ☐ Human blood, fluid, cells, tissue or cell cultures
- ☐ Transformed cells
- ☐ Other

and/or B. ☐ NIH Use of Recombinant DNA (or RNA) molecules, microorganisms use or breeding transgenic or techniques (plasmids, viral vectors, transfection); of transgenic animals or plants at NIH Category .

This approval is effective for a period of 3 years and may be renewed with an updated registration if needed at that time. Your laboratory will be inspected periodically (every 1-3 years) depending upon the materials/techniques used.

Please notify the Animal Care staff before beginning work with Biohazard agents in animals. Also, please keep in mind all individuals who will be exposed to or handle human-derived biohazardous agents will be due for Blood Borne Pathogens refresher training annually.

Please do not hesitate to contact Biological Safety at 744-2070 if you have any questions, concerns, or need any additional information. Best wishes on your research.

cc: Dr. Daniel Martin, Chair, Biosafety Committee
Dr. Cindy Putman-Evans, Department Chair
Dr. David Taylor, Department Chair

Figure D-1 Biosafety approval for working with biotoxins

Appendix E Chapter 2 Supplemental Data

Table E-1 Basic input parameters for the Ecopath with Ecosim models used in chapter 2. Detritus import (not shown) was set to 15,000 g m^{-2}. Data for Gambierdiscus spp. changed depending on the model (See Table E-2).

Group name	Biomass in habitat area (g m^{-2})	P/B (/year)	Q/B (/year)
Large sharks/Rays C	0.3	0.24	4.9
Sharks/Scombrids C	0.414	0.34	9.15
Large Jacks C	0.181	0.525	5.7
Intermediate Jacks C	1.63	1.17	9.15
Small Jacks C	1	0.83	12.5
Intermediate Reef Fish C1	3.024	0.58	6.85
Large to Intermediate Schooling Fish P	11.172	0.68	12.4
Intermediate Reef Fish C2	6.493	1.265	10.3
Hemiramphidae H	1.125	1.23	39.1
Kyphosidae H	2.42	0.6	23.6
Intermediate Reef Fish H	9.651	0.71	25.65
Large Reef Fish C	2.525	0.38	3.9
Intermediate Reef Fish C3	7.25	0.64	6.4
Small Reef Fish C1	4.419	1.665	9.8
Small Schooling Fish P	10.146	3.54	18.85
Engraulidae H	3.325	2.835	43.4
Small Reef Fish C2	1.3	3.82	14.65
Large Groupers C	0.725	0.37	2.3
Intermediate Reef Fish C4	1.431	0.55	6
Small Reef Fish O1	3.534	1.6	16.4
Small Reef Fish O2	0.99	1.505	16
Small Reef Fish O3	0.96	2.53	39.7
Large Scaridae H	18.982	0.85	13.5
Intermediate Scaridae H	5.51	1.155	20.75
Small Scaridae H	5.075	0.94	33.9
Blenniidae H	0.6	2.84	36.1
Small Gobiidae C	0.175	3.14	17.45
Sea Birds	0.017	5.4	80
Squids	1.5	1.3	17.5
Sea Turtles	0.5	0.15	3.5
Octopuses	8.4	1.9	6.76
Lobsters	3.27	1	7.4
Crabs	19	1.6	14
Shrimps/Hermit Crabs/Stomatopods	10	2.8	26.9
Small Benthic Arthropods	13.25	5	125.5
Asteroids	25	0.49	3.24
Echinoids	100	1.1	3.7
Gastropods	46.8	2.8	14
Chitons/Scaphopods	62	0.36	11.7
Polychaetes/Priapuloids/Ophiuroids	33	5.2	61.6
Holothuroids/Sipunculids/Echiuroids/Hemichordates	66.24	0.31	3.36
Bivalves	109.25	2.23	9.5
Ascidians/Banacles/Bryozoans	137.4	2.3	20
Sponges	800	1.5	5
Sea Anemones/Corals	121	1.09	9
Zooplankton	32	40	165
Microfauna	15	280	1900
Phytoplankton	40	70	
Benthic Autotrophs	1300	13.25	
Gambierdiscus spp.			
Detritus	2000		

Table E-2 Gambierdiscus spp. basic input parameters for each Ecopath simulation.

Compartment	Biomass in habitat area (g m^{-2})	P/B (/year)	Q/B (/year)
Gambierdiscus excentricus (100 cells g m^{-2})	5.620893	20.805	N/A
Gambierdiscus excentricus (100,000 cells g m^{-2})	5620.893	20.805	N/A
Gambierdiscus carolinianus (100 cells g m^{-2})	5.443886	63.51	N/A
Gambierdiscus carolinianus (100,000 cells g m^{-2})	5443.886	63.51	N/A

Table E-3 Compartment codes and diet composition matrix for the preliminary models. Data for Gambierdiscus spp. groups are missing due to the models having different values. These data can be found in Table E-4.

Compartment	Code	Compartment	Code
Large sharks/Rays C	F1	Small Gobiidae C	F27
Sharks/Scombrids C	F2	Sea Birds	B1
Large Jacks C	F3	Squids	I1
Intermediate Jacks C	F4	Sea Turtles	R1
Small Jacks C	F5	Octopuses	I2
Intermediate Reef Fish C1	F6	Lobsters	I3
Large to Intermediate Schooling Fish P	F7	Crabs	I4
Intermediate Reef Fish C2	F8	Shrimps/Hermit Crabs/Stomatopods	I5
Hemiramphidae H	F9	Small Benthic Arthropods	I6
Kyphosidae H	F10	Asteroids	I7
Intermediate Reef Fish H	F11	Echinoids	I8
Large Reef Fish C	F12	Gastropods	I9
Intermediate Reef Fish C3	F13	Chitons/Scaphopods	I10
Small Reef Fish C1	F14	Polychaetes/Priapuloids/Ophiuroids	I11
Small Schooling Fish P	F15	Holothuroids/Sipunculids/Echiuroids/Hemichordat	I12
Engraulidae H	F16	Bivalves	I13
Small Reef Fish C2	F17	Ascidians/Banacles/Bryozoans	I14
Large Groupers C	F18	Sponges	I15
Intermediate Reef Fish C4	F19	Sea Anemones/Corals	I16
Small Reef Fish O1	F20	Zooplankton	I17
Small Reef Fish O2	F21	Microfauna	I18
Small Reef Fish O3	F22	Phytoplankton	A1
Large Scaridae H	F23	Benthic Autotrophs	A2
Intermediate Scaridae H	F24	Gambierdiscus excentricus	A3
Small Scaridae H	F25	Detritus	D1
Blenniidae H	F26		

Predator/Prey Matrix	Code	F1	F2	F3	F4	F5	F6	F7
Large sharks/Rays C	**F1**	0.01	0.01	0.003	0.05	0.004	0.1	0.01
Sharks/Scombrids C	**F2**	0	0.01	0	0.034	0.05	0.015	0.176
Large Jacks C	**F3**	0	0	0	0.15	0	0.2	0
Intermediate Jacks C	**F4**	0	0	0	0	0	0.001	0.056
Small Jacks C	**F5**	0	0	0	0.002	0.001	0	0.014
Intermediate Reef Fish C1	**F6**	0	0	0	0	0	0.001	0
Large to Intermediate Schooling Fish P	**F7**	0	0	0	0	0	0	0.003
Intermediate Reef Fish C2	**F8**	0	0	0	0.001	0	0	0.001
Hemiramphidae H	**F9**	0	0	0	0	0	0	0
Kyphosidae H	**F10**	0	0	0	0	0	0	0
Intermediate Reef Fish H	**F11**	0	0	0	0	0	0	0
Large Reef Fish C	**F12**	0	0	0.004	0.024	0.02	0.03	0.04
Intermediate Reef Fish C3	**F13**	0	0	0	0	0.003	0.01	0.002
Small Reef Fish C1	**F14**	0	0	0	0	0	0	0.002
Small Schooling Fish P	**F15**	0	0	0	0	0	0	0
Engraulidae H	**F16**	0	0	0	0	0	0	0
Small Reef Fish C2	**F17**	0	0	0	0	0	0	0
Large Groupers C	**F18**	0.005	0.005	0	0.01	0.01	0.085	0
Intermediate Reef Fish C4	**F19**	0	0	0	0	0	0	0
Small Reef Fish O1	**F20**	0	0	0	0	0	0	0.003
Small Reef Fish O2	**F21**	0	0	0	0	0	0	0
Small Reef Fish O3	**F22**	0	0	0	0	0	0	0
Large Scaridae H	**F23**	0	0	0	0	0	0	0
Intermediate Scaridae H	**F24**	0	0	0	0	0	0	0
Small Scaridae H	**F25**	0	0	0	0	0	0	0
Blenniidae H	**F26**	0	0	0	0	0	0	0
Small Gobiidae C	**F27**	0	0	0	0	0	0	0
Sea Birds	**B1**	0	0.02	0.01	0.06	0.03	0.01	0.28
Squids	**I1**	0	0	0	0	0	0	0.15
Sea Turtles	**R1**	0	0	0	0	0	0	0
Octopuses	**I2**	0	0	0	0	0	0	0
Lobsters	**I3**	0	0	0	0	0	0	0
Crabs	**I4**	0	0	0	0	0	0	0
Shrimps/Hermit Crabs/Stomatopods	**I5**	0	0	0	0	0	0	0
Small Benthic Arthropods	**I6**	0	0	0	0	0	0	0
Asteroids	**I7**	0	0	0	0	0	0	0
Echinoids	**I8**	0	0	0	0	0	0	0
Gastropods	**I9**	0	0	0	0	0	0	0
Chitons/Scaphopods	**I10**	0	0	0	0	0	0	0
Polychaetes/Priapuloids/Ophiuroids	**I11**	0	0	0	0	0	0	0
Holothuroids/Sipunculids/Echiuroids/ Hemichordates	**I12**	0	0	0	0	0	0	0
Bivalves	**I13**	0	0	0	0	0	0	0
Ascidians/Banacles/Bryozoans	**I14**	0	0	0	0	0	0	0
Sponges	**I15**	0	0	0	0	0	0	0
Sea Anemones/Corals	**I16**	0	0	0	0	0	0	0
Zooplankton	**I17**	0	0	0	0	0	0	0
Microfauna	**I18**	0	0	0	0	0	0	0
Phytoplankton	**A1**	0	0	0	0	0	0	0
Benthic Autotrophs	**A2**	0	0	0	0	0	0	0
***Gambierdiscus* spp.**	**A3**	0	0	0	0	0	0	0
Detritus	**D1**	0	0	0	0	0	0	0

Code	F8	F9	F10	F11	F12	F13	F14	F15	F16	F17
F1	0.01	0.002	0.06	0.052	0.03	0.025	0.02	0.049	0.028	0.006
F2	0.04	0.115	0.02	0.02	0.014	0.084	0.02	0.136	0.04	0.02
F3	0.16	0	0	0.044	0	0.124	0	0	0	0
F4	0.11	0	0.015	0.107	0	0	0	0.11	0.107	0.001
F5	0	0	0	0.02	0	0	0	0.006	0.019	0
F6	0.009	0	0.003	0.004	0	0.057	0.005	0.062	0	0.003
F7	0	0	0	0.01	0	0	0	0.047	0.006	0
F8	0.02	0	0.003	0.004	0.001	0.015	0	0.067	0.002	0.001
F9	0	0	0	0	0	0	0	0.2	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0	0	0	0	0	0	0	0	0
F12	0.04	0	0.003	0.023	0.005	0.04	0.001	0.059	0.001	0
F13	0.01	0	0.003	0.009	0	0.012	0.01	0.002	0.002	0.01
F14	0.002	0.002	0	0	0	0	0.003	0.002	0.002	0.002
F15	0	0	0	0	0	0	0	0.002	0.002	0
F16	0	0	0	0	0	0	0	0	0	0
F17	0.002	0	0	0.001	0	0	0.008	0	0	0.003
F18	0.09	0	0.02	0.02	0.232	0	0	0	0	0
F19	0	0	0	0	0	0	0	0	0	0
F20	0.003	0.003	0	0	0	0	0.004	0.004	0.004	0.004
F21	0	0	0	0	0	0	0.003	0.003	0.003	0.003
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0	0	0
F26	0	0	0	0	0	0	0	0	0	0
F27	0	0	0	0	0	0	0	0	0	0
B1	0.04	0.1	0	0.01	0.005	0.03	0.025	0.2	0.1	0.008
I1	0	0.015	0	0	0	0	0	0.13	0.03	0
R1	0	0	0	0	0	0	0	0	0	0
I2	0.002	0	0	0.002	0.001	0.003	0.007	0	0	0.001
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0.01	0	0	0.007
I5	0.01	0	0	0	0	0	0.01	0	0.01	0.005
I6	0	0	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0	0	0
A3	0	0	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0	0	0

Code	F18	F19	F20	F21	F22	F23	F24	F25	F26	F27
F1	0.093	0.02	0.025	0.004	0.001	0.025	0.002	0.001	0	0
F2	0	0.006	0.014	0.001	0	0.03	0.001	0.002	0.001	0
F3	0	0	0	0	0	0	0	0	0	0
F4	0	0.005	0.053	0.053	0.053	0.016	0.073	0.122	0.053	0
F5	0	0	0	0	0	0	0	0	0	0
F6	0	0.003	0.02	0	0	0.03	0.016	0.016	0	0
F7	0	0	0	0	0	0	0	0	0	0
F8	0	0.003	0	0	0	0.02	0.03	0.01	0	0
F9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0	0	0	0	0	0	0	0	0
F12	0	0.003	0.017	0.006	0	0.01	0.01	0.008	0.001	0
F13	0	0.003	0.04	0.001	0.015	0.055	0.007	0.006	0.002	0.004
F14	0	0	0.005	0.002	0.006	0	0	0.002	0.006	0.002
F15	0	0	0	0	0	0	0	0	0	0
F16	0	0	0	0	0	0	0	0	0	0
F17	0	0	0.001	0	0.004	0	0	0	0.001	0.002
F18	0	0.01	0	0	0	0.248	0	0	0	0
F19	0	0	0	0	0	0	0	0	0	0
F20	0	0	0.006	0.004	0.004	0	0	0.004	0.004	0.003
F21	0	0	0	0.003	0.003	0	0	0	0.003	0.002
F22	0	0	0	0	0	0	0	0	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0	0	0
F26	0	0	0	0	0	0	0	0	0	0
F27	0	0	0	0	0	0	0	0	0	0
B1	0	0.005	0.005	0.003	0	0.002	0.005	0.002	0	0
I1	0	0	0	0	0	0	0	0	0	0
R1	0	0	0	0	0	0	0	0	0	0
I2	0	0	0.002	0.002	0	0	0	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0	0	0	0	0
I5	0	0	0	0	0	0	0	0	0	0
I6	0	0	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0	0	0	0	0
I8	0	0	0	0	0	0	0	0	0	0
I9	0	0	0	0	0	0	0	0	0	0
I10	0	0	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0	0	0
I12	0	0	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0	0	0
A3	0	0	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0	0	0

Code	B1	I1	R1	I2	I3	I4	I5	I6	I7	I8
F1	0.02	0.005	0.005	0.02	0.005	0.05	0.05	0	0	0.008
F2	0	0.01	0	0.045	0.005	0.02	0.05	0	0	0
F3	0	0	0	0	0	0.01	0.015	0	0	0.082
F4	0	0.004	0	0	0	0.004	0.031	0	0	0
F5	0	0	0	0	0	0	0	0	0	0
F6	0	0.001	0	0.014	0	0.09	0.03	0	0.001	0.05
F7	0	0	0	0	0	0	0.004	0	0	0
F8	0	0	0	0.05	0.001	0.1	0.02	0.031	0	0.1
F9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0	0	0	0	0	0	0.004	0	0	0
F12	0	0.001	0	0.009	0.002	0.032	0.03	0	0	0.1
F13	0	0	0	0.03	0	0.15	0.1	0.042	0	0.06
F14	0	0	0	0	0	0.1	0.04	0.042	0	0.11
F15	0	0	0	0	0	0	0	0.01	0	0
F16	0	0	0	0	0	0	0	0	0	0
F17	0	0	0	0.02	0	0.063	0.15	0.1	0	0
F18	0	0	0.035	0	0.16	0.07	0	0	0	0
F19	0	0	0	0	0	0.001	0.002	0	0.002	0.003
F20	0	0	0	0	0	0.01	0.01	0.056	0	0
F21	0	0	0	0	0	0.05	0.04	0.035	0.021	0.05
F22	0	0	0	0	0	0	0	0.008	0	0
F23	0	0	0	0	0	0	0	0	0	0
F24	0	0	0	0	0	0	0	0	0	0
F25	0	0	0	0	0	0	0	0	0	0
F26	0	0	0	0	0	0	0	0	0	0
F27	0	0	0	0	0	0	0	1	0	0
B1	0	0	0	0	0	0	0	0	0	0
I1	0	0.01	0	0	0	0	0.1	0	0	0
R1	0	0	0	0	0.06	0.025	0.04	0	0.02	0.02
I2	0	0	0	0.02	0	0.025	0.02	0	0	0
I3	0	0	0	0	0	0	0	0	0	0
I4	0	0	0	0	0	0.005	0.01	0.005	0	0.05
I5	0	0.005	0	0.025	0.005	0.002	0.002	0.04	0.01	0.11
I6	0	0	0	0	0	0	0	0	0	0
I7	0	0	0	0	0	0.01	0	0	0.08	0.07
I8	0	0	0	0	0	0	0	0	0.006	0.015
I9	0	0	0	0	0.002	0.002	0.004	0.006	0	0
I10	0	0	0	0	0	0	0	0	0	0
I11	0	0	0	0	0	0	0	0.015	0	0
I12	0	0	0	0	0	0	0	0	0	0
I13	0	0	0	0	0	0	0	0	0	0
I14	0	0	0	0	0	0	0	0	0	0
I15	0	0	0	0	0	0	0	0	0	0
I16	0	0	0	0	0	0	0	0	0	0
I17	0	0	0	0	0	0	0	0	0	0
I18	0	0	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0	0	0
A3	0	0	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0	0	0

Code	I9	I10	I11	I12	I13	I14	I15	I16	I17	I18
F1	0.01	0	0.05	0.05	0.067	0	0	0	0	0
F2	0.008	0	0.006	0	0.007	0	0	0	0	0
F3	0.158	0	0	0	0.057	0	0	0	0	0
F4	0.004	0	0	0	0	0	0	0	0.022	0
F5	0	0	0	0	0	0	0	0	0.938	0
F6	0.017	0	0.006	0.003	0.01	0.06	0.2	0.01	0.25	0
F7	0	0	0.12	0	0	0	0	0	0.722	0
F8	0.035	0.02	0.18	0.06	0.073	0.05	0.07	0.004	0.018	0
F9	0	0	0	0	0	0	0	0	0	0
F10	0	0	0	0	0	0	0	0	0	0
F11	0.001	0	0.002	0	0	0	0	0	0.002	0
F12	0.4	0	0	0	0.075	0	0	0	0.006	0
F13	0.004	0.01	0.05	0.01	0.023	0.04	0.07	0.003	0.15	0
F14	0.144	0.04	0.1	0.008	0.06	0	0	0	0.26	0
F15	0	0	0	0	0	0.03	0	0	0.95	0.006
F16	0	0	0	0	0	0	0	0	0.2	0
F17	0.005	0.002	0.079	0.001	0.03	0	0	0	0.5	0
F18	0	0	0	0	0	0	0	0	0	0
F19	0	0	0.006	0.006	0	0.023	0.921	0.007	0.001	0
F20	0.038	0	0.05	0	0.02	0.02	0.02	0.02	0.1	0
F21	0.035	0	0.1	0.005	0.027	0.02	0.08	0.1	0.109	0.005
F22	0	0	0.05	0	0.071	0	0	0	0.2	0
F23	0	0	0	0	0	0	0.002	0.001	0	0
F24	0	0	0	0	0	0	0.001	0	0	0
F25	0	0	0	0	0	0	0	0	0	0
F26	0.001	0	0	0	0	0	0	0	0.005	0
F27	0	0	0	0	0	0	0	0	0	0
B1	0	0	0	0	0	0	0	0	0.05	0
I1	0	0	0	0	0	0	0	0	0.565	0
R1	0.025	0	0	0	0.02	0.02	0.34	0.016	0	0
I2	0.5	0.01	0.005	0	0.4	0	0	0	0	0
I3	0.15	0	0.05	0	0.5	0	0	0	0	0
I4	0.05	0	0.004	0.001	0.05	0.02	0.05	0.01	0.03	0.03
I5	0.08	0.01	0.05	0.005	0.212	0	0	0	0.15	0.055
I6	0	0	0	0	0	0.05	0.06	0.04	0.04	0.04
I7	0.135	0.005	0.03	0.005	0.145	0.02	0.02	0.012	0	0.02
I8	0	0	0.004	0.005	0.01	0	0.05	0.05	0	0
I9	0.02	0.02	0.05	0.015	0.04	0.058	0.04	0.015	0	0.03
I10	0	0	0	0	0	0	0	0	0	0
I11	0.01	0	0.03	0	0.03	0.04	0.05	0.01	0.05	0.05
I12	0	0	0	0	0	0	0	0	0	0.1
I13	0	0	0	0	0	0	0	0	0	0.1
I14	0	0	0	0	0	0	0	0	0.15	0.1
I15	0	0	0	0	0	0	0	0	0	0.001
I16	0	0	0	0	0	0	0	0	0.15	0.1
I17	0	0	0	0	0	0	0	0	0	0.6
I18	0	0	0	0	0	0	0	0	0	0
A1	0	0	0	0	0	0	0	0	0	0
A2	0	0	0	0	0	0	0	0	0	0
A3	0	0	0	0	0	0	0	0	0	0
D1	0	0	0	0	0	0	0	0	0	0

Code	A1	A2	A3	D1
F1	0	0		0.02
F2	0	0		0
F3	0	0		0
F4	0	0		0
F5	0	0		0
F6	0	0.029		0
F7	0	0.088		0
F8	0	0.01		0
F9	0	0.8		0
F10	0	1		0
F11	0	0.533		0.458
F12	0	0		0
F13	0	0.052		0
F14	0	0.058		0
F15	0	0		0
F16	0.7	0		0.1
F17	0	0.028		0
F18	0	0		0
F19	0	0.028		0
F20	0	0.326		0.28
F21	0	0.2		0.1
F22	0	0.335		0.336
F23	0	0.997		0
F24	0	0.999		0
F25	0	1		0
F26	0	0.497		0.497
F27	0	0		0
B1	0	0		0
I1	0	0		0
R1	0	0.414		0
I2	0	0		0
I3	0	0.18		0.12
I4	0.02	0.6		0.048
I5	0	0.084		0.12
I6	0	0.6		0.17
I7	0	0.088		0.36
I8	0	0.86		0
I9	0	0.4		0.298
I10	0	0		0
I11	0.1	0.245		0.37
I12	0.01	0.13		0.76
I13	0.15	0		0.75
I14	0.2	0		0.55
I15	0	0		0.999
I16	0	0.65		0.1
I17	0.2	0		0.2
I18	0	0.05		0.95
A1	0	0		0
A2	0	0		0
A3	0	0		0
D1	0	0		0

Table E-4 Diet matrix data for the benthic autotrophs (A2) and *Gambierdiscus* spp. (A3) prey items. The benthic autotroph compartment as a proportion of the predator diet was reduced based on the biomass of dinoflagellates. This follows the assumption that the dinoflagellates reside on the benthic autotrophs.

100 cells *G. excentricus*			**100,000 cells *G. excentricus***			**100 cells *G. carolinianus***			**100,000 cells *G. carolinianus***		
Code	A2	A3	Code	A2	A3	**Code**	A2	A3	Code	A2	A3
F1	0	0	F1	0	0	**F1**	0	0	F1	0	0
F2	0	0	F2	0	0	**F2**	0	0	F2	0	0
F3	0	0	F3	0	0	**F3**	0	0	F3	0	0
F4	0	0	F4	0	0	**F4**	0	0	F4	0	0
F5	0	0	F5	0	0	**F5**	0	0	F5	0	0
F6	0.02887	0.00013	F6	0.00545	0.02355	**F6**	0.02888	0.00012	F6	0.00559	0.02341
F7	0.08762	0.00038	F7	0.01654	0.07146	**F7**	0.08763	0.00037	F7	0.01696	0.07104
F8	0.00996	4.3E-05	F8	0.00188	0.00812	**F8**	0.00996	4.2E-05	F8	0.00193	0.00807
F9	0.79654	0.00346	F9	0.15038	0.64962	**F9**	0.79665	0.00335	F9	0.15421	0.64579
F10	0.99568	0.00432	F10	0.18797	0.81203	**F10**	0.99581	0.00419	F10	0.19277	0.80723
F11	0.5307	0.0023	F11	0.10019	0.43281	**F11**	0.53077	0.00223	F11	0.10274	0.43026
F12	0	0	F12	0	0	**F12**	0	0	F12	0	0
F13	0.05178	0.00022	F13	0.00977	0.04223	**F13**	0.05178	0.00022	F13	0.01002	0.04198
F14	0.05775	0.00025	F14	0.0109	0.0471	**F14**	0.05776	0.00024	F14	0.01118	0.04682
F15	0	0	F15	0	0	**F15**	0	0	F15	0	0
F16	0	0	F16	0	0	**F16**	0	0	F16	0	0
F17	0.02788	0.00012	F17	0.00526	0.02274	**F17**	0.02788	0.00012	F17	0.0054	0.0226
F18	0	0	F18	0	0	**F18**	0	0	F18	0	0
F19	0.02788	0.00012	F19	0.00526	0.02274	**F19**	0.02788	0.00012	F19	0.0054	0.0226
F20	0.32459	0.00141	F20	0.06128	0.26472	**F20**	0.32463	0.00137	F20	0.06284	0.26316
F21	0.19914	0.00086	F21	0.03759	0.16241	**F21**	0.19916	0.00084	F21	0.03855	0.16145
F22	0.33355	0.00145	F22	0.06297	0.27203	**F22**	0.3336	0.0014	F22	0.06458	0.27042
F23	0.99269	0.00431	F23	0.18741	0.80959	**F23**	0.99283	0.00418	F23	0.19219	0.80481
F24	0.99468	0.00432	F24	0.18778	0.81122	**F24**	0.99482	0.00418	F24	0.19257	0.80643
F25	0.99568	0.00432	F25	0.18797	0.81203	**F25**	0.99581	0.00419	F25	0.19277	0.80723
F26	0.49485	0.00215	F26	0.09342	0.40358	**F26**	0.49492	0.00208	F26	0.09581	0.40119
F27	0	0	F27	0	0	**F27**	0	0	F27	0	0
B1	0	0	B1	0	0	**B1**	0	0	B1	0	0
I1	0	0	I1	0	0	**I1**	0	0	I1	0	0
R1	0.41221	0.00179	R1	0.07782	0.33618	**R1**	0.41227	0.00173	R1	0.07981	0.33419
I2	0	0	I2	0	0	**I2**	0	0	I2	0	0
I3	0.17922	0.00078	I3	0.03383	0.14617	**I3**	0.17925	0.00075	I3	0.0347	0.1453
I4	0.59741	0.00259	I4	0.11278	0.48722	**I4**	0.59749	0.00251	I4	0.11566	0.48434
I5	0.08364	0.00036	I5	0.01579	0.06821	**I5**	0.08365	0.00035	I5	0.01619	0.06781
I6	0.59741	0.00259	I6	0.11278	0.48722	**I6**	0.59749	0.00251	I6	0.11566	0.48434
I7	0.08762	0.00038	I7	0.01654	0.07146	**I7**	0.08763	0.00037	I7	0.01696	0.07104
I8	0.85628	0.00372	I8	0.16165	0.69835	**I8**	0.8564	0.0036	I8	0.16578	0.69422
I9	0.39827	0.00173	I9	0.07519	0.32481	**I9**	0.39832	0.00168	I9	0.07711	0.32289
I10	0.62927	0.00273	I10	0.1188	0.5132	**I10**	0.62935	0.00265	I10	0.12183	0.51017
I11	0.24394	0.00106	I11	0.04605	0.19895	**I11**	0.24397	0.00103	I11	0.04723	0.19777
I12	0.12944	0.00056	I12	0.02444	0.10556	**I12**	0.12946	0.00054	I12	0.02506	0.10494
I13	0	0	I13	0	0	**I13**	0	0	I13	0	0
I14	0	0	I14	0	0	**I14**	0	0	I14	0	0
I15	0	0	I15	0	0	**I15**	0	0	I15	0	0
I16	0.64719	0.00281	I16	0.12218	0.52782	**I16**	0.64728	0.00272	I16	0.1253	0.5247
I17	0	0	I17	0	0	**I17**	0	0	I17	0	0
I18	0.04978	0.00022	I18	0.0094	0.0406	**I18**	0.04979	0.00021	I18	0.00964	0.04036

Table E-5 Ecotracer proportion of contaminant excreted data. All compartments that were lower than 3.0 ETL in the model that directly consumed *Gambierdiscus* spp. were given an excretion value of 0.95 due to the metabolism and excretion of the contaminant by herbivorous fish, like *Mugil cephalus*, shown by Ledreux *et al*. (2014).

Code	Group name	Prop. of contaminant excreted
F1	Large sharks/Rays C	0
F2	Sharks/Scombrids C	0
F3	Large Jacks C	0
F4	Intermediate Jacks C	0
F5	Small Jacks C	0
F6	Intermediate Reef Fish C1	0
F7	Large to Intermediate Schooling Fish P	0
F8	Intermediate Reef Fish C2	0
F9	Hemiramphidae H	0.95
F10	Kyphosidae H	0.95
F11	Intermediate Reef Fish H	0.95
F12	Large Reef Fish C	0
F13	Intermediate Reef Fish C3	0
F14	Small Reef Fish C1	0
F15	Small Schooling Fish P	0
F16	Engraulidae H	0.95
F17	Small Reef Fish C2	0
F18	Large Groupers C	0
F19	Intermediate Reef Fish C4	0.95
F20	Small Reef Fish O1	0.95
F21	Small Reef Fish O2	0.95
F22	Small Reef Fish O3	0.95
F23	Large Scaridae H	0.95
F24	Intermediate Scaridae H	0.95
F25	Small Scaridae H	0.95
F26	Blenniidae H	0.95
F27	Small Gobiidae C	0
B1	Sea Birds	0
I1	Squids	0
R1	Sea Turtles	0
I2	Octopuses	0
I3	Lobsters	0
I4	Crabs	0
I5	Shrimps/Hermit Crabs/Stomatopods	0
I6	Small Benthic Arthropods	0
I7	Asteroids	0
I8	Echinoids	0
I9	Gastropods	0
I10	Chitons/Scaphopods	0
I11	Polychaetes/Priapuloids/Ophiuroids	0
I12	Holothuroids/Sipunculids/Echiuroids/Hemichorda	0
I13	Bivalves	0
I14	Ascidians/Banacles/Bryozoans	0
I15	Sponges	0
I16	Sea Anemones/Corals	0
I17	Zooplankton	0
I18	Microfauna	0
A1	Phytoplankton	0
A2	Benthic Autotrophs	0
A3	Gambierdiscus excentricus	0
D1	Detritus	0

Table E-6 Growth and toxicity data of the two species used in the preliminary models. (Litaker *et al.*, 2017).

Strain	**Growth rate (d^{-1})**	**Toxicity (fg CTX3c eq. $cell^{-1}$)**	**Weight g $cell^{-1}$**
G. carolinianus	0.17 ± 0.017	0.027 ± 0.43	4.2E-05
G. excentricus	0.057 ± 0.002	469 ± 10	4.3E-05

Table E-7 Ecotracer input for the four different dinoflagellate parameters in the model.

Group name	Initial conc. (t/t)	Direct absorption rate
Gambierdiscus carolinianus 100 cells	6.45E-12	5.41E-09
Gambierdiscus carolinianus 100,000 cells	6.45E-12	5.41E-06
Gambierdiscus excentricus 100 cells	1.08E-08	2.43E-07
Gambierdiscus excentricus 100,000 cells	1.08E-08	0.000243322

Appendix F Chapter 3 Supplemental Data

Questions asked to informants.

1. How many years have you been fishing?
2. Have you ever had an experience with CTX (self or someone you know)?
3. Where do you think CTX is most prevalent?
4. What months do you think the fish are most toxic?
5. Which fish species are most likely to have CTX (see pile sort)?
6. What type of gear do you use to catch those fish?
7. How do you detect if a fish has the toxin?
    a. Insect test (insects avoid tissue with CTX)
    b. Animal test (feed to cats to see adverse reactions)
    c. Anatomy test (does the tissue or organs look or smell different)
    d. The coin rub test (rub with a coin, does the coin change colors)

Interview 1 Cabo Rojo Experience: 47 years

Pile sort

Species	Hot	Cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Amberjack, Black Jack, Hogfish

Interview 2 Fajardo Experience: 39 years

Pile Sort

Species	Hot	Intermediate	Cold
Hogfish	**X**		
Barracuda	**X**		
King Mackerel			**X**
Cero			**X**
Black Jack		**X**	
Amberjack		**X**	
Bluerunner			**X**
Horse-eye Jack		**X**	
Jack Travel			**X**
Cubera Snapper			**X**
Queen Snapper			**X**
Silk Snapper			**X**
Blackfin Snapper			**X**
Lane Snapper			**X**
Mutton Snapper			**X**
Mangrove Snapper			**X**
Yellowtail Snapper			**X**
Schoolmaster			**X**
Dog Snapper			**X**
Tiger Grouper			**X**
Red Hind			**X**
Coney			**X**
Yellowfin Grouper		**X**	
Queen Parrotfish			**X**
Rainbow Parrotfish		**X**	
Stoplight Parrotfish			**X**
Stripped Mojarra			**X**
Yellowfin Mojarra			**X**
Sand Tilefish			**X**
Spadefish			**X**
Trunkfish			**X**
Redear Sardine			**X**
White Mullet			**X**
Ballyhoo			**X**
Blue Crab			**X**
Queen Conch			**X**
West Indian Topshell			**X**

Free-listing: Barracuda, Hogfish, Escolar, Amberjack

Interview 3 Fajardo Experience: 67 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, Amberjack, Black Jack, Horse-eye Jack

Interview 4 Fajardo Experience: 33 years

Pile sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack		**X**
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack		**X**
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, African Pompano, Amberjack

Interview 5 Fajardo Experience: 36

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack		**X**
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, Horse-eye Jack, Amberjack

Interview 6 Fajardo Experience: 40 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, Amberjack, African Pompano, Horse-eye Jack, Black Jack

Interview 7 Guayama Experience: 18 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster	**X**	
Dog Snapper	**X**	
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Hogfish, Barracuda, King Mackerel, Amberjack, Black Jack, Dog Snapper, Schoolmaster, Horse-eye Jack

Interview 8 Guayama Experience: 28 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster	**X**	
Dog Snapper	**X**	
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Amberjack, Bar Jack, Hogfish, Black Jack, Horse-eye Jack, Barracuda, Dog Snapper

Interview 9 Guayama Experience: 32 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster	**X**	
Dog Snapper	**X**	
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Amberjack, Hogfish, Barracuda, Dog Snapper, King Mackerel, Black Jack, Horse-eye Jack, Schoolmaster

Interview 10 Guayama Experience: 25 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster	**X**	
Dog Snapper	**X**	
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, Amberjack, Dog Snapper, Black Jack, Horse-eye Jack, Schoolmaster, King Mackerel

Interview 11 Guayama Experience: 23 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster	**X**	
Dog Snapper	**X**	
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, Amberjack, Black Jack, King Mackerel

Interview 12 Arroyo Experience: 30 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Hogfish, Barracuda, Amberjack, King Mackerel

Interview 13 Arroyo Experience: 58 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Hogfish, Barracuda, Amberjack, King Mackerel

Interview 14 Arroyo Experience: 27 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack		**X**
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, King Mackerel

Intervew 15 Juana Diaz Experience: 44 years

Pile Sort

Species	Hot	cold
Hogfish		**X**
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack		**X**
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster	**X**	
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Schoolmaster, Yellow Goatfish, Almaco Jack, Amberjack, Black Jack (no hogfish)

Interview 16 Juana Diaz Experience: 31 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel		**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, Hogfish, Blackjack, Horse-eye Jack

Interview 17 Juana Diaz Experience: 25 Years

Pile Sort

Species	Hot	cold
Hogfish		
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero	**X**	**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-List: Barracuda, Black Jack, Amberjack, King Mackerel

Interview 18 Ponce Experience: 45 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper	**X**	
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Hogfish, Barracuda, King Mackerel, Dog Snapper, Amberjack, African Pompano, Horse-eye Jack, Cobia (possibly rainbowrunner)

Interview 19 Ponce Experience: 37 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper		**X**
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper		**X**
Tiger Grouper		**X**
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Barracuda, African Pompano, Hogfish, King Mackerel, Cobia (Rainbowrunner)

Interview 20 Maunabo Experience: 20 years

Pile Sort

Species	Hot	cold
Hogfish	**X**	
Barracuda	**X**	
King Mackerel	**X**	**X**
Cero		**X**
Black Jack	**X**	
Amberjack	**X**	
Bluerunner		**X**
Horse-eye Jack	**X**	
Jack Travel		**X**
Cubera Snapper	**X**	
Queen Snapper		**X**
Silk Snapper		**X**
Blackfin Snapper		**X**
Lane Snapper		**X**
Mutton Snapper		**X**
Mangrove Snapper		**X**
Yellowtail Snapper		**X**
Schoolmaster		**X**
Dog Snapper	**X**	
Tiger Grouper	**X**	
Red Hind		**X**
Coney		**X**
Yellowfin Grouper		**X**
Queen Parrotfish		**X**
Rainbow Parrotfish		**X**
Stoplight Parrotfish		**X**
Stripped Mojarra		**X**
Yellowfin Mojarra		**X**
Sand Tilefish		**X**
Spadefish		**X**
Trunkfish		**X**
Redear Sardine		**X**
White Mullet		**X**
Ballyhoo		**X**
Blue Crab		**X**
Queen Conch		**X**
West Indian Topshell		**X**

Free-list: Hogfish, Dog Snapper, King Mackerel, Barracuda, Amberjack, Horse-eye Jack, Black Jack

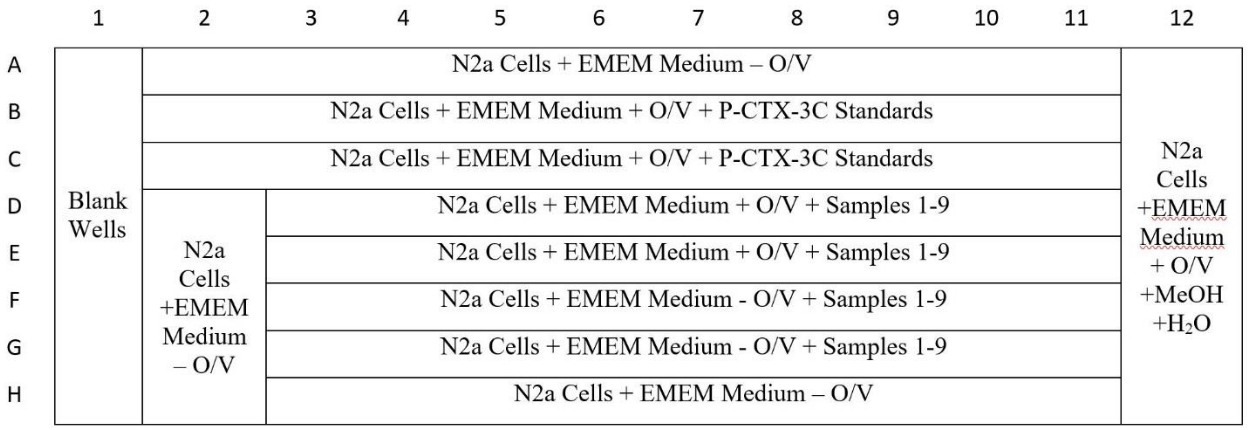
Interview 21 Naguabo Experience: 24 years

Didn't have the cards with me, so no pile sorting was done

Free list: Barracuda, Hogfish, Amberjack, Black Jack, Dog Snapper, Cero, Schoolmaster, African Pompano

Appendix G   Chapter 4 Supplemental Data

Table G-1 The 96-Well plate setup for N2-cba assay. Each well-contained 120µl of total volume. Samples and the standard curve were added in 10µl aliquots to 110µl of medium with or without O/V. Column 12, rows A-H contained 110µl of the medium, 2µl of MeOH, and 8µl H_2O as a negative control. Column 2 rows D-H were used as a positive control for O/V with expected results of 20% cell death. Row A columns 2-11 and row H columns 3-11 were used as negative controls to see the maximum growth of cells with no treatment as a baseline. Ouabain and veratridine were added in concentrations of 31.3µM and 3.13 µM, respectively.



	1	2	3	4	5	6	7	8	9	10	11	12
A	Blank Wells	N2a Cells + EMEM Medium – O/V										N2a Cells +EMEM Medium + O/V +MeOH $+H_2O$
B		N2a Cells + EMEM Medium + O/V + P-CTX-3C Standards										
C		N2a Cells + EMEM Medium + O/V + P-CTX-3C Standards										
D		N2a Cells +EMEM Medium – O/V	N2a Cells + EMEM Medium + O/V + Samples 1-9									
E			N2a Cells + EMEM Medium + O/V + Samples 1-9									
F			N2a Cells + EMEM Medium - O/V + Samples 1-9									
G			N2a Cells + EMEM Medium - O/V + Samples 1-9									
H			N2a Cells + EMEM Medium – O/V									

Table G-2 Absorbance data for plate 5 from the N2a assay.

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.04635	1.17377	1.42563	1.31086	1.07475	1.04203	1.20124	1.4878	1.00486	1.01939	1.089825	1.45683
B	0.04645	0.15867	0.16623	0.15962	0.38207	0.43218	0.60333	0.73288	0.94846	0.96198	0.985528	1.35687
C	0.04675	0.16856	0.18639	0.13714	0.39103	0.42466	0.59061	0.68887	0.87024	0.96561	0.974585	1.51632
D	0.0466	0.96901	0.0148	0.88185	0.76634	0.8869	0.91297	0.79897	0.69725	0.95012	Blank Cells	1.11371
E	0.04555	0.97695	0.03421	0.98651	0.79223	0.89197	0.84091	0.75187	0.74149	0.89874		1.41333
F	0.04578	0.95552	0.0249	1.14218	1.28274	1.12134	1.29463	1.12275	1.07585	1.40571		1.29017
G	0.04569	0.97378	0.02337	1.21673	0.99917	0.912	1.30515	0.95105	0.99588	1.10995		1.3588
H	0.04528	0.98472	1.54295	1.30827	1.35985	1.47532	1.32127	1.53815	1.66519	1.68822	1.438224	1.6675

Table G-3 Absorbance data for plate 6 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0467	0.8276	0.9202	0.992	0.8177	0.9526	1.0357	0.9236	1.0136	1.0427	1.1145	1.172
B	0.0467	0.1058	0.102	0.1056	0.184	0.2251	0.4141	0.7157	0.8575	0.7896	0.7779	1.0053
C	0.0471	0.1047	0.101	0.1188	0.1983	0.2637	0.4487	0.6497	0.7245	0.7652	0.8325	1.1193
D	0.0465	0.7979	0.6524	0.795	0.78646	0.3455	0.68453	0.793	0.2605	0.8821	Blank Cells	1.0478
E	0.0457	0.745	0.543	0.804	0.81266	0.2678	0.7887	0.814	0.2093	0.725		1.0623
F	0.0452	0.8483	0.8557	0.7971	0.707	0.7138	0.801	0.8105	0.8448	0.808		1.136
G	0.0463	0.7185	1.0866	0.814	0.832	0.8238	0.7601	0.816	0.9613	0.796		1.1998
H	0.0463	0.7393	0.9352	0.9812	1.145	1.1387	1.0691	1.2593	1.0354	1.0628	1.1899	1.1412

Table G-4 Absorbance data for plate 7 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0459	1.1347	1.003	1.1418	1.1162	1.1212	1.8356	1.1023	1.3426	2.0892	1.4463	1.3809
B	0.0469	0.044	0.0477	0.0505	0.1127	0.3075	0.6485	0.9595	1.4111	1.3359	1.3868	1.0999
C	0.0467	0.0374	0.0566	0.066	0.1843	0.3159	0.6533	0.9683	1.1401	1.145	1.1656	1.8906
D	0.0468	1.0917	0.751	0.9195	0.3349	0.4778	0.5103	0.5564	0.3127	0.4742	Blank Cells	1.5014
E	0.0466	1.1517	0.7137	0.7827	0.3276	0.3446	0.4967	0.4993	0.3718	0.4488		1.826
F	0.0458	1.2739	0.9385	1.5664	1.4953	1.5003	0.9104	0.9783	1.407	1.6269		1.5659
G	0.0464	1.2231	1.0719	1.2847	1.7675	1.4839	0.9066	1.0662	1.4453	1.7603		1.6102
H	0.0464	1.1286	1.3547	1.4127	1.5653	1.2603	1.651	1.9258	1.6484	1.5231	1.2251	0.8801

Table G-5 Absorbance data for plate 8 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0467	1.643	1.5251	1.406	1.4017	1.3051	1.3915	1.3073	1.3955	1.4008	1.3913	1.2673
B	0.0469	0.0983	0.0852	0.0876	0.2165	0.3902	0.5303	0.7786	1.0535	1.0398	1.0659	1.3265
C	0.0614	0.0677	0.0832	0.1709	0.2288	0.324	0.5704	0.8865	0.9547	1.0458	1.0452	1.2317
D	0.0448	1.0277	0.6104	0.7471	0.7538	0.7496	0.7718	0.7914	0.8045	0.357	Blank Cells	1.2451
E	0.047	1.0314	0.7436	0.7071	0.7996	0.7342	0.7909	0.703	0.7145	0.3155		1.5897
F	0.0456	1.0837	1.1668	1.5576	1.3109	1.4024	1.5134	1.4589	1.4317	1.362		1.6708
G	0.0492	1.07198	1.1627	1.3286	1.1968	1.5232	1.428	1.5193	1.6115	1.2871		1.1577
H	0.046	0.9719	1.4673	1.4938	1.2384	1.4858	1.4032	1.5155	1.5003	1.5662	1.3764	1.1873

Table G-6 Absorbance data for plate 9 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0464	2.1012	2.0803	2.0604	2.0983	2.0813	2.0963	2.0283	2.0778	2.0831	2.0699	1.8488
B	0.0462	0.2245	0.2861	0.2667	0.3883	0.6947	0.876	1.3183	1.7913	1.7393	1.7235	1.9972
C	0.0468	0.2874	0.2929	0.2842	0.3972	0.4829	0.9197	1.3511	1.3955	1.3756	1.4498	1.8949
D	0.0459	1.5654	1.1348	1.0152	1.5279	1.4177	1.4444	1.4243	1.2926	1.2228	Blank Cells	2.103
E	0.0453	1.6704	1.5946	1.5101	1.0233	1.389	1.4213	1.7484	1.5411	1.5785		1.8395
F	0.0445	1.552	2.0283	2.0993	2.5341	1.7629	1.9763	1.8125	1.8674	2.113		1.9335
G	0.0457	1.7744	2.0282	2.0279	2.0354	2.0029	1.8925	2.1979	2.1071	1.8722		1.9834
H	0.0453	1.3746	1.8067	1.9382	1.8557	1.8723	1.8807	1.8924	1.8992	1.9171	1.833	1.8239

Table G-7 Absorbance data for plate 10 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0463	1.3045	1.2328	1.2454	1.1988	1.2348	1.222	1.2493	1.2255	1.3194	1.2812	1.3304
B	0.0472	0.1022	0.1114	0.1491	0.2181	0.3684	0.5665	0.777	0.9898	1.0426	1.0619	1.2743
C	0.0475	0.0997	0.1499	0.1202	0.1859	0.3188	0.5639	0.7733	1.0001	0.9813	1.0676	1.2708
D	0.046	1.0046	0.8122	0.7947	0.2813	0.6095	0.3921	0.3266	0.6275	0.3931	Blank Cells	1.2674
E	0.0458	1.0081	0.8605	0.8136	0.3596	0.6496	0.4897	0.2741	0.6025	0.4875		1.2274
F	0.0503	1.1083	1.1314	1.0116	0.9865	1.1081	0.8614	1.1155	1.0583	0.8159		1.2992
G	0.0459	1.1084	1.0371	1.0954	1.1043	1.0467	1.1062	1.0238	1.3958	0.8208		1.3298
H	0.0466	0.9211	1.3555	1.4171	1.4331	1.5125	1.5087	1.4683	1.4362	1.3171	1.2299	1.37

Table G-8 Absorbance data for plate 11 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.046	0.8809	0.8355	0.8987	0.8792	0.8139	0.881	0.808	0.8883	0.8656	0.8921	0.8621
B	0.0456	0.0436	0.0585	0.0553	0.0675	0.1904	0.2833	0.4199	0.6499	0.6471	0.5908	1.0185
C	0.0457	0.0408	0.0442	0.0456	0.1566	0.187	0.3328	0.4287	0.543	0.6022	0.6056	0.9297
D	0.0447	0.6213	0.0748	0.0686	0.4678	0.1379	0.1647	0.3234	0.1927	0.2354	Blank Cells	0.8829
E	0.0444	0.6362	0.0702	0.0681	0.4876	0.0957	0.3717	0.3404	0.2733	0.2313		1.0091
F	0.045	0.6419	0.1496	0.0682	0.7562	0.7478	0.6062	0.5563	0.3697	0.5297		1.001
G	0.0446	0.6346	0.3011	0.0691	0.8114	0.6945	0.7038	0.5782	0.4199	0.6666		0.9634
H	0.0443	0.6573	0.8632	0.9359	0.8778	0.9481	0.913	0.9691	9.9108	0.9509	0.9812	0.967

Table G-9 Absorbance data for plate 12 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0483	0.6153	0.6838	0.6865	0.6679	0.6555	0.6554	0.6416	0.6356	0.6403	0.6719	0.6998
B	0.0519	0.0735	0.0926	0.0906	0.1537	0.1961	0.2863	0.414	0.4921	0.5258	0.5041	0.6758
C	0.0491	0.0819	0.0901	0.1159	0.1483	0.181	0.2885	0.4181	0.4989	0.495	0.4885	0.6718
D	0.0492	0.5065	0.1479	0.512	0.5106	0.1028	0.387	0.3922	0.473	0.1014	Blank Cells	0.6635
E	0.049	0.4984	0.1437	0.4886	0.4483	0.2884	0.2185	0.4533	0.4509	0.1676		0.6702
F	0.0516	0.5106	0.4961	0.6639	0.4878	0.571	0.465	0.6294	0.4225	0.5233		0.6631
G	0.0479	0.4956	0.5881	0.5558	0.4895	0.5033	0.5478	0.4976	0.4539	0.6278		0.6922
H	0.0482	0.5023	0.6789	0.6914	0.6556	0.6321	0.6185	0.6374	0.614	0.6335	0.6466	0.6665

Table G-10 Absorbance data for plate 13 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0462	1.6199	1.3651	1.5802	1.6861	1.6425	1.6495	1.6144	1.6464	1.6092	1.6237	1.7141
B	0.049	0.2456	0.3339	0.3154	0.3333	0.5702	0.9319	1.3482	1.541	1.5145	1.5776	1.6491
C	0.0494	0.2879	0.3135	0.323	0.3402	0.5314	0.9116	1.1954	1.5251	1.5638	1.5066	1.6727
D	0.0496	1.208	0.5599	0.4599	0.6822	0.4652	1.2668	1.2827	1.1514	1.1729	Blank Cells	1.7045
E	0.0497	1.5112	0.6867	0.6402	0.5848	0.5861	1.1053	1.3796	1.2092	0.9612		1.8642
F	0.05	1.3505	1.1257	1.2202	1.2041	1.543	1.2673	1.4431	1.3151	1.3162		1.7051
G	0.049	1.2916	0.9773	1.3393	1.472	1.2959	1.2973	1.3347	1.4126	1.1898		1.7002
H	0.0509	1.3299	1.9445	2.0978	1.9147	1.8434	1.8025	1.7919	1.7457	1.7092	1.6971	1.6758

Table G-11 Absorbance data for plate 14 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0461	1.1289	1.3321	1.3293	1.3312	1.354	1.0279	1.1174	1.3252	0.9252	0.7222	0.894
B	0.0468	0.237	0.2512	0.2413	0.5125	0.848	1.0047	1.2587	1.7097	1.7211	1.6983	0.9752
C	0.0478	0.2967	0.2566	0.2559	0.5754	0.8471	1.0157	1.321	1.6954	1.7959	1.779	1.4644
D	0.0445	0.8062	0.0729	0.511	0.078	0.0828	0.08	0.7644	0.0841	0.074	Blank Cells	1.1293
E	0.0476	1.001	0.0732	0.5543	0.0753	0.0735	0.0812	0.5607	0.077	0.0732		1.3826
F	0.0472	0.9322	0.1839	0.9938	0.0737	0.2151	0.1058	0.8892	0.3555	0.0679		1.0076
G	0.0459	0.892	0.1817	1.0532	0.0728	0.2589	0.0874	1.0746	0.5231	0.0712		0.9993
H	0.0467	0.8619	1.2844	1.3401	1.4017	1.3769	1.3326	1.1575	1.3818	1.4358	1.1874	1.179

Table G-12 Absorbance data for plate 15 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0469	1.2808	1.2895	1.3122	1.3586	1.3634	1.3636	1.3603	1.3698	1.3817	1.3233	1.3874
B	0.0469	0.2954	0.3851	0.3157	0.3595	0.5004	0.7197	0.9683	1.2042	1.2511	1.2373	1.4079
C	0.0471	0.2761	0.3731	0.3053	0.3507	0.5181	0.761	0.9169	1.2329	1.3181	1.2115	1.2793
D	0.0476	1.2307	1.1974	0.9781	0.8405	0.9873	0.8781	1.1185	0.7056	1.0975	0.5064	1.6657
E	0.0477	1.1827	1.1042	0.9823	0.8549	1.0798	0.8552	1.1127	0.7191	1.0548	0.5187	1.5403
F	0.0459	1.2248	1.425	1.3266	1.3736	1.4675	1.3693	1.418	1.3936	1.3962	1.4012	1.4135
G	0.0461	1.2647	2.2384	1.4072	1.3793	1.3708	1.3643	1.3761	1.4031	1.3756	1.3954	1.8231
H	0.0458	1.2711	1.4318	1.4354	1.3489	1.4131	1.3794	1.4628	1.4032	1.4442	1.5204	1.4597

Table G-13 Absorbance data for plate 16 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.047	1.5568	1.6265	1.5248	1.5358	1.5366	1.5148	1.5011	1.5211	1.5482	1.4993	1.4854
B	0.0468	0.1875	0.1952	0.2232	0.2345	0.3458	0.6259	0.9584	1.1148	1.1565	1.2015	1.4079
C	0.0469	0.1811	0.1994	0.2221	0.2415	0.3658	0.6554	0.9847	1.1365	1.1785	1.1945	1.4358
D	0.0464	1.1815	0.4516	0.5564	0.5994	1.1541	1.0259	1.1134	0.9847	1.1854	1.0215	1.4878
E	0.0482	1.1944	0.4687	0.5841	0.5941	1.1261	0.9954	1.1277	1.0172	1.1658	1.001	1.5785
F	0.048	1.1221	1.4526	1.4154	1.4359	1.4265	1.3648	1.4015	1.5348	1.5641	1.5741	1.5365
G	0.0472	1.1665	1.4148	1.3844	1.3948	1.4441	1.3698	1.4229	1.5511	1.5484	1.5662	1.5648
H	0.046	1.1548	1.4658	1.5591	1.5384	1.5214	1.4859	1.6028	1.5028	1.5489	1.5546	1.5241

Table G-14 Absorbance data for plate 17 from the N2a assay

<>	1	2	3	4	5	6	7	8	9	10	11	12
A	0.0468	1.5429	1.5171	1.5463	1.4902	1.5003	1.4951	1.5303	1.4789	1.4883	1.5217	1.5226
B	0.0488	0.2449	0.2548	0.2648	0.4152	0.5264	0.7726	1.0847	1.1997	1.1954	1.2115	1.5284
C	0.0421	0.25674	0.2627	0.2664	0.4187	0.5146	0.7594	1.0826	1.1984	1.1939	1.2168	1.5674
D	0.0445	1.213	1.1954	1.1006	0.5224	0.6524	0.7715	0.6899	1.0325	1.1154	0.8955	1.5183
E	0.0425	1.2005	1.1898	1.2366	0.5264	0.6218	0.7534	0.6548	1.0366	1.1086	0.8638	1.5295
F	0.0461	1.1954	1.4658	1.3647	1.4001	1.3554	1.4099	1.5004	1.4958	1.4537	1.4582	1.5481
G	0.047	1.2351	1.4453	1.3224	1.3996	1.3657	1.3957	1.5162	1.4871	1.4581	1.4662	1.5119
H	0.0443	1.2278	1.5272	1.4951	1.5315	1.5441	1.495	1.4986	1.4975	1.5345	1.527	1.495

Table G-15 Complete list of all fishes sampled and tested for CTX in ppb CTX3C equiv. Plate number corresponds to plates in Tables G-2 to G-14. BACH and Maunabo were not identified as hotspots or coldspots. Samples are sorted from lowest CTX3C equiv. levels to the highest.

Fish ID	Location	Spot	ppb CTX3C equiv.	Plate Number	Species	Common Name
GYHP001	Guayama	Hot	1.21833E-06	6	*Haemulon plumierii*	White Grunt
NGLM002	Naguabo	Cold	1.82603E-06	6	*Lachnolaimus maximus*	Hogfish
C1ST001	CTX-1	Cold	2.66914E-06	9	*Scarus taeniopterus*	Princess Parrotfish
GYEG001	Guayama	Hot	3.43946E-06	12	*Epinephelus guttatus*	Red Hind
C1CF002	CTX-1	Cold	3.48786E-06	5	*Cepahlopholis fulva*	Coney
C4OC003	CTX-4	Hot	4.34569E-06	5	*Ocyurus chrysurus*	Yellowtail Snapper
C1CF004	CTX-1	Cold	8.49482E-06	5	*Cepahlopholis fulva*	Coney
C1CF005	CTX-1	Cold	1.04048E-05	5	*Cepahlopholis fulva*	Coney
FASB001	Fajardo	Cold	1.087E-05	12	*Sphyraena barracuda*	Great Barracuda
MASR005	Maunabo		1.45179E-05	6	*Scomberomorus regalis*	Cero
BASB004	Bayamon		1.91776E-05	12	*Sphyraena barracuda*	Great Barracuda
FALJ001	Fajardo	Cold	2.34495E-05	6	*Lutjanus jocu*	Dog Snapper
C1SV001	CTX-1	Cold	2.41754E-05	16	*Sparisoma viride*	Stoplight Parrotfish
C1CF001	CTX-1	Cold	2.88687E-05	9	*Cepahlopholis fulva*	Coney
C2CF005	CTX-2	Cold	3.25505E-05	9	*Cepahlopholis fulva*	Coney
C4SV001	CTX-4	Hot	3.58031E-05	9	*Sparisoma viride*	Stoplight Parrotfish
GYEG002	Guayama	Hot	3.63833E-05	9	*Epinephelus guttatus*	Red Hind
C1CF003	CTX-1	Cold	4.05383E-05	5	*Cepahlopholis fulva*	Coney
BASB005	Bayamon		4.16289E-05	10	*Sphyraena barracuda*	Great Barracuda
GYHR001	Guayama	Hot	4.24913E-05	11	*Holocentrus rufus*	Squirrelfish
C4OC001	CTX-4	Hot	4.25319E-05	5	*Ocyurus chrysurus*	Yellowtail Snapper
C2OC001	CTX-2	Cold	4.58052E-05	9	*Ocyurus chrysurus*	Yellowtail Snapper
GYHR005	Guayama	Hot	4.6876E-05	12	*Holocentrus rufus*	Squirrelfish
C2CF007	CTX-2	Cold	4.7427E-05	13	*Cepahlopholis fulva*	Coney
GYHR003	Guayama	Hot	7.38761E-05	9	*Holocentrus rufus*	Squirrelfish
MACH001	Maunabo		7.76988E-05	8	*Caranx hippos*	Crevalle Jack
C4MP001	CTX-4	Hot	7.86195E-05	9	*Malacanthus plumieri*	Sand Tilefish
FASP004	Fajardo	Cold	7.86195E-05	9	*Sphyraena picudilla*	Southern Sennet
C2CF004	CTX-2	Cold	7.99984E-05	8	*Cepahlopholis fulva*	Coney
C4OC002	CTX-4	Hot	8.03192E-05	5	*Ocyurus chrysurus*	Yellowtail Snapper
C2CF003	CTX-2	Cold	8.91125E-05	16	*Cepahlopholis fulva*	Coney
FALM001	Fajardo	Cold	9.89636E-05	8	*Lachnolaimus maximus*	Hogfish
C2CE001	CTX-2	Cold	9.89971E-05	13	*Cephalopholis cruentata*	Graysby
GYEG003	Guayama	Hot	0.000101617	13	*Epinephelus guttatus*	Red Hind
NGLM004	Naguabo	Cold	0.000102441	8	*Lachnolaimus maximus*	Hogfish
GYCP001	Guayama	Hot	0.000113471	8	*Calamus pennatula*	Pluma Porgy
NGLM003	Naguabo	Cold	0.000120055	7	*Lachnolaimus maximus*	Hogfish
C4ST001	CTX-4	Hot	0.00012831	16	*Scarus taeniopterus*	Princess Parrotfish
C2CF002	CTX-2	Cold	0.00015769	8	*Cepahlopholis fulva*	Coney
NGLM005	Naguabo	Cold	0.000199693	7	*Lachnolaimus maximus*	Hogfish
GYHR002	Guayama	Hot	0.000223932	11	*Holocentrus rufus*	Squirrelfish
BASB001	Bayamon		0.000274565	12	*Sphyraena barracuda*	Great Barracuda
MASR003	Maunabo		0.0002955	17	*Scomberomorus regalis*	Cero
C3SV001	CTX-3	Hot	0.0003138	17	*Sparisoma viride*	Stoplight Parrotfish
C2CF006	CTX-2	Cold	0.000416783	16	*Cepahlopholis fulva*	Coney
C2SV003	CTX-2	Cold	0.000416783	16	*Sparisoma viride*	Stoplight Parrotfish
GYSC001	Guayama	Hot	0.00044	5	*Scomberomorus cavalla*	King Mackeral
MASR002	Maunabo		0.000448043	11	*Scomberomorus regalis*	Cero
C2SC001	CTX-2	Cold	0.000448639	16	*Sparisoma chrysopterum*	Redtail Parrotfish
NGLM006	Naguabo	Cold	0.000520883	7	*Lachnolaimus maximus*	Hogfish
C4SV002	CTX-4	Hot	0.000592376	17	*Sparisoma viride*	Stoplight Parrotfish
BASB002	Bayamon		0.00063042	10	*Sphyraena barracuda*	Great Barracuda
BASB003	Bayamon		0.000634912	10	*Sphyraena barracuda*	Great Barracuda
MASR001	Maunabo		0.0006401	17	*Scomberomorus regalis*	Cero
C2CR002	CTX-2	Cold	0.000674135	11	*Caranx ruber*	Bar Jack
C4MP002	CTX-4	Hot	0.000674135	11	*Malacanthus plumieri*	Sand Tilefish
FASP002	Fajardo	Cold	0.000674135	11	*Sphyraena picudilla*	Southern Sennet
FASB002	Fajardo	Cold	0.000780011	6	*Sphyraena barracuda*	Great Barracuda
NGSB001	Naguabo	Cold	0.000800106	7	*Sphyraena barracuda*	Great Barracuda

BASB007	Bayamon		0.000864584	13	*Sphyraena barracuda*	Great Barracuda
BASB006	Bayamon		0.000906114	13	*Sphyraena barracuda*	Great Barracuda
C2CF001	CTX-2	Cold	0.001227143	15	*Cepahlopholis fulva*	Coney
C2CC001	CTX-2	Cold	0.001280774	12	*Caranx crysos*	Blue Runner
C2CR001	CTX-2	Cold	0.001318837	13	*Caranx ruber*	Bar jack
C2HR009	CTX-2	Cold	0.001497698	15	*Holocentrus rufus*	Squirrelfish
FALJ002	Fajardo	Cold	0.001531083	6	*Lutjanus jocu*	Dog Snapper
NGLM001	Naguabo	Cold	0.001660033	10	*Lachnolaimus maximus*	Hogfish
MASR004	Maunabo		0.0018475	17	*Scomberomorus regalis*	Cero
C2HR007	CTX-2	Cold	0.002178169	15	*Holocentrus rufus*	Squirrelfish
C1AV001	CTX-1	Cold	0.002391701	14	*Anisotremus virginicus*	Porkfish
GYSB002	Guayama	Hot	0.002704755	10	*Sphyraena barracuda*	Great Barracuda
C2LM001	CTX-2	Cold	0.003119362	15	*Lachnolaimus maximus*	Hogfish
MASC002	Maunabo		0.0035815	17	*Scomberomorus cavalla*	King Mackeral
C2HR004	CTX-2	Cold	0.004324338	15	*Holocentrus rufus*	Squirrelfish
C2HR002	CTX-2	Cold	0.0056973	14	*Holocentrus rufus*	Squirrelfish
MASC003	Maunabo		0.0058664	17	*Scomberomorus cavalla*	King Mackeral
C2SV001	CTX-2	Cold	0.006269884	15	*Sparisoma viride*	Stoplight Parrotfish
MASC001	Maunabo		0.0071489	17	*Scomberomorus cavalla*	King Mackeral
C4LM001	CTX-4	Hot	0.009072407	10	*Lachnolaimus maximus*	Hogfish
C4LM002	CTX-4	Hot	0.009072407	10	*Lachnolaimus maximus*	Hogfish
C1CF006	CTX-1	Cold	0.013819703	15	*Cepahlopholis fulva*	Coney
GYCR002	Guayama	Hot	0.017388464	16	*Caranx ruber*	Bar Jack
GYCR001	Guayama	Hot	0.019755444	16	*Caranx ruber*	Bar Jack
FACR001	Fajardo	Cold	0.029821041	15	*Caranx ruber*	Bar Jack
GYSB001	Guayama	Hot	0.031492097	7	*Sphyraena barracuda*	Great Barracuda
C3CR001	CTX-3	Hot	0.0349967	16	*Caranx ruber*	Bar Jack
GYLM002	Guayama	Hot	0.0576127	7	*Lachnolaimus maximus*	Hogfish
GYLM001	Guayama	Hot	0.06136924	7	*Lachnolaimus maximus*	Hogfish
GYSB003	Guayama	Hot	0.072032767	17	*Sphyraena barracuda*	Great Barracuda
C4SB001	CTX-4	Hot	0.077304577	8	*Sphyraena barracuda*	Great Barracuda
GYSC002	Guayama	Hot	0.099918181	10	*Scomberomorus cavalla*	King Mackeral
C3SB001	CTX-3	Hot	0.1151133	15	*Sphyraena barracuda*	Great Barracuda

Appendix H Chapter 5 Supplemental Data

Table H-1 Cell count data from the screen-sampler rigs. Cells were counted and confirmed to be Gambierdiscus spp. by the NOAA Southeast Fisheries Lab in Beaufort, NC (chris.holland@noaa.gov).

		Date	mL				Average	cells/L
Station	**Repeat**	**sampled**	**counted**	**Gambierdiscus cells counted**			**cells/mL**	
					Subsamples			
CTX-1	1	10/16/2019	1	1	1	0	0.66667	666.667
	2	10/16/2019	1	2	0	1	1	1000
	3	10/16/2019	1	0	0	0	0	0
	4	10/16/2019	1	0	1	0	0.33333	333.333
	5	10/16/2019	1	0	0	0	0	0
CTX-2	1	10/16/2019	1	2	0	1	1	1000
	2	10/16/2019	1	2	0	0	0.66667	666.667
	3	10/16/2019	1	1	2	0	1	1000
	4	10/16/2019	1	1	1	0	0.66667	666.667
	5	10/16/2019	1	1	1	1	1	1000
CTX-3	1	10/22/2019	1	0	1	1	0.66667	666.667
	2	10/22/2019	1	2	2	3	2.33333	2333.33
	3	10/22/2019	1	5	4	4	4.33333	4333.33
	4	10/22/2019	1	4	4	3	3.66667	3666.67
	5	10/22/2019	1	2	2	2	2	2000
CTX-4	1	10/22/2019	1	17	15	17	16.3333	16333.3
	2	10/22/2019	1	9	10	9	9.33333	9333.33
	3	10/22/2019	1	15	15	13	14.3333	14333.3
	4	10/22/2019	1	12	10	9	10.3333	10333.3
	5	10/22/2019	1	11	11	13	11.6667	11666.7

Appendix I Chapter 7 Supplemental Data

Completed models and all data can be found online:

Luczkovich, J. J., & Raab, H. (2021, January 10). Ciguatera Ecopath Models for Puerto Rico Sea Grant. Retrieved from osf.io/btyd3

Month	Value
0	1.75
February	2.00
March	1.00
April	1.00
May	1.00
June	2.00
July	1.50
August	2.50
September	3.00
October	4.00
November	3.00
December	3.00

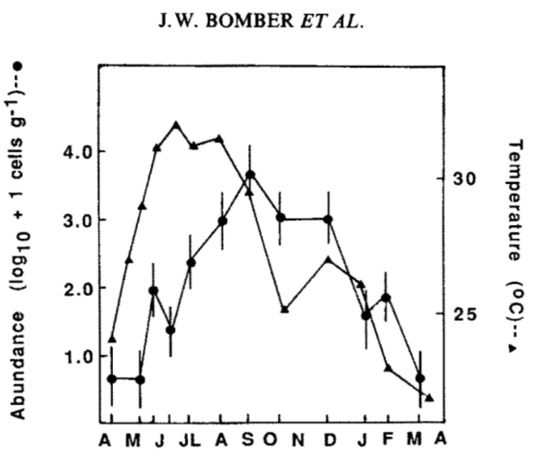


Figure I-1 Data used in the forcing function for the final models along with the original data from Bomber *et al*. (1988).

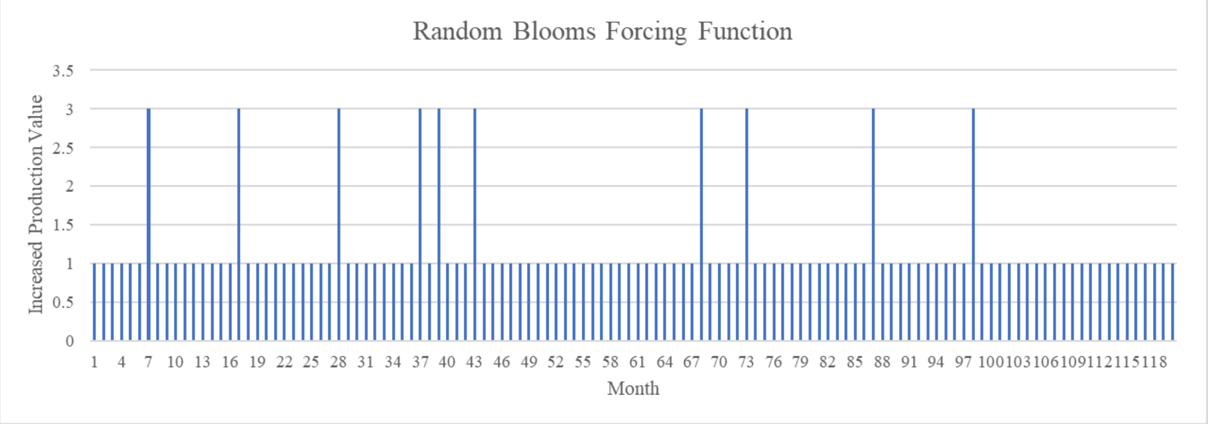


Figure I-2 The random bloom forcing function. Ten random numbers between 1-120 were generated (7,17, 28, 37, 39, 43, 68, 73, 87, 98) to simulate blooms of random months over a 10-year period. The y-axis is the increased amount of production for the *Gambierdiscus* spp. compartment and the x-axis are months. The increased production is multiplicative.

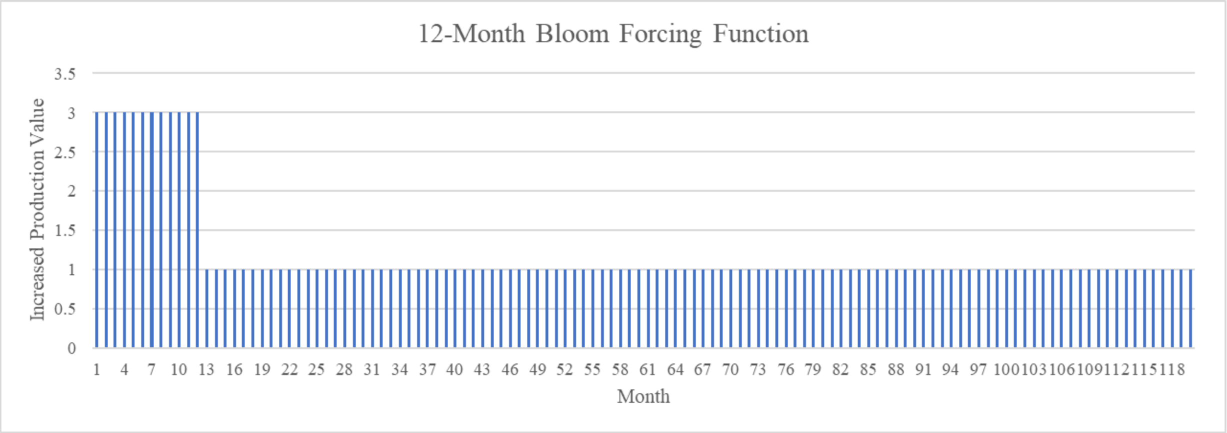


Figure I-3 The 12-month bloom forcing function. This forcing function was used to see how quickly different compartments could reach 0.1 ppb with the worst-case scenario bloom for 12 straight months. The y-axis is the increased amount of production for the *Gambierdiscus* spp. compartment and the x-axis are months. The increased production is multiplicative.