

# The Aquaculture of Sponges--Coral Reef Superheroes--in a Closed System Remi Ventura, Bobbie Renfro

# Abstract

Sponges have numerous responsibilities in coral reef ecosystems that support the survival and function of corals. Sponges provide structural support to reef corals improving their survival and assisting with coral reef regeneration following physical damage. Despite their importance to the maintenance of coral reef ecosystems, sponges are poorly understood in part because they are difficult to maintain in closed aquaculture systems used in many laboratory settings. The research conducted herein will improve our understanding of an integral part of closed-system sponge cultivation: proper food distribution. The effects of two different quantities of food on the growth and survival of the sponge, Axinella *pomponiae*, were tested in this study. Four individual sponges were subdivided to control genotype and half of each individual was used in each treatment group (Recommended food level: 60 mL and Elevated food level: 160 mL). Food treatments were calculated using a ratio derived from Reiswig (1971) who stated that 1 cm^3 of sponge tissue can filter 1 liter of water per hour. Sponges were fed live phytoplankton containing four different species of microalgae. The results of this study can be used as a guide for cultivation of sponges in laboratory settings. Lab cultivation will allow for the generation of sponge populations to be used for

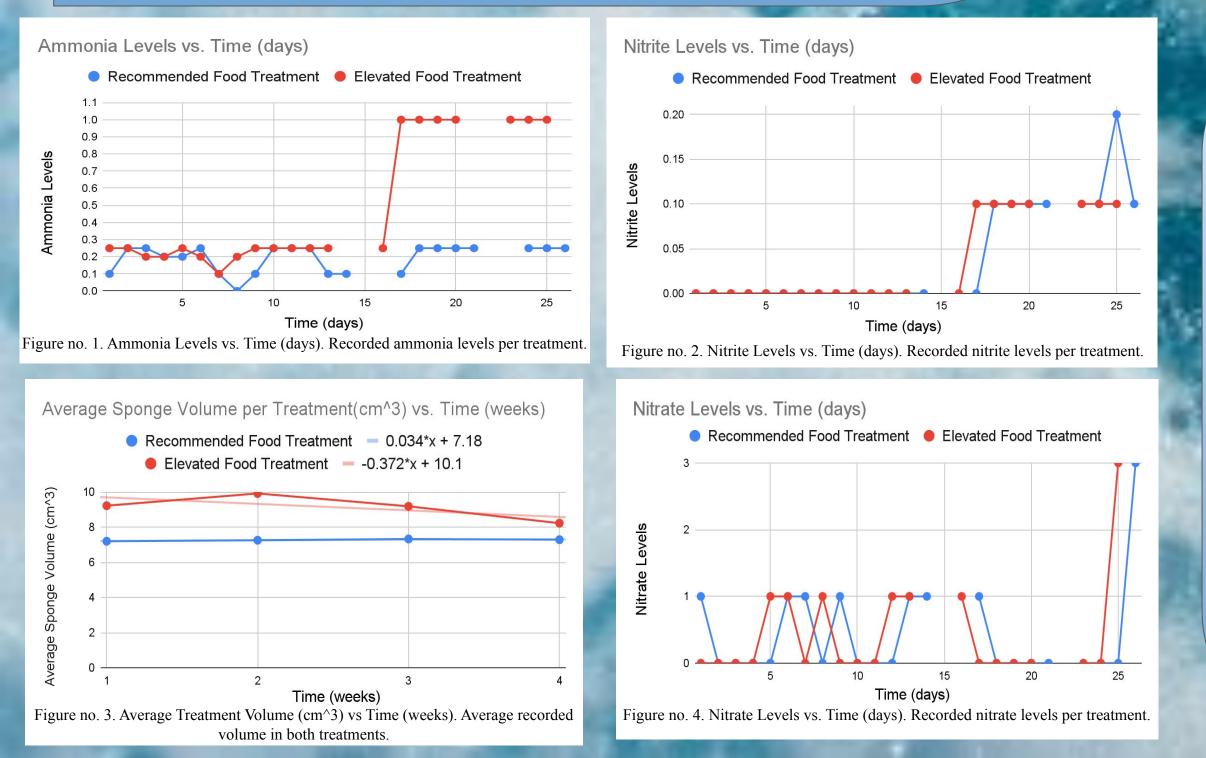
research to better understand these diverse organisms. Additionally, this will allow for the maintenance of stock populations to be used for restoration, preventing the need for fragmentation of natural sponge individuals for restoration activities.

# Introduction

Sponges play critical roles in the survival, restoration, structure, and function of coral reefs (Wulff 2016). Corals lack strong structural support due to their growth originating from a single base point. Sponges provide support by attaching to the coral and filling in vacancies, allowing corals to grow without the threat of detachment from the reef. The addition of sponges to coral reefs increased overall coral survival (Wulff 1984). Corals build the reef structure that supports both marine and human life. Coral reefs serve as coastline protection from storms and forceful currents, support over 25% of the ocean's species, are responsible for food for 500 million people, and have an immense influence on the economy (Brander 2015, Wilkinson 2000)

Researchers have found that sponges are not only able to help maintain existing reef structures, but also are able to assist in coral reef regeneration. Wulff (1984) found that coral recruit survival increases when sponges bind and consolidate reef rubble. It is important that the marine science community determines a way to cultivate sponges within a lab both to further the study of these organisms and to generate more sponges for reef restoration to reduce relying on fragmentation of wild sponges. Despite this need, researchers have found it difficult to sustain sponges within the laboratory in closed aquaculture systems.

Osinga (1999) highlighted the lack of closed-system sponge research and attempted to cultivate the sponge species *Pseudosuberites aff andrewsi* and *Pseudosuberites andrewsi*. He found that both had over 600% growth increases over a few weeks when fed roughly 150 mL of live food weekly (Osinga 1999). His research did not determine what the proper food intake was. Previous research has displayed that sponge starvation may cause low survivability. For example, A. Schmidt of Florida State University attempted to cultivate *Halichondria corrugata* in the laboratory. Throughout her experiment, her sponges survived for 13 weeks, and were fed 60 mL of phytoplankton *Nanochloropsis sp.* for an hour daily. She hypothesized the sponges struggled due to starvation (Schmidt 2016). Osinga's sponges survived for a longer period which may be accredited to the amount of food they received. The purpose of this study is to improve our understanding of one specific part of closed system sponge cultivation: proper food distribution.

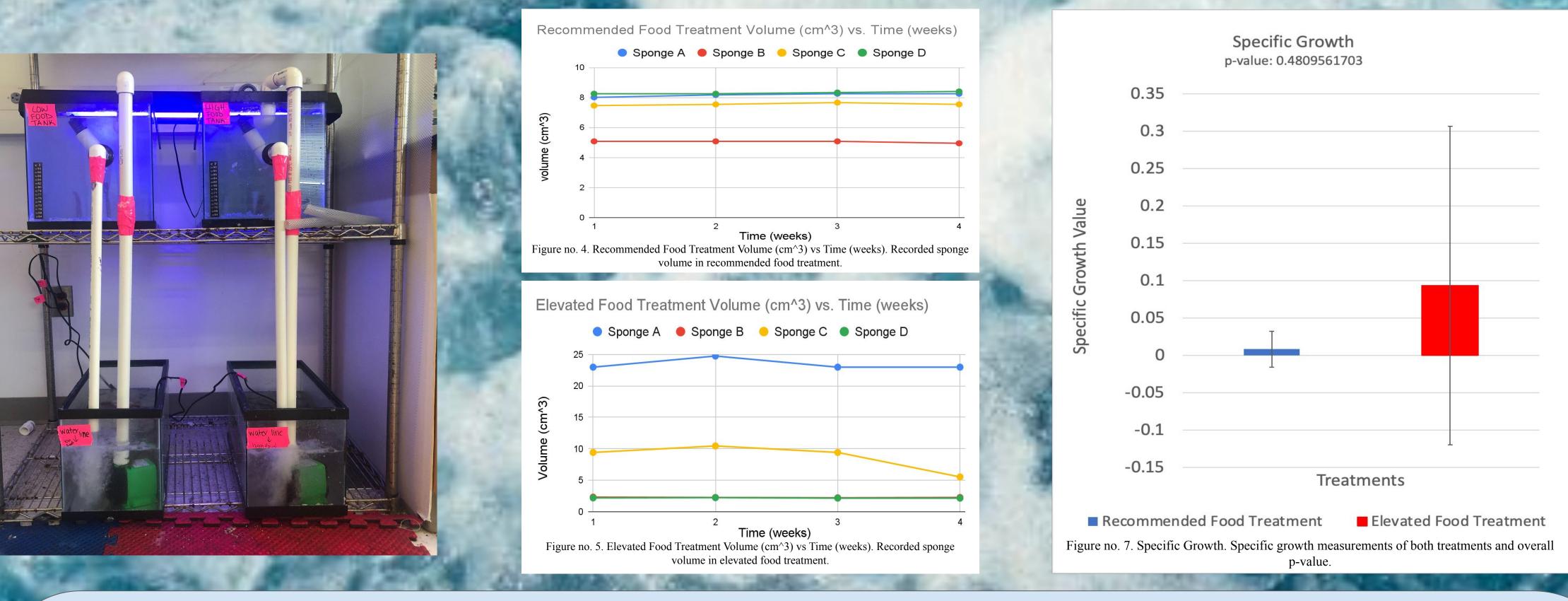




To determine what quantity of live phytoplankton allows for more successful sponge survival and growth, we constructed two 57-liter aquaria each containing four sponges. Below each aquarium is a sump containing live rock and live macroalgae for filtration and maintenance of beneficial bacteria. Each aquarium has a Vivosun water pump with a maximum flow rate of 3000 liters per hour set to its lowest setting-cycling the water about 50 times per hour. An actinic coral grows light is providing natural lighting. To provide some stability for the sponges, each aquarium is equipped with a CPVC pipe stand with six prongs. The sponge fragments were attached to these via thin nylon cable ties commonly used in sponge research.

Sponge fragments for the recommended food group had an average initial volume of 7.2 cm^3, and the elevated food group had an average initial volume of 9.2 cm^3. The water temperature was maintained between 21 to 22 degrees Celsius as per the average temperature of the northern Gulf of Mexico where they were harvested (NCEI). To maintain proper ocean salinity of 35 ppt, we used 60 ounces of salt per 15 liters of water (De Lanza Espino 2004).

The leftmost aquarium received the low amount of food: 60 mL of OceanMagik containing four types of phytoplankton: *Nannochloropsis gaditana*, *Tetraselmis sp*, *Isochrysis galbana*, and *Thalassiosira weissflogii*. The aquarium to the right received 160 mL of the same live food phytoplankton. These mL amounts were determined by using Reiswig's 1971 paper stating that sponges could filter one liter per hour per cubic centimeter of sponge tissue (Reiswig 1971). Each treatment system was 57 liters, and the sponges totaled a volume of 28.8 cm<sup>3</sup> and 36.9 cm<sup>3</sup> (respectively: recommended, elevated). Thus, the recommended food aquarium could filter the entire 57 liters in 1.97 hours and the elevated food aquarium could filter it in 1.5 hours. The OceanMagik recommends 5 mL of live food per aquarium each day—assuming the organisms within the aquarium will filter this in a 24-hour revolution. Since the sponges filter at 12-13 times this speed, this equates to 60 mL and 80 mL—recommended, elevated—per day of live food. This is the biological bare minimum amount of food the sponges need as they filter the water per day. Based on concerns found by other authors of sponge starvation (i.e., A Schmidt of Florida State University in 2016), we are also testing elevated food concentrations to ensure that starvation is not the limiting factor in sponge aquaculture. Therefore, the recommended concentration of food will be 60 mL and the elevated will 160 mL daily. (The higher amount is the aquarium's sponge average's bare minimum food intake multiplied by two.) Each day, the salinity, temperature, nitrate/ammonia/nitrite levels, and pH of the water was tested to ensure it was maintained within the correct levels. Aquariums were cleaned once a week and water were replaced as needed to compensate for evaporation. Once per week, 4 liters of water was removed and replaced.



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<u>Water Quality Parameters that Remained Constant Across Treatments:</u>	Nit	trite Le
▶ pH: 8.4-8.6		For th
► Temperature: 22–23°C		For th
Salinity: 35–36 ppm		
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<u>Specific Growth:</u>		Rema
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<ul> <li>Slight, but consistent growth over the 26-day period</li> </ul>		110 0110
<ul> <li>Average Specific Growth: 0.0081514</li> </ul>	An	nmonia
Elevated Food Treatment:		Recor
• Slight growth between the first and second weeks, then generally		• Le
consistent shrinkage over the remaining weeks		• It • Th
• Average Specific Growth: 0.0932810825.		
➤ T-Test Results: failed to support the hypothesis that specific growth		pr Elevat
would differ between the two treatments of food quantity (p =		

0.48096)

# Results

#### levels:

he beginning of the study, they stayed constant at 0.00 ppm for both treatments the last week of testing, they elevated to about 0.10 ppm in both treatments

#### <u>\_evels:</u>

ained between o-1 ppm for both treatments

e end of the study, elevated to 3 ppm for both treatments

#### ia Levels

ommended Food Treatment:

Levels stayed low for most of the experiment at around 0-0.25 ppm

These are average ammonia levels due to the other environmental factors (food

provided, live rock and algae, etc.).

ated Food:

• Ammonia levels were high from the beginning of the experiment  $\circ$  At 2/3 of the way through the testing, ammonia levels shot up to 1 ppm

Overall, the individual sponges survived successfully for the extent of the experimental period. Salinity was maintained around the average salinity of seawater (35 ppm) but varied between 34-37 ppm each day when initially tested (De Lanza Espino 2004). To counteract the constant, small fluctuations in salinity, water was replaced with either deionized water or prepared salt water. Due to lab room temperatures and open-top aquarium design, evaporation was rapid, and water was replaced daily to maintain salinity and proper flow. **Elevated Treatment Analysis:** 

It was evident that as the experiment ended, ammonia, nitrite, and nitrate levels within the elevated food treatment began to increase. This required daily water changes as a result to lower detrimental nutrient concentrations. The high nutrient concentrations were caused by the decomposition of the excess food accumulating in the elevated food treatment. The water in the aquarium used for this treatment remained consistently green-tinged even after daily water changes of 4 liters and removal of detritus. **Recommended Food Treatment Analysis:** 

The sponges in the recommended food treatment survived and grew for most of the experiment as seen by figure 4. By the fourth week, each sponge's size either plateaued, or began to stop declining. Overall, the ammonia, nitrite, and nitrate levels changed slightly (Figures 1, 2, and 4). However, the nitrite and nitrate levels did increase drastically in the final week of testing after been low around zero ppm for most of the experiment. When the ammonia level was higher, daily aquarium cleaning and four-liter water changes were completed to restore water quality. This may have had a similar effect on these sponges that it had on the elevated food treatment sponges. In contrast to the elevated food treatment, the sponge individuals in this treatment had minor green algal growth. **Relevance to the Field of Sponge Biology:** 

Between the two treatments there was not a significant difference in growth, however the sponges in the recommended treatment remained "healthier" than the other (no tissue shrinkage or algal growth). The Increased ammonia levels can cause stress to organisms and halt their growth (Francis-Floyd 2009). I aquarium. Other research has suggested a break in food distribution when ammonia levels increase to reduce Based on the results of this study, daily water changes are recommended in closed-aquaculture systems for issues in the short term but starting this task early may stop them from occurring overall. It was evident that 160 mL of live food may have been too much for the sponges to handle in the manner that minimal enough to not negatively affect the water quality. In contrast, the Elevated Food Treatment had more

water quality was better in this treatment based on the parameters recorded in the results. From this experiment there are two potential conclusions about the amount of food provided to the Elevated Food Treatment (160 mL) to explain why this aquarium had poor water quality. There may have been too much food provided each day causing the remainder to decompose into waste products polluting the treatment. attempted to counteract the ammonia level issue by completing large, daily water changes in each treatment it (Francis-Floyd 2009). However, as displayed by Schmidt's 2016 study, sponges cannot survive in a food deficit (Schmidt 2016). Therefore, pausing feedings in the elevated treatment was not an option in this study. sponges to prevent any water quality deterioration. These water changes did help to counteract water quality it was distributed. The sponges had a higher filtering capacity than the daily dose of live phytoplankton that OceanMagik recommends. They filter 12-13 times faster than the normal marine organisms that OceanMagik is geared towards. The recommended treatment individuals were able to filter and consume all the food they were provided, given that they did not have significant volume growth. However, the amount of food was overall growth, but poor water quality. Despite active efforts to counteract the water quality in the Elevated Food Treatment, the sponge individuals began to die at the end of the study. It is unclear whether the Elevated

Food Treatment may still promote the same growth with better what quality when food distribution is fewer times per week.

The amounts of food were chosen using the filtration ratio determined by Reiswig in his 1971 paper. He determined that sponges could filter one liter per hour per cubic centimeter of sponge tissue (Reiswig 1971). Each aquarium had total sponge tissue volumes of 28.8 cm<sup>3</sup> and 36.9 cm<sup>3</sup> (respectively: recommended, elevated). Based on filtration speed, sponge tissue volume, and OceanMagik's recommended daily dose, I calculated 60 mL and 80 mL--recommended, elevated—per day of live food. The elevated tank received double what they were able to filter and consume. This was inspired by the fact that A. Schmidt hypothesized that her sponges may have suffered from starvation in her 2016 sponge cultivation study (Schmidt 2016). The closed aquaculture of sponges is incredibly important for the restoration of coral reefs (Wulff 2016). It is essential that in lab cultivation of these sponges it is determined to hasten and continue the regeneration of coral reefs. From this experiment, we can determine that sponges fed substantial amounts may not need to be fed each day as it may overwhelm the individuals and that daily water changes are essential to the maintenance of proper water quality levels. I would recommend the usage of Reiswig's 1971 ratio as when used to determine the proper amount needed to maintain the Recommended Food Treatment, the individuals did well with the amount of food provided. Additionally, increasing the sample size of the experiment would be helpful in providing more overall insight and a potentially more significant p-value.

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# Conclusions

#### Aquarium Care Standards to Note

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