Experimental Uses of Integrated Multi-Trophic Aquaculture



T. Hayden Cronin, Tara Fraprie, Ian Stelzner and Kelsi O'Neil

Department of Biology, University of New Hampshire, Durham, NH, 03824

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Abstract

Integrated Multi-Trophic Aquaculture (IMTA) systems are very important for researchers to expand the large-scale aquaculture industry because it provides valuable biomass and services for waste reduction. These systems show a tremendous increase in sustainability of aquaculture, and can show benefits economically, environmentally and societally. However, aquaculture can cause pollution and disease for single species systems. The idea of IMTA is to eliminate the environmental impacts by integrating several species, each of which can benefit from wastes of others and also be value added for a more diverse group of species so if one crashes, you still have the others to provide a buffer and add more potential for making a profit. The four marine species integrated into the system were Gracilaria spp, Ostrea edulis, Nereis virens, and *Centropristis striata*. The objectives for this experiment were to maintain a closed re-circulating system with the four marine species, track the growth of *Gracilaria spp* and *N. virens*, and to track ammonia uploading versus ammonia uptake. Three re-circulating systems were set up with Tank A in each system containing *Gracilaria spp*, Tank B contained N. virens and three O. edulis. To maintain a sustainable environment the N. virens were fed every third day, and ammonia, salinity and pH levels were tested daily. To determine growth, weekly weigh-ins were successfully monitored to determine that there was indeed growth in the species. In conclusion, the closed re-circulating system was successful to allow these species to maintain a sustainable life in optimal water quality conditions. Further research can be done to ensure that N. virens will consume C. striata fecal matter, to eliminate the use of TetraMin (the food used to feed N. *virens*). The *Gracilaria spp* can take in the ammonia produced by *N. virens* to allow for a sustainable system. IMTA is beneficial for producers because it adds value to each of the products in the system.

Introduction

Aquaculture is the farming of marine and freshwater species. (Ridler, 2007) In this project four marine species were integrated in the hopes that each species would benefit off the others survival. Growth was tracked through the project to ensure that the species involved were continuously growing and sustaining each other until they reached market size. (Pillay, 1990) Each species chosen has an economic value, but also a biological value to this experiment. Integrated Multi-Trophic Aquaculture (IMTA) is a revolutionary approach to aquaculture; introduced to solve the problem of nutrient loading beneath pen cultures. (Ridler, 2007) This new approach to farming is beneficial ecologically and economically for single species processes. In our study, multi species tests were conducted in order to test the possibility of combining aquaculture. Having an integrated system reduces the amount of waste due to the organisms chosen: *C. striata*, black sea bass, excrete wastes which then in turn are consumed by *N. virens*, bait worms, *and* O. *edulis*, European oysters; while this is occurring all three of these species excrete ammonia which is consumed by *Gracilaria spp*. further regulating the systems.

Nereis virens are from the family Nereidae. *N. virens* are a marine species that live in shallow shorelines. Typically, *N. virens* grow between 20-40 cm but can reach up to a meter long. Significant structures on *N. virens* body include parapods, eyes, antenna, mouth and pygidium. Parapods are leg-like strictures used to aid mobility. (Hamaker, 2010) The pygidiums are structures on the end of the *N. virens* that tend to be the first damaged when they fight. In our experiment, the pygidium were observed and noted as to whether or not they were present. If they are absent it is an indicator that the worms are fighting. (Nielsen, 1995) *N. virens* are used commonly by fisherman as bait worms.

Gracilaria spp. is from the phylum Phodophyta. *Graacilaria spp.* is commonly referred to as red algae. It is a marine species found around reef areas or intertidal zones. The species grows 1-2 mm in diameter with a vast amount of branching. The coloring is typically reddishbrown. (Raikar, 2001) Gracilaria spp. is widely used as agar, which is a medium for culturing bacteria. (Ria, 2013)

Ostrea edulis is from the phylum Mollusca, referred to as European oysters. At maturity the oysters are typically 3.8-11 cm across with an oval shape and a rough shell. (Enright, 1986) The halves of the oyster are made of two shells on a hinge. *O. edulis* are hermaphrodites, changing sexes multiple times a year and can live for greater than 20 years. (Enright, 1986)

Centropristis striata is from the phylum Cordata. *C. striata* is a marine fish, referred to as black sea bass, found in shallow and offshore waters. (Hood, 1994) A majority of their time is spent near the sea floor around reefs or rocks for protection. At maturity, *C. striata* is characteristically around 19 cm but can reach up to 50 cm. (Hood, 1994) This species is hermaphroditic, beginning their lives as female and some changing to male. Both *C. Striata* and *O. edulis* are used as food sources.

In initial tests conducted, *U. lactuca* were used in the place of *Graacilaria spp. U. lactuca* is commonly referred to as sea lettuce because of its weak, thin, ruffled shape. The typical size is 18 cm in length and 30 cm in width. (Pizzolla, 2003) Layers of two cells make up its thickness, which is responsible for its weak makeup. The main population grows attached to rocks on the coastline.

Methods

Species Coexistence:

Our initial testing consisted of a set-up of three tanks. The first tank contained 30 *N*. *virens* with a displacement of 60 mL. The second tank contained 30 *N*. *virens* with a displacement of 30 mL and 17.4 grams of *U*. *lactuca*. The third tank contained 17.1 grams of *U*. *lactuca*. A light was set above the tanks and set on a timer. The *U*. *lactuca* were held under the surface of the water in wire baskets to ensure equal light was administered to all plants equally. These tanks all were observed as different treatments under the same environmental conditions, e.g. temperature, salinity, pH, light exposure and intensity. These conditions included optimal ranges of pH for growth, 8-8.5; a 12 hour photoperiod; a salinity of 32-35 ppt; and a temperature of 20 °C. These tanks were held at room temperature (20-24°C), to see the species growth limitations.

Each day the *N. virens* were fed 1/8 of a teaspoon of TetraMin. Revisions for our second experiment were made based on the results of this first trial. The recorded weights of *N. virens* and the end of weight of *U. lactuca* would draw a correlation that could be used in future trials. This number would be averaged from all 3 series and that calculation would be used to offset future influxes.

Revised Species Coexistence Series:

Due to the lack of ammonia consumption by *U. lactuca; Gracilaria spp.* was introduced to the same system listed above, under the same conditions. *Gracilaria spp.* was added to the system because it is known as a hardier intertidal species that also has an economic value. With this change the systems were reset to the same conditions listed above. Once the ammonia levels were sustained in all three replicates the total amount of *Gracilaria spp.* and the total amount of *N. virens* in each tank was weighed. From these numbers a ration of *N. virens* to *Gracilaria spp.* could be drawn to implement in future trials. The average was taken of all of the ratios to produce one final average that would be used on all of the future treatments.

O. edulis Consumption Rates:

European Oysters, *O. edulis*, were added into the systems in late November. The purpose of adding oysters was to remove the particulate from the water that the *N. virens* did not consume. To ensure feed of the oysters we added Phytoplex, an algal supplement, to a 1 gallon fish tank. 1 organism was placed in this tank and water samples were taken throughout the day and frozen for further examination. The samples were taken: immediately after *O. edulis* was added; after: 15 mins, 30mins, 1 hour, 2 hours, 4 hours, 8 hours, 12 hours, and lastly at 24 hours. This data would give us a daily feeding rate of the *O. edulis*. During this day experiment *O. edulis* were exposed to the same conditions as the series above. This test was run in triplicate.

After the Oysters were tested for feeding rates; 1 organism was added to each of the series explained above and ammonia testing was conducted to ensure there was no severe influx of ammonia loading. If a spike of ammonia was recorded *Gracilaria spp*. was added in 10 ml displacements to offset the influx.

Ammonia Testing and Water Quailty:

Three tanks were set up as a series to test the amount of ammonia given off by *N.virens* and taken up by *Gracilaria spp*. One of the tanks, the negative control, contained: sea water

(filtered through a 50 µm mesh), 2 liters of sand, a power filter without a filter pad (for aeration), a tank cover made of transparent cowhide, and a light source suspended 6 inches above the series. This tank was used to track the normal changes that could be expected in salinity, temperature, ammonia concentration, etc. The second tank of the series was that positive control; this tank contained all of the above parameters along with the addition of 40 *N.virens*. The 40 *N.virens* were weighed before added to the system so that a ratio between grams of worms and ammonia output could be derived. The last tank was the experimental tank, this tank included all of the factors from the previous two series however this tank also contained Gracilaria spp. We would use this tank to test the amount of ammonia *Gracilaria spp.* could consume before it reached to high of a level. To do this we started out with 10 grams of *Gracilaria spp.* added to the tank and increase the amount of *Gracilaria spp*. by 10 grams every time the tanks ammonia concentration exceeded 40 µmolar. The goal of this final tank was to establish the amount of Gracilaria spp. needed to offset the amount of ammonia that the N.virens gave off. This experiment was done in triplicate and the tanks were randomly placed underneath the light source to eliminate the chance optimal light conditions. Water samples were taken daily for the duration of the experiment and frozen for further examination through nutrient assays. The tanks containing *N.virens* were fed .5 grams of Tetramin every third day. On the feeding day the ammonia levels were tested using and amount of Gracilaria spp. was adjusted.

Final Recirculating Systems:

Three recirculating systems were set up in a temperature controlled environment. Light was added to the systems on a 12 hour period, 7am-7pm. Each system was comprised of two ten gallon tanks, two tank dividers, and one 90gph pump. Tank A of each system, the upper tank,

contained *Gracilaria spp*. This purpose of this tank was to remove the dissolved nitrogenous waste, in the form of ammonia, which are nutrients for the *Gracilaria spp*. Tank B, the lower tank contained: *N. virens* (20 counted and pre-weighed), 2 liters of sand, *O. Edulis* (3 counted measured and weighed) and an air stone for aeration. Two sources of light were hung 6 inches above Tank A, 12 inches above Tank B. Tank B was fed every third day, 0.75 grams, and the organisms were weighed once weekly. The ammonia levels were tested daily along with the salinity and pH.



Figure 1: this image represents the final recirculating system which was conducted to test overall sustainability of our three species.

Results:

Semester 1: At the start of the year the first experiment was just three tanks. One tank had nothing in it as the control, the second tank had just worms in it, and the third tank had both worms and *Ulva lactuca*. The experiment was to test the feasibility of this experiment on a small scale. The goal was to end with a system like the last experiment ran with three individual systems. Each tank had forty worms and the third tank had about a handful of Ulva lactuca (starting weight unknown). The ammonia levels were taken but were not written down. The ammonia levels were measured using parts per million. A general knowledge of the ammonia levels during this time were noted. During the first week of the experiment the ammonia levels stayed between 0 ppm and 0.5 ppm. However, after the first week the ammonia levels steadily increased to about 1 ppm in both tanks with the worms. The Ulva lactuca started to die at this point and the worms started to die as well. The Ulva lactuca was turning a clear color and the worms were found floating in the current of the tanks. This could have been a result of overcrowding and ammonia build-up. The Ulva lactuca was replaced during the start of the third week, but it died soon after. About every week the ammonia levels were lowered with water changes. The worms were also not eating all of the food, so it would get collected by the current, deposited in the corner of the tank, and rot.

The next experiment ran was the same set-up as the first, but on a bigger scale. This experiment replicated the first one by three. The results from that experiment were the same as the first. The *Ulva lactuca* started to die and the ammonia levels rose to over 1 ppm in all three systems after a week. The worms also started to die in all of the systems. The water quality was very poor because the worms were not eating all the food and it was rotting on the sand in all

three systems. That experiment lasted about two weeks, because most of the worms were dead and the ammonia levels did not decrease.

Winter Break: The third experiment was different from the others because of the air wands and the replacement of *Gracilaria spp*. for *Ulva lactuca*, but the results were the same until we started to add more *Gracilaria spp*. The ammonia levels started to rise with each day until more *Gracilaria spp*. was added to each system. The *Gracilaria spp*. was added after the first week of experiments when the ammonia levels were above 1 ppm in every system. The middle tanks without *Gracilaria spp*. rose to over 1 ppm and were then lowered by a water change. The water quality in all the systems was increased because of the feed rate change. The rate of *Gracilaria spp*. to worm density was calculated at 1 gram of *Gracilaria spp*. to 3.28 grams of worms during this experiment. However, the numerical data for that calculation is unavailable.

Semester 2: The results of this experiment are based on three graphs that show the growth rates of both the *Gracilaria spp*. and *N. virens*, and the ammonia levels in all three systems. Figure 2 shows the average growth rate of the *N. virens*. Twenty worms were placed in all three systems and were massed at the start of the trial and then each week after. System 1 started at an average of 1.70 grams, system 2 started at an average of 1.83 grams, and system 3 started at an average of 1.62 grams. Systems 1 and 3 decreased after the first week but then steady rose to a final weight of 1.90 grams and 2.06 grams respectively. System 2 increased throughout the whole study and finished at 2.09 grams. Nine worms were assumed dead through the trial. The adjusted weight was calculated by the mass of all the body sections.

Figure 3 shows the ammonia levels throughout the nineteen day trial. The ammonia levels start at 0 ppm (parts per million) at the beginning and then steadily rise to 0.2 ppm. Throughout the trial the ammonia levels stayed consistently between 0.2 and 0.6. Systems 1 and 3 both rose to 1.0 ppm but only for 2 days or less.

Figure 4 shows the growth of G*racilaria spp*. in grams over the four week time period. The amount of *Gracilaria spp*. was determined by a previous experiment. There was 1 gram of *Gracilaria spp*. added to every 3.28 grams of total worm weight. Based on those numbers, system 1 had a total of 33.90 grams in worms and 10.34 grams in *Grascilaria spp*. System 2 had 36.60 grams in worms and 11.16 grams in *Grascilaria spp*. System 3 had 32.33 grams in worms and 9.86 grams in *Gracilaria spp*. They all showed a positive increase in growth. System 1 ended at 35.34 grams, system 2 ended at 45.66 grams, and system 3 ended at 51.08 grams.



Figure 2: The growth rate of *N. virens* over the four week time period. System 1 is colored in blue, system 2 is colored in orange, and system 3 is colored in green. Each marker represents the average weight recorded in that system for the given week. The average weight is measured in grams and is the wet weight of the worms.



Figure 3: The amount of ammonia resent in each system during the nineteen day time period. System 1 is colored in blue, system 2 is colored in orange, and system 3 is colored in green. Each marker represents the amount of ammonia recorded in that system for the given day. The ammonia levels were measured in parts per million (ppm).



Figure 4: The growth rate of the *Graciliaria spp*. in each system during the four week time period. System 1 is colored in blue, system 2 is colored in orange, and system 3 is colored in green. Each marker represents the weight recorded in that system for that given week. The weight was measured in grams and was the wet weight of the *Gracilaria spp*.

Figure 2 showed the average wet weight in grams of the *N. virens* over the four week time period. Each system started with twenty worms, which were weighed at the start and every week after that for four weeks. An overall picture can be said about the graph that the average weight of all three systems increased over the time period. A general observation can be assumed that the worms therefore were striving and in their level of tolerance range. During the experiment phase a few complications arose from the system set-up. When the worms were taken from their tanks and weighed; only about 17 were collected in each tank. This meant that the other three were assumed dead and dismembered, which usually happens when there is overcrowding. The adjusted weight was measured by removing the dead and injured worms and weighing them.

For system 1, the average weight of the twenty worms started at 1.70 g and ended at 1.94 g. The average weight of the twenty worms in system 2 was 1.83 g and increased to 2.08 g. The average weight of the twenty worms in system 3 was 1.62 g and increased to 2.06. The graph also shows that systems 1 and 3 decreased in total average weight after the first week. The worms acclimating to the new systems and in System 3 there were three worms missing could have caused this. System 1 had two worms with only have a body. System 2 showed an increase in weight even though it also had three worms missing. An explanation for this could be that most of the worms in system 2 were fully intact, and that some worms were found behind the screen that is supposed to separate the worms from the pump and therefore been protected from cannibalism.



Figure 5: this image depicts O. edulis feeding in the final recirculating system.

After week 2 the worms started to increase in weight and amount of worms absent in the systems combined increased by three (6 to 9). The average weight in the systems increased by 0.15 g in system 1, 0.02 g in system 2, and 0.15 g in system 3. These numbers show the greatest growth in systems 1 and 3. A reason why there is so much growth is because the worms took a week or less to get adjusted to the tank and environment. Their environmental conditions were within their level of tolerance, which provided optimum growth conditions. At the conclusion of the four weeks the total average weights were at they're highest. System 1 total was at 1.94, system 2 was at 2.08, and system 3 was at 2.06. System 1 could be lower do to the fact that the *Gracilaria spp*. levels in system 1 were much lower than the other two systems. This probably caused the water conditions to suffer, which caused the worm growth to decrease. The *Gracilaria spp*. level in system 1 was lower than the other systems because the *Gracilaria spp*. tank cracked down the middle. The *Gracilaria spp*. was then put in the worm tank behind the screen, but the *Gracilaria spp*. was stationary and not rotating around in the tank like the other two systems.

Figure 3 shows the amount of ammonia present in each system on each day between March 21 and April 9th. The ammonia levels throughout the nineteen-day trial with varied a lot. They all started at 0 ppm and then rose to 0.25 ppm within the first day. The ammonia levels stayed within the 0.2-0.6 range. Two days within the trial system 1 went as high as 1.0 ppm. That could be because the tank broke leaving the *Gracilaria spp*. in the bottom of an empty tank for about a day. Then it was placed in the worm tank where the growth conditions were not favorable. Since the conditions in the tank were not favorable and the fact the *Gracilaria spp*. was left out of water for a day, the *Gracilaria spp*. was not growing and converting the ammonia to nitrate. For two consecutive days system 2 ammonia levels were up to 1 ppm, which cannot be explained. The ammonia levels of all three systems went down in the last four days. In figure 4, the *Gracilaria spp*. levels spiked in the last week of the *Gracilaria spp*. four-week study, which was also during the last four days of the ammonia testing. That spike can explain the lowered ammonia levels because the high amount of *Gracilaria spp*. would need more ammonia than before.

The four-week study of the *Gracilaria spp*. growth is shown in Figure 4. The amount of *Gracilaria spp*. needed at the start of each system was found in a previous study at the beginning of the year. There should be 1 gram of *Gracilaria spp*. to every 3.28 grams of total worm weight in the tank. Based on those numbers, system 1 had a total of 33.90 grams in worms and 10.34 grams in *Grascilaria spp*. System 2 had 36.60 grams in worms and 11.16 grams in *Grascilaria spp*. System 3 had 32.33 grams in worms and 9.86 grams in *Gracilaria spp*. The *Gracilaria spp*. showed a positive growth curve throughout the whole four-week trial and had exponential growth between the third and fourth weeks. System 1 did not have as much growth because it was left out of water when the tank broke and then it was placed in poor water conditions in the

worm tank. The poor water conditions were that the *Gracilaria spp*. was not rotating in the tank. In order to grow the *Gracilaria spp*. needs to be kept moving, which did not happen when it was placed in the worm tank.



Figure 6: this image depicts the *Gracilaria spp*. tumbling in the top tier of the recirculating system. The bubble wand, on the right side of the tank, creates a clockwise current in which the *Gracilaria spp*. tumbles in ensuring equal light exposer to all parts of the plant.

Discussion

The first system of three tanks failed because the *Ulva spp*. was not moving around (Friedlander, 1995) and the temperature was too high at 15-20°C instead of 10-15°C (Fortes, 1980; Vermaat, 1987). The *Ulva spp*. was not in good water quality due to the high nitrogen content and the rotting worm food. The worms were not eating because of the stress of moving into the system and also the temperature change. The worms were previously kept in the 10°C

lab room and then placed in the systems at a temperature of around 20°C. The feeding rate of the worms was 0.25 grams of fish food every other day. That was too much especially during the time that the worms were not eating. Another reason for the lack of eating could be due to the photoperiods. The worms are mostly nocturnal and swim in the water column during the night. In our system the lights would come on during the normal nighttime hours and turn off during the day. Therefore, the worms were in constant light because the room where the systems were set-up was also a classroom where the lights were on all day long. The food then started to rot on the bottom decreasing the water quality. The *Ulva spp.* started to die and not convert the ammonia into nitrate. The amount of worms was great amount more in the starting systems at around 40 worms/tank, than the ending experiment at around 20 worms/tank. The system was a failure to start, but the results were not statistically viable. Therefore, the first three-tank system was replicated three times to provide statistically significant data.

The second experiment consisted of three true replicates of the first experiment. The exact same thing happened during the second experiment as in the first experiment. The worms started to die and the algae started to die as well. The temperature of the room was in the preferential range of the *Nereis spp.*, which is between 11-20°C (Kristensen, 1983). The decision was made to switch the algae to *Gracilaria spp.* because of its temperature range of 15-20°C (Friedlander, 1995).

The third experiment composed of the three replicated systems mentioned above, but with *Gracilaria spp*. This provided to be a good replacement. In the cultivation of unanchored algae, the tank or system needs to have supplemental oxygen flow to make the algae flow in the water channel (Frielander, 1995). Another reason that the algae died in the first two systems was because the algae were unanchored and freestanding. The algae need to be tumbling and rotating

in the water column in order to grow. In order to fulfill this, bubble-wands were purchased to create a horizontal channel that enabled the algae to tumble around itself. This created optimum growth conditions. The ammonia testing of the three tanks provided evidence of the ammonia levels increasing. It was understood that the *Gracilaria spp*. converts the ammonia to nitrates and based on this knowledge more *Gracilaria spp*. was added to each system as the ammonia levels increased. This third experiment was performed in the 20°C room. The goal amount of ammonia was between 0.2 and 1.0 because it is known that there is a strong positive correlation between the nitrogen levels and *Gracilaria spp*. growth. (Glenn, 1999). It was found that the amount in grams of *Gracilaria spp*. to amount in grams of *Nereis spp*. was 1 gram of *Gracilaria spp*. to every 3.28 grams of *Nereis spp*.

The last experiment was the whole reason for this yearlong project. The goal was to create a successful IMTA system. Given the knowledge from the previous experiments, the whole systems were moved into the 15°C room and the bubble-wands were in the algae tanks. The changes to the systems were made because of the concerns for the water quality, access to both worms and algae, and the small-scale commercial set-up. The final experiment had 3 systems with 2 tanks in each system. The water in the bottom tank with the 20 worms was pumped up into the top tank that contained the algae, and then gravity-fed back into the bottom tank. This was a much better system because of the way it was set-up. It was easier to access the worms since they were in a tank of their own without the algae floating on top. This yearlong project was also performed to see if it was possible to apply this to a commercial scale. The set-up of the final fourth experiment would be about the same way that a commercial business would do. The algae and worms would be in two different tanks/sites and the water would be flowing through them (MacDonald, 2011). This set-up was actually very beneficial because during the

final week of the experiment one of the algae tanks cracked in the middle and all the water drained out. Since the two tanks were separated the worms were unaffected in the short term time span.

The results of the final experiment helped prove that the IMTA system was successful and can be applied to a commercial business. Figure 2 shows the positive growth curve of the worms. Systems 2 and 3 were about equal and figure 1 was a little below the two. Figure 1 was lower because of the cracked algae tank. The other systems flourished, because the ammonia levels were low enough to sustain life and the temperature was in the tolerance range. The ammonia levels in figure 3 were conclusive that 3.28 grams of worms to every 1 gram of *Gracilaria spp.* would keep the ammonia levels low. The ammonia levels were kept in the range of 0.2 and 1.0 only to increase because of the broken tank and odd spikes in system 3. The spikes in system 3 could be from water loss and low water level for a span of one day. The water levels of the other systems and system 3 were maintained throughout the experiment.

The *Gracilaria spp*. showed exponential growth during the third and fourth weeks. This could have been due to the decreased amount of stressors in the systems like ammonia build-up, low oxygen levels, and photoperiods.

Conclusion

In conclusion, the purpose of this experiment was to maintain a closed aquaculture system with *Gracilaria spp*, *O. edulis*, *N. virens*, and *C. striata*. This system was successful in creating a small-scale ecosystem by monitoring ammonia loading and uptake. In our first semester experiments we had many errors with trying to maintain sufficient levels of ammonia to

allow for growth in the species present in the tanks. *U. lactuca* was proven to not be successful as a species in the experiment due to its inability to survive temperatures of 15-20 degrees Celsius. The *U. lactuca* did not show successful growth rates due to the use of a basket system to hold the *U. lactuca*. Further research and remodeling occurred to design a multi-tier system to allow for a re-circulating system with different algae that would be successful in consuming ammonia levels produced by the *N. virens*.

The multi-tier system allowed for efficient data obtained by this re-circulating system that lead to the success of the experiment. The major alteration made to the multi-tier system was a species change from *U. lactuca* to *Gracilaria spp*. Adjustments were made to determine which conditions *Gracilaria spp*. survived. *Gracilaria spp*. didn't grow in tanks with sand because the sand blocked the current flow. We were able to successfully determine the *Gracilaria spp*. grew better in a flow through tank in a moving system, rather than contained in a basket sitting at the top of a tank. There was a change in the amount of *N. virens* to decrease the amount of *overcrowding in the system*. Due to the stock density being too high for the amount of *N. virens* were able to survive in the tank with fewer worms in the system. This system was successful due to the amount of worms offset by *Gracilaria spp*. to maintain ammonia levels suitable for the species to survive. The *O. edulis* and *N. virens* produced sufficient amounts of ammonia that were absorbed by the *Gracilaria spp*. to maintain an efficient re-circulating system.

Implementing an integrated aquaculture system reduces the amount of waste build-up allowing for greater productivity and sustainable growth rates for the species. Aquaculture systems are beneficial to enhancing large-scale industries of marine and freshwater species.

An experiment in the future could be conducted to test the *N. virens* intake of fecal matter excreted by *C. striata*. This could provide proof that the integration of the two species could be successful in sustaining a viable habitat.

Incorporating *C. striata* into the system will provide evidence of the beneficial idea of IMTA. The *N. virens* will consume the waste products of the *C. striata*, reducing the environmental hazards formed from a build up of fecal matter. Providing a cleaner system creates an environment that is sustainable for the *C. striata*. The idea of IMTA is to have a sustainable system that reduces pollutions and has a buffer for failed organisms. The hazards of single species aquaculture would be reduced.

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