Density dependent effects of *Austrovenus Stutchburyi* and Ulva treatments on species richness

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**Abstract**

This study was done to determine the effect *A.stutchburyi* has on nutrients fluxes and species richness as *A. stutchburyi* is considered a keystone species in the estuarine environments it inhabits. We determined if the presence of *A. stutchburyi* increases species richness by altering nutrient fluxes. Several plots of four different treatment types (ulva and *A.stutchburyi,* ulva without *A.stutchburyi*, *A.stutchburyi* without ulva, and neither) were set up in an estuarine area of Blueskin Bay. Measurements of oxygen flux, ammonia concentration, and phosphate concentration were taken at the time of set-up and two weeks after. Macrofauna were also sampled at each plot at set-up and after two weeks. Our results indicated no correlation between treatment type and species richness. We concluded that the presence of *A. stutchburyi* had no effect on species richness.

Keywords: *Austrovenus stutchburyi,* cockles, Density Dependent Effects, MPB, Nutrient Flux, Ulva

**Introduction**

Marine estuaries, or estuarine ecosystems, provide benefits ranging from food production and recreation opportunities to contaminant processing. Estuarine ecosystems have important ecological functions such as influencing the nature and rate of biogeochemical processes, processing land contaminants, and fueling productivity on adjacent coasts. Estuarine ecosystems also provide habitats and nurseries for marine organisms. This being especially relevant in a country such as New Zealand with many estuarine ecosystems that provide food and recreation strongly tied to cultural identity (Thrush et al. 2013).

High biodiversity in an ecosystem has been shown to increase resilience, the ability to resist damage and recover quickly, and resistance, to remain unchanged when disturbed, of the that ecosystem (Oliver et al. 2015), (Stachowicz et al. 2002). High biodiversity can also increase nutrient cycling, which can prevent high concentrations of nutrients from producing harmful effects such as algal blooms (Snelgrove 1998). One of the main components of biological diversity is species richness.

*A.stutchburyi* is endemic to the estuarine environments in New Zealand. They are commonly known as Cockles, although are not true Cockles, which are of genus *Cardiidae*. Cockles grow to about 40 mm in size. They are an infaunal suspension feeder. Cockles live in intertidal zones burrowing 2 - 3 centimeters below the surface on the mid to low intertidal shore (Sandwell 2013). The intertidal zones are marked by features such as increased dissolved O2 and salinity, increased temperature, higher exposure to UV radiation, as well as higher exposure to predation by terrestrial animals (Smith 2013). Cockles are often a keystone species in the intertidal zones they inhabit (Jones 2011). Cockles as filter feeders can easily bio accumulate the toxins and bacteria found in their environment. This allows cockles to be used as environmental indicators, gauging the health of an ecosystem (Fukunaga et al 2011). Cockles link primary producers to primary and secondary consumers on the food web acting as a large source of food for animals such as birds, crabs, and rock lobsters. Cockles also help reduce phytoplankton levels which can lower the chances of harmful blooms occurring in the ecosystem (Sandwell 2013). Cockles are also a popular food item in New Zealand and are often harvested by hand by New Zealanders (Cook 1999).

The aim of this study was to determine the effect Cockles had on species richness through their manipulation of nutrient fluxes by respiration, feeding, and bioturbation. We hypothesized that the presence of cockles would decrease species richness by reducing oxygen availability (through consumption), and increasing efflux of ammonia and phosphate. This would theoretically reduce the oxygen availability for other organisms and would also reduce the amount of nitrogen and phosphate primary consumers can use for vital life processes (Strong 2015).

**Methods**

We are performing this experiment to test the effect *A. stutchburyi* has on ecosystem functioning in an intertidal estuary. The intertidal estuary being studied is Blueskin Bay in the Waitaiti region of the southeastern part of the South Island of New Zealand. The study consisted of seven blocks of four treatments, each placed along the coast of the intertidal zone. The four treatments were color coded as black, yellow, blue, and green using colored markers. The black treatment consisted of no Cockles and no ulva. The yellow treatment consisted of ulva and no Cockles. The blue treatment consisted of Cockles and no ulva. The green treatment consisted of Cockles and Ulva. The plots were prepared on the first visit. Each treatment with Cockles received 300 Cockles and each treatment with Ulva received 100 grams of dead Ulva. Each plot (treatment) was 1m x1m with a 50-cm buffer zone between each. The plot area was finger plowed, the Cockles present removed and weighed, and a core of the buffer zone taken. Then each plot was prepared according the proper treatment. Core samples were taken and the benthic organisms filtered out. Core samples were analyzed by groups in a laboratory and the benthic organisms found were recorded.

Light and dark chambers were also set up to determine the effects of light on nutrient fluxes in each treatment. The methods outlined in Sandwell et al. 2009 were followed with the exception that the light and dark chambers were clear and black bowls respectively and the time between the beginning and end of incubation was over one tidal cycle (a few hours). Nutrient analysis was carried out at the Portobello Marine Laboratory.

Statistical analysis on species richness, treatment type, nutrient flux, and oxygen flux was done using the Shapiro-Wilk test, the Levene Test, and ANOVA analysis. For each treatment, nutrient concentration change and change in species richness were measured by taking the mean of all initial measurements and then the mean of all final measurements to create an average initial measurement and an average final measurement for each treatment. These measurements were then graphed.

**Results**

The change in nutrient concentrations and species richness were measured when the plots were first set up and then again two weeks later. This provided initial and final data for oxygen concentration, phosphate concentration, and ammonia concentration, as well as data on any change in species richness.

The change in oxygen concentration between initial and final readings varied between treatments. The black treatment showed a 1.35 mg/L decrease between the initial and final measurements. The blue treatment showed a 0.08 mg/L decrease between the initial and final measurements. The yellow treatment showed a 1.41 mg/L decrease between the initial and final measurements, and the green treatment showed a 0.607 mg/L decrease between the initial and final measurements (Figure 1).

The change in phosphate concentration also varied between treatments, and unlike oxygen showed an increase over time. The black treatment showed a 1.08 µg. L-1 increase between the initial and final measurements. The blue treatment showed a 1.82 µg. L-1 increase between the initial and final measurements. The yellow treatment showed a 9.79 µg. L-1 increase between the initial and final measurements, and the green treatment showed a 12.59 µg. L-1 increase between the initial and final measurements (Figure 2).

The change in ammonia concentration showed a large difference between initial and final measurements across all treatments, and unlike oxygen showed huge increases over time. The black treatment showed a 50.87 µg. L-1 decrease between the initial and final measurements. The blue treatment showed a 75.27 µg. L-1 decrease between the initial and final measurements. The yellow treatment showed a 106.7 µg. L-1 decrease between the initial and final measurements, and the green treatment showed a 143.8 µg. L-1 decrease between the initial and final measurements (Figure 3).

The changes in species richness varied across treatments between positive and negative changes. The black treatment showed a 0.286 increase between the initial and final measurements. The blue treatment showed a 0.232 decrease between the initial and final measurements. The yellow treatment showed 0 change between the initial and final measurements, and the green treatment showed a 1.054 increase between the initial and final measurements (Figure 4).

There was no significant difference between treatments in terms of species richness (F = 5.45, p > 0.05). The residual standard error was 0.395 on 4 degrees of freedom. ANOVA analysis of oxygen concentration and treatment type showed no significance (F = 0.637, p > 0.05). The residual standard error was 0.721 on 4 degrees of freedom. This was true for the ANOVA analysis of phosphate (F = 0.679, p > 0.05) and ammonia (F = 0.157, p > 0.05) as well. The residual standard error was 5.67 and 71.0 both on 4 degrees of freedom respectively.

**Discussion**

The results of our study indicated no statistically significant relationship between treatment type and richness indicating that there is no relationship between the presence of *A. stutchburyi* and species richness. Our hypothesis was not supported. The findings are unusual given insights about the effect of Cockles on species richness (Lohrer et al 2004), (epa.govt.nz 2017).

For each treatment, various effects were assumed. In the black, no Cockle, no Ulva, treatment, it was assumed that there would be no significant change as this was our control treatment. In the yellow, Ulva and no Cockle treatment it was expected that there would be a sharp increase in oxygen consumption as the Ulva added was dead and consumes oxygen upon decomposition (Teichberg et al. 2010). In the blue treatment of Cockles and no Ulva, it was expected that Cockles would consume more algae and other primary producers such as MPB from their surrounding environment as no food was provided for them. This would theoretically increase ammonia and phosphate as fewer organisms would be present that consume or fix it (Howarth et al. 2006). In the green, Cockles and Ulva treatment, it was expected that the effects of the blue treatment would be present but lesser because the Cockles would have to rely less on algae, etc. as a food source.

Estuarine ecosystems are partially enclosed coastal bodies of water most often containing brackish water with free flow between the ocean and inputs from freshwater sources such as rivers and streams. This combination of marine and freshwater and the flow of sediment from input sources makes estuaries nutrient rich ecosystems (Kaiser 2011). This influx of nutrients can create problems however by increasing primary production to a point where the decay of producers reduces the dissolved oxygen content of the water, thereby reducing the ability of many organisms to live there (Kaiser 2011).

Cockles act as regulators of the primary producers in their estuarine environments. They do this by consuming algae and other photosynthetic organisms through filter feeding. This ability to maintain an ecosystem by keeping the levels of primary producers in check and regulating nutrient fluxes makes Cockles a keystone species (Jones 2011). The removal of Cockles from a food web, if they are not replaced by another suitable species, can result in an overabundance of primary consumers with nothing around to eat them. Their decay can consume large amounts of oxygen and create dead zones, anoxic conditions, in which species richness and diversity plummet (Teichberg et al 2010).

This ability of Cockles to suppress primary producer numbers in a bottom-up trophic cascade make Cockles essential to maintaining an autochthonous community where the producers make food for the Cockles and other organisms, and these organisms, in turn, provide food for other consumers (Thrush et al. 2006). These other consumers can be second level consumers, that feed third level consumers, but can also be decomposers and parasites. Cockles also, in their ability to regulate the amount of nutrients (through feeding and bioturbation) in the estuarine system, influence benthic-pelagic coupling (Lohrer 2004), (Marinelli et al. 2009), (Norkko et al. 2001). Thus, preventing excessive amounts of phosphate and ammonia from entering the ocean which could create anoxic areas detrimental to life (Teichberg et al 2010).

In other studies, Cockles have been shown to increase ammonia levels, decrease oxygen levels, and have little effect on phosphate levels at high densities (Sandwell et al. 2009). Other studies have also associated the presence of Cockles with a higher biodiversity, specifically higher species richness (epa.govt.nz. 2017). Nutrient concentrations have also been shown to effect species richness in estuarine ecosystems. Excessive amounts of phosphate and ammonia can create algal blooms that, if not consumed quickly enough, can result in ecosystem harm by using all available oxygen in their decay. It follows then that relatively higher amounts of dissolved oxygen are associated with relatively higher species richness.

A longer study may have produced significant results. The short duration of our experiment may have masked the effect of surrounding macrofaunal communities on our study area (Sandwell et al. 2009). The presence of these communities could have allowed for geographical drift of species into our study area creating higher species richness. Thrush et al. did a similar study on ecosystem interaction networks over the course of 100 days giving significant results showing Cockles greatly affected nutrient fluxes. Our study time of two weeks may have been too short to provide accurate, relevant data. Also, our use of 300 Cockles across all treatments may have prevented us from detecting differences related to higher or lower densities of Cockles. Using much higher and lower densities of Cockles could have provided data with more pronounced differences leading to more significances in data analysis (Sandwell et al. 2009).

**List of tables**

**Results of Statistical Analysis**

Table 1: Summary of ANOVA analysis for Richness v Treatment, Oxygen v Treatment, Ammonia v Treatment, and Phosphate v Treatment. Gives FDF (factor degrees of freedom), EDF (error degrees of freedom), p-value (significance level), F-statistic (Variation within groups; if > 1 cannot distinguish between groups), and Adjusted r2 (amount of variability in dependent variable)

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| ANOVA RESULTS | Richness | Oxygen | Ammonia | Phosphate |
| FDF | 5.446 | 4 | 4 | 4 |
| EDF | 4 | 4 | 4 | 4 |
| P-value | 0.06759 | .6297 | .9201 | .6093 |
| F-statistic | 5.446 | .6368 | .1567 | .6787 |
| Adjusted r2 | .6558 | .-0.1843 | -0.5659 | -0.1597 |

**Figure legends**

Figure 1: Initial Oxygen concentration per treatment type (pink) and final Oxygen concentration per treatment type (blue) in mg/L. Initial and Final concentrations calculated by taking the mean of all initial and all final values for each treatment respectively.

Figure 2: Initial Phosphate concentration per treatment type (pink) and final Phosphate concentration per treatment type (blue) in µg. L-1. Initial and Final concentrations calculated by taking the mean of all initial and all final values for each treatment respectively.

Figure 3: Initial Ammonia concentration per treatment type (pink) and final Ammonia concentration per treatment type (blue) in µg. L-1. Initial and Final concentrations calculated by taking the mean of all initial and all final values for each treatment respectively.

Figure 4: Initial Richness concentration per treatment type (pink) and final Richness concentration per treatment type (blue). Initial and Final concentrations calculated by taking the mean of all initial and all final values for each treatment respectively.

**List of figures**



Figure 1.



Figure 2.



Figure 3.



Figure 4.

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The Portobello Marine Laboratory

My wetsuit-having classmates

My other classmates

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