Macroinvertebrate Densities and Plant Preference as Mechanisms Leading to Changes in Alternative Stable States

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Abstract

Earlier studies have suggested that alternative stable states exist in regards to aquatic plant densities in the presence of invasive rusty crayfish populations. While crayfish density appears to be a factor in causing shifts between stable states of low and high aquatic plant densities, other mechanisms have yet to be identified. Using *O. propinquus*, a previous invader, this experiment tested two possible mechanisms leading to shifts between these stable states: plant consumption in the presence of various macroinvertebrate densities and the preference of crayfish for either new or old growth plant. The data obtained from this experiment found that there was no significant difference in plant consumption over varying invertebrate densities ($p=0.360$), and crayfish have no apparent preference for either old or new growth plants ($p=0.245$). Based on the results, impossible to link either macroinvertebrate densities or new/old growth plant preferences with the alternative stables states observed in plant densities.

Introduction

Alternative stables states exist in nature when small incremental environmental changes have drastic impacts on communities (Mittelbach et al. 2006, van Ness and Scheffer 2004, Scheffer and Carpenter 2003). This phenomenon was likened to capsizing a boat by overloading one side; the boat will continue to tip until one more person is added that causes it to overturn and the “threshold” is surpassed (Scheffer and Carpenter 2003). Alternative stable states have been observed in both terrestrial and freshwater systems (Schroder et al. 2005). Schroder et al. (2005) states that further research on alternative stable states should focus on determining the mechanisms leading to the two states.

Field work conducted by Rosenthal (2004) has suggested that alternative stable states exist for the invasive rusty crayfish (*Orconectes rusticus*) in regards to their consumption and removal of aquatic plants in lakes in northern Wisconsin (gray box in Figure 1). Rusty crayfish are native to the Kentucky and Tennessee area, but has expanded its range throughout the Great Lakes region including lakes around UNDERC property, mainly by anglers using them as bait (Capelli and Magnuson 1983, USGS 2008). One major impact rusty crayfish have after they become established in lakes and reach high densities is reduce plant densities and species richness (Lodge and Lorman 1987, Lodge et al. 1994).

Currently, the rusty crayfish is not present on UNDERC property. The northern clearwater crayfish (*O. propinquus*), a previous invader (Capelli and Munjal 1982), however, is found on property and has a similar effect on its environment when it is present in high densities (J. Peters, personal communication). I will use the northern clearwater crayfish in lab experiments to test two mechanisms that may lead to alternative stable states of plant densities. These mechanisms include 1) the consumption of macroinvertebrates as an alternative food source to plants and 2) the consumption preferences between old growth and new growth vegetation.

The first mechanism that was evaluated was the presence of macroinvertebrates as an alternative food source to plants. Crayfish consumption of macroinverebrates has been shown to lead to higher growth rates compared to consumption of plants (Hill et al. 1993). I expected that the presence and abundance of macroinvertebrates would have an effect on the plant consumption by crayfish and would therefore influence which stable state of vegetation density was achieved. For plants to remain in the high density state (Figure 1, Circle A), I hypothesize that there has to be an abundance of macroinvertebrates available for crayfish consume, therefore ignoring the presence of plants. I hypothesize that plants will be in the low density state (Figure 1, Circle B) in environments where macroinvertebrates are rare or absent and the crayfish would be left no other option than to consume the surrounding plants. Mixed results of each stable state (Figure 1, Circles A and B) can be expected when levels of macroinvertebrates are moderate.

The second mechanism that was tested was the preference of old growth vs. new growth of the plants by the crayfish. The old growth plants represent the stable state in which the plants are abundant (Figure 1, Circle A), where crayfish have no affect on plant density. The new growth, on the other hand, represents the state where crayfish have clipped and continue to keep the plants at low densities (Figure 1, Circle B). For plants to remain in the high density state (Figure 1, Circle A), I hypothesize that crayfish do not prefer old growth plants. On the other hand, I hypothesize that if crayfish do prefer old growth plants then the plant state can change from high density to low density (in Figure 1: a movement from Circle A to Circle B). I hypothesize that the plants would stay in the low density state (Figure 1, Circle B) if the crayfish prefer the new plant growth as the crayfish will continually clip the new growth and the plants will never be able to regenerate.

Methods

To test these hypotheses, northern clearwater crayfish were collected from Tenderfoot Lake using traps baited with beef liver as well as by snorkeling. Tanks were filled with 600 grams of sand collected from Tenderfoot Lake. The sand was allowed to dry for 3 weeks to ensure any invertebrates in the sand were no longer living. The tanks were then filled with 2.45 liters lake water and underwent constant aeration.

The tanks were filled with three levels of macroinvertebrates. One treatment required no macroinvertebrates added to the tank, a second treatment had a moderate level (4-5) invertebrates added to the tank, and a final treatment had a high level (10-12) invertebrates added to the tank. Three different types of invertebrates were used in each of the treatments. The invertebrates used were Ephemeroptera larvae, snails of the genus *Helisoma* and *Physella*, and earthworms of the order Aporrectodea. Each invertebrate group was used in a different experimental trial.

The plant species used was *Potamogeton robbinsii* collected from Tenderfoot Lake. 2.00 ± 0.20 grams of old growth and new growth were anchored in the tanks by a black clip and with half the treatments receiving old growth plants and the other half receiving new growth plants. This resulted in six fully crossed treatments: three densities of macroinvertebrates tested with both new growth and old growth plants present.

One crayfish ranging in size from X-X mm carapace length was added to each of the tanks and left for 48 hours. After 48 hours, invertebrate survivorship was observed and the remaining plant mass was recorded.

The results were evaluated with several statistical tests. A simple correlation determined if there was a relationship between size and plant consumption, and a t-test was used to determine if the sex of the crayfish has any effect on how much it eats.
A 2-way ANOVA was used to determine if there is a significant difference among plant and macroinvertebrate consumption in the different macroinvertebrate densities. A Tukey's multiple comparison test was used to test which treatments are significantly different from each other. All analyses were completed using SYSTAT 12.

There were two null hypotheses for this experiment. The first is that the macroinvertebrate abundance does not affect the consumption of plants by the northern clearwater crayfish. The second is that the northern clearwater crayfish has no preference between old growth and new growth plants. Both results were evaluated to determine if they are possible mechanisms explaining the observed pattern of alternative stables states in plant abundance.

Results

First, a t-test was used to determine whether or not the sex of the crayfish affected the amount of plant consumption. There was no significant difference in the amount of plant consumption between males and females ($F_{1,52}$=0.571; p=0.453). Therefore, all of the data collected for both males and females was combined for the rest of the analyses.

The simple correlation between crayfish size and plant consumption indictates that there was no relationship between the two variables (R^2=0.0213, Figure 2). Therefore, plant consumption did not increase with crayfish size and the data from crayfish of all sizes was in the rest of the analyses.

Although there was a greater amount of consumption of old growth plants vs. new growth plants at all 3 invertebrate densities (Figure 3). A 2-way ANOVA demonstrated that there was no significant difference between the consumption of new growth or old growth *Potamogeton robbinsii* ($F_{1,48}$=1.385, p=0.245). Additionally, there was no significant difference for consumption of plants at differing macroinvertebrate densities ($F_{2,48}$=1.045, p=0.360) and there was also no interaction between the macroinvertebrate levels and the old growth/new growth plants in each treatment ($F_{2,48}$=0.223; p=0.801).

Next I wanted to determine if there was a difference in the amount of plants consumed when different invertebrate taxa were present. I conducted a 1-way ANOVA examining the total plant consumption (old and new growth combined) with the three types of invertebrates. There was significantly less plants consumed when snails or worms were present (Figure 4, $F_{2,51}$=5.586; p=0.006). Through the use of Tukey's Multi-Comparison Test, it was found that treatments using Ephemeroptera as invertebrates demonstrated more total plant consumption than both snails (p=0.007), and worms (p=0.024).

Separate tests were performed on each of the macroinvertebrates taxa. A two-way ANOVA demonstrated that no significant difference between old growth and new growth plant consumption for all three of the taxa (Ephemeroptera: $F_{1,12}$=1.276; p=0.281, Snails: $F_{1,18}$=2.195; p=0.156, Earthworms: $F_{1,12}$=0.377; p=0.551). Also, there was no statistical difference in the amount of plant consumption at the 3 density of the 3 taxa (Ephemeroptera: $F_{2,12}$=2.270, p=0.146, Snails: $F_{2,18}$=1.100, p=0.354, Earthworms: $F_{2,12}$=1.008; p=0.394) and there was no interaction between these two variables ($F_{2,12}$=0.343; p=0.716). The average plant consumption across all variables is seen in Figure 4.

Discussion

In general there was no significant difference in plant consumption for all of the invertebrate treatments and the null hypothesis that macroinvertebrate densities affect the plant consumption of the northern clearwater crayfish can not be rejected. In addition, there was no statistical difference in the amount of old vs. new growth plants consumed and again I fail to reject the null hypothesis that the northern clearwater crayfish has no preference for either old or new growth plants. With the data collected in this experiment, I am unable to determine if old growth or new growth of plants or if increasing invertebrate densities are mechanisms that will lead to the pattern of alternative stable states found in plant abundance when crayfish reach high densities.

With an ANOVA test, I was able to determine that crayfish consumed less of the plants when snails or earthworms were used instead of mayflies. This could indicate that in areas where snails and earthworms are abundant, plant densities could remain in a high stable state due to lower plant consumption. Future studies would be necessary before any conclusions are made. To improve upon this particular experimental design investigating the effects of macroinvertebrate densities, ideally the same vertebrate taxas would be used throughout all of the replicates. In my experiment I was limited to the invertebrates that were present and abundant, however, given more time and resources in a future experiment it would be beneficial to use representative invertebrate communities and densities found in lakes. Further investigation might also be warranted to find different preferences of crayfish consumption among different macroinvertebrates. It should be noted that consumption was noted in all taxa used in this experiment through personal observation.

There are multiple changes that could be made to this experiment in the future to make it more realistic and informative. For instance, I kept old growth and new growth plants in separate treatments to test plant preference by evaluating how much new or old growth was consumed in individual tanks. A future experiment might place both types of plants in the same tank to give crayfish the option of eating one or the other. In addition I limited my experiment to just using *P. robbinsii*, but obviously in lakes crayfish will be exposed to multiple plant species. Therefore in future experiments I suggest that multiple plant species are used. If both old and new growth are combined or if multiple plant species are used, there is one potential problem that will need to be resolved for future experiments which is that partially eaten plant pieces whether they are old or new growth or even different plant species may be challenging to distinguish between each other. Another consideration when deciding what type of plant species or plant morphologies to use is the nutritive and secondary compound content of the plants, as studies have shown that aquatic plants are also likely to have chemical defenses (Rowell and Blinn 2003). For example, perhaps while new growth plants probably are more abundant in nutrients, as is common in terrestrial new growth, there could also be some secondary compounds produced by *Potamogeton robbinsii* that deter crayfish from eating too much (Farji-Brener 2001).

A third set of suggestions has to do with the crayfish used. One factor that might have influenced the outcomes of this laboratory experiment is the unusually late molting of the crayfish this year. It is possible that recently molted crayfish or those who are about to undergo the molting process are under too much physical stress to practice normal eating patterns, thus altering the data. Future studies should use crayfish that have recently molted and which are unlikely to molt again for quite awhile. Also, there was limited success this year with trapping and snorkeling for northern clearwater crayfish. While the experiment had originally been designed for larger crayfish species, the availability of smaller specimens led to a slight alteration in the experiment. Perhaps larger crayfish would be more likely to lead to the alternative stable states compared to small crayfish. In addition, crayfish could be given more time than the 48 hours used in this experiment. If crayfish were given longer, however, the number of invertebrates should be kept at a constant level for each treatment.

Finally, this experiment might also be tried outside of a laboratory to determine whether or not crayfish act differently depending on the habitat they occupy. Ultimately, the findings in this experiment warrant further investigations of these and other mechanisms that may contribute to the changes in stable states of plant abundance.

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Appendix

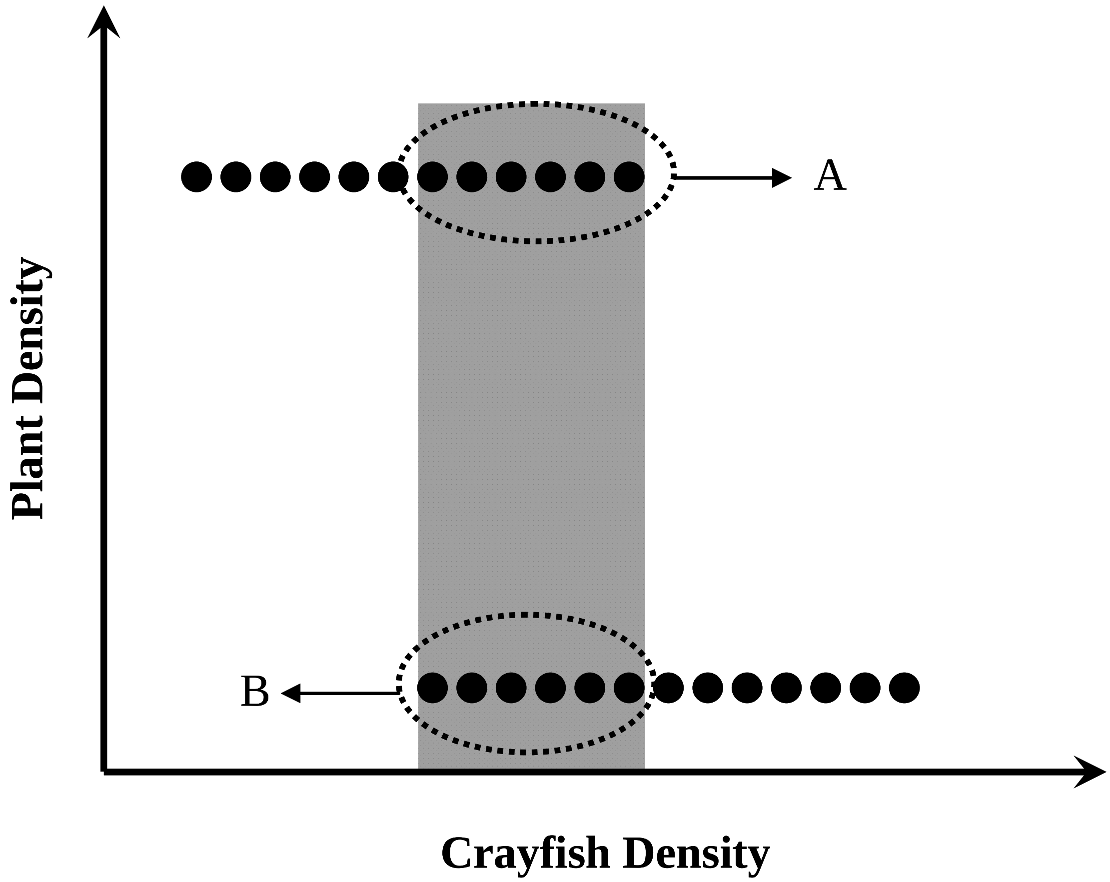

Figure 1. Pattern of alternative stable states found in lakes with rusty crayfish. The gray box is the overlapping stable states. Circle A denotes lakes with intermediate crayfish densities where high densities of plants are found, whereas Circle B denotes lakes with intermediate crayfish densities and low plant densities are found. The mechanism leading to these two overlapping states will be examined in my experiment this summer. The figure was modified from graphs by Rosenthal (2004).

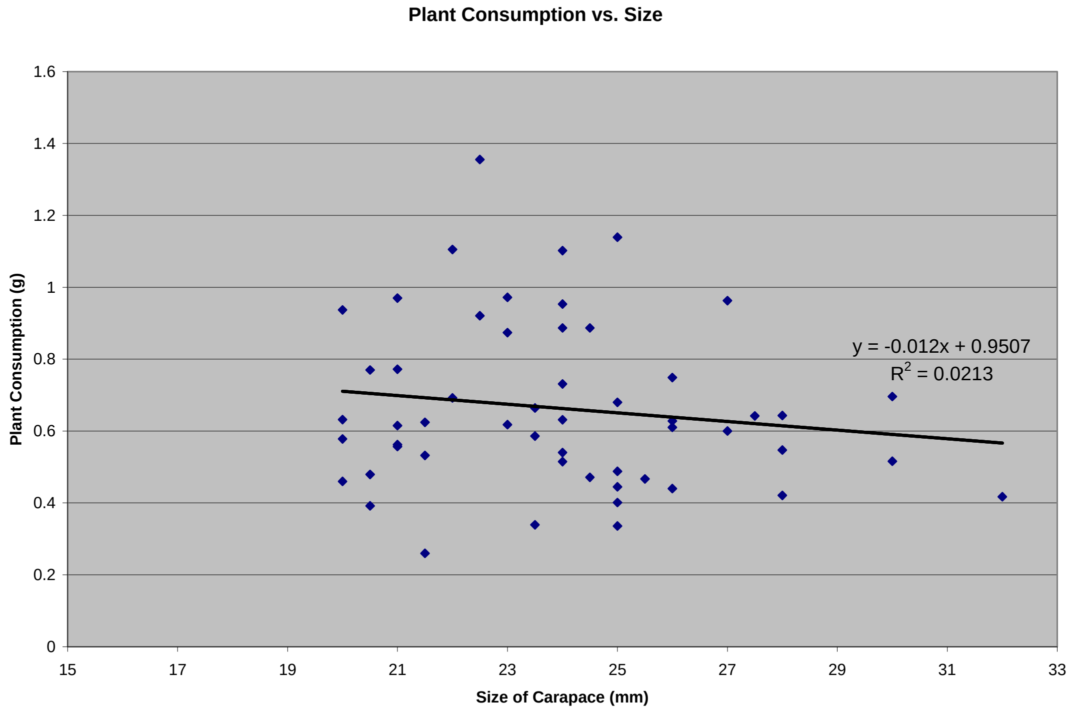

Figure 2. A simple correlation between the size of a crayfish and the amount of plant consumption observed.

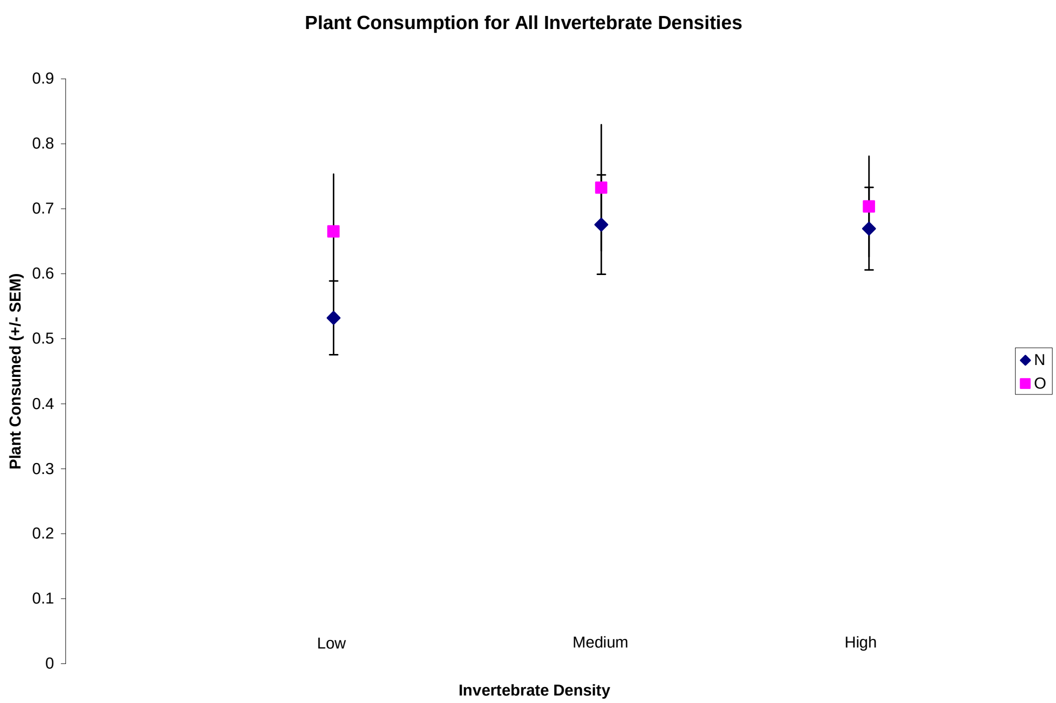

Figure 3. Average consumption for old growth and new growth plants are depicted across differing macroinvertebrate densities. Diamonds represent new growth plants and boxes represent old growth plants.

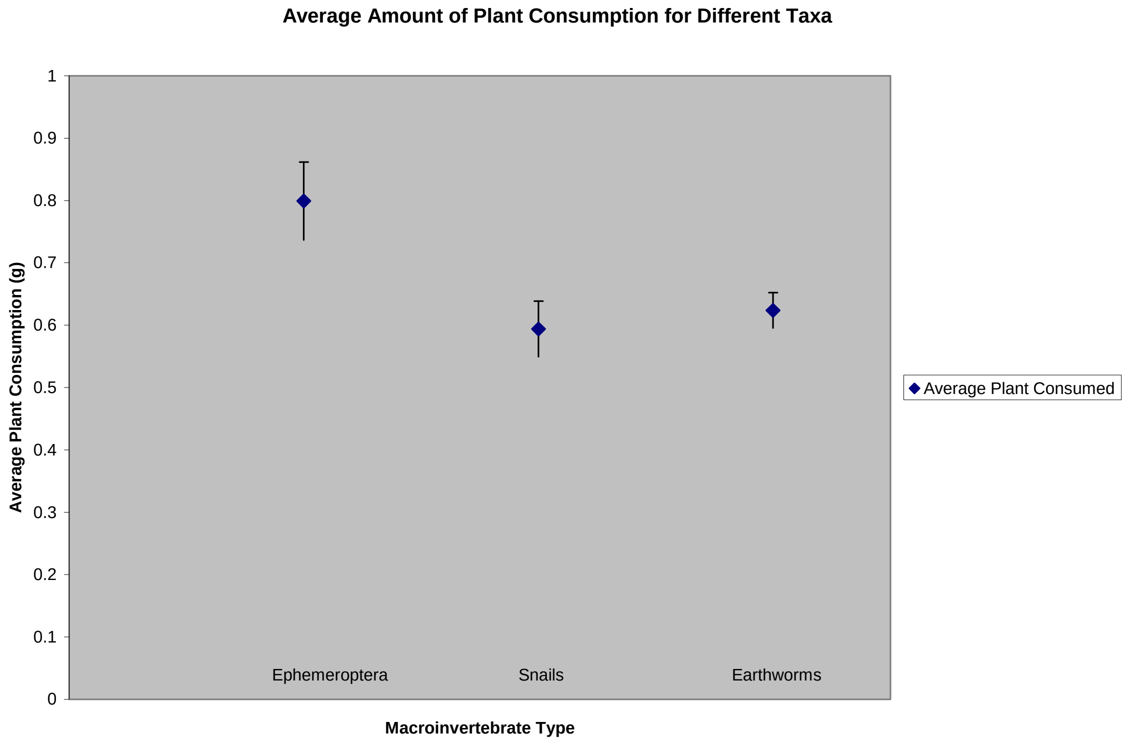

Figure 4. Average consumption of all plants are depicted across different taxa. Each diamond represents a different taxa.