The Effect of Northern Clearwater Crayfish (*Orconectes propinquus*) Abundance on Stream Invertebrate Diversity in Michigan's Upper Peninsula

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Abstract

Crayfish are an essential component of stream ecosystems in the Northern Hardwoods. Due to their diversity of roles, from shredders to herbivores to predators of macroinvertebrates, crayfish such as the Northern Clearwater crayfish, *Orconectes propinquus*, can have a significant effect on aquatic community composition. Because of the complexity and strength of the northern clearwater crayfishes's interactions with its environment, they can even be considered to be a keystone species. To test this hypothesis, sixteen artificial streams with three, two, one, and no crayfish, each with four replicates were run for eighteen days. In addition, six in situ plots with two, one, and no crayfish, each with two replicates, were placed in Tenderfoot Creek for one week on the University of Notre Dame Environmental Research Center (UNDERC) property in Gogebic County, Michigan. The results of these experiments reveal that as crayfish populations increased, algae and periphyton cover in the artificial streams decreased, and that invertebrate abundance in all plots decreased. Moreover, the crayfish number and invertebrate diversity in artificial streams were positively correlated. These findings can help understand and support noninvasive crayfish populations in the Northern Hardwoods, especially given the imminent spread of the invasive species *Orconectes rusticus*.

Introduction

An ecosystem is often intricate and complex; however, better understanding of one aspect can lead to better understanding of the whole. Studying native crayfish in northern temperate streams can serve as one viewpoint by which to examine these ecosystems. Crayfish are particularly important to temperate North American stream ecology for a variety of reasons. For instance, they are omnivorous, so their possible effects on species abundance is widespread. They are also decomposers, so their breakdown of organic matter helps support other species present in the stream. And finally, as a result of these two characteristics, crayfish arguably are a keystone species, necessary in maintaining stream health and species diversity (Creed 1994).

There have been many previous studies on the effect of crayfish on freshwater systems in the upper Midwest of North America. Most studies have focused on one of the most important functions of crayfish: vegetation breakdown (Creed 1994; Charlebois 1996). Crayfish can graze off of both allochthonous, such as fallen tree leaves, and autochthonous, such as algae, vegetation input (Nyström *et al*. 1999; Keller and Ruman 1998; Mormot *et al*. 1978). *Orconectes propinquus* also grazes on periphyton, a mixture of algae, cyanobacteria, microbes, and detritus found on submerged rocks and stream bottoms (Creed 1994). Periphyton is important for ecosystem energy flow, but a weaker competitor than filamentous algae (Creed 1994; Charlebois, 1996). Taking this into account, studies have shown that crayfish serve to decrease filamentous algae and therefore increase periphyton biomass as well herbivorous invertebrate diversity in stream ecosystems, thus providing a large base for a complex food web with high productivity (Creed 1994; Mormot *et al.* 1978). Despite the importance of crayfish herbivory, the majority of the their diet consists of macroinvertebrates (Hills *et al.* 1993). Therefore, crayfish indirectly help populations of periphyton and invertebrates grow by fostering their niche and directly control them via feeding (Charlebois 1996).

Determining native crayfish as essential to the University of Notre Dame Environmental Research Center - East property is especially important given the impending damage caused by introduction of the invasive rusty crayfish (*Orconectes rusticus*) in northern temperate freshwater zones (Charlebois 1996). Rusty crayfish were first spotted in Trout Lake, Wisconsin in 1979 (Charlebois 1996). Since then, they have been advancing through freshwater systems at an average rate of 0.68 km/year (Charlebois 1996). Although throughout this time overall crayfish abundance rose, the native population of crayfish in affected lakes almost disappeared (Charlebois 1996). The surge of invasive crayfish also correlates with decreased levels of fish with similar diets to the crayfish, as well as snails and macrophytes (Charlebois 1996). Unfortunately, experiments aimed at studying the reversibility of rusty crayfish damage do not end optimistically. Most experiments conclude that reintroductions would only happen if artificially aided and would not occur quickly (Rosenthal *et al.* 2006; Wilson *et al.* 2004).

Given the imminent spread of damage caused by rusty crayfish invasion, it would be helpful to assess the effect of noninvasive Northwoods crayfish (*Orconectes propinquus*) on pre-invasion stream community dynamics by observing the effects of varying crayfish abundance on algae and macroinvertebrate abundance. *Orconectes propinquus* is not a native species, yet it does not have a known negative impact on the surrounding ecosystem for it to be considered invasive (Hill *et al.* 1993). Studies done in both artificial and *in situ* plots may provide more accurate results on crayfish-community dynamics. Moreover, establishing the role of noninvasive crayfish could add to the debate over trophic cascades and help elucidate if low trophic level species, such as crayfish, control ecosystem structure in the way that apex predators do (Usio, 2000). This experiment could further support the idea of crayfish as keystone species, hopefully leading to increased protection and conservation (Usio, 2000).

Because of their polytrophic status, I hypothesize that crayfish will act as important controls for both algal and macroinvertebrate populations. Specifically, I predicted that crayfish presence will decrease filamentous algal biomass and macroinvertebrate abundance through predation (Creed 1994). I also hypothesize that crayfish will increase periphyton biomass and macroinvertebrate diversity by opening niches, as crayfish keep highly competitive species in check (Nyström 1999).

Methods

Research was conducted at UNDERC - East in Michigan's Upper Peninsula, in Gogebic County, Michigan. To start, two riffles along Tenderfoot Creek within the property were measured in length, width, depth, and velocity (Figure 11). Then, an area of the riffle was marked off with flags to be surveyed for its crayfish population. The plot was combed through, including under rocks, to count the amount of crayfish. Later, crayfish density was determined for each riffle. In addition, two invertebrate samples were taken from each riffle using Surber Samplers.

The results of the survey dictated the densities of the artificial streams. The highest density recorded within the riffle, three crayfish per meter squared, was the highest amount of crayfish in an artificial stream. The next stream had two crayfish, followed by one crayfish, and finally no crayfish for control. This set was replicated four times over 16 artificial streams. The artificial streams contained rocks for crayfish hiding space and shade. The flow was kept at 65 centimeters per second and with a constant slow inflow of lake water. The artificial streams ran for a total of 18 days, with a one week period used to acclimate the the crayfish. At the end of the 18 days, the contents of the artificial streams were removed and the water was strained to collect filamentous algae. The strained samples were searched through to quantify macroinvertebrate populations. Periphyton was collected by scraping a 17 centimeter by 10 centimeter rectangle off the bottom of the artificial streams. Periphyton algal was dried via vacuum filtration and then massed, while filamentous algae was partially dried by vacuum filtration and then left in a drying oven set at 40 degrees Celsius overnight and then massed.

In addition to artificial stream experiments, there were in-stream plots with manipulated crayfish densities. The in situ plots were constructed out of 11.8 inch by 4.37 inch plastic baskets with openings on the side and plastic wiring with holes a quarter of an inch in diameter stitched to the top with fishing wire. The densities for the plots were two, one, and zero for control, with two replicates each. The baskets were loaded with rocks from Tenderfoot Creek and placed in one of the riffle areas sampled. These in situ plots were left in the creek for a week and then collected with the aid of a Surber Sampler to quantify invertebrate populations.

Invertebrate abundance data were collected for each artificial stream and *in situ* plot and used to determine species richness, the Simpson index, and the Shannon-Wiener index. SYSTAT 2011 was used to statistically analyze the data. The data were checked for normalization with the Shapiro-Wilk test. To visualize the relationship between crayfish abundance in artificial streams and floating algal biomass within each plot, a regression analysis was performed. Other regressions were performed to determine the relationship between crayfish abundance in artificial streams and periphyton biomass, crayfish abundance in artificial streams and macroinvertebrate abundance, crayfish abundance in artificial streams and macroinvertebrate species richness, and crayfish abudance in artificial streams and Simpson index of macroinvertebrate diversity. The relationships between crayfish number and macroinvertebrate abundance and diversity were also analyzed using regression analysis for the in situ plots, in addition to one analyzing the relationship between crayfish abundance and Shannon-Wiener index for macroinvertebrate diversity. Finally, an ANOVA was used to analyze the difference in the Shannon-Wiener diversity index among the differing crayfish abundances of the *in situ* plots.

Results

Algae

For the artificial streams, an increase in crayfish abundances explained 87.86% of the decline filamentous algae biomass ($p = 0.000008$, $R^2 = 0.878557$; Table 1; Figure 1). An increase in crayfish abundance in artificial streams also explained 98.63% of periphyton biomass ($p = 0.013688$, $R^2 = 0.986312$; Table 1; Figure 2). However, crayfish abundance did not significantly effect percentage sugar maple leaf eaten ($p = 0.704821$, $R^2 = 0.295179$; Table 1; Figure 9).

Macroinvertebrates

In artificial streams, crayfish abundance explained 78.93% of the decrease in macroinvertebrate abundance ($p = 0.000165$, $R^2 = 0.789348$; Table 1; Figure 8). The decline in species richness was explained 59.43% by increased crayfish abundance ($p = 0.015200$, $R^2 = 0.594267$; Table 1; Figure 3). The Shannon-Wiener index showed no significant difference in diversity among plots of differing crayfish abundance ($p = 0.139734$; Table 1; Figure 10). However, crayfish abundance explained 96.77% of Simpson Index negative variance, meaning increased diversity as the crayfish population increased ($p = 0.032305$, $R^2 = 0.967695$; Table 1; Figure 6).

For the *in situ* plots, neither the Shannon-Wiener ($p = 0.867555$, $R^2 = 0.161882$; Table 1; Figure 7) nor the Simpson index ($p = 0.867555$, $R^2 = 0.070874$; Table 1; Figure 4) showed a difference in diversity with differing crayfish abundances. In addition, there was not a significant relationship between crayfish numbers and macroinvertebrate abundance ($p = 0.148792$, $R^2 = 0.851208$; Table 1; Figure 5).

Discussion

The purpose of this experiment was to determine the importance of the crayfish (*O. propinquus)* within Northwood stream ecosystems. The significant impact of crayfish on both consumers and producers found does support the idea of crayfish as a keystone species. The hypotheses that increased crayfish abundance corresponds to decreased filamentous algal cover and macroinvertebrate abundance in all plots were supported by the results (Table 1). However, the hypotheses that increased crayfish abundances correlate to increased periphyton abundance , invertebrate species richness, and invertebrate diversity in all plots were not supported (Table 1).

The effects of crayfish abundance on algae could be explained by their herbivory. Numerous studies have shown a negative relationship between crayfish number and filamentous algal biomass due to increased feeding (Keller and Ruman 1998; Dorn and Wojdak 2004; Creed 1994). The results of the artificial stream experiments also show this decrease in metaphytic algae as crayfish density increased . Given that crayfish had no significant effect on the breakdown of the green leaf allochthonous input, it appears that crayfish focused their diet on the algae available and locally present macroinvertebrates. In addition, unlike the sugar maple leaves provided, algae has relatively few chemical defenses, making it a more preferable food choice over the maple leaves (Lodge *et al.* 1994). This lack of preference for the green leaf litter could also be due to the abiotic streams factors. Creed and Reed (2004) found that velocities greater than 50 centimeters per second and temperatures greater than 19 degrees Celsius impeded grazing on leaf litter. Throughout the experiment, the artificial streams violated at least one of these conditions, possibly shifting crayfish feeding preferences. In any case, this significant effect on algae populations supports the hypothesis that top-down control plays a significant role in aquatic community structure (Lodge *et al.* 1994).

However, the artificial streams resulted in something contrary to the literature: a significant decrease in paraphyton biomass as crayfish abundance increased (Nyström *et al.*, 1999; Lodge *et al.* 1994). This unexpected effect on periphyton biomass could be due to herbivory effects as well or could also be a result of the physical conditions of the experiment. With the dietary options in artificial streams being reduced relative to those in lakes or streams, the crayfish could rely more heavily on periphyton for food than in other in situ experiments (Lodge *et al.* 1994). The presence of crayfish in the artificial streams could have also dislodged a lot of the periphyton, even if it increased its growth, leading to inaccurate estimates (Lodge *et al.*,1994). Finally, many studies on dynamics between crayfish and periphyton involve the major regional predator of periphyton, snails. In these situations, increased crayfish densities increase periphyton biomass because crayfish predation on snails increases (Lodge *et al.* 1994; Weber and Lodge, 1990; Charlebois 1996). Because this experiment predominantly involved insect larvae, which have a dramatically smaller effect on periphyton biomass, instead of snails as the macroinvertebrates, the positive effect of crayfish on streams periphyton could be improperly represented (Lodge *et al.* 1994).

The rather diverse feeding behavior of the crayfish may have impacted macroinvertebrate populations in both artificial stream and *in situ* experiments. The finding of significantly decreased macroinvertebrate abundance in artificial streams coincides with previous studies on crayfish omnivory (Usio 2000; Lodge *et al.* 1994; Charlebois 1996). Usio (2000) explains that as crayfish populations increase, the amount of feeding on invertebrates and competition for algal food sources increase, while decreasing the invertebrate abundance. Additionally, as crayfish feed more, the quality of the leaf litter remaining in the system decreases, leaving less to support other species (Usio 2000). Lastly, as with periphyton, crayfish potentially dislodge larval populations just by moving around or hiding between rocks. This may in turn increase the probability that the invertebrates get flushed out of the artificial streams (Usio 2000).

The hypothesis of increased invertebrate diversity, however, was only supported in artificial streams when looking at the Simpson index . Increased diversity is partially a result of an increase in energy flow, as crayfish are leaf and algal shredders, so their breakdown of vegetation provides food for microscopic herbivores (Momot *et al*. 1978). Crayfish predation on macroinvertebrates keeps certain populations in check and allows more vulnerable species, such as leeches, to thrive (Stenroth and Nyström, 2003; Momot *et al.* 1978). However, the reason for lack of significant results among the in situ plots could be due to the short experimental period. The plots were only kept in the stream for one week, less than half the amount of time the artificial streams ran, and, according to Nyström (1999), increased diversity of invertebrates is a long-term effect of crayfish.

The strong influence on algal and invertebrate community structure supports the definition of crayfish as keystone species. In controlling thick, fast-growing metaphytic algal populations, crayfish keep the water column open to light and allow other types of algae and vegetation to thrive within stream ecosystems (Creed 1994). This increased diversity of vegetation, in turn, leads to increased invertebrate diversity (Creed 1994; Weber and Lodge

1990). Even if crayfish decrease periphyton biomass by grazing on it, grazing has been shown to increase periphyton efficiency by increasing its levels of chlorophyll *a* (Lodge *et al.* 1994). Also, as Usio (2000) notes, shredders become increasingly important as latitude increases and decomposition rates slow. The action of crayfish as decomposers helps support a whole level of essential microbes that were not studied in this experiment (Lodge *et al*. 1994).

Overall, crayfish serve as an important source of energy flow within stream communities, providing a link between a variety of different biotic and abiotic factors (Momot *et al.* 1978). Although this polytrophism complicates the idea of a typical trophic cascade model, Lodge *et al.* (1994) shows that even complex food webs can react in chain-like manner. This is especially relevant given the prediction that the introduction of an invasive species, namely *Orconectes rusticus*, will have considerable damage on the structure of Northwoods aquatic ecosystems (Hill *et al.* 1993). Rusty crayfish prove to be a stronger competitor over the current crayfish species and may have a negative influence on vegetation and invertebrate diversity over time (Hill *et al.* 1993). These effects may seem small scale; however, changes lower down on the food web, especially among macroinvertebrates, an important aquatic food source, eventually resonate throughout the entire community (Lodge *et al.* 1994; Nyström 1999). Therefore, it is of heightened importance to protect the status of noninvasive crayfish in the Northern Hardwoods to maintain the health and biodiversity of the region’s aquatic ecosystems.

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Appendix

Table 1: Summary of P-values for Response Variables to Varying Crayfish Populations

Figure	Relationship	Plot	Analysis	R^2 value	P-value
1	Crayfish Abundance and Filamentous Algal Biomass	Artificial Stream	Regression	0.878557	0.000008
2	Crayfish Abundance and Periphyton Biomass	Artificial Stream	Regression	0.986312	0.013688
3	Crayfish Abundance and Species Richness	Artificial Stream	Regression	0.594267	0.015200
4	Crayfish Abundance and Simpson Index	*In Situ*	Regression	0.070874	0.867555
5	Crayfish Abundance and Macroinvertebrate Abundance	*In Situ*	Regression	0.851208	0.148792
6	Crayfish Abundance and Simpson Index	Artificial Stream	Regression	0.967695	0.032305
7	Crayfish Abundance and Shannon-Wiener Index	*In Situ*	Regression	0.161882	0.906284
8	Crayfish Abundance and Macroinvertebrate Abundance	Artificial Stream	Regression	0.789348	0.000165

Figure	Relationship	Plot	Analysis	R^2 value	P-value
9	Crayfish Abundance and Percentage Green Leaf Litter Eaten	Artificial Stream	Regression	0.295179	0.704821
10	Crayfish Abundance and Shannon-Wiener Index	Artificial Stream	ANOVA	---	0.139734

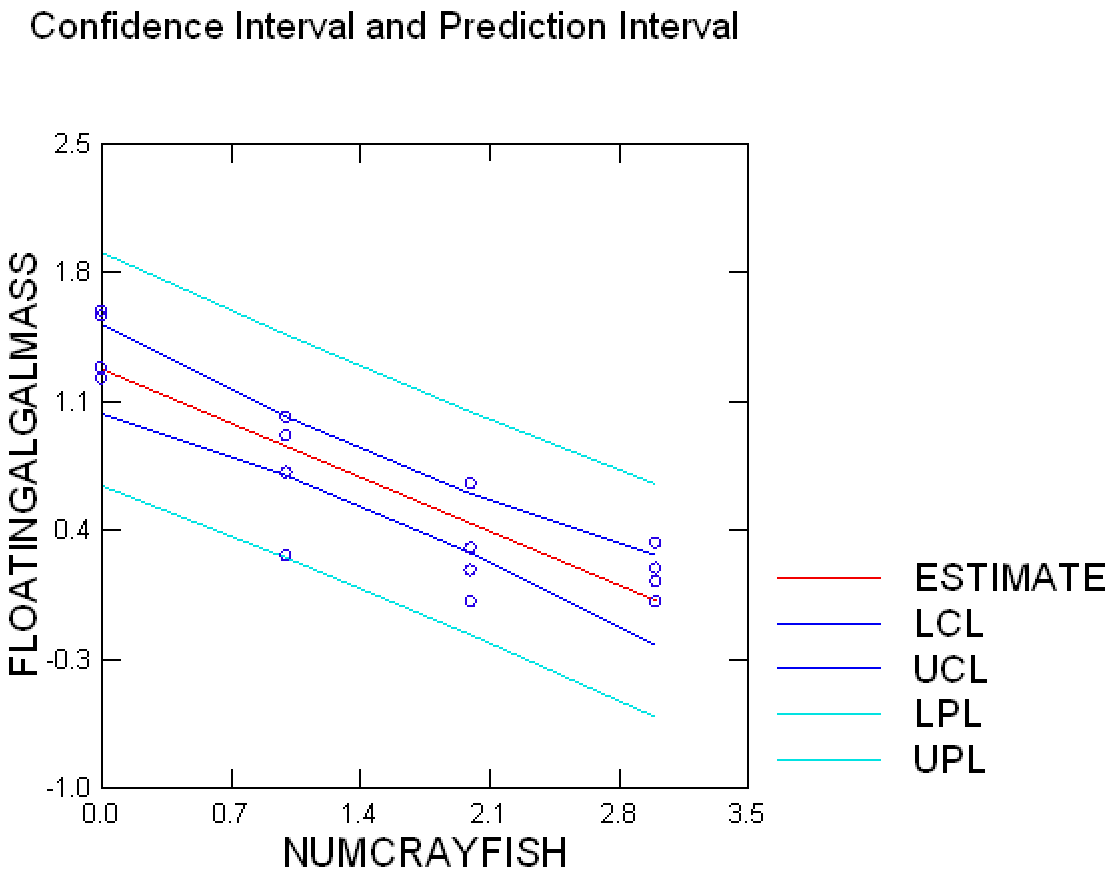

Figure 1: Biomass of Floating Algae at Different Crayfish Abundances in Artificial Streams

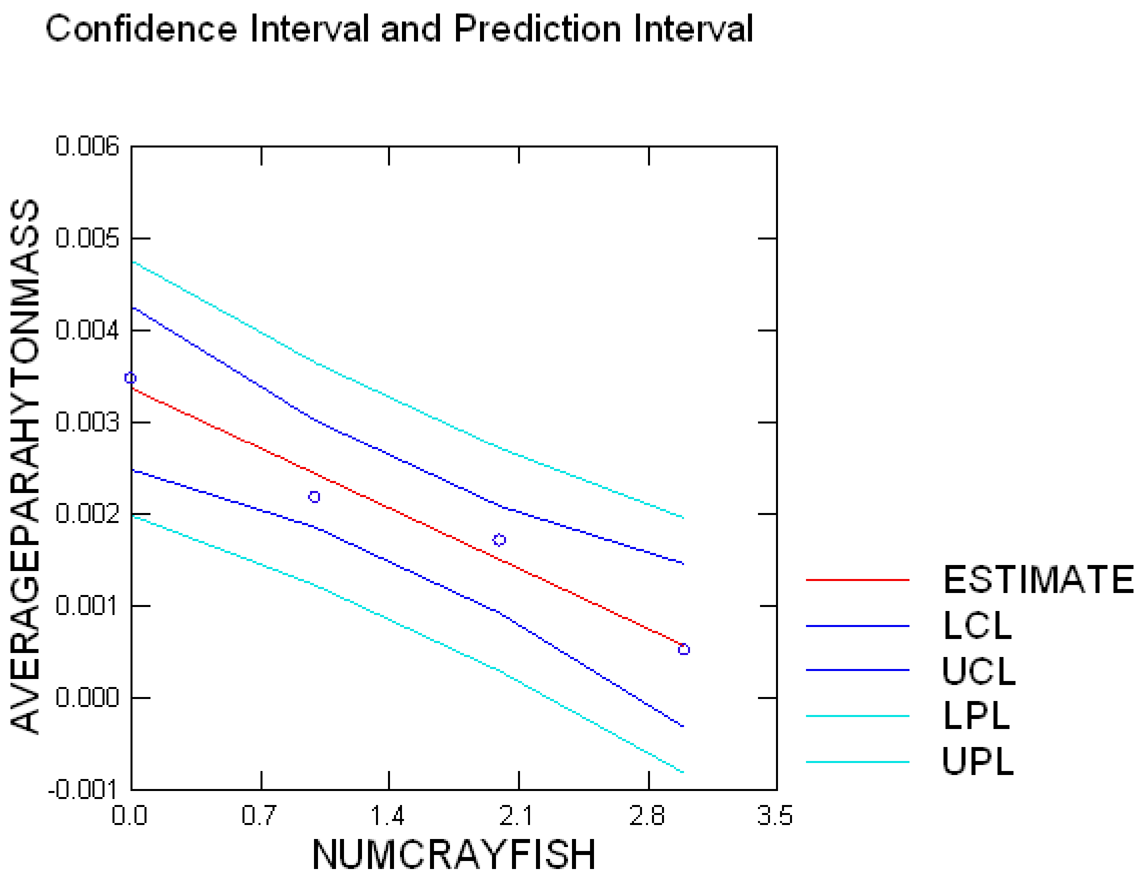

Figure 2: Biomass of Paraphyton at Different Crayfish Abundances in Artificial Streams

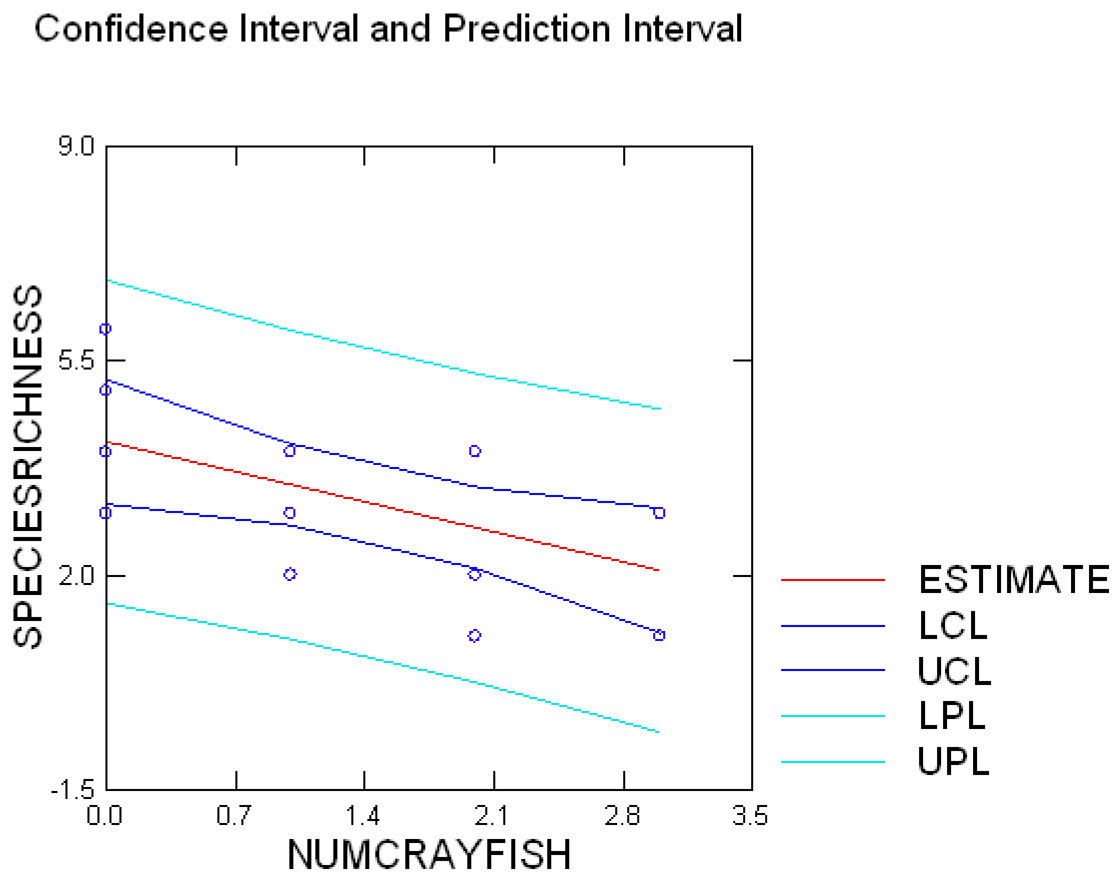

Figure 3: Macroinvertebrate Species Richness at Different Crayfish Abundances in Artificial Streams

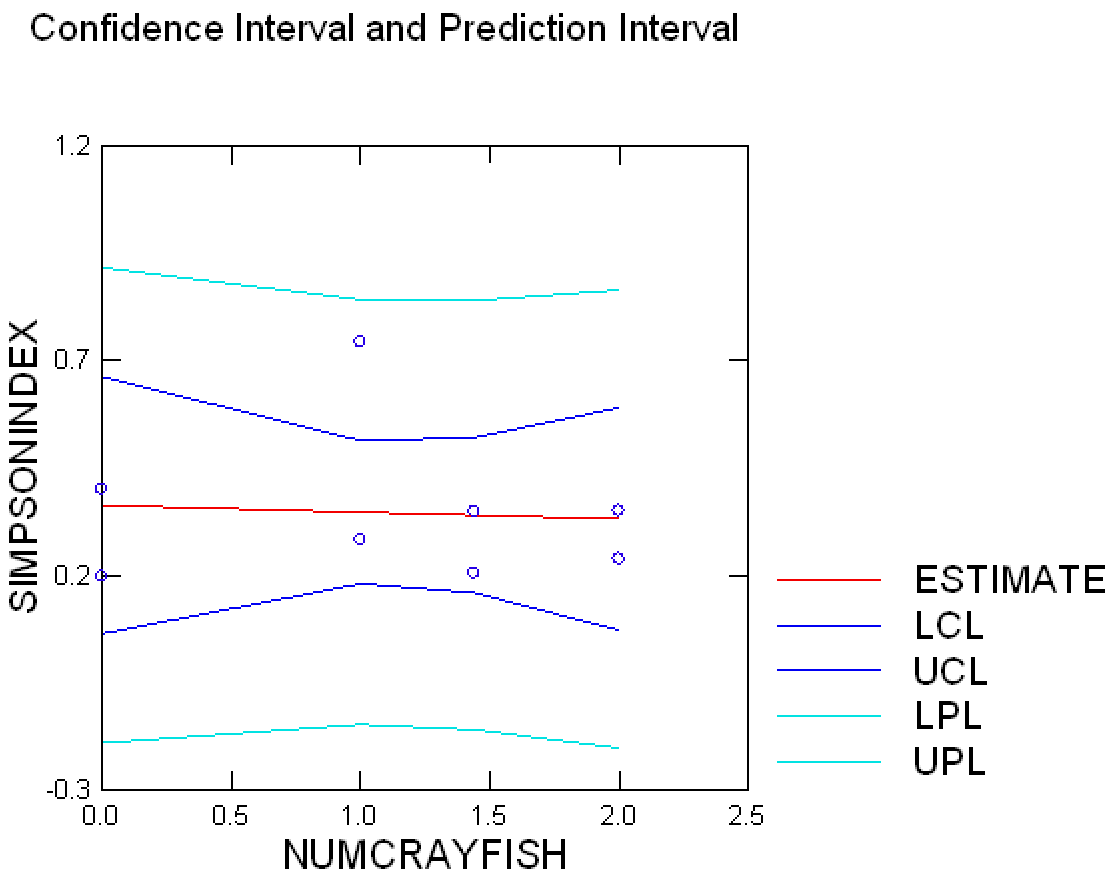

Figure 4: Macroinvertebrate Species Diversity as Measured by the Simpson Index at Different Crayfish Abundances in the *In Situ* Plots

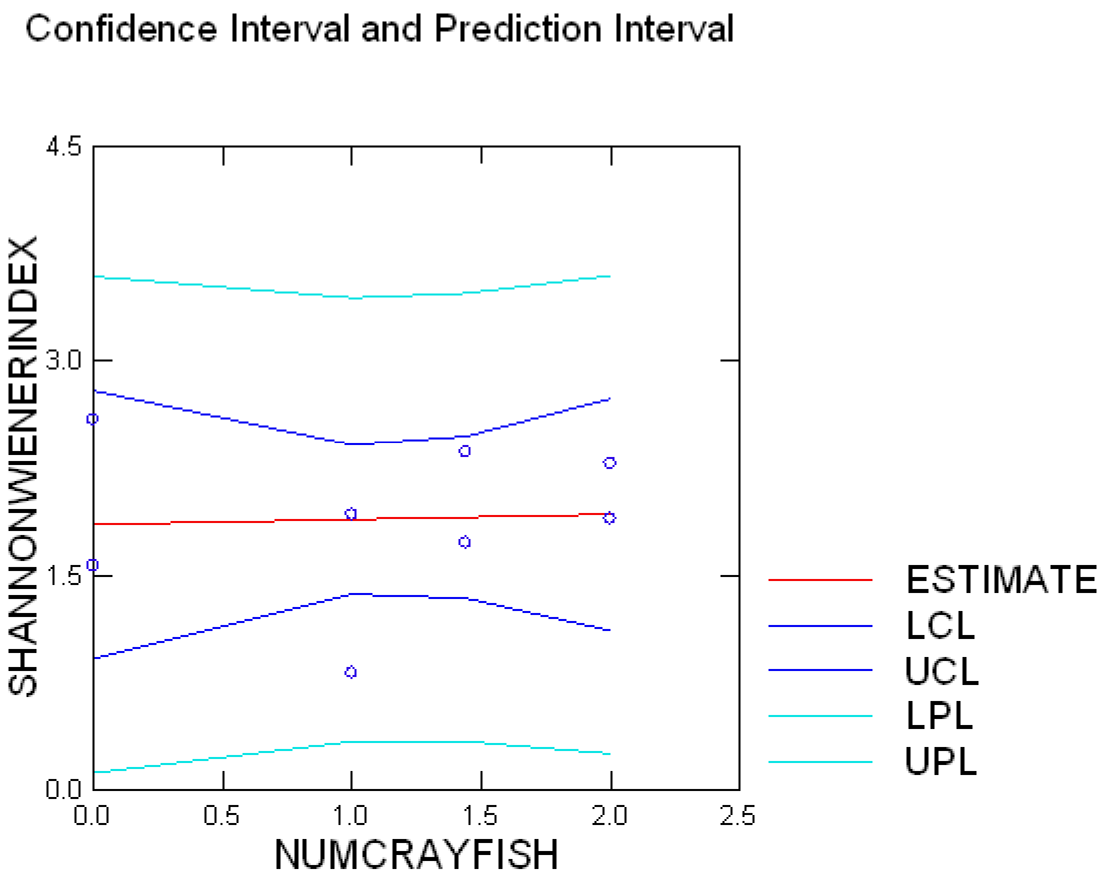

Figure 5: Macroinvertebrate Species Diversity as Measured by the Shannon-Wiener Index at Different Crayfish Abundances in the *In Situ* Plots

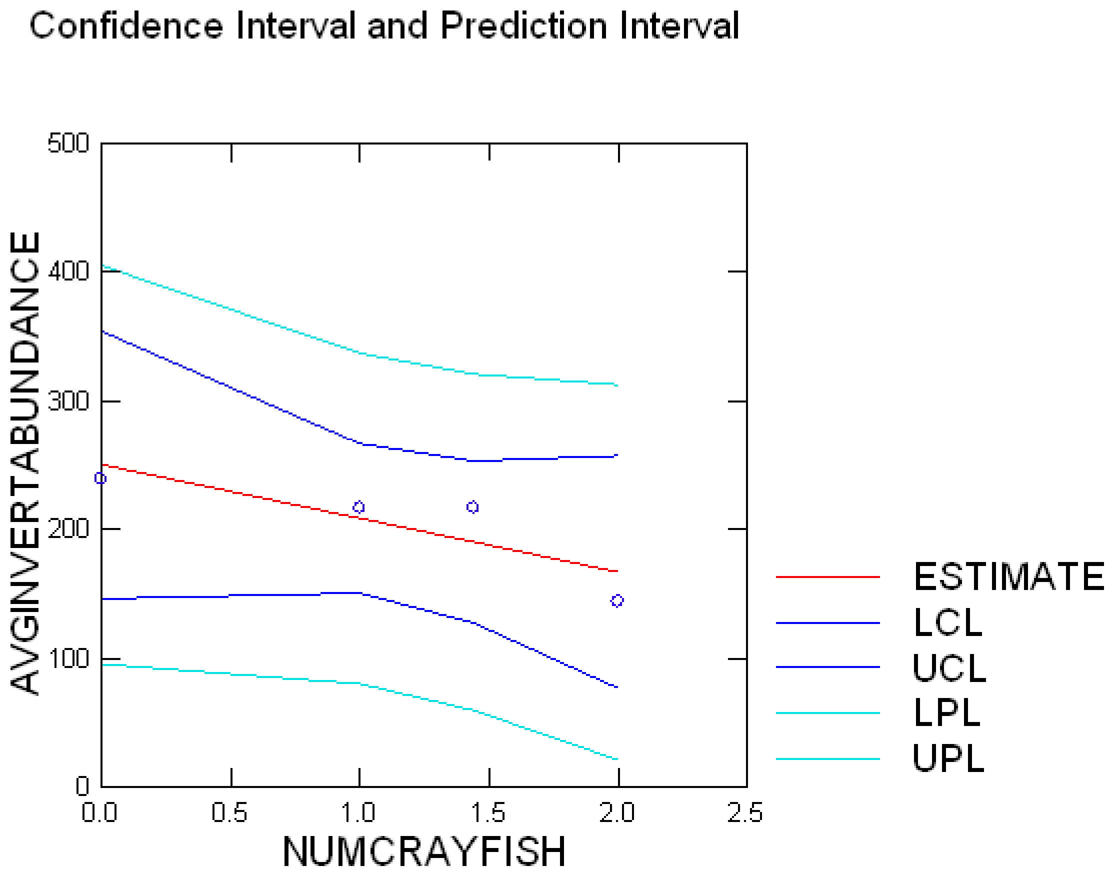

Figure 6: Average Invertebrate Abundance at Different Crayfish Abundances in the Artificial Streams

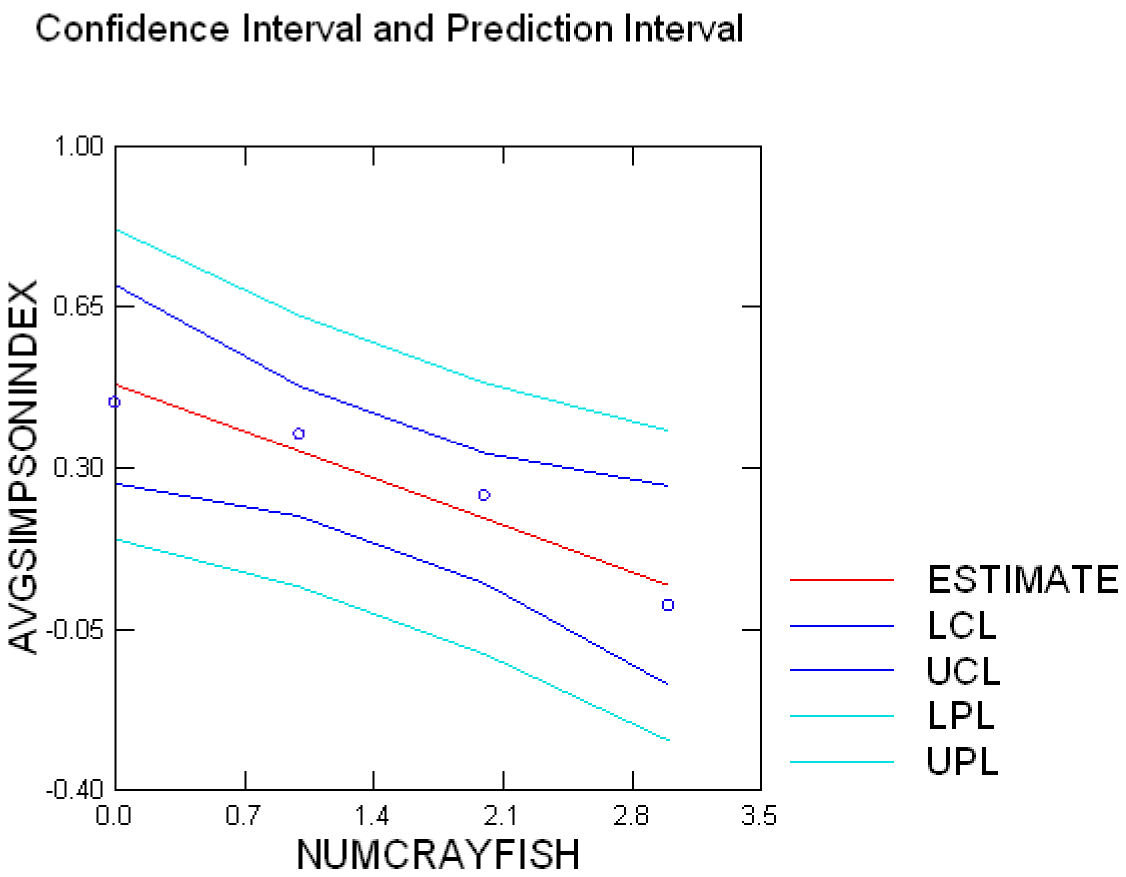

Figure 7: Simpson Index at Different Crayfish Abundances in Artificial Streams

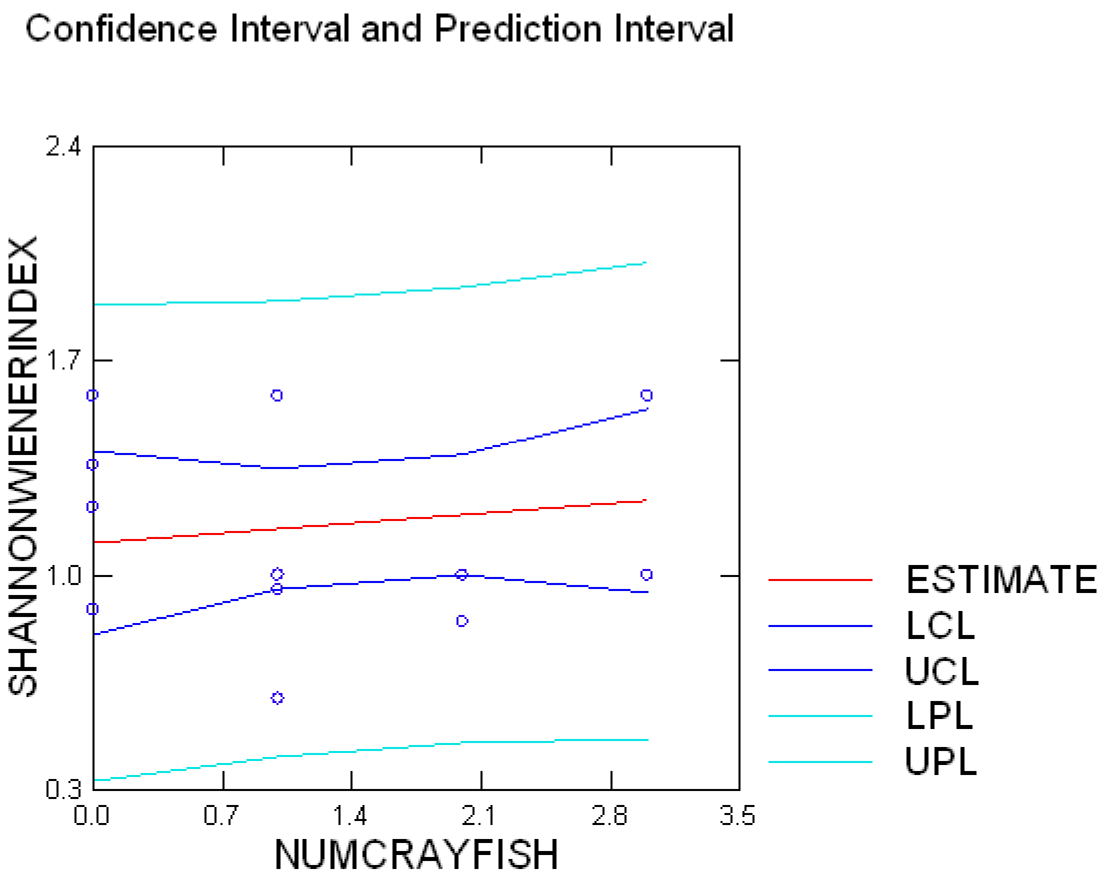

Figure 8: Shannon-Wiener Index by Crayfish Abundance in Artificial Streams

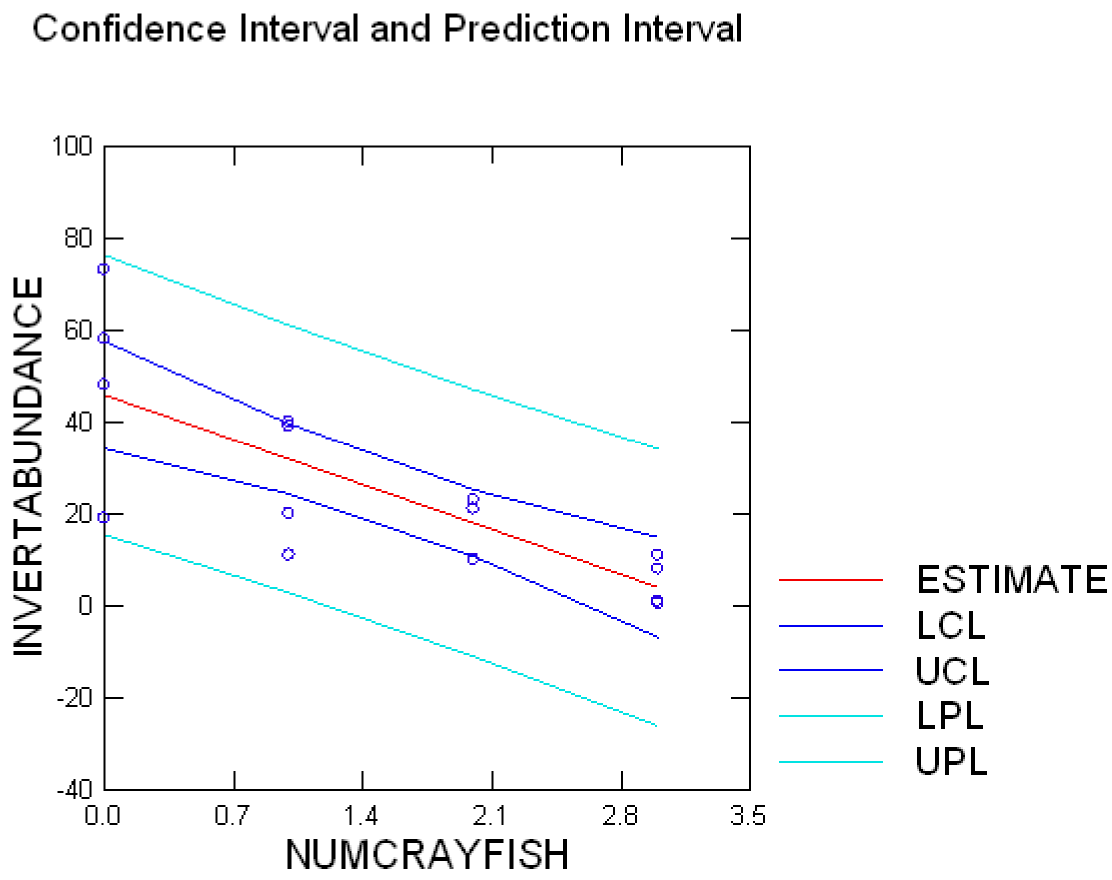

Figure 9: Macroinvertebrate Abundance by Crayfish Abundance in Artificial Streams

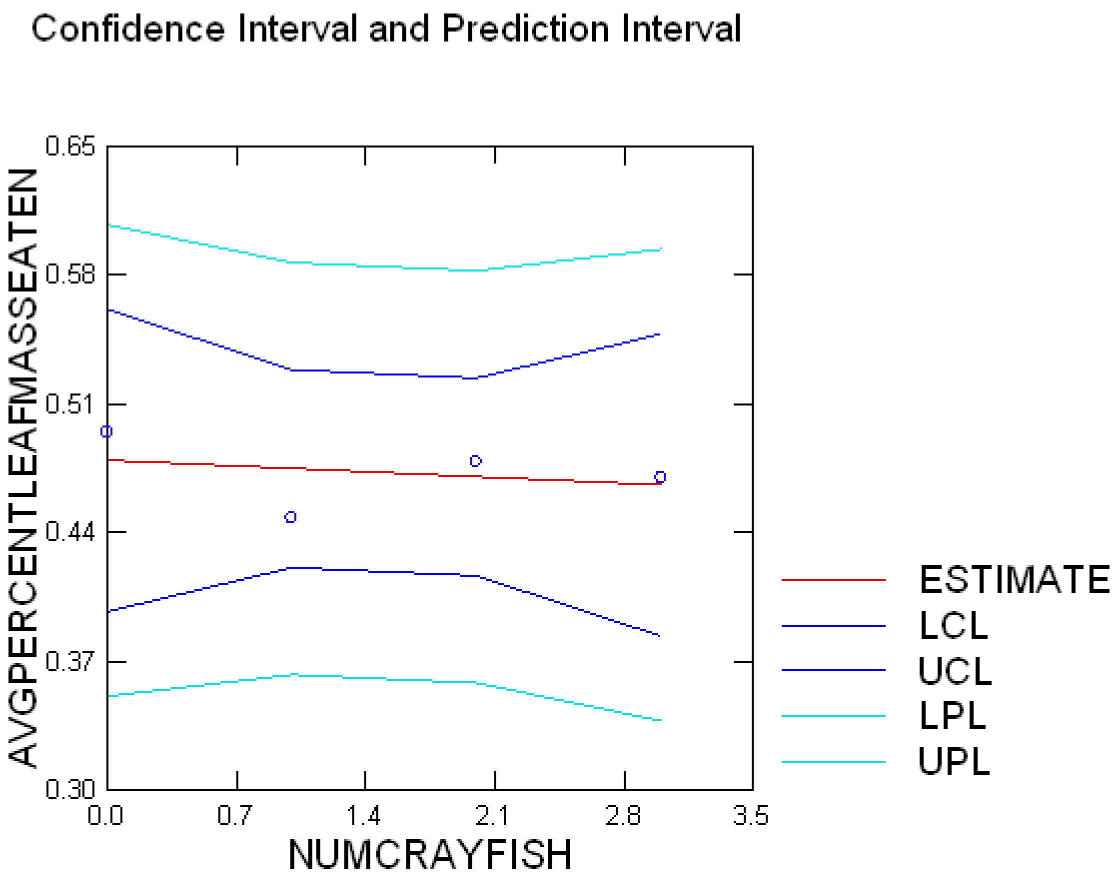

Figure 10: Percentage Maple Leaf Mass Eaten in Different Crayfish Abundances in Artificial Streams

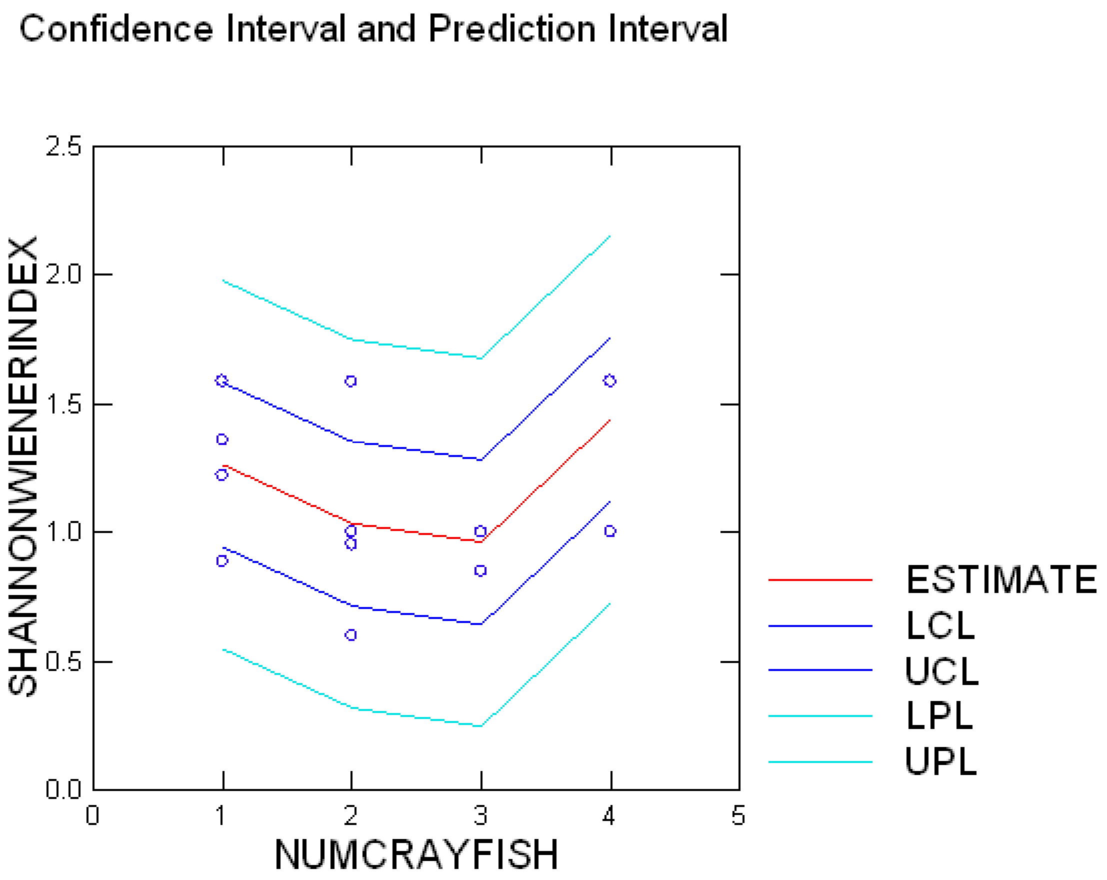

Figure 11: Shannon-Wiener Index in Different Crayfish Abundances in Artificial Streams

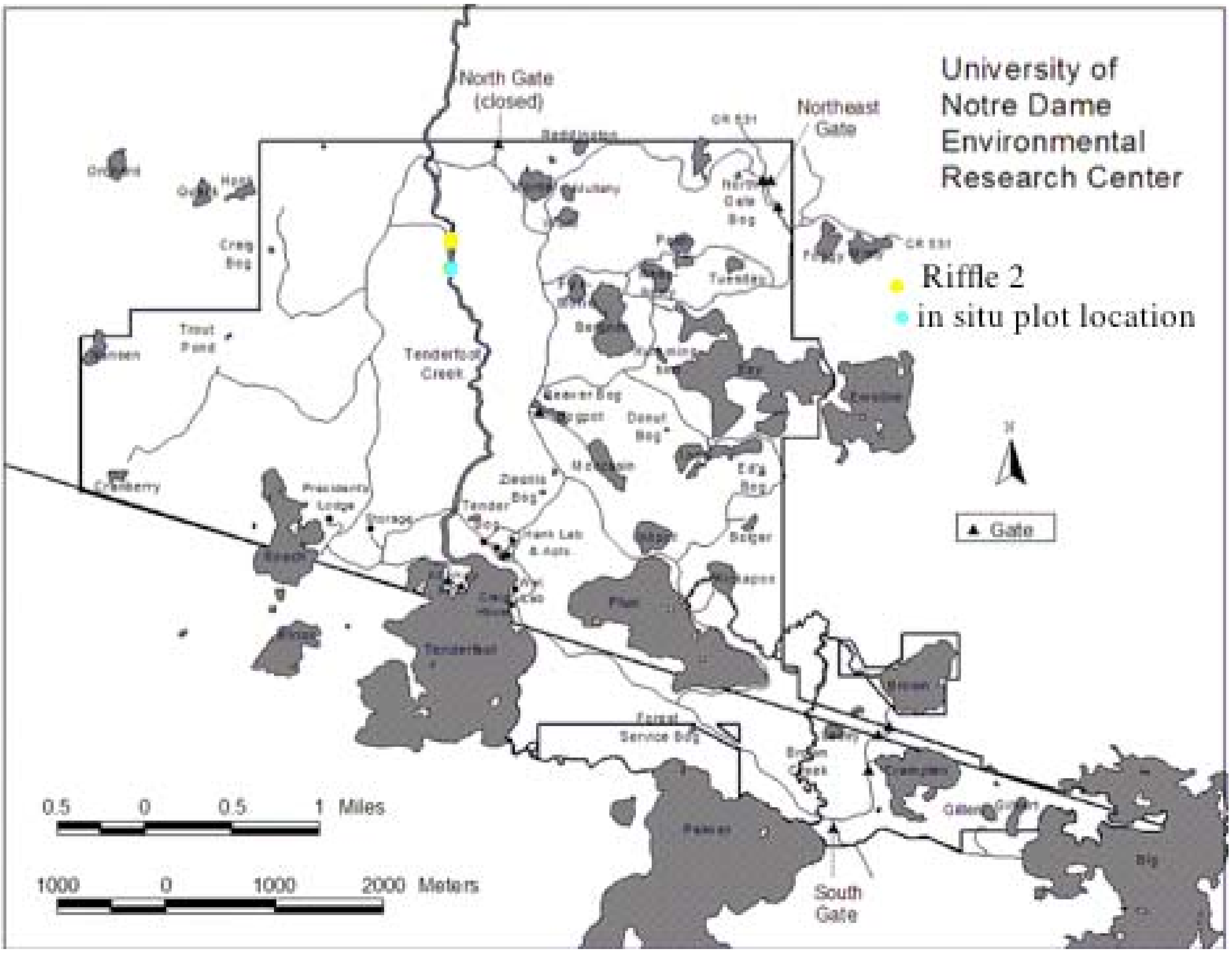

Figure 11: Location of Riffles within the UNDERC Property Involved in the Experiment