Assessing effects of supplemental feeding and Threadfin Shad addition to recreational fishing ponds using stable isotope analysis

by

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Abstract

Pond enhancements are commonly used to increase fish production or enhance angling opportunities, but their effects are often not evaluated. I used stable isotope analysis to estimate the contribution of different rates of pelleted feed to Bluegill *Lepomis macrochirus* reproduction and ultimately to Largemouth Bass *Micropterus salmoides* growth in both the presence and absence of Threadfin Shad *Dorosoma petenense*. Two approaches were used: a controlled small pond experiment and sampling of established ponds. For the controlled small pond experiment I stocked 10 0.1-ha ponds with Bluegill and Largemouth Bass in February of 2012, and fed them one of five feed rates (0, 1.3, 1.9, 3.2, and 4.4 kg – ha^{-1} d^{-1}, 2 ponds per feed treatment). Ponds were sampled through the summer and harvested in August. I also sampled 30 established ponds, 10 with neither Threadfin Shad or pelleted feed, 10 that received only pelleted feed, and 10 that received pelleted feed and contained Threadfin Shad. As expected, Bluegill growth and reproduction increased with increasing feed rates in the small pond experiment. The nitrogen isotopic signatures differed among trophic levels for all species. Bluegill nitrogen isotopic signatures were negatively related with feed rates. Largemouth Bass nitrogen isotopic signatures showed similar trends to those for Bluegill at high feed rates in the controlled small pond experiment. Pelleted feed contributed to the carbon isotopic signatures of both Bluegill and Largemouth Bass across pond types in the established pond study independent of Threadfin Shad presence. Observed effects on growth and

Gonadal investment of Bluegill as well as shifts in isotopic signatures of both Largemouth Bass and Bluegill demonstrated that adding pelleted feed to recreational Largemouth Bass-Bluegill ponds can influence multiple trophic levels; however, the ultimate impact of this enhancement on growth, production, and condition of Largemouth Bass requires further research.

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Introduction

Small impoundments or ponds are abundant throughout the United States, and provide a variety of functions, including pleasure, irrigation, water storage, and stock watering. The most common use of ponds is recreational fishing (USDA 1982; Dauwalter and Jackson 2005; Haley 2009). A small impoundment is generally defined as a reservoir that is less than 40-ha surface area (Dauwalter and Jackson 2005). These water bodies are formed by constructed dams, or as manmade depressions that store water from creeks, springs, runoff, direct precipitation, or wells. As of 2002, there were over 2.6 million privately-owned ponds across the United States, with most located in the Southeastern United States (Smith et al. 2002). According to Swingle (1950), ponds with balanced predator and prey fish populations produce consistent annual harvest. Some pond owners, however, are interested in more than a consistent harvest; size of fish and number of fish harvested are also important as fishing is a recreational activity versus a means of survival. Dauwalter and Jackson (2005) noted that Largemouth Bass *Micropterus salmoides* and Bluegill *Lepomis macrochirus* are the most common predator and prey fish species stocked into small impoundments, and this combination can provide what they termed a "balanced" predator-prey combination under a proper management regime. This combination forms part of the broader pond food web, which with proper management can provide a sustainable fishery (Swingle and Smith 1941; Swingle 1946; Swingle 1950; Modde 1980; Dauwalter and Jackson 2005).

Ponds can be managed for a variety of different purposes including aesthetics, wildlife, hunting, swimming, and recreational fishing for multiple fish species. Ponds can be managed to maximize fish catch, to produce a consistent harvest year after year,

and to produce harvestable or trophy size Bluegill or Largemouth Bass. This is where the predator-prey relationship between largemouth bass and Bluegill plays a vital role. Largemouth Bass is the top predator in such a food web, keeping the Bluegill population from increasing without control. Largemouth Bass also provides a desired fish species for many recreational anglers. Bluegill are important as prey for Largemouth Bass because they are tolerant of a wide range of temperatures, mature early, and can exhibit multiple reproduction events in a year (Swingle and Smith 1941; Haley et al. 2012). Bluegill are also popular and desired by many recreational anglers (Swingle and Smith 1941; Swingle 1946; Swingle 1950; Haley et. al 2012). Traditional pond management techniques such as liming, fertilization, appropriate harvest, and aquatic vegetation management are commonly used to maintain a "balanced" fish population, but additional techniques have been adopted to achieve the goal of maximizing fish density and size (Hampton and Lackey 1976). These enhancement techniques include stocking alternative forage fish species (in addition to Bluegill), installing fish attractors, adding aeration, altering stocking and harvest rates, addition of sterile or genetically enhanced fish, and using supplemental pellet feed to increase caloric intake of desired fish species (Haley et. al 2012).

Fertilization is the one of the most widely practiced fish enhancement techniques. The application of fertilizer in regions with low fertility soils can increase fish standing stock; for example; fertilization in Alabama ponds increased total standing stock from 100 kg-ha^{-1} to 300-400 kg-ha^{-1} (Swingle and Smith 1939; Swingle 1970). Enhancements in addition to fertilization may be implemented to attain a goal of maximized fish density and size (e.g., stocking new prey fish species).

Stocking prey fish is a common method to directly enhance growth and abundance of desired piscivorous fish. Threadfin Shad *Dorosoma petenense* is the most common forage enhancement used in the southeastern United States to increase Largemouth Bass abundance and growth rate (DeVries and Stein 1990). The addition of Threadfin Shad may increase bass production, but can sometimes have a negative effect on Bluegill production via competition for a common food source (DeVries and Stein 1990, 1992; Irwin et al. 2003). In situations where such competition may occur, other management strategies may be considered to compensate for the effects of competition. Providing pelleted feed to enhance Bluegill growth and/or abundance is another popular strategy used to minimize the effects of competition (Berger 1982; Murnyak et al. 1984; Porath and Hurley 2005). Fish feeders installed around ponds provide pelleted feed for fish, and can help to improve condition of stunted fish populations that may be limited by the abundance, type, and caloric value of food (Schalles and Wissing 1976; Berger 1982; Murnyak et al. 1984; Porath and Hurley 2005). Pelleted feed can attract Bluegill into a concentrated area of a pond thereby making them easier to catch by anglers (Berger 1982).

Stable isotope analysis is a technique that has been used to quantify the contribution of different food sources, such as pelleted feed versus natural foods. The approach is useful for quantifying energy flow in ecosystems (Jardine et al. 2003), and is also used to quantify the food source and trophic position of an organism. The ratio of ^{12}C and ^{13}C indicates differences in the source of primary production while the ratio of ^{14}N and ^{15}N indicates differences in the trophic position of an organism (DeNiro and Epstein 1978; Jardine et al. 2003). I used stable isotope analysis to determine the

contribution of pelleted feed to Largemouth Bass via their consumption of both Bluegill that have directly consumed the pelleted feed and Bluegill offspring whose parents consumed the pelleted feed. I also incorporated the presence/absence of Threadfin Shad into the study design to determine the contribution of pelleted feed to Largemouth Bass when Threadfin Shad are present versus when they are not present in the traditional Largemouth Bass and Bluegill food web.

The overall goal of my work was to assess the role of supplemental pellet feed as a recreational fishing enhancement in small impoundments using both established ponds and a controlled small pond experiment. The established ponds identified broad scale trends whereas the controlled small pond experiment identified fine scale mechanistic effects. More specifically my objectives were to:

1. Determine the influence of pelleted feed on Bluegill growth and reproduction as well as Largemouth Bass growth rate.
2. Use stable isotope analysis to estimate the contribution of carbon and nitrogen isotopes from pelleted feed to Bluegill and ultimately to Largemouth Bass.
3. Quantify the influence of Threadfin Shad on pelleted feeding effects in established ponds.
4. Quantify the effects of feeding different rations of pelleted feed on Bluegill and Largemouth Bass material flow.

Methods

Sampling Areas and Locations (Established Ponds)

I sampled ponds with established fish populations at the E.W. Shell Fisheries Center and at private ponds in southern Alabama and western Georgia. I sampled 9 ponds during 2012 and 21 ponds during 2013. I used ponds that did not contain Threadfin Shad, and also did not receive any pelleted feed ("control"; n=3 in 2012; n=7 in 2013), ponds that received pelleted feed, but did not contain Threadfin Shad ("feed only"; n=3 in 2012; n=7 in 2013), and ponds that contained Threadfin Shad, and received pelleted feed ("feed and Threadfin Shad"; n=3 in 2012; n=7 in 2013). All ponds were fertilized. I sampled the first 9 ponds from May through August 2012, and the second set of 21 ponds from April through August 2013.

Sampling Areas and Locations (Controlled Small Pond Experiments)

A small pond experiment was conducted at the E.W. Shell Fisheries Center to determine how the rate of pelleted feeding relates to shifts in ratios of stable isotopes of carbon and nitrogen over a period of five months. The experiment utilized ten 0.1-ha earthen ponds with a maximum depth of 1.5 m.

Sampling Methods (Established Ponds)

I used boat-mounted pulsed-DC electrofishing to collect both Largemouth Bass and Bluegill once at each established pond. I conducted two electrofishing transects at each pond, one centered around the feeder, and the other beginning at least 25 m away from the end of the feeder-centered transect. A subsample of Largemouth Bass (n=10 for each transect) and Bluegill (n=10 for each transect) were euthanized according to the appropriate animal care protocol for further analysis in the lab. The subsample included

individuals from across the size range collected. I measured (nearest mm total length, TL), and weighed (nearest g) all Largemouth Bass and Bluegill >80 mm TL that were not retained.

Temperature and dissolved oxygen (mg/L) were measured at the pond surface and at 1-m in the deep end of each pond (Yellow Springs Instrument Model 51 B). Secchi depth was measured (nearest cm). Surface water samples were collected for chlorophyll *a* and turbidity in 500-ml dark polyethylene bottles, and placed directly on ice. Turbidity was measured with a nephelometer (NTU; HF Scientific, Inc Microw TPW) and chlorophyll *a* concentrations were determined using a flourometer (µg/L; Turner Designs Aquaflour). Water samples (500-ml) were filtered with a 47-mm diameter glass fiber disc (Millipore®). The filters were stored frozen protected from light. Chlorophyll *a* was extracted in cold 95% ethanol for 24h, followed by flourometric analysis (Welschmeyer 1994; Woodard et al. 2013). Two replicate zooplankton samples were collected from each pond using a vertical tow zooplankton net (30-cm diameter; 50-µm mesh) from 1-m to the surface at the deepest point of each pond, and preserved in 90% ethanol for identification under a dissecting microscope. In the laboratory, zooplankton were sub-sampled so that a minimum of 200 of the most common taxa were counted. Cladocerans were identified to genus, copepodite and later instar copepods were identified to family, and nauplii were counted.

Sampling Methods (Controlled Small Pond Experiments)

In February 2012 I drained and refilled ten 0.1–ha ponds located at the E.W. Shell North Auburn Fisheries Experimental Station to ensure they did not contain any fish before stocking. Before refilling the ponds I applied an appropriate amount of

agricultural lime ($MgCaCO_3$) to the pond bottom based on soil test analysis. The agricultural lime increased the buffering capacity or alkalinity of the water which stabilized fluctuations in pH levels throughout the day allowing the fertilizer to induce a phytoplankton bloom by chemically releasing phosphorus bound in the soils (Boyd 1982). I then refilled the ponds through 300µm mesh filters to prevent larval fish from entering each pond, and began fertilizing with 10-34-0 (N-P-K) liquid fertilizer after pond temperatures stabilized above 15.6˚C. This induced a phytoplankton bloom that I maintained at a Secchi depth of approximately 80 cm using ongoing fertilization (Swingle and Smith 1939; Woodard et. al 2013). I stocked 250 50-100 mm Bluegill into each pond in early March 2012. Then in early April 2012 I stocked 25 150 – 250 mm Largemouth Bass into each pond. I randomly assigned one of five feed rates (0, 1.3, 1.9, 3.2, and 4.4 kg – ha^{-1} d^{-1}) of 3 – mm diameter, 36% protein floating catfish feed to each of the ten ponds beginning in mid-March. Each treatment was randomly assigned to two ponds. Stationary fish feeders dispersed the given ration into each pond at 0800 every day to coincide with increased Bluegill feeding activity during the early morning hours. Throughout the duration of the project, I applied herbicide as needed to help prevent and treat excess aquatic vegetation growth. During times of application, I monitored dissolved oxygen levels to determine if aeration was needed (i.e., if the DO level fell below 5.0 mg/l). Long periods of fish exposure to dissolved oxygen levels below 5mg/L can slow fish growth, and exposure to levels below 2mg/L can be lethal (Boyd 1990; Boyd and Boyd 2012).

I collected zooplankton, chlorophyll *a*, and turbidity following the same protocols described above for the established ponds (Table 1). Dissolved oxygen (mg/L) was

measured at the pond surface from the deep end of each pond (Yellow Springs Instrument Model 51 B) (Table 1). Pond temperatures were measured at a depth of 0.5 m in the deep section of five ponds, independent of feed treatment, at two-hour intervals with waterproof Hobo® temperature pendant data loggers (Model UA-002-64) (Table 1). Larval fish samples were collected using an ichthyoplankton larval fish pull net (0.5-m diameter; 500-µm mesh) fitted with a flow meter to allow determination of towing speed and water volume sampled for estimation of larval fish density (Table 1). Two samples were collected from each pond during the monthly sampling period by pulling the ichthyoplankton net the length of each pond twice (Table 1).

Original stock fish were sampled in June (midpoint) and July with boat mounted electrofishing, and all original stock fish were collected in August when the ponds were drained. I measured (nearest mm TL), and weighed (nearest g) all collected Largemouth Bass and Bluegill larger than 80 mm TL. A subsample of both Largemouth Bass and Bluegill was collected from each pond for stable isotope analysis. The subsamples were collected at the midpoint (July electrofishing; n=3 Largemouth Bass from each pond; n=20 Bluegill from each pond), and upon draining in August (all remaining Largemouth Bass; n=20 Bluegill from each pond).

In August, just prior to draining the ponds, I collected potential prey organisms for stable isotope analysis using a sweep net for macroinvertebrates in the vegetation and water column, and an Eckman dredge to collect benthic macroinvertebrate infauna both near feeders and away from feeders if present. I also collected two zooplankton samples from each pond, following the same protocol for the monthly sampling. Immediately

after collection the macroinvertebrate and zooplankton samples were frozen for stable isotope analysis.

Laboratory Processing

All adult fish collected from both the small pond experiment and established ponds were weighed (g), measured (mm, TL), and sexed. Gonads of adult Bluegill from the small pond experiment and established pond sub-samples were weighed (nearest 0.0001g). Saggital otoliths were removed from the Largemouth Bass and Bluegill collected in the established ponds and stored dry for aging. Otoliths that were difficult to read were set in an epoxy base and cross-sectioned using a diamond blade saw, and viewed under oil immersion with a compound microscope (400X magnification). All otoliths were read independently by two readers to ensure accuracy.

Stable Isotope Analysis

Samples of fish muscle tissue from the sub-sampled fish for both the small pond experiment and established ponds were removed and frozen for stable isotope analysis. Samples of pelleted feed from both the small pond experiment and established ponds, zooplankton and macroinvertebrates from the small pond experiment, and a sub-sample of Bluegill ovaries collected at the midpoint (n=5 per pond) and end (n=5 per pond) of the small pond experiment were also frozen. All frozen samples were later dried and homogenized according to standard methods for stable isotope mass spectroscopy (Jardine et al. 2003). A 5 g sample of dorsal muscle tissue was used for large fish, and whole fish were used as samples when 5 g of dorsal muscle tissue could not be extracted. Whole specimens were used as samples on prey items other than fish. The samples were dried to a constant weight at 60°C, homogenized using a mortar and pestle, and dried

again at 60°C to a constant weight. A 0.4 mg sample of the dried samples was packed into a tin capsule and stored in sample preparation trays. Stable isotope data were reported as δX values (where X represents the heavier isotope, either ^{13}C or ^{15}N), or differences from the given standards, expressed in parts per thousand or 0.001 (‰), and calculated according to the formula:

$$\delta X = [(R_{sample}/R_{standard}) - 1] \times 1000$$

where $R_{sample} = {}^{13}C/{}^{12}C$ of the sample, and $R_{standard} = {}^{13}C/{}^{12}C$ of PDB (Pee Dee Belemnite). Similar calculations were performed comparing samples and standards for nitrogen (Walsworth 2011).

Analysis (Established Ponds)

All data were analyzed using R ver. 2.15.2 software (R Development Core Team, 2012) and Statistical Analysis System ver. 9.1.3 (SAS Institute Inc., Cary, North Carolina, USA). Single factor ANOVA was used to test for differences in male and female Bluegill and Largemouth Bass gonadosomatic index (GSI = *ovary weight/body weight)* across treatments in the established ponds to indicate the reproductive allocation (Wootton 1979). When significant overall treatment effects were detected, Tukey post-hoc analysis was used to determine where pair-wise comparisons were statistically significant. Normality and homogeneity of variance were assessed to ensure that the assumptions of ANOVA were met. Male and female Largemouth Bass GSI, Largemouth Bass length at age 2, Largemouth Bass relative weight (with transect as an interaction term), and female Bluegill GSI values were log transformed to meet the assumptions of ANOVA. I used back-calculation from otolith radius measurements to determine length-at-age for both Largemouth Bass and Bluegill in the established ponds. Back-calculated

total length at the *i*th age (TL_i) was estimated using the direct proportion method (Le Cren 1947):

$$L_i = \frac{Si}{Sc} \text{ x } L_{c,}$$

where L_i is the back calculated length of the fish at the formation of the *i*th increment, L_c is the length of the fish at capture, S_c is the radius of the otolith at capture, and S_i is the radius of the otolith at the *i*th increment. Length at age allowed me to relate growth to enhancement effects (i.e., feed, feed and Threadfin Shad). One-way ANOVA was used to test for differences in back-calculated total length at age 2 for Bluegill and Largemouth Bass across pond types in the established ponds. Relative weight (W_r) was quantified to estimate Bluegill and Largemouth Bass condition, defined as the ratio of the actual weight of the fish compared to the published standard weight for that length (Hillman 1982; Henson 1991; Neumann et al. 2012):

$$W_r = \frac{W}{Ws} \text{ x } 100,$$

where W is the mass of an individual and W_s is a standard, length-specific, mass for each species (Murphy et al. 1991). One-way ANOVA was used to test for differences in relative weight of adult Largemouth Bass and Bluegill across pond types in the established ponds.

One-way ANOVA was used to test for differences in $\delta^{15}N$ and $\delta^{13}C$ across pond enhancement types for both Largemouth Bass and Bluegill in the established ponds. Using separate linear mixed effect models (LME) with an interaction term for treatment (pond type) I tested for associations between 1) relative weight and $\delta^{15}N$ and $\delta^{13}C$ for Bluegill, and 2) total length and $\delta^{15}N$ and $\delta^{13}C$ for Largemouth Bass and Bluegill from the established ponds. One-way ANOVA with an interaction term for transect was used

to test for differences in $\delta^{15}N$, $\delta^{13}C$, and relative weight near feeders versus away from feeders for adult Largemouth Bass and Bluegill in established ponds containing supplemental feed, and supplemental feed and Threadfin Shad. I was able to determine a feed rate in each of the established ponds containing supplemental feed, and feed and Threadfin Shad by the following equation:

$$\text{Feed Rate} = (FA/SH)$$

where FA= [(22.68 kg of feed/number of days to feed 22.68 kg of feed through one feeder)*number of feeders] and SH= surface hectares. I used 22.68 kg of feed because that is the weight of a bag of feed typically used by pond owners. This was the best way to get an accurate estimate of feed rates from pond owners in the established ponds (i.e., in terms of how many bags of feed they used in a given length of time). Feed rate is reported as kg – ha^{-1} d^{-1} for each established pond (Table 2). Simple linear regression was then used to test for relationships between feed rate and $\delta^{15}N$ and $\delta^{13}C$ for Bluegill and Largemouth Bass in the established ponds. Mixing model analysis was used to determine the relative contributions of the diet sources for both Largemouth Bass and Bluegill in the established ponds (Jardine et al. 2003).

One-way ANOVA was used to test for differences in temperature, dissolved oxygen, secchi depth, chlorophyll *a*, turbidity, zooplankton density, and pond size across pond enhancement types. Chlorophyll *a*, turbidity, temperature at 1m, zooplankton density, and pond size values were log transformed to meet the assumptions of normality and homogeneity of variance.

Analysis (Controlled Small Pond Experiment)

I used simple linear regression models to test for relationships between feed rate and 1) total weight for Bluegill and Largemouth Bass, 2) male and female Bluegill gonad weight, and 3) male and female Bluegill GSI. Total weight was used in the first model because all fish did not differ in mean size across ponds. Simple linear regression was used to test for relationships between feed rate and $\delta^{15}N$ and $\delta^{13}C$ for Bluegill, Largemouth Bass, and Bluegill ovaries in the small pond experiment. Normality and homogeneity of variance were assessed to ensure that the assumptions of linear regression were met. Mixing model analysis is used in stable isotope analysis when there are two or more sources of energy for growth (Jardine et al. 2003), and was employed in my project to determine the relative contribution of the diet sources for both Largemouth Bass and Bluegill in the small pond experiment.

Single factor ANOVA was used to test for differences in total length and weight of both stock size Largemouth Bass (i.e., ≥ 200 mm TL) and stock size Bluegill (i.e., ≥ 75 mm TL) across feed rates at the beginning of the small pond experiment. I tested for relationships between feed rate and dissolved oxygen, secchi depth, chlorophyll *a* concentration, turbidity, zooplankton density, and larval fish density over the duration of the small pond experiment using separate linear mixed effect models (LME). Chlorophyll *a*, secchi depth, zooplankton density, stock size Largemouth Bass length, stock size Largemouth Bass weight, stock size Bluegill length, and stock size Bluegill weight values were log transformed to meet the assumptions of normality and homogeneity of variance.

Results

Established Ponds

Abiotic Measures, Plankton, and Pond Size

Chlorophyll *a* concentration (one-way ANOVA: $F_{2,27} = 11.58$; $P < 0.01$; Figure 1A) and turbidity (one-way ANOVA: $F_{2,26} = 4.231$; $P = 0.03$; Figure 1B) were significantly greater in the control treatment than the feed only treatment. Neither differed between the feed only and feed and threadfin shad treatments. Turbidity failed homogeneity of variance after log transformation. Secchi depth was significantly lower in the control treatment than the feed only and feed and threadfin shad treatment (one-way ANOVA: $F_{2,27} = 7.046$; $P < 0.01$); which did not differ (Figure 2A). There were no significant differences in surface dissolved oxygen (one-way ANOVA: $F_{2,23} = 1.339$; $P = 0.28$; Figure 3A), dissolved oxygen at 1m (one-way ANOVA: $F_{2,23} = 0.5124$; $P = 0.61$; Figure 3B), and surface temperature (one-way ANOVA: $F_{2,25} = 2.982$; $P = 0.07$; Figure 4A) across treatments. Temperature at 1 m was significantly greater in the feed only and feed and threadfin shad treatments than the control treatment (one-way ANOVA: $F_{2,25} = 3.543$; $P = 0.04$; Figure 4B). The feed only and feed and threadfin shad treatments did not differ. Zooplankton density did not differ across treatments (one-way ANOVA: $F_{2,26} = 2.925$; $P = 0.07$; Figure 2B). The most abundant taxonomic groups were calanoid copepods, copepod nauplii, *Bosmina, Cyclopoid*, and Ostrocods (all were greater than 5% in all of the established ponds). The surface area of the feed and Threadfin Shad treatment ponds were significantly larger than in the control and feed only treatments (one-way ANOVA: $F_{2,27} = 4.992$; $P = 0.01$; Figure 5; Table 2), which did not differ.

Adult Bluegill

Bluegill relative weight (one-way ANOVA: $F_{2,27} = 0.9521$; $P = 0.4$; Figure 6A) and length at age 2 (one-way ANOVA: $F_{2,27} = 1.309$; $P = 0.29$; Figure 6B) did not differ across pond types for the established ponds. Male Bluegill GSI also did not differ across pond types (one-way ANOVA: $F_{2,27} = 0.468$; $P = 0.63$; Figure 7B); however, female Bluegill GSI was significantly lower in the feed only pond type than the control and feed and Threadfin Shad pond types (one-way ANOVA: $F_{2,27} = 4.66$; $P = 0.02$; Figure 7A), which did not differ. When comparing relative weight in transects near feeders versus away from feeders I did not find any significant differences (one-way ANOVA: $F_{3,36} = 0.5549$; $P = 0.65$; Figure 8).

There was no significant difference in Bluegill $\delta^{15}N$ values across pond types (one-way ANOVA: $F_{2,27} = 0.5659$; $P = 0.57$; Figure 9A). Bluegill $\delta^{13}C$ values were significantly more negative in the feed only treatment than the control treatment (one-way ANOVA: $F_{2,27} = 6.14$; $P < 0.01$; Figure 9B), and the feed and Threadfin Shad treatment did not differ from either the control or feed only treatment. Neither Bluegill $\delta^{15}N$ values (one-way ANOVA: $F_{3,36} = 0.6833$; $P = 0.57$; Figure 10A) nor $\delta^{13}C$ values (one-way ANOVA: $F_{3,36} = 0.799$; $P = 0.5$; Figure 10B) differed between near feeder transects versus away from feeder transects in the feed only and feed and Threadfin Shad pond types. There was no significant difference among treatments for Bluegill $\delta^{15}N$ values related to total length (LME: $F_{2,27} = 0.6276$; $P = 0.54$; Figure 11A) and relative weight (LME: $F_{2,27} = 0.645$; $P = 0.53$; Figure 12A), but the interaction effect was significant for both total length (LME: $F_{2,555} = 13.2962$; $P = <0.01$; Figure 11A) (positive interaction) and relative weight (LME: $F_{2,552} = 5.193$; $P = <0.01$; Figure 12A) (negative

interaction). There was a significant difference among treatments for Bluegill $\delta^{13}C$ values related to total length (LME: $F_{2,27} = 6.257$; P = <0.01; Figure 11B) and relative weight (LME: $F_{2,27} = 5.725$; P = <0.01; Figure 12B). Bluegill $\delta^{13}C$ values were more negative in the feed only and feed and Threadfin Shad pond types when compared to the control; however, no significant interaction effect for both total length (LME: $F_{2,555} = 0.427$; P = 0.65; Figure 11B) and relative weight (LME: $F_{2,552} = 1.216$; P = 0.3; Figure 12B) was detected. There was no association between Bluegill $\delta^{15}N$ values and feed rate in the feed only (simple linear regression: $F_{1,8} = 0.0237$; P = 0.88; $R^2 = -0.1217$; Figure 13A) and feed and Threadfin Shad (simple linear regression: $F_{1,8} = 1.505$; P = 0.26; $R^2 = 0.0532$; Figure 14A) pond types. Similarly $\delta^{13}C$ values in the feed only (simple linear regression: $F_{1,8} = 0.3152$; P = 0.59; $R^2 = -0.0824$; Figure 13B) and feed and Threadfin Shad (simple linear regression: $F_{1,8} = 1.398$; P = 0.27; $R^2 = 0.0423$; Figure 14B) pond types were unrelated to feed rate.

Adult Largemouth Bass

Adult Largemouth Bass relative weight (one-way ANOVA: $F_{2,27} = 0.3712$; P = 0.69; Figure 15A), female GSI (one-way ANOVA: $F_{2,27} = 1.152$; P = 0.33; Figure 16A), and male GSI (one-way ANOVA: $F_{2,27} = 0.155$; P = 0.98; Figure 16B) did not differ across pond types. Male GSI failed to meet the homogeneity of variance assumption even after log transformation. I found that length at age 2 was significantly greater for Largemouth Bass in the feed and threadfin shad treatment versus in the control treatment (one-way ANOVA: $F_{2,27} = 3.968$; P = 0.03; Figure 15B), while feed only treatment did not differ from either the control or feed and Threadfin Shad treatments. Adult Largemouth Bass relative weight did not differ between near feeder transects versus away

from feeder transects in the feed only and feed and Threadfin Shad pond types (one-way ANOVA: $F_{3,36} = 0.2348$; P = 0.87; Figure 17).

There was no significant difference in Largemouth Bass $\delta^{15}N$ values across pond types (one-way ANOVA: $F_{2,27} = 0.851$; P = 0.44; Figure 18A). Largemouth Bass $\delta^{13}C$ values were significantly more negative in the feed only and feed and Threadfin Shad treatments versus the control treatment (one-way ANOVA: $F_{2,27} = 5.784$; P < 0.01; Figure 18B). Largemouth Bass $\delta^{13}C$ values did not differ between feed only and feed and Threadfin Shad treatments. Neither $\delta^{15}N$ values (one-way ANOVA: $F_{3,36} = 0.8939$; P = 0.45; Figure 19A) nor $\delta^{13}C$ values (one-way ANOVA: $F_{3,36} = 0.2003$; P = 0.87; Figure 19B) differed for adult Largemouth Bass between near feeder transects versus away from feeder transects in the feed only and feed and Threadfin Shad pond types. There was no significant difference among treatments for Largemouth Bass $\delta^{15}N$ values related to total length (LME: $F_{2,27} = 1.123$; P = 0.34; Figure 20A), but the interaction effect was a significant positive association (LME: $F_{2,565} = 6.729$; P = <0.01; Figure 20A). There was a significant difference among treatments for $\delta^{13}C$ values related to Largemouth Bass total length (LME: $F_{2,27} = 5.8678$; P = <0.01; Figure 20B) with more negative $\delta^{13}C$ values in the feed only and feed and Threadfin Shad pond types compared to the control. The interaction effect was a significant positive association for both the feed only and control pond types, and a significant negative association for the feed and Threadfin Shad pond type (LME: $F_{2,565} = 4.8308$; P = <0.01; Figure 20B). Although Largemouth Bass $\delta^{15}N$ values and feeding rates were not related in the feed only pond type (simple linear regression: $F_{1,8} = 0.3811$; P = 0.55; $R^2 = -0.0737$; Figure 21A); they were positively related in the feed and Threadfin Shad pond type (simple linear regression: $F_{1,8} = 8.778$;

P = 0.02; R^2 = 0.4636; Figure 22A). In contrast, Largemouth Bass $\delta^{13}C$ values and feed rate were positively related in the feed only pond type (simple linear regression: $F_{1,8}$ = 5.31; P = 0.05; R^2 = 0.3238; Figure 21B); while being unrelated in the feed and Threadfin Shad pond type (simple linear regression: $F_{1,8}$ = 2.871; P = 0.13; R^2 = 0.1721; Figure 22B).

Controlled Small Pond Experiment

Abiotic Measures, Plankton, Larval Bluegill, and Stock Size Largemouth Bass and Bluegill

Chlorophyll *a* concentration (Figure 23A), turbidity (Figure 23B), secchi depth (Figure 24A), dissolved oxygen (Figure 27), and larval Bluegill density (Figure 26) all did not differ among treatments in the small pond experiment (Table 3). Zooplankton density also did not differ among treatments (Figure 24B; Table 3), although these data failed a test for normality even after log transformation. The most abundant zooplankton groups (i.e. those that were 5% or greater in all of the ponds over the duration of the experiment) were calanoid copepods, copepod nauplii, *Bosmina, Ceriodaphnia,* and *Cyclopoid*. Temperature was collected in five ponds regardless of feed rates. The temperatures never varied more than 1°C, so probably had no effect on the Largemouth Bass and Bluegill (Figure 27). There were no significant differences among feeding rates in total length (one-way ANOVA: $F_{4,5}$ = 0.0774; P = 0.99; Figure 28A) and total weight for stock size Bluegill (one-way ANOVA: $F_{4,5}$ = 0.081; P = 0.99; Figure 28B), or for total length (one-way ANOVA: $F_{4,5}$ = 0.087; P = 0.98; Figure 29A) and total weight (one-way ANOVA: $F_{4,5}$ = 0.027; P = 0.99; Figure 29B) for stock size Largemouth Bass at the

beginning of the experiment. Length and weight for both stock size Largemouth Bass and Bluegill failed for homogeneity of variance, even after log transformation.

Adult Bluegill and Bluegill Ovaries

Bluegill total weight at the experiment's midpoint (simple linear regression: $F_{1,8} = 17.47$; $P < 0.01$; $R^2 = 0.6467$; Figure 30A), in July (simple linear regression: $F_{1,8} = 22.95$; $P < 0.01$; $R^2 = 0.7092$; Figure 30B), and at the end (simple linear regression: $F_{1,8} = 8.34$; $P = 0.02$; $R^2 = 0.4492$; Figure 30C) showed a significant positive association with feed rate. Female Bluegill GSI (simple linear regression: $F_{1,7} = 0.2637$; $P = 0.62$; $R^2 = -0.1014$; Figure 31A) and gonad weight (simple linear regression: $F_{1,7} = 0.217$; $P = 0.21$; $R^2 = -0.1085$; Figure 32A) did not differ at the midpoint; however, at the end of the experiment both female Bluegill GSI (simple linear regression: $F_{1,8} = 8.636$; $P = 0.02$; $R^2 = 0.459$; Figure 31B) and gonad weight (simple linear regression: $F_{1,8} = 11.86$; $P < 0.01$; $R^2 = 0.5469$; Figure 32B) were positively related to feed rate. Male Bluegill GSI did not differ across feed rates at the midpoint (simple linear regression: $F_{1,7} = 0.5681$; $P = 0.48$; $R^2 = -0.0571$; Figure 33A) or end of the experiment (simple linear regression: $F_{1,8} = 4.557$; $P = 0.07$; $R^2 = 0.2833$; Figure 33B); however, male Bluegill gonad weight was positively related to feed rate at both the midpoint (simple linear regression: $F_{1,7} = 5.5751$; $P = 0.05$; $R^2 = 0.3726$; Figure 34A) and end (simple linear regression: $F_{1,7} = 29.08$; $P < 0.01$; $R^2 = 0.7573$; Figure 34B) of the controlled small pond experiment.

Bluegill $\delta^{15}N$ values were negatively associated with feed rate at both the midpoint (simple linear regression: $F_{1,8} = 16.43$; $P < 0.01$; $R^2 = 0.6316$; Figure 35A) and end (simple linear regression: $F_{1,8} = 10.78$; $P = 0.01$; $R^2 = 0.5207$; Figure 35B) of the experiment; however, bluegill $\delta^{13}C$ values did not differ across feed rates at either the

midpoint (simple linear regression: $F_{1,8} = 0.052$; $P = 0.825$; $R^2 = -0.1177$; Figure 36A) or end (simple linear regression: $F_{1,8} = 0.2241$; $P = 0.65$; $R^2 = -0.0944$; Figure 36B) of the small pond experiment. Bluegill ovary $\delta^{15}N$ values did not differ across feed rates at either the midpoint (simple linear regression: $F_{1,7} = 2.837$; $P = 0.14$; $R^2 = 0.1868$; Figure 38A) or end (simple linear regression: $F_{1,8} = 1.48$; $P = 0.21$; $R^2 = 0.0854$; Figure 37B) of the experiment. Similarly, Bluegill ovary $\delta^{13}C$ values did not differ at either the midpoint (simple linear regression: $F_{1,7} = 0.4617$; $P = 0.52$; $R^2 = -0.0721$; Figure 38A) or end (simple linear regression: $F_{1,8} = 0.344$; $P = 0.57$; $R^2 = -0.0786$; Figure 38B) of the small pond experiment.

Adult Largemouth Bass

Largemouth Bass total weight and feed rate were not related at the midpoint (simple linear regression: $F_{1,8} = 3.673$; $P = 0.09$; $R^2 = 0.229$; Figure 39A), in July (simple linear regression: $F_{1,8} = 0.0924$; $P = 0.77$; $R^2 = -0.1122$; Figure 39B), or at the experiment's end (simple linear regression: $F_{1,8} = 0.5871$; $P = 0.47$; $R^2 = -0.0481$; Figure 39C). Adult Largemouth Bass $\delta^{15}N$ values was not related to feeding rate at either the midpoint (simple linear regression: $F_{1,8} = 0.037$; $P = 0.85$; $R^2 = -0.1198$; Figure 40A) or at the experiment's end (simple linear regression: $F_{1,8} = 2.711$; $P = 0.12$; $R^2 = 0.1597$; Figure 40B). And similarly, Largemouth Bass $\delta^{13}C$ values were also not related to feed rate at either the midpoint (simple linear regression: $F_{1,8} = 0.0587$; $P = 0.82$; $R^2 = -0.1168$; Figure 41A) or at the experiment's end (simple linear regression: $F_{1,8} = 0.6162$; $P = 0.46$; $R^2 = -0.0445$; Figure 41B).

Discussion

Food availability and quality are important in determining fish size differences among populations in ponds (Hewett and Kraft 1993); this is why supplemental feed and stocking Threadfin Shad have become popular strategies for managing small impoundments. Supplemental feed has a higher caloric density versus natural foods (Schalles and Wissing 1976; Porath and Hurley 2005), and the expectation is that bluegill feeding on pelleted feed will spawn more often in a season or produce more eggs at each spawn, thus providing additional food for Largemouth Bass (Woodard et. al 2013). Relative to Threadfin Shad stocking, Largemouth Bass may shift their diet to feed on Threadfin Shad when present (Davies et al. 1979; Noble 1981), and this diet shift may alter the effects of pelleted feed on the predator – prey dynamic.

Between my small pond experiment and the established ponds I found that Bluegill total weight, gonad weight, and gonadosomatic indices increased with feed rate. Mixing models for stable isotope analysis are useful in determining the relative contributions of different food sources to an animal's diet (Hobson 1999; Phillips 2001). While it is relatively straightforward to use stable isotope ratios to evaluate food web structure and material flow within a single ecosystem (Post 2002), it can be very difficult across multiple ecosystems due to considerable variation among ecosystems in the $\delta^{13}C$ and $\delta^{15}N$ of the base of the food web from which organisms draw their nitrogen and carbon (Rounick and Winterbourn 1986; Zohary et al. 1994; Cabana and Rasmussen 1996; MacLeod and Barton 1998; Kitchell et al. 1999, VanderZanden and Rasmussen 1999; Post 2002). Due to limiting factors I was not able to estimate suitable $\delta^{13}C_{base}$ and $\delta^{15}N_{base}$ for each individual pond in both the established ponds and the controlled small

pond experiment making the mixing models more difficult to interpret; however, the $\delta^{13}C$ and $\delta^{15}N$ values of Largemouth Bass, Bluegill, and prey items collected are sufficient to derive the effects of pelleted feed and Threadfin Shad to the top of the food chain without the use of mixing models. Stable isotope analysis provided evidence that pelleted feed can affect $\delta^{13}C$ and $\delta^{15}N$ signatures of Bluegill and Largemouth Bass (Figure 42). Threadfin Shad did not affect the $\delta^{13}C$ and $\delta^{15}N$ signatures of Bluegill and Largemouth Bass in the presence of pelleted feed; however, Largemouth Bass had increased growth rates at age 2 in the presence of Threadfin Shad and pelleted feed.

Abiotic Measures and Plankton

All of my established ponds were fertilized. The use of phosphorus and nitrogen fertilizers is a common practice to stimulate planktonic algae blooms, thus increasing the productivity up the entire food web (Swingle and Smith 1939; Woodard 2011). Depending on the natural fertility of pond soils, fertilization can increase fish biomass three to four-fold (Swingle 1950; Haley et. al 2012) by increasing plankton production (Swingle and Smith 1939). I found chlorophyll *a* concentration and turbidity to be higher in control pond types versus both feeder pond types with significant differences between the control and feed only ponds. Similarly, I found the opposite for secchi depth with control ponds having significantly lower secchi depths than the feeder ponds. Zooplankton densities did not differ among the three established pond types. Although all ponds were fertilized there are several reasons that could explain these differences. First, desired fertilizer concentrations may have been easier to obtain in the control ponds because they were smaller than the feeder pond types. Second, owners of the control ponds were only concentrating efforts on fertilizing while owners of the feed only and

feed and Threadfin Shad pond types distributed efforts among fertilizing, feeding, stocking, and maintaining Threadfin Shad; as such they may not have put forth equal effort to maintain a regular fertilizing routine. Third, I only sampled each pond once such that I obtained a snapshot of these measures. Research indicates that Threadfin Shad can dramatically reduce zooplankton density (Ziebell et al. 1986; Prophet 1988; DeVries et al. 1991; Garvey and Stein 1998); however, zooplankton density in the feed only and feed and threadfin shad pond types from the established ponds did not differ. A possible explanation is the research associated with Threadfin Shad and zooplankton densities was conducted in larger reservoirs, and my research was in small fertile impoundments. This would suggest that fertilizing may affect zooplankton density more than the presence of Threadfin Shad in these small systems during summer months. Dissolved oxygen and surface temperature did not differ across pond types; however temperature at 1m was lower in the control ponds than my feeder ponds by an average of no more than 3.5°C, so therefore, probably had no effect on the Largemouth Bass and Bluegill. This is because some of the control ponds were sampled early in the two summers when surface temperatures may have been high, but temperatures at 1 m may have been lower than the ponds containing feed that were sampled later in the summer. Some of the feeder pond types were purposely sampled later in the summer to allow a longer time during which feed was present in the feeder pond types before my sampling.

In the controlled small pond experiment, there were no differences in chlorophyll *a* concentration, turbidity, secchi depth, zooplankton density, and dissolved oxygen across feed rate treatments. This suggests that any differences in growth and reproduction for Largemouth Bass and Bluegill are related to pelleted feed rates. I found

substantial variation within each feed treatment for chlorophyll *a* concentration, turbidity, and secchi depth. Because the ratio of ^{12}C to ^{13}C indicates differences in the source of primary production (DeNiro and Epstein 1978; Jardine et al. 2003), variability of chlorophyll *a* concentration, turbidity, and secchi depth may have altered the $\delta^{13}C$ isotopes of Largemouth Bass and Bluegill without regard to feed rate.

Bluegill

According to previous research, harvest of Largemouth Bass and Bluegill can affect size structure of both species in small impoundments (Swingle 1946; Gabelhouse 1987; Coble 1988; Guy and Willis 1990). In the established ponds I was generally able to control for fertilization, presence/absence of alternative forage species, and pelleted feeding. Similar to Haley et. al (2012), I was not able to control for or precisely measure fish harvest in the established ponds due to inconsistency in keeping harvest records; therefore, I did not necessarily expect to see strong differences in the size structure of Bluegill due to pelleted feed in the established ponds. There were no differences in relative weight or in length at age 2 across pond types suggesting that fish harvest may play an important role when it comes to determining bluegill size structure regardless of other management strategies. Previous research suggests using caution when estimating annual growth from any annuli other than the most recent (Gutreuter 1987; Walsworth et al. 2013); however, due to a small sample size of Bluegill (20 fish) that I was allowed to take from each established pond I had to use length at age 2. I also did not detect any differences in relative weight near the feeders versus away from the feeders in the feed only and feed and Threadfin Shad pond types, suggesting that fish are moving enough such that feeders affect the whole pond (Berger 1982). Because fecundity is often related

to fish size (Wooton 1979; Fletcher and Wooton 1995; Woodard et. al 2013), and I was not able to control for fish harvest in the established ponds, I did not expect to see any differences in male and female GSI among treatments from the established ponds. While there were no differences in male GSI across pond types; I did find female GSI to be significantly lower for the feed only pond type when compared to the control and feed and Threadfin Shad pond types. I believe that this difference is likely a consequence of a lack of control of fish harvest versus an effect of pelleted feeding.

The nitrogen and carbon composition of an animal reflects the nitrogen and carbon composition of its diet (DeNiro and Epstein 1978; DeNiro and Epstein 1981). I expected pelleted feed to affect Bluegill $\delta^{15}N$ and $\delta^{13}C$ values in the established ponds containing pelleted feed versus control ponds. The $\delta^{15}N$ values did not differ across pond types; however, $\delta^{13}C$ values were lower in the established ponds containing pelleted feed when compared to the control ponds suggesting that pelleted feed affects Bluegill $\delta^{13}C$ signatures. Berger (1982) reported that Bluegill movement in a study lake with pelleted feeders was considerable, with some moving as far as 457 m. This is reflected in the established ponds containing pelleted feed because $\delta^{15}N$ and $\delta^{13}C$ values of Bluegill did not differ near the feeders versus away from the feeders, suggesting that Bluegill may move considerable distances to consume pelleted feed in small impoundments. I expected a negative relationship with $\delta^{15}N$ and $\delta^{13}C$ values versus total fish length and relative weight in the feed only and feed and threadfin shad pond types because $\delta^{15}N$ values of pelleted feed are lower than those of natural prey items and previous studies have demonstrated pelleted feed to increase Bluegill growth (Schmittou 1969; Berger 1982; Woodard et. al 2013); therefore, I expected the larger Bluegill to have consumed

more pelleted feed than smaller Bluegill. I found a significant positive association between fish length and $\delta^{15}N$ values, and a significant negative association with $\delta^{13}C$ values for all three pond types with $\delta^{13}C$ values being significantly lower in the feed only and feed and Threadfin Shad pond types when compared to the control. This demonstrates that both the $\delta^{15}N$ and $\delta^{13}C$ values of bluegill are not affected by fish length in the presence/absence of pelleted feed and pelleted feed and Threadfin Shad; however, pelleted feed effected the overall $\delta^{13}C$ values because bluegill $\delta^{13}C$ values were more negative in the presence of pelleted feed and pelleted feed and Threadfin Shad when compared to the control. There was a significant negative association with $\delta^{15}N$ values for all pond types when compared to relative weight; however the treatments did not differ suggesting pelleted feed and pelleted feed and Threadfin Shad did not have an effect on the $\delta^{15}N$ value and relative weight relation. There was no association with Bluegill $\delta^{13}C$ values and relative weight; however, the $\delta^{13}C$ values were more negative in the presence of pelleted feed and pelleted feed and Threadfin Shad. Finally, the $\delta^{15}N$ and $\delta^{13}C$ values were not influenced by feed rate in the established ponds, likely because most of the feed rates were well below recommended feed rates. Only two out of the twenty established ponds were fed pelleted feed rates above 2 kg – ha^{-1} d^{-1}. As such the gradient between fed and non-fed ponds was likely not adequate to detect differences in $\delta^{15}N$ and $\delta^{13}C$ values.

In the controlled small pond experiment, I expected Bluegill growth rates to increase with increasing feed rates (Schmittou 1969; Berger 1982; Woodard et. al 2013). Throughout the small pond experiment, there was a significant positive relationship between total weight across and feed rate. Woodard et. al (2013) found that bluegill had

an increased GSI in the presence of pelleted feed due to Bluegills attaining a larger size in high-ration treatments. Similarly, I found that pelleted feed increased female GSI across increasing feed rates. Absolute gonad weight was measured because GSI may not be a reliable indicator of energy allocated to reproduction for a protracted spawning species such as Bluegill (Fox and Crivelli 1998; Woodard 2011). I found that pelleted feeding had a positive relationship on absolute gonad weight for both females and males at the midpoint and end of the controlled small pond experiment. These results indicate that pelleted feed increased growth rate of Bluegill within a 3-month period of stocking that continued throughout the experiment. Pelleted feed also caused Bluegill to increase the energy they allocated to reproduction; however, surprisingly this did not result in differences in larval Bluegill density across feed treatments. This contradicts previous studies stating that increased Bluegill gonad weight should correspond with an increase in the number of eggs spawned (Wootton 1979; Roff 1984; Fletcher and Wootton 1995; Woodard et. al 2013). It is possible that the monthly sampling for larval Bluegill was not adequate to detect differences among feed treatments. Woodard et al. (2013) found similar results during one year of a similar controlled experiment and attributed the finding to density dependence.

Filbrun and Culver (2013) found that in low feed treatments (3% and 1% BW/d), $\delta^{15}N$ values of channel catfish shifted toward $\delta^{15}N$ values of pelleted feed while $\delta^{13}C$ values did not differ after a 2-month exposure period. Similar to their findings, I found a significant negative relationship with $\delta^{15}N$ values related to increasing feed treatments at both the midpoint and the end of the controlled small pond experiment while the $\delta^{13}C$ values did not differ across feed rates at both the midpoint and the end of the experiment.

The $\delta^{15}N$ values of animals are usually more positive than those of their diets (DeNiro and Epstein 1981) suggesting that the significant negative relationship with the $\delta^{15}N$ values is related to Bluegill in the higher feed rates consuming more pelleted feed because the $\delta^{15}N$ values of pelleted feed are lower than those of natural prey items. It is possible that we were able to detect differences in $\delta^{15}N$ values from the small pond experiment, and not in the established ponds because established ponds were generally fed at very low feed rates (18 out of 20 established ponds fed below 2 kg – ha^{-1} d^{-1}) compared to the feed rates from the small pond experiment (up to 4.4 kg – ha^{-1} d^{-1}). DeNiro and Epstein (1978) state that it is possible to perform dietary analysis based on the determination of the $^{13}C/^{12}C$ ratio of animal carbon. This makes it difficult to understand why we were able to differentiate ponds containing feed versus control ponds based on Bluegill $\delta^{13}C$ values in the established ponds but not in the controlled small pond experiment. It is possible that the high variation in chlorophyll *a* concentration and zooplankton densities within treatments from the small pond experiment may have prevented detection of any differences in $\delta^{13}C$ values across pelleted feed treatments. Finally, there was no relationship in the $\delta^{15}N$ and $\delta^{13}C$ values for Bluegill ovaries across increasing feed rates suggesting that the sources of primary production as a result from the pelleted feed were not integrated directly into Bluegill ovaries; however, negative trends in $\delta^{15}N$ values of bluegill ovaries with increasing feed rate reflect the possibility of a relationship requiring more replicates or a longer duration of the controlled small pond experiment.

Largemouth Bass

In the established ponds there were no differences in Largemouth Bass relative weight, male GSI, or female GSI across pond types, likely due to a lack of control over fish harvest. Condition factors such as relative weight vary primarily due to environmental factors (Springer and Murphy 1990), and may be cautiously used as a working index of prey availability (Liao et al. 1995). Environmental factors and prey availability may vary annually; whereas length at age 2, as applied in my research, measures growth over multiple years. Length at age 2 was significantly greater for the feed and Threadfin Shad pond type versus the control pond type. Threadfin Shad have a higher caloric density then some other prey species including Bluegill (Wright and Kraft 2012). In response to a Threadfin Shad introduction in Lake Powell, Arizona-Utah, smaller Largemouth Bass (<324 mm) changed their diets from 78% centrarchids (volume) to 97% Threadfin Shad (volume) (May and Thompson 1974; Wydoski and Bennett 1981), and in larger systems, Threadfin Shad can enhance Largemouth Bass growth (Tharratt 1966; Miller 1971; von Geldern and Mitchell 1975; Haley et. al 2012). This provides support for Threadfin Shad contributing to greater Largemouth Bass length at age 2 in the established ponds. There were no differences in relative weight near feeders versus away from feeders in the feed only and feed and Threadfin Shad pond types suggesting that Largemouth Bass condition was not spatially limited by feeders.

In the established ponds, $\delta^{15}N$ values did not differ across pond types; however, $\delta^{13}C$ values were significantly lower in the established ponds containing pelleted feed when compared to the control ponds, suggesting that pelleted feed affects Largemouth Bass $\delta^{13}C$ values. This suggests that the lower $\delta^{13}C$ values of the Bluegill translated into

lower Largemouth Bass $\delta^{13}C$ values when feed was present. Both $\delta^{15}N$ and $\delta^{13}C$ values did not differ near feeders versus away from feeders in the feed only and feed and Threadfin Shad established ponds. With previous research suggesting considerable Bluegill movement in the presence of pelleted feed (Berger 1982; See Results), I did not expect to find any differences in $\delta^{15}N$ and $\delta^{13}C$ values near feeders versus away from feeders for Largemouth Bass.

There were no treatment effects with $\delta^{15}N$ values versus total fish length; however, a significant positive interaction in all pond types. The control pond type had a more positive slope than the feed only and feed and Threadfin Shad pond type, suggesting that larger sized Largemouth Bass were consuming Bluegill directly consuming pelleted feed, or the offspring of the Bluegill directly consuming pelleted feed in the ponds containing pelleted feed. There was a significant difference in pond types for $\delta^{13}C$ values versus total fish length with more negative $\delta^{13}C$ values in the feed only and feed and Threadfin Shad pond types. There was also a significant positive interaction with $\delta^{13}C$ values and fish length in the control and feed and Threadfin Shad pond types, and a negative relationship in the feed and Threadfin Shad pond type, suggesting that Threadfin Shad may alter the $\delta^{13}C$ values of larger sized Largemouth Bass. There was no relationship between $\delta^{15}N$ values and feed rate in the feed only established pond type; however there was a significant positive relationship between $\delta^{13}C$ values and feed rate. I expected there to be a negative relationship between $\delta^{13}C$ values and feed rate because pelleted feed has a lower $\delta^{13}C$ value than natural prey items for Bluegill. This relationship can be explained because 8 of the 10 feed ponds were fed rates below 2 kg – ha^{-1} d^{-1}, so the linear regression was heavily influenced by the two feed rates above 2 kg – ha^{-1} d^{-1}.

For the feed and threadfin shad established pond type there was a significant positive association between $\delta^{15}N$ values and feed rate, and no association between $\delta^{13}C$ values and feed rate. For the feed and Threadfin Shad pond types, all ponds were fed below 2 kg – ha^{-1} d^{-1}. There was probably not an adequate gradient of feed rates to conclude any meaningful effects of feed rate on Largemouth Bass $\delta^{15}N$ and $\delta^{13}C$ values.

I did not expect to find any significant results related to growth and feed rate for Largemouth Bass due to the small sample size and short duration of the controlled pond experiment. In a similar controlled pond experiment, there was no effect of feed rate on Largemouth Bass length, weight, or condition (Woodard et. al 2013). Similarly, I found no relationship between feed rate and Largemouth Bass total length at the midpoint, in July, and at the end of the controlled small pond experiment. There also was no effect of feed rate on Largemouth Bass $\delta^{15}N$ and $\delta^{13}C$ values at the midpoint and end of the small pond experiment. Though non-significant, the $\delta^{15}N$ values were lower for the highest feed rate than all others at the end of the controlled small pond experiment, similar to results for Bluegill at the higher feed rates, suggesting that additional replication or a longer study duration might enhance differences in $\delta^{15}N$ values for Largemouth Bass at the higher feed rates.

Threadfin Shad

Given documented preference of Largemouth Bass for Threadfin Shad when present (Davies et. al. 1979; Noble 1981; Wydoski and Bennett 1981), I expected Largemouth Bass $\delta^{13}C$ and $\delta^{15}N$ isotopic signatures to differ with presence/absence of Threadfin Shad in the established ponds that were fed pelleted feed. In contrast I found that $\delta^{13}C$ and $\delta^{15}N$ isotopic values of both Largemouth Bass and Bluegill did not differ in

the presence/absence of Threadfin Shad in the established ponds containing pelleted feed. Largemouth Bass $\delta^{13}C$ isotopic signatures were closely related to Bluegill $\delta^{13}C$ isotopic signatures in both the feed only and feed and Threadfin Shad pond types suggesting that Largemouth Bass were consuming primarily Bluegill in both pond types (Figure 43). However, I also found that Largemouth Bass length at age 2 was significantly greater when Threadfin Shad and pelleted feed were present versus absent. This suggests that Largemouth Bass may have been primarily consuming Bluegill (as indicated by stable isotope analysis), but supplementing their diets (and growth) with some Threadfin Shad.

There was a possible sampling bias in the methods used to collect the Largemouth Bass. I sampled Largemouth Bass and Bluegill using boat-mounted pulsed-DC electrofishing, in which I targeted littoral zones of the established ponds due to effectiveness. Research in two Central Florida lakes suggested a Largemouth Bass preference for littoral vegetated areas, whereas a small segment of the population remained offshore (Mesing and Wicker 1986). It is possible that the Largemouth Bass I collected were feeding primarily on Bluegill because I collected them in the littoral zone, and did not target Largemouth Bass in the open water limnetic zone. However, previous studies suggest that in some cases, even in the presence of *Dorsoma* spp., centrarchids remain the primary prey for Largemouth Bass (Timmons et al. 1980; Jackson et al. 1992; Bettoli et al. 1992; Irwin et al. 2003; Haley et al. 2012). This may be more realistic given the established ponds sampled were much smaller than the Florida lakes. Lastly, Largemouth Bass have showed yearly (Storck 1986) and seasonal variation (Storck 1986; Irwin et al. 2003) in diet items in the presence of Gizzard Shad. It is possible that there may be seasonal variation in prey selection for Largemouth Bass in the presence of

Threadfin Shad that I could not detect. I sampled in the established ponds during the summers of 2012 and 2013 because summer is when most pond owners are feeding with pelleted feed. A study conducted on the rates of carbon turnover in fish muscle reveals that adult Largemouth Bass show a $\delta^{13}C$ turnover half-life of 116-173 days (Weidel et al. 2011); therefore, the stable isotopic signatures of Largemouth Bass from the established ponds do not reflect sources of primary production in Largemouth Bass from the fall and early winter of those years.

Previous research suggests a potential for competition between Threadfin Shad and sunfishes (Davies et. al. 1979; Noble 1981); however, a literature review dealing with field manipulations of Gizzard and Threadfin Shad found mixed results for effects of Threadfin Shad on Bluegill (DeVries and Stein 1990). I expected Bluegill $\delta^{13}C$ and $\delta^{15}N$ isotopic signatures to differ in the feed only and feed and Threadfin Shad pond types because of a heavy reliance upon pelleted feed when Threadfin Shad were present due to presumed competition (Davies et. al. 1979) and a possible reduction in zooplankton densities (Ziebell et. al. 1986; Prophet 1988; DeVries et. al. 1991; Garvey and Stein 1998; Haley et. al. 2012); however, there were no differences in zooplankton densities nor Bluegill isotopic signatures between pond types. This suggests that either Bluegill relied on pelleted feed in the feed only pond type the same as in the feed and Threadfin Shad pond type, or food was not limited for Bluegill in the presence of Threadfin Shad in the established ponds. Further research needs to be conducted on the impact of Threadfin Shad introductions to both Largemouth Bass and Bluegill in small impoundments.

Management Implications

Using pelleted feed as a management strategy in small impoundments can be an effective strategy for increasing Bluegill growth (Schmittou 1969; Berger 1982; Murnyak et al. 1984; Porath and Hurley 2005; Woodard et al. 2013), condition (Schmittou 1969; Burger 1982; Murnyak et al. 1984; Woodard et al. 2013), and reproduction (Woodard 2011). Less known are the trophic effects of pelleted feed through the food web, ultimately to Largemouth Bass. Similar to previous research, I found that increasing feed rate led to increased Bluegill growth and energy allocated to reproduction. I was also able to detect differences in Bluegill $\delta^{15}N$ and $\delta^{13}C$ values in the presence versus absence of pelleted feed in established ponds, as well as with increasing feed rate in a controlled experiment. A pelleted feed signature was detectable through multiple trophic levels in the $\delta^{15}N$ and $\delta^{13}C$ values of Largemouth Bass (Figure 42). Pelleted feed also affected the $\delta^{15}N$ and $\delta^{13}C$ values of Bluegill and Largemouth Bass across whole established ponds, suggesting that pelleted feed provides more than just localized effects around a feeder. These differences were detected in the established ponds, and most of those ponds were fed at a lower rate than what is recommended, suggesting that even at low feeding rates pelleted feed can be detected through trophic levels and ultimately in the isotopic signature of Largemouth Bass. More research needs to be completed to see if that result ultimately translates into increased and sustainable growth and condition for Largemouth Bass before recommending pelleted feed as a management tool for increasing growth and condition of Largemouth Bass. My research suggests the potential is there.

Previous work indicates mixed results on the effects of Threadfin Shad for both Largemouth Bass and Bluegill (DeVries and Stein 1990). Threadfin Shad did not affect

$\delta^{15}N$ and $\delta^{13}C$ values of either Largemouth Bass or Bluegill, suggesting that pelleted feed was detectable in the isotopic signatures of fish through multiple trophic levels even in the presence of another prey item. Although Threadfin Shad did not affect the isotopic signatures of Largemouth Bass, I did detect a difference in growth for age 2 Largemouth Bass in the presence of Threadfin Shad and pelleted feed. Threadfin Shad did not have an effect on the detected isotope effects resulting from pelleted feed; however, additional research is required to determine these complex effects of Threadfin Shad in small impoundments.

According to Haley et al. (2012), pond enhancement techniques of pelleted feed and Threadfin Shad cannot by themselves consistently produce ideal fish growth and condition. The use of supplemental pelleted feed and stocking Threadfin Shad may only further enhance fish growth and condition when used in combination with other management techniques like fish harvest, water quality control, and aquatic macrophyte control. My results suggest pelleted feed as a potential strategy to enhance fish growth and condition through multiple trophic levels even in the presence of alternative forage species (i.e. Threadfin Shad) (Figure 42), and Threadfin Shad may be used as a tool in combination with pelleted feed to increase Largemouth Bass growth in small impoundments. Though, these strategies must be used in combination with other management techniques to be effective at achieving and maintaining desired fish growth and condition. My research also suggest the importance for a re-evaluation of recommended feeding rates based upon pond owner expectations given we were able to detect the isotopic signature of pelleted feed through the food web, in most cases, well below the current recommended feeding rates.

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Table 1. Summary of measures for the small pond experiment.

Small Pond Experiment 2012		
Measure	**Frequency**	**Timeline**
Temperature ℃	Every 2 hours	May-August
Secchi Depth (cm)	Once per month	April-July
Dissolved Oxygen (mg/L)	Once per month	April-July
Larval Fish (No./M^3)	Once per month	April-July
Zooplankton (No./L)	Once per month	April-July
Chlorophyll *a* (μg/L)	Once per month	April-July
Turbidity (NTU)	Once per month	April-July
Electrofishing	Once per Month	June-July

Table 2 Established pond surface hactares and feed rate.

Pond	Treatment	Surface Hectares	Feed Rate (kg – ha^{-1} d^{-1})
1	Control	3.62	0
2	Control	1.83	0
3	Control	1.63	0
4	Control	2.04	0
5	Control	1.63	0
6	Control	2.14	0
7	Control	4.21	0
8	Control	2.78	0
9	Control	0.98	0
10	Control	0.8	0
11	Feed Only	0.81	0.47
12	Feed Only	10.46	0.93
13	Feed Only	3.84	1.69
14	Feed Only	3.72	0.30
15	Feed Only	4.03	0.56
16	Feed Only	2.27	4.28
17	Feed Only	1.41	0.40
18	Feed Only	0.87	2.61
19	Feed Only	2.51	0.65
20	Feed Only	6.7	0.48
21	Feed and THSH	8.51	1.00
22	Feed and THSH	15.73	0.10
23	Feed and THSH	2.46	0.46
24	Feed and THSH	2.23	0.73
25	Feed and THSH	2.84	1.60
26	Feed and THSH	1.78	1.59
27	Feed and THSH	5.31	0.43
28	Feed and THSH	23.29	0.24
29	Feed and THSH	7.39	0.22
30	Feed and THSH	6.93	1.87

Table 3 Abiotic, plankton, and larval Bluegill model results for separate linear mixed effect models (LME) from the small pond experiment.

Treatment Effects		
Measure	**F- statistic**	**P-value**
Chlorophyll *a* (µg/L)	$F_{(1,8)}$=0.11	p=0.75
Turbidity (NTU)	$F_{(1,8)}$=0.30	p=0.60
Secchi Depth (cm)	$F_{(1,8)}$<0.01	p=0.96
Dissolved Oxygen (mg/L)	$F_{(1,8)}$=0.07	p=0.80
Larval Fish (No./M^3)	$F_{(1,8)}$=0.44	p=0.52
Zooplankton (No./L)	$F_{(1,8)}$=0.39	p=0.55
Time Effects		
Chlorophyll *a* (µg/L)	$F_{(4,32)}$=6.82	p<0.01
Turbidity (NTU)	$F_{(4,32)}$=1.34	p=0.28
Secchi Depth (cm)	$F_{(4,31)}$=0.76	p=0.56
Dissolved Oxygen (mg/L)	$F_{(4,32)}$=0.99	p=0.43
Larval Fish (No./M^3)	$F_{(4,32)}$=5.19	p<0.01
Zooplankton (No./L)	$F_{(4,31)}$=8.58	p<0.01
Interaction Effects (Time*Treatment)		
Chlorophyll *a* (µg/L)	$F_{(4,32)}$=0.31	p=0.87
Turbidity (NTU)	$F_{(4,32)}$=0.33	p=0.85
Secchi Depth (cm)	$F_{(4,31)}$=0.80	p=0.53
Dissolved Oxygen (mg/L)	$F_{(4,32)}$=1.10	p=0.37
Larval Fish (No./M^3)	$F_{(4,32)}$=0.11	p=0.98
Zooplankton (No./L)	$F_{(4,31)}$=0.13	P=0.97

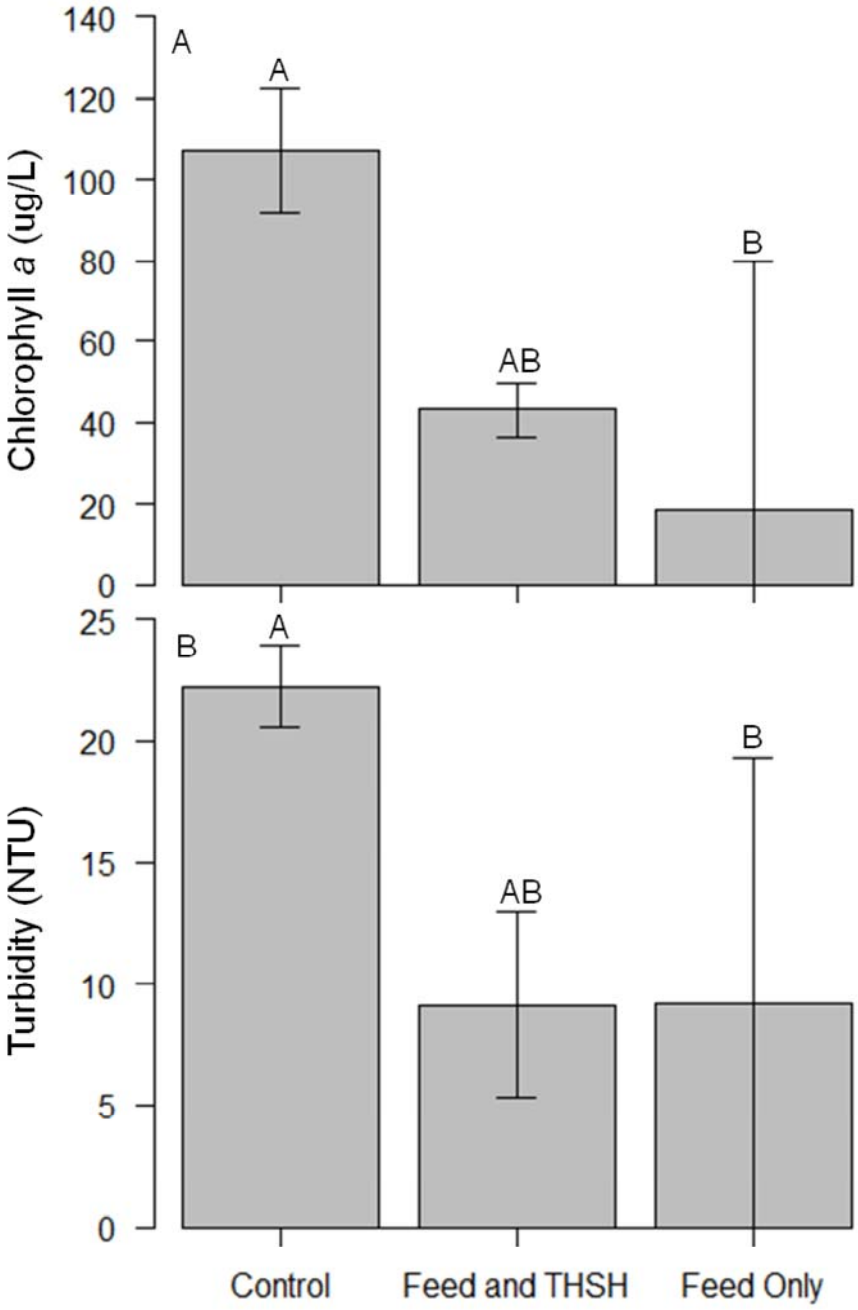
Chlorophyll a (ug/L)
140
120
100
80
60
40
20
0
A
A
AB
B
Turbidity (NTU)
25
20
15
10
5
0
B
A
AB
B
Control
Feed and THSH
Feed Only

Figure 1.

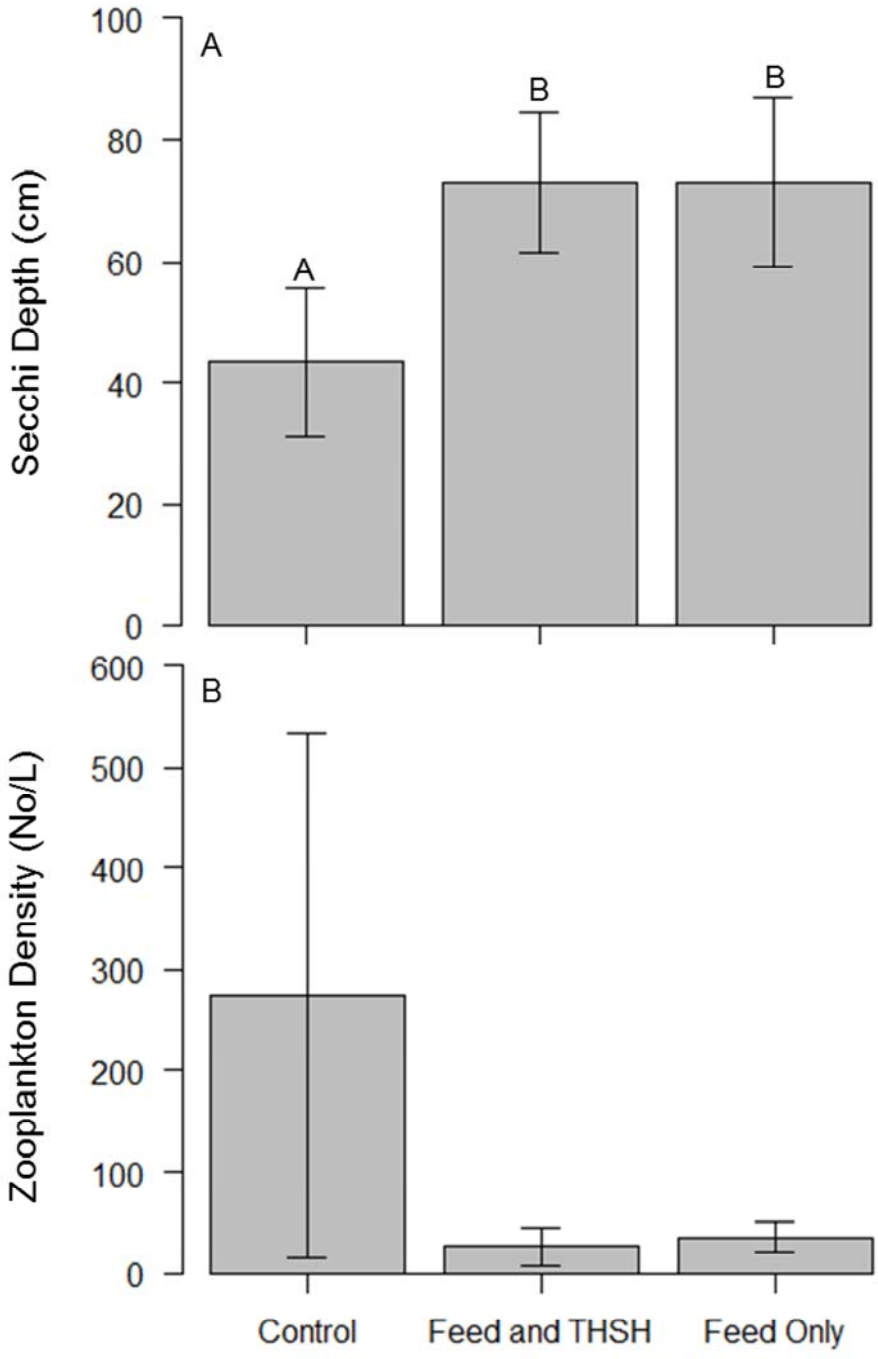
Secchi Depth (cm)
100
80
60
40
20
0
A
B
B
A
Zooplankton Density (No/L)
600
500
400
300
200
100
0
B
Control
Feed and THSH
Feed Only

Figure 2.

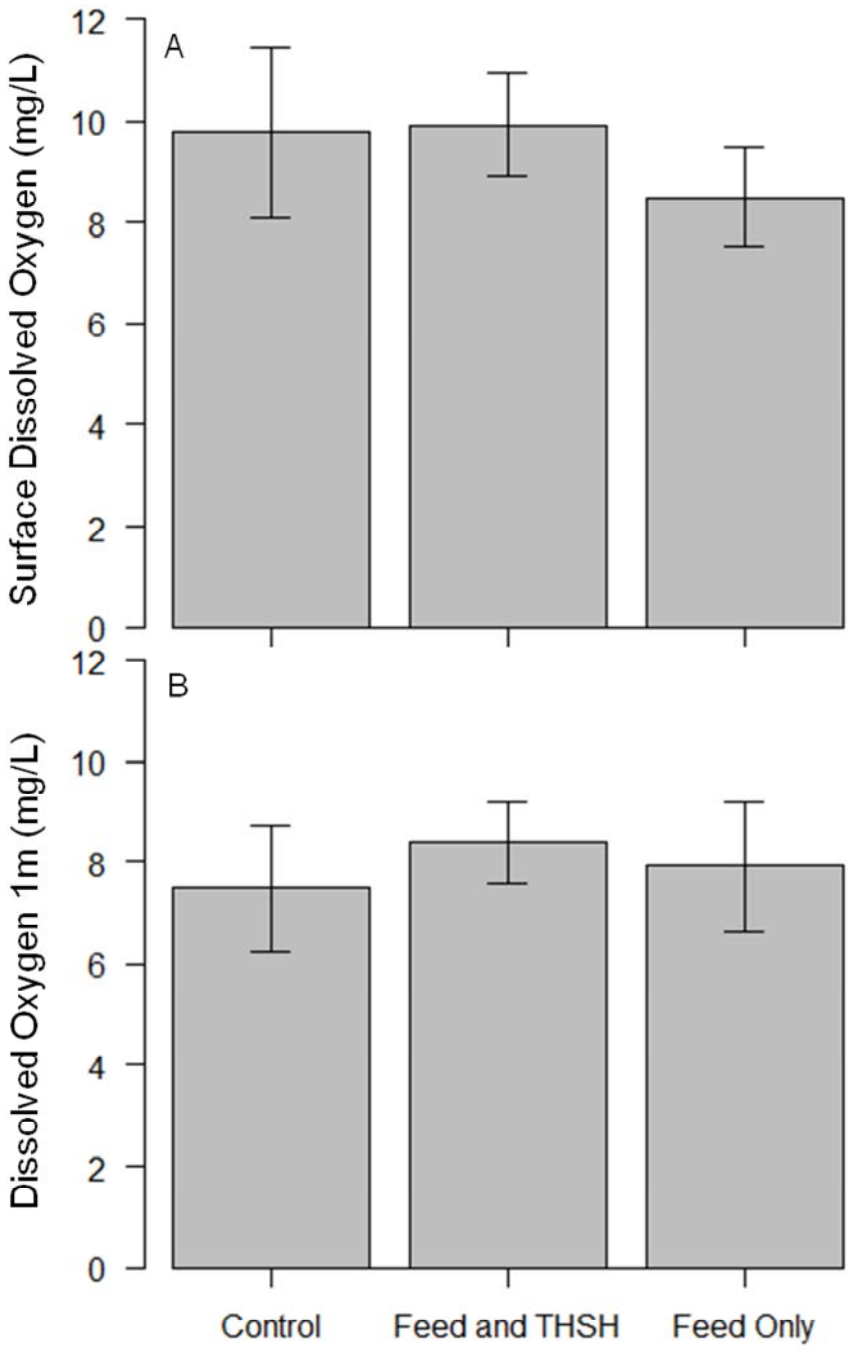
A
B
Surface Dissolved Oxygen (mg/L)
Dissolved Oxygen 1m (mg/L)
0
2
4
6
8
10
12
Control
Feed and THSH
Feed Only

Figure 3.

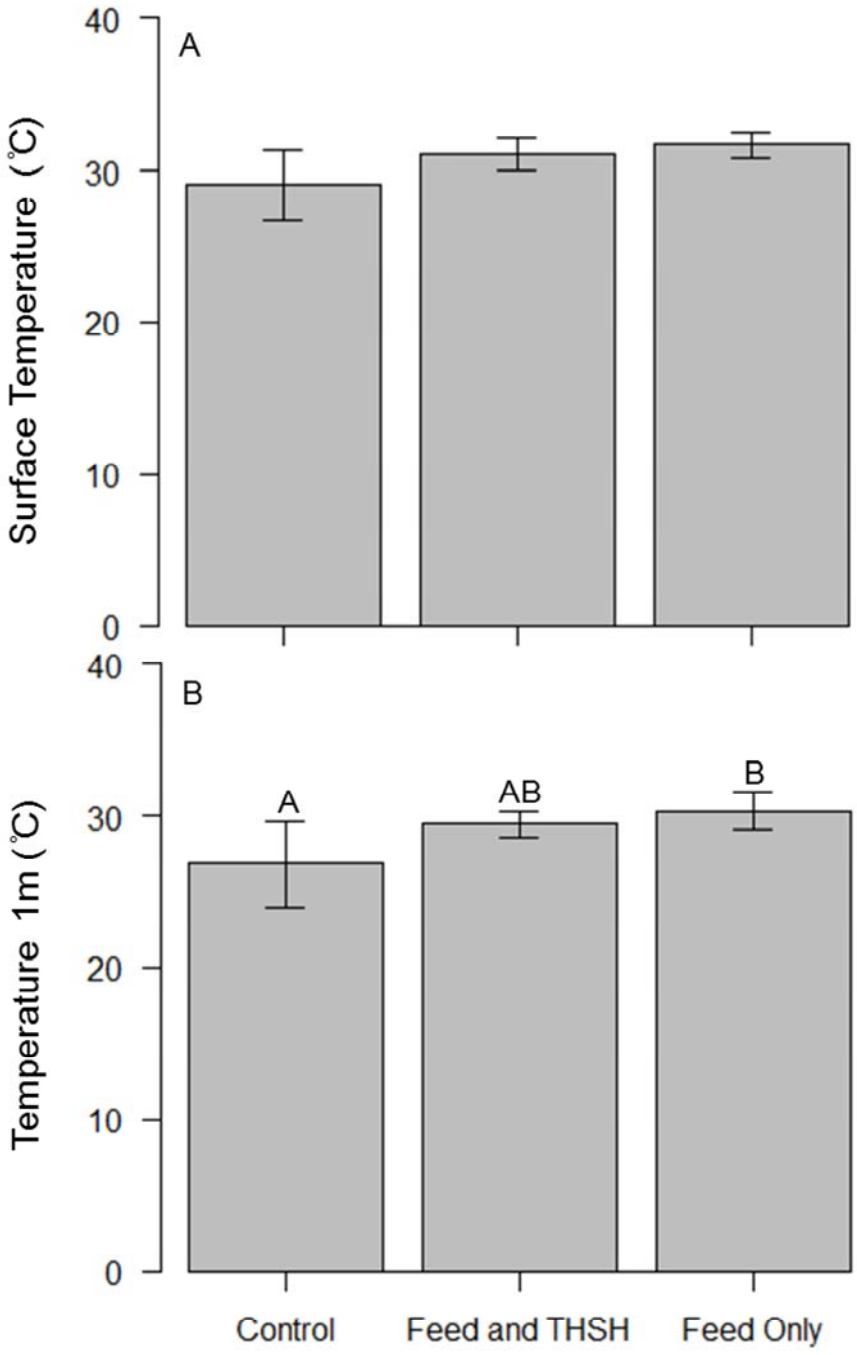
Surface Temperature (°C)
40
30
20
10
0
A
Temperature 1m (°C)
40
30
20
10
0
B
A
AB
B
Control
Feed and THSH
Feed Only

Figure 4.

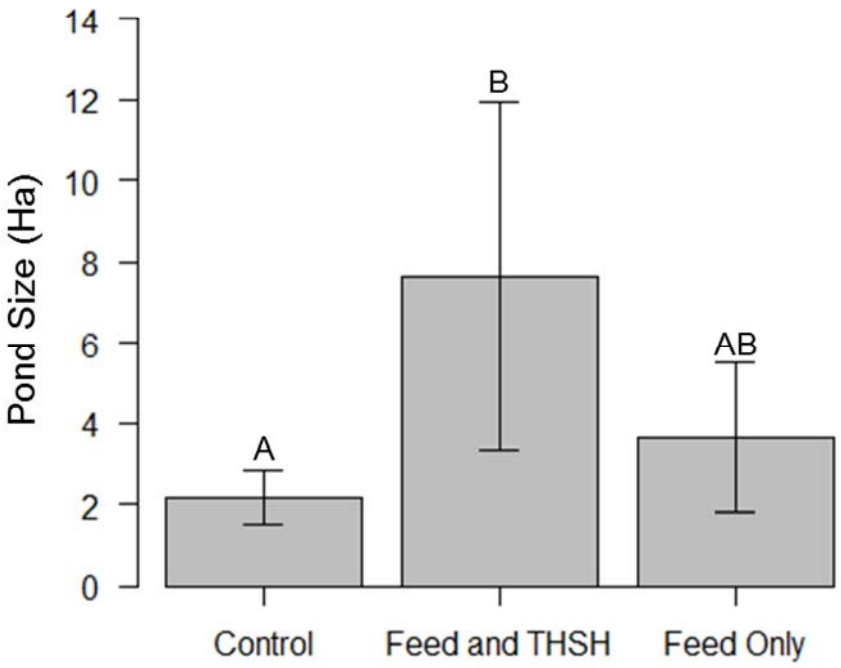
Pond Size (Ha)
14
12
10
8
6
4
2
0
A
B
AB
Control
Feed and THSH
Feed Only

Figure 5.

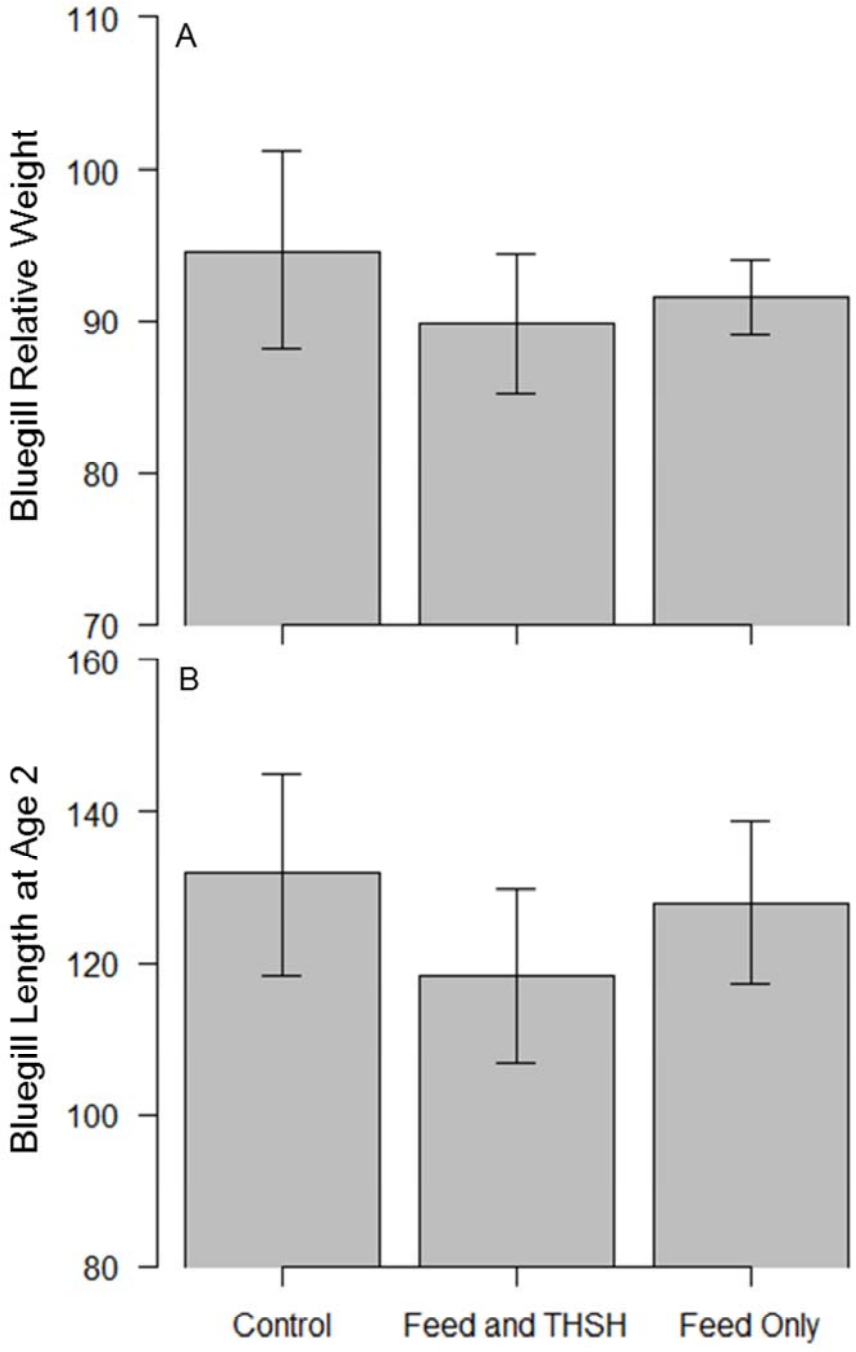
A
B
Bluegill Relative Weight
Bluegill Length at Age 2
110
100
90
80
70
160
140
120
100
80
Control
Feed and THSH
Feed Only

Figure 6.

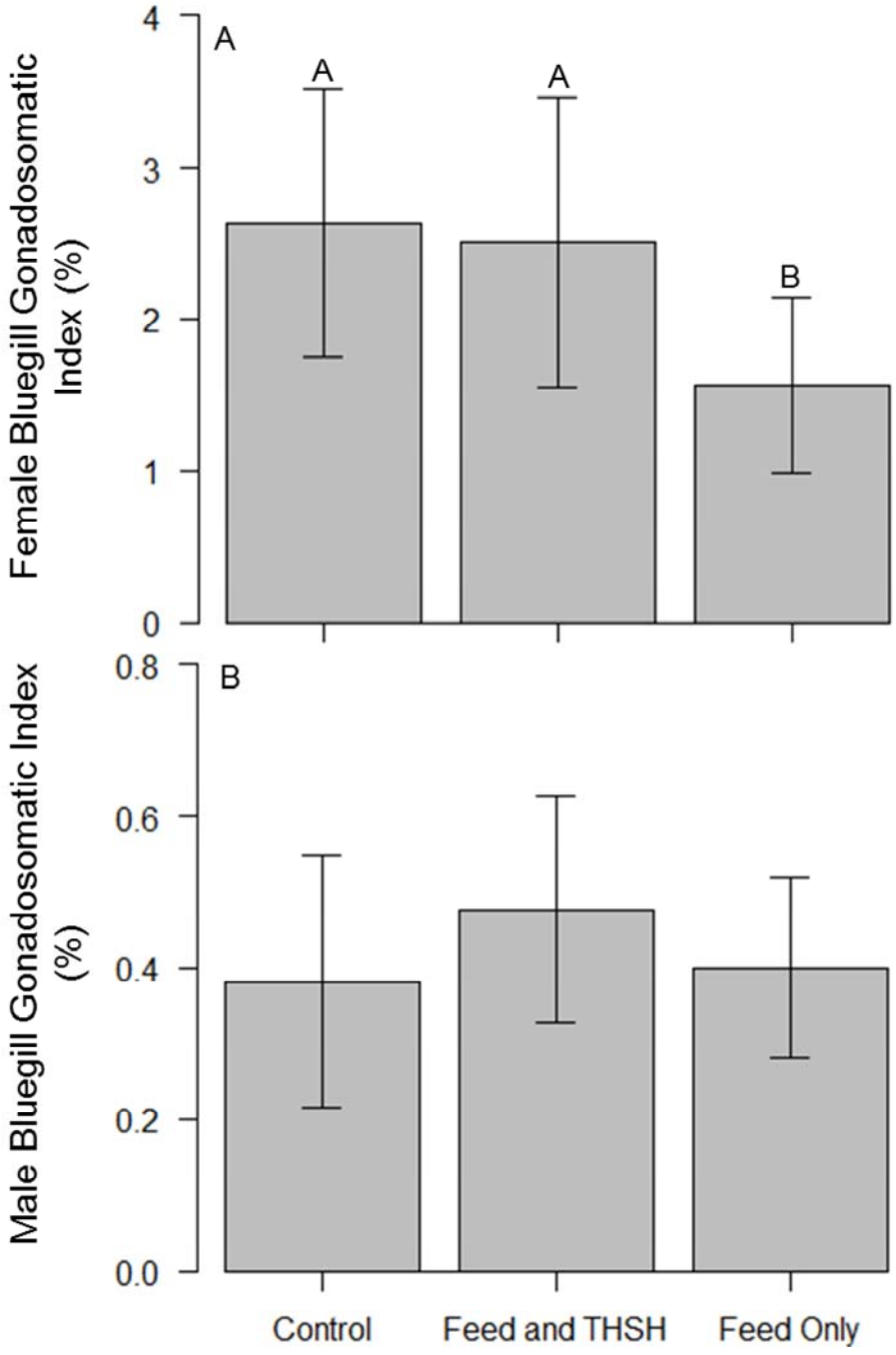
Female Bluegill Gonadosomatic Index (%)
4
3
2
1
0
A
A
A
B
Male Bluegill Gonadosomatic Index (%)
0.8
0.6
0.4
0.2
0.0
B
Control
Feed and THSH
Feed Only

Figure 7.

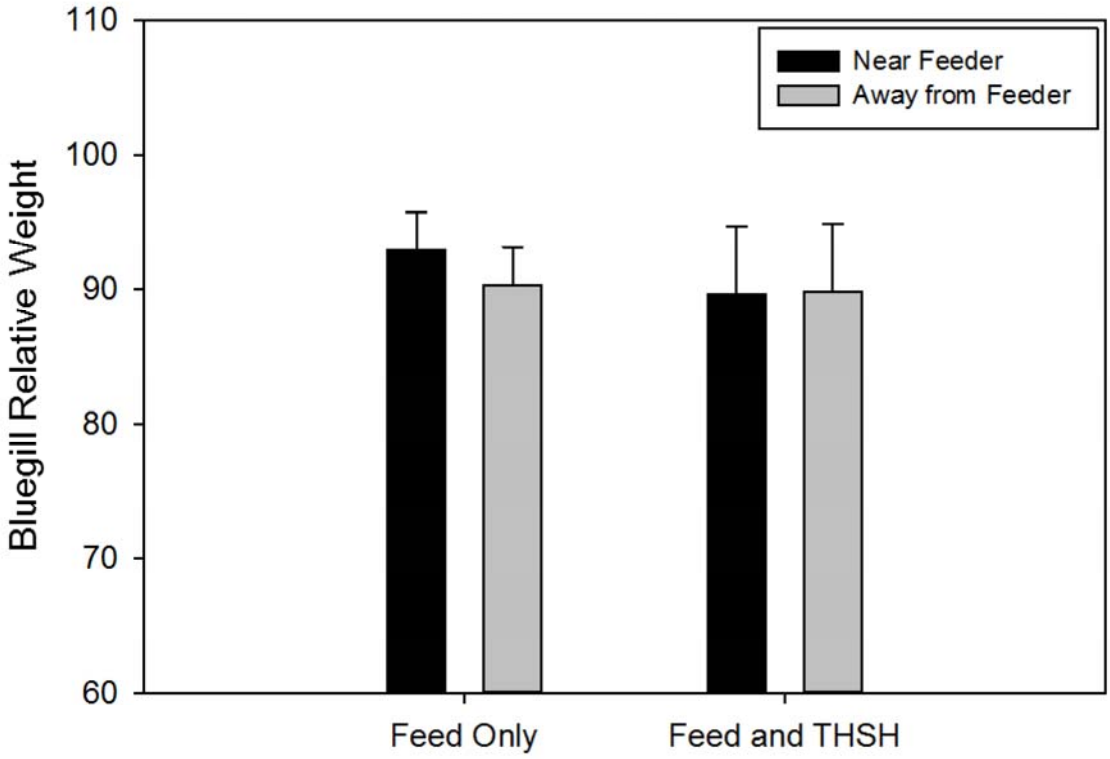
110
100
90
80
70
60
Bluegill Relative Weight
Near Feeder
Away from Feeder
Feed Only
Feed and THSH

Figure 8.

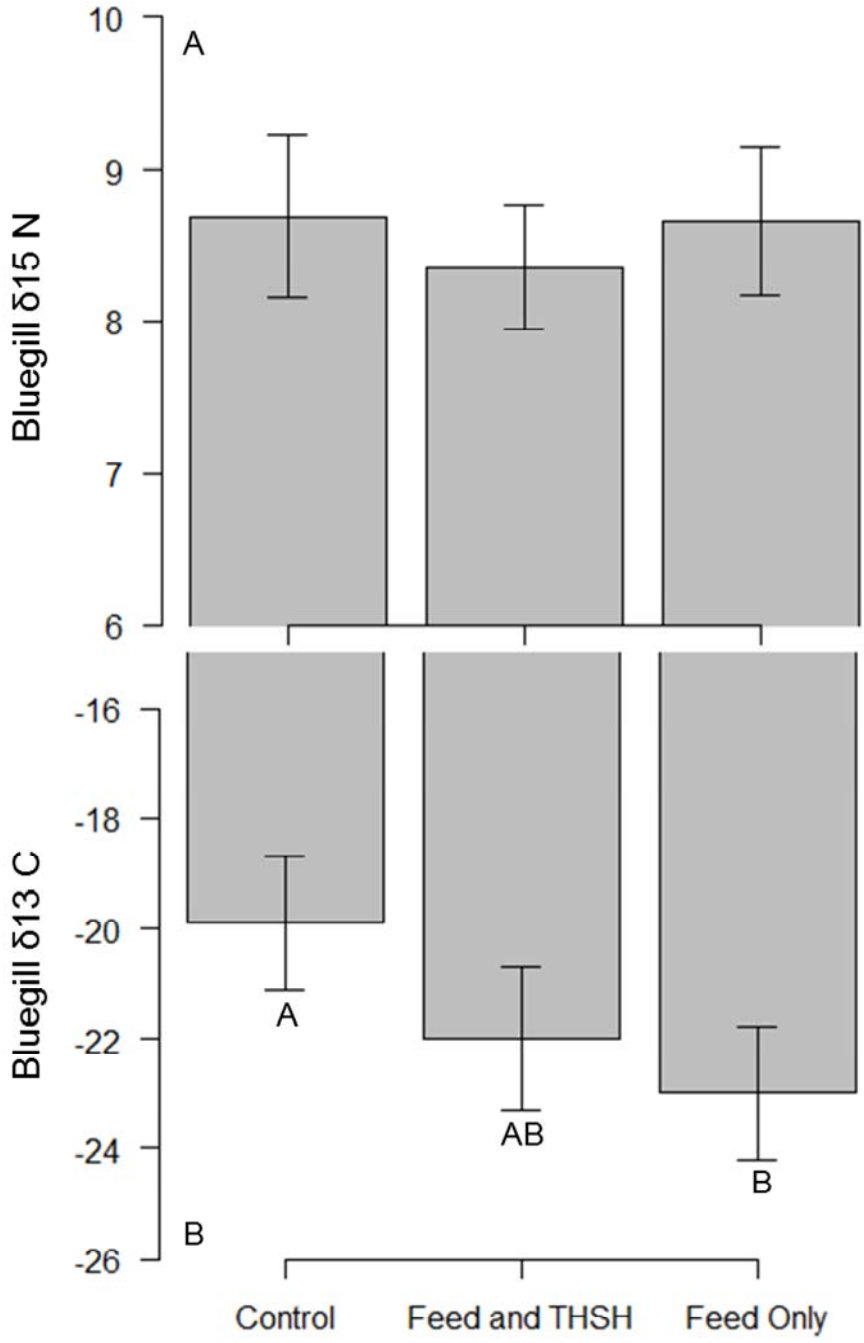
10
9
8
7
6
A
Bluegill δ15 N
-16
-18
-20
-22
-24
-26
B
Bluegill δ13 C
A
AB
B
Control
Feed and THSH
Feed Only

Figure 9.

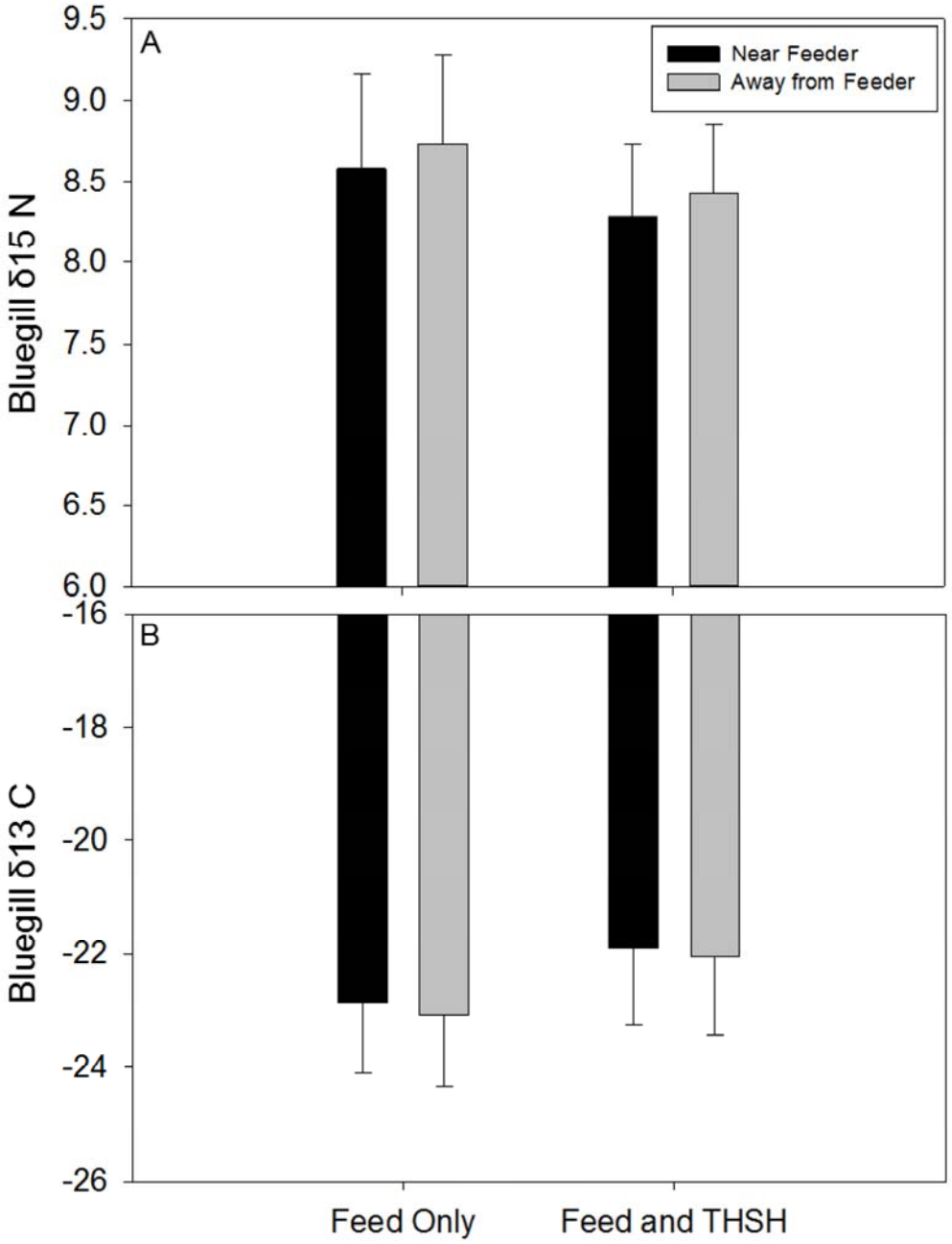
A
B
Near Feeder
Away from Feeder
Bluegill δ15 N
Bluegill δ13 C
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
-16
-18
-20
-22
-24
-26
Feed Only
Feed and THSH

Figure 10.

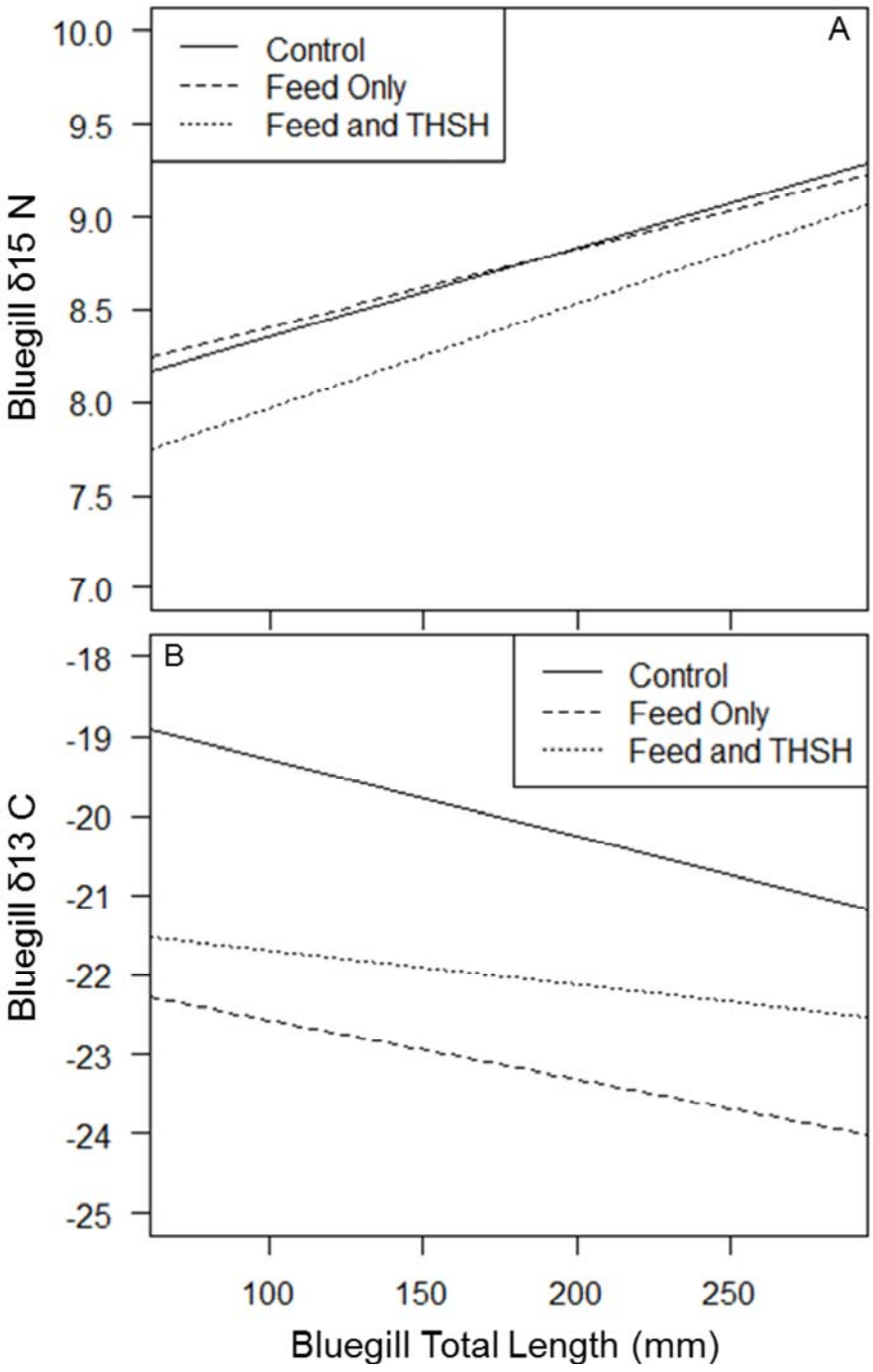
A
Control
Feed Only
Feed and THSH
Bluegill δ15 N
10.0
9.5
9.0
8.5
8.0
7.5
7.0
B
Control
Feed Only
Feed and THSH
Bluegill δ13 C
-18
-19
-20
-21
-22
-23
-24
-25
100
150
200
250
Bluegill Total Length (mm)

Figure 11.

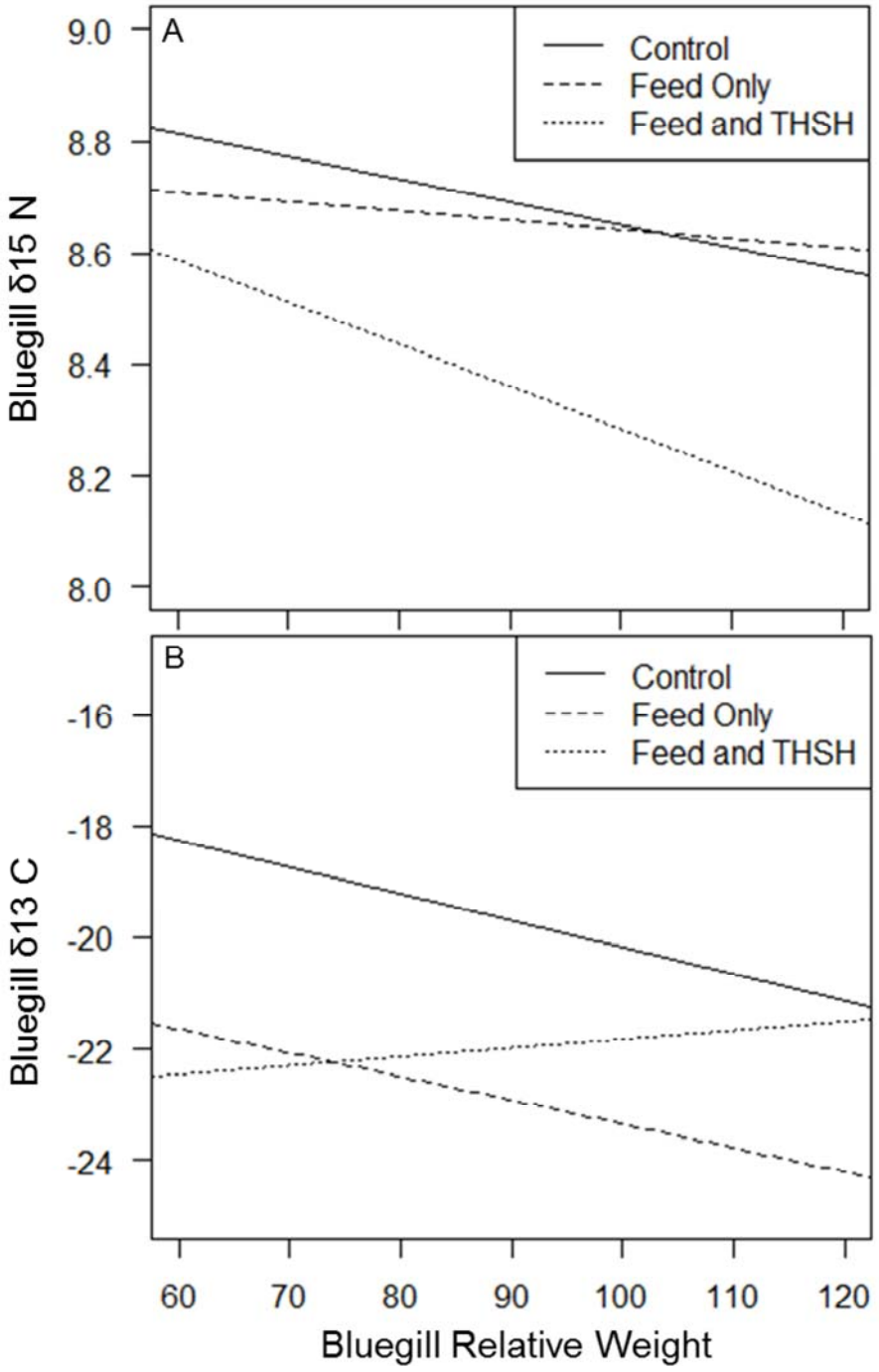
A
B
Control
Feed Only
Feed and THSH
Bluegill δ15 N
Bluegill δ13 C
9.0
8.8
8.6
8.4
8.2
8.0
-16
-18
-20
-22
-24
60
70
80
90
100
110
120
Bluegill Relative Weight

Figure 12.

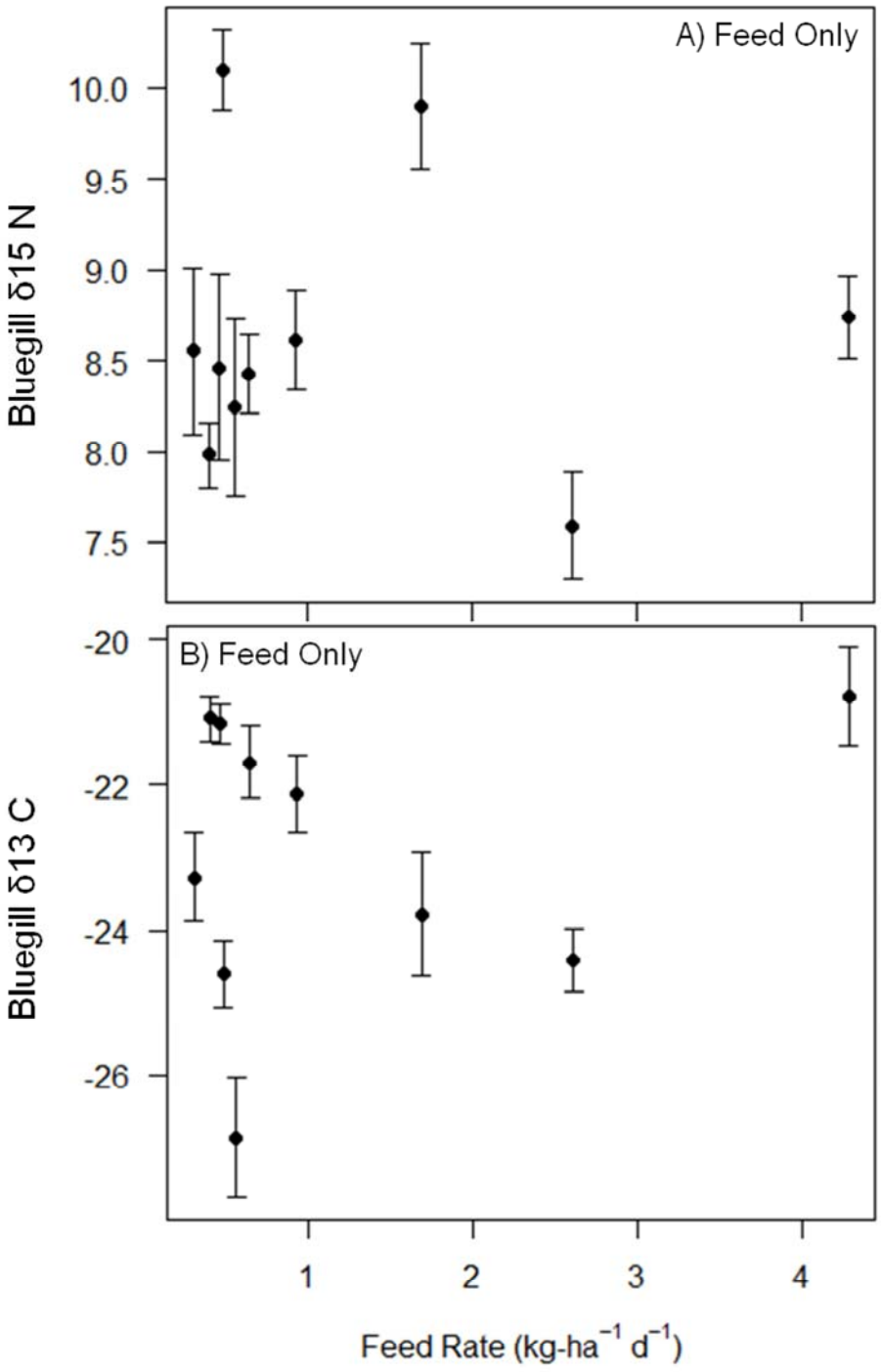
A) Feed Only
Bluegill δ15 N
10.0
9.5
9.0
8.5
8.0
7.5
B) Feed Only
Bluegill δ13 C
-20
-22
-24
-26
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 13.

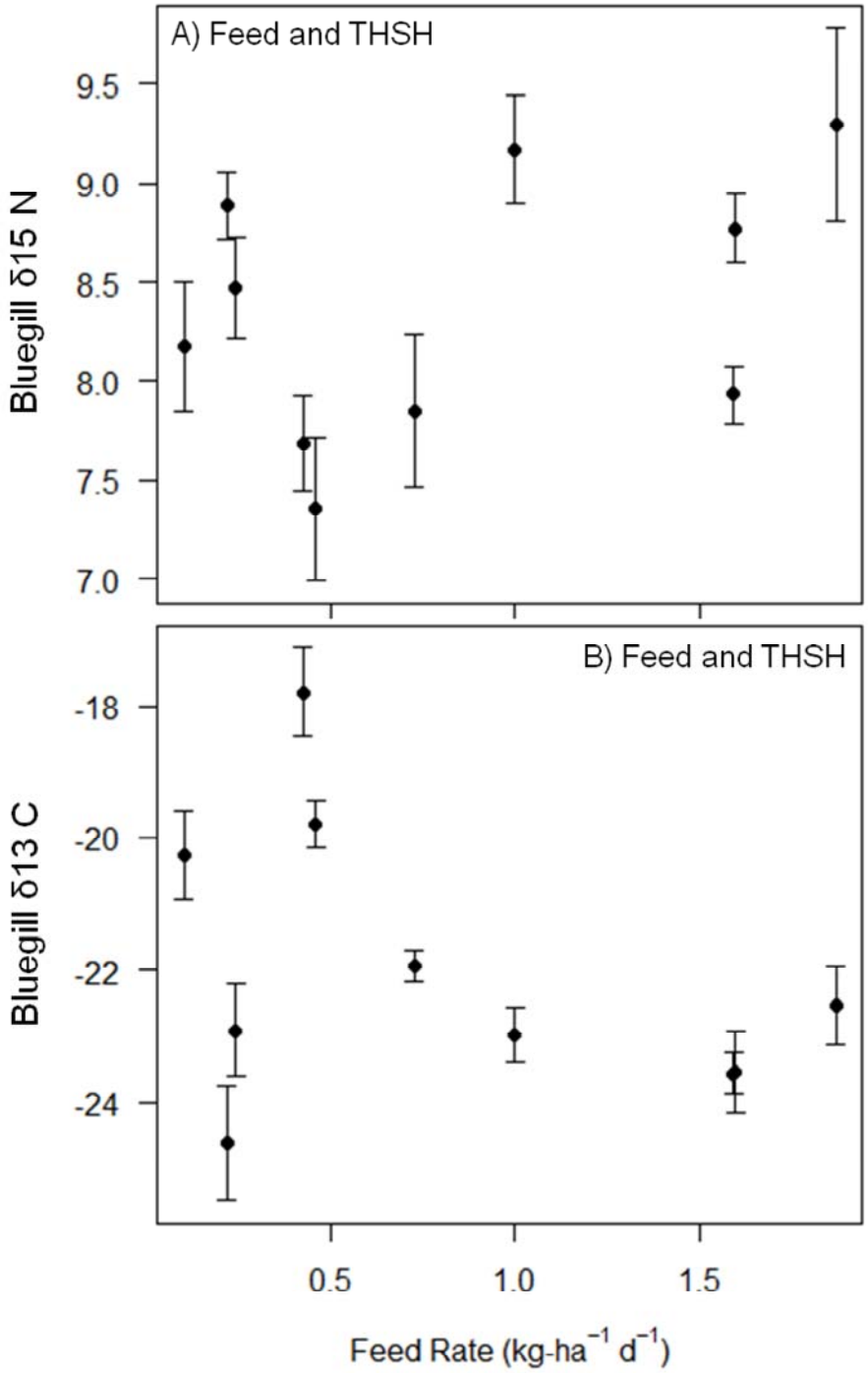
A) Feed and THSH
Bluegill δ15 N
9.5
9.0
8.5
8.0
7.5
7.0
B) Feed and THSH
Bluegill δ13 C
-18
-20
-22
-24
0.5
1.0
1.5
Feed Rate (kg-ha^{-1} d^{-1})

Figure 14.

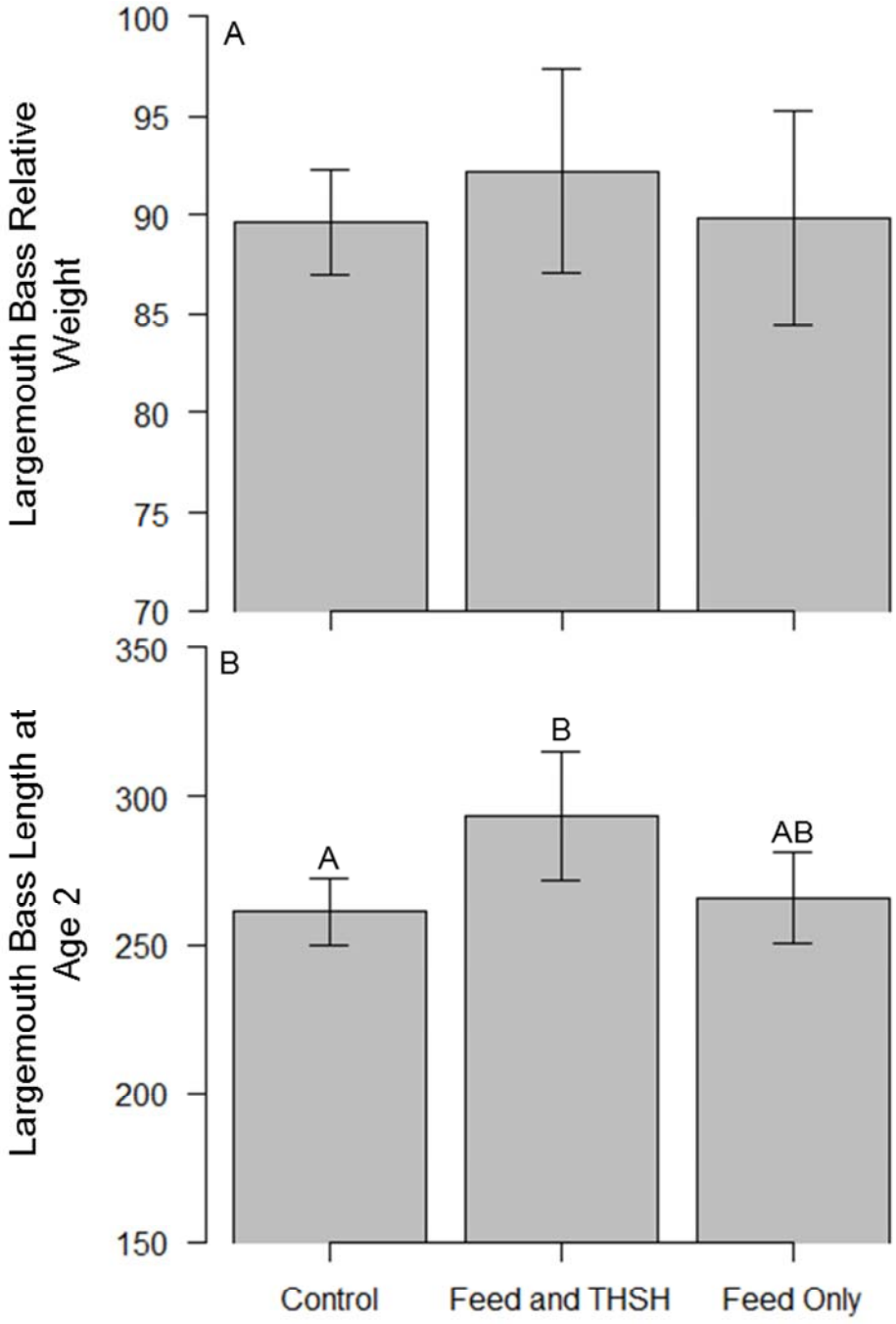
Largemouth Bass Relative Weight
100
95
90
85
80
75
70
A
Largemouth Bass Length at Age 2
350
300
250
200
150
B
A
B
AB
Control
Feed and THSH
Feed Only

Figure 15.

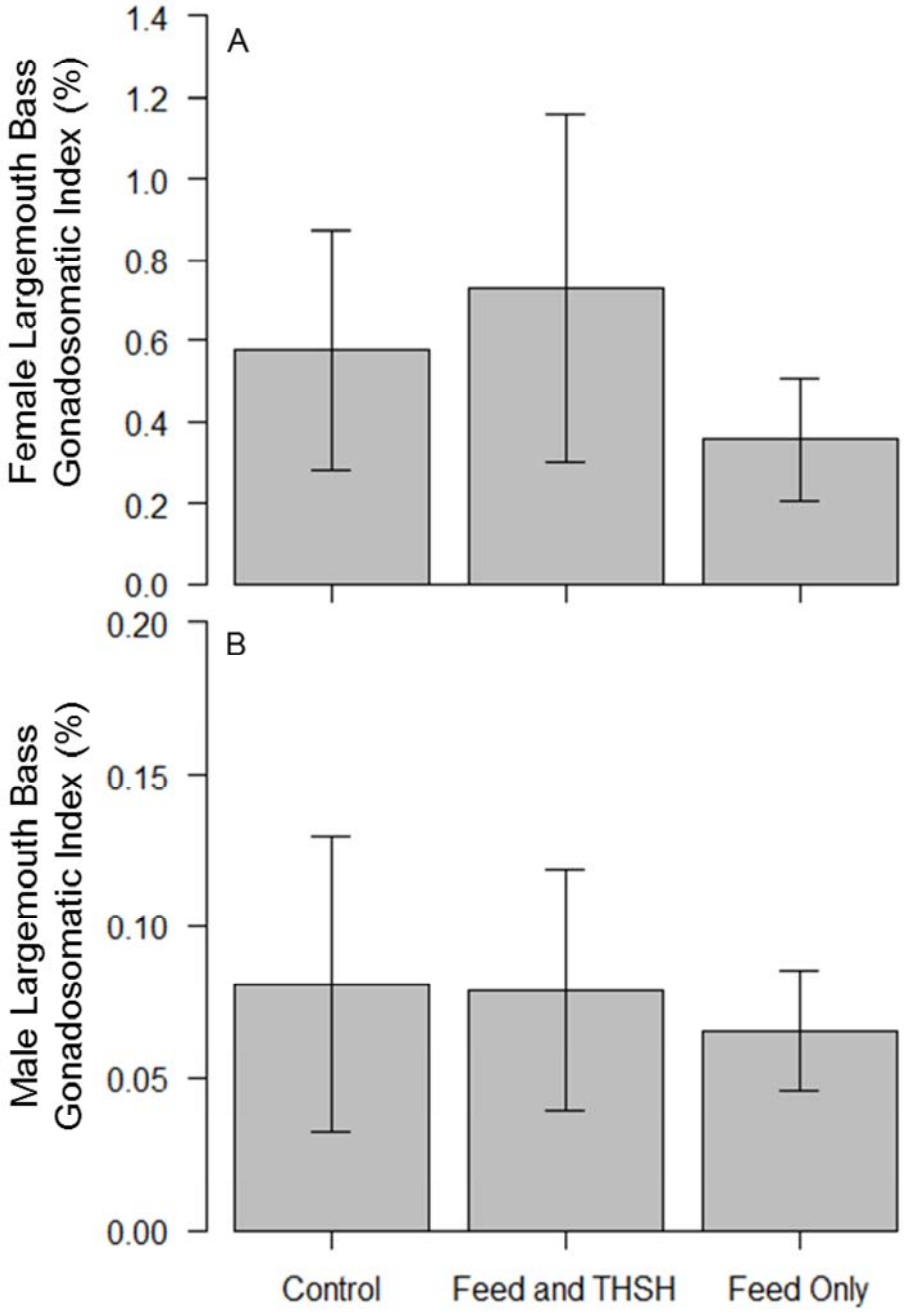
A
Female Largemouth Bass
Gonadosomatic Index (%)
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
B
Male Largemouth Bass
Gonadosomatic Index (%)
0.20
0.15
0.10
0.05
0.00
Control
Feed and THSH
Feed Only

Figure 16.

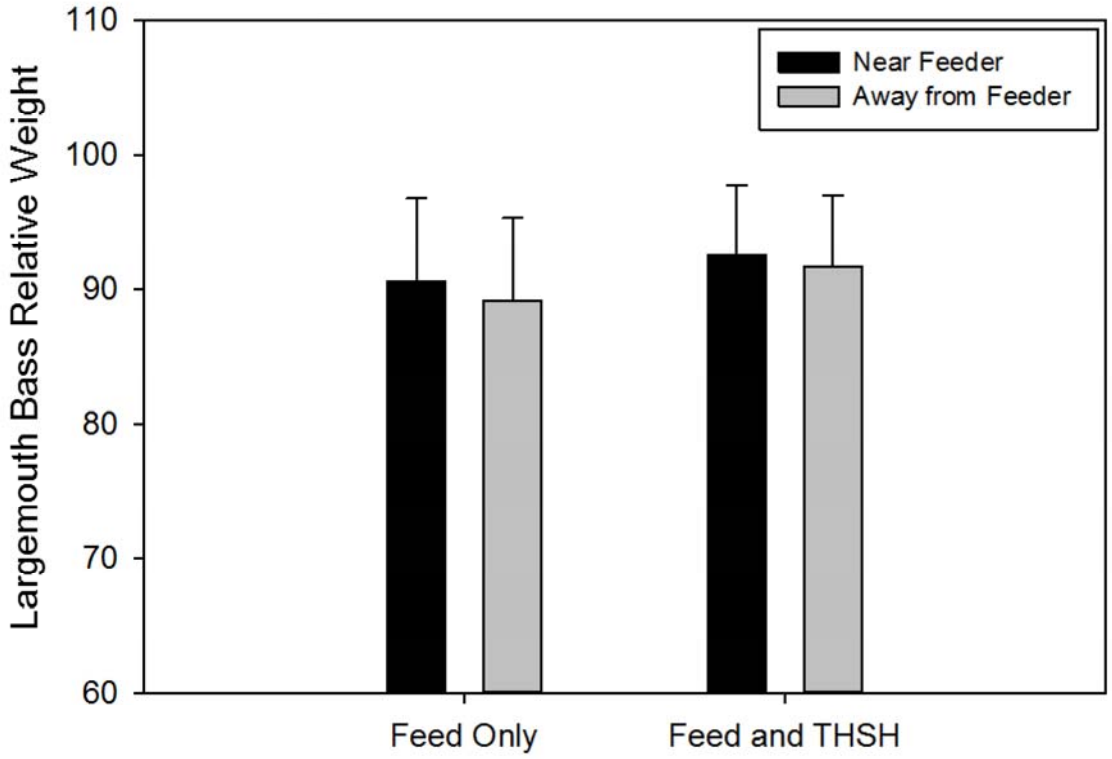
Largemouth Bass Relative Weight
110
100
90
80
70
60
Near Feeder
Away from Feeder
Feed Only
Feed and THSH

Figure 17.

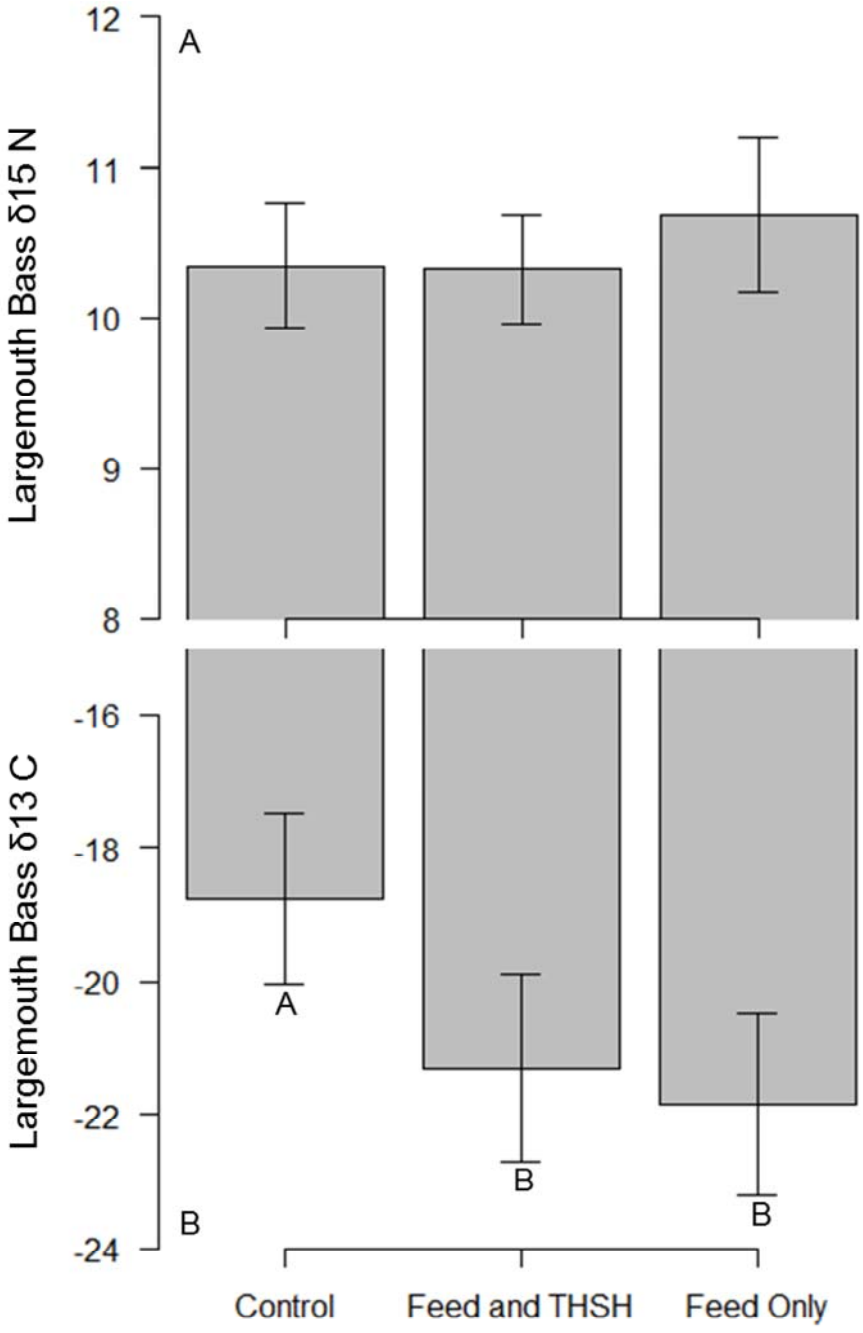
Largemouth Bass δ15 N
12
11
10
9
8
A
Largemouth Bass δ13 C
-16
-18
-20
-22
-24
B
A
B
B
Control
Feed and THSH
Feed Only

Figure 18.

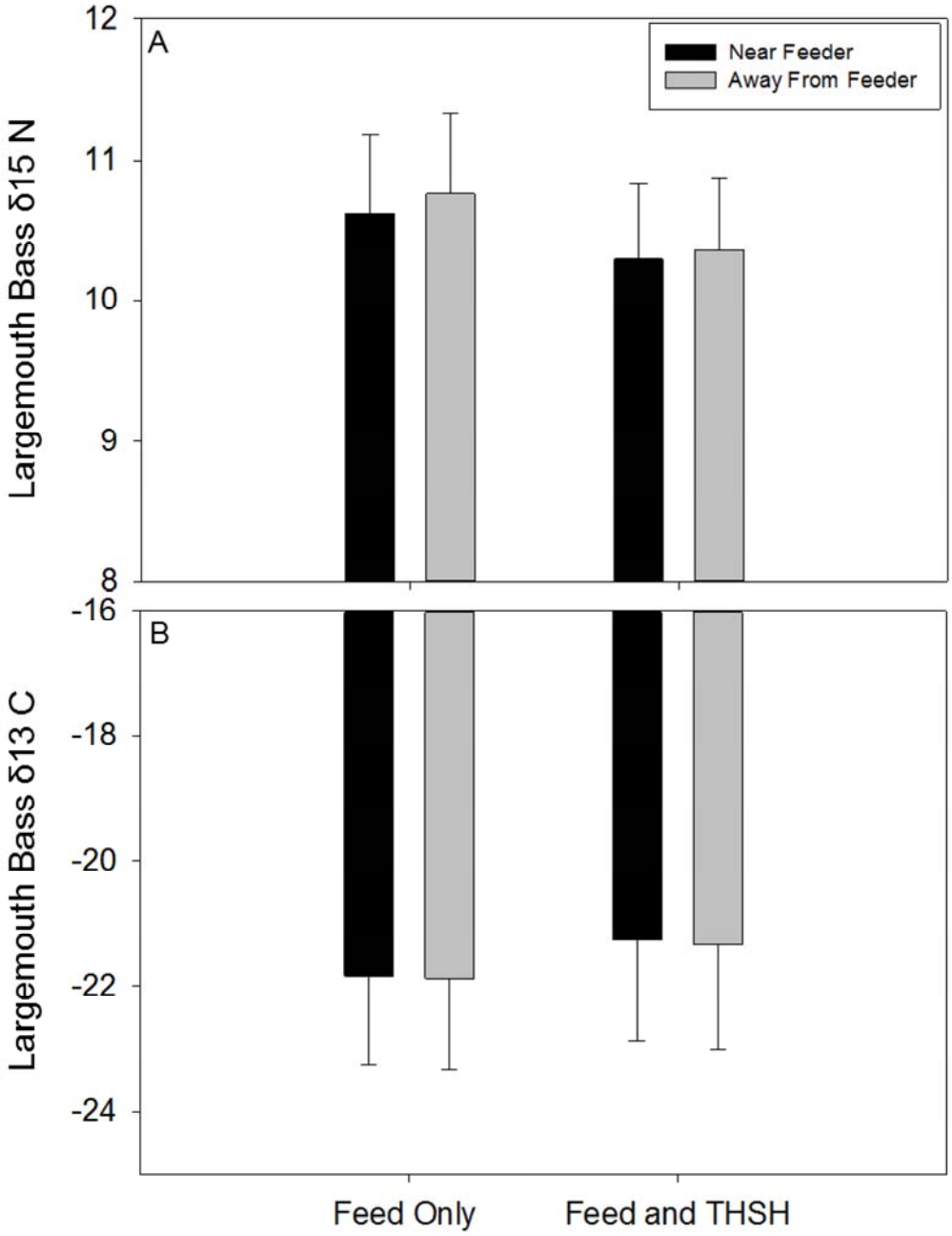
A
B
Near Feeder
Away From Feeder
Largemouth Bass δ15 N
Largemouth Bass δ13 C
12
11
10
9
8
-16
-18
-20
-22
-24
Feed Only
Feed and THSH

Figure 19.

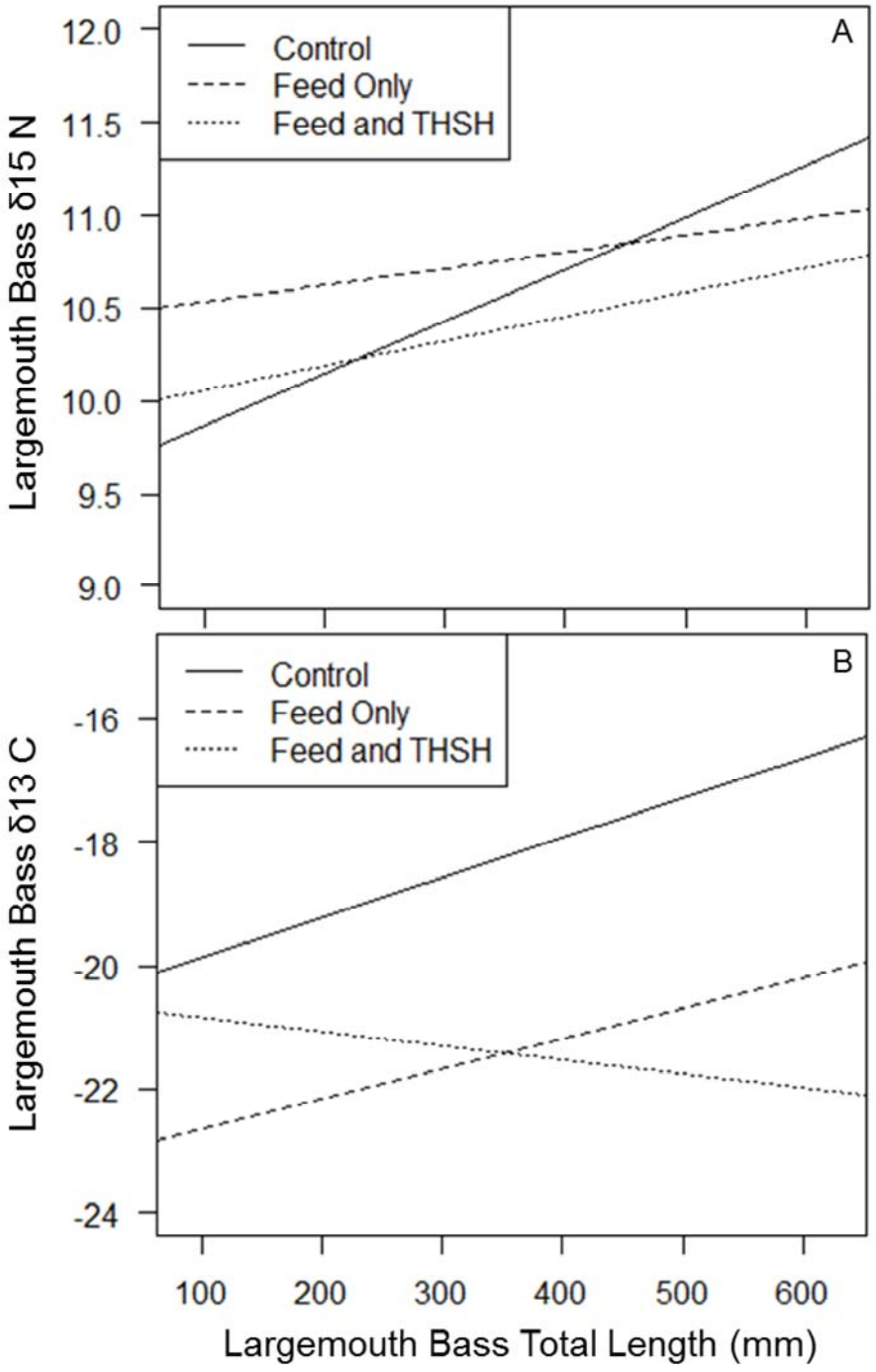
A
Control
Feed Only
Feed and THSH
Largemouth Bass δ15 N
12.0
11.5
11.0
10.5
10.0
9.5
9.0
B
Control
Feed Only
Feed and THSH
Largemouth Bass δ13 C
-16
-18
-20
-22
-24
100
200
300
400
500
600
Largemouth Bass Total Length (mm)

Figure 20.

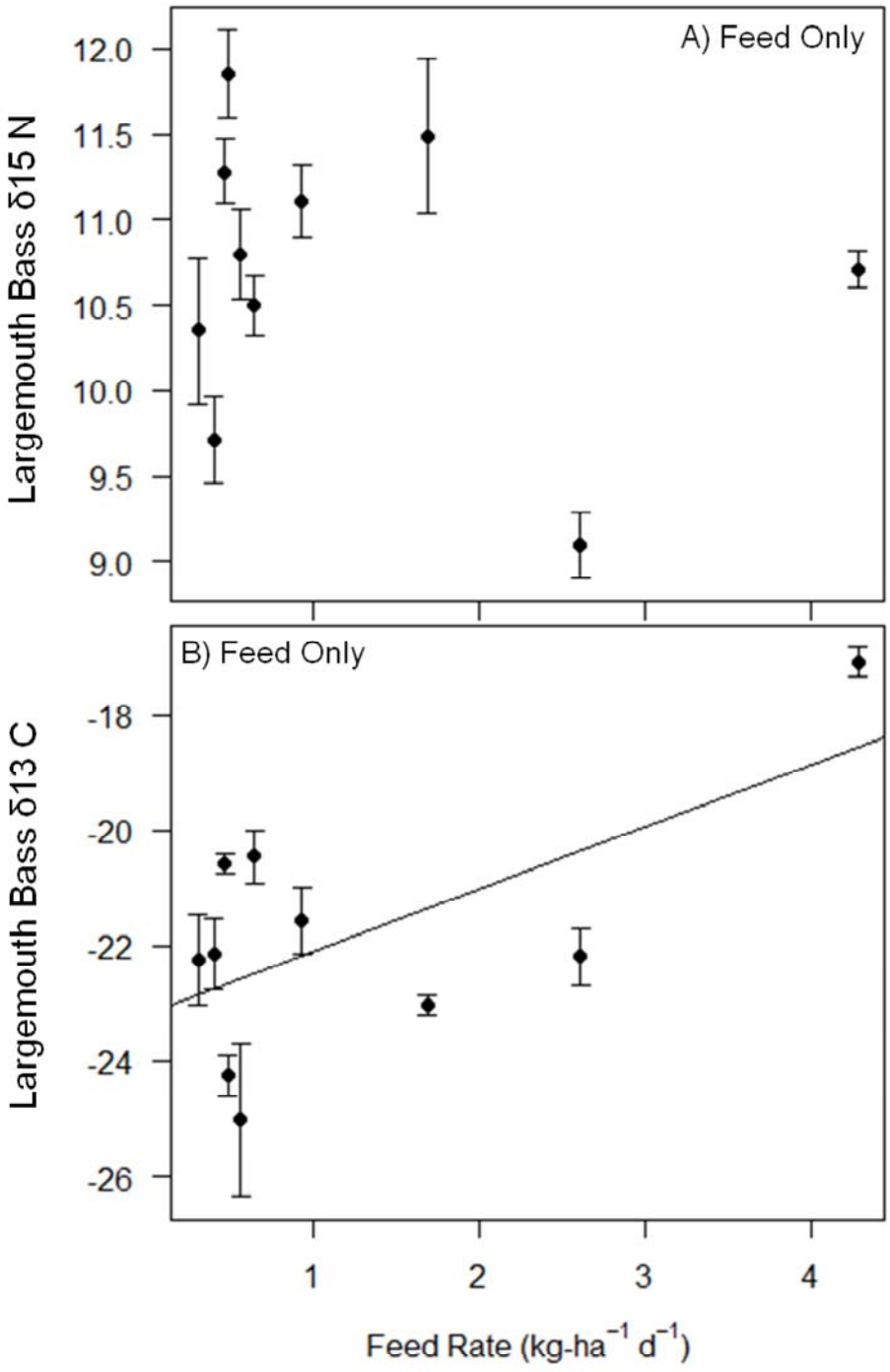
A) Feed Only
Largemouth Bass δ15 N
12.0
11.5
11.0
10.5
10.0
9.5
9.0
B) Feed Only
Largemouth Bass δ13 C
-18
-20
-22
-24
-26
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 21.

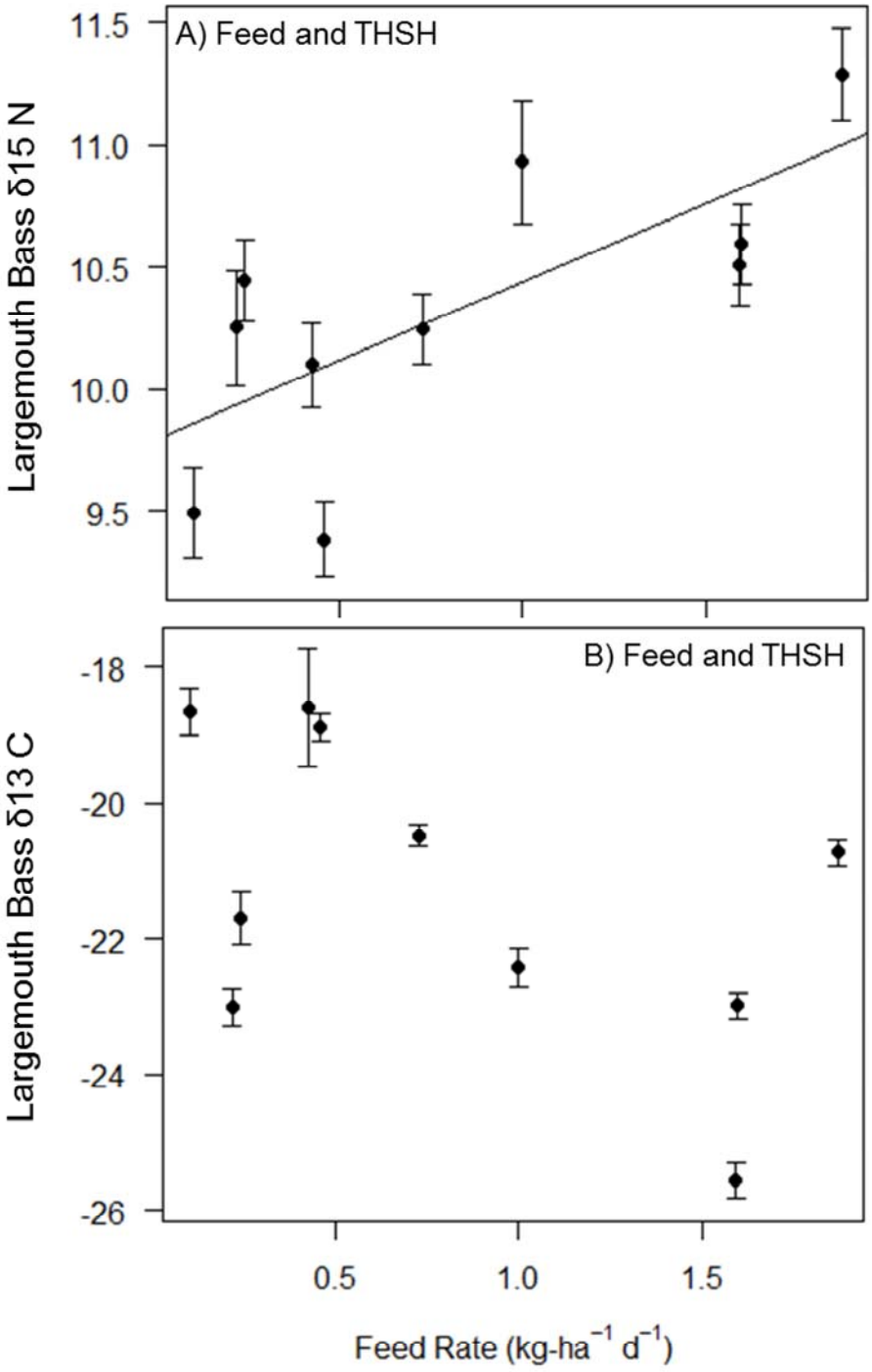
A) Feed and THSH
Largemouth Bass δ15 N
11.5
11.0
10.5
10.0
9.5
B) Feed and THSH
Largemouth Bass δ13 C
-18
-20
-22
-24
-26
0.5
1.0
1.5
Feed Rate (kg-ha^{-1} d^{-1})

Figure 22.

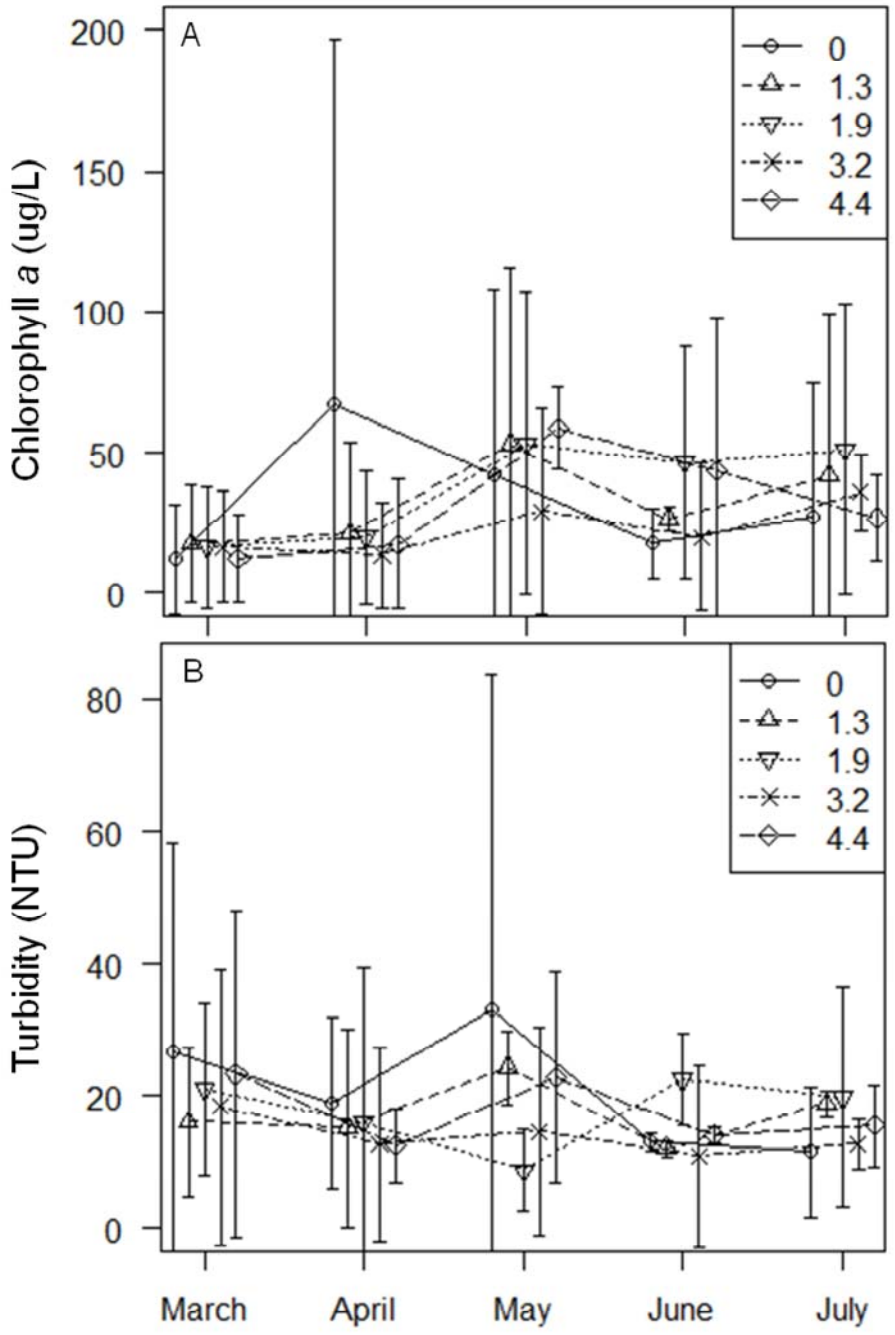
A
B
Chlorophyll a (ug/L)
Turbidity (NTU)
0
1.3
1.9
3.2
4.4
200
150
100
50
0
80
60
40
20
0
March
April
May
June
July

Figure 23.

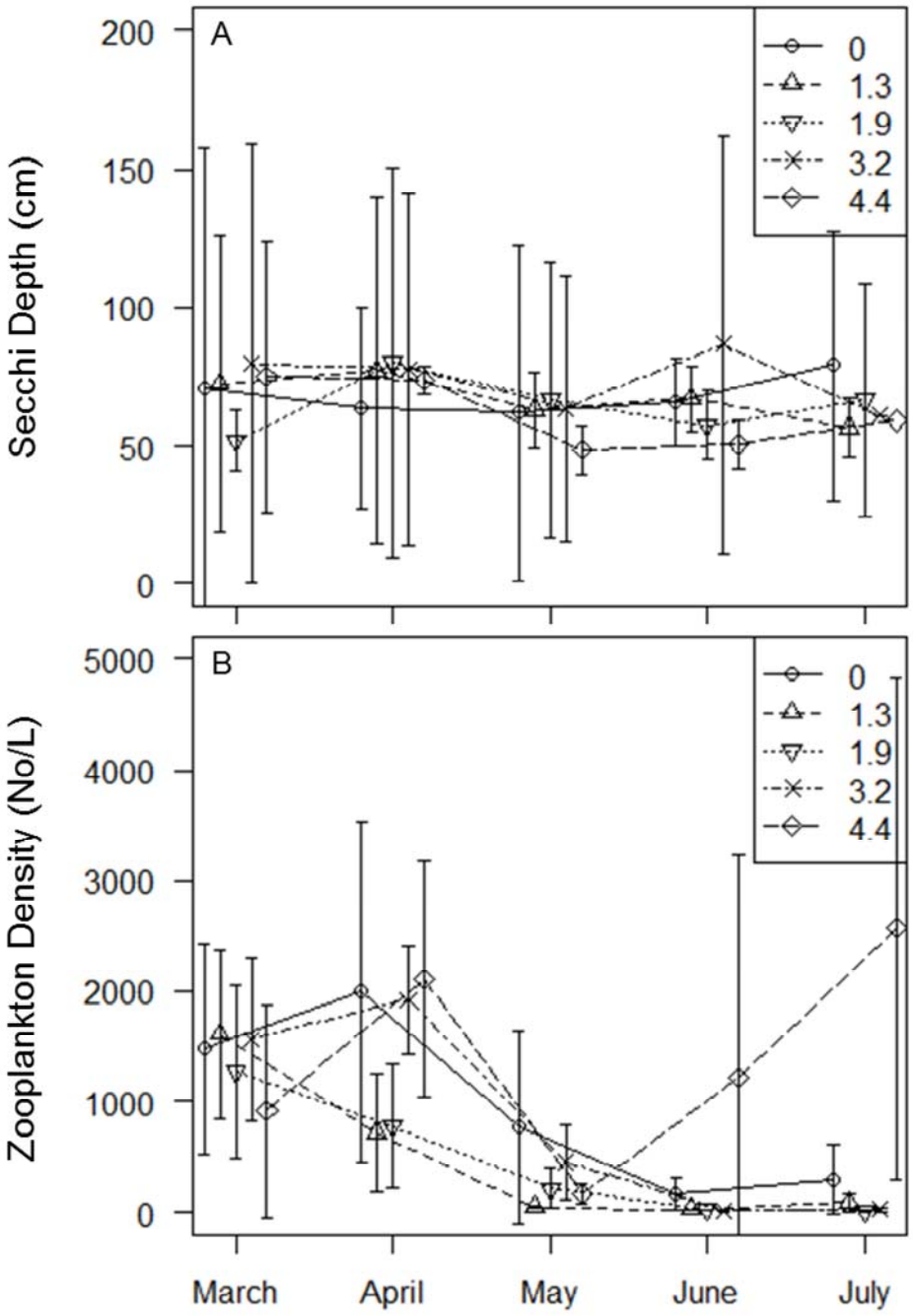
A
B
Secchi Depth (cm)
Zooplankton Density (No/L)
0
1.3
1.9
3.2
4.4
200
150
100
50
0
5000
4000
3000
2000
1000
0
March
April
May
June
July

Figure 24.

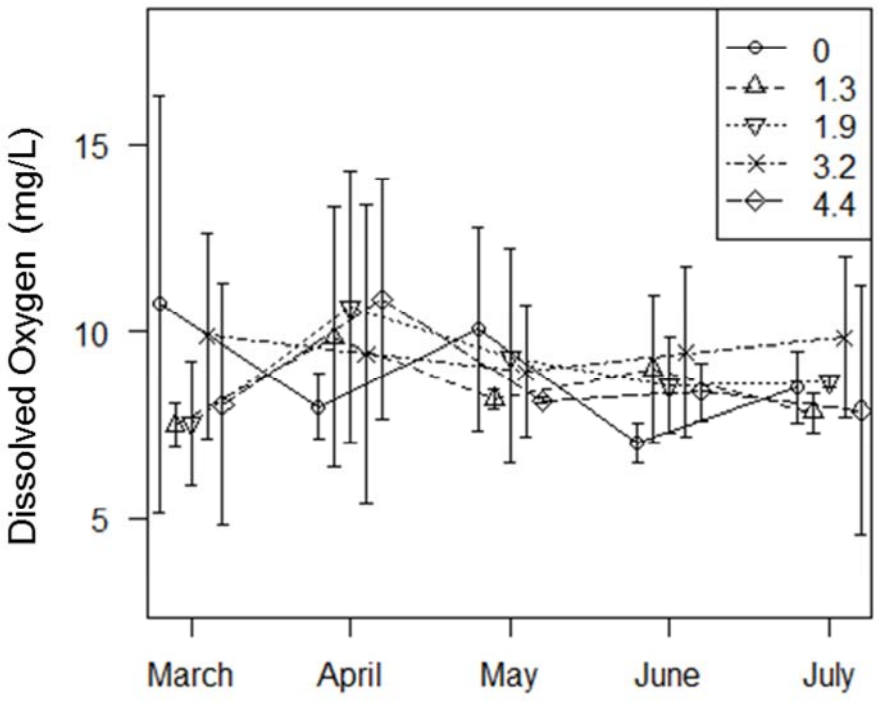
Dissolved Oxygen (mg/L)
15
10
5
March
April
May
June
July
0
1.3
1.9
3.2
4.4

Figure 25.

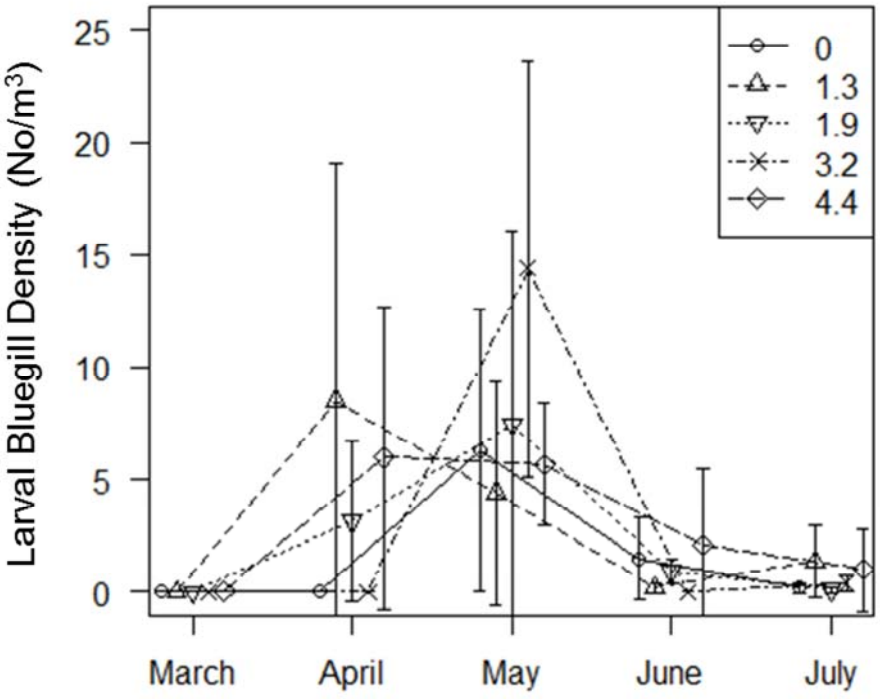
Larval Bluegill Density (No/m3)
25
20
15
10
5
0
March
April
May
June
July
0
1.3
1.9
3.2
4.4

Figure 26.

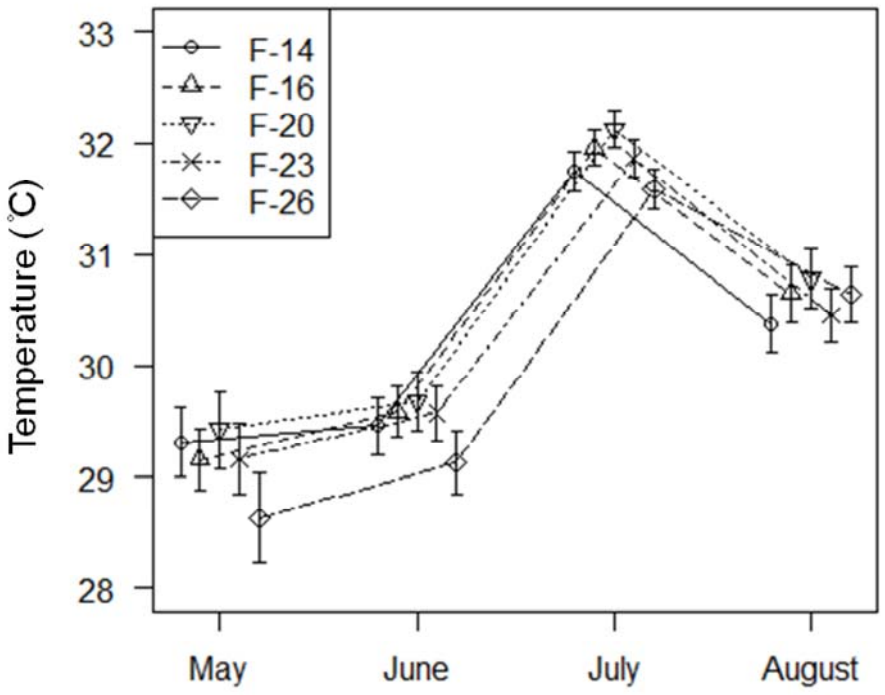
F-14
F-16
F-20
F-23
F-26
Temperature (°C)
33
32
31
30
29
28
May
June
July
August

Figure 27.

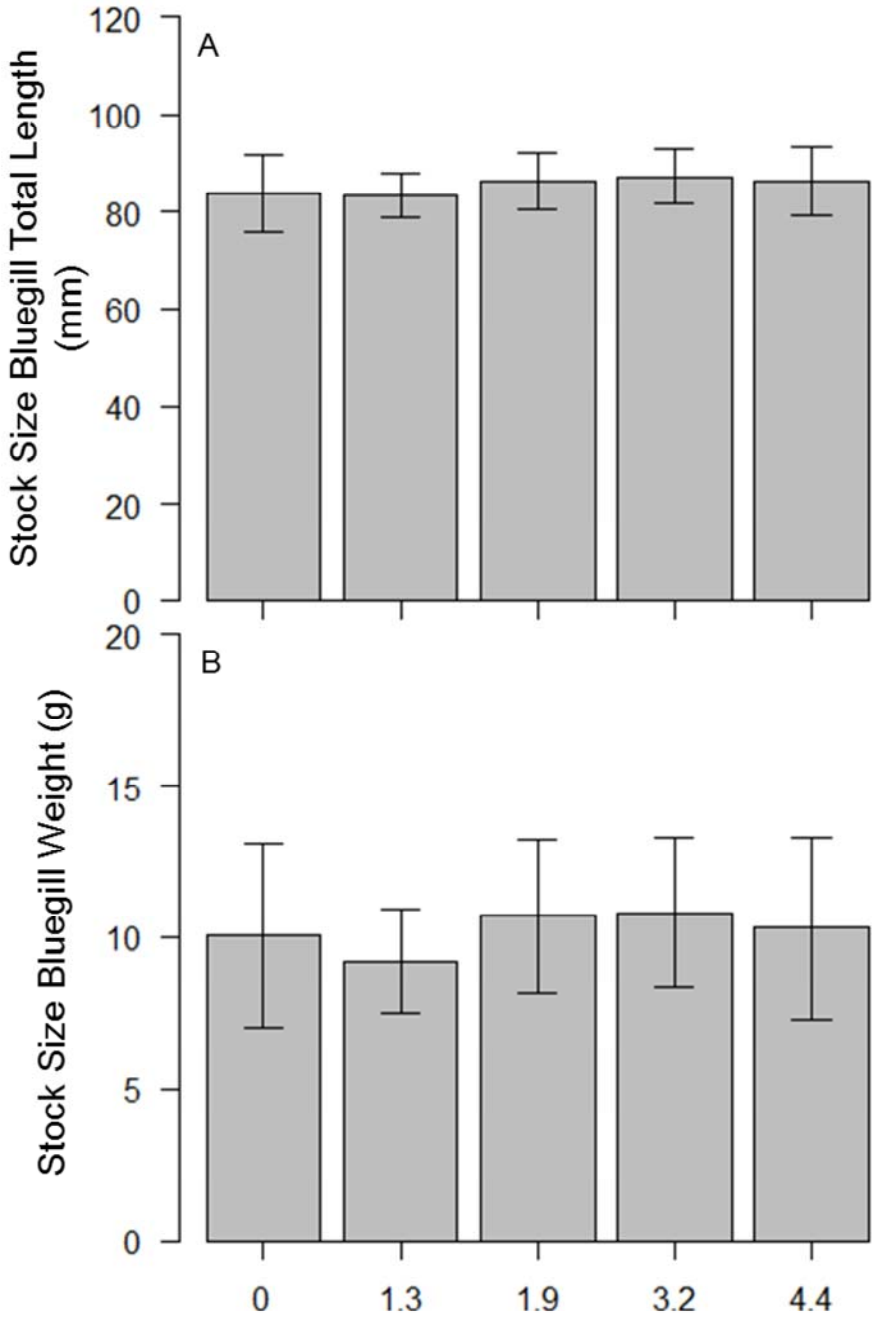
A
B
Stock Size Bluegill Total Length (mm)
Stock Size Bluegill Weight (g)
120
100
80
60
40
20
0
20
15
10
5
0
0
1.3
1.9
3.2
4.4

Figure 28.

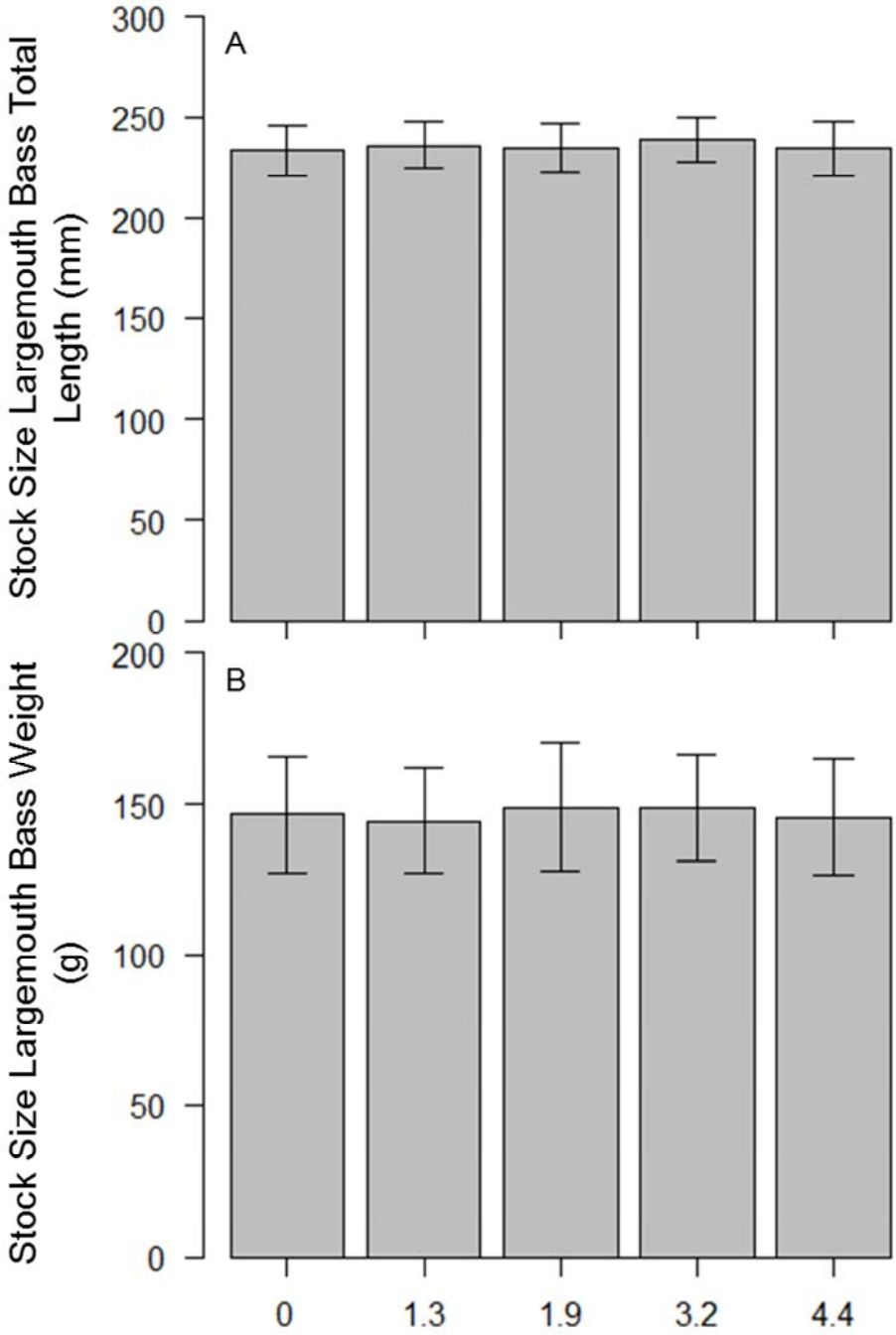
Stock Size Largemouth Bass Total Length (mm)
300
250
200
150
100
50
0
A
Stock Size Largemouth Bass Weight (g)
200
150
100
50
0
B
0
1.3
1.9
3.2
4.4

Figure 29.

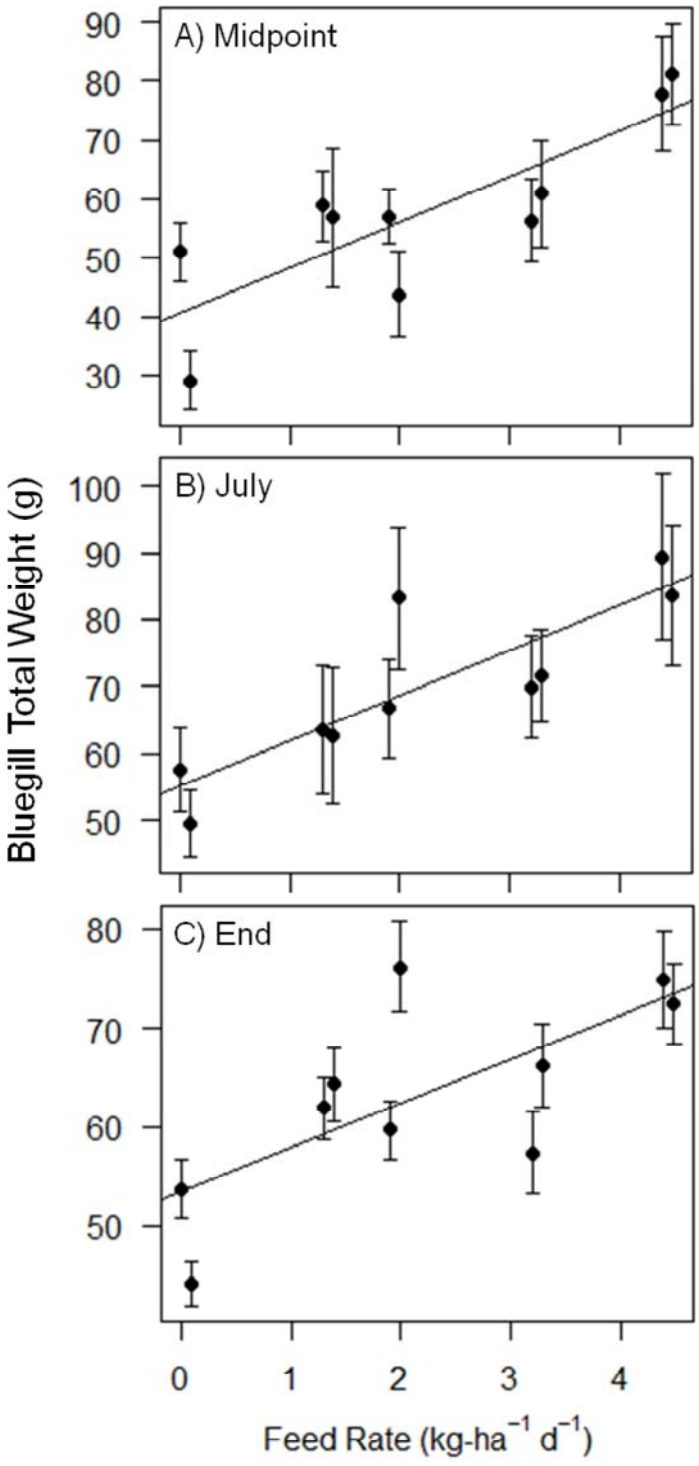
A) Midpoint
B) July
C) End
Bluegill Total Weight (g)
Feed Rate (kg-ha^{-1} d^{-1})
90
80
70
60
50
40
30
100
90
80
70
60
50
80
70
60
50
0
1
2
3
4

Figure 30.

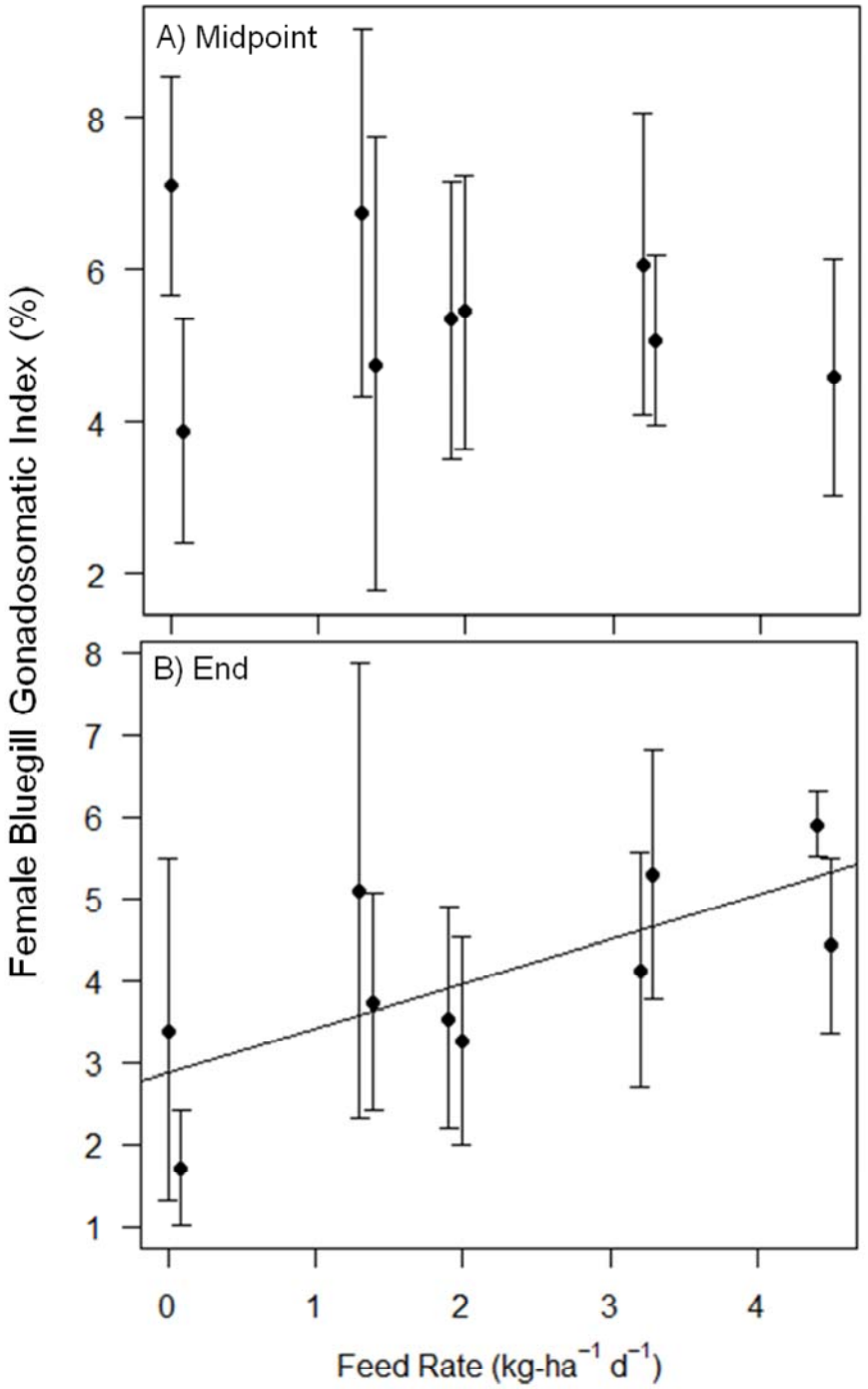
A) Midpoint
B) End
Female Bluegill Gonadosomatic Index (%)
Feed Rate (kg-ha^{-1} d^{-1})
0
1
2
3
4
1
2
3
4
5
6
7
8

Figure 31.

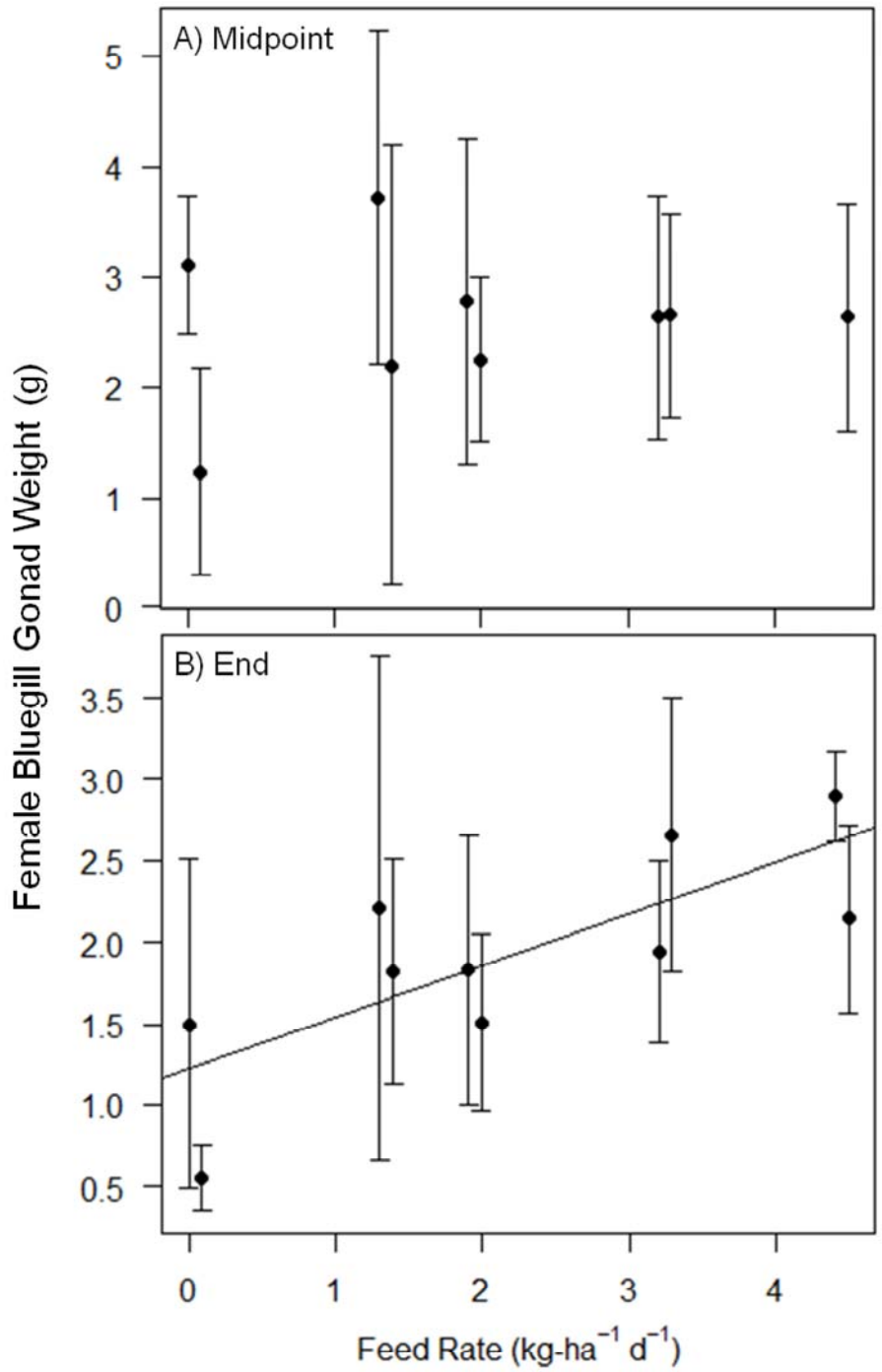
A) Midpoint
B) End
Female Bluegill Gonad Weight (g)
Feed Rate (kg-ha^{-1} d^{-1})
0
1
2
3
4
5
0.5
1.0
1.5
2.0
2.5
3.0
3.5

Figure 32.

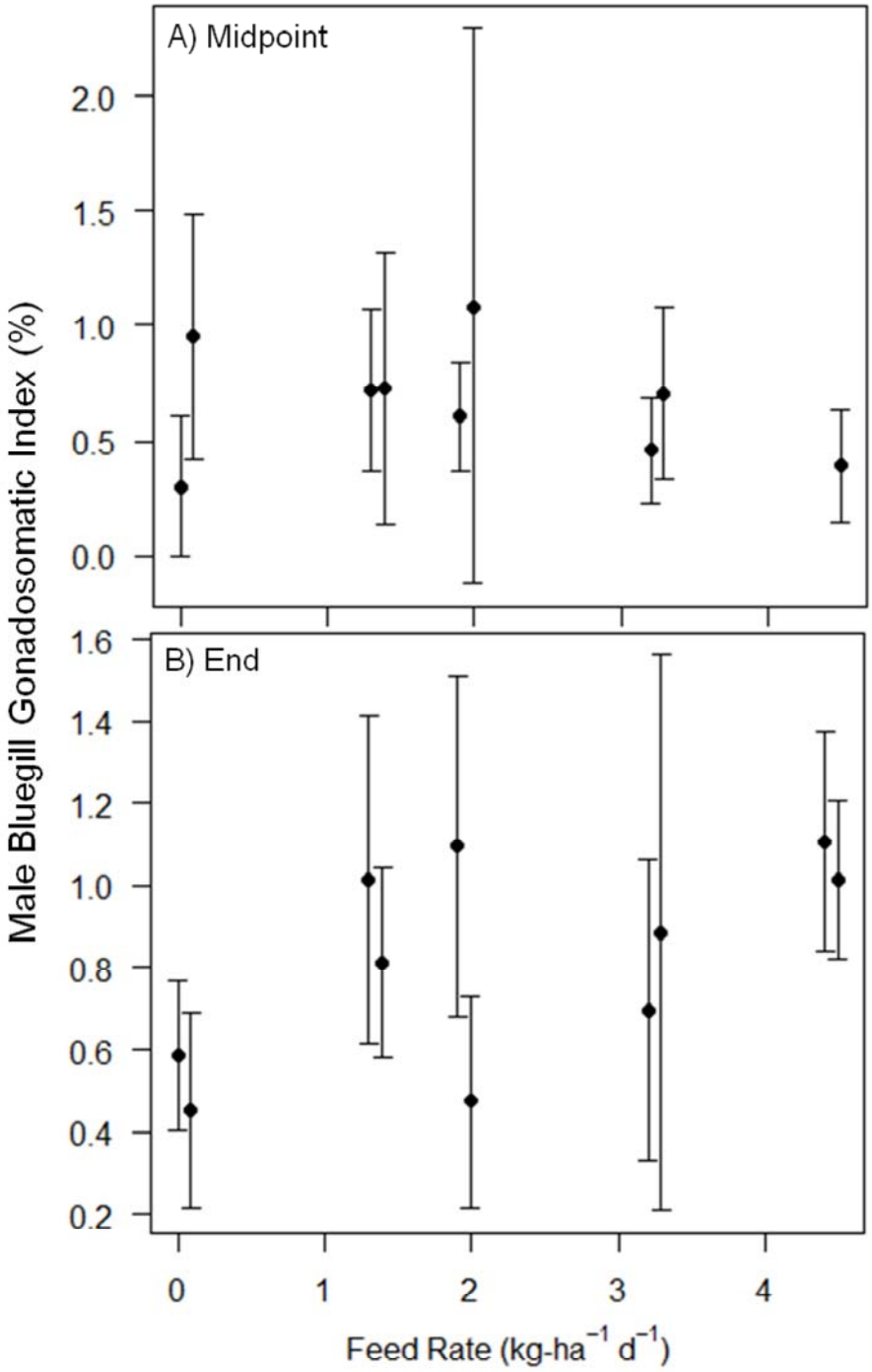
A) Midpoint
B) End
Male Bluegill Gonadosomatic Index (%)
2.0
1.5
1.0
0.5
0.0
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 33.

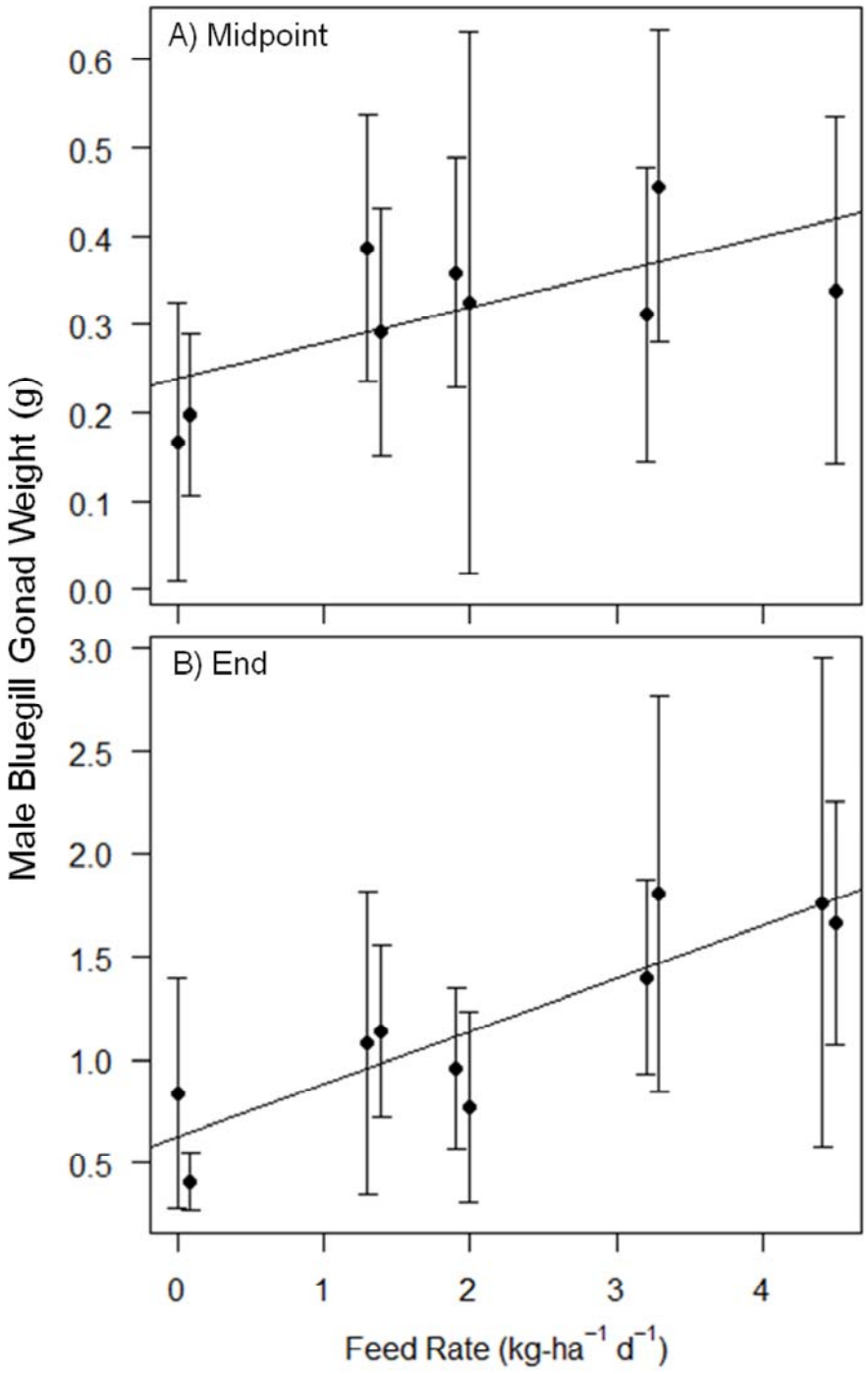
A) Midpoint
B) End
Male Bluegill Gonad Weight (g)
0.0
0.1
0.2
0.3
0.4
0.5
0.6
0.5
1.0
1.5
2.0
2.5
3.0
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 34.

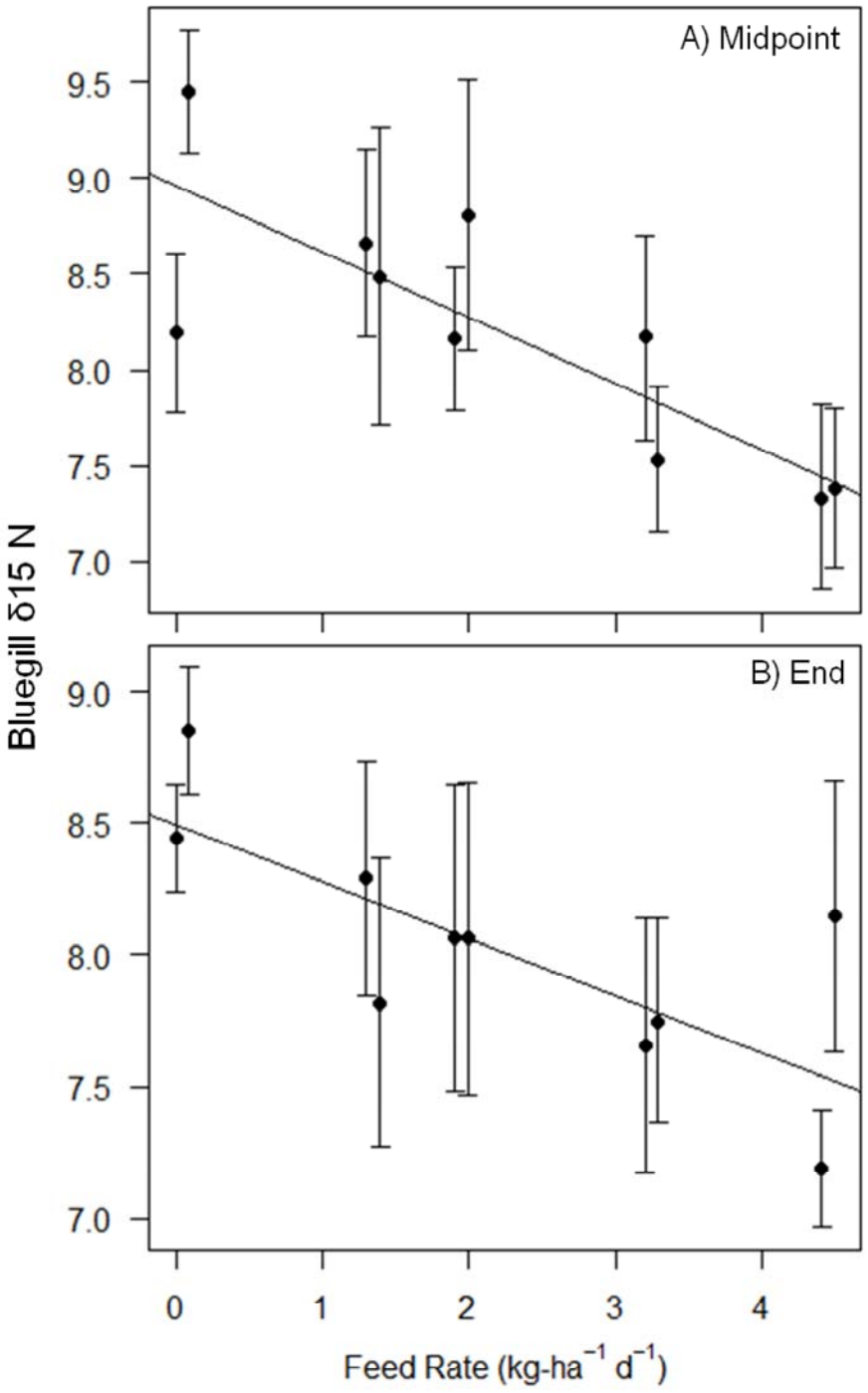
A) Midpoint
B) End
Bluegill δ15 N
9.5
9.0
8.5
8.0
7.5
7.0
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 35.

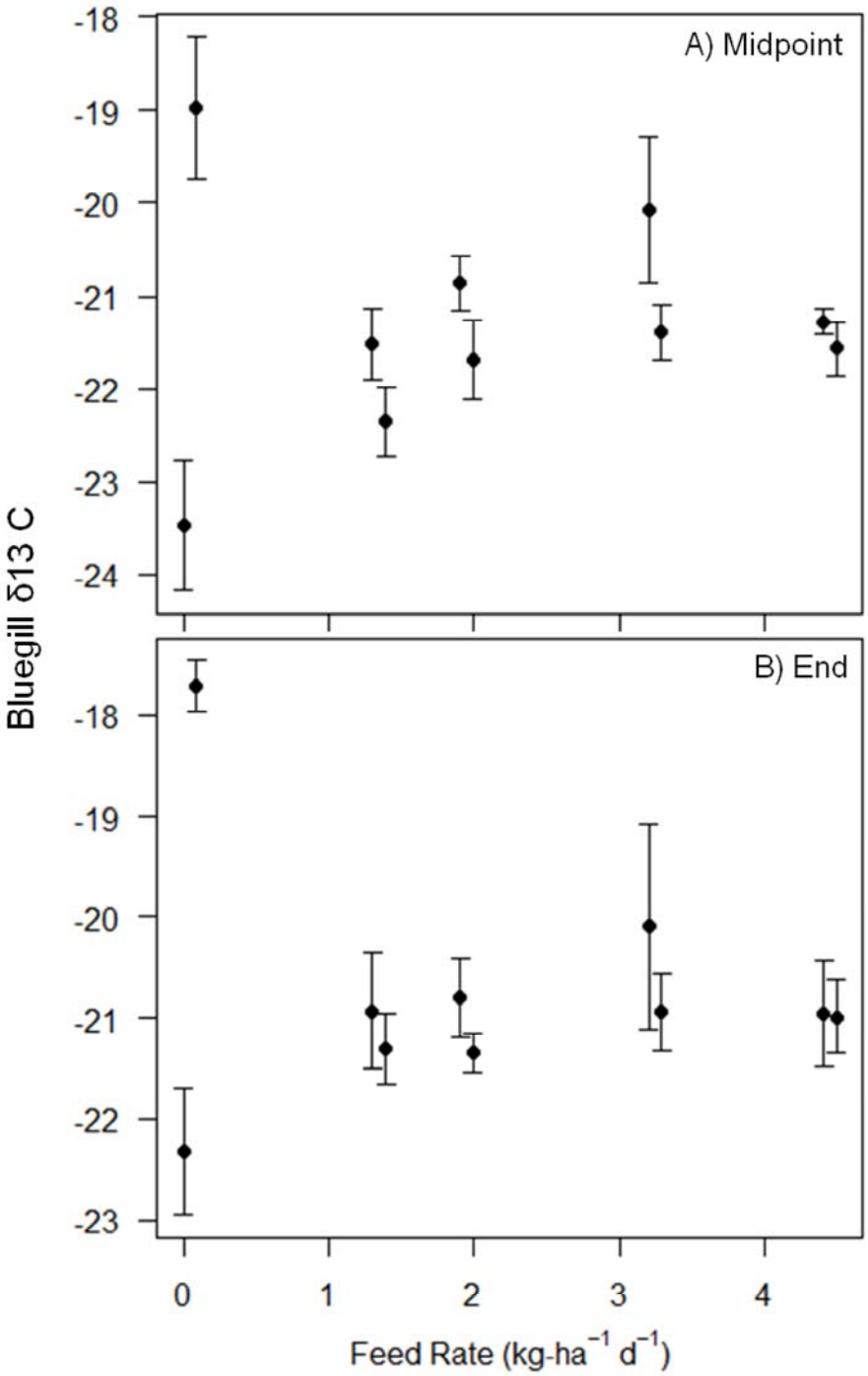
A) Midpoint
B) End
Bluegill δ13 C
-18
-19
-20
-21
-22
-23
-24
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 36.

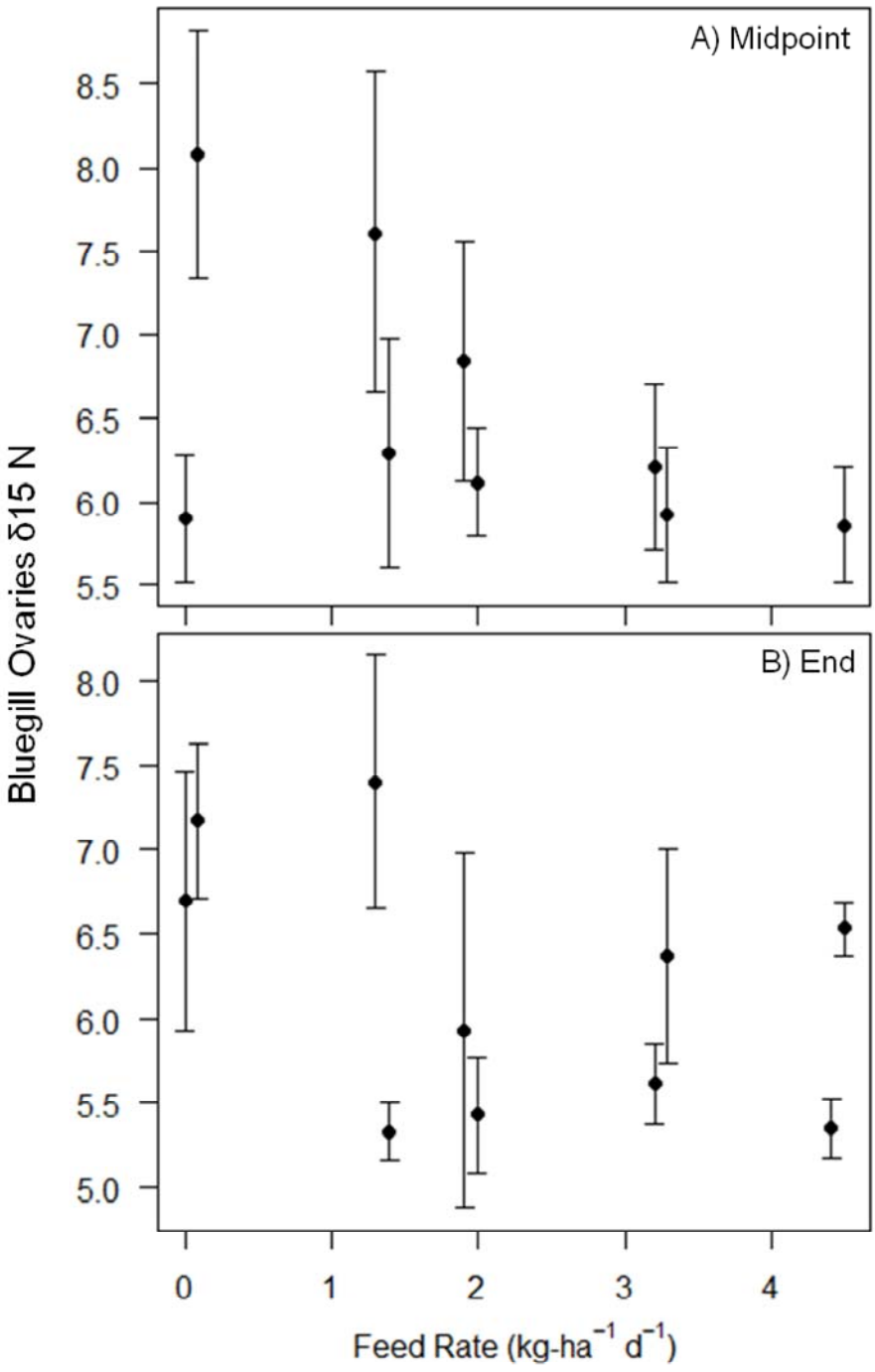
A) Midpoint
B) End
Bluegill Ovaries δ15 N
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 37.

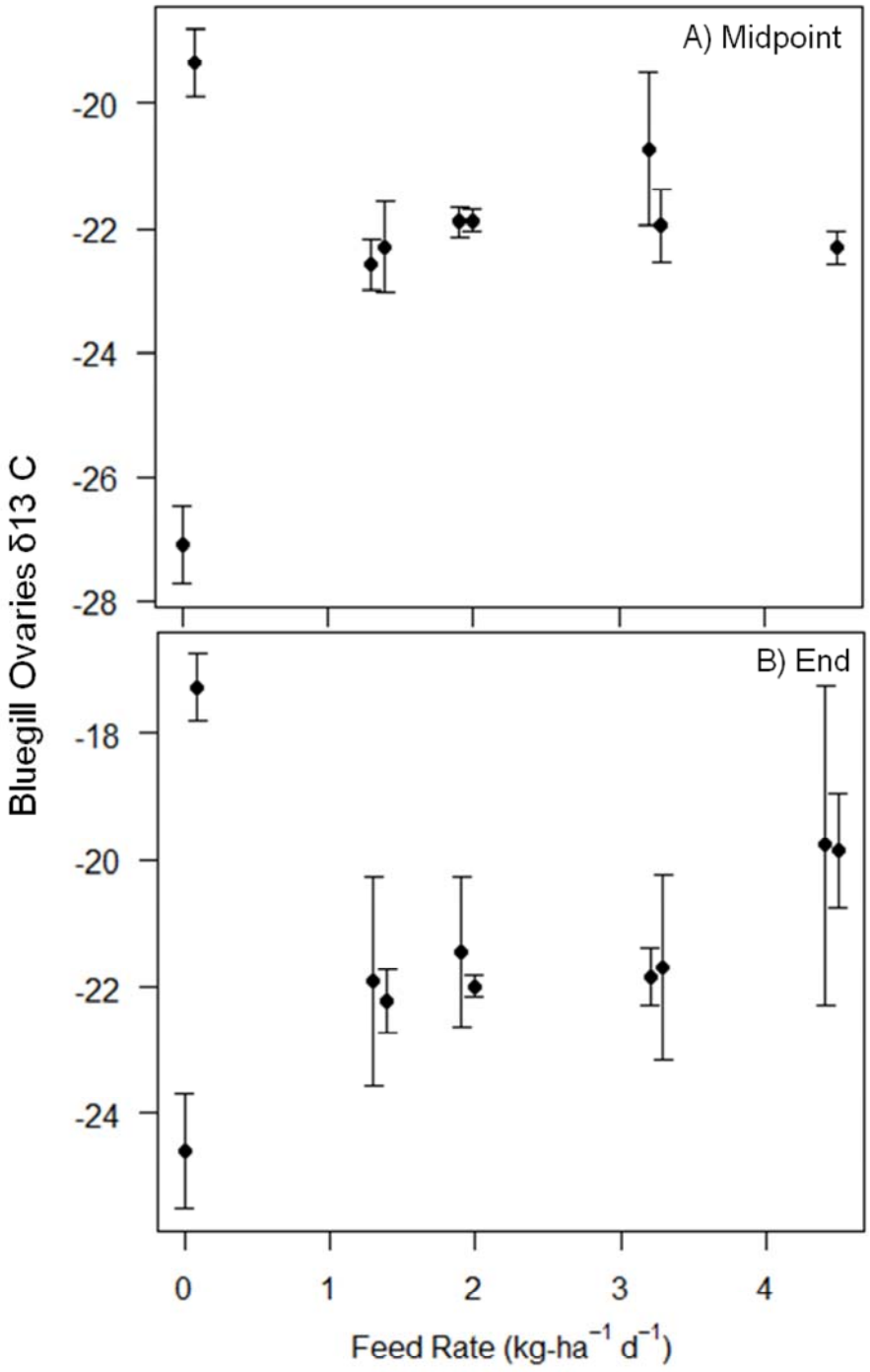
A) Midpoint
B) End
Bluegill Ovaries δ13 C
-20
-22
-24
-26
-28
-18
-20
-22
-24
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 38.

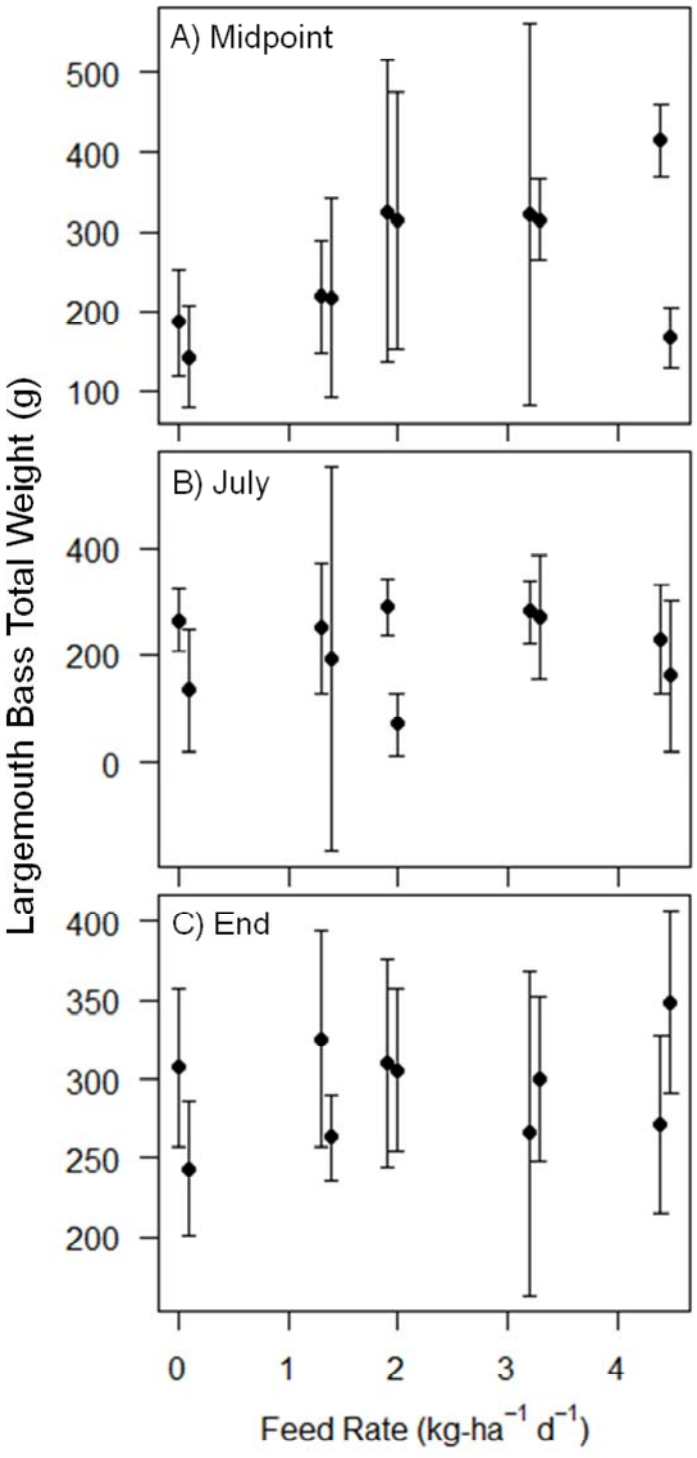
A) Midpoint
B) July
C) End
Largemouth Bass Total Weight (g)
Feed Rate (kg-ha^{-1} d^{-1})
500
400
300
200
100
0
350
250
1
2
3
4

Figure 39.

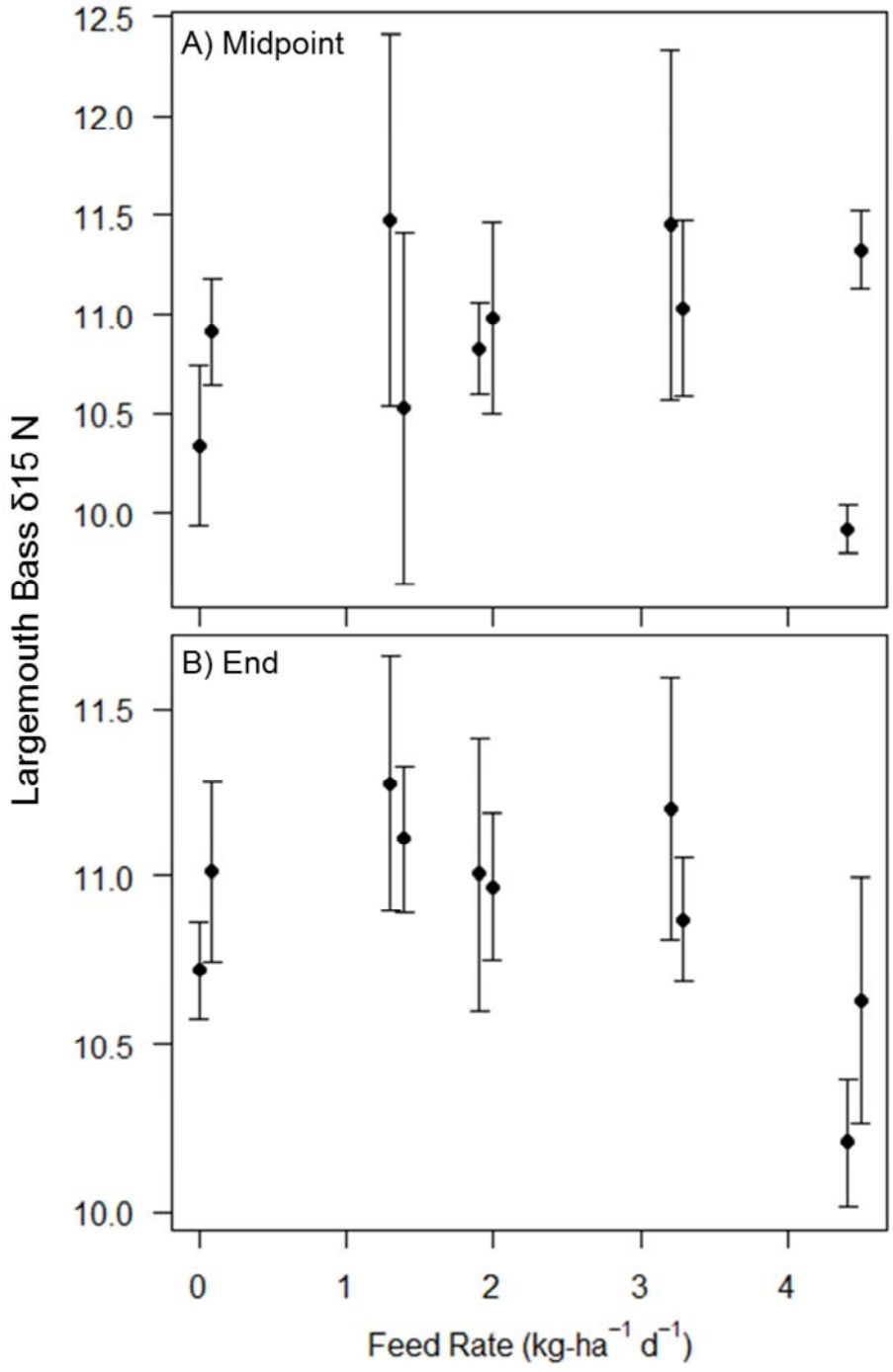
A) Midpoint
B) End
Largemouth Bass δ15 N
12.5
12.0
11.5
11.0
10.5
10.0
11.5
11.0
10.5
10.0
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 40.

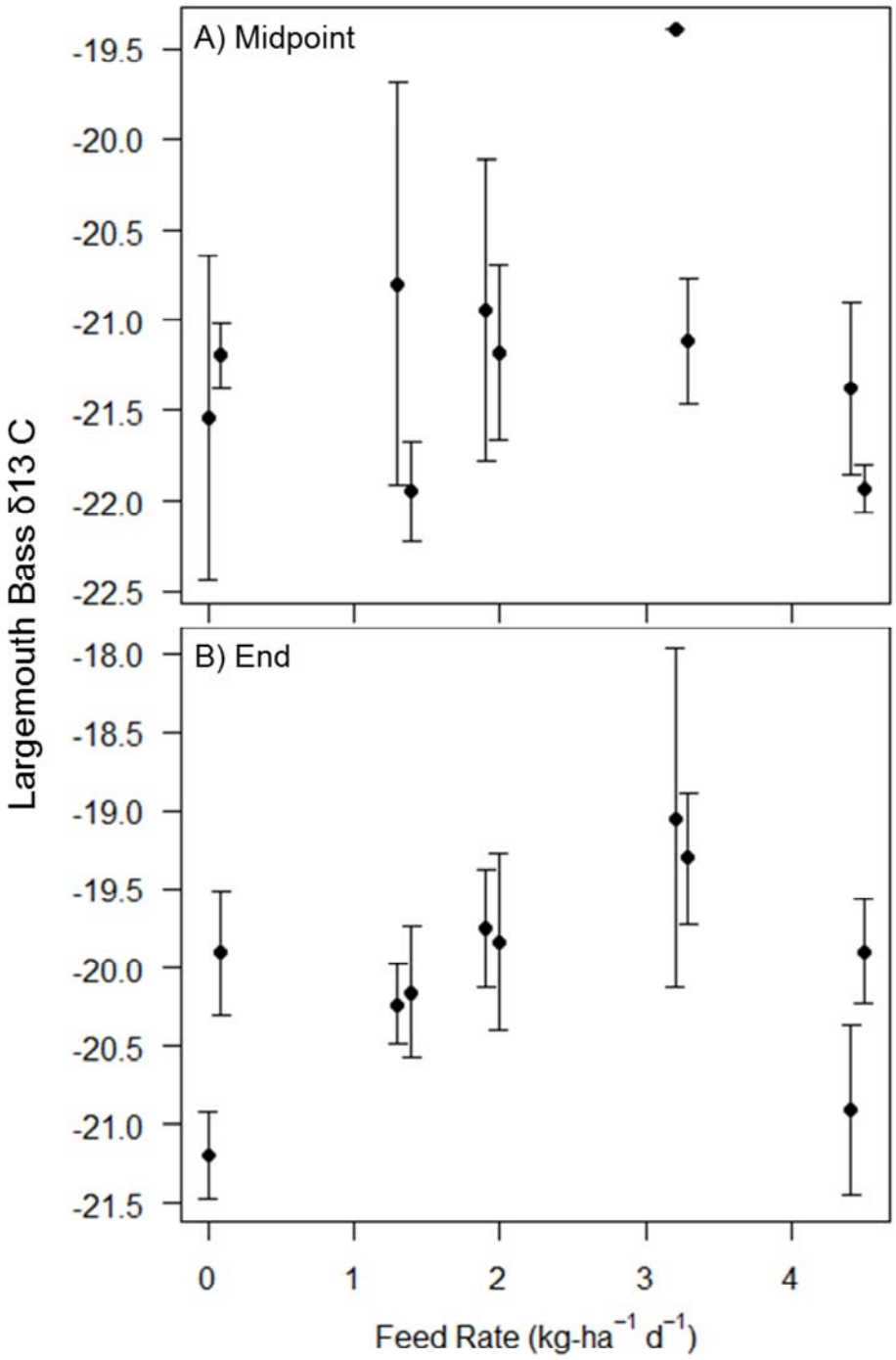
A) Midpoint
B) End
Largemouth Bass δ13 C
-19.5
-20.0
-20.5
-21.0
-21.5
-22.0
-22.5
-18.0
-18.5
-19.0
-19.5
-20.0
-20.5
-21.0
-21.5
0
1
2
3
4
Feed Rate (kg-ha^{-1} d^{-1})

Figure 41.

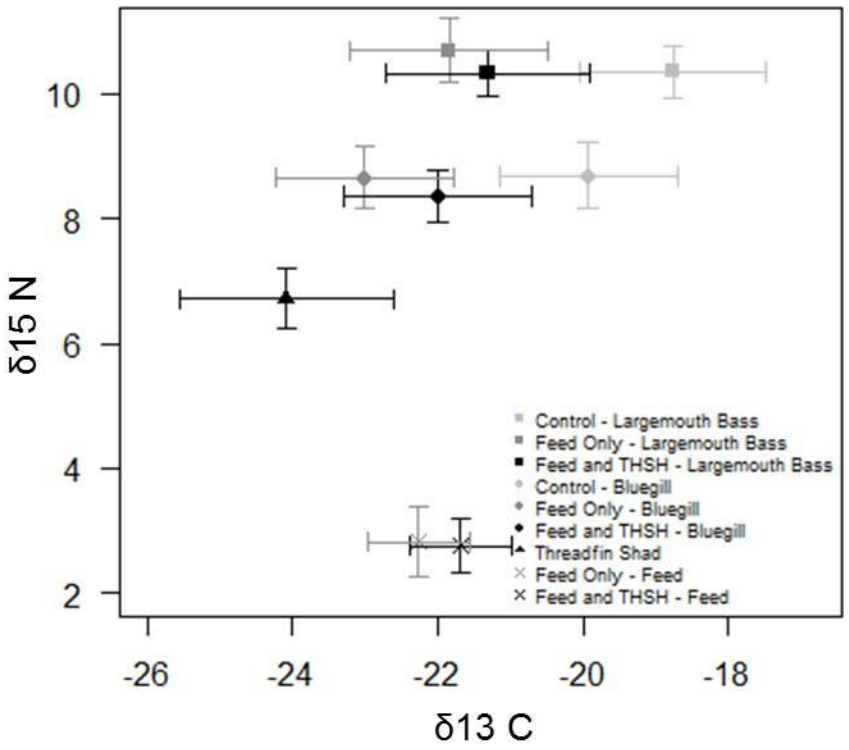
10
8
6
4
2
δ15 N
-26
-24
-22
-20
-18
δ13 C
Control - Largemouth Bass
Feed Only - Largemouth Bass
Feed and THSH - Largemouth Bass
Control - Bluegill
Feed Only - Bluegill
Feed and THSH - Bluegill
Threadfin Shad
Feed Only - Feed
Feed and THSH - Feed

Figure 42.