WINTER ENERGETICS OF YOUNG-OF-THE-YEAR BLUEFISH (*Pomatomus saltatrix*): EFFECTS OF RATION AND COHORT OF ORIGIN ON SURVIVAL

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ABSTRACT

The bluefish (*Pomatomus saltatrix*) population along the East Coast of the United States has experienced declines in both recruitment and adult abundance since the mid 1980s. At the end of their first growing season young-of-the-year (YOY) bluefish exhibit a bimodal length/frequency distribution consisting of larger, spring-spawned individuals (SP cohort) and smaller, summer-spawned individuals (SU cohort). While both SP and SU cohorts have been observed in the adult population in the past, recent studies have suggested that few SU-spawned individuals currently recruit to the adult stock. I investigated the hypothesis that the apparent recruitment failure of SU-spawned bluefish reflects negative size-selective overwinter mortality due to starvation. Due to mass allometries in energy storage and energy depletion, I predicted that larger, SP bluefish would 1) have greater energy stores prior to winter than smaller, SU bluefish, and 2) deplete their energy reserves at a slower rate than SU bluefish. Thus, I predicted that SP bluefish would exhibit greater overwinter survival (and therefore higher recruitment potential) than SU bluefish under starvation conditions.

Overwinter mesocosm experiments performed at ambient temperatures were conducted to examine the effects of cohort of origin (SP versus SU) and feeding level (fed versus unfed) on the overwinter survival of YOY bluefish. Energetic condition (non-polar lipid and ash content) and survival duration of bluefish subjects were monitored over the 192-day experiment.

SP-spawned bluefish possessed greater total lipid stores prior to winter than SU-spawned individuals, and both cohorts relied on multiple tissue depots (liver, viscera, white muscle, red muscle and skin) for the storage and mobilization of lipids. When

starved, SP and SU bluefish depleted their non-polar lipid reserves at similar rates over the first 31 days of the experiment. When food was present, both cohorts stored lipid at similar rates over the first 31 days of the experiment but depleted lipid reserves thereafter. This seasonal depletion pattern, despite the presence of food, indicates that lipid reserves are important for fueling routine metabolic requirements during winter and that bluefish may shift their energy allocation strategy from storage to mobilization/growth as winter progresses. When fed, both cohorts survived winter. When starved, SU bluefish began to exhibit starvation mortality six weeks prior to SP individuals. Although SU bluefish were more susceptible to overwinter starvation mortality than SP bluefish, their starvation endurance appears more than sufficient to permit overwinter survival under poor feeding conditions (>90% survival probability after 120 days without food and >60% after 150 days). Interestingly, SP bluefish suffered a brief mortality event during January when tank temperatures dropped below 6^{o}C, suggesting that SP individuals may be less cold tolerant than smaller, SU individuals. Wild YOY bluefish sampled from inner continental shelf waters off North Carolina during winter did not approach critical energy levels as determined from starved laboratory bluefish.

Given the high starvation endurance of SU-spawned YOY bluefish, I conducted a second winter experiment to assess the influence of forced activity and reduced pre-winter lipid storage on their overwintering ability. It was hypothesized that high activity level and reduced pre-winter lipid storage would increase the vulnerability of SU individuals to winter starvation. The experimental design was a fully-crossed 2X2 factorial design with activity level (high versus low) and pre-winter lipid storage (high

versus low) as factors. The high activity/low storage and low activity/high storage treatments were also tested in the presence and absence of winter food. Although the experiment was ended prematurely due to a system failure, lipid levels of bluefish at the time of death were quantified to examine whether the 2.5-month treatment exposures had measurable effects on bluefish energetics. Experimental results indicated that SU bluefish have a remarkable ability to store energy rapidly prior to winter. During a 30-day acclimation period SU bluefish were able to store more energy than was required to survive 2.5 months without food and at high (~0.8 body lengths sec^{-1}) activity levels. Also, pre-winter lipid storage had a greater effect on bluefish energy reserves than activity level. Furthermore, SU-spawned YOY bluefish appeared capable of assimilating food in the winter, if available, allowing them to compensate for reduced pre-winter lipid storage. These observations are consistent with the defended energy level hypothesis.

In conclusion, the remarkable starvation endurance ability of SU-spawned YOY bluefish, coupled with their capacity for rapid energy storage, and their ability to assimilate food during winter, indicates that SU bluefish are physiologically well-equipped to survive their first winter of life. These findings are consistent with recent energetics data reported for wild bluefish and do not support the overwinter starvation hypothesis as an explanation for the apparent recruitment failure of SU-spawned YOY bluefish.

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I wish to thank both Dr. Joan Willey and Dr. Robert Roer for their help and support over the past few years. I thank the Ichthyology Lab, both past and present, for unselfishly helping me maintain my experiments, interpret my data and retain my sanity. Also, I would like to thank all of my friends who stuck with me throughout my graduate experience.

Lastly, but most important, I would like to thank my family for their undying support throughout the graduate process. Who knows where I would be right now without you…

DEDICATION

This thesis is dedicated to my family. Thank you for everything that you have done for me.

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CHAPTER 1: EFFECTS OF COHORT OF ORIGIN AND FEEDING LEVEL

INTRODUCTION

The bluefish, *Pomatomus saltatrix*, is a coastal marine/estuarine fish in the Order Perciformes, Family Pomatomidae. Adult bluefish range in color from blue to green dorsally and silvery to white ventrally (Robins *et al*. 1986, Fahay *et al*. 1999). They have a darkish blotch at the base of their pectoral fins and a dusky, forked tail (Robins *et al*. 1986). Bluefish have a spiny dorsal fin that is separate from a long based soft dorsal fin, as well as a large, slightly superior mouth with prominent, flattened, and triangular teeth (Robins *et al*. 1986). *Pomatomus saltatrix* is a highly migratory, schooling species with a worldwide, subtropical distribution (Briggs 1960, Champagnat *et al*. 1983, Juanes *et al*. 1996). Along the East Coast of North America it ranges from Nova Scotia to the Florida Keys (Robins *et al*. 1986). Bluefish are thought to migrate north and south seasonally, as well as inshore/offshore, depending on prey location and water temperature (Fahay *et al*. 1999).

Bluefish eggs are spawned on the continental shelf of the SAB where they hatch and develop into juveniles (Fahay *et al*. 1999). Eggs are approximately one millimeter in diameter (Fahay 1983) and their incubation time ranges from 46-48 hours at 18-22^{o}C (Deuel *et al*. 1966). Following hatching, larval bluefish grow from ~2.0mm to 10-12mm standard length (SL) before they become pelagic juveniles, exhibiting most of the adult characteristics aside from scales (Hare and Cowen 1994, Fahay *et al*. 1999). At around 12mm bluefish start to develop scales, however it is not until ~37mm before this scale development is completed (Silverman 1975, Fahay *et al*. 1999). Physical processes such as wind-driven water currents and major ocean currents such as the Gulf Stream (Powles

1981, Lee and Atkinson 1983), along with their much-improved swimming abilities, help transport juvenile bluefish to the near-shore and estuarine habitats that serve as juvenile nursery habitats (Kendall and Walford 1979, Cowen *et al*. 1993, Hare and Cowen 1996). After entering estuaries, their growth rate increases dramatically (Juanes and Conover 1994).

Mature adult bluefish, usually age two and older, (Deuel 1964) spawn on the continental shelf starting in the South Atlantic Bight (SAB) in the springtime, as they start their annual migration northward (Kendall and Walford 1979). It is still highly debated whether these bluefish spawn continuously throughout their northward migration. Several investigations have suggested that spawning is a single, continuous event, but that young are lost from the middle portion resulting in the appearance of two discrete spawning events (Hare and Cowen 1993, Smith *et al*. 1994). Other researchers have argued that bluefish have multiple discrete spawning events (Chiarella and Conover 1990, McBride and Conover 1991). In either case, at least two and sometimes three distinct cohorts of young-of-the-year (YOY) bluefish appear in most years (Nyman and Conover 1988, McBride 1989). These different cohorts are termed spring-spawned, summer-spawned, and fall-spawned (Juanes *et al*. 1993, McBride *et al*. 1993). The spring cohort is generally composed of larger, older individuals spawned in the SAB in March-May, whereas summer cohort individuals are usually smaller, younger and presumably spawned in the Middle Atlantic Bight (MAB) in June-August (McBride *et al.* 1993). The fall cohort consists of the smallest body-sized YOY in years when it is present and is spawned in the SAB in September-January (McBride *et al*. 1993).

Bluefish spawned in the spring in the SAB recruit to estuaries in both the SAB and MAB, with assistance from the northward flowing Gulf Stream current (Kendall and Walford 1979, Collins and Stender 1987, McBride and Conover 1991, Cowen *et al*. 1993, McBride *et al*. 1993, Hare and Cowen 1996). Bluefish spawned in the summer in the MAB recruit only to MAB nurseries (Kendall and Walford 1979, Nyman and Conover 1988, McBride and Conover 1991, Able and Fahay 1998). The spawning location and juvenile habitats of fall-spawned YOY bluefish are unclear. Several researchers have used both scale analysis to demonstrate the presence of these YOY bluefish cohorts (Lassiter 1962, Chiarella and Conover 1990) and otolith analysis to determine their birthdates (Nyman and Conover 1988, Gilmore 2000).

After their spring spawning event in the SAB, adult bluefish start a long migration northward to the cooler waters of the MAB (Fahay *et al*. 1999). Here they spend their summer months feeding on anchovies, menhaden and other forage fishes, presumably to maintain themselves, to recover energy lost during breeding and migration and to store energy for their annual southward fall migration and the upcoming winter (Hartman and Brandt 1995a,b, Fahay *et al*. 1999). When ocean temperatures begin to decline in the fall, the adults leave the MAB and return to the SAB to overwinter (Fahay *et al*. 1999).

The different cohorts of YOY bluefish display similar growth rates due to the inability of late-spawned individuals to exhibit compensatory growth (McBride *et al*. 1993, Buckel *et al*. 1998). Therefore, the amount of time each fish has to grow before its first winter (which is determined by its date of birth) determines its body size at the onset of winter. This leads to the bimodal (occasionally tri-modal) length-frequency distribution of YOY bluefish at their fall estuarine egress (Wilk 1977, Kendall and

Walford 1979, Nyman and Conover 1988, McBride and Conover 1991, McBride *et al*. 1993). In the MAB, before their fall migration, spring-spawned YOY are more than twice the average length of summer-spawned YOY. During this time summer-spawned YOY average 120-140mm SL, while spring-spawned YOY average 240-280mm SL (Kendall and Walford 1979, Chiarella and Conover 1990, McBride and Conover 1991, Gilmore 2000). Thus, each cohort enters the winter at markedly different body sizes.

The difference in body size between spring- and summer-spawned YOY bluefish may have important implications for survival and recruitment potential (Sogard 1997, Campana 1996). Bluefish populations off of the East Coast of the United States appear to have experienced declines in both recruitment and adult abundance since the mid 1980s (Munch and Conover 2000). While Baird (1873) has shown that the bluefish population fluctuates naturally, the mechanisms responsible for these recent declines are unknown. Both Chiarella and Conover (1990) and Gilmore (2000) have shown that, recently, spring-spawned bluefish appear to be the main contributors to the adult stock, while summer-spawned bluefish are rare in the adult stock (Gilmore 2000). However, in 1960 and 1961, Lassiter (1962) found that both cohorts were equally present in a sample of age-1 bluefish. Sometime after the YOY summer and fall cohorts leave their estuaries in the fall and the age one and older bluefish return in the spring, the summer and fall cohorts seem to disappear.

There are several possible explanations for this apparent disappearance of summer- and fall-spawned bluefish in the adult population. First, the accuracy of the method used to back-calculate the birth date of adult bluefish might be flawed. However, this method was recently validated by Fenwick and Conover (unpublished). Second, the

summer cohort could be experiencing compensatory growth and catching up in body size to the spring cohort before their first birthday (Sogard 1997). This would give the impression of the summer cohort's disappearance despite their presence in the adult population. This explanation has not been shown to occur (McBride *et al*. 1993, Buckel *et al*. 1998). Third, the summer cohort may recruit to areas other than the MAB. Lastly, the summer cohort may not be contributing to the adult population in recent years due to negative size-selective overwinter mortality (Sogard 1997).

Winter represents a potentially stressful period for young fishes (Johnson and Evans 1990). Overwinter mortality can be an important factor regulating the recruitment success in fish populations (Toneys and Coble 1979, Shuter *et al*. 1980, Post and Evans 1989, Lankford and Targett 2001). During the winter, temperatures decrease and either acute or chronic cold stress might negatively affect survival (Lewis 1965, Holt and Holt 1983, Uphoff 1989, Thompson *et al*. 1991, Johnson and Evans 1996, Schultz and Conover 1999, Lankford and Targett 2001). Food limitation, decreased digestion rates and/or activity costs (e.g. migration) may lead to the exhaustion of energy reserves (Shul'man 1974, Oliver *et al*. 1979, Cunjak *et al*. 1987, Post and Evans 1989b, Thompson *et al*. 1991, Sogard 1997, Foy and Paul 1999, Hurst and Conover 2001, see also Schultz and Conover 1999). Chronic starvation also may function to weaken individuals, increasing their susceptibility to predation (Furuta 1998, Skajaa *et al*. 2003). Smaller individuals of a species are thought to be more vulnerable to winter starvation because of their higher mass-specific metabolic rate and reduced capacity for energy storage (Oliver *et al*. 1979, Post and Evans 1989b, Schultz and Conover 1999). In contrast, larger fishes

may be more vulnerable to acute cold stress due to lower rates of metabolism and protein turnover (Lankford and Targett 2001).

I tested the hypothesis that the apparent recruitment failure of summer-spawned YOY bluefish reflects negative size-selective overwinter mortality due to starvation. I predicted that if both spring and summer cohorts were held without food, the summer cohort would experience higher overwinter mortality associated with a faster rate of energy depletion. I also predicted that if both cohorts were fed, they would both experience a low overwinter mortality. In addition, I predicted that the energetic condition of wild fish would approach critical lipid levels as determined from mesocosm experiments (described below).

METHODS

Experimental Subjects:

Experimental subjects were collected by hook and line from the Atlantic Intra-Coastal Waterway (AICWW) at the University of North Carolina at Wilmington (UNCW) Center for Marine Science (CMS) dock and from Beaufort Inlet, NC from mid-October to mid-November 2001. Subjects were transported to the UNCW CMS and placed into 2,000-L circular fiberglass tanks. Tanks were supplied with ambient flow-through seawater (~20L/minute) obtained from the AICWW adjacent to the CMS research pier.

Mesocosm Setup:

The outdoor mesocosm setup was a flow-through system containing twelve 2,000-L tanks (1.8 meter diameter). Seawater was obtained from the AICWW at ambient salinity and temperature. Water temperature ranged from <6^{o}C at the end of December/beginning of January to 32^{o}C in May (Figure 1).

Experimental Design:

A fully-crossed 2X2 factorial design was used to test for effects of cohort (spring (SP) vs. summer (SU)) and feeding regime (fed vs. unfed) on the overwinter survival ability of YOY bluefish. Three replicate tanks were used for each treatment combination (Figure 2). On November 19, 2001 (day 0), each tank (n=6 for SP, n=6 for SU) was stocked with 15 bluefish and the experiment was initiated. On November 20, 2001, more SU cohort bluefish were captured and 3 to 4 more individuals were added to each SU cohort tank. SP bluefish FL ranged from 225-311mm, with a mean value of 264mm; while SU bluefish FL ranged from 179-229mm, with a mean value of 206mm (Figure 3). Cohort assignment was based on bimodal length/frequency distributions of YOY bluefish collected in NC during fall 2001 (Morley 2004).

During their acclimation period, all bluefish were fed to satiation once daily on a diet of dead bay anchovies (*Anchoa mitchilli*) and Atlantic silversides (*Menidia menidia*). Both species are known to be natural prey items of YOY bluefish in the wild at this time of year (Buckel and Conover 1997, Buckel *et al*. 1999). All feedings were performed in the early evening before dusk. Starting November 19, 2001, only fed treatments continued to receive this ration while unfed treatments received none. All tanks were

checked for mortalities at least twice per day, with any mortalities being immediately frozen for later analyses. Temperature was recorded throughout the experiment using a miniature data logger (Onset Computer Corp., Pocasset, Massachusetts), which recorded water temperature at 30-minute intervals.

Bluefish were sampled from each treatment on predetermined dates (days 0, 11, 31, and 89) of the experiment to monitor energetic condition of tissues. On these days three individuals were removed haphazardly from each replicate tank and immediately frozen for later analysis. Natural mortalities were also analyzed for energetic condition. The entire liver and a small sample (~0.5-2.0g wet weight) of epaxial white muscle (WM) tissue were removed from each fish and immediately flash frozen until lipid extractions were performed. In addition, other body depots (viscera, red muscle (RM), and skin) were dissected from a subsample of bluefish sampled on each date in order to determine the general distribution of lipids in this species. On day 192 all remaining experimental subjects were sacrificed and their tissues analyzed in a similar manner.

Lipid Extraction:

Total nonpolar lipids were extracted from bluefish tissues using the Soxhlet dry extraction protocol described in Schultz and Conover (1997). Nonpolar lipids were monitored because this class includes the triacylglycerols (TAG's), which are the principal energy storage lipids in fishes (Jobling 2001a). Prior to extractions, cellulose thimbles (22mmX80mm) were dried at 60^{o}C for at least 48 hours. WM, RM, viscera, and skin samples were thawed, blotted gently, and placed in a tared, pre-labeled polystyrene dish. Wet weight was measured to ±0.0001 gram (g). Tissues were then

dried at 60°C for at least 48 hours. Liver samples were handled in a similar fashion, except that they were not blotted prior to obtaining wet weights.

Soxhlet extractor flasks were filled (~110mL) with clean petroleum ether and heated sufficiently to produce a cycle rate of approximately 7.5 cycles/hour. Pre-dried thimbles were labeled and weighed (±0.0001g). Dry tissue samples to be extracted were weighed, placed into thimbles and put into the Soxhlet extraction devices. Samples were extracted for 4 hours to remove all non-polar lipids then dried at 60°C for a minimum of 48 hours. Samples were then re-weighed to obtain a post extraction thimble/tissue dry weight. This weight was then subtracted from the combined weight of the thimble pre-extraction weight and the tissue pre-extraction weight. The difference represents the weight of total nonpolar lipids removed from the tissue sample.

Ashing Data:

Ash content of tissues was measured as an indicator of their energetic condition (Ali *et al*. 2001). Lean (extracted) tissues were dried at 60°C for at least 48 hours prior to ashing. Quartz crucibles (20mL) were pre-ashed at 450°C for 24 hours prior to use. Pre-ashed crucibles were weighed (±0.0001g) and tared. Next, a tissue sample's dry weight was measured (±0.0001g) by placing it in the tared crucible. Samples were then ashed in a muffle furnace (Thermolyne 1400) at 450°C for 24 hours. Crucibles were then placed in a drying oven and allowed to cool to 60°C before being re-weighed. The ash weight was measured and expressed as a percentage of lean tissue dry weight. This ratio of inorganics to organics in the lean tissue was termed ash content (g ash/g lean tissue*100).

As starved fish utilize energy reserves, the ash content of their body tissues increases (Ali *et al*. 2001, Jobling 2001b).

Statistical Analyses:

Lipid Energetics:

Initial Condition:

Initial (day 0) lipid data were analyzed using two-way ANOVA to test for differences between cohorts and among body depots at the start of the experiment. For these analyses fed and unfed bluefish were combined within their appropriate cohorts, since day 0 subsamplings were performed prior to any feeding manipulation. Both lipid content (g) and lipid density (%) for liver, viscera, WM, RM, and skin were individually analyzed and compared across cohorts. A significance value of $\alpha=0.05$ was used. Significant main effects were analyzed post-hoc using Tukey multiple comparisons tests. Prior to ANOVA, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using an extension of the Kruskal-Wallis ANOVA by ranks to individually test for cohort and body depot effects.

To examine whether lipid values (content and density) were correlated across different body depots (liver, viscera, WM, RM, and skin), lipid data from each cohort were subjected to separate correlation analyses using the Pearson product-limit method in Statistica version 6.0.

Energetic Condition of Body Depots: Day 0-31(89)

Day 0, 11, 31, and 89 (SU cohort only) lipid data were analyzed across all body depots using separate one-way repeated-measures ANOVA within each treatment (spring fed (SP_F), summer fed (SU_F), spring unfed (SP_U) and summer unfed (SU_U)) to assess the energetic role of different body depots in lipid storage and depletion in overwintering YOY bluefish. Lipid content and lipid density data were individually tested for each treatment. Due to insufficient numbers, final (day 192) data were not included in the analyses. A significance value of $\alpha=0.05$ was used. Body depots included in the ANOVA were liver, viscera, WM, RM, and skin. Significant main effects were analyzed post-hoc using Tukey multiple comparisons tests. Prior to ANOVA, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. Occasionally, data remained somewhat heteroscedastistic following transformation; for these data, it was assumed that the ANOVA was robust enough to perform well despite the deviation from homoscedasticity (Zar 1984).

Effects of Feeding and Cohort on Energetics:

Initial (day 0) subsamples were analyzed using T-tests to test for differences between cohorts at the start of the experiment. For these analyses fed and unfed bluefish were combined within their appropriate cohorts, since day 0 subsamplings were performed prior to any feeding manipulation. Body condition indicators analyzed were: nonpolar lipid content of liver in grams (g), liver lipid density ((g liver lipid/g liver dry

weight (DWT))*100) in percent (%), liver lipid density (g liver lipid/bluefish fork length (FL) in millimeters (mm)), liver DWT (g)/FL (mm), WM lipid density (%), wet weight (g) (WWT)/FL (mm) and mean FL (mm). A significance value of α=0.05 was used. Prior to T-tests, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using the Mann Whitney U-test (MWU).

Day 11 and 31 lipid data were each analyzed using two-way ANOVA to test for any differences between cohorts and between feeding treatments on these days. Body condition indicators analyzed were the same as stated for day 0. A significance value of α=0.05 was used. Significant main effects were analyzed post-hoc using Tukey multiple comparisons tests. Prior to ANOVA, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using an extension of the Kruskal-Wallis ANOVA by ranks to individually test for cohort and feeding effects.

Day 89 lipid data were analyzed using T-tests to test for a feeding effect in the SU treatments. Treatments containing SP bluefish were not subsampled on day 89 and, therefore, could not be analyzed. Body condition indicators investigated were the same as stated for day 0. A significance value of α=0.05 was used. Prior to T-tests, variances were tested for homogeneity using Levene's test. If variances were found to be

significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using the MWU-test.

Final (day 192) lipid data were analyzed using a T-test to test for a cohort effect between the SP_F and SU_F treatments. Numbers of unfed bluefish from either treatment were insufficient for any other analyses. Final data were analyzed as stated for day 89.

Energetic Condition: Day 0-31(89)

Day 0, 11, and 31 lipid data were analyzed using one-way repeated-measures ANOVA to individually test for any cohort (SP_F vs. SU_F and SP_U vs. SU_U) or feeding effect (SP_F vs. SP_U and SU_F vs. SU_U) across time. Treatments with SU bluefish were analyzed along with day 89 lipid data, since only SU bluefish were subsampled on this day. Due to insufficient numbers of unfed bluefish, final (day 192) data were not included in the analyses. Body condition indicators analyzed were the same as stated for day 0, with the addition of WWT (g)/FL (mm). A significance value of $\alpha=0.05$ was used. Significant main effects were analyzed post-hoc using Tukey multiple comparisons tests. Prior to ANOVA, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. Occasionally, data remained somewhat heteroscedastistic following transformation; for

these data, it was assumed that the ANOVA was robust enough to perform well despite the deviation from homoscedasticity (Zar 1984).

Energetic Condition: Day 0 vs. Day 192

Initial (day 0) and final (day 192) lipid data for the SP_F, SP_U and SU_F treatments were each analyzed using T-tests to test for a time effect within a treatment. These analyses were not performed on the SU_U treatment since no bluefish from this treatment survived to day 192. Body condition indicators analyzed were the same as stated for day 0. A significance value of $\alpha=0.05$ was used. Prior to T-tests, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using the MWU-test.

All statistical analyses were performed using the computer software Statistica 6.0.

Ash Content:

Both liver and WM ash content (g ash/g lean tissue*100) were analyzed similarly to the body condition indicators mentioned in both lipid energetics sections above.

Survival Analyses:

Survival curves were calculated for each treatment combination using the Kaplan-Meier product-limit estimate method (StatSoft 2001). Survival curves were compared

statistically using non-parametric (Mantel-Haenzel log-rank test and Gehan's Wilcoxon test) survival analyses appropriate for censored data (Marubini and Valsecchi 1995). Tank effects within each treatment were also tested using the above methods. The tank effect analysis for the SU_F treatment was performed using only two tanks, since the third tank did not experience a natural mortality throughout the experiment. After analyzing for tank effects, pooled data were analyzed for treatment effects. Pair-wise comparisons were performed using the two-sample log-rank test and/or Gehan's wilcoxon test to better identify any cohort and/or feeding effects when a significant main treatment effect was found. Survival analyses were conducted using the software program Statistica, version 6.0 (StatSoft 2001).

Sampling of Wild Bluefish:

Lipid data from the mesocosm experiment were compared to lipid data collected from wild bluefish (Morley 2004) to evaluate whether wild bluefish displayed low lipid values as seen in starved laboratory bluefish. Monthly collections of wild bluefish from the inner continental shelf of Onslow Bay, North Carolina, were obtained using a 40' bottom trawl aboard UNCW's 70' R/V Cape Fear (Morley 2004).

Laboratory/Wild Statistical Analyses:

Energetic Condition: Laboratory Bluefish

Day 0, 31, and 89 energetics data from unfed bluefish, both cohorts combined, were analyzed using one-way repeated-measure analyses-of-covariance (ANCOVAs) (similar-slopes or separate-slopes models) to investigate how energetics data changed

over time as bluefish starved. Unfed bluefish that expired after March 31, 2002, were determined to be starvation deaths, and included as a single time-point in the above analyses. Body condition indicators analyzed for each of these four time-points were: ln liver DWT (g), ln (liver lipid content (g) +1), ln (WM lipid density (%) +10), ln liver ash content, and ln WM ash content. A significance value of α=0.05 was used. Significant main effects were analyzed post-hoc using Tukey multiple comparisons tests. Prior to ANCOVA, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data was either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. Occasionally, data remained somewhat heteroscedastistic following transformation; for these data, it was assumed that the ANCOVA was robust enough to perform well despite the deviation from homoscedasticity (Zar 1984).

Energetic Condition: Laboratory vs. Wild Bluefish

Energetics data from wild bluefish collected in February 2002 and May 2002 were analyzed in the above manner, along with the laboratory starvation values, to determine if wild bluefish approach critical energetic values as determined from laboratory starved bluefish. Body condition indicators analyzed for each of these three time-points were the same as previously mentioned. A significance value of α=0.05 was used. Significant main effects were analyzed post-hoc using Tukey multiple comparisons tests. Prior to ANCOVA, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data was either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. Occasionally, data

remained somewhat heteroscedastistic following transformation; for these data, it was assumed that the ANCOVA was robust enough to perform well despite the deviation from homoscedasticity (Zar 1984).

RESULTS

Lipid Energetics:

Initial Condition:

Two-way ANOVA revealed significant differences in initial lipid content (g) and lipid density (%) among body depots and between cohorts in YOY bluefish (Table 1). For SP cohort bluefish, mean lipid content was highest in WM, followed by skin, RM, viscera and liver (Figure 4a). Tukey multiple comparisons tests indicated the following significant differences: WM>RM, viscera, liver; skin>viscera=liver; RM>liver. SU cohort bluefish displayed a similar distribution of lipid among body depots with the exception that liver values were higher than viscera values (Figure 4a). Tukey multiple comparisons tests indicated the following significant differences: WM>RM=liver=viscera, skin>liver=viscera. Comparisons among cohorts revealed that SP bluefish had significantly higher lipid content in all body depots except liver (Figure 4a). There was also a significant cohort*body depot interaction, indicating that the allocation of lipids to specific body depots varied by cohort (Table 1; Figure 4a). Specifically, SU bluefish had a higher percentage of lipids in liver than SP bluefish.

For SP bluefish, mean lipid density was highest in skin, followed by RM, viscera, liver and WM (Figure 4b). Tukey multiple comparisons tests indicated the following significant differences: skin>RM>viscera=liver=WM. SU bluefish showed highest levels

in skin, followed by liver, RM, viscera and WM (Figure 4b). Tukey multiple comparisons tests indicated the following significant differences: skin>RM, viscera, WM; liver>viscera=WM; RM>WM. Comparisons among cohorts revealed that SP bluefish had significantly greater mean lipid density in every body depot except liver (Figure 4b). There was a significant cohort*body depot interaction indicating that the concentration of lipids in specific body depots varied between cohorts (Table 1; Figure 4b). Specifically, SU bluefish had a higher concentration of lipids in liver than SP bluefish.

For SP cohort bluefish, lipid content was significantly correlated among all body depots except for between liver and viscera (Table 2a; Figure 5). Lipid content of SU bluefish was also correlated among body depots except for between liver and every other depot (Table 2b; Figure 6). Lipid densities in SP cohort depots were significantly correlated with each other except for between liver and viscera, liver and WM, and liver and RM (Table 2c; Figure 7). Lipid densities in SU cohort depots were significantly correlated with each other except for between liver and every other depot (Table 2d; Figure 8).

Energetic Condition of Body Depots: Day 0-31(89)

Fed Treatments: Spring and Summer Cohort

In the presence of food, SP bluefish exhibited a significant increase in overall lipid content during the first 31 days of the experiment (Table 3). While mean lipid content increased within each individual body depot, except skin, these individual increases were not statistically significant (Figure 9a-e). The lack of a significant

depot*time interaction indicated that bluefish increased the lipid content of all body depots similarly over time (Table 3; Figure 9a-e).

Lipid densities in fed SP bluefish depots did not increase significantly during the first 31 days of the experiment (Table 3). The lack of a significant depot*time interaction indicated that the lipid density of all body depots responded similarly over time (Table 3; Figure 10a-e).

Fed SU bluefish exhibited a significant increase in overall lipid content during the first 89 days of the experiment (Table 4). Specifically, mean lipid content increased in each body depot and these increases were statistically significant for liver, viscera, and WM (Figure 9a-e). The lack of a significant depot*time interaction indicated that SU bluefish increased the lipid content of all body depots similarly over time (Table 4; Figure 9a-e).

Overall, lipid density in fed SU bluefish increased significantly during the first 89 days of the experiment (Table 4). While mean lipid density increased within each individual body depot, skin was the only depot where the increase was significant (Figure 10a-e). The lack of a significant depot*time interaction indicated the bluefish increased the lipid density of all body depots similarly over time (Table 4; Figure 10a-e).

Unfed Treatments: Spring and Summer Cohort

In the absence of food, SP bluefish exhibited a significant decrease in overall lipid content during the first 31 days of the experiment (Table 5). While mean lipid content decreased within each individual body depot over time, viscera was the only depot where the decrease was significant (Figure 9a-e). The lack of a significant depot*time

interaction indicated that bluefish decreased the lipid content of all body depots similarly over time (Table 5; Figure 9a-e).

Overall lipid density in unfed SP bluefish decreased significantly during the first 31 days of the experiment (Table 5). While mean lipid density decreased within each individual body depot, except liver; these individual decreases were not statistically significant (Figure 10a-e). The significant depot*time interaction indicated that the lipid density of different body depots responded differently over time (Table 5; Figure 10a-e). Specifically, liver lipid density increased over time, while densities in all other depots decreased.

SU bluefish exhibited a significant decrease in overall lipid content during the first 89 days of the experiment when held without food (Table 6). While mean lipid content decreased within each individual body depot, these individual decreases were not statistically significant (Figure 9a-e). The lack of a significant depot*time interaction indicated the bluefish decreased the lipid content of all body depots similarly over time (Table 6; Figure 9a-e).

Overall lipid density in unfed SU bluefish decreased significantly during the first 89 days of the experiment (Table 6). While mean lipid density decreased within each individual body depot, these individual decreases were not statistically significant (Figure 10a-e). The lack of a significant depot*time interaction indicated the bluefish decreased the lipid density of all body depots similarly over time (Table 6; Figure 10a-e).

Effects of Feeding and Cohort on Energetics:

On day 0, SP bluefish had significantly larger values for liver DWT/FL, WM lipid density, and mean FL (Table 7). SP bluefish also had 50% more lipid in their liver than SU bluefish ($p=0.083$) (Table 7). Neither liver lipid density (%) nor liver lipid density (g lipid/mm FL) differed among cohorts (Table 7).

After 11 days, SP (fed and unfed) bluefish again displayed significantly higher condition than SU bluefish for liver lipid content, liver lipid density (g lipid/mm FL), liver DWT/FL, WM lipid density, and mean FL (Table 8; Figure 11a-e). The difference between cohorts was most apparent, significantly, among unfed bluefish (Figure 11a-e). Liver lipid density (%) was the only index that did not differ significantly between cohorts (Table 8). For each cohort, fed individuals displayed significantly larger values than their unfed counterparts in terms of liver lipid content, liver lipid density (%), liver lipid density (g lipid/mm FL) and liver DWT/FL (Table 8). The difference between fed and unfed bluefish was most significant in SU individuals (Figure 11a-e). WM lipid density and mean FL did not differ between feeding treatments (Table 8). No significant cohort*feeding interaction effect was found in any body condition index explored, indicating that SP and SU bluefish reacted similarly to their different feeding treatments (Table 8; Figure 11a-e).

After 31 days, SP (fed and unfed) bluefish displayed significantly higher condition than SU bluefish for liver lipid content, liver DWT/FL, and mean FL (Table 9). Again, the difference between SP and SU bluefish was most apparent in unfed individuals (Figure 11a-e). For each cohort, fed individuals displayed significantly higher condition than unfed individuals for liver lipid content, liver lipid density (%),

liver lipid density (g lipid/mm FL), liver DWT/FL and WM lipid density (Table 9). This feeding effect was equally visible in both cohorts for most condition factors; however, the difference between fed and unfed individuals for WM lipid density was only significant in SU bluefish (Figure 11a-e). The significant cohort*feeding interactions for both liver DWT/FL and WM lipid density indicated that how these condition indices responded to their feeding treatments depended on whether they were from SP or SU bluefish (Table 9; Figure 11a-e). A greater feeding effect was seen in SU bluefish, specifically unfed, than in SP bluefish.

After 89 days, fed SU bluefish, again, had significantly higher levels for liver lipid content, liver lipid density (g lipid/MM FL), liver DWT/FL, and WM lipid density than unfed SU bluefish (Table 10; Figure 11a-e). Liver lipid density (%) and mean FL were not affected by feeding treatment (Table 10).

After 192 days, SP_F bluefish only had significantly higher values than SU_F bluefish for liver DWT/FL and mean FL (Table 11; Figure 11a-e). Liver lipid content, liver lipid density (%), liver lipid density (g lipid/mm FL), and WM lipid density no longer differed between cohorts (Table 11).

Energetic Condition: Day 0-31(89)

Fed Treatments: Spring vs. Summer Cohort

In the presence of food, both SP and SU bluefish exhibited a significant increase in liver lipid content during the first 31 days of the experiment (Table 12; Figure 11a). The lack of a significant cohort*time interaction indicated that SP and SU bluefish increased their liver lipid content similarly over time (Table 12; Figure 11a).

Liver lipid density (%) in both SP and SU bluefish increased over the first 31 days of the experiment, although the increase was not statistically significant ($p=0.083$) (Table 12; Figure 11b). The lack of a significant cohort*time interaction indicated that SP and SU bluefish increased their liver lipid densities similarly over time (Table 12; Figure 11b).

Liver lipid density (g lipid/mm FL) in both SP and SU bluefish significantly increased during the first 31 days of the experiment (Table 12; Figure 11c). The lack of a significant cohort*time interaction indicated that SP and SU bluefish increased their liver lipid densities similarly over time (Table 12; Figure 11c).

Liver DWT/FL of both SP and SU bluefish significantly increased during the first 31 days of the experiment (Table 12; Figure 11d). The lack of a significant cohort*time interaction indicated that SP and SU bluefish increased their liver DWT/FL similarly over time (Table 12; Figure 11d).

WM lipid density (%) in both SP and SU bluefish increased during the first 31 days of the experiment (Table 12; Figure 11e); however, the increase was only significant in SU bluefish. The near significant cohort*time interaction ($p=0.071$) indicated that the increase of WM lipid density in YOY bluefish was partially dependent on cohort of origin (Table 12; Figure 11e).

WWT/FL of both SP and SU bluefish did not significantly increase during the first 31 days of the experiment ($p=0.072$) (Table 12). The lack of a cohort*time interaction indicated that SP and SU bluefish increased their WWT/FL similarly over time (Table 12).

Mean FL of both SP and SU bluefish did not significantly increase during the first 31 days of the experiment (Table 12). The lack of a cohort*time interaction indicated that SP and SU bluefish increased their mean FL similarly over time (Table 12).

Unfed Treatments: Spring vs. Summer Cohort

In the absence of food, both SP and SU bluefish exhibited a near significant decrease in liver lipid content during the first 31 days of the experiment ($p=0.068$) (Table 13; Figure 11a). The lack of a significant cohort*time interaction indicated that SP and SU bluefish decreased their liver lipid content similarly over time (Table 13; Figure 11a).

Mean liver lipid density (%) in both SP and SU bluefish increased during the first 31 days of the experiment ($p=0.099$) (Table 13; Figure 11b). The lack of a significant cohort*time interaction indicated that SP and SU bluefish increased their liver lipid densities similarly over time (Table 13; Figure 11b).

Mean liver lipid density (g lipid/mm FL) in SP and SU bluefish decreased significantly during the first 31 days of the experiment ($p=0.030$), but the decrease was not significant in either individual cohort (Table 13; Figure 11c). The lack of a significant cohort*time interaction indicated that SP and SU bluefish increased their liver lipid densities similarly over time (Table 13; Figure 11c).

Liver DWT/FL in both SP and SU bluefish decreased significantly during the first 31 days of the experiment (Table 13; Figure 11d). The lack of a significant cohort*time interaction indicated that SP and SU bluefish decreased their liver DWT/FL similarly over time (Table 13; Figure 11d).

WM lipid density (%) in both SP and SU bluefish did not significantly change during the first 31 days of the experiment (Table 13; Figure 11e). The near significant cohort*time interaction (p=0.071) indicated that the change in WM lipid density of YOY bluefish was partially dependent on cohort of origin (Table 13; Figure 11e). Specifically, mean WM lipid density values decreased in SP bluefish, while they increased in SU bluefish.

WWT/FL in both the SP and SU bluefish combined decreased significantly during the first 31 days of the experiment (p=0.046) (Table 13); however, the decrease was not significant in either individual cohort. The lack of a significant cohort*time interaction indicated that SP and SU bluefish decreased their liver lipid densities similarly over time (Table 13).

Both SP and SU bluefish exhibited a marginally significant increase in mean FL during the first 31 days of the experiment (p=0.052) (Table 13). The near significant cohort*time interaction (p=0.070) indicated that the change in mean FL of YOY bluefish was partially dependent on cohort of origin (Table 13). Specifically, mean FL in SP bluefish increased, while in SU bluefish it remained stable.

Spring Cohort: Fed vs. Unfed Treatment

SP bluefish, regardless of feeding treatment, exhibited a significant increase in mean liver lipid content during the first 31 days of the experiment (Table 14; Figure 11a). The significant feeding*time interaction indicated that how liver lipid content changed was dependent on the presence of food (Table 14; Figure 11a). Specifically, mean liver lipid content increased in fed bluefish and decreased in unfed bluefish (Figure 11a).

Mean liver lipid density (%), regardless of feeding treatment, exhibited a near significant increase during the first 31 days of the experiment ($p=0.075$) (Table 14; Figure 11b). The lack of a significant feeding*time interaction indicated that fed and unfed SP bluefish increased their liver lipid densities similarly over time (Table 14; Figure 11b).

Mean liver lipid density (g lipid/mm FL), regardless of feeding treatment, exhibited a near significant increase during the first 31 days of the experiment ($p=0.093$) (Table 14; Figure 11c). The significant feeding*time interaction indicated that how liver lipid density changed was dependent on the presence of food (Table 14; Figure 11c). Specifically, mean liver lipid density decreased in unfed bluefish, while it increased significantly in fed bluefish (Figure 11c).

Mean liver DWT/FL, regardless of feeding treatment, did not significantly change during the first 31 days of the experiment (Table 14; Figure 11d). The significant feeding*time interaction indicated that how body condition changed was dependent on the presence of food (Table 14; Figure 11d). Specifically, mean liver DWT/FL increased significantly in fed bluefish and decreased significantly in unfed bluefish (Figure 11d).

Mean WM lipid density (%), regardless of feeding treatment, did not significantly change during the first 31 days of the experiment (Table 14; Figure 11e). The lack of a significant feeding*time interaction indicated that mean WM lipid density in fed and unfed SP YOY bluefish responded similarly over time (Table 14; Figure 11e).

Mean WWT/FL, regardless of feeding treatment, did not significantly change during the first 31 days of the experiment (Table 14). Although mean WWT/FL increased in fed bluefish and decreased in unfed bluefish, neither trend was significant.

The lack of a significant feeding*time interaction indicated that mean WWT/FL in fed and unfed SP bluefish responded similarly over time (Table 14).

Mean FL, regardless of feeding treatment, increased significantly during the first 31 days of the experiment (Table 14). Individually, only the increase in unfed bluefish was significant. The lack of a significant feeding*time interaction indicated that mean FL in fed and unfed SP bluefish increased similarly over time (Table 14).

Summer Cohort: Fed vs. Unfed Treatment

SU bluefish, regardless of feeding treatment, exhibited a near significant increase in mean liver lipid content during the first 89 days of the experiment ($p=0.056$) (Table 15; Figure 11a). The significant feeding*time interaction indicated that how liver lipid content changed over time was dependent on the presence of food (Table 15; Figure 11a). Specifically, mean liver lipid content increased significantly in fed bluefish and decreased in unfed bluefish (Figure 11a).

Mean liver lipid density (%), regardless of feeding treatment, increased significantly during the first 89 days of the experiment (Table 15; Figure 11b); however, the increase was not significant in either individual treatment. The lack of a significant feeding*time interaction indicated that fed and unfed SU bluefish increased their mean liver lipid content similarly over time (Table 15; Figure 11b).

Mean liver lipid density (g lipid/mm FL), regardless of feeding treatment, increased significantly during the first 89 days of the experiment (Table 15; Figure 11c). The significant feeding*time interaction indicated that how mean liver lipid density changed over time depended on feeding treatment (Table 15; Figure 11c). Specifically,

mean liver lipid density increased significantly in fed bluefish, but it decreased in unfed bluefish (Figure 11c).

Mean liver DWT/FL, in both fed and unfed bluefish, changed significantly during the first 89 days of the experiment (Table 15; Figure 11d). Mean liver DWT/FL decreased significantly in unfed bluefish, while in fed bluefish it increased significantly over the first 31 days, followed by a significant decrease from day 31 to day 89 (Figure 11d). The significant feeding*time interaction indicated that how mean liver DWT/FL changed over time depended on feeding treatment (Table 15; Figure 11d).

Mean WM lipid density (%), regardless of feeding treatment, increased significantly during the first 89 days of the experiment (Table 15; Figure 11e). The significant feeding*time interaction indicated that mean WM lipid density changed over time depended on feeding treatment (Table 15; Figure 11e). Specifically, mean WM lipid density decreased in unfed bluefish, while it increased significantly in fed bluefish (Figure 11e).

Mean WWT/FL, regardless of feeding treatment, significantly decreased during the first 89 days of the experiment (Table 15). The significant feeding*time interaction indicated that how mean WWT/FL changed over time depended on feeding treatment (Table 15). Specifically, mean WWT/FL significantly decreased in unfed bluefish, while it increased in fed bluefish.

Mean FL, regardless of feeding treatment, did not significantly increase during the first 89 days of the experiment (Table 15). The lack of a significant feeding*time interaction indicated that mean FL in fed and unfed bluefish increased similarly over time (Table 15).

Energetic Condition: Day 0 vs. Day 192

Spring Fed Treatment:

In the presence of food, SP bluefish had significantly lower condition values at the end of the experiment (day 192) than at the beginning for liver lipid content, liver lipid density (%), liver lipid density (g lipid/mm FL), liver DWT/FL and WM lipid density (Table 16; Figure 11a-e). Mean FL (±S.D.) of SP_F bluefish increased significantly from day 0 (252 mm) to day 192 (275 mm) (Table 16).

Summer Fed Treatment:

In the presence of food, SU bluefish also had significantly lower condition values at the end of the experiment than at the beginning for liver lipid content, liver lipid density (%), liver lipid density (g lipid/mm FL), liver DWT/FL and WM lipid density (Table 17; Figure 11a-e). Mean FL (±S.D.) of SU_F bluefish increased significantly from day 0 (206 mm) to day 192 (238 mm) (Table 17).

Spring Unfed Treatment:

In the absence of food, SP bluefish had significantly lower condition values at the end of the experiment than at the beginning for liver lipid content, liver lipid density (%), liver lipid density (g lipid/mm FL), liver DWT/FL and WM lipid density (Table 18; Figure 11a-e). Mean FL (±S.D.) of SP_U bluefish on day 0 (247 mm) and day 192 (256 mm) were not significantly different (Table 18).

Ash Content:

Initial Condition:

Kruskal-Wallis two-way ANOVA by ranks revealed significant differences in initial ash content among body depots, but not between cohorts in YOY bluefish (Table 19). For SP cohort bluefish, mean ash content was lowest in liver, followed by skin, viscera, RM and WM (Figure 12). Multiple comparison tests of mean rank indicated the following significant differences: liver<WM (Figure 12). For SU cohort bluefish, mean ash content was lowest in RM, followed by liver, WM, viscera and skin (Figure 12). Multiple comparisons of mean rank indicated the following significant differences: RM<viscera=skin; liver<viscera (Figure 12). Comparisons among cohorts revealed that SP bluefish did not have significantly lower ash content in any body depot, except skin (Figure 12).

Ash content in SP cohort bluefish tissues was not correlated between depots (Table 20a; Figure 13). For SU bluefish, the only significant correlation was between liver and viscera (Table 20b; Figure 14).

Energetic Condition of Body Depots: Day 0-31(89)

Fed Treatments: Spring and Summer Cohort

In the presence of food, SP bluefish exhibited a significant decrease in overall ash content during the first 31 days of the experiment (Table 21). While mean ash content decreased within each individual body depot, except viscera, liver was the only depot where the decrease was significant (Figure 15a-e). The significant depot*time interaction indicated that the ash content of bluefish body depots responded differently over time

(Table 21; Figure 15a-e). Specifically, viscera ash content increased over time, while the ash content of all other depots decreased.

Within the SU cohort, overall ash content did not significantly change during the first 89 days of the experiment when food was present (Table 21). The significant depot*time interaction indicated that the ash content of bluefish body depots responded differently over time (Table 21; Figure 15a-e). Specifically, mean ash content in viscera and RM decreased over time, while mean ash content in liver, WM and skin increased.

Unfed Treatments: Spring and Summer Cohort

In the absence of food, overall ash content in SP bluefish did not significantly change during the first 31 days of the experiment (Table 21). The significant depot*time interaction indicated that the ash content of bluefish body depots responded differently over time (Table 21; Figure 15a-e). Specifically, liver, viscera and RM mean ash content increased over time, while mean ash content in WM and skin decreased.

Within the SU cohort, overall ash content did not significantly change during the first 89 days of the experiment when food was not present (Table 21). The significant depot*time interaction indicated that the ash content of bluefish body depots responded differently over time (Table 21; Figure 15a-e). Specifically, liver, viscera, WM and RM mean ash content increased over time, while mean ash content of skin decreased significantly.

Effects of Feeding and Cohort on Energetics:

On day 0, both liver and WM ash content did not differ significantly between cohorts (Table 22).

After 11 days, neither liver nor WM ash content differed significantly between cohorts (Table 23). In each cohort, liver ash content was significantly lower in fed individuals than unfed individuals (Figure 16a). WM ash content had a near significant feeding effect ($p=0.066$); however, the difference between fed and unfed individuals was only apparent in SU bluefish (SU_F<SU_U) (Table 23; Figure 16b). The near significant cohort*feeding interaction in WM ash content indicated that how it responded to a different feeding treatment depended on whether it was from a SP or SU bluefish. ($p=0.057$) (Table 23; Figure 16b).

After 31 days, SP (fed and unfed) bluefish had significantly lower WM ash content than SU bluefish (Table 24). Specifically, unfed SP bluefish had significantly less WM ash content than unfed SU bluefish (Figure 16b). In each cohort, fed individuals had significantly less ash content, in both liver and WM, than unfed individuals (Table 24; Figure 16a-b). Again, the difference in WM ash content between fed and unfed individuals was only significant, individually, in SU bluefish. The near significant cohort*feeding interaction in WM ash content indicated that how it responded to a different feeding treatment depended on whether it was from a SP or SU bluefish. ($p=0.058$) (Table 24; Figure 16b).

After 89 days, fed SU bluefish had significantly lower ash content, in both liver and WM, than unfed SU bluefish (Table 25; Figure 16a-b).

After 192 days, ash content, liver and WM, in SP_F bluefish did not differ significantly from SU_F bluefish (Table 26; Figure 16a-b).

Energetic Condition: Day 0-31(89):

Fed Treatments: Spring vs. Summer Cohort

In the presence of food, both SP and SU bluefish exhibited a significant decrease in liver ash content during the first 31 days of the experiment (Table 27; Figure 16a). The lack of a significant cohort*time interaction indicated that SP and SU bluefish decreased their liver ash content similarly over time (Table 27; Figure 16a).

WM ash content in both SP and SU bluefish combined decreased significantly during the first 31 days of the experiment when food was present (Table 27; Figure 16b). While mean WM ash content values decreased in both cohorts, only in the SP cohort was the individual decrease significant. The lack of a significant cohort*time interaction indicated that SP and SU bluefish decreased their WM ash content similarly over time (Table 27; Figure 16b).

Unfed Treatments: Spring vs. Summer Cohort

In the absence of food, both SP and SU bluefish exhibited a significant increase in liver ash content during the first 31 days of the experiment (Table 28; Figure 16a). The lack of a significant cohort*time interaction indicated that SP and SU bluefish increased their WM ash content similarly over time (Table 28; Figure 16a).

WM ash content in both SP and SU bluefish did not significantly change during the first 31 days of the experiment when food was not present (Table 28; Figure 16b).

The significant depot*time interaction indicated that WM ash content in YOY bluefish responded differently over time depending on cohort of origin (Table 28; Figure 16b). Specifically, in SP bluefish mean WM ash content decreased, while in SU bluefish it increased.

Spring Cohort: Fed vs. Unfed Treatment

SP bluefish, regardless of feeding treatment, exhibited a significant change in mean liver ash content during the first 31 days of the experiment (Table 29; Figure 16a). Specifically, mean liver ash content increased significantly in unfed bluefish and decreased significantly in fed bluefish. The significant feeding*time interaction indicated that how mean liver ash content changed over time depended on feeding treatment (Table 29; Figure 16a).

Mean WM ash content, regardless of feeding treatment, significantly decreased during the first 31 days of the experiment (Table 29; Figure 16b), despite not significantly decreasing in either individual feeding treatment. The lack of a significant feeding*time interaction indicated that fed and unfed bluefish decreased mean WM ash content similarly over time (Table 29; Figure 16b).

Summer Cohort: Fed vs. Unfed Treatment

SU bluefish, regardless of feeding treatment, exhibited a significant change in mean liver ash content during the first 89 days of the experiment (Table 30; Figure 16a). Specifically, mean liver ash content increased significantly in unfed bluefish, while it decreased significantly in fed bluefish through day 31 and then increased significantly

from day 31 to day 89 (Table 30; Figure 16a). The significant feeding*time interaction indicated that how mean liver ash content changed over time depended on feeding treatment (Table 30; Figure 16a).

Mean WM ash content, regardless of treatment, did not significantly change during the first 89 days of the experiment (Table 30; Figure 16b). The significant feeding*time interaction indicated that how mean WM ash content changed depended on feeding treatment (Table 30; Figure 16b). Specifically, mean WM ash content decreased in fed bluefish, while it increased significantly in unfed bluefish (Figure 16b).

Energetic Condition: Day 0 vs. Day 192

Spring Fed Treatment:

In the presence of food, liver and WM ash content in SP bluefish did not differ significantly between the start and finish of the experiment (Table 31; Figure 16a-b).

Summer Fed Treatment:

In the presence of food, WM ash content in SU bluefish increased significantly over the course of the experiment; however, final bluefish still had very low WM ash content levels (Table 32; Figure 16b). Liver ash content in SU bluefish did not change significantly between the start and finish of the experiment (Table 32; Figure 16a).

Spring Unfed Treatment:

In the absence of food, the ash content in both liver and WM of SP bluefish did not differ significantly between the start and finish of the experiment (Table 33; Figure

16a-b). However, due to only two SP_U bluefish surviving to the end of the experiment, an accurate account of this treatment is not available.

Survival Analyses:

Mantel-Haenzel log-rank tests revealed that survival curves within a given treatment (SP_F, SP_U, SU_F, SU_U) did not differ significantly among replicate tanks (Figure 17a-d). Given the lack of tank effects, a log-rank test performed on pooled data from each treatment indicated survival durations of YOY bluefish differed significantly across treatments (Figure 18).

Within each cohort, fed individuals survived significantly longer than unfed individuals (log-rank: $p=0.00804$ for SP and $p=0.00831$ for SU) (Figure 18). When both cohorts were held without food during winter, SP bluefish survived significantly longer than SU bluefish (Gehan's wilcoxon: $p=0.00057$) (Figure 18). Under starvation conditions, 50% of SU bluefish survived ~158 days (Figure 18). In the presence of food, survival durations did not differ significantly between cohorts (log-rank, $p=0.55260$) (Figure 18).

Laboratory/Wild Analyses:

Energetic Condition: Laboratory Bluefish

A separate-slopes model ANCOVA revealed that size-adjusted liver dry mass (g liver DWT) decreased significantly over time in unfed YOY bluefish (November>December>February>Starvation values) ($p=0.004$) (Figure 19). Levene's

test showed a moderate heterogeneity of variance (p=0.046), however ANCOVA was assumed to be robust enough to handle the slight heterogeneity (Zar 1984).

Each of the remaining condition indices (liver lipid content, WM lipid density, liver ash content and WM ash content) declined significantly over time; however, all failed Levene's test (Figures 20-23).

Energetic Condition: Laboratory vs. Wild Bluefish

A separate-slopes ANCOVA results indicated that size-adjusted mean liver dry mass of critically starved laboratory bluefish were significantly lower than values measured in wild bluefish during February and May (p=0.000) (Figure 19). Levene's test indicated that all variances were homogeneous. For liver ash content, ANCOVA indicated that wild bluefish values from both February and May were significantly lower than critically starved laboratory bluefish (p=0.000) (Figure 22). Levene's test indicated that all variances were homogeneous.

Analyses for liver lipid content, WM lipid density and WM ash content failed Levene's test; however, graphical inspection of the data suggests that wild bluefish values never approached those of critically starved laboratory bluefish (Figures 20, 21, 23).

DISCUSSION

Overwinter Mortality and Bluefish Recruitment:

Bluefish populations off of the East Coast of the United States have declined in recent years (Munch and Conover 2000). While both SP- and SU-spawned bluefish have been shown to recruit to the adult population in the past (Lassiter 1962), recent studies

have suggested that very few SU-spawned bluefish currently recruit to the adult stock (Chiarella and Conover 1990, Gilmore 2000). The present study examined whether winter starvation might account for the disappearance of this cohort.

Winter is a potentially stressful period in the life of young fishes when survival and recruitment success may be influenced greatly (Johnson and Evans 1996, Hurst and Conover 1998; see also Hurst and Conover 2001). Low winter water temperatures are often associated with reduced prey availability and/or decreased ability to digest/assimilate food (Cunjak *et al*. 1987, Cunjak and Power 1987, Johnson and Evans 1996, Hurst and Conover 1998; see also Hurst and Conover 2001). Winter declines in prey abundance may make it difficult for wild bluefish to feed sufficiently in the winter to meet daily metabolic requirements (Morley 2004). When feeding is either ineffective or inadequate to meet daily energy requirements, fishes usually rely on stored energy (mainly lipids) to survive (Oliver *et al*. 1979, Cunjak *et al*. 1987, Cunjak 1988, Post and Evans 1989b, Thompson *et al*. 1991, Miranda and Hubbard 1994, Schultz and Conover 1997, Jobling 2001b). Extended periods of cold winter water temperatures, or chronic thermal stress, can lead to the exhaustion of energy reserves and starvation. Due to the allometries of both energy storage and energy depletion rate with body size, larger individuals usually have greater energy stores, a lower mass-specific metabolic rate and a greater overall starvation endurance (Oliver *et al.* 1979; Post and Evans 1989b; Thompson *et al.* 1991; Johnson and Evans 1996; Schultz and Conover 1997, 1999; Sogard 1997). In addition to limited food sources and chronic thermal stress, winter can also cause acute thermal stress in fishes. Sharp declines in water temperature during the winter, along with extreme cold temperatures, are examples of acute thermal stress and

can cause osmoregulatory failure in overwintering fishes (Johnson and Evans 1996, Hurst and Conover 1998, Lankford and Targett 2001). Lankford and Targett (2001) found that larger YOY Atlantic croaker (*Micropogonias undulatus*) were more vulnerable to acute thermal stress than smaller conspecifics (see also Otwell and Merriner 1975, Shafland and Pestrak 1982, Prentice 1989).

A growing theory concerning the winter energetics of fishes is that larger individuals are better suited to endure prolonged cold than smaller conspecifics (Oliver *et al.* 1979, Toneys and Coble 1979, Thompson *et al*. 1991, Miranda and Hubbard 1994, Sogard 1997, Hurst and Conover 1998, Post and Evans 1989b, Johnson and Evans 1990, Thompson *et al*. 1991). Larger individuals have been shown to have greater energy storage capacity and lower weight specific metabolic rates than smaller individuals (Paloheimo and Dickie 1966, Shul'man 1974, Oliver *et al.* 1979, Post and Evans 1989b, Schultz and Conover 1999). Larger individuals are therefore able to store more energy prior to winter than smaller individuals and tend to deplete this energy at a slower rate. Negative size-selective overwinter mortality has been observed in striped bass, *Morone saxatilis* (Hurst and Conover 1998); largemouth bass, *Micropterus salmoides* (Miranda and Hubbard 1994); smallmouth bass, *Micropterus dolomieui* (Oliver *et al.* 1979); yellow perch, *Perca flavescens* (Post and Evans 1989b); white perch, *Morone americana* (Johnson and Evans 1990); Colorado pikeminnow, *Ptychocheilus lucius* (Thompson *et al*. 1991); Atlantic silversides, *Menidia menidia* (Schultz and Conover 1997); sand smelt, *Atherina boyeri* (Henderson *et al*. 1988); and rainbow trout, *Oncorhyncus mykiss* (Smith and Griffith 1994); because of these allometries between body-size and energy storage/depletion.

Pre-winter Energy Reserves: Spring vs. Summer Cohort

An important prediction of this hypothesis is that SU bluefish enter winter with lower energy reserves than SP bluefish. Previous researchers have already shown that larger YOY fishes, including Colorado pikeminnow (Thompson *et al*. 1991), largemouth bass (Miranda and Hubbard 1994), and Atlantic siliversides (Schultz and Conover 1997) enter winter with greater energy stores than smaller conspecifics. The present findings for bluefish were generally in agreement with these studies. SP bluefish did enter winter with significantly higher lipid content (>4.75times) and significantly higher lipid densities (>13%) than SU bluefish in four out of five body depots examined, including viscera, WM, RM and skin. Interestingly, the livers of SP- and SU-spawned bluefish did not differ significantly in lipid content or lipid density, although SP liver lipid content was, on average, 1.5 times greater than that of the SU cohort. Liver body condition in SP bluefish was significantly greater than in SU bluefish suggesting that larger bluefish had larger livers. Despite differences in overall lipid values between cohorts, ash values were not consistently different indicating that the organic content of each body depot was similar among cohorts going into winter.

Fishes are known to store and metabolize lipids from multiple body depots. The relative importance of different depots differs depending on species (reviewed in Love 1980, Sheridan 1988). When analyzing lipid content of fishes, most previous studies have focused on selected depots while ignoring others (Sheridan 1988). In this study, the lipid content of liver, viscera, WM, RM, and skin were all analyzed individually. This approach revealed that both SP and SU bluefish relied on all depots for storage and mobilization of lipids. Furthermore, each cohort displayed significant correlations

between viscera, WM, RM, and skin values for both lipid content and lipid density (%). Thus, with the exception of liver, it appears that overall lipid content of bluefish can be indexed by analyzing a particular body depot. Ash content was not as strongly correlated between body depots in either cohort.

Winter Energy Depletion Rate: Spring vs. Summer Cohort

Another important prediction of the starvation hypothesis was that SU bluefish would deplete their lipid reserves at a faster rate than SP bluefish when food was witheld. Smaller YOY of other species, including yellow perch (Post and Evans 1989b), Atlantic silversides (Schultz and Conover 1999), and largemouth bass (Miranda and Hubbard 1994) have previously been shown to deplete reserves faster than larger conspecifics (see also Paloheimo and Dickie 1966). Experimental data from this study did not generally support this prediction. When starved, the smaller, SU bluefish did not deplete their lipid reserves at a significantly different rate than the larger, SP bluefish for any of the six condition indices analyzed over the first 31 days of the experiment. Also, liver ash content did not change at a significantly different rate between cohorts. Interestingly, WM ash content in SU bluefish increased at a faster rate than in SP bluefish, suggesting that SU individuals may have depleted non-lipid energy reserves (e.g. protein or glycogen) in WM at a faster rate than SP individuals. Although SP and SU bluefish depleted their lipid reserves at similar rates, SU individuals would be expected to have a lower starvation endurance since they enter winter with lower energy reserves than SP bluefish (Thompson *et al*. 1991).

Winter Energy Storage Rate: Spring vs. Summer Cohort

SU cohort individuals might compensate for their size disadvantage at the onset of winter if, when food was present, they were able to store energy more rapidly and efficiently than SP individuals. Previous studies by McBride *et al*. (1993) and Buckel *et al*. (1998) have suggested that bluefish do not exhibit compensatory growth. This study had similar findings with respect to energy storage. When food was present, both cohorts of bluefish stored lipid at similar rates over the first 31 days of the experiment. There were no significant differences in energy storage rates between cohorts for any of the six condition factors analyzed. There were also no significant differences in the rates of decrease for ash content in liver or WM over this time period, suggesting that both cohorts increased the organic content of their tissues at similar rates. These findings suggest that SU bluefish are not able to store lipids at faster rates than SP bluefish when food is present and are therefore unable to compensate for their lower energy reserves than SP individuals prior to winter.

Interestingly, when fed throughout the winter both cohorts experienced a significant loss of stored lipids. Both cohorts of bluefish in the fed treatment had considerably lower lipid reserves in May than in November and December even though these cohorts were provided unlimited prey throughout the winter. This seasonal depletion pattern has been reported for other species of fish (Reimers 1963, Hunt 1969, Post and Evans 1989b, Thompson *et al*. 1991) and is thought to reflect the importance of lipid reserves for fueling routine metabolic requirements during winter. There was not a significant change in the ash content of liver or WM for either cohort (except for WM ash content in SU bluefish), which suggests that despite lipid stores being depleted, muscle

organic content was maintained. While WM ash content in SU bluefish did increase significantly, mean values of liver and WM ash content were not suggestive that either cohort of bluefish had less organic material in its WM or liver following winter than before, meaning that the function of these depots was not compromised over time.

In the presence of food, both cohorts appeared to selectively deplete lipid reserves rather than to maintain or synthesize new lipids. Cunjak *et al.* (1987) has shown that at cold temperatures fish cannot digest and assimilate food efficiently and must rely on stored energy reserves to maintain daily energy requirements. While such temperatures likely occurred for bluefish subjects during January and February, it is surprising that lipid reserves remained virtually depleted in late May (day 192) when winter temperatures had not occurred for three months and food was unlimited. Water temperatures in the mesocosm tanks during March, April and May ranged from ~10^{o}C to ~30^{o}C, with temperatures above 15^{o}C the majority of the time. Such temperatures are within the preferred normal range of wild bluefish (Lund and Maltezos 1970, Munch 1997, Morley 2004). It is noteworthy that similar temperatures during the fall were associated with rapid lipid storage by both cohorts.

A second explanation for the depletion of lipid stores in the fed SP and SU bluefish involves the "defended energy level" hypothesis (Mrosovsky and Sherry 1980, Metcalfe and Thorpe 1992), and a seasonal energy allocation hypothesis. The defended energy level hypothesis states that fish will not allow their energy reserves to drop below a threshold level that is critical for survival. If their energy stores do drop below this level, fish will resume feeding until their depleted reserves have been restored (Metcalfe and Thorpe 1992). Thus, rates of feeding and energy storage would be inversely

proportional to energetic condition. The seasonal energy allocation hypothesis involves the trade-off in energy allocation between growth and storage (Shul'man 1974, Schultz and Conover 1997, Post and Parkinson 2000). This trade-off suggests that once the threat of winter starvation ceases and food is no longer limiting, any stored energy not utilized during the winter would be allocated towards growth and/or reproduction (Shul'man 1974). Laboratory analyses support these hypotheses. Since both cohorts depleted energy reserves at similar rates and SP bluefish started with significantly greater reserves, SP bluefish should still have greater energy reserves than SU bluefish following winter. This was not observed. While fed SP bluefish still had significantly larger livers than fed SU bluefish at the end of the experiment, they no longer exhibited significantly higher energy reserves. Liver and WM ash content were still not significantly different between cohorts suggesting that the organic content of various depots was similar. These findings are consistent with the "defended energy level" hypothesis in that if SU bluefish selectively fed throughout winter when their energy reserves approached a critical level, and SP bluefish, with their greater lipid reserves, chose not to feed; then the energy reserves of SU bluefish would be comparable to those of SP bluefish by the end of the experiment. Data from wild caught bluefish also support this argument (Morley 2004). Morley (2004) states that wild YOY bluefish likely defend their energy levels in the winter through strategic feeding.

The low energy reserves of laboratory SP and SU bluefish at the end of the experiment, despite warm water temperatures and abundant food, also support the seasonal energy allocation hypothesis. If YOY bluefish store energy in the fall in preparation for low winter temperatures and low prey abundance, and utilize any extra

stored energy after winter is over for growth/reproduction/migration, similar results would be seen. This strategy has been observed in many other species (Flath and Diana 1985, see Schultz and Conover 1997, see Jobling 2001b). While some species of fish are known to utilize energy reserves for gonadal development (Weatherley and Gill 1987, Love 1988, Jørgensen *et al*. 1997, Jobling 2001b), bluefish do not become reproductively mature until ~ age 2 (Deuel 1964). Thus, any stored energy mobilized by YOY bluefish after their first winter is likely associated with either somatic tissue growth or migration costs rather than reproduction. Analyses of wild bluefish also suggest that bluefish have a seasonal energy allocation strategy. Morley (2004) showed that, from April to May, wild bluefish simultaneously lose lipid reserves while increasing both their body weight and feeding activity.

Overwinter Survival: Spring vs. Summer Cohort

Two other important predictions of the starvation hypothesis were 1) that SU bluefish would survive winter better when food was present than when it was withheld, and 2) that when food was unavailable, SU bluefish would starve sooner than SP bluefish. Such patterns have been observed in overwintering white perch (Johnson and Evans 1990), yellow perch (Post and Evans 1989b) and Colorado pikeminnows (Thompson *et al*. 1991). Survival data for laboratory bluefish generally supported both of these predictions. YOY bluefish from both cohorts experienced significantly higher mean probable percent survival at the end of the experiment when fed than when unfed. In addition, when food was withheld from both cohorts, SU bluefish started to experience starvation mortalities approximately six weeks earlier than SP bluefish. These findings

are in accordance with the starvation hypothesis; however, due to the length of time that the SU bluefish were able to survive in the absence of food this hypothesis does not appear to be ecologically relevant. While SU bluefish did experience starvation at least six weeks before SP bluefish, these starvation mortalities were not observed until after March 31, 2002 (day 127), well after winter had ended and foraging conditions in the wild would have improved. Furthermore, unfed SU bluefish still had a > 60% survival probability by day 150, after approximately five months without food. Bluefish are not unique among fishes in their ability to endure long periods without food (Love 1980, Post and Evans 1989b, Johnson and Evans 1990, Johnson and Evans 1991, Thompson *et al*. 1991, Sogard and Olla 2000, see Jobling 2001b). Although SU bluefish were more susceptible to overwinter starvation mortality than SP bluefish, their ability to endure starvation appears more than sufficient to enable them to survive a typical winter period in the SAB. Based on these findings, the starvation hypothesis does not appear to be a suitable explanation for the apparent recruitment failure of SU-spawned YOY bluefish.

Starvation vs. Acute Thermal Stress

When analyzing the survival curves of the unfed SP- and SU-spawned bluefish, an interesting observation was made involving the different nature of both cohorts' survival curves over time. Despite unfed SU bluefish beginning to experience starvation mortalities before SP bluefish, they still had >90% survival probability over the first four months of the experiment. Unfed SP cohort bluefish, however, experienced a severe mortality event during January 2002. This mass mortality event coincided with a cold front which lowered tank temperatures below 6^{o}C. While these cold temperatures

severely affected the larger SP unfed bluefish, they did not affect the smaller SU unfed bluefish. This observation suggests that smaller, SU bluefish possess greater cold tolerance than larger, SP bluefish. Similar findings were reported for YOY Atlantic croaker by Lankford and Targett (2001). Different thermal tolerances might have important implications for the winter ecology of SP versus SU bluefish. For example, smaller SU bluefish might not have to migrate as early or as far south in order to avoid winter temperatures since they are apparently better suited than SP bluefish to handle colder temperatures. Given an increased ability to tolerate lower temperatures, it may benefit SU bluefish to overwinter at lower temperatures where reduced metabolic demands might compensate for the lower energy reserves of SU individuals. Thus, optimal winter temperatures may be lower for SU than SP bluefish. Johnson and Evans (1996) noted a possible trade-off between the risk of acute thermal stress at low water temperatures and an increased starvation risk at elevated temperatures in YOY white perch. Also, decreased migration might allow SU cohort bluefish to conserve more of their energy reserves, thus further increasing their likelihood of winter survival (see Hurst and Conover 2001). Additional studies of the effect of differing activity levels (i.e. migration costs) and energy depletion rates of SU YOY bluefish are necessary to further evaluate this hypothesis. Different thermal physiologies between cohorts might dictate when individuals of a specific cohort stop feeding for the winter. For example, smaller fishes might be better able to digest and assimilate prey items at lower temperatures than larger individuals due to their higher weight specific metabolic rate. The influence of body-size on temperature of feeding cessation in YOY bluefish is not known and further studies in this area are needed.

Overwinter Starvation: Laboratory vs. Wild

Another important aspect of the starvation hypothesis is whether wild YOY bluefish approach critical energy levels as determined from starved laboratory bluefish. Energetic analyses suggest that wild bluefish did not approach critical energy levels. Wild bluefish displayed values significantly higher than critically starved laboratory bluefish for all three body condition indices analyzed. In May, critically starved laboratory bluefish had livers almost half the size of similar-sized wild bluefish. Liver and WM ash content of wild bluefish sampled at the end of winter were also significantly less than those of starved laboratory bluefish, suggesting that the liver and WM of wild bluefish were in better energetic condition. These findings are consistent with those of the laboratory survival analyses and do not support the hypothesis that recruitment failure of SU-spawned bluefish is due to starvation. However, it is important to point out that the laboratory study only assessed the physiological capabilities of bluefish to endure starvation and did not evaluate possible indirect effects of starvation on survival potential. For example, previous researchers have suggested chronic starvation may weaken individuals, thus increasing their susceptibility to predation (Furuta 1998, Skajaa *et al*. 2003). This might explain why energy values of wild bluefish would not approach those of critically starved laboratory bluefish even if they were experiencing indirect starvation mortality. However, when analyzing wild bluefish, Morley (2004) found that YOY bluefish likely feed during winter although feeding is reduced due to low temperature. He also found evidence that SU-spawned bluefish do survive winter (Morley 2004).

In summary, both energy and survival data suggest that YOY bluefish do suffer from negative size-selective overwinter mortality when food is withheld. However, the length of time that SU-spawned YOY bluefish were able to survive without food as well as the laboratory/wild energetic comparisons suggests that negative size-selective overwinter mortality due to starvation is not a suitable explanation for the apparent recruitment failure of SU-spawned bluefish in recent years. Further investigation is necessary to determine what role activity level (i.e. migration) plays in the overwinter energetics of YOY bluefish and to determine whether smaller sizes of bluefish could endure starvation as well.

CHAPTER 2: EFFECTS OF ACTIVITY AND PRE-WINTER LIPID STORAGE

INTRODUCTION

Winter is a stressful period in the life of many young fishes characterized by declining water temperature and low prey abundance (Johnson and Evans 1996, Hurst and Conover 1998, see also Hurst and Conover 2001). Not only can low water temperature directly cause mortality through acute thermal stress (Lankford and Targett 2001), but it also slows the metabolism and digestion/evacuation rate of fishes, thus limiting their feeding rate (Cunjak *et al*. 1987, Cunjak and Power 1987). Also, low winter prey abundance limits the food availability of overwintering fishes (see Johnson and Evans 1996, see also Hurst and Conover 2001). Limited food availability combined with low feeding rates limits the amount of energy that fishes are able to assimilate in winter (Cunjak *et al*. 1987, see also Hurst and Conover 2001). When energy intake is insufficient to meet daily energetic requirements, fishes rely on stored energy (mainly lipids) to survive (Oliver *et al*. 1979, Cunjak *et al*. 1987, Cunjak 1988, Post and Evans 1989b, Thompson *et al*. 1991, Miranda and Hubbard 1994, Schultz and Conover 1997, Jobling 2001b).

Bluefish (*Pomatomus saltatrix*) are a coastal marine/estuarine fish with a worldwide, subtropical distribution (Briggs 1960, Champagnat *et al*. 1983, Juanes *et al*. 1996). Along the East Coast of North America they range from Nova Scotia to the Florida Keys (Robins *et al*. 1986), and are known to migrate north and south seasonally (McBride *et al*. 1993). Bluefish begin spawning in the South Atlantic Bight (SAB) in the springtime, before they migrate north for the summer (Kendall and Walford 1979, Collins

and Stender 1987, McBride *et al*. 1993). Then, in the fall, bluefish migrate south to overwinter (Lund and Maltezos 1970). It is widely debated whether bluefish spawn continuously (Hare and Cowen 1993, Smith *et al*. 1994) or in multiple, discrete events (Chiarella and Conover 1990), but the result is usually a bimodal length/frequency distribution of young-of-the-year (YOY) bluefish at the end of the summer growing season (McBride *et al*. 1993). This bimodal length/frequency distribution of YOY bluefish is comprised of larger-sized spring-spawned (SP) individuals and smaller-sized summer-spawned (SU) individuals (McBride *et al*. 1993). In 1960 and 1961, Lassiter (1962) found that both cohorts recruited equally to the adult population. In recent years, it has become apparent that, while the larger, SP-spawned bluefish still recruit to the adult population, the smaller, SU-spawned bluefish no longer do (Chiarella and Conover 1990, Gilmore 2000). It is currently not known what is happening to these YOY SU bluefish between the times of their estuarine egress in the fall and their recruiting to the adult population the following spring.

It is generally believed that overwinter survival probability in fishes is linked closely to the amount of lipid that is stored prior to winter (i.e. energy reserves) (Oliver *et al*. 1979, Miranda and Hubbard 1994, Schultz and Conover 1997, Schultz and Conover 1999, Jobling 2001b, Sogard and Olla 2001, see also Connolly and Petersen 2003). Many marine fish species undergo natural population fluctuations (Rothschild 1986), (Campana 1996), including those that young-of-the-year (YOY) bluefish, *Pomatomus saltatrix*, commonly feed on in the fall (i.e. bay anchovies *Anchoa mitchilli*, and Atlantic silversides *Menidia menidia*) (Able and Fahay 1998, Buckel *et al*. 1999). This can lead to yearly variations in the amount of energy that YOY bluefish have available for storage

prior to winter. Every autumn YOY bluefish migrate south in response to declining water temperatures and shorter day lengths in order to overwinter in more hospitable areas (Lund and Maltezos 1970, Munch 1997). In addition to the high activity levels associated with migration, wild bluefish also actively pursue prey while trying not to become prey (Buckel *et al*. 1999). A large amount of energy is likely required to fuel these fall/winter activities, however few studies have explored the effect activity level has on overwinter energy depletion in fish (Hurst and Conover 2001, Facey and Grossman 1990).

Under low activity levels and periods of high pre-winter lipid storage both spring (SP) and summer (SU) cohort YOY bluefish are well equipped to endure long periods of overwinter starvation (2001 experiment). However, the consequences of high activity levels or reduced opportunities for pre-winter lipid storage in YOY bluefish, particularly the SU cohort, are not known. The goal of the 2002 mesocosm experiment was to examine the importance of activity level and pre-winter lipid storage on the ability of SU-spawned YOY bluefish to survive the winter. I hypothesized that activity level and pre-winter lipid storage are important determinants for both overwinter energy levels and survival times in SU-spawned YOY bluefish.

METHODS

Experimental Subjects:

Experimental subjects, late SU-spawned YOY bluefish, were obtained using a beach seine at Sandy Hook, New Jersey on October 01, 2002. These bluefish were then held at the James J. Howard Marine Sciences Laboratory, Northeast Fisheries Science

Center in Sandy Hook, New Jersey until being transported to the University of North Carolina at Wilmington (UNCW) Center for Marine Science (CMS) via truck, on October 03. Upon arrival approximately 20 bluefish were sacrificed and immediately frozen for lipid analysis (termed Initial). All remaining bluefish were then placed into three 2,000-L circular fiberglass tanks and held until the start of the experiment. Tanks were supplied with ambient flow-through seawater (~20L/minute) obtained from the Atlantic Intra-Coastal Waterway (AICWW) adjacent to the CMS research pier.

Due to insufficient numbers of experimental subjects, more SU-spawned YOY bluefish were collected via beach seine in Beaufort, NC, on October 30. These individuals were transported to UNCW-CMS by truck on October 31. Upon arrival these bluefish, as well as the ones previously collected, were anesthetized using Tricaine-S (MS-222) and fork lengths, total lengths, and wet weights were recorded. After taking these measurements, all bluefish were revived and assigned, randomly, to one of twelve experimental tanks. For the next five days all bluefish were fed to satiation daily and allowed to acclimate to their new surroundings before the start of the experiment.

During the holding and acclimation periods, and when the treatments called for it, bluefish were fed to satiation once daily on a diet of bay anchovies and Atlantic silversides. Both species are known to be natural prey items of YOY bluefish in the wild at this time of year (Buckel and Conover 1997, Buckel *et al*. 1999). All feedings were performed in the early evening before dusk.

Mesocosm Setup:

The outdoor mesocosm setup was a flow-through system containing twelve 2,000-L tanks (1.8 meter diameter). Seawater was obtained from the AICWW at ambient salinity and temperature. Salinity ranged from 30 to 35 $^{o}/_{oo}$ and temperature ranged from <4.5^{o} to 20.8^{o}Celsius throughout the experiment (Figure 1). Temperature was recorded throughout the experiment using a miniature data logger (Onset Computer Corp., Pocasset, Massachusetts), which recorded water temperature at 30-minute intervals.

Experimental Design:

The experimental design was a fully-crossed 2X2 factorial design with activity level (high vs. low) and pre-winter lipid storage (high vs. low) as factors. Each treatment combination was replicated twice. This design occupied eight of the twelve available tanks (Figure 24).

High pre-winter lipid storage (i.e. high prey availability) was simulated by feeding bluefish to satiation once daily over the period from November 05 (Day 0) to December 06. Low pre-winter lipid storage was simulated by feeding bluefish to satiation only twice (on November 11 and November 24) during this same period. On December 07, pre-winter lipid manipulation ended and all tanks, regardless of treatment, were held without food for the remainder of the experiment (Figure 25).

Activity level manipulation was started the same day as pre-winter lipid manipulation (November 05), and was continued until the end of the experiment (Figure 25). High activity level was achieved by using Power Head PH2000 hydro-jets (1/tank) to create a continuous current averaging 11.35 cm/s. This current speed was

approximately equal to 0.8 body lengths per second and was assumed to approximate normal activity levels of wild bluefish. Tanks designated for low activity level treatments did not contain a hydro-jet and had a flow-rate of 0 cm/s. Both treatments and fish were assigned to tanks randomly.

Two additional conditions were simulated using the remaining four tanks: low activity levels and high pre-winter lipid storage (n=2) and high activity levels and low pre-winter lipid storage (n=2). Unlike the previous treatments mentioned, each of these treatments was fed to satiation once daily starting December 07, and lasting until the end to the experiment (Figure 24-25).

The two tanks designated as having low activity levels and high pre-winter lipid storage mimicked the SU-fed treatment from the previous year's experiment, while the two tanks with low activity levels and high pre-winter lipid storage that were not fed starting December 07 mimicked the SU-unfed treatment from the previous year's experiment. This set of treatments was established to help serve as a basis for comparison between this year's and the previous year's data.

The other two tanks that were fed to satiation once daily starting December 07, were designated as having high activity levels and low pre-winter lipid storage. It was hypothesized that the unfed equivalent of this treatment (described earlier-no feeding after Dec. 06) would be the most stressful to the YOY SU bluefish representing poor pre-winter (fall) prey availability coupled with the high energy demands of migration. This fed treatment tested whether YOY SU bluefish could recover from poor lipid storage in the fall coupled with high migration costs if they were able to locate sufficient prey during the winter.

On October 31, 2002, each tank was stocked with 13-14 SU-spawned YOY bluefish. Bluefish fork length (mm FL) ranged from 115-180 mm, with a mean value of 141 mm (Figure 26). These SU bluefish were smaller than those examined in the prior year's experiment (range: 179-229mm; mean: 206mm) (Figure 27); however, it is necessary to note the difference in sampling dates between the years (November 20, 2001 versus October 31, 2002). Cohort determination resulted from the bimodal length/frequency distribution of YOY bluefish as present off the coast of NC (Morley 2004), and based upon work by McBride *et al*. (1993), Munch (1997) and Gilmore (2000).

All tanks were checked for mortalities at least twice per day, with any mortalities being immediately frozen for later analyses. Once in the lab, all mortalities were dissected with the liver, viscera, white muscle (WM), red muscle (RM), and skin being removed for energetic condition analyses.

Parasitism:

Shortly after the start of the experiment it was noticed that some bluefish were suffering from parasitism by monogenetic trematodes. Due to this parasitism, all bluefish tanks underwent a series of formalin treatments to remove the parasites. Formalin treatments occurred on November 07, 11-13, and 19, and involved 600-800mL of formalin being added to each tank for up to two-hour periods.

Lipid Extraction:

Total non-polar lipids were extracted from bluefish tissues using the Soxhlet dry extraction protocol described in Schultz and Conover (1997). Non-polar lipids were monitored because this class includes the triacylglycerols (TAG's), which are the principal energy storage lipids in fishes (Jobling 2001a). Prior to extractions, cellulose thimbles (12mmX50mm) were dried at 60^{o}C for at least 48 hours. Viscera, WM, RM and skin samples were thawed, blotted gently, and placed in a tared, pre-labeled polystyrene dish. Wet weight was measured to ±0.0001 gram (g). Tissues were then dried at 60^{o}C for at least 48 hours. Liver samples were handled in a similar fashion, except they were not blotted prior to obtaining wet weights.

Soxhlet extractor flasks were filled (~110mL) with clean petroleum ether and heated sufficiently to produce a cycle rate of approximately 7.5 cycles/hour. Pre-dried thimbles were labeled and weighed (±0.0001g). Dry tissue samples to be extracted were weighed, placed into thimbles and put into the Soxhlet extraction devices. Samples were extracted for 4 hours to remove all non-polar lipids then dried at 60^{o}C for a minimum of 48 hours. Samples were then re-weighed to obtain a post extraction thimble/tissue dry weight. This weight was then subtracted from the combined weight of the thimble pre-extraction weight and the tissue pre-extraction weight. The difference represents the weight of total non-polar lipids removed from the tissue sample.

Ashing Data:

Ash content of tissues was measured as an indicator of their energetic condition (Ali *et al*. 2001). Lean (extracted) tissues were dried at 60^{o}C for at least 48 hours prior to

ashing. Ceramic crucibles (20mL) were pre-ashed at 450^{o}C for 24 hours prior to use. Pre-ashed crucibles were weighed (±0.0001g) and tared. Next, a tissue sample's dry weight was measured (±0.0001g) by placing it in the tared crucible. Samples were then ashed in a muffle furnace (Lindberg) at 450^{o}C for 24 hours. Crucibles were then placed in a drying oven and allowed to cool to 60^{o}C before being re-weighed. The ash weight was measured and expressed as a percentage of lean tissue dry weight. This ratio of inorganics to organics in the lean tissue was termed ash content (g ash/g lean tissue*100). As starved fish utilize energy reserves, the ash content of their body tissues increases (Ali *et al*. 2001, Jobling 2001b).

Statistical Analysis:

Lipid Energetics:

Two-way ANOVA were used on all final unfed treatments' ($Act._{high}/Stor._{high}$/Unfed, $Act._{high}/Stor._{low}$/Unfed, $Act._{low}/Stor._{high}$/Unfed, $Act._{low}/Stor._{low}$/Unfed) lipid data to evaluate the effects of activity level and pre-winter lipid storage on SU bluefish' energy reserves. Body condition indicators analyzed were: mean lipid content in grams (g), mean lipid density ((g lipid/g tissue dry weight (DWT))*100) in percent (%), mean lipid density (g lipid content/bluefish fork length (FL) in millimeters (mm), mean tissue DWT (g)/FL (mm) and mean FL (mm). All condition indicators were analyzed for each body depot (liver, viscera, WM, RM and skin). A significance value of α=0.05 was used. Significant main effects were analyzed post-hoc using Tukey's multiple comparisons tests. Prior to ANOVA, variances were tested for homogeneity using Levene's test. If variances were found to be significantly

heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using an extension of the Kruskal-Wallis ANOVA by ranks to individually test for activity and storage effects.

Final lipid data from the $Act._{high}/Stor._{low}$ fed and unfed treatments and the $Act._{low}/Stor._{high}$ fed and unfed treatments were each individually analyzed using T-tests to identify any significant winter-feeding effects on YOY bluefish energetics. All condition indicators were analyzed across all body depots. A significance value of $\alpha=0.05$ was used. Prior to T-tests, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using MWU-test.

Initial lipid data was individually compared to mean final lipid data from all treatments ($Act._{high}/Stor._{high}$/Unfed, $Act._{high}/Stor._{low}$/Unfed, $Act._{low}/Stor._{high}$/Unfed, $Act._{low}/Stor._{low}$/Unfed, $Act._{high}/Stor._{low}$/Fed and $Act._{low}/Stor._{high}$/Fed) using T-tests to investigate any significant treatment effect over time. All condition indicators were analyzed across all body depots. A significance value of $\alpha=0.05$ was used. Prior to T-tests, variances were tested for homogeneity using Levene's test. If variances were found to be significantly heterogeneous, then data were either log or ASIN (SQRT) transformed to meet the assumptions of equal variance. If data transformation did not remove the heterogeneity, untransformed data were reanalyzed nonparametrically using MWU-test.

All statistical analyses were performed using the computer software Statistica 6.0.

Ash Content:

Ash content from every body depot was analyzed similarly to the body condition indicators mentioned in both lipid energetics sections above.

RESULTS

The experiment was ended earlier than planned due to a system failure brought on by severe cold weather. On January 19, 2003, pipes delivering fresh seawater from the AICWW into UNCW's Center for Marine Science saltwater system froze and ruptured. Without ambient temperature seawater being supplied the tank temperatures dropped to atmospheric levels that were apparently below the lower lethal temperature for this species (<4.5^{o}C) (Figure 1). All bluefish in the experiment expired, save one. Although premature deaths prevented estimation of starvation thresholds and survival times, the lipid levels of bluefish at the time of death were quantified to examine whether the 2.5-month treatment exposures had measurable effects on bluefish energetics.

Initial bluefish subsampled on October 3, 2002, were in normal condition compared to wild bluefish at that time period. The energetic condition of initial bluefish increased over the course of the experiment. For each body condition index investigated and across all five of the body depots, the initial energy values were found to be lower than most, if not all, of the final values in each treatment (Figure 28a-d). For liver, initial mean lipid content was >30 times lower (0.001425 g) than the mean final lipid content for each treatment (Tables 34-39). Initial liver lipid densities (%) were >13 times lower

than final values in each treatment (Tables 34-39). This trend was evident in other body depots, but to a lesser extent.

Effects of Activity Level and Pre-Winter Lipid Storage:

Winter activity level had little effect on final YOY bluefish energy reserves (Table 40; Figure 28a-d). The ability to store lipids prior to winter (here after referred to as storage) was associated with significant increases in all body depots and all final condition indices except viscera DWT/FL ($p=0.257$) and WM DWT/FL ($p=0.082$) (Table 40; Figure 28a-d). There was no significant activity*storage interaction (Table 40). After nonparametric analysis, viscera lipid density (%) was found to have a significant activity*storage interaction (Table 40). Mean final bluefish FL (mm) was not affected by activity ($p=0.716$) or storage ($p=0.066$) (Table 40).

Effects of Winter Feeding:

For the two treatments that had both a fed and unfed component to their design ($Act._{high}/Stor._{low}$ and $Act._{low}/Stor._{high}$), t-tests were used to test the significance of winter food availability on bluefish energetic condition. For the $Act._{high}/Stor._{low}$ treatments, winter-feeding was found to have a significant, positive effect on all condition factors for both viscera and RM (Table 41; Figure 28a-d). All liver condition factors were significant except for lipid density (%) (Table 41). All WM condition factors were significant except for DWT/FL ($p=0.055$) (Table 41). For skin only lipid density (%) and lipid density (g lipid/mm FL) were significant, while lipid content was nearly significant ($p<0.06$) (Table 41). Mean final bluefish FL was not significantly different among those

treatments (Table 41). For the $Act._{low}/Stor._{high}$ treatments, winter-feeding had no significant effect on the energetic condition of any body depots (Table 42; Figure 28a-d). Mean final FL also did not differ significantly between treatments (Table 42).

Initial vs. Final Energetics:

Final values for the $Act._{high}/Stor._{high}$/Unfed treatment (HHU) were significantly higher than initial values for every condition factor and all body depots except viscera DWT/FL ($p=0.067$) and WM DWT/FL ($p=0.059$) (Table 34; Figure 28a-d). Mean bluefish FL did not increase significantly during the experiment (Table 34).

The $Act._{high}/Stor._{low}$/Unfed treatment (HLU) final values were significantly higher than initial values for all of the liver condition indices (Table 35; Figure 28a-d). Final condition indices for other body depots were not significantly different from initial values, including mean bluefish FL ($p=0.079$) (Table 35).

The $Act._{high}/Stor._{low}$/Fed treatment (HLF) final values were significantly higher than initial values for all condition indices in all body depots except skin DWT/FL ($p=0.051$) (Table 36; Figure 28a-d). Mean bluefish FL did not increase significantly during the experiment (Table 36).

The $Act._{low}/Stor._{high}$/Unfed treatment (LHU) final values were significantly higher than initial values for all condition indices in all body depots except viscera DWT/FL ($p=0.064$) and WM DWT/FL ($p=0.137$) (Table 37; Figure 28a-d). Mean bluefish FL did not increase significantly during the experiment (Table 37).

The $Act._{low}/Stor._{high}$/Fed treatment (LHF) final values were significantly higher than initial values for all condition indices in all body depots except WM DWT/FL

(p=0.061) and skin DWT/FL (p=0.087) (Table 38; Figure 28a-d). Mean bluefish FL did not increase significantly during the experiment (Table 38).

The $Act._{low}/Stor._{low}$/Unfed treatment (LLU) final values were not significantly different than initial values for all condition indices in all body depots except liver (Table 39; Figure 28a-d). All final liver condition factors were significantly larger than initial values (Table 39). Mean bluefish FL also increased significantly during the experiment (p=0.044) (Table 39).

Ash Content:

Two-way ANOVA revealed that activity level did not significantly affect the ash content of any of the five body compartments examined (Table 43; Figure 29). The effect of high pre-winter storage on ash content was only significant for the RM depot (Table 43). Ash content did not show a significant storage effect in any other body depot (Table 43). There was no significant activity*storage interaction (Table 43).

In the $Act._{high}/Stor._{low}$ treatment, t-tests revealed that winter food availability had significant, negative effects on ash content of liver, WM and RM (Table 44; Figure 29). For the $Act._{low}/Stor._{high}$ treatments, winter food availability did not significantly affect ash content of any body depot (Table 45; Figure 29).

Ash Content: Initial vs. Final

Within the $Act._{high}/Stor._{high}$ treatment, mean ash content of unfed individuals (HHU) did not decrease significantly in any body depot except skin (Table 46; Figure

29). The decreases in both WM (p=0.070) and RM (p=0.059) were approaching significance (Table 46).

Within the $Act._{high}/Stor._{low}$ storage treatment, mean ash content of unfed individuals (HLU) did not decrease significantly in any body depot except skin (p=0.034) (Table 47; Figure 29).

Within the $Act._{high}/Stor._{low}$ treatment, mean ash content of fed individuals (HLF) decreased significantly for every body depot except viscera (p=0.058) (Table 48; Figure 29).

Within the $Act._{low}/Stor._{high}$ treatment, mean ash content of unfed individuals (LHU) decreased significantly for every body depot except liver (p=0.058) and viscera (p=0.137) (Table 49; Figure 29).

Within the $Act._{low}/Stor._{high}$ treatment, mean ash content of fed individuals (LHF) decreased significantly for every body depot (Table 50; Figure 29).

Within the $Act._{low}/Stor._{low}$ treatment, mean ash content of unfed individuals (LLU) did not decrease significantly in any body depot except skin (p=0.046) (Table 51; Figure 29). The decreases in both WM (p=0.052) and RM (p=0.052) were approaching significance (Table 51).

DISCUSSION

Based on prior observations that SU-spawned YOY bluefish could endure winter starvation for very long periods of time, the present investigation was conducted to assess the influence of forced activity and pre-winter lipid storage on the overwintering ability of SU-spawned individuals. These two factors were analyzed to determine their role in

the hypothesis that the apparent recruitment failure of SU-spawned bluefish reflects size-selective overwinter starvation. Increased activity level has been shown to raise metabolic demands in striped bass (Hurst and Conover 2001) and other fishes (Facey and Grossman 1990), and is believed to cause energy reserves to be depleted at a faster rate. Bluefish are highly active and known to migrate south in preparation for winter (Wilk 1977). Activity level was investigated to assess whether increased activity level prior to winter (i.e. fall migration) and during winter compromises the ability of YOY bluefish to endure overwinter starvation. Bluefish are known to feed heavily and actively store energy during the fall (Buckel *et al*. 1999, Morley 2004), presumably to prepare for the winter when food availability may become low (see Shul'man 1974, Morley 2004) and low water temperatures may reduce digestive and assimilatory efficiency (Cunjak *et al*. 1987, Cunjak and Power 1987). Fall abundances of bluefish prey species may fluctuate year to year (Rothschild 1986, Campana 1996, Buckel *et al*. 1999). Such fluctuations in prey availability may directly affect how much energy bluefish are able to store prior to winter. The pre-winter lipid storage treatment was intended to simulate variable prey conditions in fall when YOY bluefish are actively storing lipids.

Unfortunately, this experiment was concluded prematurely due to a mechanical failure that caused nearly all experimental subjects to perish. Therefore, the extent to which activity level and pre-winter lipid storage influenced the overwinter survival duration of SU-spawned YOY bluefish could not be determined. However, subjects were analyzed to determine the effect of these factors on energy dynamics and their implications for the apparent recent recruitment failure of SU-spawned bluefish.

Winter Energetics:

When comparing final energy condition values between treatments a significant effect of pre-winter energy storage was observed for most condition indices and body depots. In general, activity level did not have strong effects on energy dynamics. This suggests that the amount of energy that bluefish store prior to winter would have a greater effect on their ability to endure starvation than their level of activity during late fall and winter. However, since the experiment was ended after only 2.5 months (January 19, 2003) the extent to which prolonged activity levels may adversely affect bluefish energy reserves is not known.

Energy Storage:

Based on the 2001 experiment, it was concluded that SU-spawned YOY bluefish (175-315 mm FL) have a high capacity to endure winter starvation. Data from the 2002 experiment support this conclusion and also illustrate that these fishes have a remarkable ability to store energy rapidly prior to winter. Subjects in the most energetically demanding treatment ($Act._{high}/Stor._{low}$/Unfed) that were subjected to high activity levels without food displayed higher energetic condition after 2.5 months than initial bluefish. Since bluefish in this treatment were not fed during the experiment, the only way they could have stored energy prior to the experiment was during the approximately 30-day acclimation period. Essentially, these bluefish were able to store more energy during this period than was required to survive >2.5 months without food and at high activity levels. This is evidenced by the significantly higher final values than initial values for every condition index in liver along with a significantly lower percent ash for skin. The

depletion of energy reserves in every body depot except liver suggests that SU bluefish may defend liver energy stores while preferentially depleting energy reserves in other body depots. The significantly lower skin ash content of final bluefish compared to initial bluefish implies that they were in better energetic condition. A possible explanation for the significant difference found in skin and not any other body depot lies in its function. Skin is an animal's first line of defense, protecting it from mechanical and bacterial stress (Campbell 1996). Therefore, it may be important not to utilize this reserve, and thus compromise its ability to perform, until absolutely necessary.

SU-spawned bluefish appear to have a remarkable energy storage capacity, which complements their starvation endurance, making them highly resistant to winter starvation. Furthermore, the overall condition of initial bluefish was not energetically poor and is comparable to those of similar-sized wild bluefish captured at the same time (Morley 2004).

An important question addressed by this experiment was whether SU-spawned YOY bluefish could compensate for low prey availability in the fall by feeding during the winter. Limited winter feeding has been observed in several species of fishes, including brook trout (*Salvelinus fontinalis*) and brown trout (*Salmo trutta*) (Cunjak and Power 1997), striped bass (*Morone saxatilis*) (Hurst and Conover 2001), white perch (*Morone americana*) (Johnson and Evans 1990), white crappie (*Pomoxis annularis*) (McCollum *et al*. 2003), and Atlantic salmon (*Salmo salar*) (Metcalfe and Thorpe 1992), however it does not occur in smallmouth bass (*Micropterus dolomieui*) (Oliver *et al*. 1979). It appears that bluefish were able to feed during the winter. The $Act._{high}/Stor._{low}$ treatment in which bluefish were fed starting December 6, 2002, after having been starved for the

month of November, displayed significantly better condition than the corresponding unfed treatment for most indices and body depots. There was also no significant difference in mean FL suggesting that energy storage may be prioritized over growth during winter. This agrees with Shultz and Conover (1997) and Post and Parkinson (2001), who suggested that lipid storage is a more beneficial allocation strategy for rapidly growing fishes (i.e. bluefish) in the fall than growth rate maximization. The lack of a difference in skin DWT/FL may reflect the limited amount of skin fish can posses per unit body length, regardless of feeding. The lack of a significant difference between liver lipid density (%) in winter-fed and winter-unfed bluefish in the $Act._{high}/Stor._{low}$ treatment suggests that $Act._{high}/Stor._{low}$/Unfed bluefish had not yet depleted liver energy stores. Ash content in $Act._{high}/Stor._{low}$/Fed bluefish was significantly lower than in $Act._{high}/Stor._{low}$/Unfed bluefish for liver, WM, and RM, suggesting that these depots had a higher percent of organic material and were therefore in better condition. The lack of a significant difference between fed and unfed treatments for both viscera and skin ash content might be the result of these tissues being harder to utilize organic material from in order to help endure periods without food. Interestingly, despite not being fed in the fall and only receiving food in the winter, $Act._{high}/Stor._{low}$/Fed treatment bluefish maintained comparable energy stores to the $Act._{low}/Stor._{high}$/Unfed and $Act._{low}/Stor._{high}$/Fed treatment bluefish for most condition indices and most body depots. Overall, SU-spawned YOY bluefish appeared capable of assimilating prey in the winter, when available, to compensate for poor feeding conditions prior to winter. This ability, along with their high starvation endurance, is not consistent with the starvation

hypothesis as an explanation for the apparent recent recruitment failure of SU-spawned YOY bluefish.

The effects of winter food availability were less apparent for the $Act._{low}/Stor._{high}$ treatments. There was no significant difference in final energy stores or ash content between $Act._{low}/Stor._{high}$/Unfed and $Act._{low}/Stor._{high}$/Fed bluefish. Since the significant difference between $Act._{high}/Stor._{low}$/Unfed and $Act._{high}/Stor._{low}$/Fed treatments showed that bluefish are capable of feeding and storing/maintaining energy in the winter, the lack of a difference between $Act._{low}/Stor._{high}$/Unfed and $Act._{low}/Stor._{high}$/Fed treatments supports the defended energy hypothesis suggested from the year 2001 results (see Chapter 1 'Discussion'). The defended energy hypothesis states that individuals will feed selectively in the winter to defend their energy reserves only if these reserves are depleted below a minimum level (Metcalfe and Thorpe 1992). The significant difference between the $Act._{high}/Stor._{low}$/Unfed and $Act._{high}/Stor._{low}$/Fed treatments, combined with the lack of a difference between the $Act._{low}/Stor._{high}$/Unfed and $Act._{low}/Stor._{high}$/Fed treatments, supports this hypothesis. The $Act._{high}/Stor._{low}$ treatments were energetically stressed with both high activity levels and poor feeding prior to winter, so the significant difference between the different winter-feeding treatments can be attributed to fish defending their energy stores when food is available. The $Act._{low}/Stor._{high}$ treatments were less stressed prior to winter, so the lack of an effect of winter food availability may reflect fish choosing not to store energy despite food being available. These findings suggest that SU-spawned bluefish are capable of storing energy during the winter and that the defended energy hypothesis may help to explain winter energy dynamics.

Despite the 2002 experiment ending prematurely, useful information was obtained with which to further address the winter starvation hypothesis. Results indicated that SU bluefish can endure winter starvation for long periods without significant depletion of energy reserves and that they have the ability to greatly increase their energy reserves in a short period of time, providing sufficient prey is available. Also, pre-winter energy storage appears to have a greater effect on bluefish winter energy reserves than activity level. If a bluefish does have poor food availability in the fall and is unable to store sufficient energy to survive the winter without food, then it is capable of feeding in the winter to maintain its energy reserves if prey are encountered. Overall, these experimental findings are inconsistent with the hypothesis that the apparent recruitment failure of SU-spawned bluefish results from size-dependent winter starvation.

EXPERIMENTAL SIGNIFICANCE

Our understanding of the winter energetics of marine fishes is generally poor. Decreases in temperature, food limitations, and changes in activity levels are but a few of the stresses that marine fishes have to endure throughout the winter. By further exploring these potential stresses, valuable insight into the winter energetics of marine fishes might be gained.

This experiment was designed to address the apparent recent recruitment failure of SU-spawned YOY bluefish in the western North Atlantic. Specifically, it addressed the hypothesis that SU-spawned YOY bluefish encounter negative size-selective over-winter mortality due to starvation. Overall, due to the bluefish's ability to store lipid rapidly, deplete lipid slowly, access multiple body depots for both energy storage and depletion and the incredible length of time that they are able to endure starvation, this study concludes that overwinter mortality due to starvation is not a likely explanation for this apparent recent recruitment failure of SU-spawned YOY bluefish. With bluefish being such an important species, both commercially and recreationally, further information is necessary to help better understand/explain their recent decline. Once this mechanism is identified it can more easily be determined if and how to address their decline and whether or not it can be reversed. In addition, any further knowledge on bluefish life history will lead to more informed bluefish management plans.

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Table 1. Results of two-way ANOVA used to evaluate the effects of cohort of origin (spring-spawned versus summer-spawned) and body depot (liver, viscera, white muscle, red muscle and skin) on lipid content (g) and lipid density (%) of YOY bluefish subsampled on day 0 of the experiment.

Cohort & Depot	INITIAL		Effects Cohort	Effects Cohort	Effects Body Depot	Effects Body Depot	Effects Cohort*Depot	Effects Cohort*Depot	Levene's	Levene's
Variable	Trans.	Test	F-value	P-value	F-value	P-value	F-value	P-value	F-value	P-value
lipid_g	log	2-way Anova	116.2	0.000	53.6	0.000	3.2	0.017	0.561626	0.825
lipid_%	raw	2-way Anova	32.14	0.000	35.36	0.000	4.56	0.002	1.085892	0.381

Table 2. Pearson product-limit correlation coefficients for (A) spring cohort lipid content (g), (B) summer cohort lipid content, (C) spring cohort lipid density (%) and (D) summer cohort lipid density across five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in spring- and summer-spawned YOY bluefish subsampled on day 0 of the experiment. All significant ($p<0.05$) coefficients are indicated by an asterisk (*).

A)

spring cohort	liver_lipid_g_0	viscera_lipid_g_0	WM_lipid_g_0	RM_lipid_g_0	skin_lipid_g_0
liver_lipid_g_0	1.00	0.59	0.76*	0.84*	0.90*
viscera_lipid_g_0	0.59	1.00	0.70*	0.71*	0.70*
WM_lipid_g_0	0.76*	0.70*	1.00	0.84*	0.89*
RM_lipid_g_0	0.84*	0.71*	0.84*	1.00	0.93*
skin_lipid_g_0	0.90*	0.70*	0.89*	0.93*	1.00

B)

summer cohort	liver_lipid_g_0	viscera_lipid_g_0	WM_lipid_g_0	RM_lipid_g_0	skin_lipid_g_0
liver_lipid_g_0	1.00	0.47	0.33	0.31	0.19
viscera_lipid_g_0	0.47	1.00	0.97*	0.81*	0.70*
WM_lipid_g_0	0.33	0.97*	1.00	0.82*	0.76*
RM_lipid_g_0	0.31	0.81*	0.82*	1.00	0.94*
skin_lipid_g_0	0.19	0.70*	0.76*	0.94*	1.00

C)

spring cohort	liver_lipid_%_0	viscera_lipid_%_0	WM_lipid_%_0	RM_lipid_%_0	skin_lipid_%_0
liver_lipid_%_0	1.00	0.60	0.50	0.56	0.68*
viscera_lipid_%_0	0.60	1.00	0.74*	0.75*	0.77*
WM_lipid_%_0	0.50	0.74*	1.00	0.81*	0.88*
RM_lipid_%_0	0.56	0.75*	0.81*	1.00	0.88*
skin_lipid_%_0	0.68*	0.77*	0.88*	0.88*	1.00

D)

summer cohort	liver_lipid_%_0	viscera_lipid_%_0	WM_lipid_%_0	RM_lipid_%_0	skin_lipid_%_0
liver_lipid_%_0	1.00	0.30	0.27	0.28	0.35
viscera_lipid_%_0	0.30	1.00	0.94*	0.74*	0.74*
WM_lipid_%_0	0.27	0.94*	1.00	0.76*	0.79*
RM_lipid_%_0	0.28	0.74*	0.76*	1.00	0.94*
skin_lipid_%_0	0.35	0.74*	0.79*	0.94*	1.00

Table 3. Results of repeated-measures ANOVA used to evaluate changes in lipid content (g) and lipid density (%) of different body depots (liver, viscera, white muscle, red muscle and skin) of YOY bluefish from the Spring-Fed treatment. Subsamples were taken on days 0, 11, and 31 of the experiment.

Spring Fed INITIAL-11-31 Variable	Trans.	Test	Effects Body Depot F-value	Effects Body Depot P-value	Effects Time F-value	Effects Time P-value	Effects Depot*Time F-value	Effects Depot*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
lipid g	log	R.M. Anova	49.94	0.000	3.44	0.042	0.27	0.972	0.174	0.881424	0.273256
lipid_%	raw	R.M. Anova	49.06	0.000	2.260	0.117	0.150	0.996	0.1968	0.7136	0.9471

Table 4. Results of repeated-measures ANOVA used to evaluate changes in lipid content (g) and lipid density (%) of different body depots (liver, viscera, white muscle, red muscle and skin) of YOY bluefish from the Summer-Fed treatment. Subsamples were taken on days 0, 11, 31 and 89 of the experiment.

Summer Fed INITIAL-11-31-89 Variable	Trans.	Test	Effects Body Depot F-value	Effects Body Depot P-value	Effects Time F-value	Effects Time P-value	Effects Depot*Time F-value	Effects Depot*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value	89 Levene's P-value
lipid g	log	R.M. Anova	117.4	0.000	21.1	0.000	0.9	0.591	0.976	0.932	0.431	0.623934
lipid_%	raw	R.M. Anova	71.6	0.000	19.08	0.000	0.94	0.517	0.85437	0.183928	0.194604	0.026369

Table 5. Results of repeated-measures ANOVA used to evaluate changes in lipid content (g) and lipid density (%) of different body depots (liver, viscera, white muscle, red muscle and skin) of bluefish from the Spring-Unfed treatment. Subsamples were taken on days 0, 11 and 31 of the experiment.

Spring Unfed INITIAL-11-31 Variable	Trans.	Test	Effects Body Depot F-value	Effects Body Depot P-value	Effects Time F-value	Effects Time P-value	Effects Depot*Time F-value	Effects Depot*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
lipid g	log	R.M. Anova	48.82	0.000	14.55	0.000	1.38	0.244	0.329	0.704946	0.482928
lipid_%	raw	R.M. Anova	44.89	0.000	7.000	0.003	3.540	0.005	0.2998	0.5356	0.8791

Table 6. Results of repeated-measures ANOVA used to evaluate changes in lipid content (g) and lipid density (%) of different body depots (liver, viscera, white muscle, red muscle and skin) of YOY bluefish from the Summer-Unfed treatment. Subsamples were taken on days 0, 11, 31 and 89 of the experiment.

Summer Unfed INITIAL-11-31-89 Variable	Trans.	Test	Effects Body Depot F-value	Effects Body Depot P-value	Effects Time F-value	Effects Time P-value	Effects Depot*Time F-value	Effects Depot*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value	89 Levene's P-value
lipid g	log	R.M. Anova	109.6	0.000	19.1	0.000	1	0.419	0.651	0.948104	0.594991	0.622507
lipid_%	raw	R.M. Anova	41.98	0.000	6.310	0.001	1.150	0.342	0.3757	0.3844	0.0031	0.0542

Table 7. Results of t-tests used to determine the effects of cohort of origin (SP=spring-spawned, n=19; SU=summer-spawned, n=18) on various condition indices of YOY bluefish subsampled on day 0 of the experiment. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

SP vs. SU Variable	Day 0 Trans.	Test	Mean Spring	Std.Dev. Spring	Mean Summer	Std.Dev. Summer	T- & Z-Values	p	p Levene
liver_lipid_g	log	T-test	-0.686943	0.325512	-0.846080	0.225805	1.718554	0.094531	0.035264
liver_lipid_g	raw	MWU	N/A	N/A	N/A	N/A	1.732051	0.083265	N/A
liver_lipid_%	raw	T-test	0.239130	0.107388	0.269506	0.075174	-0.991491	0.328251	0.049046
liver_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	-0.972379	0.330863	N/A
liver_lipid_g/FL	log	T-test	-3.081890	0.317512	-3.155230	0.225126	0.806299	0.425515	0.041706
liver_lipid_g/FL	raw	MWU	N/A	N/A	N/A	N/A	0.941993	0.346197	N/A
liver_DWT_g/FL	raw	T-test	0.004051	0.001174	0.002832	0.000989	3.403797	0.001680	0.126839
WM_lipid_%	ASIN	T-test	0.385878	0.136943	0.172807	0.090210	5.512608	0.000004	0.091682
Mean FL	log	T-test	2.394952	0.033872	2.309149	0.025842	8.626727	0.000000	0.215000

Table 8. Results of two-way ANOVA used to evaluate the effects of cohort of origin (spring-spawned versus summer-spawned) and feeding (fed versus unfed) on various body condition indices for overwintering YOY bluefish subsampled on day 11 of the experiment.

Cohort & Feeding	Day 11 subsample		Effects Cohort	Effects Cohort	Effects Feeding	Effects Feeding	Effects Cohort*Feeding	Effects Cohort*Feeding	Levene's	Levene's
Variable	Trans.	Test	F-value	P-value	F-value	P-value	F-value	P-value	F-value	P-value
liver_lipid_g	log	2-way Anova	15.59	0.000	15.3	0.000	1.7	0.201	0.390023	0.7610
liver_lipid_%	raw	2-way Anova	1.766	0.193	8.066	0.008	1.037	0.316	0.683049	0.5690
liver_lipid_g/FL	log	2-way Anova	7.41	0.010	14.69	0.001	1.62	0.213	0.293344	0.8299
liver_DWT_g/FL	log	2-way Anova	44.4	0.000	100.6	0.000	0.9	0.360	0.376728	0.7704
WM_lipid_%	raw	2-way Anova	9.122	0.005	2.254	0.143	0.498	0.485	2.13746	0.1149
Mean FL	raw	2-way Anova	78.96	0.000	0.07	0.790	0	1.000	0.5371	0.6602

Table 9. Results of two-way ANOVA used to evaluate the effects of cohort of origin (spring-spawned versus summer-spawned) and feeding (fed versus unfed) on various body condition indices for overwintering YOY bluefish subsampled on day 31 of the experiment. Variables found to have heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using Kruskal-Wallis ANOVA to test individually for cohort and feeding effects.

Cohort & Feeding	Day 31 subsample		Effects Cohort	Effects Cohort	Effects Feeding	Effects Feeding	Effects Cohort*Feeding	Effects Cohort*Feeding	Levene's	Levene's
Variable	Trans.	Test	F-value	P-value	F-value	P-value	F-value	P-value	F-value	P-value
liver_lipid_g	raw	2-way Anova	11.28	0.002	25.58	0.000	0.04	0.837	0.876368	0.4636
liver_lipid_%	ASIN	2-way Anova	0.429	0.517	6.249	0.018	0.963	0.334	3.123738	0.0394
liver_lipid_%	raw	Kruskal-Wallis	0.2252	0.635	6.5675	0.010	N/A	N/A	N/A	N/A
liver_lipid_g/FL	raw	2-way Anova	3.33	0.078	32.05	0.000	0.93	0.341	0.856915	0.4734
liver_DWT_g/FL	log	2-way Anova	41.3	0.000	218.3	0.000	7.2	0.012	2.7107	0.0613
WM_lipid_%	ASIN	2-way Anova	1.943	0.173	5.93	0.021	5.503	0.025	4.7726	0.0073
WM_lipid_%	raw	Kruskal-Wallis	1.4454	0.229	5.9349	0.015	N/A	N/A	N/A	N/A
Mean FL	raw	2-way Anova	178.7	0.000	0.2	0.635	0.5	0.488	0.938182	0.4337

Table 10. Results of t-tests used to determine the effects of feeding (Fed, n=9; Unfed, n=9) on various condition indices of summer-spawned YOY bluefish subsampled on day 89 of the experiment. For WM lipid density (%) the sample size of unfed individuals was n=8. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

Summer cohort	Day 89 subsample		Mean	Std.Dev.	Mean	Std.Dev.	T- or Z-		p
Variable	Trans.	Test	Fed	Fed	Unfed	Unfed	Value	p	Levene
liver_lipid_g	raw	T-test	0.293544	0.125038	0.100900	0.095784	3.669216	0.002073	0.472875
liver_lipid_%	ASIN	T-test	0.679695	0.096539	0.631113	0.215093	0.618186	0.545153	0.006484
liver_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	0.397360	0.691103	N/A
liver_lipid_g/FL	raw	T-test	0.001367	0.000564	0.000475	0.000428	3.776048	0.001654	0.554226
liver_DWT_g/FL	raw	T-test	0.003442	0.001057	0.001125	0.000505	5.936524	0.000021	0.193211
WM_lipid_%	ASIN	T-test	0.341423	0.080409	0.056244	0.024786	0.8334247	0.000001	0.038884
WM_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	3.464102	0.000532	N/A
Mean FL	raw	T-test	213.8889	7.991315	209.3333	15.70032	0.775763	0.449204	0.054470

Table 11. Results of t-tests used to determine the effects of cohort of origin (spring-spawned, n=10; summer-spawned, n=11) on various condition indices of fed YOY bluefish subsampled on day 192 of the experiment.

Fed Treatments	Day 192 subsample		Mean	Std.Dev.	Mean	Std.Dev.	T-		p
Variable	Trans.	Test	Spring	Spring	Summer	Summer	Value	p	Levene
liver_lipid_g	raw	T-test	0.045500	0.036102	0.038773	0.031501	0.456076	0.653506	0.975825
liver_lipid_%	raw	T-test	0.083056	0.054357	0.096184	0.057135	-0.538105	0.596752	0.964831
liver_lipid_g/FL	raw	T-test	0.000166	0.000133	0.000161	0.000126	0.095037	0.925281	0.866833
liver_DWT_g/FL	raw	T-test	0.001962	0.000399	0.001560	0.000468	2.107596	0.048578	0.801858
WM_lipid_%	raw	T-test	0.020328	0.018403	0.016044	0.012268	0.633483	0.533970	0.375932
Mean FL	raw	T-test	274.9000	13.62555	237.7273	9.644593	7.271281	0.000001	0.545183

Table 12. Results of repeated-measures ANOVA used to evaluate changes in various body condition indices of fed spring and summer cohort bluefish. Subsamples were taken on days 0, 11 and 31 of the experiment.

Fed INITIAL-11-31 Variable	Trans.	Test	Effects Cohort F-value	Effects Cohort P-value	Effects Time F-value	Effects Time P-value	Effects Cohort*Time F-value	Effects Cohort*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
liver_lipid_g	log	R.M. Anova	4.39	0.052	18.340	0.000	0.590	0.561	0.2025	0.2246	0.7851
liver_lipid_%	raw	R.M. Anova	6.862	0.019	2.692	0.083	0.020	0.980	0.2899	0.9525	0.6596
liver_lipid_g/FL	raw	R.M. Anova	1.95	0.182	16.890	0.000	0.460	0.637	0.1310	0.0731	0.9051
liver_DWT_g/FL	log	R.M. Anova	32.96	0.000	39.110	0.000	1.460	0.247	0.2187	0.4423	0.0667
WM_lipid_%	ASIN	R.M. Anova	2.845	0.112	6.247	0.005	2.898	0.071	0.2250	0.4904	0.9345
WWT/FL	log	R.M. Anova	123.7	0.000	2.900	0.072	0.700	0.496	0.0742	0.2631	0.9588
Mean FL	raw	R.M. Anova	136	0.000	1.700	0.191	1.500	0.246	0.1813	0.1420	0.1139

Table 13. Results of repeated-measures ANOVA used to evaluate changes in various body condition indices of unfed spring and summer cohort bluefish. Subsamples were taken on days 0, 11 and 31 of the experiment.

Unfed INITIAL-11-31 Variable	Trans.	Test	Effects Cohort F-value	Effects Cohort P-value	Effects Time F-value	Effects Time P-value	Effects Cohort*Time F-value	Effects Cohort*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
liver_lipid_g	raw	R.M. Anova	17.32	0.001	2.930	0.068	0.330	0.720	0.8319	0.0851	0.3385
liver_lipid_%	raw	R.M. Anova	0.032	0.860	2.493	0.099	0.039	0.962	0.6558	0.4378	0.0892
liver_lipid_g/FL	raw	R.M. Anova	10.61	0.005	3.900	0.030	0.260	0.769	0.9337	0.2520	0.6358
liver_DWT_g/FL	raw	R.M. Anova	25.14	0.000	26.200	0.000	0.040	0.963	0.7936	0.0525	0.7711
WM_lipid_%	ASIN	R.M. Anova	28.01	0.000	0.310	0.738	2.870	0.071	0.5178	0.1481	0.0428
WWT/FL	raw	R.M. Anova	134.8	0.000	3.400	0.046	0.900	0.398	0.0638	0.3954	0.5934
Mean FL	raw	R.M. Anova	125.9	0.000	3.300	0.052	2.900	0.070	0.1914	0.6708	0.7479

Table 14. Results of repeated-measures ANOVA used to evaluate changes in various body condition indices of fed and unfed spring-spawned YOY bluefish. Subsamples were taken on days 0, 11 and 31 of the experiment.

Spring INITIAL-11-31 Variable	Trans.	Test	Effects Feeding F-value	Effects Feeding P-value	Effects Time F-value	Effects Time P-value	Effects Feeding*Time F-value	Effects Feeding*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
liver_lipid_g	raw	R.M. Anova	5.287	0.035	3.589	0.039	6.115	0.006	0.5595	0.0872	0.5590
liver_lipid_%	raw	R.M. Anova	24.38	0.000	2.810	0.075	0.110	0.894	0.8913	0.8226	0.5994
liver_lipid_g/FL	raw	R.M. Anova	5.406	0.034	2.561	0.093	7.037	0.003	0.8067	0.0692	0.4544
liver_DWT_g/FL	log	R.M. Anova	72.59	0.000	1.360	0.272	29.500	0.000	0.4643	0.8722	0.2102
WM_lipid_%	raw	R.M. Anova	1.421	0.251	0.012	0.988	1.385	0.265	0.8721	0.9935	0.0154
WWT/FL	raw	R.M. Anova	2.428	0.139	1.648	0.208	1.855	0.173	0.4471	0.1112	0.1559
Mean FL	raw	R.M. Anova	0.076	0.787	8.146	0.001	0.437	0.650	0.7126	0.7672	0.8599

Table 15. Results of repeated-measures ANOVA used to evaluate changes in various body condition indices of fed and unfed summer-spawned YOY bluefish. Subsamples were taken on days 0, 11, 31 and 89 of the experiment.

Summer INITIAL-11-31-89 Variable	Trans.	Test	Effects Feeding F-value	Effects Feeding P-value	Effects Time F-value	Effects Time P-value	Effects Feeding*Time F-value	Effects Feeding*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value	89 Levene's P-value
liver_lipid_g	raw	R.M. Anova	23.94	0.000	2.700	0.056	7.630	0.000	0.3506	0.2454	0.1760	0.4728
liver_lipid_%	raw	R.M. Anova	1.113	0.307	3.461	0.023	0.704	0.554	0.9407	0.2812	0.0045	0.0043
liver_lipid_g/FL	raw	R.M. Anova	24.93	0.000	2.900	0.045	8.250	0.000	0.3923	0.1633	0.1642	0.5542
liver_DWT_g/FL	log	R.M. Anova	193.9	0.000	4.400	0.008	19.000	0.000	0.4735	0.3730	0.2915	0.2971
WM_lipid_%	ASIN	R.M. Anova	59.77	0.000	5.990	0.002	5.430	0.003	0.6273	0.0816	0.3684	0.0389
WWT/FL	raw	R.M. Anova	23.89	0.000	2.900	0.044	3.910	0.014	0.3508	0.2399	0.5984	0.9724
Mean FL	raw	R.M. Anova	1.848	0.193	1.902	0.142	0.268	0.848	0.8465	0.7159	0.2143	0.0544

Table 16. Results of t-tests used to determine the effects of time [Day 0 (Initial), n=9; Day 192 (Final), n=10] on various condition indices of overwintering YOY bluefish in the Spring-Fed treatment (SP_F). Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

SP_F (Initial-Final) Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	log	T-test	-0.838242	0.328427	-1.42318	0.255904	4.355616	0.000430	0.279573
liver_lipid_%	raw	T-test	0.186826	0.104072	0.083056	0.054357	2.767183	0.013184	0.073910
liver_lipid_g/FL	log	T-test	-3.23792	0.311872	-3.86187	0.256657	4.781953	0.000173	0.387541
liver_DWT_g/FL	log	T-test	-2.45488	0.145431	-2.71598	0.092860	4.716240	0.000199	0.088136
WM_lipid_%	ASIN	T-test	0.286726	0.159353	0.127592	0.069072	2.878642	0.010424	0.049984
WM_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	2.286190	0.022244	N/A
Mean FL	raw	T-test	251.7778	21.01653	274.9000	13.62555	-2.87615	0.010479	0.181496

Table 17. Results of t-tests used to determine the effects of time [Day 0 (Initial), n=9; Day 192 (Final), n=11] on various condition indices of overwintering YOY bluefish in the Summer-Fed treatment (SU_F).

SU_F (Initial-Final) Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T-Value	p	p Levene
liver_lipid_g	log	T-test	-0.889338	0.220707	-1.52468	0.324081	4.997693	0.000093	0.330146
liver_lipid_%	raw	T-test	0.250725	0.072566	0.096184	0.057135	5.334773	0.000045	0.449902
liver_lipid_g/FL	log	T-test	-3.20261	0.226454	-3.90043	0.314612	5.566877	0.000028	0.439752
liver_DWT_g/FL	raw	T-test	0.002685	0.000634	0.001560	0.000468	4.569993	0.000237	0.805733
WM_lipid_%	ASIN	T-test	0.200762	0.089702	0.118482	0.056888	2.497154	0.022440	0.445322
Mean FL	raw	T-test	206.1111	13.20459	237.7273	9.644593	-6.18913	0.000008	0.542137

Table 18. Results of t-tests used to determine the effects of time [Day 0 (Initial), n=10; Day 192 (Final), n=2] on various condition indices of overwintering YOY bluefish in the Spring-Unfed treatment (SP_U). Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

SP_U (Initial-Final) Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	raw	T-test	0.318770	0.127884	0.005000	0.007071	3.338298	0.007512	N/A
liver_lipid_g	raw	MWU	N/A	N/A	N/A	N/A	2.148345	0.031687	N/A
liver_lipid_%	raw	T-test	0.286204	0.090768	0.028686	0.040568	3.818646	0.003381	N/A
liver_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	2.148345	0.031687	N/A
liver_lipid_g/FL	raw	T-test	0.001292	0.000524	0.000019	0.000026	3.306071	0.007932	N/A
liver_lipid_g/FL	raw	MWU	N/A	N/A	N/A	N/A	2.148345	0.031687	N/A
liver_DWT_g/FL	raw	T-test	0.004386	0.001163	0.000641	0.000014	4.383954	0.001369	N/A
liver_DWT_g/FL	raw	MWU	N/A	N/A	N/A	N/A	2.148345	0.031687	N/A
WM_lipid_%	raw	T-test	0.188869	0.091088	0.008366	0.014706	2.692749	0.022593	N/A
WM_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	2.148345	0.031687	N/A
Mean FL	raw	T-test	246.5000	18.47070	255.5000	17.67767	-0.631707	0.541749	N/A
Mean FL	raw	MWU	N/A	N/A	N/A	N/A	-0.645633	0.518517	N/A

Table 19. Results of two-way ANOVA used to evaluate the effects of cohort of origin (spring-spawned versus summer-spawned) and body depot (liver, viscera, white muscle, red muscle and skin) on ash content of overwintering YOY bluefish subsampled on day 0 of the experiment. Ash content was found to have heterogeneous variance based upon Levene's test and was reanalyzed nonparametrically using Kruskal-Wallis ANOVA to test individually for cohort and feeding effects.

Cohort & Depot Variable	INITIAL Trans.	Test	Effects Cohort H-value	Effects Cohort P-value	Effects Body Depot H-value	Effects Body Depot P-value	Effects Cohort*Depot F-value	Effects Cohort*Depot P-value	Levene's F-value	Levene's P-value
%_ash	raw	2-way ANOVA	2.737	0.102	4.25	0.003	4.551	0.002	8.69088	0.0000
%_ash	raw	Kruskal-Wallis	0.9448396	0.331	16.24539	0.003	N/A	N/A	N/A	N/A

Table 20. Pearson product-limit correlation coefficients for (A) spring cohort ash content, (B) summer cohort ash content across five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in spring- and summer-spawned YOY bluefish subsampled on day 0 of the experiment. All significant ($p<0.05$) coefficients are indicated by an asterisk (*).

A)

spring cohort	liver_%ash_0	viscera_%ash_0	WM_%ash_0	RM_%ash_0	skin_%ash_0
liver_%ash_0	1.00	0.52	0.40	0.10	-0.04
viscera_%ash_0	0.52	1.00	0.29	-0.08	0.22
WM_%ash_0	0.40	0.29	1.00	0.42	0.11
RM_%ash_0	0.10	-0.08	0.42	1.00	-0.02
skin_%ash_0	-0.04	0.22	0.11	-0.02	1.00

B)

summer cohort	liver_%ash_0	viscera_%ash_0	WM_%ash_0	RM_%ash_0	skin_%ash_0
liver_%ash_0	1.00	*-0.69	0.30	-0.08	-0.35
viscera_%ash_0	*-0.69	1.00	-0.31	-0.30	0.28
WM_%ash_0	0.30	-0.31	1.00	-0.02	0.07
RM_%ash_0	-0.08	-0.30	-0.02	1.00	0.51
skin_%ash_0	-0.35	0.28	0.07	0.51	1.00

Table 21. Results of repeated-measures ANOVA used to evaluate changes in ash content of different body depots (liver, viscera, white muscle, red muscle and skin) of YOY bluefish from each treatment [spring-fed (SP-Fed), summer-fed (SU-Fed), spring-unfed (SP-Unfed), summer-unfed (SU-Unfed)]. Subsamples were taken on days 0, 11, 31 and 89 of the experiment.

Wholebody INITIAL-11-31-89 Variable	Trans.	Test	Effects Body Depot F-value	Effects Body Depot P-value	Effects Time F-value	Effects Time P-value	Effects Depot*Time F-value	Effects Depot*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value	89 Levene's P-value
%ash-spring fed	raw	R.M. Anova	13.45	0.000	9.5	0.000	3.89	0.002	0.924	0.015	0.221	N/A
%ash-summer fed	raw	R.M. Anova	14.18	0.000	2.15	0.104	2.63	0.007	0.000	0.015	0.010	0.021966
%ash-spring unfed	raw	R.M. Anova	1.721	0.198	0.103	0.902	2.554	0.030	0.0277	0.0106	0.1592	N/A
%ash-summer unfed	raw	R.M. Anova	2.791	0.054	1.654	0.187	2.945	0.003	0.0023	0.1939	0.0198	0.1597

Table 22. Results of t-tests used to determine the effects of cohort of origin (SP=spring-spawned, n=19; SU=summer-spawned, n=18) on liver and white muscle (WM) ash content of YOY bluefish subsampled on day 0 of the experiment.

Day 0 subsample Variable	Trans.	Test	Mean Spring	Std.Dev. Spring	Mean Summer	Std.Dev. Summer	T-Value	p	p Levene
liver_%ash	raw	T-test	0.054852	0.005184	0.054108	0.004710	0.455774	0.651368	0.606794
WM_%ash	raw	T-test	0.060232	0.004214	0.057968	0.004245	1.627647	0.112570	0.584821

Table 23. Results of two-way ANOVA used to evaluate the effects of cohort of origin (spring-spawned versus summer-spawned) and feeding (fed versus unfed) on liver and white muscle (WM) ash content for overwintering YOY bluefish subsampled on day 11 of the experiment.

Cohort & Feeding	Day 11 subsample		Effects Cohort	Effects Cohort	Effects Feeding	Effects Feeding	Effects Cohort*Feeding	Effects Cohort*Feeding	Levene's	Levene's
Variable	Trans.	Test	F-value	P-value	F-value	P-value	F-value	P-value	F-value	P-value
liver_%ash	raw	2-way Anova	0.9	0.362	167.1	0.000	1.3	0.267	1.245521	0.3095
WM_%ash	raw	2-way Anova	0.309	0.582	3.63	0.066	3.883	0.057	0.397769	0.7555

Table 24. Results of two-way ANOVA used to evaluate the effects of cohort of origin (spring-spawned versus summer-spawned) and feeding (fed versus unfed) on liver and white muscle (WM) ash content for overwintering YOY bluefish subsampled on day 31 of the experiment. Variables found to have heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using Kruskal-Wallis ANOVA to test individually for cohort and feeding effects.

Cohort & Feeding	Day 31 subample		Effects Cohort	Effects Cohort	Effects Feeding	Effects Feeding	Effects Cohort*Feeding	Effects Cohort*Feeding	Levene's	Levene's
Variable	Trans.	Test	F-value	P-value	F-value	P-value	F-value	P-value	F-value	P-value
liver_%ash	raw	2-way Anova	0.2	0.628	221.2	0.000	0.4	0.528	0.121293	0.9469
WM_%ash	raw	2-way Anova	7.13	0.012	10.7	0.003	3.87	0.058	3.394734	0.0296
WM_%ash	raw	Kruskal-Wallis	3.9729	0.046	10.21121	0.001	N/A	N/A	N/A	N/A

Table 25. Results of t-tests used to determine the effects of feeding (Fed, n=9; Unfed, n=9) on liver and white muscle (WM) ash content of summer-spawned YOY bluefish subsampled on day 89 of the experiment.

Summer cohort	Day 89 subsample		Mean	Std.Dev.	Mean	Std.Dev.	T-		p
Variable	Trans.	Test	Fed	Fed	Unfed	Unfed	Value	p	Levene
liver_%ash	raw	T-test	0.053721	0.007805	0.068798	0.008382	-3.94902	0.001149	0.953709
WM_%ash	raw	T-test	0.051687	0.002674	0.067508	0.003601	-10.5816	0.000000	0.433439

Table 26. Results of t-tests used to determine the effects of cohort of origin (spring-spawned, n=10; summer-spawned, n=11) on liver and white muscle (WM) ash content of fed YOY bluefish subsampled on day 192 of the experiment.

Fed treatments	Day 192 subsample		Mean	Std.Dev.	Mean	Std.Dev.	T-		p
Variable	Trans.	Test	Spring	Spring	Summer	Summer	Value	p	Levene
liver_%ash	raw	T-test	0.054236	0.005940	0.056745	0.004473	-1.10011	0.285029	0.313968
WM_%ash	raw	T-test	0.060364	0.003909	0.060544	0.001536	-0.141366	0.889069	0.105376

Table 27. Results of repeated-measures ANOVA used to evaluate changes in liver and white muscle (WM) ash content of fed spring and summer cohort bluefish. Subsamples were taken on days 0, 11 and 31 of the experiment.

Fed INITIAL-11-31 Variable	Trans.	Test	Effects Cohort F-value	Effects Cohort P-value	Effects Time F-value	Effects Time P-value	Effects Cohort*Time F-value	Effects Cohort*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
liver_%ash	raw	R.M. Anova	0.28	0.601	82.710	0.000	1.280	0.292	0.4809	0.1660	0.9200
WM_%ash	raw	R.M. Anova	1.199	0.290	6.301	0.005	1.495	0.240	0.5159	0.5055	0.1390

Table 28. Results of repeated-measures ANOVA used to evaluate changes in liver and white muscle (WM) ash content of unfed spring and summer cohort bluefish. Subsamples were taken on days 0, 11 and 31 of the experiment.

Unfed INITIAL-11-31 Variable	Trans.	Test	Effects Cohort F-value	Effects Cohort P-value	Effects Time F-value	Effects Time P-value	Effects Cohort*Time F-value	Effects Cohort*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
liver_%ash	raw	R.M. Anova	0.12	0.730	18.780	0.000	0.100	0.905	0.6487	0.4925	0.5389
WM_%ash	raw	R.M. Anova	6.645	0.020	0.268	0.767	4.426	0.020	0.8224	0.3917	0.0460

Table 29. Results of repeated-measures ANOVA used to evaluate changes in liver and white muscle (WM) ash content of fed and unfed spring cohort bluefish. Subsamples were taken on days 0, 11 and 31 of the experiment.

Spring INITIAL-11-31 Variable	Trans.	Test	Effects Feeding F-value	Effects Feeding P-value	Effects Time F-value	Effects Time P-value	Effects Cohort*Time F-value	Effects Cohort*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value
liver_%ash	raw	R.M. Anova	138.3	0.000	8.700	0.001	42.300	0.000	0.8900	0.2512	0.7708
WM_%ash	raw	R.M. Anova	0.126	0.728	5.133	0.012	0.483	0.622	0.2760	0.9970	0.1281

Table 30. Results of repeated-measures ANOVA used to evaluate changes in liver and white muscle (WM) ash content of fed and unfed summer cohort bluefish. Subsamples were taken on days 0, 11, 31 and 89 of the experiment.

Summer INITIAL-11-31-89 Variable	Trans.	Test	Effects Feeding F-value	Effects Feeding P-value	Effects Time F-value	Effects Time P-value	Effects Cohort*Time F-value	Effects Cohort*Time P-value	Initial Levene's P-value	11 Levene's P-value	31 Levene's P-value	89 Levene's P-value
liver_%ash	raw	R.M. Anova	93.15	0.000	15.320	0.000	21.120	0.000	0.8499	0.4380	0.6878	0.9537
WM_%ash	raw	R.M. Anova	53.83	0.000	1.200	0.320	11.860	0.000	0.7093	0.9282	0.7991	0.4334

Table 31. Results of t-tests used to determine the effects of time [Day 0 (Initial), n=9; Day 192 (Final), n=10] on liver and white muscle (WM) ash content of overwintering YOY bluefish in the Spring-Fed treatment (SP_F).

SP_F (Initial-Final) Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T-Value	p	p Levene
liver_%ash	raw	T-test	0.056208	0.005659	0.054236	0.005940	0.738762	0.470129	0.576910
WM_%ash	raw	T-test	0.060719	0.004076	0.060364	0.003909	0.193697	0.848709	0.856803

Table 32. Results of t-tests used to determine the effects of time [Day 0 (Initial), n=9; Day 192 (Final), n=11] on liver and white muscle (WM) ash content of overwintering YOY bluefish in the Summer-Fed treatment (SU_F). Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

SU_F (Initial-Final) Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_%ash	raw	T-test	0.056451	0.003689	0.056745	0.004473	-0.157601	0.876526	0.496503
WM_%ash	raw	T-test	0.057050	0.004232	0.060544	0.001536	-2.55272	0.019986	0.014781
WM_%ash	raw	MWU	N/A	N/A	N/A	N/A	-2.08928	0.036684	N/A

Table 33. Results of t-tests used to determine the effects of time [Day 0 (Initial), n=10; Day 192 (Final), n=2] on liver and white muscle (WM) ash content of overwintering YOY bluefish in the Spring-Unfed treatment (SP_U). Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

SP_U (Initial-Final) Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z- Value	p	p Levene
liver_%ash	raw	T-test	0.053631	0.004667	0.060079	0.013321	-1.36207	0.203068	N/A
liver_%ash	raw	MWU	N/A	N/A	N/A	N/A	-0.64450	0.519250	N/A
WM_%ash	raw	T-test	0.059793	0.004505	0.071022	0.012072	-2.52950	0.029896	N/A
WM_%ash	raw	MWU	N/A	N/A	N/A	N/A	-1.50384	0.132623	N/A

Table 34. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=13] on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{high}$/Unfed treatment (HHU) of overwintering YOY bluefish. For mean FL the sample sizes were: initial, n=167; final, n=13. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-HHU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	log	T-Test	-2.74261	0.522360	-1.13754	0.221366	-9.59648	0.000000	0.009669
liver_lipid_g	raw	MWU	N/A	N/A	N/A	N/A	-3.76588	0.000166	N/A
liver_lipid_%	raw	T-Test	0.030109	0.094583	0.568073	0.158416	-8.65214	0.000000	0.217167
liver_lipid_g/FL	log	T-Test	-4.85773	0.509625	-3.29287	0.202278	-9.77225	0.000000	0.006088
liver_lipid_g/FL	raw	MWU	N/A	N/A	N/A	N/A	-3.76588	0.000166	N/A
liver_DWT_g/FL	raw	T-Test	0.000314	0.000157	0.000971	0.000269	-6.25525	0.000005	0.094383
viscera_lipid_g	log	T-Test	-2.17542	0.435863	-1.49741	0.347912	-3.94291	0.000873	0.956706
viscera_lipid_%	asin	T-Test	0.218214	0.073618	0.392319	0.111221	-3.91199	0.000937	0.257513
viscera_lipid_g/FL	log	T-Test	-4.28512	0.409485	-3.65274	0.332602	-3.87868	0.001011	0.934333
viscera_DWT_g/FL	raw	T-Test	0.001316	0.000448	0.001701	0.000438	-1.93765	0.067674	0.761240
WM_lipid_g	log	T-Test	-1.306370	0.374316	-0.373236	0.347428	-5.80750	0.000014	0.974155
WM_lipid_%	asin	T-Test	0.148830	0.047342	0.359385	0.084538	-6.41250	0.000004	0.194333
WM_lipid_g/FL	log	T-Test	-3.416060	0.343639	-2.528560	0.328390	-5.91167	0.000011	0.946393
WM_DWT_g/FL	raw	T-Test	0.019742	0.006701	0.026481	0.007910	-2.00281	0.059675	0.473980
RM_lipid_g	log	T-Test	-2.24706	0.377203	-1.15999	0.301303	-7.30210	0.000001	0.613701
RM_lipid_%	raw	T-Test	0.085309	0.048181	0.358731	0.094340	-7.56092	0.000000	0.257132
RM_lipid_g/FL	log	T-Test	-4.35675	0.349635	-3.31532	0.281082	-7.52177	0.000000	0.545884
RM_DWT_g/FL	log	T-Test	-3.22037	0.149883	-2.85398	0.167580	-5.05546	0.000070	0.330680
skin_lipid_g	raw	T-Test	0.016013	0.016810	0.112731	0.107370	-2.50457	0.021532	0.102755
skin_lipid_%	asin	T-Test	0.254875	0.068151	0.543100	0.127888	-5.84535	0.000012	0.101760
skin_lipid_g/FL	raw	T-Test	0.000116	0.000109	0.000759	0.000651	-2.74171	0.012966	0.089513
skin_DWT_g/FL	raw	T-Test	0.001491	0.000634	0.002556	0.001048	-2.58075	0.018322	0.272512
Mean FL	raw	T-Test	140.8922	13.67613	143.3846	11.13207	-0.64024	0.522838	0.410019

Table 35. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=8] on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{low}$/Unfed treatment (HLU) of overwintering YOY bluefish. For skin lipid content (g) and skin DWT (g)/FL (mm) the sample sizes were: initial, n=8; final, n=7. For mean FL the sample sizes were: initial, n=167; final, n=8. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-HLU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	log	T-Test	-2.74261	0.522360	-1.53466	0.496136	-4.40963	0.000851	0.789014
liver_lipid_%	ASIN	T-Test	0.245805	0.129386	0.690089	0.236015	-4.14086	0.001369	0.029087
liver_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	-3.15063	0.001629	N/A
liver_lipid_g/FL	log	T-Test	-4.85773	0.509625	-3.70807	0.480661	-4.31853	0.000999	0.801361
liver_DWT_g/FL	raw	T-Test	0.000314	0.000157	0.000626	0.000309	-2.54835	0.023192	0.081855
viscera_lipid_g	raw	T-Test	0.010288	0.012032	0.012075	0.003853	-0.400176	0.695067	0.238092
viscera_lipid_%	raw	T-Test	0.051172	0.030121	0.055277	0.017944	-0.331186	0.745409	0.427808
viscera_lipid_g/Fl	raw	T-Test	0.000074	0.000074	0.000081	0.000027	-0.249648	0.806486	0.308586
viscera_DWT_g/l	raw	T-Test	0.001316	0.000448	0.001487	0.000324	-0.876283	0.395664	0.893647
WM_lipid_g	raw	T-Test	0.075662	0.098612	0.104682	0.096341	-0.595392	0.561084	0.742650
WM_lipid_%	raw	T-Test	0.023845	0.015605	0.033599	0.030622	-0.802765	0.435527	0.140032
WM_lipid_g/FL	raw	T-Test	0.000539	0.000606	0.000701	0.000654	-0.511484	0.616984	0.566958
WM_DWT_g/FL	raw	T-Test	0.019742	0.006701	0.020928	0.006801	-0.351251	0.730630	0.708922
RM_lipid_g	raw	T-Test	0.008450	0.010089	0.017750	0.015157	-1.44467	0.170560	0.137010
RM_lipid_%	asin	T-Test	0.286234	0.087578	0.358077	0.125360	-1.32880	0.205167	0.160850
RM_lipid_g/FL	raw	T-Test	0.000061	0.000062	0.000115	0.000093	-1.38813	0.186793	0.095541
RM_DWT_g/FL	raw	T-Test	0.000638	0.000265	0.000784	0.000329	-0.976054	0.345602	0.644786
skin_lipid_g	raw	T-Test	0.016013	0.016810	0.028729	0.021033	-1.30158	0.215655	0.375442
skin_lipid_%	raw	T-Test	0.067026	0.036611	0.089225	0.052324	-0.962650	0.353289	0.096895
skin_lipid_g/FL	raw	T-Test	0.000116	0.000109	0.000186	0.000127	-1.14284	0.273714	0.490563
skin_DWT_g/FL	raw	T-Test	0.001491	0.000634	0.001983	0.000491	-1.73182	0.105270	0.242050
Mean FL	raw	T-Test	140.8922	13.67613	149.6250	13.87637	-1.76325	0.079623	0.784070

Table 36. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=8] on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{low}$/Fed treatment (HLF) of overwintering YOY bluefish. For mean FL the sample sizes were: initial, n=167; final, n=8. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-HLF Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	log	T-Test	-2.74261	0.522360	-1.06755	0.256260	-7.95550	0.000004	0.054294
liver_lipid_%	raw	T-Test	0.030109	0.094583	0.416050	0.053332	-10.0532	0.000000	0.248889
liver_lipid_g/FL	log	T-Test	-4.85773	0.509625	-3.22951	0.227942	-8.10040	0.000003	0.035979
liver_lipid_g/FL	raw	MWU	N/A	N/A	N/A	N/A	-3.36067	0.000778	N/A
liver_DWT_g/FL	raw	T-Test	0.000314	0.000157	0.001577	0.000753	-4.64284	0.000380	0.091702
viscera_lipid_g	raw	T-Test	0.010288	0.012032	0.039438	0.016142	-4.09525	0.001092	0.097948
viscera_lipid_%	raw	T-Test	0.051172	0.030121	0.111729	0.044491	-3.18791	0.006577	0.180775
viscera_lipid_g/FL	raw	T-Test	0.000074	0.000074	0.000270	0.000107	-4.27219	0.000774	0.071751
viscera_DWT_g/FL	raw	T-Test	0.001316	0.000448	0.002467	0.000383	-5.52407	0.000075	0.973068
WM_lipid_g	log	T-Test	-1.30637	0.374316	-0.55173	0.334084	-4.25420	0.000802	0.856854
WM_lipid_%	asin	T-Test	0.148830	0.047342	0.282938	0.088103	-3.79251	0.001980	0.110908
WM_lipid_g/FL	log	T-Test	-3.41606	0.343639	-2.71370	0.328496	-4.17882	0.000928	0.771691
WM_DWT_g/FL	raw	T-Test	0.019742	0.006701	0.027882	0.006541	-2.45869	0.027579	0.984631
RM_lipid_g	log	T-Test	-2.24706	0.377203	-1.23956	0.339924	-5.61205	0.000064	0.973415
RM_lipid_%	asin	T-Test	0.286234	0.087578	0.587828	0.133968	-5.32966	0.000106	0.189223
RM_lipid_g/FL	log	T-Test	-4.35675	0.349635	-3.40152	0.327324	-5.64116	0.000061	0.982168
RM_DWT_g/FL	log	T-Test	-3.22037	0.149883	-2.86677	0.177822	-4.30056	0.000733	0.524786
skin_lipid_g	raw	T-Test	0.016013	0.016810	0.138600	0.136583	-2.51960	0.024520	0.092202
skin_lipid_%	asin	T-Test	0.254875	0.068151	0.574764	0.113491	-6.83464	0.000008	0.160297
skin_lipid_g/FL	raw	T-Test	0.000116	0.000109	0.000924	0.000876	-2.58995	0.021391	0.084468
skin_DWT_g/FL	raw	T-Test	0.001491	0.000634	0.002691	0.001465	-2.12597	0.051780	0.413058
Mean FL	raw	T-Test	140.8922	13.67613	145.6250	11.80723	-0.96114	0.337823	0.451517

Table 37. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=14] on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{high}$/Unfed treatment (LHU) of overwintering YOY bluefish. For mean FL the sample sizes were: initial, n=167; final, n=14. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-LHU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	log	T-Test	-2.74261	0.522360	-1.06783	0.213879	-10.4041	0.000000	0.006434
liver_lipid_g	raw	MWU	N/A	N/A	N/A	N/A	-3.82213	0.000132	N/A
liver_lipid_%	raw	T-Test	0.030109	0.094583	0.591653	0.072694	-15.6363	0.000000	0.551931
liver_lipid_g/FL	raw	T-Test	0.000011	0.000026	0.000657	0.000313	-5.76778	0.000012	0.009674
liver_lipid_g/FL	raw	MWU	N/A	N/A	N/A	N/A	-3.82213	0.000132	N/A
liver_DWT_g/FL	log	T-Test	-3.54008	0.182362	-2.99243	0.155263	-7.47737	0.000000	0.983302
viscera_lipid_g	log	T-Test	-2.17542	0.435863	-1.47344	0.316440	-4.36648	0.000299	0.966259
viscera_lipid_%	raw	T-Test	0.051172	0.030121	0.155291	0.074125	-3.76716	0.001212	0.050943
viscera_lipid_g/FL	log	T-Test	-4.28512	0.409485	-3.62914	0.303345	-4.29960	0.000349	0.918782
viscera_DWT_g/FL	raw	T-Test	0.001316	0.000448	0.001732	0.000498	-1.95354	0.064887	0.271784
WM_lipid_g	log	T-Test	-1.30637	0.374316	-0.52591	0.379454	-4.66273	0.000150	0.368085
WM_lipid_%	sqrt	T-Test	0.281382	0.083720	0.540870	0.110949	-5.72616	0.000013	0.579259
WM_lipid_g/FL	log	T-Test	-3.41606	0.343639	-2.68161	0.361623	-4.66231	0.000150	0.334907
WM_DWT_g/FL	raw	T-Test	0.019742	0.006701	0.024607	0.007302	-1.54632	0.137706	0.212055
RM_lipid_g	log	T-Test	-2.24706	0.377203	-1.30136	0.369277	-5.73490	0.000013	0.654464
RM_lipid_%	asin	T-Test	0.286234	0.087578	0.576541	0.131300	-5.55775	0.000019	0.348197
RM_lipid_g/FL	log	T-Test	-4.35675	0.349635	-3.45706	0.349758	-5.80465	0.000011	0.695608
RM_DWT_g/FL	log	T-Test	-3.22037	0.149883	-2.90414	0.197173	-3.91986	0.000848	0.176725
skin_lipid_g	log	T-Test	-1.97458	0.398774	-0.998483	0.460332	-5.00801	0.000067	0.593573
skin_lipid_%	sqrt	T-Test	0.251574	0.065347	0.525392	0.138127	-5.24102	0.000040	0.067540
skin_lipid_g/FL	log	T-Test	-4.08427	0.366344	-3.15418	0.445517	-5.00239	0.000068	0.528012
skin_DWT_g/FL	raw	T-Test	0.001491	0.000634	0.003123	0.001924	-2.30671	0.031901	0.054303
Mean FL	raw	T-Test	140.8922	13.67613	143.5714	11.94677	-0.71023	0.478489	0.550429

Table 38. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=12] on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{high}$/Fed treatment (LHF) of overwintering YOY bluefish. For mean FL the sample sizes were: initial, n=167; final, n=12. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-LHF Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	log	T-Test	-2.74261	0.522360	-1.04480	0.266489	-9.27291	0.000000	0.038254
liver_lipid_g	raw	MWU	N/A	N/A	N/A	N/A	-3.70328	0.000213	N/A
liver_lipid_%	raw	T-Test	0.030109	0.094583	0.580580	0.096309	-12.6098	0.000000	0.768255
liver_lipid_g/FL	log	T-Test	-4.85773	0.509625	-3.18406	0.237499	-9.66535	0.000000	0.021244
liver_lipid_g/FL	raw	MWU	N/A	N/A	N/A	N/A	-3.70328	0.000213	N/A
liver_DWT_g/FL	log	T-Test	-3.54008	0.182362	-2.94263	0.186242	-7.08518	0.000001	0.510013
viscera_lipid_g	log	T-Test	-2.17542	0.435863	-1.37320	0.307862	-4.84125	0.000131	0.812291
viscera_lipid_%	raw	T-Test	0.051172	0.030121	0.161857	0.064692	-4.49508	0.000280	0.053048
viscera_lipid_g/FL	log	T-Test	-4.28512	0.409485	-3.51245	0.283394	-5.00738	0.000091	0.722915
viscera_DWT_g/FL	raw	T-Test	0.001316	0.000448	0.002112	0.000496	-3.64696	0.001844	0.622718
WM_lipid_g	log	T-Test	1.30637	0.374316	-0.37744	0.330474	-5.84518	0.000015	0.839251
WM_lipid_%	asin	T-Test	0.148830	0.047342	0.360560	0.079047	-6.77348	0.000002	0.078068
WM_lipid_g/FL	log	T-Test	-3.41606	0.343639	-2.51669	0.304714	-6.14958	0.000008	0.912071
WM_DWT_g/FL	raw	T-Test	0.019742	0.006701	0.026816	0.008371	-1.99604	0.061286	0.277353
RM_lipid_g	log	T-Test	-2.24706	0.377203	-1.20211	0.364939	-6.19158	0.000008	0.658208
RM_lipid_%	raw	T-Test	0.085309	0.048181	0.358991	0.092261	-7.67425	0.000000	0.055777
RM_lipid_g/FL	log	T-Test	-4.35675	0.349635	-3.34136	0.337807	-6.49608	0.000004	0.768034
RM_DWT_g/FL	log	T-Test	-3.22037	0.149883	-2.88131	0.222883	-3.75701	0.001443	0.021772
RM_DWT_g/FL	raw	MWU	N/A	N/A	N/A	N/A	-3.08607	0.002028	N/A
skin_lipid_g	log	T-Test	-1.97458	0.398774	-1.03810	0.399561	-5.13884	0.000069	0.930679
skin_lipid_%	asin	T-Test	0.254875	0.068151	0.597893	0.146308	-6.15919	0.000008	0.089451
skin_lipid_g/FL	log	T-Test	-4.08427	0.366344	-3.17735	0.391657	-5.20126	0.000060	0.915993
skin_DWT_g/FL	raw	T-Test	0.001491	0.000634	0.002548	0.001561	-1.80461	0.087895	0.200802
Mean FL	raw	T-Test	140.8922	13.67613	138.3333	12.79441	0.62849	0.530492	0.666682

Table 39. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=6] on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{low}$/Unfed treatment (LLU) of overwintering YOY bluefish. For mean FL the sample sizes were: initial, n=167; final, n=6. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-LLU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_lipid_g	log	T-Test	-2.74261	0.522360	-1.20452	0.099279	-7.08570	0.000034	0.008101
liver_lipid_g	raw	MWU	N/A	N/A	N/A	N/A	-3.09839	0.001946	N/A
liver_lipid_%	raw	T-Test	0.030109	0.094583	0.514453	0.061342	-10.8867	0.000000	0.484084
liver_lipid_g/FL	raw	T-Test	0.000011	0.000026	0.000417	0.000083	-13.2066	0.000000	0.034091
liver_lipid_g/FL	raw	MWU	N/A	N/A	N/A	N/A	-3.09839	0.001946	N/A
liver_DWT_g/FL	raw	T-Test	0.000314	0.000157	0.000811	0.000125	-6.37273	0.000035	0.959600
viscera_lipid_g	raw	T-Test	0.010288	0.012032	0.014367	0.006348	-0.750681	0.467310	0.504145
viscera_lipid_%	raw	T-Test	0.051172	0.030121	0.055552	0.011857	-0.334547	0.743742	0.178741
viscera_lipid_g/FL	raw	T-Test	0.000074	0.000074	0.000092	0.000035	-0.562631	0.584044	0.454598
viscera_DWT_g/FL	raw	T-Test	0.001316	0.000448	0.001618	0.000336	-1.38014	0.192712	0.821612
WM_lipid_g	raw	T-Test	0.075662	0.098612	0.088395	0.052045	-2.858840	0.779842	0.599183
WM_lipid_%	raw	T-Test	0.023845	0.015605	0.025369	0.011848	-0.199263	0.845393	0.647125
WM_lipid_g/FL	raw	T-Test	0.000539	0.000606	0.000570	0.000321	-0.111738	0.912878	0.585744
WM_DWT_g/FL	raw	T-Test	0.019742	0.006701	0.021676	0.005052	-0.589835	0.566242	0.654434
RM_lipid_g	raw	T-Test	0.008450	0.010089	0.020050	0.012975	-1.88728	0.083542	0.180126
RM_lipid_%	raw	T-Test	0.085309	0.048181	0.140529	0.073066	-1.70923	0.113119	0.470540
RM_lipid_g/FL	raw	T-Test	0.000061	0.000062	0.000128	0.000081	-1.77554	0.101151	0.213716
RM_DWT_g/FL	raw	T-Test	0.000638	0.000265	0.000817	0.000258	-1.25842	0.232173	0.777440
skin_lipid_g	raw	T-Test	0.016013	0.016810	0.038100	0.024063	-2.02948	0.065188	0.365820
skin_lipid_%	raw	T-Test	0.067026	0.036611	0.123866	0.068815	-2.00515	0.068037	0.218768
skin_lipid_g/FL	raw	T-Test	0.000116	0.000109	0.000248	0.000160	-1.84161	0.090372	0.318035
skin_DWT_g/FL	raw	T-Test	0.001491	0.000634	0.001866	0.000434	-1.23998	0.238684	0.119881
Mean FL	raw	T-Test	140.8922	13.67613	152.3333	10.61446	-2.02514	0.044408	0.447736

Table 40. Results of two-way ANOVA used to evaluate the effects of activity level (high versus low) and pre-winter lipid storage (high versus low) on various body condition indices of different body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] of unfed YOY bluefish subsampled on January 19, 2003. Variables found to have heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using Kruskal-Wallis ANOVA to test individually for cohort and feeding effects.

Activ.&Storage Variable	Trans.	Test	Effects Activity F-value	Effects Activity P-value	Effects Storage F-value	Effects Storage P-value	Effects Activ*Storage F-value	Effects Activ*Storage P-value	Levene's F-value	Levene's P-value
liver_lipid_g	raw	2_way Anova	1.558	0.220	6.624	0.014	0.030	0.864	2.271	0.0963
liver_lipid_%	raw	2_way Anova	1.777	0.191	6.163	0.018	0.685	0.413	7.711	0.0004
liver_lipid_%	raw	Kruskal-Wallis	0.496	0.481	5.333	0.021	N/A	N/A	N/A	N/A
liver_lipid_g/FL	raw	2_way Anova	1.649	0.207	8.767	0.005	0.017	0.896	1.804	0.1633
liver_DWT_g/FL	raw	2_way Anova	1.929	0.173	8.353	0.006	0.121	0.730	2.046	0.1241
viscera_lipid_g	log	2_way Anova	0.190	0.666	19.380	0.000	0.030	0.853	1.738	0.1760
viscera_lipid_%	ASIN	2_way Anova	0.010	0.913	29.170	0.000	0.000	0.971	3.495	0.0250
viscera_lipid_%	raw	Kruskal-Wallis	0.360	0.549	21.844	0.000	N/A	N/A	N/A	N/A
viscera_lipid_g/FL	log	2_way Anova	0.170	0.686	23.650	0.000	0.020	0.880	1.604	0.2047
viscera_DWT_g/FL	raw	2_way Anova	0.325	0.572	1.324	0.257	0.121	0.730	1.177	0.3314
WM_lipid_g	log	2_way Anova	0.320	0.578	32.380	0.000	0.510	0.479	0.988	0.4089
WM_lipid_%	asin	2_way Anova	1.150	0.291	41.550	0.000	0.320	0.578	1.876	0.1504
WM_lipid_g/FL	log	2_way Anova	0.390	0.535	37.710	0.000	0.510	0.481	1.127	0.3507
WM_DWT_g/FL	raw	2_way Anova	0.056	0.814	3.193	0.082	0.305	0.584	0.989	0.4082
RM_lipid_g	log	2_way Anova	0.100	0.752	25.660	0.000	0.550	0.463	0.966	0.4186
RM_lipid_%	raw	2_way Anova	0.540	0.466	36.370	0.000	0.920	0.343	0.811	0.4955
RM_lipid_g/FL	log	2_way Anova	0.140	0.714	30.160	0.000	0.560	0.460	1.114	0.3557
RM_DWT_g/FL	log	2_way Anova	0.020	0.876	16.490	0.000	0.450	0.508	0.373	0.7728
skin_lipid_g	log	2_way Anova	0.610	0.439	18.250	0.000	0.100	0.753	0.985	0.4105
skin_lipid_%	asin	2_way Anova	0.660	0.422	25.160	0.000	0.170	0.678	0.968	0.4183
skin_lipid_g/FL	log	2_way Anova	0.620	0.436	21.550	0.000	0.090	0.764	1.065	0.3761
skin_DWT_g/FL	log	2_way Anova	0.071	0.791	5.610	0.023	0.465	0.499	2.187	0.1058
Mean FL	raw	2_way Anova	0.134	0.716	3.602	0.066	0.102	0.752	0.451	0.7174

Table 41. Results of t-tests used to determine the effects of winter-feeding (unfed, n=8; fed, n=8) on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{low}$ treatments (HLU and HLF) of overwintering YOY bluefish subsampled on January 19, 2003. For skin lipid content (g) and skin DWT (g)/FL (mm) the sample sizes were: unfed, n=7; fed, n=8. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

Feeding-HL Variable	Trans.	Test	Mean Unfed	Std.Dev. Unfed	Mean Fed	Std.Dev. Fed	T- or Z-Value	p	p Levene
liver_lipid_g	raw	T-Test	0.044825	0.036674	0.096938	0.044146	-2.56824	0.022314	0.637099
liver_lipid_%	ASIN	T-Test	0.690089	0.236015	0.700601	0.054292	-0.122773	0.904032	0.000155
liver_lipid_%	raw	MWU	N/A	N/A	N/A	N/A	0.00	1.000000	N/A
liver_lipid_g/FL	raw	T-Test	0.000297	0.000248	0.000653	0.000275	-2.70972	0.016931	0.768611
liver_DWT_g/FL	raw	T-Test	0.000626	0.000309	0.001577	0.000753	-3.30277	0.005233	0.281959
viscera_lipid_g	log	T-Test	-1.94143	0.159458	-1.43912	0.190743	-5.71471	0.000053	0.379225
viscera_lipid_%	asin	T-Test	0.234707	0.039176	0.334637	0.074608	-3.35415	0.004725	0.067274
viscera_lipid_g/FL	log	T-Test	-4.11484	0.155018	-3.60108	0.183216	-6.05479	0.000030	0.473727
viscera_DWT_g/FL	raw	T-Test	0.001487	0.000324	0.002467	0.000383	-5.52702	0.000075	0.812141
WM_lipid_g	log	T-Test	-1.139250	0.401651	-0.551734	0.334084	-3.18078	0.006671	0.409308
WM_lipid_%	raw	T-Test	0.033599	0.030622	0.083581	0.049089	-2.44347	0.028398	0.181591
WM_lipid_g/FL	log	T-Test	-3.312650	0.396651	-2.713700	0.328496	-3.28943	0.005374	0.449164
WM_DWT_g/FL	raw	T-Test	0.020928	0.006801	0.027882	0.006541	-2.08457	0.055905	0.713120
RM_lipid_g	log	T-Test	-1.93365	0.466040	-1.23956	0.339924	-3.40336	0.004284	0.271856
RM_lipid_%	raw	T-Test	0.133281	0.079308	0.313384	0.122360	-3.49354	0.003581	0.145202
RM_lipid_g/FL	log	T-Test	-4.10706	0.449059	-3.40152	0.327324	-3.59111	0.002950	0.247037
RM_DWT_g/FL	raw	T-Test	0.000784	0.000329	0.001459	0.000570	-2.89908	0.011663	0.084192
skin_lipid_g	raw	T-Test	0.028729	0.021033	0.138600	0.136483	-2.09696	0.056121	0.138786
skin_lipid_%	raw	T-Test	0.089225	0.052324	0.299789	0.103968	-4.83382	0.000326	0.127918
skin_lipid_g/FL	raw	T-Test	0.000186	0.000127	0.000924	0.000876	-2.20052	0.046453	0.122609
skin_DWT_g/FL	raw	T-Test	0.001983	0.000491	0.002691	0.001465	-1.29692	0.215628	0.243566
Mean FL	raw	T-Test	149.6250	13.87637	145.6250	11.80723	0.620954	0.544608	0.373551

Table 42. Results of t-tests used to determine the effects of winter-feeding (unfed, n=14; fed, n=12) on various condition indices in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{high}$ treatments (LHU and LHF) of overwintering YOY bluefish subsampled on January 19, 2003. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

Feeding-LH Variable	Trans.	Test	Mean Unfed	Std.Dev. Unfed	Mean Fed	Std.Dev. Fed	T-Value	p	p Levene
liver_lipid_g	raw	T-Test	0.095900	0.049458	0.105792	0.058036	-0.469454	0.642982	0.300658
liver_lipid_%	raw	T-Test	0.591653	0.072694	0.580580	0.096309	0.333714	0.741493	0.282416
liver_lipid_g/FL	raw	T-Test	0.000657	0.000313	0.000743	0.000363	-0.648807	0.522624	0.271049
liver_DWT_g/FL	raw	T-Test	0.001082	0.000411	0.001236	0.000487	-0.872646	0.391503	0.308605
viscera_lipid_g	raw	T-Test	0.043343	0.033568	0.052658	0.036020	-0.682137	0.501683	0.979012
viscera_lipid_%	raw	T-Test	0.155291	0.074125	0.161857	0.064692	-0.238555	0.813475	0.735720
viscera_lipid_g/FL	raw	T-Test	0.000297	0.000223	0.000368	0.000221	-0.814075	0.423606	0.921100
viscera_DWT_g/FL	raw	T-Test	0.001732	0.000498	0.002112	0.000496	-1.93962	0.064267	0.581870
WM_lipid_g	raw	T-Test	0.417493	0.335194	0.541931	0.400309	-0.862430	0.396987	0.594178
WM_lipid_%	raw	T-Test	0.102570	0.054519	0.128795	0.050990	-1.25946	0.219977	0.790985
WM_lipid_g/FL	raw	T-Test	0.002813	0.002099	0.003766	0.002482	-1.061820	0.298885	0.622264
WM_DWT_g/FL	raw	T-Test	0.024607	0.007302	0.026816	0.008371	-0.719051	0.479053	0.823097
RM_lipid_g	raw	T-Test	0.068086	0.051155	0.085733	0.068487	-0.751081	0.459906	0.145261
RM_lipid_%	raw	T-Test	0.303971	0.115074	0.358991	0.092261	-1.32901	0.196342	0.497082
RM_lipid_g/FL	raw	T-Test	0.000461	0.000331	0.000592	0.000429	-0.880247	0.387455	0.132410
RM_DWT_g/FL	raw	T-Test	0.001370	0.000614	0.001486	0.000783	-0.424716	0.674827	0.141559
skin_lipid_g	raw	T-Test	0.173957	0.205198	0.139500	0.153015	0.478271	0.636786	0.346486
skin_lipid_%	raw	T-Test	0.293753	0.148917	0.323578	0.133791	-0.533214	0.598789	0.585399
skin_lipid_g/FL	raw	T-Test	0.001161	0.001288	0.000984	0.001013	0.383592	0.704658	0.412502
skin_DWT_g/FL	raw	T-Test	0.003123	0.001924	0.002548	0.001561	0.827471	0.416122	0.423820
Mean FL	raw	T-Test	143.5714	11.94677	138.3333	12.79441	1.078793	0.291403	0.916214

Table 43. Results of two-way ANOVA used to evaluate the effects of activity level (high versus low) and pre-winter lipid storage (high versus low) on ash content of different body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] of unfed YOY bluefish subsampled on January 19, 2003.

Activ.&Storage Variable	Trans.	Test	Effects Activity F-value	Effects Activity P-value	Effects Storage F-value	Effects Storage P-value	Effects Activ*Storage F-value	Effects Activ*Storage P-value	Levene's F-value	Levene's P-value
liver_%_ash	raw	2_way Anova	0.168	0.684	1.834	0.184	0.983	0.328	0.3886	0.7618
viscera_%_ash	raw	2_way Anova	1.19	0.282	0.134	0.717	0.702	0.407	1.13	0.3494
WM_%_ash	raw	2_way Anova	0.865	0.358	1.111	0.299	0.141	0.709	2.3421	0.0889
RM_%_ash	raw	2_way Anova	1.601	0.214	5.549	0.024	0.008	0.927	2.3394	0.0892
skin_%_ash	raw	2_way Anova	0.861	0.359	0.371	0.546	0.482	0.492	2.6219	0.065

Table 44. Results of t-tests used to determine the effects of winter-feeding (unfed, n=8; fed, n=8) on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{low}$ treatments (HLU and HLF) of overwintering YOY bluefish subsampled on January 19, 2003. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

Feeding-HL Variable	Trans.	Test	Mean Unfed	Std.Dev. Unfed	Mean Fed	Std.Dev. Fed	T- or Z-Value	p	p Levene
liver_%_ash	raw	T-test	10.24093	2.872997	7.115945	1.237231	2.825629	0.013482	0.106947
viscera_%_ash	raw	T-test	8.555907	1.305694	7.620160	0.834629	1.707918	0.109720	0.179851
WM_%_ash	raw	T-test	6.470630	0.620467	5.592172	0.813201	2.429087	0.029194	0.369625
RM_%_ash	raw	T-test	7.145278	1.283518	6.015774	0.546206	2.290279	0.038045	0.110798
skin_%_ash	log	t-test	0.762036	0.064073	0.761677	0.036206	0.013776	0.989203	0.040477
skin_%_ash	raw	MWU	N/A	N/A	N/A	N/A	-0.105021	0.916359	N/A

Table 45. Results of t-tests used to determine the effects of winter-feeding (unfed, n=14; fed, n=12) on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{high}$ treatments (LHU and LHF) of overwintering YOY bluefish subsampled on January 19, 2003. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

Feeding-LH Variable	Trans.	Test	Mean Unfed	Std.Dev. Unfed	Mean Fed	Std.Dev. Fed	T- or Z-Value	p	p Levene
liver_%_ash	raw	T-test	8.800130	2.674892	7.631300	1.187682	1.397158	0.175151	0.152262
viscera_%_ash	raw	T-test	8.104053	1.988413	7.311289	0.926261	1.265711	0.217766	0.115805
WM_%_ash	raw	T-test	5.894541	0.680329	5.633355	1.219921	0.687421	0.498407	0.179577
RM_%_ash	log	T-test	0.761081	0.035332	0.791895	0.062214	-1.58240	0.126649	0.059314
skin_%_ash	log	t-test	0.732792	0.038121	0.764198	0.075696	-1.36644	0.184467	0.038255
skin_%_ash	raw	MWU	N/A	N/A	N/A	N/A	-1.13156	0.257821	N/A

Table 46. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=13] on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{high}$/Unfed treatment (HHU) of overwintering YOY bluefish. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-HHU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_%_ash	log	t-test	1.086110	0.199161	0.990955	0.078938	1.554814	0.136490	0.000166
liver_%_ash	raw	MWU	N/A	N/A	N/A	N/A	0.724207	0.468939	N/A
viscera_%_ash	raw	T-test	9.640022	2.649778	9.305838	2.200482	0.313012	0.757682	0.500570
WM_%_ash	raw	t-test	6.717920	0.468326	6.055413	1.214307	1.465506	0.159137	0.039283
WM_%_ash	raw	MWU	N/A	N/A	N/A	N/A	1.810517	0.070217	N/A
RM_%_ash	log	t-test	0.945106	0.177493	0.788159	0.077078	2.818244	0.010978	0.000250
RM_%_ash	raw	MWU	N/A	N/A	N/A	N/A	1.882938	0.059710	N/A
skin_%_ash	raw	T-test	7.315920	1.557885	5.860143	1.042363	2.577037	0.018467	0.289716

Table 47. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=8] on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{low}$/Unfed treatment (HLU) of overwintering YOY bluefish. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-HLU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_%_ash	log	t-test	1.086110	0.199161	0.997554	0.108373	1.104689	0.287919	0.011892
liver_%_ash	raw	MWU	N/A	N/A	N/A	N/A	0.630126	0.528613	N/A
viscera_%_ash	log	T-test	0.970430	0.114896	0.927884	0.065840	0.908739	0.378866	0.086453
WM_%_ash	raw	T-test	6.717920	0.468326	6.470630	0.620467	0.899754	0.383467	0.390257
RM_%_ash	log	t-test	0.945106	0.177493	0.848528	0.071887	1.426447	0.175658	0.000647
RM_%_ash	raw	MWU	N/A	N/A	N/A	N/A	0.735147	0.462250	N/A
skin_%_ash	raw	T-test	7.315920	1.557885	5.837377	0.875238	2.340326	0.034595	0.238664

Table 48. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=8] on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{high}/Stor._{low}$/Fed treatment (HLF) of overwintering YOY bluefish. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-HLF Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_%_ash	log	t-test	1.086110	0.199161	0.846528	0.075121	3.183533	0.006634	0.000670
liver_%_ash	raw	MWU	N/A	N/A	N/A	N/A	2.625525	0.008652	N/A
viscera_%_ash	log	t-test	0.970430	0.114896	0.879792	0.045882	2.072145	0.057201	0.014391
viscera_%_ash	raw	MWU	N/A	N/A	N/A	N/A	1.890378	0.058708	N/A
WM_%_ash	raw	T-test	6.717920	0.468326	5.592172	0.813201	3.393057	0.004373	0.097606
RM_%_ash	log	t-test	0.945106	0.177493	0.777813	0.037794	2.607429	0.020676	0.000015
RM_%_ash	raw	MWU	N/A	N/A	N/A	N/A	2.520504	0.011719	N/A
skin_%_ash	log	T-test	0.856333	0.087091	0.761677	0.036206	2.838576	0.013143	0.070934

Table 49. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=14] on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{high}$/Unfed treatment (LHU) of overwintering YOY bluefish. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-LHU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_%_ash	log	T-test	1.086110	0.199161	0.916044	0.187138	2.004474	0.058748	0.214972
viscera_%_ash	raw	T-test	9.640022	2.649778	8.104053	1.988413	1.545636	0.137870	0.178272
WM_%_ash	raw	T-test	6.717920	0.468326	5.894541	0.680329	3.023241	0.006715	0.551853
RM_%_ash	raw	t-test	9.503441	4.032876	5.787164	0.497805	3.465752	0.002441	0.000000
RM_%_ash	raw	MWU	N/A	N/A	N/A	N/A	3.276113	0.001053	N/A
skin_%_ash	log	t-test	0.856333	0.087091	0.732792	0.038121	4.646216	0.000156	0.026773
skin_%_ash	raw	MWU	N/A	N/A	N/A	N/A	3.344366	0.000825	N/A

Table 50. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=12] on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{high}$/Fed treatment (LHF) of overwintering YOY bluefish. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-LHF Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_%_ash	log	t-test	1.086110	0.199161	0.877732	0.068139	3.378232	0.003349	0.000053
liver_%_ash	raw	MWU	N/A	N/A	N/A	N/A	2.854612	0.004309	N/A
viscera_%_ash	log	t-test	0.970430	0.114896	0.861041	0.051947	2.909943	0.009343	0.009228
viscera_%_ash	raw	MWU	N/A	N/A	N/A	N/A	2.468854	0.013555	N/A
WM_%_ash	raw	T-test	6.717920	0.468326	5.633355	1.219921	2.382421	0.028433	0.146052
RM_%_ash	log	t-test	0.945106	0.177493	0.791895	0.062214	2.776392	0.012450	0.000024
RM_%_ash	raw	MWU	N/A	N/A	N/A	N/A	2.005944	0.044863	N/A
skin_%_ash	raw	T-test	7.315920	1.557885	5.893683	1.068390	2.432128	0.025673	0.309849

Table 51. Results of t-tests used to determine the effects of time [Oct. 3, 2002 (Initial), n=8; Jan. 19, 2003 (Final), n=6] on ash content in different body depots (liver, viscera, white muscle (WM), red muscle (RM) and skin) in the $Act._{low}/Stor._{low}$/Unfed treatment (LLU) of overwintering YOY bluefish. Variables found to have a significantly heterogeneous variances based upon Levene's test were reanalyzed nonparametrically using the Mann-Whitney U-test (MWU).

TIME-LLU Variable	Trans.	Test	Mean Initial	Std.Dev. Initial	Mean Final	Std.Dev. Final	T- or Z-Value	p	p Levene
liver_%_ash	log	t-test	1.086110	0.199161	1.020599	0.096095	0.738422	0.474453	0.009030
liver_%_ash	raw	MWU	N/A	N/A	N/A	N/A	0.387298	0.698536	N/A
viscera_%_ash	raw	T-test	9.640022	2.649778	8.398167	1.350057	1.043574	0.317250	0.069770
WM_%_ash	raw	T-test	6.717920	0.468326	6.091459	0.624490	2.152410	0.052417	0.495806
RM_%_ash	log	t-test	0.945106	0.177493	0.809098	0.110032	1.645545	0.125778	0.026304
RM_%_ash	raw	MWU	N/A	N/A	N/A	N/A	1.936492	0.052808	N/A
skin_%_ash	raw	T-test	7.315920	1.557885	5.774555	0.760946	2.217167	0.046676	0.163132

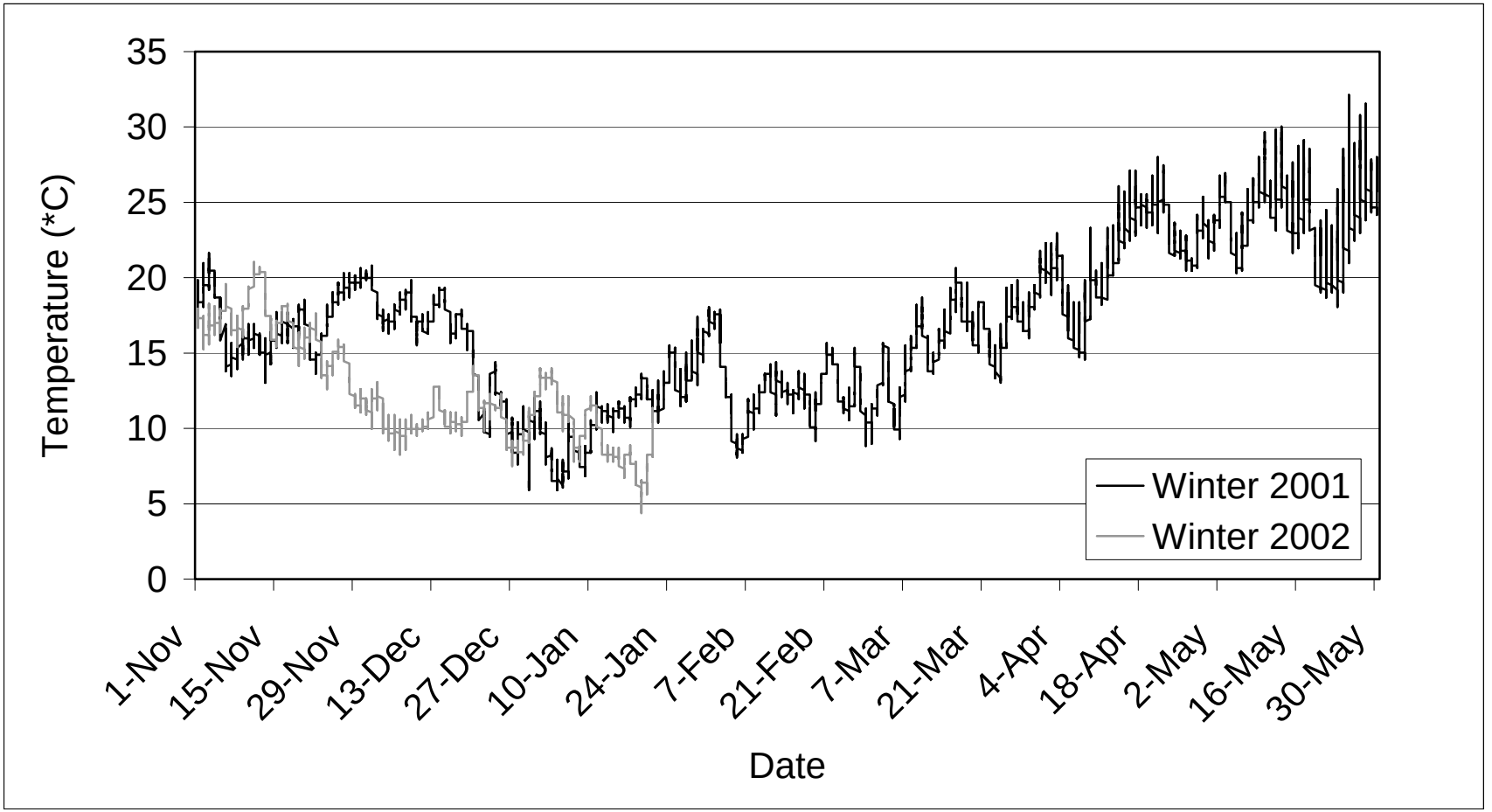


Figure 1. Water temperatures recorded in bluefish tanks during the 2001 and 2002 mesocosm experiments.

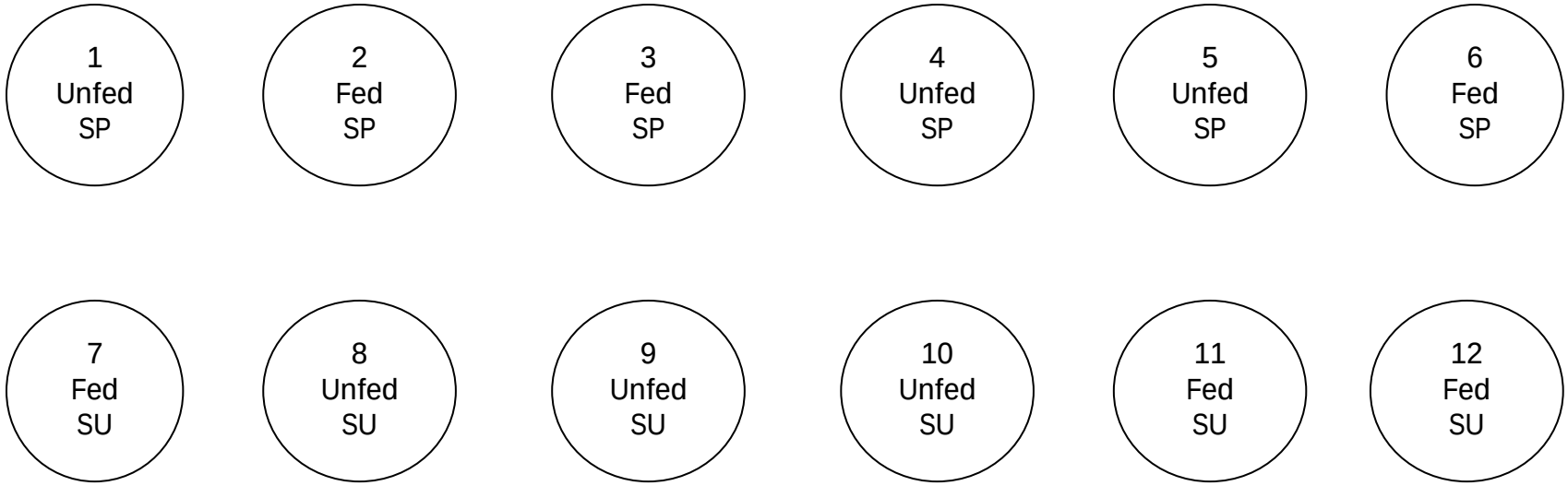


Figure 2. Tank layout for the 2001 experiment. SP=Spring-spawned YOY bluefish and SU=summer-spawned YOY bluefish.

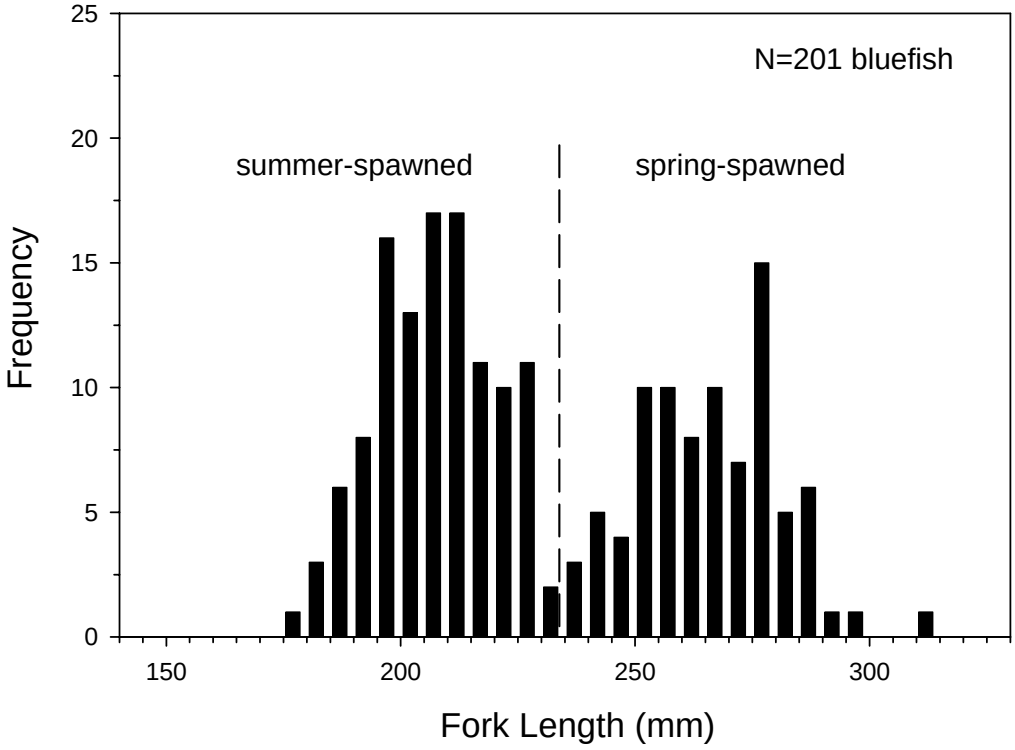
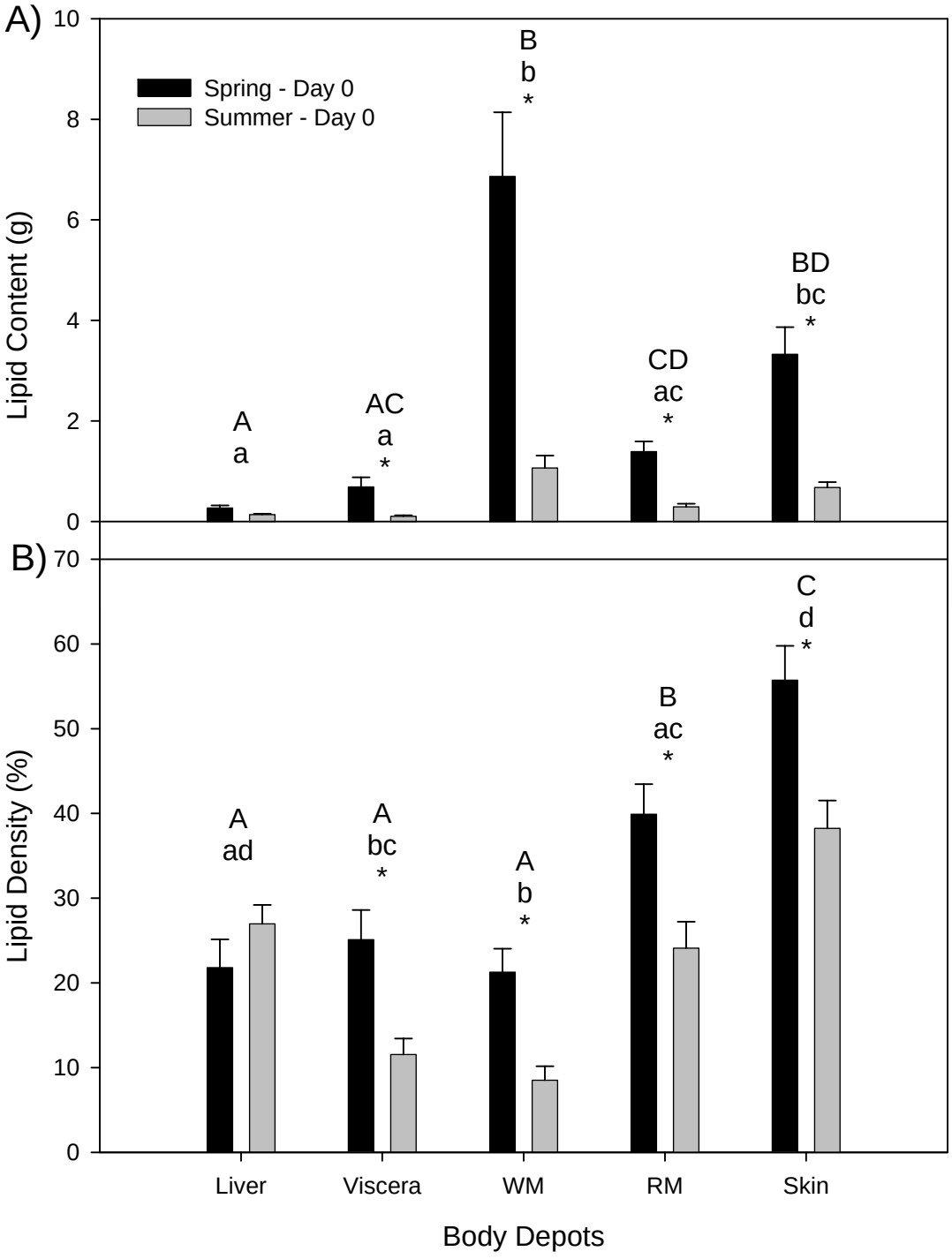


Figure 3. Initial length-frequency distributions of spring- and summer-spawned YOY bluefish subjects measured on day 0 (19 Nov 2001) of the 2001 overwinter mesocosm experiment.

Figure 4. Mean (±S.E.) lipid content (A) and lipid density (B) of different body depots (liver, viscera, white muscle, red muscle and skin) for spring versus summer cohorts of YOY bluefish subsampled on day 0 of the experiment. Tissue means sharing the same upper-case letter (spring bluefish only) are not significantly different (Tukey multiple comparisons test, α=0.05). Tissue means sharing the same lower-case letter (summer bluefish only) are not significantly different (Tukey multiple comparisons test, α=0.05). An asterisk (*) denotes a significant difference between cohorts within a given body depot (Tukey multiple comparisons test, α=0.05).


A)
B)
Spring - Day 0
Summer - Day 0
Lipid Content (g)
Lipid Density (%)
A
a
AC
a
*
B
b
*
CD
ac
*
BD
bc
*
A
ad
A
bc
*
A
b
*
B
ac
*
C
d
*
Liver
Viscera
WM
RM
Skin
Body Depots

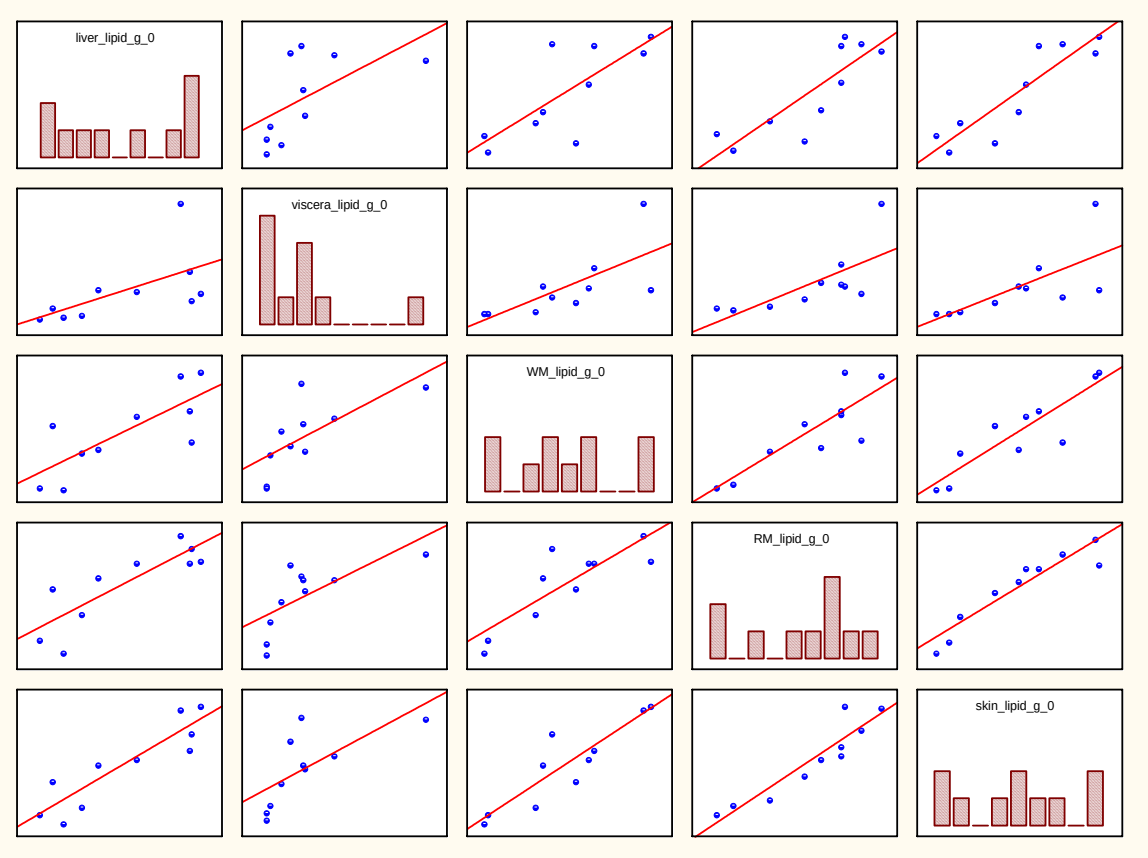


Figure 5. Scatter-plot matrices illustrating the association of lipid content (g) among five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in spring cohort bluefish subsampled on day 0 of the 2001 experiment.

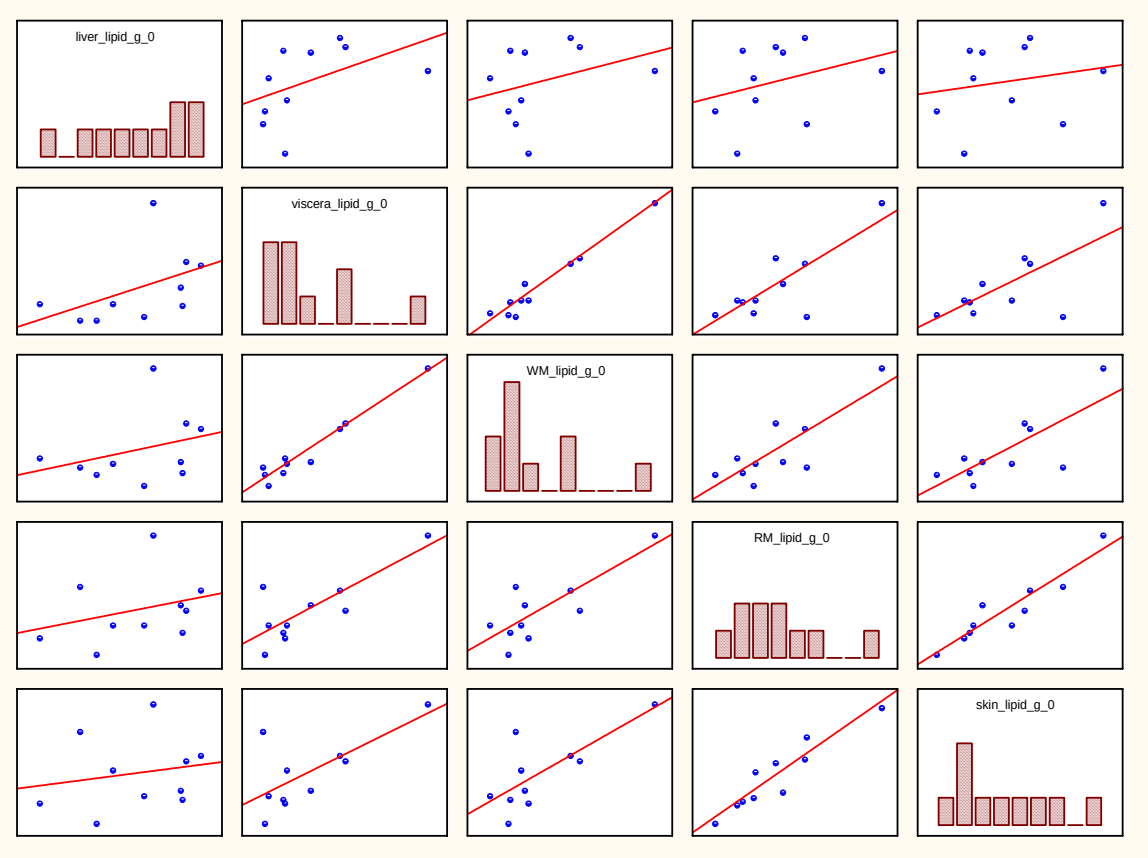


Figure 6. Scatter-plot matrices illustrating the association of lipid content (g) among five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in summer cohort bluefish subsampled on day 0 of the 2001 experiment.

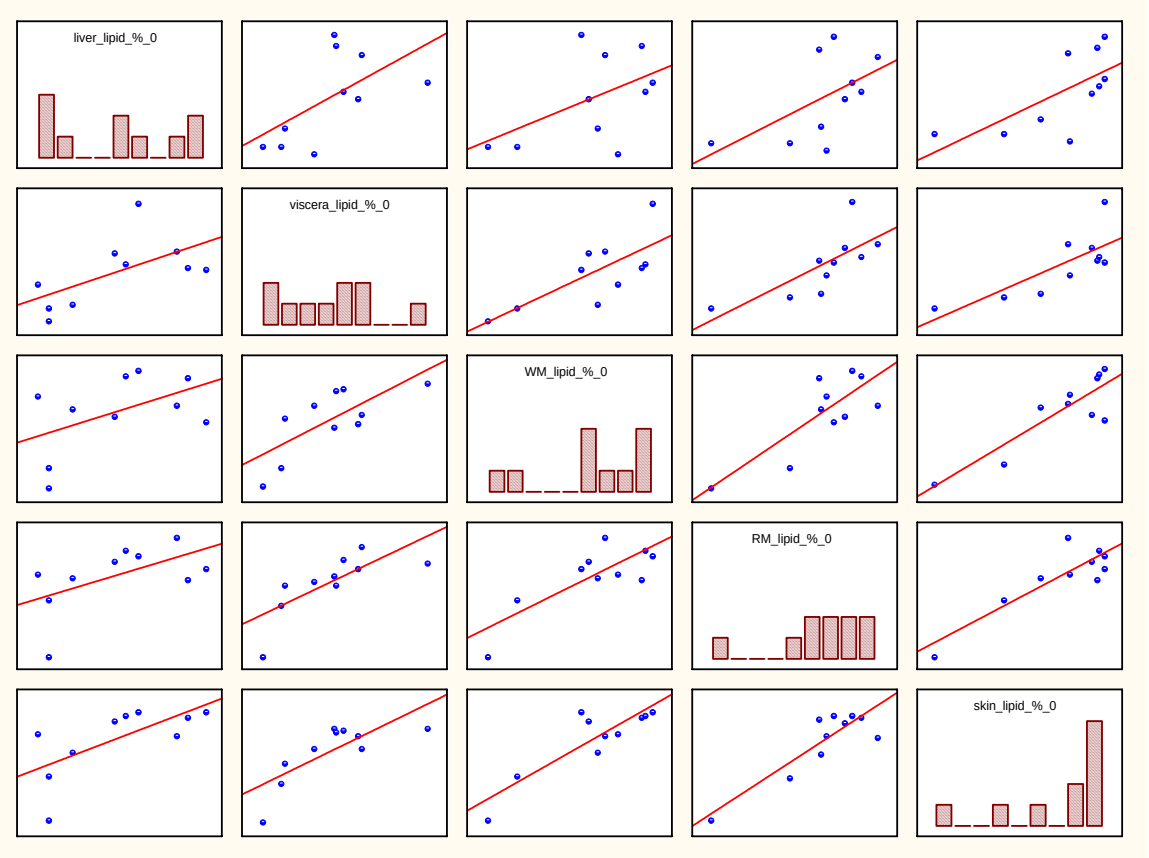


Figure 7. Scatter-plot matrices illustrating the association of lipid density (%) among five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in spring cohort bluefish subsampled on day 0 of the 2001 experiment.

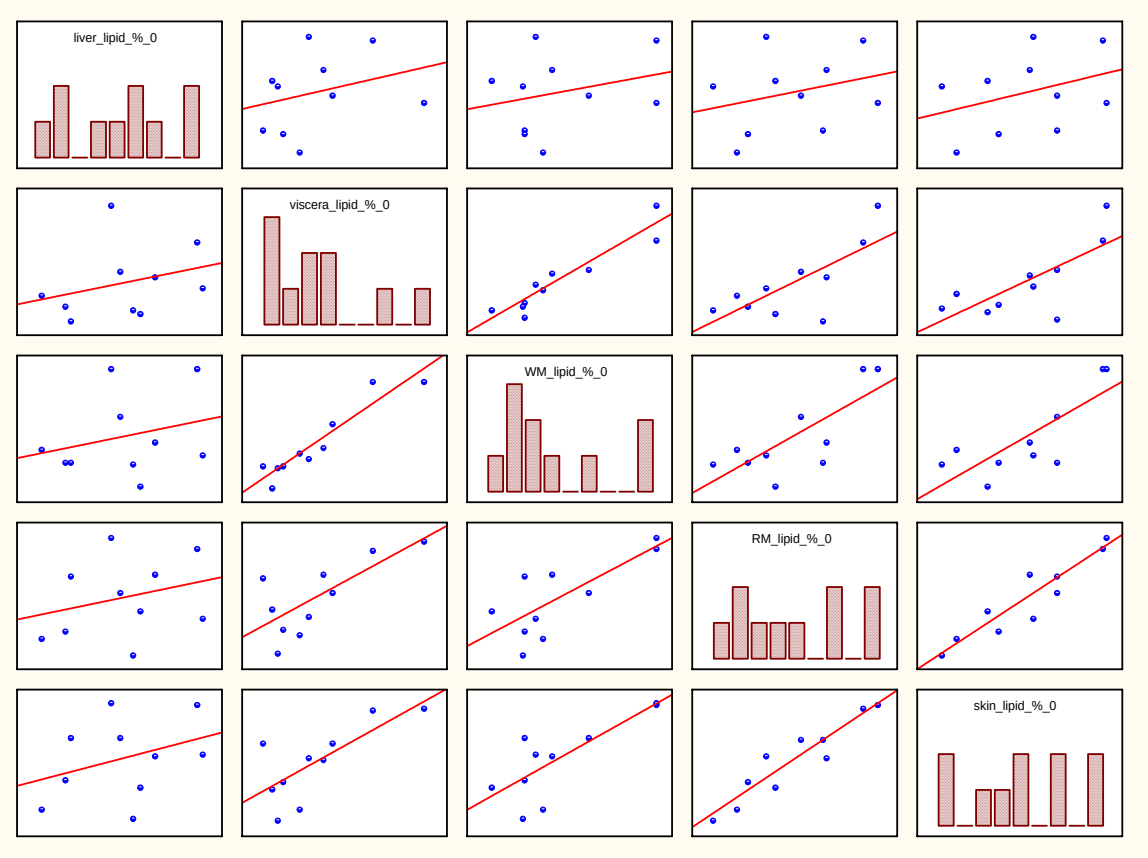


Figure 8. Scatter-plot matrices illustrating the association of lipid density (%) among five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in summer cohort bluefish subsampled on day 0 of the 2001 experiment.

Figure 9. Effects of cohort of origin (spring- versus summer-spawned) and feeding status (fed versus unfed) on the mean lipid content of (A) liver, (B) viscera, (C) white muscle (WM), (D) red muscle (RM) and (E) skin of overwintering YOY bluefish during the 2001 mesocosm experiment.

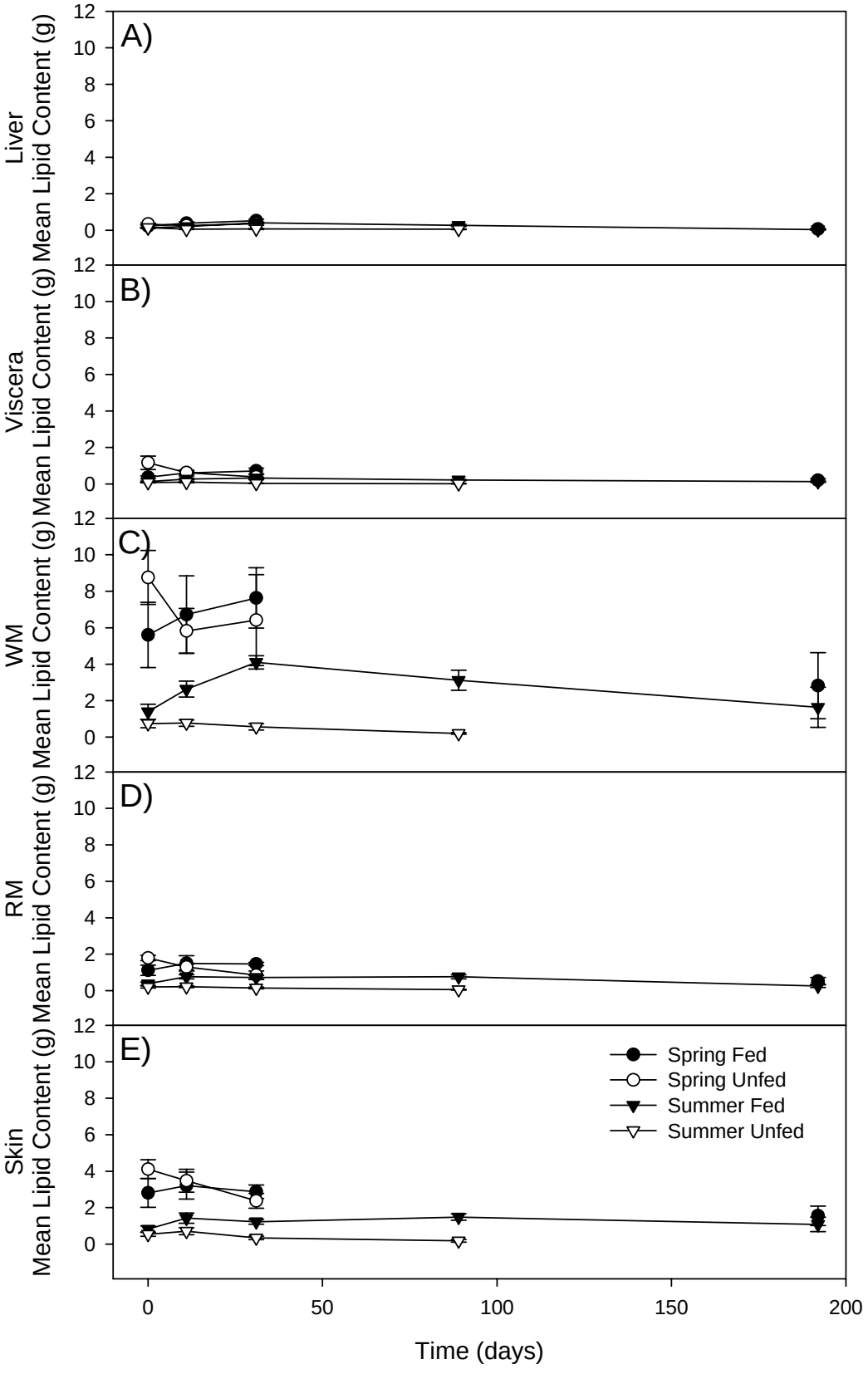

A)
B)
C)
D)
E)
Liver
Viscera
WM
RM
Skin
Mean Lipid Content (g)
Spring Fed
Spring Unfed
Summer Fed
Summer Unfed
Time (days)
0
50
100
150
200

Figure 10. Effects of cohort of origin (spring- versus summer-spawned) and feeding status (fed versus unfed) on the mean lipid density (%) of (A) liver, (B) viscera, (C) white muscle (WM), (D) red muscle (RM) and (E) skin of overwintering YOY bluefish during the 2001 mesocosm experiment.

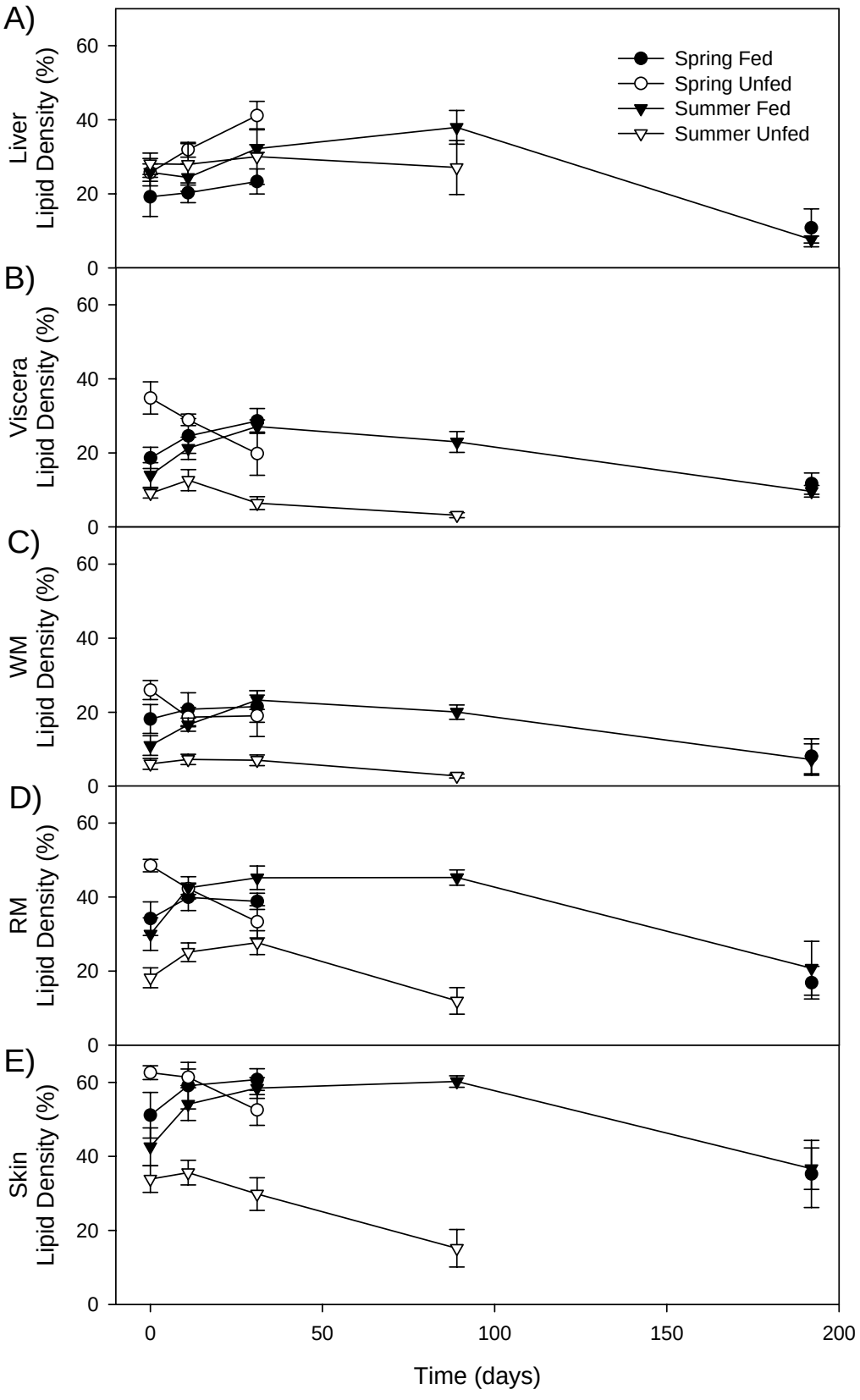
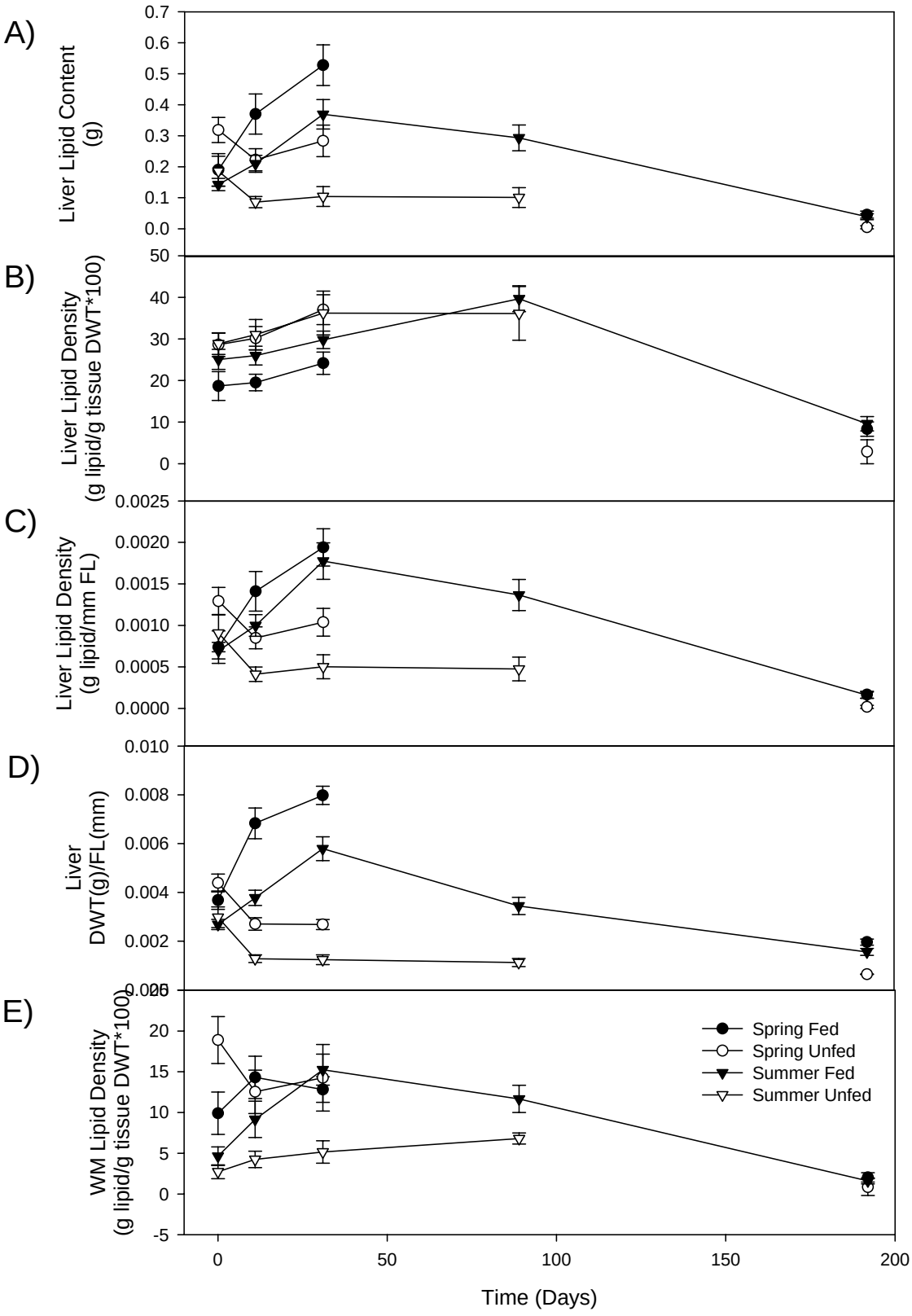

A)
B)
C)
D)
E)
Liver Lipid Density (%)
Viscera Lipid Density (%)
WM Lipid Density (%)
RM Lipid Density (%)
Skin Lipid Density (%)
Spring Fed
Spring Unfed
Summer Fed
Summer Unfed
0
20
40
60
0
50
100
150
200
Time (days)

Figure 11. Effects of cohort of origin (spring- versus summer-spawned) and feeding status (fed versus unfed) on various body condition indices [(A) liver lipid content, (B) liver lipid density, (C) liver lipid density, (D) liver dry weight (DWT)/FL, (E) white muscle (WM) lipid density] of overwintering YOY bluefish during the 2001 mesocosm experiment.


A)
Liver Lipid Content (g)
B)
Liver Lipid Density (g lipid/g tissue DWT*100)
C)
Liver Lipid Density (g lipid/mm FL)
D)
Liver DWT(g)/FL(mm)
E)
WM Lipid Density (g lipid/g tissue DWT*100)
Spring Fed
Spring Unfed
Summer Fed
Summer Unfed
Time (Days)

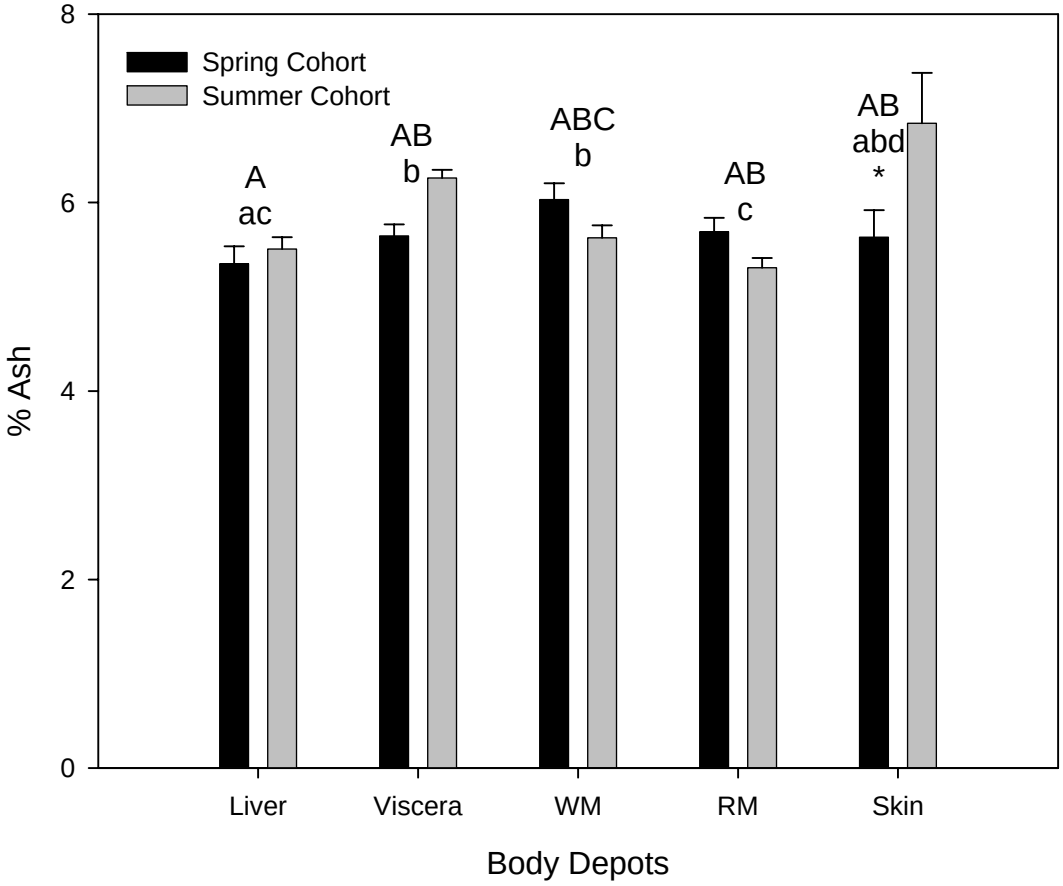


Figure 12. Mean (±S.E.) ash content (% ash) of different body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] for spring versus summer cohorts of YOY bluefish subsampled on day 0 of the experiment. Tissue means sharing the same upper-case letter (spring bluefish only) are not significantly different (Tukey multiple comparisons test, α=0.05). Tissue means sharing the same lower-case letter (summer bluefish only) are not significantly different (Tukey multiple comparisons test, α=0.05). An asterisk (*) denotes a significant difference between cohorts within a given body depot (Tukey multiple comparisons test, α=0.05).

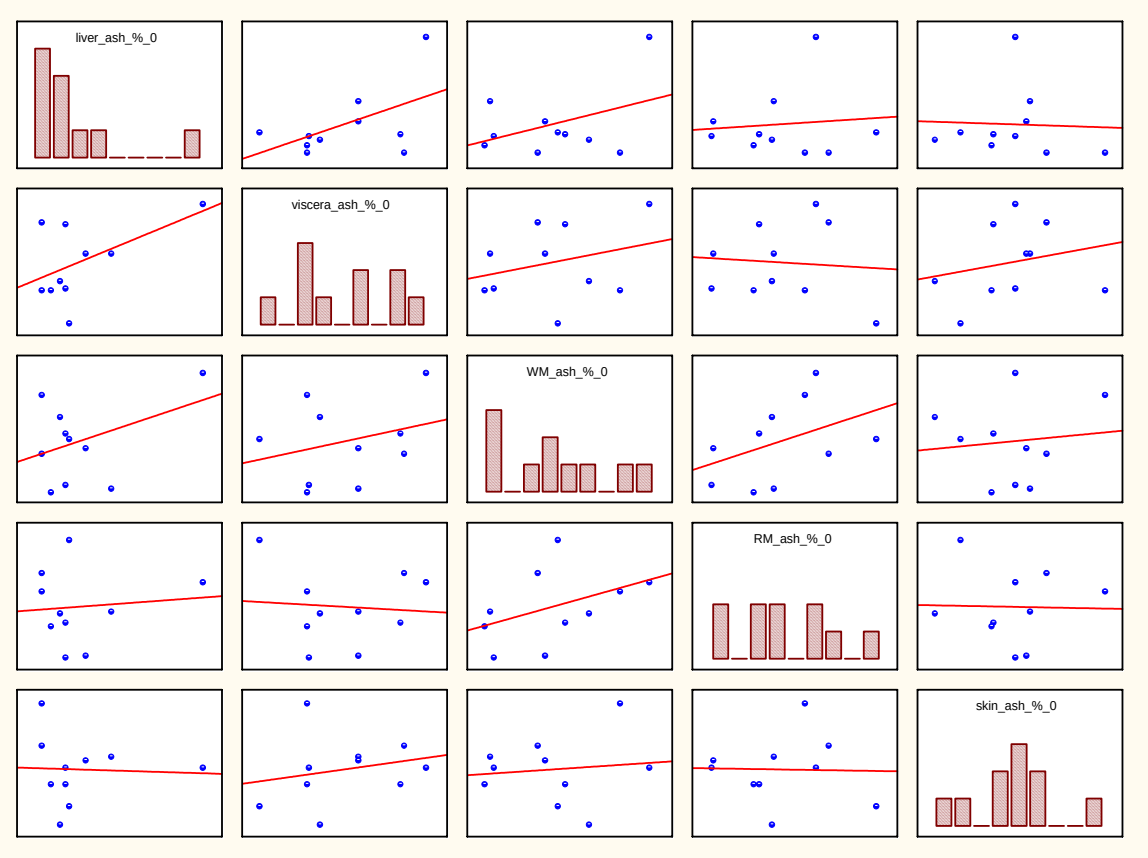


Figure 13. Scatter-plot matrices illustrating the association of ash content (% ash) among five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in spring cohort bluefish subsampled on day 0 of the 2001 experiment.

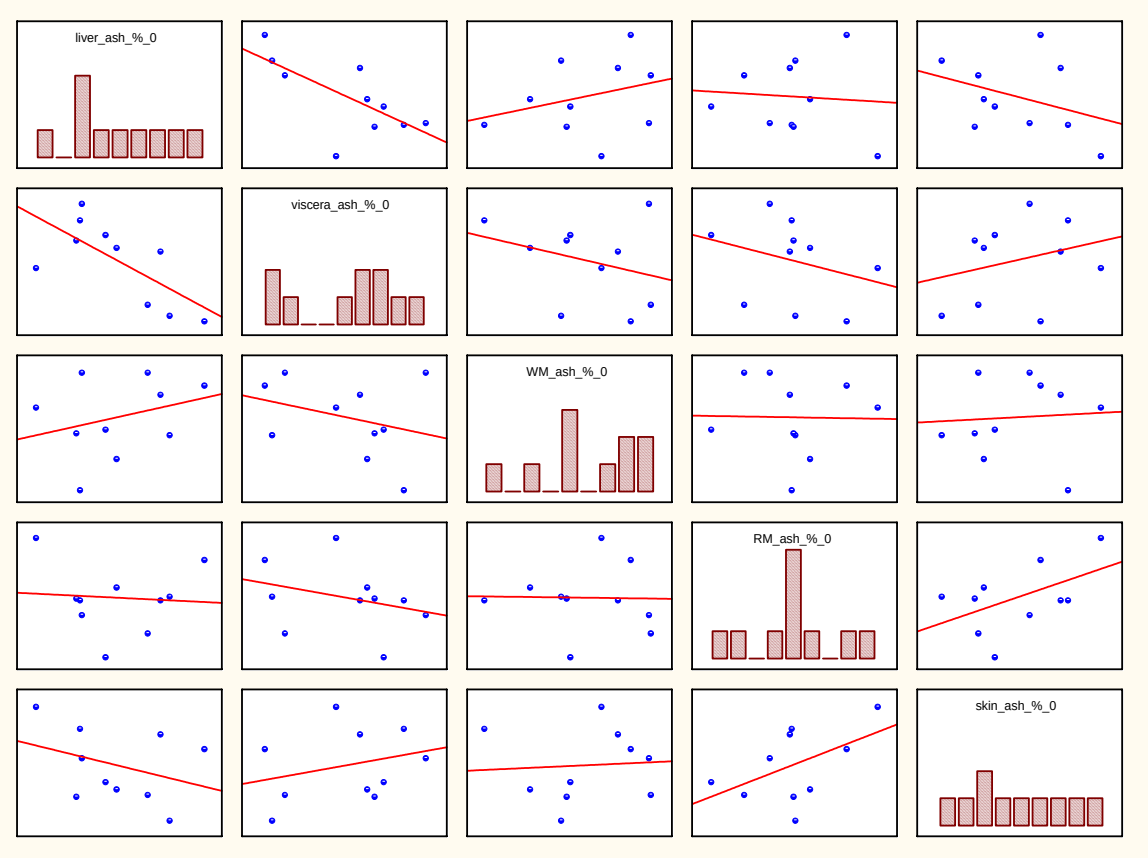


Figure 14. Scatter-plot matrices illustrating the association of ash content (% ash) among five body depots [liver, viscera, white muscle (WM), red muscle (RM) and skin] in summer cohort bluefish subsampled on day 0 of the 2001 experiment.

Figure 15. Effects of cohort of origin (spring- versus summer-spawned) and feeding status (fed versus unfed) on ash content (% ash) of (A) liver, (B) viscera, (C) white muscle (WM), (D) red muscle (RM) and (E) skin of overwintering YOY bluefish during the 2001 mesocosm experiment.

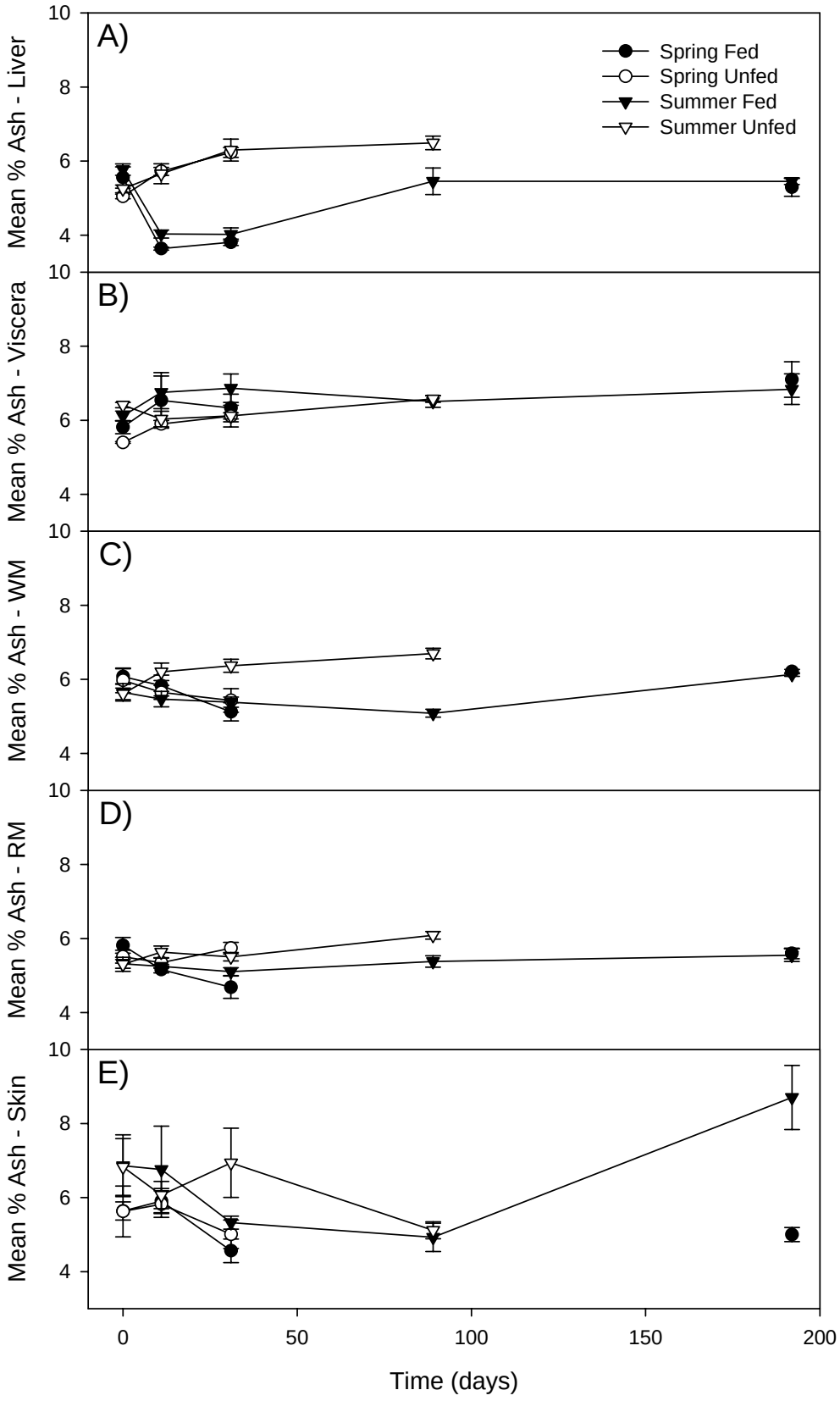

A)
B)
C)
D)
E)
Spring Fed
Spring Unfed
Summer Fed
Summer Unfed
Mean % Ash - Liver
Mean % Ash - Viscera
Mean % Ash - WM
Mean % Ash - RM
Mean % Ash - Skin
10
8
6
4
0
50
100
150
200
Time (days)

Figure 16. Effects of cohort of origin (spring- versus summer-spawned) and feeding status (fed versus unfed) on ash content (% ash) of (A) liver and (B) white muscle (WM) of overwintering YOY bluefish during the 2001 mesocosm experiment.

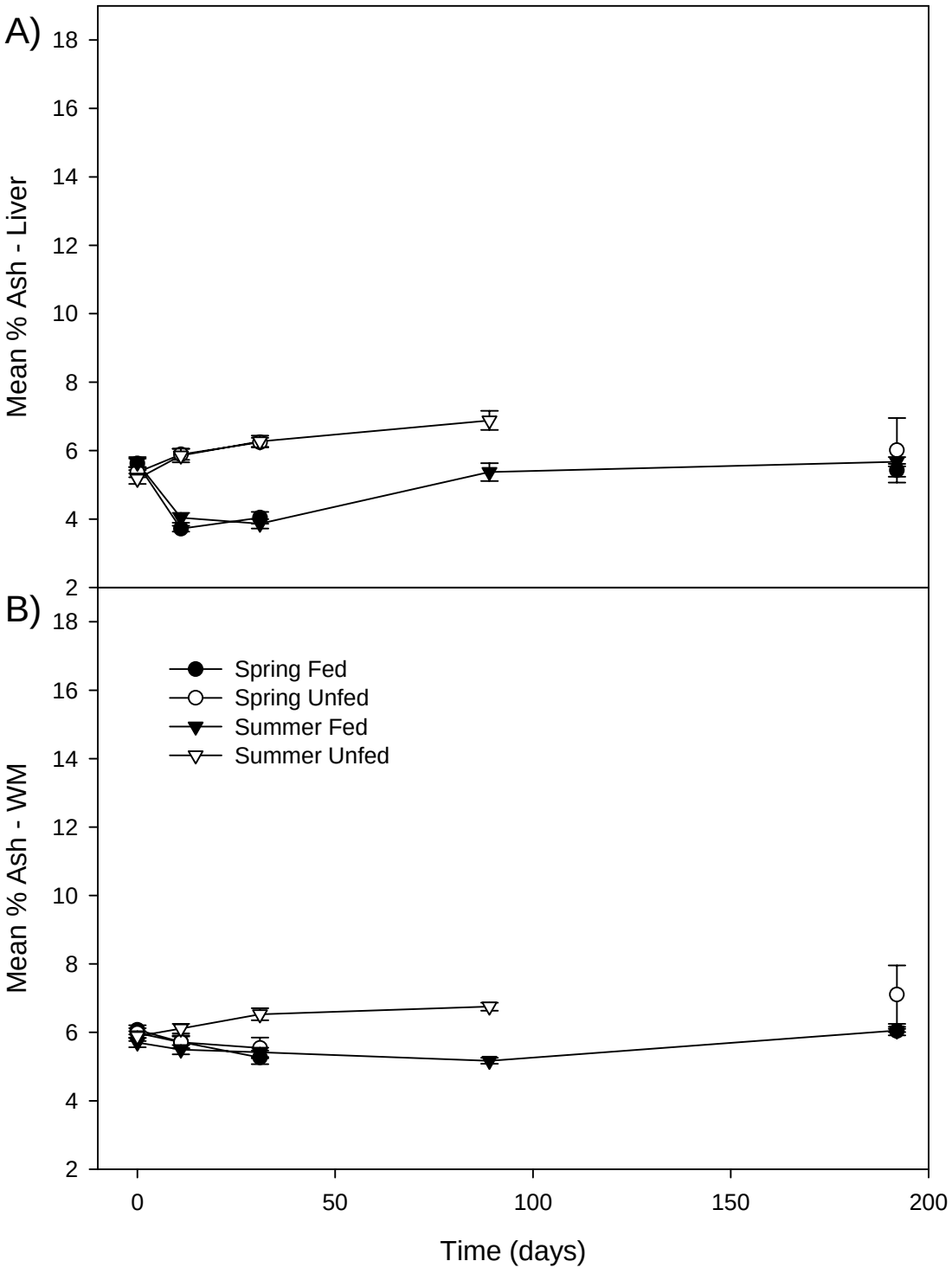
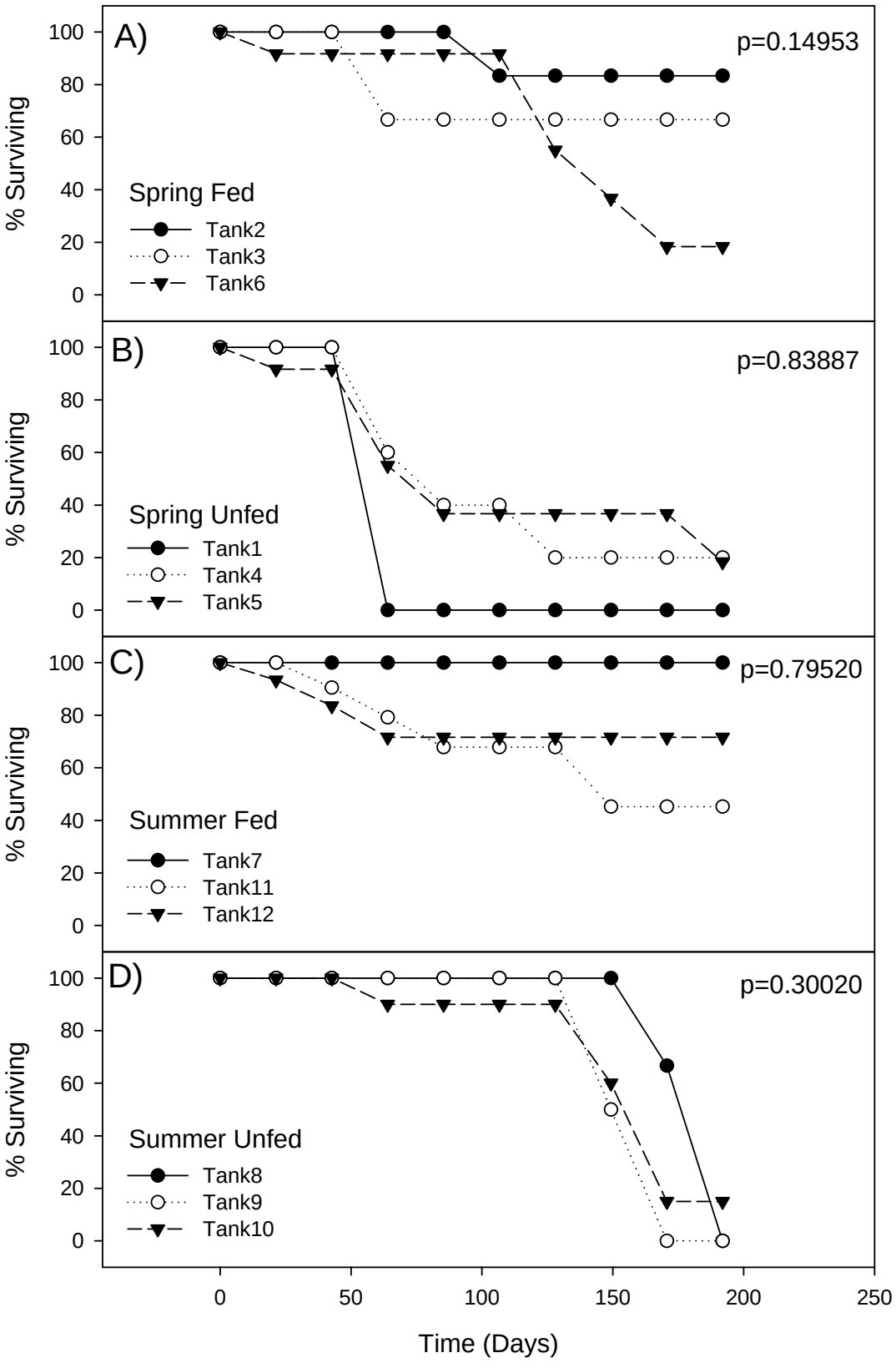

A)
B)
Mean % Ash - Liver
Mean % Ash - WM
Spring Fed
Spring Unfed
Summer Fed
Summer Unfed
Time (days)
2
4
6
8
10
12
14
16
18
0
50
100
150
200

Figure 17. Overwinter survival curves for (A) spring-fed (B) spring-unfed (C) summer-fed (D) summer-unfed YOY bluefish held in mesocosm tanks (n=3 replicate tanks per treatment). Survival curves are based on Kaplan-Meier product-limit estimates. P-values indicate results of Mantel log-rank tests comparing replicate curves within each treatment (α=0.05).


A)
p=0.14953
Spring Fed
Tank2
Tank3
Tank6
B)
p=0.83887
Spring Unfed
Tank1
Tank4
Tank5
C)
p=0.79520
Summer Fed
Tank7
Tank11
Tank12
D)
p=0.30020
Summer Unfed
Tank8
Tank9
Tank10
% Surviving
Time (Days)
0
20
40
60
80
100
0
50
100
150
200
250

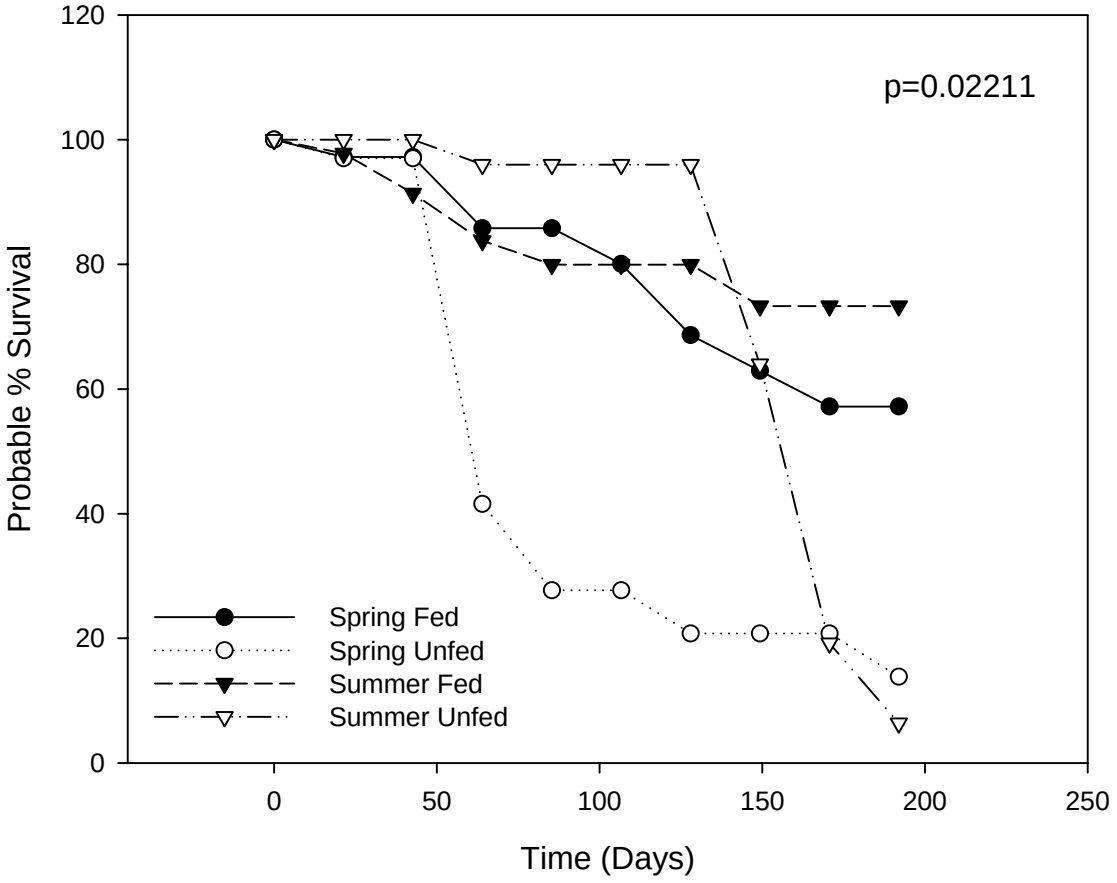


Figure 18. Mean overwinter survival curves for each treatment (spring fed, spring unfed, summer fed and summer unfed) of YOY bluefish held in mesocosm tanks (n=3 replicate tanks per treatment). Survival curves are based on Kaplan-Meier product-limit estimates. P-values indicate results of Mantel log-rank tests comparing survival curves across treatments (α=0.05).

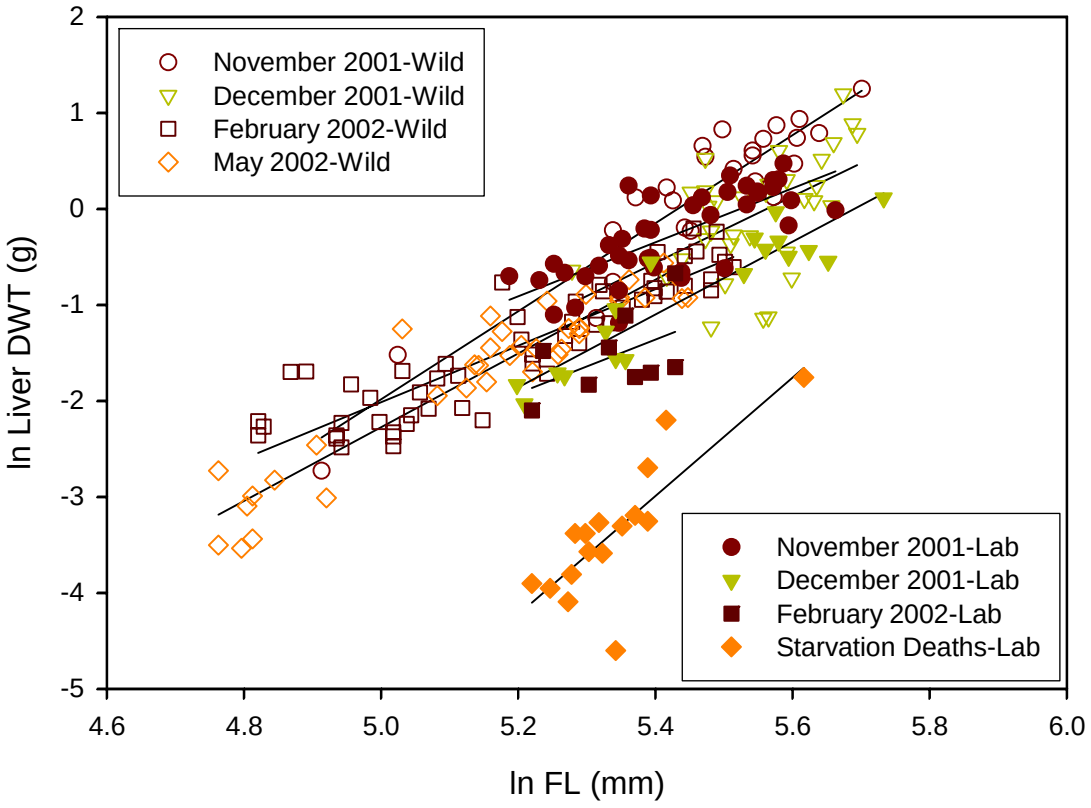


Figure 19. Comparison of overwinter changes in liver dry weights of wild versus starved laboratory bluefish.

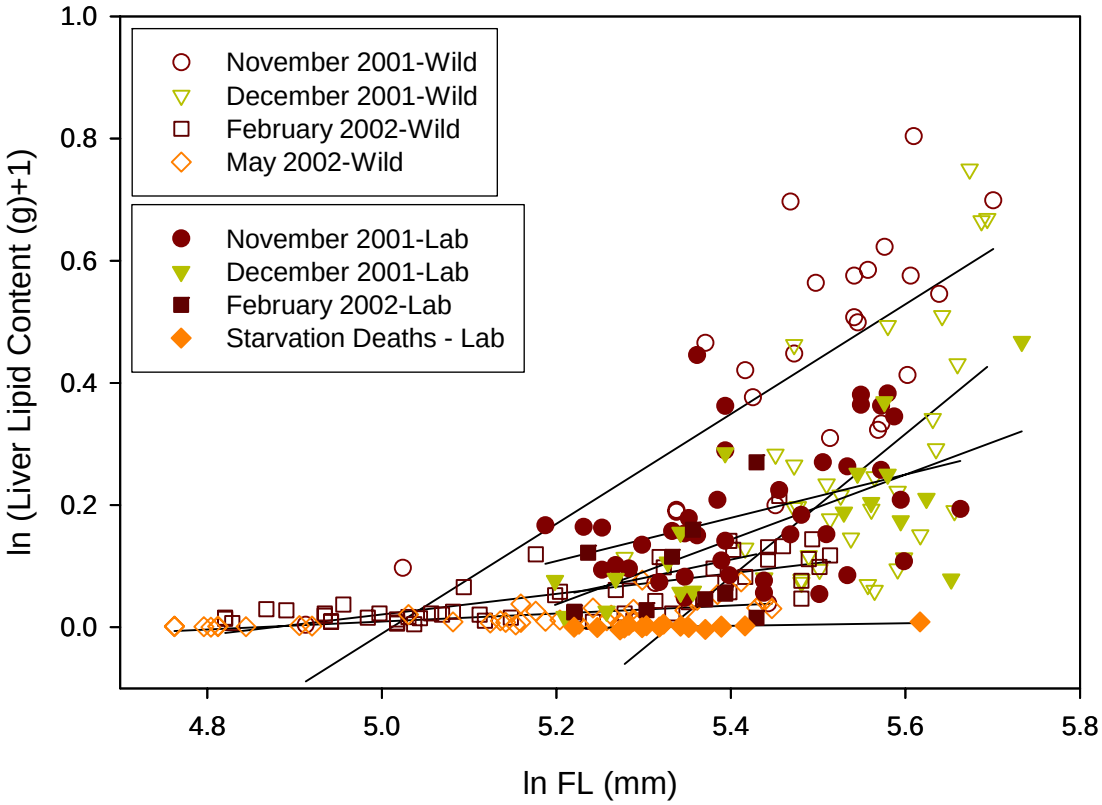


Figure 20. Comparison of overwinter changes in liver lipid content of wild versus starved laboratory bluefish.

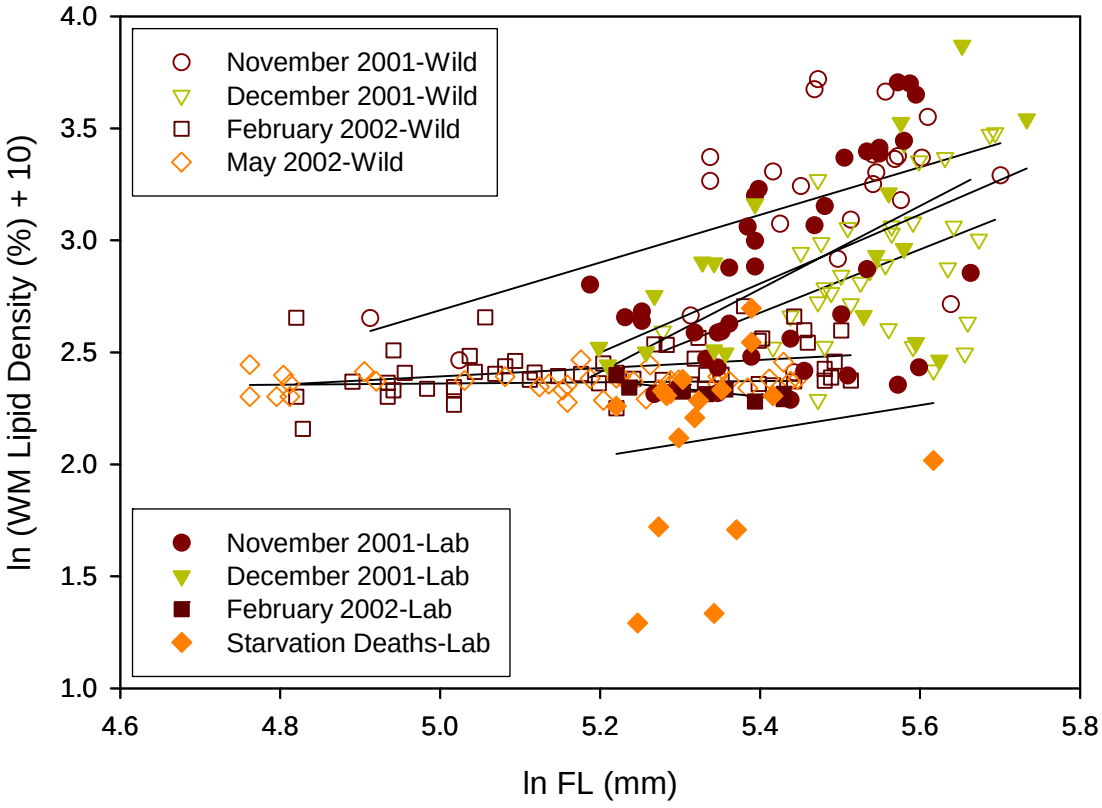


Figure 21. Comparison of overwinter changes in WM lipid density of wild versus starved laboratory bluefish.

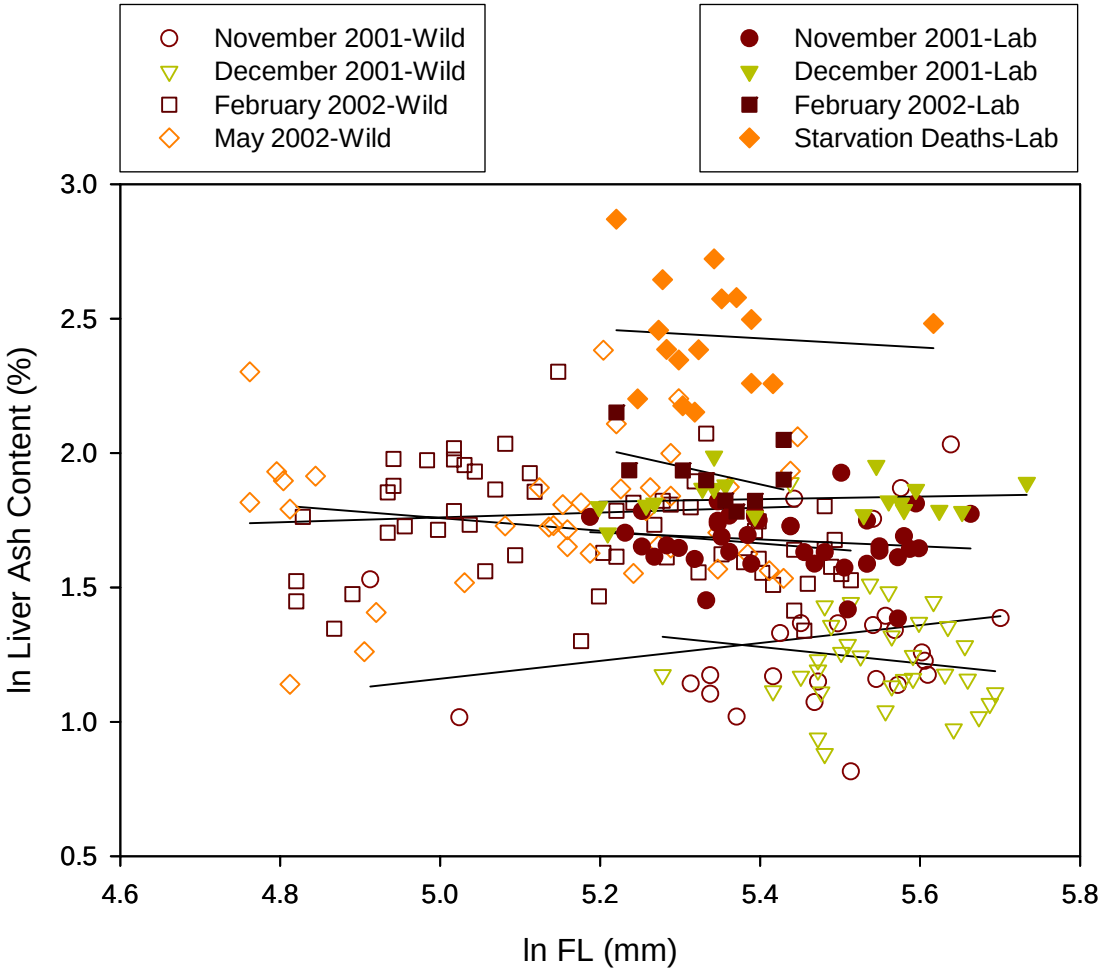


Figure 22. Comparison of overwinter changes in liver ash content of wild versus starved laboratory bluefish.

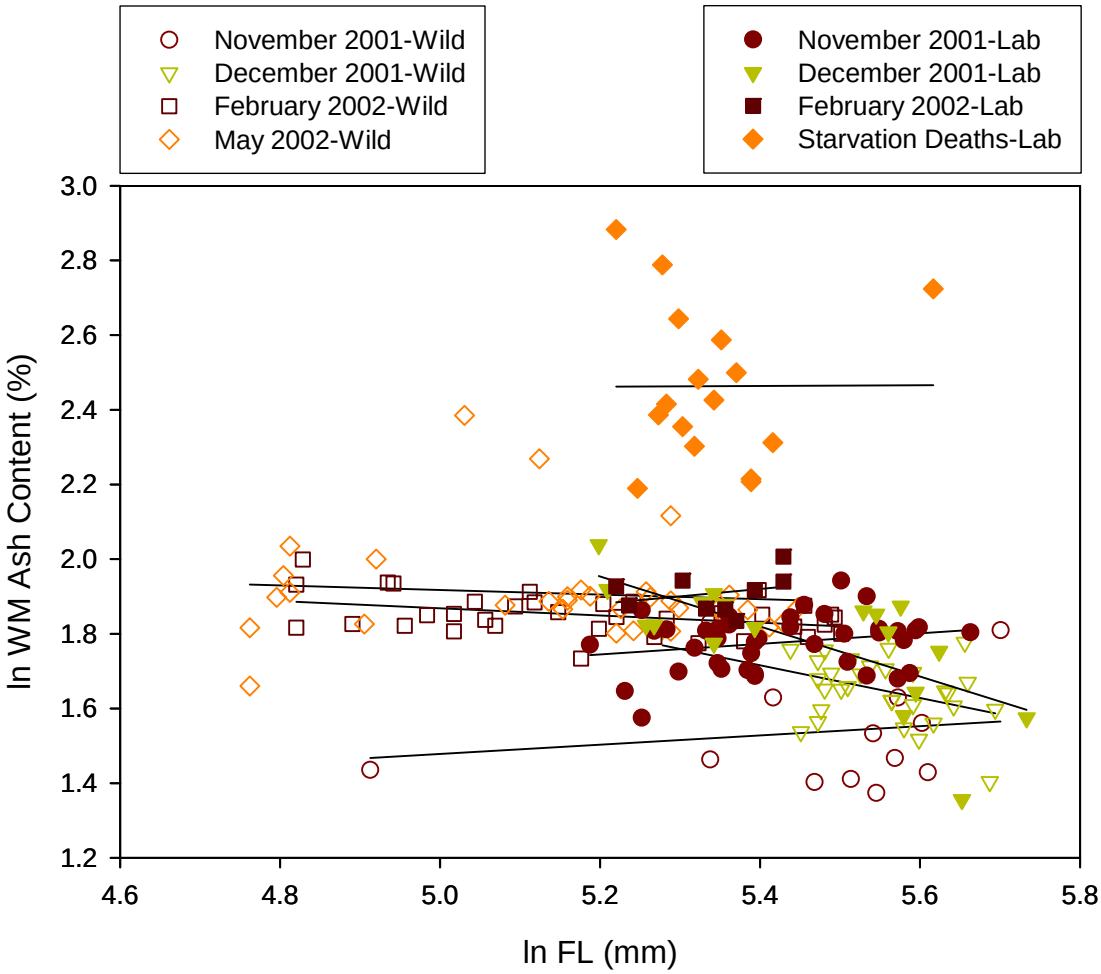


Figure 23. Comparison of overwinter changes in the WM ash content of wild versus starved laboratory bluefish.

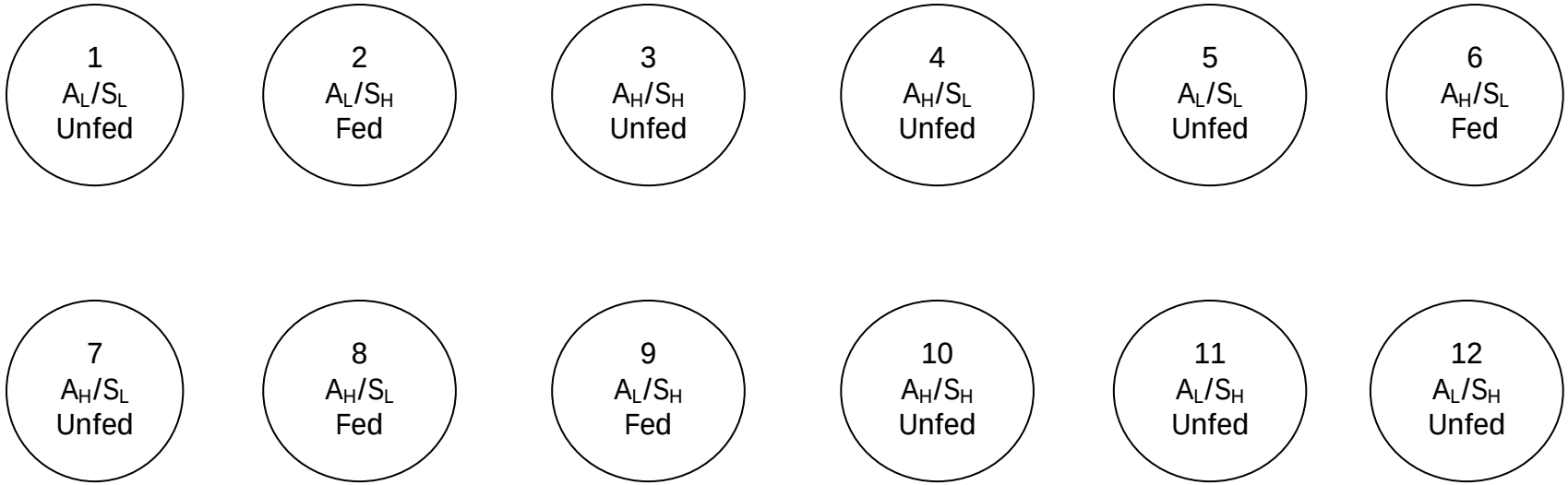


Figure 24. Tank layout for the 2002 experiment. A=activity level and S=pre-winter lipid storage level. H=high and L=low.

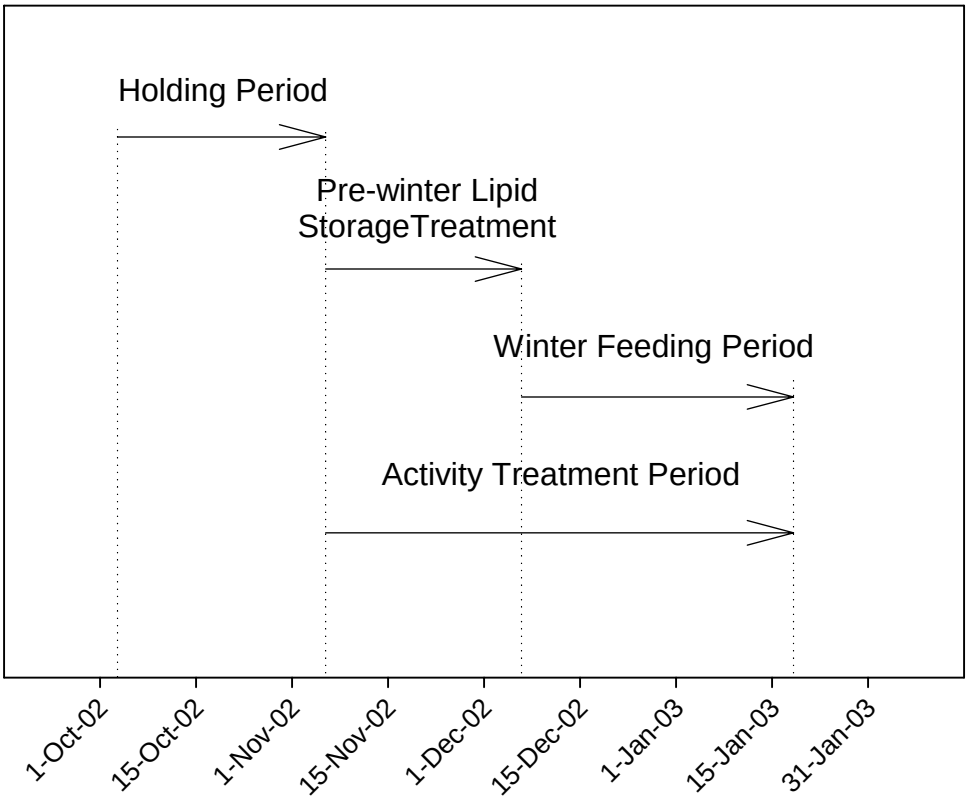


Figure 25. Timeline (2002 mesocosm experiment).

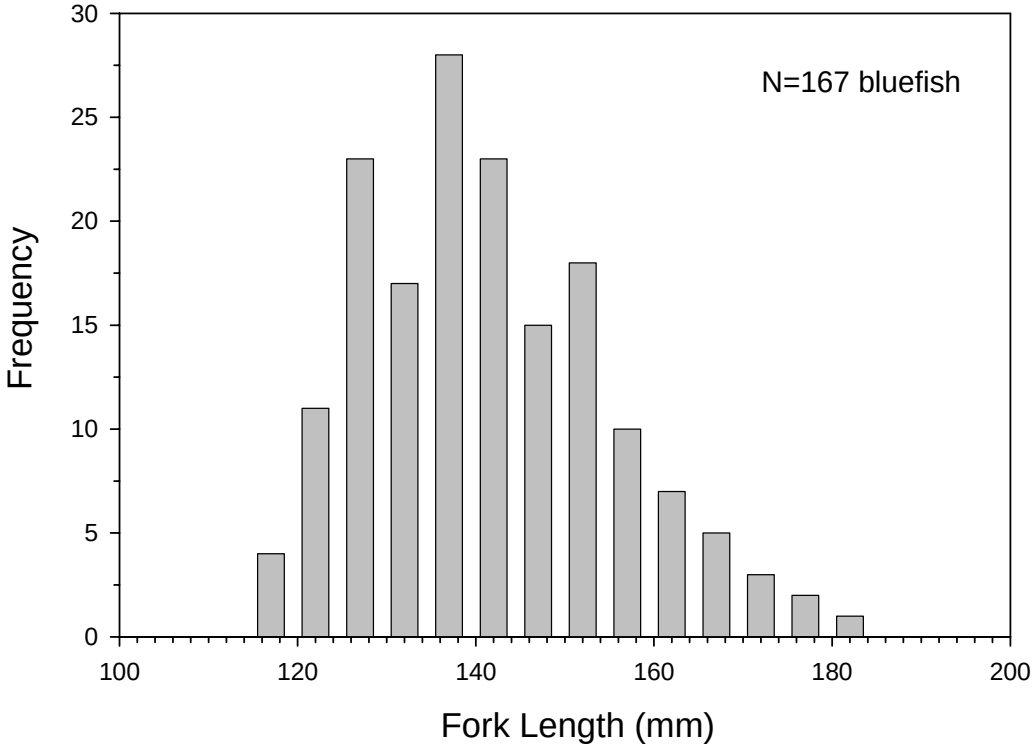


Figure 26. Initial length-frequency distributions of summer-spawned YOY bluefish subjects measured on 31 Oct. 2002 of the 2002 overwinter mesocosm experiment.

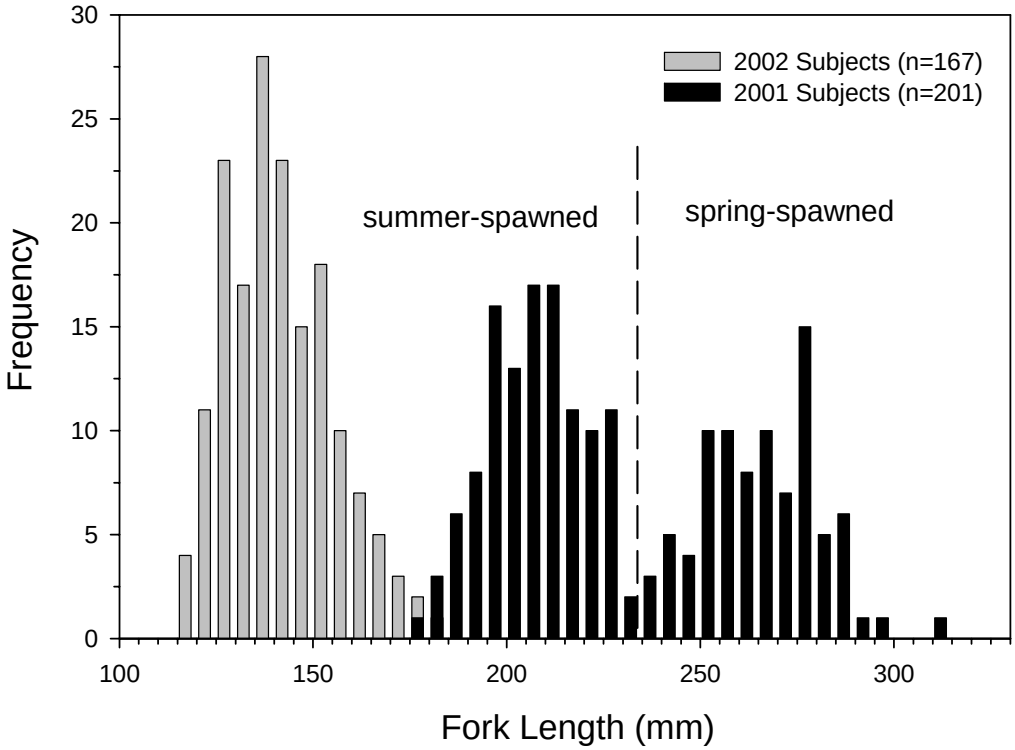
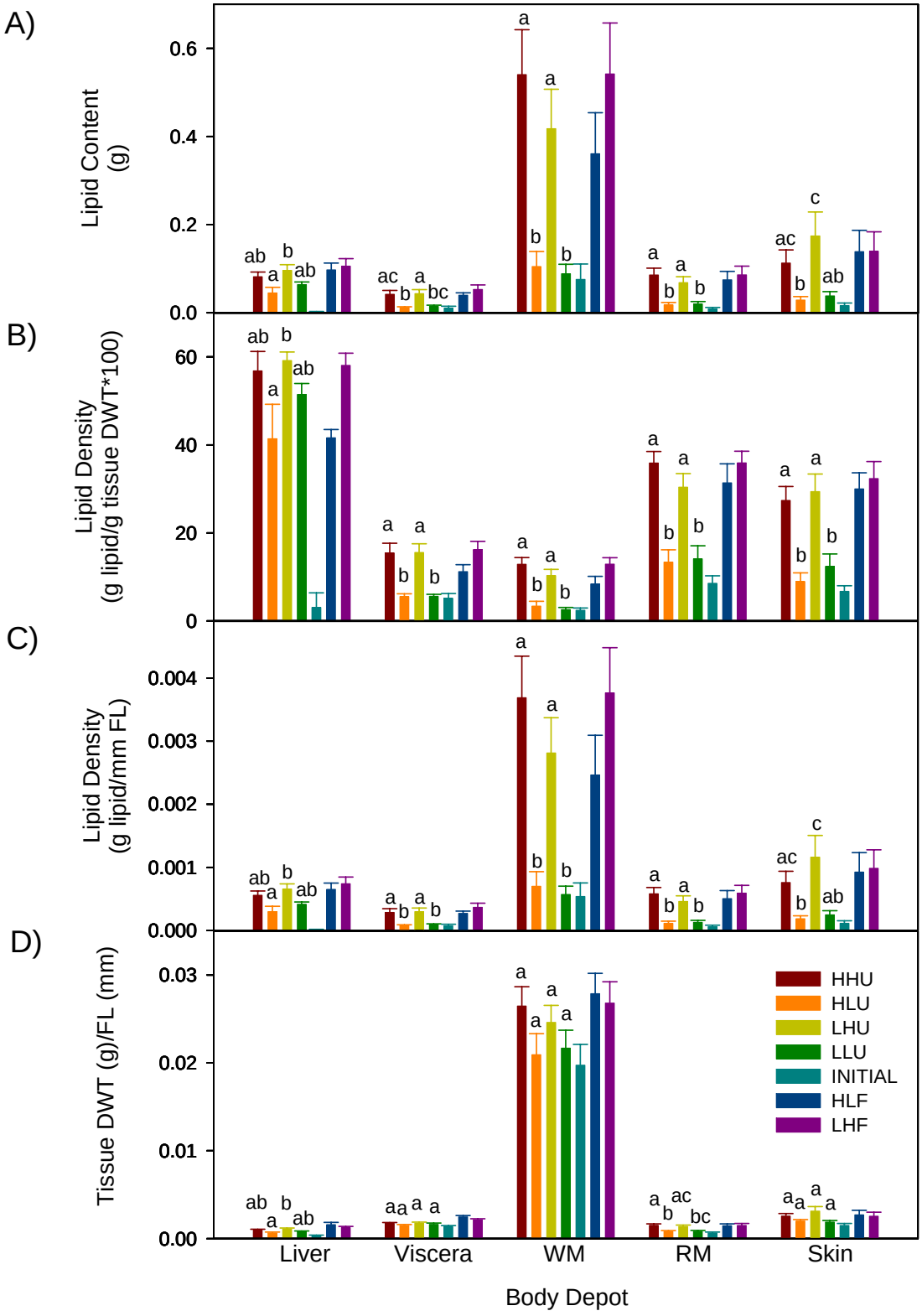


Figure 27. Initial length-frequency distributions of spring- and summer-spawned YOY bluefish subjects from the 2001 and 2002 overwinter mesocosm experiments. Subjects were measured on 19 Nov 2001 and 31 Oct. 2002, respectively.

Figure 28. Effects of activity level, pre-winter lipid storage and winter-feeding on the (A) lipid content, (B) lipid density, (C) lipid density, (D) and tissue dry weight of different body depots [(liver, viscera, white muscle (WM), red muscle (RM) and skin)] in summer-spawned YOY bluefish (±S.E.). Initial subsamples were taken on October 03, 2002. All other samples were taken on January 19, 2003. Treatment means (n=2 tanks) sharing the same lower case letter are not significantly different (Tukey multiple comparisons test, α=0.05). In the legend, the initial letter indicates activity level (H=high and L=low), the second indicates pre-winter storage level (High or Low) and the last indicates winter feeding level (U=unfed and F=fed).


A)
B)
C)
D)
Lipid Content (g)
Lipid Density (g lipid/g tissue DWT*100)
Lipid Density (g lipid/mm FL)
Tissue DWT (g)/FL (mm)
Liver
Viscera
WM
RM
Skin
Body Depot
HHU
HLU
LHU
LLU
INITIAL
HLF
LHF

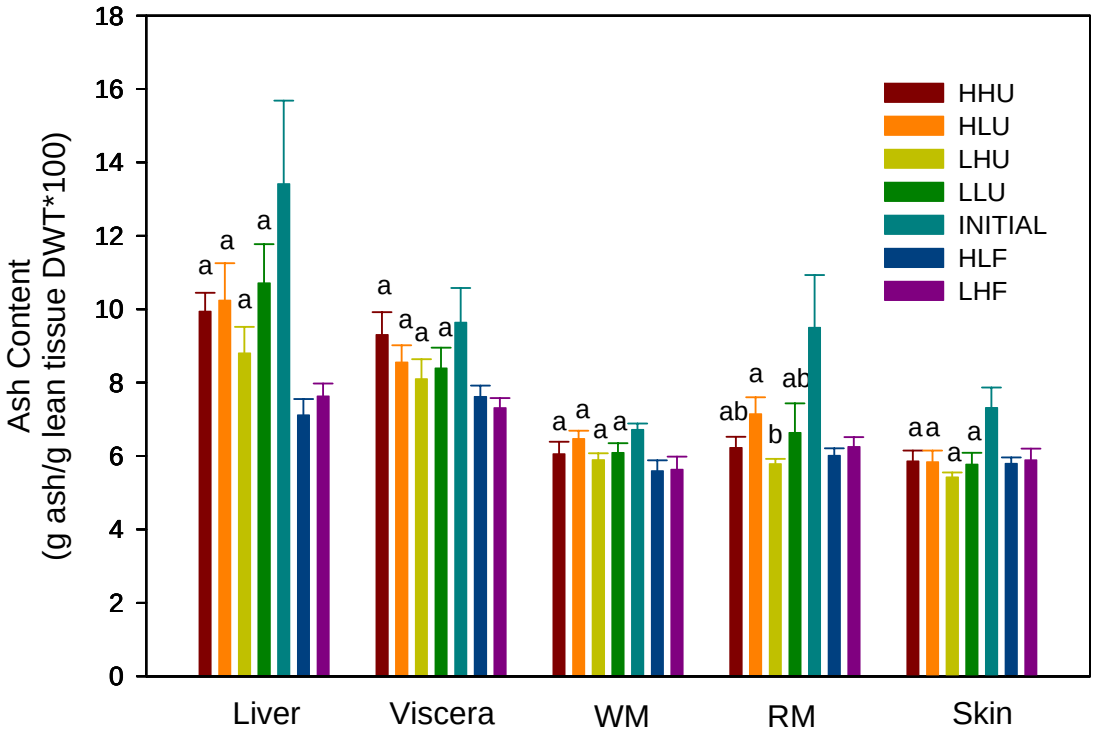


Figure 29. Effects of activity level, pre-winter lipid storage and winter-feeding on the mean ash content (±S.E.) of different body depots [(liver, viscera, white muscle (WM), red muscle (RM) and skin)] in summer-spawned YOY bluefish. Initial subsamples were taken on October 03, 2002. All other samples were taken on January 19, 2003. Treatment means (n=2 tanks) sharing the same lower case letter are not significantly different (Tukey multiple comparisons test, α=0.05). In the legend, the initial letter indicates activity level (H=high and L=low), the second indicates pre-winter storage level (High or Low) and the last indicates winter feeding level (U=unfed and F=fed).