

Autecological effects of habitat alteration: trophic changes in mangrove marsh fish as a consequence of marsh impoundment

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ABSTRACT: The key to predicting, counteracting, and managing environmental change is understanding the mechanisms through which habitat change affects populations of organisms. The mangrove ecosystem of the Indian River Lagoon (IRL), east-central Florida, has been impounded for the purpose of mosquito control and is representative of such large-scale alterations. Sailfin mollies *Poecilia latipinna* in this system exhibit significant interpopulation differences in adult body size in relation to habitat alteration. One likely hypothesis for this pattern is the apparent change in diet experienced as a result of impoundment, as has been documented in earlier studies. To address the question of whether these dietary changes would have been sufficient to cause trophic differences in *P. latipinna* populations relative to impoundment, a comparative study of trophic position in 2 size-divergent populations of *P. latipinna* using stable isotopes was employed. Results suggested that *P. latipinna* from an unimpounded site had a slightly higher trophic position than those from an impounded site and may constitute a partial explanation of differences in adult body size between the 2 populations. Results from the present study also showed that there was a fundamental difference in energy source between the study sites, which suggests greater assimilation of mangrove-based carbon in impoundments. Specific population- and community-level effects of impoundment on mangrove habitats have been demonstrated, indicating a mechanistic link between anthropogenic habitat alteration and life history of *P. latipinna*.

KEY WORDS: Habitat alteration · Life history · Autecology · Stable isotope · Trophic differences · Mangrove food webs · *Poecilia latipinna*

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INTRODUCTION

One recently developed suite of techniques which have proven to be useful in diagnosis of ecological systems (sensu Grimm et al. 1999) is the use of stable isotopes. Stable isotopes have been used to address many different types of issues in ecology, including elucidation of food webs, movements of organisms between habitats, and relative trophic structure (Peterson & Fry 1987). One application to which stable isotopes are particularly well suited is the determination of trophic status using the relative abundance of ^{15}N and ^{13}C in animal tissue (DeNiro & Epstein 1981, Peterson & Fry

1987). Isotopic ratios reflect an integration of assimilated material over a substantial time period and therefore are more representative of long-term dietary patterns than analysis of stomach contents (VanderZanden et al. 1997). Heavy isotopes of nitrogen (^{15}N) increase predictably in tissues from organisms in higher trophic levels (Minagawa & Wada 1984, Cabana & Rasmussen 1996). The fraction of heavy isotopes of carbon (^{13}C) is relatively conserved between trophic levels and reflects carbon source (Fry & Sherr, 1987, VanderZanden et al. 1999). Combined, these 2 isotopic ratios provide an effective means for comparing trophic position and the determination of the primary

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source of energy. Stable isotopes of carbon and nitrogen have been used effectively to examine both intraspecific (Gu et al. 1997) and interspecific (Beaudoin et al. 1999, Fantle et al. 1999, VanderZanden et al. 1999) differences in trophic position.

An example of an ecosystem which has been extensively altered in structure and function is the mangrove habitat found in east-central Florida. Virtually all mangrove areas along the Indian River Lagoon (IRL) were altered through impoundment, having profound impacts on the ecosystem (reviewed in Brockmeyer et al. 1997). Impoundments consist of a low head dam surrounding a mangrove marsh, by which water levels can be regulated for the purpose of controlling mosquito populations (Rey et al. 1999). One of the few studies to directly document the effects of impoundment on the mangrove ecosystem was the classic study of Harrington & Harrington (1961, 1982). Part of the study observed the changes in dietary patterns of fish remaining after impoundment using gut contents. This study suggested that *Poecilia latipinna* populations residing in these mangrove habitats shifted trophic position as a result of impoundment (Table 1). Prior to impoundment, 30% of *P. latipinna* diet consisted of insects, mostly mosquito larvae, with the balance primarily consisting of vascular plants. Following impoundment, *P. latipinna* diet consisted almost entirely of algae (mostly dinoflagellates) and detritus, and the insect fraction virtually disappeared. As impoundment was highly successful at controlling mosquito populations (Brockmeyer et al. 1997), it was the likely mechanism responsible for reduction or elimination of mosquitoes from *P. latipinna* diets. Therefore, evidence exists that *P. latipinna* may have undergone a partial trophic shift following impoundment.

Poecilia latipinna from unimpounded sites are significantly larger than those from impounded sites (Fig. 1; Kemp 2004). Taken together, a possible hypothesis to explain this pattern was that impoundment reduced the body size of *P. latipinna* through the mechanism of reduced energy availability in the form of mosquito

Table 1. *Poecilia latipinna*. Proportional volume of dietary items before and after impoundment. Data are from Harrington & Harrington (1982)

% by volume	Item
Before	
67	Vascular plants
30	Mosquito larvae
2	Other
1	Phytoplankton
After	
51	Phytoplankton
47	Detritus
2	Other

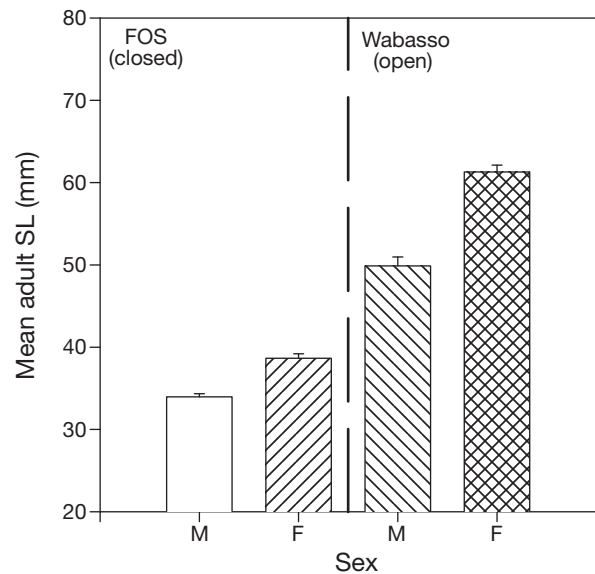


Fig. 1. *Poecilia latipinna*. Mean (+ 1 SE) adult standard length (SL, mm) by sex from open mangrove site (Wabasso) and closed mosquito impoundment (Florida Oceanographic Society [FOS]). Both males (M) and females (F) differ significantly between the open site and closed impoundment (males: 1-way ANOVA, df = 1,271, $p < 0.0001$; females: 1-way ANOVA, df = 1,147, $p < 0.0001$; Kemp 2004)

larvae. The energy density of mosquito larvae is high relative to detritus or plant material, and increased net energy availability results in greater amounts of energy available for other functions such as growth (Brett & Groves, 1979, Hewett & Johnson, 1992, Peck & Walton 2006). If energy allocated towards growth was greater in the open, unimpounded mangrove populations as a result of the presence of mosquito larvae, levels of ^{15}N would be relatively higher in body tissue (DeNiro & Epstein 1981). A comparative study of stable isotope ratios was employed to determine the difference in the contribution of plant and animal material to growth between 2 size-divergent populations and to determine if trophic shifts following impoundment may be the mechanism underlying observed patterns in body size.

MATERIALS AND METHODS

Study sites. Two study sites were chosen from an array of 8 studied mangrove marsh sites along the IRL in east-central Florida (Fig. 2). These 2 sites were selected as being representative of fully impounded and unimpounded mangrove marsh sites with size-divergent *Poecilia latipinna* populations. While a more rigorous comparison could be made through the inclusion of more study sites, logistical constraints, site

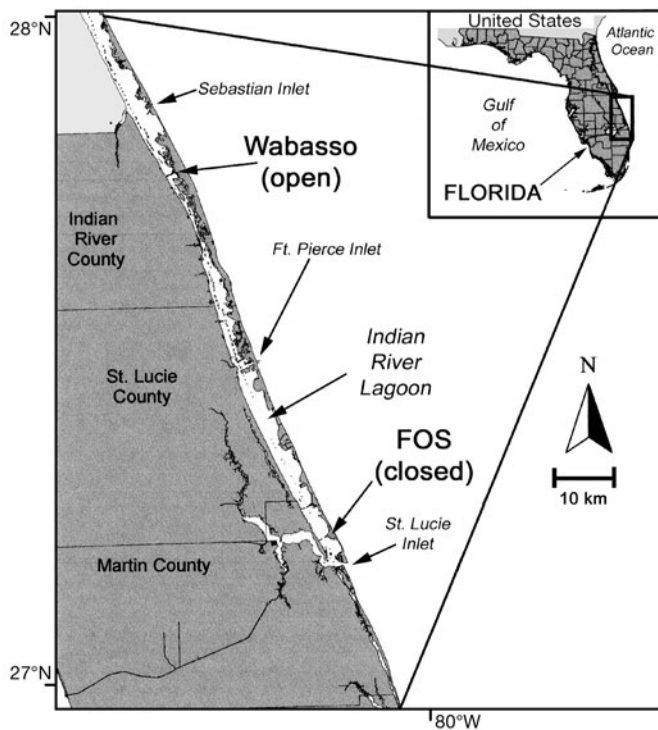


Fig. 2. Indian River Lagoon, showing locations of study sites (Florida Oceanographic Society [FOS] and Wabasso) and inlets from Atlantic Ocean

access issues, and the fact that very few unimpounded areas similar enough for comparison remain in the system precluded this possibility.

Wabasso Island (Wabasso) is located in northern Indian River County, Florida (27° 45' 50" N, 80° 26' 70" W). The site is privately owned and is currently operated as a nature preserve (Environmental Learning Center, Vero Beach, Florida). The marsh was partially ditched to facilitate tidal flushing of the island. The marsh surface is traversed by small tidal creeks running through red mangrove *Rhizophora mangle*, which in turn are bordered by black and white mangrove *Avicennia germinans* and *Laguncularia racemosa*, respectively, and buttonwood *Conocarpus erectus*, as inundation becomes less frequent. Periodically, the entire marsh surface was inundated, and water levels fluctuated throughout the year. The marsh surface was otherwise continuous with the IRL. Adult body sizes of *Poecilia latipinna* were significantly higher at this site than at Florida Oceanographic Society (FOS) (Fig. 1; Wabasso adult standard length [SL] mean [mm] for male = 49.88 ± 1.086 SE, N = 25; female = 61.30 ± 0.828 SE, N = 31; Kemp 2004).

The FOS mangrove marsh is located in Martin County along the IRL and is a semi-contained area inside a larger mosquito impoundment (26° 43' 00" N, 80° 11' 00" W), also maintained as a wildlife preserve. This area was mostly isolated from the surrounding

impoundment by a series of low berms, except during times of exceptionally high water level. Therefore, only indirect, infrequent exchange occurred with waters from the IRL. Vegetation at this site consisted of well-developed red mangrove *Rhizophora mangle* forest. Adult body sizes of *Poecilia latipinna* were significantly smaller here than at Wabasso (Fig. 1; FOS adult SL mean [mm] for male = 33.96 ± 0.391 SE, N = 193; female = 38.66 ± 0.533 SE, N = 31; Kemp 2004).

Separated by a distance of 84 km, both FOS (closed) and Wabasso (open) have generally very similar climates. Over the period 1948 to 2007, the average annual monthly maximum and minimum temperatures (°C) measured at the nearest available weather recording stations (Vero Beach and Stuart) to FOS and Wabasso were 28.4 and 18.7 and 27.7 and 18.1, respectively. In addition, average annual precipitation was 147 cm at FOS and 130 cm at Wabasso. Both sites had maximum monthly average temperatures during the month of August and minimum monthly average temperatures during the month of January. Maximum monthly average precipitation for both sites was observed during September, while the minimum monthly average precipitation was observed for FOS during January and December at Wabasso (climatic data source: Center for Ocean- Atmospheric Prediction Studies at Florida State University website www.coaps.fsu.edu). Distance to the nearest inlet was 4.87 km for FOS and 8.78 km for Wabasso, and average salinities measured in the IRL at those sites were 35.4 and 31.6, respectively.

Field sampling. A representative sample of male, female, and juvenile *Poecilia latipinna* was collected from both study sites using cast and lift nets during July to August, 2002. It was possible to determine sex and maturity externally in the field in *P. latipinna*. Morphology of the male intromittent organ, the gonopodium, indicates maturity in the male, and markings near the gonopore indicate maturity in the female (Trexler & Travis 1990). Individuals lacking these features were assumed to be juveniles. Cast nets (weighted circular nets 0.91 to 1.21 m radius and 63.5 mm bar mesh) were thrown into open areas where fish were present. Lift nets consisted of a 15.9 mm bar mesh net mounted in pound net style on a 1.21 × 1.21 m square PVC frame. The net was hoisted from above to capture fish which collected in the center of the net. *P. latipinna* were immediately placed in water-filled specimen bags and put on ice. These samples were frozen as soon as possible for later analysis. From FOS, 10 females (33 to 46 mm SL), 7 males (39 to 54 mm SL), and 5 juveniles (28 to 34 mm SL) were used for the stable isotope analysis. From Wabasso, 17 females (40 to 68 mm SL), 9 males (43 to 58 mm SL), and 5 juveniles (33 to 42 mm SL) were used for the stable isotope analysis.

For identifying site-specific differences in ambient isotopic fractions, *Melampus coffeus*, a detritivorous pulmonate snail, was collected for analysis. *M. coffeus* was chosen because it is the organism least likely to vary in trophic position between sites (Mook 1986), and thus reflect ambient differences in $\delta^{15}\text{N}$ isotopic ratio (Fry et al. 2000, Post 2002). Mangrove litter and detritus is a primary food source for this snail (Mook 1986), and this species is one of the main processors of mangrove detritus in this ecosystem (McKee & Faulkner 2000, Raulerson 2004). Organisms used for this purpose should have limited dietary breadth, must be relatively long lived, and have low vagility. As *M. coffeus* snails feed on mangrove litter and detritus, live more than a year, and have limited scope for movement to or between sites, this species serves well as a baseline comparison organism. A series of 5 *M. coffeus* snails were collected from the prop roots of *Rhizophora mangle* at different locations from each site, immediately placed on ice, and frozen for later analysis.

Finally, representative samples of both detritus and mangrove leaves were taken at both sites. Three composite grabs of detritus and live mangrove leaves were made at separate locations at each site for stable isotope analysis. These were placed on ice for later analysis.

Sample processing. All samples were processed at the Smithsonian Marine Station in Fort Pierce, Florida. Fish samples were immediately processed following defrosting. Dissection of tissue for stable isotope analysis involved removal of skin, scales, and associated conjunctiva. Dorsal muscle tissue located above the lateral line of the fish was filleted and placed in a lyophilizer overnight to completely dehydrate the sample (freeze-dried). The isotopic ratios found in muscle tissue correspond primarily to periods of growth and mainly reflect the diet of the organism during those periods (Perga & Gerdeaux 2005). Thus, they are most relevant to the question of how changes in *Poecilia latipinna* diets have affected observed adult body size differences.

Samples were temporarily stored in a dessicator and processed as soon as possible. Samples were ground using a clean, dry mortar and pestle until completely pulverized. Following the determination of the approximate carbon and nitrogen content of the samples, a sample weight was determined which would provide the analyzer with the proper amounts of each for isotopic fractionation. Samples were then weighed out to the nearest 0.1 mg using clean dissection tools and a microbalance. These samples were then placed into previously weighed 5 × 9 mm tin capsules, which were then sealed at the top and folded over into tiny cubes. These were tested for leakage before being placed into a labeled 96-well culture tray for later analysis by dropping against a dark surface.

Melampus coffeus samples were prepared by removing the shell and the gastrointestinal tract, and the remainder of the soft tissue was used in the analysis. Following dehydration using a lyophilizer, the samples were processed in an identical manner to *Poecilia latipinna* samples, as outlined above. Mangrove leaf and detritus samples were prepared by dehydrating a representative sample of the composite grab using the lyophilizer and then processed identically to *P. latipinna* samples.

Stable isotope analysis. Assay for stable isotope fractionation of carbon and nitrogen was completed at the Duke environmental stable isotope laboratory (Duke University). Fractionation was carried out using a Finnegan MAT Delta Plus XL mass spectrometer system using continuous-flow stable isotope mass spectrometry (Barrie & Prosser 1996). Stable isotopic fractions are reported in standard delta notation, which represents a proportional difference in fraction from a given standard (atmospheric nitrogen for $\delta^{15}\text{N}$, PeeDee Belemnite for $\delta^{13}\text{C}$). For both isotopic fractions determined, precision was ca. $\pm 0.2\%$ at 1 SD.

Analysis of results. In order to evaluate the differences observed between populations in nitrogen fractions, values for $\delta^{15}\text{N}$ were examined in a 2-way ANCOVA for all likely sources of variation, including site and sex/stage of individual (male, female, or juvenile) as dependent variables, and SL as a covariate. It was necessary to incorporate these sources (sex and maturity), as they have been observed to impact stable isotope fractions in other studies (e.g. Genner et al. 2003). By including SL as a covariate in the model, this analysis corrected for difference in stable isotope ratios stemming from size differences alone. All factors were treated as fixed effects in the model. Data groups were tested for normality using a Shapiro-Wilk test (Sall & Lehman 1996). Homogeneity of slopes (Sokal & Rohlf 1981) was determined by implementing a full factorial, 3-way ANOVA with sex, site, and SL as factors. The lack of significant interactions between SL and other factors affirmed homogeneity of slopes. A post hoc test to determine differences between sex/stage groups within sites was carried out using a T' test (Sokal & Rohlf 1981).

A similar 2-way ANCOVA analysis was used to evaluate intersite differences in $\delta^{13}\text{C}$, also including site and sex/stage of individuals (male, female, or juvenile) as dependent variables, and SL as a covariate. All factors were treated as fixed effects. Prior to running the analysis, homogeneity of slopes was tested in an identical manner to that described above for ratios and was confirmed. Differences between sex/stage groups within sites were analyzed using a post hoc T' test as for $\delta^{15}\text{N}$ values.

RESULTS

Stable isotope fractions of *Poecilia latipinna*

Stable isotope fractions of both carbon and nitrogen are shown in Table 2. In the ANCOVA, differences attributable to site were significant, with $\delta^{15}\text{N}$ values at Wabasso (open) significantly greater than those from FOS (closed) ($p < 0.05$, Table 3). The difference in mean $\delta^{15}\text{N}$ values between sites was equal to 0.52 ‰. Sex was a highly significant factor ($p < 0.0001$) in the model, while the sex by site interaction term was not significant. At both sites, mean $\delta^{15}\text{N}$ levels were highest in males, intermediate for females, and lowest in juveniles. The post hoc T' test comparing sex/stage groups between sites showed similar patterns at both sites. At both sites, male and female $\delta^{15}\text{N}$ levels were significantly greater than juveniles, but were not significantly different from each other.

Table 2. *Poecilia latipinna*, *Melampus coffeus*, mangrove leaf and detritus. Mean \pm SE of stable isotope fractions of carbon and nitrogen for items collected at Florida Oceanographic Society (FOS) (closed impoundment) and Wabasso (open). Values in **bold** indicate those that are significantly different ($p < 0.05$) from their counterpart measurements at the other site (*P. latipinna*: 2-way ANCOVA, *M. coffeus*, mangrove leaf, detritus: 1-way ANOVA). N indicates sample size

Source	N	$\delta^{15}\text{N}$ (‰)	$\delta^{13}\text{C}$ (‰)
<i>P. latipinna</i>			
Wabasso (open)	31	6.22 \pm 0.121	-26.22 \pm 0.155
FOS (closed)	21	5.70 \pm 0.188	-29.70 \pm 0.343
<i>M. coffeus</i>			
Wabasso (open)	5	2.58 \pm 0.043	-25.92 \pm 0.224
FOS (closed)	5	2.43 \pm 0.106	-27.50 \pm 0.487
Mangrove leaf			
Wabasso (open)	3	2.28 \pm 0.838	-27.14 \pm 0.433
FOS (closed)	3	0.85 \pm 0.802	-30.19 \pm 0.613
Detritus			
Wabasso (open)	3	1.56 \pm 0.345	-28.38 \pm 0.389
FOS (closed)	3	1.00 \pm 0.189	-29.28 \pm 0.243

Table 3. ANCOVA for *Poecilia latipinna*. $\delta^{15}\text{N}$ vs. site and sex with standard length (SL) as a covariate. All model effects were treated as fixed. Probabilities in **bold** are significant ($p < 0.05$)

Source	df	SS	F Ratio	Prob > F
Sex	2	7.34	9.03	0.0005
Site	1	2.04	5.02	0.0302
Site \times Sex	2	0.27	0.33	0.7181
SL	1	0.00	0.00	0.9468
Error	44	17.88		

In the ANCOVA performed for $\delta^{13}\text{C}$ differences, site was a highly significant ($p < 0.0001$) factor in the model, but neither sex nor the sex by site interaction was significant (Table 4). The $\delta^{13}\text{C}$ values from Wabasso (open) *P. latipinna* were significantly greater ($p < 0.0001$) than those for *P. latipinna* from FOS (closed), and the average difference in $\delta^{13}\text{C}$ between populations was equal to 3.48 ‰. The post hoc T' test comparing sex/stage groups within sites for $\delta^{13}\text{C}$ differences showed no significant differences between these groups within sites, although all groups from Wabasso (open) were significantly greater ($p < 0.05$) than those from FOS (closed).

Stable isotope fractions of *Melampus coffeus*

A 1-way ANOVA was used to evaluate intersite differences in carbon and nitrogen stable isotope fractions found in populations of the baseline organism *Melampus coffeus*. Values of $\delta^{15}\text{N}$ were similar at both sites, though there was greater variation present in samples from FOS (Table 2). Comparison of $\delta^{15}\text{N}$ values using a 1-way ANOVA with site as fixed effect revealed no significant difference in terms of $\delta^{15}\text{N}$ between *M. coffeus* from FOS and Wabasso ($df = 1, 9$; $SS = 0.05$; $p = 0.2365$). However, an identical 1-way ANOVA with $\delta^{13}\text{C}$ ratios as the response showed that *M. coffeus* from Wabasso had significantly greater $\delta^{13}\text{C}$ values over those from FOS ($df = 1, 9$; $SS = 6.21$; $p = 0.0188$). The mean difference between sites in $\delta^{13}\text{C}$ was on average equal to 1.58 ‰, under half of the average difference between FOS and Wabasso *Poecilia latipinna*.

Mangrove and detritus samples

Stable isotope fractions of both carbon and nitrogen measured for mangrove leaf and detritus samples are shown in Table 2. A 1-way ANOVA with site as a fixed effect showed no significant difference between sites

Table 4. ANCOVA for *Poecilia latipinna*. $\delta^{13}\text{C}$ vs. site and sex with standard length (SL) as a covariate. All effects in model were treated as fixed. Probability in **bold** is significant ($p < 0.05$)

Source	df	SS	F Ratio	Prob > F
Sex	2	4.42	1.74	0.1879
Site	1	85.35	67.12	< 0.0001
Site \times Sex	2	4.08	1.60	0.2125
SL	1	0.70	0.55	0.4629
Error	44	55.95		

in mangrove leaf $\delta^{15}\text{N}$ ($df = 1, 5$; $SS = 3.11$; $p = 0.2824$), and a 1-way ANOVA with site as a fixed effect showed the difference in $\delta^{13}\text{C}$ between sites to be significant ($df = 1, 5$; $SS = 13.99$; $p = 0.0152$).

For detritus samples, a 1-way ANOVA with site as a fixed effect showed that the difference in $\delta^{15}\text{N}$ between sites was not significant ($df = 1, 5$; $SS = 0.40$; $p = 0.2603$), and a 1-way ANOVA with site as a fixed effect showed that the difference in $\delta^{13}\text{C}$ between sites was also not significant ($df = 1, 5$; $SS = 1.23$; $p = 0.1200$).

In summary, no significant difference was observed between $\delta^{15}\text{N}$ values for detritus or mangrove leaf samples collected at Wabasso and FOS. For $\delta^{13}\text{C}$, only the differences in mangrove leaf means were significant ($p < 0.05$), which were elevated at the open site (Wabasso).

Poecilia latipinna individual samples

Individual *Poecilia latipinna* values for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ are plotted in Fig. 3. Most *P. latipinna* from Wabasso, except for 2 juveniles, are loosely clustered along $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ axes. Individuals from FOS show more variation in these parameters, especially among males. A single individual male from FOS appears to fall in the range of extreme values for Wabasso. Average values for $\delta^{15}\text{N}$ for fish from FOS would have been less had it not been for the variation observed within FOS males.

Comparison of *Melampus coffeus* and *Poecilia latipinna*

A visualization of the overall $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values for both *Poecilia latipinna* and *Melampus coffeus* at

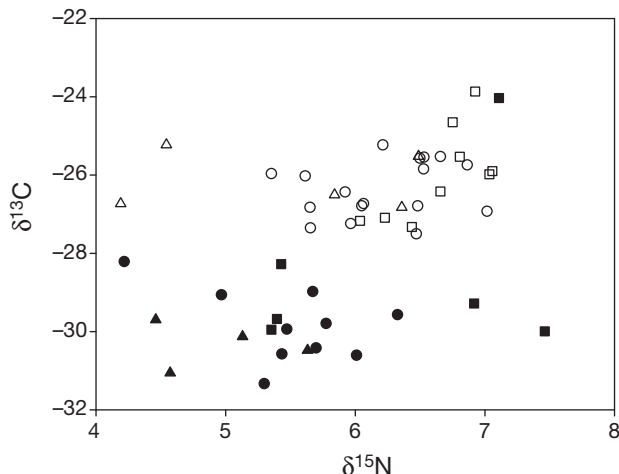


Fig. 3. *Poecilia latipinna*. Scatterplot of measured $\delta^{13}\text{C}$ (‰) vs. $\delta^{15}\text{N}$ (‰) coded by site (white markers = Wabasso [open], black markers = FOS [closed]) and sex/stage (circles = females, squares = males, and triangles = juveniles)

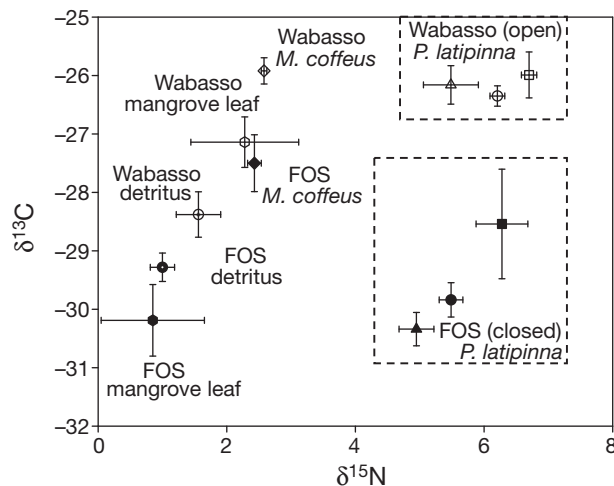


Fig. 4. Scatterplot of mean (± 1 SE) $\delta^{13}\text{C}$ (‰) and $\delta^{15}\text{N}$ (‰) of *Poecilia latipinna* grouped by sex and site, *Melampus coffeus* grouped by site, and mangrove leaves and detritus grouped by site. White symbols represent samples collected at Wabasso (open) and black symbols represent those collected at Florida Oceanographic Society (FOS) (closed). For *P. latipinna*, triangles denote juveniles, circles denote females, and squares denote males

each site is presented in Fig. 4. While *M. coffeus* are strongly differentiated between sites along the $\delta^{13}\text{C}$ axis, they are in similar, statistically identical positions along the $\delta^{15}\text{N}$ axis. *P. latipinna* from each site are also differentiated along the $\delta^{13}\text{C}$ axis in a similar manner to *M. coffeus*, although to a greater degree. The $\delta^{15}\text{N}$ values for *P. latipinna* from Wabasso (open) are shifted slightly higher than those from FOS (closed). Values of $\delta^{15}\text{N}$ for *P. latipinna* were at both sites much higher than those from *M. coffeus*. In summary, both *M. coffeus* and *P. latipinna* had intersite differences in $\delta^{13}\text{C}$, although the differences between *M. coffeus* were about half the difference observed for *P. latipinna*. The values for $\delta^{13}\text{C}$ were depleted for *P. latipinna* relative to *M. coffeus* (Fig. 4) at both sites.

DISCUSSION

Intersite stable isotope baseline comparison

The pulmonate snail *Melampus coffeus* provided a readily available organism to investigate baseline differences in carbon and nitrogen stable isotope ratios. Results indicated that $\delta^{15}\text{N}$ in *M. coffeus* at both sites was statistically identical, and there was a consistent difference in $\delta^{13}\text{C}$ between populations. Because these organisms are not likely to vary extensively in diet, are relatively long-lived, and are consumers, they most likely would reflect relative differences in ambient $\delta^{15}\text{N}$ between the 2 sites (Post 2002). One interesting

aspect of the results is that while both *Poecilia latipinna* and *M. coffeus* are mostly first-order consumers, the overall values for $\delta^{15}\text{N}$ are much lower for *M. coffeus*. In addition, the difference between *M. coffeus* populations in $\delta^{13}\text{C}$ was much lower than the difference observed between *P. latipinna* populations. In a metanalysis, Vanderklift & Posnard (2003) found that some invertebrates, particularly gastropods, appear to fractionate stable isotopes less than vertebrates. Alternatively, the results may also be explained, partially or fully, by the dietary and trophic differences between the 2 species. In any case, results indicate that there is no difference in baseline $\delta^{15}\text{N}$ between sites, and that there is a significant difference in baseline $\delta^{13}\text{C}$ between sites. This result is crucial to the interpretation of the stable isotope ratios from the 2 populations, as any relative intersite differences can be assumed to reflect intersite differences in diet.

Interpopulation differences in $\delta^{15}\text{N}$ for *Poecilia latipinna*

In summary, results show significant differences in $\delta^{15}\text{N}$ for *Poecilia latipinna* from the open (Wabasso) and closed impoundment (FOS) populations, although the interpopulation difference in $\delta^{13}\text{C}$ is much larger than that measured for $\delta^{15}\text{N}$ (Fig. 4). These results also revealed significant ontogenetic differences in these ratios within populations.

The main focus of the present study was to evaluate the hypothesis that observed patterns in body size and life history in *Poecilia latipinna* populations are due to reduced availability of animal protein (primarily mosquito larvae) as a result of impoundment. This evaluation depended strongly on the outcome of the intersite differences in $\delta^{15}\text{N}$ values. Given the equivalent baseline established by the *Melampus coffeus* samples, the expectation would be an enrichment of $\delta^{15}\text{N}$ in tissues from *P. latipinna* at the open site (Wabasso) if the hypothesis were valid and mosquito larvae had contributed to growth disproportionately. While there has been broad agreement in the literature regarding the correlation between trophic position and enrichment of $\delta^{15}\text{N}$, the degree of enrichment may not be constant across taxa. Also, other factors, such as mode of nitrogen excretion and nutritional status, may also affect it (Vanderklift & Posnard 2003). The mean difference in $\delta^{15}\text{N}$ between populations was ca. 0.52‰, a fraction of the difference expected for separation of an entire trophic level (2.96‰; Vanderklift & Posnard 2003).

The interpopulation difference in $\delta^{15}\text{N}$ suggests that the relative contribution from animal protein sources (such as mosquito larvae) to growth in *Poecilia latipinna* at the open mangrove site (Wabasso) is slightly

higher than that found in the mosquito impoundment (FOS). Perga & Gerdeaux (2005) found that muscle tissue has a relatively low level of annual variation in stable isotope ratios and that these ratios reflect the diets experienced during growth periods. In the mangrove ecosystem, the main growth periods of *P. latipinna* and the breeding period of the saltmarsh mosquito coincide and occur between the months of April and October (Snelson 1984, Smith 1988, Rey et al. 1999). Therefore, growth in these populations can be attributed primarily to diets experienced during this time, and any assimilated proteins originating from mosquito larvae as a food source would be reflected in the isotopic signature of those tissues. Any enrichment in $\delta^{15}\text{N}$ can most likely be attributed to mosquito larvae, as large broods emerge in the habitat periodically throughout this season (Brockmeyer et al. 1997). While other sources of nitrogen exist for an opportunist such as *P. latipinna*, several facts highlight the probable importance of mosquito larvae. *P. latipinna* are opportunistic predators at best and are often described as being completely herbivorous (e.g. Robins & Ray 1986). Also, the measured nitrogen content of mosquito larvae is high (10 to 12.4%; Peck & Walton 2006), relative to other potential energy sources of *P. latipinna*, such as mangrove detritus (<3% in *Rhizophora* detritus; Woitchik et al. 1997, Lugendo et al. 2006, present study). The data indicate a significant enrichment in $\delta^{15}\text{N}$ in the muscle tissue of *P. latipinna* from the open (Wabasso) site relative to those at the closed impoundment (FOS). This suggests that the relative contribution of animal protein sources at the open site during times of growth is somewhat higher than in mosquito impoundments. While not on the scale of the difference expected for an entire trophic level, the interpopulation differences measured in $\delta^{15}\text{N}$ may be important relative to the comparative life history of these populations.

The significant difference between populations in $\delta^{15}\text{N}$ is in the predicted direction of an increased intake of animal material at the open mangrove site, as suggested by Harrington & Harrington (1961, 1982). Mosquito broods in natural mangrove marsh proliferate unpredictably during the mosquito mating season, and therefore mosquito larvae comprise an irregularly timed, pulse food source for *Poecilia latipinna* populations. The fact that these pulses come during the main part of the growing season suggests that pulse inputs of mosquito larvae may be a partial contributing factor to observed interpopulation body size differences. Strong selective forces exist for both rapid growth and attainment of maturity prior to the onset of winter. Overwintering mortality is extensive in *P. latipinna* populations (Snelson 1984, Trexler et al. 1992), and survival until the next favorable reproductive period is uncertain.

Since growth in *P. latipinna* is strongly curtailed or halted following maturation (Snelson 1984), adult body size effectively approximates size at maturity. An ecological tradeoff is known to exist between larger body size and time to maturation. Larger males achieve more mating success (Ptacek & Travis 1997), and larger females produce more eggs. However, the longer it takes to achieve size at maturation, the greater the risk of predation, and therefore, a selective balance exists between survival and reproductive factors. Adult body sizes in *P. latipinna* have a large documented genetic component, with potentiality for size at maturity in males being determined by a Y-linked allele (Travis 1994). Subdivision of the mangrove system through impoundment reduced the connectivity between *P. latipinna* populations and may have also enhanced site-specific selection. Taken together, it is clear that any advantage conferred in terms of energy available during the growth season may have led to the greater body sizes seen in the open population by altering the selective balance of the ecological tradeoff in body size.

An unpredicted, consistent significant difference within populations in terms of $\delta^{15}\text{N}$ was observed between adults and juveniles (Fig. 4). Adults were consistently enriched in $\delta^{15}\text{N}$ relative to juveniles, and males were consistently enriched relative to females, though the latter difference was not significant at either site. Differences in $\delta^{15}\text{N}$ in the same direction relative to ontogeny were observed by Bouillon et al. (2002a), along with significant intersexual differences. One possible reason for these ontogenetic shifts may be related to the increase in gape size of adults relative to juveniles. This would allow for the consumption of larger prey items of increasingly higher trophic level (Cocheret de la Morinière et al. 2003). One hypothesis which can be immediately ruled out is the direct correlation of $\delta^{15}\text{N}$ with size, which has been observed in some populations (Genner et al. 2003). Within these and other *Poecilia latipinna* populations studied (Kemp 2004), females were universally larger than males. However, in the present study, females were intermediate in $\delta^{15}\text{N}$ between juveniles and males. One speculation regarding this difference may be differing physiological or allocational needs of male and female *P. latipinna*. It has been demonstrated in this system that within *P. latipinna* populations, non-reproductive females have greater length-specific mass than males (Kemp 2004), thus suggesting the possibility that males differ from females in allocation patterns.

***Poecilia latipinna* interpopulation differences in $\delta^{13}\text{C}$**

An auxiliary result to the main focus of the present study was the measurement of significant interpopula-

tion differences in $\delta^{13}\text{C}$. Because isotopic signatures of all food sources for $\delta^{13}\text{C}$ were not determined, these results could not be used to specifically identify food sources. However, there was a significant enrichment in both *Poecilia latipinna* and *Melampus coffeus* in $\delta^{13}\text{C}$ from the open mangrove site (Wabasso) versus the closed site (FOS). This may be an indication of an overall difference in carbon source between the 2 sites. Little enrichment occurs in $\delta^{13}\text{C}$ relative to trophic position, and most variation is set at the level of primary producer (DeNiro & Epstein 1978). Several other studies have shown that as distance to marine-influenced habitats (such as the IRL) increased, similar depletions in $\delta^{13}\text{C}$ were observed (Bouillon et al. 2002a, Lugendo et al. 2006). Carbon sources found in open marine systems have been found to be enriched in $\delta^{13}\text{C}$ (Fry & Sherr 1987). Pineda (2003) found that assimilation of mangrove-based carbon by organisms increased as marine influence decreased. One possible reason for this pattern is that carbon exports from other ecosystems such as seagrass may increase along with interconnectivity (Bouillon et al. 2002b, Connolly et al. 2005). Relevant to the present study, the restriction of the exchange of carbon and other materials caused by impoundment may have resulted in an increased amount of mangrove-based carbon assimilated at FOS, reflected in its isotopic signature. This suggestion is further supported by the fact that $\delta^{13}\text{C}$ signatures found in the *P. latipinna* from FOS match the relatively depleted $\delta^{13}\text{C}$ values of the mangrove leaves and detritus found there fairly closely, opposite of that found at Wabasso. Therefore, an influx of carbon sources from surrounding habitats at Wabasso appears to be a viable hypothesis to explain the consistent intersite difference in $\delta^{13}\text{C}$. In summary, the separation of the mangrove habitats from the IRL caused by impoundment may have altered mangrove food webs by increasing the importance of mangrove-based carbon sources.

CONCLUSIONS

The results of the present study have shed light on the population-level effects of impoundment of a mangrove marsh in the IRL ecosystem. Large-scale anthropogenic impacts typically have both direct and indirect effects on the ecosystem. Mosquito impoundments are no exception to this general rule. The construction of low head dams around mangrove habitat restricted tidal flushing of the area from the adjacent lagoon and movement of biota in and out of mangrove areas (Gilmore et al. 1982, Brockmeyer et al. 1997, Rey et al. 1999). The number of potential effects of impoundment on any particular population is very great, and the key to attaining understanding of an

observed effect is to isolate the primary mechanisms impacting the life history of the population (Dunham 1993). The mechanistic hypothesis proposed underlying the observed life history differences in this case made specific predictions, which were tested using stable isotope techniques. The results from the present study suggest a partial role for the effects of human habitat change (impoundment) on diet in producing observed life-history patterns in this system. The larger adult body sizes in populations from the open site (Wabasso) may have been contributed to by an increased amount of animal protein available during the growing season, as suggested by the seminal studies of Harrington & Harrington (1961, 1982). One intriguing result emerging from the present study highlights the effects that impoundment may have had on mangrove food webs. Other potential mechanisms underlying population-level effects which remain to be explored in this habitat include differential overwintering survivorship, aspects of predation pressure, and effects of resource limitation. Ultimately, the explanation for the observed interpopulation life-history differences may be multifactorial. Recognition of the importance of mangrove habitats in the IRL has led to an extensive effort in its restoration (Rey et al. 1999). While restoration efforts of mangrove ecosystems have been successful in many ways, often little attention is paid to the detailed population- and community-level information available for the ecosystem, if it exists at all (Ellison 2000). Identification of the specific effects of habitat alteration, as presented here, is an essential link between ecological science and restoration efforts in mangrove habitats.

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