VARIATION IN INVERTEBRATE DIETS AMONG WETLAND TYPES: AN APPLICATION OF STABLE ISOTOPE MIXING MODELS TO FOOD WEB COMPARISONS

by

ALANI NELSON TAYLOR

(Under the Direction of Darold P. Batzer)

ABSTRACT

Wetland function is an aspect of management that is poorly understood. Macroinvertebrates are known to have a role in these functions, but characterization of these roles is not clear, particularly for groups such as chironomid larvae and crustaceans. During 2006 and 2007, I sampled non-Tanypodinae chironomid larvae and select crustaceans from different southeastern wetland sites, along with their potential food sources. These samples were prepped for stable isotope analysis, and run for stable isotopes of carbon and nitrogen. EPA's IsoSource mixing model was used to highlight important and unlikely foods. Comparisons between forested and herbaceous wetlands within the same region suggest detrital sources are more important in forested wetlands than in herbaceous wetlands. Algae was found to be an important resource for midge larvae in all wetland types.

INDEX WORDS: stable isotope analysis, wetland food webs, chironomid larvae, crustaceans

VARIATION IN INVERTEBRATE DIETS AMONG WETLAND TYPES: AN APPLICATION OF STABLE ISOTOPE MIXING MODELS TO FOOD WEB COMPARISONS

by

ALANI NELSON TAYLOR

B.S., Cornell University, 2002

A Thesis Submitted to the Graduate Faculty of The University of Georgia in Partial Fulfillment of the Requirements for the Degree

MASTER OF SCIENCE

ATHENS, GEORGIA

2007

VARIATION IN INVERTEBRATE DIETS AMONG WETLAND TYPES: AN APPLICATION OF STABLE ISOTOPE MIXING MODELS TO FOOD WEB COMPARISONS

by

ALANI NELSON TAYLOR

Major Professor:	Darold P. Batzer
Committee:	Peter Kalla Jay Vaun McArthur

Electronic Version Approved:

Maureen Grasso
Dean of the Graduate School
The University of Georgia
December 2007

ACKNOWLEDGEMENTS

Since the beginning of my professional career, the most important lesson that I have learned is that successful science is never something that can be accomplished alone. I owe gratitude to so many people for their advice, encouraging words, hands-on help, monetary assistance, and scientific insight, all of which contributed to my completion of this project and of this chapter in my development. First and foremost, I acknowledge my advisor, Darold Batzer, for (among other things) continued encouragement through this trying experience, his gentle humor, and his manuscript critiques. My thanks to my committee Pete Kalla, Jay Vaun McArthur, and Barbara Taylor. Adrienne DeBiase, thank you for your participation out at the Savannah River Site. Broughton Caldwell, you helped make midge IDing a less daunting undertaking. I truly appreciate the insight provided by more experienced isotope minds (Tom Maddox, Chris Romanek, Seth Newsome)- your explanations were invaluable. Thanks to my fellow grad students who came to my aid in many ways (Eric Bright, Missy Churchel, Kauaoa Fraiola, Linda Lee).

TABLE OF CONTENTS

Page

CHAPTER

LIST OF TABLES

Page

LIST OF FIGURES

INTRODUCTION

Aquatic invertebrates have long been viewed as a critical link between primary producers and higher trophic levels in wetlands (Batzer and Wissinger 1996, Wissinger 1999). Chironomidae midges (Diptera) and Crustacea are often among the most abundant invertebrates in wetland ecosystems and play numerous functional roles (Golladay et al. 1999, Sharitz and Batzer 1999, Smock 1999). They contribute to decomposition and nutrient availability (Ruetz et al. 2002, Vanni 2002, Kelly et al. 2004) and are important foods for birds, fish, and other vertebrates (Pinder 1995, Euliss et al. 1999, Lemke and Benke 2003). However, these animals are often not considered in detail in community- or ecosystem-level studies (Battle and Golladay 2001). One reason that chironomids and crustaceans (particularly microcrustaceans) are sometimes overlooked is the challenge of identifying them to genus or species (Epler 2001, DeBiase and Taylor 2005).

Assessing the diets and evaluating potential feeding patterns of chironomids and crustaceans among different habitats may be a useful way to better understand wetland community dynamics. While chironomids and crustaceans as groups are considered to be trophic generalists, individual species probably serve more specific trophic roles and feeding functions, from algivory to predation. Much of the existing work on such aquatic invertebrate feeding has focused on the notion of functional feeding groups (e.g., Taylor et al. 1999, Battle and Golladay 2001, Batzer et al. 2005). Collector-gatherer is the feeding function that is most commonly attributed to chironomid larvae (Courtney et al. 1996), which means they feed predominantly on fine particulate organic matter (FPOM) (Berg 1995). Many midges are known to gather sediment-dwelling bacteria (Berg 1995). However, Caldwell et al. (1997) described *Tanytarsus* as a collector-filterer, feeding on planktonic algae or organic particulates that are suspended in the water column. Also, some Orthocladiinae taxa are labeled as scrapers, grazing matter from rocks, wood, plants, or sediments (Batzer and Resh 1991, Berg 1995). Direct feeding on macrophytes has also been observed (Berg 1995), and a few midges are wood-miners (Pinder 1995). Finally, the subfamily Tanypodinae are typically predaceous (Epler 2001). Thus, although the family consumes a broad range of foods, individual chironomid genera or species are more specialized.

Crustaceans in wetlands also consume many foods. Amphipoda are typically associated with benthic substrates. The genus *Hyalella* is known to consume epiphytes, detritus, and filamentous algae (Pickard 1996), and in some cases functions as a shredder (Thorp and Covich 1991). Warren (1996) observed *Crangonyx* consuming leaf litter and its associated microbiota. Graça et al. (2001) concluded that for some gammarid amphipods, hyphomycetes fungi were an important nutrient source. Smock and Harlowe (1983) found that living plants can sustain gammarid amphipods. Aquatic Isopoda (Asellidae) are considered detritivores. *Caecidotea* has been observed shredding CPOM (coarse particulate organic matter) (Ruetz et al. 2002). Smock and Harlowe (1983) found that smaller isopods were associated with FPOM, and functioned as collector-gatherers rather than shredders. They also suggest that *Caecidotea* may derive nutrition from living plant matter. Branchiopoda crustaceans typically exploit planktonic and benthic habitats (DeBiase and Taylor 2005). Most planktonic cladocerans are filter feeders, consuming algae, bacteria and small particles of detritus (DeBiase and Taylor 2005). Benthos-associated cladocerans are scrapers, consuming algae and FPOM (Taylor et al. 1999). Lampert (1987) and Thorp and Covich (1991) highlight the importance of small algae (1-25µm) to the diet of some cladocerans. Lemke and Benke (2003) studied three cladoceran species that were suspension feeders.

Many factors, such as quality and quantity, will influence what foods are consumed in wetlands, and it cannot be assumed that a species will always feed on the same resource. Most chironomid larvae are not restricted to a single feeding mode (Berg 1995). Some midge larvae target sediments with high organic content to maximize growth rates. It has been suggested that xylophages typically supplement their diets of wood with other higher quality resources, such as periphyton (Anderson and Cummins 1979, Pinder 1995). Shifts in feeding patterns can minimize competition in diverse communities. *Hyalella* amphipods have been shown to avoid larger leaf material, and target epiphytic fungi, algae, or bacteria when abundant (Nilsson 1974, Moore 1975). Although cladoceran crustaceans collect a wide variety of food items when filtering, they can be selective in what they actually ingest (DeBiase and Taylor 2005). As mentioned, Smock and Harlowe (1983) found that isopods can shift between feeding on FPOM and CPOM, depending on availability and age. Midges also show temporal changes in food selection from the early to late instars (Pinder 1995).

The most abundant food resources are not necessarily the most important nor the most commonly consumed. Although typically a low-quality food source, detritus may become more nutritious, and hence more important to consumers, in areas where seasonal desiccation allows for oxidation of the decomposed material (e.g., floodplain forests, Smock 1999). Kulesza and Holomuzki (2006) showed that rates of litter breakdown vary widely among macrophyte species, thereby affecting their value as food and habitat for detritivores. Habitat type and the seasonal availability of other resources often affect algae consumption (Wissinger 1999).

Biological communities and ecological processes are believed to differ between forested and herbaceous wetlands (Battle and Golladay 2001, Batzer et al. 2005). Some speculate that food webs in forested habitats are largely detritus-based, while food webs in herbaceous wetlands are driven by algae consumption (e.g., Batzer et al. 2005). Battle and Golladay (2001) compared herbaceous and forested depressional wetlands in southeastern USA and determined that marshes had more niches and a wider variety of food sources than the forested habitats. In their study, the forested wetlands were typified by high BOM (benthic organic matter) standing stocks and dissolved carbon levels, indicating a system driven by decomposition rather than primary production. Wissinger (1999) and Entrekin et al. (2001) suggest benthic algae is the driving food source in marshes. Further, others argue that algae, being often more nutritious than detritus, is a critical food source in all types of wetland habitats (Bott 1996, Goldsborough and Robinson 1996, Batzer et al. 2006).

Functional feeding group classification provides insight into what resources are ingested but it does not demonstrate what organisms are actually assimilating. Isotopic research often provides this insight. Food web and diet studies examine many aspects of isotopic information, and stable isotope analysis is an important component of this research (Bearhop 2004, McIntyre and Flecker 2006, Tillberg et al. 2006). Stable isotope analysis is a way to study nutrient flow in communities because ratios of elemental stable isotopes in tissue proteins of consumers reflect those of their food proteins in an often predictable manner (Bearhop 2004, Connolly et al. 2005, McIntyre and Flecker 2006). A common application of stable isotope analysis is to use carbon and nitrogen values of organisms to establish trophic relationships between consumers and food sources (Hecky 1995, Tillberg 2006, Layman et al. 2007). Ratios of ^{15}N to ^{14}N are used to demonstrate trophic position in community food webs, because ^{15}N values tend to be greater (more enriched) in consumers than the values in their foods, and this enrichment occurs at a consistent rate. Carbon is used to establish the basal resource of a food chain when the carbon signatures of possible resources are distinct (Bouillion 2002, Tillberg 2006). Factors determining carbon signatures include photosynthetic pathways and rates of decomposition and respiration (Boutton 1991). For example, plants with C_4 photosynthetic pathways are considerably more ^{13}C-enriched than plants with C_3 pathways. Ratios of ^{13}C to ^{12}C of consumer body tissue tend be very similar to those of their dietary sources (Bearhop 2004). One reason that stable isotope analysis is so useful is that it provides evidence about what dietary components are being assimilated (Bearhop 2004, McIntyre and Flecker 2006). Gut analysis or visual observation may demonstrate what an organism is ingesting, but cannot indicate from which food sources a consumer is deriving nutrition. Because isotopic signatures of tissues reflect the diet over time, they provide a more concrete picture of what food sources are important. More recently, mixing models have been developed to quantify source contributions in a mixture or diet (Phillips and Gregg 2003).

The objectives of this study were to use stable isotope analysis to identify important food sources for chironomids and crustaceans in wetland types common in the southeastern U.S. and to compare the diets of study organisms between forested and herbaceous wetlands with otherwise similar hydrogeologic forms. Using this information, my goal was to extrapolate the dietary habits of common aquatic invertebrates to forested and herbaceous wetlands communities in general with possible implications on changes in the community carbon bases.

STUDY SITES

The southeastern U.S.A. has an assortment of forested and herbaceous wetland habitats in both alluvial and non-alluvial settings. For this study, I selected paired forested and herbaceous wetland habitats from the Coosawhatchie River floodplain of southeastern South Carolina, Carolina bay depressional wetlands in western South Carolina, and the Okefenokee Swamp in southeastern Georgia. Each pair of forested and herbaceous sites within a region was in close proximity, which allowed for direct comparisons of consumer diets between them. The six habitats selected provided a reasonable cross-section of the range of wetland habit present in the southeastern U.S.

Coosawhatchie floodplain: The Coosawhatchie is a fourth-order blackwater river draining 1,000 km^2 of the South Carolina coastal plain (Braccia and Batzer 2001). The floodplain typically floods beginning in winter and remains partially inundated into late spring and average annual precipitation is between 127 and 152 cm. The pH of surface waters range from 6.5 to 7.3. In the Coosawhatchie floodplain sites, the forested study habitat is classified as bottomland hardwood forest and the major tree species are sweetgum (*Liquidambar styraciflua*), red maple (*Acer rubrum*), water tupelo (*Nyssa aquatica*) and various oaks. The herbaceous study area, which was logged in 1997, is located downstream of the forested site (Batzer et al. 2006). The herbaceous study area had an aquatic vegetative community of *Polygonum* knotweed and assorted emergents (*Juncus* and *Scirpus*), with sweetgum and other tree saplings beginning to regenerate.

Carolina bays: The isolated Carolina bay depressional wetlands selected for study were located on the Savannah River Site (S.R.S.) in Aiken and Barnwell counties in South Carolina. The Savannah River Site is in the Upper Atlantic Coastal Plain, which is characterized by well-drained sandy soils (DeBiase and Taylor 2005). The hydrologic cycle of these bays is dominated by precipitation and evapotranspiration, in which they fill in late autumn or early winter and dry in late spring or early summer. The importance of groundwater depends on the seasonal depth of the water table. The waters tend to be acidic (pH range: 3.5-6.5) and soft, with little seasonal variation.

The forested Carolina bay (Bay 118) was about 1 ha and was dominated by sweetgum with some assorted mixed pine (*Pinus palustris*, *P. taeda*). The herbaceous Carolina bay (Bay 5204) was about 0.5 ha and had stands of *Typha* along the perimeter, with several aquatic (*Nymphaea odorata*, *Polygonum*) and emergent macrophytes (*Panicum*, *Scirpus*) present in the interior. The habitat had also been logged 3 years before my study, and some residual wood remained and a few saplings were present.

Okefenokee Swamp: The Okefenokee Swamp is a large (1,754 km^2) southern blackwater swamp. Water inputs to the Okefenokee are primarily through precipitation and outputs are evapotranspiration and surface water outflow (Loftin 1997). Most of the wetland area is inundated throughout the year, although during some periods, the cypress domes scattered across the Okefenokee can have large dry surface patches. The water is dark and acidic, with a pH range of 3.8 to 4.7. Here, a representative cypress dome and a lily prairie habitat were chosen in Chesser Prairie, on the east side of the Okefenokee National Wildlife Refuge. The cypress dome was dominated by baldcypress (*Taxodium distichum*), while the lily prairie was dominated by fragrant water lily (*Nymphaea odorata*).

METHODS

Sample collecting and processing: I conducted the field components of the study from January 2006 to March 2007. Sampling was done at least twice in each habitat to incorporate potential seasonal or other temporal variation. Trips were scheduled when surface water levels were suitable for access and sampling. I sampled the Coosawhatchie on 14 February 2006 and 14 March 2007, the Carolina bays on 7 March 2006 and 17 April 2006, and the Okefenokee on 15 January 2006, 14 May 2006, and 13 March 2007. On each collection date, two or three replicate samples of consumer organisms and potential food sources were taken from each habitat (however, only one sample set was collected from the Okefenokee on 15 January 2006). I collected non-Tanypodinae chironomid larvae from all six wetlands, and the crustacean consumers that were most common in each of the three areas. I collected amphipods and isopods from the Coosawhatchie floodplain, cladocerans from the Carolina bays, and amphipods from the Okefenokee sites (however, amphipods were rare in the lily prairie, so data on amphipods from the Okefenokee are not presented).

Invertebrates were collected by sweeping a D-frame aquatic net (1 mm mesh, 30 cm diameter) through the water column, through aquatic plant beds, and along benthic substrate. The gathered material was placed in large buckets for field sorting of invertebrates, which were extracted with droppers and forceps and placed in scintillation vials with a small amount of water. Where present, samples of submerged wood were brought back to the lab to be searched for additional midge larvae, which were added to the field collected samples. Some authors suggest keeping organisms alive for a time to allow for gut clearing, but others maintain this is unnecessary (Jardine et al. 2005). Other researchers at the University of Georgia have found that gut clearing of midge larvae does not occur without continuous feeding (unpublished data). Thus, I did not attempt gut clearing for the organisms in this study.

At the same time as invertebrate collections, I collected potential food sources for the invertebrates as available, including leaf litter, macrophyte detritus, macrophyte new growth, epiphytic algae, periphyton, sediment, submerged wood, and phytoplankton. Macrophytes were rinsed to remove and collect epiphyton, and the macrophytes were later identified. I initially tried to sample benthic algae on clay tile substrates but this procedure failed and thus benthic algae were considered as a component of the sediment samples. Sediment samples were collected from beneath any litter layer. Wood samples were separated into outer bark and inner wood fractions. All food source samples were placed in labeled plastic bags or glass jars, stored on ice in the field, and then kept frozen in the laboratory until processing.

In the laboratory, invertebrate and food source samples were thawed, cleaned, freeze-dried, pulverized to a fine powder in a ball-mill grinder, and weighed (at least 1.5 mg of substance) into tin capsules in preparation for analysis of carbon and nitrogen isotopes (as recommended by the Analytical Chemistry Laboratory). Before grinding, leaf litter samples were separated and categorized by species. Detrital samples, indistinguishable decomposed plant fragments, were categorized, for the purposes of this study, as FPOM (particulates passing through a 1-mm mesh) or CPOM (particulates retained on a 1-mm mesh). This was done to account for the potential importance to invertebrates of microbial detrital conditioning. Sediment samples, placed in silver capsules, were acid-rinsed with 20% HCl to remove inorganic carbon, oven-dried overnight, reweighed, and analyzed as the other samples. The invertebrate samples and other samples for which only a small amount of matter could be obtained were ground in vials using a glass rod.

Analysis of carbon and nitrogen was done by the Analytical Chemistry Laboratory (Institute of Ecology, University of Georgia) using a Carlo Erba NA 1500 CHN analyzer (Carlo Erba Instrumentazione, Milan, Italy) coupled to a Finnigan Delta C isotope radio mass spectrometer (Thermo Electron, Waltham, MA) operating as a continuous flow system. The laboratory error rate is ±0.15‰. The reproducibility of each sample run was monitored by using a bovine liver standard and a poplar leaf standard. This analysis provided the stable isotope content of my samples, as well as the percent content of total carbon and nitrogen. The stable isotope data were expressed as relative difference per mil (‰) using the equation:

$$\delta X=[(R_{\text{sample}}/R_{\text{standard}}) - 1]\times 1{,}000$$

where $X={}^{13}C$ or ${}^{15}N$, and $R={}^{13}C/{}^{12}C$ or ${}^{15}N/{}^{14}N$. The ratios of ${}^{13}C$ to ${}^{12}C$ are expressed relative to the standard PDB (Pee Dee Belemnite). The ratios of ${}^{15}N$ to ${}^{14}N$ are expressed relative to N_2 in air.

Isotope and Diet Analysis: For each collection at a site, carbon and nitrogen stable isotope ratio data were first graphed to view the range of food and consumer values, as well as any sample grouping. Then I used the IsoSource mixing model (Phillips 2001), developed by the U.S. Environmental Protection Agency, to estimate the relative contribution of potential food resources to the invertebrate diet (midge or crustacean) for each sampling date. The IsoSource software is based on isotopic mass balance conservation. The program calculates the range of feasible proportional contributions of sources to a mixture (expressed in some small increment and totaling 100%) when the number of sources is too large to permit a unique solution. The user enters the isotopic data (e.g., atom %, isotopic ratios) for the mixture (e.g. the consumer isotopic data) and for each of the sources, and determines the increment value and mass balance tolerance value (e.g. within ± 0.1‰ of the observed mixture value) for each analysis. IsoSource's output files list each feasible solution combination, descriptive statistics about the distribution of these solutions (number of solutions, mean, standard deviation, minimum, maximum, 1st percentile, median, and 99th percentile for each source), and histograms of these distributions.

Using guidelines in Bunn and Boon (1993), I considered sources to be potential food for invertebrates if they had $\delta^{13}C$ values within the range of 2‰ less than or 1‰ greater than the mean consumer signature, and had $\delta^{15}N$ values within the range of 1-5‰ less than the consumer signature. Sources in this range were evaluated using the IsoSource model. I provided the model with $\delta^{15}N$ and $\delta^{13}C$ values for food (i.e., source entries) and consumer (i.e., mixture entry) samples. The entered values were replicate means. To account for trophic isotope fractionation (resulting from consumer digestion and assimilation), I subtracted 2.0‰ from the $\delta^{15}N$ value and 1.0‰ from the $\delta^{13}C$ value of my consumer sample (personal communication, S. Newsome). Source increment was set at 2% and I used a mass balance tolerance value of 0.1‰. If under these parameters a solution could not be calculated, I increased the tolerance, incrementally by 0.1 to 1.0, and if still unsuccessful, I concluded that the model could not be used to analyze that consumer data set.

Using the IsoSource output histograms for each data set run, sources with low maximum values (i.e. the source was never >20% of the mixture in feasible solutions) were assumed to be either unlikely or minor components of the consumer diet. Sources with wide distributions were assumed to be present in the diet, but their significance (or importance) was unclear. Likely major food sources were those where the source proportion was greater than 20% in more than

50% of solutions, and where none of generated solutions showed a 0% contribution. The output values are expressed in text as the 1st and 99th percentile range followed by the 50th percentile value as compiled by the mixing model. For example, if the IsoSource solution summary for a source were (20-50%: 30%), it would mean that 98% of the solutions indicated the food item contributed between 20% and 50% of the consumer diet, and that for the median solution, 30% was contributed to the diet. Additional site comparisons of food source and consumer interactions were made using total carbon and nitrogen data.

Taxonomic Confirmations: In the laboratory, a sub-sample of the midges from each site was slide-mounted for taxonomic identification. Each sub-sample consisted of at least 20 individuals. I used Epler (2001) for identifications, which were then verified by Dr. Broughton Caldwell (GAEPD, retired). Crustacean genera were identified using standard keys (Pennak 1989), and species of cladocerans in the Carolina bays were confirmed by Adrienne DeBiase (Savannah River Ecology Laboratory).

RESULTS

COOSAWHATCHIE FLOODPLAIN

Midge, Amphipods, and Isopod Taxonomy: On the Coosawhatchie floodplain, *Polypedilum tritum* was the major midge taxon in both the herbaceous (74%) and forested (76%) habitat in February 2006 (Appendix). In March 2007, *P. tritum* was again the most abundant midge in both habitats (43% of the forested community and 91% of the herbaceous community). At that time, the forested community was richer than the herbaceous community, with an additional seven taxa comprising the remaining 57% of the community. *Caecidotea* isopods (Asellidae) and *Crangonyx* amphipods (Crangonyctidae) were abundant in both habitat types and both dates.

Carbon and Nitrogen Analyses in Invertebrates: Midges, amphipods, and isopods in the Coosawhatchie floodplain had a $\delta^{13}C$ range of -30.48 to -25.86‰ and $\delta^{15}N$ range from 2.61 to 7.66‰ (Figure 1A). Forested samples were consistently more ^{13}C-enriched than corresponding herbaceous samples. Invertebrates collected from the forested floodplain had more varied isotopic signatures than those from the herbaceous area and isotopic signatures among invertebrates were more variable in March 2007 than in February 2006. Amphipods had the highest $\delta^{15}N$ values.

Carbon and Nitrogen Analyses of Food Resources: Potential food resources from the Coosawhatchie floodplain had total C compositions between 5.3 and 78.9% and total N compositions from 0.3% to 4.8%. Wood and detrital food samples tended to have the highest C:N ratios, and epiphyton and macrophyte samples had the lowest ratios (Table 1). Most of the potential food samples at the Coosawhatchie had a range of -32.5 to -25‰ for $\delta^{13}C$ values and a range of -0.5 to 5.0‰ for $\delta^{15}N$ values.

Linking Invertebrates to their Foods: In samples from February 2006, carbon signatures of midges in both the forested and herbaceous habitats were within the range of the $\delta^{13}C$ values of the potential food sources (Figure 2A-B). Amphipods and isopods in the forested habitat were most similar to wood in $\delta^{13}C$ value. In the herbaceous habitat, the amphipod carbon signature was very similar to the midge signature and within the range of the potential food resources.

In March 2007, midge $\delta^{13}C$ values were again similar to many of the potential food sources (Figure 2C-D). However, carbon values for amphipods from the forested floodplain were closest to cypress bark values, and isopod carbon values were most similar to sweetgum leaf litter. In the herbaceous habitat, both crustaceans had $\delta^{13}C$ values in the same range as many potential foods.

IsoSource analyses for the forested habitat in February 2006 suggest that midges were primarily consuming tupelo leaf litter (84-98%: 90%; refer back to the Methods for information on interpreting these values), while CPOM (0-6%: 0%), epiphyton (0-10%: 2%), sweetgum leaf litter (0-14%: 2%), and sediment (0-12%: 2%) were unlikely foods. In March 2007, sweetgum leaf litter (60-78%: 70%) and epiphyton (10-38%: 24%) were identified as major components, while sweetgum wood (0-8%: 2%) and FPOM (0-12%: 2%) were not likely foods.

For midges from the herbaceous floodplain in February 2006, *Micranthemum* mudflower (58-74%: 66%) and epiphyton (18-36%: 28%) were identified as likely foods, and sweetgum leaf litter (0-10%: 2%) and sediment (0-8%: 2%) were unlikely or minor sources. In March 2007, IsoSource analyses indicated that sediment (68-78%: 72%) was a very likely food item and periphyton (22-30%: 26%) was also important. Sweetgum leaf litter (0-4%: 0%) and phytoplankton (0-2%:0%) were not likely food sources for midges.

Mudflower (6-68%: 62%) was identified as being important in the amphipod diet in the herbaceous habitat in February 2006. Non-tanypod midges (28-38%: 34%) were also important foods for amphipods in all solutions. Sweetgum leaf litter (0-4%: 0%), epiphyton (0-12%: 2%), and sediment (0-2%: 0%) were unlikely foods. In the herbaceous habitat in March 2007, IsoSource analyses for amphipods yielded an output range suggesting a mixture of foods with none being dominant [sweetgum leaf litter (0-34%: 8%), knotweed (0-42%: 10%), phytoplankton (0-36%: 8%), epiphyton (2-40%: 26%), *Scirpus* bulrush (2-26%: 16%), non-Tanypod midges (0-82%: 22%)]. For the forested sample collected in February 2006, midges were the major food source (74-94%: 82%), for amphipods, while sweetgum leaf litter (0-20%: 4%), sediment (0-18%: 4%), CPOM (0-16%: 4%), and epiphyton (0-10%: 2%) were unlikely foods. IsoSource could not generate solutions for amphipods from the March 2007 collection.

For isopods in the forested habitat in February 2006, wood (72-74%: 74%) and sediment (16-28%: 24%) were likely food sources, and sweetgum (0-10%: 2%), tupelo (0-2%: 0%), and oak (0-0%: 0%) leaf litter, epiphyton (0-2%: 0%), and detrital CPOM (0-2%: 0%) were not likely foods. In March 2007, sweetgum leaf litter (0-56%: 12%), epiphyton (0-42%: 10%), bark (0-40%: 8%), and inner gum wood (0-50%: 10%) were possible dietary components for isopods, but their importance was equivocal. Knotweed (20-48%: 34%) was shown to be a likely food for isopods in the forested habitat. No IsoSource solutions were generated for isopods in the herbaceous habitat during March 2007.

CAROLINA BAY DEPRESSIONAL WETLANDS

Midge and Cladoceran Taxonomy: In the Carolina bays, *Polypedilum* spp. midge larvae dominated both the forested (96%) and herbaceous (56%) bays in March 2006, although the herbaceous bay also supported a number of other genera (Appendix). In April 2006, both bays supported more diverse and evenly distributed midge assemblages. At that time, *Psectrocladius psilopterus* sp. 3 was the most abundant midge in the forested bay (46%) followed by *Dicrotendipes modestus* (23%). The cladocerans in the forested bay were *Simocephalus expinosus*, and *S. expinous* and *S. serrulatus* occurred in the herbaceous bay.

Carbon and Nitrogen Analyses of Midges and Cladocerans: The Carolina bays showed a wide range in consumer isotopes, with $\delta^{13}C$ values from -32.89 to -23.11‰ and $\delta^{15}N$ values from 0.82 to 4.71‰ (Figure 1B). Within each bay, invertebrate samples collected in March 2006 were more enriched in ^{13}C than April 2006 collections. Cladoceran variation was pronounced, while midge samples from the herbaceous bay exhibited modest variation in $\delta^{13}C$ values. Midge samples from the forested bay exhibited some variation between replicates in April, and pronounced variation in March. Samples of both cladocerans and midges from the herbaceous bay were less variable than those from the forested bay.

Carbon and Nitrogen Analyses of Food Resources: Potential invertebrate foods from Carolina bays had total carbon ranging from 17.0 to 50.7% and total nitrogen ranging from 0.16 to 3.38%. Wood samples had the highest C:N ratio, with other detrital materials (leaf litter, particulate organic matter) usually having high C:N ratios (Table 1). Other potential foods had lower ratios than detritus, but were otherwise similar to each other. Algae as a category (e.g. epiphyton, metaphyton) had no consistent C:N range.

Stable isotope results of Carolina bay organic matter indicated most of the source $\delta^{13}C$ values were between -30 and -26‰ and most $\delta^{15}N$ results were in between 2.0 and 4.5‰. Detrital and sediment samples had similar stable isotope values between sampling dates, while algae and macrophytes varied temporally.

Linking Midges and Cladocerans to their Foods: Midge $\delta^{13}C$ values in the forested bay in March 2006 were similar to many potential foods (Figure 3A). In the herbaceous bay, the mean midge $\delta^{13}C$ values were between those of the metaphyton and epiphyton samples (Figure 3B). Stable isotope values for cladocerans in the March forested sample were dissimilar from any collected food source, and the herbaceous cladoceran isotope values were distant from all food samples except *Nymphaea* lily (Figure 3A-B).

In April 2006, midges continued to have isotope values within the ranges of the potential food sources in the forested habitat (Figure 3C). In the herbaceous bay, mean midge isotope values deviated from many potential foods (Figure 3D). Cladocerans in both the forested and herbaceous bays had isotope values in similar ranges of the potential food sources (Figure 3C-D).

For midges, IsoSource analyses from the forested bay in March 2006 indicated that FPOM (48-60%: 54%) and detrital sweetgum fruit (40-52%: 44%) were likely food sources, while sediment (0-2%: 0%) was not. In April 2006, IsoSource modeling identified epiphyton (74-96%: 82%) as being the principal midge food in the forested bay. Sweetgum bark (0-12%: 2%), CPOM (0-14%: 2%), sweetgum leaf litter (0-16%: 4%), and sediment (0-20%: 6%) were unlikely foods.

For midges in the herbaceous bay in March 2006, metaphyton (64-76%: 70%) was identified as the only important food source. Sediment (0-14%: 8%) and decomposed lily (0-18%: 8%) were unlikely sources and the importance of epiphyton (0-34%: 14%) could not be determined. For midges in the herbaceous bay during April, no feasible solutions could be calculated.

For cladocerans, IsoSource could not generate solutions for populations occurring in March 2006 for either the forested or herbaceous bay, but there were solutions for populations sampled in April 2006. In the forested bay, sediment (74-86%: 78%) and sweetgum wood (14-26%: 20%) were identified as likely food sources for cladocerans, while epiphyton (0-4%: 0%) and *Panicum* grass litter (0-2%: 0%) were unlikely foods. However, because cladocerans are physically unable to consume wood, the sweetgum result was considered an anomaly (personal communication, B. Taylor). In the herbaceous bay, epiphyton (2-78%: 42%) was present in virtually all feasible IsoSource solutions, but the distribution was wide, and thus importance cannot be inferred. Contributions from pine needle litter (0-34%: 8%), FPOM (0-34%: 10%), sediment (0-54%: 16%), and sweetgum bark (0-60%: 18%) also could not be interpreted.

OKEFENOKEE SWAMP

Midge Taxonomy: Midge genera collected in January 2006 were not identified. In May 2006, the midge community in the lily prairie (6 genera) was richer than in the cypress dome (4 genera), but both were dominated by the same taxon, *Chironomus* sp. (67% of the lily prarie community, and 52% of the cypress dome community) (Appendix). In March 2007, the communities were similarly rich, but dissimilar in character. Different genera dominated each habitat; *Polypedilum trigonus* comprised 42% of the lily prairie community and *Paratendipes* sp. comprised 47% of the cypress dome community.

Carbon and Nitrogen Analyses of Midges: The isotope ratio scale for midges in the Okefenokee Swamp was -31.48 to -25.34‰ for δ^{13}C values and 1.48 to 2.46‰ for δ^{15}N values. All of the separate samples were distinct from each other in their signatures (Figure 1C). All of the midge samples occurred across a narrow δ^{15}N range. Samples from lily prairies were always more ^{13}C- enriched than from the corresponding cypress dome samples.

Carbon and Nitrogen Analyses of Food Resources: Potential food categories collected from the Okefenokee Swamp had total carbon values between 31.9% and 54.7%, and total nitrogen values from 0.4% to 6.3%. The lily prairie sample set had a narrower range of both total carbon and total nitrogen than the cypress dome set. This was due to the fact that wood found in the cypress dome consistently had very low total nitrogen values (and had the highest C:N ratios). Samples not derived from wood in the cypress dome collections were in the range of the lily prairie sample sets. Epiphyton and sediment samples tended to have the lowest C:N ratios (Table 1).

Stable isotope analysis of Okefenokee sites showed most of the $\delta^{13}C$ values of potential foods were in the range of -30 and -24‰ and the majority of $\delta^{15}N$ values were between -1.8 and 1.2‰. Although stable isotope values of potential foods in each habitat shifted temporally, there was no consistent pattern of change within any of the food categories.

Linking Midges to their Foods: In general, the isotopic signatures of midges from the Okefenokee habitats were similar in $\delta^{13}C$ to a number of the potential foods and had higher $\delta^{15}N$ values (Figure 4A-F). However, in May 2006, midge signatures were lower than much of the source set, and matched few potential foods (Figure 4C-D). Here, midges in the cypress dome had $\delta^{13}C$ values similar to those of bladderwort, and in the lily prairie had $\delta^{13}C$ values similar to lily.

In the forested cypress dome, IsoSource analyses for the January 2006 samples indicated that cypress wood (34-52%: 44%) was a sizable component of all calculated solutions, while cypress bark (0-20%: 4%) was an unlikely food. Epiphyton (0-44%: 14%), cypress needle litter (0-22%: 6%) and titi leaf litter (0-40%: 24%), and sediment (0-22%: 6%) were indicated in small amounts. For the May 2006 analyses, knotweed (22-50%: 38%) was indicated in moderate amounts in all solutions, and bladderwort (0-52%: 14%) and epiphyton (0-40%: 20%) were present to some degree. Periphyton (0-12%: 2%) was an unlikely food. In March 2007, sediment (94-100%: 96%) was indicated as a major food source, while CPOM (0-4%: 0%), and bladderwort (0-4%: 0%), epiphyton (0-6%: 2%) were unlikely.

In the lily prairie, sediment (90-100%: 96%) was a very likely food source of midges in January 2006, while lily (0-10%: 2%), decomposed lily (0-2%: 0%), bladderwort (0-2%: 0%), and *Carex* sedge (0-2%: 0%) were all unlikely. The IsoSource model could not generate solutions for the May 2006 sample set in the lily prairie. In March 2007, phytoplankton (72-98%: 90%) appeared to be the dominant food for the midge, while peat (0-6%: 2%), decomposed lily (0-4%: 0%), and lily (0-4%: 0%) were unlikely foods; sediment (0-26%: 6%) was indicated in small amounts in the majority of solutions.

DISCUSSION

Invertebrates play a central role in wetland food webs (Wissinger 1999), and thus information on their diets contributes to understanding the overall ecosystem functioning of wetlands. An ongoing debate exists about whether wetland food webs are primarily detrital- or algal-based (Batzer and Wissinger 1996, Batzer et al. 2006). Because of the copious plant growth in wetlands, and the fact that most of this material enters wetland foodwebs as detritus, many believe that wetlands are detrital-based systems (e.g., Battle and Golladay 2001, Leslie et al. 1999). However, others argue that detritus is a poor quality food, and many invertebrates consume higher quality algae instead, and thus wetlands are algal-based systems (Bunn and Boon 1993, Batzer and Wissinger 1996). My study suggests that this dichotomy is an oversimplification, and depending on the habitat both detritus and algae can be important to wetland food webs. Below I discuss invertebrate diets in each of the three wetland systems that I studied, and then conclude with a more general synthesis.

Coosawhatchie Floodplain: The midges, isopods, and amphipods that occurred on the Coosawhatchie floodplain are all reported to have fairly broad food habits. *Polypedilum tritum* was the most abundant midge at the Coosawhatchie, and is known to exhibit flexibility in food consumption (Berg 1995, Golladay et al. 1999, Smock 1999). Opsahl (2006) used stable isotope analysis to show that *Caecidotea* isopods rely on a mix of basal resources. *Crangonyx* amphipods have been categorized as shredders and gatherers, which suggests consumption of detritus, but have also been observed preying on midge larvae (Thorp and Cothran 1984).

My analyses of invertebrate diets at the Coosawhatchie also suggest a range of diets, although a recurring theme was the presence of algae (Table 2). For midges, epiphyton was important in both herbaceous and forested habitats, and periphyton and sediment (of which epipelic algae was probably a component) was important in the herbaceous habitats. However, macrophytes were also important. Mudflower (which had a low C:N ratio) was important in the herbaceous habitat, and leaf litter (sweetgum and tupelo) was important in the forested habitat. *Caecidotea* were found to have consumed wood, among other resources. *Crangonyx* also consumed mudflower, but, supporting the findings of Thorp and Cothran (1984), were also apparently consuming midges. While the variability in solutions and a lack of solutions for organisms in some collections hinders generalization, the herbaceous portion of the Coosawhatchie tended to be algal-based and the forested area tended to be detrital-based.

Carolina Bays: The midge communities from the Carolina Bays showed an obvious shift in species richness between sampling dates. Still, the dominant midge taxa from each collection (*Chironomus*, *Dicrotendipes*, *Polypedilum*, *Psectrocladius*) are reported by others to consume a variety of foods and to be associated with a variety of microhabitats (Berg 1995, Golladay et al. 1999, Kelly et al. 2004, Goedkoop et al. 2006). *Simocephalus* cladocerans are considered suspension feeders and generally as algivores (Wiggins et al. 1980, Lemke and Benke 2003).

Results for the Carolina bay midge larvae again indicate consumption of algal resources in both herbaceous and forested habitats (Table 2). In the herbaceous bay, metaphyton was the only source clearly linked with midges. In the forested bay, FPOM (a detrital component) and sweetgum fruit were present in solution sets. However, later in the hydroperiod (April), epiphyton was a dominant food source. This seasonal variation might reflect shifts in consumer selection or nutritional needs, but in most cases (except sweetgum fruit), the indicated foods had low C:N ratios, indicating high nutritional quality. I had anticipated that *Simocephalus* cladocerans would be consuming algae, but the minimal overlap of the carbon signatures with any of the foods sampled in March suggests a failure to collect their specific foods. However, in April, cladocerans were linked to epiphyton in the herbaceous bay. In the forested bay, sediment was important, in addition to other sources. Elsewhere, benthic algae have been reported as important resources for cladocerans (Stevenson et al. 1996).

Okefenokee Swamp: There was seasonal variation in the midge community, but the common taxa in both the cypress dome (*Chironomus*, *Paratendipes*), and the lily prairie (*Chironomus*, *Polypedilum trigonus*) were, again, taxa that are known to use a variety of resources. *Chironomus* are known to consume green algae, as well as detritus (Berg 1995, Kelly et al. 2004). *Paratendipes* was abundant in the cypress dome in March 2007, and has been noted as a detritivore elsewhere (Ward and Cummins 1979, Berg 1995).

In the lily prairie, phytoplankton was important to midges in March. Sediment matter was important at times in solutions for both habitats (Table 2). In each instance, the sediment had a relatively high nitrogen content, and thus may be rich in microbes or algae. Detrital foods (wood, macrophytes) were moderately important in the cypress dome. Kratzer and Batzer (2007) maintain that wetland invertebrate communities in the Okefenokee are dominated by generalists that are able to tolerate wide ranges in habitat conditions. Perhaps they are also dietary generalists. Porter et al. (1999) describe Okefenokee systems as algal-based, with the majority of food web carbon originating from consumption of microbial production, algae, and living plants, rather than detritus. There is weak support for this statement in my results.

Synthesis: In this study, invertebrate consumer signatures were usually within the carbon range of food samples, suggesting that the majority of potential foods for study organisms were sampled. As expected (Tillberg 2006), invertebrate ^{15}N levels placed them one trophic level above basal resources (the high levels for amphipods suggest they were functioning in part as predators). Results generated with IsoSource mixing models reflect wide ranging diets, with evidence of herbivory, algivory, detritivory, and carnivory. In many instances, solutions could not be generated by IsoSource, possibly because the food source of that consumer was not sampled or because samples were insufficiently pure to develop unique signatures. Alternatively, a lack of resolution may suggest that many foods were being consumed simultaneously.

Food habits of an organism tended to change depending on whether the taxon was residing in herbaceous or forested habitat (Table 2). In all three wetland types (Coosawhatchie floodplain, Carolina bays, Okefenokee Swamp), invertebrates in the herbaceous habitat relied heavily on algal food resources, with macrophytes supplementing the diets. In the forested areas, detritus was more important, but in most cases, algae were supplementing diets. Wood was occasionally consumed in forested habitat (Okefenokee midges, Coosawhatchie isopods), even though it usually had the highest C:N ratio of all samples. These results in general support the notion that forested wetlands are detrital-based and herbaceous wetlands are algal-based. However, it should be recognized that overall patterns were weak. At times, solutions could not be generated, and even when a food item was identified, it was usually only for a single collection period. It could instead be argued that the wetland invertebrates in this study were opportunistic, generalist feeders, able to use a wide range of foods in a wide range of habitats, and they simply consumed foods as available.

REFERENCES

Anderson, N. H. and K. W. Cummins. 1979. The influences of diet on the life histories of aquatic insects. Journal of the Fisheries Research Board of Canada. 36:335-342.

Battle, J. M. and S. W. Golladay. 2001. Water Quality and Macroinvertebrate Assemblages in Three Types of Seasonally Inundated Limesink Wetlands in southwest Georgia. Journal of Freshwater Ecology 16:189-207.

Batzer, D. P. and V. H. Resh. 1991. Trophic interactions among a beetle predator, a chironomid grazer, and periphyton in a seasonal wetland. Oikos 60:251-257.

Batzer, D. P. and S. A. Wissinger. 1996. Ecology of insect communities in nontidal wetlands. Annual Review of Entomology 41:75-100.

Batzer, D. P., S. E. Dietz-Brantley, B. E. Taylor, and A. E. DeBiase. 2005. Evaluating regional differences in the macroinvertebrate communities from forested depressional wetlands across eastern and central North America. Journal of North America Benthological Society 24:403-414.

Batzer, D. P., R. Cooper, and S. A. Wissinger. 2006. Wetland Animal Ecology. p. 242-284. *In* D. P. Batzer and R. R. Sharitz (eds.) Ecology of Freshwater and Estuarine Wetlands. University of California Press, Berkeley, CA, USA.

Bearhop, S., C. E. Adams, S. Waldron, R. A. Fuller, H. Macleod. 2004. Determining trophic niche width: a novel approach using stable isotope analysis. Journal of Animal Ecology 73:1007–1012.

Berg, M. B. 1995. Larval food and feeding behaviour. p. 136–168. *In* P. D. Armitage, P. S. Cranston, and L. C. V. Pinder (eds.) The Chironomidae: the biology and ecology of non-biting midges, Chapman and Hall, London, UK.

Bott, T. L. 1996. Algae in Microscopic Food Webs. p. 574-608. *In* R. J. Stevenson, M. L. Bothwell, and R. L. Lowe (eds.) Algal Ecology: Freshwater Benthic Ecosystems, Academic Press, San Diego, CA, USA.

Bouillon, S., N. Koedam, A. V. Raman, and F. Dehairs. 2002. Primary producers sustaining macro-invertebrate communities in intertidal mangrove forests. Oecologia 130:441–448.

Boutton, T. W. 1991. Stable Carbon Isotope Ratios of Natural Materials: II. Atmospheric, Terrestrial, Marine, and Freshwater Environments. In D. C. Coleman and B. Fry (eds.) Carbon Isotope Techniques, Academic Press, San Diego, CA, USA.

Braccia, A. and D. P. Batzer. 2001. Invertebrates associated with woody debris in a southeastern U.S. forested floodplain wetland. Wetlands 21:18-31.

Bunn, S. E. and P. I. Boon. 1993. What sources of organic carbon drive food webs in billabongs? A study based on stable isotope analysis. Oecologia 96:85-94.

Caldwell, B. A., P. L. Hudson, D. R. Lenat, and D. R. Smith. 1997. A revised annotated checklist of the Chironomidae (Insecta: Diptera) of the Southeastern United States. Transactions of the American Entomological Society 123:1-53.

Connolly, R. M., D. Gorman, and M. A. Guest. 2005. Movement of carbon among estuarine habitats and its assimilation by invertebrates. Oecologia 144:684-691.

Courtney, G. W., H. J. Teskey, R. W. Merritt, and B. A. Foote. 1996. Aquatic Diptera. Part one. Larvae of aquatic Diptera. p. 484-548. *In* R. W. Merritt and K. W. Cummins (eds.) An introduction to the aquatic insects of North America (third edition). Kendall/Hunt Publishing Company, Dubuque, IA, USA.

DeBiase, A. E. and B. E. Taylor. 2005. Microcrustaceans (Branchiopoda and Copepoda) of Wetland Ponds and Impoundments on the Savannah River Site, Aiken, South Carolina. Savannah River Ecology Laboratory, National Environmental Research Park Program, U.S. Department of Energy, Aiken, SC, USA. SRO-NERP-28.

Dietz-Brantley, S. E., B. E. Taylor, D. P. Batzer, and A. E. DeBiase. 2002. Invertebrates that aestivate in dry basins of Carolina bay wetlands. Wetlands 22:767-775.

Entrekin, S. A., S. W. Golladay, and D. P. Batzer. 2001. The influence of plant community on chironomid secondary production in two wetland types: cypress-gum swamps and grass-sedge marshes. Hydrobiologia 152:369-394.

Epler, J. H. 2001. Identification Manual for the Larval Chironomidae (Diptera) of North and South Carolina. North Carolina State Department of Environmental Water Quality, Raleigh, NC, USA.

Euliss, N. H., Jr. and D. M. Mushet. 1999. Inverterbrates in Wetlands of the Prairie Pothole Region-Species Composition, Ecology, and Management. p. 471-514. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Goedkoop, W., N. Åkerblom, and M. H. Demandt. 2006. Trophic fractionation of carbon and nitrogen stable isotopes in *Chironomus riparius* reared on food of aquatic and terrestrial origin. Freshwater Biology 51:878-886.

Golladay, S. W., S. Entrekin, and B. W. Taylor. 1999. Forested limesink wetlands of Southwest Georgia: invertebrate habitat and hydrologic variation. 197-216. *In* D. Batzer, R. B. Rader, and

S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Goldsborough, L. G. and G. G. C. Robinson. 1996. Pattern in Wetlands. p. 78-120. *In* Stevenson, R. J., M. L. Bothwell, and R. L. Lowe (eds.) Algal Ecology: Freshwater Benthic Ecosystems, Academic Press, San Diego, CA, USA.

Graça, M. A. S., C. Cressa, M. O. Gessner, M. J. Feio, K. A. Callies, and C. Barrios. 2001. Food quality, feeding preferences, survival and growth of shredders from temperate and tropical streams. Freshwater Biology 46:947-957.

Grey, J., A. Kelly, and R. I. Jones. 2004. High intraspecific variability in carbon and nitrogen stable isotope ratios of individual lake chironomid larvae. Limnology and Oceanography 49:239–244.

Hecky, R. E., and R. H. Hesslein. 1995. Contributions of benthic algae to lake food webs as revealed by stable isotope analysis. Journal of the North American Benthological Society. 14:631–653.

Jardine, T. D., R. A. Curry, K. S. Heard, R. A. Cunjak. 2005. High fidelity: isotopic relationship between stream invertebrates and their gut contents. Journal of the North American Benthological Society 24:290-299.

Kelly, A., R. I. Jones, J. Grey. 2004. Stable isotope analysis provides fresh insights into dietary separation between *Chironomus anthracinus* and *C. plumosus*. Journal of the NorthAmerican Bentholoigcal Society 23:287-296.

Kratzer, E. and D. P. Batzer. 2007. Spatial and temporal variation in aquatic macroinvertebrates in the Okefenokee Swamp, Georgia, USA. Wetlands 27:127-140.

Kulesza, A. E. and J. R. Holomuzki. 2006. Amphipod performance responses to decaying leaf litter of *Phragmites autralis* and *Typha angustifolia* from a Lake Erie coastal marsh. Wetlands 26:1079-1088.

Lampert, W. 1987. Feeding and Nutrition in *Daphnia*. p. 143-192. *In* R. H. Peters and R. DeBernardi (eds.) *Daphnia*, Memorie dell'Istituto Italiano di Idrobiologia 45. Pallanza: Istituto Itanliano di Idrobiologia.

Layman, C. A., D. A. Arrington, C. G. Montana, D. M. Post. 2007. Can Stable Isotope Ratios Provide for Community-wide Measures of Trophic Structure? Ecology 88:42-48.

Lemke A. M. and A. C. Benke. 2003. Growth and reproduction of three cladoceran species from a small wetland in the southeastern USA. Freshwater Biology 48:589–603.

Leslie, A. J., J. P. Prenger, and T. L. Crisman. 1999. Cypress Domes in North Florida: Invertebrate Ecology and Response to Human Disturbance. p. 105-120. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Loftin, C. S. 1997. Okefenokee swamp hydrology. Proceedings of the 1997 Georgia Water Resources Conference, held 20-22 March 1997, at the University of Georgia, K. J. Hatcher (ed.), Institute of Ecology, The University of Georgia, Athens, GA, USA.

McIntyre, P. B. and A. S. Flecker. 2006. Rapid turnover of tissue nitrogen of primary consumers in tropical freshwaters. Oecologia 148:12-21.

Moore, J. W. 1975. The role of algae in the diet of *Asellus aquaticus* L. and *Gammarus pulex* L. Journal of Animal Ecology 44:719–729.

Nilsson, L. M. 1974. Energy budget of a laboratory population of *Gammarus pulex* (Amphipoda). Oikos 25:35–42.

Opsahl, S. P. and J. P. Chanton. 2006. Isotopic evidence for methane-based chemosynthesis in the Upper Floridian aquifer food web. Oecologia 150:89-96.

Pennak, R. W. 1989. Freshwater Invertebrates of the United States, third edition. John Wiley & Sons, Inc., New York, NY, USA.

Phillips, D. L. and J. W. Gregg. 2001. Uncertainty in source partitioning using stable isotopes. Oecologia 127:171-179.

Phillips, D. L. and J. W. Gregg. 2003. Source partitioning using stable isotopes: coping with too many sources. Oecologia 136:261-269.

Pickard D. P. and A. C. Benke. 1996. Production Dynamics of *Hyallela azteca* (Amphipoda) among Different Habitats in a Small Wetland in the Southeastern USA. Journal of the North American Benthological Society 15:537–550.

Pinder, L. C. V. 1995. The habitats of chironomid larvae. p. 107–135. In P. Armitage, P. S. Cranston, and L. C. V. Pinder (eds.) The Chironomidae: the biology and ecology of non-biting midges. Chapman and Hall, London, UK.

Porter, K. G., A. Bergstedt, and M. C. Freeman. 1999. The Okefenokee Swamp: invertebrate communities and foodwebs. 121-135. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Rader, R. B. 1999. The Florida Everglades: natural variation, invertebrate diversity, and foodweb stability. 25-54. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Ruetz III, C. R., R. M. Newman, and B. Vondracek. 2002. Top-down control in a detritus-based food web: fish, shredders, and leaf breakdown. Oecologia 132:307-315.

Sharitz, R. R. and D. P. Batzer. 1999. An introduction to freshwater wetlands in North America and their invertebrates. p. 1-22. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Smock, L. A. and K. L. Harlowe. 1983. Utilization and Processing of Freshwater Wetland Macrophytes by the Detritivore *Asellus forbesi*. Ecology 64:1556-1565.

Smock, L. A. 1999. Riverine Floodplain Forests of the Southeastern United States: Invertebrates in an Aquatic-terrestrial Ecotone. p. 137-166. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Stevenson, R. J. 1996. An Introduction to Algal Ecology in Freshwater Benthic Habitats. p. 3-30. *In* R. J. Stevenson, M. L. Bothwell, and R. L. Lowe (eds.) Algal Ecology: Freshwater Benthic Ecosystems, Academic Press, San Diego, CA, USA.

Taylor, B. E., D. A. Leeper, M. A. McClure, and A. E. DeBiase. 1999. Carolina Bays: ecology of aquatic invertebrates and perspectives on conservation. 167-196. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

Thorp, J. H. and M. L. Cothran. 1984. Regulation of freshwater community structure at multiple intensities of dragonfly predation. Ecology 65:1546-1555.

Thorp, J. H. and A. P. Covich (eds.). 1991. Ecology and Classification of North American Freshwater Invertebrates. Academic Press, San Diego, CA, USA.

Tillberg, C. V., D. P. McCarthy, A. G. Dolezal, and A. V. Suarez. 2006. Measuring the trophic ecology of ants using stable isotopes. Insectes Sociaux 53:65-69.

Vanni, M. J. 2002. Nutrient cycling by animals in freshwater ecosystems. Annual Review of Ecology and Systematics 33:341–370.

Ward, G. M. and K. W. Cummins. 1979. Effects of food quality on growth rate and life history of *Paratendipes albimanus* (Meigen) (Diptera: Chironomidae). Ecology 60:57-64.

Warren, P. H., and M. Spencer. 1996. Community and Food-Web Responses to the Manipulation of Energy Input and Disturbance in Small Ponds. Oikos 75:407-418.

Wiggins, G. B., R. J. Mackay, and I. M. Smith. 1980. Evolutionary and ecological strategies of animals in annual temporary pools. Archiv für Hydrobiologie Supplement 58:97-206.

Wissinger, S. A. 1999. Ecology of wetland invertebrates: synthesis and application for conservation and management. p. 1043-1086. *In* D. Batzer, R. B. Rader, and S. A. Wissinger (eds.) Invertebrates in Freshwater Wetlands of North America-Ecology and Management. John Wiley and Sons, Inc., New York, NY, USA.

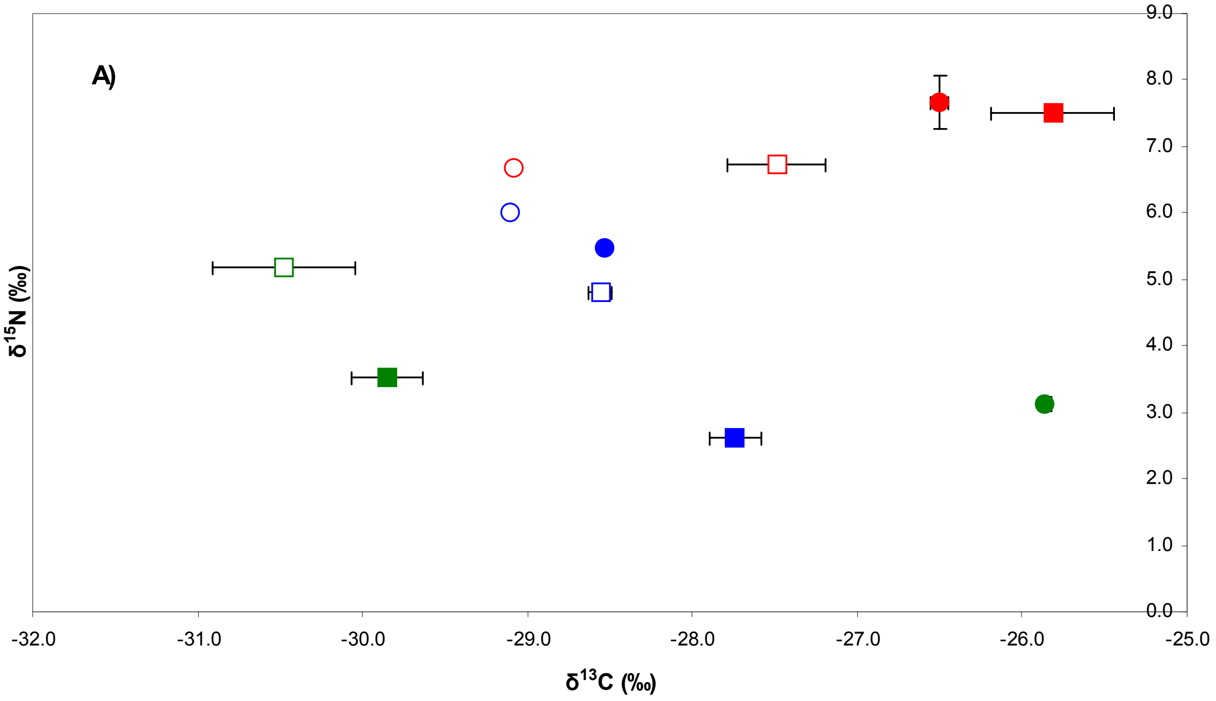
Appendix. Non-Tanypodinae community composition in sampled sites

Coosawhatchie Floodplain	**February**	**2006**	**March**	**2007**
	Forested	**Herbaceous**	**Forested**	**Herbaceous**
Apedilum			5%	
Chironomus sp.	4%	9%	10%	
Limnophyes			10%	
Micropsectra sp.				5%
Parametriocnemus		4%		5%
Polypedilum sp.			5%	
Polypedilum tritum	76%	74%	43%	91%
Psectrocladius (Monopsectrocladius) sp.		4%	5%	
Pseudosmittia	8%		19%	
Tanytarsus	12%	9%	5%	

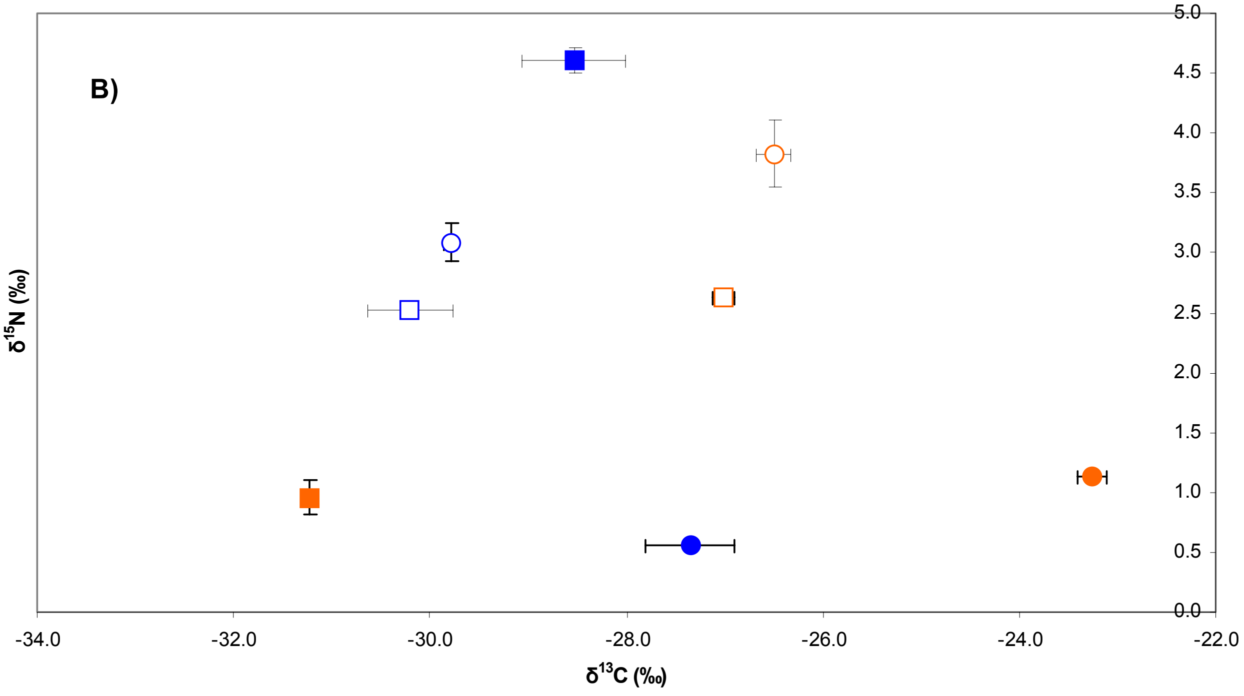
Carolina Bays	**March**	**2006**	**April**	**2006**
	Forested	**Herbaceous**	**Forested**	**Herbaceous**
Ch. (*Lobochironomus*) *austini*				30%
Chironomini genus III Epler	4%			
Chironomus decorus		2%	3%	33%
Chironomus sp.		2%	7%	7%
Dicrotendipes modestus		2%	23%	3%
Glyptotendipes testaceus				3%
Kiefferulus sp.			3%	
Micropsectra sp.		2%		
Nanocladius alternantherae		2%		3%
Paratanytarsus sp.		6%	3%	
Polypedilum spp.	96%	56%	7%	17%
Psectrocladius (Monopsectrocladius) sp.		10%		3%
Psectrocladius octomaculatus		2%		
Psectrocladius psilopterus sp. 3		8%	47%	
Tanytarsus sp. G		8%	7%	

Okefenokee Swamp	**May**	**2006**	**March**	**2007**
	Cypress	**Lily**	**Cypress**	**Lily**
Chironomini genus III Epler	39%	6%	3%	
Chironomus sp.	52%	67%	20%	4%
Cladopelma				25%
Kiefferulus dux	4%	11%		
Larsia berneri		6%		
Parachironomus chaetoalus complex	4%			
Paratendipes			47%	4%
Polypedilum illinoense grp.			17%	8%
Polypedilum sp.		6%		
Polypedilum spp.				
Polypedilum trigonus			10%	42%
Polypedilum tritum		6%		
Psectrocladius (Monopsectrocladius) sp.			3%	
Tanytarsus				17%

Figure 1. Stable isotope ratios for consumers in study habitats. A) Coosawhatchie floodplain, B) Carolina bays, C) Okefenokee Swamp. Each data point is a mean value of the sample. The values are isotope ratios relative to a standard (air for $\delta^{15}N$, PDB for $\delta^{13}C$). Circles represent first field collection, squares represent second field collection, triangles represent third collection (which only applies to the Okefenokee samples). Larval non-Tanypodinae midge data points are blue, amphipod points are red, isopod points are green, and cladoceran points are orange. Forested samples are indicated with filled data points, and herbaceous samples are indicated with unfilled data points. Error bars, indicating 1 standard error, are visible when they are larger than the data points.



A)
δ15N (‰)
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
-32.0
-31.0
-30.0
-29.0
-28.0
-27.0
-26.0
-25.0
δ13C (‰)



B)
δ15N (‰)
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-34.0
-32.0
-30.0
-28.0
-26.0
-24.0
-22.0
δ13C (‰)

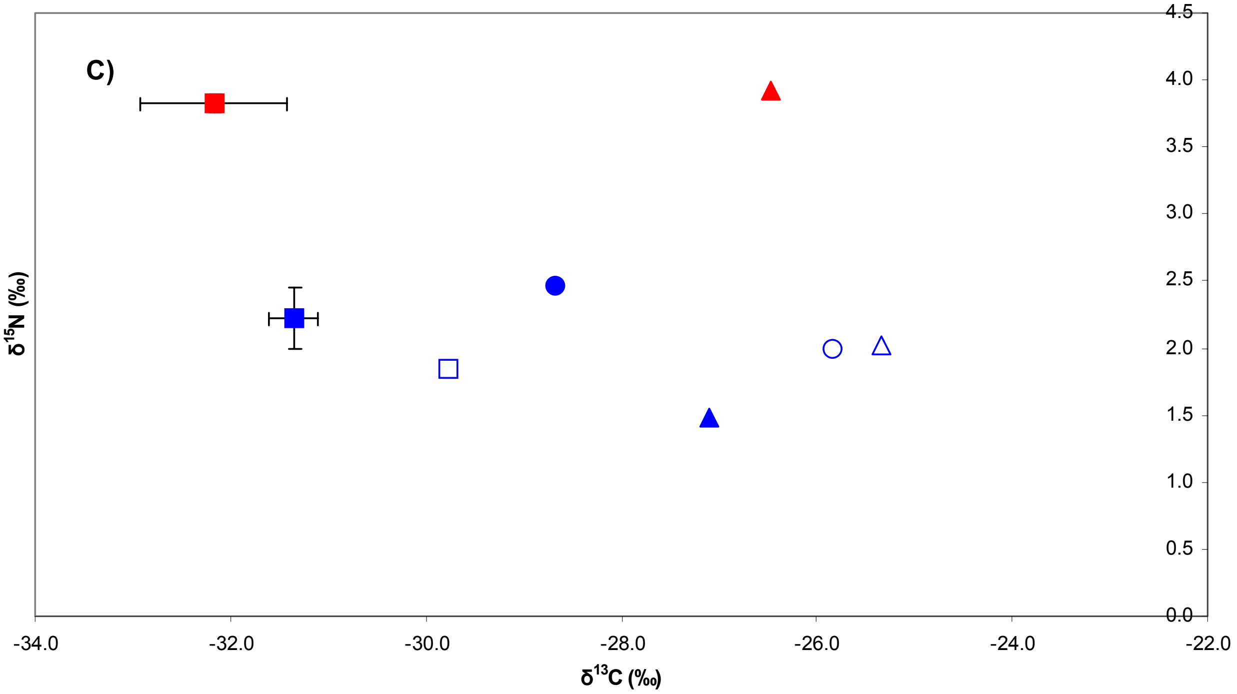
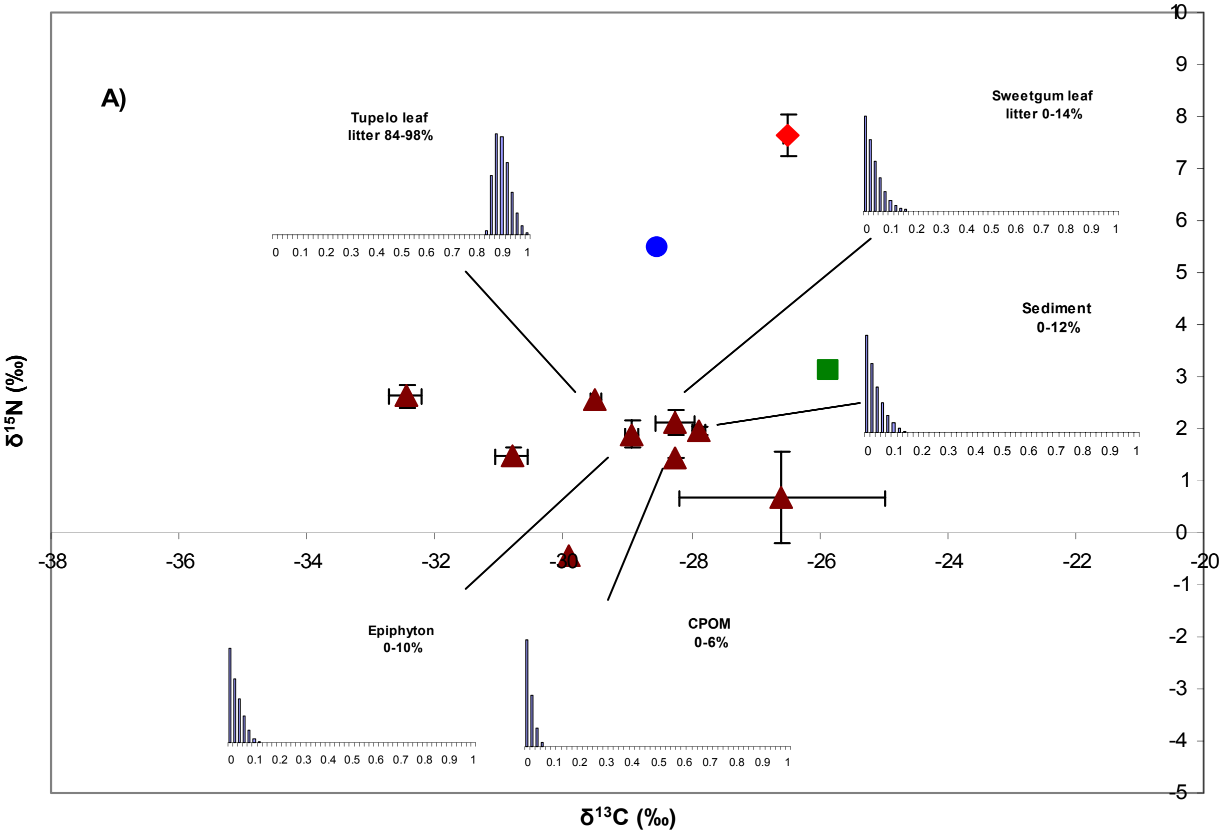
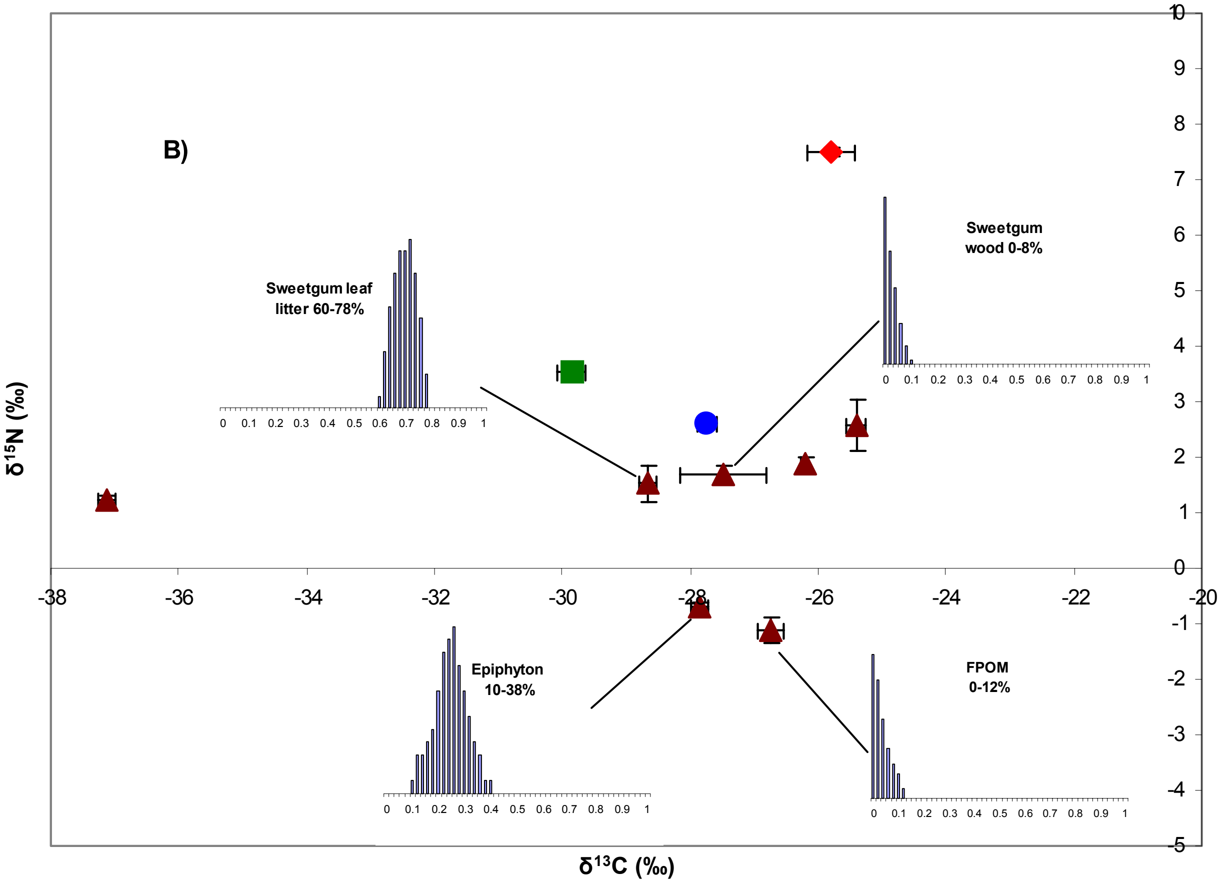
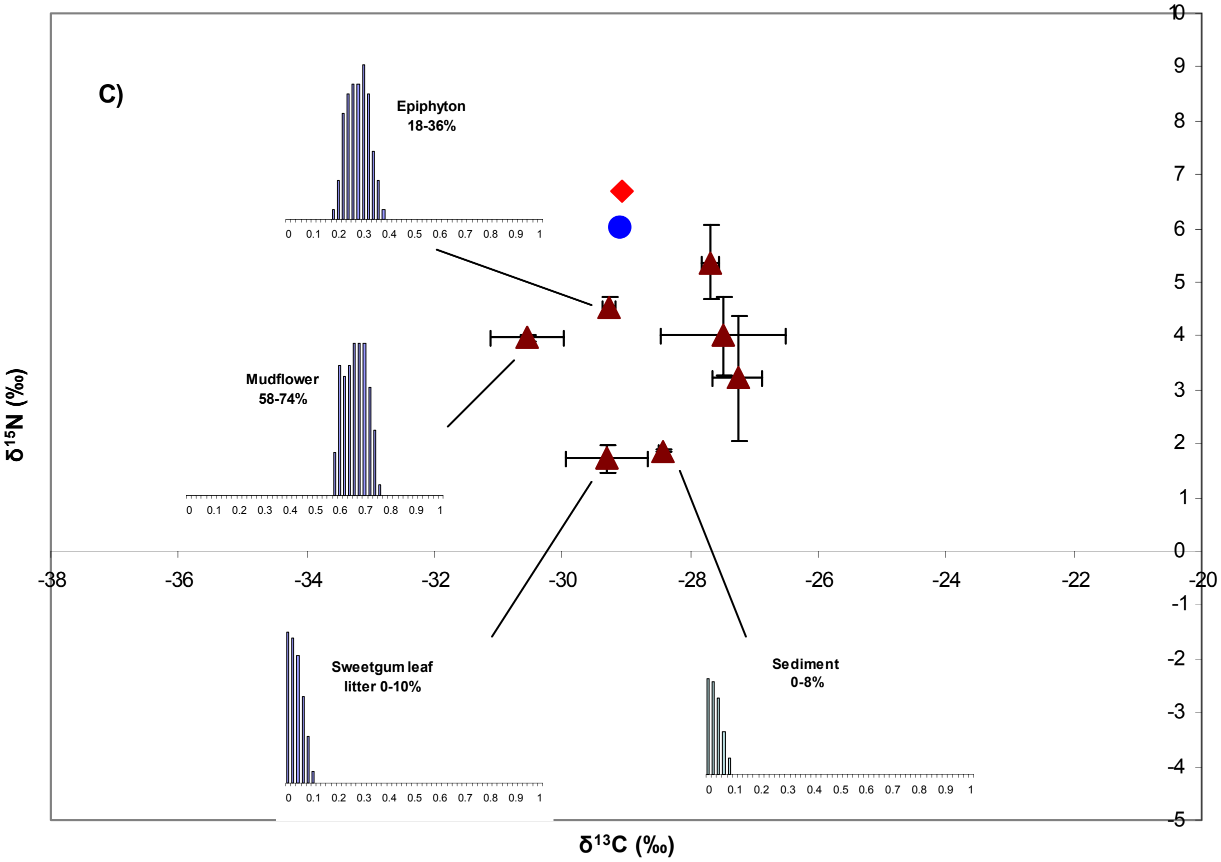
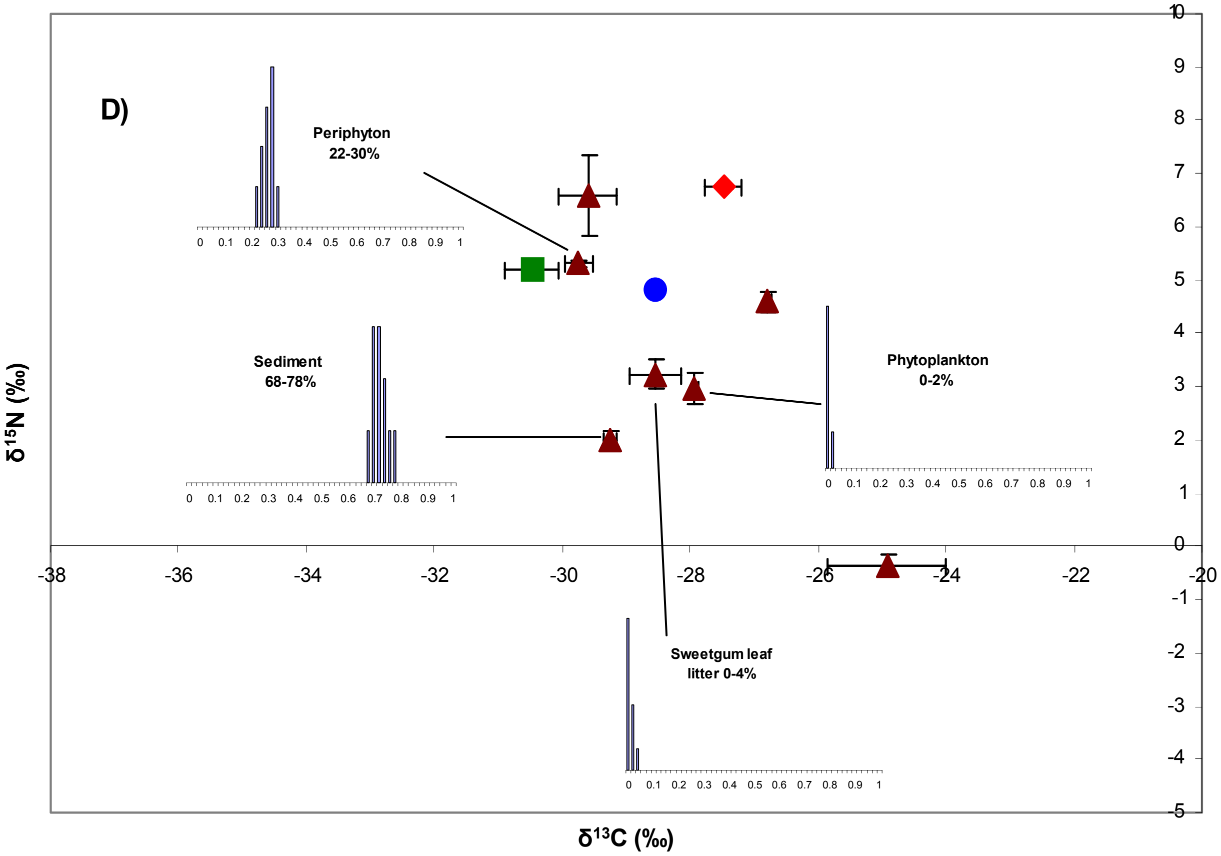

C)
δ15N (‰)
δ13C (‰)
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-34.0
-32.0
-30.0
-28.0
-26.0
-24.0
-22.0

Figure 2. Stable isotope biplots of Coosawhatchie communities. A) Forested February 2006 set, B) Forested March 2007 set, C) Herbaceous February 2006 set, D) Herbaceous March 2007 set. Each data point is the mean of the sample replicates. The isotope values are ratios relative to a standard (air for $\delta^{15}N$, PDB for $\delta^{13}C$). Circles symbolize midge larvae; diamonds symbolize amphipods; squares symbolize isopods; and triangles symbolize sampled food sources. Error bars, indicating 1 standard error, are visible when they are larger than the data points. Sources within parameters described in Bunn and Boon (1993) were analyzed using IsoSource. The histograms provide the distribution of feasible IsoSource contributions from these sources to the midge diet. Values shown in the boxes are the 1-99 percentile ranges for these distributions.


A)
Tupelo leaf litter 84-98%
Sweetgum leaf litter 0-14%
Sediment 0-12%
Epiphyton 0-10%
CPOM 0-6%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
δ15N (‰)
δ13C (‰)
-38 -36 -34 -32 -30 -28 -26 -24 -22 -20
10 9 8 7 6 5 4 3 2 1 0 -1 -2 -3 -4 -5

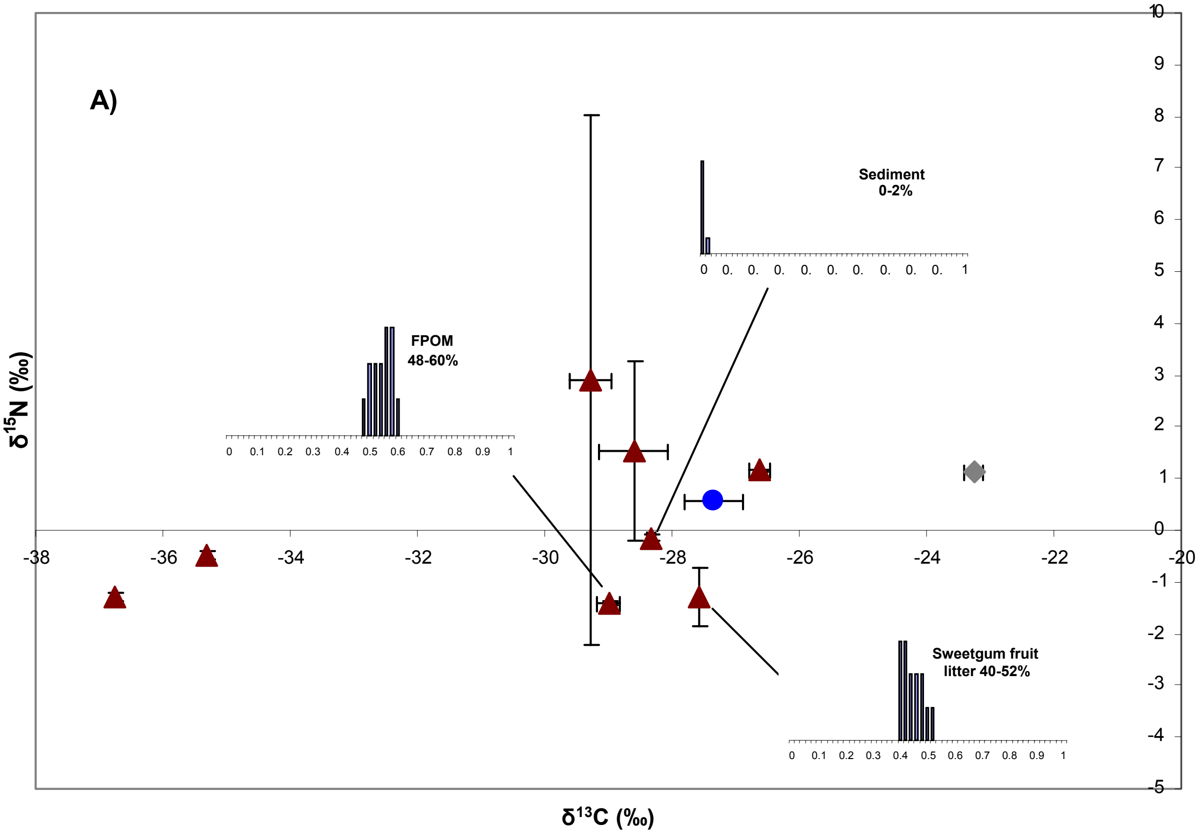

B)
Sweetgum leaf litter 60-78%
Sweetgum wood 0-8%
Epiphyton 10-38%
FPOM 0-12%
δ15N (‰)
δ13C (‰)
-38
-36
-34
-32
-30
-28
-26
-24
-22
-20
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
1

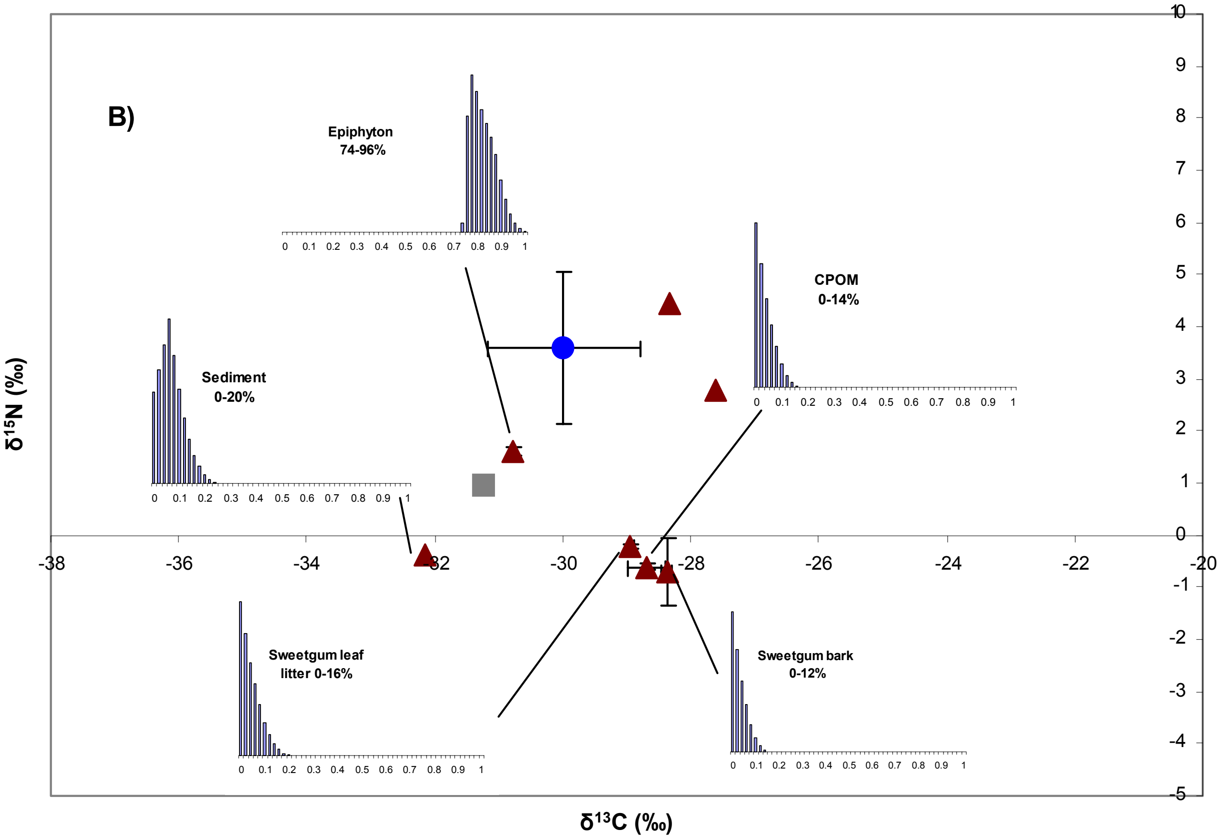

C)
Epiphyton
18-36%
Mudflower
58-74%
Sweetgum leaf
litter 0-10%
Sediment
0-8%
δ¹⁵N (‰)
δ¹³C (‰)
-38
-36
-34
-32
-30
-28
-26
-24
-22
-20
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1

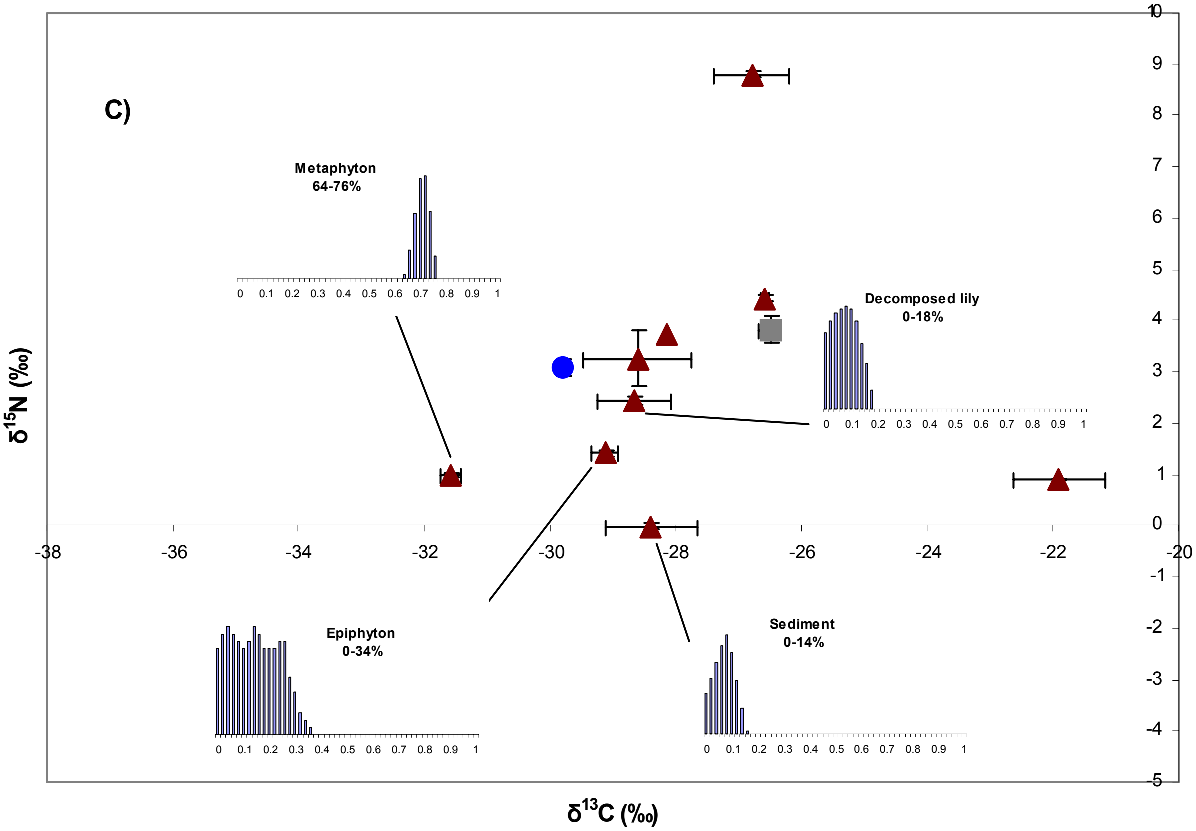


D)
Periphyton
22-30%
Sediment
68-78%
Phytoplankton
0-2%
Sweetgum leaf
litter 0-4%
δ¹⁵N (‰)
δ¹³C (‰)
-38
-36
-34
-32
-30
-28
-26
-24
-22
-20
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1

Figure 3. Stable isotope biplots of Carolina bay communities. A) Forested March 2006 set, B) Forested April 2006 set, C) Herbaceous March 2006 set, D) Herbaceous April 2006 set. Each data point is the mean of the sample replicates. The isotope values are ratios relative to a standard (air for $\delta^{15}N$, PDB for $\delta^{13}C$). Circles symbolize midge larvae; squares symbolize cladocerans; and triangles symbolize sampled food sources. Error bars, indicating 1 standard error, are visible when they are larger than the data points. Sources within parameters described in Bunn and Boon (1993) were analyzed using IsoSource. The histograms provide the distribution of feasible IsoSource contributions from these sources to the midge diet. Values shown in the boxes are the 1-99 percentile ranges for these distributions. (There were no solutions, and thus no histograms shown for Figure 3D.)


A)
δ15N (‰)
δ13C (‰)
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5
-38
-36
-34
-32
-30
-28
-26
-24
-22
-20
FPOM
48-60%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
Sediment
0-2%
0 0. 0. 0. 0. 0. 0. 0. 0. 0. 1
Sweetgum fruit
litter 40-52%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1


B)
Epiphyton
74-96%
CPOM
0-14%
Sediment
0-20%
Sweetgum leaf
litter 0-16%
Sweetgum bark
0-12%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
δ15N (‰)
δ13C (‰)
-38 -36 -34 -32 -30 -28 -26 -24 -22 -20
10 9 8 7 6 5 4 3 2 1 0 -1 -2 -3 -4 -5


C)
Metaphyton
64-76%
Decomposed lily
0-18%
Epiphyton
0-34%
Sediment
0-14%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
δ^{15}N (‰)
δ^{13}C (‰)
-38 -36 -34 -32 -30 -28 -26 -24 -22 -20
10 9 8 7 6 5 4 3 2 1 0 -1 -2 -3 -4 -5

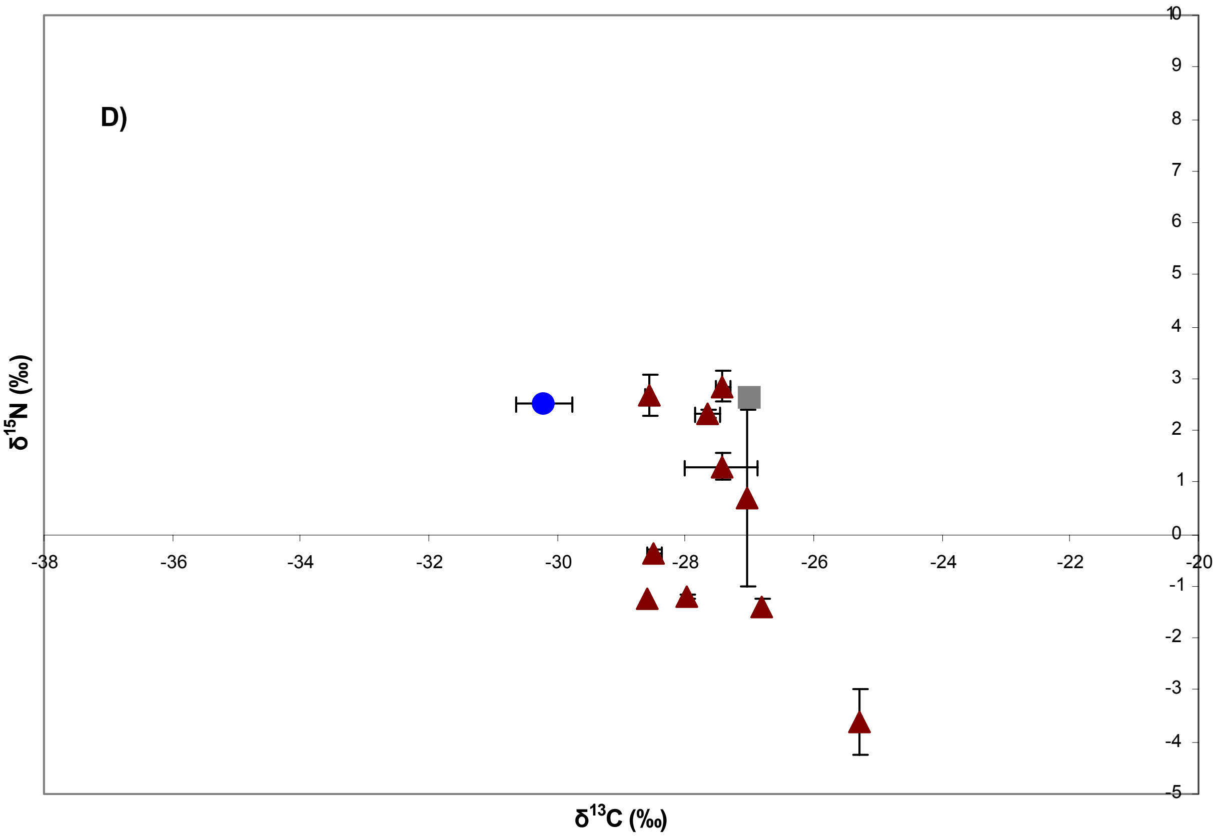
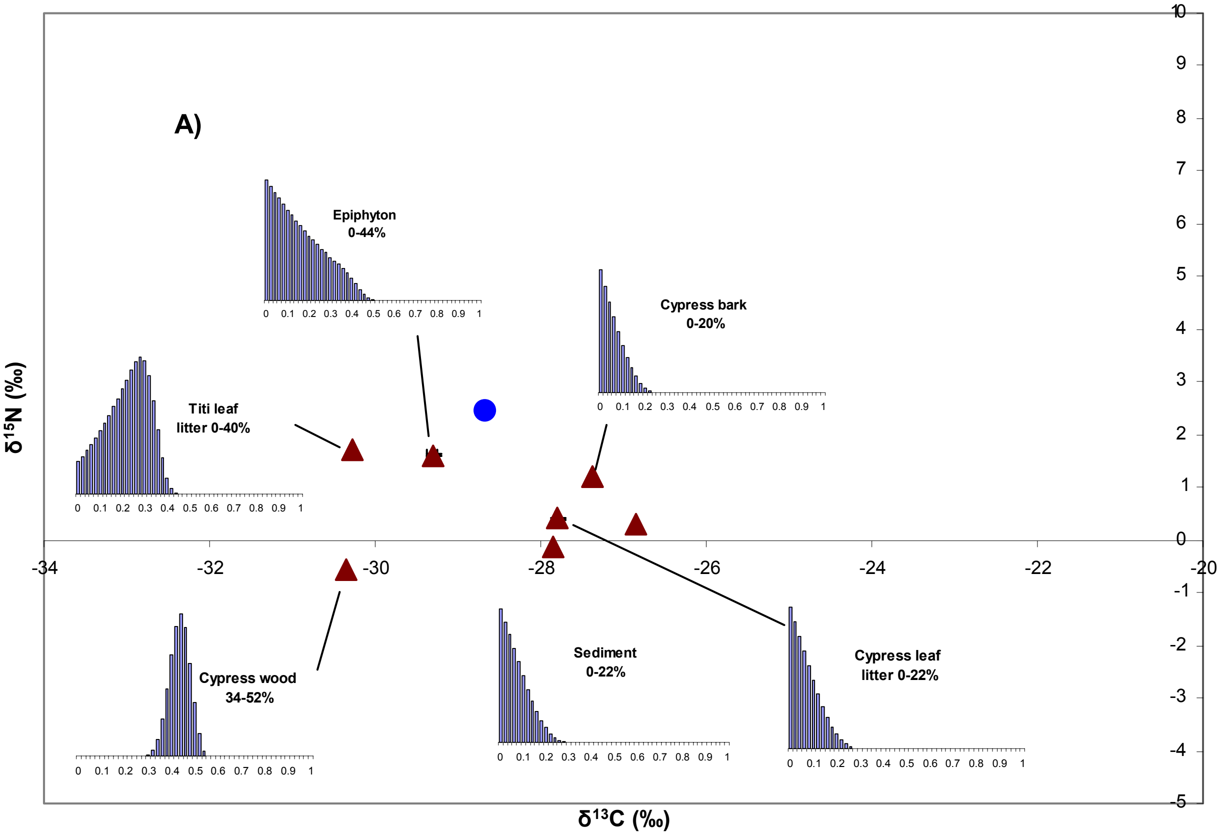
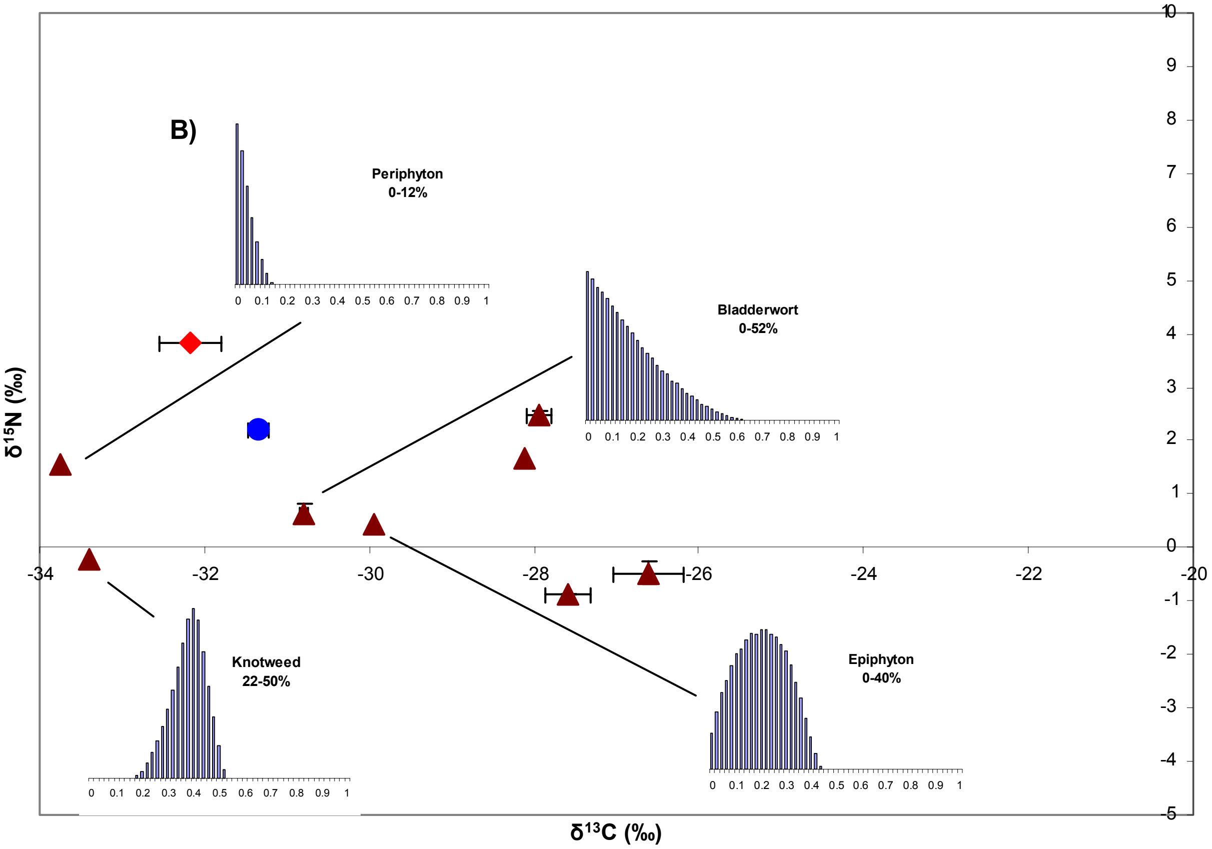
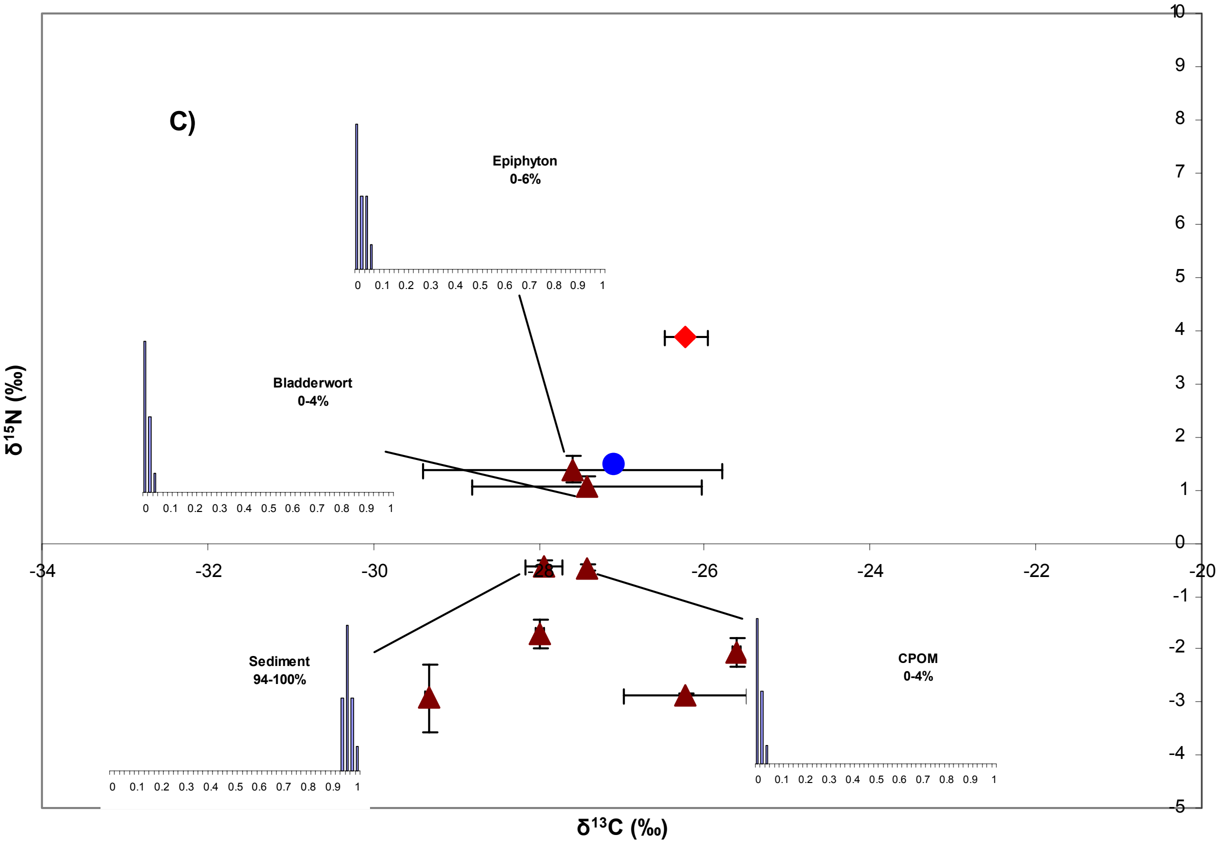
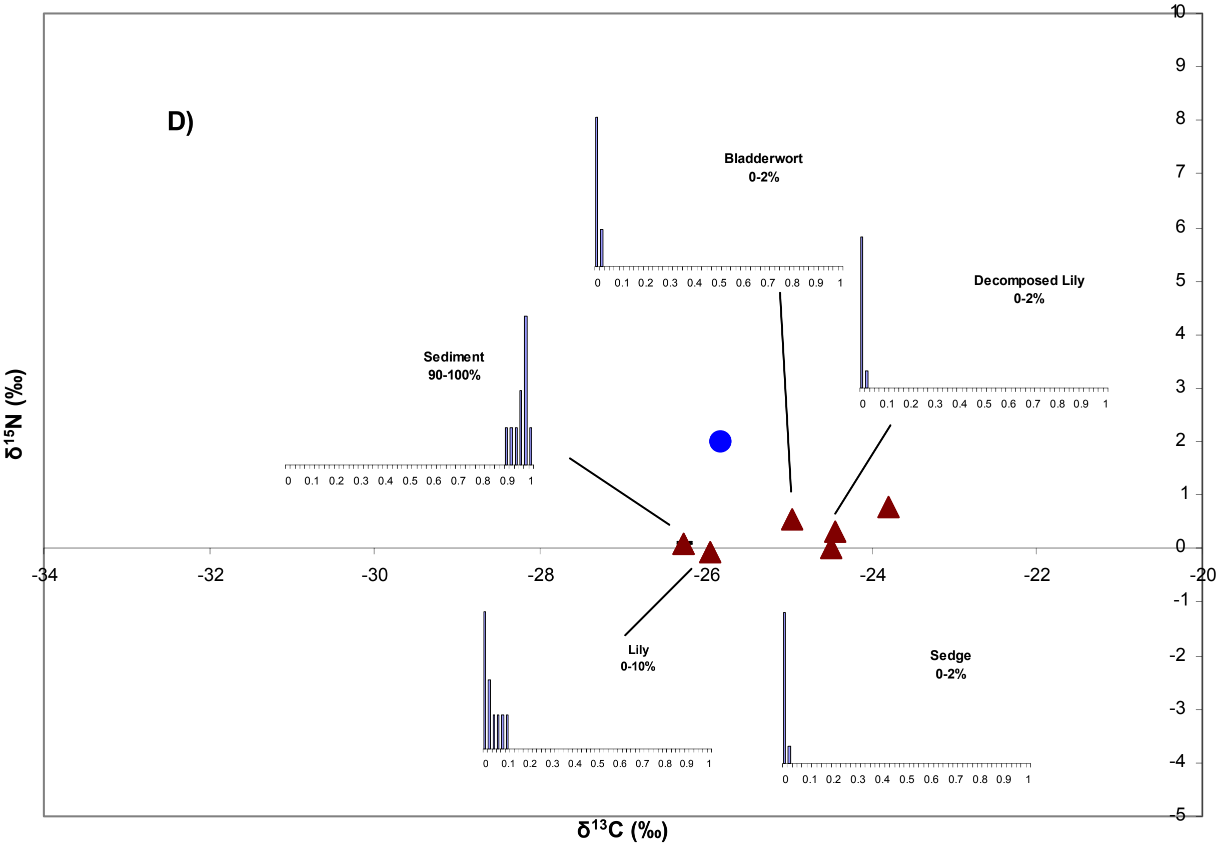

D)
δ15N (‰)
δ13C (‰)
-38
-36
-34
-32
-30
-28
-26
-24
-22
-20
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5

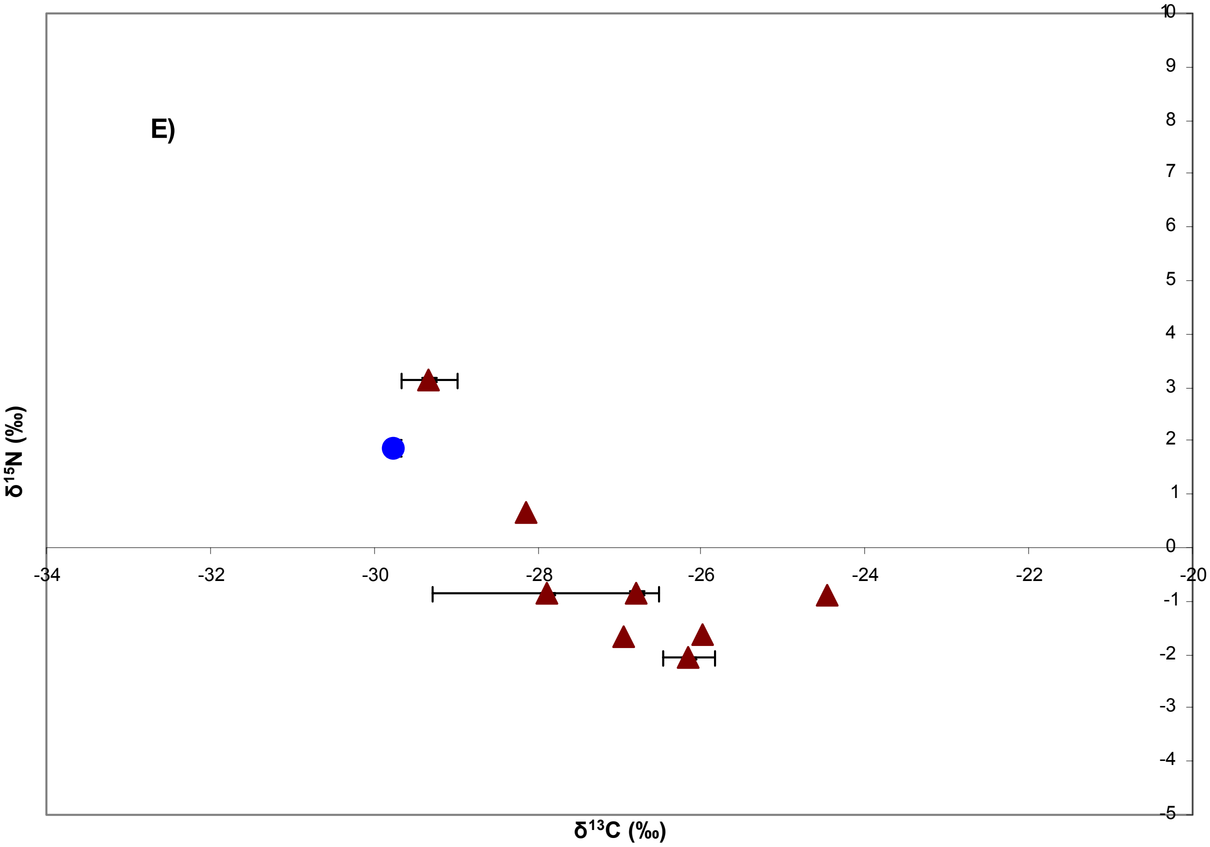
Figure 4. Stable isotope biplots of Okefenokee communities. A) Cypress dome January 2006 set, B) Cypress dome May 2006 set, C) Cypress dome March 2007 set, D) Lily prairie January 2006 set, E) Lily prairie May 2006 set, F) Lily prairie March 2007 set. Each data point is the mean of the sample replicates. The isotope values are ratios relative to a standard (air for $\delta^{15}N$, PDB for $\delta^{13}C$). Circles symbolize midge larvae; diamonds symbolize amphipods; and triangles symbolize sampled food sources. Error bars, indicating 1 standard error, are visible when they are larger than the data points. Sources within parameters described in Bunn and Boon (1993) were analyzed using IsoSource. The histograms provide the distribution of feasible IsoSource contributions from each source to the midge diet. Values shown in the boxes are the 1-99 percentile ranges for these distributions. (There were no solutions, and thus no histograms shown for Figure 4E.)


A)
Epiphyton
0-44%
Cypress bark
0-20%
Titi leaf
litter 0-40%
Cypress wood
34-52%
Sediment
0-22%
Cypress leaf
litter 0-22%
δ15N (‰)
δ13C (‰)
-34
-32
-30
-28
-26
-24
-22
-20
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1


B)
Periphyton
0-12%
Bladderwort
0-52%
Knotweed
22-50%
Epiphyton
0-40%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
δ15N (‰)
δ13C (‰)
-34 -32 -30 -28 -26 -24 -22 -20
10 9 8 7 6 5 4 3 2 1 0 -1 -2 -3 -4 -5


C)
Epiphyton
0-6%
Bladderwort
0-4%
Sediment
94-100%
CPOM
0-4%
δ15N (‰)
δ13C (‰)
-34
-32
-30
-28
-26
-24
-22
-20
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1


D)
Bladderwort
0-2%
Decomposed Lily
0-2%
Sediment
90-100%
Lily
0-10%
Sedge
0-2%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
δ15N (‰)
δ13C (‰)
-34 -32 -30 -28 -26 -24 -22 -20
10 9 8 7 6 5 4 3 2 1 0 -1 -2 -3 -4 -5


E)
δ15N (‰)
δ13C (‰)
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5
-34
-32
-30
-28
-26
-24
-22
-20

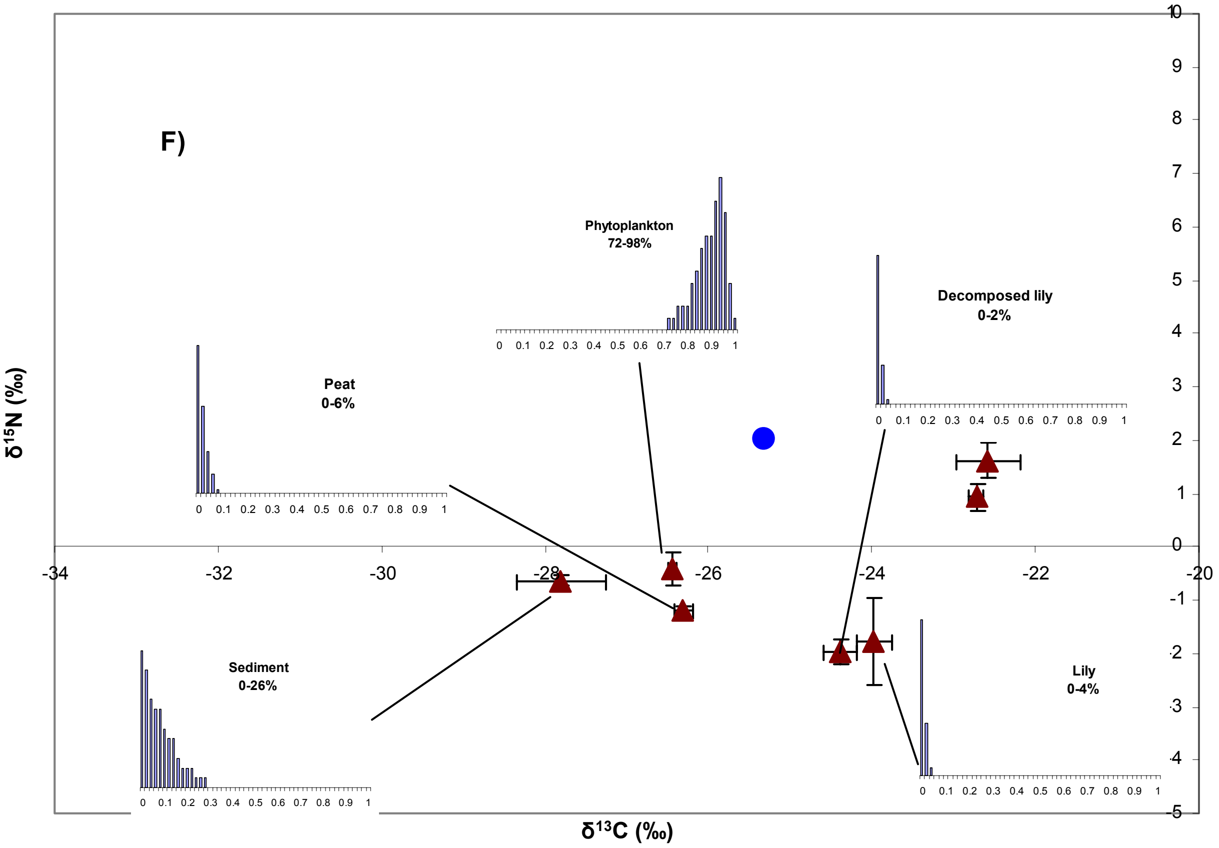

F)
δ15N (‰)
δ13C (‰)
Phytoplankton
72-98%
Decomposed lily
0-2%
Peat
0-6%
Sediment
0-26%
Lily
0-4%
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
-34
-32
-30
-28
-26
-24
-22
-20
10
9
8
7
6
5
4
3
2
1
0
-1
-2
-3
-4
-5

Table 1. Food quality of sampled food sources as reflected by C:N ratios. Bold font indicates sources that were found to be major foods by IsoSource modeling, and asterisks indicate unlikely foods. Samples with total nitrogen levels too low to be measured are indicated by a dash. Collection dates for the Coosawhatchie floodplain were 14 February 2006 and 14 March 2007, for the Carolina bays were 7 March 2006 and 17 April 2006, and for the Okefenokee swamp were 15 January 2006, 14 May 2006, and 13 March 2007.

	Forested			**Herbaceous**		
Coosawhatchie Floodplain	First collection	Second collection		First collection	Second collection	
Epiphyton	*28.5	**8.7**		**10.9**	17.4	
Metaphyton	22.4					
Periphyton					**11.2**	
Phytoplankton	10.2				*19.1	
Sediment	*--	14.9		*--	**13.3**	
Sweetgum wood	54.1	*88.6		52.5	96.7	
Sweetgum bark		79.1				
Sweetgum leaf litter	*47.5	**36.0**		*45.4	*31.0	
Tupelo leaf litter	**43.5**					
Oak leaf litter	72.0					
Detrital CPOM	*26.9	--				
Detrital FPOM		*11.3				
Micranthemum mudflower				**14.2**		
Polygonum knotweed		26.6		14.4		
Scirpus bulrush	22.4				25.2	
Carolina Bays	First collection	Second collection		First collection	Second collection	
Epiphyton		**9.8**		*14.4	16.5	
Metaphyton	21.8			**19.8**		
Periphyton	20.5	20.2			13.1	
Phytoplankton	*17.7	*13.5		14.7	--	
Sediment	*100.1	94.1		*--	77.5	
Sweetgum wood		*67.7			55.4	
Sweetgum bark	**95.3**					
Sweetgum leaf litter	48.9	*51.9				
Pine needle litter	86.8				21.7	
Decomposed lily				*13.9	19.6	
Dried *Panicum* grass		34.3			37.1	
Detrital CPOM	24.3	*26.5			29.6	
Detrital FPOM	**19.5**				26.3	
Callitriche starwort				15.9		
Lemna duckweed				17.6	29.4	
Nymphaea lily				15.4		
Polygonum knotweed				14.0		
Utricularia bladderwort				20.9		
Okefenokee Swamp	First collection	Second collection	Third collection	First collection	Second collection	Third collection
Epiphyton	10.6	14.7	*23.4	20.6	43.2	14.2
Periphyton		*6.6				

Phytoplankton	19.8				23.5	**23.5**
Sediment	25.6	13.8	**14.9**	**16.3**	12.5	9.1
Peat					30.3	17.2
Cypress wood	**70.9**	--	128.7			
Cypress bark	*55.5	67.1	45.0			
Cypress needle litter	50.2	49.8	47.5			
Titi leaf litter	54.7		50.1			
Decomposed lily				*31.5	32.5	*24.9
Detrital CPOM		26.0	*18.2		18.2	
Carex sedge				*19.0		
Nymphaea lily				*20.6	18.9	*18.6
Polygonum knotweed		**27.8**				
Utricularia bladderwort		19.3	*15.9	*26.0	21.6	12.5

Table 2. Midge food analysis summary table. The following sources were found by IsoSource analysis to be the major foods of the non-Tanypodinae midge larvae in the forested and herbaceous habitat in each wetland type, compiled from all collection dates.

	Forested	**Herbaceous**
Coosawhatchie Floodplain	Tupelo leaf litter Sweetgum leaf litter Epiphyton	Mudflower Epiphyton Sediment Periphyton
Carolina Bays	Detrital FPOM Sweetgum fruit litter Epiphyton	Metaphyton
Okefenokee Swamp	Wood Knotweed Sediment	Sediment Phytoplankton