Scientific note

Preliminary examination of food web structure of Nicola Lake (Taim Hydrological System, south Brazil) using dual C and N stable isotope analyses


Taim Ecological Reserve is located within the Taim Hydrological System and was created to protect a heterogeneous and productive landscape harboring exceptional biological diversity in southern Brazil. Using stable isotope ratio analyses of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$), we provide a preliminary description of the food web structure, including estimates of production sources supporting fish populations and vertical trophic structure, within a representative lake of this system. A total of 21 organisms (5 macrophytes, 3 mollusks and 13 adult fishes) representing 16 species were collected for isotope analysis. Fishes had $\delta^{13}C$ values ranging from -24.30 ‰ to -28.31 ‰, showing concordance with the range of values observed for macrophytes (-25.49 to -27.10 ‰), and suggesting that these plants could be a major carbon source supporting these fishes. $\delta^{13}C$ signatures of Corbicula (-30.81‰) and Pomacea (-24.26‰) indirectly suggest that phytoplankton and benthic algae could be alternative carbon sources for some consumers. Nitrogen isotope ratios indicated approximately three consumer trophic levels. The pearl cichlid Geophagus brasiliensis was a primary consumer. Two catfishes (Trachelyopterus lucenai and Loricariichthys anus) were secondary consumers. Two congeneric pike cichlids (Crenicichla lepidota and C. punctata), a catfish (Pimelodus maculatus) and the characids Astyanax fasciatus and Oligosarcus robustus were tertiary consumers.

Further studies including additional primary producers and consumers and greater sample numbers should be conducted to provide a more complete and detailed description of food web structure and dynamics within the reserve.

Key words: Subtropical wetland, Neotropical fishes, Patos-Mirim Lagoon complex, Long-Term Ecological Research (LTER), Brazilian Long Term Ecological Research (PELD).

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Taim Ecological Reserve is located within a larger area called Taim Hydrological System and was created in 1978 to protect a unique freshwater wetland ecosystem in southern Brazil that had been suffering increasing anthropogenic impacts in its surroundings, such as water diversion for rice irrigation and fishing activities. The reserve encompasses a variety of habitats such as beaches, dunes, forests, grasslands and, especially, lakes and wetlands. This heterogeneous and productive landscape harbors exceptional biological diversity (Motta Marques et al., 2002). In face of the above mentioned threats, it is critically important to develop baseline data on aquatic community attributes in this ecological reserve for effective conservation and management.

Fishes have been poorly investigated within Taim. Published information is restricted to taxonomic lists (Buckup & Malabarba, 1983; Grosser et al., 1994; Garcia et al., 2006), description of new species (Buckup, 1981; Reis, 1983) and, more recently, a feeding study (Moresco & Bemvenuti, 2005). Food web structure and dynamics in this area have not been investigated.

Based on the isotopic ratio method, we provide a preliminary description of food web structure at Nicola, a representative lake inside the reserve, including estimates of production sources supporting fish populations and vertical trophic structure. Isotopic ratios of carbon and nitrogen provide information on sources of organic matter important to consumers as well as insights about how materials are processed within trophic networks (Peterson & Fry, 1987).

Producer and consumer taxa were sampled at Nicola Lake (Taim Hydrological System) on March 25th, 2005 (Fig. 1). Our survey targeted macrofauna (fishes and mollusks) and macrophytes occurring at the study site during the day of collection. Fishes caught in Nicola Lake for isotope analysis constituted a small (9 out of 42 species occurring in this site) but a representative sample of fishes inhabiting open beaches and pelagic waters in this lake (Garcia et al., 2006). Floating and emergent macrophytes, snails and bivalves were collected by hand, and fishes were collected with beach seines, cast nets and gillnets. All samples were placed on ice for transport to the laboratory where they were frozen. Samples for isotopic analysis consisted of several leaves for plant species, muscle tissue (adductor) from snail and bivalve individuals, and approximately 5g of dorsal muscle tissue from individual fish. Thawed samples were inspected to remove contaminants (e.g. periphyton on leaves, bone or scales in fish tissue), rinsed with distilled water, placed in sterile Petri dishes and dried in an oven at 60°C to constant weight (minimum of 48 hrs). Dried samples were ground to a fine powder with a mortar and pestle and stored in clean glass vials.

Sub-samples were weighed to the nearest 10^-6 g, pressed into Ultra-Pure tin capsules (Costech, Valencia, CA), and sent to the Analytical Chemistry Laboratory, Institute of Ecology, University of Georgia for determination of stable isotope ratios (δ¹³C/¹²C and δ¹⁵N/¹⁴N). Results are reported as parts per thousand (‰) differences from a corresponding standard: δX = [(R_{sample}/R_{standard})-1] x 10³, where R = ¹³C/¹²C or ¹⁵N/¹⁴N. Standards were carbon in the PeeDee Belemnite and molecular nitrogen gas in air. Standard deviations of δ¹³C and δ¹⁵N replicate analyses were 0.14‰ and 0.13‰ respectively.

Bi-plots of δ¹³C and δ¹⁵N values were used to visualize patterns of energy flow between sources and aquatic consumers. Sources of organic carbon assimilated by consumers are indicated by relative positions of taxa on the x-axis (δ¹³C values), whereas trophic level is indicated by relative position on the y-axis (δ¹⁵N) (Peterson & Fry, 1987).

Previous studies have shown that δ¹⁵N signatures are
accurate indicators of trophic position in aquatic systems because of predictable δ¹⁵N enrichment of consumers relative to their diet (Vander Zanden & Rasmussen 1999; Post, 2002; Vanderklift & Ponsard, 2003). Considering that heavier δ¹⁵N accumulates in consumers as it moves up the food web, consumers tend to be enriched by 2.5 to 3.5‰ relative to their food resources.

Thus, nitrogen isotopic distributions were used to estimate trophic positions of consumers following the method described in Post (2002) using snails and bivalves to estimate the littoral and pelagic bases of the food web: TP = λ + (δ¹⁵N_predator - δ¹⁵N_bivalve - α + δ¹⁵N_snail (1 - α))/F, where λ is the trophic level of consumers estimating the base of the food web (in this case λ = 2 because snails and bivalves are primary consumers), δ¹⁵N_predator is the nitrogen signature of the consumer being evaluated, α is the proportion of carbon derived from the pelagic food web base: α = (δ¹³C_predator - δ¹³C_snail)/(δ¹³C_bivalve - δ¹³C_snail), and F is the per trophic level fractionation of nitrogen [in this case 2.54, following the meta-analysis of Vanderklift & Ponsard (2003)]. Previous experimental studies by DeNiro & Epstein (1981), Minagawa & Wada (1984), and Vander Zanden & Ramussen (1996) suggested a fractionation value of approximately 3.4, therefore the trophic position values presented here should be considered upper estimates.

A total of 21 organisms (5 macrophytes, 3 mollusks and 13 adult fishes) representing 16 species was collected for isotope analysis (Table 1). Macrophytes showed higher between-taxon variation in δ¹⁵N (4.00 to 8.57 ‰) than δ¹³C (-25.49 to -27.10 ‰) signatures (Fig. 2). Floating macrophytes (Pistia stratiotes, Eichhornia crassipes, Salvinia spp.) were more enriched in δ¹⁵N than emergent macrophytes (Cladium jamaicense, Scirpus californicus). The floating macrophyte P. stratiotes had δ¹⁵N values that surpassed values of some consumers (both mollusks and the fish Geophagus brasiliensis). The two mollusks (Corbicula and Pomacea) had similar δ¹⁵N values (around 8‰) but contrasting δ¹³C signatures, with Corbicula showing a lighter signature (-30.81 ‰ mean value) compared to Pomacea (-24.26 ‰). Fishes had δ¹³C values ranging from -24.30 ‰ to -28.31 ‰, showing concordance with the range of values observed for macrophytes (-25.49 to -27.10 ‰), and suggesting that these plants could be a major carbon source supporting these fishes (Fig. 2; Table 1).

Macrophytes revealed a δ¹³C range concordant with values reported in the literature for terrestrial plants using the C₃
Table 1. Taxonomic status and total length (TL mm) of the individuals collected at Nicola Lake, Taim Hydrological System. # symbols denotes different individuals of the same species.

<table>
<thead>
<tr>
<th>Family</th>
<th>Species</th>
<th>TL (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Fishes</strong></td>
<td></td>
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</tr>
<tr>
<td>Cichlidae</td>
<td>Geophagus brasiliensis</td>
<td>190</td>
</tr>
<tr>
<td></td>
<td>Crenicichla lepidota</td>
<td>165</td>
</tr>
<tr>
<td></td>
<td>Crenicichla punctata</td>
<td>190</td>
</tr>
<tr>
<td>Erythrinidae</td>
<td>Hoplias malabaricus #1</td>
<td>375</td>
</tr>
<tr>
<td></td>
<td>H. malabaricus #2</td>
<td>226</td>
</tr>
<tr>
<td>Auchenipteridae</td>
<td>Tracheylopterus lucenai #1</td>
<td>205</td>
</tr>
<tr>
<td></td>
<td>T. lucenai #2</td>
<td>210</td>
</tr>
<tr>
<td></td>
<td>T. lucenai #3</td>
<td>245</td>
</tr>
<tr>
<td>Pimelodidae</td>
<td>Pimelodus maculatus</td>
<td>305</td>
</tr>
<tr>
<td>Loraciidae</td>
<td>Loricarichthys anus #1</td>
<td>386</td>
</tr>
<tr>
<td></td>
<td>L. anus #2</td>
<td>239</td>
</tr>
<tr>
<td>Characidae</td>
<td>Oligosarcus robustus</td>
<td>172</td>
</tr>
<tr>
<td></td>
<td>Astyanax fasciatus</td>
<td>123</td>
</tr>
<tr>
<td><strong>Mollusks</strong></td>
<td></td>
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<tr>
<td>Corbiculidae</td>
<td>Corbicula sp.</td>
<td></td>
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<tr>
<td>Ampullariidae</td>
<td>Pomacea sp.</td>
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</tr>
<tr>
<td>Araceae</td>
<td>Pistia stratiotes</td>
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<tr>
<td>Pontederiaceae</td>
<td>Eichhornia crassipes</td>
<td></td>
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<tr>
<td>Salviniaae</td>
<td>Salvinia sp.</td>
<td></td>
</tr>
<tr>
<td>Cyperaceae</td>
<td>Scirpus californicus</td>
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<tr>
<td></td>
<td>Eurya macrophytes</td>
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photosynthetic pathway (-23 ‰o and -30 ‰o, Smith & Epstein, 1971; -26.8 ‰o and 28.9 ‰o, Deegan & Garritt, 1997) rather than the relatively enriched signatures shown by species that employ the $C_4$ photosynthetic pathway (between -12.9 ‰o and -15.8 ‰o, Deegan & Garritt, 1997; -10‰ and -16‰, Forsberg et al., 1993). In contrast with few $C_4$ emergent grasses (e.g., *Zizaniaxis bonariensis*), which were not sampled in this study, most aquatic macrophytes inhabiting Taim freshwater wetlands seemed to be $C_3$ plants (Motta Marques et al., 1997). High concordance between $\delta^{13}C$ values of these primary producers and consumers suggests that analyzed macrophytes could constitute a major carbon source supporting aquatic consumers at Nicola Lake. In the Baia River (Paraná River floodplain, Brazil), Manetta and collaborators (2003), found that $C_4$ plants seem to be the main carbon source for the ichthyofauna.

However, the contrasting $\delta^{13}C$ signatures of the two mollusks (*Corbicula*: -30.81 ‰o and *Pomacea*: -24.26 ‰o), which constituted the extremes values observed for the $\delta^{13}C$ signatures, also suggests that phytoplankton and benthic algae could be alternative carbon sources. Based on diet studies of congeneric species (Fellerhoff, 2002; Kasai et al., 2006), we hypothesize *Corbicula* $\delta^{13}C$ signatures reflect the isotopic composition of seston (phytoplankton, suspended detritus and bacteria) and *Pomacea* reflect $\delta^{13}C$ largely derived from benthic algae. Phytoplankton and benthic algae have been shown to be important carbon sources supporting aquatic food webs in a variety of habitats (e.g. Araujo-Lima et al., 1986; Post, 2002; Vander Zanden & Vadeboncoeur, 2002; Kang et al., 2003).

Nitrogen isotope ratios estimated four trophic levels at Nicola Lake (Fig. 2 and 3). The cichlid *G. brasiliensis* approximated the second trophic level (primary consumers, trophic position = 2.1), clustering together with suspension feeder *Corbicula* and grazer *Pomacea*. The black catfish *T. lucenai* and the armored catfish *L. anus*, had intermediate trophic position values (between 2.4 and 3.0). Pike cichlids *Crenicichla lepidota* and *C. punctata*, Bloch’s catfish *Pimelodus maculatus*, trahira *H. malabaricus* (individual #2), and the characids *Astyanax fasciatus* and *Oligosarcus robustus* occupied the third trophic level (from 3.0 to 3.4).

These results provided a first glance at vertical trophic structure at Nicola Lake. Although some species had intermediate trophic positions, our results suggested approximately three consumer trophic levels. The pearl cichlid, *Geophagus brasiliensis*, was a primary consumer. Two catfishes (T. lucenai and L. anus) were secondary consumers. Two congeneric pike cichlids (*Crenicichla lepidota* and *C. punctata*), a catfish (*Pimelodus maculatus*) and the characids *Astyanax fasciatus* and *Oligosarcus robustus* were tertiary consumers.

Stomach contents analysis performed on these species at Taim (Moresco & Bemvenuti, 2005) and elsewhere in Brazil (Arcifa & Meschiatti, 1993; Hahn et al., 2004; Casseimiro et al., 2005) largely corroborated the trophic positions obtained by the isotopic ratio method. The pearl cichlid, *Corbicula*, was on detritus/sediment, aquatic insects, fish and fish scales in a shallow eutrophic reservoir, Monte Alegre Lake, São Paulo state (Arcifa & Meschiatti, 1993). In the Iguacu River (Paraná state), the pearl cichlid diet is composed mainly of detritus/sediment, micro-crustaceans, insects, fish, algae and vascular plants (Casseimiro et al., 2005). Within the Taim Ecological Reserve, black catfish *T. lucenai* has a diet composed mainly of insects and fish, followed by mollusks and crustaceans. This species also seems to be an opportunistic forager, as evidenced by the occurrence of the blind-snake *Amphisbaena darwinii* in the stomach of one individual (Moresco & Bemvenuti, 2005). The armored catfish *Loricariichthys* sp. from the Upper Paraná River floodplain fed mostly on detritus, sediments and insects, plus lesser amounts of other invertebrates and algae (Hahn et al., 2004). Casseimiro et al. (2005) found that trahiras and pike cichlids at rio Iguacu were piscivores, and catfish of the genus *Pimelodus* fed mostly on fish and micro-crustaceans.

The two analyzed trahira specimens from Taim had different trophic positions (2.4 and 3.4). We speculated that this apparent incongruent pattern could be associated with diet changes during ontogeny. The smaller trahira (226 TL mm) likely had a higher trophic position because it fed on small insectivorous characins. The larger trahira (375 mm) may have fed heavily on relatively larger detritivorous fishes. In the Venezuelan llanos trahira experience ontogenic diet shifts from invertebrate feeding by small juveniles to piscivory by...

Despite the low number of samples analyzed and lack of adequate sample replication, our study provides a preliminary estimate of food web structure within Taim Ecological Reserve. Further food web studies in Taim’s wetlands should include additional primary producers (e.g., phytoplankton, benthic microalgae) and consumers (e.g., benthic invertebrates, zooplankton and other fish species) to provide a more complete and detailed description of food web structure and dynamics. The isotopic ratio method could also be used to investigate ontogenetic changes in diet (Genner et al., 2003), temporal and spatial variation (Deegan & Garritt, 1997; Dalerum & Angerbjorn, 2005) and material transfers associated with animal movements (Cunjak et al., 2005; Herzka, 2005). Stable isotope analyses have been underutilized in investigations of food web structure within protected areas in Brazil. Compared to the more common method of stomach contents analysis, the stable isotope method yields estimates of trophic structure without requiring large sample sizes, a significant advantage when studying protected ecosystems.

Acknowledgments

Thanks to the numerous colleagues and technicians that assist in the field and laboratory work, especially Antonio R. T. Bueno, Ana C. Mai, Fábio Roselet and Vinicius Condini; to Dr. César S. B. Costa for providing assistance during sample pre-preparation; to Dr. Cesar V. Cordazzo and MSc Juliano C. Marangooni for helping with taxonomic plant identification. AMG acknowledges fellowship support provided by CNPq (Grant 150868/2003-0). DJH was supported in part by research scholarships from the Society of Wetland Scientists. PELD-Sítio 7 (Sistema Hidrológico do Taim) provided logistical support (Grant 52.0027/98-1). Facilities were made available to DJH at the Research Nucleus in Limnology, Ichthyology and Aquaculture (Nupélia) at the State University of Maringá (UEM), Brazil.

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Received November 2005
Accepted May 2006