

Food webs of the Paraná River floodplain: Assessing basal sources using stable carbon and nitrogen isotopes



Mercedes Rosa Marchese ^{a,b,*}, Miguel Saigo ^a, Florencia Lucila Zilli ^a, Soledad Capello ^a, Melina Devercelli ^a, Luciana Montalto ^{a,b}, Graciela Paporello ^a, Karl Matthias Wantzen ^c

^a Instituto Nacional de Limnología (INALI, CONICET-UNL), Ciudad Universitaria, 3000 Santa Fe, Argentina

^b Facultad de Humanidades y Ciencias (FHUC-UNL), Ciudad Universitaria, 3000 Santa Fe, Argentina

^c Interdisciplinary Research Center for Cities, Territories, Environment and Society (CNRS UMR CITERES), Université François Rabelais, Parc Grandmont, 37200 Tours, France

ARTICLE INFO

Article history:

Received 24 February 2013

Received in revised form

23 November 2013

Accepted 25 November 2013

Available online 11 December 2013

Keywords:

Carbon sources

Detritivores

Herbivores

Isotopic niche

Benthic-pelagic food webs

ABSTRACT

Food webs in floodplain ecosystems may be based on a variety of aquatic, terrestrial or amphibious food resources. Here, we determined which of the basal resources mostly contribute to the food webs in a floodplain lake of the Middle Paraná River using isotopic composition of C and N ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) of potential food sources in the Paraná floodplain (Argentina). We analyzed if organic matter sources isotope of C and N differ between flooding and low water seasons, and analyzed the isotopic niche representations of consumers in order to characterize niches width and intraguilds overlapping. To estimate the contribution of different sources of carbon to primary consumers, we measured the stable isotopic compositions of bottom sediment organic matter, coarse particulate organic matter, biofilm, suspended particulate organic matter, epiphyton, phytoplankton, C₃ and C₄ macrophytes and riparian tree leaves, benthic macroinvertebrates, aquatic orthopterans and fishes in dry and flooding seasons. The packages Stable Isotope Analysis and the Stable Isotope Bayesian Ellipses algorithm in R were calculated to compare the C and N isotopic variability between the primary consumers and their sources. The energy sources available for benthic organisms mainly originated from autochthonous sources based on the C₃ photosynthesis pathway. The isotopic signatures of sources and primary consumers did not differ significantly between low and high water seasons. Our results demonstrated a higher contribution to primary consumers of C₃ macrophytes and low contributions of C₄ for herbivores; biofilm and benthic organic matter for gatherer collectors (Oligochaeta and Chironominae); epiphyton for ephemeropterans, amphipods and fishes, whereas biofilm was the most important source for mussels.

© 2013 Elsevier GmbH. All rights reserved.

Introduction

Classical concepts for the functioning of riverine systems are generally based on organic matter flows and carbon pathways. In the River-Continuum Concept (Vannote et al., 1980) the importance is given to the gradual transformation of allochthonous inputs from the terrestrial part of the catchment (e.g., leaves, wood, riparian and upland trees, shrubs, and grasses) into fine particulate organic matter (FPOM) and dissolved organic matter (DOM), and an increasing production of autochthonous algae along with the increasing stream order. The Flood Pulse Concept (Junk et al., 1989)

focuses on the importance of the floodplain as productive areas, whereas the Riverine Productivity Model (Thorp and DeLong, 1994) stresses the importance of benthic production even in lower river section.

In this context, the study of the relative importance of the different food sources in freshwater food webs stand as a key issue to understand the ecosystem functioning. For example, whereas C₄ grasses mainly represent the river bank production, C₃ macrophytes make up most of lakes primary production in Paraná River system (Sabattini and Lallana, 2007). In a similar way the benthic production is held by detritus inputs (particulate organic matter) and by biofilm which is a complex of primary producers (diatoms mainly) and microbes.

The Stable Isotopes Analysis (SIA) in ecological research allows a quantification of the importance of the individual compartments of organic matter (Fry, 2006). Indeed, this analytical tool is increasingly used to trace energy flow and describe food web structures because it integrates both the variation across spatial

* Corresponding author at: Instituto Nacional de Limnología (INALI, CONICET-UNL), Ciudad Universitaria, 3000 Santa Fe, Argentina. Tel.: +54 342 4511645; fax: +54 342 4511645x111.

E-mail addresses: mmarchese@inali.unl.edu.ar, mercedes.marchese@gmail.com (M.R. Marchese).

and temporal scales, providing important information to understand food web dynamics (Peterson and Fry, 1987; Paetzold et al., 2005, 2006; O'Callaghan et al., 2013). Moreover, in recent years, mixing models have been developed and applied to SIA in order to determine sources contribution to consumers (Phillips and Gregg, 2003), being carbon and nitrogen isotope ratio commonly used in food webs studies (Minagawa and Wada, 1984; Post, 2002; Rubenstein and Hobson, 2004; Kurle, 2009).

The calculation of proportional contributions of the basal resources to food webs in floodplain lakes is challenging because existing isotopic mixing model approaches are generally sensitive to the number of sources they can accommodate, based on the number of isotopic tracers used (Phillips and Gregg, 2003). The models (e.g., MixSIR and SIAR) that have recently emerged based on stable isotope data combined with Bayesian analysis techniques (Moore and Semmens, 2008; Parnell et al., 2010; Solomon et al., 2011) incorporating many sources of variability within the model can be adequate tools for quantifying such sources of uncertainty and for estimating the contribution of multiple sources to floodplain lake food webs. The isotopic values of consumers may be represented in δ -space and has been used to explore questions traditionally resided within the domain of ecological niche theory, formalized in the concept of "isotopic niche" by Newsome et al. (2007, 2012). Thus, the isotopic axes provide quantitative information on both resource (bionomic) and habitat (scenopoetic) factors commonly used to define ecological niche space (Newsome et al., 2007; Martínez del Rio et al., 2009).

Stable isotope studies have tested patterns of food web structure in large rivers of the neotropics (Araújo-Lima et al., 1986a,b; Hamilton and Lewis, 1992; Hamilton et al., 1992; Martinelli et al., 1994; Thorp et al., 1998; Lewis et al., 2001; Wantzen et al., 2002, 2011; Benedito-Cecílio et al., 2004; Jepsen and Winemiller, 2002, 2007; Lopes et al., 2009; Correa and Winemiller, in press) and a point of view arising from these studies is that river food webs have a strong watershed signal embedded in consumer isotopic compositions (Fry and Allen, 2003). The organic matter sources of aquatic food webs are highly diverse in space and time in large river-floodplain-systems. During rising inundation (the rising limb of the flood pulse), differently organic matter is driven into the floodplain area (Junk and Wantzen, 2004, 2006; Wantzen and Junk, 2006). On the other hand, autochthonous material (e.g., biomass resulting from primary production by attached algae, phytoplankton and aquatic macrophytes) produced or accumulated during the dry period is fundamental for energy flow and carbon stocks in floodplains of large rivers (Martinelli et al., 1994; Benedito-Cecílio et al., 2000). Although many studies have documented the major role of allochthonous detritus in low order streams (Füreder et al., 2003), autochthonous production is also a valuable food resource because of its high nutrient and energy contents (Forsberg et al., 1993; Thorp and Delong, 2002; Jepsen and Winemiller, 2007). Recent works on stable isotopes has proved the importance of algal primary production even for smaller streams in the tropics (Brito et al., 2006; Dudgeon et al., 2010).

Based on this conceptual background, our goals were to determine the basal resources that mostly contributed to the food webs in a floodplain lake of the Middle Paraná River, to analyze if isotopic signatures (C and N) of organic matter sources differ between flooding and low water seasons, and to analyze the isotopic niche representations of consumers to characterize niches width and intra-guilds overlapping. To address these questions, we collected different potential energy sources and primary consumers in a floodplain lake during two hydrological phases.

Materials and methods

Study site

Samplings were carried out during high (November 2009 and March 2010) and low water seasons (September and December 2010) in a floodplain lake connected to the main channel of the Middle Paraná River ($31^{\circ}41'S$, $60^{\circ}43'W$). Water temperature ranged from 24.6 to $27.7^{\circ}C$ during the study period. The floodplain lake was characterized by low conductivity ($62.0\text{--}70.3\ \mu\text{S}/\text{cm}$), water transparency did not exceed 0.8 m, and dissolved oxygen varied between 3.2 and 7.2 mg/l. The benthic particulate organic matter ranged from 10.0 to 17.4% (Mesa et al., 2012).

Sample collection and isotope analyses

We collected different energy sources and primary consumers in a floodplain lake during two hydrological seasons, such as bottom sediment organic matter (BSOM), coarse particulate organic matter (CPOM), biofilm, suspended particulate organic matter (SPOM), epiphyton (filamentous green algae attached to macrophytes), phytoplankton, macrophytes and riparian tree leaves, benthic macroinvertebrates, aquatic orthopterans, and fish for SIA.

Benthic macroinvertebrates were sampled with a Tamura grab (sampled surface: $322\ \text{cm}^2$), the bivalves with a dredge and the orthopterans with a D-net. After allowing time for gut clearance, the individuals were rinsed with distilled water to remove attached inorganic and organic materials. Depending on the body mass of taxa, samples of multiple individuals (same location, same sampling date) were pooled to provide 3–4 mg samples for replicated analysis. The legs of orthopterans and the foot muscle in mollusks were selected for SIA.

The dominant and more frequent C₃ and C₄ plants in the Paraná River system were selected to analyze the relative contribution as sources of energy. Several leaves (10–15) of the dominant C₃ floating macrophytes such as, *Eichhornia crassipes*, *Pistia stratiotes*, *Salvinia biloba*, *Limnobium laevigatum*; rooted plants, such as *Ludwigia peploides*, *Nymphoides indica*, *Polygonum* sp.; submerged plants, such as *Ceratophyllum demersum* and C₄ grass *Paspalum repens* and *Echinochloa* sp. were handpicked. Stable isotope data of riparian vegetation consisted in values of fresh leaves of dominant riparian plants in the floodplain of the Middle Paraná River, such as *Salix humboldtiana* and *Sapium haematospermum*. CPOM was represented by samples of leaf litter collected from the bottom of the lake and sorted with sieves ($>1000\ \mu\text{m}$). Water-column SPOM samples were collected from integrated subsurficial water samples and filtered onto precombusted (450°C) glass fiber filters (Whatman® GF/F). Then, filters were frozen until isotope analysis. Bottom sediment organic matter (BSOM), which is mainly associated with silt and clay, was taken with a Tamura grab in the upper 2–3 cm of sediment depositions. Stable carbon isotope analysis was performed separately for epiphytic and planktonic algae. The epiphyton filamentous algae were scraped from aquatic plant stems, whereas phytoplankton was collected with a net of 25 μm mesh. In order to separate epiphytic and planktonic algae from detritus, we performed density fractionation in colloidal silica Ludox® AM-50 (density $1.210\ \text{g}/\text{cm}^3$) diluted with deionized water according to Hamilton et al. (2005).

Biofilm was sampled with an ooze sucker sampler (Welch, 1963), collecting the interstitial water layer between the bottom sediment and the water column. The biofilm consisted in amorphous non-living organic matter, mixed with adhering bacteria and fungi, diatoms, protozoans, and mineral particles. The selected invertebrates were identified to the lowest possible taxonomic level, and then associated with different

functional feeding groups: herbivores (mollusks *Pomacea canaliculata* and *Omalonix* sp., and orthopterans *Cornops aquaticum*), filterers (Bivalvia *Diploodon parallelopipedon* and *Limnoperna fortunei*), gatherer-collectors (Oligochaeta Tubificinae, Diptera Chironominae, Ephemeroptera *Campsurus* cfr. *violaceus*, and Amphipoda *Hyalella curvispina*). Meanwhile, a detritivorous fish (*Prochilodus lineatus*) was also included in the analysis because it is the most important consumer in terms of production in the floodplain lakes of the Paraná River (Cordoviola de Yuan, 1992; Rossi et al., 2007). Fishes were collected with fishing nets. The dorsal-lateral muscle (approximately 5 g) was excised from adult individual fish to perform SIA. Scales were removed to obtain only soft tissue samples. All samples were transported on ice to the laboratory where they were rinsed with distilled water, placed in sterile Petri dishes and dried in an oven at 45–50 °C to constant weight (minimum of 48 h). After drying, samples were ground to a fine powder with a pestle and mortar, stored in clean glass vials, and frozen until further processing in the laboratory. Given that high lipid levels (indicated by high C:N) can drive $\delta^{13}\text{C}$ in a negative direction (McConaughey and McRoy, 1979; Matthews and Mazumder, 2005), the stable carbon data were normalized. For that purpose, we used the arithmetic equation $\delta^{13}\text{C} = -3.32 + 0.99(\text{C:N})$ outlined in Post et al. (2007), in which C:N is the elemental ratio. For plants with >40% of C, we used the equation $\delta^{13}\text{C} = -5.83 + 0.14\%$ Carbon (Post et al., 2007).

SIA was performed in a mass spectrometer (IRMS Finnigan MAT Delta S) coupled to an elemental analyzer (CATNAS, Montevideo, Uruguay).

The ratio of stable isotopes was expressed by convention in delta (δ) notation:

$$\delta = \left[\left(\frac{R_{\text{sample}}}{R_{\text{standard}}} \right) - 1 \right] \times 1000$$

where δ is the isotope ratio of the sample relative to a standard. R_{sample} and R_{standard} are the fractions of heavy to light isotopes in the sample and the standard, respectively. One is subtracted from the $R_{\text{sample}}/R_{\text{standard}}$ fraction so that samples with lower ratios of heavy isotopes than the standard have a negative value and those with higher ratios of heavy isotopes than the standard have a positive value. This number is then multiplied by 1000 so that the δ notation is in units of parts per thousand (‰). The mean trophic fractionation used for $\delta^{15}\text{N}$ was 3.4‰ (SD = 1‰) and for $\delta^{13}\text{C}$ was 0.4‰ (SD = 1.3‰), according to Post (2002). Initially, we compared the data of C and N isotopes using the analysis of variance (ANOVA, $p < 0.05$) to test differences between low waters and high waters seasons. One-way ANOVA ($p < 0.05$) and post hoc Tukey test analyses were performed to determine differences in mean isotopic ratios between species pairs. Statistical analyses were conducted with InfoStat software (Di Rienzo et al., 2013).

To determine the proportional contribution of each source to consumers, we used the package Stable Isotope Analysis in R (SIAR – Parnell et al., 2008, 2010). This model is based on a series of related linear equations that utilize Bayesian statistical techniques to identify proportional contributions of source pools (Jackson et al., 2009; Moore and Semmens, 2008; Parnell et al., 2010; Solomon et al., 2011). In contrast to other statistical tools used in SIA (e.g., Euclidean methods, IsoSource), outputs from the Bayesian models are true probability distributions, not just summaries of all feasible solutions (Layman et al., 2012). The sources were selected and pooled based on published information on invertebrates and fish diet (Bowen et al., 1984; Saigo et al., 2009; Capello et al., 2011, 2012; Galizzi et al., 2012; Saigo, unpublished). Sources that did not show significant differences among them ($p < 0.05$) were jointly considered in the mixing model of each consumer. The Stable Isotope

Bayesian Ellipses (R package SIBER, Jackson et al., 2011) were calculated to evaluate the distributions of C and N isotopic values of the primary consumers, providing an estimation of isotopic niche width.

Results

Isotopic signatures of sources and consumers

The variation range of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in C_3 macrophytes was considerable large (from $-25.44 \pm 0.97\%$ to $-31.33 \pm 1.67\%$ and $3.89 \pm 2.05\%$ to $9.06 \pm 1.78\%$; respectively) with the emergent *N. indica* being the most enriched in $\delta^{13}\text{C}$ and *Polygonum* sp. in $\delta^{15}\text{N}$, and the submerged *C. demersum* being the most depleted in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ (Table 1). Floating macrophytes (*E. crassipes*, *P. stratiotes*, *L. laevigatum* and *S. biloba*) were more depleted in $\delta^{13}\text{C}$ than emergent macrophytes (*L. peploides*, *N. indica*, *Polygonum* sp.). The emergent C_4 macrophytes (*P. repens* and *Echinochloa* sp.) had $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ranging from $-11.84 \pm 0.29\%$ to $-11.98 \pm 0.06\%$ and $6.80 \pm 1.35\%$ to $6.82 \pm 1.39\%$, respectively. The dominant tree species on the banks (*S. humboldtiana* and *S. haematospermum*) had similar $\delta^{13}\text{C}$ values as those observed for emergent macrophytes and $\delta^{15}\text{N}$ values like submerged macrophytes. Additionally, epiphyton was the most variable sources with SD of $\pm 8.25\delta^{13}\text{C}$. The source SPOM showed the highest level of $\delta^{15}\text{N}$ enrichment ($8.51 \pm 1.59\%$) and BSOM exhibited low $\delta^{15}\text{N}$ enrichment ($3.46 \pm 0.46\%$). The $\delta^{13}\text{C}$ isotopic signatures of detritivores presented higher variation among individuals of *H. curvispina* ($-23.17 \pm 3.69\%$), *C. cfr. violaceus* ($-29.07 \pm 3.50\%$) and *P. lineatus* ($-27.33 \pm 3.12\%$) than Chironominae ($-26.98 \pm 1.38\%$) and Oligochaeta Tubificinae ($-28.00 \pm 1.42\%$) indicating large variations among individuals (Table 1). Four mollusks (*P. canaliculata*, *Omalonix* sp., *D. parallelopipedon* and *L. fortunei*) had variable $\delta^{13}\text{C}$ values, with the filter feeding bivalve *D. parallelopipedon* showing a more depleted signature ($-28.67 \pm 0.64\%$) compared with *L. fortunei* ($-26.32 \pm 0.81\%$). An intra-guild variation was also observed for consumers with aquatic and semiaquatic herbivores (*P. canaliculata*, *Omalonix* sp., *C. aquaticum*) from $-26.57 \pm 1.31\%$ to $-27.87 \pm 0.30\%$ (Table 1). On the other hand, *H. curvispina* and *C. cfr. violaceus* exhibited the highest levels of $\delta^{15}\text{N}$ enrichment ($10.17 \pm 1.42\%$, $9.79 \pm 1.35\%$; respectively) and *L. fortunei* and Oligochaeta showed the lowest values of $\delta^{15}\text{N}$ enrichment ($7.15 \pm 1.34\%$, $7.19 \pm 0.76\%$, respectively) (Table 1).

On all data of organic matter sources (primary producers and detritus) and consumers (all taxa), no significant differences were observed in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between flooding and low water seasons (ANOVA, $p > 0.05$). Nevertheless, the basal sources analyzed were on average slightly more enriched in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ during low water levels than during high water levels. There were significant differences (ANOVA, $p < 0.001$) in the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ isotope ratios among the types of food sources and only in $\delta^{15}\text{N}$ values among primary consumers. The post hoc Tukey test of $\delta^{13}\text{C}$ values among the sources showed significant differences for C_4 plants (*P. repens* and *Echinochloa* sp.) with all the other carbon sources and between phytoplankton and *C. demersum*, *S. biloba* and epiphyton (Table 1). For the $\delta^{15}\text{N}$ values, the same test showed significant differences between BSOM and several food sources depending on the origin, e.g., epiphyton, phytoplankton, *P. stratiotes*, *L. laevigatum*, *Polygonum* sp. and SPOM (Table 1). On the other hand, $\delta^{13}\text{C}$ values did not differ significantly among primary consumers and $\delta^{15}\text{N}$ showed significant differences only among *H. curvispina*, *C. cfr. violaceus* and Oligochaeta; between *L. fortunei* and *P. canaliculata*, and between *P. canaliculata* and *P. lineatus* (Table 1).

Table 1

Stable isotope ratios of C and N and C:N ratio of sources and consumers analyzed. $\delta^{13}\text{C}$ values were arithmetically corrected for lipid content in consumers and plants. Values are mean \pm S.E.; means not sharing the same letter are significantly different from each other (post hoc Tukey test, $p < 0.05$). N = number of samples of pool of individuals, the number with asterisks number of individuals. The number of individuals in a pooled sample was 7–10 Chironominae; 3–7 *Campsurus*; 10–30 Oligochaeta; 4–6 *Omalonix*, 7–10 *Cornops*; 10–20 *Hyalella*; 20–30 *Limnoperna*.

Sources	Habitat	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	C:N	N
<i>Eichhornia crassipes</i>	C3 Floating macrophytes	-29.13 \pm 0.49 ^{b,c}	6.58 \pm 1.33 ^{a,b,c}	10.50 \pm 5.63	5
<i>Ludwigia peploides</i>	C3 Emergent macrophytes	-27.84 \pm 0.71 ^{b,c}	7.05 \pm 1.67 ^{a,b,c}	12.31 \pm 2.93	4
<i>Pistia stratiotes</i>	C3 Floating macrophytes	-29.91 \pm 0.47 ^{b,c}	7.96 \pm 1.45 ^{b,c}	12.76 \pm 1.47	4
<i>Salvinia biloba</i>	C3 Floating macrophytes	-30.75 \pm 1.82 ^c	6.85 \pm 1.01 ^{a,b,c}	9.07 \pm 7.61	3
<i>Limnobium laevigatum</i>	C3 Floating macrophytes	-28.18 \pm 1.26 ^{b,c}	8.18 \pm 1.06 ^{b,c}	9.70 \pm 0.69	4
<i>Nymphoides indica</i>	C3 Emergent macrophytes	-25.44 \pm 0.97 ^{b,c}	6.19 \pm 0.37 ^{a,b,c}	12.10 \pm 2.43	2
<i>Ceratophyllum demersum</i>	C3 Submerged macrophytes	-31.33 \pm 1.67 ^c	3.89 \pm 2.05 ^{a,b}	15.01 \pm 3.02	3
<i>Polygonum</i> sp.	C3 Emergent macrophytes	-28.18 \pm 1.25 ^{b,c}	9.06 \pm 1.78 ^c	13.75 \pm 3.16	3
Riparian tree		-26.16 \pm 0.02 ^{b,c}	3.86 \pm 0.10 ^{a,b}	10.98 \pm 2.92	2
<i>Paspalum repens</i>	C4 grass	-11.84 \pm 0.29 ^a	6.80 \pm 1.35 ^{a,b,c}	12.96 \pm 0.48	3
<i>Echinochloa</i> sp.	C4 grass	-11.98 \pm 0.06 ^a	6.82 \pm 1.39 ^{a,b,c}	12.70 \pm 0.75	3
Phytoplankton		-23.29 \pm 2.29 ^b	7.92 \pm 0.99 ^{b,c}	6.90 \pm 4.43	3
Biofilm		-29.14 \pm 2.02 ^{b,c}	5.20 \pm 4.91 ^{a,b,c}	9.38 \pm 1.20	3
Epiphyton	Attached to plants	-30.93 \pm 8.25 ^c	7.45 \pm 2.74 ^{b,c}	7.97 \pm 0.95	4
BSOM		-25.88 \pm 1.33 ^{b,c}	3.46 \pm 0.46 ^a	10.92 \pm 0.86	7
CPOM		-28.08 \pm 1.78 ^{b,c}	4.92 \pm 2.47 ^{a,b,c}	15.81 \pm 7.60	10
SPOM		-25.51 \pm 0.94 ^{b,c}	8.51 \pm 1.59 ^{b,c}	8.42 \pm 1.47	8
Consumers					
Chironominae	Benthic	-26.98 \pm 1.38	7.88 \pm 1.62 ^{a,b,c}	4.83 \pm 0.26	5
<i>Hyalella curvispina</i>	Benthic-associated to macrophytes	-23.17 \pm 3.69	10.17 \pm 1.42 ^a	5.51 \pm 0.39	4
<i>Campsurus</i> cfr. <i>violaceus</i>	Benthic	-29.07 \pm 3.50	9.79 \pm 1.35 ^a	4.75 \pm 0.07	3
<i>Prochilodus lineatus</i>	Benthic-pelagic	-27.33 \pm 3.12	9.13 \pm 0.82 ^{a,b}	4.09 \pm 0.94	12*
Oligochaeta Tubificinae	Benthic	-28.00 \pm 1.42	7.19 \pm 0.76 ^{b,c}	4.69 \pm 0.12	5
<i>Diplodon parallelipedon</i>	Infraunal	-28.67 \pm 0.64	8.60 \pm 0.62 ^{a,b,c}	5.08 \pm 0.59	3*
<i>Limnoperna fortunei</i>	Epifaunal	-26.32 \pm 0.81	7.15 \pm 1.34 ^{b,c}	4.06 \pm 0.14	3
<i>Pomacea canaliculata</i>	Benthic-associated to macrophytes	-26.57 \pm 1.31	6.50 \pm 0.71 ^c	3.83 \pm 0.37	9*
<i>Omalonix</i> sp.	Associated to floating macrophytes	-26.89 \pm 0.96	8.31 \pm 0.36 ^{a,b,c}	4.05 \pm 0.12	3
<i>Cornops aquaticum</i>	Associated to floating macrophytes	-27.87 \pm 0.30	8.07 \pm 1.26 ^{a,b,c}	3.48 \pm 0.04	3

Isotopic niche width

The isotopic niche width of the functional groups gatherer-collector (Chironominae, Oligochaeta, *C. cfr. violaceus* and *H. curvispina*), filterers (*D. parallelipedon* and *L. fortunei*) and herbivores (*P. canaliculata*, *Omalonix* sp. and *C. aquaticum*) were described.

Isotopic niche of gatherers-collectors was significantly wider than the herbivores and filterers ones ($p < 0.05$) (Fig. 1a). Among gatherer collectors, we found that *C. cfr. violaceus* and *P. lineatus* showed significantly wider isotopic niche than Chironominae ($p < 0.05$). Although *H. curvispina* showed wider isotopic niche than Chironominae and Oligochaeta, no significant differences were found. The Bayesian ellipses calculated showed a high isotopic niche overlap between Chironominae and Oligochaeta among gatherer-collector while *C. cfr. violaceus* and *H. curvispina* exhibited the highest levels of $\delta^{15}\text{N}$ (Fig. 1b). *P. lineatus* in turn showed niche overlap with the invertebrates (Fig. 1b). Within the Feeding Functional Groups (FFG) of herbivores we did not find significant differences among niche width ($p > 0.05$). The mollusks *P. canaliculata* and *Omalonix* sp. did not show isotopic niche overlap and the orthopterans *C. aquaticum* showed high levels of $\delta^{15}\text{N}$ among the herbivores (Fig. 1c). The filterers did not exhibit isotopic niche overlap and although we did not find significant differences in niche width, *L. fortunei* showed less variation in $\delta^{13}\text{C}$ values and *D. parallelipedon* slightly higher $\delta^{15}\text{N}$ values (Fig. 1d).

Isotopic contributions of the sources to consumers

The SIAR model demonstrated that C₄ macrophytes (*P. repens* and *Echinochloa* sp.) were not an important source for herbivores (Fig. 2a–c). The proportional contributions of *E. crassipes*, *L. peploides*, and *P. stratiotes* as C₃ sources, C₄ and epiphyton for herbivores (*P. canaliculata* and *Omalonix* sp.) indicated a mean dietary contribution of 37–48%, 26–29% and 26–33%, respectively

(Fig. 2a and b). The importance of C₃ plants (data from *E. crassipes*, *L. peploides*, *P. stratiotes*, and *Polygonum* sp. pooled together) to *C. aquaticum* was high, reaching a mean proportion of 90%, whereas the C₄ plants had a small dietary contribution (mean of 10%) (Fig. 2c). Likewise, the most important source for filter feeders *D. parallelipedon* and *L. fortunei* was the biofilm (mean of 58 and 44%, respectively) followed by phytoplankton (mean of 20 and 28%, respectively) and SPOM (22–28%, respectively) (Fig. 2d and e). The most important carbon sources for oligochaetes and chironomids (gatherer-collectors) were the BSOM + CPOM (mean of 40 and 54%, respectively) and the biofilm (mean 47 and 31%, respectively) with a lower contribution of epiphyton (mean of 12 and 15%, respectively) (Fig. 2f and g). On the other hand, epiphyton (40 and 45%) and BSOM + CPOM (33–34%) were important sources for *C. cfr. violaceus* and *H. curvispina* (Fig. 2h and i). The BSOM + CPOM (mean of 36%) and epiphyton (mean of 35%) were the most important sources used by *P. lineatus* (Fig. 2j).

Discussion

Isotopic signatures of sources and consumers

Macrophytes had $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ranging in concordance with values reported in the literature using the C₃ photosynthetic pathway (Smith and Epstein, 1971; Forsberg et al., 1993; Deegan and Garritt, 1997; Garcia et al., 2006). An unexpected result was that the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ signals did not show significant differences between flooding and low water seasons. However, these results could be explained by the permanent connection of the floodplain lake with the main channel of the Middle Paraná River. An earlier study focusing on fish isotope signatures in the Pantanal had shown that $\delta^{13}\text{C}$ ratios of herbivorous and detritivorous fish decreased from the wet to the dry season (Wantzen et al., 2002, 2011). In the Pantanal study, the dry season coincided with a complete isolation of the floodplain lake from the river main stem (causing a

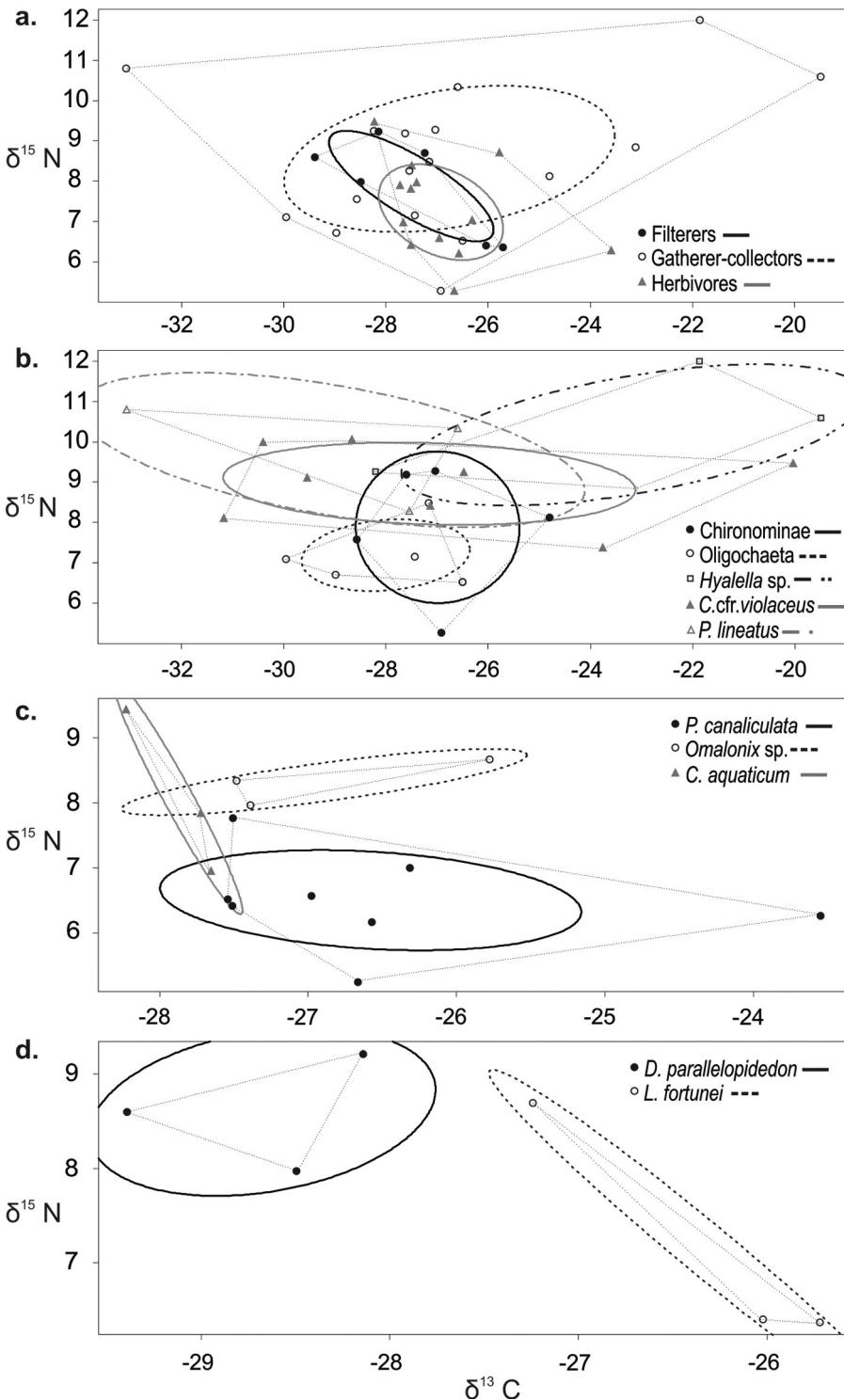


Fig. 1. Bayesian standard ellipse estimated to characterize the isotopic niche width of: (a) different feeding functional groups, (b) gatherer-collectors, (c) herbivorous and (d) filterers.

situation with the frequent reuse of available organic matter), and in the wet season the flood water arrived only partially and indirectly via a small floodplain channel from the river, whereas the flooding of the nearby meadow allowed the consumers to integrate fresh organic matter into their diets, including the direct and indirect use of C₄ plants. A study on sediment isotopes from the Upper Paraná (Gimenes et al., 2012) has shown a visible (however, not statistically significant) trend from less depleted sites near or more

directly connected to the river main stem to the more depleted sites in the more remote sections of the floodplain.

When water level increases, the allochthonous organic matter is carried to the floodplain lakes. In spite of the lack of significantly isotopic differences between seasons, the relatively enriched δ¹³C values of SPOM, CPOM, and BSOM obtained during the high water season indicated that during this season higher carbon inputs probably comes from C₄ plants and from the heavier C sediments loading

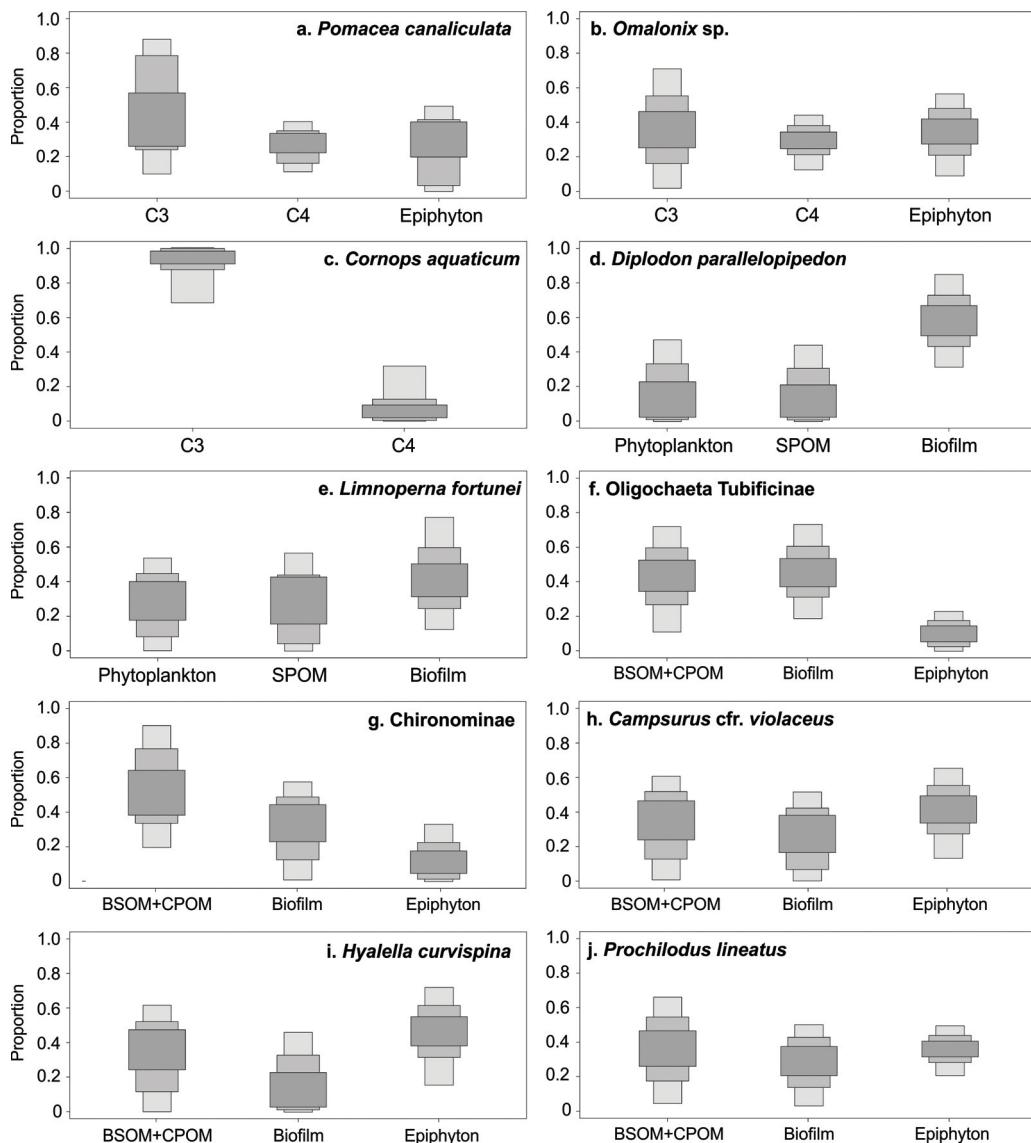


Fig. 2. Results of the Bayesian mixing model SIAR. The shaded boxes represent the 50%, 75%, and 95% credibility intervals from dark to light gray of the proportional contribution of carbon sources to (a) Gastropoda *Pomacea canaliculata*, $n=9$, (b) Gastropoda *Omalonix* sp., $n=3$, (c) Orthoptera *Cornops aquaticum*, $n=3$, (d) Bivalvia *Diplodon parallelopipedon*, $n=3$, (e) *Limnoperna fortunei*, $n=3$, (f) Oligochaeta *Tubificinae*, $n=5$, (g) Diptera *Chironominae*, $n=5$, (h) Ephemeroptera *Campsurus* cfr. *violaceus*, $n=4$, (i) Amphipoda *Hyalella curvispina*, $n=4$, and (j) Pisces *Prochilodus lineatus*, $n=12$.

into the seston. This may be supported by the associations of aquatic C₄ of genera *Echinochloa*, *Paspalum* and *Panicum* that usually cover large extension of the banks of the main channel of the Paraná River and floodplain channels (Sabattini and Lallana, 2007). Similar results were reported by Benedito-Cecílio et al. (2000) for the Amazonian floodplain and by Wantzen et al. (2002) for the Pantanal.

Isotopic niche width

We have found different levels of isotopic niche overlap and niche width among the analyzed taxa. We consider that these differences are yielded by the ecological characteristics of the organisms. Thus, the isotopic niche width of *H. curvispina*, *C. cfr. violaceus* and *P. lineatus* could be explained by their distribution on a variety of habitats, including bottom sediments and submerged and floating plants as well as by individual displacements through the water column. The genus *Campsurus* for example, is known to perform lateral migrations along with the floodpulse (Takeda and

Grzybkowska, 1996), which may be reflected in its isotope ratios and therefore, in its isotopic niche. This variety of habitats may imply a wide range of potential food sources such as BSOM, CPOM, biofilm and epiphyton. In contrast, oligochaetes and chironomids that mainly inhabit sediments and feed on surrounding surface sediments had an intrapopulation isotopic variation lower than ± 1.1 (SD). Thus, we consider that the great variation in the isotopic niche width of gatherer collectors depends upon the accessibility of different food sources with large isotopic variations. This would explain the significant differences in niche width found between *C. cfr. violaceus* and *Chironominae*.

Among the herbivores, the Orthoptera *C. aquaticum* showed the narrow range of variation in $\delta^{13}\text{C}$ in coincidence with the results of diet preference on *E. crassipes* (Capello et al., 2011). On the other hand, *P. canaliculata* and *Omalonix* sp. did not show isotopic niche overlap. Whereas $\delta^{13}\text{C}$ values of both species were similar, $\delta^{15}\text{N}$ values of *Omalonix* sp. were higher than the ones of *P. canaliculata*. This is a surprising result given that both species are considered herbivores (we assumed that this species have the same trophic

position). To interpret this result we have to consider that *P. canaliculata* feeds not only on green live parts of the plants but also on senescent tissues, whereas *Omalonix* sp. feed mainly on green parts. In addition, Fellerhoff et al. (2003) determined that $\delta^{13}\text{C}$ values of macrophytes do not change significantly along decomposition process but $\delta^{15}\text{N}$ may change largely ($\pm 6\%$). It is also known that food quality (which certainly changes along decomposition process) affects the trophic enrichment factor (TEF) of $\delta^{15}\text{N}$. Indeed, Vanderklift and Ponsard (2003) found that detritivores present lower enrichment factors than other functional feeding groups. Moreover, Kurata et al. (2001) found negative TEF for snails foraging on litter. We consider that the differences in $\delta^{15}\text{N}$ found between *P. canaliculata* and *Omalonix* sp. may be explained by the different foraging habits of these herbivores. Nevertheless, experimental testing to accurately measure TEF's of these species is needed to definitely address this issue.

The isotopic niche analysis of filterers' mussels showed a clear segregation between *D. parallelopipedon* and *L. fortunei*. This could be explained by the differences in habitat use between this species. Whereas *D. parallelopipedon* is known to be an infaunal mussel, *L. fortunei* is considered as epifaunal (Ezcurra de Drago et al., 2007). These differences in habitat use could imply correlative differences on resources uptake.

Isotopic contributions of the sources to consumers

In aquatic ecosystems, food resources for benthic invertebrates have been usually considered to be limited to detritus and microalgae in the sediment. However, in this study we have demonstrated a more diverse use of food sources not only among species, but also among individuals within populations, as also reported by Doi et al. (2010) for gastropods and by Fry (2011) for shrimp. Our results demonstrated that detritus (derived mainly from aquatic plants) and pelagic primary production contribute to supporting lake food webs, and that their relative importance varies according to the species.

The biofilm and BSOM–CPOM presented a higher contribution principally for the detritivorous oligochaetes and chironomids, because it constitutes a source with a higher colonization of bacteria and protozoans, as well as benthic algae. The detritus is an important basal food resource on which many metazoan food webs are highly dependent. This source is consumed by few fishes in temperate stream systems, but it is ingested by abundant and diverse groups of specialized fishes in tropical rivers (Winemiller, 1990; Lujan et al., 2011).

In coincidence with the results of isotopic niche analysis, we found higher contributions of epiphyton for *H. curvispina* and *C. cfr. violaceus* than for Chironomidae and Oligochaeta. We consider that the highest displacement capacity of the first ones facilitates the access to other sources such as epiphytic algae.

In relation to the analysis of source contribution to *P. lineatus*, algal contribution such as epiphyton in addition to BSOM + CPOM was the most important carbon source for this species, which was considered detritivores by different authors (Angelescu and Gneri, 1949; Bowen et al., 1984; Rossi et al., 2007). Coincidentally with our results, Lopes et al. (2007) observed that the highest carbon contribution for *P. lineatus* was C₃ plants (phytoplanktonic and periphytic algae) in the Upper Paraná River floodplain and for detritivorous Characiformes in the Amazonian River basin reported by Araújo-Lima et al. (1986a).

In the analysis of sources contribution to herbivores, we found that C₃ plants are more important than epiphyton and C₄ grasses. A study on Ampullariidae snails in the Pantanal (Fellerhoff, 2002; Fellerhoff et al., 2003) indicated that these snails can switch between C₃ and C₄ plants in their diet and Cazzaniga and Estebenet (1984) reported that *P. canaliculata* fed on plants and detritus.

According to our findings the macrophytes C₃ were the main energy sources (48%) followed by C₄ plants (26%) and epiphyton (26%) for *P. canaliculata* indicating that this species consumes a wide variety of food resources.

Filter feeders composed by mussels constitutes an important functional group in freshwater ecosystems because it transfers organic materials and nutrients from the water column to the surrounding benthic area and stimulates primary and secondary production (Howard and Cuffey, 2006; Spooner and Vaughn, 2006; Vaughn et al., 2008). *D. parallelopipedon*, a native infaunal unionid mussel, is a filter-feeding consumer that depends on microbes, algae, and detrital material available in water columns and benthic habitats. Our results imply a higher contribution of biofilm rather than phytoplankton and SPOM, for this species. This result is coincident with previous studies performed for other species of bivalves such as *Diplodon chilensis*, that feeds not only on plankton but also on benthic algae (Lara et al., 2002) and with results reported for *Corbicula fluminea* by Atkinson et al. (2010). In contrast, *L. fortunei*, an invasive epifaunal Asian clam consumes higher proportions of SPOM and phytoplankton. These findings could explain the niche segregation between these two mussels.

Conclusions

Isotopic signatures of sources did not differ between low and high water seasons. This unexpected result is explained by the connectivity characteristics of the analyzed system.

Our results demonstrated a higher contribution to primary consumers of C₃ macrophytes (mainly *E. crassipes*, *L. peploides* and *P. stratiotes*) for herbivores, biofilm and BSOM for gatherer collectors with low displacement capacity (oligochaeta and chironominae), epiphyton for more mobile collectors, whereas biofilm was the most important source for mussels and we found very low contribution of C₄ sources to herbivores. High concordance between $\delta^{13}\text{C}$ values of these primary producers and herbivores suggests that the analyzed macrophytes could constitute a major carbon source supporting aquatic primary consumers in the floodplain lake of the Middle Paraná River. In addition, the principal energy sources available for benthic organisms mainly originate from autochthonous sources and derive from the C₃ photosynthesis pathway and the pelagic primary production (mainly epiphyton) contributes to support the metazoan component of floodplain lake food webs.

Acknowledgements

We are grateful to the Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) and the Universidad Nacional del Litoral (UNL) for the financial support and the field assistance provided by E. Creus and E. Lordi. The English language was reviewed by Language Editing Services.

References

- Angelescu, V., Gneri, F.S., 1949. *Adaptaciones del aparato digestivo al regimen alimenticio de algunos peces del río Uruguay y del Río de la Plata. Tipo omnívoro e iliófago en representantes de las familias Loricariidae y Anostomidae*. Rev. Inst. Nac. Invest. Cs. Nat. Zool. 1, 161–272.
- Araújo-Lima, C.A.R.M., Forsberg, B.R., Victoria, R., Martinelli, D., 1986a. Energy sources for detritivorous fishes in the Amazon. *Science* 234, 1256–1258.
- Araújo-Lima, C.A.R.M., Portugal, L.P.S., Ferreira, E.G., 1986b. Fish-macrophyte relationship in the Anavilhas Archipelago, a blackwater system in the Central Amazon. *J. Fish Biol.* 29, 1–11.
- Atkinson, C.L., Opsahl, S.P., Covich, A.P., Golladay, S.W., Conner, L.M., 2010. Stable isotopic signatures, tissue stoichiometry, and nutrient cycling (C and N) of native and invasive freshwater bivalves. *J. N. Am. Benthol. Soc.* 29, 496–505.
- Benedito-Cecílio, E., Araújo-Lima, C.A.R.M., Forsberg, B., Bittencourt, M.M., 2000. Carbon sources of Amazonian fisheries. *Fish. Manage. Ecol.* 7, 305–315.
- Benedito-Cecílio, E., Lopes, C.A., Manetta, G.I., Gimenes, M.F., Faria, A.C.E.A., Dourado, E.C.S., Pinheiro, R.P., Martinelli, L.A., 2004. Trophic structure of the fish

- assemblage in the floodplain of the Upper Paraná River: stable isotopes. In: Agostinho, A.A., Rodrigues, L., Gomes, L., Thomaz, S.M., Miranda, L.E. (Eds.), Structure and Functioning of the Parana River and its Floodplain LTER – SITE 6 (PELD sitio 6). Eduem, Maringá, Brazil, pp. 151–156.
- Bowen, S.H., Bonetto, A.A., Ahlgren, M.O., 1984. Microorganism and detritus in the diet of a typical neotropical riverine detritivore, *Prochilodus platensis* (Pisces: Prochilodontidae). *Limnol. Oceanogr.* 29, 1120–1122.
- Brito, E.F., Moulton, T.P., De Souza, M.L., Bunn, S.E., 2006. Stable isotope analysis indicates microalgae as the predominant food source of fauna in a coastal forest stream, south-east Brazil. *Austral Ecol.* 31, 623–633.
- Capello, S., de Wysiecki, M.L., Marchese, M., 2011. Feeding patterns of the aquatic grasshopper *Cornops aquaticum* (Bruner) (Orthoptera, Acrididae) in the Middle Paraná River, Argentina. *Rev. Neotrop. Entomol.* 40, 170–175.
- Capello, S., Marchese, M., de Wysiecki, M.L., 2012. Feeding habits and trophic niche overlap of the aquatic Orthoptera associated to macrophytes. *Zool. Stud.* 51, 51–58.
- Cazzaniga, N.J., Estebenet, A.L., 1984. Revisión y notas sobre los hábitos alimentarios de los Ampullariidae (Gasteropoda). *Hist. Nat.* 4, 213–224.
- Cordiviola de Yuan, E., 1992. Fish populations of lentic environments of the Paraná River. *Hydrobiologia* 237, 159–173.
- Correa, S.B., Winemiller, K.O., 2013. Niche partitioning among frugivorous fishes in response to fluctuating resources in the Amazonian floodplain forest. *Ecology*, <http://dx.doi.org/10.1890/13-0393.1> (in press).
- Deegan, L.A., Garritt, R.H., 1997. Evidence for spatial variability in estuarine food webs. *Mar. Ecol. Prog. Ser.* 147, 31–47.
- Di Renzo, J.A., Casanoves, F., Balzarini, M.G., Gonzalez, L., Tablada, M., Robledo, C.W., 2013. InfoStat versión, 2013. Grupo InfoStat, FCA, Universidad Nacional de Córdoba, Argentina, <http://www.infostat.com.ar>
- Doi, H., Yurlova, N.-I., Kikuchi, E., Shikano, S., Yadrenkina, E.N., Vodyanitskaya, S.N., Zuykova, E.I., 2010. Stable isotopes indicate individual level trophic diversity in the freshwater gastropod *Lymnaea stagnalis*. *J. Mollus. Stud.* 76, 384–388.
- Dudgeon, D., Cheung, F.K.W., Mantel, S.K., 2010. Foodweb structure in small streams: do we need different models for the tropics? *J. N. Am. Benthol. Soc.* 29, 395–412.
- Ezcurra de Drago, I., Marchese, M., Montalto, L., 2007. Benthic invertebrates. In: Iriondo, M., Paggi, J.C., Parma, J.E. (Eds.), The Middle Paraná River: Limnology of Subtropical Wetland. Springer Verlag, Heidelberg, pp. 251–271.
- Fellerhoff, C., 2002. Feeding and growth of apple snail *Pomacea lineata* in the Pantanal wetland, Brazil – a stable isotope approach. *Isotopes Environ. Health Stud.* 38, 227–243.
- Fellerhoff, C., Voss, M., Wantzen, K.M., 2003. Stable carbon and nitrogen isotope signatures of decomposing tropic macrophytes. *Aquat. Ecol.* 37, 361–375.
- Forsberg, B.R., Araújo-Lima, C.A.R.M., Martinelli, L.A., Victoria, R.L., Bonassi, J.A., 1993. Autotrophic carbon sources for fish of the central Amazon. *Ecology* 74, 643–652.
- Füreder, L., Welter, C., Jackson, J.K., 2003. Dietary and stable isotope ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) analyses in alpine stream insects. *Int. Rev. Hydrobiol.* 88, 314–331.
- Fry, B., 2006. Stable Isotope Ecology. Springer, New York.
- Fry, B., 2011. Mississippi River sustenance of brown shrimp (*Farfantepenaeus aztecus*) in Louisiana coastal waters. *Fish. Bull.* 109, 147–161.
- Fry, B., Allen, Y.C., 2003. Stable isotopes in zebra mussels as bioindicators of river-watershed linkages. *River Res. Appl.* 19, 683–696.
- Galizzi, M.C., Zilli, F.L., Marchese, M., 2012. Diet and functional feeding groups of Chironomidae (Diptera) in the Middle Paraná River floodplain (Argentina). *Iheringia Sér. Zool.* 102, 117–121.
- Garcia, A.M., Hoeinghaus, D.J., Vieira, J.P., Winemiller, K.O., Motta Marques, D.M.L., Bembenuti, M.A., 2006. Preliminary examination of food web structure of Nicola Lake (Taim Hydrological System, south Brazil) using dual C and N stable isotope analyses. *Neot. Ichthyol.* 4, 279–284.
- Gimenes, M., Takeda, A.M., Benedito, E., 2012. The role of sediment on the energy availability for fishes in the Upper Paraná River floodplain. *Acta Sci.* 34, 391–398.
- Hamilton, S.K., Lewis Jr., W.M., 1992. Stable carbon and nitrogen isotopes in algae and detritus from the Orinoco River floodplain, Venezuela. *Geochim. Cosmochim. Acta* 56, 4237–4246.
- Hamilton, S.K., Lewis, W.M., Sippell, S.J., 1992. Energy sources for aquatic animals in the Orinoco River floodplain: evidence from stable isotopes. *Oecologia (Berlin)* 89, 324–330.
- Hamilton, S.K., Sippell, S.J., Bunn, S.E., 2005. Separation of algae from detritus for stable isotope or ecological stoichiometry studies using density fractionation in colloidal silica. *Limnol. Oceanogr. Methods* 3, 149–157.
- Howard, J.K., Cuffey, K.M., 2006. The functional role of native freshwater mussels in the fluvial benthic environment. *Freshw. Biol.* 51, 460–474.
- Jackson, A.L., Inger, R., Bearhop, S., Parnell, A., 2009. Erroneous behavior of MixSIR, a recently published Bayesian isotope mixing model: a discussion of Moore and Semmens (2008). *Ecol. Lett.* 12, 1–5.
- Jackson, A.L., Inger, R., Parnell, A.C., Bearhop, S., 2011. Comparing isotopic niche widths among and within communities: SIBER – Stable Isotope Bayesian Ellipses in *R*. *J. Anim. Ecol.* 80, 595–602.
- Jepsen, D.B., Winemiller, K.O., 2002. Structure of tropical river food webs revealed by stable isotope ratios. *Oikos* 96, 46–55.
- Jepsen, D.B., Winemiller, K.O., 2007. Basin geochemistry and isotopic ratios of fishes and basal production sources in four neotropical rivers. *Ecol. Freshw. Fish* 16, 267–281.
- Junk, W.J., Bayley, P.B., Sparks, R.E., 1989. The flood pulse concept in river-floodplain systems. In: Dodge, D.P. (Ed.), Proceedings of the International Large River Symposium (LARS). Canadian Special Publication Fisheries Aquatic Sciences 106. NRC Research Press, Ottawa, pp. 110–127.
- Junk, W.J., Wantzen, K.M., 2004. The flood pulse concept: new aspects, approaches, and applications – an update. In: Welcomme, R., Petr, T. (Eds.), Proceedings of the Second International Symposium on the Management of Large Rivers for Fisheries, Food and Agriculture Organization and Mekong River Commission. FAO Regional Office for Asia and the Pacific, Bangkok, pp. 117–149 (RAP Publication 2004/16).
- Junk, W.J., Wantzen, K.M., 2006. Flood pulsing, and the development and maintenance of biodiversity in floodplains. In: Batzer, D.P., Sharitz, R.R. (Eds.), Ecology of Freshwater and Estuarine Wetlands. University of California Press, Berkeley, pp. 407–435.
- Kurata, K., Minami, H., Kikuchi, E., 2001. Stable isotope analysis of food sources for salt marsh snails. *Mar. Ecol. Prog. Ser.* 223, 167–177.
- Kurle, C.M., 2009. Interpreting temporal variation in omnivore foraging ecology via stable isotope modeling. *Funct. Ecol.* 23, 733–744.
- Lara, G., Parada, E., Peredo, S., 2002. Alimentación y conducta alimentaria de la almeja de agua dulce *Diploodon chilensis* (Bivalvia Hyriidae). *Gayana Zool.* 66, 107–112.
- Layman, C.A., Araujo, M.S., Boucek, R., Hammerschlag-Peyer, C.M., Harrison, E., Jud, Z.R., Matisch, P., Rosenblatt, A.E., Vaudo, J.J., Yeager, L.A., Post, D.M., Bearhop, S., 2012. Applying stable isotopes to examine food-web structure: an overview of analytical tools. *Biol. Rev.* 87, 545–562.
- Lewis Jr., W.M., Hamilton, S.K., Rodríguez, M.A., Saunders III, J.F., Lasi, M.A., 2001. Foodweb analysis of the Orinoco floodplain based on production estimates and stable isotope data. *J. N. Am. Benthol. Soc.* 20, 241–254.
- Lopes, C.A., Benedito-Cecílio, E., Martinelli, L.A., Moreira, M.Z., 2007. Energy sources for explorator of bottom fish, *Prochilodus lineatus* (Prochilodontidae, Characiformes), in neotropical region. *J. Fish Biol.* 70, 1649–1659.
- Lopes, C.A., Benedito, E., Martinelli, L.A., 2009. Trophic position of bottom-feeding fish in the Upper Paraná River floodplain. *Braz. J. Biol.* 69, 573–581.
- Lujan, N.K., Donovan, P.G., Winemiller, K.O., 2011. Do wood-grazing fishes partition their niche? morphological and isotopic evidence for trophic segregation in Neotropical Loriciariidae. *Funct. Ecol.* 25, 1327–1338.
- Martinelli, L.A., Victoria, L., Forsberg, B.R., 1994. Isotopic composition of majors carbon reservoirs in the Amazon floodplain. *Int. J. Ecol. Environ. Sci.* 20, 31–46.
- Martínez del Río, C., Wolf, N., Carleton, S.A., Gannes, L.Z., 2009. Isotopic ecology ten years after a call for more laboratory experiments. *Biol. Rev.* 84, 91–111.
- Matthews, B., Mazumder, A., 2005. Temporal variation in body composition (C:N) helps explain seasonal patterns of zooplankton d13C. *Freshw. Biol.* 50, 502–515.
- McConaughey, T., McRoy, C.P., 1979. Food-web structure and the fractionation of carbon isotopes in the Bering Sea. *Mar. Biol.* 53, 257–262.
- Mesa, L., Marchese, M.R., Montalto, L., Zilli, F.L., 2012. Bidirectional exchanges of benthic invertebrates in a large river-floodplain system (Paraná River, Argentina). *Ann. Limnol. Int. J. Limnol.* 48, 425–436.
- Minagawa, M., Wada, E., 1984. Stepwise enrichment of ^{15}N along food chains: further evidence and the relation between ^{15}N and animal age. *Geochim. Cosmochim. Acta* 48, 1135–1140.
- Moore, J.W., Semmens, B.X., 2008. Incorporating uncertainty and prior information into stable isotope mixing models. *Ecol. Lett.* 11, 470–480.
- Newsome, S.D., Martínez del Río, C., Bearhop, S., Phillips, D.L., 2007. A niche for isotopic ecology. *Front. Ecol. Environ.* 5, 429–436.
- Newsome, S.D., Yeakel, J.D., Wheatley, P.V., Tinker, M.T., 2012. Tools for quantifying isotopic niche space and dietary variation at the individual and population level. *J. Mammal.* 93, 329–341.
- O'Callaghan, M.J., Hannah, D.M., Boomer, I., Williams, M., Sadler, J.P., 2013. Responses to river inundation pressures control prey selection of riparian beetles. *PLoS ONE* 8 (4), e61866, <http://dx.doi.org/10.1371/journal.pone.0061866>.
- Paetzold, A., Bernet, J.F., Tockner, K., 2006. Consumer-specific responses to riverine subsidy pulses in a riparian arthropod assemblage. *Freshw. Biol.* 51, 1103–1115.
- Paetzold, A., Schubert, C.J., Tockner, K., 2005. Aquatic-terrestrial linkages along a braided river: riparian arthropods feeding on aquatic insects. *Ecosystems* 8, 748–759.
- Parnell, A., Inger, R., Bearhop, S., Jackson, A.L., 2008. SIAR: Stable Isotope Analysis in R, <http://cran.r-project.org/web/packages/siar/index.html>
- Parnell, A., Inger, R., Bearhop, S., Jackson, A.L., 2010. Source partitioning using stable isotopes: coping with too much variation. *PLoS ONE* 5, e9672.
- Peterson, B.J., Fry, B., 1987. Stable isotopes in ecosystem studies. *Annu. Rev. Ecol. Syst.* 18, 293–320.
- Phillips, D.L., Gregg, J.W., 2003. Source partitioning using stable isotopes: coping with too many sources. *Oecologia (Berlin)* 136, 261–269.
- Post, D.M., 2002. Using stable isotopes to estimate trophic position: models, methods, and assumptions. *Ecology* 83, 703–718.
- Post, D.M., Layman, C.A., Arrington, D.A., Takimoto, G., Quattrochi, J., Montaña, C., 2007. Getting to the fat of the matter: models, methods and assumptions for dealing with lipids in stable isotope analyses. *Oecologia* 152, 179–189.
- Rossi, L.M., Cordiviola, E., Parma, M.J., 2007. Fishes. In: Iriondo, M., Paggi, J.C., Parma, M.J. (Eds.), The Middle Paraná River: Limnology of Subtropical Wetland. Springer Verlag, Heidelberg, pp. 305–326.
- Rubenstein, D.R., Hobson, K.A., 2004. From birds to butterflies: animal movement patterns and stable isotopes. *Trends Ecol. Evol.* 19, 256–263.
- Sabattini, R.A., Lallana, V.H., 2007. Aquatic macrophytes. In: Iriondo, M., Paggi, J.C., Parma, M.J. (Eds.), The Middle Paraná River: Limnology of Subtropical Wetland. Springer Verlag, Heidelberg, pp. 205–226.
- Saigo, M., Marchese, M., Montalto, L., 2009. Hábitos alimentarios de *Hyalella curvispina* Shoemaker, 1942 (Amphipoda: Gammaridea) en ambientes leníticos de la llanura aluvial del río Paraná Medio. *Nat. Neotrop.* 40, 43–59.
- Smith, B.N., Epstein, S., 1971. 2 Categories of C-13/C-12 ratios for higher plants. *Plant Physiol.* 47, 380–385.

- Solomon, C.T., Carpenter, S.R., Clayton, M.K., Cole, J.J., Coloso, J.J., Pace, M.L., Van der Zanden, M.J., Weidel, B.C., 2011. *Terrestrial, benthic, and pelagic resource use in lakes: results from a three-isotope Bayesian mixing model*. *Ecology* 92, 1115–1125.
- Spooner, D.E., Vaughn, C.C., 2006. Context-dependent effects of freshwater mussels on stream benthic community. *Freshw. Biol.* 51, 1016–1024.
- Takeda, A.M., Grzybowska, M., 1996. Seasonal dynamics and production of *Campus violaceus* nymphs (Ephemeroptera, Polymitarcyidae) in the Baia River, Upper Parana River floodplain, Brazil. *Hydrobiologia* 356, 149–155.
- Thorp, J.H., Delong, M.D., 1994. The Riverine Productivity Model – an heuristic view of carbon-sources and organic-processing in large river ecosystems. *Oikos* 70, 305–308.
- Thorp, J.H., Delong, M.D., 2002. Dominance of autotrophic autochthonous carbon in food webs of heterotrophic rivers. *Oikos* 96, 543–550.
- Thorp, J.H., Delong, M.D., Greenwood, K.S., Casper, A.F., 1998. Isotopic analysis of three food web theories in constricted and floodplain regions of a large river. *Oecologia* 117, 551–563.
- Vanderklift, M.A., Ponsard, S., 2003. Sources of isotopic variation in consumer-diet $\delta^{15}\text{N}$ enrichment: a meta-analysis. *Oecologia* 136, 169–182.
- Vannote, R.L., Minshall, G.W., Cummins, R.W., Sedell, J.R., Cushing, C.E., 1980. The river continuum concept. *Can. J. Fish. Aquat. Sci.* 37, 130–137.
- Vaughn, C.C., Nichols, S.J., Spooner, D.E., 2008. Community and foodweb ecology of freshwater mussels. *J. N. Am. Benthol. Soc.* 27, 409–423.
- Wantzen, K.M., Junk, W.J., 2006. Aquatic-terrestrial linkages from streams to rivers: biotic hot spots and hot moments. *Arch. Hydrobiol. Suppl.* 158, 595–611.
- Wantzen, K.M., Machado, F.A., Voss, M., Boriss, H., Junk, W.J., 2002. Seasonal isotopic shifts in fish of the Pantanal wetland, Brazil. *Aquat. Sci.* 64, 239–251.
- Wantzen, K.M., Fellerhoff, C., Voss, M., 2011. Stable isotope ecology of the Pantanal. In: Junk, W.J., da Silva, C.J., da Cunha, C.N., Wantzen, K.M. (Eds.), *The Pantanal: Ecology, Biodiversity and Sustainable Management of a Large Neotropical Seasonal Wetland*. Pensoft Publishers, Sofia, pp. 597–617.
- Welch, P.S., 1963. *Limnological Methods*. McGraw-Hill, New York.
- Winemiller, K.O., 1990. Spatial and temporal variation in tropical fish trophic networks. *Ecol. Monogr.* 60, 331–367.