BIVALVE CONTRIBUTION TO SHALLOW SANDY BOTTOM FOOD WEB OFF MAR DEL PLATA (ARGENTINA): INFERENCE FROM STOMACH CONTENTS AND STABLE ISOTOPE ANALYSIS

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ABSTRACT Two infaunal species, the purple clam Amiantis purpurata and the razor clam Solen tehuelchus, are common species in the 15–20 m sandy bottom sediments between southern Brazil and central Argentina. Both species are food sources for the coastal food web, but the extent of their contribution to this food web is still unknown. Based on stomach content analysis and $\delta^{15}N$ and $\delta^{15}C$ stable isotope signatures we explored the position of these clams in the food web. Stomach content shows that the ray Sympterigia bonapartei and the white croaker Micropogonias furnieri prey on entire A. purpurata and S. tehuelchus. The stable isotopic analysis confirms this result. The elephant fish Callorynchus callorynchus and the Brazilian codling Urophycis brasiliensis also show values of C and N consistent with those expected from species that are preying on A. purpurata. Based on the prediction from the isotopic analysis the clam A. purpurata is a food source for the shrimps Artemesia longinaris, Pleoticus muelleri, the anemone Antholoba achates and the gastropod Buccinanops monilifer. The gastropods Adelomelon brasiliana, Olivancillaria deshayesiana, Olivancillaria urceus and Zidona dufresnei also show evidence of consuming A. purpurata but with contribution from other species with heavier C content. According to complementary results the razor clam Solen tehuelchus may be this other species. The stable isotopic analysis shows that both clam species are at the base of the consumers in the food web. A. pupurata showed lighter C mark than S. tehuelchus, but the N isotopic mark showed higher value but still being within the same trophic level.

KEY WORDS: food web, bivalves, fish, crustaceans, gastropods, SW Atlantic

INTRODUCTION

Two infaunal bivalves, the purple clam Amiantis purpurata (Lamarck, 1818) and the razor clam Solen tehuelchus (D'Orbigny 1843), are common species in the coastal (≤20-m depth) sandy bottom of the SW Atlantic. The purple clam is distributed from Espírito Santo, Brazil (20°S) to the Golfo San Matías, Argentina (42°S) (Carcelles 1944, de Castellanos 1967, Scarabino 1977) occurring in high-density patches (maximum abundance: 632 clams m⁻²) at depths ranging from 10–18 m dominated by 1- or 2-yearclasses (Morsan 2003). The razor clam S. tehuelchus (≤61-mm length) shows similar distribution, from Río de Janeiro, Brazil (22°54'S) to Bahía Blanca, Argentina (38°44'S) (Rios 1994, Capitoli 1997). However, the abundance of this species is not truly reflected in the literature, probably because of their deep burrowing habits. There is evidence that both species are important food sources for a number of species in shallow coastal areas but the extent of their contribution to the food web is still unknown.

Several predators have been described for these species based on gut content analysis (Olivier et al. 1968, Radonic 1997). For example, siphon tips of *A. purpurata* are the main food item of the electric ray *Discopyge tschudii*, Heckel 1846 (Arrighetti et al. 2005), which is the regionally most abundant torpediniform species. Also, entire clams are preyed by the fish *Pogonias cromis* (Linnaeus, 1766) (García & Gianuca 1997). However, gut content analysis is limited and may be biased toward larger species that can hold the bivalves for longer time (see Sutela & Huusko 2000, Hyslop 1980) and toward food items recently consumed; moreover, this method cannot provide information on the rate of ingestion and regarding long-term assimilation (Creach et al. 1997).

As an alternative, measurement of stable isotope ratios has

become widely used to define relationships between consumers and their food sources (Peterson & Fry 1987, Michener & Schell 1994, Cabana & Rasmussen 1994), and they have been applied to study foraging, migration and other life history phenomena (Hesslein et al. 1991, Alisauskas & Hobson 1993, Walker et al. 1999, Best & Schell 1996, Hansson et al. 1997, Kline et al. 1998, Griffin & Valiela 2001, McGinnis & Emslie 2001). Stable isotopes are relatively consistently fractionated by biological and physical processes as they pass through food webs becoming heavier by 3% to 4% for N and by 1% to 2% for C with each trophic transfer (Peterson & Fry 1987, Cabana & Rasmussen 1994). This fractionation from potential food sources to tissues in consumers has been used to identify trophic relationships. Trophic relationships between motile consumers like fishes and their food sources may depend on size of consumers and foraging patterns. Larger animals are able to feed higher in the food web and often acquire heavier isotopic signatures in their tissues (France et al. 1998, Harvey et al. 2002). Therefore, combining isotope analysis with gut contents can provide a better understanding of trophic pathways, given that some items could be ingested but not assimilated. Also, isotope analysis can provide information on those organisms in which it is not possible to evaluate gut content.

In this study, we used stomach content analysis whenever possible and $\delta^{15}N$ and $\delta^{13}C$ stable isotope signatures to define the position of the purple clam *Amiantis purpurata* and the razor clam *Solen tehuelchus* in the food web of the shallow northern Argentinean costal ecosystem.

MATERIAL AND METHODS

Sampling and Study Areas

The study was performed at the coastal area of northern Argentina (38°20'S) between April 2002 and December 2002. This

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area is dominated by sandy and sandy-silty bottoms (Cortelezzi in Olivier et al. 1968). The community studied is typical of a temperate marine environment where few taxa are dominant over a large number of occasional species. The low diversity of the infaunal assemblages has been described (Olivier et al. 1968, Scelzo et al. 2002). Samples of the two bivalves and of the most representative and abundant organisms coexisting with them were obtained from the study area with bottom trawls using a 2-cm cup net from water depth of 10-18 m. The species sampled were the fishes Urophycis brasiliensis (Kaup 1858), Cynoscion striatus (Cuvier 1829), Sympterygia bonapartei (Müller & Henle 1841), Micropogonias furnieri (Desmarest 1823) and Callorynchus callorynchus (Meuschen 1778); the gastropods Buccinanops monilifer (Kiener 1834) Adelomelon brasiliana (Lamark 1811), Olivancillaria deshayesiana (Duclos 1857), Olivancillaria urceus (Roding 1798), and Zidona dufresnei (Donovan 1823); the anemone Antholoba achates (Drayton 1984) and the shrimps Artemesia longinaris (Bate 1888) and Pleoticus muelleri (Bate 1888).

Stomach Content Analysis

The stomach of 15 *Urophycis brasiliensis*, 35 *Cynoscion striatus*, 11 *Sympterygia bonapartei* and 42 *Micropogonias furnieri* were injected aboard with 10% formaline to stop the digestive process. In the laboratory, the total length of each fish was measured to the nearest mm. To analyze the stomach contents, the digestive tract anterior to the intestine was removed and preserved in 10% formaline. Stomach contents were observed under a dissecting microscope. Additional information was made on the gastro-vascular cavity of *Antholoba achates* and its associated snail *Adelomelon brasiliana*. This information was complemented by reported information from the same area (i.e., Olivier et al. 1968, Radonic 1997).

Stable Isotopes Analysis

Samples of white dorsal muscle of all fish species were removed and frozen for stable isotopic analysis. Samples were also

obtained from the epibenthic fauna (5 pools of 5 individuals each) for the same analysis. All these samples were dried at 60°C, tissues were ground using a mortar and pestle, and all samples were sent to the University of California-Davis Stable Isotope Facility to determine δ^{13} C and δ^{15} N signatures by mass spectrometry.

RESULTS AND DISCUSSION

Stable isotope analysis shows three trophic levels based on expected N fractionation (3.4%). The two bivalves Solen tehuelchus and Amiantis purpurata are at the base of the food web, with δ^{15} N values between 10.9% and 12.3%. S. tehuelchus has δ^{13} C values more depleted than A. purpurata but similar N isotope ratios. The fishes Callorynchus callorynchus and Cynoscion striatus and the squid Loligo gahi (d'Orbigny 1835) are in the upper trophic level with δ^{13} N values between 16.7% and 17.5%. In our study system, stomach content analysis can only be reliable with fishes and anemones. Based on information previously reported and our sampling (Table 1), the ray Sympterigia bonapartei prey on siphons of Amiantis purpurata and S. bonapartei. A similar result was previously found for other ray species, the electric ray Discopyge tschudii (Arrighetti et al. 2005). S. bonapartei showed C and N isotopic values expected from a clam diet, but N isotope ratios showed high variability and could be the result of a mixed diet of both clam species and some contribution of Artemesia longinaris (see Fig. 1, Table 1).

Stomach contents showed that the white croaker *Micropogonias furnieri* prey on entire individuals of *A. purpurata* and *Solen tehuelchus* (Table 1). This fish species showed different isotopic values between juveniles (<30 cm in length) and big individuals (Fig. 1), suggesting a shift in diet during their life cycle. Juveniles have values more depleted in C suggesting a food source other than clams, probably crustaceans or polychaetes (Olivier et al. 1968, Radonic 1997 and stomach content results). Adults, however, showed values that suggest an influence of clams. This shift in diet between sizes for this species was found for other sites (Olivier et al. 1968).

 $\label{eq:TABLE 1.}$ Biomass of the prey items identified in the stomach content analysis (%)

Prey items	C. striatus			U. brasiliensis		S. bonapartei		M. furnieri		
	Olivier et al. 1968	Radonic 1997	Ours	Olivier et al. 1968	Ours	Olivier et al. 1968	Ours	Olivier et al. 1968	Radonic 1997	Ours
Crustacea										
Artemesia longinaris	5		17.5	46.7	6	21.6	7	22.4		
Pleoticus muelleri	0.6		0.5	5.1	1	4.2	10			
Other	93	77	69.2	43.8	75	62	71	57.2	45	37.3
Polychaete	1.2	1	2.7			5.4		6.1	13	
Ostheichthyes	2	2		4.4	18	6.8		6.1	1	
Mollusca										
Bivalve										
Amiantis pupurata							4			34.7
Solen tehuelchus							3			28
Other								6.1		
Gastropoda										
Buccinanops monilifer								2		
Cephalopoda										
Loligo gahi									41	
Cnidaria		3								
Chaetognata		17								
Others			8.1							

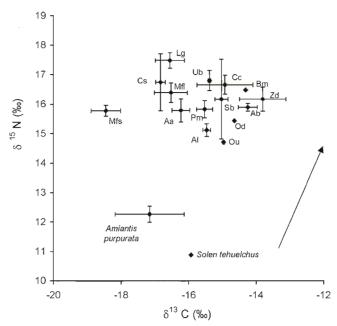


Figure 1. δ¹⁵N versus δ¹³C (mean ± 1 SE) for the subtidal sandy bottom food web. Olivancillaria urceus (Ou), Olivancillaria deshayesiana (Od), shrimp Artemesia longinaris (Al), anemone Antholoba achates (Aa), white croacker Micropogonias furnieri small (Mfs) and large (Mfl), shrimp Pleoticus muelleri (Pm), gastropod Adelomelon brasiliana (Ab), gastropod Zidona dufresnei (Zd), gastropod Buccinanops monilifer (Bm), ray Sympterigia bonapartei (Sb), Brazilian codling Urophycis brasiliensis (Ub), elephant fish Callorynchus callorynchus (Cc), stripped weakfish Cynoscion striatus (Cs), squid Loligo gahi (Lg). The arrow shows expected enrichment in one trophic level.

The Brazilian codling *Urophycis brasiliensis*, and the stripped weakfish *Cynoscion striatus* (Olivier et al. 1968, Radonic 1997 and our results; Table 1), did not show clams in their stomachs. Carbon isotope ratios of the stripped weakfish *C. striatus* showed no evidence of preying on these clams (Fig. 1). The elephant fish *Callorynchus callorynchus* and the Brazilian codling *U. brasiliensis* show values of C consistent with those expected from species that are preying on the purple clam *A. purpurata*, but higher N signatures. This could be the result of a mix diet of items in more than one trophic level for example; in this case they could be feeding on shrimps (as seen by stomach contents) and having some contribution of clams.

For invertebrates it is much more difficult, or just impossible, to recognize items in the stomach content by microscopical analysis. Based on the prediction from the isotopic analysis the clam *Amiantis purpurata* is a food source for the shrimps *Artemesia*

longinaris and Pleoticus muelleri, and the gastropod Buccinanops monilifer. However, the gastropods Adelomelon brasiliana, Olivancillaria deshayesiana, Olivancillaria urceus and Zidona dufresnei also show evidence of consuming the purple clam but with contribution from the razor clam Solen tehuelchus (Fig. 1).

One of the few invertebrate species where items in the stomach content can be recognized is the anemone Antholoba achates (see Acuña et al. 2003). Our results show that 60% of the weight was Amiantis purpurata, whereas the other species contributed with much lower values (Solen tehuelchus 5%, Encope emarginata 11.5%, Artemesia longinaris 11.5%, Olivancelaria uretai, O. Urceus and Buccinanops moniliferum 10%, the hermit crab Pagurus excilis 0.6%, Leucipa patagonica 0.4% and polychaetes 1.1%). Isotope C and N also showed values expected for a diet mainly on A. purpurata. Interestingly, this anemone lives epibiotic on the gastropod Adelomelon brasiliana (see Acuña et al. 2003). Adelomelon brasiliana is a common inhabitant of the studied area, which preys on Amiantis purpurata and Solen tehuelchus of all sizes (Cledón 2005). These snails are long-lived and can reach over 200 mm in length (Cledón et al. 2005), which means a high predatory pressure for the bivalves, because no size is out of predation risk and their burrowing in the sediment is not always effective, the snails can also do that (Cledón 2005). The anemone and the snail are at the same trophic level (i.e., same n values) but C isotope ratios suggest that their food sources are somewhat different (i.e., the C mark differ). Whereas the anemone showed δ^{13} C values expected of a predator feeding on A. purpurata and no on S. tehuelchus, the gastropod A. brasiliana showed δ^{13} C values suggesting a diet that incorporates both bivalves at similar proportion. The sea anemone and the snail, although highly associated, do not show evidence of large diet overlap. This fact could be related to the burrowing activity of A. brasiliana, whereas A. achates would remain in contact with the upper sediment levels (Arrighetti et al. 2004).

In conclusion, the purple clam *Amiantis purpurata* and the razor clam *Solen tehuelchus* are species clearly located at the bottom of the food web. These two species contribute to the diet of some gastropods and sea anemones and to some economically important fish.

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LITERATURE CITED

Acuña, F. H., A. C. Excoffon & M. A. Scelzo. 2003. Mutualism between the sea anemone *Antholoba achates* (Drayton 1846) (Cnidaria: Actiniaria: Actinostolidae) and the spider crab *Libinia spinosa* Milne-Edwards, 1834 (Crustacea: Decapoda, Majidae). *Belg. J. Zool.* 133:85–87.

Alisauskas, R. T. & K. A. Hobson. 1993. Determination of lesser snow goose diets and winter distribution using stable isotope analysis. J. Wildl. Manage. 57:49–54.

Arrighetti, F., J. P. Livore & P. E. Penchaszadeh. 2004. Ecología trófica de la anémona de mar *Antholoba achates* en la zona de Mar del Plata. In: II Reunión Binacional de Ecología. Mendoza, Argentina. pp. 158.

Arrighetti, F., J. P. Livore & P. E. Penchaszadeh. 2005. Siphon nipping of

the bivalve *Amiantis purpurata* by the electric ray *Discopyge tschudii* (Heckel 1846) in Mar del Plata, Argentina. *J. Mar. Biol. Ass. U.K.* 85:1151–1154.

Best, P. B. & D. M. Schell. 1996. Stable isotopes in southern right whale (Eubalaena australis) baleen as indicators of seasonal movements, feeding and growth. Mar. Biol. 124:483–494.

Cabana, G. & J. B. Rasmussen. 1994. Modeling food chain structure and contaminant bioaccumulation using stable nitrogen isotopes. *Nature* 372:255–257.

Capitoli, R. R. 1997. Continental Shelf Benthos. In: U. Seeliger, C. Odebrecht & J. P. Castello, editors. Subtropical convergence environments:

- the coast and sea in the southwestern Atlantic. Springer-Verlag, Berlin. pp. 117–120.
- Carcelles, A. 1944. Catálogo de moluscos marinos de Puerto Quequén (R. Argentina). Revista del Museo La Plata, Nueva Serie. Sección Zoología. 3:233–309.
- Cledón, M. 2005. Reproductive biology and ecology of Adelomelon brasiliana off Mar del Plata, Argentina. Berichte für Polar und Meeresforschung. Bremerhaven, Germany. 100 pp.
- Cledón, M., T. Brey, P. E. Penchaszadeh & W. Arntz. 2005. Individual growth and somatic production in *Adelomenlon brasiliana* (Gastropoda; Volutidae) off Argentina. *Mar. Biol.* 147:447–452.
- Creach, V., M. T. Schricke, G. Bertru & A. Mariotti. 1997. Stable isotopes and gut analyses to determine feeding relationships in saltmarsh macroconsumers. *Estuar. Coast. Shelf Sci* 44:599–611.
- de Castellanos, Z. A. 1967. Catálogo de moluscos marinos bonaerenses. Anales de la Comisión de Investigaciones Científicas 8:1-365.
- France, R., M. Chandler & R. Peters. 1998. Mapping trophic continuity of benthic food webs: body size-d15 N relationships. Mar. Ecol. Prog. Ser. 174:301–306.
- García, V. M. T. & N. M. Gianuca. 1997. Relationships and function of coastal and marine environments. In: U. Seeliger, C. Odebrecht & J. P. Castello, editors. Subtropical convergence environments: the coast and sea in the southwestern Atlantic. Berlin: Springer-Verlag. 308 pp.
- Griffin, M. P. A. & I. Valiela. 2 001. δ¹⁵N isotope studies of life history and trophic position of *Fundulus heteroclitus* and *Menidia menidia*. *Mar. Ecol. Prog. Ser.* 214:299–305.
- Hansson, S., J. E. Hobbie, R. Elmgren, U. Larsson, B. Fry & S. Johansson. 1997. The stable nitrogen isotope ratio as a marker of food-web interactions and fish migration. *Ecology* 78:2249–2257.
- Harvey, C. J., P. C. Hanson, T. E. Essington, P. B. Brown & J. Kitchell. 2002. Using bioenergetics models to predict stable isotope ratios in fishes. Can. J. Fish. Aquat. Sci. 59:115–124.
- Hesslein, R. H., M. J. Capel, D. E. Fox & K. A. Hallard. 1991. Stable isotopes of sulfur, carbon, and nitrogen, as indicators of trophic level and fish migration in the lower Mackenzie River Basin, Canada. *Can. J. Fish. Aquat. Sci.* 48:2258–2265.

- Hyslop, E. J. 1980. Stomach contents analysis—a review of methods and their application. J. Fish Biol. 17:411-429.
- Kline, T. C., Jr., W. J. Wilson & J. J. Goering. 1998. Natural isotope indicators of fish migration at Prudhoe Bay, Alaska. Can. J. Fish. Aquat. Sci. 55:1494–1502.
- Michener, R. H. & M. Schell. 1994. Stable isotope ratios as tracers in marine aquatic food webs. In: K. Lajtha & R. H. Michener, editors. Stable isotopes in ecology and environmental science. Blackwell Scientific. pp. 138–158.
- McGinnis, T. W. & S. D. Emslie. 2001. The foraging ecology of royal and sandwich terns in North Carolina, USA. *Waterbirds* 3:361–370.
- Morsan, E. M. 2003. Spatial analysis and abundance estimation of the southernmost population of the purple clam, *Amiantis purpurata* in Patagonia, Argentina. J. Mar. Biol. Ass. U.K. 83:1073–1082.
- Olivier, S. R., M. R. Torti & R. Bastida. 1968. Ecosistema de las aguas litorales. *Servicio de Hidrografía Naval. H.* 1025. 46 pp.
- Peterson, B. J. & B. Fry. 1987. Stable isotopes in ecosystem studies. Ann. Rev. Ecol. Syst. 18:293–320.
- Radonic, M. 1997. Diversidad, abundancia y aspectos tróficos de la fauna icticola acompañante de la captura del camarón y langostino, frente a las costas de Mar del Plata. Tesis de grado, Universidad Nacional de Mar del Plata. 53 pp.
- Rios, E. 1994. Seashells of Brazil. Rio Grande: FURG. 492 pp.
- Scarabino, V., 1977. Moluscos del Golfo San Matías (Prov. Río Negro, Argentina). Inventario y claves para su identificación. Comunicaciones de la Sociedad Malacológica del Uruguay IV (31–32). pp. 177–297.
- Scelzo, M. A., J. Martinez Arca & N. M. Lucero. 2002. Diversidad, densidad y biomasa de la macrofauna componente de los fondos de pesca "camarón-langostino", frente a Mar del Plata, Argentina (1998–1999). Rev. Invest. Desarr. Pesa. 15:43–65.
- Sutela, T. & A. Huusko. 2000. Varying resistance of zooplankton prey to digestion: implications for quantifying larval fish diets. *Trans. Am. Fish. Soc.* 129:545–551.
- Walker, J. L., C. W. Potter & S. A. Macko. 1999. The diets of modern and historic bottlenose dolphin populations reflected through stable isotopes. *Mar. Mamm. Sci.* 15:335–350.