Estuaries Vol. 28, No. 5, p. 714–725 October 2005

Can the Nitrogen and Carbon Stable Isotopes of the Pygmy Mussel, *Xenostrobus securis*, Indicate Catchment Disturbance for Estuaries in Northern New South Wales, Australia?

STEPHANIE K. MOORE* and IAIN M. SUTHERS

Fisheries and Marine Environmental Research Laboratory, School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, New South Wales 2052, Australia

ABSTRACT: The nitrogen and carbon stable isotope ratios ($\delta^{15}N$ and $\delta^{13}C$) of the pygmy mussel, *Xenostrobus securis*, were determined for three estuaries with varying levels of catchment disturbance in northern New South Wales, Australia. The lower Manning River catchment supported the highest human population densities and 3% residential development with some livestock agriculture (41%); the Wallamba River catchment was mostly livestock agriculture (56%) while the Wallingat River catchment was mostly vegetated (79%). Mussels, estuarine particulate organic matter (POM), and livestock and human-derived waste were collected in two stages during the austral summers of 2001–2002 and 2002–2003 for dual carbon-nitrogen stable isotope analysis. The disturbed Manning and Wallamba River catchment mussels were enriched in ^{15}N by an average of 3.2‰ and 1.5‰, respectively, compared to the vegetated Wallingat River mussels. Mussel $\delta^{13}C$ values ranged from −24.8‰ to −30.3‰ and showed an estuarine gradient becoming enriched with distance downstream within estuaries, but were unable to distinguish patterns in catchment disturbance between estuaries. The $\delta^{15}N$ and $\delta^{13}C$ values of POM showed a similar pattern to mussels, indicating a direct link between them within each estuary. A multiple regression model of mussel $\delta^{15}N$ using the fractions of land used for livestock agriculture and residential development within 5 km zones from river networks to a distance equivalent to a tidal ellipse from sites explained 67% of the variation in mussel $\delta^{15}N$ with 95% of the differences lying within 1.6‰ of observed values. Increasing fractions of land used for livestock agriculture depleted mussel $\delta^{15}N$ values estimated by the regression equation, indicating the use of cow manure as a nutrient source with a value of 2.0‰. Increasing fractions of land used for residential development enriched estimated mussel $\delta^{15}N$, indicating the use of human-derived waste with a value of 20.8‰. Pygmy mussels are a useful long-term bio-indicator for the effects of anthropogenic catchment disturbance and nutrient enrichment in estuaries.

Introduction

Anthropogenic alteration of catchments from native vegetation to residential and agricultural lands has led to increased quantities of nutrients entering coastal systems (Carpenter et al. 1998; Meeuwig 1999; Nedwell et al. 2002; Paterson et al. 2003; Scharler and Baird 2003). It is widely accepted that this can cause deteriorating water quality and persistent phytoplankton blooms, leading to large scale changes in ecosystem structure and function from eutrophication (de Jonge and Elliott 2001; de Jonge et al. 2002). The resilience of coastal systems to eutrophication depends on a number of system-specific physical and biological attributes, such as water residence times, light regimes, and the abundance of suspension-feeding grazers (see Cloern 2001).

Monitoring programs designed to detect early responses to increased nutrient loading are hampered by high levels of variability associated with estuarine environments. Tides, rainfall, wind, and temperature cycles can alter water column nutrient and chlorophyll concentrations. Bioindicators can integrate this patchiness in physicochemical properties over time. Bivalve molluscs are recognized as suitable bioindicators because they accumulate and incorporate chemicals and elements into their tissues from the large volumes of water they filter while feeding (Viarengo and Canesi 1991; Cantillo 1998).

Nitrogen and carbon stable isotope ratios ($\delta^{15}N$ and $\delta^{13}C$) of bivalves have been used to assess the influence of anthropogenic nutrient loads on estuarine ecosystems (Fry 1999; Rogers 1999; McKinney et al. 2002). Anthropogenic nutrient sources such as treated sewage can have distinct $\delta^{15}N$ and $\delta^{13}C$ values that can be traced as they pass through the food chain. The isotope ratios of nutrient sources become distinct due to fractionation and result from slight mass differences between the heavy and light isotopes. Although functionally equivalent, the rates of chemical, physical, and biologically-mediated reactions are

*Corresponding author; current address: University of Washington, School of Oceanography, Box 355351, Seattle, Washington 98195-5351; tele: 206/543-0599; fax: 206/685-3354; e-mail: mooresk@u.washington.edu

faster for lighter isotopes than for heavier isotopes. Treated sewage has a distinctly enriched $\delta^{15}N$ due to higher rates of volatilization of the lighter ^{14}N as ammonia and preferential use of ^{14}N in the bacterially-mediated denitrification pathway (Heaton 1986; Jones et al. 2001).

Values of $\delta^{13}C$ can discriminate between marine and terrestrially derived organic matter due to the different carbon sources and pathways used in photosynthesis (Boutton 1991; Thornton and McManus 1994). Increased fractionation associated with the dissolution of atmospheric carbon dioxide into water results in marine plants being comparatively enriched in $\delta^{13}C$. As a result of this, nutrient sources such as sewage organic matter, which is terrestrially formed, can also be traced using depleted $\delta^{13}C$ (Rogers 1999).

Since the isotope ratio of an animal is closely related to that of its food (De Niro and Epstein 1978, 1981) and the isotope ratio of primary producers reflects their nutrient sources (McClelland et al. 1997; McClelland and Valiela 1998), information can be obtained on original nutrient sources and the contribution of anthropogenic nutrient sources to estuarine food chains by measuring the isotope ratios of bivalves. Bivalves are close to the base of the food chain so they are particularly useful bioindicators of nutrient enrichment using stable isotope techniques because their isotope ratios are less complicated by trophic enrichment.

This study uses the nitrogen and carbon stable isotope ratios of the pygmy mussel (*Xenostrobus securis*) to assess the influence of catchment disturbance for three estuaries in northern New South Wales, Australia. The pygmy mussel is an epifaunal mytilid found in high abundance in estuaries throughout southern Australia on firm substrates, particularly mangrove pneumatophores. The adult stage is found in the tidal reaches of estuarine systems and can tolerate a wide range of salinities, actively filtering when salinities are between 5 and 32.5 (Wilson 1969). *X. securis* also has the physiological ability to tolerate sudden osmotic shock, such as that which would occur during a large rain event.

Site Description

The Manning, Wallamba, and Wallingat Rivers are subtropical east Australian estuaries on the mid-north coast of New South Wales, Australia (Fig. 1). The region generally experiences a summer rainfall regime with the wettest month often in March and the driest month often in September. All three estuaries have a semidiurnal tidal cycle that extends to approximately 54 km and 20 km for the Manning and Wallingat Rivers, respectively. The Wallamba

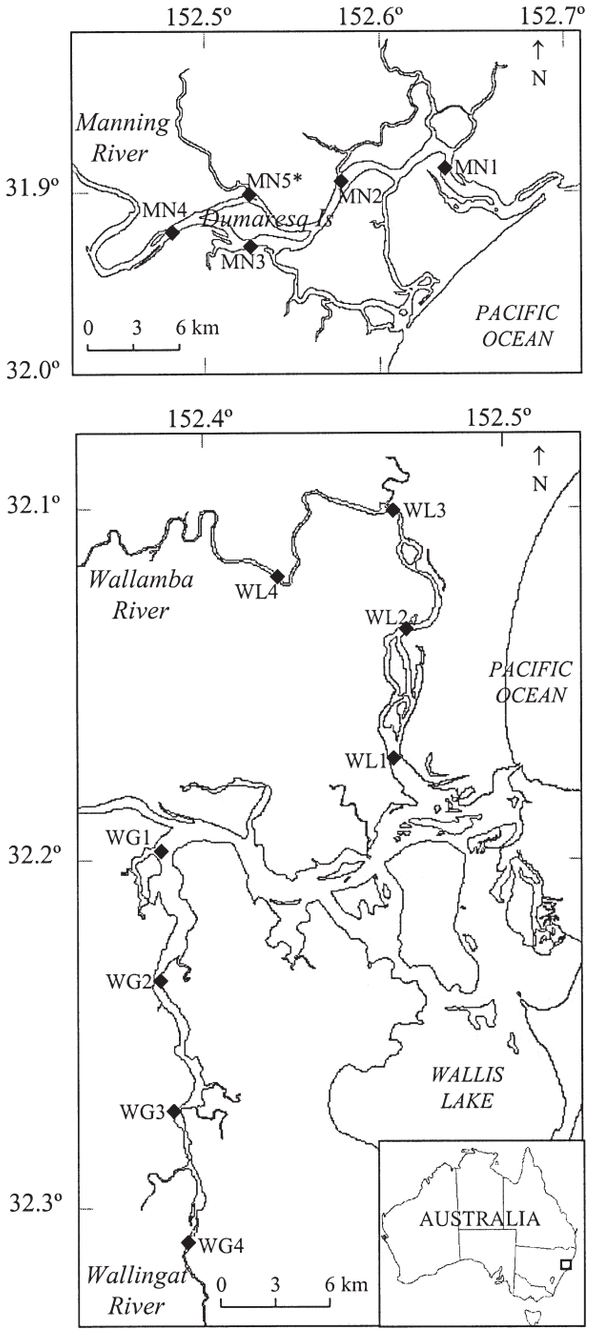

Fig. 1. Manning River sites (MN1-MN5*) and Wallis Lake region showing Wallamba (WL1-WL4) and Wallingat (WG1-WG4) River sites. Site MN5* indicates the location of the Dawson sewage treatment plant discharge point on the northern side of Dumaresq Island in the Manning River.

River is tidal to 28 km at which point the river is restricted by a weir.

The Manning River has the largest catchment area of the three estuaries covering 8,927 km^2. This

study took place in the lower Manning River catchment that is predominantly a floodplain with an elevation <50 m. The lower catchment has been extensively modified with 41% cleared for livestock agriculture (particularly beef and dairy) and 3% for residential purposes. The Dawson sewage treatment plant (STP) discharges approximately 5 Ml d^{-1} of tertiary treated effluent into the estuary 24 km upstream. The estuary has two entrances to the ocean: a main northern entrance with a secondary entrance approximately 12 km south.

The Wallamba and Wallingat Rivers flow into Wallis Lake, a permanently open coastal lagoon. The Wallamba River has a subcatchment covering approximately 500 km^2 or one third of the entire Wallis Lake catchment (1,440 km^2). Approximately 56% of the subcatchment has been extensively cleared and is used for livestock agriculture. The Wallingat River subcatchment covers approximately 185 km^2 and remains mostly vegetated (79%).

Materials and Methods

Field Study

The first stage of this study was conducted in the austral summer of 2001–2002. Mangrove pneumatophores with *X. securis* attached were collected from Site WL2 in the Wallamba River on November 8, 2001, and deployed at Sites WL1 and WL2 in the Wallamba River and at Sites WG1 and WG2 in the Wallingat River from November 2001 until March 2002 (Fig. 1). Sites were positioned at approximately 5 km intervals along each estuary, which is just outside the range of a typical tidal ellipse for the Wallamba River (Allsop and Kadluczka 1999). Mussels were deployed in cylindrical 1 cm mesh cages on one mooring at each site in 1.5–2 m depth of water. Moorings consisted of a 15 kg anchor with 1 m of chain attached to 2–2.5 m of rope with a surface buoy. Each cage contained approximately 50 mussels and was positioned on the mooring at approximately 0.5 m depth. Three mussels were sampled at fortnightly intervals from each mooring on November 25, 2001, December 6 and 18, 2001, January 4, 18, and 31, 2002, February 16 and 27, 2002, and March 13, 2002, and frozen whole until analysis. At each site on each sampling occasion temperature and salinity were measured at the surface using a calibrated Yeo-Kal 611 conductivity, temperature, and depth unit or a Hydrolab MiniSonde multiprobe interfaced with a Surveyor data logger.

The second stage of this study was carried out in the austral summer of 2002–2003 and was expanded to include the Manning River and the number of sites within each river was increased to four. Mangrove pneumatophores with *X. securis* attached were collected from Site WL2 in the Wallamba River on November 5, 2002, and deployed as before at Sites MN1-MN4 in the Manning River, Sites WL1-WL4 in the Wallamba River and Sites WG1-WG4 in the Wallingat River from December 2002 until April 2003. Three mussels were sampled from two replicate moorings approximately 15 m apart at each site (n = 2) at monthly intervals on December 3, 2002, January 6, 2003, February 6, 2003, March 8, 2003, and April 7, 2003, and frozen whole until analysis. On each sampling occasion three mussels were also sampled from two replicate moorings deployed within 50 m of the discharge point of the Dawson STP at Site MN5* (n = 2) in the Manning River (Fig. 1). The Dawson STP discharges to the north of Dumaresq Island that divides the Manning River into two channels with similar volumes and flow characteristics. Comparing the isotope ratios of *X. securis* from Site MN3 and MN5* allowed an assessment of any possible influence of the Dawson STP at a 1–5 km spatial scale within the estuary.

Different isotope ratios have been observed in conspecific suspension-feeding bivalves of different sizes as a result of ontogenetic shifts in diet (Kang et al. 1999) and differing rates of tissue turnover combined with highly variable diets of juveniles (Peterson et al. 1986). To minimize any size related effects on the isotope ratios of *X. securis* in this study, all mussels sampled were adults with a mean length on the longest axis and standard deviation of 21.9 ± 2.9 mm. Single factor analysis of covariance with Site as a categorical variable and Length as a covariate for each estuary confirmed that size did not significantly affect mussel $\delta^{15}N$ ($F_{1, 35} = 0.5$, $p = 0.472$; $F_{1, 35} < 0.1$, $p = 0.924$; and $F_{1, 35} = 0.5$, $p = 0.495$ for the Manning, Wallamba, and Wallingat Rivers, respectively) or $\delta^{13}C$ ($F_{1, 35} = 0.6$, $p = 0.431$; $F_{1, 35} = 0.1$, $p = 0.802$; and $F_{1, 35} = 1.3$, $p = 0.255$ for the Manning, Wallamba, and Wallingat Rivers, respectively) from the second stage of the study.

At each site on each sampling occasion during the second stage of the field study temperature and salinity were measured as before. Estuarine particulate organic matter (POM) samples (n = 2) were collected from each site from December 2002 to February 2003 to characterize the isotope ratios of the food sources available to the mussels. Two liters of water were sampled from the surface in darkened bottles and filtered on 47 mm diameter, 1.2 μm precombusted (450°C for 1.5 h) glass fiber filters under low vacuum within 7 h of collection and frozen until analysis.

POM samples are a complex mixture of organic sources from terrestrial and marine origin (Thornton and McManus 1994; Richard et al. 1997). To examine the possible contribution of anthropogenic derived organic matter to the estuarine POM

pool, sewage effluent and cow manure samples were collected and their isotope ratios were determined. Final tertiary treated sewage effluent (n = 2) was collected prior to discharge from the Dawson STP on three occasions during dry weather conditions (January 14 and 28, 2003, and March 14, 2003) and frozen until analysis. Fresh and dry cow manure samples (n = 3) were collected from the Wallamba River catchment at monthly intervals from December 2002 until April 2003 and frozen until analysis.

Any possible influence of gut fullness on mussel $\delta^{15}N$ was assessed during the first stage of the study using two-factor analysis of variance (ANOVA; River × Guts). Three additional mussels were collected from each site (n = 2) within each river on January 31, 2002, and placed in 5 l of 1.2 µm filtered water from the collection site for 12 h before being frozen. There was no significant effect on mussel $\delta^{15}N$ of immediately freezing mussels without first allowing their guts to evacuate ($F_{1,\ 5} = 0.5$, $p = 0.477$).

Sample Preparation and Analysis

Mussels were thawed, removed from the shell valves, and freeze dried using a Dynavac FD3 freeze drier. The small size of the mussel hindered the use of tissue specific analyses, which would have required amalgamating tissue samples from numerous individuals to obtain the minimum of 1 mg dry weight required for analyses. Whole mussels were homogenized to a fine powder using scissors and between 1–2 mg were weighed using a Cahn C33 digital microbalance to 1 µg accuracy and transferred into tin capsules for isotope analysis.

Isotope ratios are expressed in delta notation relative to international standards (atmospheric nitrogen for $\delta^{15}N$ and Pee Dee Belemnite for $\delta^{13}C$) and measured in parts per thousand (‰) according to the equation

$$\delta^{15}N \text{ or } \delta^{13}C = \left[\left(R_{sample} - R_{standard}\right)/R_{standard}\right] \times 1{,}000$$

where R is the ratio of the heavy to light isotope ($^{15}N : {}^{14}N$ or $^{13}C : {}^{12}C$). When comparing samples, those that contain more of the heavy isotope are referred to as enriched and those that contain less of the heavy isotope are depleted. Analysis was conducted using an Automated Nitrogen Carbon Analysis Mass Spectrometer (20-20 Europa Scientific) to a precision of 0.2‰.

Tissue condition was determined for mussels collected during the second stage of the study as the percentage of total dry weight represented by soft body tissue (Allen et al. 1999). Shell length was used to approximate shell weight for this calculation using a regression based on mussels collected from the three estuaries on two randomly chosen dates (December 14, 2002, and March 9, 2003; $r^2 = 0.89$, $p < 0.001$, n = 23). Tissue and shell were freeze dried using a Dynavac FD3 freeze drier and weighed using a Mettler Toledo PB303 DeltaRange balance to 0.01 g accuracy; shell length was measured using digital callipers.

Frozen filter papers with POM were freeze dried and between 2–3 mg of material was carefully removed from the surface of the filter paper using fine forceps and sealed in tin capsules for isotope analysis. Sewage effluent samples were thawed and 1 l was filtered on 47 mm diameter, 1.2 µm precombusted glass fiber filters under low vacuum. Sewage POM was analyzed as for estuarine POM. Cow manure samples were freeze dried, homogenized to a fine powder using scissors, and 2.0–2.5 mg of material were sealed in tin capsules for isotope analysis.

Catchment Land Use

Geographical Information System data describing land cover for the Manning, Wallamba, and Wallingat catchments were obtained from the New South Wales Department of Environment and Conservation Water Science Unit (unpublished data). The data were determined from 1999 satellite imagery and included 100 land cover classes with 25 m grid resolution. Subcatchment boundaries for the Wallamba and Wallingat Rivers were obtained from Great Lakes Council (unpublished data).

Three land use categories were calculated from the land cover classes following McKinney et al. (2002). Residential land use included the classes Urban Low Density, Urban Medium Density, Commercial-Industrial-Urban, Urban Residential, Urban Recreational and Caravan Parks. Agricultural land primarily used for livestock agriculture included the classes Unimproved Pasture and Improved Pasture, and areas of land with natural vegetation included the classes Conservation-Bushland, Bushland-Riparian-Wetland, and State Forests.

It was assumed that the isotope ratios of mussels would be most influenced by land-derived nutrients entering the estuaries 4 km upstream and 3 km downstream of each site, which is the distance of a typical tidal ellipse for the Wallamba River (Allsop and Kadluczka 1999). The lower catchments of the estuaries are floodplains with elevations of < 50 m above sea level, and so the flow paths of groundwater and runoff into the estuaries are difficult to determine. Streams and channels are more reliable conduits for nutrient input to the estuaries from the catchment. Drainage networks consisting of the estuary and the streams and channels entering the estuary within a tidal ellipse of each site were identified from topographic data obtained from

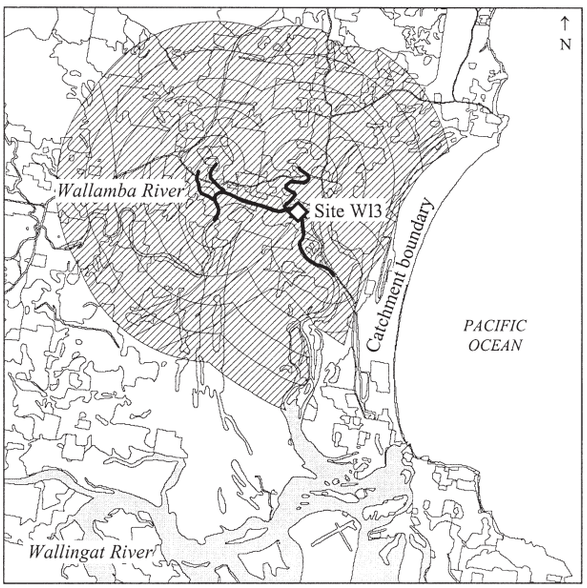

Fig. 2. Example of a drainage network for site WL3 (in bold) extending to 4 km upstream and 3 km downstream from the site location. Buffer zones to 1, 2, 3, 4, and 5 km from the drainage network within the Wallamba River catchment boundary are hashed. The fraction of land used for livestock agriculture, used for residential development, and covered in natural vegetation was calculated within these buffer zones. Different land use areas are outlined, and water bodies and drainage areas are shaded grey.

Geoscience Australia (unpublished data) using the Environmental Systems Research Institute, Inc. ArcInfo (ESRI ARC/INFO) software package. The fraction of land used for livestock agriculture, residential development, and natural vegetation within 1, 2, 3, 4, and 5 km buffers from the drainage networks within catchment boundaries (see Fig. 2) was linearly regressed against the isotope ratios of mussels at corresponding sites collected during the second stage of the study. The upper limit of the buffer area was set to 5 km from drainage networks as this was the distance between sites.

Results

Water temperature varied seasonally among sites and times during both stages of the study becoming cooler towards the end of the austral summer (Fig. 3). Salinity measurements typically showed an estuarine gradient with upstream sites fresher than downstream sites, and responded to two rain events in November 2001 and February 2002 with all sites becoming fresher. Besides these patterns there was no distinct clustering of an estuary during any one sampling occasion, indicating that the physicochemical characteristics of the estuaries were similar.

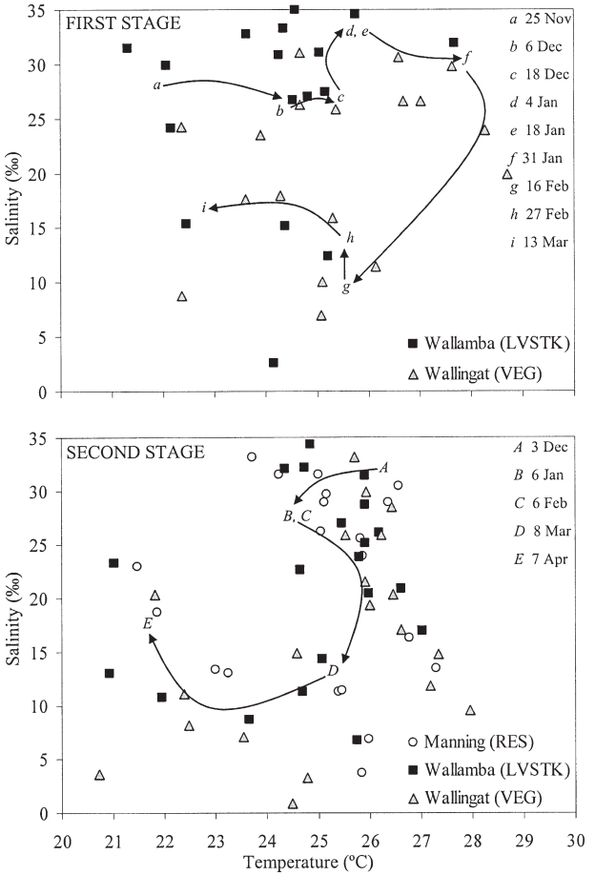

Fig. 3. Temperature and salinity measurements from the first stage of the study at sites in the Wallamba and Wallingat Rivers on nine sampling occasions from November 2001 until March 2002, and the second stage of the study at sites in the Manning, Wallamba, and Wallingat Rivers on five sampling occasions from December 2002 until April 2003.

The stable isotope ratios of *X. securis* from the estuaries were distinctly different (Fig. 4). Wallamba River mussels were consistently enriched in $\delta^{15}N$ compared to those from the Wallingat River by an average of 2.0‰ during the first stage and 1.5‰ during the second stage of the study. Mussels from the Manning River were the most enriched by an average of 1.7‰ compared to Wallamba River mussels and 3.2‰ compared to Wallingat River mussels during the second stage of the study. Mussel $\delta^{13}C$ values from the Wallamba and Wallingat Rivers differed by only 0.1–0.2‰ during the first and second stages of the study, but Manning River mussels were comparatively enriched by 1.5‰ compared to Wallamba and Wallingat River mussels during the second stage.

Although the trends of enrichment of *X. securis* from the Wallamba and Wallingat Rivers were consistent during the first and second stages of

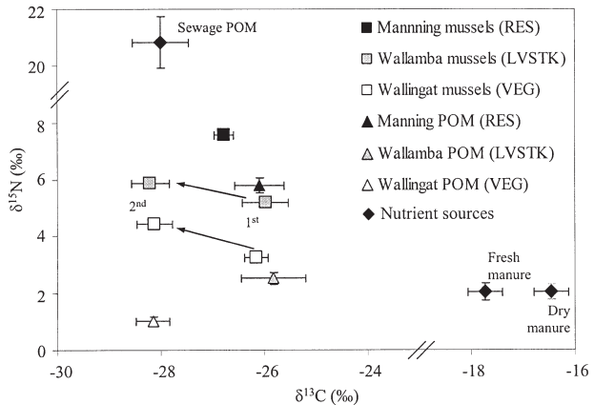

Fig. 4. Mean ± standard error $\delta^{15}N$ and $\delta^{13}C$ of *Xenostrobus securis* and POM from sites in the Manning, Wallamba, and Wallingat Rivers, and the nutrient sources sewage POM from the Dawson STP and fresh and dry cow manure. Arrows indicate the shift in isotope ratios of mussels from the first to the second stage of the study for the Wallamba and Wallingat Rivers.

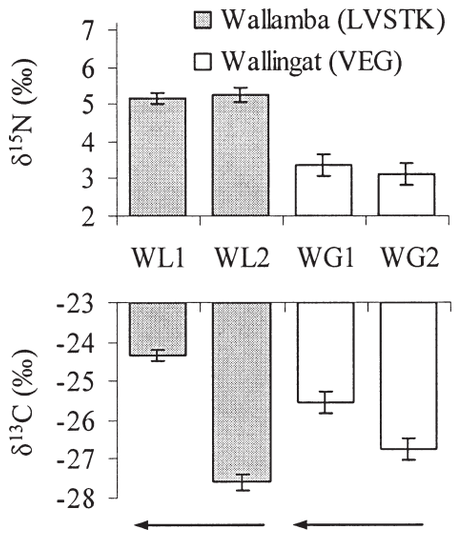

Fig. 5. Mean ± standard error $\delta^{15}N$ and $\delta^{13}C$ of *Xenostrobus securis* from sites WL1 and WL2 in the Wallamba River and sites WG1 and WG2 in the Wallingat River for the first stage of the study over nine sampling occasions. Arrows point downstream in each estuary.

the study, the actual isotope values differed (Fig. 4). Mussel $\delta^{15}N$ differed by only 0.7‰ and 1.2‰ for the Wallamba and Wallingat Rivers, respectively, during the first and second stages, but mussel $\delta^{13}C$ from the first stage of the study were depleted by 2.2‰ and 2.0‰ compared to the second stage.

POM $\delta^{15}N$ follows the same pattern as for mussel tissue during the second stage of the study with Manning River POM enriched by an average of 3.3‰ compared to Wallamba River POM and 4.8‰ compared to Wallingat River POM (Fig. 4). Values of $\delta^{13}C$ for Manning and Wallamba River POM were similar over the sampling period with values of −26.1‰ and −25.8‰, respectively. POM from the Wallingat River was comparatively depleted in $\delta^{13}C$ by approximately 2‰ with a mean value of −28.2‰.

The isotope ratios of the two nutrient sources measured during the second stage of this study were distinctly different (Fig. 4). Sewage POM from the Dawson STP that discharges into the Manning River was considerably enriched in $\delta^{15}N$ with a mean value of 20.8‰. The $\delta^{13}C$ value of sewage POM was similar to that of estuarine POM from the relatively unaltered Wallingat River, limiting its use as an indicator of human-derived effluent. Fresh and dry cow manure from the Wallamba River catchment was comparatively depleted in $\delta^{15}N$ with values of 2.0‰, but was considerably enriched in $\delta^{13}C$ with values of −16.5‰ and −17.7‰ for fresh and dry cow manure, respectively.

Patterns of enrichment in mussel isotope ratios within estuaries were consistent between the first and second stages of the study with $\delta^{15}N$ distinguishing between estuaries and $\delta^{13}C$ distinguishing between sites (Figs. 5 and 6). Repeated measures ANOVAs with River and Site as between subject factors and Time as a repeated factor were used to assess mussel isotope ratios during the second stage of the study. The effect of River on mussel $\delta^{15}N$ was highly significant with Manning River mussels the most enriched and those from the Wallingat River the most depleted (Fig. 6). The mean square value for this factor was considerably large explaining virtually all of the total variance (93%) from the analysis (Table 1). Significant differences in $\delta^{15}N$ were also found among times and all interaction effects involving time. This is because there is very little variation in the samples, evident by the low

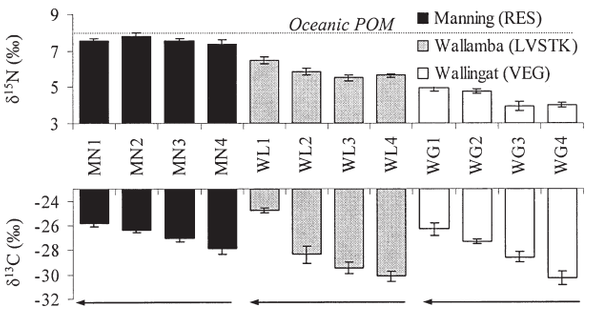

Fig. 6. Mean ± standard error $\delta^{15}N$ and $\delta^{13}C$ of *Xenostrobus securis* from sites MN1-MN4 in the Manning River, sites WL1-WL4 in the Wallamba River, and sites WG1-WG4 in the Wallingat River for the second stage of the study over five sampling occasions. The $\delta^{15}N$ value of oceanic POM from the central and south coasts of New South Wales is included for comparison (source: Gaston and Suthers 2004). Arrows point downstream in each estuary.

TABLE 1. Repeated measures ANOVAs of mussel $\delta^{15}N$, $\delta^{13}C$, and tissue condition to assess the effects of river and site over 5 sampling occasions (time) during the second stage of the study.

Stage 2: 2002–2003	$\delta^{15}N$				$\delta^{13}C$				Condition			
Source of variation	MS	df	F	p	MS	df	F	p	MS	df	F	p
Between subjects												
River	105.8	2	1769.3	< 0.001	29.1	2	331.0	< 0.001	74.39	2	56.96	< 0.001
Site	2.7	3	45.3	< 0.001	77.2	3	876.5	< 0.001	1.32	3	1.01	0.423
River × Site	1.3	6	21.2	< 0.001	8.8	6	99.6	< 0.001	5.63	6	4.31	0.015
Error	0.1	12			0.1	12			1.31	12		
Within subjects												
Time	2.0	4	33.3	< 0.001	16.7	4	141.0	< 0.001	2.00	4	1.20	0.321
Time × River	0.9	8	15.7	< 0.001	4.1	8	34.3	< 0.001	9.88	8	5.95	< 0.001
Time × Site	0.3	12	5.0	< 0.001	1.8	12	15.3	< 0.001	3.45	12	2.08	0.037
Time × River × Site	0.7	24	11.5	< 0.001	1.3	24	11.3	< 0.001	2.37	24	1.43	0.146
Error	0.1	48			0.1	48			1.66	48		

mean square error terms from the analyses, and producing large significant F-ratios for all factors and interactions (Table 1). Comparison of the mean square values is therefore more useful for this analysis, and reveals that the factor Time and all interaction effects involving Time explain only a small percentage of the total variance. The effect of Site was highly significant in the analysis of mussel $\delta^{13}C$ from the second stage of the study and explained 56% of the total variability based on mean square values (Table 1). An estuarine gradient in $\delta^{13}C$ up to 5.4‰ was apparent for each estuary with mussels from downstream sites enriched compared to those from upstream sites (Fig. 6). Due to the low variability in $\delta^{13}C$ among times, the analysis also indicated a significant effect of River, River × Site, Time, and all interactions involving Time (Table 1). These factors and interactions explain only a small percentage of the total variance.

Repeated measures ANOVAs were used to assess any influence on mussel isotope ratios of the Dawson STP at Site MN5* at the discharge point compared to Site MN3 in the Manning River. Significant Time × Site interactions were found for mussel $\delta^{15}N$ and $\delta^{13}C$ ($F_{4, 8} = 6.4$, $p = 0.013$; and $F_{4, 8} = 25.5$, $p < 0.001$, respectively), so any influence of effluent discharge was assessed using one-factor ANOVAs for each sampling occasion. Mussels were enriched in $\delta^{15}N$ at Site MN5* compared to those from Site MN3 on all sampling occasions (Fig. 7), but only significantly for December, February, and April ($F_{1, 2} = 24.9$, $p = 0.038$; $F_{1, 2} = 33.2$, $p = 0.029$; and $F_{1, 2} = 23.7$, $p = 0.040$, respectively). Mussel $\delta^{13}C$ was significantly enriched at Site MN5* compared to those from Site MN3 on April 7, 2003 ($F_{1, 2} = 277.5$, $p = 0.004$), but was not significantly different on all other sampling occasions.

Repeated measures ANOVA showed a highly significant effect of River on the tissue condition of *X. securis* with Manning River mussels having an average dry weight of 1.1% more than those from the Wallamba River and 2.8% more than those from the Wallingat River for all sites over the second stage of the study (Fig. 8). This effect had a large mean square value explaining 73% of the total variability in mussel condition (Table 1). There was also a significant River × Site interaction and significant Time × River and Time × Site interactions; a comparison of the mean square values indicated that these effects explained only a small percentage of the total variability in *X. securis* tissue condition (Table 1).

The fractions of land used for livestock agriculture, residential development, and natural vegetation within buffer zones for each site specific drainage network varied among estuaries (Table 2). The highest fractions of residential land use were associated with buffer zones in the Manning River of up to 20% for smaller zones of 1 km and 2 km. Buffer zones in the Wallamba River had the highest

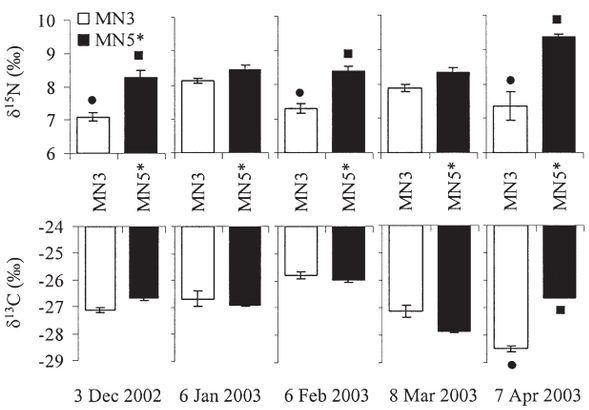

Fig. 7. Mean ± standard error $\delta^{15}N$ and $\delta^{13}C$ of *Xenostrobus securis* from site MN3 and site MN5* at the discharge point of the Dawson STP in the Manning River on December 3, 2002, January 6, 2003, February 6, 2003, March 8, 2003, and April 7, 2003. Columns with different symbols are significantly different.

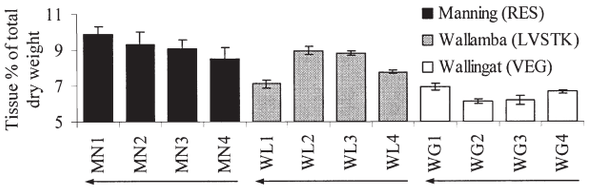

Fig. 8. Mean ± standard error tissue condition of *Xenostrobus securis* from sites MN1-MN4 in the Manning River, sites WL1-WL4 in the Wallamba River, and sites WG1-WG4 in the Wallingat River for the second stage of the study over five sampling occasions. Arrows point downstream in each estuary.

fractions of livestock agriculture land use of approximately 40% for the two most upstream sites (WL3 and WL4). Wallingat River buffer zones had the highest fractions of vegetated land of up to 95% with 0% residential development.

Fractions of land used for livestock agriculture and covered in natural vegetation were highly correlated using Pearson's Correlation ($r = -0.83, -0.91, -0.91, -0.94$, and -0.95 for 1, 2, 3, 4, and 5 km buffer zones, respectively, $n = 12$), and so the fraction of natural vegetation was removed from the multiple linear regressions with mussel $\delta^{15}N$. It was decided that the fractions of land used for livestock agriculture and residential development would remain in the regressions because they were the land uses potentially contributing to the increased $\delta^{15}N$ of mussels from estuaries with altered catchments.

Multiple linear regressions of the fractions of livestock agriculture and residential development with mussel $\delta^{15}N$ were significant for all buffer zone sizes ($p < 0.001$, $n = 120$; Table 3). The 5 km buffer zone regression explained the most variation in mussel $\delta^{15}N$ ($r^2 = 0.67$) and had the greatest standardized partial regression coefficients for both livestock agriculture and residential development ($\beta = 0.57$ and 0.96, respectively; Table 3). Calculated limits of agreement show that model estimated mussel $\delta^{15}N$ may be 1.6‰ above or 1.6‰ below observed values (Fig. 9). Limits of agreement were calculated as the mean difference of observed and estimated values ± 2 standard deviations of the differences and provided a statistical measure of assessing model fit where 95% of differences lie between the limits (Bland and Altman 1986). The model explaining the most variation in mussel $\delta^{15}N$ using the 5 km buffer zone is

$$\delta^{15}N_{pred} = -7.8\text{LVSTK} + 30.9\text{RES} + 5.6‰$$

where LVSTK and RES are the fractions of land used for livestock agriculture and residential development, respectively.

The predictive ability of the model was assessed using $\delta^{15}N$ of mussels collected during the first stage of the study. Estimated $\delta^{15}N$ was overestimated by an average of 1.0‰ compared to observed mussel $\delta^{15}N$ from the first stage of the study, with upper and lower limits of agreement of 3.3‰ and −1.3‰, respectively.

Discussion

The isotope ratios of *X. securis* varied between estuaries and sites during both the first and second stages of the study. Mussel $\delta^{15}N$ distinguished patterns in catchment disturbance between estuaries, with mussels from the Manning River the most enriched and those from the Wallingat River the most depleted. Mussel $\delta^{13}C$ was unable to distinguish patterns in catchment disturbance, but instead showed an estuarine gradient with downstream sites more enriched compared to upstream sites within each estuary. This was consistent with previously reported values from other systems and reflected the relative contribution of organic matter from terrestrial and marine sources to the diet of *X. securis* at each site (Boutton 1991; Thornton and McManus 1994).

The greater enrichment of mussel $\delta^{13}C$ in the Wallamba and Wallingat Rivers during the first stage

TABLE 2. Fractions of land used for livestock agriculture (LVSTK), residential development (RES), and natural vegetation (VEG) within 1, 2, 3, 4, and 5 km buffer zones from drainage networks for each site.

	Fraction LVSTK					Fraction RES					Fraction VEG				
Site	1 km	2 km	3 km	4 km	5 km	1 km	2 km	3 km	4 km	5 km	1 km	2 km	3 km	4 km	5 km
MN1	0.08	0.07	0.07	0.07	0.07	0.21	0.15	0.12	0.11	0.09	0.40	0.48	0.51	0.53	0.55
MN2	0.12	0.12	0.11	0.11	0.11	0.19	0.14	0.11	0.11	0.10	0.32	0.31	0.33	0.36	0.39
MN3	0.18	0.19	0.18	0.15	0.14	0.19	0.14	0.12	0.10	0.09	0.29	0.33	0.38	0.42	0.43
MN4	0.25	0.22	0.19	0.17	0.16	0.18	0.13	0.10	0.09	0.08	0.23	0.30	0.37	0.41	0.44
WL1	0.03	0.04	0.08	0.09	0.14	0.00	0.00	0.00	0.00	0.01	0.75	0.82	0.72	0.70	0.66
WL2	0.30	0.26	0.24	0.24	0.24	0.01	0.02	0.03	0.08	0.08	0.55	0.62	0.58	0.53	0.54
WL3	0.34	0.34	0.38	0.39	0.38	0.12	0.13	0.11	0.11	0.10	0.40	0.41	0.40	0.40	0.42
WL4	0.31	0.31	0.37	0.40	0.39	0.09	0.12	0.10	0.10	0.10	0.44	0.45	0.43	0.41	0.43
WG1	0.13	0.13	0.13	0.12	0.10	0.00	0.00	0.00	0.00	0.00	0.50	0.63	0.68	0.73	0.76
WG2	0.07	0.08	0.09	0.08	0.07	0.00	0.00	0.00	0.00	0.00	0.78	0.81	0.81	0.83	0.85
WG3	0.05	0.03	0.05	0.06	0.09	0.00	0.00	0.00	0.00	0.00	0.91	0.93	0.92	0.90	0.87
WG4	0.05	0.06	0.11	0.13	0.15	0.00	0.00	0.00	0.00	0.00	0.95	0.94	0.89	0.86	0.84

TABLE 3. Statistics from the multiple linear regressions of the fractions of land used for livestock agriculture (LVSTK) and residential development (RES) to predict mussel $\delta^{15}N$ in the Manning, Wallamba, and Wallingat Rivers during the second stage of the study for 1, 2, 3, 4, and 5 km buffer zone sizes.

	Single variable			Overall		
Land use	β	T	p	r^2	F	p
1 km LVSTK +	0.06	−1.00	0.321	0.61	91.26	<0.001
1 km RES	0.80	13.18	<0.001			
2 km LVSTK +	0.25	−3.43	0.001	0.54	69.54	<0.001
2 km RES	0.83	11.48	<0.001			
3 km LVSTK +	0.38	−5.73	<0.001	0.60	88.14	<0.001
3 km RES	0.88	13.27	<0.001			
4 km LVSTK +	0.52	−8.13	<0.001	0.65	107.74	<0.001
4 km RES	0.94	14.67	<0.001			
5 km LVSTK +	0.57	−9.24	<0.001	0.67	120.35	<0.001
5 km RES	0.96	15.46	<0.001			

of the study indicated that the diet of first stage mussels consisted of a greater proportion of marine-derived organic matter compared to those from the second stage. Less rainfall and runoff delivering less terrestrially-derived organic matter to the estuaries during the first stage of the study could produce this pattern in mussel $\delta^{13}C$, but average daily rainfall was 0.93 mm d^{-1} and 0.68 mm d^{-1} greater in the first stage compared to the second stage for the Wallamba and Wallingat River catchments, respectively. An alternative explanation is that wind-wave resuspension of benthic sediments was greater during the second stage increasing the amount of terrestrial-derived organic matter in the water column contributing to the diet of the mussels.

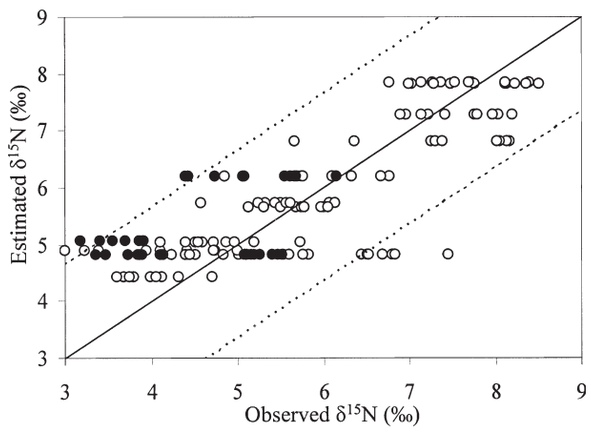

Fig. 9. Estimated versus observed $\delta^{15}N$ for mussels collected during the second stage of the study using the multiple linear regression model $\delta^{15}N_{pred} = -7.78LVSTK + 30.92RES + 5.60‰$. LVSTK and RES are the fractions of land used for livestock agriculture and residential development, respectively. A line of slope equal to one representing the model and lines representing the limits of agreement of the model are shown for comparison. Estimated $\delta^{15}N$ for mussels collected during the first stage of the study are shown for comparison in filled circles.

The enrichment of mussel $\delta^{15}N$ (1.9–3.4‰) and $\delta^{13}C$ (0–2.4‰) compared to estuarine POM for each estuary was within the typical range of trophic enrichment values (De Niro and Epstein 1978, 1981; Michener and Schell 1994). This indicated that mussels were directly feeding on POM characteristic of each estuary.

A direct link between the isotope ratios of POM and mussels has been demonstrated for other estuarine systems, with $\delta^{15}N$ reflecting increasing levels of residential development and nitrogen loading from human-derived effluent (Evgenidou and Valiela 2002). A similar pattern in $\delta^{15}N$ was observed in this study but mostly at the scale of catchments rather than being site specific within estuaries. Mussels from upstream sites were more influenced by land-derived organic matter as expected from decreased tidal flushing and longer residence times and evident from $\delta^{13}C$ values; this was not reflected in mussel $\delta^{15}N$ for all estuaries (Fig. 6). The $\delta^{15}N$ of mussels from the Manning River were fairly uniform at all sites over the 5 mo. This is probably due to the $\delta^{15}N$ value of 8.2‰ for oceanic POM from the central and south coasts of New South Wales (Gaston and Suthers 2004), which is more similar to estuarine POM at the most upstream site in the Manning River (6.0‰) compared to estuarine POM at the most upstream sites in the Wallamba (2.6‰) and Wallingat (1.4‰) Rivers. It appears that land-derived nitrogen entering the Manning River is enriching base level $\delta^{15}N$ values to levels entering the estuary via the ocean, masking any gradient in $\delta^{15}N$ with distance downstream. The land-derived nitrogen entering the Wallamba and Wallingat Rivers is likely depleted compared to oceanic inputs, resulting in a small estuarine gradient in $\delta^{15}N$ with mussels from downstream sites 0.8–0.9‰ more enriched than those from upstream sites.

Sewage effluent is a nutrient source distinctly enriched in $\delta^{15}N$ as a result of bacterially-mediated

denitrification that occurs during treatment (Jones et al. 2001). The considerably ^{15}N enriched tertiary treated sewage POM from the Dawson STP (20.8‰) consistently enriched mussels from Site MN5* at the point of sewage discharge over the 5 mo (Fig. 7). Mean mussel $\delta^{15}N$ at Site MN5* was only 8.6‰, enriched by just 1.0‰ compared to the reference site (MN3). This indicates that although mussels are using sewage-derived nitrogen on a localized spatial scale, it is largely diluted and nutrition is mostly obtained from other sources within the estuary.

Since temperature and salinity observations were similar among estuaries (Fig. 3), observed differences in tissue condition (Fig. 8) must be a result of other physicochemical or biological factors. Chlorophyll *a* concentrations observed during the second stage of the study using the method of Hallegraeffe and Jeffrey (1975) were 6.3 ± 0.6, 6.0 ± 0.6, and 4.6 ± 0.3 mg m^{-3} for the Manning, Wallamba, and Wallingat Rivers, respectively (mean ± standard error, n = 40), following the trend in mussel condition. This suggests that increased nutrient input to estuaries from altered catchments increases phytoplankton biomass, which increases food availability for filter feeding bivalves resulting in an increase in their physiological condition.

Declining physiological condition can cause tissue $\delta^{15}N$ values to become significantly enriched due to starving animals excreting the lighter ^{14}N at a faster rate than ^{15}N while neither pool is replaced by dietary protein (Hobson et al. 1993). The critical level of tissue condition below which this occurs in *X. securis* is unknown, yet starvation-enrichment is unlikely to have occurred in this study because the mussels with the lowest tissue condition from the Wallingat River were the most depleted in $\delta^{15}N$. This indicates that either the tissue condition of Wallingat River mussels did not fall below this critical level or that the starvation-enrichment that occurred in lower condition mussels was not sufficient to mask the patterns in $\delta^{15}N$ resulting from the use of different pools of nitrogen in the estuaries.

The samples sizes used in this study were small (n = 2) but each data value used in the analyses was the mean of three mussels from each mooring to reduce potential confounding. Previous research has indicated that with adequate replication at spatial scales, small samples sizes of n = 3 are sufficient to detect significant ecological variation (Gaston and Suthers 2004). Greater replication at spatial and temporal scales are recommended over increased sample sizes for future management studies.

The fractions of land used for livestock agriculture and residential development within 5 km buffer zones from drainage networks for each site explained 67% of the variation in $\delta^{15}N$ values of mussels collected during the second stage of the study. As fractions of livestock agriculture land use increased, estimated mussel $\delta^{15}N$ from the regression became depleted indicating the use of cow manure with relatively depleted $\delta^{15}N$ values (2.0‰) as a source of nutrition. This is contrary to previous research that presumed livestock-derived nitrogen would be enriched by the same bacterial transformation in groundwater that affects human-derived nitrogen from septic effluent, resulting in an elevated base level $\delta^{15}N$ with increasing fractions of land used for livestock agriculture (McKinney et al. 2002). In this study it appears that livestock-derived nitrogen undergoes little transformation or enrichment during transportation to the estuary in groundwater. The enrichment of Wallamba River mussels compared to those from the Wallingat River is likely a result of the small fractions of land used for residential development within Wallamba River buffer zones.

Although depleted in ^{15}N, fresh and dry cow manure were considerably enriched in $\delta^{13}C$ with values of −16.5‰ and −17.7‰, respectively. This nutrient source appears to be enriching estuarine POM $\delta^{13}C$ at sites with high fractions of livestock agriculture land use, with Manning and Wallamba River estuarine POM an average of 2.2‰ more enriched compared to the Wallingat River (Fig. 4). Mussel $\delta^{13}C$ values did not incorporate this pattern in POM $\delta^{13}C$ values and mussels from the Wallamba River had similar $\delta^{13}C$ values to those from the relatively unaltered Wallingat River. The similar $\delta^{13}C$ values of sewage POM from the Dawson STP and estuarine POM from the relatively unaltered Wallingat River compromises the use of ^{13}C as an indicator of anthropogenic catchment disturbance.

The enrichment of estimated mussel $\delta^{15}N$ with increasing fractions of land used for residential development has been previously demonstrated and is a result of nitrogen derived from septic systems undergoing transformation in groundwater enriching $\delta^{15}N$ values (McClelland and Valiela 1998; Lake et al. 2001; McKinney et al. 2002). In this study the highest fractions of residential development are associated with the 5 km buffer zones within the Manning River catchment, and these buffer zones also have high numbers of registered septic systems (652, 912, 1052, and 1,094 registered systems for sites MN1, MN2, MN3, and MN4, respectively; Greater Taree City Council unpublished data). It is possible that nitrogen derived from failing septic systems with a similarly enriched $\delta^{15}N$ value to sewage POM is entering the Manning River from numerous residential developments, enriching mussel $\delta^{15}N$ at all sites along the length of estuary examined during this study.

The predictive ability of the regression model for mussel $\delta^{15}N$ was reduced for the first stage of the study, and upper and lower limits of agreement (3.3‰ and −1.3‰, respectively) approached the total variation observed in $\delta^{15}N$ between estuaries. It is possible that the very small fraction of residential development in the Wallamba and Wallingat River catchments decreased the overall predictive ability of the model. A larger sample size of estuaries is required to further assess this relationship. The variability in mussel $\delta^{15}N$ values between the first and second stages of the study also likely reduced the effectiveness of the model. Sampling of more years would be required to determine the causes of this temporal variability so that it could be included in future models. In spite of these shortcomings, model estimated mussel $\delta^{15}N$ followed the observed trend of Wallamba River mussels being enriched compared to those from the Wallingat River.

The nitrogen isotope ratios of the mussel *X. securis* and estuarine POM are influenced by land-derived nutrient sources associated with anthropogenic land use characteristics within catchments. In spite of longer water residence times and decreased tidal exchange with the ocean, mussel and POM $\delta^{15}N$ were relatively uniform with increasing distance downstream at sites within estuaries, particularly in the Manning River. Mussel and POM $\delta^{15}N$ discriminated among estuaries in relation to catchment disturbance. Estuarine POM $\delta^{13}C$ values were influenced by fresh and dry cow manure at sites with high fractions of livestock agriculture land use in the surrounding catchments for the Manning and Wallamba Rivers. Mussel $\delta^{13}C$ values did not incorporate this pattern in POM $\delta^{13}C$ values and were unable to distinguish patterns in catchment disturbance between estuaries. Mussel $\delta^{13}C$ values showed an estuarine gradient between sites within estuaries. Fractions of land used for livestock agriculture and residential development explained 67% of the variation in mussel $\delta^{15}N$, so $\delta^{15}N$ values of *X. securis* are a useful integrator of the effects of catchment disturbance and anthropogenic nutrient enrichment in estuaries.

Acknowledgments

The authors acknowledge the Australian Research Council Strategic Partnership with Industry-Research and Training program (No. C00107004). Industry partners are Great Lakes Council, Department of Environment and Conservation, and the Department of Infrastructure, Planning and Natural Resources. We thank our partner investigators G. Tuckerman, T. Pritchard, and G. Carter. Mid-Coast Water collected effluent samples from the Dawson STP, Taree. Thanks to R. Piola, J. Dela-Cruz, T. Gaston, K. Wright, J. Everett, and R. and K. Moore for assistance and support at various times during field data collection and to M. Baird and M. de Graaf for providing useful comments on an earlier version of the manuscript. Thanks to E. Johnston and D. Marshall for advice on statistical analyses and particular thanks to S. Laffan and P. Hatton for assistance and advice with the processing of land use data.

Literature Cited

Allen, Y. C., B. A. Thompson, and C. W. Ramcharan. 1999. Growth and mortality rates of the zebra mussel, *Dreissena polymorpha*, in the Lower Mississippi River. *Canadian Journal of Fisheries and Aquatic Sciences* 56:748–759.

Allsop, D. and R. Kadluczka. 1999. Wallis Lake estuary tidal data collection March–June 1998. New South Wales Department of Public Works and Services and Manly Hydraulics Laboratory, MHL927, Manly Vale, New South Wales.

Bland, M. J. and D. G. Altman. 1986. Statistical methods for assessing agreement between two methods of clinical measurement. *Lancet* i:307–310.

Boutton, T. W. 1991. Stable Carbon Isotopic Ratios of Natural Materials: II. Atmospheric, Terrestrial, Marine and Freshwater Environments. Academic Press, San Diego, California.

Cantillo, A. Y. 1998. Comparison of results of Mussel Watch Programs of the United States and France with world-wide mussel watch studies. *Marine Pollution Bulletin* 36:712–717.

Carpenter, S. R., N. F. Caraco, D. L. Correll, R. W. Howarth, A. N. Sharpley, and V. H. Smith. 1998. Nonpoint pollution of surface waters with phosphorus and nitrogen. *Ecological Applications* 8:559–568.

Cloern, J. E. 2001. Our evolving conceptual model of the coastal eutrophication problem. *Marine Ecology Progress Series* 210:223–253.

de Jonge, V. N. and M. Elliott. 2001. Eutrophication, p. 852–870. *In* J. Steele, S. Thorpe, and K. Turekian (eds.), Encyclopaedia of Ocean Studies. Academic Press, London.

de Jonge, V. N., M. Elliott, and E. Orive. 2002. Causes, historical development, effects and future challenges of a common environmental problem: Eutrophication. *Hydrobiologia* 475/476:1–19.

De Niro, M. J. and S. Epstein. 1978. Influence of diet on the distribution of carbon isotopes in animals. *Geochimica et Cosmochimica Acta* 42:495–506.

De Niro, M. J. and S. Epstein. 1981. Influence of diet on the distribution of nitrogen isotopes in animals. *Geochimica et Cosmochimica Acta* 45:341–351.

Evgenidou, A. and I. Valiela. 2002. Response of growth and density of a population of *Geukensia demissa* to land-derived nitrogen loading, in Waquoit Bay, Massachusetts. *Estuarine Coastal and Shelf Science* 55:125–138.

Fry, B. 1999. Using stable isotopes to monitor watershed influences on aquatic trophodynamics. *Canadian Journal of Fisheries and Aquatic Sciences* 56:2167–2171.

Gaston, T. F. and I. M. Suthers. 2004. Spatial variation in $d^{13}C$ and $d^{15}N$ of liver, muscle and bone in a rocky reef planktivorous fish: The relative contribution of sewage. *Journal of Experimental Marine Biology and Ecology* 304:17–33.

Hallegraeff, G. M. and S. W. Jeffrey. 1975. New spectrophotometric equations for determining chlorophylls a, b, c1, and c2 in higher plants, algae and natural phytoplankton. *Biochemie und Physiologie der Pfanzen* 167:191–194.

Heaton, T. H. E. 1986. Isotopic studies of nitrogen pollution in the hydrosphere and atmosphere: A review. *Chemical Geology* 59:87–102.

Hobson, K. A., R. T. Alisauskas, and R. G. Clark. 1993. Stable-nitrogen isotope enrichment in avian tissues due to fasting and nutritional stress: Implications for isotopic analysis of diet. *Condor* 94:181–188.

Jones, A. B., M. J. O'Donohue, J. Udy, and W. C. Dennison. 2001. Assessing ecological impacts of shrimp and sewage effluent: Biological indicators with standard water quality analysis. *Estuarine Coastal and Shelf Science* 52:91–109.

Kang, C. K., P.-G. Sauriau, P. Richard, and G. F. Blanchard. 1999. Food sources of the suspension-feeding bivalve

Cerastoderma edule in a muddy sandflat of Marennes-Oléron Bay, as determined by analyses of carbon and nitrogen stable isotopes. *Marine Ecology Progress Series* 187:147–158.

LAKE, J. L., R. A. MCKINNEY, F. A. OSTERMAN, R. J. PRUELL, J. KIDDON, S. A. RYBA, AND A. D. LIBBY. 2001. Stable nitrogen isotopes as indicators of anthropogenic activities in small freshwater systems. *Canadian Journal of Fisheries and Aquatic Sciences* 58:870–878.

MCCLELLAND, J. W. AND I. VALIELA. 1998. Linking nitrogen in estuarine producers to land-derived sources. *Limnology and Oceanography* 43:577–585.

MCCLELLAND, J. W., I. VALIELA, AND R. H. MICHENER. 1997. Nitrogen-stable isotope signatures in estuarine food webs: A record of increasing urbanization in coastal watersheds. *Limnology and Oceanography* 42:930–937.

MCKINNEY, R. A., J. L. LAKE, M. A. CHARPENTIER, AND S. RYBA. 2002. Using mussel isotope ratios to assess anthropogenic nitrogen inputs to freshwater ecosystems. *Environmental Monitoring and Assessment* 74:167–192.

MEEUWIG, J. J. 1999. Predicting coastal eutrophication from land-use: An empirical approach to small non-stratified estuaries. *Marine Ecology Progress Series* 176:231–241.

MICHENER, R. H. AND D. M. SCHELL. 1994. Stable isotope ratios as tracers in marine aquatic food webs, p. 138–157. *In* K. Lajtha and R. H. Michener (eds.), Stable Isotopes in Ecology and Environmental Science. Blackwell Scientific Publications, Oxford, U.K.

NEDWELL, D. B., L. F. DONG, A. SAGE, AND G. J. C. UNDERWOOD. 2002. Variations of the nutrients loads to the mainland U.K. estuaries: Correlation with catchment areas, urbanization and coastal eutrophication. *Estuarine Coastal and Shelf Science* 54:951–970.

PATERSON, K. J., M. SCHREIDER, AND K. D. ZIMMERMAN. 2003. Effect of catchment disturbance on seston quality and quantity in Brisbane Water, NSW. *Estuarine Coastal and Shelf Science* 56:355–368.

PETERSON, B. J., R. W. HOWARTH, AND R. H. GARRITT. 1986. Sulfur and carbon isotopes as tracers of salt-marsh organic matter flow. *Ecology* 67:865–874.

RICHARD, P., P. RIERA, AND R. GALOIS. 1997. Temporal variations in the chemical and carbon isotope compositions of marine and terrestrial organic inputs in the Bay of Marennes-Oléron, France. *Journal of Coastal Research* 13:879–889.

ROGERS, K. M. 1999. Effects of sewage contamination on macro-algae and shellfish at Moa Point, New Zealand using stable carbon and nitrogen isotopes. *New Zealand Journal of Marine and Freshwater Research* 33:181–188.

SCHARLER, U. M. AND D. BAIRD. 2003. The influence of catchment management on salinity, nutrient stoichiometry and phyto-plankton biomass of Eastern Cape estuaries, South Africa. *Estuarine Coastal and Shelf Science* 56:735–748.

THORNTON, S. F. AND J. MCMANUS. 1994. Application of organic carbon and nitrogen stable isotope and C/N ratios as source indicators of organic matter provenance in estuarine systems: Evidence from the Tay estuary, Scotland. *Estuarine Coastal and Shelf Science* 38:219–233.

VIARENGO, A. AND L. CANESI. 1991. Mussels as biological indicators of pollution. *Aquaculture* 94:225–243.

WILSON, B. R. 1969. Survival and reproduction of the mussel *Xenostrobus securis* (Lamarck) (Mollusca: Bivalvia: Mytilidae) in a Western Australian estuary. Pt. II: Reproduction, growth and longevity. *Journal of Natural History* 3:93–120.

SOURCES OF UNPUBLISHED MATERIALS

GEOSCIENCE AUSTRALIA. unpublished data. Jerrabomberra Avenue (crn Hindmarsh Drive), Canberra, ACT 2600, Australia.

GREAT LAKES COUNCIL. unpublished data. Breese Parade, Forster, NSW 2428, Australia.

GREATER TAREE CITY COUNCIL. unpublished data. 2 Putney Street, Taree, NSW 2430, Australia.

NEW SOUTH WALES DEPARTMENT OF ENVIRONMENT AND CONSERVATION WATER SCIENCE UNIT. unpublished data. 59 Goulburn Street, Sydney, NSW 2000, Australia.

Received, October 10, 2004
Revised, June 1, 2005
Accepted, June 1, 2005