Vol. 501: 225–238, 2014
doi: 10.3354/meps10648

MARINE ECOLOGY PROGRESS SERIES
Mar Ecol Prog Ser

Published March 31

Assessing connectivity of a tropical estuarine teleost through otolith elemental profiles

Brad R. Moore[1,2,*], Colin A. Simpfendorfer[1]

[1]Centre for Sustainable Tropical Fisheries and Aquaculture, School of Earth and Environmental Sciences, James Cook University, Queensland 4811, Australia

[2]*Present address:* Coastal Fisheries Programme, Secretariat of the Pacific Community, BP D5, 98848 Noumea, New Caledonia

ABSTRACT: Understanding connectivity between groups of a species is fundamental to effective management and conservation practices, yet is poorly understood for tropical estuarine fishes. Here, age-related trends in otolith elemental chemistry were examined to assess the degree of connectivity of *Polydactylus macrochir*, a large, non-diadromous, tropical estuarine teleost, across the species' Australian distribution. Elemental signatures (^{7}Li, ^{43}Ca, ^{55}Mn, ^{88}Sr and ^{138}Ba) of transverse sections of otoliths of 3+ yr fish from the 2005 year class collected from 17 locations were sampled using laser ablation inductively coupled plasma mass spectrometry, providing elemental profiles from the otolith core through the first 3 yr of a fish's life. Univariate and cluster analyses revealed differences in elemental signatures of the otolith core among most locations, although similarities were evident among locations in the Gulf of Carpentaria and among 2 neighbouring locations in both Western Australia and the east coast of Queensland. Fewer differences were observed for post-settlement life history stages, although some differences were observed among neighbouring locations separated by as little as 50 km. SIMPROF analyses revealed that ^{138}Ba generally provided the greatest discrimination among locations. Positive correlations were observed between otolith ^{138}Ba concentration of individual fish and river flow indices for material laid down in all age groups, suggesting differences in flow was a significant driver of the observed patterns in ^{138}Ba. Examined in conjunction with complementary studies into the connectivity of the study species, the spatial structuring suggests that *P. macrochir* populations are susceptible to localised depletion, with limited opportunity for replenishment from neighbouring populations. The results highlight the importance of using multiple, complementary methods for assessing connectivity of aquatic organisms.

KEY WORDS: Otolith chemistry · LA-ICPMS · Connectivity · Tropics · Estuaries · Fishes

INTRODUCTION

Understanding the population structure and degree of exchange between groups of individuals from a single species (i.e. connectivity) is fundamental to the effective management and conservation of aquatic species and ecosystems (Cowen et al. 2007). Information on connectivity, which may be achieved through the dispersal of individuals as larvae, juveniles, or adults, helps to identify which groups are susceptible to localised depletion and potential sources of replenishment and recruitment to local populations (Thorrold et al. 2001, Gillanders 2009). Such knowledge is fundamental to the design of protected areas such as marine reserves or marine protected areas (MPAs), and for determining the spatial scale at which a species should be managed (Cowen et al. 2007). If managed inappropriately, spatially dis-

*Corresponding author: bradleym@spc.int

crete populations could be inadvertently subjected to localised depletion or extinction through over-fishing or environmental perturbation, as there would be limited opportunity for replenishment from neighbouring populations (Hilborn & Walters 1992).

Fishes that inhabit estuaries are regarded as particularly vulnerable to localised depletion from overfishing, given their typically patchy distributions and close proximity to human populations where fishing pressure is typically high (Blaber 2000, Secor & Rooker 2005). As estuarine species worldwide continue to face added pressures of anthropologically-induced habitat degradation and fragmentation, it becomes increasingly important to identify patterns of movement and exchange between spatially isolated groups. Although such information is considered fundamental to managing estuarine fishes, the degree of connectivity remains poorly understood for many species, particularly in tropical systems (Secor & Rooker 2005, Jones 2006, Gillanders 2009). Most studies of connectivity of tropical estuarine-associated fishes to date have focused on diadromous species, where the mode of diadromy (i.e. anadromy, catadromy) appears to be a significant driver of population connectivity (Russell & Garrett 1988, Milton & Chenery 2003). In contrast, there is a paucity of data on the connectivity of non-diadromous fishes inhabiting tropical estuaries. With anthropogenic pressures in the form of increased fishing pressure, habitat modification and climate change on tropical estuarine fishes projected to increase (Blaber 2000, Roessig et al. 2004), understanding their patterns of connectivity becomes increasingly important so that effective management can be implemented.

Analysis of elemental signatures of otoliths offers a powerful approach to examining patterns of connectivity of estuarine fishes (Thorrold et al. 2001, Gillanders 2002). As an otolith grows, elements are incorporated into its calcium carbonate structure at rates mediated by both environmental and endogenous factors, including ambient concentration, water temperature, salinity and diet (Fowler et al. 1995, Webb et al. 2012). As otoliths are metabolically inert, the deposition of elements and resulting chemical signature remains unaltered through time (Campana & Neilson 1985). Consequently, otoliths retain a chronological record of the environments experienced by a fish throughout its life (Secor & Rooker 2000). Correlating patterns of elemental signatures with temporal references within otoliths, such as the annual or daily growth increments, can facilitate examination of ontogenetic patterns of movement and connectivity (McCulloch et al. 2005).

The present study used otolith chemistry to examine connectivity of a non-diadromous estuarine-associated teleost: king threadfin, *Polydactylus macrochir* Günther, 1867, as part of a multidisciplinary study into the connectivity, movement and stock structure of this species across tropical and sub-tropical Australia. Specifically, the aim of this study was to examine elemental signatures from the otolith core (reflecting larval/early juvenile life history) through to material laid down during adult life, to assess connectivity across larval, late juvenile and adult life history stages. The following key questions were investigated: (1) Are otolith elemental signatures a useful technique of assessing connectivity of this species? (2) What patterns of connectivity are apparent from the elemental data, and how do they vary with ontogeny? (3) How do the estimates of connectivity derived from the elemental data compare against other techniques employed to assess connectivity of this species? (4) How does the spatial scale of connectivity of *P. macrochir* compare against those of the few studies examining connectivity of tropical, non-diadromous estuarine fishes?

MATERIALS AND METHODS

Study species. *Polydactylus macrochir* is a large, non-diadromous member of the Polynemidae that is endemic to tropical and subtropical estuaries and turbid coastal waters of northern Australia, southern Papua New Guinea and Irian Jaya (Motomura et al. 2000). In Australia, *P. macrochir* are distributed across the north of the continent from the Ashburton River in Western Australia to Brisbane in southeast Queensland (Motomura et al. 2000, Pember et al. 2005). The species is a protandrous hermaphrodite, has a life span of at least 22 yr and an estimated maximum attainable size of approximately 40 kg and 170 cm fork length (FL) (Kailola et al. 1993, Moore et al. 2011). They form an important component of estuarine and coastal ecosystems, and are a significant predator of crustaceans and small fishes (Brewer et al. 1995). The species is a significant component of northern Australia's commercial inshore net fisheries, second in importance only to the iconic barramundi *Lates calcarifer* (Welch et al. 2010). Spawning of *P. macrochir* on the east coast of Queensland occurs at the mouths and lower reaches of estuaries, with peak spawning occurring between October and December (Moore et al. 2011). Both eggs and larvae of *P. macrochir* are pelagic (Motomura 2004). Young-of-the-year juveniles (30 to

100 mm FL) have been observed in north Queensland estuaries in salinities ranging from 2.0 to 37.8 (I. Halliday pers. comm.), suggesting a high due of euryhalinity of these life history stages. *P. macrochir* does not use freshwater during any life history stage, and likely limits its use of estuarine habitats to permanent water areas of the main channels and tributaries (Halliday et al. 2008).

Sample collection. *Polydactylus macrochir* were collected from 17 locations (lower estuarine stretches of rivers and coastal sites) across the species' northern Australian distribution between July 2007 and February 2010 (Fig.1). At each location (with the exception of the Brisbane River and Lucinda), fish were obtained directly from commercial fishers or fish processors, or by fisheries-independent sampling that used the same gear used by commercial fishers (i.e. a combination of gillnets of 100 mm to 165 mm stretched mesh). Brisbane River samples were collected through a fishery-independent sampling program using the same gear as used by commercial fishers as outlined above, by opportunistic collections from recreational fishers, and by research line-fishing, whereas Lucinda samples were obtained from recreational fishers. The location, date of capture, sex, maturity stage, FL and total length (TL) were recorded for each fish collected, unless damaged. Sagittal otoliths (hereafter referred to as otoliths) were removed from all fish. Otoliths were rinsed in deionised (Milli-Q) water, cleaned of adhering tissue and stored to dry for later age estimation and elemental analysis. A single otolith from each fish was selected for ageing, which followed the protocol of Moore et al. (2011, 2012a), and assigned to a year class on the basis of spawning year. To minimise any confounding temporal influences on the spatial comparisons due to differences in collection time, examination of elemental profiles was conducted on 3+ yr fish that originated from the 2005 year class. Between 6 and 18 otoliths were randomly selected from those available from each of the 17 locations for the analyses (Fig. 1). Otoliths used for elemental analysis were rinsed with ultra-pure water and air-dried overnight for processing.

Otolith preparation. Each otolith was embedded in epoxy resin and a single transverse section of 400 µm thickness encompassing the core was taken using a low speed diamond-edged circular saw continuously lubricated with ultra-pure water. Each otolith section was polished down to the inner core on both sides using 1500 grit-size abrasive paper that was wet with Milli-Q water. Polished sections were triple rinsed with Milli-Q water and air-dried. Four randomly selected sections, each from an individual fish, were fixed on an acid-washed 50 × 25 mm microscope slide with resin. Each slide was wiped with a paper towel wet with 0.5 M HNO_3 to remove any surface contamination, rinsed with Milli-Q water and air-dried before being stored in an individual plastic bag for transportation to the laser ablation facility.

Elemental analysis. The otolith sections were analysed by laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS). The system con-

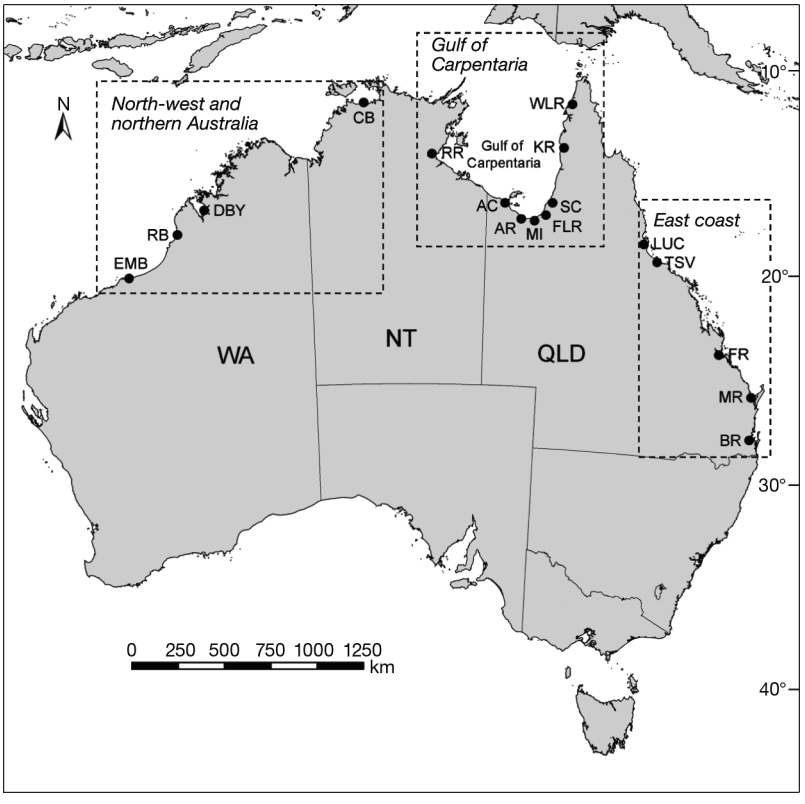

Fig. 1. *Polydactylus macrochir*. Sampling locations for examination of otolith elemental signatures (sample sizes in parentheses). From left: EMB, Eighty Mile Beach (15); RB, Roebuck Bay (16); DBY, Doctors Creek, Derby (10); CB, Chambers Bay (11); RR, Roper River (14); AC, Arthurs Creek (14); AR, Albert River (15); MI, Morning Inlet (14); FLR, Flinders River (18); SC, Spring Creek (14); KR, Kendall River (10); WLR, Wenlock River (8); LUC, Lucinda (6); TSV, Cleveland Bay, Townsville (10); FR, Fitzroy River (12); MR, Mary River (10); BR, Brisbane River (10). Dashed boxes: regions separated in the multivariate analyses

sisted of a New Wave UP-213 high performance ultraviolet laser ablation system connected to an Agilent 7500ce ICP-MS. Each slide was placed in a sealed perspex ablation chamber with helium atmosphere (1.10 l min^{-1}) and viewed remotely via a computer monitor. The laser was programmed to follow a transect from the core to the outer edge of the otolith section parallel to the edge of the sulcus. This sampling axis was chosen as it generally provided well defined opaque annuli when viewed under reflected light. Examination of the elemental profiles revealed a distinct peak of ^{55}Mn in the otolith core of most samples (see Results), consistent with studies on other teleosts (Ruttenberg et al. 2005). Accordingly, the presence of these ^{55}Mn peaks was used to confirm that each transect sampled the otolith core and to allow identification of the position of the core along each transect. Transects were ablated using an 80 µm diameter laser beam, at a pulse rate of 5 Hz, a laser energy of 80% and a scan speed of 10 µm s^{-1}. A pilot study revealed concentrations of ^{7}Li, ^{43}Ca, ^{55}Mn, ^{86}Sr, ^{88}Sr, ^{137}Ba and ^{138}Ba were generally measured above detection limits and showed no evidence of surface contamination, based on comparisons of pre-ablated and non pre-ablated otoliths. These elements were therefore recorded for all transects. As previous studies have shown Sr and Ba to be particularly useful for discriminating groups of estuarine fishes (Gillanders 2002, Milton & Chenery 2003); 2 isotopes of Sr and Ba were read in order to cross-check the occurrence of any possible mass interference of these elements.

Prior to ablating each otolith section, background counts of each elemental isotope were measured in the blank sample gas for 20 s. This provided average background counts of the analysed isotopes, which were subtracted from the sample counts for each ablation. After processing each otolith section, the chamber was purged with Argon gas for approx. 2 to 3 min to eliminate any background gases that may have contained contaminants. Otolith sections were analysed in random order to eliminate any possible biases associated with instrument drift.

Counts of elemental isotopes were calibrated against the National Institute of Standards (NIST) 612 glass standard. This standard was analysed once at the beginning of each day and once at the beginning and completion of each slide (4 ablations) to further eliminate short-term instrument drift by linear interpolation. The minimum detection limit at the 99% confidence level was calculated using GLITTER software (Van Achterbergh et al. 2001). Average detection limits (in ppm) for each isotope were estimated as: ^{7}Li 0.01, ^{43}Ca 20.62, ^{55}Mn 0.05, ^{86}Sr 0.76, ^{88}Sr 0.05, ^{137}Ba 0.02, ^{138}Ba 0.01. Estimates of precision (% relative standard deviation, RSD, expressed in molar ratios relative to mean ^{43}Ca concentration) based on the repeated analysis of the NIST 612 standard for each isotope were: ^{7}Li 7.80%, ^{55}Mn 3.18%, ^{86}Sr 4.94%, ^{88}Sr 2.81%, ^{137}Ba 3.26%, ^{138}Ba 3.01%.

Calcium (^{43}Ca) was used as an internal standard to correct for variations in ablation yield. Calcium concentration was assumed to be constant at 388 000 µg g^{-1} based on published values for certified otolith reference material (Yoshinaga et al. 2000). To control for the amount of ablated material, all elemental data were expressed as molar ratios to ^{43}Ca (hereafter referred to as 'concentrations'). Elemental values in the NIST 612 standard were derived from Pearce et al. (1997).

Relating elemental signatures to fish age. The elemental signatures were related to fish age using the peak in ^{55}Mn and the annuli of the otolith as temporal references. After each otolith was ablated using LA-ICP-MS, a digital image of the section was recorded using a Leica DC 300 digital camera mounted to a dissecting microscope. Using this image, a transect was drawn adjacent to the ablation scar, and the width of each annual increment was measured along this transect from the outer edges of the consecutive opaque zones. The elemental profiles were then divided into 4 life-history stages according to the measured distances along this transect: larval/early juvenile (first 50 µm of the transect, comprising the otolith core), and Years 1, 2 and 3. Only the chemical signatures from the core to the third opaque zone were used in the analyses. Although fish were collected from the different locations over a 12 mo period, no analyses were conducted on the elemental signatures within the marginal increment.

Data analysis. To reduce the noise in the data, concentrations of each element were lightly smoothed using a 7-point running median. The profiles for each element were plotted against fish age, to visualise age-related variation of elemental signatures within and among locations. For each otolith an age group mean was calculated from the elemental readings that were assigned to each age group. This provided 4 age group means for each otolith: one for the core (larval/early juvenile portion), and one for each of the 3 following years of the fish's life.

Concentrations of ^{7}Li and ^{55}Mn were found to be highest in the otolith core and generally decreased throughout the first 3 years of a fish's life (Fig. 2; Fig. S1 in the supplement at www.int-res.com/articles/suppl/m501p225_supp.pdf). As average ^{55}Mn

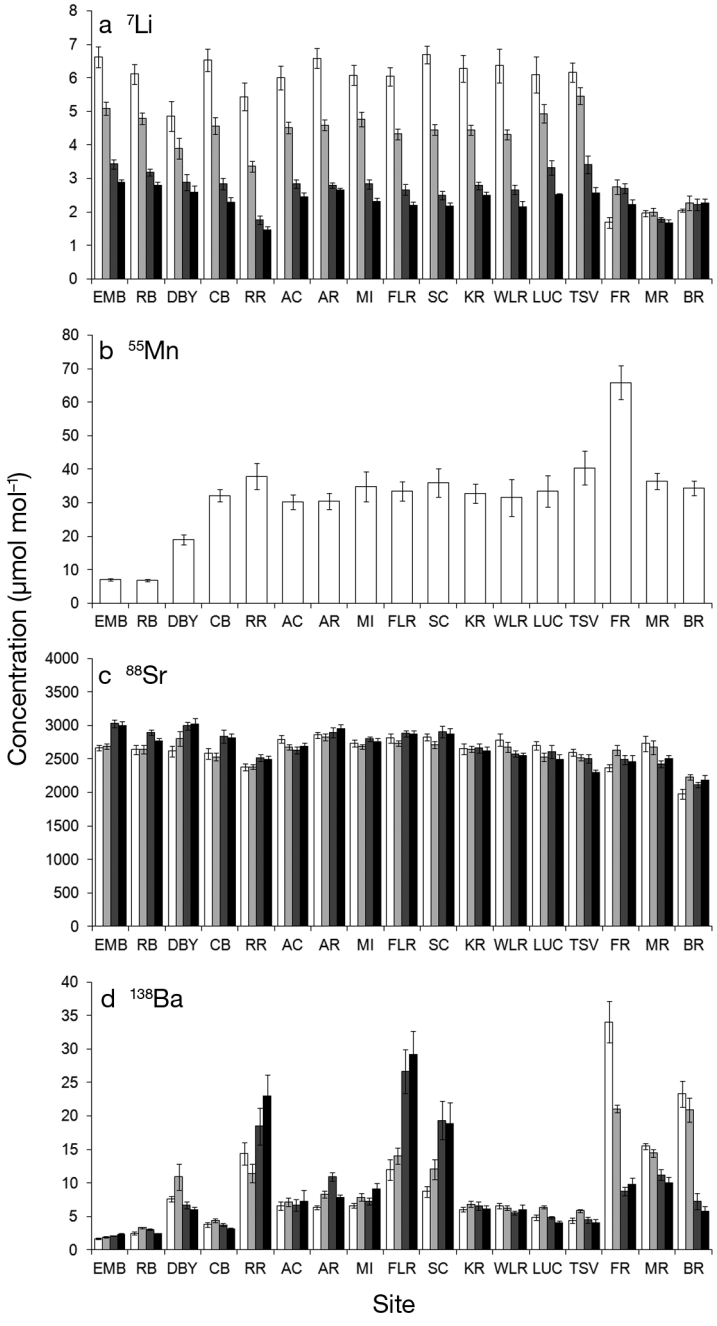

Fig. 2. *Polydactylus macrochir*. Age-related mean concentrations (µmol mol^{-1} relative to ^{43}Ca) of ^{7}Li, ^{55}Mn, ^{88}Sr and ^{138}Ba in otoliths collected from 17 locations across northern Australia (±1 SE). Larval/early juvenile = white, Year 1 = light grey, Year 2 = dark grey, Year 3 = black. See Fig. 1 for location codes

concentrations generally dropped below detection levels with increasing distance from the core (Fig. S1), data for this element were included only in the analyses of otolith cores (larval/early juvenile portion). Whilst relative concentrations of ^{7}Li showed a similar decrease at most locations, ^{7}Li concentrations exceeded the detection limits throughout a fish's life, and as such was included for analyses of all age groups. The 2 isotopes recorded for Sr (^{86}Sr and ^{88}Sr) and Ba (^{137}Ba and ^{138}Ba) showed similar trends across the otoliths, suggesting that variation in their profiles represent real trends in chemical composition. Of the 2 isotopes, ^{88}Sr and ^{138}Ba showed the lowest variance, and were therefore considered more reliable for analysis.

Patterns of individual elements. One-way ANOVA was applied to identify differences in the individual elements among the 17 locations for each age group (larval/early juvenile, Year 1, 2 and 3), using location as a fixed factor in the univariate design. As Shapiro-Wilk tests revealed the data for some elements were non-normal ($p < 0.05$), concentrations were ln(x+1) transformed prior to analysis to minimise the variance of the data. Significant results were examined using Tukey-Kramer post-hoc pairwise comparisons.

Patterns in multi-element signatures. As the ablated material was laid down over the course of a fish's life, individual fish may not have necessarily originated at the respective collection locations. As such, no assumption of location was considered in the multivariate analyses for any age group. Rather, hierarchical cluster analysis was used to group fish in each age group that contained similar elemental/Ca ratios. Cluster analysis was performed using normalized Euclidean distance and average linkage. The similarity profile permutation test (SIMPROF) was used to identify significantly different clusters in each age group at $p < 0.05$. As *Polydactylus macrochir* from the coasts of north-west and northern Australia, the Gulf of Carpentaria, and eastern Queensland are genetically distinct (Horne et al. 2012), these regions were separated in the cluster and SIMPROF analyses. All multivariate analyses were conducted on ln(x+1) transformed data using PRIMER (PRIMER-E).

Relating elemental profiles to biological and environmental variables. To gain an understanding of the factors driving the observed ^{138}Ba concentrations, and to further explore structuring of *Polydactylus macrochir*, annual otolith concentrations of individual fish were examined for correlations against biological and environmental variables via regression analysis. Ba was selected as it was

responsible for the majority of structuring among groups (see Results). Variables examined included otolith incremental width of individual fish (µm, as a proxy for growth), 3 rainfall indices: mean monthly rainfall, peak monthly rainfall and total annual rainfall, and 3 environmental flow indices: mean monthly river flow, peak monthly river flow and total annual river flow. Historical rainfall data were obtained from BoM (2013), with the exception of the Flinders River, which were obtained from the Queensland Department of Natural Resources and Mines (DNRM 2012). Rainfall data were taken from the closest gauging station to the collection location where data for all months were available. Environmental flow data for sampled estuaries were obtained from DNRM as above, using data from the most downstream gauging station. Where a river contained major water storage infrastructure (i.e. a dam or barrage), data were used only when the gauging station was situated downstream of the structure. While no flow gauging station occurs at Eighty Mile Beach or Roebuck Bay, these locations lie along an arid coastline with no proximity to any freshwater river systems (Newman et al. 2010), therefore flow rates at these locations were considered as zero. All data were calculated relative to a *P. macrochir* 'year' (i.e. November to October), and log(x+1) transformed prior to analysis. While incremental width data were available for all collection locations, and rainfall data for most locations, reliable environmental flow data were available for 6 locations only (a summary of data used in these analyses is given in Table S1).

RESULTS

Patterns of individual elements

There was significant variation among estuaries in the age-related mean concentration of each element in each of the 4 age groups (Table 1; Tables S2 & S3). In some cases, however, differences were inconsistent among age groups, with an element differing among locations for one age group but not the next. In general, average ^{7}Li concentrations were significantly lower for samples collected from the Fitzroy, Mary and Brisbane Rivers relative to all other locations for the core region (Fig. 2; Tables S2 & S3). Average ^{55}Mn concentrations were lowest in the core region of samples collected from Eighty Mile Beach and Roebuck Bay relative to all other locations, and highest in fish collected from the Fitzroy River. Average ^{88}Sr concentrations varied considerably over the life of an individual fish, showing numerous peaks and troughs within samples from all locations (Fig. S1). A slight decrease in average ^{88}Sr from the westernmost locations to the easternmost locations was evident, and typically highest in fish from the north-west and northern Australia and lowest in Brisbane River fish (Fig. 2). Fish collected from the Brisbane River had significantly lower average ^{88}Sr concentrations than those collected from the neighbouring Mary River location for all age groups (Tables S2 & S3).

Table 1. *Polydactylus macrochir*. Analysis of variance (*F*-values) of elemental concentrations (µmol mol^{-1} relative to ^{43}Ca) among locations for each otolith region. For all tests: df = 16,190; p < 0.01

Otolith region	^{7}Li	^{55}Mn	^{88}Sr	^{138}Ba
Core	44.94	41.75	12.00	80.84
Year 1	26.88	–	7.00	57.29
Year 2	11.49	–	16.00	41.50
Year 3	10.32	–	15.00	41.59

Of the 4 elements analysed, ^{138}Ba showed the most variation among locations (Fig. 2). In the north-west and northern region, average ^{138}Ba concentrations of fish collected from Derby were significantly higher than those from Eighty Mile Beach and Roebuck Bay for all age groups, and Chambers Bay for core material and Year 1 (Fig. 2; Tables S2 & S3). In the Gulf of Carpentaria, average ^{138}Ba signatures of fish from the Flinders River were significantly higher than those caught from most other locations for all age groups examined (Fig. 2; Tables S2 & S3). Average ^{138}Ba concentrations of otoliths collected from the Flinders River and Spring Creek typically increased with age, from immediately before the deposition of the first annual band (Fig. 2; Fig. S1). On the east coast of Queensland, fish caught from Lucinda and Townsville had the lowest average ^{138}Ba concentrations for all ages examined. Fish from the Fitzroy, Mary and Brisbane Rivers had relatively high average ^{138}Ba concentrations in their otolith cores that decreased with age (Fig. 2).

Patterns in multi-element signatures

North-west and northern Australia

Cluster analyses of fish from north-west and northern Australia revealed significant structuring within most age groups. For otolith core material, 3 distinct groups were apparent: one group containing all sam-

ples collected from Eighty Mile Beach and Roebuck Bay (Group A), and separate groups for all fish collected from Chambers Bay (Group B) and Derby (Group C) (Table 2). SIMPROF tests revealed that Group A was characterised by relatively low ^{55}Mn and ^{138}Ba, Group B by relatively high ^{55}Mn and relatively moderate ^{138}Ba, and Group C by relatively moderate ^{55}Mn and relatively high ^{138}Ba (Fig. 3). Similarly, 3 distinct clusters were observed for material laid down in Year 1: one group (Group A) containing all samples from Eighty Mile Beach and Roebuck Bay, and 8 fish from Chambers Bay (Group A), one group containing predominantly Derby fish and the 3 remaining Chambers Bay fish (Group B), and a third group characterised solely by 3 fish collected from Derby (Group C) (Fig. 3, Table 2). For material laid down in Year 2, 2 distinct clusters were evident: one cluster characterised by fish with relatively low ^{138}Ba that contained all fish collected from Eighty Mile Beach and Roebuck Bay and the majority of fish from Chambers Bay, and a second group characterised by relatively high ^{138}Ba that contained all fish collected from Derby and the remaining Chambers Bay fish (2 individuals) (Fig. 3, Table 2). No significant structuring was evident for material laid down in Year 3, with all samples grouping into a single cluster (Fig. 3, Table 2).

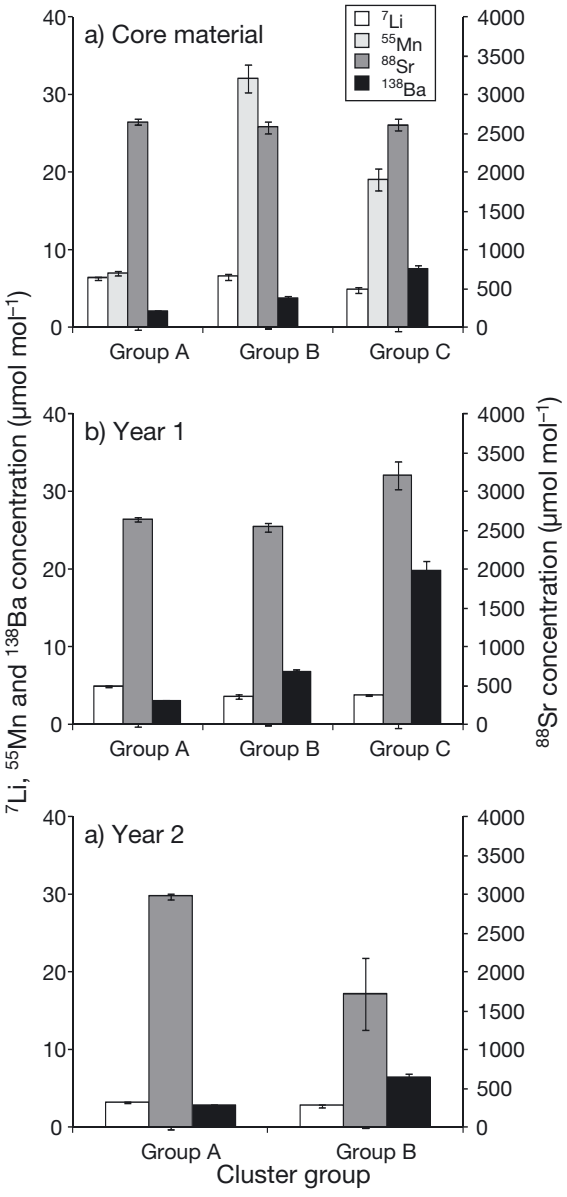

Fig. 3. *Polydactylus macrochir*. Mean elemental concentrations (µmol mol^{-1} relative to ^{43}Ca) for the groups identified by SIMPROF analysis in north-west and northern Australia (±1 SE). No significantly different groups were observed from material laid down in Year 3

Table 2. *Polydactylus macrochir*. Percentage of individuals assigned to each group identified by cluster analysis from locations in north-west and northern Australia. See Fig. 1 for location codes

Otolith region	Group	EMB	RB	DBY	CB
Core	A	100	100	0	0
	B	0	0	0	100
	C	0	0	100	0
Year 1	A	100	100	0	73
	B	0	0	70	27
	C	0	0	30	0
Year 2	A	100	100	0	82
	B	0	0	100	18
Year 3	A	100	100	100	100

Gulf of Carpentaria

Cluster and SIMPROF analyses of the otolith core material of fish from the Gulf of Carpentaria revealed 3 distinct groups: one group characterised by relatively low ^{138}Ba that contained individuals from all locations, a second group characterised by moderate ^{138}Ba concentrations that contained individuals collected from all locations except for the Albert and Kendall Rivers, and a third group characterised by relatively high ^{138}Ba that contained individuals collected from the Roper and Flinders Rivers and one individual from Spring Creek (Fig. 4, Table 3). Two distinct clusters were evident for material laid down in Year 1, one characterised by relatively low ^{138}Ba that contained individuals from all locations, and one characterised by relatively high ^{138}Ba that contained individuals from all locations except the Kendall and

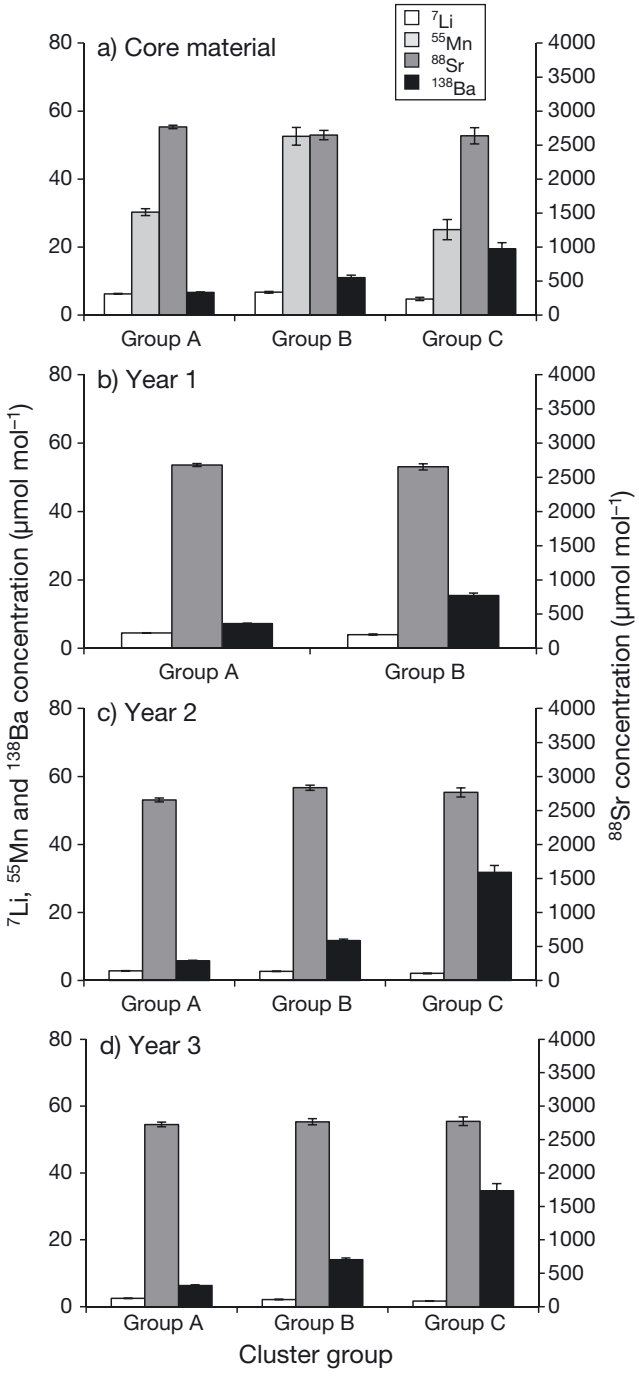

Fig. 4. *Polydactylus macrochir*. Mean elemental concentrations (µmol mol^{-1} relative to ^{43}Ca) for the groups identified by SIMPROF analysis in the Gulf of Carpentaria (±1 SE)

Wenlock Rivers in the north-east of the Gulf. For Year 2 material, 3 distinct clusters were evident: one with relatively low ^{138}Ba containing samples from all locations except the Albert and Flinders Rivers, a second group with moderate ^{138}Ba comprising samples from all locations except the Wenlock River, and a third group characterised by relatively high ^{138}Ba containing samples from the Roper and Flinders Rivers and Spring Creek (Fig. 4, Table 3). Similarly 3 clusters were evident for Year 3 material; 2 groups (Group A & B) containing samples from most locations, and a third (Group C) with high ^{138}Ba characterised predominantly by samples from the Roper and Flinders Rivers (Table 3). Of the fish collected from the Flinders River, 10 individuals consistently displayed a high ^{138}Ba signature across Years 1, 2, and 3.

East coast

Cluster and SIMPROF analyses of otolith core elemental signatures of fish from the east coast of Queensland revealed significant structuring, with 5 distinct groups apparent: one group that contained all fish collected from Lucinda and Townsville, characterised by relatively high ^{7}Li and relatively low ^{138}Ba, a second group containing a single anomalous individual collected from the Brisbane River, a third characterised by relatively low ^{7}Li, low ^{88}Sr and moderate ^{138}Ba, containing 90% of fish collected from the Brisbane River and a single fish from the Mary River, a fourth containing 90% of fish collected from the Mary River and a fifth group characterised by relatively low ^{7}Li, high ^{55}Mn and high ^{138}Ba, containing all fish from the Fitzroy River (Fig. 5, Table 4). For material laid down in Year 1, 3 distinct clusters were observed: one group characterised by relatively high ^{7}Li and low ^{138}Ba that solely contained fish collected from Lucinda and Townsville, one group containing a single fish collected from the Brisbane River, and a third group containing 90% of fish collected from the Brisbane River and all fish collected from the Fitzroy and Mary Rivers. Little structuring was evident from material in Years 2 and 3, with 2 different clusters identified in these age groups; one with relatively low ^{138}Ba (Group A), that contained a mix of fish predominantly from Lucinda, Townsville and the Brisbane River, and one with relatively high Ba (Group B), that contained a mix of fish predominantly from the Fitzroy and Mary Rivers (Fig. 5, Table 4).

Relationships between elemental concentrations and biological and environmental variables

No significant correlation was evident between otolith Ba concentration and any biological or environmental variable for material laid down in Year 1,

Table 3. *Polydactylus macrochir*. Percentage of individuals assigned to each group identified by cluster analysis from locations in the Gulf of Carpentaria, Australia. See Fig. 1 for location codes

Otolith region Group	RR	AC	AR	MI	FLR	SC	KR	WLR
Core								
A	21	93	100	93	44	64	100	88
B	43	7	0	7	28	29	0	13
C	36	0	0	0	28	7	0	0
Year 1								
A	50	93	80	86	33	50	100	100
B	50	7	20	14	67	50	0	0
Year 2								
A	7	79	0	71	0	14	80	100
B	50	21	100	29	33	50	20	0
C	43	0	0	0	67	36	0	0
Year 3								
A	0	86	93	64	6	14	100	88
B	50	0	7	36	28	64	0	13
C	50	14	0	0	67	21	0	0

with the exception of peak monthly rainfall. For material laid down in Year 2, weak, yet significant, relationships existed between otolith ^{138}Ba concentration and incremental width and the 3 rainfall indices, while strong positive relationships were observed between otolith ^{138}Ba concentration and the 3 environmental flow indices for all age groups (Table 5). Similarly for material laid down in Year 3, while no significant relationship was evident between otolith ^{138}Ba concentration and incremental width, weak yet significant relationships existed between Ba concentration and the 3 rainfall indices, and strong positive relationships were observed between otolith ^{138}Ba concentration and the 3 environmental flow indices within this age group (Table 5).

DISCUSSION

Otolith chemistry of *Polydactylus macrochir* showed considerable variation among locations and with ontogeny, a result which is generally suggestive of limited connectivity and a complex population structure of *P. macrochir* among the estuaries of tropical and sub-tropical northern Australia. Although limited connectivity over similar spatial scales has been observed for a number of estuarine-associated fishes in temperate systems (e.g. Gillanders 2002, Bradbury et al. 2008), this study provides one of the first assessments of connectivity of a tropical, non-diadromous estuarine fish species.

Factors affecting elemental deposition

In the analysis, ^{138}Ba generally provided the most discrimination among locations, particularly for material laid down in Years 1, 2 and 3. Otolith Ba concentration is considered to be closely associated with ambient Ba concentration (Webb et al. 2012), and is typically enriched in low salinity environments such as freshwater or flood plumes, where it is desorbed from fine-grained suspended particles (Elsdon & Gillanders 2005, McCulloch et al. 2005). As such, the differences observed in ^{138}Ba concentrations in *Polydactylus macrochir* otoliths likely reflect differences in terrestrial runoff and freshwater flow among locations. While river flow data were limited to only a small number of locations, the positive association observed between otolith Ba concentration and river flow indices within the respective collection locations for all age groups suggests that freshwater flow is a significant (albeit likely indirect) driver of otolith Ba concentration in the study species. Moreover, the observed significant relationships between Ba concentration and river flow indices provides further evidence for limited movement of the study species. If fish were moving among estuaries of contrasting flow regimes following deposition of otolith material such relationships would be unlikely.

Relative to ^{138}Ba, ^{88}Sr offered little discrimination among locations. Strontium has been used successfully to discriminate among estuaries in a number of estuarine and coastal fish species in temperate waters (Gillanders et al. 2003). It is generally assumed that there is a positive relationship between otolith Sr and ambient salinity; however, recent studies suggest that otolith Sr is largely mediated by ambient Sr and temperature (Webb et al. 2012). In the present study, ^{88}Sr concentrations varied considerably over the life of an individual fish, with age-related ^{88}Sr profiles showing numerous peaks and troughs that exhibited little temporal consistency, suggesting they were not related to season. Such variation may reflect movement of fish between differing salinity profiles, or episodic changes in ambient salinity or temperature at a particular location.

Concentrations of ^{7}Li and ^{55}Mn were found to be generally elevated in the otolith core relative to the surrounding material. Although several studies have reported similar patterns of core enrichment in fish otoliths across a number of phylogenetically distinct species (Ruttenberg et al. 2005), the mechanisms responsible for the elevated concentrations and spatial variation are largely unknown. The consistent occurrence of core enrichment among locations over

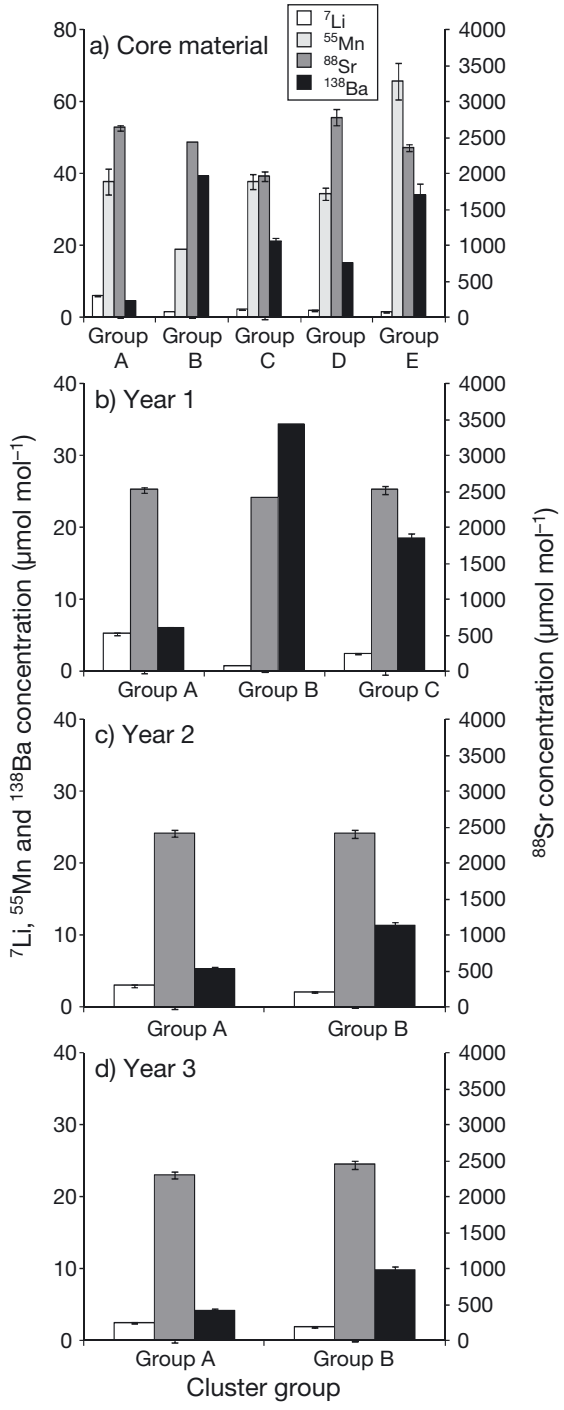

Fig. 5. *Polydactylus macrochir*. Mean elemental concentrations (µmol mol^{-1} relative to ^{43}Ca) for the groups identified by SIMPROF analysis on the east coast of Queensland (±1 SE)

such large spatial distances indicates that this phenomenon in *Polydactylus macrochir* may be attributed to similar physiological or maternal effects, as opposed to ambient concentrations or dietary influences. Furthermore, the spatial patterns in core

Table 4. *Polydactylus macrochir*. Percentage of individuals assigned to each group identified by cluster analysis from locations on the east coast of Queensland, Australia. See Fig. 1 for location codes

Otolith region Group	LUC	TSV	FR	MR	BR
Core					
A	100	100	0	0	0
B	0	0	0	0	10
C	0	0	0	10	90
D	0	0	0	90	0
E	0	0	100	0	0
Year 1					
A	100	100	0	0	0
B	0	0	0	0	10
C	0	0	100	100	90
Year 2					
A	100	100	42	10	80
B	0	0	58	90	20
Year 3					
A	100	90	8	0	70
B	0	10	92	100	30

Table 5. Correlation coefficients between otolith ^{138}Ba concentration (µmol mol^{-1} relative to ^{43}Ca) and biological and environmental variables. Significance: $^{**}p < 0.01$, ns = non-significant

Variable	Year 1 R^2	Year 2 R^2	Year 3 R^2
Increment width	$<0.01^{ns}$	0.09^{**}	0.01^{ns}
Mean monthly rainfall	$<0.01^{ns}$	0.15^{**}	0.10^{**}
Peak monthly rainfall	0.23^{**}	0.33^{**}	0.10^{**}
Total annual rainfall	$<0.01^{ns}$	0.16^{**}	0.07^{**}
Mean monthly flow	0.57^{**}	0.50^{**}	0.60^{**}
Peak monthly flow	0.51^{**}	0.50^{**}	0.62^{**}
Total annual flow	0.58^{**}	0.49^{**}	0.58^{**}

enrichment, particularly for ^{55}Mn, are consistent with results of recent genetic comparisons, suggesting that the deposition of these elements within the otolith core may be mediated at a genetic level. For example, while Horne et al. (2012) found no difference in mtDNA haplotypes among samples from Eighty Mile Beach and Roebuck Bay, these locations differed to all other locations examined; a result that is consistent with spatial patterns in otolith ^{55}Mn concentration observed in the present study. Similarly, fish from the Fitzroy and Brisbane Rivers are genetically distinct (Horne et al. 2012), and differed significantly in average Mn concentration in their otolith cores. Supporting this hypothesis, Clarke et al. (2011) observed significant differences in partition coefficients of a number of elements, including Mn, among genetically distinct populations of *Menidia menidia*

raised under identical laboratory conditions, concluding that such differences have (at least partially) a genetic basis.

Patterns of connectivity of *Polydactylus macrochir*

The patterns of connectivity evident from the otolith elemental data are generally consistent with those of concurrent investigations of mtDNA haplotypes (Horne et al. 2012), otolith stable isotope ratios (Newman et al. 2010), life history parameters (Moore et al. 2012a) and parasite assemblages (Moore et al. 2012b). However, several key discrepancies exist between the results of these complementary studies and the current study that warrant discussion, particularly for post-settlement life history stages. For example, while few differences in elemental signatures were evident among post-settlement fish from within the 3 geographic regions of this study (see Fig. 1), analysis of parasite assemblages and life history traits, conducted on many of the same individuals, indicated significant structuring and limited connectivity within the regions (Moore et al. 2012a, 2012b). The separation of populations within geographic regions, specifically for locations in north-west and northern Australia and the east coast of Queensland, is supported by the observed differences in otolith core elemental signatures.This result suggests isolation of both larval/early juvenile and post-settlement life history stages. If fish were mixing among these locations, it would be expected that their otolith core signatures would similarly show a mixed signal. In these cases it is likely that similarities in elemental signatures observed among these locations reflect homogeneity in the factors affecting elemental deposition, rather than movement of fish, among estuaries (Thresher 1999).

In contrast to the elemental data, otolith stable isotope ratios, life history parameters and parasite assemblages provided little evidence for spatial segregation of post-larval fish among estuaries in the south-eastern Gulf of Carpentaria (Newman et al. 2010, Moore et al. 2012a, 2012b). While homogeneity of elemental concentrations was observed among individuals collected from most locations, the elevated Ba signatures observed in the majority of fish from the Flinders River from Year 1 onwards relative to those from other estuaries in the region suggests that movement of post-settlement life history stages may be more restricted than that indicated by other methods. This result is consistent with conventional tagging data, which suggests that while individual fish are capable of travelling large distances, movement of adult fish among estuaries is generally limited (Welch et al. 2010). Given the significant structuring and fine spatial scale between the Flinders River and the neighbouring locations (in particular the Morning Inlet, which is approximately 50 km west of the Flinders River), it may be that similarities in elemental signatures observed elsewhere in this region reflect regional homogeneity in the factors affecting elemental deposition, in particular low freshwater flow rates, rather than broadscale movement of fish among estuaries. Ultimately, it is likely that *Polydactylus macrochir* in this region operate as a series of metapopulations, connected by a degree of non-trivial movement that is neither so low as to negate significant demographic connectivity, nor so high as to eliminate independence of local population dynamics (Kritzer & Sale 2004).

The differences observed among the various techniques used to assess connectivity of *Polydactylus macrochir* likely relate to the varying spatial and temporal scales the individual techniques operate on. Mitochondrial markers, such as those examined by Horne et al. (2012), operate over broad spatial and temporal timescales, and may be unable to distinguish among locations that share low levels of exchange or among recently separated populations (Ovenden et al. 2002). Both the parasite approach of Moore et al. (2012b) and the whole otolith isotope approach of Newman et al. (2010) were based on signals accumulated over an individual fish's entire life history, and thus may be unable to reveal short-term isolation of individuals within a mixed population. The confounding results observed amongst techniques highlights the importance of using multiple, complementary methods for assessing connectivity, in that such an approach effectively increases the chances of identifying differences between spatially distinct populations by both allowing for the varying scales and limitations of individual methods to be resolved, and allowing for greater confidence in results where consistent patterns are identified among different methodologies (sensu Begg & Waldman 1999).

In contrast to the differences observed in otolith material laid down following settlement, core elemental signatures for all locations in the Gulf of Carpentaria generally appeared similar. It is unclear whether such similarity results from mixing of these life history stages, the movement of fish following recruitment, or regional homogeneity in the factors affecting elemental deposition among locations. Spawning in *Polydactylus macrochir* in Queensland's

Gulf waters occurs between August and September (Garrett 1997), immediately prior to the austral wet season and at a time when freshwater flow into the Gulf of Carpentaria estuaries is typically low (DNRM 2012). As a consequence, the otolith cores showed little of the Ba enrichment that discriminated the later life history stages. Although little is known of the dispersal capabilities of *P. macrochir* larvae, Horne et al. (2011) observed significant differences in microsatellite loci of blue threadfin *Eleutheronema tetradactylum* among estuaries separated by as little as 15 km, concluding that such fine-scale genetic structuring resulted partly from a high rate of self-recruitment in this species. A similar recruitment strategy may occur in *P. macrochir*, and warrants further research.

The limited exchange of *Polydactylus macrochir* among estuaries evident from the elemental data and complementary studies is consistent with the few studies that have examined movements of tropical, non-diadromous estuarine fishes. Sheaves et al. (1999) reported that none of the 962 *Acanthopagrus berda* tagged in creeks in the Hinchinbrook Channel in tropical north Queensland moved to nearby creeks. Using microsatellite markers, Horne et al. (2011) revealed fine scale structuring in blue threadfin, *Eleutheronema tetradactylum*. The consistency of these results among studies suggests that limited movement of post-settlement life history stages among estuaries may be a feature of some tropical non-diadromous estuarine fish species, even where adjacent habitat appears conducive to movement, and may reflect the benefits offered by tropical estuaries. Movement among spatially-discrete habitat patches, such as estuaries, is energetically expensive, and typically renders organisms more susceptible to predation (Dingle 1996, Dodson & Godin 1997). Thus, to be evolutionarily advantageous, the benefits of movement — be they environmental, trophic or reproductive — must ultimately outweigh the associated costs (Dodson & Godin 1997). Tropical estuaries are frequently highly turbid, relatively thermally stable and inherently productive ecosystems, typically supporting greater biomass and diversity of aquatic organisms than their associated coastal foreshores or temperate counterparts (Blaber 2002). As such, there may be little biological or environmental requirement or overall benefit for fish to move among estuaries. For *P. macrochir*, it may be that the highly productive nature of tropical estuarine systems (Blaber 2002), combined with the species' generalist diet (Brewer et al. 1995), proposed high degree of euryhalinity for all life history stages, hermaphroditic nature, and overlap in the distribution of sexes (Halliday et al. 2008) results in little requirement or advantage for an individual to move frequently among estuaries. Examination of additional species, and the mechanisms influencing their connectivity, is required to further test this hypothesis.

It should be noted that the age-related approach used here places limitations on the interpretations that can be made regarding the connectivity of larval *Polydactylus macrochir* to adult populations. Despite the significant differences in core elemental signatures observed among locations on the north-western and east coasts, it cannot be unequivocally concluded using the age-related profile approach that each fish caught at a particular location originated from that specific location. Rather, the results suggest that fish caught at these locations have a different natal origin to those from the other locations examined. An increasing number of studies that have documented self-recruitment in estuarine species (North & Houde 2006, Braverman et al. 2009), and it may be that the natal origins of each population are proximate to the respective collection locations and result from self-recruitment of adult populations. More direct approaches, such as those in which otolith elemental signatures of larvae or early juveniles (which represent natal origins) are compared with those of adults of the same cohort (e.g. Gillanders 2002), are required to identify the natal origins of the adults. Nevertheless, given the variation observed in elemental signatures, we consider that the age-related approach adopted here provides a viable, albeit indirect, alternative to assessing patterns of connectivity. As such, the technique may be particularly suitable for species in which collection of larval or early juvenile life history stages is not feasible, such as those that occur in geographically remote environments or for species whose larval and/or juvenile habitats are unknown.

This study has provided further evidence that analysis of otolith elemental concentrations is a valuable tool in discerning connectivity and movement of tropical estuarine fishes across their life history, particularly among waters with differing environmental regimes. The limited connectivity evident from the otolith elemental data and other complementary studies suggests that *Polydactylus macrochir* is particularly vulnerable to localised depletion, a result that should be accounted for when making management decisions for this species. Moreover, the results highlight the importance of using multiple, complementary methods for assessing connectivity of aquatic organisms and, given the paucity of studies examining connectivity of non-diadromous tropical

estuarine fishes, provide fundamental information from which more specific hypotheses of connectivity, population structure and gene flow of such species can be tested.

Acknowledgements. We are greatly indebted to D. Welch and A. Ballagh (James Cook University), Q. Allsop (Northern Territory Fisheries), J. Stapley (Queensland Department of Primary Industries and Fisheries), and G. Mitsopoulos, S. Newman, M. Pember, B. Rome and C. Skepper (Department of Fisheries, Western Australia) for assistance with sample collection, together with the numerous commercial and recreational fishers and fish processors who kindly supplied samples. N. Munksgaard and F. Foti (Charles Darwin University) provided training and advice on the LA-ICPMS. We thank I. Nagelkerken, B. Gillanders, R. Allman, R. Lester, D. Welch and 3 anonymous reviewers for their constructive comments on earlier drafts of the manuscript. B.R.M. was supported by an Australian Postgraduate Award. Funding for this project was provided in the form of research grants from James Cook University's School of Earth and Environmental Sciences and Graduate Research School and the Australian Fisheries Research and Development Corporation (Project no. 2007/032).

LITERATURE CITED

Begg GA, Waldman JR (1999) An holistic approach to fish stock identification. Fish Res 43:35–44

Blaber SJM (2000) Tropical estuarine fishes: ecology, exploitation and conservation. Blackwell Science, Oxford

Blaber SJM (2002) 'Fish in hot water': the challenges facing fish and fisheries research in tropical estuaries. J Fish Biol 61:1–20

BoM (Bureau of Meteorology) (2013) Climate Data Online www.bom.gov.au/climate/data (accessed 15 September 2013)

Bradbury IR, Campana SE, Bentzen P (2008) Otolith elemental composition and adult tagging reveal spawning site fidelity and estuarine dependency in rainbow smelt. Mar Ecol Prog Ser 368:255–268

Braverman MS, Acha EM, Gagliardini DA, Rivarossa M (2009) Distribution of whitemouth croaker (*Micropogonias furnieri*, Desmarest 1823) larvae in the Rio de la Plata estuarine front. Estuar Coast Shelf Sci 82:557–565

Brewer DT, Blaber SJM, Salini JP, Farmer MJ (1995) Feeding ecology of predatory fishes from Groote Eylandt in the Gulf of Carpentaria, Australia, with special reference to predation on penaeid prawns. Estuar Coast Shelf Sci 40:577–600

Campana SE, Neilson JD (1985) Microstructure of fish otoliths. Can J Fish Aquat Sci 42:1014–1032

Clarke LM, Thorrold SR, Conover DO (2011) Population differences in otolith chemistry have a genetic basis in *Menidia menidia.* Can J Fish Aquat Sci 68:105–114

Cowen RK, Gawarkiewicz GG, Thorrold JP Sr, Werner FE (2007) Population connectivity in marine systems — an overview. Oceanography (Wash DC) 20:14–21

Dingle H (1996) Migration: the biology of life on the move. Oxford University Press, Oxford

DNRM (Department of Natural Resources and Mines) (2012) Water Monitoring Data Portal. http://watermonitoring.dnrm.qld.gov.au/host.htm (accessed 10 July 2013)

Dodson JJ, Godin JGJ (1997) Fish migration: an evolutionary perspective. In: Godin JGJ (ed) Behavioural ecology of teleost fishes. Oxford University Press, Oxford, p 10–36

Elsdon TS, Gillanders BM (2005) Alternative life-history patterns of estuarine fish: barium in otoliths elucidates freshwater residency. Can J Fish Aquat Sci 62: 1143–1152

Fowler AJ, Campana SE, Jones CM, Thorrold SR (1995) Experimental assessment of the effects of temperature and salinity on elemental composition of otoliths using solution-based ICPMS. Can J Fish Aquat Sci 52: 1421–1430

Garrett RN (1997) Biology and harvest of tropical fishes in the Queensland Gulf of Carpentaria gillnet fishery. Final report to the Fisheries Research & Development Corporation, Project 1992/145. Queensland Government Department of Primary Industries, Brisbane

Gillanders BM (2002) Connectivity between juvenile and adult fish populations: Do adults remain near their recruitment estuaries? Mar Ecol Prog Ser 240:215–223

Gillanders BM (2009) Tools for studying biological marine ecosystem interactions-natural and artificial tags. In: Nagelkerken I (ed) Ecological connectivity among tropical coastal ecosystems. Springer, New York, NY, p 457–492

Gillanders BM, Able KW, Brown JA, Eggleston DB, Sheridan PF (2003) Evidence of connectivity between juvenile and adult habitats for mobile marine fauna: an important component of nurseries. Mar Ecol Prog Ser 247:281–295

Halliday IA, Robins JB, Mayer DG, Staunton-Smith J, Sellin MJ (2008) Effects of freshwater flow on the year-class strength of a non-diadromous estuarine finfish, king threadfin (*Polydactylus macrochir*), in a dry-tropical estuary. Mar Freshw Res 59:157–164

Hilborn R, Walters CJ (1992) Quantitative fisheries stock assessment: choice, dynamics and uncertainty. Routledge, Chapman & Hall, New York, NY

Horne JB, Momigliano P, Welch DJ, Newman SJ, van Herwerden L (2011) Limited ecological population connectivity suggests low demands on self-recruitment in a tropical inshore marine fish (*Eleutheronema tetradactylum*: Polynemidae). Mol Ecol 20:2291–2306

Horne JB, Momigliano P, Welch DJ, Newman SJ, van Herwerden L (2012) Searching for common threads in threadfins: phylogeography of Australian polynemids in space and time. Mar Ecol Prog Ser 449:263–276

Jones CM (2006) Estuarine and diadromous fish metapopulations. In: Kritzer JP, Sale PF (eds) Marine metapopulations. Elsevier Academic Press, Burlington, MA, p 119–154

Kailola PJ, Williams MJ, Stewart PC, Reichelt RE, McNee A, Grieve C (1993) Australian fisheries resources. Bureau of Resource Sciences, Department of Primary Industries & Energy, Canberra

Kritzer JP, Sale PF (2004) Metapopulation ecology in the sea: from Levins' model to marine ecology and fisheries science. Fish Fish 5:131–140

McCulloch M, Cappo M, Aumend J, Muller W (2005) Tracing the life history of individual barramundi using laser ablation MC-ICP-MS Sr-isotopic and Sr/Ba ratios in otoliths. Mar Freshw Res 56:637–644

Milton DA, Chenery SR (2003) Movement patterns of the tropical shad hilsa (*Tenualosa ilisha*) inferred from transects of $^{87}Sr/^{86}Sr$ isotope ratios in their otoliths. Can J Fish Aquat Sci 60:1376–1385

Moore BR, Welch DJ, Simpfendorfer CA (2011) Spatial patterns in the demography of a large estuarine teleost: king threadfin, *Polydactylus macrochir*. Mar Freshw Res 62: 937–951

Moore BR, Simpfendorfer CA, Newman SJ, Stapley JM, Allsop Q, Sellin MJ, Welch DJ (2012a) Spatial variation in life history reveals insight into connectivity and geographical population structure of a tropical estuarine teleost: king threadfin, *Polydactylus macrochir*. Fish Res 125-126:214–224

Moore BR, Welch DJ, Newman SJ, Lester RJG (2012b) Parasites as indicators of movement and population connectivity of a non-diadromous, tropical estuarine teleost: king threadfin, *Polydactylus macrochir*. J Fish Biol 81: 230–252

Motomura H (2004) Threadfins of the world (family Polynemidae). An annotated and illustrated catalogue of polynemid species known to date. Food and Agriculture Organisation of the United Nations (FAO), Rome

Motomura H, Iwatsuki Y, Kimura S, Yoshino T (2000) Redescription of *Polydactylus macrochir* (Günther, 1867), a senior synonym of *P. sheridani* (Macleay, 1884) (Perciformes: Polynemidae). Ichthyol Res 47:327–333

Newman SJ, Allsop Q, Ballagh AC, Garrett RN and others (2010) Variation in stable isotope ($\delta^{18}O$ and $\delta^{13}C$) signatures in the sagittal otolith carbonate of king threadfin, *Polydactylus macrochir* across northern Australia reveals multifaceted stock structure. J Exp Mar Biol Ecol 396: 53–60

North EW, Houde ED (2006) Retention mechanisms of white perch (*Morone americana*) and striped bass (*Morone saxatilis*) early-life stages in an estuarine turbidity maximum: an integrative fixed-location and mapping approach. Fish Oceanogr 15:429–450

Ovenden JR, Lloyd J, Newman SJ, Keenan CP, Slater LS (2002) Spatial genetic subdivision between northern Australian and southeast Asian populations of *Pristipomoides multidens*: a tropical marine reef fish species. Fish Res 59:57–69

Pearce NJG, Perkins WT, Westgate JA, Gorton MP, Jackson SE, Neal CR, Chenery SP (1997) A compilation of new and published major and trace element data for NIST SRM 610 and NIST SRM 612 glass reference materials. Geostand Newsl 21:115–144

Pember MB, Newman SJ, Hesp SA, Young GC, Skepper CL, Hall NG, Potter IC (2005) Biological parameters for managing the fisheries for blue and king threadfin salmons, estuary rockcod, Malabar grouper and mangrove jack in north-western Australia. Final report to the Fisheries Research & Development Corporation, Project 2002/003. Centre for Fish and Fisheries Research, Murdoch University, Perth

Roessig JM, Woodley CM, Cech JJ, Hansen LJ (2004) Effects of global climate change on marine and estuarine fishes and fisheries. Rev Fish Biol Fish 14:251–275

Russell DJ, Garrett RN (1988) Movements of juvenile barramundi, *Lates calcarifer* (Bloch), in north-eastern Queensland. Aust J Mar Freshw Res 39:117–123

Ruttenberg BI, Hamilton SL, Hickford MJH, Paradis GL and others (2005) Elevated levels of trace elements in cores of otoliths and their potential for use as natural tags. Mar Ecol Prog Ser 297:273–281

Secor DH, Rooker JR (2000) Is otolith strontium a useful scalar of life cycles in estuarine fishes? Fish Res 46:359–371

Secor DH, Rooker JR (2005) Connectivity in the life histories of fishes that use estuaries. Estuar Coast Shelf Sci 64:1–3

Sheaves MJ, Molony BW, Tobin AJ (1999) Spawning migrations and local movements of a tropical sparid fish. Mar Biol 133:123–128

Thorrold SR, Latkoczy C, Swart PK, Jones CM (2001) Natal homing in a marine fish metapopulation. Science 291: 297–299

Thresher RE (1999) Elemental composition of otoliths as a stock delineator in fishes. Fish Res 43:165–204

Van Achterbergh E, Ryan C, Jackson S, Griffin W (2001) Data reduction software for LA–ICPMS. In: Sylvester P (ed) Laser-Ablation–ICPMS in the earth sciences: principles and applications. Mineralogical Association of Canada, Ottawa, p 239–243

Webb SD, Woodcock SH, Gillanders BM (2012) Sources of otolith barium and strontium in estuarine fish and the influence of salinity and temperature. Mar Ecol Prog Ser 453:189–199

Welch DJ, Ballagh A, Newman SJ, Lester RJ, and others (2010) Defining the stock structure of northern Australia's threadfin salmon species. Final report to the Fisheries Research & Development Corporation, Project 2007/032. Fishing & Fisheries Research Centre, James Cook University, Townsville

Yoshinaga J, Nakama A, Morita M, Edmonds JS (2000) Fish otolith reference material for quality assurance of chemical analyses. Mar Chem 69:91–97

Editorial responsibility: Ivan Nagelkerken, Adelaide, South Australia, Australia

Submitted: September 14, 2012; Accepted: November 13, 2013
Proofs received from author(s): February 26, 2014