CSIRO PUBLISHING

www.publish.csiro.au/journals/mfr *Marine and Freshwater Research*, 2005, **56**, 655–660

Retrospective determination of natal habitats for an estuarine fish with otolith strontium isotope ratios

James A. Hobbs[A,C], *Qing-zhu Yin*[B], *Jessica Burton*[A] *and William A. Bennett*[A]

[A]Bodega Marine Laboratory, University of California, Davis, John Muir Institute of the Environment, University of California, Davis, One Shields Avenue, Davis, CA 95616, USA.
[B]Department of Geology and UC Davis Interdisciplinary Center for Plasma Mass Spectrometry, University of California, Davis, One Shields Avenue, Davis, CA 95616, USA.
[C]Corresponding author. Email: jahobbs@ucdavis.edu

Abstract. We investigated the ability of strontium isotope ratios ($^{87}Sr/^{86}Sr$) in otolith cores to record the natal habitats of juvenile delta smelt *Hypomesus transpacificus* from the San Francisco Estuary, USA. Young delta smelt ($<$60 days old) were collected during the California Department of Fish and Game 20-mm Survey in May and June of 1999 at several potential natal areas: Napa River, Suisun Marsh, West Delta, North Delta, Central Delta, South Delta and East Delta. The core region of sagittal otoliths was assayed with laser ablation-multicollector inductively coupled plasma mass spectroscopy. The laser ablation technique provided precise estimates of $^{87}Sr : ^{86}Sr$ ratios with relative standard deviation of 0.003% (one sigma). Isotope ratios ranged from 0.7065 to 0.708 and were different among natal habitats. However, natal habitats within the delta region were not discernable among each other, and reflect the mixing of the two major rivers, Sacramento River and San Joaquin River within the delta. We will therefore be able to determine natal habitats for delta smelt by assaying the core region of the otoliths. The application of strontium isotope ratios ($^{87}Sr/^{86}Sr$) in fish otoliths will greatly improve conservation efforts for this protected species.

Extra keywords: delta smelt, natal origins, otoliths, strontium isotopes.

Introduction

Retrospective determination of natal habitats and migration history using otolith geochemical composition has resulted in significant advances in fish ecology and fisheries management. For example, trace elements have proven useful for recording the natal river, environmental history and migration patterns of American shad *Alossa sapidissima* in several east coast rivers, and the degree of natal homing in marine species (Limburg 1995; Thorrold *et al*. 1998, 2001). However, the application of trace elements for determining habitat use is based on consistent differences in water chemistry between different habitats used by a species. It is now well established that the chemical composition within otoliths mostly reflect the composition of the water (Simkiss 1974; Farrell and Campana 1996), although other studies have shown that temperature, salinity and diet can influence the otolith composition, including strontium isotopes (Fowler *et al*. 1995; Kennedy *et al*. 1997; Dove and Kingsford 1998). The use of strontium isotopes as natural geochemical tracers of environmental history has circumvented this problem. The ratio of strontium isotopes in fish otoliths are not physiologically regulated and may provide a much more precise geochemical signature (Kennedy *et al*. 1997; Outridge *et al*. 2002).

Geographic differences in strontium isotopic ratios in the environment are primarily derived from the underlying geological formations. Radiogenic decay of ^{87}Rb to ^{87}Sr and the resulting ratio of $^{87}Sr/^{86}Sr$ reflects the lithologies of distinct watershed. Because strontium isotope ratios vary over geologic time, they provide a stable geographic signature in ecological studies (Dickin 1995). However, differences in strontium isotope ratios in the environment are subtle, requiring high-resolution tools to quantify variability. Measurements of strontium isotope ratios with quadrapole inductively coupled plasma mass spectroscopy (ICP-MS) results in precision less than 1% relative standard deviation (RSD) for solution-based and laser ablation (LA) techniques (Machado and Gauthier 1996). Magnetic sector single or multi-collector (MC) ICP-MS provides much higher measure of precision with RSD of 0.1% and 0.04% respectively (Thorrold and Shuttleworth 2000; Outridge *et al*. 2002).

Recently strontium isotope ratios in fish otoliths have been used to identify stock structure, describe migration history (Kennedy *et al*. 2002) and to determine natal stream origins for migratory salmonids. For example, Kennedy *et al*. (1997) measured strontium isotope ratios ($^{87}Sr/^{86}Sr$) in Atlantic salmon among different rivers in two watersheds of the Connecticut River, and determined that several streams lying in

 10.1071/MF04136 1323-1650/05/050655

unique geological landscapes had unique strontium isotopic ratios. In the Sacramento–San Joaquin watersheds, (Ingram and Weber 1999) determined that Fall–Run Chinook salmon also exhibited unique strontium isotope ratios specific to natal rivers within each watershed. Delta smelt undergo similar migrations in the San Francisco Estuary (SFE), but on much smaller scales, with adults migrating upstream, landward of salinity intrusion in the estuary. Depending on the amount of freshwater flow the distance between estuarine rearing habitat and freshwater spawning habitat can be less than 10 km. Several major rivers feed directly into the SFE, across a distance of 60 km, including the Sacramento River, San Joaquin River, Mokelumne River and the Napa River. Consequently, this results in a high degree of spatial variability for natal habitats. Because delta smelt spawn in spatially variable habitats, strontium isotope ratios might be used to determine the natal source for this threatened species. To take advantage of micro-scale difference in habitat use in the life history of the delta smelt, the use of laser ablation techniques with high spatial resolution can significantly advance the application of strontium isotope in otoliths to record natal habitats. In the present study, we seek to advance the scope of strontium isotope ratios to act as natural tags for freshwater habitats at a small spatial scale <60 km in the SFE and Sacramento–San Joaquin Delta. The precision of $^{87}Sr/^{86}Sr$ ratio is significantly improved (0.003% RSD) compared with that of Outridge *et al.* (2002) at 0.04% RSD, and comparable with the thermal ionisation mass spectrometry (TIMS) technique used in Kennedy *et al.* (1997, 2000, 2002). The LA-ICP-MS technique has an added advantage of high spatial resolution compared with that of micro-drilling technique or whole otolith dissolution (Kennedy *et al.* 1997, 2000, 2002; Ingram and Weber 1999) and the analyses are done *in situ* and much less time-consuming.

Materials and methods

Juvenile delta smelt, *Hypomesus transpacificus*, were collected in May–June 1999 at seven sites: Napa River (N), North Delta, on the lower Sacramento River (ND), the Central Delta near the Mokelumne River (CD), the South Delta near the State and Federal Water Export Facilities (SD), the East Delta on the lower San Joaquin River (ED), the West Delta at the confluence of the Sacramento and San Joaquin River near Sherman Island (WD) and Suisun Marsh (SM) (Fig. 1). These areas were chosen based on the occurrence of yolk-sac larvae from monitoring surveys conducted by the California Department of Fish and Game (http://www.delta.dfg.ca.gov/data/20mm, verified June 2005). In the laboratory, the fish were measured for standard length. The sagittal otoliths were removed, weighed on a microbalance, and then cleaned in Milli-Q water. Otoliths were mounted on glass slide, affixed with Crystal bond thermoplastic wax and polished on both sides with 3-micron lapping film. Otoliths were then washed with 1% nitric acid for 5 to 10 s, rinsed in an ultrasonic waterbath for 5 min, and dried under a class 100 laminar flow hood.

Otolith strontium isotope ratios

Strontium isotopic compositions were measured at UC Davis Interdisciplinary Center for Plasma Mass Spectrometry (http://icpms.ucdavis.edu, verified June 2005). A multi-collector inductively coupled plasma mass spectrometer (*Nu Plasma HR* from Nu Instrument Inc.) is interfaced with a Nd : YAG 213 nm laser (New Wave Research UP213, Fremont, CA) for the Sr isotope measurement by laser ablation (LA-MC-ICP-MS technique). Single spots at the core of the otoliths were assayed using a laser beam size of 60 micron, 100% laser power, and 10 Hz repetition rate. Typical ^{88}Sr signals of 2–6 V were obtained during the analyses. He gas is used as a carrier gas to maximise sensitivity and minimise sample deposition at ablation site and mixed with additional Ar after the laser sample cell but before sending the aerosols to the Plasma source. Gas blank and background signals are monitored until ^{84}Kr and ^{86}Kr is stable after the sample change (i.e. exposing sample cell to the air) and measured for 30 s. Then the laser is turned on typically for 30–60 s. Background is subtracted from the measured signals automatically. The $^{86}Sr/^{88}Sr = 0.1194$ is used to correct for instrumental fractionation with the exponential law. The peak intensities for ^{88}Sr, ^{87}Sr, ^{86}Sr, ^{85}Rb, and ^{84}Sr are measured simultaneously. The ^{85}Rb peak is monitored to correct for any ^{87}Rb interference on ^{87}Sr.

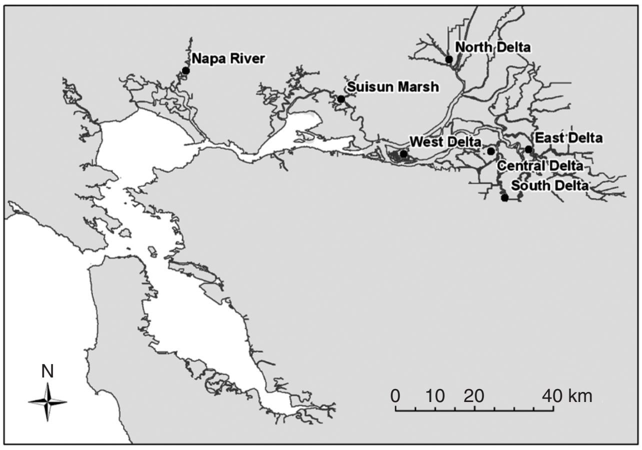

Fig. 1. Map of sampling locations – natal habitats.

Statistical analyses of strontium isotope ratios were carried out with a univariate approach. Analysis of variance (ANOVA) was used to test for differences in fish length and otolith weight from each natal habitat. Because of small differences in fish length and otolith weight among natal habitats, analysis of covariance (ANCOVA) with fish length as the covariate was used to test the null hypothesis of no significant difference in strontium isotope ratios among habitats, while adjusting means for the fish length covariate. We report only the ANCOVA results with fish length as the covariate as similar results were found for otoliths weight. The ANCOVA approach that was used is dependent on the assumption of homogeneity of slopes between the treatment (habitat) and the covariate (fish length), which was tested for by examining significance of the interaction term (habitat × fish length). Bonferroni corrections were applied to make comparisons among each habitat. The assumption of normality and the equality of the variance were assessed by examining for normal distribution of errors and homogeneity of variance using residual analysis (Winer *et al.* 1991). All statistical analyses were conducted with SYSTAT 10.0 (SYSTAT software Inc., http://www.systat.com/, verified June 2005).

Results

Juvenile delta smelt collected from the hypothesised natal areas ranged in size from 17 to 28 mm standard length (Table 1). Fish captured in the Sacramento River and Napa River were larger in length (ANOVA: d.f. = 4,26;

Table 1. Summary information for delta smelt collected for otolith strontium isotope chemistry comparisons at natal habitats
Shown are dates of collection (months per year), mean standard length (SL, mm), and mean otolith weight (OW, mg)

Location	Date	*n*	Mean SL ± s.e.	Mean OW ± s.e.
Napa	June 1999	5	27.8 ± 1.8	0.214 ± 0.031
North Delta	May 1999	4	25.5 ± 2.2	0.176 ± 0.034
West Delta	June and July 1999	3	20.4 ± 2.0	0.165 ± 0.025
Central Delta	May 1999	4	16.4 ± 1.6	0.164 ± 0.021
South Delta	May 1999	2	23.5 ± 2.6	0.147 ± 0.037
East Delta	May 1999	4	21.5 ± 2.1	0.169 ± 0.032
Suisun Marsh	May 1999	3	24.1 ± 2.4	0.170 ± 0.042

MS = 8.75; $F = 11.22$; $P < 0.001$) and otolith weight (ANOVA: d.f. = 4,26; MS = 0.019; $F = 12.74$; $P < 0.001$). Greater length and otolith weight for Sacramento River and Napa River fish reflected earlier spawning and older age.

The strontium isotope standard NIST SRM 987 was measured throughout the course of the present study. Figure 2*a* shows the reproducibility of the standard measured in solution nebulisation mode, with an average $^{87}Sr/^{86}Sr$ ratio = 0.710256 ± 45 (2σ s.d., % RSD 0.006%). The SRM 987 carbonate standard was also measured in laser ablation mode, with an average $^{87}Sr/^{86}Sr$ ratio = 0.710235 ± 49, and % RSD 0.006% (Fig. 2*b*). Both values compare favourably with the nominal value of 0.710248 for SRM 987. An in-house natural carbonate standard (WA-13) was also measured at the beginning of each analytical session to document the reproducibility of the laser ablation MC-ICP-MS technique (Fig. 2*c*). This standard is shown to be isotopically homogeneous with an average $^{87}Sr/^{86}Sr$ ratio = 0.707761 ± 38 (2σ). Collectively, these standards measurements document the accuracy and reproducibility of the $^{87}Sr/^{86}Sr$ ratio measured by the LA-MC-ICP-MS technique.

Strontium isotope ratios for fish collected near sites with known water strontium isotope ratios (Ingram and Sloan 1992; Ingram and Weber 1999) showed good agreement with a 1 : 1 relationship, suggesting the isotopic signature in the otolith reflects water isotopic ratios for that area (Fig. 3). Strontium isotopes were highly variable among individual fish varying from 0.706268 to 0.708378; with the highest values in the Napa River and the lowest in the North Delta sites (Table 2). Strontium isotope ratios differed among the sampled sites representing natal habitats for delta smelt (ANCOVA: d.f. = 6; $F = 77.33$; $P < 0.001$). (Table 3). The covariate fish length did not have a significant effect on the analysis suggesting fish of different size was not a confounding factor (ANCOVA: d.f. = 1; $F = 1.28$; $P = 0.282$) (Table 3), thus means were values for each sample were not adjusted for fish length. The homogeneity of slopes test was also not significant (d.f. = 5; $F = 0.67$; $P = 0.578$), thus fish length had a very small effect on otolith strontium isotope

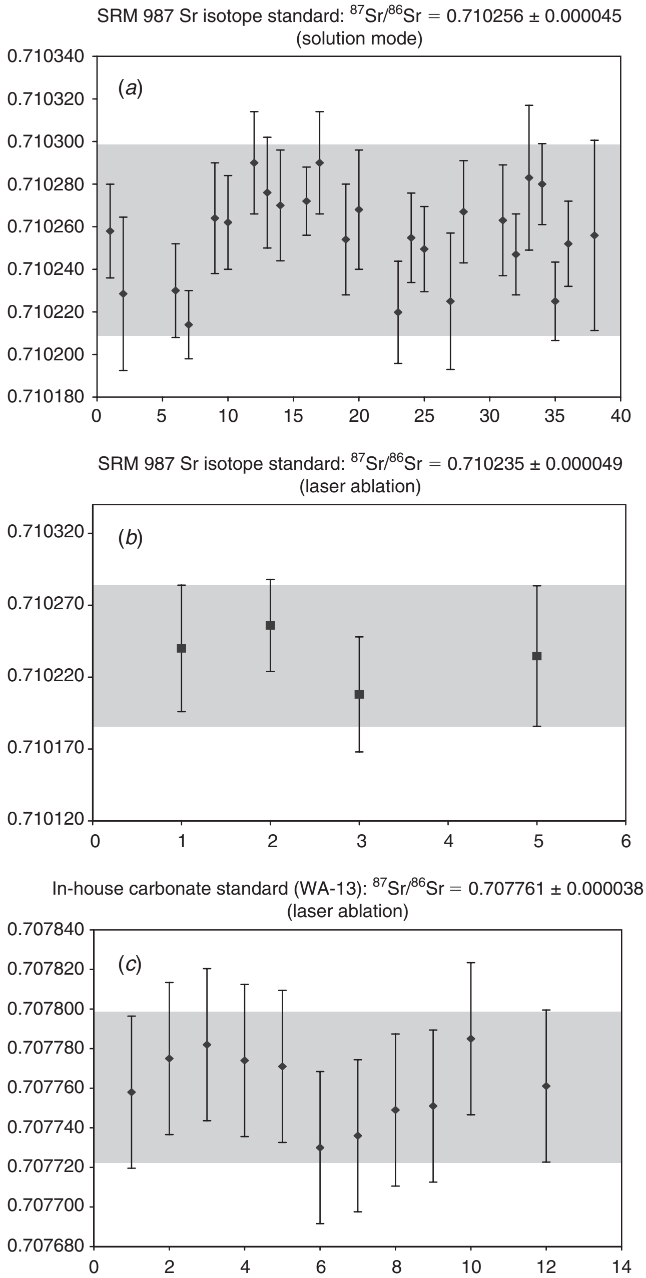

Fig. 2. (*a*) The NIST Sr isotope standard SRM 987 measured in solution nebulisation mode throughout the course of the present study, with two sigma external reproducibility shown in the grey band. The average $^{87}Sr/^{86}Sr = 0.710256 \pm 45$ (two sigma). (*b*) The NIST SRM 987 standard was also measured in laser ablation mode. The average $^{87}Sr/^{86}Sr = 0.710235 \pm 49$ (two sigma). (*c*) In-house natural carbonate standard with a homogeneous Sr isotopic composition. The average $^{87}Sr/^{86}Sr = 0.707761 \pm 38$ (two sigma). These standards demonstrate the measure of accuracy and reproducibility of the technique used for the current study.

ratio. This was to be expected considering the laser ablation technique allows for detailed sampling by focusing the laser beam at similar points on the otolith, which correspond with the same time points. Bonferonni corrections were applied

Table 2. Strontium isotope ratios for individual delta smelt collected from natal habitats in the San Francisco Estuary, USA

Region	Sample number	$^{87}Sr/^{86}Sr$		Region average
Napa River	1	0.708129	±52	
	2	0.708130	±52	
	3	0.707943	±98	
	4	0.708378	±50	
	5	0.708050	±62	0.708126 ± 160
North Delta	6	0.706467	±56	
	7	0.706268	±52	
	8	0.706267	±56	
	9	0.706336	±62	0.706334 ± 94
South Delta	10	0.706894	±40	
	11	0.706956	±106	0.706925 ± 44
East Delta	12	0.706863	±44	
	13	0.706804	±38	
	14	0.706952	±68	
	15	0.706944	±98	0.706890 ± 70
Central Delta	16	0.706622	±44	
	17	0.707021	±102	
	18	0.706810	±90	
	19	0.706724	±64	0.706794 ± 170
West Delta	20	0.706672	±38	
	21	0.706830	±80	
	22	0.706810	±74	0.706771 ± 86
Suisun Marsh	23	0.707373	±68	
	24	0.707477	±82	
	25	0.707446	±84	0.707432 ± 54

Table 3. ANCOVA table for strontium isotope ratios ($^{87}Sr/^{86}Sr$) with natal habitats

Fish length was analysed as a covariate as a result of the differences in sizes of fish from each area at capture

Variables	d.f.	MS	F	P
Site	6	1.13×10^{-6}	77.33	<0.001
Length	1	1.80×10^{-8}	1.28	0.282
Site × length	5	1.02×10^{-8}	0.67	0.578
Error	11	2.35×10^{-7}		

to compare means of each natal habitat. The Napa River, North Delta and Suisun Marsh were significantly different than the remaining Delta sites and each other at the $P = 0.001$ level. However, the West, Central, East and South Delta were indistinguishable (Fig. 4).

Discussion

The results of the present study indicated that LA-MC-ICP-MS technique we have developed provided high precision estimates of $^{87}Sr/^{86}Sr$ ratios with a laser spot size of 60 μm. Laser spot sampling of the NIST SRM 987 standard gave precision estimates of 0.006% RSD and accuracy within 2σ of the accepted values of $^{87}Sr/^{86}Sr$. Precision estimates of $^{87}Sr/^{86}Sr$ ratios at a spot size of 60 μm were an order of magnitude greater than those measured with quadrapole and sector field single collector ICP-MS, (Thorrold and Shuttleworth 2000), significantly improved over the similar technique used by Outridge *et al.* (2002) and were similar to the TIMS technique (Kennedy *et al.* 1997; Outridge *et al.* 2002).

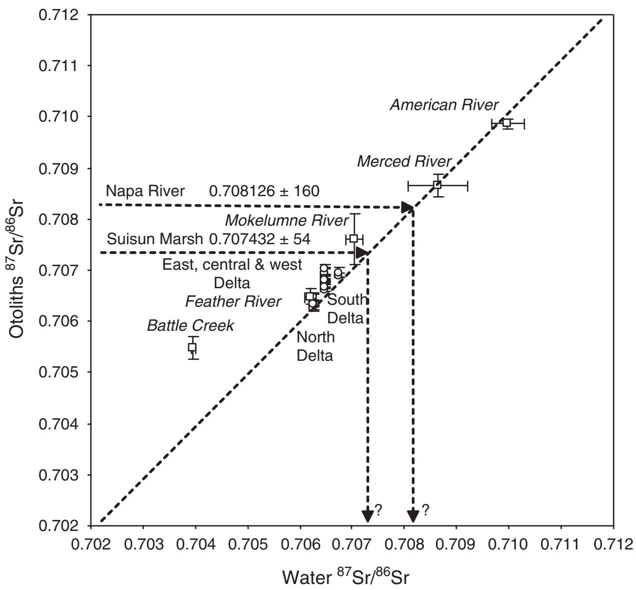

Fig. 3. Mean strontium isotope ratios (2σ) for natal habitats plotted against water strontium isotope ratios measured by Ingram and Sloan (1992) and Ingram and Weber (1999). Otolith strontium isotope values for the Napa River and Suisun Marsh are plotted near the 1 : 1 line to reflect potential water strontium isotope ratios for those areas. Italicised open squares are Chinook Salmon Sr isotope ratios from Ingram and Webber. Open circles are data for delta smelt in the present study.

Strontium isotope ratios identified four distinct natal areas within the SFE. Fish from the Napa River exhibited high $^{87}Sr/^{86}Sr$ ratio, whereas fish from the Sacramento River displayed a low $^{87}Sr/^{86}Sr$ ratio with range of 0.70636 ± 20. This value is consistent with Sacramento River water $^{87}Sr/^{86}Sr$ ratio of 0.706257 ± 40 (Ingram and Sloan 1992). Meanwhile, fish from Suisun Marsh expressed only moderately high $^{87}Sr/^{86}Sr$ ratio, while sites within the Delta revealed slightly lower $^{87}Sr/^{86}Sr$ ratio and were not significantly different from each other. Again we note that the waters at Centeral Delta ($^{87}Sr/^{86}Sr = 0.70646 \pm 36$) and South Delta ($^{87}Sr/^{86}Sr = 0.70674 \pm 50$) (Ingram and Weber 1999) compare favourably with juvenile delta smelt caught in the delta as measured in its otolith core (Table 2). This high degree of variability between locations within the SFE and positive correlations between otoliths and water $^{87}Sr/^{86}Sr$ ratio, allowed us to accurately identify delta smelt natal habitats.

Strontium isotope ratios in the environment are derived from the geological formations within the watershed and thus are useful as habitat markers in ecological timescales. Recent evidence suggests strontium isotope uptake occurs primarily

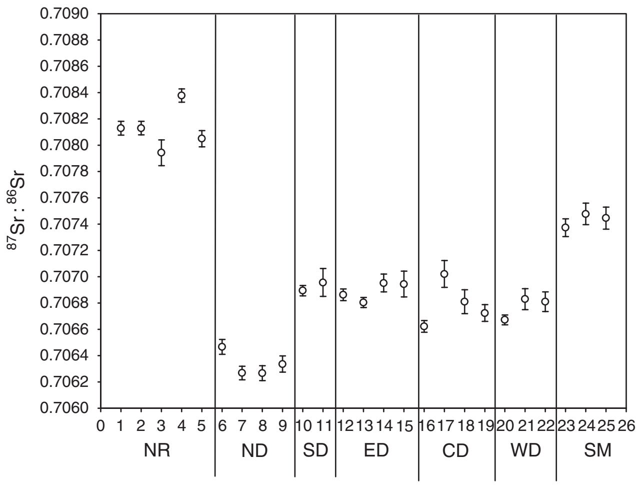

Fig. 4. Strontium isotope ratios ($^{87}Sr/^{86}Sr$) for individuals (2σ) collected at potential natal areas within the San Francisco Estuary (SFE). NR, Napa River; ND, North Delta; ED, East Delta; WD, West Delta; CD, Central Delta; SD, South Delta; SM, Suisun Marsh.

through food consumption in adult hatchery reared salmon (Kennedy *et al.* 2000), although other research has shown that a significant proportion of the signature is derived directly from the water (Simkiss 1974). In either case, strontium isotopes do not fractionate across trophic levels in natural foodwebs and thus strontium isotope ratios in fish otoliths should reflect the freshwater signature of the stream in which the fish lives (Blum *et al.* 2000). However, in estuarine habitats, small increases in salinity (i.e. values greater than two part per thousand (ppt) or 5% seawater can have significant influence on strontium isotope ratios.

Strontium isotope ratios for the Napa River and Suisun Marsh exhibited high strontium values (0.7079, 0.7074 respectively) for the SFE, reflecting the potential influence of salinity within the Napa River and Suisun Marsh (Fig. 2). Mean salinity during the spring in the Napa River was 5 ppt and 2 ppt for Suisun Marsh. However, we did not sample the freshwater end members for the Napa River or Suisun Marsh, thus strontium isotope ratios may be indicative salinity intrusions into these habitats. Regardless of the mechanism that results in the strontium signature for the Napa River and Suisun Marsh, the result was a unique isotopic signature that allowed us to accurately identify the fish to both the Napa River and Suisun Marsh habitats.

Accurate retrospective determination of natal habitats has important applications for understanding the population ecology of delta smelt. As delta smelt use a spatially variable spawning strategy, it is necessary that delta smelt are able to utilise many different areas of the SFE for spawning and rearing of young. Differences in habitat quality among these natal areas might result in different cohorts experiencing heterogeneous conditions during the critical early life stage. During the current study, water temperatures varied considerably between natal areas thus highlighting variability in environmental conditions among these natal areas. Differences in environmental conditions experienced by larvae can have a significant influence on their recruitment potential (Crecco and Savoy 1985; Houde 1989; Cushing and Horwood 1994). Moreover, contaminant levels with the SFE have been shown to vary spatially throughout the estuary and may also significantly impact the delta smelt population (Bennett and Moyle 1996; Thompson *et al.* 2000; Kuivila and Moon 2004; Bennett 2005). The ability to identify natal areas for delta smelt in a spatially heterogeneous environment will allow us to identify the habitats used by adult delta smelt and ultimately lead to better assessment of nursery habitats, factors influencing recruitment success and effective management of this threatened fish population.

Conclusions

The present study clearly shows that LA-MC-ICP-MS provided high precision measurements of otolith strontium isotope ratios with higher spatial resolution than the microdrilling or whole otolith dissolution TIMS technique. Strontium isotope ratios in delta smelt were derived primarily from the surrounding fresh waters in the natal habitats, although low salinity waters in two of the natal habitats may have an influence of strontium isotope ratios. Strontium isotope ratios were distinct among the natal habitats for the delta smelt and will provide an accurate means to identify the natal habitats for this protected species and will ultimately lead to better management and conservation strategies in the SFE.

Acknowledgments

The measurements were performed at the UC Davis Interdisciplinary Center for Plasma Mass Spectrometry (UCD-ICP-MS). Funding by NSF and UC Davis for the facilities is sincerely acknowledged. We thank the Central Valley Branch of the California Department of Fish and Game for assistance with sample collections and the Association of Bay Area Governments (ABAG) for funding. We also thank the anonymous reviewer for comments which greatly improved the manuscript. This work represents the UCD-ICP-MS contribution No. 0010 and Bodega Marine Laboratory contribution No. 2232.

References

Bennett, W. A. (2005). The population ecology of delta smelt in the San Francisco Estuary. *Francisco Estuary and Watershed Science*, in press.

Bennett, W. A., and Moyle, P. B. (1996). Where have all the fishes gone? Interactive factors producing fish declines in the Sacramento–San Joaquin Estuary. In 'San Francisco Bay: The Ecosystem'. (Ed. J. T. Hollibaugh.) pp. 519–542. (American Association for the Advancement of Science Pacific Division: San Francisco, CA.) Available online at http://www.sou.edu/AAASPD/TableContents/SFBayEco.pdf, verified June 2005.

Blum, J. D., Taliaferro, E. H., Weiss, M. T., and Holmes, R. T. (2000). Changes in Sr/Ca, Ba/Ca and $^{87}Sr/^{86}Sr$ ratios between trophic levels in two forest ecosystems in the northeastern USA. *Biogeochemistry* **49**, 87–101. doi:10.1023/A:1006390707989

Crecco, V. A., and Savoy, T. F. (1985). Effects of biotic and abiotic factors on growth and relative survival of young American Shad, *Alosa sapidissima*, in the Connecticut River. *Canadian Journal of Fisheries and Aquatic Sciences* **42**, 1640–1648.

Cushing, D. H., and Horwood, J. W. (1994). The growth and death of fish larvae. *Journal of Plankton Research* **16**, 291–300.

Dickin, A. P. (1995). 'Radiogenic Isotope Geology.' (Cambridge University Press: Cambridge.)

Dove, S. G., and Kingsford, M. J. (1998). Use of otoliths and eye lenses for measuring trace-metal incorporation in fishes: a bio-geographic study. *Marine Biology* **130**, 377–387. doi:10.1007/S002270050258

Farrell, J., and Campana, S. E. (1996). Regulation of calcium and strontium deposition on the otoliths of juvenile tilapia, (*Oreochromis niloticus*). *Comparative Biochemistry and Physiology* **115**, 103–109. doi:10.1016/0300-9629(96)00015-1

Fowler, A. J., Campana, S. E., Jones, C. M., and Thorrold, S. R. (1995). Experimental assessment of the effect of temperature and salinity on elemental composition of otoliths using laser ablation ICPMS. *Canadian Journal of Fisheries and Aquatic Sciences* **52**, 1421–1430.

Houde, E. D. (1989). Comparative growth, mortality, and energitics of marine fish larvae: temperature and implied latitudinal effects. *Fishery Bulletin* **87**, 471–495.

Ingram, B. L., and Sloan, D. (1992). Strontium isotopic composition of estuarine sediments as paleosalinity-paleoclimate indicator. *Science* **255**, 68–72.

Ingram, B. L., and Weber, P. K. (1999). Salmon origin in California's Sacramento-San Joaquin river system as determined by otolith strontium isotopic composition. *Geology* **27**, 851–854. doi:10.1130/0091-7613(1999)027<0851:SOICSS>2.3.CO;2

Kennedy, B. P., Folt, C. L., Blum, J. D., and Chamberlain, C. P. (1997). Natural isotope markers in salmon. *Nature* **387**, 766–767. doi:10.1038/42835

Kennedy, B. P., Blum, J. D., Folt, C. L., and Nislow, K. H. (2000). Using natural strontium isotopic signatures as fish markers: methodology and application. *Canadian Journal of Fisheries and Aquatic Sciences* **57**, 2280–2292. doi:10.1139/CJFAS-57-11-2280

Kennedy, B. P., Klaue, A., Blum, J. D., Folt, C. L., and Nislow, K. H. (2002). Reconstructing the lives of fish using Sr isotopes in otoliths. *Canadian Journal of Fisheries and Aquatic Sciences* **59**, 925–929. doi:10.1139/F02-070

Kuivila, K. M., and Moon, G. E. (2004). Potential exposure of larval and juvenile delta smelt to dissolved pesticides in the Sacramento–San Joaquin Delta, California. In 'Early life History of Fishes in the San Francisco Estuary and Watershed'. (Eds F. Feyrer, L. R. Brown, R. L. Brown and J. J. Orsi.) pp. 229–241. (American Fisheries Society: Bethesda, MD.)

Limburg, K. E. (1995). Otolith strontium traces environmental history of subyearling American shad *Alosa sapidissima. Marine Ecology Progress Series* **119**, 25–35.

Machado, N., and Gauthier, G. (1996). Determination of $^{207}Pb/^{206}Pb$ ages on zircon and monazite by laser ablation ICP-MS and applications to the study of sedimentary provenance and metamorphism in southeastern Brazil. *Geochimica and Cosmochemica Acta* **60**, 5063–5073. doi:10.1016/S0016-7037(96)00287-6

Outridge, P. M., Chenery, S. R., Babaluk, J. A., and Reist, J. D. (2002). Analysis of geological Sr isotope markers in fish otoliths with sub annual resolution using laser ablation-multicollector-ICP-mass spectrometry. *Environmental Geology* **42**, 891–899. doi:10.1007/S00254-002-0596-X

Simkiss, K. (1974). Calcium metabolism of fish in relation to ageing. In 'The Ageing of Fish'. (Ed. T. B. Begenal.) pp. 1–12. (Unwin Brothers: Surrey.)

Thompson, B., Hoenicke, R., Davis, J. A., and Gunther, A. (2000). An overview of contaminant-related issues identified by monitoring in San Francisco Bay. *Environmental Monitoring and Assessment* **64**, 409–419. doi:10.1023/A:1006459605924

Thorrold, S. R., Jones, C. M., Campana, S. E., McLaren, J. W., and Lam, J. W. H. (1998). Trace element signatures in otoliths record natal river of juvenile American shad (*Alosa sapidissima*). *Limnology and Oceanography* **43**, 1826–1835.

Thorrold, S. R., and Shuttleworth, S. (2000). *In situ* analysis of trace elements and isotope ratios in fish otoliths using laser ablation sector field inductively coupled plasma mass spectrometry. *Canadian Journal of Fisheries and Aquatic Sciences* **57**, 1232–1242. doi:10.1139/CJFAS-57-6-1232

Thorrold, S. R., Latkoczy, C., Swart, P. K., and Jones, C. M. (2001). Natal homing in a marine fish metapopulation. *Science* **291**, 297–299. doi:10.1126/SCIENCE.291.5502.297

Winer, B. J., Brown, D. R., and Michels, K. M. (1991). 'Statistical Principles on Experimental Design.' (McGraw-Hill: New York.)

Manuscript received 12 July 2004; revised and accepted 20 April 2005.