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Article in *Canadian Journal of Fisheries and Aquatic Sciences* · September 2013

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# Classification of hatchery and wild fish using natural geochemical signatures in otoliths, fin rays, and scales of an endangered catostomid

Brian A. Wolff, Brett M. Johnson, and Chad M. Landress

**Abstract:** Endangered and endemic June sucker (JS, *Chasmistes liorus*) have been stocked for many years to prevent extinction in Utah Lake, Utah. When unmarked fish appeared in the lake at higher rates than expected from tag loss, we sought to determine if  $^{87}\text{Sr}/^{86}\text{Sr}$  and  $\text{Sr}/\text{Ca}$  (signatures) in otoliths, fin rays, and scales could be used to identify whether stocking had created a naturally reproducing population. Signatures from otoliths and fin rays suggested that approximately 38% (12 of 31) of unmarked JS probably came from the Fisheries Experimental Station (FES) hatchery in Logan, Utah, and a minimum of 13% (four of 31) of unmarked JS had signatures that strongly indicated that they were of wild origin. The remaining JS (15 of 31) could not be assigned to any particular location because of signature overlap. While scales were not useful for determining fish provenance, we found a nearly 1:1 relationship between pelvic fin and otolith  $^{87}\text{Sr}/^{86}\text{Sr}$ . Thus, fin sections appear to provide a nonlethal structure for laser ablation microchemical analysis to determine origins of unknown origin JS in the future.

**Résumé :** L'empoisonnement du *Chasmistes liorus*, un poisson endémique en voie de disparition, se poursuit depuis de nombreuses années afin d'en prévenir la disparition dans le lac Utah (Utah, États-Unis). Quand des poissons non marqués ont commencé à apparaître dans ce lac à des fréquences plus grandes que ce que la perte d'étiquettes permet de prévoir, nous avons tenté de déterminer si les rapports  $^{87}\text{Sr}/^{86}\text{Sr}$  et  $\text{Sr}/\text{Ca}$  (signatures) dans les otolithes, les rayons de nageoire et les écailles pouvaient être utilisés pour établir si l'empoisonnement avait produit une population issue de la reproduction naturelle. Les signatures d'otolithes et de rayons semblent indiquer qu'environ 38 % (12 sur 31) des *C. liorus* non marqués proviennent probablement de l'écluse de la Fisheries Experimental Station (FES) à Logan (Utah) et au moins 13 % (quatre sur 31) des *C. liorus* non marqués ont des signatures indiquant fortement qu'ils seraient issus de la reproduction à l'état sauvage. Les autres *C. liorus* (15 sur 31) ne peuvent être affectés à un endroit précis en raison de la superposition de signatures. Si les écailles ne se sont pas avérées utiles pour déterminer la provenance des poissons, nous avons noté un rapport avoisinant 1:1 des valeurs de  $^{87}\text{Sr}/^{86}\text{Sr}$  des nageoires pelviennes et des otolithes. Les sections de nageoire semblent donc constituer une structure se prêtant à l'analyse microchimique non létale par ablation laser pour la détermination des origines de *C. liorus* d'origine inconnue. [Traduit par la Rédaction]

## Introduction

Distinguishing hatchery and wild origins of fishes is important for fisheries management and conservation questions such as evaluating effectiveness of stocking programs to supplement and maintain fisheries or to establish self-sustaining, naturally reproducing fish populations (Nickum et al. 2004). Because stocking is expensive, knowledge of the contribution of stocked fish to the population is paramount. Further, stocking can be ineffective or harmful when it occurs in the presence of wild fish (Hilborn 1999; Araki and Schmid 2010). In cases where the goal is to reestablish natural recruitment of a species, it is vital to determine whether unmarked fish are the result of natural reproduction or whether substantial loss of marks from hatchery fish is occurring. The repercussions of management decisions without this validation can be costly from both financial and ecological standpoints.

June sucker (JS, *Chasmistes liorus*) is an endemic and endangered fish in Utah Lake, Utah, USA (USFWS 1986). It is a relatively large (total length 600+ mm) and long-lived (40+ years) adfluvial lake sucker that makes annual spawning migrations from Utah Lake into tributaries of the lake (Scoppettone and Vinyard 1991; Belk

1998). Posthatch residence time (1–3 weeks) in tributaries consists of a quiescent period when JS larvae remain in the substrate and a drift period downstream into Utah Lake littoral zones (Modde and Muirhead 1994; Wilson and Thompson 2001; Ellsworth et al 2010).

JS populations began to decline in the early 1900s, mostly attributed to overexploitation by commercial anglers, introduction of non-native species, and hydrologic modifications including the dewatering, channelizing, and diverting of Utah Lake tributaries that resulted in changes in temperature, salinity, and turbidity (Whitney and Belk 2000). Substantial reductions in adult JS populations were documented through the mid-1900s via major declines in commercial catches, and by the 1970s, JS were rare or absent from both commercial catches and scientific monitoring efforts (Heckmann et al 1981; Radant and Sakaguchi 1981). Current threats are primarily interactions with non-native species and the lack of refuge resulting from habitat alterations that greatly reduced survival from the larval to juvenile stage (USFWS 1999).

As indicated by declines in commercial catches and adult JS aging studies, natural recruitment has been negligible since the mid-1900s, with natural recruitment too low to maintain a viable wild population (Radant 1986; Scoppettone 1988; Belk 1998). To

Received 23 February 2013. Accepted 1 August 2013.

Paper handled by Associate Editor Bronwyn Gillanders.

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prevent extinction, the June Sucker Recovery and Implementation Program (JSRIP) maintains JS in Utah Lake via stocking from a brood stock program at the Fisheries Experimental Station (FES), Logan, Utah, and additional refuges in northern Utah.

Most (77%) of the JS stocked into Utah Lake are reared exclusively at the FES facility in Logan, Utah. Other locations that rear JS before stocking into Utah Lake include Camp Creek and Red Butte reservoirs and Springville and Rosebud ponds. These locations serve as a backup program to supplement the FES brood stock program and as rearing facilities when FES is space-limited. These four additional locations receive JS from FES, although natural reproduction has been documented at Camp Creek and Red Butte reservoirs.

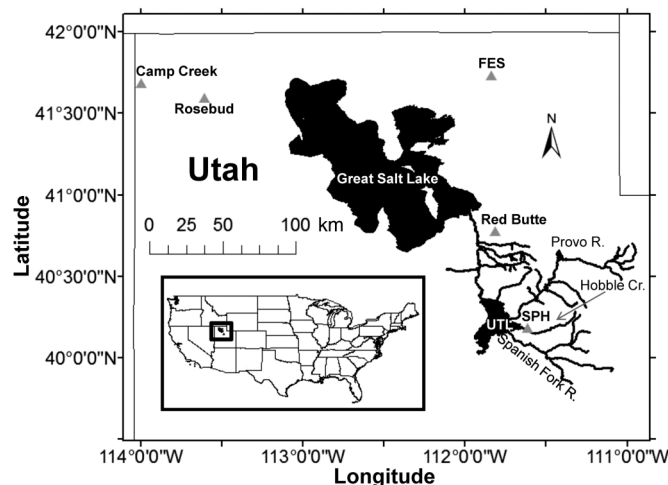
Before transfer to Utah Lake or other rearing locations, all JS are tagged following guidelines recommended by Guy et al. (1996). Prior to 2007, all JS (>150 mm) stocked into Utah Lake were injected in the abdominal musculature at the pelvic fin insertion point with passive integrated transponder (PIT) tags (12.0 mm × 2.1 mm, Biomark, Boise, Idaho). Smaller individuals received visible implant elastomer tags or a calcein mark. Beginning in 2007, because of large increases in FES production of JS and the cost limitations of PIT tags, all JS received a coded-wire tag (CWT; 2.2 mm × 0.25 mm), and some lots stocked into Utah Lake received PIT tags (e.g., Camp Creek and Red Butte). Differential CWT placement was as follows: FES (head), Rosebud (caudal peduncle), and Springville (dorsal musculature).

From 2007 to 2011, 22% of JS captured in Utah Lake during routine monitoring lacked a tag. While complete tag retention is unlikely, the observed tag loss rates were unexpectedly high. Determination of the origins of unmarked JS has important management implications. The JSRIP's primary goal is to "recover the June sucker to the extent that it no longer requires protection under the Endangered Species Act." Evidence of natural recruitment (i.e., if unmarked JS are of wild origin) would suggest a tremendous step toward recovery. However, if unmarked suckers were hatchery fish that lost their tags, then improved tag retention or an alternative method to assess wild versus hatchery contributions to the Utah Lake JS population would be needed.

Geochemical markers  $^{87}\text{Sr}/^{86}\text{Sr}$  and Sr/Ca (signatures) are among the most reliable natural tags for inferring origins of freshwater and anadromous fishes (e.g., Walther et al. 2011; Muhlfeld et al. 2012; Wolff et al. 2012). Water bodies can have unique  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios based on their underlying geology. Unique  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios are a result of radioactive beta-decay of  $^{87}\text{Rb}$ , which forms the stable, radiogenic isotope  $^{87}\text{Sr}$ . The isotope  $^{86}\text{Sr}$  remains constant, so water bodies influenced by older rocks or those containing more  $^{87}\text{Rb}$  (e.g., granites) will generally have higher  $^{87}\text{Sr}/^{86}\text{Sr}$  compared with younger rocks and (or) those with less inherent  $^{87}\text{Rb}$  (e.g., limestone; Faure and Powell 1972). Most chemical analyses in fish utilize otoliths, primarily because Sr readily substitutes for Ca in their calcium carbonate ( $\text{CaCO}_3$ ) matrix (Campana 1999). The incorporation of  $^{87}\text{Sr}/^{86}\text{Sr}$  from a fish's surrounding water into their otoliths provides a permanent record of fish origins and movements (provenance) throughout an individual's lifetime (Walther and Thorrold 2006; Barnett-Johnson et al. 2010; Hobbs et al. 2010), without fractionation by abiotic or biotic processes (Blum et al. 2000). In this paper, we used signatures in otoliths to assess the likelihood of natural reproduction of JS in Utah Lake.

In situations where sacrificing fish to obtain their otoliths is undesirable (e.g., endangered or rare fishes), it would be useful to have a nonlethal method to obtain provenance information comparable to that from otoliths. Others have used scales and fin ray chemistries (e.g., Sr/Ca, Ba/Ca, Mn/Ca, etc.) to infer fish provenance (Muhlfeld et al. 2005; Clarke et al. 2007; Smith and Whitley 2010). However, we have seen no studies that have successfully used natural  $^{87}\text{Sr}/^{86}\text{Sr}$  in fin rays or scales to determine origin or movements of fishes. Smith and Whitley (2011) examined Sr isotopes in fish fin rays; however, these authors used

**Fig. 1.** Map of the study area. The inset is the location of Utah Lake in relation to the rest of the USA. The triangles represent the locations the different June sucker rearing locations. FES, Fisheries Experimental Station; SPH, Springville ponds; and UTL, Utah Lake.



spiked  $^{88}\text{Sr}/^{86}\text{Sr}$  ratios to determine effectiveness of artificially marking lake sturgeon (*Acipenser fulvescens*). The secondary aim of this paper was to evaluate the utility of scales and fin rays as nonlethal substitutes for otoliths for  $^{87}\text{Sr}/^{86}\text{Sr}$  and Sr/Ca analysis in studies of fish provenance.

## Materials and methods

### Study area

Utah Lake is located in Utah Valley within the Great Basin, just west of the city of Provo, Utah (Fig. 1). It is one of the largest natural freshwater lakes in the western United States, with an elevation of 1368 m, surface area of 39 214 ha, and a shoreline spanning 122.3 km. Despite its large surface area, Utah Lake is very shallow, with mean depth of 2.8 m and a maximum depth of 4.2 m (Fuhrman et al. 1981). Utah Lake has well-mixed, turbid water during ice-off and a relatively short hydraulic residence time of around 2.5 years (Horn 2005). The Provo River is the largest river that flows into Utah Lake and has been the primary site of JS spawning (USFWS 1999). The lower 4.9 miles (1 mile = 1.609 km) of the Provo River is also designated critical habitat of JS (USFWS 1999). Other tributaries that are potential spawning areas for JS in Utah Lake include Hobbie Creek and the American Fork, Provo, and Spanish Fork rivers.

### Fish sampling

To determine whether unmarked JS were of hatchery or wild origin, we first needed to establish baseline signatures of Utah Lake and JS rearing locations. We examined JS from six different rearing histories (natal period to capture location): (1) Camp Creek to Rosebud; (2) FES to Red Butte; (3) FES to Utah Lake; (4) FES; (5) Springville; and (6) unmarked from Utah Lake (Table 1). Rearing histories 1–5 were marked fish whose history was known. We examined JS from these histories because they are all potential rearing areas before fish are ultimately stocked into Utah Lake. Unmarked JS could have come from any one of these locations, including Utah Lake (i.e., naturally reproduced; Fig. 2).

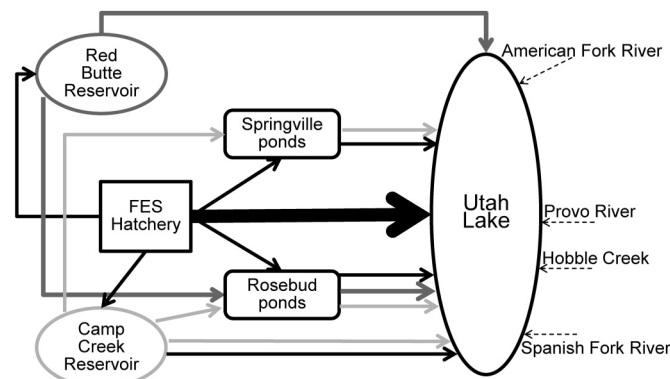
From 2004 to 2007, JS ( $n = 102$ ) were collected directly from a rearing facility (e.g., FES and Springville), from bycatch of the common carp (*Cyprinus carpio*) commercial seining fishery, or from routine monitoring efforts in Utah Lake, Camp Creek, and Red Butte by the Utah Division of Wildlife Resources (UDWR) biologists. Routine monitoring of Utah Lake included 24 h trap net sets from 22 standardized monitoring sites around the littoral zone

**Table 1.** June sucker sampling summary.

Rearing history	n							Mean			No. of ablations			Ablation zone coverage	
	2007	2008	2009	2010	2011	Unknown	Sum	TL	WM	age	Otoliths	Rays	Scales	Core	Edge
Camp Creek to Rosebud	—	—	—	—	26	—	26	285	270	6	20	18	0	Camp Creek	Rosebud
FES	—	—	—	—	13	—	13	87	13	1	13	12	12	FES	—
FES to Red Butte	—	—	3	—	5	—	8	324	331	4	7	7	3	FES	Red Butte
FES to Utah Lake	—	1	—	2	9	3	15	316	441	3	15	9	6	FES	Utah Lake
Springville	—	—	—	—	9	—	9	121	16	1	9	9	0	Springville	—
Unmarked from Utah Lake	—	—	2	1	26	2	31	435	1108	5	31	28	5	Unknown	Utah Lake
Sum	—	—	—	—	—	—	102	—	—	—	95	83	26	—	—
Mean	—	—	—	—	—	—	—	261	363	3	—	—	—	—	—

Note: TL, total length (mm); WM, wet mass (g). Mean ages are represented in years.

**Fig. 2.** Potential provenance of stocked and wild June sucker from Utah Lake. Different shading of lines represents various hatching locations. Heavy lines represent dominant sources of June sucker stocked into Utah Lake. Dashed lines represent potential natural recruitment sources.



or 1 h trammel net sets near the mouths of main tributaries where JS congregate before making their spawning run upstream.

Upon collection, fish were euthanized with a lethal dose (250 mg·L<sup>-1</sup>) of tricaine methanesulfonate (MS-222) and placed on ice until they could be frozen. Frozen fish were then sent to the Colorado State University Fisheries Ecology Lab, Fort Collins, Colorado, for extraction of hard parts and preparation for geochemical analysis.

### Fish structures preparation

We extracted otoliths (lapilli, preferred over sagittae in catostomids; [Sylvester and Berry 2006](#)), the leading pelvic fin ray ("rays"; clipped at the base closest to the body), and scales (removed below the dorsal fin and above lateral line) from each fish using nonmetallic forceps. After extraction, otoliths, rays, and scales were wiped with delicate task wipes to remove adhering tissue from the surface, sonicated in Milli-Q water for 5 min, and then air-dried. Otoliths and rays were then embedded in epoxy resin and cut transversely using an Isomet low-speed saw with diamond wafering blades to expose the inner core and annuli. Cut sections were then sanded to ~150 µm using 2000 grit sandpaper, polished with 3 µm lapping paper, rinsed with Milli-Q water, and air-dried. After polishing, fish ages were determined by counting otolith annuli under a compound light microscope. The age (years) of fish were then determined from the capture date minus the number of annuli. We also made a second assessment of age by counting ray annuli.

Following aging, otolith and ray sections were then prepared under a class-100 laminar flow hood for geochemical analysis. Otoliths and rays were placed onto 27 mm × 46 mm petrographic microslides using double-sided tape, rinsed with 3% ultrapure

nitric acid, sonicated in Milli-Q water for 5 min, and air-dried for 24 h. Scales were mounted dorsally (skin-side up) onto petrographic slides and cleaned again using the same method used for other structures described above. Petrographic sample slides were then placed in clean plastic petri dishes and taken to the Woods Hole Oceanographic Institution Plasma Mass Spectrometry Facility, Woods Hole, Massachusetts, for geochemical analysis.

### Laser ablation analysis

Geochemical analysis of otoliths, rays, and scales were performed using a Thermo Finnigan Neptune multicollector inductively coupled plasma mass spectrometer (MC-ICP-MS) coupled to a New Wave Research UP 193 nm excimer laser ablation system. The laser was configured to run at 80% intensity, 10 Hz pulse rate, 35 µm laser beam spot size, 5 µm per second laser scan speed, and 550 µm laser ablation distance within annuli. Using this method, ablated otolith material was carried from the laser cell to the MC-ICP-MS via helium gas, where it was then mixed in a spray chamber with argon gas and a wet aerosol, at which time a suite of isotopes (<sup>48</sup>Ca, <sup>83</sup>Kr, <sup>84</sup>Sr, <sup>85</sup>Rb, <sup>86</sup>Sr, <sup>87</sup>Sr, <sup>88</sup>Sr) were finally measured. This instrument method allowed for the direct quantification of <sup>87</sup>Sr/<sup>86</sup>Sr and Sr/Ca simultaneously. Since Sr/Ca is a ratio that includes all of the isotopes of both Sr and Ca, we applied the percentage of measured <sup>88</sup>Sr and <sup>48</sup>Ca from the sum of all isotopes in Sr and Ca, respectively, to derive Sr/Ca (mmol·mol<sup>-1</sup>). We adjusted for Kr and Rb interferences — the most important interferences in other studies using <sup>87</sup>Sr/<sup>86</sup>Sr laser ablation ICP-MS analyses ([Barnett-Johnson et al. 2005](#); [Woodhead et al. 2005](#); [Walther et al. 2008](#)) — following the techniques described by [Wolff et al. \(2012\)](#) as adopted from [Jackson and Hart \(2006\)](#) and [Walther et al. \(2008\)](#), respectively. To monitor instrument drift and external precision, standards were analyzed after every ten ablations. Standards used included National Institute of Standards and Technology standard reference material (SRM) 987 (<sup>87</sup>Sr/<sup>86</sup>Sr = 0.71024) and dissolved otolith certified reference materials CRM (<sup>87</sup>Sr/<sup>86</sup>Sr = 0.70918 and Sr/Ca = 2.78 mmol·mol<sup>-1</sup>; see [Yoshinaga et al. 2000](#)) and NRC (<sup>87</sup>Sr/<sup>86</sup>Sr = 0.70918 and Sr/Ca = 2.45 mmol·mol<sup>-1</sup>; see [Sturgeon et al. 2005](#)). All ablated samples and dissolved otolith standards were normalized to a daily mean SRM 987 using the following formula:

$$(1) \quad {}^{87}\text{Sr}/{}^{86}\text{Sr} \text{ normalized} = \frac{0.71024}{\text{SRM 987 measured}} \times {}^{87}\text{Sr}/{}^{86}\text{Sr} \text{ sample}$$

Normalized daily mean <sup>87</sup>Sr/<sup>86</sup>Sr (±1 SD) of CRM and NRC were 0.70919 ± 0.00002 (n = 8) and 0.70919 ± 0 (n = 2), respectively. Daily mean Sr/Ca of CRM and NRC were 3.01 ± 0.19 (n = 8) and 2.35 ± 0.01 (n = 2), respectively.

Otoliths, rays, and scales from JS were ablated within the inner-most annulus (core) and along the outermost edge (edge) to obtain signatures during the natal period and time nearest capture,



respectively. Core ablations were made just outside the primordium to minimize potential maternal influence on signatures (Macdonald et al. 2008). For instance, in JS rearing history 1 (see Fish sampling in Materials and methods), the core geochemical signature corresponded to Camp Creek and the edge geochemical signature corresponded to Rosebud. We knew that the core signature represented the prestocking location because they were not moved until after 1 year of age. Since we also knew the captured location, we were certain that the edge ablation represented the baseline signature from the water body from which they came. One important exception is unmarked JS; we could not be certain that the edge signatures from unmarked JS represented the signature from Utah Lake because we did not have any indication of when it entered Utah Lake. For this reason, we only used edge signatures from JS from rearing history 3 (FES to Utah Lake) to obtain a Utah Lake baseline signature. Baseline signatures of Utah Lake and each rearing area were then compared with core signatures of unknown origin JS (rearing history 6) to determine a likely place of origin.

Post hoc, we performed single transect ablations from the otolith core to edge on all unmarked JS that showed evidence of possible wild origin — transects were not run on unmarked JS that were classified to FES. We performed this analysis because some JS from FES (approximately 1 week old) were used in a penning study at Utah Lake in 2007 and 2008 to measure larval survivorship (Billman and Belk 2009), and it was thought some of these JS may have escaped from their enclosures and could therefore be mistaken for naturally reproduced fish. By ablating over the entire otolith core, we were able to detect the time spent in FES before being transplanted to Utah Lake, which we potentially would have been unable to detect using the core ablations as described above.

### Water sampling

To address whether unmarked JS could be of wild origin, we wanted to obtain baseline signatures of the major tributaries that flow into Utah Lake. We could not obtain JS signatures from these tributaries because larval fish structures were too small for ablation analysis. Therefore, we collected water samples from the Provo ( $n = 2$ ) and Spanish Fork ( $n = 2$ ) rivers, Hobble Creek ( $n = 2$ ), and the center of Utah Lake ( $n = 2$ ) on 14 June 2011 — we were unable to collect samples at the fourth main tributary, American Fork River, because of logistical constraints. Each river water sample was collected at the mouth of Utah Lake. Water samples were collected and filtered (0.45  $\mu\text{m}$ ) following the protocol described by Shiller (2003). We did not collect water samples from the artificial rearing areas because we already had fish samples for direct comparisons.

Filtered water samples (not acidified) were sent to the Woods Hole Oceanographic Institution Plasma Mass Spectrometry Facility, Woods Hole, Massachusetts, for  $^{87}\text{Sr}/^{86}\text{Sr}$  analysis using the same MC-ICP-MS as was used for all previously described ablation analyses. Strontium was separated and purified from the samples using Sr-Spec (Eichrom) resin in Bio-rad 1.2 mL columns. Samples were loaded in 1 mL 3.5 mol·L<sup>-1</sup> HNO<sub>3</sub>, and Sr was eluted with 4 mL deionized water. Separation from potential isobaric interferences of Ca and Rb was excellent (e.g., De Muynck et al. 2009). It is necessary to separate Ca from Sr before  $^{87}\text{Sr}/^{86}\text{Sr}$  is measured on the MC-ICP-MS because of potential isobaric interferences of Ca on Sr. For this reason, we could not get Sr/Ca from the water samples prepared for  $^{87}\text{Sr}/^{86}\text{Sr}$  analysis.

We then compared our tributary water  $^{87}\text{Sr}/^{86}\text{Sr}$  values with otolith and ray  $^{87}\text{Sr}/^{86}\text{Sr}$  values from all potential rearing areas. If the tributary values overlapped with other locations, then we assumed JS matching both a tributary and a rearing area could potentially be wild fish. This information was also useful when examining the laser transect geochemical profiles of all JS not assigned as likely FES origin. Since >99% of JS are stocked or

transferred to another rearing facility after 1 year of life, we assumed that an observed shift in  $^{87}\text{Sr}/^{86}\text{Sr}$  before the formation of the first annulus suggests possible wild origin.

### Statistical analysis

We used mixed regression analysis (PROC MIXED; SAS Institute Inc., Cary, North Carolina) for all statistical analyses. We used the full maximum likelihood estimation method for every mixed model. We addressed assumptions of normality and homogeneity of variance of signatures by examining diagnostic plots of conditional Studentized residuals. If diagnostic plots suggested non-normality or heterogeneity of variance, then Akaike's information criterion corrected for small sample size (AIC<sub>c</sub>) was used to determine the best transformations (e.g., log<sub>e</sub>, log<sub>10</sub>, or square root) necessary for the data. We re-examined conditional Studentized residual plots of the top-ranked model(s) after transformation to ensure assumptions of normality and homogeneity of variance were still met.

To evaluate otolith and ray  $^{87}\text{Sr}/^{86}\text{Sr}$  similarities, we used mixed regression of otolith  $^{87}\text{Sr}/^{86}\text{Sr}$  as the predictor variable (e.g.,  $x$  axis) and ray  $^{87}\text{Sr}/^{86}\text{Sr}$  as the response variable (e.g.,  $y$  axis). PROC MIXED was necessary because many JS had repeated measures, with two comparisons (core otolith versus core ray; edge otolith versus edge ray) per fish. Fish from a single rearing location were only ablated at the core, so they only had one comparison (otolith core versus ray core).

All our mixed models included one or more of the following fixed effects: (1) Intercept — fixed intercept (no other fixed effects); (2) Year — ablation corresponding to a given calendar year; (3) Location — ablation corresponding to location; and (4) Structure — otolith and ray. Additionally, we examined a model with interaction between ablation-location and ablation-year (Location  $\times$  Year) to determine if effect of year depended upon a given location, and vice versa. Our mixed models always included a random effect to accommodate repeated measures of individual fish (FishID) because most fish had both core and edge ablations.

We used variance components analysis (VCA) to determine the amount of variation explained by each of our fixed effects (Singer and Willet 2003; Breton et al. 2008; Wolff et al. 2012). We performed this analysis by measuring the amount of variance explained by each fixed effect (Location, Year, and Structure) as the proportional change in the total variance when each fixed effect was added to the Intercept model (model 1 from above) using the following equation:

$$(2) \quad \% \text{ Total variance reduced} = \frac{\text{Fixed effect variance} - \text{Intercept variance}}{\text{Intercept variance}}$$

We supplemented our statistics by ranking competing models (models 1–4) and all combinations of these effects using AIC<sub>c</sub>. As described by Burnham and Anderson (2002), relative support for each model  $i$  was calculated using delta AIC<sub>c</sub> ( $\Delta\text{AIC}_c$ ) and Akaike weights. In practice,  $\Delta\text{AIC}_c$  is the amount of information lost relative to the top-ranked model, where the top (or best) model has the lowest AIC<sub>c</sub> value (Burnham and Anderson 2002):

$$(3) \quad \Delta\text{AIC}_c = \text{given model AIC}_c - \text{top model AIC}_c$$

Generally, only models with  $\Delta\text{AIC}_c \leq 6$  are considered plausible (Burnham and Anderson 2002). Akaike weights provide relative support for each model by calculating model probabilities, which sum to one across the full model set (Burnham and Anderson 2002).

To compare signatures of JS from all rearing histories, we performed a VCA of each relevant fixed effect (Intercept, Location, Structure, and Year). In addition, we ranked all appropriate mixed

models and all their possible combinations using AIC<sub>c</sub> model selection. To obtain a baseline Utah Lake signature, we used edge values from JS from rearing history 3 (FES to Utah Lake). We used these JS because they were from the only rearing history that had a core signature distinctly different from Utah Lake. For instance, we could not use edge values from unmarked JS because they could have been recently stocked or moved from a tributary and therefore not reflective of the Utah Lake signature.

We examined temporal variation of all other water bodies by comparing JS from the six previously mentioned rearing histories. These fish had variable ages, which enabled us to compare signatures of the same water body at a different time (i.e., different aged fish had different core ablation-years). These age comparisons may be unbalanced, but it was the only way to compare multiple years for each location without collecting new samples from each location over multiple years. We were only able to obtain signatures from multiple years from Camp Creek, FES, Red Butte, and Utah Lake. Therefore, we could not test temporal stability for all locations (e.g., Rosebud and Springville).

A cross-validated quadratic discriminant function analysis (QDFA) was performed to determine the assignment reliability of JS signatures from all known locations of origin (PROC DISCRIM; SAS Institute Inc., Cary, North Carolina). The QDFA enabled us to examine locations with signature overlap that may potentially result in misclassification of unknown origin JS. Owing to uncertainty of when unmarked JS entered Utah Lake, we only used edge signatures from JS from rearing history 3 (FES to Utah Lake) to obtain a Utah Lake baseline signature. Since the FES signature was significantly different from all other locations, we were confident that edge values are reflecting the Utah Lake signature.

To assign the likely origin of unknown origin JS captured in Utah Lake, we used a bootstrapped maximum likelihood estimation (MLE) program HISEA (Millar 1990). We performed the bootstrapped (1000 iterations) MLE separately for otoliths and rays. The baseline signature of Utah Lake was established the same as described for the QDFA.

## Results

### <sup>87</sup>Sr/<sup>86</sup>Sr analysis

Diagnostic plots of conditional Studentized residuals revealed homogeneity of variance and no major departures from normality for <sup>87</sup>Sr/<sup>86</sup>Sr. Thus, no data transformations were performed on our <sup>87</sup>Sr/<sup>86</sup>Sr dataset.

For all JS comparisons, the VCA showed that the Location effect reduced the variance by 97%, a much greater reduction than Year (3%) and Structure (0%) (Table 2, part i). This shows that the most important effect on <sup>87</sup>Sr/<sup>86</sup>Sr was the location of origin among fish, and temporal and structure effects were relatively unimportant. We found similar results using AIC<sub>c</sub> (Table 3, part i), where Location was the top-ranked model, with 69% of the Akaike weight. The next best model included Location and Structure, with a ΔAIC<sub>c</sub> of 1.6 and 31% Akaike weight. However, since Structure showed a 0% reduction in the total variance from the Intercept model and it was the lowest ranked model by itself (ΔAIC<sub>c</sub> = 615.4), we feel that there may be some confounding of these two variables. The rest of the models had no support (ΔAIC<sub>c</sub> ≥ 16.8 and 0% Akaike weight, collectively).

Post hoc laser transect profiles of <sup>87</sup>Sr/<sup>86</sup>Sr (from the core to edge) of unmarked JS otoliths revealed that one JS whose core signature suggested Utah Lake origin (i.e., wild) was probably an escapee from the penning study at Utah Lake in 2007 and 2008 (Billman and Belk 2009), where the larval fish used were originally from FES. We also observed shifts in JS <sup>87</sup>Sr/<sup>86</sup>Sr between different (presumably) locations throughout the lifetimes for several fish (Fig. 3).

**Table 2.** Results from variance components analysis (VCA) of fixed effects.

	Percentage of variance		Total variance	Variance decrease (%)
Fixed effect	Fish ID	Residual		
<b>(i) <sup>87</sup>Sr/<sup>86</sup>Sr</b>				
Intercept	46	54	1.78×10 <sup>-5</sup>	—
Location	0	100	4.99×10 <sup>-7</sup>	97
Year	76	24	1.72×10 <sup>-5</sup>	3
Structure	46	54	1.78×10 <sup>-5</sup>	0
<b>(ii) Tranformed versus nontransformed Sr/Ca</b>				
Sr/Ca	21	79	2.87×10 <sup>-1</sup>	—
log <sub>10</sub> (Sr/Ca)	29	71	3.49×10 <sup>-2</sup>	88
SQRT(Sr/Ca)	25	75	5.07×10 <sup>-2</sup>	82
log <sub>e</sub> (Sr/Ca)	29	71	1.85×10 <sup>-1</sup>	36
<b>(iii) log<sub>10</sub>(Sr/Ca)</b>				
Intercept	29	71	3.49×10 <sup>-2</sup>	—
Location	42	58	1.92×10 <sup>-2</sup>	45
Year	58	42	3.03×10 <sup>-2</sup>	13
Structure	36	64	3.23×10 <sup>-2</sup>	7

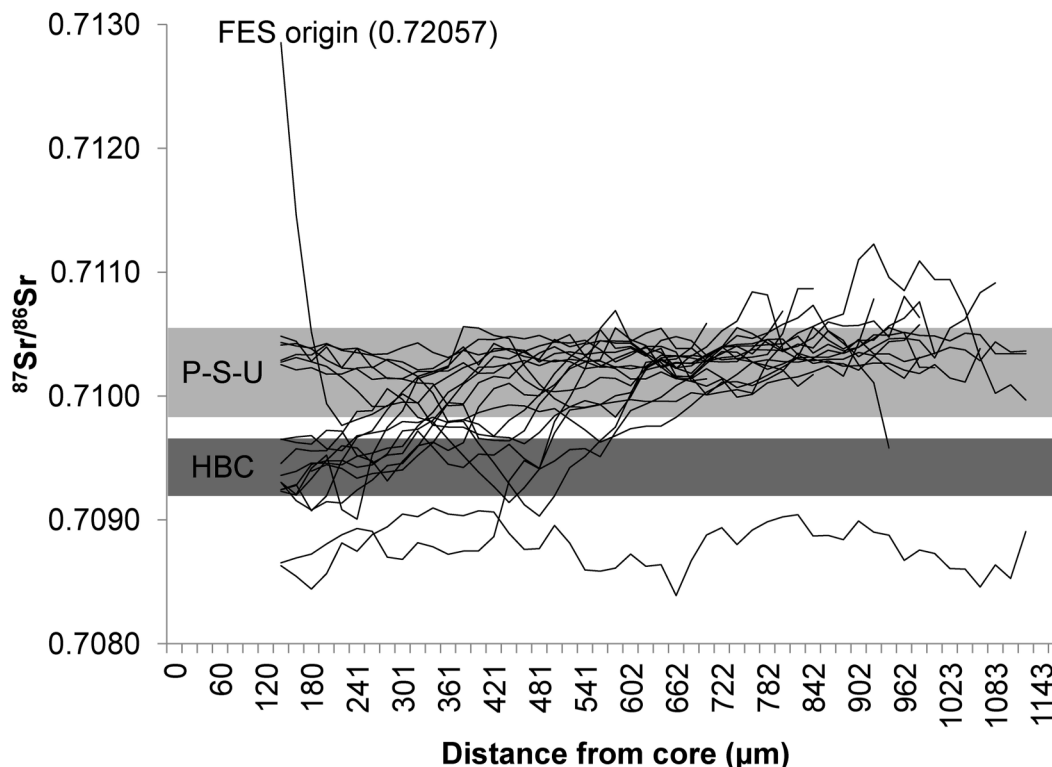
**Table 3.** Model selection results using Akaike's information criterion (AIC<sub>c</sub>).

Model	AIC <sub>c</sub>	ΔAIC <sub>c</sub>	Akaike weight
<b>(i) <sup>87</sup>Sr/<sup>86</sup>Sr</b>			
Location	-2 133.1	0.0	0.69
Location; Structure	-2 131.5	1.6	0.31
Location; Year	-2 116.3	16.8	0.00
Location; Structure; Year	-2 114.7	18.4	0.00
Location; Year; Location × Year	-2 100.3	32.8	0.00
Year	-1 583.5	549.6	0.00
Year; Structure	-1 581.6	551.5	0.00
Intercept	-1 519.8	613.3	0.00
Structure	-1 517.7	615.4	0.00
<b>(ii) Transformed versus nontransformed Sr/Ca</b>			
log <sub>10</sub> (Sr/Ca)	-104.0	0.0	1.0
SQRT(Sr/Ca)	-31.3	72.7	0.0
log <sub>e</sub> (Sr/Ca)	202.9	306.9	0.0
Sr/Ca	290.6	394.6	0.0
<b>(iii) log<sub>10</sub>(Sr/Ca)</b>			
Location; Structure	-264.7	0.0	1.0
Location; Structure; Year	-250.9	13.8	0.0
Location; Year; Location × Year	-219.2	45.5	0.0
Location	-216.0	48.7	0.0
Location; Year	-204.5	60.2	0.0
Year; Structure	-193.2	71.5	0.0
Year	-151.5	113.2	0.0
Structure	-123.2	141.5	0.0
Intercept	-104.0	160.7	0.0

### Sr/Ca analysis

Diagnostic plots of conditional Studentized residuals of Sr/Ca suggested non-normality and heterogeneity of variance. Therefore, VCA and AIC<sub>c</sub> model selection was used to determine if a log- (log<sub>e</sub> and log<sub>10</sub>) or square-root transformation was preferred (i.e., lower AIC<sub>c</sub>) over nontransformed Sr/Ca. The VCA showed that the log<sub>10</sub> transformation reduced the variance from the base (non-transformed) model by 88%, whereas the square-root- and log<sub>e</sub>-transformed model decreased the variance by 82% and 36%, respectively (Table 2, part ii). The log<sub>10</sub> transformation was also the top-ranked model (100% Akaike weight), and none of the other models (e.g., non-, square-root-, and log<sub>e</sub>-transformed) had any support (ΔAIC<sub>c</sub> ≥ 72.7; Table 3, part ii). Additionally, diagnostic

**Fig. 3.** Sr isotope profiles of otoliths of unmarked June sucker not classified to FES (Fisheries Experimental Station). To smooth the data, each line is an eight-point moving mean from each unmarked fish. The gray bars represent the mean ( $n = 2$  per tributary) water  $^{87}\text{Sr}/^{86}\text{Sr}$  signatures. P-S-U, Provo River, Spanish Fork River, and Utah Lake combined ( $n = 6$ ); HBC, Hobble Creek.



plots indicated assumptions of normality and homogeneity of variance were met for the  $\log_{10}$  transformation. Thus, the  $\log_{10}$  transformation was used for all succeeding analyses.

The VCA of Sr/Ca among JS showed that Location reduced the variance from the base (Intercept) model by 45%, compared with Year and Structure, which reduced the variance by 13% and 7%, respectively (Table 2, part iii). The top-ranked model included only Location and Structure effects (100% Akaike weight). There was no support for any other models, including those with a temporal component ( $\Delta\text{AIC}_c \geq 13.8$  and 0% Akaike weight; Table 3, part iii).

#### Concordance among structures

Analysis of scales suggested that they were unreliable structures for  $^{87}\text{Sr}/^{86}\text{Sr}$  laser ablation because the core region did not have enough Sr present to obtain a valid  $^{87}\text{Sr}/^{86}\text{Sr}$  measurement in most cases. Therefore, we excluded using scales from further analysis in this paper. However, we found a nearly 1:1 relationship between otolith and ray  $^{87}\text{Sr}/^{86}\text{Sr}$  using all otolith and pelvic  $^{87}\text{Sr}/^{86}\text{Sr}$  comparisons ( $y = 0.97x + 0.02$ ;  $R^2 = 0.94$ ;  $n = 92$ ; observations = 137). The relationship of Sr/Ca ( $\log_{10}$ -transformed) in otoliths and rays ( $y = 0.55x - 0.05$ ;  $R^2 = 0.40$ ;  $n = 92$ ; observations = 137) was less consistent than observed with  $^{87}\text{Sr}/^{86}\text{Sr}$ , where otolith Sr/Ca was generally higher than that of ray Sr/Ca. Both regression analyses were statistically significant ( $p < 0.0001$ ;  $\alpha = 0.05$ ).

#### Discrimination among water bodies

Overall reclassification accuracy ( $\pm 1$  SD) of fish back to their known ablation-location was correct  $73\% \pm 20\%$  for otoliths (Table 4, part i) and  $73\% \pm 40\%$  for rays (Table 4, part ii) using both geochemical markers. For otoliths, the classification accuracy ranged from 55% (Camp Creek) to 100% (FES). For rays, classification accuracy ranged from 0% (Red Butte) to 100% (FES, Rosebud, and Springville; Table 4, part ii). Although the SD of rays was greater than that of otoliths (driven by 100% Red Butte fish classified as Camp Creek origin), the overall discrimination among

groups was better with rays (i.e., less misclassification among different locations). Importantly, there were zero misclassifications of FES fish with JS from any other sources using both otolith and ray signatures.

#### Water $^{87}\text{Sr}/^{86}\text{Sr}$

Comparisons of water samples from Provo and Spanish Fork rivers, Hobble Creek, and the center of Utah Lake suggested that Provo and Spanish Fork rivers and Utah Lake had indistinguishable  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios; however,  $^{87}\text{Sr}/^{86}\text{Sr}$  from Hobble Creek was lower than from the other locations. Therefore, Provo and Spanish Fork rivers and Utah Lake (hereafter, P-S-U) were grouped together to facilitate comparisons with unmarked JS not classified to FES (i.e., possible tributary origin fish). We observed considerable overlap of Hobble Creek  $^{87}\text{Sr}/^{86}\text{Sr}$  with JS  $^{87}\text{Sr}/^{86}\text{Sr}$  from Red Butte and Camp Creek reservoirs and Springville ponds, whereas P-S-U  $^{87}\text{Sr}/^{86}\text{Sr}$  overlapped mostly with JS  $^{87}\text{Sr}/^{86}\text{Sr}$  from Utah Lake (Fig. 4). There was some overlap of P-S-U with Rosebud ponds and Camp Creek Reservoir, but most overlap was on the edges of both source clusters (Fig. 4).

#### Unknown origin June sucker

The MLE composition estimates of JS from known locations were generally accurate using both otoliths and rays. However, the overall difference between actual and MLE composition was much greater using otolith signatures (12.9%) than with ray signatures (1.4%; Table 5, parts i and ii, respectively). Of the 31 unmarked JS otolith signatures, the MLE assigned  $42.8\% \pm 4.2\%$  to Camp Creek,  $38.3\% \pm 3.3\%$  to FES,  $0.6\% \pm 0.0\%$  to Red Butte,  $0.1\% \pm 0.0\%$  to Rosebud,  $0.2\% \pm 0.0\%$  to Springville, and  $17.9\% \pm 1.3\%$  to Utah Lake (Table 5, part iii). The MLE using ray signatures of 28 unmarked JS (three JS did not have intact rays to examine) classified  $3.2\% \pm 0.1\%$  to Camp Creek,  $39.6\% \pm 3.7\%$  to FES,  $0\%$  to Red Butte,  $0\%$  to Rosebud,  $21.1\% \pm 1.7\%$  to Springville, and  $36.1\% \pm 3.3\%$  to Utah Lake

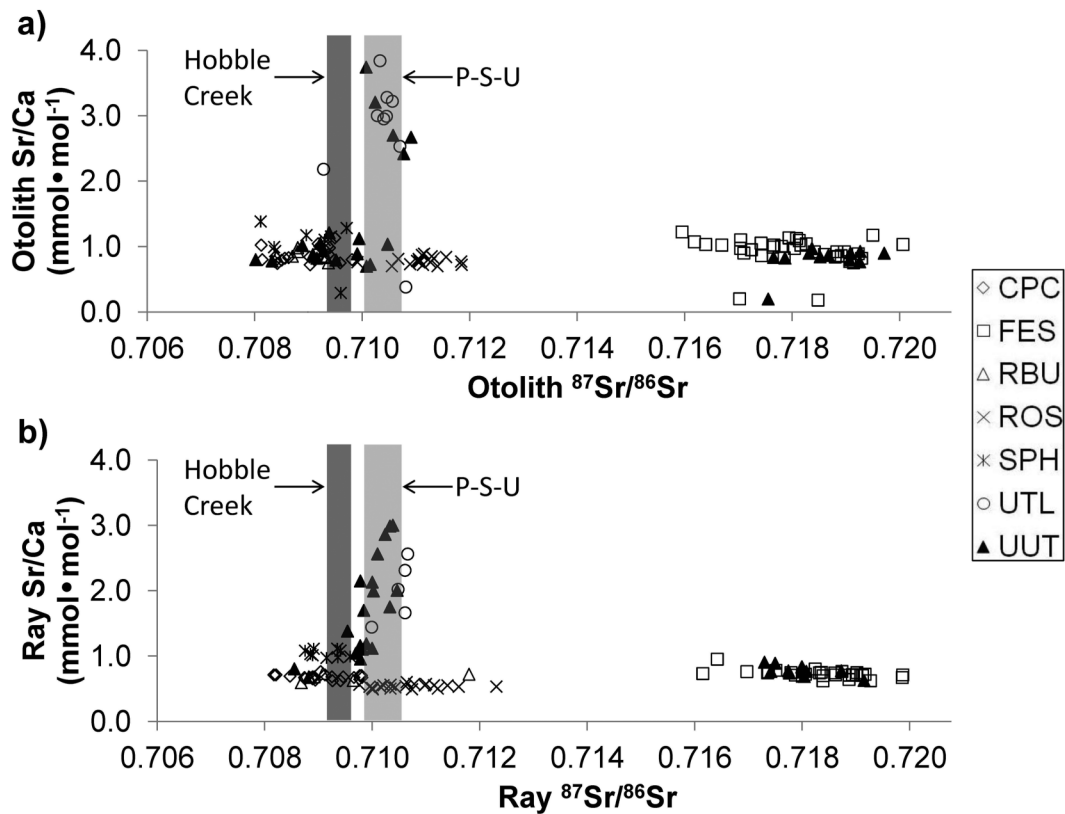


**Table 4.** Results from quadratic discriminant function analysis (QDFA) of June sucker otolith and fin ray  $^{87}\text{Sr}/^{86}\text{Sr}$  and  $\log_{10}\text{Sr}/\text{Ca}$  data from known locations.

	Assigned location						
Actual location	Camp Creek	FES	Red Butte	Rosebud	Springville	Utah Lake*	<i>n</i>
<b>(i) Otoliths (<i>n</i> = 98)</b>							
Camp Creek	55% <sup>†</sup>	—	40%	—	5%	—	20
FES	—	100% <sup>†</sup>	—	—	—	—	35
Red Butte	43%	—	57% <sup>†</sup>	—	—	—	7
Rosebud	—	—	6%	94% <sup>†</sup>	—	—	18
Springville	22%	—	22%	—	56% <sup>†</sup>	—	9
Utah Lake	—	—	—	—	22%	78% <sup>†</sup>	9
<b>(ii) Rays (<i>n</i> = 86)</b>							
Camp Creek	56% <sup>†</sup>	—	44%	—	—	—	18
FES	—	100% <sup>†</sup>	—	—	—	—	28
Red Butte	100%	—	0% <sup>†</sup>	—	—	—	6
Rosebud	—	—	—	100% <sup>†</sup>	—	—	20
Springville	—	—	—	—	100% <sup>†</sup>	—	9
Utah Lake	—	—	—	—	20%	80% <sup>†</sup>	5

\*Utah Lake signatures were established using edge ablations from June sucker transferred from FES only.  
†Correct classification to its known origin.

**Fig. 4.** Signatures of June sucker from all locations using otoliths (a) and fin rays (b). The gray bars represent the mean ( $n = 2$  per tributary) water  $^{87}\text{Sr}/^{86}\text{Sr}$  signatures (P-S-U, Provo River, Spanish Fork River, and Utah Lake combined;  $n = 6$ ). CPC, Camp Creek Reservoir; FES, Fisheries Experimental Station; RBU, Red Butte Reservoir; ROS, Rosebud ponds; SPH, Springville ponds; UTL, Utah Lake; UUT, unknown origin from Utah Lake.



(Table 5, part iii). Figure 4 shows the separation among groups with both geochemical markers.

**Discussion**

Our work suggests that fin rays appear to be reasonable surrogates for otoliths and may prove useful in future work as a non-lethal means to fingerprint fish origins. This was demonstrated by (i) a near 1:1 relationship between  $^{87}\text{Sr}/^{86}\text{Sr}$  of otoliths and rays; (ii) lower overall variation in  $\text{Sr}/\text{Ca}$  in rays than in otoliths; and

(iii) better discrimination among known locations from ray signature (i.e., greater accuracy in QDFA and the MLE analyses). The results from the MLE suggested a minimum of five and nine unmarked JS to Utah Lake origin using otoliths and rays, respectively. We also observed five unmarked JS whose signatures only overlapped with the Utah Lake signature and were not close to any other location's signature (Fig. 4). Otolith  $^{87}\text{Sr}/^{86}\text{Sr}$  transects of the 31 unmarked JS, however, suggest that one of the five previously mentioned fish was actually an escapee from a study con-



**Table 5.** Results from program HISEA bootstrapped maximum likelihood estimation (MLE) of June sucker origins.

	Assigned location					
	Camp Creek	FES	Red Butte	Rosebud	Springville	Utah Lake*
<b>(i) Otoliths (n = 98)</b>						
Actual composition	20.4%	35.7%	7.1%	18.4%	9.2%	9.2%
MLE composition	24.5%	35.7%	9.1%	18.8%	3.9%	8.1%
Absolute difference	4.1%	0.0%	2.0%	0.4%	5.3%	1.1%
Total difference = 12.9%						
<b>(ii) Rays (n = 86)</b>						
Actual composition	20.9%	32.6%	7.0%	23.3%	10.5%	5.8%
MLE composition	20.6%	32.6%	7.3%	23.4%	10.7%	5.5%
Absolute difference	0.4%	0.0%	0.3%	0.1%	0.2%	0.3%
Total difference = 1.4%						
<b>(iii) Unmarked</b>						
Otoliths (n = 31)	42.8%±4.2%	38.3%±3.3%	0.6%±0.0%	0.1%±0.0%	0.2%±0.0%	17.9%±1.3%
Rays (n = 28)	3.2%±0.1%	39.6%±3.7%	0%	0%	21.1%±1.7%	36.1%±3.3%

**Note:** Otolith and fin ray  $^{87}\text{Sr}/^{86}\text{Sr}$  and  $\log_{10}\text{Sr}/\text{Ca}$  data are from known locations. For Unmarked data (section iii), MLE origins ( $\pm 1$  SD) use otolith and ray signatures of unknown origin June sucker.

\*Utah Lake signatures were established using edge ablations from June sucker transferred from FES only.

ducted by Billman and Belk (2009), whose fish were of FES origin. Nonetheless, at least six JS whose profiles started within the P-S-U signature (Fig. 3). Signatures from both structures had very similar MLE estimates of the number of unmarked JS that were from FES (~12 of 31). However, the remaining fish could not be assigned to any one location because of substantial overlap of signatures from different sources. Our results also allowed us to exclude Red Butte Reservoir and Rosebud ponds as potential sources of the 31 unknown origin JS.

We assert that the discrepancy among otolith and ray signatures from the same fish probably resulted from temporal resolution divergence caused by structural size differences. The temporal resolution of otolith ablations is greater than that of rays because the area between the innermost core (i.e., primordium) and the first annulus were much larger in otoliths than with rays. Thus, otolith core ablations were incorporating material closer to the primordium (while staying relatively far from the first annulus), whereas the ray core ablations were incorporating more material between the inner core and the first annulus. Otolith core signatures may then be reflecting the source signature earlier in a fish's life (because the laser is integrating less material across the structure), and the ray core signatures may be incorporating material after a JS moved out from a tributary or was transplanted from a hatchery before entering Utah Lake. As a result, MLE results from ray core signatures may be overestimating the number of wild origin JS in this study — approximately 10 of 28 classified to Utah Lake with rays as opposed to approximately five of 31 with otoliths. Additionally, we observed several JS laser transect profile shifts from lower  $^{87}\text{Sr}/^{86}\text{Sr}$  values to  $^{87}\text{Sr}/^{86}\text{Sr}$  consistent with P-S-U (Fig. 3). Most of the shifts in  $^{87}\text{Sr}/^{86}\text{Sr}$  occurred prior to the formation of the first annulus — recall that artificially reared JS are stocked after 1 year of life — suggesting that some JS classified to Springville ponds or Camp Creek and Red Butte reservoirs could be fish that were spawned in the Hobbie Creek – Provo Bay area of Utah Lake. Owing to signature overlap among these locations, however, the possibility of a hatchery origin cannot be ruled out, and the movement patterns from laser transect profiles to infer a wild origin should be treated with caution.

Tag loss seems less likely to explain the origin of unknown JS classified to Camp Creek reservoir or Springville ponds, since all individuals transferred from these locations to Utah Lake were marked with a CWT or a PIT tag and checked for tag retention prior to stocking. Retention of CWT in JS from rearing areas has consistently been 85% or higher and should approach 100% after prestocking evaluations of tag retention (D. Routledge, FES Hatchery Superintendent, UDWR, unpublished data, 2012). Long-term

retention rates of intramuscular injected CWTs in other species (*Micropterus treculi*, *Centropomus undecimalis*, and *Lepisosteidae* spp.) exceeded 95% in all species, and all tag loss occurred within 30 days of tagging (Buckmeier 2001; Brennan et al 2005; Buckmeier and Reeves 2012). Retention of PIT tags (e.g., Red Butte and Camp Creek) should be higher than CWT rates based on comparative studies, with retention greater than 98% (Dare 2003; Buckmeier and Reeves 2012). PIT tag retention in juveniles (61–85 mm) of the closely related Lost River sucker (*Deltistes luxatus*) was 98% after 34 days (Burdick 2011). Retention of PIT tags improves with larger fish (Kaemingk et al 2011). Since JS are typically tagged when  $\geq 150$  mm TL, retention rates are expected to be comparable to or higher than the previously mentioned studies. Therefore, the likelihood of JS classified to outside rearing areas (except FES) actually originating from Utah Lake and its tributaries is increased by the expected negligible tag loss.

Since there are currently no published studies that have used  $^{87}\text{Sr}/^{86}\text{Sr}$  — or other Sr isotopes at natural abundance levels — in fin rays, it is difficult to assess if they are consistently correlated with otolith and water signatures. However, Clarke et al. (2007) reported that Sr concentrations in fin rays were more highly correlated with water than with otoliths. Smith and Whitley (2010) demonstrated that Sr/Ca ratios from fin rays could be used to distinguish smallmouth bass (*Micropterus dolomieu*) from different locations in northern Illinois rivers. Fin ray Sr/Ca ratios of age-0 *Scaphirhynchus* sturgeons in the Missouri and Mississippi rivers have also been demonstrated to correlate well with ambient water Sr/Ca (Phelps et al. 2012). Woodcock et al. (2013) also demonstrated that enriched, artificial stable isotopes derived from diets were retained well (86%) in fin rays of sub-adult red drum (*Sciaenops ocellatus*). Therefore, the use of fin rays to examine marks as a nonlethal method will likely continue substantially in the future.

Recall that we found consistently low Sr concentrations in JS scales, which resulted in unreliable  $^{87}\text{Sr}/^{86}\text{Sr}$  and Sr/Ca ratios. This finding makes it difficult to assess the merits of using scales as nonlethal structures for chemical analysis to reconstruct environmental histories of a fish. Clarke et al. (2007) found that Sr concentrations in Arctic grayling (*Thymallus arcticus*) scales were not well correlated with Sr concentrations of the ambient water. However, Wells et al. (2003) found that westslope cutthroat trout (*Oncorhynchus clarkii lewisi*) scale Sr/Ca did show a consistent relationship and water Sr/Ca. Additionally, Woodcock et al. (2013) showed that spiked isotope marks from scales of subadult red drum were retained 100% of the time throughout their study. Since the scales analyzed were from subadults, however, it remains uncertain as to whether those marks would be retained

throughout a fish's lifetime. Indeed, we found that older fish tended to have less Sr present in the core compared with younger fish, and Sr concentrations were higher near the edges of most fish (time closest to capture) than in the core. This further suggests that signatures in scales may not be permanently retained, as found with otoliths and presumably fin rays. Consequently, it remains unclear if scales may be appropriate nonlethal structures for all species, locations, and timescales.

The temporal variation in geochemical signatures will ultimately be determined by the geological uniqueness and the relative amount of water contributed from each tributary's watershed (Walther and Limburg 2012). Since two of the four main tributaries, Provo and Spanish Fork rivers, share similar geology (and therefore similar  $^{87}\text{Sr}/^{86}\text{Sr}$ ) and provide the majority of the water into Utah Lake, then we would not expect to observe large temporal variation on an annual basis. The VCA suggested negligible temporal effects, particularly compared with location effects. We also visually examined the temporal trends for each location, and we very rarely observed years in which the signature confidence intervals did not overlap. Moreover, we also observed very little fluctuation of the otolith signatures in walleye from 2007 to 2011 (B.A. Wolff, unpublished data, 2011). This is also in agreement with work performed in a relatively nearby watershed by Wolff et al. (2012) that found minimal temporal variation of  $^{87}\text{Sr}/^{86}\text{Sr}$  among several reservoirs over a decadal scale. Other studies utilizing  $^{87}\text{Sr}/^{86}\text{Sr}$  in otoliths have also reported low seasonal (Kennedy et al. 2000) and annual (Barnett-Johnson et al. 2008) temporal variation. Therefore, it is likely that our baseline  $^{87}\text{Sr}/^{86}\text{Sr}$  signatures could be used in future studies aimed at provenance of unknown origin JS. However, other studies have found significant temporal effects in  $^{87}\text{Sr}/^{86}\text{Sr}$ , particularly in lotic systems (Feyrer et al. 2007; Walther and Thorrold 2009; Walther et al. 2011). Sr/Ca ratios have been shown throughout the literature to be more subject to variation both intra- and inter-annually (Bacon et al. 2004; Schaffler and Winkelman 2008; Walther and Thorrold 2009), principally because otolith and ray Sr/Ca incorporation can be modified by temperatures and ambient availability (Thorrold et al. 1998; Elsdon and Gillanders 2003). Therefore, routine monitoring of baseline signatures should be made to ensure confidence in future classifications.

In summary, our study showed strong evidence of natural recruitment of JS in Utah Lake. This was demonstrated using both otoliths and rays, which is relevant to monitoring of JS populations in the future. The effectiveness of geochemical analysis of rays for determining provenance is a very important finding, providing a nonlethal method for monitoring possible recovery of the species over time. Although signatures of rays may have less temporal resolution than otoliths, rays can detect stocked fish if stocking occurs after formation of the first annulus. Future work needs to determine if clipping fin rays of live JS affects survival; however, this technique seems extremely promising for provenance studies.

## Acknowledgements

We thank the JSRIP for providing the funding for this project. Mark Capone (formerly USFWS), Mike Mills (JSRIP local coordinator), and Krissy Wilson (UDWR) provided considerable logistical support. We offer many thanks to the UDWR field technicians for providing fish samples. Jurek Blusztajn (Woods Hole Oceanographic Institution Plasma Mass Spectrometry Facility) provided support with the laser ablation mass spectrometry and performed the water  $^{87}\text{Sr}/^{86}\text{Sr}$  analysis. We are grateful for the advice provided by the anonymous reviewers that increased the quality of our manuscript.

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