

## MANAGEMENT BRIEF

# Forensic geochemistry identifies the illegal introduction of Walleye into Lake Cascade, Idaho

George Whitman<sup>1</sup>  | Jordan Messner<sup>2</sup> | Rachel C. Johnson<sup>1,3</sup>  |  
Malte Willmes<sup>4,5</sup>  | Brian P. Kennedy<sup>6</sup> | Carson Jeffres<sup>1</sup> 

<sup>1</sup>Center for Watershed Sciences,  
University of California Davis, Davis,  
California, USA

<sup>2</sup>Idaho Department of Fish and Game,  
Southwest Regional Office, McCall,  
Idaho, USA

<sup>3</sup>National Oceanic and Atmospheric  
Administration, National Marine  
Fisheries Service, Southwest Fisheries  
Science Center, Santa Cruz, California,  
USA

<sup>4</sup>Norwegian Institute for Nature  
Research, Trondheim, Norway

<sup>5</sup>The Ronin Institute for Independent  
Scholarship, Upper Montclair, New  
Jersey, USA

<sup>6</sup>Department of Fish & Wildlife  
Sciences, University of Idaho, Moscow,  
Idaho, USA

## Correspondence

George Whitman

Email: [gewhitman@ucdavis.edu](mailto:gewhitman@ucdavis.edu)

## Abstract

**Objective:** The illegal introduction of fish species can disrupt ecosystems, collapse food webs, and undermine recreational fishing opportunities. Determining whether introduced fish are locally reproducing is important for resource managers. Here, we used the geochemical analysis of otoliths to investigate the potential illegal introduction of a Walleye *Sander vitreus* caught in 2022, in Lake Cascade, Idaho. Lake Cascade is known for its recreational Yellow Perch *Perca flavescens* fishery and has no documented resident population of Walleye.

**Methods:** To determine if the Walleye was spawned in Lake Cascade or introduced from elsewhere, we analyzed otoliths for strontium isotopes ( $^{87}\text{Sr}/^{86}\text{Sr}$ ) and compared them to local water samples and three locally caught Yellow Perch.

**Result:** The Walleye otolith revealed a shift from higher  $^{87}\text{Sr}/^{86}\text{Sr}$  values (0.70878) in its early life to a period of intermediate value (0.70842) equal to Payette Lake water, then finally to a lower value (0.70807) comparable to the Lake Cascade water and resident Yellow Perch otoliths.

**Conclusion:** These results suggest that the Walleye was initially transplanted to the Payette Lake area from a currently unknown source in 2020, 2 years before its capture. It resided there briefly before migrating south into Lake Cascade. This study further highlights the benefit of geochemical analyses to identify the illegal introduction of fish and to provide resource managers with a powerful tool for early detection and prevention of the establishment of illegally introduced fish species.

## KEYWORDS

distribution, fisheries, nonindigenous species, otoliths

## INTRODUCTION

The intentional or accidental introduction and spread of invasive species threatens freshwater ecosystems and organisms worldwide (Sala et al. 2000; Jenkins 2003; Dudgeon et al. 2006; Leprieur et al. 2009). Consequently,

understanding the current state and pathways of invasions is crucial for management and conservation efforts. Throughout the previous approximately 150 years, state and federal agencies have intentionally introduced a variety of fish species to new waterways in the USA to increase recreational fishing opportunities (Nico and Fuller 1999;

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Nielsen 1999; Rahel 2004). However, the establishment of reproducing populations of introduced fish can cause drastic shifts in fish assemblage structures and food web dynamics (Pejchar and Mooney 2009; Bernery et al. 2022). This often leads to a decline of important fisheries, with consequent effects on the ecological and economic values that are associated with aquatic systems (McMahon and Bennett 1996; Haubrock et al. 2022).

In recognizing these adverse effects, there has been a shift in focus to maintaining current fish assemblages and limiting the institutional introduction of new species (Johnson et al. 2009). Illegal fish introductions are now a primary cause of nonnative fish spreading into novel waterways in the USA (Rahel 2004; Johnson et al. 2009). These introductions circumvent regulatory oversight and planning and can have catastrophic effects on ecosystems and undermine recreational angling opportunities. Furthermore, illegal introductions divert scarce agency resources that could be better used to manage and improve existing fisheries.

The rapid detection and evaluation of newly introduced species is crucial because reducing the densities of introduced species is difficult or impossible to reverse once a species becomes established (Vander Zanden and Olden 2008; Marr et al. 2010). Consequently, determining whether an introduced species is already reproducing locally or the result of continued introductions is crucial for resource managers.

The geochemical analysis of strontium isotopes ( $^{87}\text{Sr}/^{86}\text{Sr}$ ) in otoliths (ear stones) is a robust and well-established method for determining the provenance of invasive fishes (Campana 1999; Morissette and Whitley 2022). Otoliths are formed throughout the life of a fish by the incremental accretion of layers of calcium carbonate and protein with trace elements such as Sr substituting for Ca atoms in the  $\text{CaCO}_3$  crystalline matrix (Campana and Thorrold 2001; Doubleday et al. 2014). The value of the  $^{87}\text{Sr}/^{86}\text{Sr}$  ratio for a water body is determined by the chemical weathering of the watershed geology. Variation across watersheds is created by differences in the age and composition of the local geology based on the radioactive decay of  $^{87}\text{Rb}$  to  $^{87}\text{Sr}$  over geologic time. Therefore, watersheds that have older lithology or higher rubidium concentrations (e.g., granites) have higher  $^{87}\text{Sr}/^{86}\text{Sr}$  isotope ratios (Faure 1977). The  $^{87}\text{Sr}/^{86}\text{Sr}$  ratio in the otoliths is in equilibrium with the  $^{87}\text{Sr}/^{86}\text{Sr}$  ratio of the surrounding water, owing to negligible biological fractionation. Strontium isotopes have become an ideal marker for tracking provenance and fish movement among water bodies (Kennedy et al. 2000, 2002; Barnett-Johnson et al. 2008; Brennan et al. 2015). For example, Wolff et al. (2012) used  $^{87}\text{Sr}/^{86}\text{Sr}$  to investigate the origins of invasive piscivores in the upper Colorado river. Crook

### Impact statement

Otolith geochemical analyses reveal that an invasive Walleye caught in Lake Cascade, Idaho, did not originate there but was instead illegally introduced. This showcases the potential of this tool to provide independent evidence of illegal introductions.

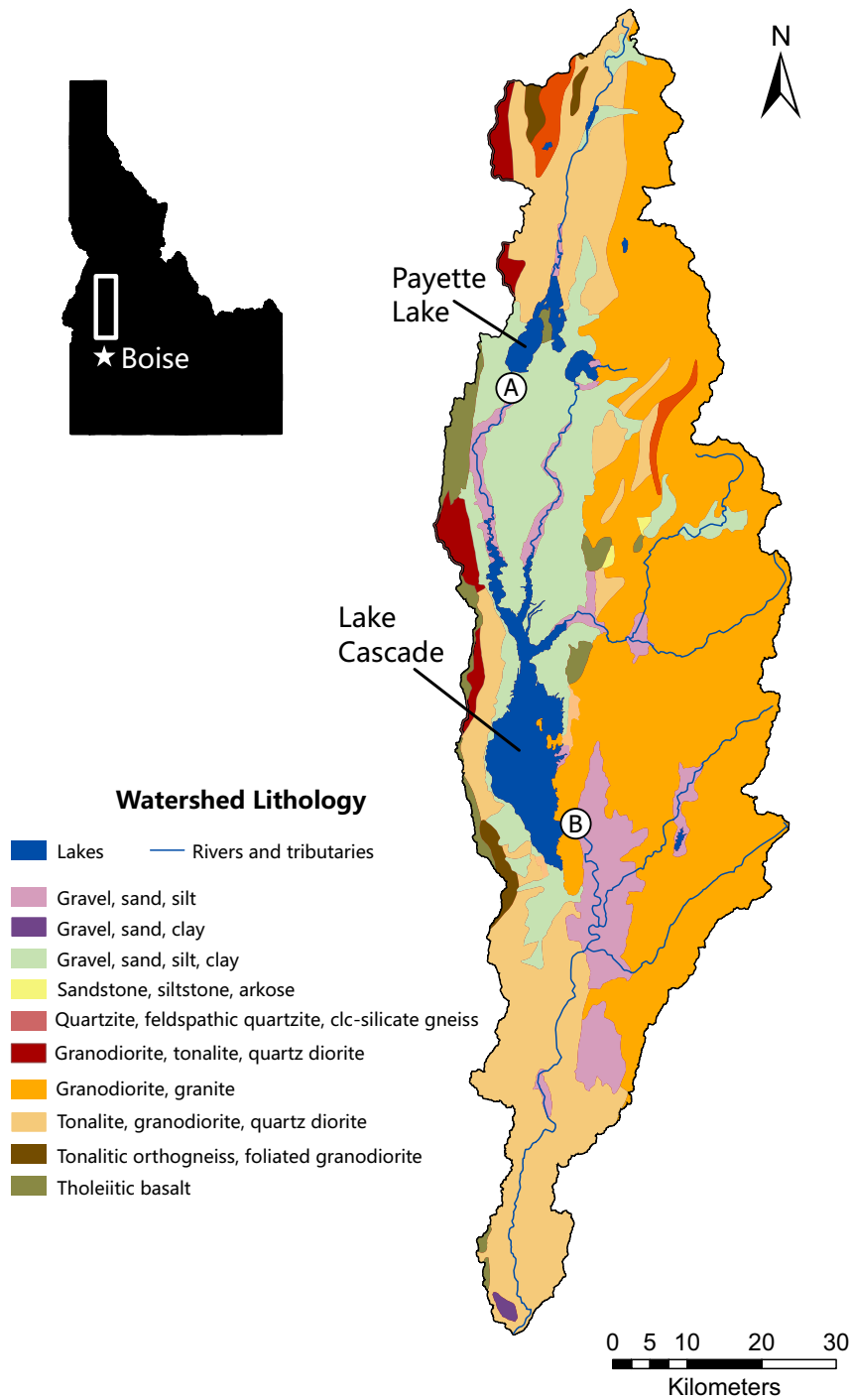
et al. (2013) employed  $^{87}\text{Sr}/^{86}\text{Sr}$  isotopes combined with trace elements to identify recruitment hot spots of nonnative carp in the Lachlan River, Australia. Bourret and Clancy (2018) tracked the timing and source of Walleye *Sander vitreus* introductions in Swan Lake, Montana.

Here, we used the geochemical analysis of otoliths to investigate the potential illegal introduction of a Walleye that was caught in 2022 in Lake Cascade, Idaho (Figure 1). Walleye are historically native to most of Canada and the northern USA (Colby et al. 1979; Hartman 2009). Their culinary and economic value make them popular among anglers (Hoslmann and Scott 2021). This popularity has led to their introduction into areas where they are not native, such as in the lakes and waterways of the intermountain western USA. In many of these locations where they become established outside of their native range, Walleye drastically alter the existing fisheries as prolific predators (Yule et al. 2000; Baldwin et al. 2003; Quist et al. 2005; Ryan et al. 2021). Lake Cascade is a reservoir that is known for providing a world-class recreational fishery for Yellow Perch *Perca flavescens*, which draws more than 250,000 h of angling effort each year (Idaho Fish and Game, unpublished data). The establishment of Walleye in this fishery, where Yellow Perch serve as the main forage base, could compromise the status of this world-class fishery. No Walleye had been found in Lake Cascade, Idaho, until 2018, when an angler reported catching a 483-mm Walleye. The fish was reportedly released back to the lake. A second individual was caught in May 2022, which is the subject of this study. These captures raised concerns of the presence of a resident, self-sustaining Walleye population, necessitating the need to evaluate whether the recently caught fish hatched locally or was an illegal transplant from another location.

## METHODS

### Fish collection

The Walleye specimen was captured via hook and line in Lake Cascade on May 7, 2022, and was brought to the



**FIGURE 1** Map showing Payette Lake, Lake Cascade, the major tributaries of Lake Cascade, and the geology of the surrounding watershed. The sites of water collection for the  $^{87}\text{Sr}/^{86}\text{Sr}$  analysis are labeled as follows: Payette Lake outflow (A) and Lake Cascade outflow (B). Sources: Reed S. Lewis, Paul K. Link, Loudon R. Stanford, and Sean P. Long: Idaho Geological Survey and Esri, National Atlas of the United States and the U.S. Geological Survey.

Idaho Department of Fish and Game office in McCall, Idaho, to determine total length (mm) and weight (g) and remove hard structures and tissue samples for analysis. Yellow Perch that are natal to Lake Cascade

were captured using sinking-style experimental gill nets (46×2 m; six panels of 19-, 25-, 32-, 38-, 51-, and 64-mm bar-measure mesh) set in Lake Cascade overnight in June 2022.

## Otolith preparation and aging

The sagittae otoliths were extracted from the Walleye and three Yellow Perch from Lake Cascade. The otoliths from the three locally reproduced Yellow Perch functioned as a proxy for the  $^{87}\text{Sr}/^{86}\text{Sr}$  ratio of species that are local to Lake Cascade over time to compare with the isotopic value of the Walleye otoliths. All the otoliths were given a double-sided polish using 600 and 1500 grit wet/dry sandpaper on the sagittal plane to expose the core and annuli. The otoliths were given a final fine polish with 3- $\mu\text{m}$  lapping film. The otoliths were then imaged using Image-Pro Premier at 100 $\times$  magnification and the annuli counted following established methods (Dembkowski et al. 2017; Long and Grabowski 2017).

## Otolith strontium isotope analysis

The  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios of the otoliths were measured at the University of California Davis Interdisciplinary Center for Plasma Mass Spectrometry using a Nd:YAG 213-nm laser (New Wave Research UP213) coupled to a Nu Plasma HR MC-ICP-MS (Nu032) following established methods (Barnett-Johnson et al. 2008). The otolith material was ablated with a transect of laser spots (40  $\mu\text{m}$  in diameter) from the core to edge at a laser pulse rate of 10 Hz. The data-reduction process included normalization for mass bias,  $^{87}\text{Rb}$  interference correction, and on-peak subtraction for  $^{86}\text{Kr}$ . The accuracy and precision of the instrument was evaluated using a marine White Seabass *Atractoscion nobilis* otolith as an in-house reference material. Replicate analyses for the White Seabass yielded an  $^{87}\text{Sr}/^{86}\text{Sr}$  value of  $0.70907 \pm 0.00009$  (mean  $\pm$  2SD;  $n=12$ ) in good agreement with the global average  $^{87}\text{Sr}/^{86}\text{Sr}$  value of modern seawater of 0.70918. To account for any instrument drift during the analysis, all the sample  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios were corrected based on the measured versus the known values of the White Seabass reference material (correction factor 1.000155). The individual spots displaying a transitional signature underwent a reexamination of their analysis profiles, and only the surface data were used if they exhibited significant isotopic changes in the Z direction.

The otoliths were reimaged after analysis, and the laser spots were compared with the annuli. The occurrence and location of potential shifts in the Walleye  $^{87}\text{Sr}/^{86}\text{Sr}$  profile were identified based on break-point analysis, where the model selection was performed comparing the Bayesian information criterion and residual sum of squares using the R package “strucchange” (Zeileis et al. 2002, 2003). All the data analysis was carried out using R 4.2.3 (R Core Team 2023).

## Water collection and Sr isotope analysis

To compare the Sr isotopic composition of the rearing water to that of the otolith samples, water was collected from the outlet of Lake Cascade as well as the outlet of Lake Payette (Figure 1), which is approximately 25 river kilometers upstream of Lake Cascade and provides between 15% and 30% of the inflow to Lake Cascade. Samples were taken from locations that presented thoroughly mixed lake water before ice-out in April 2023. The water samples were prepared for isotopic measurement using standard laboratory procedures (see Kennedy et al. 2000), and the isotope ratios were measured on an IsotopX Phoenix multicollector thermal ionization mass spectrometer (MC-TIMS) at the University of Idaho's Laboratory of Integrative Fish Ecology. Two samples of NIST – SRM 987 ( $^{87}\text{Sr}/^{86}\text{Sr}=0.710248$ ) analyzed concurrently with samples measured  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios of 0.710243 (SD=0.000019) and 0.710243 (SD=0.000020). A long-term analysis of NIST – SRM 987 over the lifetime of the instrument (2106–present,  $n=140$ ) reports an average value of 0.710244 (SD=0.000005).

## RESULTS

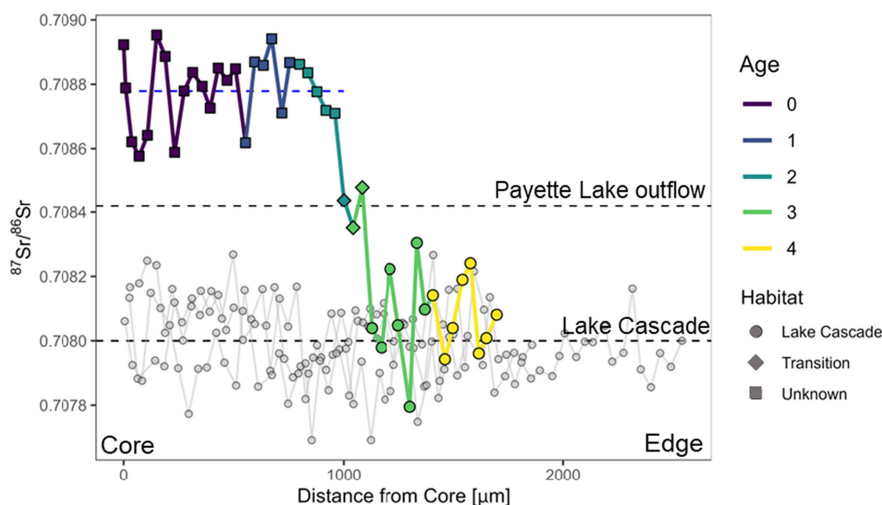
The Walleye was estimated to be age 4 (Figure 2), whereas the Yellow Perch varied from age 3 to age 5 (Table 1). The  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios of the three Yellow Perch (Table 1) showed no shifts in their lifetime profiles and similar values of  $0.70806 \pm 0.00021$  (mean  $\pm$  2SD;  $n=37$ ), of  $0.70803 \pm 0.00022$  ( $n=45$ ), and of  $0.70793 \pm 0.00023$  ( $n=65$ ). The water samples that were taken from Lake Cascade's largest input (Payette Lake's outflow) and largest output provided an  $^{87}\text{Sr}/^{86}\text{Sr}$  baseline across the local watershed and measured 0.70842 and 0.70800 respectively. The  $^{87}\text{Sr}/^{86}\text{Sr}$  values across the lifetimes of all three Yellow Perch overlapped with those of Lake Cascade (Table 1; Figure 3).



**FIGURE 2** A 100 $\times$  microscope image of the prepared Walleye otolith with laser spots and corresponding annuli labeled.

**TABLE 1** Summary of the  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios and ages of the Walleye; three locally caught Yellow Perch in Lake Cascade, Idaho; and water measurements of the outflows of Payette Lake and Lake Cascade.

Sample	Species	Habitat	Mean $^{87}\text{Sr}/^{86}\text{Sr}$	2 SD	Spot <i>n</i>	Age
Walleye	Walleye	Unknown	0.70878	0.00022	26	4
		Transition	0.70842	0.00013	3	
		Lake Cascade	0.70807	0.00027	15	
Yellow Perch 1	Yellow Perch	Lake Cascade	0.70806	0.00021	37	3
Yellow Perch 2	Yellow Perch	Lake Cascade	0.70803	0.00022	45	3
Yellow Perch 3	Yellow Perch	Lake Cascade	0.70793	0.00020	64	5
Payette Lake outflow	Water	Payette Lake	0.708422	0.00004		
Lake Cascade outflow	Water	Lake Cascade	0.708000	0.00003		



**FIGURE 3** The lifetime transect of  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios for the Walleye (colored by age) and three Yellow Perch (gray lines) caught in Lake Cascade, Idaho, in 2022. The measured  $^{87}\text{Sr}/^{86}\text{Sr}$  water values for Lake Cascade and the outflow of Payette Lake are shown as black dashed lines. The first 26 spots of the Walleye otolith show an elevated ratio (mean = blue line) before transitioning to lower values that are similar to those of the Yellow Perch and Lake Cascade water.

The break-point analysis indicated that the Walleye  $^{87}\text{Sr}/^{86}\text{Sr}$  profile was characterized by a single shift, splitting the lifetime profile into two distinct parts (Figure 3). The early life had a higher value of  $0.70878 \pm 0.00022$  ( $n$  spots = 26), and the later period had a lower value of  $0.70807 \pm 0.00027$  ( $n$  spots = 15). At the boundary is a narrow transitional zone with a  $^{87}\text{Sr}/^{86}\text{Sr}$  ratio of  $0.70842 \pm 0.00013$  ( $n$  spots = 3). Transitional spots were not included in the calculation of either early or late life mean  $^{87}\text{Sr}/^{86}\text{Sr}$ . Compared with the mean  $^{87}\text{Sr}/^{86}\text{Sr}$  value of the Yellow Perch samples from Lake Cascade, the Walleye early life  $^{87}\text{Sr}/^{86}\text{Sr}$  values were significantly different ( $t = 36.32$ ,  $df = 25$ ,  $p < 0.0001$ ), whereas the later period  $^{87}\text{Sr}/^{86}\text{Sr}$  values were similar to the mean for Lake Cascade ( $t = 1.92$ ,  $df = 14$ ,  $p = 0.074$ ). This transition in the Walleye from high to low  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios started between the second and third annuli.

## DISCUSSION

In Idaho, great effort has gone into preventing Walleye from spreading into new watersheds because once they are initially introduced, their population can rapidly increase and become difficult if not impossible to eradicate (Ryan et al. 2021). Across the western United States, there are many examples of prized sport fisheries being disrupted by illegal Walleye introductions (e.g., McMahon and Bennett 1996; Baldwin et al. 2003; Johnson et al. 2017). Lake Cascade currently supports a world-renowned Yellow Perch fishery. Walleye have the potential to reduce the value of this nationally recognized fishery by decreasing the abundance of Yellow Perch through competition and predation (Pierce et al. 2006). Consequently, the discovery of even a single Walleye is great cause for concern to fisheries managers.

In this study, we used otolith geochemistry to investigate the potential illegal introduction of a Walleye that was caught in Lake Cascade, Idaho, in 2022. The  $^{87}\text{Sr}/^{86}\text{Sr}$  values for water from the Lake Cascade outflow are consistent with our expectations based on the values for resident fish and can be considered representative of the entire lake, as Lake Cascade is a relatively shallow and well-mixed reservoir. There are also no known examples in the literature of heterogeneity in  $^{87}\text{Sr}/^{86}\text{Sr}$  in large lake systems, even when the source water from different tributaries varies in  $^{87}\text{Sr}/^{86}\text{Sr}$  (Feyrer et al. 2019).

The otoliths from locally caught Yellow Perch supplemented the water data and tracked any possible  $^{87}\text{Sr}/^{86}\text{Sr}$  changes over time. The analysis of Lake Cascade's inputs and outflows supports variation in Sr isotope ratio data for resident perch between 0.7082 and 0.7078, suggesting that the tributary inputs from the north are slightly more radiogenic (higher  $^{87}\text{Sr}/^{86}\text{Sr}$ ) but that other inputs (groundwater or unmeasured tributaries) provide lower Sr isotope values, with a mean lake value approximating 0.7080. A lack of significant shifts in their lifetime  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios indicate that these fish spent their entire life in Lake Cascade.

In the Walleye, we identified a shift that began between the second and third annuli from higher to lower  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios, which indicated a change in watershed, consistent with being transplanted. The lower  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios of the Walleye matched those of the Lake Cascade water and the locally caught Yellow Perch, suggesting that it arrived in Lake Cascade in 2020, 2 years before its capture. The 120- $\mu\text{m}$  span of transitional material at the boundary is larger than a quick trip between water bodies would suggest. The average  $^{87}\text{Sr}/^{86}\text{Sr}$  value of this area is 0.70842, identical to the water value of Payette Lake's outflow. The most likely explanation for the presence of the Walleye in Lake Cascade is that it was originally stocked in the Payette Lake area, resided there briefly, and then migrated into Lake Cascade. The lack of a comprehensive Sr isoscape of Idaho currently prohibits determining the Walleye's exact original source.

The possible source regions for the illegal introduction of a Walleye into Lake Cascade span a diverse geologic landscape with large regions of isotopic similarity (e.g. Columbia River basalts) interspersed by smaller regions of tremendous geologic heterogeneity. The geologic diversity of potential natal habitats provides the opportunity to identify sources of illegal introductions when stocked fish are captured and sampled. Lake Cascade geographically straddles a well-established suture zone between the island arc terranes that remained following subduction under the old North American craton to its east, a region that is now characterized as the western edge of ancient Laurentia (Armstrong et al. 1977; Kistler and

Peterman 1973). The suture has resulted in the identification of an isotopic transition from west to east named "the 0.706 isopleth" (Armstrong et al. 1977; Gaschnig et al. 2011). Large regions to the west of Lake Cascade have lower Sr isotopic values, whereas rivers draining the eastern watersheds of the Snake and Salmon rivers can have either very radiogenic (higher) Sr isotopic values that reflect billion-year-old mountain building or intermediate values that reflect either the Columbia River Basalt or the Idaho batholith (0.7083–0.7090; Hegg et al. 2013; B. P. Kennedy, unpublished data). Consequently, there are several reservoirs that sit along the southern Snake River plain that represent isotopically plausible sources for this introduction.

This study highlights the potential of geochemical analyses to identify the illegal introduction of fish and provides resource managers with a powerful tool for early detection to prevent the establishment of nonnative species. Our results suggest that this Walleye was illegally transplanted to the Payette Lake area before migrating to Lake Cascade. Targeted monitoring for Walleye in 2022 did not find any further indications of the species in the system. Additionally, biologists with the Idaho Department of Fish and Game and the Idaho Cooperative Fish and Wildlife Research Unit set more than 10 km of gill nets over a 12-month period in 2022 and 2023 and did not find any other Walleyes. Additionally, no other Walleye catches have been reported. This is encouraging, as it suggests that the current abundance of Walleye in Lake Cascade is low to absent. Consequently, management to prevent illegal stocking is likely the best management response.

## ACKNOWLEDGMENTS

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## CONFLICT OF INTEREST STATEMENT

There is no conflict of interest declared in this article.

## DATA AVAILABILITY STATEMENT

All data are openly available in a public repository (<https://github.com/MalteWillmes/WalleyeForensics>) and upon reasonable request from the authors.

## ETHICS STATEMENT

This study adheres to the principles and practices of the “Guidelines for the Use of Fishes in Research” published by the American Fisheries Society.

## ORCID

George Whitman  <https://orcid.org/0000-0002-0701-0282>

Rachel C. Johnson  <https://orcid.org/0000-0002-0278-7826>

Malte Willmes  <https://orcid.org/0000-0001-8734-5324>

Carson Jeffres  <https://orcid.org/0000-0001-6532-6851>

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