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RESEARCH

Contemporary evolution of neutral genetic structure in introduced Klamath smallscale suckers (*Catostomus rimiculus*) recapitulates native patterns

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Abstract
We investigated the riverscape genetic structure of Klamath smallscale suckers (*Catostomus rimiculus*) in the Smith River, California, a population believed to have been introduced and founded by approximately six individuals within the last 100 years. Analyzing 15 microsatellites from larval samples across the species' entire known Smith River range, we discerned: (1) increased genetic diversity downstream, (2) increased within-site differentiation upstream, and (3) isolation by river distance. These introduced suckers displayed a genetic profile expected for native species in river networks, indicating post-introduction contemporary evolution of neutral genetic structure unconstrained by a small founding population size. Application of available models of processes generating genetic variation in river networks suggested that upstream colonization generated the patterns we observed. The Smith River population of Klamath smallscale suckers may be in genetic equilibrium or may be continuing to expand since a likely introduction between the 1890s and 1970s.

Keywords Riverscape genetics · Genetic diversity · Fish · Catostomidae · Klamath smallscale sucker · *Catostomus rimiculus* · Introduced species · Evolution

Introduction

Globally, introduced species pose significant challenges, affecting biodiversity, ecosystem services, and economic structures (Clavero and García-Berthou 2005; Sax et al. 2007). Oftentimes, introduced species undergo founder events that reduce genetic diversity, creating the potential for adverse population-level consequences (Dlugosch and Parker 2008). These events may increase homozygosity and the risk of inbreeding depression, and create greater vulnerability to random genetic fluctuations, all of which threaten population sustainability (Frankham 2005). Interestingly, contrary to these expectations, many populations of introduced species have been established and have expanded their geographic range despite evidence that they possess limited genetic diversity (Dlugosch and Parker 2008).

Analyzing neutral genetic variation can provide key insights into the status of introduced populations. The gradual development of neutral patterns, as expected under drift-migration equilibrium, can indicate changes in genetic structuring over time without necessarily implying adaptive evolution (Slatkin 1993; Excoffier and Ray 2008). Additionally, specific genetic patterns, such as a decrease in genetic diversity along routes of colonization, can suggest whether an introduced species is in a phase of range expansion or has reached an equilibrium in its new territory (Estoup et al. 2004; Paz-Vinas et al. 2015).

The propensity of introduced populations to evolve neutral genetic structuring patterns similar to those in the native range is not well understood. However, studies of post-introduction contemporary evolution of heritable phenotypes have shown that introduced species may recapitulate evolutionary patterns observed within native habitats (Gilchrist et al. 2008; Alexander et al. 2009). Post-introduction evolution suggests the absence of genetic constraints hindering introduced species (Bock et al. 2015). Within introduced populations, evolutionary change can occur rapidly, with

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numerous examples documenting changes within 50 years (Whitney and Gabler 2008).

We studied Klamath smallscale suckers (*Catostomus rimiculus*) that are believed to have been introduced to the Smith River, California, likely between the 1890s and 1970s (Kinziger et al. 2021). The introduced population shows a 75% loss in allelic richness and a 30% drop in heterozygosity compared its native range. These losses in genetic diversity are consistent with founder effects, with analyses suggesting a single introduction of six or possibly fewer effective individuals (Kinziger et al. 2021). In spite of such a constrained starting point, these suckers now occupy much of the Smith River drainage.

In this study we describe within-basin patterns of genetic diversity and structure of the introduced Smith River population of Klamath smallscale suckers by evaluating variation in 15 microsatellites from larval samples collected throughout the species' current known distribution in the drainage. We specifically evaluate our data for evidence of downstream increases in genetic diversity, a prevalent pattern observed in native species, including catostomids (Douglas et al. 2003; Dowling et. al. 2012; Paz-Vinas et al. 2015; Thomaz et al. 2016). We also explore the mechanisms that can produce downstream increases in genetic diversity, including downstream-biased dispersal, increased habitat availability downstream, upstream-directed colonization, and their possible interactions (Paz-Vinas et al. 2015). Our study incorporates two sets of samples collected 4 years apart, allowing us to gauge the temporal resilience in patterns of genetic diversity and differentiation.

Materials and methods

Sample collection

We collected and analyzed 933 Klamath smallscale sucker larvae from the Smith River, California. The sampling effort sought to cover the entire distribution of the species in the drainage. It included 16 sites, encompassing all major tributaries (North, South, and Middle forks) and downstream areas (Fig. 1; Table 1). The pairwise distance between sites averaged 22 river km, with a maximum of 58 river km. To evaluate whether genetic patterns were stable through time, collections were made at 14 sites in 2013 and 6 sites in 2017, with 4 of these sites sampled in both 2013 and 2017.

We collected larvae because adults are rarely encountered in the Smith River. This difficulty is due to the fact that adults commonly occupy deep pools (> 10 m) with substantial habitat complexity, and frequently exhibit concealment

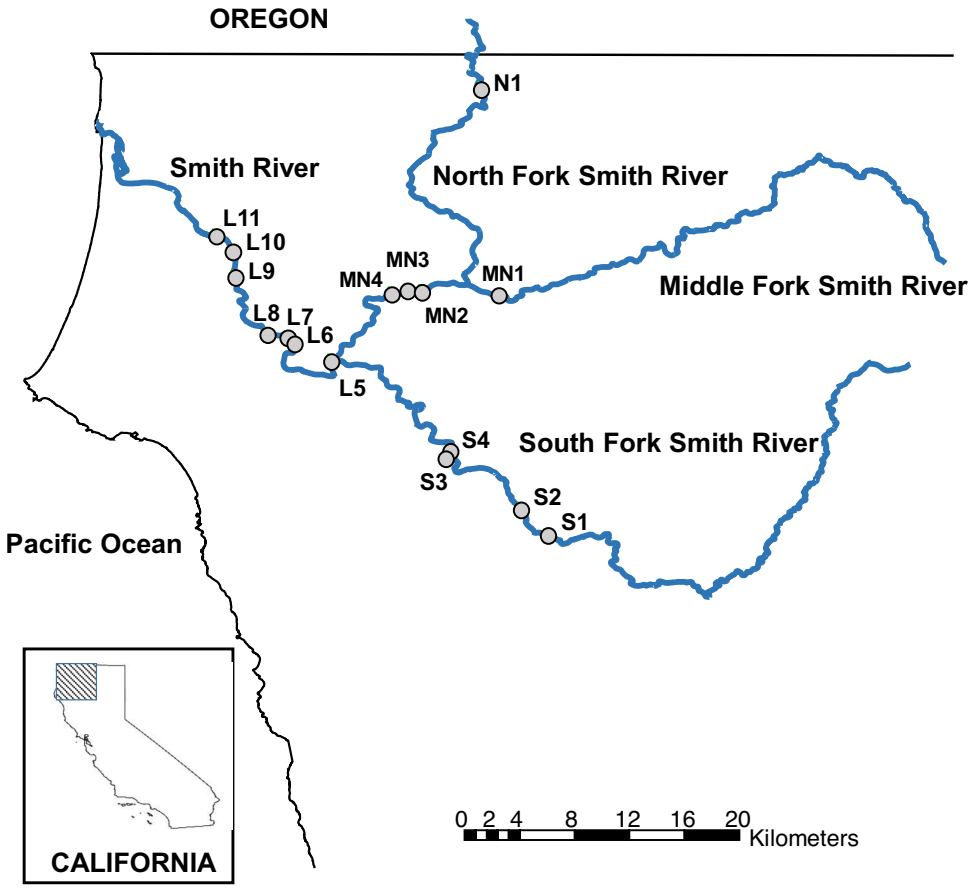

Fig. 1 Distribution of the 16 collection locations for larval Klamath smallscale suckers (*Catostomus rimiculus*) in the Smith River, California. Site labels identify their positions in the stream network. N = North Fork, M = Middle Fork, MN = below the confluence of the Middle and North Forks, S = South Fork, L = Lower river

Table 1 Site, collection year, sample size (N), observed heterozygosity (H_O), expected heterozygosity (H_E), observed allelic richness (A), standardized allelic richness (A_R) using rarefaction, within-site F_{ST}, and distance upstream from the river mouth for collections of Klamath smallscale suckers from the Smith River, California

Site	Year	N	H_O	±SE	H_E	±SE	A	±SE	Ar	±SE	Within-Site F_{ST}	Distance upstream (river km)
N1	2013	77	0.55	0.02	0.53	0.06	4.13	0.14	3.320	0.200	0.052	62.53
N1	2017	20	0.6	0.04	0.6	0.13	3.87	0.22	3.550	0.230	0.044	62.53
MN2	2013	37	0.61	0.02	0.61	0.10	4.73	0.26	3.680	0.240	0.024	38.93
MN3	2013	96	0.61	0.01	0.61	0.06	5.33	0.24	3.580	0.230	0.016	38.53
MN4	2013	66	0.61	0.02	0.61	0.08	4.87	0.23	3.650	0.260	0.018	37.13
L5	2013	55	0.63	0.02	0.62	0.08	4.40	0.22	3.600	0.220	0.015	29.23
L6	2013	54	0.62	0.02	0.62	0.08	4.67	0.24	3.610	0.230	0.016	22.63
L7	2013	79	0.63	0.01	0.61	0.07	4.93	0.17	3.600	0.210	0.012	21.83
L7	2017	24	0.63	0.03	0.63	0.13	4.00	0.26	3.580	0.240	0.012	21.83
L8	2013	96	0.64	0.01	0.64	0.07	4.93	0.17	3.640	0.230	0.016	20.63
L9	2013	16	0.61	0.04	0.65	0.16	4.07	0.34	3.760	0.270	0.012	14.93
L9	2017	37	0.66	0.02	0.65	0.11	4.40	0.29	3.680	0.270	0.022	14.93
L10	2013	53	0.65	0.02	0.64	0.09	5.00	0.29	3.720	0.240	0.024	12.93
L11	2013	20	0.64	0.04	0.64	0.14	4.07	0.30	3.660	0.250	0.019	11.73
S4	2013	23	0.57	0.04	0.54	0.11	3.73	0.18	3.300	0.170	0.033	42.13
S3	2013	32	0.54	0.03	0.57	0.10	3.80	0.20	3.340	0.190	0.030	42.43
S2	2013	42	0.57	0.02	0.58	0.09	3.93	0.20	3.410	0.210	0.032	49.43
S2	2017	39	0.57	0.02	0.56	0.09	4.27	0.14	3.440	0.170	0.033	49.43
S1	2017	33	0.6	0.02	0.54	0.09	3.80	0.19	3.300	0.210	0.045	51.73
M1	2017	34	0.59	0.02	0.6	0.10	4.47	0.19	3.650	0.210	0.019	43.53

behavior during the day making them difficult to sample (B. C. Harvey, R. J. Nakamoto, and J. L. White, personal observations). We also concluded that the collection of larvae would only slightly affect population dynamics, due to the minimal residual reproductive value of larval fish. Larvae were collected using hand nets in backwater areas (< 0.5 m depth) in late-June and July. Larvae ranged 12 to 18 mm in total length and thus were likely to have recently settled at the sampling sites. Larvae were euthanized with an overdose of tricaine methanesulfonate and preserved in 95% ethanol until DNA extraction.

Molecular methods

Whole genomic DNA was extracted from fin tissue using chelex methods (Walsh et al. 1991). We used 15 microsatellite loci developed by Tranah et al. (2001). Genotyping was conducted using the M13 fluorescent labeling protocol of Schuelke (2000). Amplifications were conducted in 12.7 µL reaction volumes with 6.25 µL Promega® PCR Master Mix® (Catalog #M7505), 4.25 µL nuclease-free water, 0.5 µL of the reverse primer (10 µmol), 0.1 µL forward M13 tailed primer (10 µmol), 0.5 µL of dye (1 µmol), and 1.1 µL of template DNA. Thermal cycling conditions were 94 °C for 3 min, 20 cycles of 94 °C for 1 min, 52 °C for 30 s, and 72 °C for 30 s followed by 15 cycles of 94 °C for 30 s, 48 °C for 45 s, 72 °C for 45 s and final extension of 72 °C for 5 min. Amplification products were separated and allele sizes determined using the Beckman Coulter CEQ™ 8000 Genetic Analysis System. Relative allele sizes were determined manually from electropherograms.

We used ARLEQUIN 3.5.2.2 (Excoffier and Lischer 2010) to perform tests for Hardy–Weinberg proportions, linkage disequilibrium, and calculate pairwise F_{ST}, and conduct tests of their significance. This software was also used to calculate the proportion of polymorphic loci, as well as observed and expected heterozygosity, and allelic richness. Corrections for multiple tests were implemented using the Bonferroni method (Rice 1989). In addition, we computed allelic richness, standardized to a sample size of 20 individuals, with ADZE (Szpiech et al. 2008).

Incorporating full-sibling groups may lead to the incorrect interpretation of population structure (e.g., Hansen et al. 1997; Anderson and Dunham 2008). Because our samples predominantly consisted of larvae, frequently gathered from the same backwater locations, we investigated whether the sampling of related individuals affected our genetic structuring analysis. We used the software COLONY 2.0 (version 2.0.6.1) (Jones and Wang 2010) to identify full-siblings and half-siblings in our collections from 2013. COLONY was run using the following settings: a polygamous mating system, no inbreeding, species are dioecious and diploid, allele frequencies unknown, length of run and likelihood precision were set to medium, and full likelihood analysis. To evaluate

the effect of family sampling on our estimates of genetic structure, we used linear regression to compare pairwise estimates of genetic differentiation (F_{ST}) between the full data set and a data set with one member of all full-sibling and half-sibling pairs identified by COLONY removed.

Downstream increase in genetic diversity

We evaluated the hypothesis of downstream increase in genetic diversity using analyses of covariance, with river kilometer as the continuous independent variable, year of collection as the categorical independent variable and allelic richness, standardized for sample size, and expected heterozygosity as alternative response variables that were analyzed separately. Analysis of both response variables included the river kilometer*year interaction and were conducted by building generalized linear models (glms) using the GLM Procedure in SAS (SAS 2023).

To evaluate the likely causes of a downstream increase in genetic diversity, we applied the Approximate Bayesian Computation (ABC) methods of Paz-Vinas et al. (2015). The approach compares summary statistics calculated from simulations under different assumptions about the processes driving patterns in genetic diversity to those statistics calculated from observed data. The simulation methods specifically test models of downstream-biased gene flow asymmetry, increased downstream habitat availability, upstream-directed colonization, two-way and three-way interactions among these factors, and a null model governed by dendritic connectivity. We used a set of summary statistics identified by Paz Vinas et al. (2015) as informative for discriminating among alternative hypotheses to explain downstream increases in genetic diversity: COR_{IBD}, COR_{IBF}, Global F_{ST}, COR_{AR} (Paz-Vinas et al. 2015). These parameters were calculated using software and methods identical to those in Paz-Vinas et al. (2015). Specifically, COR_{IBD} and COR_{IBF} are regression coefficients from multiple regression of pairwise F_{ST} (dependent variable) and two independent variables: (i) pairwise river distances (km) between collection sites, and (ii) a pairwise matrix indicating flow connected sites (= 1) and flow unconnected sites (= 0). Sites are flow connected when water flows from the upstream site to the downstream site. For example, in the study at hand, the N, MN and S sites are flow connected to all the L sites, but none of the N sites are connected to the S sites (Fig. 1). Multiple regression on the distance matrices were conducted using the MRM function with Spearman correlation ranked distances in the R package 'ecodist' (Goslee & Urban 2007). Global F_{ST} was calculated by AMOVA using ARLEQUIN 3.5.2.2 (Excoffier and Lischer 2010). Lastly, COR_{AR} was calculated as the Pearson's correlation coefficient between allelic richness, standardized to a sample size of 20 individuals using the software ADZE (Szpiech et al. 2008), and distance upstream of the river mouth. To ensure independent observations, this analysis only used 2013 collections. Analyses of downstream increase in genetic diversity was conducted using the statistical software package R (R Core Team 2023).

Upstream increase in genetic differentiation and isolation by distance

To evaluate the hypothesis that genetic differentiation declines with distance downstream (see Finn et al. 2011; Paz-Vinas et al. 2015) we used analysis of covariance, with river kilometer as the continuous independent variable and year of collection as the categorical independent variable. We used F_{ST} / (1—F_{ST}) to quantify genetic differentiation, with F_{ST} calculated as the mean pairwise F_{ST} values observed between each site*year observation and all other site*year observations in the network (e.g. Coleman et al. 2013).

We evaluated the hypothesis of isolation by distance using analyses of covariance, with river kilometer as the continuous independent variable, year of collection and site connectivity (0 or 1) as categorical independent variables and pairwise F_{ST} / (1—F_{ST}) as the response variable. We included the river distance*year and the river distance*connectivity interactions in this analysis. This analysis used a dataset without pairwise comparisons between years, comprising a total of 106 observations—91 from 2013 and 15 from 2017. We used the GLM Procedure in SAS (SAS 2023) to complete the analyses of genetic differentiation with distance downstream and isolation by distance.

Results

The 15 microsatellite loci surveyed in 933 individuals ranged from 4 to 13 alleles per locus and averaged 8.1 alleles per locus. A small fraction (1.7%) of the 13,995 genotypes scored (933 individuals at 15 loci) were missing; missing data were not associated with particular loci or sub-populations. Of the 300 tests for conformance to Hardy–Weinberg proportions (20 collections at 15 loci), none were significant following Bonferroni correction for multiple tests (critical value = 0.00017). Only 14 of the 300 tests for conformance to Hardy–Weinberg proportions had $P < 0.05$. None of the 1470 tests for linkage disequilibrium were significant following Bonferroni correction for multiple tests (critical value = 0.00002). All loci were polymorphic in all populations, except for S3, where the proportion of polymorphic loci was 0.87. Catostomidae are tetraploid but are undergoing diploidization (Ferris and Whitt 1977); we found no evidence of tetraploidy in the examined microsatellite loci.

Analysis with the software COLONY identified two full-sibling pairs in our data, one pair at site N1 (probability 0.838), and one pair at site L10 (probability 0.99). A total of 33 half-sibling pairs were identified with probability > 0.90, and 244 at a probability > 0.80. Out of the 244 half-sibling pairs identified with a probability > 0.80, 42 included half-sibling pairs from the same site whereas 202 half-sibling pairs were collected from different sites. A data set with related individuals removed was constructed by deleting one member of each full-sibling and half-sibling pair (defined by probability > 0.80). Comparison of genetic differentiation (F_{ST}) in the full data set and the data set with related individuals removed revealed a strong relationship (r = 0.96, slope = 0.919, intercept = −0.0005), thus we concluded that family-sampling was unlikely to mislead our interpretations of genetic structuring. Additionally, comparing genetic diversity levels in the datasets with and without related individuals showed no differences in the estimates significant enough to affect the conclusions.

Downstream increase in genetic diversity

Both measures of genetic diversity (rarefied allelic richness and expected heterozygosity) exhibited downstream increases, with no apparent effect of collection year (Fig. 2). Downstream distance explained 51% of the variation in allelic richness and 69% of the variation in expected heterozygosity. The analysis of covariance indicated significant increase in rarefied allelic richness with downstream distance ($F_{1,16} = 14.42$, $p = 0.002$), no significant effect of year ($F_{1,16} = 0.67$, $p = 0.425$), or the interaction of year and distance ($F_{1,16} = 1.31$, $p = 0.269$). Similarly, the analysis of covariance indicated significant increase in expected heterozygosity with downstream distance ($F_{1,16} = 33.55$, $p < 0.001$), no significant effect of year ($F_{1,16} = 0.11$, $p = 0.746$), or the interaction of year and distance ($F_{1,16} = 0.54$, $p = 0.472$).

Application of the methods to identify the processes responsible for downstream increases in genetic diversity developed by Paz-Vinas et al. (2015) yielded a clear result. The summary statistics calculated from the observed data were: $COR_{IBD} = 0.70$, $COR_{IBF,} = -0.45$, Global $F_{ST} = 0.0175$, and $COR_{AR} = -0.80$. The ABC model of Paz-Vinas et al. (2015) indicated that upstream colonization was highly likely (posterior probability = 0.98) to be the process responsible for the downstream increase in genetic diversity observed in Klamath smallscale suckers in the Smith River. The combined "Habitat/colonization" model came next with a probability of 0.02, while all remaining models registered negligible probabilities.

The primary factor influencing the selection of the upstream colonization model for the introduced population of Klamath smallscale sucker in the Smith River appears

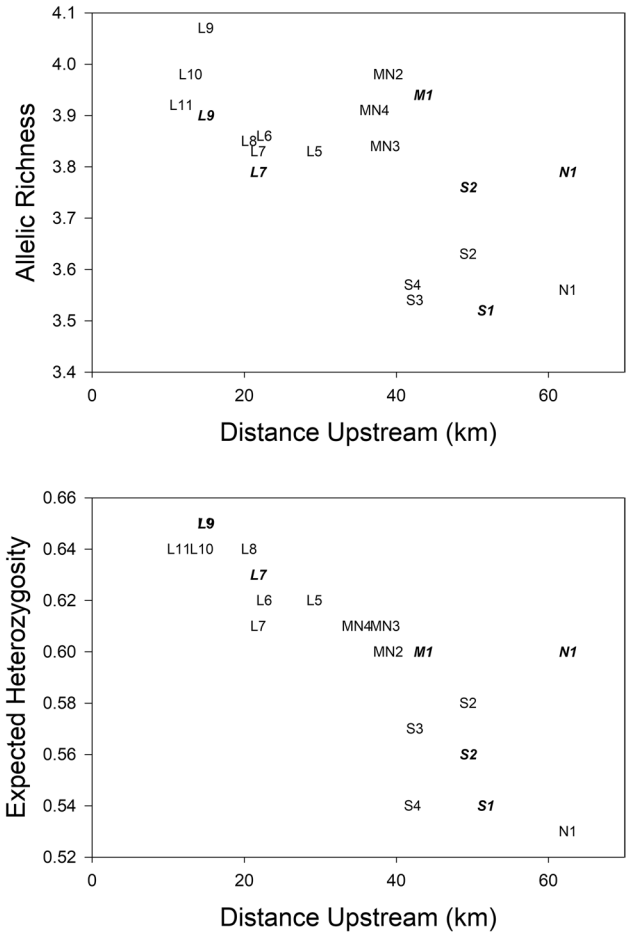

Fig. 2 Rarefied allelic richness (top) and expected heterozygosity (bottom) versus distance upstream for Klamath smallscale suckers in the Smith River, California. Symbols correspond with site labels in Fig. 1. Data labels with standard font indicate 2013 samples; bold, italicized data labels indicate 2017 samples. In the bottom panel, two pairs of data points (L10/L11 and MN3/MN4) were slightly offset for clarity. Also note in the bottom panel that samples from 2013 and 2017 at site L9 exhibited very similar values

to be low isolation by flow. However, high isolation by distance and low F_{ST} also provided support for the upstream colonization hypothesis according to the analysis of Paz-Vinas et al. (2015; see Figure S2). In contrast to our results, high connectivity by river flow would support the hypotheses of downstream-biased dispersal or increased habitat downstream, because high flow connectivity corresponds with movement of individuals downstream, leading to higher gene flow and greater dispersal in the downstream direction.

Upstream increase in genetic differentiation and isolation by distance

The overall level of genetic differentiation between sites was relatively low (mean 0.0206), but a total of 60 out of 91 pairwise comparisons of genetic differentiation (F_{ST})

achieved statistical significance (critical value = 0.0098). The analysis of covariance indicated a strong effect of distance upstream (Fig. 3, top panel, $p < 0.001$) on within-site genetic differentiation and no effects of year ($p = 0.96$) and the site*year interaction ($p = 0.93$) A simple regression with distance upstream as the predictor of genetic differentiation explained the same amount of variation ($R^2 = 0.63$) as the analysis of covariance.

Pairwise genetic differentiation conformed to an isolation-by-distance model of gene flow (Fig. 3, bottom panel).

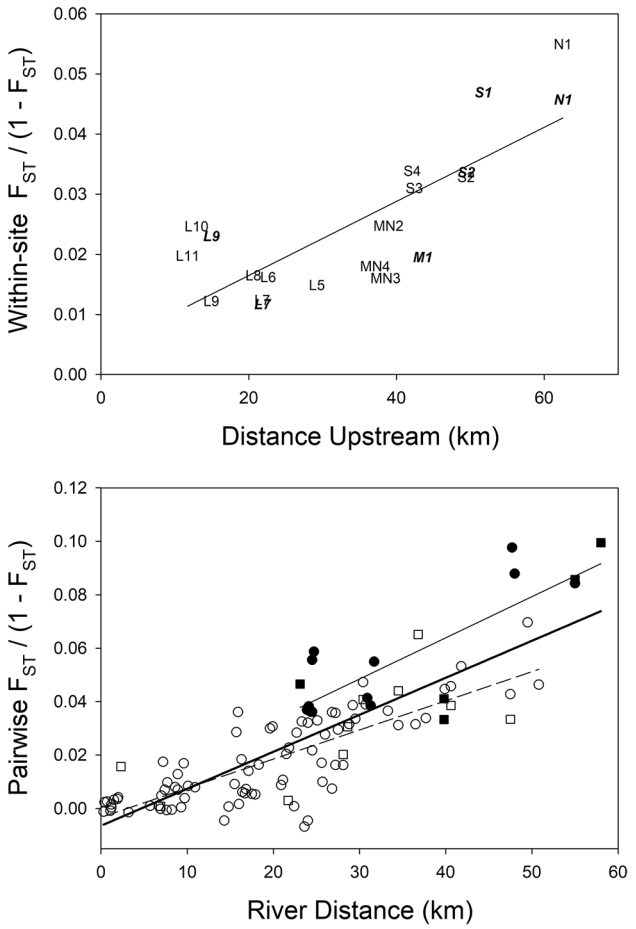

Fig. 3 Top panel: Within-site genetic differentiation (F_{ST}/[1—F_{ST}]) versus distance upstream for Klamath smallscale suckers in the Smith River, California. Data labels correspond with site labels in Fig. 1. Standard font indicates 2013 samples; bold, italicized data labels indicate 2017 samples. Note samples from 2013 and 2017 at sites L7 and S2 exhibited very similar values. Bottom panel: Relationship between pairwise genetic differentiation (F_{ST}/[1—F_{ST}]) and river distance for Klamath smallscale suckers (*Catostomus rimiculus*) in the Smith River, California. Open symbols indicate comparisons between flow connected sites (i.e. the downstream flow of water from one site flows through the second site) and filled symbols indicate comparisons of flow disconnected sites. Circles indicate 2013 data; squares indicate 2017 data. Thicker solid line indicates overall regression; lighter lines indicate separate regression results for flow connected (dashed line) and flow disconnected (solid line) sites

The analysis of covariance revealed a significant effect of river distance ($F_{1,100} = 96.03$, $p < 0.001$) and the river distance*connectivity interaction ($F_{1,100} = 5.01$, $p = 0.027$) on genetic differentiation, but no detectable effects of year ($F_{1,100} = 0.86$, $p = 0.357$), connectivity ($F_{1,100} = 2.080$, $p = 0.152$), nor the river distance*year interaction ($F_{1,100} = 1.49$, $p = 0.225$). River distance alone explained 69% of the variation in pairwise genetic differentiation, while the 5-term analysis of covariance model explained 79% of the variation.

Discussion

In a barrier-free network across sites spanning up to 58 river kilometers, we identified riverscape genetic structure in an introduced population of Klamath smallscale suckers in the Smith River, California, including downstream increase in genetic diversity, upstream increase in genetic differentiation, and isolation by distance. The genetic patterns we documented seem to have evolved from a small genetically limited founding group introduced sometime between the 1890s and 1970s. Notably, the observed genetic signature closely aligns with that expected for native populations. Our analysis suggested upstream colonization as the process responsible for the observed patterns in genetic structure, indicating either a stable or expanding range within the drainage.

Genetic diversity

Increased genetic diversity downstream is a consistent finding for animals occupying dendritic river networks, as indicated by both empirical research on varied taxa (Paz-Vinas et al. 2015) and simulation studies (Paz-Vinas et al. 2015; Paz-Vinas and Blanchet 2015; Thomaz et al. 2016). This pattern has also been observed in other catostomid fishes on larger spatial scales (Dowling et al. 2012; Douglas et al. 2003).

Our simulation analysis suggested that the downstream increase in genetic diversity among Klamath smallscale suckers in the Smith River can be attributed to consecutive upstream founding events accompanied by decreasing genetic diversity. Upstream colonization may be expected for introduced suckers, given that adult catostomids frequently migrate upstream for spawning and several studies highlight the propensity of some species to revisit spawning areas over multiple reproductive cycles (Villa 1985; Moyle 2002; Olson and Scidmore 1963; Grabowski and Isely 2006; Neely et al. 2009). The lack of Klamath smallscale suckers in the Smith River's upper reaches, as documented by field observations (BCH, RJN, JLW observations), supports a hypothesis of ongoing territorial expansion since their probable introduction. Nevertheless, the influence of climatic events like

droughts or floods potentially displacing upstream sucker populations to downstream refuges before moving back upstream cannot be dismissed. These findings offer a different perspective than earlier studies of catostomid riverscape genetic structure conducted at larger geographic scales that mainly associated upstream colonization with historic climate phenomena such as droughts and glaciations (Dowling et al. 2012; Douglas et al. 2003).

Genetic differentiation

We found a strong correlation between within-site F_{ST} and upstream distance in a natural setting, a relationship inconsistently observed in empirical studies (reviewed in Paz-Vinas et al. 2015) but frequently predicted in theoretical and simulation studies (Paz-Vinas and Blanchet 2015; Thomaz et al. 2016). Only two of the 12 studies reported on by Paz-Vinas et al. 2015) exhibited stronger relations between within-site F_{ST} and distance upstream than the one observed in this study; both examples involve fish in smaller watersheds than the Smith River that contain multiple barriers to upstream dispersal (Crispo et al. 2006; Raeymaekers et al. 2009).

Isolation by distance in riverine fish is a frequently documented phenomenon across diverse taxa (Kinziger et al. 2013; Crookes and Shaw 2016; Wiesenfeld et al. 2017; Bohonak 1999; Jenkins et al. 2010). The strong pattern of isolation by distance we observed suggests equilibrium between migration and drift at the scale of the Smith River basin. Riverscape-scale studies of catostomids have yielded mixed results regarding isolation by distance. For instance, some studies have found evidence for isolation by distance in species such as *Pantosteus clarkii* (desert sucker) (Pilger et al. 2017). In contrast, other studies have observed a lack of overall isolation by distance in species like *Catostomus santaanae* (Santa Ana sucker), *Catostomus catostomus* (longnose sucker), and *Catostomus insignis* (Sonora sucker) (Salisbury et al. 2016; Pilger et al. 2017; Richmond et al. 2018). Notably, some of these latter cases occurred in systems with significant barriers to migration and invoked higher than expected influence of genetic drift.

Given consistent observations of passive downstream transport of larvae in many sucker populations, including Klamath smallscale suckers in the Smith River (White and Harvey 2003), one might anticipate an effect of larval drift on patterns of genetic structure. Consistent with this, we observed a modest negative effect of flow connectivity on genetic differentiation, but river distance per se had a much stronger effect. The ABC analyses corresponded with these results by indicating that upstream-directed colonization was a more likely explanation for the patterns in the data than downstream-biased dispersal. While long distance downstream transport of some larval suckers probably occurs in the Smith River, relatively limited downstream transport may predominate. Considering that larval catostomids emerge from the substrate at relatively large sizes (ca. 10 mm total length), they may have significant capability to resist displacement by flow, given that this capability has been observed to rapidly increase at lengths of 8 to 10 mm (Harvey 1987). Kennedy and Vinyard (1997) observed drift avoidance capabilities and relatively low propensity to drift by larval Warner suckers (*C. warnerensis*), which they hypothesized could be related to unreliable habitat in the terminal lake in the river system occupied by the species. The Smith River may similarly disfavor drift because of its limited extent, steepness, lack of lentic habitat, and marine terminus. In addition, consistently high densities of juvenile salmonids throughout the Smith River may increase the risk of predation for drifting larval suckers. While the unidirectional flow of water can be a major factor for increased downstream genetic diversity, a variety of studies highlight the importance of other processes (e.g., Hänfling & Weetman 2006; Kikuchi et al. 2009; Paz-Vinas et al. 2015).

Genetic change in introduced species

Theoretical frameworks indicate that introduced populations can be hotspots for evolutionary transformations, including both adaptive and non-adaptive changes (Baker and Stebbins 1965; Bock et al. 2015). Consistent with these predictions, our findings indicate at least non-adaptive genetic change within the introduced population of Klamath smallscale sucker. This population, initiated by a severe founder event, evolved highly structured genetic variation coinciding with predictions for organisms in idealized dendritic networks (e.g., Paz-Vinas et al. 2015; Thomaz et al. 2016). However, for catostomids in nature, these patterns can be easily disrupted by historical and contemporary factors, such as urbanization, that constrain connectivity and elevate the importance of non-equilibrium processes (Salisbury et al. 2016; Pilger et al. 2017; Richmond et al. 2018).

The extent of adaptive genetic change within the introduced Smith River population of Klamath smallscale suckers is yet to be determined (Colautti and Lau 2015). The pronounced founder event implies that adaptation from standing genetic variation would be unexpected (but see Dlugosch and Parker 2008). The evolutionary pressure for adaptive genetic change in the introduced population is also unclear: the Smith River ecosystem exhibits broad similarities to the source environment.

Numerous studies have demonstrated that introduced species often undergo rapid genetic change, both neutral and adaptive, sometimes in less than 50 years (reviewed by Whitney and Gabler 2008). While the exact timeline of the non-adaptive evolutionary change we observed in the Smith River population of Klamath smallscale suckers

remains uncertain, it is estimated to have occurred within 40 to 110 years (Kinziger et al. 2021), which corresponds with about 7 to 16 generations, assuming a 6-year generation time for Klamath smallscale suckers (Moyle 2002). This finding provides insights on the timescale for the emergence of neutral genetic structures within introduced populations.

Coleman et al. (2013) emphasize that the protection of genetic uniqueness might come at the expense of overall genetic diversity within species: focusing solely on genetic uniqueness could lead to a loss of genetic variation necessary for a species' adaptation and long-term survival. Our findings of increasing genetic differentiation upstream and increasing genetic diversity downstream reflect the conservation trade-off acknowledged by Coleman et al. (2013). However in this case, the distinctiveness of upstream populations seems likely to be mostly explained by limited diversity rather than significant adaptation to local conditions. Downstream areas harbor a large proportion of overall genetic diversity, presumably because downstream areas have relatively recently acted as source areas for upstream sub-populations. However, if conservation efforts focused on downstream habitat in the Smith River become necessary, they would face typical challenges for these areas, including patterns in land ownership and relatively high levels of human influence.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s10592-024-01651-5.

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Author contributions Bret Harvey and Andrew Kinziger identified the research question and designed the study. Rodney Nakamoto led the sample collection and genotyping, assisted by the coauthors. Andrew Kinziger and Bret Harvey analyzed the data and wrote initial drafts of the paper. All authors reviewed and commented on previous drafts of the manuscript. The final manuscript was read and approved by all authors.

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Data availability Sample metadata and associated multilocus genotypes are available as supplementary material. Link to the simulations and scripts by Paz-Vinas et al. (2015) are available in the Dryad repository: https://datadryad.org/stash/dataset/doi:https://doi.org/10.5061/dryad.38c1g.

Declarations

Competing interests The authors declare no competing interests.

Ethical approval This research was reviewed and approved the Cal Poly Humboldt Institutional Animal Care and Use Committee (Protocol: 13/14.F.09-A).

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