ABSTRACT
Biological invasion by non-native species has been identified as one of the major threats to native fish communities worldwide. The fish community of San Francisco Estuary is no exception, as the estuary has been recognized as one of the most invaded on the planet and the system has been impacted significantly by these invasions. Here, we summarize the introduction and probable establishment of a new species in the Sacramento–San Joaquin Delta, the Bluefin Killifish (*Lucania goodei*), as discovered by the US Fish and Wildlife Service Delta Juvenile Fish Monitoring Program (DJFMP). The DJFMP has conducted a large-scale beach seine survey since 1976, and it is the longest-running monitoring program in the San Francisco Estuary that extensively monitors the shallow-water nearshore habitat. Possibly introduced as discarded aquarium fish within the vicinity of the Delta Cross Channel, Bluefin Killifish is a close relative of the Rainwater Killifish (*Lucania parva*), another non-native fish species that has been present in the San Francisco Estuary system for decades. Studies in their native range suggest that Bluefin Killifish will fill a similar niche to Rainwater Killifish, albeit with a more freshwater distribution. The potential ecological impact of Bluefin Killifish remains unclear in the absence of additional studies. However, we have been able to track the spread of the species within the Sacramento–San Joaquin Delta through the existence of long-term monitoring programs. Our findings demonstrate the value of monitoring across various habitats for the early detection and proactive management of invasive species.

KEY WORDS
biological invasion, introduced species, Bluefin Killifish, *Lucania goodei*, life history

INTRODUCTION
Invasion by exotic species is one of the main causes of biodiversity loss worldwide (Sala et al. 2000; Bellard et al. 2016; Blackburn et al.)
Although species invasions have been a constant throughout geologic history, the scale at which species move between systems today is unprecedented (Mooney and Cleland 2001). As a result of human intervention (intentional or otherwise), every region around the world is currently experiencing an accelerated rate of biological invasion several orders of magnitude higher than that of prehistoric rates (Ricciardi 2007). Invasive species are topics of interest in natural resources management because they can have profound detrimental effects on local native biota, and the systems they invade can incur severe economic costs (Pimentel et al. 2005).

The San Francisco Estuary (estuary) is an estuary of significant ecological and socio-economic importance that for many decades has been one of the most invaded estuaries in the world (Cohen and Carlton 1998). The Sacramento–San Joaquin Delta (Delta) is a network of dredged channels that make up the tidal freshwater portion of the estuary. The Delta supplies water to over 27 million people and supports an agricultural industry valued at over USD $38 billion (Lund et al. 2008; Delta Stewardship Council 2018). The Delta also hosts various aquatic species endemic to the estuary that include the previously listed Sacramento Splittail (Pogonichthys macrolepidotus) and the endangered Delta Smelt (Hypomesus transpacificus). Invasive species have significantly affected the Delta ecosystem over the years, and have been directly and indirectly linked to declines of native species. The Central Valley-endemic Sacramento Perch (Archoplites interruptus) was extirpated from the Delta largely from a combination of habitat degradation and interaction with invasive Centrarchid fish species (Marchetti 1999; Crain and Moyle 2011). The accidental introduction of the invasive clam Potamocorbula amurensis in the mid-1980s, most likely through ballast water, led to a severe decline in the lower trophic food web of the estuary, and subsequently caused a drop in abundance of multiple pelagic fish species (Nichols et al. 1990; Kimmerer et al. 1994; Thomson et al. 2010). Water hyacinth (Eichhornia crassipes), which originates from the Amazon basin, has altered turbidity and dissolved oxygen levels in the Delta (Tobias et al. 2019) and, at times, has blocked waterway navigation and affected water delivery operation (Marineau et al. 2019).

Although the majority of key invasive species in the Delta arrived through ballast water (Nichols et al. 1990; Choi et al. 2005; Winder and Jassby 2011) or intentional human-assisted introduction (e.g. Striped Bass Morone saxatilis and Largemouth Bass Micropterus salmoides) (Moyle 2002), past releases of ornamental aquarium species have at times led to extensive ecosystem shifts. For example, the Brazilian Waterweed (Egeria densa), a popular ornamental species that quickly proliferated in the Delta, has been shown to facilitate the spread of the non-native Largemouth Bass (Conrad et al. 2016) and caused a large-scale decline of turbidity in the region (Hestir et al. 2016). Numerous species from aquaria and aquatic ornamental culture have invaded natural ecosystems worldwide, and a large majority of these are freshwater fishes (Padilla and Williams 2004). The freshwater fish community of the Delta today is largely dominated by invasive species in both total number of species and abundance (Cohen and Carlton 1998; Brown and Michniuk 2007; Mahardja et al. 2017). Mediterranean-climate estuaries, such as the San Francisco Estuary, generally support a high level of endemism, which suggests that the system’s island-like biota are more vulnerable to invasion (Marr et al. 2010). Introduced species can displace native species through competition, predation, environmental modification, disease transfer, and hybridization (Moyle et al. 1986). Given the decline of multiple species of concern in the Delta and the increased global invasion rate in recent years, it is critical for existing monitoring programs to be vigilant and aware of the potential establishment of new introduced species.

Since 1976, the US Fish and Wildlife Service (USFWS) has conducted beach seine surveys to evaluate the abundance and distribution of juvenile Chinook Salmon (Oncorhynchus tshawytscha) and various resident fish species in the estuary, with focus on the Delta (Kjelson
et al. 1982; IEP et al. 2019). Dubbed the Delta Juvenile Fish Monitoring Program (DJFMP), the beach seine survey has been the primary monitoring program of the estuary that evaluates fish community changes in nearshore, littoral habitat (Mahardja et al. 2017). On October 10, 2017, DJFMP staff encountered what appeared to be a new killifish (*Lucania* spp.) species at the Delta Cross Channel (DCC) beach seine station (Figure 1). No specimens from this date were collected; however, photographs were taken that allowed staff to tentatively identify them as Bluefin Killifish (*Lucania goodei*). A second observation of this species, at the same location on November 3, 2017, allowed DJFMP staff to collect specimens and further confirm the original species identification in a laboratory setting. However, external morphological traits are solely relied on, misidentification can sometimes occur because of the lack of distinctive traits between species, degradation of specimens, or the presence of interspecific hybrids (Godbout et al. 2009; Hull et al. 2010; Benjamin et al. 2018). In addition, it is unclear if this putative Bluefin Killifish observation constitutes the establishment of a new species in the estuary, given that even intentional species introductions have failed in the past (Dill and Cordone 1997). Here, we describe our effort to genetically confirm the occurrence of Bluefin Killifish, conduct literature review on the species’ biology, and describe their initial spread in the Delta.

Figure 1  Map of the San Francisco Estuary and the sites (triangles) regularly sampled by the DJFMP beach seine survey
METHODS

Field Sampling

The DJFMP beach seine survey began in 1976 with the initial goal of monitoring the abundance and distribution of juvenile Chinook Salmon in the estuary, with a particular focus on the Delta region (Kjelson et al. 1982; IEP et al. 2019). Since then, the survey expanded several times, and its objective now includes the monitoring of other fish species that are of interest to natural resource management agencies (e.g., Sacramento Splittail, Delta Smelt, etc.). The DJFMP currently samples over 60 beach seine sites throughout the estuary and the lower Central Valley of California, either weekly or every 2 weeks year-round (Figure 1). Because of the extensive spatio-temporal coverage of this sampling effort, data from the beach seine survey has been used over the years to better understand fish habitat and community changes within the shallow, near-shore habitat of the Delta (Sommer et al. 2001; Feyrer et al. 2005; Brown and May 2006; Mahardja et al. 2016; Mahardja et al. 2017; Munsch et al. 2019).

DJFMP beach seine sampling was conducted by hauling a single 15.2-m x 1.3-m beach seine net with 3-mm\(^2\) mesh and a 1.3-m x 1.3-m bag into shore. After each seine haul, all fish larger than 25-mm fork length were identified to species and then counted (with the exception of a few species that can be identified even at <25-mm fork length). Up to 30 fish per species from each seine haul were measured for fork length, after which any additional fish were simply counted. For fish species listed under the Endangered Species Act, up to 50 fish per species were measured. At every sampling occasion, a YSI PRO 2030 meter was used to measure water temperature (°C) and conductivity (μS cm\(^{-1}\)); a HACH 2100Q turbidity meter was used to measure turbidity levels in nephelometric turbidity units (NTU).

The first observation of the putative Bluefin Killifish occurred on October 10, 2017 at a beach seine site within the DCC (Figures 2 and 3; Table 1). As part of regular monitoring, DJFMP staff again encountered the species on November 3, 2017 at the DCC and captured two fish. These two fish were collected and sent to University of California, Davis for genetic verification. After the two initial observations of the putative Bluefin Killifish, DJFMP crew conducted additional beach seining at the same location on November 29, 2017 (not part of regularly scheduled sampling). They collected over 100 putative Bluefin Killifish, many of which were in the small juvenile size range (fork length ≤ 15 mm) (A. Goodman, pers. observation). Of the fish collected on November 29, 2017, 13 individuals were preserved in ethanol for genetic analysis. On September 11, 2018, California Department of Fish and Wildlife (CDFW) also collected a single putative Bluefin Killifish from a beach seine haul near Decker Island in the western portion of the Delta (Figure 2, Table 1) (CDFW et al. 2018). Given the relatively long distance between Decker Island and the DCC, we also conducted genetic analysis for the fish collected at Decker Island. In this article, we summarize all known occurrences of Bluefin Killifish in the estuary (identified based on morphology and/or genetics) up to September of 2019.

Genetic Analysis

The Genomic Variation Laboratory at the University of California, Davis performed DNA barcoding to confirm the species of the 16 killifish collected by the USFWS DJFMP and CDFW. DNA was extracted from fin tissue of the collected killifish using the DNEasy Blood and Tissue DNA extraction kit (Qiagen). DNA was amplified and sequenced at the cytochrome oxidase I mitochondrial DNA (mtDNA) gene using primers FishF2 and FishR2 (Ward et al. 2005). The 25 μl PCR reaction contained 2.5 μl 10x PCR buffer, 1.25 μl MgCl\(_2\) (50 mM), 1 μl dNTPs (0.2 μM), 0.5 μl forward and reverse primers (10 μM), 1 μl 1X BSA (bovine serum albumin, New England Biolabs), 0.225 μl FastStart Taq DNA Polymerase (Roche), 2 μl genomic DNA template, and 16.025 μl ultrapure water. Amplifications were performed using an ABI GeneAmp PCR System 9700 with the following protocol: 2 min at 95 °C followed by 35 cycles of 30 sec at 95 °C, 45 sec at 53 °C, and 1 min at 72 °C, followed in turn by 10 min at 72 °C. PCR amplicons were cleaned using Ampure XP Beads (Beckman Coulter) following
the manufacturer’s guidelines. Sanger sequencing was conducted by QuintaraBio (Richmond, California).

We trimmed low-quality base pair reads at each end of the cytochrome oxidase I sequences in the program Sequencher 4.8 (Gene Codes Corporation). The resulting high-quality sequences were used to query the public sequence repositories Barcode of Life BOLD System repository (BoL; http://www.boldsystems.org/) and the NCBI Nucleotide Database (https://www.ncbi.nlm.nih.gov/nucleotide/). We determined preliminary species identification based on similarity to known species-specific sequences in those repositories. We downloaded cytochrome oxidase I sequence data from the two closest species matches from BOLD and NCBI and aligned

Figure 2  Locations where Bluefin Killfish collections or observations occurred up to September 2019. DJFMP beach seine stations are labeled by their station ID as seen in Table 1.

Figure 3  Photograph of the first Bluefin Killfish captured at the Delta Cross Channel on October 10, 2017. Photo Source: Phil Voong.
to unknown killifish sequences using Clustal W in the program MEGA7 (Thompson et al. 1994; Kumar et al. 2016). We created three groups, one for each species identified as being closely related (Lucania goodei $n = 5$, Lucania parva $n = 6$) and the unknowns ($n = 16$), and we calculated the between-group mean pair-wise genetic distances between them in MEGA7. This analysis used all nucleotide substitutions (transitions and transversions, coding and non-coding), assumed uniform evolution among lineages and sites, and used a gap treatment of complete deletion (i.e., any sites with gaps are eliminated from analysis). A thousand bootstrap replicates were performed to estimate variance.

RESULTS

Only one killifish species, the non-native Rainwater Killifish ($L. ~parva$), was known to be present in the Delta system before 2017. The DJFMP distinguished the original killifish specimens collected in 2017 from Rainwater Killifish, and visually identified them as Bluefin Killifish based on the conspicuous dark lateral stripe on the fish that extends from the snout to the tail (Figures 3 and 4). Subsequent genetic analysis of these specimens from 2017 confirmed their visual identification as Bluefin Killifish. The two closest species matches to the 15 unknown killifish samples were the Bluefin Killifish and Rainwater Killifish. The unknown samples showed greatest sequence similarity to the Bluefin Killifish (Tables 2 and 3). The final alignment for the between-group mean pair-wise genetic distance analysis was 571 base pairs, and the analysis showed that the unknowns were more similar to the Bluefin Killifish than the Rainwater Killifish (Table 4).

After the first confirmed observations of Bluefin Killifish at the DCC in October and November of 2017, the DJFMP beach seine survey collected another Bluefin Killifish in December of 2017 at the Wimpy’s Marina site along the Mokelumne River over a kilometer south of the original DCC site (Figure 2). A year later on September 11, 2018, the CDFW also collected a Bluefin Killifish by beach seine near Decker Island in the western Delta. A similar genetic analysis was conducted
on this fish, given the distance between Decker Island and the DCC (~ 30 river km), which confirmed that it was indeed a Bluefin Killifish (99.85% similarity to Rainwater Killifish, Table 2). On December 14, 2018, during a boat electrofishing study, the DJFMP captured a single Bluefin Killifish at Beaver Slough near the Mokelumne River. On May 1, 2019, a boat electrofishing survey conducted by the East Bay Municipal Utility District (EBMUD) also collected a couple of Bluefin Killifish at Hog Slough adjacent to the lower Mokelumne River. In 2019, the DJFMP observed Bluefin Killifish at two new beach seine locations along the lower Sacramento River: Sherman Island and Brannan Island (Figure 2, Table 1).

**DISCUSSION**

Sound management of the San Francisco Bay–Delta system requires a proper understanding of the presence and role of non-native species. Prompt detection of new invasive species would allow for swift actions that could potentially stop or slow their advance and/or mitigate their effects. Such early detection is more likely when long-term monitoring programs with staff well trained in species identification are in place. There remains some level of uncertainty on whether Bluefin Killifish have become established in the Delta system. However, given that Bluefin Killifish have been observed from the Mokelumne

![](figure4.png)

**Figure 4** Side-by-side photographs of male (top) and female (bottom) Bluefin Killifish (left) and Rainwater Killifish (right) from Florida. Photos reprinted with permission: Zachary Randall, Florida Museum of Natural History (UF 236230 and UF 238088).

**Table 2** Sequence similarities between unknown samples and closest species matches identified by Barcode of Life public sequence repository. %Sim refers to percent sequence similarity between the unknown specimen and the online accession. Unk #16 is the specimen collected at Decker Island by CDFW.

<table>
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<th>Species 2</th>
<th>%Sim</th>
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</tbody>
</table>
River to the confluence between the Sacramento River and the San Joaquin River (>30 river km) over the span of 2 years (lifespan of the species) (Rohde et al. 1994), the species has probably become established in the region. It is not really surprising to see that another species has joined the extensive list of invasive fish species already present in the Delta (Cohen and Carlton 1998; Moyle 2002), especially given the accelerating invasion rate observed worldwide in the past few decades (Ricciardi 2007). Nonetheless, our finding highlights the types of key information that long-term monitoring programs can provide.

### Identification

Bluefin Killifish is a small-sized fish species that generally only reaches up to 50 mm in total length. They have small, upturned mouths and are fairly slender, with compressed bodies and a rounded tail (Page and Burr 2011). The species has a distinctive wide black stripe along the midline of the entire length of its body (Figures 3, 4). This black stripe starts from the tip of the snout, then appears to go through the eye, and ends at a black spot at the base of the caudal fin (Nunziata 2010). This characteristic makes Bluefin Killifish relatively easy to identify against morphologically similar fishes in the Delta (e.g., Rainwater Killifish, Western Mosquitofish *Gambusia affinis*). The origin of their dorsal fin

### Table 3

Sequence similarities between unknown samples and closest matches identified by blasting the NCBI Nucleotide database. Individuals often matched to multiple individuals from each species but the top match for *Lucania goodei* and *L. parva* are shown here. %QC refers to query coverage, or overlap between the unknown killifish alignment and individual sequences within the Nucleotide database. %Identity is the sequence similarity between the unknown sample and species. Unk #16 is the specimen collected at Decker Island by CDFW.

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<th>%Identity</th>
<th>Species 2</th>
<th>%QC</th>
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### Table 4

Between group mean pairwise genetic distance, or number of base differences per site averaging across sequence pairs, as calculated in MEGA7 *(below diagonal).* Standard error based on 1000 bootstrap iterations is *above the diagonal.* Group compositions were as followed: 5 Bluefin Killifish, 6 Rainwater Killifish, 16 unknown specimens.

<table>
<thead>
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<th>Rainwater Killifish</th>
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is anterior to the origin of their anal fin, which distinguishes killifish species from the Western Mosquitofish that are fairly ubiquitous throughout the Delta. Bluefin Killifish and Rainwater Killifish are closely related, and aside from the distinct stripe of the Bluefin Killifish, we found no key meristic trait that can distinguish the two species. However, Hubbs et al. (2008) indicated that Bluefin Killifish tends to be more slender-bodied, with standard length that is roughly 4.5 to 5 times their body depth, whereas Rainwater Killifish are expected to have standard length that is about 3.5 to 4 times their body depth.

Bluefin Killifish are sexually dimorphic. The fish likely derives its name from the fact that the anterior of the dorsal fin on males is generally colored blue. Adult Bluefin Killifish males typically have red pigmentation at the base of their caudal fin, and may have brightly colored pelvic, dorsal, anal, and caudal fins (Fuller 2002). These male color patterns are largely driven by the genetic makeup of individuals, although the transmission of ultraviolet/blue wavelengths (360–478 nm) through the various bodies of water they inhabit has also been shown to influence male color patterns (Fuller et al. 2005). Males with blue anal fins are commonly found in highly turbid swamps and lakes (waters with low transmission of ultraviolet and blue wavelengths); males with red or yellow anal fins are commonly found in clear water (waters with high transmission of ultraviolet and blue wavelengths) (Fuller and Travis 2004). Female Bluefin Killifish, on the other hand, lack colored fins (Page and Burr 2011). Both sexes are dusky brown to olive in color above their midline, and a mix of olive/brown and silvery white below. Their scales have dark edges, and their anal and dorsal fins both have thin black edges.

**Distribution**

Bluefin Killifish are native to the Ogeechee River drainage in southern Georgia, the Chipola River drainage in southeastern Alabama, and throughout most of Florida (only absent west of Choctawhatchee River drainage in the panhandle) (Page and Burr 2011). There are currently self-sustaining introduced populations of Bluefin Killifish in North Carolina, South Carolina, and Texas (Gallaway et al. 2008; Fuller 2019). The species was established in the Cooper River, South Carolina in 1973 (Christie and Curtis 1983); in Cape Fear River, North Carolina in 1977; and near the city of Victoria, Texas in 1998 (Fofonoff et al. 2019).

**History in California**

Bluefin Killifish made its initial appearance in California in 1959, when a single fish was found in the first shipment of Florida-strain Largemouth Bass to San Diego County from the Holt State Fish Hatchery of the Florida Game and Fresh Water Fish Commission (Hubbs and Miller 1965; Dill and Cordone 1997). It would be another 20 years before Bluefin Killifish was accidentally introduced into a water body in California. In 1980, a shipment of Asian milfoil (Myriophyllum spp.) from Florida was sent to Los Angeles to be sold at local aquarium/pond supply stores. The shipped Asian milfoil contained the eggs of Bluefin Killifish, and the hatchlings survived several months in a few ponds in the area (Swift et al. 1993). Although the species was present in these isolated ponds, there was no record of Bluefin Killifish in any public waters in the state until 2000 (Dill and Cordone 1997). In July 2000, the Marine Sciences Institute at the University of California, Santa Barbara captured seven Bluefin Killifish by beach seine on the upper part of the San Dieguito River in San Diego County while conducting their annual monitoring. In September of the same year, they captured five more Bluefin Killifish in the same location using dip nets (Huang et al. 2003). After these observations, Bluefin Killifish was thought to be established on the San Dieguito River; however, no Bluefin Killifish individuals have been found in the area since 2001 (2017 personal conversation between D. Huang and A. Goodman, unreferenced, see “Notes”). Major tidal wetland restoration effort has been conducted on the San Dieguito River since 2006, which led to higher salinity variability within the river (Nordlie and Haney 1998) that may have created unfavorable conditions for Bluefin Killifish.
Life History
Multiple aspects of Bluefin Killifish biology contribute to a potentially high population growth rate. First, Bluefin Killifish are extremely iteroparous: a single female may spawn every day for several weeks (Breder and Rosen 1966). Females release 1 to 2 eggs per spawn, and can deliver up to 20 eggs per day across multiple spawning attempts with one or more males. This high degree of iteroparity makes it difficult to estimate lifetime fecundity, because fecundity depends on the length of time that females remain reproductive. The high degree of iteroparity, combined with their mating behavior, predisposes Bluefin Killifish to have a high degree of outcrossing and low FST among populations (Creer and Trexler 2006; Fuller and Johnson 2009; Johnson et al. 2018).

Second, Bluefin Killifish have a long spawning season in comparison to temperate killifish and topminnows. Foster (1967) states that Bluefin Killifish breed from late January to mid-September. Arndt (1971) generally agrees with this, but adds that there is a great deal of variation between Bluefin Killifish populations. At some localities, ripe females can be found throughout the year (Arndt 1971; Rohde et al. 1994). In a laboratory setting, Bluefin Killifish can be induced to spawn year-round under the appropriate light ratio and temperature, albeit with a dramatic drop in egg production between September and January (R. Fuller, pers. observation).

Third, like many small cyprinodontiform fishes, Bluefin Killifish have a short time to adulthood. Foster (1967) stated that sex differences emerge by 29 days post-hatching, and that species-specific courtship behavior develops in males by 52 days post-hatching (see also Arndt 1971). In a laboratory setting, Bluefin Killifish can be induced to spawn year-round under the appropriate light ratio and temperature, albeit with a dramatic drop in egg production between September and January (R. Fuller, pers. observation).

Bluefin Killifish have a few notable habitat requirements: submerged aquatic vegetation and hard, fresh water with somewhat alkaline pH (Foster 1967; Arndt 1971; Gilbert and Burgess 1980; Dunson and Travis 1991; Page and Burr 2011). In nature, males guard patches of vegetation from other competing males and also from heterospecific fish (typically minnows). Females visit males in their territories, where the female fish are then courted. If courtship continues, females may spawn their eggs on a single male's territory or, if disrupted, may disperse their eggs among multiple males. The submerged aquatic vegetation serves as a spawning substrate for the eggs, refuge from large fish predators (particularly for juveniles), and as a source of food (i.e., small invertebrates, crustaceans, epiphytes, and vascular plants) (Gilbert and Burgess 1980; Mettee et al. 1996). Water chemistry also largely determines the distribution of Bluefin Killifish in its native range. In Florida, Bluefin Killifish are found in hard, fresh water with pH > 7. These habitats range from springs to rivers to lakes/ponds to swamps (Foster 1967; Arndt 1971; Fuller 2002; Fuller and Noa 2008). In Florida, Bluefin Killifish are notably absent in soft water with pH < 7, presumably because of their low tolerance of soft water (Dunson and Travis 1991). They can occasionally be found in slightly brackish waters, but they are largely absent in salinities > 10 ppt. While Bluefin Killifish can tolerate and are occasionally found in salinities up to 10 ppt (Foster 1967; Fuller and Noa 2008), they are typically outcompeted and replaced by Rainwater Killifish at slightly brackish salinities (Dunson and Travis 1991; Berdan and Fuller 2012). In contrast, Rainwater Killifish have low overwinter survival in cold, fresh water (Fuller et al. 2007).

Introduction into the Sacramento–San Joaquin Delta
Successful invasion often occurs when there is a close match between the invading species’ original and new environments (Moyle and Marchetti 2006). The conditions where Bluefin Killifish were originally found in this study (i.e., warm, low water velocity, shallow, freshwater habitat) likely played a key role in the species’ persistence within the Delta. Our first observation of Bluefin Killifish took place at the DCC, a
Burr 2011), with anecdotal evidence of the species surviving at ~ 4.5 °C in captivity (Nunziata 2010). Bluefin Killifish was observed in the Delta at temperatures measuring 8.9 °C, and the species appeared to have persisted for multiple winters in the area (Table 1), indicating that Bluefin Killifish may be more thermally tolerant than previously thought.

In their native range, Bluefin Killifish appears to have undergone speciation from Rainwater Killifish based on local adaptation to different salinity ranges (Dunson and Travis 1991; Fuller et al. 2007; Berdan and Fuller 2012). Bluefin Killifish is more associated with freshwater habitat, while Rainwater Killifish seems to prefer brackish water. The two Lucania species are closely related (Whitehead 2010) and can hybridize with one another; however, reduced hybrid fitness (Fuller 2008) and sexual selection for conspecifics (Kozak et al. 2015) suggest that minimal introgression will occur. Given that the two Killifish species seem to have diverged somewhat recently and differ primarily in their salinity tolerance, we expect that if Bluefin Killifish continue to be present in the system, they would occupy a niche in the estuary similar to the established Rainwater Killifish, albeit with a more upstream (i.e., freshwater) distribution.

What future effect Bluefin Killifish will have on the Sacramento–San Joaquin Delta ecosystem is unclear, because the ecological role of small-bodied resident fishes such as Rainwater Killifish have not been well studied in the region. Rainwater Killifish in the estuary is mostly presumed to have low ecological impact, given that the species has been mainly restricted to low-order, shallow tidal marsh habitat alongside Western Mosquitofish and Threespine Stickleback (Gasterosteus aculeatus) (Visintainer et al. 2006; Gewant and Bollens 2012; Grimaldo et al. 2012). Nonetheless, the establishment of a new invasive species rarely, if ever, benefits the native biota of the system it invades (Moyle et al. 1986).

Submerged aquatic vegetation that serves as spawning habitat for Bluefin Killifish has spread rapidly in the Sacramento–San Joaquin Delta (Hestir et al. 2016). Meanwhile, climate change is
expected to increase water temperature (Cloern et al. 2011), which would likely improve overwinter survival for the species. It is probable that Bluefin Killifish would expand rather quickly into shallow-edge habitats within the vicinity of the Delta in the next several years.

CONCLUSION
We were able to record the introduction and likely establishment of Bluefin Killifish in the Sacramento–San Joaquin Delta through the DJFMP, one of the multiple long-term ecological monitoring programs within the estuary. The DJFMP regularly and frequently surveys a sizeable part of the estuary (Figure 1), providing a consistent data set on the littoral fish assemblage that makes possible the early detection of new invasive species within shallow-water habitat. This information will become more essential in the coming years, as shallow-water habitat is expanded through tidal wetland restoration efforts, and climate change creates conditions more favorable to invasive littoral fish species (Brown and May 2006; Moyle et al. 2013; Mahardja et al. 2017). Climate change is projected to increase water temperature and the occurrence of droughts in California to the detriment of California’s native fish species (Cloern et al. 2011; Dettinger 2013; Davis et al. 2019). As the San Francisco Bay–Delta system continues to change, it is increasingly important to be more proactive than reactive to the challenges posed by invasive species. Bluefin Killifish was possibly introduced into the system as discarded aquarium fish. Outreach programs that provide invasive species education to the public remain critical to prevent the future introductions of new species (Chang et al. 2009).

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NOTES

Huang D. 2017. Email communication between A. Goodman and David Huang, University of California, Santa Barbara, regarding any recent observations of L. goodei in the San Dieguito River area.