State of California Department of Fish and Wildlife

Memorandum

Date: 27 April 2023

- To: Leslie Alber, Senior Environmental Scientist; Sierra District Supervisor; North Central Region Fisheries
- From: Isaac Chellman, Environmental Scientist; High Mountain Lakes; North Central Region Fisheries
- Cc: Region 2 Fish Files
- Ec: CDFW Document Library

Subject: Native amphibian restoration in Bucks Lake Wilderness, Plumas County.

• 2021–2022 update on *Rana sierrae* captive rearing, release, and monitoring.



California Department of Fish and Wildlife (CDFW) is focusing on two areas in Bucks Lake Wilderness (**Figure 1**), the Gold Lake area and the Mount Pleasant area, to benefit state threatened and federally endangered Sierra Nevada Yellow-legged Frogs (*Rana sierrae*, SNYLF). The Gold Lake area (**Figure 2**) includes Gold Lake, Rock Lake, Mud Lake, and tributaries. Gold Lake is a site from which CDFW removed introduced Brook Trout (*Salvelinus fontinalis*; BK) to benefit SNYLF. The Mount Pleasant area (**Figure 2**), which includes several small ponds, is located about 2 kilometers (km) northwest of the Gold Lake area. CDFW has designated both areas as Native Species Reserves (NSRs) in the Aquatic Biodiversity Management Plan for the Bucks Lake Wilderness Management Unit (ABMP; CDFW 2015).

Amphibian monitoring data from 2004–2022 reveals small, persisting SNYLF populations in both areas. However, the population sizes have remained low for years, and biologists have consistently observed a small number of dead SNYLF in the Mount Pleasant area. Additionally, SNYLF populations in Bucks Lake Wilderness, like many populations at the northern extent of the species' range, are small and isolated, resulting in high risk of extirpation. Therefore, these populations are of particular conservation concern to CDFW.

The Interagency Conservation Strategy for Mountain Yellow-legged Frogs in the Sierra Nevada (hereafter "Strategy"; MYLF ITT 2018) highlights reintroductions as a principal method for SNYLF recovery. As a result, in September 2018, Plumas National Forest (PNF) staff from the Mount Hough Ranger District collected 64 larval and metamorphic SNYLF for captive rearing at the San Francisco (SF) Zoo. SF Zoo staff raised the SNYLF to maturity, and PNF and CDFW staff released a subset of these frogs (those large enough for release in 2019) back into the Gold Lake and Mount Pleasant areas on 28 June 2019. In October 2019, PNF staff collected an additional 42 larval SNYLF from the Mount Pleasant area for captive rearing at the SF Zoo. On 19 June 2020, PNF and CDFW staff released a subset of these frogs (those large enough for release in 2020), including all remaining individuals from the 2018 cohort. On 25 June 2021, CDFW and PNF released the remaining captive-reared SNYLF housed at the SF Zoo back into the wild. CDFW and PNF staff will continue annual amphibian monitoring to document SNYLF response to reintroductions and BK removal.



Figure 1. Bucks Lake Wilderness, Plumas County, CA. The areas discussed in this memorandum are circled.



Figure 2. Focal areas for Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) management by California Department of Fish and Wildlife (CDFW) and Plumas National Forest (PNF) in Bucks Lake Wilderness, Plumas County, CA. The orange area highlights the Mount Pleasant Native Species Reserve (NSR), and the purple area highlights the Gold Lake NSR. CDFW and PNF biologists from the Mount Hough Ranger District regularly monitor the SNYLF populations in both areas. Numbers displayed are CDFW Site IDs.

ENVIRONMENTAL SETTING

Bucks Lake Wilderness is located in western Plumas County, south of state route 70 and north of Bucks Lake Reservoir. The Pacific Crest Trail bisects the wilderness from north to south, and local elevations range from around 2,400 feet (730 meters [m]) above mean sea level near the northern border along the Feather River, to 7,067 feet (2,154 m) at the summit of Mount Pleasant. CDFW staff initially observed SNYLF populations in Bucks Lake Wilderness while conducting baseline surveys in 2003 and 2004. Surveys conducted in the intervening years have identified two SNYLF populations: one south of Silver Lake in the Gold Lake area, and another persisting in a small drainage southeast of Mount Pleasant.

INTRODUCTION

Due primarily to the <u>Dixie Fire</u>, CDFW did not conduct SNYLF monitoring in the Gold Lake and Mount Pleasant areas in 2021. However, CDFW and PNF staff returned to Bucks Lake Wilderness for amphibian surveys in 2022. Those interested in previous detailed results from SNYLF population surveys and past non-native fish removal at Gold Lake should <u>consult the</u> <u>previous Bucks Lake Wilderness memorandum</u>, which presents results through summer 2020 (CDFW 2021). This memorandum provides an update on the CDFW, PNF, and SF Zoo collaborative effort to release captive-reared SNYLF adults back to their natal lakes in Bucks Lake Wilderness in 2021, and continued long term monitoring in the Mount Pleasant and Gold Lake areas conducted by CDFW and PNF in 2022.

THREATS

Marginal Habitats

VES data show that the Gold Lake and Mount Pleasant area SNYLF populations are currently reproducing in Rock Lake (**Figure 3**) and Dot Lake (**Figure 4**), both of which are small, isolated, and shallow. Any disturbance, natural or otherwise, that threatens overwintering habitats presents a potential extirpation risk. Among the risks to the population are habitat disturbance by humans, wildfire, possible exposure to severe winter conditions, and desiccation during drought conditions (e.g., the 2012–2015 and 2020–2022 drought periods, which lowered water levels in many small ponds throughout the Sierra Nevada), any one of which could threaten these small SNYLF populations.

Introduced Fish

Gold Lake formerly contained a self-sustaining BK population. However, in 2015, CDFW initiated mechanical fish removal using gill nets. CDFW field staff have not observed BK in Gold Lake since July 2016 (CDFW 2018). During summer 2018, CDFW field staff observed BK in isolated pools along the Gold Lake outlet stream, which CDFW subsequently removed by electrofishing (CDFW 2019). These pools are below the Gold Lake streamflow maintenance dam, which provides a barrier to upstream fish movement. Trout may have been precluding SNYLF from successful breeding and recruitment in Gold Lake, which is the only additional deep-water habitat near the breeding SNYLF site at Rock Lake. Furthermore, Speckled Dace

(*Rhinichthys osculus*; DC-S) are present in Gold Lake and Blixie Lake. Speckled Dace may compete with, or directly harm, smaller life stages of SNYLF (e.g., eggs and larvae). However, little information is available regarding effects of DC-S on SNYLF (for more in-depth discussion of potential interactions between early life stage SNYLF and minnows, please refer to the discussion section of the 2020 Native amphibian restoration and monitoring in Bucks Lake Wilderness memorandum; CDFW 2020).

Disease

All SNYLF populations in Plumas County are chytrid fungus (*Batrachochytrium dendrobatidis*; *Bd*) positive. In 2008, 2010, and 2011, SNYLF captured at Rock Lake, Dot Lake, and Blixie Lake were genetically sampled with epithelial swabs for the presence of *Bd*. Field staff collected 27 swabs, which partner scientists screened for the presence of *Bd* DNA using real-time quantitative polymerase chain reaction (qPCR) analysis. Results for all three years indicated very light to moderate *Bd* zoospore loads.

PNF staff consistently collect epithelial swabs on a subset of SNYLF in Bucks Lake Wilderness to monitoring *Bd* status in these populations. For example, in 2018, PNF analyzed 20 additional swabs collected from SNYLF at Dot Lake (n = 14) and Rock Lake (n = 6). Most of the swabbed frogs at Dot Lake were recent metamorphs, which are typically the life stage most susceptible to *Bd* (Rachowicz et al. 2006), whereas four of the six swabs from Rock Lake were collected from adults. Results from the swabs collected in 2018 indicated moderate to high *Bd* zoospore loads. However, high *Bd* loads are typical for recent SNYLF metamorphs, even in populations persisting with *Bd* (Ellison et al. 2019; R. Knapp, pers. comm.). In 2020, CDFW staff collected one swab from an adult frog in the Mount Pleasant drainage, which partner scientists analyzed in fall 2020. Results indicated a light *Bd* load.

CDFW and PNF staff have consistently observed low numbers of dead SNYLF of various life stages at Dot Lake, including during VES in 2004, 2005, 2010, 2015, 2017, 2018, and 2022 (observations during the latter two years were by PNF staff). These observations suggest that there may be consistent *Bd*-induced mortality in this population. CDFW and PNF staff have not observed any recent signs of *Bd* epizootic events in the Gold Lake or Mount Pleasant areas. However, VES results suggest that the Mount Pleasant population may have experienced a major die-off during a *Bd*-induced epizootic event sometime between 2005 and 2008 (CDFW 2019). A similar die-off may have occurred around the same time in the Gold Lake area, but VES do not suggest as pronounced a decline as the one suggested by Mount Pleasant observational data. The Gold Lake and Mount Pleasant SNYLF populations may have not been previously exposed to *Bd* before these potential outbreaks.

Loss of Genetic Diversity

Like many SNYLF populations at the northern extend of the species range, the Bucks Lake Wilderness SNYLF populations are small, isolated, relegated to marginal habitats, and face

potential threats from multiple land uses (MYLF ITT 2018). The northern Sierra Nevada also includes some of the lowest elevation SNYLF populations in the species' range. Additionally, populations in Plumas County are widely separated from one another, which limits gene flow between populations and increases risk for local extirpation. This isolation can lead to factors such as inbreeding depression, genetic drift, fixation of deleterious alleles, and loss of genetic diversity, all of which are population genetic factors exacerbated in small populations (Frankham et al. 2009).

SNYLF CAPTIVE REARING AND RELEASE: 2021 Update

The consistent low number of SNYLF detections in the Mount Pleasant and Gold Lake areas has been concerning to both CDFW and PNF. During VES in August 2018, CDFW field staff observed very few SNYLF in both the Mount Pleasant and Gold Lake areas. Although observer bias and variation in survey conditions can affect the number of SNYLF detected during any given VES, the low numbers were troubling, especially when observed in already threatened populations. In response to these observations and long-term trends, PNF personnel, in collaboration with CDFW, the U.S. Fish and Wildlife Service (USFWS), and SF Zoo, undertook a collection of early life stage SNYLF from Rock Lake (Site ID 12069; **Figure 3**) and Dot Lake (Site ID 12052; **Figure 4**) for captive rearing (a.k.a., "headstarting") at the SF Zoo. The Strategy highlights captive rearing as one of the primary actions to restore SNYLF populations (MYLF ITT 2018, pgs. 17–19). CDFW also mentions the potential for translocations (which are one of the methods, along with captive rearing, broadly considered under "Reintroductions" in the Strategy) in the Mount Pleasant area in the Bucks Lake Wilderness ABMP (CDFW 2015, pg. 20).



Figure 3. Rock Lake (Site ID 12069) in August 2022, looking northwest. (CDFW)



Figure 4. Dot Lake (Site ID 12052) in August 2022, looking north. (CDFW)

During VES in early September 2018 (the purpose of which was locating early life stage SNYLF for captive rearing at the SF Zoo), PNF staff observed more SNYLF than CDFW had detected in August 2018. These early September 2018 surveys by PNF were fortunately timed because they coincided with the emergence of very recently metamorphosed SNYLF, which had likely not been available for detection during surveys the previous month. PNF staff collected 60 recently metamorphosed SNYLF at Dot Lake, plus four early life stage SNYLF at Rock Lake. On 6 September 2018, PNF staff transported all 64 collected SNYLF to the SF Zoo. Once at the SF Zoo, staff treated the young frogs with an antifungal drug called itraconazole, which removes amphibian chytrid fungal infection (for more details, see the *Disease* section above). The SNYLF were raised to maturity at the SF Zoo and 42 captive-reared frogs were mature enough for release back into Bucks Lake Wilderness in summer 2019. On 8 October 2019, PNF staff collected SNYLF from Dot Lake for captive rearing at the SF Zoo. The SNYLF were raised to maturity at the SF Zoo and 38 captive-reared frogs were mature enough for release back into Bucks Lake wilderness in summer 2020 (PNF 2021).

CDFW and PNF staff release all captive-reared SNYLF back into the sites from which young frogs were originally collected. However, since staff detected nearly all early life stage SNYLF at Dot Lake, most captive-reared SNYLF that staff released back into Rock Lake were originally collected from Dot Lake. Given that Dot Lake and Rock Lake are close together (< 2.5 km), contain very similar habitat, and both contain *Bd*-positive SNYLF populations, moving captive-reared frogs from Dot Lake to Rock Lake poses negligible risk to the resident SNYLF populations or local environment. Additionally, recent population genetic analyses indicate that little genetic substructuring exists between SNYLF populations located in such close proximity

(Rothstein et al. 2020; A. Byrne et al., *in prep*). Therefore, CDFW, PNF, USFWS, and partner scientists have determined that movements like those undertaken between Dot Lake and Rock Lake are an appropriate conservation measure for SNYLF (MYLF ITT 2018).

On 28 June 2019, CDFW and PNF biologists released 40 young adult SNYLF (18 females and 22 males) back into the Bucks Lake Wilderness after being cleared of *Bd* infection and raised to maturity at the SF Zoo. These 40 individuals were a subset of the 64 individuals collected by PNF staff on 6 September 2018. CDFW staff picked up 42 SNYLF from SF Zoo staff on the morning of 28 June 2019 and transported the frogs to the PNF Mount Hough Ranger Station. There, staff split the frogs into two groups, each containing 9 females and 12 males. CDFW and PNF staff took one group to Rock Lake and one group to Dot Lake for release. Before release, staff scanned and recorded each frog's passive integrated transponder (PIT) tag (a tiny, glasscoated chip that provides a unique identifier that biologists insert under the frog's skin). Staff also recorded snout-urostyle length (SUL) and weight measurements of each frog (Figure 5). Staff successfully reintroduced 21 frogs into Dot Lake and 19 frogs to Rock Lake. Two male frogs intended for release into Rock Lake perished after exposure to excessive heat from briefly sitting in sunlight while in transport containers. The mortality incident was very unfortunate, but an occurrence that will be easily preventable in the future. CDFW staff informed the U.S. Fish and Wildlife Service of the mortality incident immediately after returning from the field. The 40 remaining frogs were all in good condition and returned to their original habitat.

On 19 June 2020, CDFW and PNF biologists used the methods described above to release another 38 adult SNYLF (23 females, 12 males, and 3 of unknown sex) back into Bucks Lake Wilderness. SNYLF released by staff in 2020 included the remaining frogs from the cohort originally collected in 2018 (n = 20), plus a subset of frogs that PNF staff collected as late stage tadpoles in October 2019 (i.e., those large enough for release in June 2020; n = 18). Of the 38 frogs, staff released half (n = 19) into Dot Lake and the other half into Rock Lake. When dividing up SNYLF into the two release groups, CDFW and PNF staff allotted 13 females/unknowns and 6 males to each site. All 38 SNYLF were behaving normally and appeared to be in excellent body condition during the release.

On 25 June 2021, CDFW and PNF biologists used the aforementioned methods to release the remaining cohort of 22 adult SNYLF (11 females and 11 males) back into Bucks Lake Wilderness. SNYLF released by staff in 2021 included all remaining frogs from the cohort originally collected in 2019 as late stage tadpoles. Of the 22 frogs, staff released half (n = 11) into Dot Lake and the other half into Rock Lake (**Figure 6**). When dividing up SNYLF into the two release groups, CDFW and PNF staff allotted 6 females and 5 males to Dot Lake, and 5 females and 6 males to Rock Lake. All SNYLF were behaving normally during release, and all but one appeared to be in excellent body condition. One male was significantly smaller than all other individuals (i.e., less than half the weight of the next smallest frog).

As of June 2021, SF Zoo no longer houses any SNYLF from Bucks Lake Wilderness. However, the SF Zoo may receive additional SNYLF for future captive-rearing efforts, depending on the results of future surveys in Bucks Lake Wilderness and management goals of CDFW, PNF, and USFWS.



Figure 5. A Plumas National Forest biologist collecting morphological data from a captive reared adult Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) at Rock Lake on 25 June 2021. (CDFW)



Figure 6. A CDFW staff member releases a captive reared adult Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) into Rock Lake on 25 June 2021. (CDFW)

SNYLF MONITORING: 2022 Update

VES in the Gold Lake area

CDFW performed the baseline VES in the Gold Lake area in 2004, during which staff encountered a small breeding SNYLF population. Nineteen years of periodic monitoring data suggest that this population is persisting at low numbers (**Figure 8**). Most signs of breeding, including observations of egg masses, larvae, and recent metamorphic SNYLF, have occurred at Rock Lake (Site ID 12069; **Figure 9**). SNYLF detections of all life stages have remained relatively consistent, albeit low, since 2010. Observer bias, variation in survey conditions, and the low number of detections all make deriving trends difficult. Additionally, recent population augmentation (<u>discussed above</u>) may be providing benefits to the Bucks Lake Wilderness SNYLF populations, which may partially account for higher frog detections in recent years. Additionally, the more recent inclusion of capture-mark-recapture (CMR) techniques (<u>discussed</u> <u>below</u>) has allowed CDFW and PNF to more accurately monitor population demographics and trends.

In 2022, CDFW surveyed the Gold Lake area once, and PNF staff visited the area on several additional occasions. Although this report focuses on the joint CDFW-PNF surveys in late August 2022, the survey results presented below include observations by PNF staff during additional visits in which CDFW did not participate. The joint CDFW-PNF survey occurred 22–24 August 2022 and included Gold Lake (Site ID 12067; **Figure 7**), Rock Lake (Site ID 12069; **Figure 3**), Mud Lake (Site ID 12062), Site ID 12064, and surrounding stream segments (Site IDs 50140, 50142, 52705, and 52709; **Figure 9**). In 2022, CDFW and PNF staff observed a total of 13 adult and 14 subadult SNYLF in the Gold Lake area. As part of the captive rearing and release, PNF partners surveyed portions of the Gold Lake and Mount Pleasant areas on several additional occasions in summer 2022. PNF and CDFW staff observed SNYLF egg masses and one larva in Rock Lake in May 2022, and post-metamorphic life stages (subadults and adults) from late spring through early fall.

For several years, PNF and CDFW field staff have been marking newly captured adult SNYLF (≥40 mm SUL) in the Gold Lake and Mount Pleasant areas with PIT tags, which provide a unique identifier for keeping track of individuals. Staff first scan each captured adult with a PIT tag reader, which displays a unique identification code when a PIT tag is detected, to determine if the frog is marked (i.e., a recapture). Field staff then record the global positioning system (GPS) point, SUL, weight, and the frog's sex. If the adult is a recapture, staff release the frog without further processing. For new adult captures, in addition to the data collected above, staff insert an 8 x 1.4 mm PIT tag under the dorsal skin (using methods recommended by McAllister et al. 2004), scan and record the PIT tag number, and release the frog at the point of capture. PIT tags can be used to monitor demographic trends in the population using CMR analyses (Williams et al. 2001). Additional details on CMR results from Bucks Lake Wilderness are discussed below in the <u>SNYLF CAPTURE-MARK-RECAPTURE</u> section.

Both CDFW and PNF will continue surveying the area at least once each year, keeping records on the location and identity of recaptured adult SNYLF, and PIT tag any newly captured adults. These data will help CDFW better understand the status and trends of the Bucks Lake Wilderness SNYLF populations.



Figure 7. Gold Lake (Site ID 12067) in August 2022. (CDFW)



Figure 8. Histogram of Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) detections during visual encounter surveys (VES) by life stage and year in the Gold Lake area. Survey data includes SNYLF detections at Rock Lake (Site ID 12069), Gold Lake (Site ID 12067), and the associated stream segments (Site IDs 50140, 50141, 50142, 52705, and 52709). California Department of Fish and Wildlife (CDFW) and Plumas National Forest (PNF) field staff have detected most SNYLF at Rock Lake and in the outlet stream close to Rock Lake (i.e., upstream end of Site ID 50142). SNYLF detections at Gold Lake and the connected streams have been composed primarily of adults. If more than one VES was conducted in a given year, the data shown are from the survey day resulting in the highest number of SNYLF detections. Survey effort, in terms of Site IDs visited and the number of visits to each Site ID, varied between survey years.

⁺Beginning in 2018, adult frog totals are based on passive integrated transponder (PIT) tag identifications by CDFW and PNF staff (i.e., captured adults are either known recaptures or new individuals that received a PIT tag once caught). Therefore, adult totals from 2018 to 2022 reflect all unique, tagged individuals captured by PNF and CDFW. CDFW did not add stream segment 52705 (the segment below the confluence of the Rock and Gold Lake outlets) until 2018. Additionally, CDFW added a Site ID for an old mining trench (Site ID 52709).

++In 2019, only PNF staff surveyed Rock Lake. *Total does not include young adult SNYLF released at Rock Lake in 2019 (n = 19).*

*Total does not include young adult SNYLF released at Rock Lake in 2020 (n = 19).

[End of figure caption.]



Figure 9. Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) observations and surface water status during visual encounter surveys (VES) in the Gold Lake area during late August 2022. SNYLF letter codes in the legend, which indicate the life stage(s) observed during VES, are as follows: "A" = adults, "SA" = subadults, "L" = larvae, and "EM" = egg masses. Rock Lake is the only known SNYLF breeding location in the drainage. California Department of Fish and Wildlife (CDFW) and Plumas National Forest (PNF) field staff consistently observe post-metamorphic SNYLF in the drainages downstream of Rock Lake and Gold Lake. In 2018 and 2019, PNF staff collected early life stage SNYLF from Rock Lake (Site ID 12069, in yellow) and Dot Lake (Site ID 12052, which is northwest of Rock Lake; **Figure 11** below). Following each collection, biologists transported the SNYLF to the San Francisco Zoo for captive rearing. In 2019, 2020, and 2021, PNF and CDFW staff released portions of the captive-reared SNYLF (now adults) back into Rock Lake and Dot Lake. CDFW has designated the area displayed as a Native Species Reserve (NSR) in the Aquatic Biodiversity Management Plan for the Bucks Lake Wilderness Management Unit (CDFW 2015).

VES in the Mount Pleasant area

CDFW performed the baseline VES in the Mount Pleasant area in 2004, during which staff encountered a small breeding SNYLF population. Nineteen years of periodic monitoring data suggest that this population is persisting at low numbers (**Figure 10**). Most signs of breeding, including observations of egg masses, larvae, and recent metamorphic SNYLF, have occurred in Dot Lake (Site ID 12052; **Figure 11**). Similarly to the Gold Lake area SNYLF population, detections have remained relatively consistent, albeit low, since 2010. In part due to these low detections, PNF and CDFW staff have undertaken the same adult SNYLF PIT-tagging methods described above in the <u>VES IN THE GOLD LAKE AREA</u> section.

In 2022, CDFW surveyed the Mount Pleasant area once, and PNF staff visited the area on several additional occasions. Although this report focuses on the joint CDFW-PNF surveys in late August 2022, the survey results presented below include observations by PNF staff during additional visits in which CDFW did not participate. The joint CDFW-PNF survey occurred 22–24 August 2022 and included Dot Lake (**Figure 12**), Blixie Lake (Site ID 12049; **Figure 13**), the stream segment that connects Dot and Blixie (Site ID 50157), Coco Lake (Site ID 52583; **Figure 14**), Site ID 12057 (**Figure 15**), upper Mill Creek (Site ID 52918; **Figures 16 and 17**), plus all other sites shown in **Figure 11**.

In 2022, CDFW and PNF staff observed a total of nine adults, 87 subadults (the highest subadult count during a single survey of the site), eight larvae, and 18 egg masses in the Mount Pleasant area. Staff detected all larvae and egg masses at Dot Lake, and all subadults at Dot and Blixie Lakes. In addition to traditional VES counts, PNF staff examined chin spot patterns to uniquely identify subadult individuals captured at Dot Lake (Marlow et al. 2016). Based on these chin spot pattern recognition techniques, PNF detected approximately 230 subadult individuals at Dot Lake when combining several visits from August to October 2022 (PNF 2023).

As part of the captive rearing and release (<u>discussed above</u>), PNF partners surveyed portions of the Gold Lake and Mount Pleasant areas on several additional occasions in summer 2022. PNF staff observed SNYLF egg masses in Dot Lake in early June 2022, and all other life stages (tadpoles, subadults, and adults) from late spring through early fall. Additionally, PNF staff detected a large dead adult SNYLF lying on the bottom of Dot Lake in May 2022, and one dead subadult on 23 August 2022.



Figure 10. Histogram of Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) detections during visual encounter surveys (VES) by life stage and year in the Mount Pleasant area. Survey data includes SNYLF detections at Blixie Lake (Site ID 12049), Dot Lake (Site ID 12052), Coco Lake⁺ (Site ID 52583), nearby ponds (the largest of which are Site IDs 12056 and 12057), and the associated stream segments (Site IDs 50157 and 50158). In 2022, CDFW added Site IDs 52918 (upper Mill Creek) and 53460 (a marshy area that feeds upper Mill Creek). Survey effort, in terms of Site IDs visited and the number of visits to each Site ID, varied between survey years. Tree stars] indicate outliers above the y-axis scale (see table).

The high subadult counts during VES in 2008, 2010, and 2018 may be due to survey timing coinciding with late stage SNYLF tadpoles metamorphosing into young subadult frogs. Only a small subset of early life stages typically survive to sexual maturity and recently metamorphosed frogs are very susceptible to mortality from chytridiomycosis, the disease caused by the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*). These factors could explain why the VES detections were low in many other years.

⁺Beginning in 2018, adult frog totals are based on passive integrated transponder (PIT) tag identifications by CDFW and PNF staff (i.e., captured adults are either known recaptures or new individuals that received a PIT tag once caught). Therefore, adult totals from 2018 to 2022 reflect all unique, tagged individuals captured by PNF and CDFW. CDFW did not add Coco Lake, which Plumas National Forest staff have occasionally monitored, until 2018.

* Total does not include young adult SNYLF released at Dot Lake in 2020 (n = 19).

‡In 2022, PNF staff collected chin spot pattern photos of all subadult SNYLF captured. Based on analysis of these chin spot photos, staff observed approximately 230 unique subadults at Dot Lake (Site ID 12052) during several visits to the site between August and October 2022. However, for better comparability with previous years, the total shown in the histogram reflects the highest number of subadults observed during any single VES in 2022. [End of figure caption.]



Figure 11. Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) observations and surface water status in the Mount Pleasant drainage during late August 2022. California Department of Fish and Wildlife (CDFW) updated several existing GIS polygons to more accurately reflect the composition of the depicted waterbodies. SNYLF letter codes in the legend, which indicate the life stage(s) observed during VES, are as follows: "A" = adults, "SA" = subadults, "L" = larvae, and "EM" = egg masses. CDFW staff observed adult and subadult SNYLF at Dot Lake and Blixie Lake. Plumas National Forest (PNF) staff observed the SNYLF larvae and egg masses at Dot Lake. In 2018 and 2019, PNF staff collected early life stage SNYLF from Dot Lake (Site ID 12052, in yellow) and Rock Lake (Site ID 12069, which is southeast of Dot Lake; **Figure 9** above). Following each collection, biologists transported the SNYLF to the San Francisco Zoo for captive rearing. In 2019, 2020, and 2021, PNF and CDFW staff released portions of the captive-reared SNYLF (now adults) back into Dot Lake and Rock Lake. CDFW has designated the area displayed as a Native Species Reserve (NSR) in the Aquatic Biodiversity Management Plan for the Bucks Lake Wilderness Management Unit (CDFW 2015, pg. 28).



Figure 12. Dot Lake (Site ID 12052) on 23 August 2022, looking northeast. On 25 June 2021, Plumas National Forest (PNF) biologists released 11 captive-reared Sierra Nevada Yellow-legged Frogs (*Rana sierrae*; SNYLF) adults at the site. Most signs of Sierra Nevada SNYLF breeding (including observations of egg masses, larvae, and recent metamorphic SNYLF) in the Mount Pleasant area have occurred at Dot Lake. Large areas of forest burned during the 2021 Dixie Fire are visible in the background.



Figure 13. Blixie Lake (Site ID 12049) on 23 August 2022, looking southwest. The burn scar visible on the left is from the Dixie Fire, which burned through the area in 2021. (CDFW)



Figure 14. Coco Lake (Site ID 52583) on 23 August 2022.



Figure 15. Site ID 12057 in August 2022. Plumas National Forest (PNF) staff detected an adult Sierra Nevada Yellow-legged Frog (*Rana sierrae*) at this location in 2022. The burn scar visible around the pond is from the Dixie Fire, which burned through the area in 2021. (CDFW)



Figure 16. Site ID 52918 in August 2022. California Department of Fish and Wildlife (CDFW) staff detected an adult Sierra Nevada Yellow-legged Frog (*Rana sierrae*) at this site in August 2022. The burn scar visible in the background is from the Dixie Fire, which burned through the area in 2021. (CDFW)



Figure 17. Looking north from the downstream end of Site ID 52918 in August 2022. This site includes a portion of the Mill Creek headwaters. A series of steep cascades over bedrock are located just below the downstream end of the survey reach. During surveys in late August 2022, very little water remained in this portion of Mill Creek. California Department of Fish and Wildlife (CDFW) staff detected an adult Sierra Nevada Yellow-legged Frog (*Rana sierrae*) at this site in August 2022. Extensive areas of forest burned during the Dixie Fire are visible in the background. (CDFW)

SNYLF CAPTURE-MARK-RECAPTURE

U.S. Forest Service (USFS) personnel have been PIT-tagging adult SNYLF (**Figure 18**) for monitoring purposes in Bucks Lake wilderness since 2015. The initial tagging efforts were subsequently halted due to changes in research direction and personnel. More recent PIT-tagging work undertaken by current PNF and CDFW staff, beginning in 2017 and expanding in 2018, seeks to PIT tag all newly captured (untagged) adults in the Gold Lake and Mount Pleasant areas. Data presented here only include 2018 onward, which is the period during which there has been more consistent surveying and tagging effort. PIT-tagging allows CDFW and PNF to more accurately keep track of SNYLF population demographics through time by using CMR analyses. Additionally, PIT-tagging allows biologists to learn more about the potential benefits of the recent SNYLF headstarting work, <u>discussed in the previous section</u>.

A brief summary of PIT-tagging effort by PNF and CDFW during summer and early fall from 2018 to 2022 is below (**Table 1**).

Overall, among the SNYLF that have been marked and released since 2018, recapture rates have been low, particularly among zoo-reared frogs. For example, in 2019, staff only recaptured one zoo-reared adult (a frog observed in Dot Lake) from among the 40 frogs that had been released in June. Similarly, of all adults released in 2019, staff only observed one in 2020 (a frog observed in Rock Lake in late June 2020). The same result occurred during surveys in 2021, when staff detected one frog released in 2019 (a frog observed in Rock Lake, which was a different individual than the recaptured zoo-reared frog observed in 2020). However, same season recaptures were more common. Of the 38 zoo-reared individuals release in June 2020, staff recaptured seven at least once later in the same summer and early fall (three in Dot Lake and four in Rock Lake; **Table 1**). Of the 22 zoo-reared individuals released in 2021, staff recaptured six at least once later in the summer and early fall, all at Rock Lake. None of the zoo-reared frogs released into Dot Lake in June 2021 were seen later that same year. However, the Dixie Fire limited opportunities to revisit the site to look for frogs.

In total, from 2019 to 2021, CDFW and PNF staff released 100 zoo-reared frogs back into Rock and Dot Lakes. Of those 100 frogs, staff have detected 16 individuals during follow-up surveys of the Gold Lake and Mount Pleasant areas. Only two of the 16 recaptured zoo-reared frogs have been detected in a subsequent year (i.e., 14 of the 16 zoo-reared recaptures were only detected later in the same summer and/or early fall of year they were released). In 2022, CDFW and PNF staff did not detect any zoo-reared frogs.

Although recaptures have so far been highly limited among the zoo-reared frogs, CMR work with SNYLF in other locations (e.g., Mossy Pond in Tahoe National Forest and Desolation Valley in Eldorado National Forest; CDFW unpubl. data) has demonstrated that SNYLF will often go undetected during surveys one year and later be available for detection in following years. Such an example is also provided among wild frogs from Bucks Lake Wilderness. For example, of the seven wild frogs PIT-tagged in the Mount Pleasant area in 2018, five were not observed during surveys in 2019, but were recaptured during surveys in 2020. Of the remaining two Mount Pleasant area SNYLF marked in 2018, staff observed one in both 2019 and 2020, and staff have not observed the other since 2018. These data demonstrate that more zoo-reared SNYLF may still be extant on the landscape and detected during future surveys in Bucks Lake Wilderness.

Wild frog recapture events have been more common than those staff have currently observed among the zoo-reared frogs (**Table 1**). Since 2015, USFS and CDFW staff have PIT-tagged 130 wild SNYLF in the Mount Pleasant and Gold Lake areas. Of those 130 wild-caught and PITtagged individuals, staff have recaptured 82 at least once (i.e., 48 wild frogs have not been recaptured since the initial detection). Of those 82 recaptured wild frogs, 29 are known to have survived overwinter (i.e., 29 wild PIT-tagged frogs have been recaptured after at least one winter has passed since staff initially detected the frog). So far, the remaining 53 recaptured wild frogs have only been observed within the same season during which they were originally captured by CDFW or PNF staff.

Continued CMR work is needed to obtain a better understanding of survival among the captive-reared and wild-caught SNYLF. This work will also provide useful information on abundance, growth, and movement of SNYLF within the Gold Lake and Mount Pleasant areas. Therefore, CDFW and PNF staff plan to continue this work annually.



Figure 18. An adult Sierra Nevada Yellow-legged Frog (*Rana sierrae*) at Rock Lake (Site ID 12069) in August 2022. (CDFW)

Table 1. Summary of released and wild caught Sierra Nevada Yellow-legged Frogs (*Rana sierrae*; SNYLF) PIT-tagged in the Mount Pleasant and Gold Lake areas of Bucks Lake Wilderness between 2018 and 2022. Data are divided into SNYLF captive-reared at the San Francisco Zoo (Zoo Frogs), and wild caught SNYLF (Wild Frogs). The "# Same summer recaps" tallies include the total number of SNYLF individuals, of those individuals that were released (zoo frogs) or newly tagged (wild frogs) in the given year, that were recaptured during the same summer. The remaining two columns of data show the number of recaptured SNYLF individuals that were originally marked in a previous year. Therefore, individuals from the last two columns 1) survived at least one winter, 2) were available for detection during survey periods in the subsequent year(s), and 3) were detected by observers.

*Note: the number contained in each cell is the number of <u>unique SNYLF individuals</u> within each category for that year, <u>not</u> the number of capture <u>events</u> within that category (i.e., some frogs were recaptured more than once in the same summer, but those repeat recapture events are excluded from this table).

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‡Although CDFW staff assisted with the release of zoo-reared SNYLF in June 2021, staff from Plumas National Forest conducted all subsequent amphibian surveys in 2021. Surveys in 2021 were limited due to the Dixie Fire.

⁺One recapture from the Mt. Pleasant drainage that had been tagged in a previous year was a mortality (i.e., the frog was found dead underwater during visual encounter surveys).

DISCUSSION

As of 2022, relatively few of the zoo-reared SNYLF have been recaptured during follow-up surveys in the Gold Lake and Mount Pleasant areas. In total, since 2019, PNF and CDFW staff have recaptured 10 captive-reared SNYLF in the Mount Pleasant area and 6 captive-reared SNYLF in the Gold Lake area, respectively. These 16 recapture events are all unique frogs: as of spring 2022, staff have not recaptured any captive-reared SNYLF more than once. Additionally, most captive-reared SNYLF recaptures have been detected by staff within approximately two weeks post-release. However, a small number of captive-reared SNYLF recaptures have occurred months later, or the following year. Additionally, although there have been no notable recruitment pulses in the Gold Lake and Mount Pleasant SNYLF populations in response to captive-rearing and release, staff have still been detecting signs of breeding in late spring during the past few years. For example, staff detected SNYLF egg masses at Rock Lake and Dot Lake in late spring during each of the past four years (2019–2022).

Based on similar efforts with SNYLF in other parts of the Sierra Nevada, the slow start to population augmentation is not surprising. SNYLF are long-lived and tadpoles often take at least two years to metamorphose. Under the right conditions, captive-rearing work has demonstrated that SNYLF can metamorphose in one year, but the timeline for SNYLF development in the captive environment likely represents an upper limit on growth rates. Any individual component of those conditions (e.g., controlled climate, constant supply of high quality food, lack of predators, active disease mitigation, etc.), let alone all factors, is often not present in the wild. However, SNYLF may naturally experience rapid development in relatively low elevation populations like Bucks Lake Wilderness (~6,000 ft. [~1,830 m]), particularly during drought periods with long growing seasons and relatively mild winters (e.g., the 2012–2016 drought, and winters 2017–2018 and 2019–2022). Despite some longer warm periods in recent years, most SNYLF in the Bucks Lake Wilderness populations likely overwinter at least once before metamorphosis. Therefore, more time may be needed to observe potential benefits of headstarting.

Population augmentation—in the form of conservation efforts such as headstarting, translocations, or *in situ* rearing—can take years to manifest, or may require many separate augmentations, before there is a detectable increase in abundance (Joseph and Knapp 2018). Delays in population growth can be the result of many factors, including limited recruitment. Low recruitment appears to be a consistent attribute of SNYLF populations in the northern Sierra Nevada, in which many populations are stream-based (Brown et al. 2020). Current SNYLF research in Yosemite, being conducted by University of California, Santa Barbara (UCSB) scientists, suggests that recruitment is often rare and episodic, despite reproduction (T. Smith, pers. comm.). *Bd*-induced mortality, particularly among highly susceptible early life stages, is one factor often leading to low recruitment in SNYLF populations (Rachowicz et al. 2006). However, other factors can also affect recruitment, including severe winters (Bradford 1983,

Joseph and Knapp 2018), snake predation (Jennings et al. 1992, Matthews et al. 2002; T. Smith, pers. comm.), and non-native fish (Knapp and Matthews 2000).

Given the close proximity and hydrologic connection of Gold Lake and Rock Lake, CDFW is not planning to conduct short distance translocations of SNYLF from the Rock Lake drainage to Gold Lake. CDFW and PNF surveys have demonstrated that SNYLF are moving throughout the Gold Lake drainage. For example, one PIT-tagged female was seen in Gold Lake in September 2018, next recaptured in the Gold Lake outlet stream in September 2019, and most recently recaptured in Rock Lake in mid-October 2019. The most recent SNYLF observation at Gold Lake was another adult female observed by PNF in early November 2020. SNYLF will likely continue moving between Gold Lake and Rock Lake via the connected streams. However, as opportunities allow—based on SNYLF population status, funding, SF Zoo capacity, and availability of personnel—CDFW and PNF may collect additional early life stage frogs for future captive rearing efforts to help further augment the Bucks Lake Wilderness SNYLF populations. Such an effort is anticipated for early summer 2023, during which PNF and CDFW staff plan to collect portions of any egg masses found for captive rearing at the SF Zoo.

Finally, portions of Bucks Lake Wilderness were affected by the <u>Dixie Fire</u>, which burned through the area in early August 2021. The landscape surrounding portions of the trail to Gold and Rock Lakes burned at high severity (**Figures 19 and 20**). However, the primary locations that SNYLF occupy in Bucks Lake Wilderness did not burn, or were minimally affected. These areas include Rock Lake (**Figure 21**), Gold Lake (**Figures 22**), and Dot Lake (**Figure 23**). In the Mount Pleasant area, the fire burned a small amount of vegetation around Blixie Lake, but most of the surrounding vegetation was unaffected (**Figure 24**). The areas surrounding primary SNYLF habitat in Bucks Lake Wilderness are rocky and contain sparser vegetation than nearby locations more heavily impacted by the fire. In addition to the dedicated efforts of wildland firefighters, the granitic landscape likely helped prevent the SNYLF habitats from burning severely.



Figure 19. California Department of Fish and Wildlife (CDFW) and Plumas National Forest (PNF) staff hiking to Rock Lake, Bucks Lake Wilderness in late June 2021 for the release of adult Sierra Nevada Yellow-legged Frogs (*Rana sierrae*) that had been captive-reared at the San Francisco Zoo. (CDFW)



Figure 20. The same segment of trail (which leads to Rock and Gold Lakes) shown in **Figure 19**, post-<u>Dixie Fire</u>, on 20 May 2022. (CDFW)



Figure 21. Rock Lake (photo taken from the western shore, looking east) on 20 May 2022. The landscape directly surrounding Rock Lake was not burned in the Dixie Fire. (CDFW)



Figure 22. The landscape surrounding Gold Lake (a small portion of which is visible in center background) on 20 May 2022. Spanish Peak is visible at the upper left of the photograph. The area immediately surrounding Gold Lake was not burned in the Dixie Fire. (CDFW)



Figure 23. Dot Lake on 20 August 2021, soon after the Dixie Fire had burned in the nearby vicinity. However, the fire did not burn the area immediately surrounding Dot Lake. (C. Dillingham, Plumas National Forest, Mt. Hough Ranger District)



Figure 24. Blixie Lake on 20 August 2021, soon after the Dixie Fire had burned in the nearby vicinity. Some vegetation on the northeast side of the lake was burned during the fire. However, areas with thick willow on the southwest side of the lake were not burned. (C. Dillingham, Plumas National Forest, Mt. Hough Ranger District)

CONCLUDING REMARKS

CDFW, in close partnership with PNF, will continue monitoring the Gold Lake and Mount Pleasant area SNYLF populations every year to assess population status (i.e., determine relative abundance, look for signs of continued breeding and recruitment, and assess distribution of SNYLF on the landscape). CDFW and PNF will also continue PIT-tagging adult SNYLF to obtain more detailed abundance and survival data on wild and SF Zoo-reared frogs. Additionally, staff will focus on locating additional SNYLF that may be moving out of Rock Lake and into Gold Lake or its tributaries. These efforts will require thorough VES in challenging terrain, such as stream channels with dense willow growth and steep, rocky substrates with abundant refugia for SNYLF.

CDFW may also work with PNF and zoo partners to collect additional early life stage SNYLF for future captive rearing efforts. Success of captive rearing efforts may be determined in several ways, including staff observing, 1) released adult SNYLF persisting after the first winter following release, 2) additional signs of breeding (higher counts of egg masses, tadpoles, and/or recently metamorphosed frogs), and 3) evidence of new recruitment into the adult population. Augmenting these populations through captive rearing and enhancing deep-water habitat through the successful removal of introduced trout has increased the odds of long-term SNYLF persistence in Bucks Lake Wilderness.

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