## State of California Department of Fish and Wildlife

# Memorandum

## Date: 17 April 2020

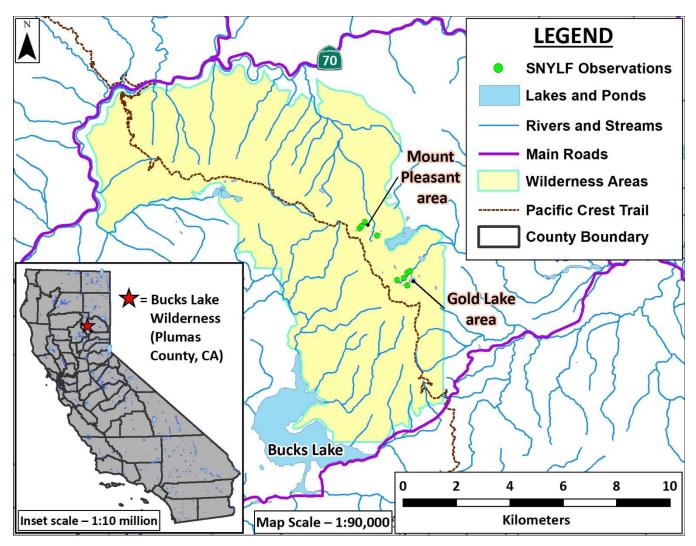
- To: Sarah Mussulman, Senior Environmental Scientist; Sierra District Supervisor; North Central Region Fisheries
- From: Isaac Chellman, Environmental Scientist; John Imperato, Scientific Aide; High Mountain Lakes; North Central Region Fisheries
- Cc: Region 2 Fish Files
- Ec: CDFW Document Library

## Subject: Native amphibian restoration and monitoring in Bucks Lake Wilderness.

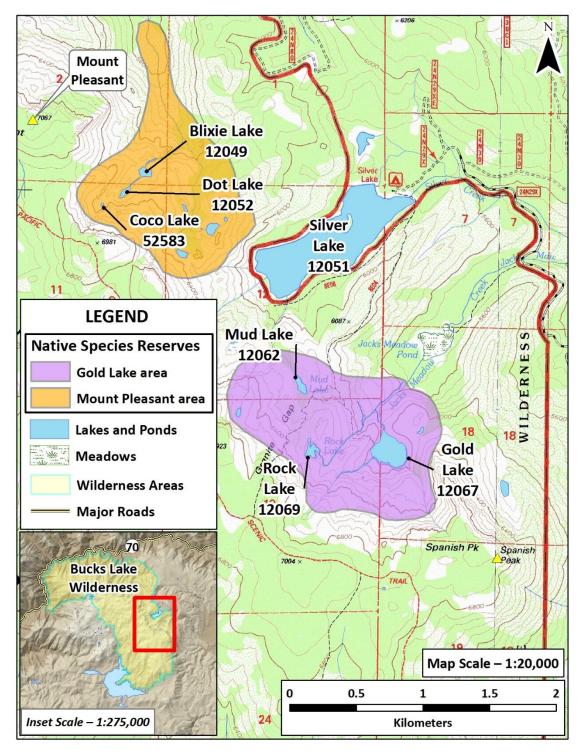
- Gold Lake area Rana sierrae monitoring.
- Rana sierrae captive rearing and release.

California Department of Fish and Wildlife (CDFW) is focusing on two drainages surrounding Silver Lake in Bucks Lake Wilderness to benefit state threatened and federally endangered Sierra Nevada Yellow-legged Frogs (*Rana sierrae*, SNYLF; **Figure 1**). Gold Lake is a site from which CDFW staff removed introduced Brook Trout (*Salvelinus fontinalis*; BK) to benefit SNYLF. The Gold Lake area includes Gold Lake, Rock Lake, and tributaries. The Mount Pleasant area, which includes several small ponds, is located about 2 kilometers (km) northwest of the Gold Lake area (**Figure 2**). 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 2004–2019 suggest 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 are part of a genetic clade at high risk of extirpation (Vredenburg et al. 2007). 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 Zoo. 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. Additionally, 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. Green dots showing *Rana sierrae* (SNYLF) sites include positive detections by CDFW staff during visual encounter surveys (VES) between 2004 and 2018.



**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

Gold Lake (Site ID 12067; **Figure 3**) represents the only fish removal project to benefit SNYLF undertaken by CDFW in Plumas County. For detailed background on aquatic ecosystem management in the Bucks Lake Wilderness (**Figure 1**), <u>see the ABMP</u> (CDFW 2015). The ABMP identifies Gold Lake as one of the few, and possibly only, feasible habitat restoration opportunities available in Plumas and Sierra Counties (CDFW 2015). CDFW selected Gold Lake because 1) the site exhibits the physical habitat characteristics beneficial to state threatened and federally endangered SNYLF, 2) CDFW environmental scientists determined that BK eradication using mechanical methods is feasible, and 3) there is an extant SNYLF population in close proximity to Gold Lake.



**Figure 3**. Gold lake (Site ID 12067) on 3 July 2019. Gold Lake is the site from which CDFW removed non-native Brook Trout (*Salvelinus fontinalis*; BK) from 2015–2018. (CDFW).

Fish removal was complicated by Gold Lake being a popular recreational destination easily accessible from Silver Lake. Members of the local community strongly opposed the action. However, CDFW is mandated to conserve threatened and endangered species (Fish & G. Code, § 2050 et seq.), and Gold Lake offered the only feasible option known in Plumas County to improve habitat for an extant SNYLF population by mechanically removing non-native fish. Additionally, several other lakes are available for trout fishing in the vicinity, including Silver Lake, Lost Lake, Lower Three Lakes, Bucks Lake, Lower Bucks Lake, and Grizzly Forebay. There are numerous other fishing opportunities in Plumas County, including the Mount Hope area of Lassen National Forest to the west of Bucks Lake Wilderness, and approximately 20 different locations in Plumas County that are regularly stocked by CDFW.

Those interested in detailed background about fish removal at Gold Lake may consult the 2018 memorandum "<u>Gold Lake fish removal and Rock Lake *Rana sierrae* monitoring</u>" (CDFW 2018). Final BK removal results are in the 2019 memorandum "<u>Gold Lake and Mount Pleasant areas –</u> *Rana sierrae* monitoring; Gold Lake fish removal update" (CDFW 2019).

During a 2004 baseline visual encounter survey (VES) for SNYLF at Rock Lake (Site ID 12069; **Figure 4**), CDFW observed eight adults, seven juveniles, and 21 larvae – the only sign of breeding in the drainage. An 8-hour gill net set at Rock Lake yielded no fish. However, a 9-hour net set at nearby Gold Lake, which includes the only deep-water lake habitat in Bucks Lake Wilderness (29 m maximum recorded depth), captured eight BK and indicated a self-sustaining fishery. Both Gold and Rock Lakes are beautiful northern Sierra Nevada lakes accessible via a short hike from Silver Lake trailhead, all factors that make for heavy recreational use of the area. Based on extensive surveys during the summers of 2002–2005, Rock Lake contains one of only three documented lake-based SNYLF populations in Plumas County. In accordance with the CDFW mission statement to balance native species diversity and recreational opportunity, Gold Lake (**Figure 4**) was identified as a potential restoration site for SNYLF.

The Mount Pleasant SNYLF population is another of only a few populations remaining at the northern extent of the species' range. Additionally, this population is one of even fewer lakebased populations in Plumas County. In 2004, CDFW conducted a baseline VES, during which staff observed adult, subadult, and larval SNYLF at Dot Lake (Site ID 12052; **Figure 2**), plus adult SNYLF at Blixie Lake (Site ID 12049; **Figure 2**). In 2013, PNF staff surveyed the Mount Pleasant area and observed adult frogs in all wetted habitat, including eight adult SNYLF in Coco Lake (Site ID 52583; **Figure 2**; CDFW 2015).

#### THREATS

#### Marginal Habitats

VES data show that the Gold Lake and Mount Pleasant area SNYLF populations are currently reproducing in Rock Lake and Dot Lake, both of which are small, isolated, and shallow (**Figure 5**; CDFW 2019). 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, possible exposure to severe winter conditions, and desiccation from drought conditions (e.g., the 2012–2015 drought, which lowered water levels in many small ponds throughout the Sierra Nevada), any one of which could eliminate 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 (see <u>DISCUSSION</u> below).

#### 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. Twenty seven swabs were collected and 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. 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.). 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, and 2018 (the latter observation 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 (Figure 5). The Gold Lake and Mount Pleasant SNYLF populations may have not been previously exposed to Bd before these potential outbreaks.

#### Loss of Genetic Diversity

The Bucks Lake Wilderness SNYLF populations represent a unique genetic unit (known as Clade 1; Vredenberg et al. 2007, MYLF ITT 2018). Clade 1 is by far the most threatened of the three currently recognized genetic clades (as determined by mitochondrial DNA analysis; Vredenburg et al. 2007), due to few remaining extant populations, marginal habitats, and potential threats from multiple land uses (MYLF ITT 2018). Clade 1 also includes some of the lowest elevation SNYLF populations in the range of the species. Additionally, populations in Clade 1 are widely separated from one another, which limits potential for 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).

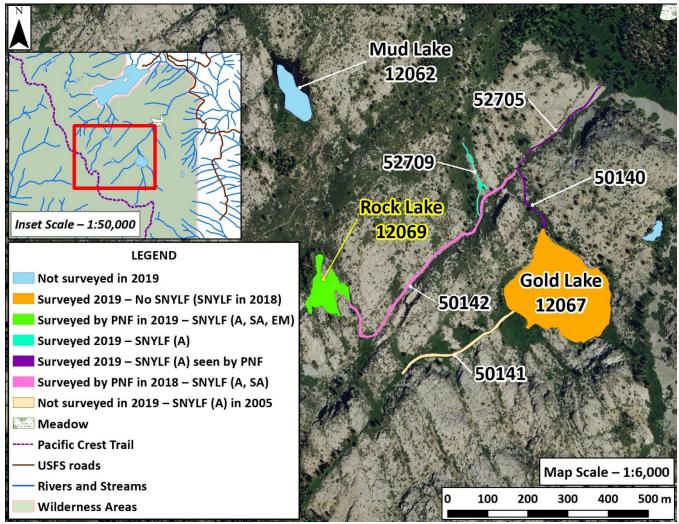


Figure 4: Gold Lake area in Bucks Lake Wilderness, Plumas County, CA. Sierra Nevada Yellowlegged Frog (Rana sierrae; SNYLF) observations occurred during recent visual encounter surveys (VES). SNYLF letter codes in the legend, which indicate the life stage(s) observed during VES, are as follows: "A" = adults, "SA" = subadults, and "EM" = egg masses. In 2005, California Department of Fish and Wildlife (CDFW) staff observed two SNYLF in the Gold Lake inlet (Site ID 50141), but no SNYLF have been subsequently observed during VES of that tributary. The most recent SNYLF observations at Gold Lake were two adults observed by a Plumas National Forest (PNF) biologist in September 2018. Rock Lake is the only known SNYLF breeding location in the drainage. CDFW and PNF field staff consistently observe post-metamorphic SNYLF in the drainages downstream of Rock Lake and Gold Lake. In early July 2019, CDFW staff observed one adult SNYLF in an old mining trench (Site ID 52709) that is fed water from Site ID 50142. In early September 2018, PNF staff collected 64 SNYLF, which had recently undergone metamorphosis, from Rock Lake (n = 4; Site ID 12069, in yellow) and Dot Lake (n = 60; Site ID 12052, which is northwest of Rock Lake and not shown in this figure). Biologists transported the young frogs to the San Francisco Zoo for captive rearing. On 28 June 2019, PNF and CDFW released 19 of the captive-reared SNYLF, which were now young adults, into Rock Lake. Apart from the area below Site ID 52705, 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).

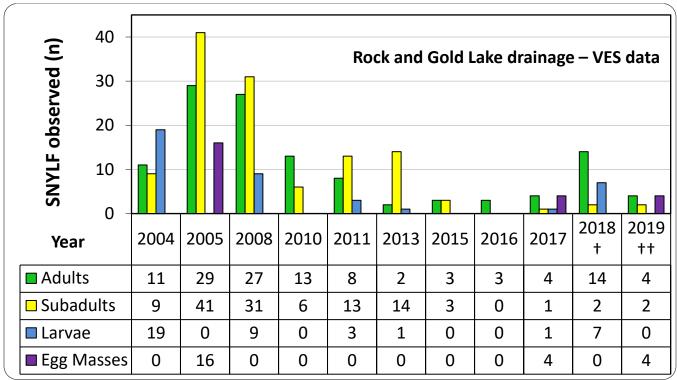
#### VES IN THE GOLD LAKE AREA

CDFW performed the baseline VES in the Gold Lake area in 2004, during which staff encountered a small breeding SNYLF population. Fifteen years of periodic monitoring data suggest that this population may be slowly declining (**Figure 5**). 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.

For several years, PNF and CDFW field staff have been marking newly captured adult SNYLF (>40 mm snout-to-urostyle length [SUL]) in the Gold Lake area with passive integrated transponder (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 and the frog's sex. If the adult is a recapture, staff release the frog without further processing. For new adult captures, 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.

CDFW surveyed the Gold Lake area once in 2019. The survey included Gold Lake and portions of the stream segments below Gold Lake and Rock Lake (Site IDs 50140, 50142, 52705, and 52709; **Figure 4**). In 2019, CDFW did not survey Rock Lake, Site ID 50141, or Site ID 50142 upstream of the hiking trail crossing (UTM Zone 10 S, 659029 E, 4422977 N). However, as part of the captive rearing and release (discussed below), PNF partners surveyed portions of the Gold Lake and Mount Pleasant areas on a few occasions in summer 2019 (C. Dillingham, PNF, pers. comm.). CDFW staff observed one adult SNYLF during VES on 3 July 2019 (**Figure 6**). Field staff located the frog in the vicinity of an old mining trench (Site ID 52709), which is fed water by the adjacent stream segment (Site ID 50142). CDFW first surveyed this trench in 2018, during which staff did not detect SNYLF. CDFW did not have PIT tagging equipment available during the 2019 survey, so field staff did not scan the frog for a tag. However, the visual examination suggested that the frog was not tagged. PIT tags are normally located subcutaneously and, therefore, often easily visible through the skin of marked SNYLF.

During many past surveys of the Gold Lake area, a majority of adult and subadult SNYLF observations have been made at Rock Lake (Site ID 12069; **Figure 7**) and portions of the outlet relatively close to the lake. In addition, most signs of breeding (including observations of egg masses, larvae, and recent metamorphic SNYLF) have occurred in Rock Lake. In 2019, CDFW did not survey Rock Lake or the stretch of Site ID 50142 from Rock Lake to the Silver Lake–Gold Lake trail crossing. Therefore, the exclusion of these areas from surveys likely explains the limited SNYLF detections by CDFW staff in 2019. However, PNF field staff surveyed Rock Lake and some portions of the stream segments below Rock Lake and Gold Lake (**Figure 5**). Both CDFW and the PNF will continue surveying the area at least once each year, keeping records on the location and identity of recaptured adult SNYLF, and PIT tagging any newly captured adults. These data will help CDFW better understand the status and trends of the Gold Lake area SNYLF population.



**Figure 5**: 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, and 50142). Most SNYLF detections have occurred at Rock Lake and the outlet stream in close proximity to Rock Lake (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 (2015, 2016, 2018, and 2019), the data shown are from the survey day resulting in the highest number of SNYLF detections.

CDFW only conducted VES at the following sites during the years listed: Gold Lake during all years shown except 2008; Site 50140 in 2005, 2010–2013, 2016, 2018, and 2019; Site 50141 in 2005, 2010–2011, and 2018; and Site 50142 in 2011–2013 and 2016–2018.

<sup>+</sup> Totals in 2018 include SNYLF detected by PNF staff. CDFW did not add stream segment 52705 (the segment below the confluence of the Rock and Gold Lake outlets) until 2018.

<sup>++</sup>Totals in 2019 include SNYLF detected by PNF staff. In 2019, only PNF staff surveyed Rock Lake (Site ID 12069); additionally, CDFW staff did not survey Site ID 50141 or most of Site ID 50142. *Total does not include young adult SNYLF released at Rock Lake (n = 19) and Dot Lake (n = 21).* 

## VES IN THE MOUNT PLEASANT AREA

CDFW staff did not survey the Mount Pleasant area in 2019. However, CDFW visited the area in late June 2019 for the release of captive-reared SNYLF (**Figure 8**; see **SNYLF CAPTIVE REARING AND RELEASE** section below). For the most recent details on CDFW VES in the Mount Pleasant area, please consult the memorandum "<u>Gold Lake and Mount Pleasant areas – Rana sierrae</u> monitoring; Gold Lake fish removal update" (CDFW 2019).



**Figure 6:** Adult female Sierra Nevada Yellow-legged Frog (*Rana sierrae*; SNYLF) observed during visual encounter surveys (VES) in an old mining trench (Site ID 52709), on 3 July 2019. The trench in which this SNYLF was found is fed water from the main Rock Lake outlet stream (Site ID 50142).

## SNYLF CAPTIVE REARING AND RELEASE

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 and the U.S. Fish and Wildlife Service (USFWS), undertook a collection of early life stage SNYLF from Dot Lake (Site ID 12052; **Figures 9 and 10**) and Rock Lake (Site ID 12069; **Figures 4 and 7**) for captive rearing at the San Francisco 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). During VES in early September 2018 to locate early life stage SNYLF for captive rearing at the San Francisco Zoo, PNF staff observed more SNYLF than CDFW had detected in August. 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 San Francisco Zoo. Once at the zoo, staff treated the young frogs with an antifungal drug called Itraconazole, which clears *Bd* infection. The SNYLF were raised to maturity at the zoo and about two-thirds of captive-reared frogs were mature enough for release back into Bucks Lake Wilderness in summer 2019. CDFW and PNF staff plan to release the remaining SNYLF into Dot Lake and Rock Lake in summer 2020.

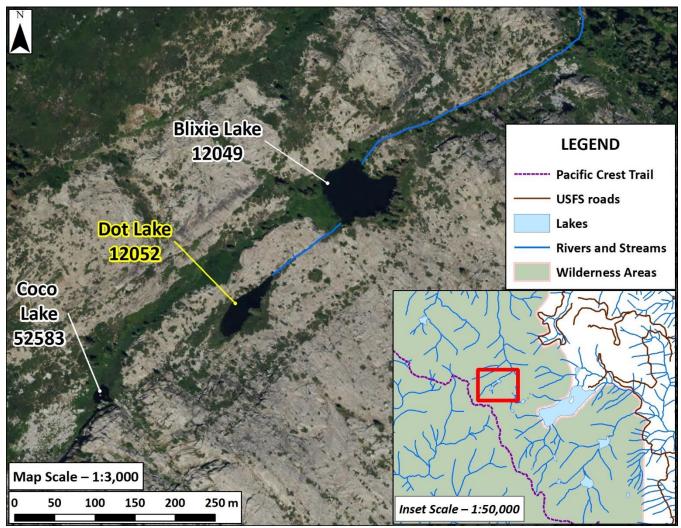
CDFW and PNF staff release all captive-reared SNYLF back into the sites from which young frogs were originally collected. However, since staff detected nearly all early life stage SNYLF at Dot Lake, most captive-reared SNYLF that staff released back into Rock Lake were originally collected from Dot Lake. Given that Dot Lake and Rock Lake are close together (< 2.5 km), contain very similar habitat, and both contain *Bd*-positive SNYLF populations (see *Disease* section above), 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 very little genetic differentiation exists between SNYLF populations located in such close proximity (Andy Rothstein, UC Berkeley, pers. comm., 3/2019). Therefore, CDFW, PNF, and partner scientists have determined that short distance movements like those undertaken between Dot Lake and Rock Lake are an appropriate conservation measure for SNYLF (MYLF ITT 2018).



**Figure 7**. Rock Lake (Site ID 12069) on 21 August 2018. On 28 June 2019, California Department of Fish and Wildlife (CDFW) and Plumas National Forest (PNF) biologists released 19 captive-reared Sierra Nevada Yellow-legged Frogs (*Rana sierrae*; SNYLF), most of which were originally collected from Dot Lake. Most signs of SNYLF breeding (including observations of egg masses, larvae, and recently metamorphosed SNYLF) in the Gold Lake area have occurred at Rock Lake.



**Figure 8:** Two captive-reared young adult Sierra Nevada Yellow-legged Frogs (*Rana sierrae*; SNYLF) recently released at Dot Lake on 28 June 2019. (CDFW)



**Figure 9**: Mount Pleasant drainage in Bucks Lake Wilderness, Plumas County, CA. In early September 2018, Plumas National Forest (PNF) staff collected 64 SNYLF, which had recently undergone metamorphosis, from Dot Lake (n = 60; Site ID 12052, in yellow) and Rock Lake (n = 4; Site ID 12069, which is southeast of Dot Lake and not shown in this figure). Biologists transported the young frogs to the San Francisco Zoo for captive rearing. On 28 June 2019, California Department of Fish and Wildlife (CDFW) and PNF biologists released 21 captive-reared Sierra Nevada Yellow-legged Frogs (*Rana sierrae*; SNYLF) at Dot Lake. PNF field staff returned to Dot Lake for multiple follow-up surveys in 2019 (C. Dillingham, PNF, pers. comm.). During PNF follow-up surveys, only one captive-reared SNYLF was recaptured. However, PNF field staff observed six newly captured (wild) adult SNYLF at Dot Lake, all of which field staff marked with passive integrated transponder (PIT) tags. Additionally, PNF staff observed eight newly captured SNYLF adults in Mill Creek, which is the stream flowing northeast out of Blixie Lake. All SNYLF that PNF staff observed in Mill Creek were located in the reach visible in the aerial image. 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 10**. Dot Lake (Site ID 12052) on 28 June 2019, the day California Department of Fish and Wildlife (CDFW) and Plumas National Forest (PNF) biologists released 21 captive-reared Sierra Nevada Yellow-legged Frogs (*Rana sierrae*; SNYLF) 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.

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 San Francisco Zoo. These 40 individuals are a subset of the 64 individuals that were collected by PNF staff on 6 September 2018. CDFW staff picked up 42 SNYLF from SF Zoo staff on the morning of 28 June and transported them to the PNF Mt. Hough Ranger Station. There, the frogs were split 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, each frog's PIT tag was scanned (**Figure 11**) and snout-urostyle length (SUL) and weight measurements were taken. Twenty-one frogs were successfully reintroduced to Dot Lake and nineteen to Rock Lake. Two frogs intended for release into Rock Lake perished after brief exposure to excessive 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.

While at Rock Lake, field staff incidentally observed three SNYLF egg masses. Additionally, staff observed one resident adult SNYLF at Dot Lake during a snorkel survey (**Figure 12**).



**Figure 11**: A Plumas National Forest biologist scanning a young adult Sierra Nevada Yellow-legged Frog (*Rana sierrae*) with a PIT tag reader prior to its release in Dot Lake on 28 June 2019. (CDFW)



**Figure 12**: A Plumas National Forest biologist holding a large female Sierra Nevada Yellow-legged Frog (*Rana sierrae*) captured during a snorkel survey of Dot Lake on 28 June 2019. (CDFW)

#### DISCUSSION

The Gold Lake fish removal project has been successful at mechanically removing BK using monofilament gill nets. The BK population was small at Gold Lake: only about 250 trout were removed from the lake after nearly constant gill-netting efforts for over three years. Removing a non-native predatory fish from the deepest lake in Bucks Lake Wilderness is highly desirable for the SNYLF population in this basin, especially given that few remaining lake-based SNYLF populations exist in Plumas County. Gold Lake will potentially supply additional foraging and overwintering habitat for SNYLF that immigrate from the Rock Lake drainage. Now lacking predatory non-native BK, Gold Lake is more likely to provide suitable deep-water habitat for SNYLF.

Gold Lake and Blixie Lake continue to contain robust DC-S populations. CDFW does not know what effect DC-S may have on SNYLF breeding (e.g., potential for DC-S to prey on or damage egg masses and early life stage tadpoles). Blixie Lake provides some circumstantial evidence for DC-S limiting SNYLF breeding because SNYLF eggs and tadpoles have not been observed. Conversely, Goose Lake, located at the southern edge of PNF in Sierra County, contains an abundant DC-S population sympatric with SNYLF. CDFW and PNF staff regularly monitor Goose Lake and the other waterbodies in the drainage east of Gold Lake reservoir (Sierra County). During recent surveys, in addition to observations of SNYLF adults and subadults, staff have observed egg masses in 2010, 2016, 2017, and 2019. CDFW is not aware of additional fishless deep water breeding habitat in the Goose Lake area, and VES results suggest that the SNYLF population in this area appears to be doing well. Therefore, the presence of DC-S clearly does not preclude successful SNYLF breeding. However, little quantitative data are available regarding direct interactions between DC-S and early life stage SNYLF.

There is some evidence that other cyprinids can cause reduced survival and growth in amphibians. For example, a mesocosm study found that Fathead Minnows (*Pimephales promelas*) reduced the survival and growth rates of salamander larvae through competition and inflicting injury (Pearson and Goater 2009). Other small fish species have been linked with sublethal effects on amphibians, including Mosquitofish (*Gambusia* sp.; Pyke and White 2000; studies summarized in Kats and Ferrer 2003). Experimental evidence has shown that Three-spined Sticklebacks (*Gasterosteus aculeatus*) cause limb and tail damage in larval Western Toads (*Anaxyrus boreas*) identical to damage observed in the field (Bowerman et al. 2010). Undoubtedly, numerous fish species can affect amphibian larvae in different ways, and many of those effects may be deleterious, if not necessarily lethal (Wells 2007 pgs. 657–659).

The evidence for other common aquatic predators affecting larval amphibians further complicates isolating potential effects of minnows. For example, various species of dragonfly nymphs are voracious predators of amphibian larvae (See Table 14.2 in Wells 2007; Ballengée and Sessions 2009, Bowerman et al. 2010). Additionally, other amphibian larvae found in the northern Sierra Nevada are known to prey on conspecifics (e.g., *Ambystoma macrodactylum*; Wildy et al. 1998, Wildy et al. 2001).

In light of these complications and unknowns regarding interactions between minnows and SNYLF, CDFW and PNF will continue to monitor the Gold Lake and Blixie Lake populations to detect recruitment (particularly tadpoles), and observe potential interactions between minnows and SNYLF. This site, in combination with SNYLF populations in the Gold Lake Reservoir area (in Sierra County, some of which are also sympatric with minnows), could provide a unique opportunity to study the ecological interactions of native cyprinids and early life stage SNYLF.

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 in 2018 and 2019 have already resulted in an increase in SNYLF observations in the Gold Lake drainage. SNYLF will likely continue migrating toward Gold Lake via the streams connecting to Rock Lake. However, as opportunities allow–based on SNYLF population status, funding, zoo capacity, and availability of personnel–CDFW and PNF may collect additional early life stage frogs for future captive rearing efforts to help supplement the Bucks Lake Wilderness SNYLF populations.

#### RECOMMENDATIONS

CDFW 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). Staff will especially 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.

Additionally, CDFW may work with local zoo and university partners to develop a research project (e.g., a graduate research masters study) on the interactions between cyprinids and SNYLF, especially early life stages, such as eggs and recently hatched larvae. The interactions of large predatory fish (e.g., trout) and SNYLF are well-studied, but there is much less known about the interactions of smaller forage fish and amphibians, especially potential sublethal effects (e.g., limited breeding success, reduced size at metamorphosis, limb damage) on frog populations.

If results from the releases of captive-reared SNYLF in 2018–2020 are encouraging, 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.

#### LITERATURE CITED

- Ballengée, B., and S.K. Sessions. 2009. Explanation for missing limbs in deformed amphibians. Journal of Experimental Zoology (Molecular Development and Evolution) 312B:770–779.
- Bowerman, J., P.T.J. Johnson, and T. Bowerman. 2010. Sublethal predators and their injured prey: linking aquatic predators and severe limb abnormalities in amphibians. Ecology 91:242–251.
- California Department of Fish and Wildlife (CDFW). 2015. Aquatic Biodiversity Management Plan for the Bucks Lake Wilderness Management Unit. North Central Region, Rancho Cordova. Available from: <u>http://nrm.dfg.ca.gov/FileHandler.ashx?DocumentID=102579</u>
- CDFW. 2018. Native amphibian restoration and monitoring in Bucks Lake Wilderness: Gold Lake fish removal and Rock Lake *Rana sierrae* monitoring. Available from: <u>http://nrm.dfg.ca.gov/FileHandler.ashx?DocumentID=156673</u>
- CDFW. 2019. Native amphibian restoration and monitoring in Bucks Lake Wilderness: Gold Lake and Mt. Pleasant areas *Rana sierrae* monitoring and Gold Lake fish removal update. Available from: <u>http://nrm.dfg.ca.gov/FileHandler.ashx?DocumentID=165859</u>
- Ellison, S, R.A. Knapp, W. Sparagon, A. Swei, and V.T. Vredenburg. 2019. Reduced skin bacterial diversity correlated with increased pathogen infection intensity in an endangered amphibian host. Molecular Ecology 28:127–140.
- Frankham, R., J.D. Ballou, and D.A. Briscoe. 2009. Introduction to Conservation Genetics. Cambridge University Press, New York, NY, USA.
- Kats, L.B., and R.P. Ferrer. 2003. Alien predators and amphibian declines: review of two decades of science and the transition to conservation. Diversity and Distributions 9:99–110.
- McAllister, K.R., J.W. Watson, K. Risenhoover, and T. McBride. 2004. Marking and radiotelemetry of Oregon spotted frogs (*Rana pretiosa*). Northwestern Naturalist 85:20–25.
- Mountain Yellow-legged Frog Interagency Technical Team (MYLF ITT). 2018. Interagency conservation strategy for mountain yellow-legged frogs in the Sierra Nevada (*Rana sierrae* and *Rana muscosa*). California Department of Fish and Wildlife, National Park Service, U.S. Fish and Wildlife Service, U.S. Forest Service. Version 1.0.
- Pearson, K.J., and C.P. Goater. 2009. Effects of predaceous and non-predaceous introduced fish on the survival, growth, and antipredation behaviors of long-toed salamanders. Canadian Journal of Zoology 87:948–955.
- Pyke, G.H., and A.W. White. 2000. Factors influencing predation on eggs and tadpoles of the endangered green and golden bell frog *Litoria aurea* by the introduced plague minnow *Gambusia holbrooki*. Australian Zoologist 31:496–505.

- Rachowicz, L.J., R.A. Knapp, J.A.T. Morgan, M.J. Stice, V.T. Vredenburg, J.M. Parker, and C.J.
  Briggs. 2006. Emerging infectious disease as a proximate cause of amphibian mass
  mortality. Ecology 87:1671–1683.
- Vredenburg, V.T., R. Bingham, R. Knapp, J.A.T. Morgan, C. Moritz, and D. Wake. 2007. Concordant molecular and phenotypic data delineate new taxonomy and conservation priorities for the endangered mountain yellow-legged frog. Journal of Zoology 271:361– 374.
- Wells, K. 2007. The ecology and behavior of amphibians. The University of Chicago Press, Chicago, Illinois.
- Wildy, E.L., D.P. Chivers, J.M. Kiesecker, and A.R. Blaustein. 1998. Cannibalism enhances growth in larval long-toed salamander, (*Ambystoma macrodactylum*). Journal of Herpetology 32:286–289.
- Wildy, E.L., D.P. Chivers, J.M. Kiesecker, and A.R. Blaustein. 2001. The effects of food level and conspecific density on biting and cannibalism in larval long-toed salamanders, *Ambystoma macrodactylum*. Oecologia 128:202–209.