Giant Gartersnake (*Thamnophis gigas*) Surveys on the Cosumnes River Ecological Reserve: Sacramento County, California

2017 Monitoring Report

Prepared for:

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October 30, 2017

EXECUTIVE SUMMARY

This report describes the results of post-drought monitoring of giant gartersnake (*Thamnophis gigas*) presence and distribution at the Cosumnes River Ecological Reserve (CRER). Once supporting the most robust and genetically diverse *T. gigas* population known to remain throughout the species' historic range, the CRER has experienced dramatic curtailments to hydrologic inputs in recent decades. Exacerbated by an historic drought affecting the Central Valley from 2012-2016, remaining inputs failed to provide adequate surface water during the spring and summer *T. gigas* active season. This resulted in the premature drying of crucial aquatic habitat from 2014-2016 despite the addition of groundwater augmentation in 2015 and 2016. The described monitoring from 2017 was conducted to assess the status of the CRER *T. gigas* population following the drought and to compare the resulting data with those collected in association with restoration work from 2008-2009.

To maximize statistical power and to account for potential spatiotemporal variation in detection rates, *T. gigas* were sampled during four discrete and equal survey periods between May 30 and September 16, 2017. Additional survey efforts included periodically measuring water chemistry, cataloguing prey items collected in traps, and collecting and archiving *T. gigas* tissue samples for future genetic analysis. In total, 26 individual *T. gigas* (16 females and 10 males) were captured in 35 capture events at the CRER in 2017 (4 females and 2 males were captured two or more times). Of the 35 *T. gigas* capture events, 2 occurred in the residually-wetter portion of the East Marsh, 12 occurred in the West Marsh and 21 were captured in the in the area that was restored to increase water depth and open water surface in 2008 (Restoration Site). None of the *T. gigas* captured in 2017 were among the 429 captured and marked in previous years (86 from 2001-2003, 331 from 2008-2009, and 13 in 2016). Twenty-six tissue samples were collected and archived for future genetic analyses.

In 2009, we recorded 281 captures of 194 snakes, and in 2017, we recorded 35 captures of 26 snakes. The estimated population in 2009 was 651.1 (409-962). As expected, the post-drought abundance estimate for the CRER *T. gigas* population in 2017 was much smaller at 98.4 (41-226). The estimate of the difference between years is 536 (284-878), with abundance estimated for 2017 representing only 13% (6% to 40%) of that calculated for 2009.

All pH and electrical conductivity (EC) measurements obtained in 2017 fell within the ranges believed to be tolerable for *T. gigas* prey species. While the composition and of prey species collected in traps was comparable to that observed at other occupied sites, abundance was low. Though water was present, levels were highly variable, and the floating accumulation of biomass from yellow water primrose (*Ludwigia hexapetala*) appeared to compromise wetland function by occluding the open water surface.

INTRODUCTION

Habitat fragmentation and alteration caused by the intensification of human uses on the landscape have numerous, negative impacts on habitat quality and biodiversity. For wetlands in California, draining and conversion to cropland and urban development has reduced the once vast expanses of marshland to less than 10% of their extent prior to European settlement (Dahl et al., 1991). In the Central Valley, 43% of freshwater wetlands in the have been lost or converted since 1939 (Frayer et al. 1989). Even relatively natural habitat that remains physically intact in these highly altered landscapes is often functionally impaired. Wetlands are important natural features that provide critical ecosystem functions including regulation and maintenance of hydrologic processes through flood attenuation, groundwater recharge and water quality improvement (Wilen and Bates 1995). Wetlands also provide critical habitat for fish and wildlife species, with over one third of all threatened and endangered species occurring in wetlands according to the U.S. Environmental Protection Agency (https://www.epa.gov/wetlands/why-are-wetlands-important).

One such species, the giant gartersnake (*Thamnophis gigas*), is classified as a threatened species under both State and Federal Endangered Species Acts. The species historically occurred from the northern portions of the Sacramento Valley southward to the southern portion of the San Joaquin Valley. *T. gigas* are associated with low-gradient streams, valley-floor wetlands, and marshes in California's Central Valley and are considered the most aquatic of the gartersnakes in California (Fitch 1940). They require freshwater wetlands for foraging on fish and amphibians (their primary prey species), upland areas for basking, upland burrows as summer shelter, and higher elevation uplands for winter habitat (Hansen and Brode 1980; USFWS 1993; USFWS 2017). Extant *T. gigas* populations are distributed discontinuously in fragmented patches of remaining natural wetlands, created and restored wetlands, and areas where natural wetlands were converted to and have since remained in rice agriculture. The loss of historical habitat for *T. gigas* has resulted in extirpations or serious declines throughout much of the species' former range.

Part of the Cosumnes-Mokelumne Basin, the Cosumnes River Ecological Reserve (CRER) supports one of 9 distinct and separate populations of *T. gigas* recognized by the U.S. Fish and Wildlife Service (2017). Concentrated at the confluence of Badger and Willow creeks in a wetland known as Snake Marsh by CRER staff, the CRER population is unique in that natural marsh and creeks make up most of the existing habitat. Recent genetic analysis demonstrates that *T. gigas* in Snake Marsh harbor the greatest genetic diversity compared to *T. gigas* in other population areas (Paquin et al., 2006; Engstrom and Olson 2007; Wood et al. 2015).

The historical hydrology at Badger Creek was apparently much different prior to reclamation, with the pre-development water table in the Cosumnes River vicinity

existing near ground levels, providing persistent surface water in channels and depressions year-round, despite varying seasonal precipitation in the basin (Phillip Williams and Associates. 1997). Current hydrology relies upon varying seasonal precipitation and agricultural runoff, and provides no guarantee that suitable habitat can be sustained. In addition, excessive groundwater pumping in south Sacramento County has severely depleted regional groundwater levels, resulting in diminished surface flows in the nearby Cosumnes River during the dry season (Fogg et al. 2001, Mount et al. 2001). Regional groundwater depletion may also be impacting surface flows on tributaries such as Badger Creek.

The Badger Creek *T. gigas* population once occupied wetlands throughout the Cosumnes River watershed, but now appears to be largely restricted to Snake Marsh. This population was once the healthiest known population in the species' extant range, harboring important genetic variation that could be used to repatriate areas where the species has been extirpated. However, after three consecutive years of drought, much of Snake Marsh went dry in 2014, and owner/manager California Department of Fish and Wildlife (CDFW) used emergency drought funds to install a well and contour ditches to deliver water to the northern pools. Despite the ability to augment water supply, the pools prematurely dried again in 2015 and 2016, leaving the post-drought status of this vital population unknown. To assess the status of the CRER *T. gigas* population following the drought and to compare the resulting data with those collected in association with restoration work from 2008-2009, CDFW undertook two surveys in 2016, but limitations on the amount and timing of available drought funding as well as difficulty assembling adequately trained staff and necessary equipment prevented the type of survey effort required to determine population status. Due to these constraints, the surveys were limited in duration, geographic scope, and were poorly aligned with peak *T. gigas* activity periods.

To ameliorate these constraints, CDFW in 2017 turned to the Governor's Drought Executive Order 4-25-2014, which directed the CDFW to immediately implement projects that respond to drought conditions through habitat restoration and through water infrastructure projects on property owned or managed by CDFW for the benefit of fish and wildlife impacted by the drought. The Executive Order authorized CDFW to enter into contracts to accomplish this work, and work was subsequently contracted to complete the following objectives:

1. Conduct a trapping survey of all accessible habitat on the CRER during the optimal activity period for *T. gigas*;

2. Collect genetic samples for future analyses to determine if the effective population size has been reduced or genetic variability has been lost;

3. Collect water samples for eDNA analyses on tributaries Snake Marsh (i.e., North and South Forks of Badger Creek; and

4. To the extent possible, provide summary statistics comparing the post-drought distribution, demography, and density (or CPUE) of *T. gigas* to pre-drought population estimates.

Project Location

The project site is in the Badger Creek Unit of the CRER in south Sacramento County, California (**Figure 1**). The Badger Creek Unit is located at the confluence of Badger Creek and the Cosumnes River, extending east to highway 99 between the Arno and Dillard Road interchanges (**Figure 2**). The Unit is entirely within the 100-year floodplain and supports several natural communities including valley oak riparian forest, annual grasslands, and freshwater wetlands. The 1275-acre Badger Creek Unit is owned and managed by the California Department of Fish and Wildlife.

West of the Union Pacific Railroad tracks to Valensin Forest, Snake Marsh is connected directly to Badger Creek via Horseshoe Lake. The area east of the Union Pacific Railroad tracks east to Highway 99 is connected directly to Willow Creek via a box culvert under the highway and is divided by a vegetated land berm that demarks the Preserve's southern property boundary. Badger Creek from Valensin Forest west to its confluence with the Cosumnes River is largely characterized by oak woodland and riparian overstory.

Figure 1. Project location

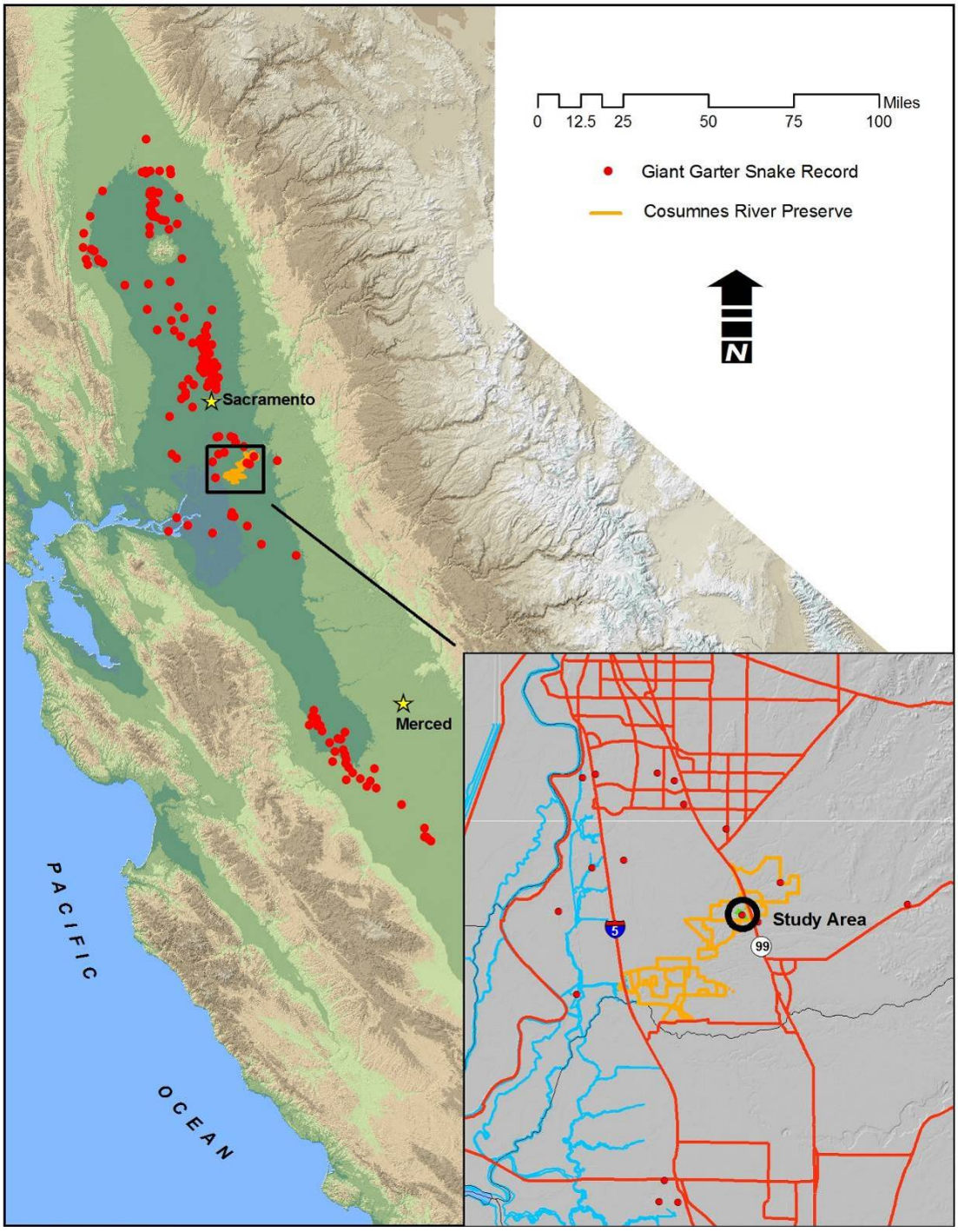
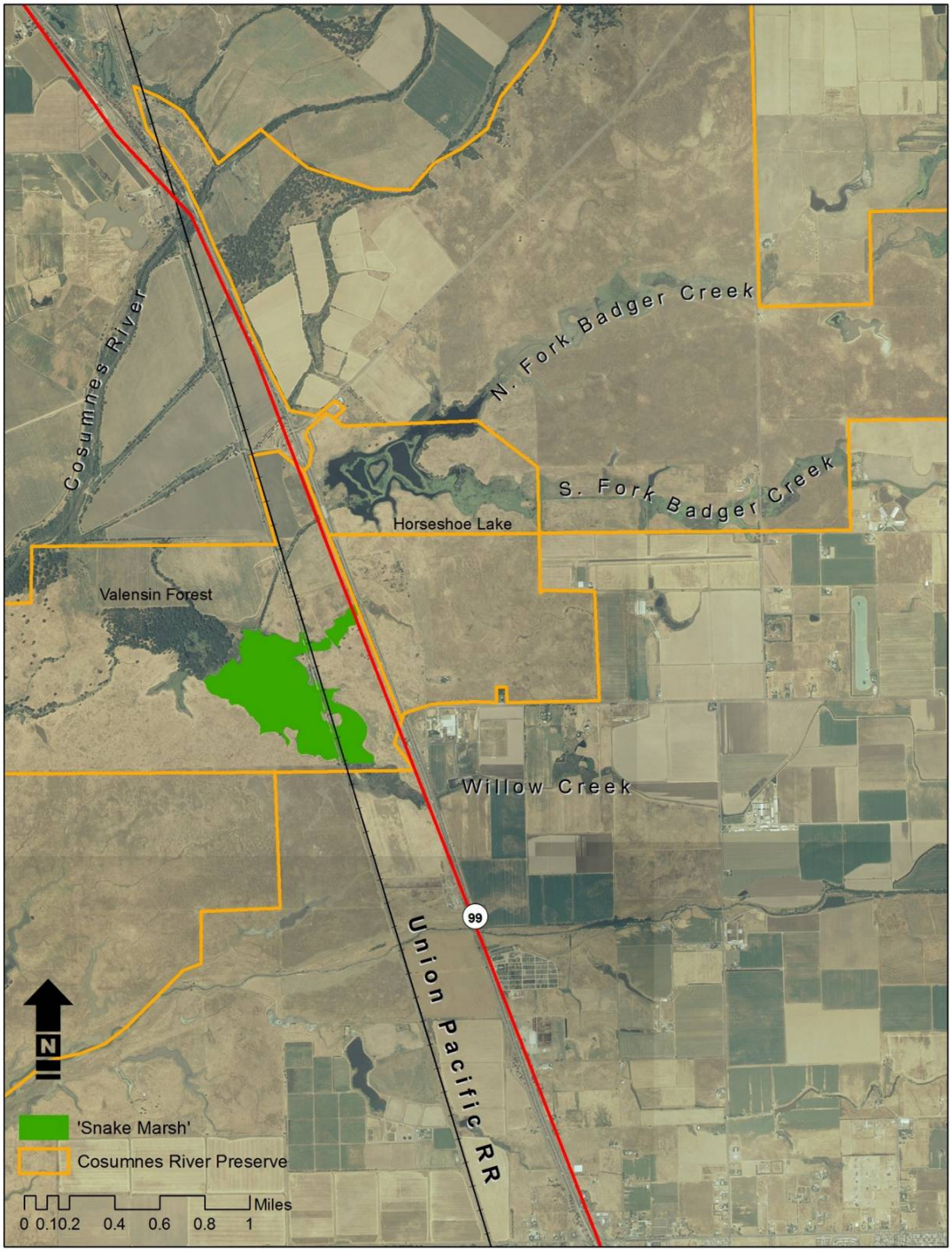


Figure 2. Project area detail

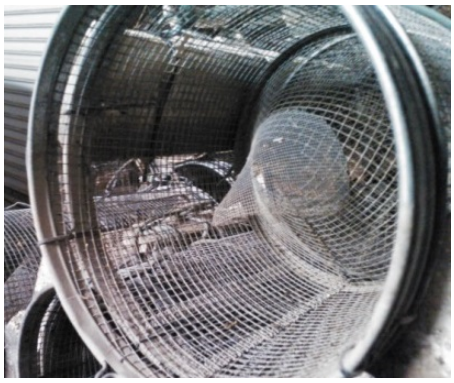


METHODS

Giant Gartersnake Surveys

Sampling for *T. gigas* entailed a combination of aquatic trapping and visual encounter surveys (VES). Both trapping and VES were conducted during four discrete 15-day periods temporally distributed between May 30 and September 16. The four 15-day periods sample a greater range of dates (and thus a greater range of snake movements and seasonal habitat changes) throughout the active season.

While divided among four discrete and equal periods between May 1 and September 30, the precise timing of surveys is determined by weather and seasonal conditions and deliberately chosen to maximize the probability of capture success. VES are conducted by walking or kayaking along channels and nearby upland areas to search for basking and mating snakes. Primary searching areas include vegetated banks, channels, drainages, and marshland edges, as well as upland basking sites. When possible, snakes discovered during these searches are captured either by hand or using reptile snares. Global positioning system (GPS) units are used to determine the geo-coordinates of capture locations, and associated data, such as vegetation type, approximate water depth, substrate type, time of day, and ambient temperature are recorded. VES are also conducted incidental to daily trap checks.



Surveys implemented a sampling design based on multiple habitat strata or types with transects distributed at a spatial scale consistent with current methods described by Halstead et al. (2009, 2011) that are designed to maximize the probability of detection. Trapping methods were also implemented to decrease escape rates (Halstead et al. 2013). Flexible screen mesh covers or hoods extending below the waterline were used on the aperture of each trap funnel to reduce escape rates (Hansen, unpublished data). Trapping entailed the placement and daily monitoring of 150 floating aquatic traps divided into three (3) 25- and one (1) 75-trap transects placed at suitable areas of aquatic habitat and monitored for 15 days during four (4) separate survey periods spanning from May 30 to September 16. Conditions in 2017 were drier than those experienced previously, so sites that dried in less than 15 days were moved to new locations to maintain the intended trapping intensity. A summary of 2017 trap effort is provided in **Table 1**.

Table 1. 2017 *T. gigas* Trapping Quadrat Geocoordinates[1] and Survey Dates

	Quadrat ID	Centroid Easting	Centroid Northing	Start Date	End Date
Phase 1	BC27	645249	4243744	5/30/2017	6/14/2017
	BC28A	645486	4243197	6/1/2017	6/15/2017
	BC28B	645251	4243127	6/1/2017	6/15/2017
	BC28C	645129	4243437	6/1/2017	6/15/2017
Phase 2	BC27	645249	4243744	6/26/2017	7/11/2017
	BC28A	645486	4243197	6/27/2017	7/12/2017
	BC28D	645279	4243203	6/27/2017	7/12/2017
	BC28E	645349	4243263	6/27/2017	7/12/2017
	BC29	645744	4243050	6/27/2017	7/2/2017
Phase 3	BC27	645249	4243744	8/5/2017	8/19/2017
	BC28E	645349	4243263	8/5/2017	8/19/2017
	BC28F	645253	4243251	8/5/2017	8/19/2017
	BC28G	645387	4243293	8/5/2017	8/19/2017
P4	BC27	645249	4243744	9/1/2017	9/16/2017
	BC28I (= F-H)	645319	4243305	9/1/2017	9/16/2017

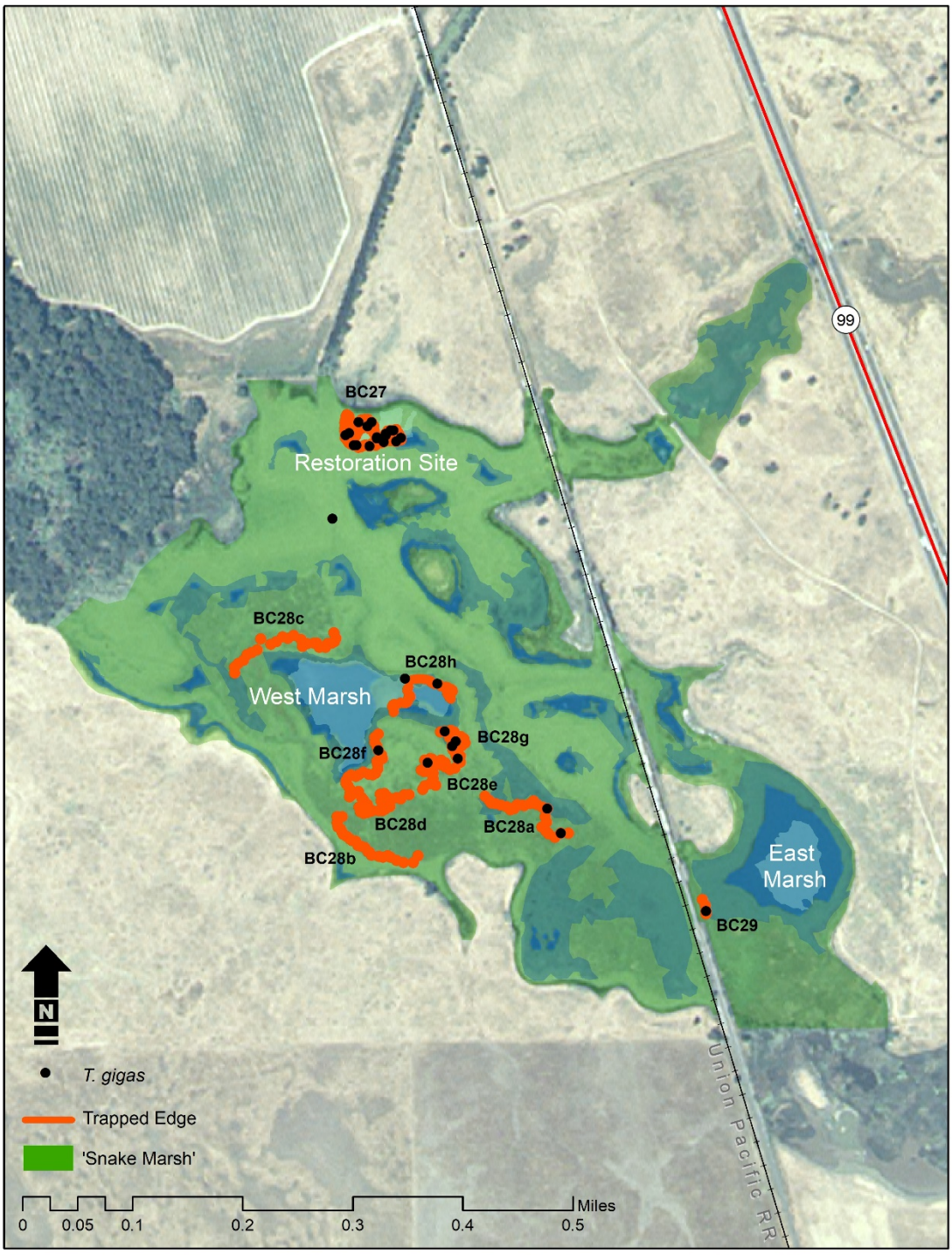
1. North American Datum 1983, UTM Zone 10

Data recorded at each trap location include geo-coordinates and environmental characteristics, (e.g., distance from upland refuge, bank slope, approximate water depth, dominant vegetation type, height, and density). Digital photographs are also taken at each quadrat at the start and end of each survey period. Each deployed trap is checked daily.

To assess demographics according to this protocol, weight, total length, snout to vent length (SVL), and sex is recorded for all captured *T. gigas*. Other physical features such as scars and tumors, as well as identifying characteristics, such as scale counts and measurements on head and midbody, are also noted. Captured snakes are photographed and implanted with passive integrated transponder (PIT) tags for permanent identification. For snakes that are too small to implant with PIT tags (≤ 30 grams), medical cautery units are used to microbrand caudal scutes in a pattern consistent with established scale-clip marking techniques (Brown and Parker 1976, Winne et al. 2006). Marking snakes is essential for estimating population size, density, male to female ratios, and fecundity of the species (E. Hansen 2004, USFWS 1999, Wylie et al. 1997). Snakes are released at their capture location immediately following data recordation.

Surveys were conducted per the terms and conditions of the PI's US FISH AND WILDLIFE SERVICE RECOVERY PERMIT 10(a) (1) (A) ESA TE-018177-7 (valid through 08/06/2019) and DEPARTMENT OF FISH AND GAME SCIENTIFIC COLLECTING PERMIT 003881 (valid through 04/16/2018).

Figure 2. Snake Marsh 2017 *T. gigas* trap and capture locations



Water Quality Metrics

Water chemistry metrics, including pH, electrical conductivity (EC) (mS/cm), and temperature, were measured at each trapping location (where sufficient water is present) at the beginning and end of each survey period using a portable YSI 556 Multi-Probe unit. Fluctuations in water surface levels were also noted. While water quality measurements are not a requirement for *T. gigas* monitoring at SBCB, these data are collected to help interpret observed *T. gigas* and aquatic prey distributions.

Prey Samples

Although traps are not purposely baited, frogs, tadpoles, and fish are frequently caught in the traps and likely serve as attractants for *T. gigas*. Wherever traps remained in place without interference, organisms within the traps (by-catch) were identified and counted at the end of each survey period to compare prey composition and densities between trapping sites.

Abundance estimates

We collected data from 2009 and 2017 from the population of *T. gigas* at Snake Marsh in the CRER. The primary goal was to estimate annual abundance, and we used a capture-recapture model for closed populations (Otis et al. 1978) to separately analyze the data for each year. A second goal was to compare estimates of abundance at Badger Creek between 2009 and 2017. The site at Badger Creek experienced considerable drying, and we wanted to compare estimates to quantify the effect of the drying on the population *T. gigas*. Capture-recapture models were necessary for analyses, because they account for the fact that, for most species, a proportion of the individuals in a population are not captured on sampling occasions, regardless of the method of capture and the intensity of effort (a phenomenon referred to as imperfect detection). Imperfect detection is particularly relevant to species of snakes, because their wariness, coloration and other behaviors make them difficult to observe and capture (Lind et al. 2005, Breininger et al. 2012). Therefore, counts of the number of individuals that are trapped will nearly always be lower than the actual number of individuals in a population. Capture-recapture models for closed populations estimate the probability of capturing individuals, which allows abundance to be estimated. Because traps were checked daily, we treated each day as a sampling occasion, and our estimates of capture probability pertain to each day during the sampling period.

Estimates of abundance from models for closed populations can be negatively biased when capture probability varies among individuals (Williams et al. 2002), and variation in capture probability can be addressed using multiple modeling approaches (Kery and Schaub 2012). We fit the logistic-normal, mixture model to the data (Kery and Schaub 2012). The model included a random effect by treating the capture probabilities of individuals as if they arose from a normal distribution with an estimated mean and variance. We analyzed the data in a Bayesian framework. Prior to fitting models, we

augmented the capture-recapture data by adding many zero-only capture histories. Data augmentation is necessary to fix the dimensions of the parameter vector for Markov Chain Monte Carlo simulation, a central feature of Bayesian methods (Kery and Schaub 2012).

We fit all models using OpenBUGS, an open source version of WinBUGS (Lunn et al. 2000). We used Uniform(0, 1) priors on all probability parameters. We ran models on 3 chains for 200,000 iterations each after a burn-in of at least 50,000 iterations. We thinned the chains by a factor of 5, which resulted in a total of 120,000 samples in each posterior. We visually inspected trace plots to ensure chains were well-mixed, and evaluated convergence for each model using the $\hat{R}$ statistic. We concluded that convergence had been achieved, if $\hat{R}$ was less than 1.1 (Gelman et al. 2004). Finally, to determine if the posteriors contained enough samples, we examined the ratio of Monte Carlo error to the standard deviation of the posterior for parameters (Kery and Schaub 2012). If the ratio was less than 0.05, we concluded that the number of samples was adequate.

RESULTS

Giant Garter Snake Surveys

Trapping surveys resulted in the accrual of 4,006 total trap days. The respective number of trap days accrued in the East Marsh, West Marsh, and Restoration Site was 30; 4,500; and 4,500. Survey efforts were constrained to some extent by habitat conditions and other factors predominantly driven by varying water volume and surface within the system. For instance, drying of the system necessitated relocating established traplines in the West Marsh during later trapping sessions. Regardless, except for the small trap cluster placed in the East Marsh, continuous sampling effort was maintained in all regions during all sessions. Sampling results are summarized in Table 1 and Table 2, below. Survey period dates and trapping effort (i.e., trap days accrued) are summarized in **Table 1**.

In total, 26 individual *T. gigas* (16 females and 10 males) were captured in 35 capture events at the CRER in 2017 (4 females and 2 males were captured two or more times). Of the 35 *T. gigas* capture events, 2 occurred in the residually-wetter portion of the East Marsh, 12 occurred in the West Marsh and 21 were captured in the in the area that was restored to increase water depth and open water surface in 2008 (Restoration Site). None of the *T. gigas* captured in 2017 were among the 429 captured and marked in previous years (86 from 2001-2003, 331 from 2008-2009, and 13 in 2016). All *T. gigas* capture locations are depicted in Figure 2.

Overall catch per unit effort (CPUE), calculated as the total number of individuals trapped per trap day, was 0.0035. Trapping effort and capture results (counts and CPUE) for each habitat type are reported in **Table 1**.

Table 1: 2017 (post-drought) *T. gigas* trapping effort and results summary by area

Unit	Trap Days	Individuals Captured	CPUE
Restoration Site	4500	17	0.0047
West Marsh	4500	13	0.0038
East Marsh	30	2	0.067
Snake Marsh Total	9030	32	0.0035

Table 2: 2008-2009 (pre-drought) *T. gigas* trapping effort and results summary by area

Unit	Trap Days	Individuals Captured	CPUE
2008 Field Season			
East Marsh	2150	82	0.038
West Marsh	1500	83	0.055
Restoration Site	1043	7	0.007
2009 Field Season			
East Marsh	2250	39	0.017
West Marsh	2250	112	0.050
Restoration Site	1910	42	0.022
Snake Marsh Total	11103	365	0.033

Abundance

At the CRER, the duration of sampling was 46 days in 2009 and 65 days in 2017. In 2009, we recorded 281 captures of 194 snakes, and in 2017, we recorded 35 captures of 26 snakes. The estimated population in 2009 was 651.1 (400-962). As expected, the post-drought abundance estimate for the CRER *T. gigas* population in 2017 was much smaller at 98.4 (41-226). The estimate of the difference between years is 536 (284-878), with abundance estimated for 2017 representing only 13% (6% to 40%) of that calculated for 2009 (i.e., a reduction of 87%). Abundance estimates and credible intervals for the CRER *T. gigas* population in 2009 and 2017 are presented in **Table 3.**

Table 3: 2009 (pre-drought) and 2017 (post-drought) *T. gigas* abundance estimates

Survey Year	Trap Days	Estimated Abundance	Credible Interval
2009 Field Season (pre-drought)	6410	651.1	(409-962)
2017 Field Season (post-drought)	9030	98.4	(41-226)

Water Quality Metrics

Where sufficient water was present, water chemistry metrics, including pH and electrical conductivity (EC) (mS/cm), were measured at each trapping quadrat twice during the full survey period. The mean electrical conductivity (EC) of all samples at CREC was 0.42mS/cm; mean salinity was 0.20 parts per trillion (PPT); mean percent saturation of dissolved oxygen was 35.68; and mean pH was 7.183. All water chemistry measurements obtained in the CRER during 2017 are reported in **Table 4**.

Table 4. 2017 Water Chemistry Metrics

Quadrat ID	Date	Time	H2O Temp (C)	Conductivity (mS/m)	Salinity (PPT)	DO % Saturation	pH
BC27RS	7/12/2017	9:18	22.48	0.40	.19	19.7	6.96
BC27RS	8/15/2017	9:10	20.12	0.46	.22	8.2	7.23
BC28	7/12/2017	10:10	24.53	0.40	.19	97.3	7.16
BC28	8/15/2017	9:27	22.15	0.43	.21	17.5	7.38
Mean	--	--	22.32	0.42	0.20	35.68	7.183

Prey Samples

By-catch consisted of both adult and larval American bullfrogs (*Lithobates catesbeianus* [=*Rana catesbeiana*]), mosquitofish (*Gambusia affinis*), Centrarchids (*Lepomis* spp.), and crayfish (*Procabarus clarkii*).

Table 2. 2017 Prey Catch Per Unit Effort (CPUE)

Quadrat ID	Trap Days	Phase	CPUE (Bullfrog)	CPUE (Mosquitofish)	CPUE (Sunfish)	CPUE (Crayfish)
BC27	1125	1	0.00356	0.07467	0.00356	0.59467
BC28A	375	1	0.00267	0.00800	0.00000	2.43467
BC28B	375	1	0.00000	0.06933	0.00000	1.95733
BC28C	375	1	0.00000	0.03733	0.00000	1.79200
BC27	1125	2	0.10756	0.21778	0.00356	0.47556
BC28A	375	2	0.01600	0.14133	0.00000	3.68533
BC28D	375	2	0.01067	0.07200	0.00267	1.73333
BC28E	375	2	0.00533	0.02933	0.00000	2.68533
BC29	30	2	0.00000	0.16667	0.10000	0.70000
BC27	1125	3	0.01600	0.05778	0.00178	0.46311
BC28E	375	3	0.00267	0.01067	0.00000	0.34667
BC28F	375	3	0.01867	0.01867	0.00267	0.27733
BC28G	375	3	0.04533	0.02133	0.00000	0.21067
BC27	1125	4	0.01244	0.07911	0.00000	0.48178
BC28I	1125	4	0.00978	0.22578	0.00800	0.26222

DISCUSSION

The number of *T. gigas* captured at the CRER has decreased substantially since trapping was last conducted, with CPUE decreasing by an order of magnitude and estimated abundance decreasing by 87%. Typical CPUE for most ostensibly stable populations can be illustrated by captures in the Natomas Basin, where overall CPUE ranged from 0.0017 to 0.0039 between 2004 and 2010 (ICF International 2011). Although Natomas Basin sites include some locations that are not occupied, these figures align well with those reported for the CRER in 2017. However, the highest CPUE we have observed at a site in a single year is 0.0356 (168 snakes captured in 4,718 trap days) (E. Hansen et al. 2010), which occurred on the CRER in 2008. Unlike the rice cultivation and managed marsh that characterize the Natomas Basin, natural topography and hydrological profiles have previously characterized the CRER. Prior to the drought, the CRER was home to the densest recorded population of *T. gigas* (USFWS 1999, E. Hansen unpublished data).

The diversity of prey species found in aquatic traps was comparable to other sites occupied by *T. gigas*, yet the abundance was somewhat low (E. Hansen, unpublished data). While information regarding suitable pH and EC ranges for *T. gigas* prey species is generally lacking, a cursory review of the scientific literature indicates that most freshwater fish species thrive in waters with a pH range of 6.5 to 9 (U.S. Environmental Protection Agency 1986), and most juvenile freshwater fishes can tolerate EC values up to 3 mS/cm without adverse effects (adults can generally tolerate EC values up to 13 mS/cm) (James et al. 2003). For most amphibian species, a pH value below 5 is usually harmful or lethal (Al-Aqtum 1999, Dale et al. 1985, Glos et al. 2003) (maximum suitable pH values were not found in the literature reviewed), and salinity levels above 4.5 parts per thousand (approximately 7.03 mS/cm EC) are considered unsuitable for the California red-legged frog (*Rana draytonii*) (Jennings and Hayes 1994); tolerable salinity ranges for other Ranid frogs (i.e. bullfrogs) are likely similar.

All pH and EC measurements obtained at the CRER fall within the tolerable ranges identified in the literature. Furthermore, all pH measurements obtained to date have fallen within the range of values observed between 2006 and 2011 at other sites occupied by *T. gigas* (range=6.79 to 9.59, mean=7.954, SE=0.025, SD=0.450, n=329 (E. Hansen unpublished data). Although *T. gigas* have been documented in habitats with EC ranging from 0.062 to 1.476 mS/cm (mean=0.6062, SE=0.0168, SD=0.3051, n=329), capture rates are typically lower where EC values are elevated (E. Hansen unpublished data).

The CREC's Badger Creek Unit is home to one of 9 distinct and separate populations of *T. gigas* recognized by the U.S. Fish and Wildlife Service (USFWS 2017). Recent genetic analysis demonstrates that *T. gigas* in Snake Marsh are relatively unique compared to *T.*

gigas in other population areas (Paquin et al., 2006; Engstrom and Olson 2007, Wood et. Al 2015). Genetic population metrics characterize a population's viability and its ability to sustain itself indefinitely. As such, these measurements of a population's genetic viability satisfy recommended recovery guidelines under the Endangered Species Act. Furthermore, by measuring genetic viability within a population over time, one can evaluate the status of a population and whether trends in the population are positive or negative.

From the standpoint of conservation and population recovery, the key genetic metric that must be measured to document a population's viability is the effective populations size (Ne). Ne is a critical metric used to gauge a population's status, genetic diversity, and its long-term viability. Importantly, genetic diversity within a population is not dictated by its census number, denoted N (i.e., the number of total individuals present on the landscape). Rather, diversity reflects how the variability in genetic information is distributed within and among individuals present in the population. Genetic variation (diversity) prevents the negative effects of inbreeding depression and provides the trait variability needed for populations to respond to environmental changes. Ne is estimated from genotype data generated from non-lethally sampled material (e.g. oral swab; tail clip); therefore, samples can be collected from individual GGS encountered during existing surveys.

Measuring Ne over time gauges a population's status and can function as an early warning system for things such as negative trends in viability over time, inbreeding, and probability of extinction. In this way Ne can also be used as a metric to gauge the effectiveness of ongoing recovery and conservation actions. If recovery and conservation actions are beneficial to the species, Ne should remain stable and possibly increase over time. Conversely, if management actions (such as those that influence water allocations to flooding and/or fallowing of rice fields) are detrimental to the species, Ne would be expected to decrease over time.

In addition to continued restoration to ameliorate the deteriorating conditions within Snake Marsh, we strongly recommend reevaluating the population's genetic structure, both now using the provided tissue samples, but into the future to evaluate the continued health of this valuable population of *T. gigas.*

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Table 1: 2017 *T. gigas* survey period dates and trapping effort

	Survey Period 1					Survey Period 2					Survey Period 3					Survey Period 4					Totals	
Quad ID	Start Date	End Date	Quad Days	Trap Days	T. gigas Caps	Start Date	End Date	Quad Days	Trap Days	T. gigas Caps	Start Date	End Date	Quad Days	Trap Days	T. gigas Caps	Start Date	End Date	Quad Days	Trap Days	T. gigas Caps	Trap Days	T. gigas Caps
BC27	5/31	6/15	15	1125	2	6/27	7/12	15	1125	10	8/4	8/19	15	1125	5	9/1	9/16	15	1125	4	4500	21
BC28	5/31	6/15	15	1125	1	6/27	7/12	15	1125	1	8/4	8/19	15	1125	4	9/1	9/16	15	1125	6	4500	12
BC29	NA					6/27	7/2	6	30	2	NA					NA					30	2

Table 2: 2017 prey catch per unit effort (CPUE)

Quadrat ID	Survey Period	Trap Days	Ranid Adult		Ranid Larvae		Mosquitofish		Black Bass		Other Sunfish		Carp		Silverside		Black Bullhead		Crayfish		Combined Prey	
			Count	CPUE	Count	CPUE	Count	CPUE	Count	CPUE	Count	CPUE	Count	CPUE	Count	CPUE	Count	CPUE	Count	CPUE	Count	CPUE
BC2	1	1	4	.0035	2	.0257	84	.0746	0	0	4	.0035	0	0	0	0	0	0	669	.0594	790	.702
	2	1	12	.1075	2	.0231	24	.2177	0	0	4	.0035	1	.0008	0	0	0	0	535	.0475	932	.828
	3	1	18	.0160	5	.0044	65	.0577	0	0	2	.0017	2	.0017	0	0	0	0	521	.4631	613	.544
	4	1	14	.0124	8	.0071	89	.0791	0	0	0	0	0	0	0	0	0	0	542	.4817	653	.580
BC2	1	1	1	.0008	0	0	43	.0382	0	0	0	0	0	0	0	0	0	0	231	2.061	236	2.10
	2	1	12	.0106	0	0	91	.0808	0	0	1	.0008	3	.1000	0	0	0	0	303	2.701	314	2.79
	3	1	25	.0222	0	0	19	.0168	0	0	1	.0008	0	0	0	0	0	0	313	.2782	358	.318
	4	1	11	.0097	0	0	25	.2257	0	0	9	.0080	0	0	0	0	0	0	295	.2622	569	.505
BC2	2	6	0	0	0	0	5	.1666	0	0	2	.0017	2	.0017	0	0	0	0	21	.7000	30	.700

Appendix B

1. BC27RS 6/1/2017

2. BC27RS 7/11/2017

3. BC27RS 8/19/2017

4. BC27RS 9/16/2017

5. BC28A 6/1/2017

6. BC28A 6/15/2017

7. BC28A 7/12/2017

8. BC28B 6/1/2017

9. BC28B 6/15/2017

10. BC28C 6/1/2017

11. BC28C 6/15/2017

12. BC28D 6/27/2017

13. BC28D 7/12/2017

14. BC28E 6/27/2017

15. BC28E 7/12/2017

16. BC28F 8/4/2017

17. BC28F 8/19/2017

18. BC28G 8/4/2017

19. BC28G 8/19/2017

20. BC28H 9/1/20178

21. BC28H 9/16/2017

22. BC29 6/27/2017

23. BC29 7/2/2017