



Lower Colorado River Multi-Species Conservation Program

Balancing Resource Use and Conservation

Relict Leopard Frog Monitoring and Management, 2009 Annual Report



August 2011

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National Park Service
Bureau of Land Management
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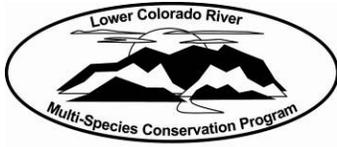
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Lower Colorado River Multi-Species Conservation Program

Relict Leopard Frog Monitoring and Management, 2009 Annual Report

*Prepared by Jef R. Jaeger, Dana Drake, and Joseph Barnes,
Public Lands Institute, University of Nevada, Las Vegas*

Lower Colorado River
Multi-Species Conservation Program
Bureau of Reclamation
Lower Colorado Region
Boulder City, Nevada
<http://www.lcrmscp.gov>

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EXECUTIVE SUMMARY

- All sites containing Relict Leopard Frogs (*Rana onca*) were surveyed at least once diurnally during spring and twice nocturnally during spring and fall.
- To better understand breeding phenology multiple additional diurnal surveys were conducted at some sites during the spring.
- Multiple surveys were also conducted at Blue Point Spring in attempts to find egg masses for head starting (none were found) and as part of mark-recapture estimation.
- The translocation program was completed by early summer. Egg masses were collected from Black Canyon sites and 1,286 frogs or tadpoles were translocated to 5 sites.
- Currently there are six active translocation sites, with Pupfish Refuge Spring no longer being augmented with head-started animals. This (2009) was the last year for translocations to Goldstrike and Grapevine (Meadview) sites.
- The latest experimental site at Quail Spring shows early success with high survivorship and evidence of successful breeding.
- Habitat conditions for successful breeding at Bighorn Spring remain extremely limited.
- Habitat conditions at Rogers Spring for any life stage appear very limited, and frogs were only seen in an extremely small patch of disturbed habitat.
- Habitat conditions at lower Blue Point Spring have deteriorated, but at upper Blue Point adult frogs appear to show high survival; however, exotic fish in this stream greatly limit recruitment.

SUMMARY

During 2009, all natural and experimental sites containing *Rana onca* (14 sites total) were surveyed at least once diurnally during spring and twice nocturnally, spring and fall (Tables 1-6). As reported previously, the experimental sites at Sugarloaf and Lower Grapevine Springs, Nevada were not surveyed as both systems had dried in previous years. As stipulated in the CAS, visual encounter surveys (VES) were used, although at upper Blue Point Spring mark-recapture efforts were continued from previous years. All surveys for *R. onca* were conducted by at least one trained biologist with experience in amphibian surveys. To improve understanding of breeding phenology, multiple diurnal VES at four coldwater and four warmwater sites were conducted through this spring; this effort began in September 2008. Multiple diurnal VES were also conducted in spring 2009 at upper Blue Point Spring and the upper 200-300 meters of lower Blue Point Spring in an effort to locate egg masses for head-starting; none were found. In general, frogs of *R. onca* were observed at all sites in 2009, and egg masses or tadpoles have been observed at 11 sites. A detailed summary of data by spring site is provided in this report.

Translocation efforts moved 1,286 frogs or late-stage tadpoles raised from eggs collected at three Black Canyon sites to five experimental sites (Table 7). Currently there are six active translocation sites, with Pupfish Refuge Spring no longer being augmented to evaluate the self-sustainability of the population. As per protocol, 2009 was also the last year for translocations to Goldstrike and Grapevine (Meadview) sites. The latest experimental site at Quail Spring shows early success with high survivorship and breeding activity.

MONITORING OF NATURAL SITES

Black Canyon Sites

Bighorn Sheep Spring, NV. – This site was targeted for numerous diurnal VES in 2009, as well as two nocturnal surveys (Tables 1 and 2). All life stages of *R. onca* except the juvenile stage were observed over the course of these surveys. Frog numbers remain low and single-digit counts of adults and egg masses have become typical during surveys. No overwintering tadpoles have been observed in recent years.

The eight pools constructed using sandbags in September 2008 were washed out during rains in December 2008, and the site is back to having very limited habitat for larval development. As part of the translocation program, portions of three egg masses observed in puddles at this site were brought in for rearing; the thought was that few animals would be recruited from these egg masses given the poor habitat conditions for tadpoles. The red-spotted toad (*Bufo punctatus*) is common at this site and reproduces here, as is true of most Black Canyon sites, except Dawn's Canyon and Salt Cedar Spring.

Boy Scout Spring, NV. – Over the course of four diurnal and two nocturnal VES at this site all life stages of *R. onca* were observed in 2009 (Tables 1 and 2). Five egg masses were found in small side pools during the spring surveys, and as part of the translocation program, portions of two of these egg masses were brought in for rearing.

Dawn's Canyon, NV. – One diurnal and two nocturnal VES were conducted at this relatively small site in 2009. All life stages of *R. onca* were observed including juveniles during the fall survey (Tables 1 and 2). A single egg mass was observed in spring, and as the pool in which it was located has limited habitat, part of the egg mass was brought in for rearing and translocation.

Salt Cedar Spring, NV. – Four diurnal and six nocturnal VES were conducted at this site during 2009 (Tables 1 and 2). All life stages, including two egg masses, were observed during these surveys.

Crayfish remained abundant in the stream and pool at the base of the survey area near the confluence with the Colorado River, but have not moved up over the earthen dam and dry section of channel into upper portions of the stream. No other amphibians have been seen in this spring in 2009. Vegetation is also starting to reestablish within the system and some areas of stream have become quite choked.

Black Canyon Spring, NV. – Seven diurnal and three nocturnal VES were conducted at this site in 2009, but only single adult *R. onca* were seen on any given survey (Tables 1 and 2) and none were seen during the fall nocturnal survey. The portion of the stream where the survey is conducted does not represent good *R. onca* habitat. Further efforts to gain a better understanding of upstream conditions (above the large waterfall used as the survey endpoint) may be useful in assessing the status of frogs in this system.

Black Canyon Spring Side Canyon, NV. – This site is off of the main Black Canyon Spring drainage and the only natural coldwater site containing *R. onca*. Eight diurnal and two nocturnal VES were conducted at this site during 2009 (Tables 1 and 2). Several large, slow pools, which looked like good frog habitat in spring, were greatly reduced by fall. Adults of *R. onca*, larvae, and an egg mass were observed at this site in 2009, but numbers were very low.

Table 1. Summary of *Rana onca* observed at natural sites in the Black Canyon during nocturnal visual encounter surveys (spring and fall) conducted in 2009. Temperatures (degrees C) are ambient air temperature during surveys (Temp^A) and water temperature (Temp^W) generally taken at sites where animals were seen. Count totals (Total) include adult and juvenile frogs. Also shown are tallies of animals observed by age stage: (A) adult, (J) juvenile, tadpoles (L), and egg masses (E).

Site	Date	Temp ^A	Temp ^W	Total (A+J)	A	J	L	E
Bighorn Sheep Spring	04/20/09	20.9	23	10	10	0	300+	5
	10/15/09	22.8	24	11	11	0	0	0
Boy Scout Canyon Spring	04/20/09	26	18	23	23	0	117	0
	10/22/09	23.3	-	21	20	1	0	0
Dawn's Canyon Spring	04/20/09	27	22	2	2	0	6	0
	10/22/09	21.7	23	8	4	4	0	0
Salt Cedar Canyon Spring	01/24/09	19.6	20	20	19	1	32	0
	02/23/09	16.2	18	33	20	13	56	0
	03/31/09	20.3	25	47	41	6	137	0
	04/27/09	26.5	24.4	42	36	6	141	2
	06/04/09	29	29.3	44	31	13	0	0
Black Canyon Spring	10/15/09	25	-	20	20	0	7	0
	03/31/09	19.3	28	1	1	0	0	0
	04/27/09	29	35	1	1	0	0	0
Black Canyon Side Spring	10/15/09	25.6	-	0	0	0	0	0
	03/31/09	19	19	7	7	0	0	0
	10/15/09	27.2	23	2	2	0	0	0

Table 2. Summary of *Rana onca* observed at natural sites in the Black Canyon during diurnal visual encounter surveys conducted in 2009 by age class: adult (A), juvenile (J), tadpoles (L), or eggs (E). Temperatures (degrees C) are ambient air temperature during surveys (Temp^A) and water temperature (Temp^W) generally taken at sites where animals were seen.

Site	Date	Temp ^A	Temp ^W	A	J	L	E
Bighorn Sheep Spring	01/12/09	13	8	0	0	0	0
	01/24/09	20.4	15	2	0	0	1
	01/29/09	14.1	10	0	0	0	3
	02/11/09	11.4	8	0	0	0	0
	02/23/09	18.2	12	2	0	0	4
	03/09/09	16.7	15	0	0	300+	0
	03/31/09	25.5	22	0	0	300+	3
	04/20/09	20.9	23	10	0	300+	5
	04/28/09	22	22	0	0	5	0
Boy Scout Canyon Spring	06/06/09	26.1	25	0	0	6	0
	01/12/09	17.9	14	9	0	0	1
	01/24/09	21	10	2	0	0	2
	02/23/09	17.2	12	10	0	53	2
Dawn's Canyon Spring	03/23/09	16	21	6	0	5	0
	01/24/09	20.5	13	1	0	0	1
Salt Cedar Canyon Spring	01/12/09	18.1	17	4	7	16	0
	02/11/09	16.5	15	2	5	21	0
	03/09/09	20.6	21	11	8	23	0
	04/16/09	20.3	25	8	9	42	0
Black Canyon Spring	01/12/09	18	24	0	0	0	0
	01/29/09	16.6	24	0	0	0	0
	02/11/09	16.1	24	0	0	0	0
	02/23/09	19.6	26	1	0	0	0
	03/09/09	22.3	25	0	0	0	0
	04/16/09	22.3	32	0	0	0	0
	06/06/09	26.6	35	0	0	0	0
Black Canyon Spring Side	01/12/09	17.4	9	1	0	2	0
	01/29/09	15.3	9	0	0	0	0
	02/11/09	12.8	9	0	0	0	0
	02/23/09	18.8	11	1	0	1	1
	03/09/09	21.4	14	4	0	0	0
	04/16/09	20.6	20	6	0	0	0
	04/27/09	28.7	21	5	0	0	0
	06/06/09	27.7	24	2	0	0	0

Northshore Spring Complex

Upper Blue Point Spring, NV. – The nocturnal surveys reported for ‘upper’ Blue Point Spring (Table 3) consist of representative spring and fall surveys from more extensive mark-recapture efforts at this site. Mark-recapture efforts included a total of six surveys in spring (between April 6 and May 24) and five surveys in fall (between September 27 and November 10). Mark-recapture estimates based on the Schumacher-Eschmeyer multiple-census method provided estimates of 91.1 (95% CI 72.6–122.2; 67 individuals observed) in spring and 69.2 (95% CI 49.3–115.9; 43 individuals observed) in fall. These data indicate high survivorship of head-started frogs placed into the system in 2008 from eggs collected at ‘lower’ Blue Point Spring. Although some evidence of natural recruitment was observed during the mark-recapture efforts, the number of juveniles appeared quite low. Habitat at upper Blue Point has benefited from the habitat improvement study conducted at this site over the last couple years, but these benefits appear short-lived and are now being reversed. This area also benefits from limited use by park visitors where trampling reduces riparian vegetation density and cover along small portions of the stream.

In addition to the nocturnal surveys at this site, 10 diurnal VES were conducted during the spring to search for egg masses and tadpoles as part of continuing augmentation effort. These surveys focused on upper Blue Point Spring, but generally included searches along the upper 200–300 m of lower Blue Point Spring within areas of favorable habitat and where egg masses have been previously seen. Only the survey on February 3 consisted of a complete survey of both upper and lower Blue Point (Table 4). Unfortunately, only adults of *R. onca* have been seen in 2009. No other amphibians have been seen at these sites this year.

Lower Blue Point Spring, NV. – At lower Blue Point Spring, nocturnal surveys continued to document adult frogs, with a relatively large number observed during the fall survey (Table 3). These frogs were observed only along the upper stretch of the stream where patches of open habitat remain. Some of these areas were manipulated as part of the habitat experiments noted above, but the system is becoming very overgrown with dense vegetation throughout. These frogs were within areas where head-started frogs were released last year as part of the augmentation effort.

Rogers Spring, NV. – Nocturnal VES at Rogers Spring have continued to document adult frogs within the system (Table 3). These frogs likely consist of head-started animals placed into the system from eggs collected at lower Blue Point in 2008. We cannot confirm that all the individual frogs observed are from that augmentation, but those that have been captured show the toe-clip mark used to identify these individuals. One diurnal survey was conducted during the spring to search for egg masses and tadpoles, but none were seen (Table 4).

Habitat conditions for *R. onca* at Rogers Spring have continued to deteriorate despite experiments over the last couple of years to improve habitat, and there are no good habitat patches other than that formed by the power line road that crosses the main stream channel and at areas just below the main springhead pool (however, few frogs have ever been seen at the upper end of this system). All the frogs observed during surveys in 2009 have been concentrated at the small opening in the otherwise dense vegetation formed by the power line road. At this time, water flows over the dirt road and aquatic vegetation is limited by the minimal road traffic and by soil compaction from vehicle traffic. No other amphibians were seen at this site in 2009.

Table 3. Summary of *Rana onca* observed at Blue Point and Rogers Springs during nocturnal visual encounter surveys conducted in the spring and fall, 2009. The surveys of ‘upper’ Blue Point Spring included here consist of representative spring and fall surveys from more extensive mark-recapture efforts at this site. Temperatures (degrees C) are ambient air temperature during surveys (Temp^A) and water temperature (Temp^W) generally taken at sites where animals were seen. Count totals (Total) include adult and juvenile frogs. Also shown are tallies of animals observed by age class: adult (A), juvenile (J), tadpoles (L), or egg masses (E).

Site	Date	Temp ^A	Temp ^W	Total (A+J)	A	J	L	E
Blue Point Spring Upper	04/06/09	21.6	-	38	38	0	0	0
	11/10/09	19.7	-	25	25	0	0	0
Blue Point Spring Lower	05/06/09	29.5	24	4	4	0	0	0
	11/05/09	21	-	16	16	0	0	0
Rogers Spring	04/30/09	20.3	24	8	8	0	0	0
	11/05/09	21.1	-	7	7	0	0	0

Table 4. Summary of *Rana onca* observed at Blue Point and Rogers Springs during diurnal visual encounter surveys conducted in the spring of 2009 by age class: adult (A), juvenile (J), tadpoles (L), or egg mass (E). An asterisk (*) indicates that the survey only consisted of the upper 200-300 m of lower Blue Point. Temperatures (degrees C) are ambient air temperature during surveys (Temp^A) and water temperature (Temp^W) generally taken at sites where animals were seen.

Spring Site	Time	Date	Temp ^A	Temp ^W	A	J	L	E
Upper Blue Point	Diurnal	01/11/09	14.5	22	0	0	0	0
Lower Blue Point*			14.2	5	0	0	0	0
Upper Blue Point	Diurnal	01/27/09	13.1	19	0	0	0	0
Lower Blue Point*			11.6	16	2	0	0	0
Upper Blue Point	Diurnal	02/03/09	20	20	1	0	0	0
Lower Blue Point			20	16	1	0	0	0
Upper Blue Point	Diurnal	02/19/09	18.6	20	3	0	0	0
Lower Blue Point*			19.7	16	3	0	0	0
Upper Blue Point	Diurnal	03/05/09	22.3	26	2	0	0	0
Lower Blue Point*			21.2	24	8	0	0	0
Upper Blue Point	Diurnal	03/12/09	17.1	23	4	0	0	0
Lower Blue Point*			20.1	21	5	0	0	0
Upper Blue Point	Diurnal	03/20/09	16.5	24	0	0	0	0
Lower Blue Point*			15	21	0	0	0	0
Upper Blue Point	Diurnal	04/03/09	15	22	0	0	0	0
Lower Blue Point*			16.7	21	0	0	0	0
Upper Blue Point	Diurnal	04/15/09	8.8	26	0	0	0	0
Lower Blue Point*			8.2	24	0	0	0	0
Upper Blue Point	Diurnal	05/01/09	28.9	29	3	0	0	0
Lower Blue Point*			26.9	26	5	0	0	0
Rogers	Diurnal	02/03/09	19.3	15	1	0	0	0

MONITORING OF EXPERIMENTAL TRANSLOCATION SITES

Black Canyon Sites

Goldstrike Canyon, NV. – Numbers of frogs observed at Goldstrike appear somewhat limited; our highest count for 2009 was 21 frogs, but these frogs were generally large and appeared healthy. We also continued to observe evidence of breeding at this site, including egg masses and non-translocated tadpoles (Tables 5 and 6). As part of the continued translocation program, 143 tadpoles were released at this site this spring.

Pupfish Refuge Spring, NV. – This site continued to be successful, with a high count this year of 40 frogs (Table 5). We also continued to note evidence of breeding in 2009 (Table 6), and at least one small, late season juvenile indicative of natural recruitment. As stipulated by protocol, translocations to this site were terminated in 2008 to assess the natural sustainability of the population.

As noted previously, exotic snails have proliferated throughout this stream and have visibly reduced algae. Indeed, in some pools algae is not readily visible, and the lack of algae is likely to have a negative impact on the growth and development of tadpoles within the main channel. Breeding, however, often occurs in the drainage ditch along the road and the snails do not appear to prefer this habitat. The Bureau of Reclamation has proposed extensive habitat restoration efforts at this site, which began on November 30 with the cutting and herbicide treatment of tamarisk on an upslope location by NPS EMPT crews. As part of this effort, personnel from UNLV have reduced vegetation and removed debris from pools in the road ditch used for breeding.

Sugarloaf Spring, AZ. – As noted in previous reports, this site was discontinued as a translocation experiment, and no survey was conducted at this site during this period.

Gold Butte Sites

Quail Spring, NV. – Quail Spring has continued as a surprising success in 2009. Observations from nocturnal surveys indicate very high survival for the initial releases of head-started frogs. The count this fall was 71 frogs (Table 5). Evidence of breeding, including calling, amplexus, and young tadpoles, has been observed at the site this year. An additional 115 juvenile frogs were released in 2009 as a continuing part of the translocation program, although the site appears saturated from the 2007 release. No official diurnal survey was attempted at this site this year. Although this site is fairly isolated, we observed calling Woodhouse toads (*Bufo woodhousii*) during the spring survey, but none were observed in the fall.

UNLV field personnel worked with BLM and a hired NCC crew to conduct habitat improvements at this site during late fall (October 28-29). The lower half of the pool was mucked out by hand with materials added to the banks to retain pool depth in the face of bank erosion from cattle and burros. Prior to the initial frog release at this site, the upper half of the pool was mucked out with the general idea that only half the pool at any one time would be impacted. As part of the effort this year, a small pool of about 3 m in length, 2 m wide, and 0.75 m deep was dug out below the main pool in the outflow channel. This pool was mostly constructed under desert willows and was lined with rubber and surrounded by rocks; two frogs were observed in this pool during the nocturnal survey five days later.

Red Rock Spring, NV. – The Red Rock Spring site continues to maintain *R. onca*, but not in large numbers (Table 5). Multiple diurnal surveys were conducted this spring to better understand breeding at this site, and both egg masses and young tadpoles were observed (Table 6). Several of the egg

masses were found on subsequent visits to have desiccated as a result of lower water levels in the pools. Cows continue to be present over much of the site, but the fencing around the main spring seep has been repaired. The site also contains *B. woodhousii* and *B. punctatus*. One hundred young frogs were released this spring as part of the translocation program at this site.

Tassi Spring – Tassi Spring continues to be a successful site with up to 87 frogs counted during nocturnal surveys this spring; fewer frogs were seen during the nocturnal fall survey but that survey was conducted by relatively novice observers (Table 5 and 6). Multiple diurnal surveys were also conducted at this site and all life stages, except juveniles, were observed this year. As part of the continuing translocation effort at this site, 223 frogs were released this year. Both *B. punctatus* and *B. woodhousii* are common at this site and both species were breeding this spring. No canyon treefrogs (*Hyla arenicolor*) have been seen or heard at this site in 2009.

The NPS EPMT crew conducted some vegetation reduction along the main ditch below the springhead in May (05/05/09). Approximately 30-50 m of vegetation was cut in one large section in and around the channel to improve flows. Some vegetation was treated with herbicide.

Other Sites

Grapevine Spring (Meadview), AZ. – Grapevine Spring, Arizona continues as a successful translocation site with large numbers of frogs, up to 115, observed on nocturnal visits (Table 5). The survey this fall went past a common stopping point for previous surveys at the waterfall, and 18 frogs were counted in the upper area. Multiple diurnal surveys were conducted at this site during the spring (Table 6); tadpoles were seen and frogs were heard calling in pools upstream of the survey area on 02/20/09, and then within the survey area on 02/25/09. As part of the translocation effort at this site, an additional 705 tadpoles were released this spring. No *H. arenicolor* have been seen or heard at this site this year.

Lower Grapevine Spring, NV. – As noted in previous reports, this site was discontinued as a translocation experiment, and no survey was conducted at this site during this period.

Table 5. Summary of *Rana onca* observed at experimental sites during nocturnal visual encounter surveys conducted in 2009 by age class: adult (A), juvenile (J), tadpoles (L), or egg mass (E). Temperatures (degrees C) are ambient air temperature during surveys (Temp^A) and water temperature (Temp^W) generally taken at sites where animals were seen.

Site	Date	Temp ^A	Temp ^W	Total (A+J)	A	J	L	E
Black Canyon Sites								
Goldstrike Canyon	04/13/09	22	18	21	21	0	15	1
	10/22/09	23.9	25	15	15	0	0	0
Pupfish Refuge	04/21/09	26.1	29	23	23	0	0	0
	11/05/09	27	-	40	39	1	0	0
Gold Butte Sites								
Red Rock Spring	03/19/09	20	14	12	12	0	0	7
	11/03/09	12.8	15	13	13	0	0	0
Tassi Spring	03/19/09	18.8	18	68	68	0	15	1
	05/05/09	23.9	24	87	82	5	12	1
	05/28/09	30.2	23	81	76	5	13	0
	11/04/09	20.6	22	18	18	0	0	0
Quail Spring	04/02/09	21	18	42	42	0	40	0
	11/03/09	19.4	21	71	71	0	0	0
Other Sites								
Grapevine Spring, AZ	03/14/09	19.5	12	47	46	1	4	4
	04/29/09	22.8	16	115	107	8	7	1
	10/21/09	23.9	18	69	69	0	9	0

Table 6. Summary of *Rana onca* observed at experimental sites during diurnal visual encounter surveys conducted in 2009 by age class: adult (A), juvenile (J), tadpoles (L), or eggs (E). Temperatures (degrees C) are ambient air temperature during surveys (Temp^A) and water temperature (Temp^W) generally taken at sites where animals were seen.

Site	Date	Temp ^A	Temp ^W	A	J	L	E
Black Canyon Sites							
Goldstrike Canyon	02/06/09	18.2	15	0	0	0	0
	03/23/09	13.5	23	0	0	57	2
Pupfish Refuge	02/04/09	23.9	22	4	0	300+	0
Gold Butte Sites							
Red Rock Spring	01/13/09	16.5	0	0	0	0	0
	01/28/09	13.3	-2	0	0	0	0
	02/10/09	12.9	0	0	0	0	0
	02/27/09	20.6	8	8	0	0	2
	04/03/09	20	11	0	0	300+	4
	05/05/09	34.8	24	3	4	0	0
Tassi Spring	01/13/09	19.1	14	11	0	0	0
	01/28/09	14.9	12	13	0	0	5
	02/10/09	18.2	13	15	0	1	1
	02/27/09	22.9	17	21	0	0	2
	04/03/09	18.1	19	23	0	2	0
Other Sites							
Grapevine Spring, AZ	01/10/09	11.4	1	0	0	0	0
	01/30/09	17	3	2	0	0	0
	02/20/09	15.2	4	5	0	0	0
	02/25/09	23.2	7	41	0	0	0
	03/10/09	24.4	13	15	0	0	2
	03/14/09	19.5	12	46	1	4	2

HEADSTARTING AND TRANSLOCATIONS

For translocation in 2009, a total of six partial egg masses from three Black Canyon populations were collected (3 from Bighorn Sheep Spring, 1 from Dawn's Canyon, and 2 from Boy Scout Canyon). These eggs resulted in a total of 1,476 hatchlings. As is common, we generally lose a percentage of the hatchlings during early rearing, but approximately 60 larger tadpoles were lost in a mortality event in one tank at the NPS laboratory this season. A handful of tadpoles and a metamorphic frog survived the event, and thoughts lean toward a run-up of temperature from a faulty heater as the cause of the loss. In total, 1,286 late-stage tadpoles and post-metamorphic frogs were released at five sites in 2009 (Table 7).

As noted above, no animals were released at Pupfish in 2009, as 2008 was the fifth year of translocations. As outlined in the CAS, experimental sites should have translocations for five years and then be assessed for sustainability. As per this protocol, 2009 was the fifth year of translocations

to Goldstrike and Grapevine (Meadview) springs and further augmentations will not be conducted over the next several years to assess the success of populations at these sites.

For rearing this spring, 796 early stage tadpoles were taken to Willow Beach National Fish Hatchery. Because of concerns over calcium deficiency in the tadpoles encountered last season, Willow Beach had samples of the water from the new warmwater well (first used in 2008) and old coldwater well (in which tadpoles had been reared prior to 2008) analyzed for calcium content in February. The calcium concentration was 137 and 108 ppm, respectively. The tadpole diet was modified at the hatchery this year to include alfalfa-based rabbit chow, romaine, endive, mustard and dandelion greens, spirulina, and flake fish food. Cuttlebones were also added to the raceway to allow the tadpoles to graze directly on a calcium source if desired. The raceway used in 2009 is within a couple of feet of a window, allowing for additional natural lighting. Despite these efforts, some tadpoles still developed kinked tails later in development, as was seen with the tadpoles in 2008 at the hatchery. Tadpoles from each egg mass collected from the field were also kept at the NPS facility and shared greens and rabbit chow with the hatchery in an effort to address specific potential issues in diet or genetics as possibilities for developmental issues seen with tadpoles at the hatchery. No tadpoles in the NPS laboratory developed kinked tails, and tadpoles returned from the hatchery and raised for a couple weeks at the NPS facility developed into visibly healthy frogs and were released. There are preliminary plans to conduct experiments at the hatchery in 2010 to determine whether the water source may be at issue.

Table 7. Tadpole and post-metamorphic frog release data for *Rana onca* translocation in 2009.

Date	Translocation Site	Tadpoles Released (n)	Frogs Released (n)	Total
04/16/09	Goldstrike Canyon	143	0	143
03/15/09	Grapevine, AZ	705	0	705
04/02/09 & 04/18/09	Quail Spring	0	50 & 65	115
04/06/09	Red Rock Spring	0	100	100
04/06/09 & 04/18/09	Tassi Spring	0	123 & 100	223
Total		848	438	1286

Translocation Site Reconnaissance

On 01/29/09, UNLV and NPS personnel investigated a small, unnamed warm spring located on the Nevada side of the Black Canyon, just upstream of Dawn’s Canyon (NAD 83, 703550, 3985200). No frogs were seen during the survey. The site was thick with cattail, arrowweed, and tamarisk. The main spring pool and small side pool contained crayfish of several sizes, and the site does not likely harbor *R. onca* nor does it seem like a good potential site for translocations.

Site visits were conducted by UNLV personnel to Stewart Ranch and Perkins Pond in fall (08/03/09 and 9/10/09) to consult with BLM personnel and their private consultant regarding modifications to these sites for future translocations.

On 09/23/09, UNLV personnel investigated Chill Heal Spring, near Grand Wash, Arizona (NAD 83, 213238, 4011975, zone 12). No *R. onca* were seen during the survey, although 12 adult *H. arenicolor* and 3 unidentified large black tadpoles (≤ 5 cm) were observed primarily mid-way up the stream course. Springsnails were observed on the lower portion of the spring. The cool spring site consisted of approximately 100 m of flowing stream and intermittent pools before it became choked with

vegetation (cattail, arrowweed, etc.). This site is impacted moderately by cattle and has potential as a translocation site for *R. onca*. The site was previously visited on 01/22/09 and had flow equal to, or greater than, the flow observed on the current survey, and it is listed as persisting year-round in an assessment of springs completed for Grand Canyon-Parashant National Monument in 2008.

On 09/24/09, UNLV personnel investigated Burro Spring, east of Temple Bar, Arizona (NAD 83, 755599, 3989037). No frogs were seen during the survey. This was a follow-up visit after tamarisk was removed in 2006 by an NPS EPMT crew. The site had good flow and several areas relatively free of vegetation, with large pools interspersed among areas thick with cattails and *Phragmites*. The site contained large numbers of crayfish in the lower portions and a few crayfish were also observed in an upper stretch of stream, and so is not likely a good potential site for translocations.