FWCP Northern Leopard Frog Project: 2014 Field Season Report

contains sensitive information – not for distribution



Submitted to: Fish and Wildlife Compensation Program-Columbia Basin

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April 2015 Revised

ACKNOWLEDGEMENTS

Thanks to the Fish and Wildlife Compensation Program (FWCP) – Columbia Basin for administering this project, to Irene Manley for project support, Amy Waterhouse for GIS support and mapping and Trevor Oussoren for providing valuable direction as FWCP-Columbia Program Manager. Special thanks to the field staff, Claire Schadeli, Emily Ackroyd and Thomas Hill for many long hours, late nights and dedication in the field, and to Kat McGlynn for her help during the summer YOY surveys. Thanks also to Vince Nitchie for his volunteer efforts throughout the field season. Acknowledgements also to Marc-Andre Beaucher and the Creston Valley Wildlife Management Area (CVWMA) for providing support and allowing us to carry out our fieldwork on the WMA.

SENSITIVE SPECIES

Since the northern leopard frog (*Lithobates pipiens*) is listed on the *Provincial List of Species Considered Data Sensitive*, and *all sites* are considered sensitive site information for this species due to *the potential for indirect harassment*, all geographic location information is secure (locked) and is not openly available to the public. This report contains specific geographic information and maps of northern leopard frog locations as it was prepared for internal use by the FWCP and members of the British Columbia Northern Leopard Frog Recovery Team (BC-NLFRT) and is not intended to be distributed beyond that audience.

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1. INTRODUCTION

The main objectives for the 2014 field season were to monitor the British Columbia remnant wild population of northern leopard frogs (*Lithobates pipiens*), in the Creston Valley Wildlife Management Area (CVWMA), bolster the reintroduced population at Bummer's Flats by continuing with phase 2 reintroductions from Creston and provide additional founder tadpoles to enhance the captive assurance colony at the Vancouver Aquarium.

2. METHODS

In order to meet the objectives listed above, 6 types of survey methods were utilized during the 2014 field season: Songmeters, nocturnal calling surveys, egg mass surveys, visual encounter surveys, road surveys and tadpole trapping.

2.1 Songmeters

Two separate projects using Songmeters (produced by Wildlife Acoustics Inc.) were carried out during the 2014 field season. During the first project, 2 Songmeters were deployed at DLNA, one at WDLNA and another at the EDLNA pond. They were deployed from March 25-April 29 and set to record daily from 20:00 to 22:00 to determine when spring calling begins; however due to a malfunction the unit at the EDLNA pond only recorded from March 25-March 31. The second project involved setting up 4 Songmeters in the northern region of Leach Lake, in ponds 3 and 4; each compartment had 2 units deployed from May 20-June 5 which were set to record daily from 21:00-23:00. Ideally the Songmeters would have been deployed in early May but we were awaiting replacement microphones. The map in Appendix 1 shows the locations of the Songmeters in 2014.

Data analysis (presence/not-detected) was carried out using Songscope software, a spectrogram viewing program developed by Wildlife Acoustics Inc.; .wav files were scanned at high speed while visually searching for the digital signature of a *Lithobates pipiens* (LIPI) call and verified by listening to any positive matches.

2.2 Nocturnal Calling Surveys (NCS)

The main objective of the NCS is to monitor breeding activity and determine the number of calling LIPI in each area during the breeding season. The methodology for calling surveys follows that outlined by the Resource Inventory Committee (Heyer et al. 1994, MELP, 1998) with modifications. Calling surveys were only carried out at the CVWMA in 2014; no NCS were conducted by our crew at Bummer's Flats.

NCS are carried out as night falls during the breeding season (late April to early June) at fixed calling stations (for CVWMA NCS stations see map in Appendix 1). Two types of NCS are utilized: standardized auditory surveys and blitz style surveys.

Standardized Auditory Surveys (SAS)

The SAS methodology is used at fixed calling stations in known breeding areas to approximate the relative abundance of calling males and determine their locations for follow up egg mass searches. Surveys begin with one minute of silence to ensure that any disruptions created by the surveyor reaching the site do not impact the calling activity. The survey itself consists of three 3-minute intervals, each separated by 1 minute (Adama and Beaucher, 2006). The surveyor listens and estimates the number of calling LIPI during each interval, and at the end of the survey also estimates the minimum and maximum number of calling LIPI individuals heard during the entire survey.

Blitz Style Surveys

Blitz style surveys are used in areas where breeding is not documented regularly, but either has been in the past or the area contains suitable habitat so it is reasonable to think that breeding may occur. Surveyors listen for a minimum of 7 minutes at each station and record the number of LIPI detected calling.

Efforts are made to survey each of the main breeding areas at least once per week, and if time permits conduct blitz style surveys at other areas at least once during the peak of the breeding season; all surveys are weather permitting. Surveys are cancelled when winds are excessive or there is heavy rain, as it would be impossible to detect calling activity under these conditions. Surveys are also cancelled if the air temperature drops below 5°C, as it is not likely LIPI would be calling.

Upon arrival at the first station within a breeding site the start time is recorded, GPS location noted, and environmental conditions documented, including: air temperature, water temperature, cloud cover, precipitation (current and last 24 hours), approximate wind speed, pH, and conductivity. NCS are then carried out and special attention is given to determine the location of each calling LIPI, as this enables the surveyors to pinpoint the area in which they should focus their efforts during the egg mass survey the next day. Once the NCS is completed, the end time, end air temperature and end water temperature are noted. If time permits, the surveyor attempts to capture and processes some of the calling male LIPI, starting with the core calling groups in the area (see Section 2.8 Animal Capture and Tissue Collection for methods). Capture is important as it not only enables surveyors to pinpoint the exact location of the calling males, which allows them to determine the focal areas for follow up egg mass searches, but it also enables them to identify the age class of the calling males. Determining the age class (whether juvenile or adult) indicates if they are of reproductive age or not and is important information taken into account when prioritizing search areas for follow up egg mass searches. By capturing the animals, surveyors are also able to collect morphometrics, assess the animal's health, collect a swab for chytridiomycosis testing and take a dorsal spot pattern photo for the recognition image library.

2.3 Egg Mass Surveys (EMS)

Egg mass surveys are conducted to monitor and quantify the breeding activity of LIPI during the spring breeding season (late April to early June), which gives a measure of the minimum size of the breeding population so it can be compared from year to year to give an indication of how the population is doing. It is through these surveys that egg masses are located, which are the source for the Bummers Flats reintroduction program and the additional founders provided each year to the Vancouver Aquarium captive assurance colony.

It has been found that the most efficient way to conduct EMS is to return to the breeding site where a NCS was conducted within the last few nights, as this enables the surveyor to focus on key areas of concentrated calling; due to the vast amount of habitat there is not enough time to search all areas so this method increases efficiency. At the beginning of the survey environmental conditions as described above (for NCS) are recorded and a GPS tracklog is started, then the surveyors begin searching the area systematically on foot. Weather is a very important factor in conducting EMS, as any wind, rain, or cloud cover can obscure the surveyor's visibility, even though polarized sunglasses are always used. When an egg mass is detected, surveyors immediately make every effort to neutralize disturbance to the area, to prevent siltation of the egg mass. Once that is completed, the location is marked by GPS, egg mass volume and water depth measurements recorded, and the vegetation species of attachment noted; a photo is also taken when possible. The condition of the egg mass, percent fertilization and estimated age (based on development) is noted, and finally, the area is marked by a ring of flagged stakes or cattail stems driven into the substrate (at a radial distance of 2 meters from the egg mass). When possible, egg mass enclosures (see photo in Figure 1) are utilized to facilitate collection of hatchlings for the Vancouver Aquarium captive assurance colony, the reintroduction at Bummers Flats and to provide protection from predators until free swimming. Whether or not egg masses are caged depends on numerous factors, including the developmental stage, position in the water column, attachment and general condition. When caged, native aquatic vegetation is added to the enclosure and any potential predatory aquatic invertebrates are removed; bladderwort, Genus Utricularia was not used as it is a carnivorous plant and can cause mortality.



Figure 1. Egg mass enclosure

Since capturing and processing LIPI is not the main objective of an EMS, LIPI that are observed during these surveys are generally just recorded as an auditory or visual observation with a UTM to mark the location and are not captured and fully processed as this would take away time from the main priority of the EMS which is searching for egg masses.

2.4 Visual Encounter Survey (VES)

The main objective of the VES is to gather information on the health and status of the northern leopard frog population, as well as to get an indication of habitat use, dispersal patterns and migration corridors. As habitat use for the northern leopard frog varies by life stage and season, VES are targeted towards specific life stages during different seasons. During the summer, daytime VES are carried out in an effort to target LIPI young of year (YOY) in their preferred habitat within natal ponds before late summer and fall dispersal. During late summer and fall, migration corridors and over-wintering areas are surveyed during the daytime in the CVWMA; methodology usually involves walking the perimeter of the water body searching for LIPI, but in some cases surveys are conducted by canoe.

LIPI are captured and processed using methods outlined in Section 2.8 Animal Capture and Tissue Collection.

2.5 Tadpole Trapping

The main objective of tadpole trapping is to gather presence/not detected data. The primary focal area in 2014 was Leach Lake pond 4 to determine if there was any evidence of breeding since calling was detected during the spring breeding season. Tadpole trapping was also conducted at EDLNA ponds, a known breeding site to determine the efficacy of revised methods and to establish what developmental stage tadpoles were at. See Appendix 1 for location of tadpole traps within the CVWMA and Appendix 9 for tadpole trap locations in Bummers Flats.

Tadpoles are trapped using un-baited 21 inch wire minnow traps (shown in Figure 2). The traps are partially submerged in warm shallow areas with high levels of emergent vegetation along the perimeter of the marsh. They are set up so that at least a portion of the trap is up out of the water to prevent drowning of post-metamorphic individuals even though there is a very small probability they would enter and not escape on their own. Traps are checked within 8-12 hours to decrease the likelihood of predation on trapped tadpoles and everything trapped is released on site. If handling tadpoles for identification to species level Nitrile gloves are used as there is evidence latex gloves can be harmful to tadpoles (Gutleb, 2001).

If necessary either the United States Geological Survey (USGS) document entitled *Tadpoles* of the United States and Canada: A Tutorial and Key tadpole key (Altig et al, 1998) or the field guide entitled Amphibians of Oregon, Washington and British Columbia: a field identification guide (Corkran and Thoms, 1996) were consulted to identify tadpoles to the species level.



Figure 2. Wire minnow trap used for tadpole trapping

2.6 Late-summer and Fall Migration Road Surveys

The objective of late-summer and fall migration road surveys is to gather information on road mortality along the Duck Lake dike while moving from summer foraging grounds in the DLNA to the primary over-wintering area at the Goat channel in late summer and early fall. To avoid causing any road mortality, surveyors park their vehicle outside the migration corridor and begin walking or very slowly cycling the roadway starting at dusk and note the location of all LIPI encountered (live or dead) by GPS. If time permits, live animals are captured and processed using methods outlined in Section 2.8 Animal Capture and Tissue Collection; if time is restricted a dorsal spot pattern photo and UTM location is taken at a minimum. Dead frogs are removed from the roadway to ensure they are not double counted and mortally wounded animals are euthanized.

2.7 Animal Capture and Swab Collection for Chytridiomycosis Testing

During all survey types, when time permits effort is made to capture LIPI detected for full processing (recorded as a capture). When the surveyor is unable to make a capture, or time prohibits it, the observation is noted as a *visual*; if it is heard calling, but not seen or captured, it is recorded as an auditory detection; if only a photo and UTM are taken it is recorded as a photo only observation. Each animal that is captured is processed to collect information about the physical attributes, including: snout to vent length (SVL), weight, health (good, fair, poor, dead), visible signs of chytridiomycosis, age class (YOY, juvenile, adult), and sex. Animals are not usually checked for visual implant elastomer (VIE) marks any longer, unless they are very large individuals, since the last detection of a VIE marked individual from the 2001-2005 captive rearing program was in 2008 and it is not believed that any of the VIE marked frogs are still alive. The UTM location of all observations is marked by GPS, the habitat features and distance to shore noted, and a digital photo of the animal's dorsal spot pattern is taken for identification. When an animal is captured, the surveyor puts on a pair of single-use disposable gloves, and place the animal into a one-time use Ziploc bag. The animal is then weighed, measured, visually assessed for health and any signs of chytridiomycosis, swabbed, put back in the net for a photo and then released as quickly as possible to minimize stress (processing occurs in order listed and the length of time the animal is in the bag is very minimal). Photos documenting the process of weighing, SVL measurement and swabbing are provided in Figure 3. Fieldwork methods follow the Ministry of Environment Standard Operating Procedures: Interim Hygiene Protocols for Amphibian field staff and researchers (MoE 2008).

The health of each LIPI captured is assessed in the field by visual inspection. The surveyor looks for any abnormalities or injuries and for signs of chytridiomycosis such as sloughing skin, redness, vascularisation, lethargy, abnormal body positioning, or loss of righting reflex. General health and whether or not an animal is suspected of a chytridiomycosis infection is noted. An animal's general health is defined to be *good* if it has no injury or signs of illness; it is deemed to be *fair* if it has a minor injury such as a wound, which it is expected to fully recover from, or minor symptoms suspected to be from chytridiomycosis; and it is considered to be in *poor* health if it has a major injury, that will likely cause death, or if it is showing major signs of disease, such as chytridiomycosis. If chytridiomycosis is suspected, details of the symptoms are noted. If health is deemed to be anything other than good, the reason for the designation is noted and where possible photos taken.

For all animals captured, ventral body swab samples are collected to test for the presence of *Batrachochytrium dendrobatidis* (*Bd*), which causes the disease chytridiomycosis. Body swabs are taken using sterile Mediwire MW100 rayon tipped swabs stored in a dry labelled test tube. The animal is swabbed a total of 33 times, in the following order to minimize the spread of *Bd*: 5 times on each side, 5 times on the ventral surface, 5 times on each thigh, and once on the webbing between each toe. Once swabbing is completed the swab is put back into the sterile, dry test tube, labelled with observation number, species, sex, age class, site, health and whether or not *Bd* was suspected. Swabs are stored in the refrigerator until submitted to the lab for testing. The swab is not stored in any type of fixative, as it impairs the DNA extraction process, and is not required to maintain the integrity of the *Bd* DNA.

If the number of swabs collected during the field season is in excess of what the budget for lab analysis will cover, some are discarded; adult samples are the highest priority followed by juvenile and finally YOY. Tissue samples are analyzed by the Animal Health Centre of the Ministry of Agriculture Fisheries and Food Lab in Abbotsford, BC for the presence of *Bd* DNA using Polymerase Chain Reaction (PCR) assay under the direction of Dr Tomy Joseph using methods developed by Boyle (Boyle, 2004).

If a recently deceased animal is found in relatively good body condition (i.e., decomposition has not begun) it is immediately submitted to Dr Stephen Raverty of the Animal Health Centre of the Ministry of Agriculture Fisheries and Food Lab in Abbotsford, BC for a full work up to determine the cause of death and general condition.



Figure 3. Photos (left to right): the process of weighing, measuring SVL and swabbing for *Batrachochytrium dendrobatidis* (*Bd*), which causes the disease chytridiomycosis.

2.8 Dorsal Spot Pattern Recognition

Dorsal spot pattern photos were taken for all captures during the 2014 field season, they were labelled by observation number, cropped, resized, and organized by site and a library of 2014 images was created. However, it was decided that the recognition would not be completed this year as it has in the past due to the large amount of effort required to do it.

2.9 Vancouver Aquarium Captive Assurance Colony

In 2014 the Vancouver Aquarium requested 40 additional founder tadpoles for the captive assurance colony from 4 different egg masses, this is fewer than in previous years due to the fact that they are experiencing space constraints. Tadpoles awaiting transfer to Vancouver were held and cared for in 4 different egg mass enclosures in situ at the WDLNA natal site. Native vegetation with algal growth for grazing is added to each enclosure to provide a food source and shade for the developing hatchlings; when set up, potential predatory aquatic invertebrates are removed from the cages. Enclosures are carefully monitored; native vegetation replaced as needed and efforts made to keep it free of large amounts of waste build up. Once ready for transfer to the Vancouver Aquarium tadpoles are collected from the natal site in thermos coolers filled with 1/3 water and transported carefully, as quickly as possible to the CVWMA office where they are held overnight and packaged the following morning in plastic bags used for live fish transport with 1/3 water and 2/3 air tied off by an elastic band (Figure 4); no food is packed in the bags for transport to prevent the buildup of wastes (as recommended by the Vancouver Aquarium, Lee Newman, pers comm.).



Figure 4. Photos (left to right): egg mass enclosure to house tadpoles in situ until ready for transfer to Vancouver, Thermos used to collect tadpoles for transport to CVWMA, packaged tadpoles for transport to Vancouver Aquarium

2.10 Bummers Flats Reintroductions

In order to bolster the reintroduced population at Bummers Flats that was started in 2003 a second phase of reintroductions was started in 2011, using eggs and/or hatchlings from wild laid egg masses in Creston. It was decided by the British Columbia Northern Leopard Frog Recovery Team (BC-NLFRT) that if greater than 10 egg masses were detected in Creston, a portion of each one above that level would be moved to Bummers Flats. This year the source of all animals was WDLNA and all reintroduced animals were moved at the hatchling stage, upon becoming free-swimming; where logistically feasible a portion (ideally not more than

half) of the healthy egg masses detected over and above the threshold level of 10 were moved in insulated Thermos containers. Pond water was placed in each container to approximately the 1/3 level and hatchlings were collected from egg mass enclosures using stainless steel strainers and quickly transferred into the Thermos for transport to the recipient site. Methods for transport developed by Kendell and Prescott (2007) in the *Northern Leopard Frog Reintroduction Strategy for Alberta* were utilized with some minor modifications. Once at the release site, egg mass cages were set up to hold hatchlings overnight and vegetation native to the site was added (bladderwort *Genus Utricularia* was not used as it is a carnivorous plant and can cause mortality), with special attention to ensure no invertebrates were added to the enclosures. The following day, hatchlings were counted and released at various locations along the length of the release ditch. Figure 5 shows photos of some of the steps in the reintroduction process.



Figure 5. Photos (left to right): Thermos container with hatchlings for transport to reintroduction site; field technician Claire Schadeli carrying containers; Bummer's Flats reintroduction site with caged hatchlings and *please do not disturb* signage.

3. RESULTS

Throughout the results, observations are noted as 1 of 4 different detection types: auditory, capture, visual or photo only. Since it was decided dorsal spot pattern recognition would not be done this year there is no information available on recaptures. As a result all data summaries are based on point in time observations of animals or detections; recaptures (if any) are not taken into account so the data does not reflect the number of unique individuals.

3.1 Songmeters

Data analysis of .wav files from 2 Songmeters deployed for the early spring (March 25 – April 29) at known breeding areas at DLNA to determine when calling first started indicate that calling began on April 9 at WDLNA. Unfortunately due to a malfunction the Songmeter deployed at EDLNA ponds only recorded from March 25 – 31; no calling was detected during this period.

Through data analysis of .wav files calling was detected on 3 of the 4 Songmeters deployed at Leach Lake from May 20 - June 5. Calling was detected on unit D deployed in the northern section of pond 3 on 4 nights (May 26, 29 and June 2, 3), on unit B deployed in the northern section of pond 4 on 4 nights (June 1 - 4) and on unit A deployed in the south-eastern section of pond 4 on 5 nights (May 24, 27, 29, 30, 31). The 3 calling males detected in-person during a NCS in pond 4 (a distance of 386 m away from unit A) on May 22 were not detected on the Songmeter recordings. No calling was detected on unit B deployed in the southern section of pond 3. See Appendix A for location of deployed Songmeters.

3.2 Survey Effort

In total 438:44 person-hours were spent surveying for LIPI in the CVWMA and Bummer's Flats (n=135 surveys) between January 17 and October 20, 2014. Of this total, 429:22 person-hours were spent surveying at the CVWMA (n=128 surveys). Table 1 outlines the effort put forth by survey type at the CVWMA during the 2014 field season and Table 2 indicates the number of surveys completed. Survey effort is reported in person-hours (survey time multiplied by the number of surveyors).

Table 1. Survey effort in person-hours (hh:mm) by survey type at CVWMA

| Season | NCS | EMS | Incidental | Road | Tadpole | VES | Total |
|--------|-------|--------|------------|-------|---------|--------|--------|
| Spring | 98:29 | 120:52 | 07:12 | * | * | 01:09 | 227:42 |
| Summer | * | * | 00:05 | 23:26 | 13:21 | 111:01 | 147:53 |
| Fall | * | * | 00:05 | 13:01 | * | 39:06 | 52:12 |
| Winter | * | * | * | * | * | 01:35 | 01:35 |
| Total | 98:29 | 120:52 | 07:22 | 36:27 | 13:21 | 152:51 | 429:22 |

^{*} indicates that no surveys of this type were carried out during this time period

Table 2. Number of surveys in the CVWMA by survey type per season in 2014

| Season | NCS | EMS | Incidental | Road | Tadpole | VES | Total |
|--------|-----|-----|------------|------|---------|-----|-------|
| Spring | 21 | 16 | 2 | * | * | 1 | 40 |
| Summer | * | * | 1 | 8 | 8 | 45 | 62 |
| Fall | * | * | 1 | 5 | * | 19 | 25 |
| Winter | * | * | * | * | * | 1 | 1 |
| Total | 21 | 16 | 4 | 13 | 8 | 66 | 128 |

^{*} indicates that no surveys of this type were carried out during this time period

At Bummer's Flats 9:20 person-hours were spent surveying (n=7 surveys). Of the total, 6:47 person hours were spent on tadpole trapping surveys (n=6 surveys) and 2:33 were spent summer VES for YOY (n=1 survey).

3.3 Nocturnal Calling Surveys (NCS)

In the CVWMA, 98:29 person-hours were dedicated to NCS (n=21 surveys) during the peak breeding season from April 28 to June 6; this tally of survey hours includes time spent conducting blitz style surveys, standardized auditory surveys (SAS) and time spent locating and capturing calling males. During these surveys 167 LIPI detections were made; catch per effort was 1.7 LIPI per person-hour of NCS (Table 3).

Table 3. Summary of NCS efforts in the CVWMA

| | NCS |
|------------------------------|-------|
| Number of surveys | 21 |
| Survey effort (person-hours) | 98:29 |
| Number of LIPI observations | 167 |
| LIPI catch/effort during NCS | 1.7 |

Table 4 summarizes the LIPI observations by stage and detection type during NCS. Of the 167 total observations, 43 were adults (42 male, 1 female), 52 were juvenile (49 male, 3 sex unknown) and 72 were unknown age class (all males). Of the observations, 45 (27.0%) were auditory (calling heard only), 56 (33.5%) were captures and 66 (39.5%) were visuals (seen but not captured and no photo taken). Locations of LIPI observations in the CVWMA are provided in maps in Appendices 2, 3 and 7.

Table 4. LIPI observations by stage and detection code during NCS at CVWMA

| Detection code | Adult | Juvenile | Unknown | Total |
|-----------------------|-------|----------|---------|-------|
| Auditory | * | * | 45 | 45 |
| Capture | 25 | 31 | 0 | 56 |
| Visual (no photo) | 18 | 21 | 27 | 66 |
| Total | 43 | 52 | 72 | 167 |

^{*}Auditory detection so impossible to determine age class so assigned to age class unknown

Within the CVWMA 21 NCS were conducted, these include: SAS stations in WDLNA (n=8 surveys) plus 2 reconnaissance level surveys into new habitat in the southern portion of the area, EDLNA ponds (n=3 surveys), East ditch (n=3 surveys), Leach Lake pond 4 (n=2 surveys) and Leach lake blitz (n=2 surveys). See map in Appendix 1 for locations of calling stations. Calling was detected at WDLNA and EDLNA ponds.

As shown in Table 5, calling activity was documented at WDLNA, the EDLNA pond, Leach Lake pond 4 and new areas in the south-west and middle portions of DLNA (see Appendix 3 for map showing location of calling males in DLNA by age class and Appendix 7 for map showing location of calling males in Leach Lake pond 4 by age class). Of these sites, the WDLNA had the greatest amount of calling activity with at least 53 calling males detected May 20-21 (dates pooled since it is not possible to survey the entire area in one night due to the large number and widespread location of calling males); calling activity was documented at the site each week it was surveyed from April 28-June 1 (not surveyed week 1), calling was not detected during the last week it was surveyed (June 2-6). At the EDLNA ponds calling was detected each week it was surveyed from April 24-June 1 (not surveyed week 5 or 7) the greatest amount of calling was detected on May 13, when 9-12 LIPI were detected calling. At pond 4 of Leach Lake calling was detected on May 21 and 22 only, when there were 3 LIPI detected calling; calling was not detected during surveys at the site during week 2, 4 or 6.

Table 5. Summary of LIPI calling detected (yes or no) by site during breeding season

| Week # | Date range | WDLNA | EDLNA ponds | East ditch | Leach #4 | New** |
|--------|----------------|-------|--------------------|------------|----------|-------|
| 1 | April 24-27 | n/a* | yes | n/a* | n/a* | n/a* |
| 2 | April 28-May 4 | yes | yes | no | no | n/a* |
| 3 | May 5-11 | yes | yes | no | n/a* | yes |
| 4 | May 12-18 | yes | yes | no | no | yes |
| 5 | May 19-25 | yes | n/a* | n/a* | yes | n/a* |
| 6 | May 26-June 1 | yes | yes | no | no | n/a* |
| 7 | June 2-6 | no | n/a* | n/a* | n/a* | n/a* |

^{*} n/a means site not surveyed for that week

Due to the fact that there was so much calling detected in the spring of 2014 coupled with the fact that it was so widespread across the landscape it was not possible to document it all. As a result, a Table summarizing amount of calling activity by week and a Figure showing proportion of calling males by week is not provided here as it was in previous years because it would not be representative of the total amount of calling.

By looking at the total number of observations of male LIPI of known age class (n=91) encountered during NCS provided in Table 6 (recaptures included) we can get an estimate of the observed adult to juvenile male ratio during NCS. The data shows an estimated adult to juvenile male ratio of 6:7 (42 of 91 (46.2%) adults and 49 of 91 (53.8%) juveniles); however, it is important to keep in mind that this ratio cannot be assumed to be a direct representation of the actual breeding male population as recaptures are included in this estimate and more importantly, surveyors are not able to capture all males during the spring breeding season. Mean weight of adults from this dataset was 55.1 g (n=24, SD=5.7, range 50.0 - 73.0), mean

^{**} new site in south west region of DLNA (see Appendix 4 for location of these calling males)

SVL of adults was 79.5 mm (n=24, SD=4.4, range 71.6-88.8). For the juvenile dataset, mean weight was 42.3 g (n=31, SD=7.1, range 22.0-49.0) and mean SVL was 73.1 mm (n=31, SD=5.5, range 58.4-81.6).

Table 6. Observed ratio of juvenile to adult male LIPI from NCS at DLNA (includes recaptures)

| | | | | \overline{x} weight | | | | \overline{x} SVL | | | |
|-----------|----|--------|-------|-----------------------|----|-----|-------------|--------------------|----|-----|-------------|
| Age class | # | % | ratio | (g) | n | SD | range | (mm) | n | SD | range |
| Adult | 42 | 46.2% | 6 | 55.1 | 24 | 5.7 | 50.0 - 73.0 | 79.5 | 24 | 4.4 | 71.6 - 88.8 |
| Juvenile | 49 | 53.8% | 7 | 42.3 | 31 | 7.1 | 22.0 - 49.0 | 73.1 | 31 | 5.5 | 58.4 - 81.6 |
| Total | 91 | 100.0% | 13 | 47.9 | 55 | 9.1 | 22.0 - 73.0 | 75.9 | 55 | 6 | 58.4 - 88.8 |

No NCS were conducted by our crew at Bummer's Flats in 2014.

3.4 Egg Mass Surveys (EMS)

In the CVWMA, there were 120:52 person-hours (n=16 surveys) dedicated to EMS between April 24 and May 27, 2014 (Table 7). EMS were not conducted at any other sites in 2014; no EMS were conducted at Leach Lake as the area where calling was detected in pond 4 was flooded reed canary grass and it would be impossible to detect an egg mass there. In total, there were 39 egg masses detected in the CVWMA (see Appendix 1 for map of egg mass locations). This amount of effort translates to a catch per effort of 0.32 egg masses per hour of survey-effort.

Table 7. Summary of LIPI detection rate during EMS

| | 0 |
|------------------------------|--------|
| | EMS |
| Number of surveys | 16 |
| Survey effort (person-hours) | 120:52 |
| Number of LIPI observations | 77 |
| LIPI catch/effort during NCS | 0.64 |

As shown in Table 8, EMS effort by site, there were 97:18 person-hours of survey time in WDLNA (n=11 surveys) and 23:34 person-hours of survey time in EDLNA ponds (n=5 surveys). Given that there were 33 egg masses detected in WDLNA, this is a detection rate of 0.34 egg masses per person-hour of survey effort; at EDLNA ponds there were 6 egg masses detected, a detection rate of 0.26 egg masses per person-hour of survey effort.

Table 8. Summary of egg mass detection rate by site in the CVWMA for spring 2014

| | WDLNA EMS | EDLNA pond EMS | Grand Total** |
|--------------------------------|-----------|-----------------------|----------------------|
| Number of surveys | 11 | 5 | 16 |
| Survey effort (person-hours) | 97:18 | 23:34 | 120:52 |
| Number of egg masses detected* | 33 | 6 | 39 |
| Egg mass catch/effort | 0.34 | 0.26 | 0.32 |

^{*}includes 5 detected outside of EMS effort (0.28 if not included)

^{**}both sites combined

Table 9 summarizes the LIPI observations by stage and detection type during EMS. Of the 77 total observations, 46 were adults (44 male, 2 female), 18 were juvenile (17 male, 1 sex unknown), and 13 were unknown age class (12 males and 1 unknown sex). Of the observations, 7 were auditory (9.1%), 16 (20.8%) were captures, 3 (3.9%) had photos taken only and 51 (66.2%) were visuals. Location of LIPI observations in the CVWMA are provided in maps in Appendices 2, 3 and 7.

Table 9. LIPI observations by stage and detection code during EMS at CVWMA in 2014

| Detection code | Adult | Juvenile | Unknown* | Total |
|-----------------------|-------|----------|----------|-------|
| Auditory | 0 | 0 | 7 | 7 |
| Capture | 9 | 7 | 0 | 16 |
| Photo only | 3 | 0 | 0 | 3 |
| Visual (no photo) | 34 | 11 | 6 | 51 |
| Total | 46 | 18 | 13 | 77 |

^{*}unknown age class because auditory or visual detections

The greatest proportion of egg masses were detected at WDLNA, where 33 of the total 39 egg masses were found, detection dates were: April 30 (n=3), May 1 (n=5), May 5 (n=1), May 6 (n=2), May 7 (n=5), May 8 (n=4), May 13 (n=4), May 14 (n=4) and May 15 (n=2), May 21 (n=2) and May 22 (n=1). There were 6 of the total 39 egg masses detected at EDLNA ponds, detection dates were: April 25 (n=1), April 28 (n=1), May 6 (n=2) and May 8 (n=2). While there were no egg masses detected at Leach lake, there were YOY detected in the summer, providing evidence that there was successful breeding somewhere in Leach lake in the spring of 2014 (exact location unknown).

Of the 39 egg masses detected in 2014, 34 were detected as pre-hatch egg masses and 5 were detected in the process of hatching.

Of the 34 detected as pre-hatch egg masses 24 (70.6%) were in good condition and 10 (29.4%) were in fair condition (≤95% fertilized, small size egg mass). The egg masses were estimated to be 1-7 days old upon detection and were estimated to be between 87-99% fertilized. Of the 24 deemed to be in good condition, fertilization was estimated to be between 97-100%, of those 10 deemed to be in fair condition, fertilization was estimated to be between 87-99%.

Of the 5 egg masses detected in the process of hatching, 4 (80.0%) were in good condition (appeared to have a good hatch-out, average size egg mass, well attached to vegetation and not slumped to bottom of substrate) and 1 (20.0%) was in fair condition because there appeared to be low hatching success. See Table 10 for details. None of the egg masses detected in 2014 were deemed to be in poor condition.

Egg mass volume data is no longer being reported since it is not possible to gather any meaningful conclusions by comparing volumes of egg masses detected at different developmental stages since the size and volume of the egg mass changes over time during development.

Table 10. LIPI egg masses detected in the CVWMA during the spring of 2014

| | 86 | | | <u> </u> |
|---------------|--------------|--------------|-----------|--------------------------------|
| Egg Mass #* | ~Date Laid** | Site | Condition | Comments |
| EM140425-CS01 | 24-Apr | EDLNA | Good | 99% fertilized |
| EM140429-CS02 | 27-Apr | EDLNA | Good | 99% fertilized |
| EM140430-EA03 | 29-Apr | WDLNA | Fair | 99% fertilized; small |
| EM140430-BH04 | 28-Apr | WDLNA | Good | 99% fertilized |
| EM140430-BH05 | 26-Apr | WDLNA | Fair | 95% fertilized |
| EM140501-CS06 | 29-Apr | WDLNA | Good | 99% fertilized |
| EM140501-EA07 | 29-Apr | WDLNA | Fair | 99% fertilized; small; slumped |
| EM140501-EA08 | 29-Apr | WDLNA | Fair | 99% fertilized; slumped |
| EM140501-CS09 | 29-Apr | WDLNA | Good | 99% fertilized |
| EM140501-CS10 | 29-Apr | WDLNA | Good | 99% fertilized |
| EM140505-BH11 | 04-May | WDLNA | Good | 99% fertilized |
| EM140506-CS12 | 03-May | WDLNA | Fair | 95% fertilized |
| EM140506-EA13 | 02-May | WDLNA | Fair | 98% fertilized; slumped |
| EM140506-EA14 | 29-Apr | EDLNA | Fair | 95% fertilized |
| EM140506-EA15 | 27-Apr | EDLNA | Good | hatching out |
| EM140507-BH16 | 05-May | WDLNA | Good | 99% fertilized |
| EM140507-BH17 | 05-May | WDLNA | Good | 99% fertilized |
| EM140507-TH18 | 02-May | WDLNA | Good | 97% fertilized; double lobe |
| EM140507-TH19 | 05-May | WDLNA | Good | 98% fertilized |
| EM140507-EA20 | 05-May | WDLNA | Good | 99% fertilized |
| EM140508-TH21 | 01-May | EDLNA | Good | 98% fertilized |
| EM140508-EA22 | 06-May | WDLNA | Good | 98% fertilized |
| EM140508-BH23 | 05-May | EDLNA | Good | 99% fertilized |
| EM140508-CS24 | 06-May | WDLNA | Good | 99% fertilized |
| EM140508-EA25 | 07-May | WDLNA | Good | 100% fertilized |
| EM140508-CS26 | 06-May | WDLNA | Good | 99% fertilized |
| EM140513-CS27 | 12-May | WDLNA | Good | 99% fertilized |
| EM140513-CS28 | 04-May | WDLNA | Fair | hatching out; low numbers |
| EM140513-TH29 | 11-May | WDLNA | Fair | 98% fertilized; slumped |
| EM140513-TH30 | 11-May | WDLNA | Good | 99% fertilized |
| EM140514-EA31 | 08-May | WDLNA | Good | hatched out |
| EM140514-TH32 | 04-May | WDLNA | Good | hatched out |
| EM140514-BH33 | 12-May | WDLNA | Good | 97% fertilized |
| EM140514-BH34 | 12-May | WDLNA | Good | 99% fertilized |
| EM140515-EA35 | 10-May | WDLNA | Fair | 90% fertilized |
| EM140515-TH36 | 07-May | WDLNA | Good | hatched out |
| EM140521-BH37 | 19-May | WDLNA | Good | 99% fertilized |
| EM140521-BH38 | 14-May | WDLNA | Good | 99% fertilized |
| EM140522-CS39 | 20-May | WDLNA | Fair | 87% fertilized (not well) |

^{*}Number format is based on date of detection (YYMMDD)

^{**}Date laid is approximated based on developmental stage of egg mass when detected

While releasing tadpoles from egg mass cages at the free-swimming stage, the number of hatchlings were estimated (not counted precisely) for 10 of the egg masses. The mean number of hatchlings was 2355 (n=10, SD=425.5, range 1442 – 2986), see Table 11 for details.

Table 11. Hatchling Counts upon release from egg mass cages

| Egg mass # | Site | # of hatchlings |
|---------------|-------|-----------------|
| EM140430-EA03 | WDLNA | 2986 |
| EM140430-BH04 | WDLNA | 2406 |
| EM140501-CS06 | WDLNA | 2464 |
| EM140501-EA07 | WDLNA | 2027 |
| EM140501-CS09 | WDLNA | 2438 |
| EM140501-CS10 | WDLNA | 2500 |
| EM140513-CS27 | WDLNA | 2318 |
| EM140513-TH30 | WDLNA | 1442 |
| EM140514-BH33 | WDLNA | 2811 |
| EM140514-BH34 | WDLNA | 2159 |

This year was the fourth consecutive year of above average egg mass detections and is the largest number of egg masses detected at the CVWMA since the project began (Table 12). The mean was 8.2 (n=11, SD=3.8, range 4-16) from 2000-2010. An estimate of the minimum size of the breeding population can be calculated by doubling the number of egg masses detected to account for the breeding pair, giving us 78 for 2014; this is likely a conservative estimate since the male to female ratio is not thought to be 1:1.

Table 12. LIPI egg masses detected in the CVWMA 2000-2014

| Year | EDLNA pond | | WDLNA | Leach #4 | Total |
|-------|------------|----|-------|----------|-------|
| 2000 | 8 | 0 | 8 | * | 16 |
| 2001 | 12 | * | 0 | * | 12 |
| 2002 | 1 | 2 | 2 | * | 5 |
| 2003 | 4 | 0 | 2 | * | 6 |
| 2004 | 3 | 0 | 1 | * | 4 |
| 2005 | 0 | 0 | 4 | 3 | 7 |
| 2006 | 3 | 2 | 2 | 0 | 7 |
| 2007 | 3 | 1 | 4 | 5 | 13 |
| 2008 | 3 | 0 | 1 | 2 | 6 |
| 2009 | 4 | 0 | 3 | 0 | 7 |
| 2010 | 5 | ** | 2 | ** | 7 |
| 2011 | 2 | 0 | 15 | ** | 17 |
| 2012 | 3 | 0 | 19 | ** | 22 |
| 2013 | 2 | 0 | 19 | ** | 21 |
| 2014 | 6 | 0 | 33 | *** | 39 |
| Total | 59 | 5 | 115 | 10 | 189 |

^{*} indicates area not surveyed

^{**}indicates no EMS in area because no calling detected during NCS

^{***}YOY observed in summer so possible undetected breeding occurred

In 2014, the egg mass detection rate was 0.32 egg masses per person-hour of survey time. The mean egg mass detection rate between the years of 2009-2013 was 0.15 egg masses per person-hour of survey time; egg mass detection rate during these years ranged from a minimum of 0.11 in 2009, to a high of 0.21 in 2012 (Table 13). It should be noted that this is just a general comparison, for a precise comparison it would be necessary to compare values between years by specific site within the CVWMA as survey effort by site varies from year to year.

Table 13. Summary of egg mass detection rate in the CVWMA (all sites); 2009-2014

| | 2009 | 2010 | 2011 | 2012 | 2013 | 2014 |
|------------------------------|-------|-------|-------|--------|--------|--------|
| Number of surveys | 30 | 17 | 23 | 29 | 26 | 16 |
| Survey effort (person-hours) | 62:20 | 58:26 | 97:22 | 104:17 | 167:42 | 120:52 |
| # of egg masses detected | 7 | 7 | 17 | 22 | 21 | 39 |
| Egg mass catch/effort | 0.11 | 0.12 | 0.17 | 0.21 | 0.13 | 0.32 |

Figure 6 shows the range in the egg mass detection rate during the spring breeding season in the CVWMA from 2009 to 2014. The highest detection rate during this period was in 2014, at 0.32 egg masses per person-hour of survey time, which is more than twice the mean detection rate of 0.15 egg masses per person-hour of survey time (n=5, SD=0.04, range 0.11-0.21) for the previous 5 years (2009-2013).

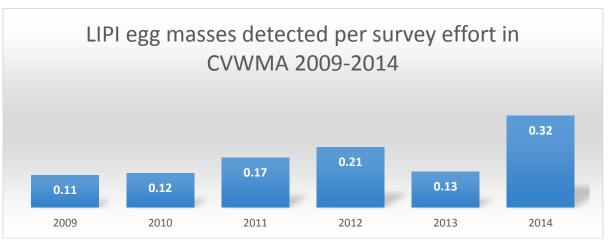


Figure 6. LIPI egg masses detected per survey effort in the CVWMA (all sites combined) from 2009-2014.

Air temperatures recorded at DLNA by HOBO temperature data logger #5 show that the air temperature for the surveyed spring breeding season (April 24-June 1, 2014) ranged from a low of -1.1°C on April 29 at 06:00 to a high of 29.1°C on June 1, mean for the period was 11.8°C (SD=6.4, n=946). Water temperatures recorded at DLNA by HOBO temperature data logger #6 show that the water temperature for the same period ranged from a low of 6.6°C on April 27 at 07:00 to a high of 23.6°C on May 14 at 18:00, mean for the period was 15.4°C (SD=3.7, n=946); Figure 7.

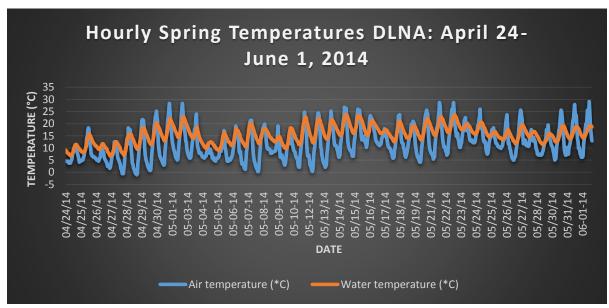


Figure 7. Daily spring temperatures at DLNA for the spring breeding season.

As shown in Figure 8, there were multiple peaks and troughs in the nightly air temperature profile spanning several degrees for the spring breeding season (data source: HOBO temperature data logger #5 at DLNA breeding site, temperatures at 22:00 nightly). This temperature profile which is characterized by a series of peaks and troughs appears to trigger egg mass deposition as temperatures begin to warm after a night or more of colder temperatures at dusk.

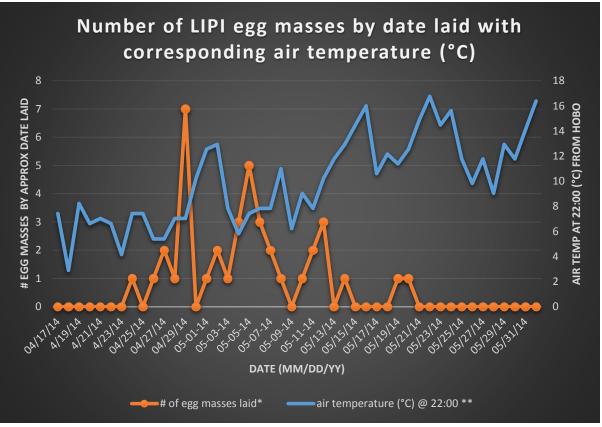


Figure 8. LIPI egg masses by date laid with corresponding air temperature (°C)

- * indicates # of egg masses detected (by date laid, not date of detection)
- ** source for air temperature is HOBO data logger #5 at DLNA temperature at 22:00 Note: EMS began on April 24 but temperature is shown for 1 week period prior to show trend; there was no evidence of any egg masses being laid earlier than those first detected

3.5 Visual Encounter Surveys (VES)

In 2014 at the CVWMA 152:51 person-hours were dedicated to visual encounter surveys (n=66 surveys) throughout the year, Table 14 provides a summary of VES and catch per effort by season. Locations of LIPI observations in the CVWMA are provided in maps in Appendices 2, 4, 5 and 7.

Table 14. Summary of VES efforts in the CVWMA for 2014

| | Spring | Summer | Fall | Winter | Total |
|------------------------------|--------|--------|-------|--------|--------|
| Number of surveys | 1 | 45 | 19 | 1 | 66 |
| Survey effort (person-hours) | 01:09 | 111:01 | 39:06 | 01:35 | 152:51 |
| Number of LIPI observations | 0 | 225 | 92 | 1 | 318 |
| LIPI catch/effort during NCS | 0 | 2.0 | 2.4 | 0.6 | 2.1 |

Only 1 spring VES was conducted, on 24 March with a survey effort of 1:09. This survey was done in the goat channel over-wintering area to determine if there were any signs of LIPI at this time of year; nothing was detected.

During summer 111:01 person-hours were spent on VES (n=45 surveys) from July 15 to September 17. Summer surveys were done in known breeding ponds (*) to search for YOY and in the migration corridors and overwintering areas to search for all age classes. Surveyed areas include: WDLNA*, EDLNA ponds*, East ditch, goat channel, Kootenay River east channel, Kootenay River main channel and inlets at Leach Lake, south shoreline of Duck Lake, Pumphouse channel, south-west ditch and Leach lake (compartments 1, 3, 4 and 5); attempts were made to search the frog-bear channel but it has become too overgrown with weeds to survey. During these summer VES, a total of 225 LIPI observations were made, a catch per effort of 2.0 LIPI per person-hour of surveying. Of the total 225 summer VES observations 1 was an adult (female), 222 were YOY (3 male, 219 unknown sex) and 2 were tadpoles, see Table 15 for details.

During fall VES a total of 39:06 person hours (n=19 surveys) were spent surveying for LIPI in the migration corridors and over-wintering areas between Sept 25 and October 20. Areas surveyed include: East ditch, squiggle channel, Leach Lake compartments 5 and 6, goat channel, Kootenay River east channel, Kootenay River main channel and inlets at Leach Lake, Pumphouse channel, Duck Lake (south shoreline) and Six mile slough compartments 4 and 5; attempts were made to search the frog-bear channel but it has become too overgrown with weeds to survey. During fall VES, a total of 92 LIPI observations were made, a catch per effort of 2.4 LIPI per person-hour of survey effort. Of the total 92 fall VES observations, 4 were adults (all female) and 88 were YOY (10M/78U), see Table 15 for details.

During winter, one VES was conducted by canoe along the goat channel over-wintering area to determine if over-wintering LIPI could be detected. Survey effort was 01:35 person-hours, and 1 adult was detected resting on the substrate, a catch per effort of 0.6.

Table 15. LIPI observations by stage, sex and season during VES at CVWMA in 2014

| | Adult | YOY | Tadpole | Total |
|--------|---------|------------|---------------|---------------|
| Summer | 1 | 222 | 2 | 225 |
| | (1F) | (3M/219U) | (2U) | (1F/3M/221U) |
| Fall | 4 | 88 | 0 | 92 |
| | (4F) | (10M/78U) | n/a | (4F/10M/78U) |
| Winter | 1 | 0 | 0 | 1 |
| | (1U) | n/a | n/a | (1U) |
| Total | 6 | 310 | 2 | 318 |
| | (5F/1U) | (13M/297U) | (2U) | (5F/13M/300U) |

Note: sex is noted in brackets: F=female, M=male, U=unknown

Table 16 summarizes the LIPI observations made during VES throughout the year by stage and detection type. Of the 318 total observations, 115 (36.2%) were captured (5 adult, 108 YOY and 2 tadpoles), 2 (0.6%) had photos taken only (2 YOY) and 201 (63.2%) were visuals with no photo (1 adult and 200 YOY).

Table 16. LIPI observations by stage and detection code during VES at CVWMA in 2014

| Detection code | Adult | YOY | Tadpole | Total |
|-----------------------|-------|-----|---------|-------|
| Capture | 5 | 108 | 2 | 115 |
| Photo only | 0 | 2 | 0 | 2 |
| Visual (no photo) | 1 | 200 | 0 | 201 |
| Total | 6 | 310 | 2 | 318 |

At Bummer's Flats only one VES was conducted (additional surveys were planned but due to an overlap in scheduling with another survey crew from the BC-NLFRT our surveys were cancelled). In total 2:33 person-hours of survey effort were put in on July 17, 2014 at the release ditch of North Bummer's Flats where we released 7253 hatchling tadpoles between May 15 to 31, 2014; the objective of this survey was to determine the stage of development and to get a tally of YOY metamorphs out of the water on that date; size data was also collected on a few individuals. There were 24 YOY detected, 5 were checked for tail stubs and it was found that only 2 of the 5 had tail stubs present; length of the tail stubs ranged from 5.0 – 25.0 mm. Size data was collected on 3 individuals, mean weight was 5.3 grams (n=3, SD=0.6, range 5.0 - 6.0), mean SVL was 37.4 mm (n=3, SD=0.5, range 37.0 - 37.9). A map of observations is provided in Appendix 10.

3.6 Tadpole Trapping

In the CVWMA 13:21 person-hours were spent tadpole trapping (n=8 surveys), Table 17. Trapping was conducted at the EDLNA pond, east ditch and Leach Lake pond 4 between July 2 and 8. LIPI tadpoles were only detected in the EDLNA pond (n=15 tadpoles, Gosner stages 34-41 on July 2 and 3). See map in Appendix 1 for trap locations and map in Appendix 4 for location of tadpole observations.

Table 17. Summary of 2014 tadpole trapping effort

| | Tadpole Trapping |
|---|------------------|
| Number of surveys | 8 |
| Survey effort (person-hours) | 13:21 |
| Number of LIPI observations | 15 |
| LIPI catch/effort during tadpole trapping | 1.1 |

Tadpole trapping was conducted at Bummers Flats in the release ditch at North Bummers and at the pond connected to it via culvert located between the railway tracks and highway (referred to as the railway pond where an egg mass was detected in spring 2014 by Penny Ohanjanian; pers comm P. Ohanjanian); a total of 6:47 person-hours were spent trapping on July 9, 10 and 11. On July 10, 3 LIPI tadpoles were detected in the railway pond (Gosner stages 27-28). On July 11, 7 LIPI tadpoles were detected in the release ditch (Gosner stages 37-45). All tadpoles appeared to be in good health. See map in Appendix 9 for trap locations and map in Appendix 10 for location of tadpole observations.

3.7 Late-summer and Fall Migration Road Surveys

Night time road surveys during the late-summer and fall migration period were conducted along the Duck Lake dike between September 9 and October 9 (n=12 night surveys and 1 day survey to look for carcasses). A total of 36:27 person-hours of survey effort was dedicated to these surveys, 219 LIPI observations were made, a catch per effort of 6.1 LIPI/person-hour of survey effort (Table 18). See maps in Appendices 2, 4 and 5 for locations of LIPI observations made during road surveys.

Table 18. Summary of 2014 late-summer & fall road survey effort

| | Road surveys |
|---------------------------------------|--------------|
| Number of surveys | 13* |
| Survey effort (person-hours) | 36:27 |
| Number of LIPI observations | 219 |
| LIPI catch/effort during road surveys | 6.1 |

^{*12} night surveys and 1 day survey

Of the 219 LIPI observations made during night time road surveys 7 were adults (3 female, 1 male and 3 unknown sex) and 212 were YOY (6 male and 206 unknown sex). There were 199 live LIPI detected on the dike (7 adults, 192 YOY) and 20 dead (all YOY), which translates to 90.9% live and 9.1% dead (Table 19).

The majority of the LIPI observations made during the road surveys, 140 of 219 (63.9%) were made during 2 night surveys (September 17 and September 22). On the night of September 17 there were a total of 68 LIPI detections on the road (all YOY), of these 55 were live (80.9%) and 13 (19.1%) were freshly killed road mortalities, all attributed to only 2 car passes; there were also 21 LIPI YOY detected staging along the south shoreline of Duck Lake during this night survey. On the night of September 22 there were a total of 72 LIPI detections on the road, 67 live (4 adults, 63 YOY) and 5 dead (6.9%; all YOY).

Table 19. LIPI detections on night road surveys at Duck Lake dike (n=12 surveys)

| Stage | Total | Live | Dead | Proportion Live (Dead) |
|-------|------------------|------------------|----------|-------------------------------|
| Adult | 7 (3F/1M/3U) | 7 (3F/1M/3U) | 0 | 100.0% (0.0%) |
| YOY | 212 (6M/206U) | 192 (6M/186U) | 20 (20U) | 90.6% (9.4%) |
| Total | 219 (3F/7M/209U) | 199 (3F/7M/189U) | 20 (20U) | 90.9% (9.1%) |

Mean air temperature at dusk for the subset of night surveys where LIPI were encountered was 15.8° C (n=6, SD=3.4, range 10.3° C -20.9° C). LIPI were encountered in 6 of the 12 night road surveys (50%). Movement onto the road surface seemed to begin at dusk, and as darkness set in the number of encounters increased. Some of the frogs moved across the road quite quickly, but the majority seemed to be resting on the road, likely for the main purpose of thermoregulating and possibly to opportunistically forage as well. In some cases the frogs did not even flee when approached.

Other herptile observations during late summer and fall night time road surveys along Duck Lake dike included: 3 live Western toads (*Anaxyrus boreas*), 2 dead Common garter snakes

(Thamnophis sirtalis) and 3 Western terrestrial garter snakes (Thamnophis elegans), 1 live and 2 dead.

Traffic data from the area is collected 24-hours per day during the migration period via Trafx traffic counter and is available from the CVWMA.

3.8 Animal Health

Of the captured LIPI whose health was assessed visually in the field (n=232), 80.6% were deemed to be in good health (n=187), 9.1% in fair health (n=21), 0 % in poor health and 10.3% were found dead (n=24), see Table 20 for details by season and age class.

Table 20. Health assessment of LIPI 2014 field observations in the CVWMA (n=232)

| Season | Age Class | Good | Fair | Poor | Dead | Total |
|--------------------|-----------------|-------|------|------|-------|--------|
| Spring | adult | 26 | 7 | 0 | 1 | 34 |
| Spring | juvenile | 30 | 8 | 0 | 0 | 38 |
| Spring | all age classes | 56 | 15 | 0 | 1 | 72 |
| Summer | adult | 3 | 0 | 0 | 0 | 3 |
| Summer | YOY | 66 | 2 | 0 | 15 | 83 |
| Summer | all age classes | 69 | 2 | 0 | 15 | 86 |
| Fall | adult | 6 | 1 | 0 | 0 | 7 |
| Fall | YOY | 56 | 3 | 0 | 8 | 67 |
| Fall | all age classes | 62 | 4 | 0 | 8 | 74 |
| Grand Total | all age classes | 187 | 21 | 0 | 24 | 232 |
| Percentage | all age classes | 80.6% | 9.1% | 0.0% | 10.3% | 100.0% |

Of the 187 LIPI deemed to be in good health, there were 35 adults, 30 juvenile and 122 YOY. Of the 21 live LIPI deemed to be in fair condition, there were 8 adults, 8 juvenile and 5 YOY. The reason they were deemed to be in fair condition included the following: chytridiomycosis suspected (n=6; 2 adults, 3 juvenile, 1 YOY), chiggers (n=6; 3 adults, 3 juveniles), chiggers and chytridiomycosis suspected (n=2 adults), necrotic digits (n=1 YOY), red-orange stained ventral surface (n=1 YOY), missing distal portion of limb (n=1 YOY) and wound (n=4; 1 adult, 2 juvenile, 1 YOY). There were no LIPI detected in poor condition during the 2014 field season.

Of the 24 dead LIPI detected, there was 1 adult male and 23 YOY. The dead adult was a male detected in the WDLNA breeding area on April 30, 2014, the body was in the advanced stages of decomposition so it was not possible to conclusively determine the cause of death as it could not be submitted to the lab in such advanced stage of decomposition. Of the 23 YOY, 1 was killed and being consumed by a giant water bug (Family Belostomatidae), 1 was a road mortality on the Leach Lake dike and the remainder (n=21) were road mortalities at Duck Lake on the dike in late summer and fall. The photos in Figure 9 document some of the health issues detected in the LIPI population in Creston during the 2014 field season as noted above.

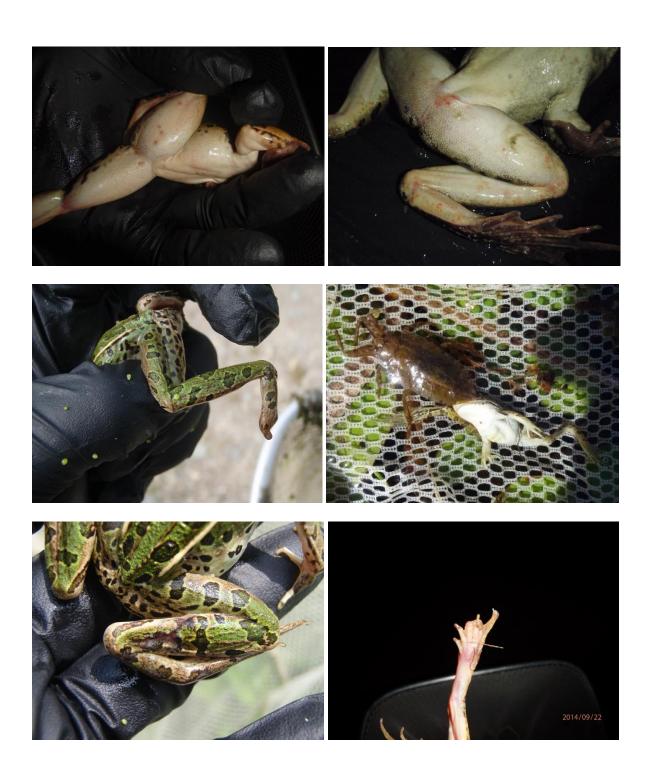






Figure 9. Health issue photos (from top row, left to right): chiggers infection, chiggers infection, YOY missing distal portion of left hind limb, YOY being eaten by giant water bug (Belastomatidae), wound (right hind leg), necrotic tissue on the feet, road mortality (freshly killed) and YOY with stained skin.

Of the 232 LIPI observations which were visually inspected by surveyors, chytridiomycosis was suspected in 8 (3.4%) of the frogs; it was suspected in 1.7% of adults, 1.3% of juveniles and 0.4% of the YOY observations. It was not possible to conclusively determine whether or not the 1 dead male adult (0.4% of total) found in the WDLNA breeding area in the spring had chytridiomycosis because the body was too decomposed to visually inspect or send to the lab for analysis; however there is a high probability that it did. Chytridiomycosis was not suspected in 223 of the 232 observations visually inspected for signs of the disease (96.1%). Table 21 provides a detailed summary of these results.

Table 21. Field observer's determination if chytridiomycosis suspected by visual inspection of LIPI captured at CVWMA (n=232); includes dead specimens (n=24)

| Chytridiomycosis Suspected | | | | | |
|----------------------------|-------------|----------|----------|--------------------|--|
| Stage | No | Yes | Unknown | Grand Total | |
| Adult | 39 (16.8%) | 4 (1.7%) | 1 (0.4%) | 44 (19.0%) | |
| Juvenile | 35 (15.1%) | 3 (1.3%) | 0 | 38 (16.4%) | |
| YOY | 149 (64.2%) | 1 (0.4%) | 0 | 150 (64.6%) | |
| Grand Total | 223 | 8 | 1 | 232 | |
| Percentage | 96.1% | 3.4% | 0.4% | 100.0% | |

Lab results of PCR assay for *Batrachochytrium dendrobatidis* (*Bd*) on LIPI tested in the CVWMA in 2014 are outlined in Table 22. Results indicate that 78 of the 144 (54.2%) samples were positive. Of the 39 adults tested 36 were positive (92.3%), of the 38 juveniles tested, 36 were positive (94.7%) and of the 67 YOY tested, 6 were positive (9.0%). PCR assay Ct values ranged from 21.08 - 39.24 in the samples that tested positive.

Table 22. Results of 2014 PCR assay for Bd on LIPI in the CVWMA

| Age class | Total # sampled | Total # positive | Total % positive |
|-----------|-----------------|------------------|------------------|
| Adult | 39 | 36 | 92.3% |
| Juvenile | 38 | 36 | 94.7% |
| YOY | 67 | 6 | 9.0% |
| Total | 144 | 78 | 54.2% |

Of the LIPI tested for *Bd* during the spring (n=71), 95.8% tested positive (n=68), including 97.0% of adults (n=32) and 94.7% (n=36) of juveniles. Of the LIPI tested for *Bd* during the summer (n=20), 25.0% tested positive (n=5), including 50.0% of adults (n=1) and 22.2% (n=4) of YOY. Of the LIPI tested for *Bd* during the fall (n=53), 9.4% tested positive (n=5), including 75.0% of adults (n=3) and 4.1% of YOY (n=2). Table 23 provides a summary of the lab results by age class and season.

Table 23. Bd results for captured LIPI in CVWMA by age class and season (n=144)

| Stage | Spring | | Sun | ımer | Fall | | |
|----------|------------|-----------|-----------|-----------|-----------|-----------|--|
| | Positive | # Samples | Positive | # Samples | Positive | # Samples | |
| Adult | 32 (97.0%) | 33 | 1 (50.0%) | 2 | 3 (75.0%) | 4 | |
| Juvenile | 36 (94.7%) | 38 | * | * | * | * | |
| YOY | * | * | 4 (22.2%) | 18 | 2 (4.1%) | 49 | |
| Total | 68 (95.8%) | 71 | 5 (25.0%) | 20 | 5 (9.4%) | 53 | |

^{*} no samples

Table 24 provides a comparison of lab results and field observers health classification of whether or not Bd was suspected. The results show that of the 7 live LIPI where Bd was suspected, 100% tested positive in the lab (4 adults and 3 juveniles); however, of the 137 live LIPI where Bd was not suspected, surprisingly 51.8% tested positive (n=71, including 32 adults, 33 juveniles and 6 YOY) and only 48.2% tested negative (n=66, including 3 adults, 2 juveniles and 61 YOY).

Table 24. 2014 Results of field observer's health classification compared to results of *Bd* lab testing for live LIPI captured in the CVWMA (n=144 total samples tested)

| | Chytrid suspec | ted: No (n=137) | Chytrid susp | Grand | |
|--------------------|--------------------------|--------------------------|--------------------------|--------------------------|---------|
| Stage | Bd lab test: Negative | Bd lab test: Positive | Bd lab test: Negative | Bd lab test: Positive | Total |
| Adult | 3 | 32 | 0 | 4 | 39 |
| Juvenile | 2 | 33 | 0 | 3 | 38 |
| YOY | 61 | 6 | 0 | 0 | 67 |
| Grand Total | 66 | 71 | 0 | 7 | 144 |
| % of category* | 48.18% | 51.82% | 0.00% | 100.00% | n/a |
| % of Total | 45.83% | 49.31% | 0.00% | 4.86% | 100.00% |

^{*2} categories chytridiomycosis suspected: yes and chytridiomycosis suspected: no

3.9 Dorsal Spot Pattern Recognition and Recapture Information

While dorsal spot pattern photos were taken for all captures during the 2014 field season and a library was created, it was decided that the recognition would not be completed this year due to the large amount of analysis time required and the low number of recaptures.

3.10 Morphometrics of Creston LIPI

Size data was gathered on 205 LIPI observations (includes all field captures where both a weight and snout vent length were recorded; includes recaptures) during the 2014 field season. Throughout the course of the entire field season, there were measurements taken on 125 YOY (67 in summer, 58 in fall), 38 juveniles 38 in spring) and 42 adult captures (33 in spring, 3 in summer, 6 in fall), see Table 25 and 26 for details. Figures 10, 11 and 12 provide a visual representation of the morphometrics of Creston LIPI detailed in Table 25 and 26

Table 25. Weight measurements of 2014 LIPI observations at CVWMA

| Weight (g) | | n | mean | SD | range (min) | range (max) |
|---------------------------|-----------------|----|------|------|-------------|-------------|
| Spring (March 21-June 20) | juvenile | 38 | 42.2 | 7.5 | 21.0 | 49.0 |
| | adult | 33 | 57.0 | 8.8 | 50.0 | 95.0 |
| | all age classes | 71 | 49.0 | 11.0 | 21.0 | 95.0 |
| Summer (June 21-Sept 20) | YOY | 67 | 16.1 | 8.8 | 4.0 | 34.0 |
| | adult | 3 | 88.7 | 1.5 | 87.0 | 90.0 |
| | all age classes | 70 | 19.2 | 17.1 | 4.0 | 90.0 |
| Fall (Sept 21-Dec 20) | YOY | 58 | 20.9 | 9.0 | 6.0 | 39.0 |
| | adult | 6 | 84.0 | 15.6 | 63.0 | 103.0 |
| | all age classes | 64 | 26.9 | 20.9 | 6.0 | 103.0 |

^{*}recaptures included

Table 26. SVL measurements of 2014 LIPI observations at CVWMA

| SVL (mm) | | n | mean | SD | range (min) | range (max) |
|---------------------------|-----------------|----|------|------|-------------|-------------|
| Spring (March 21-June 20) | juvenile | 38 | 73.4 | 5.9 | 58.4 | 83.1 |
| | adult | 33 | 80.5 | 4.9 | 71.6 | 89.7 |
| | all age classes | 71 | 76.7 | 6.5 | 58.4 | 89.7 |
| Summer (June 21-Sept 20) | YOY | 67 | 52.2 | 9.4 | 31.1 | 72.0 |
| | adult | 3 | 88.4 | 3.2 | 85.4 | 91.7 |
| | all age classes | 70 | 53.8 | 11.8 | 31.1 | 91.7 |
| Fall (Sept 21-Dec 20) | YOY | 58 | 56.1 | 8.8 | 37.6 | 70.8 |
| | adult | 6 | 84.5 | 7.4 | 74.9 | 92.8 |
| | all age classes | 64 | 58.8 | 12.0 | 37.6 | 92.8 |

^{*}recaptures included

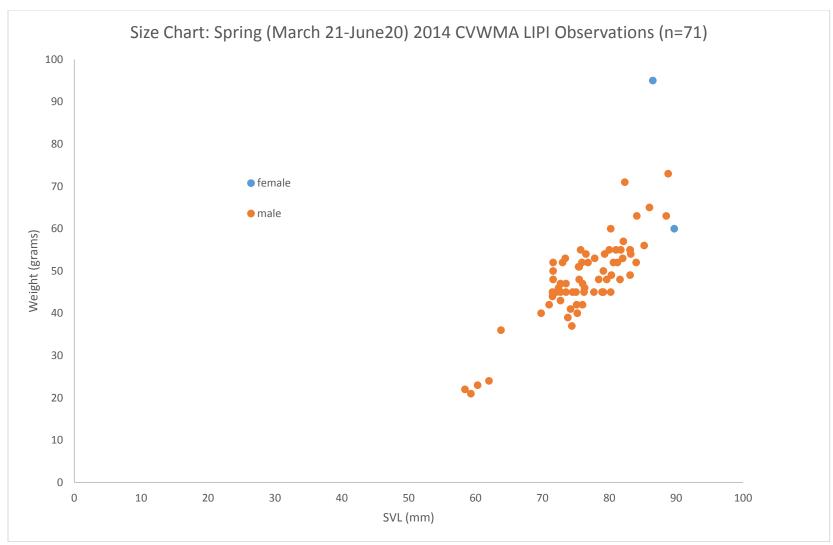


Figure 10. Size Chart: Spring (March 21-June 20) 2014 CVWMA LIPI Observations (n=71)

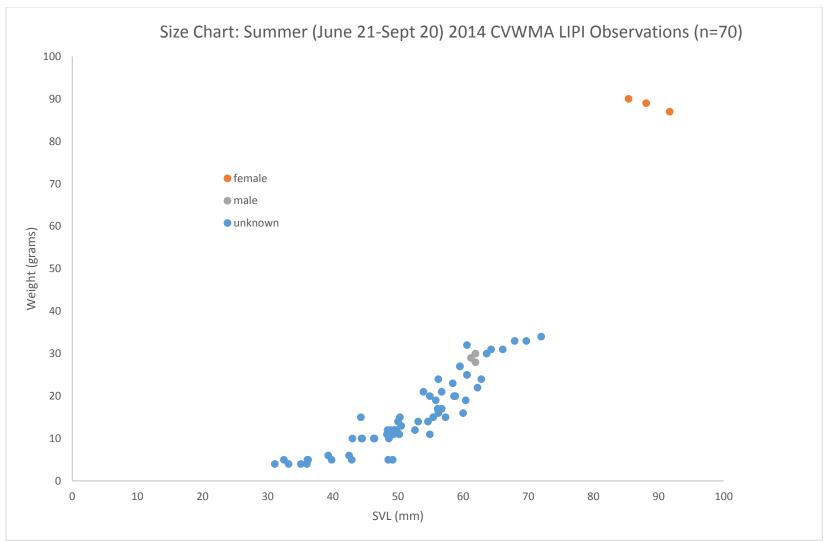


Figure 11. Size Chart: Summer (June 21-September 20) 2014 CVWMA LIPI Observations (n=70)

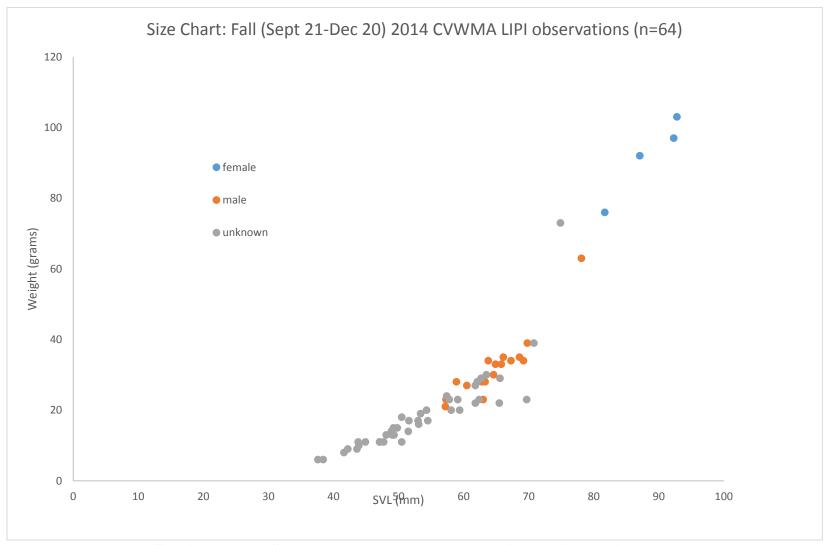


Figure 12. Size Chart: Fall (September 21-December 20) 2014 CVWMA LIPI Observations (n=64)

3.11 Vancouver Aquarium Captive Assurance Colony

In 2014, a total of 40 free-swimming tadpoles, from 4 different egg masses (10 tadpoles from each) were transferred to the Vancouver Aquarium May 13, 2014. In 2014 a new approach for transferring the tadpoles was used, for the first time they were packaged up in Creston and driven to the Trail Airport (YZZ) where they were flown to Vancouver via Pacific Coastal Airlines. The source of all the tadpoles was WDLNA, which were collected from egg masses EM140430-EA03, EM140430-BH04, EM140501-CS06, EM140501-EA07; these egg masses were estimated to have been laid April 28-29.

As of the last update received from the Vancouver Aquarium in the fall of 2014, when prebrumation morphometrics were taken, there were 20 survivors of the original 40 transferred as tadpoles in 2014; all metamorphosed into YOY. Table 27 shows the fall 2014 pre-brumation morphometrics of the Creston and the captive bred animals. Mean weight of the Creston YOY was found to be 8.56 g (n=20, SD=1.18, range 6.15-10.37), mean SVL was 3.70 mm (n=20, SD=0.16, range 3.38-3.70). The mean weight of the Vancouver Aquarium captive bred YOY was 6.88 g (n=27, SD=0.08, range 5.06-8.41), mean SVL was 3.56 mm (n=27, SD=0.19, range 3.07-3.83).

Table 27. Pre-brumation morphometrics of 2014 Vancouver Aguarium LIPI

| Morphometric | mean | n | SD | range (min) | range (max) | p-value |
|----------------------------|------|----|------|-------------|-------------|----------|
| Creston source weight (g) | 8.56 | 20 | 1.18 | 6.15 | 10.37 | 0.000005 |
| VA captive bred weight (g) | 6.88 | 27 | 0.08 | 5.06 | 8.41 | 0.000005 |
| Creston source SVL (mm) | 3.70 | 20 | 0.16 | 3.38 | 3.70 | 0.006 |
| VA captive bred SVL (mm) | 3.56 | 27 | 0.19 | 3.07 | 3.83 | 0.006 |

As shown in Table 27 and Figure 13 Creston source YOY at the Vancouver Aquarium are larger, mean weight of 8.56 grams compared to 6.88 grams for the captive bred animals, a difference of 1.68 grams. Mean SVL for the Creston source YOY was 3.70 mm compared to 3.56 mm for the captive bred animals, a difference of 0.14 mm (Figure 14). Unpaired t-test results show that both of these differences in size are statistically significant at the 95% level (p-value <0.05).



Figure 13. Comparison of mean weight in YOY animals at Vancouver Aquarium (Creston source animals versus captive bred)

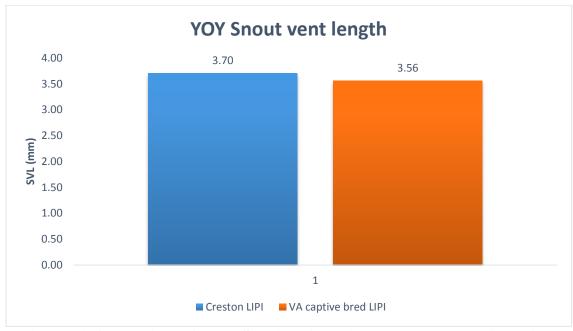


Figure 14. Comparison of mean SVL in YOY animals at Vancouver Aquarium (Creston source animals versus captive bred)

3.12 Bummers Flats Reintroductions

In 2014, year 4 of phase 2 reintroductions at Bummers Flats, a total of 7253 hatchlings were transferred from 8 different egg masses from the source population in Creston (Table 28). The source egg masses were all from WDLNA and were estimated to have been laid between April 29 and May 12, 2014. Transfers were done using hatchlings that had recently become free-swimming. Hatchlings were brought to the site, acclimatized using the protocol developed by the Alberta Northern Leopard Frog Recovery Team (Kendell and Prescott 2007) with some minor modifications and held in egg mass cages overnight. Hatchlings were counted the following day and released in various locations throughout the length of the release ditch. Appendix 9 shows the location of the release ditch at North Bummers Flats. Since 2011, when phase 2 reintroductions at Bummers Flats began a total of 28,679 LIPI hatchlings have been released (Table 29).

Table 28. Summary of LIPI Bummers Flats Reintroduction in 2014

| Transfer Date | Source Site | Egg mass # | ~Date laid | # Released |
|----------------------|-------------|---------------|------------|------------|
| May 15-16 | WDLNA | EM140430-EA03 | 29-Apr | 1486 |
| May 15-16 | WDLNA | EM140430-BH04 | 28-Apr | 1206 |
| May 15-16 | WDLNA | EM140501-CS06 | 29-Apr | 1264 |
| May 15-16 | WDLNA | EM140501-EA07 | 29-Apr | 1027 |
| May 30-31 | WDLNA | EM140513-CS27 | 12-May | 711 |
| May 30-31 | WDLNA | EM140513-TH30 | 11-May | 599 |
| May 30-31 | WDLNA | EM140514-BH33 | 12-May | 442 |
| May 30-31 | WDLNA | EM140514-BH34 | 12-May | 518 |
| Grand Total | | | | 7253 |

Table 29. Summary of phase 2 reintroductions at Bummers Flats 2011-2014

| Year | Source | Release Site | # LIPI released |
|-------|--------|-----------------------------|-----------------|
| 2011 | WDLNA | North Bummers release ditch | 4948 |
| 2012 | WDLNA | North Bummers release ditch | 7600 |
| 2013 | WDLNA | North Bummers release ditch | 8878 |
| 2014 | WDLNA | North Bummers release ditch | 7253 |
| Total | | | 28, 679 |

4. DISCUSSION

4.1 Songmeters

During 2013-2014 efforts were made by an audio specialist to develop a filter/recognizer to automate northern leopard frog detection during analysis using the Songscope software; a filter was developed but unfortunately it did not function as well as was hoped and is not being used due to the extremely large number of false positives in the results during trials. As a result, the only other options for analysis are: 1) listen to audio files manually in real time, or 2) use Songscope software to visually scan the files at high-speed searching for the characteristic digital signature of the northern leopard frog. The second method is the one that we have always used, and while it is much faster than method 1, it still requires a large amount of analysis time. It is estimated that for every hour of recording it takes a minimum of 10 minutes for an experienced analyzer to visually scan the file, and that is without stopping at all to take a closer look or listen to possible matches.

During the spring 2014 breeding season, 4 Songmeters were deployed for 17 days set to record daily for 2 hours each, resulting in 136 hours of recordings. At a conservative rate of 10 minutes of analysis per hour that is 22.7 hours of analysis time. At a more realistic rate of 20 minutes of analysis per hour of recording (more realistic given the large amount of background noise in the productive CVWMA wetlands and the fact that much of it is within the same frequency as the northern leopard frog call so masks the target), that is 45.4 hours of analysis time. Given these large amounts of analysis time coupled with set up time, field deployment time, field retrieval time and equipment costs it might be worth re-evaluating their use as the resources might be better allocated to in-person field surveys, if surveyors are available, especially if the sites are relatively easy to access.

In addition to the large amount of analysis time, detection distance is not known and likely varies depending on site-specific characteristics such as vegetation and topography as well as environmental conditions. Surprisingly, 3 calling males that were detected in-person during a NCS on May 22 at Leach Lake pond 4 were not detected on Songmeter unit A that was deployed 386 m away or on Songmeter B, deployed 460 m away. This indicates that in this setting, detection distance was less than 386 m, which is surprising since calling can be detected by ear at distances greater than this under ideal conditions. Since environmental conditions were ideal on May 22 (no wind or rain) and most of the habitat between the calling males and the Songmeter was open water (as the drawdown completed recently removed the dense stands of cattails that were previously there) these are no plausible explanations for the lower than expected detection distance of the Songmeter.

4.2 Survey Effort

With the observed increases in the spring breeding population detected over the last few years it is has become increasingly more difficult to continue with the status quo spring survey program. With the increased number of calling males and the large geographic area over which they are calling at DLNA it is no longer possible to record all calling activity or search all areas of calling activity for egg masses. In the future changes will have to be made to the spring survey program unless extra resources are made available.

4.3 Nocturnal Calling Surveys (NCS)

Since no calling has been detected at the East Ditch of DLNA since 2012 it is recommended that this site be removed from the NCS field program to free up time to focus on other tasks.

The new calling areas detected during the spring of 2014 in the south-western and central regions of DLNA should be followed up on during the 2015 field season and it is recommended that if possible, surveys at Leach Lake be expanded to determine how extensive the breeding population is at the site.

4.4 Egg Mass Surveys (EMS)

With the increased area of calling activity in the WDLNA over the last few years it is not possible to thoroughly survey all areas where calling has been documented; to do so would require additional resources. Use of a stand up paddle board could be one way to cover larger amounts of area, and may have the added benefit of increasing the range of visibility from a higher perspective above the water in comparison to standing on the substrate, however dense aquatic vegetation may make navigation difficult in some areas.

The spring 2014 purchase of a new light weight canoe designed for use in shallow water was very beneficial; it worked well and saved time travelling between egg masses for caging, counting and releasing as well as being very beneficial in streamlining the time required for travel during the collection of tadpoles for both the Bummers Flats reintroduction and the Vancouver Aquarium captive assurance colony.

Since it takes a considerable amount of effort to cage and care for egg masses, it would be beneficial to determine whether or not caging has a significant impact on survival if possible.

4.5 Tadpole trapping

Since tadpole trapping is not a conclusive method to determine whether or not breeding occurred in an area, it is recommended that it primarily be used as a method to determine what developmental stage tadpoles are at in the known breeding areas to inform timing of metamorphosis which is important for YOY surveys. Tadpole trapping can be used as one of many tools that may help to determine if breeding occurred at a suspected site but should not be relied upon exclusively; it should be completed along with survey methods such as NCS, EMS and VES to really identify whether successful breeding occurred.

4.6 Late-summer and fall migration road surveys

In 2014 there were not a large number of road surveys done as it was felt that sufficient data has been collected since 2011 to document there is road mortality occurring. It should be noted that in the spring-summer of 2014, the CVWMA resurfaced the Duck Lake dike which has resulted in some vehicles travelling at much higher speeds now that they no longer have to navigate a very rough road with large potholes; as a result this may have impacts on the number of road mortalities so it recommended that now more than ever a mitigation plan be decided upon and implemented. It is recommended that the BC-NLFRT work with land managers to implement a mitigation plan.

4.7 Animal health

Aside from the road mortalities, there were only 2 dead LIPI detected during the 2014 field season, a YOY that was killed and being eaten by a giant water bug (Family Belostomatidae) and an adult male at WDLNA on April 30. The adult was already in the advanced stages of decomposition so could not be sent to the lab for a full work up to determine the cause of death but since there were no obvious signs of trauma it is suspected that it died from chytridiomycosis.

There were not a large number of animals detected showing overt symptoms of chytridiomycosis during the 2014 field season (8 of 232 observations); however lab results show that a large number of animals without symptoms tested positive for Bd (71 of 137 observations where chytridiomycosis was not suspected or 51.8%). This provides further evidence that it could be possible that the population in Creston has developed a resistance to chytridiomycosis as suggested in the paper entitled Prevalence of the pathogenic chytrid fungus, *Batrachochytrium dendrobatidis*, in an endangered population of northern leopard frogs, *Rana pipiens* (Voordouw et al, 2010). As expected, the highest incidence of chytridiomycosis was in adults in the spring (97.0%), followed by juveniles in spring (94.7%) and adults in fall (75.0%). Surprisingly 22.2% of summer YOY tested positive, which is much higher than the 3.1% reported by Voordouw (Voordouw et al, 2010). It is recommended that the BC-NLFRT review the multi-year summary of Bd sampling results prepared by the author and provide recommendations for the future sampling program.

There were a few cases of what is believed to be frog chiggers infection (genus Hannemania) detected during the spring breeding season of 2014 (n=8; 5 adult and 3 juvenile observations – not individuals); without lethally collecting specimens for analysis it is impossible to make an accurate diagnosis but it is believed to be the cause. Detections were made at both WDLNA (n=3 of the observations) and EDLNA ponds (n=5 of the observations). Of the 8 observations, 3 were large juveniles > 40 grams and 5 were adults. Most of the red nodules were observed on the ventral surface of the animals, with the highest concentration on the hind legs and feet. By reviewing dorsal spot pattern photos it was determined that of the 8 observations, 7 were unique individuals infected (5 adults and 2 juveniles; one juvenile was a recapture). This type of infection was first detected in the population in the spring of 2011, when Dr. David Earl Green, DVM, with the USGS National Wildlife Health Centre was contacted for advice in determining the cause, he suggested it was most likely a chiggers infection of the genus Hannemania, an ectoparasite (pers. comm. May 23, 2011).

There was 1 case of necrotic tissue during the 2014 field season, but it was on September 22, not during the spring breeding season which is when it was detected previously. The animal was a YOY detected at night on the Duck Lake dike during the fall migration period, it was missing the distal portion of tissue on 3 of the 5 digits of its left hind leg (for a photo see Figure 9 in Section 3.8). It was suggested by amphibian health specialists in the past that the necrotic tissue could be due to frostbite. Initially it was thought that this detection would disprove the frostbite theory since it was not expected that below freezing temperatures would have been encountered by a YOY between time of metamorphosis and September 22; however upon review of the temperature data collected by HOBO temperature data loggers it was discovered that there were a few days in September which were unseasonably cold with air temperatures

reaching a minimum of -0.61 °C, which could potentially provide evidence to support the frostbite theory since LIPI are very terrestrial at this time of year.

It is not clear if the YOY that was missing the distal portion of its foot was the result of a predatory attack in which it lost a portion of its limb or if it was a malformation known as ectromelia (absence of part of a limb) which could potentially have been caused by a trematode parasite such as Ribeiroia ondatrae which has been associated with malformed limbs (Johnson et al, 2002) or another cause.

4.8 Doral Spot pattern recognition and Recapture information

While dorsal spot pattern photos were taken for all captures during the 2014 field season and a library was created, it was decided that the recognition would not be completed this year due to the significant amount of effort required and given that the number of between-year recaptures is too low to enable a population estimate. This also means that within year recapture data is not available.

It should be noted that if obtaining a population estimate were deemed to be a high priority, then changes could be made to the current field program to try and increase the number of between-year recaptures; this is not currently the focus of the field program. Given the vast amount of working knowledge we have accumulated over the years about the Creston population, LIPI habitat use and behaviour it should be possible to custom tailor the surveys to achieve this goal. This would involve dedicating resources to surveying at specific sites at specific times of the year when and where it is known that there is a high probability of detection, but given finite resources it may involve a shift away from the current priority of the project which is to locate egg masses for the Bummers Flats reintroduction and bolstering the Vancouver Aquarium captive assurance colony. It is recommended that this be discussed by the BC-NLFRT and program priorities decided upon.

This is the first year that photo recognition has not been done. As a result, none of the data summaries provided in the results for the 2014 field season include information on unique individuals as it is impossible to differentiate between observations of individuals versus recaptures without doing the photo recognition.

4.9 Morphometrics of Creston LIPI

Weight and SVL measurement data was collected for 205 LIPI observations during the 2014 field season. This data could be used to compare YOY sizes between regions, for example at the Bummers Flats reintroduction site or the Brisco reintroduction site, however in order to be accurate, comparisons would have to be made on animals observed at the other sites during the same time frame, and date of metamorphosis would have to be factored in. There would likely be some regional variation in rate of development in YOY due to site specific differences in habitat (for example Brisco site being flooded with cold river water in 2013 which would slow the rate of tadpole development since it is temperature dependant) and foraging quality but data such as size at metamorphosis and growth rates could be compared to provide an index of site suitability.

4.10 Vancouver Aquarium Captive Assurance Colony

The new method of packaging the tadpoles up for transfer and flying them from Trail to Vancouver via Pacific Coastal Airlines worked well and was cost effective. No issues with tadpole metamorphosis were reported this year.

A comparison of pre-brumation YOY morphometrics between the captive bred animals held back from the Brisco release and the Creston wild source indicate that the captive bred animals appear to be smaller. The reason for this difference should be investigated.

4.11 Bummers Flats Reintroduction

For the 2015 field season, it is recommended to continue with phase 2 of the North Bummers Flats reintroductions status quo. As this will be year 5 of an initial 5-year commitment to the project, results of the project should be reviewed by the BC-NLFRT prior to the F17 funding cycle with discussions on how to proceed for 2016.

4.12 Leach Lake drawdown

In the fall of 2013 the drawdown at Leach Lake 4 was completed by the CVWMA to remove the dense monotypic stands of cattails that had formed since 2009 and during the spring of 2014 LIPI were detected calling in the area. This provides further evidence that LIPI may naturally recolonize an area if the habitat is restored, as the same results were observed the last time a drawdown was completed in the area in 2004. While no egg masses or tadpoles were detected at the site this year, YOY were detected in the summer, providing evidence that successful breeding did occur in Leach Lake; however it is unclear whether or not pond 4 was their natal pond or if they came from another pond in Leach Lake. YOY surveys would have to be conducted while metamorphosis was occurring to conclusively determine if breeding occurred in pond 4.

In 2014 other amphibian species were also detected using the restored site, including Western toads, long-toed salamanders and Columbia spotted frogs. Since some cattail ingrowth was detected during the summer of 2014, it is recommended that the CVWMA monitor the vegetation ingrowth to ensure it does not become a dense monotypic stand of cattails again.

4.13 Habitat Concerns

A number of habitat-related concerns that could potentially have negative impacts on the Creston LIPI population have been identified and brought to the attention of the BC-NLFRT and land managers over the years but to date these complex issues have not been prioritised for action. It is recommended that the BC-NLFRT discuss and rank these threats for the Creston population.

4.14 Additional Recommendations

In addition to recommendations outlined in the Discussion, below is a list of some additional recommendations for consideration:

- Continue to provide founder tadpoles to bolster the Vancouver Aquarium captive assurance colony
- Ensure future LIPI work planning at Bummers Flats involves everyone planning to do work on site so that work plans can be developed cooperatively
- Health, condition and hatch out success of wild laid egg masses sourced from Creston should be factored into the BC-NLFRT decision matrix for egg mass translocation; egg masses that are not in good health or condition and have below average hatch out should be omitted from the tally.
- If resources available, survey agricultural fields to follow up on anecdotal reports of LIPI in the fields during harvest as this could potentially be a significant source of mortality
- The BC-NLFRT should have a workshop to discuss priorities of the field program, this could be especially important now that population appears to be increasing
- Resolve the road mortality issue
- Continue working with the Nature Conservancy of Canada on the Frog-bear property management and farm planning:
 - Ensure new farm lease holder is aware of LIPI use of property and preferred farming practices
 - Discuss possible enhancement projects

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Appendix 1. CVWMA Overview Map

| H0GO station | Nochamol caling station (SAS) |
| Nochamol caling station (

East Duck Lake esting Area (EDLNA) West Duck Lake esting Area (WDLNA CVWMA 2014 - Northern Leopard Frog Observations - Duck Lake Date Updated: 16/02/2015 Coordinate System: WGS 1984 UTM Zone 11N (b) LIPI YOY summer - Dead 🔳 LIPI adult spring - Dead 🛧 LIPI Juvenile spring

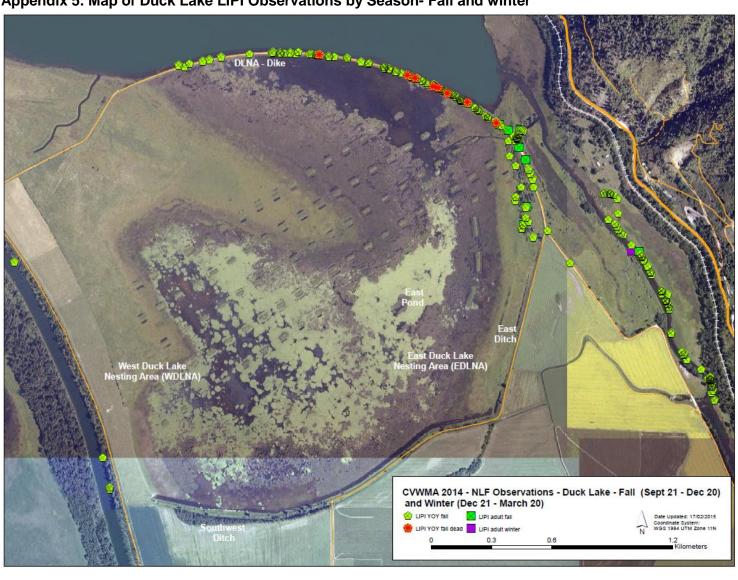
Appendix 2. Map of Duck Lake LIPI Observations – all seasons

DLNA - Dike East Ditch East Duck Lake esting Area (EDLNA) CVWMA 2014 - NLF Observations - Duck Lake - Spring (March 21 - June 20) 0.25

Appendix 3. Map of Duck Lake LIPI Observations by Season- Spring

East Duck Lake esting Area (EDLNA) CVWMA 2014 - NLF Observations - Duck Lake - Summer (June 21 - Sept 20) Date Updated: 17/02/2015 Coordinate System: WGS 1994 UTM Zone 11N

Appendix 4. Map of Duck Lake LIPI Observations by Season- Summer



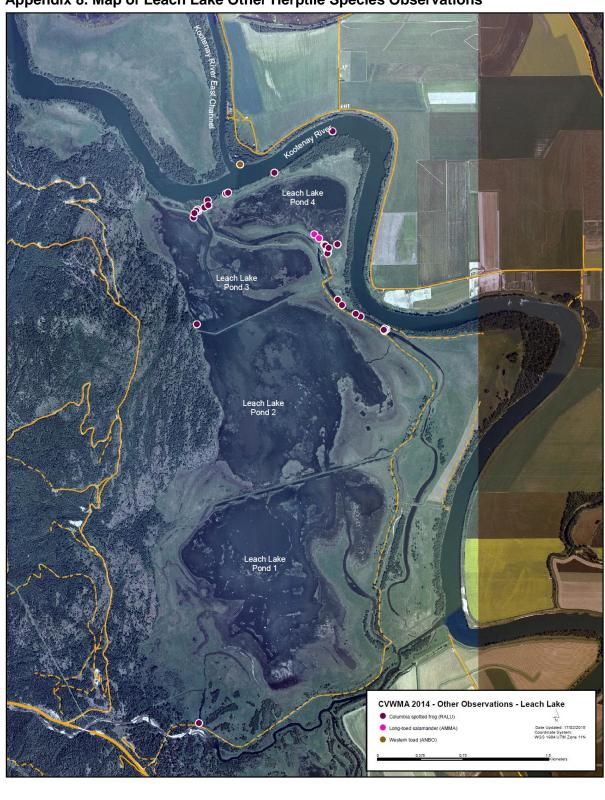
Appendix 5. Map of Duck Lake LIPI Observations by Season- Fall and winter

DLNA - Dike East Duck Lake sting Area (EDLNA) Date Updated: 17/02/2015 Coordinate System: WGS 1984 UTM Zone 11N CVWMA 2014 - Other Species Observations - Duck Lake Pacific chorus frog (PSRE)
 western terrestrial garter snake (THEL) O Common garter snake (THSI) - dead Western painted turtle (CHPI) 🚳 western terrestrial garter snake (THEL) - dead Long-toed salamander (AMMA)

Appendix 6. Map of Duck Lake Other Herptile Species Observations

Leach Lake Pond 2 CVWMA 2014 - NLF Observations - Leach Lake

Appendix 7. Map of Leach Lake LIPI Observations



Appendix 8. Map of Leach Lake Other Herptile Species Observations

Bummer's Flats release site, tadpole trapping and temperature data logger locations 2014 ear of Orthophotography - 2004 North Bummers Map Extent

Appendix 9. Bummer's Flats Overview Map

Bummer's Flats Northern Leopard Frog Observations 2014 North Bummers

Appendix 10. Map of Bummer's Flats LIPI Observations