Western Painted Turtle visual surveys, habitat assessment and restoration, and eDNA sampling on the east coast of Vancouver Island, 2015

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Executive Summary

The distribution of the endangered Western Painted Turtle (*Chrysemys picta bellii*) remains poorly understood in many areas on Vancouver Island. This project builds on previous efforts during 2012 and 2013, 2014, and the objectives for the 2015 field season were to:

1) Conduct visual surveys in areas of poor coverage to determine presence of the Western Painted Turtle on the east coast of Vancouver Island
2) Conduct assessments for Western Painted Turtle habitat on the east coast of Vancouver Island
3) Collect and filter water samples for developing an eDNA sampling method to identify new turtle populations on Vancouver Island.
4) Plan and carry out turtle habitat enhancement at Diver Lake, Nanaimo

We searched Google Earth imagery of the east coast of Vancouver Island for with seemingly good turtle habitat and used our database of previously assessed wetlands to identify survey locations. We surveyed 65 wetlands and did 81 surveys. Of these wetlands 43 were surveyed for the first time, and we did a habitat and threat assessment at these wetlands. We found the Western Painted Turtle at Maple, Chemainus, and Summit Lakes for the first time and a photo was received of a Western Painted Turtle in the Koksilah River by the Kinsol Trestle. The habitat assessments identified 20 wetlands with highly suitable turtle habitat. Due to the rural or backcountry settings of the four new Western Painted Turtle locations, there were few threats to these populations. The main threat at Maple and Chemainus Lakes was sport fishing because turtles are known to bite at some types of fishing lures and could be injured or killed if hooked.

Sliders (*Trachemys scripta*) were observed in five wetlands and the Mississippi Map Turtle (*Graptemys pseudogeographica*) and the Reeves Pond Turtle (*Mauremys reevesii*) were observed in one location each.

The team decided to develop an eDNA marker system for Sliders to help improve the eDNA detection method for turtles, to increase the number of known populations of this species on eastern Vancouver Island, and to determine the overlap between WPT and RES. In collaboration with Dr. Brent (UNBC) and Melissa Todd (FLNR), we developed the field method for eDNA water sampling and sample preparation for the subsequent laboratory identification of Western Painted Turtle and Red-eared Slider DNA. Each water body was sampled at three sites, 50 m or more apart and environmental parameters (depth, turbidity, pH, dissolved oxygen and light intensity) were measured at each site. We took 1 L of water for each filter size and 15ml water samples for the Ficetola *et al*. (2008) precipitation method. In spring we collected 2 L of water to be filtered through a 0.45 µm and a 1 µm mixed cellulose ester (MCE) filter and 6 x 15 ml for the precipitation method from 7 locations. In the fall we re-sampled these locations as well as sampling 12 other locations. At the fall sample locations we collected 1x15 ml samples for the precipitation method, and 2 L of water to be filtered through a 0.45 µm and a 1 µm MCE filter, and 1L for filtering with a 10 µm polycarbonate track-etched (PTCE) filter. At three locations we also sampled 1L for filtering with a 5 µm PCTE filter. The 132 MCE filters, 40 PCTE filter and 303 precipitation vials were stored in a freezer in solutions appropriate for extraction methods and shipped to the UNBC molecular ecology lab after the field season, where the Biolinx-UNBC MITACS student has developed a set of objectives for prioritization of sample analysis, as well as completed the development and testing of the RES marker system. A separate UNBC report summarizes this analytical component.

Recommendations:

- Conduct visual surveys at these locations:
    - Chemainus, Maple, Divers, Morrell and Cathers Lake to estimate population size
    - Re-survey wetlands where Western Painted Turtle have not been seen and the habitat assessment resulted in a moderately or highly suitable turtle habitat score
    - Survey areas with anecdotal observations that have highly or moderately suitable turtle habitat or wetlands with such as the wetlands in the vicinity of the Kinsol Trestle Western Painted Turtle observation
    - Survey the many wetlands around Nanaimo to better understand the local distribution of the species
- Habitat enhancement at Divers Lake
    - Monitor use of installed logs
    - Enhance nesting grounds and protect Western Painted Turtle nests
- eDNA sampling
    - Investigate correlations between DNA detection and environmental parameters such as turbidity, pH, temperature, oxygen concentration, nuclease activity, conductivity, UV radiation, temperature, type of sediment and salinity measured when collecting water samples
    - Investigate correlations between DNA detection and time of year by sampling during spring, summer, and fall, and/or by sampling a few locations throughout the year
    - Investigate the effect of wetland type and size (lake, marsh and pond) and sampling effort on DNA detection

Cover photo: Western Painted Turtle photographed at Maple Lake on 1 Oct 2015, © C Engelstoft

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Acknowledgments

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Introduction

Currently the distribution of the Endangered (COSEWIC, 2006) Western Painted Turtle (*Chrysemys picta bellii*) on Vancouver Island is best known in the Capital Region District (CRD) and the Alberni Valley (Engelstoft and Ovaska, 2015), and less so in other parts of the island (Engelstoft and Thorpe 2015). More than 370 surveys in 134 wetland have been conducted in the Capital Region District (CRD) and Alberni Valley (Engelstoft 2015 unpublished data), and 103 surveys in 52 wetlands have been surveyed in other areas of Vancouver Island (Engelstoft and Thorpe 2015). Despite these efforts there are still no observations of Western Painted Turtles in Duncan, Courtenay and Quadra Island where we have anecdotal sightings of the species (Chatwin, pers. com). Clarifying the distribution on Vancouver Island is a critical first step for recovery planning and management of Western Painted Turtle populations and their habitat.

Visual surveys for Western Painted Turtles (RISC 1998) are a relatively expensive and time-consuming approach because of many factors influencing detection such as unexpected change in weather, disturbance from recreational users, and logistical obstacles. It is often necessary to re-visit sites multiple times in order to increase the probability of detection when turtles are not observed. "Not detected" does not rule out the presence of the species, which is an issue in locations with small populations. Environmental DNA (eDNA) methods hold promise as a complementary, and more cost-effective approach for this species (Diaz-Ferguson *et al*. 2014). This method has been effective at detecting aquatic amphibians (e.g., Ficetola *et al*., 2008; Pilliod *et al*., 2013), fish (e.g., Thomsen *et al*., 2012; Jerde *et al*., 2011) and snakes (Piaggio *et al*., 2013). We believe that the Western Painted Turtle is a good candidate for using the eDNA detection approach because it spends most of its time in the water. The genetic primer for the species have been developed for the Painted Turtle (pers. com. Dr. Brent Murray, UNBC), which are necessary for eDNA-based species inventory.

The objectives for this project were to:

1) Conduct visual surveys in areas of poor coverage to determine presence of the Western Painted Turtle on the east coast of Vancouver Island
2) Conduct assessments for Western Painted Turtle habitat on the east coast of Vancouver Island
3) Collect and filter water samples for developing an eDNA sampling method to identify new turtle populations on Vancouver Island.
4) Plan and carry out turtle habitat enhancement at Diver Lake, Nanaimo

This report presents results of the visual surveys and habitat assessments conducted from June to September 2015 and water sampling and filtering efforts. The genetic analysis will be reported separately by the UNBC molecular ecology laboratory.

Methods

Visual Surveys and Habitat and Threat Assessments

Visual surveys followed the BC standard (RISC 1998), and we used three approaches to select visual survey sites. We re-surveyed sites that in previous years have been assessed to have good habitat; surveyed new wetlands identified using Google Earth; and surveyed locations with anecdotal turtle observations.

We identified wetlands on Google Earth imagery and selected previously assessed wetlands without Western Painted Turtle sightings. On Google Earth we looked for wetlands that were at low elevations (<250 m asl[1]) and had areas of shallow water; both attributes help predict suitable turtle habitat. Accessibility was also a factor in site selection. From our database of Western Painted Turtle habitat assessment from previous years, we selected wetlands 21 wetlands with good turtle habitat that had previously been surveyed but where no Western Painted Turtles had been observed (Engelstoft and Thorpe 2015, Engelstoft and Ovaska, 2012). Wetlands were also selected in three areas (Quadra, Courtney, and Cobble Hill) with anecdotal observations of Western Painted Turtles and on Denman Island where un-identified turtles had been observed. This provided us with at list of 60 potential wetlands to survey.

The BC standards for visual turtle surveys at the present/not detected level (RISC 1998) require visual inspection of potential basking sites and the water surfaces, particularly in areas along shoreline and sheltered spots. On warm summer days, it is more effective to scan shallow waters for swimming turtles and protruding heads or carapaces as the turtles do not need to come out of the water to bask. We used binoculars and a telescope to scan for basking turtles from either a boat or from vantage points on land. For smaller water bodies, we examined the entire perimeter and for larger lakes, we examined habitats most likely to support turtles (such as shallow bays and sheltered areas that were accessible). We noted the date, start and end time of each survey, wind speed, precipitation, air and water temperature (C^o), and recorded the number and species of turtles observed.

We conducted a habitat suitability and threat assessment during the first visit to each site (Ovaska and Engelstoft, 2010). The habitat features described included:

- landscape context (backcountry, rural, urban);
- upland habitat (coniferous, deciduous, mixed-wood forest, other);
- wetland type (lake, pond, marsh, other) with modifications noted (dug-out, dams, beaver maintained); percentage cover of littoral zone with emergent and submerged vegetation;
- percentage cover of vegetation types within riparian zone along the perimeter of the water body (forest, shrubs, grass-herbs, development, or other);
- availability of basking logs (none, low, moderate, high);
- availability of nesting sites (confirmed, potential, not identified), including a description of identified sites.

The last step was to indicate the overall habitat suitability as low, moderate, or high based on the availability of basking, foraging, and nesting opportunities (Ovaska and Englestoft, 2010).

[1] asl – above sea level

We assessed potential threats by examining the degree of threats from:

- roads (paved or unpaved);
- recreation (motorized or non-motorized);
- pets; exotic species (bullfrogs and alien turtle species);
- residential development; other urban development; agriculture; forestry; grazing; water withdrawal; and other sources.

Environmental DNA

eDNA Water Sampling Approach

During the course of the project we decided to develop an eDNA primers or marker system for Sliders (*Trachemys scripta*) to increase the number of turtle-occupied ponds from which to sample for the study of turtle eDNA detection probability. There are 29 lakes or ponds on Vancouver Island where the Western Painted Turtle are known to occur and another 36 lakes or ponds where the Sliders have been found. We sampled all known Western Painted Turtle locations but we chose to select a subset of 39 easily accessible locations that could be sampled (Table 1). Perimeter length (<1 km, >=1 & <2 km, and >=2 km) and maximum sighted numbers of turtle (1 turtle, >1 & < 5 turtles, and >5 turtles) were used to categorize the pool of sampling locations (Table 2). We did not have any more reliable indicator of turtle density for categorization of wetlands for eDNA sampling because the inconsistencies of count methods and unusual weather patterns over the past years.

Our objective was to investigate the impact of filter sizes (0.45 and 1.00 µm) of mixed cellulose ester (MCE) on detection probability. A further review of recent literature (i.e., Turner et al. (2014)) prompted us to also try a 5 and 10 µm polycarbonate track-etched (PTCE) filter during the fall sampling. We collected samples from wetlands with known Western Painted Turtles and Slider populations. At these wetlands we collected the water samples from three sites[2] that were ≥50 m apart to ensure independence, and we attempted to sample within 15 m of a Western Painted Turtle sighting or in areas known to be used by turtles. At each of these sites, we collected depending on sampling treatment between 1 and 4 L of surface water samples in Nalgene bottles (Thermo Scientific Nalgene Wide-Mouth Economy Bottle, HDPE, 1000mL) similar to Grob (2014) and 1 to 6 x 15 ml surface water samples to precipitate the DNA (Ficetola et al. 2008). We took 6 “Ficetola” -samples from a sub-set of sampling sites in order to test whether higher samples intensity would increase detection probability. We used new gloves at each site, and rinsed the bottle in surface water at the sampling site at least three times before collecting the water. Our samples were transported and stored in a cooler with ice-packs. We also collected environmental covariate data such as light intensity, pH, turbidity, dissolved oxygen, and weather data (Appendix 1).

Because Western Painted Turtles often live in slow-moving and eutrophic waters, the high turbidity in the waters can cause filters to clog up. At sampling sites with dense vegetation, we attempted to obtain our water samples from areas patches of water without vegetation among the floating aquatic vegetation.

Boat, equipment, boots, waders and paddles were cleaned as prescribed by the “Interim Hygiene Protocols for Amphibian field staff and researchers" (MOE, 2008) when moving between sites.

[2] Site refer to the site where water samples were obtained

Table 1. List of 39 selected eDNA water sampling locations on Vancouver Island

Location	WPT max # observed[A]	RES max # observed[B]	Unk Turtle	Previous eDNA Sample	Elevation	Perimeter (km)	Wetland Size/ Turtle Density[C]
Buttertubs Marsh	3	10		2014	57	2.1	L/H
Cathers Lake	1	10			99	1.4	M/H
Morrell Lake	1	1			175	1.3	M/L
Divers Lake	13			2014	105	1.7	M/H
Lost Lake		1			165	0.6	S/L
Westwood Lake		4			163	5.2	L/M
Sywash Pond			1		99	2.9	L/L
Long Lake			2		109	3.6	L/M
Chemainus Lake	2				115	1.4	M/M
Mayo Lake		1			187	0.8	S/L
Jim's Pond		20			62	0.8	S/H
Kwassin Lake		2			183	1	M/M
Wild Play Pond		1			171	0.1	S/L
Airport Wetlands	37	1			83	3.2	L/H
Summit Lake	9				382	1.2	M/H
Sumner Lake	9		1		79	2.3	L/H
Ash main 26K	4				116	0.9	S/M
Patterson Lake	1				92	3.4	L/L
Turnbull Lake	5				267	3.6	L/H
Loon Lake		3			365	1.2	M/M
Little Turtles Lake	4	1			88	1.7	M/M
Tilley Drive Pond		1			43	0.4	S/L
Beaver Lake	20	13		2014	59	3.6	L/H
Florence Lake	2	3			76	1.7	M/M
Glen Lake	3	7			65	2	L/H
Eagle Lake	1	1			90	1.3	M/L
Kemp Lake	1	3			34	2.6	L/M
Langford Lake	37	9		2014	64	5.0	L/H
Matheson Lake	3	5			32	3.4	L/H
Swan Lake	11	4		2014	14	1.2	M/H
McKenzie Lake	4	1			62	1.7	M/M
Beckwith Park		15			62	0.2	S/H
Durrance Lake		6			184	1.7	M/H
Kings Pond		3			62	0.3	S/M
Matson Lake		1			123	1.4	M/L
Prospect Lake		1			123	4.4	L/L
Thetis Lake		16			52	4.4	L/H
Maple Lake	1				136	2.6	L/L
Saratoga Golf Course		1			5	0.1	S/L

[A] WPT – Western Painted Turtle

[B] RES – Red-eared Slider

[C] See Table 2 for explanation of codes for Large, Medium, or Small sized wetlands and Low, Medium, or High turtle density

Table 2. Categorization of pond and lake sizes based on the perimeter length and estimated relative turtle density based on number of observed turtles.

Pond size (km perimeter)	Low (L) (1 turtle)	Medium (M) (>1 & < 5 turtles)	High(H) (>5 turtles)	Total
< 1 km (S)	5	2	2	9
>=1 & <2 km (M)	4	6	5	15
>=2 km (L)	3	3	9	15
Total	12	11	16	39

Water sample preparation

We operated two filtering set-ups, each consisting of a Masterflex L/S economy variable-speed drive (20 to 600rpm, 115 VAC) with a peristaltic Masterflex Standard pump head (L/S 24 tubing, PC housing, SS rotor) that was attached to a Thermo Scientific Nalgene polypropylene filtering flask (1000ml). A disposable Thermo Scientific™ Nalgene™ Analytical Test Filter Funnel was used and changed between each sample. We used mixed cellulose ester (MCE) membranes with a diameter of 47mm and pore size of 0.45 µm and 1.0 µm, and as many filters that were necessary to filter 1 L of water (Appendix 1). During the fall sampling we also filtered 1 L of water with a pore size of 10 µm polycarbonate track-etched (PTCE) filter and a few sites using a 5 µm PTCE filters. New filters were handled with sterile forceps (flamed with a lighter). Filters were carefully removed from the filtering funnels, folded and placed in a 2 ml cryogenic vial. Each vial could hold up to two folded filters from the same sample, and filters were stored in 95% ethanol at -20°C until extraction (3-6 months). We terminated filtering when we reached ten vials from one sample. To avoid cross contamination, we changed gloves every time we switched tasks (e.g., writing notes, labelling cryogenic vials, sterilizing tweezers etc.). MCE samples were submerged in 95% ethanol and the PTCE filters were in 700 µM of CTAB buffer (Turner et al 2014) in 2 ml cryovials, and stored in a freezer before being shipped to the laboratory.

To increase the detection probability of the Ficetola et al (2008) precipitation method, we prepared 6 x 50 ml vials per site during summer sampling. We followed the procedure outlined in Appendix 1.

In fall only one 15 ml water sample was obtained per location that had not been sampled previously that year. A full summary of the number and type of replicate filter and precipitation samples collected by season at a total of 19 wetlands can be found in Appendix 2.

Divers Lake Turtle Habitat Enhancement Project

The Western Painted Turtle was first reported at Diver Lake (Nanaimo) on 20 June 2012 (pers. com. H. Kimura, 2015). A turtle habitat assessment conducted in 2014 indicated that there were few basking logs and only floating rhizome mats available for basking at the site (Engelstoft and Thorpe 2015). Nesting is known from two areas around the lake. We found nesting attempts under the canopy along the trail that runs from the parking lot to the fishing dock and in a more suitable nesting habitat located

on the field with the bike tracks in 2015. Turtles have been known to nest on this field in previous years as well.

The threats to the Western Painted Turtle at Diver Lake include bait on fishing lures that occasionally catch adult turtles, the threats to juvenile turtles from Bullfrogs (*Lithobates catesbeianus*), and disturbance of nesting areas located in a mountain bike park. Watermilfoil (possibly Eurasian Watermilfoil, *Myriophyllum spicatum*) is abundant, but is not considered a threat to the turtles (Engelstoft and Thorpe 2015).

Results

Visual Surveys

Surveys took place between 28 May and 5 November, 2015, from Victoria in the south to Black Creek in the north on the east coast of Vancouver Island (Figure 1). We conducted a total of 81 surveys of which 4 surveys were in collaboration with a Habitat Acquisition Trust project in the Alberni area (Table 3). We surveyed a total of 65 wetlands out of which 43 were surveyed for the first time in 2015 (Figure 1, Table 3).

A total of 4 wetlands were added to the list of known Western Painted Turtle locations in 2015 (Figure 2). Western Painted turtles were observed in Maple Lake (1 July, 2015, n=1) in the Comox Valley, Chemainus Lake (17 July 2015, n=2) by Chemainus, and Summit Lake (28 May, 2015; n=9) by Port Alberni. Todd Carnahan, a former colleague from Habitat Acquisition Trust observed and photographed a Western Painted Turtle in the Koksilah River by the Kinsol Trestle on 23 Aug 2015.

Two areas were targeted after getting reports of turtle observations. We conducted 6 wetland surveys in the vicinity of the Western Painted Turtle observation in the Koksilah River, in an attempt to find the origin or destination of the observed turtle. The late survey time and hot weather impeded our ability to detect turtles; it is recommended that several of these wetlands be surveyed during the spring in future years. On Denman Island a landowner informed me (CE) about turtle observations in Chickadee Lake, so I took the opportunity to assess turtle habitat in 7 wetlands on the island. Six of the 7 wetlands had highly suitable habitat, and more surveys should be undertaken on this island during the correct survey conditions.

We attempted surveying several ponds in the Comox and Merville area which were either not in the expected location (n=1) or on private land we could not gain access to (n=3).

The introduced Slider (*Trachemys scripta*) was found at 5 wetlands (Table 3). At Sywash Marsh, north of Nanaimo, we saw holes that indicated that turtles had attempted to nest on the shore of this wetland, but we saw no turtles during the survey. A Mississippi Map Turtle (*Graptemys pseudogeographica*) was seen in Mayo Lake, Cowichan Vallley, again this year, and we saw a Reeves Pond Turtle (*Mauremys reevesii*) at Buttertubs Marsh in Nanaimo.

Survey efforts were measured using person-hours, and we surveyed for 63 person-hours (Table 3). The average time spent at a site was 1 person-hour (Standard deviation = 1 person-hour), and the most time at one site was 4.8 person-hours.

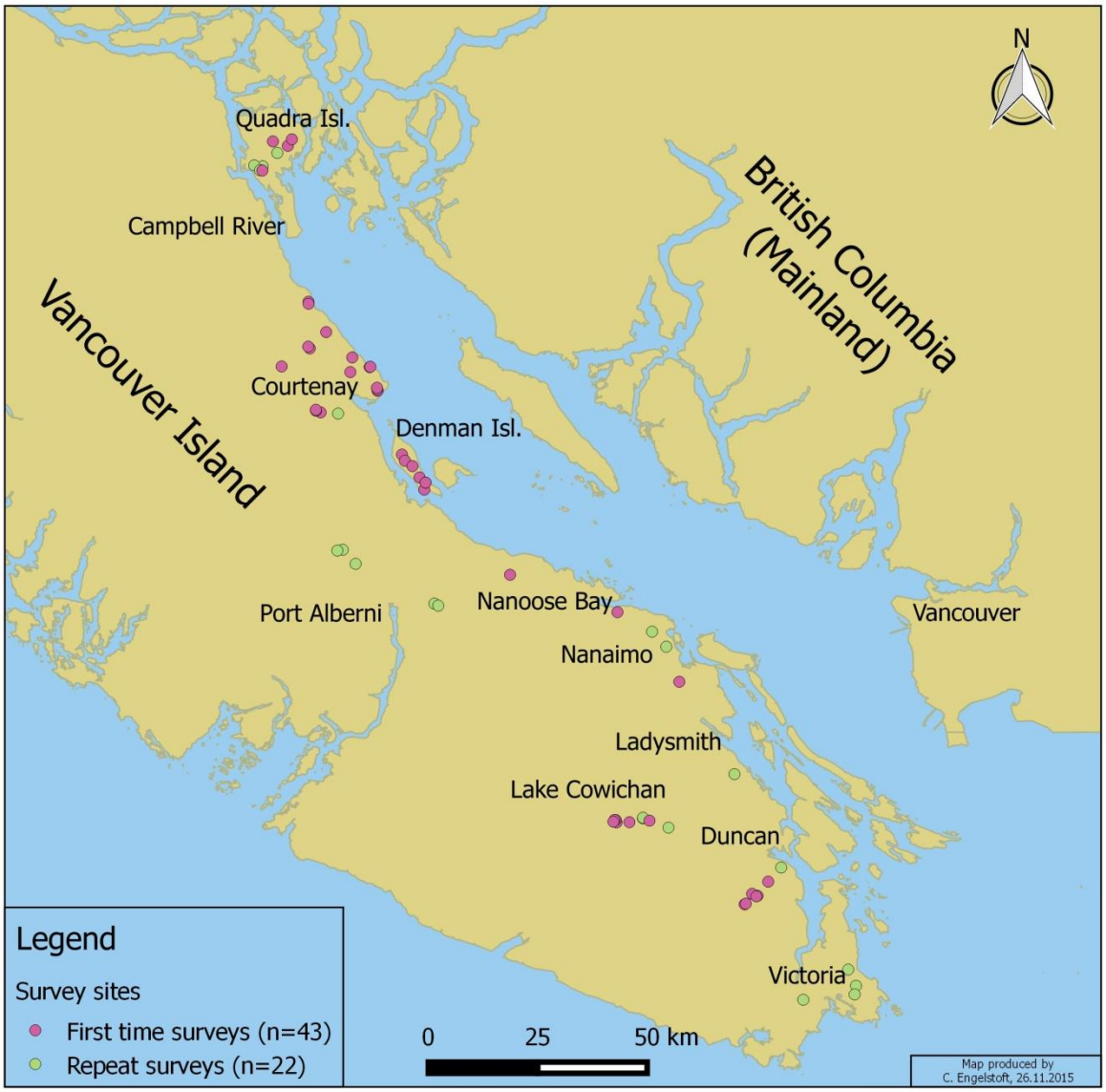

Figure 1. Western Painted Turtle survey locations (n=65) on Vancouver Island, 2015.

Table 3. Western Painted Turtle survey locations, coordinates, dates and results, on the east coast of Vancouver Island, 2015

WPT = Western Painted Turtle; RES = Red-eared Slider

Site ID	Location	Easting (UTM)	Northing (UTM)	Year of 1st survey	2015 Survey Dates[1]	Max WPT #[1]	Max RES #	Person-hrs	# surveys in 2015
1	Airport Wetlands	358400	5466114	2008	(28 May), 24 Jun, 29 Oct	(14)	0	1.5	3
13	Loon Lake	376385	5457036	2008	(28 May), 29 Oct	0	0	1.0	2
19	Sumner Lake	355463	5469326	2008	24 Jun, 29 Oct	0	0	3.0	2
28	Beckwith Park	472743	5369846	2008	5 Nov	0	0	1.0	1
47	Langford Lake	460686	5366651	2008	4 Nov	0	0	2.0	1
70	Swan Lake	472414	5367870	2008	5 Nov	0	0	1.0	1
91	Buttertubs Marsh	429378	5447204	2008	10 Jun, 20 Oct	0	1	1.5	2
120	Ash Main, 26 km pond	354228	5469136	2013	24 Jun, 29 Oct	0	0	2.0	2
132	Summit Lake (slough)	377274	5456612	2013	(28 May), 23 Jun, 29 Oct	(9)	0	3.0	3
27a	Beaver Lake	470962	5373534	2008	4 Nov	0	0	1.5	1
Cw06	Grant Lake	424054	5408055	2014	9 Jun	0	0	0.3	1
Cw07	Kwassin Lake	429889	5405928	2014	9 Jun	0	0	1.0	1
Cw09	Private Pond, Cowichan Bay	455700	5397000[2]	2014	22 Oct	0	0	0.7	1
E01	Little Morte Lake	337175	5556822	2012	5 Jun	0	1	1.2	1
E02	Morte Lake	335228	5557042	2012	5 Jun	0	0	2.8	1
E03	Mud Lake	336551	5555919	2012	4, 5 Jun	0	1	1.8	2
E04	September Lake	340496	5559859	2012	5 Jun	1	0	4.0	1
E11	Maple Lake, Cumberland	354392	5500395	2012	1 Jul, 21 Oct	2	0	4.8	2
E16	Chemainus Lake	444964	5418126	2012	9, 17 Jun. 2 Jul, 22 Oct	2	0	3.5	4
E17	Mayo Lake	429889	5405928	2012	9 Jun	0	0	0.5	1
E26	Diver Lake	426134	5450644	2014	2 Jun, 15 Oct	0	0	0.5	2
E29	Lost Lake	428041	5453127	2014	23 Oct	0	0	1.0	1
E35	Village Bay Lake	342910	5561487	2015	4 Jun	0	1	0.2	1
E36	Main Lake	343836	5562976	2015	4 Jun	0	0	0.3	1
E37	Hamilton's Marsh	393689	5463611	2015	4 Jun	0	0	0.5	1
E38	Rees Lake	337079	5555840	2015	4, 5 Jun	0	0	1.6	2
E39	Stramberg Lake	339504	5562494	2015	5 Jun	0	0	0.7	1
E40	Mesachie Lake	418021	5407127	2015	9 Jun	0	0	1.2	1
E41	Wild Play Pond	417838	5407653	2015	9 Jun	0	0	0.3	1
E42	Imadene Pond	417579	5407690	2015	9 Jun	0	0	1.2	1
E43	Bear Lake	417306	5407285	2015	9 Jun	0	0	0.7	1
E44	Beaver Lake (Cowichan)	420959	5407169	2015	9 Jun	0	0	0.7	1
E45	Hudson's Lake	425551	5407520	2015	9 Jun	0	0	0.7	1
E46a	Lazo Nature Park	363422	5505582	2015	1 Jul	0	0	1.7	1

Site ID	Location	Easting (UTM)	Northing (UTM)	Year of 1st survey	2015 Survey Dates[1]	Max WPT #[1]	Max RES #	Person-hrs	# surveys in 2015
	(a)								
E46b	Lazo Nature Park (b)	363352	5505915	2015	1 Jul	0	0	1.2	1
E46c	Lazo Nature Park (c)	363277	5506291	2015	1 Jul	0	1	1.2	1
E47	Black Lake, Cumberland	350404	5500666	2015	1 Jul	0	0	0.0	1
E48a	Courtenay Fish and Game (Lower)	349351	5501084	2015	2 Jul	0	0	0.3	1
E48b	Courtenay Fish and Game (Middle)	349337	5501093	2015	2 Jul	0	0	0.3	1
E48c	Courtenay Fish and Game (Upper)	349331	5501318	2015	2 Jul	0	0	0.5	1
E49	Anderson Lake	341501	5511147	2015	30 Jun	0	0	0.3	1
E50	Meldas Marsh	357626	5513194	2015	30 Jun	0	0	0.3	1
E51a	Saratoga Golf Course	347634	5525904	2015	30 Jun, 21 Oct	1	0	0.3	2
E51b	Saratoga Golf Course	347641	5525443	2015	30 Jun	0	0	0.3	1
E52	6930 Railway Ave. (Cheri and Sally)	347930	5515265	2015	30 Aug	0	0	0.3	1
E53	4705 Gail Creek	357192	5509862	2015	29 Aug	0	0	0.3	1
E54a	Little River Nature Park, Pond W	361604	5511000	2015	29 Aug	0	0	0.3	1
E54b	Little River Nature Park, Pond E	361772	5511067	2015	29 Aug	0	0	0.3	1
E56	Spur Rd	347564	5515668	2015	30 Aug	0	0	0.3	1
E57	William Beach/Hagen Wetland	351663	5518992	2015	29 Aug	0	0	0.3	1
E58	Kinsol Trestle,	448993	5390820	2015	10 Sep	(1)	0	0.3	1
E59	Chickadee Lake	368984	5491042	2015	23 Sep	0	0	0.3	1
E60	Inner Isl Nature Reserve	369656	5489632	2015	23 Sep	0	0	0.5	1
E61	Lacon/Duster Rd	374079	5483047	2015	23 Sep	0	0	0.5	1
E62	Graham Lake	373006	5485829	2015	23 Sep	0	0	0.5	1
E63	Central Park Wetland	371386	5488403	2015	23 Sep	0	0	0.5	1
E64	Lake Side Dr Pond	432356	5439202	2015	22 Sep	0	0	0.5	1
E65	Morrison Marsh	374384	5484640	2015	23 Sep	0	0	0.0	1
E66	McFarlane Wetland	374384	5484640	2015	23 Sep	0	0	0.0	1
E67a	E Kingburn Pond,	450233	5390372	2015	10 Sep	0	0	0.0	1
E67b	W Kingburn Pond	449962	5390245	2015	10 Sep	0	0	0.0	1
E68	3681 Kingburn Dr	452676	5393596	2015	10 Sep	0	0	0.0	1
E69	Pond A, Renfrew Rd (K7)	447284	5388427	2015	10 Sep	0	0	0.0	1

Site ID	Location	Easting (UTM)	Northing (UTM)	Year of 1st survey	2015 Survey Dates[1]	Max WPT #[1]	Max RES #	Person-hrs	# surveys in 2015
E70	Pond B, Renfrew Rd (K6)	447556	5388581	2015	10 Sep	0	0	0.0	1
E71	Sywash Pond	418221	5455087	2015	10 Jun	0	0[3]	3.5	1
Total	*65 Locations*							*63*	*81*

[1] - Numbers in () indicate surveys done in collaboration with Habitat Acquisition Trust
[2] - Coordinates randomized to project privacy
[3] - Saw holes from turtle diggings

Figure 2. Known Western Painted Turtle (WPT) populations north of the Capital Region District on the east coast on Vancouver Island, 2015

Habitat Assessment

The very warm weather during June, July and August hampered our ability to spot turtles, because instead of basking in the sun out of the water they "basked" submerged only with their tip of their nose out of the water. We observed this "basking" behaviour among floating vegetation that makes detection even harder. As a consequence we assessed both known occupied and potential turtle wetlands.

We assessed 43 wetlands during June and July surveys, of which 19 had highly suitable habitat and 17 had moderately and 5 had low suitable habitat (Appendix 3). One wetland (E70) could not be assessed because it was dry and another visited site (not in Appendix 3) was a river, but they could both functions as "stepping stone" habitat used during movement form one wetland to another. The river bed where the Western Painted Turtle was observed had no aquatic vegetation.

All the assessed wetlands were classified as rural (n=32) or backcountry (n=11). The wetlands were categorized as lakes (n=10), natural or human-made ponds (n=22), marsh/fen/slough (n=10) and river (n=1). The emergent vegetation along the shoreline varied from 0 to 100% cover, There were 8 wetlands with 0% emergent vegetation and 54% of the wetlands (n=23) had over 50% emergent vegetation cover along the shoreline. Floating vegetation also varied tremendously, but 26% of the wetlands (n=11) had more than 85% cover along the shoreline. The riparian upland areas had shrub cover greater than 60% at 21 of 42 sites, and forest patches or stands were present at all sites (see Appendix 3 for details about habitat assessment).

The habitat assessment of the three new lakes where the Western Painted Turtle were observed for the first time in 2015 indicated that they were situated in rural or backcountry settings and all had emergent and floating aquatic vegetation such as cattails, sedges and pond lilies. They had over 50% upland forest cover and the shrub cover around the water's edge were higher than 30%. Basking opportunities were good at all the lakes and nesting habitat in close proximity to the lake was restricted in all cases. The overall assessment for the three lakes was that they all had highly suitable habitat. Maple and Chemainus Lakes were assessed in 2012 prior to any knowledge of the Western Painted turtles being present at the site suggesting that habitat assessments can be used to guide visual survey efforts in the future.

Threat Assessment

We conducted a threat assessment at 42 sites (Appendix 4) regardless of whether the Western Painted Turtle was observed or not. The Kinsol Tressle site was not assessed because there was no real turtle habitat. At the Maple, Chemainus and Summit Lakes with new Western Painted Turtle observations this year only Summit Lake was assessed this year whereas the other two lakes were assessed in 2012. The assessments indicated that there was a low threat from non-motorized boats and fishing at these lakes. Summit Lake is a 1 km hike so it is well protected. Disturbance of the few nesting sites at these lakes might be a low threat.

Environmental DNA Sampling and Filtering

We sampled 19 ponds and lakes varying in size and turtle density (Figure 3). A total of 7 water bodies were sampled in the Alberni Valley (n=4), Nanaimo (n=2) and Chemainus (n=1) during June. For each of these locations we prepared 6 samples for the precipitation analysis (Ficetola et al. 2008), and filtered the water with a 0.45 and a 1.0 µm MCE filters. This was repeated in October for these locations, and we added filtering of 1 L with a 10 µm PCTE filter in the fall (Appendix 2).

During October and early November, sampling took place at 12 locations that were not sampled in June, including Langford, Swan and Beaver Lakes in the Victoria area (Appendix 2) that were sampled in 2014 (Engelstoft and Thorpe 2015). Filtering of samples at one of these locations (a private pond) failed due to high content of suspended organic matters that clogged the filters immediately and only precipitation samples were collected. In the fall the 6x15 ml Ficetola et al. (2008) sampling took place in Maple Lake, a private pond and Saratoga Golf Course pond.

With the exception of Ash Main 26 K and the Saratoga Golf Course ponds that were too small for more than 2 sample sites, we took samples from 3 sites established at each wetland (Appendix 2).

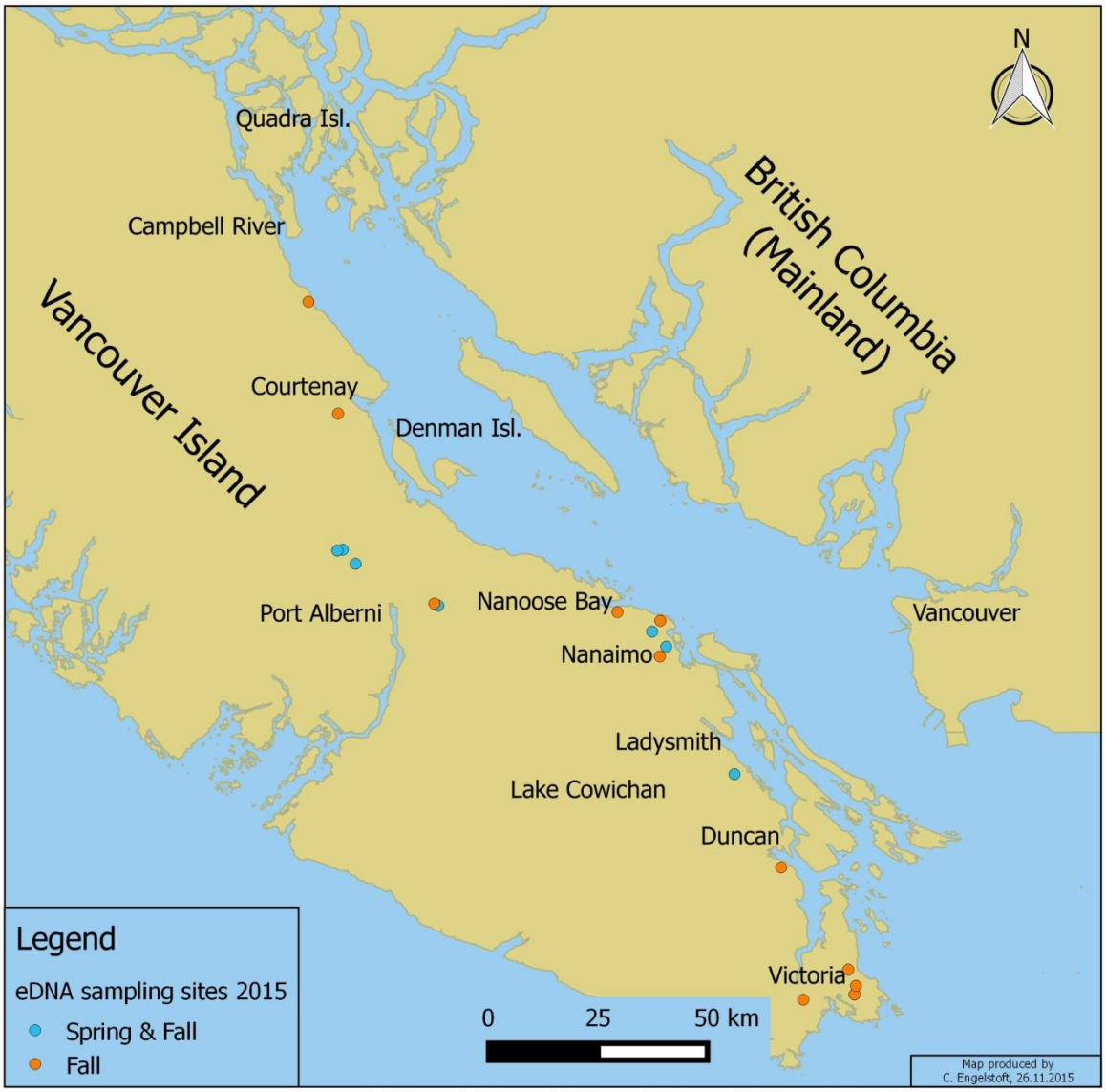

Figure 3. eDNA water sampling locations and seasonal sampling regime on Vancouver Island, 2015

Diver Lake enhancement project

Habitat enhancement activities and management guidelines for the Western Painted Turtle were developed during the summer of 2015 in the document called "Western Painted Turtle Habitat Restoration and Management Guidelines at Diver Lake, Nanaimo" (see plan in Appendix 5).

Following this plan, on 3 December 2015, we installed 10 logs to enhance turtle basking opportunities at Diver Lake (Figure 4). They were all about 3 m long and greater than 25 cm in diameter. Four larger logs were installed singly and six smaller logs were tied together in pairs to prevent rolling and improve suitability for basking turtles (Figure 4). They were anchored to trees and shrubs along the water's edge

with UV resistant nylon rope. They were all installed so they could be seen from the fishing dock for easy monitoring.

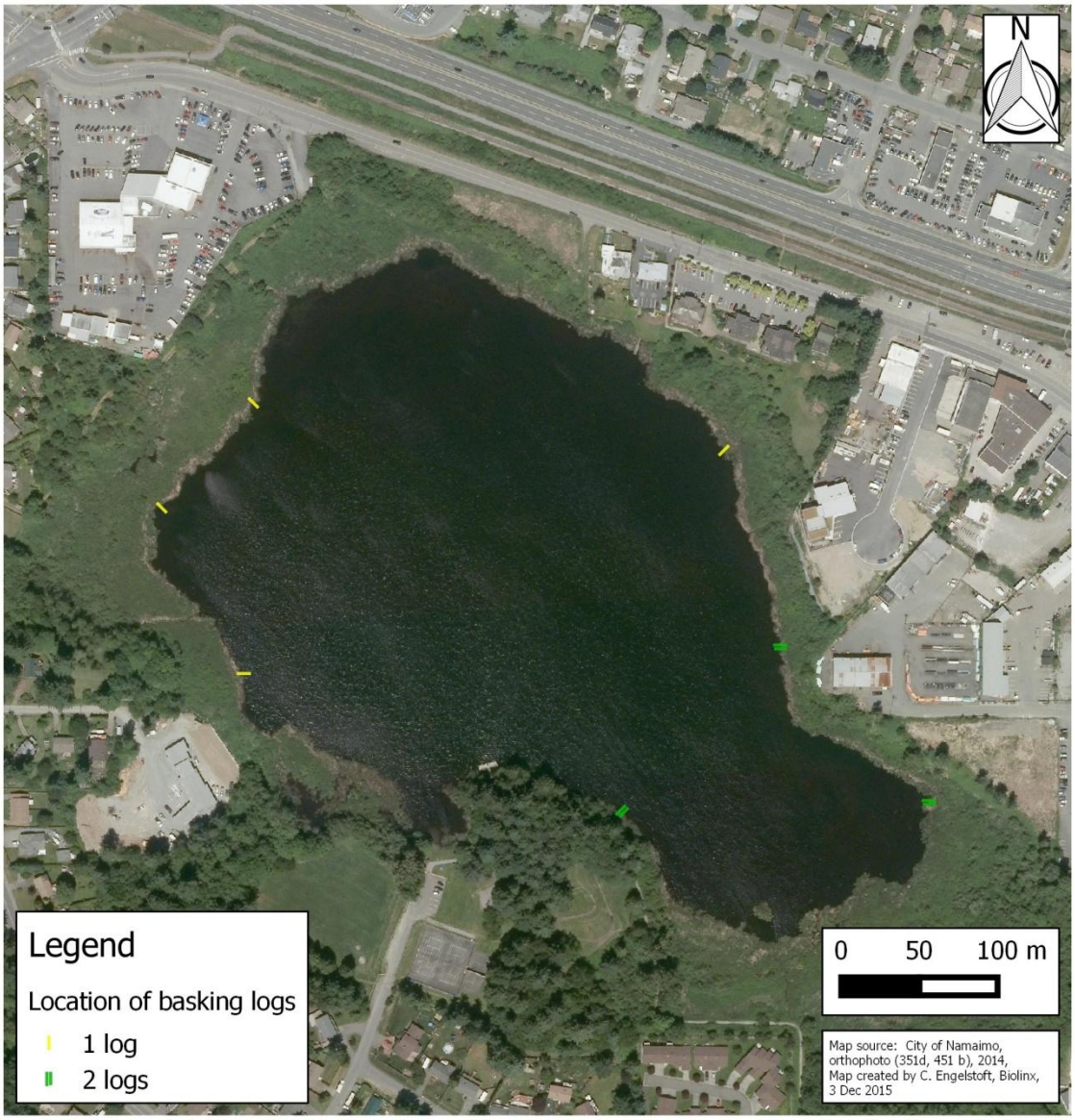

Figure 4. Location of turtle basking logs installed on 3 December 2015 in Diver Lake, Nanaimo

Discussion

Visual surveys

During the summer of 2015, even though hot weather impeded visual surveys, 4 new Western Painted Turtle populations were identified (Figure 2); this brings the total of known populations up to 33 on

Vancouver Island. The new Western Painted Turtle sites were broadly dispersed from just north of the CRD in the south to Courtney in the North (Figure 2).

More surveys need to be done to further our understanding of the distribution of this species on Vancouver Island, and with the identification of 40 new locations with highly or moderately suitable habitat there are many possible survey locations in the following years. These wetlands are clustered around Courtney, Nanaimo, Denman Island, and Cowichan Valley,. Denman Island should be surveyed in the future because of the many wetlands with good turtle habitat, the history of beaver activity that creates new wetlands, and the observation of unidentified turtles. Another area that should be surveyed is around Merville, because there are many wetlands with good turtle habitat. The Hamilton Marsh by Coombs, that we only had a chance to visit once, has good habitat, and it should be surveyed more in the future. Another area that look very interesting from Google imagery are the Illusion Lakes by Spider Lake that were not on our 2015 list of selected wetlands but should included in future turtles surveys..

At Maple Lake we had no prior information to suggest that the wetland was occupied by the Western Painted Turtle, but a survey and a habitat assessment in 2012 suggested that the location was suitable for the species. The observation occurred on the 1 July 2015 (see cover photo) at Maple Lake. Prior to the observation of Western Painted Turtles at Chemainus Lake, we had conducted 3 visits to the lake because there was good habitat, but the 2 Western Painted Turtles observed in the lake were seen during an opportunistic visit by C. Engelstoft on 17 Jun 2015. Interviews with fishers and other visitors had not suggested that the species was present. In 2013 pictures of Western Painted Turtles were sent to us from Summit Lake, but a previous survey in 2013 had failed to verify the presence of the species. During our joint training session with Habitat Acquisition Trust contractor on 28 May 2015, we observed 9 individual Western Painted Turtles.

Most of the known Western Painted Turtle locations are found at lower elevations on the East coast of Vancouver Island, but the new location at Summit Lake occurs at 380 m asl that is 120 m higher than Turnbull Lake that sits at 260 m asl. This suggests that all wetlands below 400 m asl should be considered for surveys in the future.

Even though we visited 43 new wetlands in 2015, there are still many wetlands that could be surveyed for Western Painted Turtle around Nanaimo, Courtenay, Port Alberni and Quadra Island, all areas with known or suspected Western Painted Turtle populations. The anecdotal observations from Duncan also suggest that wetlands in this area should be re-surveyed.

eDNA water sampling and filtering

The 19 locations from which we took water samples were located from Saratoga north of Courtenay to locations in the Victoria area. The wetlands that were sampled in the summer were also sampled in the fall in an attempt to compare detection seasonally, but the relatively late sampling in the fall took place when turtles had moved to their hibernation sites. One sampling location (#3) at Swan Lake was situated within 30 m of a known hibernation place in the lake, determined by following 4 radio tagged turtles in 2010 to 2012 (unpublished data). Unfortunately none of the hibernation areas were known in any of the other ponds and lakes sampled.

Due to scarcity of visual detections during the warm summer, it was only possible to obtain a few water samples within 15 m of observed turtles, but we relied on prior knowledge of where turtles had been spotted in previous surveys. This rule of taking water samples within 15 m of basking turtles, assumes

that this is also the area where the turtle spend time in the water, which might or might not be the case. This highlights the importance of learning more about the movement of turtles during the day and night to better understand how to increase the detection probability of turtle DNA in water samples. As we better understand how turtle DNA disperses in water and because hibernating turtles presumably shed DNA into the water column, it might someday be possible to determine hibernation areas using this eDNA method by sampling potential hibernation areas during the winter or very early spring.

Turtle Habitat Restoration at Diver Lake

Logs were installed to enhance basking opportunities for turtles in Diver Lake. The logs may also be used by many other species as shown by camera monitoring in Swan Lake in Saanich. Snakes, ducks and Great Blue Herons have been observed using logs installed for turtles (Umphrey et al 2012). Because the logs were installed in sight of the fishing dock, visitors have increased chance of observing the turtles and other wildlife.

The turtle basking restoration plan also provides guidelines for parks managers to reduce some threats to the Western Painted Turtle at Diver Lake Park (Appendix 5).

Recommendations

- Conduct visual surveys at these locations:
    - Re-survey wetlands where Western Painted Turtle have not been seen and the habitat assessment resulted in a moderately or highly suitable turtle habitat score including the Illusion Lakes by Spider Lake and wetlands on Denman Island
    - Survey areas with anecdotal observations that have highly or moderately suitable turtle habitat or wetlands with such as the wetlands in the vicinity of the Kinsol Trestle Western Painted Turtle observation
    - Chemainus, Maple, Divers, Morrell and Cathers Lake to estimate population size
    - Survey the many wetlands around Nanaimo to better understand the local distribution of the species
- Habitat enhancement at Divers Lake
    - Monitor use of installed logs
    - Enhance nesting grounds and protect Western Painted Turtle nests
- eDNA sampling
    - Investigate correlations between DNA detection and environmental parameters such as turbidity, pH, temperature, oxygen concentration, nuclease activity, conductivity, UV radiation, temperature, type of sediment and salinity measured when collecting water samples
    - Investigate correlations between DNA detection and time of year by sampling during spring, summer, and fall, and/or by sampling a few locations throughout the year
    - Investigate the effect of wetland type and size (lake, marsh and pond) and sampling effort on DNA detection

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Appendices

Appendix 1. Surveying, sampling, fixing and filtering protocols

Field Sampling Protocol

Field Equipment needed:

Sample collection

- 3 extra-large ziploc bags and 3 medium ziploc bags (both labelled with sampling location)
- 3-1L Nalgene bottle for each filter treatment (0.45 um MCE, 1.00 um MCE, PCTE, GF; maximum 12 bottles)
- 3 sets of gloves for each person
- 18-15ml tubes (or 3-15ml tubes)
- cooler with ice packs
- sampling extension pole

Environmental data collection

- Kestrel Weather Meter
- Spherical Densiometer
- Easy View Light Meter
- YSI probe (DO and Temperature)
- pH Meter
- Conductivity metre
- GPS
- Secchi Disk

Turtle observations

- Camera
- Telescope and binoculars

General

- Field data sheets and clipboard
- Field notebook
- Sharpies
- pencils

Field Protocol:

<u>Do NOT sample in moderate to heavy wind or rain</u>.

Decontamination

- All 1L Nalgene bottles and lids should be rinsed with 50% bleach then rinsed three times with tap water. Wear protective eyewear and gloves when working with bleach.
- Between sites, the sampling pole needs to be sprayed with 5% bleach, and then rinsed at the next site (off to side of sampling spot).
- Between locations the boat, paddles, boots, waders, and other equipment (e.g., secchi disk) need to be cleaned with 5% bleach (for eDNA and chytrid decontamination)

Survey

- Record survey data for location, including date, start and end time, observers, weather (wind (none or light), precipitation (none or light), cloud cover) and date of last rain.

Sampling

- Collect samples at 3 independent (>50m apart) sites at a location (wetland or lake).
- Locate sample sites at or near current or reported turtle sightings or occurrences.

- Get as close as possible to turtle locations by sampling from shore or by boat. Avoid bringing boots or the boat close to the sampling locating to avoid contamination by using the extension sample pole.
- All vials and bottles need to be rinsed at sampling sites three times.
- Gloves should be changed between sampling sites.
- Samples are taken from surface water and are collected first, before any environmental data collection.
- the sample location can then be approached on foot or by canoe to facilitate environmental data collection.

1. Record coordinates (UTM) for each sampling site
2. Collect 6- 15ml (or 1-15ml, depending on location) water samples from each of 3 sampling sites per location
3. Collect 1- 1L water sample from each of 3 sampling sites per location for EACH of the filter treatments for that location.

Water Quality

4. Submerge YSI probe in surface water to determine water temperature (^{o}C) and dissolved O_2 (mg/l) at each sampling site.
5. Record conductivity. If YSI does not have conductivity, use a conductivity metre.
6. Submerge pH meter at each sampling site.
7. Submerge Secchi disk to determine turbidity (m) and depth (m) at each sampling site.

Biophysical Characteristics

8. Use Kestrel Weather Meter to determine average wind speed (km/h) and air temperature (^{o}C) at each sampling site.
9. Use EasyView Digital Light Meter to determine light intensity (lux) at each sampling site.
10. Use spherical densiometer to determine canopy cover (%) at each sampling site.
11. Determine distance from nearest shore and nearest turtle observation (m) for each sampling site.
12. Record Aquatic vegetation (emergent or submergent; leading species) for each sampling site.
13. Record Substrate type (organic/inorganic; fines/muck, detritus/organics, sand, gravel, other) at each sampling site.

Western Painted Turtle eDNA Environmental Variable Datasheet, 31 Oct 2015

Location Name:		
Date:		Observers:
Start Time:	Wind Speed: Calm Light	Precipitation: None Light
End Time:	Cloud Cover (%):	Last Rain:
Location Comments:		
Site 1 Way Point #	UTM: E N	Sampling Time:
Air Temp (°C):	Water Temp (°C,YSI):	Dissolved O_2(mg/L):
Avg Wind Speed (km/h):	Conductivity:	pH:
Light Intensity (Klux):	Depth (m, Secchi):	Turbidity (m, Secchi):
Distance from Turtle Obs (m):	Substrate Type:	
Distance from Shoreline (m):	Aquatic Veg Species:	
Site Comment:		
Site 2 Way Point #	UTM: E N	Sampling Time:
Air Temp (°C):	Water Temp (°C,YSI):	Dissolved O_2(mg/L):
Avg Wind Speed (km/h):	Conductivity:	pH:
Light Intensity (Klux):	Depth (m, Secchi):	Turbidity (m, Secchi):
Distance from Turtle Obs (m):	Substrate Type:	
Distance from Shoreline (m):	Aquatic Veg Species:	
Site Comment:		
Site 3 Way Point #	UTM: E N	Sampling Time:
Air Temp (°C):	Water Temp (°C,YSI):	Dissolved O_2(mg/L):
Avg Wind Speed (km/h):	Conductivity:	pH:
Light Intensity (Klux):	Depth (m, Secchi):	Turbidity (m, Secchi):
Distance from Turtle Obs (m):	Substrate Type:	
Distance from Shoreline (m):	Aquatic Veg Species:	
Site Comment:		

Laboratory Protocol

Decontamination

- Wipe down the work area and outer surfaces of all equipment to be used with 50% bleach prior to sample processing, and between locations.
- Wear powder-free nitrile gloves for all processing, and change gloves between sites.

Ficetola Standard Protocol

1. Transfer 15ml of water sample into a 50ml vial
2. Add 1.5ml 3M Sodium Acetate. *Do not touch the inside of the 50m vial with the pipette tip OR change pipette tips between each 50ml vial*.
3. Fill the 50 ml vial with 100% (or 95%) molecular grade ethanol.
4. Shake.
5. Label Ficetola samples: species (Pt for painted turtle); 3 letter code for site (e.g., DIV for Divers Lake); site number (01 to 03); sample letter (a to f); and date of sample collection (day, month, year).
   E.g., Pt DIV 01 a 25Oct15
6. Store in Fridge/Freezer

Vacuum Filtration

- Set-up peristaltic pump, filter flask with stopper and filter funnel adapter, and decontaminate all surfaces with 50% bleach.
- Decontaminate filter tweezers in 50% bleach between sites and rinse in distilled water
- Use one disposable plastic filter funnel for each site plus the blank (4 funnels per location)
- One disposable plastic filter funnel can be used to filter all 1-L water samples collected at one site in a location through different filter treatments (i.e., differ substrates and pore sizes). Not all locations will have all filter treatments applied.
- Replace the absorbent pad in the filter funnel with a sterile one if it becomes soiled or clogged.
- When all 1-L water samples for all 3 sites plus the blank have been filtered for a location, decontaminate the pump tubing, filter flask and funnel adapter with 50% bleach followed by a thorough distilled water (or tap water) rinse.

Filter Treatment	Treatment #
MCE 0.45 um	01
MCE 1.00 um	02
PCTE 10 um	03
GF 1.5 um	04

MCE Filter Treatments 01 (0.45 um) and 02 (1.0 um)

1. Remove contamination-free 250ml plastic filter funnel with 0.45 um MCE filters from plastic pack and install on filter funnel adapter, taking care not to remove the cap or touch the inside of the funnel
2. Pour 250ml of 1-L water sample for a site into filter funnel, and turn on pump.
3. Refill cup (before filter dries out) until 1-L sample is completed; this may take up to an hour for a 1-L sample.
4. If the filter becomes clogged before 1-L is completed (i.e., no more water appears in the clear plastic adapter below the filter funnel), turn off the pump and replace the filter with another 0.45 um filter (remove the blue paper disc between filters).

5. Continue filtering and changing filters until the sample is completed (keep the number of filters low, only changing if filtration completely stops; try not to use more than 10 filters).
6. To remove a filter, use filter tweezers to carefully lift the filter off the absorbent pad, and fold in half and half again until the filter is in a small cone.
7. Place the filter cone in a contamination-free 2ml cryogenic vial (2 filters max per vial), and fill to top with 95-100% molecular grade ethanol. Be careful not to touch the inside of the cryovial with the ethanol pipette.
8. Label cryovial with ethanol-resistant pen: 3 letter code for site, site number (01 to 03), filter treatment (01 to 04 by substrate and pore size), filter letter (A to J for number of filters per 1-L sample) and date of sample collection (day month year).

E.g., DIV 01-02-A 25Oct15

Divers Lake, site 1, filter treatment 2 (MCE 1.0 um), filter A

9. When the 1-L sample for the 0.45 um MCE treatment is completed, repeat for next 1-L filter treatment sample for that site (e.g., 1.0 um MCE), using the same plastic filter funnel.

PCTE Filter Treatment 03 (10 um)

1. Proceed as above, replacing the MCE filters with PCTE filters.
2. Place the filter membrane in a contamination-free 2ml cryogenic vial (2 filters max per vial), and add 700 ul of CTAB extraction buffer using the 1ml pipette and contamination free pipette tip. *Do not touch the inside of the cryovial with the pipette tip OR change pipette tips between each*.
3. Label as per MCE protocol, using filter treatment 03.

GF Filter Treatment 04 (1.5 um)

1. Proceed as above, replacing the MCE filters with GF (glass) filters.
2. Place the filter membrane in a contamination-free 2m cryogenic vial (2 filters max per vial) and freeze immediately.
3. Label as per MCE protocol, using filter treatment 04.

Blank (using 0.45 um MCE filter)

1. After all samples for all 3 sites have been completed for a location, filter a minimum of 250m of contamination-free water through a new 250ml plastic filter funnel with the 0.45 um filter in place.
2. Record the volume filtered and the type of water used. Preferentially use nuclease free, sterile or distilled water if available. Municipal tap water can be used if necessary.

Filter Storage

1. Place cryovials containing filters for one 1-L sample together in a small baggie, and write the date, 3-letter location code, site number and filter treatment on the outside in ethanol resistant marker.
2. Place the baggies for all 1-L samples collected at a site and filtered using the same treatment (filter substrate and pore size; 01 to 04) in one baggie and label the outside with the date, 3-letter location code, site number, and filter treatment.
3. Place the baggies for all samples and filter treatments collected at a site together in one medium size site baggie and again label the outside with the date, 3-letter location code, and site number.
4. Place the 3 site baggies together in one large location baggie. Label the outside with the date and 3-letter location code.
5. Place all location bags containing all cryovials with all filter treatments in a -20°C freezer.

Appendix 2. Summary of water sampling sites and dates, sample preparation procedures, and eDNA lab priorities

Lab Priority	Autumn 2015 Sample Dates	Spring 2015 Sample Dates	Autumn 2014 Sample	Region	Location	# Sites	6 Ficetola samples	1 Ficetola sample	MCE (0.45, 1.0 uM)	MCE 0.45 uM only	PCTE 10.0uM	PCTE 5.0uM	WPT max # observed	RES max # observed	Unk Turtle	Elev. (m asl)	Perimeter (km)	Population Size / density
Priority 1: 2 seasons, all ppt, all filter types																		
	29-Oct	23-Jun		Port Alberni	Airport Wetlands	3	Jun & Oct		Jun & Oct		Oct		37	1		83	3.2	L/H
	29-Oct	23-Jun		Port Alberni	Summit Lake	3	Jun & Oct		Jun & Oct		Oct		9			382	1.16	M/H
	29-Oct	24-Jun		Port Alberni	Sumner Lake	3	Jun & Oct		Jun & Oct		Oct		9		1	79	2.3	L/H
	29-Oct	24-Jun		Port Alberni	Ash main 26K	2	Jun & Oct		Jun & Oct		Oct		4			116	0.87	S/M
Priority 2: 2 seasons, all ppt, all MCE filters																		
	20-Oct	10-Jun	Sep	Nanaimo	Buttertubs Marsh	3	Jun & Oct		Jun & Oct				3	10		57	2.1	L/H
	15-Oct	03-Jun	Sep	Nanaimo	Divers Lake	3	Jun & Oct		Jun & Oct				13			105	1.7	M/H
	22-Oct	02-Jul		Cowichan/ Chemainus	Chemainus Lake	3	Jun & Oct		Jun & Oct				2			115	1.4	M/M
Priority 3: 1 season, all ppt, all MCE filters																		
	21-Oct			Comox Valley	Maple Lake	3	Oct		Oct				1			136	2.6	L/L
Priority 4: 1 season, 1 ppt, all filter types																		
	04-Nov		Sep	Capital Regional District	Beaver Lake	3		Nov	Nov		Nov	Nov	20	13		59	3.61	L/H
	04-Nov		Sep	Capital Regional District	Langford Lake	3		Nov	Nov		Nov	Nov	37	9		64	5.03	L/H
	05-Nov		Sep	Capital Regional District	Swan Lake	3		Nov	Nov		Nov	Nov	11	4		14	1.2	M/H
Priority 5: 1 season, 1 ppt, 1 MCE filter																		
	27-Oct			Nanaimo	Cathers Lake	3		Oct		Oct			1	10		99	1.4	M/H
	23-Oct			Nanaimo	Morrell Lake	3		Oct		Oct			1	1		175	1.3	M/L
Priority 6: RES ONLY																		
	21-Oct			Comox Valley	Saratoga Golf Course	2	Oct		Oct					1		5	0.1	S/L
	29-Oct			Port Alberni	Loon Lake	3		Oct	Oct					3		365	1.2	M/M
	05-Nov			Capital Regional District	Beckwith Park	3		Nov		Nov				15		62	0.2	S/H
	23-Oct			Nanaimo	Lost Lake	3		Oct		Oct				1		165	0.6	S/L
	27-Oct			Nanaimo	Sywash Pond	3		Oct		Oct					1	99	2.9	L/L
Priority 7: 1 season, all ppt, MCE filter clogged																		
	22-Oct			Cowichan/ Chemainus	Private Pond	2	Oct		Oct					20		62	0.8	S/H

Appendix 3. Western Painted Turtle habitat assessment on Vancouver Island, 2015

Site ID	Location	Region	Date assessed	WPT found	Elev. (m)	Wetland perimeter (km)	Emergent veg. (% perim.)	Floating veg. (% perim)	Forest (% perim.)	Shrubs (% perim.)	Grass/ herbs (% perim.)	Developed (% perim.)	Assess basking opportunities	Assess nesting opportunities	Assess foraging opportunities	Rate overall habitat quality
E35	Village Bay Lake	Quadra Isl.	09-Jun	N			7	5	100	80		50	Moderate	Moderate	Low	Moderate
E36	Main Lake	Quadra Isl.	04-Jun	N			10	5	100	40			Moderate	Moderate	Low	Moderate
E37	Hamilton's Marsh	Coombs	04-Jun	N			100		5	100			Low	Low	Moderate	Moderate
E38	Rees Lake	Quadra Isl.	04-Jun	N			90	10	5	90			Low	Low	Low	Low
E39	Stramberg Lake	Quadra Isl.	05-Jun	N			90	15	100	90			High	Low	Moderate	Moderate
E40	Mesachie Lake	Cowichan Valley	09-Jun	N				5	70	80		40	Low	Moderate	Low	Moderate
E41	Wild Play Pond	Cowichan Valley	09-Jun	N	171	0.12		95	40	60		50	Low	Moderate	Low	Moderate
E42	Imadene Pond	Cowichan Valley	09-Jun	N			50	70	45	100			Low	Moderate	Low	Moderate
E43	Bear Lake	Cowichan Valley	09-Jun	N				60	5	90		10	Moderate	Moderate	Moderate	Moderate

Site ID	Location	Region	Date assessed	WPT found	Elev. (m)	Wetland perimeter (km)	Emergent veg. (% perim.)	Floating veg. (% perim)	Forest (% perim.)	Shrubs (% perim.)	Grass/ herbs (% perim.)	Developed (% perim.)	Assess basking opportunities	Assess nesting opportunities	Assess foraging opportunities	Rate overall habitat quality
E44	Beaver Lake, Cowichan	Cowichan Valley	09-Jun	N				95	80	75		20	High	High	High	High
E45	Hudson's Lake	Cowichan Valley	09-Jun	N			80	10	80	100			High	Low		High
E45	Sywash Pond	Nanaimo	10-Jun	N	99	2.9	60	100	60	100			Low	High	High	High
E46a	Lazo Nature Park	Comox Valley	01-Jul	N			100	100	60	100			Low	Low	Low	Low
E46b	Lazo Nature Park	Comox Valley	01-Jul	N					70	30			Low	Moderate	Moderate	Moderate
E46c	Lazo Nature Park	Comox Valley	01-Jul	N				10	60	10	100		Low	Moderate	Low	Low
E47	Black Lake	Comox Valley	01-Jul	N				60	10	100			Low	Moderate	Moderate	Moderate
E48a	Courtenay Fish and Game	Comox Valley	02-Jul	N			30		50	10		30	Moderate	Moderate	Low	Moderate
E48b	Courtenay Fish and Game	Comox Valley	02-Jul	N			60		70	30		20	High	Moderate	Low	Moderate
E48c	Courtenay Fish and Game	Comox Valley	02-Jul	N			100		90	100		40	High	Moderate	moderate	High
E49	Anderson Lake	Comox Valley	30-Jun	N			80	10	80	100			Moderate	Low	Low	Moderate

Site ID	Location	Region	Date assessed	WPT found	Elev. (m)	Wetland perimeter (km)	Emergent veg. (% perim.)	Floating veg. (% perim)	Forest (% perim.)	Shrubs (% perim.)	Grass/ herbs (% perim.)	Developed (% perim.)	Assess basking opportunities	Assess nesting opportunities	Assess foraging opportunities	Rate overall habitat quality
E50	Meldas Marsh	Comox Valley	30-Jun	N			100	100	30	100			Low	Low	Low	Low
E51a	Saratoga Golf Course	Comox Valley	30-Jun	N	5	0.1		20	40	60		60	Low	High	Low	Moderate
E51b	Saratoga Golf Course	Comox Valley	30-Jun	N	5	0.1		100		100		98	Low	High	Low	Moderate
E52	6930 Railway Ave.	Merville	29-Aug	N	71	1.62	95	90	90	80	2	5	good	moderate	good	High
E53	4705 Gail Creek	Merville	29-Aug	N			90	80	0	10	90		many		good	High
E54A	Little River Nature Park, Pond W	Comox	29-Aug	N	5	0.23	60	80	0	5	100	5	good	good	good	High
E54B	Little River Nature Park, Pond E	Comox	29-Aug	N	5	0.24	50	30	0	10	90	0	good	good	good	High
E56	Spur Rd	Merville	29-Aug	N	71	0.34	90	90	10	80	5	10	good	moderate	good	High
E57	Hagen Wetland	Merville	29-Aug	N			100	95	80			10	good	moderate	good	High
E59	Chickadee Lake	Denman Isl	23-Sep	N	51	2.7	95	85	100	20	0	0	good	poor	good	High
E60	Inner Isl Nature Reserve	Denman Isl	23-Sep	N	65	3.7	100	20	100	20	10	0	good	poor	good	High
E61	Lacon/Duster Rd	Denman Isl	23-Sep	N	8	1.2	100	0	100	10	0	0	poor	poor	good	High
E62	Graham Lake	Denman Isl	23-Sep	N	34	2.4	60	60	95	5	0	5	good		good	High
E63	Central Park Wetland	Denman Isl	23-Sep	N	60	1.9	100	0	100	0	0	0	poor	poor	poor	Poor

Site ID	Location	Region	Date assessed	WPT found	Elev. (m)	Wetland perimeter (km)	Emergent veg. (% perim.)	Floating veg. (% perim)	Forest (% perim.)	Shrubs (% perim.)	Grass/ herbs (% perim.)	Developed (% perim.)	Assess basking opportunities	Assess nesting opportunities	Assess foraging opportunities	Rate overall habitat quality
E64	Richard Lake	Nanaimo	22-Sep	N	21	1.8			20	20	20	10	poor	good	good	High
E65	Morrison Marsh	Denman Isl	23-Sep	N	14	4.6	70	70	98	10	2	2	good	moderate	good	High
E66	McFarlane Wetland	Denman Isl	23-Sep	N	15	2.9	100	80	80	30	0	1	good	moderate	good	High
E67a	E Kingburn Pond E (K1)	Cobble Hill	10-Sep	N	182	1.7	5	60	90	10	0	0	good	moderate	good	High
E67b	E Kingburn Pond E (K2)	Cobble Hill	10-Sep	N	182	0.9	5	50	50	100	0	0	good	moderate	good	High
E68	3681 Kingburn Dr (K17)	Cobble Hill	10-Sep	N	81	0.3	0	10	50	100	0	0	low	moderate	moderate	Moderate
E69	Logging road parallel to Renfrew Rd (K7)	Cobble Hill	10-Sep	N	150	0.8	80	100	60	70	0	0	low	poor	good	Moderate
E70	Logging road parallel to Renfrew Rd (K6)	Cobble Hill	10-Sep	N	154	0.8			Wetland Dry				low	poor	good	

[1] assessment with 10 m of shoreline

Appendix 4. Western Painted Turtle threat assessment on Vancouver Island, 2014

WPT = Western Painted Turtle

Site ID	Location	Date assessed	Roads - paved	Roads - unpaved	Recreation (motorized)	Recreation (non-motorized)	Pets	Exotic species	Housing/ industrial dev.	Urban activities	Forestry	Grazing	Agriculture	Water use/control	Other	Comments about threats
E71	Sywash Pond	10-Jun	L				L	Y		L					Hydro line	Bullfrogs
E37	Hamilton's Marsh	04-Jun					L	Y								Bullfrogs
E38	Rees Pond	04-Jun		H							M					
E40	Mesachie Lake	09-Jun	L	L		L				L						
E41	Wild Play Pond	09-Jun		L												
E42	Imadene Pond	09-Jun		L						L						
E43	Bear Lake	09-Jun		L	L	L										
E44	Beaver Lake	09-Jun		M	L	M								L	Great Location	Use water to fill salmon streams
E45	Hudson's Lake	09-Jun	L								M					
E35	Village Bay Lake	04-Jun		L	M	M			L	L				M		Dam, Water consumption
E36	Main Lake	04-Jun		L		M										
E39	Stramberg Lake	05-Jun														
E46a	Lazo Nature Park (a)	01-Jul		L												
E46b	Lazo Nature Park (b)	01-Jul														
E46c	Lazo Nature Park (c)	01-Jul														
E47	Black Lake	01-Jul						Y							Bullfrogs	
E48a	Courtenay Fish and Game Club (a)	02-Jul		M						M						
E48b	Courtenay Fish and Game Club (b)	02-Jul		M			L									

Site ID	Location	Date assessed	Roads - paved	Roads - unpaved	Recreation (motorized)	Recreation (non-motorized)	Pets	Exotic species	Housing/ industrial dev.	Urban activities	Forestry	Grazing	Agriculture	Water use/control	Other	Comments about threats
E48c	Courtenay Fish and Game Club (c)	02-Jul	H					Y							Bullfrogs	
E49	Anderson Lake	30-Jun		L							H					
E50	Meldas Marsh	30-Jun		L												Water Level Low
E51a	Saratoga Golf Course (a)	30-Jun	L							H						
E51b	Saratoga Golf Course (b)	30-Jun								H						
E59	Chickadee Lake	23-Sep				L					L			L		
E60	Inner Isl ture Reserve	23-Sep		L							L					
E61	Lacon/Duster Rd	23-Sep			L						L					
E62	Graham Lake	23-Sep			L											drinking water
E63	Central Park Wetland	23-Sep	M						L		L					
E65	Morrison Marsh	23-Sep	L						L		L					
E66	McFarlane Wetland	23-Sep	L													
E64	Richard Lake	22-Sep	M	M					L			L				Bullfrog
E67a	E Kingburn Pond	10-Sep		L	L		L				L			L		Beaver?
E67b	W Kingburn Pond	10-Sep		L	L						M			L		
E68	3681 Kingburn Dr	10-Sep	M	L			L		L				M	M		
E69	Logging road parallel to Renfrew Rd (K7)	10-Sep		L	L						L					Bullfrogs?
E70	Logging road parallel to Renfrew Rd (K6)	10-Sep														dried up on day of assessment

Site ID	Location	Date assessed	Roads - paved	Roads - unpaved	Recreation (motorized)	Recreation (non-motorized)	Pets	Exotic species	Housing/ industrial dev.	Urban activities	Forestry	Grazing	Agriculture	Water use/control	Other	Comments about threats
E52	6930 Railway Ave.	29-Aug				L	L	H	L							Bullfrogs
E54a	Little River Nature Park, Pond W	29-Aug	L				M	M	L							
E54b	Little River Nature Park, Pond E	29-Aug	L				M	M	L							
E56	Spur Rd	30-Aug	M				L	Y	L							bullfrogs
E57	Hagen Wetland	30-Aug	L	L			L		L	L	L			L		raining when assessed
E53	4705 Gail Creek	29-Aug					L									

H- high, M – medium, L – low

Y – yes there are exotic species

Western Painted Turtle

Habitat Restoration and Management Guidelines at Diver Lake, Nanaimo

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Synopsis of Restoration and Management Guidelines

A) Habitat Restoration or creation

1. Assessment of habitat use and condition

- Monitor basking turtles in ponds
- Keep track of turtle observations
- Identify and monitor nesting sites
- Monitor water quality

2. Basking log installations

- Source logs and organize pickup or delivery of appropriately sized logs. The diameter should be greater than 25 cm.
- Cut the notches for the rope attachment
- Collect and prepare anchoring ropes (or chains)
- Identify a site to launch the logs and drag them to the chosen sites in the lake
- Install the logs

3. Restore or create turtle nesting sites

- Identify exact locations of 4 restoration sites on south facing mounds
- Remove grasses in the 4 one meter square nesting site previously used by the Western Painted Turtle.
- Maintain open soil patches and prevent weeds from taking over. It might be possible to plant native non-invasive plants that will leave areas with exposed soil and create minimal shading when invasive species have been removed from the area. The goal is that the digging of the nesting turtles will in the long run create a dynamic between digging and vegetation growth that will ensure bare soil patches for the turtles.
- Fence the area with a cedar-log stack fence, so turtles can access the area but it prevents visitors to inadvertently entering the sites.
- Install signs to inform visitors of our efforts.

- Install a time-lapse monitoring camera pole for monitoring turtle use.

4. Maintenance of restoration activities

- Annually weeding or tilling of the turtle nesting areas will control invasive grasses and will ensure bare patch of soil for female turtles to nest in. This should take place in the last two weeks of May
- Nurturing native plants that might be planted on or around nesting grounds
- Inspecting basking logs for shore line vegetation overgrowth, problems with anchoring and possibly repositioning of the logs

5. Monitoring

- Conduct one to two turtles surveys a month in Mar, Apr and May
- Monitor nesting grounds using time-lapse camera (10 min intervals) during nesting season (May, June, July)
- Protect known nests with a 2 x 2 inch stucco mesh that are located outside the nesting areas
- Monitor emergence of hatchings on nesting grounds weekly (mid-April to May), by looking for holes where the turtles escaped

B) Management Guidelines

1. ***General Guidelines:***

- Check for Western Painted Turtle occurrences or nesting grounds before undertaking projects that can potentially harm turtles

2. Maintenance of Trails, roads and parking lots maintenance

- Where turtles are known to nest along trials, roads and parking lots and restored nesting sites, conduct maintenance to trials before spring emergence of hatchlings after the turtles hatchlings

have emerged by mid May (this is weather dependent) and the adult females are nesting in June (also weather dependent). The period between emerging hatching and egg laying females is time where there is the least chance to do harm.

- In other areas where trail are immediately adjacent to the lake, either take above precautions *or* survey for signs of turtle nesting activity on trail before starting work.

Table of Contents

Background

The Pacific Coast Populations of the Western Painted Turtle (*Chrysemys picta*) are *Red* listed provincially and listed as *Endangered* federally by COSEWIC (2006). These populations occur in the Lower Mainland, on the Sunshine Coast, and on Vancouver and the Gulf Islands.

The Western Painted Turtle is known from the Nanaimo area, but was first reported from Diver Lake on 20 June 2012 (pers. com. H. Kimura, 2015).

The habitat assessment conducted in 2014, indicated that basking opportunities were poor because there were very few basking logs and only Cattail (*Typha* sp) rhizome mats or floating vegetation were available for basking at the site.

Nesting is known from two areas. In 2015 nesting attempts found under the canopy along the trail that runs from the parking lot to the fishing dock, and in a more suitable nesting habitat located on the filed with the bike tracks. Turtles have been known to nest on this field in the previous years as well. The threats to the Western Painted Turtle at Diver Lake include bait on fishing lures that occasionally catch adult turtles, the threats to juvenile turtles from Bullfrogs (*Lithobates catesbeianus*), and disturbance of nesting area. Watermilfoil (possibly Eurasian Watermilfoil, *Myriophyllum spicatum*) is abundant, but is not considered a threat to the turtles.

Western Painted Turtle Biology

Habitat Requirements

Western Painted Turtles spend most of their lives in ponds, lakes, oxbows, or quiet waters of rivers, where they prefer shallow, sheltered areas with abundant aquatic vegetation. The diet of northern populations of Western Painted Turtle is more carnivorous compared to that of southerly populations (COSEWIC 2006). Young turtles feed mostly on small pond invertebrates such as aquatic insects, crayfish, snails and tadpoles. As they mature they switch to larger prey such as small animals, frogs, and crayfish, as well as consuming aquatic vegetation. They are also known to scavenge when opportunities arise. On sunny days, they can be seen basking on logs or other objects in the water or shorelines. Turtles need to bask to raise their body temperature, which in turn promotes digestion, synthesis of vitamin D, and other vital processes and basking helps remove ectoparasites and/or algae on the carapace.

Turtles can move across the landscape to reach nesting, over-wintering, or foraging sites. While longer movements probably take place along water courses, turtles also regularly move overland, for example when moving to their nesting grounds and between water bodies (Figure 5). We demonstrated this at Swan Lake, Victoria, using radio tags (Engelstoft and Ovaska 2011, Ovaska

and Engelstoft 2012), but it is unknown if Western Painted Turtles at Diver Lake undertake any of these seasonal movements. Movement patterns at a landscape level of Western Painted Turtles inhabiting coastal areas are virtually unknown, and we must rely on information from other areas. Movements of 10 species of freshwater turtles in the United States suggest that riparian buffer zone width of 150 m is necessary to protect terrestrial habitat around water bodies (Bodie 2001), and Semlitsch (2003) found that turtles need at least 300 m of terrestrial buffer. On the Sunshine Coast turtles moved among lakes throughout the summer (M. Evelyn, 2009 per com). Not only is the aquatic habitat important to turtles, so is the terrestrial habitat along the riparian zones, nesting grounds, and migration routes between foraging, breeding, and hibernation sites. Nothing is known about these longer dispersal movements around Diver Lake.

Nesting sites are generally located close to the ponds where the turtles live. All known nesting sites on Vancouver Island occur within 150 m from water bodies (Engelstoft and Ovaska, unpublished data), which concur with other areas in Canada (COSEWIC 2006).

Figure 5. Areas of observed nesting attempts of Western Painted Turtle at Diver Lake, 2015

Life History

Individual Western Painted Turtles can live 50 years or more. While mortality of hatchlings and young turtles is often high, adults have few natural predators. Great Blue Herons, ravens, crows, raccoons, otters, and rats prey on turtle eggs and hatchlings and otters are known to prey on adult hibernating turtles. Bullfrogs are also known to eat Western Painted Turtle hatchlings (Jancowski and Orchard 2013).

The following links provide more information about the Western Painted Turtle:

- Habitat Acquisition Trust Western Painted Turtle project
- COSEWIC Western Painted Turtle Status Report
- CDC Species Explorer
- Wildlife in British Columbia At Risk, Painted Turtle brochure

Because turtle populations depend on high female survival for persistence, one of the main threats to adult Western Painted Turtle in urban areas is road mortality of turtles during their seasonal travels or while dispersing (Tingley and Herman. 2008). It was shown in a computer model that annual kills of 3 adult females in a population of 100 individuals will cause the populations to go extinct in 50 years. Road mortality of females traveling to the nesting ground is a widespread phenomenon, and can result in serious population declines. In the United States, high road densities have been shown to be associated with male-biased sex ratios of Painted Turtle populations, probably as a result of mortality of females on nesting migrations (Steen and Gibbs 2004). There are no reports of Western Painted Turtle road kills at Diver Lake.

Suitable nesting sites are often limited along the Pacific Coast, but suitable turtles nesting habitat needs to have the following features: southern aspect ensuring good exposure to the sun; light clay, loamy and sandy soils, usually hard-packed; bare ground or sparse vegetation at well-drained sites; proximity to water, usually within about 200 m. Turtles are also attracted to nest along the sides of trails and roads and bare soil patches in gardens.

Western Painted Turtles often nest at well established communally nesting-sites that are used year after year. The female digs a flask-shaped nest in the substrate and can lay more than 18 eggs in early summer. The eggs hatch in the fall, but hatchling turtles usually overwinter in the nest and dig themselves out in the spring, then head to water. Males mature in about 8-10 years, females in 12-15 years.

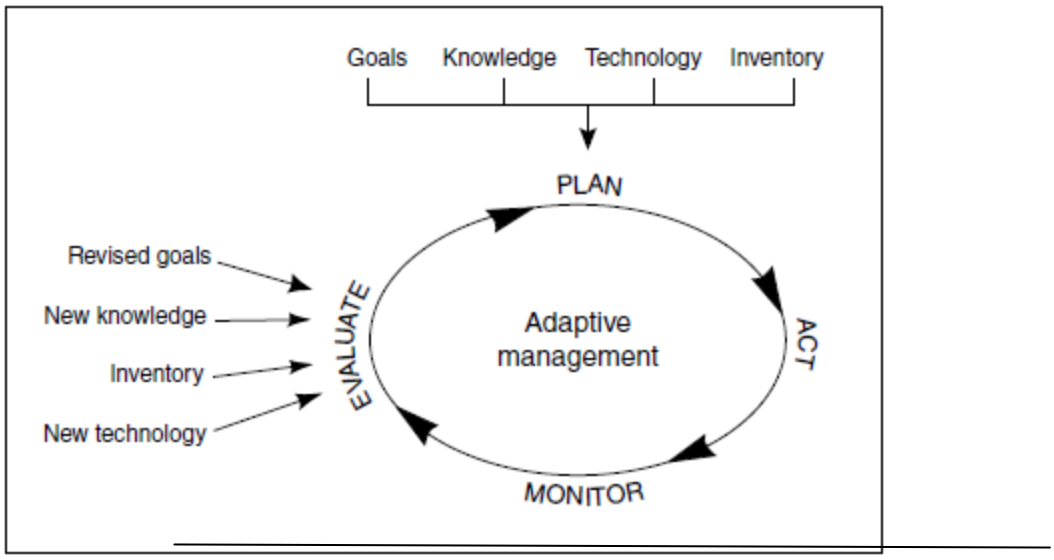
Knowledge Gaps and Threats to Western Painted Turtles

- Poor understanding of how the Western Painted Turtle use the area, such as seasonal movements to and from nesting, mating, feeding and hibernation sites
- Limited number of nesting grounds that compel turtles to use edges of roads and trails, to seek nesting sites in surrounding areas, and to cross the highway and other roads, activities that increase risk of mortality and disturbance
- Potential predation by Bullfrogs, Great Blue Herons, Raccoons, cats, and dogs, and rabbits on nesting sites can inadvertently dig up and destroy nests
- Number of turtles per year caught by fishermen and the impact injury

Proposed Habitat Enhancement

Basking opportunities and nesting grounds are two important Western Painted Turtle habitat requirements that are only partially met in the area around Diver Lake. Below is a list of suggestions that seek to improve the Western Painted Turtle habitat. Due to the limited understanding of how Western Painted Turtles use the area even the best made plans might not result in the desired outcome due to unforeseen complications. Because we cannot wait with attempting to restore turtle habitat until we know it all, we suggest that restoration efforts adopt an Adaptive Management approach (Starkey et al 2005). Briefly this means that project design, on-the-ground work, monitoring and evaluation of results are linked in a feedback loop (Figure 6).

Figure 6. Adaptive management cycle



(Source: Figure 2 in Starkey et. al 2005)

1. Assessment of habitat use and condition

Rationale:

Currently we know that the Western Painted Turtle is in the area. Knowing more about how the species uses the area is important

What?

- Monitor basking turtles in ponds
- Keep track of turtle observations
- Identify and monitor nesting sites
- Monitor water quality

When?

- Any time

Where?

- Throughout the property

2. Basking log installation

Rationale:

It has already been established that basking sites were limited to floating vegetation mats and flattened shoreline vegetation. Basking opportunities can be improved by installing basking logs in the pond.

Our experience from other sites shows that the installation of real logs with a diameter of more than 25 cm and 4 to 6 meters long is the best option. Stabilizing boards by installing them in pairs has shown to work well in Swan Lake because it will prevent the logs from rolling with the waves and when turtles climb up to bask. Logs should be installed along the shoreline throughout the lake, preferably in sheltered sites. Some of the logs should be installed perpendicular to the shore and others along the shore. We have seen at other sites that the shore vegetation overgrow the logs and sink them if there is quick vegetation growth, so they should be anchored in such a manner that they can be moved if that happens. The logs should be tied with UV resistant

polypropylene ropes (or chains depending on budget) to either the shore line vegetation or trees, and/or anchored with concrete blocks.

Basking logs will also benefit other species, such as waterfowl, snakes, and insects, as well as providing better wildlife viewing possibilities for visitors.

What?

- Source logs and organize pickup or delivery of appropriately sized logs. The diameter should be greater than 25 cm.
- Cut the notches for the rope attachment
- Collect and prepare anchoring ropes (or chains)
- Identify a site to launch the logs and drag them to the chosen sites in the lake
- Install the logs

When?

- Any time in 2015 or before the turtles emerge from hibernation in 2016

Where?

- Along the shore line throughout the lake with some in sheltered locations.

3. Restore or create turtle nesting sites

Rationale:

Nesting attempts have been observed in several areas on the south side of the lake (Figure 5), but the site under the canopy on the path to the fishing dock is likely not a productive site. The other site is located on a field with mountain bike tracks (Figure 7). Based on our experimentation and testing at Beaver Lake of clearing several smaller areas to create more edge habitat where the turtles like to dig their nests, we suggest that 4 plots of about 1 m^2 each are cleared for vegetation on the south side of the mounds created for the bike track Figure 7. It might be advisable to protect the enhanced areas with a fence the turtles can pass under to prevent dogs and people from destroying nests. We also suggest a sign be posted to inform the public of the purpose of the cleared sites.

Figure 7. Approximate location of Western Painted Turtle nesting site enhancement at Diver Lake

What?

- Identify exact locations of 4 restoration sites on south facing mounds

- Remove grasses in the 4 one meter square nesting site previously used by the Western Painted Turtle.
- Maintain open soil patches and prevent weeds from taking over. It might be possible to plant native non-invasive plants that will leave areas with exposed soil and create minimal shading when invasive species have been removed from the area. The goal is that the digging of the nesting turtles will in the long run create a dynamic between digging and vegetation growth that will ensure bare soil patches for the turtles.
- Fence the area with a cedar-log stack fence, so turtles can access the area but it prevents visitors to inadvertently entering the sites.
- Install signs to inform visitors of our efforts.
- Install a time-lapse monitoring camera pole for monitoring turtle use.

When?

- Creation of nesting sites in unused area can be created at any time,
- Enhancing used sites should take place from mid May after hatchlings have emerged and the females are starting to lay eggs in early June (see next section for detail about timing).

Where?

- At suitable sites on the back side of the mounds of the mountain bike tracks

4. Maintenance of restoration activities

Rationale:

Restoration is our way of trying to repair or enhance habitat. For example historically, the dynamic between the digging turtles and vegetation cover was such that the disturbance of the turtles ensured the next year's nesting ground. In a system with aggressive early succession introduced species it is necessary to control these species.

Finding the best time for nesting area maintenance can be tricky because the timing of hatchling emergence and females laying eggs is weather depended and may vary from year to year. Based on our surveys in the Victoria area, most of turtle hatchlings generally emerge in April (Figure 8), but in years with mild winters and spring they can start emerging in early March (our earliest record is March 3) and in colder years they will emerge later. Adult females generally lay her eggs in June (Figure 8), but that can be earlier if the spring is mild or later of it is cold. Generally maintenance of the nesting areas should take place in the time between mid- May to end of May when hatchlings are emerged and females have not yet started laying their eggs.

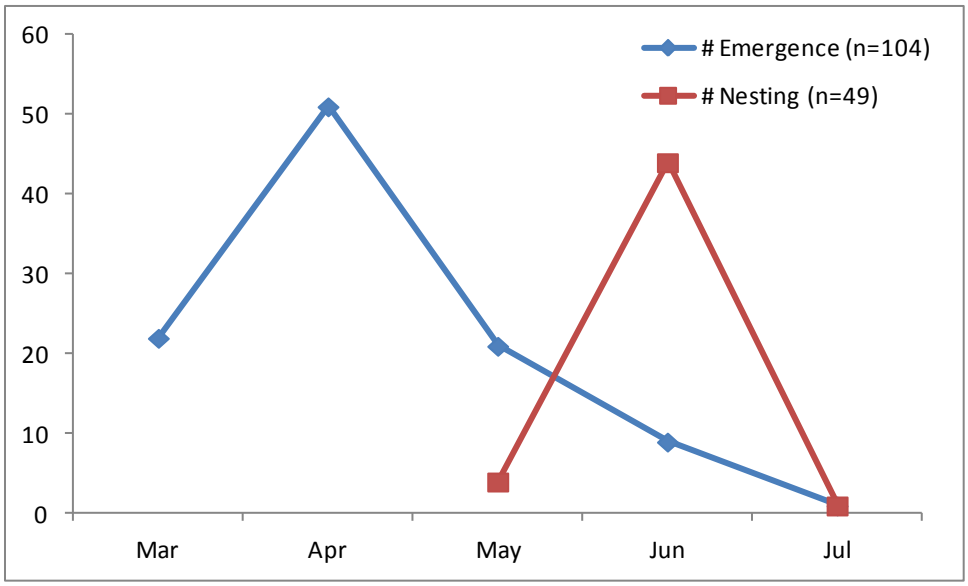


Figure 8 Timing of Western Painted Turtle hatchling emergence and adult female egg laying on Vancouver Island from 2008 to 2015.

Basking logs might rip loose during winter storms or be overgrown with encroaching shore vegetation. As a consequence it is important to inspect installed basking logs to ensure that they function as they were intended.

What?

- Annually weeding or tilling of the turtle nesting areas will control invasive grasses and will ensure bare patch of soil for female turtles to nest in. This should take place in the last two weeks of May
- Nurturing native plants that might be planted on or around nesting grounds
- Inspecting basking logs for shore line vegetation overgrowth, problems with anchoring and possibly repositioning of the logs

When?

- Most of the maintenance is ongoing because it is important to keep an eye on the various aspect of turtle habitat restoration initiatives

- Basking logs should be inspected in February to make sure they are in good order when the turtles come out of hibernation
- Nesting sites should be ready for the nesting season starting in June

Where?

- Where restoration took place

6. Monitoring

Rationale:

Planning and restoring turtle habitat is exciting and possible, but is only the first steps towards ensuring the persistence of the Western Painted Turtle population on the property. The less exciting but equally important aspect of restoration is the monitoring and evaluation of the effect of the on the ground work. These two steps document and ensure that the restoration objectives are met and provide important information for the Adaptive Management feedback loop (Figure 6).

To determine the long term effect on the turtle population in it is necessary to establish turtles counts in Mar, Apr and May, when the turtles are easiest to see (Engelstoft and Ovaska 2008). Turtle species should be identified, and their size categorized as hatchling (<5 cm), medium (5-15 cm), large (> 15 cm). The basking log use can be noted at the same time.

The use of the nesting grounds could be monitored using time-lapse cameras during the nesting season (mid-May to Mid July, at 10 to 15 min intervals), and hatchling emergence should be monitored (Mar, April and May). Known nests outside designated nesting areas should be protected with stucco mesh to prevent rabbits, raccoons, cats, dogs and dust bathing birds and others from digging up the nests.

What?

- Conduct one to two turtles surveys a month in Mar, Apr and May
- Monitor nesting grounds using time-lapse camera (10 min intervals) during nesting season (May, June, July)
- Protect known nests with a 2 x 2 inch stucco mesh that are located outside the nesting areas

- Monitor emergence of hatchings on nesting grounds weekly (mid-April to May), by looking for holes where the turtles escaped

When?

- See above

Where?

- Where restoration took place

A few General Management Guidelines in the park

The following guidelines are general measures that are recommended before any maintenance activities or infrastructure development is undertaken:

1. General Guidelines:

- Check for Western Painted Turtle occurrences or nesting grounds before undertaking projects that can potentially harm turtles

2. Maintenance of Trails, roads and parking lots maintenance

Rationale:

The main threat from maintenance of trails, roads and parking lots is inadvertent destruction of turtle nests located in the gravel in these areas. Adding gravel can destroy nests by preventing eggs from hatching and hatchings from successfully dig themselves out of the nest. It is, however, possible to minimize these risks, because nests are usually located on the side of the roads and the shoulder rather on the roadbed itself.

See section ***4. Maintenance of restoration activities*** for details about timing, as these also pertain to maintenance of trails, roads and parking lots.

Suggested BMPs:

What?

- Where turtles are known to nest along trials, roads and parking lots and restored nesting sites, conduct maintenance to trials before spring emergence of hatchlings after the turtles hatchlings have emerged by mid May (this is weather dependent) and the adult females are nesting in June (also weather dependent). The period between emerging hatching and egg laying females is time where there is the least chance to do harm.
- In other areas where trail are immediately adjacent to the lake, either take above precautions *or* survey for signs of turtle nesting activity on trail before starting work.

When?

- Grade and repair trails, roads and parking lots in late spring, after hatchlings have emerged from nests but before egg-laying begin. There is often a narrow seasonal time-window when these activities can be conducted safely in sensitive turtle habitats. There is, however, variation form year to year, and occasionally the two events overlap. This seems to happen when the spring is long and cold. It is best to monitor known nest to identify about the exact timing of this biological window. If that is not possible the rule-of-thumb for the optimal period is in last week of May.
- Surveys for signs of turtle nesting activity can be conducted anytime from about March to July.

Where?

- The BMPs apply to sections of trails, road and parking lots where the Western Painted Turtle is known to nest, and where trails are immediately adjacent to the pond

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