

**Alaksen National Wildlife Area: Reservoir suitability
for the introduction of the endangered Western
Painted Turtle**

**by
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Abstract

Alaksen National Wildlife Area located in Delta, BC is home to freshwater species in the former tidal marsh. The current agricultural landscape has left a legacy of high concentrations of heavy metals, trace amounts of organochlorine pesticides, and excess nutrients within the sediments and water of the brackish Fuller and Ewen Reservoirs. Arsenic and phosphorous exceeded Canadian water quality guidelines, while arsenic, chromium, copper, iron, manganese, nickel, and phosphorus exceeded sediment quality guidelines. There were trace pesticides known to be endocrine disruptors detected in the water and sediment, and combined low level toxicity effects are a concern. A preliminary ecological risk assessment on the metals was completed and the results indicate that there is a possibility of adverse effects for benthic invertebrates, but negligible risk for endangered Western Painted Turtles. However, compounding all the ecosystem stressors along with rising sea levels leads ANWA not an ideal place to introduce this species.

Keywords: Western Painted turtles; ecotoxicology; risk assessment; agricultural reservoir; heavy metal; endocrine disruptors

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List of Acronyms

1,2,3,4-TeCB	1,2,3,4-Tetrachlorobenzene
α -HCH	Alpha-hexachlorocyclohexane
ANWA	Alaksen National Wildlife Area
BCIT	British Columbia Institute of Technology
β -HCH	Beta-hexachlorocyclohexane
CCME	Canadian Council of Ministers of the Environment
CEPA	Canadian Environmental Protection Agency
CWS	Canadian Wildlife Service
COPC	Contaminant of potential concern
DDD	Dichlorodiphenyldichloroethane
DDE	Dichlorodiphenyldichloroethylene
DDT	Dichlorodiphenyltrichloroethane
δ -HCH	Delta-hexachlorocyclohexane
ERA	Ecological Risk Assessment
EDC	Endocrine Disrupting Chemical
γ -HCH	Gamma-hexachlorocyclohexane
HCB	Hexachlorobenzene
HQ	Hazard quotient
LEL	Lowest effects level
OCP	Organochlorine pesticides
PCA	Pentachloroanisole
PCNB	Pentachloronitrobenzene
PeCB	Pentachlorobenzene
POP	Persistent organic pollutants
PCDD	Polychlorinated dibenzodioxins
RES	Red-eared Sliders
ROC	Receptor of concern
SFU	Simon Fraser University
USEPA	United States Environmental Protection Agency
WPT	Western Painted Turtles

Preface



The sole 'Lone Ranger' Western Painted Turtle at Alaksen National Wildlife Area resting in front of one of my traps. I did not catch him.

Introduction

History of Alaksen National Wildlife Area

Alaksen National Wildlife Area (ANWA) located on Reifel Island, Smoky Tom, and Westham Islands in the Corporation of Delta was originally part of the Fraser River Delta Marshlands. Starting in 1898, portions of Westham Island were dyked and the marshes were converted to agricultural fields. By 1930, Reifel and Smoky Tom Islands were connected by causeways and similarly dyked, only connected to the Fraser River through culverts and control gates (Retfalvi, 1986). The Canadian Wildlife Service (CWS) acquired the land in 1972 to create a protected wildlife area and bird sanctuary after more than 40 years of private ownership and agricultural practices. ANWA encompasses 349 ha of remnant marshes, riparian shrubs and forests, agricultural fields, and anthropogenic sloughs (ECCC, 2017). As such, it is an important habitat and feeding area for migratory birds and part of the land overlaps Reifel Migratory Bird Sanctuary. In addition, portions of ANWA are included as a Ramsar site, in the Boundary Bay – Roberts Bank – Sturgeon Bank Important Bird Area, and the Fraser River Estuary Western Hemisphere Shorebird Reserve Network (ECCC, 2017).

Westham Island, where ANWA is situated, has had a history of over 80 years of agriculture use and thus contamination of this area by agricultural related contaminants (i.e. pesticides, fertilizers, etc) is likely and may have implications for wildlife inhabiting or using this area. Recently, two pesticide surveys in 2009 and 2012 by CWS were undertaken. The purpose of these past surveys was to determine the legacy and current levels of several pesticide contaminants in this area, and several organochlorine, organophosphate, and phenylurea pesticides were detected (CWS 2009, 2012 unpublished data). Although numerous wild species inhabit and use ANWA, this site is of particular importance as critical habitat for the Western Painted Turtle (WPT). Historically this site was Fraser River tidal marshlands, and would not be the typical place to find WPT as the water would be too saline; however there is one WPT that lives at ANWA based on turtle trapping and basking surveys completed in 2009, and visual observation in 2018 by CWS. The WPT found in ANWA is part of a genetically unique and Endangered species in the Lower Mainland and as such, this site is designated as Canadian government 'Critical Habitat.' With ANWA used and inhabited by numerous

wildlife species, including the endangered WPT, ongoing research to better understand the impacts of anthropogenic contaminants and physical habitat changes limiting healthy wildlife populations is key to managing this area.

Contaminants in Agricultural Landscapes

Wetlands have a large ecological significance and support high numbers of species (Bartzen et al., 2010; Zedler and Kercher, 2005). However, over 50% of the wetlands on the planet have been lost, primarily because of agriculture (Zedler and Kercher, 2005). Compounding these losses, many of the remaining wetlands have been degraded through salinization, hydrologic manipulation, eutrophication, sedimentation, invasive species invasion, and climate change (Zedler and Kercher, 2005). Ecosystems within agricultural areas tend to experience multiple stressors that can be cumulative and adversely affect wildlife populations and communities (Çavaş and Könen, 2007; Zedler and Kercher, 2005). Indeed, wetlands are often a sink for various organic and inorganic contaminants and nutrients from nearby anthropogenic sources since they are typically the lowest points in a landscape. These unseen contaminant stressors can affect the soil biogeochemical equilibrium by concentrating in soil, sediments, and water after extended periods of time, and ultimately, reduce biodiversity (He et al., 2005; Zedler and Kercher, 2005). Two major groups of anthropogenic contaminants of growing concern globally are pesticides and metals due to their frequent use and detection above environmental quality guidelines for the protection of humans and wildlife. In particular, some synthetically produced organic pesticides can resist degradation in the environment for many years, biomagnify in food chains, and have negative ecotoxicological effects (Jones and de Voogt, 1999). Monitoring and understanding all the effects of these pesticides in non-target organisms is difficult and complex because of all the different mixtures and their interactions in the environment.

There are over 7,000 registered pesticides in Canada with an estimated total of 101 million kilograms of active ingredients sold in 2014 (Health Canada, 2014). Many of the highly toxic and persistent pesticides and numerous other types of anthropogenic chemicals have been progressively banned since their first registration and use in Canada. However, repeated historical applications over time and long half-lives of these chemicals have resulted in continued pollution in many areas of the world that merits ongoing investigation (Jiao et al., 2012). For example, several organochlorine pesticides

(OCP) have been banned in Canada since the 1970s but concentrations are still detected in the environment globally (i.e. DDT, HCB, HCB, Aldrin, and mirex) due to their persistent nature (Moss et al., 2009). In addition, there is mounting evidence that the environmental persistence of pesticides can change when there are numerous chemical contaminants interacting in an area (Swarcewicz et al., 2013). These interactions may have additive, synergistic, potentiating, or antagonistic effects. However, the fate, exposure, and toxicity of these pesticides when present in mixtures is largely unknown.

Pesticides can be taken up and affect non-target organisms in the environment through contamination of water, sediments, or air. Several adverse effects after exposure to many types of pesticides have been shown to cause mortality, immunotoxicology, genotoxicity and cytotoxicity (Çavaş and Könen, 2007; D'Souza et al., 2005), behaviour alterations (Saglio and Trijasse, 1998), and disrupt the endocrine system in non-target organisms (Hayes et al., 2003; Sumpter, 2005; Zhou et al., 2010; WHO and UNEP, 2012). Endocrine disrupter chemicals (EDCs) have gained considerable attention globally and are a more recently discovered mode of toxic action. Furthermore, the EDC issue has brought to light concerns regarding the health risks of low level (parts per million, parts per billion, and parts per trillion) continuous exposure of humans and wildlife to chemicals and mixtures of chemicals that are a common exposure scenario in most areas inhabited by humans. The definition of an EDC has been added to several government jurisdictions, including the Canadian Environmental Protection Act (CEPA) and is defined as chemicals that interfere with natural hormones by “disrupting the synthesis, secretion, transport, binding, action, or elimination of natural hormones in an organism or its progeny” (CEPA, 1999). Although few chemicals, including pesticides, have undergone testing to identify an endocrine disrupting mode of action, many different chemical classes (e.g. organochlorine pesticides, organophosphate pesticides, phenylurea herbicides, etc) have been deemed EDCs (McKinlay et al., 2008). Since the endocrine system controls major biological processes, a chemical that can mimic a natural hormone (i.e. estrogen, thyroid hormone, cortisol) and bind or activate hormone receptors has been shown to impair growth, development, and gonadal maturity in organisms, as well as increasing the risk of being sensitive to environmental stresses (Mnif et al., 2011; Zhou et al., 2010).

Current use anthropogenic chemicals, pesticides and fertilizers often have trace or heavy metals in their formulations, and when deliberately released on agricultural fields, it has resulted in their prevalence in the environment in many areas (He et al., 2005; Jiao et al., 2012). Unlike organic synthetic contaminants, metals are natural substances and do not degrade in the environment. Accumulation of heavy metals in soil, sediment, and water outside their natural geologic sources is assumed to be from human influence rather than the natural variability and origin of macronutrients (Bradl, 2005; He et al., 2005). Elevations above natural background levels can have adverse impacts on biota and are redistributed by plant and animal uptake, their ability to bind onto soil particles, and in their dissolved forms in water (Bradl, 2005). The high levels of trace heavy metals found in wetland water and sediments around agricultural lands can originate from a few different sources through repeated use on crops over time such as with phosphorus fertilizers, pesticide mixtures, manure fertilizers, fungicides, chemical fertilizers, and biosolids (Ashley and Stockner, 2003; Bradl, 2005; He et al., 2005; Jiao et al., 2012; Karstens et al 2016). However, not all these excess metal concentrations are bioavailable at a given time, which is when they are accessible for uptake by organisms, as it is dependent upon the chemical properties in a wetland including pH, biological processes, and anaerobic/aerobic conditions (including different reactions of precipitation-dissolution, adsorption-desorption, complexation-dissociation, and oxidation-reduction) (He et al., 2005).

Metal speciation, which includes all the phases of the metal and whether it exists as a free ion or complexed to other chemicals, can be difficult to quantify because of the interactions between the different biogeochemical factors in a waterbody (Bradl et al., 2005; Camerlynck and Kiekens, 1982). To understand the mobility of heavy metals in the environment, knowing the influences of pH and redox potential are important (Bradl et al., 2005). Mobility can be increased by reducing pH, altering redox conditions to moderate/high, and increasing salinity. Factors such as dissolved organic matter, suspended particulate matter, ionic strength, and alkalinity are also important (Bradl et al., 2005). Understanding metal speciation in the environment is further complicated with the metals interactions with other metals in the environment, and further with organic pesticides. In total, there are many different anthropogenic impacts that can affect wildlife around agricultural areas; the difficulties lie in the understanding of the physical, chemical, and biological environment as a whole in an ecosystem, and then further

looking at the many genetically and phylogenetically unique species that can inhabit and interact within the environment.

The effects of pesticides and metals on non-target organisms, especially in real-world exposure scenarios, is only marginally understood and is understudied. Reptiles especially are one of the most underrepresented taxa, with most of the ecotoxicological data available being more qualitative than quantitative (Moss et al., 2009; Weir et al., 2015). Turtles are an important indication of environmental conditions and can be bioindicators of contamination (Meyer et al., 2016; Meyers-Schöne and Walton, 1990; Moss et al., 2009). They have long lifespans, generalist diets, low generational turnover, live in a variety of different ecosystems around the world, and have interactions with sediments, water, and macroinvertebrates (Meyer et al., 2016; Meyers-Schöne and Walton, 1990; Moss et al., 2009). Living among the sediments and part of a turtle's diet, benthic invertebrates can transfer sediment bound contaminants up the food chain (Li et al., 2017). As well, because turtles lay eggs within sediments and soils near wetlands, there is a concern for inorganic and organic contaminants to enter incubating eggs (Allan et al., 2017; de Solla and Fernie, 2004; de Solla and Martin, 2011). While habitat destruction and human exploitation have negatively impacted about two-thirds of turtle species around the world, one of the largest unseen threats to reptile populations and conservation efforts is environmental contamination (Charruau, 2013; Moss et al., 2009).

Ecological Risk Assessment – Western Painted Turtle and Alaksen National Wildlife Area

A method of quantifying the risks the chemical contaminants and human impacts in general at ANWA pose to the Western Painted Turtle is through an ecological risk assessment (ERA). An ERA is a way to evaluate the potential adverse effects of anthropogenic stressors on individuals, populations, or communities of non-human organisms (EC, 2012). An ERA of ANWA is warranted because of the many anthropogenic influences in terms of historical, physical, chemical, and biological changes to this wetland, and because of its historical and current land use introducing several anthropogenic contaminants. Currently, the reservoirs on site have no emergent or submergent vegetation so function more as a sink and resuspension area for contaminants than a wetland that can naturally filter, biotransform, and phytoremediate.

ERA's can inform restoration plans by addressing areas of management concern before restoration activities take place and having a better understanding of the physical, biological, and chemical stressors in the environment (Kapustka et al., 2016; Wagner et al., 2016). Knowing more about the unseen stressors can aid in long term restoration goals and increase ecosystem resiliency and integrity, which will be a key factor for ANWA as it functions as a wildlife area. A successful ERA should integrate restoration management goals in the planning phase by incorporating assessment endpoints related to ecosystem services and functions, which has not always been the case in the past (Kapustka et al., 2016). It is essential to understand the biophysical environment to be able to predict the stability and mobility of contaminants when developing restoration goals as that could limit restoration potential (Wagner et al., 2016).

ERA's can complement ecological restoration by having a broader understanding of the drivers that influence the current condition of the site (Kapustka et al., 2016). This understanding can help determine if remediation is required or if restoration is a feasible option. However, there are still limitations regarding the utility and success of an ERA based on several poorly understood scientific phenomena as well as the translation of scientific findings into government regimes aimed at protecting the environment. For example, there are considerable knowledge gaps related to the complexity of nature such as stochasticity within ecosystems, diverse food webs, trophic cascades with feedback loops, and the often varied provincial and federal legislation of government and associated policies that affect environmental interpretation (Kapustka et al., 2016). Yet, having a greater understanding of the geochemical and biophysical environment, having an integrated and iterative process between the two different approaches, and knowing more in-depth about the risks to ecosystem services in degraded landscapes is a more robust approach than either method alone. An ERA is multi-disciplinary and is concerned about both the science and management of the lands (EC, 2012). Completing a preliminary ERA within this project will help in the long term management at ANWA and future restoration initiatives.

Objectives

The Coastal population of the Western Painted Turtle (*Chrysemys picta bellii*) (WPT) is at its northern range in British Columbia and is the only freshwater turtle left in the province. Introducing an endangered species to a wetland that may have high levels

of contaminants in the water and sediments are a concern for hatchlings with developing reproductive systems as reptile eggs are porous and allow gases and water to pass through (de Solla and Fernie, 2004; de Solla and Martin, 2011). The data from this study will assist the WPT Recovery Team and the Ministry of Forests, Lands & Natural Resource Operations with assessing the suitability of this site to support a sustainable WPT population. Thus, the objectives of this study were to determine 1) select pesticide and metal contaminant concentrations in water and sediments in ANWA and the risk these levels of contaminants may pose to a WPT population; 2) the overall suitability of this site for future introductions of juvenile WPT; and 3) to describe potential restoration prescriptions at ANWA.

Methods

Study Site

Alaksen National Wildlife Area is located in Delta, BC and has been a Wildlife Area for over 40 years. The site under investigation in this study was located around and within both Ewen and Fuller Reservoirs in ANWA (Figure 1). The bottom substrate is largely sand and gravel covered by a thick organic detritus layer with some muddy/clayey areas (Kilburn and Mitchell, 2011).

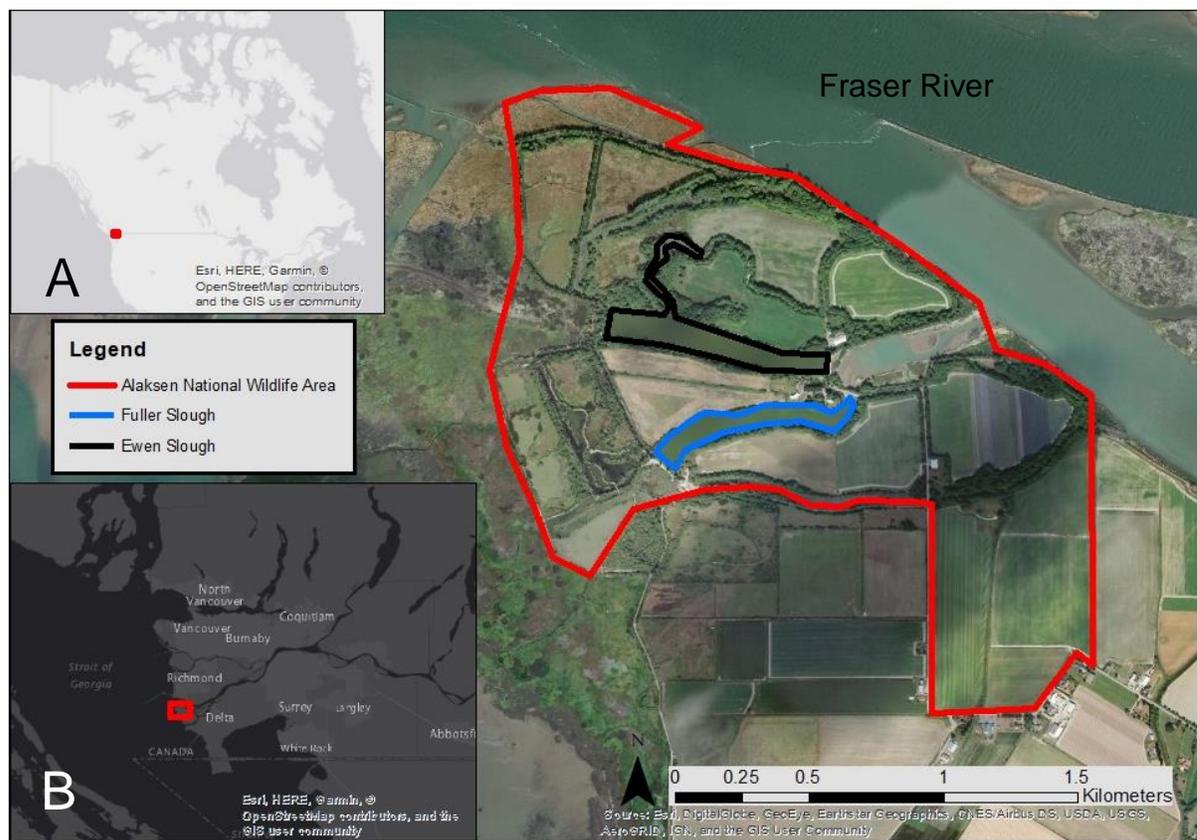


Figure 1 - Alaksen National Wildlife Area in Delta, BC is outlined in red. The blue and black outlines represent the two reservoirs that were the main areas of study. Inset map B shows where in the Lower Mainland ANWA is located, while inset map A shows the study area in relation to Canada. Basemaps were obtained from ESRI.

Two former deltaic channels form the agricultural reservoirs of Ewen and Fuller. These reservoirs have steep banks and on average are only 1.5 m deep, though they have been dredged throughout the last century and have deep pockets up to 10 metres

at the Western ends (Figure 2). Water from the Fraser River is pumped in during freshet and into July to fill and maintain the reservoirs for agricultural use on fields. This water is pumped through a series of shallow dyked channels and are controlled by floodgates.



Figure 2 – The bathymetry profiles of both Ewen and Fuller Reservoirs. These maps were compiled from individual depth readings ($n \approx 100/\text{reservoir}$). There are two deep (10 m) ‘pockets’ at the western ends of the reservoirs, while the remainder of the water level is around 1.5–2 m in depth.

Animal species at ANWA include a plethora of migratory birds, a few species of amphibians, and listed species such as the Western Painted Turtle (*Chrysemys picta bellii*), Great Blue Heron (*Ardea herodias*), and Barn Owl (*Tyto alba*) (ECCC, 2017). The sole western painted turtle on site was identified in 2009, and trapped in Fuller reservoir in 2010 (Kilburn and Mitchell, 2011); WPT would not have been native in deltaic marshlands normally and it is uncertain how this one arrived at ANWA. The WPT has eluded trapping since then, but has been observed basking on a log in Fuller reservoir in successive years and most recently in 2018. Animal species also include a number of invasive species like Red-eared Sliders (*Trachemys scripta elegans*), common carp

(*Cyprinus carpio*), catfish (*Ameiurus* spp.), and pumpkinseed (*Lepomis gibbosus*). These were opportunistic sightings and catches and do not reflect an invasive species survey or count. Most recently in 2018, an invasive plants survey was conducted over the whole site. The study site is located around and in both Ewen and Fuller reservoirs as a possible area for introducing more headstarted WPT to maintain a population of WPT.

Habitat and Landscape Analysis

Water Quality

Water quality was monitored from June to August twice a week between the hours of 9:00-16:00. The sampling dropped to once a month between September and November. Water temperature, dissolved oxygen, conductivity, salinity, pH, and turbidity were the parameters measured with an YSI Professional Plus Multiparameter (YSI Incorporated, Yellow Springs, Ohio, USA) and LaMotte 2020we Turbidimeter (LaMotte, Maryland, USA). There were two locations in Fuller reservoir and three locations in Ewen reservoir where water quality was measured (Figure 3). Three depths were sampled depending on the bathymetry at the sampling location and were as follows: Fuller reservoir, the depths were 0.2 m, 1.5 m, and 4.75 m; Ewen reservoir, the depths were 0.2 m, 2.5 m, and 4.75 m.

Turtle Trapping

To ensure there was no unnecessary pain and distress, the Canadian Council on Animal Care species specific recommendations on amphibians and reptile standards were followed (CCAC, 2004). This research was also completed under approval from BC FrontCounter Wildlife Act Permit (SU18-357631), Alaksen National Wildlife Area General Wildlife Permit, and an Environment Canada SARA permit (BC-18-0044).

Western Painted Turtles (WPT) and Red-eared Slider (RES) turtles were trapped over a period of 9 days from July 3, 2018 to July 18, 2018. The trapping entailed deploying D-nets (a style of hoop trap), and in total 14 D-nets were set up on the banks/edge of water in both Ewen and Fuller reservoirs (Figure 3). The D-nets were suspended in shallow water on large woody debris as near to basking logs as possible,

following the South Coast Western Painted Turtle Recovery Project's trapping procedure (Kilburn and Mitchell, 2011). The D-nets were baited with canned cat food. The cat food was inside small mesh bags and suspended from the frame of the trap with the lid partially opened. The D-nets were deployed on Mondays with fresh bait each week, and the bait was removed from the traps on Friday as traps were disassembled for the weekend.

The D-nets were secured in place with 4-5 inches of air space above the surface of the water to allow any reptiles caught to breathe and allow easy access for retrieval. All of the D-nets were checked twice daily during the Canadian Wildlife Service's hours of operation between 8:00-16:00 to minimize time spent in the net for any species caught. Although no WPT were caught in the D-nets, the protocol upon capture of a WPT was to weigh and measure the individual, take a blood sample for DNA analysis, and check for previous marking of the shell for identification. If the turtle shell had no notches, it would then be marked using the shell notching methods outlined in Kilburn and Mitchell (2011). Any turtles caught were weighed and measured; the RES that were collected were brought to the veterinarian clinic in Maple Ridge for euthanasia, while any WPT caught were identified, and those unmarked were to be marked (using unique shell notching) and a blood sample taken according to Kilburn and Mitchell, 2011. Invasive fish species caught as bycatch were euthanized with 0.4 mg/L MS222 (tricaine methanesulfonate), buffered to neutral pH with sodium bicarbonate, and disposed of at SFU.

Water and Sediment Sample Collections

Water Collections

Water sampling procedures were obtained from the BC Field Sampling Manual (MOE, 2013). One litre of surficial water was collected at a depth of 0.1 m in both Ewen and Fuller reservoirs to measure organochlorine pesticides, linuron, metals, and nutrients. There was one sample collected at one location in Fuller (Site 1) and two in Ewen (Sites 2 and 3) (Figure 3). Water samples were collected in water free of vegetation and a minimum of two metres from the reservoir's banks. ALS Environmental (Burnaby, BC, Canada) provided all cleaned water collection vials, coolers, and ice packs to store samples at 4°C.

Sediment Collections

Sediment sampling procedures were followed from the BC Field Sampling Manual (MOE, 2013). The sediment was extracted from the uppermost 0.15 m of the reservoir bottom as this uppermost region near the interface with water has been proposed to contain the most organochlorine residues, even after extensive time has passed (Marburger et al., 2002). This is also a sediment region that WPT are expected to encounter when they overwinter. One sediment sample was collected from each reservoir at Site 1 in Fuller, and Site 2 in Ewen (Figure 3). An Eckman Grab Sampler (Van Walt Ltd., Haslemere, UK), as suggested by the BC Field Sampling Manual, was deployed from a canoe three times, at each sample location and these 3 cores/site were

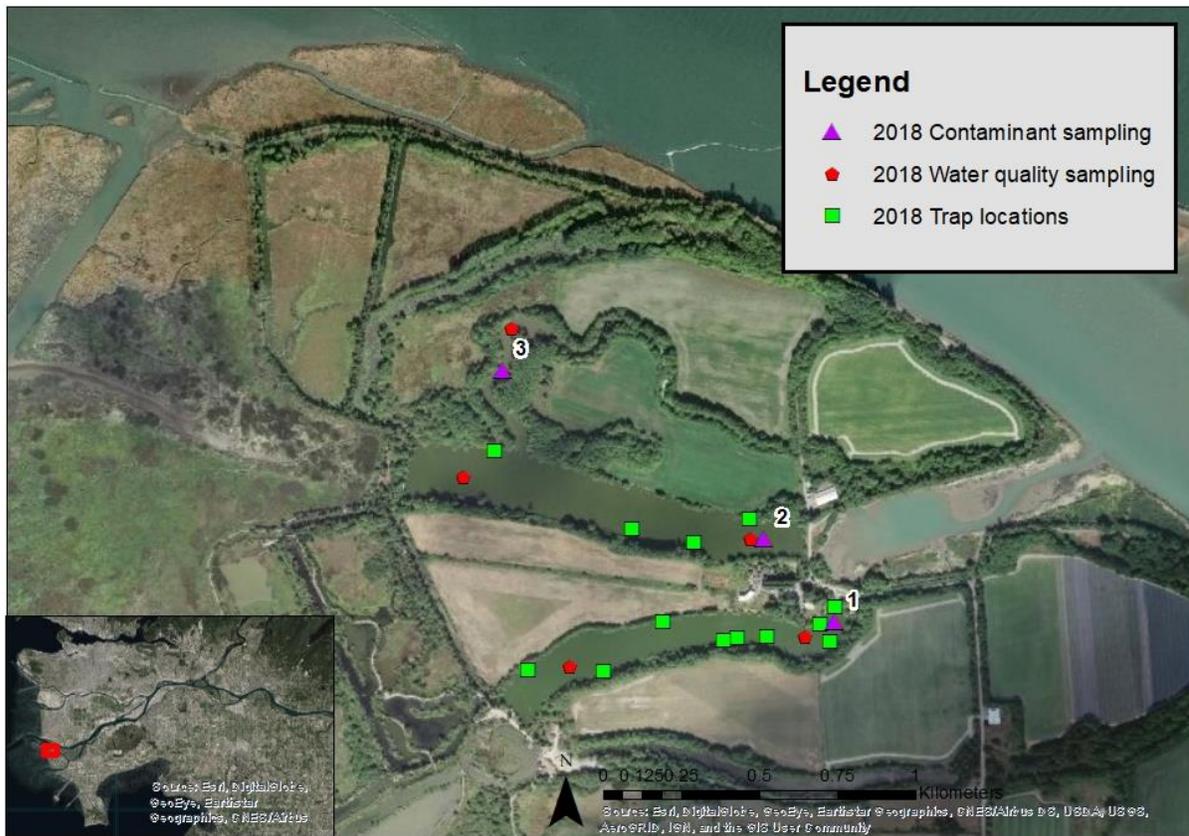


Figure 3 - Alaksen National Wildlife Area sampling map. Purple triangles are the contaminant sampling locations in 2018; Sites 1 and 2 had both sediment and water collections while Site 3 had solely water collections. The red dots are the water quality sampling locations and green squares are the trap locations that were placed along the banks of the reservoirs. Basemap from ESRI.

combined in a large stainless steel bowl, mixed thoroughly before a sample was removed and placed in a single glass jar to form one composite sediment sample (MOE, 2013). Sampling equipment was cleaned with deionized water and 5% nitric acid prior to

field activities and between each deployment of the Eckman Grab Sampler to ensure no cross contamination of the sediment sample. All samples were stored at 4°C after collections at each site, and submitted to ALS Environmental within 8 hours of collection.

Water and Sediment Pesticide, Metal, and Nutrient Analyses

Contaminant analyses were completed by ALS Environmental in Burnaby, BC. The following metals, organochlorine pesticides, nutrients were analysed in the lab according to different analytical techniques (Table 1).

Table 1 - Pesticide Analysis methods by ALS. Each parameter is broken down by water and sediment techniques.

Parameter	Analytical Method	Analytes
Sediments		
Metals	EPA 200.2/6020A (mod)	Al, Sb, As, Ba, Be, Bi, B, Cd, Ca, Cr, Co, Cu, Fe, Pb, Li, Mg, Mn, Hg, Mo, Ni, P, K, Se, Ag, Na, Sr, S, Ti, Sn, Ti, W, U, V, Zn, Zr
Extractable metals	EPA 821/R-91-100; EPA 6010B	Cd, Cu, Pb, Hg, Ni, Zn
Nutrients	Nitrate-n Alberta Ag 1988	Available nitrate-n
Nutrients	Phosphate-p Comm. Soil. Sci. Plant. Anal. 25 (5&6)	Phosphate-p
Water		
Metals	EPA 200.2/6020A (mod)	Sb, As, Ba, Be, Bi, B, Cd, Cs, Ca, Cr, Co, Cu, Fe, Pb, Li, Mg, Mn, Hg, Mo, Ni, P, K, Rb, Se, Si, Ag, Na, Sr, S, Te, Tl, Th, Sn, Ti, W, U, V, Zn, Zr
Nutrients	Methods adapted from APHA4500-P(J)/NEMI9171/USGS03-4174 for total N	Total nitrogen
Nutrients	Phosphorous 4500-P – (Standards Method Committee, 1999)	Total phosphorous in water
Organochlorine pesticides	Method 1699 – pesticides extracted with Soxhlet, prepared with	α-HCH, β-HCH, γ-HCH, δ-HCH, t-Nonachlor, c-nonachlor, endrin-ketone, t-chlordane, c-chlordane,

	column chromatography, and analysed with high resolution gas chromatography/high resolution mass spectrometry (Quote and EPA, 2007)	alpha-endosulphan, beta-endosulphan, heptachlor-epoxide, HCB, Aldrin, dieldrin, o,p/p,p DDD,DDE,DDT, endrin, mirex, oxychlorane, endosulphan-sulphate, methoxychlor, heptachlor
Other pesticide	SW846 8270 Analysed by gas chromatography/mass spectrometry	Linuron

Water and Sediment Guidelines

The quality standards that were used for metals, pesticides, and nutrients followed the hierarchy of British Columbia as the first option for guidelines when it was available, followed by Canada (CCME), Ontario, and then the United States Environmental Protection Agency (USEPA).

Risk Assessment Methods

An ecological risk assessment is a way to look at current contamination in a site and determine the exposure risks to ecological receptors (EC, 2012). The ERA followed the methods outlined in the Federal Contaminated Sites Action Plan on Ecological Risk Assessment Guidance (EC, 2012). For this research, a modified risk assessment approach was used because of the data limitations of point measurements in the reservoirs, the lack of aquatic vegetation in the reservoirs, no soil samples collected on land, and no receptor tissue to determine contaminant concentrations. This ecological risk assessment will be a preliminary assessment of risk for wildlife in ANWA and for quantifying the degree of metal contamination to determine if further action should be taken. Fuller reservoir had heavy metals exceeding the quality guidelines, therefore only Fuller was used for the ERA. A weight of evidence approach uses different lines of evidence to quantify an assessment endpoint (EC, 2012). It is a way to combine information on both exposure and effects of different contaminants in the environment to determine the risk to receptors of concern. For this preliminary ERA, a weight of evidence approach with three lines of evidence were used using sediment and water

using chemistry data (Table 2). The protection goals within the ERA look at community level and individual level assessment and measurement endpoints within the condensed foodweb to look at the risk for WPT. This approach uses a hazard quotient (HQ) to determine risk with hazard quotients ranging from negligible to adverse effects possible (Table 3; EC, 2012). This calculation is a measure between the calculated exposure for the receptor and the effect based threshold known as a toxicity reference value (Equation 1; EC, 2012). The methods and equations to follow for risk assessments are based on current scientific knowledge, but still have limited data sets because of the complex interactions chemicals have in the environment (EC, 2012). As a result, some substances cannot be well characterised by a risk assessment because of there is no adequate endpoint testing available (on neurotoxins, behavioural effects, endocrine disruptors), or there is limited understanding and information on substances that are released from different stages of the life cycle.

The ecological receptors of concern will have a basic food web focus, looking at what interacts with both the sediments and water of Fuller reservoir. There is no plant tissue data so the aquatic plant community will not be looked at within this ERA. As well, Fuller has very limited to no submergent and emergent vegetation in the reservoir so this group will not be used. The three different receptor groups were benthic invertebrate communities, fish communities, and the WPT with a surrogate species of painted turtles (*Chrysemys picta*) used for the WPT during analysis (Table 4). This surrogate species was chosen as there was enough ecotoxicological and life history data available. WPT also has a diet mainly composed of plants and benthic invertebrates, along with some fish species, usually consumed as carrion (Ernst et al., 1994). However, the limitations of no plant tissue data and only sediment quality data mean that the diet of the WPT is assumed to be primarily from benthic invertebrates and fish carrion within this ERA. These three receptors were chosen because they are potentially exposed to contaminants of concern at Fuller reservoir, and lower trophic levels were looked at from a community perspective while the upper trophic level was defined at the species level within the lines of evidence (EC, 2012). Each receptor had a different exposure to contaminants based on the feeding guilds and diet, as well as different foraging behaviour that could influence exposure rates (EC, 2012). Inhalation exposure is assumed to be negligible for the WPT as the sites are vegetated and there is limited risk

of sediments leaving the reservoir. Operable pathways included sediment contact, water contact, and soil ingestion, water, and food ingestion (Table 5).

The concentration based exposure scenario is used for benthic invertebrates and fish as they have direct contact with sediment and water respectively (Equations 2 and 3; EC, 2012). The painted turtle is exposed to contaminants of potential concern through sediment and water and a dose based approach that is the sum of exposures (Equation 4) from different pathways was used to estimate the amount of contamination through diet, water ingestion, and sediment ingestion (Equation 5; EC, 2012). Risk characterization was calculated by determining the hazard quotient which consisted of the total dose or concentration calculated for the receptors divided by a toxicological reference value (TRV). A TRV is a threshold dose or concentration found through literature, or water and sediment quality guidelines that determine what level of contaminants would not harm the receptor (EC, 2012). TRV's used in the hazard quotient were based of US EPA values, and when those were not available values were pulled from the CCME water and sediment quality guidelines as they are protective of the most sensitive receptor and equivalent to TRVs (CCME, 2007).

Table 2 - Ecological risk assessment lines of evidence at ANWA.

Lines of evidence	Assessment endpoint	Measurement endpoint	criteria	weighting
Sediment	Benthic invertebrate community diversity and abundance	Comparison of concentration based exposure to toxicological reference values to calculate an HQ	HQ<1	100%
Water	Fish species survival at community level	Comparison of concentration based exposure to toxicological reference values to calculate an HQ	HQ 5-10	100%
Dose based	Painted turtles individual survival	Comparison of concentration based exposure to toxicological reference values to calculate an HQ	HQ<1	100%

Table 3 - Hazard quotient risk characterization

Hazard Quotient	Risk	Action
HQ < 1	Negligible Risk	None
HQ > 1	Adverse effects possible	Yes, more precise evaluation needed to address uncertainty

Table 4 - Receptors chosen through different aquatic groups and types. The exposure pathway describes the methods that were followed for each receptor.

Aquatic Receptor Group	Aquatic Receptor Type	Freshwater	Exposure pathway
Benthic invertebrate	Epifauna and infauna	Benthos community	Direct contact with sediment (concentration based)
Fish	Piscivorous	Fish community	Direct contact with water (concentration based)
Reptile	Omnivore	Painted turtle	Water, food, and incidental sediment ingestion (dose based)

Table 5 - Exposure pathways for the different receptors of concern at Fuller reservoir in ANWA.

Receptor	Dermal contact with soil	Dermal contact with water	Food ingestion	Water Ingestion	Soil Ingestion
Benthic invertebrates					
Fish					
Painted Turtle					

Equation 1.

$$HQ = \frac{Dose_{total} \text{ or } [C]}{TRV}$$

Dose_{total} or [C] = the calculated exposure through total dose or concentrations

TRV = toxicity reference value

Equation 2.

$$C_{invb} = BCF * [COPC_{sediment}]$$

BCF = bioconcentration or bioaccumulation factor for benthic invertebrates

C_{invb} = exposure to benthic invertebrate (mg/kg)

[COPC_{sediment}] = concentration of contaminant in sediment (mg/kg)

Equation 3.

$$C_{fish} = BCF * [COPC_{water}]$$

BCF = bioconcentration or bioaccumulation factor for fish species

C_{fish} = exposure to fish community (mg/L)

[COPC_{water}] = concentration of contaminant in water (mg/L)

Equation 4.

$$Dose_{total} = Dose_{food} + Dose_{water} + Dose_{sediment}$$

Equation 5.

$$Dose_{fws} = \sum_{fws} \frac{A}{HR} * \frac{IR_i * C_{ij} * Bio_{oral}}{BW} * P_i$$

$Dose_{fws}$ = exposure to contaminant attributed to food, water, or sediment (sum of three separate equations) (mg/kg/d)

$\frac{A}{HR}$ = Area of contaminated land divided by habitat range. Assumed to be 1 for Fuller reservoir.

IR_i = ingestion rate (kg/individual/day)

C_{ij} = concentration of contaminant in food type (mg/kg)

Bio_{oral} = bioavailability of COPC (unitless) and assumed to be 1

BW = body weight (kg)

P_i = Proportion of the food type in the diet

Results

Water and Sediment Quality

Dissolved oxygen (DO) had consistently the highest levels at 0.2 m in depth in both Ewen and Fuller reservoirs during June to September 2018 (Figure 4). The highest level for Fuller was just above 18 mg/L (~180% saturation) in June before decreasing to 15 mg/L (~150% saturation) by August, and again increasing to 18 mg/L in September. Ewen had a high DO level just above 15 mg/L in June before decreasing steadily to below 12 mg/L (~120% saturation) by August and September. The midlevel depths see the opposite trend, starting at 2-3 mg/L (~20-30% saturation) in June and increasing DO concentration until about 4-7 mg/L (~40-70% saturation). This could mean that there was some mixing throughout the water column. Both Ewen and Fuller had close to 0 mg/L throughout the sampling period at 4.75 m depth, thus both the holes at the western ends of the reservoirs are anoxic all the way to the sediments at ~10 m depth.

The pH at both Ewen and Fuller had very similar data between the different depths throughout the sampling period of June to September (Figure 5). The pH remained above 9 at 0.2 m in both reservoirs during the day, reaching a peak in July at 9.42 and ending at 9.26 by September. The midlevel depths started in June at a pH around 8 before increasing by the end of September to 9. At 4.75 m of depth, the pH hovered around 7, both Ewen and Fuller diverging very slightly in July by 0.40.

Conductivity varied between the reservoirs and depths during June to September (Figure 6). The conductivity values at Ewen ranged from around 6,000 $\mu\text{S}/\text{cm}$ at 0.2 m depth to around 9,000 $\mu\text{S}/\text{cm}$ at the deepest point. There is a general trend of conductivity increasing from May to September. However, these are less than Fuller reservoir, which has a higher conductivity at all depths starting with a 0.2 m depth reading of around 9,000 $\mu\text{S}/\text{cm}$ in May and increasing to 11,000 $\mu\text{S}/\text{cm}$ by September. The midlevel reading very closely follows the 0.2 m depth trend. At 4.75 m in the hole, the conductivity is ~12,000 $\mu\text{S}/\text{cm}$ in May, and increases to ~15,000 $\mu\text{S}/\text{cm}$ by September. In August there is an increase back to 12,000 $\mu\text{S}/\text{cm}$. However, this increase in August at 4.75 m is not seen at the upper level depths in Fuller, which have a steady decrease throughout the sampling period.

The salinity at ANWA generally increased at all depths throughout the sampling period in both reservoirs (Figure 7). The salinity at 0.2 m and 1.5 m depth is very similar in both Ewen and Fuller separately. In Fuller, the salinity surface reading at 0.2 m and 1.5 m in June was around 5-6 ppt before increasing in September to ~6.5 ppt. At 4.75 m depth, the salinity starts in June at ~7.5 ppt before increasing to ~9 ppt in September. In Ewen, the 0.2 m and 2.5 m depth decreases from ~3/4 ppt to ~3 ppt from June to August before decreasing to back to 4 ppt in September. At 4.75 m depth, the salinity is around 5 ppt from June to August before decreasing to 6 ppt in September. It is more saline near the bottom in both reservoirs.

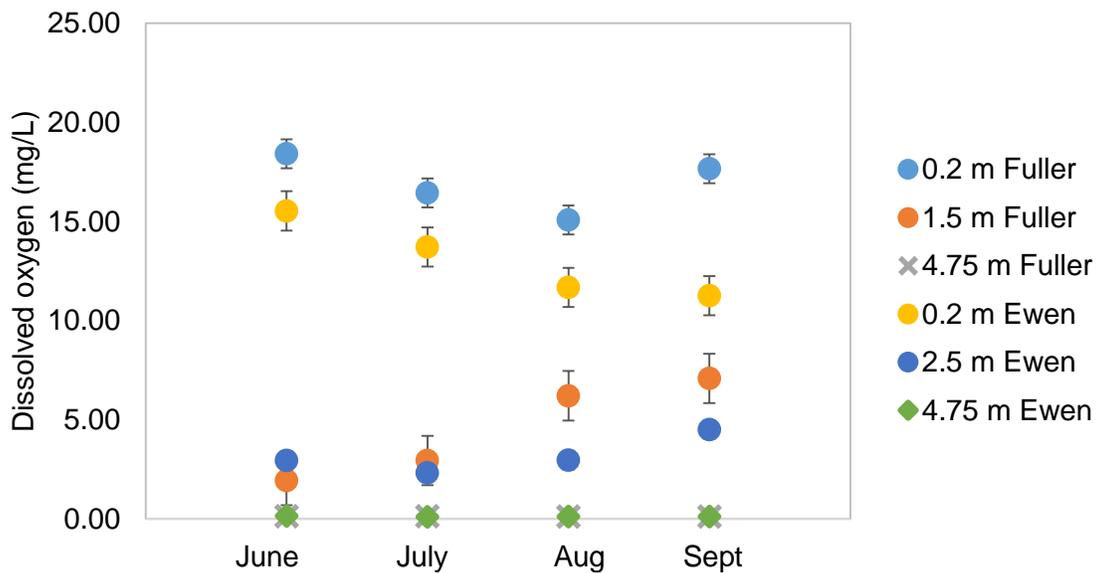


Figure 4 – Monthly averages for dissolved oxygen (mg/L) in Ewen and Fuller reservoirs at ANWA, Delta, BC, Canada at different depths from June to September, 2018. Dissolved oxygen was measured twice weekly in July and August and once weekly in June and September (at 0.2 m and 1.5 m/2.5 m depth, $n = 10$, $n = 16$, $n = 7$, $n = 7$ for each month respectively; at 4.75 m depth, $n = 4$, $n = 7$, $n = 4$, $n = 4$).

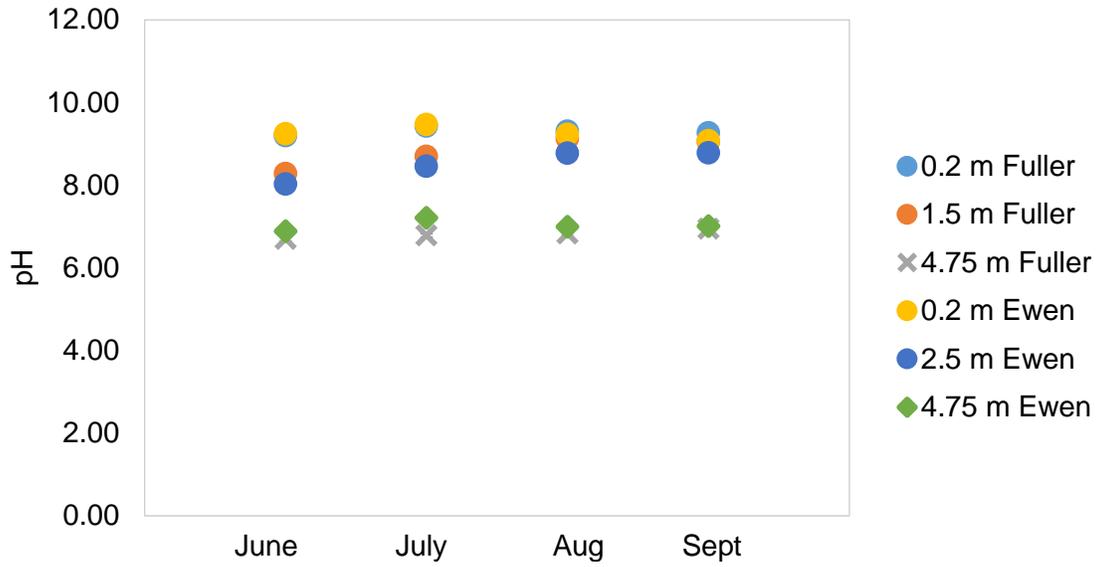


Figure 5 – Monthly averages for pH in Ewen and Fuller reservoirs at ANWA, Delta, BC, Canada at different depths from June to September, 2018. The pH was measured twice weekly in July and August and once weekly in June and September (at 0.2 m and 1.5 m/2.5 m depth, $n = 10$, $n = 16$, $n = 7$, $n = 7$ for each month respectively; at 4.75 m depth, $n = 4$, $n = 7$, $n = 4$, $n = 4$).

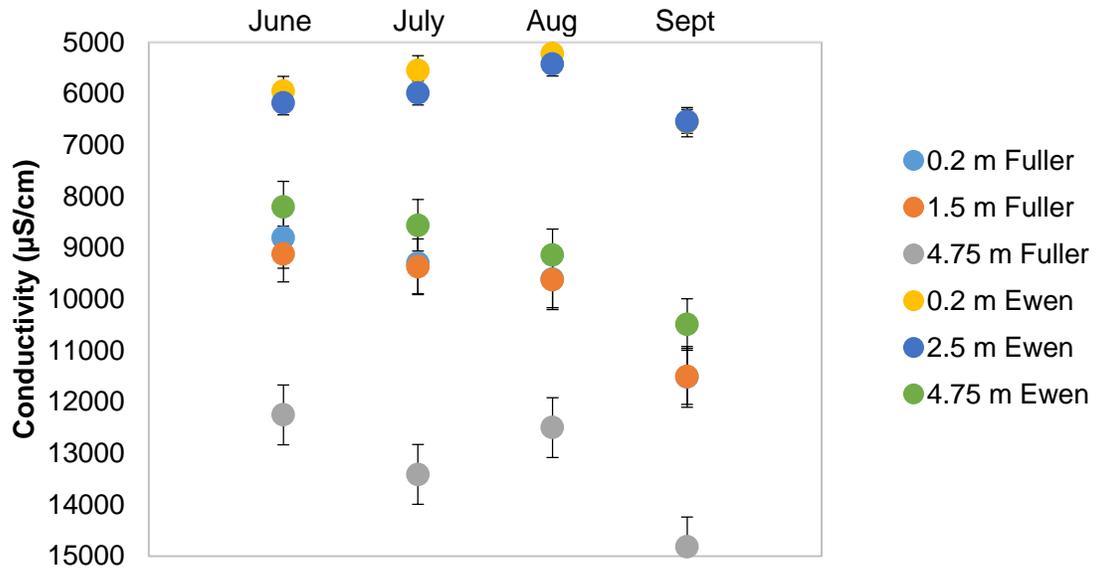


Figure 6 - Monthly averages for conductivity ($\mu\text{S}/\text{cm}$) in Ewen and Fuller reservoirs at ANWA, Delta, BC, Canada at different depths from June to September, 2018. The conductivity was measured twice weekly in July and August and once weekly in June and September (at 0.2 m and 1.5 m/2.5 m depth, $n = 10$, $n = 16$, $n = 7$, $n = 7$ for each month respectively; at 4.75 m depth, $n = 4$, $n = 7$, $n = 4$, $n = 4$). The conductivity axis was flipped to denote conductivity increases with depth in each of the reservoirs.

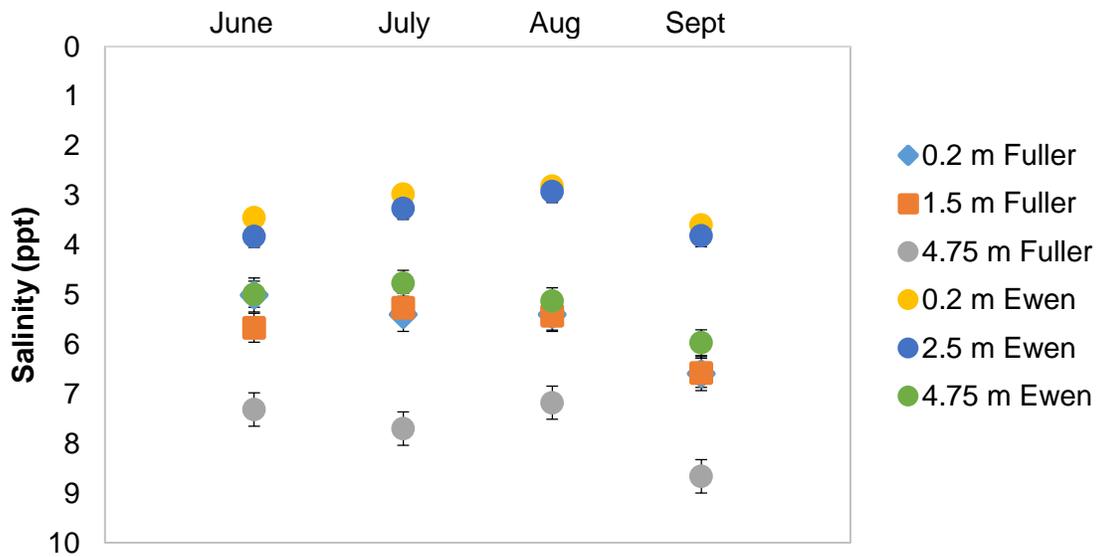


Figure 7 - Monthly averages for salinity (ppt) in Ewen and Fuller reservoirs at ANWA, Delta, BC, Canada at different depths from June to September, 2018. The salinity was measured twice weekly in July and August and once weekly in June and September (at 0.2 m and 1.5 m/2.5 m depth, $n = 10$, $n = 16$, $n = 7$, $n = 7$ for each month respectively; at 4.75 m depth, $n = 4$, $n = 7$, $n = 4$, $n = 4$). The salinity axis was flipped to denote salinity increasing with depth in the water.

Nutrients in Sediment and Water

The levels of total phosphorus in water and sediment were found to exceed CCME and Ontario guidelines for the protection of aquatic life in almost all the sites (Table 5). The total phosphorous in water is above the 0.1 mg/L threshold, indicating that the water is hyper-eutrophic (CCME, 2004). There are no guidelines for total hardness or total nitrogen measured in water in CCME, BC, or Ontario water quality guidelines.

Table 6 – Water nutrient concentrations (mg/L), sediment nutrient concentrations (mg/kg), hardness in water (as CaCO₃), and sediment pH in both Fuller and Ewen reservoirs at ANWA, Delta, BC, Canada. These are single point collections taken in August, 2018.

Water parameter measured	Site 1 Fuller (mg/L) n=1	Site 2 Ewen (mg/L) n=1	Site 3 Ewen (mg/L) n=1	Water Quality Guidelines (mg/L)
Hardness (as CaCO ₃)	929	563	559	N/A ¹
Total Nitrogen	2.45	1.61	1.56	N/A ²
Phosphorus (P)-Total	0.36	0.183	0.216	0.1 (hyper-eutrophic)³
Sediment parameter measured	Site 1 Fuller (mg/kg) n=3	Site 2 Ewen (mg/kg) n=3		Sediment quality guidelines (mg/kg)
pH	7.93	8.0		N/A ⁴
Available Nitrate-N	<2.0	<1.0		N/A ⁵
Available Phosphate-P	2.9	<2.0		N/A ⁵
Phosphorus (P)	1073.7	430.0		600⁶

¹There are no guidelines for total hardness in CCME or USEPA guidelines. However, water that is more than 180 mg/L is considered very hard (Durfor and Becker, 1964).

²There are no guidelines for total nitrogen measured in water, the convention is to measure the nitrate ion (NO₃⁻) to determine the water quality (CCME, 2012).

³ According to CCME (2004), concentrations over 0.1 mg/L are hyper eutrophic.

⁴There are no guidelines for pH in sediment.

⁵There are no available sediment quality guidelines in CCME or from the US EPA.

⁶MEEQ, 1993.

Contaminant Data

Metals

Total metals in sediment and water were measured in samples collected from both reservoirs in August 2018. There are three sites for total metals in water (1 in Fuller and 2 in Ewen) and only two sites for metals in sediments (1 in Fuller and 1 in Ewen). There were 27 metals in water detected out of 27 different metals measured (Table 6); and 32 metals in sediment detected out of the 32 different metals measured (Table 7). The water hardness (CaCO_3) was also measured in each water sample collected and was 929 mg/L in Fuller, and 563 mg/L and 559 mg/L in Ewen. This indicates that the water is very hard, thus impacts the biological availability of some of the metals such as cadmium, lead, and manganese. These CaCO_3 values exceed the hardness limits within the equation in the water quality guidelines for each metal, and very likely are not above the aquatic freshwater life threshold. A specific site assessment is recommended instead. Chronic exposures within the water and sediment quality guidelines were chosen over the acute exposure guidelines to look at exposure rates over a lifetime, more indicative for a long lived species.

Arsenic exceeded water quality guidelines at Fuller reservoir, while phosphorus exceeded guidelines at Ewen reservoir. Both Ewen and Fuller reservoirs exceeded the sodium water quality guidelines. None of the other metals exceeded water quality guidelines. The Ministry of Environment in BC and CCME water quality guidelines were the primary sources used, but when no guidelines were available, USEPA was secondary. For the sediment quality, Fuller had more metal exceedances than Ewen with arsenic, chromium, copper, iron, manganese, and phosphorus all exceeding applicable guidelines. Nickel was the only metal that was exceeded at both Ewen and Fuller reservoirs. CCME and Ontario sediment quality guidelines were the primary sources used, but when no guidelines were available, guidelines used by the EPA were secondary. The raw results of both water and sediment quality sampling can be found in Appendix A.

Table 7 – Water concentrations (mg/L) of inorganic metals at three different sites at both Fuller (Site 1) and Ewen (Sites 2 and 3) reservoirs in ANWA. Samples were collected in August, 2018. These are point collections with n = 1 at each site. The metals that exceeded guidelines are bolded.

Analyte	Site 1 Fuller (mg/L) n=1	Site 2 Ewen (mg/L) n=1	Site 3 Ewen (mg/L) n=1	Water Quality Guidelines (mg/L)
Aluminum (Al)-Total	0.433	0.4	0.374	N/A ¹
Antimony (Sb)-Total	<0.0010	0.00075	0.0007	N/A ²
Arsenic (As)-Total	0.0054	0.0048	0.0044	0.005³
Barium (Ba)-Total	0.166	0.086	0.091	1 ⁴
Boron (B)-Total	0.64	0.382	0.391	1.2 ⁵
Cadmium (Cd)-Total	<0.000050	0.000034	<0.000025	0.00037 ⁶
Calcium (Ca)-Total	50.1	49.1	50.2	*related to alkalinity
Chromium (Cr)-Total	0.0011	0.00081	0.0007	N/A ⁷
Cobalt (Co)-Total	<0.0010	0.00057	0.00057	0.11 ⁴
Iron (Fe)-Total	0.85	0.635	0.687	1 ⁴
Lead (Pb)-Total	0.00058	0.00053	0.00049	0.007 ¹³
Lithium (Li)-Total	0.011	0.009	0.0092	N/A ⁸
Magnesium (Mg)-Total	195	107	105	*related to alkalinity
Manganese (Mn)-Total	0.35	0.16	0.17	*Hardness dependent
Molybdenum (Mo)-Total	0.0016	0.0020	0.0022	1 ⁴
Nickel (Ni)-Total	<0.0050	0.0032	0.003	0.15 ¹¹
Phosphorus (P)-Total	<0.50	0.29	0.26	0.005-0.015¹²
Potassium (K)-Total	56.3	30.9	31	N/A ⁸
Rubidium (Rb)-Total	0.0083	0.0073	0.0069	N/A ⁸
Selenium (Se)-Total	<0.00050	0.00025	0.00029	0.002 ⁴
Silicon (Si)-Total	5	3.06	2.92	N/A ⁸
Sodium (Na)-Total	1560	878	884	N/A ⁸
Strontium (Sr)-Total	0.94	0.74	0.74	1.5 ⁹
Sulfur (S)-Total	27.3	33.1	31.8	N/A ¹⁰
Titanium (Ti)-Total	0.019	0.018	0.014	N/A ⁸
Uranium (U)-Total	0.00062	0.00086	0.00081	0.0085 ⁴

Vanadium (V)-Total	0.0091	0.0089	0.0084	0.02 ⁹
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¹Aluminum guidelines are based off dissolved Al, not total Al in CCME.

²Antimony guidelines based off the Sb (III) oxidation state, not total Sb in CCME.

³CCME, 2001.

⁴MOE, 2018. The phosphorous range is based from aquatic lakes with salmonids as predominant species.

⁵CCME, 2009.

⁶CCME, 2014. This guideline is based on a maximum hardness of 360 mg/L CaCO₃, therefore a site specific assessment is needed because of the high hardness level found at ANWA. This guideline may not be an accurate reflection of the chronic toxicity.

⁷Chromium guidelines based off Cr (III) and Cr (VI) oxidation states, not total Cr.

⁸There are no water quality guidelines.

⁹TNRCC, 2001.

¹⁰Guidelines based off dissolved sulphate or sulphide in CCME.

¹¹CCME, 1999f. This guideline is based on a maximum hardness of 180 mg/L CaCO₃, therefore a site specific assessment is needed because of the high hardness level found at ANWA. This guideline may not be an accurate reflection of the chronic toxicity.

¹² MOE, 2018. This range is based off aquatic lakes with salmonids as predominant species.

¹³CCME, 1999g. This guideline is based on a maximum hardness of 180 mg/L CaCO₃, therefore a site specific assessment is needed because of the high hardness level found at ANWA. This guideline may not be an accurate reflection of the chronic toxicity.

Table 8 – Sediment concentrations (mg/kg) of inorganic metals in the sediment at both reservoirs: Fuller (Site 1) and Ewen (Site 2) in ANWA. Samples were collected in August, 2018. These are point collections with n = 3 at each site. The metals that exceeded guidelines are bolded.

Analyte	Site 1 Fuller (mg/kg) n=3	Site 2 Ewen (mg/kg) n=3	Sediment quality guidelines (mg/kg)
Aluminum (Al)	16166.7	8653.3	N/A ¹
Antimony (Sb)	0.5	0.2	N/A ²
Arsenic (As)	7.6	3.3	5.9³
Barium (Ba)	147.0	37.2	N/A ²
Beryllium (Be)	0.4	0.2	N/A ²
Boron (B)	12.7		N/A ²
Cadmium (Cd)	0.3	0.1	0.6 ⁴
Calcium (Ca)	13466.7	4430.0	N/A ²
Chromium (Cr)	40.8	26.6	37.3⁵
Cobalt (Co)	12.5	8.2	50 ⁶
Copper (Cu)	43.2	11.7	35.7⁷
Iron (Fe)	33700.0	18766.7	20000⁸
Lead (Pb)	16.3	3.4	35 ⁹
Lithium (Li)	18.8	8.2	N/A ¹
Magnesium (Mg)	10966.7	6966.7	N/A ²
Manganese (Mn)	839.3	416.3	460⁸
Mercury (Hg)	0.1	0.0	0.17 ¹⁰
Molybdenum (Mo)	1.5	0.2	N/A ²
Nickel (Ni)	42.1	29.1	16⁸
Phosphorus (P)	1073.7	430.0	600⁸
Potassium (K)	1886.7	616.7	N/A ²
Selenium (Se)	0.4		N/A ²
Silver (Ag)	0.1		0.5 ⁶
Sodium (Na)	9753.3	620.0	N/A ²
Strontium (Sr)	136.7	23.1	N/A ²
Sulfur (S)	12733.3	1766.7	N/A ²

Thallium (Tl)	0.1		N/A ²
Titanium (Ti)	887.7	814.0	N/A ²
Uranium (U)	0.8	0.2	N/A ²
Vanadium (V)	55.7	40.7	N/A ²
Zinc (Zn)	85.8	37.3	123 ¹¹
Zirconium (Zr)	5.8	4.9	N/A ²

¹No sediment quality guidelines; values should be normalized to reference sites (CCME, 1999c).

²There are no sediment quality guidelines in CCME or USEPA.

³CCME, 1998a.

⁴CCME, 1997a.

⁵CCME, 1998b.

⁶TNRCC, 2001.

⁷CCME, 1998c.

⁸MEEQ, 1993.

⁹CCME, 1998d.

¹⁰CCME, 1997b.

¹¹CCME, 1998e.

Pesticides

Pesticide water and sediment data were collected from Fuller and Ewen reservoirs at ANWA in August 2018. These were point collections with one sample per reservoir. There were 8 pesticides detected out of 40 that were measured in the water between the two sites, including hexachlorobutadiene (HCBd), hexachlorobenzene (HCB), Aldrin, dieldrin, 2,4'-DDE, 4,4'DDE, endrin aldehyde, and mirex (Table 8). There were 15 pesticides detected out of 61 measured in the sediment samples, including HCBd, 1,2,3,4-tetrachlorobenzene, 1,2,3,4-tetrachlorobenzene, pentachlorobenzene, HCB, pentachloroanisole, pentachloronitrobenzene, heptachlor, aldrin, chlorpyrifos, trans-nonachlor, dieldrin, 4,4'DDE, 4,4'DDD, and endrin aldehyde (Table 9). The raw results from the sampling period can be found in Appendix A.

There is very limited pesticide data in the BC Approved and Working Water Quality guidelines, therefore Canadian Council of Ministers of the Environment (CCME)

guidelines and guidelines used by the Environmental Protection Agency in the USA were the secondary source. The sediment sample pesticide concentrations at each site are the average of a triplicate sample. No pesticides exceeded any water or sediment quality guidelines. Raw data tables from ALS are provided in Appendix A.

Table 9 - Pesticide concentrations in water (ng/L) in both Fuller and Ewen reservoirs at ANWA, Delta, BC, Canada. These are single point collections that were sampled in August, 2018.

Analyte	Site 1 Fuller (ng/L) n=1	Site 2 Ewen (ng/L) n=1	Water Quality Guidelines (ng/L)
Hexachlorobutadiene (HCBd)	0.0476	0.0556	1300 ¹
Hexachlorobenzene (HCB)		0.111	3680 ²
Aldrin	0.00952		300 ³
Dieldrin	0.0762		56 ²
2,4'-DDE	0.0381		N/A
4,4'-DDE		0.278	10500 ²
Endrin Aldehyde	0.0667		1,210,000 ³
Mirex	0.0381	0.0556	1 ²

¹ CCME, 1999b.

² USEPA, 2002.

³ TNRCC, 2001.

Table 10 - Pesticide concentrations in sediment (ng/kg) in both Fuller and Ewen reservoirs at ANWA, Delta, BC, Canada. These concentrations are based off the average of three samples collected in August, 2018.

Analyte	Site 1 Fuller (ng/kg) n=3	Site 2 Ewen (ng/kg) n=3	Sediment Quality Guidelines (ng/kg)
Hexachlorobutadiene (HCBd)	15.0	3.27	26,500 ¹
1,2,3,4-Tetrachlorobenzene (1,2,3,4-TeCB)	16.6	BD*	N/A ²
Pentachlorobenzene (PeCB)	23.97	5.44	24,000 ¹
Hexachlorobenzene (HCB)	37.5	7.84	20,000 ³
Pentachloroanisole (PCA)	28.8	6.35	N/A ²
Pentachloronitrobenzene (PCNB)	BD*	16.2	N/A ²
Heptachlor	BD*	3.47	600 ¹
Aldrin	BD*	1.24	2000 ³
Chlorpyrifos	105	14.6	N/A ²
trans-Nonachlor	34.4	BD*	N/A ²
Dieldrin	43.2	5.15	2850 ⁴
4,4'-DDE	749.3	56.6	1420 ⁴
4,4'-DDD	172.95	14.14	3540 ⁴
Endrin Aldehyde	BD*	2.57	480,000 ¹

*BDL (below detection).

¹TNRCC, 2001.

²No sediment quality guidelines have been derived for this pesticide.

³MEEQ, 1993.

⁴CCME, 1998.

Turtle Trapping

After 9 days of turtle trapping, 1 RES, 4 pumpkinseed, and 4 carp were caught in the D-nets. The RES was weighed, measured, and euthanised. If there is further funding, it can be analysed in the lab to determine body burden concentrations.

Risk Assessment

The contaminants of potential concern for the ecological risk assessment were the metals and pesticides detected in water and sediments in Fuller reservoir since the majority of the exceedances of those that had formal guidelines were within the waterbody. Contaminants of concern that were carried forward in the risk assessment were those that exceeded water and sediment quality guidelines. These include arsenic in water, and arsenic, chromium, copper, iron, manganese, and nickel in sediments. A concentration based exposure was used for lower trophic species that interact with the contaminants directly, while a dose based exposure scenario was used for higher trophic level species that interact with contaminants indirectly (EC, 2012). The risk assessment results indicate that there is a low level of risk from copper, manganese, and nickel for benthic invertebrates, which exceeds the lines of evidence criteria of an HQ<1 (Table 10). There is negligible risk for the fish communities and for the WPT at Fuller reservoir. The receptor characteristics, exposure calculations and values, and hazard quotient calculation can all be found in Appendix B.

Table 11 - Hazard quotient characterization for the different receptors. Those that are considered high risk are bolded. As fish only had exposure to arsenic in water, no other contaminants were considered. Iron was dropped because of limited CCME and USEPA data.

COPC	As	Cr	Cu	Fe	Mn	Ni
Benthic communities	0.89	0.91	2.20		4.85	1.24
Fish communities	0.18					
Painted turtle	0.00024	0.0054	0.011		0.0021	0.00017

Discussion

The main goals of this study were to assess water and sediment metal and select pesticides contaminant levels, the suitability of ANWA as a future site for reintroduction of WPT, and offer ecological restoration solutions if needed. The two main reservoirs at ANWA examined in this study, Ewen and Fuller, appear to be hyper-eutrophic and brackish with some metals exceeding water and sediment environmental guidelines. Specifically, in Fuller high arsenic exceeding water quality guidelines was observed while several metals exceeded guidelines in the sediment (arsenic, chromium, copper, manganese, iron, and nickel). In Ewen reservoir within ANWA the metal profile was different with only high nickel measured in the sediments exceeding environmental quality guidelines, and no elevations in metals in the water column. Furthermore, although measureable levels of several organochlorine pesticides and one organophosphorus pesticide in water and sediment were observed in this study, there were no guideline exceedances. However, although some of the pesticides measured are not current use pesticides and even some that are current use pesticides, there were few government (provincial or federal) guidelines available for these contaminants. Collectively, this study suggests that there is low level metal and pesticide contamination in these reservoirs at ANWA that would result in chronic exposure scenarios for organisms of this area; the adverse effects of which are poorly understood for most wildlife, including a long lived reptile such as WPT. The risk assessment indicated that there is a high risk for lower trophic species such as plants and benthic invertebrates, but a low risk for WPT. Furthermore, the water and sediment habitat in these reservoirs can be generally considered as poor for the whole ecosystem, especially at lower trophic levels, and this is likely in large part due to the historical and current agricultural uses of this land. These agricultural influences combined with additional future salt water intrusion in to the system with sea levels projected to rise one metre in the next century, are cause for concern regarding acceptable habitat for future WPT introductions as well as other wildlife inhabiting ANWA.

Water Quality at ANWA

The low dissolved oxygen at the bottom of Ewen and Fuller reservoirs in the ANWA, oversaturation at the surface, pH profile and elevations in total phosphorus is

indicative of a eutrophic waterway (CCME, 2004). Based on the water temperatures measured at ANWA in this study, it was predicted that the dissolved oxygen in the reservoir would be closer to 8-10 mg/L rather than maximum saturation of 18 mg/L midday of June. However, upon factoring the measured salinity at ANWA, the expected DO would be lower than the predicted 8-10 mg/L since brackish water cannot hold as much dissolved oxygen as freshwater. Nonetheless, it is likely that there is an overabundance of photosynthetic activity at the surface that is causing DO levels to oversaturate, and then as is characteristic in eutrophic waterbodies, considerable decrease at the sediment-water interface from bacterial respiration and oxidation from organic matter throughout the water column (CCME, 1999; Reddy and DeLaune, 2008). In turn, the removal of CO₂ in water during photosynthesis is likely responsible for an increase in the pH of the water column in Ewen and Fuller reservoirs in the present study, which was evidenced by measured values as high as a pH of 9 during the day. Conversely, the low pH in the water column at deeper depths is likely attributed to bacteria at the bottom of the reservoirs in the sediments releasing acidic by-products into the water column during decomposition of organic matter, resulting in the low pH observed at 1.5 m, 2.5 m, and 4.75 m.

Photosynthetic activity of aquatic plants and algae are dependent not only on pH, DO, but also phosphorus which is often the limiting nutrient for algal growth. According to the CCME (2004), concentrations of total phosphorous (TP) in a waterbody over 0.1 mg/L are hyper-eutrophic. All TP water concentrations at ANWA were found to be higher than this 0.1 mg/L trigger value which indicates that this could be an environmental problem due to well-studied attributes instigating the process of eutrophication (CCME, 2004). Phosphorus can be removed from waterbodies by binding onto sediment and settling out at the bottom of reservoirs and the amount of phosphorus in sediments in Fuller reservoir in particular, were found to exceed sediment guidelines for the protection of the majority of benthic organisms (MEEQ, 1993). This settling out of phosphorus into sediment can remove the immediate availability of this nutrient, but also has long term implications in a reservoir, even if external loading of overland nutrients stops. The process of phosphorous settling out and resuspending in waterbodies is regulated by oxidation-reduction (redox) reactions, the metabolic characteristics of bacteria and fungi, water temperature, dissolved oxygen, pH, and turbulence of the sediments from biota or human activities (CCME, 2004). As ANWA is a shallow unstratified waterbody (warm

water polymictic), sediments can also be resuspended by wind (Luettich et al., 1990). Further, bacterial metabolism in wetland sediments can release the inorganic phosphate bound in sediments back to the water column, increasing the internal phosphorus load in the system (Carlton and Wetzel, 1988). This internal loading from sediments can exceed the external loading and produce a positive feedback cycle even if external loading is reduced. It is generally understood that anoxic conditions at the sediment and water interface and lowering oxidation-reduction potential can release phosphate bound to hydrous oxides (Carlton and Wetzel, 1988). As well, a steady supply of organic matter that is easily decomposed can cause reducing conditions. It is typical in these systems to see a diel pattern, with oxygen concentrations drastically dropping at night because of the high respiratory demand of microorganisms (Reddy and DeLaune, 2008). This most likely occurs at ANWA following the similar fashion of other shallow hypereutrophic reservoirs. Thus, the concern at Fuller reservoir is that the system has been overloaded with phosphorus to the extent that it will be a self sustaining cycle that will continuously affect the dissolved oxygen and pH, increasing the risk for unpredictable anoxic conditions, increasing organic matter content and sedimentation rates, increasing turbidity, increasing the potential for cyanobacteria blooms, and reducing ecologically sensitive species in favour of more tolerant ones (EC, 2004).

Reduced oxygen and nutrients (i.e. P, C, N) and can have lethal or a variety of sublethal effects on aquatic wildlife unless an aquatic organism has specific adaptations to tolerate such deficiencies in the ambient environment (CCME, 1999a). For example, Northern subspecies of freshwater painted turtles (*Chrysemys picta bellii*) are known to have a tolerance for surviving metabolic acidosis in anoxic conditions (Jackson, Herbert, and Ultsch, 1984; Rollinson, Tattersall, and Brooks, 2008; Warren and Jackson, D. C., 2004). Western painted turtles demonstrated the ability to exchange O₂, CO₂, and water with the surrounding environment, relying on their own energy reserves largely from their shell (Jackson, 2000). Anoxic overwintering conditions have been found to have a large effect on skeletal magnesium and carbonates with little effect on body stores of calcium and phosphates in freshwater turtles (Warburton and Jackson, 1995). Freshwater turtles can replenish carbonates with metabolic CO₂ processes while magnesium is obtained through their diet (Warburton and Jackson, 1995). While northern subspecies of painted turtles have evolutionary adaptations to anoxic conditions largely because of overwintering in often ice covered ponds and lakes (Ultsch, 2006), other organisms throughout the

food web such as benthic invertebrates, zooplankton, and fish can be harmed and even killed in stressful anoxic conditions. In freshwater, the guideline for lowest acceptable dissolved oxygen in cold water has a guideline of 9.5 mg/L for early life stages and 6.5 for other life stages for cold water biota (CCME, 1999a). The large variability in oxygen levels daily, seasonally, and yearly can induce chronic stress and be harmful to long-term survival of different species living in waterbodies (CCME, 1999a), and this is likely the scenario in both Ewen and Fuller reservoirs in the ANWA. It is hypothesized that these conditions would reduce the food types and amount for future introduced WPT in ANWA since this study strongly suggests that the reservoir chemistry is unstable and sub-optimal for benthic invertebrates and other smaller aquatic organisms that are not adapted for anoxic conditions.

Another potential stressor for WPT at ANWA is the brackish water in the water column. The range for brackish water 1,301 to 28,800 $\mu\text{S}/\text{cm}$ (Li and Migliaccio, 2011) and both reservoirs fall into this range at all depths. Fuller reservoir is generally more brackish with higher conductivity readings at all depths than Ewen reservoir. Sodium, chloride, calcium, and magnesium are all ions that can conduct electricity, and as both Fuller and Ewen are brackish with very high total hardness, the conductivity reflects both of those inputs. At ANWA, values exceed 500 mg/L CaCO_3 in both reservoirs and are categorized as very hard according to Durfor and Becker (1964; >180 mg/L). The brackish water and higher salinity content in this area could be from a few different factors. As this was Fraser tidal marshlands before it was diked, there could be residue salt in sediments and the land. These reservoirs have been dredged a few times, so other possibilities are that there are small amounts of brackish water entering the reservoirs when they fill them every spring from the Fraser River freshet, or that there is salt water intrusion through the diking system and brackish water is breaching the reservoirs through a weakness during certain times of the year as the diking system is not up to code (pers. comm). Red eared sliders currently live within Fuller and Ewen reservoir, indicating that it is liveable for turtles, but there may be some long term risks at ANWA with projected sea level rise and further increases in salinity based on studies in turtles.

Agha et al., (2018) found that 30% of coastal freshwater turtle species have been reported in brackish water ecosystems with their phylogeny, behavioural, and life-history traits enabling them to tolerate various salinities over short time frames. However, this is

highly variable for different freshwater turtle species including WPT. Painted turtles (*Chrysemys picta*) were found to have a mean loss of 1.80% mass loss when exposed to 35 ppt saltwater (100% seawater) (Dunson, 1986). Other turtles within the family Emydidae have mass losses ranging from 0.3% - 2.4% with 35 ppt saltwater (Agha et al., 2018). Water loss in salt water is inversely proportional to body size with larger turtles having a better tolerance to varying salinity levels (Dunson, 1986), thus the impacts of higher salinity in WPT may be high, particularly in juveniles. Furthermore, with a rise in mean sea level of at least 1 m, ~90% of coastal turtle species, or those that inhabit brackish water, may be vulnerable to saltwater intrusion into their habitats (Agha et al., 2018; Jevrejeva et al., 2012; Rasmussen et al., 2011). In addition, Red eared sliders (RES) (*Trachemys scripta elegans*) also exhibit some tolerance to brackish water opportunistically since the pet trade has drastically expanded their range to areas they normally would not inhabit; a feral population of RES was found among both freshwater and brackish ponds in Bermuda (Outerbridge, 2008). Ultimately, either WPT an introduced WPT would have to physiologically adapt or migrate away from increased salinity in their ambient water, the latter resulting in reduced habitat. This could be of concern in an introduction area like ANWA which is isolated as an island in the Fraser River. Though the effects are unknown for most freshwater North American turtle species, it is possible that most of these species, including the WPT may undergo physiological challenges where there is increased salinization (Bower et al., 2016). It was observed that the eastern painted turtle (*Chrysemys picta picta*) had occasionally inhabited brackish tidal waters, was found with rusty deposits on the shell in a salt marsh, but the amount of time spent the species spent in these areas are unknown (Pope, 1967). This salt water tolerance merits further research since WPT are native to coastal habitats; thus, it is possible WPT may have unique adaptations to deal with increased salinities at different life history stages (Agha et al., 2018).

As a whole, the water quality is poor within the two main reservoirs at ANWA with high nutrient loads undoubtedly affecting the internal recycling of phosphorous as well as both the pH and dissolved oxygen in the water column to some extent. Furthermore, the high salinity content makes these reservoirs more brackish than freshwater. These cumulative stressors are likely to change the species composition from those normally present in freshwater wetlands to those that can tolerate large diurnal changes in chemistry. Whether the current invasive species present in ANWA are more tolerant to

eutrophication is not well studied, currently the combined effects of these water quality parameters does appear to support numerous invasive fish species and red eared sliders (*Trachemys scripta*; RES). With respect to the RES, data describing the tolerance for RES to anoxic conditions is sparse, but one study suggested less tolerance for anoxic conditions, with juveniles at more risk than adults (Ultsch, 2006). There have been no population surveys of RES done since Kilburn and Mitchell (2011) at ANWA, but this may merit further studies to determine changes in populations. It is likely that WPT would survive in this area short term, but the threat of dike breach and further salinization of this water may indicate a poor quality of life.

Metal Contamination at ANWA

Within Fuller reservoir arsenic was the only metal found to exceed water quality guidelines while arsenic, copper, manganese, iron, chromium, and nickel were found to exceed sediment quality guidelines. This metal profile was different compared to Ewen reservoir where only nickel exceedances were observed in the sediment, and none in the water. Interestingly, sediment has a great adsorption capability for heavy metals, thus it is not surprising to see high metal concentrations in sediments (Ivask et al., 2002). Sediment accumulations of heavy metals can be harmful for benthic invertebrates and other benthic microbial organisms, affecting organic matter recycling (Schwarz et al., 2007), pollutant biotransformation and degradation, and biomass production (Ahmed et al., 2018). In waterbodies, heavy metals can change the dynamic nature of wetlands and interfere with natural recycling processes, and together, finding water and sediment contamination by heavy metals further decreases the habitat quality at ANWA. While many heavy metals (i.e. copper, iron) are essential micronutrients at low levels, most are pollutants at high levels in the environment. However, there is little evidence to date for a arsenic and nickel as essential micronutrients exists, thus elevations of such metals above natural background levels is often a contaminant of concern for biota. Additional characteristics of waterbodies are known to modify the potential toxicity of heavy metals and should be considered when predicting site specific toxicity of metals. Of particular importance at ANWA is the high total hardness in the reservoirs that can act as a buffer for some metal toxicity (including Cu, Mn, and Ni) by forming insoluble complexes with other elements (Pascoe et al., 1986). However, the arsenic found in concentrations above water and sediment guidelines in ANWA is one metal with toxic effects that is

unaffected by hardness (CCME, 2001). One obvious and likely source of metals and nutrients in the waters and sediments at ANWA is the surrounding agricultural lands and associated pesticide and fertilizer use. Although pesticides and fertilizers often have unknown components for proprietary reasons, these chemicals have frequently been shown to have metals in addition to various nutrients (Ashley and Stockner, 2003; Bradl, 2005; He et al., 2005; Jiao et al., 2012; Karstens et al 2016). Regardless of land applications, there can be runoff into nearby waters and to wetlands, typically the lowest points in the landscape.

In Fuller reservoir, the speciation and mobility of arsenic from water to sediment is likely different depending on the environmental conditions within the various areas of this reservoir. The two main species of inorganic arsenic are arsenite (III) and arsenate (V), both which readily enter cells, but As(III) is more mobile and toxic to aquatic organisms (Bodwell et al., 2003; Bradl et al., 2005). Arsenobetaine is an organic form that is also taken up by wildlife (Kunito et al., 2008). Under oxygenated conditions, As(III) is oxidized to As(V) (Liber et al., 2011) but in a reducing environment with low pH like at the deepest point in Fuller that is permanently anoxic, As(III) ions could form. In general, As does not tend to biomagnify up the food chain, but different As species have high affinities for proteins, lipids, and other cellular components and can bioaccumulate in living organisms to some extent (Bradl et al., 2005; Spehar et al., 1980). Indeed, bioaccumulation factors vary among species, and background concentrations as low as 0.000113-0.00037mg/L As can have BAF of 265 for pumpkinseed (*Leponis gibbosus*) (Chen and Folt, 2000), 541 and 270 for small and large brooktrout (*Salvelinus fontinalis*) respectively (Mason et al., 2001), while at a concentration at 0.008 mg/L, carp (*Cyprinus carpio*) were found to have a BAF of 13 and 25 (Baker and King, 1994). These bioaccumulation factors do not exceed the BAF>5000 criteria for categorizing a contaminant as bioaccumulative in the Canadian Environmental Protection Act (CEPA, 1999). This low tendency to bioaccumulate and biomagnify in fish is further supported by observations that the largest uptake of arsenic is by phytoplankton with decreasing amounts up higher trophic levels (Kunito et al., 2008). Specifically, Kunito et al (2008) showed that marine phytoplankton bioconcentrated arsenic 1,000-50,000 times the concentration of surrounding seawater that has a background concentration of 0.002-0.003 mg As/L. Both Fuller and Ewen reservoirs have background concentrations above this average level. Kunito et al. (2008) used for seawater, though Fuller exceeds the

0.005 mg/L water quality guidelines while Ewen arsenic concentrations are just below it. However, there appears to be some controversy or uncertainty surrounding As bioaccumulation based on more recent studies in different species in the upper levels of the food chain. Specifically, including harp seal (*Pagophilus groenlandicus*), pilot whale (*Globicephala melas*), hawksbill turtle (*Eretmochelys imbricate*), and green turtle (*Chelonia mydas*), to name a few, demonstrated that As tissue concentrations were largely dependent upon diet and life history and not related to the trophic position (Kunito et al., 2008).

The high levels of As in the Fuller reservoir water and sediments at ANWA is a concern because of its known multiple modes of toxic action in animals. Arsenic at low levels can be carcinogenic, cause cardiovascular and pulmonary diseases, impair neurological and developmental functions (Bates et al., 1992; Tian et al., 2001), and is observed to be an endocrine disruptor in many vertebrate taxa (Bodwell et al., 2003; Kunito et al., 2008; Shaw et al., 2006). As well, there are synergistic and potentiating effects of arsenic in tandem with other toxic and carcinogenic stressors in the environment (Kaltreider et al., 2001; Shaw et al., 2006). While also causing severe adverse effects with acute exposure (Liber et al., 2011), arsenic also can impair cellular function at very low concentrations. As (III) was found to cause oxidative DNA damage at 0.000075 mg/L in cultured human cells (*Homo sapiens*) (Schwerdtle et al., 2003) and at concentrations of ~0.001- 0.1 mg/L were found to impair different hormone functions in rat cells (*Rattus spp*) (Bodwell et al., 2003; Kaltreider et al., 2001; Shaw et al., 2006). In three different marine turtle species (*Chelonia mydas*, *Caretta caretta*, and *Eretmochelys imbricate*) Saeki et al. (2000) found As(V) levels were higher in muscle tissue than in liver, but As(III) had higher concentrations in liver than in the muscle tissue, with the ratio rising in relation to the turtle's weight. It was hypothesized that the turtles had exposure through the food chain consuming a large proportion of bottom-dwellers and through absorption of sea water which has a background concentration of 0.002-0.003 mg As/L (Saeki et al. 2000; Storelli et al., 2000). Saeki et al. (2000) did not know what the sensitivity of the species were to As(III) toxicity, but presumed metabolic processes within the liver could be altered (Saeki et al. 2000; Storelli et al., 2000). Within Fuller reservoir, chronic exposure from As is the primary concern for a long lived species such as WPT and their different food sources which includes different benthic organisms. Long lived species such as turtles can bioaccumulate metals in their tissues,

causing long term cytotoxicity or endocrine disruption (Tan et al., 2010); in particular, Cu and Fe were found predominantly in the liver in sea turtles, Ni was more concentrated in the kidneys while Cr was found within the lungs (Gardner et al., 2006; Tan et al., 2010). One study found no differences between sex, and age to explain variation in metal concentrations of As, Cr, Cu (among others) in tissues of box turtles (*Terrapene carolina carolina*), but they did note that concentrations are expected to be lower in females than males due to females offloading metals in eggs (Allender et al., 2015). Additional studies are recommended to investigate more areas within Fuller and Ewen reservoirs in light of the many low level, chronic exposure adverse effects of As on multiple biological systems in vertebrates. This would aid in better understanding how prevalent this contaminant is and the range of concentrations of As that exists in ANWA and how these compare to toxic concentrations in vertebrates.

Another metal in this study that was high in the Fuller reservoir at a concentration known to impact biota was Cu in sediment (43 mg/kg). Within sediment, reports of levels starting at 33 mg/kg Cu have been shown to reduce the ability of bacteria to use carbon sources and can alter community structure of heterotrophic bacteria and reduce the density (Ahmed et al., 2018; Zhao et al., 2014). Copper has a greater toxicity at a lower hardness (Liber et al., 2011) that the water quality guideline factors in up to a certain concentration, and this resulted in the water concentrations of Cu not exceeding guidelines in Fuller and Ewen reservoirs suggesting that adverse effects of Cu at ANWA would impact sediment dwelling or benthic organisms in contact with sediment or the sediment-water interface. This is of consideration for many benthic invertebrates and thus the base of the ANWA food chain and potentially a concern for WPT that bury into sediments during brumation in the winter season.

Total chromium exceeded guidelines in the sediment in Fuller reservoir, but there are no guidelines for total Cr in the water, and further water quality testing is recommended to determine what species of Cr are present in ANWA. The two relevant species of chromium that determine its potential toxicity to biota are Cr(III) and Cr(VI). Cr(VI) is toxic in the environment while Cr(III) is essential in humans and animals before becoming toxic beyond a small threshold (Bradl et al., 2005). The cumulative effects of Cr toxicity can be seen from molecules to the ecosystem level (Freitas and Rocha, 2011). Cr can negatively affect population growth rates, decrease reproduction and survival of individual species (such as in different species of zooplankton and algae) by

interfering with the biochemical activity of certain enzymes (Freitas and Rocha, 2011). Cr(VI) is the stable speciation of chromium and is dominant in surface waters and oxygenated sediments while Cr(III) is dominant in reducing environments (CCME, 1999d) indicating that Cr(III) may be more present within Fuller as there is little oxygen within the sediment-water interface.

Nickel was the only metal to exceed guidelines in both Ewen and Fuller. Nickel is a heavy metal with hazardous properties depending on its oxidation state, with soluble nickel compounds more toxic than insoluble compounds (Bradl et al., 2005). In humans, effects of nickel can cause gastrointestinal distress, pulmonary fibrosis, renal edema, and skin dermatitis (Akhtar et al., 2004). However, there is limited data about whether species at higher trophic levels are more sensitive to Ni than lower trophic levels, nor is there enough data available to determine if Ni biomagnifies in organisms (DeForest et al., 2011). Nonetheless, it is known that nickel can disrupt DNA repair and affects epigenetic histone modification, thus has high potential for harm in plants and animals (Goodson et al., 2015). With different water concentrations of 0.01, 0.1, and 1 mg/L, marine copepods (*Tigriopus japonicas*, *Apocyclops borneoensis*, and *Acartia pacificia*) egg production and hatchling success were reduced, with more detrimental effects with a higher concentration (Mohammed et al., 2010). The sediments in the reservoirs present a risk for overwintering turtles, the benthic communities and food sources for turtles, and other wildlife species present at ANWA.

Metal complexes and associated toxicity are poorly understood and complicated, and present considerable uncertainty for estimating the toxic effects of the mixtures of metals observed in the water and sediment at ANWA. For example, the speciation of As(V) is more common in aquatic environments due to the complexes As can form with Ca, Fe and Mn₃ (Bradl et al., 2005). Arsenic and iron are coupled in nature, with iron oxyhydroxides within the water affecting the speciation of As, though this depends on a pH of around 7 or slightly under (Lizama et al., 2011). As well there are competing ions (phosphate, sulphate, carbonate, bicarbonate, and chloride) which can desorb As(V) from sediments and replace it (Lizama et al., 2011). Fuller has all these ions within the water and sediment, with a high diel variance in pH, which means there could be complex speciation occurring. Further, iron becomes mobile in reducing environments in a dissolved form and can precipitate in oxidized conditions. Iron is an essential micronutrient in wetlands for plants and wildlife, but can be harmful above trace levels.

Too much iron uptake can impair liver functions, have some endocrine disruptions, and cause cardiovascular effects in humans (Syakalima, 2000). Manganese within the same study by Kalisinska et al., 2004 found that the highest manganese concentrations were found in the bones and the brain. Manganese is essential in metabolic processes in wildlife and plants, but excesses of the mobile form of Mn are known to be a neurotoxin since it can affect the central nervous system (Oweson and Hernroth, 2009). During reducing conditions, Mn, which usually forms a speciation with oxygen among the sediments, is converted to its bioavailable form (Oweson and Hernroth, 2009). Excessive Mn exposure can lead to behavioural changes, motor disturbances, and altered cognitive function as shown in laboratory studies on rats (*Rattus spp.*) (Santos et al., 2012). Similar to copper, Mn is buffered by total hardness to an extent, so there is a reduced risk within Fuller reservoir. Overall, the metal concentrations in Fuller and Ewen reservoirs do pose some concern for the reproduction and development of juvenile and adult WPT if they were to be introduced into this area. However, future studies with expanded sampling areas and analyzing different metals species and complexes would assist in better understanding the extent of the metals.

Pesticides at ANWA

A suite of organochlorine pesticides (OCP), one organophosphate pesticide (OPP; chlorpyrifos), and a phenylurea pesticide (PP; linuron) were examined in the water and sediment samples collected at ANWA in this study. There were between 8-15 organochlorine pesticides (OCPs) detected in the water and sediment and all except one (pentachloronitrobenzene/quintozene; PCNB) are not currently used in Canada. Chlorpyrifos is still a registered pesticide in Canada and was also detected in ANWA in low concentrations (ng/L and ng/kg). None of the OCP, OP, PP pesticides detected exceeded any water or sediment quality guidelines, and based on assessing each pesticide individually using guidelines it is possible that there may be a relatively low perceived risk for wildlife inhabiting the ANWA. However, there is mounting evidence emerging about low dose effects and non-monotonic dose response curves where there are environmental effects that are observed at levels below traditional toxicology studies (Goodson et al., 2015). The implications of this suggest that the responses observed at higher doses cannot predict the low dose responses, and further, many water and sediment quality guidelines do not take this into account (Vandenburg et al., 2012).

Furthermore, guidelines do not take into account chemical interactions, low level effects, or indirect toxicity effects (Goodson et al., 2015; Vandenburg et al., 2012). As a result, there are large knowledge gaps in understanding the effects of low dose chemical mixtures and how pesticide mixtures interact in the environment, including in wildlife. Most OCP detected at ANWA with the exception of pentachloroanisole and PCNB, were added to the List of Toxic Substances in Schedule 1 of the Canadian Environmental Protection Act (CEPA) during the 1990's and 2000's to prevent pollution and protect the environment and human health after the persistent and toxic nature of the pesticides were proven (CEPA, 1999). As a result, detecting some of these OCPs at the ANWA further demonstrates the persistent nature of the chemicals, and it is likely that wildlife inhabiting the ANWA are undergoing low level, chronic exposures to many of these highly toxic OCPs.

The relative measured concentrations of pesticides in the water at Fuller were dieldrin > endrin aldehyde > HCB > 2,4'-DDE > mirex > aldrin. In sediment within Fuller, the order of decreasing concentrations is 4,4'-DDD > 4,4'-DDE > chlorpyrifos > dieldrin > HCB > trans-nonachlor > PCA > PeCB > 1,2,3,4-TeCB > HCB. Historically, aldrin and dieldrin were both synthesized independently, but dieldrin is also a degradation metabolite of aldrin in the environment as aldrin is volatile and readily degrades by photodegradation or from bacteria (Jorgenson, 2001). Dieldrin has been observed to be teratogenic and estrogenic in tadpoles (*Xenopus laevis*) (Moresco et al., 2014; Schuytema et al., 1991; Palmer et al., 1998). Endrin aldehyde is from the breakdown of endrin and does not dissolve in water, but its environmental fate is less understood than with other organochlorines (ATSDR, 1996). DDT and DDE are known endocrine disruptors, with the oestrogen receptor antagonised by DDT and the androgen receptor antagonised by DDE (Kelce et al., 1995). Chlorpyrifos is an organophosphate insecticide still used in Canada that disrupts nerve impulse transmission by inhibiting the enzyme acetylcholinesterase (AChE) through phosphorylation, leading to convulsions, paralysis, and death in invertebrates (CCME, 1999e). Chlorpyrifos is also an endocrine disrupter and can lower serum estrogen and testosterone levels, as observed in fish species *Oreochromis niloticus* (Oruç, 2010). The concentration of 4,4'-DDE was below the sediment quality guidelines, but was the contaminant that came the closest to the guideline out of all the pesticides. Further sediment sampling in the waterbodies and

waterways at ANWA would help determine the spatial variability and the different ranges of OCP pesticides.

Within the water column at Ewen, the concentrations of pesticides are 4,4'-DDE > HCB > HCBd > mirex. There is a higher amount of total OCP and chlorpyrifos in water in Ewen (0.5 ng/L) compared to Fuller (0.27 ng/L), but a higher amount of OCP and chlorpyrifos in sediment in Fuller (1226.72 ng/L) compared to Ewen (136.87 ng/L). There were less pesticides found in the water at Ewen reservoir than at Fuller, but Ewen has higher total concentrations of pesticides in the water. This is an interesting find since Fuller reservoir has a larger amount of total OCP pesticides in the sediments, therefore one possible explanation is that resuspension rates may be influenced by different chemical parameters or bioturbation in the sediments. In Ewen reservoir, concentrations in sediment are 4,4'-DDE > PCNB > chlorpyrifos > 4,4'-DDD > HCB > PCA > PeCB > dieldrin > heptachlor > HCBd > endrin aldehyde. The higher presence of 4,4'-DDE may indicate degradation of DDT by benthic organisms in aerobic riparian areas (Pandit et al., 2002; Zhou et al., 2008; Aly Salem et al., 2014), though environmental conditions such as pH, salinity, and organic matter content also play a role (Pandit et al., 2002). The OCP 4,4'-DDE is more toxic to aquatic organisms than other DDT metabolites because of its environmental persistence and high bioaccumulation potential, especially through the food chain (Hitch and Day, 1992; Wu et al., 2013). This has the potential to be a mode of chronic toxicity for wildlife, especially for those more at risk including neonatal and juveniles where the timing and exposure to EDC in the environment can lead to irreversible development and reproductive alterations (Guillette et al., 2000; Palanza et al., 1999).

Trace OCPs in the environment are positively correlated with soil organic matter content and have an affinity for wetland sediments (Doong et al., 2002; Malik et al., 2009; Tao et al., 2008). It is also hypothesized that sediments can be a secondary emission source of OCP since they degrade slower when bound to sediment than in water or the atmosphere. Both reservoirs have a thick detritus-organic layer (Kilburn and Mitchell, 2011) that provide many binding areas and provide the opportunity for subsequent resuspension in the water column. Similar to metals, benthic communities can be directly affected by contaminants in the sediment, and any subsequent disturbances (mechanical, physicochemical changes) may allow a resuspension of contaminants into the water column, thus resulting in exposure routes via the water as

well (Ahmed et al., 2018). It was observed in a burrowing sea crab (*Chasmagnathus granulata*) that the beds they create are a sink for OCPs because of their habitat preferences for high organic matter and resulting bioturbation when they dig 0.4 m into the sediments (Menone et al., 2004). Yet, there was less bioaccumulation in the crabs when the sediments had high organic matter and clay content since OCPs have a strong affinity to the binding areas, leading Menone et al., (2006) to conclude that the physicochemical characteristics of the habitat may be more important than the concentrations. Conversely, different invertebrate species such as chironomids have been shown to remobilize OCPs in sediment by bioturbation activity (Goedkoop and Peterson, 2003; Menone et al., 2006). This bioturbation is seen by many different species that live in wetlands, including WPT when they overwinter as they are known to bury in sediments, and this could be a source of potential OCP resuspension from sediments. However, there is usually large spatial variability in distribution of contaminants in water and sediments so more extensive pesticide sampling efforts are recommended within ANWA. Two sampling rounds completed in 2009 and 2012 examining the spatial variability of pesticides in the upper water column found traces of other OCPs that were not found in the water or sediment of the single grab samples at Fuller and Ewen reservoirs in the present study. These include 4,4-DDT, alpha-HCH, and endosulfan-sulphate (CWS, unpublished data). It is possible that degradation or movement of the additional OCPs out of Ewen and Fuller reservoirs occurred since the 2009 and 2012 sampling event, or that the present studies design with fewer samples per reservoir was not extensive enough to capture the distribution of OCPs at ANWA. Nonetheless, the present study shows OCP water concentrations that were at similar levels comparing to the 2009 and 2012 sampling events. However, this is the first study to examine pesticides in the sediments at the ANWA and indicates that several OCPs and a current use OPP (chlorpyrifos) were present in both Fuller and Ewen reservoir sediments. This data supports the categorization of many of the OCPs and OPP as persistent, mobile from bioturbation and physicochemical changes, and within the same magnitude ranges for water that were found in this study, though sediment sampling for contaminants has never been completed at ANWA.

A general assumption is that as omnivores, turtles are expected to have a lower ecotoxicological risk than carnivores, but higher foraging rates in warmer weather and foraging in a contaminated food web could indicate that there is more of a risk than

thought (Komoroske et al., 2011). Many OCPs and chlorpyrifos are endocrine disruptors can cause reproductive and developmental abnormalities, as well as skewing sex ratios in many taxa, including turtles (Moss et al., 2009). In the RES (*Trachemys scripta elegans*), Willingham and Crews (1998) noted that eggs incubated at a temperature that would produce a male biased sex ratio were subject to a sex reversal after exposed to different OCP chemicals, with more significant affects when multiple OCP's were present (Willingham and Crews, 1998). Males turtles from different species (*Trachemys scripta troostii* and *Sternotherus odoratus*) were observed to have higher concentrations of OCP than females which is thought to be due to differences in prey selection, maternal offloading to eggs, or sex differences in biotransformation and elimination of different compounds (Moss et al., 2009). Guirlet et al. (2010) found maternal transfer of all OCP, even at low levels, detected in the eggs, with the OCP concentration decreasing in successive clutches of eggs – showing how the mother turtles offload contaminants to juveniles. Van de Merwe (2009) looked at POP concentrations to hatchling mass/length ratios and found that there is a reduction in the size ratio with increasing POP concentrations at low concentrations indicating that there is some combined effects of different POPs (DDT, mirex, HCHs, endosulfan, aldrin, dieldrin, etc) and metals (As, Co, Cu, Zn, etc) on embryonic development. Heavy metals and POPs can also compete for binding sites as dieldrin and numerous heavy metals competed with the natural hormone oestradiol in green turtles (*Chelonia mydas*) while Cu was shown to compete with testosterone (Ikonomopoulou and Bradley, 2009). Within this same study, it was observed that DDT and dieldrin both affected the testosterone binding affinity to the androgen receptor by decreasing or increasing its binding affinity (Ikonomopoulou and Bradley, 2009). Dieldrin also competed for binding sites with oestradiol, indicating the different potentiating affects with mixtures of contaminants (Ikonomopoulou and Bradley, 2009). These studies highlight the risk for juveniles during development through offloading of OCP from adult females, and where there is background concentration of low level pesticides. Since WPT have a long lifespan, there is more of a risk over time of juvenile exposure to endocrine disruptors that would affect their development and reproduction. Future studies examining future tissue/body burdens of pesticides as well as egg pesticide concentrations in invasive turtles at the ANWA (i.e. RES) as surrogates for WPTs to predict the extent of bioaccumulation that coincides with the measured sediment and water concentrations of pesticides in the present study is recommended.

The presence of trace pesticides in the environment can affect the reproduction and development of WPT. There is no universal 'low dose' concentration that can be used for all chemicals and there is no safe threshold of daily exposure to endocrine disrupting chemicals (EDCs), especially during early life exposures (CELA, 2017; Vandenberg et al., 2012). EDCs can produce irreversible effects on development and critical physiological systems at low doses, especially when there are mixtures of the chemicals in the environment (CELA, 2017). This area is a vastly understudied due to the huge amount of synthetic chemicals in the environment, the complex interactions in the environment and in different species, and the difficulty of studying all the different interactions and their effects in the lab that can often produce confounding results (Combarrous, 2017). Though no guidelines were exceeded within Ewen and Fuller reservoirs, there are many unknowns with synthetic chemical interactions in the environment and the long term developmental and reproductive effects in a population.

Risk Assessment and Uncertainty

The results of the preliminary ERA indicate that organisms at the bottom of the food chain are more at risk from the levels of copper, manganese, and nickel than upper trophic levels. The WPT, through a surrogate painted turtle, was determined to not be at risk from the levels of metals found at Fuller reservoir. However, the food sources of the WPT which include benthic invertebrates are at risk from metal exposures and therefore the potential introduction of WPT should be met with caution. The findings of this project extend beyond just WPT that have the potential to be introduced at ANWA. There are numerous wildlife that continue to inhabit the site and restoration should be undertaken to reduce some of the contaminant loads, nutrient loads, and improve the ecological integrity of the ecosystem as a whole.

This ERA has both an overestimation and underestimation of risk and there are a few areas of uncertainty. Since there was a limited dataset, there was an assumption that Fuller reservoir had the same level of metals in water and sediment throughout the reservoir based off a point sample of each. In reality, there could be a large spatial variation or seasonal variation of contaminants on site. There also could be more transport pathways such as movement of contaminants through groundwater, overland flow, or within pore water (EC, 2012). Further, there could be some contaminants of concern not accounted for within the initial sampling that are at high levels in the

sediments and water. As there was no terrestrial soil data, there was a large assumption that the WPT eats only within an aquatic environment which is not the case for turtle species. Not having plant tissue samples to determine the bioconcentration factor in plants also meant that plants were removed from the ERA (EC, 2012). As well, turtle diets are site specific and can vary seasonally, so there is inherent uncertainty in what the turtles eat. All of the values used for WPT are based off adult turtles, and thus it does not account for risk for juveniles. The TRV values had different sources and may have had different derivation methods used, as well for metals that did not have a derived TRV through US EPA, the CCME water or sediment quality guidelines were used. Since reptiles are often understudied and there is limited data in literature, an avian placeholder was used for the TRV for the painted turtle hazard characterisation calculation. Most TRVs are from studies using bioavailable forms of metals than can overestimate actual availability on site, and since there is only total metals available within the data set, it is assumed that the metals are 100% bioavailable (EC, 2012). Some of these uncertainties could be removed with a more robust sampling regime in the sediments and water, along with plant tissue data at ANWA.

Overall further studies and a more robust weight of evidence approach would be beneficial to further characterise the risk based off the measured contaminants. A weight of evidence approach would look at multiple lines of evidence (biological community structure, toxicity, and bioaccumulation) to support the ERA (EC, 2012). Lines of evidence would be derived from different assessment and measurement endpoints including some site specific toxicity tests or looking at community structure (EC, 2012). Lastly, the contaminants have many additive, antagonistic, and synergistic properties with other contaminants in the environment that were not accounted for within this preliminary model. These are complex interactions and there is limited data on chemical properties and interactive effects in the environment with the thousands of different compounds at large.

Restoration Recommendations and Implications for Restoration

Based off the current chemistry data from the water and sediments and the risk for wildlife in the area, ecological restoration should occur at ANWA. This site is not an ideal habitat for the introduction of WPT, but if deemed there are no other options, a freshwater wetland with a nesting beach and basking areas should be created away from the main reservoirs. This would open up a different area of the NWA that possibly has less contamination with more submergent and emergent vegetation as a food source for WPT. More aquatic plants would aid in phytoremediation of the water and sediment if there are any residual pesticide and heavy metals in the area (Guittonny-Philippe et al., 2015). However this only partially addresses the nutrient loading on site, so efforts should be made to reduce the amount of fertilizer used for current land management practices in ANWA. The hyper-eutrophic waters at Fuller and Ewen are most likely in a positive feedback cycle at this stage, so any reduced external loading may not lower nutrient levels and improve water quality.

Since Fuller and Ewen reservoirs have extremely steep gradients, to improve the ecological integrity and help the reservoir function more like a wetland the sides could be recontoured to a gentler slope. This would provide space and a platform for aquatic vegetation to grow with the added benefits of long-term phytoremediation in the sediments and water as wetlands are known to filter out contaminants and nutrients. This would also create more habitat space and food to be used by organisms and wildlife at ANWA. The largest risk is for the lower trophic organisms on the site that interact with the sediments directly, so this may lower the contamination over time. The riparian areas between the fields and the waterbodies and channels at ANWA could also be increased to prevent direct runoff from the land to the water. Lastly, it is worth a suggestion that one or both of the reservoirs could be opened back up to the Fraser River to restore the historic foreshore tidal marshes that used to be there before the 1930s, as this area is not suitable for WPT and could support at risk species in tidal marshes (Thorne et al., 2012). Maintaining the dikes around the reservoirs is costly, may reduce habitat space at ANWA, and further maintain the reservoirs in their current state. ANWA is an ecologically important landscape within the Boundary Bay – Roberts Bank – Sturgeon Bank Important Bird Area and the Fraser River Estuary Western

Hemisphere Shorebird Reserve Network and restoration of the site would be beneficial for all species present.

Future Studies

Future studies should be undertaken to understand the spatial and seasonal distribution of metals and contaminants in the sediments, water, and soil. These results could inform a more robust ecological risk assessment, along with further studies about the body burden concentrations in RES on site as a receptor species. With the high phosphorus loading, there should be further studies to see if there is still an external load to the system from the agricultural fields around Fuller and Ewen or if the phosphorus is primarily from the self-potentiating internal loading. Furthermore, an assessment of the benthic invertebrate diversity at ANWA should be completed to determine what species are currently present and how this changes over time as benthics are a good indication of contamination. Invasive species monitoring in both Ewen and Fuller reservoirs would also be beneficial to look at both RES and fish species population trends over time.

Conclusion

There are many different variables when considering if this area is a suitable site for the introduction of WPT. A few of the potential stressors in Fuller and Ewen reservoirs identified in this report include high nutrient loads and the subsequent effects on water chemistry, brackish waters and the risk of increased salinity with sea level rise, and the levels of heavy metals and pesticides in the water and sediment. Beyond interactions with the chemistry on WPT development and reproduction, there are other important factors to consider related to the life history requirements of WPT that include aquatic and terrestrial food availability, nesting beaches for reproduction, and overwintering sites. With numerous invasive fish species in the water, there is risk from predation on juvenile WPT and there should be considerations for the competition of habitat and resources between invasive RES at ANWA and WPT. There are numerous basking logs available primarily from trees that have fallen in from the banks, but the steep sided banks and phytoremediation mobilization of contaminants is inhibited by the lack of submergent and emergent vegetation in the reservoirs.

This report was a preliminary assessment of risks, and could be confirmed through a more thorough sampling across the site with a weight of evidence approach. Regardless, the levels of heavy metals and pesticides found in the ecosystem are concerning, especially for the lower trophic organisms, and restoration (reducing the nutrient load, trying to create a more functional wetland, or reverting back to tidal estuary) should be top priority for sustainability. ANWA is not an ideal location for the long term success of an endangered population of WPT.

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Appendix A. Pesticide, Metals, and Nutrient Sampling in 2018

Table A1 – Raw sediment data results completed during August 2018. OCP units are all in ng/g. Site 1 replicates (Figure 3).

Sample Name	SITE 1 REP A	Duplicate of SITE 1 REP A	SITE 1 REP B	SITE 1 REP C
Sample Size	2.15 g	2.17 g	3.14 g	4.85 g
Percent Moisture	89.4%	89.3%	84.4%	75.8%
Target Analytes	ng/g	ng/g	ng/g	ng/g
Hexachlorobutadiene	0.0145	<0.13	0.0218	0.00872
1,2,4,5-Tetrachlorobenzene	<0.0016	<0.028	<0.00098	<0.00058
1,2,3,4-Tetrachlorobenzene	<0.0016	<0.028	0.0166	<0.00057
Pentachlorobenzene	0.0277	<0.033	0.0293	0.0149
Hexachlorobenzene	0.0438	<0.067	0.0435	0.0252
3,4,5,6-Tetrachloroveratrole	<0.0062	<0.096	<0.0061	<0.0030
Pentachloroanisole	0.0288	<0.22	<0.021	<0.021
alpha-BHC	<0.037	<0.50	<0.021	<0.013
beta-BHC	<0.061	<0.87	<0.034	<0.021
gamma-BHC	<0.041	<0.66	<0.024	<0.015
delta-BHC	<0.045	<0.74	<0.027	<0.016
Pentachloronitrobenzene	<0.023	<0.66	<0.014	<0.0063
Heptachlor	<0.0043	<0.093	<0.0022	<0.0027
Aldrin	<0.0066	<0.14	<0.0041	<0.0024
4,4'-DDNU	<0.014	<0.28	<0.0085	<0.0048
Dacthal	<0.021	<0.30	<0.012	<0.0061
Chlorpyrifos	<0.052	<0.78	<0.064	0.105
Octachlorostyrene	<0.0067	<0.084	<0.0038	<0.0018
Heptachlor Epoxide B	<0.11	<0.13	<0.079	<0.038
Heptachlor Epoxide A	<0.74	<0.85	<0.53	<0.26

Oxychlordane	<0.30	<0.42	<0.27	<0.17
4,4'-DDMU	<0.61	<15	<0.47	<0.25
trans-Chlordane	<0.027	<0.39	<0.021	<0.012
cis-Chlordane	<0.026	<0.38	<0.021	<0.012
trans-Nonachlor	<0.024	<0.35	0.0344	<0.011
Dieldrin	<0.032	<0.35	0.0432	<0.013
Endrin	<0.048	<0.84	<0.021	<0.0072
cis-Nonachlor	<0.032	<0.36	<0.014	<0.0096
Endosulfan I	<0.066	<1.2	<0.036	<0.024
Endosulfan II	<0.11	<1.7	<0.069	<0.043
Endosulfan Sulfate	<0.018	<0.73	<0.018	<0.0082
2,4'-DDE	<0.017	<0.31	<0.010	<0.0049
4,4'-DDE	0.622	0.812	1.08	0.546
2,4'-DDD	<0.018	<0.45	<0.055	<0.0072
4,4'-DDD	<0.091	<0.42	0.250	0.0959
2,4'-DDT	<0.039	<0.60	<0.032	<0.012
4,4'-DDT	<0.042	<1.0	<0.13	<0.024
Endrin Aldehyde	<0.022	<1.0	<0.011	<0.0066
Endrin Ketone	<0.051	<1.3	<0.029	<0.018
Methoxychlor	<0.031	<1.6	<0.050	<0.023
Dicofol	<1.2	<13	<1.6	<0.63
Mirex	<0.0087	<0.049	<0.0088	<0.0044
Parlar 26	<0.13	<2.6	<0.16	<0.079
Parlar 50	<0.14	<1.7	<0.16	<0.063
Parlar 62	<0.19	<2.8	<0.23	<0.089
Extraction Standards	% Rec	% Rec	% Rec	% Rec
Pentachlorobenzene, 13C6-	46	66	59	64
Hexachlorobenzene, 13C6-	35	41	40	50
alpha-BHC, 13C6-	51	58	67	72
gamma-BHC, d6-	51	55	65	72
Heptachlor, 13C10-	47	48	63	73
Oxychlordane, 13C10-	40	47	52	63

trans-Nonachlor, 13C10-	50	55	68	76
Dieldrin, 13C12-	52	51	67	74
Endrin, 13C12-	47	39	65	76
Endosulfan II, 13C9-	49	56	54	66
4,4'-DDE, 13C12-	56	54	69	79
4,4'-DDD, 13C12-	41	39	38	49
4,4'-DDT, 13C12-	27	32	21	28
Methoxychlor, d6-	21	32	12	18
Mirex, 13C10-	32	48	24	30

Table A2 – Sediment analysis raw data continued. Sampling completed during August 2018. OCP units are all in ng/g. Site 2 replicates (Figure 3).

Sample Name	SITE 2 REP A	SITE 2 REP B	SITE 2 REP C
Sample Size	15.51 g	13.98 g	14.81 g
Percent Moisture	23.9%	31.1%	27.0%
Target Analytes	ng/g	ng/g	ng/g
Hexachlorobutadiene	0.00316	0.00337	<0.0026
1,2,4,5-Tetrachlorobenzene	0.00760	<0.00018	<0.00022
1,2,3,4-Tetrachlorobenzene	0.00817	<0.00018	<0.00022
Pentachlorobenzene	0.0102	0.00313	0.00299
Hexachlorobenzene	0.0111	<0.0050	0.00457
3,4,5,6-Tetrachloroveratrole	<0.0062	<0.00068	<0.00068
Pentachloroanisole	0.00635	<0.0012	<0.0017
alpha-BHC	<0.0039	<0.0037	<0.0036
beta-BHC	<0.0064	<0.0061	<0.0060
gamma-BHC	<0.0050	<0.0046	<0.0040
delta-BHC	<0.0055	<0.0051	<0.0044
Pentachloronitrobenzene	0.0162	<0.0016	<0.0028
Heptachlor	0.00347	<0.0011	<0.00086
Aldrin	0.00124	<0.00059	<0.00037
4,4'-DDNU	<0.0014	<0.0011	<0.0010

Dacthal	<0.0018	<0.0018	<0.0015
Chlorpyrifos	<0.0047	0.0146	<0.0046
Octachlorostyrene	<0.00062	<0.00044	<0.00050
Heptachlor Epoxide B	<0.019	<0.015	<0.018
Heptachlor Epoxide A	<0.13	<0.099	<0.12
Oxychlordane	<0.051	<0.051	<0.050
4,4'-DDMU	<0.051	<0.061	<0.056
trans-Chlordane	<0.0035	<0.0053	<0.0027
cis-Chlordane	<0.0033	<0.0052	<0.0026
trans-Nonachlor	<0.0031	<0.0048	<0.0024
Dieldrin	<0.0018	0.00515	<0.0018
Endrin	<0.0041	<0.0028	<0.0023
cis-Nonachlor	<0.0024	<0.0016	<0.0014
Endosulfan I	<0.0057	<0.0051	<0.0078
Endosulfan II	<0.0073	<0.011	<0.0096
Endosulfan Sulfate	<0.0018	<0.0028	<0.0028
2,4'-DDE	<0.0013	<0.0016	<0.0014
4,4'-DDE	0.0186	0.100	0.0512
2,4'-DDD	<0.0015	<0.0026	<0.0017
4,4'-DDD	0.00521	0.0237	0.0135
2,4'-DDT	<0.0019	<0.0032	<0.0025
4,4'-DDT	<0.0032	<0.0087	<0.0039
Endrin Aldehyde	<0.0016	0.00257	<0.0027
Endrin Ketone	<0.0055	<0.0045	<0.0048
Methoxychlor	<0.0042	<0.0050	<0.0025
Dicofol	<0.12	<0.11	<0.090
Mirex	<0.00092	<0.0011	<0.00076
Parlar 26	<0.013	<0.014	<0.011
Parlar 50	<0.013	<0.013	<0.014
Parlar 62	<0.019	<0.018	<0.020
Extraction Standards	% Rec	% Rec	% Rec
Pentachlorobenzene, 13C6-	49	73	66
Hexachlorobenzene, 13C6-	25	45	38

alpha-BHC, 13C6-	60	87	78
gamma-BHC, d6-	55	83	80
Heptachlor, 13C10-	58	85	79
Oxychlorane, 13C10-	50	70	64
trans-Nonachlor, 13C10-	74	91	89
Dieldrin, 13C12-	68	87	81
Endrin, 13C12-	71	92	86
Endosulfan II, 13C9-	69	84	80
4,4'-DDE, 13C12-	77	98	91
4,4'-DDD, 13C12-	61	71	75
4,4'-DDT, 13C12-	40	51	55
Methoxychlor, d6-	35	31	44
Mirex, 13C10-	54	53	64

Table A3 – Water quality analysis raw values from ALS for Sites 1 and 2 (Figure 3). Sampling completed during August 2018. OCP units are all in ng/L.

	Site 1	Site 2
Target Analytes	ng/L	ng/L
Sample Size	1.05 L	1.08 L
Hexachlorobutadiene	0.0476	0.0556
Hexachlorobenzene	<0.14	0.111
alpha-BHC	<0.058	<0.082
beta-BHC	<0.10	<0.082
gamma-BHC	<0.065	<0.052
delta-BHC	<0.073	<0.059
Heptachlor	<0.012	<0.020
Aldrin	0.00952	<0.025
4,4'-DDNU	<0.027	<0.028
Chlorpyrifos	<0.084	<0.23
Heptachlor Epoxide B	<0.023	<0.038
Heptachlor Epoxide A	<0.037	<0.11
Oxychlorane	<0.018	<0.037
4,4'-DDMU	<1.0	<1.4

trans-Chlordane	<0.060	<0.15
cis-Chlordane	<0.057	<0.15
trans-Nonachlor	<0.054	<0.14
Dieldrin	0.0762	<0.036
Endrin	<0.074	<0.11
cis-Nonachlor	<0.040	<0.067
Endosulfan I	<0.043	<0.041
Endosulfan II	<0.11	<0.063
Endosulfan Sulfate	<0.043	<0.020
2,4'-DDE	0.0381	<0.035
4,4'-DDE	<0.087	0.278
2,4'-DDD	<0.036	<0.077
4,4'-DDD	<0.036	<0.073
2,4'-DDT	<0.050	<0.072
4,4'-DDT	<0.056	<0.12
Endrin Aldehyde	0.0667	<0.047
Endrin Ketone	<0.067	<0.047
Methoxychlor	<0.038	<0.060
Dicofol	<1.6	<1.2
Mirex	0.0381	0.0556
Parlar 26	<0.15	<0.40
Parlar 50	<0.15	<0.46
Parlar 62	<0.26	<0.79
Extraction Standards	% Rec	% Rec
Pentachlorobenzene, 13C6-	33	50
Hexachlorobenzene, 13C6-	39	40
alpha-BHC, 13C6-	66	43
gamma-BHC, d6-	69	50
Heptachlor, 13C10-	39	18
Oxychlordane, 13C10-	53	26
trans-Nonachlor, 13C10-	50	22
Dieldrin, 13C12-	58	67
Endrin, 13C12-	48	60

Endosulfan II, 13C9-	49	59
4,4'-DDE, 13C12-	54	22
4,4'-DDD, 13C12-	46	26
4,4'-DDT, 13C12-	46	21
Methoxychlor, d6-	43	54
Mirex, 13C10-	42	14

Table A4 – Triplicates of sediment parameters at Sites 1 and 2 (Figure 3) completed during August 2018. Soil parameters such as pH, available Nitrate-N, Phosphate-P, pesticides, and a suite of metals are included in the table.

Parameter	Lowest Detecti on Limit	Units	SITE 1 REP A	SITE 1 REP B	SITE 1 REP C	SITE 2 REP A	SITE 2 REP B	SITE 2 REP C
Physical Tests (Soil)								
% Moisture	0.10	%	84.3	81.4	74.0			
pH (1:2 soil:water)	0.10	pH	7.86	7.96	7.96	7.82	8.16	8.03
Plant Available Nutrients (Soil)								
Available Nitrate-N	1.0	mg/kg	<2.0	<2.0	<2.0	<1.0	<1.0	<1.0
Available Phosphate -P	2.0	mg/kg	3.2	3.1	2.3	<2.0	<2.0	<2.0
Metals (Soil)								
Aluminum (Al)	50	mg/kg	17400	16800	14300	8910	8780	8270
Antimony (Sb)	0.10	mg/kg	0.51	0.51	0.62	0.20	0.18	0.35
Arsenic (As)	0.10	mg/kg	7.83	8.31	6.70	3.91	2.76	3.23
Barium (Ba)	0.50	mg/kg	163	151	127	30.9	44.3	36.5
Beryllium (Be)	0.10	mg/kg	0.39	0.39	0.36	0.18	0.17	0.18
Bismuth (Bi)	0.20	mg/kg	<0.20	<0.20	<0.20	<0.20	<0.20	<0.20

Boron (B)	5.0	mg/kg	12.4	11.6	14.1	<5.0	<5.0	<5.0
Cadmium (Cd)	0.020	mg/kg	0.241	0.295	0.311	0.078	0.103	0.117
Calcium (Ca)	50	mg/kg	13100	12300	15000	4620	4230	4440
Chromium (Cr)	0.50	mg/kg	41.4	41.2	39.9	27.8	25.3	26.7
Cobalt (Co)	0.10	mg/kg	12.7	12.6	12.3	8.26	8.06	8.29
Copper (Cu)	0.50	mg/kg	37.4	41.2	50.9	11.2	12.1	11.9
Iron (Fe)	50	mg/kg	36000	34500	30600	19100	18000	19200
Lead (Pb)	0.50	mg/kg	12.6	14.0	22.2	2.50	4.41	3.35
Lithium (Li)	2.0	mg/kg	20.4	19.6	16.5	8.1	8.4	8.2
Magnesium (Mg)	20	mg/kg	11400	10800	10700	7090	7070	6740
Manganese (Mn)	1.0	mg/kg	710	768	1040	436	420	393
Mercury (Hg)	0.0050	mg/kg	0.0583	0.0597	0.0686	0.0307	0.0252	0.0352
Molybdenum (Mo)	0.10	mg/kg	1.68	1.72	1.02	0.17	0.21	0.22
Nickel (Ni)	0.50	mg/kg	40.7	42.5	43.2	29.8	28.9	28.6
Phosphorus (P)	50	mg/kg	1160	1070	991	415	442	433
Potassium (K)	100	mg/kg	2080	1940	1640	580	660	610
Selenium (Se)	0.20	mg/kg	0.40	0.41	0.42	<0.20	<0.20	<0.20
Silver (Ag)	0.10	mg/kg	0.12	0.14	0.15	<0.10	<0.10	<0.10
Sodium (Na)	50	mg/kg	12500	8940	7820	519	730	611
Strontium (Sr)	0.50	mg/kg	139	134	137	21.0	24.5	23.7
Sulfur (S)	1000	mg/kg	13700	13700	10800	2100	1400	1800
Thallium (Tl)	0.050	mg/kg	0.109	0.107	0.096	<0.050	<0.050	<0.050

Tin (Sn)	2.0	mg/kg	<2.0	<2.0	3.6	<2.0	<2.0	<2.0
Titanium (Ti)	1.0	mg/kg	902	887	874	868	795	779
Tungsten (W)	0.50	mg/kg	<0.50	<0.50	<0.50	<0.50	<0.50	<0.50
Uranium (U)	0.050	mg/kg	0.778	0.829	0.650	0.241	0.254	0.252
Vanadium (V)	0.20	mg/kg	53.9	56.1	57.1	44.4	39.1	38.7
Zinc (Zn)	2.0	mg/kg	79.5	84.3	93.7	35.9	38.7	37.4
Zirconium (Zr)	1.0	mg/kg	5.5	6.1	5.8	5.2	4.7	4.9
Herbicides (Soil)								
Bromoxynil	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
Clopyralid	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
2,4-D	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
Dicamba	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
2,4-DB	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
2,4-DP	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
Dinoseb	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
MCPA	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
MCPB	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
Mecoprop	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
Picloram	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
2,4,5-T	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			
2,4,5-TP	0.0050	mg/kg	<0.0050	<0.0050	<0.0050			

Triclopyr	0.0050	mg/kg	<0.005 0	<0.005 0	<0.005 0			
2,4-Dichlorophenylacetic Acid		%	96	94	93			
Pesticides (Soil)								
Bromacil	0.00050	mg/kg	<0.000 50	<0.000 50	<0.000 50			
Linuron	0.00050	mg/kg	<0.000 50	<0.000 50	<0.000 50			

Table A5 – Raw values for water parameters at Sites 1, 2, and 3 (Figure 3) sampled during August 2018. Water parameters of hardness, anions and nutrients, metals, and pesticides are included in the data.

Parameter	Lowest Detection Limit	Units	SITE 1	SITE 2	SITE 3
Physical Tests (Water)					
Hardness (as CaCO ₃)	0.63	mg/L	929	563	559
Anions and Nutrients (Water)					
Total Nitrogen	0.30	mg/L	2.45	1.61	1.56
Phosphorus (P)-Total	0.020	mg/L	0.360	0.183	0.216
Total Metals (Water)					
Aluminum (Al)-Total	0.015	mg/L	0.433	0.400	0.374
Antimony (Sb)-Total	0.00050	mg/L	<0.0010	0.00075	0.00070
Arsenic (As)-Total	0.00050	mg/L	0.0054	0.00475	0.00443
Barium (Ba)-Total	0.00050	mg/L	0.166	0.0858	0.0906
Beryllium (Be)-Total	0.00050	mg/L	<0.0010	<0.00050	<0.00050
Bismuth (Bi)-Total	0.00025	mg/L	<0.00050	<0.00025	<0.00025

Boron (B)-Total	0.050	mg/L	0.64	0.382	0.391
Cadmium (Cd)-Total	0.000025	mg/L	<0.000050	0.000034	<0.000025
Calcium (Ca)-Total	0.25	mg/L	50.1	49.1	50.2
Cesium (Cs)-Total	0.000050	mg/L	<0.00010	<0.000050	<0.000050
Chromium (Cr)-Total	0.00050	mg/L	0.0011	0.00081	0.00070
Cobalt (Co)-Total	0.00050	mg/L	<0.0010	0.00057	0.00057
Copper (Cu)-Total	0.0025	mg/L	<0.0050	<0.0025	<0.0025
Iron (Fe)-Total	0.050	mg/L	0.85	0.635	0.687
Lead (Pb)-Total	0.00025	mg/L	0.00058	0.00053	0.00049
Lithium (Li)-Total	0.0050	mg/L	0.011	0.0090	0.0092
Magnesium (Mg)-Total	0.025	mg/L	195	107	105
Manganese (Mn)-Total	0.00050	mg/L	0.347	0.156	0.174
Mercury (Hg)-Total	0.0000050	mg/L	<0.0000050	<0.0000050	<0.0000050
Molybdenum (Mo)-Total	0.00025	mg/L	0.00158	0.00201	0.00217
Nickel (Ni)-Total	0.0025	mg/L	<0.0050	0.0032	0.0030
Phosphorus (P)-Total	0.25	mg/L	<0.50	0.29	0.26
Potassium (K)-Total	0.25	mg/L	56.3	30.9	31.0
Rubidium (Rb)-Total	0.0010	mg/L	0.0083	0.0073	0.0069
Selenium (Se)-Total	0.00025	mg/L	<0.00050	0.00025	0.00029
Silicon (Si)-Total	0.50	mg/L	5.0	3.06	2.92

Silver (Ag)-Total	0.000050	mg/L	<0.00010	<0.000050	<0.000050
Sodium (Na)-Total	0.25	mg/L	1560	878	884
Strontium (Sr)-Total	0.0010	mg/L	0.942	0.736	0.735
Sulfur (S)-Total	2.5	mg/L	27.3	33.1	31.8
Tellurium (Te)-Total	0.0010	mg/L	<0.0020	<0.0010	<0.0010
Thallium (Tl)-Total	0.000050	mg/L	<0.00010	<0.000050	<0.000050
Thorium (Th)-Total	0.00050	mg/L	<0.0010	<0.00050	<0.00050
Tin (Sn)-Total	0.00050	mg/L	<0.0010	<0.00050	<0.00050
Titanium (Ti)-Total	0.0015	mg/L	0.0188	0.0177	0.0142
Tungsten (W)-Total	0.00050	mg/L	<0.0010	<0.00050	<0.00050
Uranium (U)-Total	0.000050	mg/L	0.00062	0.000856	0.000807
Vanadium (V)-Total	0.0025	mg/L	0.0091	0.0089	0.0084
Zinc (Zn)-Total	0.015	mg/L	<0.030	<0.015	<0.015
Zirconium (Zr)-Total	0.00030	mg/L	<0.00060	<0.00030	<0.00030
Pesticides (Water)					
Bromacil	0.10	ug/L	<0.10	<0.10	<0.10
Iprodione	0.10	ug/L	<0.10	<0.10	<0.10
Linuron	0.10	ug/L	<0.10	<0.10	<0.10

Appendix B. Risk Assessment Calculations

Table B1 - Receptor of concern (painted turtle) characteristics used in the exposure calculations.

Receptor	Painted Turtle
Feeding type	Omnivore
Diet proportion	98% animals <ul style="list-style-type: none"> - 91.8% benthic invertebrates - 6.2% fish, as carrion ~2 % incidental sediment
Body weight (kg)	0.240 ¹
Food ingestion rate (kg/day ww)	0.0003 ²
Water ingestion rate (kg/day)	0.0048 ³
Soil ingestion (% of food intake)	5.9 ¹
Soil ingestion rate (kg/day)	0.0000177 ⁴
Habitat range	4.31 ha ¹
Area of study (Fuller) (ha)	5.8825 ha

¹USEPA, 2003.

²Ernst et al., 1972. Calculated from ingestion rate of 1.25 mg/g/day at 25°C using body weight of 240 g.

³USEPA, 1993. Calculated from the ingestion rate of 0.02 g/g/day using body weight of 240 g.

⁴USEPA, 2003. Calculated from the soil ingestion percentage and food ingestion rate.

Table B2 – BCF or BAF factors for the receptors for each contaminant.

Contaminant	Family/species	BCF/BAF
As	Fish - <i>Cyprinus carpio</i>	19 ⁴
As (mg/kg)	Benthic – <i>Hyalella azteca</i>	0.8 ³
Cr	Benthic – nine families of macroinvertebrates	2.13 ²
Cu (mg/kg)	Benthic - chironomids	4.58 ¹
Fe	Benthic – nine families of macroinvertebrates	1.15 ²
Mn	Benthic – nine families of macroinvertebrates	2.66 ²
Ni (mg/kg)	Benthic - chironomids	0.47 ¹

¹ Kļaviņš et al., 1998. Averages of different BCF's used.

² Chiba et al., 2010. The nine benthic families include Baetidae, Hydrobiosidae, Libellulidae, Gomphidae, Ceratopogonidae, Chironomidae, Elmidae, Glossiphoniidae and Tubificidae; BAF numbers are averaged from 6 different sites.

⁴ Baker and King, 1994. Average of two values.

Table B2 - Contaminants of potential concern estimated exposure through water for the fish community at Fuller reservoir.

Receptor of Concern exposure	COPC estimated exposure (water)
	As (mg/L)
BCF	19
Fish communities	0.0054
Total exposure	0.1026

Table B3 - Contaminants of potential concern estimated exposure through sediments (mg/kg) for benthic communities at Fuller.

Receptor of Concern exposure	COPC estimated exposure with bioaccumulation factor (sediment)						
	As	Cr	Cu	Fe	Mn	Ni	
BCF	0.8	2.13	4.58	1.15	2.66	0.47	
Benthic communities	7.6	40.8	43.2	33700	839.3	42.1	
Total exposure	6.1	87	198	38755	2233	20	

Table B4 - Contaminants of potential concern estimated through a dose based approach based on the painted turtle characteristics.

Dose estimate exposure	COPC						
	As	Cr	Cu	Fe	Mn	Ni	
Dose food (mg/kg-BW/day) (invertebrates 91.8%)	0.0095	0.136	0.31	60.70	3.50	0.03	
Dose food (mg/kg-BW/day) (fish 6.2%)	0.000011	0.0	0.0	0.0	0.0	0.0	
Dose sediment (mg/kg-BW/day) (soil 2%)	0.000015	0.000082	0.000087	0.068	0.0017	0.000085	
Dose water (mg/kg-BW/day)	0.00015	0.00003	0.0	0.023	5.3	0.00014	
Dose total (mg/kg-BW/day)	0.0097	0.14	0.31	60.78	8.82	0.031	

Table B5 – Example Calculation for Painted Turtle using arsenic.

Painted turtle	Dose (total)
Arsenic	$[\text{benthic}] + [\text{fish}] + [\text{soil}] + [\text{water}] = \text{total dose}$ $[(5.88 \text{ ha}/4.31 \text{ ha}) * (0.0003 \text{ kg/day} * 6.08 \text{ mg/kg} / 0.24 \text{ kg}) * 0.918] +$ $[(5.88 \text{ ha}/4.31 \text{ ha}) * (0.0003 \text{ kg/day} * 0.1026 \text{ mg/kg} / 0.24 \text{ kg}) * 0.062] +$ $[(5.88 \text{ ha}/4.31 \text{ ha}) * (0.000015 \text{ kg/day} * 7.6 \text{ mg/kg} / 0.24 \text{ kg}) * 0.02] +$ $[(5.88 \text{ ha}/4.31 \text{ ha}) * (0.0048 \text{ kg/day} * 0.0054 \text{ mg/kg} / 0.24 \text{ kg})]$ $= 0.0097 \text{ mg/kg-BW/day}$

Table B6 - TRV values used in the ERA to calculate hazard quotients. Values were obtained from US EPA and CCME derived toxicity data when available, and using sediment quality guidelines when not available.

Receptor group (exposure type)	COPC	Methods	TRV (water) (mg/L)	TRV (sediment) (mg/kg)
Benthic Invertebrates [C]	As	Chironomids	NA ²	6.87 ³
	Cr	Amphipoda and Chironomidae	NA ²	95 ⁴
	Cu	<i>Hyalella azteca</i>	NA ²	89.8 ⁵
	Fe	Guideline	NA ²	20000 ⁶
	Mn	Guideline	NA ²	460 ⁶
	Ni	Guideline	NA ²	16 ⁶
Fish [C]	As	<i>Oncorhynchus mykiss</i> (LC ₅₀)	0.55 ³	NA ²
	Cr		N/A ²	NA ²
	Cu		N/A ²	NA ²
	Fe		N/A ²	NA ²
	Mn		N/A ²	NA ²
	Ni		N/A ²	NA ²
Painted turtle (dose) (avian receptor)	As	EPA derived	NA ²	43 ¹
	Cr	EPA derived	NA ²	26 ¹
	Cu	EPA derived	NA ²	28 ¹

Fe		NA ²	ND ⁷
Mn	EPA derived	NA ²	4300 ¹
Ni	EPA derived	NA ²	210 ¹

¹USEPA, 2016.

²These TRV values not needed for calculations.

³ CCME, 2001.

⁴CCME, 1999d.

⁵CCME, 1998c.

⁶MEEQ, 1993.

⁷No appropriate guidelines.

Table B7 - Hazard calculation example for arsenic for the different receptors.

Arsenic	Hazard quotient calculation
	[dose or concentration] / TRV
Benthic invertebrates	6.08/6.87 = 0.89
Fish	0.1026/0.55 = 0.19
Painted turtle	0.0097/43 = 0.00023