Using 10-Years of Population Monitoring Data to Assess Breeding Productivity of the Oregon Spotted Frog (*Rana pretiosa)*

By

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Abstract

Relationships between changing environmental variables and amphibian populations have been understudied. Yet, alterations to temperature and precipitation have been suggested as contributors to the decline of some pond-breeding species, such as the Oregon Spotted Frog (*Rana pretiosa). R. pretiosa* has been classified as the most endangered amphibian in Canada, yet the cause for its decline is unknown. Therefore, this paper examined associations between temperature and precipitation, and *R. pretiosa* population trends, using a 10-year data set from two breeding populations in the Lower Mainland of British Columbia. Timing of oviposition was positively related to higher temperature and increased precipitation within both populations ($p<0.05$). No statistical relationship was determined between egg mass productivity and temperature or precipitation; however, this paper proposes that further research, consistent protocols and longer study periods, is necessary in order to determine environmental variables as possible predictors of population success. This paper recommends the evaluation of breeding success through survivorship studies, as such methods provide insight into productivity as the primary determinant for population recruitment. Further, ecological restoration efforts can be implemented to help ameliorate negative consequences climate change poses on reproductive success.

Keywords: amphibian, climate change, conservation, ecological restoration, endangered, population dynamics, population monitoring, survivorship

For Mom.

Completing this was one of your last wishes - we did it.

Love you for always.

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List of Acronyms

CMR	Capture-Mark-Recapture
COSEWIC	Committee on the Status of Endangered Wildlife
COSFRT	Canadian Oregon Spotted Frog Recovery Team
GPS	Global Positioning System
OSF	Oregon Spotted Frog
PIT	Passive Integrated Transponder
SARA	Species at Risk Act
SUL	Snout Urostyle Length
SVL	Snout Ventricle Length
UTM	Universal Transverse Mercator

Photo 1. Juvenile Oregon Spotted Frog

Chapter 1. Introduction

Over the last few decades, scientists have seen increasing evidence that amphibian populations are experiencing dramatic declines (Blaustein et al. 1994, Stuart, Simon N. et al. 2004, Beebee and Griffiths 2005). The causes for these declines are numerous and range from loss of habitat to invasive species to poor water quality to disease (Beebee and Griffiths 2005, Salvidio 2009)**.** Perhaps most concerning in the coming years for amphibian populations, however, are the consequences of climate change and the effects on temperature and moisture regimes, two key determinants that affect the health of amphibian populations (Donnelly, Maureen A. and Crump, Martha L. 1998, Blaustein et al. 2010, Benard 2015). Notably, these two fundamental abiotic variables directly impact amphibian biological function (Carey and Alexander 2003), including timing of breeding, oviposition and larvae development (Benard 2015). It is furthermore expected that, as a consequence of climate change, changes to these environmental variables hold potential to increase stress on amphibian species by altering the frequency of weather events, melting of freshet, changing the length of the dry season, and enhancing UV-B radiation. This is especially true for highly aquatic species that rely on pooled water for breeding, as the timing of breeding is an important life history event that may either positively or negatively alter the success of an amphibian population (Benard 2015)**.** It follows, that changes in temperature and water availability by precipitation could present significant consequences for the onset of breeding and the development of amphibian offspring.

Population monitoring programs have been developed for amphibian species to provide researchers with baseline information to detect declining population trends before a critical state has been reached (Scherer and Tracey 2011). The use of egg mass surveys (Campbell Grant et al. 2005) and capture-mark-recapture (CMR) studies (Petit and Valiere 2006) are commonly used methods for surveying pond-breeding amphibians. While these techniques are useful for monitoring species distributions and highlighting changes in population size, few are capable of providing causal evidence. For pond-breeding amphibians, for example, monitoring must be conducted over multiple generations for trends to be reliably detected (Blaustein et al. 1994). Thus, long-term studies prove critical for amphibian species experiencing significant population fluctuations from year to year.

It is now abundantly clear that the climate on earth is noticeably different from previous centuries, with temperature and precipitation patterns continuing to change at relatively accelerated rates (Blaustein et al. 2010). Despite this knowledge, there exists a lack of robust studies investigating the effects of climate change on pond-breeding amphibians, specifically with regard towards reproduction and larvae development. It has been noted that research has primarily focused on the direct effects of climate change, as linking indirect effects and population dynamics can be challenging (Corn 2005). Further, species demonstrating consistently insufficient reproduction and recruitment behaviours may fail to maintain an adult breeding population, ultimately leading to population decline (Semlitsch 2002). Specifically, pond-breeding amphibians exhibit greater fluctuations in population size due to their population stochasticity and reliance on environmental variables (Green 2003). In this sense, long-term studies on amphibian populations are sparse (Blaustein et al. 1994).

In considering that climate change may be severely impacting the success of pond-breeding amphibians, and consequently contributing to the decline in local species populations, the need for reliable long-term examination of these species becomes apparent (Blaustein et al. 2001). To fully understand the changes in population numbers, this paper analyzes a 10-year data set to argue for the development of pond-breeding amphibian monitoring programs that carefully follow consistency, have an identifiable state variable to measure change, and provide guidance for conservationists to conduct sound management decisions. Therefore, this study identifies variables and methods that may best capture productivity and breeding trends among amphibian populations within the prospective context of changing climatic conditions. Additionally, suggestions are made in regards to restoration efforts as well as implementation of citizen science programs.

1.1. Amphibians in Decline

Around the globe, the prevalence of amphibian species is declining at a rapid rate (Blaustein and Wake 1995, Carey and Alexander 2003, Stuart et al. 2004)**.** Moreover, as temperatures and precipitation patterns continue to change, there is increasing concern regarding the effects that climate change will have on already threatened amphibian populations around the world (Blaustein and Wake 1990, D'Amen and Bombi 2009). While the cause for some species declines have been documented, the reasons behind the declines of many pond-breeding amphibian species, in particular, remains unknown. The limited existing literature attributes identified patterns of species decline to habitat degradation and urbanization, as well as increasing threats caused by invasive species, disease and the effects of pesticides and fertilizers from agriculture (Beebee 1995). Furthermore, secondary changes to the physical environment related to climate change, such as increased UV-B radiation, temperatures and reduction in water levels, have also been suggested for many species (Houlahan 2000, Beebee and Griffiths 2005).

Amphibians are integral components of the ecosystem, as they represent strong ecological indicators, add to the biological complexity of the environment (Blaustein et al. 1994), and play vital roles in the ecological food chain as both prey and secondary consumers (Donnelly and Crump 1998). For instance, tadpoles are involved in nutrient cycling as they transition from herbivores to omnivores; as adult frogs, they prey upon invertebrates (Blaustein et al. 1994). Amphibians also serve as a primary food source for predators such as birds, fish and mammals, therefore substantiating important components across various food webs (Blaustein and Wake 1990). These species are notable determinants of ecosystem health, as their permeable skin readily absorbs any toxicity exposures within their environments (Blaustein and Wake 1990, Donnelly and Crump 1998). Their presence and health, therefore, is a determinant of the health of the environment in which they live.

Changes in climatic conditions increase the susceptibility of pond-breeding amphibians to alterations in oviposition and larval development, subjecting them to a greater risk of population decline (Benard 2015). As demonstrated by Benard (2015), timing of breeding, oviposition, female fecundity and timing of metamorphosis may be negatively affected by variable and unpredictable climatic conditions**.** In effect, breeding may be delayed and possibly even absent for a full year, as some species are capable of reabsorbing their eggs without laying (Donnelly and Crump 1998). Egg masses also face an increased mortality threat as a result of the reductions in water depth, fluctuations in water temperatures, and higher UV-B radiation that are accompanying climate change (Kiesecker 2001)**.** Once hatched, factors such as temperature and limited water availability may constrain successful recruitment into the population (Semlitsch 2002)**.**

This combination of low reproductive effort, increased embryonic risks and high larvae mortality suggests that large scale climatic oscillations may prove devastating for the productivity and success of many amphibian species around the world (Blaustein and Wake 1995, Semlitsch 2002)**.**

1.2. Oregon Spotted Frog (*Rana pretiosa*)

With only four extant populations in the Fraser Valley, the Oregon Spotted Frog (OSF) (*Rana pretiosa)* is Canada's most endangered amphibian species. Appropriately, the *Committee on the Status of Endangered Wildlife* (COSEWIC) also classifies the OSF as endangered, and it is further protected under the *Species at Risk Act* (SARA) as a Red-Listed species. Membership within the family *Ranidae* imparts that the OSF is considered a true frog. However, the OSF was previously considered one species with the Columbia Spotted Frog (*Rana luteiventris*) and was differentiated from this subspecies on a molecular level in 1997 (Baird and Girard 1853; COSEWIC 2011). Its common name nods to the unique dark spots present on its head, back and legs, which grow in size and deepen in colour as each individual age (COSEWIC 2011). Another distinguishing characteristic of the OSF is its upturned eyes which contrast most other frog species exhibiting eyes on the sides of their heads.

The OSF is a Pacific Northwest endemic whose known range once spread from northern California through Oregon, Washington, and southern British Columbia (Cushman and Pearl 2007). Today, however, the species remains in less than 10% of its historical distribution, including complete extirpation from California (Canadian Oregon Spotted Frog Recovery Team 2012)**.** Of the remaining US populations, the OSF is only known to inhabit 30 locations in Oregon and 8 locations in Washington (COSEWIC 2011). All known historical Canadian populations are extant but have been genetically isolated from one another by distance and human development (Canadian Oregon Spotted Frog Recovery Team 2012). Still, the reason for the rapid decline of this species is relatively unknown and with less than 500 estimated mature individuals remaining in Canada, there is an immediate need to recover the population to a naturally sustainable level.

Monitoring and researching efforts, that seek to determine causative reasons for decline of the OSF, are primarily focused towards the effects of habitat loss and degradation (Canadian Oregon Spotted Frog Recovery Team 2012). Additionally, land development, agriculture, and hydrological alternations have all been proven to negatively affect pond-breeding species, including remaining populations of OSF. However, this paper will highlight that there is a significant need to additionally study emergent stressors posed by climate change. Even further, in order to protect the OSF from extirpation in Canada, advocacy and management decisions for remaining isolated populations must be developed from a strong understanding of OSF behavioural patterns and productivity. Successful species protection and advocacy, an example seen with the restoration of bald eagle populations, stems from a foundational understanding of patterns, threats, and needs that relate to the endangerment of the species (Blood and Anweiler 1994, Stinson et al 2001). In this respect, the Canadian Oregon Spotted Frog Recovery team (COSFRT) was established to organise efforts to monitor, conserve, and manage the OSF within its Canadian range and to increase the few remaining populations to self-sustaining levels. Critical recovery objectives for the COSFRT in Canada are focused towards preventing further habitat degradation and loss; sustaining and improving survivorship rates of all life stages; to establish/re-establish self-sustaining populations; and to improve conservation and recovery efforts by addressing knowledge gaps (Canadian Oregon Spotted Frog Recovery Team 2012).

1.3. Study Rationale

Since the 1970's discovery of OSF populations in the Fraser Valley lowlands, scientists and wildlife groups have maintained ongoing monitoring of OSF subpopulations. Results from such data suggest both population variation and scientific recognition of the importance of this keystone population, as demonstrated by increased interest in monitoring egg mass numbers and raw data accumulation over recent years (COSEWIC 2011). While natural rises and falls are to be expected within any amphibian population, the last few years have seen an unprecedented reduction in breeding success; as such, OSF populations are considered to be in decline (COSEWIC 2011). In fact, for a few known historical populations, there has been no evidence of OSF in recent years, concluding extirpation from these localities (e.g. Aldergrove Detachment, Little Campbell River). The cause for the species decline within these Canadian populations is unknown.

Currently there are eight amphibians registered as endangered under both COSEWIC and SARA in Canada (COSEWIC, SARA). Still, one of the biggest challenges of recovering endangered species is the uncertainty posed by the cause of decline (Kissel et al. 2017), as well as the need for proper action to be taken**.** On this note, however, a number of hypotheses have attempted to explain fluctuations in individual OSF numbers. For example, although hypotheses have suggested that poor water quality create insufficient breeding conditions for the OSF, follow-up studies have been conducted to disprove this theory (McKibbin et al. 2008).

This project attempts to identify and quantify effects between changes in temperature and precipitation and the annual productivity of OSF egg masses. A long-term data set, such as the 10-year data assessed for this study, is unique for small, local populations and therefore provides an unparalleled opportunity towards gaining valuable site-specific knowledge for this threatened population. A major question that remains unanswered is how changing climate has affected populations living at the northern limit of their range. The results from this study will provide evidence whether changing climate is a contributor to the variation and decline of OSF numbers. As such, this study utilizes egg mass and monitoring data to analyze population trends and population productivity related to changing temperature and precipitation patterns. This is important because, although a population may include an abundant number of females reproducing each breeding season, it is the reproductive success that contributes to actual population recruitment. Using the life history of this species, then, this project proposes that certain methods of monitoring are insufficient, while also explaining how changes to environmental variables can be problematic. Results suggest that further studies be conducted to monitor the survivorship of egg masses and proposes specific approaches to data collection moving forward. The results of this study seek not only to add to the scientific knowledge of the species, but also to prompt site specific restoration to support conservation efforts.

1.4. Objective

This study adds to the scientific-based knowledge of the OSF by examining the relationship between climatic variables and population trends. The research and data analyses will contribute to filling species and site-specific population knowledge gaps, identifying population trends over both spatial and temporal scales, and helping to inform ecological restoration decisions. As well, this study points to citizen science as being an appropriate additional resource. Ultimately, this project strives to contribute critical information towards meeting the recovery objectives for this species, as outlined by COSEWIC (COSEWIC 2011).

In order to offer specific and precise data outcomes, this paper focuses on the Morris Valley wetland and Maria Slough populations of OSF within British Columbia, Canada. These populations were selected based on the long-term data set, and the close proximity of the two sites to one another. In doing so, this study chose to utilize egg mass counts to evaluate amphibian productivity, as well as to provide baseline and causative data for assessing population declines of the OSF within these populations. This project is unique as it uses data collected over a 10-year study period from 2008 to 2017, inclusive, to examine associations between population fluctuations and environmental variables that might direct future restoration and conservation attempts.

From the information presented thus far, it follows that temperature and precipitation perhaps represent critical components imperative to the success of any pond-breeding amphibian, including the OSF. It is therefore possible for any change in climatic conditions to contribute to the success, decline or constraint of this species to such a narrow range. This study thus proposes and attempts to evaluate the following hypotheses:

> H_0: Changes in temperature and precipitation regimes are not a contributor of population success of *Rana pretiosa.*
>
> H_1: Changes in temperature and precipitation regimes do contribute to the decline of the *Rana pretiosa* population.

Chapter 2. Study

2.1. Background of the Oregon Spotted Frog

As an aquatic species, OSF experiences three separate seasons during the year that determine microhabitat use**.** This annual cycle can be separated into oviposition (breeding), active summer season and overwintering period (Pearl and Hayes 2004). Perhaps in parallel to such defined seasonal distinctions, the OSF inhabits different regions of its wetland habitat with each season, and thus takes on certain unique seasonal characteristics, as described below.

The subpopulations focused on by this study are located within the Morris Valley and Maria Slough wetlands of British Columbia. The OSF breeding season in British Columbia is initiated when these amphibians become active as temperatures begin to rise to approximately 5^{o}C, typically towards the end of February (Licht 1969). When this occurs, OSF migrate along habitat corridors from overwintering sites towards their traditional, communal breeding areas of shallow pooled water (McAllister and Leonard 1997). Males move to these oviposition sites first, where they begin their breeding calls to females (Licht 1969). Typically, these calls are composed of 6-9 short, low-volume bass notes (Licht 1969). Once females have arrived, they pair up to mate and oviposit their egg masses, then leave the area to continue living solitary lives (Backhouse et al. 2002). Males will remain at the oviposition sites until breeding season is complete, which continues for approximately 2 to 4 weeks (Licht 1969).

OSF oviposition sites are shallow (<35 cm in depth) and gently sloping, typically within slow-moving water (McAllister and Leonard 1997, Cushman and Pearl 2007). Egg masses are not attached to vegetation, and thus OSF eggs risk stranding and desiccation due to shoreline placement of egg masses and the shallow waters of oviposition sites (Backhouse et al. 2002). In order for breeding to occur, however, conditions must be suitable, including adequate solar radiation, water depth and temperature - measures which can be unique to the population or even climatic conditions by year. The number of egg masses per mating pair varies amongst subpopulations and ranges, but averages between 600-1500 eggs per mass have been evidenced at Canadian sites (COSEWIC 2011). Development of egg masses (from hatching through to metamorphosis) can take 3 to 4 months (Licht 1974); if conditions are not ideal, egg masses may take longer to develop. Eggs typically hatch within 18 to 30 days (McAllister and Leonard 1997)**.** This study questions whether survivorship of OSF egg masses and tadpoles varies dramatically between years, perhaps leading to the fluctuations in population trends. Thus, prompting speculation that hydrological alterations and changing climate conditions hold a significant effect on the productivity of this species. Within its Canadian range, the OSF reaches sexual maturity by the age of 3, however OSF females may not breed until into their fourth year (McAllister and Leonard 1997, COSEWIC 2011). OSF males and females are thought to live from 5 to 9 years, which does suggest that OSF longevity varies amongst populations. It is possible that climate plays a role amongst such a wide estimate for the OSF lifespan, and so this knowledge gap highlights our need for further research towards understanding the OSF species.

In British Columbian populations, the OSF ranges in size from 40 to 80 mm snout-ventricle length (SVL), a slightly smaller average compared to southern populations (COSEWIC 2011). Mature females are relatively larger than mature males, and a study of all Canadian OSF populations determined the average SVL size of females as 66 mm and males as 57 mm (McAllister and Leonard 1997)**.** It follows that this smaller average size in males may make male frogs more susceptible to predation.

Once hatched, the tadpoles remain within their cluster jelly (McAllister and Leonard 1997). As they develop, this jelly diminishes and the tadpoles begin their free-swimming stage of life (McAllister and Leonard 1997). At this time, tadpoles are herbivorous graze-feeders, ingesting plant tissue and bacteria by scraping plant surfaces with their tooth rows, and detritus and carrion make-up the majority of their diet (McAllister and Leonard 1997). More mature juvenile and adult frogs are omnivorous, obtaining a range of nutrients from detritus to small macroinvertebrates to other small amphibians (Richardson 2011). Interestingly, OSF consume their prey by lunging forwards underwater, as opposed to attacking upwards above the surface like other subspecies from the *Ranidae* family.

2.1.1. Climate Change and Reproduction

Inherently, climate change provokes variations in temperature and precipitation patterns around the world. As noted in the literature, dramatic observed declines in amphibian populations suggest similar correlations with climate change (Beebee 1995, Carey and Alexander 2003, Kiesecker et al. 2001, Reading 2007), however studies are sparse and warrant further research (Carey and Alexander 2003). Of note, behaviour, hibernation and reproduction are directly impacted by changes in temperature and moisture, and these effects are amplified in more temperate areas such as the Lower Mainland of British Columbia (Reading 1998). Canadian populations of the OSF may already be highly susceptible to weather conditions as they are living at the northern limit of their range. If environmental variables continue to change, pond-breeding amphibians may fail to successfully reproduce or mature into offspring, ultimately spurring population decline and perhaps extinction for threatened species such as the OSF (Blaustein and Kiesecker 2002).

2.1.2. Timing of Breeding and Oviposition

Amphibians exhibit behavioural preferences that ensure optimal environmental variables before instigating breeding and oviposition (Blaustein et al. 2010). In this regard, Donnelly and Crump (1998) posit that climate changes lead to major change in instinctual reproductive activities. When temperatures begin to warm, for instance, frogs emerge from winter hibernation to initiate their breeding season (Licht 1969). As previously discussed, males migrate to traditional communal oviposition sites and females follow shortly thereafter (Backhouse et al. 2002). Colder winters, however, may result in adults leaving their overwintering sites later, which delays breeding behaviours until conditions warm later in the season. This delay in breeding and oviposition results in reduced length of development time for egg masses and larvae (Benard 2015).

Conversely, a warmer winter may result in earlier breeding and reproduction for amphibians, such as the OSF. This can present a physiological stressor, as there may not yet be adequate resources to replenish energy and nutrient stores lost throughout hibernation (Blaustein et al. 2001). Another issue associated with earlier breeding involves flash cold temperatures that could freeze egg masses and cause significant embryo mortality (Backhouse et al. 2002). As well, warmer temperatures may lead to lower water levels in breeding ponds, resulting in desiccation of egg masses prior to tadpole development (Backhouse et al. 2002). Some do argue, however, that earlier breeding may be beneficial to offspring, as it lengthens development periods and allows for metamorphoses to occur prior to ponds drying up (Donnelly and Crump 1998).

Climate change also threatens amphibians with unpredictable precipitation patterns, which represents an important variable for the onset of breeding. For the OSF, breeding ponds tend to be created by the onset of snow meltwater (Canadian Oregon Spotted Frog Recovery Team 2012). During years with warmer spring temperatures, areas may have water earlier in the season due to the melting of the freshet. With warmer temperatures and a lesser snow pack, this may result in lower than adequate water levels within breeding ponds. Adults may also be more susceptible to predation, as they may be compelled to migrate to the warm breeding sites sooner, however sites may not be adequate for the onset of breeding to begin. When faced with the inadequate conditions at breeding locations, females may choose to delay reproduction until the following year by reabsorbing their eggs and leaving the breeding site (Donnelly and Crump 1998). Another possibility is that males may migrate to the breeding sites much earlier than necessary to begin calling to females, thus depleting their energy reserves in a manner non-conducive to successful breeding (Donnelly and Crump 1998).

Another potential impact of climate change is that altered temperature and precipitation patterns may prompt different species to overlap breeding seasons (Blaustein et al. 2010). This will increase competition for food and oviposition sites and could even cause acoustical interference amongst male amphibians calling out for female mates (Donnelly and Crump 1998).

2.1.3. Egg Mass Survivorship and Larvae Development

All amphibian species and life history stages involve upper and lower thermal tolerance limits. Temperatures outside of a tolerable range for any given species will result in high rates of adult, embryo and/or larval mortality (Blaustein et al. 2010). Licht (1971) demonstrated that the lethal thermal limits for young OSF embryos ranges from approximately 6 to 28^{o}C, however older embryos can withstand short-term exposure to lethal cold temperatures. As well, the amount of available water in standing breeding pools acts as a determining factor for the success of egg masses to develop to metamorphosis (Licht 1974). It follows that changes to temperature and precipitation may affect egg mass survivorship and larvae development.

It should be considered that the OSF holds a strong preference to oviposit near sloped shorelines, even though this activity poses a heightened risk for egg mass stranding (McAllister and Leonard 1997). In other words, slopes expose egg masses to desiccation in times of warm, dry weather, or freezing during colder periods.

Warmer temperatures often hasten embryo development, and conversely cooler temperatures tend to slow down this process (Donnelly and Crump 1998). However, temperature extremes leave OSF embryos susceptible to freezing or desiccation (Licht 1974, Backhouse et al. 2002). Warmer breeding seasons enable faster progression to metamorphoses, but also decrease frog size at metamorphosis (Blaustein et al. 2010). While this rapid development allows the OSF a better chance at metamorphosis before ponds dry up, their small size at maturity makes these frogs more vulnerable to both predation and desiccation (Donnelly and Crump 1998). Developing tadpoles may also be greatly affected during warmer, drier seasons due to the potential for higher densities of developing tadpoles to compete for shared resources (Donnelly and Crump 1998). Further, such concentrated populations make a highly susceptible concentration of food source for potential predators (Donnelly and Crump 1998). This combination could lead to overall higher mortality rates during this vulnerable life stage. As well, Blaustein et al. (2003) determined an association between high UV-B levels and amphibian mortality.

Another considerable aspect prompted by climate change involves levels of dissolved oxygen within aquatic environments. With warmer temperatures, dissolved oxygen decreases in concentration. Not only does this hinder proper development of amphibian embryos and larvae, but it can also cause physiological stress and behavioral changes to adults (Blaustein et al. 2010).

While only one or two years of significantly high mortality rates in embryos and tadpoles may not hold a significant effect on long-term population dynamics, consistently poor environmental conditions secondary to climate change overtime may severely deplete population numbers, especially for species as recognizably endangered as the OSF (Blaustein et al. 2010). In fact, as the OSF cannot reach sexually maturity until a minimum of three years, it should be considered that the effects of high embryo mortality will not be recognized immediately. Once these changes become recognizable, however, saving the remaining population may prove unmanageable.

2.1.4. Habitat Availability

Amphibian oviposition sites tend to be temporarily inundated, shallow (approximately 5-30 cm), warm pools connected to larger bodies of water that provide placid water movement (McAllister and Leonard 1997, Pearl and Hayes 2004). For example, a subpopulation of the OSF studied by this paper oviposit within the Morris Valley wetland of British Columbia, which feeds into the larger Harrison River. The movement of water is thought to provide optimal oxygenation and cleanse the eggs of algae or fungus', therefore providing a robust qualitative measure of oviposition site selection (Licht 1974). Features such as solar radiation, water temperature, day length, insolation, wind exposure, substrate slope, distance from shore and distance from vegetation may additionally influence site selection among amphibian populations (Pearl and Hayes 2004). Potential oviposition sites that do not meet sufficient criteria may deter adult amphibians from congregating at historical breeding sites, or alternatively, prompt females to reabsorb their eggs and delay attempts at breeding until the next year.

Changes in environmental variables, should then be expected to alter the amount of available breeding habitats, as well as the timing of ideal breeding conditions. Warmer spring time temperatures may result in earlier available breeding habitat due to the melting of the freshet. Conversely, colder temperatures may delay the incoming freshet and therefore limit the water available for breeding ponds to fill. Warmer temperatures will also likely increase the temperatures in the breeding ponds, which may also increase competition, reduce resources and increase levels of UV-B radiation exposure for the OSF (Donnelly and Crump 1998, Blaustein et al. 2010). Colder spring time temperatures could leave breeding ponds covered with snow or ice, limiting available breeding habitat.

The effects of these potential conditions have yet to be formally recognized within the literature, but without question these factors would affect OSF mortality and susceptibility to disease (Blaustein and Kiesecker 2002). If environmental variables are in fact limiting the success of OSF within these localities, ecological engineering provides the opportunity to restore areas of these wetlands to promote successful breeding.

2.2. Materials and Methods

In order to gain understanding into the productivity and patterns of remaining OSF populations, this study examined existing data sets provided by the Canadian Oregon Spotted Frog Recovery Team (COSFRT). This paper acknowledges that attempts to revive this endangered species may be bolstered by specificity, and therefore focused on data in two of its major extant locations, Maria Slough and Morris Valley wetlands – both within the city of Agassiz in British Columbia, Canada – from 2008 to 2017, inclusive. Since recorded discovery of the OSF at these present locations, surveying and monitoring of the populations by COSFRT has been ongoing. Egg mass surveys, in particular, have been conducted in these locations to estimate the size of the breeding population since 2008. More recently in 2011, CMR methods were initiated to assess the abundance, age, size, and health of individuals at these sites. This study draws upon outcomes from both of these methods in the following analysis.

2.2.1. Study Sites

Across British Columbia, ongoing data outcomes have revealed fluctuations to the OSF population size over the last decade (Environment Canada 2014). The Maria Slough and Morris Valley (Figure A. 1) wetlands are two of only three known remaining breeding populations of the OSF in Canada (Environment Canada 2014). These two sites are located within the Fraser Valley floodplain of British Columbia, in the district of Kent. Kent is bound by Harrison Lake to the north, Harrison River to the west, Fraser River to the south, and by Hope and the Fraser Canyon to the east, and thus these OSF populations are surrounded by calm, shallow waters that have historically supported OSF survival and productivity.

Previous radio-telemetry studies have determined the maximum expected movement of OSF as 2 to 3 kilometres (Environment Canada 2014). The distance between Morris Valley and Maria Slough is approximately 8 km, which therefore indicates that these two populations are too geographically separated from one another for movement between populations to occur.

2.2.2. Morris Valley Wetland

Morris Valley (Figure A. 2) is a 13-hectare floodplain wetland, located on Sts'ailes First Nation Band territory, near the Weaver Creek salmon hatchery and fed by the Harrison River. Egg masses of the Morris Valley OSF population *(UTM 49.311812, -121.885501*, Fraser Valley Regional Electoral Area "C") were first discovered in 2008 by the BC Hydro ILM Transmission project. Although the area was privately owned at this time, it was subsequently purchased by the BC Hydro in 2009 as a right-of-way for high-voltage electrical transmission lines (Pearson 2011). Currently, the wetland is in the process of transferring hands to become Crown Land, and potentially an extension of the Harrison-Chehalis Wildlife Management Area (WMA).

Along with neighbouring farmland, this site has been historically subjected to grazing livestock, active beavers, and historical burning to control exotic grass species (Environment Canada 2014). In their habitat summary report, Pearson (2011) described how the 1970s farmers dug a large ditch through the wetland to lower the water table and increase the range area for livestock. However, in combination with climatic change overtime, this topographical alteration has since altered natural fluctuations in water levels throughout the wetland inhabited by OSF (Environment Canada 2014). As a result, the meadows are seasonally inundated during the freshets of the Harrison and Fraser Rivers, with peak flows occurring in June (Environment Canada 2014). Further, the presence of active beavers in this region can enhance structural complexity and overwintering areas for the OSF but can also negatively alter oviposition sites by lowering water levels (COSEWIC 2011). Disturbances from livestock, agriculture, and changing water levels have prompted recognition that the oviposition sites and low-water refuge habitats provided by the Morris Valley wetlands are becoming fewer and shallower overtime (Pearson 2011).

Until as recently as 2014, the American Bullfrog (*Lithobates catesbeianus)* was absent from the Morris Valley wetlands (Environment Canada 2014). However, the 2017 discovery of *L. catesbeianus* egg masses and tadpole capture in this region indicate egg mass survivorship and successful infiltration of this species into Morris Valley wetlands (C.Feischl pers. observation). Additionally, although reed canary-grass (*Phalaris arundinacea*) has been present on-site for many years, this perennial species has recently evolved into a dominant species in areas inhabited by OSF. It follows that increasing competition amongst invasive species adds considerable challenges to breeding ground selection, resource availability, food web dynamics, and survivorship of the OSF.

2.2.3. Maria Slough Wetland

Located east of Agassiz, British Columbia, the Maria Slough wetland (Figure A. 3)*(UTM 49.306155, -121.685541)* measures approximately 15 hectares in size, including an additional 1 hectare that was constructed in 2009 (Pearson 2011). Here, the extant OSF population was discovered in 1997 and extends from private land in the district of Kent through the First Nations Reserve of the Sto:lo Nation Seabird Island band (Pearson 2011). Although this wetland was historically an arm of the Fraser River, its waters became blocked from the Fraser main-stem with construction of the Canadian Pacific Rail embankment in the 1890s (Pearson 2011). A groundwater upwelling provides most of the water to the slough, while other water sources include Hicks Creek to the north and a cold-water spring in the middle of Maria Slough (Pearson 2011). Water exits the wetland by flowing south into the Fraser River, east of Agassiz (Pearson 2011). Much like Morris Valley, the slough becomes fully inundated with fast-flowing water during the spring freshet (Environment Canada 2014). The water level is at its lowest during the breeding season in February and March (Environment Canada 2014).

Despite efforts such as creation of a small breeding pond for OSF near Chaplin Road, recent projects are likely disturbing this population at Maria Slough. For instance, spawning platforms at Hick's Creek have been implemented for Chinook salmon spawning by the Department of Fisheries and Ocean (DFO) and construction of the Canadian Pacific Rail embankment has fragmented the natural OSF habitat (Pearson 2011). Additionally, areas surrounding the Maria Slough wetlands are primarily agricultural, used for dairy, hay, and silage production (Pearson 2011). Such surrounding agriculture activities represent a significant threat to water quality, and therefore the success of OSF at Maria Slough. For example, reports show that algal blooms, which are often caused from excessive nutrient loading, are common to this region (Pearson 2011). Further complicating OSF productivity is the observation that, over recent years, Maria Slough vegetation has become predominantly composed of invasive reed-canary grass (Pearson 2011).

2.2.4. Climate Variables

Temperature and precipitation data used within this study were reported by the AGASSIZ CDA Station, located approximately 9 kilometers from Maria Slough and 12 kilometers from Morris Valley. Minimum, maximum, and mean daily and monthly temperatures were collected for February to April from 2008-2017, inclusive, as these months represent the typical breeding season for OSF. Daily and monthly precipitation values were calculated in millimeters as total daily/monthly rainfall plus total daily/monthly snowfall for February till April 2008-2017. (Agassiz CDA weather station: *Latitude: 49°14'35.000"N. Longitude:121°45'37.000"W. Elevation: 19.3 m. Climate ID: 1100119. WMO ID: 7113. TC ID: WZA. (www.climate.weather.gc.ca))*

2.2.5. Population Monitoring

Egg Mass Surveys

A commonly used technique for estimating the annual reproductive success of various amphibian species is to conduct systemic surveys of egg-mass counts (Raithel et al. 2011)**.** In utilizing these surveys, biologists can make inferences on population size based on the number of egg-masses produced, as well as monitor fluctuations in population productivity. Furthermore, egg masses provide a proxy for estimating the number of females in amphibian populations. The literature posits that the reproductive output of anurans fluctuates across species, perhaps due to various factors such as environmental variables, developmental success, population stability and cyclical reproduction (Pechmann et al. 1989, Blaustein and Wake 1990). On this note, for example, extrapolation from annual egg mass counts suggests that the OSF experienced a 35% population decline in its historical regions of Aldergrove, Mountain Slough, and Maria Slough from 2000 to 2010 (COSEWIC 2011).

Annual systemic surveys of OSF egg masses are conducted by COSFRT at the Morris Valley and Maria Slough wetlands during their breeding season, which spans from late February through to early May. Egg masses are typically oviposited within the months of February and March. In order to identify egg masses, surveyors walk through wetland study sites during the breeding period to look for egg masses and listen for calling frogs. Systemic surveys typically involved 1 to 2 COSFRT researchers visually searching for egg masses by foot or kayak in shallow water. Surveyors varied within and between breeding seasons, and the same surveyor may have surveyed different regions throughout data collection. Egg mass surveys for the entire wetland are conducted upon identifying a minimum of one egg mass and repeated at least once weekly until most of the masses have hatched. Since 2011, with the initiation of CMR studies, observers tend to be present at oviposition study locations throughout the entirety of the breeding season. This has allowed for daily monitoring and recording as new egg masses are laid and hatched. Such ongoing study of OSF breeding and oviposition may have increased accuracy and comprehensiveness of the available data.

The information collected for each egg mass cluster include location (via GPS), number of masses, Gosner stage range, and percentage of dead eggs, as possible. Water depth to both soft and hard substrates were measured using collapsible rulers. Water temperature, pH, dissolved oxygen, and conductivity were measured using handheld metres. COSFRT surveyors maintained daily record forms to track weather conditions, including cloud cover, wind intensity, air temperature, and general condition descriptions. Each egg mass was flagged using coloured metals flags, and surveyors detailed waypoints in one set of descriptive notes that were shared with all surveyors. Actual survey collection dates were recorded in the COSFRT database; however, of note, survey effort hours were inconsistently recorded.

Although Maria Slough surveys began in 1997, the Morris Valley surveys were only initiated in 2008, when the population was first discovered there. Moreover, it is expected that better protocols were implemented and survey efforts improved with each year of survey experience. To capture the most consistent and reliable variables overtime, then, this study opted to evaluate OSF data from Maria Slough and Morris Valley for 2008-2017, inclusive.

Capture-Mark-Recapture (CMR)

From 2011-2017, COSFRT has also monitored the Morris Valley and Maria Slough Oregon Spotted frog populations using CMR methods. Due to missing data, 2014 to 2017 is used for the analysis of CMR data. CMR studies estimate quantitative population sizes and structural population changes overtime by repeatedly capturing, marking, releasing and re-capturing members of the target species, thus providing insight into regional and temporal patterns of the population (Pradel 1996, Courtois et al.

2013). Thus, principal information regarding survival, recruitment, and the overall size of a population can be collected to measure amphibian populations (Pradel 1996).

Between the end of February and beginning of March each year, OSF tend to become active as temperatures climb above 5^{o}C. It is at this time that COSFRT initiate CMR studies by placing funnel traps throughout known OSF breeding regions, including the Maria Slough and Morris Valley wetlands examined by this study. In particular, traps are placed at oviposition sites and along migration corridors, then checked daily and moved according to changing water depths. Trap placement is based on site fecundity, as this species lays their egg masses communally, typically in the same locations as previous years. UTM coordinates are recorded and are also utilized to anticipate oviposition sites for future capture seasons.

Captured OSF are measured for body mass (g), SUL, shank length and sex, if identifiable. Body mass is determined by digital scale, shank length by calipers, and SUL by ruler. To determine previous capture, each individual is scanned for a PIT tag using a Biomark 601 reader, and hind feet checked for florescent dye using a small flashlight. PIT tag number or color of dye are then recorded, or unmarked frogs longer than 40mm and weighing more than 9g are administered a PIT tag by making a small insertion hole with sanitized clippers, manually inserting the tag, and gently pushing the tag towards the rear. Smaller unmarked juveniles are administered injected florescent dye on feet by alternating patterns. All data is recorded on an iPad in the field, then later uploaded to the master COSFRT database.

2.3. Data Analysis

Data was statistically analyzed through Excel. Data was plotted in histograms, bar and plot graphs, and linear regressions applied to determine the presence or absence of relationships. Significance was accepted at $p<0.05$. Data ran as regression tested as normal distribution using the Shapiro-Wilks test.

2.4. Results

2.4.1. Survey Effort

Table 1 shows the total number of survey days for both the Morris Valley and Maria Slough locations over the course of the study period (2008-2017, inclusive). Despite attempts by this study to match sites based on years of active data collection, large variation exists between sites and amongst years. This suggests that bias may have been introduced due to the lack of consistent survey protocols. The greatest survey effort in Morris Valley was 23 days in 2015, whereas this value was only 16 days in 2013 in Maria Slough (Table 1). The least survey effort occurred for 3 days in 2008 in Morris Valley, and 2 days in 2009 and 2011 in Maria Slough (Table 1). These gaps suggest that the data may not accurately represent temporal differences between breeding seasons overtime, posing issues for the questions of this study. The largest same-year difference in survey effort was 10 days in 2013, which attests to the potential for disproportionate capture of data between sites (Table 1).

Table 1. Total number of survey days conducted at each site per year

	Site	
Year	Maria Slough	Morris Valley
2008	10	3
2009	2	4
2010	4	14
2011	2	8
2012	8	10
2013	16	6
2014	11	9
2015	15	23
2016	15	13
2017	11	12

The trend lines in Figure 1 show that survey effort increased overtime. Maria Slough shows a strong level of significance for number of survey days and breeding season year, showing that level of surveying increased over the study period at this site (Figure 1). This increase in effort might explain why an increase in numbers of egg masses were identified for both sites in later years, as opposed to when monitoring first began. For example, in 2009, the number of survey days was only 2 for Maria Slough and 4 for Morris Valley, which corresponded to low numbers of only 43 egg masses at Maria Slough and 29 at Morris Valley (Figure 5). In 2015, in contrast, 15 days of surveying in Maria Slough yielded 118 egg masses and 23 days of surveying in Morris Valley resulted in 85 egg masses.

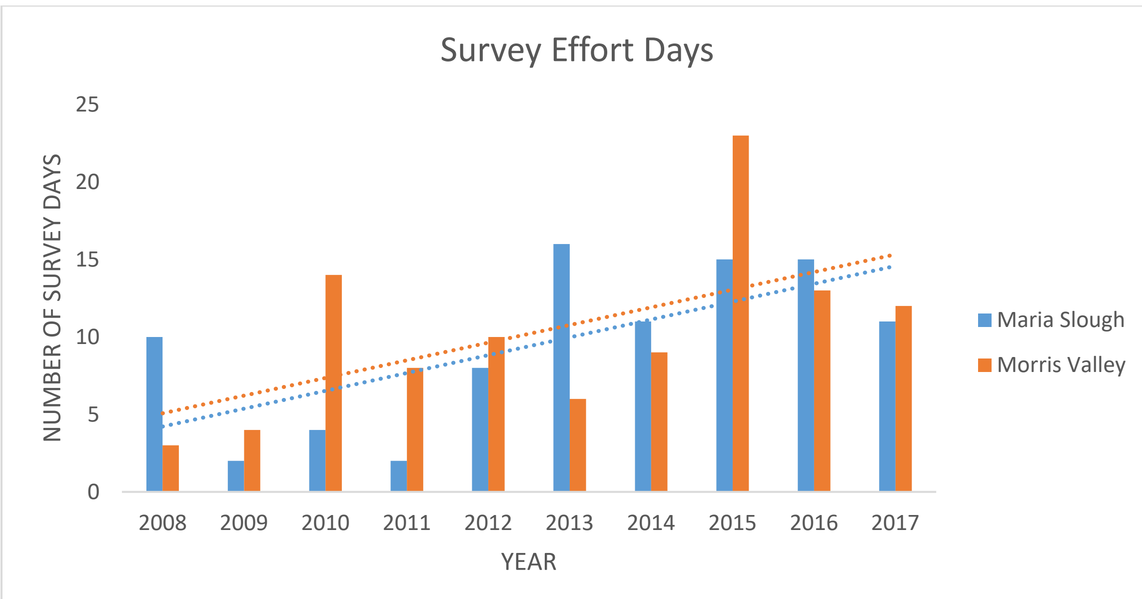

Figure 1. **Survey effort recorded as number of days for Maria Slough and Morris Valley. (R^2=0.64; $p<0.05$ and R^2= 0.23; $p>0.05$ for Maria Slough and Morris Valley respectively).**

In order to determine whether or not a relationship exists between effort and productivity, number of survey days are compared to egg masses discovered at the study locations for each breeding season (Figure B. 1, Figure B. 2). Both sites demonstrate a positive relationship between the number of days surveyed and number of egg masses discovered, however no significance was determined. However, when outliers from 2010 and 2015 are removed, Morris Valley site shows a positively significant relationship between number of survey days and annual egg masses discovered ($p<0.04$; R^2=0.63) (Figure 2). Climatic conditions in 2010 and 2015 corresponded with environmental extremes that involved high temperatures and low precipitation during the OSF breeding season (Figure B. 3, Figure B. 7).

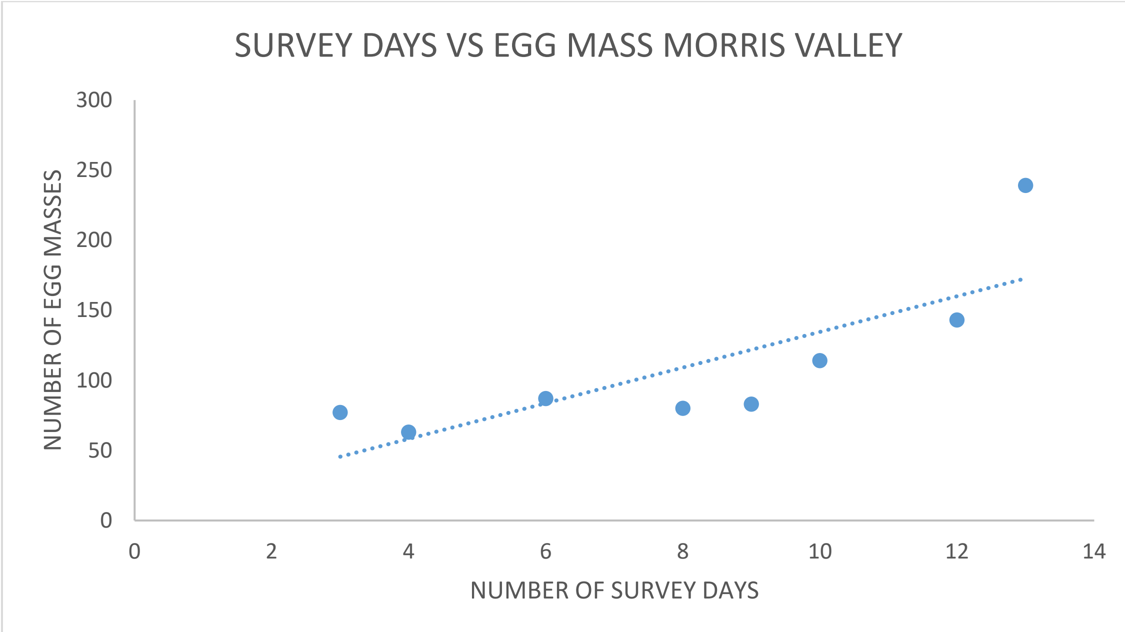

Figure 2. **Number of survey days compared to identified egg masses at Morris Valley excluding outlier years of 2010 and 2015. Results show statistical significance (R^2=0.63419; p<0.04) for a positive correlation between number of egg masses and number of survey days in this study.**

2.4.2. Egg Mass Counts and CMR

Fluctuations in egg mass numbers were observed at both study sites throughout the study period (Figure 3, Figure 4, Figure 5). The greatest and lowest counts, respectively, at Maria Slough were 155 egg masses in 2015 and 35 egg masses in 2017 (Figure 3). These results indicate a 77% decline in egg masses discovered at Maria Slough after 2015.

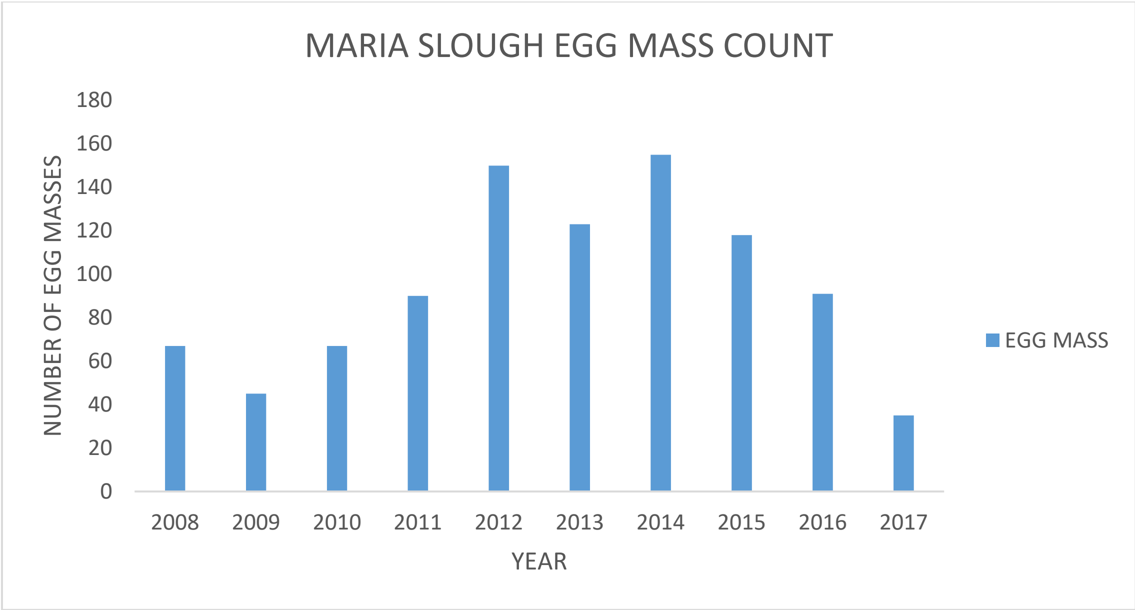

Figure 3. **Results of *OSF* egg mass surveys at Maria Slough from 2008-2017. The population has been affected by fluctuations in egg mass numbers, including a recent decline since 2014. (R^2=0; $p>0.05$)**

The highest number of egg masses surveyed in Morris Valley occurred in 2016 with a total of 239, and the lowest was in 2010 with 39 (Figure 4).

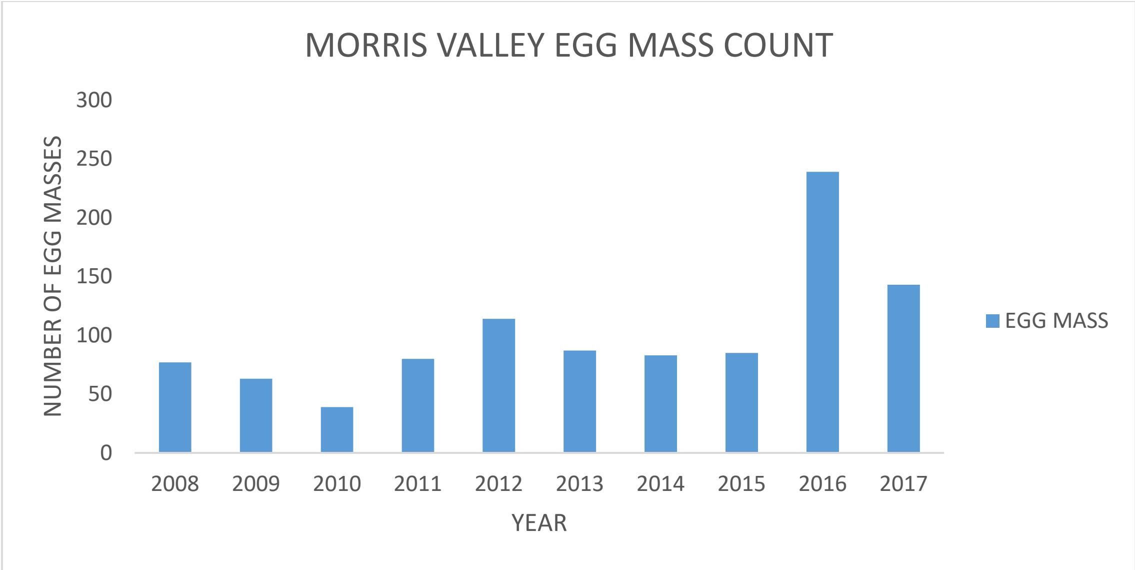

Figure 4. ***OSF* egg mass survey results at Morris Valley Wetland for 2008-2017. Egg mass numbers in Morris Valley have fluctuated overtime, peaking in 2016. *This rise in egg masses may have been attributed to increased survey effort.* (R^2=0.49; $p<0.05$).**

Both populations demonstrate similar fluctuations in productivity which raises the question over what primary variable is causing these fluctuations (Figure 5). However, a substantial increase in Morris Valley egg masses occurred in 2016, which was not observed at Maria Slough (Figure 5). As well, a significant relationship ($p<0.05$) is demonstrated between number of egg masses and study year for Morris Valley. This significance may not be representative of an increasing population but attributed to the sudden increase of egg masses that were discovered in 2016.

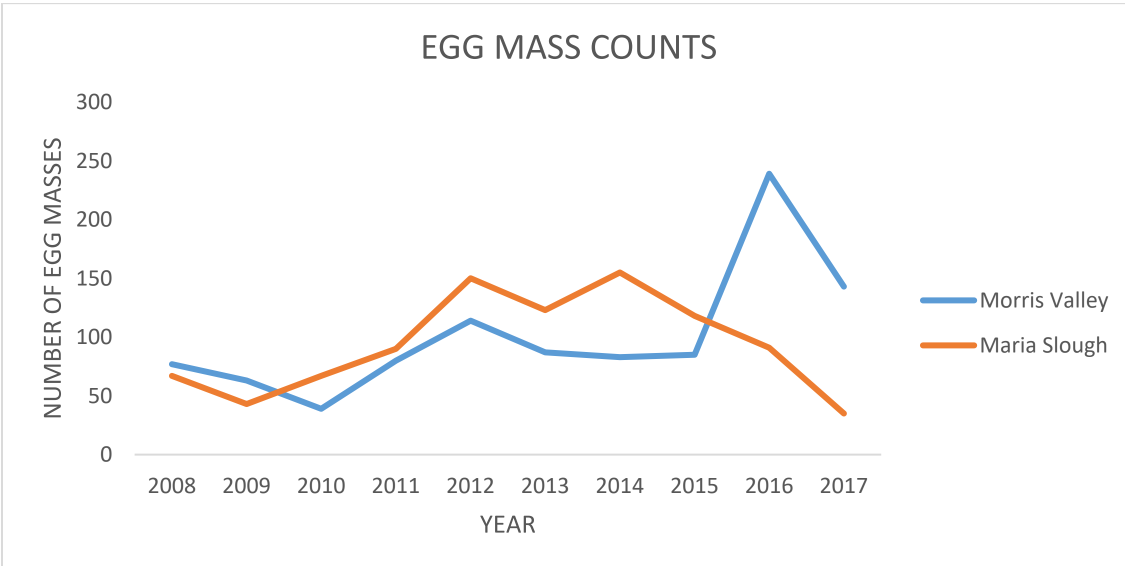

Figure 5. ***OSF* egg mass survey results compared between Morris Valley wetland and Maria Slough for 2008-2017, inclusive. Both populations have experienced fluctuations within the populations overtime and Morris Valley shows significance relating egg mass numbers to study year. (R^2=0; $p>0.05$; R^2=0.49; $p<0.05$ for Maria Slough and Morris Valley inclusively).**

Results from ongoing CMR studies is investigated from 2014 to 2017 at both Maria Slough and Morris Valley and demonstrate that Morris Valley has increased in capture rates over time. 2016 yielded the highest overall number of captured individuals in Maria Slough, with a total of 283 events; the lowest capture year for Maria Slough was in 2017 with 157 events (Figure 6). 2017 saw the greatest number of captured individuals in Morris Valley with 281; the least number of CMR events in Morris Valley occurred in 2014, with 158 events (Figure 6). Also of note, Maria Slough demonstrated a significant decrease in CMR events from 2016 to 2017, falling from 283 to 157 (45% decrease), while CMR events in Morris Valley steadily increased from 2014 to 2017 (Figure 6).

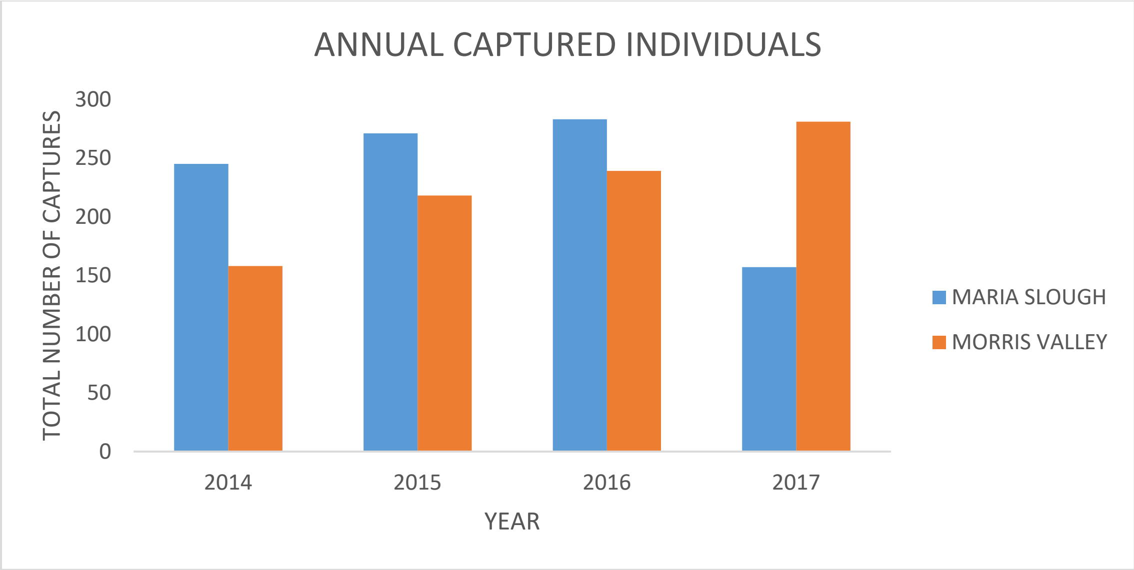

Figure 6. **Annual Capture-Mark-Recapture results from both Maria Slough and Morris Valley from 2014-2017. Number of captures has increased over time at Morris Valley, whereas capture numbers declined most recently at Maria Slough.**

Results varied between CMR and egg mass survey results, as illustrated by female sampling throughout this study. While CMR provides direct evidence of females within the population, egg masses provide an indirect estimate of the minimum population of viable females per breeding season. The number of females captured at Maria Slough fluctuated between 27 to 55 individuals (Figure 7). Through CMR methods, for example, the highest number of female OSF captured at Maria Slough was 55 in 2015, out of 271 total OSF captures (Figure 7). In other words, females captured during this breeding season represented only 20% of the total captured individuals. When considering that 118 egg masses were surveyed during 2015, this suggests that less than 50% of the female population was captured by CMR. 2014 shows the largest discrepancy between CMR and egg mass outcomes for females, as only 28% of the minimum female population was captured at Maria Slough (Figure 7).

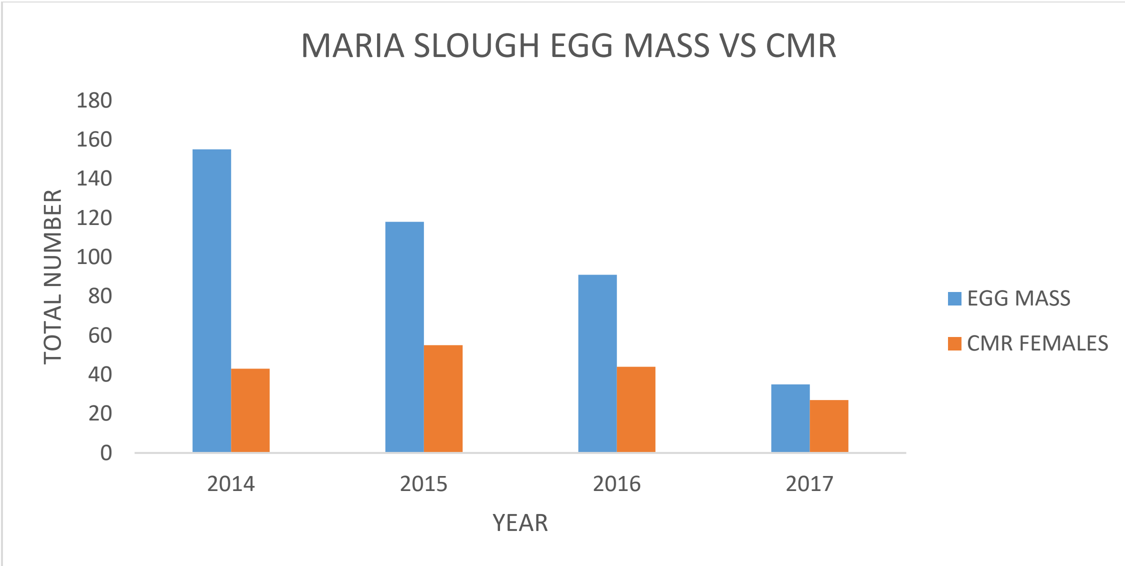

Figure 7. **Capture-Mark-Recapture events during breeding season for 2014-2017 compared to number of egg masses at Maria Slough.**

The number of females captured at Morris Valley fluctuated between 29 to 70 individuals (Figure 8). The highest number of captured total individuals at Morris Valley was in 2017 with a total of 281 (Figure 8), only 56 of these captures were female which represented 39% of known females based on egg masses. In 2016 however, the highest number of female captures occurred with 70 females out of 239 (29%) total captures, representing 29% of the female population based on egg mass numbers for that year. The least females were captured at Morris Valley in 2015, with 29 events (Figure 8). Females captured during this breeding season represents only 13% of the total captured individuals, and only 34% of the minimum female population when considering the 85 egg masses surveyed at the site that year.

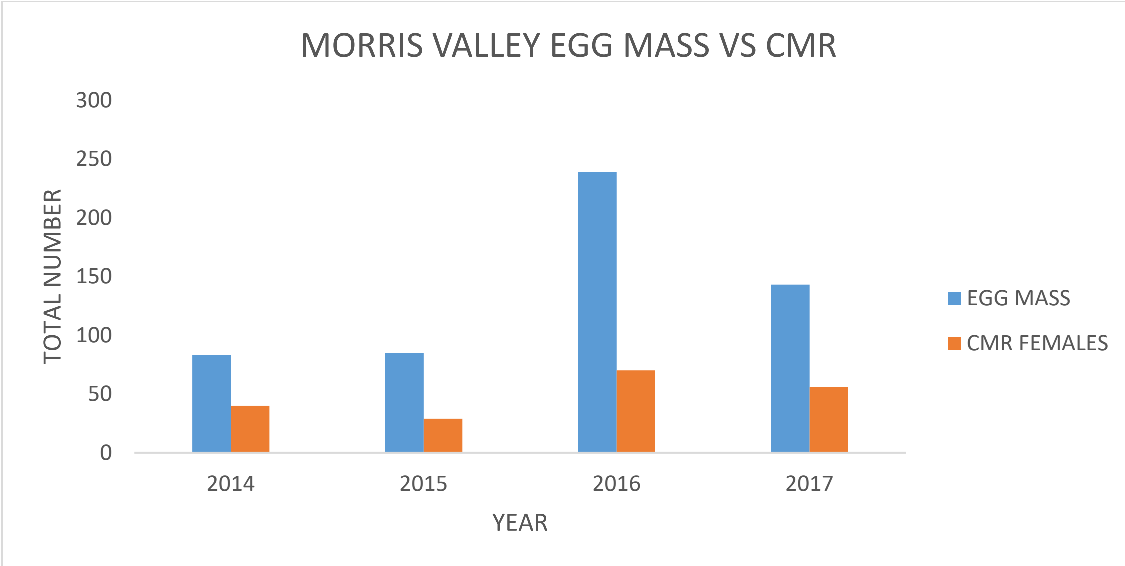

Figure 8. **Capture-Mark-Recapture during breeding season compared to number of egg masses at Morris Valley from 2014-2017.**

2.4.3. Egg Mass Related to Temperature and Precipitation

This study considered both minimum and maximum average monthly and breeding season temperatures by relating temperature statistics for Maria Slough and Morris Valley wetlands with egg mass production. Based on the trend lines, average monthly breeding temperatures from February to March have increased slightly over the course of the study, as shown in (Figure B. 3, Figure B. 4).

For February, the coldest year was in 2014 with an average monthly low of -0.7^{o}C and warmest in 2015 with a high of 12.3^{o}C. The coldest March was in 2009 with a low of 0.4^{o}C, and warmest in 2015 with a high of 14.6^{o}C. Across the study years the coldest breeding season was in 2009 with an average low of 0.5^{o}C and the warmest in 2015 with an average high of 13.5^{o}C.

No statistical relationships were detected between minimum or maximum temperatures and number of egg masses produced per year. However, a number of trends were observed.

Breeding seasons with cold minimum temperatures demonstrated lower productivity of egg masses. For example, in 2009 the coldest recorded year, both populations represented low numbers in egg mass productivity where 45 egg masses were discovered at Maria Slough, and 63 at Morris Valley (Figure B. 6). During another cold breeding season in 2017, Maria Slough demonstrated the lowest recorded egg masses over the study period with only 35. This same year Morris Valley dropped by 96 egg masses from the previous year.

Breeding season precipitation represents total precipitation for February through March, which comprises the oviposition season for OSF, across study years of 2008-2017, inclusive (Figure B. 7). The driest breeding season was in 2010 (144mm) and the wettest in 2011 (365mm). A slight, but non-significant increase occurred overtime, and there is a high variability of average breeding season precipitation. Breeding season precipitation levels are graphed alongside egg mass counts at each site in Figure B. 8. No significant relationship was demonstrated between precipitation levels and number of egg masses produced during breeding seasons at Morris Valley or Maria Slough.

2.4.4. Timing of Egg Mass Deposition

Although date of first seasonal egg mass discovery was available for most years over the course of the study, this data was missing for the Maria Slough population in 2008 and 2012 (Table 2**).** The start of oviposition varies over 40 calendar days throughout the study period. The average date for oviposition at Morris Valley is around the 71st day of the year, and Maria Slough around the 65th day of the year.

Table 2. Date of first *OSF* egg mass deposition discovery at Maria Slough and Morris Valley sites, with corresponding climate data for that day (average daily min-max temperature (ºc), precipitation (mm)). Data points were unavailable for Maria Slough in 2008 and 2012.

Year	Maria Slough (Temp; Precipitation)	Morris Valley (Temp; Precipitation)
2008	N/A	March 3 (3.3-5.9ºc; 10.2mm)
2009	April 3* (3.2-11.4ºc; 0mm)	March 15 (1-4.5ºc; 7.4mm)
2010	March 12 (2.4-8.4ºc; 21.6mm)	February 26 (6.2-8.6ºc; 4.8mm)
2011	March 28 (6.5-11.6ºc; 1.6mm)	March 16 (4.7-10.3ºc; 10.2mm)
2012	N/A	March 6 (-0.3-6.9ºc; 0.3mm)
2013	March 12 (3.1-8.2ºc; 53mm)	March 3 (2.1-9.5ºc; 0mm)

Year	Maria Slough (Temp; Precipitation)	Morris Valley (Temp; Precipitation)
2014	March 13 (2.8-14.4^{o}c; N/A)	March 13 (2.8-14.4^{o}c; N/A)
2015	February 22 (3-12.5^{o}c; 0mm)	February 26 (5.1-9.8^{o}c; 1.2 mm)
2016	February 24 (6.2-12.8^{o}c; 0mm)	February 25** (4.2-15.6^{o}c; 0mm)
2017	March 14 (7.2-11.6^{o}c; 24.3mm)	March 11 (3.9-7.1^{o}c; 21.9mm)

*First date egg masses identified, may have been deposited earlier ; **Older egg masses identified suggesting breeding starting as early as February 18

Table 2 shows that the earliest date of oviposition (calendar day number) recorded between both sites was at Maria Slough on February 22, 2015 (53), and the latest egg mass, also discovered at Maria Slough, was on April 3, 2009 (93). It should be noted that this egg mass may have been laid at a date prior to its discovery by researchers. The earliest egg mass discovery date at Morris Valley was February 25, 2016 (55) (Table 2), although this egg mass was believed to have been deposited as early as February 18 (48), thus possibly substantiating the earliest recorded date of oviposition among both sites.

Table 2 shows that both sites had the same, early deposition dates in February of 2015 (53). Corresponding precipitation levels for this month were relatively low compared to other years, at 178 mm (Figure B. 7). Furthermore, maximum temperatures in February of 2015 were the highest recorded during this study period, averaging 12.3^{o}C (Figure B. 3). The earliest recorded deposition date at Morris Valley in 2016 corresponds to average February temperatures of 11^{o}C and precipitation of 168.8 mm. These variables, much like 2015 represent drier and warmer conditions over the study period. Notably, 2016 was also the second earliest deposition date at Maria Slough.

In comparison, 2011 boasted the latest oviposition discovery date in Morris Valley; 2011 also corresponds to the second latest recorded oviposition discovery at Maria Slough (Table 2). The lack of survey visits in 2009, however, suggests that its April 3 oviposition discovery date may represent inaccurate timing of data collection and therefore 2011 may in fact represent the latest date of oviposition for Maria Slough as well. From here, we see that the average breeding season precipitation in 2011 increased to 365mm, the wettest during the study period (Figure B. 3), and the average temperature during breeding season temperature was between 1.05-8°C, one of the lowest during the study (Figure B. 5).

Minimum and maximum temperatures were averaged over the breeding season and compared to first date of oviposition at each study site (Figure 9, Figure 10). The results exhibit statistical significance relating temperature to timing of oviposition at both study sites ($p<0.001$). The results demonstrate that oviposition is instigated by warmer temperatures. In breeding seasons with colder temperatures, oviposition is delayed later into the breeding season.

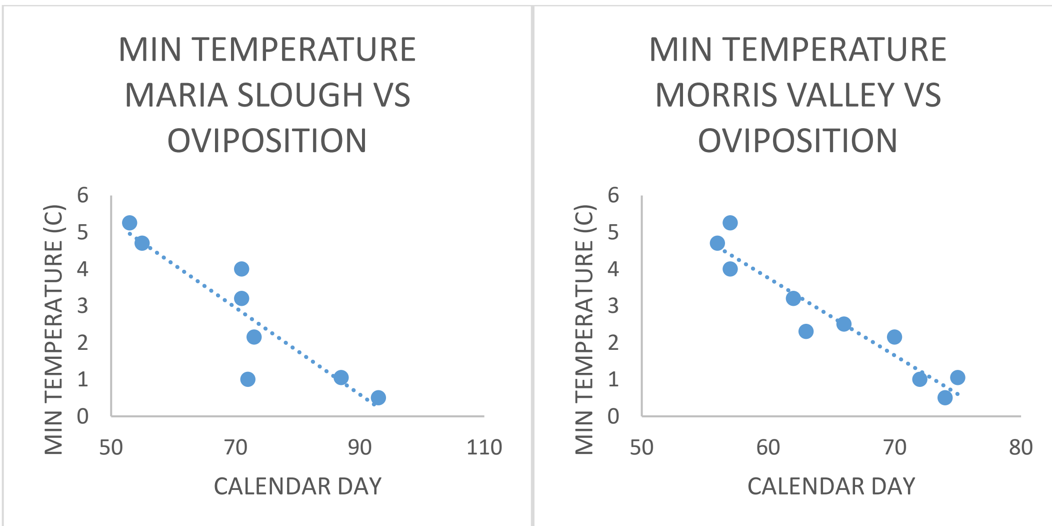

Figure 9. **Minimum breeding season temperatures (February-March) are compared to the first calendar day of oviposition at both (a) Maria Slough ($p<0.001$; $R^2=0.73$) and (b) Morris Valley ($p<0.001$; $R^2=0.94$). These results show significance relating onset of breeding with minimum temperatures.**

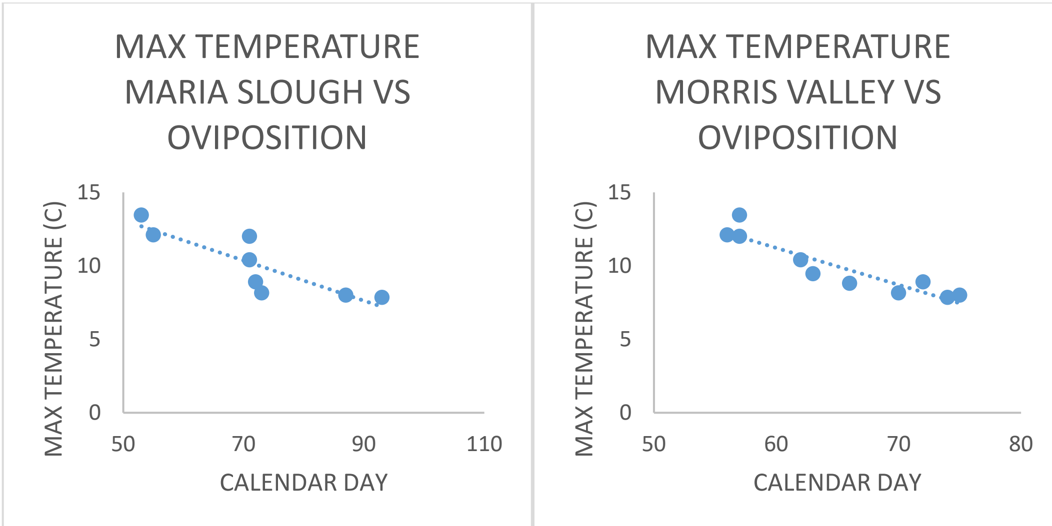

Figure 10. **Maximum breeding season temperature (February-March) are compared to the first calendar day of oviposition at both (a) Maria Slough (p<0.001; R^2=0.71) and (b) Morris Valley (p<0.001; R^2=0.88). These results show significance relating onset of breeding with maximum temperatures.**

Precipitation during February and March were also compared to first date of egg mass discovery at both sites throughout the study period. The trend lines suggest that in years with less precipitation, egg mass deposition occurs earlier in the breeding season, and conversely years with increased levels of precipitation oviposition is delayed. This is represented significantly at Morris Valley (p<0.02)(Figure 11).

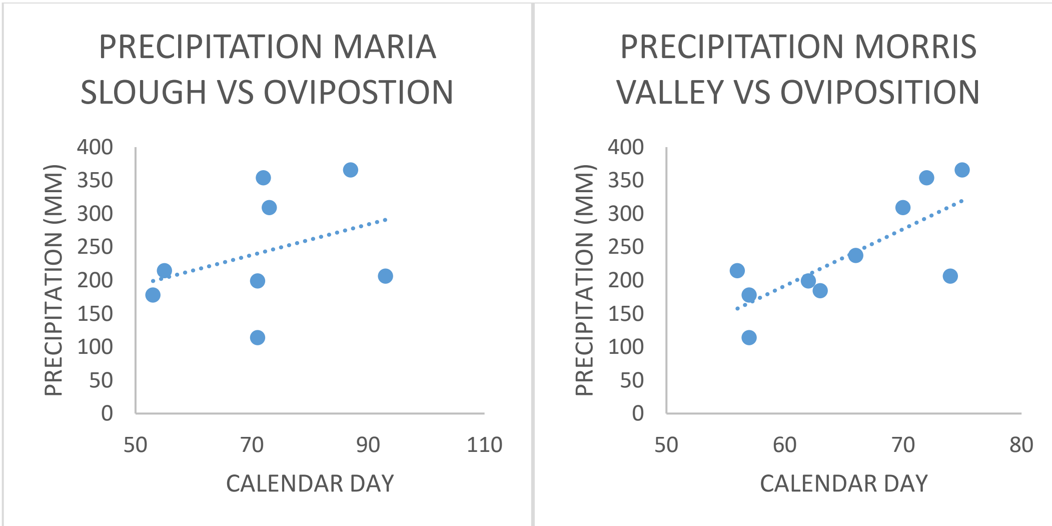

Figure 11. **Precipitation during breeding season (February-March) is compared to the first calendar day of oviposition at both (a) Maria Slough ($p>0.05$; $R^2=0.34$) and (b) Morris Valley ($p<0.02$; $R^2=0.58$). Maria Slough did not show significance, however, Morris Valley showed significance relating precipitation to onset of breeding.**

2.5. Discussion

Population monitoring is useful for assessing trends in species productivity and population status overtime (Scherer and Tracey 2011), as well as indicating the health of the overall ecosystem. Such monitoring provides researchers insight into significant population changes, which aids specifically in preventing species decline or extinction (Scherer and Tracey 2011). Additionally, population monitoring conducted scientifically, with a priori hypothesis, can counter management decisions that might be passed too quickly or with limited/incorrect data or supporting evidence (Pollock et al. 2002).

As such, this study argues that the primary cause(s) of the decline of the OSF must first be determined so that strategies may be developed towards helping this species persist within its northern niche. From here, this study provides species information that should be accurately collected in order to best study these endangered populations overtime, especially considering that environmental variables such as temperature and moisture regimes are expected to continue to change. In doing so, the primary objective of this study is to improve the baseline knowledge of the status of the OSF within the Morris Valley and Maria Slough wetlands, with a focus on environmental conditions as possible determinants for successful OSF breeding. Recommendations are also proposed for future monitoring and potential restoration projects as the effects of global climate change become increasingly concerning for this species. As well, the implementation of a citizen science program is discussed.

2.5.1. Population Monitoring

For the last 10 years, OSF populations at Morris Valley and Maria Slough have been monitored using systemic egg mass surveys. The results of the data illustrate what appear to be natural fluctuations in these populations, as shown in Figure 5. Based on this data, both populations saw a decline in population numbers in 2017; however, the Maria Slough population has shown a steady decline without any evidence of population increase since 2014. These observations spark concern that these OSF populations may be at risk of extirpation. Still, the Morris Valley population did experience a significant growth in egg mass counts across study years ($p<0.05$) which was not observed for the Maria Slough population, which suggests that breeding habits within Morris Valley may be less susceptible to environmental variables than the Maria Slough population.

While counts of egg mass numbers are a valid state variable to monitor productivity patterns among pond-breeding amphibians, the literature lacks evidence that this method is capable of representing changes in overall OSF population status, perhaps due to differences between egg mass counts and actual embryo survival. Previous studies that have looked at the statistical significance of wildlife monitoring programs have suggested that selection of an appropriate state variable is imperative to population monitoring (McKelvey 2006)**.** Not only must this variable reflect population status, but any changes in population size should have a high probability of being detected by this chosen variable. Through this logic, egg mass counts alone can neither represent alterations in actual OSF population size nor indicate causative factors influencing population productivity. However, considering the limited available population trend data for the studied OSF populations, this study utilized egg mass counts as a state variable in the context of breeding productivity. By examining causative environmental factors influencing OSF population size, then, this study drew comparisons between numbers of OSF egg masses and the environmental variables of temperature and precipitation.

Time series studies, such as the 10-year data used within this study, tend to involve high levels of variance that may confound accurate representations of species decline (Green 2003; Buckley and Beebee 2004)**.** In anticipation of this limitation, study protocols must be established and strictly enforced across study years and sites in order to draw reliable scientific comparisons among distinct populations. The use of two study sites, however, allows for comparisons between the populations to be made in order to determine whether or not population trends are merely natural cycles or instigated by a stressor. The fact that this study examined two populations within a shared climate (*e.g.,* the Maria Slough and Morris Valley sites are located within close proximity to each other) improved this study with regard to relationships between environmental variables and OSF egg mass productivity. Climatic data in this study was generalized, as it was collected from one weather station near both of the study sites. This entails reduced specificity, but the author upholds that this outcome should be taken in context of the similar climatic and geographical conditions shared by both wetlands. Still, this study proposes that future studies could focus on microclimate data in order to draw more specific conclusions regarding the influences of temperature and precipitation on OSF populations. Future studies might consider site specific monitoring stations to collect ambient air temperature, precipitation, and water temperature and depth to improve accuracy of climatic data.

A key observation by this study was that when survey effort increased, higher overall numbers of egg masses were discovered. This finding was particularly significant when outliers representing years of environmental extremes were excluded for the Morris Valley population ($p<0.04$). The findings of such results underline the importance of maintaining consistent survey effort across both study sites and years to prevent inconsistencies in population-level data. This study therefore maintains that it is imperative that future OSF studies abide by fixed standards in order to strengthen reliability and conclusions.

In addition to egg mass counts, CMR studies have been conducted since 2011 and are expected to continue into 2018. Data, however, was only available for years 2014-2017 for this study. Furthermore, the results of CMR are used to estimate population size, as opposed to answering a priori hypothesis (e.g. scientific objective) (Polluck et al. 2002). In this sense, the primary objective for CMR data collection of these populations has been primarily management oriented.

This study aims to highlight that CMR may incur more negative consequences - such as financial cost, time expenditure, (Polluck et al. 2002) and risk for habitat destruction that may occur during survey periods – that may in fact outweigh beneficial research or management outcomes. CMR studies must be conducted in a manner inclusive of strict study designs and sampling methods, in order to produce significantly reliable results (Polluck et al. 2002; Petit and Valiere, 2006). Therefore, a large amount of effort must be expended to ensure replicability is maintained from year to year (Lettink 2012). In this regard, this paper echoes previous reports that suggest the costs of CMR outweigh its benefits for elusive species such as the OSF, and that resources should be more wisely allocated towards other methods of study (Polluck et al. 2002) such as egg mass survivorship.

This claim is substantiated by the data collected within this study. Considering that female OSF are only capable of producing one egg mass per breeding season, we would expect CMR to identify a relationship between female OSF captured by CMR and the number of egg masses identified by wetland surveys. Although the results did demonstrate a relationship between caught females and egg masses at Maria Slough ($p<0.05$), no correlation was found between these variables for the Morris Valley population. Therefore, the results from this study posit that CMR is not consistently reliable for determining female OSF population counts across geographical sites. This discrepancy prompts consideration that CMR is also unreliable for providing proper estimates of male OSF and total population size. Thus, this study proposes that CMR proves both insufficient and inaccurate for sizing a given OSF population size and gender demographics. Still, it should be noted that researchers may find value in using CMR as a secondary method of population observation to monitor longevity and health of individuals. As well, CMR may be used as a valid measure to assess habitat use within a wetland. Importantly, however, CMR should not be conducted during the sensitive breeding season.

2.5.2. Quantity and Timing of Egg Mass Deposition

Previous studies have shown that climate change has the potential to cause changes in reproductive timing for amphibian species (Licht 1971, Todd et al. 2011). For the OSF, in particular, temperature has been theorized as a significant determining factor for the onset of breeding (Licht 1971). With this consideration, this study utilized daily air temperature as a proxy for breeding activity, as Licht (1971) has shown higher temperatures to correlate with OSF breeding activity and egg mass production. Breeding is most likely to occur during the daytime, when maximum temperatures are reached (Licht 1971). Even further, results from this analysis add new data that minimum temperatures also delay OSF oviposition. The onset of oviposition was proven statistically to be related to temperature in that years with more extreme minimum average temperatures, resulted in delayed onset of egg mass deposition. This could have negative consequences for the development of tadpoles, as a delay in oviposition will delay the time to metamorphism, threatening tadpoles with stranding or even desiccation as breeding ponds dry up. Conversely, with warmer temperatures earlier in the spring, oviposition may be instigated prematurely, warranting concern that tadpoles will be exposed to the risks of flash cold weather. Also of consideration, colder temperatures lead to cooler water temperatures, and water temperatures that drop below the minimum threshold of 6^{o}C increase the susceptibility of OSF embryos to freezing (Licht 1971).

The data shows that precipitation through breeding seasons may also play a role in OSF oviposition onset. As shown in Table 2, dates of oviposition during the study period varied by as much as 40 calendar days, indicating that the timing of breeding season onset varied considerably between study years. As true for any species, this shift in reproductive timing for the OSF likely impacts other members of the community. At both Maria Slough and Morris Valley, the onset of oviposition correlated with breeding season precipitation, a result that proved significant for the Morris Valley population ($p<0.05$). In years with drier breeding seasons, oviposition was initiated earlier than years with more precipitation. These drier conditions are most likely correlated with warmer temperatures, and therefore the environmental variables that instigate OSF to breed and consequently lay their eggs. The negative consequences of earlier oviposition must be considered here. If OSF oviposit when precipitation is low, then egg mass survivorship may be conditional on water levels in breeding ponds because lower levels of precipitation leave egg masses more susceptible to desiccation. Although not examined by this study, lower levels of precipitation during February and March may also be correlated with years of lesser snowpack. Such conditions could incur lower levels of freshet, therefore inciting water stressed conditions relatively earlier in the year. Not only might egg masses be lost due to desiccation, but tadpoles would be threatened by shortened developmental periods. Tadpoles may metamorphose into smaller individuals as a result, leaving them more susceptible to predation and higher surface area-to-volume ratios that experience greater degrees of moisture loss (Donnelly and Crump 1998).

It is also possible that low precipitation levels created unfavourable conditions that prevented some females from ovipositing during these drier seasons (Donnelly and Crump 1998). As floodplain wetlands, the OSF depends highly on melting freshet to fill breeding ponds prior to oviposition. In years with relatively low snowfall, these oviposition sites may fill with shallower water levels that delay oviposition. In contrast, winters with more precipitation result in higher levels of snowpack, which could yield more available breeding habitat once the freshet floods the breeding wetlands. On this note, future studies may consider how OSF oviposition dates are affected by varying levels of accumulated snowfall and their corresponding freshet levels, as data available for this study was limited to monthly precipitation levels. For example, in 2016, a substantial increase in egg mass numbers was seen at Morris Valley in concurrence with significantly higher water levels during breeding season, but it is difficult to determine how much additional breeding area was added once the accumulated freshet melted. It may be for this reason that no significant relationship was determined between egg mass counts and precipitation levels alone.

As another consideration, earlier seasonal flooding may occur due to a combination of increased seasonal temperatures, precipitation and an earlier and larger freshet, and such conditions may increase available breeding habitat by providing a larger area of breeding water within floodplains. In support of this notion, egg masses were more widely geographically distributed in 2016 at Morris Valley, thus implying that the OSF pond-breeding habitat was broader this year in comparison to others.

Further, Table 2 shows that the earliest dates of oviposition at both sites (February 24th at Maria Slough; February 25th at Morris Valley) occurred on days with 0 mm of precipitation and relatively higher maximum temperatures. It follows that a combination of increased temperatures and lower precipitation levels may indicate superior breeding conditions for these populations at the level of daily analysis.

In combination, the observations set forth by this paper highlight the importance of not only assessing the environmental conditions during breeding season, but also considering daily climatic conditions as well as conditions during months prior. For instance, trends may be observed related to the amount of snowpack that explain some of the patterns observed in breeding and oviposition behaviour in these populations.

2.5.3. Embryonic Survivorship

Embryonic survivorship studies promise insight into relationships between environmental variables and successful reproductive output of OSF. In order to utilize egg mass counts as a measurement of population size, the number of breeding adults should correlate with the number of egg masses observed, and also the number of egg masses observed should approximate reproductive success (Richter et al. 2003). While egg mass surveys provide a useful tool for monitoring annual variations in reproductive efforts of pond-breeding amphibians, these trends do not provide quantitative evidence for the embryonic survivorship rates. Whereas researchers have proposed that the number of tadpoles produced per egg mass be used as an indicator of breeding success (Buckley and Beebee, 2004), this study notes that egg mass data at Morris Valley and Maria Slough was underpowered and therefore inconclusive. From here, this discussion elaborates on the need for further research in the area of embryonic survivorship as a means towards successful study and protection of the OSF.

Embryonic survivorship probability entails that egg mass and/or ova number represent potential population recruits, rather than actual members of the population. An individual with high capacity for producing offspring is recognized as having a high level of fertility. In some amphibian species, such as the OSF, fertility is often correlated with female body size, as the larger bodied females produce more ova per egg mass (Licht 1974). In turn, the number of ova in each egg mass combined with the degree of embryonic survivorship represents the fecundity of a female.

Population size is ultimately determined by both birth and death rates. Where adult population size may be theoretically adequate to maintain a viable population, low embryonic survivorship may stunt population growth; in contrast, small population sizes may experience growth if embryonic survivorship is high (Semlitsch 2002 from McKibbin et al. 2008). It has been proposed that amphibian species have evolved to produce large numbers of offspring in order to compensate for low rates of embryonic survival. Within its Canadian range, for example, the OSF produces approximately 249 to 935 ova per egg mass (Licht 1974). Within the context of this theory, it follows that climate change may impose stressful conditions that further hinder embryonic survivorship and consequently population growth (Licht 1974).

Here, this paper underscores the lack of available data surrounding embryonic survivorship for the OSF at its northern limits. A literature review did yield a study by McKibbin et al. (2008) that examined survivorship of egg masses within the Canadian range; however this research was notably limited by study design, as observations were performed under artificially constructed conditions. For example, this study employed study cages, thus eliminating the threat of predation on egg masses. In another study, Licht (1974) estimated the natural mortality of embryos within egg masses, and, as a result, offered guidance for obtaining accurate estimates of embryonic survivorship in natural conditions (Licht 1974). It was noted in this study that due to the shallow water level oviposition condoned by OSF, a number of egg masses would have desiccated had intervention not been imposed (Licht 1974). This behavior in OSF is believed to have evolved where the threat of desiccation was non-existent, and these local populations have yet to adapt to the dry breeding conditions in the Lower Mainland (Licht 1974). This form of human intervention has been conducted at study sites in the past, influencing the survivorship of egg masses, even in years during this study period, 2008 to 2017. It is imperative for future OSF embryonic survivorship studies to mandate specific protocols that indicate whether human intervention occurs, as well as which specific egg masses are affected, as such actions will skew data outcomes.

Even further, this study advocates that accurate measurements of reproductive OSF output can only be gauged by examining embryonic survivorship successes for egg masses unaffected by human intervention.

2.5.4. Survey Limitations

From its outset, this study was limited by relatively scarce literature and existing data pertaining to the OSF. Also, as discussed above, data examining factors such as OSF embryonic survivorship are virtually non-existent for the studied populations and therefore could not be studied with regard to temperature or precipitation. This substantiates a primary argument of this study that more research surrounding OSF productivity and population dynamics is warranted. In effect, the suggested protocols and evaluation of existing research methods provided by this paper are intended to encourage reliable, accurate examination of this species in future research.

One significant issue with this study is potential inaccuracies involving survey data. Maria Slough is missing information related to oviposition timing, including dates that may have been incorrectly recorded. Stricter survey protocols for future data collection efforts should aim to ensure that oviposition dates are accurate, which in turn will provide more accurate conclusions regarding relationships between oviposition and changing environmental variables such as temperature and precipitation.

Furthermore, this study determined inconsistencies of survey effort amongst years and between sites. Insufficient guidelines and protocols were likely responsible for these deficiencies, which unfortunately render the data outcomes less reliable. For example, both sites varied in the amount of survey effort days and number of observers. Establishment of more comprehensive survey protocols would promote consistent effort between geographical sites and across survey years, thus rendering more comparable results between Maria Slough and Morris Valley.

Relatedly, several years of Maria Slough data were accrued by only one surveyor; in contrast, other years for both sites involved multiple surveyors. This limitation presents a risk for potential bias, reduced diversity in observations, and possible exclusion of certain areas from being explored for egg masses. At minimum, this study advocates for the establishment of double-observer protocols for future years of study in order to prevent such shortcomings (Pollock et al. 2002; Faccio 2011; see Figure A. 5 for survey protocols used in COSFRT reports).

As has been concluded in other studies, an important facet of population studies involves training for the individuals conducting systemic surveys (Faccio 2011). While all surveyors underwent training, it is possible that some surveyors were better trained or more experienced than others, which may have been reflected in the data collected. One method for countering this limitation in future studies may be to use the same surveyors at both sites and across years, as possible.

Considerable funding has been allocated to conducting the annual surveys evaluated by this study, and therefore these projects might benefit from defining additional guidelines that ensure the data is collected and recorded reliably. An example would be providing all surveyors with a shared standardized table to enter their data and reviewing how this data should be entered during the training session. Training should also address foundational education on the appearance and behaviours of the OSF, especially considering that this species shares its habitat with other pond-breeding amphibians and egg masses that may be mistaken for those of the OSF.

The consequences of these limitations are illustrated by the relationship between survey effort and number of egg masses found at each site. During 2010 and 2015, for example, high temperatures and low precipitation levels correlated with reduced egg mass productivity in Morris Valley, thus substantiating the conclusion that extreme climate conditions influenced OSF productivity. Also for these two years, greater survey effort was observed. As such, these two years were identified as outliers and excluded from analysis, which resulted in a significant positive relationship between survey effort and egg masses numbers. This suggests that greater survey effort may lead to discovery of more egg masses. This discrepancy should emphasize the importance to standardize protocols, training and data collection methods when testing these relationships in the future.

In combination, the limitations discussed may have prevented this study from detecting significant relationships between egg mass productivity, temperature and precipitation. Once these limitations are rectified to yield more reliable data outcomes, potential relationships between climatic variables and OSF productivity should be re-examined.

2.6. Conclusion

The data used in this study offers valuable baseline data to begin assessing OSF trends at the northern limit of its range. This paper also proposes that changing climatic conditions will affect reproductive output for this species, but further research is warranted to implement stricter protocols towards assessing dependent climatic variables such as temperature and precipitation.

This study showed a statistical relationship between the timing of initial OSF oviposition and both ambient air temperatures and precipitation levels during the breeding season. Notable procedural observations may explain why this study was unable to conclude any significant associations among egg mass counts and environmental variables at the locations studied. Firstly, the data has determined to be unreliable in producing consistent statistically significant results at both sites, as an unreliable level of survey effort and data recording has been condoned. Secondly, the data suggests a correlated relationship between the number of survey days conducted and the number of egg masses found, statistically so at Maria Slough.

Furthermore, this paper demonstrated that CMR failed to accurately represent the breeding female population at Morris Valley, as determined by discrepant results between female CMR events and egg mass counts. This finding suggests that CMR should not be considered a reliable estimator for OSF breeding habits or population size.

This paper seeks to emphasize that productivity trends evaluated by this study, as evidenced by egg mass counts, were difficult to correlate with environmental variables due to discrepancies in data collection. However, high environmental fluctuations show fluctuating productivity, while steady environmental variables across years seem correlated with steady productivity at both sites.

2.6.1. Recommendations for Future Monitoring

In order to properly assess the causal variable for decline, a number of suggestions are made for future population monitoring efforts.

It is recommended that egg mass surveys be utilized as the primary method for assessing OSF population trends over time and between populations. CMR studies provide useful insight to the health of individuals, but notable drawbacks – such as reduced reliability for population monitoring, financial and time investments and its potential for habitat destruction – prompt consideration that CMR studies do not necessarily need to be conducted on an annual basis. In fact, this study proposes that CMR studies be limited to every 3-4 years and focus on capturing anatomical behavioural observations rather than population trends. The effort and resources for CMR studies may instead be reallocated to more constructive scientific studies.

Since 2009, survey reports submitted to the Ministry of Environment have encouraged that population study designs include multiple-observer surveys (Pearson 2009). In a dependent double-observer protocol, two observers survey a pool together with observers trading off between primary and secondary roles (Nichols et al. 2000). Conducting field methods in this manner permits computation of detection probability estimates for egg masses, as well as reduces observer biases (Grant et al. 2005). Studies surrounding other amphibian species, for instance, have concluded that double-observer methods may draw inferences about population changes by reducing the likelihood of missing or misidentifying egg masses (Grant et al. 2005). However, within the context of OSF studies conducted to date, such protocols have yet to be implemented. As a result of this slow procedural uptake, data sets are less comparable between sites and it is therefore more difficult to determine reliable, generalizable trends. Double-observer protocols should be established and followed hereafter during breeding season monitoring.

Also related to study design, this paper suggests that future surveying efforts follow strict schedules and protocols in order to equalize survey effort across years. This approach can ensure, for example, that no survey days are missed, and may also facilitate easier detection of first oviposition date. Researchers may then form more reliable conclusions regarding OSF behavioural and population changes and climate fluctuations from year to year. Future studies might also make data more easily accessible by promoting the use of electronic data sheets, as currently used by the COSFRT, that ensure all necessary information is recorded and can be shared amongst researchers.

Timing of egg mass surveying also constitutes an important factor moving forward. As environmental variables continue to change, it is possible that there will be a change in the breeding patterns of amphibians, including the OSF. The onset of egg mass surveying should occur across study sites on the same days in order to ensure that data outcomes are comparable. Due to the fact that the Morris Valley and Maria Slough wetlands are located within the same climate regime, the onset of breeding should theoretically occur around the same day for both populations if breeding is driven by environmental variables, as this study determined. The number of egg masses and site conditions (*e.g.*, temperature, precipitation and freshet levels) per study day also provide critical variables that should be recorded to monitor changes in productivity.

This study also noted that egg masses were sometimes altered or moved by researchers to prevent their desiccation. While this management approach poses beneficial outcomes for the population, such involvement interferes with researchers' abilities to identify underlying population stressors. Therefore, egg mass survivorship outcomes have been altered, and the data does not wholly represent natural trends. If intervention occurs in future studies, such actions must be diligently recorded in order to accrue accurate data. Even further, results might evaluate differences between egg masses that survive with versus without human intervention. Also to consider, OSF egg masses that are susceptible to desiccation or freezing could be considered for the husbandry program at the Greater Vancouver Zoo (Aldergrove, BC) and the Vancouver Aquarium (Vancouver, BC). Of course, protective measures and protocols must be established for such situations.

The amount of survey effort contributed towards conservation and research projects, such as the COSFRT, is often limited by funding. It is suggested that the implementation of a citizen science program may not only promote OSF conservation, but also would draw upon the valuable insights and capabilities of students and community members who are interested in recovery programs. Citizen science depends on volunteers to conduct ecological research and have proven especially useful for programs with limited resources (Dickinson et al. 2010). Moreover, by promoting public involvement, citizen science programs engage the community and create a connection between nature and humankind. Citizen science programs have been implemented for large-scale projects surrounding avian species, for instance, from which recommendations can be applied to future projects regarding the OSF. Such recommendations include coordination and communication between researchers and organizations, ensuring quality over quantity of data, utilizing repeated sampling techniques and establishing clear objectives and sampling design (Tulloch et al. 2013).

This paper acknowledges that, although citizen science programs do provide inexpensive data collection, their ability to uphold standardized methods and protocols are often questioned. Most often, citizen science programs do not seek to answer a particular hypothesis but rather involve surveillance monitoring with hopes that outcomes will prove useful (Dickinson et al. 2010). The argument of data quality also arises; however, this study supports the notion by Dickinson et al. (2010) that this issue is not unique to citizen science. As such, perhaps the implementation of a citizen science program could be implemented towards OSF research by offsetting costs associated with adhering to stricter protocols, such as the multiple-observer approach. An early study by Terhivuo (1988), which collected over 140 years of data through citizen science, determined that one particular frog species began breeding 2 to 13 days earlier over the course of 40 years. Evidently, such a significant result illustrates the potential usefulness of citizen science to help determine scientific outcomes. This paper thus maintains that successful citizen science programs offer an opportunity to strengthen evidence supporting the effects of climate change on the OSF.

Moving forward, a strong determining factor of success for the Morris Valley and Maria Slough populations involves ensuring that reproductive effort results in reproductive success. Therefore, further studies relating to survivorship of egg masses in the wild are vital to our understanding of the constraints that environmental variables incur for this species. As climate change will continue to play a considerable role in altering the timing of phenological activities, continual and reliable data collection will prove critical to understanding causes of OSF population changes.

Yet, despite the fact that climate change has been recognized as a major threat to amphibian biodiversity, there are few examples of studies and actions to address this issue (Shoo et al. 2011). It is possible that engineering solutions may provide solutions towards ameliorating some of the negative consequences of climate change on pond-breeding amphibians. Shoo et al. (2011) discuss useful interventions that can reduce exposures to temperature and moisture stressors that may be applicable for the OSF. For example, installation of microclimate and microhabitat refugees, enhancement and restoration of breeding sites and manipulation of water levels at breeding sites may provide notable benefits to extant OSF populations (Shoo et al. 2011). Of particular interest for this study is the application of restoring breeding habitat complexes, as this intervention could buffer amphibian populations from environmental stressors, provide heterogeneous aquatic breeding sites and may help to increase population size by alleviating reproductive failure (Shoo et al. 2011). This study upholds that any artificial interventions should be diligently monitored in conjunction with population studies in order to denote their actual effects on the OSF population, as well as to ensure that interventions are not in fact harming this species.

Furthermore, it is possible for management actions to manipulate hydroperiod water levels at breeding sites in order to increase the likelihood that developing tadpoles will survive to metamorphosis (Shoo et al. 2011). Potential interventions may include irrigation, filling drainage ditches and managing evapotranspiration through vegetation manipulation (Shoo et al. 2011). Similar management ideas have been implemented and discussed in an OSF management plan in Oregon, which future researchers may benefit from studying more closely (Cushman and Pearl 2007).

Based on the data analysis within this study, restoration should be considered an immediate option to improve breeding conditions for OSF populations. The risks associated with egg mass and tadpole stranding and desiccation is much greater than the risks associated with improving breeding ponds. The development of more and deeper breeding ponds may improve conditions during warmer, drier years when breeding occurs earlier in the year. In years when OSF face flash cold temperatures, or when dry conditions follow oviposition, conservation efforts should consider removal of these early egg masses to incorporate into head starting programs.

Above all, restoration decisions should aim to recover OSF populations by first determining the underlying cause(s) of OSF population declines. If changing climatic variables are not the primary cause, it must still be considered that climate change invokes stress on OSF populations from year to year, particularly during the breeding season. Moving forward, the results from this study imply that climate will continue to affect the onset of breeding, oviposition onset and larvae development. Therefore, future studies are warranted to monitor how the OSF populations will change in response to environmental variables over a longer period of time.

The recommendations of this study poise future studies to build study protocols towards ensuring reliable data outcomes. Survey methods should follow consistent standards overtime and between populations in order to evaluate data comparably, and a strong causal variable must be selected. In effect, this study concludes that definitive effective restoration decisions for the OSF will not be possible until scientific evidence is able to prove the determining factor(s) causing population decline. Future answers should aim to guide conservation management and restoration decisions to support, not only, population success of the OSF but also other at-risk pond-breeding amphibians around the world.

References

Backhouse, F., British Columbia, and L. Ministry of Environment and Parks. 2002. Oregon spotted frog. British Columbia Ministry of Environment, Lands and Parks. Victoria, BC

Beebee, T.J.C., and R.A. Griffiths. 2005. The amphibian decline crisis: A watershed for conservation biology? Biological Conservation 125:271–285.

Beebee, Trevor J.C. 1995. Amphibian Breeding and climate. Nature 219-220.

Benard, M.F. 2015. Warmer winters reduce frog fecundity and shift breeding phenology, which consequently alters larval development and metamorphic timing. Global Change Biology 21:1058–1065.

Blaustein, A.R., Belden, L.K., Olson, D.H., Green, D.M., Root, T.L., and Kiesecker, J.M. 2001. Amphibian Breeding and Climate Change. Conservation biology 15(6):1804-1809.

Blaustein, A.R., and J.M. Kiesecker. 2002. Complexity in conservation: lessons from the global decline of amphibian populations. Ecology letters 5:597–608.

Blaustein, A.R., Wake, David B., and Sousa, Wayne P. 1994. Amphibian Declines: Judging Stability, Persistence, and Susceptibility of Populations to Local and Global Extinctions. Conservation biology 8(1):60-71.

Blaustein, A.R., and D.B. Wake. 1990. Declining amphibian populations: a global phenomenon? 203-204.

Blaustein, A.R., and D.B. Wake. 1995. The puzzle of declining amphibian populations. Scientific American 272:52–57.

Blaustein, A.R., S.C. Walls, B.A. Bancroft, J.J. Lawler, C.L. Searle, and S.S. Gervasi. 2010. Direct and Indirect Effects of Climate Change on Amphibian Populations. Diversity 2:281–313.

Blood, D. A., and Anweiler, G. G. 1994. Status of the bald eagle in British Columbia. Wildlife Branch, Ministry of Environment, Lands, and Parks.

Buckley, J., and T.J.C. Beebee. 2004. Monitoring the conservation status of an endangered amphibian: the natterjack toad Bufo calamita in Britain. Animal Conservation 7:221–228.

Campbell Grant, E.H., R.E. Jung, J.D. Nichols, and J.E. Hines. 2005. Double-observer approach to estimating egg mass abundance of pool-breeding amphibians. Wetlands Ecology and Management 13:305–320.

Canadian Oregon Spotted Frog Recovery Team, British Columbia, and Ministry of Environment. 2012. Recovery strategy for the Oregon spotted frog (Rana pretiosa) in British Columbia. British Columbia, Ministry of Environment, Victoria

Carey, C., and M.A. Alexander. 2003. Climate change and amphibian declines: is there a link? Diversity and distributions 9:111–121.

Corn, P. S. 2005. Climate change and amphibians. Animal biodiversity and Conservation 28(1): 59-67.

COSEWIC. 2011. COSEWIC Assessment and Status Report on the Oregon Spotted Frog Rana pretiosa in Canada.

Cushman, K.A., and C.A. Pearl. 2007. A conservation assessment for the Oregon spotted frog (Rana pretiosa). USDA Forest Service and USDI Bureau of Land Management, Oregon

D'Amen, M., and P. Bombi. 2009. Global warming and biodiversity: Evidence of climate-linked amphibian declines in Italy. Biological Conservation 142:3060–3067.

Dickinson, J. L., Zuckerberg, B., and Bonter, D. N. 2010. Citizen science as an ecological research tool: challenges and benefits. Annual review of ecology, evolution, and systematics 41: 149-172.

Donnelly, Maureen A., and Crump, Martha L. 1998. Potential Effects of Climate Change on Two Neotropical Amphibian Assemblages. Potential impacts of climate change on tropical forest ecosystems 401-421.

Environment Canada. 2014. Recovery Strategy for the Oregon Spotted Frog (Rana pretiosa) in Canada [Proposed]. Species at Risk Act Recovery Strategy Series. Environment Canada, Ottawa.

Faccio, S.D. 2011. Using Egg Mass Surveys to Monitor Productivity and Estimate Population Sizes of Three Pool-breeding Amphibians at Marsh-Billings-Rockefeller National Historical Park.

Green, D.M. 2003. The ecology of extinction: population fluctuation and decline in amphibians. Biological conservation 111:331–343

Houlahan, Jeff. E., Findlay, C. Scott, Schmidt, Benedikt R., Meyers, Andrea H., and Kuzmin, Sergius L. 2000. Quantitative Evidence for global amphibian population declines. Nature 404:748

Kiesecker, J.M, Blaustein, A.R., and Belden, L.K. 2001. Complex causes of amphibian population declines. Nature 410:681-684.

Kissel, A.M., W.J. Palen, and P. Govindarajulu. 2017. A Decision-theory Approach to Cost-effective Population Supplementation for Imperiled Species. Ecological Economics 142:194–202

Lettink, M. 2012. Herpetofauna: population estimates (using capture-mark-recapture data) v1. 0. Inventory and monitoring toolbox: herpetofauna. Department of Conservation Te Papa Atawhai.

Licht, Lawrence E. 1969. Comparative breeding behavior of the red-legged frog (rana aurora aurora) and the western spotted frog (rana pretiosa pretiosa) in southwestern British Columbia. Canadian Journal of Zoology 47(6):1287-1299.

Licht, L. E. 1971. Breeding habits and embryonic thermal requirements of the frogs, Rana aurora aurora and Rana pretiosa pretiosa, in the Pacific Northwest. Ecology 52(1): 116-124.

Licht, L.E. 1974. Survival of embryos, tadpoles, and adults of the frogs Rana aurora aurora and Rana pretiosa pretiosa sympatric in southwestern British Columbia. Canadian Journal of Zoology 52:613–627

McAllister, K.R., and Leonard, W.P. 1997. Washing State status report for the Oregon spotted frog. Washington Department of Fish and Wildlife, Wildlife Management Program.

Nichols, J. D., Hines, J. E., Sauer, J. R., Fallon, F. W., Fallon, J. E., and Heglund, P. J. 2000. A double-observer approach for estimating detection probability and abundance from point counts. The Auk, *117*(2): 393-408.

Noon, Barry R., and Kevin S. McKelvey. "The process of indicator selection." In: *Aguirre-Bravo, C.; Pellicane, Patrick J.; Burns, Denver P.; Draggan, Sidney, eds. 2006. Monitoring Science and Technology Symposium: Unifying Knowledge for Sustainability in the Western Hemisphere. Proceedings RMRS-P-42CD. Fort Collins, CO: US Department of Agriculture, Forest Service, Rocky Mountain Research Station* 42:944-951.

McKibbin, R., W.T. Dushenko, G. vanAggelen, and C.A. Bishop. 2008. The influence of water quality on the embryonic survivorship of the Oregon spotted frog (Rana pretiosa) in British Columbia, Canada. Science of The Total Environment 395:28–40.

Pearl, Christopher A., and Hayes, Marc P. 2004. Habitat associations of the Oregon spotted frog (rana pretiosa): a literature review.

Pearson, Monica. 2009. Inventory and long-term monitoring Oregon spotted frog and red-legged frog egg masses in the Fraser Valley Lowlands. Report.

Pearson, Monica. 2011. Project Report: Chaplin Road Wetlands Oregon Spotted Frog Habitat Expansion 2010. Report.

Pechmann, J.H., D.E. Scott, J.W. Gibbons, and R.D. Semlitsch. 1989. Influence of wetland hydroperiod on diversity and abundance of metamorphosing juvenile amphibians. Wetlands Ecology and Management 1:3–11

Petit, E., and N. Valiere. 2006. Contributed Papers: Estimating Population Size with Noninvasive Capture-Mark-Recapture Data: Noninvasive Capture-Mark-Recapture Data. Conservation Biology 20:1062–1073.

Pradel, R. 1996. Utilization of Capture-Mark-Recapture for the Study of Recruitment and Population Growth Rate. Biometrics 52:703.

Raithel, C.J., P.W. Paton, P.S. Pooler, and F.C. Golet. 2011. Assessing long-term population trends of wood frogs using egg-mass counts. Journal of Herpetology 45:23–27.

Reading, C.J. 1998. The effect of winter temperatures on the timing of breeding activity in the common toad Bufo bufo. Oecologia 117:469–475.

Reading, C.J. 2007. Linking global warming to amphibian declines through its effects on female body condition and survivorship. Oecologia 151:125–131.

Richardson, John. 2011. Movement, Habitat Use, and Dispersal of Juvenile Oregon Spotted Frogs (Rana pretiosa) on Joint Base Lewis McChord. Doctoral dissertation, Evergreen State College

Salvidio, S. 2009. Detecting amphibian population cycles: The importance of appropriate statistical analyses. Biological Conservation 142:455–461.

SARA A to Z Species Index http://www.registrelep-sararegistry.gc.ca/ Accessed January 27, 2018

Scherer, R.D., and J.A. Tracey. 2011. A power analysis for the use of counts of egg masses to monitor wood frog (Lithobates sylvaticus) populations. Herpetological Conservation and Biology 6:81–90.

Semlitsch, R.D. 2002. Critical elements for biologically based recovery plans of aquatic-breeding amphibians. Conservation biology 16:619–629.

Shoo, L. P., Olson, D. H., McMenamin, S. K., Murray, K. A., Van Sluys, M., Donnelly, M. A., …and Bishop, P. J. 2011. Engineering a future for amphibians under climate change. Journal of Applied Ecology, *48*(2): 487-492.

Stinson, D. W., Watson, J. W., & McAllister, K. R. (2001). Washington State status report for the bald eagle. Washington Department of Fish and Wildlife, Wildlife Program.

Stuart, Simon N., Chanson, Janice S., Cox, Neil A., Young, Bruce E., Rodrigues, Ana S. L., Fischman, Debra L., and Waller, Robert W. 2004. Status and Trends of Amphibian Declines and Extinctions Worldwide. Science 306(5702):1783-1786.

Terhivuo, J. 1988. Phenology of spawning for the common frog (Rana temporaria L.) in Finland from 1846 to 1986. In *Annales Zoologici Fennici.* Finnish Academy of Sciences, Societas Scientiarum Fennica, Societas pro Fauna et Flora Fennica and Societas Biologica Fennica Vanamo 165-175

Todd, B.D., Scott, D.E., Pechmann, J.H., and Gibbons, J.W. 2010. Climate change correlates with rapid delays and advancements in reproductive timing in an amphibian community. Proceedings of the Royal Society of London B: Biological Sciences.

Tulloch, A. I., Possingham, H. P., Joseph, L. N., Szabo, J., and Martin, T. G. 2013. Realising the full potential of citizen science monitoring programs. Biological Conservation 165:128-138.

Appendix A.

Figure A.1. **Map of the two study locations, Morris Valley and Maria Slough wetland, and surrounding area. (Map: Caroline Feischl; Google Earth)**

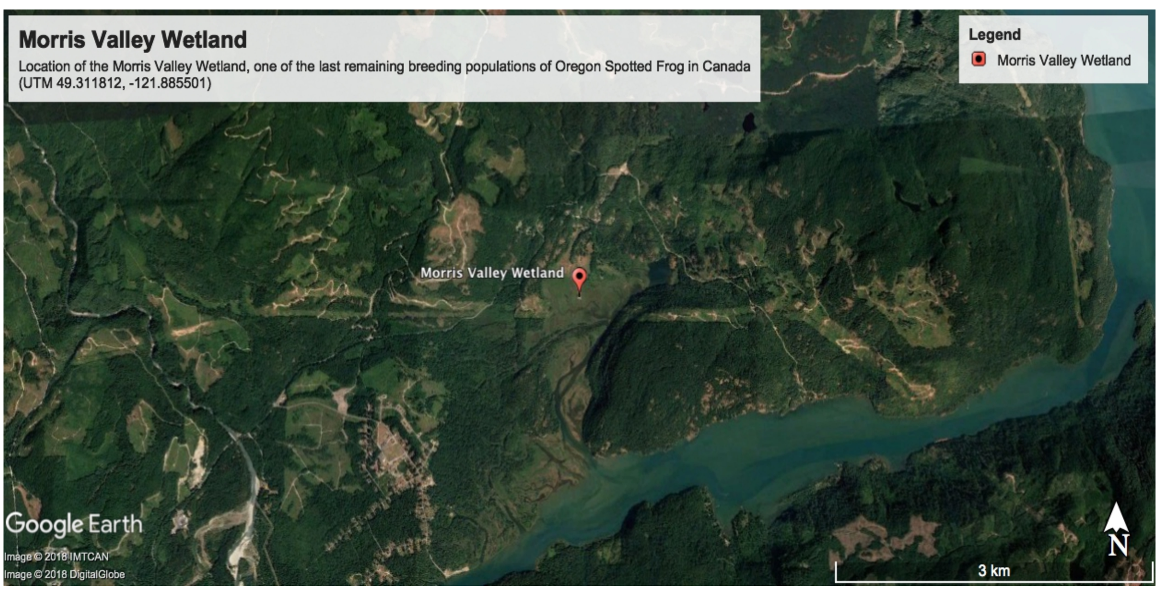

Figure A.2. **Map of Morris Valley location. (Map: Caroline Feischl; Google Earth)**

Figure A.3. **Map of Maria Slough location. (Map: Caroline Feischl; Google Earth)**

Figure A.4. **Photo of young Oregon Spotted Frog partially submerged in the water. Taken at the Morris Valley Wetland, 2017. (Photo: C. Feischl)**

Egg Mass Survey Protocols for Oregon spotted frogs and Red-legged frogs 2011

This document anticipates multiple site visits by potentially different observers, and will ensure that all observers are following the same protocols. Systematic surveys will be completed by multiple observers at multiple locations.

Materials:

- Polarized sunglasses
- GPS unit, set to NAD83/UTM Zone 10
- Clipboard, data forms, pencil
- Digital camera (polarized lens if you have one)
- Flagging tape, if planning to return to the site (use yellow if in locations with public access)
- Sharpie marker
- Thermometer
- Conductivity/pH meter
- Depth-measuring device (folding extensible ruler, or a heavy bob on a string)
- Waders
- Canoe / kayak, if necessary to move effectively in the habitat.

GPS settings:

Ensure that your GPS is set to collect data in NAD83/UTM Zone 10, and to track your movements. Turn the GPS on when you begin your search, and mark the time in your notes. This will allow us to track effort. When using your GPS for other activities during the breeding window, please ensure that other tracks are not recorded in the same file as your OSF search tracks.

Waypoints should be 'marked' in your GPS, and the waypoint number transcribed onto your notes along with the UTM coordinates. **Please write the UTM coordinates AND waypoint # in your notes.**

When all surveys are completed, you will need provide your GPS files to the data processor in .xls and .gpx format, along with your field notes (either digitized into the excel data form, or scanned / original forms).

Single-Visit Protocols:

At locations not known to have OSF on-site, single visits will be made to count RLF egg masses and search for OSF egg masses. These visits will begin once OSF egg masses have been seen in all known locations, ensuring that breeding season has begun. Depending on density, RLF egg mass locations may be grouped into logical landscape units (i.e. # of egg masses in a pond or section of stream). If using this method, ensure that the marked GPS location is central within the landscape unit.

Please fill in each field on the spreadsheet for every waypoint, according to your available tools. Temperature, conductivity and depth should be collected for every waypoint. Please make a note of pictures taken, and submit them with your data. Photographs of overall habitat conditions are also very useful.

If Oregon spotted frog egg masses are suspected, collect all data (water temperature, conductivity, Gosner stage, est % dead eggs), flag the location of the egg mass and alert Monica Pearson immediately to arrange for confirmation.

Multiple-Visit Protocols

At locations known to have OSF on-site, multiple site visits will be made to ensure that all egg masses are counted. The following instructions are intended to help observers track changes more easily.

When OSF egg masses are identified, ensure that each **cluster** has a unique waypoint.

- In your notes:
  - Time
  - Waypoint #
  - UTM coordinates
  - Species
  - Water temperature
  - Conductivity
  - pH
  - Dissolved oxygen (if possible)
  - Gosner stage
  - Estimate % dead eggs
  - Photo number and drawing of the cluster showing individual egg masses.
  - Comments
  - Access details if necessary
- In your GPS waypoint file:
  - note in the waypoint file (in the 'notes' section) how many egg masses are in each cluster.
- In the field:
  - mark each cluster in the field with flagging tape as close as possible to the cluster
  - write your initials, the waypoint #, date, and # egg masses on the flagging tape.
  - install a second flag at a visible location (in yellow if in a sensitive area)
  - When returning to an egg mass, take the same information in your notes, and add # egg masses / cluster to your GPS file. Compare new data to flagging tape. Add flagging with updated information (leave existing flagging where it is.) If you know that no-one will return to the site, remove flagging and submit it with your notes to Monica Pearson.

Figure A.5. Egg mass survey protocol as suggested in annual COSFRT annual reports.

Appendix B.

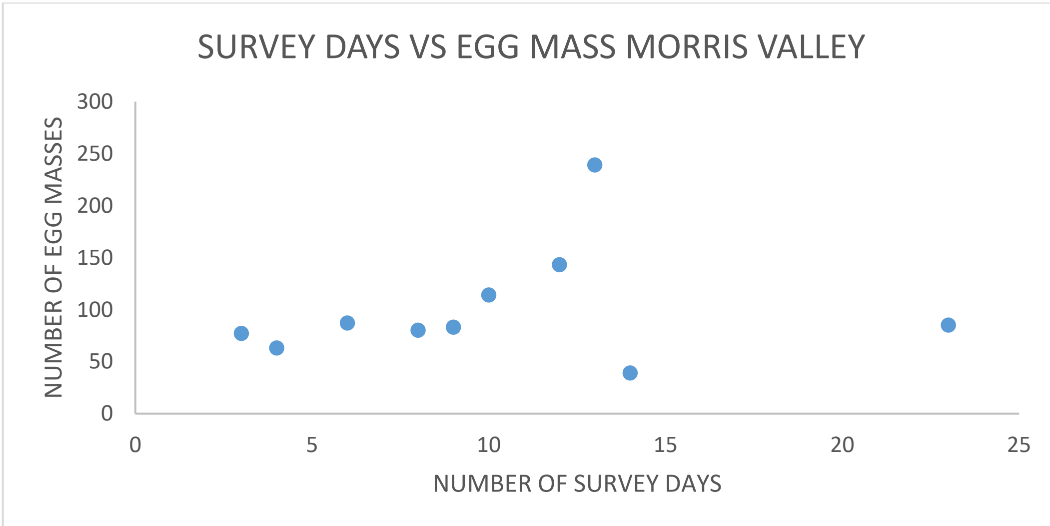

Figure B.1. **Number of survey days compared to number of egg masses at Morris Valley. Results include outlier years 2010 and 2015, suggest a weak relationship and demonstrate no significance (R2=0.03603; p>0.05).**

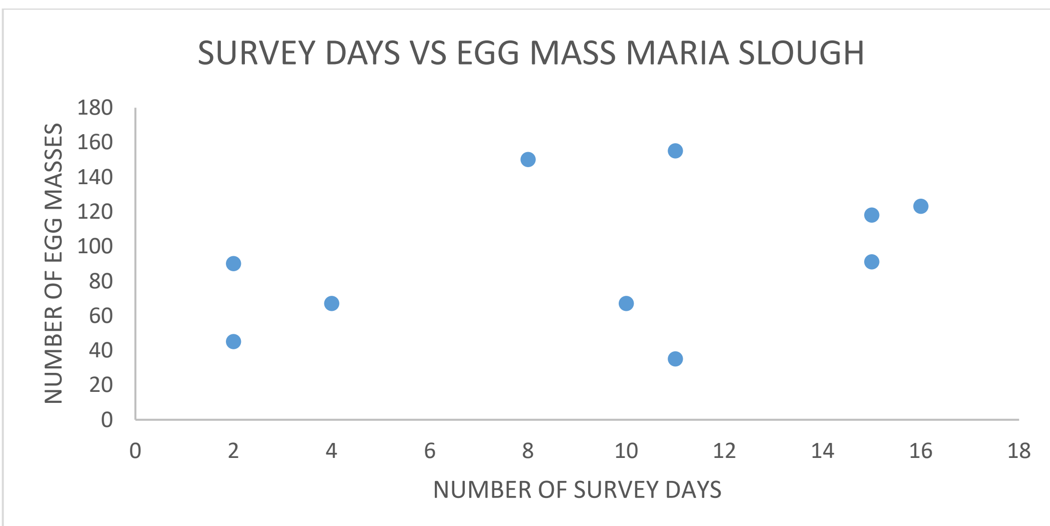

Figure B.2. **Number of survey days related to egg masses at Maria Slough. Results suggest a weak relationship and demonstrate no significance (R2=0.14464; p>0.05).**

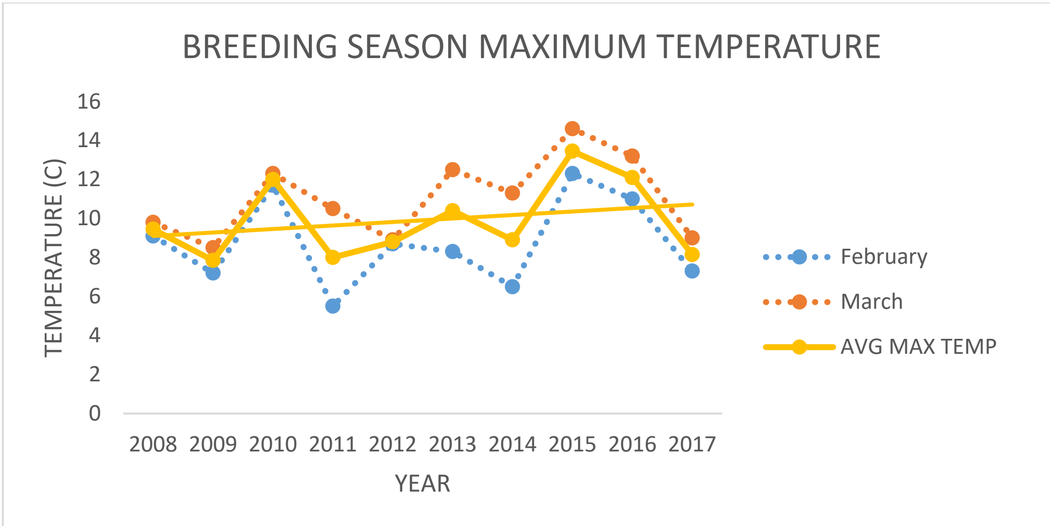

Figure B.3. **Average maximum temperatures in Maria Slough and Morris Valley wetlands for February through March for 2008-2017, inclusive. Results show a slight increase in temperatures overtime throughout the breeding season of OSF as observed by the trendlines.**

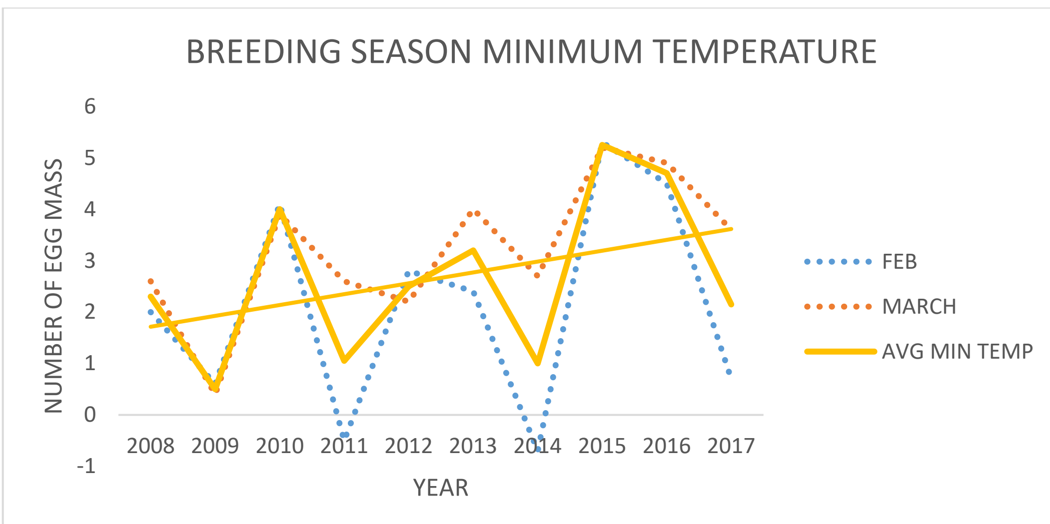

Figure B.4. **Average minimum temperatures in Maria Slough and Morris Valley wetlands for February through March for 2008-2017, inclusive. Results show a slight increase in temperatures overtime throughout the breeding season of OSF as observed by the trendlines.**

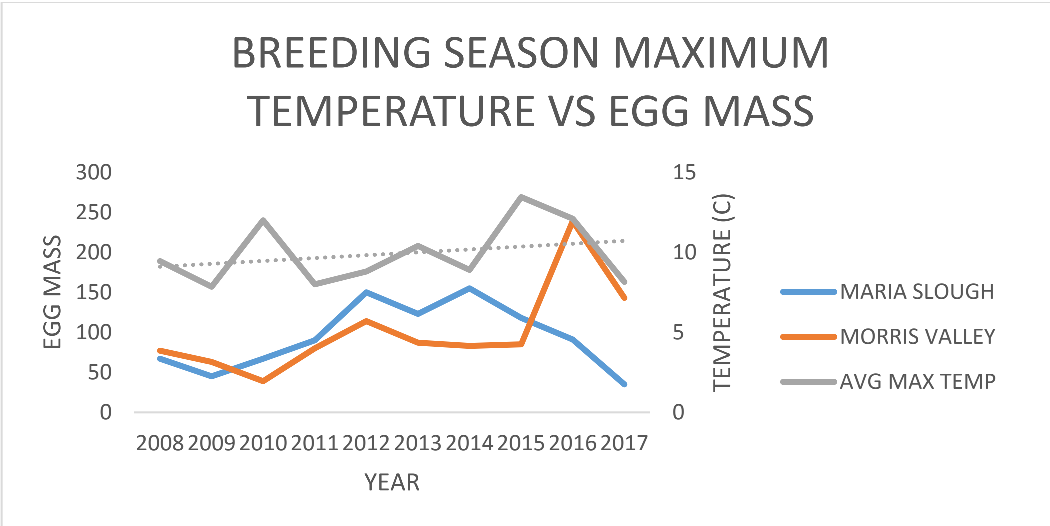

Figure B.5. **Maximum average breeding season (February, March) temperatures for Morris Valley and Maria Slough from 2008-2017, inclusive, as related to number of egg masses produced at these sites. There is no significance between temperatures and egg masses at either study site (p>0.05).**

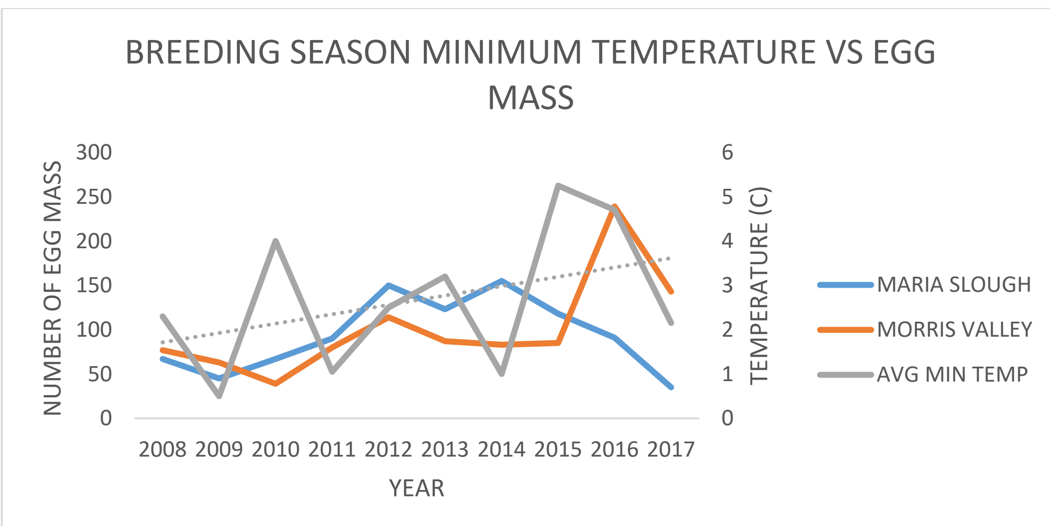

Figure B.6. **Minimum average breeding season (February, March) temperatures for Morris Valley and Maria Slough from 2008-2017, inclusive, as related to number of egg masses produced at these sites. There is no significance between temperatures and egg masses at eith**

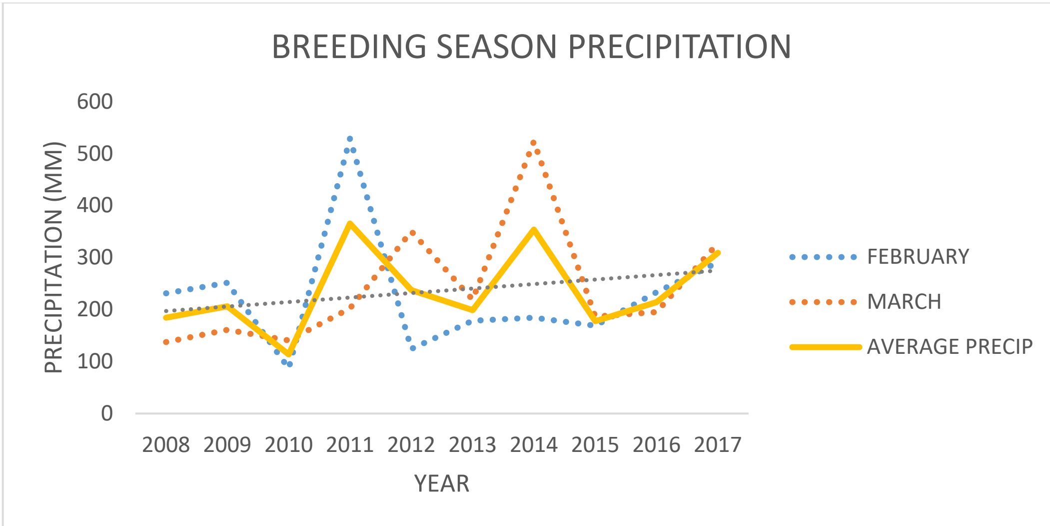

Figure B.7. **Average precipitation (mm) during breeding season (February, March) of OSF for 2008-2017, inclusive. The trend line indicates a slight increase in average breeding season precipitation levels over the study period.**

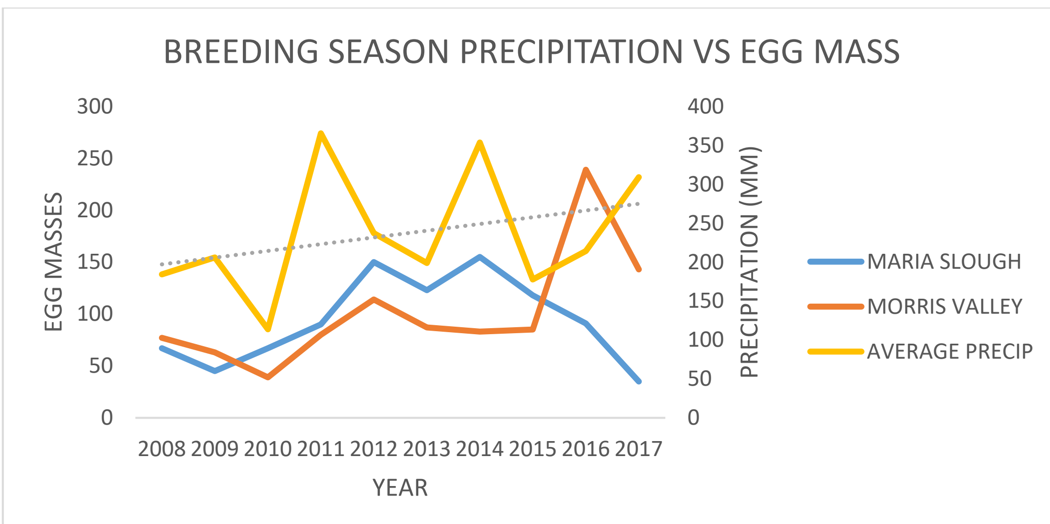

Figure B.8. **Average monthly precipitation (mm) during OSF breeding seasons (February, March) from 2008-2017, inclusive, as related to number of egg masses produced at Morris Valley and Maria Slough. No significant relationship was determined between precipitation levels and quantity of egg masses at either site ($p>0.05$).**