Temporal Calling Patterns of Seven Anuran Species in Southern Ontario

by

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ABSTRACT

TEMPORAL CALLING PATTERNS OF SEVEN ANURAN SPECIES IN SOUTHERN ONTARIO

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Globally, anurans have experienced significant declines and reliable monitoring protocols are required to track population trends. This thesis compares the effectiveness of manual call surveys (MCS) and automated recording systems (ARS) and describes the calling profiles of seven anuran species in southern Ontario.

Using detailed audio recordings, we identify the seasonal and hourly calling patterns of seven anuran species at Warwick and Silver Creek Conservation Areas in the Credit River watershed. We employ descriptive and graphical methods to establish calling profiles. Given the low detection rate of American toads, grey treefrogs and northern leopard frogs, a survey protocol comprised of 9 weekly site visits are recommended in May and June. The optimal time of day for detecting the greatest abundance and species richness is 23h30 whereas optimal survey duration varies with hour and season given the target species. Climatic variables may induce or inhibit call activity and until the extent of these variables can be established, protocol guidelines should implement longer and more frequent site visits. The urgency of this revision is exacerbated for commercial surveys used to make contentious land management decisions.
Acknowledgments

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I would also like recognize to CVC staff for granting us access to Warwick and Silver Creek Conservation Areas as well as the time they have contributed to numerous meetings throughout the duration of this study. More specifically I would like to thank Kata Bavrlic and Kirk Bowers for their statistical insight.
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Chapter 1

1.0 Introduction to the Amphibian Crisis

Reported amphibian declines and extinctions have led to many recent amphibian studies (Heyer et al. 1994, Houlnahan et al. 2000, Wake and Vredenburg 2008). Currently over 70% of amphibian populations are in decline and one third of the world’s amphibian species are endangered due to habitat modification, over-exploitation, climate change (Stuart et al. 2004) and pathogens (Hayes 2010). Amphibians have existed for 220 million years (Jensen et al. 2008) and their populations have been historically resilient (Stuart et al. 2005). Some amphibians alive today are among the longest lived species on Earth, with records dating back to the Miocene (5.3 million years ago) or Pliocene (1.8 million years ago) epochs (Delfino 2005). The extinction rate of amphibians was far less than the extinction rate of birds and mammals with only 34 extinctions documented historically since the 1500’s, thus amphibian populations were presumed to be relatively stable (Stuart et al. 2004). Present-day trends have changed dramatically; 122 amphibian extinctions are believed to have occurred since the 1980’s, far exceeding that of birds (5 extinctions) and mammals (no extinctions) (Stuart et al. 2004). This relatively stable taxon has become the most threatened group of organisms on the planet (Wake and Vredenburg 2008).

Anurans (frogs and toads) have a complex lifecycle, in North America, requiring a variety of connected habitats (Marsh and Trenham 2001) causing them to migrate hundreds of metres from the wetland boundary (Semlitsch and Bodie 2003). These species require extensive terrestrial habitat for foraging, refugia, hibernation and migration (Gibbons 2003, Guerry and
Hunter 2002). With rapidly increasing global urbanization rates (Seto et al. 2012), their need for extensive habitat is making them highly susceptible to the effects of habitat loss because they are slow moving, small bodied animals with poor dispersal abilities in urban environments (Gibbs 1998). Urban landscapes produce many impassable barriers resulting in habitat fragmentation (Gibbs 1998, Trenham et al. 2003), moreover anurans have permeable skin that makes them particularly sensitive to environmental contaminants found therein (Quaranta et al. 2009). Furthermore, larvae are susceptible to environmental variables caused by early drying of ponds that can negatively affect food supply, tadpole density, size at metamorphosis and postmetamorphic effects (Carey and Alexander 2003). Given the nature of these sensitivities and requirements during larval and adult stages, conservation challenges are compounded (Hayes 2010).

Much effort is required to ensure that these animals are able to persist and thrive in conjunction with rapid urban development (Pillsbury and Miller 2008) and the Environmental Commissioner of Ontario recommends that amphibian biodiversity should be given priority protection in Ontario as “These declines and extinctions appear to represent a unique amphibian crisis ... the worst we have ever faced” (Miller 2009, p.44). To mitigate declines in Ontario, environmental consulting firms are hired to evaluate anuran populations prior to development using standardized monitoring practices (PPS 2005).

1.1 Monitoring Anuran Populations
An increased awareness of anuran population decline has prompted the establishment of many monitoring programs across North America (Heyer et al. 1994). The Amphibian Road Call
Counts, the Backyard Frog Survey, the Marsh Monitoring Program (Konze and McLaren 1997) and the North American Amphibian Monitoring Program (USGS 2012) are a few examples of programs that utilize auditory data to assess population structure. These surveys are based on the premise that male anurans emit species-specific calls during breeding season that can be used to determine site occupancy (Weir and Mossman 2005). Observers conduct a series of manual call surveys (MCS) consisting of three to four site visits per season. These surveys begin no earlier than half an hour after sunset and must be completed by midnight (Konze and McLaren 1997) or 1 am (NAAMP n.a.). In recent years, skepticism regarding the effectiveness of conventional protocols has surfaced (Brander et al. 2007, Bridges and Dorcas 2000, de Solla et al. 2005, Shirose et al. 1997). As an alternative, some researchers have utilized automated recording systems (ARS) to evaluate anuran populations using a consistent recording schedule conducted over a 24 period (Peterson and Dorcas 1994). These systems can record calls at a fine temporal scale to identify patterns of calling activity with little disruption to calling anurans (Mohr and Dorcas 1999, Bridges and Dorcas 2000).

The development of an effective monitoring strategy is the first step towards successful anuran conservation (Heyer et al. 1994); consequently protocols should be based on a clear understanding of anuran calling strategies. Detection of species richness at a site may vary annually as a result of metapopulation dynamics or due to temporal variability of calling anurans (de Solla et al. 2006). Inaccurate detection of species richness at a discrete location is acceptable as site occupancy is likely to be confirmed if surveys are conducted consistently on an annual basis (Shirose et al. 1997). In the case where short-term surveys seek to produce an accurate
account of species richness, frequency and survey duration must be designed to maximize species detectability, especially when these studies influence land management decisions.

1.2 Study Design

This thesis compares the effectiveness of widely used manual call surveys (MCS) to data collected from automated recording systems (ARS) to test the hypothesis that current protocol guidelines for MCS are under-representing annual species richness. If the hypothesis is correct, data will be used to identify the temporal calling patterns of anuran species and provide critical knowledge for the development of improved MCS guidelines. Temporal patterns can be formulated by creating species-specific profiles from detailed audio recordings conducted during the breeding season. Relating these profiles to survey guidelines could significantly improve species detectability as well as our knowledge and ability to preserve populations. The need to establish the distribution of population presence and the location of population declines in the presence of development pressures highlights the significance and immediacy of this study.

Objectives:

1. Assess the effectiveness of MCS in producing an accurate account of species presence.
2. Evaluate the ability of ARS techniques in detecting anuran calls.
3. Identify seasonal and hourly trends to create species-specific calling profiles.
4. Identify optimal survey duration.
5. Integrate species trends to identify interspecific seasonal and hourly variances to formulate biologically representative monitoring recommendations.
An in-depth approach was taken to compare MCS and ARS methods and create calling profiles for species occurring within the Credit River watershed. Detailed recordings were collected from two wetlands at Warwick and Silver Creek Conservation Areas which have been monitored for 10 years utilizing conventional MCS methods. These data provided the necessary information required to evaluate the effectiveness of both survey methods. In addition, intensive recordings provided the detail required to construct species-specific profiles. These data were analysed using a series of descriptive methods and results were represented graphically depicting various temporal aspects of species calling patterns. Species profiles were assembled by identifying daily fluctuations within the breeding season. Hourly trends were identified by periods of peak frequency and intensity of call activity and statistically verified using chi-square tests. Patterns were synthesized to establish interspecific variances and similarities in anuran calling activity while the number of recommended site visits was estimated using a rudimentary calculation based on species-specific detection rates. Associations between hour, month and survey duration were identified to produce relevant protocol recommendations that incorporate dynamic anuran calling patterns.

The chapters that follow emphasize the need for an improved monitoring protocol. Chapter 2 explores the ecology of anuran species within the Credit River watershed and expands upon the auditory methods utilized to quantify these populations. Chapter 3 provides a detailed description of the auditory methods used to collect call data as well as the descriptive methods employed to analyse these data. Chapter 4 summarizes the results of these analyses and presents the calling profiles of each species identified during field recordings. Concluding chapters integrate seasonal and hourly interspecific similarities to produce recommendations that reflect
calling patterns of seven anuran species and explain how these methods and findings can be implemented into management practices.
Chapter 2

2.0 Background

Amphibians have a complex life cycle requiring both aquatic and terrestrial habitats. The structure and composition of their populations are dependent upon the availability and configuration of local habitat. Some species exist primarily in terrestrial habitats and frequent wetlands during the breeding season while others reside solely in permanent ponds. Despite specific habitat requirements, all anurans gather in wetlands during species-specific breeding seasons where they emit distinctive calls that can be used to identify species and abundance. Monitoring programs have developed protocol guidelines to maximize species detection by exploiting these seasonal congregations. These protocols are utilized across North America and monitor a wide range of wetland habitats to produce databases used to track population trends.

In recent years, these protocols have come under scrutiny and the effectiveness of their guidelines has been challenged as timing, frequency and duration of surveys are believed to under-represent species richness and abundance (de Solla et al. 2006). Protocols should be revised to maximize species detectability and observer effort by establishing species-specific calling profiles to detect all species during their peak calling period. An automated recording system (ARS) can be used to collect detailed auditory data required to define anuran calling strategies. Increasing survey accuracy will improve our knowledge of species distribution and habitat use so that informed decision makers can effectively manage public and private lands.
2.1 Monitoring Anuran Populations

Frogs and toads comprise a diverse group of amphibians classified under the order Anura. Anurans are the most numerous order within the amphibian class and can be found in a wide range of habitats from tropical forests to arid deserts (Hulse et al. 2001). Ontario is home to 13 of these species (MacCulloch 2002); however, due to rapid development and increasingly urbanized habitats, their populations are in peril (Miller 2009). In response to these concerns, numerous agencies have begun implementing anuran monitoring programs to track population trends (de Solla et al. 2005).

Credit Valley Conservation (CVC) has addressed this issue by commissioning Dr. Lorne Bennett, University of Guelph, and Dr. Robert Milne, Wilfrid Laurier University, to monitor anuran populations throughout the watershed. The CVC is concerned with documenting patterns of species richness, site occupancy and abundance as well as species response to landscape and vegetation parameters (CVC 2009). Monitoring began in 2003 and continues today using a revised form of the Marsh Monitoring Protocol (MMP). In addition to the MCS, an in-depth study began in the summer of 2011 focusing on temporal calling patterns of anuran species using an automated recording system.

2.2 Credit River Watershed

Temporal data for this study were collected in the Credit River watershed. The watershed is approximately 980 km², 6% of which is classified as wetlands and 12% as forested land cover (CVC 2007). As it is located in the Greater Toronto Area (GTA), the watershed contains large areas of urban development including the cities of Brampton, Mississauga and Oakville. Urban
land use covers 31% of the watershed while agricultural activities occupy 34% of the total area (CVC 2007). The headwaters of the Credit River begin in the town of Mono and extend 100 km south to Lake Ontario (Fig. 2.1).

Figure 2.1. Credit River watershed.
2.3 Description of Anuran Species

Adult anurans are characterized by their protruding eyes, compact tailless bodies and large hind legs (Jensen et al. 2008). Though there are no taxonomic differences between frogs and toads, frogs typically have smooth skin while toads are defined by their warty skin (Jensen et al. 2008). Anurans of Ontario reproduce by laying eggs in aquatic environments (MacCulloch 2002). Larvae (tadpoles) hatch and undergo a transformation called metamorphosis (Mazerolle et al. 2005). When metamorphosis is complete, froglets emerge from their aquatic environment (Rothermel 2004).

2.3.1 Anurans of the Credit River watershed

In the province of Ontario, there are 13 anuran species (MacCulloch 2002); seven of which were identified in the Credit River watershed during the MCS conducted in the spring of 2011 at Silver Creek and Warwick Conservation Areas (Table 2.1).

Table 2.1. Anurans of the Credit River watershed.

<table>
<thead>
<tr>
<th>Image</th>
<th>Species name</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image.png" alt="American Bullfrog" /></td>
<td>American Bullfrog <em>Rana catesbeiana</em></td>
</tr>
<tr>
<td><img src="image.png" alt="American Toad" /></td>
<td>American Toad <em>Bufo americanus</em></td>
</tr>
<tr>
<td><img src="image.png" alt="Grey Treefrog" /></td>
<td>Grey Treefrog <em>Hyla versicolor</em></td>
</tr>
<tr>
<td><img src="image.png" alt="Green Frog" /></td>
<td>Green Frog <em>Rana clamitans</em></td>
</tr>
<tr>
<td><img src="image.png" alt="Northern Leopard Frog" /></td>
<td>Northern Leopard Frog <em>Rana pipiens</em></td>
</tr>
<tr>
<td><img src="image.png" alt="Pickerel Frog" /></td>
<td>Pickerel Frog <em>Rana palustris</em></td>
</tr>
<tr>
<td><img src="image.png" alt="Spring Peeper" /></td>
<td>Spring Peeper <em>Pseudacris crucifer</em></td>
</tr>
</tbody>
</table>

(Pictures from MacCulloch 2002)
2.3.1.1 Bufonidae

Bufonidae are true toads (Hulse et al. 2001). This family contains nearly 300 species globally and two species occur in Ontario, namely the Fowler’s toad and American toad (MacCulloch 2002). The American toad is of the genus *Bufo* and is found in the Credit River watershed. This genus has short, stocky legs with a neutral coloration and thick warty epidermis for protection against water loss (Hulse et al. 2001, MacCulloch 2002).

American Toad (*Bufo americanus*)

American toads are brown in colour with several dark patches on their back containing one to two warts and colour can vary depending on temperature and light intensity (Hulse et al. 2001). Toads are aquatic breeders and larvae can develop in both temporary and permanent bodies of water (Houlahan and Findlay 2003). Adults are terrestrial species that inhabit a variety of habitats from urban gardens to rural forests (Jensen et al. 2008).

Males begin calling as early as April and continue calling into late June. Toadlets hatch in 4 to 10 days following oviposition (Petranka et al. 1994). The advertisement call of an American toad is a long, piercing, high frequency trill that can last up to 30 seconds and sounds like the high pitch squeal of distant traffic. The release call of a male is a series of squeaky chirps (Elliott 2004).
2.3.1.2 Hylidae

Treefrogs are classified in the family Hylidae. There are 740 confirmed species (Hulse et al. 2001), five of which occur in Ontario (MacCulloch 2002) and two species, namely the grey treefrog and spring peeper, occur in the Credit River watershed. Treefrogs are naturally adapted to arboreal environments with pads on their fingers and toes that allow them to adhere to surfaces (Hulse et al. 2001).

Grey Treefrog (*Hyla veriscolor*)

The grey treefrog is of the genus *Hyla* which contains over 260 species. As their name indicates, treefrogs of Ontario are predominantly grey in colour; however, they can also exhibit various shades of green. The underside of their hind legs is bright orange or yellow in colour to distract predators as they hop away (Hulse et al. 2001). This arboreal species is typically found in deciduous forests. Frogs emerge from their terrestrial hibernacula in May (Hulse et al. 2001) and begin migrating to breeding ponds. Treefrogs breed in temporary ponds as they have no larval predatory defenses (Heenar and McCloskey 1998) and oviposit batches of 20-90 eggs which hatch within 4-5 days of egg mass deposition (Johnson and Semlitsch 2003).

Grey treefrogs begin calling in May and continue throughout the month of June. The advertisement call is a short melodic trill that is repeated every few seconds. The treefrog also emits an antagonistic squeaky chirp when approached by another male (Elliott 2004).
Spring Peeper (*Pseudacris crucifer*)

Spring peepers are also classified as treefrogs and organized under the genus *Pseudacris*; however, they are ground-dwelling frogs that are rarely found in arboreal environments. Members of the genus *Pseudacris* are slender species with small discs on their fingers and toes (Hulse et al. 2001). Spring peepers are tan in colour and have a characteristic dorsal X-shaped marking which differentiate them from juvenile grey treefrogs (MacCulloch 2002). They have a preference for deciduous woodlands (Hehnar and McCloskey 1998). They emerge from their terrestrial hibernacula as early as March and congregate in ponds a short time later to begin breeding (Houlahan and Findlay 2003). Froglets hatch within 6 to 12 days following oviposition (University of Acadia, n.d.) and can complete their metamorphosis as early as June in ponds with little canopy cover (Halverson et al. 2003).

Frogs begin calling in April but may start earlier in response to climatic conditions. The spring peeper has several different calls including an advertisement call which is a series of short peeps: *peep-peep-peep*, an aggressive call which is a stuttering trill: *purrreeek* and a rain call which is a chirp or harsh squeak (Elliott 2004).

2.3.1.3 Ranidae

The family of true frogs, Ranidae, contains over 700 species with 270 classified as of the genus *Rana* (Hulse et al. 2001). These frogs have long limbs which allow them to exhibit a superior jumping ability and slender webbed-toes making them proficient swimmers (Hulse et al. 2001). There are six species of the *Rana* genus in Ontario (MacCulloch 2002) and four species
including, American bullfrog, green frog, northern leopard frog and pickerel frog, were found in the Credit River watershed during auditory surveys.

American Bullfrog (*Rana catesbeiana*)

Bullfrogs are green in color and occasionally have a mottled pattern on their back (MacCulloch 2002). They are smooth with no dorsal folds and have a large tympanum. Bullfrogs are aquatic species which forage, breed and overwinter in large permanent ponds (Pillsbury and Miller 2008). Following oviposition, froglets hatch within 4-5 days (University of Acadia, n.d.). They call throughout the month of June and into July and may begin calling as early as May if conditions are favorable. Bullfrogs emit a deep, basal call that is commonly described as *jug-o-rum*.

Green Frog (*Rana clamitans*)

Green frogs vary in colour from green to brown and often have small black spots on their back. Frogs are aquatic and are positively correlated with the presence of forested landscape (Houlahan and Findlay 2003). They breed and overwinter in permanent ponds. They develop quickly and hatch within 4 days of egg mass deposition (University of Acadia, n.d.).

Males begin calling in May but their peak calling period occurs in June and can persist into July. The green frog has 6 call types. The primary mating call is similar to a plucked banjo string: *Goonk-gunk-gunk* (Elliott 2004). The second is guttural stuttering call used during encounters with other frogs: *r-u-u-u-ng… r-u-u-u-ng…gunk* (Elliott 2004). When frightened, green frogs will leap into the water sounding an alarm call: *Eeek!* The release call of a green frog
is a long series of guttural notes (Wells 1978). Occasionally, males will utter 3 to 4 notes in rapid succession in a chorus that lasts 1 – 2 minutes. A call that is rarely heard is a low frequency growl 1.5-5.5 s long uttered by a male warning an intruder (Wells 1978).

Northern Leopard Frog (*Rana pipiens*)

Northern leopard frogs are green with black dorsal spots and a white line runs across the snout to the tympanic membrane. They are terrestrial frogs that prefer meadow and pasture habitat (Gibbs et al. 2005) with a high percentage of marsh and open water (Houlahan and Findlay 2003). They are aquatic breeders and overwinter in permanent ponds. Larvae hatch in 9 days or less and froglets emerge late July to early August (Seburn and Seburn 2000). Breeding activity begins in April and continues through the month of May. The call of a northern leopard frog sounds like a rattling snore followed by grunts or chucking noises (Elliott 2004); from a distance, its call is similar to the tapping of a woodpecker.

Pickerel Frog (*Rana palustris*)

The pickerel frog is green in colour with black dorsal spots similar to the northern leopard frog, it is smaller in size and the spots on its back are square and aligned along two dorsal lateral folds. Pickerel frogs breed in cold water streams (Holomuzki 1995) and inhabit deciduous forests as well as open field habitats (Hulse et al. 2001). Breeding activity begins in May, and tadpoles hatch within 12 to 18 days following deposition (University of Acadia, n.d.). The call of a pickerel frog is similar to that of a northern leopard frog; however, it is a quicker and higher pitch rattling tone and lacking the *chuck-chuck* note at the end of its call.
2.4 Anuran Breeding Habits

Anurans are semiaquatic species with a complex life cycle during which they depend on both wetland and terrestrial habitats. The species analyzed in this thesis breed in wetlands and many of them spend 11 months of the year foraging in terrestrial habitats. They congregate in ponds during species-specific breeding seasons which typically occur from April to July in southern Ontario.

Males begin advertising their arrival at ponds with species-specific choruses. When females arrive, males latch onto them with their thumbs hooked under the female; this is called amplexus (Kirby 1983). Male wood frogs will try to latch onto other males but will immediately dismount when the second male emits a release call (Berven 1981). Some females are unceptive towards latching males; and may also emit a release call causing the male to dismount (Diakow and Nemiroff 1981). Following successful amplexus, females will deposit eggs and males will fertilize them as they emerge.

Anurans are selective of their breeding habitat (Kiesecker and Skelly 2000) as the survival of their offspring is dependent upon the characteristics of the chosen pond (Crump 1991). The breeding habitat of some species can be identified by the defences that larvae have evolved against fish predation such as chemical repellents and predatory cues (Kats et al. 1988). Most species that utilize permanent ponds such as the bullfrog, green frog and American toad are toxic to fish (Kats et al. 1988). Non-toxic anuran larvae rely on behavioural defences to avoid predation; however, species that require fishless ponds typically lack these defences (Kats et al. 1988). Palatable species such as grey treefrogs may coexist with fish using chemical cues for
predatory detection; however, species that breed in isolated ponds, such as the wood frog, rarely encounter fish and consequently, do not exhibit these behaviours.

Species such as spring peppers and grey treefrogs breed in ephemeral ponds (Starnes et al. 2000). Spring peepers select ponds with little canopy cover (Skelly et al. 2002) because they develop more quickly in ponds with light exposure than shaded ponds covered by the forest canopy (Halverson et al. 2003). The grey treefrog can deposit egg masses in a range of habitats from shallow ditches to woodland pools (Starnes et al. 2000).

American toads are generalists but typically choose to oviposit eggs in ponds absent of wood frog tadpoles. Wood frog hatchlings begin to emerge when American toads deposit their eggs, and despite the toxicity and distaste of American toads, wood frog tadpoles consume them when available (Petranka et al. 1994).

Bullfrogs, green frogs (Kats et al. 1988) and northern leopard frogs (Light 1991) prefer to breed in permanent ponds. Bullfrogs select sites with substantial vegetation in order to reduce predation (Crump 1991). They territorially defend oviposition sites by floating in the water with their gular sac exposed while emitting both territorial and mating calls (Ryan 1980). Male green frogs also defend oviposition sites (Shepard 2002) by inflating their gular sac and occasionally utilize antagonistic behaviour such as patrolling, splashing, chasing, attacking and wrestling (Wells 1978).
The breeding habits of pickerel frogs differ from those discussed above. These frogs typically oviposition in channel margins of rivers within 10 cm of the shoreline (Holomuzki 1995). Frogs have antipredatory mechanisms so females deposit eggs in the head waters of river systems with a low density of predatory fish (Holomuzki 1995).

2.5 Monitoring Protocols

Several anuran monitoring programs have been initiated in the province of Ontario including the Amphibian Road Call Count, the Ontario Backyard Frog Survey and the Marsh Monitoring Program (Konze and McLaren 1997). These programs have been designed to monitor long-term trends in anuran populations using MCS conducted by volunteers across the province (Weeber and Vallianatos 2000). Anurans typically congregate in wetlands where male anurans voice species-specific breeding calls to attract females (Mohr and Dorcas 1999) and observers can use this opportunity to estimate species and abundance at a discrete location (Weir and Mossman 2005). MCS has been in use since the 1980’s and continues to be widely employed regionally and nationally as a standard monitoring technique (Dorcas et al. 2009).

The Amphibian Road Call Count and the Backyard Frog Survey were the first long-term anuran monitoring programs to be implemented in Ontario followed by the Marsh Monitoring Program (Konze and McLaren 1997). The Amphibian Road Call Count and the Backyard Frog Survey are coordinated by the Canadian Wildlife Service. Program coordinators provide volunteers with auditory learning material to identify the vocalization calls of each species (de
Solla et al. 2005). Volunteers survey sites by identifying species of calling males during mating season and noting the number and/or call level (Konze and McLaren 1997).

### 2.5.1 Amphibian Road Call Count

The Amphibian Road Call Count is conducted along road transects with count locations occurring at regular intervals. Road call transects are approximately 7 km in length with 10 stops and a minimum distance of 0.8 km between each stop (Konze and McLaren 1997). Observers must wait silently for a period of 30 seconds, before recording any calls to allow disrupted anurans to resume calling. Calls are recorded from all directions (360°) for a period of three minutes. Call counts are classified using 4 categories: call level 0 indicates no calling males; call level 1 is used when individual calls are not overlapping; call level 2 is used when the number of calling individuals is discernible with some overlap and call level 3 indicates a full chorus and is used when calls overlap and individuals are too numerous to count (Konze and McLaren 1997). Volunteers will record the number of individuals calling if calls are discernible (call level 1 or 2) (Konze and McLaren 1997). The call level ranks the number and intensity of anuran vocalizations and is a direct measure of calling males which is correlated with anuran abundance (de Solla et al. 2005).

There are three site visits that occur between April and July. In central Ontario (43°N – 47°N), the first site visit is conducted between 15-30 April when the temperature is at least 8°C (Konze and McLaren 1997). The second site visit occurs between 15-30 May, when the temperature is at least 13°C and the third site visit occurs between 15-30 June, when the temperature is at least 21°C (Konze and McLaren 1997). Volunteers are permitted to sample at
any time during these respective periods; however, they are encouraged to sample during optimal calling conditions. Such conditions consist of warm nights with light rain or fog and wind speeds less than 3 on the Beaufort scale (12-19 km/h) (Konze and McLaren 1997). Call counts begin at least 30 minutes after sunset and must be completed by midnight.

2.5.2 Ontario Backyard Frog Survey

The Backyard Frog Survey coordinates volunteers that collect data from their properties. This survey began in 1992 to validate results from the Amphibian Road Call Counts. The purpose of the survey was to determine whether Amphibian Road Call Counts were being conducted during the peak calling season for each species (Konze and McLaren 1997). In the southern tip of Ontario, volunteers begin documenting calls April 1 and northern sections of southern Ontario, volunteers begin surveys May 1. However, if anurans are heard calling before these dates, volunteers may begin recording prior to the official start date. The sampling period lasts four months; however, data collected outside the sampling period are also accepted (Konze and McLaren 1997).

Participants conduct daily surveys by standing outside their homes and listening quietly for a period of 3 minutes (Konze and McLaren 1997). Calls for each species are classified using call level 0-3 (Konze and McLaren 1997) and weather conditions are also documented. These surveys are very useful, as they document daily anuran calling patterns over a four month period.
2.5.3 Marsh Monitoring Program

The Marsh Monitoring Program (MMP) is a bi-national monitoring program that collects data in the Great Lakes region (Weeber and Vallianatos 2000). The MMP was founded by Bird Studies Canada and Environment Canada in 1994 and later expanded to include the U.S. Environmental Protection Agency and the Great Lakes Protection Fund (Weeber and Vallianatos 2000). The Long Point Bird Observatory coordinates volunteer observers in collecting amphibian and bird data within the Great Lakes marshes (Konze and McLaren 1997).

MMP surveys are conducted yearly and consist of three site visits per season. This protocol is utilized for marshes which are described as “…vegetated wet area, periodically inundated up to a depth of 2 metres with standing or slowly moving water.” (Konze and McLaren 1997, p.116). In the central portion of southern Ontario (43°N – 47°N), the first site visit occurs between 15-30 April when the temperature is at least 5°C (MMP 2008). The protocol states that surveys should begin at least 30 min after sunset and must be completed before midnight. Observers quietly wait for a period of 1 min to allow disrupted anurans to resume calling and listen for an additional 3 min documenting species presence using four letter species codes (Table 3.1). The number of individuals or call level for each species is noted using a categorical numbering system between 1 and 3 (Konze and McLaren 1997). Calls are counted within a 180° listening arc in front of the observer and indicated as calling within or beyond a 100 m radius (Fig. 2.2). The level of background noise is indicated using an index of 0-4 with 0 indicating no appreciable effect, 1 slightly affecting sampling, 2 moderately affecting sampling, 3 seriously affecting sampling and 4 profoundly affecting sampling MMP (2008).
Optimal survey conditions are believed to be warm, damp, foggy nights with a wind speed less than 3 on the Beaufort scale (12-19 km/h); however, the absence of wind is preferable (MMP 2008). Surveys should not be conducted in heavy wind or rain as these conditions can affect the ability of the observer to detect calls. The second site visit occurs between 15-30 May when the temperature is at least 10°C and a third site visit occurs between 15-30 June when the temperature is at least 17°C (MMP 2008). These dates are provided as guidelines and surveys may be conducted at an earlier date if climatic conditions are favourable.
2.5.4 North American Amphibian Monitoring Protocol (NAAMP)

Following the formation of MMP, 44 provinces and states came together under NAAMP to develop a standard protocol that was implemented throughout the United States (Nelson and Graves 2004). NAAMP establishes monitoring routes and distributes these to coordinating agencies. Each route is approximately 24 km long and has 10 stops (Weir and Mossman 2005). These stops can either be divided by habitat or by equidistance (at least 0.8 km apart) (Weir and Mossman 2005). A survey is conducted at each stop, where observers record species and call level (0-3) while listening quietly for a period of 5 min (Genet and Sargent 2003). Each state establishes 3 site visits based on NAAMP recommendations while an optional fourth site visit early in the season is permitted to target wood frogs (de Solla et al. 2005). Surveys begin at least half an hour after sunset and must be completed by 1 am (NAAMP n.a.). Surveys should not be conducted if wind speed is greater than level 3 on the Beaufort scale, nor should they be conducted during heavy rainfall as these factors may impair the ability of the observer to detect calls. The minimum temperatures for conducting surveys are established on a regional basis (NAAMP n.a.).

2.6 Effectiveness of Manual Call Surveys

MCS is the primary sampling method used for surveying anurans in Ontario (Konze and McLaren 1997). It is cost-effective, requires moderate training (Shirose et al. 1997) and may be conducted from a discrete location (Scott and Woodward 1994). It is also versatile where both ground dwelling and arboreal species can be detected during the same survey (Zimmerman 1994). Surveys contain information regarding species richness and population size (de Solla et al. 2005) and provide scientists, policy makers and organizations with long-term population data
(Weeber and Vallianatos 2000). The data collected by these programs allow interested parties to monitor densities and distributions of anuran populations over a long period of time at different spatial scales. These data can aid in planning and conservation activities, and raise awareness by getting citizens involved in the conservation process (Weeber and Vallianatos 2000, Brander et al. 2007). MCS protocols are also employed by environmental consulting firms in southern Ontario to formulate environmental impact reports for urban developments. Due to the widespread use of this sampling method, many researchers have sought to determine its effectiveness (Brander et al. 2007, Bridges and Dorcas 2000, Crouch and Paton 2002, de Solla et al. 2005).

Current protocols are touted as adequate for sampling the breeding activity of all species with minimal observer effort (Genet and Sargent 2003, Weir and Mossman 2005), while others have identified inadequacies. There is an agreement among some researchers that three to four site visits per year are sufficient for detecting species richness and abundance; however, some species call so infrequently that they may be difficult to detect using the current MCS protocol (Brander et al. 2007, Corn et al. 2000, Crouch and Paton 2002). Several anuran species including the pickerel frog, American toad, bullfrog, northern leopard frog and wood frog could be overlooked during MCS and may require more intensive survey methods such as egg mass counts (Brander et al. 2007) or drift fences (Crouch and Paton 2002).

Problems with call detection while employing the current protocol are well recognized (Brander et al. 2007, Bridges and Dorcas 2000, Crouch and Paton 2002, de Solla et al. 2005, Steelman and Dorcas 2010). Corn et al. (2000) evaluated the NAAMP protocol and found two
additional species using an ARS; they concluded that the protocol is poor at detecting rare species. Other studies which utilized an ARS found that 30% of species would have been missed if the NAAMP protocol were employed (Mohr and Dorcas 1999, Bridges and Dorcas 2000). Using results from the Ontario Backyard Frog Survey, where annual survey frequency can exceed 100, de Solla et al. (2005) found that three site visits per year underestimates species richness. To account for low detection probabilities, some programs conduct site occupancy adjustments (de Solla et al. 2005) by using a model to statistically adjust the occupancy rate of species (Mackenzie et al. 2002, Weir et al. 2005). However, this does not address the issue of species non-detection (de Solla et al. 2005) and imperfect species detection reduces survey effectiveness (Genet and Sargent 2003, Mazerolle et al. 2007, Weir et al. 2003, Weir et al. 2005). MCS can be an effective survey method if properly revised with an appropriate number of site visits (de Solla et al. 2005). Species richness is strongly correlated with sampling effort (de Solla et al. 2005, Gibbons et al. 1997) thus current knowledge leads us to believe that by increasing survey effort, the detectability of inconspicuous species can be improved.

Current protocols assume that all animals are active during the recommended sampling time. Due to interspecific daily and seasonal variation in anuran calling patterns, protocols are unable to consistently detect the presence of certain species (Bridges and Dorcas 2000, Brander et al. 2007, Crouch and Paton 2002). Detectability varies by species and guidelines need to be revised to reflect species-specific inconsistencies (Peterson and Dorcas 1992, Shirose et al. 1997). Peak calling periods of some species are known to occur outside the recommended sampling period. An ARS used to record variations in anuran calling activity over a 24 hour period found that some species, such as the southern leopard frog, call later in the evening once
the sampling period has ended (Bridges and Dorcas 2000). The peak call period of green frogs and bullfrogs also falls outside the recommended guidelines as they peak during the early morning hours, before sunrise (Mohr and Dorcas 1999). Chorus frogs and northern leopard frogs call with greater intensity during daylight hours (de Solla et al. 2005), while wood frogs were observed to call most persistently during the afternoon (Crouch and Paton 2002). Seasonal interspecific variances in calling patterns can also create inaccurate accounts of species presence or abundance if surveys are conducted outside the peak breeding season (Zimmerman 1994). While males with a prolonged breeding season have a greater probability of being detected by an observer, species with shorter breeding periods might be missed entirely if surveys are conducted infrequently.

Anurans with sporadic calling patterns present a problem for conventional MCS methods. In Rhode Island, spring peppers and green frogs called continuously during 16 min surveys, while pickerel frogs were detected infrequently during their peak (Crouch and Paton 2002). The pickerel frog is one of the most difficult species to detect utilizing MCS (Brander et al. 2007, de Solla et al. 2005) so it was concluded that call surveys should be at least 10 min in duration to detect all species including the pickerel frog (Crouch and Paton 2002). Shirose et al. (1997) evaluated the effectiveness of 3 min versus 5 min surveys and found little difference; however, when the survey time was extended, 18% of first detections occurred between 5 min and 15 min. Sporadicity of species calling patterns can also affect their daily detection probabilities. When survey duration was extended to 60 min, some species remained undetected despite calling several nights before or after the survey (Shirose et al. 1997) indicating that sampling once within a species breeding period does not guarantee detection.
Species non-detection can also occur as a result of pitch and timing of a species calling strategy. Northern leopard frogs, for example, were difficult to detect due to their calling characteristics. These frogs emit a basal call at low densities which make them difficult to distinguish amongst the concurrent high-frequency calling patterns of spring peepers (de Solla et al. 2006).

The effect of environmental variability on anuran call activity is complex and it is uncertain whether protocols recommend a sufficient number of site visits throughout the breeding season to accommodate for these effects (de Solla et al. 2005). Cues for the commencement of a species-specific breeding season as well as general variance in calling activity during the breeding season are influenced by a variety of factors including, air and water temperature, rainfall, humidity, etc. (Saenz et al. 2006, Steelman and Dorcas 2010). Many variables converge to provoke anuran call activity and these aspects may be difficult to measure experimentally. The impact of short-term climatic variability can be mitigated by employing more effective surveys established upon species-specific calling patterns.

MCS provide base line data for many studies ranging from identifying ecological criteria (Guerry and Hunter 2002) and causes for species decline (Knutson et al. 1999) to studying long-term population trends (Gibbs et al. 2005) and effects of metapopulation dynamics (Trenham et al. 2003). Species non-detection resulting from inadequate sampling effort (de Solla et al. 2005, Gibbons et al. 1997), can severely compromise the reliability of any study and if utilized by
environmental consulting firms, inaccurate results may impact the level of conservation within development projects.

Understanding the dominant factors controlling anuran breeding habits provide the necessary basis for auditory monitoring programs; however, temporal variability has never been studied in great detail. Further research is required to develop an effective protocol that maximizes species detection while minimizing observer effort. To achieve this, optimal survey frequencies can be identified using a detailed yet holistic approach to anuran calling patterns.

2.7 Establishing a Protocol using an ARS

The goal of this study is to maximize observer effort by determining the minimum sampling effort required to achieve an accurate portrayal of species richness. The optimal frequency and duration of seasonal surveys depends on a few key factors. It is important to recognize that calling strategies of anuran species are specific to individual breeding periods (de Solla et al. 2005). Protocol guidelines should maintain a biological approach by stipulating survey periods which coincide with the peak calling cycle of each species (de Solla et al. 2006). Distinctive and overlapping breeding periods pose a unique challenge in developing a set of comprehensive survey timelines and should be carefully studied. Equally important is the consideration of hourly calling patterns; recommendations must reflect the time of day during which each species calls most actively (Bridges and Dorcas 2000, Scott and Woodward 1994).

The information needed to understand these temporal trends requires that anuran calls be observed over a 24 hour period for the entirety of the breeding season (Peterson and Dorcas
Several studies have sought to identify anuran calling patterns by employing an ARS (Automated Recording System) (Saenz et al. 2006, Steelman and Dorcas 2010). This recording device can be left unattended, at a site of interest, for an extended period of time (Peterson and Dorcas 1994) and can minimize problems such as disruption to calling anurans and disturbance to the environment (Mohr and Dorcas 1999, Bridges and Dorcas 2000). It is a timer-based system that records data at a fine temporal scale to provide detailed information regarding the temporal variation in call activity, species composition and call intensity at a discrete location, without necessitating numerous site visits.

Peterson and Dorcas (1992) were one of the first to employ this method for the study of anuran call activity. They utilized a tape recorder controlled by a data logger set to record samples for 10 seconds every 5 minutes. Studies were conducted in Utah for a period of 5 days from 18-22 March, 1991 (Table 2.2). Solar radiation, relative humidity, wind speed and temperature were also recorded during this study.

In 1995, a series of ARS devices were utilized in northern Colorado (Corn et al. 2000). Systems were pre-assembled and included a voice clock and microphone. They were installed within 0.5 to 1.5 m above ground at water’s edge and calls were recorded for 12 seconds every half hour, on a 90-minute cassette tape. Tapes were changed every 5 days, requiring 9 to 16 visits per site. Each site had a weather station to document precipitation and changes in air and water temperatures. The ARS and data-logging systems were set up prior to the commencement of the breeding season and remained until the end of the breeding season.
In South Carolina, an ARS was used to record anuran activity from June 24 to July 18, 1998 (Mohr and Dorcas 1999). The system was composed of a cardioid microphone attached to a tape recorder which was controlled by a recycling timer. The equipment was placed in a tool box and a microphone was affixed to a nearby tree 2 m from the ground, at waters’ edge. Calls were recorded for 12 seconds every 30 minutes, for a period of 25 days.

Bridges and Dorcas (2000) employed an ARS to identify variations in calling activity in South Carolina. The system comprised a cardioid microphone and an analog tape recorder which was controlled by a recycling timer, and a voice clock was used to record an audio time stamp of the recording. This system was contained in a plastic box and the microphone was attached to a tree 2 m from the ground at waters’ edge. Data was collected for 12 seconds every half hour, for a period of 26 days between June 16 and July 12, 1997. Recordings were reviewed in the laboratory and the NAAMP call index was utilized to evaluate the call level.

Saenz et al. (2006) used an ARS to track the calling phenology of anurans in the state of Texas and identify the influences of abiotic factors on their breeding activity. Recordings occurred for a 2 year period from January 1, 2001 to December 31, 2002 at 8 sites. Instrumentation consisted of a cassette recorder, condenser microphone, voice clock and 6-cycle timer connected to a circuit board. One minute recordings were conducted at the beginning of each hour from 21h00 to 2h00. The number of calling individuals was noted. A maximum of 4 individuals at time could be discerned; therefore, if the number of calling anurans was > 4, a value of 5 was assigned. Temperature and rainfall were also documented throughout the period of this study.
Table 2.2. Recording frequency of ARS studies

<table>
<thead>
<tr>
<th>Study</th>
<th>Timeline</th>
<th>Recording Frequency</th>
<th>Daily Recording</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peterson and Dorcas (1992)</td>
<td>5 days</td>
<td>10 sec every 5 min</td>
<td>48 min/day</td>
</tr>
<tr>
<td>Corn et al. (2000)</td>
<td>Up to 74 days</td>
<td>12 sec every 30 min</td>
<td>9.6 min/day</td>
</tr>
<tr>
<td>Mohr and Dorcas (1999)</td>
<td>25 days</td>
<td>12 sec every 30 min</td>
<td>9.6 min/day</td>
</tr>
<tr>
<td>Bridges and Dorcas (2000)</td>
<td>26 days</td>
<td>12 sec every 30 min</td>
<td>9.6 min/day</td>
</tr>
<tr>
<td>Saenz et al. (2006)</td>
<td>2 years</td>
<td>1 min/hour between 21h00 to 2h00</td>
<td>6 min/day</td>
</tr>
<tr>
<td>Steelman and Dorcas (2010)</td>
<td>75 days</td>
<td>30 sec every 30 min</td>
<td>24 min/day</td>
</tr>
</tbody>
</table>

Steelman and Dorcas (2010) utilized an ARS to evaluate anuran calling activity at an ephemeral wetland in North Carolina from January 30 to April 15, 2007. The ARS consisted of a recorder, a condenser microphone and a 6-V battery which was charged using a solar panel. Anuran calls were recorded for 30 seconds every half hour. Recordings were copied to a computer on a 2 week interval. Environmental data loggers were also used to measure ground, water and air temperature, humidity, rain fall, barometric pressure, solar radiation, wind speed, gust speed, dew point, and light intensity during each auditory sample.

Using an ARS to create detailed calling profiles of anuran breeding patterns can help reduce uncertainties concerning detectability of elusive species as well as seasonal and hourly calling peaks of each species. ARS data can provide the necessary basis for creating species-specific calling profiles which can be used for the revision of monitoring protocols (Peterson and Dorcas 1992).
Chapter 3

3.0 Methods

Data for this study were collected at two sites within the Credit River watershed where MCS have been conducted for a period of 10 years. These data were utilized to test the effectiveness of MCS methods and to quantify detailed anuran call activity. Detailed calling profiles were developed using 10 min recordings conducted each hour of the day over a three month period. Graphical methods were employed to identify the hourly and seasonal trends exhibited by each species at peak frequency and intensity. Species-specific detection rates were calculated and trends were synthesized to establish interspecific daily and seasonal variations and similarities.

3.1. Site Selection and Description

Two sites within the Credit River watershed, namely, Warwick Conservation Area and Silver Creek Conservation Area, were chosen for this study. The criteria for this selection were based on species diversity and accessibility.

The Warwick Conservation Area is located at the corner of Heart Lake Road and Olde Base Line Road. Fig. 3.1 shows the property is located in a rural setting comprised of forests and agricultural fields. A narrow forest patch leads to a large permanent pond at the back of the property. The edges of the bog feature a diverse array of dense emergent vegetation and has a wet, spongy substrate composed primarily of organic material. A wide variety of shrubs, trees
and herbaceous plant life grow in the shallow regions of this bog. Towards the centre of the pond vegetation becomes sparse until it becomes open water habitat.

Figure 3.1. Warwick Conservation Area (adapted from Google Maps 2012).
Warwick was chosen for its isolation, species diversity and accessibility. The site has been monitored using the MMP protocol by Dr. Lorne Bennett and Dr. Rob Milne for 10 years and is known to provide habitat to six anuran species including, the American toad, bullfrog, green frog, grey treefrog, northern leopard frog and spring peeper. MCS and ARS surveys are conducted in the north-west portion of this property adjacent to the bog, indicated by the green arrow in Fig. 3.1. The property is owned by the CVC; however, access to the general public is prohibited as the entrance to the property is locked to prevent trespassers. Despite its limited access to the general public, this site is accessible to personnel, via a long driveway leading to the field house.

Silver Creek Conservation Area is located on Fallbrook Trail (9th Line) in Halton Hills. It is located within a gorge of the Niagara Escarpment with slopes rising 75 m above the valley on either side. A coldwater creek (Silver Creek) flows through the centre of the gorge providing fresh oxygenated water to the perennially inundated marsh. The marsh habitat acts as a transition between open water habitat at the centre of the site to the base of the forested hills above. The edges of the stream are characterized by emergent herbaceous vegetation as it is predominantly a cattail marsh.

Silver Creek was chosen for an ARS study because of its species diversity and accessibility. The location of the ARS is indicated by the green arrow in Fig 3.2. MCS have been conducted at this site for 10 years from a road side location. The site provides habitat to six anuran species including, the American toad, green frog, grey treefrog, northern leopard frog, pickerel frog and spring peeper. The pickerel frog is the primary species of interest as its calling
pattern is sporadic and site occupancy can be difficult to detect (de Solla et al. 2005). This is the only site in the Credit River watershed where it has been documented consistently. Silver Creek is owned by the CVC; however, it is open to the public and commonly used for recreational purposes. The Bruce Trail and several side trails in the vicinity are frequented by hikers and the site is occasionally visited by paddlers.

Figure 3.2. Silver Creek Conservation Area (adapted from Google Maps 2012).
3.2 Sampling

3.2.1 Wildlife Acoustics Song Meter

Detailed chronological data are required to determine whether current protocol guidelines are effective. These data were collected using an ARS to record calls on an hourly basis for the duration of the breeding season. A similar system was used by Peterson and Dorcas (1992), Mohr and Dorcas (1999), Corn et al. (2000), Bridges and Dorcas (2000), Saenz et al. (2006) and Steelman and Dorcas (2010) to document anuran calling patterns. This method was chosen because it is able to provide quality recordings of call activity throughout the day without necessitating frequent site visits. While it is otherwise impossible to physically survey wetlands at the frequency required, this recording device provides the means for an intensive survey schedule. Any potential bias caused by human presence or disturbance during MCS can also be eliminated using this method (Mohr and Dorcas 1999, Bridges and Dorcas 2000).

Systems utilized in previous studies were assembled by attaching a cardioid microphone to a tape recorder and using a timer to control the recording frequency while a voice clock time stamped the recording (Mohr and Dorcas 1999, Bridges and Dorcas 2000, Saenz et al. 2006). Other systems were pre-assembled containing a microphone and voice clock (Corn et al. 2000).

This study utilizes a comprehensive system called the Wildlife Acoustics Song Metre (SM2) Terrestrial Acoustic Package. The Song Metre is a computerized system which digitally records calls in stereo using two microphones with a sensitivity of -36±4dB and a frequency response of 20Hz to 20,000Hz. The recording frequency is based on the schedule entered into the
built-in timer. Each sample is saved onto a 32 GB-SD card using a 16-bit PCB (.wav) format. The sound file is time-stamped with the date and time of the recording. The system is contained within a waterproof metal casing; however, the microphones remain exposed, as seen in Fig. 3.3.

**Figure 3.3.** Wildlife Acoustics Song Meter at Warwick.

Wildlife Acoustics Song Meter Sampling Design

The Song Meter was used at two anuran breeding sites to record calls during a 24 hour period (Corn et al. 2000, Mohr and Dorcas 1999). Recording at Warwick began April 5 at 6:30 pm and concluded July 7 at 2:30 pm. The recording device was attached to a tree 2 m above ground at waters’ edge (Mohr and Dorcas 1999). Recording at Silver Creek began April 5 at 6:30 pm and
concluded July 6 at 7:30 pm. The recording device was attached to a tree 0.5 m above ground at water’s edge (Corn et al. 2000). Silver Creek is accessible to the public and for this reason the recorder was installed behind a small bush to hide the device from potential vandals. Unfortunately, this made the system more accessible to wildlife and the microphones which were attached to either side of the system were destroyed resulting in a loss of 24 days (May 25 to June 17) of call data.

Both loggers were programmed to begin recording for a period of 10 min (Crouch and Paton 2002) every hour on the half hour; resulting in 4 hours of recording per site/day. This differs greatly from other similar studies which have recorded between 6 to 24 min per day (Table 2.2). The SD cards used in the device provided 32 GB of storage and cards were changed every 3 to 4 weeks. At Warwick, the first batch of files was collected April 28, followed by subsequent collections May 20 and June 25 and the retrieval of the recorder on July 7. At Silver Creek, the first batch of files was collected April 26, subsequent collections occurred May 9 and June 7, and the system was retrieved July 6. The .wav files were downloaded to a computer and were converted to mp3 format for easy storage using the Lame Drop program. There were no discernible differences between the .wav and mp3 files. Sound files were played on a laptop and were carefully reviewed using headphones. Several different methods were explored such as computer speakers and a Denon sound system; however, headphones were found to provide the greatest level of detail with the least white noise.
Species presence was recorded in a spreadsheet while detailed information was recorded in a database. A new entry was created in the database for each species detected during a 10 min recording. Every entry contained information regarding the site, species, date, time and call level for each 3, 5 and 10 min interval. Any wind and/or rainfall that may have impacted the clarity of the audio recording were also documented for each entry. The main table contained a unique record for each date and time a species was heard calling at a location. The maximum call level was recorded for each interval: within the first three minutes, from 3 min to 5 min and from 5 min to 10 min. Numerous queries were created to data mine the main table to retrieve information described in Section 3.3.

3.2.2 Manual Call Surveys

For the past 10 years, MCS were conducted at Warwick and Silver Creek Conservation Areas using the MMP protocol described by Konze and McLaren (1997). The MMP was created by Bird Studies Canada to monitor anuran populations in marshes and the method was adapted by Dr. Lorne Bennett and Dr. Rob Milne to monitor all inundated areas including bogs, fens and swamps. This method has been employed for the past 10 years and has yielded useful population data.

The MMP protocol stipulates that the optimal sampling period is dependent upon the geographical location of the monitoring site. The Credit River watershed is located north of the 43° N parallel and falls within the region of central Ontario (43° N – 47° N). The MMP states that within central Ontario, the first batch of surveys will occur within the last two weeks of
April when the air temperature is at least 5°C (MMP 2008). Over the past 10 years, this first round of surveys has been conducted between 15-30 of April. In accordance with the protocol, the second site visits were conducted between 15-30 of May, when the temperature was at least 10°C. The third site visits were conducted between 15-30 of June when the temperature was at least 17°C (MMP 2008).

Surveys began at least 30 min after sunset and were completed by midnight (MMP 2008). Observers stood quietly for 1 min before beginning the count to allow any disrupted anurans to resume calling. The approximate location of calls occurring within a 180° listening arc were documented relative to 100 m radius. The number of individuals and call level of each species were also recorded on the diagram. If individuals were too numerous to count, the call level was the only abundance measure indicated. Call counts were classified using three categories: call level 1 was used when individual calls were not overlapping and easily counted; call level 2 was used when the number of calling individuals was discernible with some overlap and counts were estimated; and call level 3 was used when calls overlap and individuals were too numerous to count (Konze and McLaren 1997).

Climatic conditions such as wind speed and humidity (Saenz et al. 2006, Steelman and Dorcas 2010) affect call frequency and intensity, consequently the MMP specifies that optimal survey conditions are warm, damp, foggy nights with a wind speeds less than 3 on the Beaufort scale (12-19 km/h) (MMP 2008). These recommendations were followed as closely as possible
in order to optimize survey effectiveness. If winds became too strong as the evening progressed, surveys were cancelled and repeated during more appropriate weather conditions.

3.3 Data Analysis

Data for this study have been collected to measure the effectiveness of current monitoring protocols and to recommend improvements based on the trends that surface during the analysis. A range of descriptive methods have been utilized to address the objectives of this study.

The MMP, among others, utilizes a standard coding system to record the presence of calling species. Consistent with the standard protocol, this study has opted to use the same coding system throughout this analysis. The species codes for anurans found within the study area are listed in Table 3.1. Additional codes have also been utilized to indicate whether a species is calling from Warwick (-W) or Silver Creek (-S) locations.

Table 3.1. Site codes and standard species codes from MMP (2008).

<table>
<thead>
<tr>
<th>Common Name</th>
<th>Species Code</th>
<th>Common Name</th>
<th>Species Code</th>
<th>Site</th>
<th>Site Code</th>
</tr>
</thead>
<tbody>
<tr>
<td>American toad</td>
<td>AMTO</td>
<td>Northern leopard frog</td>
<td>NLFR</td>
<td>Warwick</td>
<td>-W</td>
</tr>
<tr>
<td>Bullfrog</td>
<td>BULL</td>
<td>Pickerel frog</td>
<td>PIFR</td>
<td>Silver Creek</td>
<td>-S</td>
</tr>
<tr>
<td>Green frog</td>
<td>GRFR</td>
<td>Spring peepers</td>
<td>SPPE</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grey treefrog</td>
<td>GRTR</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### 3.3.1 Evaluation of Survey Methods

This study seeks to compare the effectiveness of standard MCS protocols with data acquired from an ARS. To achieve this, it is important to determine the ability of each method to accurately detect the presence of each species at a site during a given season. If conducted correctly, standard MCS should return consistent results of species presence while the Song Meter should provide accurate recordings of calling anurans. Both methods must be evaluated for their effectiveness and species detectability.

#### Evaluation of Manual Call Surveys

A temporal analysis of species detection utilizing the standard MCS protocol was conducted by summarizing 10 years of survey data. A table was created for each site, with the species/sites codes listed horizontally and the years enumerated vertically. Annual site visits were colour coded according to number of times a species was detected during a season. The number of species detections occurring during each first, second and third site visit were summed and divided by 10 (number of years surveys were conducted). The detection rate was converted to a percentage to display the calling rate for each species.

#### Evaluation of Song Meter Detection

To evaluate the effectiveness of the Song Meter, it was important to determine whether the calls recorded on the device were reflective of those occurring in the field. This can provide us with information regarding the sensitivity of the recording system and clarify the accuracy of our results. To identify the level of sensitivity of the instrumentation, three MCS were conducted
while the Song Meter was recording. Results listed in Table 4.1 indicate the species and call level heard during each of the three surveys (early, mid and late) for both the ARS and MCS.

3.3.2 Species Richness Measures

General calling trends provide a foundation upon which protocol guidelines can be based. Identifying general seasonal and hourly calling trends also creates a basis upon which further analysis can be conducted.

Anuran Call Period

General species richness trends can be easily compared by displaying the call period of each species along a timeline to identify periods of overlap that feature the greatest species diversity. These represent important periods of the season during which surveys should be conducted to obtain accuracy of site occupancy. The calling period of a species can be determined by noting the first and last call detected during the breeding season. Periods of time with the greatest species overlap provide a basic estimate of when site visits should occur to detect the greatest number of species.

The seasonal call period of each species was plotted onto a single horizontal bar graph to illustrate the overlap between breeding periods. The first and last day of species detection was plotted to display the duration of the species calling period. The calendar date was converted into a numerical value beginning with the first day of recordings, April 5, as Day 1, ending with July
7 as Day 94. The species/site codes were used as a descriptor and each bar displayed the temporal range of the species within the confines of the study period.

Interspecific Daily Variation

Identifying hourly trends with the greatest species diversity is the first step to selecting an optimal time for site visits. By combining species diversity with a measure of call intensity, an hourly representation of call activity was used to identify the time of day with the greatest overall detection. This was used to determine the hourly call period during which all surveys should be conducted.

This was completed by isolating hours with the greatest species diversity and call intensity, a graph was created to illustrate the hourly surveys with the greatest activity. This was done by creating a query to isolate all the sampling times with call levels > 1. Call level 1 is often heard throughout the day; meanwhile, call level 2 or 3 indicate periods of heightened activity. To identify periods with the greatest activity and species diversity, all entries with a minimum of two species each calling with a minimum call level 2, were sorted by time. All sorted data fell within the hours of 20h30 to 5h30 and a count was conducted to produce a sum of surveys for each hour. Data were plotted onto a vertical bar graph to display interspecific hourly calling trends.
A chi-square test was performed on these nominal data to determine whether there was a significant difference in the hourly variation of anuran diversity and intensity. This test was selected because it verifies the goodness-of-fit by comparing frequencies of a nominal variable to the expected frequency (average of observed values). Further testing revealed that there was no significant difference in call activity from 21h30 to 4h30 hence these parameters were used for subsequent analysis. Chi-square tests were also conducted for individual species to determine whether there is a significant difference in their hourly calling patterns; results were summarized in Table 4.2.

3.3.3 Species Calling Profiles

Monitoring surveys should seek to detect species during their peak calling periods to produce dependable population profiles. Identifying the optimal time of day and time of year to conduct MCS is important when trying to develop an effective protocol. The need increases when trying to detect species of interest, thus individual calling profiles must be created for each species within a region. If the calling strategies of each species can be clearly defined and reflected in monitoring guidelines, the probability of detecting site occupancy will increase.

Frog Grapher

Seasonal and hourly graphs were produced using a program specifically designed by Mark Sohm. The graphing program was created using a custom Java application. The graphs were drawn using the JFreeChart version 1.0.14 library (http://www.jfree.org/jfreechart/). The stacked bar graph type was used with a customized StackedBarRender, CategoryAxis and ValueAxis.
Data for this application were retrieved from the main table. The application connected to the database using JDBC (Java Database Connectivity) and iterated through the data using multiple loops and SQL queries. The application combined and calculated the relevant elements and inserted them into unique tables. The data were queried from a table, by counting the frequency of each call level $> 0$ for each day/hour for a specified species and location.

Seasonal Calling Frequency

The seasonal calling period of each species as well as the time of year during which each species calls with the greatest frequency and intensity is represented by a series of graphs. These graphs feature total daily call frequencies of each call level to determine the time of year during which a species calls with the greatest frequency and intensity. A series of queries were used to determine the number of days each species was detected. These results were used to calculate the species daily detection rate at a given site to identify the periods of time that experience the greatest calling activity. This was subsequently combined with graphical data to determine the time of year during which each species calls with the greatest frequency.

Seasonal graphs were created using the Frog Grapher which queried the daily call level frequencies of each species/site from the database and plotted it in a vertical stacked bar graph as seen in Fig. 3.4. The daily frequency count of call codes is indicated by the left axis, while the detection rate (%) is indicated by the right axis. The daily frequency count is a simple count of call levels beginning with the frequency of call level 3 counts at the bottom of the bar, the frequency of call level 2 counts in the middle and the frequency of call level 1 at the top of the
bar. Each bar represents a single day during which the species was heard calling and the height of the bar indicates the number of times (surveys) the code was detected during a given day. The detection rate is a percentage which represents the number of times a species was heard calling on a given day divided by 24 (number of surveys conducted per day). Since hourly surveys were conducted on a daily basis, the maximum possible count is 24 as indicated by the frequency axis.

![Graph Example of daily call level graphs.](image)

**Figure 3.4.** Example of daily call level graphs.

The daily detection rate is expressed using a tabular format that is used to identify the days with the greatest species activity and the time of year with which each species calls with the greatest frequency. A query was created that produced a total daily count of 10 min surveys when calls were detected for each species/site. The data were reduced to a binary detection/non-detection (1, 0 respectively) to facilitate the calculation of detection probabilities (Brander et al. 2007). The daily detection rate was expressed as the proportion of surveys with detections. The
detection rate was calculated by dividing the total daily call count by 24 (number of surveys conducted per day), and converting to a percentage.

Hourly Calling Frequency

The hourly calling pattern of each species was determined using a series of tables and graphs. A series of stacked bar graphs were used to identify the hour(s) of the day during which each species calls with the greatest frequency and intensity, representing the period of time with the greatest probability of species detection. A table was used to determine the probability of hearing a given species at a specified hour of the day by calculating the species detection rate. This facilitates the determination of the number of site visits required to detect the species if surveys are conducted at a given hour.

Hourly stacked bar graphs were created for each species/site using the Frog Grapher (Fig. 3.5). Graphs contain 24 bars, one for each hour of the day, representing the number of times a species was heard at a given time during its call period. The x axis represents the hour of the day when species were detected rather than the time of day. This was done to avoid confusion with the word “time” and to simplify the text in the axis label. The frequency count of call levels was included, beginning with the frequency of call level 3 at the bottom of the bar, the frequency of call level 2 in the middle and the frequency of call level 1 at the top of the bar. The frequency count indicated by the left axis, represents the number of days the species was heard calling at a given time; therefore, the maximum possible height of the bar is equal to the total number of days the species was detected during its call period.
Figure 3.5. Example of hourly call level graphs.

A table was created to illustrate the probability of hearing a given species/site during a specified hour of the day. It was created by conducting a query for each species/site that produced a total hourly count of surveys with calls for each interval (3 min, 5 min and 10 min). The hourly detection rate was calculated for each time interval by dividing each count result by the call period. The data were expressed as a proportion of surveys with detections to facilitate the calculation of detection probabilities. The call period was determined by summing the number of days between the first detection of a given species/site, to the last detection of the species/site. If a single call was followed or preceded by an extended period of silence, it is assumed to be an outlier and possibly provoked by other factors (e.g. startle or warning call) and not part of the calling period. In this case, several days may have been subtracted from the calling range to reflect the actual call period.
Using the hourly detection rates calculated above for the hours between 21h30 and 4h30 (period of equal detection), monthly bar graph were created for each species/site depicting their nocturnal hourly calling patterns (Fig. 3.6). Monthly average nocturnal detection rates (MANDR) were calculated by averaging the detection rates between 21h30 and 4h30 for the 3 min survey and results were summarized in Table 4.4. The nocturnal detection rates before and after midnight, were calculated by averaging the detection rates from 21h30 to 23h30 for before midnight and averaging detection rates from 0h30 to 4h30. Results were summarized in Table 4.5.

Figure 3.6. Example of hourly detection graphs.

3.3.4 Interspecific Seasonal Variation

The formulation of an effective monitoring protocol requires that the calling profiles of each species be combined into a series of optimal calling periods. To identify these optimal calling periods, call strategies of all anuran species must be analysed holistically to uncover temporal
trends. If general trends can be established, then effective guidelines can be produced to improve survey accuracy and efficiency. In selecting the optimal number and frequency of sampling nights, it is important to identify the detection probabilities of each species. Any species with a detection rate < 100%, is not guaranteed to return accurate results of species occupancy. Increasing the number of site visits can produce a better estimation of occupancy rates (MacKenzie et al. 2002); therefore, the number of site visits should aim to accurately affirm species detection (100%) during a given sampling season.

A rudimentary estimate of site visits was determined based on species-specific detection rates. The MANDR was used to estimate the minimum number of site visits required to detect a given species during its call period. A simple calculation (equation: \# site visits = \frac{100\%}{\text{MANDR}}) was used to determine the number of site visits required to reach 100% detection. Any species with a detection rate < 100% can be missed if sampled during its breeding season (Shirose et al. 1997) thus the species should be sampled at least twice during their calling period to increase chances of detection. The total number of site visits was determined by plotting the number of site visits required for each species on a calendar during its peak calling period and identifying those which overlap. With successive years of study, a more statistically accurate estimate of site visits can be determined.

3.3.5 Survey Duration
Identifying the optimum survey duration has been highly debated over the past decade. Studies by Crouch and Paton (2002), de Solla et al. (2005), and Shirose et al. (1997) have sought to
determine the most effective amount of time required to detect all anuran species. The intensive sampling frequency utilized in this study provides the ideal data set to answer such questions.

Monthly Detection

The magnitude of call activity varies with time and the sampling effort required for species detection can vary by month. The increase in detectability with sampling effort was calculated by querying the number of calls occurring within the first 3 min. A second query was conducted summing the number of first detections between 3 min and 5 min (when no calls were detected in the first 3 min). Lastly, the number of first detections between 5 min and 10 min were queried (when species was not heard within the first 5 min). The increase in first detection with time was calculated for each interval (3 min-5 min and 5 min-10 min) by dividing the total number of detections within the interval by the total number of detections heard in the previous interval. Table 4.8 summarizes the increase in detection from 3 min to 5 min and 3 min to 10 min.

A series of monthly graphs have been created to display the time to first detection for each species. Data above were tallied for each species/month to identify the increase in monthly detection with sampling effort. The total for each interval was divided by the number of surveys with species detections for a given month. Surveys with detections were graphed to illustrate the percentage of first detections occurring during the 3 min, 5 min and 10 min intervals. Graphs were limited to nocturnal calling patterns (from 21h30 to 4h30), as these hours were found to produce significantly higher calling frequencies. The graphs were used to determine the amount of time required to detect the species calling within a given month (Fig 3.7).
Figure 3.7. Example of graph illustrating monthly patterns of time required for first species detection.

Hourly Detection
To indentify the optimal survey duration, the temporal variation in calling strategies of each species were established. The amount of time required to detect a species should be analyzed based on time of day and time of year as calling frequency can vary throughout the breeding season. A table was produced summarizing the increase in species detectability from 3 min to 5 min and from 3 min to 10 min. The increase in detection with sampling effort was calculated for the period of time from 21h30 to 4h30. A count of first detections occurring within a given month for each species/site was determined for each time interval. The increase in detection was calculated by dividing the number of first detections within a time interval by the total number of detections occurring during that hour within a given month. Table 4.7 depicts the nocturnal hours with the highest and lowest species detection rate within the first 3 min. These results provide an estimate of times when an observer is likely to hear the greatest diversity in the shortest amount of time for a given month.
The increase in detection with sampling effort was also determined by calculating the average increase before midnight (21h30-23h30) and after midnight (0h30-4h30). The average increase in species detection from 3 min to 5 min and 3 min to 10 min was calculated by taking the average (before and after midnight) of the hourly detection increase calculated above. These results are summarized in Table 4.8. This table provides the necessary information for determining the most effective survey duration for a given month based on the hour of the survey.
Chapter 4

4.0 Results

A comparison of MCS and ARS techniques revealed that MCS did not provide an accurate portrayal of species richness at Silver Creek. Using the conventional protocol, in 2011, < 50% species richness was detected at Silver Creek while detection at Warwick was perfect. During the 10 year study, American toads and grey treefrogs experienced the lowest detection rates while pickerel frogs, bullfrogs and northern leopard frogs experienced moderate levels of detection. Spring peepers and green frogs were detected most consistently during the study period (2003 to 2012).

Detailed auditory data confirmed calling patterns of seven anuran species. A loss of data made it difficult to establish the calling trends of species detected at Silver Creek. American toads and grey treefrogs exhibited sporadic calling patterns and had the lowest detection rates while spring peepers and green frogs called consistently throughout their breeding period. Bullfrogs had high detection rates while pickerel frogs were difficult to detect. Species-specific calling profiles were depicted using a series of graphs and tables, and general calling trends were inferred.
4.1. Comparison of Survey Methods

4.1.1 Manual Call Survey Results

MCS were conducted for a period of 10 years at both sites and species detection was inconsistent from year to year at both sites. The evaluation of the MMP standard protocol varied significantly as species detection ranged from 0% to 100%.

![Species detection rates of MCS at Warwick using MMP guidelines.](image)

**Figure 4.1.** Species detection rates of MCS at Warwick using MMP guidelines.

Species detection at Warwick was not consistent year to year (Fig. 4.1). Green frogs and spring peepers were detected most regularly using the MMP and were often detected on more than one visit per season. Bullfrogs, grey treefrogs and northern leopard frogs were frequently detected at a rate of 70% over the past 10 years. American toads were the most difficult to detect.
at 60% hence were absent from 40% of surveys over the past 10 years. Bullfrogs were primarily restricted to detection in June and northern leopard frogs were restricted to detection in April, while American toads called during all three visits. In 2011, all species were detected using both MCS and the Song Meter.

Species detection using MCS at Silver Creek varied from the results acquired during the ARS study (Fig. 4.2). The Song Meter permitted the use of an intensive protocol and the instrument was capable of recording the presence of American toads which were previously undetected using MMP guidelines. Grey treefrogs, northern leopard frogs and a bullfrog were also detected by the Song Meter in May and June of 2011 despite being undetected by MCS conducted during the same time period. Grey treefrogs exhibited a low detection rate at Silver
Creek; being noted only twice in the past 10 years using MMP guidelines. Northern leopard frogs had a high detection rate in MCS over the 10 year study period but were undetected in 2011 MCS. This non-detection was inaccurate as frogs were recorded in April and May using the Song Meter. Overall, MCS produced low detection results in 2011 with only three of the seven species (43% species richness) detected during breeding season.

### 4.1.2 Song Meter Detection

MCS and Song Meter results were documented simultaneously to compare the detection capabilities of each method. A comparison of three surveys, one for each visit (early, mid and late visits), resulted in the detection of four different species (Table 4.1).

<table>
<thead>
<tr>
<th>Site</th>
<th>Date</th>
<th>Time</th>
<th>Species</th>
<th>MCS - CL</th>
<th>ARS - CL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Silver Creek</td>
<td>April 26</td>
<td>22:34</td>
<td>SPPE</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>May 24</td>
<td>22:30</td>
<td>GRFR</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>SPPE</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>PIFR</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Warwick</td>
<td>June 20</td>
<td>21:30</td>
<td>BULL</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>GRFR</td>
<td>1</td>
<td>1</td>
</tr>
</tbody>
</table>

The ability of the Song Meter to detect species presence and call level was similar to that of the MCS. Spring peepers, green frogs and bullfrogs were detected at the same call levels using both survey methods. The pickerel frog was detected May 24 during MCS but was undetected by recordings occurring concurrently.
4.2 Audio Recordings

Recordings at both Silver Creek and Warwick Conservation Areas began April 5, 2011 at 18h30 and continued until early July. The Song Meter recordings at Warwick were discontinued July 7, resulting in 94 sampling days and 22,390 minutes of audio. At Silver Creek, recordings were terminated July 6; however, due to damage to the equipment, audio was interrupted from May 25 until June 17, a total of 24 days. Damage appeared to have been caused by wildlife chewing on exposed microphones. When the recorder was restored, wire mesh was fastened over the microphones for protection. Despite the disruption, 71 days of data were recorded at Silver Creek, resulting in 16,646 minutes of audio.

4.3 General Daily and Seasonal Trends

General trends provide an overview of seasonal and hourly periods with the greatest species diversity and abundance. These trends also provide a basis upon which protocol guidelines can be formulated.

4.3.1 Anuran Calling Period

Breeding commenced with spring peepers, on April 9 (recording day 4) (Fig. 4.3). With the exception of April 18 and 19, anurans were heard nightly at Warwick until audio was terminated July 7 (day 94). While the recorder was in operation at Silver Creek, frogs were heard nightly from April 9 to July 6 (day 93), when the audio was terminated. Calls were detected for 88 days at Warwick, and 67 days at Silver Creek.

The succession of species is illustrated in Fig. 4.3. At Warwick, the breeding season began with spring peepers (April 9) and continued with northern leopard frogs (April 14-day 10),
American toads (May 5-day 30), green frogs (May 19-day 44), grey treefrogs (May 19) and bullfrogs (May 23-day 48). The succession of species at Silver Creek was similar to that of Warwick beginning with spring peepers (April 9) followed by northern leopard frogs (April 30-day 25), American toads (May 10-day 35), green frogs (May 5), grey treefrogs (May 11-day 36) and pickerel frogs (May 23). Fig. 4.3, also illustrates the greatest species overlap occurred between day 40 (May 15) and day 50 (May 25).

![Call Period Graph](image)

**Figure 4.3.** The calling period of each species detected during the ARS survey.

### 4.3.2 Interspecific Daily Variation

Surveys with a call level and species count > 1 were queried and a total of 248 surveys were returned. All surveys fell between 20h30 and 5h30. A chi-square test was conducted on this data set. The test indicated a significant difference in the level of detection ($p < 0.000$) during this time. As seen in Fig. 4.4, the greatest account of species presence and abundance occurred between 21h30 and 4h30 with a maximum frequency at 1h30. To isolate a period of equal
detection, a second chi-square test was conducted with the removal of 5h30 and 20h30 from the
data set, as these hours had much lower results. The second test indicates that no significant
difference in detection \((p=0.196)\) exists between 21h30 and 4h30.

![Figure 4.4](image)

**Figure 4.4.** Hourly frequency of surveys with call level and species count > 1.

Chi-square \((\chi^2)\) tests were conducted for each species/site to determine whether frequency of
calls varied significantly throughout the day. The \(p\) value for most species was low \(<0.000\),
indicating a significant difference in frequency of calls throughout the day. However, the \(p\)
values for American toads \((0.261)\) and green frogs \((1)\) at Silver Creek indicate that there is no
significant difference in frequency of calls throughout the day (Table 4.2).

**Table 4.2.** Results of chi-square \(\chi^2\) tests \((p\) value\) conducted on hourly call frequencies.

<table>
<thead>
<tr>
<th></th>
<th>BULL-W</th>
<th>AMTO-W</th>
<th>AMTO-S</th>
<th>GRFR-W</th>
<th>GRFR-S</th>
<th>GRTR-W</th>
<th>NLFR-W</th>
<th>NLFR-S</th>
<th>SPPE-W</th>
<th>SPPE-S</th>
</tr>
</thead>
<tbody>
<tr>
<td>All</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
<td>0.261</td>
<td>&lt;0.000</td>
<td>1</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
</tr>
<tr>
<td>21h30 - 4h30</td>
<td>21h30 - 4h30</td>
<td>&lt;0.000</td>
<td>0.196</td>
<td>&lt;0.000</td>
<td>1</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
<td>&lt;0.000</td>
</tr>
</tbody>
</table>


4.4 Species Calling Profiles

Calling profiles outline the temporal breeding variability of each species. The calling period is summarized in Table 4.3 highlighting the seasonal and hourly periods of heightened frequency and intensity.

<table>
<thead>
<tr>
<th>Warwick</th>
<th>Breeding Season</th>
<th>Duration of CP</th>
<th># Days Detected</th>
<th>Day (Max Freq.)</th>
<th>Day (Max Intensity)</th>
<th>Time (Max Freq.)</th>
<th>Time (Max Intensity)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AMTO</td>
<td>May 5 – June 20</td>
<td>47</td>
<td>25</td>
<td>May-13</td>
<td>May-13</td>
<td>0h30</td>
<td>23h30-0h30</td>
</tr>
<tr>
<td>BULL</td>
<td>May 23 - July 7</td>
<td>46</td>
<td>37</td>
<td>June 5, July 3</td>
<td>June 17, 18</td>
<td>0h30, 2h30</td>
<td>1h30-2h30, 23h30</td>
</tr>
<tr>
<td>GRFR</td>
<td>May 19 - July 7</td>
<td>50</td>
<td>49</td>
<td>June 15, 21, July 5</td>
<td>July-03</td>
<td>0h30</td>
<td>3h30</td>
</tr>
<tr>
<td>GRTR</td>
<td>May 19 - June 26</td>
<td>39</td>
<td>27</td>
<td>June 7, 9</td>
<td>June 7, 9</td>
<td>21h30-23h30</td>
<td>22h30</td>
</tr>
<tr>
<td>NLFR</td>
<td>April 14 - May 29</td>
<td>46</td>
<td>35</td>
<td>May-02</td>
<td>May 7, 9, 20</td>
<td>0h30-2h30</td>
<td>1h30-2h30</td>
</tr>
<tr>
<td>SPPE</td>
<td>April 9 - June 26</td>
<td>79</td>
<td>52</td>
<td>Apr-29</td>
<td>May-14</td>
<td>1h30, 21h30</td>
<td>21h30</td>
</tr>
</tbody>
</table>

Silver Creek

<table>
<thead>
<tr>
<th>Breeding Season</th>
<th>Duration</th>
<th># Days Detected</th>
<th>Day (Max Freq.)</th>
<th>Day (Max Intensity)</th>
<th>Time (Max Freq.)</th>
<th>Time (Max Intensity)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AMTO</td>
<td>May 10 - May 23</td>
<td>14</td>
<td>8</td>
<td>May-14</td>
<td>May-23</td>
<td>21h30</td>
</tr>
<tr>
<td>GRFR</td>
<td>May 5 - July 6</td>
<td>63</td>
<td>30</td>
<td>N/A</td>
<td>N/A</td>
<td>0h30 - 4h30, 22h30</td>
</tr>
<tr>
<td>GRTR</td>
<td>May 11 to June 28</td>
<td>49</td>
<td>9</td>
<td>N/A</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>NLFR</td>
<td>April 30 - May 25</td>
<td>26</td>
<td>25</td>
<td>May-08</td>
<td>May-14</td>
<td>0h30</td>
</tr>
<tr>
<td>PIFR</td>
<td>May 23 - June 24</td>
<td>33</td>
<td>5</td>
<td>N/A</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>SPPE</td>
<td>April 9 - June 28</td>
<td>81</td>
<td>59</td>
<td>Apr-27</td>
<td>Apr-23</td>
<td>21h30-22h30</td>
</tr>
</tbody>
</table>

4.4.1 American Toad

In 2011, the peak calling period of American toads occurred in May. American toads began calling at Warwick on May 5 and were last heard calling June 20 (Fig. 4.5) and were heard 25 days of this 47 day calling period. At Warwick, there were 14 days when the American toad was detected > 1 survey per day. These periods occurred from May 8 to May 15 and May 19 to May
23 displaying a slightly bimodal distribution (Fig. 4.5). The initial period of heightened call frequency and intensity (call level) exhibited perfect species detection on May 13. The secondary peak in call activity occurred between May 19 and May 23, thereafter toads were heard infrequently at call level 1 and 2 until June 20.

**Figure 4.5.** Daily call level graph depicting calling patterns of American toads at Warwick.

At Silver Creek, toads were heard calling at low densities for eight days of their 14 day calling period (May 10 to May 23). Elevated frequency occurred for a period of three days when the American toad called > 1 survey per day (May 13, 14 and 23). Peak frequency occurred May 14 with a detection rate of 58%, while intensity (call level 3) peaked May 23. Consistent with Crouch and Paton (2002), the duration of the toad’s calling period was sporadic making it difficult to detect. Since there were some technical difficulties with instrumentation between May 25 and June 17, toads might have continued calling at Silver Creek without detection.
Figure 4.6. Hourly call level graph depicting calling patterns of American toads at Warwick.

At Warwick, American toads called with the greatest frequency and intensity from 21h30 until 4h30 (Fig. 4.6), with a peak frequency at 0h30 and peak intensity from 23h30 to 0h30. In May, calls peaked at 22h30 with a detection rate of 48% (Fig. 4.7). In June, calls peaked again at 0h30, with a detection rate of 50%. At Silver Creek, the highest call frequency occurred at 21h30 (43%), while intensity peaked (call level 3) from 3h30 to 4h30. In May, American toads had a monthly average nocturnal detection rate (MANDR) of 36% and 20% at Warwick and Silver Creek, respectively (Table 4.4).

Figure 4.7. Hourly detection rate of American toads at Warwick in May.
Table 4.4. MANDR (Monthly average nocturnal detection rate) from 21h30 to 4h30 of species.

<table>
<thead>
<tr>
<th></th>
<th>AMTO -W</th>
<th>AMTO -S</th>
<th>BULL -W</th>
<th>GRFR -W</th>
<th>GRFR -S</th>
<th>GRTR -W</th>
<th>GRTR -S</th>
<th>NLFR -W</th>
<th>NLFR -S</th>
<th>SPPE -W</th>
<th>SPPE -S</th>
</tr>
</thead>
<tbody>
<tr>
<td>April</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>44%</td>
<td>0%</td>
<td>65%</td>
<td>94%</td>
<td></td>
</tr>
<tr>
<td>May</td>
<td>36%</td>
<td>20%</td>
<td>0%</td>
<td>24%</td>
<td>18%</td>
<td>29%</td>
<td>72%</td>
<td>&lt;50%</td>
<td>86%</td>
<td>98%</td>
<td></td>
</tr>
<tr>
<td>June</td>
<td>12.5%</td>
<td>0%</td>
<td>65%</td>
<td>91%</td>
<td>94%</td>
<td>22%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>65%</td>
<td></td>
</tr>
<tr>
<td>July</td>
<td>0%</td>
<td>0%</td>
<td>93%</td>
<td>93%</td>
<td>88%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td></td>
</tr>
</tbody>
</table>

4.4.2 Grey Treefrog

The grey treefrog was heard calling at Warwick from May 19 to June 26 for a period of 39 days (Table 4.3). Frogs called sporadically for 27 days of this period and the highest frequencies were heard from May 28 to June 1 and June 5 to June 9. Call intensity (call level 3) peaked May 31 and call frequency peaked June 7 and 9, with a detection rate of 33%. The MANDR in May and June were 29% and 22%, respectively (Table 4.4).

Grey treefrogs called exclusively from 19h30 to 4h30; with the exception of one call which was detected at 17h30 on June 7 (Fig. 4.8). Detection of the grey treefrog was highest before midnight with an average detection rate of 49% in May and June (Table 4.5). The peak intensity and detection rate occurred at 22h30 (59%).
Figure 4.8. Hourly call level graph depicting calling pattern of grey treefrogs at Warwick.

At Silver Creek, the grey treefrog was heard calling 13 times between May 11 and June 28. Missing audio was consistent with the period of greatest call intensity at Warwick, which occurred from May 28 to June 9. A greater call frequency and intensity might have been detected at Silver Creek if audio had been available during that time.
Table 4.5. Species detection rates before and after midnight.

<table>
<thead>
<tr>
<th>April</th>
<th>AMTO-W</th>
<th>AMTO-S</th>
<th>BULL-W</th>
<th>GRFR-W</th>
<th>GRFR-S</th>
<th>GRTR-W</th>
<th>NLFR-W</th>
<th>NLFR-S</th>
<th>SPPE-W</th>
<th>SPPE-S</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before Midnight</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>39</td>
<td>73</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>After Midnight</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>49</td>
<td>65</td>
<td>95</td>
<td></td>
</tr>
</tbody>
</table>

May

| Before Midnight | 42     | 31     | 36     | 24     | 49     | 71     | 45     | 87     | 96     |
| After Midnight  | 40     | 21     | 45     | 27     | 34     | 81     | 71     | 92     | 98     |

June

| Before Midnight | 13     | 71     | 94     | 100    | 49     |        |        | 75     |
| After Midnight  | 18     | 79     | 98     | 93     | 20     |        |        | 65     |

July

| Before Midnight |        |        | 86     | 86     | 78     |        |        |        |
| After Midnight  |        |        | 100    | 100    | 93     |        |        |        |

4.4.3 Spring Peeper

At Warwick, spring peepers began calling April 9 and were last heard June 26. The duration of
the calling period was 79 days; however, frogs were only heard calling for 52 days (Table 4.3).
The peak frequency of call level 3 occurred May 14. At Silver Creek, spring peepers began
calling April 9 and continued until June 28. Following a short spike in activity, frogs experienced
a period of reduced calling intensity from April 17 to 22. The highest detection rate of 92%
occurred April 27 and the maximum intensity (call level 3) was detected April 23. Intensity and
consistency of their calling pattern increased after April 23 at both sites and they continued
calling at high frequencies and intensities for the first 3 weeks of May.
Hourly detection of spring peepers was high and they called vigorously during all hours of the night (Fig. 4.12). The MANDR of spring peepers peaked in May with at least a 90% detection at both sites (Table 4.4). Detection peaked before midnight in April and after midnight in May at both sites.

### 4.4.4 Bullfrog

At Warwick, bullfrogs called between May 23 and July 7 (Table 4.3). A bullfrog was heard calling once on May 23 and was silent for eight days following the initial call. On June 1, bullfrogs resumed and continued calling consistently until July 7 (Fig. 4.10). The calling pattern of the bullfrog illustrates a tri-modal distribution represented by three distinct phases of elevated call activity over a 37 day period. The first occurred from June 3 to June 12, the second occurred June 14 to June 23 and the third occurred June 30 to July 7, with a maximum detection rate of 67% on June 5 and July 3. It was also during these periods that the greatest call levels were
detected. Call level 2 was heard frequently throughout the peak calling period but a call level of 3 was only detected on 3 separate days: June 17, 18 and 20.

Figure 4.10. Daily call level graph depicting calling patterns of bullfrogs at Warwick.

Hourly detection rate of 89% occurred at 2h30, and call intensity peaked at 23h30 and 1h30. Bullfrogs were most prevalent after midnight and MANDR for June and July was 65%, 93% respectively (Table 4.4).

A bullfrog was also detected at Silver Creek at such low frequencies that no trends could be discerned from these data. Over the past 10 years, this species had never been detected using MCS and due to its random low frequency calls during recordings its presence is believed to be an anomaly.
4.4.5 Green Frog

At Warwick, green frogs called for a period of 50 days from May 19 until July 7. Similar to bullfrogs, green frogs had a tri-modal calling distribution. The first period occurred June 5 to June 12, the second occurred June 14 to June 24 and the third occurred June 30 to July 7. A detection rate of 100% occurred June 15, 21 and July 5. The MANDR of green frogs at Warwick in May was 24%; meanwhile, detection increased to 91% and 93% in June and July, respectively (Table 4.4).

![Daily call level graph depicting calling patterns of green frogs at Warwick.](image)

**Figure 4.11.** Daily call level graph depicting calling patterns of green frogs at Warwick.

Frogs were heard consistently throughout the day with call intensities rarely exceeding level 1. Call intensity increased after 22h30 while the highest detection rate (90%) occurred at 2h30 and intensity (call level 3) peaked at 3h30. Call frequency peaked at 2h30 in May (62%), while in June, frogs were heard calling with a detection rate of 94% before midnight and 98% after midnight (Table 4.5). In July, the detection rate of green frogs was 86% and 100% before and after midnight, respectively.
At Silver Creek, green frogs began calling May 5 and continued until the recording ended July 6. Peak frequency of call level 2 and 3 occurred June 18. The MANDR in May was 18%; however, detection increased to 94% and 88% in June and July, respectively (Table 4.4). Green frogs called consistently throughout the day and night with a peak detection rate of 59% at 3h30 and peak intensity at 0h30. The detection rate of green frogs before midnight was 24% and 100%, in May and June, respectively. The consistent low frequency calling pattern of the green frog at Silver Creek differed significantly from other species in this study.

4.4.6 Northern Leopard Frog

The northern leopard frog called from April 14 to May 29 at Warwick. Frogs called for a total of 35 days during the 46 day calling period (Table 4.3) and the greatest call frequency was heard May 2 with a detection rate of 83%. Call level 3 was detected on three separate occasions; May 7, 9 and 20. The MANDR of northern leopard frogs at Warwick in April and May was 46% and 78%, respectively (Table 4.4). Frequency and intensity peaked from 21h30 to 4h30, with an overall maximum detection of 72% from 0h30 to 2h30. The maximum frequency in May also occurred at 0h30 with a detection rate of 86% (Fig 4.11), while the maximum frequency in April occurred from 1h30 to 3h30 with a detection rate of 53%.
At Silver Creek, northern leopard frogs were heard calling from April 30 to May 25, for a total of 26 days. Frogs called consistently from May 7 to May 16, with a maximum detection rate of 38% on May 8. A peak call level of 2 was detected from May 10 to May 15 and the highest frequency occurred May 14. Detection of the northern leopard frog at Silver Creek occurred in May with a MANDR of < 50% (Table 4.4). Frogs were found to call most prominently after midnight (Table 4.5). Frogs called with the greatest frequency at 0h30 and 1h30, with a detection rate of 76%, while the peak intensity occurred at 1h30.

4.4.7 Pickerel Frog

The pickerel frog was detected at Silver Creek from May 23 to June 24 (Table 4.3). Little is known about the call frequency and intensity of the pickerel frog as it was only detected seven times throughout the duration of the study. The peak call period likely occurred between May 25 and June 17, when audio was unavailable.

**Figure 4.12.** Hourly detection rate of northern leopard frogs at Warwick in May.
4.5 Survey Duration

The time to first species detection was analysed to identify the optimal survey duration for each species based on month. The detection rates during each 3 min, 5 min and 10 min segment varied depending on species and time of year.

4.5.1 General Trends

General calling trends were identified by querying the call database. A total of 1299 surveys were returned for Warwick and 1223 surveys were returned for Silver Creek. Survey duration had a greater influence at Warwick than at Silver Creek. There were 22.7% more species detected when the survey duration was extended from 3 min to 10 min (Table 4.6). The smallest improvement occurred from 3 min to 5 min at Silver Creek with a 6.7% increase in species detection.

Table 4.6. Average increase in species detection with sampling effort.

<table>
<thead>
<tr>
<th></th>
<th>5min/3min</th>
<th>10min/3min</th>
<th>10min/5min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Warwick</td>
<td>9.3%</td>
<td>22.7%</td>
<td>12.3%</td>
</tr>
<tr>
<td>Silver Creek</td>
<td>6.7%</td>
<td>15.6%</td>
<td>8.3%</td>
</tr>
</tbody>
</table>

4.5.2 April

In April, 2 species, namely, spring peeper and northern leopard frog were detected at Warwick (Fig. 4.13). Spring peepers were heard throughout April at Silver Creek, while northern leopard frogs were only detected April 30, as a result they were not included in Fig. 4.13. At Warwick, spring peepers were heard calling actively after 20h30, with 95% of first species detections occurring within the first 3 min. Northern leopard frogs were also heard in April at Warwick and
little time was required to detect this species, with 96% of first detections occurring within the first 3 min.

![Bar chart depicting detection rates of species over time.](chart)

**Figure 4.13.** Time to first species detection in April.

### 4.5.3 May

The highest species richness was detected in May. Five species were detected at Warwick including, American toads, green frogs, grey treefrogs, northern leopard frogs and spring peepers (Fig. 4.14). Six species were detected at Silver Creek including, American toads, green frogs, grey treefrogs, northern leopard frogs, pickerel frogs and spring peepers. Due to technical difficulties, the last week of May was not recorded; therefore, the grey treefrog was only detected on two occasions at 18h30 and 19h30.
General monthly trends for May demonstrate that species requiring the greatest amount of time to first detection were green frogs and pickerel frogs with < 75% of detections occurring within the first 3 min. Spring peepers and northern leopard frogs required the least amount of time to first detection with at least 93% of detections occurring within the first 3 min. The northern leopard frog was less conspicuous at Silver Creek than at Warwick, with 80% of detections occurring within the first 3 min. Grey treefrogs were the most difficult to detect, with 75% of detections occurring within the first 3 min. By increasing the survey duration to 5 min at Silver Creek, detection of American toads, green frogs and pickerel frogs increased by 10% or more. Increasing survey duration to 10 min increased detection of grey treefrog, green frog and pickerel frog by 20% or more.
4.5.4 June

In June, five species were detected at Warwick including, American toads, bullfrogs, green frogs, grey treefrogs and spring peepers (Fig. 4.15). At the Silver Creek, four species were detected including green frogs, grey treefrogs, pickerel frogs and spring peepers. The American toad was not detected at Silver Creek in June; however, the first 17 days of recordings were lost due to technical difficulties.

![Detection Rate Graph]

**Figure 4.15.** Time to first species detection in June.

At Warwick grey treefrogs, American toads and spring peepers required the longest amount of time to first detection. Detection of the American toad decreased in June, with only 66% of detections occurring within the first 3 min. Green frogs and spring peepers at Silver Creek required the least amount of time to first detection, with at least 94% occurring within the first 3 min. Grey treefrogs had lower detection rates with 75% of first detections occurring within the first 3 min. By increasing survey duration to 5 min, the detection of American toads, grey treefrogs and spring peepers increased by at least 25%. Increasing the survey duration to 10 min increased the detection rate of the grey treefrog and pickerel frog by 50%.
4.5.5 July

Green frogs and bullfrogs were the only species detected in July (Fig. 4.16). Both species called consistently throughout the night. There was no increase in detectability with an increase in survey duration to 5 min; however, a slight increase of 2% occurred when survey duration was extended to 10 min.

Figure 4.16. Time to first species detection in July.

4.5.6 Hourly Trends

An analysis of hourly variation in monthly calling patterns was conducted by hour and by time of day (before midnight and after midnight). It was found that trends of survey duration varied greatly by hour depending on the calling patterns of individual species.
In April, species richness peaked at 0h30 to 1h30 within the first 3 min (Table 4.7). It took the longest amount of time to detect species richness during the early evening (21h30) and later morning hours (3h30-4h30). During the month of May, species richness peaked at 0h30 within the first 3 min. At 21h30 and 22h30 the longest amount of time was required to detect species richness. In June, the hours of 0h30 and 3h30 required the least amount of time to detect species richness while it took the longest amount at 21h30. During the month of July, the period of time between 0h30 and 4h30 required the least amount of time to detect species richness, while surveys occurring at 21h30 to 22h30 took the longest amount of time.

Table 4.7. Hours experiencing maximum and minimum species detection within the first 3 min.

<table>
<thead>
<tr>
<th></th>
<th>April</th>
<th>May</th>
<th>June</th>
<th>July</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Max</td>
<td>Min</td>
<td>Max</td>
<td>Min</td>
</tr>
<tr>
<td>AMTO-W</td>
<td>-</td>
<td>-</td>
<td>21h30, 23h30</td>
<td>22h30</td>
</tr>
<tr>
<td>AMTO-S</td>
<td>-</td>
<td>-</td>
<td>22h30, 0h30, 3h30, 4h30</td>
<td>1h30</td>
</tr>
<tr>
<td>BULL-W</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>GRFR-W</td>
<td>-</td>
<td>-</td>
<td>0h30</td>
<td>21h30</td>
</tr>
<tr>
<td>GRFR-S</td>
<td>-</td>
<td>-</td>
<td>0h30-1h30</td>
<td>21h30</td>
</tr>
<tr>
<td>GRTR-W</td>
<td>-</td>
<td>-</td>
<td>21h30, 23h30</td>
<td>22h30</td>
</tr>
<tr>
<td>NLFR-W</td>
<td>0h30-4h30</td>
<td>21h30</td>
<td>2h30-4h30</td>
<td>0h30</td>
</tr>
<tr>
<td>NLFR-S</td>
<td>-</td>
<td>-</td>
<td>0h30</td>
<td>23h30</td>
</tr>
<tr>
<td>SPPE-W</td>
<td>22h30, 1h30, 3h30, 4h30</td>
<td>2h30</td>
<td>3h30</td>
<td>-</td>
</tr>
<tr>
<td>SPPE-S</td>
<td>21h30-23h30</td>
<td>3h30-4h30</td>
<td>21h30-4h30</td>
<td>N/A</td>
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</table>
Table 4.8. Variation in species detection with survey duration before midnight (21h30 to 23h30) and after midnight (0h30 to 4h30).

<table>
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<th>AMTO-W</th>
<th>AMTO-S</th>
<th>BULL-W</th>
<th>GRFR-W</th>
<th>GRFR-S</th>
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Table 4.8 summarizes improvement in detection rates with time and survey duration. Detection and intensity of spring peepers is higher than most other species thus negligible improvement in species detection (0-5%) can be made with increased sampling effort. Green frogs have high detection rates during their peak calling period in June and July (Table 4.4) thus little improvement in survey detection can be made. The increase in detection with time for the American toad is moderate during peak breeding season in May and the greatest levels of improvement (17%-30%) occur at Silver Creek where populations are less prominent. Bullfrogs have high detection rates after midnight during peak breeding season, in June and July; therefore, improvement is low (0-12%). If surveys are conducted before midnight in June, bullfrog detection can increase by 33% when extending survey duration to 10 min. Grey treefrogs experience the greatest level of improvement with increase sampling effort. During seasonal and hourly peaks (before midnight), the detection of grey treefrogs increased by 36% and 56% using a 10 min survey. Northern leopard frogs experience a low to moderate increases in detection with time. Improvement in detection levels was minimal at Warwick (5%-11%), while at Silver Creek, where populations are less prominent, detection increase ranged from 34% and 21% before and after midnight, respectively.

Some species such as the spring peeper, green frog and bullfrog experienced a low level of improved detection with sampling effort during seasonal and hourly peaks. When surveys were conducted outside the peak calling period, 10 min survey duration could greatly increase the effectiveness of observer effort (117% for green frog). American toads, grey treefrogs and northern leopard frogs experienced the greatest improvement in detection with increased sampling effort.
Chapter 5

5.0 Discussion

The standard MCS protocol under-estimates species richness on an annual basis and presumably influences interpretation of long-term trends. Hourly and seasonal variability and inconsistent detection rates during peak breeding season, indicate that detection probabilities are less than perfect during the breeding season. A minimum of nine site visits occurring weekly are recommended in May and June to increase species detectability. The optimal time of day for detecting the greatest species richness occurs at 23h30; however, if multiple sites are sampled in one evening, routes should be alternated throughout the month of June to allow for equal detection of species.

Specific calling trends could not be explained temporally and evidence leads us to believe that climatic variability affected anuran calling activity in this study. Future studies should employ an intensive recording schedule while documenting environmental variables such as air and water temperature, rainfall, barometric pressure, and relative humidity as these factors are likely to provide important environmental cues for anuran breeding activity. Until effects of these variables can be established, more intensive MCS protocols should be utilized. The importance of increasing survey accuracy becomes more acute when MCS methods are employed by environmental consulting firms, as the data collected by these firms provides the basis for land management and conservation decisions. In such cases where site specific studies are being conducted, an ARS may be employed to increase survey frequency with minimal cost.
5.1 Manual Call Surveys

Spring peepers and green frogs had the highest level of detection (90% Warwick, 100% Silver Creek) at both sites during the 10 year period, demonstrating that MMP guidelines are effective at detecting these two species (Fig. 4.1 and 4.2). In 2011, a species detection rate of 43% (3 of 7 species) at Silver Creek indicates that species detections are inconsistent when employing standard MCS guidelines.

American toads were the most difficult species to detect at both sites using standard MCS guidelines. At Silver Creek, toads had never been detected during the 10 year study period when employing MSC guidelines (including 2011); however, they were detected throughout the month of May using the Song Meter. The presence of this species may have resulted from newly migrated individuals; however, a call level of 3 leads us to believe that the toad population is established in the region. The low detection probability of American toads in MCS is not uncommon and has been flagged by other studies (Brander et al. 2007). Grey treefrogs were also difficult to detect at Silver Creek as they were detected in only two of the past 10 years. Treefrogs remained undetected using standard MMP guidelines in 2011; however, the species was detected at call level 3 during one third of the surveys in May and June, 2011 utilizing the Song Metre. Focusing on increasing the detection of these two elusive species is key to the improved protocol guidelines.

American toads (60%, 0%) and grey treefrogs (70%, 20%) had the lowest detection rates. Northern leopard frogs were detected in April and May at Silver Creek using the Song Meter, but they failed to be detected in 2011 while employing standard MCS guidelines. Frogs had a 70%
and 80% detection rate at Warwick and Silver Creek, respectively, indicating that even high detection rates can result in species non-detection during infrequent MCS monitoring surveys. Bullfrogs (70%) and pickerel frogs (60%) have similar MCS detection rates as the northern leopard frog; consequently their calling patterns were closely examined. These concerns are supported by other studies which also question the effectiveness of MCS in detecting northern leopard frogs, bullfrogs and pickerel frogs, as these species may be overlooked when using conventional MCS methods (Brander et al. 2007).

5.1.2 Metapopulation Dynamics

It is important to acknowledge that anuran populations can fluctuate significantly with varying abiotic factors (Gibbons et al. 1997). Populations can experience high turnover rates depending on the colonization and extinction rates at a given site, consequently, presence and abundance of anuran communities is highly variable as a result of metapopulation dynamics (Gill 1978, de Solla et al. 2006). Metapopulations are communities comprised of two or more subpopulations which oscillate in response to stochastic (Semlitsch 2002) and/or deterministic factors (Marsh and Trenham 2001, Skelly and Meir 1997) and populations can be characterised by high extinction and colonization rates (Gill 1978).

It is possible that non-detection over the past 10 years represents characteristics of metapopulation dynamics; however, results from the Silver Creek Song Meter may suggest otherwise. In 2011, three species, namely, the American toad, grey treefrog and northern leopard frog, were detected using the Song Metre but conventional MCS guidelines were unsuccessful at detecting these species during the same year. Corn et al. (2000) compared these two methods
using a recording schedule of 12 seconds every half hour and found two additional species using the ARS compared to surveys utilizing standard NAAMP protocol at the same site. The detection of a species indicates occupancy; however, non-detection of a species, as at Silver Creek, cannot be assumed to reflect species absence (MacKenize et al. 2002). Results of presence and abundance of anuran populations is highly dependent on sampling effort (de Solla et al. 2005); therefore, without accurate data, extinction and colonization rates cannot be reliably calculated, nor can the extent of metapopulation dynamics be assumed. Erroneous non-detection can be presumed to represent a local extinction or emigration (de Solla et al. 2006) and if assumed, will lead to an over-estimate of population fluctuations (Marsh and Trenham 2001, Peterson and Dorcas 1992). The southern leopard frog (*Rana sphenocephala*) is rarely detected during the month of June; however, its non-detection is not indicative of species absence or metapopulation dynamics because this species calls actively between midnight and sunrise and is likely to be missed utilizing standard MCS protocols (Bridges and Dorcas 2000). It is possible that the breeding populations in the Credit Valley are present year to year; however, peak call periods may be undetected due to protocol guidelines producing false negatives.

5.2 Song Meter

Results from the Song Meter were similar to those documented during MCS. Detection and call intensity of spring peepers, green frogs and bullfrogs were consistent using both methods (Table 4.1). However, the pickerel frog was undetected by the Song Meter, despite being detected during MCS which were conducted simultaneously. In this case, the range and basal frequency of the pickerel frog’s call is believed to have been the reason for its absence from instrument recordings. The call of the pickerel frog is similar to that of the northern leopard frog which does
not transmit well over long distances (de Solla et al. 2005). Since MCS are conducted approximately 100 m from the Song Meter, the instrument was not able to detect the calling frequency of the pickerel frog at this distance. This leads us to believe that the call rates determined during this study are conservative and calls might occur with greater frequency depending on distance and pitch of a given species. Distance of calling anurans at Warwick was also a factor; however, by increasing the volume on the computer calls were discernible.

The 24 days of missing data at Silver Creek has introduced bias into this data set. The calling period of the northern leopard frog and American toad began in early May and may have concluded when the audio was unavailable. The duration of their calling period might have continued into June without our knowledge. The intensity of their calls is also unknown during that time, making it difficult to determine the peak calling period of these species at Silver Creek. This missing data also prevented us from studying the calling patterns of grey treefrogs and pickerel frogs at this location. Such information could have greatly improved the accuracy and applicability of our results.

5.3 Two-Tier Approach

Program coordinators have the responsibility of developing protocol guidelines that effectively record site occupancy while trying to attract and maintain a large group of qualified observers (Shirose et al. 1997). Although the literature indicates that greater sampling effort yields higher species detectability (Crouch and Paton 2002, de Solla et al. 2005, Gibbons et al. 1997), this may not be an option at the level of a volunteer based organization as tradeoffs must be made between volunteer effort and survey accuracy. Programs must compromise accuracy for broad geographic
coverage; however, 100% detection is unnecessary as non-detection can be reduced if sample size is large and sites are surveyed consistently (Shirose et al. 1997).

Problems of detection arise when environmental consulting firms in the province of Ontario employ the same protocol prescribed by the MMP and the Amphibian Road Call Count. These firms are commissioned to study one specific site to determine species richness; however, anuran surveys are often only conducted for 1 or 2 years, using 3 to 4 site visits per year. These data are used to determine the significance and biodiversity of a site which can be used to establish the level and configuration of protection required (PPS 2005). Rare or elusive species have low detection probabilities using standard techniques, due to limited sampling effort, and have a greater probability of being missed during brief surveys (Gibbons et al. 1997). In short-term surveys where impending development depends upon the results of an EIR (Environmental Impact Report), accuracy is invaluable. In such cases, serious errors in land management decisions could be made. Therefore, a secondary protocol should be developed for short-term commercial surveys that will maximize species detection within a one or two year period. A two-tier approach is recommended as both volunteer organizations and environmental consulting firms have very different, goals (long-term versus short-term), resources (volunteer versus paid personnel), study area (regional/national versus site-specific) and agenda (general trends versus accurate species account).

5.4 Daily and Seasonal Trends

A comparison of auditory methods demonstrates that current monitoring protocols are ineffective at reliably detecting all species at a discrete location (Fig. 4.2). Results from this preliminary
study provide a general understanding of calling patterns of seven anuran species and indicate that increased sampling effort is required to produce a more accurate account of species presence.

Consistent with the literature, spring peepers and green frogs were found to have the highest detection rates because they call consistently throughout the season (Bridges and Dorcas 2000, Crouch and Paton 2002, de Solla et al. 2005, Stevens et al. 2002). Also consistent with the literature, ranids peak after midnight (Bridges and Dorcas 2000, Mohr and Dorcas 1999) while hylids (grey treefrogs) called primarily during the early evening hours (Runckle et al. 1994, Bridges and Dorcas 2000). The calling pattern of American toads was sporadic making them the most difficult to detect. This species has a low detection rate using both ARS and MCS methods and its breeding period can occur in April, May or June or any combination of the 3 months (Fig 4.1 and 4.2). This species has no significant hourly calling pattern (Table 4.2) unlike that observed for hylids and ranids. The calling patterns of bullfrogs and pickerel frogs also require special attention during protocol revision having been undetected at a rate of 30% and 40% respectively by standard MCS methods (Fig. 4.1 and 4.2). MMP guidelines have yielded high detection rates for spring peepers and green frogs; therefore, revision of current guidelines should focus on the calling patterns of American toads, grey treefrogs, northern leopard frogs, pickerel frogs and bullfrogs.
Table 5.1. Recommended frequency of site visits.

<table>
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<tr>
<th>MANDR</th>
<th>AMTO</th>
<th>BULL</th>
<th>GRFR</th>
<th>GRTR</th>
<th>NLFR</th>
<th>PIFR</th>
<th>SPPE</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>20%</td>
<td>93%</td>
<td>93%</td>
<td>29%</td>
<td>&lt;50%</td>
<td>N/A</td>
<td>86%</td>
</tr>
<tr>
<td>Month(s)</td>
<td>May, June</td>
<td>June, July</td>
<td>June, July</td>
<td>late May, June</td>
<td>early- mid May</td>
<td>late May, June</td>
<td>April, May</td>
</tr>
<tr>
<td># Visits</td>
<td>5</td>
<td>2</td>
<td>2</td>
<td>5</td>
<td>3</td>
<td>N/A</td>
<td>2</td>
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</table>

Using detailed calling data from this single year of study, general trends can be inferred. Species with the lowest MANDR are key to identifying the minimum number of site visits required for effective survey frequency; these species include American toads, grey treefrogs and northern leopard frogs (Table 4.5). With minimum detection rates of 20% and 29% respectively during peak season, American toads and grey treefrogs require a minimum of 5 site visits. A rate of < 50% during peak season indicates that a minimum of 3 site visits are required to detect northern leopard frogs. Bullfrogs, green frogs and spring peepers have high detection rates; however, with only one site visit and detection rates < 100%, they could potentially be missed during their respective call periods (Fig. 4.1). A minimum of 2 site visits can increase the probability of detecting these species as increasing the number of site visits produces more accurate occupancy rates (MacKenzie et al. 2002).

5.4.1 American Toad

The calling pattern of American toads is sporadic. In 2011, the toad called primarily in May and was also detected on several occasions throughout June at Warwick. During MCS, the toad was most commonly detected in May but was also detected in April 2006, 2008 and June 2008; therefore, its calling pattern spans all 3 site visits recommended by the standard protocol. Despite the duration of its calling period, this species remains the most difficult to detect at both
Warwick and Silver Creek locations. Low detection probabilities of <50% (Crouch and Paton 2002) are not uncommon for this species as they are difficult to detect using MCS (Brander et al. 2007).

Fig. 4.5 illustrates that even during their peak calling period, American toads do not call nightly and have a possibility of being missed if a single survey is conducted during their breeding season (Fig. 4.1 and 4.2). When modelling for site occupancy, it was found that American toads had the greatest variation between observed and modelled occupancy rates (MacKenzie et al. 2002, Weir et al. 2005). Shirose et al. (1997) found that when employing one-60 min survey, some species remained undetected during their breeding period; however, increasing the number of site visits can produce a better estimation of the occupancy rate (MacKenzie et al. 2002). A MANDR of 20% at Silver Creek (Table 4.4) indicates that toads should be surveyed a minimum of 5 times during their breeding period for detection to occur (Table 4.5). At sites with low densities such as Silver Creek, species detection can increase by 17% and 28% by utilizing a 5 min and 10 min survey, respectively, in May (Table 4.8). In June, a 10 min survey is highly recommended to increase the detection of this species.

5.4.2 Grey Treefrog
Grey treefrogs had a long and consistent calling pattern; however, there calls at Warwick and Silver Creek were sporadic during ARS recordings. The calling period of treefrogs spans the last two site visits recommended by the standard protocol. The detection rate of the grey treefrog utilizing the standard MCS guidelines is 70% at Warwick and 20% at Silver Creek and also spans both site visits (Fig. 4.1 and 4.2).
Though detection of the grey treefrog occurs during the end of May and throughout June, the MANDR remains low (29% and 22%, respectively – Table 4.4). To increase the detection of the grey treefrog in Ontario, a minimum of 5 site visits should be conducted throughout its calling period (Table 4.5). Detection of the grey treefrog primarily occurred before midnight with an average detection rate of 49% in May and June. Frogs from the genus hyla are typically found to peak in the early evening hours (Bridges and Dorcas 2000, Mohr and Dorcas 1999, Runckle et al. 1994), for this reason surveys should be conducted before midnight to increase the detection probabilities of the species.

The detection rate of grey treefrogs can also be improved by increasing survey duration. A moderate detection rate of 62% was heard in Rhode Island (Crouch and Paton 2002) using a 16 min survey. Effectiveness of surveys in May can increase by 33% and 56%, by increasing the survey duration to 5 min and 10 min, respectively (Table 4.8). Effectiveness of surveys in June can also increase by 18% and 36%, by increasing the survey duration to 5 min and 10 min, respectively. A minimum of 5 site visits of 10 min in duration are recommended to detect this species during its calling period at sites with low detection probabilities.

5.4.3 Spring Peeper

Utilizing the standard MMP protocol, detection of the spring peeper was nearly perfect at both sites (Fig. 4.1 and 4.2). MCS data indicate that spring peepers call during all three recommended sampling periods and the detection probability of this species is very high. MCS at Warwick demonstrate that spring peepers are typically detected in April; however, results from the ARS demonstrate that the peak calling period of this species occurred in May. Spring peepers were the
easiest species to detect and were found to have the highest detection rate in this and several similar studies (Crouch and Paton 2002, de Solla et al. 2005, Stevens et al. 2002). At both sites, the MANDR (86% and 98%) of this species peaked in May (Table 4.4). Other studies found that the detection of spring peepers in Ontario ranged from 89% (de Solla et al. 2005) to 95% (de Solla et al. 2006). With such high species detection rates, current protocol guidelines are adequate for detecting the presence of spring peepers.

5.4.4 Bullfrog

During ARS recordings, a bullfrog was heard on one occasion in May but actually began its calling period June 1. The detection of the bullfrog is generally limited to the last site visit recommended by the standard protocol; however, with 70% detection over the past 10 years (Fig. 4.1), it is possible this number could be improved.

Bullfrogs are known to have low detection probabilities (Crouch and Paton 2002, Saenz et al. 2006); however, during our ARS study, they were detected at a rate of 65% and 93% in June and July, respectively (Table 4.4). For optimal detection of bullfrog populations, a minimum of two site visits per season are recommended (Table 4.5). The first site visit should occur in mid-late June and the second site visit should occur early July.

Call frequency and intensity of bullfrogs peaked after 23h30. In South Carolina bullfrogs peaked between 2h00 and 6h00 from mid-June to mid-July (Bridges and Dorcas 2000, Mohr and Dorcas 1999). In other surveys conducted before midnight, bullfrogs were found to call infrequently (Crouch and Paton 2002). Surveys should be conducted after 23h30 (Bridges and
Dorcas 2000, Mohr and Dorcas 1999); however, if conducted before this time in June, detection of the bullfrog can increase by 33% if survey duration is increased to 10 min (Table 4.8).

5.4.5 Green Frog

The detection rate of green frogs was high at both sites. The green frog was heard consistently at Warwick from May 20 to July 7; however, frequency and intensity (call level 2 and 3) increased after May 31. The call frequency of the green frog was also very high at Silver Creek while recordings were available. Detection of the green frog utilizing the standard MCS protocol occurred in May and June and was nearly perfect.

Detection of green frogs using the MMP guidelines has been consistent and is supported by results from the ARS survey. There is little concern regarding the detectability of this species. A study conducted in Rhode Island found that green frogs had one of the highest occupancy rates (88%), second to spring peepers (Crouch and Paton 2002) and in South Carolina green frogs called nightly from mid-June to mid-July (Bridges and Dorcas 2000). One site visit per month in May and June has sufficed over the past 10 years and should continue to yield accurate results. Due to low detection rates (Table 4.4) in May (18% and 24%) and much higher detection rates in June (91% and 94%) and July (93% and 88%), detection of green frogs can be optimized by focusing on latter months. To increase detection of population densities, it is recommended that surveys be conducted after 22h30; this calling pattern is consistent with other studies which found that green frogs peaked after midnight (Bridges and Dorcas 2000, Mohr and Dorcas 1999, Weir et al. 2005).
5.4.6 Northern Leopard Frog

Using the current protocol detection of northern leopard frogs occurs at a relatively high frequency at both Warwick (70%) and Silver Creek (80%); however, results from the 2011 ARS study reveal a need for improvement (Fig 4.1 and 4.2). All detections at Warwick and the majority of detections at Silver Creek have occurred in April. Song Meter results indicate that the detection of northern leopard frogs occurred periodically in April and intensity peaked during the second and third week of May.

This species was identified as one of the most difficult to detect using MCS (de Solla et al. 2005). The MANDR of the northern leopard frog in April (46% and 0%) and May (78% and <50%) differed significantly (Table 4.4). To improve detection of this species, a minimum of three site visits should be conducted at the beginning of May (Table 4.5). Detection of the frog peaked at both sites after midnight. Since the peak calling period for ranids typically occurs between midnight and sunrise (Bridges and Dorcas 2000), the optimal time to detect this species should occur at this time. If surveys are conducted before midnight, survey effectiveness can increase by 34%, by extending the survey duration to 10 min (Table 4.8).

5.4.7 Pickerel Frog

Little can be surmised from the data collected for the pickerel frog. This species calls in May at a detection rate of 60% (Fig. 4.2); however, it is difficult to detect using MCS (Brander et al. 2007, Crouch and Paton 2002, de Solla et al. 2005). Detection using MMP guidelines has been limited to May; however, utilizing the Song Meter, we are now aware that the pickerel frog calls until
the end of June. From these data, the breeding period of this species can be identified as occurring from the end of May and throughout the first 3 weeks of June.

5.5 Recommendations

The period of time with highest species diversity occurred from April 30 (Day 25) to June 28 (Day 85) (Fig 4.3). A summary of calling patterns in Table 5.1 indicates that some species reach their peak calling period in early May such as northern leopard frogs, while others such as green frogs and bullfrogs peak in June and July. The grey treefrog; however, peaks later in May and throughout the month of June. Spring peepers call consistently through May while American toads display sporadic calling patterns throughout the breeding season (Fig 4.1). Surveys should be conducted during the months of May and June as the greatest species richness (Fig 4.9 and 4.10) and calling peaks occur during this period.

Given the low detection rate of species such as American toads (20%), grey treefrogs (29%) and northern leopard frogs (50%) (Table 4.4) which peak during this period, a more intensive monitoring schedule is recommended. It is estimated that a minimum of 3 site visits (Table 4.5) are required for the detection of northern leopard frogs and these should occur early-mid May (Table 4.3). A minimum of 5 site visits (Table 4.5) are recommended for the detection of the grey treefrog at sites with the lowest densities and these visits should occur late May through the month of June (Table 4.3). American toads have sporadic calling patterns; however, they regularly call in May (Fig 4.1 and 4.5); therefore, the 5 recommended site visits should occur during this time.
Based on species-specific trends, the site visit recommended at the end of April does not maximize observer effort. Spring peepers and northern leopard frogs were the only species detected during this time period. Spring peepers call consistently throughout the season and northern leopard frogs peak at the beginning of May; therefore, the April visit is unnecessary to detect these species. One species that does influence the commencement of the monitoring season is the wood frog which was not detected during this study. Wood frogs are known to call in April; however, these prolific breeders can peak at call level 3 as early as March 16 (pers. observ.); consequently, surveys occurring at the end of April are likely to miss this species (de Solla et al. 2006). Additional studies would need to be conducted to identify the calling patterns of this species; however, for the purpose of early callers such as spring peepers, a site visit during the month of April is unnecessary.

### Table 5.2. Recommended survey frequency and duration.

<table>
<thead>
<tr>
<th></th>
<th>AMTO</th>
<th>BULL</th>
<th>GRFR</th>
<th>GRTR</th>
<th>NLFR</th>
<th>PIFR</th>
<th>SPPE</th>
<th>Recommended</th>
</tr>
</thead>
<tbody>
<tr>
<td># Visits</td>
<td>5</td>
<td>2</td>
<td>2</td>
<td>5</td>
<td>3</td>
<td>N/A</td>
<td>2</td>
<td>9</td>
</tr>
<tr>
<td>Monthly Peak</td>
<td>May, June</td>
<td>June, July</td>
<td>June, July</td>
<td>late May, June</td>
<td>early-mid May</td>
<td>late May, June</td>
<td>April, May</td>
<td>May, June</td>
</tr>
<tr>
<td>Peak Time</td>
<td>21h30-4h30</td>
<td>after 23h30</td>
<td>after 22h30</td>
<td>before midnight</td>
<td>after midnight</td>
<td>N/A</td>
<td>21h30-4h30</td>
<td>alternating</td>
</tr>
<tr>
<td>Duration</td>
<td>10 min</td>
<td>3 min</td>
<td>3 min</td>
<td>10 min</td>
<td>3 min</td>
<td>N/A</td>
<td>3 min</td>
<td>10 min</td>
</tr>
</tbody>
</table>

If surveys were to be more intensively conducted between May 1 and June 30, all species reviewed in this study could be detected with greater confidence. This period of time is a crucial for anuran detection, as all seven species analyzed in this study peak within this time period. Though all species were detected between May 10 (Day 35) and May 30 (Day 55) (Fig. 4.3), the
The optimal daily time period to detect the maximum species richness with the greatest intensity occurs between 21h30 and 4h30 (Table 4.2). However, Fig 4.4 illustrates that the frequency of peak activity is nearly 40% higher between 22h30 and 3h30 as most species peak later in the evening (Table 4.3). During the month of May, spring peepers, northern leopard frogs (Fig. 4.11) and green frogs experience peak detection between the hours of 0h30 and 2h30. It is also between these hours that the greatest number of species can be detected in the least amount time (Table 4.7); therefore, the peak sampling period in May occurs within these times. In June, green frogs call consistently after 22h30; however, optimal detection of species richness during the month of June is difficult to identify due to opposing peaks of grey treefrogs and bullfrogs. The detection rate of the bullfrog peaks between 23h30 and 2h30 while the grey treefrog peaks
between 21h30 and 23h30; therefore, if a single site is to be sampled, the optimal time for detecting the greatest species diversity and intensity is 23h30. Surveys conducted earlier or later than this are likely to miss bullfrogs or grey treefrogs, respectively.

The grey treefrog can be heard at call level 3 until 3h30; however, the frequency and intensity of its calling pattern drops after midnight. Ranids consistently peak after midnight; however, bullfrogs particularly call at low intensities during the early evening and should be sampled later in the night. If routes are utilized to sample numerous sites within one evening, the sequence of stops should be altered throughout the month of June to increase the probability of detecting both species at their peak.

Optimal survey duration depends on the level of detection being sought by the researcher. Table 4.8 outlines the increase in detection with sampling effort for each species during a given month. Surveys of 10 min in duration occurring in May significantly increase the detection rates of northern leopard frogs and American toads before and after midnight at sites with the lowest densities. A 10 min survey is also highly recommended for June to improve the detection rate of grey treefrogs. A 3 min survey is sufficient to detect bullfrogs in July; however, if surveys are conducted before midnight in June, a 10 min survey can increase detection by 33% (Table 4.8). Therefore, a 10 min survey is recommended to increase species detection throughout May and June at sites with the lowest detection rates and when sampling species outside their peak call periods.
These trends are based on a single year of study and provide preliminary data for the development of a calling chronology for these seven species. Annual variations in calling patterns are likely to occur due to environmental conditions which act as cues for calling activity (Brander et al. 2007, Mossman et al. 1998, Saenz et al. 2006, Shirose et al. 1997, Tupper et al. 2007, Weir et al. 2005). Continued intensive seasonal studies utilizing ARS are recommended to gain a clearer understanding of anuran calling patterns.

5.6 Beyond Temporal Analysis

Recognizing the temporal calling patterns of all anuran species is essential; notwithstanding, certain patterns cannot be predicted by temporal analysis alone. Each of the species studied demonstrates patterns of elevated and reduced activity within their respective calling periods. Data in this study have been used to identify optimal time for detecting species during their calling periods; however, variability within the calling period cannot be explained by temporal trends alone. The tri-modal calling pattern of the bullfrog (June 3-12, June 14-24 and June 30 to July 7) and green frog (June 5-12, June 14-24 and June 30 to July 7) may be the most prominent. The patterns occurred simultaneously, indicating that distinct factors are affecting both species in a similar manner. The sporadic calling activity of American toads and grey treefrogs exhibit very little temporal regularity and the randomness of their calling patterns is likely affected by other factors. A one week interruption in calling activity of the northern leopard frog at the end of April also coincides with the reduced frequency and intensity of spring peepers during this time period.
It is known that breeding activity is strongly influenced by abiotic factors (Saenz et al. 2006) and can be interrupted if conditions are unfavorable (Shirose et al. 1997). Due to their body structure, ectotherms, such as anurans, are more acutely affected by environmental variations than endotherms, such as mammals (Pough et al. 1989) as they rely on external conditions to regulate their body temperature (Purves et al. 2000). As a result, anuran body temperature is an important determinant of calling activity (Tupper et al. 2007). As such anuran calling patterns were found to be closely correlated with temperature (Mossman et al. 1998, Weir et al. 2005). In Massachusetts, calling patterns of fowler’s toad are strongly correlated with water temperature which is coincidentally correlated with body temperature (Tupper et al. 2007). In response to annual variation in temperature, the peak call period of the fowler’s toad varied annually and peaked at different times of the season.

Though spring peepers call at a wide range of temperatures between 5°C to 22°C (Saenz et al. 2006), green frogs and grey treefrogs are highly correlated with temperature (Brander et al. 2007). Shirose et al. (1997) experienced similar results when hot dry conditions in June interrupted calling activity of the grey treefrog, while green frog breeding activity continued unimpeded. Saenz et al. (2006) also found the breeding activity of green frogs and bullfrogs to be associated with warm temperatures, while pickerel frogs were correlated with intermediate temperatures between 10° to 22°C. Rainfall can also affect anuran calling patterns. Calling strategies of green frogs were found to correlate with drought conditions (Mossman et al. 1998), while grey treefrogs are correlated with periods of rainfall (Bishop et al. 1997). The calling frequencies of these frogs were also positively correlated with a temporal lag following rainfall.
(Saenz et al. 2006). Factors such as barometric pressure, relative humidity, wind speed light and intensity were also found to influence anuran calling patterns (Steelman and Dorcas 2010).

As such calling patterns of these species cannot be explained by temporal analysis alone. To determine the source of the non-temporal variance, calling trends were generally compared to weather data from a nearby weather station at Lester B. Pearson International Airport (Environment Canada 2012). The calling periods of American toads were associated with periods of increased temperature and precipitation and days of peak frequency and intensity corresponded with peak temperature and/or precipitation. Calling trends for the grey treefrog were associated with increasing temperatures and calling periods ended when temperatures reached their peak and treefrogs were not heard as temperatures began to decrease. The beginning of the spring peeper’s calling period commenced as temperatures began to rise and the initial calling spike ended when the temperature dipped. The calling period slowed during the cooler period and recommenced when the temperature peaked once again. The tri-modal calling pattern of the bullfrog and green frog did not follow any specific temperature or precipitation pattern; however, June 13, a day of noted reduced activity for both species, was associated with very low temperatures. The initial spike of the northern leopard frog was not associated with any specific temperature or precipitation pattern; however, the second spike in calling activity of both northern leopard frogs and spring peepers on April 23 coincided with a peak in temperature and precipitation occurring on the same day. May 19 and May 23 were also periods of significant calling activity when several new species were heard calling for the first time. On May 19, green frogs and grey treefrogs began calling at Warwick; this was a day of no precipitation which occurred during a temperature increase and was preceded by a week of wet weather. A bullfrog
and pickerel frog were heard calling for the first time May 23 when temperature peaked and a precipitation event occurred. These associations provide a general understanding of species response to climatic variables; however, to infer specific calling trends, detailed localized data is required. As such these trends lead us to believe that further investigation into the effects of temperature and precipitation on calling activity is necessary.

5.7 Future Studies

The trends presented here provide base line data for future studies. To optimize survey frequency, further investigation into climatic variables is necessary. Anuran calling strategies should be recorded annually to document variations in occupancy and abundance utilizing a rigorous recording schedule while documenting potential abiotic influences. Calling activity is controlled by variations in abiotic conditions, therefore, future studies should include documentation of environmental variables such as air and water temperature as well as rainfall. Since the tri-modal distribution of bullfrogs and green frogs did not correlate with temperature and precipitation patterns, barometric pressure, light intensity and relative humidity should also be documented as these factors are likely to provide important environmental cues for anuran breeding activity. Factors such as increased wind speed and traffic noise have also been associated with increased detection of grey treefrogs, pickerel frogs and south western chorus frogs (*P. feriarum*) but were discounted as spurious effects (Weir et al. 2005). During Song Meter recordings, American toads were often detected when a jet plane was heard passing overhead. These events were also discounted as an anomaly; however, due to the regularity of their occurrences, further investigation into such phenomena is required.
The environmental and anthropogenic influences on anuran call activity are complex. MCS should be designed to target species found within a given region. To improve the applicability of future studies and the effectiveness of our recommendations, a greater number of sites should be studied to include all species within this region. A species such as the wood frog is key to defining the seasonal and abiotic conditions which cue their calling period hence commencement of the annual anuran breeding season. Defining their calling strategies would improve recommendations for protocol revision. Gaining a comprehensive understanding of the calling phenology for all species within this region can provide us with unprecedented detail to formulate an effective protocol based on detailed yet holistic patterns of anuran calling strategies.

5.8 Management Implications

This thesis emphasizes that the complexity of anuran calling strategies can significantly affect species detectability. Cues for the commencement of a species-specific breeding season and general calling activity within the breeding season are influenced by a variety of factors including, air and water temperature, rainfall, humidity, etc. Many aspects converge to provoke anuran call activity and these factors may be difficult to measure experimentally. The impact of short-term climatic variability and local disturbances can be mitigated by employing effective surveys established upon species-specific detection rates.

Until the effects of climatic variability can be established, a greater number of site visits should be recommended by protocol guidelines to account for variations in species detectability. The nine weekly site visits recommended in this thesis provide a rudimentary estimate of the minimum number of surveys required to detect species richness within a given year, while a
greater number of site visits is likely to yield more accurate results. When intensive short-term surveys are conducted to evaluate the species richness of an area, standard MCS protocols should not be used. These protocols are not rigorous enough to portray anuran biodiversity given the dynamic patterns of their life cycle over a one or two year period. This study demonstrated that, even when monitoring is conducted over 10 year period following the standard protocol, some species could potentially be missed.

Trends identified in this thesis should be utilized to produce effective standards for intensive short-term surveys such as those employed by environmental consulting firms. Well established long-term surveys could also benefit from these results as species richness may be unable to acquire full species detection when utilizing MCS given the low species detection at Silver Creek when employing MCS.

5.9 Automated Recording Systems

Automated recording systems can be very useful instruments for constructing detailed acoustic patterns. In their summary of MCS methods, Dorcas et al. (2009) recommend that MCS be used for large scale regional surveys, monitoring multiple anuran populations, over an extended period of time. Conversely, they recommended that an ARS be used to monitor discrete populations at one or a few sites of interest. An automated recording system can provide a substantial amount of call data, and depending on the frequency of recordings, has a better chance of detecting rare species (Dorcas et al. 2009). Some studies have found this method to be expensive and time consuming (Corn et al. 2000); however, they used cassette tapes with relatively limited storage capabilities which had to be changed every few days. With advances in
technology, current systems record calls using a digital format (.wav or mp3) and depending on storage capacity, recorders can remain in the field for an entire season and no retrieval of data is necessary. With the use of new technologies, a researcher’s time can focus on listening to recorded calls rather than on the retrieval of field data.

If the goal of the study is to document species occupancy at a given site, the use of an ARS would reduce the number of site visits required and permit a greater sampling frequency. However, if the goal is to document specific abundance numbers, digital recordings make it difficult to decipher individuals and MCS might be more appropriate in this case. Some calls such as that of the pickerel frog can be difficult to detect using an ARS so the use of a recording system should be accompanied by occasional site visits. For commercial studies such as those conducted by environmental consulting firms, reducing the number of site visits is economical as it reduces cost of paid personnel as well as fuel costs for travel to and from the study site. An initial investment in a recording system could provide profitable return on investment after several seasons of use, while improving species detection with a feasible increase in survey frequency.

Reviewing recorded data can involve considerable effort. Corn et al. (2000) required 1.5 hours to review 1 hour of cassette tapes. However, the 10 min recording sample utilized in this study permitted ample time to review and document recorded data. In maximizing efficiency and accuracy, sample recordings should be long enough to permit trained personnel to document all necessary information within the time period of the recording. The 12 second sampling period utilized by Corn et al. (2000) was not sufficient to document all necessary information; however,
a longer sample recording will increase time for species detectability in the field, while allowing
the listener to precisely document all necessary information. Audio recognition software is also
available and may reduce the number of hours required to review recordings.

One of the most important concerns in the use of this method is the safety of the
equipment. Recording equipment left unattended in the field is vulnerable to theft, vandalism and
wildlife activity (Corn et al. 2000). During this study, 24 days of data were lost due to destructive
wildlife activity. Corn et al. (2000) also experienced some technical difficulties while in the field
resulting in lost data. Despite initial cost of the recording system and risk of damage to the
equipment, this method has the potential to become a widely utilized method in the field
environmental consulting or any field of research that employs acoustic methods at a discrete
location.
Chapter 6

6.0 Conclusion

This thesis illustrates the complexity of anuran breeding habits. Evidence of significant interspecific daily and seasonal variation supports our hypothesis that current protocol guidelines for MCS are ineffective at representing species richness. The current frequency and timing of site visits cannot detect all species calling during a given season and may also be unable to detect site occupancy in long-term surveys. Each species peaks at different periods of the season and different hours of the day. Inter-seasonal variation has presented a problem in detecting species during their breeding season as environmental variables may control the detectability of anurans by cueing or inhibiting calling activity. Species-specific chronologies and detection rates must be incorporated into new and revised protocols so that accurate accounts of anuran populations can be collected for years to come. Data collection using ARS techniques can supplement MCS methods by intensively monitoring sites for a short period with minimal survey effort. Such methods are highly encouraged for consulting firms on which critical and sometimes contentious land management and conservation decisions are made. This method can also be employed to clarify the calling patterns of threatened species such as the western chorus frog and the fowler’s toad.
6.1 Summary of Results

A comparison of MCS and ARS methods uncovered significant differences in species calling patterns. ARS data provided insight into the true calling patterns of anuran species in southern Ontario. With an intensive recording schedule, we were able to evaluate the effectiveness of MCS methods and create detailed calling profiles for each species. MCS methods were unable to create an accurate portrayal of species richness at Silver Creek. Two new species were undetected in 2011 and only 50% of populations were detected during the same year. Species detectability was inconsistent at both sites during the 10 years of study. American toads and grey treefrogs experienced the lowest level of detection using MCS. Pickerel frogs, bullfrogs and northern leopard frogs experienced moderate levels of detection while spring peepers and green frogs experienced the highest level of detection.

Daily and seasonal trends of anuran calling activity reflect differences in species detectability. Some species such as the American toad are difficult to detect and exhibit sporadic calling patterns while other species such as the spring peeper call consistently in great numbers throughout the duration of their breeding period. Green frogs call consistently during their breeding season but at much lower densities than spring peepers. Despite calling at lower call intensities, green frogs have high detection rates in MCS similar to that of the spring peeper. Bullfrogs also had high detection rates throughout their breeding season. Grey treefrogs called sporadically throughout the season and were difficult to detect with any consistency. Pickerel frogs were the most elusive species as their basal tone made them difficult to detect using recording instrumentation and system failure caused a loss of data during their calling period.
The high degree of variability indicates that the timing and frequency of site visits recommended by standard MCS protocol requires some amendments to improve the accuracy of species richness estimates. The use of three site visits per season under-estimates the true species richness during a given year and presumably influences the detection of rare or elusive species for long-term studies as well. Results indicate that short-term studies should require more intensive sampling frequencies to increase accuracy of findings within a given year. A minimum of nine weekly site visits are recommended in May and June and these should occur when target species are most likely to call. In May, target species such as northern leopard frogs and spring peepers experience the highest detection levels after midnight. In June, grey treefrogs peak before midnight, while bullfrogs and green frogs peak after midnight. To increase detectability of species richness surveys should occur at 23h30; however, if multiple sites are sampled in one evening, routes should be alternated throughout the month of June to provide equal detection for all species.

6.2 Management Implications

Establishing biologically relevant guidelines is critical to the foundation of anuran monitoring protocols. Identifying and applying these species-specific chronologies to current and proposed survey methods will improve survey accuracy which can provide greater insight into long-term population trends and improve our understanding of metapopulation dynamics. MCS data provide the basis for many anuran studies. A data set with improved accuracy may uncover patterns that can be used to examine phenomena at larger scales to better understand the ecological, biogeographical and climatic processes that control population dynamics. An
accurate data set can help researchers recognize the complex processes that influence the growth and distribution of anuran populations in a region and slow the accelerated rate of species decline that we are currently experiencing.

Automated recordings systems can also contribute to an improved data set by reducing disruption to the environment and calling anurans while allowing for increased survey frequency with minimal observer effort. This technique has proven to yield useful results that can advance knowledge within the field. When combined with other instruments, an ARS can identify underlying environmental determinants that control anuran behaviour. Barriers to this method include potential damage to unsupervised instrumentation as well as inability of the audio equipment to detect species calling at low basal frequencies. Despite these limitations, ARS is a monitoring technique that has the potential to become widely utilized as a method for intensively surveying an area of interest over a short period of time.

Much of the power for preservation of critical habitat in the developing regions of southern Ontario resides in the ecological data collected by environmental consulting firms (PPS 2005). An effective, efficient and accurate survey method is required to produce relevant and representative depictions of anuran populations in areas that are to be developed so that informed land management decisions can be implemented. Identification of these populations is the first step to long-term conservation. This method has the potential to become a standard monitoring practice in the environmental consulting industry.
Accurate survey methods become critically important when surveying threatened species such as the western chorus frog and fowler’s toad. Further investigation into the calling patterns of these species will allow us to make specific recommendations for optimal survey frequency. More appropriate survey guidelines could increase the detection of these species thus more effectively defining areas that require habitat protection. This would lead to improved long-term preservation of current anuran populations in Ontario.
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Appendices

Time to first detection of American Toads at Warwick
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Figure A.57. Time to first detection of spring peeper at Silver Creek in May.
Figure A.58. Time to first detection of spring peeper at Silver Creek in June.