Ecological Monitoring 2015

rare Charitable Research Reserve

Prepared by:
Tim Skuse
Jenna Quinn

Ecological Monitoring Report Executive Summary

The ***rare*** **Charitable Research Reserve** is a not-for-profit environmental organization that preserves over 900 acres of land along the Grand River in Waterloo Region, Ontario. In 2006, ***rare*** joined Environment Canada's Ecological Monitoring and Assessment Network (EMAN) to establish long-term ecological monitoring programs for the property with the objective of determining the status of ***rare's*** ecosystems and tracking how they change over time. Since 2006, several ongoing monitoring programs have been established at ***rare*** and have been carried out in each subsequent year. In 2015 ecological monitoring programs occurred for butterflies, plethodontid salamanders, forest health, and soil humus decay rates.

Butterfly Monitoring

Butterfly monitoring occurs at ***rare*** across four separate transects for four weeks during the late spring and summer. Butterfly monitoring in 2015 saw the third highest total butterfly abundance and the most species ever recorded in a monitoring season with 4931 individual observations. The most abundant butterfly species was the Cabbage White (*Pieris rapae*), followed by the European Skipper (*Thymelicus lineola*) and the Clouded Sulphur (*Colias philodice*). Cabbage White and European Skippers are both non-native butterflies, which have seen high abundances in past years. The Cabbage White has been the most abundant butterfly in every past monitoring year, but, despite being the most abundant butterfly overall, Cabbage Whites had their lowest year on record in 2015 at ***rare***. In past years Cabbage Whites have often accounted for 40-50% of all the butterflies seen during monitoring, however they only accounted for 16% of the butterflies seen in this year. In 2015, Cabbage Whites may have been impacted by cold weather during the winter or from a dip in their natural population fluctuations. The low number of Cabbage Whites may have had far reaching consequences for other butterflies on the property as they no longer needed to compete with the Whites for resources.

It may be because of the lack of competition that 2015 had the greatest number of species seen during a monitoring season. Many new observations were recorded on the property in 2015, including the rarely seen Eastern Pine Elfin (*Callophrys niphon*) and several Silvery Blues (*Glaucopsyche lygdamus*), which are not known to Waterloo Region. These new species observations may be indicative of changes in historical ranges and it will be interesting to see if more and more new species will be seen at ***rare*** in the future.

A large amount of change has occurred across the property, overlapping with some transect locations. Sparrow field has been allowed to naturalize and Blair Flats was subjected to a prescribed burn. Interestingly, the lowest number of butterflies was seen in Sparrow field in 2015 and the highest number of butterflies was seen in Blair Flats. Tracking the impact these changes have had on the abundance and richness of butterflies on the property should be a priority during subsequent monitoring years.

Plethodontid Salamander Monitoring

Monitoring of lungless (Plethodontid) salamanders occurs at ***rare*** by turning over pre-placed wooden cover boards in Indian Woods and the Hogsback once a week for nine-weeks each fall.

Eastern Red-backed Salamanders (*Plethodon cinereus*) were overwhelmingly the most abundant species found in both Indian Woods and the Hogsback in 2015 and in every other monitoring year. In Indian Woods, two Blue-spotted Salamanders *(Ambystoma laterale)* were observed in 2015, and in the Hogsback; one Blue-spotted Salamander, one Yellow-spotted Salamander (*Ambystoma maculatum*) and four Four-toed Salamanders (*Hemidactylium scutatum*) were seen. The number of salamanders observed was roughly average compared to data in previous years, but the number in Indian Woods was the second lowest on record. Furthermore, trends in salamander age may indicate that in Indian Woods adult salamanders have decreased in number over the past year, perhaps because of the extreme cold during this past winter. The causes are unclear but a number of other environmental variables and human disturbance may also be playing a role.

These changes in Indian Woods are concerning and the cause, whether temporary or permanent, is unknown. As the salamander monitoring program acts as a warning sign for environmental change, falling numbers coupled with ongoing human pressures from agriculture, development projects, and the potential for accumulative effects from aggregate extraction highlight the need for continued salamander monitoring at ***rare***.

Forest Canopy and Tree Biodiversity Monitoring

The forest canopy and tree biodiversity monitoring program at ***rare*** occurs in in all three major forest areas; the Hogsback, Indian Woods and the Cliffs and Alvars. Three permanent plots are set-up within each area to track changes in the health of the trees within these forests.

The most dominant trees in all forest areas is the Sugar Maple (*Acer saccharum*) followed by American Beech (*Fagus grandifolia*). As forests change slowly there have been few noticeable differences since the beginning of the monitoring program in 2009. Each forest area seems stable overall; however, some species have seen concerning trends. All species of Ash located within the forest monitoring plots are either dead or in severe decline, likely due to the epidemic of the invasive Emerald Ash Borer. Beech trees are in similar peril as at least one quarter of all American Beech within monitoring plots are showing signs of Beech bark disease; a fungal infection which can cause mortality. There are also the remains of one of the 12 mature Butternut trees at ***rare*** located within the monitoring plots. This tree has likely died due to Butternut canker, a fungus that has imperiled Butternut throughout North America. The future of these trees with active invasive pests is unknown and monitoring programs and recovery strategies are recommended to retain these trees.

Soil Humus Decay Rate Monitoring

Decay rate monitoring occurs in late October and early November around one of the permanent forest canopy plots in each forest area. Decay rates are measured by burying wooden tongue depressors below the soil surface and comparing their mass lost over a period of a year to those left on the soil surface.

Decay rates were low in 2015 compared to those seen in previous years. Low decay rates were likely the result of both a long and harsh winter and very low annual precipitation, which both influence rates of decay by making soil conditions dry and cold. Dry and cold conditions limit soil microbial activity and therefore limit the amount of decay that occurs over time.

Decay rates in Indian Woods and Cliffs and Alvars are typically close to even, but the Hogsback has had the lowest rates of decay in every year. Low rates of decay in the Hogsback are most likely caused by the saturated soils as some portions of the monitoring take place within a wetland area where anaerobic conditions limit microbial activity. Overall, no trends of immediate concern have arisen from soil monitoring. Still, soil processes can occur long time scales, therefore monitoring of soil humus decay rates is recommended to continue into the foreseeable future.

Acknowledgements

Many thanks to Employment Ontario and Natural Resources Canada (NRCan) for funding the Ecological Monitoring Intern position; without their support, this monitoring program and report would not have been possible.

I would also like to thank all of the ***rare*** staff. I would like to sincerely thank Jenna Quinn for her support and supervision in collecting the monitoring data and writing this report, it would not have possible without her guidance. Also thank you to my committed volunteers: Gillian Preston, Dan Root and Adelle Strobel who volunteered many hours of their time to help with the monitoring. A big thanks to Erin Sonser as well who helped me get on my feet with monitoring and for all her hard work on the butterfly monitoring report from 2014. Staff from ***rare***; Brock Trojahn, Mike Achtymichuk and Dave Winger also helped in monitoring so I thank them for their support. Thank you to Glenn Richardson from the Toronto Entomologists' Association who helped in identifying skippers. Finally, a special thanks to Julie Reid whose passion for butterflies has helped us discover so many species on the property this year.

NATIONAL CONSERVATION PLAN
PLAN DE CONSERVATION NATIONAL

Canada

This project was undertaken with the financial support of the Government of Canada.
Ce projet a été réalisé avec l'appui financier du gouvernement du Canada.

Cover Photography by Tim Skuse. Clockwise from top right; Blue-spotted Salamander, Silvery Blue, Indian Woods forest scene, and Monarch butterfly.

Table of Contents

List of Figures

List of Tables

List of Appendices

Appendix A: Maps and Coordinates

Appendix B: Equipment Lists

Appendix C: Data Sheets and Codes

Appendix D: Species Lists

Appendix E: Results from Annual Butterfly Count

Appendix F: 2015 Milkweed Survey Results

Appendix G: Additonal Data

Lists of Acronyms

Acronym	Description
EMAN	Ecological Monitoring and Assessment Network
ACO	Artificial Cover Object
IN	Indian Woods
HO	Hogsback
CA	Cliffs and Alvars
SVL	Snout-Vent Length
VTL	Vent-Tail Length
ANOVA	Analysis of Variance
CPUE	Catch Per Unit Effort
AIC	Akaike's Information Criterion
SD	Standard Deviation
dbh	Diameter at Breast Height
IV	Importance Value
SARA	Species at Risk Act
SARO	Species at Risk in Ontario
ADR	Annual Decay Rate

1.0 Introduction

Prepared by: Jenna Quinn & Amy Reinert

1.1 Ecological Monitoring

Ecological monitoring involves measuring a set of environmental variables at regular intervals over a long period of time (Vaughan et al. 2001). The consistent monitoring of these abiotic or biotic environmental variables can provide information about the environmental changes that are occurring within an ecosystem (Lovett et al. 2007). The fundamental reasons for conducting long term ecological monitoring are to establish baseline data, which represents the current status of an ecosystem, and to facilitate the detection of environmental changes over time. Observations of environmental variables that exceed the natural variation in baseline data can be indicative of an environmental change (Vaughan et al. 2001).

The importance of continued long term ecological monitoring has been stressed in the scientific literature as it can provide important information for evaluating ecosystem health (Wolfe et al. 1987; Jeffers 1989; Vos et al. 2001; Lovett et al. 2007). The results of monitoring programs should be considered during policy development in order to create suitable strategies for mitigating and responding to environmental changes (Wolfe et al. 1987; Noss 1990; Beever 2006; Lovett et al. 2007).

Due to the broad scope of biological diversity throughout an entire ecosystem, the limited time, personnel, and money available for monitoring programs often means that only the highest priority indicators can be monitored (Beever 2006). Therefore, measuring the occurrence of a few indicator species is much more feasible than conducting comprehensive species inventories throughout the entire ecosystem (Fleishman et al. 2005). Indicator species are particularly sensitive to changes in their environment and are relatively cost effective and easy to monitor, making them ideal representatives for identifying changes in ecosystem health (Noss 1990).

1.2 Ecological Monitoring and Assessment Network

In 1994, Environment Canada initiated the Ecological Monitoring and Assessment Network (EMAN) which connected the various groups and individuals conducting ecological monitoring across Canada (Craig & Vaughan 2001). These members worked towards the collective goal of determining “what is changing and why in Canadian ecosystems” by achieving the following objectives: 1) determine how Canada’s ecosystems are being influenced by environmental stresses, 2) demonstrate scientific rationale for resource management policies, 3) evaluate the effectiveness of resource management policies, and 4) promptly detect new environmental issues (Vaughan et al. 2001).

The EMAN coordinating office was responsible for developing standardized protocols for the ecological monitoring of marine, freshwater and terrestrial ecosystems across Canada (Environment Canada 2012). The use of standardized protocols improves the ability to detect, describe, and report ecosystem changes by encouraging the collection of comparable data sets. In addition, collected data was uploaded to a shared database to facilitate the analysis of large scale ecosystem changes (Vaughan et al. 2001).

The EMAN coordinating office was closed in September 2010 and the future of EMAN is currently unknown. Protocols can still be accessed from the Environment Canada website but data can no longer be uploaded or accessed.

1.3 Ecological Monitoring at ***rare Charitable Research Reserve***

The ***rare*** **Charitable Research Reserve** provides a unique opportunity for monitoring. Located at the confluence of the Speed and Grand River within Waterloo Region, it is 900+ acres of preserved land surrounded by expanding urban development. A high diversity of habitats supports a wide biodiversity of flora and fauna, providing a good representation of local species (Figure A.1).

An ecological monitoring program was established at ***rare*** in 2006 following EMAN protocols, with the goal of developing baseline data and the hope of creating a long-term protocol to observe changes over time. Due to limitations, such as funding and manpower, monitoring is restricted to indicator species, which are closely tied to environmental changes. Butterfly monitoring began in 2006 on two transects, Cliffs and Alvars and South Field, and was expanded in 2009 to include the newly acquired Thompson's Tract, and again in 2010 to Blair Flats. Plethodontid salamander monitoring began in 2006 in Indian Woods and was expanded in 2008 to include the Hogsback forest. Benthic invertebrate monitoring occurred at Bauman and Cruickston creeks in 2006, and, continuing on a three year cycle, occurred again in 2009 and 2012. In 2009, the monitoring program was expanded to include forest canopy tree biodiversity plots in the Indian Woods and Cliffs and Alvars forests, with soil humus decay rate monitoring also occurring within the Cliffs and Alvars plot. In 2010, an additional forest health plot was added to the Hogsback forest, and soil humus decay rate monitoring was included in all forest plots. Here, the results of the 2013 monitoring year are reported and discussed.

1.4 Literature Cited

Beever, E.A. 2006. Monitoring biological diversity: Strategies, tools, limitations and challenges. *Northwestern Naturalist*. 87(1):66-79.

Craig, B., & Vaughan, H. 2001. EMAN and biosphere reserves: cooperating in providing information for ecozone and local ecosystem management. *Niagara Escarpment Commission- Leading Edge*.

Environment Canada. 2012. Ecological Monitoring. Available at: http://www.ec.gc.ca/faunescience-wildscience/default.asp?lang=En&n=B0D89DF1-1. Accessed June 2012.

Fleishman, E., Thomson, J.R., MacNally, R., Murphy, D.D. & Fay, J.P. 2005. Using indicator species to predict species richness of multiple taxonomic groups. *Conservation Biology*. 19(4):1125-1137.

Jeffers, J.N.R. 1989. Environmental monitoring. *Biologist*, 36:171.

Lovett, G.M., Burns, D.A., Driscoll, C.T., Jenkins, J.C., Mitchells, M.J., Rustad, L., Shanley, J.B., Likens, G.E., & Haeuber, R. 2007. Who needs environmental monitoring? *Frontiers in Ecology and the Environment*. 5(5):253-260.

Noss, R. 1990. Indicators for monitoring biodiversity: a hierarchical approach. *Conservation Biology*, 4(4):355-363.

Vaughan, H. Brydges, T., French, A., & Lumb, A. 2011. Monitoring long-term ecological changes through the ecological monitoring and assessment network: Science-based and policy relevant. *Environmental Monitoring and Assessment*, 67:3-28.

Vos, P., Meelis, E., & Ter Keurs, W.J. 2000. A framework for the design of ecological monitoring programs as a tool for environmental and nature management. *Environmental Monitoring and Assessment*, 61:317-344.

Wolfe, D.A., Champ, M.A., Fletcher, D.A., & Mearns, A.J. 1987. Long-term biological data sets: their role in research, monitoring, and management of estuarine and coastal marine systems. *Estuaries*, 10:181-193.

2.0 Butterfly Monitoring Prepared by: Tim Skuse

2.1 Introduction

2.1.1 Lepidoptera Taxonomy

The order Lepidoptera, meaning “scaled wings”, is made up of butterflies and moths. There are six families of butterflies, including five true families (*Papilionidae*, *Nymphalidae*, *Pieridae*, *Lycaenidae*, and *Riodinidae*) and the skipper family (*Hesperiidae*). An estimated 17,500 species of butterfly species exist, with approximately 13,750 of those being true butterflies (Robbins and Opler 1997). Although exceptions do exist, there are some general rules regarding their appearance and behaviour that can be used to distinguish moths and butterflies from one another. Butterflies are predominately diurnal, have clubbed antennae, and fold their wings vertically over their body while at rest, whereas moths are predominately nocturnal, have feathered or tapering antennae, and hold their wings out flat when resting (Pyle 1981).

2.1.2 Why Monitor Butterflies?

Long term monitoring of butterfly populations can provide valuable insight into the overall health of ecosystems and environmental change. Butterflies have short life spans, allowing them to respond quickly to various ecological pressures, both locally and on a broader scale (Fleischmann and Murphy 2009). Butterflies are sensitive to regional weather conditions, as unseasonably cold or wet periods can delay their development and reproduction (Pollard 1988). Further, global climate change can result in an extension or shift of butterfly populations outside of their typical ranges (Roy et al. 2001). Climate warming is expected to allow butterflies to expand their ranges to higher elevations and latitudes, particularly along the geographic margins of their current ranges (Oliver et al. 2012). However, this ability to increase geographic range is largely species specific, with species with high dispersal ability or those willing to use a range of host plants appearing to best exploit warmer temperatures (Kallioniemi 2013). Therefore, the presence or absence of butterfly species within geographic regions could provide useful information to better understand environmental change.

Throughout their life cycle, butterflies have specific requirements, namely the host plants they require for egg laying and feeding as both caterpillars and adults. Changes in the availability of host plants through both natural and human-caused disturbances (i.e. habitat loss) can have negative effects on butterfly populations. Invasive plant species can out-compete the native plants butterfly require. However, some butterfly species are now taking advantage of these alien plants and using them as effective hosts (Tallamy and Shropshire 2009). For example, the Wild Indigo Duskywing (*Erynnis baptisiae*); this butterfly species historically was uncommon, and restricted to habitats in southwestern Ontario where its larval food plant, Wild Indigo (*Baptisia tinctora*), was found (Hall 2009). However, its range has been expanding as a result of it using a non-native foodplant, Crown Vetch (*Coronilla varia*), and since 2010, it has been more commonly observed in the Waterloo Region (Linton 2012) and at ***rare***.

In addition to their rapid responses to ecological change, butterflies make good indicator species because of the ease in which they can be monitored (Fleischmann and Murphy 2009). Their size and the colourful distinctions between species make observation and identification relatively simple. Finally, butterflies invoke a positive response from the public, allowing for recruitment of volunteers and the promotion of citizen science programs.

Over the past few decades, populations of butterflies have declined at an alarming rate in many parts of the world (Merckx et al. 2013; Brereton et al. 2014). Habitat loss, pesticide use, habitat degradation and fragmentation are just some of the proposed causes that have led to these declines (Merckx et al. 2013). Continued pressure from these sources and the response from butterfly populations highlight the urgent need for monitoring efforts to help in making conservation management decisions.

2.1.3 Butterfly Monitoring at ***rare Charitable Research Reserve***

The standardized Ecological and Monitoring Assessment Network (EMAN) protocol for long term butterfly monitoring was developed and piloted at ***rare*** in 2006. The purpose of this pilot program was to determine if the Transect Walk Method (Pollard 1977) was a feasible technique to examine butterfly abundance and diversity in Canada (Grealey 2006), and it marked the start of the long term monitoring program at ***rare***.

In 2006, two transects were established: one located in the Cliffs and Alvars and one in South Field/Sparrow Field. Baseline data were collected over a five week period during the initial pilot study. Butterfly monitoring at ***rare*** continued in 2009 and subsequent years, during which time two more transects were established: one in 2009 in the newly acquired Thompson Tract, and one in 2010 in Blair Flats. Monitoring took place over thirteen weeks in 2009, and fourteen weeks in 2010 through to 2015. It is important to note that due to a change in property boundaries, the South Field/Sparrow Field transect had to be slightly altered in 2014; the changes are described below (Section 2.2).

2.2 Methods

2.2.1 Monitoring Protocol

One of the most commonly used monitoring methods around the world is the Transect Walk Method, originating in Britain in 1976 (Pollard 1977; Pollard and Yates 1993). This method involves walking established routes (i.e. transects) at a uniform pace, and making observations within a given radius (Pollard 1977). Butterfly monitoring at the ***rare*** **Charitable Research Reserve** is conducted using the Transect Walk Method, as it does not require extensive effort or time, and limits disturbances to the butterflies' behaviour.

Ideally, butterfly monitoring programs should take place over a 26 week period, from April to September (Layberry et al. 1998). At ***rare***, this time period has been reduced due to both time and monetary constraints. In 2006, monitoring took place over a five week period in mid-July and August. In 2009, the monitoring period was expanded to thirteen weeks. Starting in 2010 and for all subsequent years, monitoring has taken place over fourteen weeks, starting mid-May and ending mid-August. Monitoring typically begins on the third Monday of May; however, this may be either advanced or delayed, depending on weather conditions (i.e.

particularly cold or warm local temperatures). Butterflies are most active during the warmest part of the day, and thus monitoring is completed between the hours of 10am and 3pm. It is also recommended that monitoring be completed on sunny days, when the temperature is above 13°C; if it is overcast, the temperature must be at least 17°C (UK Butterfly Monitoring Scheme; Butterfly Monitoring Scheme Germany). Wind should also be less than five on the Beaufort Wind Scale (refer to table C.1. in the Appendix).

Butterfly monitoring at ***rare*** took place at four transects. Each transect is broken into sections, each section with a stopping point, as described in Appendix A. Each individual section was created based on changes in habitat type. Prior to beginning monitoring, the observer walked the transects and flagged the section breaks and stopping points, as required. Observations were recorded during optimal weather conditions; in the absence of rain, observations were recorded in suboptimal conditions, as this is more valuable than not collecting data at all. In order to minimize observer bias, all observations were made by one individual with occasional assistance in spotting individual butterflies from volunteers .

In 2015, monitoring began on May 19th, and each of the four transects were walked once per week for fourteen weeks. A recommended list of field equipment can be found in Appendix B. At the start and end of each transect, the time was recorded, and a hand-held Kestrel 3000© (Nielson-Kellerman, Boothwyn, PA, USA) was used to determine air temperature. The transects were walked at a uniform pace, and butterflies observed within a ten metre radius were recorded. Halfway through each section, ten minute stops were made at predetermined locations, again recording any butterflies observed within a ten metre radius. At the stopping points, the percent of blue sky was estimated (0-100; 0 being full cloud cover), and the Kestrel 3000© was used to determine average wind speed. Butterflies were visually identified in the field, and caught with a net when necessary to aid in identification. Unknown species were photographed and sent to local experts for identification. If identification was not possible, the individual was recorded as the most common species based on data from previous years and a note was made due to the uncertainty. While walking the transects, occasional stops were permitted to properly identify butterflies, and recording continued from where the stop was made. All observations were recorded in a standard field form, found in Appendix C and on the ***rare*** server.

2.2.2 Transect Descriptions

Butterfly monitoring occurred across the following transects at the ***rare* Charitable Research Reserve**. Refer to Appendix A for a map of the property which outlines the transect routes.

The **Cliffs and Alvars** transect is 3.5 km and follows primarily the River and Grand Trunk trails. A large part of the transect consists of mature hardwood forest stands, dominated by American Beech (*Fagus grandifolia*) and Sugar Maple (*Acer saccharum*). This transect also passes through deciduous swamps, limestone cliffs, open alvar habitats, and an extensive floodplain.

The **South Field/Sparrow Field** transect is 2.9 km, running along the edge of agricultural fields, hedgerows, and through a recently restored tall grass prairie. Several fields in the area are currently in agricultural production, including hay in South Field West and corn in South Field East in 2015. Prior to the 2014 monitoring year, this transect traveled along the south-eastern perimeter of Indian Woods. However, due to a change in the ***rare*** property boundary in early 2014, this part of the transect (formerly section 6 and 7) was eliminated and an alternative route was used. To minimize the effects of this change, the new section is referred to as 6/7 (allowing the other sections to remain as they were).

The **Thompson Tract** transect is 2.2 km and follows established trails through meadows, plantations, and lowland and upland forest dominated by American Beech (*Fagus grandifolia*) and Sugar Maple (*Acer saccharum*). Thompson Tract is located at the western boundary of the ***rare*** property.

The **Blair Flats** transect is a 1.3 km loop that walks the perimeter of a restored tall grass prairie. Prior to 2010, Blair Flats was in agriculture production. As part of a long term study, the area was restored to a tall grass prairie, and is currently dominated by Goldenrod (*Solidago*), Queen Anne's Lace (*Daucus carota*), Black Eyed Susan (*Rudbeckia hirta*), and Tansy (*Tanacetum vulgare*). In 2015 Blair Flats was burned as part of a five year prescribed burn program, which intends to encourage and promote native prairie plants and overall tallgrass prairie ecosystem health. Beginning at the large Bur Oak (*Quercus macrocarpa*) just off of Blair Road, the transect heads north towards the river, turns west and runs parallel to the river, then turns south and follows the property boundary, and finally traveling eastward, parallel to the road and ending at the Bur Oak.

2.2.3 Data Analysis

Data were analyzed using SPSS Statistics Version 20 and Microsoft Excel 2010. Most of the analyses compare data within each individual transect over time rather than between each transect, as each transect is of varying length and habitat, and direct comparisons are of limited use. Within transects, the number of individuals observed were fit to a Poisson regression model . An ANOVA was used to determine if significant differences occurred in the data and pairwise comparisons (TukeyHSD) were used to identify where differences occurred (P=0.05).

The Shannon Diversity Index and species evenness for 2015 were calculated and compared with those from previous years (Figure 2.1). Species evenness refers to the relative abundance of individuals of different species, and the Shannon Diversity Index takes into account the evenness and the total number a species to produce a score from 0-4. Zero (0) indicates very low diversity, while 4 is very high diversity; real world values typically fall between 1.5 and 3.5 (Magurran 2004).

$$H = -\sum_{i=1}^{S} p_i \ln p_i$$

Shannon Diversity Index: Where p_i is the proportion of individuals belonging to the *i*th species and S is the number of species.

$$E_H = H/\ln(S)$$

Species Evenness: Where H is the Shannon Diversity Index and S is the number of species

$$G_j = \left(\left(\prod_{i=1}^{N} \frac{x_j}{x_1} \right)^{1/N} \right)$$

Geometric Mean Index: Where G is the index in year *j* calculated from the Nth root of the product of relative abundances (*x*) for each species compared to the base year (2010)

Figure 2.1: Formulas used for calculating the Shannon Diversity Index, species evenness value (Magurran 2004), and Geometric Mean Index (Buckland et al. 2011).

Calculation and comparison of a composite index of Geometric Means was also used to analyze observations within transects over time (See Figure 2.1). Geometric Means are sensitive to both species abundance as well as evenness (Buckland et al. 2011). Calculation of Geometric Means has the benefit over the Shannon Diversity Index in that it will be able to detect overall population changes if there is no change in evenness (i.e. all butterfly species have had a drop in population) whereas Shannon Diversity will register no change (Buckland et al. 2005;van Swaay et al. 2008; Buckland et al. 2011). However, Geometric Mean results will be negatively impacted by rarely recorded species, which is negligible for the Shannon Index (Buckland et al. 2011). Therefore, all Geometric Mean calculations excluded those species that were not recorded in every monitoring year. Geometric Means were calculated using 2010 as a baseline year, since this is the first year of complete fourteen week monitoring across all four transects. The base year has an index value of 1.0 and subsequent years will either be higher or lower indicating greater abundance and/or evenness or a decline in abundance and/or evenness, respectively.

2.3 Results

2.3.1 Overall Abundance and Diversity

The 2015 butterfly monitoring period saw a total of 4931 individual butterflies from 55 species within the four transects at the ***rare*** **Charitable Research Reserve**. Of these 55 species, four were new sightings never before observed during monitoring, bringing the total number of species recorded by the monitoring program to 67. The most abundant species observed in 2015 were; Cabbage White (N=779), European Skipper (N=687), Clouded Sulphur (N=546), Common Wood Nymph (N=448), Inornate Ringlet (N=392), and the Northern Crescent (N=383). From 2012-2014 the most abundant species were the Cabbage White followed by the Clouded Sulphur and the Common Wood Nymph. The European skipper supplanted the Clouded Sulphur and Common Wood Nymph in 2015 to make it the second most abundant species observed. The total number of individuals observed by species in 2015, as well as the Waterloo Regional Status for each species, can be seen in Table 2.1. The total number of individuals and species in each transect, the Shannon Diversity Index, the Geometric Means Index and species evenness values from 2015 are compared to those from previous monitoring years in Table 2.2.

Table 2.1: Summary of all observed butterflies in the 2015 monitoring season at the ***rare* Charitable Research Reserve**. The Waterloo Regional Status for each of the observed species is also included from Linton (2012).

	TRANSECT					
SPECIES	**1**	**2**	**3**	**4**	**TOTAL**	**REGIONAL STATUS**
American Lady		3	1		**4**	**Common**
Banded Hairstreak	18		8		**26**	**Uncommon**
Black Dash	3				**3**	**Uncommon**
Black Swallowtail	3	14	2	11	**30**	**Very Common**
Bronze Copper		1		2	**3**	**Very Common**
Cabbage White	330	256	87	106	**779**	**Very Common**
Clouded Sulphur	108	179	102	157	**546**	**Very Common**
Columbine Duskywing		1			**1**	**Rare**
Common Sootywing	1	26			**27**	**Rare**
Common Wood Nymph	98	76	191	83	**448**	**Very Common**
Coral Hairstreak	1				**1**	**Uncommon**
Crossline Skipper		1	1		**2**	**Rare**
Delaware Skipper	5	8	7	20	**40**	**Common**
Dun Skipper	7		7	1	**15**	**Very Common**
Eastern Comma	11	3	7		**21**	**Very Common**
Eastern Pine Elfin			1		**1**	**Rare**
Eastern Tailed Blue	1	8		12	**21**	**Uncommon**
Eastern Tiger Swallowtail	14	2	12	3	**31**	**Very Common**
European Skipper	255	23	400	9	**687**	**Very Common**
Eyed Brown	35				**35**	**Very Common**
Giant Swallowtail	10		7		**17**	**Uncommon**
Great Spangled Fritillary	28	2	39	2	**71**	**Very Common**
Grey Comma	1		3	1	**5**	**Uncommon**
Harvester			1		**1**	**Rare**
Hobomok Skipper	25	13	37		**75**	**Common**
Inornate Ringlet	70	146	159	17	**392**	**Common**
Juvenal's Duskywing	39	2	29		**70**	**Rare**
Least Skipper	2				**2**	**Uncommon**
Little Glassywing	2		1		**3**	**Uncommon**
Little Wood Satyr	114	6	142	1	**263**	**Very Common**
Long Dash			4		**4**	**Uncommon**
Milbert's Tortoiseshell	23	4		3	**30**	**Uncommon**
Monarch	8	15	24	6	**53**	**Very Common**
Mourning Cloak	3	4	8	1	**16**	**Very Common**
Northern Broken Dash	11		12	1	**24**	**Common**

Northern Crescent	91	55	222	15	**383**	**Uncommon**
Northern Pearly Eye	34	9	123		**166**	**Common**
Orange Sulphur	5	3		6	**14**	**Very Common**
Painted Lady		2		4	**6**	**Common**
Pearl Crescent	17	74	10	12	**113**	**Common**
Peck's Skipper	2		16		**18**	**Very Common**
Question Mark	5	1	3		**9**	**Very Common**
Red Admiral	62	21	30	30	**143**	**Very Common**
Red Spotted Purple	41	1	23	3	**68**	**Common**
Silver-Bordered Fritillary			31		**31**	**Rare**
Silver-Spotted Skipper	3	1	13		**17**	**Uncommon**
Silvery Blue		5	16		**21**	**Unknown**
Spring Azure	36	1	17	1	**55**	**Common**
Striped Hairstreak	7	1	1		**9**	**Uncommon**
Summer Azure	33	11	21	6	**71**	**Very Common**
Tawny Emperor			4	1	**5**	**Uncommon**
Tawny-edged Skipper	19	3	1		**23**	**Common**
Viceroy	3	7	8	3	**21**	**Very Common**
White Admiral			1		**1**	**Uncommon**
Wild Indigo Duskywing	6	1	2	1	**10**	**Unknown**
Totals	**1590**	**989**	**1834**	**518**	**4931**	

Table 2.2: Summary of butterfly observations for each transect by year, the Shannon Diversity Index, Species Evenness, and Geometric Mean Index for every 14 week monitoring period.

		Number of Individuals (n)	Species Richness (S)	Species Evenness (E)	Shannon-Diversity Index (H)	Geometric Mean Index (G)
Transect One	**2009**	620	25	0.59	1.90	
	2010	1063	33	0.59	2.07	1
	2011	1453	35	0.50	1.77	1.47
	2012	2826	46	0.57	2.19	3.23
	2013	1494	43	0.65	2.45	1.97
	2014	1365	47	0.71	2.72	1.9
	2015	1590	43	0.76	2.85	2.48
Transect Two	**2009**	717	24	0.52	1.65	
	2010	1778	26	0.44	1.42	1
	2011	1146	30	0.47	1.60	1.5
	2012	2427	37	0.49	1.76	2.45
	2013	1751	35	0.57	2.02	2.45
	2014	1130	31	0.62	2.12	2.11
	2015	989	38	0.67	2.43	1.58
Transect Three	**2010**	938	30	0.70	2.37	1
	2011	911	35	0.72	2.56	1.34
	2012	2116	38	0.71	2.56	2.2
	2013	1636	36	0.71	2.55	2.15
	2014	1354	38	0.72	2.62	1.5
	2015	1834	44	0.73	2.75	2.78
Transect Four	**2010**	270	14	0.49	1.30	1
	2011	298	20	0.42	1.26	0.9
	2012	497	35	0.60	2.12	2.47
	2013	381	21	0.63	1.93	1.87
	2014	256	26	0.74	2.42	2.19
	2015	518	29	0.67	2.26	2.5

2.3.2 Transect One: Cliffs and Alvars

A total of 1590 individual butterflies and 43 different species were observed in the 14 week monitoring period in 2015 in Transect One. This monitoring year was the second most abundant year for total butterflies, but still substantially behind the 2826 individuals recorded in 2012. The number of species observed in 2015 (N=43) was the third most across monitoring years. The 2015 species evenness and Shannon Diversity Index were both the highest ever for Transect One at 0.76 and 2.85, respectively, and the Geometric Mean was second highest at 2.48 (See Table 2.2).

The total number of butterflies recorded in 2012 was significantly higher than all monitoring years ($p<0.05$) with the exception of 2015 ($p < 0.05$) (See Figure 2.2). These were the only significant differences observed between all monitoring years.

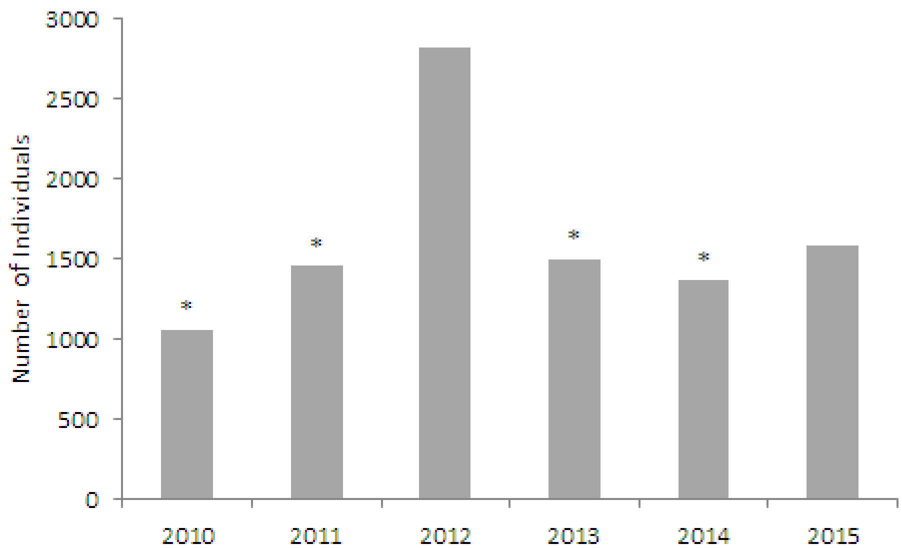


Figure 2.2: Comparison of total observed butterflies in Transect One for each monitoring year. Asterisk (*) denotes a significant difference ($p<0.05$) in observations between monitoring year and 2012, the most abundant monitoring year for Transect One.

The most abundant species in Transect One were the Cabbage White (N=330), European Skipper (N=255) and Little Wood Satyr (N=114), accounting for 44.0% of the total observations in Transect One. The Cabbage White and European Skipper have been the most abundant species in this transect across all monitoring years, with the exception of 2013, where the European Skipper was third most abundant. A total of thirteen species had their highest abundance in 2015 at this transect, they are; Banded Hairstreak, Great Spangled Fritillary, European Skipper, Hobomok Skipper, Juvenal's Duskywing, Milbert's Tortoiseshell, Northern Broken Dash, Red-spotted Purple, Silver-spotted Skipper, Spring Azure, Striped Azure, Summer Azure, Tawny-edged Skipper, Wild Indigo Duskywing. The Coral Hairstreak was spotted in this transect for the first time since 2009 and the Grey Comma was seen for the first time ever. Figure 2.3 and Figure 2.4 show abundances of butterfly species for Transect One.

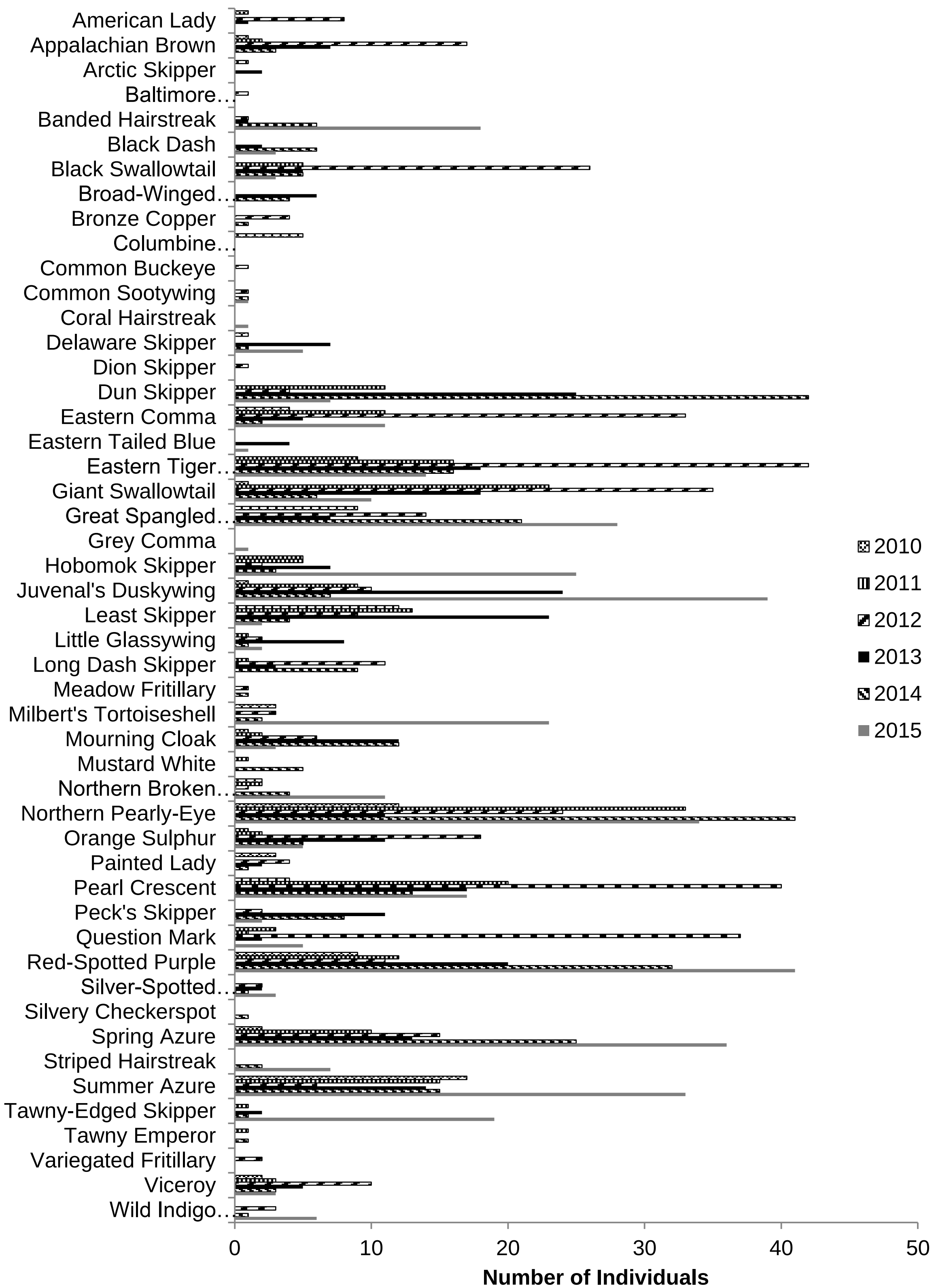


Figure 2.3: Comparison of the number of individuals of each species observed on Transect One for 2010-2015 monitoring periods. Only species with less than 50 observations are shown.

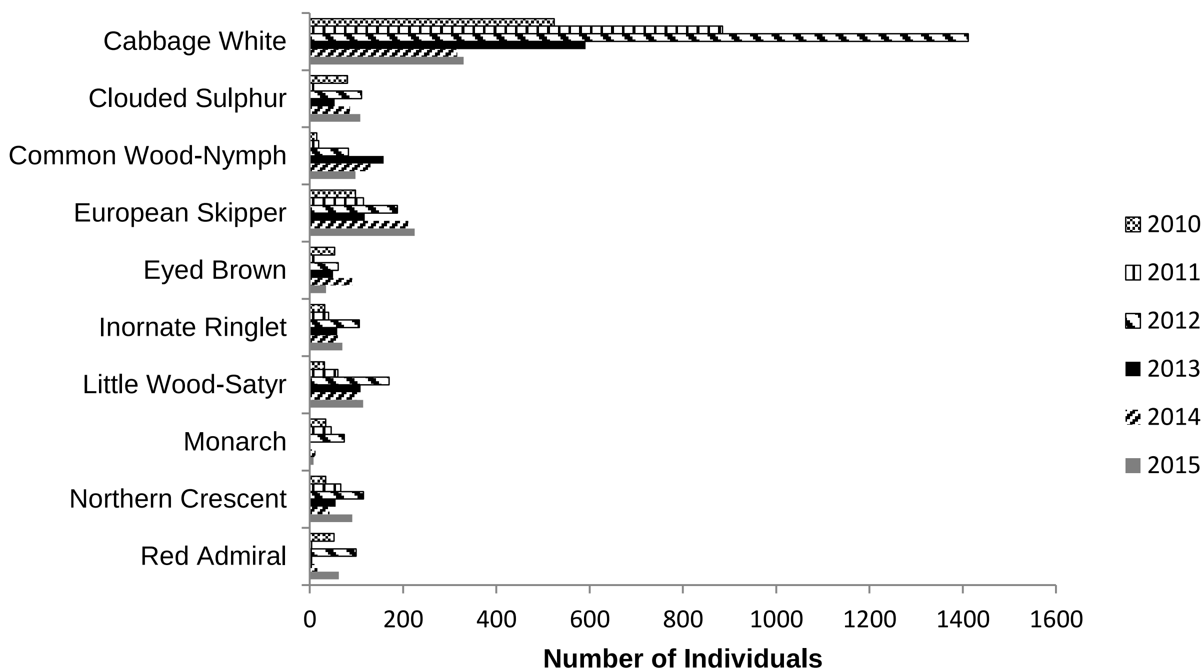


Figure 2.4: Comparison of the number of individuals of each species observed on Transect One for 2010-2015 monitoring periods. Only species with more than 50 observations are shown.

2.3.3 Transect Two: South Field/Sparrow Field

A total of 989 individuals and 38 different species were observed in 2015 at Transect Two; the lowest ever recorded abundance since 2009, but the highest ever recorded species richness. Similar to that of Transect One, the species evenness and Shannon Diversity Index were the highest recorded in Transect Two for any monitoring year at 0.67 and 2.43, respectively, and the Geometric Mean was the second lowest at 1.58 (See Table 2.2).

Although the total number of observations in Transect Two has been fluctuating across years, the only significant differences in abundance were between 2015 and 2012 ($p<0.05$), with 2012 as the most abundant year for monitoring ($p < 0.05$) (See Figure 2.5).

The most abundant species in this transect were the Cabbage White (N=256), Clouded Sulphur (N=179) and Inornate Ringlet (N=146) accounting for 58.7% of all observations on this transect. Cabbage White and Clouded Sulphur butterflies have consistently been the two most abundant species within this transect across all monitoring years with the Clouded Sulphur occasionally being the most abundant (2010 and 2014). In 2015, five species of butterfly were seen for the first time ever in Transect Two, they are; Columbine Duskywing, Bronze Copper, Crossline Skipper, Silvery Blue and Tawny-edged Skipper. Common Sootywings, Milbert's Tortoiseshells, Common Wood Nymphs and Pearl Crescents had their most abundant year on record in 2015. Figure 2.6 and Figure 2.7 show abundances of butterfly species for Transect Two.

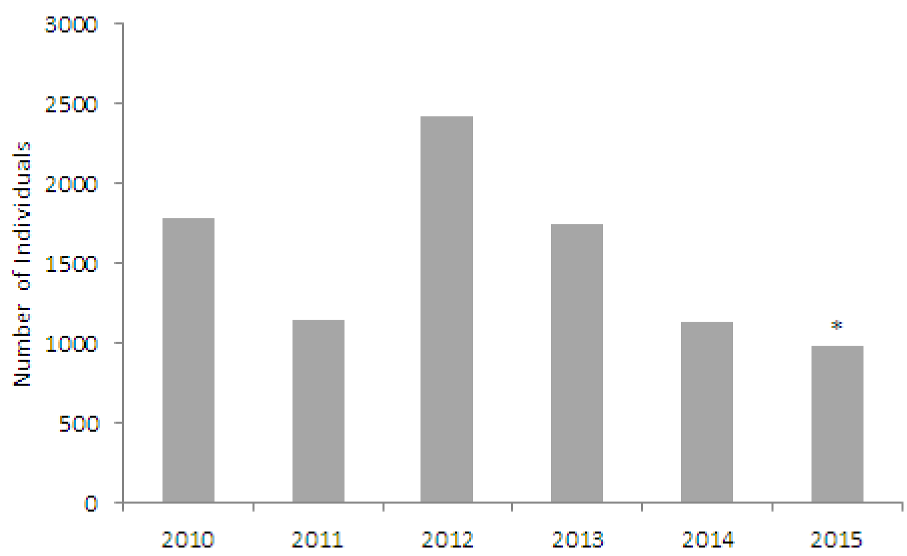


Figure 2.5: Comparison of total observed butterflies in Transect Two for each monitoring year. Asterisk (*) denotes a significant difference ($p<0.05$) in observations between monitoring year and 2012, the most abundant monitoring year for Transect Two.

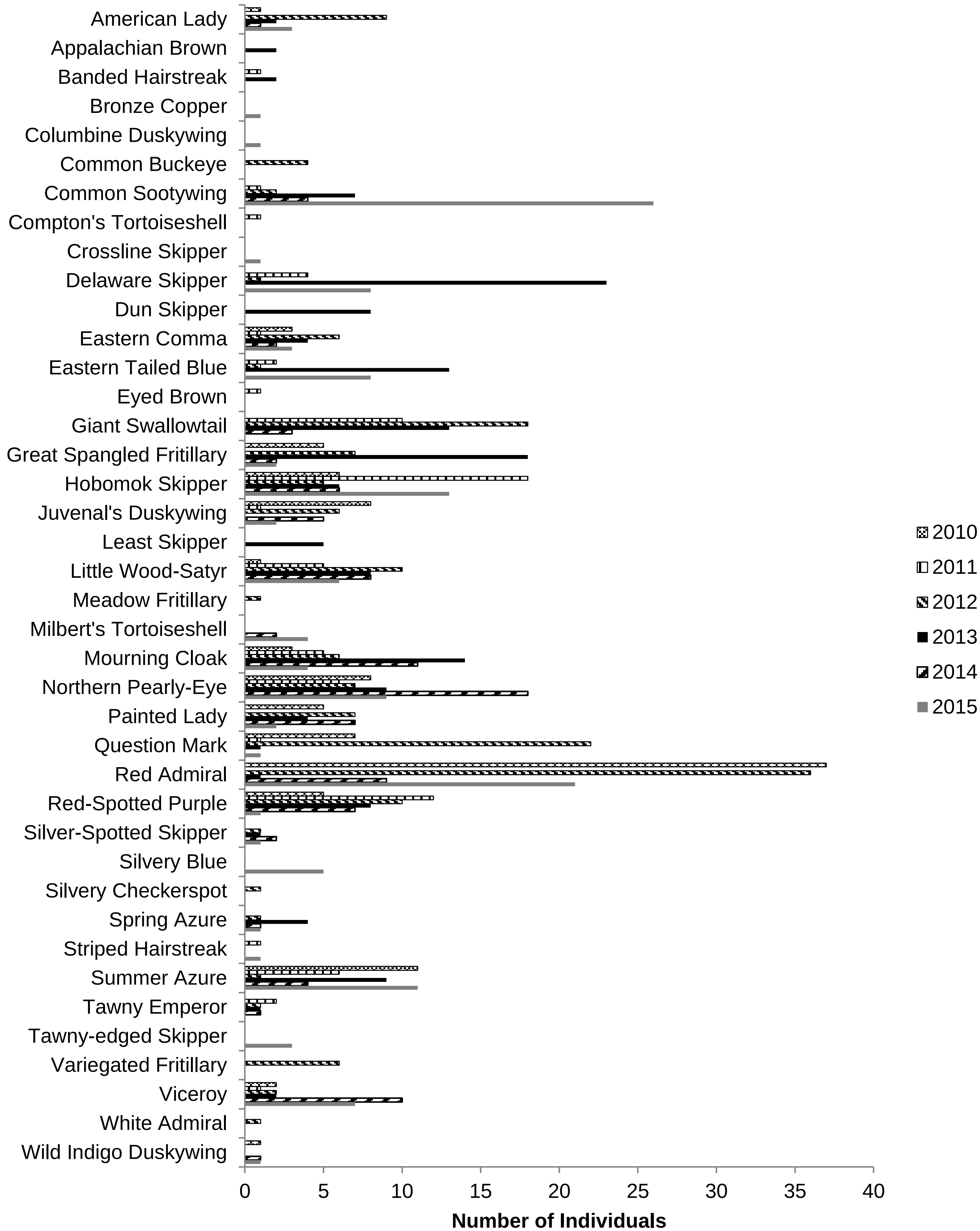


Figure 2.6: Comparison of the number of individuals of each species observed on Transect Two for 2010-2015 monitoring periods. Only species with less than 50 observations are shown.

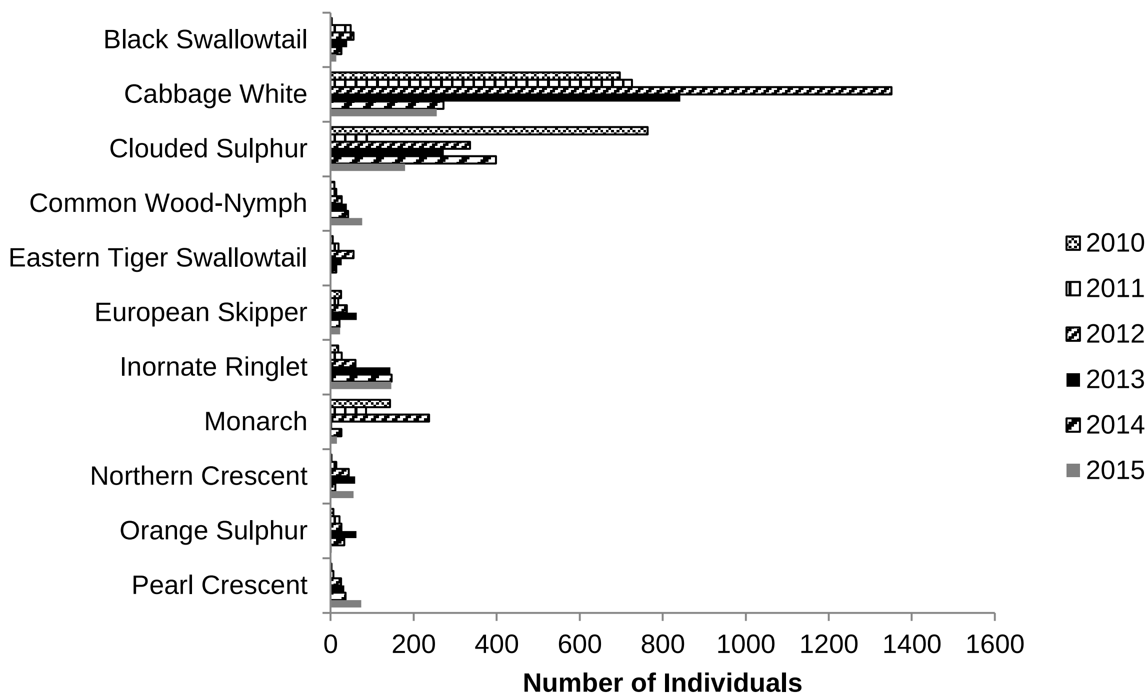


Figure 2.7: Comparison of the number of individuals of each species observed on Transect Two for 2010-2015 monitoring periods. Only species with more than 50 observations are shown.

2.3.4 Transect Three: Thompson Tract

There were a total of 1834 individuals and 44 different species observed in 2015 in Transect Three. This year recorded the second most individual butterflies and the highest number of species ever. Species evenness was 0.73, Shannon Diversity Index was 2.75, and the Geometric Mean was 2.78, all the highest on record (See Table 2.2)

The only significant differences in abundance existing across years were between 2012 and 2010 ($p<0.05$), and 2012 and 2011 ($p<0.05$) ($p < 0.01$) (See Figure 2.8).

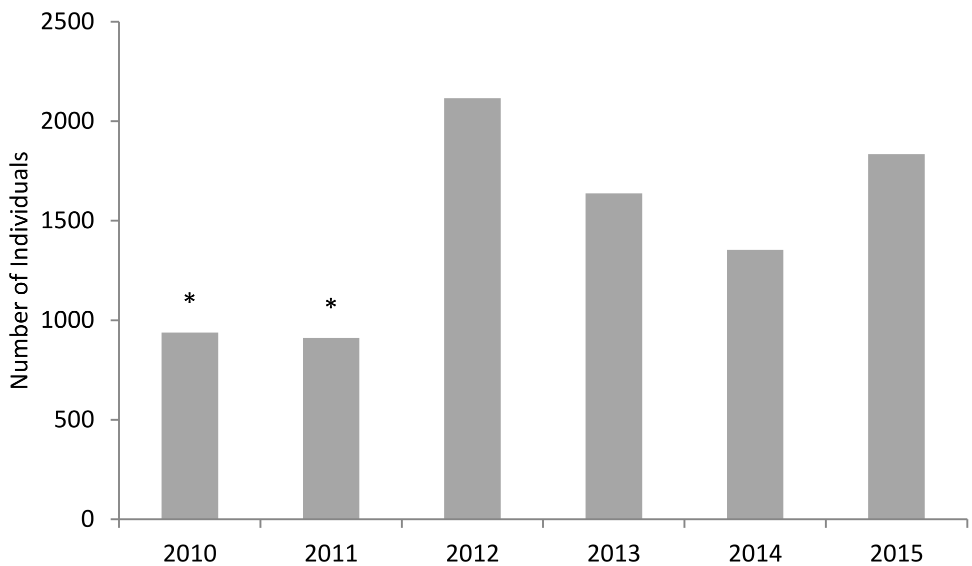


Figure 2.8: Comparison of total observed butterflies in Transect Three for each monitoring year. Asterisk (*) denotes a significant difference ($p<0.05$) in observations between monitoring year and 2012, the most abundant monitoring year for Transect Three.

European Skipper (N=400) was the most abundant species observed, followed by the Northern Crescent (N=222) and the Common Wood Nymph (N=191), accounting for 44.3% of total observations on this transect. Figure 2.9 and Figure 2.10 show abundances of butterfly species for Transect Three. Past years have had most common species as the Cabbage White (2011, 2012) or the Common Wood Nymph (2013, 2014) and this is the first time the European Skipper was recorded as the most abundant species. Seven species were recorded for the first time ever on Transect Three in 2015, they are; Crossline Skipper, Northern Broken Dash, Eastern Pine Elfin, Grey Comma, Silvery Blue, Striped Hairstreak, and Tawny-edged Skipper. Ten species had their most abundant years ever record in Transect Three, they are; Banded Skipper, Delaware Skipper, European Skipper, Great Spangled Fritillary, Hobomok Skipper, Long Dash Skipper, Northern Crescent, Silver-bordered Fritillary, Summer Azure, Tawny Emperor and Viceroy.

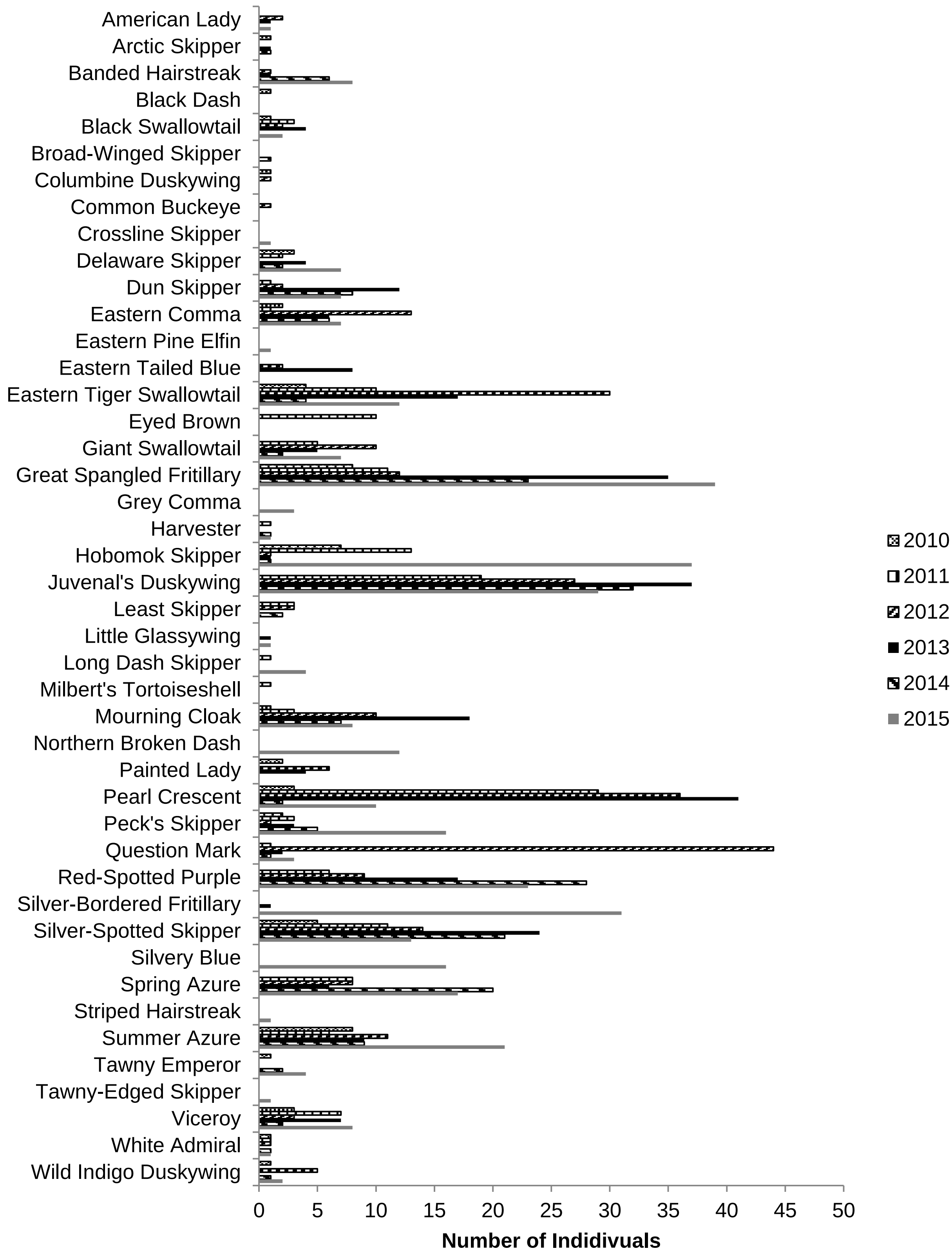


Figure 2.9: Comparison of the number of individuals of each species observed on Transect Three for 2010-2015 monitoring periods. Only species with less than 50 observations are shown.

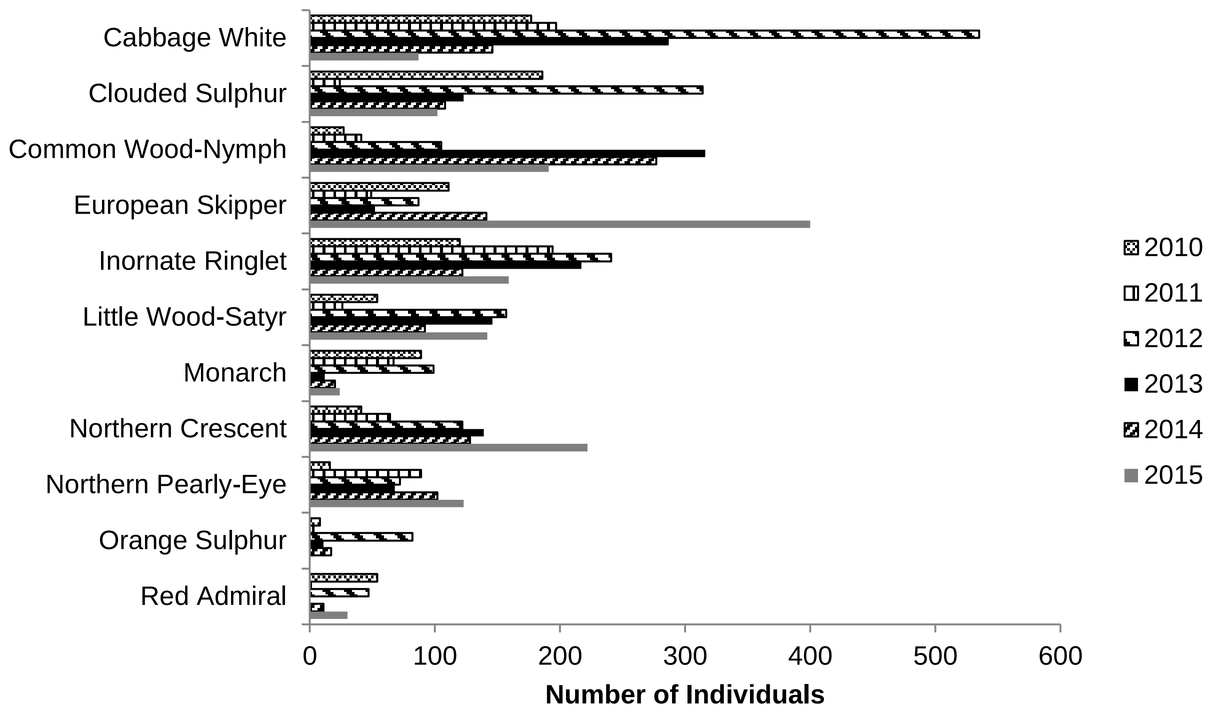


Figure 2.10: Comparison of the number of individuals for each species observed on Transect Three for 2010-2015 monitoring periods. Only species with more than 50 observations are shown.

2.3.5 Transect Four: Blair Flats

The total number of butterflies observed in 2015 at Transect Four was the highest of all monitoring years, at 518 individuals, and the second highest species richness at 29. The species evenness was 0.67 and the Shannon Diversity Index was 2.26; the second highest of any monitoring year, surpassed only by 2014, while the Geometric mean the highest at 2.5 (See Table 2.2).

Despite recording the highest ever abundance in 2015 there was no significant difference between 2015 and any other year and in fact there was no significant difference between all years on record ($p>0.05$) (See Figure 2.11).

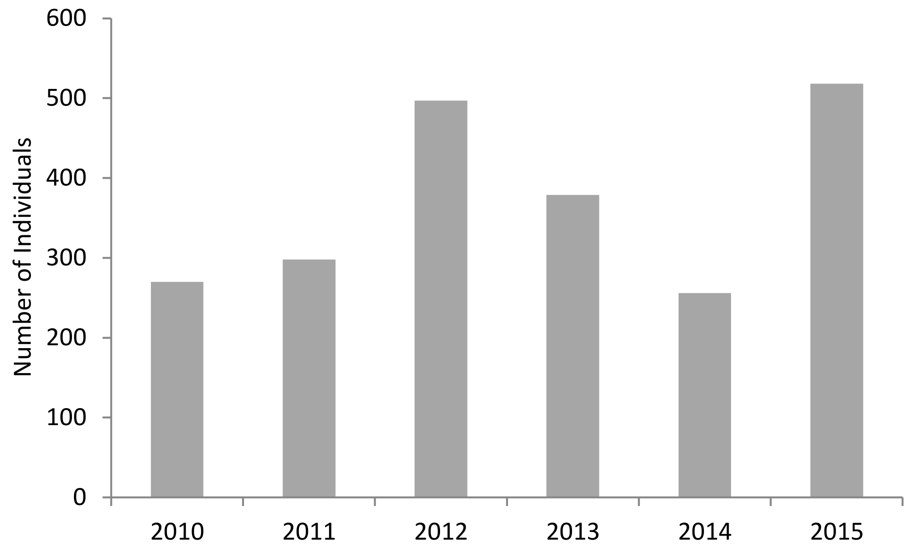


Figure 2.11: Comparison of total observed butterflies in Transect Four for each monitoring year. No significant differences observed ($p>0.05$).

The most abundant species observed in this transect were the Clouded Sulphur (N=157), Cabbage White (N=106), and the Common Wood Nymph (N=83), accounting for 66.8% of observations. These three species have been the most abundant across all monitoring years; however the Clouded Sulphur has experienced a population surge in 2014 and 2015 overtaking the previously most abundant species, the Cabbage White. 2015 was the first year both the Milbert's Tortoiseshell and the Grey Comma were observed in Transect Four. 2015 also saw the most ever observations for Bronze Cooper, Delaware Skipper, Eastern Tailed Blue, Inornate Ringlet, Northern Crescent, Red Admiral, Summer Azure, Clouded Sulphur and the Common Wood Nymph. Figure 2.12 and Figure 2.13 show abundances of butterfly species for Transect Four.

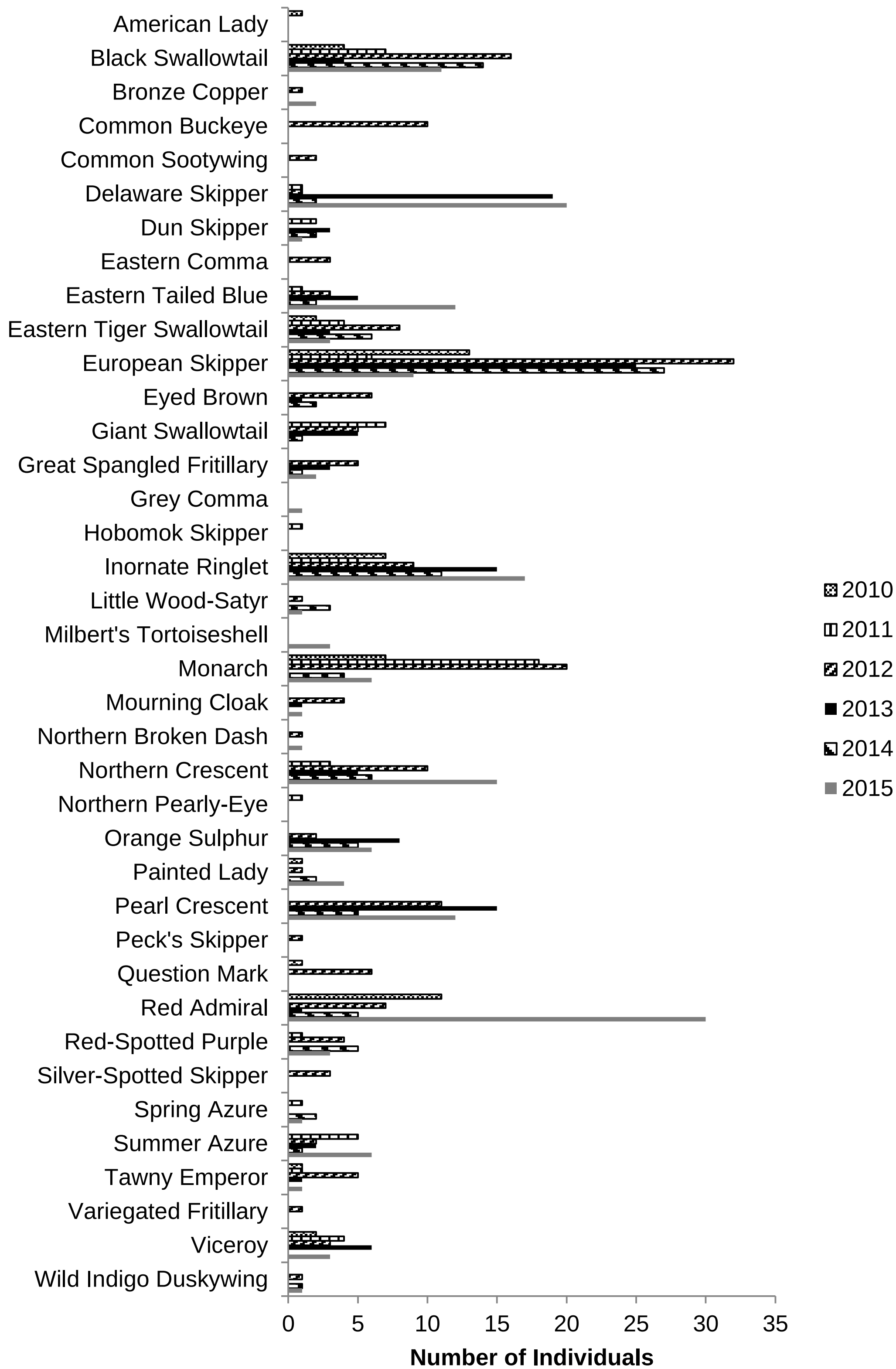


Figure 2.12: Comparison of the number of individuals for each species observed on Transect Four for 2010-2015 monitoring periods. Only species with less than 50 observations are shown.

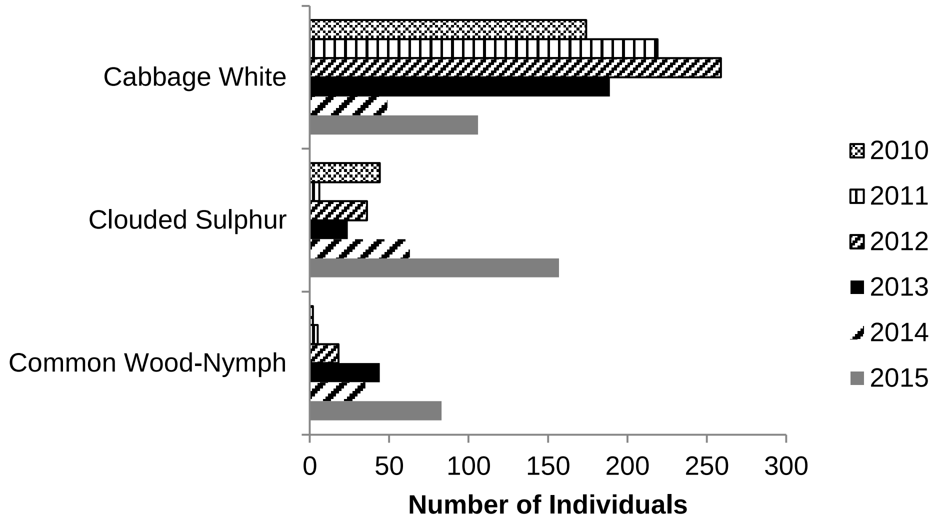


Figure 2.13: Comparison of the number of individuals of each species observed on Transect Four for 2010-2015 monitoring periods. Only species with more than 50 observations are shown.

2.3.6 Species of Interest and New Observations

Monarch butterflies have seen a sharp decline in population in the past three monitoring seasons (See Figure 2.14). Populations peaked in 2012 with 430 individuals observed and bottomed out at 17 individuals the following year in 2013. It is important to note that there was a route change in Transect Two beginning in 2014 (see Section 1.3 for further information).

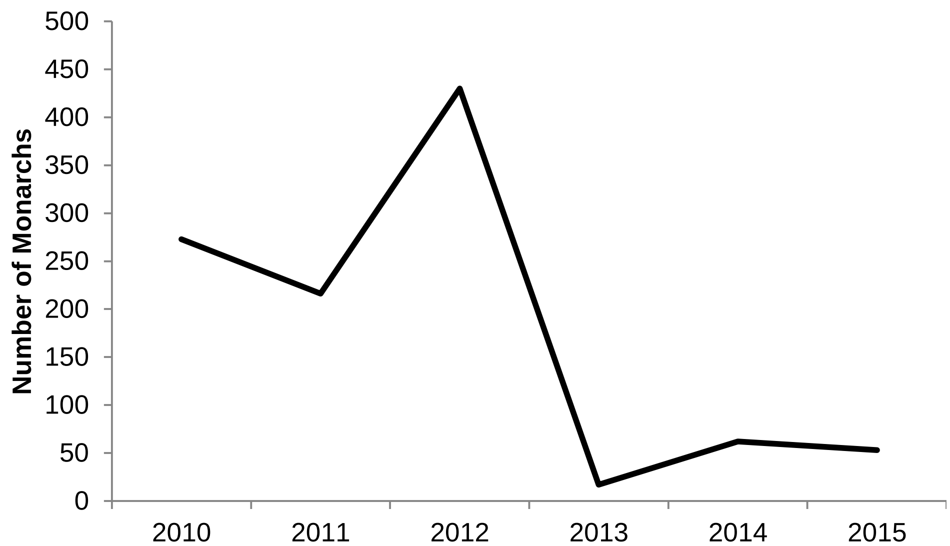


Figure 2.14: Representation of total Monarchs recorded for all transects for 2010-2015 monitoring.

2015 saw changes to total abundances of species invasive to Waterloo Region, the European Skipper and the Cabbage White. Cabbage Whites experienced their lowest ever populations on record with 779 observations accounting for 15.8% of total butterflies observed.

The highest populations of Cabbage Whites were seen in 2012 at 3557 individuals accounting for 46% of the total individuals observed. European Skippers have exhibited the opposite trend of the Cabbage White and have had their most abundant year on record (N=687) and were the second most commonly observed species, accounting for 13.9% of total observations in 2015 compared to the next highest year of 9.7% of total observations in 2014 (See Figure 2.15).

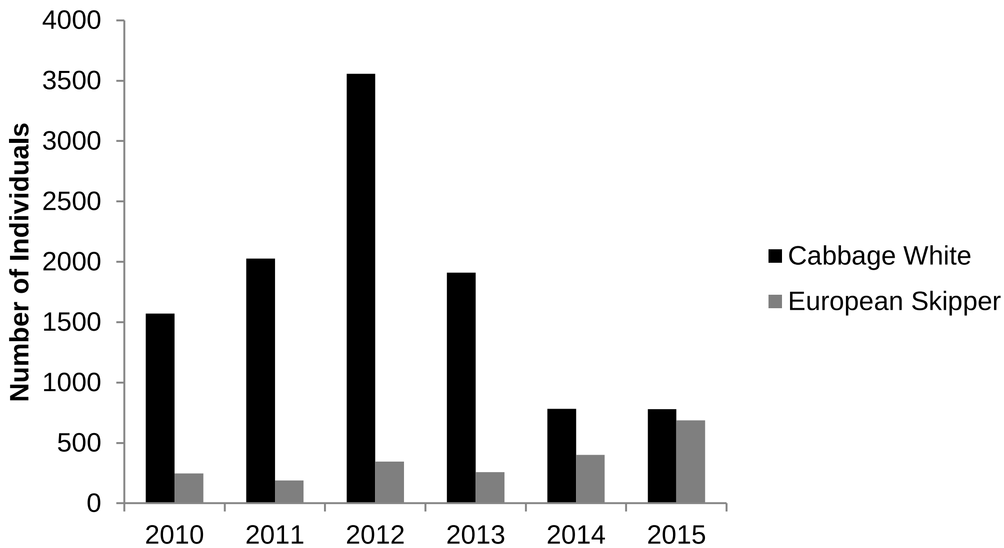


Figure 2.15: Representation of total Cabbage Whites and European Skippers observed in each monitoring year.

Four new species were added to the list of butterflies observed while conducting the monitoring program in 2015, they are; Grey Comma, Silvery Blue, Crossline Skipper, and Eastern Pine Elfin. The Crossline Skipper and Eastern Pine Elfin are both considered rare in Waterloo Region, and the Silvery Blue was not known to the region before several reported sightings in 2015 including at ***rare*** (Linton 2012; Larrivee et al. 2014 (eButterfly)). Note that the Crossline Skipper and Grey Comma have been previously observed during the annual butterfly counts.

In addition to the four species added in the monitoring program, five first observations of species were made by ***rare*** volunteer and butterfly enthusiast Julie Reid as opportunistic sightings. Julie first spotted the Silvery Blue, Eastern Pine Elfin, Dreamy Duskywing, Ocola Skipper and Indian Skipper on the property this year. The Dreamy Duskywing is considered rare in Waterloo Region, the Ocola Skipper was not previously known to this area (Linton 2012). The last confirmed sighting of an Indian Skipper in the Waterloo region dates back to 1950 (Linton 2012). These new additions bring the total number of butterfly species observed on ***rare*** property to a total of 75. This is 90% of the total number of butterflies known to Waterloo Region (81 species reported in Linton 2012 plus two new species this year).

2.3.7 Weather Conditions

Mean temperature was the highest on record in the month of May (15.2˚C) and the lowest on record for June (16.6˚C). Mean temperature for all four months was 17.4˚C, which the second coldest monitoring period overall (tied with 2013). A comparison of all monitoring years' temperature data can be found in Figure 2.16.

Precipitation was the highest ever recorded in the month of August and middling for all other months. Total precipitation was the highest of all full monitoring years at 366mm. Precipitation data for all monitoring years can be visualized in Figure 2.17.

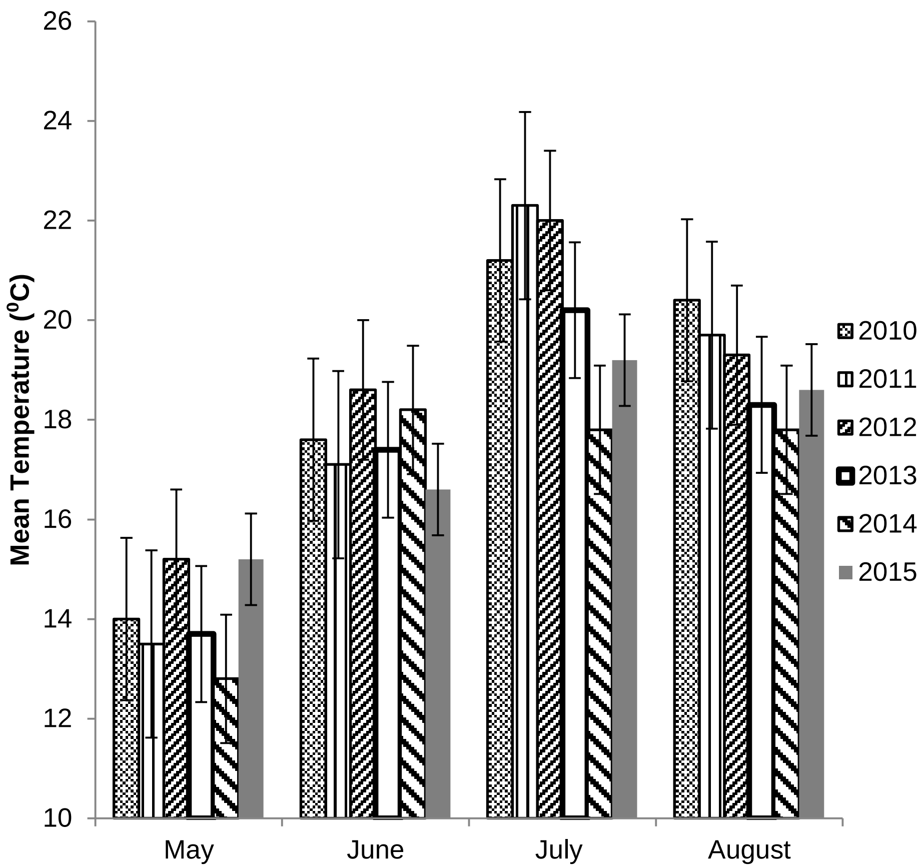


Figure 2.16: Mean monthly temperatures for Kitchener/Waterloo for all months of all butterfly monitoring seasons at the ***rare*** **Charitable Research Reserve**. Weather data for 2006 and 2009 are from the Waterloo International Airport Weather Station, and data for 2010-2015 are from the Kitchener Waterloo Weather Station (Accessed from Environment Canada 2015). Error bars represent +/- one standard error.

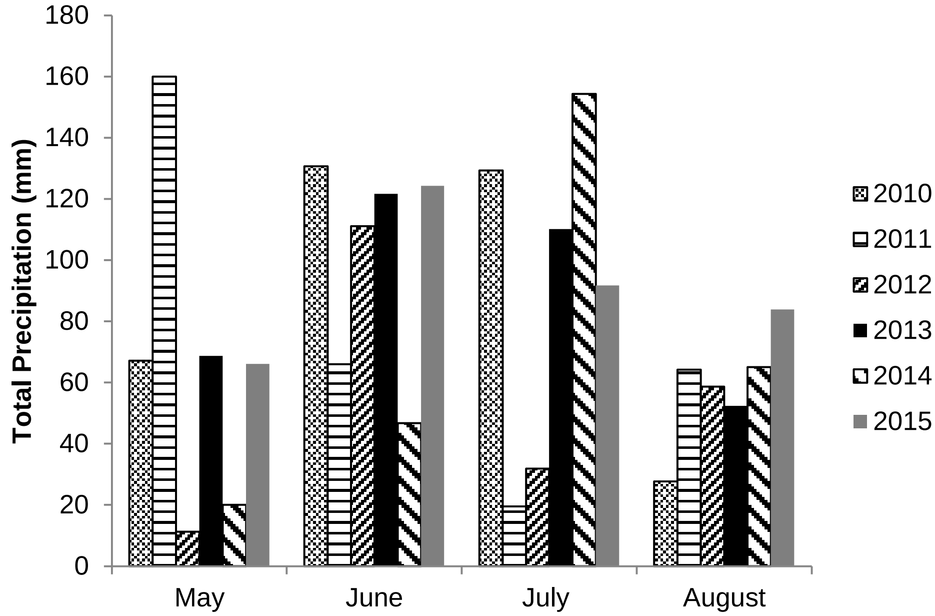


Figure 2.17: Total monthly precipitation for Kitchener/Waterloo for all months of all butterfly monitoring seasons at the ***rare*** **Charitable Research Reserve**. Weather data for 2006 and 2009 are from the Waterloo International Airport Weather Station, and data for 2010-2015 are from the Kitchener Waterloo Weather Station, with the exception of July 2015 due to missing values. July 2015 data was taken from Roseville Weather Station 43°21'13.026" N 80°28'25.056" W (Accessed from Environment Canada 2015).

2.3.8 Annual Butterfly Count

The 10th Annual Butterfly Count was held at ***rare*** on July 11, 2015. A total of 31 species and 883 individuals were observed. Data collected from the count was submitted to the North American Butterfly Association and results from the count can be found below. Data from counts in previous years can be found in **Appendix D**.

Observations: Black Swallowtail 14, E. Tiger Sw. 3, Cabbage White 247, Clouded Sulphur 162, Coral Hairstreak 1, Banded Ha. 1, E. Tailed-Blue 3, 'Summer' Spring Azure 7, Gr. Spangled Fritillary 20, Pearl Crescent 12, N. Cr. 28, Question Mark 2, Mourning Cloak 8, Red Admiral 13, Red-spotted Admiral 3, N. Pearly-eye 15, Eyed Brown 1, Little Wood-Satyr 24, Com. Wood-Nymph 212, Monarch 5, Silver-spotted Skipper 3, Least Sk. 1, European Sk. 46, Peck's Sk. 10, Tawny-edged Sk. 13, N. Broken-Dash 12, Little Glassywing 1, Delaware Sk. 4, Mulberry Wing 2, Dun Sk. 7. **Unidentified:** Polygonia spp. 3. **Field Notes:** Immatures: Giant Sw. 1 caterpillar on Northern Prickly Ash (recently predated)

2.4.0 Discussion

2.4.1 Overall Abundance and Diversity

2015 saw the third highest total butterfly abundance and the most species ever recorded in a monitoring season with 4931 individual observations. Only 2013 and 2012 had more observed individuals with 5263 and 7866, respectively. 2012 in particular had a substantial difference and the extreme abundance appears to be the cause of ideal circumstances. Butterflies are highly sensitive to shifts in local weather (Wikstrom et al. 2009). Butterfly populations respond positively to warm and dry weather by increasing their rate of development during the egg and larval stages and by having improved reproductive success (Roy et al. 2001). This would explain the high abundance in 2012 as it was both the warmest (Average Temp. = 17.8 ˚C) and driest (Total Precip. = 213 mm) on record. 2015 experienced the highest precipitation on record (Total Precip.= 366mm) and had middling temperatures (mean temp.= 17.4 ˚C), which likely yielded the mid-range butterfly abundances seen.

Another possible explanation for the abundances seen is the occurrence of population cycling. Many species of butterfly have populations that fluctuate between high and low abundance (Harrison et al. 2015). This is particularly evident at ***rare*** in species like the Red Admiral, which has consistently had observations over 100 individuals (2010, 2012, 2015) or less than 50 (2009, 2011, 2013, 2014). Populations of butterflies are known to synchronize having either high or low abundance across their range (Pollard 1991). True to this, the same trend can be seen in Red Admirals, as well as many other species, when comparing count data across Ontario. Counts in Ottawa, Windsor and Kitchener all show the same trend for Red Admirals in their data (Moore 2014; Ojibway Nature Centre; Ottawa Field Naturalist's Club, City of Kitchener). Some species had above average abundance in 2015, like Red Admirals, while others had lower than average numbers, such as Cabbage Whites, helping account for the differences observed.

While not the most abundant year for butterflies, 2015 was the most diverse. The newly observed species played an important role in making this year reach the highest species richness and are likely the result of expanding ranges. For instance, the abundance of Silvery Blue butterflies is probably due to the increasing range of non-native plants the butterfly will use as a host, such as Cow Vetch (*Vicia cracca*) (Layberry et al. 1998). A similar pattern has been observed in the Wild Indigo Duskywing, which has become more common in Waterloo Region in recent years, due to increased availability of vetch as a host plant (Layberry et al. 1998). The same phenomenon has been seen elsewhere in the world with butterflies shifting into new ranges as habitat conditions change (Diamond et al. 2014; Oliver et al. 2012).

Reponses to habitat change are most often species specific, with species who are habitat generalists or those with strong ability to disperse often best suited to adapt to the changing landscape (Oliver et al. 2012.). Species like the Silvery Blue are willing to use the non-native Cow Vetch as a host, which is actively expanding its range into disturbed areas and roadsides (OMAFRA). The interests of ***rare*** involve conservation of its natural areas and promotion of native species and while Cow Vetch is present on ***rare*** property, the more actively growing populations of the vetch are likely in the changing urban environment surrounding ***rare***. The disturbances from urban areas will provide new opportunities for plants that thrive in these types of habitats and, as a consequence, for species that will use those plants. It is then likely that more of these types of species will be seen on the property in the future just through proximity and regardless if habitat requirements are met on the property.

It must also be noted that there are changes happening on the property. Ongoing restoration efforts are taking place in many areas, in particular on or nearby Transect Two and Transect Four. In these areas, efforts have been made to promote native species (i.e. prescribed burns and plantings), which may have had a positive impact on butterfly observations. The potential impact of restoration activities on the transects is discussed at length in their respective sections of the discussion (Section 4.2 and 4.3).

Global changes in weather patterns may also have an effect on the species observed during the monitoring season at ***rare***. Parmesan (2006) discusses how increases in global temperatures may lead to shifts in the typical range of certain butterfly species. Rare species as well as species not thought to exist in this region may become commonplace. This is perhaps why the Ocola Skipper was spotted on the property in 2015. Typically a more southern species, the Ocola Skipper is a rare migrant to Ontario (Layberry et al. 1998) and a number of sightings in Ontario in 2015 suggest it may be increasing the size of its range northward (Larrivee et al. 2014 (eButterfly)).

Another possible explanation is the hot start to the monitoring period in May (average temp.=15.2˚C), which may have increased sightings of spring migrants. Early warm temperatures coupled with all the explanations given are likely acting in tandem to have caused the greatest number of species observed yet.

2.4.2 Transect One: Cliffs and Alvars

Cliffs and Alvars is the longest of the four transects, travelling through various habitat types, such as alvars, maple-beech forest and meadow areas. Transect One has undergone very little change over the course of the monitoring and, on top of containing a diverse range of habitats, the transect has a wide permanent gravel trail. This trail fragments the natural areas, which has been shown to be beneficial to species which prefer edge habitat, such as Swallowtails and Little Wood-satyrs (Siu 2014). For these reasons, it is not surprising that it has had consistently high abundance and richness throughout all monitoring periods.

Despite having high abundance and richness, most of these observations occurred in only three of the 11 sections located within the transect. More than half (50.4%) of all observations occurred in these three sections, which correspond to Floodplain and Alvar habitat, supporting an abundance and variety of butterflies. This trend is common across all years, with sections 1, 2 and 7 making up roughly 50% of total observations on Transect One. Many of the other sections in this transect, and most of the transect itself, is located within partially or fully enclosed forest canopy and thus is not ideal butterfly habitat for most species. This would explain the very low abundance seen in many of the sections of the transect.

Shannon Diversity Index values for 2015 were the highest of any monitoring year. Shannon Diversity was likely so high because of the lower than average numbers of Cabbage Whites. Fewer Cabbages Whites might create more available resources for historically less abundant species like Milbert's Tortoiseshells, Summer Azures and Juvenal's Duskywings, which all had their best years on record. The drop in Cabbage Whites then increases species evenness and as a result causes a high Shannon Diversity Index. This also explains the high

GM Index value because of the high species evenness, but the GM Index is also very sensitive to abundance and, as a consequence, the highly abundant 2012 had a higher Index.

Transect One has continuously had a positive Geometric Mean (GM) (see Figure 2.18), indicating overall positive growth in abundance and species evenness, compared to the base year of 2010. This trend is surprising given that there has been few changes to the transect since the start of the monitoring program. This may be due to natural habitat improvement over time, but another possibility is our increased knowledge of this area causing greater numbers and more species of butterflies to be noticed along the transect. However, the GM Index is only calculated from the species seen across all years, most of which are very conspicuous such as Monarchs and Swallowtails. The most likely cause of this upward trend is that Transect One had very low numbers in the base year for data, 2010. 2010 is in fact the year of not only the lowest abundance, but also the lowest richness on record, suggesting that every year since has registered growth because they were compared to a year of particularly low abundance. Continued monitoring of Transect One should prove useful in helping determine whether or not habitat for butterflies has improved here.

Figure 2.18: Representation of Geometric Mean Index compared to the base year of 2010 for Transect One.

2.4.3 Transect Two: South Field/Sparrow Field

At 989 individual observations, 2015 is the year of the lowest abundance on record for Transect Two. While Transect two is the second longest transect, in comparison to Transect One and Three it is relatively poor in terms of habitat variety. Changes in this transect in 2014 have further diminished the number of habitats covered within this transect (see section: 2.2 Transect Descriptions). Many of the habitats that do exist within the transect are areas of high disturbance or areas poor in plant species richness (i.e. agricultural fields and old field borders). This has resulted in a fewer number of both species and individuals observed here, as it does not meet the habitat requirements for a number of species seen elsewhere on the property. With that being said, the species which are supported here, in particular Inornate Ringlets, Common

Wood Nymphs and Northern and Pearl Crescents, were in abundance, especially compared to previous years.

Shannon Diversity Index for Transect Two was the highest of any monitoring year. Although number of individual observations was low, richness and evenness were high resulting in a high Shannon Diversity Index. Similar to Transect One, Cabbage Whites have had their lowest abundance ever, potentially leading to increased opportunities for other species. This may then explain why 2015 also saw the highest number of recorded species, as a result of increased resource availability. However, the GM Index scored low compared to the previous three years. This suggests abundance and evenness of the most productive species to the ecosystem is falling. As mentioned, the Cabbage White has had a particularly low year, but so did Clouded Sulphurs and Monarchs, which were some of the most productive species in 2010. It must be noted that the overall trend of the GM Index indicates increased abundance and evenness for all years since 2010 (See Figure 2.19).

One possibility for the recent decline in GM Index could be the change in habitat in and around the transect over time. For instance, Sparrow Field was used for agriculture up until 2011. Starting in 2012, the field was allowed to naturalize and was partially mowed for research purposes. 2015 was the first year in which the field was not managed, providing the most available habitat for the butterflies since the start of the monitoring program. Transect Two also undergoes dramatic changes year to year from the impact of agriculture. South Field West is harvested each year during the butterfly monitoring period for use as feed, while South Field East is on a three year corn, soy, winter wheat rotation. There does not appear to be any correlation between crop type and butterfly abundance. The sections of the transect located on South Field East (section two, three and four) have typically accounted for 30-35% of the observations on Transect Two, with the exception of 2014 where 47% of all observations were seen in these sections. Given that there is only two years of data for each crop type, it is not possible to determine if one type is more beneficial to butterflies and there is a lack of research in the literature examining this topic. This issue is further complicated by the adjacent fields to the transect, which are on a different rotating schedule. For instance, the field adjacent to South Field East was planted with soy in 2015. While this is a difficult problem to isolate, it is generally agreed that agriculture is detrimental to most butterfly species, namely from the loss of habitat and the use of pesticides (Merckx et al. 2013; Pleasants and Oberhauser 2012).

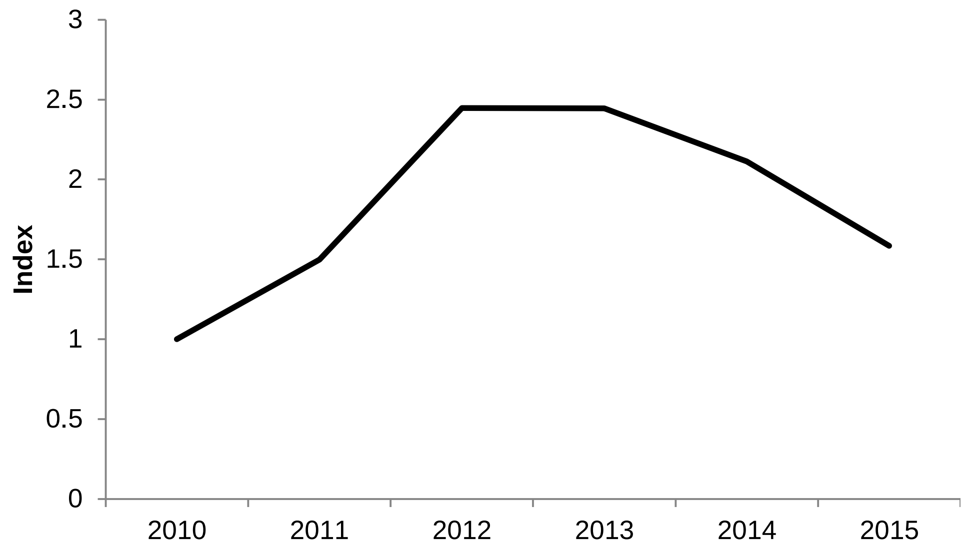


Figure 2.19: Representation of Geometric Mean Index compared to the base year of 2010 for Transect Two.

Being the second year of walking the modified transect route, comparisons can start to be made of the influence the route changes have caused. The total number of butterflies observed at this transect in 2015 was the lowest it has been since 2009 (N=717). In past years, a fairly large portion of the number of total butterflies were observed along both section six and seven (18.5% of total observations between 2010 and 2013) while the relative number of observations in the new section six/seven has so far shown to be of less weight (11.5% of observations in 2014 (N=130) and 13% in 2015(N=127)). Section five was also altered by the change in 2014. Between 2010 and 2013 the average percent total observations on section five was 9.75% compared to 10.5% in 2014 and 9.3% in 2015. Overall the changes from the shortened section five seem minimal in comparison to the change in sections six and seven.

Another factor of note is the nearby restoration efforts that have taken place along the edges of Indian Woods and the nearby Hogsback. These areas should provide habitat for forest edge species and may have encouraged the high species diversity seen. The expectation would be that the restoration efforts as well as the naturalization of Sparrow field would result in not only greater numbers of species, but also higher abundance, however this was not the case in 2015. Given the data here, the most prominent cause of falling numbers at Transect Two appears to be the shortening of the transect rather than changes in the butterflies environment. Future years of monitoring should examine this issue and should note whether populations increase from the data gathered in 2014 and 2015.

2.4.4 Transect Three: Thompson Tract

For the first time on record, Transect Three had the highest abundance of any transect. This transect offers a diversity of habitats, including a number of different forest types and a meadow area which provide opportunities for a variety of species. In 2015, much of the abundance within the transect occurred along section four (N=635 or 35% of total observations). This section is adjacent to a highly productive meadow with forested borders and a small wetland area, providing both refuge and food plants for larval and adult butterflies alike. These numbers are considerably higher than in previous years (9% of total observations in section four in 2012, 16% in 2013, and 22% in 2014) with an upward trend, suggesting that more and more butterflies are using this area productively. Similar to Transect One, Transect Three has undergone little change throughout the monitoring program and so observed changes should not be attributed to anthropomorphic enhancement or restoration of the area.

In all metrics (Shannon Diversity, Evenness, Geometric Mean) 2015 had the highest values for this transect. Gradual upward trends are seen in Shannon Diversity and Evenness and an overall positive trend is seen in the GM Index (See Figure 2.20). This indicates butterfly diversity and abundance appear to be increasing in this transect over time as more butterflies are drawn to this area. It may be these trends are from improved habitat quality for butterflies which could be occurring naturally or a lack of nearby habitat making this a refuge for butterflies. More likely than these reasons seems to be changes in how the butterflies are detected.

Every new species seen during monitoring was observed in Transect Three. Some of these species, like the Silvery Blue, are probably entirely new species to the area. However, given that most of the new species observed were small and inconspicuous, there is a strong possibility many of these species have been overlooked in past monitoring years. New species this year may have been noticed because of our increased ability to observe species from our accumulated knowledge of the area. This is particularly true for skippers.

There was a greater number of different species and abundance of skippers (*Hesperiidae*) seen in 2015 than any other year. Over time, the data shows a general upward trend in both species richness and abundance for skippers. Many skippers are small, brown, inconspicuous and often difficult to distinguish from one another. While there may in fact be more species of skipper present along Transect Three, given that the transect itself has changed little over the course of the monitoring program, it is more likely this change has occurred from changes in the observer's knowledge. Unsurprisingly, this is a known phenomenon, with an observer's prior knowledge influencing data gathered through monitoring (van Swaay et al. 2008). This bias is unavoidable as the knowledge of Transect Three grows between each monitoring period. While the knowledge should give a more accurate data set for Transect Three it will make interpretation of the data more difficult to determine if changes between years are real or solely based on gaps in knowledge. The bias may be minimized as observers are changed between each year due to the short-term nature of the position that performs ecological monitoring, but the access to data from previous years may still influence their decision making on difficult identifications. This is an issue for monitoring across all transects, but the problem seems most pertinent for Transect Three given the abundance of species that are difficult to detect and identify.

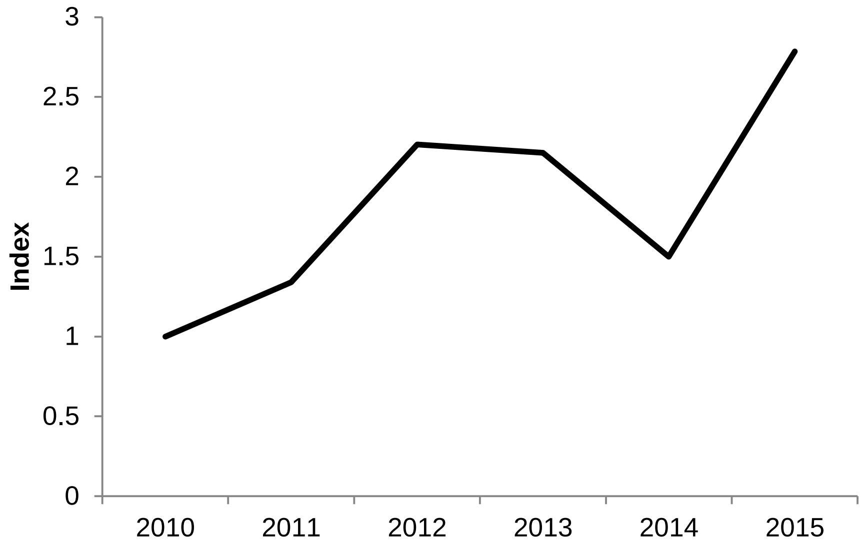


Figure 2.20: Representation of Geometric Mean Index compared to the base year of 2010 Transect Three.

2.4.5 Transect Four: Blair Flats

Over the course of butterfly monitoring at ***rare***, Transect Four has constantly been changing as it transitions from an old agricultural field to a native tall grass prairie. This past spring Blair Flats was subjected to a controlled burn in an effort to further restore the field to a tall grass prairie by simulating a wild fire.

This transect has had the lowest abundance and species richness of all transects over all monitoring years. This is due, in part, to the comparatively small size, length of this transect and the historically species poor old field habitat. However, the species evenness and Shannon Diversity Index have increased dramatically since the start of monitoring in Blair flats in 2010 (See Table 2.2). This suggests that the relative abundance of species as well as the overall biodiversity within this transect seems to be improving. An overall upward trend is seen in the Geometric Mean Index as well (Figure 2.21).

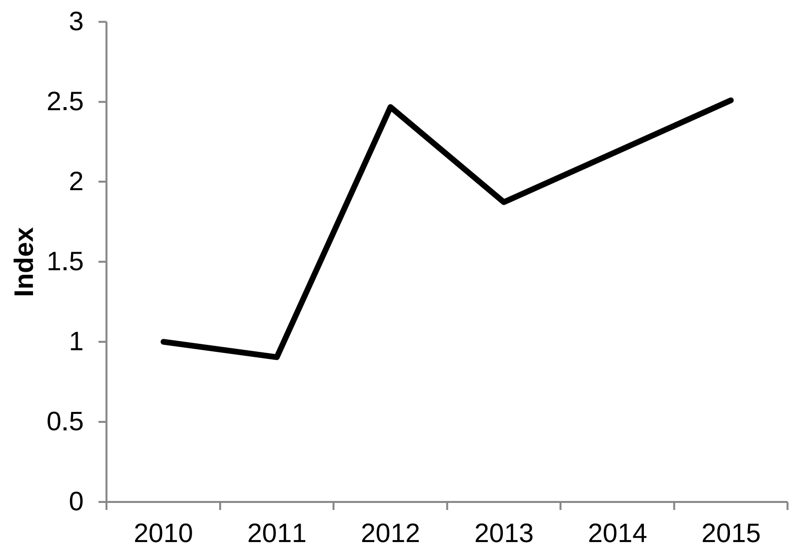


Figure 2.21: Representation of Geometric Mean Index compared to the base year of 2010 Transect Four.

While the burn appears to have had a positive impact on butterflies in Transect Four, the impact of prescribed burns on butterflies and invertebrates in general is contested. There is no consensus as to whether the butterflies are positively or negatively impacted and response to fire is typically species specific (Panzer 2002; Vogel et al. 2007; Vogel et al. 2010). Overall it seems there is a tendency for species to perform better in the years following a burn rather than the year of the burn (Vogel et al. 2010). Only 80% of the prairie at ***rare*** was burned, leaving 20% unburned as a refuge for species, particularly to avoid complete elimination of any larval stages inhabiting the area. Providing this refuge may have resulted in an avoidance of the trends observed by Vogel et al. (2010), however only continued monitoring will determine the long term effects of this field restoration on the butterfly and other pollinator communities. Given the abundance found in 2015 for Transect Four, it will be interesting to see how numbers will change in subsequent years.

2.4.6 Comparison of Geometric Means and Shannon Diversity

For all transects, except Transect Four, 2015 calculations of Shannon Diversity were the highest for all years. Geometric Mean Index calculations did not register the same trend, instead showing only Transect Three and Four having the highest values. Trends in Shannon Diversity exhibit a positive trend over time and trends in Geometric Mean, while being overall positive, were not nearly so uniform. This indicates that butterflies in Transect One and Two appear to be increasing in terms of species richness and evenness, but not in abundance. Transect Four is becoming more abundant, but not as species rich than it has been before and Transect Three appears to be increasing in richness, number of individuals, and evenness.

The differences seen in the two metrics are largely attributable to the how each is calculated. This difference can be best exhibited by the calculations from Transect Three (See Table 2.3). Species evenness is highly consistent across all years in Transect Three; however richness and abundance are not. Shannon Diversity will fail to register these changes in abundance from year to year if evenness is more or less consistent. The GM Index is a calculation of both abundance and evenness and this is reflected in the results of the calculations. However, the GM Index is only calculated from a portion of the total butterflies. Only butterflies present across all years were used in the calculation as the Geometric Mean is highly sensitive to rarely recorded species (Buckland et al. 2011). Therefore the Geometric Mean is not as representative of all of the species within the population as the Shannon Index, but the species included are the most abundant and, as a consequence, the most impactful on the communities they inhabit (Buckland et al. 2011). Both metrics have shortcomings that need to be kept in mind when interpreting the results, but both metrics have value in helping inform decision making and thus should be used in conjunction to help make future management decisions.

Table 2.3: Number of individuals, Species Richness, Species evenness, Shannon Diversity and Geometric Mean Index calculations for Transect Three across all years.

		Number of Individuals (n)	Species Richness (S)	Species Evenness (E)	Shannon-Diversity Index (H)	Geometric Mean Index (G)
Transect Three	**2010**	938	30	0.70	2.37	1
	2011	911	35	0.72	2.56	1.34
	2012	2116	38	0.71	2.56	2.2
	2013	1636	36	0.71	2.55	2.15
	2014	1354	38	0.72	2.62	1.5
	2015	1834	44	0.73	2.75	2.78

2.4.7 Species of Interest and Species of Special Concern

There are two species of non-native butterfly abundant within Waterloo Region, the Cabbage White and the European Skipper, and both have had interesting trends across monitoring years. This year the Cabbage White had its lowest abundance year on record. Furthermore, Cabbage Whites had only a fraction of the proportion of total observations seen in previous years (See Figure 2.22). Originally from Europe, the Cabbage White was introduced to Canada in the 1860's in Quebec. As with other invasive alien species, it spread rapidly, quickly out-numbering native butterflies across many areas of Canada (Layberry et al. 1998). Part of its advantage is that it can use many habitat types, and has a lengthy flight season (Layberry et al. 1998). This butterfly's required host plants belong to the Mustard Family, including another non-native invasive species, Garlic Mustard (*Alliaria petiolata*). This plant is widespread throughout the ***rare*** property (***rare*** Environmental Management Plan 2014; Robson et al. 2012), and has provided above average host plant availability to the Cabbage White butterfly.

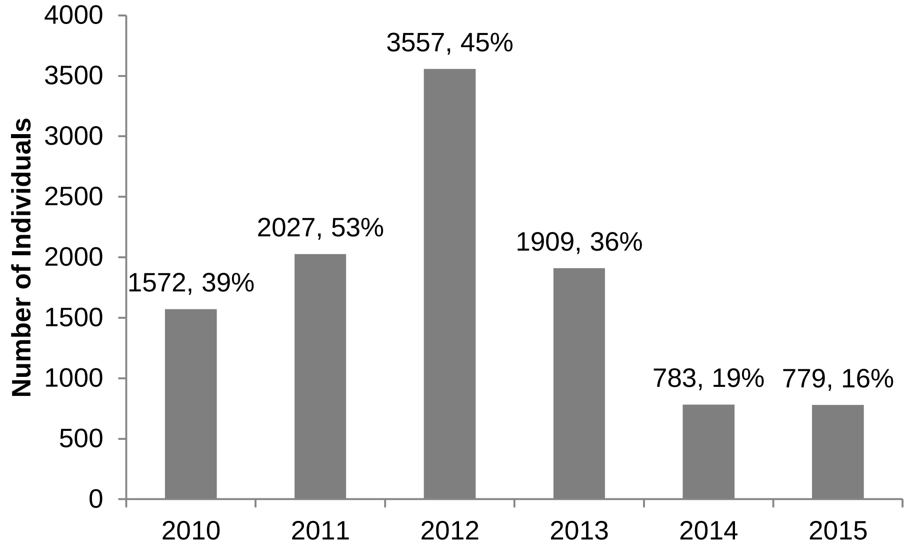


Figure 2.22: Total number of Cabbage Whites seen in each year of butterfly monitoring. Number of Individuals observed followed by the percent of total observations Cabbage Whites accounted for in that year are displayed above each bar.

Despite still being the most common species observed, in 2015 the Cabbage White had the lowest numbers recorded since 2010. The Cabbage White has consistently been the most common butterfly observed since the induction of the monitoring program, but overall numbers have declined sharply in 2014 and 2015, compared to previous years. This downward trend may be indicative of a number of things. Weather likely played a significant role in curbing Cabbage White populations early in the season in 2014 and 2015, as mean May temperatures were the lowest on record in 2014 and there were late frost dates in 2015 (May 22,23) (Environment Canada). The other possibility is the shift in habitat or vegetation cover. However, the notion that Cabbage Whites have less potential host plants seems unlikely, due to the overall rise in potential hosts, such as Garlic Mustard. Further investigation is needed into the host plants of Cabbage Whites and their populations on the property to see if this is impacting their abundance.

In 2014, 50 Cabbage Whites were collected for research purposes. While this may have had an impact on the populations seen in 2015, given the ubiquity of Cabbage Whites and the relatively small number collected this seems unlikely. In all likelihood, this was a poor year for Cabbage Whites and subsequent years will attain numbers seen in previous years, especially given their long establishment in this region. Further monitoring of this trend is needed.

The opposite trend is seen in the European Skipper which had its most abundant year ever in 2015 (See Figure 2.15). The European Skipper is another invasive species that has been present in Canada for quite some time. Original introduction of the species occurred near London Ontario around 1910 in seed for livestock feed (Layberry et al. 1998). Since that time, the European Skipper has attained incredibly abundant populations throughout Ontario, aided by their fondness for using many common invasives as host plants, such as Timothy Grass (*Phleum pratense*), Orchard Grass (*Dactylis glomerata*) and Quackgrass (*Elymus repens*) (Layberry et al. 1998). European Skippers in Waterloo Region tend to last about five weeks between early June and mid-July (Layberry et al. 1998). Given that this June had the lowest temperatures of any monitoring season as well as late frost dates in May make it unlikely weather was the cause of the high populations in 2015.

Another possible explanation is the increase of available habitat, but the host plants of the European Skipper are both widespread and abundant and have been for quite some time in Ontario. Most likely the abundance seen in 2015 is from fluctuations in the population as the European Skipper is known to reach very high densities in some years (Blatt et al. 2005). This trend is also present in data gathered during monitoring at ***rare*** and in other counts across Ontario (Moore 2014; Ojibway Nature Centre; Ottawa Field Naturalists Club). Therefore, the population may continue to grow then crash over the next monitoring years. Further attention to this trend is needed in the future to determine the cause.

Monarchs are a species of special concern in Ontario due to their falling populations in recent years. Total Monarch observations in the past three monitoring periods have fallen under 100 individuals, while before this all years have registered more than 100. Although Monarchs are considered very common in Waterloo Region, their declining population trends constantly catch the attention of ecologists and the general public alike. The Monarch is listed as a species of Special Concern in Ontario (Government of Canada 2014), and their North American migration is in peril from a number of causes including; deforestation in their overwintering grounds, land development, pesticide use, climate change, and the loss of their host plant, Milkweed (*Asclepidaceae*) (Brouwer et al. 2011). Recent research by Flockhart et al. (2014) has suggested the most critical cause of their decline is caused by the loss of Milkweed and until this past year, Ontario had Common Milkweed (*Asclepias syriaca)* on its list of noxious weeds (OMAFRA). Large numbers of the plant have been removed not only in Ontario, but throughout the United States, where some states still consider this plant a noxious weed (OMAFRA). The Monarch is vulnerable as it requires adequate numbers of food plants as it makes its way north in each successive generation throughout the summer (Pleasants and Oberhauser 2012).

Efforts have been made at ***rare*** to increase viable habitat for Monarchs, mainly through the seeding of Milkweed. In 2015, a Milkweed survey took place, which identified areas with existing Milkweed populations on the property and estimated the abundance of Milkweed stems at these sites. Seeding of Milkweed and other nectaring plants occurred across three acres within these designated areas and will be re-evaluated in future years. It remains to be seen whether the plants will have any impact on the population of Monarchs seen at ***rare.*** Results from the Milkweed Survey can be seen in Section F of the appendix and the full report can be found on the ***rare*** server.

2015 was also the year of new sightings inside and outside of usual monitoring. Most notably was the Silvery Blue, which was seen in some abundance on Transect Two and Three. The Silvery Blue is not a species known to the Waterloo area (Linton 2012), but changes in the availability and range of one of its non-native host plants, such as Cow Vetch (*Vicia cracca*) or White Sweet Clover (*Melilotus alba*) have likely facilitated this change (Layberry et al. 1998). It remains to be seen if the Silvery Blue has permanently established in Waterloo Region. The Eastern Pine Elfin was also a new addition to monitoring this year. The species is known to Waterloo Region, but only two records before this year exist from 1997 and 2010 (Linton 2012). Unsurprisingly, the Pine Elfin caterpillar feeds on White Pine (*Pinus strobus*), which is present on ***rare*** property (Linton 2012). The single observation during monitoring this year, along with incidental observations of the elfin, suggests this species may become more common in Waterloo Region and at ***rare*** in the future. Given these trends, it is very possible more and more species not known to this area will expand their ranges into Waterloo Region over time.

2.4.8 Comparison with Baseline Data

In order to accurately identify trends and averages for populations, EMAN protocol suggests the first five years of data collection be used to create a baseline for monitoring programs. 2013 was the fifth year that butterfly monitoring at ***rare*** took place for either 13 or 14 weeks, and thus data from these years were used to identify averages and standard deviations for both abundance and species richness across the four transects (Table 2.4).

Using these data, we can compare the 2015 results to the averages for each transect to determine if this monitoring season fell within or outside of these averages. Values that are outside of the given ranges may indicate environmental change that has potentially had either positive or negative impacts on the populations. Monitoring results are heavily dependent on local weather patterns which vary from year to year, meaning a wide range of values are considered acceptable, as seen by the standard deviations reported with each average.

Table 2.4: Mean butterfly abundance and species richness, with standard deviations, for monitoring seasons 2009-2013.

Transect	Number of Individuals		Species Richness	
	Average	Standard Deviation	Average	Standard Deviation
Transect One	1491	+/- 825	36	+/- 8
Transect Two	1563	+/- 655	30	+/- 6
Transect Three	1203	+/- 670	35	+/- 3
Transect Four	361	+/- 101	23	+/- 9

The number of individuals in Transects One, Two, and Three all are considered average, as they fall within the average range of the baseline data. The total number of individuals at Transect Four was above average ranges by 56 individuals (total individuals was 518). This is likely as a result of ongoing restoration efforts (discussed in section 4.5).

Transect One and Four fell within average ranges for species richness while Transect Two and Three were above average at 38, and 44 species, respectively. Causes for this increased species richness are speculated to be from declines in numbers of Cabbage Whites, or shifting ranges of new species (see section 4.7) or improvement of butterfly habitat variety. As previously noted, further monitoring of this trend is needed.

2.5 Conclusions and Recommendations

The main driver of change in this season compared to others was the difference in Cabbage White populations. Cabbage White numbers have heavily impacted calculations of evenness and abundance in previous years and have played a major role in the butterfly monitoring. While still being the most abundant species in 2015, the proportion of Cabbage Whites has dropped dramatically, resulting in increases in calculations of evenness and Shannon Diversity across the board. This drop in Cabbage Whites appears to have benefitted the other butterfly species on the property as many of their abundances have increased, perhaps to ‘fill the gap’ of unused resources left by Cabbage Whites. As speculated earlier in the discussion, this drop in Cabbage White numbers is likely temporary and a natural dip in population that will recover over time. Monitoring of this trend should continue to determine if it is permanent or temporary.

The 2015 season was also characterized by new species resulting in the highest richness seen over any monitoring year. Part of this high richness is attributable to the instances of rare and never before seen species in the region. This is likely telling of how butterfly populations on ***rare*** property will change in the future with more and more species not thought to exist or to have established populations in the region turning up. While it may be exciting to see more species of butterfly, it may be at the expense of species commonly found on the property now. Should issues in populations of historically abundant species dramatically decline as a result of encroaching species of butterfly, it may be prudent to develop recovery strategies for these species, such as food plant seeding.

The most promising trend of 2015 was the high abundance and species richness seen on Transect Four. Given the ongoing restoration of the field in Blair Flats (i.e. controlled burning) it appears the efforts have been successful in promoting butterflies in the area. Further restoration activities may promote more butterflies and careful monitoring of this trend could prove fruitful.

Another prime candidate for promoting butterfly habitat is Transect Two, in which the current population of butterflies is under heavy pressure on all sides from agricultural activity. Cessation or scaling back of farming activity on ***rare*** property is underway and plans are made to convert South Field East to hay in 2016. Sparrow field could also be a candidate for a restoration project as the field has previously been managed as a research project and plants along the edges of the field are largely non-native.

It is recommended that the monitoring program at ***rare*** continue in its full capacity in the years to come. With a constant urban growth surrounding the ***rare*** property, including new subdivisions, increased vehicle traffic, and continued aggregate mining, the butterfly monitoring program will play a key role in detecting changes in ecosystem health. Identifying potential issues early on will also allow for further creation and implementation of management plans for the property. The data collected during butterfly monitoring at the ***rare*** **Charitable Research Reserve** continue to be useful on a broader scale, adding to the knowledge of environmental health and species within the Region of Waterloo as a whole.

2.6.0 Literature Cited

Blatt, S.E., Crowder A., and Harmsen, R. 2005. Secondary succession in two south-eastern Ontario old-fields. *Plant Ecology*, 177:25–41.

Brereton, T.M., Botham, M.S., Middlebrook, I., Randle, Z. Noble, D.G. and Roy, D.B. 2014. United Kingdom Butterfly Monitoring Scheme report for 2013. *Centre for Ecology and Hydrology and Butterfly Conservation.*

Buckland, S.T., Magurran, A.E., Green, R.E., and Fewster, R.M. 2005. Monitoring change in biodiversity indices through composite indices*. Phil. Trans. R. Soc. B.,* 360:243-254.

Buckland, S.T. Studeny, A.C., Magurran, A.E., Illian, J.B., and Newson, S.E. 2011. The geometric mean of relative abundance indices: A biodiversity measure with a difference. *Ecosphere,* 2(9):100.

City of Kitchener. 2015 Butterfly Monitoring Results. Provided by Joshua Shea from the City of Kitchener. Available on ***rare*** server.

Craig, S., Damstra, E., Kelly, P., Leather, P., Marko Dunn, J., McDonald, L., McLeod, K., Moore, R., Pope C., Quinn, Reinert, J., Robson, L., Sobek-Swant, S., Whelan, S., Woodcock, T. 2014 . ***rare*** Environmental Management Plan. Available on ***rare*** server.

Diamond, S.E., Cayton, H., Wepprich, T., Jenkins, C.N., Dunn, R.R., Haddad, N.M., and Ries, L. 2014. Unexpected phenological responses of butterflies to the interaction of urbanization and geographic temperature. *Ecology*, 95(9):2513-2521.

Flockhart, T.D.T., Pichancourt, J-B., Norris, D.R., and Martin, T.G. 2014. Unravelling the annual cycle in a migratory animal: breeding-season habitat loss drives population declines of monarch butterflies. *Journal of Animal Ecology*, 1-11.

Grealey, J. 2006. EMAN Butterfly Report: 2006. Available on ***rare*** server and from http://raresites.org/pdf/research_reports/research_EmanButterflyFinalreport2006.pdf.

Grealey, J., Stricker, F.A., Campbell, C. and Lamb, L. 2010. The Status of Butterflies in the Region of Waterloo. Available on ***rare*** server.

Hall, P.W. 2009. *Sentinels on the wing: The status and conservation of butterflies in Canada*. NatureServe Canada. Ottawa, Ontario.

Harrison, J.G., Shapiro A.M., Espeset, A.E., Nice, C.C., Jahner, J.P., and Forister, M.L. 2015. Species with more volatile population dynamics are differentially impacted by weather. Biol. Lett. 11: 20140792.

Kallioniemi, E.P. 2013. Effects of morphology, habitat and weather on the movement behaviour of range-expanding butterfly species. *University of East Anglia*. Thesis.

Layberry, A.L., Hall, P.W., and Lafontain J.D. 1998. The Butterflies of Canada. University of Toronto Press, Toronto, Ontario.

Larrivee, M., Prudic, K.L., McFarland, K., and Kerr, J. 2014. eButterfly: a citizen-based butterfly database in the biological sciences. Retrieved From: http://www.e-butterfly.org, Oct 21, 2015

Linton, J.E. 2012. The butterflies of Waterloo Region: An annotated checklist, a species scarcity ranking, and a transect-based analysis of urban vs. rural populations. *Toronto Entomologist's Association*. Toronto, Ontario.

Magurran. A.E. 2004. Measuring biological diversity. *Blackwell Publishing*, Malden, MA.

Merckx, T., Huertas, B., Basset, Y., Thomas, J. 2013. A global perspective on conserving butterflies, moths and their habitat. *Key topics in conservation biology*. 1:240-257.

Moore, C. 2014. Testing the effectiveness of citizen science using a volunteer butterfly monitoring program. *University of Waterloo*. Thesis.

Ojibway Nature Centre. 2013. Windsor Butterfly Count. Retrieved From: www.ojibway.ca/naba.html, Oct 16,2015.

Oliver, T.H., Thomas, C.D., Hill, J.K., Brereton, T., Roy, D.B. 2012. Habitat associations of thermophilous butterflies are reduced despite climatic warming. *Global Change Biology*, 18 (9): 2720-2729.

Ontario Ministry of Agriculture, Food, and Rural Affairs (OMAFRA). Weeds Profile – Milkweed spp.
Retrieved From: http://www.omafra.gov.on.ca/english/crops/facts/info_milkweed.htm, Oct. 6, 2015.

Ontario Ministry of Agriculture, Food, and Rural Affairs (OMAFRA). Ontario Weeds: Tufted Vetch.
Retrieved From: http://www.omafra.gov.on.ca/english/crops/facts/ontweeds/tufted_vetch.htm, Oct. 21, 2015.

Ottawa Field Naturalists Club. 2015. Annual Ottawa area butterfly count results. Retrieved From:
www.ofnc.ca/butterfly-count/NABAcount.html, Oct 16, 2015.

Panzer, R. 2002. Compatibility of prescribed burning with the conservation of insects in small, isolated prairie reserves. *Conservation Biology*, 16(5):1296-1307.

Parmesan, C. 2006. Ecological and Evolutionary Responses to Recent Climate Change. *Annual Review of Ecology, Evolution, and Systematics*, 37:637-669.

Pleasants, J.M., and Oberhauser K.S. 2012. Milkweed loss in agricultural fields because of herbicide use: effect on the monarch butterfly population. *Insect Conservation and Diversity*, 10(1): 1752.

Pollard, E. 1977. A method of assessing change in the abundance of butterflies. *Biological Conservation*, 12(2):115-134.

Pollard, E. 1988. Temperature, rainfall and butterfly numbers. *Journal of Applied Ecology*, 25:819-828.

Pollard, E. 1991. Synchrony of population fluctuations: The dominant influence of widespread factors on local butterfly populations. *Oikos* 60(1):7-10.

Pollard, E., and Yates, T.J. 1993. Monitoring butterflies for ecology and conservation. *London, United Kingdom, Chapman and Hall*.

Pyle, R.M. 1981. National Audubon Society Field Guide to North American Butterflies. *Alfred A. Knopf, New York, USA.*

Robbins, R.K. and Opler, P.A. 1997. Butterfly diversity and a preliminary comparison with bird and mammal diversity*. Biodiversity II*, 78-82.

Roy, D.B., Rothery, P., Moss, D., Pollard, E., and Thomas, J.A. 2001. Butterfly numbers and weather: Predicting historical trends in abundance and future effects of climate change. *Journal of Animal Ecology,* 70:201-217.

Siu, J. 2014. Testing for positive edge responses in fragmented landscape in the Eastern Tiger (*Papilio glaucus*) and the Spicebush (*P. Troilus*) Swallowtail butterflies. *University of Western Ontario.* Thesis.

Van Swaay, C.A.M., Nowicki, P., Settele, and van Strien, A.J. 2008. Butterfly monitoring in Europe: methods, applications and perspectives. *Biodiversity Conservation*, 17:3455-3469.

Vogel, J.A., Debinski, D.M., Koford, R.R., Miller, J.R. 2007. Butterfly responses to prairie restoration through fire and grazing. *Biological Conservation*, 140:78-90.

Vogel, J.A., Koford, R.R., and Debinski D.M. 2010. Direct and indirect responses of tallgrass prairie butterflies to prescribed burning. *J Insect Conserv.*

Wikström, L., Milberg, P., and Bergman, K. 2009. Monitoring butterflies in seminatural grasslands: diurnal variation and weather effects. *Journal of Insect Conservation,* 13:203-211.

3.0 Plethodontid Salamander Monitoring

Prepared by: Tim Skuse

3.1Introduction

3.1.1 Salamander Taxonomy

Ontario is home to salamanders representing four different families (Proteidae, Salamandridae, Ambystomatidae, and Plethodontidae), of which two families are known to be present at ***rare***. The mole salamanders (Ambystomatidae) are large burrowing salamanders with an aquatic juvenile phase and a terrestrial adult phase (Conant and Collins 1998). Members of this family such as Yellow-spotted salamanders (*Ambystoma maculatum*) and Blue-spotted salamanders (*A. laterale*) are occasionally observed at ***rare***. Members of the *jeffersonian-laterale* complex are also present on the property. An additional report on the occurrence of these species can be found on the ***rare*** server.

The lungless salamanders (plethodontids) are the most frequently observed salamander family at ***rare.*** Primarily observed are Eastern Red-backed Salamanders (*Plethodon cinereus*), with occasional sightings of Four-toed Salamanders (*Hemidactylium scutatum*). Plethodontids are the largest family of salamanders worldwide representing 27 genera and over 370 recognized species (Larson et al. 2006). These salamanders are generally long and slender and are lungless, breathing through their thin, moist skin (Behler and King 1979). This reliance on cutaneous respiration across moist body surfaces makes plethodontid salamanders particularly sensitive to environmental changes in their micro-habitat (Zorn et al. 2004). Gas exchange requires skin to be moist (Welsh and Droege 2001) resulting in high absorption rates potentially exposing the salamander to contaminants in the soil.

The Eastern Red-backed Salamander is the most abundant plethodontid in Eastern Canada (Zorn et al. 2004) and at ***rare***. They are completely terrestrial and therefore do not require ponds or vernal pools for development. They can generally be found in moist soil under downed woody debris in mature forests (Conant and Collins 1998). There are two main colour phases of the Eastern Red-backed Salamander- a red-backed morph that has dark grey sides and a rough edged red stripe down the back, and a lead-backed morph that lacks the red stripe and is entirely grey.

3.1.2 Global Amphibian Decline

It is estimated that one-third of all amphibian species worldwide are endangered or threatened with extinction (Stuart et al. 2004). Amphibians experience both aquatic and terrestrial stressors, and therefore are uniquely valuable as indicators of environmental stress. As such, there is significant concern over the noted amphibian declines world-wide; however, the causes of such declines are still largely undecided, and are seemingly both variable and context dependent (Blaustein and Kiesecker 2002, Caruso and Lips 2012). Alford and Richards (1999) suggest decline of amphibian populations are a global problem with local complex causes. Habitat destruction and alteration, global climate change, diseases, contaminants, and introduced species are all examples of such causes that have likely contributed to this global decline (Blaustein and Kiesecker 2002, Hof et al. 2011, Bruhl et al. 2013). Given the difficulty in neutralizing or reversing these threats, the future for amphibians around the world is seemingly bleak (Beebee and Griffiths 2005).

3.1.3 Plethodontid Salamanders as Indicator Species

Woodland plethodontids, which complete their entire life cycle on the forest floor, are useful indicator species for a forested ecosystem (Welsh and Hodgson 2013). This is due to their life history traits, sensitivities to anthropogenic stresses, and population sampling properties (Zorn et al. 2004).

Under normal conditions, plethodontid salamanders typically have stable population sizes due to long life spans (10+ years), high annual survivorship, and low birth rates. They have small home ranges (13m^2 for males and juveniles and 24m^2 for females (Kleeberger and Werner 1982)) and display site fidelity, with some species exhibiting occasional territorial behaviours (Peterson et al. 2000; Maerz and Madison 2000). Due to these traits, observed changes in population from long-term monitoring are more likely to be indicative of ecosystem stresses than typical home range shifts or population fluctuations. The role of plethodontid salamanders in the forest ecosystem is an important one. They are efficient predators and quickly metabolize insect and other invertebrate prey, which can result in plethodontid densities equalling or surpassing other vertebrate groups (Butron and Likens 1975). These high densities provide an ample food source for predators such as snakes, rodents, and birds. Their role, therefore, in transferring energy up trophic levels is invaluable (Zorn et al. 2004). Walton's 2013 study supports a hypothesized top-down regulatory role of plethodontid salamanders in the terrestrial detrital food web. As predators of invertebrate species that have substantial impact on decomposition and nutrient cycling on the forest floor, plethodontid salamanders help in managing these important ecosystem roles.

Being lungless, plethodontid respiration is strongly affected by body moisture and the contact between their skin and contaminants (Droege et al. 1997). This sensitivity makes woodland plethodontids useful indicators of ecological stresses, as they are influenced by their micro-climate and water and air quality. Potential stresses include both human activities (development, pollution, etc.) and natural disturbances (storms, fires, etc.) or any event that may alter soil moisture, quality, or sun exposure (Zorn et al. 2004).

Finally, monitoring and identifying plethodontid salamanders can be done with relative ease. With a limited number of salamander species inhabiting the area, accurate identification can occur with minimal training, and reliable data can be collected from year to year with varying observers and/or volunteers. Additionally, since woodland plethodontids are attracted to artificial cover boards (ACOs) they can be easily sampled, avoiding destruction of habitat and unnecessary stress or harm to individuals. Since populations remain relatively stable, population trends can still be detected with small sample sizes (Zorn et al. 2004).

*3.1.4 EMAN Plethodontid Salamander Monitoring at **rare***

In 2004, the Ecological Monitoring and Assessment Network (EMAN) and Parks Canada published a joint National Monitoring Protocol for plethodontid salamanders. The goals of this protocol were to work alongside a suite of other standardized protocols to act as an early detection of ecological change and to environmental issues. First and foremost, this protocol aims to provide a standardized methodology for plethodontid monitoring across Canada (Zorn et al. 2004). The protocol involves the establishment of permanent forest monitoring plots which contain a series of wooden ACOs (artificial cover objects) spaced evenly across the forest floor. Zorn et al. (2004) suggest that monitoring should ideally occur in both spring and fall of each year to achieve the best results relating to salamander abundance and community structure as an indicator of ecosystem health.

The salamander monitoring program at ***rare*** is conducted exclusively in the fall due to monetary and time constraints. The program was established in 2006 with the installation of 29 ACOs in Indian Woods. Following a pause in 2007, the monitoring resumed in 2008 and was expanded to include a second monitoring plot in the Hogsback consisting of twenty ACOs, running for only five weeks. In 2009, the program was once again expanded with the addition of three ACOs to the already established monitoring plot in Indian Woods, bringing the total number of ACOs in that plot to 32 and increasing the length of monitoring in the Hogsback to the full nine weeks. Monitoring has been ongoing with consistent a nine-week sampling effort each fall since 2009 at both sites.

Salamanders successfully began using the ACOs within weeks of establishment and continue to use them despite resultant disturbances from the monitoring process. The initial years of this monitoring have resulted in the collection of valuable baseline data regarding salamander populations at ***rare*** with which data from future years can be compared in order to determine how ***rare*'s** salamander populations are changing over time. Additionally, McCarter (2009) identified specific research questions regarding the goals and mandates of this monitoring initiative at ***rare***:

1. What is the current state (species diversity, abundance, age structure) of the salamander populations in ***rare*** forests, and how do they compare to one another?
2. What are the long-term trends in Eastern Red-backed salamander abundance and population structure taking place within Indian Woods and the Hogsback?
3. Is the ecosystem integrity of Indian Woods and the Hogsback being maintained or improved under ***rare*** management?
    - Ecosystem integrity is defined as an ecosystem that has its native abiotic and biotic components intact and likely to persist (Parks Canada 2009)
4. Is either the ecological health or integrity of Indian Woods and the Hogsback being affected by on-site and nearby changes in land use (i.e. restoration, agriculture, residential development and aggregate extraction)?
    - Ecosystem health is defined as an ecosystem that has the capacity to resist and recover from a range of disturbances, while maintaining its functions and processes (Styers et al. 2010; Twery and Gottschalk 1996)

3.2 Methods

3.2.1 Monitoring Locations

Indian Woods (IW) is an old-growth Sugar Maple-American Beech (*Acer saccharum-Fagus grandifolia*) dominated forest located on the western side of the ***rare*** property, south of Blair Road and north of Whistle Bare Road. The forest covers approximately 20 acres and contains trees as old as 240 years. The Indian Woods salamander monitoring plot is located on the east side of an ephemeral pond near the south edge of the forest (Appendix A, Figure A.3).

The plot is accessed by parking at the South Gate on Whistle Bare Road, and walking north along the Grand Allée trail until a second path merges from the west (left) side. This second trail is marked by a blue square sign with a white arrow. From the point of the trail junction, walk east (right) into the forest towards a large ephemeral pond (approximately 100m). The 32 ACOs are distributed in a large square made up of four lines of eight ACOs each (Appendix A). Boards five, six, and seven were missing prior to 2009.

The Hogsback (HB) is a 57-acre forest located approximately 700m southeast of Indian Woods, south of Blair Road, and just west of the Newman Drive subdivision. It is comprised of mixed swamp interspersed with ridges of upland forest characterized by Red Maple (*Acer rubra*) and White Pine (*Pinus stroba*). The Hogsback salamander plot can be accessed from the Springbank Community Gardens by travelling across the farm field adjacent to the gardens to the edge of the forest. At the forest's edge, on foot, keep left and walk north and then east along the edge of the forest, finally heading south into the stand at an area of downed fence marked by pink flagging tape on a fallen log. Continue south into the stand for approximately 50m to the monitoring plots. Twenty ACOs are distributed in a large rectangle with eight ACOs on the north and south sides and two ACOs on the east and west sides (Appendix A). Each board is identified with a writeable aluminum tag marked as follows: SITE-YEAR -NUMBER (ex.HB-08-01) and is flagged with pink or orange flagging tape on an adjacent shrub or tree.

3.2.2 Monitoring Protocol

One month prior to the start of monitoring, all ACOs in both Indian Woods and the Hogsback were visited to ensure proper positioning and clear labelling. If necessary, boards were repositioned so that they were flush against the soil and reoriented into their original location. As the boards have been in place for multiple years, the proper positioning is generally noticeable as an area of bare soil. Labels and flagging tape were replaced as needed, and any holes in the boards were packed with soil to prevent salamanders from hiding during monitoring. Boards that were missing or too damaged or decomposed to be viable were replaced by newly cut boards, and relabelled with the current year.

Each plot was monitored once a week for nine successive weeks from the end of August to the end of October. Indian Woods and the Hogsback were monitored for only five weeks in their pilot years, 2006 and 2008, respectively.

At the beginning of each monitoring session, water was collected into a squeeze bottle from the education pond behind Lamb's Inn. This water was used to calibrate the soil moisture meter (Lincoln Irrigation Corporation, Lincoln, Nebraska, USA) by adjusting the meter with a screw driver so that it read a moisture rating of "10: saturated" when the probe was completely immersed in the water. The start time for the entire monitoring plot and Beaufort's wind and sky codes were recorded on the data sheet at the start of monitoring (see Appendix C). Additionally, the precipitation from the 24hrs prior to monitoring was recorded using the data collected by the Environment Canada Weather Office. In Indian Woods, the depth of the ephemeral pond was recorded using the measuring stick permanently in place.

Boards were always visited in sequential order starting with one. Soil temperature (°C) and moisture measurements were collected at each ACO by inserting the probes of the soil thermometer (Ashcroft® Thermometers, USA) and soil moisture meter to a depth of 10cm, as marked with tape on the probes, in the soil beside the board. The ACO was then gently turned over and any salamanders underneath were collected by the observers wearing nitrile gloves and placed into a plastic container with a sponge dampened with pond water previously collected in squeeze bottle. Each salamander was identified to species (colour phase was indicated for Eastern Red-backed salamanders) and any noticeable physical defects were recorded. A list of common and scientific names for all salamanders observed at ***rare*** and their abbreviated codes is available in Appendix D, Table D.2. Salamanders were weighed on a digital scale (Equal Digital Scale, model #23-D-50, capacity 50g) in grams to two decimal places. Snout-vent length (SVL) and vent-tail length (VTL) were recorded for each individual using a set of digital calipers (TuffGrade IDI, Commercial Solutions, Alberta, Canada). To ensure measurements were recorded accurately from the vent, individuals were measured through a clear lid while pressed up against moist sponges in the base of the container to secure the salamander and view the ventral side. Following measurements, salamanders were released next to the board. Disturbances under or near the ACOs (e.g. snakes, ant nests, turkey scratches, fungus/mold, ACO movement) were also recorded. Data sheets can be found in Appendix C and on the ***rare*** server.

In each monitoring plot, specific ACOs were assigned the status of weather station and each weather station represents a specific subset of ACOs. Table 3.1 and 3.2 show which ACOs are associated with each weather station in Indian Woods and the Hogsback respectively. When each weather station is reached during the monitoring of boards in sequential order, weather variables including average wind speed (taken as the average after ten seconds), air temperature (°C) and percent relative humidity were collected using the Kestrel 3000 (Nielson-Kellerman, Boothwyn, PA, USA). Additionally, soil samples for pH testing were collected from both Indian Woods and the Hogsback at each weather station on the last day of monitoring. A complete list of required equipment is available in Appendix B, List B.3.

Table 3.1: Weather stations and the artificial cover objects (ACOs) associated with them in Indian Woods salamander monitoring plot.

Weather Station ACO Number	Associated ACOs
3	1,2,3,4
7	5,6,7,8
11	9,10,11,12
15	13,14,15,16
18	17,18,19,20
23	21,22,23,24
27	25,26,27,28
31	29,30,31,32

Table 3.2: Weather stations and the artificial cover objects (ACOs) associated with them in the Hogsback salamander monitoring plot.

Weather Station ACO Number	Associated ACOs
2	1,2,3,4,5
7	6,7,8,9,10
12	11,12,13,14,15
17	16,17,18,19,20

3.2.3 Data Analysis

Data were analysed using Microsoft Excel 14.0.6 (Microsoft 2010) and SPSS Statistics Version 20. Prior to analysis, assumptions of parametric testing were examined and appropriate transformations were used when assumptions were not met. If assumptions could not be met through transformation, the most robust tests were used, followed by cautious interpretation of results. Each salamander monitoring plot (Indian Woods and the Hogsback) was interpreted as representing a unique population, and each ACO within that plot was interpreted as representing a sample of that population.

Since each monitoring plot had a differing number of ACOs and since in 2006 and 2008 the Indian Woods monitoring plot had three less ACOs than in later years, data had to be standardized to allow for comparisons. Abundance was therefore transformed into catch per unit effort (CPUE) for each monitoring session, as is commonly used in fisheries science (Krebs 2001). To calculate CPUE, the total salamander count for each monitoring session was divided by the number of ACOs in that plot to get the mean weekly catch per ACO. The CPUE calculation included only Eastern Red-backed Salamanders.

A univariate analysis of variance (ANOVA) with one recoded combination fixed factor (plot and year) was used to look for differences in salamander abundance represented by CPUE. A two-way ANOVA split by plot was used to investigate weekly differences in salamander abundance, with week and year as independent variables. A two-way ANOVA split by plot was used to examine differences in species composition across all years. When interactions occurred data were either split (Zar 1999) or variables were combined and recoded into plot/year combination variables (Leech et al. 2008) depending on the question of interest. This was followed by Bonferroni post hoc testing to determine where the differences between the levels occurred.

Only Eastern Red-backed Salamanders (both colour phases) were considered in a size class comparison. Individuals were classified as either an adult, intermediate, or juvenile based on their snout-vent length as outlined in Zorn et al. (2004). Age classes were defined as follows: juveniles <25mm; intermediates 25mm-35mm; adults >35mm. Eastern Red-backed Salamanders are capable of dropping their tail as a defense mechanism (Wise and Jaeger 1998), and while vent-tail length was also measured, it is not a reliable indicator of size class. An ANOVA with three fixed factors (plot, year, and size class) was used to look for differences in salamander size class. Interactions between factors would signify that a size class varies among plots or years. Bonferroni post hoc testing followed to determine where differences occurred.

Each plot was analysed separately for their relationship with environmental parameters, as sampling effort varied with plot. 2006 and 2008 data for both plots were eliminated from this analysis since sampling effort varied from other years. To determine which environmental factors (year, soil temperature, soil moisture, soil pH, pond depth, precipitation, sky and wind codes, wind speed, relative humidity, and air temperature) affected total salamander abundance, multiple linear regressions were used. Multicollinearity of independent environmental variables was assessed with Pearson's r. Preliminary assessments indicated that soil moisture, Beaufort sky and wind codes, and precipitation had a significant impact in the Hogsback, and in Indian Woods soil temperature and Beaufort sky codes significantly impacted abundance. Hierarchal multiple regressions followed with total abundance as the dependent variable and related parameters as the independent variables. Variables were entered into models based on their inherent relationship with salamanders (i.e. since salamanders live in the soil, soil factors were likely important). How well each model predicted the dependent variable- the goodness of fit of each model- was determined using the Akaike's Information Criterion (AIC) model selection technique.

3.3 Results

3.3.1 Total Observations

A total of 224 salamanders were observed between September 1^{st} and October 28^{th} at the ***rare* Charitable Research Reserve** in 2015. In Indian Woods, 83 salamanders were observed and in the Hogsback 141 salamanders were observed.

Eastern Red-backed Salamanders represented 96.4% of detections; 84.3% were the red-backed form, 16.7% were the lead-backed form of the same species. The remaining 3.6% of salamanders found under ACOs were comprised of three Blue-spotted Salamander observations, one Yellow-spotted Salamander and four observations of Four-toed Salamanders. Using age classes outlined in Zorn et al. (2004), 60.1% of the total detections of the Red-backed Salamanders were adults. There were 27 instances with two salamanders under one board, and 12 instances of three or more.

3.3.2 Salamander Abundance

Plot differences varied with years (interaction $F_{8,146}$ =3.925, p<0.001), so both factors were considered simultaneously in an sixteen-level combination variable of plots and years (Leech *et al.* 2008). Some significant differences were found between these levels ($F_{16,128}$=9.437, p<0.001). Within years, CPUE at each plot generally did not significantly differ, although largely more observations were documented in the Hogsback (Figure 3.1). Three exceptions exist.

In the Hogsback, CPUE in 2013 was significantly greater than what was observed between 2008 and 2012 (p<0.05), excluding 2009 (p>0.05). 2013 was also the highest ever recorded CPUE at this site. After 2013, subsequent years have seen a drop in CPUE. CPUE in 2014 in the Hogsback was only significantly greater than CPUE from 2008 (p<0.05), the lowest year on record, and CPUE for 2015 was not significantly different from any other year (p>0.05). CPUE in 2006 and 2008 for Indian Woods was significantly greater than all proceeding years

(p<0.05). CPUE in 2015 at Indian Woods was low and only slightly greater than the lowest year on record, 2011, significantly differing from the first two years of Indian Woods sampling (p<0.001)

Differences in salamander abundance were examined across weeks (Figure 3.2). This analysis used total weekly salamander abundance as the dependent variable as opposed to CPUE, and excluded years 2006 and 2008 as sampling efforts differed. Since number of ACOs in each plot differed, Indian Woods and Hogsback were examined independent of one another. No significant differences occurred between weeks at either plot (IW: $F_{8,146}$=0.968, p=0.463; HB: $F_{8,146}$=1.311, p=0.242). In Indian Woods in 2015, number of salamanders seen was the lowest on record in four separate weeks (weeks two, five, six, and eight) and was tied for the highest number of salamanders seen in week nine. For the Hogsback in 2015, week one tied for highest number of salamanders seen and all subsequent weeks had middling abundances.

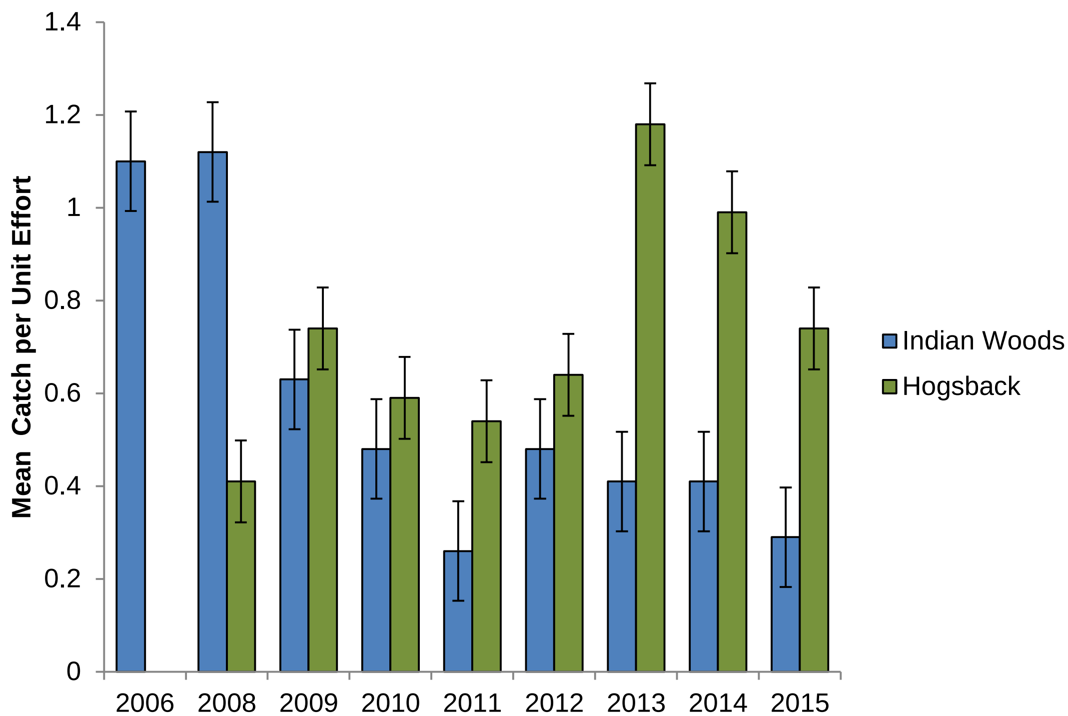


Figure 3.1: Meanweekly salamander observation per artificial cover object (ACO) (Catch per Unit Effort) for both Indian Woods and Hogsback for all monitoring years. Error bars represent +/- one standard error.

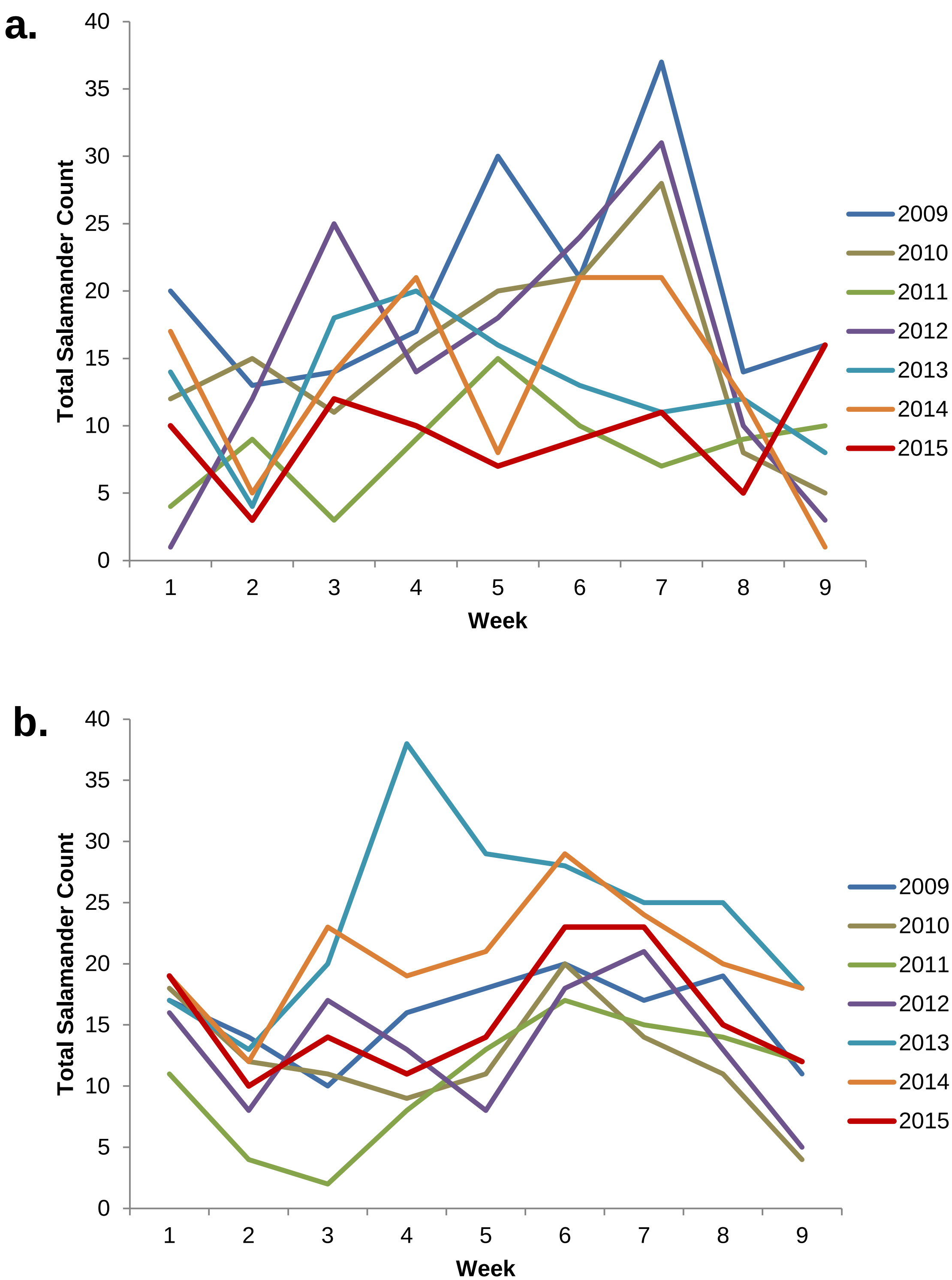


Figure 3.2: Total weekly salamander counts in Indian Woods (**a.**) and the Hogsback (**b.**) from 2009-2015. Data from 2006 and 2008 is excluded due to unequal sampling effort.

3.3.3 Salamander Species Composition

Plot differences varied with year ($F_{7,253}$=7.700, p<0.001), so data were split by plot (Zar 1999) and Indian Woods and Hogsback were each considered independently of one another. In both plots, significant differences occurred between species (p<0.001), with significantly more Eastern Red-backed Salamanders occurring than any other species, regardless of year (p<0.001). Red-backed and Lead-backed salamanders are two colourmorphs of the same species, the Eastern Red-backed salamander. There were significantly more Red-backed morphs than Lead-backed morphs in both plots (p<*0*.001). Four species have been observed in the Hogsback since 2008, and only two species have been observed in Indian Woods since 2006 (see Figure 3.3).

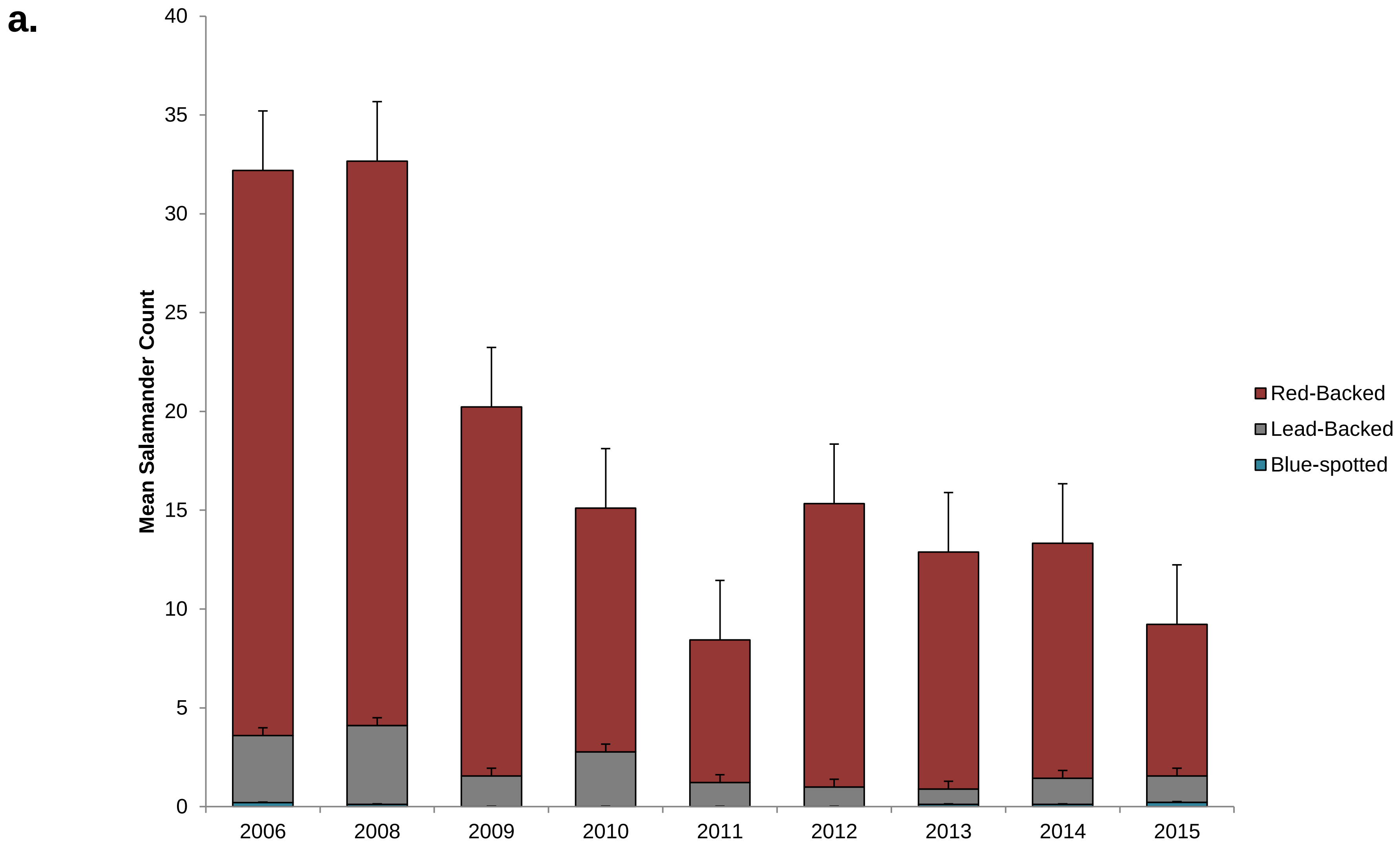


Figure 3.3a: Mean salamander abundance by species for each monitoring year in Indian Woods. Red-backed and Lead-backed are two colour morphs of the same species, the Eastern Red-backed Salamander.

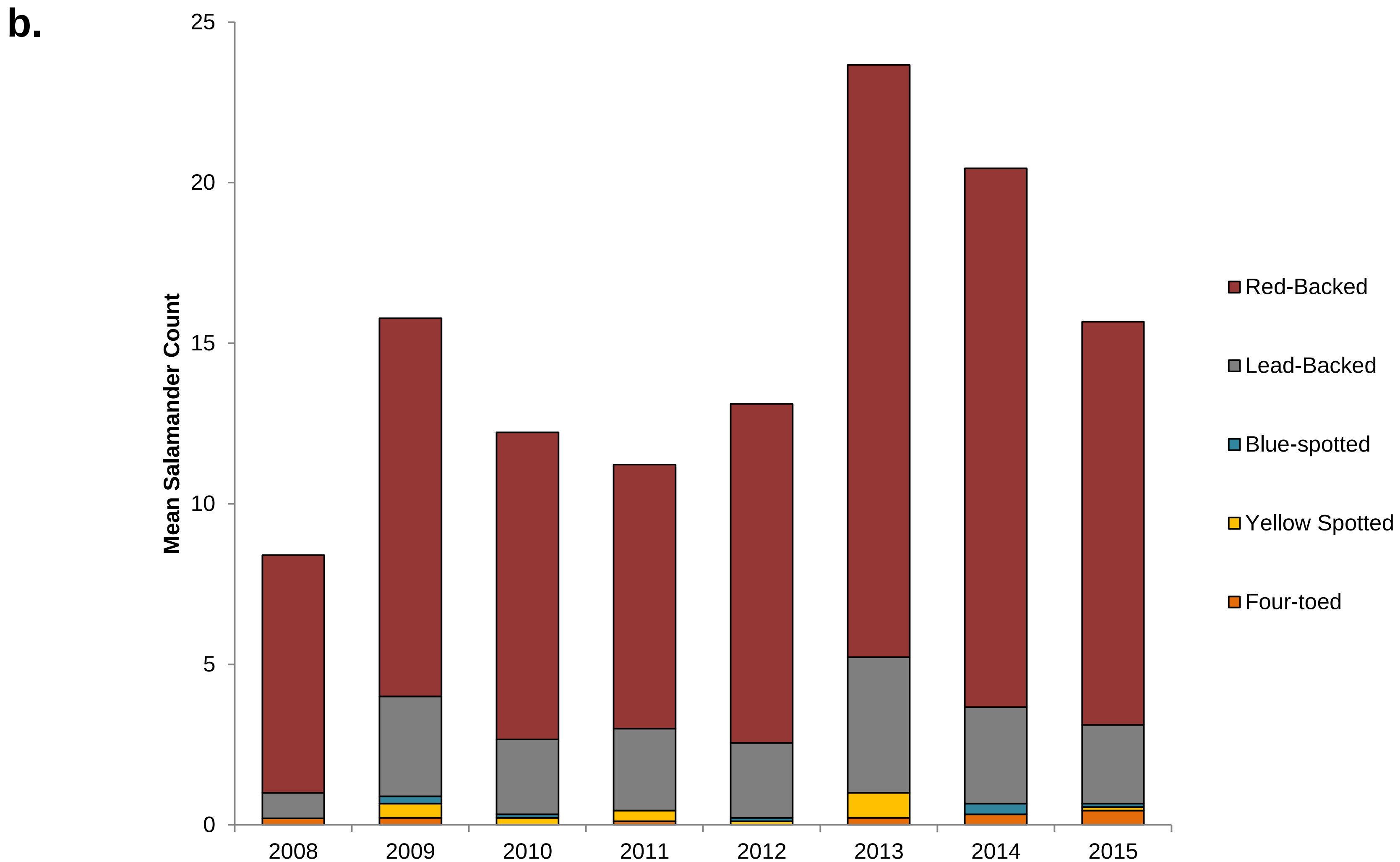


Figure 3.3b: Mean salamander abundance by species for each monitoring year in Indian Woods (**a.**) and the Hogsback (**b**.). Red-backed and Lead-backed are two colour morphs of the same species, the Eastern Red-backed Salamander. A table of abundance means and their corresponding standard errors can be found in Appendix G.

3.3.4 Eastern Red-Backed Salamander Size Class Distribution

Plot interacted with size class ($F_{7,284}$=3.335, p=<0.01) so data were split by plot and Indian Woods and the Hogsback were investigated separately. In both plots total weekly salamander observations significantly differed by size class with significantly more adult salamanders than intermediate and significantly more intermediate salamanders than juveniles (IW: $F_{2,153}$=83.159, p<0.001; HB: $F_{2,131}$=111.199, p<0.001). Size classes did not differ significantly across years for both plots (p>0.05). While not statistically significant, size classes in 2015 were more evenly distributed among adults and intermediates than in previous years in Indian Woods and there is a noticeable decrease in the number intermediate salamanders in the Hogsback (see Figure 3.4)

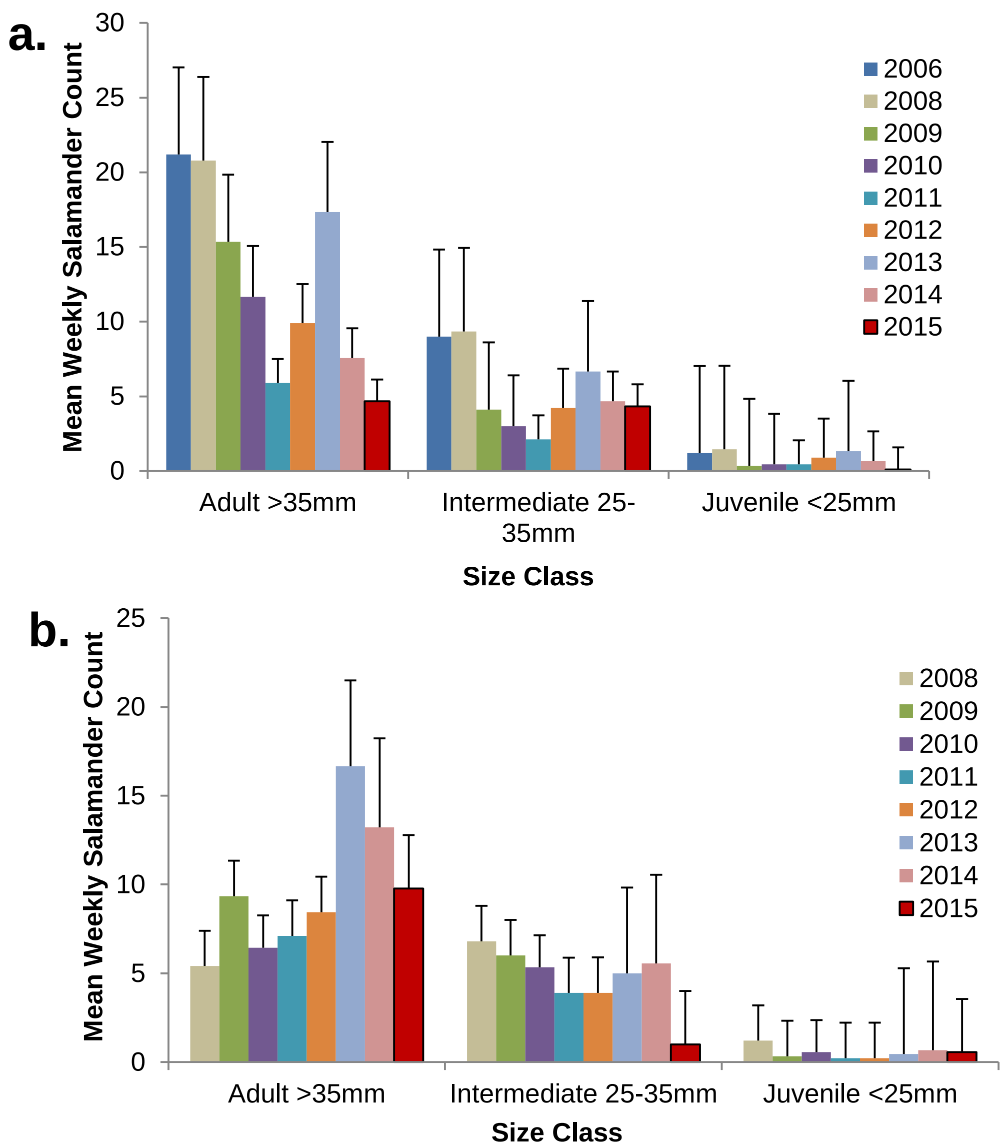


Figure 3.4: Mean size distribution of salamanders observed weekly during monitoring in Indian Woods (**a.**) and the Hogsback (**b.**) from 2006-2015. Error Bars represent + one standard error.

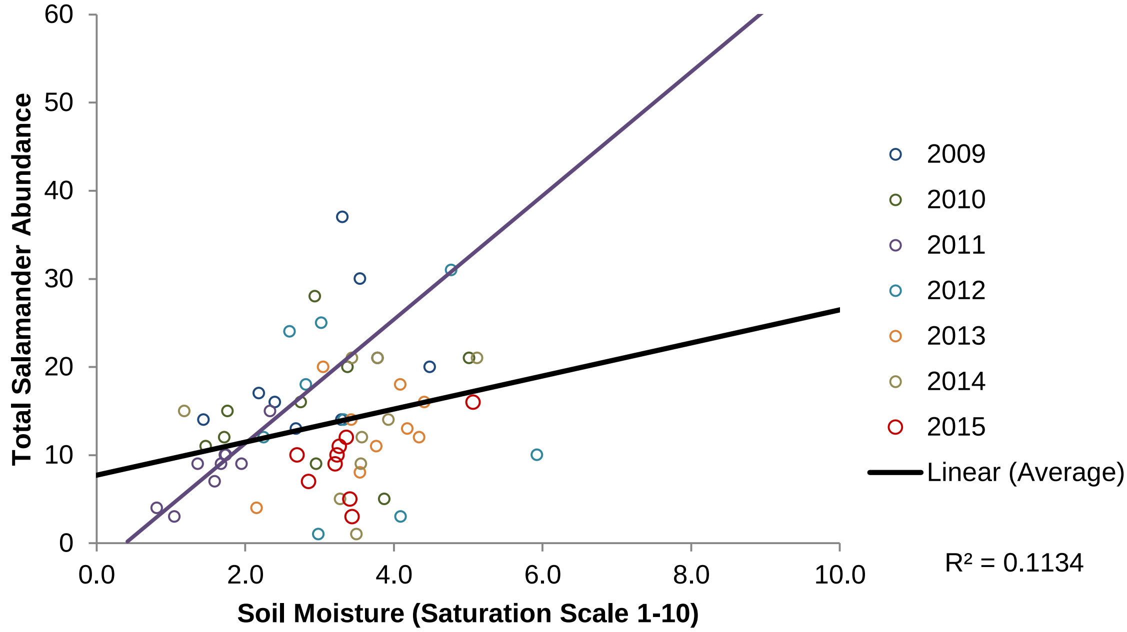
3.3.5 Environmental Parameters

Since air temperature and soil temperature were found to be highly correlated ($r=0.864$; $p<0.001$), only one parameter could be used in analysis, so air temperature was not included. It should be noted that week was not a significant predictor of salamander abundance in preliminary analysis, but week is also highly correlated with air and soil temperatures. This is not surprising as we would expect soil and air temperature to decrease with time as we move from September to October. For Indian Woods, Beaufort sky codes and soil temperature were found to have a significant impact on and a negative relationship with abundance ($F_{2,60}=6.337$, $p<0.01$ $r^2=0.174$).

For the Hogsback, soil moisture, Beaufort sky and wind codes and precipitation were all found to have a significant impact on the abundance of salamanders ($F_{3,53}=5.948$, $p<0.001$, $r^2=0.291$) with soil moisture being the most influential factor on abundance ($r^2=0.112$). Soil moisture was the only factor shown to have an overall positive relationship with abundance, with the exception of 2015 where soil moisture had a negative relationship with abundance. Soil moisture has been found to have the strongest relationship with salamander abundance in past years as well (See Figure 3.5).

Weather during the 2015 monitoring months was roughly average compared to other years for October, but far different than other years in September (See Figure 3.6 and Figure 3.7). September 2015 was the warmest on record by several degrees and rainfall was the second lowest since the start of monitoring.

a.



b.

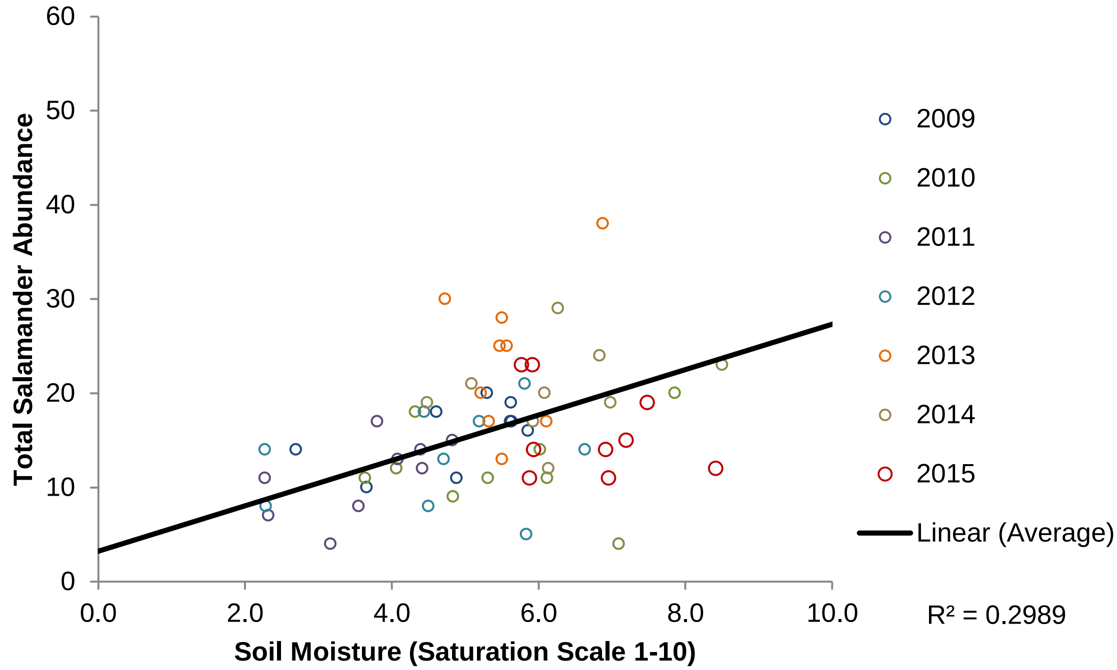


Figure 3.5: Relationship between total salamander abundance in Indian Woods (a.) and the Hogsback (b.) and measured soil moisture for 2009-2015. Trendlines are only displayed for significant relationships (p<0.05). Average abundance-moisture relationship for all years and corresponding r^2 of the relationship is displayed as well.

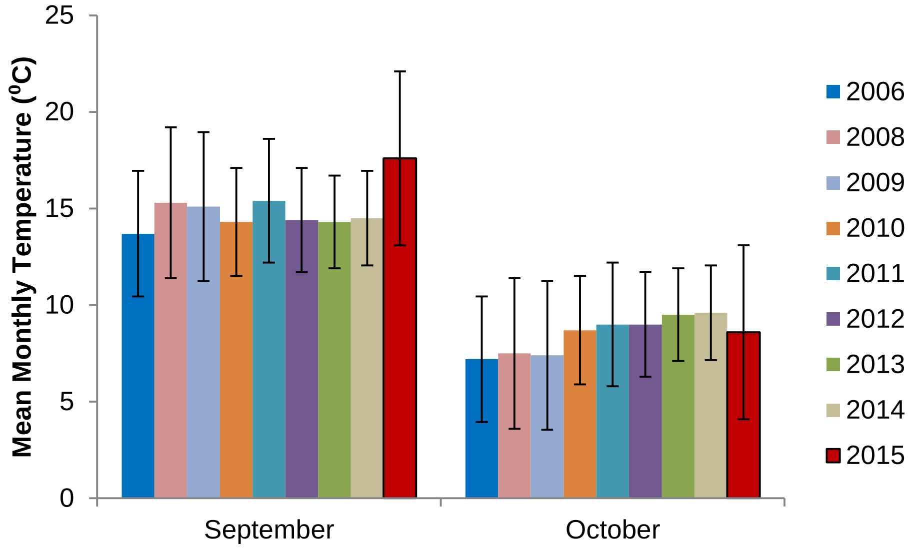


Figure 3.6: Mean monthly temperatures for Waterloo Region during the salamander monitoring season in 2006, 2008-2015 (Environment Canada- 2006, 2008-2009 data from Waterloo International Airport Weather Station, and 2010-2015 data from Kitchener-Waterloo Weather Station). Error bars represent +/- one standard error.

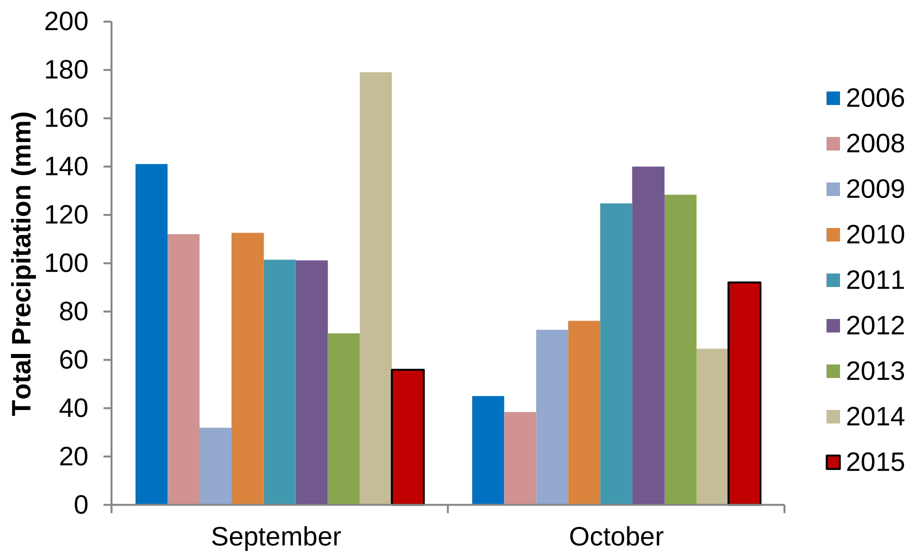


Figure 3.7: Total monthly rainfall for Waterloo Region during the salamander monitoring season in 2006, 2008-2015 (Environment Canada- 2006/2008-2009 data from Waterloo International Airport Weather Station, and 2010-2015 data from Kitchener-Waterloo Weather Station).

3.4.0 Discussion

3.4.1 Eastern Red-Backed Salamander Abundance

Given their importance in food web dynamics and their sensitivity to changes in forest floor conditions, significant changes in plethodontid salamander populations over time may be an early warning of ecosystem stress. Recognizing a population change that may be acting as an early warning sign as opposed to natural population fluctuations requires a monitoring target or threshold to be set (Zorn et al. 2004). Zorn et al. (2004) recommends a monitoring threshold set at "a statistically significant change in plethodontid counts at a plot level over 5 or more years". With variable sampling effort in the first years of data collection, five consecutive and consistent years of data collection were achieved in 2013. Information gathered on salamander populations in the inaugural years does not contribute to the EMAN protocol for testing monitoring thresholds. It is suggested that the ACOs weather in situ for a winter prior to monitoring to avoid skewing abundance estimates due to the disturbance of plot establishment. Therefore thresholds for the first five consistent and consecutive years of salamander monitoring (2009-2013) are; **Indian Woods: 130 +/- 31** and for the **Hogsback: 136 +/- 38**.

In Indian Woods, the first two years of monitoring had the highest abundances on record, followed by a steady decline, culminating in the lowest abundance recorded in 2011 (Figure 3.1). Between 2012 and 2014 abundances rebounded to levels similar to 2009 and 2010, but 2015 registered numbers nearly as low 2011 and had values below calculated five-year thresholds (N=83). Numbers seen in 2006 and 2008 may have been exceedingly high as these years are when initial establishment of ACOs in Indian Woods occurred. This may have impacted the observed abundances by providing additional cover, acting as an artefact in attracting salamanders in early years and levelling out as ACOs became weathered and established over time (Van Wieren 2003). Studies on this topic are varied, with some reporting salamanders almost immediately making use of cover boards (Ballantyne 2004; Bennett et al. 2003; Monti et al. 2000) and others suggesting boards must be left for a year to weather before data collected is valid (Zorn et al. 2004; Droege et al. 1997). It may also be dependent on other factors, as suggested by Ballantyne (2004), where excess precipitation just prior to and at the start of monitoring may have sped the weathering process, making the boards more appealing to salamanders. Given this, the low abundance observed in 2011 may be attributed to the high precipitation levels. Jaeger (1972, 1980) reports that cover objects become more important during dry periods, acting as a moisture refuge for salamanders. Therefore, salamanders may be less dependent on cover boards in wetter years, having more moist spaces to use for foraging, and thus lower abundances may be observed under ACOs (Van Wieren 2003). However, fall precipitation in 2012, 2013, and 2014 was higher than in 2011, yet these years recorded higher salamander abundance.

A similarly counterintuitive trend is seen in 2015, as September was particularly dry and hot, high numbers of salamanders taking refuge under ACOs would be expected, however low abundances were recorded. A possible explanation for these low abundances may be that salamanders in Indian Woods retreated underground to reach the moisture they need to survive. Terrestrial salamanders will only spend time on the surface if moisture conditions are adequately high; if it is too dry salamanders will retreat underground to stay moist (O'Donnell and Semlitsch, 2015). Given the poor weather conditions, it is understandable that numbers of salamanders would be less than average. While weather conditions returned to about average in October, it is possible that salamanders retreated underground in September and never resurfaced during October.

Another possible explanation for these years of low abundance in Indian Woods is temperature outside of the September and October monitoring months. Winter in 2015 was the coldest on record, and winter in 2011 was colder than most years, perhaps causing low survivorship and thus low abundance during salamander monitoring in the fall. However, winter 2014 was the second coldest since 2009 and a similar drop in 2014 monitoring abundances was not observed. The major winter strategy of Eastern Red-Backed Salamanders is avoidance of sub-zero temperatures by retreating into the soil column (Storey and Storey 1986), and these salamanders have been observed as deep as one meter in the soil (Grizzell 1949; Hoff 1977). Harsh winters may have been particularly difficult to survive in if there was little or no snow covering the soil during these years. Snow pack acts as an insulator against ambient air temperatures protecting animals beneath the snow (Aitchinson 2001). Although it does not appear to be an area of active research for salamanders, given the inability of Red-backed salamanders to survive sub-zero temperatures, snow cover should benefit their winter survival rates. While snow cover has not been measured for salamander monitoring, both 2011 and 2015 had the lowest average precipitation during winter months. Therefore it is possible harsh winter temperatures and a lack of snow cover created a lethal environment for some salamanders. In the future it may be pertinent to take measures of snow depth and/or soil temperatures from November to March to discover if this is indeed the case.

In terms of abundances observed per week, most years show a peak number of salamanders at the sixth or seventh week and a rapid decline in the weeks following (Figure 3.2). Week nine in 2015 does not follow this trend as the highest abundances in Indian Woods were seen during this week. The reason for high numbers the final week may be from soil moisture. Soil moisture levels were highest in 2015 in week nine at 5.05 compared to the yearly average of 3.4. This may be why the last week of monitoring in 2015 was highest on record (tied with 2009).

As Indian Woods fell below the recommended five-year population thresholds in 2015, it is possible some external stressor is impacting populations beyond measured environmental factors. Notably, anthropogenic stresses from nearby aggregate mining and agriculture could be having an impact. In 2015 the field adjacent to Indian woods has been converted into a grazing pasture for cattle. The effects of cattle on terrestrial salamanders are poorly understood. Riedel et al. (2008) report fewer Red-backed salamanders are found in fields with cattle, but interestingly these salamanders can still be found in these fields, despite grazing activity. This indicates that Red-backed salamanders may be resilient to the impacts of cattle on an ecosystem. While Red-backed salamanders may not be impacted by cattle, there is evidence that indicates livestock have a negative impact on aquatic juvenile salamanders in streams and ponds (Knutson et al. 2004). This may mean our other three species of salamander at ***rare*** are in peril and because fall salamander monitoring poorly captures other species, any impact cattle are causing is unlikely to be captured through fall monitoring alone. While this may be of great concern for salamander populations, given the distance from the cattle to the ACOs in Indian Woods and the lack of water features along the margin of the forest in this area, the impact from cattle may be negligible, at least on monitored populations. Still it may be prudent to include additional years of spring monitoring to assess impact from increased agricultural activity on other species of salamander.

The other major anthropogenic concern is from nearby aggregate mining operations. Aggregate mining is known to lower the water table in surrounding areas (Environment Canada 2004). In past monitoring years, the pond near the ACOs in Indian Woods has been filled with water in one or more monitoring weeks. In 2015 there was no water present in this pond throughout monitoring, indicating the water table may have fallen in this area. If the pond fails to fill with water in the future, this area may cease to be a productive breeding site for salamanders with aquatic juvenile phases. Because Red-backed salamanders live a completely terrestrial life they do not require this pond for breeding, still a reduced water table may mean they must burrow deeper into the soil to find moisture in periods of drought, potentially reducing their visibility during salamander monitoring. It must be noted that 2015 also had less total annual rainfall than all other monitoring years, which may have caused the pond to stay dry. Also, aggregate mining operations have been ongoing for years in the area and any actual reduction in the water table within the past year is only speculative at this point. However, the issue of the potential of a reduced water table should not be overlooked in future years and should be investigated as it has the potential to seriously impact salamanders in this area.

Canopy cover may also be impacting the abundance in Indian Woods. In winter 2014, a storm knocked down several large trees in Indian Woods opening canopy gaps over several ACOs. Canopy cover has been directly linked to salamander abundance in past studies (Dupuis et al. 1995, Riedel et al. 2008). Therefore, the reduced canopy cover caused by the storm would be expected to lower salamander abundances, at least locally, under affected ACOs. Since 2013, canopy cover was monitored by assigning each board into one of three categories: complete, incomplete or no canopy cover. Thus far there has been no significant relationship found between salamander abundance and canopy cover. As there are relatively few ACOs where major canopy gaps have opened, it is possible this storm event has had minimal impact on abundance. Still, in terms of abundance for ACOs directly under where canopy gaps have opened, there has been a noticeable decline in the number of salamanders (2013 n=31, 2014 n=25, 2015 n=14). This indicates that we may be seeing localized loss of abundance in these ACOs, which may be one of the main reasons 2015 had so few salamanders in Indian Woods. If this is the case, salamander abundance would be expected to stay low until canopy gaps fill from trees in the understory.

In the Hogsback, 2013 abundances were significantly higher than all previous years, with the exception of 2009 (Figure 3.1). However, 2014 and 2015 were not significantly less than 2013 and comparably high abundances were observed. Contrary to the establishment years in Indian Woods, the original monitoring session in the Hogsback (2008) is the lowest of all years, suggesting perhaps acclimatization of the newly placed ACOs was taking place (Zorn et al. 2004; Droege et al. 1997). The relatively consistent observed abundances following the first year of monitoring in the Hogsback are an encouraging trend, reflecting a likely stable population. In 2013 and 2014, a large population spike occurred, with a greater number of salamander observations than previous years in nearly every week. In particular, week four of 2013 saw an extremely high abundance when salamanders were found under seventeen of the twenty boards in 2013. Given the unremarkable environmental data gathered in this week, it is unknown what caused this large increase in observations.

September 2014 saw the highest rainfall ever recorded during monitoring, and while precipitation was lower in September and October 2013 than in previous years, the annual rainfall was higher so conditions in these years may have been well suited for populations to thrive. This certainly appears true when considering the soil moisture levels measured throughout monitoring (Table 3.3). 2013 and 2014 in the Hogsback have higher moisture levels than most previous years indicating that the ACOs (as the reading was taken within the microclimate created by the ACO) were an ideal location for salamanders. The average monthly temperatures in October 2013 and 2014 were also higher than in previous years, which may have prevented the typical drop-off in observations that has been previously observed nearing the end of the monitoring season. This would then cause an inflation of observation numbers as salamanders typically retreat underground to avoid winter temperatures. As can be observed in Figure 3.3, the increase in salamanders observed was primarily observations of adult Eastern Red-Backed Salamanders.

Salamander abundances in 2015 for the Hogsback are roughly average and fall within five-year population thresholds. High September temperatures and low rainfall coupled with moderate temperatures and rainfall in October may have caused the middling abundances observed. Also of note is the highest average soil moisture of any year was in 2015, but as mentioned, a particularly cold winter may have decreased overwinter survivorship.

In all likelihood there is no one cause of the abundances seen in any year and factors including temperature, moisture, and available cover can be having an impact(Heatwole 1962; Spotila 1972; Feder and Pough 1975; Jaeger 1972, 1979, 1980; Feder 1983; Feder and Londos 1984; DeMaynadier and Hunter 1998; Herbeck and Larsen 1999). This view is also supported in the data at ***rare*** (see section 4.4 for an analysis of environmental parameters impacting salamander abundance). Overall, population size in the Hogsback appears to be stable and the population in Indian Woods may be suffering. Careful monitoring in subsequent years will determine if this is indeed the case.

Table 3.3: Average soil moisture levels during the salamander monitoring season in 2009-2015 at Indian Woods and the Hogsback.

Plot	Year	Mean Soil Moisture Level
Indian Woods	2009	3.02+/-0.93
	2010	2.87+/-1.14
	2011	1.58+/-0.460
	2012	3.53+/-1.18
	2013	3.68+/-1.91
	2014	4.65+/-2.55
	2015	3.4+/-2.12
Hogsback	2009	4.87+/11.07
	2010	5.47+/-1.42
	2011	3.65+/-0.912
	2012	4.63+/-1.37
	2013	5.63+/-2.34
	2014	4.94+/-2.45
	2015	6.7+/-2.77

3.4.2 Salamander Species Composition

While the monitoring program at ***rare*** is primarily designed for plethodontid salamanders (Zorn et al. 2004), other species have also been observed on the property. Between 2009 and 2012 in Indian Woods, only Red-backed Salamanders were observed with the red-backed colour phase being dominant (Figure 3.3a). For the past three monitoring seasons, one or two Blue-Spotted Salamanders were observed in Indian Woods, a species not recorded since 2008. This is possibly connected to the vernal pond adjacent to the plots which appears to be used by this species for breeding; more information on this species can be found in the report on the jeffersonanium-laterale complex investigation completed in spring 2015. During fall monitoring in 2015, no water was recorded in this pond, some water was found in 2014 (average 5cm) and the most water of any monitoring season was found in 2013 (average 29cm). Mole salamanders are more easily found in the spring during their breeding season (Whitford and Vinegar 1966) and therefore their presence during fall monitoring may be seen as an abnormality. Continued monitoring of this trend may inform us whether Blue-spotted salamanders may be seen more commonly in Indian Woods in the future (e.g. from increases in population or displaced salamanders) or whether these are incidental observations.

In the Hogsback, Red-backed Salamanders are again dominant with the red-back phase more abundant than the lead-back phase (Figure 3.3b). This is unsurprising, as the lead-backed phase salamanders are known to experience preferential predation pressures (Moreno 1989; Venesky and Anthony 2007) and the red-backed phase is known to be proportionately higher in more areas and at higher latitudes (Lamond 1994; Harding 1997). Studies of spatial variation indicate that the lead-backed phase is more closely associated with warmer, drier climates, experiencing higher mortality in colder sites, and retreating from the surface earlier than red-backed individuals in the fall (Lotter and Scott 1977; Moreno 1989). Since there is a temperature preference between colour morphs, Gibbs and Karraker (2006) suggest increasing global temperatures may be resulting in a shift from red-backed individuals to lead-backed ones in temperate areas. In both the Hogsback and Indian Woods, the proportion of lead-backed salamanders does not show an increasing trend; however continued monitoring of Eastern Red-Backed Salamanders and the ratio of the varying colour morphs should be analysed as it may be indicative of important global temperature changes affecting the entire forest ecosystem.

Species diversity is higher in the Hogsback than Indian Woods (Figure 3.3). Four-toed Salamanders, another member of the plethodontid family, have been observed in most monitoring years in the Hogsback, with a record number of four observations in 2015. These salamanders are typically found in sphagnum moss or boggy woodlands (Conant and Collins 1998), the latter of which is found in the Hogsback forest stand. Multiple mole salamanders have been observed; Blue-spotted Salamanders in 2009, 2010, and every year since 2012 including one observation in 2015. Observations of Yellow-spotted Salamanders occurred between 2009 and 2013 and one observation was seen in 2015. In 2013, several Yellow-spotted Salamanders of different sizes were observed under different ACOs and a single Yellow-spotted was seen under a different board in 2015. However, from 2009 to 2012, it has been suggested that the same individual Yellow-spotted salamander was repeatedly observed as it was roughly the same size and consistently observed under the same ACO near what appeared to be a burrow or underground tunnel. This suggests salamanders may exhibit fidelity to ACOs. High site fidelity for salamanders has also been seen in other studies (Marvin 2001, Peterson et al. 2000). Expanding monitoring efforts at ***rare*** to include individual identification and possibly gender identification may be of benefit. Methods such as toe clipping to identify already sampled salamanders have been used in other studies (Trenham et al. 2000). With these identification methods we would be able to eliminate oversampling of individual salamanders, especially for poorly represented species like the Yellow-spotted salamander. This type of sampling, however, is more invasive and would require additional permitting. Programs have been explored in other studies to identify individual yellow spotted salamanders based on the location of their spots (Grant and Nanjappa 2006). This is perhaps something less invasive that ***rare*** could apply, but would only work for species with easily identified unique markings like yellow spotted salamanders.

3.4.3 Eastern Red-backed Salamander Size Class Distribution

In both Indian Woods and the Hogsback, the greatest proportion of Eastern Red-backed Salamanders in 2015 fell within the snout-vent length range of 35mm-45mm. Salamanders measured in the Hogsback and Indian Woods were of similar length (HB: mean SVL: 37.26+/-6.29; IW: mean SVL: 35.06+/-6.17), although individuals recorded in Hogsback were slightly heavier (HB: mean weight: 0.98+/-0.86; IW: mean weight: 0.80+/-0.32). Based on size class categories outlined in Zorn et al. (2004), significantly more adults were found in both plots than intermediates and juveniles, and further there are significantly more intermediates observed than juveniles (Figure 3.4). A significant positive correlation between unsexed salamander size and age in their first four years has been documented (LeClair et al. 2006). Based on their results, the majority of salamanders found under ACOs at ***rare*** are between the approximate ages of two and six (Figure 3.8). If other size class distinctions had been used to categorize salamanders at ***rare***, such as those outlined in Sayler (1966) and subsequently used in additional studies (Brooks 1999; Ballantyne 2004), data would have shifted toward more intermediate sized salamanders.

Trends in size-class distribution for 2015 are markedly different than those seen in previous monitoring years. In the Hogsback, there was a substantial drop in the number of intermediate salamanders seen. Trends in previous years have seen less of a drastic difference between adult and intermediate salamanders, but it appears many intermediate salamanders are no longer within the sampling area for the Hogsback. A probable scenario is a die-off of intermediates, potentially from the particularly cold winter in 2015 as they would likely have a lower survivability than adults in the harsh weather. If there continues to be a noticeable age gap in proceeding monitoring years it is likely due to intermediate salamander death. Interestingly this trend is not seen in Indian Woods, where numbers of adult salamanders were nearly equivalent to intermediate salamanders. In all other years of monitoring there have been noticeably more adult salamanders than intermediate salamanders, but in 2015 the difference has nearly disappeared. Given that the lowest ever number of adult salamanders were seen this year, it seems as though adult populations have declined compared to previous years, perhaps not being replaced as quickly by the next generation which could have suffered slowed growth from cold winter temperatures or some other factor.

A loss of adults instead of intermediates would be puzzling as they would be expected to be hardier than younger salamanders. One possibility is the level of ACO disturbance recorded in 2015. Disturbances under ACOs such as mould and predators are recorded for each ACO in each week. Average levels of ACO disturbance were found to be highest in 2015 with a significant negative relationship between salamander abundance and ACO disturbances ($p<0.01$). Adult salamanders may be avoiding disturbances as they are more likely to come into contact with them under ACOs due to their large size. The majority of recorded disturbances are related to the presence of mould. Mould disturbances may be increasing as the age of ACOs increases, which could impact monitoring in future seasons. A search of the literature indicates that this is a topic that has not been previously explored or is not considered an issue for salamanders. This may be due to the known anti-fungal properties of the mucus salamanders, including Red-backed salamanders, excrete from their skin (Lauer et al. 2008). The lack of impact from disturbance is supported by the data in the Hogsback where number of disturbances in 2015 was the highest ever recorded for both plots and all years, yet no significant relationship was found between abundance and disturbance ($p>0.05$). Still, no other measured variable has substantially changed in 2015 for Indian Woods making it possible that disturbance is playing a role in causing the low abundances. As mentioned, it is likely a combination of factors, including yearly weather, soil moisture, and temperature are working in tandem with high-levels of disturbance to cause low abundances. Should abundances in Indian Woods continue to decrease, the possibility of disturbance playing a role should not be overlooked.

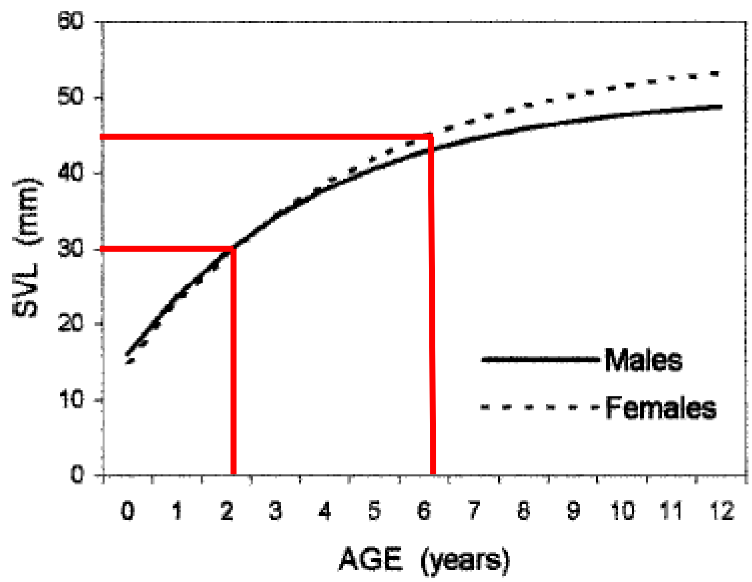


Figure 3.8: Growth in length (SVL) of Red-Backed Salamanders modified from LeClair et al. 2006. Red lines bound the dominant size range observed at ***rare*** plots.

Across all years, few juveniles (or first year young) have been found under the ACOs at ***rare*** in either forest stand for all monitoring years. Juvenile populations may be underrepresented by ACO sampling. Adults will exhibit territorial behaviours and outcompete juveniles for space (Marsh and Goicochea 2003), or, in the fall, this behaviour could be in connection to mating (Van Wieren 2003). Of 13 occasions in Indian Woods where multiple Red-backed Salamanders were found under the same ACO, only one occasion involved juveniles. Similarly in the Hogsback, of 27 occasions with multiple salamanders located under a single ACO, only one involved juveniles, and in this case both salamanders found under that ACO were juveniles. Red-backed Salamanders have been shown to exhibit kin selection, allowing related juveniles into their territories in stressful conditions (Horne and Jaeger 1988; Jaeger et al. 1995; Simons et al. 1997) however this seems to be occurring minimally, if at all, during the fall months at ***rare***. Territoriality of boards in connection to mating may be part of the cause for the underrepresentation of juveniles in this study.

Another likely hypothesis is that larger salamanders prefer the wider cover provided by ACOs. Mathis (1990) and Moore et al. (2001) found significant positive correlations between salamander size and cover object size. Therefore, ACOs used in this study may be more attractive to larger adults. Gabor (1995) found this relationship with cover object size and salamander size existed only where direct sunlight reached the board. In cases where direct sunlight does not heat the boards, cover objects were chosen in relation to food quality and quantity in surrounding areas. As mentioned, data so far have shown no significant relationship between salamander abundance and canopy cover.

3.4.4 Environmental Parameters

Many factors have been shown to impact plethodontid salamanders including temperature (Spotila 1972; Feder & Pough 1975), moisture (Grover 1998; Feder & Londos 1984), soil pH (Wyman and Hawksley-Lescault 1987; Sugalski and Claussen 1997; Moore and Wyman 2010) and other environmental conditions (Heatwole 1962; Feder 1983; DeMaynadier and Hunter 1998; Jaeger 1972, 1980). This study found that the location of the plot had a large effect on which environmental or temporal variables had an impact on abundance. In the Hogsback, a forest-wetland complex with a thick canopy, it was found that soil moisture, Beaufort sky and wind codes and precipitation were all significantly impacting salamander abundance. In Indian Woods, a remnant old-growth forest with a thin, sparse canopy in areas, total abundance was found to be significantly linked to Beaufort sky codes and soil temperature.

Soil temperature was found to significantly negatively impact abundance in Indian Woods. High temperatures cause salamander skin to dry out more quickly and, as a consequence, limit their surface activity (Spotila 1972, Feder and Pough 1975). Given this, a negative relationship between soil temperature and abundance is to be expected.

The negative relationship found between Beaufort sky codes and abundance in both forest areas is not surprising. As plethodontid salamanders must stay moist to survive, one would expect to see more salamanders hiding under boards when there are low sky codes (corresponding to sunny or partly cloudy) and less salamanders with high sky codes (corresponding to overcast or raining) as they will disperse from under the boards to more moist areas. The negative relationship between Beaufort wind codes and abundance in the Hogsback is, however, more surprising.

Higher wind codes correspond to higher wind speeds and higher wind speeds saw fewer salamanders. One would expect fewer salamanders to be found under boards when wind codes were low and more salamanders taking refuge under boards when wind speeds were high to avoid desiccation. This pattern was not seen, but the negative relationship between wind codes and abundance is likely from poorly distributed data. The vast majority of weeks of salamander monitoring had Beaufort wind codes of 0 or 1 and only three weeks (of a possible 63) have had wind codes higher than 4, causing these points to have a disproportionate impact on the data. Removing the data from these three weeks greatly weakens the linear relationship. This is coupled with the failure to find any significant relationship between ACO microclimate wind speed and salamander abundance, lending evidence that the relationship seen in the complete dataset over exaggerates the importance of wind codes.

Precipitation data in the Hogsback also appears to encounter the same problem as wind codes. The majority of precipitation data (recorded precipitation in the past 24hr) has 0mm values with only 12 of 63 weeks having greater than 10mm of rainfall. These comparatively few values inflate the relative importance of precipitation within this study. It must be noted that time since rainfall has been shown to have a substantial impact on salamander surface activity (O'Donnell and Semlitsch 2015), but as the monitoring program occurs at the same time every week it is difficult to quantify the real impact precipitation is having. While beyond the scope of this monitoring program, a targeted study measuring amount of and time since rainfall, compared to abundances seen would shed more light on this relationship.

The significant relationship between soil moisture and total abundance in Hogsback is overall a positive one, where more salamanders are observed when soil moisture is higher (Figure 3.5). The impact of soil moisture on salamander abundance is not surprising. Plethodontid salamanders require moist skin to facilitate gas exchange across their cutaneous membrane for respiration (Behler and King 1979; Welsh and Droege 2001). During cool, moist weather they can disperse across the forest floor, while in drier conditions they would be confined to moist microhabitats or spend very little time in dry exposed areas (Jaeger 1972, 1980; Feder 1983; Droege et al 1997). This relationship was particularly strong in 2013, a year with the second highest average soil moisture (Table 3.3). High soil moisture in this year potentially allowed many salamanders to stay on the surface for longer periods resulting in the high abundances seen. Interestingly the only year to have a negative relationship between soil moisture and salamander abundance was 2015. 2015 had the highest average soil moisture of any monitoring year. It is possible that soil moisture was so high fewer salamanders sought refuge under ACO as the cover they provided was unnecessary than compared to 2013 and 2014.

While soil pH does not appear to be playing a significant role in controlling the abundances seen at ***rare,*** other studies found pH to be the more influential factor (Sugalski and Claussen 1997). This is not the case in this monitoring program, likely because soil conditions in both forest stands fall within or close to their preferred pH range of 6.0 to 6.8 for plethodontid salamanders (Heatwole 1962), and it is suggested that these salamanders avoid soils with pH outside of these ranges (Wyman and Hawksley-Lescault 1987; Wyman 1988). In future monitoring years, if pH becomes a more accurate predictor of salamander abundance, it may be an early warning sign of soil acidification.

Overall, most of the environmental variables show intuitive relationships with salamander abundance. The relative importance of some variables, such as Beaufort sky and wind codes appears to be over-exaggerated, while some variables such as soil moisture in Indian Woods appear to be under-represented. Many of the variables (i.e. wind codes, precipitation, etc.) are poorly distributed and contain mainly zero values, making their inclusion into statistical tests (even when transformed) produce suspect results. Any interpretation in future years of these variables should keep these limitations of the data in mind.

3.5 Conclusions and recommendations

After seven years of consistent and consecutive monitoring, this program has established baseline data of expected salamander populations in both Indian Woods and Hogsback and will continue to compare future years to these baselines. Populations of salamanders in the Hogsback seem stable; however salamanders in Indian Woods have fallen outside of this baseline threshold. Age class distribution for both forest stands has also changed quite dramatically compared to previous years. These changes are concerning and the cause, whether temporary or permanent, is unknown. As this program acts as a warning sign for environmental change, falling numbers coupled with ongoing human pressures from agriculture, development projects and the potential for accumulative effects from aggregate extraction highlight the need for continued monitoring at ***rare***. Only by continuing long-term monitoring, can ***rare*** best assess the impact of land management decisions both on an adjacent to the property. Therefore, it is recommended that a full nine week monitoring program continue at both forest sites.

In addition, the inclusion of Mirrored Research salamander boards into the Cliffs and Alvars forest will continue to expand the program to include all three forest stands on ***rare*** property, and allow for a more complete analysis of ecological health. Salamander monitoring also took place in spring 2015. This one year data collection may also allow ***rare*** to better understand the true biodiversity of species on the property. In the future, a long term spring monitoring program may be explored.

3.6.0 Literature Cited

Aitchinson, C. W, 2001. The effect of snow cover on small animals. In: H.G. Jones, J.W. Pomeroy, D.A. Walker and R.W. Hoham (eds.). Snow ecology, pp. 229-265. Cambridge University Press, Cambridge.

Alford, R.A., and Richards, S.J. 1999. Global amphibian declines: A problem in applied ecology. *Annual Review of Ecology and Systematics,* 30:133-165.

Ballantyne, K. 2004. Monitoring *Plethodon cinereus*, the eastern red-backed salamander, in Kejimkujik National Park and Historic Site. B.Sc. Hons thesis, Dalhousie University, Halifax, Nova Scotia, Canada.

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

Behler, J.L., and King, F.W. 1979. The Audubon Society Field Guide to North American Reptiles and Amphibians. Alfred A. Knopf, Inc. New York, USA.

Bennett, R.M., Ross, R.M., Lellis, W.A., and Redell, L.A. 2003. Terrestrial salamander preference for artificial cover objects made from four species of wood. *Journal of the Pennsylvania Academy of Science*, 76: 77-79.

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

Brooks, R.T. 1999. Residual effects of thinning and high white-tailed deer densities on northern redback salamanders in southern New England oak forests. *Journal of Wildlife Management*, 63: 117-1180.

Bruhl, C.A., Schmidt, T., Pieper, S. and Alscher, A. 2013. Terrestrial pesticide exposure of amphibians: An underestimated cause of global decline? *Scientific Reports*, DOI: 10.1038/srep01135.

Burton, T.M., and Likens, G.E. 1975. Salamander populations and biomass in the Hubbard Brook experimental forest, New Hampshire. *Copeia*, 3:541-546.

Caruso, N.M., and Lips, K.R. 2012. Truly enigmatic declines in terrestrial salamander populations in Great Smoky Mountains National Park. *Diversity and Distributions*, 1-11.

Conant, R., and Collins, J.T. 1998. A field guide to reptiles and amphibians. Eastern and Central North America (3^{rd} edition). Houghton Mifflin Company, Boston, USA.

DeMaynadier, P.G., and Hunter, M.L. Jr. 1998. Effects of silvicultural edges on the distribution and abundance of amphibians in Maine. *Conservation Biology*, 12(2):340-352.

Droege, S., Monti, L., and Lantz, D. 1997. USGS terrestrial salamander monitoring program. http://www.mp2-pwrc.usgs.gov/sally/., Accessed: February 18, 2013.

Dupuis, L.A., Smith, J.N.M, and Bunnell, F. 1995. Relation of terrestrial salamander abundance to tree-stand age. *Conservation Biology*, 9(3):645-653.

Environment Canada. 2004. Threats to Water Availability in Canada. National Water Research Institute, Burlington, Ontario. NWRI Scientific Assessment Report Series No. 3 and ACSD Science Assessment Series No. 1. p.128.

Feder, M.E. 1983. Integrating the ecology and physiology of plethodontid salamanders. *Herpetologica*, 39(3):291-310.

Feder, M.E., and Londos, P.L. 1984. Hydric constraints upon foraging in a terrestrial salamander, *Desmognathus ochrophaeus* (Amphibia: Plethodontidae). *Oecologia*, 64(3):413-418.

Feder, M.E., and Pough, F.H. 1975. Temperature selection by the Red-backed salamander, *Plethodon c. cinereus* (Green) (Caudata: Plethodontidae). *Comparative Biochemistry and Physiology*, 50A:91-98.

Gabor, C.R. 1995. Correlational test of Mathis' hypothesis that bigger salamanders have better territories. *Copeia*, 1995(3):729-735.

Gibbs, J.P., and Karraker, N.E. 2006. Effects of warming conditions in eastern North American forests on Red-Backed Salamander morphology. *Conservation Biology,* 20(3):913-917.
Harding, J.H. 1997. Amphibians and Reptiles of the Great Lakes Region. University of Michigan Press. Ann Arbor, Michigan, USA.

Grant, E.H.C., and Nanjappa, P. 2006, Addressing error in identification of Ambystoma maculatum (spotted salamanders) using spot patterns. *Herpetological Review,* 37(1): 57-60.

Grizzell, R.A. jr. 1949.The hibernation site of three snakes and a salamander. *Copeia*, 1949:231-232.

Grover, M.C. 1998. Influence of cover and moisture on abudances of the terrestrial salamanders *Plethodon cinerus* and *Plethodon glutinosus. Journal of Herpetology*, 32(4):489-497.

Heatwole, H. 1962. Environmental factors influencing local distribution and activity of the salamander *Plethodon cinereus*. *Ecology*, 43(3):460-472.

Herbeck, L.A., and Larsen, D.R. 1999. Plethodontid salamander response to silvicultural practices in Missouri Ozark Forests. *Conservation Biology*, 13(3):623-632.

Hof, C., Araujo, M.B., Jetz, W., and Rahbek C. 2011. Additive threats from pathogens, climate and land-use change for global amphibian diversity. *Nature*, doi:10.1038/nature10650.

Hoff, J. G. 1977. A Massachusetts hibernation site of the red-backed salamander, Plethodon cinereus. *Herpetological Review*, 8:33.

Horne, E.A., and Jaeger, R.G. 1988. Territorial pheromones of female red-backed salamanders. *Ethology*, 78:143-152.

Jaeger, R.G. 1972. Food as a limited resource in competition between two species of terrestrial salamanders. *Ecology*, 53(3):535-546.

Jaeger, R.G. 1979. Spatial distributions of the terrestrial salamander *Plethodon cinereus*. *Herpetologica*, 35(1):90-93.

Jaeger, R.G. 1980. Microhabitats of a terrestrial forest salamander. *Copeia*, 2:265-268.

Jaeger, R.G., Schwarz, J., and Wise, S.E. 1995. Territorial male salamanders have foraging tactics attractive to gravid females. *Animal Behavior*, 49:633-639.

Knutson, M.G., Richardson, W.B., Reineke, D.M., Gray, B.R., Parmlee, J.R., and Weick, S. 2004. Agricultural ponds support amphibian population. *Ecological Applications,* 14(3):669-684.

Krebs, C.J. 2001. Ecology: the experimental analysis of distribution and abundance. (5th edition). Benjamin Cummings, San Francisco, USA.

Lamond, W.G. 1994.The Reptiles and Amphibians of the Hamilton Area: A Historical Summary and the Results of the Hamilton Herpetofaunal Atlas. Hamilton Naturalists' Club. Hamilton, Ontario.

Larson, A., Wake, D., and Devitt, T. 2006. Plethodontidae: lungless salamanders (version 26). Available at: http://tolweb.org/Plethodontidae *in* The Tree of Life Project, http://tolweb.org/ Accessed: February 18, 2013.

Lauer, A. Simon, M.A., Banning, J.L., Lam, B.A., and Harris, R.N. 2008. Diversity of cutaneous bacteria with antifungal activity isolated from female four-toed salamanders. *The ISME Journal*, 2:145-157.

LeClair, M.H., Levasseur, M., and LeClair, R. 2006. Life-history traits of *Plethodon cinereus* in the northern parts of its range: variations in population structure, age, and growth. *Herpetologica*, 62(3):265-282.

Leech, N.L., Barrett, K.C., and Morgan, G.A. 2008. SPSS for intermediate statistics use and interpretation (3rd edition). Lawrence Erlbaum Associates (Taylor Francis Group), New York, NY, USA.

Lotter, E., and Scott, N.J. 1977. Correlation between climate and distribution of the color morphs of the salamander *Plethodon cinereus*. *Copeia*, 1977:681-690.

Maerz, J.C. and Madison, D.M. 2000. Environmental variation and territorial behavior in a terrestrial salamander. Pp. 395–406. *In:* Bruce, R.C., Jaeger, R.G., and Houck. L.D. 2000. The biology of Plethodontid salamanders. Kluwer Academic/Plenum Publishers. New York, USA.

Marsh, D.M., and Goicochea, M.A. 2003. Monitoring terrestrial salamanders: biases caused by intense sampling and choice of cover objects. *Journal of Herpetology*, 37(3):460-466.

Marvin, G.A. 2001. Age, growth, and long-term site fidelity in the terrestrial Plethodontid Salamander *Plethodon kentucki*. *Copeia*, 1:108-117.

Mathis, A. 1990. Territoriality in a terrestrial salamander: The influence of resource quality and body size. *Behaviour*, 112: 162-178.

McCarter, J. 2009. Ecological monitoring at ***rare*** **Charitable Research Reserve**. Available on ***rare*** server.

Moore, A.L., Williams, C.E., Martin, T.H., and Moriarity, W.J. 2001. Influence of season, geomorphic surface and cover item on capture, size and weight of *Desmognathus ochrophaeus* and *Plethodon cinereus* in Allegheny Plateau riparian forests. *American Midland Naturalist*, 145: 39-45.

Moore, J.D., and Wyman, R.L. 2010. Eastern red-backed salamanders (*Plethodon cinerus*) in a highly acid forest soil. *American Midland Naturalist*, 16:95-105.

Monti, L., Hunter, M. and Witham, J. 2000. An evaluation of the artificial cover object (ACO) method for monitoring populations of the redback salamander *Plethodon cinereus*. *Journal of Herpetology*, 34:624-629.

Moreno, G. 1989. Behavioural and physiological differentitation between colour morphs of the salamander *Plethodon cinereus*. *Journal of Herpetology*, 23:335-341.

O’Donnell, K.M., and Semlitsch, R.D. 2015. Advancing terrestrial salamander population ecology: the central role of imperfect detection. *Journal of Herpetology*, 49(3) DOI: 10.1670/14-100.

Parks Canada. 2009. National Parks of Canada: Ecological Integrity. Available at: http://www.pc.gc.ca/progs/np-pn/ie-ei.aspx. Accessed: February 28, 2012.

Peterson, M.G., Gillette, J.R., Franks, R., and Jaeger, R.G. 2000. Alternative life styles in a terrestrial salamander: do females preferentially associate with each other? Pp. 417–429. *In:* Bruce, R.C., Jaeger,

R.G., and Houck, L.D. 2000. The biology of Plethodontid salamanders. Kluwer Academic/Plenum Publishers. New York, USA.

Riedel, B.L., Russell, K.R., Ford, W.M., O'Neill, K.P., and Godwin, H.W. 2008. Habitat relationships of eastern red-backed salamanders (*Plethoden cinereus*) in Appalachian agroforestry and grazing systems. *Agriculture, Ecosystems and the Environment,* 126:229-236.

Sayler, A. 1966. The reproductive ecology of the red-backed salamander, *Plethodon cinereus*, in Maryland. *Copeia*, 2:183-193.

Simons, R.R., Jaeger, R.G., and Felgenhaur, B.E. 1997. Competitor assessment and area defense by territorial salamanders. *Copeia*, 1:70-76.

Spotila, J.R. 1972. Role of temperature and water in the ecology of lungless salamanders. *Ecological Monographs*, 42(1):95-125.

Storey, K.B., and Storey, J.M. 1986. Freeze tolerance and intolerance as strategies of winter survival in terrestrially-hibernating amphibians. *Comparative Biochemistry and Physiology Part A*, 83(4):613-617.

Stuart, S.N., Chanson, J.S., Cox, N.A., Young, B.E., Rodrigues, A.S.L., Fischman, D.L., and Waller, R.W. 2004. Status and trends of amphibian declines and extinctions worldwide. *Science,* 306:1783-1786.

Styers, D.M., Chappelka, A.H., Marzen, L.J., and Somers, G.L. 2010. Scale matters: Indicators of ecological health along the urban-rural interface near Columbus, Georgia. *Ecological Indicators*, 10:224-233.

Sugalski, M.T., and Claussen, D.L. 1997. Preference for soil moisture, soil pH, and light intensity by the salamander, *Plethodon cinerus. Journal of Herpetology*, 31(2):245-250.

Trenham, P.C., Shaffer, B., Koenig, W.D., and Stromberg, M.R. 2000. Life history and demographic variation in the California Tiger Salamander (*Ambystoma californiense*). *Coepia,* 2:365-377.

Twery, M., and Gottschalk, K.W. 1996. Forest health: another fuzzy concept. *Journal of Forestry*, 94(8):20.

Van Wieren, J. 2003. Long Point world biosphere reserve *Plethodon cinereus* monitoring program: data analysis. Available at: http://longpointbiosphere.com/Publications/Salamander%20Monitoring%20Data%20Analysis.pd

Venesky, M.D. and Anthony, C.D. 2007. Antipredator adaptations and predator avoidance by two colour morphs of the eastern red-backed salamander *Plethodon cinereus*. *Herpetologica*, 63:450-458.

Walton, B.M. 2013. Top-down regulation of litter invertebrates by a terrestrial salamander. *Herpetologica,* 69(2):127-146

Welsh, H.H. Jr., and Droege, S. 2001. A case for using plethodontid salamanders for monitoring biodiversity and ecosystem integrity of North American forests. *Conservation Biology*, 15(3):558-569.

Welsh, H. H., Jr., and Hodgson G.R. 2013. Woodland salamanders as metrics of forest ecosystem recovery: a case study from California's redwoods. *Ecosphere* 4(5):59. http://dx.doi.org/10.1890/ES12-00400.1

Whitford, W.G., and Vinegar, A. 1966. Homing, survivorship, and overwintering of larvae in Spotted salamanders, *Ambystoma maculatum*. *Copeia*, 1966(3):515-519.

Wise, S.E., and Jaeger, R.G. 1998. The influence of tail autonomy on agnostic behaviour in a territorial salamander. *Animal Behaviour*, 55:1707-1716.

Wyman, R.L. 1988. Soil acidity and moisture and the distribution of amphibians in five forests of southcentral New York. *Copeia*, 1988(2):394-399.

Wyman, R.L. 1998. Experimental assessment of salamanders as predators of detrital food webs: effects of invertebrates, decomposition and carbon cycle. *Biodiversity Conservation*, 7:641-650.

Wyman, R.L., and Hawksley-Lescault, D.S. 1987. Soil acidity affects distribution, behavior, and physiology of the salamander Plethodon cinerus. E*cology*, 68:1819-1827.

Zar, J.H. 1999. Biostatistical analysis (3rd edition). Prentice Hall, Upper Saddle River, New Jersey, USA.

Zorn, P., Blazeski, V., and Craig, B. 2004. Joint EMAN/Parks Canada national monitoring protocol for plethodontid salamanders. *Ecological Monitoring and Assessment Network*. Available at: http://www.ec.gc.ca/faunesciencewildlifescience/default.asp?lang=En&n=9C4F74C5-1.

4.0 Forest Canopy and Tree Biodiversity Monitoring

Prepared by: Tim Skuse

4.1Introduction

4.1.1 Forest Health Monitoring

Forests are critical to environmental health and stability (Environment Canada and Canadian Forest Service 2004). They house a significant amount of the world's biodiversity and provide numerous ecosystem services such as; soil conservation, water cycling, and air quality mediation (Butt 2011). Establishing global policies and protocols related to the safeguarding of forests are of high priority.

In southern Ontario, forests have experienced a great deal of change in the past 200 years. Prior to European settlement, southern Ontario was largely covered by a patchwork of deciduous and mixed hardwood forests (Ontario Ministry Natural Resources 1999). Due to rapid development and land use changes, forest species have been removed and land cover has been significantly altered. What remains are highly fragmented forests which are much smaller in size than they were historically (Waldron 2003). Forests are also under pressure from many other biotic and abiotic factors. Widespread invasive species have caused drastic changes to forest stand composition and forest nutrient cycles, threatening to alter the ecology of forest systems profoundly (Moser et al. 2009). Impacts to forests from climate change are thought to be equally far-reaching (Allen et al. 2010). Natural disturbances to forests from insects and disease will become more severe with warmer climates (Weed et al. 2013). Forests will also have to adapt to more instances of extreme weather such as storms and drought (Allen et al. 2010). These factors demonstrate the number of pressures impacting our forests and highlight the need to monitor the health of our remaining forest stands.

Establishing long-term ecological monitoring across a network of forest sites can help develop a more thorough understanding of baseline levels of both variability and health in natural systems (Gardner 2011). Monitoring crown conditions and stem defects is essential to detect early warning signs and recognising changes in tree health of Canadian forests and Canada's urban areas (Environment Canada and Canadian Forest Service 2004). Records of tree damage and mortality can help with identifying and understanding the causes and effects of tree and forest decline. Information on populations and species decline can be used as a platform to launch conservation initiatives (Gardner 2011), and may influence management objectives when considering human-impact on forests.

Although the age, diversity, and overall health of a forest stand can be derived from canopy tree monitoring, it says little about the likely successional trajectory of the stand. Beneath the canopy, the rate of sapling recruitment and survivorship in the shrub and small tree stratum can be informative of the health and progression of a forest stand (Roberts-Pichette & Gillespie 1999). Shrub and small tree monitoring can provide valuable insight into the successional direction of a forest stand by observing saplings that may eventually be a part of the forest canopy. Historical records can aid in understanding a forest's past dynamics and structure, while ongoing, long-term monitoring of both canopy tree and shrub/small tree forest strata can shed light on the present influential factors affecting its development. Together, these can contribute to effective long-term best management practices that have been developed to meet the challenges of dynamic forest ecosystems.

*4.1.2 EMAN Forest Monitoring at **rare***

With the rapid development of southern Ontario, there are very few undisturbed remnant old-growth forests remaining (Ontario Ministry Natural Resources 1999). At the ***rare*** **Charitable Research Reserve**, one such remnant old growth exists: a Sugar Maple-American Beech (*Acer saccharum – Fagus grandifolia*) dominated forest named Indian Woods, which has trees more than 240 years old. Additional forest stands at ***rare*** include the Cliffs and Alvars, a mixed deciduous forest that was partially grazed by cattle within the last century, and the Hogsback, a relatively undisturbed mixed swamp forest. All of these forest ecosystems contribute invaluable services to the region by sequestering carbon dioxide and improving air and water quality (Führer 2000), as well as providing increasingly uncommon habitat to countless plants and animals that require mature forest interior (Ontario Ministry of Natural Resources 1999).

Forests face diverse challenges in the landscape of Waterloo Region; ***rare*** is bordered by conventional farm fields, aggregate mining operations, subdivisions, and busy roads. Many of these neighbouring lands are scheduled for drastic changes and development within the next few years. By acquiring baseline records of the conditions of the ***rare*** forests and continuing long-term monitoring, changes in the forest stands may be detected early, allowing for the development and implementation of an effective management plan to protect ***rare*** forest ecosystems.

The research questions being addressed with long-term forest canopy tree biodiversity monitoring were identified at the establishment of the program (McCarter 2009) and subsequent questions were asked based on new objectives established in 2013:

1. What is the current state (biodiversity, composition, health) of ***rare's*** forests, and how do they compare to one another?
2. What are the long-term trends in tree mortality, recruitment, and replacement taking place within the forests at ***rare***?
3. Is the ecosystem integrity of the forests being maintained or improved under ***rare*** management?
4. Is either the ecological health or integrity of ***rare*** forests being affected by on-site and nearby changes in land use (i.e. restoration, agriculture, residential development, and aggregate extraction)?
5. How does the canopy tree stratum influence the species composition of the shrub and small tree stratum?
6. What is the most likely successional trajectory as suggested by the recruitment and mortality rates of saplings in the forests at ***rare***?

The forest canopy tree biodiversity monitoring program at the ***rare*** **Charitable Research Reserve** began in 2009 with the establishment of three plots in the Cliffs and Alvars forests and three plots in the Indian Woods. Preliminary monitoring data, such as trees species, location within the plot, and diameter at breast height (dbh), were collected in this first year. In the 2010 monitoring year, three plots were established in the Hogsback forest so that all three major forested areas on the ***rare*** property would be represented in the monitoring program. An Ecological Monitoring and Assessment Network (EMAN) Tree Health Protocol was added to the monitoring program in 2010, and all nine forest plots have been monitored in full each subsequent year. In 2013, a shrub and small tree monitoring program was added as a pilot study to the existing protocol at ***rare***. Based on the results of the pilot study, a more tailored shrub and small tree monitoring program specific to this forest stratum was developed and implemented in 2014. Shrub and small tree monitoring is to occur every five years with 2014 as the initial year. Methodology, results and discussion for shrub and small tree monitoring can be found in the Forest Canopy and Tree Biodiversity report from 2014 and on the ***rare*** server.

4.2 Methods

4.2.1 Forest Plot Locations

Forest biodiversity monitoring plots are established in three forest stands on ***rare*** property. Each of these stands contains three monitoring plots, which together are used to describe their respective stands.

Cliffs and Alvars: A mature Sugar Maple-American Beech dominated forest located on the north side of Blair Road, bordered by Cruickston Creek on the west, Newman Creek on the east and the Grand River to the north. The three plots in the Cliffs and Alvars forest are located approximately 50m north of the Grand Trunk Trail, arranged parallel to the trail (Appendix A). To access these plots, walk from the ECO Centre to the Grand Trunk Trail. Follow the Grand Trunk Trail to the east (right) until completely under the canopy (approximately 200m). Shortly after, the forest opens up and a small seasonal trail heads north towards the river. The plots are located to the left and right of this trail, past the large fallen trees. Plot corners are marked with pigtail stakes and orange or pink flagging tape.

Indian Woods: A remnant old-growth forest located south of Blair Road and north of Whistle Bare Road, on the west side of the property. The three forest plots in Indian Woods are oriented in a north-south line in the centre of the forest, approximately 100m east of the Grand Allée. The third plot can be accessed by turning east into the forest off the Grand Allée towards the salamander monitoring plot and continuing to the top of the hill overlooking the pond. The first and second plots can be found by heading north from the third plot (Appendix A). The plots are approximately 30m apart and the flagging tape on the corners of each plot should be visible from the adjacent plot.

Hogsback: Located at the south-west corner of the property, the Hogsback is bisected by Cruickston Creek and bordered by the Newman Drive subdivision to the west. The Hogsback is a mixed swamp forest with upland ridges dominated by White Pine, Red Maple, American Beech, and Sugar Maple. The three forest biodiversity plots were established on these elevated ridges as the lower areas will likely be too swampy to access in wetter years. The second forest plot overlaps with the Hogsback salamander monitoring plot. The first plot is found approximately 30m north of the second plot on the same elevated ridge, and the third plot is located 30m southwest of the second plot, separated by a small boggy area (Appendix A). This area can be accessed by driving east down South Gate Road to edge of the forest stand, and following the hedgerow around the forest (north, east, north, east). Alternatively, the site can be accessed by parking at Springbank Gardens, turning south at the pavilion, travelling south along the small hedgerow, then east along the forest perimeter. The forest can be entered at part of fence lowered with a fallen log, at the southern edge of Hogsback Field (303).

4.2.2 Plot Establishment

Following the EMAN Forest Canopy Tree Biodiversity Monitoring Protocol (Environment Canada and Canadian Forest Service 2004), the plots established in 2009 and 2010 at ***rare*** are permanent 20m x 20m plots located in the forest interior. According to EMAN, plots should not be closer than three times the average tree height to any forest edge (estimated at 90m-100m for our forests); however this was not always possible due to the small size of Indian Woods and swampy topography of the Hogsback; in these cases, plots were established as far from any edge as possible. The plots were oriented along the cardinal directions and the corners were marked with galvanized steel pigtail stakes with labelled flagging tape (Figure 4.1). All trees within the plot with a diameter equal to or greater than 10cm at breast height (dbh) were included in the monitoring. Trees in Indian Woods and Hogsback were labelled with pigtail stakes inserted in the ground at the base of the tree with pre-printed aluminum tags attached. The trees in the Cliffs and Alvars forest plots were originally marked with forestry tags, each with unique identification codes (ex. CA-02-08, Cliffs and Alvars – Plot 2 – Tree 8) which were fixed to the tree with a downward angled nail. In 2013, these forestry tags were removed from the trees in Cliffs and Alvars and were replaced with steel pigtails with numbered aluminum tags in a manner consistent with Indian Woods and Hogsback.

The trees were tagged in a clockwise spiral inward from the northwest corner of the plot. The species of each tree was recorded at the time of plot establishment, and its distance to two plot corners was recorded for plot map generation. In this plotting technique, one observer stands with their back to the tree, facing the nearest line (i.e. edge) of the plot. The line number was recorded, and the “A” distance and “B” distance were measured; “A” distance was measured from the tree to the corner to the right-hand side of the observer facing the line, while the “B” distance was measured from the tree to the corner to the left-hand side of the observer (Figure 4.1). Trees that split into multiple stems under breast height had each stem measured independently.

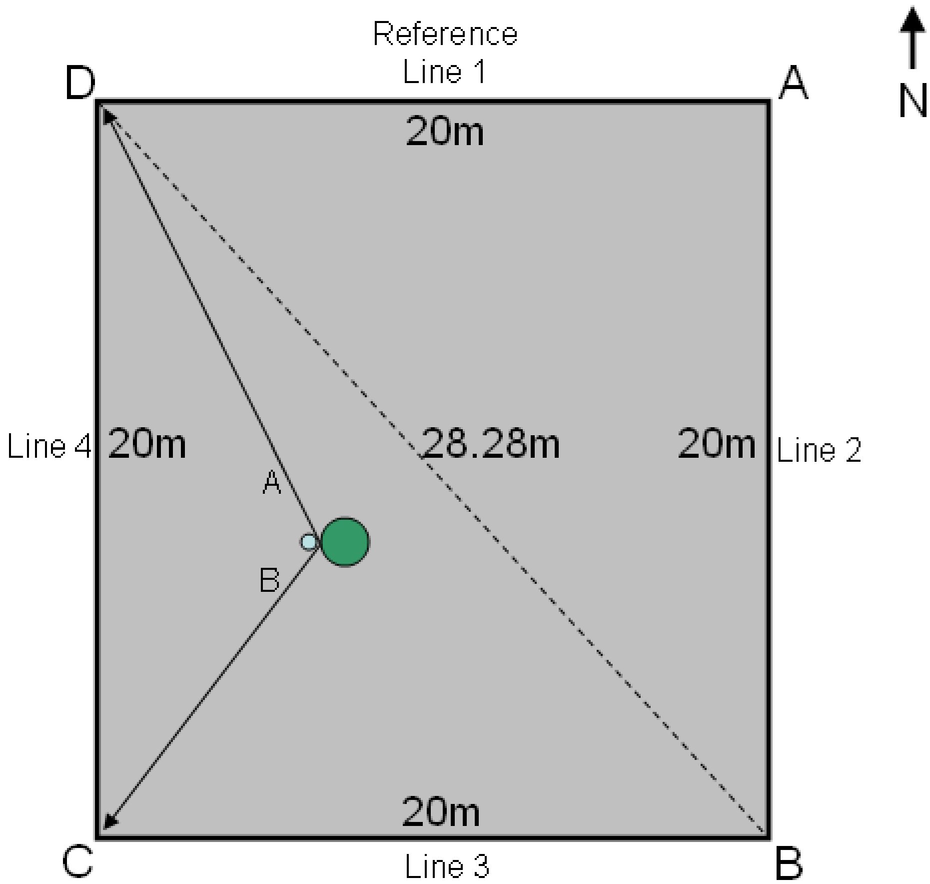


Figure 4.1: Diagram of an EMAN forest canopy tree biodiversity plots from McCarter 2009. The A and B distances are used to map the position of the tree within the plot. The A distance is measured from the tree to the corner to the right of the observer standing facing the reference line. The B distance is measured to the corner on the left side of the observer.

4.2.3 Monitoring Procedure: Canopy-Tree Monitoring

Each plot is visited once annually, ideally in the summer or when leaves are still present on trees for ease of identification and canopy assessments. In 2015, forest plots were visited September 8th, 18th and 28th for Cliffs and Alvars, September 22nd, September 29th, and October 1st for Indian Woods, and September 24th and 25th in the Hogsback. The following variables were recorded for each tree in the monitoring plots: diameter at breast height (Woven Fibre Glass 5m Diameter Tape, Richter Measuring Tools), tree height (Haglöf Electronic Clinometer & Mastercraft© Fibre glass measuring tape), and tree condition based on Environment Canada and Canada Forestry Services EMAN codes (Table 4.1). Tree health was monitored by recording stem defects, crown class, crown rating (Table 4.2), and any other health notes, again based on Environment Canada's EMAN protocol. Marginal trees in each plot were checked to see if they had graduated into the 10cm dbh size class (minimum for inclusion). Trees that had newly met minimum requirements were tagged in a manner consistent with their plot and measured into the plot using distance from adjacent corners as described above. Initially, all trees were plotted into BioMon (BioMon *for* Windows Suite Version 2), a biodiversity monitoring software package, to generate tree species maps for each forest plot (Appendix A). BioMon software is now outdated and no longer compatible with recent operating systems. No program has been identified as a suitable substitute for plotting trees and new additions since 2011 have been placed manually onto the original forest plots with best estimates of location and size.

Table 4.1: Tree condition codes from EMAN protocol (Environment Canada and Canada Forestry Service 2004)

Code	Condition
AS	Alive Standing
AB	Alive Broken
AL	Alive Leaning
AF	Alive Fallen/Prone
AD	Alive Standing with Dead Top
DS	Dead Standing
DB	Dead Broken
DL	Dead Leaning
DF	Dead Fallen/Prone

Table 4.2: Crown class and rating codes from EMAN protocol (Environment Canada and Canada Forestry Service 2004).

Crown Class	Code	Crown Rating
Dominant: Crown extends above the general canopy level and receives full sunlight from above and partly from the sides; larger than the average trees in the stand	**1**	**Healthy**: Appears in good health, no major branch mortality, <10% branch/twig mortality
Co-dominant: Crown forms the general canopy level and receives full sunlight from directly above and comparatively little from the sides	**2**	**Light-Moderate Decline**: Branch and twig mortality <50% of the crown, <50% branch/twig mortality
Intermediate: Shorter than the two preceding classes, and receiving little direct sunlight from above and from the sides; their crowns extend into the base of the canopy of the dominant and co-dominant trees	**3**	**Severe Decline**: Branch and twig mortality >50% of the crown, >50% branch/twig mortality
Suppressed: Receives no direct sunlight from above or the sides, their crowns are entirely below the general level of the crown cover.	**4**	**Dead, Natural**: Tree is dead; either standing or downed

Open: Exposed to full sunlight from directly above and on all sides; typically growing in a field or along a boulevard.	**5**	**Dead, Human**: Tree cut down, removed, or girdled

4.2.4 Data Analysis

All data were analyzed using SPSS Statistics Version 20 and Microsoft Excel 2010. For each location, summary statistics were calculated by combining the data from the three plots which together represent the forest stand. For each stand, the number of trees present, the number of species present, the mean diameter at breast height, and the total basal area (sum of cross sectional area of all trees within a plot, based on dbh measurements) were recorded. These data were used to calculate the Shannon Diversity Index (H) and Species Evenness Value (E_H) for each forest stand. The relative density, relative frequency, and relative dominance were also calculated, and results were combined to give an Importance Value to each species within each stand (Roberts-Pichette & Gillespie, 1999). Only living trees were included in these calculations; formulas used for all calculations are found in Figures 2.1 and 4.2 to 4.5. Measures of forest health were also included, such as presence of select diseases, trends in canopy health, rates of mortality and recruitment, and individual species health. Canopy health trends were calculated with crown ratings given to trees proportional to numbers of total trees. Only three categories were used for these trends; dead, trees in severe decline, and a combination category of healthy and light-moderate decline. Healthy and light to moderate decline were included together due to the inconsistency of crown rating estimates between years and the high possibility for a tree to recover even if it is in moderate (up to 50%) decline (Campbell and Valentine 1972).

In addition to the summary statistics, a univariate analysis of variance (ANOVA) was used to investigate differences between size classes at each forest stand. Trees were assigned to one of eight size classes based on their dbh measurements in meters (0.1-0.19, 0.2-0.29, 0.3-0.39, 0.4-0.49, 0.5-0.59, 0.6-0.69, 0.7-0.79, 0.8+; hereon referred to as size class 1 through 8). For each forest stand, data from 2009 to 2015 were used to conduct the ANOVA, where year and size class were the independent variables and tree abundance was the dependant variable. When results were significant, a Tukey HSD Post-hoc test was used to determine where the differences existed. In previous years, a combination variable for size class and location was used because of an interaction between these two variables (Leech et al. 2008). However, since size class differences *within* a forest stand is of more interest than differences *across* forest stands (and in order to simplify the analysis), a separate ANOVA was performed for each forest stand, and the combination variable was not necessary. An alpha level of 0.05 was used to determine significance.

$$\textbf{Relative Density} = \frac{\text{Number of tree species A in plots}}{\text{Total number of trees in the plots}}\ x100$$

Figure 4.2: Formula for calculating the relative density of tree species in a forest stand, with all three plots per stand combined.

$$\textbf{Relative Frequency} = \frac{\text{Frequency of species A in plots}}{\text{Total frequency of in all trees in the plots}} \text{ x } 100$$

$$\text{Where: Frequency} = \frac{\text{number of plots with species A}}{\text{total number of plots in the stand}}$$

Figure 4.3: Formula for calculating the relative frequency of tree species in a forest stand, with all three plots per stand combined.

$$\textbf{Relative Dominance} = \frac{\text{Basal area of species A } (\text{m}^2)}{\text{Total basal area of all species in the plots } (\text{m}^2)} \text{ x}100$$

Figure 4.4: Formula for calculating the relative dominance of tree species in a forest stand, with all three plots per stand combined.

$$\textbf{Importance Value} = \text{Relative Density} + \text{Relative Frequency} + \text{Relative Dominance}$$

Figure 4.5: Formula for calculating the importance value of each tree species in a forest stand.

4.3.0 Results

4.3.1 Canopy Tree Monitoring: Abundance and Dominance

In 2015, the Cliffs and Alvars forest plots contained five tree species from five families (Figure 4.7). Sugar Maple and American Beech continue to be the dominant species in this forest for all plots and there was significantly more of these two species (p<0.001). Sugar Maple and American Beech have been the two dominant species since the start of the monitoring program in 2009. One new tree mortality was recorded for Cliffs and Alvars in 2015, a White Ash (*Fraxinus americana*); the last of three White Ash alive at the start of the monitoring program in 2009. Shannon Diversity Index for 2015 is the lowest on record at 1.34 and species evenness has increased to the highest value on record at 0.833 (Table 4.3).

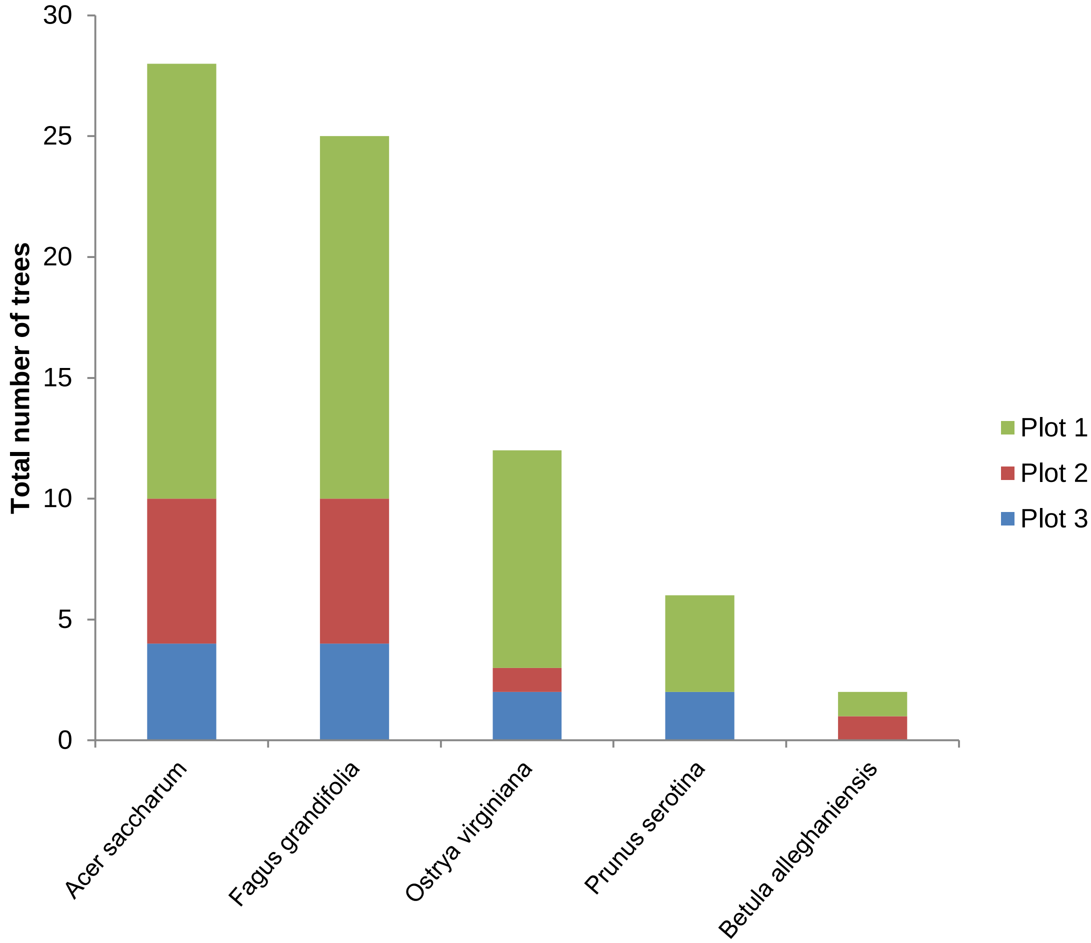


Figure 4.6: Tree species composition and abundance for each of the three forest plots in the Cliffs and Alvars.

Indian Woods forest plots had the lowest number of species with only four different species present from two different families (Figure 4.8). Indian Woods experienced more change between 2014 and 2015 than between any other year with four mortalities and two new recruits. Three mortalities occurred in Indian Woods Plot One where a large Sugar Maple fell onto two trees in the plot. New recruits were also from Plot One. Sugar maple is the dominant species for Plot Two and Plot Three and was previously the dominant species in Plot One. American Beech has now over taken Sugar Maple in Plot One as the most dominant species, but overall there are significantly more sugar maples than any other species (p<0.001). The Shannon Diversity Index and Species Evenness were the highest ever in 2015 at 0.848 and 0.611, respectively (Table 4.3).

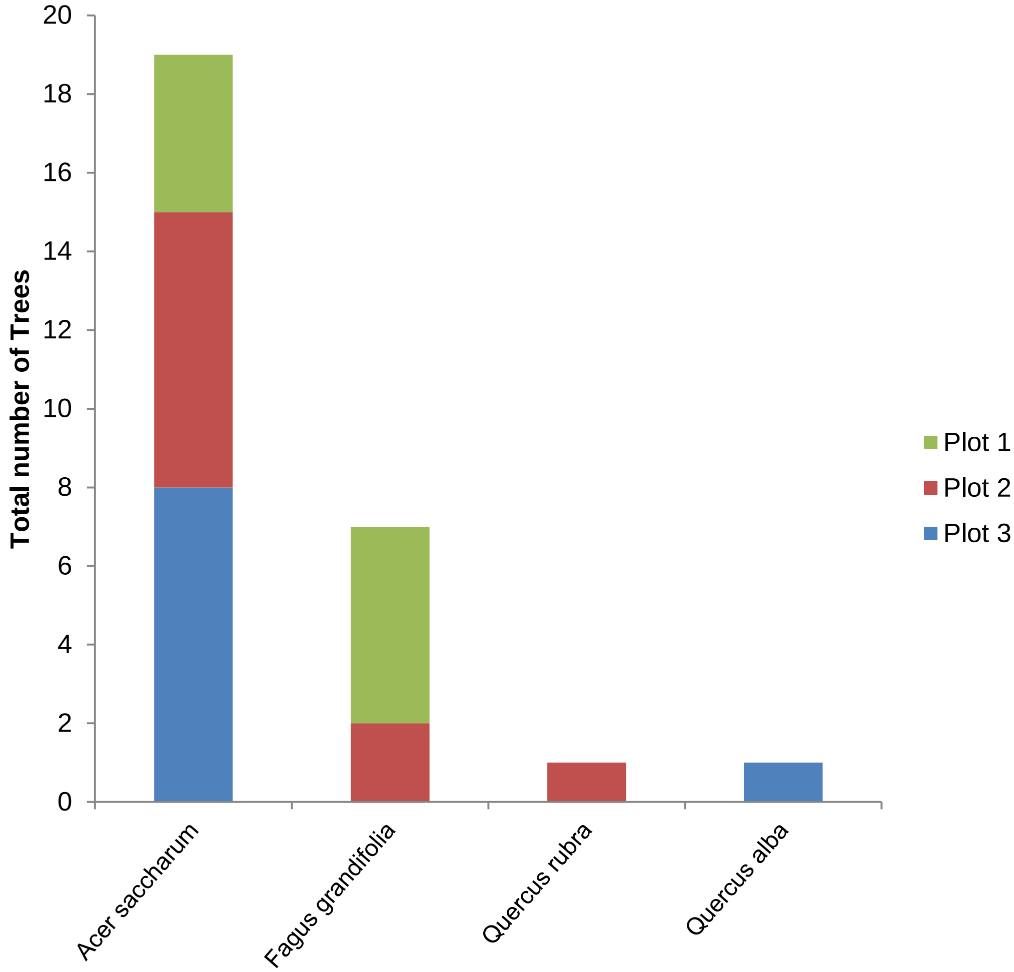


Figure 4.7: Tree species composition and abundance for each of the three forest plots in Indian Woods.

The Hogsback Forest has consistently has the highest species abundance across all forest stands, with ten different species representing six different families (Figure 4.9). Sugar Maple was the dominant species for the forest stand as a whole, but American Beech and Hop Hornbeam (*Ostrya virginiana*) were both dominant species in individual plots. There was significantly more Sugar Maple than every species except for American Beech and Red Maple (p<0.05). One new mortality was recorded in 2015, which was a Green or Red Ash*. Shannon Diversity Index values were the lowest for any monitoring period at 2.026 and have been on a gradual decline since the start of monitoring in 2010. A similar trend is seen in species evenness, but 2015 had an evenness of 0.88, up from last year's 0.855.

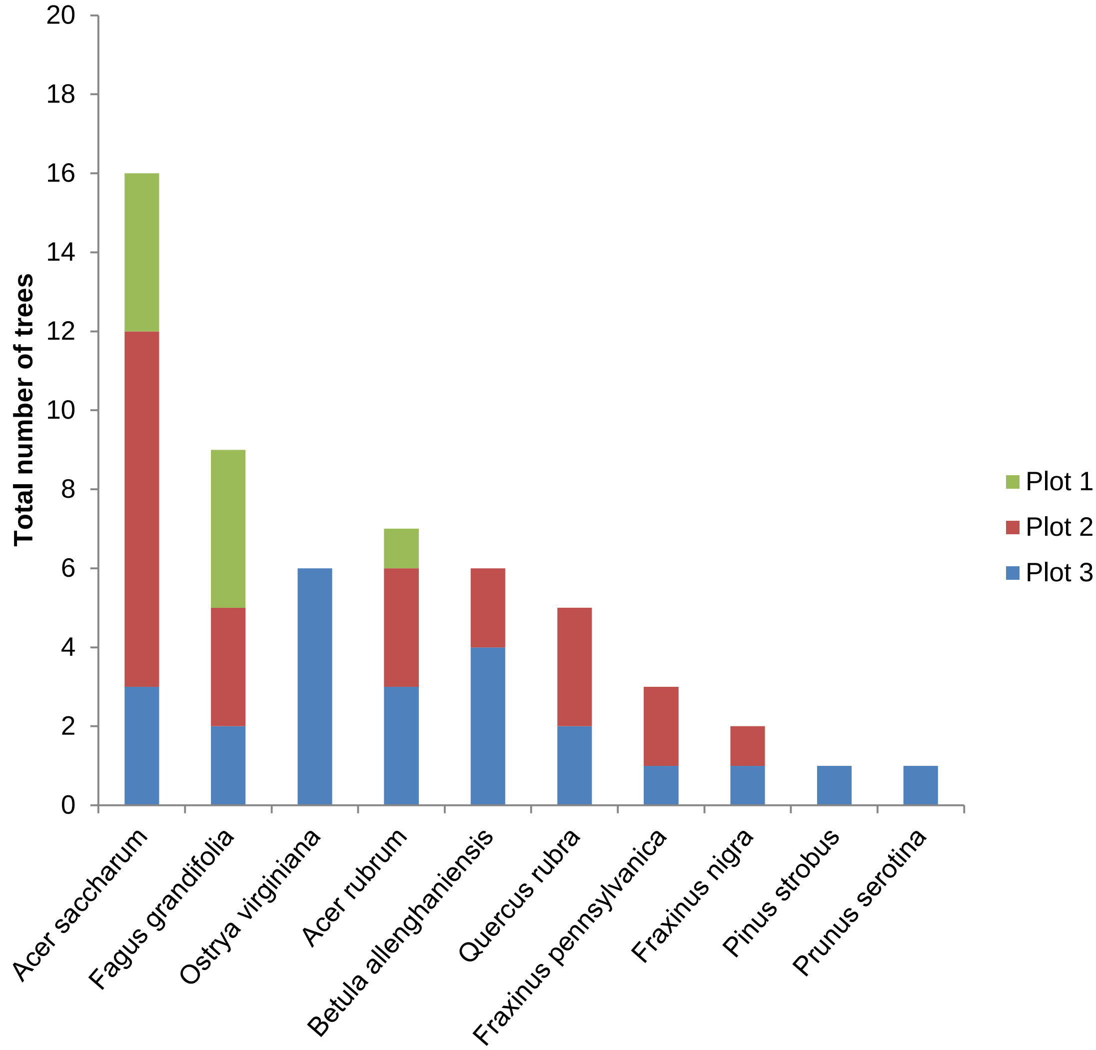


Figure 4.8: Tree species composition and abundance for each of the three forest plots in the Hogsback.

*Note that due to the difficulty in distinguishing between Green and Red Ash and their similar ecological role, they are included as one species in this report as *Fraxinus pennsylvanica*. This lack of distinction is also used in reports by other conservation authorities (Credit Valley Conservation 2009).

Table 4.3: Summary of forest monitoring plot observations with numbers of live and dead trees, number of species, mean dbh, Shannon-Weiner Diversity Index and evenness value for each forest stand. All three plots from each forest stand are included in the calculations.

		Measures					
		Number of Live Trees	Number of dead trees	Number of Species	Mean Stem dbh (cm)	Shannon-Weiner Diversity Index	Species Evenness Value
Cliffs and Alvars	2009	48	7	7	23.07	1.487	0.83
	2010	50	6	7	23.34	1.56	0.8
	2011	49	8	7	23.3	1.479	0.76
	2012	49	9	6	23.4	1.404	0.784
	2013	49	9	6	23.4	1.404	0.784
	2014	48	10	6	23.9	1.414	0.789
	2015	47	11	5	24.3	1.34	0.833
Indian Woods	2009	34	4	5	32.97	0.843	0.524
	2010	32	7	4	32.11	0.746	0.538
	2011	32	7	4	32.3	0.746	0.538
	2012	29	10	4	33.1	0.792	0.571
	2013	31	10	4	32.9	0.761	0.549
	2014	30	10	4	33.3	0.776	0.56
	2015	28	14	4	30.1	0.848	0.611
Hogback	2010	54	6	10	24.92	2.077	0.902
	2011	54	6	10	25.1	2.077	0.902
	2012	54	6	10	24.49	2.077	0.902
	2013	56	6	10	25.3	2.052	0.891
	2014	57	6	10	24.7	2.039	0.855
	2015	56	7	10	25.2	2.026	0.88

4.3.2 Canopy Tree Monitoring: Stand Characteristics and Size Class Abundance

Across all monitoring stands, Sugar Maple had the highest Importance Value. The Importance Value, along with the Abundance, Basal Area, Relative Density, Relative Frequency, and Relative Dominance for all species present in the three forest stands can be found in Table 4.4.

When investigating the differences in the number of trees within each size class by year, it was found that monitoring year did not have a significant effect (p=0.967). However, within each forest stand, there were significant differences across size classes. It is important to note that the abundance of trees represents an average abundance across all monitoring years (Figure 4.10).

In the Cliffs and Alvars, the mean abundance of trees in size class 1 was significantly greater than all other size classes (p<0.001). Indian Woods also had the highest mean abundance of trees in size class 1, however this was only significantly greater from that of size class 5, 6, 7 and 8 (p<0.01). Size class 3 in Indian Woods had the same trend as 1 being significantly greater than that of size class 5, 6, 7, and 8 (p<0.01). The majority of trees in the Hogsback were classified in size class 1 or 2. The mean abundance of trees in size class 1 was significantly greater than that of all other size classes (p<0.001). In addition, the mean abundance of trees in size class 2 was significantly greater than that in size class 3 through 8 (p<0.05).

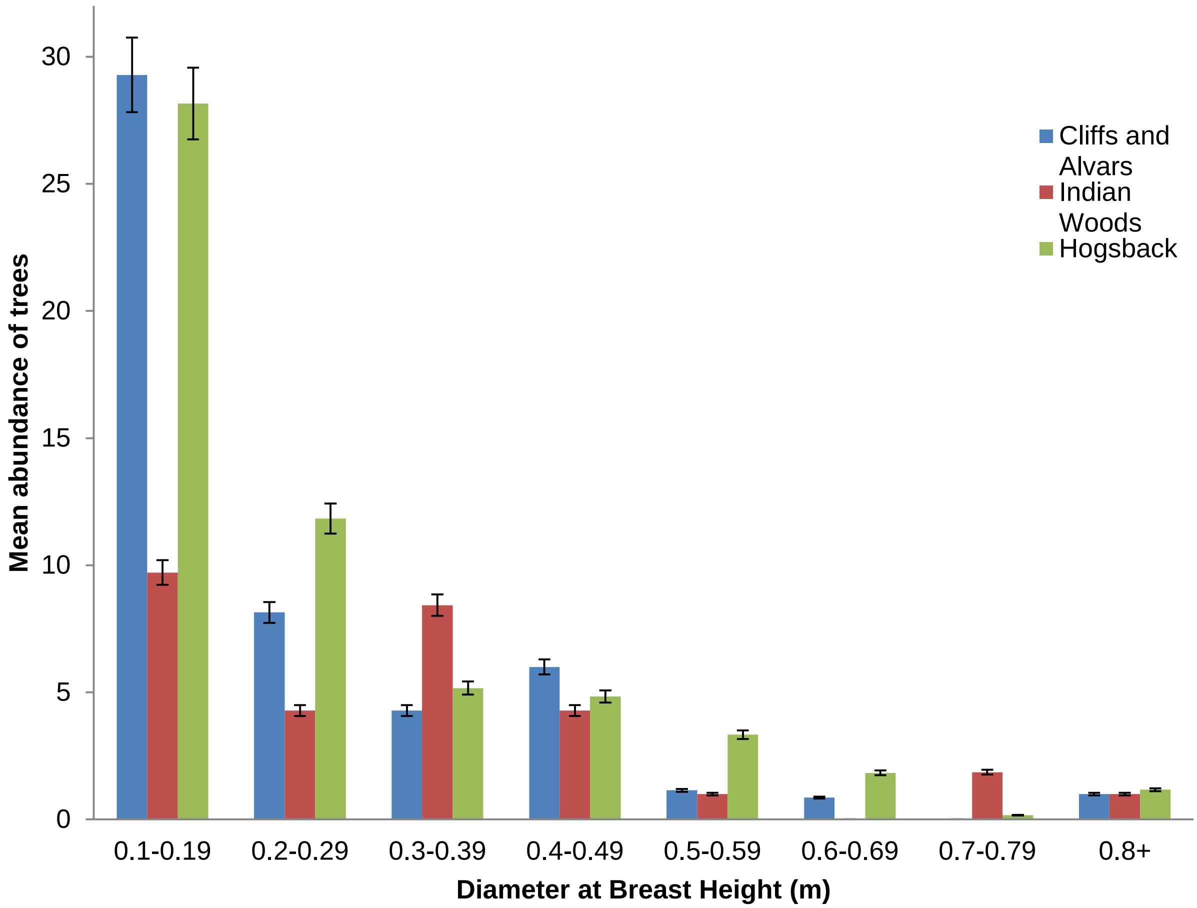


Figure 4.9: Mean size class distribution of all trees measured during forest health monitoring at rare from 2009-2015. Error bars represent Confidence Intervals at 95%.

Table 4.4: 2015 tree species composition and summary statistics for the three forest stands monitored at ***rare***.

Location	Species name	Abundance	Basal Area (m2)	Relative Density	Relative Frequency	Relative Dominance	Importance Value
Cliffs and Alvars	*Acer saccharum*	18	1.61	36.73	25.00	49.06	110.79
	Betula alleghaniensis	1	0.16	2.04	8.33	4.78	15.16
	Fagus grandifolia	15	1.16	34.69	25.00	35.43	95.12
	Ostrya virginiana	4	0.11	18.37	25.00	3.20	46.57
	Prunus serotina	4	0.25	8.16	16.67	7.53	32.36
Indian Woods	*Acer saccharum*	19	1.81	67.86	42.86	64.12	174.83
	Fagus grandifolia	7	0.22	25.00	28.57	7.69	61.26
	Quercus alba	1	0.17	3.57	14.29	6.11	23.96
	Quercus rubra	1	0.62	3.57	14.29	22.09	39.94
Hogsback	*Acer rubrum*	7	1.16	12.28	15	28.49	55.77
	Acer saccharum	16	0.94	28.07	15	23.13	66.20
	Betula alleghaniensis	6	0.16	10.53	10	3.90	24.42
	Fagus grandifolia	9	1.17	15.79	15	28.90	59.69
	Fraxinus nigra	2	0.02	3.51	10	0.39	13.90
	Fraxinus pennsylvanica	3	0.12	5.26	10	3.08	18.34
	Ostrya virginiana	6	0.11	12.28	5	2.75	20.03
	Pinus strobus	1	0.01	1.75	5	0.28	7.04
	Prunus serotina	1	0.06	1.75	5	1.59	8.35
	Quercus rubra	5	0.30	8.77	10	7.48	26.25

4.3.3 Measures of Tree Health

Mortalities for co-dominant and dominant trees for each forest area show trees in Indian Woods have declined at a greater rate than in the other two forest areas (see Table 4.5). This is reflected in the total mortalities for Indian Woods, which have also been substantially higher, however Indian Woods has also had the greatest number of new recruit trees since 2009 (see Table 4.6). Cliffs and Alvars has had an average 2.12% loss of dominant and co-dominant trees over the course of monitoring and has experienced more tree mortality than recruitment. As the Hogsback has only had one new dominant or co-dominant tree mortality since 2009, the change is only a fraction of the other two forested areas. The Hogsback is also the only forest to have gained more new trees than it has lost.

Table 4.5: Change in number of living dominant and co-dominant trees between each year over the course of the monitoring period. Note that numbers of dominant and co-dominant trees were calculated from crown rating assessments made in 2015 as crown assessments are highly inconsistent between years.

	Indian Woods			Cliffs and Alvars			Hogsback		
	Dominant/Co-Dominant Trees	Newly Dead	% Change	Dominant/Co-Dominant Trees	Newly Dead	% Change	Dominant/Co-Dominant Trees	Newly Dead	% Change
2014-2015	16	3	15.75	29	1	3.33	31	1	3.13
2013-2014	19	0	0	30	1	3.23	32	0	0
2012-2013	19	0	0	31	0	0	32	0	0
2011-2012	19	1	5.0	31	1	3.13	32	0	0
2010-2011	20	0	0	32	1	3.03	32	0	0
2009-2010	20	3	13.04	33	0	0	32	0	0
	Average Change		5.64	Average Change		2.12	Average Change		0.52

Table 4.6: Number of new recruit trees and mortalities between each monitoring year. New recruits are trees that have grown to be at least 10cm dbh.

	Indian Woods		Cliffs and Alvars		Hogsback	
	New Recruits	Mortalities	New Recruits	Mortalities	New Recruits	Mortalities
2009-2010	1	3	1	0	No data	No data
2010-2011	0	0	0	1	0	0
2011-2012	0	3	1	1	0	0
2012-2013	2	0	0	0	2	0
2013-2014	0	1	0	1	1	0
2014-2015	2	4	0	1	0	1
Totals	5	11	2	4	3	1

Trends in crown health for forest plots overall show no major changes since 2010 when crown rating data was gathered for the first time (see Figure 4.11). There has been a 6% decrease in the number of trees with healthy/light-moderate decline since 2010. Gradual upward trends in numbers of dead trees and gradual downward trends of healthy trees of are to be expected as the trend lines were calculated with the total number of trees observed since the beginning of monitoring. Overall, severe decline has remained nearly stagnant for the majority of the monitoring period.

Examining each forest area individually shows slightly different results. For crown health in the Hogsback, all ratings have stayed mostly even over the course of the monitoring with only a 1% decrease in numbers of healthy trees since 2010. Cliffs and Alvars have had a slight downward trend in overall numbers of healthy trees, with a 4% decrease in healthy trees since 2010. The greatest changes have happened in Indian Woods, with a 15% decrease in the number of trees in healthy/light-moderate decline and a 15% increase in the number of dead trees.

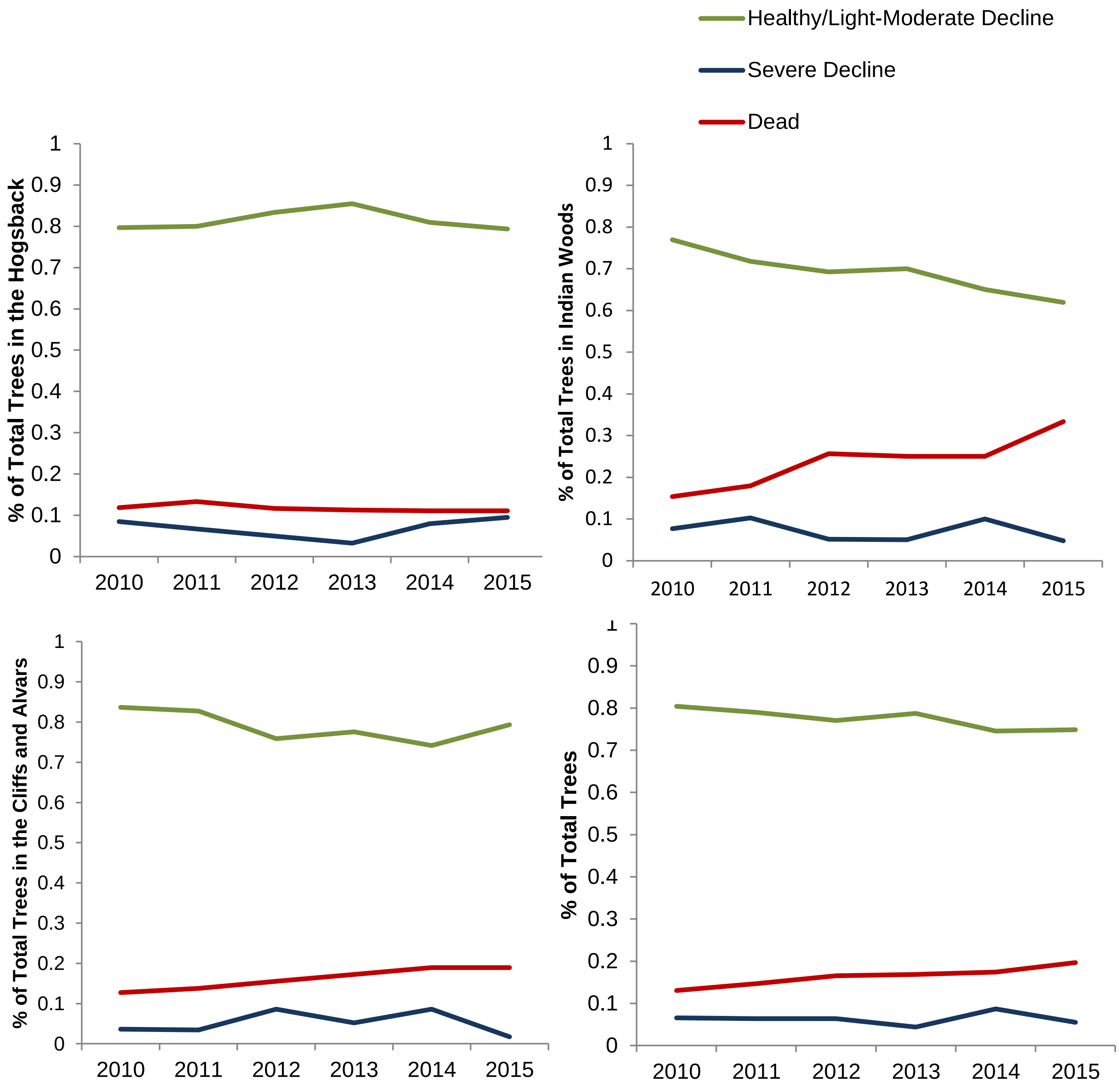


Figure 4.10: Comparison of the total number of trees for each forest plot found in EMAN Crown Rating categories since 2010. Bottom right graph is a combined representation of the Crown Ratings for all three forest plots.

Crown rating trends for each individual species were also calculated (see Figure 4.12). Sugar Maple shows a gradual downward trend of healthy individuals, while American Beech has had sharper declines since 2012. Crown ratings for all Ash species were added together to see how the genus is faring as a whole on ***rare*** property. The majority of individuals are dead with White Ash no longer existing in the forest plots in 2015 and all surviving Green/Red Ash (*Fraxinus pennsylvanica*) and Black Ash (*Fraxinus nigra*) in severe decline. For Hop Hornbeam, Yellow Birch (*Betula alleghaniensis*), Black Cherry (*Prunus serotina*) and Red Maple, the majority of individuals in the healthy and light-moderate decline category and populations have all experienced little or no changes since 2010. All other species (Butternut (*Juglans cinerea*), White Pine, White Oak (*Quercus alba*), and Red Oak (*Quercus rubra*)) have too few individuals to determine any crown rating trends. Percent of total dead trees and trees in severe decline for each species can be seen in Figure 4.13.

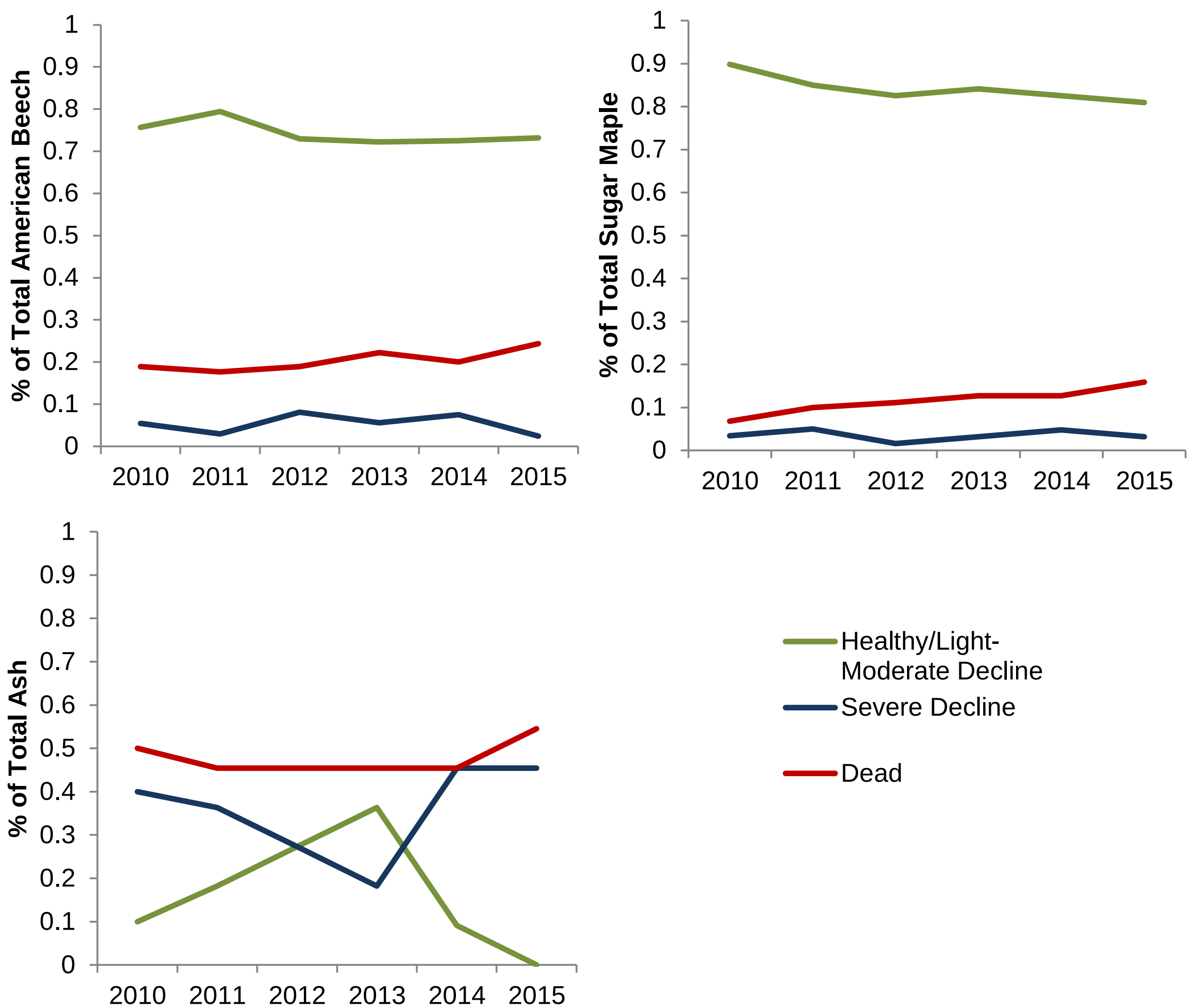


Figure 4.11: Trends in crown ratings for select species from 2010-2015. Crown rating trends for Ash are calculated from all ash species (White Ash, Green/Red Ash, Black Ash, and Ash sp.) found on the property.

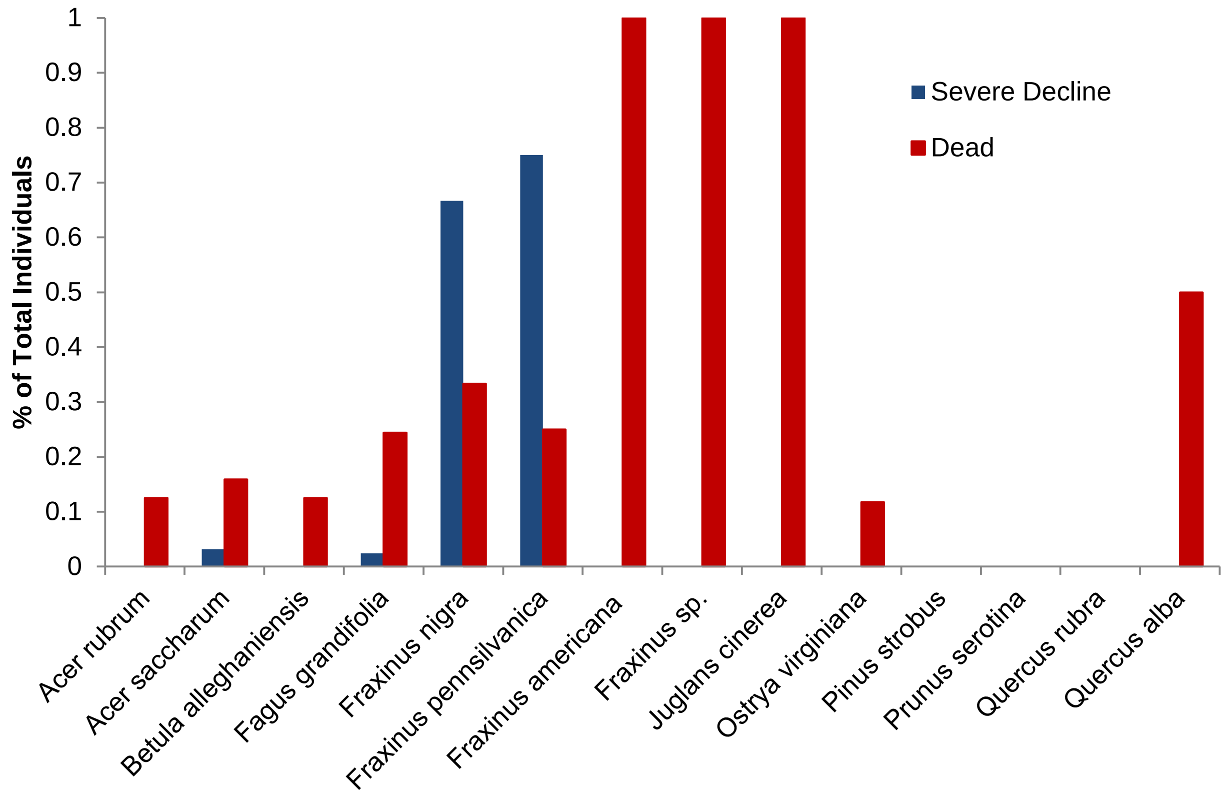


Figure 4.12: Percent of total individuals with severe decline or dead crown ratings for each species in 2015.

4.4 Discussion

4.4.1 Tree Species Diversity

All the forest areas monitored are characterized by two species; American Beech and Sugar Maple. These trees are the most abundant species found in all three forest stands with Sugar Maple being the overall most dominant species. Sugar Maples and American Beech are very commonly seen together as co-dominant species in northeastern North American late-successional forests (Takahashi & Lechowicz 2008). In these Maple-Beech forest stands there doesn't appear to be a clear rule for determining which of the two species will be more dominant (Poulson and Platt 1996; Gravel et al. 2011). There is evidence that American Beech has been recruiting more new canopy trees over the past 40 years than Sugar Maple has in Maple-Beech forests throughout North America (Gravel et al. 2011). This trend is seen weakly in forest plots at ***rare*** with five new American Beech recruits and four new Sugar Maple recruits since 2009. However, there is a high incidence of Beech Bark Disease on the American Beech within the plots making it unlikely Beech will come to dominate the forest stands at ***rare*** in the near future. Beech Bark Disease and its impact on American Beech trees at ***rare*** is discussed in-depth in section 4.3.

The Cliffs and Alvars forest is a mature stand co-dominated by Sugar Maple and American Beech, which together make up 70.4% of the trees in the three monitoring plots**.** Most trees found within this forest stand prefer well drained, upland habitats and are tolerant of shade (Laird Farrar 1995), thus performing well in the complete canopy. The exception is the Yellow Birch which favours moist soils as well as Black Cherry which is intolerant of shade and found only in canopy gaps, but both commonly occur in mixed woods with Sugar Maple and American Beech (Laird Farrar 1995). Given these limitations, it is unsurprising these species are found in low densities in the Cliffs and Alvars. Shannon Diversity Index was the lowest value recorded over the monitoring period and Evenness was the highest. These changes are caused by the death of the last White Ash in the forest plots at Cliffs and Alvars. Three of the four White Ash present in the forest plots were alive at the start of monitoring in 2009, but severe canopy dieback has been recorded on all White Ash since 2010. The most probable cause of death for these trees is Emerald Ash Borer although cause of death is not confirmed. Emerald Ash Borer's impact on Ash trees at ***rare*** is discussed at length in section 4.4.3.3. Despite the loss of the last White Ash, changes to Shannon Diversity over the course of monitoring have been small, as no drastic changes have occurred in the composition of trees within these plots.

Indian Woods is an eastern deciduous remnant old-growth forest dominated by Sugar Maple (67.8% of all trees); such an ecosystem is rare not only to the region but also to southwestern Ontario (OMNR 1999). The Shannon Diversity Index and Evenness values were the lowest in Indian Woods out of the three forest stands at ***rare*** due to poor tree species diversity and the dominance of a single species. Old-growth forests like Indian Woods are often considered to have reached a climax community stage, wherein the community structure will continue in a state of dynamic equilibrium subject to environmental conditions (Krebs 2011). Four trees in Indian Woods were found newly dead in 2015 and appear to have been blown over or killed by other falling trees. Many of the tree deaths in Indian Woods appear to be caused from windthrow during extreme weather events, which has created canopy gaps for new trees to fill.

Canopy dynamics (i.e. gaps and closures) are known to influence the regeneration and growth rates of forests, as well as species composition (Weiskittel and Hix 2003). Forest canopies are in constant flux; openings are created by disturbance and subsequently filled by individuals in the understory. For this reason, the canopy dynamics of a forest continue to be the major influence on forest microhabitat (Jennings et al. 1999). As succession progresses and the canopy closes, the composition of canopy trees shifts toward more shade tolerant species like Sugar Maple and American Beech in eastern deciduous forests (Fox 1977). In 2015, two new American Beech recruits were added to the forest plots and all new recruits since 2009 have either been American Beech or Sugar Maple. These species are able to grow suppressed in the understory and exploit canopy gaps when they occur, outcompeting other shade-intolerant species (Weiskittel and Hix 2003). Given the age of trees in Indian Woods and their shown susceptibility to be damaged by weather events and create canopy gaps, it is possible this forest area will show more noticeable canopy changes on a shorter time scale than the other forested areas.

The Hogsback forest is a forest-wetland complex and as such offers a greater diversity of habitats than the other two forest stands monitored at ***rare*****.** Each of the plots in the Hogsback forest is dominated by a different species: American Beech (Plot 1), Sugar Maple (Plot 2), and Hop Hornbeam (Plot 3). The Hogsback forest also exhibited the highest Shannon Diversity Index and Evenness of the three forest monitoring areas. The wetland areas within the Hogback plots are likely a source of increased diversity, as Yellow Birch, Black Ash, Green/Red Ash, and Red Maple all thrive in wet soils (Sibley 2009). While Sugar Maple and American Beech do dominate the Hogsback, they tend to thrive in areas of better drainage or drier soils (Laird Farrar 1995). This explains why Sugar Maple and American Beech are not able to dominate the Hogsback to the extent they do in the Cliffs and Alvars and Indian Woods. The increased diversity of the Hogsback should allow this forest area to be more resilient to disease, disturbance, and the loss of certain species than the other two forest areas, as there are a greater variety of species available as replacements.

4.4.2 Stand Characteristics and Size Class

The importance value (IV) in forestry is calculated as a means of characterizing the importance of a particular species to the forest community (Roberts-Pichette and Gillespie 1999). The IV is calculated for each species in each plot by incorporating its relative density, relative frequency, and relative dominance. The IV examines each species within forest stand, and takes into consideration how abundant that species is, as well as the total amount of forest area that species occupies within each plot (i.e. basal area). From a forest management perspective, the IV is indicative of the overall influence of a particular species in the community structure and contributes to defining a community based upon its species assemblage.

Despite the differences between Indian Woods, Cliffs and Alvars, and the Hogsback forests, Sugar Maple was found to have the highest IV across all stands. Consistent with the trends in species dominance, the IV of American Beech was second highest in all three forest stands. As mentioned, this combination is commonly associated with late-successional northern hardwood forests, and is typical of the Carolinian forest region (Takahasi & Lechowicz 2008). In the Cliffs and Alvars, Sugar Maple and American Beech are relatively close in IV, however the overall greater abundance and basal area of Sugar Maple gives it a greater value.

The difference between these two species is more striking in Indian Woods where Sugar Maple has an exceedingly high IV in comparison with all other species due to its comparatively high abundance. Although still the second most abundant species in Indian Woods, the number of American Beech trees present is considerably less than Sugar Maples, and they are generally smaller in size. Indian Woods only consisted of one individual of the other two species present, Red Oak and White Oak, which have relatively high IVs due to their large dbh. Prior to 2013, American Beech had the highest IV in the Hogsback forest. In recent years, greater abundance of Sugar Maples have allowed them to surpass American Beech IVs. Red Maple IVs are also of note as they have an importance value closely ranked with that of Sugar Maple and American Beech in the Hogsback. Although they are few in number, the Red Maples present are large and thus exert a strong influence on the forest community as a whole. While generally Red Maple tends to give way to the more shade tolerant Sugar Maple and American Beech in a mature forest, in wet areas that reach a climax status, Red Maple may be able to remain a dominant presence with a relatively high IVs (Walters and Yawney 1990).

The dbh size class distribution of trees can be used to estimate the age of a forest stand, and in conjunction with height and species composition, can help characterize a forest's structure (Burns and Honkala 1990). The size class distribution is useful baseline data for future comparisons examining recruitment and replacement patterns of each stand (Forrester & Runkle 2000; Parker 2003). Both the Cliffs and Alvars and the Hogsback forests had significantly greater abundances of trees in size class one. This indicates that, although the forests stands are late-successional, many younger individuals, particularly shade tolerant species such as Sugar Maple and American Beech, are successfully becoming part of the larger canopy. Size class distribution in both of these forests appears to be right-skewed, typical of a stand comprised mostly of young trees with fewer in the larger size classes. One exception exists in the Cliffs and Alvars, where size class 4 has higher abundance than size class 3 (not significant). Historically, this forest stand was grazed by cattle in the early twentieth century, and this could account for this increased number of trees in size class 4. These trees may have been large enough at the time of the grazing to not be stripped completely by the cattle. Smaller trees, particularly those now in size class 3, were more likely to be grazed, potentially resulting in fewer individuals today.

Indian Woods, a remnant old growth forest, has different distribution than the other two forest stands. Tree abundances are more evenly dispersed among all of the size classes, particularly the first four. No significant differences occurred between these first four size classes, but abundance in size class 1 and 3 were both significantly greater than that of size classes 5 through 8. This indicates that, although regeneration is occurring in this forest stand, it is settling as a climax community forest where Sugar Maple and Beech trees are stable in the understory for many years using a series of gaps to reach the canopy (Forrester & Runkle 2000).

4.4.3 Forest Health

4.4.3.1. Measures of Overall Forest Health

Crown dieback in trees can be used as an early indicator of many of the stresses a tree faces and, as a consequence, as a measure of forest health (Schomaker et al. 2007). Sajan

(2006) outlines thresholds for forest stand health using EMAN crown rating codes. They propose that if the annual mortality rate of dominant and co-dominant trees is less than 5%, a forest stand can be considered healthy (Sajan 2006). Table 4.5 shows the percent change in mortality between each monitoring year for dominant and co-dominant trees. Both the Hogsback and Cliffs and Alvars do not exceed the 5% mortality threshold, but Indian Woods does in three separate monitoring years. Based on this threshold, Cliffs and Alvars and the Hogsback are healthy forest stands with no major problems occurring in their canopies. Being above the 5% threshold in several instances, Indian Woods can be considered an unhealthy forest stand. While this may be true, other factors should be considered before conclusions are made.

Indian Woods has far fewer dominant and co-dominant trees than Cliffs and Alvars and the Hogsback so any tree that dies within Indian Woods has a higher impact within these forest plots. Indian Woods has also experienced more dead dominant and co-dominant trees than the other two forest stands, but the cause of death for these trees appears to be mostly weather related. For instance, in 2015 a dominant tree, likely falling from high winds, knocked over two other co-dominant trees within the plot. Trees in Indian Woods are comparatively larger and older and are thus likely more susceptible to extreme weather events than the other forest stands which have smaller, younger trees. The Hogsback has had only one new tree mortality since 2009 and the cause of death was most likely from the forest pest the Emerald Ash Borer. Cliffs and Alvars has had four deaths since 2009, most of them Ash, and also likely from the Emerald Ash Borer. The difference in how trees are dying between the plots is directly a result of forest stand age and species composition, making direct comparisons of rates of mortality difficult. Indian Woods' trees have died mostly from natural disturbance and the forest is in a state characteristic of old-growth forests with occasional death of the oldest largest trees which are replaced by understory trees (Krebs 2011). Deaths in the other two forests have been mostly caused by an invasive insect, which is an additional stress causing more tree mortalities than would be expected. There are no more ash within the forest plots in Cliffs and Alvars, but several ash still live in the Hogsback. It is likely that these trees will die over the next few years putting the Hogsback on par with Cliffs and Alvars for number of mortalities.

As a consequence of having the most tree deaths, Indian Woods has also had the most new recruits over the monitoring period. New recruits are able to capitalize on canopy gaps left by falling large trees eventually taking their spot in the canopy. The number of new recruits indicates this is an area of active growth, however so far there has been a disproportionate number of deaths to recruits (see table 4.6). In all likelihood an influx of new recruits will be seen over the next years as several large canopy gaps have opened. While the threshold put forth by Sajan (2006) may be a good baseline for examining canopy health, in this case, it seems overly simplistic. Based on the information presented here, it seems as though tree health in Indian Woods is faring better than in Cliffs and Alvars or the Hogsback.

The health of each species was also examined individually using crown ratings as an indicator of health (see Figure 4.13). Unsurprisingly, ash species are faring poorly, likely in large part due to the Emerald Ash Borer epidemic. White Ash trees no longer exist within the forest plots and for Green/Red and Black Ash some trees are dead while the majority are in severe decline. Several species are performing very well in the forest plots including; Red Maple, Hop Hornbeam, Yellow Birch and Black Cherry. These species are not known to have any widespread serious pests of diseases and therefore are of low concern (Davis and Meyer 1997). Similarly, Red Oak, White Oak, and White Pine appear to be healthy and are not known to contract serious diseases or pests (Davis and Meyer 1997), but there are very few individuals of these species within the forest plots making it difficult to make any conclusions about the health of these species for the larger forest stand. Only one Butternut is present in the plots and is no longer alive, the implications of this are discussed in section 4.3.4.

Assessments of Sugar Maple health are of great interest given its high importance value within each forest area. Overall Sugar Maples appear to be healthy with a small percentage (3.2%) in severe decline; however there have been six new Sugar Maple deaths since 2009. Three of those deaths occurred in 2015 being the direct or indirect result of extreme weather. The causes of the other three deaths are unspecified. Sugar Maple is not associated with any serious diseases or pests, and for the time being, populations should be stable and healthy (Bavrlic and Bowers 2009). Given the dominance of Sugar Maples, should a serious threat emerge, the forest areas at ***rare*** could be drastically altered.

Unfortunately the other dominant tree in the forests at ***rare*** does have a serious pest. Populations of American Beech on the property are under threat from Beech Bark Disease. While currently Beech trees are showing little to no decline since the start of monitoring, the recent arrival of Beech Bark Disease in Waterloo Region could change this. Beech Bark Disease is discussed at length in section 4.3.2.

The majority of tree species on ***rare*** property appear to be healthy. Given the information collected from monitoring, no specific management practices are recommended for most species. However, three problem groups have been identified, they are; American Beech, Ash species and Butternut. Each of these three problem groups have a serious pest or disease associated with them that has the potential to cause significant tree mortality. These groups are each given their own separate section to assess the extent of the problem and identify potential management practices.

4.4.3.2 American Beech Pests

A major concern for American Beech at ***rare*** is Beech Bark Disease (BBD). BBD is caused by an infestation of one or more species of a fungus called *Neonectria* (Cale et al. 2015). The fungus typically enters a tree that has been stressed due to feeding from a non-native scale insect called *Cryptococcus fagisuga* (Cale et al. 2015). The fungal infestation causes a whole range of health problems from reduced growth, to crown dieback and potentially death. Mortality from individuals infected by the fungus can be up to 50% (Kasson and Livingston 2011). The non-native scale insect has been known to be present in southern Ontario since 2003 (Morin et al. 2007) and has been documented in the Kitchener area in Steckles Woods (Burtt 2005). Potential instances of BBD have been identified by forest health monitoring on ***rare*** property since 2010. American Beech is an important food tree for many species (Cale et al. 2015) and also constitutes a considerable portion of the over-story in the Cliffs and Alvars and the Hogsback forests. Widespread infection by BBD at ***rare*** could potentially cause significant losses in these forest areas.

Signs of BBD were present in all forest areas at ***rare***. Of the 31 living American Beech within the forest plots, eight (25.8% of trees) were found to have signs of early or late infestation. BBD infestations proceed in three phases; (1) the advancing front where feeding from the scale insect begins, (2) a killing phase, which can last between 3-20 years (typically 3-5) where *Neonectria* infects trees causing death, followed by (3) the 'aftermath' forest in which the fungus is still present, but is no longer actively causing rapid tree mortality (Morin et al. 2007). Three Beech deaths have occurred since 2009 and we are likely in the killing phase of

BBD at ***rare***. Control measures such as selective cutting and removal of severely infected individuals have been recommended within stands to avoid major losses (Davis & Meyer 1997). A less costly measure with greater potential mortality is to allow BBD to progress and gather seed from resistant trees for reseeding (McLaughlin and Greifenhagen 2012).

Note that the *Neonectria* fungi associated with BBD are most visible in the fall when they begin to fruit and are bright red in colour (McLaughlin and Greifenhagen 2012). Due to this, only in the Hogsback were actual fruiting *Neonectria* found on Beech stems as these plots were surveyed latest in the season. This may mean some infected trees are overlooked, especially if monitoring is done within the recommended time frame in late summer. Pictures of fruiting *Neonectria* on Beech stems can be found forest health monitoring photos on the ***rare*** server.

In the 2013 monitoring season, several of the American Beech individuals included in canopy tree monitoring were observed to be covered in Beech Blight Aphids (*Grylloprociphilus imbricator*). Beech Blight Aphids have been speculated to kill or damage small limbs and branches of infested individuals (Childs 2011). While this insect on its own is unlikely to kill or severely harm its host tree, in combination with other stresses (i.e. drought, BBD, etc.) it may result in tree damage or death (Childs 2011). While monitoring in 2014 and 2015 did not note Beech Blight Aphids within the plots, several heavily infested trees were observed just outside of plots in Indian Woods. Since the effects of the Beech Blight Aphid on American Beech are not fully understood or an apparent problem it is recommended no control measures be taken to reduce infestations, but further monitoring should continue.

4.4.3.3 Emerald Ash Borer and the Ash Tree Decline

Emerald Ash Borer (EAB; *Agrilus planepennis*) is a major pest for Ash trees in North America as it is capable of infesting and killing even the healthiest of Ash trees (OMNR 2010). Native to Asia and Eastern Russia, Emerald Ash Borer was first detected in Canada in 2002 just outside of Windsor, Ontario (OMNR 2010). It has since spread over much of southern Ontario and Quebec, making its first appearance in the Waterloo Region in 2010 at Highway 401 and Homer Waterson Boulevard in Kitchener (Region of Waterloo 2010); a location only a few kilometers away from the ***rare*** property.

Evidence of EAB within the forests at ***rare*** has been noted, although no individual adults have actually been observed. Evidence includes small D-shaped exit holes left by adults that emerge from under the bark, as well as dieback of Ash tree crowns. Eleven ash are located within the forest monitoring plots with six being classified as dead and five in severe decline. While they do not make up a large portion of the canopy, the trend appears that the majority of ash will die off in these forested areas changing the canopy composition in these forests (see figure 4.12).

Given that a population of EAB can travel up to 20km over a single year (Prasad et al. 2010), it is likely that populations made their way to ***rare*** around the time of the first observation in Kitchener in 2010. Early detection of EAB is difficult, as the insects first attack the canopy of all but the smallest Ash trees (Cappaert et al. 2005). Because of this, visual surveys rarely detect infestations until the populations have increased and multiple generations have dispersed (Cappaert et al. 2005). Unlike many other insect infestations, EAB will not only attack stressed or damaged trees, but will also attack the healthiest of Ash trees, resulting in death only three to four years after infestation; saplings and small trees may succumb to the insects damage in as little as one year (Herms and McCullough 2014).

Management of EAB infestations is difficult. In many cases, removal of infested/dead tree is required. Where invasion is detected early on, and/or trees are very significant to the landscape, insecticide can be applied. Recent research on the success of this insecticides has had mixed results; some insect populations have continued to increase even after ongoing treatment (Herms et al. 2009), while in others insecticides have proven to have a 100% success rate (Herms 2010). The management options and actions taken should be a site specific strategy (Herms and McCullough 2014). Insecticides however are costly and reapplication is often necessary in subsequent years (Herms et al. 2009). However, insecticides can often still be less costly than tree removal (Herms et al. 2009). Only a few select trees would be able to be saved in this way, but they may provide re-seeding potential once the EAB epidemic has passed. Selective cutting of infested trees is similarly difficult and costly and is probably not a feasible method of managing trees on a property wide scale. At the very least seed should be collected over the next years from any individual ash trees that appear to resist EAB to be used for reintroducing ash back into the forests at ***rare***.

Although the effects of EAB on ***rare's*** forests' diversity are negative, the presence of EAB does present opportunity for research on long term effect and success of various management strategies. In order to limit the further spread of EAB, best management practices include not transporting Ash wood, branches, or logs, including firewood (buy local, burn local), dropping off infested trees at the appropriate drop off sites, and reporting any signs of EAB to the Canadian Food Inspection Agency (Region of Waterloo 2010). A more detailed report and management plan for EAB and Ash will be available on the ***rare*** server in early 2016.

4.4.3.4 Butternut Death

One of twelve Butternut trees of ***rare*** property falls within the monitoring plots in the Cliffs and Alvars area. It had been previously misidentified as dead-standing, but was determined to be living with severe crown dieback and extensive wounds, until it was found dead and broken just below a height of 5m in 2012**.** Butternut is listed as Endangered by both the Federal Species at Risk Act (SARA) and provincially on the Species at Risk in Ontario (SARO). The decline of Butternut in North American is attributed to Butternut canker caused by a fungal pathogen (*Sirococcus clavigignenti-juglandacearum*) that evidence suggests is a relatively recent introduction to North America (Broders & Boland 2010). Symptoms of the disease are elongated, sunken cankers, which commonly originate at leaf scars, buds, or wounds (Davis & Meyer 1997). There is currently no prevention, control, or treatment for the disease and most Butternut conservation efforts are focused on the detection of resistant individuals for seed banking and grafting (Forest Gene Conservation Association 2010, Broders et al. 2015). As no remaining living Butternut are located in any monitoring plots, continued observation of other Butternuts on the property should occur outside of this monitoring program, and continued review of new literature and policy should occur to effective manage this species at risk.

4.4.3.5 Difficulties, Limitations, and Recommended Changes to Forest Canopy Tree Monitoring

The forest canopy tree monitoring program at ***rare*** has a lot of potential to identify changes within the forested areas at ***rare***. Perhaps its greatest strength is to create a yearly picture of change for species actively undergoing serious stresses from invasive pests and diseases, such as American Beech and Ash. However, because it follows EMAN protocol, much of the data collected through forest canopy monitoring is qualitative and highly subjective. As the individual collecting information (i.e. the ecological monitoring intern) each year is never the same, evaluations have sometimes varied substantially between years. Crown class, crown ratings, and stem defects are often not consistent between years as much of this data is determined by the recorder. Due to this, reports from past years have completely ignored or used this qualitative data very sparingly. In this report, an effort was made to include crown class and crown ratings in evaluating forest health, but categories were expanded (i.e. healthy and light/moderate crown decline all considered healthy trees) to reduce potential errors from an individual's judgement. This issue is also identified in a 2009 report by the Credit Valley Conservation Authority in which the authors feel tree crown assessments from previous years are unreliable (Bavrlic and Bowers 2009).

An analysis of stem defect assessments has been excluded from the final report as there is little consistency between years. For instance, a recorder in one year might record a canker while the recorder in the next year sees no canker and only the stump of a tree limb. Aside from a recorder's subjectivity perhaps the most glaring problem with stem defect assessments is they do not assess the severity of a defect. A 5cm open wound on a tree's lower stem is certainly different from one nearly girdling the entirety of the stem. Furthermore, many minor stem defects have little impact on the health of a tree and are commonplace on many species (USDA 2015). The United States Department of Agriculture (USDA) Forest Service contains a more in-depth protocol for assessing stem defects (called tree damages) and sets minimum thresholds for recording on certain types of defects such as open wounds. It is recommended these guidelines or other some other threshold be implemented with the stem defect assessments to ensure recorded defects are indicative of a danger to a tree's health. The USDA Forest Inventory and Analysis (2015) can be found on the ***rare*** server.

Another measure of data not directly incorporated into this report, or most others, is measurements of tree height. For many of the tree heights measured, large changes (up to 23m) have been registered since the start of the monitoring program for many alive and standing trees. These differences make using this data in yearly comparisons challenging. Tree height measurements may be more easily taken after leaves have fallen from the trees. However, due to time constraints, tree heights are taken along with the other measures during monitoring when leaves are still on the trees. Given the difficulty of getting accurate measures with fully leafed out trees, it is recommended that tree height measurements be taken after leaves have fallen from the trees. Many mature trees experience little to no upward growth, such as Sugar Maples (Burns and Honkala 1990). Sugar Maples between 30 and 40 years of age are found to grow at a rate of 30cm per year (Burns and Honkala 1990), and such a small change is unlikely to accurately be captured on a yearly basis with a clinometer. As trees in the forest experience only small changes in height from year to year, it is also recommended tree heights be taken every five years instead of every year. Measurements every five years should still capture changes in forest structure and should allow for time to be allotted after leaves have fallen to measure tree heights.

One measure not included within forest health monitoring at ***rare*** that is present in the EMAN protocol is the inclusion of estimates of stand age (Roberts-Pichette and Gillespie 1999). Tree age can be estimated with the use of a tree corer and gives insight into stand age and growth patterns across time. EMAN protocol recommends coring five trees from each species found in each forest plot, but from trees outside of established plots to avoid damage to trees within plots (Roberts-Pichette and Gillespie 1999). Based on this protocol, a one-time tree coring effort could be made to discover average stand age and historical growth as replicating the coring effort by finding the same trees may prove difficult. Roberts and Gillespie (1999) also recommend taking cores from newly dead trees. This could be compared to average stand age to help determine if the death was directly caused by an external stressor and could easily be added to yearly canopy assessments.

The past six years of forest canopy data at ***rare*** have shown that many measurements change little from year to year. For most of their life, trees in the forest grow and die slowly and because of this yearly forest canopy reports have little new to report. EMAN protocol suggests monitoring trees every five years for mortality and growth (Roberts-Pichette and Gillespie 1999) and this was the suggested interval in the original forest canopy monitoring report in 2009. The protocol at ***rare*** was changed in 2010 to monitor canopy tree data every year to capture the variability in the measurements within the forests at ***rare.*** Yearly measurements of forests are particularly important for tree species in which rapid change is taking place, such as American Beech as BBD typically kills within a 3-5 year period. For this reason, it is recommended that most of the data for the forest canopy monitoring program be gathered each year to capture these rapid changes, but future forest reports are written on a five year interval rather than a yearly interval.

4.5 Conclusions and a Summary of Recommendations

Over the past seven years of monitoring the forests at ***rare***, there have been few changes in the forest stands in terms of diversity, size class, dominance, and canopy composition. The most appreciable difference over the monitoring period has been the decline of Ash trees as a result of Emerald Ash Borer. American Beech are also showing signs of decline and developing management and/or monitoring programs targeting species of special concern is of the utmost importance. The three following types of trees should be targeted;

1. **Ash species**: Create a recovery program for ash on the property by collecting seed from ash trees, especially those that appear unaffected by EAB. For further information, an in-depth survey and management plan for EAB is currently in draft and will be available on the ***rare*** server.

2. **American Beech**: Create a targeted survey to examine the extent of BBD on the property, which should take place in the fall. At the very least, examine Beech trees within the plots for signs of BBD. Collect seed for future Beech recovery program prioritizing trees that appear unaffected by BBD. Consider options of selective cutting and removal of heavily infested trees.

3. **Butternut**: Monitor condition of other 11 Butternut located outside of the forest plots. Continue partnership with GRIPP for more seed collection from living individuals, especially those with no signs of Butternut canker.

Long term data collection and analysis is required in order to fully understand if the integrity of the three forest stands is in fact being maintained or improved through management strategies, which have come as a result of forest health monitoring. With constant changes in the surrounding land use, continued monitoring will be important so that any changes in the health of the forests can be detected early on. However, there are some changes that need to be made to Forest Health Monitoring should the program and data be viable into the future. The following modifications to data collection and forest canopy monitoring report are recommended;

1. Write the forest monitoring report on a 5-year interval instead of every year, but continue to collect yearly data

2. Measure the tree height after leaves have fallen and only every five years.

3. Modify the current stem defect system to incorporate minimum damage thresholds and take more detailed notes and/or pictures about the extent of observed damage.

4. Incorporate tree-aging into the monitoring program with a one-time core assessment from trees outside of plots to calculate average stand age and by taking cores from all newly dead trees within plots.

These modifications are not drastic changes that will substantially alter the data collected for forest monitoring, but will allow more useful data to be collected and summarized on a more efficient timescale.

4.6 Literature Cited

Allen, C.D., Macalady, A.K., Chenchouni, H., Bachelet, D., McDowell, N., et al.. 2010. A global overview of drought and heat-induced tree mortality reveals emerging climate change risks for forest. Forest Ecology and Management. *Elsevier*, 259(4): 660-684.

Bavrlic, K., and Bowers, K. 2009. Monitoring forest integrity within the Credit River Watershed, Chapter 3: Forest Tree Health 2005-2009. Credit Valley Conservation.

Broders, K.D. and Boland, G.J. 2010. Molecular diagnostic assay for detection of the Butternut canker pathogen Sirococcus clavigignenti-juglandacearum. *The American Phytopathological Society*, 94(8):952-958.

Broders, K.D., Boraks, A., Barbison, L., Brown, J. and Boland G.J. 2015. Recent insights into the pandemic disease butternut canker caused by invasive pathogen *Ophiognomonia clavigignenti-juglandacearum. For. Path.* 45:1-8.

Butt, S. 2011. South Central Ontario Forest Biodiversity: Monitoring Plots Analysis. Association for Canadian Educational Resources. Available at: http://acer-acre.ca/wp-content/uploads/2011/12/SCOFBP.pdf Accessed: May 31, 2012.

Burns, R.M., and Honkala, B.H. 1990. Silvics of North America: 1. Conifers; 2. Hardwoods. Agriculture Handbook 654(2):877. US Department of Agriculture, Forest Service, Washington, DC.

Cale, J.A., Teale, S.A., Johnston, M.T., Boyer, G.L., Perri, K.A., and Castello, J.D. 2015. New ecological and physiological dimensions of beech bark disease development in aftermath forests. *Forest Ecology and Management,* 336:99-108.

Cappaert, D., McCullough, D.G., Poland, T.M., and Siegert, N.W. 2005. Emerald ash borer in North America: A research and regulatory challenge. *American Entomology*, 51:152-163.

Childs, R. 2011. Beech Blight Aphid. Landscape, Nursery & Urban Forestry Program, University of Massachusetts. Available at: extension.umass.edu/landscape/fact-sheets/beech-blight-aphid Accessed: November 29, 2013.

Craig, S., Damstra, E., Kelly, P., Leather, P., Marko Dunn, J., McDonald, L., McLeod, K., Moore, R., Pope C., Quinn, Reinert, J., Robson, L., Sobek-Swant, S., Whelan, S., Woodcock, T. 2014 . Draft ***rare*** Environmental Management Plan. Available on ***rare*** server.

Davis, C.N., and Meyer, T.R. 1997. Field guide to tree diseases of Ontario. Natural Resources Canada, Canadian Forest Service, Great Lakes Forestry Centre, Sault St. Marie, Canada.

Environment Canada and Canadian Forest Service. 2004. EMAN ecosystem monitoring partnership: Tree health. Ecological Monitoring and Assessment Network. Available at: http://www.ec.gc.ca/faunescience-wildlifescience/default.asp?lang =En&n=9C4F74C5-1 Accessed: May 31, 2012.

Forrester, J.A., and Runkle, J.R. 2000. Mortality and replacement patterns of an old-growth Acer-Fagus woods in the Holden Arboretum, Northeastern Ohio. *American Naturalist*, 144(2):227-242.

Forest Gene Conservation Association. 2010. Butternut Tree- A Landowner's Resource Guide. Available at:http://www.fgca.net/conservation/sar/butternut.aspx Accessed: November 30, 2012.

Fox, J.F. 1977. Alternation and coexistence of tree species. *American Naturalist*, 111(977):69-89.

Führer, E. 2000. Forest function, ecosystem stability and management*. Forest Ecology and Management*, 132:29-38

Gardner, T. 2010. Monitoring forest biodiversity: improving conservation through ecologically-responsible management. Earthscan Ltd., London, UK.

Gravel, D., Beaudet, M., and Messier, C. 2011. Sapling age structure and growth series a shift in recruitment dynamics of sugar maple and American beech over the last 40 years. C*anadian Journal of Forest Research,* 41: 873–880.

Herms, D.A. 2010. Multiyear evaluation of systemic insecticides for control of Emerald Ash Borer. Proceedings: Emerald Ash Borer Research and Technology Development Meeting of 2009. pp 71-75.

Herms, D.A. and McCullough, D.G. 2014. Emerald ash borer Invasion of North America: History, Biology, Ecology, Impacts, and Management. *Annual Review of Entomology*, 59:13-30.

Herms, D.A., McCullough, D.G., Smitley, D.R., Sadof, C.F., Williamson, R.C., Dixon, P.L. 2009. Insecticide options for protecting Ash trees from Emerald Ash Borer. Urbana, IL. National IPM Centre. pp 12.

Jennings, S.B., Brown, N.D., and Sheil, D. 1999. Assessing forest canopies and understory illumination: canopy closure, canopy cover, and other measures. *Forestry,* 72(1):59-74.

Kasson, M.T., and Livingston W.H. 2011. Relationships among beech bark disease, climate, radial growth response and mortality of American beech in northern Maine. *Forest Pathology*, 10.1111/j.1439-0329.2011.00742.x

Krebs, C.J. 2011. Ecology: The experimental analysis of distribution and abundance (5th edition). Benjamin Cummings, San Francisco, USA.

Laird Farrar, J. 1995. Trees in Canada. Fitzhenry and Whiteside Ltd., Markham, Ontario, Canada.

Leech, N.L., Barrett, K.C., and Morgan, G.A. 2008. SPSS for intermediate statistics use and interpretation (3rd edition). Lawrence Erlbaum Associates (Taylor Francis Group), New York, NY, USA.

McCarter, J. 2009. Ecological monitoring at rare Charitable Research Reserve. Available on rare server.

McLaughlin, J., and Greifenhagen S. 2012. Beech bark disease in Ontario: A primer and management recommendations. *Ontario Forest Research Institute*, note No.71.

Morin, R.S., Liebhold, A.M., Tobin, P.C., Gottschalk, K.W., and Luzader, E. 2007. Spread of beech bark disease in the eastern United States and its relationship to regional forest composition. Can. J. For. Res. 37:726-736.

Moser, K.W., Barnard, E.L., Billings, R.F., Crocker, S.J., Dix, M.E., Gray, A.N., Ice, G.G., Kim, M., Reid, R., Rodman, S.U., and McWilliams W.H. 2009. Impacts of non-native invasive species on US forests and recommendations for policy and management. *Journal of Forestry*, 320-327.

Ontario Ministry of Natural Resources. 1999. Extension Notes: the old-growth forests of southern Ontario. LandOwner Resource Centre. Available at: http://www.lrconline.com/Extension_Notes_English/pdf/oldgrwth.pdf. Accessed: February 1, 2011.

Ontario Ministry of Natural Resources. (2010). Forest Health Alert: Emerald Ash Borer. Available at:http://www.mnr.gov.on.ca/en/Business/Forests/2ColumnSubPage/STEL02_166994.html . Accessed: November 29 2013

Parker, B.H. 2003. Monitoring program status report. Long Point World Biosphere Reserve. Available at http://longpointbiosphere.com/library/statusrt.pdf Accessed on: May 31, 2012.

Poulson, T.L., and Platt, W.J. 1996. Replacement patterns of Beech and Sugar Maple in Warren Woods, Michigan. *Ecology,* 77(4):1234-1253.

Parker, B.H. 2003. Monitoring program status report. Long Point World Biosphere Reserve. Available at http://longpointbiosphere.com/library/statusrt.pdf Accessed on: May 31, 2012.

Prasad, A.A., Iverson, L.R., Peters, M.P., Bossenbroek, J.M., and Matthews, S.N. 2010. Modeling the invasive emerald ash borer risk of spread using a spatially explicit cellular model. Landscape Ecology, 25:353-369.

Roberts-Pichette, P., and Gillespie, L. 1999. Terrestrial vegetation biodiversity monitoring protocols. Ecological Monitoring and Assessment Network. Available at: http://www.ec.gc.ca/faunescience-wildlifescience/default.asp?lang=En&n =9C4F74C5-1 Accessed: May 31, 2012.

Sajan, R. 2006. Tree health-data analysis. Draft unpublished manuscript. Canadian Forest Service, Sault Ste. Marie, Ontario.

Schomaker, M.E., Zarnoch, S.J., Bechtold, W.A., Latelle, D.J., Burkman, W.G., and Cox, S.M. 2007. Crown-condition classification: A guide to data collection and analysis. USDA Forest Service.

Sibley, D.A. 2009. The Sibley Guide to Trees. Alfred A. Knopf, Inc., New York, USA.

Takahashi, K., and Lechowicz, M.J. 2008. Do interspecific differences in sapling growth traits contribute to the co-dominance of *Acer saccharum* and *Fagus grandifolia*? *Annals of Botany*, 101(1):103-109.

USDA Forest Service. 2015. Forest inventory and analysis national core field guide, Volume I: Field data collection procedures for phase 2 plots. Version 7.0.

Waldron, G. 2003. Trees of the Carolinian Forest: A guide to species, their ecology, and uses. The Boston Mills Press. Erin, Canada.

Walters, R.S., and Yawney, H.W. 1990. Red Maple. In: Burns, R.M., and Honkala, B.H. 1990. Silvics of North America: 1. Conifers; 2. Hardwoods. Agriculture Handbook 654(2):877. US Department of Agriculture, Forest Service, Washington, DC.

Weed, A.S., Ayres, M.P., and Hicke, J.A. 2013. Consequences of climate change for biotic disturbances in North American forests. *Ecological monographs*, 83(4):441-470.

Weiskittel, A.R., and Hix, D.M. 2003. Canopy gap characteristics of an Oak-Beech-Maple old-growth forest in northeastern Ohio. *Ohio Journal of Science*, 103(4):111-115.

5.0 Soil Humus Decay Rate Monitoring

Prepared by: Tim Skuse

5.1 Introduction

5.1.1 Soil Characteristics and Functions

Decomposition is defined as the physical, chemical, and biological breakdown of organic material into simpler matter, and it is a significant global producer of carbon dioxide, as well as methane and nitrogen gases (Berg and McClaugherty 2008). Soil humus, the stable organic material remaining after initial decomposition, acts as the reservoir for the carbon that was not released during decay, as well as storage for the nutrients that support plant growth and the microbial and fungal communities of the soil (Berg and McClaugherty 2008). The rate at which decomposition occurs is dependent on many factors, including the composition of the material being decomposed, the ecology (species composition and abundance) of the decomposer organisms available in the soil, and a suite of environmental variables, including soil temperature, moisture, pH and aeration (Parks Canada 2006, Singh and Gupta 1977).

*5.1.2 Soil Humus Decay Rate Monitoring at **rare***

In response to concerns that climate change may affect forest carbon budgets, Natural Resources Canada developed the Canadian Intersite Decomposition Experiment (NRC 2007) to examine the long-term litter decomposition rates and nutrient mineralization of forests across Canada. In Canadian forests, large amounts of carbon are stored in trees, soils, and decaying plant litter and any change in the balance between the uptake of carbon through photosynthesis and the release of carbon through decay and other activities could have an impact on levels of atmospheric carbon dioxide, an important greenhouse gas linked to global climate change (Bardgett et al. 2013). Thus, warmer temperatures could increase decay rates, which in turn would release carbon stored in the soils and litter and potentially accelerate rises in atmospheric carbon dioxide (Bardgett et al. 2013).

The moderate temperature zone of southwestern Ontario was excluded from the NRC long-term decomposition study. As long-term monitoring of soil decay rates can provide valuable information on the relationship between soil decomposition and environmental factors, it may serve to inform forest management decisions at ***rare***. For example, the effects that nearby aggregate mining or pesticide application may have on the health of our forest soils are unknown. Decay rate monitoring, together with the other biological monitoring protocols in place at ***rare*** such as forest tree biodiversity and plethodontid salamander monitoring, can provide us with a greater understanding of the integrity and stability of our forest ecosystems.

The first EMAN soil humus decay rate monitoring plots at ***rare*** were established on November 9, 2009 at the Cliffs and Alvars forest canopy tree biodiversity plot one. The success of the first monitoring year resulted in an expansion of the study in 2010 by the establishment of monitoring plots in both the Indian Woods and the Hogsback forest stands, within the first tree plot at each location.

At ***rare***, the objective of this monitoring procedure is to contribute to the assessment of forest ecosystem functioning by monitoring yearly mass loss in standardized decay sticks as a representation of soil decomposition rates. As per the EMAN soil humus decay rate monitoring protocol (Parks Canada 2006), Annual Decay Rate (ADR) plots were located at the corners of the permanent forest canopy tree biodiversity plots in each forest stand. The information gained from decay monitoring can then be directly linked to the forest health and productivity data. Decay rates compared over years are expected to remain relatively stable and sticks positioned on the surface of the soil are expected to experience less mean weight loss than those placed below the surface, where they are more accessible to soil microorganisms responsible for decomposition. A change in decay rates would reflect a change in the physical or biological soil environments.

5.2 Methods

5.2.1 Monitoring Protocol: Decay Stick Installation

Decay sticks were prepared in-house prior to ground installation. To prepare the tongue depressors (MedPro, 100% natural birch wood, ultra-smooth finish) a 2mm hole was drilled at one end of each stick to allow for the attachment of identification tags. While only 144 decay sticks are used during monitoring, it is best to prepare approximately fifteen sticks in excess in case of damage prior to or during installation. Once drilled, decay sticks were oven-dried at 70°C for 48 hours (Quincy 0Gc-181512 Gravity Convection Oven). Following this, decay sticks were left for at least 24 hours at room temperature and then weighed (to 0.001g) on a Sartorius 1265MP balance. A sample datasheet to record stick weight pre and post decay can be found in Appendix C.6. After recording their mass, decay sticks were tagged with pre-labelled aluminum tags attached with approximately 30cm of extra-strong (40LB) fishing line. With the exception of the initial year of monitoring, decay sticks were placed in 100% vinyl mesh bags (dimension: 17cm x 4cm with an approximate pocket size of 16cm x 3cm; hole size: 3mmx 2mm). Vinyl mesh bags were prepared in advance of decay stick placement, with an excess created in case of damage during installation. These bags were an amendment to the monitoring protocol added in 2010 in an attempt to keep all the decay stick's pieces together and increase the number of decay sticks excavated intact. Mesh bags are often used in studies of leaf litter decay rate (Moore et al. 2005; Albers et al. 2004; Gallardo et al. 1995). A complete list of equipment required for installation can be found in List B.4.

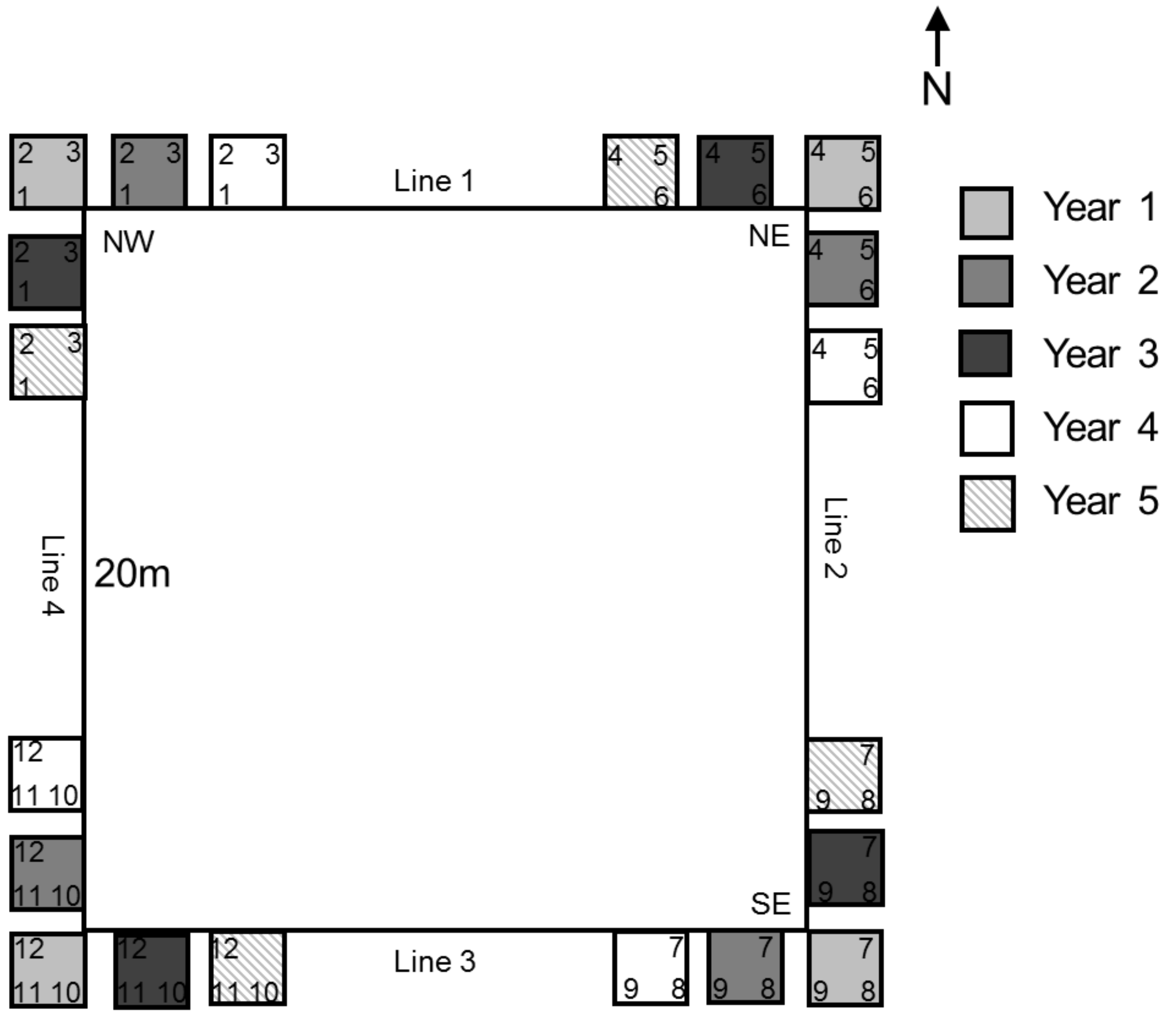


Figure 5.1: Distribution of annual soil humus decay rate (ADR) plots (numbered 1-12) around a forest canopy tree biodiversity plot. Twelve ADR plots are arranged around the corners of each plot; three located in the originally recommended location of the corner and moved counter-clockwise and clock-wise in alternating years from the original location to avoid previously sampled soil areas. Plots are colour coded by monitoring year and rotate back to 1 after 5 consecutive years.

A 1m^2 quadrat was marked on each corner of the forest plots and three ADR plots were positioned within each quadrat on the corners radiating out from the corner of the forest plot (Figure 5.1). At each ADR plot, a 30cm x 30cm hole was excavated with the soil plug removed intact if possible and placed to the side. Using a knife or chisel, three slots were made parallel to the forest floor on the north wall of the excavated hole. The slots were of large enough size to accommodate the bagged decay sticks snuggly. Slots were measured 5cm below the soil surface and were re-measured upon completion with the accurate depth below the surface recorded. The three slots were measured to be approximately 10cm apart. The bagged decay sticks were inserted into the slots, with the pre-labelled aluminum tags previously attached via fishing line left on the soil surface. A pigtail stake marked with flagging tape labelled with the forest stand and ADR plot number (i.e. CA-ADR-2) was inserted into the centre of the excavated hole. Fishing line was used to attach each bagged decay stick to one another and the centre pigtail stake with enough excess that they would not be shifted. This fishing line is to be used as a guide to locate the sticks upon excavation and therefore should not be so taut as to affect their movement throughout the year. A fourth bagged decay stick was attached to the centre pigtail stake via fishing line and left on the soil surface (Figure 5.2). The excavated hole was then refilled with the displaced soil and soil plug, and the exposed tags were covered with leaf litter to prevent public or wildlife tampering. In 2015, decay sticks were installed on October 30th in the Cliffs and Alvars, November 4th in the Indian Woods, and November 6th in the Hogsback.

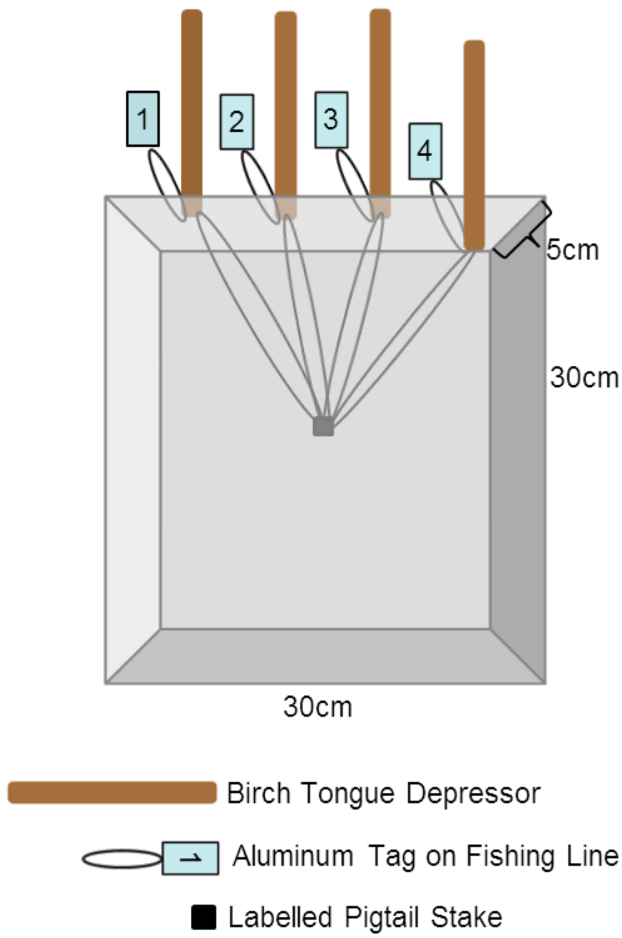


Figure 5.2: Diagram of annual soil humus decay rate (ADR) monitoring plot set-up as viewed from above. Decay sticks 1-3 are installed parallel to the soil surface at a depth of 5cm, separated 10cm from each other. Stick 4 is placed on the soil surface, and all decay sticks are tied to the central pigtail stake. Figure from Robson (2010).

5.2.2 Monitoring Protocol: Decay Stick Excavation

Decay sticks were excavated one year following their installation. In the event of an early frost and ground freeze, the date of excavation should be moved forward. Using a trowel, soil surrounding the pigtail stake in each ADR, where decay sticks were suspected to be, was slowly removed. As tags and fishing line were uncovered, they were used to help locate the decay sticks and to gently pull the bagged decay sticks from the ground once a hole has been dug. Each decay stick and its associated tag were placed in an individual re-sealable plastic bag or envelope. A complete list of equipment required for excavation can be found in List B.4.

Decay sticks were each removed from their vinyl bags and any dirt that adhered to the stick was removed. When possible, vinyl bags were cleaned and kept for reuse. Each stick was gently brushed with a dry paintbrush and then gently scrubbed with a second paintbrush in water. Decay sticks were placed in individual paper envelopes following cleaning, and each envelope was labelled with the site and tag number. Decay sticks, inside their envelopes, were then oven-dried at 70°C for 48 hours and subsequently let to sit for 24 hours at room temperature before being weighed (to 0.001g). Weights were recorded on a datasheet available on the ***rare*** server and in Figure C.5.

5.2.3 Data Analysis

Data were analysed using Microsoft Excel 14.0.6 (Microsoft 2010) and SPSS Statistics Version 20. Prior to analysis, parametric assumptions were examined. When transformation was required, the appropriate transformation to decouple variance and mean was determined using Taylor's Power Law (Perry 1981). Otherwise, the best transformation was applied and the most robust tests were used, followed by cautious interpretation of the results.

Percent dry weight loss for each decay stick was calculated, as changes in dry weight can be examined as a proxy for soil decomposition (NRC 2007). Weight loss was compared across years and sites using a univariate analysis of variance (ANOVA) followed by Bonferroni post hoc testing to determine where differences occurred. Decay rates were also examined in relation to measured weather variables (temperature and precipitation) to determine how much of the variation in rates is explained by the weather or if other factors are influencing decay.

5.3 Results

In 2015, a 142 of a possible 144 sticks were recovered from annual decay rate plots. Two decay sticks were lost during the sampling year from the Cliffs and Alvars forest stand, one stick lost was located on the surface and one was located below the surface. Decay sticks positioned below ground were found to have lost significantly more mass than those positioned on the soil surface ($F_{1,\,753}$ =376.474 p<0.001) (see Table 5.1). Across years decay rates differed significantly, in particular, 2011 and 2015 were both significantly lower than 2012, 2013, and 2014 ($F_{5,755}$=11.198 p<0.001). Decay rates in the Hogsback were also found to be significantly lower than both Indian Woods and Cliff and Alvars ($F_{2,752}$=12.864 p<0.001).

Average monthly temperature and average monthly rainfall were compared to the decay rates seen. Temperature showed no significant correlation with decay rates and the relationship with rainfall was significant, but had a weak relationship (p<0.001, r=0.130).

Table 5.1: Annual decay rates measured as percent mass loss of decay sticks from Cliffs and Alvars, Indian Woods, and the Hogsback forest stands for all monitoring years (a.) and for 2015 (b.). Decay sticks below and above ground had significantly different mass losses, regardless of site or year. SD= Standard Deviation.

a.	**Cliffs and Alvars**		**Indian Woods**		**Hogsback**	
	Mean (%)	SD	Mean (%)	SD	Mean (%)	SD
All Sticks	35.4	19.0	39.0	20.6	30.0	17.9
Sticks Below Ground	42.9	15.6	45.0	18.2	33.5	17.0
Sticks Above Ground	11.5	10.9	20.1	15.0	18.5	17.0

b.	**Cliffs and Alvars**		**Indian Woods**		**Hogsback**	
	Mean (%)	SD	Mean (%)	SD	Mean (%)	SD
All Sticks	29.9	15.6	36.4	18.6	26.3	17.6
Sticks Below Ground	35.1	14.5	43.7	14.4	29.7	14.8
Sticks Above Ground	12.9	10.8	14.4	10.5	15.9	12.1

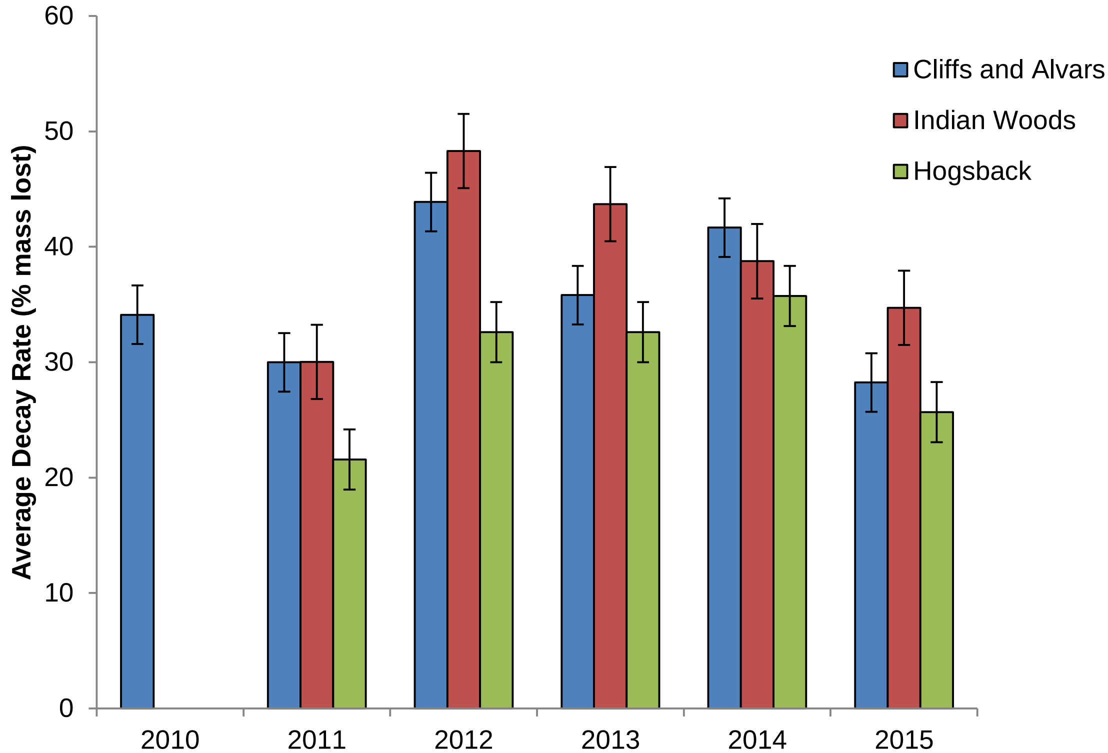


Figure 5.3: Average decay rate comparison over monitoring years for each site. Only Cliffs and Alvars was monitored in 2010. Error bars represent +/- one standard error.

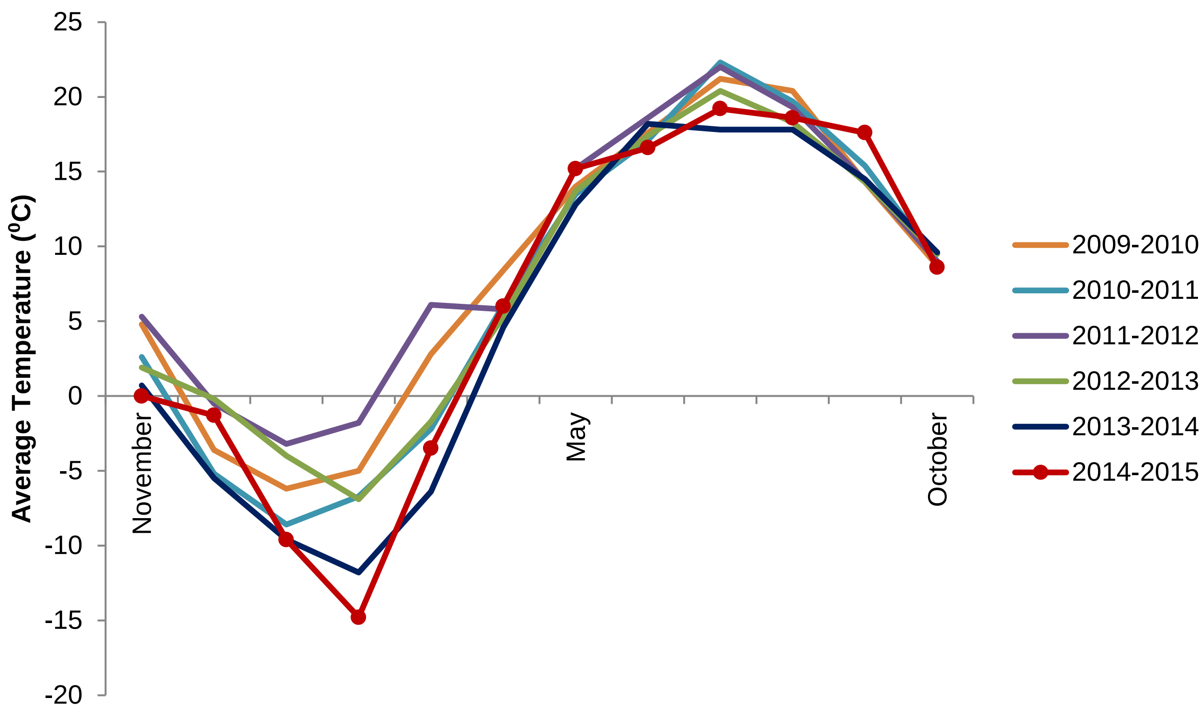


Figure 5.4: Temperature data for Waterloo Region by month during soil humus decay monitoring years, where average temperature is the average monthly temperature (Environment Canada- data from Kitchener-Waterloo Weather Station).

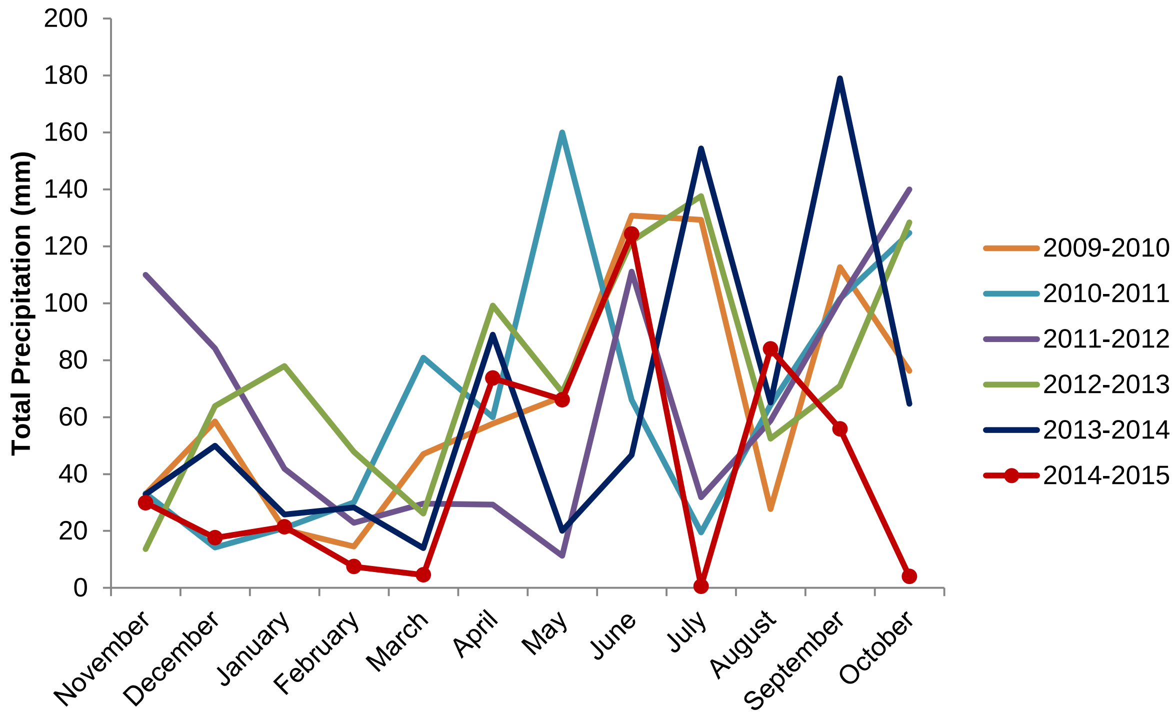


Figure 5.5: Precipitation data for Waterloo Region by month during soil humus decay monitoring years, where the total precipitation from each month is displayed (Environment Canada- data from Kitchener-Waterloo Weather Station).

5.4 Discussion

Rates of decay can be influenced by a variety of factors including climate, temperature, substrate type, nutrient concentrations and availability, litter type and size, and soil organisms (Parks Canada 2006). Weight loss associated with decomposition is strongly dependent on aerobic microbial activity (Bunnell et al.1977). Decay sticks that were placed below ground were more accessible to soil microorganisms, fungi, and moisture, which could explain the higher decay rate observed below ground (Table 5.1).

Moisture and temperature, which vary greatly with local conditions, are the principle factors that affect rate of decay (Singh and Gupta 1977), as they strongly influence microbial activity (Bunnell et al. 1977). Higher temperatures cause higher microbial activity and, as a result, generally cause increases in decay rates (Olson 1963; Van Cleave 1971; Singh and Gupta 1977). The same trend is seen in soil moisture, as increased soil moisture permits increased microbial and plant activity leading to higher rates of decay (Xu et al. 2004). Soil decay rates appear to be most strongly impacted by changes in whichever factor, temperature or moisture, is most limiting. Lower moisture contents result in a limited response to temperature changes and lower temperatures result in a limited response to changes in moisture level (Schlentner and Van Cleve 1984).

2011 and 2015 both had significantly lower decay rates than other full monitoring years (2012-2014). 2015 had the second lowest average monthly temperature (6.05^{o}C) and the second lowest overall decay rates. For 2011, average monthly temperatures were the third lowest (6.9^{o}C), but overall decay rates were the lowest. Interestingly, the coldest year on record, 2014 (average monthly temp. 5.2^{o}C), had the second highest overall rates of decay. These trends suggest precipitation may also have been playing a role in driving comparatively high rates of decay in 2014.

2015 had the lowest average monthly precipitation (48.1mm), but 2014 and 2011 had nearly identical average monthly precipitation (64.1mm and 64.3mm, respectively). According to this data, 2014 was both colder and less moist than 2011, yet decay rates were significantly higher. This disparity may be explained by the number of rainy days in each year. A high percentage of rainy days are known to increase rates of decomposition as the distribution of rain and, as a consequence, soil moisture will be higher more often (Singh and Gupta 1977). Based on climate data from the Kitchener-Waterloo Weather Station (Environment Canada), it rained more than 0.5mm on 26% of days in 2011 and on 29% of days in 2014.This small difference in the number of days may have caused the significant difference in the decay rates seen between 2014 and 2011, although it does not explain why 2011 had lower decay rates than 2015. In 2015 it rained at least 0.5mm on 23% of days, and there was lower average temperature and rainfall than 2011. These factors suggest 2015 should have had lower decay rates than 2011.

Another possibility is that the number and length of winter frosts are impacting decay rates (See table 5.2). The longest period of time maximum daily temperature did not rise above 0^{o}C was highest in 2015 followed by 2011. The number of freeze and thaw periods may also be of interest as frequent freezing and thawing is known to increase rates of decay (Taylor and Parkinson 1988). There were only three freeze and thaw periods in 2015 and there were nine in 2011. Given this data, it is still unclear why rates of decay in 2011 are lower than in 2015. It must be noted the difference between the decay rates in 2015 and 2011 is small (2.5%). This difference seen may be from variability inherent in differences in weather at each site and from rotating plot locations. The variability may also be difficult to explain as there is no site specific soil moisture and soil temperature data, but rather total precipitation and air temperature from a nearby weather station used as a proxy. This issue is also seen in correlating weather variables to decay rates in statistical analysis.

When relating average yearly precipitation and temperatures to the rates of decay seen, temperature showed no significant response and precipitation was only weakly related. As discussed, these weather factors have been shown to have a positive relationship with decay rates in other studies, making the results from statistical analysis here seem out of place. Given that soil monitoring at ***rare*** has only five years of consistent data and climate data was collected from one weather station and applied to all forest stands, sample sizes are very small for statistical analysis. Due to the small sample sizes and lack of site specific temperature and moisture measurements, statistical analysis will be unlikely to find accurate relationships between these variables. To improve this analysis for future monitoring years, site specific variables, such as soil moisture and temperature, could be measured with the use of data logging equipment. From this data we would better be able to correlate decay rates with climate data to determine if climate or other underlying factors are driving decomposition.

Table 5.2: Highest number of consecutive days below 0^oC, total number of days above 0^oC and number of freeze and thaw periods for winter months (approximately December 20th-March 20th) during each monitoring year. Data was collected based on maximum daily temperatures from Environment Canada-data from Kitchener-Waterloo Weather Station.

	2015	2014	2013	2012	2011
Highest consecutive # of days below 0^oC	55	16	11	5	24
Total Days >0^oC	19	27	39	65	29
# of Freeze and Thaw Periods	3	8	15	14	9

The Hogsback forest significantly differed from both other stands with lower decay rates. The Hogsback is a forest-wetland complex that has a mixture of upland and lowland areas with swampy features. In particular, one corner of the monitoring plot located here is found within a swamp. Sticks in this corner have had consistently lower rates of decay (average 23% mass lost) than the other three corners in this plot (average 32% mass lost). This difference is likely from anaerobic conditions. If decay sticks are continuously exposed to extremely high moisture levels or are completely submerged in water, decay rates can be slowed by lack of oxygen to support microbial activity (USDA 2007, Schlentner and Van Cleve 1984). Because 25% of the sticks in the Hogsback will be found in these anaerobic conditions, decay rates in the Hogsback are expected to remain lower than the other forest stands in future years.

Variation in decay rates at ***rare*** appear to correspond closely to similar studies. A soil decay study in Nova Scotia with a similar EMAN-based methodology recorded an 11% range in decay rates over five years (Ure et al. 2010). The range of decay rates seen at ***rare*** were 10% in the Hogsback, 12% in Indian Woods and 15% in Cliffs and Alvars. Variation in decay rates from forests in Nova Scotia are difficult to compare to variation in decay rates from forests in southern Ontario as the climate, soil, and forest composition are different. However, there is a lack of studies in southern Ontario and throughout Canada that have assessed this type of data, especially over long periods of time. Still, this comparison shows that the fluctuations in decay rates in another study are not wildly different than those seen at ***rare*** and lends credibility to the rates seen in the study here.

2015 represents the fifth full and consecutive year for monitoring soil decay rates. From the data collected thus far we can calculate thresholds for decay rates in each forest stand. Thresholds for each forest stand and for stick positions can be found in table 5.1. Future monitoring years should compare measured decay rates to those seen in thresholds to determine if and where significant changes are occurring.

5.5 Conclusions and Recommendations

Changing decay rates can be indicative of global climate change or local development and agricultural pressures. Only continued monitoring can investigate these potential trends. Past reports on soil monitoring have indicated the program should run for a minimum of five years to establish baseline decay thresholds in which to distinguish between weather changes and actual changes in decay rates. While baseline thresholds are important to establish, monitoring soil decay rates into the foreseeable future is advised. Soil processes and carbon turnover does not occur quickly. Turnover rates are highly variable based on location, but tend to occur on scales of upwards of 10 years (Schlesinger and Andrews 2000, Trumbore et al. 1996). As these processes occur over long timescales, it is pertinent to continue monitoring to be able to separate changes in climate from changes caused by other external pressures.

Furthermore, improvements can be made to the soil monitoring program by monitoring soil moisture and/or other climate variables in plots each month to allow for a closer comparison of these variables to decay rates. This would require additional and potentially expensive field equipment.

5.6 Literature Cited

Albers, D., Migge, S., Schaefer, M., and Scheu, S. 2004. Decomposition of beech leaves (*Fagus sylvatica*) and spruce needles (*Picea abies*) in pure mixed stands of beech and spruce. *Soil Biology and Biochemistry*, 36(1):155-164.

Bardgett, R.D., Manning, P., Morrien, E., and De Vries, F.T. 2013. Hierarchical responses of plant-soil interactions to climate change: Consequences for the global carbon cycle. *Journal of Ecology*, 101:334-343.

Berg, B. and McClaugherty, C. 2008. Plant litter: Decomposition, humus formation, carbon sequestration (2nd edition). Springer-Verlag, Berlin, Germany.

Bunnell, F.L., Tait, D.E.N., Flanagan, P.W., and Van Cleve, K. 1977. Microbial respiration and substrate weight loss-I: A general model of the influences of abiotic variables. *Soil Biology & Biochemistry*, 9:33-40.

Environment Canada. 2013. National Climate Data and Information Archive. Available at: http://www.climate.weatheroffice.gc.ca/climate Data/canada_e.html. Accessed: April 1, 2013.

Gallardo, J.F., Santa-Regina, I., Harrison, A.F., and Howard, D.M. 1995. Organic matter and nutrient dynamics in three ecosystems of the “Sierra de Bejar” Mountains (Salamanca Province, Spain). *Acta Oecologica-International Journal of Ecology,* 16(4):447-459.

Moore, T.R., Trofymow, J.A., Siltanen, M., Prescott, C., and CIDET Working Group. 2005. Patterns of decomposition and carbon, nitrogen, and phosphorus dynamics of litter in upland forest and peatland sites in central Canada. *Canadian Journal of Forest Research*, 35:133-142.

NRC. 2007. Natural Resources Canada: Canadian Intersite Decomposition Experiment (CIDET). Canadian Forest Service. Available at: http://cfs.nrcan.gc.ca/projects/76. Accessed: April 4, 2013.

Olson, J.S. 1963. Energy storage and balance of producers and decomposers in ecological systems. *Ecology*, 44:322-331.

Parks Canada. 2006. Ecological Monitoring and Assessment Network (EMAN): Terrestrial monitoring protocol annual soil humus decay rates (Draft 3). EMAN Coordinating Office, Burlington, Ontario.

Perry, J.N. 1981. Taylor’s power law for dependence of variance on mean in animal populations. Journal of the Royal Statistical Society. Series C (Applied Statistics), 30(3):254-263.

Robson, L. 2010. Ecological Monitoring at ***rare*** Charitable Research Reserve 2010. Available on ***rare*** server.

Schlentner, R.E. and Van Cleve, K. 1985. Relationships between CO_2 evolution from soil, substrate temperature, and substrate moisture in four mature forest types in interior Alaska. *Canadian Journal of Forest Research*, 15:97-106.

Schlesinger, W.H., and Andrews, J.A. 2000. Soil respiration and the global carbon cycle. *Biogeochemistry*, 48(1):7-20.

Singh, J.S. and Gupta, S.R. 1977. Plant decomposition and soil respiration in terrestrial ecosystems. *Botanical Review,* 43(4):449-528.

Taylor, B.R., and Parkinson, D.1988. Does repeated freezing and thawing accelerate decay of leaf litter? *Soil and Biogeochemistry*, 20(5):657-665.

Trumbore, S.E., Chadwick, O.A, and Amundson R. 1996. Rapid exchange between soil carbon and atmospheric carbon dioxide driven by temperature change. *Science*, 272(5260):393-396.

Ure, D., Chisholm, S., and Kehler, D. 2010. Kejimkujik National Park and National Historic Site: Technical Compendium to the 2010 State of Park Report. Parks Canada. Available at: http://www.merseytobeatic.ca/userfiles/file/products/publications/Kejimkujik%20NPNHS%202011%20SOPR%20Technical%20compendium.pdf. Accessed: Dec. 16, 2015.

USDA. 2007. United States Department of Agriculture: The Encyclopedia of Wood. Skyhorse Publishing Inc., Washington, D.C., USA.

Van Cleave, K. 1971. Energy and weight loss functions for decomposing foliage in birch and aspen forests in interior Alaska. *Ecology*, 52:720-723.

Xu, L., Baldocchi, D.D., and Tang, J. 2004. How soil moisture, rain pulses, and growth alter the response of ecosystem respiration to temperature. *Global Biogeochemical Cycles*, DOI: 10.1029/2004GB002281

APPENDIX A: Maps and Coordinates

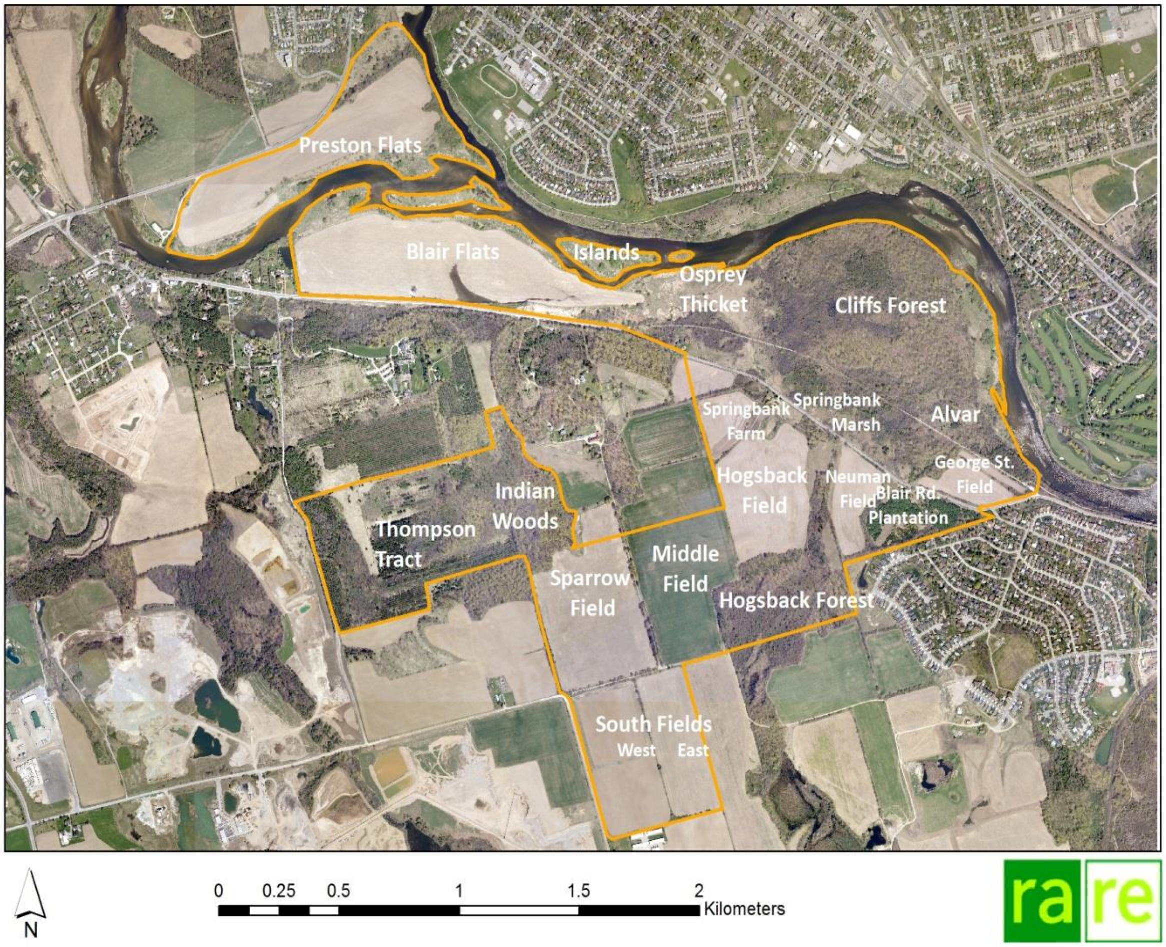


Figure A.1: Property map of the ***rare*** **Charitable Research Reserve** with colloquial names of properties.

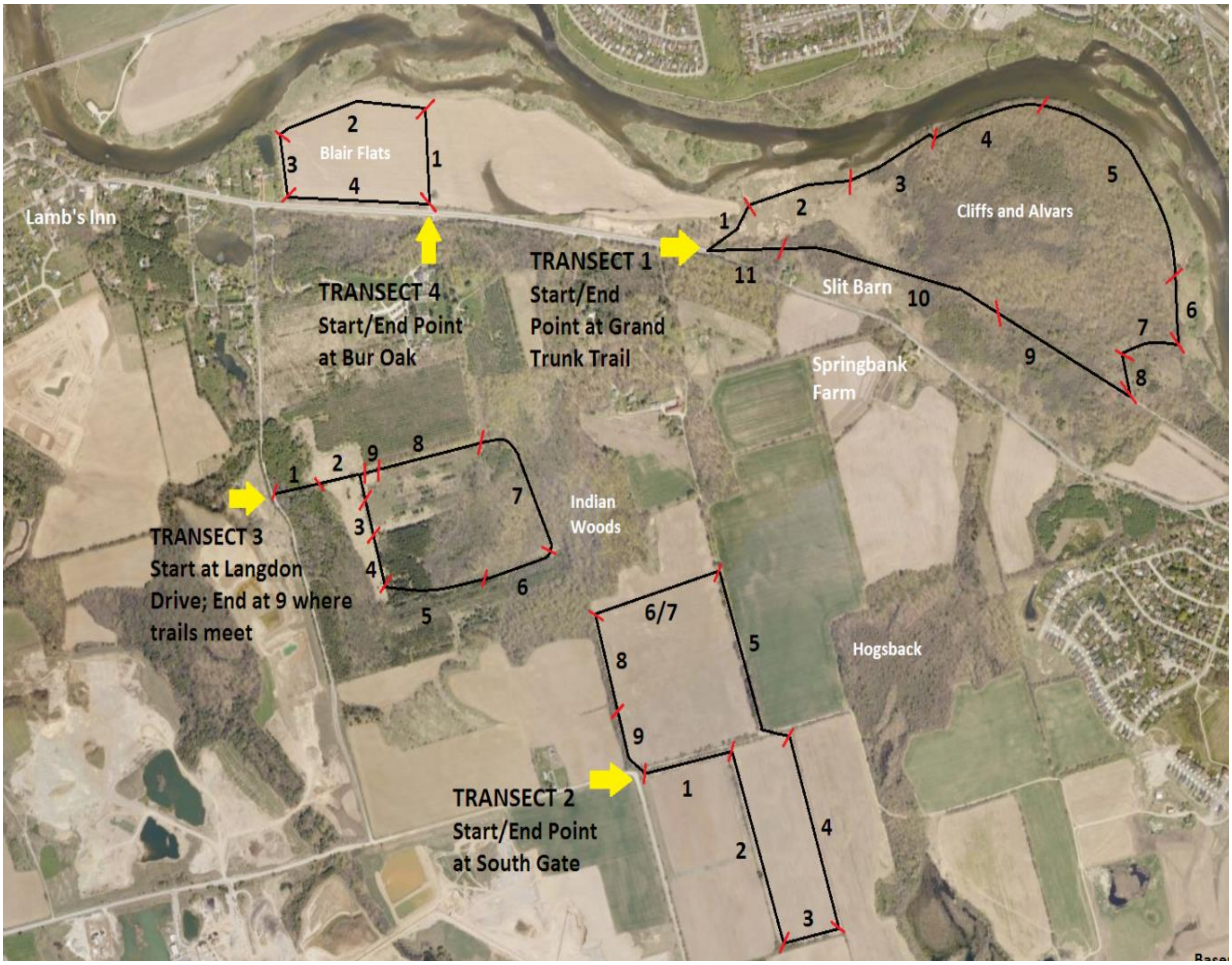


Figure A.2: Location of the four butterfly monitoring transects at the ***rare* Charitable Research Reserve** with start/end points and section break.

Figure A.3: Location of forest health, humus decay rate, and salamander monitoring plots in Indian Woods, Cliffs and Alvars and the Hogsback forest areas. Each forest stand has three forest health monitoring plots and one humus decay rate plot located at plot one. Salamander plots overlap with plot three in Indian Woods and the Hogsback and with plot two in Cliffs and Alvars.

List A.1: Description of Transect One sections with stopping point coordinates (GPS coordinate accuracy less than 10m).

Section one (N 43° 22.980' W 80° 21.541')

- Riparian grassland (milkweed, goldenrod, grasses)
- Stop past the sedge wetland, toward the river at the solitary shrub

Section two (N 43° 23.025' W 80° 21.426')

- Riparian meadow with trees and shrubs on south side
- Stop at old fallen tree in middle of field, within direct view of the osprey tower, 100m

Section three (N 43° 23.058' W 80° 21.222')

- Riparian area with trees on south side (grasses, sedges, small shrubs, goldenrod)
- Stop in open grass area with small hill on right hand side just after trail turns away from river, before continuing into forest

Section four (N 43° 23.120' W 80°21.017')

- Mainly coniferous forest trail with open canopy areas, on cliffs
- Stop when path forks to small lookout over the river to the left, break in cedar dominance

Section five (N 43° 22.986' W 80°20.625')

- Deciduous forest trail
- Stop at large fallen tree over trail, trail has moved around log; cliffs on south side and open meadow (milkweed, raspberry, goldenrod, one Oak) on north side

Section six (N 43° 22.761' W 80°20.617'

- Open shrub land
- Stop at alvar on the left hand side of trail right after the old car on the right hand side, large red pine on the trail edge and large white pine further back near alvar

Section seven (N 43° 22.767' W 80° 20.697')

- Deciduous forest trail
- Stop at large alvar, ~10m after tall Oak tree

Section eight (N 43° 22.749' W 80° 20.734')

- Open shrub land
- Stop on second boardwalk

Section nine (N 43° 22.793' W 80° 20.901')

- Grand Trunk Trail, deciduous forest
- Stop at culvert in wetland

Section ten (N 43° 22.901' W 80° 21.250')

- Grand Trunk Trail, dense shrub growth on both sides of trail
- Stop at entrance to Osprey Tower path to the north, and path to Slit Barn to the south

Section eleven (N 43° 22.927’ W 80° 21.546’)

- Grand Trunk Trail, wetland on either side of trail (sedges, cattail, milkweed, goldenrod, purple loosestrife)
- Stop at culvert near Blair Road entrance to Grand Trunk Trail, several Trembling Aspen trees, direct line of sight to stopping point for section one

List A.2: Description of Transect Two sections with stopping point coordinates (GPS coordinate accuracy less than 10m).

Section one (N 43° 22.177' W 080° 21.691')

- Agricultural field (mix of alfalfa, red fescue, perennial wild rye, buckwheat, winter wheat and oats) to south of transect, deciduous trees and shrubs to the north
- Stop at north side of South Field West in naturalized buffer, directly across from silo at farm to the south

Section two (N 43° 22.048' W 080° 21.560')

- Hedgerow along winter wheat field edge, mostly open with some shrubs
- Stop halfway along west side of South Field East, near solitary Buckthorn shrub & old collapsed wooden structure

Section three (N 43° 21.909' W 080° 21.438')

- Hedgerow of deciduous trees along edge of winter wheat field
- Stop halfway along south side of South Field East, at the end of the tree line to the north, before the row of three single trees

Section four (N 43° 22.050' W 080° 21.404')

- Hedgerow on east side of winter wheat field, mostly open with few shrubs along fence
- Stop halfway along field edge, blue post on east side of fence

Section five (N 43° 22.402' W 080° 21.620')

- Deciduous hedgerow of mostly Oak trees; bordering winter wheat field on east side and naturalized agricultural field on west side
- Stop after open canopy, once there is partial canopy coverage again

Section six/seven (N 43° 22.423' W 080° 21.771')

- Naturalized agricultural field, with grasses, wildflowers, and some saplings (maple)
- Stop halfway across field, just before the bird boxes to the south

Section eight (N 43° 22.299' W 080° 21.892')

- Hedgerow of deciduous trees (mostly Maple) bordering naturalized agricultural field
- Stop at top of hill at fallen tree, can see apartment building to the east

Section nine (N 43° 22.212' W 080° 21.857')

- Hedgerow (east of Grand Allee Trail) of mainly shrubs, vines and grasses bordering naturalized agricultural field
- Stop on incline past large group of young maple trees, 20 meters before path to Grand Allee

List A.3: Description of Transect Three sections with stopping point coordinates (GPS coordinate accuracy less than 10m).

Section one (N 43° 22.584’ W 080° 22.569’)
- Coniferous forest (Ash trees, Cedar trees, shrubs)
- Stop at swampy meadow just past culvert (goldenrod, cattails, milkweed)

Section two (N 43° 22.601’ W 080° 22.469’)
- Meadow (milkweed, goldenrod, grasses, sedges)
- Stop at junction of trails

Section three (N 43° 22.541’ W 080° 22.454’)
- Black Locust plantation and meadow
- Stop halfway through plantation area, where tree has grown around top wire of fence on east side

Section four (N 43° 22.482’ W 080° 22.430’)
- Meadow (milkweed, goldenrod, grasses, sedges) on west side of transect, Spruce tree forest on east side
- Stop at third large Spruce tree on east side, about halfway down the straight portion of the trail

Section five (N 43° 22.424’ W 080° 22.301’)
- Spruce and deciduous forest
- Stop where wet area ends (will change from year to year), small clearing to the north, several small trees leaning across path

Section six (N 43° 22.476’ W 080° 22.064)
- Meadow (grasses, sedges) and Walnut tree plantation
- Stop halfway down straight section of walnut trees, dead and fallen White Pine on north side with young maples around it

Section seven (N 43° 22.568’ W 080° 22.158’)
- Grand Allee Trail in Indian Woods (deciduous forest of Sugar Maple, Beech and Oak trees with woodland plants and flowers such as may apple, solomon’s seal, trillium and ferns)
- Stop on cement bridge over Bauman Creek

Section eight (N 43° 22.635’ W 080° 22.273’)
- Maple Lane Trail (deciduous forest of Sugar Maple and shrubs)
- Stop near small pile of logs on south side of trail

Section nine (N 43° 22.606’ W 080° 22.437’)
- Meadow (vetch, goldenrod, grasses, sedges, scattered trees and shrubs)
- Stop halfway before the junction of trails, between two stumps on north side of trail

List A.4: Description of Transect Four sections with stopping point coordinates (GPS coordinate accuracy less than 10m).

Section one (N 43° 23.090' W 080° 22.307')

- Weedy meadow planted for tall grass prairie, recovering from agricultural use (horseweed, black-eyed susan, goldenrod)
- Walk from Bur Oak toward tower in distance, stop halfway before field edge in between two University of Guelph plant enclosures

Section two (N 43° 23.131' W080° 22.523')

- Regeneration area to the north side of transect and planted tall grass prairie to the south (black-eyed susan, burdock, goldenrod, horseweed, tansy, thistles)
- Stop halfway along field edge, just after the bird boxes

Section three (N 43° 23.056' W 080° 22.641')

- Hedgerow of shrubs and trees to the west of transect and planted tall grass prairie to east of transect (black-eyed susan, burdock, goldenrod, horseweed, tansy, thistles)
- Stop halfway along field edge, hot tub on west side of transect

Section four (N 43° 22.998' W 080° 22.473')

- Hedgerow along Blair Road to the south of transect and planted tall grass prairie to north of transect (Black-eyed Susan, Horseweed, Manitoba Maple, Tansy, thistles, and shrubs)
- Stop halfway along field edge, where shrubs are tallest

Table A.1: GPS coordinates of artificial cover objects (ACO) used for plethodontid salamander monitoring in Indian Woods and the Hogsback (from McCarter 2009).

Monitoring Plot	ACO	Latitude and Longitude	UTM (zone 17T)
Indian Woods	1	N43°22'32.05" W80°21'55.49"	551408E 4802718N
	9	N43°22'31.97" W80°21'53.71"	551448E 4802716N
	17	N43°22'30.97" W80°21'53.63"	551450E 4802685N
	25	N43°22'30.85" W80°21'55.37"	551411E 4802681N
Hogsback	1	N43°22'23.93" W80°21'12.74"	552372E 4802475N
	8	N43°22'22.99" W80°21'13.32"	552359E 4802446N
	11	N43°22'22.44" W80°21'12.84"	552370E 4802429N
	18	N43°22'23.57" W80°21'12.30"	552382E 4802464N

A)   B)

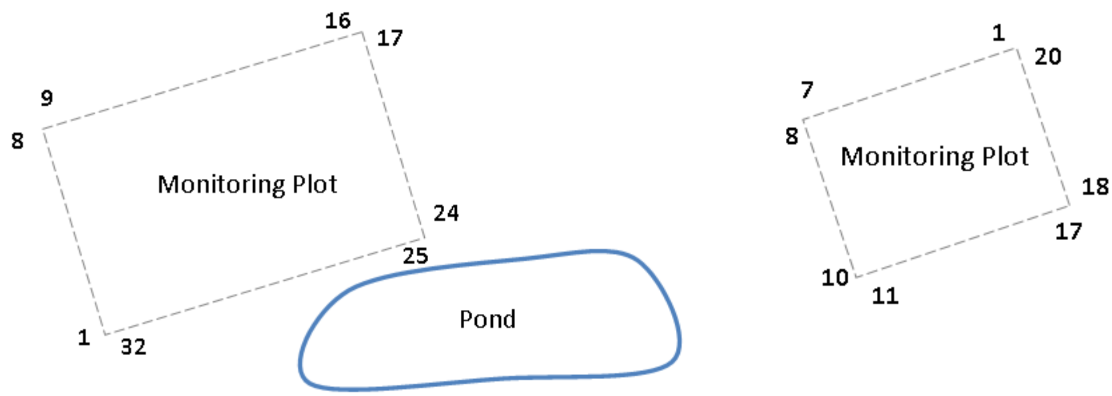
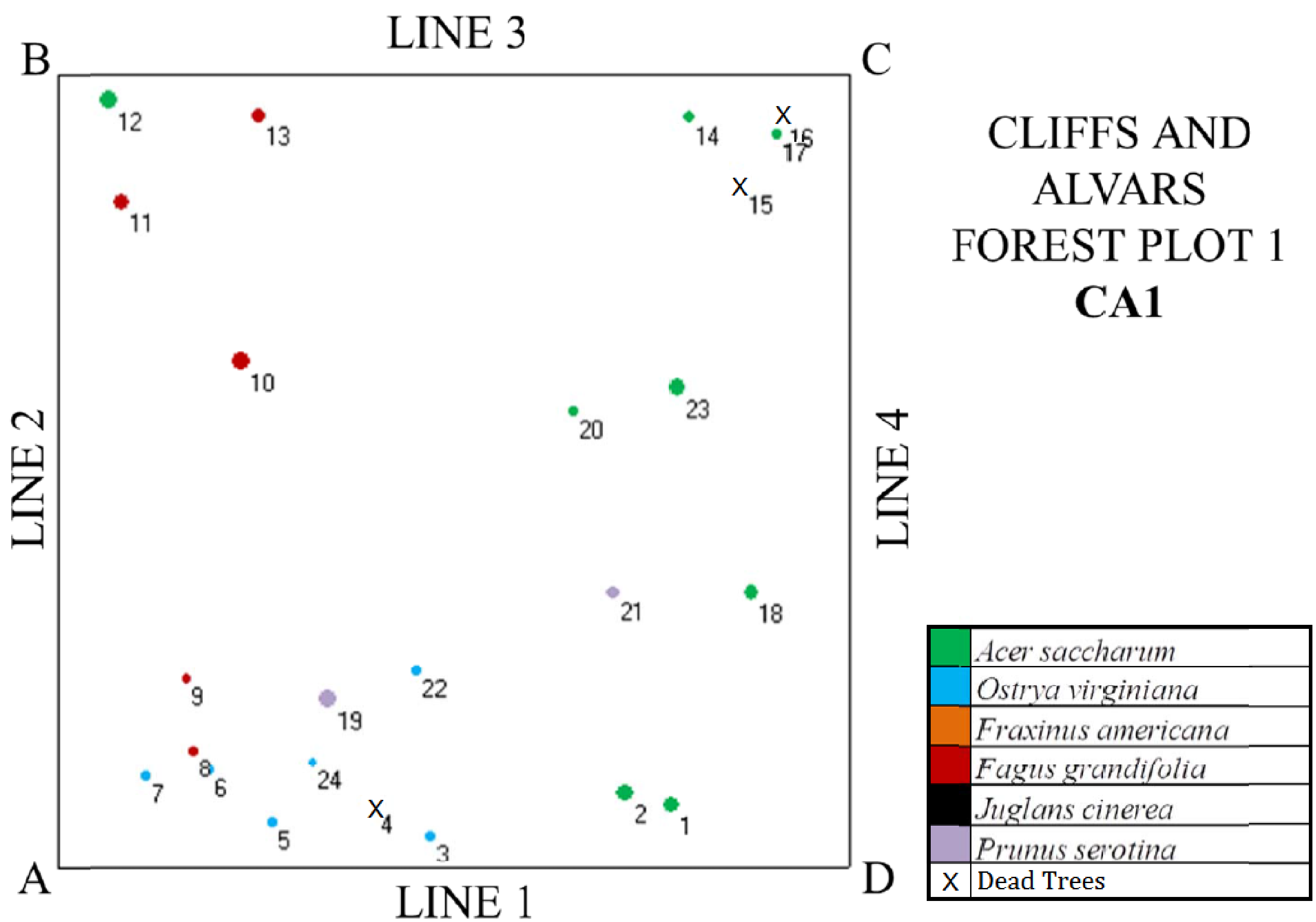


Figure A.4: Layout of artificial cover objects (ACOs) on salamander monitoring plots in A) Indian Woods and B) Hogsback.

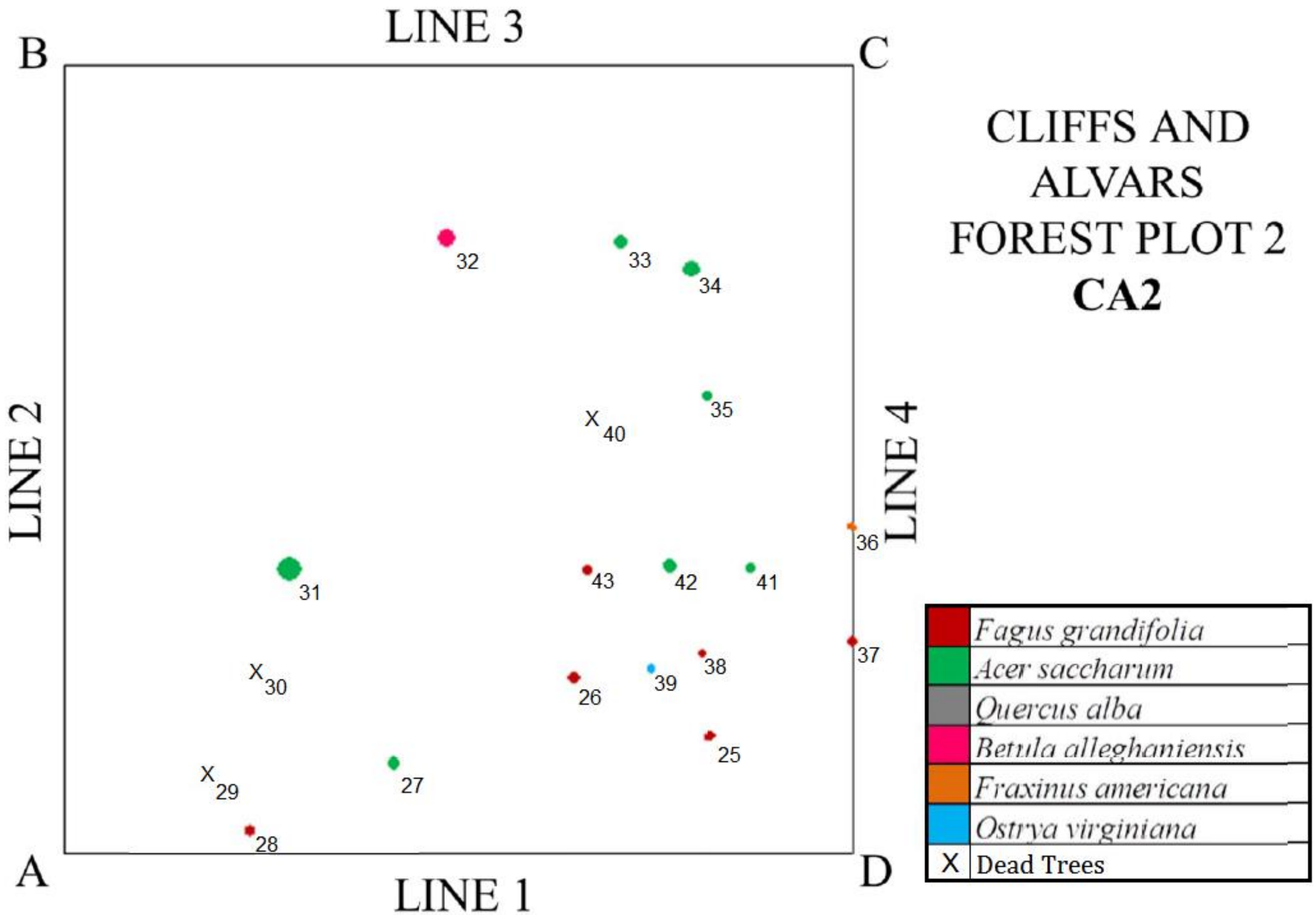
Table A.2: GPS coordinates of forest canopy tree biodiversity and health monitoring plots in Cliffs and Alvars, Indian Woods, and the Hogsback (from Robson 2010). The coordinates describe the location of the northwest corner of each plot. The annual soil decay rate monitoring plots are located on all four corners of forest plot one in each stand.

Monitoring Location	Plot	Latitude and Longitude	UTM (zone 17T)
Cliffs and Alvars	1	N43°22’46.30” W80°21’1.34”	552623E 4803167N
	2	N43°22’44.64” W80°21’0.21”	552649E 4803116N
	3	N43°22’43.72” W80°20’57.91”	552701E 4803088N
Indian Woods	1	N43°22’27.27” W80°21’51.45”	551500E 4802571N
	2	N43°22’26.12” W80°21’56.08”	551396E 4802535N
	3	N43°22’23.62” W80°21’54.78”	551426E 4802458N
Hogsback	1	N43°22’24.18” W80°21’11.10”	552409E 4802483N
	2	N43°22’23.28” W80°21’12.66”	552374E 4802455N
	3	N43°22’22.08” W80°21’14.46”	552334E 4802418N

Figure A.5-A.13: Maps of Cliffs and Alvars, Indian Woods, and the Hogsback forest biodiversity monitoring plots showing location of all standing, live trees with a diameter at breast height (dbh) greater than 10.0cm. Sizes of circles are proportional to real tree diameters, colours indicate different species.

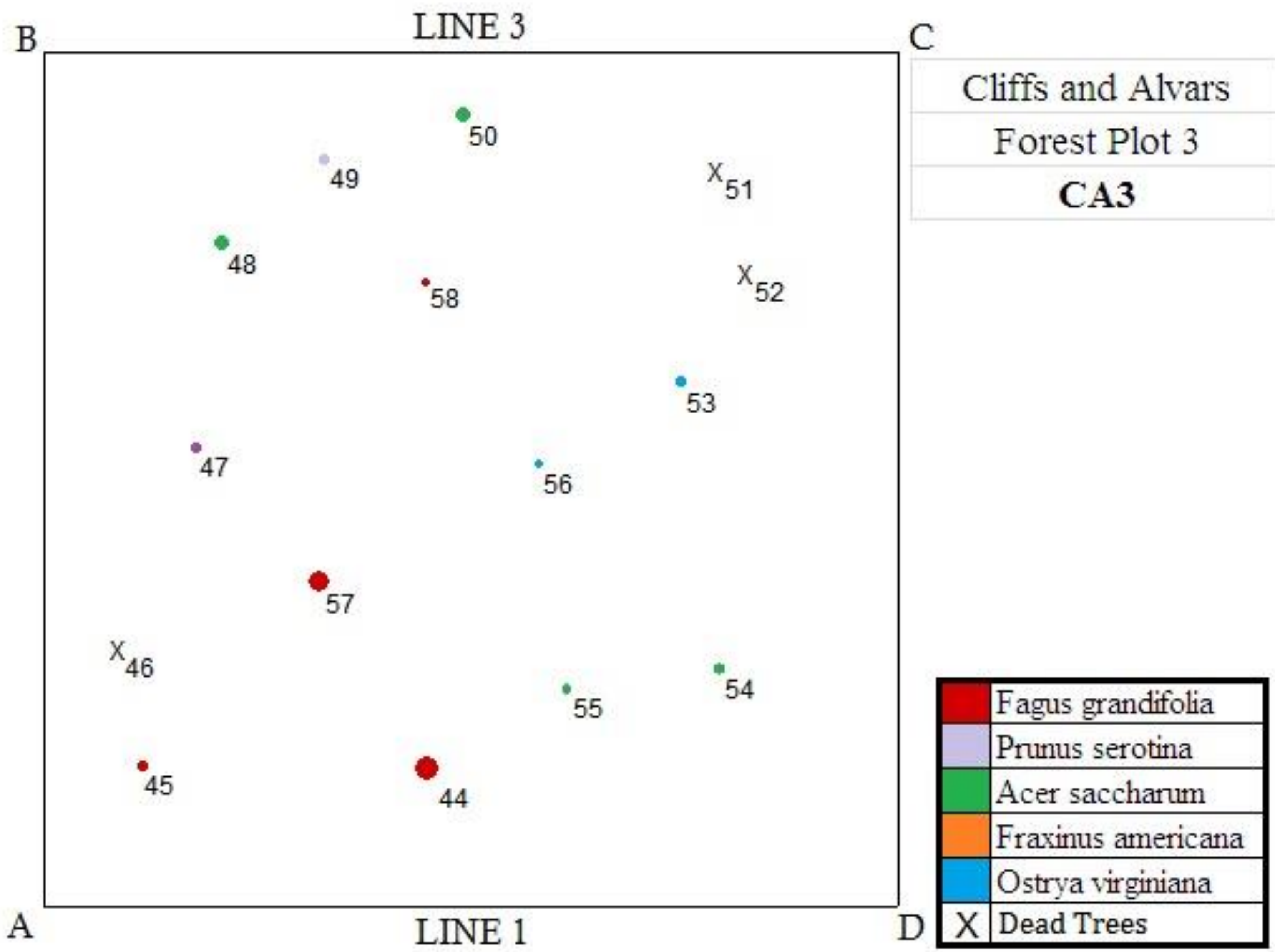


CLIFFS AND ALVARS FOREST PLOT 1
CA1

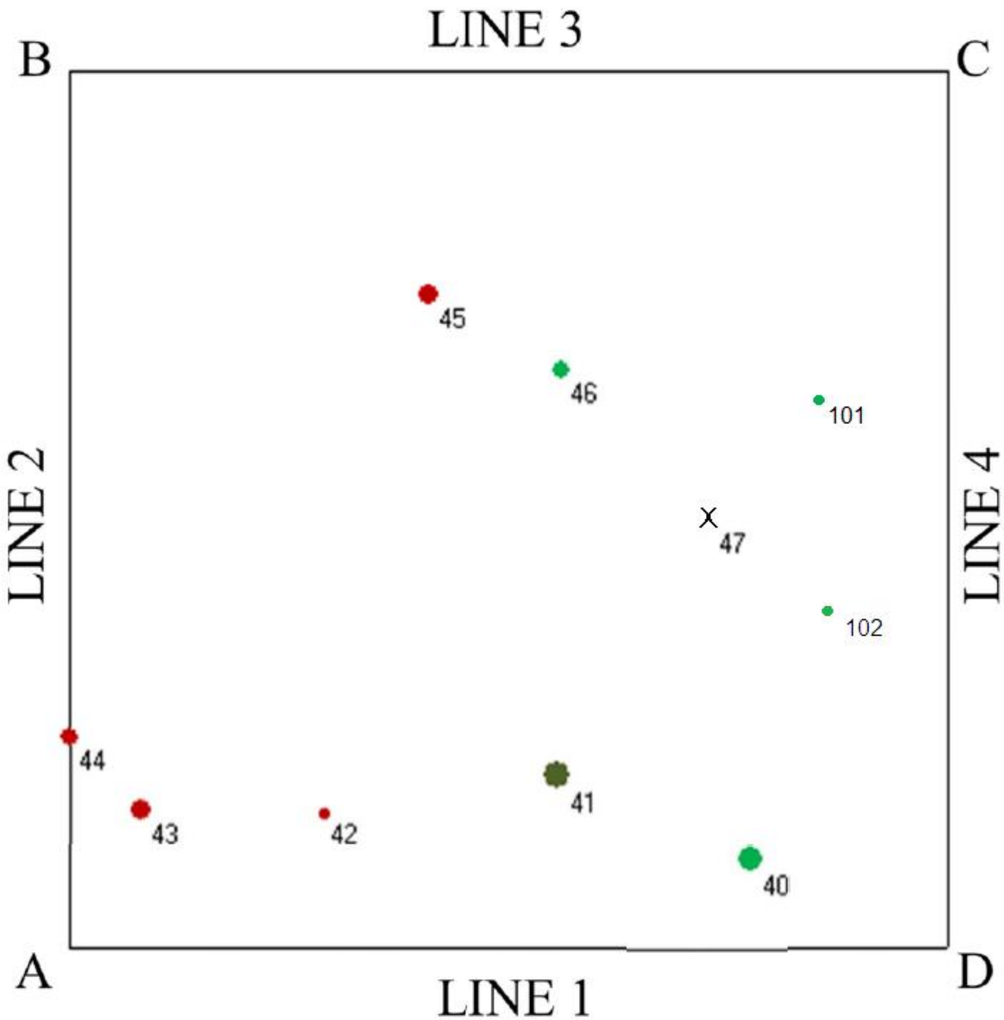


CLIFFS AND ALVARS FOREST PLOT 2

CA2

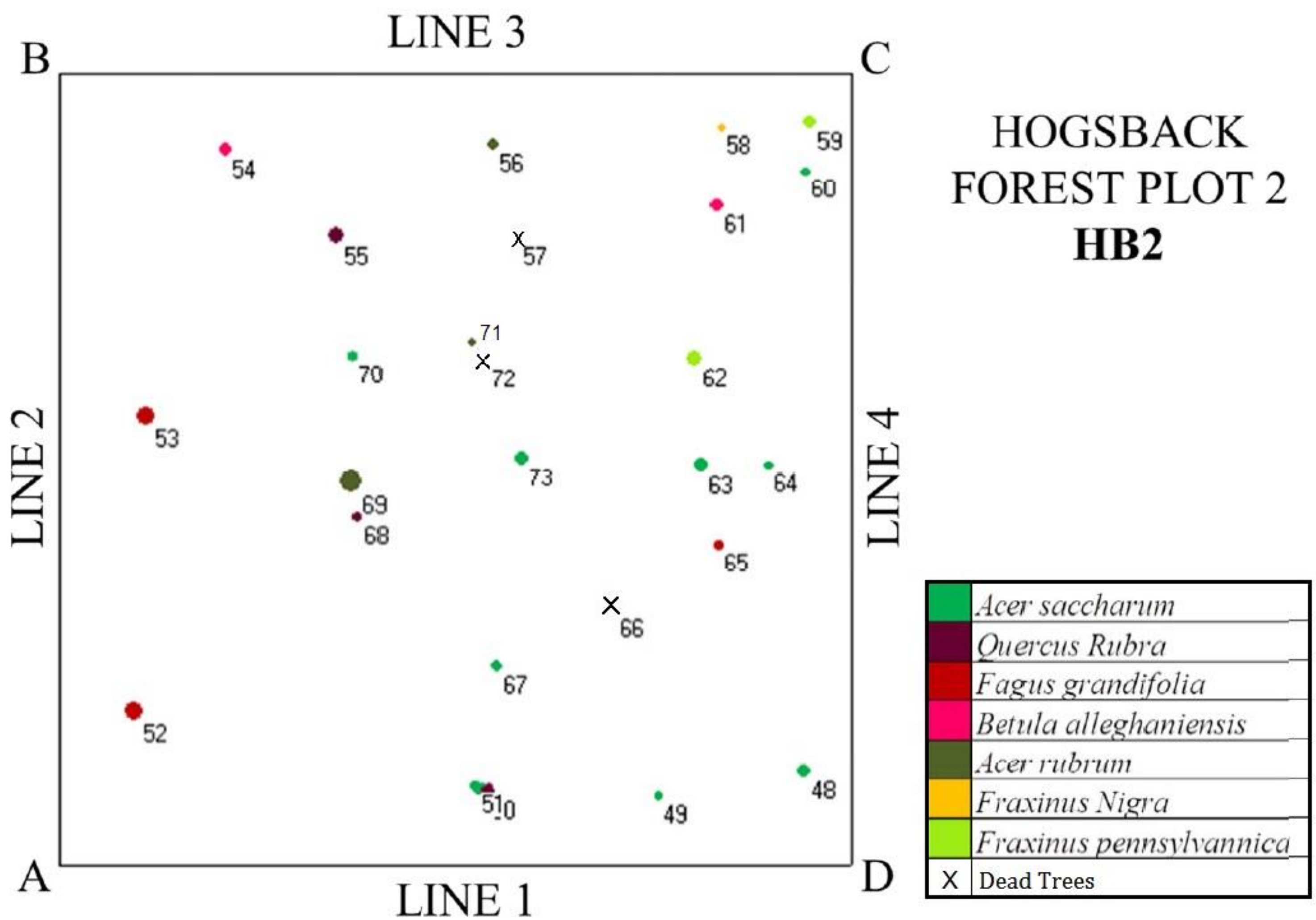


LINE 3
B
C
A
D
LINE 1
50
49
51
48
58
52
53
47
56
57
46
55
54
45
44
Cliffs and Alvars
Forest Plot 3
CA3
Fagus grandifolia
Prunus serotina
Acer saccharum
Fraxinus americana
Ostrya virginiana
X Dead Trees

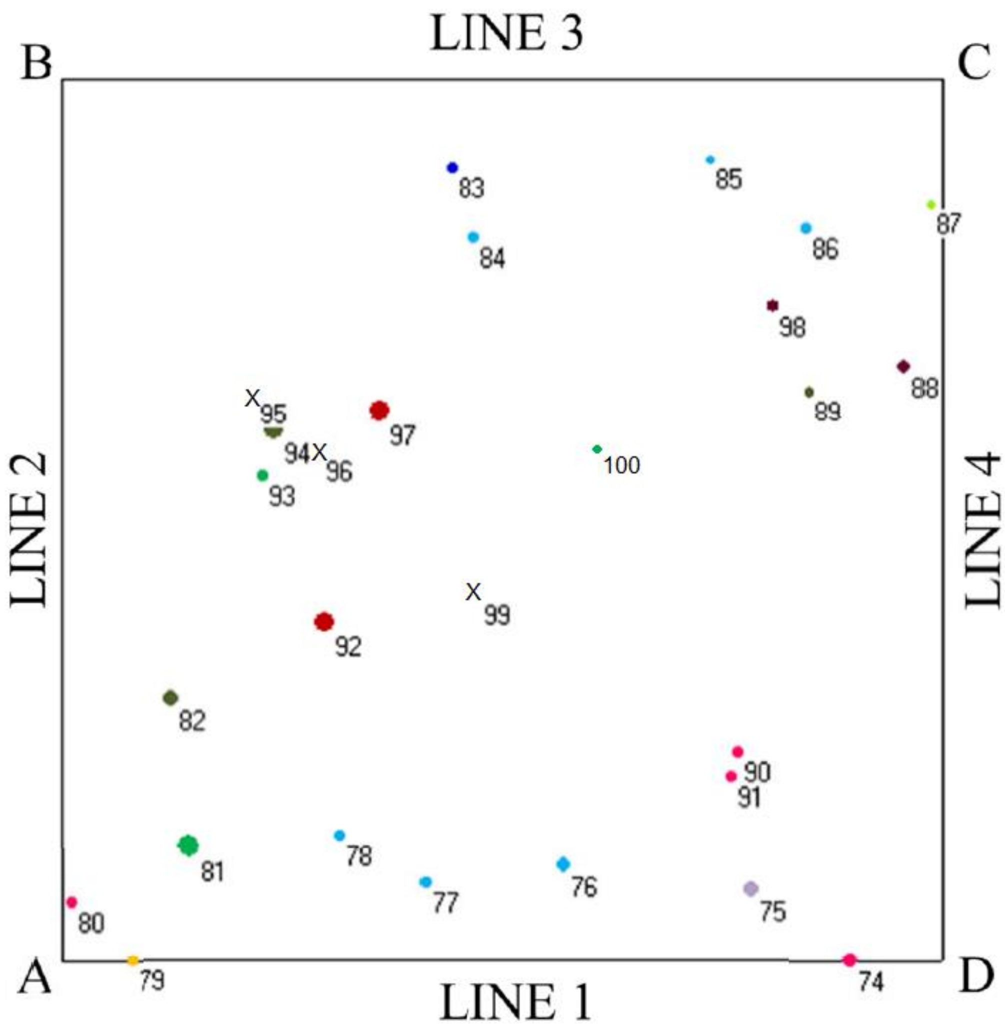


HOGSBACK
FOREST PLOT 1
HB1

	Acer saccharum
	Acer rubrum
	Fagus grandifolia
X	*Dead Trees*

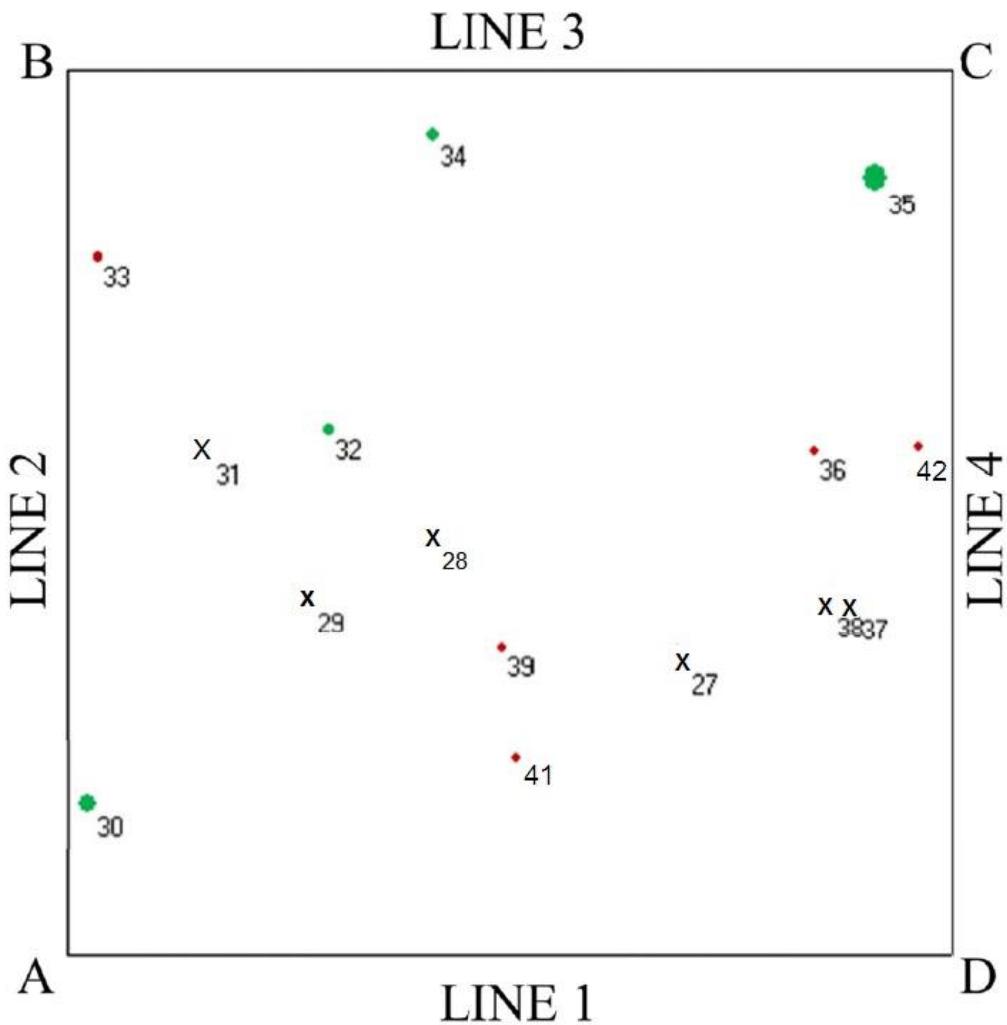


HOGSBACK
FOREST PLOT 2
HB2



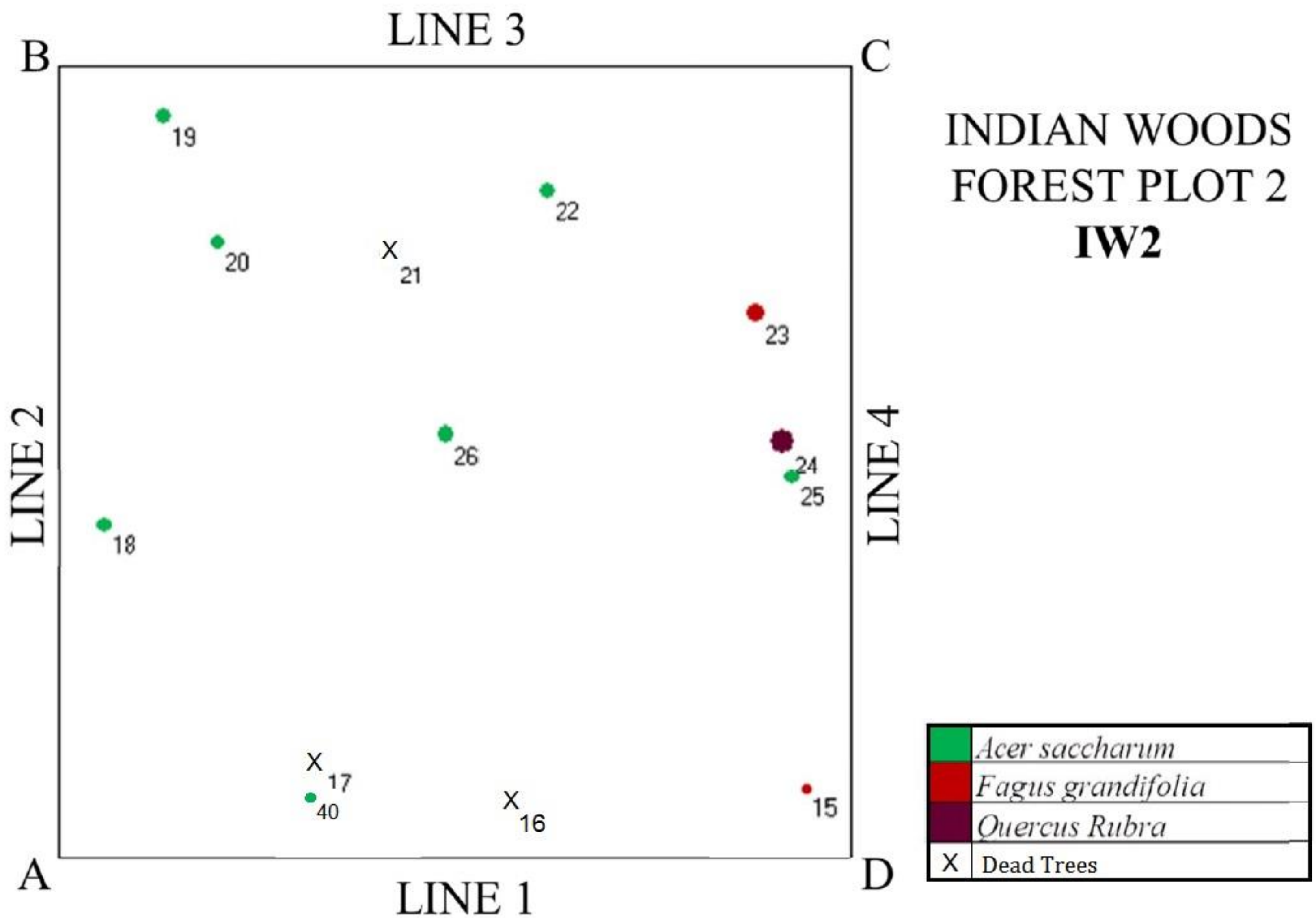
HOGSBACK FOREST PLOT 3
HB3

	Betula alleghaniensis
	Prunus serotina
	Ostrya virginiana
	Fraxinus Nigra
	Acer saccharum
	Acer rubrum
	Pinus strobus
	Fraxinus pennsylvannica
	Quercus Rubra
	Fagus grandifolia
X	Dead Trees

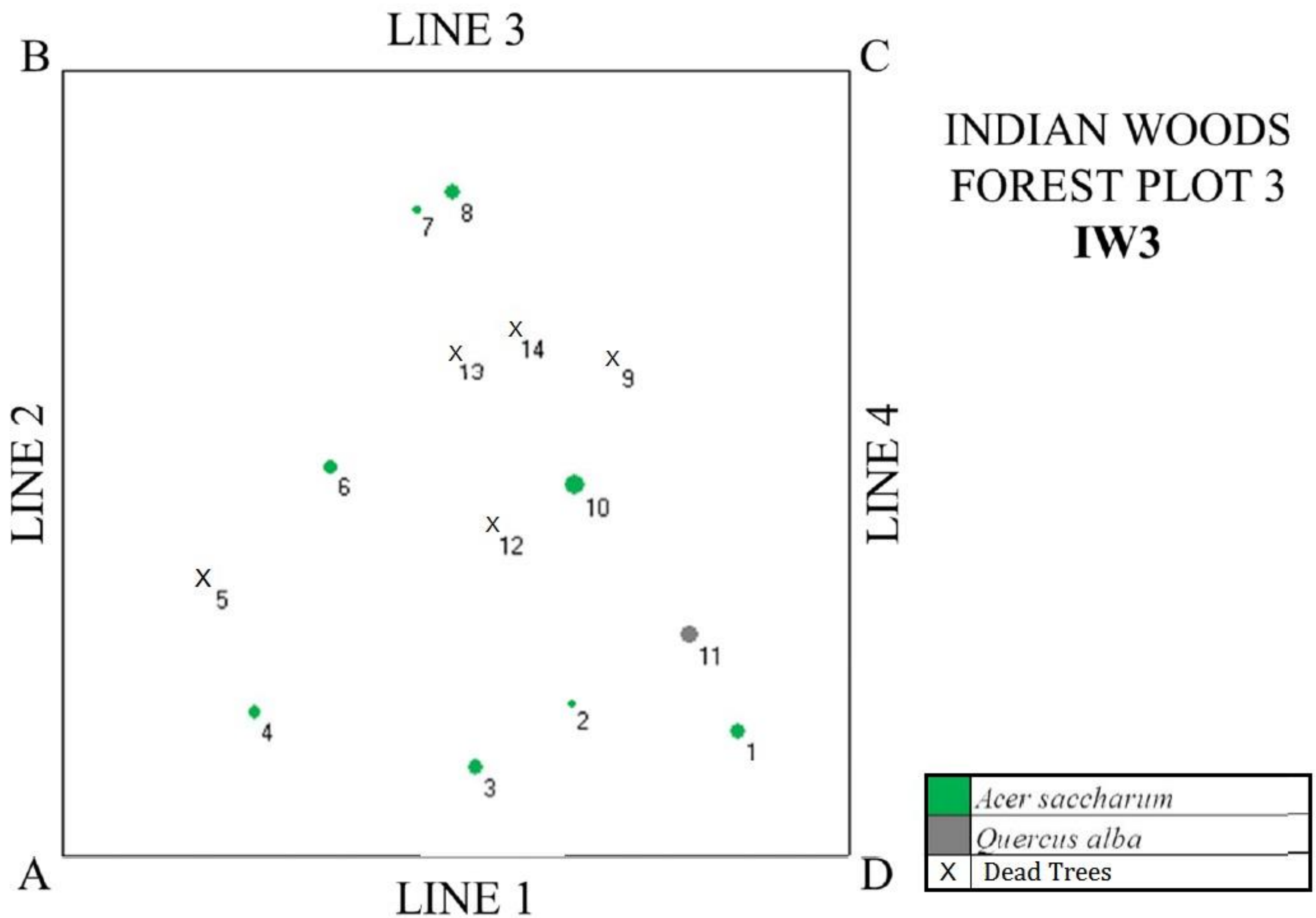


INDIAN WOODS
FOREST PLOT 1
IW1

(green)	*Acer saccharum*
(red)	*Fagus grandifolia*
X	Dead Trees



LINE 3
B
C
INDIAN WOODS
FOREST PLOT 2
IW2
19
22
20
X 21
23
LINE 2
26
24
25
LINE 4
18
Acer saccharum
Fagus grandifolia
Quercus Rubra
X Dead Trees
X 17
40
X 16
15
A
LINE 1
D


LINE 3
B
C
LINE 2
LINE 4
A
D
LINE 1
8
7
14
13
9
6
10
12
5
11
4
2
1
3
INDIAN WOODS
FOREST PLOT 3
IW3
Acer saccharum
Quercus alba
X Dead Trees

APPENDIX B: Equipment List

List B.1: Suggested butterfly monitoring field equipment.

- Field data sheet
- Clipboard
- Pencils
- Stopwatch
- Kestrel 3000©
- Butterfly net
- Binoculars
- Field guide (Recommended: Carmichael, I. and Vance, A. 2003. Photo Field Guide to the Butterflies of Southern Ontario. St. Thomas Field Naturalist Club Inc., St. Thomas, ON.)
- Clear jar with mesh lid
- Digital Camera

List B.2: Salamander monitoring equipment list

- Field data sheets A and B on waterproof paper
- Clipboard
- Pencils
- Nitrile gloves
- Kestral 3000 pocket weather station
- Soil moisture meter (calibrated with screw driver)
- Soil thermometer
- Digital calipers
- Ruler
- Soil pH Meter
- Digital pocket scale (with spare batteries)
- Sandwich sized plastic container filled with moist sponges
- Larger plastic container with some moist sponges
- Wash bottle filled with pond water from education pond
- Flagging tape
- Aluminum tags
- Digital camera

List B.3: Forest canopy tree biodiversity monitoring equipment list

- Blank canopy-sample and tree condition field data sheets on waterproof paper
- Past year data sheets & EMAN reference package
- Clipboard
- Pencils
- Flagging tape
- Diameter tape
- Two nylon tape measures (30m)
- Field guide
- Binoculars
- Clinometer
- Pre-labelled tags and steel pigtail stakes

List B.4: Soil humus decay rate monitoring equipment list

Installation

- Field data sheet on waterproof paper
- Clipboard
- Pencils
- Nitrile gloves
- Shovel
- Trowel
- Chisel
- Pigtail stakes (12 per plot)
- Tongue depressors (decay sticks), pre-weighed, dried, and labelled
- Pre-prepared mesh bags
- Fishing line

Extraction

- Field data sheet on waterproof paper
- Clipboard
- Pencils
- Nitrile gloves
- Trowel
- Scissors
- Utility knife
- Re-sealable plastic bags
- Permanent marker

Cleaning

- Nitrile gloves
- Scissors
- Two paint brushes (one wet and one dry)
- Paper envelopes

APPENDIX C: DATA SHEETS AND CODES

Table C.1: Beaufort wind codes (Zorn et al. 2004)

Beaufort Scale	Wind Speed (mph)	Wind Speed (km/h)	Description
0	1	1.6	Calm. Smoke rises vertically.
1	2	3.2	Light. Smoke drifts.
2	5	8	Light breeze. Leaves rustle.
3	10	16	Gentle breeze. Lighter branches sway.
4	15	24	Moderate breeze. Dust rises. Branches move.
5	21	33.6	Fresh breeze. Small trees sway.
6	28	44.8	Strong breeze. Larger branches move.
7	35	56	Moderate gale. Trees move.
8	42	67.2	Fresh gale. Twigs break.
9	50	80	Strong gale. Branches break.
10	59	94.4	Whole gale. Trees fall.
11	69	110.4	Storm. Violent blasts.
12	75	120	Hurricane. Structures shake.

Table C.2: Beaufort sky codes (Zorn et al. 2004)

Sky Code	Description
0	Clear. No clouds.
1	Partly cloudy. Scattered or broken clouds.
2	Cloudy (broken) or overcast.
3	Sandstorm. dust storm, or blowing snow.
4	Fog, thick dust or haze.
5	Drizzle.
6	Rain.
7	Snow, or snow rain mixed.
8	Shower(s).
9	Thunderstorm(s).

BUTTEFLY MONITORING FIELD NOTES									
DATE:				START:			TEMP_START:		
TRANSECT:				FINISH:			TEMP_END:		
1			S:	SUN:	2			S:	SUN:
W1:				WIND:	W1:				WIND:
W2:					W2:				
3			S:	SUN:	4			S:	SUN:
W1:				WIND:	W1:				WIND:
W2:					W2:				
5			S:	SUN:	6			S:	SUN:
W1:				WIND:	W1:				WIND:
W2:					W2:				
NOTES:									

Figure C.1: Sample butterfly monitoring field data sheet (available on the ***rare*** server).

Field Data Sheet A			
Plot Name:		Group Name: rare Charitable Research Reserve	
Observer Name(s):			
Pond depth (mm; Indian Woods):		Date:	Time:
Precip.(last 24hrs):	Beaufort Sky Code:		Beaufort Wind Code:

ACO Number	Species	Count	ACO: Type	Age	Soil: Temp	Moisture	ACO Disturbance

Additional Comments:

	North Perimeter		**East Perimeter**		**South Perimeter**		**West Perimeter**	
ACO #:	IN-02-03	IN-02-07	IN-02-11	IN-02-15	IN-02-19	IN-02-23	IN-02-27	IN-02-31
WS (mph)								
RH (%)								
AT (C)								

WS= Wind Speed RH= Relative Humidity AT= Air Temperautre

Figure C.2: Sample of salamander monitoring field sheet A (available on ***rare*** server).

Field Data Sheet B							
Plot Name:				Group Name: rare Charitable Research Reserve			
Observer Name(s):							
Pond depth (Indian Woods):				Date:		Time:	
Precip.(last 24hrs):			Beaufort Sky Code:			Beaufort Wind Code:	
ACO Number	Cumulative Number of Salamanders	Species	Length (mm)			Weight (g)	Comments
			S-V	V-T	Total		
Additional Comments:							

Figure C.3: Sample of salamander monitoring field sheet B (available on ***rare*** server).

CANOPY-TREE SAMPLE: FIELD DATA SHEET (1-ha. plot or 20 m x 20 m stand-alone quadrats). Stand name.. Date..

Stand location (lat. & long.) ....Hectare plot and quadrat nº...........................OR Stand-alone quadrat nº........... Av. stand height..

Identification manual................................... Observer(s)..

Tag #	Species name	Number of stems	dbh (cm)	Line (1,2,3,4)	A distance (m)	B distance (m)	Height (m)	Condition	Notes

Figure C.4: Sample of forest canopy tree biodiversity monitoring sheet (available on ***rare*** server).

CANOPY-TREE SAMPLE: SUMMARY SHEET. Stand name.. Date..

Stand location (lat. & long.).. Number of 1-ha plots .... **OR** Number of 20 m x 20 m stand-alone quadrats ....

Sample area (m^2)................ Average canopy height .... Data processor(s)...

Species name	Abundance	Density	Basal Area	Dominance	Frequency	Relative Density	Relative Frequency	Relative Dominance	Importance Value	Notes

Figure C.5: Sample of forest canopy tree health monitoring field sheet, tree condition (available on ***rare*** server).

Annual Decay Rate Data Sheet										
Fieldworker(s):										
Notes:										
			YEAR INSTALLED						YEAR EXTRACTED	
Stand ID	Plot ID	ADR Station ID	Tag Number	Original weight (g)	Placement (s/ b)	Humus depth (cm)	Buried depth (cm)	Date Buried	Date Retrieved	Decayed weight (g)

Figure C.6: Sample of annual soil humus decay rate monitoring field sheet (available on ***rare*** server)

APPENDIX D: Species Lists

List D.1: Common and scientific names of all butterflies observed at the ***rare*** **Charitable Research Reserve** during all previous butterfly monitoring seasons and annual butterfly counts since 2006. A total of 75 butterfly species have been observed.

Common Name	Scientific Name
Acadian Hairstreak†	*Satyrium acadicum*
American Lady	*Vanessa virginiensis*
American Snout†	*Libytheana carinenta*
Appalachian Brown	*Satyrodes appalachia*
Arctic Skipper	*Carterocephalus palaemon*
Baltimore Checkerspot	*Euphydryas phaeton*
Banded Hairsteak	*Satyrium calanus*
Black Dash	*Euphyes conspicua*
Black Swallowtail	*Papilio polyxenes*
Broad-Winged Skipper	*Poanes viator*
Bronze Copper	*Lycaena hyllus*
Cabbage White	*Pieris rapae*
Clouded Sulphur	*Colias philodice*
Columbine Duskywing	*Erynnis lucilius*
Common Buckeye	*Junonia coenia*
Common Sooty Wing	*Philodice catullus*
Common Wood-Nymph	*Cercyonis pegala*
Compton Tortoiseshell	*Nymphalis vaualbum*
Coral Hairstreak	*Satyirum titus*
Crossline Skipper	*Polites origines*
Delaware Skipper	*Anatrytone logan*
Dion Skipper	*Euphyes dion*
Dreamy Duskywing*	*Erynnis icelus*
Dun Skipper	*Euphyes vestris*
Eastern Comma	*Polygonia comma*
Eastern Pine Elfin	*Callophrys niphon*
Eastern Tailed Blue	*Cupido comyntas*
Eastern Tiger Swallowtail	*Papilio glaucus*
European Skipper	*Thymelicus lineola*
Eyed Brown	*Satyrodes Eurydice*
Giant Swallowtail	*Papilio cresphontes*
Grey Comma	*Polygonia progne*
Great Spangled Fritillary	*Speyeria Cybele*
Harvester	*Feniseca tarquinius*
Hickory Hairstreak	*Satyrium caryaevorum*
Hobomok Skipper	*Poanes hobomok*
Indian Skipper*	*Hesperia sassacus*
Inornate Ringlet	*Coenonympha tullia*

Common Name	Scientific Name
Least Skipper	*Ancloxypha numitor*
Little Glassywing	*Pompeius verna*
Little Wood-Satyr	*Megisto cymela*
Little Yellow†	*Eurema lisa*
Long Dash	*Polites mystic*
Meadow Fritillary	*Boloria Bellona*
Milbert's Tortoiseshell	*Nymphalis milberti*
Monarch	*Danaus plexippus*
Mourning Cloak	*Nymphalis antiopa*
Mulberry Wing†	*Poanes massasoit*
Mustard White	*Pieris oleracea*
Northern Broken-Dash	*Wallengrenia egeremet*
Northern Crescent	*Phyciodes cocyta*
Northern Pearly-Eye	*Enodia anthedon*
Ocola Skipper*	*Panoquina ocola*
Orange Sulphur	*Colias eurytheme*
Painted Lady	*Vanessa cardui*
Pearl Crescent	*Phyciodes tharos*
Peck's Skipper	*Polites peckius*
Question Mark	*Polygonia interrogationis*
Red Admiral	*Vanessa atalanta*
Red-Spotted Purple	*Limenitis arthemis astyanax*
Sachem*	*Atalopedes campestris*
Silver-Bordered Fritillary	*Boloria selene*
Silvery Blue	*Glaucopsyche lygdamus*
Silvery Checkerspot	*Chlosyne nycteis*
Silver-Spotted Skipper	*Epargyreus clarus*
Spring Azure	*Celastrina ladon*
Striped Hairstreak	*Satyrium liparops*
'Summer' Spring Azure	*Celastrina neglecta*
Tawny-Edged Skipper	*Polites themistocles*
Tawny Emperor	*Asterocampa clyton*
Variegated Fritillary	*Euptoieta claudia*
Viceroy	*Limenitis archippus*
White Admiral	*Limenitis arthemis arthemis*
Wild Indigo Duskywing	*Erynnis baptisiae*

†Denotes observation only seen during annual butterfly count
*Denotes incidental observation outside of monitoring or annual count

Table D.1: The earliest record of observation for each butterfly species historically observed at the ***rare*** Charitable Research Reserve. The first date of observation is noted for each previous monitoring year and each annual butterfly count, as well as the overall earliest observation.

Species	Earliest Record By Year								Annual Butterfly Counts	Earliest Record at *rare*
	2006	2009	2010	2011	2012	2013	2014	2015		
Acadian Hairstreak									July 13 (2008)	July 13 (2008)
American Lady			May-20		May-15	May-22	Jul-17	May-20	July 10 (2010)	May 15 (2012)
American Snout					Jul-11				July 10 (2010)	July 10 (2010)
Appalachian Brown				Jul-06	Jun-18	Jul-02	Jul-03		July 2 (2011)	June 18 (2012)
Arctic Skipper			Jun-03			Jun-04	Jun-03		July 10 (2010)	June 3 (2010)
Baltimore Checkerspot					Jun-26				July 3 (2011)	June 26 (2012)
Banded Hairstreak	Jul-18	Jul-16		Jul-12	Jun-25	Jul-15	Jul-03	Jun-29	July 2 (2011)	June 25 (2012)
Black Dash			Jun-08		Jul-14	Jul-30	Jul-30	Jul-28	July 10 (2010)	June 8 (2010)
Black Swallowtail	Jul-21	May-20	May-04	May-30	May-14	May-22	May-23	May-20	July 10 (2010)	May 4 (2010)
Broad-Winged Skipper		Jul-24			Jul-14	Jul-12	Jul-18		July 10 (2010)	July 10 (2010)
Bronze Copper	Aug-18				Jun-06		Jun-20	Jun-24	July 2 (2011)	June 6 (2012)
Cabbage White	Jul-18	May-12	May-03	May-19	May-14	May-21	May-21	May-20	July 2 (2011)	May 3 (2010)
Clouded Sulphur	Jul-18	May-22	May-04	May-31	May-14	May-21	May-24	May-19	July 10 (2010)	May 4 (2010)
Columbine Duskywing			May-19		May-31			May-19		May 19 (2010)
Common Buckeye				Sep-15	Jun-06					June 6 (2012)
Common Sooty Wing	Jul-21	Jun-02		Aug-04	Jun-07	May-22	Jun-06	May-26	July 10 (2010)	May 22 (2013)
Common Wood-Nymph	Jul-18	Jun-16	Jun-25	Jun-14	Jun-18	Jun-13	Jun-19		July 2 (2011)	June 13 (2013)
Compton Tortoiseshell				Jul-12						July 12 (2011)
Coral Hairstreak		Jul-16						Jul-08	July 2 (2011)	July 2 (2011)
Crossline Skipper								Jul-15	July 2 (2011)	July 2 (2011)
Delaware Skipper		Jun-02	May-24	Jul-11	Jul-09	Jul-04	Jul-10	Jul-06	July 10 (2010)	June 2 (2009)
Dion Skipper					Jul-14				July 13 (2008)	July 13 (2008)
Dun Skipper		Jul-24		Jul-06	Jun-26	Jul-12	Jul-04	Jul-08	July 10 (2010)	June 26 (2012)
Eastern Comma	Aug-02	Jun-30	May-14	Jun-01	May-15	May-27	Jun-19	Jun-16	July 10 (2010)	May 14 (2010)
Eastern Pine Elfin								May-20		May 20 (2015)

Species	Earliest Record By Year							2015	Annual Butterfly Counts	Earliest Record at rare
	2006	2009	2010	2011	2012	2013	2014			
Eastern Tailed Blue	Aug-18			Jul-27	Jul-14	Jul-15	Aug-06	Jun-26	July 11 (2006)	July 11 (2006)
Eastern Tiger Swallowtail	Jul-18	May-21	May-19	Jun-01	May-14	May-22	Jun-06	May-21	July 2 (2011)	May 14 (2012)
European Skipper	Jul-18	Jun-24	May-24	Jun-14	May-15	May-30	Jun-10	May-19	July 2 (2011)	May 15 (2012)
Eyed Brown	Aug-02	Jul-16	Jun-15	Jul-05	Jun-08	Jun-25	Jun-20	Jun-16	July 2 (2011)	June 8 (2012)
Giant Swallowtail	Jul-24			Jun-08	May-15	May-30	Jun-10	May-27	July 11 (2006)	May 15 (2012)
Grey Comma								Jun-22	July 19 (2009)	July 19 (2009)
Great Spangled	Jul-18	Jul-24	Jun-21	Jul-11	Jun-18	Jul-02	Jun-25	Jun-29	July 10 (2010)	June 18 (2012)
Harvester				Aug-19			Jun-21	Jun-22		Aug 19 (2011)
Hickory Hairstreak	Jul-18								July 11 (2006)	July 11 (2006)
Hobomok Skipper			May-26	Jun-01	May-30	Jun-04	Jun-06	May-27	July 2 (2011)	May 26 (2010)
Inornate Ringlet	Aug-02	Jun-02	May-19	Jun-06	May-14	May-21	Jun-02	May-26	July 2 (2011)	May 14 (2012)
Juvenal's Duskywing			May-26	May-25	May-14	May-21	May-23	May-20		May 14 (2012)
Least Skipper				Aug-05	May-28	Jul-30	Jun-19	Jun-23	July 19 (2009)	May 28 (2012)
Little Glassywing				Jul-06	Jul-10	Jul-12	Jun-30	Jul-03	July 2 (2011)	July 2 (2011)
Little Wood-Satyr	Jul-18	Jun-10	Jun-03	Jun-08	May-30	Jun-04	Jun-10	May-28	July 2 (2011)	May 28 (2015)
Little Yellow									July 11 (2006)	July 11 (2006)
Long Dash				Jun-14	May-28	Jul-04	Jun-27	Jun-17	July 2 (2011)	May 28 (2012)
Meadow Fritillary					Jul-18		Jul-22		July 10 (2010)	July 10 (2010)
Milbert's Tortoiseshell			Jun-21	Jul-19	Jun-11		Jun-16	May-21		June 11 (2012)
Monarch	Jul-18	Jun-22	Jun-25	May-30	May-14	Jun-19	May-30	Jun-11	July 2 (2011)	May 14 (2012)
Mourning Cloak		May-25	May-04	Jun-07	May-14	May-21	May-24	Jun-22	July 10 (2010)	May 4 (2010)
Mulberry Wing									July 20 (2013)	July 20 (2013)
Mustard White				Aug-12						Aug 12 (2011)
Northern Broken-Dash					Jun-26		Jul-04	Jul-03	July 10 (2010)	June 26 (2012)
Northern Crescent		May-21	Jun-03	Jun-07	Jun-04	Jun-12	Jun-03	Jun-02	July 10 (2010)	May 21 (2009)
Northern Pearly-Eye	Jul-18	Jun-30	Jun-03	Jun-20	Jun-11	Jun-13	Jun-19	Jun-10	July 10 (2010)	June 3 (2010)
Orange Sulphur	Aug-24		Jun-30	Jul-19	May-14	Jun-04	May-30	Jun-02	July 10 (2010)	May 14 (2012)
Painted Lady		Jun-04	May-04		May-15	May-21	Jul-07	May-19		May 4 (2010)

Species	Earliest Record By Year							2015	Annual Butterfly Counts	First Record at *rare*
	2006	2009	2010	2011	2012	2013	2014			
Pearl Crescent	Jul-18			May-25	May-14	May-22	May-26	May-19	July 2 (2011)	May 14 (2012)
Peck's Skipper				Jul-11	Jun-18	Jul-06	Jul-03	Jun-22	July 2 (2011)	June 18 (2012)
Question Mark	Jul-18	Jun-10	May-19	Jun-07	May-17	Jun-14	Jul-21	May-28	July 10 (2010)	May 17 (2012)
Red Admiral	Aug-18	May-14	May-03	May-25	May-14	Jun-04	May-21	May-19	July 10 (2010)	May 3 (2010)
Red-Spotted Purple		Jun-16	Jun-01	Jun-14	May-25	Jun-04	Jun-19	Jun-02	July 10 (2010)	May 25 (2012)
Silver-Bordered						Jun-03		May-27	July 2 (2011)	May 27 (2015)
Silver-Spotted Skipper		Jul-30	Jun-08	Jun-20	Jun-25	Jun-13	Jun-13	Jun-11	July 10 (2010)	June 8 (2010)
Silvery Blue								Jun-02		June 2 (2015)
Silvery Checkerspot					Jun-20		Jul-18			June 20 (2012)
Spring Azure		May-13	May-04	May-20	May-15	May-21	May-21	May-20		May 4 (2010)
Striped Hairstreak				Jul-26		Jul-12	Jul-18	Jul-02	July 11 (2006)	July 02 (2015
Summer Azure	Aug-02	Jul-22	Jun-08	Jul-05	Jun-11	Jun-13	Jun-13	Jun-11	July 2 (2011)	June 8 (2010)
Tawny Emperor	Jul-21	Jul-30		Aug-04	Jul-17	Jul-25	Jul-28	Jul-16	July 10 (2010)	July 16 (2015)
Tawny-Edged Skipper		Jul-16		Jul-22		Jul-16	Jul-17	Jun-16	July 2 (2011)	June 16 (2015
Variegated Fritillary					Jul-05					July 5 (2012)
Viceroy	Aug-02	Jun-10	Jun-08	Jun-20	May-25	Jun-04	May-28	May-26	July 10 (2010)	May 25 (2012)
White Admiral		Jul-14		Jun-14	Aug-01		Aug-18	Jun-22	July 11 (2006)	June 14 (2011)
Wild Indigo Duskywing			May-17		Jul-11		Jul-28	May-28	July 2 (2011)	May 17 (2010)

Table D.2: Common and scientific names with shorthand appreviations of all salamander species observed at ***rare*** **Charitable Research Reserve** since 2006. The Eastern Red-backed salamander has two colour phases, red- and lead-backed, which are distinguished during sampling.

Common Name	Scientific Name	Abbreviation
Yellow-spotted Salamander	*Ambystoma maculatum*	YESA
Blue-spotted Salamander	*Ambystoma laterale*	BLSA
Four-toed Salamander	*Hemidactylium scutatum*	FOSA
Eastern Red-backed Salamander*	*Plethodon cinereus*	RESA/LESA

Table D.3: Common and scientific names with shorthand abbreviations of all tree species observed in forest canopy biodiversity monitoring plots at ***rare*** **Charitable Research Reserve** since 2009.

Common Name	Scientific Name	Abbreviation
American Beech	*Fagus grandifolia*	FAGUGRAN
Black Ash	*Fraxinus nigra*	FRAXNIGR
Black Cherry	*Prunus serotina*	PRUNSERO
Butternut	*Juglans cinerea*	JUGLCINE
Green Ash	*Fraxinus pennsilvanica*	FRAXPENN
Hophornbeam	*Ostrya virginiana*	OSTRVIRG
Red Maple	*Acer rubrum*	ACERRUBR
Red Oak	*Quercus rubra*	QUERRUBR
Sugar Maple	*Acer saccharum*	ACERSACC
White Ash	*Fraxinus americana*	FRAXAMER
White Oak	*Quercus alba*	QUERALBA
White Pine	*Pinus strobus*	PINUSTRO
Yellow Birch	*Betula alleghaniensis*	BETUALLE

APPENDIX E: Annual Butterfly Count Results

Note: 2007 and 2014 Annual Counts were cancelled due to inclement weather.

List E.1: Results from Annual Butterfly Count 2013.

An annual butterfly count for the North American Butterfly Association was held at ***rare*** on July 20, 2013. A total of 39 species and 429 individuals were observed. The annual butterfly count has occurred yearly on the ***rare*** property since 2006, with the exception of 2007. Results of the 2013 count can be found below and results from previous years can be found in Appendix D.

Black Swallowtail 7, E. Tiger Sw. 3, Cabbage White 104, Clouded Sulphur 39, Orange Su. 5, Acadian Hairstreak 1, Banded Ha. 1, E. Tailed-Blue 3, 'Summer' Spring Azure 2, Gr. Spangled Fritillary 6, Pearl Crescent 29, N. Cr. 4, Question Mark 1, Mourning Cloak 2, Red Admiral 2, Red-spotted Purple 3, Viceroy 5, Tawny Emperor 4, N. Pearly-eye 2, Eyed Brown 3, Little Wood-Satyr 15, Com. Wood-Nymph 111, Monarch 5, Silver-spotted Skipper 3, Wild Indigo Duskywing 9, European Sk. 2, Peck's Sk. 1, Tawny-edged Sk. 2, N. Broken-Dash 2, Little Glassywing 1, Delaware Sk. 16, Mulberry Wing 3, Hobomok Sk. 1, Broad-winged Sk. 5, Dion Sk. 2, Black Da. 8, Dun Sk. 14. **Unidentified:** skipper species 1, Polygonia species 2. **Total:** 39 species, 429 individuals. **Field Notes:** The previous evening to the count (July 19), the area experienced a large storm. Winds up to 200 mph, significant amounts of rain, thunder, lightning, etc. A good amount of damage was done to trees in the area.

List E.2: Results from Annual Butterfly Count 2012.

Cambridge (*rare* Charitable Research Reserve), ON. Yr. 6, 43.3817°, -80.355°, center at N of Blair Rd. about 1.7 mi E of jct. of Blair Rd. and Fountain St. in Cambridge. See 2006 report for habitats. Imminent threats to habitat: None. Habitat changes since last year: Researchers have planted one area previously which was active agriculture with tall grass prairie. This will be an improvement to habitat. **14 July 2012;** 0900-1500 hrs; sun AM 10%, PM 10%; 82-89°F; wind 2-2 mi/hr. 14 observers in 5 parties. **Total party-hours** 12; **total party-miles on foot** 9. **Observers:** J. Guenther, M. Hulme, S. Hulme, Jessica Linton (245 Rodney Street, Waterloo, ON, N2J 1G7; jlinton@nrsi.on.ca), A. MacNaughton, J. Quinn, G. Richardson, S. Shiplo, A. Turchin, E. Turchin, J. Turchin, B. Wilson, B. Woodman, E. Woodman.

Black Swallowtail 40, Giant Sw. 6, E. Tiger Sw. 18, Cabbage White 169, Clouded Sulphur 39, Orange Su. 29, E. Tailed-Blue 1, 'Summer' Spring Azure 1, Am. Snout 1, Variegated Fritillary 1, Gr. Spangled Fr. 3, Pearl Crescent 7, N. Cr. 2, Question Mark 1, Mourning Cloak 1, Am. Lady 1, Painted La. 4, Red Admiral 12, Com. Buckeye 1, Red-spotted Purple 4, Viceroy 8, Tawny Emperor 1, N. Pearly-eye 1, Eyed Brown 2, Appalachian Brown 5, Little Wood-Satyr 2, 'Inornate' Com. Ringlet 2, Com. Wood-Nymph 29, Monarch 61, Silver-spotted Skipper 3, Wild Indigo Duskywing 12, European Sk. 1, Peck's Sk. 1, N. Broken-Dash 2, Broad-winged Sk. 2, Dion Sk. 2, Black Da. 12, Dun Sk. 11. **Unidentified:** Skipper Species 3. **Total** 39 species, 501 individuals. **Immatures:** Black Sw. 15 eggs; Am. Snout 1 caterpillar. **Field Notes:** 2012 has been exceptionally dry and hot in southern Ontario.

List E.3: Results from Annual Butterfly Count 2011.

Cambridge (*rare* Charitable Research Reserve), ON. Yr. 5, 43.3817°, -80.355°, center at N of Blair Rd. about 1.7 mi E of jct. of Blair Rd. and Fountain St. in Cambridge. See 2006 report for habitats. **03 July 2011;** 0930-1530 hrs; sun AM 76-100%, PM 76-100%; 24-26°F; wind 7-34 mi/hr. 6 observers in 3 parties. **Total party-hours** 10; **total party-miles on foot** 7. **Observers:** E. Damstra, H. Dodds, B. Foell, Jessica Grealey (709 Keatswood Crescent, Waterloo, ON, N2T 2R6), P. Raspberry, G. Richardson.

E. Tiger Swallowtail 1, Cabbage White 95, Bronze Copper 4, Coral Hairstreak 2, Banded Ha. 3, 'Summer' Spring Azure 3, Silver-bordered Fritillary 2, Pearl Crescent 3, N. Cr. 26, Baltimore Checkerspot 12, Red-spotted Admiral 3, Viceroy 1, Tawny Emperor 2, N. Pearly-eye 13, Eyed Brown 62, Appalachian Brown 3, Little Wood-Satyr 13, Com. Ringlet 4, Com. Wood-Nymph 3, Monarch 10, Wild Indigo Duskywing 1, European Skipper 196, Peck's Sk. 2, Tawny-edged Sk. 5, Crossline Sk. 3, Long Dash 2, Little Glassywing 5, Hobomok Sk. 8. **Total** 28 species, 487 individuals.

List E.4: Results from Annual Butterfly Count 2010.

Cambridge (*rare* Charitable Research Reserve), ON. Yr. 4, 43.3817°, -80.355°, center at N of Blair Rd. about 1.7 mi E of jct. of Blair Rd. and Fountain St. in Cambridge. Floodplain; riparian; agricultural field and hedgerow; open meadow; wet meadow; forested; thicket; alvar; gravel trail; marsh. Habitat changes since last year: A large area has been seeded this year for a tall grass prairie restoration project. This will no doubt increase and improve butterfly habitat within the reserve. **10 July 2010;** 0930-1530 hrs; sun AM 76-100%, PM 76-100%; 68-83°F; wind 2-2 mi/hr. 19 observers in 6 parties. **Total party-hours** 25; **total party-miles on foot** 9. **Observers:** R. Beaubien, T. Beaubien, E. Damstra, S. Fogo, G. Grainge, Jessica Grealey (709 Keatswood Cresent, waterloo, ON, N2T 2R6; jgrealey@nrsi.on.ca), J. Grealey, K. Hodder, L. Lamb, A. MacNaughton, G. Michalenko, C. Moore, G. Richardson, B. Snider, E. Snider, E. Turchin, J. Turchin, W. Watson, M. Wolosinecky.

Black Swallowtail 27, E. Tiger Sw. 6, Cabbage White 187, Clouded Sulphur 93, Orange Su. 3, 'Summer' Spring Azure 2, Am. Snout 1, Gr. Spangled Fritillary 5, Meadow Fr. 1, Pearl Crescent 1, N. Cr. 2, Question Mark 8, E. Comma 2, Mourning Cloak 1, Am. Lady 5, Red Admiral 78, Red-spotted Purple 1, Viceroy 2, Tawny Emperor 4, N. Pearly-eye 18, Eyed Brown 7, Appalachian Brown 2, Little Wood-Satyr 8, Com. Wood-Nymph 73, Monarch 70, Silver-spotted Skipper 1, [1]**Wild Indigo Duskywing 9**, Com. Sootywing 1, Arctic Sk. 1, European Sk. 18, Peck's Sk. 1, Tawny-edged Sk. 6, N. Broken-Dash 1, Little Glassywing 2, Delaware Sk. 3, Broad-winged Sk. 1, [2]**Black Da. 24**, Dun Sk. 5. **Unidentified:** Polygonia sp. 3. **Total** 39 species, 683 individuals. **Field Notes:** [1]This species is widespread in Waterloo Region for the first time in 2010. Previously very rare. [2]Local population known from this area but uncommon in the Region of Waterloo.

List E.5: Results from Annual Butterfly Count 2009.

Cambridge (*rare* Charitable Research Reserve), ON. Yr. 3, 43°22.9'N, 80°21.3'W, center at N of Blair Rd. about 1.7 mi E of jct. of Blair Rd. and Fountain St. in Cambridge. Floodplain; agricultural; old field; cliffs & alvars; hedgerows; old growth forest; early successional; roadside. **19 July 2009;** 1030-1530 hrs; sun AM 11-25%, PM 11-25%; 64-70°F; wind 13-24 mi/hr. 16 observers in 5 parties. **Total party-hours** 24; **total party-miles on foot** 9. **Observers:** E. Damstra, G. Grainge, Jessica Grealey (709 Keatswood Cresent, WAterloo, ON, N2T 2R6), K. Hodder, L. Lamb, C. Moore, I. Moore, S. O'Neil, C. Pomeroy, G. Richardson, J. Shea, V. Slocombe, B. Snider, C. Snider, E. Snider, W. Watson.

Black Swallowtail 1, E. Tiger Sw. 1, Cabbage White 151, Clouded Sulphur 25, Orange Su. 3, Coral Hairstreak 1, Banded Ha. 8, Gr. Spangled Fritillary 4, Pearl Crescent 12, N. Cr. 2,

E. Comma 3, Gray Comma 1, Red Admiral 1, Red-spotted Admiral 1, Tawny Emperor 2, N. Pearly-eye 20, Eyed Brown 24, Appalachian Brown 11, Little Wood-Satyr 20, Com. Wood-Nymph 75, Monarch 11, Least Skipper 1, European Sk. 62, Peck's Sk. 1, Tawny-edged Sk. 2, Delaware Sk. 6, Broad-winged Sk. 1, Black Dash 1, Dun Sk. 12. **Total** 29 species, 463 individuals. **Field Notes:** Count originally scheduled for July 18th but was re-scheduled for the 19th. Conditions were not ideal (cool, overcast) but were consistent with the unusually cool and rainy weather experienced in southern Ontario this summer. On average, temperatures are 6 degrees Celsius cooler.

List E.6: Results from Annual Butterfly Count 2008.

Cambridge (*rare* Charitable Research Reserve), ON. Yr. 2, 43°22.9'N 80°21.3'W, center at center N of Blair Rd. about 1.7 mi E of jct. of Blair Rd. and Fountain St. in Cambridge. See 2006 report for habitats. Elevation: 928-928 ft. **13 July 2008**; 0930-1500 hrs; sun AM 76-100%, PM 51-75%; 15-28°F; wind 13-17 mi/hr. 14 observers in 5 parties. **Total party-hours** 6; **total party-miles on foot** 9. **Observers:** E. Barkley, M. Burrell, M. Cassidy, Jessica Grealey (709 Keatswood Cresent, Waterloo, ON N2T 2R6), S. Hentsch, C. Humphrey, K. Jackson, L. Lamb, G. Michalenko, M. Muir, G. Richardson, J. Turchin, M. Wolosinecky, L. Work.

Black Swallowtail 4, E. Tiger Sw. 19, Cabbage White 816, Clouded Sulphur 85, Orange Su. 10, Coral Hairstreak 15, Acadian Ha. 4, Banded Ha. 59, Hickory Ha. 1, Striped Ha. 20, E. Tailed-Blue 2, 'Summer' Spring Azure 2, Am. Snout 2, Gr. Spangled Fritillary 8, Meadow Fr. 2, Pearl Crescent 3, N. Cr. 12, Question Mark 2, E. Comma 1, Mourning Cloak 29, Am. Lady 4, Red Admiral 4, Red-spotted Admiral 12, Viceroy 1, Tawny Emperor 1, N. Pearly-eye 23, Eyed Brown 25, Appalachian Brown 3, Little Wood-Satyr 63, Com. Wood-Nymph 154, Monarch 14, Silver-spotted Skipper 2, European Sk. 127, Peck's Sk. 1, Tawny-edged Sk. 24, Long Dash 1, N. Broken-Da. 3, Delaware Sk. 15, Dion Sk. 2, Black Da. 6, Dun Sk. 8, Polygonia sp. 1. **Total** 42 species, 1,590 individuals. **Note**: Giant Swallowtail butterfly observed at Springbank garden during the summer of 2008

List E.7: Results from Annual Butterfly Count 2006**.**

Cambridge (rare Charitable Research Reserve), ON. Yr. 2, 43°22.9'N 80°21.3'W, center at center N of Blair Rd. about 1.7 mi E of jct. of Blair Rd. and Fountain St. in Cambridge. See 2006 report for habitats. Elevation: 928-928 ft. 11 July 2006; **Observers**: J. Grealey and L. Lamb.

Species Observed: Black dash, Broad-winged Skipper, Common Sootywing, Crossline Skipper, Delaware Skipper, Dun Skipper, European skipper, Northern Brokendash, Peck's Skipper, Tawny-edged Skipper, Banded Hairstreak, Eastern Tailed Blue, Hickory Hairstreak, Striped Hairstreak, Summer Azure, Appalachian Brown, Common Wood Nymph, Eastern Comma, Eyed Brown, Great Spangled Fritillary, Little Wood Satyr, Monarch, Mourning Cloak, Northern Cresent, Northern Pearly eye, Pearl Cresent, Question Mark, Red Admiral, Tawny Emperor,Viceroy, White Admiral, Eastern Tiger Swallowtail, Giant Swallowtail, Black Swallowtail, Cabbage White, Clouded Sulphur, Little Yellow, Long Dash. Total Species: 38.

APPENDIX F: 2015 Milkweed Survey

Full report and description of the protocol can be found on the ***rare*** server:

Z:\LEVEL4\RESEARCH & MONITORING\ECOLOGICAL MONITORING\MILKWEED MONITORING

Figure F.1: Map of ***rare*** property showing the four study sites delineated in red. The sites are as follows; 1 - Northern portion of Blair Flats, 2 - The Butterfly Meadow in Thompson Tract, 3 - The field adjacent to the Community Gardens, 4 – Field adjacent to ECO Centre.

Study Site	Potential Milkweed Area (ha)	Milkweed Stem Density (#individuals/m²)	Estimated Total Stems at Site
Blair Flats	1.91	0.25	4775
Thompson Tract	1.63	5	81500
Community Gardens	2.08	0.18	3744
ECO Centre	0.79	0.76	6004

Table F.1. Summary of total potential area milkweed was found, stem densities, and estimates of total number of Milkweed stems for each of the four study areas

APPENDIX G: Additional Data

Table G.1: Mean abundance and standard error for each species of salamander in each monitoring year.

	BLSA		FOSA		LESA		RESA		YESA	
	Mean Abundance	SE	Mean Abundance	SE	Mean Abundance	SE	Mean Abundance	SE	Mean Abundance	SE
2008	0	n/a	0.2	0	0.8	0.272	7.40	1.615	0.00	n/a
2009	0.22	0	0.22	0	3.11	0.597	11.77	1.075	0.44	0
2010	0.11	0	0	n/a	2.33	0.557	9.56	0.983	0.22	0
2011	0	n/a	0.11	0	2.55	0.850	7.67	1.391	0.33	0
2012	0.11	0	0	n/a	2.33	0.466	10.67	1.432	0.11	0
2013	0	n/a	0.22	0	4.22	0.508	18.44	2.126	0.78	0
2014	0.33	0.353	0.33	0.353	3	0.544	16.89	1.494	0.00	n/a
2015	0.11	0	0.44	0.272	2.44	0.552	12.56	1.067	0.11	0