Ecological Monitoring at ***rare*** Charitable Research Reserve 2010

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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 Cambridge, 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, monitoring programs established at ***rare*** include Plethodontid salamander monitoring, benthic invertebrate monitoring, butterfly monitoring, forest canopy tree biodiversity and health monitoring and soil humus decay rate monitoring. All of these programs are ongoing. In accordance with the monitoring schedule for the property, the monitoring conducted in 2010 included Plethodontid salamander monitoring, forest canopy tree biodiversity and health monitoring and annual soil humus decay rate monitoring.

Plethodontid Salamander Monitoring

The 2010 field season was the 4th year of monitoring at the Indian Woods salamander plot (which was previously monitored in 2006, 2008 and 2009) and the 3rd year of monitoring at the Hogsback salamander plot (which was previously monitored in 2008 and 2009). Each plot is composed of a series of wooden boards (32 in Indian Woods, 20 in the Hogsback) placed on the forest floor to act as artificial cover objects (ACO) for Plethodontid salamanders. Once weekly for nine weeks, the plots were monitored by turning over the ACOs and determining the species, snout-vent length, vent-tail length and weight of the salamanders underneath the boards. Environmental data including soil temperature, soil moisture, soil pH, air temperature, wind speed, relative humidity and precipitation from the previous 24 hours were also collected.

Eastern Red-backed salamanders (*Plethodon cinereus*) were the most abundant species in both Indian Woods and the Hogsback, with the lead-backed morph making up close to 20% of the *Plethodon cinereus* observations for both plots (IW=18.2%, HB=19.6%). In Indian Woods, mean weekly *Plethodon cinereus* abundance (measured as weekly catch per ACO) has declined every year since the start of monitoring, although the only significant decline was between 2008 and 2010. The pattern of salamander abundance in the Hogsback was not similar to that of Indian Woods: mean weekly *Plethodon cinereus* catch per ACO was highest in 2009, significantly greater than that of 2008.

To examine the factors influencing salamander abundance in the forest plots, a selection of soil parameters (including mean weekly soil temperature, mean weekly soil moisture, and mean soil pH) and temporal parameters (including the week and the year) were regressed on weekly salamander abundance (measured as catch per ACO). In Indian Woods, we did not detect any significant relationships between salamander abundance and the soil and temporal variables. However, in the Hogsback, the year, soil moisture and soil pH were each found to have a significant positive effect on the salamander abundance under the artificial cover objects.

Finally, the size-class distribution (measured as snout-vent length) of the salamanders observed under the artificial cover objects was analyzed for each plot. In 2010, the greatest proportion of Eastern Red-backed salamanders in Indian Woods fell within the upper snout-vent length size classes ranging from 35mm to 45mm, and the greatest proportion of salamanders in the

Hogsback plot fell within a larger range from 30mm to 45mm. Juvenile salamanders (less than 25mm snout-vent length) were underrepresented under the ACOs in both plots. In Indian Woods, there was a trend for increasing mean snout-vent length over the monitoring years, with mean salamander size in 2010 significantly exceeding that in 2006 and 2008. These findings could suggest that the same individual salamanders are returning to the ACOs each year, and we are consequently detecting the increasing size of this ageing co-hort. These findings may also indicate reduced recruitment of juvenile salamanders to the monitoring plot.

Forest Biodiversity Plot Monitoring

In 2009, permanent 20m x 20m Forest Biodiversity and Health Monitoring plots were established in Indian Woods (three plots) and the Cliffs and Alvars forest (three plots). In 2010, three additional forest plots were established in the Hogsback, and the complete monitoring data (including tree health assessments) for all nine plots was collected. During annual monitoring, the following information was collected for each tree: diameter at breast height, tree height, and tree condition (first classified as either alive or dead and then classified as standing, leaning, fallen, broken, or dead top). Tree health was monitored by recording stem defects, crown class (which indicates the level of dominance or suppression in the canopy), crown rating (which indicates the percent of crown dieback) and any other health notes.

The three plots within each forest were pooled for the calculations of the stand characteristics, and the diversity of each stand was calculated using Brillouin diversity and evenness measures. The Importance Value for each species monitored within the stand was calculated as the sum of that species' relative density, relative frequency and relative dominance in the plots. The Indian Woods plots had the lowest tree species diversity (Brillouin diversity measure= 0.631, evenness= 0.516) of the three forests, while the Hogsback plots were the most diverse (Brillouin diversity= 1.803, evenness= 0.891).The diversity of the Cliffs and Alvars forest plots was high (Brillouin diversity = 1.382, evenness = 0.797), with one plot containing an endangered Butternut tree (Juglans cinerea). Sugar Maple (*Acer saccharum*) and American Beech (*Fagus grandifolia*) were the most dominant tree species in each stand.

Between the 2009 and 2010 monitoring sessions, the Cliffs and Alvars forest plots experienced a single recruitment to the 10cm dbh size class (by an Ironwood, *Ostrya virginiana*) and no new mortalities. In the Indian Woods forest plots, there was one recruitment (by an American Beech, *Fagus grandifolia*) and three mortalities (two Red Maples, *Acer rubrum*, and one American Beech). The majority of the Ash trees observed in the forests plots showed signs of severe decline and protocols could be appended to the existing forest monitoring program to determine whether the Emerald Ash Borer is the cause.

Soil Humus Decay Rate Monitoring

Soil Humus Decay Rate Monitoring plots were established at the Cliffs and Alvars Forest Plot 1 in 2009 and additional plots were established at Indian Woods Forest Plot 1 and the Hogsback Forest Plot 1 in 2010. For each forest plot being monitored, three annual decay rate (ADR) monitoring plots were set up on each of the four plot corners. At each ADR plot, three pre-weighed tongue depressors were installed in the ground parallel to the soil surface at a depth of 5 cm, and one pre-weighed tongue depressor was positioned on the soil surface. The decay sticks were excavated and re-weighed one year after installation, and percent weight loss was calculated as a measure of decay rate.

For the decay sticks installed in the Cliffs and Alvars in 2009 and excavated intact in 2010, there was a significant mean mass loss of 29.7%. Many of the decay sticks that were excavated in 2010 were broken and missing pieces. We assume these pieces were lost due to physical disturbance and not decomposition and therefore broken decay sticks were not included in our analysis. Prior to installation in 2010, we placed each decay stick in a small nylon mesh bag in an effort to prevent stick pieces from being lost over the monitoring year. Hopefully this change to the protocol will increase the number of decay sticks that are excavated intact and included in the analysis in 2011.

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1.0 Introduction

1.1 Ecological Monitoring

Ecological monitoring measures changes in ecosystems over time through the regular observation and evaluation of organisms, populations and communities (Parks Canada 2009, McCarter 2009). It is impractical and often impossible to monitor every single species within an ecosystem, and ecological monitoring therefore relies on the study of a few carefully selected indicator species. These species are selected because they are convenient to study and because they are particularly sensitive to changes in their environment. Changes in indicator species abundance or population structure indicates change in the ecosystem in general and provides an early warning of environmental stress or ecosystem function decline (Parks Canada 2007).

Ecological responses to changes in the environment occur on longer time scales (i.e. decades, Vaughn *et* al. 2001) than most academic funding packages or political initiatives, and consequently there is a paucity of long-term monitoring datasets for ecosystems in Canada. The findings of long-term ecological monitoring programs are essential in determining priority issues for ecological management and stewardship, and play an important role in informing environmental regulations and policies (Environment Canada 2010).

1.2 Ecological Monitoring and Assessment Network

In 1994, Environment Canada established the Ecological Monitoring and Assessment Network (EMAN) as a Canada-wide network of ecological monitoring organizations, including various levels of government, academic institutions, private organizations and community groups (Environment Canada 2010). The objective of the EMAN Coordinating Office was to develop a series of standardized protocols for ecological monitoring, so that data collected by these diverse organizations could be easily compared or even compiled into meta-datasets. Since 1994, the EMAN coordinating office has developed a large variety of monitoring protocols for terrestrial, freshwater and marine ecosystems (Environment Canada 2010). All protocols are available without cost and, until recently, members of the network were encouraged to upload and share their monitoring data so that meta-analyses examining larger-scale patterns in ecosystem change could be conducted.

The EMAN coordinating office was closed in September 2010, and at present it is not clear as to whether the EMAN program will continue in any form. The monitoring protocols are still available online at the Environment Canada website, but it is no longer possible to access or upload monitoring data. According to Environment Canada (2010), some aspects of the work done by the EMAN coordinating office will now be handled by the Wildlife and Landscape Science Directorate in the Landscape Science and Technology Division.

1.3 Ecological Monitoring at *rare* Charitable Research Reserve

The ***rare*** Charitable Research Reserve is a not-for-profit environmental organization that preserves over 900 acres of land along the Grand River in the village of Blair in Cambridge, Ontario. The vision of ***rare*** is to offer the community a healthy natural area preserved intact in perpetuity. The ***rare*** lands are ecologically diverse, with cold-water streams, floodplains, alvars, old-growth forests, pine plantations and tall-grass prairies counted among the many habitat types on the property. The land use surrounding the ***rare*** property is likewise diverse; our neighbours include subdivisions, gravel extraction pits, busy roads, residential estates and conventional agricultural fields. In keeping with ***rare***'s vision for healthy lands intact in perpetuity, a number of ecological monitoring programs have been established on the property to determine the health of ***rare***'s ecosystems and to study the responses of these ecosystems to the environmental stresses presented by the changing world.

In 2006, ***rare*** joined the Ecological Monitoring and Assessment Network to establish a monitoring program including Plethodontid salamander monitoring in Indian Woods, benthic invertebrate monitoring in Bauman and Cruickston creeks, and butterfly monitoring in various locations on the property. Plethodontid salamander monitoring was expanded to the Hogsback forest in 2008. In 2009, both the butterfly and benthic invertebrate monitoring programs were expanded to include more sites, and forest canopy tree biodiversity plots were established in Indian Woods and the Cliffs and Alvars forests. Annual soil humus decay rate monitoring plots were established at the Cliffs and Alvars forest plot. In 2010, the forest canopy tree monitoring program was expanded to include the Hogsback forest, and annual soil humus decay rate monitoring began at the forest plots in Indian Woods and the Hogsback. The findings of the 2010 Plethodontid salamander monitoring, forest canopy tree biodiversity and health monitoring and annual soil humus decay rate monitoring will be discussed in this report.

2.0 Plethodontid Salamander Monitoring

2.1 Introduction

2.1.1 Plethodontid Salamanders

The salamander species that have been observed on the ***rare*** Charitable Research Reserve can be classified in two families: the mole salamanders (Ambystomatidae) and the lungless salamanders (Plethodontidae). Mole salamanders, like the Yellow-spotted salamanders (Ambystoma maculatum) and Blue-spotted salamanders (Ambystoma laterale) occasionally observed at ***rare***, are large burrowing salamanders that lay eggs in ephemeral ponds where the juveniles develop until the terrestrial adult phase moves into the soil of the forest floor (Conant and Collins 1998). Lungless salamanders observed at ***rare*** include the Eastern Red-backed salamander (*Plethodon cinereus*) and the Four-toed salamander (*Hemidactylium scutatum*). Salamanders in the family Plethodontidae are small and slender, characterized by their lack of lungs and their possession of chemoreceptor-lined naso-labial grooves used for hunting prey (Conant and Collins 1998). Plethodontidae is the largest salamander family in the world, with 27 genera representing 376 recognized species (Larson *et* al. 2006). Most members of the Plethodontidae family have both an aquatic juvenile stage and a terrestrial adult stage, although there are a number of species that exhibit only one the terrestrial or aquatic phases (Larson *et* al. 2006).

Plethodontid salamanders breathe solely by cutaneous respiration, relying on gas exchange across the moist surfaces of their skin and the roof of their mouth (Conant and Collins 1998). This feature of their biology makes them particularly sensitive to changes in the environment that may alter the air and water conditions of their micro-habitat (Zorn *et* al. 2004). Cutaneous gas exchange can only occur when skin is adequately moist (Welsh and Droege 2001), and the highly absorptive nature of their skin makes them susceptible to contaminants in the soil. Given this high sensitivity to their soil microhabitat, Plethodontid salamanders have been identified as suitable indicator species for the forest ecosystem (Zorn *et* al. 2004, Welsh and Droege 2001). Of the five species of Plethodontidae in Ontario, the Eastern Red-backed salamander (*Plethodon cinereus*) is the most common on the ***rare*** Charitable Research Reserve.

2.1.2 Monitoring *Plethodon cinereus*

Eastern Red-backed salamanders (*Plethodon cinereus*) are the most abundant Plethodontid in Eastern Canada (Zorn *et* al. 2004) and on rainy days, they are nearly ubiquitous under the rocks and logs of ***rare***'s old forests. As they are completely terrestrial, Eastern Red-backed salamanders do not require ponds or vernal pools for their development, and they are most often found in the moist soil under downed woody debris in mature forests (Conant and Collins 1998). There are two major colour variants of *Plethodon cinereus*; the red-backed morph has dark grey sides with a red, rough-edged stripe down its back, whereas the lead-backed morph lacks the red stripe and is completely grey. A number of life history characteristics of the Eastern Red-backed salamander make it an ideal species for monitoring the forest ecosystem.

Eastern Red-backed salamanders are very abundant in North America's temperate forests (Welsh and Droege 2001). In a study of Eastern red-back salamander abundance in the Hubbard Brook forest in New Hampshire, Burton and Likens (1975) estimated that the biomass of this species surpassed that of any other vertebrate group, and exceeded the biomass of all small mammals in the forest combined. Given this impressive abundance, the importance of this species in the nutrient cycling of the forest is clear. Eastern Red-backed salamanders are among the top predators of invertebrates in the soil and leaf litter, feeding on springtails, earthworms, ants and many other detritivores of the forest floor (Casper 2011). Plethodon cinereus in turn provides an ample food source for predators like snakes, rodents, rove beetles and birds, all while moving energy and nutrients into the higher trophic levels (Casper 2011, Zorn *et* al. 2004).

Given their low mortality and reproductive rates and their relatively long lifespan (which may reach nine years in some cases, LeClair *et* al. 2006), Eastern Red-backed salamanders typically have stable population sizes under normal conditions (Zorn *et* al. 2004). This is an essential characteristic for the focal species of a long-term monitoring program: the year-to-year fluctuations in population size are low or negligible for *Plethodon cinereus*, therefore large changes in abundance likely indicate changes to the ecosystem and not just normal population cycling (Welsh and Droege 2001, Zorn *et* al. 2004). Eastern Red-backed salamanders are also known to have small home ranges, and they often return to the same cover objects (logs, rocks, or study boards) season after season (Welsh and Droege 2001), so it can also be assumed that changes in salamander abundance are not being caused by shifting home ranges. Finally, Eastern Red-backed salamanders are known to readily use artificial cover objects added to the forest floor, allowing for simple, repeatable and non-destructive monitoring (Zorn *et* al. 2004). *Plethodon cinereus* is therefore a highly suitable study system for long-term monitoring of the forest ecosystem.

In 2006, a Plethodontid salamander monitoring program was established at the ***rare*** Charitable Research Reserve following the protocols developed by the Ecological Monitoring and Assessment Network (EMAN) and Parks Canada. The research questions that we hope to address with long-term salamander monitoring were identified by McCarter (2009):

1. What is the current state (species diversity, abundance, age structure) of the salamander populations in ***rare***'s 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?
    - an ecosystem with integrity is identified as having all of its native abiotic and biotic components and processes intact, and it is 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 when an ecosystem has the capacity to resist and recover from a range of disturbances and maintain its functions and processes (Styers *et* al. 2010, Twery and Gottschalk 1996).

2.1.3 EMAN Plethodontid Salamander Monitoring at *rare*

The Ecological Monitoring and Assessment Network and Parks Canada published a joint National Monitoring Protocol for Plethodontid Salamanders in 2004. This protocol involves the establishment of permanent forest monitoring plots containing a series of wooden artificial cover objects evenly spaced on the forest floor (Zorn *et* al. 2004). These plots are to be monitored in the spring or fall every year to detect changes in Plethodontid salamander abundance and community structure (Zorn *et* al. 2004) as an indicator of forest health.

The salamander monitoring program at the ***rare*** Charitable Research Reserve began in 2006 with the installation of 29 artificial cover objects in the Indian Woods monitoring plot. Monitoring was not conducted in 2007, but resumed in 2008 and has continued annually to 2010. In 2009, three additional artificial cover objects were installed to the Indian Woods plot, bringing the total to 32. In 2008, the Hogsback monitoring plot was established with 20 artificial cover objects, and monitoring has since been conducted annually.

The salamander monitoring program at ***rare*** has been successful to date in that salamanders started using the artificial cover objects within weeks of plot establishment and they continue to use the boards despite the disturbance inherent in the monitoring procedure. The monitoring data collected in these early monitoring years will provide valuable baseline data to which the data from future years can be compared in order to determine how ***rare***'s salamander populations are changing over time.

2.2 Methods

2.2.1 Monitoring Locations

Indian Woods (IW) is an old-growth Sugar Maple-American Beech dominated forest located on the western side of the ***rare*** property south of Blair Road. The forest is approximately 20 acres in area and contains trees as old as 240 years. The Indian Woods salamander monitoring plot is located on the east side of the ephemeral pond near the south end of the forest (Appendix A, Figure A.1). The plot is accessed by parking at the South Gate and walking north along the Grand Allee until a second path merges with it from the west (marked by a post with a blue square and white arrow), and then walking east into the forest toward the pond for approximately 100m. The 32 artificial cover objects (ACOs) of the monitoring plot are distributed in large square, with eight ACOs on each side. Each board is identified with a writeable aluminum tag and is marked for visibility on a nearby shrub with orange flagging tape. ACO boards 5, 6, and 7 were missing from the 2006 and 2008 monitoring years and they were added to the plot in 2009.

The Hogsback (HB) is a 57 acre forest found approximately 0.7km 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 and white pine. The Hogsback salamander plot is accessed by entering the South Gate and heading east along the lane to where it meets the Hogsback and then walking north and then east around the perimeter of the forest, finally heading south into the stand (over the fallen tree that lowers the fence) for 50m to the monitoring plot (Appendix A, Figure A.1). The Hogsback monitoring plot was established in 2008 and is comprised of 20 ACOs distributed in a large rectangle with eight ACOs on the long (north-south) sides.

2.2.2 Monitoring Protocol

One month prior to the start of monitoring, all ACOs in both Indian Woods and the Hogsback were visited to make sure that they were properly positioned and labelled. If necessary, the boards were re-positioned so that they were square to the plot and flush against the soil. Any holes in the board were packed with soil to prevent salamanders from hiding in them during monitoring.

Each plot was monitored by a minimum of two observers once a week for nine successive weeks from the end of August to the end of October. Indian Woods was monitored for only five weeks in 2006, and the Hogsback was monitored for five weeks in 2008.

At the beginning of each monitoring session, Beaufort's wind and sky codes (Appendix B, Tables B.1 and B.2) were recorded for the monitoring plot and the soil moisture meter (LIC: Lincoln Irrigation) was calibrated in pond water. To calibrate, the meter was adjusted with a screw driver so that it read a moisture rating of "10: saturated" when the probe was completely immersed in water. The precipitation for the 24 hours prior to monitoring (as reported by the Environment Canada Weather Office for the Region of Waterloo Airport) was recorded for each monitoring day. In Indian Woods, the pond depth was measured using the depth stick in the centre of the pond. The first 5cm of the stick are submerged in mud, so 5cm were subtracted from the measured depth to get the true water level. The start time of the monitoring session was recorded and each board was then visited in order. Soil temperature (in degrees Celsius) and moisture measurements were collected for each ACO by inserting the probes of the soil thermometer and the soil moisture meter to a depth of 10cm in the soil beside the board. The ACO board was then gently turned over and any salamanders underneath it were collected (by observers wearing nitrile gloves) and placed into a plastic container with a sponge dampened with pond water. Each salamander was identified to species (colour morph was indicated for *Plethodon cinereus*) and any noticeable physical defects were recorded. Each salamander was then placed on the clear lid of a small sandwich-sized plastic container, and the container (filled with dampened sponges) was then fitted on top of the lid, gently pressing the salamander between the lid and the sponges. With a bit of manipulation by the observer, the salamander was repositioned until its body and tail were straight enough for its snout-vent and vent-tail lengths to be measured through the lid using digital callipers. The salamander was then removed from the measuring container and weighed on a digital scale (Equal Digital Scale, model #23-D-50, capacity 50g, measures to 0.01g) and then released next to the board. If it was raining, a spring scale (Pesola, capacity 10g, measures to 0.1g) was used to prevent damage to the digital scale. Each salamander was placed in a clear, pre-weighed plastic bag suspended from the spring scale to measure its mass. The bags were dry to ensure accurate mass measurements, so this measurement was taken as quickly as possible to prevent desiccation. Any disturbances under or near the boards (e.g. snakes, ant nests, turkey scratches, or an ACO moved from its proper location) were also recorded.

Weather variables such as average wind speed (taken as the average after ten seconds), air temperature (degrees Celsius) and percent relative humidity were collected for a subsample of the ACOs called weather stations. Tables 2.1 and 2.2 provide a list of the boards represented by the measurements at the weather stations.

Soil samples for pH testing were collected on October 19, 2010 for Indian Woods and October 20, 2010 for the Hogsback. We collected three soil samples from a depth of 10cm from the ground near each of the ACO weather stations. The soil samples were placed in individual open deli containers and left to dry for one week prior to pH testing. A Hellige-Truog Soil Reaction (pH) tester kit was used to determine the pH for each sample. The pHs for the three samples from the same ACO were averaged to give a mean board pH.

Table 2.1 Weather stations and the artificial cover objects (ACOs) they represent in the Indian Woods salamander plot.

Weather Station ACO number	ACOs represented by the weather station
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 2.2 Weather stations and the artificial cover objects (ACOs) they represent in the Hogsback salamander plot.

Weather Station ACO number	ACOs represented by the weather station
2	1,2,3,4,5
7	6,7,8,9,10
12	11,12,13,14,15
17	16,17,18,19,20

2.2.3 Data analysis

All statistical analyses were performed either in Excel (Microsoft Office 2007) or R 2.11.1.

Salamander Abundance

Each salamander monitoring plot (IW or HB) was interpreted as representing a unique population, and each ACO within that plot was interpreted as representing a sample of that population. A number of boards in Indian Woods (ACOs 6-8) were missing in 2006 and 2008, meaning that fewer samples were taken at each monitoring session during those years. To correct for this inconsistency, abundance was transformed into "catch per unit effort" (CPUE) for each monitoring session, as is commonly used in fisheries science (Krebs 2001). To calculate the CPUE, the total salamander count for each monitoring day was divided by the number of ACOs in the plot to get the mean weekly catch per ACO. Only Eastern Red-backed salamanders (*Plethodon cinereus*) were included in the abundance comparison calculations.

As only five weeks of monitoring data were collected for both Indian Woods in 2006 and the Hogsback in 2008, only these five weeks (the last week of September to the last week of October) were compared between years. A T-test (paired by week) was used to determine whether any two years were significantly different in salamander abundance (measured as mean weekly catch per ACO). Because multiple comparisons were made, the p-values were corrected with a sequential Bonferroni adjustment (Rice 1989, α= 0.05).

Relationships between Salamander Abundance and Environmental Parameters

Data for a large number of environmental parameters were collected during monitoring, including soil variables like soil moisture, soil temperature and soil pH and climatic variables like air temperature, wind speed, and relative humidity. While we would ideally test for relationships between salamander abundance and each of these variables, the dataset is currently too small to provide the necessary degrees of freedom, and many of these variables would be highly correlated anyway. As such, a subset of variables was selected based upon our knowledge of salamander biology. Plethodontid salamanders live in the soil (Conant and Collins 1998), and soil parameters were therefore included as these are assumed to be the environmental conditions most relevant to salamander occurrence under the boards. Mean weekly soil temperature, mean weekly soil moisture and mean yearly soil pH were calculated for each plot by taking the average of the ACO values. These parameters were then included in a multiple linear regression on the mean weekly catch per ACO (CPUE) of the plot. Monitoring week was included in the regression, as ACO use by the salamanders is predicted to change across the season with changes in behaviour (i.e. mating, egg-guarding, hunting, and departure for hibernacula). Finally, the year was also included in the analysis to account for any yearly environmental changes beyond the soil parameters. By including each of these variables in the regression, we will be able to determine their relationship with salamander abundance independent of the effects of the other variables that are included in the regression. Only Eastern Red-backed salamanders (*Plethodon cinereus*) were included in this analysis. All weeks of monitoring data were included.

As the distributions of many of the parameters included in the regression were non-normal, the significance of the linear selection coefficients was determined non-parametrically with permutation testing in which the F-statistic distribution was calculated from 1000 iterations of the regression model in which the abundance variable (catch per ACO) was shuffled (Legendre and Legendre 1998).

Eastern Red-backed Salamander Size

Two measurements were collected for salamander size: snout-vent length and vent-tail length. As Eastern Red-backed salamanders are capable of dropping their tails (autotomy, Wise and Jaeger 1998), only snout-vent length was used to indicate salamander size. Salamander snout-vent length is known to a have a significant positive correlation with *Plethodon cinereus* age (for salamanders four years old or younger, LeClair *et* al. 2006), which allows us to estimate the approximate age structure of the population under the boards using size-class distribution. To test for differences in mean salamander snout-vent length between years, non-parametric independent 2-group Mann-Whitney U tests were used because many of the snout-vent length distributions were non-normal.

2.3 Results

2.3.1 Salamander Abundance

Figure 2.1 shows the total weekly salamander count for each monitoring year at Indian Woods. The only significant difference in Plethodontid salamander abundance after sequential Bonferroni correction for multiple comparisons was between 2010 and 2008 (t-stat= 5.023, p=0.0074); the mean catch per ACO in 2010 was nearly half that of 2008 (CPUE 2010=0.519, CPUE 2008=0.986).

Figure 2.2 shows the total weekly salamander count for each monitoring year at the Hogsback. Mean Plethodontid salamander abundance was highest in 2009, significantly greater than in 2008 (t-stat = 7.628, p=0.0016, CPUE 2008=0.41, CPUE 2009=0.81). Tables 2.3 and 2.4 summarize the mean weekly *Plethodon cinereus* catch per ACO for the two plots.

Figures 2.3 and 2.4 show the abundance for each salamander species by year for Indian Woods and the Hogsback respectively.

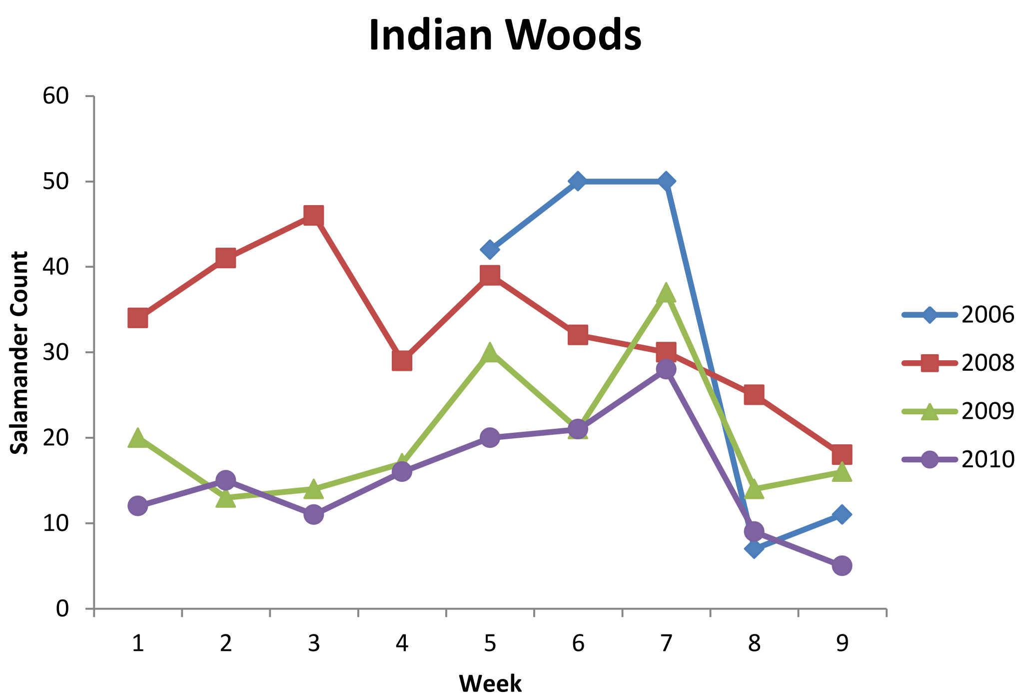


Figure 2.1 Total weekly salamander counts for each monitoring year at the Indian Woods monitoring plot.

Table 2.3 Mean weekly salamander catch per artificial cover object (ACO) and standard error for each monitoring year in Indian Woods. Values annotated with the same superscript were found to be significantly different at the p=0.05 level.

Year	2006	2008	2009	2010
Mean CPUE	1.097	0.986[a]	0.738	0.519[a]
S.E.	0.325	0.127	0.136	0.131

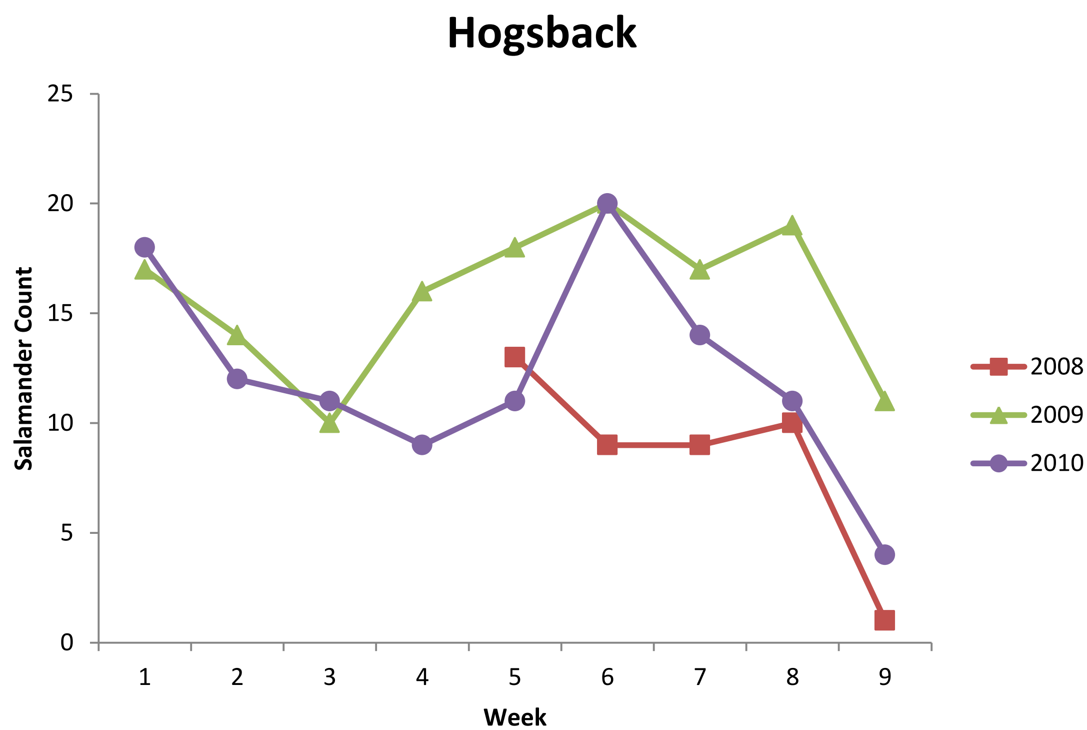


Figure 2.2 Total weekly salamander counts for each monitoring year at the Hogsback monitoring plot.

Table 2.4 Mean weekly salamander catch per artificial cover object (ACO) and standard error for each monitoring year in the Hogsback. Values annotated with the same superscript letter were found to be significantly different at the p=0.05 level.

Year	2008	2009	2010
Mean CPUE	0.410[a]	0.810[a]	0.600
S.E.	0.098	0.086	0.129

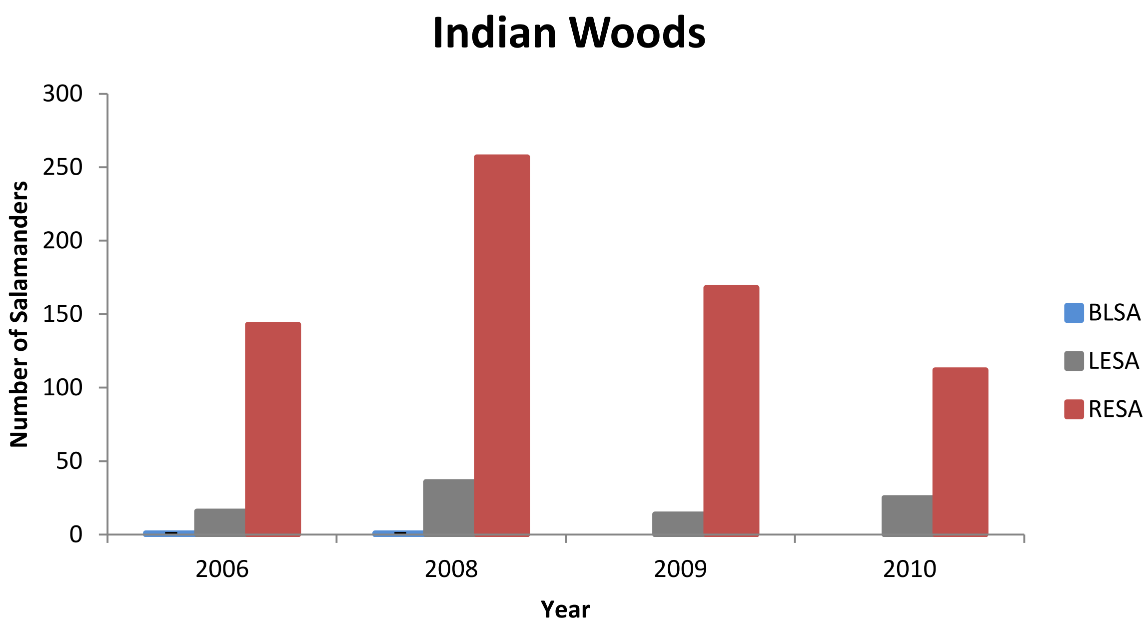


Figure 2.3 Salamander abundance by species for each monitoring year in Indian Woods. Species codes: BLSA = Blue-spotted salamander (*Ambystoma laterale*), LESA and RESA represent the lead-backed and red-backed forms of the Eastern Red-backed salamander (*Plethodon cinereus*).

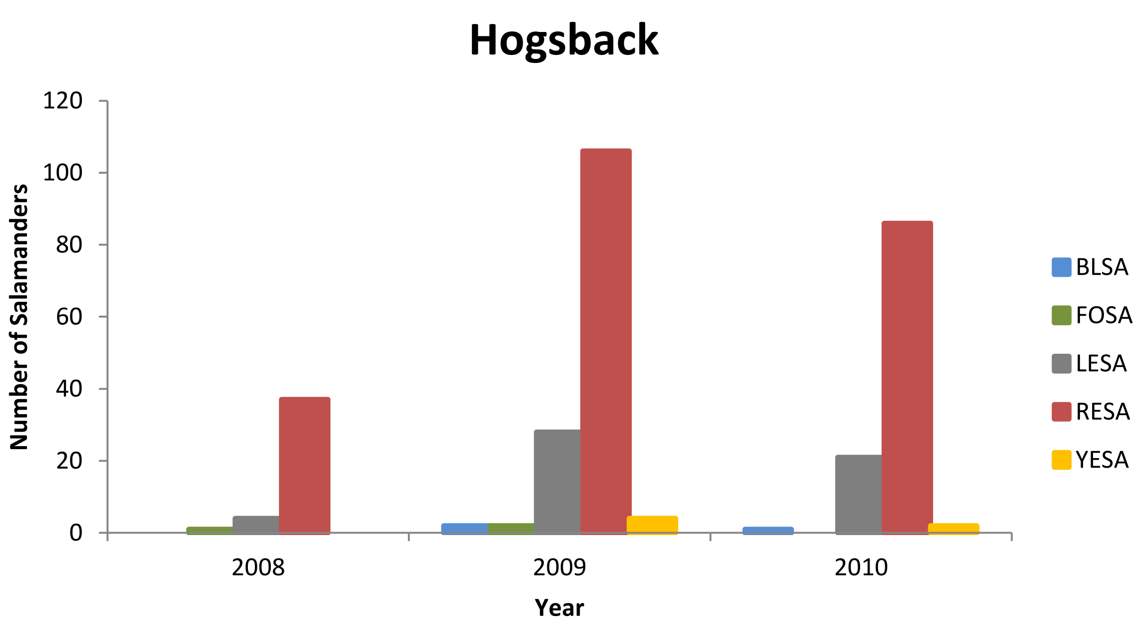


Figure 2.4 Salamander abundance by species for each monitoring year in the Hogsback. Species codes: BLSA = Blue-spotted salamander (*Ambystoma laterale*), FOSA= Four-toed salamander (*Hemidactylium scutatum*), LESA and RESA represent the lead-backed and red-backed forms of the Eastern Red-backed salamander (*Plethodon cinereus*), YESA= Yellow-spotted salamander (*Ambystoma maculatum*).

2.3.2 Relationships between Salamander Abundance and Environmental Parameters

Table 2.5 gives the selection coefficients (β) and their standard errors from the multiple linear regression of soil and temporal variables on salamander abundance (measured as mean weekly catch per ACO, CPUE) at the Indian Woods monitoring plot. For Indian Woods, none of the variables included in the analysis had a significant effect on the differences in salamander abundance observed between years, although there was a trend for a slight positive relationship between mean soil moisture and CPUE.

Table 2.6 gives the selection coefficients (β) and their standard errors from the multiple linear regression of soil and temporal variables on salamander abundance (measured as mean weekly catch per ACO, CPUE) at the Hogsback monitoring plot. Year, mean soil moisture and mean soil pH all show a significant positive effect on salamander abundance in the Hogsback.

Table 2.5 Selection coefficients for the multiple linear regression of temporal and soil parameters on weekly salamander abundance (measured as weekly catch per artificial cover object) for Indian Woods.

	β	**SE**	**P**
Year	-0.135	0.117	0.253
Week	-0.038	0.040	0.334
Mean soil temperature	-0.009	0.025	0.753
Mean soil moisture	0.103	0.052	0.066
Mean soil pH	2.090	2.223	0.346

Table 2.6 Selection coefficients for the multiple linear regression of temporal and soil parameters on weekly salamander abundance (measured as weekly catch per artificial cover object) for the Hogsback.

	β	**SE**	**P**
Year	0.395	0.134	0.007*
Week	-0.062	0.033	0.075
Mean soil temperature	-0.013	0.023	0.575
Mean soil moisture	0.104	0.047	0.035*
Mean soil pH	0.821	0.277	0.007*

2.3.3 Eastern Red-backed Salamander Size

Figures 2.5 and 2.6 show the snout-vent size class distributions for Eastern Red-backed salamanders for each of the monitoring years in Indian Woods and the Hogsback respectively. For Indian Woods, the mean snout-vent length in 2010 was significantly greater than that in both 2006 ($U=7352.5$, $p=6.045 \times 10^{-6}$) and 2008 ($U=14885.5$, $p=0.00017$), and the mean snout-vent length in 2009 was significantly greater than that of 2006 ($U=10937.5$, $p=0.00082$) (Table 2.7). There were no significant differences in mean snout-vent length between years in the Hogsback monitoring plot (Table 2.8).

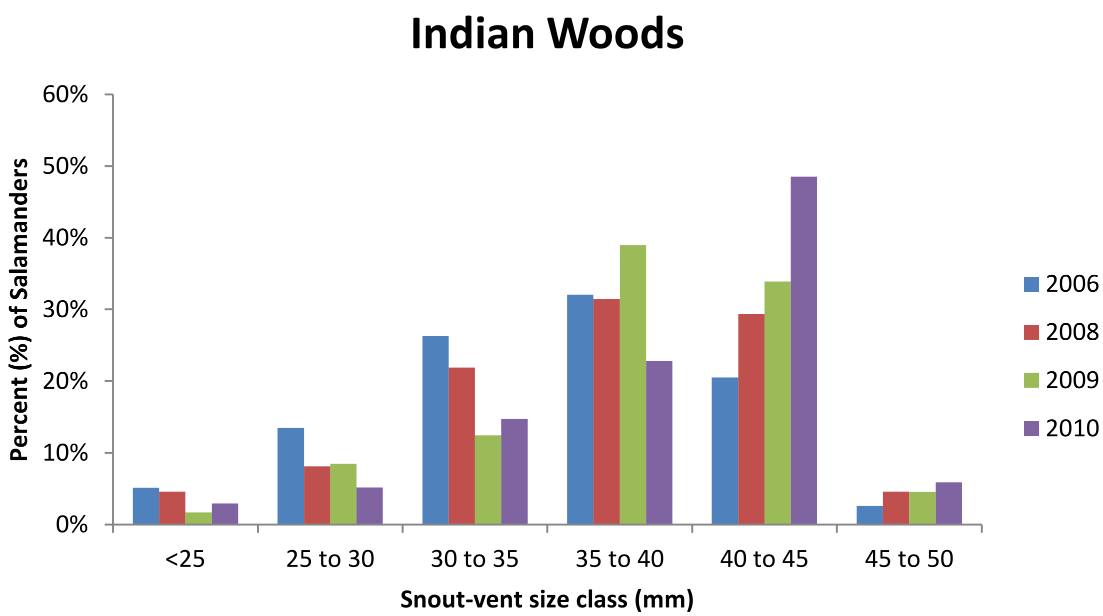


Figure 2.5 The percentage of Eastern Red-backed salamanders found in each snout-vent size class in Indian Woods for each monitoring year.

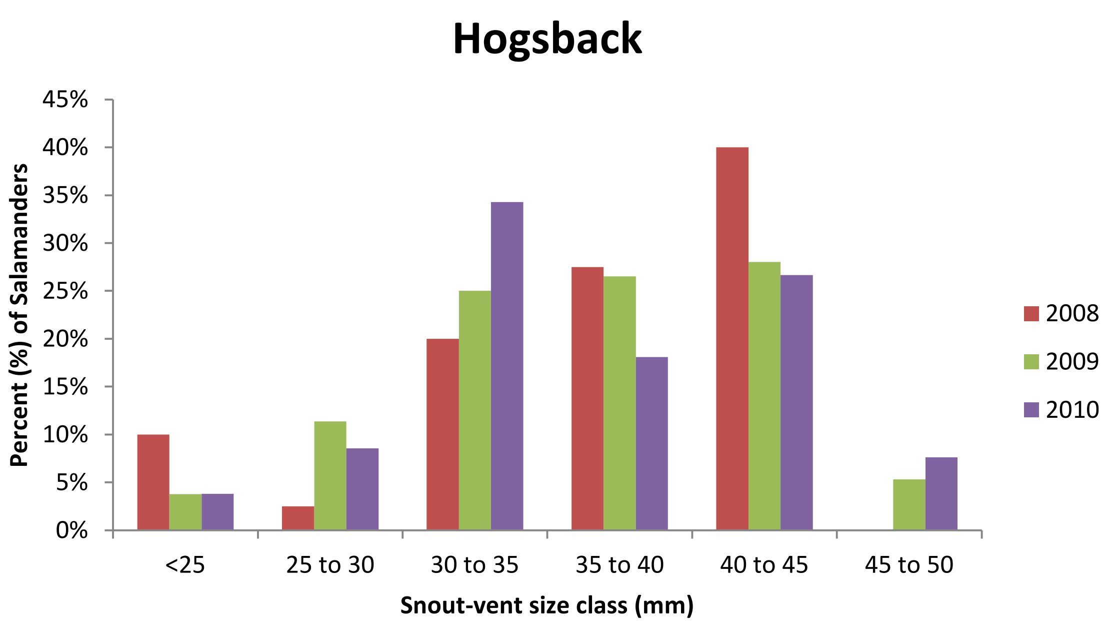


Figure 2.6 The percentage of Eastern Red-backed salamanders found in each snout-vent size class in the Hogsback for each monitoring year.

Table 2.7 Mean snout-vent length (SVL) (mm) and standard error of Eastern Red-backed salamanders from four monitoring years in Indian Woods. Values annotated with the same superscript letter were found to be significantly different at the p=0.05 level.

Year	2006	2008	2009	2010
Mean SVL	35.827^{ab}	36.466^{c}	38.118^{b}	38.486^{ac}
S.E.	0.492	0.382	0.501	0.519

Table 2.8 Mean snout-vent length (SVL) (mm) and standard error of Eastern Red-backed salamanders from three monitoring years in the Hogsback. Values annotated with the same superscript letter were found to be significantly different at the p=0.05 level.

Year	2008	2009	2010
Mean SVL	36.237	36.232	36.125
S.E.	1.106	0.582	0.631

2.4 Discussion

2.4.1 Salamander Abundance

The primary objective of Plethodontid monitoring at ***rare*** is to detect changes in the salamander populations, which in turn indicate changes in the forest ecosystem in general (Zorn *et* al. 2004). While many valuable ecological questions can be asked and answered with this monitoring data, we are most interested in knowing whether salamander abundance at the two forest monitoring plots is changing significantly over time. In the EMAN protocol, Zorn *et* al. (2004) suggest setting the monitoring thresholds at "a statistically significant change in Plethodontid counts at plot level over five or more years". The 2010 monitoring season constituted the 4th monitoring year at the Indian Woods forest (but 5th year since monitoring began – no monitoring was done in 2007) and the 3rd monitoring year in the Hogsback forest. While monitoring commenced immediately after the ACOs were established in both Indian Woods and the Hogsback, Zorn *et* al. (2004) suggest that the ACO boards weather in situ for a winter prior to monitoring because the disturbance of plot establishment may skew abundance estimates. Therefore, following the EMAN protocol, we will not be able to test whether the five year monitoring threshold has been surpassed until 2012 in Indian Woods and 2013 in the Hogsback. We can, however, compare the yearly salamander abundance data collected so far, keeping in mind that we may be detecting annual fluctuations instead of long-term population trends.

In Indian Woods, mean weekly salamander abundance (measured as weekly catch per ACO) has declined every year since the start of monitoring (Table 2.3), although the only significant decline was between 2008 and 2010. These findings reject our predictions that the first year of monitoring (2006) would have the lowest salamander abundance due to the disruptions caused by plot establishment and the time lag for the salamanders to begin using the ACO boards. This may suggest that competition for natural cover objects is sufficiently high to drive salamanders to use the ACOs despite the unfamiliarity of the boards and the recent disturbances in the area. While the trend of declining abundance is concerning, the year to year variation is fairly high and it may be too soon to tell if a significant biological change is taking place.

The pattern of salamander abundance in the Hogsback was not similar to that of Indian Woods: mean weekly *Plethodon cinereus* catch per ACO was highest in 2009, significantly greater than that of 2008, the year that the plot was established. The fact that two forests as close as 600m are demonstrating different trends could imply that the factors influencing Eastern Red-backed salamander population abundances are extremely local. In fact, Welsh and Droege (2001) suggest that Plethodontid salamanders are ideal for monitoring forest health because of their elevated sensitivity to the within-stand microclimatic conditions of the forest floor (like soil moisture and temperature) created by fine-scale characteristics of the stand such as canopy layering and gaps, soil type, and the quantity and type of downed woody debris and leaf litter.

2.4.2 Species Diversity

While the EMAN salamander protocol was designed for monitoring the Plethodontidae family of salamanders (Zorn *et* al. 2004), a variety of salamander species have been encountered over the monitoring years. Eastern Red-backed salamanders (*Plethodon cinereus*) were by far the most abundant species in both Indian Woods and the Hogsback, with the lead-backed morph making up close to 20% of the observations for both plots (IW=18.2%, HB=19.6%). Salamander species diversity was particularly low in Indian Woods, where there have only been two non-*Plethodon cinereus* observations over the four monitoring years (Blue-spotted salamanders (*Ambystoma laterale*), recorded in 2006 and 2008, Figure 2.3).

The Hogsback monitoring plot was similarly dominated by Eastern Red-backed salamanders, although the diversity of other salamander species observed was much greater than that of Indian Woods (Figure 2.4). Four-toed salamanders (*Hemidactylium scutatum*) were observed in both 2008 and 2009. This species belongs to the same family as the Eastern Red-backed salamander (lungless salamanders: Plethodontidae) and is usually associated with sphagnum moss or boggy woodlands (Conant and Collins 1998), the latter of which is found in the Hogsback forest. A number of mole salamanders belonging to the family Ambystomatidae have been observed in the Hogsback, with both Blue-spotted (*Ambystoma laterale*) and Yellow-spotted (*Ambystoma maculatum*) salamanders observed in both 2009 and 2010. It is possible that a number of the yellow-spotted salamander observations from 2009 and 2010 were from the same individual, as one was observed regularly under the same board and was reasonably consistent in size (with a snout-vent length ranging from 67.59mm to 80.70mm) and weight (ranging from 15.09g to 17.8g). This may suggest that the artificial cover objects are suitable habitat for mole salamanders as well as Plethodontid salamanders, and that some salamanders may exhibit board fidelity from year to year.

2.4.3 Relationships between Salamander Abundance and Environmental Parameters

In our analysis of salamander abundance of Indian Woods, we found no significant relationships between the weekly salamander abundance (catch per ACO) and the temporal (year and week) and soil parameters (soil moisture, soil temperature and soil pH) (Table 2.5). However, in the Hogsback forest, the year, mean weekly soil moisture and mean yearly soil pH all had a significant positive effect on salamander abundance in the plot (Table 2.6). Superficially, these results appear to contradict the findings of ***rare***'s 2009 salamander monitoring report which described a significant positive effect of soil temperature and no effect of soil moisture on the salamander abundance of Indian Woods (McCarter 2009), although a number of factors can account for the differing results. In the 2009 analysis, the data was organized by board: the mean yearly abundance, soil temperature and soil moisture was calculated for each ACO in the plot. In the 2010 analysis, the data was organized by plot: the soil moisture and temperature values from each ACO were averaged across the plot for each monitoring session (week) and these plot-wide mean weekly values were regressed onto the salamander catch per ACO. While it would be interesting to know the fine-scale ACO level habitat choices made by the salamanders, the dataset does not have sufficient degrees of freedom to adequately address the pseudoreplication inherent in treating each ACO individually. Pseudoreplication occurs when observations that are related to one another to differing degrees are all treated as being independent; for example, pooling together the mean yearly observations for each ACO for all monitoring years fails to address the fact that some observations represent the same ACOs, an error leading to incorrect estimates of statistical significance (Crawley 2005). The EMAN protocol was designed with the assumption that the monitoring plot represents a single population, and the ACOs represent sampling points within this population, not individual populations themselves (Zorn *et* al. 2004), therefore averaging the ACO values to give a mean plot value is appropriate.

The temporal variables "Year" and "Week" were included in the analysis in 2010, in part to address some of the autocorrelation (i.e. temporal pseudoreplication caused by weeks of the same year being treated as if they are equally related to weeks of different years) in the data, but also to determine if there are any temporal effects beyond the soil parameters that may influence salamander abundance. Interestingly, year had a significant positive effect on salamander catch per ACO in the Hogsback. This means that there is some factor beyond soil moisture, temperature and pH that correlates with year and influences salamander abundance. Plethodontid salamanders typically have high population stability (Welsh and Droege 2001, Zorn *et* al. 2004), but it is possible that some form of population cycling could account for the observed effect of year on abundance. As abundant predators of soil invertebrates (Casper 2011), Eastern Red-backed salamanders are capable of significantly reducing soil detritivore numbers (Wyman 1998), which suggests that predator-prey cycling could occur.

The relationship between abundance and soil pH has been well studied in *Plethodon cinereus*: Wyman and Hawksley-Lescault (1987) determined that salamanders would avoid soil of pH less than 3.7, and Heatwole (1962) reported that their preferred range of soil pH was 6.0 to 6.8. In our analysis of the Hogsback, we found a significant correlation between salamander abundance and soil pH, with pH values in the plot ranging from 6.5 to 7.0 over the course of the study. Wyman and Hawksley-Lescault (1987) suggest that the salamanders may serve as "canaries in the coal mine" should any significant changes occur in the soil pH of the forest, like, for example, soil acidification resulting from acid rain. In an experimental study manipulating soil pH, soil moisture and light intensity, Sugalski and Claussen (1997) found that *Plethodon cinereus* distribution was most affected by pH, even though inadequate soil moisture can be immediately lethal to the salamanders. A number of studies have detected a similar positive relationship between soil moisture and salamander abundance to that seen in the Hogsback (Heatwole and Lim 1961, Francl *et* al. 2010). Heatwole (1962) estimated that Eastern Red-backed salamanders cannot tolerate soil with interstitial humidity less than 85%. It is interesting to note that this relationship was not detected in Indian Woods. Mean soil temperature was significantly higher in the Hogsback than in Indian Woods (t-stat=4.917, $p=1.3\times10^{-5}$), but it is possible that the soil moisture regime in Indian Woods is more consistent within the salamander's ideal range.

2.4.4 Eastern Red-backed Salamander Size

The greatest proportion of Eastern Red-backed salamanders in Indian Woods fell within the upper snout-vent length size classes ranging from 35mm to 45mm, and the greatest proportion in the Hogsback fell in a larger range from 30mm to 45mm. Using skeletochronology (age estimation from long bone growth rings), LeClair *et* al. (2006) calculated the mean snout-vent lengths of *Plethodon cinereus* salamanders aged 0 (neonates) to seven years in the Mastigouche Reserve in Quebec, and they found a significant positive correlation between salamander size and age, although the strength of this correlation declined once the salamanders reached four years and their growth began to slow. Assuming that the *Plethodon cinereus* at ***rare*** have similar growth rates to those in Mastigouche, then the large majority of salamanders found under the ACOs in Indian Woods and the Hogsback would be adults aged three years and older. Individuals with snout-vent lengths less than 15mm were likely neonates (LeClair *et* al. 2006; Zorn *et* al. (2004) classify individuals with S-V lengths less than25mm as juveniles), an age demographic that appears to be underrepresented by ACO sampling. Marsh and Goicochea (2003) propose a number of possible reasons for the low proportion of juveniles under artificial cover objects compared to natural cover objects: 1. adults may be better dispersers and territory defenders, so they are able to reach and secure the new cover objects more quickly than juveniles; 2. larger salamanders may prefer the wider cover provided by the artificial cover objects; 3. reproductive success may be lower under artificial cover boards than natural cover objects and therefore there are fewer hatchlings and juveniles under the new boards. While these findings indicate that ACO sampling method does not provide a complete representation of all age demographics of the population of *Plethodon cinereus* in the ***rare*** forests, the data obtained from the monitoring is still valuable for within-site, between-year comparisons.

For example, in Indian Woods, there was a trend for increasing mean snout-vent length over the monitoring years (Table 2.7). These findings could suggest that the same individual salamanders are returning to the boards each year, and we are detecting the increasing size of the co-hort. Upon emergence from their hibernacula, salamanders that formerly occupied an ACO would be more familiar with its location, and may therefore be able secure it as a territory faster than new salamanders. However, a mark-recapture study of Eastern Red-backed salamanders by Monti *et* al. (2000) found both recapture rates and ACO fidelity to be low, and this trend of increasing snout-vent length was not observed in the Hogsback. Parker (2003) detected a similar trend for salamander size increasing over the monitoring years at the Long Point World Biosphere Reserve. This trend could be examined further using mark-recapture procedures.

2.4.5 Conclusion

As of 2010, four years of monitoring data have been collected for the Indian Woods salamander monitoring plot and three years of data have been collected for the Hogsback monitoring plot. Indian Woods shows a trend of declining salamander abundance over time, whereas salamander abundance in the Hogsback was highest in 2009. Our monitoring threshold will be surpassed if there is a significant decline in salamander abundance after five years of monitoring, and further investigations and possible management actions will be required should that occur.

The majority of Eastern Red-backed salamanders detected under the artificial cover objects in both plots were adults, as estimated using snout-vent length. This indicates that the cover board monitoring method does not produce a sample representative of the entire *Plethodon cinereus* population.

Eastern Red-backed salamander abundance in the Hogsback was found to have a significant positive relationship with year, soil moisture, and soil pH, whereas there were no significant relationships between abundance and the temporal and soil parameters in Indian Woods.

3.0 Forest Canopy Tree Biodiversity Monitoring

3.1 Introduction

3.1.1 Forest Tree Biodiversity

Before the time of European settlement, southern Ontario was largely covered by a patchwork of deciduous and mixed hardwood forests at differing seral stages (Ontario Ministry of Natural Resources 1999). These large tracts of forest were promptly destroyed after settlement for timber and to clear land for farming, and the rapid development of southern Ontario left very few undisturbed remnant old-growth forests in its wake (Ontario Ministry of Natural Resources 1999). The ***rare*** Charitable Research Reserve is fortunate enough to have one such remnant old-growth forest on the property, in the form of Indian Woods, a Sugar Maple-American Beech dominated forest with trees as old as 240 years. The forests at ***rare*** also include the Cliffs and Alvars forest, a mixed deciduous stand 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 rare habitat to countless plants and animals that require mature forest interior (Ontario Ministry of Natural Resources 1999).

These forests face diverse challenges in the landscape of Waterloo Region; ***rare*** is bordered by conventional farm fields, aggregate mining operations, subdivisions and busy roads, and many of these neighbouring lands are scheduled for drastic changes and development. By taking stock of the current conditions of the ***rare*** forests and monitoring them long-term, we may be able to determine how our forests respond to their changing conditions and predict what our forests will be like in the future.

The research questions that we hope to address with long-term forest canopy tree monitoring were identified at the establishment of the program (McCarter 2009):

1. What is the current state (biodiversity, composition, health) of ***rare***'s forests, and how to 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?
    - an ecosystem with integrity is identified as having all of its native abiotic and biotic components and processes intact and it is likely to persist (Parks Canada 2009).
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)?
    - ecosystem health is defined as when an ecosystem has the capacity to resist and recover from a range of disturbances and maintain its functions and processes (Styers *et* al. 2010, Twery and Gottschalk 1996).

3.1.2 EMAN Forest Monitoring at rare

Environment Canada's Ecological Monitoring and Assessment Network published a series of Terrestrial Vegetation Monitoring Protocols in 1999, which included protocols for monitoring the biodiversity of the canopy-tree stratum, the shrub and small-tree stratum and the ground vegetation stratum (Roberts-Pichette and Gillespie 1999). Each of these protocols involves the establishment of permanent plots to be monitored repeatedly over many years to detect changes in species abundance, richness and community structure for forest vegetation (Roberts-Pichette and Gillespie 1999).

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 forest and three plots in Indian Woods. Preliminary monitoring data, such as tree species, location 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 wooded areas on the ***rare*** property would be represented in the monitoring program. All nine forest plots were completely monitored in 2010 and an EMAN Tree Health Protocol was added to the monitoring program. This protocol simply extends the tree-by-tree inspection to include stem defects, canopy dieback and dominance (EMAN 2004).

3.2 Methods

3.2.1 Forest Plot Locations

The Cliffs and Alvars (CA) woods is 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 20m north of the Grand Trunk Trail, arranged parallel to the trail (Appendix A, Figure A.1).

Indian Woods is a rare remnant of old-growth forest located south of Blair 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 Allee. The third plot can be accessed by turning east into the forest off the Grand Allee towards the salamander monitoring plot and continuing to the top of the hill overlooking the pond. The second and first plots can then be found by heading north from the third plot (Appendix A, Figure A.1). The plots are approximately 30m apart and the flagging tape on the corners of each plot should be visible from the adjacent plot.

The Hogsback is located at the south-west corner of the property, bisected by Cruickston Creek and bordered by the Newman Drive subdivision on 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 and can be reached by following the directions given in section 2.2.1. 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 (and separated by a small boggy area) (Appendix A, Figure A.1).

3.2.2 Monitoring Protocol

Plot Establishment

In accordance with the EMAN Forest Canopy Tree Biodiversity Monitoring Protocol, the forest monitoring sites established in 2009 and 2010 are 20m x 20m permanent plots located in the forest interior. EMAN suggests that the plot should not be closer than three times the average tree height to any forest edges (estimated at 90m-100m for our forests), but because of the small size of Indian Woods and swampy topography of the Hogsback, this was not always possible and the plots were simply established as far from any edges as possible. The plots were oriented along the cardinal directions and the corners were marked with galvanized steel pigtails with labelled with flagging tape (see Figure 3.1 for a diagram of plot labels). All trees within the plot with a diameter equal to or greater than 10cm at breast height (dbh) (equal to a circumference of 31.4) were given unique identification codes and included in the monitoring. For example, a tag labelled with “IW-02-09” indicates tree number nine in Indian Woods forest plot 2.

The trees were tagged in a clockwise spiral inward from the northwest corner of the plot. Trees in the Cliffs and Alvars plots were labelled with pre-printed aluminum forestry tags and fixed to the trees with downward angled nails. Forest plots in Indian Woods and the Hogsback were labelled with pre-printed aluminum tags on galvanized steel pigtails inserted into the ground at the base of the tree. 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 of the plot. The line number was recorded, and the “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. Trees with multiple stems at breast height were labelled with letters as well as their unique tree ID, with the largest stem labelled with “a” (i.e. CA-01-13a, CA-01-13b and CA-01-13c represent three stems of the same tree).

Annual Monitoring Procedure

At each annual monitoring session the following variables should be recorded for each tree: dbh (calculated from circumference), tree height (obtained using a clinometer (Suunto Helsinki)), and tree condition (first classified as either alive or dead and then as standing, leaning, fallen, broken, dead top). Tree health was monitored by recording stem defects, crown class (indicates level of dominance or suppression in the canopy), crown rating (indicates percent of crown dieback) and any other health notes (e.g. leaf damage, woodpecker excavations). During each monitoring session, any larger untagged trees should be checked to see if they have graduated into the 10cm dbh size class. If so, they should be tagged in a manner consistent with their plot and measured into the plot using the “A” and “B” distances previously described.

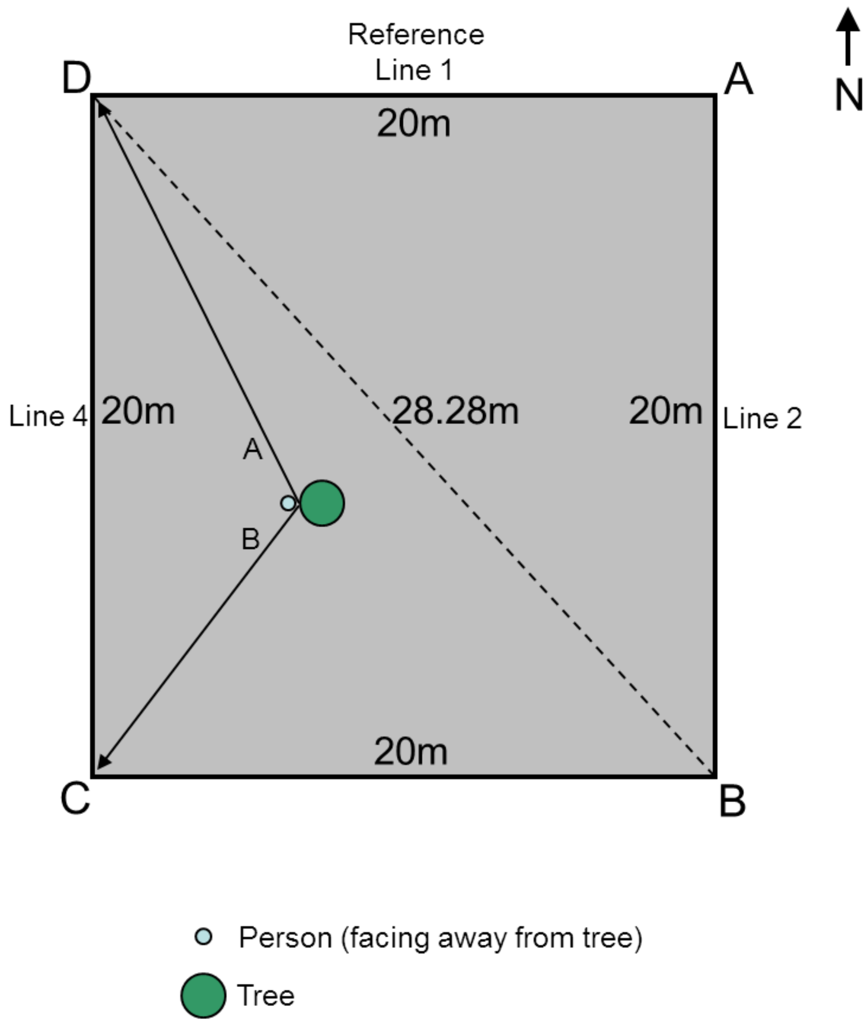


Figure 3.1 Diagram of an EMAN forest canopy tree biodiversity plot (from McCarter 2009). The A and B distances are used to map the position of the tree within the plot. The A distance is measured from the tree to the corner to the right of an observer standing facing the reference line. The B distance is measured to the corner on the left side of the observer.

3.2.3 Data Analysis

For each forest (CA, IW and HB), summary statistics were calculated by combining the data for the three plots, as they all represent the same stand. For each forest, we recorded the number of tree families present, the number of species present, the number of trees and stems present, the mean diameter at breast height (cm) for the stems included in the plots (i.e. stems over 10cm dbh), and the total basal area (m^2ha^{-1}) for the three plots combined. Basal area was calculated as the cross sectional area of all tagged tree stems in the plot and was determined using the diameter at breast height data. Species diversity and evenness were calculated using Brillouin diversity measure, which is intended for use when the randomness of the sample is suspect (Krebs 2001) or for collections where the full composition of the community is known, as is the case for the completely censused forest plots. Because it describes a known collection, the Brillouin diversity index is an exact measure (with no statistical uncertainty), and it is a composite measure of both the number of species present in the plot and their relative abundances (Van Dyke 2008). The Brillouin evenness measure increases as the number of species increases and as the species become more equal in their abundances (Magurran 2004).

Brillouin Index:

$$HB = \frac{\log(N!) - \sum \log(n_i!)}{N}$$

where n_i = # of individuals in a species and N = total # of individuals

Brillouin Evenness:

$$E = HB/HB_{max} \quad \text{where} \quad HB_{max} = \frac{1}{N} \times \ln \frac{N!}{\{[N/S]!\}^{S-r} \times \{([N/S]+1)!\}^{r}}$$

with [N/S] = integer of N/S and r = N – S[N/S]

For each forest, the relative density, relative frequency, relative dominance and importance value were calculated for each species (Roberts-Pichette and Gillespie 1999).

$$\text{Relative Density} = \frac{\text{\# of trees of species A in the sample}}{\text{Total \# of trees of all species in the sample}} \times 100$$

where trees with multiple stems are counted as single individuals

$$\text{Relative Frequency} = \frac{\text{frequency of species A in the sample}}{\text{Total frequency of all species in the sample}} \times 100 \quad \text{where freq.} = \frac{\text{\# of plots with species A}}{\text{Total \# of plots in stand}}$$

$$\text{Relative Dominance} = \frac{\text{basal area of species A } (m^2)}{\text{Total basal area of all species } (m^2)} \times 100$$

Species Importance Value = Relative Density + Relative Frequency + Relative Dominance

Species diversity, tree abundance and size class (dbh) distribution were compared graphically between the forest plots to give a general idea of differences in stand composition between the forests. The size class (dbh) distribution for each species was graphed for the three forests as an examination of recruitment and replacement patterns. The Cliffs and Alvars and Indian Woods forest plots were established in 2009, making this year the second monitoring season. The stand summary statistics from both years are presented side by side for comparison. To determine whether there was a significant increase in mean stand dbh over the year, a paired Wilcoxon signed rank test was used for both the Cliffs and Alvars and Indian Woods. This non-parametric analysis was used because the dbh distribution for both stands in both years were right skewed towards an abundance of smaller trees.

3.3 Results

3.3.1 Tree Species Diversity

Figure 3.2 shows the tree species diversity and abundance for all nine of the forest plots and Figure 3.3 shows the diameter at breast height size class distributions for each plot. Tables 3.1, 3.2 and 3.3 summarize the stand characteristics (for CA, IW and HB respectively) and Tables 3.4a, 3.4b, 3.5a, 3.5b, and 3.6 describe the stand species composition by year (for CA 2009, CA 2010, IW 2009, IW 2010 and HB respectively). Table 3.7 gives the scientific name and family for each of the species tagged in the forest plots.

The Indian Woods plots have the lowest tree species diversity (Brillouin diversity measure= 0.631, evenness= 0.516) of the three forests, containing only five species of trees belonging to only two families (Table 3.2). The Hogsback plots are the most diverse (Brillouin diversity= 1.803, evenness= 0.891) with ten tree species belonging to six families (Table 3.3). The diversity of the Cliffs and Alvars forest plots was also far greater than that of Indian Woods (Brillouin diversity = 1.382, evenness = 0.797) with eight tree species belonging to six families (Table 3.1). Plots in both Indian Woods and Cliffs and Alvars were dominated by Sugar Maple (*Acer saccharum*), while plots in the Hogsback were closely co-dominated by Sugar Maple and American Beech (*Fagus grandifolia*).

3.3.2 Stand Composition and Size Classes

Figure 3.3 shows the size (dbh) class distribution of tagged trees in each of the nine plots. Figures 3.4 to 3.6 show the species size class distributions for the three forest stands.

In the 2010 monitoring year, a single *Ostrya virginiana* was recruited to the 10cm dbh size class and added to the Cliffs and Alvars forest plot 1(CA1). There were no new mortalities between 2009 and 2010 in any of the Cliffs and Alvars plots.

The Indian Woods plots experienced one recruitment and three mortalities between the 2009 and 2010 monitoring years. In Indian Woods Forest Plot 1, a *Fagus Grandifolia* graduated to the 10cm dbh size class, while a large *Fagus grandifolia* died standing and an *Acer rubrum* fell across the plot. In Indian Woods Forest Plot 2, the stem of an *Acer rubrum* split and the tree is now suspended as a snag.

The plots in both Cliffs and Alvars and Indian Woods showed a significant increase in mean stand dbh between the monitoring years (paired Wilcoxon signed rank test: For CA, $V=110$, $p=3.615\times10^{-07}$; for IW, $V=22$, $p= 3.95\times10^{-05}$).

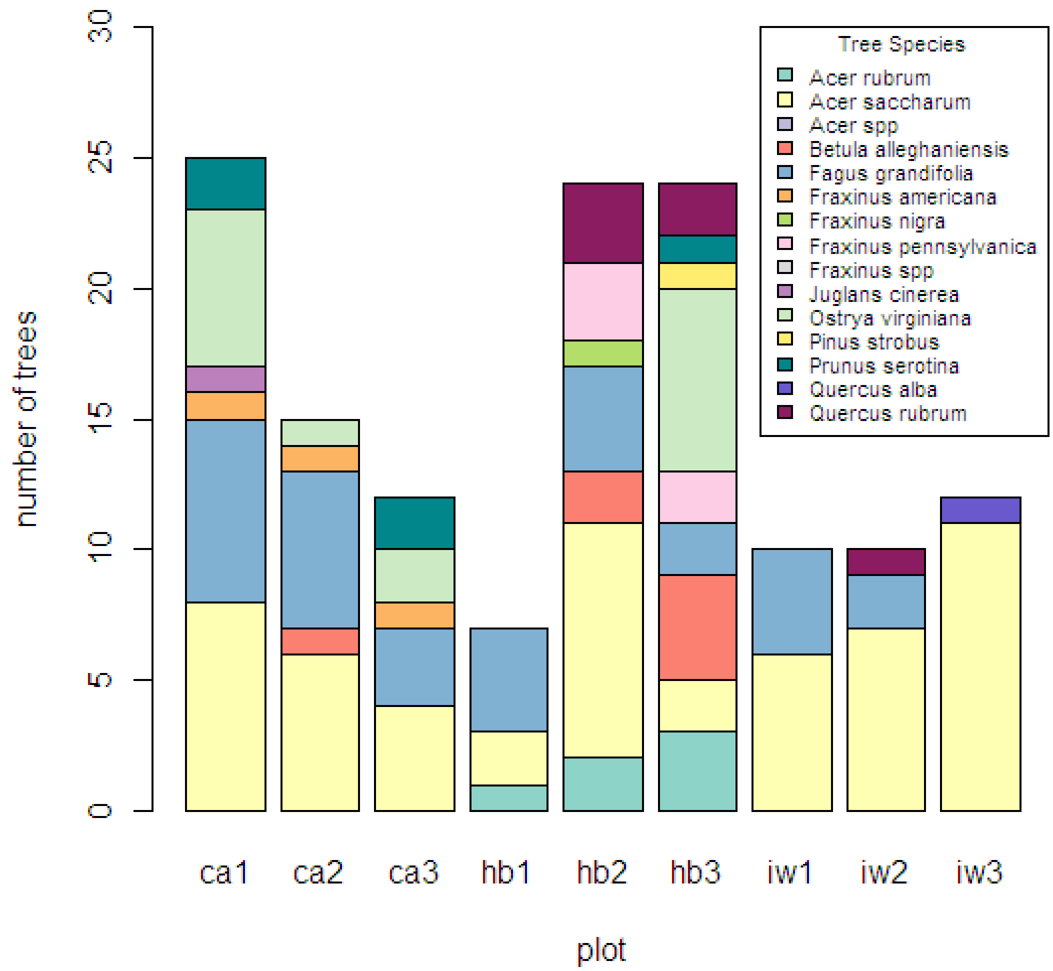


Figure 3.2 Tree species diversity and abundance for the nine EMAN forest plots. Cliffs and Alvars forest plots are represented by plot names ca1-ca3, Indian Woods forest plots are represented by plot names iw1-iw3 and Hogsback forest plots are represented by plot names hb1-hb3. Species common names: Red Maple (Acer rubrum), Sugar Maple (*Acer saccharum*), Yellow Birch (*Betula alleghaniensis*), American Beech (*Fagus grandifolia*), White Ash (*Fraxinus americana*) Black Ash (*Fraxinus nigra*), Green Ash (*Fraxinus pennsylvanica*), Butternut (*Juglans cinerea*), Ironwood (*Ostrya virginiana*), White Pine (*Pinus strobus*), Black Cherry (*Prunus serotina*), White Oak (*Quercus alba*), Red Oak (*Quercus rubra*).

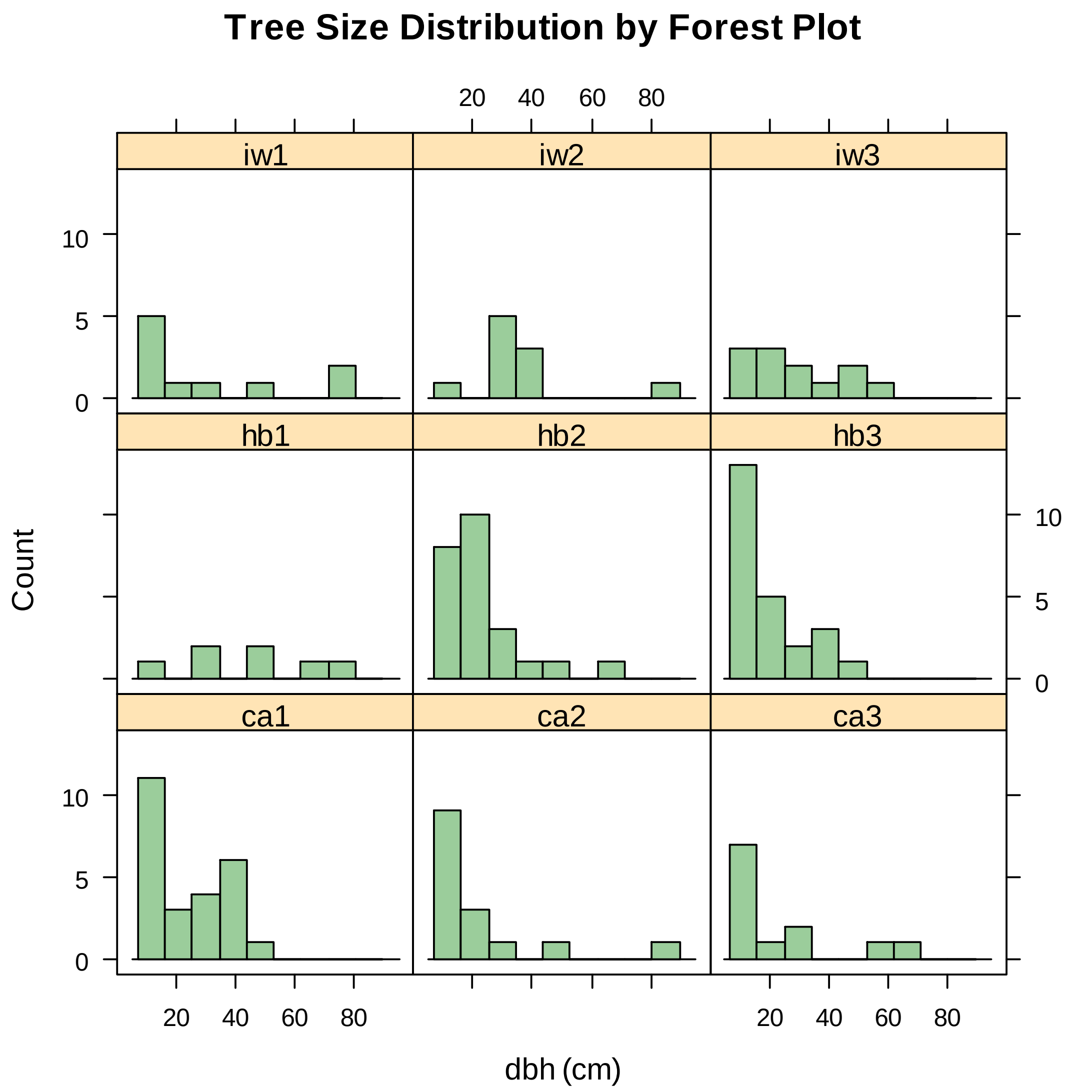


Figure 3.3 The canopy tree size class distributions, measured as diameter at breast height (dbh), for each of the nine forest plots. Cliffs and Alvars forest plots are represented by plot names ca1-ca3, Indian Woods forest plots are represented by plot names iw1-iw3 and Hogsback forest plots are represented by plot names hb1-hb3.

Table 3.1 Scientific name, common name and family name of the canopy tree species found on the ***rare*** Charitable Research Reserve property.

Species	Common name	Family
Betula alleghaniensis	Yellow Birch	Betulaceae
Ostrya virginiana	Ironwood (Eastern Hophornbeam)	Betulaceae
Fagus grandifolia	American Beech	Fagaceae
Quercus alba	White Oak	Fagaceae
Quercus rubra	Red Oak	Fagaceae
Juglans cinerea	Butternut	Juglandaceae
Fraxinus americana	White Ash	Oleaceae
Fraxinus pennsylvanica	Green Ash	Oleaceae
Fraxinus nigra	Black Ash	Oleaceae
Pinus strobus	White Pine	Pinaceae
Prunus serotina	Black Cherry	Rosaceae
Acer saccharum	Sugar Maple	Sapindaceae
Acer rubrum	Red Maple	Sapindaceae

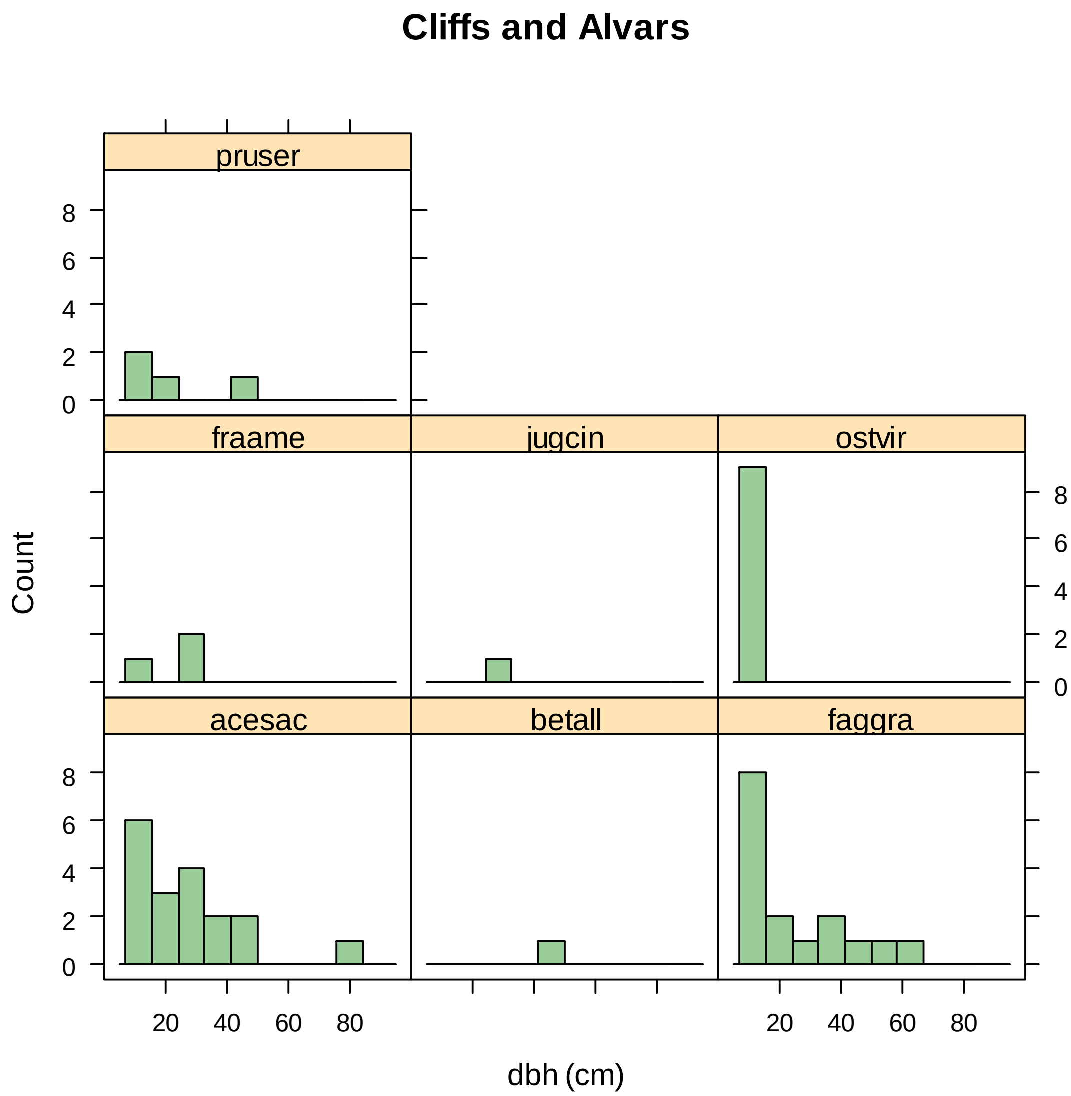


Figure 3.4 The canopy tree size class distributions, measured as diameter at breast height (dbh), for each of the seven species of living trees tagged within the Cliffs and Alvars forest plots. Species codes: acesac: Sugar Maple (*Acer saccharum*), betall: Yellow Birch (*Betula alleghaniensis*), faggra: American Beech (*Fagus grandifolia*), fraame: White Ash (*Fraxinus americana*), jugcin: Butternut (*Juglans cinerea*), ostvir: Ironwood (*Ostrya virginiana*), pruser: Black Cherry (*Prunus serotina*).

Table 3.2 Summary statistics of the stand characteristics of the Cliffs and Alvars forest for the 2009 and 2010 monitoring years. The data from the three forest plots in the Cliffs and Alvars were pooled to calculate the stand values.

Cliffs and Alvars	**2009 census**	**2010 census**
# families (A)	6	6
# species (A)	8 + 1 unknown	8 + 1 unknown
# species (L)	7	7
# unknown species	1	1
# of trees (L, K)	49	50
# of trees (A)	56	57
# stems (L, K)	51	52
# stems (A)	59	60
# stems (dead)	8	8
Brillouin Diversity Index (L, K)	1.376	1.382
Brillouin Evenness Index (L, K)	0.794	0.797
# tree recruitment 2009-2010	1	
# tree mortalities 2009-2010	0	
Mean stem dbh (cm)	23.07	23.34
Stem dbh SD (cm)	15.57	15.58
Total basal area (m^2ha^{-1})	25.69	26.65

A = all trees or stems, living, dead and of unknown species
L = only living trees/stem included
K = only trees/stems of known species included

Table 3.3a Tree species composition for the Cliffs and Alvars forest in 2010. Data from the three forest plots within the Cliffs and Alvars were pooled and only living trees were included in the calculations.

Species 2010	Abundance	Basal Area (m^2)	Relative Density	Relative Frequency	Relative Dominance	Importance Value
Acer saccharum	18	1.43	36.00	18.75	44.79	99.54
Betula alleghaniensis	1	0.16	2.00	6.25	4.91	13.16
Fagus grandifolia	14	1.08	28.00	18.75	33.88	80.63
Fraxinus americana	3	0.13	6.00	18.75	3.95	28.70
Juglans cinerea	1	0.07	2.00	6.25	2.03	10.28
Ostrya virginiana	9	0.10	18.00	18.75	3.11	39.86
Prunus serotina	4	0.23	8.00	12.50	7.33	27.83

Table 3.3b Tree species composition for the Cliffs and Alvars forest in 2009. Data from the three forest plots within the Cliffs and Alvars were pooled and only living trees were included in the calculations.

Species 2009	Abundance	Basal Area (m^2)	Relative Density	Relative Frequency	Relative Dominance	Importance Value
Acer saccharum	18	1.37	36.73	18.75	44.48	99.96
Betula alleghaniensis	1	0.15	2.04	6.25	4.94	13.23
Fagus grandifolia	14	1.05	28.57	18.75	34.16	81.48
Fraxinus americana	3	0.13	6.12	18.75	4.08	28.95
Juglans cinerea	1	0.06	2.04	6.25	2.08	10.37
Ostrya virginiana	8	0.09	16.33	18.75	2.89	37.97
Prunus serotina	4	0.23	8.16	12.50	7.37	28.03

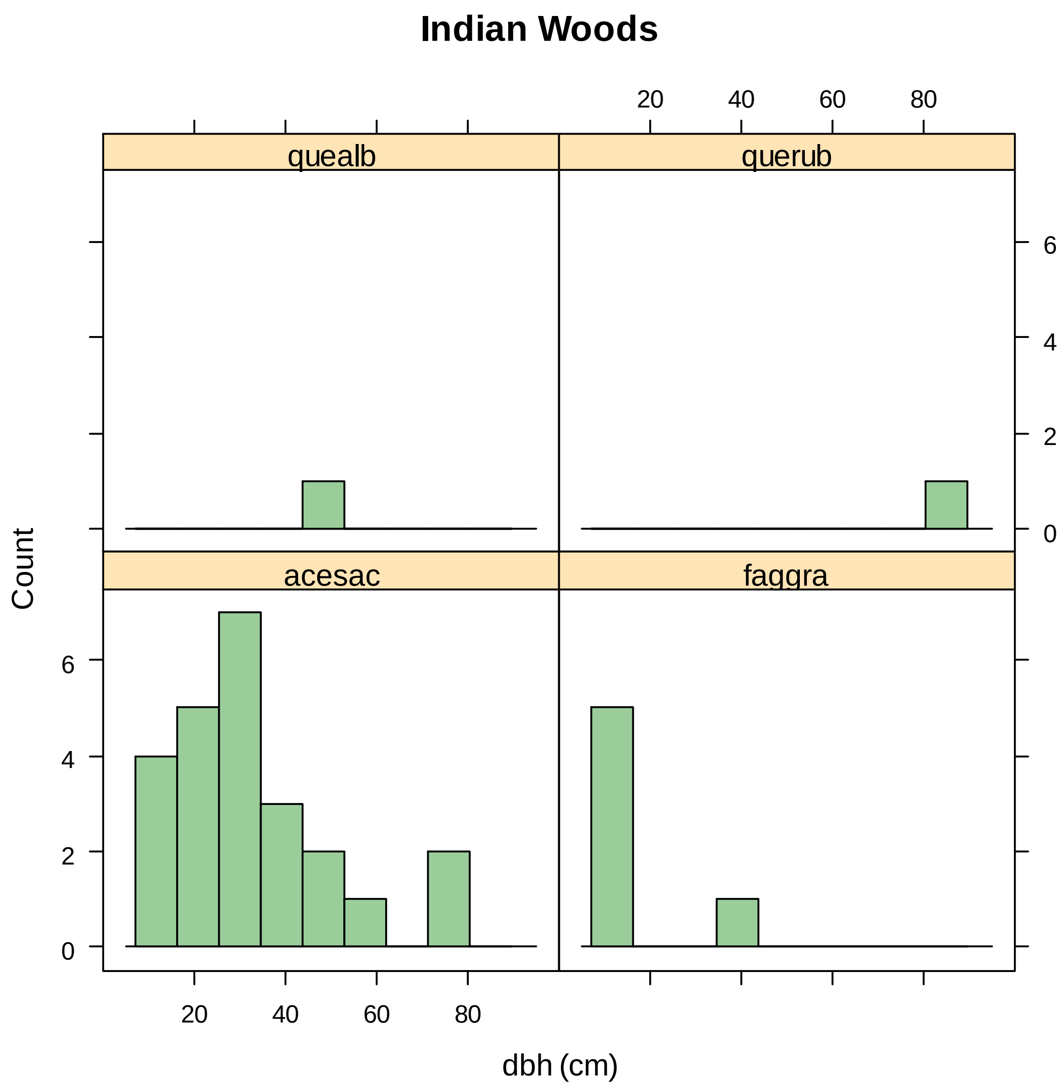


Figure 3.5 The canopy tree size class distributions, measured as diameter at breast height (dbh) for each of the four species of living trees tagged within the Indian Woods forest plots. Species codes: acesac: Sugar Maple (*Acer saccharum*), faggra: American Beech (*Fagus grandifolia*), quealb: White Oak (*Quercus alba*), querub: Red Oak (*Quercus rubra*).

Table 3.4 Summary statistics of the stand characteristics of the Indian Woods forest for the 2009 and 2010 monitoring years. The data from the three forest plots in the Indian Woods were pooled to calculate the stand values.

Indian Woods	**2009 census**	**2010 census**
# families (A)	2	2
# species (A)	5 + 1 unknown	5 + 1 unknown
# species (L)	5	4
# unknown species	1	1
# of trees (L, K)	34	32
# of trees (A)	38	39
# stems (L, K)	34	32
# stems (A)	38	39
# stems (dead)	4	7
Brillouin Diversity Index (L, K)	0.780	0.631
Brillouin Evenness Index (L, K)	0.554	0.516
# tree recruitment 2009-2010	1	
# tree mortalities 2009-2010	3	
Mean stem dbh (cm)	32.97	32.11
Stem dbh SD (cm)	18.82	19.96
Total basal area (m^2ha^{-1})	31.85	29.69

A = all trees or stems, living, dead and of unknown species
L = only living trees/stem included
K = only trees/stems of known species included

Table 3.5a Tree species composition for the Indian Woods forest in 2010. Data from the three forest plots within the Indian Woods were pooled and only living trees were included in the calculations.

Species 2010	Abundance	Basal Area (m^2)	Relative Density	Relative Frequency	Relative Dominance	Importance Value
Acer saccharum	24	2.6013	75.00	42.86	73.02	190.88
Fagus grandifolia	6	0.2076	18.75	28.57	5.83	53.15
Quercus alba	1	0.1646	3.13	14.29	4.62	22.03
Quercus rubra	1	0.5887	3.13	14.29	16.53	33.94

Table 3.5b Tree species composition for the Indian Woods forest in 2009. Data from the three forest plots within Indian Woods were pooled and only living trees were included in the calculations.

Species 2009	Abundance	Basal Area (m^2)	Relative Density	Relative Frequency	Relative Dominance	Importance Value
Acer rubrum	2	0.23	5.88	22.22	5.98	34.08
Acer saccharum	24	2.53	70.59	33.33	66.24	170.16
Fagus grandifolia	6	0.37	17.65	22.22	9.66	49.53
Quercus alba	1	0.15	2.94	11.11	3.99	18.04
Quercus rubra	1	0.54	2.94	11.11	14.14	28.19

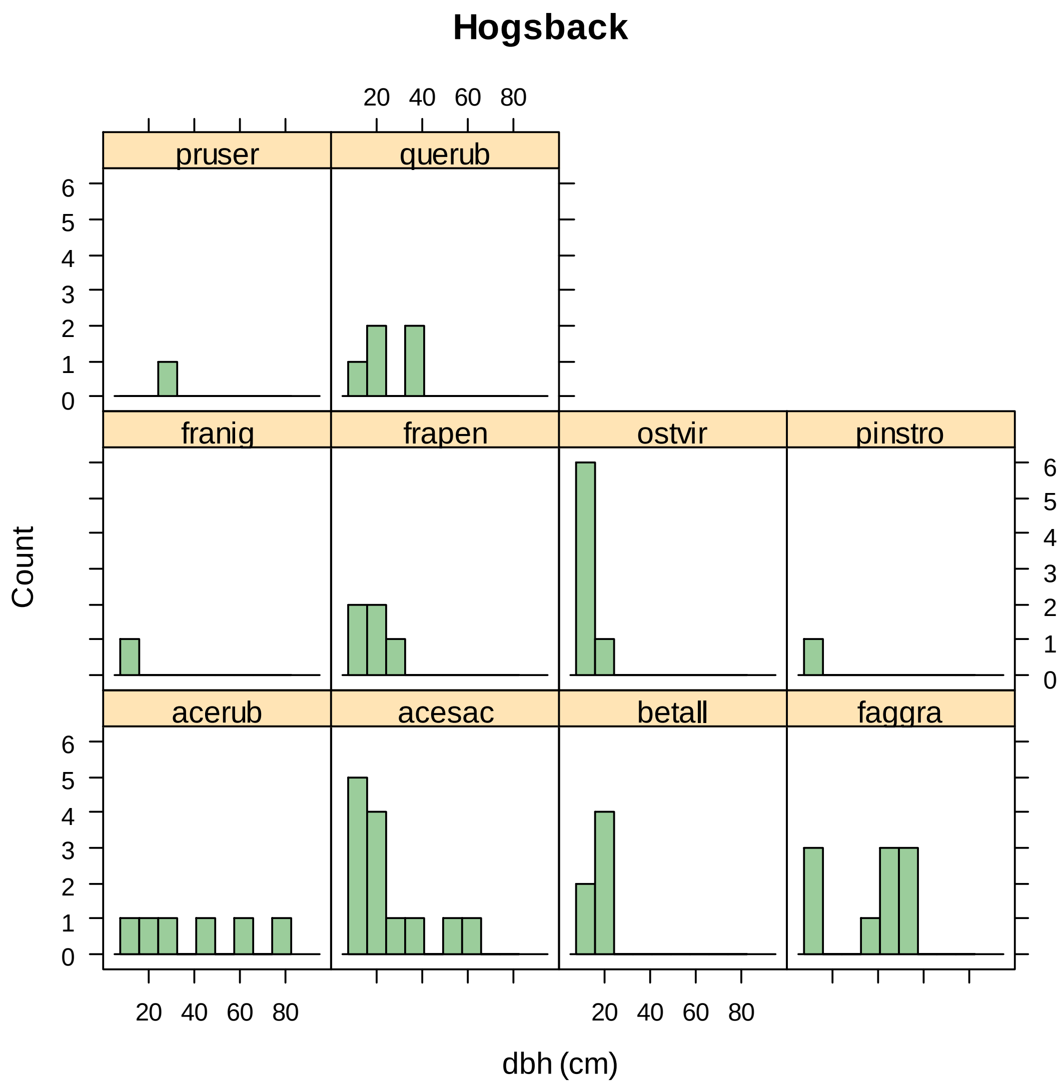


Figure 3.6 The canopy tree size class distributions, measured as diameter at breast height (dbh), for each of the ten species of living trees tagged within the Hogsback forest plots. Species codes: acerub: Red Maple (Acer rubrum), acesac: Sugar Maple (*Acer saccharum*), betall: Yellow Birch (*Betula alleghaniensis*), faggra: American Beech (*Fagus grandifolia*), franig: Black Ash (*Fraxinus nigra*), frapen: Green Ash (*Fraxinus pennsylvanica*), ostvir: Ironwood (*Ostrya virginiana*), pinstro: White Pine (*Pinus strobus*), pruser: Black Cherry (*Prunus serotina*), querub: Red Oak (*Quercus rubra*).

Table 3.6 Summary statistics of the stand characteristics of the Hogsback forest for the 2010 monitoring year. The data from the three forest plots in the Hogsback were pooled to calculate the stand values.

Hogsback	2010 census
# families (A)	6
# species (A)	10
# species (L)	10
# unknown species	0
# of trees (L, K)	54
# of trees (A)	60
# stems (L, K)	55
# stems (A)	61
# stems (dead)	6
Brillouin Diversity Index (L, K)	1.803
Brillouin Evenness Index (L, K)	0.891
Mean stem dbh (cm)	24.92
Stem dbh SD (cm)	16.37
Total basal area (m^2ha^{-1})	31.83

A = all trees or stems, living, dead and of unknown species

L = only living trees/stem included

K = only trees/stems of known species included

Table 3.7 Tree species composition for the Hogsback forest in 2010. Data from the three forest plots within the Hogsback were pooled and only living trees were included in the calculations.

Species 2010	Abundance	Basal Area (m^2)	Relative Density	Relative Frequency	Relative Dominance	Importance Value
Acer rubrum	6	1.07	11.11	15.79	28.14	55.04
Acer saccharum	13	0.84	24.07	15.79	21.94	61.80
Betula alleghaniensis	6	0.15	11.11	10.53	3.84	25.48
Fagus grandifolia	10	1.16	18.52	15.79	30.35	64.66
Fraxinus nigra	1	0.01	1.85	5.26	0.26	7.37
Fraxinus pennsylvanica	5	0.18	9.26	10.53	4.63	24.41
Ostrya virginiana	6	0.11	11.11	5.26	2.81	19.19
Pinus strobus	1	0.01	1.85	5.26	0.27	7.38
Prunus serotina	1	0.05	1.85	5.26	1.37	8.49
Quercus rubra	5	0.24	9.26	10.53	6.39	26.17

3.4 Discussion

3.4.1 Tree Diversity and Health

Indian Woods is an eastern deciduous old-growth remnant forest, an ecosystem that is rare in the region and to southwestern Ontario in general (***rare*** EMP 2006). The diversity of the Indian Woods forest plots was the lowest of the three forest stands examined, dominated almost exclusively by Sugar Maple (*Acer saccharum*). American Beech (*Fagus grandifolia*) was the second most important tree species in the forest, and single individuals of both Red and White oaks (*Quercus alba* and *rubra* respectively) were found within our plots. Old-growth forests are often viewed as a final stage in forest succession, representing a climax community that will persist in a state of dynamic equilibrium in the prevailing environmental conditions (Krebs 2001). As succession progresses and the canopy becomes more closed, the composition of canopy trees shifts towards more shade tolerant species such as Sugar Maple and American Beech (for eastern deciduous forests) (Fox 1977). These species are able to grow suppressed in the understory and they are then primed to exploit canopy gaps when they occur, outcompeting less shade tolerant species (Weiskittel and Hix 2003).

Hogsback forest plots were distributed over a slightly greater range of habitats than the Cliffs and Alvars and Indian Woods, with the plots positioned on top of upland ridges bordered by wet bog. The wet margins of the plots may account from some of the increased tree species diversity in the Hogsback, as Yellow Birch, Black Ash, Green Ash and Red Maple all thrive in wet soils (Sibley 2009).

The tree diameter at breast height (dbh) distributions of the three forests were plotted in Figures 3.3 through 3.6 to give a visual representation of the size-class composition of the stands. This information will be useful as baseline data to which the monitoring data from future years may be compared to examine the recruitment and replacement patterns of the stand (Parker 2003, Forrester and Runkle 2000). For example, in the Indian Woods forest plots (Figure 3.5) there is a single very large Red Oak, but no other Red Oaks in the smaller size classes, which are almost completely dominated by shade tolerant Sugar Maple recruits, a pattern similarly seen in another eastern deciduous old-growth forest in Ohio (McCarthy *et* al. 2001). From this, we could predict that in a century, Indian Woods will be increasingly dominated by Sugar Maple, likely at the expense of species richness. Long-term monitoring of the plots will allow predictions such as these to be tested.

Of the 12 known Butternut trees on the ***rare*** property, one individual was contained within our forest monitoring sample in the Cliffs and Alvars Forest Plot 1. Butternut is classified as Endangered by the Ontario Ministry of Natural Resources Species at Risk in Ontario (SARO) list (Ontario Ministry of Natural Resources 2008). The Butternut's decline is attributed to Butternut Canker (*Sirococcus clavigignenti-juglandacearum*), an introduced fungal disease that has been present in Ontario since the early 1990s. There is currently no prevention or treatment for the disease and most Butternut conservation efforts are focussed on the detection of resistant individuals for seed banking and grafting (Forest Gene Conservation Association 2010). Unfortunately, the Butternut surveyed in CA1 was found to be in severe decline: it had been classified as dead-standing in the 2009 monitoring season, but during the 2010 monitoring we found that it is in fact still living, albeit with severe crown dieback (>80%) and extensive wounds covering the entire height of the stem.

Severe decline was also detected in a large proportion of the Ash trees included in the plots. Of the five White Ash trees included in the Cliffs and Alvars forest plots, two were dead, two exhibited complete crown dieback with epicormic shoot growth, and the remaining tree showed only light to moderate crown dieback. Four of the five Green Ashes in the Hogsback monitoring plot were in severe decline with greater than 50% crown dieback. The remaining Green Ash and its Black Ash neighbour in HB2 demonstrated only light dieback. This high proportion of declining Ash trees is of particular concern given the recent discovery of the Emerald Ash Borer beetle (EAB - *Agrilus planipennis*) in the Waterloo region (CFIA 2010). Not all of the Ash declines observed in southern Ontario are thought to be caused by the Emerald Ash Borer; inspection of failing trees has pinned some of the blame on fungal root rot, bacterial infection (Pokorny and Sinclair 1994), other pest insects such as the Redheaded Ash Borer (*Neoclytus acuminatus*) and the Lilac Borer (*Podosesia syringae*) (Lyons *et* al. 2007) and the intolerance of the Ash's shallow root system to large fluctuations in precipitation regime (Cleland 2009). Should ***rare*** choose to supplement the EMAN forest biodiversity plots with an Ash specific monitoring program, the Canadian Food and Inspection Agency has developed a number of protocols for the detection and monitoring of EAB (Ryall *et* al. 2010). At the very least, the current forest plots will allow us to estimate the rate of Ash decline and to detect any resistance or resiliency in our tagged trees.

3.4.2 Recommendations for Future Forest Monitoring and Research

The nine established forest plots should be monitored annually. Roberts-Pichette and Gillespie (1999) suggest that the EMAN monitoring could be done as infrequently as once every five years, but the data obtained from yearly monitoring will be useful in determining the baselines and fluctuation patterns of ***rare***'s dynamic forests. Frequent monitoring is particularly important for populations at risk of disease or infestation; a small Ash tree infected by the Emerald Ash Borer can die within as few as one or two years from the time that it first displays symptoms (U.S. Department of Agriculture *et* al. 2010). Early detection of health threats like these will hopefully provide time for any appropriate management actions. Additional monitoring protocols including branch sampling (Ryall *et* al. 2010) and chemical-lure prism traps (using green leaf volatiles, Lyons 2010) could be incorporated into the forest monitoring program if desired.

Provided that forest plot monitoring could commence a few weeks earlier, the ground vegetation and shrub and small tree monitoring protocols could be added to the monitoring program. The 5m x 5m shrub and small tree plots are nested inside the corners of the 20m x 20m forest canopy tree plots, and the 1m x 1m ground vegetation plots are established along the sides of the canopy tree plots. All three types of plots should be monitored in the late summer and fall, but the ground vegetation plots must be monitored first in case of trampling during the tree and shrub monitoring (Roberts-Pichette and Gillespie 1999).

One of the goals of the forest tree monitoring program at ***rare*** is to determine the tree mortality and replacement patterns so that we can detect and predict changes to the stand composition. Canopy gaps are created when an overstory tree dies, increasing light to the understory where suppressed trees will then compete to fill its place (Forrester and Runkle 2000). As gap characteristics such as size, shape and the species of tree neighbours influence what species will graduate to the canopy (Weiskittel and Hix 2003), adding a canopy gap monitoring procedure to the forest monitoring program could provide information about the replacement patterns in our forest so that we can predict how forest composition will change in the coming years (see Forrester and Runkle 2000 for an example of canopy measurements).

Finally, a number of trees in the Cliffs and Alvars forest plots had developed small wet wounds around the nails for the tags. While the overall health of these trees appeared to be fine, future tagging should be done using pre-engraved tags on galvanized steel wire pigtails inserted into the soil at the base of the trees.

3.4.3 Conclusion

As of 2010, nine forest canopy tree plots (three plots in each of the three forest areas; Cliffs and Alvars, Indian Woods and the Hogsback) have been established and monitored. Monitoring should continue at minimum once every five years, but could be done every year without too much effort (approximately 1-2 hours per plot) now that the initial set up is complete.

The three forest stands differed greatly in their tree species diversity, with the slightly boggy Hogsback having the greatest diversity, and the old-growth Indian Woods having the lowest diversity, dominated strongly by Sugar Maple. The Cliffs and Alvars forest stand was similarly dominated by Sugar Maple and American Beech, but it had greater species richness, including an endangered Butternut tree.

There was little mortality between the two years of monitoring, although many of the ash trees within the plots were observed to be in decline. Additional monitoring procedures may be used to determine the cause of these declines.

4.0 Soil Humus Decay Rate Monitoring

4.1 Introduction

4.1.1 Soil Function

Decomposition is defined as the physical, chemical and biological breakdown of organic material into simpler matter, and it is a significant producer of carbon dioxide, as well as methane and nitrogen gases (Berg and McClaugherty 2008). Soil humus, the stable organic layer remaining after initial decomposition, acts as a 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 (Tenney and Waksman 1929). Much attention of late has been focused on the impact of global climate change on decay rate, with debate arising over whether climbing temperatures will necessarily increase decomposition rate and subsequently the release of carbon gasses into the atmosphere (Davidson and Janssens 2006, Ise *et* al. 2008, Giardina and Ryan 2000). In response to these concerns, Natural Resources Canada developed the Canadian Intersite Decomposition Experiment (Natural Resources Canada 2007) to examine the long-term litter decomposition rates and nutrient mineralization of forests across Canada. The moderate temperate zone of southwestern Ontario is the one area excluded from the study. Long-term monitoring of soil decay rates can provide valuable information on the relationship between soil decomposition and environmental factors, and it may serve to inform management decisions. For example, we currently can only guess at the effects that nearby aggregate mining or pesticide application may have on the health of our forests; decay rate monitoring, together with the other biological monitoring protocols in place at ***rare*** such as forest tree biodiversity and Plethodontid salamander monitoring, can only provide us with a greater understanding of the integrity and stability of our forest ecosystems.

4.1.2 Soil Humus Decay Rate Monitoring at *rare*

The objective of the EMAN soil humus decay rate monitoring procedure is to contribute to the overall assessment of forest ecosystem integrity by monitoring yearly mass loss in standardized decay sticks as a representation of soil decomposition. The EMAN (2006) decay rate protocol suggests locating the Annual Decay Rate (ADR) plots at the corners of the permanent Forest Canopy Tree Biodiversity plots. The information gained from decay monitoring can then be directly linked to the forest health and productivity data.

The first EMAN soil humus decay rate monitoring plots at ***rare*** were established on November 9, 2009 at the Cliffs and Alvars Forest Plot 1(CA1). The success of the first monitoring year encouraged us to expand the study in 2010 by establishing monitoring plots at Indian Woods Forest Plot 1 (IW1) and the Hogsback Forest Plot 1 (HB1).

4.2 Methods

4.2.1 Soil Humus Decay Plot Locations

For the 2010 soil decay monitoring, annual decay rate (ADR) plots were established on the corners of three of the forest plots, one in each forest on the property (CA1 on November 15 -1 6, 2010; IW1 on November 18, 2010; HB1 on November 19, 2010). Each forest plot had 12 ADR plots established around it, three at each corner of the plot (henceforth called corner stations). Section 3.2.1 provides detailed descriptions of the Cliffs and Alvars, Indian Woods and Hogsback forest plots. The 2009 soil decay monitoring included only the ADR stations at CA1.

4.2.2 Monitoring protocol

Decay Stick Installation

In preparation of decay stick installation, a 2mm hole was drilled at the end each tongue depressor (MedPro, 100% natural birch wood, ultra smooth finish). We prepared 160 tongue depressors for installation although only 144 are required for the three plots (48 sticks per plot). The tongue depressors were oven-dried at 70˚C for 48 hours and then let to sit for 24 hours at room temperature before being weighed (to ±0.001g) on a Sartorius 1265MP balance. After their mass was recorded, the tongue depressors were tagged with pre-labelled aluminum tags attached with approximately 30cm of extra-strong (40LB) fishing line. For the 2010 installation, depressors were placed in 100% vinyl mesh bags (total dimensions were 17cm x 4cm with a pocket size of 16cm x 3cm and a hole size of 3mm x 2mm) that were tied closed with fishing line. Many of the 2009 decay sticks extracted in 2010 were broken and missing pieces due to forces other than decay (e.g. hasty extraction or shifting due to ground freeze); the mesh bags were added to the protocol in an attempt to keep all the stick's pieces together and increase the number of decay sticks excavated intact in 2011.

A $1m^2$quadrat was marked on each corner of the forest plots, and three ADR plots were positioned on the corners not touching the forest plot (Figure 4.1). At each ADR plot, a 30cm x 30cm hole was excavated with the soil plug removed intact if possible and placed to the side. In the 2010 installation, the quadrat at each corner was shifted approximately 1m clockwise from the corner to ensure that the soil was undisturbed by the excavation of the previous year's sticks. Three incisions placed 5 cm apart were made with a chisel (chisel dimensions were 11cm x 2.5cm) into the north wall of the hole at a depth of 10cm from the surface or at the horizon of the organic and mineral layers if visible. The incisions were parallel to the soil surface.

The decay stick in its mesh bag was then inserted into the incision made in the soil. The pre-numbered aluminum forestry tags (attached to the stick with fishing line) were then placed on the soil surface. Each stick was also individually attached by fishing line to a galvanized steel pigtail labelled with the ADR plot number and inserted into the middle of the ADR hole. A forth decay stick (similarly strung, tagged and bagged) was placed on the soil surface on the north face of the plot (Figure 4.2). The weight and insertion depth of each stick was recorded. The hole was then refilled with the soil that was set aside and the tags were lightly covered with leaf litter in hopes of preventing them from being discovered and chewed by animals.

Decay Stick Excavation

The sticks installed on November 9, 2009 at the Cliffs and Alvars Forest Plot 1 (CA1) were excavated November 10, 2010. Decay sticks should be excavated close to the same date one year after their installation, but this date should be moved forward if there is a risk of the ground freezing. The tag and fishing line should help to indicate the position of the sticks under the soil. Using a trowel, gradually remove layers of soil above the stick's estimated location until it is visible. A butter knife may then be useful in gently extracting the stick from the soil. Place each stick and its tag together in individual plastic bags or paper envelopes.

To remove any dirt adhered to the sticks, each stick was gently brushed with a dry paintbrush and then gently scrubbed with a paintbrush in a pan of water. The sticks were placed in labelled paper envelopes and then oven-dried at 70˚C for 48 hours and then let to sit for 24 hours at room temperature before being weighed (to ±0.001g) on a Sartorius 1265MP balance. Because a large proportion of the sticks installed in 2009 fell apart during extraction, we roughly estimated the percent of the stick remaining by comparing it to an intact non-decayed tongue depressor.

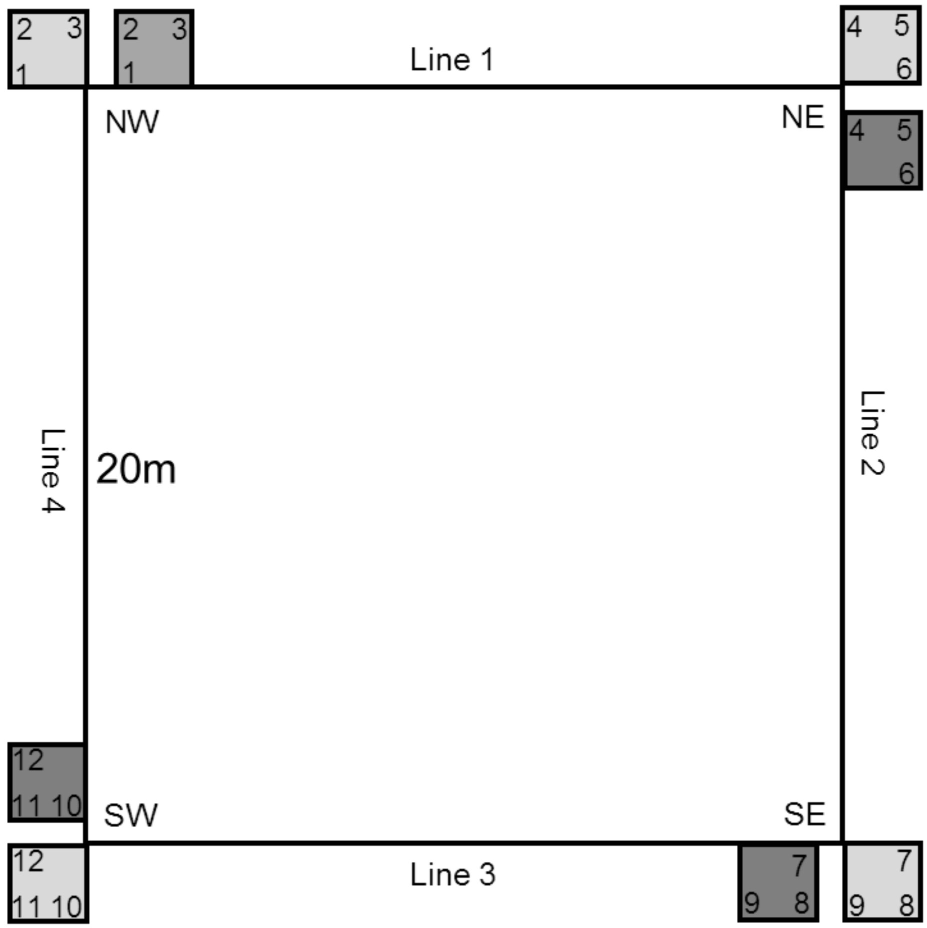


Figure 4.1 Distribution of annual soil humus decay rate (ADR) plots around a Forest Canopy Tree Biodiversity Plot. The light grey ADR stations on the corners are at the EMAN recommended locations, and represent the station locations during the first year of monitoring for each plot (i.e. CA1 in 2009, IW1 2010 and HB1 2010). For the second year of monitoring (CA1 2010), the ADR stations were shifted clockwise (shown as dark grey on the figure) to avoid installing sticks in the soil that was disturbed during extraction of the previous year's sticks. Adapted from McCarter (2009).

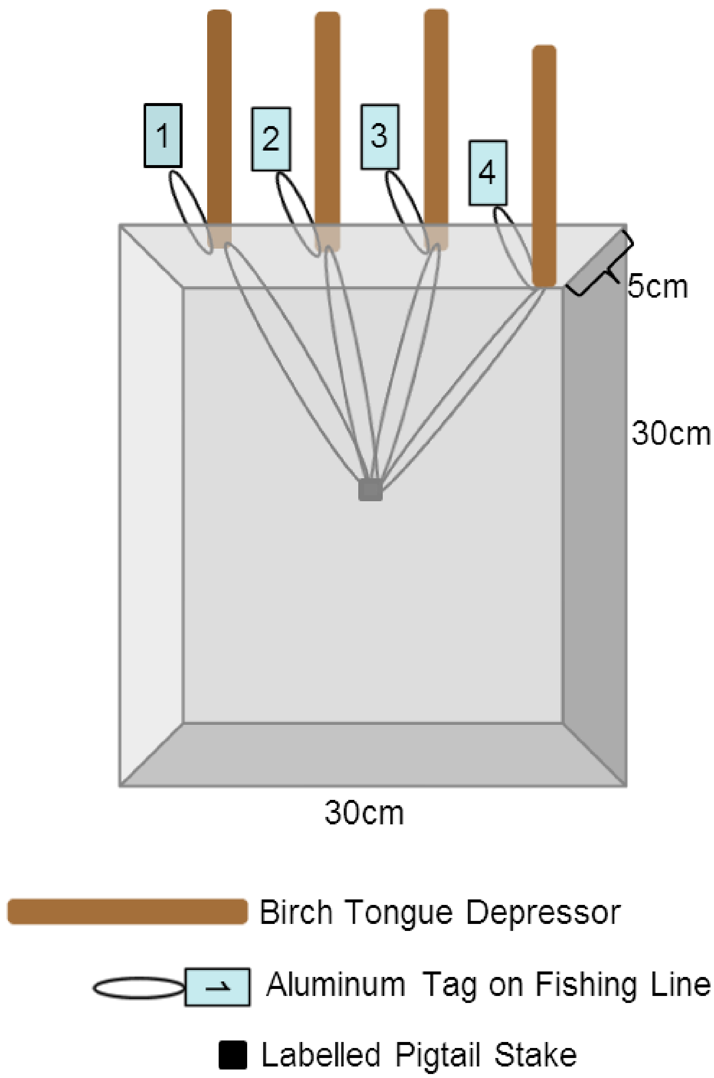


Figure 4.2 Diagram of annual soil humus decay rate (ADR) monitoring plot set-up as viewed from above. Decay sticks 1-3 are installed parallel to the soil surface at a depth of 5cm, separated 10cm from each other. Stick 4 is placed on the soil surface. Each decay stick is individually tied to a central pigtail stake with fishing line. Adapted from McCarter (2009).

4.2.3 Data analysis: 2009-2010 Decay Rate

The annual soil decay rate is calculated as the mean percent mass loss of the decay sticks. For each stick, the difference in mass between the 2009 and 2010 weight measurements was calculated as a percent of the stick's original mass. During extraction of the 2009 decay sticks, we found that many of the sticks were broken and we were unable to find the missing pieces. While it is possible that these pieces had simply decayed entirely, this seemed unlikely given that other unbroken or "intact" sticks from the same plot showed only surface pitting. Inclusion of the sticks with missing pieces into the calculations could overestimate the true mass loss due to decay. To deal with this issue, percent mass loss was calculated for five subsets of the decay sticks: Set 1 includes all decay sticks regardless of percentage of the stick broken off or missing; Set 2 includes only sticks with >50% of their original dimensions; Set 3 includes only intact sticks without any visibly missing pieces; Set 4 includes only visibly intact below-surface sticks; Set 5 includes only surface sticks. We predict that the mean mass loss of the sticks positioned on the surface of the soil will be less than the mean mass loss of the intact sticks placed below the soil, where they are more accessible to the soil organisms responsible for decomposition.

To determine whether the pre- and post-decay stick masses were significantly different, a non-parametric paired Wilcoxon signed rank test was used for each of the five sets examined. This non-parametric analysis was used because the stick mass distribution showed a departure from normality (right skew) for 2010 in both Set 1 and Set 5.

4.3 Results

4.3.1 2009-2010 Soil Humus Decay Rate

Table 4.1 provides the mean percent mass loss for each of the five subsets of the data as well as the mean magnitude of the mass loss. All of the data subsets, including the most conservative (set 5, which only included fully intact surface sticks) showed significant differences in mean mass from 2009 to 2010. As predicted, the intact below-soil sticks showed nearly double the percent mass loss shown by the intact sticks from the soil surface. Figure 4.3 shows the mass of the sticks pre- and post- decay for each of the five subsets of the data.

Table 4.1 Mean mass loss over the decay period (November 9, 2009 – November 10, 2010) for birch decay sticks installed in annual soil humus decay rate monitoring plots at the Cliffs and Alvars Forest Plot 1. Starred mass loss values indicate significant differences (at the $p<0.05$ level) in the mean pre- and post-decay masses for the subset of the decay sticks. “B” sticks were installed at a depth of 5cm below the soil surface, while “S” sticks were placed on the soil surface.

Set Number	Description	2009 mass (g)	2010 mass (g)	mass loss (g)	% mass loss
Set 1	All sticks	2.185	1.443	0.743*	34.0
Set 2	Sticks with <50% missing	2.206	1.558	0.648*	29.4
Set 3	Visibly intact sticks	2.186	1.713	0.473*	21.6
Set 4	Intact "B" sticks	2.204	1.550	0.654*	29.7
Set 5	"S" sticks	2.147	1.817	0.330*	15.4

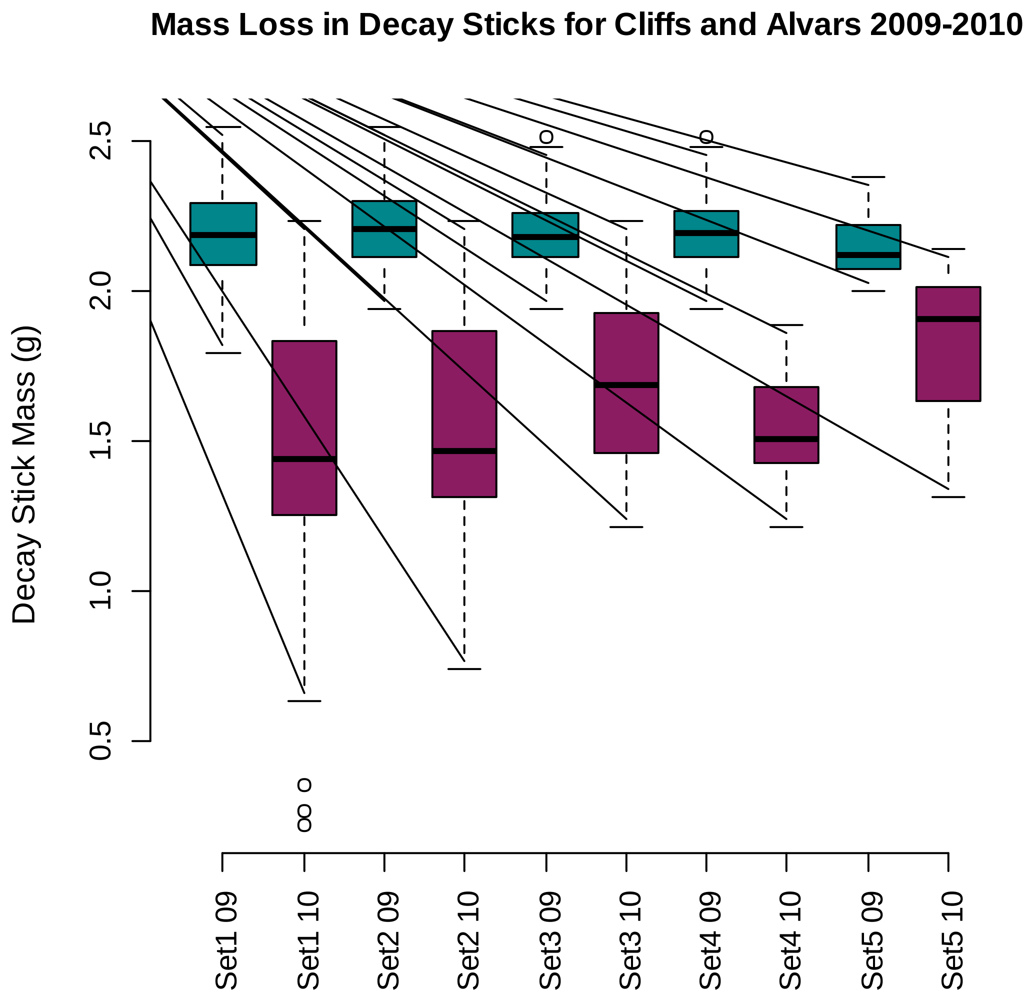


Figure 4.3 Mean pre- (2009) and post – (2010) masses of decay sticks in the Cliffs and Alvars. The decay sticks are divided into subsets based on their level of damage from extraction or position in the decay plot. Set 1 includes all decay sticks regardless of percentage of the stick broken off or missing; Set 2 includes only sticks with >50% of their original dimensions; Set 3 includes only intact sticks without any visibly missing pieces; Set 4 includes only visibly intact below-surface sticks; Set 5 includes only surface sticks.

4.4 Discussion

4.4.1 Annual Soil Humus Decay Rate

As predicted, there was a significant decrease in decay stick mass after one year in the soil at the CA1 ADR monitoring plot. Using our most conservative estimates, we found an annual decay rate of 29.7% for decay sticks installed to a depth of 10cm in the soil and 15.4% for sticks placed on the soil surface.

With the data obtained from the 2010-2011 ADR monitoring, we will be able to compare the decay rates at our different forests (Cliffs and Alvars, Indian Woods and the Hogsback). The ADR data from the next few monitoring years will allow us to determine how much variation in decay rate occurs between successive years, and this will hopefully provide us with a baseline Annual Decay Rate to which future monitoring results may be compared. Once this baseline is established and many years of data have been collected, the relationships between ADR and environmental conditions such as mean annual temperature, total annual moisture and ecological disturbance (i.e. annual pollution indices, nearby development) can be examined. Hopefully, these future finding will serve not only to increase the knowledge base of soil decomposition processes, but also to inform the land management practises on the ***rare*** property (e.g. Should we increase forest buffers from pesticide treated fields?).

4.4.2 Recommendations for Future Monitoring and Research

During excavation of the 2009 decay plots at CA1, many of the sticks were either broken during extraction or missing large pieces for unknown reasons. While it is possible that these missing pieces had decayed entirely, we suspect that the sticks had been broken by mechanical disturbances (such as ground shifting during freezing or hasty extractions). Because of these difficulties, we made a number of changes to the installation procedure to reduce the number of sticks that are missing pieces and therefore excluded from the more conservative analyses. While it is still possible that the sticks may break during extraction, we anticipate that the nylon mesh bag may serve to prevent any pieces from being lost in the soil. Mesh bags are often used in studies of leaf litter decay rate (Moore *et* al. 2005, Albers *et* al. 2004, Gallardo *et* al. 1995) We chose a wide-weave mesh in hopes that the holes would be large enough to allow for the passage of any micro and macro invertebrates involved in the soil decay process; it will be interesting to compare the decay rate of non-bagged intact sticks from the 2009 sample to those placed in bags for the 2010 monitoring year to examine the impact of the mesh on decay rate. Other protocol changes made during the 2010 installation include placing each tag on the soil surface instead of in the ADR hole and stringing each stick individually to the central pigtail stake instead of to a shared loop on the stake. These changes were intended to make it easier to locate individual decay sticks, which may have shifted apart from the rest of the sticks in the plot throughout the year.

4.4.3 Conclusion

The first year of decomposition monitoring was successful in that an adequate number of the decay sticks were excavated intact to calculate an estimate of annual soil decay rate for the Cliffs and Alvars Forest Plot 1 (CA1). Changes to the protocol, such as the use of nylon mesh bags and the placement of the tags on the surface of the soil, were made during the 2010 installation in hopes of increasing the proportion of decay sticks that are excavated from the soil intact. The monitoring data collected over the next few years will not only allow for the comparison of soil decay rates between forest sites, but it will also allow any changes in decomposition rate to be tracked and related to environmental variables or events.

5.0 Conclusion – Summary of Monitoring at *rare*

The monitoring season of 2010 was the 4th year of Plethodontid salamander monitoring in the Indian Woods forest and the 3rd year of Plethodontid salamander monitoring at the Hogsback. While the EMAN monitoring threshold for changes in Plethodontid salamander abundance is set at 5 years, we were able to detect a number of trends in the data collected so far. In Indian Woods, there is a trend towards decreasing abundance of Plethodontid salamanders, although this trend has not yet been observed in the Hogsback. The size of salamanders found under the artificial cover objects in Indian Woods has increased significantly over the monitoring years, perhaps suggesting that the same individuals are returning to the boards year after year. Finally, significant relationships between salamander abundance and the year, mean soil moisture and mean soil pH were detected in the Hogsback forest.

Forest canopy tree biodiversity monitoring plots were established in the Hogsback, and the previously established plots in the Cliffs and Alvars and Indian Woods forests were monitored for growth, recruitment, mortality and health. Forest tree biodiversity and community structure was markedly different in each forest stand, with the Hogsback having the greatest tree species diversity and Indian Woods having the lowest. The majority of Ash trees observed within the forest plots were in severe decline and additional monitoring protocols can be included in the forest tree monitoring program to determine whether the Emerald Ash Borer is responsible for the observed decline.

Decay sticks installed in the Cliffs and Alvars in November 2009 were excavated in November 2010, and additional decay rate monitoring plots were installed in Indian Woods and the Hogsback. The monitoring protocol was altered slightly by placing the tongue depressors in mesh bags in hopes of increasing the proportion of sticks recovered intact. Monitoring data collected over the next few years will allow for the comparison of soil decay rates between forest sites and across years.

Together, these three monitoring programs provide a broad picture of the function and health of the forests at ***rare***. As each program is relatively new, the data collected so far will serve to build a valuable baseline to which future years can be compared. Continuation of this long-term monitoring will allow for the early detection of any changes in the health and structure of the forest ecosystems at the ***rare*** Charitable Research Reserve.

References

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

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

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

Casper, G.S. 2011. *Plethodon cinereus*. Amphibiaweb: Information on Amphibian Biology and Conservation. amphibiaweb.org. Web. Accessed January 20, 2011.

CFIA: Canadian Food Inspection Agency. 2010. News Release: Emerald Ash Borer confirmed in the Regional Municipality of Waterloo, and the County of Oxford, Ontario. August 23, 2010.

Cleland, E. 2009. Forest Health Update. Federation of Ontario Cottagers' Associations. www.foca.on.ca

Conant, R., and J.T. Collins. 1998. A field guide to reptiles and amphibians. Eastern and Central North America. 3rd edition. Houghton Mifflin Company. Boston, United Sates of America.

Crawley, M.J. 2005. Statistics: An Introduction Using R. John Wiley and Sons. London, United Kingdom.

Davidson, E.A., and I.A. Janssens. 2006. Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. Nature 440:165-173.

EMAN Ecosystem Monitoring Partnership. 2004. Tree Health. Ecological Monitoring and Assessment Network, Environment Canada. http://www.ec.gc.ca/faunescience-wildlifescience/default.asp?lang=En&n=9C4F74C5-1 *Microsoft Word* file.

EMAN Ecosystem Monitoring Partnership and Parks Canada. 2006. Terrestrial monitoring protocol: annual soil humus decay rates – Draft 3. Ecological Monitoring and Assessment Network Coordination Office. Burlington, Ontario, Canada.

Environment Canada. 2010. Ecological Monitoring. http://www.ec.gc.ca/faunescience-wildlifescience/default.asp?lang=En&n=B0D89DF1-1 . Web. Accessed January 21, 2011.

Forest Gene Conservation Association. 2010. Butternut Tree: A land owner's resource guide. Ontario Woodlot Association. http://www.ont-woodlot-assoc.org/pdf/A%20Landowners%20Resource%20Guide%20To%20Butternut.pdf PDF file

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

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

Francl, K.E., C.R. Faidley, and C.J. Small. 2010. Salamander use of karst sinkholes in Montgomery County, Virginia. Southeastern Naturalist 9(1):35-46.

Führer, E. 2000. Forest functions, ecosystem stability and management. Forest Ecology and Management 132:28-38.

Gallardo, J.F., I. Santa-Regina, A.F. Harrison, and D.M. Howard. 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.

Giardina, C.P., and M.G. Ryan. 2000. Evidence that decomposition rates of organic carbon in mineral soil do not vary with temperature. Nature 404:858-861.

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

Heatwole, H., and K. Lim. 1961. Relation of substrate moisture to absorption and loss of water by the salamander *Plethodon cinereus*. Ecology 42(4):814-819.

Ise, T., A.L. Dunn, S.C. Wofsy, and P.R. Moorcroft. 2008. High sensitivity of peat decomposition to climate change through water-table feedback. Nature Geoscience 1(11):763-766.

Krebs, C. J. 2001. Ecology: The experimental analysis of distribution and abundance. 5th Edition. Benjamin Cummings. San Francisco, United States of America. pp.423-424.

Larson, A., D. Wake, and T. Devitt. 2006. Plethodontidae. Lungless Salamanders. http://tolweb.org/Plethodontidae/15441/2006.09.26 Web. Accessed January 26, 2011

LeClair, M.H., M. Levasseur, and R. LeClair Jr. 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.

Legendre, P., and L. Legendre. 1998. Numerical Ecology. 2nd English edition. Elsevier Science BV. Amsterdam, Netherlands.

Lyons, D.B. Emerald Ash Borer. 2010. Frontline Forestry Research Applications. Canadian Forest Service, Natural Resources Canada. Technote 110.

Lyons, D. B., C. Caister, P. De Groot, B. Hamilton, K. Marchant, T. Scarr, and J. Turgeon. 2007. Survey guide for detection of Emerald Ash Borer. Canadian Forest Service, Natural Resources Canada.

Magurran, A. 2004. Measuring biological diversity. Blackwell Publishing. Oxford, United Kingdom. pp. 113.

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

McCarter, J. 2009. Ecological Monitoring at ***rare*** Charitable Research Reserve. http://www.raresites.org/cms/en/Research/ResearchMonitoringandInventoryReports.aspx Web.

McCarthy, B. C., C.J. Small, and D.L. Rubino. 2001. Composition, structure and dynamics of Dysart Woods, an old-growth mixed mesophytic forest of southeastern Ohio. Forest Ecology and Management 140:193-213.

Monti, L., M. Hunter, and J. Witham. 2000. An evaluation of the artificial cover object (ACO) method for monitoring populations of the Redback Salamander *Plethodon cinereus*. Journal of Herpetology 34(4): 624-629.

Moore, T.R., J.A. Trofymow, M. Siltanen, C. Prescott, and CIDEY 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 Forestry Research 35:133-142.

Natural Resources Canada. 2007. Canadian Intersite Decomposition Experiment (CIDET). Canadian Forest Service. http://cfs.nrcan.gc.ca/subsite/cidet Web. Accessed January 20, 2011.

Ontario Ministry of Natural Resources. 1999. The old-growth forests of southern Ontario. Extension Notes. LandOwner Resource Centre. http://www.lrconline.com/Extension_Notes_English/pdf/oldgwth.pdf PDF file.

Ontario Ministry of Natural Resources. 2008. Species at risk in Ontario: Butternut. http://www.mnr.gov.on.ca/en/Business/Species/2ColumnSubPage/276504.html Web. Accessed January 20, 2011.

Parks Canada. 2007. Species at Risk: Glossary. http://www.pc.gc.ca/nature/eep-sar/itm6.aspx Web. Accessed January 20, 2011.

Parks Canada. 2009. National Parks of Canada: Ecological Integrity. http://www.pc.gc.ca/progs/np-pn/ie-ei.aspx Web. Accessed January 21, 2011.

Parker, B. H. 2003. Monitoring program status report. Long Point World Biosphere Reserve. http://longpointbiosphere.com/library/statusrt.pdf PDF file.

Pokorny, J.D., and W.A. Sinclair. 1994. How to identify and manage ash yellows in forest stands and home landscapes. USDA Forest Service Northeaster Area State and Private Forestry Report NA-FR-03-94.

rare Charitable Research Reserve. 2006. Draft Updated Environmental Management Plan for ***rare***.

Rice, W.R. 1989. Analyzing tables of statistical tests. Evolution 43(1):223-225.

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

Ryall, K.L., J.G. Fidgeon, and J.J. Turgeon. 2010. Detection of Emerald Ash Borer in urban environments using branch sampling. Frontline: Forestry Research Applications. Canadian Forest Service, Natural Resources Canada. Technote 111.

Sibley, D.A. 2009. The Sibley Guide to Trees. Alfred A. Knopf, Inc. New York, United States of America.

Styers, D.M., A.H. Chappelka, L.J. Marzen, and G.L. Somers. 2010. Scale matters: indicators of ecological health along the urban-rural interface near Columbus, Georgia. Ecological Indicators 10(2):224-233.

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

Tenney, F.G., and S.A. Waksman. 1929. Composition of natural organic materials and their decomposition in the soil: IV. The nature and rapidity of decomposition of the various organic complexes in different plant materials, under aerobic conditions. Soil Science 28(1):55-84.

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

U.S. Department of Agriculture Forest Service, Animal and Plant Health Inspection Service, State of Michigan, and Michigan State University. 2010. Emerald Ash Borer. www.emeraldashborer.info Web. Accessed January 26, 2011

van Dyke, F. 2008. Conservation biology: foundations, concepts, applications. 2nd edition. Springer BV. pp.91.

Vaughn, H., T. Brydes, A. Fenech, and A. Lumb. 2001. Monitoring long-term ecological changes through the Ecological Monitoring and Assessment Network: Science-Based and Policy Relevant. Environmental Monitoring and Assessment 67:3-28.

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

Welsh, H.H. Jr., and S. Droege. 2000. A case for using Plethodontid salamanders for monitoring biodiversity and ecosystem integrity of North American forests. Conservation Biology 15(3):558-569.

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

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

Wyman, R.L., and D.S. Hawksley-Lescault. 1987. soil acidity affects distribution, behaviour, and physiology of the salamander *Plethodon cinereus*. Ecology 68(6):1819-1827.

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

Appendix A: Maps

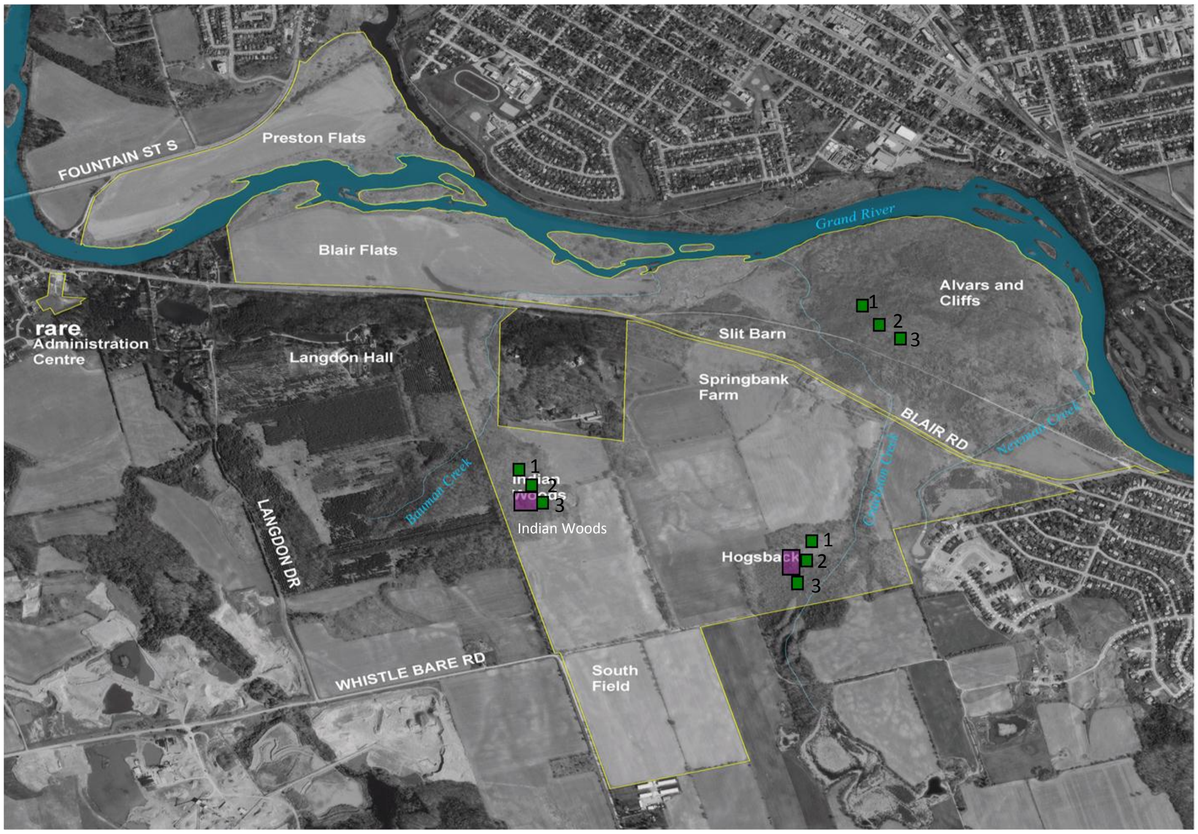


Figure A.1 Monitoring map of the ***rare*** Charitable Research Reserve. Green squares represent forest canopy tree biodiversity and health monitoring plots. Purple rectangles represent the Plethodontid salamander monitoring plots. Annual soil humus decay monitoring plots are located at the Cliffs and Alvars forest plot 1, Indian Woods forest plot 1 and the Hogsback forest plot 1. Scale= 1: 20,000.

Table A.1 Geographic coordinates of artificial cover objects used for Plethodontid salamander monitoring in Indian Woods and the Hogsback (from McCarter, 2009).

Monitoring Plot	**Artificial Cover Object**	**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

Table A.2 Geographic coordinates of the forest canopy tree biodiversity and health monitoring plots in Cliffs and Alvars, Indian Woods and the Hogsback (Adapted from McCarter 2009). 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 plots CA1, IW1 and HB1.

Forest	**Plot**	**Latitude and Longitude**	**UTM (zone 17T)**
Cliffs and Alvars (CA)	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 (IW)	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 (HB)	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

Cliffs and Alvars Forest Plot 1 – CA1

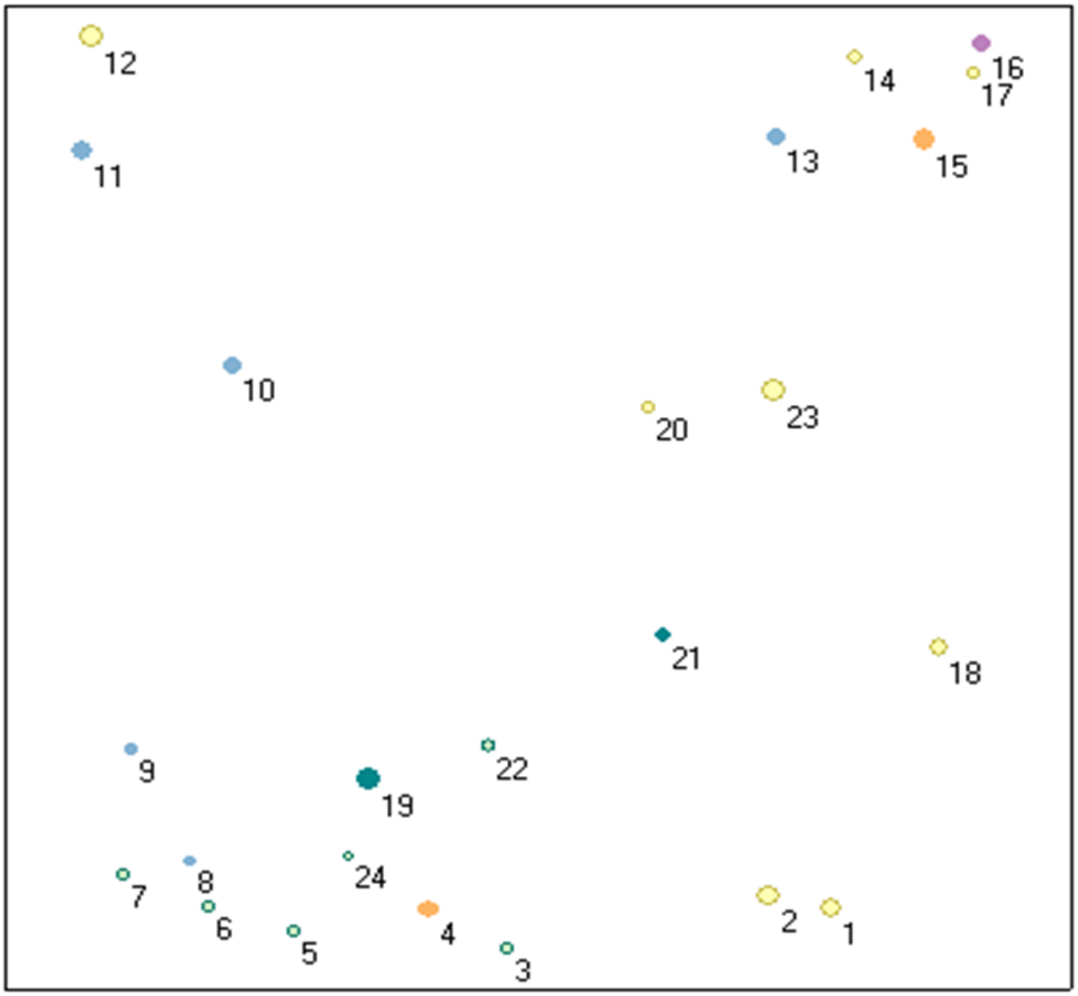
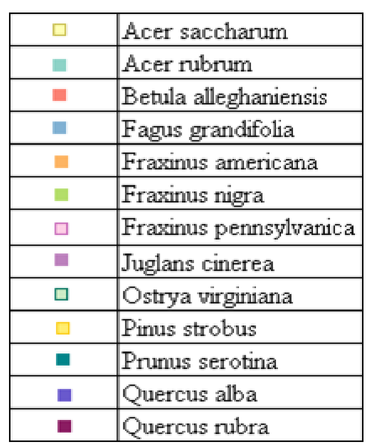


Figure A.2 Map of Cliffs and Alvars Forest Plot 1 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.



Cliffs and Alvars Forest Plot 2 – CA2

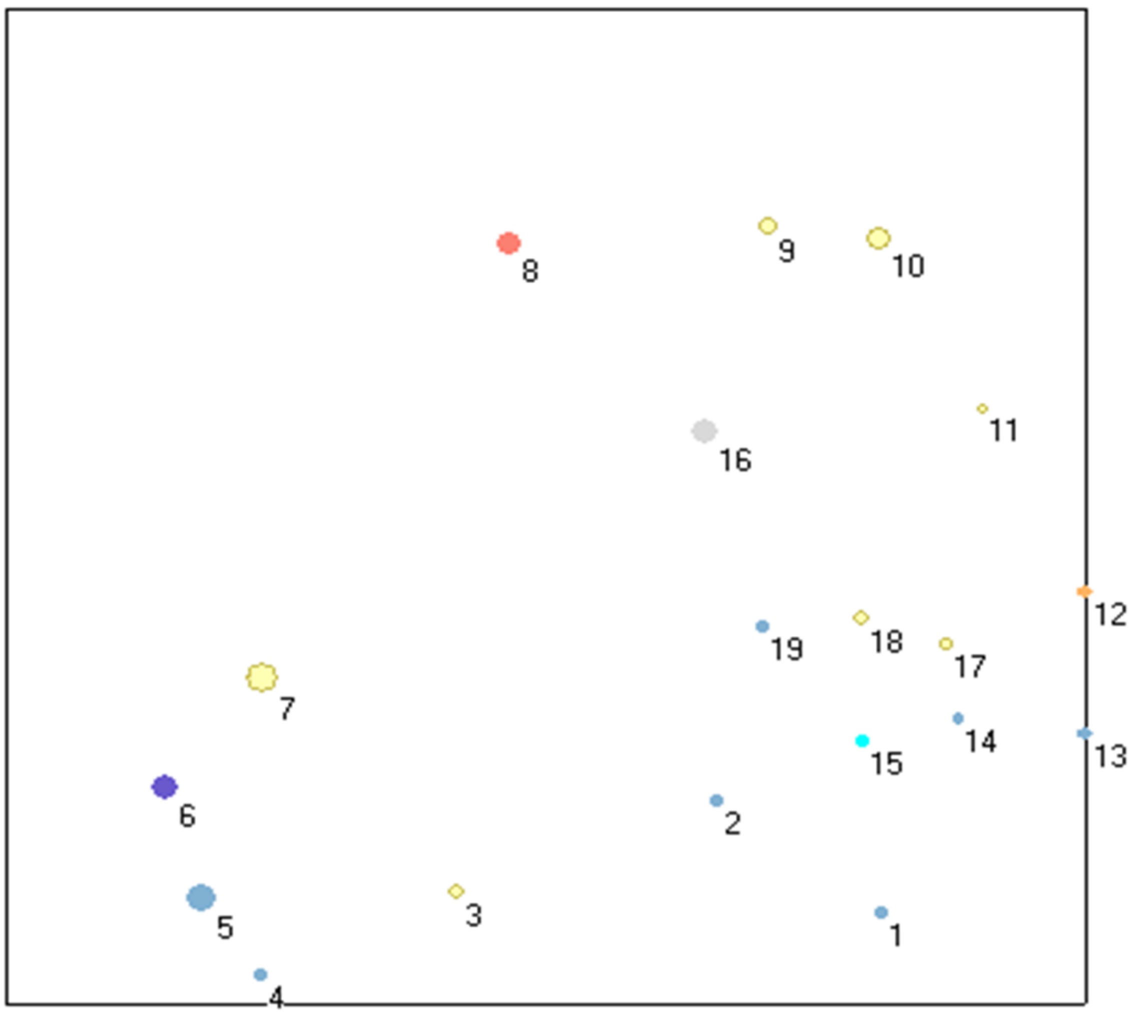


Figure A.3 Map of Cliffs and Alvars Forest Plot 2 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.

	Acer saccharum
	Acer rubrum
	Betula alleghaniensis
	Fagus grandifolia
	Fraxinus americana
	Fraxinus nigra
	Fraxinus pennsylvanica
	Juglans cinerea
	Ostrya virginiana
	Pinus strobus
	Prunus serotina
	Quercus alba
	Quercus rubra

Cliffs and Alvars Forest Plot 3 – CA3

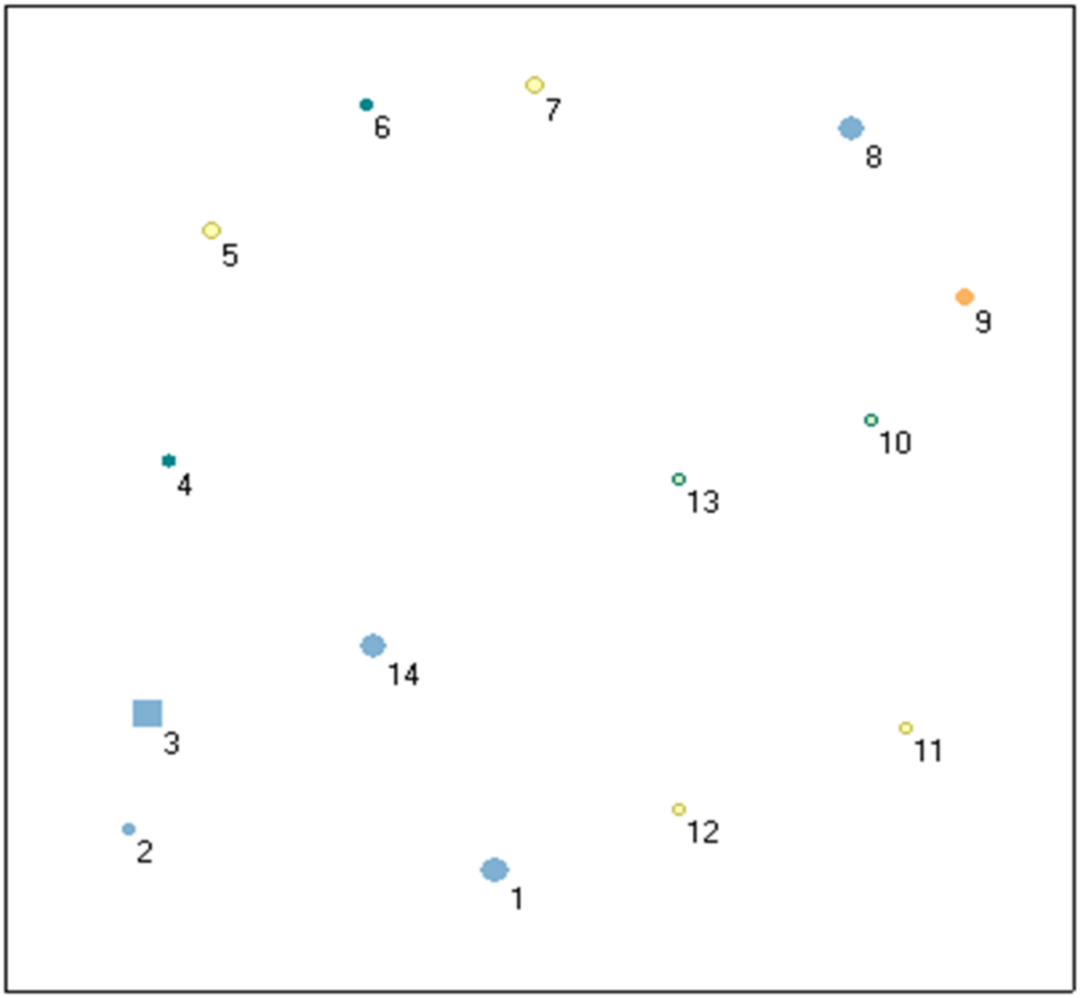
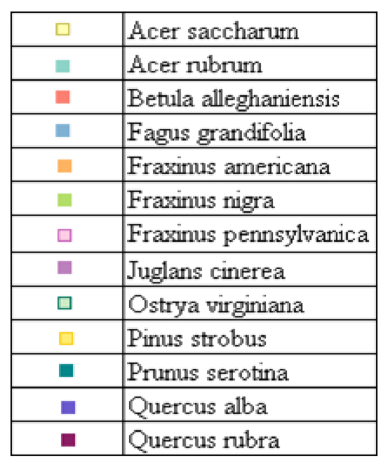


Figure A.4 Map of Cliffs and Alvars Forest Plot 3 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.



Indian Woods Forest Plot 1 – IW1

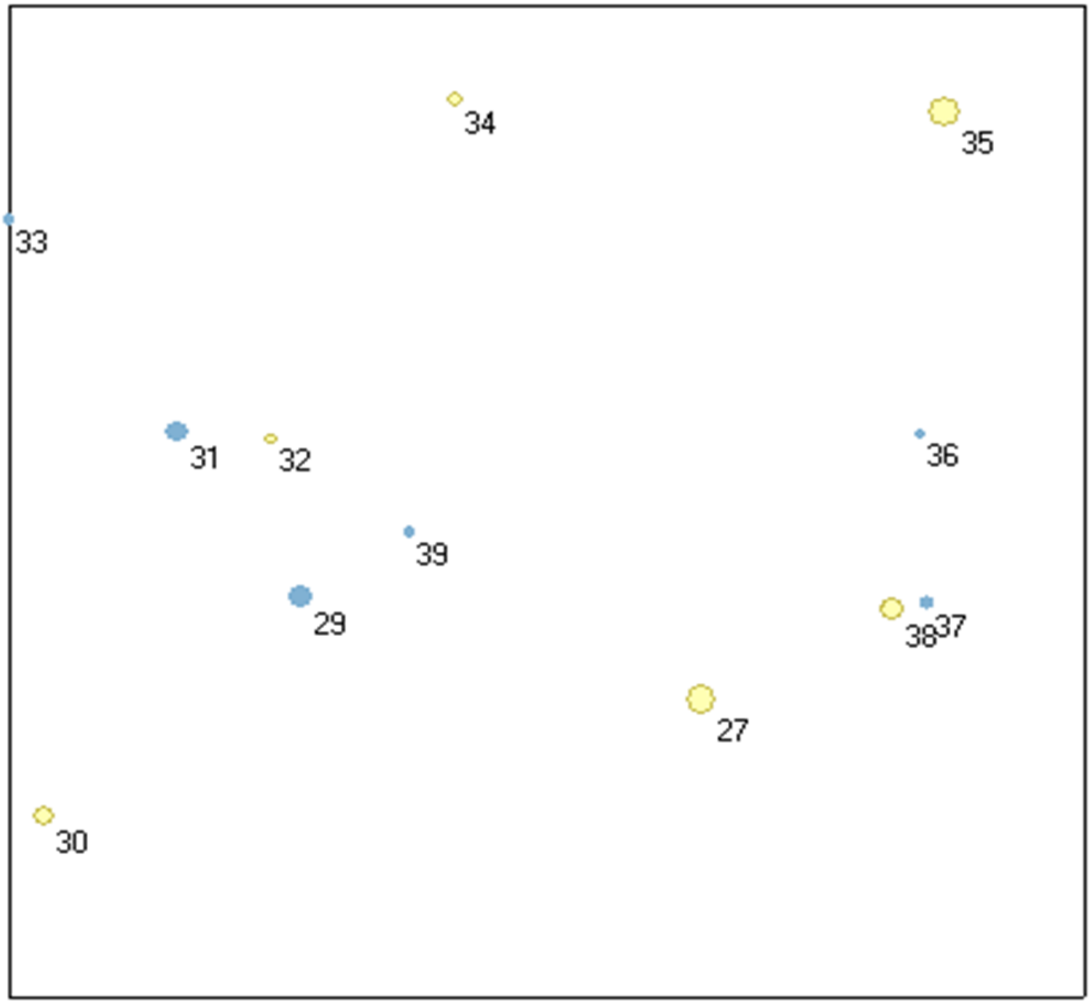


Figure A.5 Map of Indian Woods Forest Plot 1 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.

	Acer saccharum
	Acer rubrum
	Betula alleghaniensis
	Fagus grandifolia
	Fraxinus americana
	Fraxinus nigra
	Fraxinus pennsylvanica
	Juglans cinerea
	Ostrya virginiana
	Pinus strobus
	Prunus serotina
	Quercus alba
	Quercus rubra

Indian Woods Forest Plot 2 – IW2

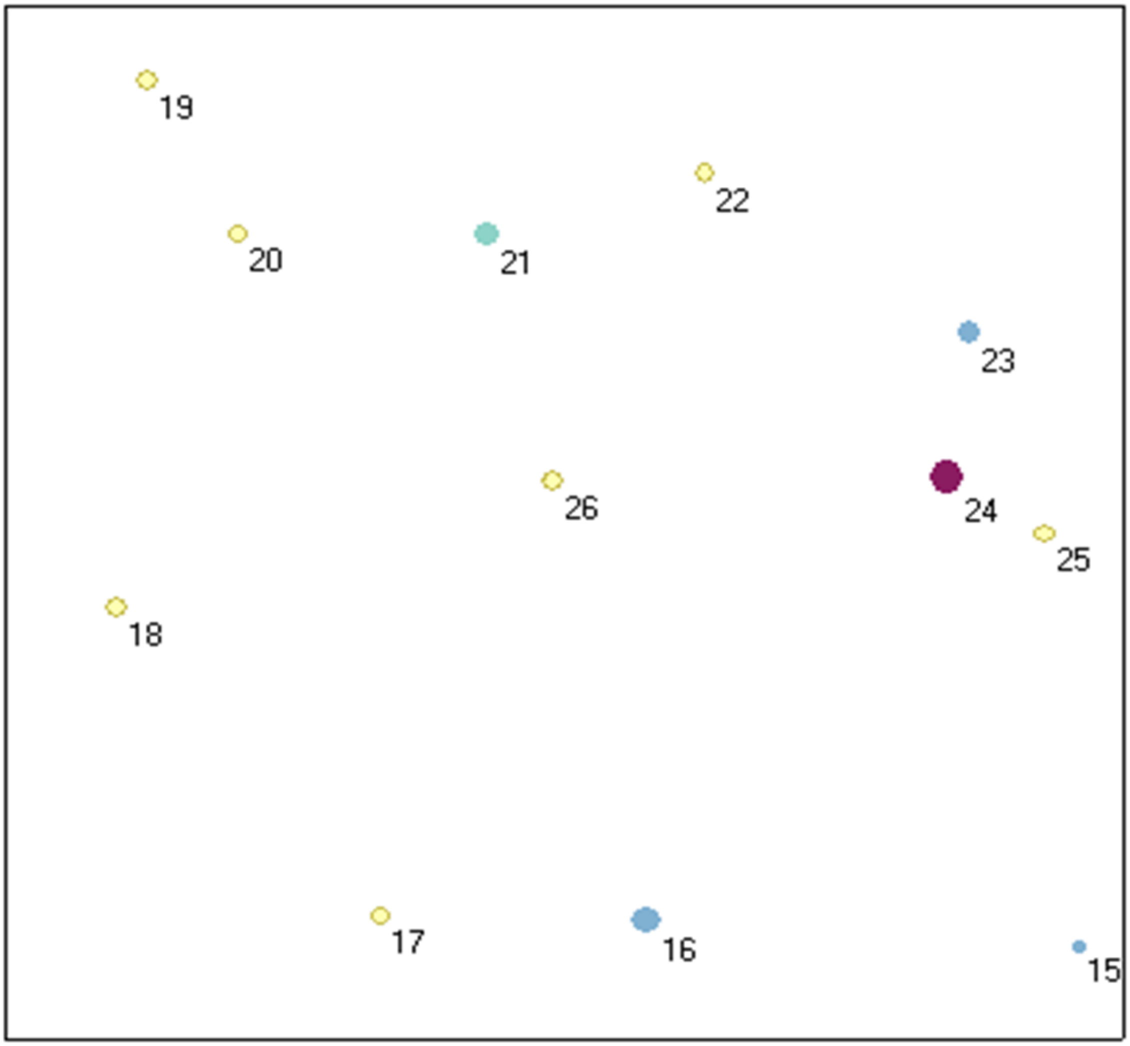


Figure A.6 Map of Indian Woods Forest Plot 2 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.

	Acer saccharum
	Acer rubrum
	Betula alleghaniensis
	Fagus grandifolia
	Fraxinus americana
	Fraxinus nigra
	Fraxinus pennsylvanica
	Juglans cinerea
	Ostrya virginiana
	Pinus strobus
	Prunus serotina
	Quercus alba
	Quercus rubra

Indian Woods Forest Plot 3 – IW3

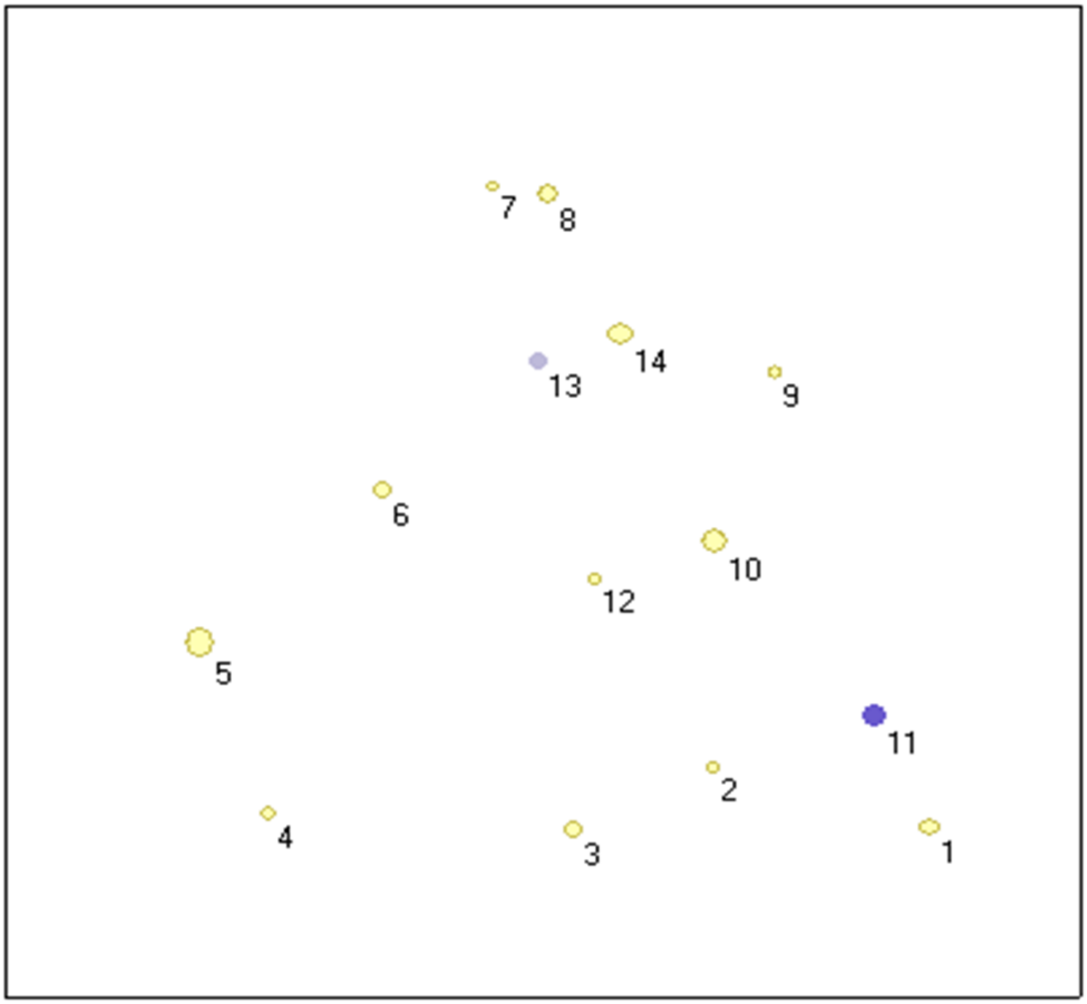


Figure A.7 Map of Indian Woods Forest Plot 3 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.

	Acer saccharum
	Acer rubrum
	Betula alleghaniensis
	Fagus grandifolia
	Fraxinus americana
	Fraxinus nigra
	Fraxinus pennsylvanica
	Juglans cinerea
	Ostrya virginiana
	Pinus strobus
	Prunus serotina
	Quercus alba
	Quercus rubra

Hogsback Forest Plot 1- HB1

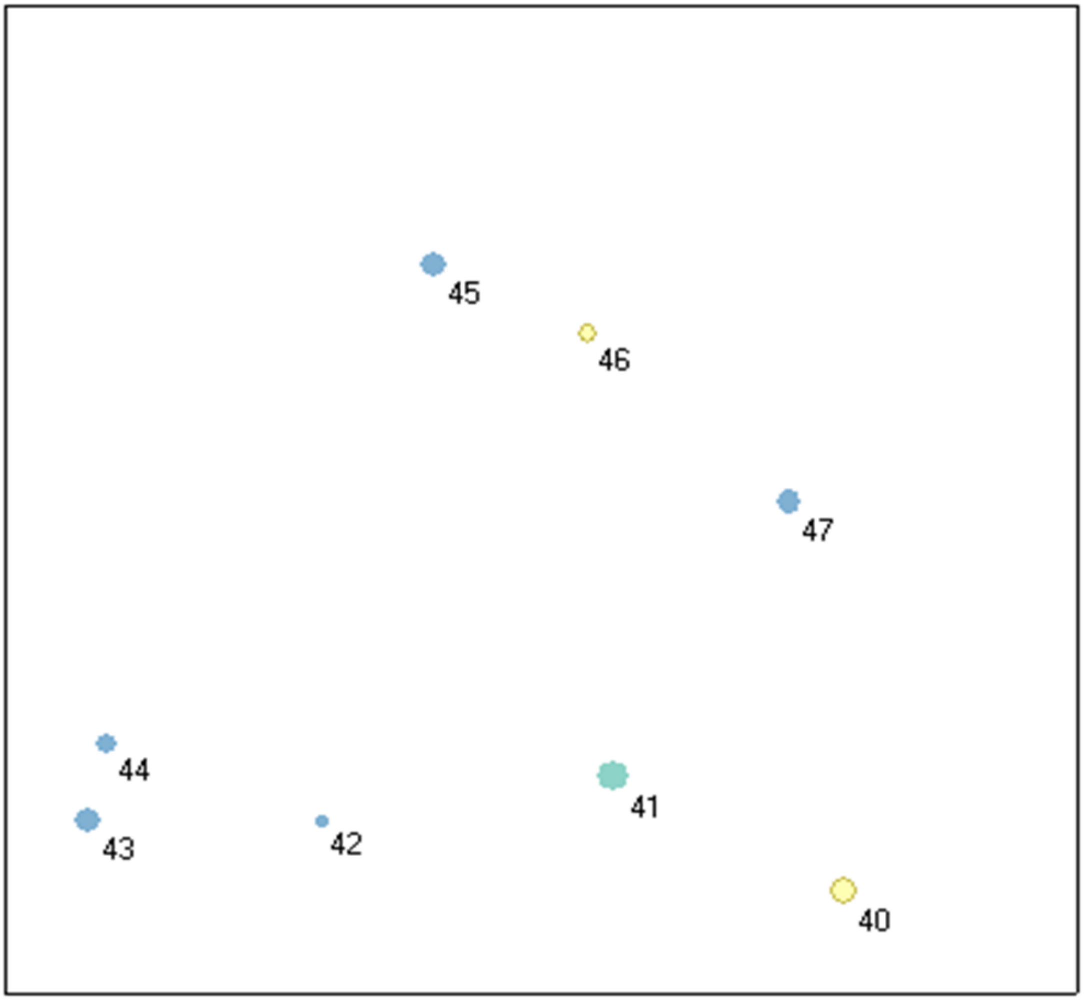


Figure A.8 Map of Hogsback Forest Plot 1 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.

	Acer saccharum
	Acer rubrum
	Betula alleghaniensis
	Fagus grandifolia
	Fraxinus americana
	Fraxinus nigra
	Fraxinus pennsylvanica
	Juglans cinerea
	Ostrya virginiana
	Pinus strobus
	Prunus serotina
	Quercus alba
	Quercus rubra

Hogsback Forest Plot 2- HB2

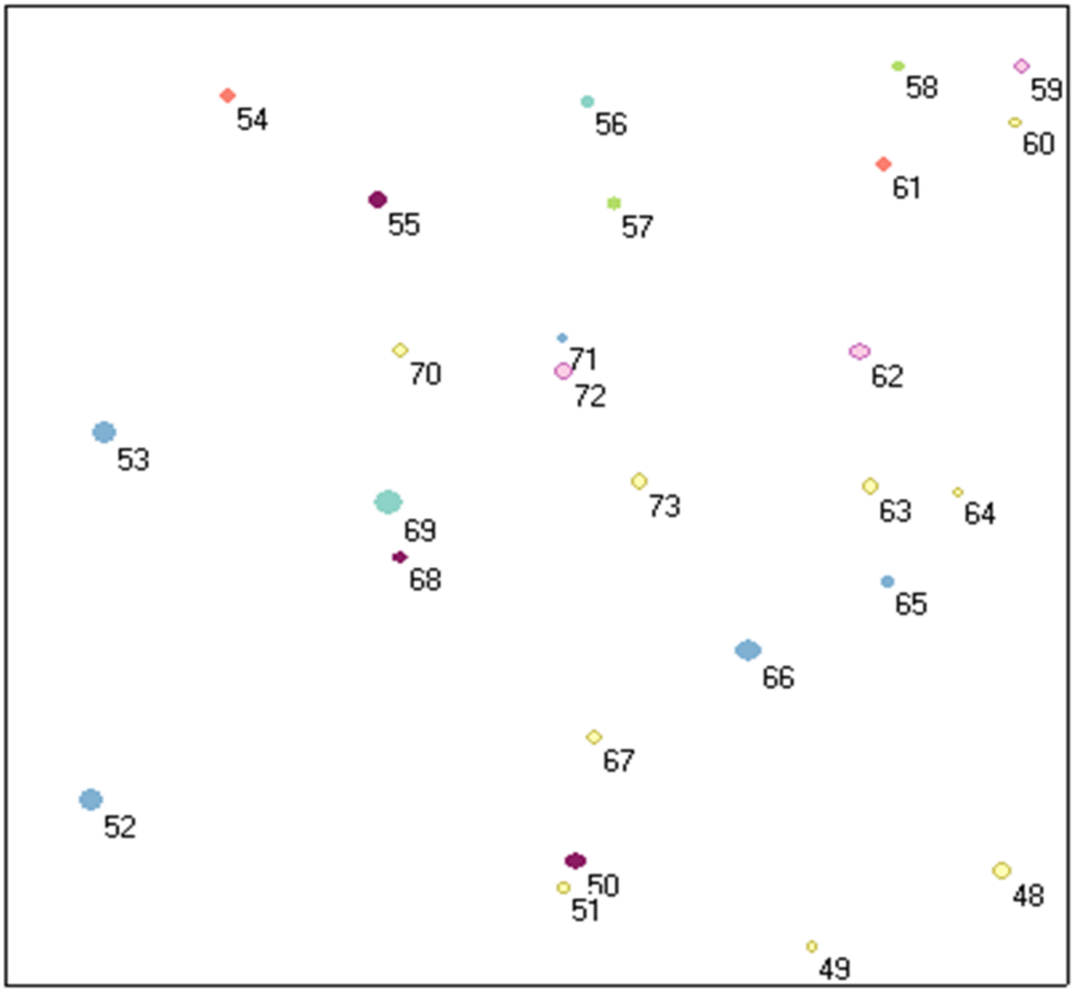


Figure A.9 Map of Hogsback Forest Plot 2 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.

	Acer saccharum
	Acer rubrum
	Betula alleghaniensis
	Fagus grandifolia
	Fraxinus americana
	Fraxinus nigra
	Fraxinus pennsylvanica
	Juglans cinerea
	Ostrya virginiana
	Pinus strobus
	Prunus serotina
	Quercus alba
	Quercus rubra

Hogsback Forest Plot 3- HB3

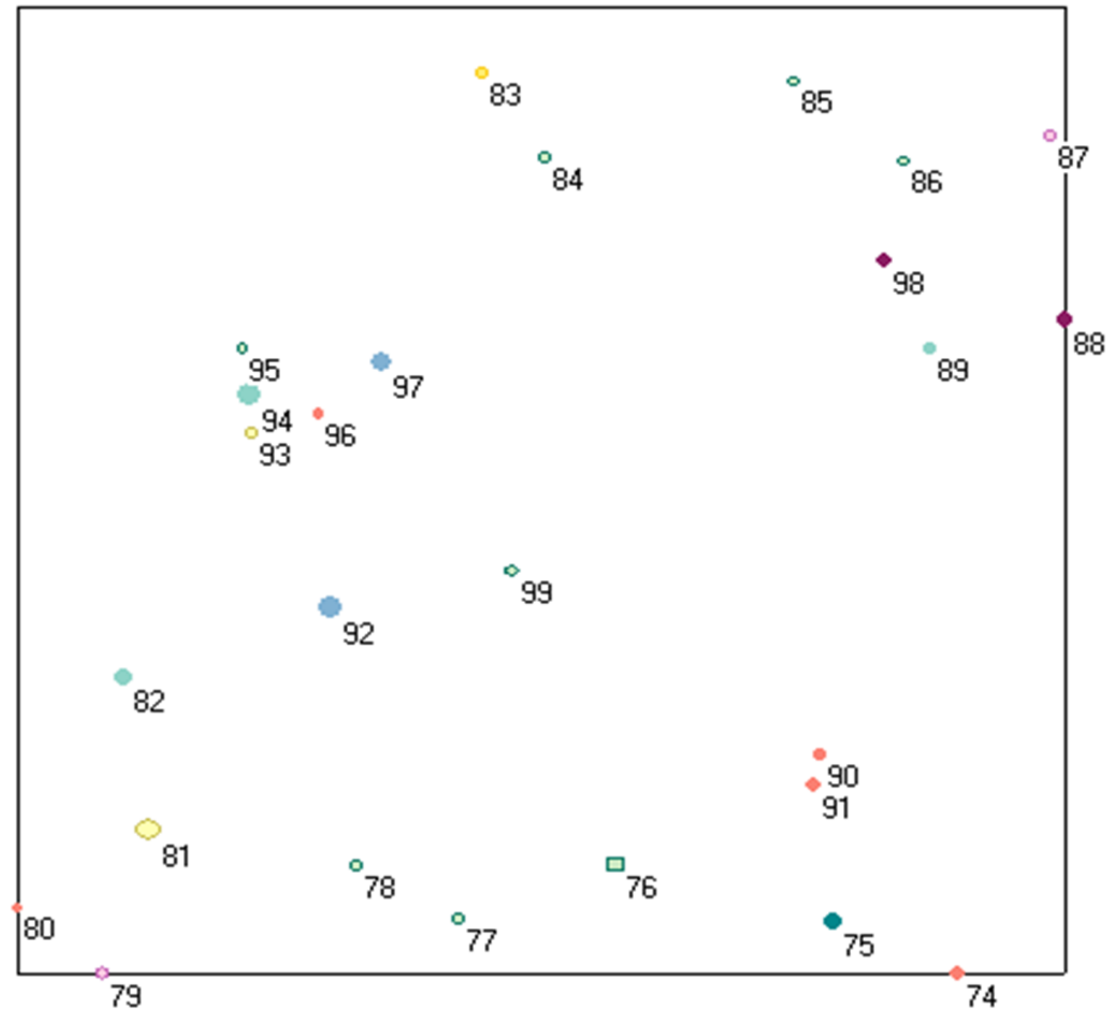
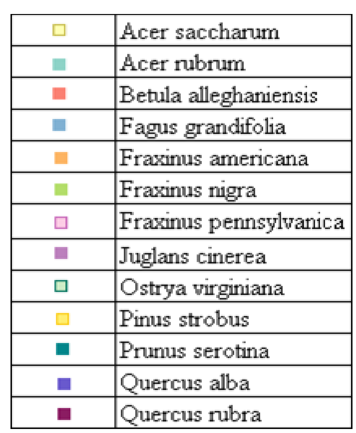


Figure A.10 Map of Hogsback Forest Plot 3 showing the location and species of all trees with dbh > 10.0cm standing within the plot. Point size increases with tree dbh, and the top left corner of the map represents the northwest corner of the plot.



Appendix B: Equipment Lists and Field Sheets

Salamander Monitoring Equipment List

- Clip board with field sheets A and B, blank paper
- Several pencils and a permanent marker
- Nitrile gloves
- Kestral 3000 pocket weather station
- Soil moisture meter and screwdriver for calibration
- Soil thermometer
- Digital calipers
- Ruler
- Digital pocket scale (give make)
- Pesola spring scale
- Small clear plastic bags
- Salamander measuring container (clear sandwich container fitted with moist sponges)
- Salamander holding container (large container with moist sponges)
- Bottle of pond water
- Flagging tape
- Utility knife
- Aluminum tags for ACOs if needed
- Camera

Soil pH Testing Equipment List

- Small containers or plastic sandwich bags for sample collection (24 for IW, 12 for HB)
- Trowel
- Spoon
- Nitrile gloves
- Soil pH testing kit

Forest Canopy Tree Monitoring Equipment List

- Clipboard with EMAN field data sheet and health data sheet – also bring previous year's data sheets and plot maps to refer to any notes or comments
- Several pencils and permanent markers
- Flagging tape
- 2 tape measures (30 m)
- Tree identification manual
- Binoculars
- Clinometer
- A few pre-labelled tags and steel pigtails for new trees
- Utility knife

Decay Rate Monitoring Equipment List

For decay stick extraction:

- Nitrile gloves
- Trowels
- Scissors or utility knife
- Envelopes or plastic bags for storing sticks individually
- Butter knives

For decay stick installation:

- Nitrile gloves
- Clipboard with decay rate field data sheet
- Shovel
- Trowels
- Tool with name I don't know
- 1 pigtail per ADR station (3 per forest plot)
- Pre-weighed, bagged and tagged tongue depressors
- Fishing line

Table B.1 Locations of Key Documents on the *rare* Server

All folders are located on Level 4 > Research and Monitoring > Ecological Monitoring

File (s)	Folder
Plethodontid Salamander Monitoring Protocol	EMAN Documents
Terrestrial Vegetation Monitoring Protocol	EMAN Documents
Tree Health Protocol	EMAN Documents
Soil Decomposition Draft Protocol	EMAN Documents
Salamander Field Data Sheets 2006 -2010	Salamanders
Forest Plot Field Data Sheets (Inventories) 2009 - 2010	Forest Health
Soil Humus Decay Rate Data Sheets 2009-2010	Soil Humus Decay Rate

Table B.2 Beaufort Wind Codes from 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 B.3 Beaufort Sky codes adapted from Zorn *et* al. (2004).

Sky Code	Description
0	Clear. No clouds at any level
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 and rain mixed
8	Shower(s)
9	Thunderstorm(s)

Plethodontid Salamander Monitoring Field Sheet A

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	ACO Age	Soil Temp	Soil Moisture	ACO Disturbance
Additional Comments:							

	North Perimeter		**East Perimeter**		**South Perimeter**		**West Perimeter**	
ACO #								
WS (mph)								
RH (%)								
AT (°C)								

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

Plethodontid Salamander Monitoring Field Sheet B

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:							

Forest Canopy Tree Biodiversity Monitoring Field Sheet

Canopy-Tree Sample: Field Data Sheet (20m x 20m stand-alone quadrats) Stand name: Date:

Stand Location: Plot no.: Avg. 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

Forest Canopy Tree Health Monitoring Field Sheet

Tree Condition Data Sheet

Site Name:	Date:	Observation Area Name and Description
Stand Location (lat/long):	Nearest Named Place:	
County/Township:	Province:	
Observer(s):	Observer Address: Telephone:	

Tree #	Species	dbh (cm)	Tree Status	Stem Defect 1		Stem Defect 2		Crown Class	Crown Rating	Comments
				Type	Location	Type	Location			

Annual Soil Humus Decay Rate Monitoring Field Sheet

Forest Plot ID:

Forest Plot Location (lat/long from NW corner):

Observer(s):

Stand	Plot	ADR Station	Tag #	Original weight (g)	Placement (s/ b)	Humus depth (cm)	Buried depth (cm)	Date Buried	Date Retrieved	Decayed weight (g)	% Stick Missing	Comments