

**Sexual segregation and comparative life history of Macoun's arctic
butterfly**

by

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ABSTRACT

SEXUAL SEGREGATION AND COMPARATIVE LIFE HISTORY OF MACOUN'S ARCTIC BUTTERFLY

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Macoun's arctic (*Oeneis macounii*) is a biennial satyrine butterfly found predominantly in boreal forests of North America. I monitored populations of *O. macounii* in Riding Mountain National Park (RMNP) in 2011 and in Sandilands Provincial Forest in 2012. I captured, sexed, marked and took UTM coordinates of butterflies to compare population sizes, sex ratios, longevity, dispersal and behaviour. I measured habitat variables around capture sites at RMNP, to establish environmental characteristics associated with butterfly occurrence. Population estimates at Sandilands show that the population size there is robust, but could not be calculated at RMNP. The number of butterflies observed at RMNP suggests that the population there is small and localized. In RMNP, males were more often in forested sites with sunny clearings, while female encounter sites were dominated by grasses, suggesting sexual segregation. The data collected from this survey is important for conservation strategies for the declining population at RMNP.

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CHAPTER 1

GENERAL INTRODUCTION

1.1 General Butterfly Background and Insect Life History

Butterflies are large, charismatic, and arguably the most popular of all invertebrates to the general public. Increasing interest in butterfly gardening, watching and conservatories all suggest that many people are interested and captivated by these showy insects. Because butterflies are generally easy to mark and observe, they also serve as excellent organisms for ecological and behavioural studies (Gilbert & Singer, 1975; Boggs *et al.*, 2003). They are also recognized as strong predictors of biodiversity and climactic change, as they are acutely sensitive to environmental change (Beccaloni & Gatson, 1995; Parmesan, 1996, 2003; Roy *et al.*, 2001; Thomas, 2005). Some endangered and charismatic “umbrella” species of butterflies, when protected, extend benefits to other species in the protected areas where they reside (New, 1997).

Life strategies vary greatly across related insect taxa. For example, fruit flies on the Hawaiian islands have evolved from a few colonizing individuals into hundreds of species that exhibit extensive morphological as well as behavioural differences (Carson & Kaneshiro, 1976). Also in Hawaii, some geometrid moths, a taxon with herbivorous larvae on the mainland, have evolved into highly modified predators of other arthropods (Montgomery, 1983). These examples highlight dramatic differences that can develop between species. More subtle differences in life history strategies, both between and within species, are widely encountered

(Stearns, 1989; Saether & Bakke, 2000). Intraspecific variation in traits frequently arises when gene flow between populations is restricted (i.e., the populations are geographically, temporally, or behaviourally isolated from each other). Once gene flow is restricted, the respective populations may encounter different selective environments due to differing physical conditions, social conditions, or a combination of both. Differential selection can result in adaptations that influence demography and life history traits of populations (Saether & Bakke, 2000; Rader *et al.*, 2005).

1.2 Introduction to the Genus *Oeneis* and to *Oeneis macounii*, Macoun's arctic

Butterflies in the genus *Oeneis* (Nymphalidae: Satyrinae), commonly known as the 'arctics', are a cryptic group of medium to large-sized butterflies found in arctic, alpine, boreal, and prairie biomes. Twelve species are found in North America, with many more being present in the Old World (Opler & Wright, 1999; Layberry *et al.*, 2001; Brock & Kaufman, 2003). *Oeneis* species are poorly studied and not often seen, as they are dark and muted in colour and their wings have cryptic undersides that make them nearly impossible to detect when at rest (Layberry *et al.* 2001). It is generally accepted that most, if not all, species in this genus have a biennial life cycle, taking two years to develop from egg to adult. Because of this biennialism, some populations occur as adults only in even-numbered years or only in odd-numbered years. This is most evident in North America in Macoun's arctic (*Oeneis macounii*).

Oeneis macounii (Figure 1.1) is found almost exclusively in Canada, ranging from central British Columbia to western Quebec, and in the United States in northern Minnesota and

Michigan. In contrast to most other arctic species in North America, *O. macounii* is a boreal forest species, usually found in association with mature jack pine stands (Masters, 1972; Opler & Krizek, 1984; Layberry *et al.*, 2001). Like other species in the genus, *O. macounii* exhibits a biennial life cycle, occurring as adults in even-numbered years in the eastern part of its range and in odd-numbered years across central Canada. The divide between the eastern even-year and central odd-year populations is in central Manitoba (Figure 1.2; Masters, 1974).



Figure 1.1. *Oeneis macounii*, the Macoun's arctic butterfly, showing a field identification number from this study on the hind wing.

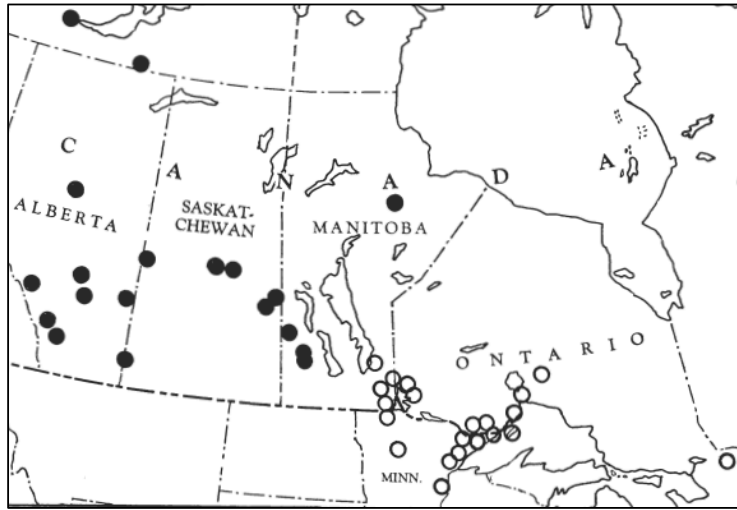


Figure 1.2. Distribution of *Oeneis macounii* in Canada and the United States, as shown by J.H. Masters (1974, *Journal of the Lepidopterist's Society* 28: p. 239.) Odd-year populations are depicted with a solid black circle, even-year populations are denoted by a open circle. Note the transition between odd-year and even-year populations in south-central Manitoba.

Within Manitoba, even-year populations of Macoun's arctic are common in the extensive mature jack pine forests broadly distributed in Sandilands, Wampum, Northwest Angle, and Agassiz Provincial Forests in the southeastern corner of the province (Klassen *et al.*, 1989, Elliott, 1997). This region is characterized as the Lake of the Woods section of the Manitoba Lowlands (Manitoba Wild Lands: Manitoba Land Use Planning). Sandilands Provincial Forest (hereafter referred to as "Sandilands") covers nearly 3000 km². Soils are comprised mainly of medium-textured sand (Sims, 1975), with numerous eskers left behind by the retreat of the most recent glaciers (Saylor, 1975). The upland areas of Sandilands north through Belair Provincial Forest are dominated by jack pine stands (Saylor, 1975) where *O. macounii* is common in sites with trees >25 years old (Elliott, 1997; G.W. Otis, pers. comm.). Populations of *O. macounii* are extensive in area and in close proximity within this region.

In western Manitoba, the odd-year population of central Canada reaches its most southeasterly limit in Riding Mountain National Park (“RMNP”). RMNP is located in the Western Upland region of the province (Manitoba Wild Lands: Manitoba Land Use Planning) that is higher in elevation than the surrounding farmlands due to the presence of the Manitoba escarpment. The Upland region is characterized by dead-ice topography, where a large block of ice from the Wisconsin glacier remained after the retreat of the main glacier; as it melted it left unsorted moraines and lakes (Field Studies Council). The population of *O. macounii* is isolated from the closest recorded population approximately 80 km to the northwest in the Duck Mountains. In addition, the habitat where it has been recently recorded within RMNP is different from the “typical” mature jack pine habitat (Opler & Krizek, 1984; Layberry *et al.*, 2001): patches of prairie surrounded by mixed forests of aspens, spruce, and old jack pines (Otis, 2010). Numerous dense young jack pine stands resulting from a number of forest fires in the last century populate sandy glacial deposits in the eastern portion of RMNP (Caners & Kenkel, 2003), but there are no known sites with open, mature jack pine forest such as those in Sandilands where Macoun’s arctics are common (G.W. Otis, pers. comm.). RMNP is a popular tourist location with a town site located within the park borders. Numerous visitors hike the Brulé Trail that traverses the study site.

I studied the even-year population of *O. macounii* in Sandilands and the odd-year population in RMNP. They are temporally and geographically separated (Figure 1.3) from each other and inhabit seemingly different habitats, as briefly described above. This separation prevents gene flow between the eastern and central populations of this species. Habitat differences in the two study sites may have created different environments that, when coupled with differences in natural or even sexual selection, may have resulted in differences in life history and demography of the butterflies, such as mating strategy, population size and adult movements. In Chapter 2, I explore this concept by quantifying and comparing population size, survival, dispersal, sex ratio, and adult behaviour of populations of Macoun's arctic in these two study sites.



Figure 1.3. Locations of the two field sites, the prairie site on the Brulé Trail, Riding Mountain National Park and the Lonesand town site, Sandilands Provincial Forest. Map courtesy of canada-maps.org.

1.3 Conservation of Macoun's arctic in Riding Mountain National Park

Historically, *O. macountii* may have declined in abundance in Riding Mountain National Park (RMNP) (Figure 1.4), and vicinity, Manitoba, as inferred from museum records (Figure 1.5). Data compiled from museum specimens show that they were collected in RMNP every year between 1932-1939. More significantly, the even-year population seems to have always been less common than the odd-year population (only 10 even-year records vs. 121 odd-year records prior to research by G. Otis and myself). Having not been collected since 1968 despite searches by G. Otis in 2008, 2010, and 2012, it is probably extirpated. However, it is difficult to use museum records to extrapolate patterns of abundance due to differences in sampling effort among collectors. However, the decline and disappearance of the even-year population at RMNP suggests that the odd-year population also may have declined over time and could also be at risk.

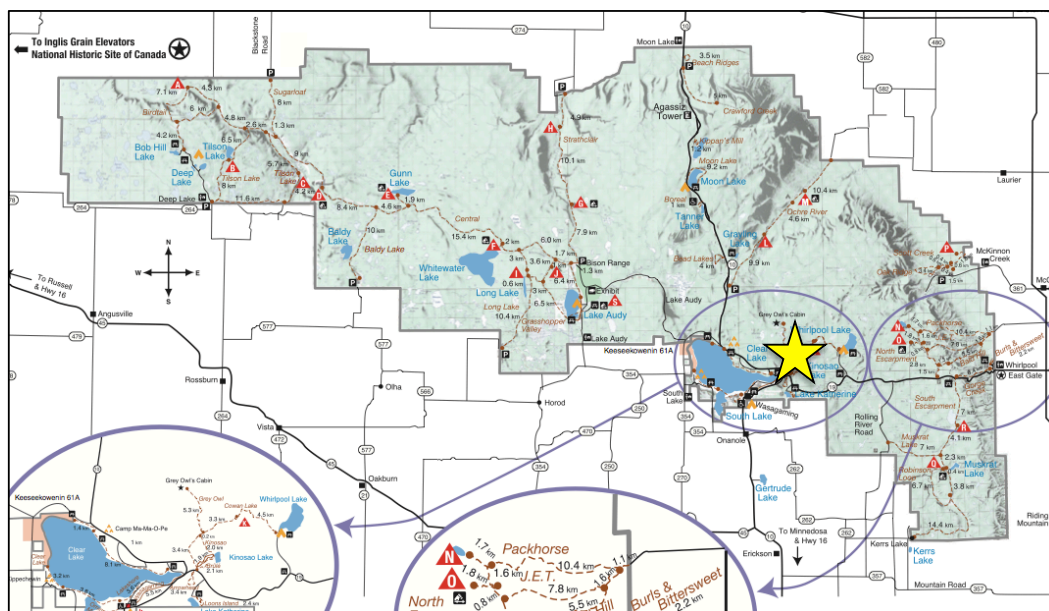


Figure 1.4. Map of RMNP with location of field site within the park marked with a star. Map courtesy of Parks Canada.

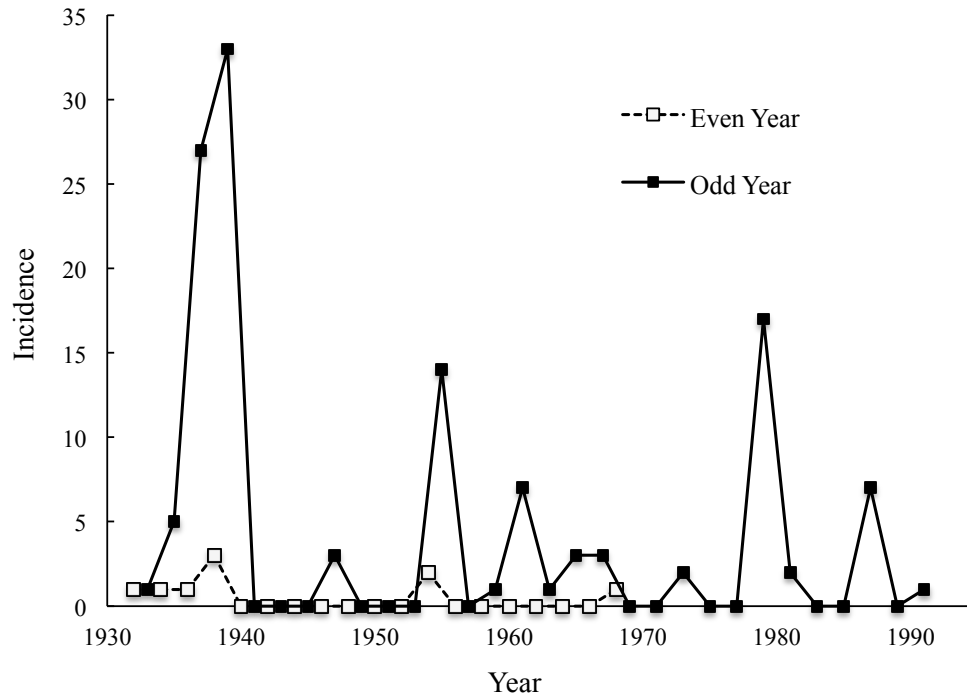


Figure 1.5. Historical museum record collection of *O. macounii* in Riding Mountain National Park. The even-year population within the park, last seen in 1968, may be extirpated. Records were obtained from the following collections: University of Manitoba; Peabody Museum; Yale University; McGuire Center for Lepidoptera and Biodiversity, University of Florida; Manitoba Museum of Man and Nature, Winnipeg; Carnegie Museum, Pittsburgh; American Museum of Natural History, New York; U.S. National Museum, Washington, D.C.; Milwaukee Public Museum; Royal Ontario Museum, Toronto; Canadian National Collection, Ottawa; Toronto Entomologists' Association records.

If conservation efforts are initiated to increase populations of Macoun's arctic in the park, detailed information about its life history, ecology and habitat requirements will be needed. For *O. macounii*, information on population size, mating behaviour, movements, and larval host plant are not currently available.

One of the objectives of this project was to more thoroughly examine the conservation status of *O. macounii* at RMNP. In Chapter 2, life history parameters are compared between the populations at RMNP and at Sandilands. In Chapter 3, I quantify the distribution and abundance of Macoun's arctic in the park. I identify and discuss differences I discovered in microhabitats occupied by males and females. Finally, I explore the implications that this sexual segregation has on the conservation of the small population of this species at Riding Mountain National Park.

In the final Chapter 4, I summarize key results of the study and discuss implications of my research.

CHAPTER 2

COMPARATIVE POPULATION BIOLOGY AND ADULT BEHAVIOUR OF THE MACOUN'S ARCTIC BUTTERFLY (*OENEIS MACOUNII*: SATYRINAE) IN TWO MANITOBAN POPULATIONS

2.1 ABSTRACT

Macoun's arctic (*Oeneis macounii*) is a biennial satyrine butterfly found in boreal forests in northern North America. Populations that are isolated from one another or in different habitat types sometimes show varying life history traits. I monitored populations of *O. macounii* in Riding Mountain National Park (RMNP) in 2011 and in Sandilands Provincial Forest of southeastern Manitoba in 2012, where the butterfly is found in two distinctly different isolated physical settings in order to obtain information about life history traits, movements and behaviour of the species in these two locations. I captured, sexed, marked and took UTM coordinates of butterflies to quantify and compare population sizes, sex ratios, longevity, dispersal and behaviour. A logistic regression of sex ratio by week showed that the ratio of males to females decreased significantly over the four-week flight at RMNP, indicating slight protandry in the population. Maximum recorded longevity was 10 days at RMNP and 9 days at Sandilands. Population estimates at Sandilands using Jolly-Seber mark-recapture analysis show that the population size there is robust. Population size could not be calculated at RMNP, but the small number of butterflies observed there in a specialized habitat suggests that the population there is small and localized.

2.2 INTRODUCTION

Macoun's arctic, *Oeneis macounii*, is predominantly a Canadian species, ranging from the interior of British Columbia to western Quebec (Layberry *et al.*, 2001). The northernmost-recorded populations live near Great Slave Lake in the southern Northwest Territories; the southernmost are found in northern Minnesota and Isle Royale, Michigan. With the considerable habitat variation over this extensive range, it is possible populations that live in different habitats will differ in some life history traits. The life history and ecology of this species are documented in the reports of Masters (1972, 1974) and Masters & Sorensen (1969). Unlike most other *Oeneis* species in North America that predominantly inhabit arctic and alpine zones, *O. macounii* is a boreal forest inhabitant (Masters & Sorensen, 1969). Populations in Manitoba typically inhabit mature jack pine forests growing on sandy soils (Masters, 1972; Opler & Krizek, 1984). In Sandilands Provincial Forest, which is characterized by this habitat type, populations of *O. macounii* can be large and occur over an extensive area. On the western side of the province an isolated population of *O. macounii* inhabits Riding Mountain National Park (RMNP). There it seems to be highly localized to a small prairie patch surrounded by aspen, spruce and jack pine forest (Otis, 2010), a habitat that is dissimilar from the mature jack pine forests that this species typically inhabits.

Oeneis macounii has a biennial life cycle, with individuals taking two years to complete their life cycle from egg to adult (Masters, 1974; Layberry *et al.*, 2001). Consequently, this contributes to the unusual phenomenon of adults occurring in Sandilands, and the rest of the eastern part of the range, exclusively in even-numbered years. By contrast, populations in

western Manitoba and across northern Alberta they are only present in odd-numbered years (Klassen *et al.*, 1989). Riding Mountain National Park historically had both even- and odd-year populations, but it seems that as of 1970, only the odd-numbered year population exists there (see Chapter 3).

In populations such as these that are isolated from each other in time or space, intraspecific variation of life history traits, including preferred habitat, sexual behaviours, dispersal, protandry (sex ratio), and phenology, can exist (Baur & Raboud, 1988; Blanckenhorn, 1991). This can occur as a consequence of genetic drift or founder effects, or both (Groeters & Dingle, 1987). However, life history variation can also occur due to environmental variation that creates different selection environments (Caswell, 1983; Fairbairn, 1984; Baur & Raboud, 1988, Rader *et al.*, 2005). Populations of the same species that are separated from each other often become adapted to local environmental conditions (Rader *et.al.*, 2005). Some reports show that the genus *Oeneis* can have interesting variation in life history and mating systems, where subspecies can behave differently (Warren, 2005). It is possible that populations of *O. macounii* show similar patterns of divergent behaviour.

In this study I compare population size, survival, dispersal, sex ratio, and adult behaviour of two spatially and temporally isolated populations of *O. macounii*. In Sandilands, southeastern Manitoba, this species is frequently encountered in 25+ year-old jack pine forests with frequent grassy clearings (Elliott, 1997). In Riding Mountain National Park, the other study site in western Manitoba, Macoun's arctic is uncommon in prairie enclaves surrounded by rather dense mixed boreal forest (Chapter 3). These two populations are temporally (even-

year vs. odd-year, respectively) as well as geographically isolated. By studying the aforementioned aspects of population biology and the behaviour of adults, I seek to uncover biological differences in these aspects between the populations and possible factors that underlie them.

2.3 METHODS

2.3.1 Study Areas

The study site in 2011 was located in Riding Mountain National Park (RMNP). The park is a large ‘wilderness island’ of boreal forest and prairies higher in elevation than the surrounding agricultural lands (Bronson & Noble, 2006). The study site was in the predominantly forested eastern portion of the park, and covered approximately 63 ha. This area was subjected to frequent burns in the early 1900s; a controlled burn along the Brulé Trail in 1971, and a large natural fire in 1980 (Caners & Kenkel, 2003). The area now has extensive stands of immature jack pine (*Pinus banksiana*). In some pockets that were untouched by fire, mature *P. banksiana* can still be found, along with open prairie enclaves that are slowly changing due to succession and lack of fire. Along the western portions of the Brulé trail (UTM: 433303, 5636816, zone 14U) there is a mosaic of open prairie patches set among relatively dense forests with a few mature jack pines, aspens, and spruces.

The study in 2012 took place near the Lonesand locality near the southern edge of Sandilands Provincial Forest, in southeastern Manitoba, Canada (UTM: 694562, 5454044, zone 14U).

Sandilands Provincial Forest covers nearly 3000 km² of low sandy hills interspersed by eskers left behind when the late Wisconsin glacier retreated approximately 10,000 years ago. It is close to Belair, Agassiz, and Wampum Provincial Forests, creating an extensive area of jack pine habitat where *O. macounii* is common. My study site consisted mostly of jack pine forest with numerous clearings, as well as minor shrubby and grassy portions, and was approximately 61 ha in size; it was surrounded by bogs and grazing land. The Lonesand Road bisected the site. There were also several small ATV tracks through the site that created small sandy openings within the forest. A slight hill ~250m long and with a maximum elevation difference of 4 meters from bottom to top was located in the east-central part of the site.

2.3.2 Sampling Design

One field assistant and I conducted surveys to catch *O. macounii* throughout both sites for the duration of its flight on days with appropriate weather (partly to completely sunny, > 18°C, wind speed < 20km/h, between 10:00-17:00h). The survey period in 2011 ranged from 1 June to 16 July (16 suitable sampling days); the 2012 sampling period ranged from 12 May to 17 June (17 suitable sampling days). The routes we walked were consistent from day to day, encompassing the entirety of suitable habitat and some of the surrounding area. Each route was traversed 3-5 times per day. In Sandilands, we did not include the boggy areas in the extreme southeast and northeast corners of the site where Macoun's arctic would be very unlikely to occur. When a butterfly was encountered, we captured it with an aerial net. Each individual was sexed by observing abdomen size (Figure 2.1) and presence or absence of

male genitalic claspers. We recorded GPS coordinates using a handheld Garmin Oregon 450t[®] device (accuracy <10m). We weighed butterflies to the nearest 0.001 g using a Mettler-Toledo 203E Precision Balance. Because of the dark ventral hindwing colouration, the butterflies were marked by first applying a dot of white Golden Fluids[®] acrylic paint on each hind wing; a unique number was then written with a Sigma Micron[®] 0.25mm pen on the paint dot for individual identification upon subsequent sightings or recaptures. We released every individual at the site of capture by gently removing it from the net and placing it on vegetation. Because the butterflies had cooled during handling, most remained at the release site.



Figure 2.1. Depiction of the difference in abdomen size in adult female (left) and male (right) *Oeneis macounii*. Females have larger and rounder abdomens when compared to males.

I also recorded all butterfly species encountered in both sites before and during the *O. macounii* flight to establish its phenology relative to other species. At RMNP records continued for three days after the last observation of *O. macounii*; at Sandilands, I ended observations when only two butterflies were caught and several days of rain were forecasted. Additionally at RMNP, flowers in bloom before, during, and after the flight period of Macoun' arctic were noted (see Appendix 1).

2.3.3. Data Analyses

Sex ratios at both sites were analyzed using linear regression to determine if there were changes over the flight period in the proportion of males captured. In order to have adequate sample sizes to estimate the percentages of male, I combined numbers of individuals caught into two-day intervals. Sample dates with <5 individuals captured were not included in the analysis because of the high stochastic variability that results from extremely small samples. This analysis would show if males are protandrous and emerge prior to females, a common phenomenon in insects. These tests were conducted using SAS version 9.2 (SAS Institute, Cary, NC).

Locations of captures of both males and females were plotted and calculations of straight-line recapture distances were made using the measure tool in ArcGIS (ESRI). For the base map at RMNP, I used an orthorectified image from 2007 air photos provided by Riding Mountain National Park, Parks Canada, and the Manitoba Land Initiative. Base maps for the Sandilands site were all obtained from the Manitoba Land Initiative. I compared recapture distances

between sexes in each site and between sites. I used Satterthwaite's t-tests, as recapture distances between groups did not meet the assumption of equal variances, and within-group variances needed to be used. I calculated densities of male butterflies by estimating the area of the study site that was searched with the Area Measure tool in ArcGIS, and dividing the number of male butterflies by the estimated area.

2.3.4 Mark-Recapture Analyses: Model Selection and Parameter Estimation in MARK

I estimated daily population sizes (N_i) and total population size (N_{tot}) for 2012 data using the population analysis software MARK (Vers. 7.0, G.C. White, 2009, Colorado State University, Fort Collins, CO). I used The Jolly-Seber method, with POPAN parameterization that allows for emigration, immigration, births and deaths. Jolly-Seber models are used to estimate population size in mark-recapture studies, and have been used on insects in the past (e.g., Daily *et al.*, 1991; Tikkamaki & Komonen, 2010; Zimmerman *et al.*, 2009). Other underlying assumptions of the Jolly-Seber models were equal catchability of marked and unmarked individuals, that animals retained their tags throughout the field season, and that survival was constant between marked and unmarked individuals throughout sampling occasions.

The analysis generated estimates of three parameters: ϕ (phi), the probability of survival between sampling occasions; p , the probability of recapture between sampling occasions; and b , the probability of new individuals entering the population from a super population between sampling occasions. I conducted the analysis with all three of these parameters being either

time-dependent (t), or fixed (.). I adjusted the model for inclement weather days by manually inputting days between sampling occasions. I assessed the outputted estimates derived from the different models by comparing Akaike's Information Criterion (AIC) and confidence intervals of population estimates. Lower AIC values help choose the best-fit model that is not over-parameterized.

2.4 RESULTS

2.4.1 Riding Mountain National Park – 2011

General Observations

At RMNP, the *O. macounii* flight began on 18 June 2011, and lasted for 25 days. The pattern of male and female captures is plotted in Figure 2.2. Of the 34 females and 30 males captured, only 14.3% and 23.3% were recaptured, respectively (Table 2.1). There was no difference in the number of males and females captured ($\chi^2 = 0.25$, d.f. = 1, $p = 0.62$). The density of males across the entire study area was approximately 0.5/ha. The longest period between recaptures for males and females was 7 and 10 days, respectively (Table 2.1). There was no difference in recapture distances between the sexes ($t = 0.51$, $df = 19$, $p = 0.62$; Figure 2.3a). The shortest distance between recapture instances was 4m; the longest was 346m (recapture distances are +/- 20m due to GPS error). These movements were both made by males. Males had a mean weight of 0.123g (se = 0.007; $n = 30$); females had a mean weight of 0.170g (se = .009; $n = 34$).

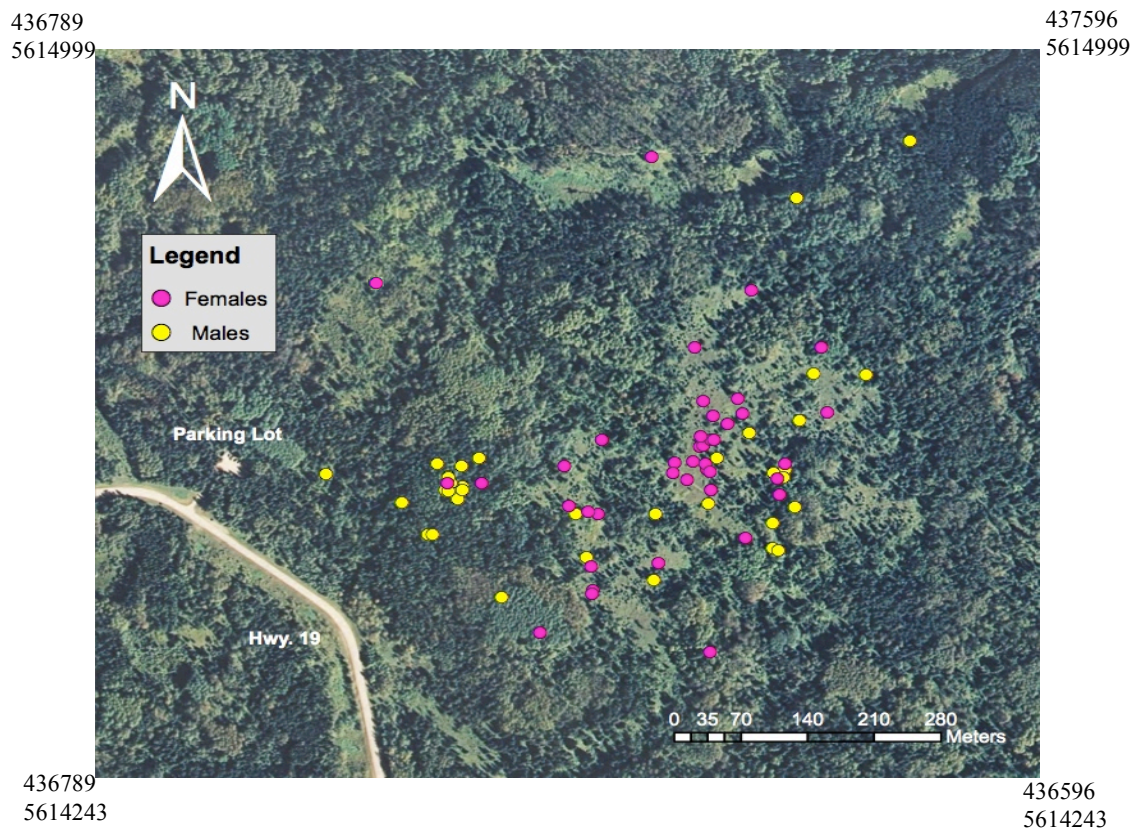
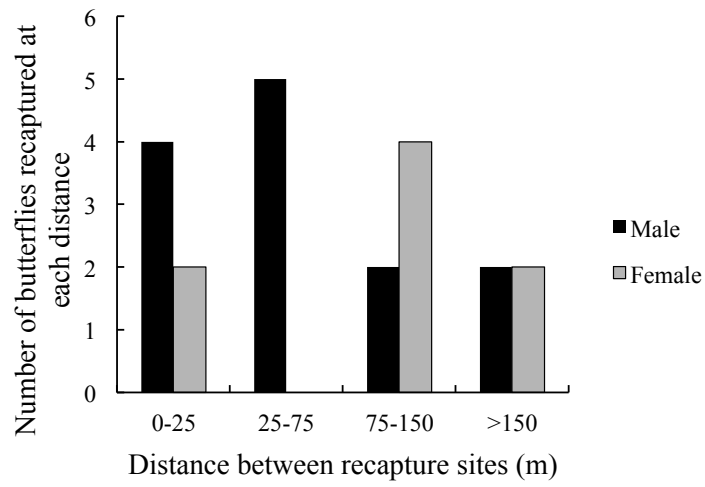


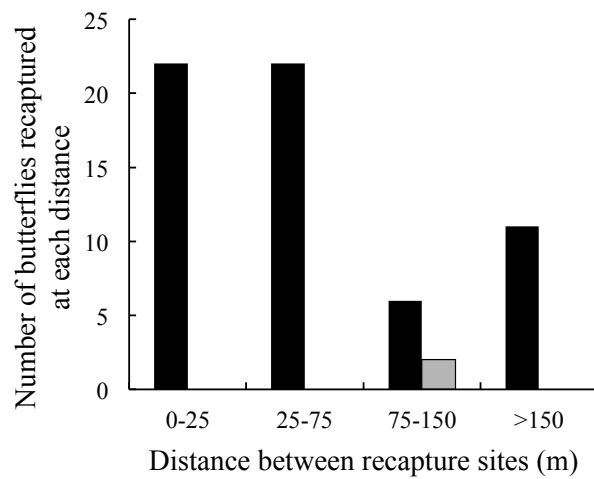
Figure 2.2. Locations of male and female butterflies in the RMNP site on the Brulé Trail. The light green areas represent the grassy prairie habitat; dark green areas are forested. Ortho photo courtesy of Parks Canada. Numbers outside the map indicate UTM coordinates of the corners of the map.

Table 2.1. Summary of capture data from two study sites in Manitoba. The recapture rates of males in both sites were similar, but female recapture rate was lower in Sandilands.

	Individuals captured	Density/ ha	Individuals recaptured	Proportion recaptured (%)	Capture events*	Max. residency (days)	Recapture distances (m)			
							Mean	se	Min.	Max.
RMNP										
<i>Males</i>	30	0.5	7	23.3	39	7	85	±30.9	4	345
<i>Females</i>	34		5	14.3	40	10	105	±22.9	13	205
Sandilands										
<i>Males</i>	176	2.9	39	22.2	231	9	94	±20.3	0	856
<i>Females</i>	45		2	4.4	47	4	109	±28.2	80	137
* Capture events are greater than total of individual captures and recaptures due to some individuals being recaptured several times over the flight period										



(a) RMNP



(b) Sandilands

Figure 2.3. Frequency chart of recapture distances of males and females in both field sites.

Population Parameters

Because so few individuals were captured at RMNP, the sample size was too small for formal analysis. Therefore the mark-recapture data were not analyzed with MARK.

Sex Ratio

The percentage of butterflies caught during the entire flight period that were male was 47% (30 males, 34 females). Sex ratio declined significantly over the four weeks of flight, from male biased at the beginning of the flight period to female biased at the end of the flight. Slope of the linear regression line differed significantly from zero (Figure 2.4).

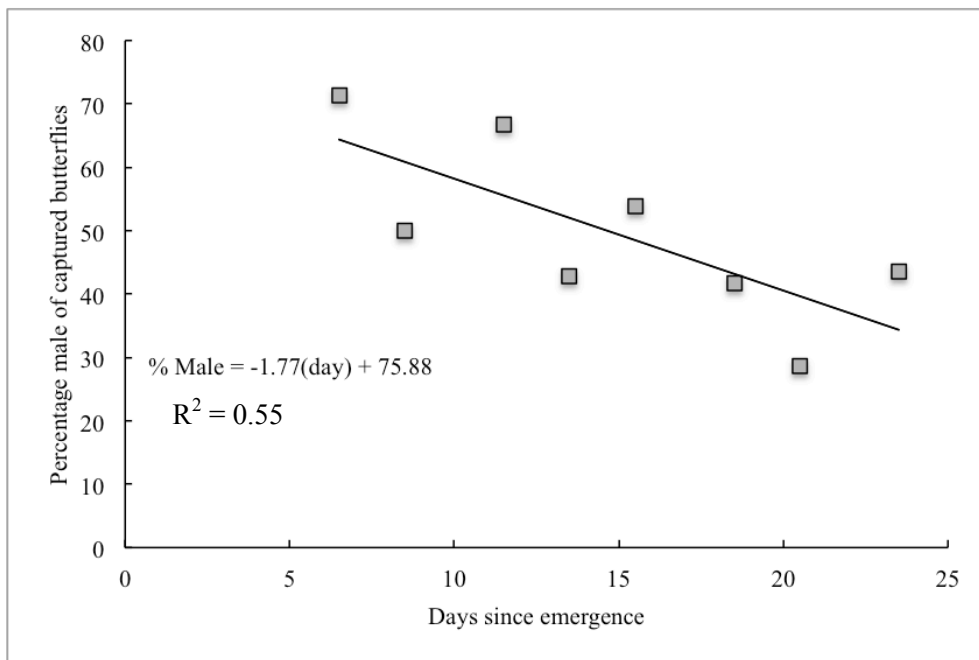


Figure 2.4. Proportion of captured butterflies that were male during *Oeneis macounii*'s flight at Riding Mountain National Park. The species is moderately protandrous, with a male biased sex ratio at the beginning of the flight. The slope differed significantly ($p < 0.05$) from zero.

Butterfly Phenology

Figure 2.5 shows the phenology of the 24 species of butterflies encountered at the RMNP site. Red-disked alpine (*Erebia discoidalis*) were abundant when I first arrived at the site, and their flight was almost at an end when *O. macounii* were first observed. The flight of white admiral (*Limentis arthemis*), a large conspicuous species, began almost exactly half way through *O. macounii*'s flight, and persisted throughout the second half. Finally, two days before the last sighting of *O. macounii* in 2011, northern pearly eyes (*Enodia anthedon*) first appeared.

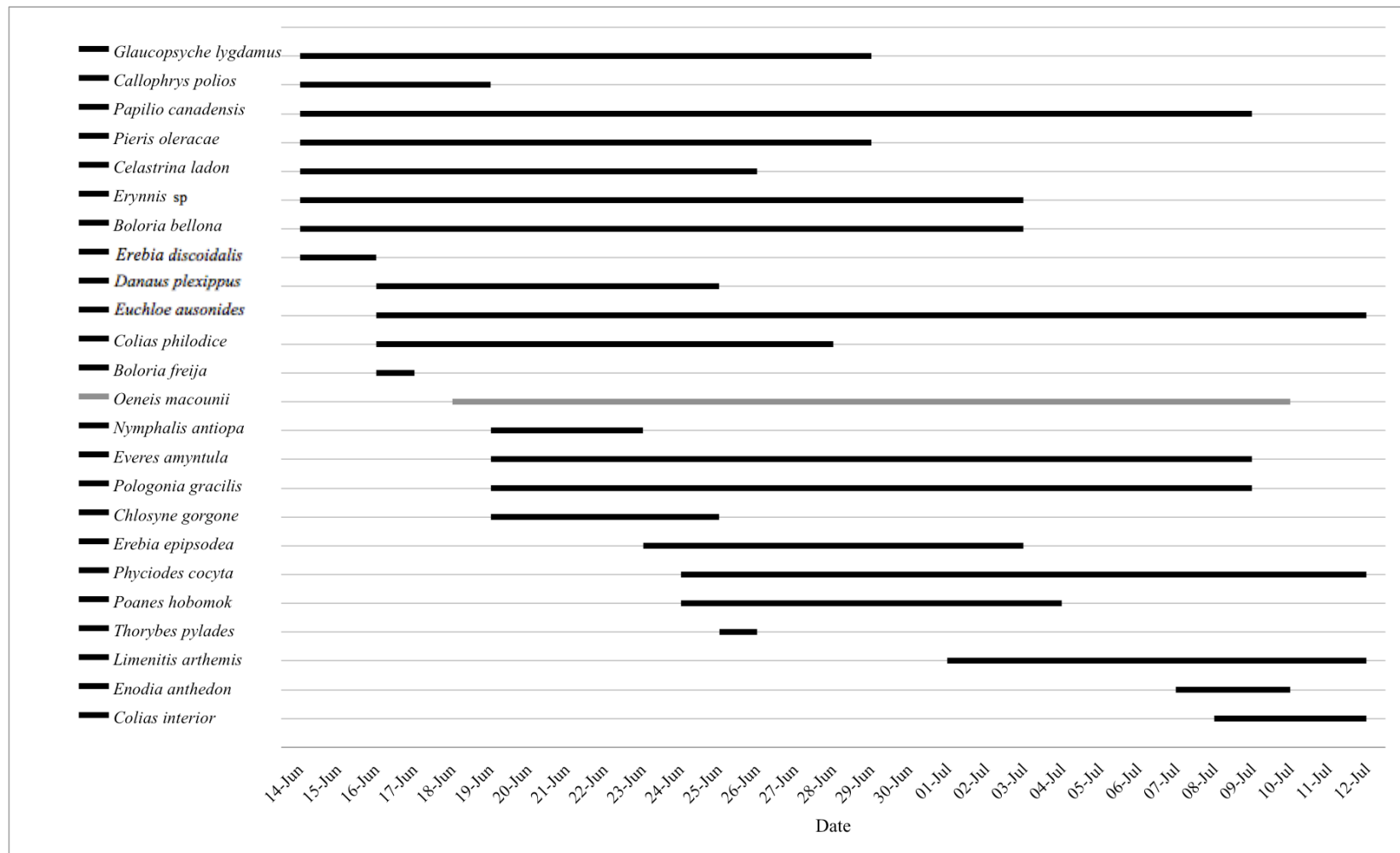


Figure 2.5. Phenology of butterfly species observed at the Riding Mountain National Park field site before, during and after the *O. macounii* flight period.

2.4.2 SANDILANDS PROVINCIAL FOREST - 2012

General Observations

The *O. macounii* flight at Sandilands started on 31 May 2011 and lasted 27 days (Figure 2.6). The number of male and female butterflies captured was not equal ($\chi^2 = 7.03$, d.f. = 1, $p < 0.01$). Of the 221 butterflies that were captured at the Sandilands site, the numbers recaptured also differed by sex: 39 of 176 males captured compared to only 2 of 45 females captured. Male density was estimated to be 2.9 butterflies/ha over the entire site, but density in the two main clusters of butterfly occurrence was 28.9/ha in the more northern site and 66.3/ha in the site to the south of the road. Male and female recapture points are plotted in Figure 2.7. There was no difference in recapture distance (Table 2.1) between males and females ($t = 0.42$, $df = 2$, $p = 0.70$). Males in general were often encountered within 75m from the original capture point (Figure 2.3b, 2.8). The overall minimum distance between recapture sites of all butterflies was 0m, and the maximum was 856m (Table 2.1), which were both traversed by males. Of 39 recapture events of males, only 5 crossed the sandy road (12.8%) (Figure 2.8). The maximum duration between recaptures was 9 days for males and 4 days for females (Table 2.1). A major storm with hail and heavy rains occurred 18 days into the flight period. This storm caused a notable decline in butterfly captures and recaptures (Figure 2.9). Specifically, I noted a loss of most worn individuals in the population. In Figure 2.9, the last day of the flight period shows a large increase in percentage recaptured. This is because only two individuals were caught on the last day, one of which was a recapture.

Males had a mean weight of 0.105g (se = 0.001; n = 176), whereas females had a mean weight of 0.181g (se = 0.007; n = 45).

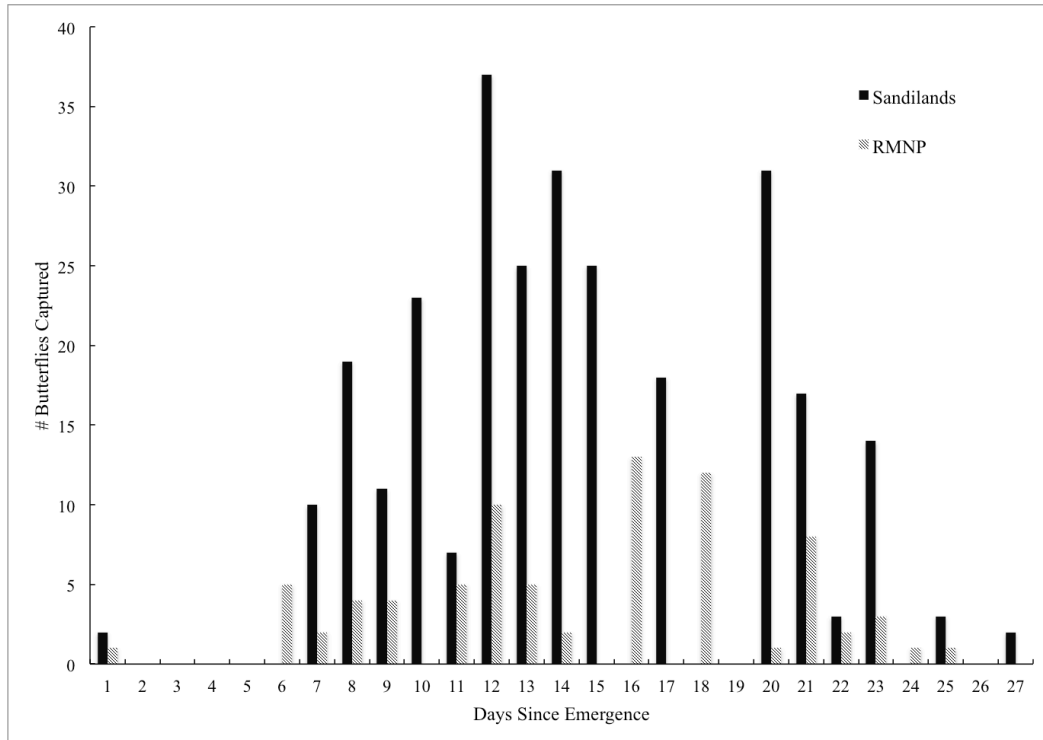
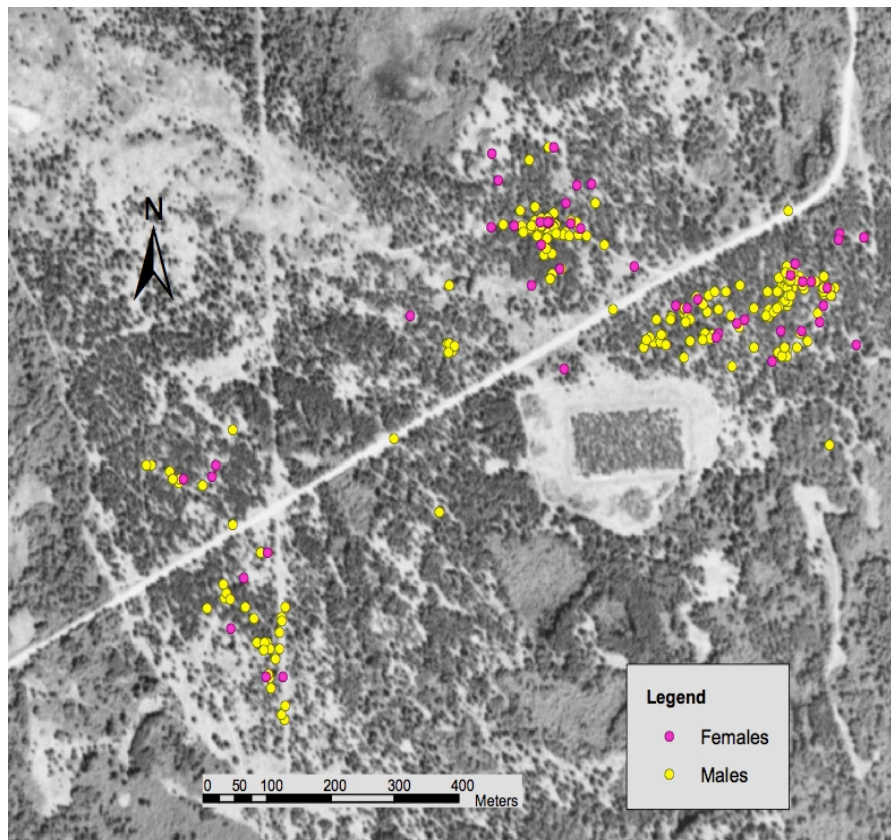


Figure 2.6. Daily captures of *Oeneis macounii* in both Manitoba field sites, Riding Mountain National Park (2011) and Sandilands Provincial Forest (2012). Data have been standardized to the day of initial sighting (18 June, 2011, at RMNP; 31 May, 2012, at Sandilands).

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Figure 2.7. Map showing locations of male and female captures in the Sandilands site. Ortho photo courtesy of the Manitoba Land Initiative.

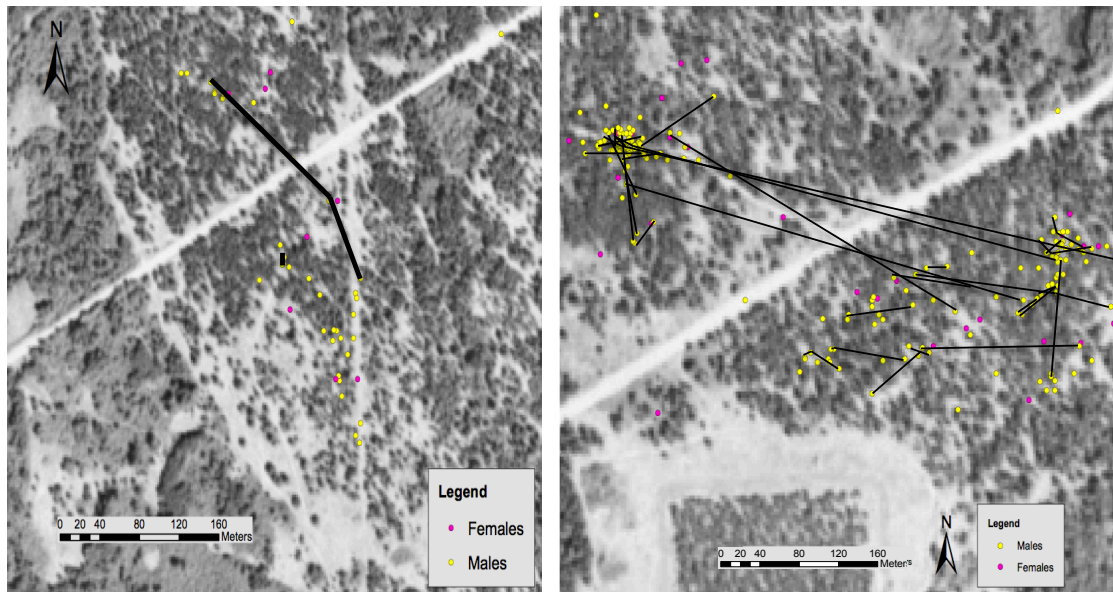


Figure 2.8. ‘Zoomed’ view of the two regions with most of the capture locations at the Sandilands site. Lines are drawn between recapture points. Only male recapture lines are pictured; only 2 females were recaptured at this field site.

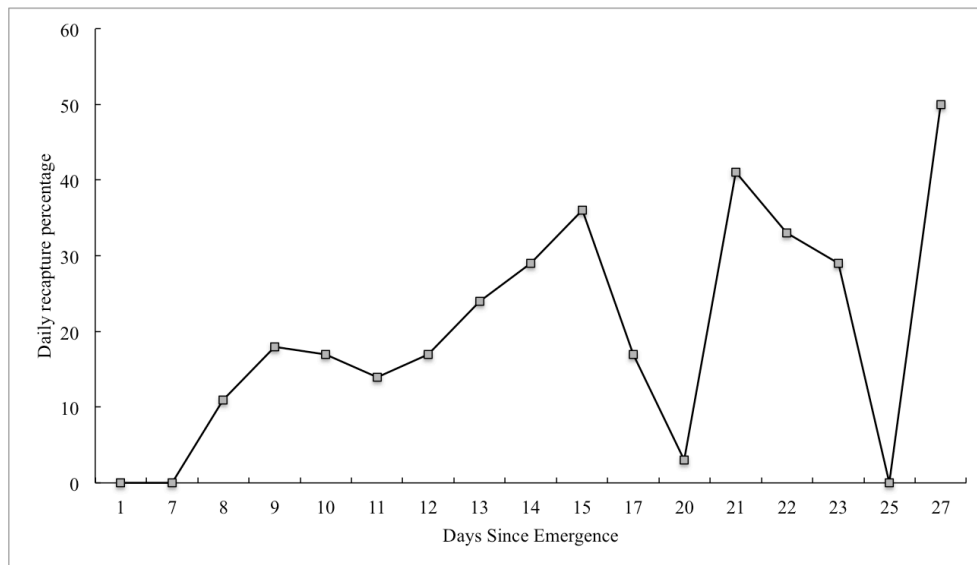


Figure 2.9. Daily percentage of captures that were recaptures of previously marked *Oeneis macounii* in the Sandilands site in 2012. Note the large drop in recaptures after day 18, when the large storm event occurred. The final day had only 2 captures, one of which was a recapture.

Population Parameters

I applied the population parameter modeling to the data from the 2012 field season in Sandilands. Due to the low capture and recapture rates of females at the Sandilands site, they were not included in the population analysis. Female butterflies are occasionally left out of population analyses due to their less conspicuous behaviour that reduces encounter rates (Ehrlich & Gilbert, 1973). The output from the models that had probability of entry (b) as a fixed parameter (\cdot), were not included because butterflies emerged over a period of several weeks, with new individuals entering the population at lower rates at the beginning and end of the flight period. This decision was reinforced when these models with b fixed also yielded extremely high AIC values, indicating that they did not fit the data as well as the time-dependent (b) models.

Figure 2.10 shows daily population estimates for each of the four models. AICc scores and total population estimates from each of the four models are shown in Table 2.2. The fully time-dependent model $\{p(t), \phi(t), b(t)\}$ had a lower AICc value than the other three models, indicating it may be the most appropriate for the data, and estimated male population size to be 2491. However, because this estimate had extremely large 95% confidence limits, the three other model outputs were examined. The second and third models, $\{p(\cdot), \phi(t), b(t)\}$ and $\{p(\cdot), \phi(\cdot), b(t)\}$, yielded very similar AICc values (Table 2.2). The population size estimates for the second and third models were 5598 (se = 1929) and 615 (se = 80), respectively. The AICc value for fourth-ranked model in Table 2.2 was substantially higher; therefore, this model was not considered further.

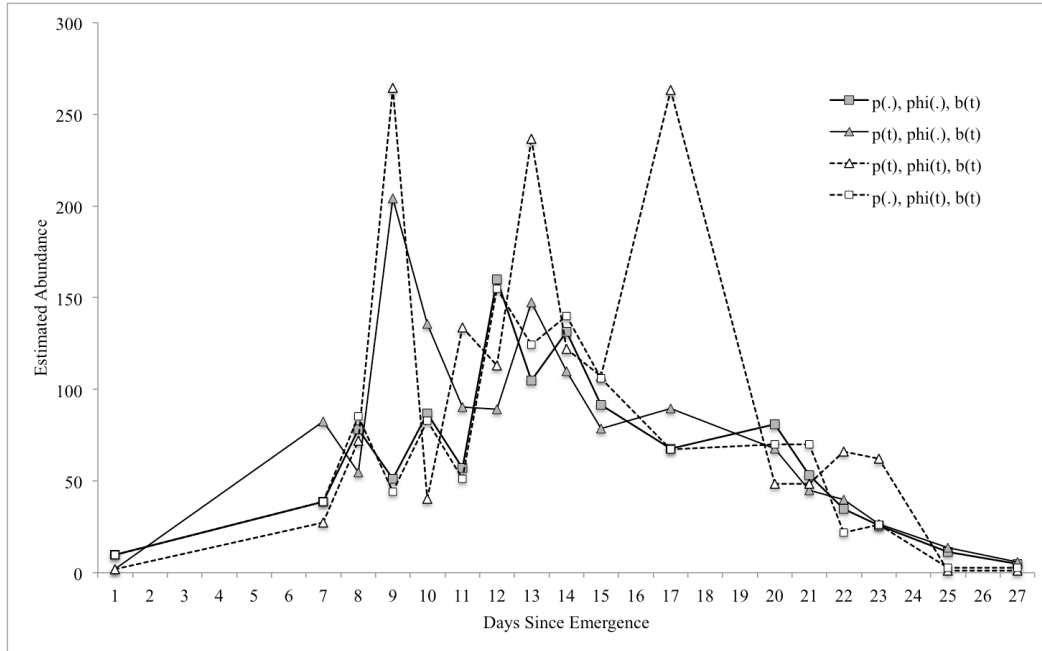


Figure 2.10. Mean estimated daily population size of *Oeneis macounii* at the Sandilands site using four different models outputted from MARK. ϕ represents the probability of survival; p the probability of recapture, and b , the probability of entry from a superpopulation. (.) Indicates constant probability over capture occasions, where (t) designates time-dependent probability.

Table 2.2. Population modeling outputs and population size estimates of male Macoun's arctic butterflies in Sandilands, Manitoba. p = recapture probability, ϕ = survival probability, b = probability of entry; subcategories (t) designate a parameter as time dependent, and (.) designate the parameter as fixed. Models are compared and ranked in ascending AICc values (Akaike's Information Criterion). $\Delta AICc$ refers to the difference of each model from the lowest AIC value.

#	Model	AICc	$\Delta AICc$	No. parameters	N_{tot}	95% confidence interval	
						Lower	Upper
1	$\{p(t), \phi(t), b(t)\}$	450.17	0.0000	28	2490	-281705	286687
2	$\{p(.), \phi(t), b(t)\}$	461.66	11.49	21	5598	1817	9379
3	$\{p(.), \phi(.), b(t)\}$	461.68	11.51	12	615	458	772
4	$\{p(t), \phi(.), b(t)\}$	476.15	25.98	28	730	400	1062

Sex Ratio

Over the course of the flight, 176 males were captured compared to only 45 females (79.6% male; Table 1). Sex ratio over the four-week flight period decreased significantly ($p < 0.01$) over the flight period, from male biased at the beginning of the flight, to slightly female biased at the end (Figure 2.11).

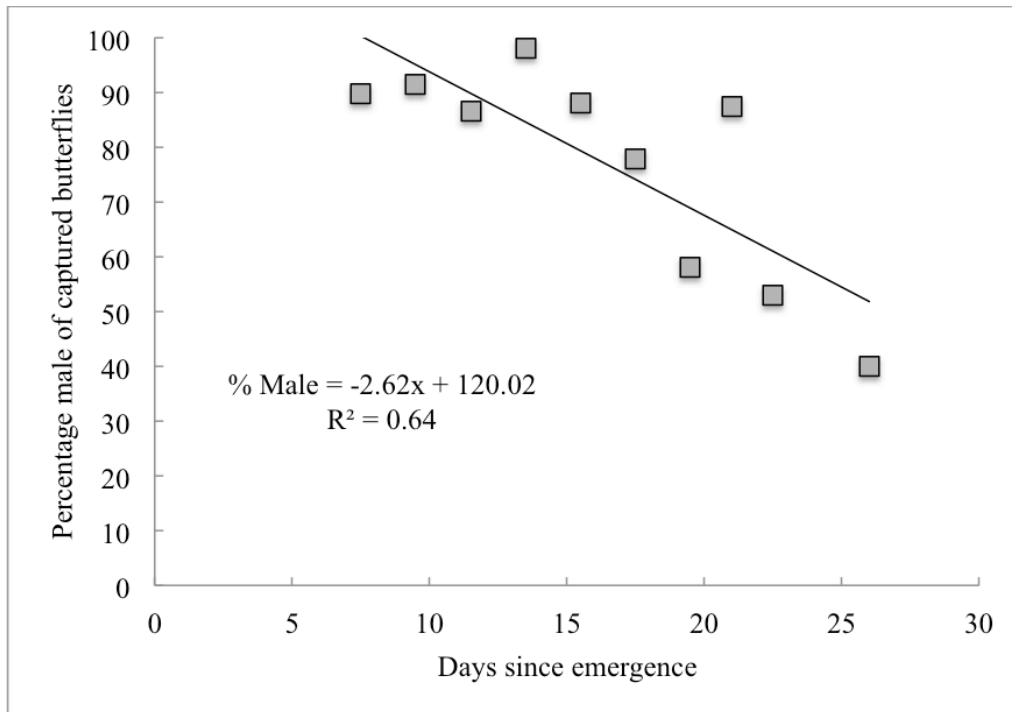


Figure 2.11. Proportion of captured butterflies that were male during the flight of *Oeneis macounii* at the Sandilands site. The slope differed significantly from zero ($p < 0.01$).

Butterfly Phenology

In total, I identified a total of 33 species of butterfly in the Sandilands site prior to and during the *O. macounii* flight period (Figure 2.12). The silvery blue (*Glaucopsyche lygdamus*) and the clouded sulphur (*Colias philodice*) first appeared one week before the first Macoun's arctic did. As was true in RMNP, white admirals (*Limentis arthemis*) emerged half way through *O. macounii*'s flight period. The little wood satyr (*Megisto cymela*) and the common wood nymph (*Cercyonis pegala*) began to appear at the end of *O. macounii*'s flight period.

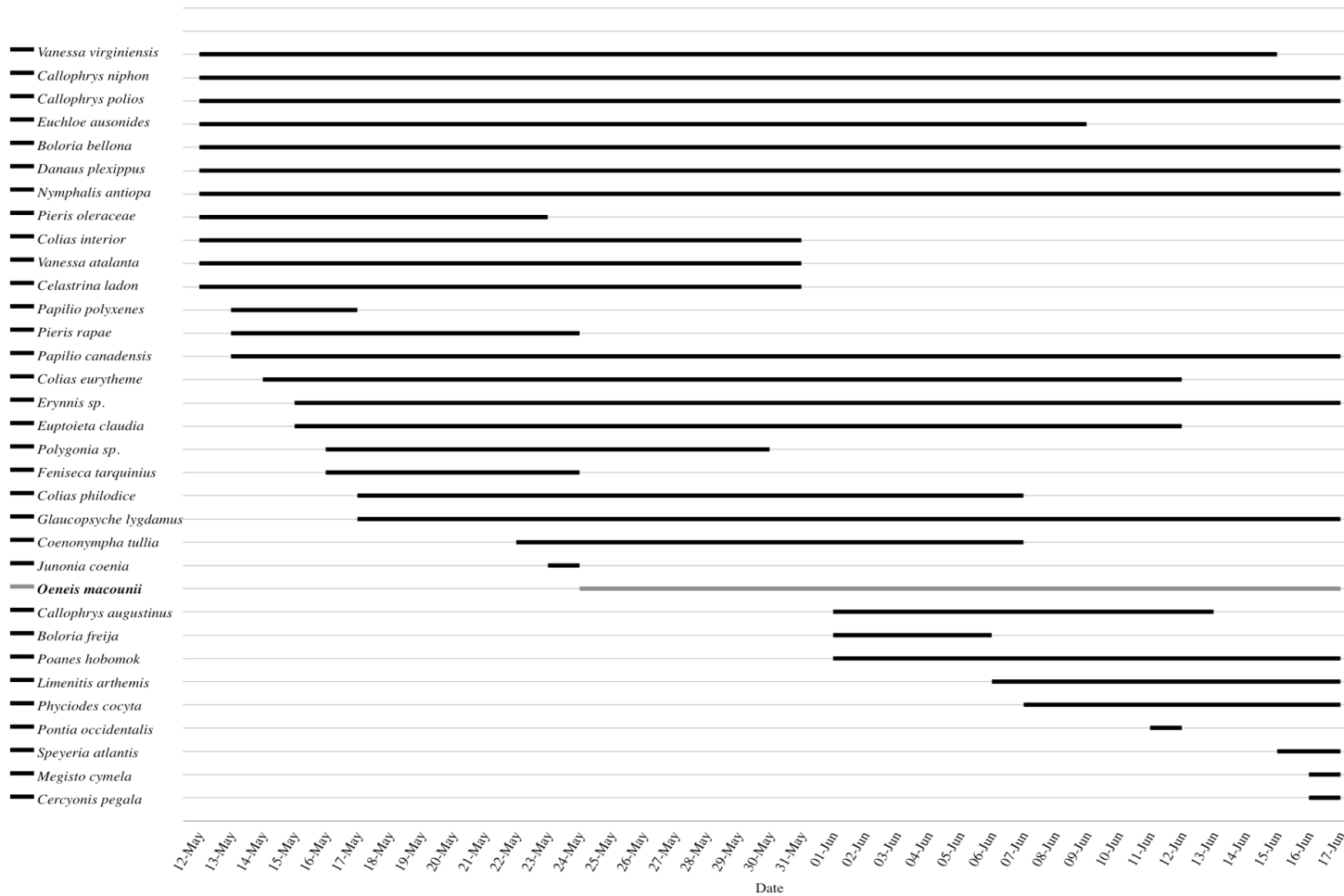


Figure 2.12. Phenology of butterfly species observed at the Sandilands site before and during *O. macounii*'s flight period.

2.5 DISCUSSION

2.5.1 General Observations and Behaviour Comparison

At the Brulé Trail site (RMNP), females were concentrated in the open, grassy meadow, whereas males were most often in areas with more tree cover in the surrounding mixed forest (Figure 3.2; see Chapter 3). Males commonly perched on branches of trees and bushes 1-3m in height in small clearings. Females were found sitting in long grass, undetectable until a researcher walked close by and startled them into flight. No oviposition was observed, despite watching females in grassy areas where *Oryzopsis angustifolia*, the suspected host plant (K.E. Johnson, pers. comm.), was present. No mating or courtship behaviour was observed.

At the Sandilands site, there were three main clusters of butterfly occurrence in the study area (Figure 2.7). The densities within these clusters were much higher than the estimated density of the entire study site. Males were usually resting on the ground. The first cluster, in the southeast corner of the site, encompassed the small hill that ran through a jack pine stand with abundant sunny openings. The second cluster north of Lonesand Road centered around a small rise on a sandy ATV track that ran through a jack pine stand. Males in this location were encountered daily sitting on the sandy track, and both males and females were caught nearby in the surrounding forest. The third cluster with the smallest number of butterfly captures was in the western portion of the site. A wide sandy trail ran through this location as well, and males were found on the sand and flying through the surrounding trees. Males in Sandilands were usually perched on the ground (such as on a trail) in sunny spots within jack

pine habitat. Females were most often caught in flight within jack pine forest, but the capture pattern largely overlapped with males (Figure 2.7). One pair of butterflies *in copula* was caught at rest on a shrub 1m from the ground in a sunny clearing of the forest. Upon capture, the pair quickly detached from each other. No oviposition was observed, despite searching sites with suspected host plants.

2.5.2 Recapture Rates and Population Sizes

The percentages of males recaptured at the two sites, with sampling effort approximately the same, were similar (23.3% vs. 22.2%), suggesting that the encounter rates of males and their behaviour were similar. In contrast, female recaptures rates were lower than those of males and differed greatly between the two sites (no statistical test, 14.3% vs. 4.4%; Table 2.1). The low recapture rate of females in the Sandilands site was unexpected. It may be partially an artifact of the habitats at each site. In RMNP, females were naturally constrained by dense surrounding forest to the large grassy prairie patch in the center of the site. There were few sites with similar habitat nearby to disperse to (Chapter 3). The habitat at Sandilands was not surrounded by dense forest and females had fewer constraints to emigration. Female capture rate may also have been lower due to behavioural differences from males. If females are more sedentary than males, encounter rates and consequently capture rates would be correspondingly lower (e.g. Shuey & Szymanski, 2012).

The population estimates of males at the Sandilands site from modeling in MARK indicate that there is a robust population of *O. macounii* at the Lonesand site in Sandilands of

uncertain magnitude. Estimates of mean total population size of males (N_{tot}) ranged from 615 (Model 3), to 2490 (Model 1) to 1820 (Model 2; Table 2.2); if the sex ratio is 1:1, then the populations including females would be about two times greater. Based upon the AICc values, the model that best fit the data had all variables as time dependent (Model 1). Model 1 the only model that reflects the effects of the large storm event that occurred mid-way through the field season and was associated with a reduced number of butterfly recaptures (Figure 2.9). However, the large confidence interval associated with this estimate makes acceptance of Model 1 questionable (Table 2.2), and with no test for significance of AICc values, it is possible that Model 1 is not the best fit. Model 2 and Model 3 have almost identical AICc values ($\Delta \text{AICc} < 7$), very similar daily population estimates (Figure 2.9), but widely differing estimates of the total population size (Table 2.2). The only difference between these two models is the parameter of survival from one sampling occasion to the next as being time-dependent or fixed. Within the limitations of the data (e.g., modest numbers of individuals captured and low recapture rate), it may be impossible to determine the ‘best’ model, thereby making the choice of model fairly subjective.

Captures of male *O. macounii* was about six times less in Riding Mountain National Park than in Sandilands (Table 2.1), despite nearly equal sampling effort. The small population size and correspondingly low recapture rate at RMNP prevented me from performing a mark-recapture analysis. However, the pattern of captures (Figure 2.6), the duration of the flight period (Figure 2.6), and the maximum daily recorded longevity (Table 2.1) were similar at the two sites. Other than population size, the demography of males seems to have been similar in the two field sites.

2.5.3 Movements

Recapture distances showed only moderate attenuation over increasing distances for both sexes at both sites (Figure 2.3), with mean movements of ~100 m (Table 2.1). The patterns of dispersal were similar for males and females at the two sites (Figure 2.3). For a species in which males are reported to defend territories that serve as mating sites (Masters *et al.*, 1967) and that rarely need to move to food resources (see Chapter 3), the extent of movements is greater than expected and probably reflects their low affinity to particular locations. *O. macounii* males may be using both “perching” and “patrolling” mating strategies (Wickman, 2009). Males were usually perched on the ground, bushes or dead trees, especially at RMNP. However, some males were observed patrolling through the habitat, especially at Sandilands. The relative frequency of these two mate-locating strategies is presumably affected by both the habitat and the population density (Kemp, 2011). Males at Sandilands infrequently crossed the Lonesand Road (12.8% of 39 recapture instances), suggesting that the unpaved road may have been a barrier that restricted movement. Roads have been shown to not impede the movements of species with open populations that travel long distance, but in species that are seemingly ‘closed’, roads can potentially restrict movement (Munguira & Thomas, 1992).

2.5.4 Sex Ratio

The significant regression of male sex ratio over time in the RMNP population (Figure 2.4) and Sandilands (Figure 2.11) indicates that the species is modestly protandrous, with males emerging slightly earlier than females. Protandry is a strategy that allows males to be physiologically capable of mating with females when they first encounter them (Wiklund & Fagerström, 1977). Butterfly sex ratio at eclosion from pupae is almost always 1:1 (Scott, 1974), as I observed for *O. macounii* at RMNP. In contrast, males comprised 80% of the individuals captured at Sandilands. Different capture probabilities between the sexes can arise for many reasons (Stoks, 2001) and it is important to understand these sampling biases when interpreting data (Foster & Soluk, 2006).. The low proportion of females captured may reflect behaviours that reduce the probability of encountering females (c.f., Ehrlich and Gilbert, 1973) or emigration by females from the study site after mating as they searched for oviposition sites.

2.5.5 Phenology

Butterfly phenologies at the two sites showed broad similarities, although there were substantial differences in the species present. The only dissimilarity in flight period was in *Boloria freija*, emerging just prior to *O. macounii* in RMNP and mid-way through the *O. macounii* flight in Sandilands. There were overall more species present at the Sandilands site, which may have been due to the abundance of wild flowers along the road, which were commonly used as a nectar resource for many species. Good predictors for the start of the

flight of Macoun's arctic were the end of the annual flight of *Erebia discoidalis* and the appearance of *Euchloe ausonides* at RMNP and fairly large numbers of *Glaucopsyche lygdamus* and *Colias philodice* at Sandilands. Highly conspicuous white admirals (*Limenitis arthemis*) first appeared near the mid-point of *O. macounii*'s flight at both sites. The end of the *O. macounii* flight was associated with the first appearance of *Enodia anthedon* and *Colias interior* at RMNP and *Megisto cymela* and *Cercyonis pegala* at Sandilands. My phenology for Sandilands corresponds closely with the research of Brent Elliott (1997) in Sandilands provincial forest in 1994, with the exception of *C. philodice* which Elliott had very few records of which appeared later (August-September) in the year than what I recorded. Notably, there was a difference of 18 days between the date of first Macoun's arctic capture in 2011 and the exceptionally early first date of capture in the unusually warm 2012 season. Butterflies are known to be good indicators of acute climactic differences (Parmesan, 2003). Despite the large difference in emergence date, the length of flight was similar.

2.5.6 Conclusions

The two populations of *O. macounii* that I studied show many similarities in life history despite differences in their habitat and different emergence dates. The RMNP population was small in comparison to the Sandilands population, potentially due to lack of suitable habitat (Chapter 3). Maximum residency, protandry, the pattern of recapture distances, and recapture rates of males were all similar in both populations, while capture and recapture rates of females were much lower at Sandilands than at RMNP. Additional study of *O. macounii* will

help to uncover undescribed aspects of its life-history, most notably the larval host plant species and courtship behaviour.

This is the first recapture study of this species, and the first qualitative data for population size, encounter rates and movements reported for this genus. This study will enable others to generate more in depth questions and make predictions about this species that were impossible to do prior to this information being made available.

CHAPTER 3

SEXUAL SEGREGATION OF THE MACOUN'S ARCTIC BUTTERFLY IN RIDING MOUNTAIN NATIONAL PARK, CANADA

3. 1 ABSTRACT

Sex-related behavioural traits, such as sexual segregation, aid in defining the life history and mating strategy of a species by describing habitat preference differences between the sexes. Understanding such traits may also help to inform conservation initiatives for endangered populations. The Macoun's arctic butterfly, *Oeneis macounii*, is a biennial satyrine butterfly that typically inhabits mature jack pine forests. However, at Riding Mountain National Park (RMNP), the species is highly localized, being found almost exclusively in and near several small prairie patches surrounded by mixed jack pine, aspen and spruce forest. Individuals of *O. macounii* were captured, marked and released in RMNP. To establish environmental characteristics associated with each butterfly occurrence, I measured habitat variables in a 5-meter radius around each butterfly capture site. A discriminant analysis correctly predicted 34 of the 41 female butterfly occurrences, and 32 of the 41 male butterfly occurrences using measured habitat variables. Males were more often in forested sites with sunny clearings, while female encounter sites were dominated by grasses and herbaceous plants. The need for two habitat types is discussed in relation to conservation strategies for the declining population of *O. macounii* at RMNP.

3.2 INTRODUCTION

Sexual segregation occurs when males and females of a species occupy different habitats. Sexual segregation most often occurs in ruminants (Kie & Bowyer, 1999), but also occurs in a wide variety of vertebrate taxa, including catsharks (Sims *et al.*, 2001), kestrels (Stinson *et al.*, 1981; Mills, 1976), turtles (Lindeman, 2003), frogs (Lamb, 1984), and elephants (Stokke & Du Toit, 2002). Sexual segregation has been infrequently reported in insects. Females of the Hine's emerald dragonfly (*Somatochlora hineana*) occupy dry meadows, and only visit the wetland habitat that males primarily inhabit to mate and to oviposit (Foster & Soluk, 2006). Among butterflies, purple-edged coppers (*Lycaena hippothoe*) segregate sexually based upon the resource defense mating strategy of males: males defend sites with high nectar source density, and females visit these areas to mate. After mating, females move to areas with larval host plants and a lower density of nectar sources (Turlure & Van Dyck, 2009). Males of the chryxus arctic butterfly (*Oeneis chryxus*) in submontane habitats spend most time on rocky ridge tops, whereas females are usually found in grassy clearings (Clayton & Petr, 1992). Several hypotheses have been proposed to explain the occurrence of habitat segregation between the sexes (Ruckstuhl, 1997; Ruckstuhl & Neuhaus, 2002). The most common explanations relate to different dietary preferences of males and females (Lindeman, 2003) and sexual behaviour practices such as lekking, where males aggregate and defend sites that females visit only to mate (Clayton & Petr, 1992; Singh *et al.*, 2010).

Oeneis macounii (Nymphalidae: Satyrinae) is found almost exclusively in Canada, ranging from British Columbia to western Quebec in sites with suitable habitat (Layberry *et al.*, 2001; Howe, 1975). As in most arctic butterflies, *O. macounii* exhibits biennialism, with cohorts

taking two years to mature fully and emerging as adults every other year (Masters, 1974). Unlike the many arctic and alpine *Oeneis* species in North America, *O. macounii* is a boreal forest inhabitant (Masters & Sorensen, 1969; Masters, 1974), usually found in association with stands of mature jack pines (*Pinus banksiana*) (Masters, 1972; Opler & Krizek, 1984). Males frequently perch in sunny spots on the ground or on bushes or trees and chase intruders, but little has been written about female behaviours. In parts of their range, male *O. macounii* form leks, a subset of perching behavior in which males aggregate and defend sites that lack food resources and oviposition sites. Females have the potential to choose their mate from among the several in the lek. Lekking is common in the foothills of the Rocky Mountains (Bird *et al.*, 1995; Guppy and Shepard, 2001), where the only reliable way to encounter Macoun's arctic is to search hilltops or open woodlands near the tops of hills ("hilltopping" behaviour *sensu* Shields, 1967; see also review by Alcock, 1987).

Historical records of *O. macounii* collected from RMNP and the surrounding region (Otis, 2010). The odd-year population in RMNP has been reported regularly since the 1930s. Interestingly, there was formerly an even-year population at RMNP as well, but no specimens have been recorded for nearly half a century (See Chapter 1; Figure 1.5), suggesting that this species may be in decline there. Though it is difficult to use museum records to extrapolate patterns of abundance due to differences in sampling effort among collectors, the disappearance of the even-year population at RMNP suggests that the odd-year population also may have declined over time and could also be at risk.

Gard W. Otis has recently searched extensively for *O. macounii* in Riding Mountain National Park (RMNP), the southeastern edge of the range of the odd-year population of central Canada. In both 2005 and 2009, he encountered a small population inhabiting grassy prairie enclaves surrounded by boreal aspen and spruce forest in one small region of the park. This habitat is unusual compared to habitats where he and others typically encounter the species (Masters, 1972; G.W. Otis, pers. comm.). During my field studies in the summer of 2011, I confirmed the occurrence of this species in the grassy prairie. However, captured individuals were predominantly female while I usually encountered males in surrounding forested areas more typical of the species. This separation of males and females within the habitat is suggestive of sexual habitat segregation.

Sexual segregation is an important factor to consider when planning conservation strategies for threatened animal species (Caro, 1999; Buchholz, 2007). Understanding habitat needs of both sexes during all life stages is a critical aspect of conservation. Some species have such dramatic habitat differences between the sexes that researchers go as far as suggesting that the sexes should be considered equivalent to two different species when planning conservation initiatives (Kie & Bowyer, 1999). Without understanding the differential utilization of the landscape by males and females, the requirements of a species are likely to be miscalculated (Foster & Soluk, 2006).

The concern that this species has shown decline in recent years at RMNP, coupled with the observations that its habitat is atypical to what normally has been reported, led to the research question of whether *O. macounii* exhibits sexual segregation in this location and how that

may relate to conservation in terms of habitat requirements of the species within Riding Mountain National Park. I performed a cluster analysis and discriminant analysis of habitat variables where males and females were captured to determine if segregation was, in fact, occurring.

3.3 METHODS

3.3.1 Study Area

The study was conducted in RMNP, in western Manitoba, Canada, at the extreme southeastern limit of the range of the odd-year population Macoun's arctic (Layberry *et al.*, 2001). The park is considered a 'wilderness island', as it is a large block of boreal forests and prairies higher in elevation than the surrounding agricultural lands (Bronson & Noble, 2006). The area of focus was within the eastern portion of the park where boreal forests predominate. This area of the park was subjected to many fires in the early 1900s (Caners & Kenkel, 2003) and a controlled burn along the Brulé Trail in 1971. Another major fire that burned 21,000 ha of land in 1980 (Caners & Kenkel, 2003) resulted in dense stands of immature jack pines (*Pinus banksiana*) through most of the area. In some pockets that were untouched by recent fires, mature *P. banksiana* can still be found, along with open prairie enclaves that are slowly changing due to succession and lack of fire since the 1970's. It is in one of these prairie patches surrounded by remnant stands of the old jack pine forest that the main field site was located. Along the Brulé Trail, close to the parking lot, there is a mosaic of mature jack pines, aspen stands and open prairie patches (UTM: 433303, 5636816). In

previous visits to the park by Gard W. Otis (pers. comm.), this was the only site in which Macoun's arctic was encountered despite extensive searching in other locations.

3.3.2 Sampling Design

Initially, one field assistant and I set up 25x25m plots (n=50) in a grid covering the Brulé meadow and extending into the surrounding forest. We measured habitat variables (tree cover, shrub cover, grass cover, wildflower species and their abundance, and elevation) in each plot prior to the emergence of adult *O. macounii*. When weather was suitable (i.e., > 18°C and partly to completely sunny) we walked through the plots 3-5 times per day along regular routes designed to provide complete coverage of the Brulé prairie and adjacent forest. Surveys were conducted between the hours of 10:00-17:00 h from 1 June to 16 July 2011. The first Macoun's arctic was seen on 18 June, and the last on 12 July, with 16 days of suitable weather for observation in the field site during the flight. As the season progressed, we noticed *O. macounii* outside the original grid and extended the region we surveyed. When a Macoun's arctic was observed, we captured it with a net, and recorded the UTM coordinates with a Garmin Oregon 450t handheld GPS unit (accuracy <10m). We marked each butterfly with a unique number written with a black Sigma Micron 0.25mm pen on a dot of white Golden Fluids acrylic paint. The behaviour each butterfly exhibited at the time of capture was recorded. Every individual was sexed by observing abdomen size and presence/absence of male genitalic claspers. Butterflies were always released where they had been captured, by gently removing them from the net and placing them on vegetation, in order to minimize the influence of capture and

handling on their behaviour. If an individual was recaptured, UTM coordinates were again recorded unless it had been captured earlier that same day. Occasionally during the flight period, we searched other sites with suitable habitat in the eastern portion of the park for Macoun's arctic.

When I began to analyze the data collected in 2012, it appeared that males and females had been caught in different habitats. Additionally, because I had extended the area I surveyed, many butterfly capture sites fell outside of the initial grid area. Finally, the 25-meter scale of the plots was too coarse to allow me to accurately associate each butterfly with the microhabitat at the site of capture. For these reasons, I returned to RMNP in 2012 to better quantify the microhabitat at each of the 2011 butterfly capture sites. On 5 June 2012, I relocated each capture site using the GPS data coupled with my memory of visual landmarks. Habitat variables were assessed within 5 m of each capture site by visually estimating the type of cover to the nearest 5%. Habitat characteristics have been assessed in circles around capture points of kestrels (Stinson *et.al.*, 1981) and elk (Mao *et al.*, 2005). I chose a radius of 5m to reflect the scale at which butterflies probably assess their immediate surroundings. The variables chosen seemed to define important aspects of the site and included percentage tree cover, percentage shrub cover, percentage grass and other herbaceous vegetation (those three variables summed to 100%), elevation (from the GPS unit), and number of *P. banksiana* trees present.

3.3.3 Analyses of Habitat Data

Butterfly capture locations were mapped using the ESRI software ArcGIS. For the base map, I used an orthorectified image from 2007 air photos provided by Riding Mountain National Park and the Manitoba Land Initiative. Habitat variables were tested against one another for normality and equality of variances using a Shapiro-Wilk test for normality, and were found to be not normally distributed and to not have equal variances. A Cochran's t-test was run to compare male and female means of each habitat measure.

An exploratory cluster analysis (Ward's method) was conducted to assign individual butterflies into groups based on the habitat characteristics that had been quantified in the field. In order to test if habitat measures could separate males and females into separate clusters, the first and second canonical coefficients generated in this analysis were plotted against each other. If habitat measures can predict sex of butterfly occurrence, then males and females are expected to be found in different clusters. Alternatively, if habitat variables do not differ between the sexes, then I expect males and females to be equally represented in the clusters.

In order to test if habitat measures could predict the sex of a butterfly, I completed a discriminant function analysis. This test was used to assess how well the model could correctly predict sex of a butterfly captured based upon the habitat variables that were measured. All five variables were used in the model: tree cover, shrub cover, grass/herbaceous cover, number of mature *P. banksiana* trees, and elevation. Shapiro-Wilk tests showed that the data were not

normally distributed, and therefore I used a discriminant analysis with unequal variance. . Analyses were all completed using SAS 9.2, Cary, NC.

3.4 RESULTS

Oeneis macounii was moderately common at the Brulé Trail prairie and surrounding forest, but they were largely absent from other sites in the park. In total, I captured 30 males a total of 39 times, and 34 females a total of 40 times at the study site. The numbers of males and females captured was approximately equal ($\chi^2 = 0.25$, d.f. = 1, $p = 0.62$).

3.4.1 *Habitat Analysis*

I ran Cochran's t-tests assuming unequal variance to compare measured male and female habitat variables. Microhabitats of males and females differed significantly from each other. Male capture locations were associated with greater percentage of tree cover ($t = -7.89$, d.f. = 40, $p < .001$), lower percentage of grass ($t = 7.51$, d.f. = 40, $p < .001$), and higher number of jack pines present ($t = -5.36$, d.f. = 40, $p < .001$) than females. Shrub cover did not differ between the sexes ($t = 0.14$, d.f. = 40, $p = 0.89$) (Table 3.1). A map (Figure 3.1) depicts the differences in male and female encounter locations.

Table 3.1. Means of three habitat measures in a 5-meter diameter circle surrounding the capture points. Males and females showed different preference in habitat measures.

		Mean measures of habitat variables			
	<i>n</i>	Tree Cover*	Grass Cover*	Shrub Cover	Number of <i>P. banksiana</i> *
Male	30	58.5%	13.5%	28.0%	3.7
Female	34	16.2%	55.1%	28.7%	0.7

* Indicates significance of $< .001$ between sexes of that variable

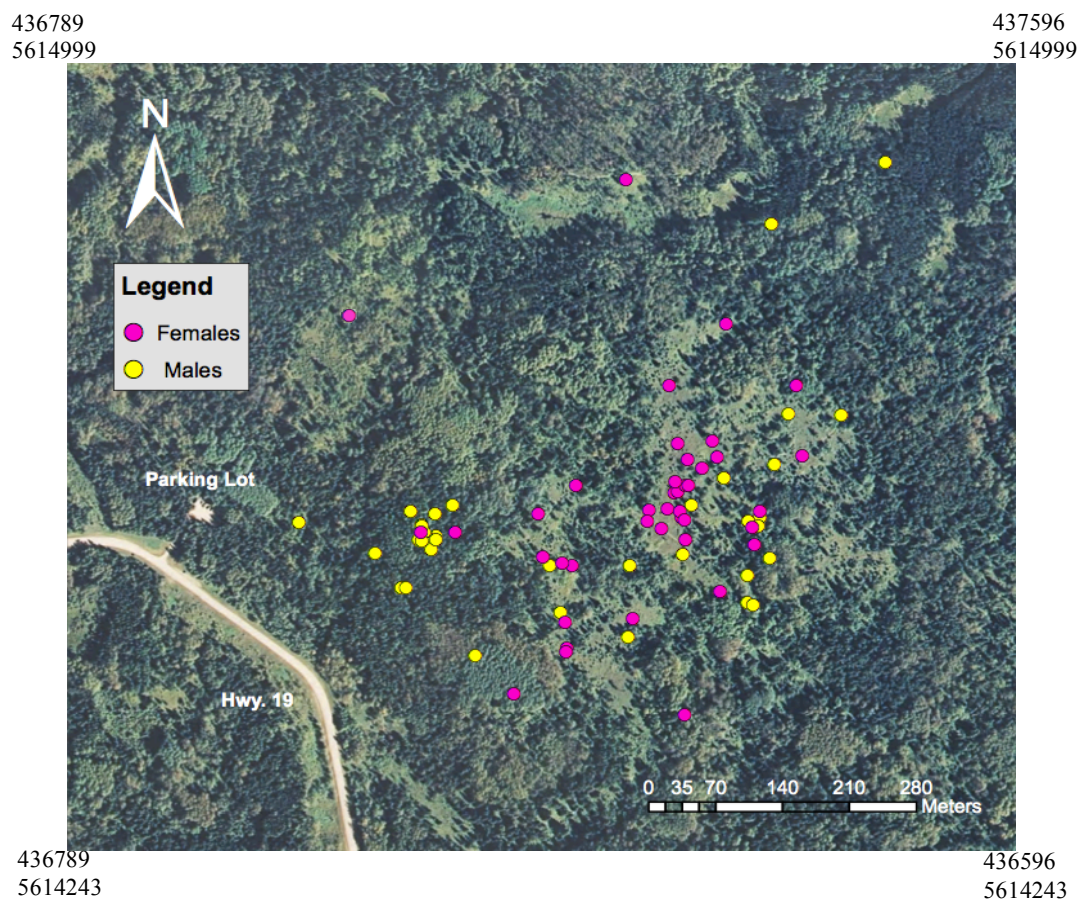


Figure 3.1 The study site along the Brulé Trail, RMNP. Butterfly capture points are indicated by coloured symbols: males yellow, females pink. Lighter green areas are predominated by grass and herbaceous cover, and darker green are heavily wooded areas. Males and females were found in different areas, suggesting sexual segregation. Base photos courtesy of Riding Mountain National Park, Parks Canada.

The discriminant analysis of habitat variables from *O. macounii* capture locations confirmed this sexual habitat segregation. Female occurrence was correctly predicted 34 of 41 times (83%), and male occurrence was correctly predicted 32 of 41 times (78%). The overall error rate of the model is 19%.

The canonical coefficients derived from the cluster analysis of the habitat variables yielded three clusters (Figure 3.2). The first canonical variable was the basis of the primary separation of the three clusters. Cluster 1, which was formed based on higher levels of tree cover, contains predominantly males (24 male; 3 female), whereas Cluster 3, associated with high levels of grass and herb cover, contains almost entirely females (2 male; 17 female). Cluster 2 had an even distribution of male and female butterflies. The canonical coefficient analysis defined the individuals in Cluster 2 with a higher level of shrub cover. Figure 3.3 shows the three clusters plotted onto the site map.

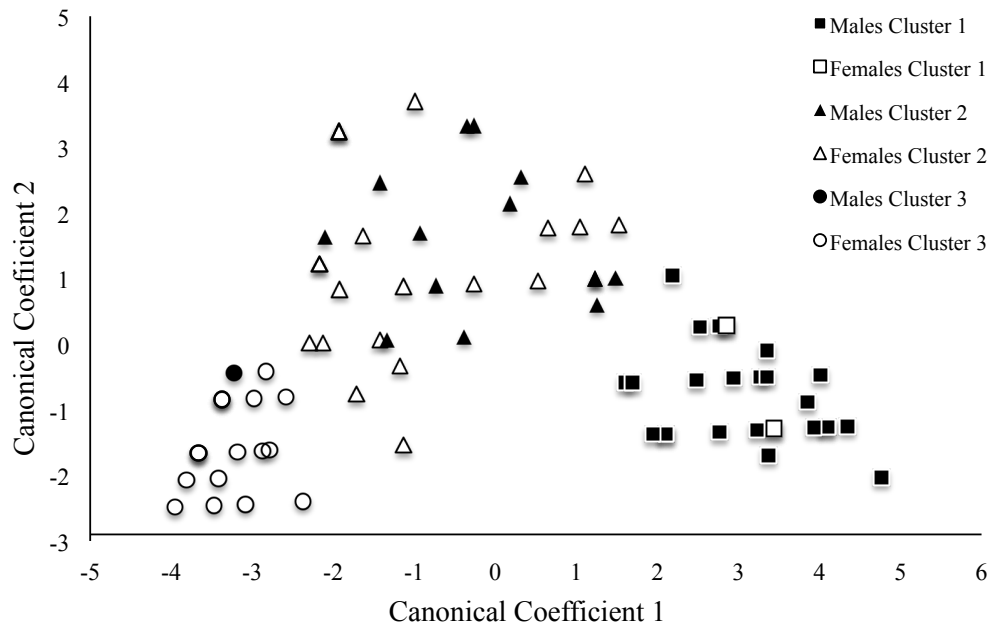


Figure 3.2 Biplot of the first and second canonical correlation coefficients that form the three clusters for *O. macounii* based upon environmental variables measured for each butterfly. Symbols indicate each cluster group, with open symbols representing females and closed symbols representing males. Males predominate in cluster 1, whereas females in cluster 3. Cluster 2 has both sexes. Figure 3.3 projects these clusters onto the base map of the site.

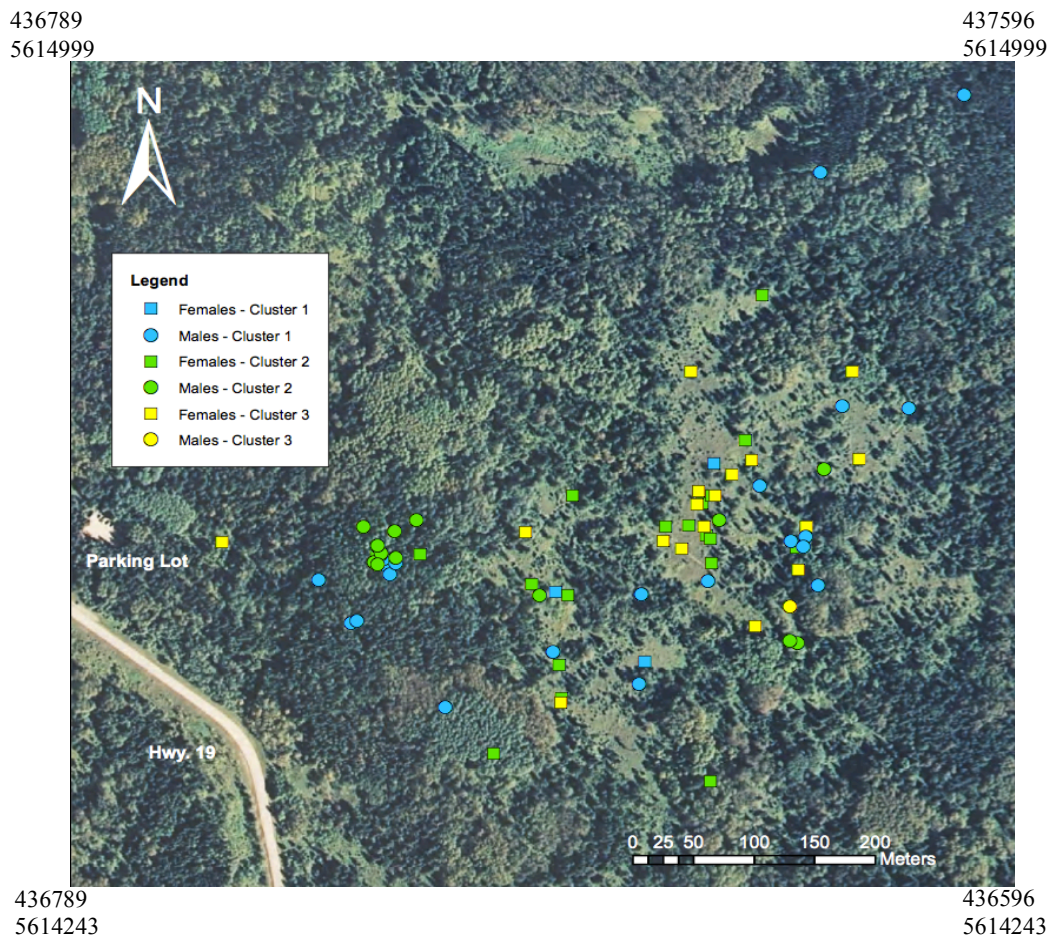


Figure 3.3 Clusters plotted onto the map of the study site. Males predominate cluster 1, females predominate cluster 3, while cluster 2 has a mixture of both sexes.

3.5 DISCUSSION

3.5.1 Sexual Segregation

Male and female Macoun's arctic butterflies at RMNP were found to inhabit different locations within the landscape. Males were most commonly observed perched in small sunny clearings in wooded areas as well as in sunny sites along small paths within the forest. In contrast, females were most frequently observed in sites with little to no tree cover in the grass- and shrub-dominated meadow surrounded by boreal forest. The differences in microhabitats where I observed male and female *O. macounii* were unanticipated based on descriptions of the habitat of the species as being dry, sandy clearings in jack pine forests, with no mention of differences between the sexes (Masters, 1972; Layberry *et al.*, 2001). The ability to predict butterfly sex based upon measured habitat variables, as shown by my discriminant analysis, confirms that sexual segregation occurs in this species at this site. This study is the first to identify these contrasting habitat needs of male and female Macoun's arctic butterflies. Furthermore, I came across very few published examples of sexual habitat segregation in the class Insecta (Turlure & Van Dyck, 2009; Clayton & Petr, 1992; Foster & Soluk, 2006).

Lindemann (2003) pointed out that dietary differences between males and females often underlie sexual habitat segregation. This is not the case for Macoun's arctic. Despite being at the study site on every day with suitable weather for butterfly activity, I never saw adult *O. macounii* feed at any potential food source (i.e., flowers, sap, scat, fungal exudates). Bird *et*

al. (1995) also reported that adult *O. macounii* rarely feed. Being largely freed from the requirement to feed is conducive to the evolution of sexual habitat segregation, as males and females have no need to visit common areas containing food sources. With daily time budgets freed from the constraint of finding food, most time can be spent on reproductive behaviours (i.e., male territoriality and mate searching, female oviposition).

I believe that habitat segregation occurs more frequently in insects than is reported in the literature because lekking has not been defined as habitat segregation. For example, in lekking chryxus arctics (*Oeneis chryxus*), males aggregate on rocky ridge tops while females are found in grassy meadows (Clayton & Petr, 1992; Daily *et al.*, 1991). This description matches the definition of habitat segregation, but the authors did not refer to it as such. Numerous other insect species are reported to be hilltoppers, but are not referred to as being sexually segregated (Shields, 1967; Alcock, 1983; 1985).

Mating strategy of male *Oeneis macounii* varied depending on the habitat of the site they occupied. In the more open sites males perched extensively and only infrequently interacted with other males; when two males did interact, their spiral flights were short and they sometimes perched again within a few meters of each other. However, at perches defended in the scarce clearings within extensive forested areas, individual males had more frequent territorial contests with intruding males. In that setting, on two occasions males successfully defended a sunspot for several days in succession. The variation in male mating behavior seems to reflect both differential density of males at perch sites and the scarcity of suitable clearings in the forest that presumably served as mate-rendezvous sites. Similar variation in

behavior has been reported for the well-studied speckled wood butterfly (*Pararge aegeria*). In open oak forest in England with abundant sun-spots, male-male contests are short-lived and largely symbolic (e.g., the resident individual always reclaims the sun-spot following a short interaction with an intruding male; Davies, 1978). However, in coniferous forests of Sweden where sun-spots are scarce, contests are intense and of long duration (Wickman and Wiklund, 1983).

The mating system of *O. macounii* is variable over its range. In sites in central and eastern Canada with little topographic relief, males wander extensively and show little or no fidelity to specific sites (see data for southeastern Manitoba, Chapter 2). However, when there are hills, even if they rise only a few meters above the surrounding landscape, males show hilltopping tendencies (i.e., they have a tendency to be aggregated on high points in the landscape; G.W. Otis, pers. comm.). At the other extreme, males in population of *O. macounii* in the foothills of the Rocky Mountains show strong lekking tendencies; they are regularly encountered in clearings in forests on the tops of hills and small mountains (Bird *et al.*, 1995). At these locations one encounters predominantly males, and contests between males over sites are frequent (G.W. Otis, pers. comm.); these traits match the definition of lekking. Among other North American species in the genus *Oeneis*, hilltopping and consequently some spatial separation of males and females is reported for *O. alberta*, *O. melissa*, and some populations of *O. nevadensis* (Bird *et al.*, 1995; Warren, 2005). The variable male-mating strategies exhibited within and between different *Oeneis* species offer opportunities for further study of factors such as population density, habitat type, and topographic relief that may affect mating strategies and sexual segregation.

In contrast to males, females were usually encountered in open, grassy sites. Males were never observed interacting with conspecifics. They were not noticed until they were disturbed, when they would fly away in evasive, fast flights usually at a height of <1 m. The flights ended when the female was either captured or lost in a shrubby area.

3.5.2 Conservation

I observed relatively few *O. macounii* at Riding Mountain National Park: only 64 individuals were caught and marked, and those marked butterflies were recaptured only 11 times. Low capture and recapture rates prevented a meaningful mark-recapture analysis (i.e., calculated confidence intervals of population size would be very large). A number of factors may have contributed to the low recapture rate. Apart from a few conspicuous males in forest clearings, there was little evidence of residency at particular sites. In a companion study in southeastern Manitoba, distances between recaptures also showed little attenuation at increasing distances (see Chapter 2, Table 2.1), suggesting high vagility of this species. Males may have dispersed from the study site but no observations enable me to verify that. The low recapture rate undoubtedly reflects the relatively short adult life span: the maximum recorded longevity was only 10 days at RMNP and 9 days in a companion study (see Chapter 2). Additionally, Macoun's arctics are usually sedentary and very cryptic unless disturbed. When at rest with their wings closed, they are nearly impossible to detect, thereby contributing to low encounter rates (see Shuey & Szymanski, 2012). These factors affected the number of butterflies I observed. However, when compared to the 2012 field season at Sandilands

Provincial Forest, during which the sampling effort was approximately the same, the Riding Mountain population was much smaller (Chapter 2, table 2.1). Despite the constraints that are inherent with sampling this species, the small number of *O. macounii* I observed in the Brulé site, their localization to that site (see below), and their apparent decline historically (see figure 3.4) suggest that this population is small and potentially at risk of extirpation.

An effective conservation strategy requires accurate knowledge of habitat usage of both sexes and all life stages of a species. In the case of *Oeneis macounii* at RMNP, males and females occupy different habitats, both of which occur in and around the prairie clearings along the Brulé Trail. Absence of sites suitable for male territories (small openings in forests) could reduce effective population size (Anthony & Blumstein, 2000). Females occupy more open grassy sites, presumably in close proximity to oviposition sites (Brues, 1920; Ehrlich & Raven, 1964). The grass species required for oviposition and larval development of Macoun's arctic are unconfirmed. However, *Oryzopsis angustifolia*, the likely host plant species (K.E Johnson, pers. comm.) and several other grass species are common in the Brulé prairie but largely absent from densely forested areas. Absence or low abundance of host plants can also result in reduced populations of butterflies (Schultz & Dlogosch, 1999).

Despite searching in many boreal regions of the park to the east of Clear Lake, only four individuals were observed outside of the Brulé site: one female near the eastern park boundary on the trail to Bald Hill and three males 5-7 km east of the Brulé site. This low number of encounters outside the Brulé site suggests that at least one of the habitats required by the two sexes is absent over most of the boreal forest region of the park. At the Brulé trail

site, the proximity of grasses required for oviposition in the open prairie to sunny clearings within forested areas required by males may be critical for the maintenance of this population within Riding Mountain National Park.

As Insecta is a very beneficial group for ecosystem services (Losey & Vaughan, 2006), and as behaviour is important to consider in conservation planning (Caro, 1999), sexual segregation should be considered when making conservation decisions about insect species. Conservation planning for Macoun's arctic at Riding Mountain National Park should consider and reflect the requirement for male and female microhabitats to be in close proximity to each other. Further research into female *O. macounii* (especially their host plants and oviposition behaviour) is needed to create a more complete conservation plan for the species within the park. Based on my research, preservation of prairie meadows (through small controlled burns or mowing) near the Brulé Trail coupled with the creation of forest clearings within the forest near those prairie patches are recommended to maintain populations of Macoun's arctic until extensive young jack pine forests have matured into more typical habitat.

CHAPTER 4

GENERAL DISCUSSION AND CONCLUSIONS

4.1 Summary of Results

The most important discovery I made with this project was made when I assessed habitat associations of male and female *Oeneis macounii* in Riding Mountain National Park, and found clear evidence of sexual segregation occurring among adults. Males were more frequently found in areas of high tree cover, and females in open, grassy areas. I hypothesize that males are using forested sites as mate attraction locations, and females were found in grassy areas that serve as oviposition sites. Sexual segregation is not reported often in insects, even though quite a few insect species use lekking as a mating strategy that effectively results in males and females occupying different parts of their habitat. It seems that researchers have not framed lekking in terms of sexual segregation. It is possible that *O. macounii*'s lack of feeding as adults is conducive to the sexual segregation I observed. With individuals freed from the need to find food resources, a majority of adult life can be focused on reproductive behaviours such as mate finding and oviposition.

While the evidence for sexual segregation was clear at RMNP, I did not record habitat variables around capture points at Sandilands to quantify segregation at this location. Casual observation did not suggest that segregation was occurring. This may reflect the greater openness of the jack pine forests there, which led to congruence between the microhabitats sought by the two sexes. Extremely low recapture rates of females in Sandilands (4.4%,

compared to 14.3% at RMNP) suggest that females were either leaving the study area, or were behaving in ways that reduced encounter rate with them. Unfortunately, I obtained no direct evidence in support of either explanation.

Capture and recapture rates at both sites were lower than I anticipated at the onset of this project. Originally, I had intended to evaluate various factors that may contribute to male mating success, but over the course of two field seasons I only encountered one mating pair and briefly witnessed one attempted courtship. Because of the sedentary nature of adults, their cryptic colouration, and their lack of nectaring and mudpuddling behaviours, I now realize that *Oeneis macounii*, and possibly all members of the genus, are likely poor candidates for investigations of factors affecting mate choice.

Estimates of population size were calculated for the Sandilands population, with two of the three main parameters, p (recapture probability) and ϕ (survival probability) being either fixed or time-dependent. I had concerns about the outputs of all four models. The “best” model, based on AICc values, had an unacceptably large confidence interval. The second and third best models yielded similar daily population estimates, but differed by a factor of 4 for total population size. Compared to other studies of insects using similar models (e.g., Tikkamaki, 2011), my estimated population sizes were much more variable. It is possible that none of the models derived from the mark-recapture data accurately portrayed the actual population size. The problems with modeling may have been influenced by the short lifespan of these insects (maximum observed longevity of 9-10 days). Another event that undoubtedly affected population estimates was the storm with heavy rains (62.8 mm), low temperatures

(5.9°C) and high winds that occurred midway through the season. This storm notably reduced recapture rates: 17% of butterfly captures the day before the storm consisted of recaptures, whereas only 3% of captures were recapture events the day after the storm. This unexpected high loss of individuals from the population most likely affected the analyses performed in MARK. The small number of captures at RMNP (30 males, 34 females) coupled with modest recapture rates (23.3% for males, 14.3% for females) did not allow for meaningful abundance estimates to be made.

I compared many aspects of life history and population parameters in the two study sites. These are summarized in table 4.1. Moderate protandry in this species was statistically significant at both sites; protandry is common in insects. Butterfly phenology data allowed me to identify indicator species that frame the beginning and end of the annual flight of *O. macounii* in each site. However, because of different butterfly faunas in the two study sites, the species differed. The one similarity in phenology at the two sites was the white admiral butterfly (*L. arthemitis*) that was a good indicator of the mid-point of *O. macounii*'s flight. Phenological data from Sandilands largely corresponds with previous phenological data reported by Brent Elliott (1997), including similar calendar dates of emergence.

My results from RMNP, coupled with historical collection records of *O. macounii* (Otis, 2010), indicate that the population at RMNP is small and highly localized. I believe that the Macoun's arctic should become a species of special concern in the park. With almost identical sampling efforts at the two study sites, the large difference in numbers of butterflies

Table 4.1 Summary of sexual segregation, population size, longevity, recapture rates and phenological indicators of Macoun's arctic butterfly from the two field sites.

	RMNP (2011)	Sandilands (2012)
Evidence for sexual segregation?	Yes	No
Marked males	30	176
Marked females	34	45
Recapture rate (%)		
<i>Males</i>	23.3	22.2
<i>Females</i>	14.3	4.4
Population size estimate	Not calculated	1230 - 4980
Maximum observed longevity	10 days	9 days
Phenology indicators		
<i>Beginning</i>	<i>Erebia discoidalis</i> <i>Euchloe ausonides</i>	<i>Glaucopsyche lygdamus</i> <i>Colias philodice</i>
<i>Middle</i>	<i>Limenitis arthemis</i>	<i>Limenitis arthemis</i>
<i>End</i>	<i>Enodia anthedon</i> <i>Colias interior</i>	<i>Megisto cymela</i> <i>Cercyonis pegala</i>
Protandry	Significant	Significant

captured at RMNP population and Sandilands (i.e., 176 vs. 30 males) indicates that the RMNP population is very small. This locale is situated on the southeastern edge of the distribution of the odd-year population of central Canada, and is geographically and genetically isolated from other the nearest known population in the Duck Mountains, 80 km to the northwest. Immigration therefore must be extremely rare, which further disadvantages the small population within the park (no gene flow or supplementation of the population from migrants).

4.2 Future Research Directions

The Macoun's arctic, like most species of *Oeneis*, is understudied. Prior to my research, the last publication on this species was by Masters in 1974. Despite contact with this species on 34 days over two field seasons, my research failed to uncover some unanswered ecological questions about this butterfly. For example, the host plant that females lay eggs on remains unknown. The suggestion by Kyle Johnson (pers. comm.) that the primary host plant may be *Oryzopsis angustifolia* may help future researchers to observe oviposition in the field. Also, I only observed one courtship, thereby making it impossible to study this behaviour or characteristics that males that successfully mate possess. Further study of mating behaviour would be valuable, especially to the at-risk RMNP population, as behavioural data is essential to successful conservation planning (Caro, 1999).

Continued monitoring of Macoun's arctic at the Brulé Trail site at RMNP and searching at other localities within the park are warranted given the currently limited identified distribution in the park and the small numbers of adults observed during the entire flight period in 2011. My fieldwork and observations of others indicate that *O. macounii* rarely feeds as an adult. Could this potentially be of benefit in terms of conservation planning? Often conservationists suggest that enhancing nectar sources is the best and easiest way to ensure the continued survival of a butterfly species. With limited necessity to feed as adults, other strategies for the conservation of this species in small isolated populations can be focused upon as opposed to actively planting adult nectar sources as has been done for some rare nectar-feeding species (Blake *et al.*, 2011; Schultz & Dlugosch, 1999).

My thesis has focused on two populations of this widespread butterfly. Indications of lekking in populations in the foothills of the Rocky Mountains could be examined to see how greater topographic variation affects mating strategy. Additional comparative studies of even- and odd-year populations may reveal differences in ecology or behaviour that have never been observed for a biennial species. However, it seems likely that such studies would encounter the same issues I faced while studying this butterfly, such as low encounter rates and difficulty observing behaviour. The Macoun's arctic, in the context of this project, has proven to be a poor study organism for behavioural research, largely due to its cryptic nature and short life span. However, the interesting result of sexual segregation at RMNP indicates that further study of *Oeneis* populations may result in unexpected discoveries.

LITERATURE CITED

- Alcock, J. 1983. Territoriality of hilltopping males of the great purple hairstreak, *Atildes halesus* (Lepidoptera, Lycaenidae): convergent evolution with pompilid wasp. *Behavioural Ecology and Sociobiology* 13: 57-62.
- Alcock, J. 1985. Hilltopping in the nymphalid butterfly *Chlosyne californica* (Lepidoptera). *American Midland Naturalist* 113: 69-75.
- Alcock, J. 1987. Leks and hilltopping in insects. *Journal of Natural History* 21: 319-328.
- Anthony, L.L. & Blumstein, D.T. 2000. Integrating behaviour into wildlife conservation: the multiple ways behaviour can reduce N_e . *Biological Conservation* 95: 303-315.
- Baur, B., Raboud, C. 1988. Life history of the land snail *Arianta arbustorum* along an altitudinal gradient. *Journal of Animal Ecology* 57: 71-87.
- Beccaloni, G.W. & Gatson, K.J. 1995. Predicting the species richness of neotropical forest butterflies: Ithomiinae (Lepidoptera: Nymphalidae) as indicators. *Biological Conservation* 71: 77-86.
- Bird, C.D., Hilchie, G.J., Kondla, N.G., Pike, E.M & Sperling, F.A.H. 1995. Alberta Butterflies. The Provincial Museum of Alberta, Edmonton. AB.
- Blake, R.J., Woodcock, B.A., Westbury, D.B., Sutton, P. & Potts, S.G. 2011. New tools to boost butterfly habitat quality in existing grass buffer strips. *Journal of Insect Conservation* 15: 221-232.
- Blanckenhorn, W.U. 1991. Life-history difference in adjacent water strider populations: phenotypic plasticity or heritable responses to stream temperature? *Evolution* 45: 1520-1525.
- Boggs, C.L., Watt, W.B. & Ehrlich, P.R. 2003. Butterflies: Ecology and Evolution Taking Flight. University of Chicago Press, Chicago.
- Brock, J.P. & Kaufman, K. 2003. Kaufman Field Guide to Butterflies of North America. Houghton Mifflin Harcourt, Boston.
- Bronson, J. & Noble, B. 2006. Measuring the effectiveness of Parks Canada's environmental management system: a case study of Riding Mountain National Park. *Canadian Geographer* 50: 101-113.
- Brues, C.T. 1920. The selection of food-plants by insects, with special reference to Lepidopteran larvae. *The American Naturalist* 54: 313-332.

- Buchholz, R. 2007. Behavioural biology: an effective and relevant conservation tool. *Trends in Ecology and Evolution* 22: 401-407.
- Caners, R.T. & Kenkel, N.C. 2003. Forest stand structure and dynamics at Riding Mountain National Park, Manitoba, Canada. *Community Ecology* 4: 185-204.
- Caro, T. 1999. The behaviour-conservation interface. *Trends in Ecology and Evolution*. 14: 366-369.
- Carson, H.L. & Kaneshiro, K.Y. 1976. *Drosophila* of Hawaii: systematics and ecological genetics. *Annual Review of Ecology and Systematics* 7: 311-345.
- Caswell, H. 1983. Phenotypic plasticity in life-history traits: demographic effects and evolutionary consequences. *American Zoologist* 23: 35-46.
- Clayton, D.L. & Petr, D. 1992. Sexual differences in habitat preference and behavior of *Oeneis chryxus* (Nymphalidae: Satyrinae). *Journal of the Lepidopterists' Society* 46: 110-118.
- Daily, G.C., Ehrlich, P.R., Wheye, D. 1991. Determinants of spatial distribution in a population of the subalpine butterfly *Oeneis chryxus*. *Oecologia* 88: 587-596.
- Davies, N.B. 1978. Territorial defence in speckled wood butterfly (*Pararge aegeria*): the resident always wins. *Animal Behaviour* 26: 138-147.
- Ehrlich, P.R. & Gilbert, L.E. 1973. Population structure and dynamics of the tropical butterfly *Heliconius ethilla*. *Biotropica* 5: 69-82.
- Ehrlich, P.R. & Raven, P.H. 1964. Butterflies and plants: a study in coevolution. *Evolution* 18: 586-608.
- Elliott, B.G. 1997. Implications of forest management techniques for biodiversity, species and ecosystems diversity among Lepidoptera in planted and naturally regenerated jack pine stands. M.Sc. Thesis, University of Manitoba, Fall 1997, 282pp.
- Fairbairn, D.J. 1984. Microgeographic variation in body size and development time in the waterstrider, *Limnopus notabilis*. *Oecologia* 61: 126-133.
- Field Studies Council: Background Information. <<http://www.geography-fieldwork.org/icefieldwork/glacial/stage1.htm>> Date accessed: 24 January 2013.
- Foster, S.E. & Soluk, D.A. 2006. Protecting more than wetland: The importance of biased sex ratios and habitat segregation for conservation of the Hine's emerald dragonfly, *Somatochlora hineana* Williamson. *Biological Conservation* 127: 158-166.

- Gilbert, L.E. & Singer, M.C. 1975. Butterfly ecology. *Annual Review of Ecology and Systematics* 6: 365-397.
- Groeters, F.R. & Dingle, H. 1987. Genetic and maternal influences on life history plasticity in response to photoperiod by milkweed bug. *The American Naturalist* 129: 332-346.
- Guppy, C.S. and Shepard, J.H. 2001. Butterflies of British Columbia. University of British Columbia Press, Vancouver. BC.
- Howe, W.H. 1975. The Butterflies of North America. Doubleday & Company, New York.
- Johnson, K.E. 2011. *Personal communication*. Department of Entomology, University of Wisconsin, Madison.
- Kie, J.G. & Bowyer, R.T. 1999. Sexual segregation in white-tailed deer: density-dependent changes in use of space, habitat selection, and dietary niche. *Journal of Mammalogy* 80(3): 1004-1020.
- Kemp, D.J. 2001. Investigating the consistency of mate-locating behavior in the territorial butterfly *Hypolimnys bolina* (Lepodoptera: Nymphalidae). *Journal of Insect Behavior* 14: 129-147.
- Klassen, P., Westwood, A.R., Preston, W.B., and McKillop, W.B. 1989. The Butterflies of Manitoba. Manitoba Museum of Man and Nature, Winnipeg.
- Knapton, R.W. 1985. Lek structure and territoriality in the chryxus arctic butterfly, *Oeneis chryxus* (Satyridae). *Behavioral Ecology and Sociobiology* 17: 389-395.
- Lamb, T. 1984. The influence of sex and breeding condition on the microhabitat selection and diet in the pig frog *Rana grylio*. *American Midland Naturalist* 111: 311-318.
- Layberry, R.A., Hall, P.W. & Lafontaine, J.D. 2001. The Butterflies of Canada. University of Toronto Press, Toronto.
- Lindeman, P.V. 2003. Sexual difference in habitat use of Texas map turtles (Emydidae: *Graptemys versa*) and its relationship to size and diet. *Canadian Journal of Zoology* 81: 1185-1191.
- Losey, J.E. & Vaughan, M. 2006. The economic value of ecological services provided by insects. *Bioscience* 56: 311-323.

- Manitoba Land Initiative: Government of Manitoba. <<http://mli2.gov.mb.ca/>> Date accessed: 13 July 2012.
- Manitoba Wild Lands: Manitoba Land Use Planning. <<http://manitobawildlands.org>> Date accessed: 24 Jan 2013.
- Mao, J.S., Boyce, M.S., Smith, D.W., Singer, F. J., Vales, D.J., Vore, J.M. & Merrill, E.H. 2005. Habitat Selection by elk before and after wolf reintroduction in Yellowstone National Park. *Journal of Wildlife Management* 69: 1691-1707.
- Masters, J.H. 1972. Habitat: *Oeneis macounii* Edwards. *Journal of Research on the Lepidoptera* 10: 301.
- Masters, J.H. 1974. Biennialism in *Oeneis macounii* (Satyridae). *Journal of the Lepidopterists' Society* 28: 237-242.
- Masters, J.H. & Sorensen, J.T. 1969. Field observations on forest *Oeneis* (Satyridae). *Journal of the Lepidopterists' Society* 23: 155-161.
- Masters, J.H., Sorensen, J.T. & Conway, P.J. 1967. Observations on *Oeneis macounii* (Satyridae) in Manitoba and Minnesota. *Journal of the Lepidopterists' Society* 21: 258-260.
- Mills, G.S. 1976. American Kestrel sex ratio and habitat segregation. *Auk* 93:740-748.
- Montgomery, S.L. 1983. Carnivorous caterpillars: the behavior, biogeography and conservation of *Eupithecia* (Lepidoptera: Geometridae) in the Hawaiian Islands.
- Munguira, M.L. & Thomas, J.A. 1992. Use of road verges by butterfly and butnet populations, and the effect of roads on adult dispersal and mortality. *Journal of Applied Ecology* 29: 316-329.
- New, T.R. 1997. Are Lepidoptera an effective 'umbrella group' for biodiversity conservation? *Journal of Insect Conservation* 1: 5-12.
- Opler, P.A. & Krizek, G.O. 1984. Butterflies East of the Great Plains: An Illustrated Natural History. John Hopkins University Press, Baltimore.
- Opler, P.O. & Wright, A.B. 1999. A Field Guide to Western Butterflies. Houghton Mifflin Company, New York.

- Otis, G.W. 2010. Riding Mountain National Park: Annual Report- 2010. Speciation in the Biennial Butterfly, Macoun's arctic (*Oeneis macounii*). Report to Parks Canada.
- Otis, G.W. 2011-2012. *Personal communication*. School of Environmental Science, University of Guelph.
- Parmesan, C. 1996. Climate and species' range. *Nature* 382: 765-766.
- Parmesan, C. 2003. Butterflies as bioindicators for climate change effects. *In*: Boggs, C.L., Watt, W.B., and Ehrlich, P.R. Butterflies (eds) *Ecology and evolution taking flight*. University of Chicago Press, Chicago and London.
- Rader, R.B., Belk, M.C., Shiozawa, D.K. & Crandall, K.A. 2005. Empirical tests for ecological exchangeability. *Animal Conservation* 8: 239-247.
- Roy, D.B., Rothery, P., Moss, D., Pollard, E. & Thomas, J.A. 2001. Butterfly numbers and weather: predicting historical trends in abundance and the future effects of climate change. *Journal of Animal Ecology* 70: 201-217.
- Ruckstuhl, K.E. & Newhuas, P. 2002. Sexual segregation in ungulates: a comparative test of three hypotheses. *Biological Reviews* 77: 77-96.
- Ruckstuhl, K.E. 1997. Foraging behaviour and sexual segregation in bighorn sheep. *Animal Behaviour* 56: 99-106.
- Saether, B-E., Bakke, O. 2000. Avian life history variation and contribution of demographic traits to the population growth rate. *Ecology* 81: 642-653.
- Saylor, S.G. Prehistoric human occupation and ecology in the Sandilands Forest Reserve, southeastern Manitoba. M.A. thesis, The University of Manitoba, Winter 1975, 157pp.
- Schultz, C.B. & Dlugosch, K.M. 1999. Nectar and hostplant scarcity limit populations of an endangered Oregon butterfly. *Oecologia* 119: 231-238.
- Scott, J.A. 1974. Mate-locating behavior of butterflies. *American Midland Naturalist* 91: 103-117.
- Shields, O. 1967. Hilltopping. *Journal of Research on the Lepidoptera* 6: 69-178.
- Shuey, J. & Szymanski, J. 2012. Modified pollard transects do not predict estimated daily population size for the secretive butterfly, *Neonympha mitchellii mitchellii* french. *Journal of the Lepidopterists' Society* 66: 221-224.

- Sims, H.P. 1975. The effect of prescribed burning on some physical soil properties of jack pine sites in southeastern Manitoba. *Canadian Journal of Forest Research* 6: 58-68.
- Sims, D.W., Nash, J.P. & Morritt, D. 2001. Movements and activity of male and female dogfish in a tidal sea lough: alternative behavioral strategies and apparent sexual segregation. *Marine Biology* 139:1165-1175.
- Singh, N.H., Bonenfant, C., Yoccoz, N.G. & Côté, S.D. 2010. Sexual segregation in Eurasian wild sheep. *Behavioral Ecology* 21: 410-418.
- Stearns, S.C. 1989. The evolutionary significance of phenotypic plasticity. *Bioscience* 39: 436-445.
- Stinson, C.H., Crawford, D.L. & Lauthner, J. 1981. Sex differences in winter habitat of American kestrels in Georgia. *Journal of Field Ornithology* 52: 29-35.
- Stokke, S. & Du Toit, J.T. 2002. Sexual segregation in habitat use by elephants in Chobe National Park, Botswana. *African Journal of Ecology* 40: 360-371.
- Stoks, R. 2001. Male-biased sex ratios in mature damselfly populations: real or artifact? *Ecological Entomology* 26: 181-187.
- Thomas, J.A. 2005. Monitoring change in the abundance and distribution of insects using butterflies and other indicator groups. *Philosophical Transactions of the Royal Society* 360: 339-357.
- Tikkimaki, T. & Komonen, A. 2010. Estimating population characteristics of saproxylic beetles: a mark-recapture approach. *Journal of Insect Conservation* 15: 401-408.
- Turlure, C. & Van Dyck, H. 2009. On the consequences of aggressive male mate-locating behaviour and micro-climate for female host plant use in the butterfly *Lycaena hippothoe*. *Behavioral Ecology and Sociobiology* 63: 1581-1591.
- Warren, A.D. 2005. Lepidoptera of North America 6. Butterflies of Oregon: Their Taxonomy, Biology, and Distribution. Contributions of the C. P. Gillette Museum of Arthropod Diversity, Fort Collins, CO.
- Wickman, P.-O. 2009. Mating behavior in butterflies. In Settele, J., Shreeve, T., Konvicka, M. & Van Dyck, H. (eds) *Ecology of Butterflies in Europe*. Cambridge University Press, Cambridge.
- Wickman, P.O. & Wiklund, C. 1983. Territorial defense and its seasonal decline in the speckled wood butterfly (*Pararge aegeria*) 31: 1206-1216.

- Wiklund, C. & Fagerström, T. 1977. Why do males emerge before females? A hypothesis to explain protandry in butterflies. *Oecologia* 31: 153-158.
- Zimmerman, K., Konvicka, M., Fric, Z. & Cihakova, V. 2009. Demography of a common butterfly on humid grasslands: *Argynnis aglaja* (Lepidoptera: Nymphalidae) studied by mark-recapture. *Polish Journal of Ecology* 57: 715-727.

APPENDIX 1 – Phenology of flower species on the Brulé trail before, during and after *O. macounii*'s flight.

Flower Species	Before	During	After
Dandelion (<i>Taraxacum officinale</i>)			
Early Blue Violet (<i>Viola adunca</i>)			
Starwort (<i>Stellaria longipes</i>)			
Bearberry (<i>Arctostaphylos uva-ursi</i>)			
Strawberry (<i>Fragaria virginiana</i>)			
Hoary Puccoon (<i>Lithospermum canescens</i>)			
Pussy Toe (<i>Antennaria neglecta</i>)			
Golden Corydalis (<i>Corydalis aurea</i>)			
Three Toothed Cinquefoil (<i>Sibbaldiopsis tridentata</i>)			
Nanny Berry (<i>Viburnum lentago</i>)			
Licorice Root (<i>Glycyrrhiza glabra</i>)			
Prairie Rose (<i>Rosa woodsii</i>)			
Pale Vetchling (<i>Lathyrus ochroleucus</i>)			
Blue Eyed Grass (<i>Sisyrinchium bellum</i>)			
Star Flowered Solomon's Seal (<i>Maianthemum stellatum</i>)			
Seneca Snakeroot (<i>Polygala senega</i>)			
Hairy Honeysuckle (<i>Lonicera hirsuta</i>)			
Striped Coralroot (<i>Corallorhiza striata</i>)			
Tall Lungwort (<i>Mertensia paniculata</i>)			
Canada Anemone (<i>Anemone canadensis</i>)			
Bunchberry (<i>Cornus canadensis</i>)			
Yellow Avens (<i>Geum aleppicum</i>)			
Leafy Spurge (<i>Euphorbia esula</i>)			
Shrubby Cinquefoil (<i>Dasiphora fruticosa</i>)			
Early Meadow-Rue (<i>Thalictrum dioicum</i>)			
Three Flowered Avens (<i>Geum triflorum</i>)			
Common Yarrow (<i>Achillea millefolium</i>)			
Tall Cinquefoil (<i>Potentilla arguta</i>)			
Wood Lily (<i>Lilium philadelphicum</i>)			
Wild Lily-of-the-valley (<i>Maianthemum canadense</i>)			
White Clover (<i>Trifolium repens</i>)			
Ox-Eye Daisy (<i>Leucanthemum vulgare</i>)			
Harebell (<i>Campanula rotundifolia</i>)			
Blue Giant Hyssop (<i>Agastache foeniculum</i>)			
Black Eyed Susan (<i>Rudbeckia hirta</i>)			
Northern Bedstraw (<i>Galium boreale</i>)			
Tufted Vetch (<i>Vicia cracca</i>)			
Indian Paintbrush (<i>Castilleja minata</i>)			
Common Burdock (<i>Arctium minus</i>)			
Golden Alexander (<i>Zizia aurea</i>)			